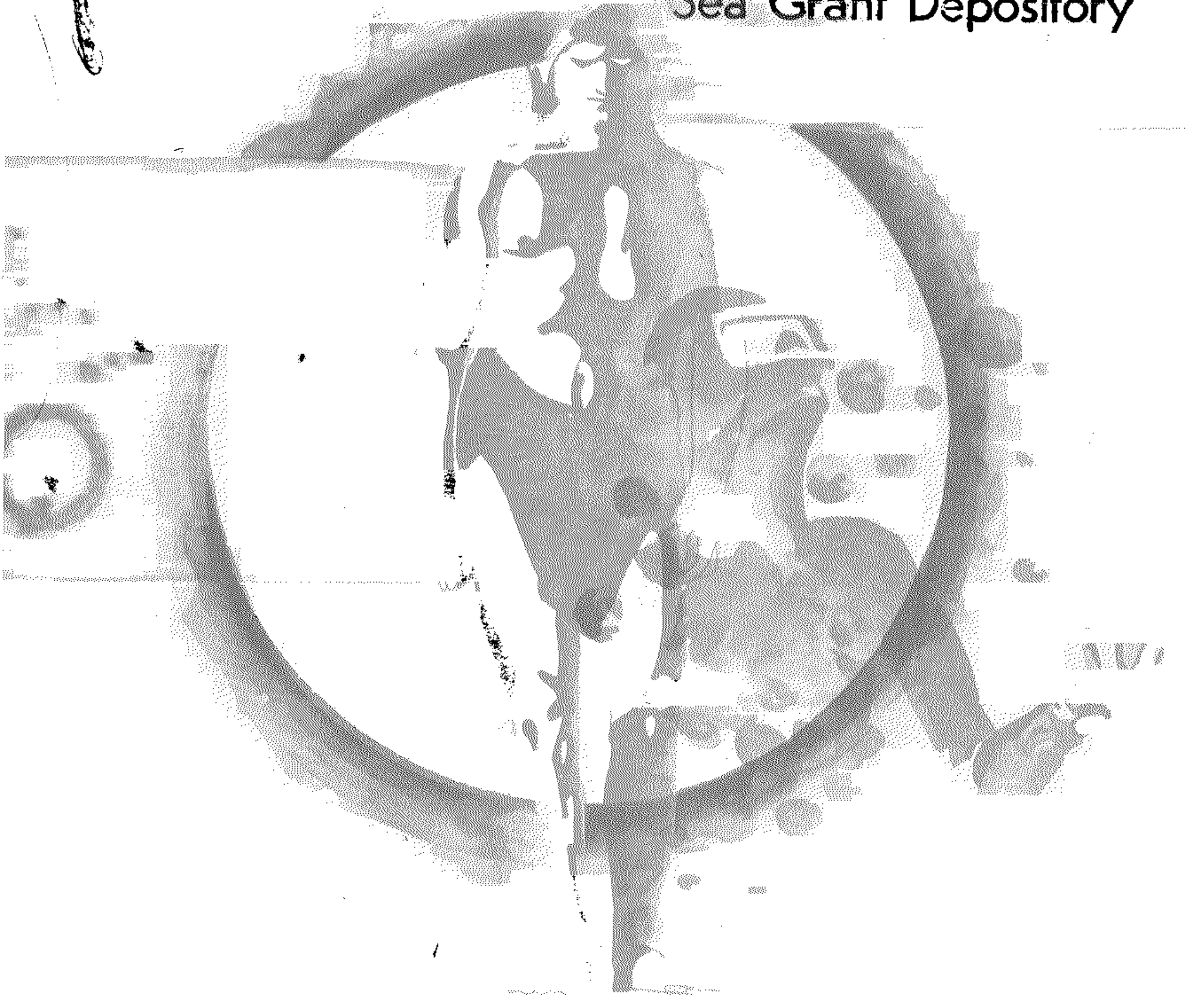
STUDIES ON HUMAN PERFORMANCE IN THE SEA

VOLUME



UNIVERSITY OF HAWAII · SEA GRANT COLLEGE PROGRAM · JULY 1975

The late Wallace Fenn once said of applied research that it should be conducted with "basic research thinking." To this end, we began at the beginning--simple immersion and breath-holding to gain basic data on the most primitive of man's methods of entering the sea.

STUDIES ON HUMAN PERFORMANCE IN THE SEA

VOLUME I

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PREFACE

This compilation of papers represents the efforts of many people; scientific, technical and clerical -- too many to be acknowledged individually. We hope our gratitude to the team is implicit and explicit in our summary of many hours of hard work.

The University of Hawaii is a fledgling member of Sea Grant Institutions, but then, the Sea Grant Program is also a relatively recent endeavor.

We present this series as a contribution from only one of many Sea Grant projects which are conducted at the University of Hawaii. Our theme has been "Human Performance in the Sea", with the implication that future efforts to utilize resources from this environment will ultimately involve the human element -- despite the degree of technological sophistication available.

Our approach has been to examine physiologic responses to hyperbarism which range from basic to highly applied research -- never losing sight of the fundamental question -- what are the limits, and why? We believe we have arrived at some of the answers, and have asked some new questions.

While the major support of the diving-related program is under the auspices of the Sea Grant Office, other agencies have contributed significantly to our research effort and should be acknowledged.

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Many of the authors and co-authors represented here were from other institutions or members of the Department who now pursue research activities at another institution. Some, to whom we are grateful, are colleagues from other departments at this university, whose collaboration brought better perspective to our joint investigations.

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chapter 1

PHYSIOLOGY of WATER IMMERSION and BREATH-HOLDING

Chapter 1 deals mainly with a fundamental mammalian reflex initiated by breath-holding per se, which evolved as an oxygen-conserving mechanism when terrestrial mammals made excursions into the sea. In marine mammals, the reflex is highly developed; in the land-bound mammals, less so. However, vestiges of the reflex, including bradycardia, peripheral vasoconstriction, increased central blood volume, etc., remain even in man and other non-aquatic mammals. They demonstrate limited ability to become "heart-lung-brain" machines during as simple a maneuver as apnea.

We were intrigued by two facets of this reflex, (i) what factors other than breath-holding had a modifying or modulating effect on the basic response, and (ii) is the reflex more highly developed in terrestrial mammals who "return often to the sea," i.e., divers? Can an evolutionary defense mechanism, rendered less necessary by non-aquatic habitation, be re-acquired?

Although, in the applied sense, we were most interested in the response of man in the sea, certain techniques were better pursued in experiments with other animals; therefore, the chapter includes papers dealing with diving responses in the rat and dog--non-aquatic mammals--and the sea lion, a superbly adapted marine mammal.

STUDIES ON MAN

Reprinted from *Journal of Applied Physiology* 30(4):540-547, 1971

Alveolar gas exchanges and cardiovascular functions during breath holding with air

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HONG, S. K., Y. C. LIN, D. A. LALLY, B. J. B. YIM, N. KOMINAMI, P W HONG, AND T. O. MOORE. *Alveolar gas exchanges and cardiovascular functions during breath holding with air.* J. Appl. Physiol. 30(4): 540–547. 1971.—During 4 min breath holding (BH) beginning with TLC or 160 sec BH with FRC $P_{A_{O_2}}$ decreased continuously, reaching approximately 30 mm Hg at the end, while $P_{A_{CO_2}}$ increased during first 30 sec after which it leveled off at approximately 50 mm Hg. During 4 min BH the lung supplied 700 ml of O_2 into the blood while it gained only 160 ml of CO_2 from the blood. indicating a significant retention of CO_2 in the blood and tissues. Both Pa_{O_2} and Pa_{CO_2} changed during BH in a manner similar to those of alveolar gas. (A-a)O_2 gradient was approximately 10 mm Hg throughout the entire BH period, while the usual (a-A)CO_2 gradient was reversed during BH. Mixed venous O_2 pressure and content approached arterial blood values toward the end of BH, whereas mixed venous CO_2 pressure and content approached arterial values after 30–60 sec of BH. The systolic pressure increased only slightly during 1–2 min of BH, after which it increased rather sharply. The pulse pressure tended to increase toward the end of BH. The right atrial and intraesophageal pressures increased immediately to and were maintained at 10–20 mm Hg during BH. Cardiac outputs measured during 1.5–2 min BH by dye-dilution method were not significantly different from those measured before BH.

blood gas; blood pressure; cardiac output; intraesophageal pressure

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IN 1963 LANPHIER AND RAHN (19) described a technique for obtaining sequential alveolar gas samples during breath holding, and reported that the course of events in breath holding with air is quite different from that reported in breath holding with oxygen. For instance, the alveolar P_{CO_2} tends to reach a plateau while oxygen uptake from the lungs diminishes markedly during 1 min breath holding with air (8, 19). Since then, similar results have been reported for breath holding longer than 1 min by Tibes and Stegemann (33) and Hong et al. (16). However, the latter authors collected gas samples only at the end of breath holding and hence the time course of alveolar gas exchange during a prolonged breath hold could not be assessed quantitatively.

In understanding the course of events in breath holding, it is equally important to know changes in the blood gas composition. On the basis of theoretical analysis, Lanphier and Rahn (19) concluded that the arterial P_{CO_2} rapidly becomes equal to the mixed venous P_{CO_2} and thereafter exceeds it by an increasing margin. Actual verification of this conclusion requires measurements of the arterial blood gas and the cardiac output, in addition to the amount of alveolar gas exchange. Although Paulev (25) actually determined both the arterial blood gas and the cardiac output during breath holding, he did not determine the alveolar gas exchange.

Hence, this investigation was undertaken to study, during a prolonged breath hold with air, *1)* changes in the alveolar gas pressures and the time course of alveolar O_2 and CO_2 exchange, *2)* changes in the cardiac output, *3)* changes in arterial blood gas pressures and contents, and *4)* changes in the arterial, right atrial, and esophageal pressures.

METHODS

Nine male subjects (5 divers and 4 nondivers; average age of 30 years) selected for an earlier breath-holding study (16) were employed in the present work. These subjects are able to breath hold for 2–4 min following a full inspiration of air without preliminary hyperventilation. The residual volume of each subject was measured by the nitrogen-dilution method of Rahn et al. (27) and the resting O_2 consumption by using a 13-liter Collins recording spirometer. Breath holding was conducted in a sitting position without previous hyperventilation, but each experiment began, unless stated otherwise, with maximal expiration followed by a single, maximal inspiration (TLC group). In this case, the volume inspired was recorded on a spirogram. In one series breath holding began at the end of normal expiration (FRC group). The FRC was determined separately before the breath-holding experiment, using a 13-liter Collins recording spirometer. Relevant lung volumes for each group are summarized in Table 1 along with the values of resting $\dot{V}_{O_2}$.

A) Alveolar gas sampling. Immediately after the initial inspiration, the subject expired into a sequential sampling device developed by Lanphier and Rahn (19). As soon as a sample had been obtained, he reinspired this gas from the sampler bag (500-ml capacity) and held his breath in a normal manner. The sampling process was repeated every 30 sec in case of breath holding with TLC and every 20 sec in case of breath holding with FRC throughout the breath hold. The subject was encouraged to breath hold until the breaking point. Gas samples were analyzed for O_2 and CO_2 in a Scholander micro-gas analyzer (30).

TABLE 1. *Lung volumes and oxygen consumption of subjects at rest*

Measurements	TLC Group (N = 9)	FRC Group (N = 5)
RV, ml BTPS	1,470 ± 130	1,490 ± 133
ERV, ml BTPS		1,500 ± 116
FRC, ml BTPS		2,990 ± 235
VC,* ml BTPS	3,980 ± 440	
TLC, ml BTPS	5,450 ± 305	
$\dot{V}_{O_2}$, ml/min STPD	247 ± 11	240 ± 23

Values are means ± SE. *Volume inspired after a maximal expiration immediately preceding breath hold.

B) Arterial blood sampling. After local anesthesia with lidocaine, an arterial catheter-needle (Longdwel) was inserted into the brachial artery. At the same time, a long cardiac catheter was also inserted into the basilic vein and guided into the right atrium for the measurement of pressure and cardiac output (vide infra). The position of the cardiac catheter was ascertained by fluoroscopy. Arterial blood samples were collected anaerobically into heparinized syringes before, during, and 20 sec after breath holding. With the exception of the last blood sampling after a breath hold, the time of blood sampling was always synchronized with the alveolar gas sampling. Immediately after each sampling, a 1-ml aliquot of blood was transferred into a test tube and deproteinized with tungstic acid for the measurement of lactic acid concentration by a method using *p*-hydroxydiphenyl (1). The rest of the blood samples were stored in a packed-ice box and were analyzed for P_{O_2}, P_{CO_2}, and pH within 60 min by using a microelectrode unit (Radiometer, Copenhagen). The CO_2 electrode was calibrated with 3 and 6% CO_2 in O_2 gas mixture (humidified) and the O_2 electrode with saline equilibrated with air. The pH electrode was calibrated with standard solutions provided by the manufacturer. Blood O_2 and CO_2 capacities were assumed to be normal, and the values of blood P_{O_2} and P_{CO_2} were employed to determine blood O_2 and CO_2 content using the nomogram of Dill et al. (10).

C) Measurements of blood pressures and cardiac output. Both the arterial and the right atrial pressures were recorded before and during breath holding on a Picker MC-IV photographic recorder, using Statham P23AC and P23BC transducers, respectively. The cardiac output was also determined before and during breath holding by a Cardio-Green-dilution method. Cardio-Green (5 mg; Hynson, Westcott and Dunning, Inc.) was injected rapidly into the right atrium through a cardiac catheter (vide supra) after which the brachial arterial blood was withdrawn at a constant rate (20 ml/min) into a Gilford densitometer and the dye-dilution curve was obtained on a strip-chart recorder (Texas Instruments). The entire procedure required approximately 30 sec. The area under the dye-dilution curve was subsequently determined by planimetry. The heart rate was determined from the ECG (standard lead I).

D) Measurement of intraesophageal pressure. An esophageal balloon made of latex with dimensions of 15 x 1 cm (Anode Rubber Plating Company) was used for the measurement of intraesophageal pressure according to the procedure described by Knowles et al. (18). The pressure was recorded using a Statham P23BC transducer.

RESULTS AND DISCUSSION

A) Alveolar gas exchange. Average percent concentrations of O_2 and CO_2 in alveolar gas samples obtained during breath holding are summarized in Table 2, while the changes in the corresponding gas pressures are presented in Fig. 1. Although the duration of breath holding varied with subjects, the alveolar gas composition at a given time during breath holding was quite similar in all experiments and hence the data obtained at each sampling time have been pooled. In general, O_2 values fell continuously but at a diminishing rate, whereas CO_2 values increased rapidly during the first 20–30 sec after which they increased very slightly. Although the initial levels of O_2 and CO_2 values were dependent upon the initial lung volume, the time course of changes in these gas values was parallel. Moreover, at the end of maximal breath holding (i.e., 240 sec for breath holding with TLC and 160 sec with FRC), there was no significant difference between TLC and FRC groups in the O_2 and CO_2 values. The terminal alveolar P_{O_2} and P_{CO_2} values were approximately 30 (range of 24–36) and 56 (range of 51–66) mm Hg, respectively.

The observed changes in alveolar O_2 and CO_2 levels as a

TABLE 2. *Percent O_2 and CO_2 in alveolar gas samples during breath holding (BH) with air*

BH with TLC*				BH with FRC†			
Time, sec	No. of exp	O_2	CO_2	Time, sec	No. of exp	O_2	CO_2
0	15	18.9 ± 0.3	3.4 ± 0.1	0	11	15.0 ± 0.4	4.9 ± 0.2
30	15	15.4 ± 0.4	5.6 ± 0.1	20	11	12.4 ± 0.5	6.2 ± 0.1
60	15	12.6 ± 0.5	6.3 ± 0.1	40	11	9.9 ± 0.5	6.6 ± 0.1
90	15	10.0 ± 0.4	6.7 ± 0.1	60	11	8.0 ± 0.5	6.9 ± 0.1
120	15	8.1 ± 0.3	7.0 ± 0.1	80	10	6.9 ± 0.5	7.3 ± 0.2
150	12	6.7 ± 0.3	7.3 ± 0.2	100	8	5.6 ± 0.5	7.5 ± 0.1
180	8	5.5 ± 0.5	7.4 ± 0.2	120	7	4.9 ± 0.3	7.7 ± 0.1
210	5	5.0 ± 0.4	7.6 ± 0.3	140	5	4.6 ± 0.2	7.9 ± 0.2
240	5	4.3 ± 0.3	7.8 ± 0.4	160	3	3.9 ± 0.4	8.0 ± 0.4

Values are means ± SE. *A total of 15 experiments on 9 subjects were performed. † A total of 11 experiments on 5 subjects were performed.

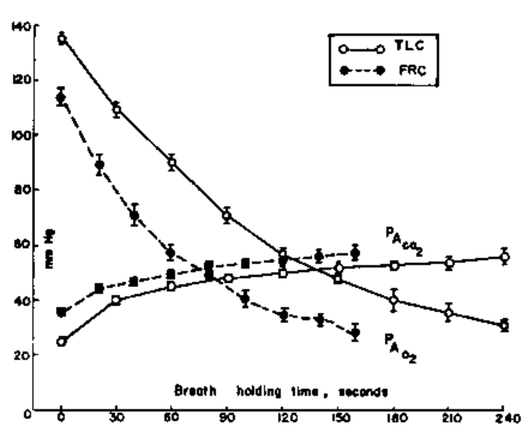

FIG. 1. Alveolar P_{O_2} and P_{CO_2} during breath holding with total lung capacity (TLC) or with functional residual capacity (FRC). See Table 1 for the number of measurements for each point. Vertical bars indicate one standard error of the mean.

TABLE 3. *Rates of removal of O_2($\Delta V_{L_{O_2}}$) from and of addition of CO_2 ($\Delta V_{L_{CO_2}}$) into the lung and alveolar gas exchange ratio (R) during breath holding (BH) with air*

BH with TLC				BH with FRC			
Time, sec	$\Delta V_{L_{O_2}}$	$\Delta V_{L_{CO_2}}$	R	Time, sec	$\Delta V_{L_{O_2}}$	$\Delta V_{L_{CO_2}}$	R
	ml/30 sec; STPD				*ml/20 sec; STPD*		
0–30	180	93	0.52	0–20	97	28	0.29
30–60	141	23	0.16	20–40	67	5	0.07
60–90	125	15	0.12	40–60	48	5	0.10
90–120	90	3	0.03	60–80	25	8	0.32
120–150	63	9	0.14	80–100	33	0	0
150–180	49	3	0.06	100–120	18	5	0.28
180–210	23	6	0.26	120–140	7	3	0.43
210–240	27	8	0.30	140–160	16	2	0.12
Total	698	160	0.23	Total	311	56	0.18

function of breath-holding time are quantitatively similar to the data reported by Lanphier and Rahn (19) and by Craig and Harley (8) on 1 min breath holding and are qualitatively similar to those by others (16, 33) on longer breath holds. Moreover, the same pattern has been shown in animals (2, 4, 11, 28) during breath holding.

To quantify the alveolar exchange of O_2 and CO_2, the amounts of these gases in the lungs ($V_{L_{O_2}}$ and $V_{L_{CO_2}}$) at various stages of breath holding were estimated by multiplying the fractional concentration of alveolar gas by the lung volume (V_L). The latter value was estimated from the relative increase in the alveolar N_2 concentration (8, 19), assuming that the volume of N_2 in the lungs remains constant during breath holding.[1] The difference in $V_{L_{O_2}}$ (or $V_{L_{CO_2}}$) between two successive sampling times, designated as $\Delta V_{L_{O_2}}$ (or $\Delta V_{L_{CO_2}}$), should represent the volume of O_2 (or CO_2) exchanged during the respective period of breath holding. Values of $\Delta V_{L_{O_2}}$ or $\Delta V_{L_{CO_2}}$ computed on the basis of data shown in Table 2 are summarized in Table 3. It is evident that the rate of O_2 removal from the lungs during breath holding was considerably greater than that of the addition of CO_2 to the lungs. Moreover, the transfer of CO_2 from the blood to the lungs was almost negligible after 1 min of breath holding with TLC and after 20 sec with FRC, in agreement with the finding reported by previous workers (8, 19). The overall alveolar gas exchange ratio (R) during breath holding was approximately 0.2, regardless of the initial lung volume.

A comparison of cumulative $\Delta V_{L_{O_2}}$ with $\dot{V}_{O_2}$ (247 ml/min, STPD) indicates that the lungs were supplying a sufficient amount of O_2 during the first 2 min of breath holding with TLC and during the first 40 sec with FRC (left side of Fig. 2). By the end of maximal breath holding, the total amount of O_2 removed from the lungs was equivalent to 70% and 47% of $\dot{V}_{O_2}$ during breath holding with TLC and FRC, respectively. In other words, the tissue has to receive the rest of the O_2 supply (about 300 ml in both TLC and FRC groups) from the blood O_2 reservoir, the size of which is estimated to be approximately 900 ml (26). Figure 2 (right side) also indicates that the major portion of CO_2 produced during breath holding was retained in the blood and other tissues. On the average, 630 ml of CO_2 (about 80% of total CO_2 produced, assuming that $\dot{V}_{CO_2} = 0.8 \times \dot{V}_{O_2}$ during breath holding) are retained by the end of 4 min breath holding with TLC and 470 ml (about 90% of total CO_2 produced) by the end of 160 sec breath holding with FRC. Such a marked retention of CO_2 in the tissue during simple breath holding or during breath-hold diving has been reported by many previous workers (8, 9, 19, 21, 25, 29). Naturally any change in $\dot{V}_{O_2}$ or $\dot{V}_{CO_2}$ during breath holding would slightly alter the above approximation. However, there is no direct evidence in man that the metabolic rate changes during breath holding, although certain indirect evidence for reduction has been given recently (4, 5).

Values of cumulative $\Delta V_{L_{O_2}}$ and $\Delta V_{L_{CO_2}}$ obtained during the 1st min of breath holding in the present work are quite similar to those obtained by Lanphier and Rahn (19) but are somewhat lower than those reported by Craig and Harley (8). This small discrepancy is most likely due to the difference in the way the initial volume of O_2 and CO_2 in the lung is calculated. In the present work as well as in the work of Lanphier and Rahn (19), zero time was defined as the time at which the first sample was obtained after a maximal expiration followed by an inspiration. However, Craig and Harley (8) pointed out recently that this would tend to underestimate the magnitude of gas exchange, especially $\Delta V_{L_{CO_2}}$, because of the rapidity with which CO_2 floods into the alveoli in a breath hold beginning at TLC.

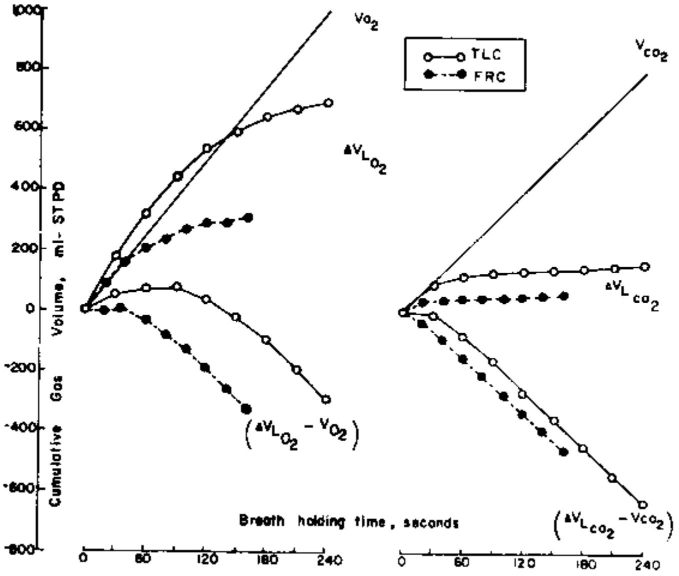

FIG. 2. Cumulative volume of alveolar gas exchange during breath holding. Left and right graphs refer to the O_2 and CO_2 exchange, respectively. See text for calculation of each parameter.

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[1] Strictly speaking, a progressive increase in $P_{A_{N_2}}$ during breath holding would result in a diffusion of N_2 from the lungs to the blood. However, this amount of N_2 loss is very small because of low solubility (1.3 vol % per 1 atm P_{N_2}) of this gas. A rough estimate based on the assumptions that the mean P_{N_2} gradient across the alveolar-capillary membrane, as calculated by the following formula,

$$\left(\frac{P_{A_{N_2}} \text{ at 4 min BH} - P_{A_{N_2}} \text{ at zero time}}{2} - P_{A_{N_2}} \text{ at zero time}\right)$$

is 34 mm Hg and the pulmonary blood flow is 20 liters/4 min indicates a loss of 12 ml of N_2 during 4 min breath holding. This amount is negligible as compared to 4,200 ml of N_2 present in the lungs at the beginning of breath holding.

TABLE 4. *Alveolar and arterial blood gas composition before, at beginning (0 min), and at 2 min breath holding (BH)*

Measurements	Before BH	0 min BH*	2 min BH†
$P_{A_{O_2}}$, mm Hg	111 ± 4	123 ± 2	58 ± 3
$P_{a_{O_2}}$, mm Hg	94 ± 3	98 ± 3	48 ± 2
$(A\text{-}a)O_2$, mm Hg	17 ± 2	25 ± 3	10 ± 3
$P_{A_{CO_2}}$, mm Hg	34 ± 3	25 ± 1	48 ± 2
$P_{a_{CO_2}}$, mm Hg	34 ± 2	31 ± 1	43 ± 2
$(a\text{-}A)CO_2$, mm Hg	0 ± 1	6 ± 1	−5 ± 1
$C_{a_{O_2}}$, vol %	19.4 ± 0.1	19.7 ± 0.1	16.2 ± 0.3
$C_{a_{CO_2}}$, vol %	45.4 ± 1.1	43.1 ± 0.9	50.8 ± 0.6
Art blood pH	7.44 ± 0.01	7.46 ± 0.01	7.36 ± 0.01

Each value represents the mean (± SE) of 13 measurements on 6 subjects. * Samples obtained immediately after a full inspiration preparatory to breath holding. † All values obtained at 2 min BH were significantly different from the corresponding values obtained at 0 min BH ($P < 0.05$ for paired difference).

B) Blood gas composition. When the artery and vein were catheterized, the subjects became somewhat apprehensive and hence the experiment was limited to 2 min breath holding with TLC. Results of analysis of simultaneously collected alveolar gas and arterial blood samples from six subjects are summarized in Table 4. Before breath holding, alveolar P_{O_2} was somewhat higher while P_{CO_2} was lower than usual, suggesting that the subject tended to hyperventilate slightly, probably as a result of apprehension. There was a substantial (A-a) gradient for O_2 but not for CO_2.

At 2 min of breath holding, alveolar P_{O_2} and P_{CO_2} were 58 and 48 mm Hg, respectively, in agreement with the earlier series (Fig. 1). Both P_{O_2} and P_{CO_2} of arterial blood were significantly lower than those of alveolar gas, the difference being 10 mm Hg for O_2 and 5 mm Hg for CO_2. A similar reversal in the CO_2 gradient across the alveolar-capillary membrane was also noted in another series of experiments (Fig. 3). Despite such a reversal of CO_2 gradient, there was no evidence for the reversal of usual CO_2 movement although CO_2 loss to the lung decreases to near zero after 1 min of breath holding (Table 3). It should, however, be realized that, even though blood and alveolar samples were obtained simultaneously in respect to time, they are not really simultaneous samples from the point of view of gas exchange because the arterial site is removed in time from the events at the alveolar-capillary membrane. When gas exchanges are rapid this can make quite a difference as compared to steady-state conditions (7). As pointed out earlier, the rate of CO_2 exchange after 1 min of breath holding is very low and hence the small differences in real sampling time can be ignored in the present work. In other words, the observed reversal of CO_2 gradient across the alveolar-capillary membrane seems to be real. Similar reversal of CO_2 gradient has been reported by Gurtner et al. (14) under conditions of no gas exchange. These authors also proposed a mechanism for the development of this interesting phenomenon.

During 2 min breath holding the arterial O_2 content decreased by 3.5 vol % while CO_2 content increased by 7.7 vol %. Such a greater increase in CO_2 content coupled with a lesser reduction in O_2 content was actually expected because the lungs supplied a considerable amount of O_2 into the blood while removing a rather small amount of CO_2 from the blood during breath holding (Table 3 and Fig. 2). Despite the considerable retention of CO_2 in the blood, the arterial blood pH decreased by only 0.10 probably because of an increase in the concentration of reduced hemoglobin which can carry more CO_2 and H^+ at a given P_{CO_2} as compared to oxyhemoglobin (21, 22).

To assess the time course of changes in the blood gas during prolonged breath holding, both alveolar gas and arterial blood samples were simultaneously collected every 30 sec during 4 min breath holding (TLC) in two subjects who were less apprehensive of the catheterization procedure. As shown in Fig. 3, arterial P_{O_2} decreased continuously during breath holding, reaching 20 mm Hg at the end. On the average, the arterial P_{O_2} was approximately 10 mm Hg lower than alveolar P_{O_2} throughout the entire breath-holding period. Arterial P_{CO_2}, which was higher at zero time than alveolar P_{CO_2}, also became consistently lower than alveolar P_{CO_2} after 60 sec of breath holding. The maximum

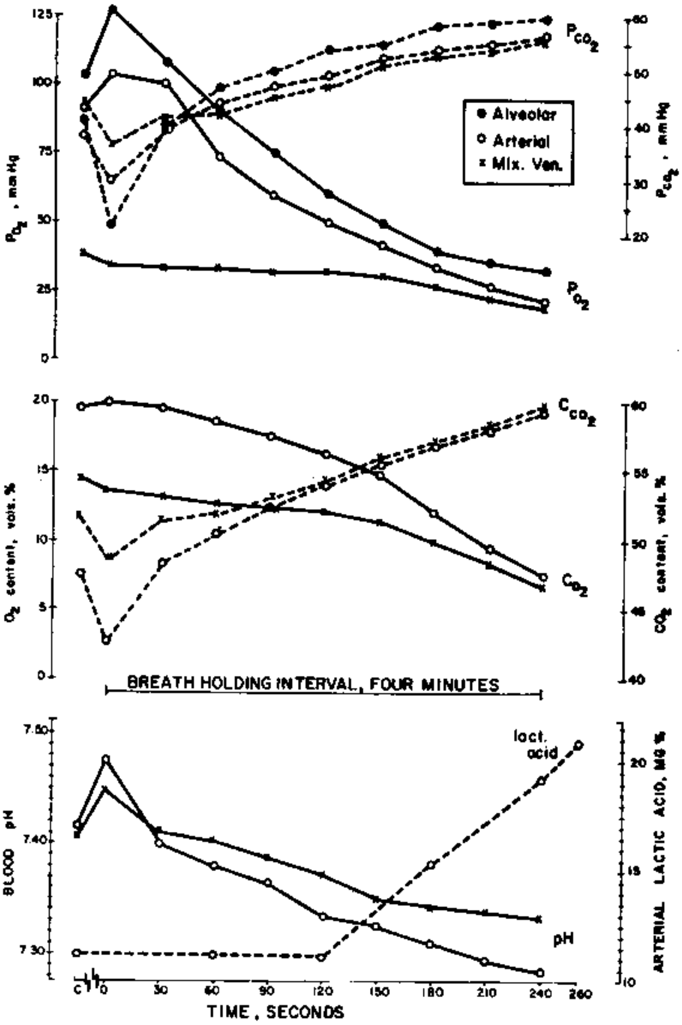

FIG. 3. Alveolar and arterial and mixed venous blood gas composition during breath holding. Each point represents data obtained from two subjects. See text for calculation of mixed venous blood gas composition.

(A-a)CO_2 gradient was as high as 5 mm Hg. Arterial pH declined continuously during breath holding, with the lowest level of 7.29 at the end. Arterial O_2 content decreased slowly during the first 2 min after which it decreased at a faster rate. This finding is consistent with the time course of $\Delta V_{L_{O_2}}$ during breath holding (see the left side of Fig. 2). Arterial CO_2 content increased sharply during the first 30 sec after which it increased linearly but at a lower rate than before. During 4 min breath holding, arterial O_2 content reduced by 12.5 vol % while the CO_2 content increased by 17.0%. Again, such a great increase in arterial CO_2 content in the face of a meager increase in P_{CO_2} is due to an increase in the concentration of reduced hemoglobin during prolonged breath holding.

The gas composition of mixed venous blood was estimated in the latter series by using the following Fick principle:

$$\dot{V}_{L_{O_2}} = \dot{Q}\,(Ca_{O_2} - C\bar{v}_{O_2})$$

or

$$\dot{V}_{L_{CO_2}} = \dot{Q}\,(C\bar{v}_{CO_2} - Ca_{CO_2})$$

In using these formulas, the values of $V_{L_{O_2}}$ (or $V_{L_{CO_2}}$) were first computed for each 30-sec interval according to the procedure described in the previous section and plotted as a function of breath-holding time. The slope of these curves at a given time (i.e., $\dot{V}_{L_{O_2}}$ or $\dot{V}_{L_{CO_2}}$) was determined and used, assuming that at any time the blood R is equal to the alveolar R (19). For the calculations of control $C\bar{v}_{O_2}$ and $C\bar{v}_{CO_2}$ before breath holding, the measured values of resting $\dot{V}_{O_2}$ (211 ml/min, STPD) and the estimated $\dot{V}_{CO_2}$ (0.8 × $\dot{V}_{O_2}$) were used, respectively. The initial lung volume was 5,800 ml (BTPS) in these two subjects. The cardiac output, $\dot{Q}$, was estimated to be 5 liters/min and was obtained before breath holding. As will be shown in the subsequent section, the cardiac output does not change significantly, at least within 2 min of breath holding (Table 6). Actually determined values of Ca_{O_2} or Ca_{CO_2} as shown in Fig. 3 were also used. Once $C\bar{v}_{O_2}$ and $C\bar{v}_{CO_2}$ were estimated from the above formulas, the mixed venous P_{O_2}, P_{CO_2}, and pH were then determined by using the nomogram of Dill et al. (10). These estimates of mixed venous blood gas and pH are included in Fig. 3 for comparison. The O_2 pressure and content of mixed venous blood did not change appreciably until 2 min of breath holding after which they began to fall. At the end of 4 min, the O_2 pressue and content of mixed venous blood became almost equal to those of arterial blood. On the other hand, the mixed venous P_{CO_2} became almost equal to the arterial P_{CO_2} at 30 sec of breath holding although it became slightly lower than the arterial P_{CO_2} during the rest of breath-holding period. The difference in the CO_2 content between mixed venous and arterial blood also became smaller and eventually disappeared after 90 sec of breath holding. As a result of such anomalous behavior of mixed venous P_{CO_2}, mixed venous pH became higher than arterial pH. It should be cautioned, however, that although the cardiac output did not change during the first 2 min of breath holding (Table 6) it could change toward the end of breath holding when the blood P_{O_2} is exceedingly low. If such a change occurs, one would expect the mixed venous blood gas composition to be different from the above estimate. A more systematic analysis of cardiac output during a prolonged breath holding is needed.

The pattern of changes in blood gas during breath-holding with air as described above confirms and extends the work of Lanphier and Rahn (19). Although they did not analyze the blood gas composition, they speculated on theoretical grounds that the arterial P_{CO_2} rapidly becomes equal to the mixed venous P_{CO_2} and thereafter exceeds it by an increasing margin. In agreement with the above prediction, the arterial P_{CO_2} indeed increased rapidly during the first 20 sec of breath holding to the mixed venous level and thereafter exceeded it (Fig. 3). However, in contrast to the prediction, the difference in P_{CO_2} between the arterial and the mixed venous blood did not become greater toward the end of breath holding. The present work also qualitatively agrees with the observation made by Mithoefer (21) on the dog during breath holding with O_2. Within 4 min after the onset of breath holding, the gas exchange ratio had fallen to 0.26, the arterial blood was more acidic and had a slightly higher P_{CO_2}, though the venous blood still carried a higher content of the gas. Interestingly enough, Mithoefer also observed a reversal of CO_2 movement at 20 min of breath holding in this dog (21). However, no evidence for this reversal of CO_2 movement was found in the present work.

During 4 min breath holding the two subjects lost as much as 657 ml (STPD) O_2 from the lungs while consuming 850 ml of O_2. In other words, the blood must have supplied approximately 200 ml O_2 to the tissue during this period to meet the demand. Calculations based on the assumption that the arterial and mixed venous blood volumes are 1,250 and 3,750 ml, respectively, indicated a reduction of blood O_2 stores by 415 ml (Table 5). Considering all the assumptions involved for the calculation, this is rather close to the value predicted above (also see Fig. 2). As will be described later there was a slight progressive increase in the lactic acid concentration of arterial blood after 2 min of breath holding (Fig. 3), indicating that the tissues began anaerobic metabolism toward the end of breath holding as the O_2 supply was decreasing steadily. Similar calculations on the changes of CO_2 stores indicate that the lungs increased their CO_2 volume by 231 ml while the tissues produced approximately 680 ml of CO_2 during 4 min breath holding. This means that the blood and other tissues must have stored approximately 450 ml of CO_2. Calculations based on the same assumptions as above showed an increase of blood CO_2 store by approximately 600 ml. In other words, increases in the level of CO_2 stores in both the lungs and the blood account for the amount of CO_2 produced during a prolonged breath holding. However, this should not be taken to mean that there is no storage of CO_2 in tissues (other than the blood) during breath holding. Since the tissue P_{CO_2} should increase during breath holding, a fraction of CO_2 produced must be stored in the metabolizing tissue. The present work suggests, however, that this storage of CO_2 in the tissue is far less than that in the blood at least during breath holding with air. Other investigators (4, 6, 12, 13, 20) also reported that the volume of distribution of CO_2 is rather small during short-term experiments (<10 min). The overall storage capacity of CO_2 on the basis of increases in alveolar P_{CO_2} during 4 min breath holding was 0.35 ml/kg per mm Hg, in agreement with other reports on short-term experiments (12, 13, 17, 22).

Changes in the alveolar and blood gas composition during breath holding have been excellently explained by Lanphier and Rahn (19) and more recently by Mithoefer (22) who visualizes two interconnected, self-perpetuating cycles of gas exchange, one in the lungs, one in the blood and tissues (see Fig. 8 of ref 22). Explanations put forward by these authors can account for the findings obtained in the present work and hence we will briefly recapitulate these views in the light of present data. In breath holding with air, O_2 initially represents less than 20% of the lung volume. Since a large fraction of N_2 is present, uptake of O_2 progressively reduces the concentration and partial pressure of O_2 in the lungs. As a result, the O_2 saturation of arterial blood decreases. However, the transfer of O_2 from lungs to the blood is not appreciably affected during the early phase of breath holding because of the relatively high alveolar P_{O_2}. As the level of P_{O_2} drops further, the transfer of O_2 from lungs to the blood decreases progressively as the result of which the arteriovenous O_2 difference becomes smaller and eventually disappears by the end of 4 min of breath holding. Moreover, during the later phase of breath holding, the tissue demand for O_2 is primarily met by the blood O_2 store. The large initial inspiration preparatory to breath holding lowers $P_{A_{CO_2}}$ markedly and CO_2 is transferred to the lungs at a rapid rate during the very early phase of breath holding, thereby rapidly increasing the level of $P_{A_{CO_2}}$. As $P_{A_{CO_2}}$ increases, the transfer of CO_2 from the blood to lungs decreases as the result of which the P_{CO_2} gradient between the mixed venous and arterial blood falls and eventually disappears within 30 sec of breath holding. However, the transfer of O_2 still continues at a much higher rate as compared to that of CO_2 during the rest of breath-hold period (Table 3), thus resulting in a decrease in lung volume. This volume loss concentrates the CO_2 present in the lung, thereby raising $P_{A_{CO_2}}$ (and $P_{a_{CO_2}}$) in spite of the fact that the total amount of CO_2 in the lung increases only slightly. In addition, uptake of O_2 by hemoglobin reduces the CO_2 capacity of the venous blood (Haldane effect) and thus tends to raise $P_{a_{CO_2}}$. In other words, both the Haldane effect and the elevation of $P_{A_{CO_2}}$ by lung volume shrinkage (i.e., the cycle in the lungs) are evident contribuotrs to the fact that alveolar-arterial P_{CO_2} exceeded the mixed venous P_{CO_2} after 30 sec of breath-hold period (Fig. 3).

On the other hand, the rise in alveolar-arterial P_{CO_2} decreases the transfer of CO_2 from the tissue into the blood, thus suppressing a rise in venous CO_2 content and pressure. In addition, the Haldane effect of hemoglobin reduction in the tissue capillaries also lowers the P_{CO_2} of venous blood for a given CO_2 content. Thus a rise in venous P_{CO_2} comparable to that which has occurred in arterial blood is suppressed by the blood-tissue cycle. The disparity in the rate of rise of arterial and mixed venous P_{CO_2} lowers the gradient for CO_2 transfer still further, and the cycle in the lung is reinforced by that in the blood and tissues. Further details of these events, including quantitative analysis of Haldane effect as well as the significance of R, are given by the above authors (19, 22).

TABLE 5. *Changes in amount (ml, STPD) of O_2 and CO_2 in circulating blood during 4 min breath holding (BH)*

Blood Gas	A: Begin of BH	B: 4 min of BH	B − A
Arterial O_2	250	94	−156
Mix ven O_2	510	251	−259
Total	760	345	−415
Arterial CO_2	535	743	+208
Mix ven CO_2	1,820	2,220	+400
Total	2,355	2,963	+608

Arterial and mixed venous blood volumes were assumed to be 1,250 and 3,750 ml, respectively. Values of appropriate blood gas contents were taken from Fig. 3.

C) Cardiac output. After the measurements of cardiac output and of the arterial and the right atrial pressures, the subjects held breath with TLC. At 1.5 min of breath holding, Cardio-Green was injected into the right atrium. The subjects continued to breath hold until the dye-dilution curve was completed. It took approximately 30 sec. During this period, the recording of arterial blood pressure was discontinued. The ECG was taken throughout the entire period. Duplicate measurements were made in five subjects and the results are summarized in Table 6. For the calculation of total peripheral resistance during breath holding, the values of the mean arterial and right atrial pressures were taken from the separate experiments (see *section D* and Fig. 4). On the average, the cardiac output as well as the cardiac index tended to increase during breath holding but the changes were not statistically significant ($P > 0.05$). On the other hand, the heart rate tended to decrease[2] while the stroke volume tended to increase during breath holding ($P > 0.05$). Although the driving pressure (i.e., mean arterial pressure minus right atrial pressure) increased significantly during breath holding, it was not big enough to indicate a significant increase in the total peripheral resistance. However, there was a clear tendency for the peripheral resistance to increase during breath holding, which may be attributed to peripheral vasoconstriction (3, 15, 24, 25, 32). Perhaps as a result of peripheral vasoconstriction, the central blood volume (estimated by the flow times the mean transit time) tended to increase somewhat.

Similar results have been reported by Paulev (25) and Smith and Rankin (31) when their subjects held breath without Valsalva maneuver. Both the cardiac output and the stroke volume tended to increase in these subjects during 20–70 sec breath holding. On the other hand, when breath holding was coupled with conscious Valsalva maneuver, both the cardiac output and the stroke volume decreased significantly. As will be shown in the following section, the magnitude of increase in the intrathoracic pressure (as estimated by the intraesophageal pressure) was rather small,

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[2] Changes in the heart rate in this series were not significant. On the other hand, significant reductions in the heart rate were noted in the subsequent series (see Fig. 4) where the subjects were not disturbed during the experiment. It is entirely possible that the subjects respond differently during the procedure to measure the cardiac output.

TABLE 6. *Cardiac output, stroke volume, and total peripheral resistance before and during breath holding (BH)*

Measurements	Before BH	During BH*
Cardiac output		
liters/min	5.77 ± 0.94	6.12 ± 0.97
liters/min per m^2	3.17 ± 0.47	3.39 ± 0.47
Heart rate/min	85 ± 10	81 ± 7
Stroke volume		
ml	72 ± 14	78 ± 13
ml/m^2	39 ± 7	42 ± 7
Mean art press, mm Hg†	97 ± 14	126 ± 9‡
Mean rt art press, mm Hg	2 ± 4	10 ± 1‡
Tot periph res, mm Hg/ml per sec	1.08 ± 0.18	1.26 ± 0.28
Mean transit time, sec§	16.3 ± 1.4	18.8 ± 2.4
Central blood vol		
liters	1.58 ± 0.30	1.81 ± 0.14
liter/m^2	0.87 ± 0.16	1.00 ± 0.07

Each value represents the mean (± SE) of 10 measurements on 5 subjects. * Measured during 1.5-2 min of breath holding. † Calculated by (diastolic pressure + 3/8 pulse pressure). ‡ Significantly different from the corresponding value obtained before BH ($P < 0.025$). § Transit time from right atrium to the brachial artery.

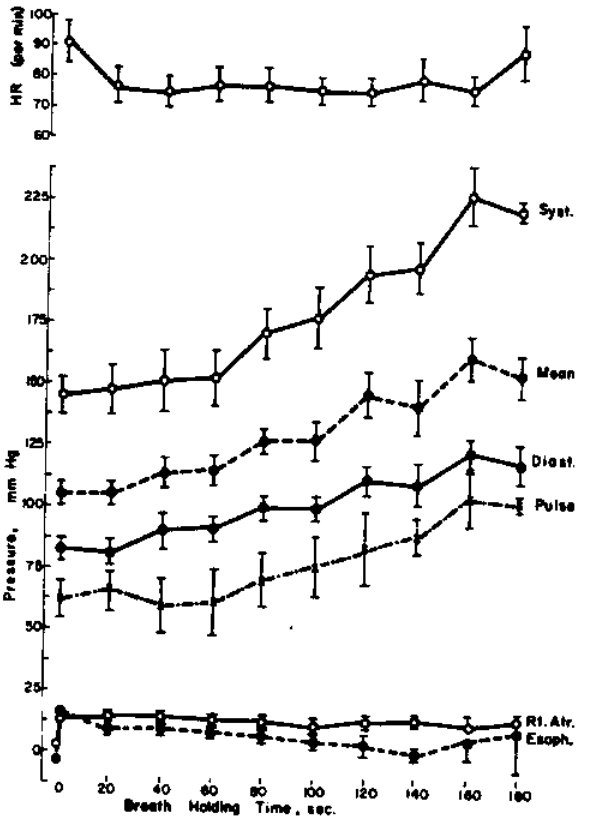

FIG. 4. Changes in heart rate and arterial, right atrial, and intraesophageal pressures during breath holding. Each point is the mean (± SE) of data obtained from six subjects.

indicating that the breath holding in the present work is not coupled with Valsalva.

D) Blood pressure and intraesophageal pressure. The time course of changes in the arterial and the right atrial pressures, the intrathoracic pressure, and the heart rate was studied in five subjects (Fig. 4). In this series, the subjects were not disturbed by any experimental procedures such as sampling of alveolar gas or blood. The subjects were encouraged to continue breath holding as long as possible. All subjects were able to breath hold for 3 min. In general, the arterial blood pressure increased only slightly during the 1st min after which it increased progressively toward the end of breath holding. On the average, the systolic pressure increased to a greater extent than the diastolic pressure causing a progressive increase in the pulse pressure during the later phase of breath holding. The mean arterial blood pressure (as estimated by diastolic pressure plus 3/8 pulse pressure) increased by 65 mm Hg during 3 min breath holding. In agreement with Paulev's finding (25), the right atrial pressure increased instantly with the onset of breath holding to approximately 10 mm Hg and maintained this level throughout the entire breath-holding period. The intraesophageal pressure increased in the same manner as the right atrial pressure at the beginning of breath holding and then gradually decreased to zero at 140 sec. A much more extensive reduction in the intraesophageal pressure was noted by Mithoefer (22) in anesthetized, paralyzed dogs during breath holding with O_2 and was attributed to a reduction in the lung volume. In case of breath holding with air, the lung volume should decrease to a lesser extent as compared to breath holding with O_2, thus giving rise to a lesser reduction in the intraesophageal pressure.

The heart rate decreased by approximately 15% within 20 sec of breath holding and more or less maintained this level during the rest of the breath-holding period. It is important to note that this bradycardia developed in the absence of any significant change in the arterial blood pressure, indicating that the breath-hold bradycardia is not elicited by a baroreflex (15, 16).

The fact that the arterial blood pressure began to increase at a faster rate after 1 min of breath holding is interesting in that at this point the arterial P_{CO_2} begins to level off at a higher level while P_{O_2} begins to decrease rapidly (Fig. 3). This suggests that the primary causes of hypertension during breath holding are central effects of high P_{CO_2} and/or low P_{O_2}. However, a greater increase in the systolic pressure as compared to that in the diastolic pressure indicates that there may be other factors. One possibility is the local effect of CO_2 or lactic acid in the tissue, which would counteract the central effect by inducing local vasodilatation. Another possibility is the increased secretion of catecholamines (especially epinephrine) during breath holding as a result of increases in P_{CO_2} and $[H^+]$ (23).

E) Blood lactic acid. In diving mammals, circulatory shunts bypass the muscle capillaries and other regional

TABLE 7. *Changes in lactic acid concentration of arterial blood during and 20 sec after breath holding (BH)*

Time of Sampling	Lactic Acid Concn, mg/100 ml
Before BH	10.1 ± 1.6
0 min BH	11.2 ± 0.8
2 min BH	11.3 ± 0.8
20 sec after BH	12.1 ± 0.7*

Each value represents the mean (± SE) of 10 measurements on 5 subjects. * Significantly different from that obtained at 2 min BH ($P < 0.025$ for paired difference).

vascular beds during diving (29). As a result, lactic acid formed in these regions during diving is trapped until the end of diving, at which time it enters the circulation. However, Olsen et al. (24) observed a very slight but significant rise in blood lactate during diving in human divers, suggesting some exchange between the muscles and the circulation. The results obtained during 2 min breath holding indicate that blood lactate level did not increase significantly during breath holding. However, a slight increase ($P < 0.05$ for the paired difference) was noted after breath holding (Table 7). Serial blood samplings during 4 min breath holding in two subjects showed that blood lactate level begins to increase linearly after 2 min of breath holding (Fig. 3). Moreover, there was a further increase in blood lactate at 20 sec after breath holding. These findings are in agreement with the report of Olsen et al. (24) and indicate that the extent of circulatory shunts during breath holding in man is much less than that observed in diving mammals during diving.

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Heart rate response to apneic face immersion in hyperbaric heliox environment

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HONG, SUK KI, T. O. MOORE, D. A. LALLY, AND J. F. MORLOCK. *Heart rate response to apneic face immersion in hyperbaric heliox environment.* J. Appl. Physiol. 34(6): 770–774. 1973.—Bradycardial responses to simple breath holding (BH) and apneic face immersion (FI) in 30° C water were studied in five divers in 4.0, 6.6, and 12.5 Ata heliox environments and in 1 Ata air. Maximal percent reduction in heart rate (Δ HR_{max}) during apnea at 1 Ata averaged 5.3 and 15% during BH and 30° C FI, respectively. As ambient pressure increased, Δ HR_{max} increased to 22% (BH) and 37% (30° C FI) at 12.5 Ata. During both BH and FI, Δ HR_{max} was greater at 6.6 and 12.5 Ata than in 1 Ata air ($P < 0.05$). Δ HR_{max} was correlated with ambient pressure in the FI series ($r = 0.485$, $P < 0.01$). Δ HR_{max} during 30° C FI in 4.0, 6.6, and 12.5 Ata heliox were equivalent to those in 1 Ata air during FI in 12.5, 10, and 6° C water, respectively. In addition, eight male subjects performed BH and FI experiments after breathing 75% He-25% O_2 at 1 Ata for 15 min. In the lower heart rate (HR) group (mean resting HR $\simeq$ 60 min^{-1}, 4 subj), Δ HR_{max} during BH and FI were generally potentiated. Resting HR was unchanged. In the higher HR group (mean resting HR $\simeq$ 80 min^{-1}, 4 subj), resting HR was lower during He-O_2 breathing while Δ HR_{max} in BH and FI tended to be attentuated. Breathing 75% Ar-25% O_2 did not alter resting HR or Δ HR_{max} in the lower HR group. These results are discussed in terms of *1*) altered behavior of thermal receptors and *2*) possible suppression of the sympathetic system by He.

apneic bradycardia; helium

OVER THE LAST 10 years, many factors which alter the magnitude of bradycardial responses have been studied extensively using the apneic face immersion technique. According to current theories (3, 4, 7, 16), there are two major factors which alter the magnitude of apneic bradycardia during diving in man: one is a mechanical factor which is an integral part of the well-known baroreflex and the other a neural factor which seems to be temperature dependent through some unknown mechanism. Despite the fact that actual diving involves changes in the environmental pressure, the possible role of pressure in the development of diving bradycardia has been almost totally neglected.

To gain insight into this question, Moore and his associates in our laboratory previously conducted a series of breath-hold experiments in the open sea at depths to 18 m, as well as face immersion experiments in a dry recompression chamber to simulated depths of 30 m (8). They observed a significant attenuation of apneic bradycardia at depth. However, the interpretation of the data was rather difficult because of an elevation of P_{O_2} at depth. Subsequent studies, in which the level of P_{O_2} was maintained constant, suggested that there are certain mechanical stimuli due to hydrostatic effects which attenuate the bradycardial response to apnea (8). The present investigation was designed to reexamine the role of ambient pressure on the apneic bradycardia in the face of constant P_{O_2}.

METHODS

Hyperbaric experiments. The experiments were carried out in January 1972, during the decompression phase of a 9-day dry heliox saturation dive in the undersea habitat "Aegir" (11). The dive profile involved 17 hr at 8.6 Ata (250 ft) and 43 hr at 16.1 Ata (500 ft) before decompression was carried out at a rate of approximately 100 ft/day. Average environmental data was: $P_{O_2} \approx 220$ mm Hg, $P_{CO_2} < 2.0$ mm Hg, ambient temperature = 27.9–29.0° C, relative humidity = 77.5%. In addition, the thermal cost of saturation diving was studied (9) during the saturation phase of this dive.

The heart rate response to simple breath holding and to apneic face immersion in 30° C water was studied in five male subjects (on the average, 28 years old, 174 cm in height, and 73.5 kg in weight) at 12.5 Ata (375 ft), 6.6 Ata (185 ft), and 4.0 Ata (100 ft). The gas pressure profile of these hyperbaric environments is shown in the top of Fig. 1. The apneic maneuver was conducted in duplicate in a sitting position without previous hyperventilation; however, each experiment began following a single, full inspiration. Heart rates were measured by recording ECG with a Physiograph located outside the habitat. The number of QRS waves for every 10-s period during apnea was compared with that obtained during a 10-s period, before the onset of apnea. Following the saturation dive, control experiments were carried out using the same subjects in air (25° C) at 1 Ata. In this case, apneic face immersions in 25, 15, and 5° C water were carried out in addition to simple breath holding and apneic immersion in 30°C water. In all phases of this study, the duration of apnea was 1 min. In the face immersion experiments, immersion was accomplished immediately following the deep inspiration and was maintained for 1 min.

Normobaric experiments. The effect of breathing He at 1 Ata was studied by comparing the heart rate responses to apnea (with and without face immersion) during periods

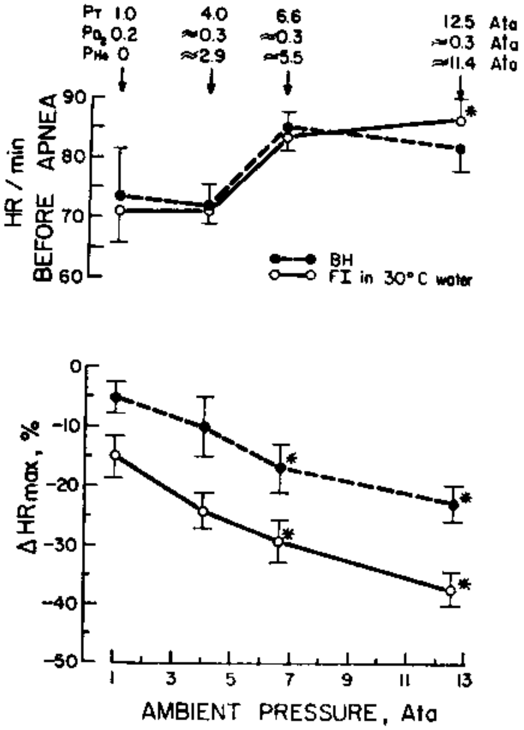

FIG. 1. Resting heart rate during 10 s before the onset of apnea and the maximal apneic bradycardia (ΔHR_{max}) as a function of ambient pressure. Each point represents the mean of 10 experiments in 5 divers. Vertical bars in this and other figures ±1 SE. *Value is significantly different ($P < 0.05$) from corresponding value at 1 Ata.

of breathing air and 75% He-25% O_2. The water temperatures for face immersion were 30, 25, 15, and 5° C, as above. Eight male subjects (averaging 29 years old, 172 cm in height, and 66 kg in weight) were used in this series of experiments. Two subjects occasionally scuba dive. Using the same breathing apparatus for all gases, the subject breathed the given gas for 15 min, after which the various apneic maneuvers were executed in random order. At the end of each 1-min apnea, the subject returned to the breathing apparatus and rested until his heart rate returned to normal prior to the next apnea. This procedure was followed for air, He-O_2 and also for a third gas, 75% argon-25% O_2, which was used to test the specificity of any He effect.

RESULTS

Hyperbaric experiments. The average heart rates during the 10 sec immediately before the onset of each apneic maneuver and the maximal bradycardial response to apnea (ΔHR_{max}) are shown as a function of ambient pressure (Ata) in the top and the bottom of Fig. 1, respectively. The ΔHR_{max} is expressed as percent of control.

The heart rate immediately before the onset of apnea averaged 70–75/min in 1 Ata (air) and 4 Ata, respectively, but increased to approximately 85/min in 6.6 and 12.5 Ata. However, the only significant increase was noted in 12.5 Ata before face immersion ($P < 0.05$).

In air at 1 Ata, the ΔHR_{max} averaged only 5.3% during simple breath holding and 15% during face immersion in 30° C water. However, ΔHR_{max} increased progressively as the ambient pressure increased, reaching 22% during breath holding and 37% during face immersion in 30° C water at 12.5 Ata. In both apneic maneuvers, ΔHR_{max} at 6.6 and 12.5 Ata was significantly greater than the respective values at 1 Ata air ($P < 0.05$). Moreover, there was a significant correlation between ΔHR_{max} and the ambient pressure for the face immersion series ($r = 0.485$, $P < 0.01$).

ΔHR_{max} during face immersion in 1 Ata air, expressed as a function of water temperature, is shown on the right of Fig. 2. In agreement with previous reports (7, 8, 16), ΔHR_{max} increased as the water temperature was lowered.

Normobaric experiments. Preliminary analysis of the results obtained from all eight subjects employed in this series indicated that there are two distinct responses to He breathing at 1 Ata which depend upon the resting heart rate in air. The results obtained from four subjects with a resting heart rate of approximately 60/min (lower HR group) and the other four with a resting heart rate of approximately 80/min (higher HR group) are shown, respectively, on the left and right of Fig. 3. The lower HR group maintained a stable heart rate during both air and He breathing and He consistently potentiated the bradycardial response to all apneic maneuvers except in the case of face immersion in 5° C water. This potentiation was significant for simple breath holding ($P < 0.05$) and was marginally significant for face immersion in 15° C ($0.05 < P < 0.10$). In contrast, the higher HR group showed an increase in heart rate during air breathing before face immersion but maintained a relatively stable, lower heart rate during He breathing; moreover, the bradycardial response, which was not altered during simple breath holding, tended to be attenuated during face immersion in water of 30, 25, and 15° C. This attenuation was significant during face immersion in water of 15° C ($P < 0.05$).

To study whether the potentiation of apneic bradycardia observed in the lower HR group during He breathing is due to the replacement of N_2 with He, the effect of breathing Ar-O_2 was studied on apneic bradycardia in the lower HR group. The resting heart rate before apnea was maintained at 60–65/min in all cases and the magnitudes of ΔHR_{max} for simple breath holding and for face immersion in water of various temperatures were not significantly altered.

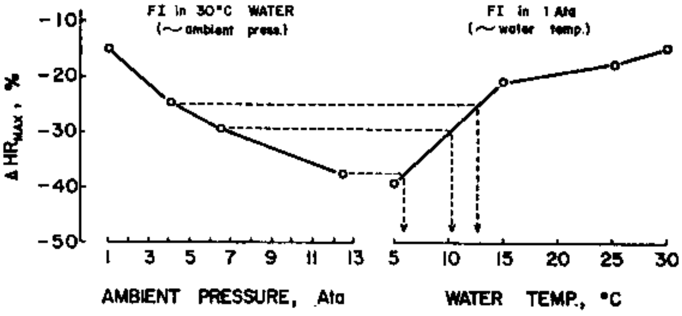

FIG. 2. *Left:* maximal bradycardial response to face immersion in water of 30° C as a function of ambient pressure. (This graph is taken from Fig. 1.) *Right:* maximal bradycardial responses to face immersion in water of various temperatures in 1 Ata air. Each point is the mean of 10 experiments in 5 divers.

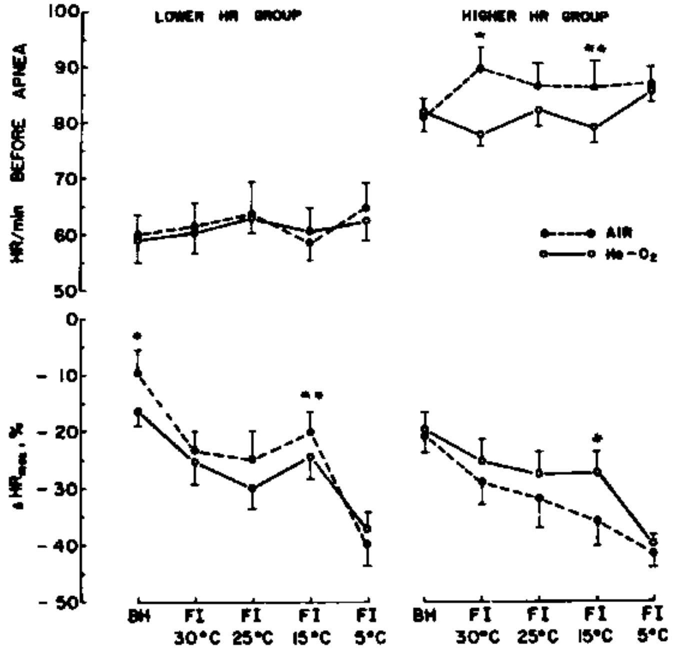

FIG. 3. Resting heart rate during 10 s before the onset of apnea with air or He-O_2 in 1 Ata air as a function of face immersion water temperature in lower (*left*) and higher (*right*) heart rate groups. Each point is the mean of 8 experiments in 4 subjects. *Significant difference between air and He-O_2 series ($P < 0.05$ for each pair). **Marginally significant difference between air and He-O_2 series ($0.05 < P < 0.10$ for each pair).

DISCUSSION

The results obtained in the hyperbaric heliox environment clearly indicate a significant potentiation of the bradycardial response to apnea (see Fig. 1). Although the resting heart rate prior to the onset of apnea tended to be slightly higher at 6.6 and 12.5 Ata, there was no correlation between the resting heart rate and the degree of bradycardia encountered in this series. For instance, there was no difference in the resting heart rate at pressures between 1 and 4 Ata, yet the bradycardial response increased from 5.3 ± 2.3 (SE) % at 1 Ata to 10.2 ± 4.9 at 4 Ata in the case of simple breath holding and from 15.1 ± 3.7 to 24.0 ± 3.0% in the case of face immersion in 30° C water. Similarly, the bradycardia at 12.5 Ata was consistently greater than that at 6.6 Ata in the absence of any difference in the resting heart rate between the two pressures. This dependence of bradycardial response on ambient pressure was more evident during face immersion in water, in which case a significant correlation between the two variables was found.

These findings are in contrast to our earlier findings in which the bradycardial response to face immersion was attenuated in a 4 Ata air environment. This suggests that the potentiation of apneic bradycardia observed in the present work is attributable to the presence of He in the environment and not to the high ambient pressure per se.

A hyperbaric heliox environment may be characterized by its greater heat dissipating capacity as compared to air, for both the density and specific heat of the gas at 12.5 Ata are about 3 times greater than those of air at 1 Ata (Table 1), while its conductivity is 6 times greater. Consequently, the heat capacity of the environmental gas at 12.5 Ata is about 9 times greater than that of 1 Ata air, while the convective character, to use Webb's definition (18), is 44 times greater. Because of this, divers in a hyperbaric heliox environment are known to feel cold even at the ambient temperature of 26° C (18). We also noted that the skin and respiratory heat loss at 27° C was significantly greater in the hyperbaric environment than in 1 Ata air (9). However, all subjects felt comfortable at the ambient temperature of 28 °C at which all the apneic experiments were carried out.

TABLE 1. *Gas composition and its thermal characteristics in various ambient pressures*

	1 Ata	4 Ata	6.6 Ata	12.5 Ata
A. Gas composition, %				
O_2	20	7.4	4.5	2.4
N_2	80	20.0	11.7	6.2
He	0	72.6	83.8	91.6
B. Thermal characteristics of gaseous environment				
ρ, g/l	1.150	1.755	2.177	3.134
c_P, cal/g·°C	0.256	0.517	0.657	0.836
ρc_P, cal/l·°C	0.295	0.908	1.431	2.619
Relative heat capacity	1.0	3.1	4.9	8.9
Convective character*	1.0	12.9	22.5	44.0

ρ = density; c_P = specific heat at constant pressure; ρc_P = heat capacity. * See Webb (18).

One of the most important factors found to consistently potentiate the magnitude of face immersion bradycardia is low water temperature (7, 8, 16). The results obtained from the same subjects indicate the bradycardial response to face immersion in 30° C in 4.0, 6.6, and 12.5 Ata heliox environments is equivalent to that observed during face immersion in water of 12.5, 10, and 6° C, respectively, in 1 Ata air (see Fig. 2). As shown in Fig. 4, the 1 Ata air-equivalent water temperature obtained from Fig. 2 decreases exponentially as a function of ambient pressure. In other words, the bradycardial response to face immersion in relatively warm water in a hyperbaric environment is equivalent to that in much colder water in 1 Ata air, suggesting that the facial cold receptors may respond differently to a given thermal stimulus in hyperbaric heliox environments.

If the hypothesis is correct that the facial cold receptors respond differently to a given thermal stimulus in hyperbaric heliox environments, one might further speculate that the presence of He in the external rather than the internal environment is responsible for this phenomenon. Recently, however, Raymond et al. (13) reported that breathing a 75% He-25% O_2 instead of 75% N_2-25% O_2 at 1 Ata reduced the occurrence of cardiac arrhythmias after ligation of the circumflex coronary artery in anesthetized dogs and also that He reduced the base-line heart rate and the level of plasma catecholamines. In other words, the presence of He in the internal environment seems to interfere with the cardiac excitability through changes in sympathetic activity. If a similar mechanism exists in

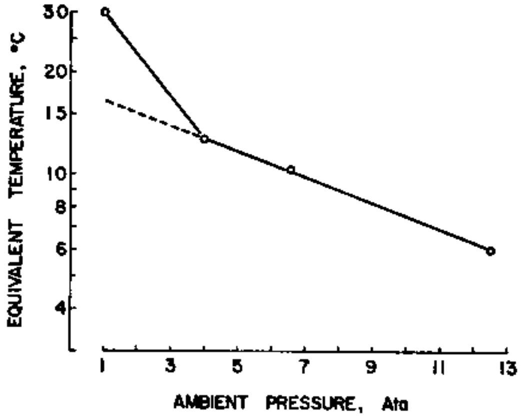

FIG. 4. Face immersion water temperature in 1 Ata which induced the equivalent degree of bradycardia to face immersion in 30° C water in various hyperbaric environments. Points were taken from Fig. 2.

man, one might expect to find a potentiation of apneic bradycardia during $He\text{-}O_2$ breathing, independent of the possible effect of He in the external environment on thermal receptors.

The results obtained from the normobaric series clearly support this hypothesis. In the lower HR group, in which the resting heart rate was not altered by He, the bradycardial responses to simple breath holding and to apneic face immersion were generally potentiated by He except in the case of face immersion in 5° C water (see Fig. 3). In this case, the extreme cold may have stimulated maximally so that the subtle effect of He may have been obliterated. In contrast, the resting heart rate was kept lower during He breathing than during air breathing in the higher HR group, in which the bradycardial response to apnea tended to be attenuated by He. Since the substitution of Ar for He failed to alter either the resting heart rate or the bradycardial response, the above He effects appear to be specific. These findings indicate that the presence of He in the internal environment alone exerts certain subtle effects on cardiac activity, which are compatible with the inhibition of sympathetic activity. However, the difference in response to He between lower and higher heart rate group suggests that the effect of He is somehow dependent upon the balance of intrinsic sympathetic and parasympathetic tones. In a similar study recently conducted in man, Raymond et al. (12) also noted a small but significant reduction in resting blood pressure during He breathing. Rhoades et al. (14) reported a significantly lower heart rate in rats kept in 79% He-21% O_2 environment than in air of 33° C. It is also of interest that the daily excretion of catecholamines tends to decrease in the hyperbaric heliox environment (2, 17, and unpublished data of Moore et al.).

In many saturation dives conducted previously, a consistent reduction in the resting heart rate was noted in man (1, 5, 6, 10, 15). Although the mechanisms underlying this interesting phenomenon are not understood at present, it is possible that the presence of He at high pressure may be responsible. In the present dive, the resting heart rate also decreased during the compression phase but increased somewhat during the early phase of decompression which agrees with the report of Hamilton (5). The latter phenomenon could well be due to the apprehension usually present during the early phase of decompression.

These considerations lead us to speculate that the observed potentiation of apneic bradycardia in the hyperbaric heliox environment may be attributed to suppression of the effect of the sympathetic nervous system. However, the possibility of altered behavior of thermal receptors cannot be completely excluded. More definitive studies are needed to elucidate the mechanism(s) underlying this interesting phenomenon.

The authors gratefully acknowledge five saturation divers who served as subjects.

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Lung volumes and apneic bradycardia in divers

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HONG, S. K., T. O. MOORE, G. SETO, H. K. PARK, W. R. HIATT, AND E. M. BERNAUER. *Lung volumes and apneic bradycardia in divers.* J. Appl. Physiol. 29(2): 172–176. 1970.—Various cardiopulmonary functions and cardiovascular responses during apnea to breaking point were studied in 26 male divers and 11 nondivers (control). There was no significant difference between the two groups in lung volumes, maximal voluntary ventilation, maximal oxygen intake (MOI), resting heart rate, or alveolar gas composition. The average duration of breath holding (BH) was 15 sec longer in divers than controls. Although the apneic bradycardia was more pronounced during face immersion (FI) in cold water (5 C) than during simple BH in air or FI in warm water (30 C) the magnitude of bradycardia in a given apneic maneuver was greater by 10% in the diver than in the control. There was a significant regression between the maximal bradycardia response (ΔHR_{max}) and age in each individual. The slope of the regression line was significantly lower while ΔHR_{max} for a given age was higher in the divers than controls. There was a significant regression between ΔHR_{max} and RV/TLC ratio but the regression lines were not different between the two groups. ΔHR_{max} was poorly correlated with MOI, BH time, or % body fat. $P_{A_{O_2}}$ continuously decreased during apnea, whereas $P_{A_{CO_2}}$ reached a plateau after 1 min up to 4 min of apnea in several subjects.

breath holding; face immersion

A MARKED REDUCTION in heart rate associated with the act of diving was originally observed in diving animals (1, 13, 20, 28) and subsequently in man (12, 18, 19, 29). A similar bradycardia can be induced in the laboratory by simple breath holding or by apneic immersion of the face in water (7, 11, 13). With these simple laboratory maneuvers, the nature of the apneic (or diving) bradycardia has been extensively studied in recent years and several theories have been advanced so far (7, 11–13, 19, 32). Unfortunately, however, the majority of these studies were carried out on subjects who had little diving experience primarily because of difficulty in recruitment. The only systematic study using well-trained divers was carried out recently by Corriol and Rohner (9) who observed an increased bradycardial response to simple breath holding or face immersion. Although several other workers (18, 29) also studied the apneic bradycardia in professional divers, no parallel data on nondivers are available for comparison.

The present investigation was undertaken to study the magnitude of apneic bradycardia in experienced divers. In addition, certain cardiopulmonary functions of these subjects were measured before and during apnea. It will be shown that the diver indeed develops a significantly greater bradycardia during apnea and the possible mechanism(s) will be discussed.

METHODS

Twenty-six male divers served as subjects. They are members of two of many diving clubs in Hawaii and regularly perform skin or SCUBA diving, or both, at least 1 day a week the year round. All but three of these divers are certified by the NAUI (National Association of Underwater Instructors) or the YMCA, or both. They have a history of skin diving for 6.5 years (range of 1–22 years) and of SCUBA diving for 3.9 years (range of 0.3–21 years). Their estimated diving hours are more than 1,000 hr in 4 subjects, 500–1,000 hr in 3, 100–500 hr in 13, and less than 100 hr in the rest. They skin dive to a depth of 46 ft (range of 15–120 ft) and SCUBA dive to 117 ft (range of 40–250 ft). In contrast, the 11 control subjects (male) have no diving experience. Most of these control subjects are members of the Department of Physiology and are occasional swimmers. The age and physical characteristics of subjects are summarized in Table 1. Each subject was invited to the laboratory for the following measurements.

Lung volumes and maximal voluntary ventilation (MVV). Upon arrival in the laboratory, a complete diving history was first taken on each subject during a 30-min rest period after which the following measurements were taken in the standing position. The residual volume (RV) was measured by the three-breath method, as modified by Rahn et al. (25); in each subject the determination was made three times and the smallest value was taken. The vital capacity (VC) was measured on a 13-liter Collins spirometer. For the determination of MVV, each subject was connected to the spirometer with the soda lime removed to minimize the resistance to breathing. The subject was then instructed to breathe in and out of the spirometer for 12 sec at maximal rate and depth. During this maneuver the subject was continuously encouraged to exert himself. The cumulative volume of ventilation was recorded on the fast-running spirogram. Both VC and MVV were determined three to four times and the highest values were taken. All values are expressed in BTPS.

Heart rate and pressor response to apnea. After the measurements of lung volumes and MVV, the subject rested in a sitting position for 10 min during which electrodes and a pressure cuff were attached to record the ECG (standard lead I) and blood pressure (in response to signals from an auscultation microphone built into the pressure cuff). Two to three alveolar gas samples were then collected into a Haldane-Priestley tube.

The subject, sitting in a forward bending position, was instructed to breathe normally, to take a maximal breath and then hold the breath as long as possible. At the breaking point of breath holding, the subject exhaled maximally into a Haldane-Priestley tube for collection of another alveolar gas sample. The ECG was continuously recorded before and during the apneic period while the blood pressure was taken every 30 sec. In the second and third apneic maneuvers, the same breath-hold procedure was coupled with the immersion of face in water of 30 and 5 C, respectively. Each maneuver was repeated twice. The subject was allowed to recover for 3–5 min after each apneic maneuver. Alveolar gas samples were analyzed for O_2 and CO_2 by a Scholander micro-gas analyzer (27).

Maximal oxygen intake (*MOI*). After the breath-holding experiments the MOI was estimated by the method of Åstrand (2) using the bicycle ergometer (AB Cykelfabriken Monark Co., Sweden). Two Beckman electrodes were attached to positions on the sternum for continuously recording the ECG during the 6-min exercise period. On the basis of the work load and the steady-state heart rate during the last minute of exercise, the value of MOI was estimated from the table prepared by Åstrand. This value was subsequently corrected for age, again using the table prepared by Åstrand.

RESULTS

Cardiopulmonary functions. The average values of various cardiopulmonary functions are summarized in Table 2. As compared to the control group, the RV tended to be lower while the VC was higher in the diver and hence the ratio of RV to total lung capacity (TLC) tended to be lower in the diver ($0.1 > P > 0.05$). As expected there was a significant correlation between the RV/TLC ratio and the age of divers ($r = 0.767$, $P < 0.01$), but the majority of control values are also distributed around the regression line for the diver. Neither the MVV, the MOI, nor the resting alveolar gas pressures was significantly different between the two groups. Although the systolic blood pressure was significantly greater in the diver as compared to the control ($P < 0.05$), the magnitude is very small.

Heart rate and blood pressure response to apnea. The average breath-holding time, the initial heart rate (obtained during a 10-sec period preceding a full inspiration preparatory to the breath hold), the maximal percent reduction in heart rate during apnea (ΔHR_{max}) and the blood pressure before and at the end of apnea are summarized in Table 3.

In general, the breath-holding time tended to be longer in the diver as compared to the control subject, although a significant difference between the two groups was noted only during face immersion in water of 30 C. The initial heart rate was approximately 15–20% higher in both groups than the resting rate, suggesting an emotional or psychological tachycardia which precedes the anticipated breath hold.

With the initiation of apnea after a maximal inspiration, the heart rate either slightly increased or decreased during the first 10 sec. After that the heart rate decreased continuously until 30–40 sec of apnea after which it tended to increase again. As compared with the control, this initial reduction in heart rate was better sustained in the diver in

TABLE 1. *Physical characteristics of subjects*

Measurements	Control (N = 11)	Diver (N = 26)
Age, yr	27.9 ± 2.0	30.4 ± 1.8
Height, cm	174.6 ± 2.9	177.3 ± 1.7
Weight, kg	66.4 ± 3.8	77.2 ± 2.0*
Body surface area, m^2	1.81 ± 0.07	1.94 ± 0.03*
Body fat, %†	12.1 ± 1.3	15.3 ± 1.0*

Values are means ± SE. * Significant difference from control ($P < 0.05$). † Estimated by method of Sloan (30), which is based on the measurement of skinfold thickness of the front thigh and subscapular regions.

TABLE 2. *Various cardiopulmonary functions of subjects*

Measurements	Control	Diver
A. Lung volumes and other pulmonary functions		
RV, ml BTPS	1,639 ± 100	1,486 ± 67
VC, ml BTPS		
Measured	5,188 ± 300	5,306 ± 172
Predicted	4,301 ± 101	4,298 ± 50
Measured/predicted, %	120 ± 5	123 ± 3
TLC, ml BTPS	6,827 ± 367	6,797 ± 197
RV/TLC, %	24.1 ± 1.0	22.0 ± 0.9
MVV, liters/min BTPS		
Measured	224 ± 10	223 ± 8
Predicted	131 ± 6	138 ± 3
Measured/predicted, %	173 ± 7	162 ± 5
Max O_2 intake		
liters/min STPD	2.80 ± 0.30	3.06 ± 0.12
ml/min per kg STPD	42.1 ± 3.1	40.1 ± 1.7
Resting $P_{A_{O_2}}$, mm Hg	104 ± 2	106 ± 1
Resting $P_{A_{CO_2}}$, mm Hg	39 ± 1	37 ± 1
B. Resting heart rate and blood pressure		
Heart rate/min	73 ± 2	73 ± 2
Blood pressure, mm Hg		
Systolic	116 ± 2	121 ± 2*
Diastolic	77 ± 1	78 ± 1

Values are means ± SE. Predicted values are based on the formulas developed by Baldwin et al. (4) * Significant difference from the control ($P < 0.05$).

TABLE 3. *Heart rate (HR) and pressor response to breath holding (BH) in air and to apneic face immersion (FI) in water of 5 and 30 C*

Measurements	BH in Air		FI 30 C Water		FI 5 C Water	
	Control	Diver	Control	Diver	Control	Diver
BH time, sec	94±6	103±5	102±7	132±6*	97±10	105±7
Initial HR/min	88±5	89±2	83±2	83±2	87±3	88±2
ΔHR_{max}, %	19±2	28±2*	14±2	26±2*	30±2	40±2*
Systol BP, mm Hg						
Before BH	124±3	129±2	123±2	128±2*	126±3	129±2
End of BH	139±3	145±2*	139±4	149±2*	152±4	154±2
Diastol BP, mm Hg						
Before BH	79±1	83±1*	81±1	87±2*	85±2	88±2
End of BH	102±3	105±2	99±4	107±2*	105±4	110±3

Values are means ± SE. Each mean value is based on 22 measurements in the control and on 52 measurements in divers. * Significant difference from the corresponding control value ($P < 0.05$).

whom certain oscillatory changes were often noted. The time at which the maximal bradycardia is observed varied considerably with the individuals. The degree of bradycardia is less pronounced during simple breath holding in air or during apneic face immersion in water of 30 C as compared with that seen during apneic face immersion in water of 5 C, in both groups. However, the diver manifested a significantly greater bradycardia than the control in a given apneic maneuver.

Both the systolic and the diastolic blood pressures increased continuously during apnea. Although the magnitude of increase in diastolic pressure tended to be greater than that of increase in systolic pressure, the difference was not only very small but the trend was also inconsistent. As expected, the pressor response was greatest during face immersion in water of 5 C, but there was very little difference in the response between the two groups. The only consistent difference in the pressor response between the two groups was noted during face immersion in water of 30 C, which may be attributable to a large difference in the breath-holding time.

Alveolar gas tensions at the end of apnea. The individual values of $P_{A_{O_2}}$ and $P_{A_{CO_2}}$ observed at the breaking point of apnea are plotted as a function of the breath-holding time (Fig. 1). As expected, $P_{A_{O_2}}$ decreased progressively as the breath-holding time was prolonged. The lowest $P_{A_{O_2}}$ observed was 29 mm Hg. In contrast, $P_{A_{CO_2}}$ increased slightly during the early phase of breath holding and reached a plateau after 1 min at the average level of 47 mm Hg. These changes in alveolar gas tension were independent of the apneic maneuvers.

DISCUSSION

Certain physical adaptations to long-term diving have been noted in the US Navy Escape Training Tank Instructors (8, 26) and, to a lesser extent, in the women divers of Korea (ama) (31). These include an increase in VC, a decrease in RV, and thus a lowered RV/TLC ratio. Although the divers employed in the present work also showed such a tendency, there was no significant difference in either VC or RV between the divers and the control (see Table 2). Moreover, the RV/TLC ratio as a function of age was comparable in both groups (see Fig. 2). The failure of these divers to show a significant adaptation may be partly due to their lesser diving time as compared to the Navy or ama divers (17). As stated under METHODS, the divers used in this study are not engaged in daily diving but dive, on the average, 1 day a week.

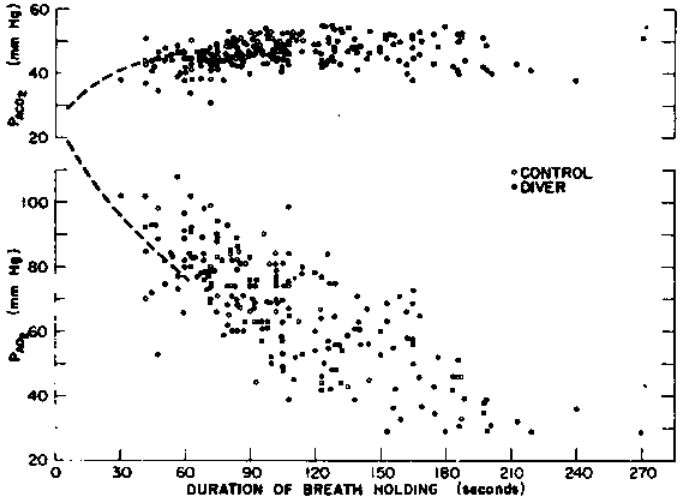

FIG. 1. Alveolar P_{O_2} and P_{CO_2} as a function of breath-holding time. Two broken curves indicate the changes in average P_{O_2} (*bottom*) and P_{CO_2} (*top*) during 1-min breath holding at rest, as reported by Lanphier and Rahn (22).

Although it is beyond the scope of this paper to discuss the mechanism(s) of the development of physical adaptation to long-term diving, it is of considerable interest to note that similar changes in VC and RV are seen during athletic training (3). This suggests that changes in the lung volumes as seen in professional divers may be somehow related to physical training rather than to certain factors unique to aquatic existence as postulated earlier (8, 31). In the preent work the values of MOI and MVV of the diver were similar to those of the control (see Table 2), suggesting that physical fitness is comparable in both groups. Evidently the divers used in this study are not subjected to any more physical training than the control and this may also account for the failure to demonstrate the adaptive changes in the lung volume of the diver.

The most significant finding in the present work is the greater bradycardia observed in the diver during apnea (see Table 3). In agreement with other studies (10, 21, 24, 32, 33), the magnitude of apneic bradycardia was significantly greater during apneic face immersion in cold water as compared to simple breath holding in air or to apneic face immersion in warm water. However, in a given apneic maneuver, the diver always showed a greater bradycardia than the control. In the only study available on well-trained divers, Corriol and Rohner (9) also observed that the degree of apneic bradycardia is greater in the diver during simple breath holding or during face immersion in water. On the basis of this finding, they concluded that diving training increases bradycardial response to breath holding, but offered no explanation how this is brought about. These authors (9) also found that the breath-holding time is longer in the diver, but found no correlation between the magnitude of bradycardia and the duration of apnea.

To assess the interrelationship between variables and apneic bradycardia, intercorrelation (r) for major variables

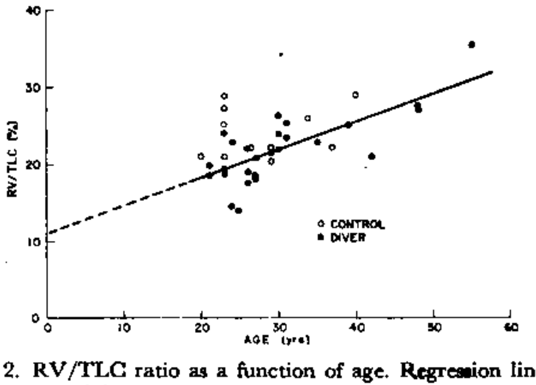

FIG. 2. RV/TLC ratio as a function of age. Regression line ($y = 0.378\,x + 10.50$) for the diver is indicated.

was computed. In the control, the degree of maximal bradycardia (ΔHR_{max}) in each individual was best correlated with the age ($r = -0.778, P < 0.01$). The correlation between Δ HR_{max} and age in the diver was lower ($r = -0.366, 0.10 > P > 0.05$), and there was a significant regression ($P < 0.05$). The difference in the slopes of regression lines between the two groups is highly significant ($t = 4.33$, df = 33, $P < 0.005$), indicating that the reduction in the degree of apneic bradycardia with advancing age is less pronounced in the diver as compared to the control (Fig. 3). Figure 3 also shows that Δ HR_{max} for a given age is greater, especially in older subjects, in the diver than in the control. Unlike the control, Δ HR_{max} of the diver was best correlated with the RV/TLC ratio ($r = -0.416, P < 0.05$) (Fig. 4). The correlation between Δ HR_{max} and the RV/TLC ratio in control was lower ($r = -0.318, P > 0.05$), although there was a significant regression ($P < 0.05$). However, there is no significant difference in the slopes of the two regression lines ($t = 0.58$, df = 33, $P > 0.10$), indicating that the bradycardial response for a given RV/TLC ratio is not significantly different between the two groups. Leith and Mead (23) proposed that the RV is determined by the static balance of muscle and recoil forces (mainly in the chest wall) in young individuals and by a dynamic one operating mainly in the lung in older men. It would be of interest to study if these forces are related to the development of apneic bradycardia.

Physical fitness seems to alter the bradycardial response to apnea (6, 10, 14, 19, 24). Although there was a trend for the bradycardial response to be positively correlated with the MOI, it was not significant ($P > 0.10$). Neither the breath-holding time nor percent body fat showed any significant correlation with the bradycardial response in either group.

A rise in blood pressure due to peripheral vasoconstriction during apnea could contribute to the bradycardia. However, apneic bradycardia does not parallel the rise in blood pressure (15) or the reduction in peripheral blood flow (7, 32). The present work also failed to demonstrate a correlation between Δ HR_{max} and the pressor response to apnea. This would suggest that a baroreflex component would not account for the observed difference in bradycardial response between the two groups. However, it is possible that either the sensitivity of baroreceptors or the vasomotor response to a given stimulus may be different in the diver who is engaged in repetitive breath-hold dives. Certain cardiovascular functions such as arterial blood pressure can be made to rise or fall predictably after conditioning by environmental stimuli (5, 16). A systematic study along this line is needed.

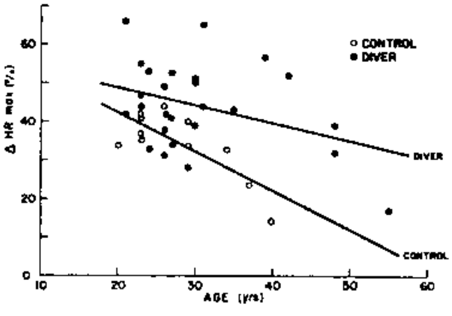

FIG. 3. Maximal percent reduction of heart rate (ΔHR_{max}) in individual subjects during apnea as a function of age. Regression equation is $y = -1.03x + 63.16$ for the control and $y = -0.46x + 58.00$ for the diver. Standard errors of the slope and of the intercept are 0.27 and 5.89, respectively, for the control and are 0.24 and 6.0, respectively, for the diver.

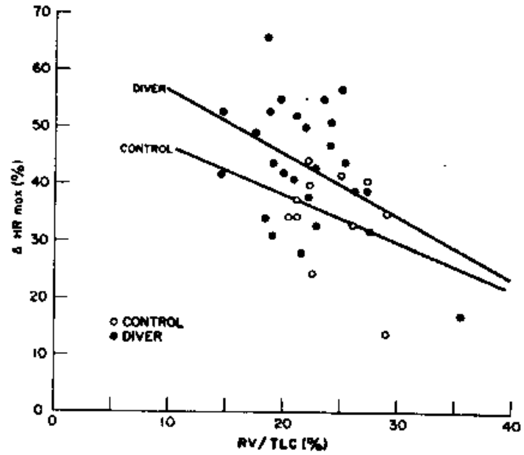

FIG. 4. Maximal percent reduction of heart rate $(\Delta HR)_{max}$ in individual subjects during apnea as a function of the RV/TLC ratio. Regression equation is $y = -0.85x + 54.84$ for the control and $y = -1.06x + 67.34$ for the diver. Standard errors of the slope and of the intercept are 0.84 and 7.67, respectively, for the control and are 0.47 and 4.15, respectively, for the diver.

The significant potentiation of apneic bradycardia by immersion of face in cold water has been widely acknowledged and is attributed to a reflex initiated by the cold-sensitive receptors in the face (10, 21, 24, 32, 33). Recently, Kawakami et al. (21) pointed out that the alveolar gas exchange during face immersion may not be quite the same as that during simple breath holding because the pulmonary capillary blood flow has been found to be somewhat decreased in their study. Actual analysis of alveolar gas samples obtained at the breaking point of breath holding indicated no difference in the level of P_{O_2} and P_{CO_2} between simple breath holding and apneic face immersion. Thus, there is no reason to speculate that the greater bradycardia observed during apneic face immersion in cold water is due to the greater hypercapnea which has been shown to intensify and maintain the apneic bradycardia (21).

The alveolar gas pressure as a function of breath-holding time (see Fig. 1) deserves additional comment. As expected, P_{O_2} decreased continuously as the breath-holding time was prolonged, indicating that long-breath holders are able to tolerate low P_{O_2}. Hence, the fact that the grand average breath-holding time (especially apneic face immersion time in warm water) was longer in the diver as compared to the control suggests a greater tolerance to hypoxia of the diver, in agreement with the work of Schaeffer (26). No such adaptation to hypoxia was observed in ama (31). Lanphier and Rahn (22) reported earlier that the alveolar P_{CO_2} tends to reach a plateau while oxygen uptake from the lungs diminished markedly during 1-min breath holding. The present work now indicates that the above CO_2 plateau is remarkably maintained up to 4.5 min of breath holding. It will be of great interest to study where and how CO_2 is stored during the prolonged breath holding.

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GASTROESOPHAGEAL DYNAMICS DURING IMMERSION IN WATER TO THE NECK

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ABSTRACT

This investigation was undertaken to study the effect of hydrostatic pressure on gastroesophageal dynamics during immersion in thermoneutral water to the neck. In five healthy male subjects normal end-expiratory, gastric pressure (P_G), esophageal pressure (P_E), location and pressure of distal esophageal sphincter (DES), location of respiratory inversion point (RIP) and gastroesophageal pH gradient were measured standing in air (A), standing in water to the neck (B) and standing in air with abdominal compression (C). The pressure was measured with a Honeywell esophageal catheter (model 31) with built-in pressure transducer. A Beckman stomach pH electrode (No. 39042) was positioned adjacent to the pressure transducer. P_G increased from 4.6 ± 0.6 (SE) mmHg in A to nearly 20 mmHg in B and C, while P_E increased from -6.0 ± 0.8 mmHg in A to -0.8 ± 1.0 and -3.4 ± 0.9 mmHg in B and C, respectively. However, P_{DES} was always 11-15 mmHg higher than P_G. The superior limit of DES was displaced cephalad by 3.0 cm while the inferior limit was displaced by 1.6 cm in B, indicating a stretching of DES and a shortening of the esophagus. Qualitatively similar findings were obtained in C. In all experiments, the esophageal pH remained above 6, and no alteration in the amplitude of primary peristaltic waves was seen. It is concluded that a head-out immersion with increased gastroesophageal pressure gradient predisposes to gastric reflux in the absence of a competent DES mechanism.

Key words: Abdominal compression, distal esophageal sphincter, esophageal pressure profile, gastroesophageal pressure gradient, immersion.

INTRODUCTION

When the human body is immersed in water up to the neck, a graded hydrostatic pressure is exerted over the whole body, resulting in a greater compression of the abdomen as compared to the chest. As a result, the transdiaphragmatic pressure (i.e. the abdominal pressure minus the intrathoracic pressure) increases while the diaphragm moves cephalad (1), which are responsible for other changes in cardio-respiratory functions observed during immersion (Arborelius et al., 1972; Craig and Ware, 1967; Hong et al., 1969; Hong et al., 1960).

Altered gastroesophageal dynamics may be one consequence of such an increase in the transdiaphragmatic pressure, coupled with a cephalad movement of the diaphragm during immersion. In fact, pyrosis and regurgitation of gastric contents have been reported by some swimmers, snorkeling and SCUBA enthusiasts while immersed in these aquatic pursuits. Pulmonary aspiration of gastric contents is a pathophysiologic event in some drownings (Cave, 1971; Martin and Barrett, 1971).

The purposes of this investigation are 1) to accurately establish the gastroesophageal pressure gradient along with the distal esophageal sphincter pressure, and 2) to monitor the gastroesophageal pH gradient as an indication of possible gastric reflux, during immersion in water up to the neck, and standing in air, with and without abdominal compression.

METHODS

Five male volunteers without symptoms of gastroesophageal reflux were used as subjects. Their age and physical characteristics are shown in Table 1.

Subjects	Age (Year)	B.W. (kg)	Height (cm)	ERV (ml, BTPS)	
				Air	Immersion
1	37	70	170	1,100	240
2	33	73	163	1,960	330
3	30	64	165	2,150	600
4	45	65	172	2,040	530
5	35	80	180	2,360	450
Mean	36	70.4	170.0	1,922	430
S.E.	2.5	2.91	2.98	216	65

Table 1. Physical characteristics and expiratory reserve volume (ERV) of subjects.

Experimental protocol

On the day of experiments, each subject reported to the laboratory at around 2 p.m. without lunch. Following an approximately 15 minute rest, the catheter assembly (an esophageal pressure probe and a pH electrode; see below for the specifications) was passed per oral while standing in air without topical anesthesia. While the subject was breathing quietly, the catheter assembly was slowly advanced at 1 cm increments until all pressure gauges and pH probe had transversed the distal esophagus and distal esophageal sphincter (DES) and were in the stomach (antigrade passage). Withdrawal of the catheter assembly was then accomplished in a similar manner until the proximal pressure gauge had transversed the cricopharyngeus and was in the hypopharynx (retrograde passage). During passage of the catheter assembly, both the pressures and pH were continuously recorded on a Beckman 8-Channel Type R Dynograph. In order to minimize the artifact due to peristalsis, the subject was constantly encouraged not to swallow during passage of the catheter assembly.

Identical procedures were repeated while the subject was standing in air with abdominal compression and also was standing in a water tank with the head out. The abdominal compression was achieved by inflating a securely fastened thigh blood pressure cuff about the abdomen. The cuff pressure was raised until the gastric pressure reached a level of 20 mmHg. The temperature of water in the tank was maintained at about 33°C. The subject rested standing in water for approximately 5 minutes before the measurements were taken. The effect of increased gastric pressure both with and without deglutition was monitored during a slow withdrawal of the catheter from the stomach to the cricopharyngeal high pressure zone.

Measurements of pressure and pH

The Honeywell (Model 31) esophageal mobility probe (air-filled) with three transistorized pressure gauges 5 cm apart was used for the pressure measurements. The use of this new probe allows the measurement of pressure in mmHg at any given region with much more certainty than with the conventional balloon method which measures the pressure over an area covered by a balloon. This enabled us to establish a more precise topography of esophageal pressure than with the use of a balloon.

A Beckman stomach pH electrode (No. 39042) was used for pH measurement. The pH electrode was positioned adjacent to the distal pressure gauge and pH recording was transcribed simultaneous with distal pressure determinations. The reference electrode for pH determination was placed on the mucosal surface of the cheek.

Determinations of DES pressure and location

These were done during both antigrade and retrograde passages of the catheter assembly. Typical pressure tracings obtained with retrograde passages of the catheter are shown in Figure 1. The changes in gastric pressure during quiet breathing are shown on the far left of each tracing, indicating an increase in the pressure during inspiration. As the catheter is being withdrawn slowly, the end-expiratory pressure and/or the amplitude of each wave begins to increase at point "a" which signifies the inferior limit of DES. As the catheter transverses the sphincter, the pressure reaches a plateau (DES pressure) and at point "b" the pressure decreases during inspiration (Respiratory Inversion Point -- RIP). Beyond this point the pressure continues to decrease until it begins to level off at point "c" which signifies the superior limit of DES. Though not shown in this figure, the pH increases from about 2 at point "a" to about 6 at "c".

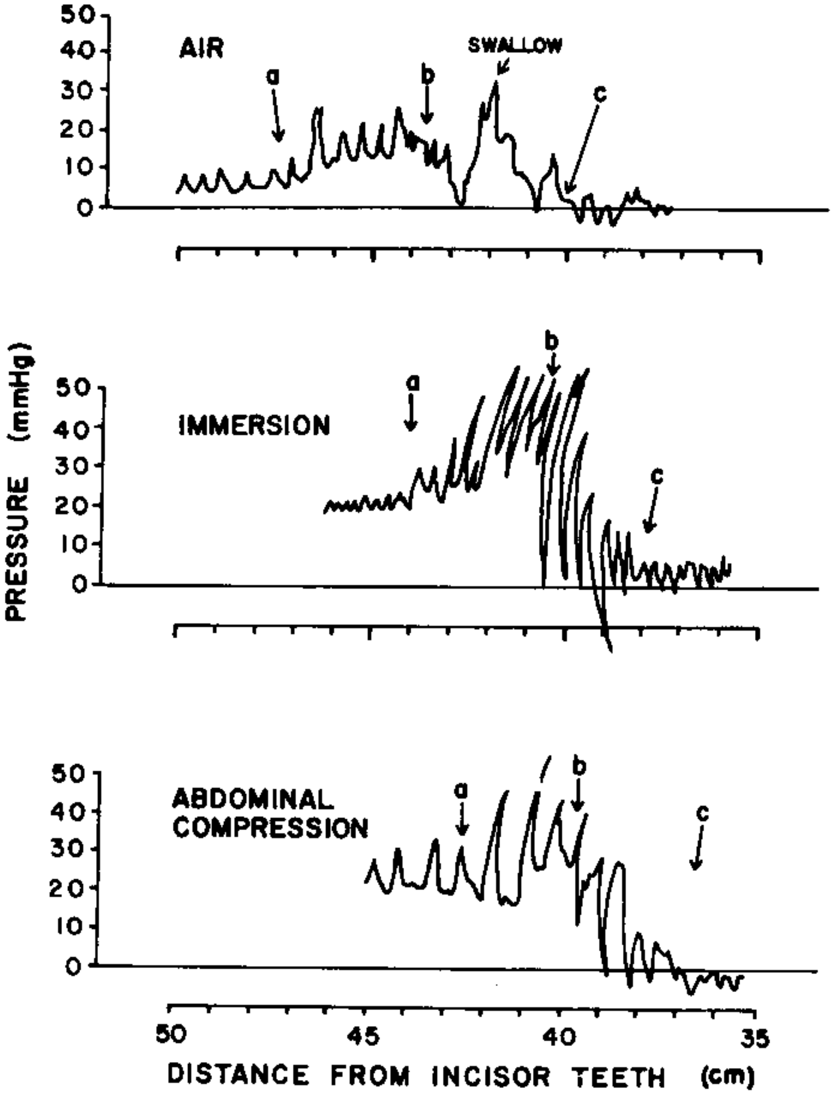

Figure 1. Typical pressure tracings obtained with retrograde passage of the catheter assembly in a subject standing in air (top), immersed in water to the neck (middle) and standing in air with abdominal compression (bottom). a: inferior limit of distal esophageal sphincter; b: respiratory inversion point; c: superior limit of distal esophageal sphincter. See the text for more detailed descriptions of these pressure tracings.

RESULTS

Topography of esophageal pressure

The mean gastric (P_G), DES (P_{DES}) and esophageal pressures (P_E) determined at the end of normal expiration in 5 subjects standing in air, with and without abdominal compression, and in water up to the neck are shown in Figure 2. The relative length of the esophagus between the bottom of the circopharyngeus (0%) and the superior limit of the DES (100%) is indicated on the abscissa.

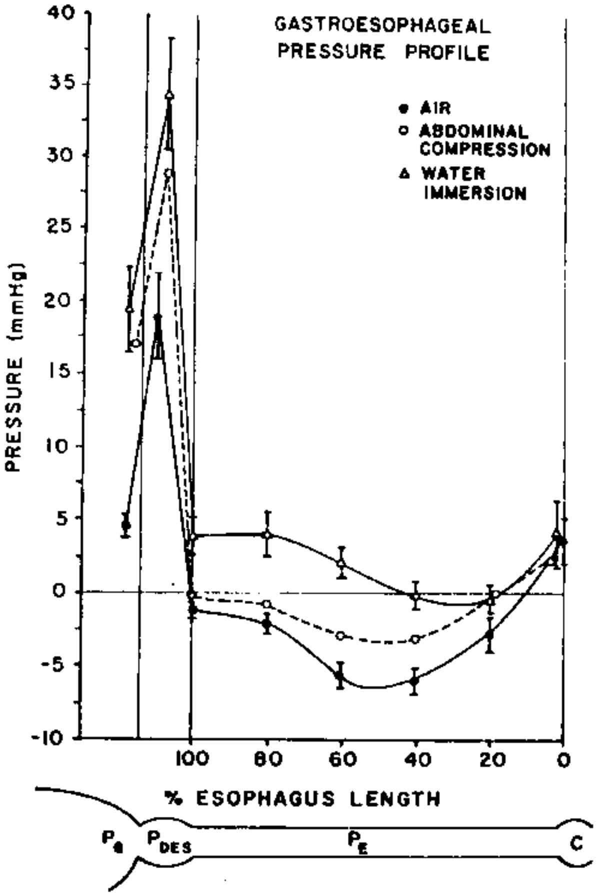

Figure 2. Gastroesophageal pressure profiles during standing in air (●), immersion in water to the neck (△), and standing in air with abdominal compression (o). P_G: gastric pressure; P_{DES}: distal esophageal sphincter pressure; P_E: esophageal pressure; C: cricopharyngeus. Esophageal length expressed as % for all subjects refers to a vertical distance between cricopharyngeus and the superior limit of distal esophageal sphincter. Each point represents the mean (± SE) of data from 5 subjects. Actual esophageal length for all subjects in 3 experimental conditions are shown in Table 4.

While standing in air the P_G averaged to 4.6 ± 0.6 (SE) mmHg while P_{DES} was 19.4 ± 3.2 mmHg. The P_E at the superior limit of DES was only slightly below atmospheric pressure but became progressively more negative toward the mid-portion of the esophagus where it reached approximately -6 mmHg. From this point on the P_E again increased progressively to above atmospheric pressure. The gastro-esophageal pressure gradient (i.e., ΔP between the stomach and the superior limit of DES) was 5.8 ± 0.6 mmHg (Table 2).

During immersion in water to the neck the P_G increased to nearly 20 mmHg and the P_{DES} to about 32 mmHg. These values represent an increase of about 15 mmHg over the respective values in air ($P < 0.01$ for P_G; $0.05 < P < 0.10$ for P_{DES}). As compared to such large increases in P_G and P_{DES} in water, the P_E values were higher in water by only about 5 mmHg than in air ($P < 0.01$). Thus, the gastroesophageal pressure gradient is nearly tripled during immersion as compared to that in air ($P < 0.01$) (Table 2). It is also of interest to note that the lowest P_E point shifted proximally in water (Figure 2).

During standing in air with abdominal compression, both the P_G and P_{DES} were similar to those during immersion in water but the P_E was found between the water and air (without abdominal compression) values (Figure 2).

DES location

The distance from the incisor teeth to the superior and inferior limits of DES, and to RIP under three different experimental conditions are tabulated in Table 3. Included in the same table are the values of pH gradient which is defined as a location (cm from incisor teeth) where the nearly neutral esophageal pH (≥ 6) changes to the acidic gastric pH (≤ 2). When available, values obtained with the antigrade and retrograde passages are shown separately.

It should be noted that both the DES (both superior and inferior limits) and the RIP are located further away from the incisor teeth with the antigrade than with the retrograde passage as Thurer et al. (1974) have previously shown. We feel the changes in the location of DES and RIP during immersion in water were more precise with the retrograde as compared to the antigrade passage of the catheter. Using the data obtained with the retrograde passage, one can see that the superior limit of DES is displaced cephalad by 3.0 cm ($P < 0.05$) while the inferior limit is displaced by only 1.6 cm ($P < 0.05$), during immersion in water as compared to standing in air without abdominal compression. Though not statistically significant, there was a cephalad displacement of RIP quantitatively similar to that of the inferior limit of DES. Essentially identical changes in the location of the inferior limit of the DES and RIP were seen with mechanical abdominal compressions standing in air as during immersion; but the cephalad displacement of the superior limit of DES tended to be smaller than during immersion.

As stated earlier the pH abruptly changes from a nearly neutral value in the esophagus to a strongly acidic value as the catheter transverses the DES during the antigrade passage. The location where the pH was last 6 (defined as "pH gradient" in Table 3) during antigrade passage also shifted cephalad by 4 cm ($P < 0.01$) during immersion.

Subjects	Standing in Air				Immersion				Abd. Compression			
	P_G	P_E	P_G-P_E	P_{DES}	P_G	P_E	P_G-P_E	P_{DES}	P_G	P_E	P_G-P_E	P_{DES}
1	6	-1	7	32	21	1	20	33	15	1	14	20
2	4	0	4	16	19	5	14	45	20	0	20	40
3	4	-2	6	18	12	1	11	17	16	-1	17	22
4	3	-2	5	16	15	3	12	25	16	-2	18	32
5	6	-1	7	15	30	9	21	42	17	0	17	-24
Mean	4.6	-1.2	5.8	19.4	19.4^b	3.8^b	15.6^b	32.4^a	16.8^b	-0.4^a	17.2^b	27.6
SE	0.6	0.4	0.6	3.2	3.1	1.5	2.1	5.2	0.9	0.5	0.9	3.7

Table 2. Intragastric (P_G), intraesophageal (at the superior limit of DES; (P_E) and distal esophageal sphincter (P_{DES}) pressure (mmHg)* during standing in air, immersion to the neck, and standing in air with abdominal compression.

* All pressure values were taken at the end of expiration.

a and b denotes the level of significance at $0.1 < p < 0.05$ and $p < 0.01$, respectively, for paired comparison between standing in air and water immersion, or between standing in air and abdominal compression.

Subject		DES Location (cm)*									pH gradient (cm) **	
		Superior Limit			Inferior Limit			Rip				
		A	B	C	A	B	C	A	B	C	A	B
1	AG	40	38	--	44.5	44	--	42	42.5	--	46	40
	RG	40	36	37	44	42	42.5	42	42	41	--	--
2	AG	42	40	--	50	47	--	44	43.5	--	45	42
	RG	41	38	37.5	46	44	42	44	42	40	--	--
3	AG	42	41	--	49	50	--	44	45	--	46	44
	RG	40	37	38.5	45	44	45	42	42	41.5	--	--
4	AG	41	41	--	45	45	--	43.5	44	--	49	43.5
	RG	41	38	40	44	43	44	42	39	42	--	--
5	AG	45	43	--	50	47	--	47	45	--	47	44.5
	RG	41	39	39.5	47	45	45	45	44	41.5	--	--
Average	AG $\bar{X}$	42.0	40.6[a]	--	47.7	46.6	--	44.1	44.0	--	46.8	42.8[c]
	SE	0.8	0.8	--	1.2	1.0	--	0.8	0.5	--	0.9	0.8
	RG $\bar{X}$	40.6	37.6[a]	38.5[b]	45.2[b]	43.6[a]	43.7	43.0	41.8	41.2	--	--
	SE	0.2	0.5	0.6	0.6	0.5	0.6	0.6	0.8	0.3	--	--

Table 3. Distal esophageal sphincter (DES) location, respiratory inversion point (RIP) and pH gradient during standing in air (A), immersion to the neck (B), and standing in air with abdominal compression (C).

*Distance from incisor teeth AG and RG refer to anti- and retrograde passages, respectively.
**Defined as a location (cm from incisor teeth) where the nearly neutral intraesophageal pH ($\gtrsim 6$) changed to the acidic intragastric pH ($\lesssim 2$).
a, b, and c denote the level of significance at $p < 0.05$, $p < 0.02$, and $p < 0.01$, respectively for paired comparisons between A and B or between A and C.

These results suggest a greater shortening of the esophagus during immersion than during standing in air with abdominal compression. As summarized in Table 4, the length of esophagus between cricopharyngeus and the superior limit of DES shortened by 11% ($P < 0.05$) during immersion and 9.7% ($P < 0.05$) during standing in air with abdominal compression. However, there was no significant difference between the above two values. Similar findings, though quantitatively less, were obtained for the length between circopharyngeus and RIP.

	DES - Cricopharyngeus			RIP - Cricopharyngeus		
Subject	length (cm)	Percent shortening		length (cm)	Percent shortening	
	air	Immersion	Abd. Comp.	air	Immersion	Abd. Comp.
1	22.5	20.0	20.0	24.0	4.2	8.3
2	22.0	11.4	13.6	25.0	8.7	12.0
3	23.0	8.7	6.5	24.0	4.2	2.1
4	23.0	8.6	4.3	25.0	10.0	6.0
5	24.0	6.3	4.2	28.0	10.7	14.3
Mean	22.9	11.0*	9.7*	25.2	7.6*	8.5*
S.E.	0.3	2.4	3.1	0.7	1.4	2.2

Table 4. Shortening of length (as percent of length during standing in air) between the superior limit of distal esophageal sphincter (DES) and cricopharyngeus and between respiratory inversion point (RIP) and cricopharyngeus during immersion to the neck and standing in air with abdominal compression.

*$p < 0.05$ for paired comparisons between standing in air and water immersion, or between standing in air and abdominal compression.

Despite the large increase in gastroesophageal pressure gradient during immersion and standing in air with abdominal compression, the esophageal pH remained above 6 in all subjects. Similarly, the amplitude of primary esophageal peristaltic waves was not altered during immersion or during standing in air with abdominal compression.

DISCUSSION

One of the most remarkable changes consistently observed during head-out immersion is a drastic reduction in the expiratory reserve volume (ERV) (Agostoni et al., 1966; Hong et al., 1969; Hong et al., 1960). In agreement with previous reports, the ERV showed an 80% reduction during immersion in the present work (Table 1). The finding that the inferior limit of DES as determined with the retrograde passage of the catheter shifted cephalad by 1.6 cm during immersion (Table 3) probably indicates a corresponding movement of the diaphragm since simultaneous manometric and cine-radiographic observations in normal subjects show presence of DES on either side of the diaphragm (Donner and Margulies, 1972). Harris and Pope (1966) have shown the RIP does not correlate with the anatomical diaphragm, but rather the location of DES. Interestingly enough, the superior limit of DES showed a greater cephalad displacement than the inferior limit during immersion, indicating a stretching of the DES muscle from 4.6 cm in air to 6.0 cm during immersion. Most probably, this is attributable to contraction of the longitudinal muscle of the esophagus as indicated by a 11% shortening of the length between the cricopharyngeus and the superior limit of DES (Table 4). Such a contraction of the esophagus cannot be readily explained at present. It is, however, interesting to note that the extent of cephalad displacement of the superior limit of DES tended to be less during standing in air with abdominal compression although both the inferior limit of DES and RIP shifted cephalad to the same extent, as during immersion (Table 3). In other words, the compression of both the chest and abdomen during immersion to the neck is responsible for the above mentioned stretching of DES and the greater shortening of the esophagus.

While standing in air without abdominal compression mean gastric pressure was 4.6 mmHg at the end of normal expiration (Table 2), which agrees well with the value reported by Agostoni and Rahn (1960), and Hong et al. (1969). The esophageal pressure was lowest at the mid-portion with an average of -6.0 ± 0.8 mmHg at the end of normal expiration, which again agrees rather well with generally accepted values and most probably reflects the intrathoracic pressure (Banchero et al., 1967; Milic-Emili et al., 1964; Milic-Emili et al., 1964). In the upper third of the esophagus, the pressure became less negative or even positive (Figure 2). Milic-Emili et al. (1964) using a short balloon also noted that the esophageal pressure in the upper third is extremely variable and is not related to changes in transpulmonary pressure. They attributed this to traction on, or compression of, the esophagus by neighboring structures, principally the trachea. In the lower third of the esophagus, the esophageal pressure became progressively less negative toward the DES with the gradient of approximately 0.4 mmHg/cm (Figure 2), in contrast to 1 cm H_2O/cm reported by Banchero et al. (1967). This increase in pressure in the lower third of the esophagus is due to downward and outward pull of the heart and lungs on pleura, rather than being intrinsic to the esophageal lumen (Banchero et al., 1967; Knowles et al., 1959; Milic-Emili et al., 1964). Nevertheless, the pressure in the lowest portion of the esophagus is still slightly negative and is 5.8 ± 0.6 mmHg below the gastric pressure (Table 2). This gastroesophageal pressure gradient would induce reflux of the gastric content into the esophagus were it not for the higher DES pressure (Cohen and Harris, 1971).

During immersion in water to the neck, the gastric pressure increased to 19.4 mmHg and the lowest esophageal pressure was -0.8 ± 1 mmHg; both values in close agreement with those obtained by Agostoni et al. (1966). However, the lowest pressure region of the immersion esophageal vertical pressure profile shifted orad (Figure 2). This shift of the lowest pressure region must be related to forces external to the esophagus which also shifted orad. In the lower portion of the esophagus the pressure increased toward DES with a gradient of approximately 0.3 mmHg/cm, a value slightly lower than 0.4 mmHg/cm observed during standing in air ($P > 0.05$). It is quite possible that the position of the heart may shift slightly due to cephalad displacement of the diaphragm by immersion abdominal compression, thus counteracting the gravity dependent downward pull of the heart and lungs. The lowest esophageal pressure region did not shift with mechanical abdominal compression, thus suggesting factors other than immersion abdominal compression may cause a shift of the heart such as thoracic anterior-posterior compression due to hydrostatic pressure. It is also possible that immersion induced reduction in ERV may have played a role in shifting minimal P_E cephalad. Because of greater increase in gastric pressure as compared to esophageal pressure during immersion, the gastroesophageal pressure gradient increased from 5.8 mmHg in air to 15.6 ± 2.1 mmHg during immersion (Table 2). It has been widely reported that increased transdiaphragmatic pressure induces blood shift into the intrathoracic space, resulting in reduction of vital capacity and an increment in the filling pressure of the heart. Functional consequences to these changes have been discussed in detail elsewhere (Arborelius et al., 1972; Hong et al., 1969).

No evidence of gastric reflux was found in the present work during immersion because the tone of DES was maintained sufficiently high in normal subjects (Figure 2). It is quite conceivable that gastric reflux could be seen in those whose DES tone is low. DES pressure in response to ΔP_G both with immersion and mechanical abdominal compression always exceeded the respective intragastric pressure, thus protecting the esophageal environment from acid regurgitation despite an accentuated gastroesophageal pressure gradient encouraging reflux. Lind et al. (1968) have shown this DES response to ΔP_G is related to a cholinergic mechanism, mediated by a reflex via the vagus nerves. Absence of this DES response to ΔP_G has been shown by Butterfield et al. (1972) in patients with gastroesophageal reflux symptoms and results in 30% transmission of ΔP_G to resting intraesophageal pressure. We feel poor DES response to immersion ΔP_G results in the gastroesophageal reflux symptoms some individuals experience during aquatic activity for we have shown similar DES response to ΔP_G in normal subjects regardless of whether due to immersion or mechanical abdominal compression. An abdominal compression syndrome with gastroesophageal reflux symptoms was described by White (1973) and attributed to tight abdominal garments. In support of this, we have shown an even greater gastroesophageal pressure gradient with mechanical abdominal compression than with immersion (Table 2) which would create a favorable pressure environment for reflux. Orad displacement of both pH change, DES, and shortening of the esophagus with immersion showed the stress the elongated DES must withstand to maintain its competency and keep gastric contents out of the esophagus.

In conclusion, we feel immersion with increase in intragastric pressure and expanded gastroesophageal pressure gradient predisposes to reflux. A competent distal esophageal sphincter mechanism prevents acid content from flowing into the esophageal environment. Drugs which diminish distal esophageal sphincter pressure and thus its competency (Pope, 1972) should be avoided during immersion. We feel individuals with severe gastroesophageal reflux may be at risk from aspiration of gastric contents with occupations or recreation involved with water immersion. Our clinical observations and those of others (Cave, 1971) would support this concern.

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Physiological and conventional breath-hold breaking points

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LIN, Y. C., D. A. LALLY, T. O. MOORE, AND S. K. HONG. *Physiological and conventional breath-hold breaking points.* J. Appl. Physiol. 37(3): 291–296. 1974.—The course of breath-hold (BH) can be characterized by *a*) voluntary inhibition of respiratory muscular activity, *b*) onset and maintained involuntary ventilatory activity (IVA) with glottis closed, and *c*) voluntary opening of glottis. While type *c* has been widely studied, the present study focused on type *b*. The intraesophageal pressure (IEP), BH time and oxygen-demand relationship, and factors responsible for the onset of IVA were studied during BH with air and BH with oxygen at rest and during steady-state exercise at 167 kgm·min^{-1}. IVA generates periodic subatmospheric pressure in the thoracic cavity, while at the onset of BH the IEP is above ambient. The effect of this periodic subatmospheric intrathoracic pressure on the cardiovascular functions is discussed. The relationship between $\dot{V}_{O_2}$ (ml·s^{-1}) and BH time (T, s) can be described by $T = a\,(1/\dot{V}_{O_2}) + b$, where b is small and insignificantly different from zero; a represents the amount of O_2 supplied by the lung and the blood during the period of BH. $P_{A_{CO_2}}$ at the onset of IVA was independent of $P_{A_{O_2}}$ over a wide range of $P_{A_{O_2}}$ (150–660 mmHg). It is concluded that the onset of IVA is P_{CO_2} and $\dot{V}_{O_2}$ dependent, nonsubjective, and serves as a physiological breaking point for the BH.

breath holding; involuntary ventilatory activity, $\dot{V}_{O_2}$ and breath-hold time

THE ABILITY TO BREATH HOLD is influenced to a great extent by psychological factors (23), such as the ability to withstand the sensation of suffocation and discomfort. Thus, the conventional breath-hold (BH) breaking point varies greatly in duration as does alveolar gas composition within and between individuals. Breaking point time from less than 20 s (26) to 270 s (11) has been reported for BH with air after expiring maximally and then inspiring fully without previous hyperventilation. The voluntary inhibition of the ventilatory muscles, easily achieved at the beginning of BH, becomes untenable as BH progresses while voluntary closure of the glottis is still possible (1). The factors that contribute to overcoming voluntary closure of the glottis (the conventional breaking point) have been studied in terms of alveolar gas composition (3, 11, 12, 17), and its dynamic features (10, 15, 18), lung volume (11, 19, 20), mechanical intervention such as rebreathing during a BH (7, 9), and psychological factors (23, 26). Godfrey and Campbell (8) summarized these factors and provided a working hypothesis for the control of breath holding. They concluded that rigid concepts of chemical and mechanical thresholds are inadequate in interpreting BH breaking point. Instead, they proposed an accumulation of a central excitatory state arising from loss of respiratory movement. Less attention has been paid to the ventilatory effort occurring prior to the conventional breaking point. Diaphragm activity (1) and return of the "urge to breathe" (6) have been noted prior to reaching the conventional breaking point during the course of a BH.

The onset of involuntary ventilatory activity (IVA) prior to the conventional breaking point has been suggested (1) and supported (16) as a physiological breaking point. This breaking point would presumably be more reliable than the conventional breaking point, since the latter is subjected to psychological influences. The factors which contribute to the onset of IVA and the conventional breaking point are the subject of this study.

METHOD

Five male volunteers, 25–42 yr old, served as subjects. With the exception of one, all subjects could breath hold for 2–4 min with a full lung of air without prior hyperventilation. All subjects were experienced as subjects in a previous study (10). Their pertinent anthropometric and physiological measurements are listed in Table 1. All subjects were in apparent good health as far as the cardiopulmonary systems were concerned. A 15 x 1 cm latex esophageal balloon (Anode Rubber Plating Co.) was introduced orally and intraesophageal pressure was measured in accordance with the method of Knowles et al. (14). The pressure was recorded continuously using a Statham P23BC transducer and a Beckman Dynograph. The method of alveolar gas sampling was the same as described by Hong et al. (10). The alveolar gas samples were analyzed for oxygen and carbon dioxide with a Scholander micro-gas analyzer (24) for BH with air, and with a Radiometer PHM 71 gas analyzer (Copenhagen, Denmark), for BH with oxygen. The calibration gases for the Radiometer PHM 71 were checked with the Scholander micro-gas analyzer. The residual volume was determined by the three-breath method of Rahn et al. (22). The inspiratory volume and the lung volume at the end of a BH were measured with a 13-liter Collins spirometer with the CO_2 absorber removed. All experiments were carried out during midday, 10:00 AM to 2:00 PM.

BH at rest. The subject in sitting position began the BH after a maximal expiration followed by a maximal inspiration. The inspired volume was recorded. An alveolar gas sample was taken at the end of maximal expiration, repre-

TABLE 1. *Physical characteristics and pulmonary functions of the subjects*

Measurements	Mean ± SE	*N*
Age, yr	32.2 ± 2.96	5
Ht, cm	168.8 ± 1.24	5
Body wt, kg	62.4 ± 2.9	5
Surface area, m^2	1.79 ± 0.20	5
RV, ml (BTPS)	1,723 ± 210	5
IV, ml (BTPS)*	3,944 ± 92	15
$\dot{V}_{O_2}$ rest, $ml \cdot min^{-1}$ (STPD)	305 ± 18	15
$\dot{V}_{O_2}$ exericse, $ml \cdot min^{-1}$ (STPD)†	747 ± 48	10

* IV, inspiratory volume, was measured just prior to BH. The subjects were asked to expire to residual volume and then inspire fully. † The subjects exercised at 167 $kgm \cdot min^{-1}$ and the $\dot{V}_{O_2}$ was measured during steady-state exercise.

senting the gas composition of the residual lung gas. Thus, the gas composition at the beginning of a BH can be calculated. The second alveolar gas sample was obtained at the time when a sharp reduction in the esophageal pressure appeared on the recording, signaling expansion of the thoracic cavity with the glottis closed. The subjects continued to BH until the maximal BH time was reached (conventional breaking point). Another alveolar gas sample was obtained at this breaking point. The final lung volume at the end of maximal BH was also recorded. The entire process required 2 (for BH with air) to 4 min (for BH with O_2). A 10- to 15-min rest period was provided following each BH. The oxygen consumption ($\dot{V}_{O_2}$) was then measured with a 13-liter Collins spirometer. The subject, upon completion of $\dot{V}_{O_2}$ measurement, was switched swiftly without inhalation of air to a 200-liter oxygen bag which was equipped with a one-way exhaust valve. The subject continued to breathe oxygen until a total of 10 min elapsed since the beginning of $\dot{V}_{O_2}$ measurement. The subject then began BH, as previously described for air, with the inspiration of oxygen. The sequence of alveolar gas sampling was the same as that in the BH with air. The entire sequence was repeated two more times on the same subject on separate days.

BH during exercise. The subjects exercised on a bicycle ergometer at the rate of 167 $kgm \cdot min^{-1}$. The $\dot{V}_{O_2}$ was raised 2–3 times over resting levels at this rate of exercise (Table 1). All subjects could perform this rate of exercise for a prolonged period. The BH began at 10 min following the onset of exercise, and the entire process as described above for BH at rest on air and on oxygen was performed without stopping the exercise. The experiment was also repeated for the same subjects on different days.

Oxygen appearance time from lung to ear. To estimate the level of arterial P_{CO_2} at the brainstem from the measured values of $P_{A_{CO_2}}$, it is necessary to know the time delay from the lung to the brain. The oxygen appearance time from the lung to the ear was estimated by an ear oximeter (Waters Instruments, Inc.). The subject was instructed to hold his breath until a significant drop in the oxygen saturation was observed on the record and then to inspire quickly. The onset of increase in oxygen saturation following the inspiration was taken as the oxygen appearance time from the lung to the ear (see Fig. 1), as an estimate for the lung-to-brain time.

Statistical analysis. The group *t* test was employed for comparison of results obtained from various experimental procedures, unless otherwise indicated. Simple linear regression analyses were performed for $P_{A_{CO_2}}$ on $P_{A_{O_2}}$ at the onset of IVA during the course of BH (Fig. 4), and for the BH time on $\dot{V}_{O_2}$ from all available data points (Fig. 5).

RESULTS

Intraesophageal pressure. Upon commencement of breath holding (A) the intraesophageal pressure (IEP) increased from a subatmospheric pressure to above the ambient pressure and remained steadily at this level for a period of time (Fig. 2). A sharp reduction in IEP was then observed (B), indicating inspiratory effect with the glottis closed. Thereafter, this involuntary drive became more frequent and progressively stronger, until the conventional breaking point was reached (C). The sharp upward and then downward deflections just prior to BH (A in Fig. 2) represent maximal expiration followed by maximal inspiration. This level of IEP changes during the course of BH was quantified during BH at rest and BH during exercise (Fig. 3). The IEP for 10 determinations from five subjects at the onset of BH were 7.7 ± 2.7 and 3.3 ± 1.1 mmHg at rest and during exercise, respec-

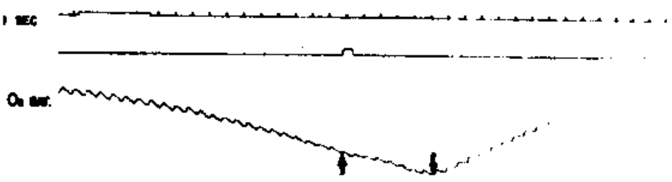

FIG. 1. Lung-to-ear oxygen appearance time. Inspiration following a period of breath holding is indicated by first arrow and oxygen appearance at the ear is indicated by second arrow. Breath hold begins at the far left of the graph.

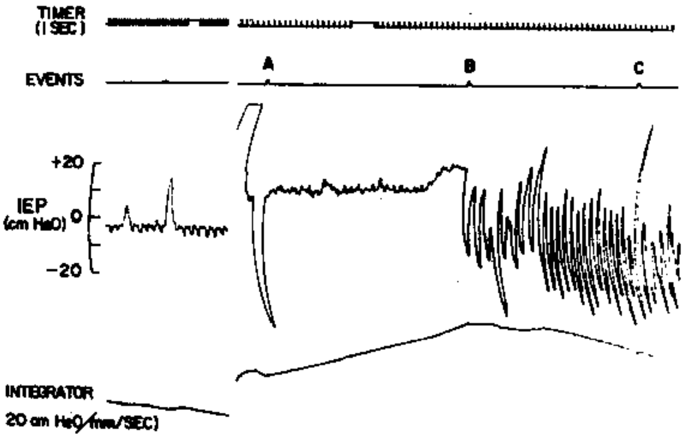

FIG. 2. A recording of esophageal pressure during a course of breath hold. Traces, from the top, represent 1-s timer, events, intraesophageal pressure (IEP) in cm of water, and integrator for the IEP, respectively. Slope of the integrator trace represents pressure; a rising slope indicates a positive and a falling slope indicates a negative pressure. Level of pressure is proportional to the slopes. Events are A, beginning of breath-hold (BH); B, onset of involuntary ventilatory activity; and C, termination of BH. Upward and downward deflections just prior to BH represent maximal expiration and maximal inspiration. Subject was exercising at a rate of 167 $kgm \cdot min^{-1}$ while breath holding. Entire BH lasted 70 s. A similar result was observed during BH at rest, but with a much longer BH time.

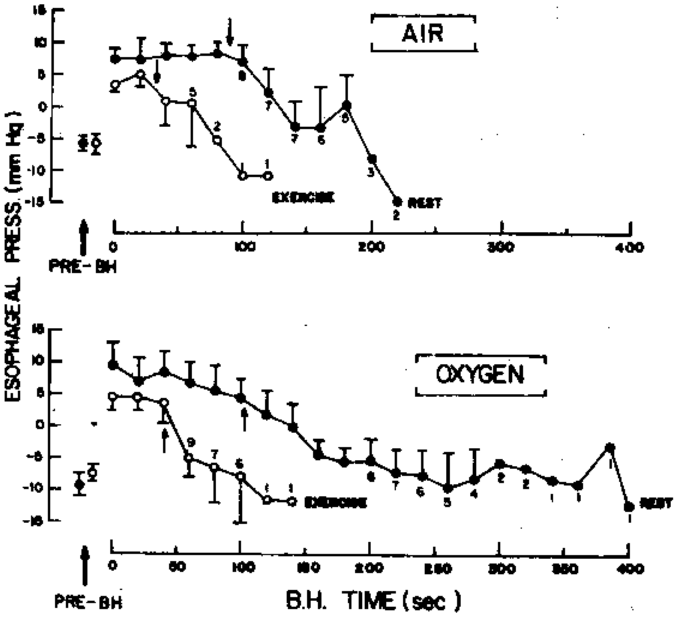

FIG. 3. Intraesophageal pressure (IEP) during breath hold (BH) with air and with oxygen at rest and during exercise at 167 kgm·min^{-1}. For clarity, only one side of standard errors are shown. Means of 10 determinations are shown unless otherwise indicated by numbers associated with each point. Each point represents the mean intraesophageal pressure for each consecutive 20-s period. Arrows indicate the onset of involuntary ventilatory activity.

tively, for BH with air; and were 10.5 ± 3.7 and 4.7 ± 2.1 mmHg at rest and during exercise, respectively, for BH with oxygen (Fig. 3). The mean IEP decreased continuously following the onset of involuntary ventilatory activity indicating the general predominance of inspiratory over expiratory activity. The magnitudes of pressure decreases are similar for BH at rest and BH during exercise, except for the time course.

Alveolar gas compositions. The calculated $P_{A_{CO_2}}$ values at the onset of BH were not statistically different between conditions, namely BH with air at rest and during exercise, and BH with oxygen at rest and during exercise. The mean values ranged from 12.0 to 13.4 mmHg. The calculated $P_{A_{O_2}}$ values at the onset of BH were also not different at rest and during exercise, either BH with air or BH with oxygen (Tables 2 and 3).

During BH with air, $P_{A_{CO_2}}$ at the onset of IVA was 45.8 ± 0.90 (SE) and 47.9 ± 1.47 mmHg at rest and during exercise, respectively ($P > 0.05$), whereas $P_{A_{O_2}}$ was 95.2 ± 4.89 and 85.9 ± 3.43 mmHg, respectively ($P > 0.05$). The BH could be continued after this time for an additional 72 s at rest and 34 s during exercise, until the conventional breaking point was reached. The alveolar gas compositions were also not different between rest and exercise at the conventional breaking point (Table 2).

$P_{A_{CO_2}}$ at the onset of the IVA was higher during exercise than at rest ($P < 0.01$) during BH with oxygen. However, there was no difference at the conventional breaking point. The $P_{A_{O_2}}$ at the onset of IVA was lower during exercise than at rest ($P < 0.05$), but no difference was observed at the conventional breaking point (Table 3).

TABLE 2. *$P_{A_{CO_2}}$ and $P_{A_{O_2}}$ during breath hold with air at rest and during steady-state exercise (167 kgm·min^{-1})*

Alveolar Gas Samples	$P_{A_{CO_2}}$, mmHg		$P_{A_{O_2}}$, mmHg		BH Time, s	
	Rest	Exercise	Rest	Exercise	Rest	Exercise
Calculated value at beginning of BH	12.0 ±0.69	13.20 ±0.94	134.6 ±0.85	132.4 ±0.94		
End-tidal before BH	39.0 ±0.59	42.8 ±1.67*	101.9 ±1.80	93.3 ±1.31†		
Onset of involuntary ventilatory activity	45.8 ±0.90	47.9 ±1.47	95.2 ±4.89	85.9 ±3.43	90 ±4.7	32 ±2.8†
Breaking point	53.5 ±1.65	56.2 ±2.61	61.9 ±4.10	57.5 ±4.31	162 ±13	66 ±7.0†

Values are means ± SE of 15 and 10 breath holds from five subjects at rest and during exercise, respectively. * Rest vs. exercise: $P < 0.05$. † $P < 0.001$.

TABLE 3. *$P_{A_{CO_2}}$ and $P_{A_{O_2}}$ during breath hold with oxygen at rest and during steady-state exercise (167 kgm·min^{-1})*

Alveolar Gas Samples	$P_{A_{CO_2}}$, mmHg		$P_{A_{O_2}}$, mmHg		BH Time, s	
	Rest ($N = 10$)	Exercise ($N = 10$)	Rest ($N = 6$)*	Exercise ($N = 10$)	Rest ($N = 10$)	Exercise ($N = 10$)
Calculated value at beginning of BH	12.0 ±0.73	13.4 ±0.75	693.7 ±2.75	685.8 ±1.83†		
End-tidal before BH	39.9 ±0.81	45.0 ±1.45‡	649.8 ±8.99	629.0 ±3.77†		
Onset of involuntary inspiratory activity	48.6 ±0.62	54.0 ±1.64‡	604.8 ±11.28	581.0 ±3.14	104 ±6.0	40 ±3.6§
Breaking point	63.8 ±2.59	66.9 ±3.35	553 ±15.6	534.5 ±10.10	275 ±21	102 ±9.3§

Values are means ± SE; (N = no of experiments). * Failure in P_{O_2} electrode in four experiments. † Rest vs. exercise: $P < 0.05$. ‡ $P < 0.01$. § $P < 0.001$.

When the alveolar gas data were plotted on a O_2-CO_2 diagram, the $P_{A_{CO_2}}$ at the onset of IVA differed little at various levels of $P_{A_{O_2}}$. The regression equation for this relationship was $P_{A_{CO_2}} = 0.00903\ P_{A_{O_2}} + 43.8$ (Fig. 4). The slope was not significantly different from zero. In other words, the $P_{A_{CO_2}}$ was nearly constant at the onset of IVA independent of $P_{A_{O_2}}$ level. The range of $P_{A_{O_2}}$ at the onset of IVA was 520-660 mmHg for BH with oxygen. However, when the $P_{A_{CO_2}}$ from these data were averaged, it was higher with oxygen than BH with air (Tables 2 and 3).

Estimation of Pa_{CO_2} at the brainstem. From the rate of $P_{A_{CO_2}}$ change and the oxygen appearance time from the lung to the ear, the Pa_{CO_2} at the brain can be estimated by:

$$Pa_{CO_2} = P_{A_{CO_2}} - \dot{P}_{A_{CO_2}}(T_1 + T_2) \qquad (1)$$

where T_1 = sampling time (3 s); T_2 = lung-to-ear time (s); and $\dot{P}_{A_{CO_2}}$ = rate of change of alveolar P_{CO_2}.

An example of the oxygen appearance time (T_2) at the ear following inspiration can be seen in Fig. 1. The average appearance time was 5.28 s at rest and 4.77 s during exercise which are insignificantly different from each other (Table 4).

The $P_{A_{CO_2}}$ between the onset of IVA and the conventional breaking point can be assessed by assuming a linear increase during this period (10, 11). $\dot{P}_{A_{CO_2}}$ was 0.09 and 0.21 mmHg·s^{-1} at rest and during exercise, respectively,

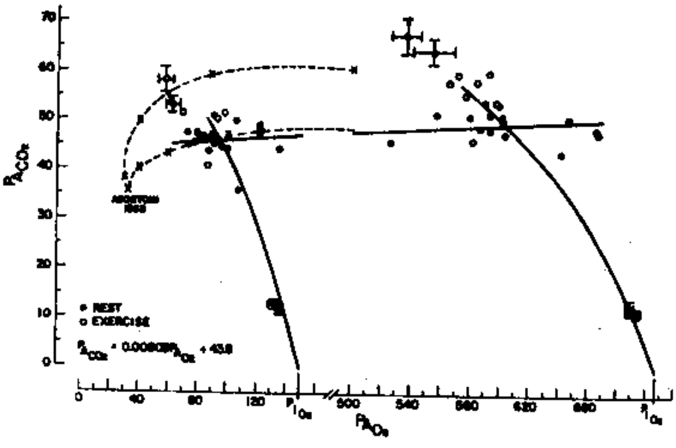

FIG. 4. Alveolar compositions at the onset of involuntary ventilatory activity (IVA) and the conventional breaking points. Individual data are plotted on O_2-CO_2 diagram for the onset of IVA. Solid dots indicate the breath hold (BH) at rest and the circles for BH during steady exercise at 167 kgm·min^{-1}. Average data (mean ± SE) at the conventional breaking point are grouped for clarity. Gas exchange paths are indicated with curved lines for BH with air ($P_{I_{O_2}}$ = 150 mmHg), and BH with oxygen ($P_{I_{O_2}}$ = 713 mmHg). Calculated alveolar compositions at the onset of BH are also indicated at the lower part of the graph on the gas exchange paths. Crosses (X) and broken lines are obtained from Agostoni's paper (1) for comparison. He obtained these curves for the onset of diaphragm activity (lower curve) and the conventional breaking point (upper curve) by varying the inspired gas mixtures.

TABLE 4. *Lung-to-ear oxygen appearance time*

Subj	Lung-to-Ear Time, s*	
	Rest	Exercise†
EH	5.15	5.25
YCL	4.43	4.38
SKH	6.85	5.10
DL	6.10	5.75
BR	3.85	3.40
Mean ± SE‡	5.26 ± 0.54	4.77 ± 0.41

* Average of four determinations each. † Exercise at 167 kgm·min^{-1}. ‡ Paired $t = 1.52$, $P > 0.2$.

during BH with oxygen and 0.11 and 0.24 mmHg·s^{-1} during BH with air.

Substituting the above values into *Eq. 1*, the arterial brain blood CO_2 partial pressure (Pa_{CO_2}) can be estimated. At the onset of IVA, the Pa_{CO_2} levels were 45 and 46.1 during BH with air, and 47.9 and 52.5 during BH with oxygen, respectively, at rest and during exercise. At the conventional breaking point, the Pa_{CO_2} levels were 52.7 and 54.4 during BH with air and 63.1 and 65.4 during BH with oxygen, respectively, at rest and during exercise. The difference of P_{CO_2} at the brainstem was small between rest and exercise conditions.

Oxygen consumption and BH time. Figure 5 is a plot of BH time (seconds) against the reciprocal of oxygen consumption in ml·s^{-1}. The conventional oxygen consumption unit is also indicated. The BH time, to either the onset of IVA or the conventional breaking point, can be described by: $T = a(1/\dot{V}_{O_2}) + b$, where T is BH time in s; $\dot{V}_{O_2}$ is oxygen consumption in ml·s^{-1}; and a and b are constants.

The duration of BH was significantly correlated with the reciprocal of oxygen consumption. The correlation coefficients are 0.72 and 0.78 for BH with oxygen and air, respectively, at the conventional breaking point; and 0.85 and 0.89 for BH with oxygen and air, respectively, at the onset of IVA. All correlation coefficients are statistically significant at the 0.001 level (Table 5).

DISCUSSION

Agostoni (1) reported that the activity of the muscles of the abdominal wall is negligible during a BH. Recording of the electrical activity from the diaphragm during a BH proved that the contraction of the diaphragm accounts for the lowering of the intrathoracic pressure (or IEP) observed during the later phase of BH (1). It has been the experience of divers that the respiratory movement with glottis closed relieves the urge to initiate breathing. The tolerance to hypercapnic and hypoxic stimuli is higher if respiratory movements are made during the course of a breath hold (7, 18). Consistent with these observations, Godfrey and Campbell (8) hypothesized that the cessation of respiratory movement elevates the central excitatory state, hence the urge to resume breathing.

The course of a BH thus can be characterized by *a*) voluntary inhibition of inspiratory muscular activity with a steady intrathoracic pressure which is above the ambient,

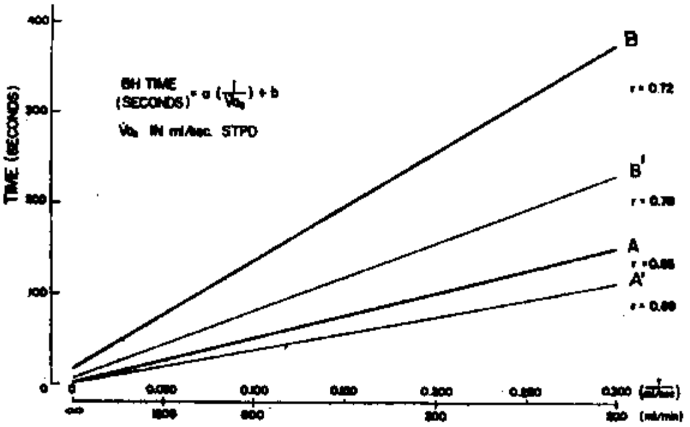

FIG. 5. Relationship between the oxygen consumption and breath-hold time. *Abscissa* is the oxygen consumption in ml·min^{-1} (lower figures) and in 1/ml·s^{-1}. Lines A and B represent the onset of involuntary ventilatory activity (IVA) and the conventional breaking point (CBP), respectively, for BH with oxygen. Line A′ and B′ represent IVA and CBP, respectively, for BH with air. Regression equations are listed in Table 5.

TABLE 5. *Oxygen supply-demand relationship*

Breaking Points	BH with	$T = a\ (1/\dot{V}_{O_2})$	+ b	N	r*
A	air	T = 388 $(1/\dot{V}_{O_2})$	+ 0.1	25	0.89
	O_2	T = 491 $(1/\dot{V}_{O_2})$	+ 2.7	23	0.85
B	air	T = 747 $(1/\dot{V}_{O_2})$	+ 7.4	25	0.78
	O_2	T = 1,100 $(1/\dot{V}_{O_2})$	+ 19.0	23	0.72

Breaking points: A, at the onset of involuntary ventilatory activity; B, at the conventional breaking point; *T:* breath-holding time, in s; $\dot{V}_{O_2}$: ml·s^{-1} (STPD); *a* and *b* are constants; *N:* total number of determinations on five subjects. * $P < 0.001$.

b) involuntary inspiratory muscular activity with the glottis closed while the intrathoracic pressure is subatmospheric and cyclic with variable intervals, and *c*) voluntary opening of the airway. The involuntary ventilatory activities (IVA) with the glottis closed generate periodic subatmospheric pressures in the thoracic cavity, which are beneficial to the venous return and, hence, the cardiac output. This activity may explain why in our previous studies, the cardiac output is not altered during the later portion of a BH during which the heart rate is decreased (11). In the early part of a BH when the intrathoracic pressure is above ambient and the respiratory pumping activity is suspended, one would expect to find a reduction in venous return and hence the cardiac output, except immediately following a deep inspiration (21).

We have not found differences in $\mathrm{PA_{CO_2}}$ at the conventional breaking point, between rest and during exercise. It has been reported that the $\mathrm{PA_{CO_2}}$ at the breaking point of BH is higher during exercise than at rest under various exercise conditions (2–5). The significance of these findings has been discussed by Mithoefer (17). The differences between rest and exercise tend to be exaggerated by heavy exercise rates, and short BH time. It is well documented that the $\mathrm{CO_2}$ transfer from the blood into the lung is greatest during the first 20 s of BH after which it proceeds at a much lower rate (10, 16). In our subjects, the average BH time to conventional breaking point was 162 s at rest and 66 s during exercise. The rate of $\mathrm{PA_{CO_2}}$ changes was low during these periods which may account for the insignificant difference in $\mathrm{PA_{CO_2}}$ between rest and during exercise. However, the time to IVA was considerably shorter than the conventional breaking point, and a higher $\mathrm{PA_{CO_2}}$ was found during exercise than at rest (Table 3).

The appearance time of $\mathrm{O_2}$ from the lung to the ear was essentially identical at rest and during exercise (Table 5). This finding is consistent with stability of cerebral blood flow at rest and during exercise (13, 25). The average appearance time of 5.28 s at rest and 4.77 s during exercise is considerably shorter than that assumed by Agostoni (1), and that calculated by Craig and Babcock (3) for circulation time from the pulmonary capillary to brainstem. Our results are within the range as measured by Williams with a similar method (27).

The variability of breath-hold time for a given oxygen consumption is much greater at the conventional breaking point than at the beginning of IVA. For example, at $\dot{V}\mathrm{O_2}$ of around 300 $\mathrm{ml \cdot min^{-1}}$, the BH time to the conventional breaking point varied from 160 to over 400 s for BH with oxygen, and from less than 100 to over 200 s for BH with air.

The values of *b*, the intercept in this relationship, are small and insignificantly different from each other and from zero, implying that when the oxygen consumption is infinitely large, it is impossible to breath hold. Therefore, the above equation can be simplified to:

$$T \times \dot{V}\mathrm{O_2} = a \qquad (2)$$

The constant *a* is the amount of oxygen consumed during the period of BH, and is equal to that supplied by the blood and the lung. The amount of oxygen consumed up to the beginning of IVA is 388 and 491 ml for breath hold with air and with oxygen, respectively. For the conventional breaking point, the corresponding values are 747 and 1,100 ml for breath hold with air and oxygen (Table 5).

Previous studies carried out in our laboratory indicated that the lung can provide enough oxygen for the first 2 min of BH with air at total lung capacity, prior to the depression of blood oxygen content (10), implying that the lung supplies the entire amount of oxygen consumed up to the beginning of IVA. The $\mathrm{PA_{O_2}}$ was 95.2 and 85.9 mmHg at rest and during exercise, at the onset of IVA. These $\mathrm{PA_{O_2}}$ levels could keep the arterial blood $\mathrm{Po_2}$ greater than 90 % saturated (10). Again, the IVA appears to be initiated by the level of $\mathrm{PA_{CO_2}}$, and, it is obviously a metabolic rate-dependent phenomenon.

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Effects of alveolar P_{O_2} and P_{CO_2} on apneic bradycardia in man

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MOORE, T. O., R. ELSNER, Y. C. LIN, D. A. LALLY, AND S. K. HONG. *Effects of alveolar P_{O_2} and P_{CO_2} on apneic bradycardia in man.* J. Appl. Physiol. 34(6): 795-798. 1973.—Heart rate to 40 sec of apnea was monitored under conditions which would selectively alter alveolar P_{O_2} and P_{CO_2}—air or O_2 prebreathing, rest, exercise, and rebreathing maneuvers. Face immersions were also included to potentiate the observed apneic bradycardia. During both rest and exercise, the bradycardial response was potentiated in the presence of low alveolar P_{O_2}, attenuated under conditions of high $P_{A_{O_2}}$, and appeared to be independent of $P_{A_{CO_2}}$ levels. These data support the theory that the "diving" bradycardia in man is also an oxygen-conserving mechanism as has been postulated for diving marine mammals. While $P_{A_{O_2}}$ levels do not initiate the bradycardial response to apnea, they should be included in the multiple factors which modify the response.

oxygen conservation; diving

IT HAS BEEN SUGGESTED that the breath-hold bradycardia observed in marine mammals during diving is related to an oxygen-conserving mechanism (9, 14). This bradycardia, coupled with peripheral vasoconstriction, leads to an increase in central blood volume. The end result is a maintained perfusion of heart, lungs, and brain at the expense of peripheral, less "important" tissues.

Apneic bradycardia is also demonstrable in man and may be modified by a variety of sensory stimuli (2, 4, 15). Song et al. (15) recently postulated a multiple-factor theory to summarize the experimental data from several laboratories. This theory states that there is a bradycardia in man accompanying apnea, per se, which may be modified by both "mechanical" and more purely "neural" factors. The mechanical component is predominantly induced, experimentally, by hydrostatic pressure during whole-body immersion and involves a reduction in extrathoracic blood volume. In consequence, peripheral resistance is elevated, intrathoracic blood volume is increased, and the resultant increase in central blood pressure leads to a barorefléxive decrease in heart rate (5, 12). The neural components involve *1)* the observed accentuation of apneic bradycardia when apnea is initiated at elevated lung volumes (and attenuation at low lung volumes) and *2)* the potentiation of the bradycardia under conditions of face immersion, becoming more pronounced as water temperature decreases. In the one case, intrathoracic stretch receptors have been implicated (8, 10). In the other, cold receptors of the facial trigeminal nerve endings are involved (1, 2). The final common path in all cases is an increase in vagal tone resulting in a decreased heart rate.

If the presence of an apneic bradycardia in man also represents an oxygen-conserving mechanism, a sensory pathway susceptible to alteration in oxygen partial pressure might be expected. Elsner et al. (6) have reported that the fall in arterial oxygen tension during "diving" after hyperventilation with O_2 varied inversely with a fall in forearm blood flow. This was supportive of the oxygen-conserving concept. However, the authors cautioned that, since a significant diving response was evoked by apnea during face immersion in the presence of arterial hyperoxia (with or without hypocapnia), the response, per se, is basically independent of asphyxial blood gas changes.

The experiments reported here attempt to ascertain the existence and relative importance of the potential "chemocomponent" in the heart rate response to apnea in man.

METHODS

Five adult male subjects performed breath holds (BH) for 40 sec after the following maneuvers: *1)* after rebreathing (RB) air (to end-tidal alveolar CO_2 levels of 6% as indicated by a Beckman model LB-1 CO_2 analyzer) with consequent lowering of $P_{A_{O_2}}$ and elevation of $P_{A_{CO_2}}$ compared to controls, *2)* after RB air, but with BH initiated on one breath of O_2, leading to elevated $P_{A_{CO_2}}$ in the presence of hyperoxia, *3)* after mild nonsteady-state exercise breathing air, resulting in lowered $P_{A_{O_2}}$ and elevated $P_{A_{CO_2}}$, *4)* after mild nonsteady-state exercise breathing air, with BH begun on one breath of pure O_2, leading to elevated $P_{A_{CO_2}}$ and hyperoxia, *5)* after mild steady-state exercise on O_2 (with prior O_2 breathing for 10 min) resulting in greater hyperoxia plus hypercapnea. Control (preapneic) $P_{A_{O_2}}$ and $P_{A_{CO_2}}$ values were 104 ± 6 and 39 ± 2 mm Hg, respectively, in the resting state.

All maneuvers were performed with and without face immersion (FI) in 26 C water, to take advantage of the potentiating effect of FI. ECG was monitored for heart rate, alveolar gas samples were delivered into a Haldane tube at the 40-sec break point, and control heart rates were taken just prior to BH in all cases. Alveolar gas samples were analyzed by Scholander micro-gas analyzer (13) and a Radiometer PHM 71 gas analyzer (Copenhagen, Den-

TABLE 1. *Control (premaneuver) heart rates*

	Rest		
	Air/no RB	Air/RB	Air/RB O_2 (1B)
Face out	73 ± 6	81 ± 7	80 ± 6
Face imm	74 ± 5	83 ± 7	83 ± 8
	Exercise		
	Air/exer	Air/exer O_2 (1B)	O_2 exer/SS WO
Face out	98 ± 8	102 ± 10	115 ± 7
Face imm	100 ± 11	99 ± 8	119 ± 7

Values are means ± SE, in beats/min.

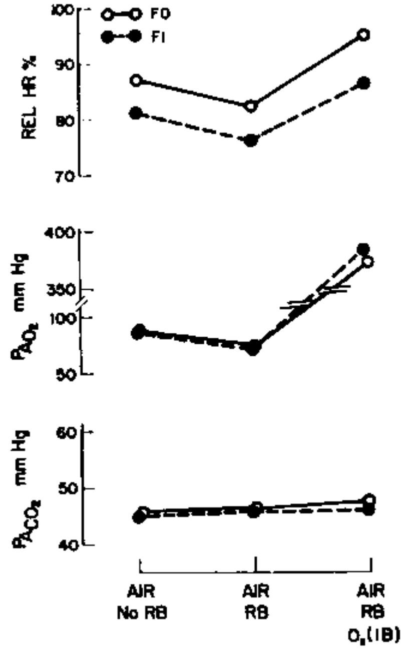

FIG. 1. Heart rate responses (as % control values), $P_{A_{O_2}}$, and $P_{A_{CO_2}}$ levels at the end of 40-sec breath holds on air, after rebreathing (RB) and on one breath of O_2 after RB on air. Dashed lines represent the face immersion (FI), and solid lines the nonimmersed condition (FO).

mark) in the case of oxygen breathing. Calibration gases for the latter were analyzed via Scholander micro-gas analyzer. In the nonsteady-state (NSS) exercise series, the exercise began approximately 10 sec before the onset of BH. In the steady-state (SS) exercise trial, a leveling of heart rate was taken to indicate steady state (approximately 3 min). Exercise was performed on a bicycle ergometer; work level was 200 kpm/min in all cases and continued throughout the period of apnea.

Heart rate results are expressed as maximum relative (percent) change from control levels and statistical significance was determined by paired sample analysis. Control heart rates in beats per minute are given in Table 1 for reference. These represent rates just prior to apnea under the breathing conditions indicated by the table.

RESULTS AND DISCUSSION

Figure 1 summarizes the data from the resting trials. BH in air resulted in a 12 ± 2% (SE) decrease in heart rate. Upon FI, the decrease was 18 ± 2%. These data are consistent with other, similar experiments (8, 11). Breakpoint $P_{A_{O_2}}$ and $P_{A_{CO_2}}$ were 87 ± 8.1 and 46 ± 4.0 mm Hg under this condition. Alveolar gas tensions were essentially the same with or without FI.

After RB on air, with $P_{A_{O_2}}$ of 68 ± 5.8 and $P_{A_{CO_2}}$ of 47 ± 5.2 mm Hg, the reduction in heart rate was 18 ± 2.0% without FI and 24 ± 2.1% with FI. Both are significantly different from control levels ($P < 0.01$). Thus, while $P_{A_{CO_2}}$ remained constant, a significant ($P < 0.05$) 30% potentiation of bradycardia occurred concomitant to a 22% decrease in $P_{A_{O_2}}$. Again, no difference in alveolar gas pressures were obtained whether or not FI accompanied the apnea.

After RB on air, with subsequent BH on one breath of 100% O_2, both the FI and no-FI heart rate responses were significantly attenuated ($P < 0.05$) compared to the responses in the preceding BH maneuver. $P_{A_{CO_2}}$ was again fairly constant (48 ± 3.0 mm Hg without FI; 47 ± 3.3 mm Hg with FI) while $P_{A_{O_2}}$ was elevated to approximately 358 ± 16 mm Hg under both conditions. However, the degree of bradycardia was only 5% and 12%, respectively, approximately 50% of the response seen in air BH controls.

The data from the resting series, therefore, suggest that

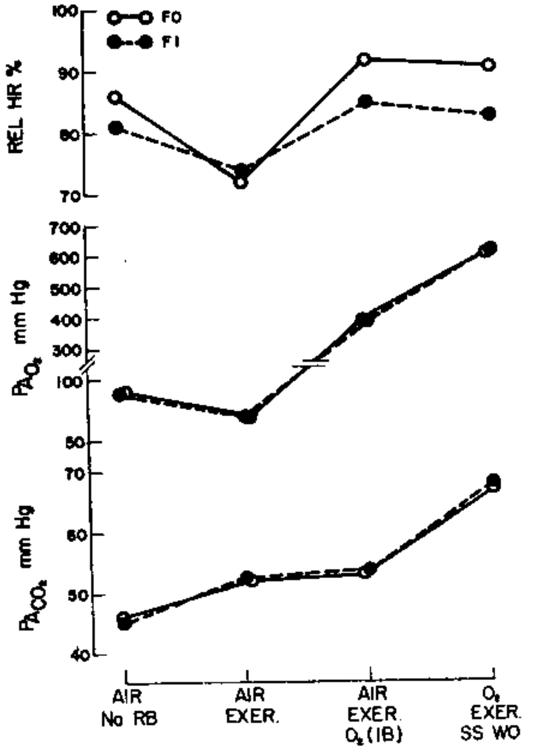

FIG. 2. Heart rate responses (as % control values), $P_{A_{O_2}}$, and $P_{A_{CO_2}}$ levels at the end of 40-sec breath holds during exercise after breathing air, after breathing air and starting apnea with one breath of O_2, and under steady-state (SS) exercise following oxygen washout (WO). The air no-RB points are as in Fig. 1 and given for reference. Dashed lines = FI; solid lines = FO.

the degree of apneic bradycardia qualitatively follows the change in $P_{A_{O_2}}$ in the presence of constant $P_{A_{CO_2}}$.

This is also consistent with the earlier report (6) describing the inverse relationship between arterial P_{O_2} and forearm blood flow, if one considers both bradycardia and reduced peripheral flow to be integral components of the diving response in man. From a previous study (7) in this laboratory, decreases in alveolar and arterial P_{O_2} were seen to parallel one another during BH of intervals up to four minutes. Heart rates were found to fall maximally within the first 40 sec of apnea. Further falls in Pa_{O_2} did not elicit further bradycardia beyond that apneic interval. Mean arterial pressure, however, continued to rise through 3 min of apnea, suggesting that peripheral resistance changes are a more tenacious component of response to asphyxia than bradycardial responses.

In the exercise experiments (Fig. 2), bradycardia is again seen to be potentiated or attenuated in the same directions as seen in the resting series, namely, exercise on air, with superimposed apnea, led to a greater degree of hypoxia ($P_{A_{O_2}}$ = 61 ± 4 mm Hg) and hypercapnia ($P_{A_{CO_2}}$ = 52 ± 3 mm Hg) and a greater degree of bradycardia than during rest (72% of control levels without FI, 73% with FI).

During the next maneuver (NSS exercise, but with initiation of apnea after one breath of O_2), $P_{A_{CO_2}}$ levels did not change from those of the previous maneuver (53 ± 3 mm Hg) while $P_{A_{O_2}}$ was elevated to 377 ± 23 mm Hg (this is approximately the same as the equivalent experiment without exercise). In this case, bradycardial response (no FI) was attenuated from the air exercise response (72%) to only 93 ± 6% of control levels. Comparable FI values were 73% and 88%. These data again support the argument that $P_{A_{CO_2}}$ changes are not relevant to the responses observed in HR.

In the final experiment, in which SS exercise was performed on O_2 (after 10 min of prior O_2 breathing) no further attenuation of HR response was seen. $P_{A_{O_2}}$ was 617 ± 31 mm Hg in this case. Unfortunately, no attempt was made to control $P_{A_{CO_2}}$ levels, which rose to 68 ± 4 mm Hg, raising the question of a hypercapnic contribution to the "leveling off" of the HR response.

However, two previous studies tend to dismiss $P_{A_{CO_2}}$ levels as being of any great importance to the response. Elsner et al. (6) performed experiments in which subjects both breathed normally on O_2 and hyperventilated on O_2 prior to apnea. Heart rate responses were not different between the two cases. Additionally, in this laboratory (11) we have reported alterations in responses found during apnea in deck-decompression chambers under conditions in which $P_{A_{CO_2}}$ was not a variable but $P_{A_{O_2}}$ was. The inverse relation between $P_{A_{O_2}}$ and degree of apneic bradycardia was found to hold, up to, but not beyond, approximately 450 mm Hg.

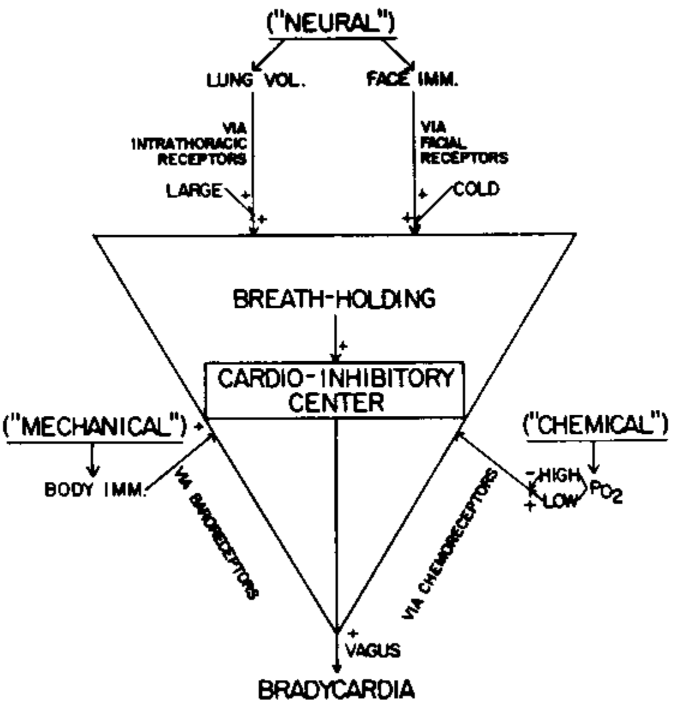

FIG. 3. Schematic model of factors modifying apneic bradycardia, including the P_{O_2} "chemocomponent." + Indicates potentiation; − indicates attenuation of bradycardial responses. Triangle symbolizes the different types of modifying stimuli to the basic bradycardia accompanying apnea, per se.

In summary, the simulation of mildly asphyxic conditions (and selective reversal of those conditions) by two different methods has yielded data on apneic bradycardia consistent with the theory that the diving response in man is indeed modified by the oxygen-exchange state of the body, and therefore, plays an oxygen-conservation role during a dive. Figure 3 represents a proposed modification of Song's "multiple-factor" theory which incorporates an oxygen-related chemocomponent. We second the suggestion of Elsner et al. (6) that the oxygen-conservation role is relatively weak, and serves only to modify, not to initiate, the response.

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Effects of temperature, immersion, and ambient pressure on human apneic bradycardia

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MOORE, T. O., Y. C. LIN, D. A. LALLY, AND S. K. HONG. *Effects of temperature, immersion, and ambient pressure on human apneic bradycardia.* J. Appl. Physiol. 33(1): 36–41. 1972.—Heart rate (HR) responses to 60-sec breath holds (BH) were recorded during *1*) face immersion (FI) with varied water and air temperatures, *2*) FI with and without face wetting, *3*) whole-body immersion at different temperatures, *4*) wet dives to 18.3 m, *4*) dry chamber dives to 30.2 m, and *5*) chamber dives with gases of different percent oxygen. HR decreased during apnea in air, the response was potentiated by FI, and greatest bradycardia appeared with 5 C FI. This pattern was similar in 25 and 5 C ambient air, but bradycardia, as percent of control HR, was less in the latter. BH bradycardia was not different during FI in 5 C water whether the face was wetted or protected. Apneic bradycardia during whole-body immersion was similar to FI alone and appeared to be temperature rather than immersion dependent. Wet ocean dives and dry chamber dives indicated an attenuation of bradycardia with depth (wet), with some evidence that elevated oxygen tension was responsible within certain ranges. It was concluded that multiple rather than single factors are responsible for the HR response to apnea.

pressure and bradycardia; immersion bradycardia

A SUBMERSION BRADYCARDIA in diving animals was described by Paul Bert a century ago (4), and the phenomenon has subsequently been reported in a variety of diving mammals (2, 10, 16) and man (5, 8, 14, 15).

Irving, Scholander, and associates (15, 16, 22) have speculated that apneic bradycardia during diving might represent part of a reflex in which oxygen conservation is the end result.

Since intrapleural pressures (8, 23) and water temperature (2, 8, 13, 17) do indeed affect the apneic bradycardia during immersion, several explanations of the mechanisms have arisen. Craig (7, 8) and others (12, 19, 20) have explained the bradycardia on the basis of hemodynamic and mechanical properties (i.e., baroreflexes) of the cardiovascular system during apnea, while others have reported that the reflex arises via afferent discharge from peripheral receptors notably in the nose, on the face, or in thoracic muscles (2, 6, 10, 17, 24).

Recently, Song and co-workers (23) have proposed a "multiple-factor" theory which incorporates both mechanical and neural stimuli ultimately impinging on a final common efferent pathway—the vagus nerve. They proposed that the act of breath holding (BH) itself initiates a bradycardial response, which may then be potentiated by at least three factors, one mechanical (due to immersion with increased central blood volume) and two neural (one lung volume dependent and the other temperature dependent).

The study reported here extends the data to the apneic bradycardial response observed, *1*) with face immersion (FI) alone but with varying water and ambient air temperatures, *2*) upon face immersion with and without face wetting, *3*) with whole-body immersion at various water temperatures with and without face immersion, *4*) with breath holding at water depths from surface to 18.3 m at constant water temperature, and in a dry recompression chamber to simulated depths of 30.2 m, and *5*) to assess the potential role of a P_{O_2} "chemocomponent" in apneic bradycardia. Data from the latter were gathered both by direct manipulation of inspired P_{O_2} by gas mixing and by natural alterations occurring with increased air pressures.

Though certain human adaptations to the breath-hold diving situation do occur which are similar to other diving mammals, the content of the present experiments pertains only to man and no generalization is presently possible regarding applicability to diving mammals as a whole.

METHODS

In all cases, ECG was recorded on a polygraph (E & M Physiograph model 4A, Houston, Texas) via the use of two precordial electrodes (Beckman). Heart rates were counted over 10-sec intervals throughout apnea. Breath-hold duration was standardized at 1 min and all were performed with full vital capacity. A total of 24 (avg age = 27.5 ± 2.3 years) healthy male adult subjects were monitored: 21 in the face immersion (FI) series, 7 in the whole-body immersion series, and 6 in the depth studies. The numbers were reduced in the latter two studies because certification with scuba was a requirement. The subjects in the whole-body immersion study had all participated in the FI experiments. One new subject was added in the chamber experiments who had not participated in the others.

Experiment 1: FI at different water and ambient air temperatures. The subjects, wearing only shorts, sat quietly for 10 min until control heart rate became stabilized. Recovery to stable resting heart rate was allowed between apneic maneuvers. Water temperatures were maintained ± 1.0 C with either ice or hot water. This series was done in ambient room air of 25 and 5 C (Hotpack environmental room model 683). Temperature thermistors (General Transducer) were attached to the skin at the following points: forehead, cheek, upper arm, hand, finger, scapula, abdomen, thigh, calf, foot, and toe; and rectal temperature was monitored. Blood pressures were monitored by standard auscultation. Total exposure time was approximately 30–40 min in each ambient air temperature, with two consecutive and identical apneic series consisting of BH in air, and with FI in 25, 15, and 5 C water. Air temperatures were maintained ± 1 C throughout the exposure.

Experiment 2: wetting vs. nonwetting on FI bradycardia. The procedures above were repeated with FI in 5 C water, 25 C ambient air, under FI conditions where the face was wetted or dry. The latter was achieved by covering the face closely with thin plastic wrap. Forehead skin temperatures were not different, wet or dry, during the course of breath holding in this series.

Experiment 3: immersion studies. A record was made during 60-sec apnea while the subjects stood quietly in 25 C ambient air outside the immersion tank. Each subject then stood in the tank for 5 min with water level to the neck (lung apex) and performed breath holds with and without FI. This procedure was repeated in water of 35, 25, and 15 C.

Experiment 4: open ocean and simulated depths. One study was carried out in the open ocean with certified scuba divers as subjects. ECG was recorded via umbilical hard-wire on a portable four-channel FM tape recorder (Lockheed Electronics model 411C) and played back onto a polygraph in the laboratory. The subjects performed their breath holds from scuba at water depths of 18.3, 13.8, 9.2, and 4.6 m, and at the water surface (still immersed).

The series was repeated with FI only in water of 25 C in a deck decompression chamber (Makai Undersea Test Range, Inc.) at air pressure-simulated depths of 30.2, 20.1, and 10.1 m and at the surface. Ambient air temperatures were approximately 26–27 C.

Experiment 5: effects of altered $P_{I_{O_2}}$. Additional experiments were carried out in which one minute BH were performed with FI in 25 C water after breathing air, 40% O_2 and 100% O_2 at 1 Ata; and air, 50% O_2, and 10.3% O_2 at 2 Ata simulated depth.

Heart rate responses are reported as the minimum heart rate (%) of initial control rate (taken in the 10 sec just prior to breath hold) which occurred during the course of the apneic maneuver. In the experiments described in this study, with the exception of the ambient air studies (below *exp 1*), no difference was observed in control rates to warrant the use of an expression of maximal response in terms other than percent of control.

RESULTS AND DISCUSSION

1) Face immersion: effects of water and ambient air temperatures. These data are presented in Fig. 1. Statistical differences were calculated by paired sample analysis. Control heart rates were those counted in the last 10 sec rest prior to BH in every case.

In ambient air of 25 C, two effects were seen: a significant ($P < 0.01$) bradycardia due to BH per se in air, and a higher degree of bradycardia when the BH was performed with FI, the greatest response being with FI in 5 C water ($56 \pm 2\%$ SE of control rate, $P < 0.001$). In 5 C ambient air a similar pattern was observed with a maximum reduction upon FI in 5 C water ($63 \pm 3.5\%$ of control rate, $P < 0.001$).

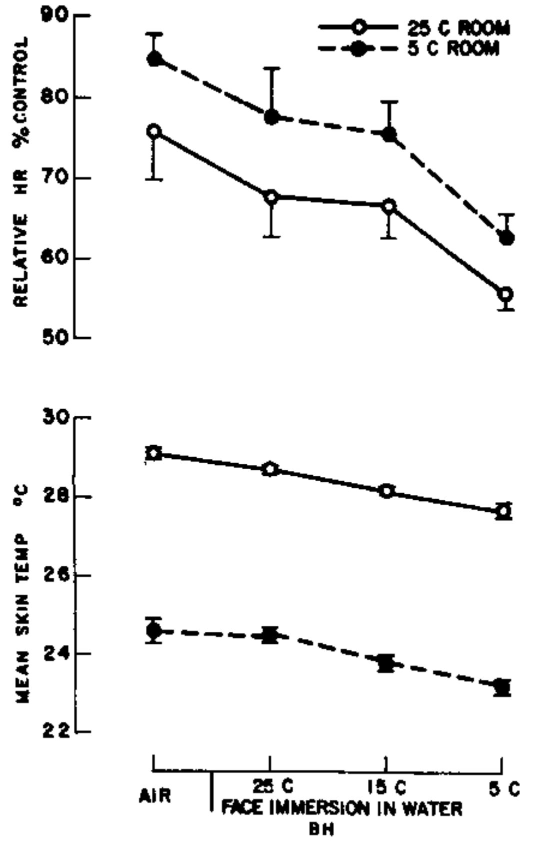

FIG. 1. Relative heart rate and skin temperatures during 60-sec BH in air and with FI in 25, 15, and 5 C water in 25 and 5 C ambient air temperature.

Calculated mean skin temperature (3) decreased over the course of the experiment in both ambient air temperatures and, in the 5 C room, was accompanied by an increase in diastolic blood pressure of 10 mm Hg. In the 5 C room, resting diastolic pressure was 94.8 ± 8.4 mm Hg compared to 84.7 ± 9.0 in the 25 C room, while systolic pressures were not different.

The resting control heart rate also decreased by 13 beats/min. While the general pattern of response was similar to that in 25 C room, the relative degree of bradycardial response to a given apneic maneuver was clearly less in 5 C room. However, there was no difference in terms of the lowest absolute heart rate during apnea between the two room temperatures. These data on absolute heart rates under the various BH conditions appear in Table 1.

Between groups (25 vs. 5 C room) there was significant ($P < 0.01$) attenuation of the maximum relative response in 5 C ambient air under all conditions. During FI in 5 C water, less difference was observed.

The temperature dependence of the bradycardia response to face immersion was well illustrated by the data in this experimental series, with similar patterns whether the subjects were exposed to 25 or 5 C ambient air. Bradycardia upon face immersion was most pronounced in 5 C water,

TABLE 1. *Absolute heart rates during BH in air, and during FI in water of 25, 15, and 5 C in ambient air temperatures of 25 and 5 C*

	Room Temperature	
	25 C	5 C
Control	96 ± 7	83 ± 5
BH air	76 ± 4	71 ± 5
$FI_{25\ C}$	70 ± 6	67 ± 5
$FI_{15\ C}$	66 ± 5	65 ± 4
$FI_{5\ C}$	56 ± 4	55 ± 4

Values are means ± SE, given in beats/min.

less so and approximately the same in 25 and 15 C water (see Fig. 1).

The attenuation of the response in 5 C ambient air merits comment. The decrease in mean skin temperature under this experimental condition was consistent with the occurrence of peripheral vasoconstriction as reported by others upon exposure to 5 C ambient air (21). With the attendant increase in peripheral resistance, it seems likely that the slower resting heart rates in the cold were the result of baroreflexes with a resulting increase in vagal discharge to the heart. In terms of the lowest absolute heart rate observed, there was no difference in the 25 or 5 C room during any BH maneuver (see Table 1), suggesting a similar degree of vagal tone.

2) Face immersion: effects of wetting vs. nonwetting on BH bradycardia. To assess the role of wetting the face on the degree of response to FI, this study was done with FI in 5 C water, which condition produces a marked bradycardia.

Breath holding in air (25 C) produced a heart rate decrease to 83 ± 6% of control rates ($P < 0.025$). When the BH was performed allowing face wetting, the mean response was 65 ± 4% of control values ($P < 0.001$). With the face protected from wetting, the comparable values were 66 ± 5 % of control ($P < 0.001$). There was no significant difference in response to the BH in 5 C water between the wet and dry conditions.

The results of this FI series suggest that wetting the face is not a requisite for the response to BH during FI. However, Elsner reports (personal communication) that this may not be the case in more moderate water temperatures (approximately 22–23 C), but rather an additional slowing of heart rate occurs when the face is wetted, compared with covered. Preliminary data in this laboratory support this finding as does that of Kawakami et al. (17) during FI at 10–17 C and with ice bag application. In this respect, the time course for facial temperature change would be extended with the face covered in 25 C water to a greater degree than with the higher temperature gradient existing during FI in 5 C water. While wetting the face, per se, may not be required to elicit a particular degree of bradycardia, it seems likely that it potentiates heat transfer from the skin. At moderate face immersion water temperatures, therefore, wetting can contribute to stimulus perception more so than at more extreme temperature, because in the latter, heat transfer is already rapid, even through plastic wrap.

3) Whole-body immersion experiments. The data are presented in Fig. 2. Eupneic (no BH) rates while standing outside the tank averaged 83 ± 5.0 beats/min. Room temperature was 26 C. The comparable rates standing quietly in water (to neck) of 35, 25, and 15 C for 5 min were 88 ± 4.5, 85 ± 8.2, and 90 ± 9.5 beats/min, respectively, with no significant differences between any of the four conditions.

Upon BH in 35 C water, there was no significant deviation from nonimmersed BH heart rate under any immersion condition. However, when the subjects were immersed to the neck in 25 and 15 C water for 5 min, the degree of apneic bradycardia was greater, and a further potentiation of bradycardial response was observed during BH with face immersed. The subjects also submerged totally and breathed on scuba, followed by BH with and without a face mask. The degree of bradycardia observed was not different from that during FI while immersed to the neck.

These involved both immersion and temperature variation. Song et al. (23) reported the effects of elevated intrapleural pressure on the attenuation of the bradycardial response to BH at several lung volumes, TLC among them. Agostoni's group (1) found that intrapleural pressure at the end of normal expiration became approximately 4 cm H_2O relatively positive when submerged to the neck compared to that in air. Their volume-pressure curves indicate that at lung volumes approaching TLC this difference from the nonsubmerged condition might be somewhat less. Thus, according to Song's data, the intrapleural pressure changes at TLC due only to immersion would predict only slight changes in the heart rate response to apnea. Our data generally agree, showing no great difference between BH in air and in water of 35 C (see Fig. 2). In 25 and 15 C water, the tendency was toward a potentiation of the response. However, the contribution of nonfacial receptors appears to be less than that of facial cold receptors. With immersion common to all conditions, it appears that any effects seen are due to thermal stimulus rather than immersion, per se, at shallow depth, and that the maximal bradycardial responses during whole-body immersion are the

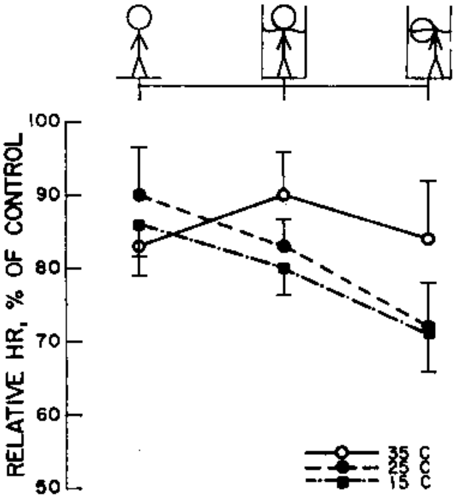

FIG. 2. Relative heart rate responses to 60-sec BH in air, immersed to the neck, and immersed to the neck with FI in water of 35, 25, and 15 C.

same as if only the face were immersed at any identical water temperature. In no case was there a difference in response between FI alone and whole-body immersion with or without face mask. The thermal experience of whole-body immersion is apparently sufficient to elicit the response even with areas of the face covered by a mask. This finding has practical advantage for relating open-ocean scuba conditions to laboratory conditions.

One major inconsistency appeared in the immersion data. Whereas in the 5 C room air a slower eupneic heart rate was observed, compared with the 25 C room (ascribed to peripheral vasoconstriction and consequent baroreflex), no reduction in resting rate appeared during immersion in water of any temperature ($\bar{x}$ = 88 ± 4.0 beats/min). No blood pressure measurements were made during immersion. However, in the colder water especially, peripheral vasoconstriction would also be expected. There is, unfortunately, no broad and systematic literature relating air and water temperatures comparable in terms of whole-body thermal experience. In an earlier study (18), a transient decrease in resting heart rate was observed immediately upon immersion in 25 and 15 C water, disappearing after a few minutes exposure. In the present case, it is perhaps plausible to assume that the excitement/apprehension associated with the immersion provided a sympathetic response counter to the expected baroreflex slowing of the heart. In colder water, an increase in metabolic rate might contribute to a higher heart rate, and increased $\dot{V}_{O_2}$ was noted (18) during exposure to 25 C and 15 C immersion. Air values were 0.36 liter/min vs. 0.52 and 0.45 for 25 and 15 C water, respectively.

4) Open ocean and simulated depth experiments. In this study, to depths of 18.3 m, BH was performed using scuba. Subjects were relaxed and maintained position by hanging onto an anchor line. No surge or current was encountered and effort to hold position was minimal. The data appear in Fig. 3 (wet dives).

Breath-hold bradycardia was significantly different from control heart rates at all levels of submersion. There was no significant difference in responses to BH between depths below the surface, but all were significantly attenuated with respect to the surface bradycardia ($P < 0.05$). The degree of bradycardia at the ocean surface was comparable to that measured totally submerged with mask in the laboratory immersion tank in 25 C water, 32% and 34%, respectively. Ocean water temperature was 26 ± 0.4 C from the surface to depths of 18.3 m.

Figure 3 also reflects measurements in a dry recompression chamber to 30.2 m, in which only face immersion, rather than whole-body immersion, was encountered. No significant differences were noted between surface and simulated depths (in the absence of whole-body immersion) under these FI conditions, which approximated water temperatures encountered in the open ocean series (25–26 C).

The attenuation of the BH bradycardia with depth of submersion (Fig. 3) poses some interesting questions. Craig (7) reported no difference in response to depth in an experiment in which a subject submerged to a pool bottom on scuba and there initiated a breath hold. This experiment was at a depth of 1.7 m and water temperature was reported to average approximately 32.5 C. From the data reported here for 35 C water and data previously reported (13) on FI in warm water, it would appear that the thermal effects of warm water on BH response (attenuation) might mask any depth effects under Craig's experimental condition. This would be particularly true during a shallow exposure. Since water temperature was relatively constant in our initial open-ocean experiment, the variable was depth. This variable may be separated into two components under these conditions: *1*) variation in breathing gas partial pressure, and *2*) mechanical effects of immersion at increased hydrostatic pressures.

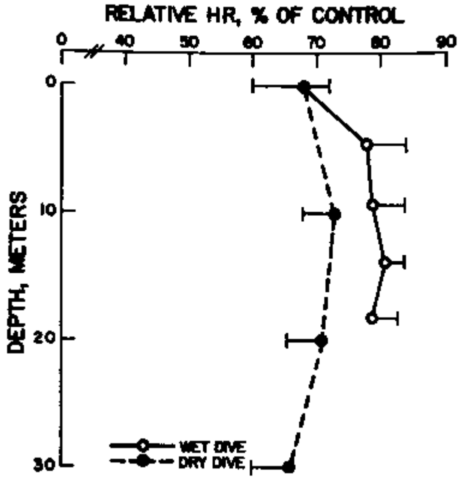

FIG. 3. Relative heart rate responses to 60-sec BH as a function of depth in open ocean (wet dive) and with FI in 25 C water in a recompression chamber (dry dive).

TABLE 2. *Mean maximal heart rate responses to 60-sec BH upon FI in 25 C water as a function of* PI_{O_2}

Condition	% of Control*	PI_{O_2} Equivalent,† mm Hg
1 Ata—air	68 ± 4.4	150
2 Ata—10.3% O_2	63 ± 7.0	150
1 Ata—40% O_2	73 ± 7.0	285
2 Ata—air	68 ± 4.0	310
1 Ata—100% O_2	62 ± 3.1	713
2 Ata—50% O_2	60 ± 6.8	736

* Means ± SE. † Corrected for water vapor pressure.

5) Altered PI_{O_2} *experiments.* Since the depth simulation experiments also necessarily involved PI_{O_2} changes with depth changes, the counter experiment was undertaken; controlled depth with experimentally altered PI_{O_2}, at sea level and at 2 Ata. A surface BH on air was duplicated (in terms of PI_{O_2}) by a 2 Ata BH (after washout for 10 min) on 10.3% O_2 mix. The 40% O_2 surface BH corresponded to the 2 Ata PI_{O_2} of an air BH, and surface BH on 100% O_2 duplicated a PI_{O_2} of 50% O_2 at 2 Ata. The results appear in Table 2. No significant differences are apparent between any of the experimental conditions. However, there is a trend toward attenuation of response in the PI_{O_2} range of approximately 253 mm Hg, disappearing on either side of this partial pressure range.

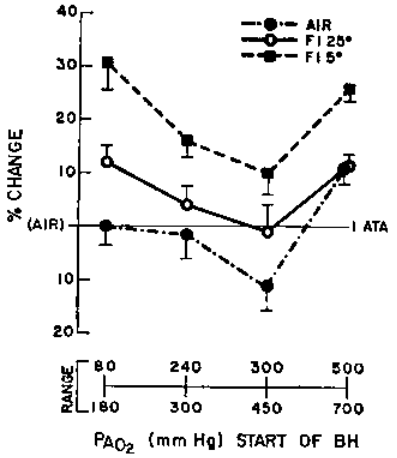

FIG. 4. Relative heart rate responses to BH in air, 25 and 5 C water as a function of $P_{A_{O_2}}$ range. Responses are normalized to BH on air at 1 Ata. Positive numbers indicate relative potentiation of bradycardial response. Negative numbers indicate relative attenuation of response.

There is considerable confusion in the literature as to the effect of $P_{I_{O_2}}$ on BH bradycardia. Daly et al. (9) found that pure O_2 in the lungs slowed the heart by vagal discharge. Raper et al. (20) report no difference from air BH response with 100% O_2 breathing. Kawakami et al. (17) reported FI experiments in which O_2 mixtures were used for BH. In water of 10–17 C, face immersion BH on air caused a decrease in HR from about 78 to about 48 beats/min, or 38%. A companion experiment on BH with 100% O_2 led to face immersion bradycardia from about 86 to about 60 beats/min, or 30%. A gas mix of 10% O_2, however, produced a slowing of only 11%. This group mainly argued against the effect of asphyxic conditions on potentiation of BH bradycardia and did not dwell upon the attenuation of response seen with 100% O_2. Our experiment was designed to explore the possibility of an elevated $P_{I_{O_2}}$ chemocomponent to the bradycardial response. The attenuation of the response on high $P_{I_{O_2}}$ would make a certain teleologic sense if indeed the converse BH bradycardia in man is an "oxygen-conserving" mechanism. As Table 2 indicates, no significant dependence upon $P_{I_{O_2}}$ could be demonstrated. However, the data in Fig. 3 and Table 2 suggest a tendency toward attenuation of BH response at certain levels of $P_{I_{O_2}}$. Elsner's group (11) has reported similar results.

Since alveolar P_{O_2} varies between individuals even at the same $P_{I_{O_2}}$, a comparison between heart rate response to apnea and $P_{A_{O_2}}$ was warranted. Results from the several experiments in which $P_{I_{O_2}}$ was altered, both by artificial gas mix or by elevated pressure, were examined in terms of the measured pre-BH $P_{A_{O_2}}$ vs. apneic heart rate response. These composite data appear in Fig. 4. The relative results are normalized or referenced against control responses to BH on air at 1 Ata without face immersion.

When viewed in terms of $P_{A_{O_2}}$, there does appear to be a significant ($P < 0.05$) attenuation of response in the $P_{A_{O_2}}$ range of 300–450 mm Hg, with an unexplained return to normal response upon further elevation of $P_{A_{O_2}}$. This pattern is qualitatively consistent whether BH is performed with face out or face in water. At these elevated $P_{A_{O_2}}$ levels, the question arises about the potential contribution of elevated P_{CO_2} (via the Haldane effect) to the bradycardia response. In an ancillary experiment in this laboratory, BH in air and with face immersion was performed after rebreathing maneuvers on both air and 100% O_2. Upon initiation of BH, $P_{A_{CO_2}}$ levels were elevated to approximately 44 mm Hg by both rebreathing procedures. On room air, bradycardia was 4% greater with elevated $P_{A_{CO_2}}$; on 100% O_2 ($P_{A_{O_2}}$ = 374 mm Hg), bradycardia was approximately the same. It appears, therefore, that the attenuation seen in Fig. 4 is not related to higher P_{CO_2} levels.

In summary, when combining the BH response with 1 Ata immersion to any degree (face in or whole body), the bradycardia appears to be due predominantly to thermal stimulus. In constant temperature, the attenuation during immersion at depth may be related to elevated $P_{A_{O_2}}$ under those conditions and/or influenced by mechanical stimuli imposed by hydrostatic effects. The nature of the latter is unknown at present. As the data and literature indicate, however, BH bradycardia may be modified by multiple factors, and espousal of hemodynamic or neural mechanisms on an "either-or" basis seems unwarranted.

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STUDIES ON ANIMALS

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Autonomic nervous control of cardiovascular response during diving in the rat

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LIN, Y. C. *Autonomic nervous control of cardiovascular response during diving in the rat.* Am. J. Physiol. 227(3): 601–605. 1974.—The diving response of the unanesthetized rats was produced by submerging the head up to eye level in 30°C water by head-down tilting. The head-down tilting without head immersion served as the predive control. A steady response of blood pressure (BP) and heart rate (HR) to diving was established within 5 s following head immersion. HR was reduced by 64% and BP increased by 15% during diving. Sympathetic tone was reduced by 61% and parasympathetic tone increased 306% from the predive values, whereas intrinsic heart rate was not altered during diving, as deduced from selective autonomic blockade by atropine or propranolol. Cardiac output was reduced by 72% and total peripheral resistance increased fourfold from the predive values. Stroke volume was not changed, stroke work increased by 24%, and left ventricular power decreased by 65% during diving. The unanesthetized rats indicated a diving response qualitatively and quantitatively similar to that of diving mammals rather than nondiving species. It is a potential model for diving studies so far as cardiovascular responses are concerned.

cardiac output; aortic blood pressure; heart rate; total peripheral resistance; cardiac power; apnea

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APPROPRIATE HEMODYNAMIC ADJUSTMENTS enable the diving mammals to effectively channel the limited amount of oxygen preferentially to the heart and the brain (1, 31). The work requirement of the heart is lowered during diving in diving mammals, by instantaneous and intense bradycardia, and reduction in cardiac output, while central systemic arterial blood pressure is maintained at normal level, reflecting severe peripheral vasoconstriction (4, 21). On the contrary, nondiving species (nonnaturally diving species), such as man and dog, develop bradycardia gradually and to a much less extent (4, 11, 15, 30), change in cardiac output is negligible during breath-holding in man (11, 30), and hypertension develops gradually but continuously during the course of breath-holding in man (11) and in the dog (19). The present study is aimed at comparing the diving responses of a nondiving species, the rat, to that of the classical description of a diving mammal and to that of man.

METHODS

Preparation of animal. Male Wistar white rats weighing 350–405 g were surgically prepared, under pentobarbital (30 mg/kg) anesthesia, as follows: an aortic catheter was introduced via the left carotid artery for purposes of recording blood pressure and collection of blood samples for cardiac output determination. A right-atrial catheter was introduced via the right jugular vein for purposes of injection of indicator during cardiac-output determinations. These catheters were threaded subcutaneously, with their exits at the back of the head between the two ears, sutured onto the skin. The animals were allowed to recover for a minimum of 3 days prior to experimentation.

Diving response. The diving response of the unanesthetized rats was produced by submerging the head up to the level of the eyes in 30°C water by head-down tilting. The surgically prepared rat (see above) was confined in a wire-mesh cone for the diving maneuver. The head-down tilting position without head immersion served as predive control.

Systemic arterial blood pressure. This pressure was recorded with a Statham P23Db transducer and a Beckman Dynograph, via the implanted aortic catheter. The transducer was mounted at the level of the heart, and it was arranged so that the level of the transducer changed in accordance with the level of the heart during the diving maneuver. The mean arterial blood pressure was determined planimetrically over a 5-s steady-state response during diving.

Heart rate. Heart rate (HR) was counted from the pressure tracings. It was confirmed on several occasions that the pressure wave can be used to assess the heart rate during the diving response by recording the electrocardiogram and blood pressure simultaneously.

Autonomic control of heart rate. Autonomic control was deduced from selective blockade of sympathetic or parasympathetic nervous activity by treating the rat with propranolol hydrochloride (8 mg/kg) or atropine sulfate (1 mg/kg) according to a procedure described by Lin and Horvath (18). We could have administered these drugs via the implanted right atrial catheter, but we administered them intraperitoneally since a procedure, using intraperitoneal injection, had been established previously regarding dosage and time course of the effectiveness of the drugs (18).

Heart rates obtained at 20 min after the administration of atropine and/or propranolol were used in the above computations since the sympathetic blockade by propranolol was effective for a prolonged time, but the response to atropine began to decline 20 min after injection. The 20-min time interval, therefore, represented the most effective blockade in both divisions of the autonomic nervous system, and because of this the diving experiments were performed 20 min after the injection of blocking drugs.

Reserpine treatment. The importance of peripheral catecholamine stores on the integrity of the cardiovascular response to diving was examined by pretreating the animal with reserpine sulfate (5 mg/kg). The diving response was initiated before treatment and 4 h posttreatment with reserpine.

Cardiac output. Output was calculated from the dilution data using cesium-137 as an indicator, taking advantage of its long half-life (30 yr), gamma-emitting property. The indicator (10 μCi/kg), in a volume of less than 0.5 ml, was injected into the right atrium via the implanted right atrial catheter and serial arterial blood samples were collected via the implanted aortic catheter onto a motorized collecting disk. The circular disk was equipped with 60 depressions located on the outer perimeter and spaced 1 s apart. The motor (Universal timer, Dimco-Gray Company, Dayton, Ohio) rotated at precisely 1.0 rpm. Ten microliters of blood were measured from each collected depression using disposable pipettes (Eppendorf Brinkmann Instruments, Westbury, N. Y.) and the radioactivity of each sample was counted in a Packard gamma counter. The downslope of the dilution curve was reconstructed to exclude the recirculation of the indicator. Both the reconstruction of dilution curve and the calculation of the cardiac output were performed in accordance with the method of Kinsman et al. (16).

Cardiac outputs were determined after heart rate and aortic blood pressure were recorded, once before and once during diving, in the same rat. The blood radioactivity level increased following the first injection of the indicator and this is subtracted from each sample for the second determination. Cardiac outputs, predive and during dive, were determined randomly. When cardiac output during dive was determined first, 0.5 h was allowed for resting immediately after the dive.

RESULTS

Control response to diving. A typical response to diving as initiated by immersion of the head of a rat in 30°C water is shown in *panel A* of Fig. 1. Initiation and termination of a dive are indicated by left and right arrows, respectively. The 1-s mark is indicated between *panels B* and *C*. Arterial blood pressure increased immediately (1–2 s) following head immersion, and a prominent bradycardia developed 1–3 s later. A steady response of arterial blood pressure and heart rate to diving was established within 5 s following the head immersion. Blood pressure and heart rate values were averaged over the 10-s period after the steady response was obtained.

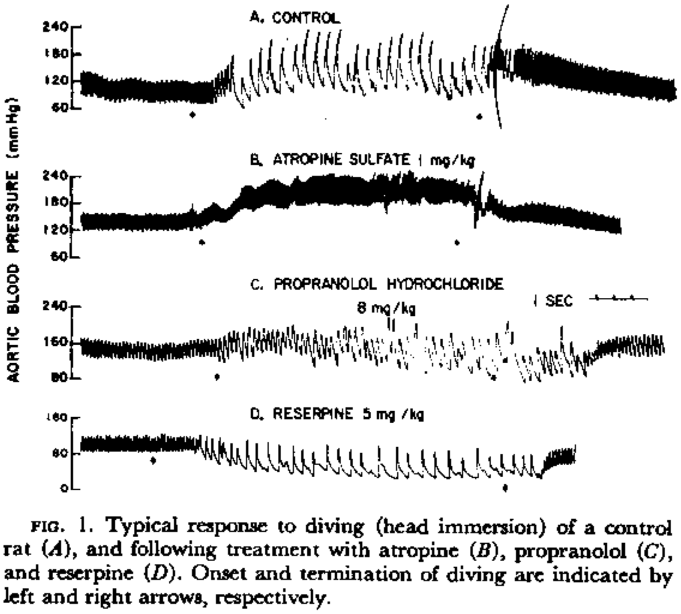

FIG. 1. Typical response to diving (head immersion) of a control rat (*A*), and following treatment with atropine (*B*), propranolol (*C*), and reserpine (*D*). Onset and termination of diving are indicated by left and right arrows, respectively.

TABLE 1. *Heart rate and mean aortic blood pressure response in unanesthetized rats before and during diving*

	Control (Saline)	Atropine, 1 mg/kg	Propranolol, 8 mg/kg	Reserpine, 5 mg/kg
		Heart rate, beats/min		
Predive	399 ± 12	464 ± 8	297 ± 9	209 ± 11
Dive	143 ± 6*	418 ± 17‡	103 ± 23*	133 ± 8*
		MABP, mmHg		
Predive	118 ± 2	123 ± 2	140 ± 9	105 ± 5
Dive	136 ± 5†	168 ± 3*	169 ± 10	77 ± 4†

Values are means ± SE (6 rats in each group). MABP = mean aortic blood pressure. * $P < 0.001$. † $P < 0.01$. ‡ $P < 0.05$.

Heart rate and mean arterial blood pressure responses. These responses to head immersion of the normal (control, saline injected), atropine-, propranolol-, or reserpine-treated rats are presented in Fig. 1 and in Table 1.

The control rats responded to diving by a 64% reduction ($P < 0.001$) in heart rate and 15% increase ($P < 0.01$) in mean arterial blood pressure. Atropine treatment did not abolish the bradycardia response completely, but the magnitude of reduction in heart rate was much less than that of control rats. Heart rate was reduced by 10% ($P < 0.05$) from the predive value. Mean arterial blood pressure increased by 37% ($P < 0.001$) from the predive value. Propranolol treatment decreased the predive heart rate from 399 ± 12 to 297 ± 9 ($P < 0.001$). A 65% reduction ($P < 0.001$) in heart rate from the predive value was observed during diving in the propranolol-treated rats. The magnitude of this reduction was similar to that of the control group. For reasons unclear to us, the arterial blood pressure was more variable following propranolol treatment than that following atropine or reserpine (Table 1). Mean arterial blood pressure rose 21% above the predive value ($0.1 > P > 0.05$). Reserpine treatment depressed predive values of both heart rate and mean arterial blood pressure as compared to the control rats. Diving bradycardia was still present but the magnitude of the bradycardia was reduced when compared to the control group. Hypotension, rather than hypertension, was observed during diving in reserpine-treated rats, signifying failure of peripheral vasoconstriction in the face of cardiac output reduction (Fig. 1).

Autonomic nervous control of heart rate. Autonomic control of heart rate was deduced from selective blockade of sympathetic or parasympathetic nervous activity. The primary results, obtained from Table 1, along with the calculated results are shown in Table 2. The heart rate free of autonomic control—the intrinsic heart rate HR_o—was essentially unchanged (+4%). Sympathetic tone was reduced 61%

TABLE 2. *Autonomic nervous control on heart rate of unanesthetized rats before and during diving*

Conditions	Predive	Dive	% Change
Control	399 ± 12	143 ± 6	−64
Atropine, 1 mg/kg*	464 ± 8	418 ± 17	−10
Propranolol, 8 mg/kg*	297 ± 9	103 ± 23	−65
Intrinsic heart rate, HR_o†	362	378	+4
Sympathetic tone, % HR_o‡	28.2	10.6	−61
Parasympathetic tone, % HR_o‡	17.9	72.6	+306

Values are means or means SE (six rats in each experiment). *The dive was performed 20 min after the administration of drug(s). †Calculated from selective blockade of the divisions of autonomic nervous system by atropine or propranolol. See text. ‡Sympathetic tone and parasympathetic tone represent the percentage of intrinsic heart rate that is controlled by sympathetic activity and by parasympathetic activity, respectively. They are expressed as percentages of the intrinsic heart rate. See text.

TABLE 3. *Hemodynamic changes in unanesthetized rats before and during diving*

	Predive	Dive	*P*
Heart rate, beats/min	401±17	114±16	<0.001
Mean systemic arterial blood pressure, mmHg	111±10	144±10	<0.05
Cardiac output, ml/min	91±10	27±6	<0.01
Stroke volume, ml/beat	0.227±0.040	0.237±0.059	>0.9
Stroke work, g-m/beat	0.34±0.5	0.47±0.13	>0.3
Left ventricular power, g-m/min	137±22	53±12	<0.01
Total peripheral resistance, (dynes-s·cm^{-5})·10^{-5}	0.973±0.137	4.256±1.08	<0.02

Values are means ± SE, six rats in each group.

and parasympathetic tone increased 306 % from the predive values during diving.

Cardiac output. Cardiac output obtained from a separate group of six rats during diving indicated a 70 % reduction ($P < 0.01$) from the predive value (Table 3).

Hemodynamic profile. Hemodynamic changes during diving are given in Table 3. Stroke volume, stroke work, left ventricular power, and total peripheral resistance were calculated from the measured parameters. Stroke volume was not changed ($P > 0.9$), and stroke work was also unchanged ($P > 0.3$); left ventricular power decreased by 61 % ($P < 0.01$), whereas total peripheral resistance increased fourfold ($P < 0.02$).

DISCUSSION

Since the discovery of diving (apneic) bradycardia in the duck just over 100 yr ago (3), a similar response has been demonstrated in many other vertebrates including man (1, 25, 27). Moreover, it has been shown that fish taken out of water (diving in reverse) exhibit profound bradycardia (9, 17), indicating the universality of the bradycardia response to respiratory arrest, differing among species only in the magnitude of the response. It is in general agreement that the final common pathway of the diving-bradycardia response is the vagus nerve in all species thus far reported, as ascertained by vagotomy or by atropine administration. Our observation, a threefold increase in parasympathetic tone (Table 2), is in consonance with this view. The contribution of sympathetic nervous activity to the bradycardia response, however, has not been reported previously. Our results indicate a 61 % reduction in sympathetic tone during diving, in the face of a 306 % increment in parasympathetic tone from predive values, as far as heart rate control is concerned. Thus, the diving bradycardia in the rat is produced by concomitant elevation of parasympathetic tone and reduction in sympathetic tone, with parasympathetic control dominating.

The circulatory response to diving consists of various intensities of bradycardia and peripheral vasoconstriction with or without alterations in the mean arterial blood pressure among mammals with wide ranges of tolerance to respiratory arrest. The rat exhibits greater and faster developing bradycardia than that of man, but they are similar in hypertensive response. The central systemic arterial blood pressure is maintained at the predive level in marine mammals (13, 20, 21), nutria (6), and beaver (12, 25). The diving hypertensive response observed in the unanesthetized rat in our study, on the other hand, is consistent with the observations in man (11), in the dog (4, 19), in the cat (5), and in the sheep (28).

The dosages of atropine and propranolol we used in this study represent our own lengthy trials of lowest dosage for the highest heart rate attainable with atropine sulfate (1 mg/kg) at resting condition and also for the lowest heart rate attainable with propranolol hydrochloride (8 mg/kg). The dosages that we tried are 0.5, 1, 2, and 5 mg/kg, and 1, 2, 4, 8, and 16 mg/kg for atropine and propranolol, respectively. The reasons for expressing autonomic tone in terms of the fractions of intrinsic heart rate rather than the recorded heart rate have been discussed previously (18). Under conditions in which HR and HR_s vary in the same direction, such as during exercise, the use of HR as the denominator will result in underestimation of sympathetic tone, where HR_s is the heart rate augmented over HR_o by sympathetic activity. Referring to Fig. 1 of the paper by Robinson et al. (24) it can be seen that HR_s/HR_o but not HR_s/HR was increased during exercise, due to the fact that HR_s and HR were both increased. HR_s/HR_o rather than HR_s/HR will be the appropriate expression concerning sympathetic activity during exercise. They demonstrated that HR_o remained essentially constant during exercise and during tilting experiments. Obviously, conclusions concerning the autonomic tone are valid, only if the denominator remains unchanged during the experimental procedures. This condition has been met during head immersion, since the HR_0 was essentially unchanged (Table 2).

Whether or not baroreceptor activity contributes to the development of diving bradycardia is an unsettled question. Andersen (1) cautioned against the dismissal of baroreceptors as the cause of diving bradycardia, since the baroreceptors contain a rate-sensitive element which responds to rate of pressure change in addition to the level of blood pressure. Our results favor the dismissal of baroreceptors as a contributing factor in diving bradycardia. Bradycardia was evident in the reserpine-treated rats during diving while aortic pressure was actually falling (Fig. 1*D* and Table 1).

The reduction in cardiac output during diving can be accounted for by the diving bradycardia alone, indicating an unaltered stroke volume. In a recent study by Jones and Holeton (14) in the duck, the stroke volume increased slightly on the average, except in one duck which consistently displayed a reduction in stroke volume during diving. Jones and Holeton reasoned that favorable conditions exist for cardiac pumping during diving since preload (right atrial pressure) is increased, while afterload is unchanged or decreased during diving. But the elevated vagal activity during diving may nullify this favorable condition by suppressing the contractility of the myocardium. It is expected that considerable species variation exists for the stroke output during diving since the anatomical arrangement of the vagal innervation to the ventricles may be predictably variable from species to species. The negative inotropic effect of the elevated vagal activity has been shown in the duck (7), in nutria (6), in the dog during diving as reviewed by Higgins et al. (10), and in the baboon (23) during vagal stimulation.

Circulatory responses to diving have been studied infrequently in the nonnatural diving species because of drastic attenuation of the response with anesthesia (4, 5, 28, 29) and difficulties in obtaining voluntary head immersion in all of these species except man. Nevertheless, diving bradycardia (breath-hold bradycardia, apneic bradycardia) has been reported in nonnaturally diving species, such as the rabbit (8, 28), dog (4, 19, 22), sheep (28), and cat (5). The present study employing unanesthetized rats indicates a diving response qualitatively and quantitatively similar to that of the diving mammals rather than the nondiving species. It is a potential model for diving studies, as far as the cardiovascular responses are concerned, without the technical difficulties and high expenses often encountered in employing the marine mammals in the laboratory.

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CARDIAC OUTPUT AND ITS DISTRIBUTION DURING DIVING IN THE RAT

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ABSTRACT

The diving response was produced by submerging the head of the unanesthetized rat for 60 seconds, while confined in a mesh-wired cone. Heart rate and cardiac output decreased by 73% and 74% from the predive values, respectively, indicating insignificant change in stroke volume. Central systemic arterial blood pressure rose by 22% during diving, while a 4 fold increase in total peripheral resistance was observed. Blood flow to the coronary, cerebral and bronchial circulations remained unchanged while a 95% reduction in the intestine and the spleen, a 97% reduction in the kidney, and greater than 99% reduction in the tail and skin were observed during diving. The blood flow reduction from predive values range from 50% for liver and skeletal muscle, 75% for the adrenals, and 65% for the diaphragm. The redistribution of the drastically reduced cardiac output during head immersion in the rat is similar to that reported for diving mammals. It is suggested that rat may serve as a useful cardiovascular model for further studies of the diving response in mammals.

Key words: Cardiac output, fractional distribution of cardiac output, regional blood flow, redistribution of regional blood flow, diving, apnea, rat.

INTRODUCTION

There is little doubt that intensive peripheral vasoconstriction occurs during apnea in non-naturally diving species (Angell and Daly, 1972; Hong et al., 1971; Lin, 1974), similar to that occurring in diving mammals. More importantly, selective vasoconstriction during apnea results in redistribution of blood flow in favor of the heart and the brain. The significance of this profound redistribution of drastically reduced cardiac output (due to bradycardia) has been discussed extensively, ascribing to it a role of oxygen conservation during diving in the diving species (Anderson, 1966; Anderson, 1967; Elsner et al., 1966; Scholander, 1963; Yonce and Folkow, 1970). While several isolated examples of apneic redistribution of blood flow have been reported (Elsner et al., 1966; Forster and Nyboer, 1955; Gooden, 1971), the objective of the present study is to define quantitatively the redistribution of the drastically reduced cardiac output to various organs in the unanesthetized rat during apnea as induced by head immersion.

METHOD

Preparation of animal: A group of six male Wistar white rats weighing 394 ± 9 grams were surgically prepared under pentobarbital (30 mg/kg) anesthesia, as follows: The aortic catheter was introduced via left carotid artery for the purpose of recording systemic arterial blood pressure and collecting arterial blood samples for cardiac output determination. The right atrial catheter was introduced via right jugular vein for indicator injection. These catheters were threaded subcutaneously, having their exits at the back of the head between the two ears and were sutured onto the skin. The animals were allowed to recover for a minimum of three days prior to experimentation.

Another group of 6 rats weighing 390 ± 5 grams were prepared exactly as above for studying the distribution of cardiac output. Although the procedure for the cardiac output distribution (see below) does not require the arterial catheter, the animals were prepared in the manner described above so that the blood flow could be estimated from the data obtained in these two groups of rats.

Diving response of the unanesthetized rats was produced by submerging the head up to the eye level in 30°C water for 60 seconds by head-down tilting. The surgically prepared rat (see above) was confined in a mesh-wire cone for the diving maneuver. The head-down tilting without head immersion served as the predive control.

Systemic arterial blood pressure was recorded with a Statham P23Db pressure transducer and a Beckman dynograph via the implanted aortic catheter. The mean arterial pressure was determined planimetrically over the entire 60 second diver period. Heart rate was also counted from the pressure trace for the entire dive period.

Cardiac output was calculated from the dilution curves using Cesium-137 as an indicator. The indicator (10 µCi/kg) in a volume of less than 0.5 ml was injected rapidly into the right atrium via the implanted right atrial catheter and serial arterial blood samples were collected via the implanted aortic catheter onto a motorized collecting disk (Figure 1). The circular disk was equipped with 60 depressions located on the outer perimeter and spaced 1 second apart. The motor (Universal time, Dimco-Gray Co., Dayton, Ohio) rotated at precisely 1.0 revolution per minute. Ten microliters of blood were measured from each collecting depression using disposable pipettes (Eppendorf, Brinkmann Instruments, Westbury, N.Y.) and the radioactivity of each sample was measured in a Packard gamma counter. The down slope of the dilution curve was reconstructed to exclude the recirculation of the indicator. Both the reconstruction of dilution curve and the calculation of the cardiac output were performed in accordance with the method of Kinsman et al. (1929).

Cardiac output was determined both before and during dive in the same rat. The sequence was randomized. A half hour was allowed between tests. The heart rate and aortic blood pressure were recorded before the determination of cardiac output. The increased blood radiation level following the first

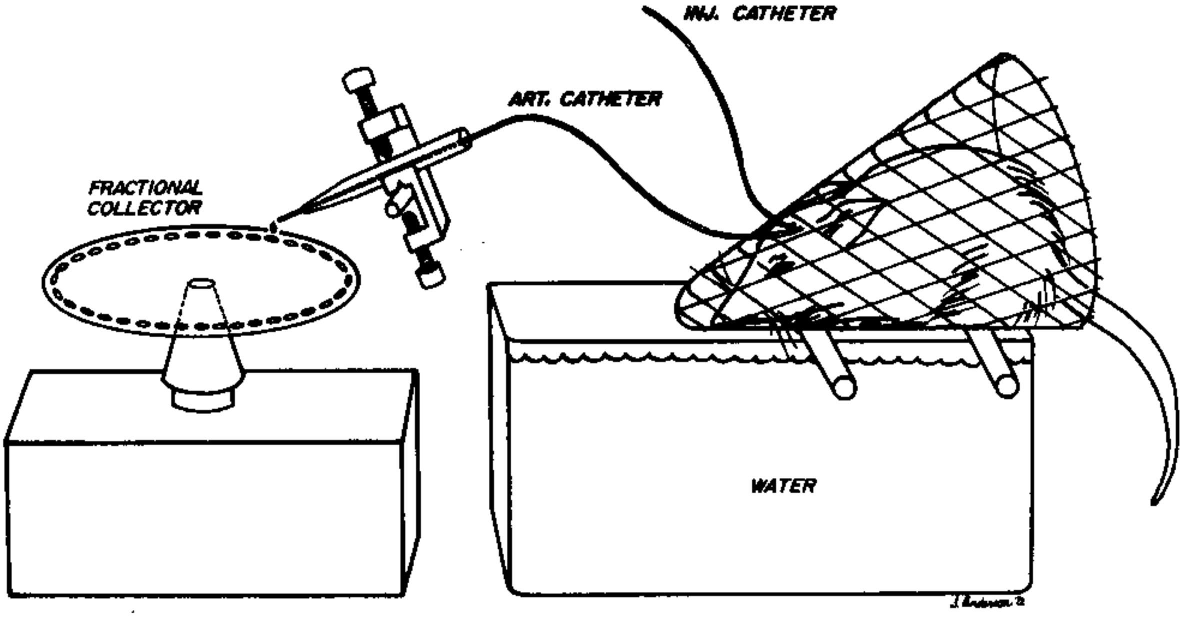

Figure 1. Diving (head-immersion) apparatus and fractional arterial blood collector. Unanesthetized rat was confined in a meshed-wire cone. Head was immersed up to level of eyes by removal of cross bar beneath head. Fractional collector is made of a timer motor and a collector plate equipped with 60 depressions at its periphery. See text for details on arterial blood pressure recording and cardiac output determinations.

indicator injection was subtracted from each sample in the second determination of cardiac output in the same rat. Arterial blood collection began just prior to immersion. These samples were used to establish the background radiation counting. The indicator was injected 5 seconds following head immersion. Blood sample collection continued for 40 to 50 seconds.

Distribution of cardiac output: The fractional distribution of cardiac output was determined by a procedure established by Sapirstein (1958). ^{137}Cs was used as the radioactive indicator. The fractional distribution of cardiac output using ^{137}Cs have been reported in the rat during exercise in warm and cold water by Horvath et al. (1970).

The rats were injected with 1.0 µCi/100 grams of $^{137}CsCl$ dissolved in a saline solution (10 μC_i/ml), via the atrial catheter. The cardiac pumping was terminated 60 seconds later by the injection of 1.0 ml of saturated KCl.

Regional blood flow was calculated by multiplying cardiac output ($\bar{X} \pm X$) and the fractional distribution of ^{137}Cs ($\bar{Y} \pm \sigma Y$) from two different groups of rats, where σX and σY are standard deviations of cardiac output and fractional distribution of ^{137}Cs, respectively. The standard deviation of the product of two independently determined quantities, σXY, was calculated as follows (6):

$$\sigma XY = \left[(\bar{X}^2 + \sigma X^2)\ (\bar{Y}^2 + \sigma Y^2) - (\bar{X}^2)\ (\bar{Y}^2)\right]^{1/2}$$

RESULTS

Diving response of the unanesthetized rat was an immediate and sustained bradycardia, and hypertension (Figure 2). The hemodynamic profile before and during diving is presented in Table 1. Central systemic blood pressure rose by 22% ($p < 0.05$) and the heart rate was reduced by 73% ($p < 0.001$) from the predive values during head immersion.

Measurements	Predive	Dive
Heart rate (bt/min)	411 ± 11	118 ± 14 ***
Cardiac output (ml/min)	95 ± 18	25 ± 5 **
Mean aortic blood pressure (mm Hg)	106 ± 5	129 ± 7 *
Stroke volume (ml/bt)	0.231 ± 0.039	0.212 ± 0.054
Total peripheral resistance (dynes-sec-cm^{-5} .10^{-5})	0.890 ± 0.216	4.118 ± 0.943**
Stroke work (g-m/bt)	0.333	0.372
Cardiac power (g-m/min	136.9	43.9
Mean transit time (sec)	9.2 ± 3.1	23.6 ± 4.6*

Table 1. Hemodynamic changes during diving in the unanesthetized rats
Means and S.E. of 6 rats weighing 394 ± 9 grams
* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$
Stroke work and cardiac power are estimated from the mean values of aortic blood pressure, stroke volume and cardiac output

Cardiac output decreased by 74% ($P < 0.01$) from 95 ml/min to 25 ml/min during head immersion (Table 1). An insignificant change in stroke volume was noted. Figure 3 shows a typical radioisotope dilution curve before and during diving with an identical amount of isotope injected. The mean transit time, from right atrium to aorta, increased from 9.2 seconds to 23.6 seconds during diving. A striking increase in the calculated total peripheral resistance was observed (Table 1).

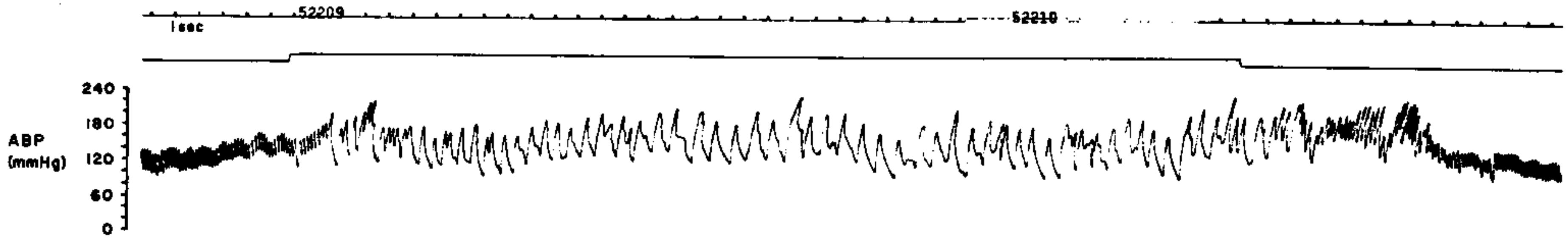

Figure 2. Typical response to diving (head immersion) of rat. Traces from top: 1-s timer, event marker signaling onset and termination of head immersion, and aortic blood pressure.

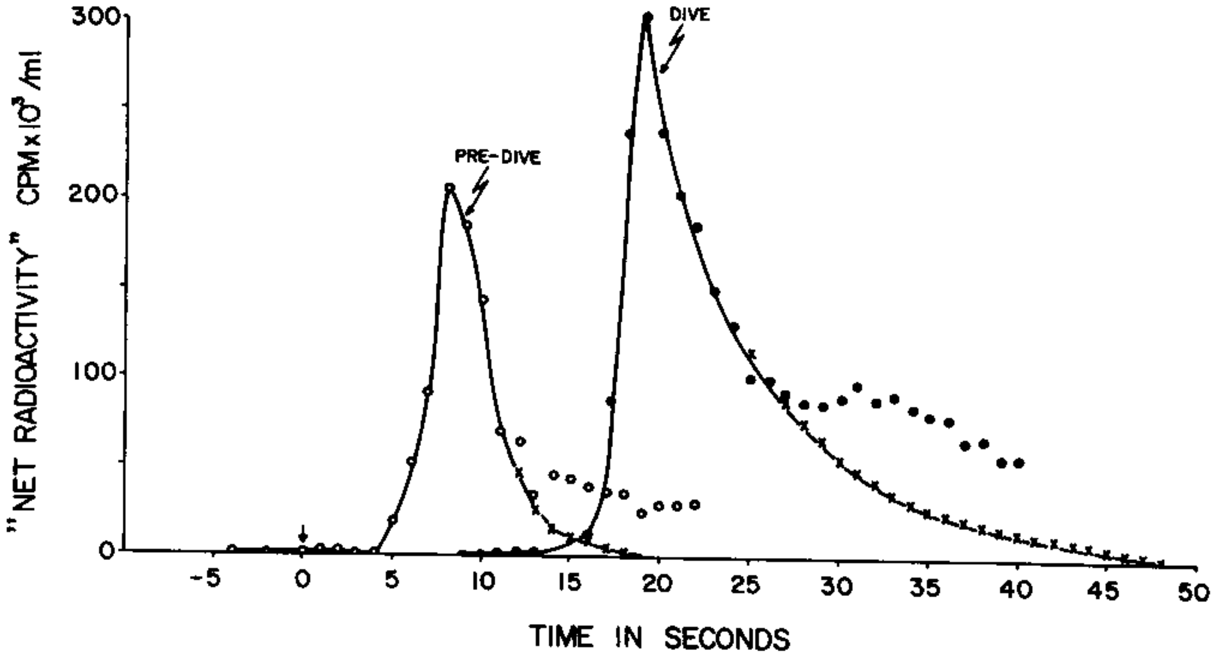

Figure 3. Typical radioisotope dilution curves obtained before and during diving maneuver in same rat. Crosses are extrapolated values. Indicator (same amount in both instances) was injected at time 0. Note increase both in area under dilution curve and in mean transit time of indicator travel from right atrium to aorta.

Fractional distribution of ^{137}Cs and regional blood flow are summarized in Tables 2 and 3, respectively.

During diving, the blood flow to the coronary, the bronchial, and the cerebral circulations were essentially maintained at pre-diving levels (Table 3). This was achieved by obtaining a greater share of the reduced cardiac output. The fractional cardiac output to each gram of the ventricle, the lung, and the brain was increased approximately three-fold (Table 2).

The increased fractional distribution of the cardiac output in the heart, the lung and the brain was at the expense of other tissues, namely the intestine, spleen, kidney, tail and skin. The fractional cardiac output they received ranged from 16% of the pre-dive level for the intestine to 3% of the pre-dive level for the skin. The corresponding values for the spleen, the kidney and the tail were 14, 10, and 4% of the pre-dive values, respectively (Table 2).

Greater than 95% reduction in the blood flow was observed in the intestine and the spleen, 97% reduction in the kidney, and greater than 99% reduction in the tail and skin during diving (Table 3).

Tissue	% Cardiac Output/g Tissue	
	Pre-dive	Dive*
Ventricles	1.55 ± 0.047	3.234 ± 0.263**
Bronchial	1.101 ± 0.057	3.543 ± 0.302**
Brain	0.049 ± 0.004	0.184 ± 0.014**
Intestine	0.658 ± 0.064	0.104 ± 0.031**
Spleen	0.426 ± 0.021	0.062 ± 0.008**
Kidney	2.589 ± 0.169	0.271 ± 0.034**
Tail	0.036 ± 0.007	0.0015 ± 0.0004**
Skin	0.096 ± 0.008	0.0026 ± 0.0009**
Liver	0.574 ± 0.029	1.124 ± 0.084**
Gast. M.	0.240 ± 0.014	0.450 ± 0.049*
Diaphragm	0.628 ± 0.034	0.829 ± 0.181
Adrenals	0.816 ± 0.072	0.788 ± 0.107

Table 2. Fractional distribution of cardiac output before and during diving in the rat

* $P < 0.01$, ** $P < 0.001$

Means and S.E. of 6 rats weighing 390 ± 5 grams

Tissue	Blood Flow (ml/min g)	
	Pre-dive	Dive
Ventricles	1.097 ± 0.219	0.809 ± 0.197
Bronchial	1.046 ± 0.212	0.886 ± 0.217
Brain	0.047 ± 0.010	0.046 ± 0.011
Intestine	0.625 ± 0.236	0.026 ± 0.011*
Spleen	0.406 ± 0.082	0.016 ± 0.004**
Kidney	2.459 ± 0.511	0.068 ± 0.018**
Tail	0.034 ± 0.010	0.0004 ± 0.0001*
Skin	0.091 ± 0.019	0.0007 ± 0.0003**
Liver	0.545 ± 0.111	0.281 ± 0.068
Gast. M.	0.228 ± 0.047	0.112 ± 0.029
Diaphragm	0.597 ± 0.122	0.207 ± 0.069*
Adrenals	0.775 ± 0.169	0.197 ± 0.054*

Table 3. Regional blood flow before and during diving in the rat

* $P < 0.01$, ** $P < 0.001$

Means and S.E. of 6 rats weighing 390 ± 5 grams

The fractional distribution of the cardiac output to the liver, the gastrocnemius muscle, diaphragm and the adrenals was either maintained or even increased during diving. However, in the face of 74% reduction in the cardiac output during diving, the blood flow to these tissues was decreased. The reduction from the pre-dive values ranged from 50% for the liver ($0.1 > p > 0.05$) and the skeletal muscle ($0.1 > p > 0.05$) to 75% for the adrenals ($p < 0.01$). The corresponding reduction in the diaphragm was 65% ($p < 0.01$) from the pre-dive value (Table 3).

DISCUSSION

Intense peripheral vasoconstriction occurs during apnea in the rat just as it occurs in diving mammals and birds, as indicated by a four fold increase in total peripheral resistance. Significant and widespread vasoconstriction as indicated by an increase in arterial blood pressure, has also been reported in several "non-naturally" diving species such as man (Hong et al., 1971; Kawakami et al., 1967; Whayne et al., 1972), dog (Angell and Daly, 1972; Elsner et al., 1966; Mithoefer, 1965), cat (Ferrante and Frankel, 1971), rat (Lin, 1974), and sheep (Tchobroustsky et al., 1969). While general vasoconstriction is valueless to the organism as a whole in regard to oxygen conservation, selective vasoconstriction in skeletal muscle and splanchnic region during apnea effectively shunts the limited amount of oxygen to the heart and the brain. Selective vasoconstriction has been reported in several non-naturally diving mammals; blood flow is reduced during apnea in the calf of man (Elsner et al., 1966), in the abdominal aorta, renal artery, and superior mesenteric artery of the dog (Angell and Daly, 1972; Elsner et al., 1966), in the skinned hind limb of the dog (Angell and Daly, 1972) in the kidney of the rabbit (Forster and Nyboer, 1955), and in the tail of the rat (Gooden, 1971). Our findings are in apparent agreement with these reports in that drastic reduction of blood flow occurred in areas other than the heart, the brain and the bronchial tissue. Quantitatively, the blood flow was reduced by 99% in the tail during head immersion, which agrees with Gooden's (1971) measurements with venous occlusion plethysmography in the tail of the rat. It is of interest to note that the four-fold increase in total peripheral resistance in the present observation is similar in magnitude to that reported by Angell and Daly (1972) in the femoral vascular bed of the dog during stimulation of nasal mucosa with flowing liquid. Furthermore, the 50% reduction in the gastrocnemius muscle blood flow which we observed during head immersion is also similar to that observed by Angell and Daly (1972) in the lower limb muscle (58% reduction) during mucosal stimulation.

Redistribution of blood flow during apnea achieves the purpose of rationing the limited amount of oxygen in accordance with the priority demand by various tissues. Maintenance of blood flow at the predive (pre-apneic level to the heart and the brain alone cannot supply these organs with oxygen at the predive rate, since the arterial P_{O_2} is falling during the course of apnea. We have not determined the blood gases as a function of time during the course of head immersion in this study. However, Hong et al. (1971) reported that the arterial P_{O_2} was reduced precipitously 30 seconds following the onset of breath-holding and face immersion in man, and the arterio-venous P_{O_2} difference narrowed progressively as the breath-hold continued. A similar response may exist in the rat, but with a much quickened pace due to its high metabolic rate. Therefore, it is clear that these vital organs must either reduce their level of activity in the face of diminishing oxygen supply, or obtain their energy anaerobically, or by both.

For the heart, the present measurement indicated a reduction of 68% in the cardiac power output during head immersion, which can be accounted for by the magnitude of apneic bradycardia (71% reduction from the predive value, Table 1). Consequently, the demand for oxygen reduced accordingly. The reduction in the cardiac power output, however, is not secondary to hypoxia, since the bradycardia is attained within three seconds following head immersion and maintained throughout the period of head immersion (Figure 2). The bradycardia response can be abolished by treatment with a parasympatholytic agent - atropine - indicating a vagal mediated reflex in response to cessation of respiration (Lin, 1974). Our recordings showed that the maximum peripheral vasoconstriction is at the onset of apnea prior to the bradycardia and maintained throughout the period of reversible asphyxia (Figure 2). Under such a constant vascular resistance, the maintained aortic blood pressure indicates a non-changing cardiac output during apnea. There is no sign of cardiac failure during the sixty second period of head immersion, suggesting that the energy supplied aerobically and anaerobically sufficed for the level of cardiac activity. It is well recognized that full expression of vasoconstriction is much slower in comparison to vagal mediated responses. But there is no hypotension upon apnea in the face of profound bradycardia and unchanged stroke volume indicating vasoconstriction prior to bradycardia and maintained throughout the entire apneic period. It may be that sympathetic activity have been elevated at the onset of head immersion, ahead of the elevation of vagal activity.

For the brain, the well-known hypoxia intolerant tissue, it is not known whether a similar reduction in activity occurs during apnea. However, it is certain that the cerebral function, as far as the integration of cardiovascular responses to apnea are concerned, is adequate since the full expression of appropriate "diving" response requires the participation of brain center(s) as reasoned from the attenuation or abolishment of the response by general anesthesia (Elsner et al., 1966; Ferrante and Frankel, 1971; Tchobroustsky et al., 1969; Whayne et al., 1971)

The classical bolus injection indicator dilution method for the measurement of blood flow is not applicable during unsteady flow. This is apparently the case if time-averaged sampling is employed for recording the dilution curve (Bassingthwaighte et al., 1970; Cropp and Burton, 1966). The bolus injection method, however, can be accurately applied in the unsteady flow condition if the flow stream can be sampled by obtaining a volume in proportion to the instantaneous flow past the sampling point - a volume-averaged sampling (Bassingthwaighte et al., 1970; Gonzalez-Fernandez, 1962). Our procedure represents a naturally occurring volume-averaged sampling. Whether the sampling thus obtained is exactly proportional to the flow variation, however, has not been proven. Our belief that the method is valid is based on a) a graphically validated exponentiality of the downslope of the dilution curve (Figure 3), and b) the relative stability of the cardiovascular system during the entire diving period, except for the first few seconds (Figure 1). In addition, it has the advantage of the cessation of respiratory movement during diving, since the low frequency effects on the circulation as produced by the respiratory or Mayer waves can produce large errors (Bassingthwaighte et al., 1970; Gabe et al., 1969; Hoffman et al., 1965; Killip, 1962). Contrarily, variations in cardiac frequencies produce much smaller errors (Bassingthwaighte et al., 1970). It is realized that errors inherent to unsteady flow and the indicator dilution method cannot be completely eliminated. But it is safe to conclude from the present study that cardiac output is drastically reduced during dive in the rat.

Sapirstein (1958) has studied and concluded that gamma-emitting Group I alkaline metals (K, Rb, or Cs) would be ideal soluble indicators for estimating tissue blood flows in small mammals. We now know that radioisotope-tagged insoluble microspheres (20 - 50 μ) are optimal since the extraction ratio of 1.0 will unquestionably be attainable, and measurement is not subjected to time limitation following injection, since entrapment in the capillaries will eliminate the problem of washout (Forsyth, 1970; Sasaki and Wagner, 1971). However, it has been proven that complete extraction (extraction ratio = 1.0) can also be achieved in the rat from 5 - 60 seconds following injection by using K^{42} (Sapirstein, 1956) and within a similar period by using Rb^{86} (Goldman, 1966; Sapirstein, 1958). The process is apparently unaffected by deep hibernation and arousal in ground squirrels (Bullard and Funkhouser, 1962), and deep hypothermia (T_r = 9 - 10°C) in the rat (Willard and Horvath, 1961), conditions in which cardiac activity is greatly reduced, or unstable.

It should be noted that the rate of uptake of ^{137}Cs is not uniform in all tissues. It is particularly slow crossing the blood-brain barrier and consequently the blood flow to the brain is underestimated. A similar conclusion is reached by using rubidium or potassium as an indicator (Goldman, 1966; Sapirstein, 1956; Sapirstein, 1958). This condition may render the comparison of blood flow among various tissues on a less solid ground. But, it should have no effect on the conclusion reached by comparing the same tissue under various experimental conditions.

Comparatively, the bradycardial response of the rat as induced by head immersion is strikingly similar, both qualitatively and quantitatively, to the diving response of the marine mammals and diving aves, and both responses are reflex in nature. On the other hand, the slow and continually developing bradycardia observed in man and anesthetized dog during apnea is possibly attributable to a response to hypertension, to hypoxia and hypercapnia, and to mechanical events altering the intrathoracic pressure (Lin et al., 1974; Pauley, 1969). From this point of view, the rat may serve as a useful model in further exploration of cardiovascular physiology during diving.

ACKNOWLEDGMENTS

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Respiratory variation of heart rate in the California sea lion

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LIN, Y.-C., D. T. MATSUURA, AND G. C. WHITTOW. *Respiratory variation of heart rate in the California sea lion.* Am. J. Physiol. 222(2): 260–264. 1972.—The respiratory variation of heart rate was studied in the lightly restrained but unanesthetized California sea lion. The respiratory cycle of the California sea lion consists of a rapid expiration and inspiration, in succession, followed by a long period of apnea until the next expiration. The apnea accounts for 84% of the respiratory cycle. A tachycardia occurred, on the average, 0.25 sec after the onset of inspiration. The heart slowed down on completion of inspiration, and remained at a low level throughout the apneic and expiratory phases of the normal respiratory cycle. The lowest level of heart rate during a normal respiratory cycle was comparable to that of experimental diving. The normal heart rate of the California sea lion is represented by a low level for 84% of the time and for the remaining period is at a high level, approximately twice that during the apneic and expiratory period of the respiratory cycle. Since the lag between the start of inspiration and the increase in heart rate was negligible, it is reasoned that a central control mediated by the vagus nerves, as deduced from atropine experiments, was involved.

respiratory sinus arrhythmia; apnea, apneic bradycardia; vagus activity; atropine treatment; marine mammals

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THERE ARE numerous reports on diving bradycardia in marine mammals (1, 6, 22); increased vagal tone is the accepted mechanism of the bradycardia, although the sympathetic component has not been studied. On the other hand, spontaneous variations of heart rate, with respiration, in the resting animal have been mentioned only casually in a few species of seals and porpoises. In 1935, Irving et al. (13) reported that the respiratory variations in cardiac frequency and peripheral vascular tonic rhythmicity were more obvious in seals than in terrestrial mammals. They noted that cardiac slowing is associated with the respiratory pause. Bartholomew (3) observed in the adult northern elephant seal that the pulse rate during apnea was approximately 15% lower than during eupnea. The arrhythmia was not obvious in the young pups. Similar results were obtained in the Weddell seal (18), while 2- to 4-year-old fur seals showed marked arrhythmia (10). The accelerated heart rate was associated with the period of rapid expiration and inspiration, which was completed in 1 or 2 sec. A similar respiratory-induced variation in the cardiac frequency had also been reported in the Pacific bottlenose dolphin (4, 11). Thus, it appears that a pronounced respiratory sinus arrhythmia is a wide-spread phenomenon among marine mammals. The purpose of this communication is twofold: *a*) to study, in detail, the variations of heart rate in relation to respiratory movements in the California sea lion; and *b*) to study the role of the vagus nerves in the control of heart rate during normal respiratory movements.

METHODS

Three California sea lions were restrained in a specially designed harness and pen (23). Respiratory movements were monitored with needle electrodes, attached subcutaneously at each side of the thoracic region, at the level of posterior end of the foreflippers, via an impedance pneumograph (E & M, Houston). The output of the pneumograph was then recorded on a Beckman type R oscillographic recorder. Utilizing the same electrodes and a ground lead attached to the sacral region of the body, the ECG and instantaneous heart rate were recorded via a ECG coupler and a cardiotachograph (Beckman Electronic Instrumentation, Calif.), respectively, and recorded on the above-mentioned recorder. Thus the respiratory movements, ECG, and heart rate could be displayed simultaneously. The respiratory flow rate was also monitored via a face mask and a pneumotachograph (Statham, Calif.) to determine whether the impedance record was an accurate representation of the respiratory movements. The agreement was excellent (Fig. 1).

To assess the role of the vagus nerves in the regulation of the heart rate, 1.0 mg of atropine sulfate (Lilly, Indianapolis) was administered subcutaneously to each of two sea lions, weighing 40–50 kg. Their ECG and respiratory movements were recorded continuously for more than 1 hr. The effect of atropine on the heart rate was clear and lasted for more than 1 hr in both cases. No ill effect attributable to the administration of atropine was observed during or after the experiment.

RESULTS

Relationship between respiratory cycle and heart rate. A and B in Fig. 1 signify the beginning and end of expiration, and B and C the beginning and the end of inspiration. The respiratory cycle of the California sea lion consists of a rapid expiration and inspiration, in succession, followed by a long postinspiratory apnea until the next expiration. The respiratory frequency is 4–5/min at normal body temperatures. When the body temperature is elevated, the respiratory frequency may increase slightly but the sea lions do not pant under hyperthermic conditions (23). The duration of the

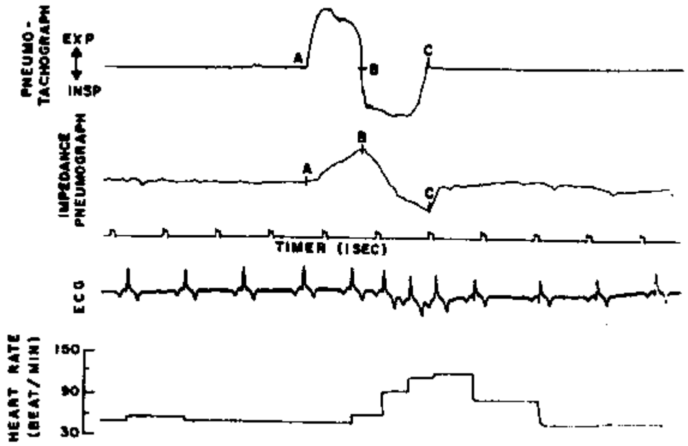

FIG. 1. Effect of respiration on heart rate of a California sea lion. Traces, from top, represent pneumotachogram, impedance pneumogram, 1-sec time signal, ECG, and heart rate, respectively. A-B, and B-C are expiratory and inspiratory phases, respectively.

TABLE 1. *Duration of phases of respiratory cycle in California sea lion*

Animal	N	Expiration	Inspiration	Apnea	Total
Corky	12	1.57±0.23	1.18±0.08	13.02±1.15	15.77±1.12
Corky	9	1.05±0.19	0.81±0.13	12.03±2.90	13.89±2.99
Oily	28	1.37±0.12	0.96±0.04	14.39±1.29	16.73±1.27
Oily	10	1.30±0.02	1.45±0.05	8.64±1.58	11.39±1.60
Zal	24	1.01±0.11	0.97±0.05	11.72±1.79	13.70±1.79
Zal	17	1.31±0.14	1.06±0.10	13.38±1.32	15.75±1.31
Zal	15	1.20±0.07	1.08±0.03	12.23±2.39	14.51±2.41
Avg		1.25	1.07	12.20	14.53

Values are means ± SE seconds. N = number of cycles studied.

entire period of respiratory gas movement is about 2–3 sec, on the average (Table 1). The remaining time of a respiratory cycle is a period of apnea (Table 1). Thus respiration in the California sea lion is characterized by a periodic post-inspiratory breath hold. The apnea accounts for 84 % of the respiratory cycle (Table 1). The inspiratory phase was shorter than the expiratory phase (Table 1). At the end of inspiration (C in Fig. 1), the sea lion expired a very small amount of air before holding its breath.

The heart rate accelerates almost immediately following the beginning of inspiration. The onset of cardioacceleration in relation to the beginning of inspiration was studied in 115 respiratory cycles, from three sea lions on seven separate occasions. The tachycardia started, on the average, 0.25 sec after the onset of inspiration (Table 2). The onset of tachycardia occasionally appeared before or at the beginning of inspiration. The heart rate remained low during the expiratory phase (A–B in Fig. 1). The average heart rate during this period was lower than that of the apneic period (Table 2).

Heart rate during apneic phase. The apneic phase of the respiratory cycle is defined as that from the end of inspiration to the beginning of the next expiration. The heart rate slows down to a low level following completion of inspiration, abruptly in the great majority of cases, occasionally gradually (Fig 1). The changes in heart rate during the course of apnea are qualitatively similar among the sea lions when the successive R-R intervals are plotted against the time course of apnea, taking the end of inspiration as zero time, regardless of the duration of apnea. Figure 2 depicts four apneic cycles with two long apneic periods and two short periods. The similarity in cardiac response is striking. The R-R intervals started to lengthen immediately after the completion of inspiration, returned to end-expiratory levels 2–3 sec later, then settle down to a low level of heart rate.

TABLE 2. *Heart rate during various phases of respiratory cycle in California sea lion*

Animal	N	Inspiration	Apnea	Expiration	Onset of Tachycardia, sec
Corky	12	117.5±1.7	78.5±4.0	63.6±3.6	0.17±0.08
Corky	9	122.3±3.7	96.2±6.1	93.2±6.1	0.15±0.06
Oily	28	94.4±3.8	57.2±1.4	58.4±2.1	0.39±0.03
Oily	10	112.8±1.8	83.4±8.5	76.0±8.1	0.20±0.07
Zal	24	123.8±2.4	103.2±4.5	95.5±4.8	0.23±0.04
Zal	17	112.8±6.1	82.3±3.2	74.0±3.0	0.27±0.06
Zal	15	115.7±3.0	93.1±6.2	75.5±4.4	0.36±0.06
Avg		114.2	84.8	76.6	0.25

Values are means ± SE beats per minute. Heart rate = the number of heart beats in each phase of the respiratory cycle was counted, then converted to minute rate. Onset of tachycardia = the starting point of tachycardia in relation to the beginning of inspiration.

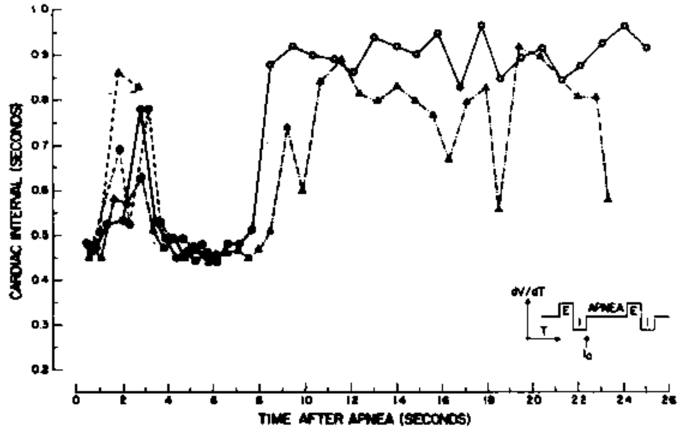

FIG. 2. Cardiac interval during course of apneic phase of a normal respiratory cycle of a male California sea lion (*Corky*). Inset is a schematic drawing of pneumotachogram of a respiratory cycle. E, expiration; I, inspiration; Apnea, from end of inspiration to beginning of next expiration. End of inspiration was taken as zero time. Different symbols represent different respiratory cycles.

During the course of apnea there are occasional bursts of two or three fast heart beats occurring either regularly (Fig. 2) or irregularly. The actual record of the occasional bursts of heart rate can be seen in Fig. 3.

Body temperature and heart rate during apneic phase. The longest R-R interval during the apneic phase tended to be less when the body temperature was elevated, by exposure to air at 35C for 3 hr, than during normothermic conditions (Fig. 4). In Fig. 4, the longest R-R interval during a given apnea is plotted as a function of the total duration of the apneic period. There is a clear trend for the longest R-R interval to be positively correlated with the duration of the apneic period.

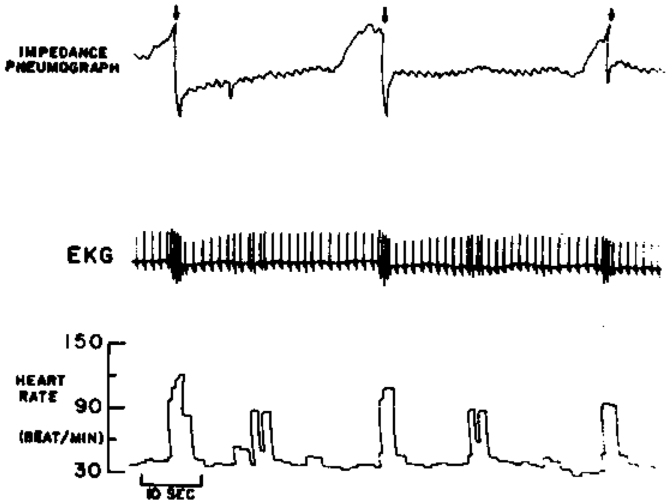

FIG. 3. Occasional bursts of rapid heart rate during apneic phase of respiratory cycle. Arrows on top trace indicate beginning of inspiration.

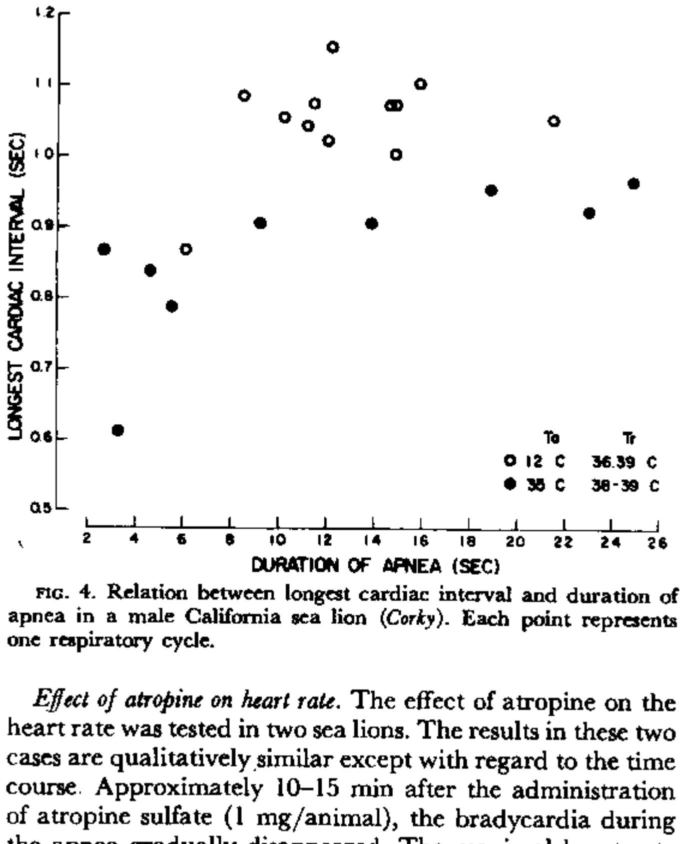

FIG. 4. Relation between longest cardiac interval and duration of apnea in a male California sea lion (*Corky*). Each point represents one respiratory cycle.

Effect of atropine on heart rate. The effect of atropine on the heart rate was tested in two sea lions. The results in these two cases are qualitatively similar except with regard to the time course. Approximately 10–15 min after the administration of atropine sulfate (1 mg/animal), the bradycardia during the apnea gradually disappeared. The maximal heart rate during the inspiratory phase remained unchanged. The maximal effect of atropine was observed 20 min in one animal, *Oily* (Fig. 5), and 40 min in the other animal, *Zal*, after the administration of atropine. At that time, the respiratory effect on the heart rate was negligible, only a very small variation of heart rate with respiratoion culd still be seen (Fig. 5).

DISCUSSION

California sea lions exhibit a clear respiration-related variation in heart rate. The cardiac acceleration was in phase with the inspiratory phase of a normal respiratory cycle. The time lag between the onset of inspiration and the cardioacceleration was less than one-half of the shortest cardiac interval. This suggests that the tachycardia in response to the inflation of the lungs was effected directly by the central nervous system, i. e., the inspiratory activity in the respiratory center inhibits the cardioinhibitory activity of the vagus nerves. The final common pathway is by way of vagus nerves. Alternatively, it is possible that without any changes in the activity of the vagus nerves, the excitation of the inspiratory center reactivates the cardiac accelerator nerves, assuming that the sympathetic activity to the heart was minimal during the apneic and expiratory phase of the cycle. Our results showed that after treatment with atropine, the maximal heart rate during inspiration was not altered, and the variation in the heart rate with respiration diminished in spite of the fact that the inflation and deflation activities of the lung continued, suggesting that the activity of the cardiac sympathetic component remained constant in all phases of the respiratory cycle and that heart rate variations attributable to respiration reflect the marked periodic fluctuation of vagal activity. There was no phase shift with the respiratory frequencies observed (4–8/min). Whether the phase angle of the respiration-heart rate response in sea lions is respiratory frequency dependent, as in man (2, 15), should be investigated. A small residual respiratory effect on the heart rate can still be observed after atropine treatment (Fig. 5). It may be argued that this residual variation in heart rate may represent a respiratory fluctuation in sympathetic activity. However, the magnitude of this effect is small and it can play no significant role in the heart rate response to respiration. It may in fact have been due to the fact that the parasympathetic blockade by atropine, at the dosages used, was not complete. The California sea lion appears to be more sensitive than man and the dog to atropine sulfate (0.020–0.025 mg/kg, sc). In comparison, Jose and Collison (14) used 0.04 mg/kg to block the parasympathetic component of the heart in man and 0.2 mg/kg in the dog (15). It has been established in the harbor seal (20), porpoise (12), and the duck (21), either by vagotomy or by injection of atropine, that the diving bradycardia is the result of an increased vagal tone. It appears that the same mechanism is operating during the naturally occurring periodic apnea. The increased vagal tone continued through the expiratory phase of the cycle. On the other hand, it can be argued that the constant vagal tone is periodically suspended during the inspiratory phase.

The maximal heart rate in the inspiratory phase of the respiratory cycle was relatively constant among animals (Table 2, Figs. 2 and 5). However, the heart rate is more labile during the apneic-expiratory period (Table 2, Figs. 2 and 3). There is a trend for the longest R-R interval to be positively correlated with the duration of apnea. This is to be expected since the cardiac slowing is a continuous process throughout the apneic-expiratory period, unless interrupted by an occasional burst of rapid heart rate. The physiological significance and mechanism of these occasional bursts of rapid heart rates have not been examined. It has been shown in man also that the heart rate oscillates occasionally during the course of a breath hold (9).

The normal heart rate of the California sea lion consists of

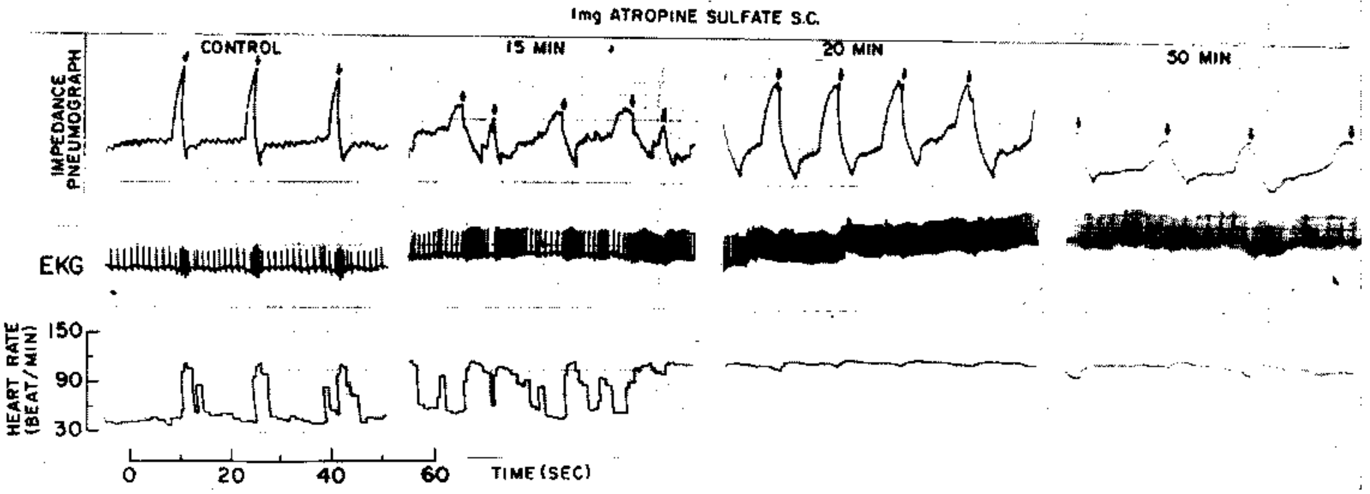

FIG. 5. Effect of atropine on heart rate of a female California sea lion (*Oily*). Traces from top represent impedance pneumogram, ECG, and cardiotachogram, respectively. 15, 20, and 50 min indicate time after administration of atropine. Arrows indicate beginning of inspiration.

a low level for 84 % of the time (Table 1) and for the remaining period it is at a high level, approximately twice that during the apneic and expiratory phases of the respiratory cycle. It is interesting to note that during a voluntary dive the California sea lion and harbor seal (5, 7) achieved no greater bradycardia than that of the normal periodic apnea in air (Fig. 5). Thus it appears that the diving bradycardia in the California sea lion involves only the elimination of the periodic tachycardia during a respiratory cycle. While it is undeniable that diving bradycardia exists and that it is an important adaptation to diving, the magnitude of the bradycardia may have been overestimated in the literature. It is concluded from these experiments that the naturally occurring bradycardia associated with the apnea and expiratory phases of the respiratory cycle was achieved by elimination of periodic tachycardia which was associated with the inspiratory phase of the respiratory cycle. Since the phase shift of the respiratory-heart rate response was negligible, it is argued that a central, direct control was involved.

The postinspiratory apnea is an added advantage to the sea lion. The study of Knelson et al. (17) indicated that introducing an end-inspiratory pause in the dog significantly increased the alveolar gas exchange, without increase in total ventilation. They observed a high correlation between length of end-inspiratory pause and fractional increase in calculated alveolar gas exchange. During the course of evolution the sea lion seems to have chosen precisely this mechanism. The gradual return of thoracic impedance to the pre-expiratory level (Fig. 1) may represent a gradual change in the thoracic configuration as a result of redistribution of air inside the lung. The redistribution of the gas inside the lung may allow the animal to take full advantage of the post-inspiratory pause. It's also possible that it resulted from a drastic reduction in the gas exchange ratio during apnea (8, 19).

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OXYGEN CONSUMPTION AND CONSERVATION DURING APNEA IN MAN AND IN THE ANESTHETIZED DOG

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ABSTRACT

Apnea was initiated by clamping off the tracheal tube in 6 anesthetized dogs and voluntarily in 4 men with or without face immersion. Bradycardia, reduction in cardiac output and peripheral vasoconstriction developed gradually throughout the entire apneic period. O_2 consumption during apnea was measured by monitoring the rate of O_2 removal from the lung (V_{LO_2}), from the arterial blood ($\dot{V}_{aO_2}$), and from the venous blood ($\dot{V}_{vO_2}$). Total O_2 consumption ($\dot{V}_{TO_2}$) was calculated by summing ($\dot{V}_{LO_2} - \dot{V}_{aO_2}$), $\dot{V}_{aO_2}$ and $\dot{V}_{vO_2} \cdot \dot{V}_{LO_2}$, estimated by the product of arterio-venous O_2 difference and cardiac output, decreased continuously during apnea. At the end of 80 seconds the rate of O_2 removal from the lung was one-fourth of the pre-apneic rate, 5.29 ± 0.51 ml-min^{-1}kg^{-1}. O_2 disappearance from the blood was estimated by measuring arterial and venous O_2 content at 20 second intervals and by assuming a constant blood volume throughout the apneic period of 86 ml/kg, 25% of which is in the arterial tree. During the last 20 seconds of an 80 second apnea, $\dot{V}_{vO_2}$ represented 69% of $\dot{V}_{TO_2}$. Identical blood gas and pH time courses were found in the human during 3 minute apnea with or without face immersion. Blood levels of 2,3-DPG were not reduced during apnea in the dog and in man, playing no role in altering the oxygen affinity of erythrocytes. It is concluded from the present study that an oxygen conservation mechanism, if existent, was not operative during an 80 second apnea in the anesthetized dog and in a 3 minute apnea in man.

Key words: Apnea, oxygen consumption, oxygen conservation, blood gas, 2,3-DPG.

INTRODUCTION

Cessation of respiratory movement either at inspiratory or expiratory position induces bradycardia and peripheral vasoconstriction. These responses have been reported in wide varieties of species, including man, varying only in the quantitative but not in the qualitative aspect of the response. These responses are appropriate in rationing the limited amount of oxygen available to the heart and the central nervous system during diving, or apnea in general, and hence, have been described as oxygen conservation mechanisms (Scholander, 1940; Scholander, 1963; Elsner, et al., 1966). The existence of an oxygen conservation mechanism in marine mammals is convincing, although the evidence for it is quite circumstantial. The exchangeable oxygen stores of a harbor seal weighing approximately 20 kg was determined by Packer, et al., (1969) to be 800 ml. Since the maximal diving time for this species is 20 minutes, and its predive oxygen consumption was approximately 200 ml/min, a drastic reduction in oxygen consumption occurred during diving. Scholander (1940) measured and showed a reduced oxygen repayment following a dive, also implying the existence of an oxygen conservation mechanism.

The existence of a similar mechanism in man during voluntary apnea has been proposed (Elsner, et al., 1971; Moore, et al., 1973), but has also been questioned (Hong, et al., 1971). The purpose of the present study is to examine the existence of an oxygen conservation mechanism by monitoring oxygen disappearance from the lung, from arterial blood, and from venous blood during apnea in the anesthetized dog, who, like man, is a non-diving terrestrial mammal whose response to apnea is quantitatively similar to that of a conscious man during voluntary apnea (Hong, et al., 1971; Mithoefer, 1965).

A reduction in blood organic phosphate, especially that of 2,3-diphosphoglycerate (2,3-DPG), is known to increase the affinity of erythrocytes for oxygen in man and in other species (Benesch, et al., 1967, Benesch, et al., 1968), resulting in greater retention of oxygen in the blood at any given Pa_{O2}. If a reduction in the 2,3-DPG level occurs in acute hypoxia, in contrast to chronic hypoxia which increases 2,3-DPG level and lowers P_{50} (Brewer and Eaton, 1971), this could be considered a mechanism for conserving O_2 during apnea. Hence, it is of interest to examine whether acute hypoxia and hypercapnia of short duration reduces blood 2,3-DPG level and hence elevates P_{50}, and, by inference, demonstrates the existence of an oxygen conservation mechanism.

METHODS

Dog studies

Six male mongrel dogs weighing 14 to 20 kg were anesthetized with an initial dose of 30 mg/kg sodium pentobarbital (Abbott Laboratories, North Chicago, Illinois), and were supplemented as required. The dogs were intubated with cuff-tracheal tubes. Catheters were introduced into the right ventricle or right atrium for indicator (cardiogreen) injection, aortic arch for pressure recording, descending aorta for blood samples (for indicator density, or for blood gas measurements), via right jugular vein, left carotid artery, and femoral artery, respectively. Standard lead II ECG was monitored continuously.

Apnea was induced by clamping the tracheal tube at the end of inspiration either with a piece of dry gauze (dry apnea) or ice-cold gauze (wet apnea) covering the nostrils, for a total of 80 seconds. The sequence of these experiments was randomized.

Blood gas and pH. One ml each of mixed venous blood and arterial blood of the dog was obtained via the right heart and aorta, respectively, at the onset of apnea, at the end of 20 seconds, 40 seconds, 60 seconds, and 80 seconds, following the onset of apnea. The blood was analyzed for PO_2, PCO_2, and pH on a blood gas analyzer (Radiometer, Copenhagen), and for hemoglobin concentration using a standard cyanmethemoglobin method. Oxygen content was calculated by using Severinghaus blood gas calculator, Type BGC-1 (Severinghaus, 1966; Radiometer, Copenhagen), using the values above.

Blood 2,3-DPG. Arterial and mixed venous blood of the dog and arterial blood of man were obtained and analyzed for 2,3-diphosphoglycerate by the method of Lowry et al. (1964) as modified by Keitt (1966), and expressed in micromoles per gram of hemoglobin (μM/g.Hb).

Cardiovascular parameters. Heart rate was obtained by counting the total number of R-waves from the ECG in each successive 20 second interval during apnea and converted to minute rate. Although a cardiotachometer was available and was routinely utilized (Figure 1), it was used only for observing the trend of response during apnea. Cardiac output was determined prior to and 40 seconds following the onset of apnea with an indicator dilution method (Cardiogreen, Hynson, Westcott, and Dunning, Inc., Baltimore, Maryland). Heart rate during cardiac output determination for calculating stroke volume was obtained by averaging over the period of the primary dye-dilution curve. Central arterial pressure was measured continuously at the descending aorta via a catheter (P.E. 160, Clay Adams), pressure transducer (P23Pb, Statham), and an oscillographic recorder (Beckman, Type R Dynograph). Mean arterial pressure was determined by averaging over each successive 20 second period during apnea.

Oxygen consumption. The amount of oxygen consumed for each successive 20 second period during apnea was estimated by summing the oxygen disappearance from the lung, from arterial blood, and from venous blood. All values were converted to minute rate and per kg of body weight.

A. Oxygen contributed by arterial blood ($\dot{V}_{aO_2}$): Oxygen disappearing from the arterial blood in each 20 second period during apnea was calculated according to:

$$\dot{V}_{aO_2} = (60/20)\ (V_a)\ (C_{aO_2},\ t_1 - C_{aO_2},\ t_2) \qquad \text{(Equation 1)}$$

Where, 60/20 is the conversion factor from 20 second values to 1 minute values, C_{aO_2}, t_1 and C_{aO_2}, t_2 are arterial O_2 contents 20 seconds apart, i.e. $t_2 = t_1 +$ 20 seconds, and V_a is arterial blood volume which was not measured but assumed as 25% of the total blood volume of 86.2 ml/kg of body weight (Hoff, et al., 1966; Green, 1950).

B. Oxygen contributed by venous blood ($\dot{V}_{vO_2}$): The oxygen contributed by venous blood during each 20 second period was calculated similarly as in A:

$$\dot{V}_{vO_2} = (60/20)(V_v)\ (C_{\bar{v}O_2},\ t_1 - C_{\bar{v}O_2},\ t_2) \qquad \text{(Equation 2)}$$

C. Oxygen contributed by the lungs (V_{LO_2}): Oxygen disappearance from the lungs was computed at 0, 20, 40, 60, and 80 seconds of apnea, according to Fick's principle:

$$\dot{V}_{LO_2} = \dot{Q}\ (C_{aO_2} - C_{\bar{v}O_2}) \qquad \text{(Equation 3)}$$

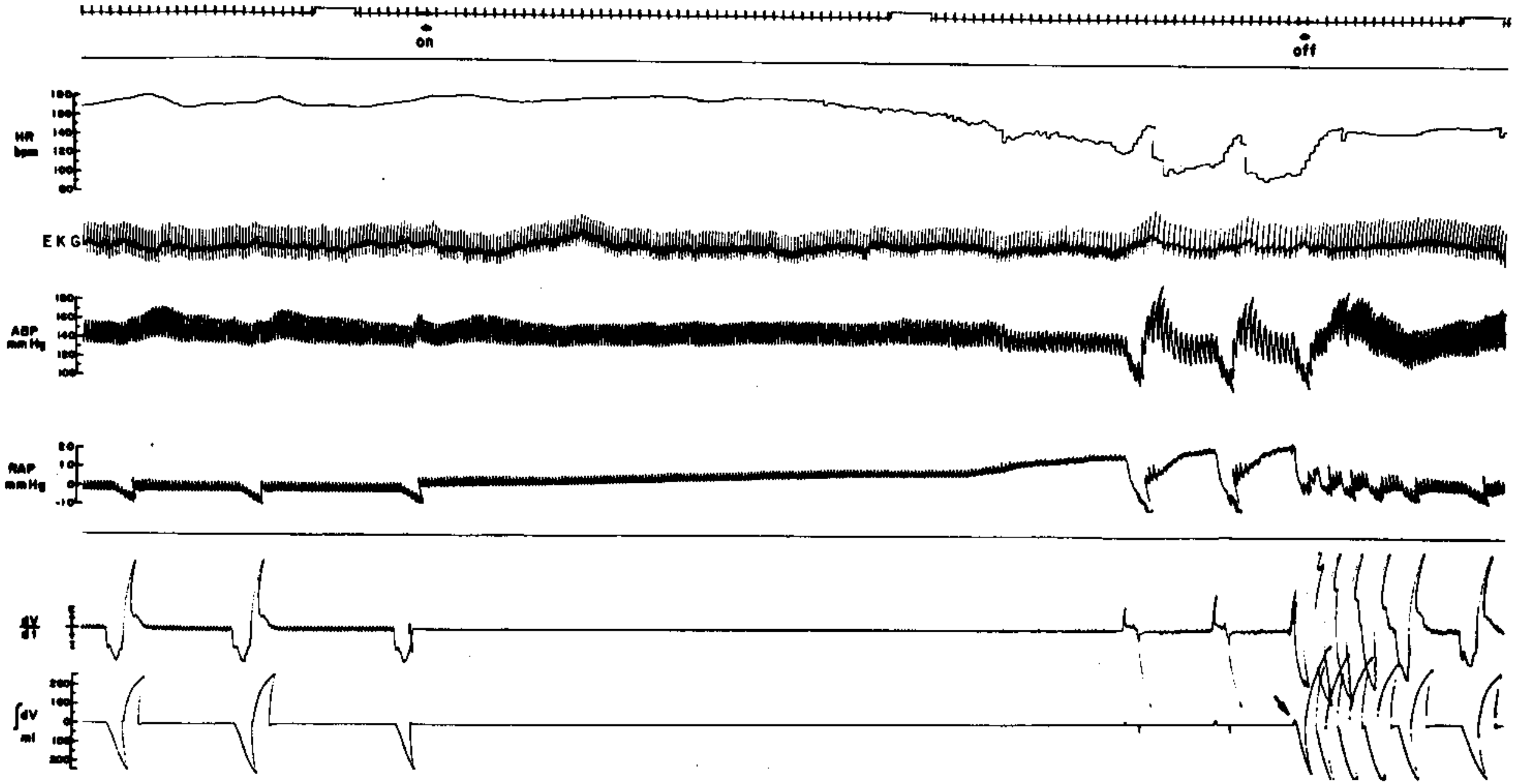

Figure 1. Typical responses of the anesthetized dog to apnea. The record was obtained from a male dog weighing 15 kg anesthetized with 30 mg/kg sodium pentobarbital. The traces from the top are: one-second timer and event marker for onset (on) and release (off) of tracheal clamping; heart rate (HR) in beats per minute: aortic blood pressure (ABP) in mmHg; right atrial pressure (RAP) in mmHg: pneumotachograph showing inspiration (I) and expiration (E); and tidal volume (∫dV) obtained from integration of pneumotachographic signals (dV/dt), in ml (ATPS). The apnea was initiated at the end of inspiration. Small expired volume (arrow on the bottom trace) following release of tracheal clamping indicating near exhaustion of alveolar oxygen.

Where $\dot{Q}$ is cardiac output and C_{aO_2} and $C_{\bar{v}O_2}$ are arterial and mixed venous oxygen contents, respectively.

D. Total oxygen consumption ($\dot{V}_{TO_2}$) was calculated as:

$$V_{TO_2} = (\dot{V}_{LO_2} - \dot{V}_{aO_2}) + \dot{V}_{vO_2} + \dot{V}_{aO_2} \qquad \text{(Equation 4)}$$

or $$\dot{V}_{TO_2} = \dot{Q}\ (C_{aO_2} - C_{\bar{v}O_2}) + \dot{V}_{vO_2} \qquad \text{(Equation 5)}$$

$\dot{V}_{aO_2}$ was deduced from $\dot{V}_{LO_2}$, since the loss from the lungs was gained by the arterial blood.

Human studies

Four male volunteers of apparent good health who were able to breath-hold for 3 minutes with full lung volume of air without prior hyperventilation served as subjects. Arterial blood samples were obtained for blood gas, pH, and 2,3-DPG measurements via an arterial catheter needle (B-D Co., Rutherford, New Jersey) situated in the brachial artery.

Apnea. Human volunteers were instructed to inhale maximally and hold their breath with (wet) or without (dry) face immersion in water (25°C), following a deep expiration without prior hyperventilation.

Heart rate. Heart rate was obtained by counting the total number of R-waves from standard lead II ECG in each successive 10 second interval during dry and wet apneas.

2,3-DPG. Face immersion is known to accentuate cardiovascular responses during apnea (Brick, 1966; Kawakami et al., 1967; Song et al., 1969). Thus, 2,3-DPG samples were obtained during wet apnea only in the human studies, and was analyzed as in dog blood.

P_{aO_2}, P_{aCO_2} and pH. Arterial blood samples from human subjects were withdrawn at 30 second intervals beginning at 1 minute of apnea for 3 minutes. The samples were stored in a packed ice-box and were analyzed for P_{O_2}, P_{CO_2} and pH at 37°C within 60 minutes by using Radiometer electrode units.

RESULTS

Cardiovascular responses to apnea in the dog

A typical response to clamping off the trachea at the end of inspiration is shown in Figure 1. Bradycardia was not evident until 40 seconds of apnea. Bradycardia became prominent during a rapid rise in arterial pressure which was preceded by a pressure decrease. Aortic blood pressure (ABP) and right atrial pressure (RAP) became cyclic with a frequency of approximately 6 per minute and with an excursion amplitude of approximately 90 mmHg for ABP and 30 mmHg for RAP. The mean arterial pressure, however, rose slowly, on the order of 10 mmHg/min. RAP rose from -2 to 22 mmHg at the end of apnea. Cardiac index, in this experiment, decreased from 139 ml · min^{-1} · kg^{-1} before apnea to 98 ml · min^{-1} · kg^{-1} during the period of 50 to 80 seconds following the onset of apnea. Inspiratory and expiratory volumes were recorded in this experiment on the bottom most trace. Apnea began with inspiratory volume of 250 ml. Drastic reduction in lung volume occurred as evident from the small volume of first expiration following the release of tracheal clamp. Results obtained from 6 dogs are summarized in Table 1. There was no quantitative difference in response during dry apnea and wet apnea. Hence, data were pooled for subsequent analyses. Heart rate, and cardiac index decreased 25% and 24%, respectively, from the pre-apneic values of 174 ± 11 bt.min^{-1} and 109 ± 4 ml-min^{-1}kg^{-1}, respectively. Stroke index was not different for all conditions. Mean arterial blood pressure and calculated total peripheral resistance rose 8% and 43%, respectively, from pre-apneic levels of 131 ± 6 mmHg and 72 ± 5 mmHg-kg-ml^{-1}-sec^{-1}, respectively (Table 1).

The pre-apneic heart rate in 4 human subjects averaged 84 beats per minute. Heart rate increased by 20 beats per minute as the subjects expired maximally and then inspired fully in preparation for breath holding. With the cessation of respiratory movement, heart rate rapidly decreased to 17% below the pre-apneic level within 50 seconds of apnea, prior to leveling off with minor oscillations. Heart rate responses from the onset to 50 seconds of apnea were identical in dry and wet apneas, after which heart rate during wet apnea was consistently lower than during dry apnea throughout the entire remaining period of 3 minute apnea (Figure 4).

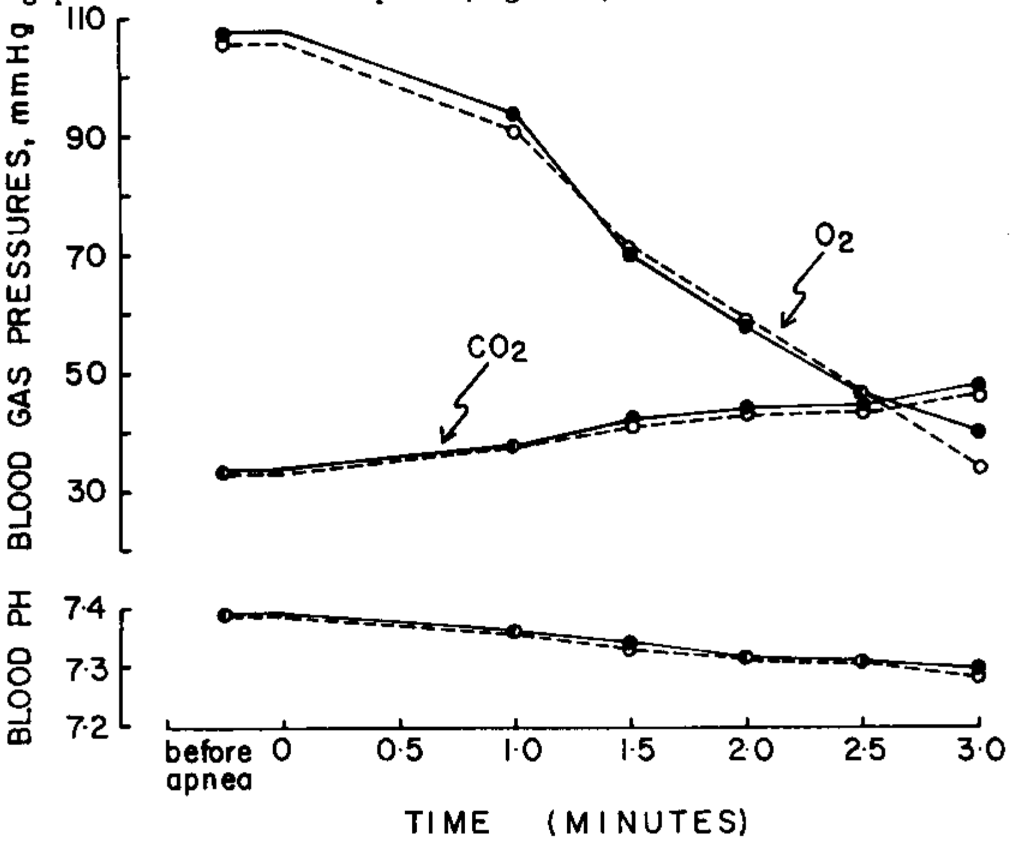

Figure 4. Arterial P_{O_2}, P_{CO_2} and pH during 3 minutes of apnea with (o) and without (•) face immersion in 25°C water. Each point represents the average of 8 experiments in 4 subjects.

Measurements	A Before apnea	B Dry apnea	C Wet apnea	D B + C
Heart rate, min^{-1}	174 ± 11	135 ± 7	125 ± 8	130 ± 5
Cardiac output, ml-min^{-1}	1935 ± 185	1533 ± 143	1402 ± 161	1468 ± 105
Cardiac index, $\text{ml-min}^{-1}\text{-kg}^{-1}$	109 ± 4	87 ± 4	79 ± 6	83 ± 4
Stroke index, $\text{ml-bt}^{-1}\text{-kg}^{-1}$	0.640 ± 0.048	0.622 ± 0.047	0.637 ± 0.072	0.649 ± 0.041
Mean aortic blood pressure mmHg	131 ± 6	144 ± 7	140 ± 7	142 ± 5
T.P.R., $\text{mmHg-kg-ml}^{-1}\text{-sec}^{-1}$	72 ± 5	99 ± 7	106 ± 13	103 ± 8
2,3-DPG, µM/g Hb**	9.80 ± 0.95	9.83 ± 0.79	9.47 ± 0.77	9.65 ± 0.54

Table 1. Cardiovascular responses during apnea* in anesthetized dogs.

Values are means ± S.E. for 6 dogs with mean body weight of 17.60 ± 1.27 Kg (range, 13.77-20.80Kg). Paired tests showed that no statistical difference existed between B & C; and that there were statistical differences ($P < 0.05$) between A & B, between A & C, and between A & D, for all measurements listed above, except that of stroke index which is not significantly different ($P > 0.05$) between conditions.

* Dry and wet apnea values were obtained during 50-80 second period following the onset of apnea.
**2,3-DPG: 2,3-diphosphoglycerate in µg per gram hemoglobin was obtained in only 4 dogs.

Oxygen contents in the dog

C_{aO_2} declined linearly at a rate of 6.15 vol% per minute, where $C_{\bar{v}O_2}$ was unchanged for the first 20 seconds, and then declined at a slower rate, 4.57 vol% per minute, during apnea. Arterial-venous oxygen content difference (C_{aO_2} - $C_{\bar{v}O_2}$) decreased from 4.84 ± 0.46 vol% before apnea to 1.86 ± 0.50 vol% at the end of 80 seconds of apnea (Figure 2).

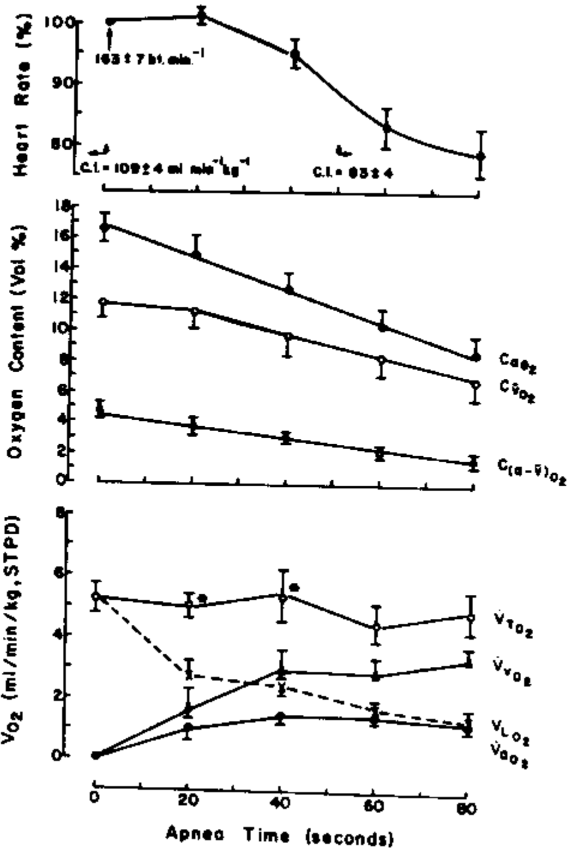

Figure 2. Heart rate, blood oxygen content and oxygen consumption ($\dot{V}_{O_2}$) of six anesthetized dogs during an 80 second apnea. C_{aO_2}, $C_{\bar{v}O_2}$, and $C_{(a-v)O_2}$ are arterial, mixed venous and arterial minus mixed venous oxygen contents, respectively. $\dot{V}_{aO_2}$, $\dot{V}_{vO_2}$, and $\dot{V}_{LO_2}$ are rates of oxygen disappearance from arterial blood from venous blood, and from the lung, respectively. $\dot{V}_{TO_2}$ equals the sum of $\dot{V}_{aO_2}$, $\dot{V}_{vO_2}$, and $\dot{V}_{LO_2}$ - $\dot{V}_{aO_2}$ (see METHOD). C.I. is cardiac index in $ml \cdot min^{-1} \cdot Kg^{-1}$. * The $\dot{V}_{LO_2}$ at these two periods was calculated by assuming the same cardiac output as that measured during the 50 - 80 second period. Therefore, the actual values of $\dot{V}_{TO_2}$ should have been greater than indicated (see DISCUSSION).

Oxygen consumption in the dog

Summation of the rate of oxygen disappearance from arterial blood, from venous blood, and from the lung (see METHOD) indicated that the oxygen consumption was essentially unchanged throughout the entire apneic period. $\dot{V}_{TO_2}$ was 5.29 ± 0.5 ml/min/kg at the onset of apnea, and was 4.59 ± 0.62 and 4.91 ± 0.67 at the end of 60 seconds and 80 seconds apnea, respectively. $\dot{V}_{LO_2}$ at the end of 20 seconds and 40 seconds apnea was calculated by assuming the same cardiac output as that measured at 50-80 second periods of apnea. The V_{TO_2} at 20 seconds and 40 seconds of apnea were 5.01 ± 0.42 and 5.44 ± 0.88 ml/min/kg, respectively. $\dot{V}_{TO_2}$ values were not statistically different for all periods. $\dot{V}_{aO_2}$ leveled off after 40 second apnea at 1.5 ml/min/kg. $\dot{V}_{vO_2}$ also leveled off after 40 second apnea, and at the end of 80 second apnea venous blood contributed to the $\dot{V}_{TO_2}$ at a rate of 3.38 ± 0.46 ml/min/kg accounting for 69% of the V_{TO_2} (Figure 2).

Blood 2,3-DPG

Arterial and venous 2,3-DPG levels of the dog were not significantly different before apnea and during wet or dry apnea. The arterial and venous pooled data are shown in Table 1. Blood 2,3-DPG levels in human during apnea were also statistically unchanged during 1, 2 and 3 minutes of wet apnea from the values at the onset of apnea and 5 minutes following termination of apnea (Table 2).

P_{O_2}, P_{CO_2}, and pH

Arterial and mixed venous blood gases and pH analyzed in 6 dogs are as follows: arterial P_{O_2} decreased from 67.7 ± 4.9 mmHg just before apnea to 31.9 mmHg at the end of the 80 second apnea. Mixed venous P_{O_2} decreased from 38.3 ± 3.6 mmHg to 26.9 ± 3.6 mmHg at the end of 80 second apnea. Arterial and mixed venous P_{CO_2} increased from 28.8 ± 1.5 and 36.7 ± 1.7 mmHg, respectively at pre-apnea to 44.2 ± 1.7 and 42.8 ± 1.5 mmHg at the end of the 80 second apnea. Blood pH decreased from 7.29 ± 0.022 to 7.23 ± 0.016, and from 7.27 ± 0.014 to 7.23 ± 0.018 for arterial and mixed venous blood, respectively (Figure 3).

Arterial P_{O_2}, P_{CO_2}, and pH values from 4 human subjects are summarized in Figure 4. Pa_{O_2} decreased continuously from 107 mmHg before apnea to close to 40 mmHg at the end of the 3 minute apnea, while P_{aCO_2} increased from 33 to 47 mmHg, and pH decreased from 7.38 to 7.30. No statistical difference exists between dry and wet apneas.

DISCUSSION

Results of the present study indicated that the oxygen consumption was unaltered throughout the entire period of 80 second apnea in the anesthetized dog. This observation is consistent with our earlier study of man that oxygen consumption was essentially unchanged during a simple voluntary apnea

Subjects	Duration of apneic face immersion (min)				Recovery (5 min)
	0	1	2	3	
EH	2.86	2.53	2.71	2.68	2.68
	2.20	3.10	3.08	----	3.19
BR	3.46	3.55	4.09	3.78	4.21
	4.25	4.06	3.99	4.83	3.61
DL	2.40	2.41	2.52	----	2.36
SH	6.11	7.00	6.88	6.49	6.75
Mean	3.546	3.775	3.878	4.445	3.80
SE	0.596	0.693	0.656	0.810	0.648

Table 2. 2,3-DPG concentration (mM/l RBC) during apneic face immersion.

*All mean values were statistically insignificant, according to paired t tests.

in the face of 15% reduction in heart rate and an apparent peripheral vasoconstriction (Hong, et al., 1971), and in agreement with the reports by Raper, et al. (1967) and Heinstad and Wheeler (1970). Oxygen conservation mechanisms in man may be demonstrable under certain unusual conditions (Elsner, et al., 1971; and Moore, et al., 1973), but it is also conceded that if the O_2 conservation mechanism exists in man, it is a very weak one. The present results failed to demonstrate an oxygen conservation mechanism during apnea. In fact, there was elevation in oxygen consumption during the 20 second and 40 second period of apnea. The oxygen disappearance from the lung was calculated by assuming the same cardiac output as that during the 50 to 80 second period of apnea. The cardiac output values should have been greater than that assumed for these 2 periods since heart rate values were higher at 20 and 40 second period of apnea than that during the 50 to 80 second period of apnea, where stroke index remained unchanged (Figure 2 and Table 1). If one accepts the extrapolation that the cardiac index remained unchanged throughout the entire apneic period and cardiac output is proportional to the heart rate, then the V_{TO_2} should have been 5.90 and 6.12 ml/min/

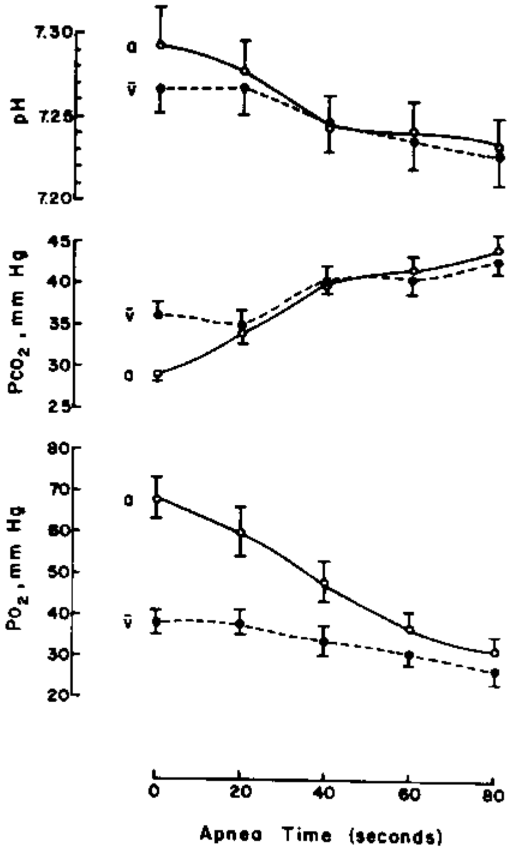

Figure 3. Arterial (a) and mixed venous (v̄) pH, P_{O_2}, and P_{CO_2} of six anesthetized dogs during an 80 second apnea.

kg, at the end of 20 seconds and 40 seconds of apnea, which is 12% and 16% higher than the pre-apneic value of 5.29 ± 0.51 ml/min/kg. Considering the overall accuracy of the determinations all parameters needed for the calculation, these figures may not significantly differ from the pre-apneic level. The important point is that they are increased rather than decreased from the pre-apneic level, making the existence of an oxygen conservation mechanism doubtful.

We have failed to demonstrate any alteration in 2,3-DPG levels during an 80 second apnea in the anesthetized dog and during the course of 3 minute apnea in man, despite the presence of bradycardia and vasoconstriction during the course of apnea. The result is not surprising since the time constant for 2,3-DPG metabolism was reported to be 6 hours (Brewer and Eaton, 1971), but it has also been reported that erythrocyte 2,3-DPG level can be altered within a transit time from arterial side to the venous side as deduced from a demonstrable arterial-to-venous concentration difference (Hamasaki, et al., 1971). We were unable to confirm their results in the present study in the dog and in man during face immersion, and, by inference, it was not altered in dry apnea as well, since the cardiovascular responses during dry apnea were much less than wet apnea (Brick, 1966; Elsner, et al., 1963; Kawakami, et al., 1967; and Song, et al., 1969).

Several assumptions were made for the calculation of $\dot{V}_{TO_2}$. They are:

a) Constant blood volume throughout the entire apneic period at 86.2 ml/kg. $\dot{V}_{TO_2}$ will be less than indicated if a smaller blood volume was assumed. In using an average standard error of 0.5 ml/min/kg in the determination of $\dot{V}_{TO_2}$, one has to assume a blood volume at 60% of the level assumed in the previous calculation in order to show a statistical difference from 5.29 ml/min/kg at the pre-apneic level which is unreasonably small, provided that the distribution of arterial and venous blood remained at the ratio of 1 to 3 (see METHOD). The larger the blood volume one assumes, the greater the calculated $\dot{V}_{TO_2}$ will be during apnea, due to the proportionally greater contribution of oxygen from the blood. The assumption of constant blood volume throughout the entire apneic period may be justified since the total blood withdrawn for gas, pH, Hb, and 2,3-DPG measurements was less than 30 ml for the entire experiment in a given dog. Blood loss in association with cardiac output measurements was negligible since the withdrawn blood was reinfused back into the animal.

b) Since the central blood volume and intrathoracic pressure tend to increase during apnea (Hong, et al., 1971; Lin, et al., 1974 and present study, Figure 1), the venous compartment may increase as a result. Hence, a distribution of 20% arterial and 80% venous was also tried in calculating the arterial and venous oxygen contribution during apnea. In such calculations, all $\dot{V}_{TO_2}$ values subsequent to the onset of apnea were greater than 5.29 ml/min/kg at the pre-apneic level. In view of the low compliance of the arterial tree (Guyton, et al., 1956; Shoukas and Sugawa, 1973), and the stability of arterial compliance under baroreceptor control (Shoukas and Sugawa, 1973), it is generally agreed that arteries and capillaries consist of much less than 25% of the circulating blood volume at any given time. Therefore, the presently reported $\dot{V}_{TO_2}$ probably represents an underestimation of the actual $\dot{V}_{TO_2}$, due to an underestimation of the venous compartment which becomes the major contributor of oxygen during apnea (Figure 2).

Arterial and mixed venous blood pH and P_{CO_2} become indistinguishable between 40 and 80 seconds of apnea. The difference between arterial and venous P_{O_2} narrows as apnea progresses. These observations are qualitatively similar in man and in the anesthetized dog, as shown by the present study, and in agreement with our previous study (Hong, et al., 1971). The low P_{aO_2} at the onset of apnea may be a result of depressed ventilation due to pentobarbital anesthesia.

Bradycardia and selective vasoconstriction can be considered a universal response to cessation of respiration, varying only in degree in a wide variety of species, including terrestrial non-diving mammals. Bradycardia occurs in voluntary apnea, in involuntary apnea, and even in spontaneous apnea during a normal respiratory cycle (Lin, et al., 1971). The functional significance of bradycardia during a dive in the marine mammals (Elsner, et al., 1966) and in the rat (Lin, 1974; Lin and Baker, 1974) is clear and undoubtedly plays a role in O_2 conservation, but it is ambiguous in non-diving mammals, such as the dog and man. Although the mechanism of apneic bradycardia was not the objective of the present study, it is of interest to note that the bradycardia developed slowly in the anesthetized dog, but it occurred prior to apparent arterial blood pressure changes which became conspicuous during the later part of apnea (Figure 1). The heart rate rise and fall in phase with aortic pressure changes is undoubtedly a vagus nerve mediated baroreceptor reflex, but bradycardia prior to these changes is still unclear. The cyclic nature of ABP and RAP as well as pneumotachographic evidence (Figure 1) indicates respiratory effort against a closed trachea, and confirms our previous observation in man whose intrathoracic pressure rose steadily during the early part of apnea but also becomes cyclic during the later part of apnea (Lin, et al., 1974).

The lack of an oxygen conservation mechanism during apnea in the present study is shown by: 1) constant oxygen consumption throughout an 80 second apnea 2) constant blood 2,3-DPG levels in the anesthetized dog, and 3) unchanging blood 2,3-DPG levels and identical time courses for blood gas pressures during wet and dry apnea in man, despite the presence of bradycardia and vasoconstriction during apnea.

ACKNOWLEDGMENTS

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chapter 2

THERMOREGULATION and EXERCISE IN WATER and/or HYPERBARIC ENVIRONMENT

Chapter 2 asks the fundamental and pragmatic question, What is the limiting factor in the performance of useful work in the wet hyperbaric environment? To relatively shallow (i.e., 600 FSW) depths, inert gas narcosis, oxygen toxicity, and decompression sickness can be avoided by judicious application of available technology. A major limitation is simply the physiologic impact of cold stress, due to the high thermal conductivity of water and/or a high density inert gas environment.

This area has been a major endeavor of our research. We have been interested primarily in (i) quantification of heat losses under practical conditions, including the routes of loss, (ii) recovery from exposure to thermal stress, (iii) predictive characteristics for heat loss on an individual basis for given environmental conditions.

In the process of this monitoring, certain techniques and measurements were developed and are represented by two of the papers (Kanwisher et al. and Morlock and Dressendorfer).

As in chapter 1, adaptive trends were investigated by measurements on divers vs. non-divers (Hanna and Hong, Hong, and Lally et al.) and insights into these adaptations were gained by investigation of the responses of marine mammals to variations in the ambient thermal environment.

STUDIES ON MAN

Recovery Parameters Following Underwater Exercise

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CRITTENDEN, G., J. F. MORLOCK and T. O. MOORE. *Recovery parameters following underwater exercise.* Aerospace Med. 45-(11):1255-1260, 1974.

Five male subjects were monitored for energy cost and recovery profiles after mild (433 ± 13 kg·m/min) work in air and immersed at water temperatures of 26° and 20°C. Recovery measurements included heart rates, rectal temperatures, and oxygen consumptions. Heart rates were not different during work under all conditions, but post-exercise recovery to resting rate was faster after exposure to 20°C water. Rectal temperatures dropped faster during working immersions than during immersions without work, and continued to fall during recovery, with the greatest rate of fall following 20°C water exposure. Oxygen uptakes were greater in water than air, the highest cost being in 20°C water, although the oxygen cost of the work per se was not different. The results suggest that thermal recovery is retarded after work in colder water and the energy cost may constitute a significant percentage of daily caloric intake even for mild exposures.

A BROAD literature exists detailing the energy cost of underwater work at a variety of depths and water temperatures (4,6,12,17). Yet, for the compressed-air SCUBA diver, periods of immersion are limited by such factors as cold, limited air supply, and tissue accumulation of nitrogen. As a result, the diver must undergo repetitive exposures of relatively short duration. Thus it is of importance to know the course of recovery after immersion in order to provide adequate time and conditions to maximize work productivity. There are little data in this area, although it is now empirically known that thermal recovery is as necessary a factor in underwater work efficiency as coping with physical fatigue (2,3,19). The present investigation was carried out to study the interactions between thermal, cardiovascular, and respiratory recovery after underwater work at differing water temperatures as compared to recovery in air at 1 ATA for the same workloads.

MATERIALS AND METHODS

Five adult males served as subjects. Selected characteristics of the subjects appear in Table I. All were familiar with experimental procedure, the use of SCUBA, and all but one (R.S.) had previous experience with the underwater ergometer described in an earlier publication (12). Average work loads were 433 ± 13 (S.E.) kg.m/min, approximately the intensity of effort exerted by self-paced divers (17).

Heart rates were recorded continuously on a polygraph from two Beckman electrodes on the chest and a ground electrode on the back. To record respiration rates, a thermister was sealed in the mouthpiece, and changes in temperature during inspiration and expiration were recorded on the polygraph. Rectal temperatures were taken from a thermister inserted 10 cm into the rectum and were read every 1 to 2 min on a telethermometer (Yellow Spring, etc.) to within 0.1°C. Gas collections were taken with Douglas bags. Volumes were measured on a dry gas meter and samples of the collected air were analyzed for oxygen and carbon dioxide on a Scholander micro-gas analyzer. Duplicate gas collections were taken, and the measurements were averaged for the resting, submerged resting, and exercise conditions.

The ergometer was placed in an immersion tank 5 × 5 × 3 ft which allowed the subject to be completely immersed. During the immersion experiments, the subject breathed from standard sport SCUBA gear. Air was supplied from a 72-ft^3 tank by a double-hose, double-stage demand regulator (U.S. Divers, Santa Ana, Ca). For the experiments in air, the subjects used the same SCUBA mouthpiece but breathed room air.

The water temperature was measured by telethermometer and was regulated to within ±0.5°C by the addition of hot water or ice. Two one-third horsepower electric pumps ran continuously to stir the water while the subject was immersed. Air temperatures averaged 27.0°C, and water temperatures were 26°C and 20°C.

During the rest period prior to immersion, and the recovery period following, the subjects wore swimming trunks, slippers, and a terry cloth robe. In the water, they wore only swimming trunks, rubber slippers, a weight belt, and a standard sport diving mask.

The subjects came into the laboratory after a light breakfast and were requested not to exert themselves prior to the experiment. In the laboratory, each subject sat quietly for 30 min to insure resting conditions and, during the last 5 min, two 2-min gas collections were taken. The subject then climbed into the tank and sat for a 10-min equilibration period, the purpose being to allow dissipation of heat stores and the onset of protective

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TABLE I. PHYSICAL CHARACTERISTICS OF THE SUBJECTS.

Subject	Age	Height (cm)	Weight (kg)	Surface Area (m^2)	Average Skinfold (mm)	% Body Fat*	$\dot{V}_{O_2}$ max** (l/min)	ml/kg)
T.M.	38	186.7	75.5	2.00	17.7	17.4	2.85	38.0
R.S.	29	172.7	64.0	1.76	12.8	11.4	2.63	41.0
J.M.	25	184.2	69.2	1.90	10.0	10.7	3.58	52.8
B.R.	29	169.4	67.0	1.76	20.5	21.2	2.37	35.3
R.D.	29	178.6	71.0	1.88	8.8	7.8	4.46	62.5
$\bar{X}$	30	178.3	69.3	1.86	13.9	13.7	3.18	45.9
S.E.	2.1	3.3	1.9	0.05	2.2	2.4	0.37	5.1

* (Sloan and Weir, 1970) (15)

** Treadmill method (9)

thermoregulatory responses. During the last 2.5 min of this period, two 1-min gas collections were taken (submerged values). After the 10-min submersion period, the subject began pedaling for 7 min, and two 1-min gas collections were taken during the last 2.5 min of this period and averaged for the exercise condition. Seven minutes were sufficient to achieve a constant heart rate and minute ventilation at this work load. In the control immersions with no work (still immersion), the procedure was identical except that the subject remained resting in the water during the entire 17 min.

Immediately after the exercise or the 17-min still immersion, the subject climbed out of the tank, put on the terry cloth robe, and sat down. Thermal, cardiovascular, and respiratory recovery was followed for 30 min. One-minute gas collections were made after 2 min (R_1), 4 min (R_2), 6 min (R_3), and 8 min (R_4) of recovery, and 2-min collections were taken after 18 min (R_5) and 28 min (R_6) of recovery. Respiration rates were recorded for the first 9 min and during the last two gas collections. Rectal temperature was taken every other minute. Statistical probabilities were calculated by paired sample analysis.

RESULTS

Rectal Temperature: The mean changes in rectal temperature during immersion and recovery are shown in Table II. There were no changes in rectal temperature during or following work in air.

During 17 min of still immersion in 26°C water, the rectal temperature dropped 0.14°C and during 17 min of immersion with work the drop increased to 0.30°C. The drop was greater for 20°C water both during the still immersion (0.26°C) and the immersion with work (0.64°C.).

Rectal temperature continued to drop during the 30-min recovery period. The drop was 0.4°C following the 26°C still immersion and was not different after immersion with work. Following still immersion in 20°C water, the rectal temperature dropped 0.7°C and a similar change was seen after immersion with work.

The total drop from the beginning of the immersion to the end of the recovery period was 0.54°C for the still immersion in 26°C water and 0.96°C for the 20°C still immersion. The difference between the total drop in rectal temperature at the two water temperatures was significant both in work and still immersion ($p < 0.05$).

Heart Rate: Table III gives the mean values of the heart rates recorded during the experiments.

The average resting heart rate in air was 70 beats/min and, during the first 10 min of immersion, it remained approximately the same in the 26°C water and rose slightly in the 20°C water. The rise during exercise to 127 beats/min was the same for air, 26°C water, and 20°C water.

TABLE II. CHANGES IN RECTAL TEMPERATURE.

Water Temperature	Still Immersion	Immersion with work
I. Mean changes in rectal temperature from rest to the end of 17 min of immersion (°C ± S.E.).		
26°C	−0.14 ± 0.07	−0.30 ± 0.14
20°C	−0.26 ± 0.05	−0.64 ± 0.12
II. Mean changes in rectal temperature from the cessation of exercise to the end of the 30-min recovery period.		
26°C	−0.40 ± 0.07	−0.20 ± 0.13
20°C	−0.70 ± 0.14	−0.63 ± 0.05
III. Total mean changes in rectal temperature from rest to the end of the 30-min recovery period.		
26°C	−0.54 ± 0.14	−0.50 ± 0.20
20°C	−0.96 ± 0.16	−1.27 ± 0.10

TABLE III. HEART RATES (beats/min ± S.E.)
DURING AND FOLLOWING EXERCISE IN AIR, 26°C WATER, AND 20°C WATER.

Environment	Rest	Submersion rest 0-10′	Submersion exercise 10-17′	R_1 2-3′	R_2 4-5′	R_3 6-7′	R_4 8-9′	R_5 18-20′	R_6 28-30′
Air	70 ± 6		124 ± 11	82 ± 11	79 ± 10	77 ± 9	75 ± 8	68 ± 8	67 ± 7
26°C water	66 ± 6	67 ± 7	129 ± 12*	81 ± 14	73 ± 12	72 ± 11	72 ± 11	65 ± 10	67 ± 9
20°C water	70 ± 7	74 ± 6	127 ± 11	83 ± 15	73 ± 13	72 ± 11	71 ± 11	65 ± 1	63 ± 8
DURING AND FOLLOWING STILL IMMERSION IN 26°C WATER AND 20°C WATER									
26°C water	68 ± 6	67 ± 8	65 ± 8	60 ± 7	60 ± 7	59 ± 6	59 ± 7	56 ± 7	56 ± 8
20°C water	69 ± 7	74 ± 7	78 ± 8	63 ± 3	62 ± 5	60 ± 7	62 ± 6	59 ± 5	58 ± 6

TABLE IV. OXYGEN CONSUMPTION (liters/min, STPD ± S.E.)
DURING AND FOLLOWING EXERCISE IN AIR, 26°C WATER, AND 20°C WATER.

Environment	Rest	Submersion rest	Submersion exercise	Recovery R_1	R_2	R_3	R_4	R_5	R_6
Air	.290		1.347	.580	.438	.353	.329	.272	.273
	±.007		± .083	±.071	±.038	±.026	±.024	±.016	±.014
26°C water	.289	.374	1.570	.619	.452	.332	.303	.286	.273
	±.023	±.005	± .102	±.071	±.072	±.026	±.028	±020	±.014
20°C water	.288	.651	1.675	.653	.484	.373	.359	.417	.399
	±.016	±.076	± .142	±.073	±.069	±.022	±.013	±.003	±.063
DURING AND FOLLOWING STILL IMMERSION IN 26°C WATER AND 20°C WATER									
26°C water	.284	.374	.409	.511	.377	.284	.299	.267	.279
	±.010	±.033	± .048	±.034	±.049	±.027	±.015	±.033	±.036
20°C water	.291	.424	.761	.730	.446	.374	.345	.313	.348
	±.021	±.081	± .096	±.028	±.029	±.031	±.035	±.045	±.060

After the cessation of work, the heart rate fell in an exponential fashion, with time, to or below the resting values by minute 19 of recovery (R_5). The heart rates were not different after the three exercise conditions during the third minute of recovery (R_1). The overall recoveries of heart rate after exercise in 26°C and 20°C water were similar but faster than recovery after exercise in air. The mean values of the recovery heart rate suggest that there is a tendency for heart rate recovery to be progressively faster with progressively colder exercise conditions.

Oxygen Consumption: The oxygen consumption (Table IV) during work in air was statistically lower than during work in 26°C water and 20°C water ($p < 0.05$), and the oxygen consumption for work in 26°C water was slightly lower than in 20°C water. The oxygen consumption due to work, as estimated by subtracting the oxygen uptake during the period of rest immersion preceeding the exercise, was not different for any of the exercise conditions (Table V).

The recovery patterns after immersions with exercise and still immersions are different. Rather than dropping off in an exponential curve, as after the immersions with work, the oxygen consumption during the third minute of recovery (R_1) was almost the same as the oxygen consumption during the immersion in 20°C water, and after the still immersion in 26°C water the oxygen consumption during R_1 was actually higher than during the immersion.

TABLE V. OXYGEN CONSUMPTION ATTRIBUTABLE TO WORK (liters/min, STPD ± S.E.).

Exercise Environment	Oxygen Consumption
Air	1.051 ± .089
26°C water	1.158 ± .130
20°C water	1.024 ± .136

The oxygen consumption during the last two recovery periods (R_5 and R_6) showed a progressive rise that tended to be greater after immersion with work than after still immersion and was more apparent after the immersions in 20°C water.

DISCUSSION

Body Temperature: Exercise in air increases heat production and raises body temperature to a level directly related to the work load. It would be expected that exercise would at least decrease the rate at which body temperature falls in cold water. Keatinge found that for a 20-min immersion with exercise heat balance could be maintained in water temperature above 24°C, but in temperatures below 24°C, heat loss was accelerated beyond metabolic heat production (10). The data in the present experiment, however, show that during immersion with work the rectal temperature fall was twice the drop seen during still immersion at both 26°C and 20°C. The conditions for this experiment were slightly different from those of Keatinge's. The work load was greater, and there was a 10-min rest immersion (before the work) not included in Keatinge's protocol.

Keatinge suggested that increased work loads would be associated with little increase in muscle blood flow and that the vasodilatory effect of exercise reaches its maximum at relatively low work loads (11). He found that men working as hard as possible in cold water suffered rather smaller mean falls in core temperature than when they worked at 350 kg·m/min at the same water temperature. The increment in the intensity of work therefore increased heat production more than heat loss, which is dependent on muscle blood flow.

The 10-min rest in the tank depleted the subject's heat stores to some extent as the periphery became vasoconstricted and was allowed to cool. The decrease in the core temperature may be a result of rewarming cool tissues that would not have cooled had the exercise started immediately on immersion as well as a result of the decreased tissue insulation concomitant with exercise.

The heat transfer coefficient of still water is 38 kcal/m^2-h-°C (1). Insufficient stirring of the water during rest would result in an insulation by stagnant water next to the skin and a significant decrease in body heat loss. Even in still water, shivering will increase the heat transfer coefficient from 38 to 53 kcal/m^2-h-°C (1). All the subjects shivered in the 20°C water, and most in the 26°C water. The combination of shivering and stirring probably was quite effective in preventing very much insulation by stagnant water.

The values for whole body conductance could not be calculated since the subjects were not at thermal equilibrium, but the increase in conductance during exercise increases the heat loss by an amount proportional to the temperature gradient between the core temperature and the water temperature (11). The mean decrease in core temperature after work was about twice as great as after rest in water of 26°C. After exposure to 20°C water, the decrease was about 2.5 times as large. Robinson (13) found an increase in tissue heat conductance during exercise in air of about the same magnitude.

During recovery, the data suggest that core temperature continued to drop, probably as a result of the influx of cold blood from the periphery rather than continued heat loss to the environment. Though there was no statistical difference between the drop in rectal temperature after still immersion and immersion with work it appears that, after the 26°C immersion, thermal recovery was facilitated by work during the immersion. The muscles involved in the exercise would not be as cold after work and so would not contribute as much to the thermal debt. Perhaps the thermal debt incurred is not affected by exercise during immersions of short duration. There is a certain amount of heat (heat store) that can be gained or lost from the body with no effect on the core temperature and results from thermal gradients from the core to the periphery. The greater fall recorded in rectal temperature during work may simply be a result of the accelerated circulation and decreased vasoconstriction causing the core and periphery to be closer to equilibrium.

Heart Rate: The heart rates during work were essentially the same in all three conditions of the experiment in spite of the variations in oxygen consumption. This was also true during rest immersion control conditions. The heart rates remained the same while oxygen consumption increased. The greater oxygen pulse in cold water could be attributed to one or a combination of three factors: a) an increase in the oxygen carrying capacity of the blood; b) increased cardiac output as a result of an increased stroke volume; and c) an increased oxygen extraction by the tissues.

Arterial blood is almost completely saturated under normal working conditions, and there is little increase in the partial pressure of oxygen of the inspired air from the slight increase in pressure from the shallow immersion. The oxygen-carrying capacity of the plasma increases with decreasing temperatures, and the affinity of hemoglobin for oxygen also increases as it is cooled. The temperature changes seen in this study were small though, being less than 1°C in most cases, so this factor would have only a very small influence.

Venous return is facilitated during immersion by the removal of the opposing gravitational forces, and the increased total peripheral resistance in response to the cold maintains a high thoracic blood volume. The resulting filling pressure would cause the blood pressure to rise, stimulating the baroreceptor reflexes to slow the heart rate. Denison et al. (7) found a 10% increase in cardiac output and pulse rate associated with work in thermoneutral water as compared to air. Counteracting this tendency, however, is the increased total peripheral resistance due to the cold, and the volume of tissues being perfused is decreased as a result of vasoconstriction (blood flow to cold, idle tissues is decreased).

The tissue oxygen extraction per unit of time would be expected to increase as a result of the decreased blood flow through the vasoconstricted tissues. When the linear velocity is low, the tissue tension of oxygen drops. The oxygen gradient from blood to tissue then increases, allowing greater oxygen uptake by the tissues.

Considering all the factors, the increased oxygen pulse is probably the result of an increased stroke volume and increased oxygen extraction by the tissues.

In the last two recovery periods after work in cold water, the heart rates dropped below resting values, in spite of an increased thermoregulatory oxygen consumption. This phenomenon and the decrease in heart rate below resting after the still immersion might be explained as a reflex bradycardia in response to the vasoconstriction from the cold stress and as a reduction in the heart rate caused by the fall in core temperature, in the case where the fall in rectal temperature was large.

Oxygen Consumption: A discussion of recovery and oxygen debt depend on the criteria for recovery. Three different baselines have been used commonly in the literature: a) the basal metabolic rate; b) the resting metabolic rate; and c) light work or working baseline (e.g., free-wheeling on a bicycle ergometer) (5,15).

The resting metabolic rate was chosen for this study because it is the most commonly used baseline and it yields reproducible results (15). This recovery oxygen measured in this way does not include the full recovery, but it does measure recovery oxygen related to the exercise directly.

Recovery to resting metabolism assumes that the conditions influencing the resting metabolism remain unchanged throughout the exercise and recovery period. The cold exposure during the immersion and exercise produced a thermal debt sufficient to cause variation in the recovery baseline. This situation is particularly obvious in the oxygen consumption values after the 20°C water exposure due simply to the onset of shivering. The control still immersion experiments involved a smaller heat loss than did the work immersions and so did not cause as much of a rise in oxygen consumption in the later part of the 30-min recovery period. However, the oxygen consumption immediately after the 17-min still immersion was higher than the oxygen consumption during the immersion. Perhaps this can be explained by the shivering observed during this period. Thus, it is not possible to determine the fraction of the increased oxygen consumption that is due to thermoregulatory responses. The first 10 min of recovery, however, were free of thermal shivering even after work in 20°C water, though this was not the case after the immersions without work. It is assumed that the elevated oxygen consumption during the period free of shivering is the recovery oxygen and that, during the last 20 min, it is primarily attributable to thermal recovery. It is doubtful that the pattern of oxygen uptake after exercise in cold water is similar to that after immersion with no work because the stimulus for shivering must have been circumvented or changed by the increased energy output of the exercise or, perhaps, the maintenance of thermogenic responses was precluded by the muscular activity.

The oxygen consumption during recovery was consistently higher after work in 20°C water. The difference is not as large as one would expect from looking at the oxygen uptakes during work. Oxygen debt, then, is related to the oxygen consumption required by the work load and apparently not affected by the absolute oxygen uptake during work under these conditions.

During R_2, R_3, and R_4, the oxygen uptake tended to be higher after work in 20°C water. This increase could be due to thermoregulatory responses, but though the subjects were obviously cold during that period, none started to shiver until after being out of the water at least 10 min. It could be a result of increased muscle tone since the differences are so small. However, the increase could also be due to the metabolism of lactate that was slow to diffuse out of cold, vasoconstricted muscle.

Energy Requirements: On the basis of the group means, it is possible to estimate the energy requirements of the immersion, work, and recovery from oxygen consumption and the drop in rectal temperature.

The values for the energy requirement (H) in Table VI were estimated from the equation:

$$H = (\dot{V}_{O_2} \times 44.8 \text{ kcal/liter})\, t + \Delta H_S$$

where ΔH_S is the change in heat stores and t is the duration of the period being considered. The ΔH_S can be approximated from the rectal temperature as

$$H_S = (T_{re}) \times (.83 \text{ kcal/°C-kg}) \times (\text{b.w.}) \times (0.67)$$

where T_{re} is the change in rectal temperature; 0.83 kcal/°C-kg is the specific heat of the body; b.w. is the body weight in kg; and 0.6 is a unitless correction factor for the use of T_{re} for mean body temperature.

These values in Table VI are only gross estimations, but they are indicative of the magnitude of the metabolic demands of immersion and cold. The total energy cost above the resting metabolic rate for the experiment in 26°C water was almost twice as much as in air, and for the experiment in 20°C water the cost was almost three times greater.

The total requirement during the 47 min was about 10% of the average daily caloric intake. The increased caloric demands of immersion may be one of the factors responsible for the subjective fatigue that lingers long after the exercise in cold water. This high caloric requirement may cause a depletion of metabolic substrates and the concomitant exhaustion.

In summary, the high metabolic demands of immersion and work in cold water may be a contributing factor to the subjective fatigue and should be considered in order to provide adequate caloric intake for the working diver. The results of this experiment suggest that recovery is, indeed, retarded by cold stress induced during work. This is an important point emphasizing again that there are other factors to be considered in planning repetitive dives besides nitrogen accumulation. With each dive, the diver not only increases his thermal debt, but he further decreases his capacity for physical recovery.

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TABLE VI. ESTIMATED ENERGY REQUIREMENTS (kcal).

	Submersion	Exercise	Recovery	Total	
	10 min	7 min	30 min	45 min	above resting
Environment					
Air	13.9	45.3	59.0	118.2	52.8
26°C water	22.8	73.7	59.1	115.6	90.1
20°C water	40.3	100.6	64.9	205.8	140.3

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Critical water temperature and effective insulation in scuba divers in Hawaii

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HANNA, JOEL M., AND SUK KI HONG. *Critical water temperature and effective insulation in scuba divers in Hawaii.* J. Appl. Physiol. 33(6): 770–773. 1972.—Thirteen regular scuba divers from Honolulu, Hawaii, were compared with fifteen control subjects in a water immersion test as developed by Rennie et al. (in: *Physiology of Breath Hold Diving and the Ama of Japan*). Water temperature ranged from 28 to 33° C. Subjects were immersed for 3 hr or until the onset of shivering; trials were repeated each day until the minimum temperature which could be tolerated for three hours with no visible shivering was determined. This was called critical water temperature (CWT). Divers tended toward a lower CWT at any level of subcutaneous fat thickness, but there were no apparent differences in maximal tissue insulation (I_{max}) that has been reported in other studies. The responses of 10 subjects of Japanese ancestry and 10 of European ancestry were examined separately. There were no great differences in response between ethnic groups.

cold tolerance; physical insulation; ethnic differences in cold tolerance

RECENT STUDIES OF DIVERS have suggested that regular diving in cold water can lead to acclimation to cold (14). Over a period of 45 days diving in cold water (2.5–3.5° C) acclimational changes favor chiefly a reduced heat loss rather than elevation in heat production, so that rectal temperatures fall without appreciable increases in metabolic heat production. The final result was a lower level of heat transfer from core to shell and a minor increase in metabolic heat production. In another instance, 26 well-trained divers in polar waters (near 0° C) experienced a mean drop in core temperature to 34° C during 55-min dives (1). That study also recognized the value of a high tissue insulation, for dives were routinely terminated upon the onset of shivering which would indicate the reduction of insulation.

Korean studies have also illustrated the reliance of divers upon mechanisms of heat conservation. At the outset it is women, rather than men, who are recruited because they have naturally thicker layers of subcutaneous fat (2). These diving women, called ama, also show a reduced critical water temperature (CWT) which permits them to tolerate a reduced core temperature without shivering (13). Moreover as compared to nondivers, the ama show a higher maximal tissue insulation (I_{max}) for a given thickness of subcutaneous fat throughout the year (3). Although they also show seasonal changes in metabolism (2), these do not seem to be of major thermogenic significance (7).

Since Arctic scuba divers, as well as the ama, were observed after frequent dives in cold water there can be little doubt that they were somewhat cold acclimated. The series of adjustments which they present are compatible with those reported for cold air exposure (9, 11). The present study was undertaken to determine to what degree divers in a more tropical setting were cold acclimated and to determine what adaptive mechanisms for cold tolerance were involved in the absence of a severe cold stress.

METHODS

Thirteen regular scuba divers were recruited in Honolulu, Hawaii. Six were professional divers from a local oceanographic institute, four were members of a Fire Rescue Squad specializing in ocean rescues, and the remaining three were sport divers, native to Hawaii and active spearfishermen. Three of the professionals were not native to the Islands, the other ten were. Two of the three non-Hawaiians were native to California. The professionals used wet suits for diving but the others frequently did not.

Fifteen control subjects were recruited from among the students at the University of Hawaii. Twelve had lived in Hawaii for life, two were from Southern California, and the remaining individual had lived in Hawaii since early childhood. None of the control subjects was a scuba diver, but five were regular surfers, engaging in the sport more than one time per week. All subjects in both groups were swimmers at infrequent intervals.

Anthropometric characteristics of all subjects are presented in Table 1. The major ethnic groups and numbers of subjects represented are Chinese (2), Japanese (10), Caucasian (14), and Chinese-Caucasian (2). The diver group tends to be older and somewhat heavier; however they have about the same subcutaneous fat thickness as controls. Since the groups do not differ in height or skinfold thickness, the weight difference probably results from greater muscularity in the divers.

Diver and control groups show a considerable age difference; however, earlier work has shown that the parameters considered in the present study are not greatly influenced by aging (10).

The basic procedure was developed by Rennie et al. (13) and followed in detail. A plywood tank of dimensions—84-inch length, 30-inch width, and 28-inch depth—

was lined with rubber and filled with water. The tank was provided with two pumps of high capacity which removed water through intakes at each end and discharged it through a number of holes along the sides. This was a particularly effective system of mixing; indeed, refractometric studies of ink dilution indicated that complete mixing was accomplished in 60–75 s.

Water temperatures were regulated to 0.01° C by maintaining the bath in a room at 24° C and heating the water to the desired temperature. This was accomplished with the aid of an immersion heater at either end coupled to a thermistor thermostat which was in turn connected to one of the water intakes as part of the recirculation system. This proved to be an extremely efficient system in that no appreciable deviation from the set temperature was noted during the course of any experiment.

Subjects reclined on a plastic mesh cot, immersed in water with only their faces exposed. Oxygen consumption was measured at 20-min intervals with a 13-l Collins spirometer. Rectal temperature (T_{re}) was measured with an indwelling rectal thermistor and recorded every 10 min. T_{re} was recorded to the nearest 0.01° C and the the thermistor calibration was checked after each three trials. Skin temperature was assumed to equal water temperature as described by Rennie et al. (13), who could detect no difference between skin and water temperatures after 1-hr immersion. In the present study several measurements of thigh and toe temperatures at the end of 3-hr immersion yielded less than 0.2° C difference in temperature between skin and water.

Each was tested at least 1 hr after his last meal and was required to sit quietly in the laboratory 1 hr before testing. Initial water temperature was set subjectively, the temperature being selected on the basis of subcutaneous fat thickness. The period of immersion was 3 hr and if an individual completed 3 hr without shivering, he was tested the next day at a temperature of 1° C lower. If he shivered during the 3 hr the experiment was terminated and he was tested the following day at 1° C higher. Shivering was defined as 10 min of visible tremor. Thus most subjects were tested twice but in some cases three or four trials were necessary to determine the critical temperature. The lowest temperature which an individual could tolerate for three hours without shivering was called his critical water temperature (CWT).

TABLE 1. *Physical characteristics and responses of control and diver groups*

Variable	Control	Diver	*t*
Age, yr	20.8 ± 2.3	31.4 ± 4.8	6.9*
Height, cm	175.8 ± 6.8	176.0 ± 7.9	0.0
Weight, kg	73.2 ± 11.5	81.6 ± 7.6	2.2*
Mean skinfold, mm	3.3 ± 2.0	3.7 ± 1.4	0.6
CWT, ° C	31.2 ± 1.2	29.9 ± 1.3	2.8*
Nonshivering			
T_{reo}, ° C	37.76 ± 0.28	37.62 ± 0.20	0.4
T_{re180}, ° C	36.44 ± 0.44	36.64 ± 0.42	1.2
Shivering			
T_{reo}, ° C	37.65 ± 0.39	37.59 ± 0.23	0.5
T_{res}, ° C	37.07 ± 0.33	36.94 ± 0.31	0.6
I_{max}, ° C·hr·m²/kcal	0.144 ± 0.032	0.164 ± 0.032	1.7*

See text for explanation. Values are means ± SD. * Significant at 5% or less.

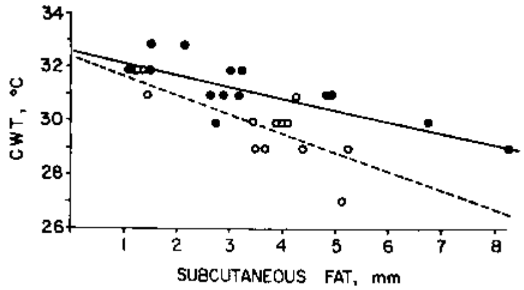

FIG. 1. Critical water temperature (CWT) as a function of subcutaneous fat thickness in 13 diver and 15 control subjects in Hawaii. Solid points and line (●——●) are controls; and open points and lines (○-----○) are divers. Difference between slopes is statistically significant between 0.10 and 0.05.

Insulation (I) was computed for 20-min intervals following Rennie et al. (13).

$$I = \frac{T_{re} - T_w}{M - 0.08\,M + S}$$

where: T_{re} is rectal temperature; T_w is water temperature; M is metabolic rate; S is change in body heat stores. A respiratory heat loss of 0.08 metabolic rate was assumed and changes in heat stores were assumed equal to body weight (kg) × 0.83 × 0.67 Δ T_{re}, calculated in 20-min intervals. Maximum insulation (I_{max}) was taken to be the highest I observed during the two trials. In practice this usually occurred shortly before shivering was observed.

Subcutaneous fat was measured at nine sites and totaled; 36 mm was subtracted to account for skin thickness and the remainder divided by the total number of single skinfolds, 18. This was called the average subcutaneous fat thickness (13).

RESULTS

Differences between diver and control subjects in CWT, I_{max}, initial rectal temperature (T_{reo}), temperature at the end of the nonshivering trial (T_{re180}), as well as rectal temperature at the visible onset of shivering (T_{res}) are presented in Table 1. When compared by means of a *t* statistic, divers show significantly lower CWT, and significantly higher I_{max}. Since subcutaneous fat thickness is greater in divers (Table 1) and insulation as well as CWT are influenced by fat thickness (12), a regression analysis was undertaken to determine if these differences could be accounted for by subcutaneous fat thickness and CWT are presented in Fig. 1. Both lines have negative slopes indicating a decreasing CWT with increasing amounts of subcutaneous fat. Linear correlations for divers and controls are −0.74 and −0.78, respectively, both values being significant beyond the 0.01 level. A test of the differences between slopes (t = 1.46, df = 24, $0.05 < P < 0.10$) suggests some real difference between the groups may exist. The actual distribution of points about the regression also suggests a difference; only one of the control group falls below the diver regression while only two divers fall above the control regression.

Figure 2 presents the relationship between I_{max} and

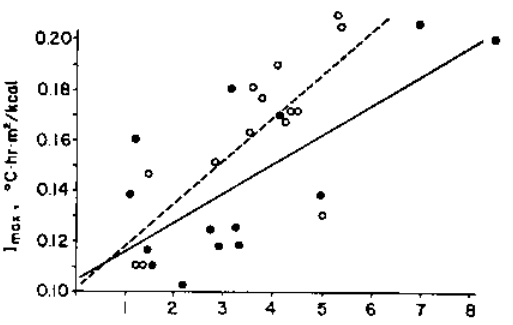

FIG. 2. Maximal tissue insulation (I_{max}) as a function of subcutaneous fat thickness in 13 diver and 15 control subjects in Hawaii (1° C·hr·m²/Kcal is equivalent to 0.485° C·m²/W). Solid points and line (●— — ●) are controls; and open points and lines (○-----○) are divers. There is no significant difference between slopes.

TABLE 2. *Summary of responses of Japanese and Caucasian subjects*

Variable	Japanese	Caucasians	*t*
Age, yr	24.3 ± 6.1	24.4 ± 5.9	0.0
Height, cm	171.3 ± 5.6	179.8 ± 5.5	3.2*
Weight, kg	76.8 ± 12.1	79.2 ± 8.9	0.4
Fat, mm	4.3 ± 1.3	3.3 ± 2.2	1.1
CWT, ° C	30.2 ± 0.9	30.8 ± 1.8	0.8
T_{reo}, ° C, nonshivering	37.65 ± 0.29	37.54 ± 0.40	0.6
T_{rem}, ° C, nonshivering	36.55 ± 0.33	36.53 ± 0.44	0.1
T_{reo}, ° C, shivering	37.54 ± 0.27	37.73 ± 0.41	1.1
T_{res}, ° C, shivering	36.95 ± 0.36	37.09 ± 0.54	0.6
I_{max}, ° C·hr·m²/kcal	0.170 ± 0.027	0.142 ± 0.037	18.5*

Values are means ± SD. * Significance at 5% or greater.

subcutaneous fat thickness. Correlations for both divers (0.75) and controls (0.69) are significant. Although I_{max} for a given subcutaneous fat thickness tended to be higher in divers as compared to controls, a test of differences between the slopes yields a *t* of 0.55 which is not significant. Likewise, an inspection of Fig. 2 shows no apparent difference between the distribution of the two populations.

Both diver and control groups were made up of subjects of different ethnic backgrounds. To determine if race had an effect, the responses of six controls and four divers of Japanese ancestry and a similar group of Caucasian ancestry were considered independently. Individuals were selected so that the groups were closely matched in anthropometric variables. Their mean anthropometric measurements and responses to the immersion test are summarized in Table 2. The Caucasians are taller and heavier and have somewhat less subcutaneous fat. There are no significant differences in any of the responses to immersion except in I_{max} where the Japanese are somewhat higher. To determine if this represented a true ethnic difference or was simply the result of a greater fat thickness, a regression of I_{max} on mean skinfold thickness was performed and is presented in Fig. 3. The correlation in Japanese was 0.62 and 0.82 in Caucasians. The slopes of the lines are quite similar. A comparison yielded a *t* of only 0.14. There is some difference in intercept; however, computation of confidence limits (Fig. 3) shows these differences are not significant. Hence there is no apparent difference in I_{max} between the Japanese and Caucasian subjects which cannot be explained by differences in subcutaneous fat thickness.

DISCUSSION

The lower CWT of divers at any level of subcutaneous fat suggests that regular scuba diving, even at mild Hawaiian water temperatures of about 23° C, confers some degree of cold acclimation. This lower shivering threshold has characterized other divers as well, but always in conjunction with a higher level of cold exposure. Thus Rennie (12) compared ama with nondiving Korean women from the same villages and found the divers could tolerate considerably lower water temperatures before shivering. When differences were corrected for subcutaneous fat thickness, as in the present study, the divers still maintained a lower CWT. At 2-mm subcutaneous fat thickness the diving women had a CWT 3° C below nondiving women. This is somewhat greater than the 0.8° C advantage of divers in the present study (Fig. 1) and probably results from the greater cold stress in Korea. Similarly scuba divers show reduced shivering thresholds after diving in the Arctic. Skreslet and Aarefjord (14) compared the responses of three scuba divers in a standardized cold immersion test before and after 45 days diving in the Arctic. During the diving period there was a progressive diminution of shivering and at the end of the 45-day period they shivered less when exposed to the standard cold stress.

The degree of cold stress to which the divers are exposed is probably mild because only CWT among the parameters measured shows any modification which could be related to cold acclimation. Other studies of more severe cold conditions have shown increases in I_{max} (12, 13), as well as a greater reduction in core temperature in acclimated subjects (9, 14). The central temperature at which shivering is initiated has also been shown to be lower in acclimated subjects (9, 14). Table 1 indicates no great differences in I_{max}, T_{rc}, or T_{res} between diving and control groups. In fact, there was no significant dif-

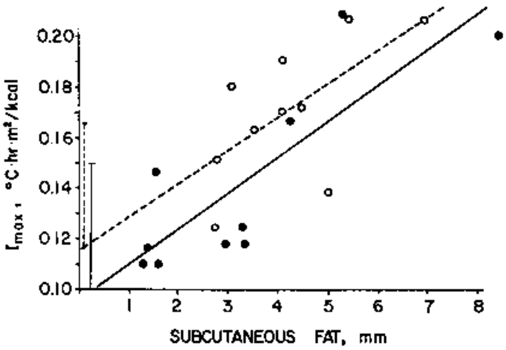

FIG. 3. Maximal tissue insulation (I_{max}) in subjects of Japanese and European ancestry (1° C·hr·m²/Kcal is equivalent to 0.485° C·M²/W). Open points and lines (○----- ○) are 10 subjects of Japanese ancestry; and solid points and line (●——●) are 10 subjects of European ancestry. Brackets at intercepts illustrate 95% confidence limits for Japanese (± 0.053) and European (± 0.054) regression.

ference in $I_{\max}$ for a given subcutaneous fat thickness between the two groups (see Fig. 2).

These results also suggest that adaptation to cold develops in steps. The first step of cold adaptation seems to be an elevation of shivering threshold, indicated by a significant lowering of CWT, as observed in the present and other studies (12, 13). Most probably, this phenomenon may be attributable to habituation and is very important in economizing the body heat balance in cold. Theoretically, one can maintain the body heat balance in cold by increasing heat production (i.e., shivering), but the latter process may accelerate rather than retard loss of central body heat in unclothed or lightly clothed individuals (8). As the degree of cold stress increases, $I_{\max}$ for a given subcutaneous thickness becomes higher as a result of more extensive lowering of CWT, possibly coupled with either more extensive vascular constriction or more effective countercurrent heat exchange in the limbs. Such an adaptation to cold has been observed in ama (5) but not in Hawaiian divers.

The responses of the subjects of Japanese and European ancestry (see Table 2 and Fig. 3) show the regression of $I_{\max}$ on subcutaneous fat. The sample size in the present study is rather small and Fig. 3 shows the variation within groups is large; hence some qualification is necessary until a larger sample is obtained. Still some suggestions relevant to ethnic differences can be made. There is no statistical difference in slopes ($t = 0.14$); the slope for Japanese is 0.014 and for Europeans is 0.013. Similarly, intercepts, which represent nonfat insulation, are not significantly different ($t = 0.65$). Thus there seems to be no great difference between the Japanese and white subjects that cannot be accounted for by differences in subcutaneous fat.

Most other studies which have compared subjects of Oriental with European ancestry have compared residents of an Asian country with residents of the United States or Europe (6, 13). Because of technological differences it is unlikely that both peoples had similar life histories of cold exposure. If real inherited differences do separate the groups they may be enhanced or diminished by cold acclimation. Technological differences would probably favor less cold exposure and less acclimation in people living in Europe or the United States. Thus Rennie et al. (13) compared Koreans with white Americans and found Koreans to maintain a greater $I_{\max}$ at any level of subcutaneous fat thickness. Although the regressions had similar slopes, the intercept (representing nonfat insulation) for Koreans was higher. At 0-mm fat thickness Koreans had an $I_{\max}$ of 0.092 while white Americans had an $I_{\max}$ of 0.050. This was interpreted to show that Orientals had a thicker nonfat insulative shell. The present study suggests that with more similar life histories of cold exposure, Orientals and whites may respond in a more similar manner than indicated by the work of Rennie et al. Specifically the higher $I_{\max}$ at zero subcutaneous fat in the present white group casts some doubt upon the intercept for the American group tested in the earlier study. Studies of Koreans (3) and native Japanese (6) yielded intercepts of 0.096 and 0.085, respectively. These values are close to the present intercepts (0.096 and 0.115). Indeed all such $I_{\max}$, subcutaneous fat regression yielded intercept values which cluster about 0.100 with the single exception of the white subjects tested by Rennie et al. (13). True ethnic differences in nonfat insulation may exist, but the present observations indicate they might be less than anticipated from earlier work.

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Pattern of cold adaptation in women divers of Korea (ama)[1,2]

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ABSTRACT

Pattern of cold adaptation in ama who daily dive in water of 27–28 C in summer and of 10 C in winter has been systematically studied from 1959 to 1969. The total extra heat loss for diving work is estimated to be 1,000 kcal/day throughout the year. The following adaptive phenomena to cold are found in Korean ama as compared with non-diving Korean women: *1)* a consistent, reversible increase in basal metabolic rate of ama in winter (i.e., metabolic adaptation), which seems to be associated with increased utilization of thyroid hormone; *2)* a very small (but significant) increase in $\dot{V}_{O_2}$ in response to exogenous norepinephrine in winter; *3)* a lower critical water temperature at a comparable subcutaneous fat thickness throughout the year (i.e., insulative-hypothermic adaptation); *4)* a significantly greater maximal thermal insulation at a comparable subcutaneous fat thickness throughout the year; *5)* a lower heat flux from the limb coupled with a higher blood flow throughout the year during whole-body immersion in water of critical temperature, suggesting a more efficient countercurrent heat exchange system in the limb; and *6)* a lower finger skin temperature and blood flow in winter during hand immersion in 6 C water (i.e., vascular adaptation). These results indicate that the basic pattern of cold adaptation in man is qualitatively similar to that observed in other homeothermic animals.—Hong, S. K. Pattern of cold adaptation in women divers of Korea (ama). *Federation Proc.* 32: 1614–1622, 1973.

Along the southern coast and islands of South Korea, there live an estimated 15,000 breath-hold women divers who each day harvest the ocean floor, gathering seaweeds, abalones, snails, and sea urchins. They are initiated into their profession at age 12 or so and continue to dive during most of their lives. They are known as hae-nyo (sea women) or jamsoo (diving women). Their counterpart is the ama (sea women) of Japan. I shall refer to them by the latter name since the first scientific description of their diving was given by Teruoka (40) for the Japanese divers.

There are many indirect lines of evidence to indicate that many thousands of Korean women have been actively engaged in daily diving work for centuries (10). Evidently, both men and women were engaged in diving work at the beginning but somehow men dropped out later. In contrast, one finds both men and women diving in Japan where diving is allowed only during the late spring and summer.

Inasmuch as these diving women present physiologists with many interesting problems (33), I have, since 1959, made a series of physiological studies in close collaboration with Dr. Hermann Rahn and Dr. D. W. Rennie of the State University of New York at Buffalo and with Dr P. H. Lee, Dr. D. H. Kang, Dr. B. S. Kang, Dr. S. H. Song, Dr. P. K. Kim, and others of Yonsei University, Seoul, Korea. Everything I will describe in this symposium is the direct result of highly coordinated international scientific collaboration carried out during 10 years (1959–1969) across the Pacific Ocean. I gratefully acknowledge all of these collaborators for their splendid, unselfish efforts. Most of all, I take this opportunity to thank the many divers who so willingly cooperated with us and tolerated all the rigid experimental protocols.

Originally, we were interested in the mechanics of diving (17, 18), respiratory mechanics (12) and gas exchange during repetitive diving (17, 18), diving bradycardia (19), physical fitness (14, 24), and the long-term adaptation of respiratory system in these divers (39). However, it immediately became clear that there are even more challenging problems associated with diving in cold water without any protective clothing. These women divers wear only a cotton bathing suit throughout the year, despite a great seasonal variation in the air and water temperatures (Fig. 1).

COLD WATER STRESS

To estimate the extent of cold water stress, the oral temperature was initially measured before and after the dive during four seasons of a year (23). As shown in Fig. 2, the oral temperature dropped by 2–3 C during a diving shift in all seasons. The greatest reduction was seen in winter (January) when it routinely fell to 33 C or less. Evidently, the ama continue diving until their body temperature is reduced to a certain intolerable level. Since the rate of reduction of body temperature is an inverse function of water temperature, the duration of a diving shift was a direct function of the water temperature. Moreover, the number of daily diving shifts was also related to the water temperature, resulting in only one or, rarely, two shifts in the middle of winter, in contrast to two or three shifts in the middle of summer. As a result, the total working time in the

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[1] From the Symposium on *Problems in Temperature Regulation and Exercise* presented at Indiana University, Bloomington, Indiana, December 13, 1971.

[2] This investigation was supported by National Science Foundation Grants G-8794 and G-24044, U.S. Air Force Contract AF 41 (657)-302, U.S. Army Grants DA-MD-49-193-64-G111, DA-CRD-AG-592-544-66-G36, and DA-CRD-AFE-S92-544-67-G76, and Public Health Service Research Grant 1 R01 GM14337.

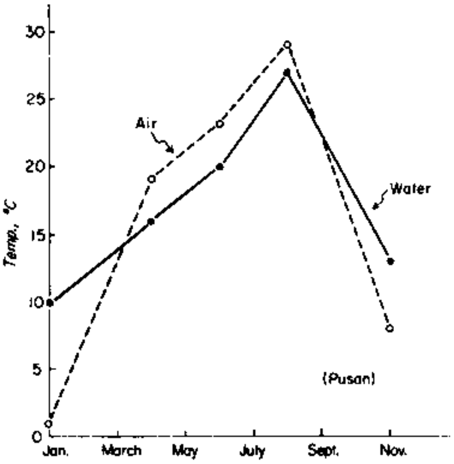


Figure 1. Seasonal variation in air and seawater temperatures in Pusan, Korea, area. (From Hong (9).)

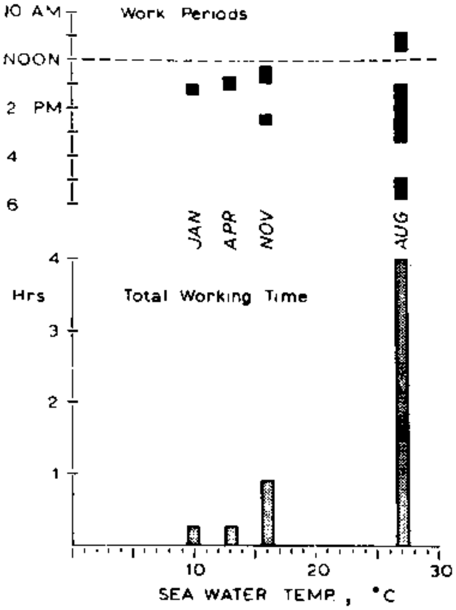


Figure 3. Time and duration of each work shift (top) and the total working hours as a function of the seawater temperature.

water varied from 15 to 20 min in winter (water temperature 10 C) to 4 hours in summer (water temperature 27–28 C) (Fig. 3). These observations clearly indicate that the working time in water is primarily determined by the water temperature.

As an attempt to estimate the amount of heat lost to the water over and above the resting heat loss in air, both the rectal temperature and the oxygen consumption were measured during a work period (24). Although the ama stayed in the water for 40 min in summer and 25 min in winter, the rectal temperature at the end of a work period was approximately 35.0 C in both seasons. However, due to a marked difference in the water temperature (and hence the skin temperature), the estimated mean body temperature decreased to 30 C in summer and to 25 C in winter. At the same time, the oxygen consumption increased from approximately 400 ml/min at rest in air to approximately 1,000 ml/min in summer and to 1,400 ml/min in winter (Fig. 4). Based on these findings, the cumulative extra heat loss per diving shift is estimated to be 388 kcal in summer and 557 kcal in winter (Fig. 5). Since they take two or three shifts a day in summer and one or two in winter, the total extra heat loss is estimated to be on the order of 1,000 kcal/day. Actual dietary surveys indicated that the ama consumes approximately 3,000 kcal a day whereas the caloric intake of nondiving women of comparable age was of the order of 2,000 kcal a day.

These observations suggest that the ama subject themselves to a daily cold stress greater than that of any other group of human beings yet studied. In view of such a severe cold water stress,

Figure 2. Diurnal changes in oral temperature of the ama during various seasons of the year in relation to the diving work. Each line represents one individual subject. Continuous descending lines indicate period of water immersion. (From Kang et al. (23).)

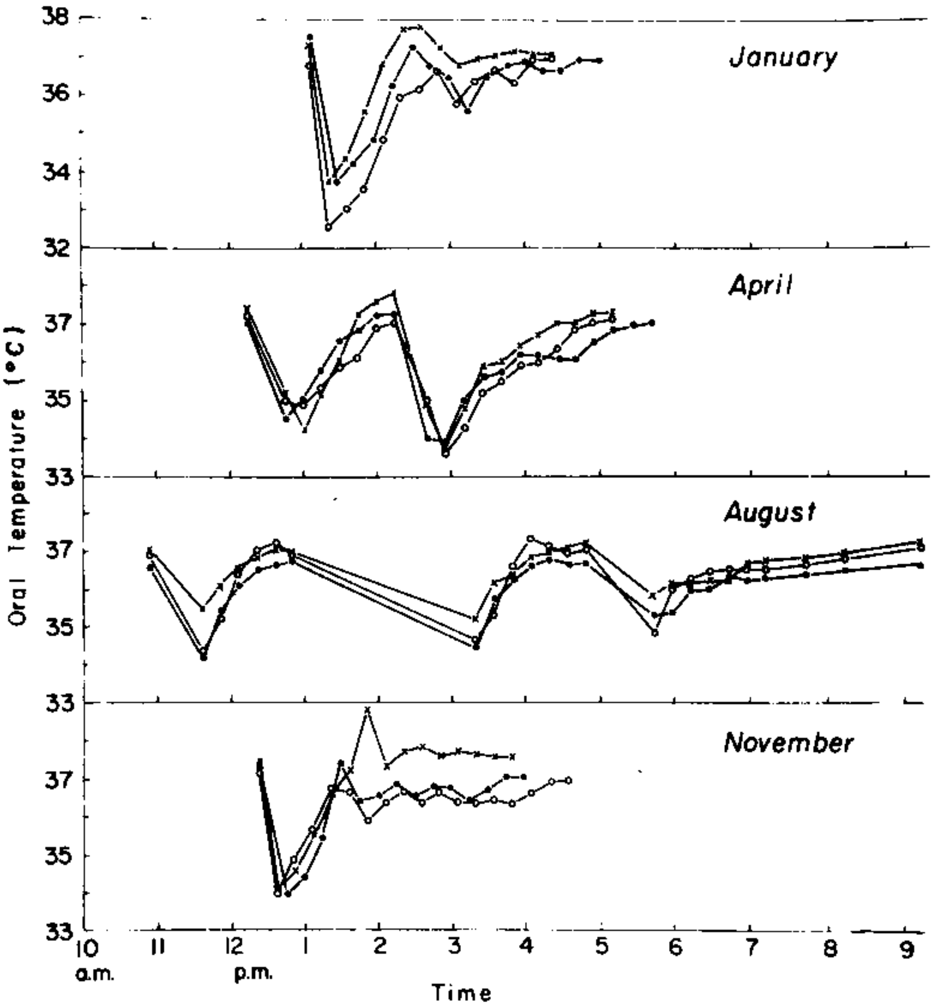
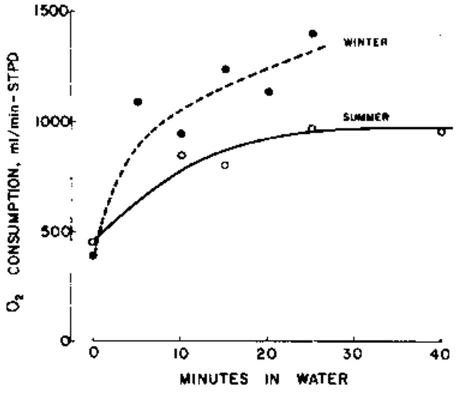


Figure 4. Oxygen consumption during the period of diving work in summer and winter. Each point represents the average of 5 to 12 measurements in three ama. (Individual data are given in ref. 24.)



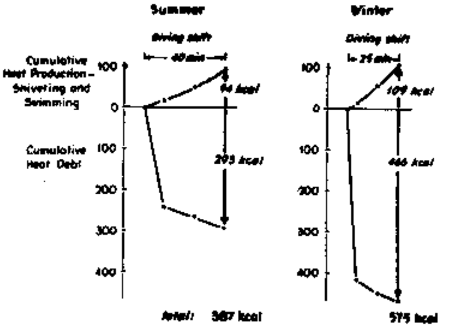


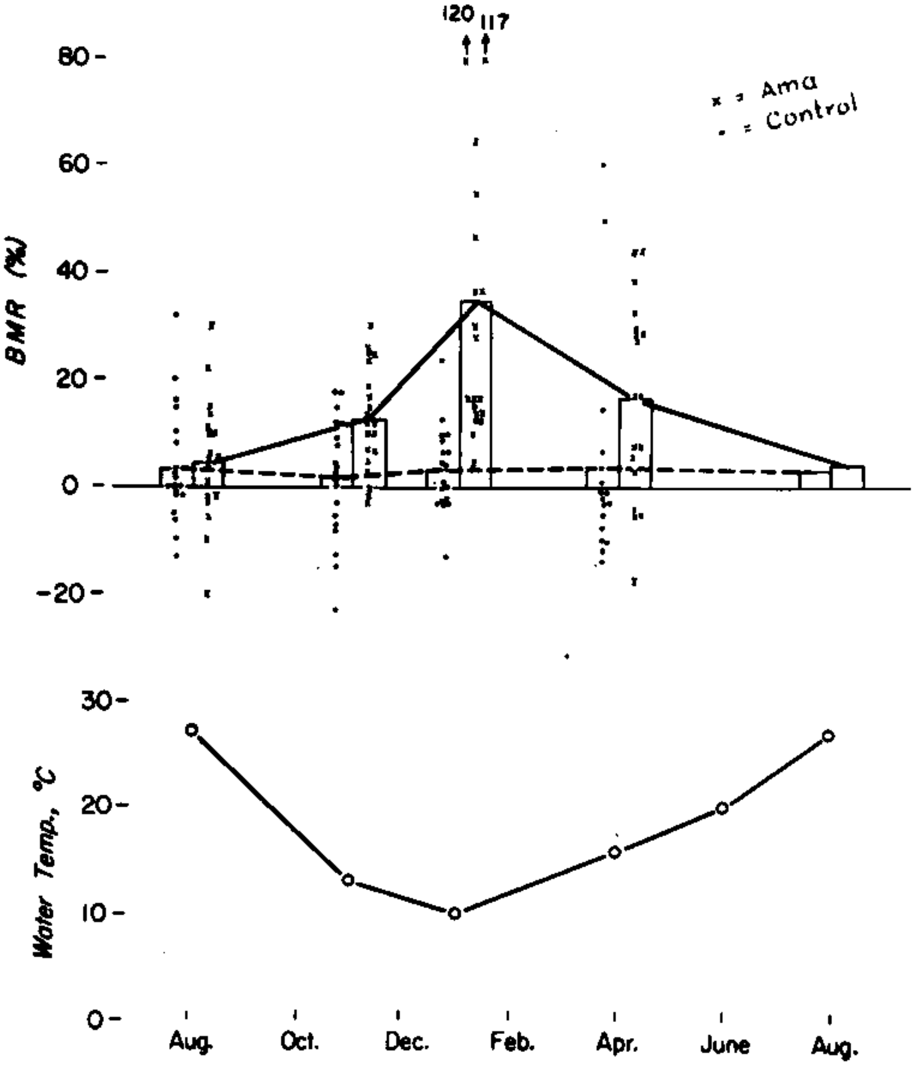
Figure 5. Increment in caloric expenditure due to diving work in summer and winter. Cumulative calories generated by shivering and swimming, estimated from oxygen consumption, are indicated as upward deflections. Cumulative heat debt during a diving shift, estimated from weighted water temperature and rectal temperatures, are indicated as downward deflections. (From Hong (11).)[3]

to which the ama are exposed the year around, evidence for possible adaptation to cold was sought.

METABOLIC ADAPTATION

The occurrence of metabolic adaptation to cold has been widely observed in many animals. Although many physiologists attempted to demonstrate this phenomenon in man, only Japanese groups have been able to document it in Japanese (but not in Canadian missionaries) living in Japan (41). We also failed to observe any seasonal variations in the basal metabolic rate (BMR) of nondiving women living in Pusan, a city located in the southeastern tip of Korean peninsula. However, the ama living in the same community showed a marked seasonal variation in BMR (Fig. 6). In general, the level of BMR in the ama was comparable to that of nondivers during summer but was approximately 30% greater in winter when the seawater temperature is lowest (23). Since the level of BMR is known to be altered by the amount of protein intake or anemia, we determined the erythrocyte counts (along with the hemoglobin concentration) as well as the urinary nitrogen output (23). There were no differences between the two groups, strongly suggesting that the observed increase in BMR in the ama in winter is primarily due to the cold stress. To ascertain this finding, we subsequently repeated the measurements of BMR on three more occasions (11, 16, unpublished data of Song et al.). As summarized in Table 1, there was a significant increase of BMR in winter in the ama (but not in nondivers) in all studies. The physiological meaning of a small difference in BMR between the ama and nondivers in summer is not clear at present. These observations clearly indicate that the ama are subjected to a reversible increase in BMR during cold weather. Admittedly, the practical value of increased BMR is minor as a defense against body cooling in water. It seems likely, however, that increased resting metabolism is a latent human adaptation to severe body cooling; rarely, if ever, aroused in modern man by cold air exposure, but manifest in the ama during winter because of the unusual degree of cold water stress that they endure.

Figure 6. Seasonal variations in basal metabolic rate. Bar graph represents average BMR for each group. Lower part of graph shows surface water temperature during the year. (From Kang et al. (23).)



What is the mechanism underlying such an increase in BMR in winter? One likely possibility is that there may be a simultaneous increase in the activity of the thyroid gland in winter in these women divers. To test this possibility, ^{131}I uptake of the thyroid gland was studied in both summer and winter (16, unpublished data of Song et al.). The elevated BMR of the ama in winter was not associated with an increased amount of thyroid ^{131}I uptake. However, the rate of release of ^{131}I from the thyroid gland was significantly faster in the ama as compared with nondivers, especially in winter (Fig. 7). Resin T-3 ^{131}I uptake was the same in both groups and unchanged throughout the year. While

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[3] Figures 5, 9, and 10 are reprinted with permission from *Physiology of Breath-Hold Diving and the Ama of Japan*. Washington, D. C.: Natl. Acad. Sci.—Natl. Res. Council, publ. 1341, 1965.

TABLE 1. Seasonal changes in basal metabolic rate[a] in the ama and nondivers[b]

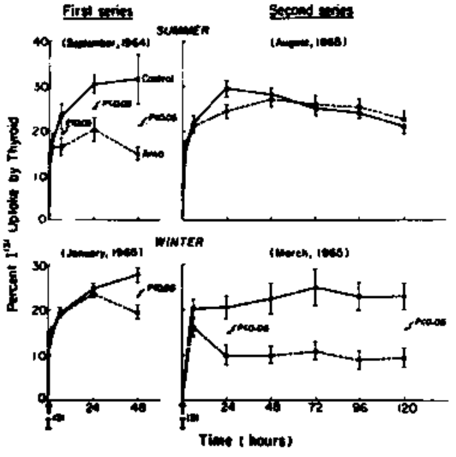
Year	Summer		Winter	
	Amà	Nondivers	Ama	Nondivers
1961 (23)	$+5.0\pm2.2$[d]	$+3.7\pm2.2$	$+35.1\pm7.5$	$+3.5\pm1.5$
1963–64 (11)	$+7.9\pm1.6$	$+2.0\pm2.4$	$+19.9\pm3.4$	$+0.8\pm3.2$
1964–65[c]	$+12.0\pm2.7$	$+3.1\pm2.4$	$+21.4\pm3.7$	$+5.7\pm3.0$
1965[c]	$+11.2\pm2.4$	$+7.1\pm1.6$		
Average	$+9.0\pm1.2$	$+4.0\pm1.1$	$+25.4\pm2.3$	$+3.3\pm1.7$
	($P < 0.005$)		($P < 0.005$)	
			(NS)	
	($P < 0.005$)			

[a] % deviation from DuBois' standard. [b] Number of subjects in each series was 20 each of the ama and nondivers. [c] Unpublished data of S. H. Song et al. [d] Values are means ± SE.

more critical experiments are needed along this line, these results suggest that the most likely reason for the seasonal increase of BMR is increased utilization of thyroid hormone by peripheral tissues.

Having thus failed to establish a relation between the metabolic adaptation and the thyroid function, we then began to wonder if this metabolic adaptation is an extension of the shift from shivering to nonshivering thermogenesis described by Davis (3) and Joy (21) for chamber cold-acclimated men. To this end, we investigated the calorigenic action of norepinephrine (22). As tabulated in Table 2, a significant increase in oxygen consumption in response to exogenous norepinephrine was observed in the ama only in winter. However, the magnitude of this increase in oxygen consumption was equivalent to only approximately 7.5% of the prenorepinephrine level. Although this figure is much lower than that observed in cold-acclimated small mammals (20), it is strikingly similar to that observed in chamber cold-acclimated (3, 4) or in Antarctic cold-acclimatized men (2). The urinary excretion of norepinephrine also increased significantly in winter in the ama (from 42 to 65 µg/day) but not in nondivers (22). These observations led us to conclude that norepinephrine induces only a small calorigenic action in the ama in winter, suggesting that the development of nonshivering thermogenesis is not the main feature of cold acclimatization, at least in the ama.

Figure 7. Thyroid ^{131}I uptake (percent oral dose) in summer (left) and in winter (right). (From unpublished data of Song et al.) (Mean ± SE.)



The data presented above leave no doubt, at least in our minds, that a metabolic adaptation indeed does occur in the ama in winter. However, the mechanism underlying this metabolic adaptation is not at all clear at present.

MAXIMAL TISSUE INSULATION

In contrast to controversial nature of metabolic adaptation to cold in man, an increase in maximal tissue insulation as a cold-adaptive mechanism appears to be more widely accepted among physiologists. The most outstanding example of the latter mechanism may be found in naked Australian aborigines who have fat thickness equal to nonadapted control subjects but are able to increase tissue insulation to values almost double those of the scientific party (5).

TABLE 2. Changes in oxygen consumption[a] during norepinephrine infusion

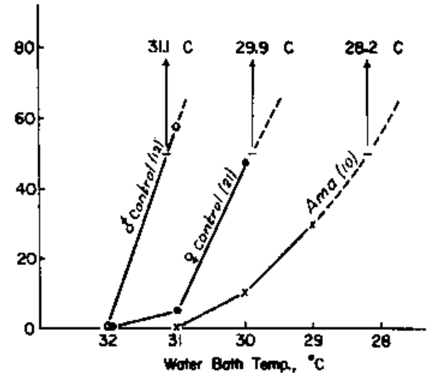
	Ama	Control
Summer		
A) Before NE	134.6 ± 5.0[b]	138.4 ± 4.0
B) During NE	133.7 ± 4.0	140.2 ± 4.5
$B - A$	-0.9 ± 1.4 (NS)	$+1.8\pm1.4$ (NS)
Winter		
A) Before NE	158.0 ± 6.8	150.0 ± 4.7
B) During NE	169.9 ± 6.7	149.9 ± 3.6
$B - A$	$+11.9\pm1.8$ ($P < 0.025$)	-0.1 ± 3.2 (NS)

[a] ml/min per m². [b] Values are mean ± SE obtained from 8 subjects (see ref. 22 for individual results).

For the measurement of maximal tissue insulation (I) in the ama we adopted the water-bath method developed by Rennie et al. (36) for their studies on the Eskimo. To provide a reproducible, uniform degree of cold exposure in all experiments, subjects clothed in cotton swim suits were immersed except for their faces in a well-stirred, constant temperature water bath regulated to within 0.01 C of the desired temperature. The water temperature was kept slightly warmer than "critical water temperature" (CWT), defined as the lowest water temperature the subject tolerated for 3 hours without shivering. In this series of studies, Korean male subjects were also included in order to see if there is any sex difference.

In an effort to obtain a more quantitative measure of CWT, the water temperature at which 50% of the subjects shivered was compared (9, 13). As shown in Fig. 8, this temperature varied from 31.1 C in the males to 29.9 C in the nondiving females and 28.2 C in the ama. In other words, CWT is considerably lower in the ama as compared with nondiving females and the males. The observed difference in CWT between the nondiving females and the males can be accounted for by the difference in subcutaneous fat thickness, as indicated by an inverse relationship between CWT and the subcutaneous fat thickness in subjects from a nondiving population (Fig. 9). Since women generally had a thicker fat layer than men, they generally had a lower CWT. However, the ama, who are usually lean, tolerated without shiv-

Figure 8. Incidence of shivering at various water temperatures. Numerical figures in parentheses indicate number of subjects. Water temperatures at which 50% of subjects shivered are shown on the top for each group. (From Hong (9).)



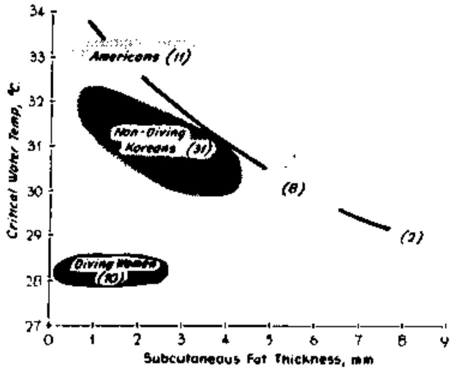
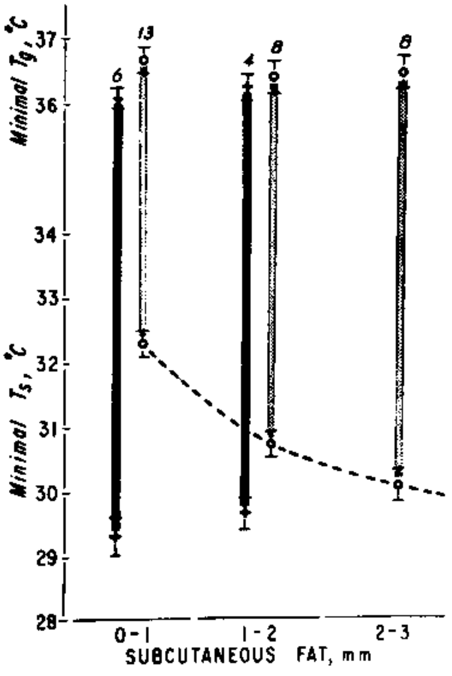


Figure 9. Critical water temperature (defined as the coldest water subjects tolerate for 3 hours without shivering) as a function of mean subcutaneous fat thickness of U.S. men and women, nondiving Korean men and women, and ama. The areas encompass the range of values for the indicated number of subjects. (From Rennie (34).)

ering much cooler water than nondivers of comparable fat thickness (see Fig. 9). Such an elevation of shivering threshold in the ama was observed in both summer and winter and may represent true cold adaptation.

As a consequence of their elevated shivering threshold, the ama sustained for long periods—at least 3 hours—a relatively large temperature gradient from body core to body surface (water temperature). This is illustrated in Fig. 10, which summarizes the minimal average rectal and skin temperatures (measured after 3 hours in water) of ama and nondiving women of comparable age. Among women having less than 1 mm fat thickness, the maximal rectal temperature–water temperature gradient sustained for 3 hours without shivering was 6.8 C for the ama compared to 4.4 C for nondivers (34). For instance, most ama were able to remain in 29 C water for 3 hours without shivering despite a decrease in rectal temperature to 36 C or below, whereas nondivers of comparable fat thickness shivered in water any colder than 33–32 C and at a time when their rectal temperatures exceeded 36.5 C. Considering the fact that voluntary muscular activity in cold water accelerates rather than retards loss of central body heat in unclothed or lightly clothed individuals (25), the advantage of suppressing shivering in the ama, who are usually lean, is rather clear in terms of economizing the body heat balance.

Figure 10. Minimal rectal temperature (top) and average skin temperature (bottom) of ama (+) and nondiving women (○) immersed for 3 hours in water of critical temperature. Numbers above symbols indicate the number of subjects. (From Rennie (34).)



Values of maximal tissue insulation were computed from the measurements of rectal temperature, water-bath temperature, and oxygen consumption during the nearly steady-state conditions of the third hour of immersion: I = (rectal temp. − bath temp.)/rate of skin heat loss. Skin heat loss was assumed to equal metabolic rate minus respiratory heat loss plus loss of stored body heat during the third hour (37). In several series (15), I values were computed during the last 20 min of 2-hour immersion. The results obtained from 89 nondiving women and men in both summer and winter are summarized in Fig. 11. There is a highly significant linear correlation between I and subcutaneous fat thickness ($R = 0.71$; $P < 0.001$). The regression of maximal body insulation on fat thickness for all subjects, indicated at the top of Fig. 11, clearly illustrates the insulative importance that such a fat thickness has. Based on the slope of this regression, one can calculate the insulative value of fat to be equivalent to 0.8 clo/cm fat. Superimposed on this regression are the mean values for fat thickness and I for different groups of subjects tested during the last 10 years (Table 3). In agreement with pioneering work of Hardy et al. (7), women subjects had a 30% greater I than men of comparable age due entirely to a thicker layer of subcutaneous fat. This extra insulation against cold provided by subcutaneous fat may be one reason why, centuries ago, women were selected for diving in Korea. Although there are perhaps other unknown socioeconomic factors which would account for this distribution of labor, it is interesting to note that, in Japan, where divers work only during warm seasons, both men and women engage in diving.

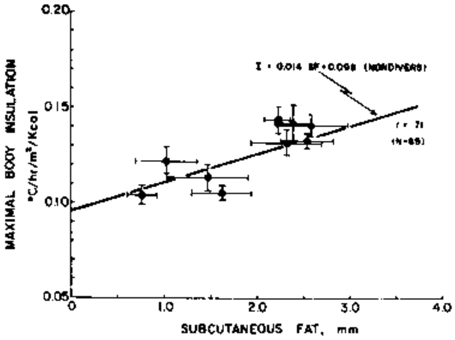


Figure 11. Maximal thermal insulation as a function of mean subcutaneous fat thickness in nondiving Korean men and women. Mean body insulation ± 1 SE obtained from different groups (see Table 3) has been superimposed on the single regression computed for all 89 subjects used in four studies (13, 15, 29, 37).

The results obtained from 30 ama are shown in Fig. 12 along with the regression line for nondivers. As in the control (nondivers) series, there was a highly significant correlation between I and fat thickness ($R = 0.50$; $P < 0.01$). Superimposed on this regression for the ama are the mean values for fat thickness and I for different series of experiments (see Table 3). The slope of the regression line for the ama is not different from that for nondivers while the intercept with the Y-axis is significantly greater in the ama compared with nondivers ($P < 0.025$), indicating an increase of the nonfatty insulative shell of the body in the ama. In other words, the value of I for a given fat thickness is significantly greater in the ama than in nondivers. Although our original studies (9, 13, 34) showed that the value of I in the ama for a given fat thickness is greater than in nondivers only during winter, subsequent studies indicated that I for a given fat thickness is higher in the ama than in nondivers throughout the year (15).

Since the value of I is primarily determined by the rectal–water temperature gradient, it is not surprising to see higher I in the ama compared with nondivers whose CWT is considerably

TABLE 3. Maximal tissue insulation (I)[a] in the ama and nondiving Korean men and women

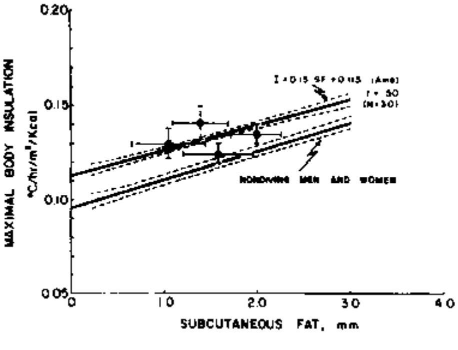
Year	Summer				Winter			
	Age, years	No. subj.	Mean fat thickness, mm	I	Age, years	No. subj.	Mean fat thickness, mm	I
				Ama				
1961–62 (13, 37)	42±3	8	1.99±0.26	0.135±0.005	41±6	10	1.04±0.20	0.130±0.008
1966–67[b] (15)	40±2	6	1.38±0.30	0.141±0.009	41±2	6	1.57±0.38	0.124±0.006
				Nondiving women				
1961–62 (13, 37)	33±2	4	2.21±0.14	0.144±0.007	37±2	8	2.53±0.28	0.133±0.004
1962 (29)	63±1	17	1.02±0.33	0.122±0.007	22±1	13	2.59±0.38	0.141±0.006
1966–67[b] (15)	36±2	6	2.39±0.21	0.142±0.010	36±2	6	2.31±0.38	0.132±0.007
				Nondiving men				
1961–62 (13, 37)	30±2	7	1.46±0.44	0.113±0.007	31±2	12	1.61±0.33	0.105±0.004
1962 (29)	66±1	17	0.76±0.16	0.104±0.005				

Each value represents means ± SE. [a] °C/kcal per hour per m². [b] I values were obtained during the last 30 min of 2-hour immersion.

higher than that of the ama (*Vide supra*). In other words, the elevation of I in the ama appears to be secondary to the elevation of the shivering threshold, which may be attributed to habituation. On the other hand, this could be due to more extensive vascular constriction or more effective countercurrent heat exchange in their limbs. Regardless of its mechanism, it is of interest to note that the basic pattern is identical to that observed in Australian aborigines (5).

Another interesting fact with regard to I was obtained during the early phase of investigation by Rennie et al. (37). They found that for a given fat thickness, American males and females living in the Buffalo, N.Y. area have a significantly lower I than Koreans living in the Pusan area. This observation is of considerable interest since the average annual temperature in Buffalo is considerably lower than that in Pusan. This difference between the two ethnic groups may suggest either that there exist racial differences in man's physical regulation of temperature, or that the actual living environment (especially the subclothing temperature) is warmer in peoples living in Buffalo because of superior heating systems. To answer this question, Dr. Hanna in our laboratory in Hawaii, measured I values in male subjects drawn from various ethnic groups. These subjects were born and raised in Hawaii and thus their living environment is the same. When comparisons were made for a given fat thickness, I values were not different between the two groups, Caucasians and Orientals (unpublished data). This finding suggests that the difference in I between Americans in Buffalo and Koreans in Pusan may not be an ethnic difference.

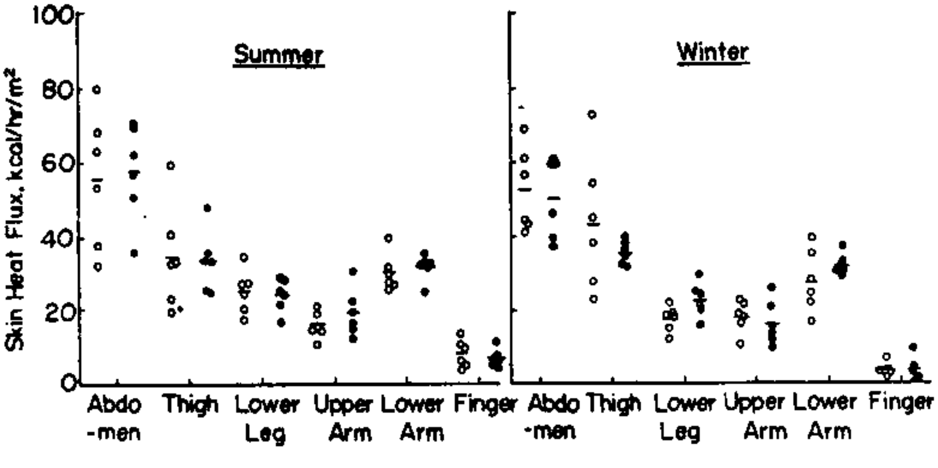
Figure 12. Maximal thermal insulation as a function of mean subcutaneous fat thickness in the ama (top). Mean body insulation ± 1 SE obtained from different groups (see Table 3) has been superimposed on the single regression computed for all 30 ama used in three studies (13, 15, 37). At the bottom is indicated the regression line for nondivers taken from Fig. 11. The shaded area represents 1 SE of estimated value of I.



REGIONAL HEAT LOSS AND BLOOD FLOW

In an attempt to better interpret the above insulation data, we measured both the regional heat flux and the limb blood flow during 2–3 hours of immersion in water of 30, 31, and 33 C (15, 35). The heat flux was measured by calibrated gradient calorimeter discs of the type described by Hatfield (8), and the blood flow by using mercury-in-rubber gauges (42). The heat flux was considerably higher during the first 30 min of immersion, following which it leveled off. The magnitude of average heat flux from various regions during the last 30 min of immersion in water of 30–33 C is shown in Fig. 13. As expected, heat flux from the abdominal region is the highest. However, it is of interest to note that the limbs also lose a considerable amount of heat, even at lower water temperatures. Despite such a regional difference, the magnitude of heat flux in each region was not significantly different between the ama and nondivers, indicating that

Figure 13. Average regional skin heat flux during the last 30 min of immersion in 30, 31, and 33 C water. Each point represents the average of values obtained in three different temperatures from each subject. Open and solid circles represent the ama and nondivers, respectively. (From Hong et al. (15).)



the higher I in the ama cannot be attributed to preferential shut-down of heat loss from any particular region.

In general, the time course of changes in the limb blood flow during immersion was similar to that of the limb heat flux. However, at a given water temperature, the limb blood flow, especially the finger blood flow, was greater in the ama than in nondivers in both summer and winter (Table 4). In other words, the heat flux for a given blood flow tended to be lower in the ama, suggesting that the ama may have a more efficient countercurrent heat exchange system in the limbs. Perhaps through this mechanism arterial blood is precooled before it reaches the peripheral zone, thereby reducing the thermal gradient. As a result, peripheral heat loss is economized despite greater peripheral blood flow.

The fact that the limb heat flux was maintained high during immersion in water of critical temperature merits further comments. As stated earlier, the lower arm blood flow was very low, after 1 hour in water. Moreover, the magnitude of the heat flux did not appreciably decrease even when the arterial circulation of the lower arm was occluded at the end of 3 hours of immersion (15, 35). This implies that in the cooler water, metabolic heat produced in the limbs is conducted directly to the skin surface to be lost to the water environment at a rate equal to metabolic heat production. The limb circulation evidently contributes little to limb heat loss in water of critical temperature although it may play an appreciable role in delivering heat to the skin surface in warmer water. In contrast, the heat flux from the finger was entirely dependent on the blood flow, probably because local metabolism plays little role as a source of heat. Since the finger blood flow decreases drastically in cold water, heat transfer from these most distal areas can be disregarded insofar as body temperature is concerned in all water temperatures cooler than 35 C.

TABLE 4. Average limb blood flow[a] during the last 30 min of 2 hour immersion in 30, 31, and 33C water.

Region	Water Temp. °C	Ama	Control	P value
		August 1966 (*Summer*)		
Lower arms	30	1.68±0.98[b]	2.04±0.93	NS[c]
	31	2.13±0.72	1.78±0.61	NS
	33	2.13±0.36	3.11±0.77	NS
Finger	30	6.48±0.81	3.40±0.57	<0.025
	31	7.20±1.28	3.72±0.83	<0.1
	33	19.02±3.10	11.82±2.32	<0.1
		January 1967 (*Winter*)		
Lower arms	30	3.00±0.50	2.19±0.40	NS
	31	4.09±0.34	2.89±0.85	NS
	33	5.10±0.91	4.16±0.61	NS
Finger	30	9.91±2.11	5.69±0.61	<0.1
	31	10.23±1.78	5.43±0.83	<0.05
	33	11.50±2.17	8.19±1.57	NS

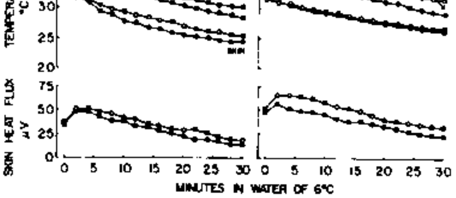
[a] ml/min per 100 ml. [b] Values are means ± SE. [c] NS indicates the nonsignificant difference between the two groups ($P > 0.05$).

VASCULAR RESPONSES TO HAND IMMERSION IN COLD WATER

In the past, many investigators compared the vascular responses of presumably cold-adapted subjects to immersion of one hand in ice-cold water and reported a significant attenuation of vasomotor response in these subjects. When a hand is immersed in ice-cold water, the return of cooled venous blood from the hand will decrease the temperature of lower arm. More specifically, the cooled blood returning from the hand via deep veins would lower the muscle temperature while that returning via superficial veins would lower the skin temperature of the lower arm. Moreover, the overall rate of cooling of the lower arm would be determined by the local skin and hand blood flow as well as by the rate of local heat production.

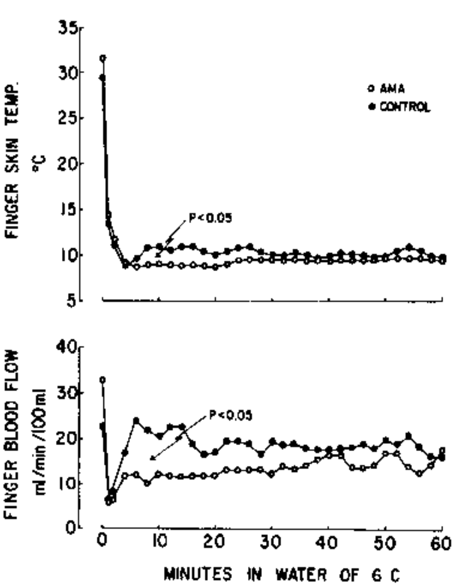
To study this problem, we conducted a series of studies in the ama during immersion of one hand in 6 C water for 30–60 min (32). As shown in Fig. 14, the skin temperature of the lower arm decreased in a similar manner in both the ama and nondivers while the muscle temperature decreased at a slower rate (especially in winter) in the ama as compared with nondivers. The most likely explanation for this finding is that the local heat production in the lower arm muscle is higher (especially in winter) in the ama, as suggested by higher BMR in these divers (see Table 1). In addition, this finding may also suggest that a smaller fraction of cooled blood returns via deep veins in the ama as compared with nondivers, and/or that the hand blood flow is maintained lower in the ama than in nondivers. Actual measurements of finger skin temperature and blood flow indeed indicated that these values are significantly lower in the ama (Fig. 15). Although the water temperature was not low enough to induce classic "hunting" reactions, there were definite signs of hunting in nondivers, but not in the ama, during immersion. These results clearly prove that the degree of vasoconstriction of finger blood vessels in response to hand immersion in 6 C water is significantly greater in the ama than in nondivers. When venous return from the hand was occluded, there was no appreciable change in the rate of reduction of lower arm muscle temperature while the respective skin temperature increased in the ama and leveled off in nondivers. This result also supports a view that a greater fraction of hand blood flow returns via superficial veins (or a smaller fraction via deep veins) in the ama than in nondivers.

Figure 14. Lower arm muscle and skin temperatures and skin heat flux during hand immersion in 6 C water. (From Paik (32).)



These findings from the ama are at variance with many previous reports. For instance, Brown et al. (1) showed in the Eskimo a faster reduction in the muscle temperature coupled with a higher blood flow in the lower arm and in the hands, which they attributed to a greater fraction of venous return via deep veins. The magnitude of pressor re-

Figure 15. Finger skin temperature and blood flow during hand immersion in 6 C water. (From Paik (32).)



sponse and of reduction in the finger temperature during hand immersions are known to be considerably less in Gaspé fishermen (27, 28), fish-filleters (31), Arctic Indians (4), Lapps (26), and Eskimos (30). However, these differences in findings may be less disturbing if one considers the basic difference in the type and degree of exposure to cold. In contrast to fishermen and Eskimos in whom only the hands are exposed to either cold water or air, the ama are totally exposed to cold water. It is highly conceivable that the type of adaptation to local exposure to cold is basically different from that of whole-body exposure. Recently, Hampton (6) reported that prolonged exposure to cold in the Antarctic initiates a more sensitive response to local cooling of the hands, while Skreslet and Aarefjord (38) also noted a reduced blood supply in the periphery following prolonged cold water exposure in scuba divers, in general agreement with our finding on the ama.

SUMMARY

Results described above may be summarized as follows:

1) The ama are daily exposed to severe cold water stress which limits both the duration of each work shift and the total working time. While the rate of heat loss to the surrounding water is approximately two times greater in winter than in summer, the estimated total extra heat loss is in the order of 1,000 kcal/day throughout the year.

2) There is a consistent, reversible increase in BMR of ama in winter when the water temperature is lowest. Nondiving women fail to show such a seasonal variation. This increase in BMR in winter (i.e., metabolic adaptation) is not associated with an increased amount of thyroid ^{131}I uptake but most likely with increased utilization of thyroid hormone by peripheral tissue.

3) There is a very small (but significant) increase in oxygen consumption in response to exogenous norepinephrine in the ama in winter but not in summer This indicates that the development of nonshivering thermogenesis is not the main feature of cold adaptation in the ama.

4) The critical water temperature in the ama at a comparable subcutaneous fat thickness is significantly lower than in nondivers in both summer and winter In other words, the ama are able to maintain a larger temperature gradient from body core to body surface. This type of adaptation (i.e., insulative-hypothermic adaptation) may represent habituation.

5) The maximal thermal insulation in the ama at a comparable subcutaneous fat thickness is significantly greater than in nondivers in both summer and winter. However, the magnitude of regional heat flux is not different between the ama and nondivers.

6) During 2- to 3-hour immersion in water of critical water temperature, the heat flux from the limb (especially the finger) for a given blood flow tends to be lower in the ama as compared with nondivers in both summer and winter. This suggests that the ama may have a more efficient countercurrent heat exchange system in the limbs.

7) During immersion of one hand in 6 C water, the lower arm muscle temperature is maintained higher in the ama than in nondivers, especially in winter. Moreover, both the finger skin temperature and blood flow are maintained lower in the ama than in nondivers, suggesting the development of vascular adaptation in the ama.

It is thus concluded that the ama definitely develop various types of adaptation to cold as a result of repeated exposure to cold water stress. I fully realize that certain conclusions in our studies are not fully substantiated and are at variance with results obtained from other presumably cold-adapted men. However, one should also realize that our effort is clearly limited by the lack of certain critical techniques which can be applied to nonmotivated subjects in remote field laboratories.

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Acoustic telemetry from human divers

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Kanwisher, J., K. Lawson, and R. Strauss. 1974. Acoustic telemetry from human divers. Undersea Biomed. Res. 1(1): 99-109.–Acoustic telemetry can be applied to human divers to assess the physiological stresses of diving in open water. This method provides data in real time as contrasted to recording on the diver for later playback. The report describes the use of constraints on sound as a means of transmitting ekg and respiration data; discusses the design and construction of sample devices for transmitting, receiving, and interpreting the data; and finally, shows how these devices have been applied in actual human diving research:

telemetry/acoustic
physiological monitoring
divers

Acoustic telemetry has been used routinely for over a decade to monitor physiological parameters on free-ranging aquatic animals such as porpoises, fish, and turtles (Kanwisher and Sundnes 1967; Kanwisher and Lawson 1974). It is shown here how the method can be applied to human divers to assess physiological stresses of diving under more realistic conditions than those possible in pressure chambers.

Telemetry also provides data in real time as contrasted to recording on the diver for later playback. Monitoring by surface personnel allows the diver to be recalled to the surface

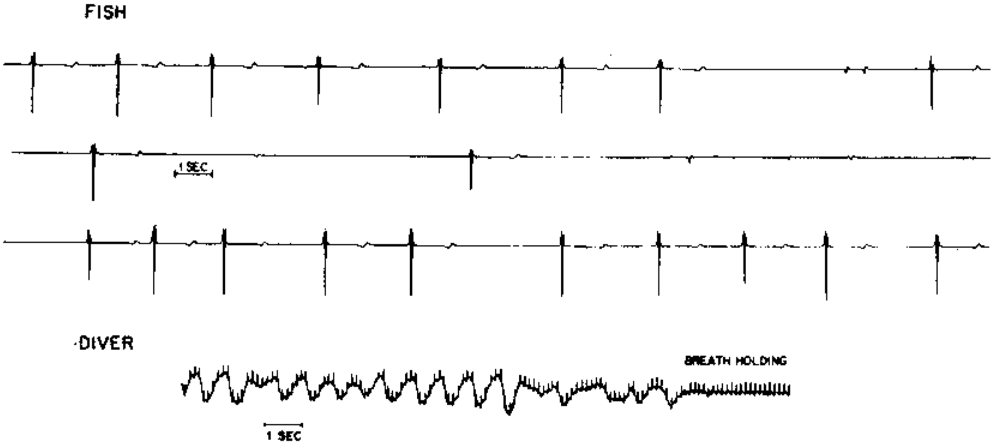

Fig. 1. EKG from free-swimming fish, and EKG plus respiration from diver. The fish shows severe tachycardia from fright.

when physiological limits are exceeded. Alternatively, the diver can be cautioned to *go slow*. This monitoring removes the need for the diver (who might be distracted by the task at hand and prideful about his professional capabilities) to take all responsibility for his own well-being. Hopefully, the method of monitoring by telemetry will add to the safety of what is, at best, a dangerous task.

Continuous improvements in electronic techniques now permit construction of acoustic transmitters the size of a little finger. These will send data such as heartbeat and temperature (from a free-swimming fish) over ranges of several hundred meters for as long as a month (Kanwisher and Lawson 1974). An example of an EKG from a codfish is shown in Fig. 1. Use of such transmitters on humans has awaited motivation, not development of the method. In the development of closed-circuit diving gear, investigators have monitored EKG and P_{O_2} extensively. This report describes the use of constraints on sound as a means of transmitting such data; discusses the design and construction of sample devices for transmitting, receiving, and interpreting the data; and, finally, shows how these devices have been applied in actual human diving research.

SOUND AS A TELEMETRY MEDIUM

Sound is the only practical form of energy for telemetry for ranges beyond a few meters through water. It travels with little loss, whereas radio waves and light are rapidly absorbed. Several properties of sound in water are important. For example, greater ranges are possible in fresh water than salt (one rarely has a choice in this); also, low frequencies transmit farther than high frequencies. For ranges up to several hundred meters, any frequency below 100 kHz is suitable. If a range of several kilometers is needed, the frequency should be less than 20 kHz; however, low frequencies involve longer wavelengths, which implies larger transducers, and in small devices these are difficult to use. Thus, frequencies employed most often are between 30 and 80 kHz. To monitor large bluefin tuna, a transmitter big enough to work efficiently at 20 kHz was used; it alternately sent tissue- and water-temperature transmissions with an open-sea range of 8 km. Such a fish has been followed in the open sea for a distance of 200 km over a period of several days (Carey et al. 1971).

Sensitivity of the divers' ears limits the use of low-frequency telemetry. For example, one 20-kHz EKG transmitter putting out 0.25W proved objectionably painful; for this reason it is doubtful whether human instruments will be tolerated below 30 kHz. This is a fortunate coincidence. A tubular piezo-electric transducer, the most convenient engineering compromise for generating sound, is about 4 cm in diameter at this frequency. The cylindrically cast instruments used in these experiments were most convenient in this size range.

Finally, interfering background noise, which tends to obscure the signal, varies greatly at different places in the water. In general, the shallow-water tropics are noisiest. At Coconut Island in Hawaii the natural acoustic energy may be 100 times greater than that at Friday Harbor in Puget Sound. Most of the noise appears to be from bottom animals such as snapping shrimp. Man-made noise (e.g. noise from boat motors or airflow in the diving gear itself) can also be troublesome.

These observations are meant to make one's ambitions more modest when considering acoustic telemetry, particularly for work in the open sea. It is not possible to send across oceans with a miniscule device. But almost any small amount of energy will work in laboratory tanks if they are acoustically quiet at the frequency used.

POWER REQUIREMENTS

An acoustic transmitter ideally sends out sound in all directions. At some remote point a receiving hydrophone intercepts the sound and feeds a converted electrical signal into a receiver. A good signal-to-noise ratio is obtained when the electrical amplitude coming from the hydrophone is 1 μV or greater. Since hydrophones used in these experiments have a typical resistance of 1,000 Ω, the amount of acoustic power intercepted to produce this signal is 10^{-15}W ($P = E^2/R$). This low minimum-detectable energy is the key to the successful functioning of the acoustic links. As an example, one of the smaller fish transmitters may put out 100 μW (10^{-4}W) of acoustic power. A 10%-efficient receiving hydrophone must *see* only 10^{-10} of this for such a transmitter to be heard, which accounts for the impressive ranges that small amounts of power allow under ideal conditions. Size, and therefore power, can be greatly increased on an instrument to be carried by a diver. One thousand times the above amount (100 mW) was minimally used in these experiments; this does not, however, lead to 1,000-fold increase in range, because of the quadratic decrease in power density with distance.

TYPES OF MODULATION

It is necessary to encode the parameter (such as EKG) to be transmitted. This is done most routinely by transforming the electrical signal into analogous frequency variations (FM). At the receiver these changes are converted back to the original electrical form and recorded on a pen writer. Such frequency modulation is fortunately the simplest to accomplish technically. It requires only sufficient frequency excursion to swamp out the small

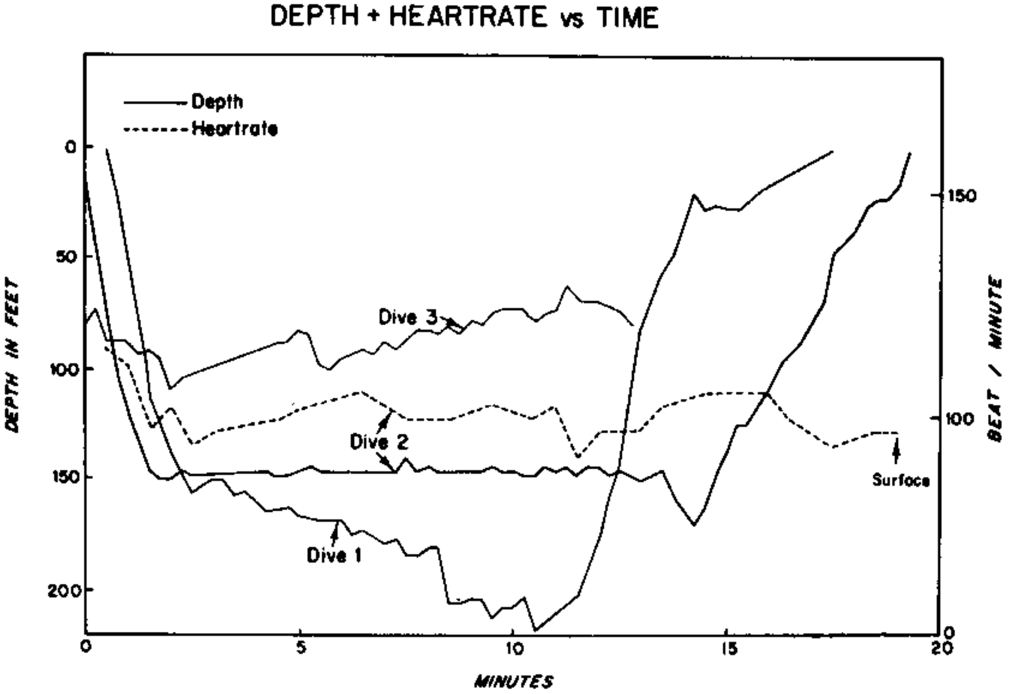

Fig. 2. Dive time versus depth and heart rate for scuba divers. Note the low heart rate during Dive 2.

amount of frequency jitter that is inherent in the multipath nature of most real acoustic situations in the water—usually below 0.1%.

Where information is needed infrequently, such as with the body temperature of a diver, the rate at which short pulses of sound are sent can be varied. The number of pulses can be counted over a period such as a minute and this number can be referred to a calibration curve to determine the temperature. Values of body temperature received once a minute provide sufficient detail to the time changes of temperature in man.

Another method of temperature encoding is to change the value of the transmitter's carrier frequency by means of a thermistor in the electronic circuit. Excursions about this slowly changing frequency can then be used to convey the EKG, in addition to temperature. At the receiver the two variables are represented by different frequency bands. Temperature is carried in the region below 0.1 Hz; the EKG is carried by modulation frequencies between 10 and 100 Hz. The two are easily separated by filtering. In this way two-channel transmission is simply accomplished.

Still another method of information encoding is to use time instead of frequency. The diving-depth transmitter sends an index pulse every 15 seconds; at a variable time after this, a second pulse is transmitted (PPM). The time is made proportional to the depth. A factor of 20 fsw in depth is used for every second of delay, and the time at the maximum calibrated depth of 200 fsw is set at 10 seconds. This still leaves a 5-second dead period before the next index pulse. If a diver goes below 200 fsw, the information pulse will be in this interval (Fig. 2).

RECEIVING

The receiving hydrophone used in these experiments has an amplifier (40 dB or 100 times voltage gain) built into it. The higher level signal in the connecting cable inhibits interference from ignition noise and radio transmitters. The hydrophone connects to a tunable superheterodyne receiver (Fig. 3); a mechanical filter at 455 kHz sharply defines the bandpass. This intermediate frequency signal is mixed with a beat frequency oscillator to produce an audible tone, and variations in the original transmitter frequency are translated to changes in the frequency of this tone. Heart rate can be determined readily by counting the distinctive chirps due to the QRS complex in the EKG. With practice the T-wave is also discernible, as well as unique events such as extra systoles. A simpler receiver that gives nearly equivalent performance has been described by Kanwisher and Lawson (1974). A directional hydrophone allows the surface monitor to keep track of a diver's position.

A written record of the EKG requires the frequency excursions to be translated into voltage changes. A discriminator working at the audible frequency has been used, but cleaner records have been obtained (Fig. 1) by using a phase-lock circuit at the intermediate frequency. In general, an audible heartbeat is easier to count, but more significant cardiac detail can be seen with a strong signal producing a clean, written record than can be interpreted from listening.

For written records in the field a phase-lock receiver set directly at the 31-kHz frequency of the EKG transmitter is built into a battery-operated pen writer. An example of a record showing respiration and heartbeat from a diver is shown in Fig. 1.

ELECTRONIC DESIGN

Any apparatus for use on scuba divers working from a small open boat is exposed to severe physical abuse. Such operating conditions also tax the capabilities of the person at the surface monitoring the diver. Both of these factors reward operational simplicity. The equipment presented here is that which has survived the attrition of past failures.

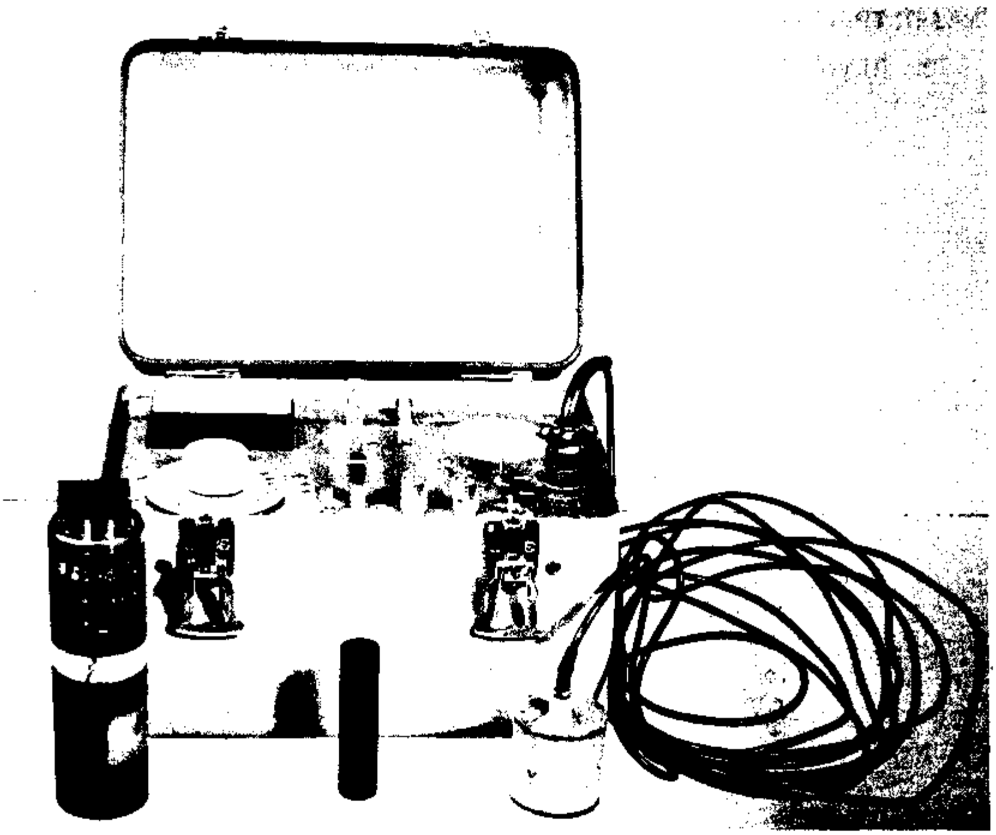

Fig. 3. Receiver plus hydrophone and also respiration and heartbeat transmitter. A fish transmitter is shown for comparison.

DESIGN PHILOSOPHY

Modern solid-state electronic components allow increasing circuit complexity with reasonable limits of size, battery drain, and cost. Proven reliability of such components gave the investigators confidence to cast finished circuits in epoxy resin instead of mounting them in a watertight case. The circuits are powered by sealed rechargeable nickel-cadmium batteries. Two metal pins, projecting from the plastic, function as the seawater switch and as charging terminals for the battery. Such instruments have functioned with good reliability to depths of over 100 msw. There is no possibility of repairing such instruments, but there has been little need to do so.

Complexity per se is rarely sufficient reason for rejecting an electronic-circuit approach to realize a certain functional end in the equipment. For versatility the investigators have used a combination of digital and analog elements. Digital is attractive because of the lower power possibilities provided by COSMOS, and analog is greatly facilitated by devices such as low-power operational amplifiers. The particular solid-state elements used undergo continuous change as better components appear on the market. An example of a significant recent improvement would be in bilateral switching, which has been improved by COSMOS gates.

DEPTH TRANSMITTER

The delayed-pulse depth transmitter is designed to give a signal which can be interpreted easily with only a receiver and a wrist watch. It uses many of the above circuit functions and is an example of the complexity allowed. The entire circuit occupies nine 1½-in. diameter printed-circuit boards, assembled in a stack before casting. The battery capacity is sufficient for 100 hours of operation.

The 15-second index pulse is generated by frequency division from a crystal oscillator. Various gates are needed to give this pulse the proper length; these are provided in an integrated circuit made for an electronic wrist watch. This circuit alone contains the equivalent of hundreds of transistors.

The measuring circuit itself is designed to convert the output of a silicon semiconductor strain-gauge bridge to a time delay. Its operation is initiated by the index pulse. Upon receipt of this trigger, the circuit integrates a constant current until the integral is equal to the amplified output voltage of the bridge. Since this voltage is linearly related to pressure, the time necessary for the integration is also. At the completion of this integration another pulse is generated. Because the integrating current is supplied from the same supply voltage that powers the bridge, the circuit is essentially independent of operational amplifier voltage and current offsets, and offset drifts. It is also immune to power-supply changes between 8 and 15 volts.

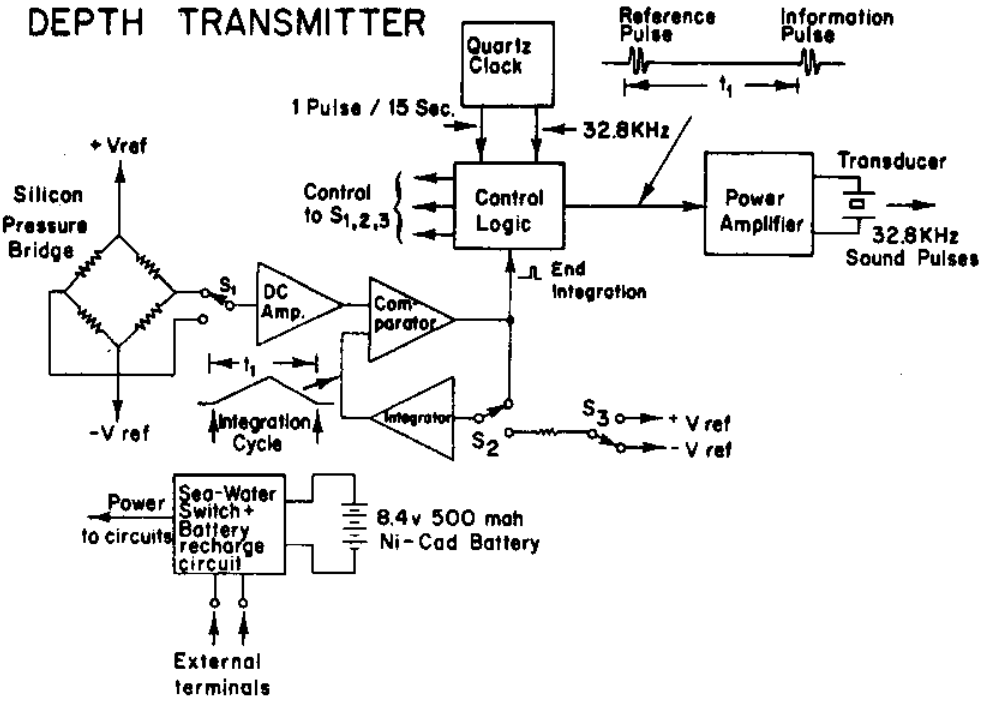

Fig. 4. Functional diagram of diver-depth transmitter circuitry.

The next index pulse, after 15 seconds, starts the integration over again whether it has been completed or not. This happens in the circuit used in these experiments if the depth is greater than 300 fsw. In such a case there will be no information pulse generated. Any malfunction in the measuring circuit still allows these index pulses to be generated.

The index and information pulses, which last 30 and 100 milliseconds (msec) respectively, are used to turn on the depth transmitter. The output of the timing oscillator (32.768 kHz) is gated to a power amplifier by these pulses. The power is 2 watts (W). A block diagram and some of the waveforms are shown in Fig. 4.

The 15-second interval between the index pulses is determined with crystal precision (.001% over a range of 10°-25° C). As the diver moves farther from the boat the pulse will arrive later because of the added time of sound traveling from the diver to the boat; at 1 mile the pulse will be approximately 1 second late. Even though this time delay is affected by both water temperature and salinity, it can still be used to determine the diver's distance from the boat. Accumulated time-error over 1 hour for a reasonable oscillator is less than 36 msec, which represents a distance error of 180 ft. To make use of the potential accuracy in this technique it is necessary to use an electronic method of time determination. As the diver leaves the boat the oscillator located in the boat is synchronized with the one worn by the diver. Its pulse is then used to start counting pulses from the oscillator worn by the diver. The received index pulse stops the count and the number of cycles tells the distance to the diver. Depth is judged most accurately by the time between the two pulses from the diver's transmitter, since the information pulse is delayed in the same way that the index pulse is delayed.

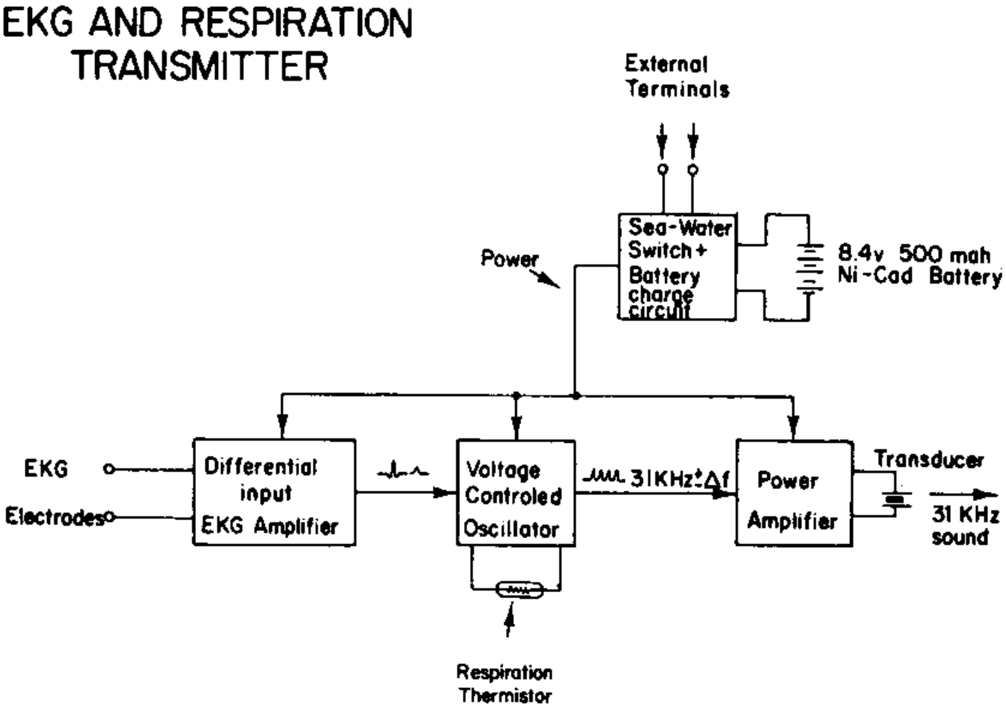

Fig. 5. Functional diagram of diver EKG and respiration transmitter.

EKG AND RESPIRATION TRANSMITTER

It has been mentioned how frequency modulation is the simplest method of transmitting low-frequency information. For heartbeat and respiration a composite modulating waveform is generated. The human EKG is generally 1 to 2 mV in amplitude, and is amplified 100 times in a circuit that strongly rejects components in phase at the two electrodes. Such common-mode rejection helps eliminate unwanted muscle potentials due to exercise.

Respiration is sensed with a thermistor in the scuba mouth-piece; the thermistor senses air-temperature fluctuation from inhalation and exhalation. Amplitude of this respiration signal is kept below that of the EKG; the ear can readily count the breathing and the heartbeats separately.

The oscillator modulated by this composite waveform is an astable multivibrator controlled at the common-base return; its stability is not critical. It can directly drive a power amplifier connected to a transducer. As in the depth transmitter, a frequency near 30 kHz is used, which works well with a 4 cm-diameter lead zirconate piezo-electric transducer. The seawater switch and charging electrode is also identical to that of the depth transmitter. The thermistor and EKG electrodes go to a watertight connector on one end (Fig. 3). The received composite signal from a diver is shown in Fig. 1 and the circuit in Fig. 5.

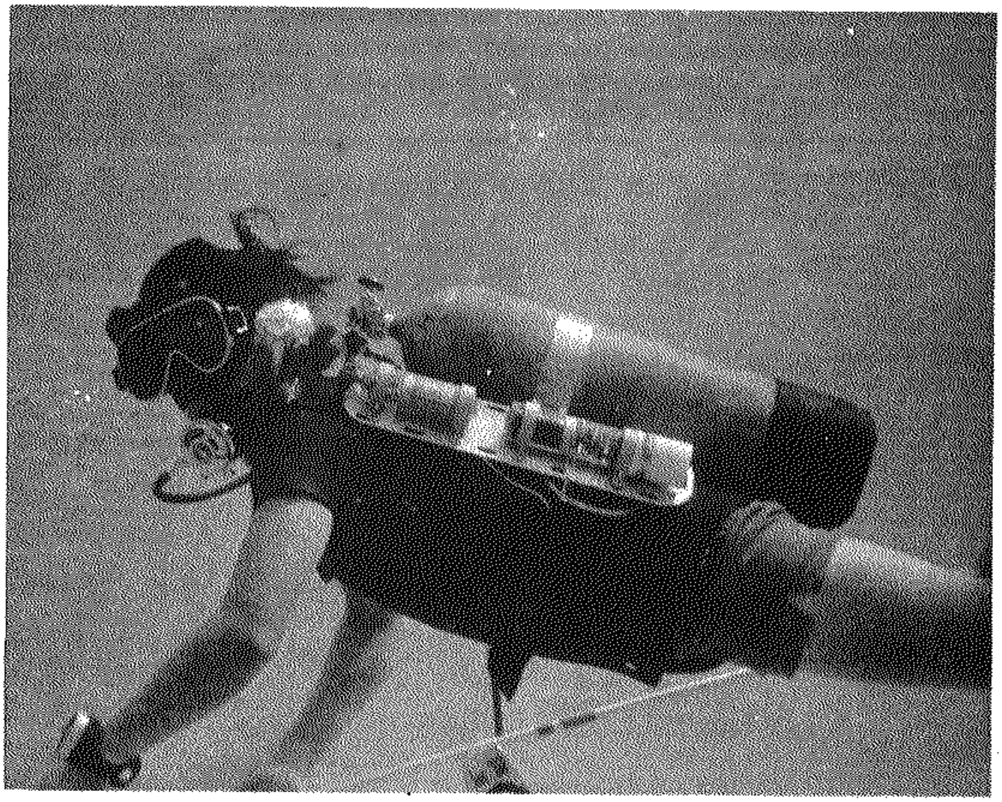

Fig. 6. Underwater photograph showing two separate transmitters alongside the scuba tank.

METHODS AND RESULTS

SCUBA DIVING

Equipment described here was used to study a group of Hawaiian fishermen, who used scuba gear for spear fishing and trapsetting at depths of 200 fsw or greater. The fishermen were experienced divers, yet they apparently did not follow recommended decompression procedures and they seldom were able to give precise information on the depth history of a dive. In this study depth and heartbeat were telemetered with the acoustic transmitters shown in Fig. 6. A diver proceeded with his diving in a normal fashion after the EKG electrodes had been applied.

To enable investigators to listen from the boat, the receiving hydrophone was dangled a few meters beneath the surface, which reduced boat and motor noises to a tolerable level. It is possible to hear and count heartbeats in the presence of considerable interference since the EKG produces a distinctive signature. Most prominent is the excursion due to the QRS complex, which can readily be counted. A feeling of rhythm makes it easy to sense the large changes that undue strain produces. The heart rate can be accurately counted later, on shore, by tape-recording the receiver acoustic output. The tone from the beat-frequency oscillator receiver is left on continuously when a diver is down; large changes in pitch reflect the

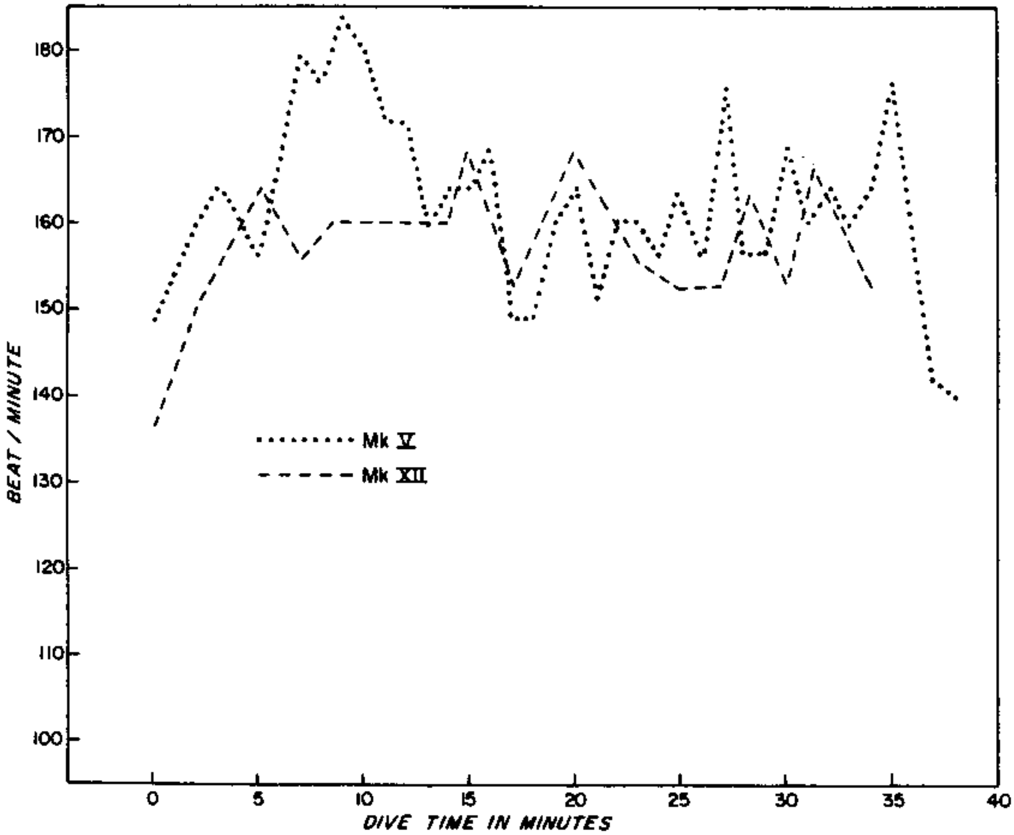

Fig. 7. Dive time versus heart rate with Navy standard (Mark V) and prototype (Mark XII) hardhat diving rigs. Note the 184 beats/min after 10 minutes with the Mark V. (From Armstrong et al. 1974.)

waveform of the diver's EKG.

The depth and heart rate curves in Fig. 2 illustrate the type of data collected using this method. Dive 2 in particular shows the rapid rate of ascent that these fishermen sometimes use. During this dive the fishermen's heart rate stayed in the neighborhood of 100 beats/min. Such a diver is clearly at ease in the water. The low rate implies a low cardiac output. This may contribute to a smaller amount of gas in the tissues, which, in turn, allows a more rapid decompression.

HARDHAT DIVING

In some recent hardhat diving tests (Armstrong et al. 1974) the decision was made to use telemetry rather than a handwire connection. Past experience had shown that it was awkward to maintain the integrity of an electrical umbilical cord. In the tests a diver was required to perform standard work tasks while his physiological stress was monitored. The sound transmitter was put in a pocket on the diving suit and the EKG leads were run through the cuff and up the sleeve.

Data in Fig. 7 indicate that a diver in an improved diving rig can perform standard work with less strain. The maximum heart rate of 184 beats/min must be close to maximum for the diver, which demonstrates the value of having real-time data during a dive. A diver would probably not be allowed to work very long at such a rate.

CONCLUSIONS

For over a decade the investigators have routinely done physiological monitoring of free-swimming animals by acoustic telemetry. In spite of its seemingly obvious advantages for human divers there has been a reluctance among many investigators either to believe it is practical or even possible. The value of such a method is presented here. The equipment involves straightforward and not particularly complex engineering, which should be easily realizable.

Unsatisfactory telemetry results are usually due to the complex nature of the acoustic path that exists in most real situations. As an example, a few feet of sea weeds in shallow water can introduce 40 dB of acoustic loss. However, in nearly all situations where the acoustic path is under 1,000 yards, it is possible to get acceptable results. Such a loss, for instance, can be offset by using a directional receiving hydrophone, pointed for maximal signal, which provides the additional advantage of locating the diver.

A wide variety of data has been transmitted acoustically. It is only necessary to transmit body temperature occasionally, such as with depth. Electroencephalograms are analogous to electrocardiograms in their frequency components, but 25 times more gain in the signal amplifier must be added because the voltages are smaller. It is even possible to send 1-msec nerve spikes from a microelectrode in the cerebellum of free-swimming fish (Kotchabhakdi et al. 1973). The bandwidth to accomplish this would require 10 times that used for EKGs and is probably close to the maximum capability in frequency response of the technique.

This study, contribution No. 3278 from the Woods Hole Oceanographic Institution, was supported by National Science Foundation Grant GA-31987. Professor S. K. Hong provided the welcome opportunity to initiate the free-diving tests. The authors are also indebted to Ed Hayashi whose skill in the field contributed importantly to these measurements. The enthusiasm of two skilled professional divers, Kenneth J. Conda and Frederick W. Armstrong, Jr., encouraged the authors to make equipment suitable for the military hardhat tests. More complete engineering and acoustic details will be supplied in a forthcoming manual.

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ANOMALOUS VENTILATORY RESPONSES OF DIVERS

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ABSTRACT

Results are presented from a study comparing ventilatory responses to treadmill exercise in divers, sedentary non-divers and non-diving runners. These results indicate a lower ventilatory response to exercise, with consequent CO_2 retention, in divers. This finding appears not to be fitness-related. The divers also exhibited a markedly slower and deeper breathing pattern than the other two groups. Results from two other studies comparing exercise in air with exercise in water in divers and non-divers indicated that divers shifted to a slower, deeper breathing pattern underwater than in air whereas non-divers did not, and that divers tended towards lower ventilations underwater than in air whereas non-divers did not. We interpret these results to imply the existence of conditioned responses in divers; diving-related stimuli elicit "air-conserving" ventilatory behavior.

INTRODUCTION

For many years, physiologists have been interested in the respiratory effects of hyperbarism, particularly in the context of diving. This interest intensified when Schaefer (1955) reported in 1955 that submarine escape tank instructors (who engaged in extensive breath-hold diving) exhibited CO_2 response curves with slopes about 30% less than found in non-divers, and that they showed an unusually slow, deep breathing pattern. Song et al. (1963) reported that the diving women of Korea also had a low ventilatory response to CO_2 (compared to non-diving Korean women), but that their response to hypoxia was normal. Schaefer (1965) subsequently reported that the characteristics of reduced CO_2 response and slow, deep breathing pattern in escape tank instructors diminished significantly after several months of non-diving and returned when they resumed diving. He also reported that active divers had a lower response to hypoxia. It seems clear that breath-hold diving influences control of ventilation.

This may also be the case in SCUBA divers. Goff and Bartlett (1957) found that experienced underwater swimmers showed end tidal P_{CO_2} values that were 9-14 mm Hg higher than inexperienced subjects, suggesting reduced chemosensitivity in the divers. In contrast, Froeb (1960) compared 16 highly experienced divers and 16 non-divers, and found no difference between the two groups in their response to exogenous CO_2, either at rest or during exercise. However, Lanphier (1963) reported finding end tidal P_{CO_2}'s of 50 mm Hg in a group of divers performing moderate exercise underwater at 4 ATA. The more pronounced "CO_2 retainers" in this group were found to show marked increases in Pa_{CO_2} even during moderate exercise in air at 1 ATA on low-resistance breathing apparatus.

The purpose of this communication is to report some observations that we have made concerning the ventilatory response to exercise of divers (compared to non-divers) both in air and underwater. These observations suggest that the slow, deep breathing pattern and reduced ventilatory responsiveness in breath-hold divers are also typical of SCUBA divers.

METHODS

The data to be presented here were accumulated during three separate studies. Two studies compared cardio-respiratory responses to exercise during immersion in water of various temperatures and one study was specifically aimed at comparing the ventilatory response to treadmill exercise of divers and two groups of non-divers.

Of the first two studies, one employing non-divers as subjects has been reported by Moore et al. (1970) and the second, employing divers as subjects, has been reported by Lally et al. (1971). Both of these studies used the same exercise machine, the same levels of exercise, the same breathing apparatus, and had two water temperatures in common, so that results from these two studies may be readily compared. The exercise was graded leg exercise on a submersible ergometer described by Moore et al. (1970), in which a pulley system is used to lift variable weights repetitively by horizontal power strokes of the legs. The loads employed were 25 lbs/leg 60 strokes/min ("heavy"), and 35 lbs/leg 100 strokes/min ("forced"). These work loads corresponded to oxygen costs (exercise minus resting) of approximately .70, 0.96 and 1.28 liters/min respectively.

In both of these studies, air was supplied to the subjects from standard SCUBA tanks, using a double-hose regulator with standard mouthpiece-valve assembly (U.S. Divers) for experiments both in air and underwater. During experiments in air, mixed expired air was collected directly from the exhaust hose in rubberized canvas (Douglas) bags. During experiments underwater, the expired air was trapped over water and, again, collected in Douglas bags, using the very simple system shown in Figure 1. Volume of gas collected was determined by emptying the bags through a dry gas meter (American Meter Co.) which had been calibrated against a 350 liter Tissot spirometer. All gas collections were made after the attainment of a steady state, as judged by a plateau of heart rate.

The third study, comparing ventilatory responses to treadmill exercise in divers and non-divers, has been reported by Lally et al. (1974). The subjects were 8 divers, 9 sedentary non-divers ("controls") and 11 non-diving distance runners. The exercise was walking up a 10% grade at speeds of 1.6, 3.2, and 4.8 km/hr (1, 2, and 3 mph). Each experiment consisted of a 7 minute walk at each of the three speeds with rest periods in between. During the 2 minutes immediately preceding each walk, $\dot{V}_E$, $\dot{V}_{O_2}$, and $\dot{V}_{CO_2}$ were measured by the open circuit method with the subject at a standing rest. The expired air was analyzed by the Scholander method (1947). These measurements were repeated over the last 2 minutes of each walk. The steady-state $\dot{V}_E$ response was measured as the difference between these two $\dot{V}_E$ values.

Inspiratory flow was also measured via a screen pneumotach and differential pressure transducer (Statham PM 197). The flow signal was integrated to

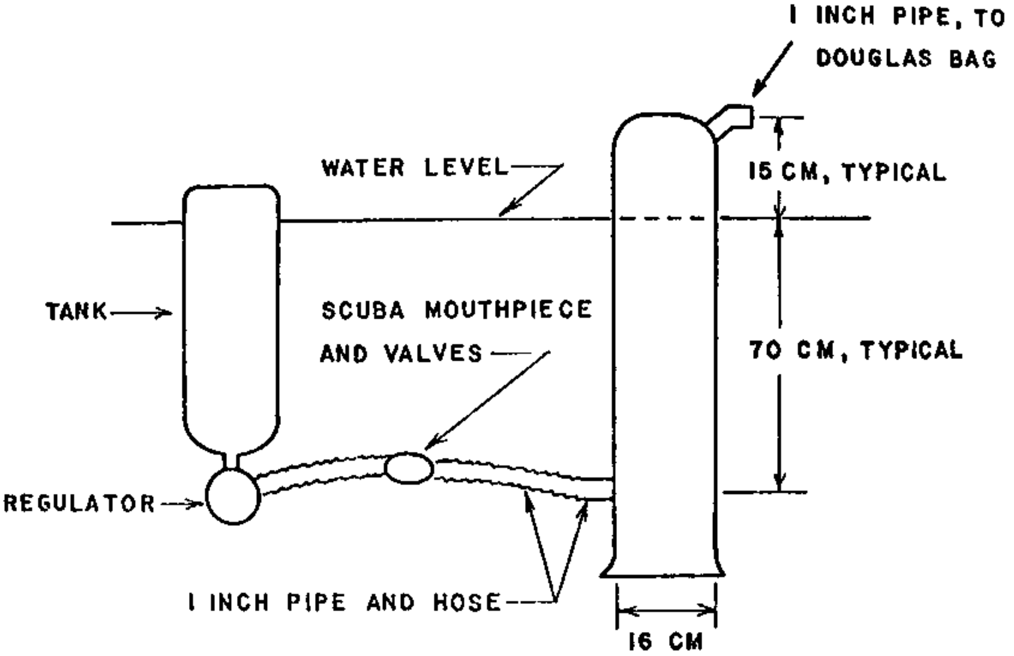

Figure 1. Schematic of the breathing apparatus used in experiments comparing exercise in air and underwater. With the subject underwater, expired air was trapped over water and collected in Douglas bags; with the subject in air, the expiratory hose was extended directly to Douglas bag.

provide breath-by-breath inspired tidal volume. Expired air was continuously sampled from the mouthpiece and analyzed for CO_2 (Beckman LB1). In all experiments, the subjects breathed through a standard SCUBA mouthpiece-valve assembly and hosing (U.S. Divers). The expired air was collected in rubberized canvas bags, which were then emptied through a dry gas meter.

As mentioned above, the steady-state $\dot{V}_E$ response was measured as the difference between the resting $\dot{V}_E$ and the $\dot{V}_E$ during the last 2 minutes of exercise. The initial response was also recorded, as an estimate of the neurogenic component of the exercise hyperpnea. This was measured as the $\dot{V}_I$ increase observed during the first 15 seconds (approximately) of exercise, as determined from the breath-by-breath inspired volume record. The exact time interval over which the initial response was measured depended on the time required to complete an integer number of ventilatory cycles. In no case was an inspiration counted if the preceding expiration exhibited an increase in end-expiratory CO_2. Due to the breath-by-breath variability of tidal volume, the number of observations of the initial response was increased by including two or three short walks of 15 - 20 seconds duration prior to each 7 minute walk. Evidence that this initial response is an estimator of the neurogenic ventilatory drive has been provided by Asmussen and Nielsen (1948), Dejours and his colleagues (1955, 1959, 1963), and D'Angelo and Torelli (1971).

The results presented from this study are based on complete data on exercise at all three speeds obtained in 13 experiments on 8 divers, 17 experiments on sedentary non-divers and 20 experiments on 11 runners. The V_{O_2} and V_E data have been normalized by body weight.

RESULTS

We would like to present the results of the third study first. These results are summarized in Table and Figure 2. As one can see, the steady-state ventilatory response of the divers was less than that of the control group at all three speeds; significantly so at 3.2 and 4.8 km/hr ($P < 0.01$). At the two higher speeds, the decrements in $\dot{V}_E$ (from controls to divers) amounted to 20% of the control values. The initial ventilatory responses were also less in divers, but not significantly so ($P < 0.1$ at 4.8 km/hr). The decrements in the initial response contributed 35 - 40% of the total (steady-state) decrements.

Groups	Rest	1.6 km/hr	3.2 km/hr	4.8 km/hr
		$\dot{V}_{O_2}$ (ml/kg min)		
Controls	4.31±0.14	11.27±0.30	17.32±0.28	24.14±0.38
Divers	4.02±0.19	11.21±0.34	17.00±0.31	24.33±0.43
Runners	3.99±0.08	10.87±0.23	16.68±0.30	22.62±0.40
		Steady state $\dot{V}_E$ (L/kg min)		
Controls		0.140±0.005	0.268±0.007	0.455±0.066
Divers		0.134±0.006	0.215±0.010	0.367±0.017
Runners		0.135±0.009	0.243±0.008	0.382±0.014
		Initial $\Delta\dot{V}_I$ (L/kg min)		
Controls		0.101±0.008	0.122±0.016	0.163±0.017
Divers		0.102±0.019	0.104±0.009	0.126±0.010
Runners		0.093±0.011	0.118±0.011	0.140±0.010

Table 1. Oxygen uptakes and ventilatory responses.
Values are mean ± SE

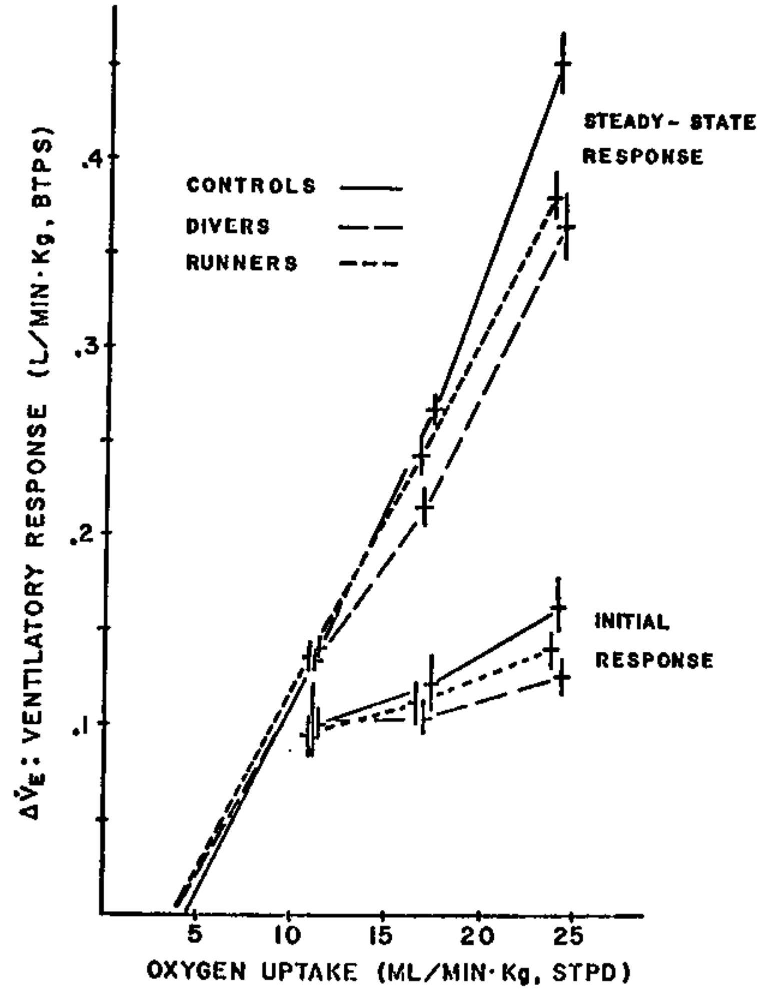

Figure 2. Steady-state and initial ventilatory responses to treadmill exercise of the three groups of subjects. Ventilatory responses and oxygen uptakes are normalized by body weight.

As one would expect, this 20% reduction in $\Delta\dot{V}_E$ resulted in an elevated P_{ACO2} (estimated from end-expiratory P_{CO2}) and CO_2 retention as the exercise progressed, as indicated in Table 2. The divers had a significantly higher P_{ACO2} at 3.2 and 4.8 km/hr and a significantly lower R at 4.8 km/hr, as measured from the fifth to seventh minutes of exercise.

Examination of the breathing patterns (Table 3) reveals that the breathing frequency is uniformly lower in divers while the tidal volume is significantly higher in all cases.

The divers also exhibited a lower heart frequency than controls at similar values of oxygen uptake (Figure 3). We interpreted this as an athletic bradycardia, which suggested that the difference in ventilatory response between controls and divers might be fitness related. The data on runners test that idea.

The ventilatory responses of the runners are included in Table 1 and Figure 2. The responses of the runners were uniformly intermediate between those of the divers and controls. The mean values of P_{ACO2}, R, breathing

	Controls Mean 13 ± SE	Divers Mean 13 ± SE	Runners Mean 13 ± SE
		$P_{A_{CO_2}}$ (mm Hg)	
Rest	38.29 ± 0.43	38.72 ± 2.0	37.35 ± 0.58
1.6 km/hr	42.07 ± 0.71	44.99 ± 1.6	*44.70 ± 1.1
3.2 km/hr	44.85 ± 0.43	*48.41 ± 1.4	*47.34 ± 0.1
4.8 km/hr	44.92 ± 0.36	*49.63 ± 1.3	47.34 ± 1.6
		R	
Rest	0.774 ± 0.009	0.784 ± 0.021	*0.810 ± 0.1010
1.6 km/hr	0.767 ± 0.012	0.745 ± 0.024	0.795 ± 0.020
3.2 km/hr	0.844 ± 0.011	0.821 ± 0.015	0.845 ± 0.016
4.8 km/hr	0.934 ± 0.011	*0.877 ± 0.016	*0.860 ± 0.019

Table 2. Estimated $P_{A_{CO_2}}$ and R.

Values marked with asterisk are significantly different ($P < 0.05$) from control values.

	Controls	Divers	Runners
		Breathing frequency (min^{-1})	
Rest	13.57 0.47	*10.07 0.64	13.58 0.55
1.6 km/hr	16.84 1.35	13.33 1.57	14.94 0.86
3.2 km/hr	18.01 0.65	*13.16 1.17	17.48 1.04
4.8 km/hr	20.36 1.06	*15.32 1.11	19.27 1.28
		Tidal volume (liters, BTPS)	
Rest	0.722 0.031	*1.025 0.046	0.945 0.138
1.6 km/hr	1.172 0.076	*1.692 0.147	1.347 0.086
3.2 km/hr	1.548 0.054	*2.203 0.166	1.650 0.102
4.8 km/hr	2.018 0.076	*2.679 0.171	2.005 0.141

Table 3. Values marked with asterisk are significantly different ($P < 0.05$) from control values.

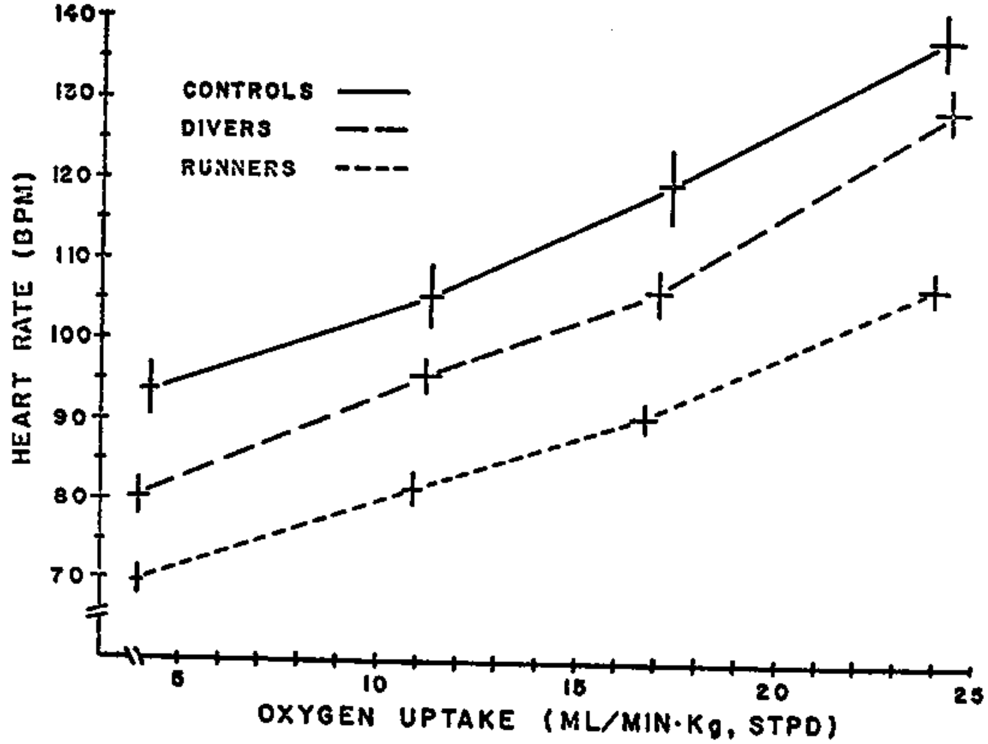

Figure 3. Heart rates of the three groups of subjects vs. oxygen uptake, indicating three distinct degrees of fitness.

frequency, tidal volume, and heart rate for runners are given along with those for controls and divers in Tables 2 and 3 and Figure 3. In general, runners exhibited the same trends as divers, but to a lesser extent, while their heart rates were considerably lower than those of the divers.

One factor not controlled for in this study was the possibility that divers might exhibit a conditioned response to the presence of the mouthpiece (which, to them, is a diving-related stimulus) whereas non-divers might not. To get some idea whether such a thing might be happening, we reviewed the data obtained during the experiments described earlier, comparing exercise in air and in water of various temperatures. We discovered the results given in Table 4. This table gives the breathing frequency and ventilation during exercise underwater as a percentage of the values measured during the same levels of exercise in air, both for the subjects of Moore et al. (1970) (non-divers) and those of Lally et al. (1971) (divers). As can be seen, the divers respond to immersion by considerable reductions in breathing frequency (even in 15 - 16°C water, where $\dot{V}_{O_2}$ is elevated due to pronounced shivering) whereas non-divers are not. The divers also exhibit a tendency towards lower ventilation underwater, while non-divers exhibit the opposite tendency (perhaps due to anxiety). These latter results could readily be interpreted as a conditioned response to a diving-related stimulus (immersion).

Breathing frequency, % of value in air

	Air	30°C Water	15-16°C Water
		(Non-divers)	
Rest	100	100±22	122±36
Moderate	100	92±32	117±36
Heavy	100	93±28	128±40
Forced	100	92±44	121±60
		(Divers)	
Rest	100	57±21	74±25
Moderate	100	57±21	57±23
Heavy	100	53±16*	60±20
Forced	100	56±17*	59±19

$\dot{V}_E$, % of value in air

	Air	30°C Water	15-16°C Water
		(Non-divers)	
Rest	100	110±16	132±23
Moderate	100	122±26	148±47
Heavy	100	100±28	136±39
Forced	100	143±39	138±31
		(Divers)	
Rest	100	116±36	152±52
Moderate	100	83±20	102±29
Heavy	100	71±18	94±24
Forced	100	78±28	81±27

Table 4. Breathing frequency and ventilation during exercise underwater as a percentage of the corresponding values in air. Values marked with an asterisk are significantly different ($P < .05$) from 100.

DISCUSSION

The principal finding of the treadmill study was the hypoventilation seen in divers relative to controls. The steady-state ventilatory responses of the divers at the 3.2 and 4.8 km/hr speeds were about 20% less than those of the control subjects, these differences being statistically significant. Since the divers used a slower, deeper breathing pattern, it may be that they had a higher ratio of alveolar to dead space ventilation, so that their alveolar ventilation might not have been markedly lower. We did not attempt any dead space measurements, so we cannot attack this matter directly. We have, however, estimated V_D by two methods: A) Using Radford's (1955) rule-of-thumb that V_D in ml equals body weight in pounds, and assuming V_D to be constant at this value, and B) using the equations given by Millhorn and Brown (1970) which provide for increases in V_D with increasing tidal volume. By method (A), we estimated that the lower $\dot{V}_D$ in divers compensated for 21 and 12.5% of the decrements in $\dot{V}_E$ observed at 3.2 and 4.8 km/hr respectively. By method (B), the reductions in V_D compensated for only 11.3 and 8.6% of the $\dot{V}_E$ decrements at 3.2 and 4.8 km/hr respectively. It therefore appears that the divers exhibited a relative alveolar, as well as pulmonary, hypoventilation.

Consequently, it is not surprising that the estimated $P_{A_{CO_2}}$'s in divers were significantly higher than in control subjects at the 3.2 and 4.8 km/hr speeds. The lower R values in divers (significant at 4.8 km/hr) suggest that their $P_{A_{CO_2}}$ would have exceeded that in control subjects by an even greater margin in exercise of longer duration. Thus, it does appear that a lower chemosensitivity occurs in divers, as suggested by Lanphier (1963, 1969).

With respect to the effect of fitness, we consider that Figure 3 indicates three distinct levels of fitness, as one would expect in sedentary subjects, divers, and runners. The ventilatory responses of the runners were intermediate between those of controls and divers, while their heart rates were appreciably lower. It appears that the hypoventilation found in divers is not related to greater fitness in divers, but is probably peculiar to the activity of diving.

Another striking difference between the divers and the other two groups was the considerably slower and deeper breathing pattern seen in divers. Seventeen years ago, Schaefer (1958) noted a relationship between ventilatory responsiveness and breathing pattern. He tested the response to exogenous CO_2 in many subjects, who were classified into high and low ventilation groups depending on their position on the ventilatory response distribution. The low ventilation group was found to have a slower, deeper breathing pattern under normocapnic conditions than the high ventilation group. To some extent, then, the divers in this study were reminiscent of Schaefer's low ventilation group. However, this latter group had a lower tidal volume during hypercapnia (than the high ventilation group) in contrast to the present finding of consistently (and significantly) higher tidal volumes in divers. Hence, we do not believe that the divers we studied were simply a sample of subjects from the low end of the ventilatory responsiveness distribution.

While the runners showed a trend toward lower $\dot{V}_E$ and higher $P_{A_{CO_2}}$ (compared to controls), they did not exhibit a trend towards the slow, deep breathing pattern. Because of these considerations, and in view of the previous observations described in the introduction to this paper, we believe the combined low ventilatory responsiveness and slow, deep breathing pattern to be attributable to the activity of diving.

In this connection, it is interesting to note that the divers studied in the earlier series of exercise experiments consistently exhibited lower breathing frequencies underwater than in air, even in cold water where metabolic rate was appreciably increased by shivering. This response to immersion was quite apparent even when the exercise was of sufficient intensity to fully occupy the subject's attention. The divers also exhibited a tendency towards lower V_E underwater than in air. The non-divers, in contrast, exhibited the opposite tendencies.

We interpret these findings in light of the subjective reports of divers, and our own experience, that one quickly "learns" to conserve air while diving by slowing the breathing frequency, or even "skip breathing"; i.e., a deep inspiration followed by an end-inspiratory pause of several seconds. We suggest that continuing diving activity may condition the diver to this breathing pattern, and that this breathing pattern (with consequent hypoventilation) may be elicited as a conditioned response to diving-related stimuli, such as breathing from a mouthpiece and water immersion.

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VENTILATORY RESPONSES TO EXERCISE IN DIVERS AND NON-DIVERS

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Abstract. The initial (neurogenic) and steady-state ventilatory responses to three levels of treadmill exercise (+10% grade; 1.6, 3.2 and 4.8 km/hr) were studied in divers, sedentary non-divers, and non-diving athletes. The steady-state ventilatory responses of divers were lower than the sedentary group, significantly so at 3.2 and 4.8 km/hr where the differences were 20% of the sedentary values. The initial responses of divers were also lower, but with marginal significance. Both initial and steady-state responses of athletes were intermediate between divers and sedentary non-divers. Both divers and athletes tended to have higher $P_{A_{CO_2}}$ and lower R values than sedentary non-divers; divers more so than athletes. Divers showed a markedly slower and deeper breathing pattern than the other two groups. We suggest that the unusual ventilatory behavior of divers is not fitness-related, reflects more than reduced chemosensitivity, and may involve a conditioned response phenomenon.

Alveolar carbon dioxide
Athletes
Breathing pattern
Control of breathing
Divers
Muscular exercise
Training

The literature contains several reports on the subject of ventilatory adaptations to breath-hold and SCUBA diving which are not entirely in mutual agreement. Schaefer (1955) studied a group of submarine escape tank instructors (essentially breath-hold divers) and found that the slope of their CO_2 response curves was about 30% less than those of non-divers, and that they exhibited a pattern of slow, deep breathing. Song *et al.* (1963) found that the hae-nyo, the diving women of Korea, had a lower ventilatory response to CO_2 than non-diving women, but that their response to hypoxia was not different. In a more detailed study, Schaefer (1965) reported that the characteristics of reduced CO_2 response and slow, deep breathing pattern in training tank instructors diminished significantly after several months of non-diving and returned when they resumed diving. He also reported that active divers had a lower response to hypoxia. Consequently, it

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seems clear that intensive breath-hold diving does induce reduced chemosensitivity.

It is not clear that this is so in SCUBA divers. Goff and Bartlett (1957) examined end-tidal P_{CO_2} during underwater swimming, and found that experienced swimmers showed P_{CO_2} values that were 9–14 mm Hg higher than inexperienced subjects. The experienced group used a pattern of deep inspiration followed by an end-inspiratory pause ("skip-breathing"). However, even when instructed not to pause, this group had end-tidal P_{CO_2}'s in the range of 44–49 mm Hg, suggesting reduced ventilatory responsiveness to chemical and/or neural stimuli in the experienced group. In contrast, Froeb (1960) compared 16 subjects having extensive SCUBA diving histories with 16 non-divers and found no difference between these groups in their response to exogeneous CO_2, either at rest or during exercise. However, Lanphier (1963) has reported studies on a group of SCUBA divers who exhibited end-tidal P_{CO_2}'s of 50 mm Hg during moderate exercise underwater at 4 ATA breathing air. The outstanding "CO_2 retainers" among this group were subsequently found to show marked increases in Pa_{CO_2} during moderate exercise in air at 1 ATA on low-resistance breathing apparatus.

The question of whether SCUBA diving induces a reduction in ventilatory responsiveness, with consequent CO_2 retention, has considerable practical importance. Lanphier (1969) has indicated that CO_2 retention lowers the threshold for oxygen convulsions, and Hesser *et al.* (1971) have shown that elevated Pa_{CO_2} potentiates N_2 narcosis. There is also the more fundamental question of the lability of the ventilatory control system. To what extent, and by what mechanisms, can the generation, central integration, and motor expression of ventilatory drives be modified by various types of physical activity?

The purpose of this study was to determine whether SCUBA divers have a ventilatory response to exercise or a breathing pattern that is demonstrably different from non-divers under dry, normobaric conditions, and, if so, to gain some insight into the cause of such differences.

Methods

The ventilatory responses to exercise of three groups of healthy, adult males were studied. These groups were divers, control subjects, and runners. "Divers"

TABLE I

Physical characteristics of subjects.

	Controls	Divers	Runners
N	9	8	11
Age (years)	29.5±2.7	32.6±4.0	27.2±1.9
Weight (kg)	67.6±2.8	80.4±6.6	68.0±2.4
Height (cm)	175.0±0.9	176.4±5.6	173.1±1.3
V.C. (ml)	4231±879	4910±793	4450±980

Physical characteristics of subjects. Values are means ±SE.

refers to SCUBA divers with 5 or more years (mean 11.3 ± 1.7 years) of diving experience. In most cases, they also had ample breath-hold diving experience during childhood. Their recent activity ranged from daily diving (20 h/week) down to weekend diving (1 or 2 h/week). "Controls" refers to laboratory personnel and graduate students with no diving experience. All were sedentary. "Runners" refers to distance (road) runners. While generally not outstanding in performance these men were in daily training, running from 30 to 100 km/week. The basic physical characteristics of each group are given in table 1.

The exercise was treadmill walking up a 10% grade at speeds of 1.6, 3.2, and 4.8 km/hr (1, 2, and 3 mph). The protocol was similar to that of D'Angelo and Torelli (1971). Each experiment comprised a 7-min walk at each of the three speeds. During the 2 min immediately preceeding each walk $\dot{V}_E$, $\dot{V}_{O_2}$ and $\dot{V}_{CO_2}$ were measured by the open circuit method with he subject at a standing rest. Mixed expired air was analyzed by the Scholander method (Scholander, 1947). These measurements were repeated over the last 2 min of each walk. The steady-state ventilatory response was measured as the difference between these two $\dot{V}_E$ values.

Inspiratory flow rate was recorded throughout the experiment by means of a screen pneumotachograph and a differential pressure transducer (Statham PM197). The flow rate was integrated (Beckman Type 9873 B resetting integrator) to provide breath-by-breath inspired tidal volume. Expired air was sampled continuously from the mouthpiece at a flow rate of 0.5 L/min and was analyzed for CO_2 with a Beckman LB-1 whose sensor was connected to the mouthpiece

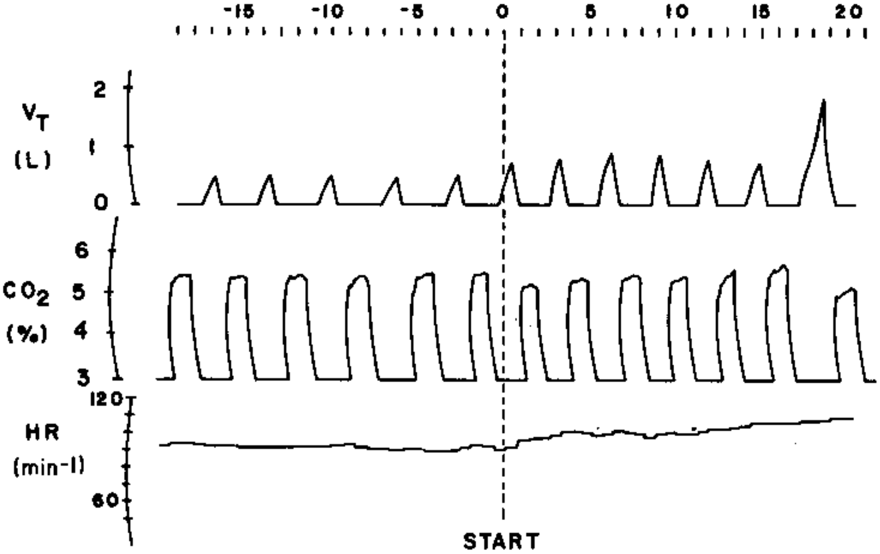

Fig. 1. Tracing of the response of a typical control subject to the onset of exercise at the 4.8 km/hr (3 mph) speed. Time, in seconds, is indicated across top of figure.

by 25 cm of PE 205 tubing (volume = 0.5 ml, transit time = 0.06 sec). ECG was recorded continuously via surface chest electrodes, and was used to derive a beat-by-beat heart frequency (Beckman type 9857 Cardiotachometer). All signals were recorded on a Beckman type R Dynograph. In all experiments, the subjects breathed through a standard SCUBA mouthpiece-valve assembly and hosing (U.S. Divers). Expired air was collected in rubberized canvas Douglas bags. The volume collected was measured by emptying the bag through a dry gas meter (American Meter Co.) which had been calibrated against a 350-liter Tissot spirometer.

Calibration of the LB-1 record was performed several times during each experiment using CO_2 in O_2 standard gases that had been analyzed on the Scholander apparatus. Calibration of the pneumotach (and integrator) was performed at three flow rates using a Flowrator (F&P Co., type B4-27-10/27).

None of the subjects were experienced treadmill walkers, so that differing degrees of familiarity with the treadmill did not enter into our comparison of the three groups. Each subject was given a few minutes practice to ensure against mishaps. To avoid a startle response, the subject started the treadmill himself, on a cue from the investigator, by means of a switch conveniently located by his side. In any case, there is evidence to indicate that the choice of starting procedure has little effect on the ventilatory response to the onset of exercise (Jensen *et al.*, 1971).

As mentioned earlier, the steady-state $\dot{V}_E$ response was measured as the difference ($\Delta\dot{V}_E$) between the resting $\dot{V}_E$ and the $\dot{V}_E$ during the last 2 min of exercise. The initial response was also recorded as an estimate of the neurogenic component of the exercise hyperpnea. This was measured as the increase in $\dot{V}_I$ observed during the first 15 sec (approximately) of exercise as determined from the breath-by-breath inspired volume record. Since $\dot{V}_I$ and $\dot{V}_E$ are not equal, this $\Delta\dot{V}_I$ would not be the same as the corresponding $\Delta\dot{V}_E$. However, this error is small, since the R values were not greatly different from 1.0, the $\dot{V}_{O_2}$ values were low, and the initial $\Delta\dot{V}_I$ was not large. The exact time interval over which the initial response was measured depended on the time required to complete an integer number of ventilatory cycles. In no case was an inspiration counted if the preceeding expiration exhibited an increase in end-tidal CO_2. Because of the problem of the breath-by-breath variability of tidal volume, the number of observations of the initial response was increased by including two or three short walks of 15–20 sec duration prior to each rest interval. The concept that this initial response is an estimator of neurogenic ventilatory drive has a long history, evidence for it having been provided by Asmussen and Nielsen (1948) and by Dejours and his colleagues (Dejours, 1959, 1963; Dejours *et al.*, 1955). This concept has been reaffirmed by D'Angelo and Torelli (1971).

$P_{A_{CO_2}}$ was estimated as the P_{CO_2} in the expired air at the time corresponding to 60% of the duration of the expiratory phase on the breath-by-breath record. Dubois *et al.* (1952) indicated that this is a reasonable approximation to the

TABLE 2

Oxygen uptakes and ventilatory responses. Values are mean ±SE

Groups	Rest	1.6 km/hr	3.2 km/hr	4.8 km/hr
			(ml/kg. min)	
Controls	4.31±0.14	11.27±0.30	17.32±0.28	24.14±0.38
Divers	4.03±0.19	11.21±0.34	17.00±0.31	24.33±0.43
Runners	3.99±0.08	10.87±0.23	16.68±0.30	22.62±0.40
			Steady state $\Delta\dot{V}_E$ (L/kg. min)	
Controls		0.140±0.005	0.268±0.007	0.455±0.066
Divers		0.134±0.006	0.215±0.010	0.367±0.017
Runners		0.135±0.009	0.243±0.008	0.382±0.014
			Initial $\Delta\dot{V}_I$ (L/kg. min)	
Controls		0.101±0.008	0.122±0.016	0.163±0.017
Divers		0.102±0.019	0.104±0.009	0.126±0.010
Runners		0.093±0.011	0.118±0.011	0.140±0.010

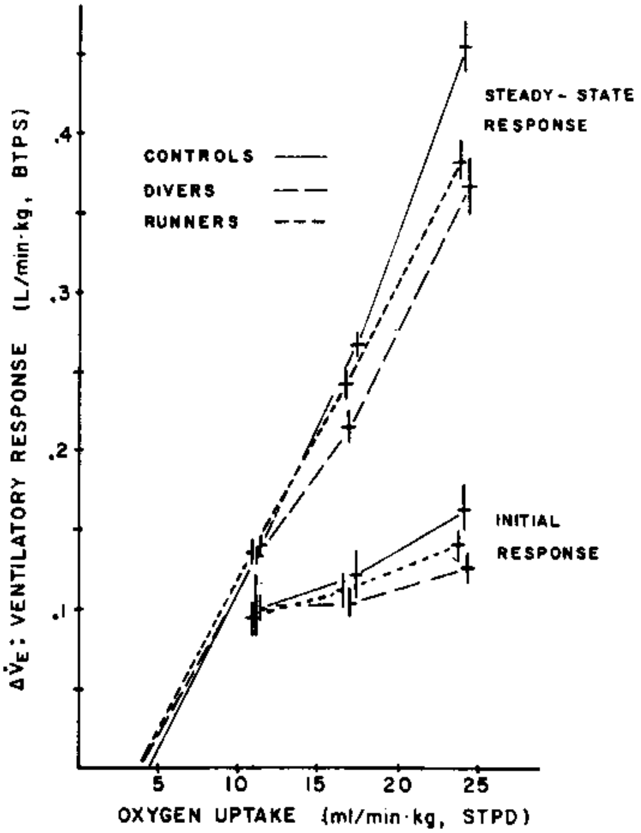

Fig. 2. Steady-state and initial (neurgenic) ventilatory responses to exercise of the three groups of subjects as a function of $\dot{V}_{O_2}$.

TABLE 3

Mean initial response as % of mean steady-state response.

	1.6 km/hr	3.2 km/hr	4.8 km/hr
Controls	71	45	36
Divers	70	48	35
Runners	76	46	37

TABLE 4

ESTIMATED $P_{A_{CO_2}}$ and R.

	Controls	Divers	Runners
	Mean 13 ESE	Mean 13 SE	Mean 13 SE
		$P_{A_{CO_2}}$ (mm Hg)	
Rest	38.29 ± 0.43	38.72 ± 2.0	37.35 ± 0.58
1.6 km/hr	42.07 ± 0.71	44.99 ± 1.6	*44.70 ± 1.1
3.2 km/hr	44.85 ± 0.43	*48.41 ± 1.4	*47.34 ± 1.0
4.8 km/hr	44.92 ± 0.36	*49.63 ± 1.3	47.34 ± 1.6
		R	
Rest	0.774 ± 0.009	0.784 ± 0.021	*0.810 ± 0.010
1.6 km/hr	0.767 ± 0.012	0.745 ± 0.024	0.795 ± 0.020
3.2 km/hr	0.844 ± 0.011	0.821 ± 0.015	0.845 ± 0.016
4.8 km/hr	0.934 ± 0.011	*0.877 ± 0.016	*0.860 ± 0.019

Values marked with asterisk are significantly different ($P < 0.05$) from control values.

$P_{A_{CO_2}}$ averaged over the ventilatory cycle.

The results to be presented represent complete data on exercise at all three speeds obtained in 13 experiments on 8 divers, 17 experiments on 9 non-diving controls, and 20 experiments on 11 non-diving runners. The $\dot{V}_{O_2}$ and ventilatory response data were divided by body weight to achieve grouping of the $\dot{V}_{O_2}$ results. The normalized ventilatory responses of the three groups were then compared by the standard procedure of testing the differences between group means by a two-tailed *t* test for groups of unequal sizes.

Results

Fig. 1. is a tracing of a typical response of a control subject to the onset of exercise at the 4.8 km/hr speed. Time, in seconds, measured from the start of exercise is indicated along the top of the figure. An immediate increase in ventilation is apparent with increases in both Vt and frequency. This is accompanied by an initial reduction in expired CO_2 concentration. The time required for the expired CO_2 to equal and then exceed the resting value is typically 12–15 sec.

TABLE 5

Breathing patterns.

	Controls	Divers	Runners
		Breathing frequency (min^{-1})	
Rest	13.57±0.47	*10.07±0.64	13.58±0.55
1.6 km/hr	16.84±1.35	13.33±1.57	14.94±0.86
3.2 km/hr	18.01±0.65	*13.16±1.17	17.48±1.04
4.8 km/hr	20.36±1.06	*15.32±1.11	19.27±1.28
		Tidal volume (liters, BTPS)	
Rest	0.722±0.031	*1.025±0.046	0.945±0.138
1.6 km/hr	1.172±0.076	*1.692±0.147	1.347±0.086
3.2 km/hr	1.548±0.054	*2.203±0.166	1.650±0.102
4.8 km/hr	2.018±0.076	*2.679±0.171	2.005±0.141

Values marked with asterisk are significantly different ($P < 0.05$) from control values.

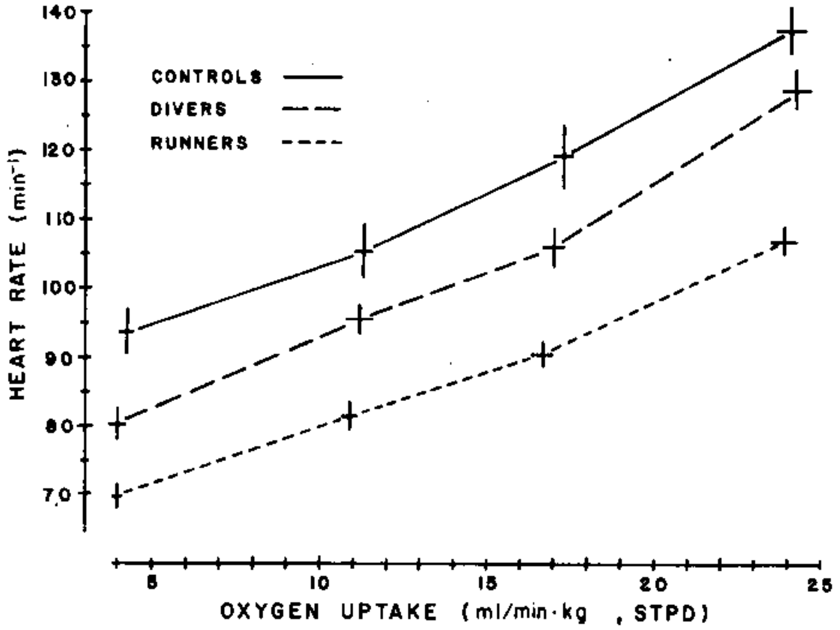

Fig. 3. Heart rate of all three groups as a function of normalized $\dot{V}_{O_2}$, suggesting three different levels of "fitness" (see text).

This time decreases with increasing fitness, and was 8–10 sec in the runners.

The initial phase of this study was a comparison of divers and control subjects, with the results shown in table 2 and the two thin lines in fig. 2. The steady-state ventilatory response of the divers was less than that of the control group at all three speeds, significantly so at 3.2 and 4.8 km/hr ($P < 0.01$). At the two higher speeds, the $\Delta\dot{V}_E$ decrements (from controls to divers) were about 20% of the control values. The initial ventilatory responses were also less in divers, but with only marginal significance ($P < 0.1$ at 4.8 km/hr). The decrements in the initial response are 35–45% of the decrements in the steady-state response. The mean values of the initial response as percentages of the mean steady-state responses are given in table 3.

A 20% reduction in $\dot{V}_E$ should result in an elevated P_{CO_2} and CO_2 retention as the exercise progressed. Table 4 indicates that this is so. The divers have a significantly higher estimated $P_{A_{CO_2}}$ at 3.2 and 4.8 km/hr and a significantly lower R at 4.8 km/hr, as measured from 5 to 7 minutes of exercise.

Examination of the breathing patterns (table 5) shows that the breathing frequency is lower in divers in all cases, while their tidal volume is significantly higher in all cases.

The divers also exhibited a lower heart fequency than controls at the same normalized oxygen uptake, as shown in fig. 3. This was interpreted as an athletic bradycardia, and suggested that the difference in ventilatory response between divers and controls might be fitness-related. The experiments on runners provide a test of that idea.

The ventilatory responses of the runners are included in table 2 and fig. 2. The responses of the runners were intermediate between controls and divers. Relative to control subjects, the steady-state response of the runners was not different at 1.6 and 3.2 km/hr, but was significantly lower at 4.8 km/hr. Relative to divers, the steady-state response of the runners was not different at 1.6 or 4.8 km/hr but was significantly higher at 3.2 km/hr. There were no significant differences in the initial response.

The mean values of $P_{A_{CO_2}}$, R, breathing frequency, tidal volume, and heart frequency for runners are given along with those for controls and divers in tables 4 and 5 and fig. 3. In general, the runners, compared to controls, exhibited the same trends as divers, but to a lesser extent.

Discussion

The principal finding of this study was the relative hypoventilation seen in divers compared to controls. This was significant at the 3.2 and 4.8 km/hr speeds. The absence of a difference at 1.6 km/hr apparently is due to the fact that the neurogenic responses were 70% of the total, and that they happened to be the same in both groups. As shown in table 3, the neurogenic component of the hyperpnea became a progressively smaller fraction of the total with increasing treadmill speed, and significant differences occurred in the total response despite the lack of significant differences in the neurogenic responses at the 3.2 and 4.8 km/hr speeds.

RESPONSES OF THE CONTROL SUBJECTS

In comparing the responses of the control subjects with data in the recent literature, one finds that the slope of the total response ($\Delta\dot{V}_E/\Delta\dot{V}_{O_2}$, liter BTPS/liter STPD)

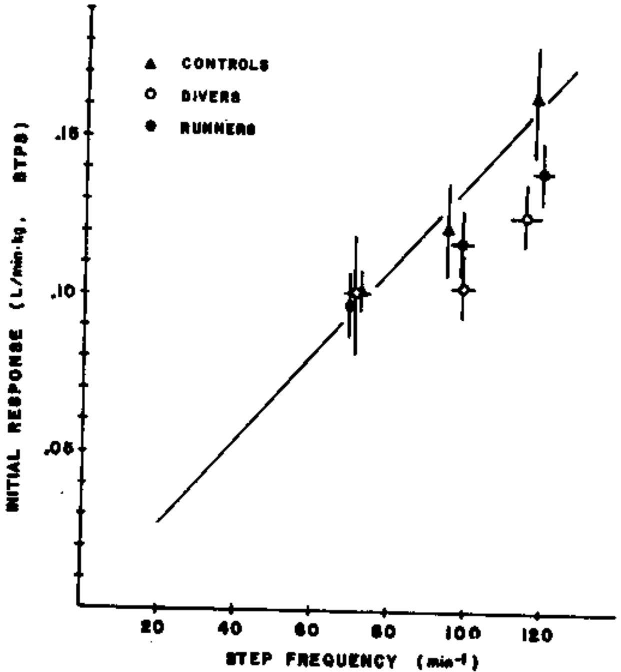

Fig. 4. Initial ventilatory response as a function of step frequency. The line is drawn to the control points.

in our data of about 18.0 compares very well with the values of 18.6 and 17.7 observed by D'Angelo and Torelli (1971) in level and uphill (+20%) treadmill walking. However, these authors found the neurogenic response to vary linearly with $\Delta\dot{V}_{O_2}$, on a line that went through the origin. Consequently, the neurogenic response was a fixed fraction of the total (for a given exercise) independent of $\dot{V}_{O_2}$. This is clearly not the case in the present data. However, there was a relationship between the neurogenic response and step frequency in control subjects (fig. 4) which could reasonably be extrapolated through the origin.

In any case, it has previously been found that frequency of limb movement does partially determine the neurogenic response (Dejours, 1964) and D'Angelo and Torelli's comparison of walking on different grades bears this out. The present data suggest that step frequency may be a more important determinant of the neurogenic drive in walking than is metabolic intensity. Hence, we agree with the observation of Bainton (1972) that the neurogenic ventilatory drive is not directly related to metabolism and should be thought of as a coarse adjustment.

The magnitudes of the neurogenic responses in the present data are intermediate between the high values reported by D'Angelo and Torelli (1971) and the low values reported by Craig *et al.* (1963) and Beaver and Wasserman (1968).

RESPONSES OF DIVERS COMPARED TO CONTROLS

The steady-state ventilatory responses of the divers at the 3.2 and 4.8 km/hr speeds were about 20% less than those of the control subjects, which was statistically significant. This decrement in ventilation in divers leads to a tendency toward higher estimated $P_{A_{CO_2}}$ (significant at 3.2 and 4.8 km/hr) and a tendency toward lower R's (significant at 4.8 km/hr). These results support the characterization of divers as "CO_2 retainers." The lower R's in divers suggest that their $P_{A_{CO_2}}$ would have exceeded that in controls by a greater margin in exercise of longer duration. Since the decrements in the neurogenic response account for only 35–45% of the decrements in the total response, it appears that a lower chemosensitivity does occur in SCUBA divers, as suggested by Lanphier (1963, 1969).

It is very curious that the decrements in the neurogenic response were in the 35–45% range, insofar as the contribution of the neural drive to the total response was also on the order of 35–45%. This implies that the decrements in the chemogenic drive were also roughly proportional to their relative contribution, and raises the possibility that the respiratory centers in divers are uniformly less sensitive to all stimuli.

Since the divers have a slower, deeper breathing pattern than controls, they should have a lower $\dot{V}_D$ and should attain a higher $\dot{V}_A$ per unit of $\dot{V}_E$ than controls. The observed relative pulmonary hypoventilation may be misleading. We have estimated $\dot{V}_D$ in these two groups by two methods: (a) using the rule-of-thumb of Radford (1955) that V_D in ml equals body weight in pounds, and assuming V_D to be constant at this value, and (b) using the relations given by Milhorn and Brown (1970) which provide for increases in V_D with increasing V_T. By method (a), we estimated $\dot{V}_D$ to be lower in divers by only 0.011 L/kg min at both the 3.2 and 4.8 km/hr speeds, so that the reduction in $\dot{V}_D$ compensated for 21% of the decrement in $\dot{V}_E$ at 3.2 km/hr and only 12.5% of the decrement at 4.8 km/hr. By method (b), the estimated differences in $\dot{V}_D$ are even less, compensating for only 11.3 and 8.6% of the decrements in $\dot{V}_E$ observed at 3.2 and 4.8 km/hr, respectively. Consequently, one cannot regard the slow, deep breathing pattern in these divers as compensatory for the reduced ventilatory responsiveness, since it is wholly inadequate for that purpose: Indeed, the disproportionality low f is the source of the hypoventilation.

EFFECT OF FITNESS

In this context, we consider "fitness" to be indicated by those physiological parameters that vary with the degree of physical activity, rather than genetic factors related to natural athletic endowment. One of the hallmarks of physical training is the development of an "athletic bradycardia," with ventricular enlargement. The relation between cardiac output and $\dot{V}_{O_2}$, however, is relatively constant (Åstrand and Rodahl, 1970). The data of fig. 3 show considerably different mean heart frequencies in the three groups at very comparable levels of $\dot{V}_{O_2}$. Insofar as stroke volume tends to be proportional to body weight (Holt *et al.*, 1968), a difference in mean body weight between groups could contribute to a difference in heart frequency. However, $\dot{V}_{O_2}$ in fig. 3 is normalized by body weight. Consequently, we consider that fig. 3 indicates three distinct levels of fitness, as one would expect in sedentary subjects, divers, and runners.

The ventilatory responses of the runners are intermediate between those of controls and divers (fig. 2), while their heart frequency responses are considerably lower (fig. 3). It appears that the hypoventilation found in divers (relative to controls) is not attributable to greater fitness in divers, although fitness may entail a minor contributing factor.

BREATHING PATTERN

Sixteen years ago, Schaefer (1958) attempted to relate ventilatory responsiveness and breathing pattern. Schaefer tested the response to exogenous CO_2 in a large number of subjects. Subjects were classified into high or low ventilation groups, depending on their relative positions on the ventilatory response distribution. He found that the low ventilation group exhibited a slower, deeper breathing pattern under normocapnic conditions than the other group. Thus, to some extent, the breathing patterns of divers in the present study are reminiscent of Schaefer's low ventilation group. However, the latter group showed a lower V_T than the other subjects during hypercapnia, which is in contrast to the present finding of uniformly (and significantly) higher V_T in divers. It does not seem likely that the divers in this study are merely a sample of subjects from the low end of the ventilatory responsiveness distribution.

While runners showed a tendency towards reduced ventilatory responsiveness relative to controls, their breathing pattern did not tend toward the slow, deep pattern seen in divers. Thus, the slow, deep pattern seems to be peculiar to divers, and to be intimately related to their reduced ventilatory responsiveness, insofar as the hypoventilation in divers is attributable to their lower f.

Interestingly, an earlier study at the University of Hawaii comparing exercise in air and water in a different group of divers revealed that the breathing pattern became slower and deeper underwater than in air, even though the breathing apparatus was the same in both cases (Lally *et al.*, 1971). This response to immersion was not seen in non-divers (Moore *et al.*, 1970) under the same experimental conditions.

Any attempt to explain these ventilatory anomalies in divers is complicated by the fact that the factors governing the trade-off of f and V_T are not well understood in "normal" subjects. It has been proposed that pulmonary and thoracic afferents are integrated with chemical stimuli in such a way as to minimize the work of breathing (Otis *et al.*, 1950) or the mean ventilatory muscle tension (Mead, 1960) at any given $\dot{V}_A$. Otis *et al.* and Mead constructed curves of work or mean muscle tension *vs.* f which had minima at values of f which agree roughly with observed values. However, Dejours (1966) has noted that both approaches yield broad minima; *i.e.*, work or mean tension are not very sensitive to f and are, therefore, not likely determinants of it. He also notes that these general considerations do not explain the wide individual variation in f, for which no thoraco–pulmonary mechanical explanations have been demonstrated.

Similarly, these considerations do not explain the observation that divers exhibit a drop in f and an increase in VT upon water immersion whereas non-divers do not. On the basis of the present data, we cannot identify the peripheral and/or central control mechanism alterations responsible for the divers' breathing pattern or reduced ventilatory response to exercise. The frequently expressed comment of SCUBA divers that they "learned", with experience, to conserve air by breathing less often, together with the observations reported here, suggests that the unique ventilatory behavior of divers may reflect a conditioned response phenomenon.

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Effect of Immersion at Different Water Temperatures on Graded Exercise Performance in Man

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MOORE, T. O., E. M. BERNAUER, G. SETO, Y. S. PARK, S. K. HONG and E. M. HAYASHI. *Effect of immersion at different water temperatures on graded exercise performance in man.* Aerospace Med. 41(12):1404-1408.

Eight subjects performed graded leg exercise at loads from light to forced maximal in air and totally submerged in water at 30°, 22°, and 16°C. There was no significant decrement in performance between the air and immersed environments. Heart rate, minute volume ($\dot{V}_E$), oxygen consumption ($\dot{V}_{O_2}$), and carbon dioxide production had high linear correlation coefficients with imposed work load. $\dot{V}_E$ and $\dot{V}_{O_2}$ were higher in water under all work loads and at the two lower water temperatures. Heart rate was the same at rest under all conditions, but significantly less at high work loads in 16°C water when compared to air. It is concluded that monitoring of a diver's heart rate will cause underestimation of work load in surface-equivalent terms at high loads in water of low temperature. The data confirm and extend information on underwater work to lower temperatures and higher work loads.

WITH INCREASING WORK performance by man in the sea, considerable research has involved the assessment of energy cost of underwater tasks,[5,7] the effects of water temperature,[5,6,7] and the simplest physiologic monitoring to provide accurate prediction of energy cost and stress level of underwater work.[11] Studies of correlates of underwater work have seldom provided data for a broad range of work output and/or water temperatures below 20°-25°C. This investigation combines the two, concentrating especially upon the extreme case, high work output and low temperature.

METHODS

Eight healthy adult males, ranging in age from 23 to 42 years ($\bar{x} = 32.2$), served as subjects. All but one were members of the Department of Physiology, Univ. of Hawaii. The exception was a local diving instructor. All subjects but one had previous underwater experience, either SCUBA or skin diving. Table I summarizes the physical characteristics, lung volumes, and estimated maximal oxygen uptake ($\dot{V}_{O_2}$ max) of the subjects.

The experiments consisted of graded exercise levels performed in air and submerged in water at 30°, 22°, and 16°C. In all cases, the subject's air source was a standard 71.2 cu. ft. SCUBA tank utilizing a double-stage, double-hose demand regulator (U.S. Divers Co., Santa Ana, California). Respiration rate (f_r) was recorded by sealing a thermistor into the regulator exhaust near the mouthpiece. Air temperature changes during expiration were transmitted to a polygraph for recording. Rectal temperature ($T°_r$) was read directly from a Yellow Springs telethermometer.

Heart rate (f_h) was continuously monitored on the polygraph via the attachment of two precordial Beckman electrodes to the subject.

Expired gas was collected from the regulator exhaust either into 250-liter Douglas bags or into a 350-liter Tissot respirometer. Gas samples were subsequently analyzed for CO_2 and O_2 on a Scholander micro-gas analyzer. Oxygen was also analyzed on a Beckman Oxygen analyzer. A standard diving mask was worn and insured respiration by mouth only.

An exercise machine was designed and built for use in an immersion tank (3x5x5 ft. deep) (Figure 1). The subject, in a sitting position, pushed alternately with each leg against independent pedals. The pedals were attached, through a pulley system, to weights external to the immersion tank, where loads could be changed easily. Work was restricted to leg muscles in all trials. Weight displacement was limited by a subject's leg length, but there was room for maximal pedal swing within the tank for each subject.

During exercise trials at any given load, the subjects maintained a cadence of either sixty or one hundred

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strokes per minute, as detailed below. Strokes were recorded on a polygraph by means of impulses from a photocell attached to the chassis of the machine, positioned so as to be activated by the passage of the pedals through their swing. Actual weight displacement was measured in several trials on each subject.

The following regimen was adhered to during both air and submerged exercise experiments: Figure 2 illustrates a typical recording regimen.

1. The subject sat quietly at rest, breathing from SCUBA, for five minutes. During the last two minutes, his steady-state heart rate, respiratory rate, rectal temperature, room (or water) temperature were recorded, and expired gas samples taken.
2. The subject immediately began light exercise (4.5 to 6.8 Kg./leg) at the rate of 60 strokes/min., for five minutes. The same steady-state parameters were recorded during the fifth minute.
3. Following a three-minute rest, the procedure repeated step 2 with a moderate load (9.1-13.5 Kg./leg).
4. The rest period was repeated, and heavy exercise (13.5-18.2 Kg./leg) began and proceeded for four minutes. Recordings and collections were made during the fourth minute. At the end of the fourth minute, the cadence was increased (without a break) to the forced exercise level of 100 strokes/minute for one and one-half minutes. The final gas collection occurred in the last thirty seconds of the period of forced exercise.

Three trials were performed in 23°-25°C air, both inside the empty immersion tank and outside. Two trials occurred before the submersion experiments, the third after. The schedule of experiments was such that any given subject performed the experiment, either in air or submerged, only once per week, in order to avoid any training effects.[3,8] No decreases in resting heart rates were noted over the weeks of the experimental period.

During submersion experiments, the subjects were seated on the exercise machine in approximately four

TABLE I. MEAN VALUES FOR PHYSICAL CHARACTERISTICS, LUNG VOLUMES AND ESTIMATED $\dot{V}_{O_2}$ MAX.

	$\bar{X}$ ± S.E.M.
Age (yrs)	32 ± 2.3
Height (cm)	172.1 ± 2.8
Weight (kg)	70.6 ± 1.7
Surf. area (m^2)	1.84 ± .05
Body fat (%)**	18.0 ± 2.3
Vital cap. (ml*)	4755 ± 246
Resid. vol. (ml*)***	1353 ± 102
max breathing cap (L/min*)	
with respirometer	224 ± 11
with SCUBA	153 ± 6
$\dot{V}_{O_2}$ max (L/min)****	2.63 ± .16

*BTPS

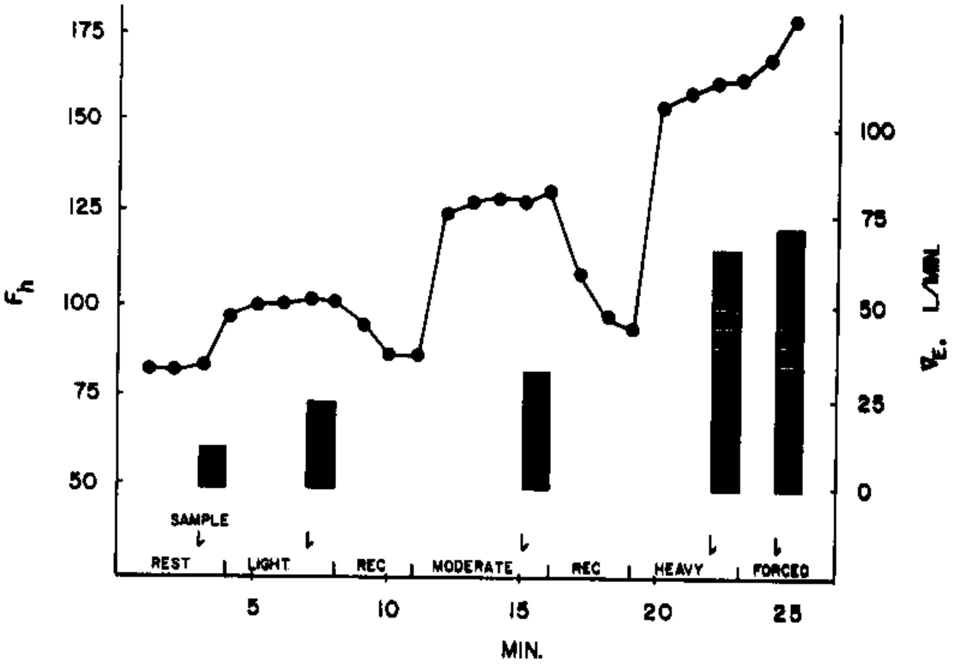
**Estimated by the method of Brozek *et al.* (4).

***Determined by the 3-breath method of Rahn *et al.* (10).

****Estimated by the method of Astrand (2).

Fig. 1. Submersible ergometer.

Fig. 2. Experimental procedure with typical results from one subject.



feet of water. Water temperature was maintained at any level within one degree centigrade by the addition of either hot water or ice.

Heart rates are expressed as steady-state levels taken from the last minute or half-minute at any work level. Work level is calculated as kilogram • meters per minute. Minute ventilation ($\dot{V}_E$) is corrected to BTPS, O_2 consumption ($\dot{V}_{O_2}$) and CO_2 production ($\dot{V}_{CO_2}$) to STPD.

RESULTS

A summary of the experimental results appears in Table II. Under all conditions, $\dot{V}_{O_2}$, f_h, $\dot{V}_E$, $\dot{V}_{CO_2}$ were directly related to external work with linear correlation coefficients above 0.8 (Table III). In the case of heart rate versus external work, the correlation coefficient was + 0.92 in air and reduced approximately 0.1 upon immersion, regardless of water temperature.

TABLE II. EXPERIMENTAL RESULTS FROM EXERCISE IN AIR AND DURING IMMERSION (MEANS ± 1 S.E.M.)

	Air	30° H_2O	22° H_2O	16° H_2O
External Work (Kg.M/min)				
Light	188 ± 10	191 ± 11	189 ± 11	193 ± 12
Moderate	314 ± 19	315 ± 18	326 ± 22	322 ± 19
Heavy	475 ± 24	469 ± 36	466 ± 28	473 ± 34
Forced	727 ± 29	750 ± 63	711 ± 49	745 ± 58
Heart Rate (beats/min)				
Rest	84 ± 4	82 ± 6	79 ± 5	82 ± 5
Light	102 ± 5	104 ± 5	100 ± 5	103 ± 4
Moderate	116 ± 6	117 ± 6	111 ± 6	113 ± 5
Heavy	136 ± 6	134 ± 6	126 ± 6	127 ± 5
Forced	157 ± 4	159 ± 5	149 ± 5	143 ± 4
Respiration Rate (breaths/min)				
Rest	9 ± 1	9 ± 1	9 ± 1	11 ± 2
Light	10 ± 2	9 ± 2	12 ± 2	13 ± 3
Moderate	12 ± 2	11 ± 2	14 ± 1	14 ± 2
Heavy	14 ± 2	13 ± 2	16 ± 1	18 ± 3
Forced	24 ± 7	22 ± 4	34 ± 6	29 ± 6
Minute Volume (L/min)				
Rest	9.6 ± 0.9	10.6 ± 0.5	15.1 ± 2.3	12.7 ± 1.0
Light	15.2 ± 1.6	19.5 ± 2.2	26.7 ± 3.5	22.6 ± 3.9
Moderate	20.8 ± 1.7	25.4 ± 3.2	35.3 ± 4.2	30.8 ± 4.3
Heavy	31.2 ± 3.9	31.3 ± 4.7	45.1 ± 4.5	42.3 ± 6.7
Forced	39.6 ± 4.0	56.7 ± 9.7	64.3 ± 6.8	54.7 ± 6.8
O_2 Consumption (L/min)				
Rest	0.36 ± .03	0.34 ± .02	0.52 ± .06	0.45 ± .04
Light	0.69 ± .05	0.86 ± .07	1.20 ± .12	1.10 ± .09
Moderate	0.97 ± .07	1.07 ± .09	1.20 ± .07	1.40 ± .12
Heavy	1.30 ± .12	1.30 ± .14	1.50 ± .15	1.60 ± .19
Forced	1.60 ± .11	2.00 ± .19	1.90 ± .15	1.80 ± .14
CO_2 Production (L/min)				
Rest	0.32 ± .02	0.31 ± .03	0.48 ± .04	0.43 ± .09
Light	0.71 ± .04	0.75 ± .04	1.07 ± .06	0.94 ± .06
Moderate	0.90 ± .03	0.98 ± .06	1.12 ± .05	1.33 ± .08
Heavy	1.30 ± .06	1.17 ± .07	1.53 ± .10	1.63 ± .08
Forced	1.60 ± .06	2.02 ± .09	1.98 ± .11	1.98 ± .08
Rectal Temperature (°C)				
Rest	37.4 ± .07	37.4 ± .10	37.6 ± .10	37.4 ± .08
Light	37.3 ± .08	37.4 ± .16	37.3 ± .16	37.3 ± .09
Moderate	37.3 ± .05	37.3 ± .12	37.0 ± .26	37.2 ± .13
Heavy	37.4 ± .08	37.3 ± .06	36.7 ± .44	37.0 ± .21
Forced	37.4 ± .04	37.3 ± .13	36.6 ± .56	36.8 ± .28

At rest, $\dot{V}_{O_2}$ was generally higher in water than in air, especially in 22° and 16°C water, while that in 30°C water was comparable to $\dot{V}_{O_2}$ in air. During light and moderate work, $\dot{V}_{O_2}$ was higher than in air during all immersion trials at 22° and 16°C. At the higher work levels, however, there was less difference between air and immersion trials. Similar results at submaximal work levels have been reported by Craig and Dvorak[7] for water temperatures from 35° to 25°C. Our data extend the findings to 16°C water. At the forced work level, $\dot{V}_{O_2}$ reached approximately 75% of estimated $\dot{V}_{O_2}$ max.

Ventilation followed a pattern similar to that of $\dot{V}_{O_2}$. $\dot{V}_E$ plotted against $\dot{V}_{O_2}$ (Figure 3) yielded no significant difference, again confirming and extending Craig's work to the lower temperature.

The gas exchange ratio ($\dot{V}_{CO_2}/\dot{V}_{O_2}$) tended to increase in all trials with increased work load, indicating that a mild degree of hyperventilation may have occurred. This was especially noted at the forced work level and there is a marginally significant difference ($P =$ approximately 0.05) at this level between air and 16°C water.

The relationship between heart rate and $\dot{V}_{O_2}$ is shown in Figure 4. Heart rate for a given $\dot{V}_{O_2}$ was less in water than in air, particularly 22° and 16°C water. In 30°C water, heart rate averaged 14 beats/L $\dot{V}_{O_2}$ less than in air, while in 22° and 16°C water, heart rate was 31.5 and 31.6 beats/L $\dot{V}_{O_2}$ less than air, respectively. The slopes of the regression lines were not different in any trial.

Heart rate and external work were linearly related with no significant differences between work in air and in 30° and 22°C water. At work loads above light, however, in 16°C water, there was a tendency toward lower heart rate per unit work (Figure 5). At the forced work level, heart rate was 143 ± 4 (S.E.) in cold water vs. 157 ± 4 in air. This decrease in 16°C water is significant below the 2% level. While the intercepts are not different for the regression lines, the slopes were significantly different ($P < 0.02$). In air, the slope was 0.103 ± 0.004, and 0.08 ± 0.006 in 16°C water.

Rectal temperature did not change significantly in air, 30° and 16°C water. There was a progressive decrease in mean temperature recorded in 22°C water but this was primarily the effect of one subject who spent additional time in the water before the work trial began and violent shivering was noticeable. This subject had one of the lowest body fat estimates (9.5%).

None of the physical characteristics correlated significantly with the heart rate or $\dot{V}_{O_2}$ response to work level. Respiratory rate did not differ and correlation with external work was not high (0.63-0.69 for all trials). In part, this was due to the fact that the subjects generally

TABLE III. CORRELATION COEFFICIENTS

	Air	30°C water	22°C water	16°C water
$\dot{V}_{O_2}$ vs. external work	+0.90	+0.90	+0.87	+0.85
$\dot{V}_{CO_2}$ vs. external work	+0.90	+0.88	+0.87	+0.86
$\dot{V}_E$ vs. external work	+0.88	+0.85	+0.84	+0.92
f_h vs. external work	+0.92	+0.82	+0.84	+0.83

adjusted their rate to some harmonic of the exercise cadence and regulated ventilation by altering tidal volume, and method was left to the subject. While maximal breathing capacity was not approached during this experiment, it was noted in early trials that MBC with SCUBA was reduced 31% from that attainable with a spirometer (Table I).

DISCUSSION

This study confirms work from Craig's laboratory and extends the data to lower water temperatures and higher work loads. Under the conditions of this experiment, the ventilation equivalent ($\dot{V}_E/\dot{V}_{O_2}$) remained unchanged even at 16°C water immersion (see Figure 3). This is in contrast to the findings of Costill et al.,[5] who reported a decreased efficiency of breathing in 17.4°C water in subjects swimming vigorously ($\dot{V}_{O_2} = 3.0$ L/min.). It is probable that the higher O_2 demand and decreased breathing efficiency recorded in the latter study reflects the use of more muscle mass than in the present study and those of Craig. Normal SCUBA swimming technique requires predominantly leg exercise.

Weltman and Egstrom[11] have reported from their studies that monitoring heart rate or minute ventilation allowed reasonable approximation of imposed workload in surface-equivalent terms. Craig suggested[7] that ventilation during work seemed to be somewhat greater in the coldest water tested (25°C). This was most pronounced at highest work loads. Our results confirm this, with an interesting qualification. While $\dot{V}_E$ in 30°C water was generally the equivalent of that in air at any given load, the ventilation in 22° and 16°C water was higher than in air under all work loads, particularly at the forced level. On the average, $\dot{V}_E$ was higher in 22°C water (though not significantly so) than in air or other water temperatures (Table II). Since Hensel and Wurster[9] have reported that frequency of discharge of facial cold receptors of the cat shows a maximum of about 25°-27°C with lower frequencies at higher and lower temperatures, it is an intriguing speculation that the increased $\dot{V}_E$ is not only cold receptor mediated, but that there is an optimum temperature range instead of simple linearity involved. The data reported here do not contribute to a resolution of the question, however.

Of particular interest is the relationship between heart rate and work load. The data indicate that there is a significantly lower heart rate at high work levels in cold water compared to air and that the regression

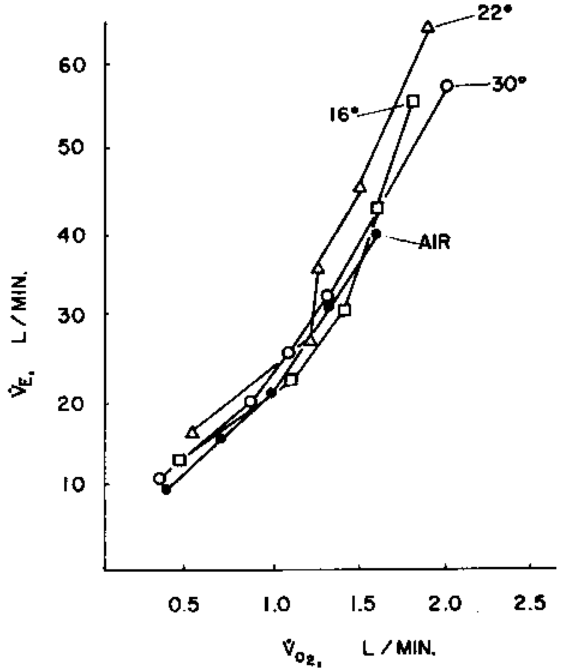


Fig. 3. Minute ventilation versus oxygen consumption during exercise in air and submerged in 30°, 22° and 16°C water.

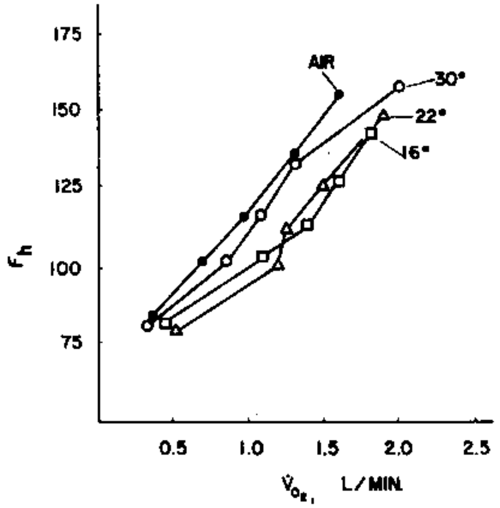


Fig. 4. Heart rate versus oxygen consumption during exercise in air and submerged in 30°, 22° and 16°C water.

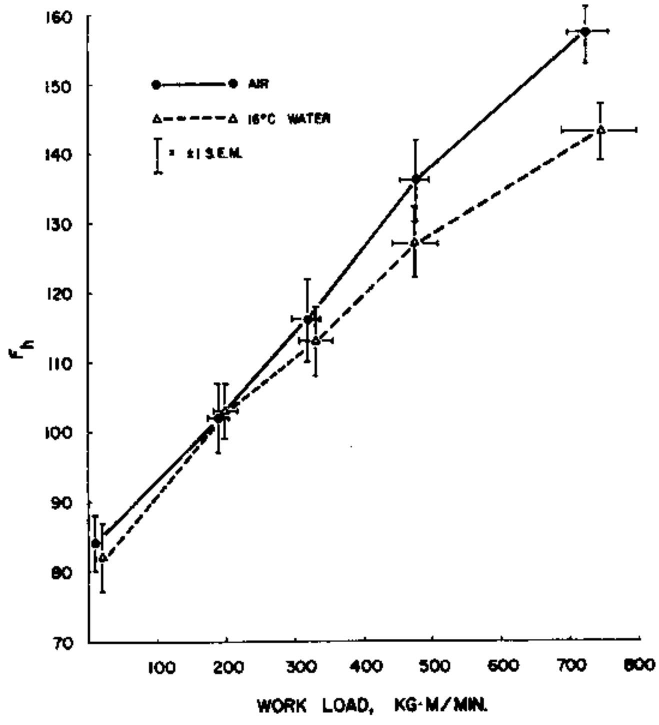


Fig. 5. Heart rate versus external work load during exercise in air and submerged in 16°C water.

slopes are also different (see Figure 5). It is possible that the peripheral vasoconstriction that occurs upon immersion in cold water[1] is maintained in 16°C water even throughout the work regimen and that heart rate is depressed via baroreceptor reflex mechanisms. In this context it would appear paradoxical that resting heart rate was not significantly lower in 16°C water. There was indeed, in most subjects, a bradycardia upon initial immersion in cold water, but f_h had returned to normal by the time (last 2 minutes) that the steady-state resting rate was recorded. Subjective impressions indicated that there was no cold discomfort during this period of rest as long as one remained very still. Upon initiation of exercise, however, the cold became quite apparent as the water was stirred up by leg motion. The possibility exists that, under the latter conditions, vasoconstriction became more pronounced.

A reasonable conclusion is that, under these extreme conditions, surface monitoring of diver's heart rate cannot be evaluated in surface-equivalent terms. In this experiment, at forced work levels in 16°C water, such evaluation would have underestimated the work load actually imposed. At 743.3 Kg • M/min the predicted heart rate would have been 161 beats/min, whereas it was actually measured at 143 beats/min. Therefore, some caution must be urged when surface monitoring under these conditions. There was no significant decrement in ability to perform the graded work task in water compared to air, although subjective reports were that it was becoming more difficult to maintain the forced work cadence underwater at the termination of the experiment.

Studies are currently underway to determine the effects of water depths to four atmospheres absolute upon work performance at various water temperatures.

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RESPIRATORY HEAT LOSS AND VENTILATION ON AIR AND HELIUM OXYGEN

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ABSTRACT

Convective, evaporative and total respiratory heat losses have been measured for $\dot{V}_E$ from resting levels to those simulating moderate work. Two gas mixes were inspired (air and heliox) at temperatures of 10 and 20C. Increasing $\dot{V}_E$ was achieved by selectively varying tidal volume (respiratory rate constant), respiratory rate (tidal volume constant) or both. While the most thermally efficient ventilation maneuver was an increase in $\dot{V}_E$ via increased rate of breathing only at each temperature, gas density was a major factor in determining total loss through the convective component. Predictive equations are given for heat loss versus respiratory maneuver, gas mix, and inspired gas temperature, with applications to hyperbaric environments.

Key words: Respiratory heat loss, heliox breathing, ventilation and heat loss, exercise and respiratory heat loss.

INTRODUCTION

For appropriate reasons of compromise in both physical and physiologic terms, helium has become the diluent gas of choice, replacing nitrogen, for exposures of men and experimental animals to the hyperbaric environment. However, the compromise is not without cost, in that the helium-oxygen (heliox) environment is one with a relatively high thermal conductivity and constant pressure specific heat (Cp). The thermal situation becomes aggravated at increased atmospheric pressures where density is increased and the impact of the resulting heat drain has been well described by Webb (1970), and others (Moore et al., 1974; Timbal et al., 1974).

While cutaneous heat losses are controlled practically by protective external insulation, both during habitat dwelling and water excursions, respiratory losses are more difficult to ameliorate and can constitute a significant portion of daily caloric intake (Moore et al., 1974; Timbal et al., 1974; Webb, 1970).

The objective of the experiments described here was to quantify the modes of respiratory heat loss as a function of gas mix, inspired gas temperatures, level of ventilation and manner of ventilation. All were performed at 1 ATA.

METHODS

Six adult male subjects participated in the experiments, and certain of their characteristics appear in Table 1. Minute ventilations ($\dot{V}_E$) from resting levels to those expected during moderate work (Moore et al., 1970; Weltman and Egstrom, 1969) were achieved via various combinations of alteration of either respiration rate (f_R) or tidal volume (V_T) or both.

Working $\dot{V}_E$ is simulated in these data. A comparison study was run on each subject exercising (bicycle ergometer) at 300, 600, and 900 Kg M/min to determine his own choice of $\dot{V}_E$. Based on these data, another exercise series was performed in which each diver controlled his ventilation at 12, 24, or 36 L/min at the appropriate work level via the methods described below.

In terms of respiratory heat loss, there was no difference between losses at a given $\dot{V}_E$ while exercising or simulated without exercise, so long as no change in core temperature occurred. Therefore, all subsequent experiments were with simulated $\dot{V}_E$ corresponding to measured work loads.

The subjects were trained to ventilate at given levels in given ways with reasonable accuracy. Frequency of breathing was regulated by metronome while V_T was monitored by the subject from a dry gas meter in series with the expired air hose. All maneuvers were maintained for five minutes, and $\dot{V}_E$ was measured during the last two minutes in a 350 L Tissot spirometer (Collins, Inc., Boston, Massachusetts). Table 2 depicts the ventilatory combinations attempted. Two gas mixes were utilized; compressed air (79% N_2: 20.9% O_2) and heliox 79.7% He: 20.3% O_2) as analyzed by micro-Scholander technique. All mixes were delivered via standard double hose, double-stage SCUBA (U.S. Divers, Santa Ana, California) with appropriate monitoring of inspired and expired air temperatures and relative humidities. All studies were carried out in a temperature controlled environmental room (Forma Scientific, Marietta, Ohio) at either 20C or 10C. SCUBA bottles were immersed in water of 20C or 10C in order to aid in control of inspired gas temperatures.

Respiratory heat loss was partitioned into the evaporative and convective components utilizing the following equations:

$$\text{Convective Loss } (\dot{Q}\text{conv}) = \dot{V}_E \times \rho \times C_p \times (T_E - T_I) \qquad \text{(Equation 1)}$$

$$\text{Evaporative Loss } (\dot{Q}\text{evap}) = \dot{V}_E \times (\Delta P_{H_2O}/760) \times (18/22.4) \times (273/273 + T_{amb}) \times 580 \qquad \text{(Equation 2)}$$

Where:

$\dot{V}_E$ = minute volume, L/min BTPS
T_E = expired gas temperature, °C
T_I = inspired gas temperature, °C
T_{amb} = ambient temperature, °C
$\Delta P_{H_2O} = (P_{H_2O})\text{ exp} - (P_{H_2O})\text{ insp}$, mmHg
760 = 1 ATA pressure, mmHg

Subject	Age (yrs.)	Ht. (cm)	Wt. (Kg)	Surface area (M^2)	V_{O2} max (L/min)*
TM	39	186.7	75.5	2.0	3.26
JM	25	184.2	69.2	1.9	3.89
RD	29	178.6	69.3	1.9	4.89
MS	23	172.2	68.0	1.8	2.87
RS	29	172.7	64.0	1.8	2.73
DB	25	180.3	74.9	1.9	4.49
$\bar{X}$	28.3	179.1	70.2	1.9	3.69
S.E.	2.3	2.4	1.8	0.03	1.36

Table 1. Characteristics of experimental subjects.
* Treadmill method (3)

Combination	f_R (breaths/min)	V_T (L)	$\dot{V}_E$ (L/min-approximate)
1	12	1.0	12
2	12	2.0	24
3	12	3.0	36
4	24	1.0	24
5	36	1.0	36
6	16	1.5	24
7	18	2.0	36

Table 2. Ventilatory combinations of V_T and f_R attempted to control $\dot{V}_E$.

273 = $0^{\circ}C$ in absolute temperature scale, $^{\circ}K$
18 = molecular weight of H_2O
22.4 = molar volume of H_2O, L/mole
580 = heat of vaporization of H_2O, cal/gm
ρ = density of gas mix, gm/liter
C_p = constant pressure specific heat, cal/gm$^{\circ}$C

The product of the gas mixture was calculated by weighting each of the individual component gases $\rho \times C_p$ product (1) by its fractional volume percent. In terms of ideal gases, the heat carrying capacity on a unit volume basis ($\rho \times C_p$) is directly proportional to absolute pressure and inversely proportional to absolute temperature. This statement is derived as follows:

A) Density of ideal gas (2)

$$\rho = (PM)/(RT) \qquad \text{(Equation 3)}$$

Where:

P = absolute pressure, ATA
M = molecular weight, gm/mole
R = ideal gas constant
1.986 cal/mole$^{\circ}$K or 0.82057 L x ATA/mole$^{\circ}$K
T = absolute gas temperature $^{\circ}$K

B) Specific heat of ideal gas (2).

$$\begin{aligned} C_p &= (5/2) \times (R/M);\ \text{for a monatomic gas} \qquad \text{(Equation 4)} \\ &= (7/2) \times (R/M);\ \text{for a diatomic gas} \\ &= (8/2) \times (R/M);\ \text{for a polyatomic gas} \end{aligned}$$

Combining equations 3 and 4 results in the density - specific heat product of an ideal gas being a function of only absolute temperature, pressure, and the number of atoms per molecule (Equation 5).

$$\begin{aligned} \rho C_p &= (5/2) \times (P/T);\ \text{for a monatomic gas} \qquad \text{(Equation 5)} \\ &= (7/2) \times (P/T);\ \text{for a diatomic gas} \\ &= (8/2) \times (P/T);\ \text{for a polyatomic gas} \end{aligned}$$

The result is ρC_p in ATA/$^{\circ}$K. Conversion to cal/liter$^{\circ}$K, obtained upon multiplication of ρ in grams/liter and C_p in cal/gram$^{\circ}$K, involves the following steps:

$$(1\text{ATA}/^{\circ}\text{K}) \times (1.0132 \times 10^{5}\text{Newton/meter}^{2})/(\text{ATA}) \times (1.0 \times 10^{-3}\text{meter}^{3}/\text{liter}) \times (1\ \text{joule/Newton} \times \text{meter}) \times (.2389\ \text{cal/joule}) = \text{cal/liter}^{\circ}\text{K}$$

or simplified

$$1\ \text{ATA}/^{\circ}\text{K} \times 24.205 = \text{cal/liter}^{\circ}\text{K}.$$

RESULTS AND DISCUSSION

The absolute caloric costs for the experimental conditions are found in Table 3, based on breathing pattern.

At inspired temperatures of approximately 20C, total heat losses are not statistically different between heliox and air breathing for any respiratory maneuver. However, there were lower convective losses breathing heliox during several breathing patterns (Table 3), mostly involving elevated ventilations.

At T_I of approximately 10C there were again only minor differences in terms of $\dot{Q}_{total}$, and few statistical differences between breathing patterns (Table 3). Ventilation levels were not different for any given breathing maneuver, whether on air or heliox.

For similar ranges, the data appear in Figure 1 normalized to heat loss per liter ventilated, and separated into convective and evaporative components. The major portion of heat loss at 1 ATA is evaporative. At the higher inspired gas temperatures, there are both qualitative and quantitative similarities in the pattern of total heat loss in terms of breathing maneuver. Convective losses are greater breathing air, as would be predicted by equation 1, on the basis of the higher heat carrying capacity per unit volume ($\rho \times C_p$) of air versus heliox at 1 ATA.

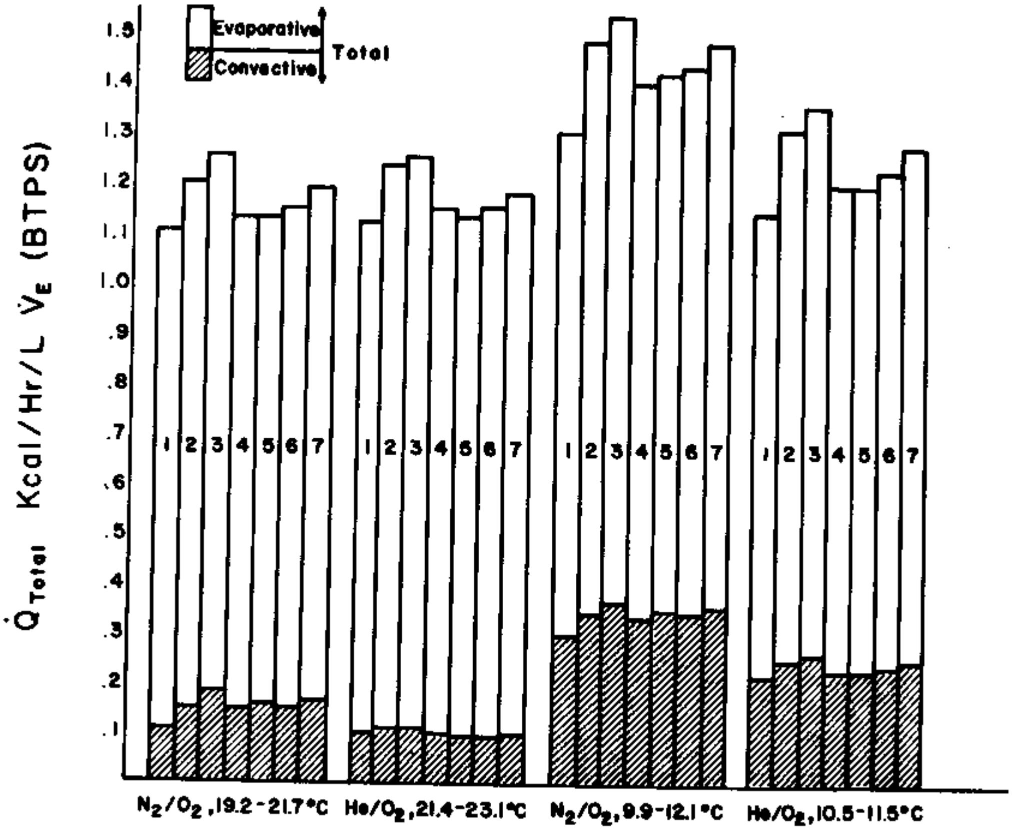

Figure 1. Total respiratory heat loss versus ventilatory pattern and gas mix. Data are normalized to heat loss per liter of gas ventilated.

Condition	Ventilatory Combinations	$\dot{V}_E$ (actual, L/min)	$\dot{Q}$ evap (Kcal/Hr)	$\dot{Q}$ conv (Kcal/Hr)
N_2/O_2	REST	8.83 ± 1.16 (S.E.)	8.32 ± 1.04	1.02 ± 0.18
T_I = 19.2-21.7°C	1	12.76 ± 0.29	12.73 ± 12.73	1.59 ± 0.09
	2	25.35 ± 0.65	26.82 ± 0.79	2.87 ± .17*
	3	37.82 ± 0.92	40.85 ± 1.13	6.94 ± 0.18*
	4	25.50 ± 0.36	25.20 ± 0.54	3.85 ± 0.08*
	5	37.16 ± 1.25	36.53 ± 1.45	5.99 ± 0.3*
	6	24.09 ± 0.47	24.42 ± 0.74*	3.64 ± 0.20*
	7	37.50 ± 0.64	38.38 ± 1.20*	6.30 ± 0.28
N_2/O_2	REST	9.73 ± 1.30	8.82 ± 1.40	2.57 ± 0.39
T_I = 9.9-12.1°C	1	15.43 ± 1.03	15.65 ± 0.89	4.56 ± 0.32
	2	28.54 ± 0.53	32.66 ± 0.99	9.75 ± 0.38*
	3	40.43 ± 1.85	47.50 ± 2.62	14.77 ± 0.89
	4	26.99 ± 0.43	29.14 ± 0.75	8.91 ± 0.36
	5	39.42 ± 0.55	42.58 ± 0.99	13.62 ± 0.50*
	6	24.59 ± 0.99	26.95 ± 0.91	8.51 ± 0.28

Table 3. Mean respiratory heat losses during air or heliox breathing at different temperatures and different breathing patterns. Ventilatory combinations are numbered as listed in Table 2.

Condition	Ventilatory Combinations	$\dot{V}_E$ (actual, L/min)	$\dot{Q}$ evap (Kcal/Hr)	$\dot{Q}$ conv (Kcal/Hr)
	7	38.50 ± 1.49	43.54 ± 2.06	14.05 ± 0.75*
He/O_2	REST	8.30 ± 1.00	7.70 ± 1.00	0.76 ± 0.14
T_I = 21.4-23.1°C	1	12.35 ± 0.19	12.73 ± 0.20	1.28 ± 0.05
	2	25.54 ± 0.64	28.73 ± 0.75	3.02 ± 0.10
	3	37.34 ± 0.19	42.47 ± 0.51	4.36 ± 0.12
	4	26.05 ± 0.21	27.56 ± 0.55	2.52 ± 0.10
	5	38.36 ± 0.39	39.88 ± 1.07	3.39 ± 0.16
	6	25.60 ± 0.21	27.45 ± 0.59	2.43 ± 0.12
	7	38.10 ± 0.24	41.28 ± 0.90	3.66 ± 0.20
He/O_2	REST	8.82 ± 0.85	6.80 ± 0.70	1.64 ± 0.16
T_I = 10.5-11.5°C	1	14.26 ± 0.20	13.26 ± 0.36	3.15 ± 0.13
	2	28.58 ± 0.41	30.70 ± 0.29	7.18 ± 0.19
	3	42.63 ± 0.65	47.50 ± 1.32	11.10 ± 0.32
	4	26.65 ± 0.76	26.14 ± 1.08	6.08 ± 0.31
	5	37.46 ± 0.65	36.75 ± 1.13	8.63 ± 0.34
	6	27.21 ± 1.27	27.31 ± 1.51	6.40 ± 0.43
	7	42.87 ± 2.77	44.63 ± 2.83	10.55 ± 0.81

Table 3. Continuation. *Significantly different from similar maneuver breathing heliox at $P < 0.05$.
All statistical inferences were based on paired t analysis.

The differences in heat loss become exaggerated at the lower inspired gas temperatures, again due to increases in convective loss, but largely through an increase in the $T_E - T_I$ gradient.

In all cases, the most efficient breathing pattern (in terms of respiratory heat loss) for increasing $\dot{V}_E$ was an increase in f_R with constant V_T. Constant f_R and increasing V_T was the least efficient, with an increase in both being intermediate. The highest efficiency is gained apparently by reducing the time interval for heat exchange between inspired air and respiratory tract.

The correlation coefficients and regression equations for $\dot{V}_E$ versus heat loss as a function of ventilatory pattern appear in Table 4. The regressions are linear for the range of $\dot{V}_E$ encountered in this experiment.

The highest total cost occurs breathing cold air, regardless of method, compared to heliox. During deep dives, the switch to helium diluent only lessens the impact of the density factor, but, combined with the high specific heat of the mix, the convective heat losses become prohibitive at high ventilations. Evaporative losses change relatively little. In a saturation experiment previously reported (Moore et al., 1974), respiratory heat losses for $\dot{V}_E$ from 10 to 26 L/min inside a habitat could be quantified as follows:

Depth		8 ATA	16.1 ATA
$He/N_2/O_2$ mix	=	91.6/6.2/2.4%	95.7/3.1/1.2%
$\dot{Q}$ conv.	=	$0.50\ (\dot{V}_E) + 0.5$	$1.19\ (\dot{V}_E) + (-2.0)$
$\dot{Q}$ evap.	=	$0.47\ (\dot{V}_E) + 0.8$	$0.52\ (\dot{V}_E) + 0.6$
$\dot{Q}$ total	=	$0.91\ (\dot{V}_E) + 1.3$	$1.70\ (\dot{V}_E) + 3.9$

These data compare favorably with losses predicted by atmospheric density changes with depth, especially as regards the convective component, i.e.

Predicted $\dot{Q}_{total}$ at 16.1 ATA at $\dot{V}_E$ of 10 L/min = 12.5 Kcal/Hr. The loss actually measured was 14 Kcal/Hr.

In summary, equations have been experimentally determined for the estimate of respiratory heat loss with increasing ventilations on air and heliox at 2 inspired gas temperatures. The least thermally wasteful method of increasing ventilation is by increasing breathing rate rather than tidal volume. Combined with the general equations for convective and evaporative heat loss, these data may be used to extrapolate to depth, as long as the ambient environmental characteristics are known.

Gas mix	Ventilatory Combination	$\dot{Q}$ conv	R	$\dot{Q}$ evap	R	$\dot{Q}$ Total	R
N_2/O_2	1,2,3	$= 0.21\ (\dot{V}_E) + (-1.2)$	0.986	$= 1.11\ (\dot{V}_E)+(-1.3)$	0.997	$= 1.33\ (\dot{V}_E)+(-3.1)$	0.997
$T_I = 19.2\text{-}21.7^{\circ}C$	1,4,5	$= 0.18\ (\dot{V}_E) + (-0.7$	0.992	$= 0.98\ (\dot{V}_E)+0.2$	0.978	$= 1.16\ (\dot{V}_E)+(-0.6)$	0.996
	1,6,7	$= 0.19\ (\dot{V}_E) + (-0.8)$	0.996	$= 1.04\ (\dot{V}_E)+(-0.6)$	0.992	$= 1.23\ (\dot{V}_E)+(-1.4)$	0.994
He/O_2	1,2,3	$= 0.12\ (\dot{V}_E) + (-0.2)$	0.981	$= 1.19\ (\dot{V}_E)+(-1.9)$	0.997	$= 1.31\ (\dot{V}_E)+(-2.2)$	0.996
$T_I = 21.4\text{-}23.1^{\circ}C$	1,4,5	$= 0.08\ (\dot{V}_E) + 0.3$	0.964	$= 1.05\ (\dot{V}_E)+(0.8)$	0.994	$= 1.13\ (\dot{V}_E)+0.2$	0.993
	1,6,7	$= 0.09\ (\dot{V}_E) + 0.1$	0.962	$= 1.11\ (\dot{V}_E)+(-1.0)$	0.995	$= 1.21\ (\dot{V}_E)+(-1.0)$	0.994
N_2/O_2	1,2,3	$= 0.41\ (\dot{V}_E) + (-1.8)$	0.989	$= 1.27\ (\dot{V}_E)+(-3.9)$	0.996	$= 1.68\ (\dot{V}_E)+(-5.8)$	0.996
$T_I = 9.9\text{-}21.1^{\circ}C$	1,4,5	$= 0.38\ (\dot{V}_E) + (-1.2)$	0.982	$= 1.11\ (\dot{V}_E)+(-1.3)$	0.990	$= 1.49\ (\dot{V}_E)+(-2.5)$	0.989
	1,6,7	$= 0.41\ (\dot{V}_E) + (-1.6)$	0.991	$= 1.20\ (\dot{V}_E)+(-2.6)$	0.994	$= 1.60\ (\dot{V}_E)+(-4.2)$	0.995
He/O_2	1,2,3	$= 0.28\ (\dot{V}_E) + (-3.8)$	0.992	$= 1.20\ (\dot{V}_E)+(-3.8)$	0.992	$= 1.48\ (\dot{V}_E)+(-4.6)$	0.993
$T_I = 10.5\text{-}11.5^{\circ}C$	1,4,5	$= 0.24\ (\dot{V}_E) + (-0.3)$	0.986	$= 1.02\ (\dot{V}_E)+(-1.3)$	0.993	$= 1.26\ (\dot{V}_E)+(-1.6)$	0.993
	1,6,7	$= 0.26\ (\dot{V}_E) + (-0.7)$	0.992	$= 1.09\ (\dot{V}_E)+(-2.2)$	0.995	$= 1.35\ (\dot{V}_E)+(-2.9)$	0.996

Table 4. Regression equations and correlation coefficients (R) for respiratory heat losses as a function of $\dot{V}_E$ and breathing pattern. Ventilatory combinations are as defined in Table 2.

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THERMAL COST OF THE HYPERBARIC ENVIRONMENT

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INTRODUCTION

Since the early 1960's, it has been demonstrated that man can dwell and do useful work in the undersea hyperbaric environment. "Mixed-gas" technology has largely solved most of the physiologic/medical problems associated with high pressure breathing gas environments. The two immediate problems being 1) the narcotic effect of some inert gases at high partial pressure (Bennett, 1969) and 2) the increased density of inert gases under pressure leading to increased ventilatory effort with consequent limitations on the diver's ability to perform useful work tasks (Miller et al., 1971).

The difficulties above may be ameliorated by substitution for the nitrogen in air of a diluent inert gas with low narcotic effect and low density. The substitute gas most often used currently is helium, which, at this writing, has been successfully utilized in chamber divers to 2001 feet (Anonymous, 1972), and in "wet" (open sea excursion) divers to 1010 feet of sea water (Anonymous, 1972), Table 1 gives the relevant physical characteristics of several inert gases which have been used in connection with hyperbaric exposure.

	Thermal Conductivity (BTU/hr-ft-^{o}F)	Kinematic Viscosity (ft^2/sec)	Density (Lbm/ft^2)	Specific Heat (BTU/Lbm^{o}F)
Helium	0.0861	136.4×10^{-5}	0.01045	1.2420
Nitrogen	0.0151	16.8×10^{-5}	0.0713	0.2486
Argon	0.0102	14.9×10^{-5}	0.1014	0.1253
Neon	0.0280	41.4×10^{-5}	0.0512	0.2460
Hydrogen	0.1050	117.9×10^{-5}	0.0051	3.4190

Table 1: Selected Physical Properties of Diving Inert Gases at 1 Ata and 26.6^{o}C (80^{o}F)

Considering in addition that the narcotic potency of these gases is (from high to low) Argon > Nitrogen > Hydrogen > Neon > Helium (1), it is obvious that helium is a reasonable compromise both from the standpoint of physical properties and cost.

However, helium has certain disadvantages in another, very practical, aspect. Both the heat capacity and thermal conductivity of this gas indicate that body heat loss is a potential problem area to working divers. That the problem is not only potential but actual is the subject of the following discussion.

THEORETICAL ASPECTS OF HEAT LOSS

For man dwelling in the surface (1 Ata) air environment, heat production is achieved largely by metabolism of nutritional constituents and balanced for thermoneutrality by heat loss via several different routes, e.g., conduction, convection, radiation and evaporation.

The overall equation for heat exchange (H) between man and his environment is:(2)

$$H = M \pm S$$
$$= E \pm K \pm C \pm R + W$$

where

S = heat storage
M = metabolic heat production
E = evaporative heat loss
K = conductive heat exchange (negative for heat gain by body)
C = convective heat exchange (negative for heat gain by body)
R = radiant heat exchange (negative for heat gain by body)
and W = external work performed

When the body is resting, $W = 0$, and thermal equilibrium, $S = 0$, heat loss from the skin surface is:

$$H = M = h_{kCRS} (T_s - T_a) + E$$

where

h_{kCRS} = heat transfer coefficient for K, C, and R
E = (gm of water evaporated) x (heat of vaporization at T_s)
T_s = skin temperature
and T_a = ambient temperature

Under hyperbaric conditions including chamber simulations, convective losses, normally a significant pathway in 1 Ata air, becomes the dominant route of loss in helium rich environments (Webb, 1970). While the other routes are not particularly dependent upon variables occurring with increased pressure, the convective term is, viz.

$$C = \frac{hc}{d} \quad 1 + a \frac{(dv\rho)}{\mu}^{1/2} + b \frac{(dv\rho)}{\mu} \quad (T_s - T_a)$$

where

d = characteristic dimension of the object (e.g., diameter of a sphere or cylinder)
v = velocity of the air or fluid
ρ = density of the air or fluid
μ = viscosity of the air or fluid
a & b = constants determined by the units used
h_c= convective transfer coefficient

Webb (1970) has derived relative constants for various ambient media illustrating the importance of convective characteristics under typical hyperbaric operational conditions. Two derived constants are presented:

1) Convective constant, which is the product of density (ρ) in gm/liter, specific heat (Cp) in cal/gm-°C, and thermal conductivity (K) in cal/min-cm-°C, divided by viscosity in centipoises.

2) Convective character, which is a ratio normalizing fluids to air by dividing the convective constants of the fluid by those for air at 1 Ata. Table 2 depicts the convective character of several fluids at an ambient temperature of 28°C.

	Convective Constant	Convective Character
Air at 1 Ata	0.61	1
98.3% He-1.7% O_2 at 20.7 Ata	52.0	85.2
Water at 1 ATA	102	167

Table 2: Convective Character of Several Fluids at 28°C

Before proceeding to an examination of convective heat losses generally, consideration of an additional type of loss is relevant. This is the heat loss from the respiratory tract.

During ventilation, air (or any breathing medium) inspired is both heated and humidified before expiration. Thus, an additional heat loss can be incurred. The equations below illustrate the convective and evaporative terms of this loss.

$$\text{Convective loss} = \dot{V}_E \cdot \rho \cdot C_p \cdot (T_E - T_I)$$

where

$\dot{V}_E$ = minute vol in L/min ATPS
ρ = gas density in gm/L
C_p = specific heat of the gas in Kcal/gm .°C

T_E = temperature of expired gas
and T_I = temperature of inspired gas

$$\text{Evaporative loss} = V_E \cdot \frac{\Delta P_{H_2O}}{760} \cdot \frac{18}{22.4} \cdot \frac{273}{273 + T_a} \; .580$$

where

ΔP_{H_2O} = vapor pressure of water in Torr
= $(P_{H_2}O)$ expired - $(P_{H_2}O)$ inspired
760 = 1 atm of pressure in Torr
18 = molecular weight of water in gm/mol
22.4 = molar volume of water in L/mol
273 = absolute temperature
T_a = ambient temperature in ^{o}C
580 = heat of vaporization in cal/gm

Considering then the general whole body convective loss and the special respiratory loss, it is obvious that both will be exacerbated in an environment of high density (ρ), high specific heat (C_p) and hence, high heat capacity (ρC_p).

We can predict, therefore, on theoretical grounds, that the hyperbaric helium environment would constitute one in which a depth related heat drain could occur. We will proceed to examine this in quantitative terms under an actual field operation.

Experimental Data (5)

In January of 1972, 6 divers completed a 500' dry dive in the mobile habitat "Aegir" of Makai Range, Inc. on Oahu, Hawaii as first in a series of dives which later entail wet dives with controlled water excursions and work levels.

Aegir consists of 3 chambers - one a laboratory/galley - one a diving ready room - one living quarters. Each chamber has separate and autonomous life support systems. The detailed features have recently been reported by Pegg (1971). Dive profile: As shown in Figure 1, beginning at 1900 hours on Day 1, pressurization was begun. The first overnight stop was made at 250 feet (approximately 8.6 ATA). After psychomotor testing during the remaining evening hours, the divers slept for 6 hours. At 0600 the normal regimen began. Vital signs - pulse rate, blood pressure, oral temperature and body weight were recorded, followed by breakfast.

After breakfast, the subjects were taken in turn (standardized) for measurement of heat loss. Each subject, with rectal temperature probe in place and clad only in light shorts, sat quietly on a bicycle ergometer (Monark, Varberg, Sweden) for 15 or 20 minutes. He then breathed normally through a standard commercial mouthpiece double hose assembly (U.S. Divers, Santa Ana, California)

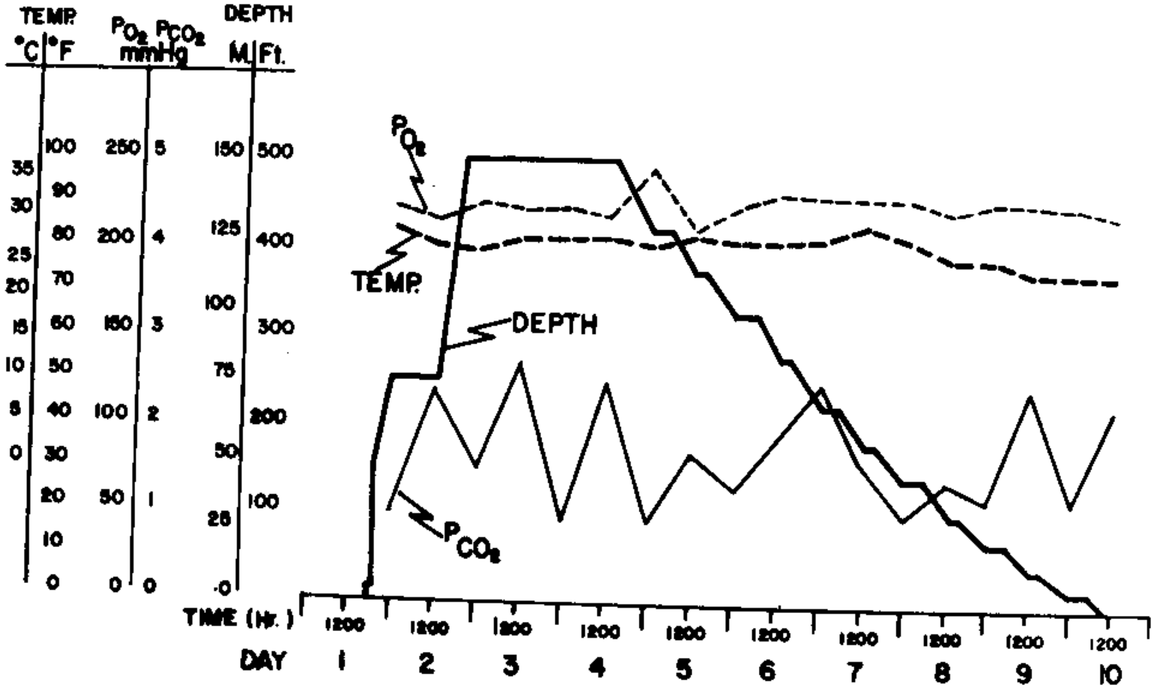

Figure 1. Experimental dive profile.

whose intake side had been shortened to approximately 2 inches upstream of the intake valve. Inserted into this segment was a hygrometer sensing probe, thermistor and thermocouple probe. The exhaust side (just downstream from the exhaust valve) was similarly equipped, with the exhaust hose terminating at a connection with a Douglas bag valve for expired gas collection. Intake and exhaust relative humidities, rectal temperatures, and intake air temperatures were recorded in the habitat at appropriate intervals during the experimental regimen. The EMF difference between intake and exhaust thermocouples was hard-wired to a polygraph located in the main control room outside the habitat, as was EKG for heart rate count.

During the last 2-3 minutes of the rest period, expired gas was collected and EMF, rectal and ambient temperatures, and relative humidities were recorded. The Douglas bag was then emptied through a horizontal plexiglass mixing chamber (approximately 12 inches long, 4 inches diameter) which was fitted and sealed at the bottom over the sensing probe of a P_{O_2} meter.

In series downstream from the mixing chamber was an attached gas flowmeter (A.H. Thomas Co., Philadelphia). Approximately half-way through the bag emptying procedure, the input and output sides of the mixing chamber was clamped off and the P_{O_2} of the contained gas recorded (approximately 30-40 sec equilibration time was required). The bag volume was then completely emptied and the expired volume recorded from the flowmeter. The P_{O_2} meter was checked daily against a range of calibration gases carried on board. (These had been analyzed previously by the micro-Scholander technique in the laboratory). $\dot{V}_E$ and $\dot{V}_{O_2}$ (O_2 consumption, L/min) were determined from these procedures.

Simultaneously with the expired gas collections, temperature and heat flux were recorded at several points on the body surface of the subject: forehead, chest, abdomen, upper back, upper arm, forearm, hand, thigh, calf, and foot. These were monitored by use of an exploring probe consisting of a thermistor and heat flow disc mounted side-by-side in a lucite plate. The plate was attached to a handle and spring loaded (with a marker) so that uniform tension was applied in each case. Each subject was indelibly marked at each of the various skin points for consistency throughout the dive duration. Skin temperatures were recorded via telethermometer (Yellow Springs Instruments, Yellow Spring, Ohio) in the habitat while heat flux was monitored on the external polygraph.

The subject then proceeded with mild exercise (67 watts) for 10 minutes, at the end of which (last 2 min) the recording procedures above were repeated, and during recovery from minutes 2-3, 5-6, and 9-10.

One ATA control values were gathered in the unpressurized habitat in the control days just prior to the dive.

At 1300 hours on Day 2, after 17 hours at 250 feet depth, pressurization continued to a depth of 500 feet, where the divers remained for 43 hours. The experimental protocol was followed as described above while at this depth.

Decompression began at 1400 hours on Day 4 (Standard U.S. Navy helium Schedule), with overnight holds from 0000-0600 hours and a rest stop from 1400-1600 hours daily, until surfacing at 1200 hours on Day 10. The experimental procedures (minus the exercise portion) were adhered to until the habitat reached 150 feet (late on Day 7) when the habitat went on "power-down" condition to pass through the fire hazard zone. From that point, only the morning vital signs were monitored.

Table 3 describes the thermal characteristics of the environment, indicating the change in heat capacity (ρC_p) associated with the elevated pressure.

Skin heat fluxes are depicted in Figure 2 for both rest and exercise at the various simulated depths. For the resting condition, the salient points appear to be an alteration in distribution and degree of flux upon exposure to the heliox environment at depth. For instance, while forehead flux appears high under all conditions, a notable decrease, especially in the distal extremities, (especially foot, and to a lesser extent, upper arm, and hand below 250 feet) was found to occur. On the other hand, the back, lower arm, thigh and lower leg were found to have relatively higher heat flux in the heliox environment at all depths.

During exercise, the qualitative pattern was not dissimilar to the resting condition; high fluxes from back, lower arm, thigh and lower leg, with hand and foot (especially the latter) showing low heat flows.

Physical Characteristics	Air at Surface	250 Feet (He 96.6%) (O_2 3.4%)	500 Feet (He 98.2% (O_2 1.8%)
Density (ρ), gm/liter	1.11	1.63	2.82
(Relative ρ)	(1.0)	(1.5)	(2.5)
Specific heat (C_p) cal/gm/°C	0.26	1.11	1.17
(Relative C_p)	(1.0)	(4.3)	(4.5)
Heat capacity (ρC_p) Cal/liter/°C	0.29	1.82	3.32
(Relative ρC_p)	(1.0)	(6.2)	(11.3)

Table 3: Thermal Characteristics of Air at Surface and He-O_2 at Hyperbaric Environment

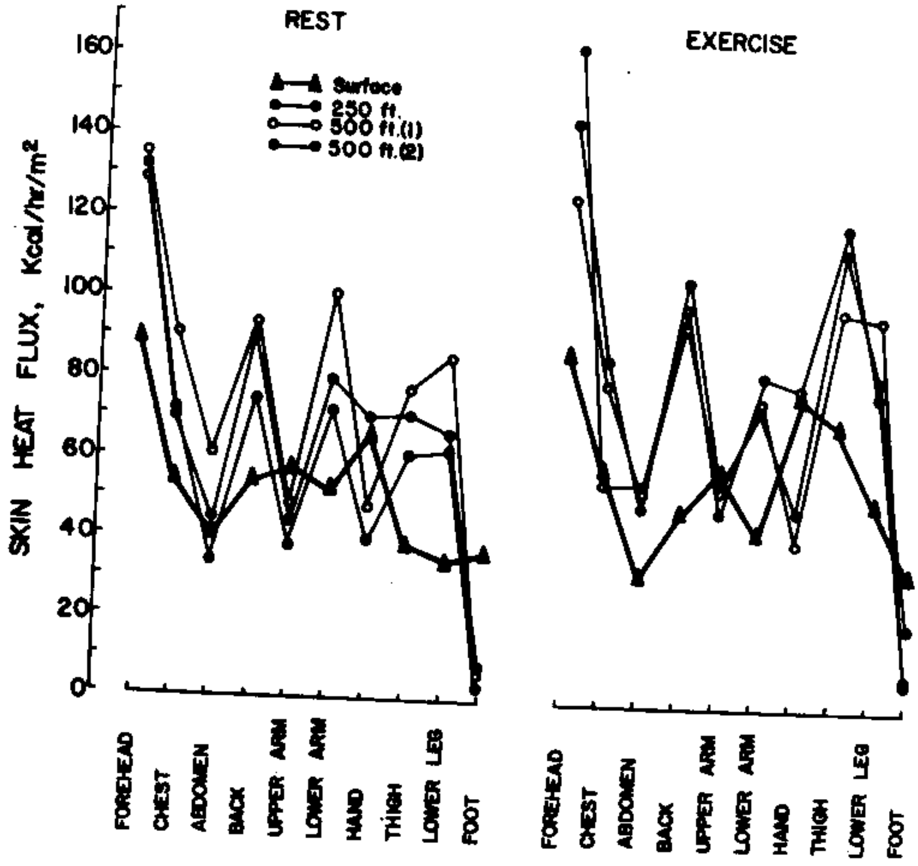

Figure 2. Cutaneous heat flux during rest and exercise as a function of depth.

Figure 3 represents the respiratory heat loss components during the experimental conditions of rest, exercise, and these recovery points. There was no difference between the first and second days at 500 feet, so the data represent the second day at that depth. The major feature of these data is the fact that with increasing depth and consequent increasing ρC_p of the ambient heliox, the convective term of respiratory loss increases dramatically while only small increases are seen in the evaporative term. This is in marked contrast to the case of air at 1 ATA, where losses are almost completely evaporative. The respiratory "thermal" recovery was essentially complete after 5 minutes from termination of exercise at this level.

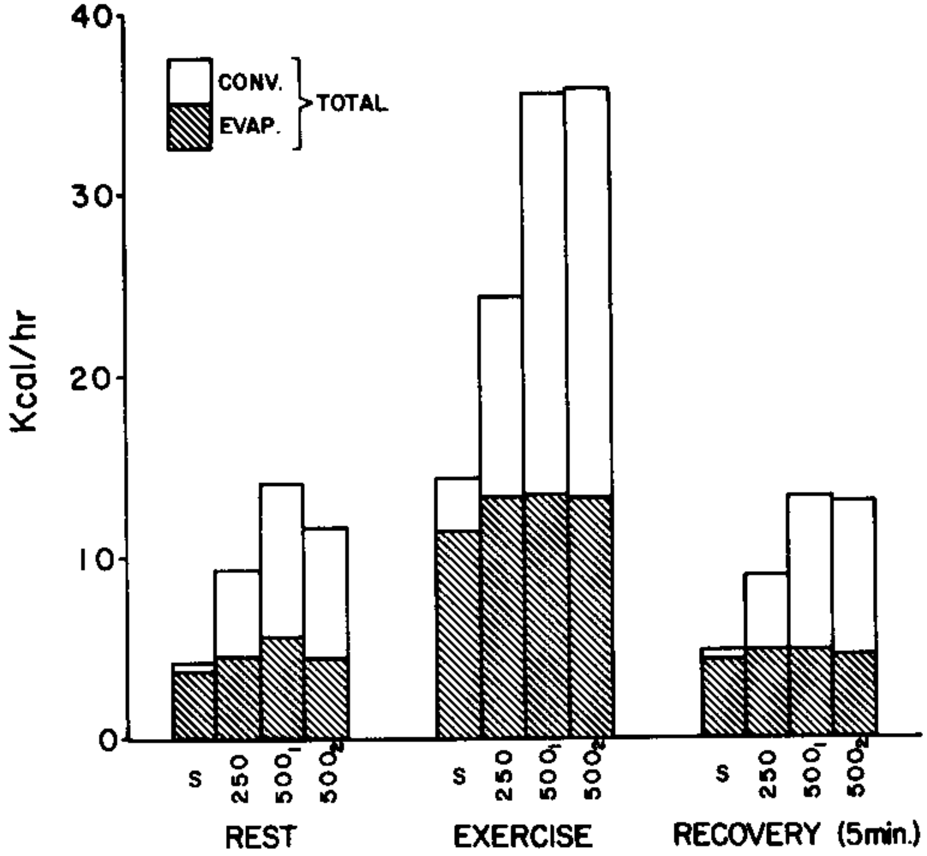

Figure 3. Convective and evaporative respiratory heat loss during rest, exercise, and recovery as a function of depth.

Considering then the overall pattern of heat exchange under these conditions, Table 4 provides a summary during rest and exercise. At rest, mean skin temperature (Hong et al., 1969) falls steadily through the first day at 500 feet; rises slightly on the second, presumably via the addition of heat to the chamber environment, when the chamber heaters were turned on. Mean body temperature follows the same pattern. Heat production, as gauged by metabolic rate, rises to 90.8 Kcal/hr/m^2, then drops on Day 2 at depth, while heat loss, both cutaneous and respiratory, follows the same qualitative pattern. The net result is a negative heat balance for all days except 500' (1) where metabolic heat production peaks. During exercise, positive heat balance was maintained throughout, even in the face of higher heat loss, through a higher level of metabolic heat production. Again, the inflection from 500' (1) to 500' (2) was seen.

	Depth (feet)			
Heat Exchange	0	250	500(1)	500(2)
Chamber temperature, °C	29.0	27.8	27.8	29.0
Chamber humidity, %	78	68	72	76
A. Rest				
Rectal temperature, °C	37.43	36.91	36.66	37.04
Mean skin temperature, °C	33.54	31.01	30.13	30.88
Mean body temperature, °C	36.14	34.96	34.50	35.00
Heat production, Kcal/hr/m^2	44.7	55.7	90.8	60.6
Heat loss, Kcal/hr/m^2	52.3	74.1	87.0	67.5
Heat balance, Kcal/hr/m^2	-7.6	-18.4	+3.8	-6.9
B. Exercise 67 watts				
Rectal temperature, °C	37.38	36.83	36.80	36.96
Mean skin temperature, °C	33.26	30.78	30.06	30.86
Mean body temperature, °C	36.02	34.83	34.57	34.94
Heat production, Kcal/hr/m^2	191.5	183.5	251.0	203.5
Heat loss, Kcal/hr/m^2	64.5	99.2	103.0	107.5
Heat balance, Kcal/hr/m^2	+127.0	+84.3	+148.0	+96.0

Table 4: Heat Exchange at Rest at Exercise

It can be seen in Figure 4 that the relative contribution of respiratory heat loss to total loss increased predictably with depth and was exacerbated with exercise to the point of comprising approximately 20% of total loss even under the mild work load imposed. Skin flux decreased overall as skin temperature decreased. Figure 5 shows the narrowing of the skin to air gradient under conditions of rest, exercise and at the 5 minute recovery point, when associated with increasing depth. Again, on the second day at 500 feet, the gradient narrows as T_a was raised.

In summary, then, experimental results follow theoretical predictions with regard to the very real, and perhaps limiting problem of thermal cost to the working diver. If one also considers the additional cold stress imposed with attendant exposure to cold water during excursions from an undersea vehicle, it is apparent that diver life support engineering requires more than appropriate provision of oxygen and elimination of carbon dioxide. Based on experimental data relative to respiratory heat losses alone, it has recently been proposed that a method of heating respired gas must be found (Anonymous, 1972), and the minimum breathing gas temperatures as a function of depth will be required to avoid "significant" (degree unspecified) respiratory heat losses.

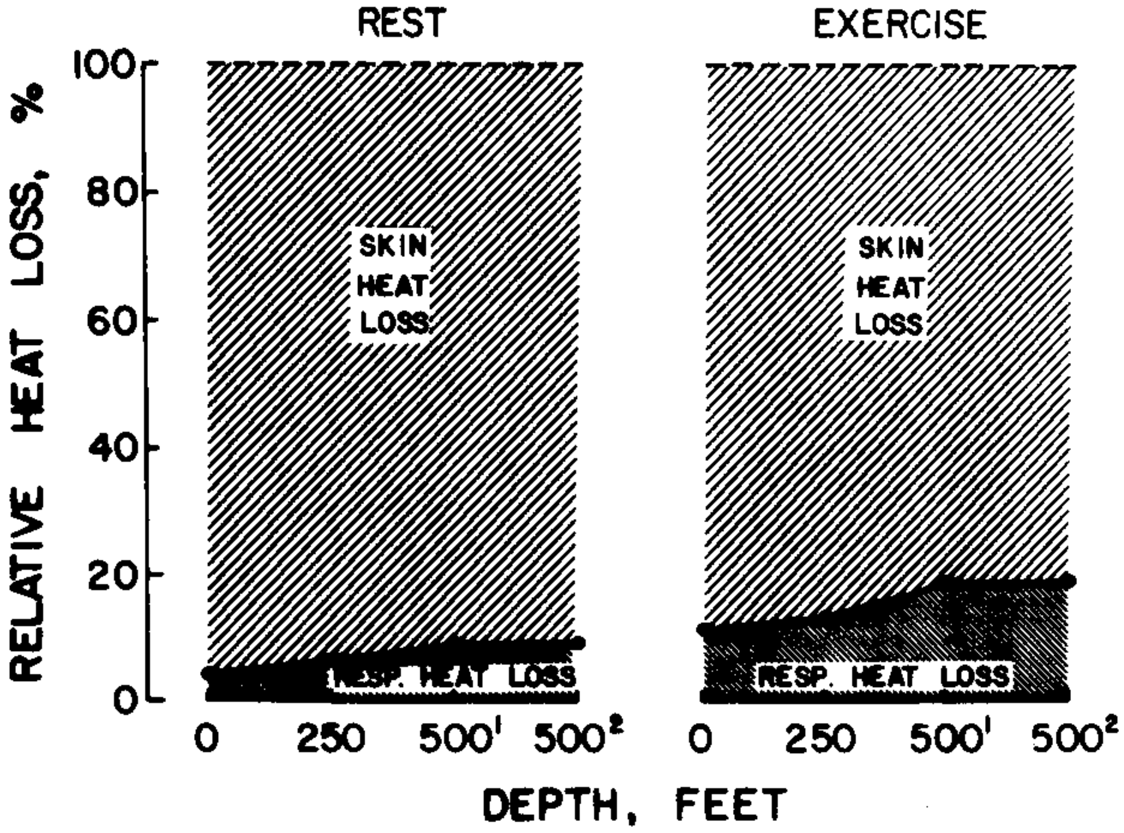

Figure 4. Respiratory heat loss as proportion of total loss during rest and exercise as a function of depth.

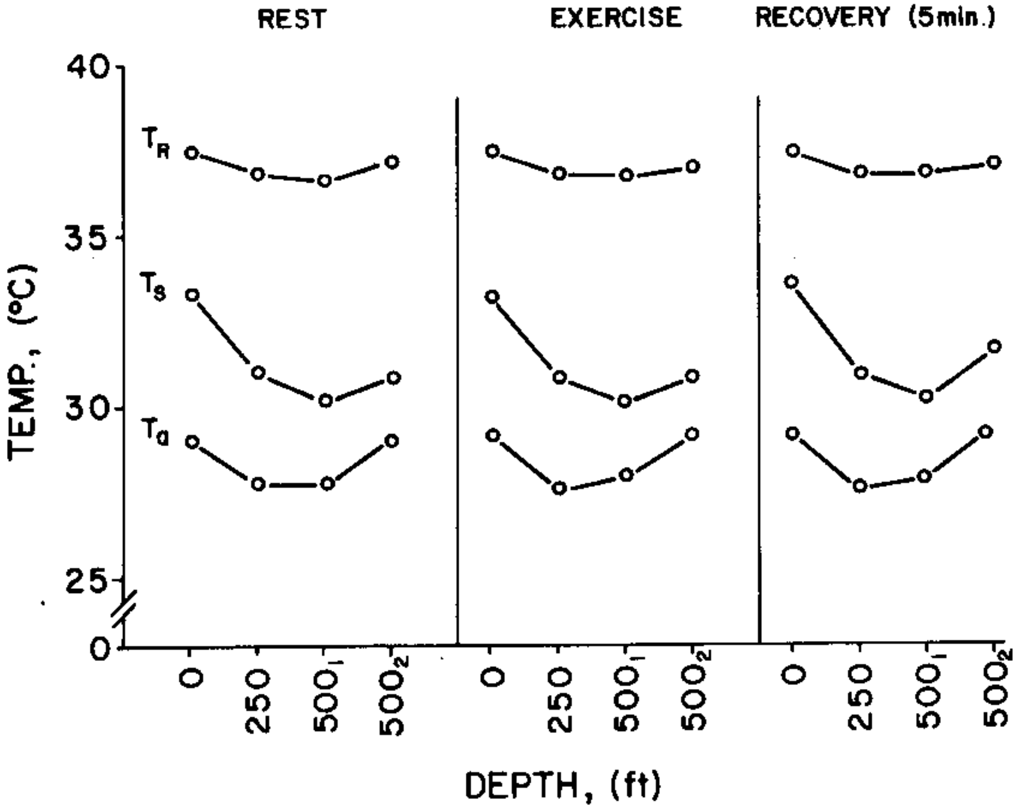

Figure 5. Rectal, skin, and ambient temperature during rest, exercise, and recovery as a function of depth.

From the physiologic/medical point of view, cold stress induces responses which are in themselves often debilitating, i.e., constriction of peripheral blood vessels, body fluid shifts, dehydration, etc. It also seems unlikely that nutritional management is a very practical method of maintaining heat balance. In the dive reported here, daily caloric intakes of well over 4000 Kcal failed to ameliorate the thermal effects of the ambient environment.

The obvious solution is to reduce the physiologic impact of the environment through appropriate means of bioengineering life support technology. This accomplished, most of the incapacitating medical problems would cease to occur.

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THERMAL COST OF SATURATION DIVING: RESPIRATORY AND WHOLE BODY HEAT LOSS AT 16.1 ATA

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INTRODUCTION

Since the early 1960's, the demonstrated ability of man to dwell in artificial helium-oxygen mixtures under saturation conditions has brought about many subsequent industrial, military, and scientific exposures to this environment. The reasons have appeared to be as many and as varied as the exposures. "Feasibility" and "screening" dives have been abundant and pragmatically impressive.

Now, in the 1970's, certain environmental features and their biomedical concomitants have become more visible, if not more serious, in man's dwelling under hyperbaric conditions for extended periods of time. Among these are the need to compromise in inert gas components in order to avoid, variously, inert gas narcosis, high gas densities (and the attendant ventilatory limitations), and of course, decompression sickness, -- all the while providing adequate but safe levels of oxygen for the divers.

A series of ingenious experiments are currently balancing these factors in several ways and continually extending the depth capability of "resident" man. (Biihlmann, A.A. et al., 1970; Morrison and Florio, 1971; Overfield et al., 1969; Strauss et al., 1973).

There are, howevet, some "on-going" physiologic problems which will continue to accompany men to depth unless overcome by engineering or medical technology.

One of these, recently summarized by Webb (1970) is the cumulative body heat drain which occurs particularly in the heliox environment at depth, and is due primarily to the high thermal conductivity, specific heat and density of that medium. The result is a high convective loss, with respiratory losses increasing with depth, which could lead to nutritional management problems in order for divers to maintain metabolic balance.

A second, fluid and electrolyte balance, enjoys a lesser abundance of documentation, but can be likened to the situations which obtain in Arctic survival, where the imbalances may tip the scale away from survival (Rogers and Setliff, 1964; Rogers et al., 1964).

In order to systematically quantify the physiologic impact on these three - thermal, fluid, and metabolic balances, a "dry" dive was undertaken in January 1972, in which six divers occupied the undersea habitat "Aegir" (Makai Range, Inc.) for a total of 9 days in a helium-oxygen environment. Maximum depth exposure was 500 feet (16.1 ATA)

The anthropometric and physiologic characteristics of the divers appear in Table 1, the dive "profile" and pertinent characteristics of the environment in Figure 1.

Divers	Age (yrs)	Ht (cm)	Wt (kg)	SA (m^2)	Vit. Cap. (l)	Pulse Rate (per min)	Blood Pr. Syst.	(mm Hg) Diast.
B.R.	29	167.0	64.9	1.74	3.90	56	110	60
J.M.	25	182.9	69.4	1.92	4.65	48	108	70
E.H.	32	165.1	75.3	1.82	3.65	80	128	90
W.B.	41	174.0	69.7	1.84	4.20	78	134	86
Cy. C.	33	185.7	81.2	2.05	4.91	64	146	94
C. Ch.	22	170.5	84.6	1.90	4.90	56	108	70
Mean	30.3	174.2	74.2	1.89	4.368	63.7	122.3	78.3
SE	2.7	3.5	3.1	0.046	0.217	5.3	6.6	5.5

Table 1. Physical characteristics of divers.

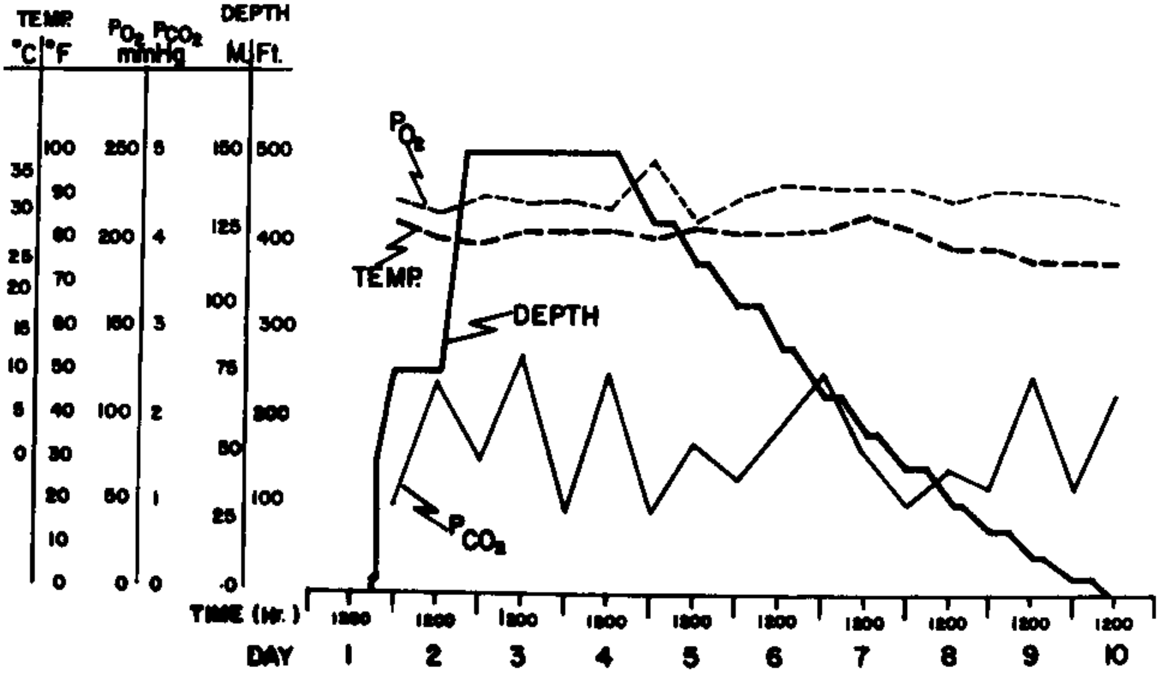

Figure 1. Dive profile and values of environmental parameters.

While each diver had particular housekeeping and scientific duties, a typical daily regimen appears in Table 2. The exercise portions were terminated during the decompression schedule.

Hours	Activity
0600	Reveille
0630	Recording of vital signs
0700	Breakfast
0730-1130	Heat loss monitoring (rest and exercise)
1200	Lunch
1300-1600	Physiologic monitoring (cold pressor test, water diuresis test, etc.)
1600-1800	Psychomotor testing*
1800	Supper
1900-2200	Psychological and Psychomotor testing
2200-0600	Sleep

Table 2. Daily regimen.

*Psychological and Psychomotor data are not included in this report.

"Aegir" consists of three separate compartments, each with autonomous life support and control capability -- laboratory/galley, central sphere (diving ready-room), and living quarters. All are equipped with P_{O_2}, P_{CO2} sensors (Minos-Beckman Instruments, Inc.) which continuously monitor ambient air. Relative humidity was monitored by Hygrometers. The environmental parameters in Figure 1 are those reported by the noon and midnight watch each day. A detailed description of "Aegir" has been reported previously by Pegg (1971).

METHODS

Dive profile: Beginning at 1900 hours on Day 1, pressurization was begun, The first overnight stop was made at 250 feet (approximately 8.6 ATA). After psychomotor testing during the remaining evening hours, the divers slept for 6 hours. At 0600 the normal regimen began. Vital signs -- pulse rate, blood pressure, oral temperature and body weight were recorded, followed by breakfast.

After breakfast, the subjects were taken in turn (standardized) for measurement of heat loss. Each subject, with rectal temperature probe in place and clad only in light shorts, sat quietly on a bicycle ergometer (Monark, Varberg, Sweden) for 15 or 20 minutes. He then breathed normally through a standard commercial mouthpiece double hose assembly (U.S. Divers, Santa Ana, California) whose intake side had been shortened to approximately 2 inches upstream of the intake valve. Inserted into this segment was a hygrometer sensing probe, thermistor and thermocouple probe. The exhaust side (just downstream from the exhaust valve) was similarly equipped, with the exhaust hose terminating at a connection with a Douglas bag valve for expired gas collection. Intake and exhaust relative humidities, rectal temperatures, and intake air temperatures were recorded in the habitat at appropriate intervals during the experimental regimen. The EMf difference between intake and exhaust thermocouples was hard-wired to a polygraph located in the main control room outside the habitat, as was EKG for heart rate count.

During the last 2-3 minutes of the rest period, expired gas was collected and EMf, rectal and ambient temperatures, and relative humidities were recorded. The Douglas bag was then emptied through a horizontal plexiglass mixing chamber (approximately 12 inches long, 4 inches diameter) which was fitted and sealed at the bottom over the sensing probe of a P_{O_2} meter. In series downstream from the mixing chamber was an attached gas flowmeter (A.H. Thomas Co., Philadelphia). Approximately half-way through the bag emptying procedure, the input and output sides of the mixing chamber were clamped off and the P_{O_2} of the contained gas recorded (approximately 30-40 seconds equilibration time was required). The bag volume was then completely emptied and the expired volume recorded from the flowmeter. The P_{O_2} meter was checked daily against a range of calibration gases carried on board. (These had been analyzed previously by the micro-Scholander technique in the laboratory.) $\dot{V}_E$ and $\dot{V}_{O_2}$ were determined from these procedures.

Simultaneously with the expired gas collections, temperature and heat flux were recorded at several points on the body surface of the subject: Forehead, chest, abdomen, upper back, upper arm, forearm, hand, thigh, calf, and foot. These were monitored by use of an exploring probe consisting of a thermistor and heat flow disc mounted side-by-side in a lucite plate. The plate was attached to a handle and spring loaded (with a marker) so that uniform tension was applied in each case. Each subject was indelibly marked at each of the various skin points for consistency throughout the dive duration. Skin temperatures were recorded via telethermometer (Yellow Springs Instruments, Yellow Springs, Ohio) in the habitat while heat flux was monitored on the external polygraph.

The subject then proceeded with mild exercise (410 KPM/min) for 10 minutes, at the end of which (last 2 minutes) the recording procedures above were repeated, and during recovery from minutes 2-3, 5-6, and 9-10.

One ATA control values were gathered in the unpressurized habitat in the control days just prior to the dive.

At 1300 hours on day 2, after 17 hours at 250 feet depth, pressurization continued to a depth of 500 feet, where the divers remained for 43 hours. The experimental protocol (Table 2) was followed as described above while at this depth, and fasting venous blood samples were taken on the morning of day 4 for comparison with pre-dive control samples.

Decompression began at 1400 hours on day 4 (Standard U.S. Navy helium schedule), with overnight holds from 0000-0600 hours and a rest stop from 1400-1600 hours daily, until surfacing at 1200 hours on day 10. The experimental procedures (minus the exercise portion) were adhered to until the habitat reached 150 feet (late on day 7) when the habitat went on "power-down" condition to pass through the fire hazard zone. From that point only the morning vital signs were monitored.

Food and fluid intake were closely monitored throughout. The regular meals were of the frozen variety, (SKY CHEF, Inc., Honolulu, Hawaii), and all divers received the identical meal at each time (though the menu varied). The divers began the controlled diet 2 days prior to the dive. Each meal, on an individual basis, was pre-weighed item by item, tagged as to recipient, and all residue weighed to determine individual consumption. The divers themselves scrupulously logged fluid consumption in its several forms. Sample meals were subsequently analyzed for sodium and potassium content by flame photometry, again on a per item basis, and caloric and water content estimated from nutritional tables (Heinz Handbook of Nutrition, 1959). Extra food and snacks were allowed for reasons of morale, but again carefully logged and subsequently analyzed.

Feces was not retained, but all urine was collected as voided, the volume noted and triplicate 50 cc aliquots locked out to the surface for subsequent analysis for sodium, potassium, chloride, urea, osmolarity, and creatinine.

Blood drawn on day 4 (Diver WB was an ex-medical corpsman, U.S.N.) were centrifuged in the habitat (after hematocrit determination) and plasma locked to the surface for analysis of osmolality, sodium, potassium, chloride, creatinine, total protein, urea, and glucose.

Water diuresis tests (one liter load) were run on 4 divers on day 4 at 500 feet, day 6 at 213 feet and at the surface. Cold pressor tests (6°C water) were done by 3 divers at 500 feet on day 4, at 180 feet on day 7, and at the surface.

RESULTS

Vital signs

The daily vital sign data are shown in Figure 2. Generally speaking, all parameters showed slight decreases early during the compression stage. The systolic blood pressure fell slightly more than diastolic and both remained somewhat depressed throughout the dive. A significant bradycardia was observed initially with a slow return of heart rate to normal by the end of the dive. A two day episode of higher rates (compared to control-o-day) occurred on days 6 and 7. Oral temperatures were depressed between 1 and 2°C throughout; rectal temperature was down an average of 1°C. A body weight loss of approximately 1 KG maximum (day 3) was observed, and remained continually lower than control weights. These data will be discussed in detail below, but are generally consistent with peripheral vasoconstriction but only as coupled with reduction of extracellular fluid volume.

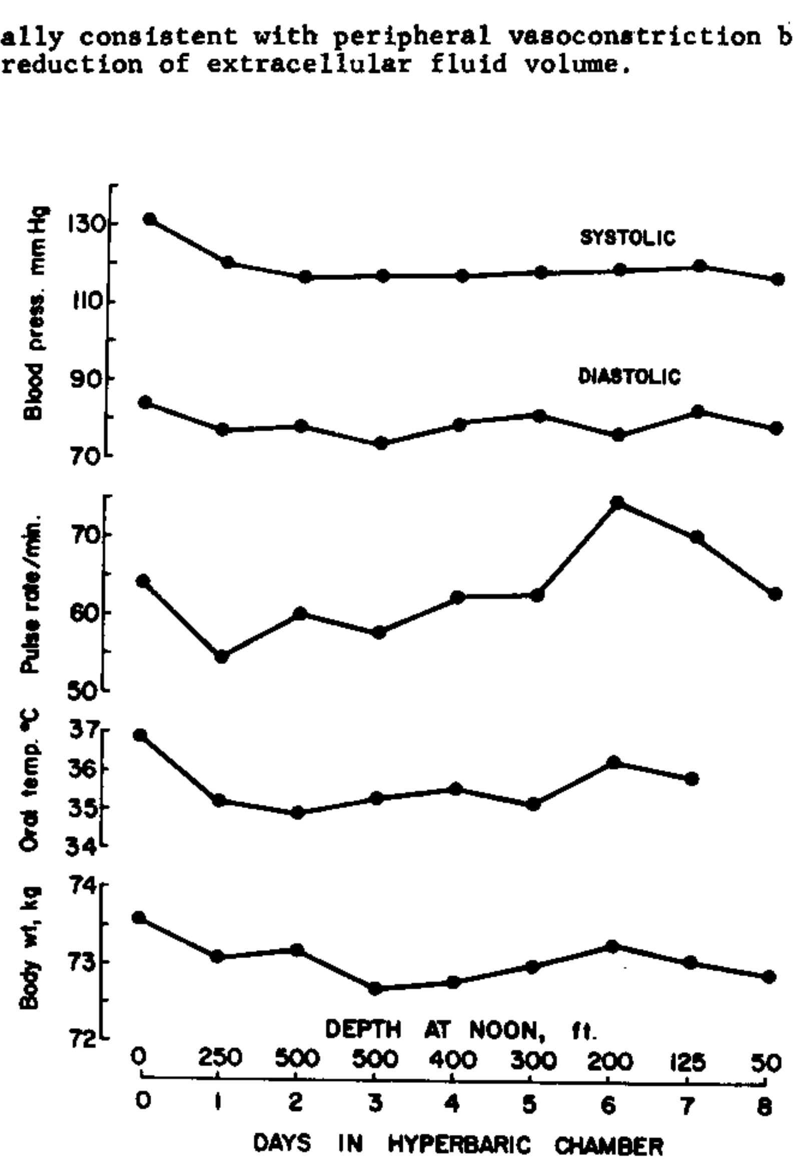

Figure 2. Daily vital sign data.

Cardio respiratory responses

The cardiopulmonary responses recorded at depth are presented in Figure 3, showing that even during exercise, heart rates are maintained at lower levels than surface controls. V_{O_2} during both rest and exercise peaks on the first day at 500 feet; returning toward surface values during the second day's exposure at that depth. $\dot{V}_E$ rises at rest, falls during exercise, again inflecting on the second day at 500 feet. This inflection also characterizes the O_2 pulse and ventilation equivalent curves. We believe the common denominator in all these reversals to be the change in ambient temperature which occurred on day 3 (500 feet). As seen in Figure 1, the internal habitat temperature had dropped more than a degree C from day 1 through day 2. On day 3, the second day at 500 feet, the habitat heaters were turned on, bringing the temperature back to approximately 29°C, within the comfortable thermal zone cited by Webb (1970). This was subjectively noted by the divers. As will be shown in a table below, mean body temperature was indeed slightly higher on day 3 compared to day 2.

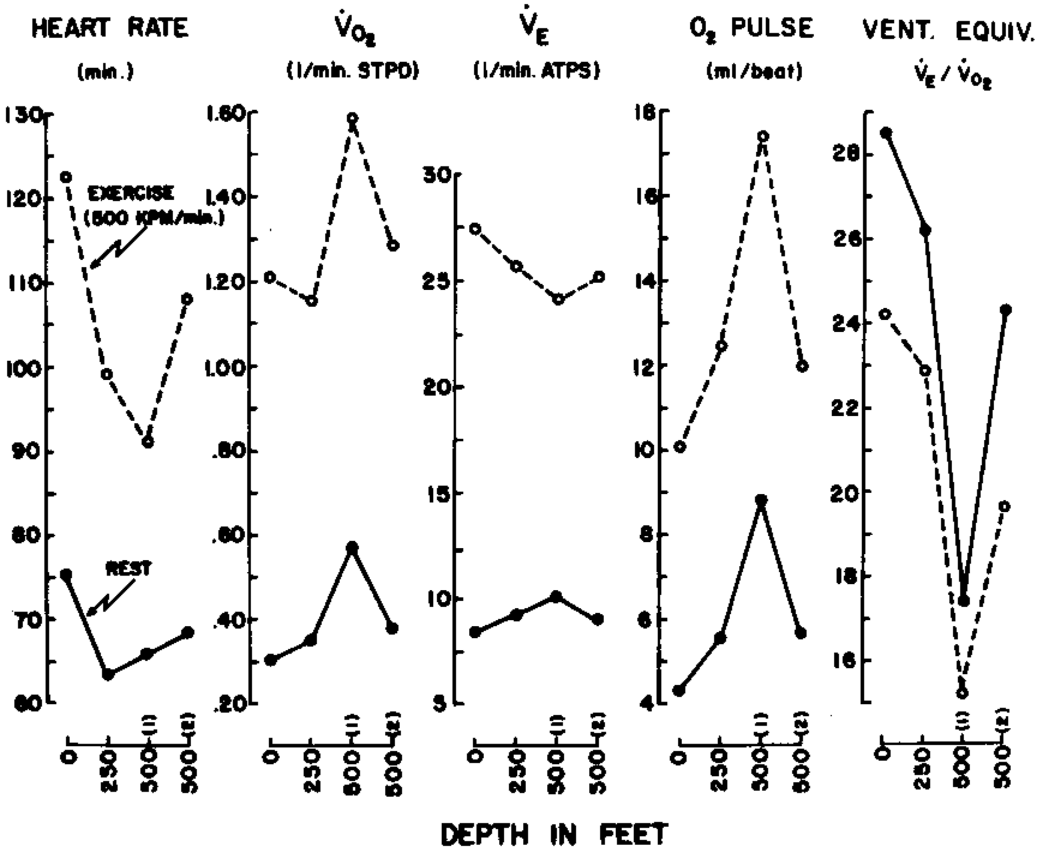

Figure 3. Cardiopulmonary responses during rest and exercise as a function of depth.

Heat loss responses

Table 3 describes the relevant thermal characteristics of the environment, indicating, as shown by the relative specific heat (C_p) and heat capacity (ρC_p), the potential thermal drain facing the divers in this environment..

Respiratory heat loss becomes an increasing problem as gas density and relative specific heat of the breathing medium increase. Total respiratory heat loss is the sum of the convective loss and the evaporative loss. The following expressions were used for calculation in this experiment.

(1) Convective loss: = $\dot{V}_E \cdot \rho \cdot C_p \cdot (T_E - T_I)$
where $\dot{V}_E$ = minute volume in L/min ATPS
ρ = gas density in gm/L
C_p = specific heat of the gas in Kcal/gm·°C

T_E = temperature of expired gas
and T_I = temperature of inspired gas

(2) Evaporative loss = $\dot{V}_E \cdot \dfrac{\Delta P_{H_2O}}{760} \cdot \dfrac{18}{22.4} \cdot \dfrac{273}{272, + T_{amb}} \cdot 580$

where $\dot{V}_E$ = minute volume
ΔP_{H_2O} = vapor pressure of water in mmHg
and = (P_{H_2O}) expired - (P_{H_2O}) inspired
760 = 1 atmosphere of pressure in mmHg
18 = molecular weight of water in gm/mol
22.4 = molar volume of water in L/mol
273 = absolute temperature
T_{amb} = ambient temperature in °C
580 = heat of vaporization in cal/gm

Physical Characteristics	Air at Surface	250 feet (He 96.6%) (O_2 3.4%)	500 feet (He 98.2%) (O_2 1.8%)
Density (ρ), gm/liter	1.11	1.63	2.82
(Relative ρ)	(1.0)	(1.5)	(2.5)
Specific heat (C_P) cal/gm/°C	0.26	1.11	1.17
(Relative C_p)	(1.0)	(4.3)	(4.5)
Heat capacity (ρC_p) Cal/liter/°C	0.29	1.82	3.32
(Relative ρC_p)	(1.0)	(6.2)	(11.3)

Table 3. Thermal characteristics of air at surface and He-O_2 at hyperbaric environment.

Figure 4 represents the respiratory heat loss components during the experimental conditions of rest, exercise, and these recovery points. There was no difference between the first and second days at 500 feet, so the data represent the second day at that depth. The major feature of these data is the fact that with increasing depth and consequent increasing ρC_p of the ambient heliox, the convective term of respiratory loss increases dramatically while only small increases are seen in the evaporative term. This is in marked contrast to the case of air at 1 ATA, where losses are almost completely evaporative. The respiratory "thermal" recovery was essentially complete after 5 minutes from termination of exercise at this level.

Skin heat fluxes are depicted in Figure 5 for both rest and exercise at the various simulated depths. For the resting condition, the salient points appear to be an alteration in distribution and degree of flux upon exposure to the

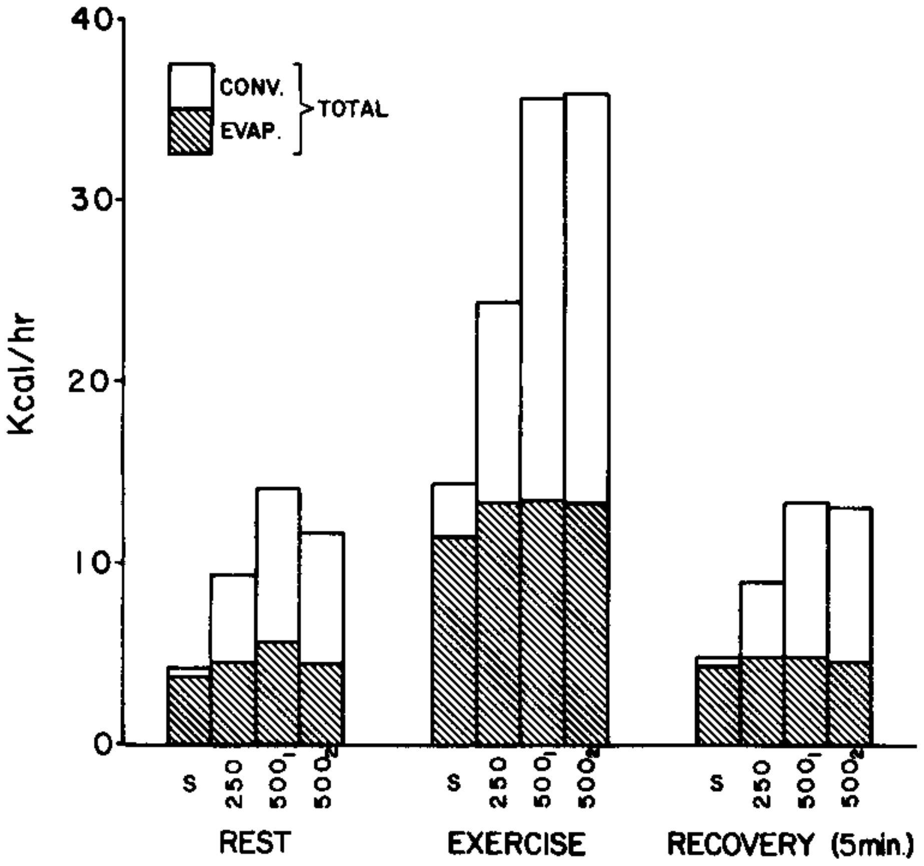

Figure 4. Convective and evaporative respiratory heat loss during rest, exercise, and recovery as a function of depth.

heliox environment at depth. For instance, while forehead flux appears high under all conditions, a notable decrease, especially in the distal extremities, (especially foot, and to a lesser extent, upper arm, and hand-below 250 feet) was found to occur. On the other hand, the back, lower arm, thigh and lower leg were found to have relatively higher heat flux in the heliox environment at all depths.

During exercise, the qualitative pattern was not dissimilar to the resting condition, high fluxes from back, lower arm, thigh and lower leg, with hand and foot (especially the latter) showing low heat flows.

Considering then the overall pattern of heat exchange under these conditions, Table 4 provides a summary during rest and exercise. At rest, mean skin temperature falls steadily through the first day at 500 feet; rises slightly on the second, presumably via the addition of heat to the chamber environment. Mean body temperature (2) follows the same pattern. Heat production, as gauged by metabolic rate, rises to 90.8 Kcal/hr/m^2, then drops at day 2 at depth, while heat loss, both cutaneous and respiratory, follows the same qualitative pattern. The net result is a negative heat balance for all days except 500' (1) where metabolic heat production peaks. During exercise, positive heat balance was maintained throughout, even in the face of higher heat loss, through a higher level of metabolic heat production. Again, the inflection from 500' (1) to 500' (2) was seen.

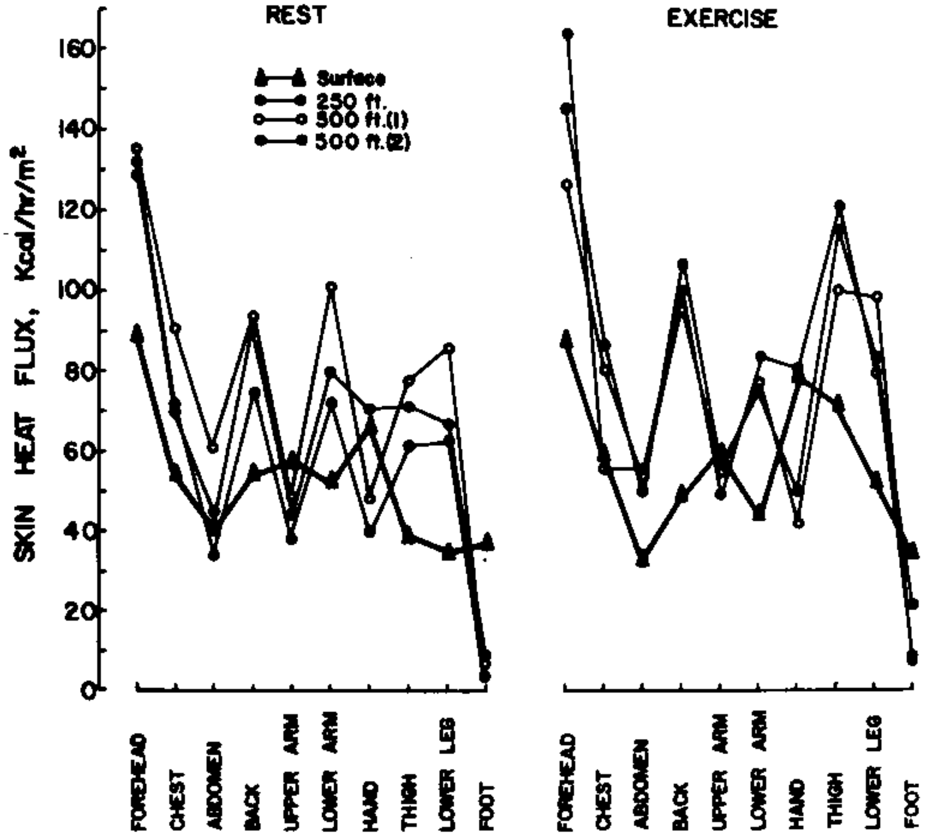

Figure 5. Cutaneous heat flux during rest and exercise as a function of depth.

It can be seen in Figure 6 that the relative contribution of respiratory heat loss to total loss increased predictably with depth and was exacerbated with exercise to the point of comprising approximately 20% of total loss even under the mild work load imposed. Skin flux decreased overall as skin temperature decreased. Figure 7 shows the narrowing of the skin to air gradient under conditions of rest, exercise and at the 5 minute recovery point, when associated with increasing depth. Again, on the second day at 500 feet the gradient narrows as Ta was raised.

The intensity of the peripheral vasoconstrictive reflex is well illustrated by the results in Figure 8, depicting the administration of a standard cold-pressor test with the hand immersed in 6°C water for 30 minutes. The blood pressure records are generally unremarkable and typical, but a noticeable lack of "hunting" or vasodilatation and consequent increase of finger temperature was found. The peripheral constriction was maintained even in the face of this added challenge, most apparent at the greatest depths.

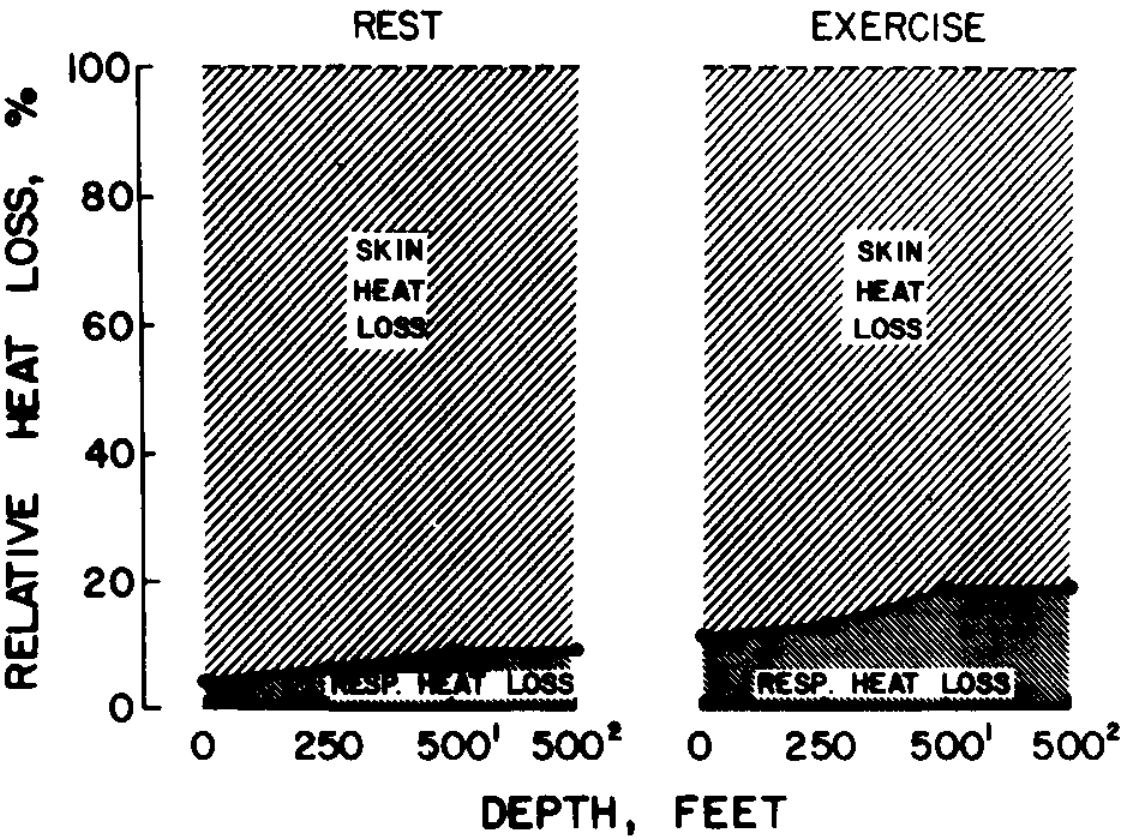

Figure 6. Respiratory heat loss as proportion of total loss during rest and exercise as a function of depth.

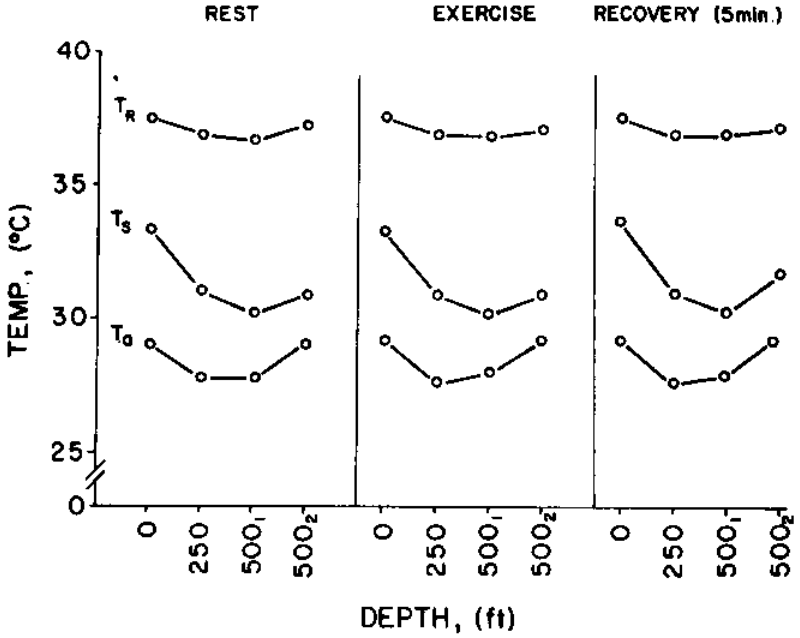

Figure 7. Rectal, skin, and ambient temperature during rest, exercise, and recovery as a function of depth.

	Depth (ft)			
Heat exchange	0	250	500(1)	500(2)
Chamber temp., °C	29.0	27.8	27.8	29.0
Chamber humidity, %	78	68	72	76
A. Rest				
Rectal temp., °C	37.43	36.91	36.66	37.04
Mean skin temp., °C	33.54	31.01	30.13	30.88
Mean body temp., °C	36.14	34.96	34.50	35.00
Heat prod., Kcal/hr/m^2	44.7	55.7	90.8	60.6
Heat loss, Kcal/hr/m^2	52.3	74.1	87.0	67.5
Heat balance, Kcal/hr/m^2	-7.6	-18.4	+3.8	-6.9
B. Exercise (410 KPM/min)				
Rectal temp., °C	37.38	36.83	36.80	36.96
Mean skin temp., °C	33.26	30.78	30.06	30.86
Mean body temp., °C	36.02	34.83	34.57	34.94
Heat prod., Kcal/hr/m^2	191.5	183.5	251.0	203.5
Heat loss, Kcal/hr/m^2	64.5	99.2	103.0	107.5
Heat balance, Kcal/hr/m^2	+127.0	+84.3	+148.0	+96.0

Table 4. Heat exchange at rest and during exercise.

Fluid and electrolyte balance

As found in other dives (Webb, 1970), the nutritional intake was high in caloric content, averaging over 4000 Kcal/day and ranging as high as 6400 on day 4 of the dive. External water intake was also high, approximately 2.5 L per day, however, negative fluid balance was a characteristic of the entire dive period. Hematocrit changes are given in Table 5; in which mean values are misleading, in that two of the 6 divers were found to have greater than 10% increase. For therapeutic reasons, these divers were instructed to increase both water and salt intake, but unfortunately, no further blood studies were made. On the other hand, the two suffered no untoward effects upon decompression, so presumably, the attempts at proper hydration were effective.

Figure 9 illustrates the renal excretion patterns found during the dive (pooled 24 hour samples). Urine flow was generally elevated throughout the dive, while creatinine excretion decreased and plasma creatinine levels did not change (Table 5). Sodium and potassium excretion remained approximately unchanged as was also true of plasma levels. Thus, in the face of decreased GFR, the relatively constant electrolyte excretion, and increased urinary volume, there was the implication of an impaired electrolyte reabsorption and consequent

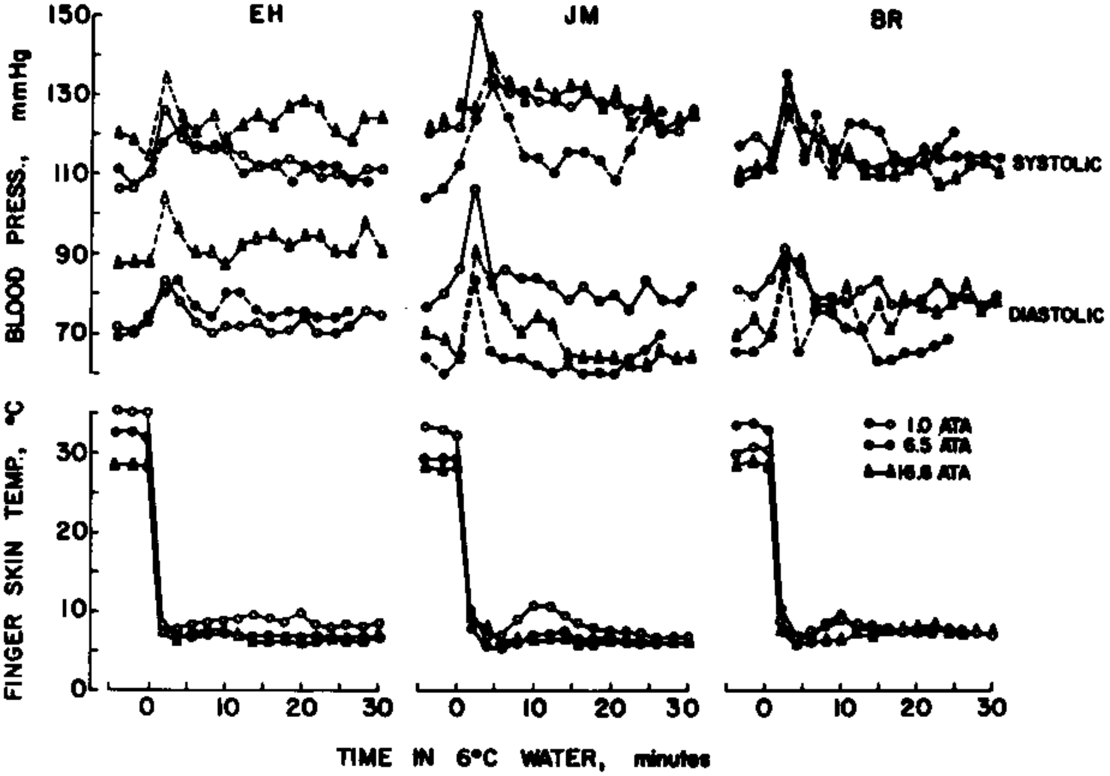

Figure 8. Blood pressure and finger temperature responses to immersion of the hand in 6^{O}C water as a function of depth.

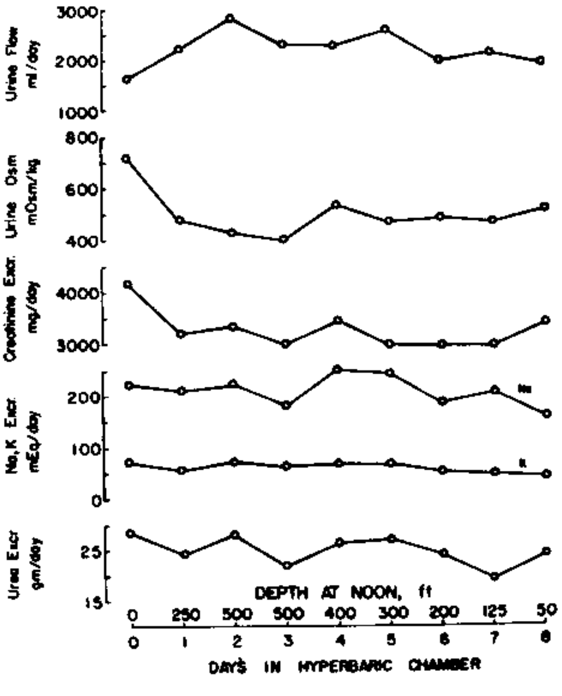

Figure 9. Urinary excretion data as a function of time and depth of pressurization.

Composition	Before dive	500 feet
Hematocrit ratio, %	40.5 ± 1.3	46 ± 1.5*
Plasma conc.		
Osmolality, mOsm/kg	285 ± 1.3	273 ± 0.7*
Na, mEq/l	142 ± 0.7	141 ± 1.3
K, mEq/l	4.0 ± 0.1	4.2 ± 0.1
Cl, mEq/l	110 ± 0.2	106 ± 1.5
Creatinine, mg%	1.03 ± 0.05	1.02 ± 0.05
Proteins, gm%	7.10 ± 0.17	7.83 ± 0.49**
Urea, mm/l	8.2 ± 1.2	7.7 ± 0.7
Glucose, mg%	86.3 ± 7.3	97.0 ± 8.8

Table 5. Blood chemistry before dive and at 500 feet depth in 4 divers.

* Significantly different from the corresponding value obtained before the dive ($P < .05$).

**Marginally different from the corresponding value obtained before the dive ($0.10 > P > .05$).

solute diuresis. The decreased urinary osmolality is supportive of this. This same pattern of contraction of extracellular fluid volume and diuresis has been observed in cold-exposed men during field experiments in the winter sub-arctic and ascribed to a decreased aldosterone secretion (Moore et al., 1968).

It was of interest in the context of fluid shifts and decreased plasma osmolality to determine whether the anti-diuretic hormone (ADH) system was in any way affected. Under the conditions of this experiment, this was most simply checked by following the elimination of a one-liter water load. These data appear in Figure 10 as taken at the surface, at 213 feet, and at 500 feet depths. While it appears as though peak urine flow occurs somewhat sooner at 500 feet, there was no statistical difference in flow pattern or urine osmolality related to simulated depth. Therefore, it appears on the basis of this test, that there is no change in time course or amplitude of the ADH system response.

In summary, these data suggest that, in the face of the thermal drain associated with a high pressure helium-oxygen environment, there are attendant deviations which occur in fluid and electrolyte balance which are similar to responses seen in other cold stress situations. This thermal drain, with the increasing importance of respiratory heat loss at depth, would be exacerbated during water excursions and the concomitant cutaneous heat losses. In any event, the relative dehydration which accompanies cold-induced vasoconstriction could be a pre-disposing factor in the onset of decompression sickness. Thus, fluid imbalance constitutes another worthy area of research with a view toward potential saturation diver support.

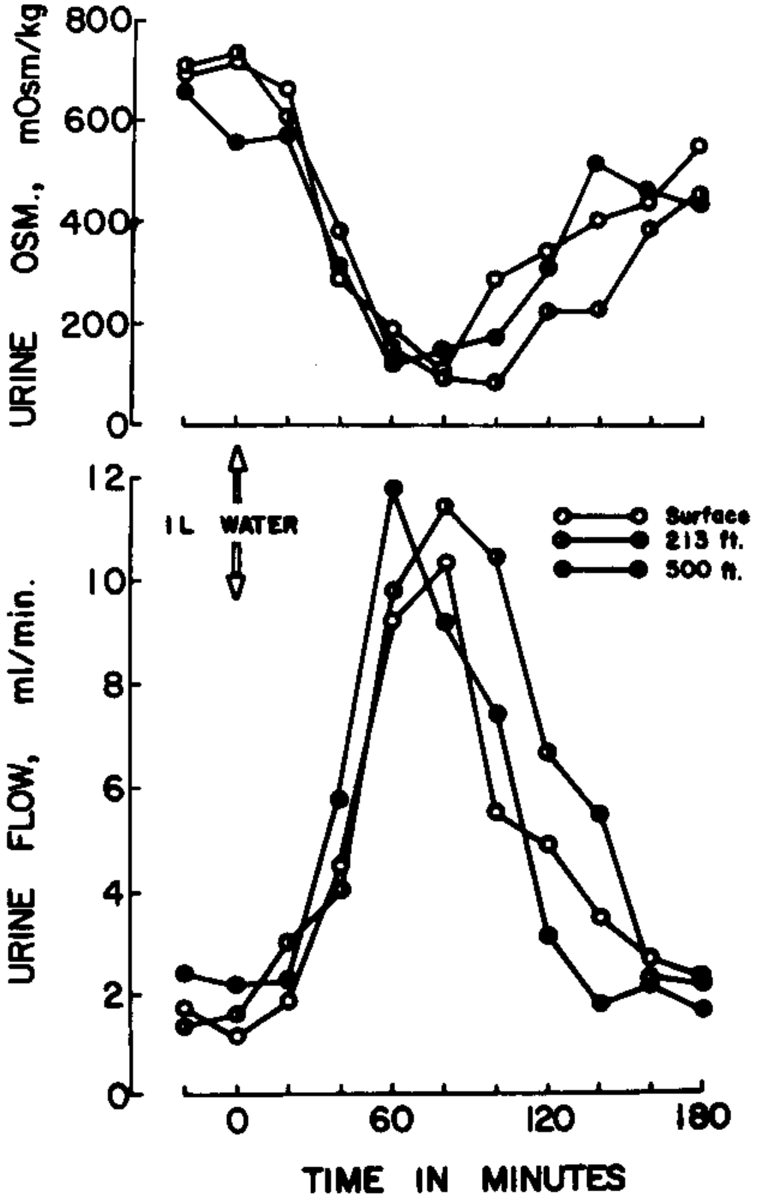

Figure 10. Time course of urine flow and osmolality in response to a 1 L water load as a function of depth.

ACKNOWLEDGMENTS

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MODIFICATION OF THE QUINTON-MONARK BICYCLE ERGOMETER FOR UNDERWATER USE

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ABSTRACT

With a few simple modifications, the standard Quinton-Monark bicycle ergometer can be made suitable for underwater use. The best fit regression equation of oxygen consumption ($\dot{V}_{O_2}$) as a function of pedaling frequency (f) has a correlation coefficient (r) of .996. The general form of the regression is a third order polynominal: $\dot{V}_{O_2} = .274 + .002025f - .000059f^2 + .000008f^3$. The extremely high accuracy of this predictive equation makes this simple ergometer of practical importance to the investigator interested in physiological responses to underwater exercise.

A theoretical discussion on the physical meaning of each of the zero, first, second, and third order terms of pedaling frequency is presented.

Key words: Underwater ergometer; underwater exercise; underwater oxygen consumption.

INTRODUCTION

With a few simple modifications, the Quinton-Monark bicycle ergometer (Åstrand 1960) can be converted to a very simple, yet very accurate, underwater ergometer. Its high predictive accuracy and subject non-dependency makes this ergometer of practical importance to investigators interested in physiological responses of the working diver.

METHODS

The modifications are as follows: Two standard automotive grease nipples were installed to provide easy regreasing of both the bottom bracket crank bearing and the flywheel bearing. A magnetic reed switch (Calectro Electronics, Rockford, Illinois) was installed to provide continual monitoring of the pedaling frequency. This reed switch was fastened securely to the frame and a small magnet was taped to the inside of the left pedal crank arm. The reed switch was then connected in series with a standard 1.5 volt flash light battery. The circuit can be wired directly to the input terminals of any standard pen recorder preamplifier. As the crank arm passes adjacent to the reed switch the circuit is completed and a small pen deflection is produced resulting in a direct count of pedaling frequency for any given time period. The third modification was to remove the friction belt completely and immerse the bicycle ergometer in a water tank, 152 cm long, 64 cm wide and 146 cm deep (see Figure 1).

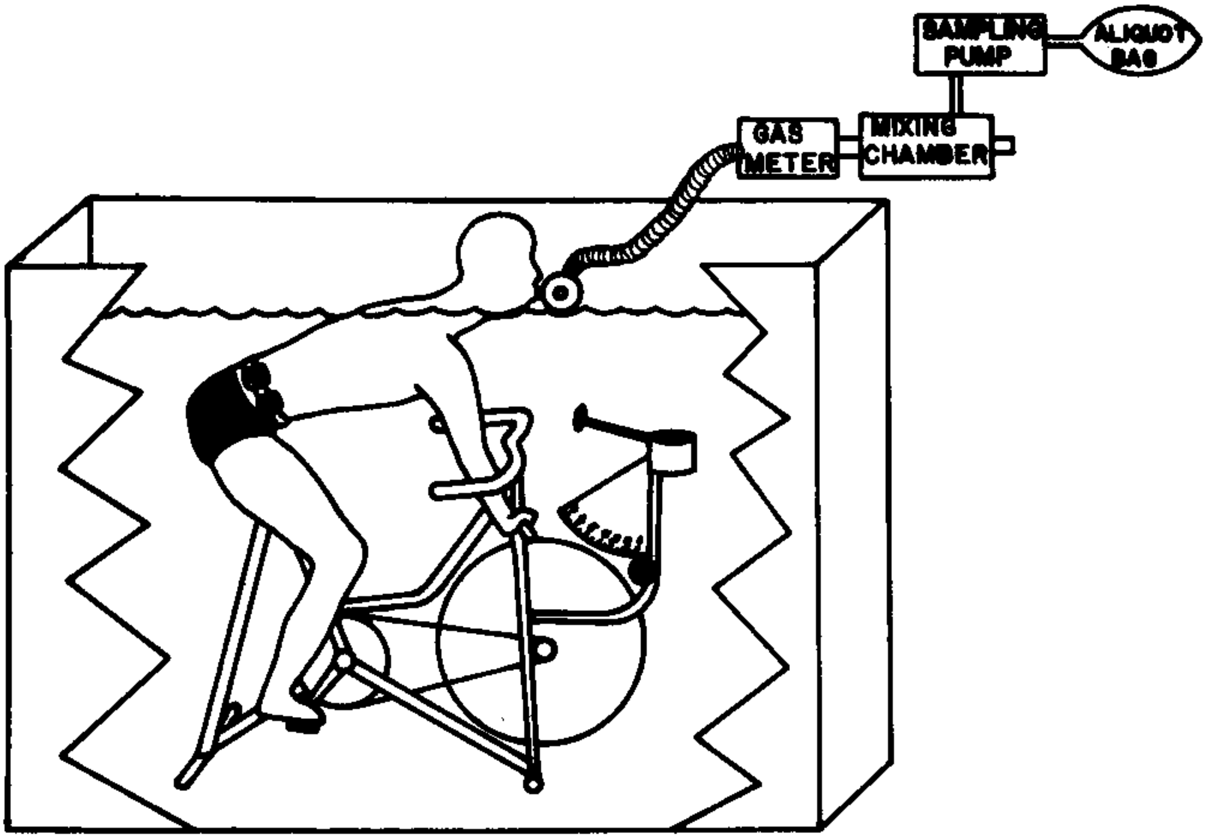

Figure 1. Diagrammatic of experimental lay-out.

Subjects	Age (years)	Height (cm)	Weight (kg)	% Fat	VO_2 max liters/min
DB	24	180	75.5	15	3.35
RD	29	178	70.8	9	4.08
JM	26	184	69.2	10	3.75
BR	29	170	65.4	16	2.05
RS	29	172	66.2	12	2.32
LW	24	164	58.1	13	2.66

Table 1. Subject characteristics

Six male subjects, characterized in Table 1, were immersed to the neck while sitting on the ergometer. The seat height was adjusted so that the legs were fully extended at the bottom-most pedal position. The subject gripped the resting foot pegs rather than the handle bars. This position was chosen in order to maintain the subject's upper body nearly horizontal to better simulate a swimming scuba diver. The position is very similar to that used by racing cyclists.

The subjects breathed either room air via a Collins Triple-J low resistance mouthpiece or compressed air via a two stage, double-hose regulator (U.S. Divers, Santa Ana, California). In the latter system, the compressed air bottle and the attached regulator were mounted to the inside wall of the tank.

Each subject was asked to sit quietly on the ergometer for 10 minutes before taking a two-minute expired gas collection. The subject then proceeded to pedal consecutively at 20, 40, 50, 60, 70 and maximum rpm for five minutes. The desired pedaling frequency was obtained by having the subject keep time with a metronome. A continual check on the pedaling rate was given by the magnetic reed switch discussed above. Two one-minute expired gas collections were obtained at each frequency from minutes 3 to 4 and 4 to 5 in order to verify that a steady state oxygen consumption had been reached. The protocol above was followed for each of the six subjects breathing either room air or compressed air while immersed in 30°C.

Oxygen consumption was measured by one of two standard open circuit methods by collection of expired gas. The first method, which was later replaced, consisted of passing the expired gas through a high speed Parkinson-Cowan flowmeter in order to measure V_E and then into a four liter baffled mixing chamber. An aliquot sample of 1-liter was continuously withdrawn from the mixing chamber over the entire collection period via a Beckman microcatheter sample pump. This was then analyzed for O_2 and CO_2 content by either a Beckman E-2 or OM-11 oxygen analyzer and a Beckman LB-1 CO_2 analyzer. The second method consisted of collecting the entire expired gas volume as described by Daniels (1971) in meteorological balloons and analyzed for O_2 and CO_2 content as described above. Expired gas volume was measured in a 350 liter Tissot Spirometer. $\dot{V}_{O_2}$ (STPD) was then calculated according to the principles outlined by Consolazio, Johnson, and Pecora (1963).

RESULTS AND DISCUSSION

By Student's paired t-test, no significant difference in $\dot{V}_{O_2}$ was found between breathing room or compressed air in 30°C water. Therefore, all of the data were pooled and the relationship between $\dot{V}_{O_2}$ and pedaling frequency is illustrated in Figure 2.

For reasons discussed later a polynomial regression analysis of the following form was applied:

$$\dot{V}_{O_2} = C_o f^o + C_1 f^1 + C_2 f^2 \ldots\ldots C_n f^n \qquad \text{(Equation 1)}$$

Results of this analysis from the IBM 360 computer gave the best fit (r) = .996 when n = 3. The coefficients (± SE) are as follows:

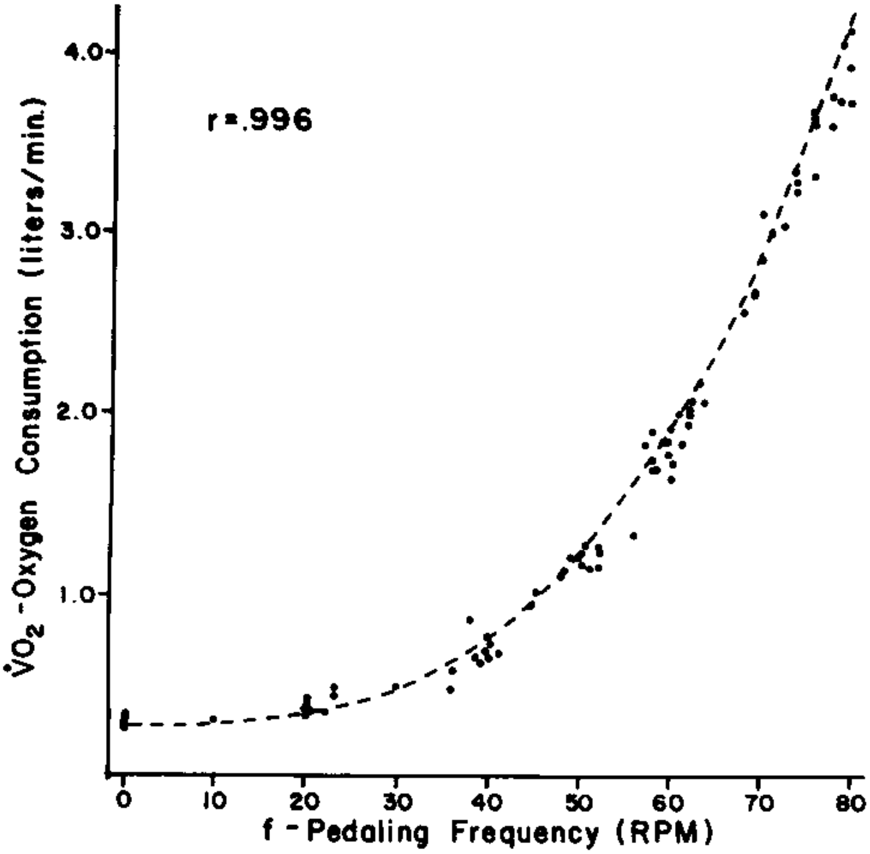

Figure 2. Energy costs as a function of increasing work load.
r = correlation coefficient.

$C_0 = + .274 \pm .80236 \times 10^{-3}$
$C_1 = + .002025 \pm .12980 \times 10^{-4}$
$C_2 = - .000059 \pm .12586 \times 10^{-7}$
$C_3 = + .000008 \pm .85568 \times 10^{-12}$

Table 2 illustrates both the absolute value and the percentage of total (in parenthesis) of each of these terms as pedaling frequency varies from 10 to 80 rpm. One sees that the f^o term remains constant but becomes relatively less important as frequency increases. Both the f^1 and f^2 terms are of relatively little importance when expressed as a percentage of total $\dot{V}_{O_2}$, and because they are opposite in sign and of approximately equal magnitude, they tend to cancel each other out. Finally, Table 2 illustrates that the f^3 term becomes progressively more important as frequency increases until, at 80 rpm, it represents almost the entire energy expenditure.

An f^o term was expected since, even if the subject performs no external work, he would still have a resting $\dot{V}_{O_2}$. The fact that the C_o coefficient turned out to be .274 liters O_2/min, (well within the range of expected resting O_2 consumption) helps to substantiate this point.

The f^1 term represents the power needed to be applied to overcome friction due to solid-to-solid contact of the moving parts of the ergometer. In fact, it is on this linear relationship that the original Quinton-Monark bicycle ergometer was based (Von Dobeln 1955), the frictional work being predominantly due to the friction between the flywheel and the adjustable tension belt. This is further illustrated by the fact that when the belt is removed for underwater use, this term is of relatively small importance even at the higher frequencies.

	VO_2	=	.274	+	.002025 f	-	.000059 f^2	+	.000008 f^3
10	.29635		.274 (92.45)		.02025 (6.83)		.005900 (1.99)		.008000 (2.69)
20	.35490		.274 (77.20)		.04050 (11.41)		.023600 (6.64)		.064000 (18.03)
30	.49765		.274 (55.05)		.06075 (12.20)		.053100 (10.67)		.216000 (43.40)
40	.77260		.274 (35.46)		.08100 (10.48)		.094400 (12.21)		.512000 (66.26)
50	1.22775		.274 (22.31)		.10125 (8.24)		.147500 (12.01)		1.0000000 (81.44)
60	1.91110		.274 (14.33)		.12150 (6.35)		.212400 (11.11)		1.7280000 (90.41)
70	2.87065		.274 (9.54)		.14175 (4.93)		.289100 (10.07)		2.7440000 (95.58)
80	4.15440		.274 (6.59)		.16200 (3.89)		.37760 (9.08)		4.0960000 (98.59)

Table 2. Partitioned estimated oxygen consumption. (% contribution of total at each level of f.)

In order to provide a complete discussion of the physical meaning of the f^2 and f^3 terms, a brief review of basic fluid dynamics is in order. A more complete and excellent discussion of this subject is given by Shapiro (1961).

Reynolds number (R_e), which can be thought of as an index of the degree of laminar and turbulent flow, is calculated as follows:

$$R_e = \frac{\text{Density x Velocity x Size}}{\text{Viscosity}} \qquad \text{(Equation 2)}$$

Density (gm/cm^3) and viscosity (dynes x sec)/cm^2, or poise) are physical characteristics of the fluid moving relative to an object which the fluid flows through or around. Velocity (cm/sec) is the velocity of the fluid relative to the object, whereas size expressed in units of length (cm) represents a linear dimension perpendicular to the direction of fluid flow. For simple cases, like fluid flowing in a rigid tube or around a sphere, this size term is represented by the diameter of the tube or sphere. For more complex shapes a concept known as hydraulic radius is often introduced. Hydraulic radius is the quotient of the cross sectional area perpendicular to the flow path and the perimeter of this area. The complex mechanisms and shapes of the subject-ergometer system immersed in water make it virtually impossible to estimate either size or velocity in order to calculate R_e.

Nevertheless, it seems safe to assume that the size factor, density and viscosity remain constant at the various pedaling frequencies, and that the relative speed between the various moving components and the enveloping fluid increases as pedaling frequency increases and therefore, so does R_e.

In addition, R_e is the ratio of the inertial drag forces to the viscous drag forces of the system. Viscous drag is due to the deformation of the fluid by an object moving, in this case, through it. Inertial drag forces are due to acceleration of the fluid particles caused by continual impact between object and fluid particles. Thus, at very low R_e, the inertial drag forces can be ignored as compared to the viscous drag forces. At high R_e, the viscous drag forces become less important and the inertial forces dominate.

Drag forces for low R_e numbers are described by Stokes Law of drag which states:

$$\text{Viscous drag force } \alpha \text{ (Velocity) x (Viscosity) x (Size)}$$

$$\text{(Equation 3)}$$

The fact that power is proportional to the drag/force velocity product (Equation 4) indicates that the power needed to overcome viscous drag is proportional to velocity squared (Equation 5),

i.e. $\quad \text{Power } \alpha \text{ (Drag force) x (Velocity)} \qquad \text{(Equation 4)}$

Combining equations (3) and (4) results in:

$$\text{Power } \alpha \text{ (Viscosity) x (Size) x (Velocity)}^2 \qquad \text{(Equation 5)}$$

At high R_e the inertial drag force dominates. There is no exact law of drag for high R_e systems but empirical results for geometrically simple systems indicate that inertial drag forces are related in the following manner to velocity, density and size (Shapiro, 1961):

$$\text{Inertial drag } \alpha \text{ (Velocity)}^2 \times \text{(Density)} \times \text{(Size)}^2 \qquad \text{(Equation 6)}$$

Combining equations (6) and (4) results in the power to overcome inertial drag being proportional to velocity cubed:

$$\text{Power } \alpha \text{ (Density)} \times \text{(Size)}^2 \times \text{(Velocity)}^3 \qquad \text{(Equation 7)}$$

As pedaling frequency increases, the energy expenditure becomes extremely dependent on the f^3 term (as seen in Table 2) and indicates that the majority of the O_2 consumed is used to overcome inertial drag which dominates at high R_e. This hypothesis was further substantiated when the data were analyzed by a standard stepwise polynomial regression analysis. As a result, when one only entered the C_0 and C_3f^3 terms, a $(r)^2$ value of .9928 was obtained. This may be interpreted to mean that 99.28% of the O_2 consumption can be explained by the resting and f^3 term. This is not to say that the f^1 and f^2 terms are not significant, but that they are of relatively little importance.

Due to the 52:14 gear ratio between the crank sprocket and the flywheel sprocket, the flywheel is spinning almost four times as fast as the subject is pedaling. This makes the flywheel by far the fastest moving of all the moving parts in the water and the most likely cadidate for the cause of most of the inertial drag. This allows one to postulate that the inertial drag on the flywheel and possibly the chain and crank mechanisms is the cause of most of the energy expenditure. This hypothesis is also consistent with the small amount of subject variability found in the oxygen consumption measurement.

The simple modifications described enable one to convert a standard Quinton-Monark bicycle ergometer found in most human performance laboratories for underwater use. Even though the actual workload imposed on the subject is not known in conventional units of power, it can be indexed by pedaling frequency. In the practical sense, this allows the workload to be variable and under the control of the investigator over the entire physiological range of the subjects tested. Additionally, this ergometer enables the investigator to predict accurately the energy expenditure of subjects in terms of V_{O2} prior to the actual exercise test. This is of practical importance in allowing the investigator to hold $\dot{V}_{O2}$ constant for any given series of experiments or to hold percent $\dot{V}_{O2max}$ constant if one previously determines the given individual's $\dot{V}_{O_{2max}}$ for this particular type of exercise.

Finally, the high correlation coefficient between energy expenditures and work load, indexed by pedaling frequency, and the simplicity of the type of exercise imposed, indicates that the ergometer is extremely subject independent, with a very small learning factor.

ACKNOWLEDGMENTS

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SKINFOLDS AND RESTING HEAT LOSS IN AIR AND WATER: TEMPERATURE EQUIVALENCE

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ABSTRACT

Fourteen male subjects with average unweighted mean (9 sites) skinfolds (MSF) of 10.23 mm underwent several 3 hour exposures to cold water and cold air of similar velocities in order to compare by indirect calorimetry the rate of heat loss in water and air. Measurements of heat loss (excluding the head) at each air temperature (Ta = 25, 20, 10°C) and water temperature (Tw = 29 - 33°C) were used in a linear approximation of overall heat transfer from body core (T_{re}) to air or water. We found the lower critical air and water temperatures to fall as a negative linear function of MSF. The slope of these lines was not significantly different in air and water with a mean of -.237°C/mm MSF. Overall heat conductance was 3.34 times greater in water. However, this ratio was not fixed but varied as an inverse curvilinear function of MSF. Thus, equivalent water-air temperatures also varied as a function of MSF. Between limits of 100 to 250% of resting heat loss the following relationships between MSF and equivalent water-air temperatures were found:

MSF (mm)	Regression	Useful Ta Range of Regression (°C)
5	Tw = .24 Ta + 28.11	24.9 - 6.6
10.23	Tw = .30 Ta + 25.92	23.7 - 4.5
15	Tw = .36 Ta + 23.73	22.6 - 1.0
20	Tw = .405 Ta + 22.02	21.5 - (-) 1.8

Key words: Water-air thermal equivalence, skinfolds and human thermoregulation, lower critical water and air temperatures, oscillations in oxygen consumption in the cold.

INTRODUCTION

Aside from the hydrostatic effects of water immersion, the essential difference between air and water immersion lies in the very large combined heat transfer coefficient of still water as compared to still air (Bullard and Rapp, 1970; Colin et al., 1970; Nadel et al., 1974; Witherspoon et al., 1971). This much higher "convective character" (Webb, 1970) of water causes the skin temperature to be clamped near that of the water temperature (Tw), and the normal longitudinal temperature gradient along the limbs to be rendered very small. In spite of the orders of magnitude greater convective character of water, heat loss to water has been estimated to be only about 2 to 5 times that seen in air of the same temperature (Molnar, 1946), indicating that heat loss in water is largely limited by the body (core to skin) tissue insulation and not the skin to water heat transfer coefficient.

Gagge (1940) showed that below the lower critical temperature (Tc) the tissue heat conductance in air and water was similar although Tw was 8 to 11°C warmer than the "equivalent" air temperature (Ta). Gagge argued that the physiological mechanisms which increase tissue insulation and heat production in cold air and water must be similar. While data such as these could be used to construct an approximate water-air equivalent temperature scale, the problem is complicated by several factors. Firstly, the head is usually excluded from immersion in water studies and included in air studies. Since the head is known to be a major site of heat loss in the cold (Froese and Burton, 1957), a way to correct this must be found. In the present study, the head was excluded from the water immersion bath but head heat loss was estimated in the air and water experiments and the result subtracted from the total heat loss. Secondly, the state of acclimation should be controlled since cold acclimation seems to decrease the shivering response in air (Davis, 1963; Newman, 1969) and water (Hanna and Hong, 1972; Hong, 1973; Rennie, 1965; Skreslet and Aarejord, 1968). Thirdly, the major factors determining heat loss from resting vasoconstricted subjects are the subcutaneous insulation in both air (Buskirk et al., 1963; Daniels and Baker, 1961; Jequier et al., 1974; Wyndham et al., 1968) and water (Cannon and Keatinge, 1960; Carlson et al., 1958; Hanna and Hong, 1972; Hong, 1973; Kollias et al., 1974; Pugh, 1965; Rennie, 1965; Sloan and Keatinge, 1973), and the velocity of the media. Medium velocity often is not reported in water studies and subcutaneous fat varies among those studies in which it is reported. These problems preclude a quantitative comparison of the relative rates of heat loss in air and water and the effect of subcutaneous fat from a review of the literature.

The present study eliminates the second and third problems mentioned above by comparing the air and water-immersed heat loss of subjects with similar thermal histories exposed to cold air and water of a similar low velocity. We have simplified the problem of analysis by using a linear approximation of overall heat transfer (Bakken and Gates, 1974; Bullard and Rapp, 1970; Scholander et al., 1950; Wilkerson et al., 1972) along the temperature gradient (ΔT) from rectal temperature (T_{re}) to ambient. The data obtained were used to derive the critical temperature gradient (ΔTc), the lower critical water (Tcw) and air (Tca) temperatures and finally, resting heat loss scales relating equivalent air-water temperatures as a function of unweighted mean skinfold thickness (MSF). The results are discussed with emphasis on water immersion, with a view toward aiding those interested in comparing resting thermoregulation in air and water.

METHODS

Subjects and skinfolds. Fourteen male students were recruited as subjects. All were born in Hawaii except one who was born in Southern California. Their individual characteristics are presented in Table 1. Most had travelled to the mainland United States, but none had done so within the preceeding six months. Thus, their recent thermal history was similar and they were probably not cold acclimatized. Table 1 suggests that the men were heterogeneous in body size, shape, and subcutaneous fat thickness. They also differed in ethnic background, 9 were of Japanese ancestry, and 5 of Caucasian ancestry. Ethnic group had no important effects upon the variables considered here (Hanna and Smith).

Subject[+]	Age, yr	Weight, kg	Surface Area, m^2	Mean Skinfold, mm	%[++] Body Fat
1 (J)	22	79.9	1.88	18.60	38.0
2 (J)	20	59.1	1.67	5.55	12.0
3 (J)	19	84.5	1.98	13.90	31.4
4 (J)	24	62.4	1.70	9.72	23.7
5 (J)	20	53.4	1.56	6.61	15.9
6 (J)	18	61.4	1.66	9.11	22.3
7 (J)	20	73.5	1.81	15.89	34.3
8 (J)	20	84.5	2.05	13.50	30.7
9 (J)	18	50.4	1.53	5.60	12.3
10 (C)	27	69.0	1.85	9.60	23.5
11 (C)	18	59.9	1.79	4.06	2.0
12 (C)	22	74.8	1.97	8.16	20.2
13 (C)	22	66.6	1.84	7.44	18.3
14 (C)	20	97.3	2.20	15.44	33.7
mean	20.7	69.76	1.82	10.23	22.7
SD	±2.5	±13.37	±0.19	±4.49	±10.2

Table 1. Physical characteristics of subjects

+J, Japanese; C, Caucasian

++ $\frac{\text{Adiposity}}{\text{Body Weight}}$ x 100; adiposity calculated from skinfolds after (1).

calipers by one experienced observer at the following sites: Chin, dorsal upper arm, largest part of lateral forearm, juxta-nipple, midaxillary line at level of sternum, waist midway between umbilicus and midaxillary line, center of anterior thigh, widest part of dorsal calf and subscapular point.

Resting heat production. Heat production at rest in a thermoneutral environment (29°C) was measured in a separate experiment. After resting in the laboratory for one hour, the subjects, except for the head, were enclosed in a plastic suit at 29°C. Oxygen consumption ($\dot{V}_{O_2}$), T_{re}, and $\bar{T}s$ were measured after 30 minutes in the supine position. The head was enclosed in an insulated hood and the face further protected by a collar and head heat loss is assumed to be negligible. Since all individuals were relaxed during this 29°C exposure, the heat exchange values were taken as resting and the temperature was taken to be thermoneutral (Hardy and Soderstrom, 1938). The procedure is part of a controlled hyperthermia test which has been described by Fox et al. (1968).

Cold water experiments. The subjects reported to the laboratory one hour before the experiment and rested in a nonstressful environment (24°C) fully clothed. They were asked to abstain from food for two hours before reporting. The cold water regimen has been described elsewhere (Hanna and Hong, 1972; Rennie et al., 1962), hence will be only summarized here.

Each man was immersed to the neck in a stirred water bath for three hours. The posture assumed was the same as for the cold air exposure (see below). Mean water velocity over the body surface measured via dye injections was approximately 3 m/min.[1] Temperature of the bath (Tw) was controlled to ± 0.01°C. T_{re} accurate to ± 0.01°C was measured with an indwelling rectal thermistor inserted 10 cm and noted every 10 minutes. $\dot{V}_{O_2}$ was measured for at least 3 minutes every 20 minutes with a Collins recording spirometer.

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[1] In order to attempt to equate the water and air velocities, we measured the water velocity at three depths at 8 locations in the immersion tank by injecting a small bolus of dye from a syringe needle inserted through the center of a thin sheet of plexiglass about 1 ft^2. The time required for the dye to spread in any direction until it crossed one of three concentric lines etched in the plexiglass at known radii from the center was noted. Although the water was circulated by two pumps, causing complete mixing in 60-75 sec (Hanna and Hong, 1972), it appears that the mixing was primarily effective at the water surface. Depths and mean water velocities (± SE) in m/min were as follows: surface, 13.0 (± 3.0); 4 cm beneath the surface, 2.8 (± 0.51); at thigh level (about 35 cm beneath the surface), 0.9 (± 0.01). Thus, we chose an air velocity of 3 m/min as being approximately equal to the mean water velocity flowing over the body surface. In air, this low velocity is well below that required to cause appreciable forced convection, while in water, this velocity probably leads to primarily forced convection (Bullard and Rapp, 1970). However, free convection at least partly prevailed in the water bath also, since the subjects reported a cold sensation upon limb movement after 3 hours of immersion, indicating that skin temperature was not exactly equal to the bulk water temperature, and, therefore, at least a thin layer of insulating water existed.

Subjects were immersed at least two times on different days and the lowest temperature which could be tolerated for three hours without visible shivering was established. Usually this required only two trials, but on some occasions three or more were needed. Minimal Tw ranged from 29°C in fat subjects to 31°C in lean ones. Total exposure time on the non-shivering trial was 3 hours.

Cold air exposure. The cold air studies also included a 1 hour pre-trial rest. The general procedure followed that reported by Wilkerson et al. (1972) except 10, 20, and 25°C Ta were used. Subjects were exposed to these Ta's in an unordered fashion such that some experienced the 10°C cold first while others experienced the 20 or 25°C first. Exposure was in a climate chamber whose temperature was controlled within ± 0.5°C. Relative humidity ranged from 40 to 60% and air velocity measured with an Alnor hot-wire anemometer was approximately 3 m/min flowing over the long axis of the subjects. Subjects reclined with limbs apart and hands open on a mesh lounge chair, clad in thin athletic shorts or swimming trunks. $\dot{V}_{O_2}$ was measured for 5 minutes every 30 minutes with a Collins spirometer. Total exposure time at each Ta was three hours.

Rectal temperature was taken as before, and surface temperatures were recorded every 30 minutes with thermistors attached with two-sided tape. Surface sites included forehead, midlateral upper arm, dorsum of hand, juxta nipple, midaxillary line at level of xyphoid, subscapula, waist (midway between midaxillary line and umbilicus), midanterior thigh, midlateral calf, and dorsum of foot.

Derived measurements. Heat loss was computed by adding to the metabolic heat production the 20 (water) or 30 (air) minute changes in heat storage. For the latter, mean body temperature was established by the following formula: $\bar{T}b = 0.7\ T_{re} + 0.3\ \bar{T}s$. A caloric value of 4.8 kcal/l O_2 was assumed with an 8% respiratory heat loss. In water, skin and water temperatures were assumed to be equal. Mean skin temperature in air was computed as presented in (1).

(1) $\bar{T}s$ = (.30 juxta nipple + .15 upper arm + .10 dorsum hand + .15 thigh + .05 calf + .05 dorsum foot)/0.8 (Buskirk et al., 1963).

Head temperature was excluded from $\bar{T}s$ in air to facilitate comparison of air and water media since the head was not immersed. For each subject, forehead heat flow was measured with a Hatfield-Turner heat flow disc and this value, expressed per unit of head surface area derived from linear measurements (Weiner and Lourie, 1969), was subtracted from the total heat loss at each temperature. Head heat loss for each subject was extrapolated to 30°C air and this value was subtracted from total heat loss in water. Thus, all air and water heat loss data reported here exclude the head heat loss.

In order to make the comparison of heat loss in water and air as real as possible, we included the cutaneous evaporation in the air heat loss calculation.

Linear regression of percent of resting body heat loss versus the rectal to ambient temperature gradient (ΔT) was performed for each subject in air and water. A sample regression is presented in Figure 1. The data points represent temperature and heat loss values obtained during the final 140 minutes of water immersion and final 90 minutes in air. The slope of each regression curve was computed by ordinary linear regression and again with the intercept fixed at zero (Steel and Torrie, 1960). This latter procedure assumes zero heat loss when ΔT is zero. The slope is an estimate of body core to ambient conductance and was found to be 4.43 times greater in water than in air (Figure 1) for the leanest subject. A similar computation was undertaken for each of the 14 men. Each subject's Tca and Tcw (Table 2) was determined by solving each regression equation in air and water for the critical temperature gradient (ΔTc) (Scholander et al., 1950) at 100% of resting heat loss and subtracting the result from 37°C. Further treatment of the data is explained below.

RESULTS

Heat production and critical temperature. Figure 1 shows typical results obtained in this study for water and air immersions. This subject, our leanest, occasionally shivered in 25°C air. It is interesting that in water the fluctuations in heat production are not as pronounced as in air. This tendency was noted in other lean subjects as well.

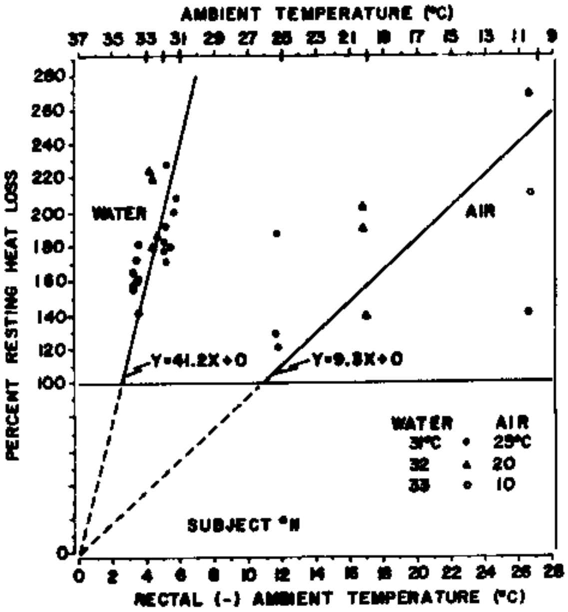


Figure 1. Linear regression analysis through the origin of heat loss (expressed as a percentage of that measured in thermoneutral air) as a function of the measured temperature gradient between rectal and several ambient temperatures in water and air.

	CONTROL	WATER				AIR (Mean Ta=18.33)		
Subject	Heat Loss (watts/m^2)	Mean Tw, °C	Tcw, °C (T_{re}-Tw)	Heat Loss (watts/m^2)	Tcw, +°C	Tca, °C (T_{re}-Ta)	Heat Loss (watts/m^2)	Tca, +°C
1	45.11	29.77	7.06	51.92	30.90	17.45	57.88	23.00
2	43.23	30.80	6.26	79.02	33.58	18.61	62.29	23.61
3	44.37	30.73	6.65	57.19	31.84	18.55	57.39	22.20
4	37.02	29.70	6.21	61.23	33.25	17.95	50.98	22.71
5	26.56	30.73	5.23	44.99	33.94	17.13	48.37	26.90
6	46.19	32.21	4.70	48.87	32.45	18.66	55.85	20.47
7	35.19	30.73	5.60	43.35	32.12	18.24	49.81	23.24
8	37.62	30.62	6.45	52.78	32.34	18.90	42.50	19.34
9	30.31	30.67	6.08	59.77	33.91	17.99	51.07	26.38
10	33.91	31.00	5.58	47.23	32.94	18.31	56.36	25.85
11	34.08	32.00	4.34	62.36	34.57	18.36	61.41	26.21
12	36.72	31.80	4.64	44.89	33.22	18.03	59.45	25.12
13	49.35	31.80	4.18	61.49	33.70	18.22	64.58	22.65
14	36.87	29.00	6.60	43.02	31.27	18.22	52.83	23.72
$\bar{X}$	38.32	30.83	5.68	54.15	32.86	18.19	55.05	23.67
SD	±6.49	±0.92	±0.94	±10.09	±1.08	±0.47	±6.11	±2.23

+ Note that Tcw and Tca are based on linear regression analysis through the origin (Figs. 1 and 3) and not on the average data shown in this table.

Table 2. Resting body heat loss in 29°C air (control) and other average responses of each subject exposed to several cold water and air immersions.

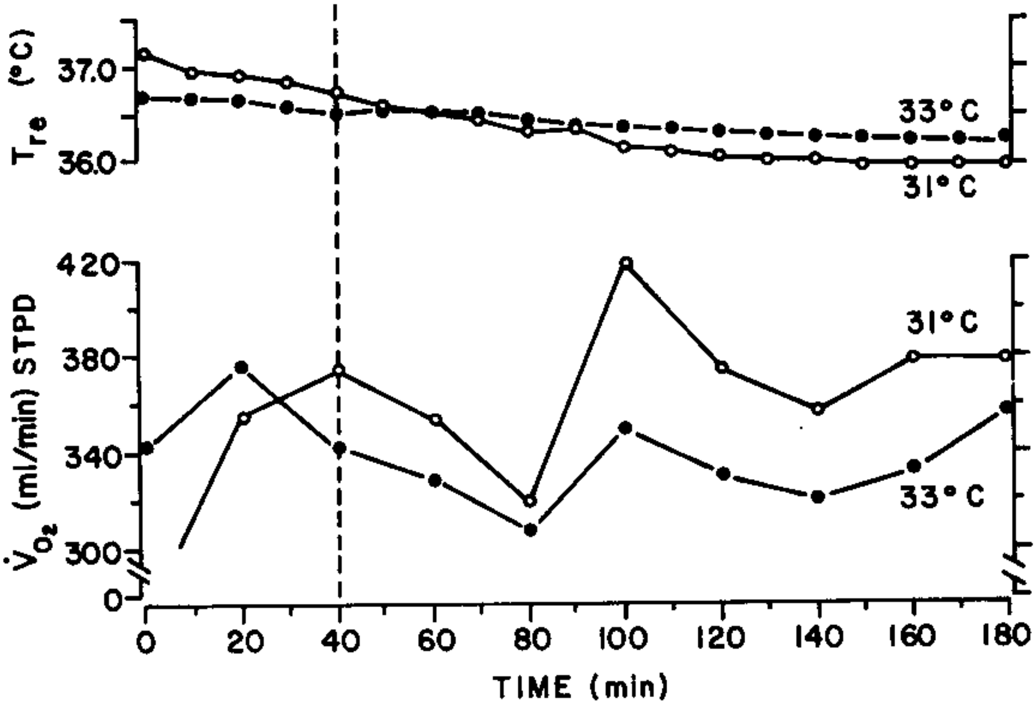


Figure 2. Oxygen consumption and rectal temperature of Subject 11 as a function of time during immersion to the neck in water at 31 and 33°C. This subject's $\dot{V}_{O_2}$ in 29°C air was 218 ml/min, STPD.

Plots of $\dot{V}_{O_2}$ versus time (Figure 2) in water (where we have more frequent measurements than in air) disclosed regular oscillations in four subjects' data, even when visible shivering was not detected. In these four subjects, the oscillations had a mean peak to peak period of 120 minutes (range 60-160 minutes). For a single subject, the period did not seem to change in the narrowly different water temperatures used here, but the amplitude of the oscillations tended to increase with decreasing water temperature (Figure 2), and the leanest subjects tended to have shorter periods. The average peak to trough change in $\dot{V}_{O_2}$ was 15% of the mean value for the four subjects. No obvious correlation with T_{re} could be seen (Figure 2).

Subject 11 showed no visible shivering in the 33°C water experiment yet the heat production was obviously increased above the 29°C air control value (Figure 1). According to the method of Rennie et al. (1962) the Tcw for this subject would be 33°C (no sustained visible shivering during three hours at 33°C). According to our linear approximation method, this extremely lean subject's Tcw is 34.57°C (Table 2). The difference in Tcw is obtained by the two methods is probably the result of the ability of the linear approximation method to include the effect of a general increase in muscle tone which often occurs before visible shivering can be detected. Perhaps this explains why the group mean Tcw of 32.86°C (Table 2) is somewhat higher than the value of 31.2°C previously determined in this laboratory using the method of Rennie et al. on 15 residents of Hawaii with MSF = 10.6 mm (Hanna and Hong, 1972).

The individual and group mean data are summarized in Figure 3 and Table 2. In two cases, individual's regression lines are not visible in Figure 3 due to overlap. Table 2 shows that the overall mean body heat loss in water and air was not different in spite of a much smaller ΔT in water. Linear regression of % of resting body heat loss (Y) versus the T_{re} to ambient temperature gradient (X) for each individual yielded individual conductance values (slopes in Figure 3). The mean result ($\pm$ SE) of these calculations for water and air respectively were: $Y = 25.72\ X\ (\pm\ 1.96) + 0$ and $Y = 7.70\ X\ (\pm\ 0.22) + 0$. In terms of watts/($m^2 \cdot {}^{\circ}C$) this is equivalent to $Y = 9.81\ X + 0$ and $Y = 2.93\ X + 0$. The mean conductance ratio (water/air) was 3.34 (Figure 3), ranging from 2.30 for the fattest subject to 4.45 for the leanest subject. The mean % resting body heat loss and ΔT gradient ($\pm$ SE) are indicated for these two subjects in water (Figure 3) to demonstrate the importance of subcutaneous fat on the heat conductance in water.

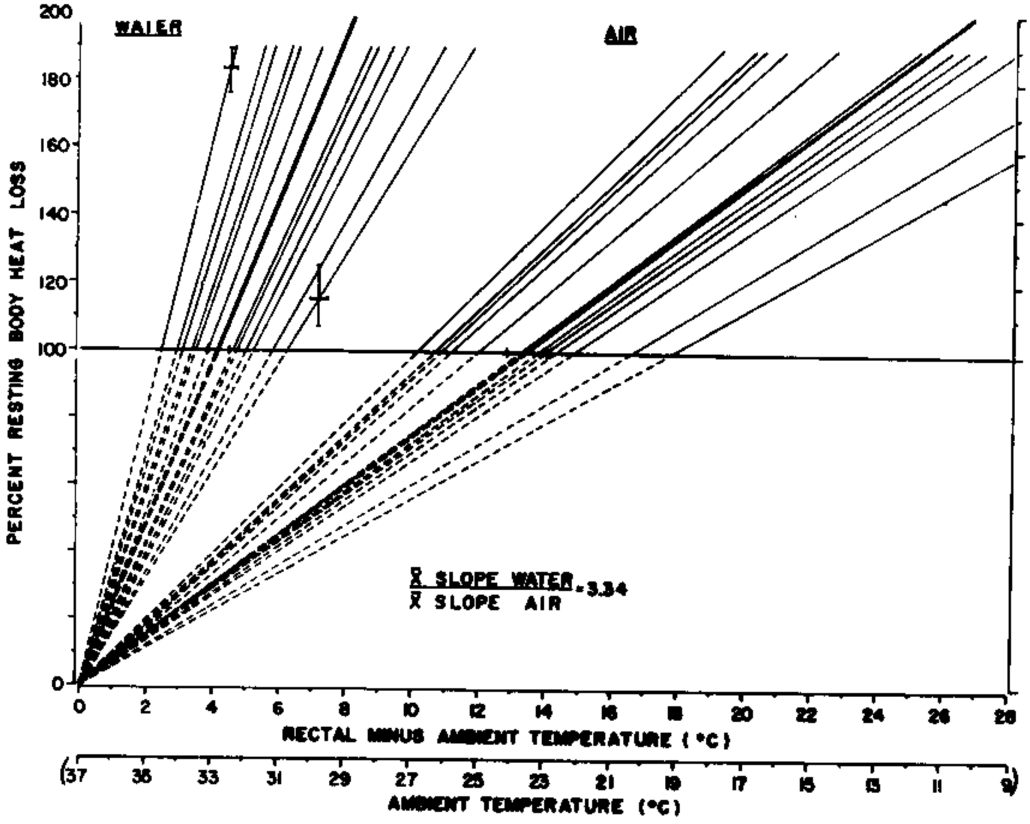


Figure 3. Percent of resting body heat loss versus the T_{re} to ambient temperature gradient in water and air for individuals (thin lines) and the group means (thick lines) The average water and air critical temperatures ($\pm$ SE) are shown on the 100% horizontal line. Also shown, in water, are mean ($\pm$ SE) values for the fattest and leanest subjects (nos. 1 and 11, respectively). Ambient temperature is shown in parenthesis for convenience.

As theory would predict (Bakken and Gates, 1974; Scholander, 1950), and in agreement with other workers (Wilkerson et al., 1972), body heat loss was a linear function of Ta in this study. Mean body heat loss in 25, 20, and 10°C air was 43.90, 53.00, and 70.78 watts/m^2 respectively (r = .99)

Regression through the origin (Figures 1 and 3) is only accurate for dry (non-evaporative) heat loss. In spite of this, we have not subtracted an estimate of cutaneous evaporative heat loss in our air heat loss calculation because we felt to do so would dilute our purpose of comparing normal body heat loss in air and water. Including the cutaneous evaporation was probably responsible for our finding a positive intercept in air of 27 ± 9.2% with ordinary linear regression, which means we have over-estimated the slope of the "dry" heat loss curve in air by about 15%. Thus, we have slightly underestimated the "dry" water/air conductance ratio. Interestingly, the included skin evaporative heat loss was nearly balanced by the excluded head heat loss in air (on the average, 19.6% of the body heat loss) so that the Tca's reported here are similar to values previously reported using a similar method but including head heat loss (Wilkerson et al., 1972).

<u>The effect of subcutaneous fat</u>. The importance of subcutaneous fat on the lower critical water and air temperatures determined by regression analysis is shown in Figure 4. The linear regression analysis indicates that the range of critical temperatures found in this study (Table 2) is mainly a result of variability in the subjects' subcutaneous fat thickness. The linear correlation coefficient in water (-0.95) is surprisingly high considering the several assumptions inherent in the method of analysis. The linear correlation in water is considerably stronger than that previously determined in this laboratory (-0.74) by one of us (Hanna and Hong, 1972) using the method of Rennie et al. (1962) on 15 subjects. Thus, the extra computations necessary with the regression method may be justified by what appears to be improved accuracy in the determination of Tcw as a function of MSF.

The linear correlation in air is much lower than in water (Figure 4). Nevertheless, the regressions for both air and water are considered to be significantly different from zero ($t = 1.60$, $df = 12$, $0.1 > P > 0.5$; and $t = 10.81$, $df = 12$, $P < 0.001$, respectively). Interestingly, the slope in water and air was not different ($t = 0.0065$), which suggests that subcutaneous fat has a quantitatively similar influence on Tcw and Tca. Assuming the slopes are indeed identical, the Tcw was 9°C higher than Tca irrespective of MSF. This observation agrees well with the 8-11°C difference in operative temperature reported by Gagge (1940) at minimal values of conductance.

<u>Equivalent air-water temperatures</u>. The purpose of this study was to produce equivalent water-air temperatures. This purpose was met by constructing Figure 5 in the following manner. Firstly, we solved for Tca and Tcw for the values of MSF illustrated in Figure 5 using the relationships shown in Figure 4. The mean slope for water and air was used (-.237°C/mm MSF, Figure 4) since the slopes in air and water were not different. Using these critical air and water temperature values, we solved for Tc assuming a T_{re} of 37°C, and determined the heat conductance value for a given skinfold thickness. Then, assuming a constant conductance below Tc (see DISCUSSION), the

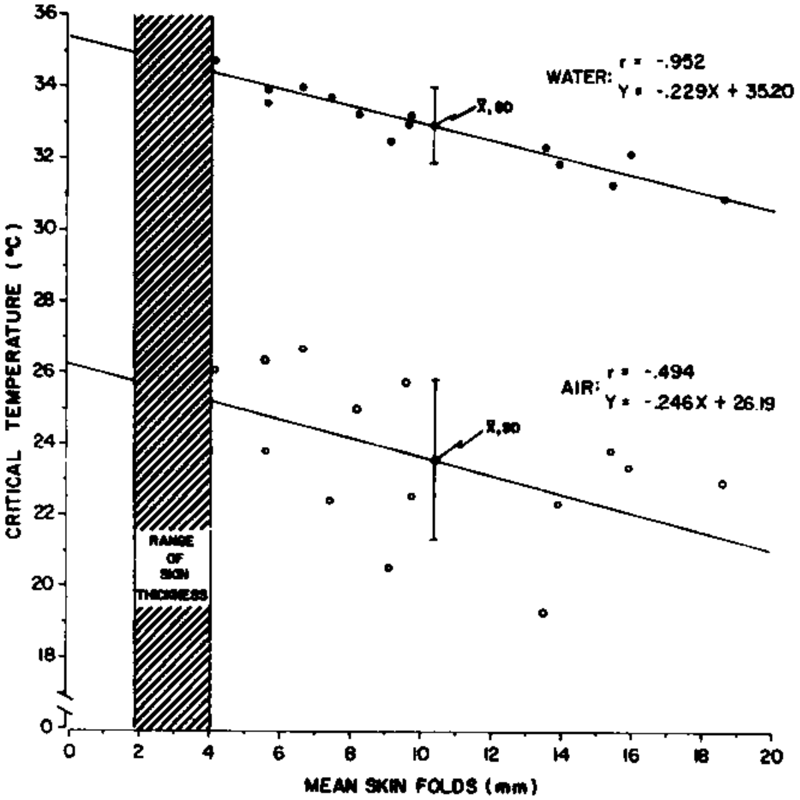
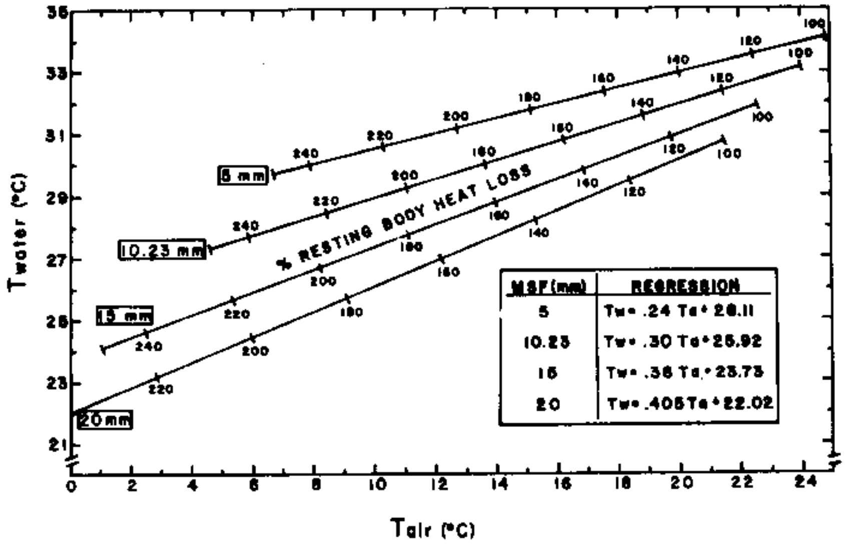


Figure 4. Linear regression of lower critical air and water temperatures versus mean skinfold thickness. The approximate range for a double thickness of skin is shown.



MSF (mm)	REGRESSION
5	Tw= .24 Ta+ 28.11
10.23	Tw= .30 Ta+25.92
15	Tw= .36 Ta+23.75
20	Tw= .405Ta+22.02

Figure 5. Water and air temperatures evoking equivalent resting heat loss for several mean skinfold thicknesses. The regression equations shown should not be applied beyond the limits of 100 to 250% of resting body heat loss as indicated by the lines in the figure.

values of ΔT (T_{re} - Ta or Tw) were computed for various levels of heat loss up to 250%. By subtracting ΔT values from 37°C, actual air and water temperatures at each heat loss level were computed. Finally, we performed linear regression analysis on the metabolically equivalent air and water temperatures at each MSF to produce the regression equations shown in Figure 5. The final equations are too highly derived to allow an error estimate, and although the lines in Figure 5 appear very precise, Figures 1 and 4 indicate the variability inherent in these data. It should be noted that all the lines in Figure 5 originate at 37°C where heat loss in both air and water is nil, and that the slopes are simply the inverse of the water/air conductance ratios at the various MSF.

The rationale for extending the straight lines in Figure 5 beyond the maximal value of about two times the resting metabolic rate which we recorded for the thinnest subjects is as follows. Craig and Dvorak (1966) found that $\dot{V}_{O_2}$ increased linearly with decreasing Tw, until it plateaued at about 2.4 times rest in Tw = 24-26°C. Cold air studies also suggest a plateau in the $\dot{V}_{O_2}$ at about 2.5 times rest in a lean individual at Ta 5°C (Wyndham et al., 1968). Although other authors (Behnke and Yaglou, 1951) have recorded a 5 to 6-fold increase in $\dot{V}_{O_2}$ during a brief exposure to ice cold water, the maximum sustained $\dot{V}_{O_2}$ during a three-hour exposure to cold air and water may be only about 2 to 3 times resting. Indeed, a summary of the earlier literature (Hemingway and Stuart, 1963) of cold air and water immersion indicates that of 8 studies which used relatively low media velocities only one reported a mean increase in $\dot{V}_{O_2}$ greater than 3 times the resting nonshivering rate. Thus, we extrapolated the straight lines in Figure 5 to only 2.5 times the resting heat production, and we do not recommend making predictions from the equations in Figure 5 beyond the limits of Tw and Ta indicated by 250% of resting metabolism.

Another assumption in the construction of Figure 5 is that T_{re} is regulated at 37.0°C in spite of a marked decrease in $\bar{T}_b$. Wilkerson et al. (1972) found that T_{re} increased slightly with decreasing Ta during three hours at Ta as low as 8°C. Data of Wyndham et al. (1968) suggests that T_{re} is lowest (about 36°C) in both fat and lean men in 10°C air but rises at Ta's above or below 10°C. We found a tendency for T_{re} to fall slightly with decreasing Ta's. However, the maximum mean decrease from the thermoneutral control value of 36.71 ± .09°C was only 0.19°C in water and 0.09°C in 10°C air. Thus, assuming a regulated T_{re} of 37°C at rest does not introduce an important error in the water/air heat loss relationship.

Figure 5 can be used to predict the body heat loss from a resting man in water or air if MSF are known, as well as to determine equivalent air-water temperatures. Craig and Addae (1969) studied finger temperature in air and water and found an air temperature of 20°C to be equivalent to 32°C water, which agrees reasonably well with our findings at near minimal MSF. In addition, Figure 5 shows that the effect of MSF is slight at Tc but becomes increasingly important at higher rates of heat production. It is interesting to note that men with MSF = 10.23 mm need not be exposed to Tw's lower than 27°C in order to evoke a metabolic response comparable to air of about 5°C. Hence, it is not surprising that thermally nude divers who habitually enter the 26°C waters around Hawaii experience a significant cold stress which may lead to a cold adaptation (Hanna and Hong, 1972).

DISCUSSION

Oscillations in $\dot{V}_{O_2}$

It is by no means clear why there was more variability in the metabolic response to cold air than to cold water (Figures 1 and 4). (Part, but certainly not all, of the difference may be artifactual due to assuming Ts in water, using a narrower range of Tw than Ta and to having more data points in water (189) than in air (126)). This difference in the shivering response in air and water has not been specifically reported in the literature and deserves further investigation. It may be that nearly abolishing the longitudinal gradients of $\bar{T}s$ in water somehow altered the control of shivering. Or perhaps warmer peripheral tissues in water depressed the amplitude of the shivering cycles (the overall mean Ta of 18.33°C led to an overall $\bar{T}s$ of 26.82°C, while the overall mean Tw was 30.83°C). In addition, the near weightless state in water may alter the proprioceptive input and hence the shivering response Perkins, 1945).

Oscillations strikingly similar in amplitude and period to those in Figure 2 have recently been reported by Bailey et al. (1973) for resting subjects exposed to presumably thermoneutral air (ambient conditions not specified). No definite explanation was given for the periodic changes in $\dot{V}_{O_2}$ in air, but the similarity to changes seen in the present study during immersion in moderately cold water suggests a similar mechanism. If this is true, it would tend to favor the second of the three hypotheses mentioned.

Peripheral vasoconstriction

The comparison of heat loss in air and water by linear approximation of heat loss via indirect calorimetry requires assumptions, some of which have been discussed above, and others will be dealt with where appropriate below. However, the most important assumption is that the degree of peripheral vasoconstriction in these two media was (ideally) near maximal, but more importantly, similar. The degree of vasoconstriction in this study is not known but can be inferred from the literature to be near maximal by a consideration of immersion time, Tw and Ta, and the degree of shivering:

Immersion time. The importance of immersion time in estimating heat loss by indirect calorimetry is seen in a recent report by Nadel et al. (1974). Resting heat loss after 7 to 10 minutes in still water as estimated by $\dot{V}_{O_2}$ was only 22% of that measured with heat flow discs, while the two methods were in "close agreement" after 30 minutes immersion during steady-state exercise. The authors' conclusion that the time-honored technique of indirect calorimetry is reliable in water during exercise when the peripheral circulation is increased but not during rest seems unwarranted. Rather, their data show that 7-10 minutes is not enough time to achieve steady heat flow from cooling peripheral tissues (Mitchell et al., 1970) or to allow the $\dot{V}_{O_2}$ to increase (Craig and Dvorak, 1966). We found no tendency for the group mean heat production to rise or fall as a function of time after 40 minutes in water or after 90 minutes in air and thus felt justified in using all data after these intervals. While it is possible that our assumption of similar core-shell weighting factors after these intervals in water and air introduced a systematic error in the calculation of changes in heat storage, we doubt that the error can be large. As noted previously, changes in T_{re} were modest during the final 90 minutes in air and in the final 140 minutes in water. In addition, the average $\bar{T}s$ in each of the 3 cold air temperatures used fell less than 0.5°C during the final 90 minutes.

Tw and Ta. Recently, Kollias et al. (1974) have questioned whether maximal vasoconstriction is possible in water as warm as 30°C. This conclusion was based upon the higher insulation values they obtained with women immersed for 60 minutes in 20°C water than those reported by Rennie (1962) during immersion for 3 hours in water at 30°C and above. This argument is not a strong one since others (Hanna and Hong, 1972; Hong, 1973) have also used 30°C immersions and find insulation values similar to those calculated by Kollias et al. Indeed, earlier work (Hanna and Hong, 1972) from this laboratory (which followed Rennie's protocol) specifically noted the unusually low insulation values reported in the one study by Rennie. More direct evidence that vasoconstriction in non-obese subjects can be maximal in 30°C water is as follows. Rennie (1965) has shown that forearm blood flow is almost zero after about 40 minutes of body immersion in 30°C water. Craig and Dvorak (1966) found body tissue conductance to be minimal after 60 minute immersions in Tw's from 30 to 24°C. Except for two very obese subjects, the data of Cannon and Keatinge (1960) demonstrate minimal tissue conductance during immersion to the neck in 30°C water. Direct calorimetry studies indicate maximal vasoconstriction at Ta below about 28°C (Hardy and Soderstrom, 1938) or Tw below 33°C (Burton and Bazett, 1936). Simply surrounding the forearm with 30°C water while the rest of the body was presumably comfortable led to near minimal forearm blood flow (Barcroft and Edholm, 1943). While fatter subjects were exposed to colder Tw's than lean subjects, we found for our group of subjects with MSF = 10.23 mm the same overall degree of vasoconstriction to MSF tissue in water at about 31°C as in air at 18°C (i.e. similar slopes, Figure 4). Figure 5 and the data of others (Kollias et al., 1974; Sloan and Keatinge, 1973) show that average subjects probably would be unable to approach a steady state during 3 hours in water at 20°C, which would tend to increase the uncertainty associated with the assumptions of indirect calorimetry. Thus, we doubt that water as cold as 20°C must be used to provoke maximal vasoconstriction, although we recognize the important influence of MSF (Figure 4), which might cause obese subjects to require the use of temperatures colder than 30°C and/or longer immersion times than one hour to accurately establish Tcw.

Shivering. Below Tc shivering is thought to increase tissue perfusion (Burton and Bazett, 1936). However, the resulting change in tissue conductivity is slight, increasing from about 11.0 watts/($m^2 \cdot {}^oC$) when not shivering to about 15 watts/($m^2 \cdot {}^oC$) during maximal shivering (about 5 met) in 20°C water (Spealman, 1949). This should be compared to the 4-fold change in effective conductivity found in resting humans between maximal vasoconstriction in the cold to maximal vasodilation in the heat (Robinson, 1949). Importantly, the average degree of shivering in the present study in air and in water was moderate (about 50% above rest) and not significantly different (Table 2). Under these conditions the assumption of minimal blood flow to peripheral tissue is approximated while the necessary increase in heat production required to determine Tc is still obtained.

However, even if the intensity of shivering had been greater and different in air and water, similar vasoconstrictor tone probably would have existed since forearm occlusion studies (Craig, 1971; Hong et al., 1969; Rennie and Marder, 1968) indicate that most of the increased heat conductance accompanying shivering does not depend on changes in muscle perfusion, but rather is a direct consequence of increased forearm metabolism due to shivering. It seems most likely that increased values of tissue conductance (see above) calculated during bouts of shivering are not due to warm core blood suddenly perfusing the muscles, but rather a result of using the unchanging (T_{re}-$\bar{T}s$) temperature gradient to calculate tissue conductance when much of the increased heat production is occurring in the shell. The increase in shell deep muscle temperature during shivering would provide an increased gradient for conductive heat loss with a smaller increase in true tissue conductivity than one calculates on the basis of a core to surface temperature gradient.

We conclude from the above that tissue insulation in air and water in the present study was near maximal and similar. Thus, the use of straight lines (constant tissue conductance) in Figure 5 is justified, and observed differences in heat loss (at fixed MSF) in air and water must be due to differences in the air and water insulation.

Lower critical water temperature and MSF

Our method of comparing heat loss in air and water is based on the concept of lower critical temperature (Tc) as discussed by Scholander et al. (1950). Strictly speaking, the definition of Tc is simply that ambient temperature below which the resting heat production is increased (Bligh and Johnson, 1973). However, implicit in this definition are the ideas that maximal insulation is reached before the metabolism is increased, that this maximal physical insulation is maintained below Tc, and that the core temperature is controlled at near 37°C by increases in heat production linearly proportional (within limits) to the core to ambient temperature gradient. While neither Newton's law of cooling nor Fourier's law of heat conductance is strictly applicable here (Bakken and Gates, 1974), empirically, these ideas are sound in both air (Scholander et al., 1950; Wilkerson et al., 1972) and water (Bullard and Rapp, 1970). However, Cannon and Keatinge (1960) have challenged the validity of these concepts in water since they found that fat and lean men alike increased heat production in Tw around 33°C, leading these workers to propose an alternate term, the "metabolic threshold temperature" which is nearly the same for all subjects. However, obvious difficulty in determining the first increase in metabolism (which is the reason for the linear approximation method) and the relatively modest effect of MSF on Tcw (Figure 4) do not, by themselves, invalidate the concept of Tcw.

When Tc is determined by the linear approximation method, it is based on not only the first increase in metabolism but also the intensity of the metabolic response below Tc as well as any change in T_{re} (Figures 1 and 3). According to Figure 4 this method seems to be even more suited for use in water than in air. We constructed Figure 6 to further demonstrate the usefulness of Tcw as determined by the "intercept" (Wilkerson et al., 1972) or linear approximation method. Based on the mean regression in Figure 4 (-.237°C/mmMSF) we calculated Tcw at progressively more negative (hypothetical) values of MSF (Table 3) until a Tcw near 37.0°C was reached. At this point, one may imagine a man with all shell insulation (skin, fat, and some muscle (Carlson et al., 1958)) removed, so that only a core of tissue at 37°C remains generating and losing the resting heat production via only the thermal resistance of the water. Since T_{re} is assumed fixed at 37°C, and heat loss is also assumed constant (resting), $\bar{T}c$ is inversely proportional to ΔTc which itself is proportional to insulation; and we have indicated the approximate insulation provided by muscle, fat, skin, water, and air within the figure. This allowed us to estimate the percentage of body weight represented by the shell and core at Tc[2]. For the group, the effective core was 69% of body weight. For subjects 1 and 11 (fattest and leanest) the values were 64% and 72% respectively. This is within the range of core weight found by direct calorimetry in air (Burton, 1935) or by using a bath calorimeter at Tw around 30°C (Burton and Bazett, 1936).

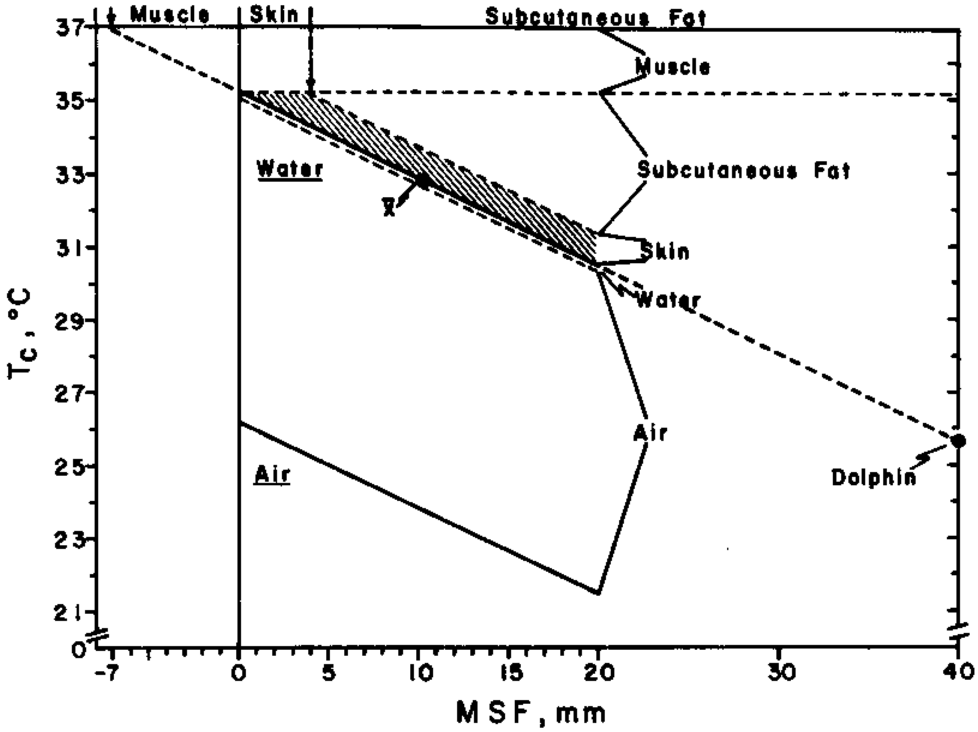


Figure 6. Lower critical water and air temperatures versus mean skinfolds redrawn from Figure 4 using the average slope for water and air (-.237°C/mm MSF). The relationship in water has been extrapolated (heavy dashed line) assuming negligible insulation due to water. Negative values of MSF are hypothetical and represent a double thickness of muscle acting as shell insulation if its heat conductivity were the same as MSF tissue (see text). Four mm have been allowed for a double layer of skin.

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[2] This calculation used an average skin and fat thickness of 5.12 mm (10.23/ 2) and a thickness of muscle of 7 mm since its conductivity is twice that of fat (Mitchell et al., 1970). The volume of shell tissue was then obtained by multiplying this shell thickness by surface area. Shell volume was converted to shell mass using the known density of skin, fat, or muscle (Mitchell et al., 1970); the relative contribution of each tissue was estimated from Figure 6, assuming the same insulating muscle and skin thickness for all subjects. We also assume negligible heat production by the cooled shell tissue.

		WATER			AIR		WATER/AIR
MSF	Tcw, °C (T_{re}-Tw)	Tcw, °C	Conductance, watts/(m^2·°C)	Tca, °C (T_{re}-Ta)	Tca °C	Conductance, watts/(m^2·°C)	Conductance Ratio
-7.5	0.02	36.98	1,916.00	9.03	27.97	4.24	451.89
-7.0	0.14	36.86	273.71	9.15	27.85	4.19	65.32
-6.0	0.38	36.62	100.84	9.39	27.61	4.08	24.72
-5.0	0.61	36.39	62.82	9.62	27.38	3.98	15.78
0	1.80	35.20	21.29	10.81	26.19	3.54	6.01
40	11.28	25.72	3.40	20.29	16.71	1.89	1.80
100	25.50	11.50	1.50	34.51	2.49	1.11	1.35

Table 3. Theoretical water and air critical temperatures (Tc), gradients (ΔTc), and heat conductance values+.

+ Negative values for MSF are hypothetical and represent layers of skeletal muscle insulation. Calculations based on relationships shown in Figure 4 assuming a mean value of -.237 °C/mmMSF, a T_{re} of 37°C, and constant resting heat loss of 38.32 watts/m^2.

A more serious objection to the concept of Tcw raised by Cannon and Keatinge (1960) is the observation that two obese subjects with MSF = 26.75 mm failed to achieve maximal insulation unless Tw as cold as 14°C was used. Our data, based on non-obese subjects predict a Tcw of about 29°C for these obese subjects, and further predict that an extremely obese man with MSF = 40 mm would have a Tcw of 25.7°C and a water to air conductance ratio below Tc of 1.8 (Table 3). We know of no human data with which to test this latter result; but the Atlantic bottlenose dolphin which has 2 cm of subcutaneous fat (MSF = 40 mm) is known to require about twice the resting metabolic rate of a similarly-sized land mammal to remain in thermal equilibrium (Kanwisher and Sundnes, 1966). Indeed, a study (Hampton et al., 1971) of regional surface temperatures and heat flows of an Atlantic bottlenose dolphin in 26°C sea water indicated that heat loss from the extremities was actively reduced at rest. In spite of this, T_{re} fell during periods of sleep and the animal awoke periodically, apparently for the purpose of rewarming. Thus, 26°C water must be near this "obese" animal's Tcw as predicted from the relationships derived in this work from non-obese human subjects.

To summarize, the fact that our most important relationship (Figure 4) may be extrapolated at either extreme to make apparently reasonable predictions reassures us that the concept of Tc upon which we have based this work has intrinsic value in water as well as air (Scholander et al., 1950; Wilkerson et al., 1972). The less than maximal insulation found in obese humans during exposure to cold (Cannon and Keatinge, 1960; Jequier et al., 1974) may be due to lower muscle insulation in these subjects (Jequier et al., 1974) and a likely reason for this is inadequate exposure times to allow adequate peripheral cooling (Mitchell et al., 1970). However, the interesting possibility remains that obese humans used to living in air differ from normal humans and the dolphin in that they are unused to exercising their potential for developing maximal muscle insulation including counter-current heat exchange (the efficiency of which is believed to increase with repeated cold exposure (Hong et al., 1969)).

Water/air conductance ratios

Effect of media properties. Assuming tissue heat conductance to be minimal and similar in these water and air experiments, the mean water/air conductance ratio of 3.34 (at fixed MSF = 10.23 mm) reflects the relative effect of the water/air combined heat transfer coefficients. For still air (including 15% for insensible evaporation) various authors report similar values of about 8 to 9.6 watts/$(m^{2}\cdot{}^{\circ}C)$ (Colin et al., 1970); while for still water where convection is the primary avenue of heat loss, a wide range of convection heat transfer coefficient (hc) has been reported, from 43.6 (Colin et al., 1970) to greater than 200 watts/$(m^{2}\cdot{}^{\circ}C)$ (Nadel et al., 1974). The literature is not clear in explaining the wide difference in values of hc reported in water but the difference is probably not due to errors in calculating heat storage for reasons discussed above (see *Immersion time*). Rather, Table 3 shows how sensitive hc is to small changes in insulation at the tissue-water interface. Similar behavior has been demonstrated with a copper manikin (Goldman et al., 1966; Witherspoon et al., 1971) in which a "thin" electrical insulation coating caused hc to be underestimated by a factor of 3. Table 3 shows that as the "core surface" is reached at about -7 mm MSF (this allows a 0.14°C temperature gradient due to water insulation) adding 1 mm of MSF insulation reduced "hc" from 273 to 101 watts/$(m^{2}\cdot{}^{\circ}$CO.

Thus, if the copper manikin's coat had the same insulating property as MSF tissue, a thickness of only about 0.5 mm could account for the observed factor of three differences in hc.

It should be emphasized that this extreme behavior only occurs near the interface of tissue and water. The points plotted in Figure 3 for the heaviest and leanest subjects show that if substantial tissue is placed between the core and water, that differences in conductance depend largely on differences in heat flow among subjects rather than subtle differences in the temperature gradient. However, at increasingly negative (hypothetical) values of MSF (Figure 7 and Table 3) where the "core-water interface" is approached, the calculated conductance has little to do with heat flow and becomes increasingly sensitive to small differences in the temperature gradient which approaches zero. This is precisely the situation facing those interested in measuring hc in water from T̄s and Tw measurements where the thermal gradient seems to be only about 0.2°C (Bullard and Rapp, 1970; Nadel et al., 1974). Thus, the wide range of hc values reported in water studies is most probably due to differences in the method of measuring T̄s in water. A difference in T̄s of only 0.5°C could account for the range of values for hc reported in the literature (see MSF values -5 to -7 mm, Table 3). Perhaps a dialog to standardize the difficult measurement of T̄s in water should be initiated so that the measurement of hc in water can be more rigidly defined.

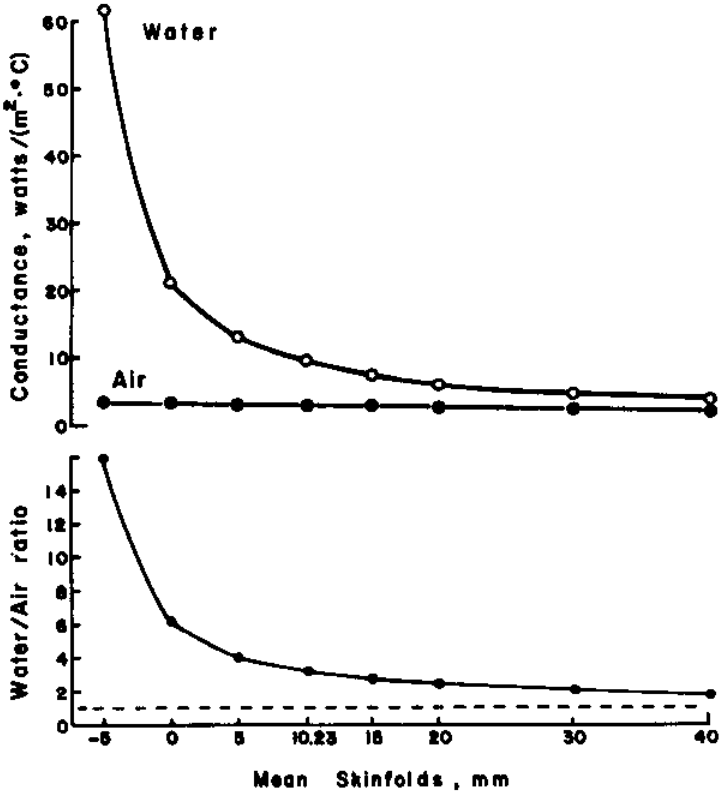

Figure 7. Heat conductance in water and air (top and their ratio (bottom) as a function of MSF. Dashed line (bottom) indicates a ratio of one. Assumptions are the same as for Table 3.

In view of the extremely small temperature gradients between skin and water and the nearly microscopic distance over which these gradients exist, and further difficulty in knowing at what point conduction yields to convection, a combined heat transfer coefficient may be more appropriate than a pure hc (Colin et al., 1970). Webb (1970) combined density, specific heat, thermal conductivity and viscosity into a "convective constant" and found a water/air ratio of 167. In fact, the correct ratio is 1670 (P. Webb, personal communication). That a ten-fold error in the water/air convective constant did not cause difficulty in drawing conclusions shows that such comparisons of water and heat transfer properties are of limited practical usefulness because heat flow through subcutaneous tissues is the rate-limiting step, and, particularly in water, very great changes in "hc" produce only modest changes in $\bar{T}s$. Calculations based on the example given by Bullard and Rapp (1970) further demonstrate this (D. Lally, personal communication). Viewed this way, the conductance ratio of 3.34 while influenced by media properties, must be determined mainly by the subcutaneous tissue insulation.

Effect of subcutaneous fat. Molnar (1946) was one of the first to point out that the 25 times greater thermal conductivity of water leads to only 2 to 4 times greater heat loss in water compared to air at the same temperature. We observed a range of water/air conductance ratios below Tc of from 2.3 to 4.5 for our fattest and thinnest subjects respectively. It is apparent from Figure 7 and Table 3 that this is largely due to changes in heat conductance to water rather than to air. Similar behavior has been demonstrated on a copper manikin with various clothing assemblies (Goldman et al., 1966). That is, the better the insulating properties of the clothing, the lower the water/air conductance ratio. Thus, a range of water/air heat conductance ratios is to be expected when subcutaneous insulation is taken into account. As MSF decreases, the much greater convective character of water exerts its effect at an increasing rate; each mm of MSF is exceedingly important for thin subjects who are greatly disadvantaged in cold water relative to cold air. Except for substantial skeletal muscle insulation and counter-current vascular heat exchange the conductance ratio in these thin individuals would increase towards that expected from a consideration of the media physical properties. The reason for this behavior is that in still air, more than half of the total insulation is due to the air itself (Figure 6) while even a poorly stirred water bath adds practically no additional insulation in series with the tissue shell. As body tissue insulation increases and becomes more rate limiting, the relative importance of the insulative power of the air decreases, and the water/air conductance ratio asymptotically approaches one (Figure 7). This behavior is responsible for the progressive increase in the slopes of Figure 5 with increasing MSF.

Surface area to mass ratio. Kollias et al. (1974) indicate that in 20°C water a family of water heat conductance curves may exist (Figure 7) depending on the SA/mass ratios at any fixed value of fat insulation, in agreement with Sloan and Keatinge's (1973) work on rate of fall of T_{re} in 20.3°C water. We tested the effect of including the SA/mass ratio on our data by regressing Tc against the product of MSF x mass/SA. This had the effect of increasing r (see Figure 4) in water by 0.006 and of decreasing r in air by 0.08, although the "t" statistic increased slightly in both water and air. Thus, if heat loss is low (as it was in this study) the effect of SA/mass ratio (which varied by a factor of 1.3) is not important compared to MSF (which varied by a factor of 4.6). However, both MSF and SA/mass ratio would have an increasingly important effect in colder temperatures when heat flow is greater.

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STUDIES ON ANIMALS

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EVAPORATIVE HEAT LOSS IN THE CALIFORNIA SEA LION AND HARBOR SEAL

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Abstract—1. Total evaporative heat loss was measured in four California Sea Lions and one Harbor Seal at air temperatures between 13 and 30°C. The total heat loss was partitioned into respiratory and cutaneous components.

2. Thermal polypnea was not observed to occur in any of the animals and respiratory evaporative heat loss was estimated to represent less than 3 per cent of the heat production when the animals were hyperthermic in a hot environment (31°C).

3. Active sweating was noted on the bare skin of the flippers of the sea lions but not on the haired surface of the torso. The total cutaneous evaporative heat loss accounted for approximately 16 per cent of the heat produced by metabolism, in a hot environment.

4. It was concluded that evaporative heat loss mechanisms were relatively ineffective in California Sea Lions and in the Harbor Seal also.

INTRODUCTION

In a previous investigation (Whittow *et al.*, 1970, 1972), it was observed that the respiratory frequency of the California Sea Lion did not increase during hyperthermia. Qualitative tests indicated that active sweating does not occur on the torso of sea lions during exposure to heat. These results suggested that evaporative heat loss is relatively unimportant in sea lions in a hot environment. This conclusion appeared to be borne out by the poor heat tolerance of California Sea Lions exposed to warm conditions out of doors (Whittow *et al.*, 1971). The purpose of the experiments described in this paper was to obtain quantitative information on the magnitude of evaporative heat loss in the California Sea Lion and to partition the total evaporative heat loss into its respiratory and cutaneous components. This information is not available for any pinniped, although many species experience hot conditions when they are on land, and their tolerance of such conditions may be important for their survival. The opportunity was also taken to examine evaporative heat loss mechanisms in a single specimen of Harbor Seal.

MATERIALS AND METHODS

The main series of experiments was conducted on three young California Sea Lions (*Zalophus californianus*), aged 3–4 years, body weight, 46–70 kg. One animal was a male while the other two were females. Additional experiments were performed on a younger female sea lion (body weight, 32 kg) and on an adult female Harbor Seal (*Phoca vitulina*, body weight, 98 kg). Each animal was kept out of the water for 1·5–2 hr before an experiment began, so that its coat was dry before it entered the climatic room. The animals were fed approximately 18 hr prior to the commencement of each experiment. The duration of each experiment was approximately 3 hr.

The main series of experiments consisted of four parts: (1) Respiratory evaporative water loss was determined, using a ventilated mask technique, at air temperatures of 16 and 31°C. Two measurements were made at each environmental temperature, approximately 1·5 and 2·5 hours after the animal entered the climatic room. (2) Cutaneous water loss from the haired dorsal surface of the animal was determined by a ventilated sweat capsule at air temperatures of 15 and 30°C. The sweat capsule was held in place by an elastic belt around the animal's abdomen. (3) Cutaneous water loss from the bare skin on the dorsal surface of the flippers was measured by a ventilated sweat capsule, at air temperatures of 15 and 30°C. (4) The total evaporative water loss from the animals was measured at air temperatures of 13, 16, 21, 26 and 30°C, in a special chamber placed inside the climatic room. In the hot environment (30°C) the experiment often had to be terminated before the 3-hr period had expired because the animal urinated (see Whittow *et al.*, 1971, 1972), thereby wetting part of its skin surface and invalidating the measurements of water loss.

In the first three procedures described above, the animals were restrained by a lightweight harness and they were confined within a special cage, as described previously (Whittow *et al.*, 1972). In the fourth type of experiment, the sea lions were unrestrained within the confines of the chamber which was used to measure total evaporative water loss. Rectal temperature of the animals was measured in all experiments. In the second, third and fourth parts of the experiment, measurements were made at intervals of 15 min.

The relative humidity of the air in the climatic room could not be controlled; it varied between 39 and 73 per cent in different experiments, the higher humidities being associated with the lower air temperatures. However, during any one experiment, the relative humidity was constant within 1–3 per cent.

The ventilated mask used to measure respiratory evaporative water loss was constructed from a soft plastic cup which covered the muzzle of the animal, and it was retained in position by an elastic harness fashioned from a diver's Neoprene "wet" suit, which fitted over the head of the sea lion. The mask was equipped with side-tubes through which air was drawn, at a flow rate of 79·6–126·4 l/min, by means of a vacuum pump, the flow rate being measured by a gas meter. With these flow rates, the relative humidity of the effluent air from the mask was kept below 72%, and condensation of moisture in the tubing was avoided. The relative humidity of the air entering and leaving the mask was measured by narrow-range humidity sensors ("Hygrosensors", Hygrodynamics, Inc.) connected to a Beckman "Dynograph" recorder. Because of the discontinuous pattern of respiration in sea lions (Lin *et al.*, 1972), each breath was detected as an abrupt change in relative humidity. Due to the accumulation of saliva within the mask, the water content of the air that the animal inspired tended to be higher than that of the air entering the mask. The relative humidity of the effluent air from the mask during the period of apnea preceding each breath was, therefore, taken to be the humidity of the air that the animal breathed. The mask was kept on the sea lion for 1–4 min so that the respiratory water loss was averaged over 3–17 breaths. The changes in relative humidity were averaged by means of planimetry.

The rate of evaporation of moisture from the haired skin on the dorsal surface of the animal was measured by a Plexiglas ventilated sweat capsule similar in design to that described by McLean (1963). The area of skin enclosed by the cup was 37·3 cm^2. Air was drawn through the capsule at a flow rate of 750 ml/min by means of a vacuum pump. The flow rate was regulated by screw clamps on the tubing between the sweat capsule and pump. The air flow rate was measured by Gilmont flow meters (No. 2); recalibration of the flowmeters indicated agreement with the manufacturer's calibration, within 3 per cent. The humidity of the air entering and leaving the sweat capsule was detected by hygrosensors similar to those used to measure respiratory water loss (see above). The humidity of the effluent air was approximately 9% higher than that of the air entering the sweat capsule. A calibrated thermistor placed near the hygrosensor provided a measure of the air temperature in the vicinity of the hygrosensor, and this information permitted the calculation of the relative humidity of the air. From the flow rate of air through the sweat capsule and the difference in relative humidity between the air entering and leaving the capsule, the rate of water loss could be computed. The calculations are given in detail elsewhere (Matsuura, 1972). Calibration of the ventilated sweat capsule system was accomplished by evaporating a known weight of water over a measured time period. The sweat capsule and sensors underestimated the water loss by 3·0–3·9 per cent.

The technique for measuring the cutaneous water loss from the bare skin of the flippers was similar to that described above, for the haired skin. However, a smaller sweat cup (surface area, 7·3 cm^2) was used and it was mounted on the end of a rod so that it could be hand-held on the skin surface until a constant reading was obtained. The rim of the sweat cup was covered with soft Neoprene, ensuring a good contact with the skin.

The total evaporative water loss from the animals was measured by enclosing the unrestrained sea lion or seal in a chamber through which air was drawn at a rate of 153–162 l/min. This chamber was also used to measure the oxygen uptake of the sea lions and both the chamber and the data for oxygen uptake are described elsewhere (Matsuura & Whittow, 1973). The relative humidity of the effluent air was measured with a hygrosensor sited in a by-pass arrangement so that air flow through the chamber could be continued while the sensors were changed. The humidity of the effluent air was compared with that of the air entering the chamber. The principle of the measurement of total body water loss was similar to that described above for the measurement of cutaneous water loss. The system was calibrated by evaporating known weights of water within the chamber. The chamber and sensors underestimated the actual water loss by an average value of 11·9 ± 3·8 (S.E.) per cent.

In the first three procedures described above, the rectal temperatures were measured by a thermistor probe (Yellow Springs Instrument Co., No. 401) inserted into the rectum to a depth of 15 cm. In the experiments on unrestrained sea lions, rectal temperatures were measured by a telemetry capsule (Fox *et al.*, 1962) which was attached to the end of a short, flexible, insulated wire. The wire facilitated the insertion of the capsule into the rectum. The free end of the wire was lightly taped to the animal's tail. The advantage of this method was that it allowed the animal freedom of movement and, at the same time, the capsule could be easily and quickly retrieved at the end of the experiment.

Respiratory frequency was determined either by observing the flank movements of the animal or from records of respiratory heat loss.

The statistical significance of the results was determined by Student's *t*-test or analysis of variance (Snedecor & Cochran, 1967).

RESULTS

California Sea Lions

Cutaneous water loss. Figure 1 shows the rate of moisture loss from the haired dorsal surface and bare areas of the flippers of three sea lions at an air temperature of 15°C. In general, the rate of evaporation of moisture from all areas was low and there was no consistent trend with time. Occasional high readings from the flippers appeared to be associated with movement, which is known to result in an increase in the skin temperature of the flippers (Whittow *et al.*, 1972).

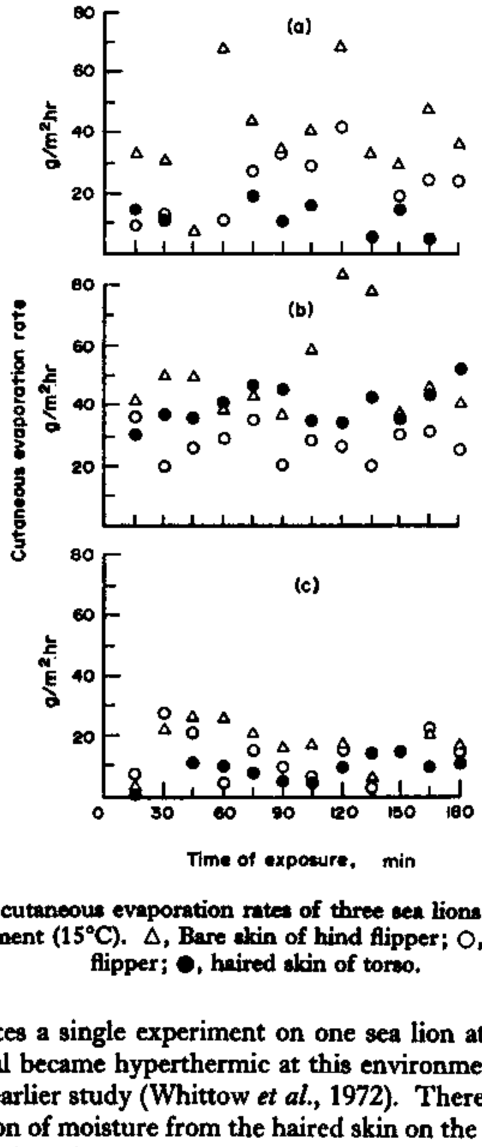

FIG. 1. Regional cutaneous evaporation rates of three sea lions (a, b, c) exposed to a cool environment (15°C). △, Bare skin of hind flipper; ○, bare skin of fore flipper; ●, haired skin of torso.

Figure 2 illustrates a single experiment on one sea lion at an air temperature of 30°C. The animal became hyperthermic at this environmental temperature, as was observed in an earlier study (Whittow *et al.*, 1972). There was little change in the rate of evaporation of moisture from the haired skin on the dorsal surface of the torso of the animal, but the evaporation rate from the flippers increased significantly, as the animal became hyperthermic, and beads of sweat were observed on the skin surface. The sweat droplets appeared in clusters on the skin of the flippers and their appearance was periodic, which may explain the variations in evaporation rates apparent in Fig. 2. The appearance of sweat droplets was not observed below rectal temperatures of approximately 38°C. The highest evaporation rate recorded from the flippers was 152 g/m² per hr. The other two animals presented a similar pattern of response to that shown in Fig. 2.

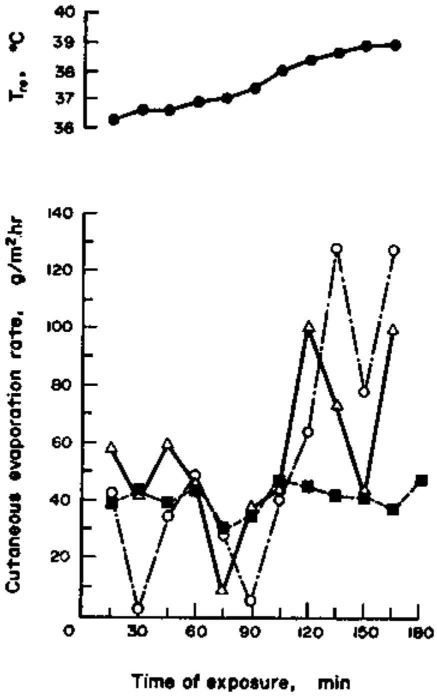

FIG. 2. Regional cutaneous evaporation rates and rectal temperatures (T_{re}) of a sea lion (b) exposed to an air temperature of 30°C. △, Bare skin of hind flipper; ○, bare skin of fore flipper; ■, haired skin of torso.

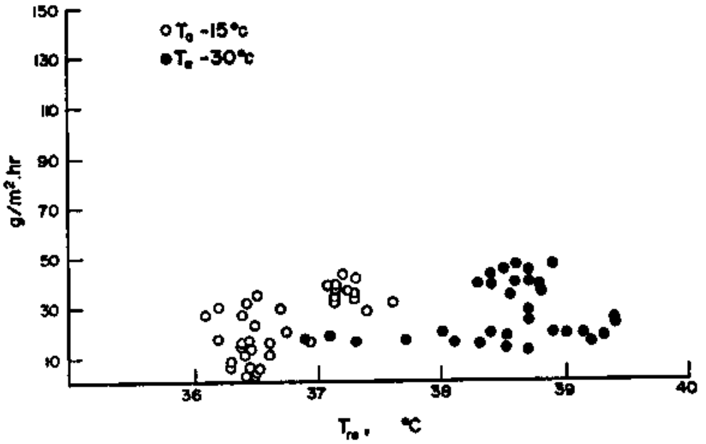

FIG. 3. Cutaneous evaporation rate from the haired skin of the torso as a function of rectal temperature (T_{re}), at the ambient temperatures (T_a) indicated. Data from three sea lions.

In Figs. 3 and 4, the cutaneous evaporation rates from the haired skin and the flippers, respectively, are plotted against rectal temperature. The two figures reveal that, in contrast to the cutaneous evaporation rate from the haired skin of the torso, the rate of moisture loss from the flippers increased as the sea lions became hyperthermic. There did not appear to be any consistent difference between the fore and hind flippers in this respect.

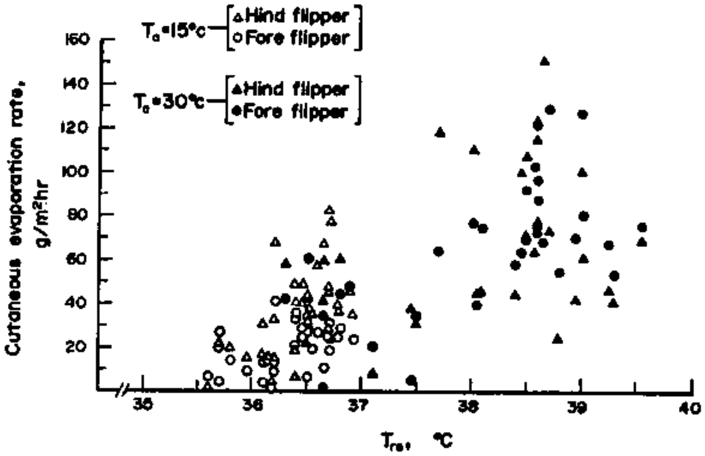

FIG. 4. Cutaneous evaporation rate from the hairless areas of the flippers, as a function of rectal temperature (T_{re}), at the ambient temperatures (T_a) indicated. Data from three sea lions.

Respiratory water loss. Table 1 and Fig. 5 show that, in confirmation of a previous study (Whittow *et al.*, 1972), there was little or no increase in respiratory frequency during exposure to a warm environment (31°C), in which the animals became hyperthermic. Although the respiratory water loss increased at higher ambient and body temperatures, the increases were small and not statistically significant. At an air temperature of 16°C, the measured respiratory evaporative

TABLE 1—RECTAL TEMPERATURE, RESPIRATORY FREQUENCY AND RESPIRATORY WATER LOSS AT AIR TEMPERATURES OF 16°C AND 31°C

N	Ambient temperature (°C)	Rectal temperature (°C)	Respiratory frequency (resp/min)	Respiratory water loss (mg/min per kg)
6	16·3 ± 0·3	36·82 ± 0·25	3·3 ± 0·5	1·277 ± 0·286
3	30·8 ± 0·1	38·48 ± 0·12	3·9 ± 1·0	1·512 ± 0·203
3	30·8 ± 0·1	39·78 ± 0·11	4·9 ± 1·7	2·562 ± 0·776

Values indicate mean ± S.E. Data from three sea lions. *N* = number of experiments.

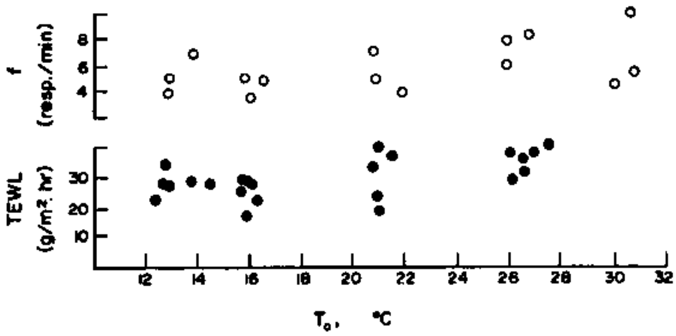

FIG. 5. Total evaporative water loss (TEWL) and respiratory frequency (f) at ambient temperatures (T_a) between 12 and 31°C for three sea lions. Each point represents a single measurement.

water loss was only approximately 40 per cent of the theoretical maximum, based on data for respiratory minute volume (Matsuura & Whittow, unpublished data) and the assumption that air is expired, saturated with water vapor, at deep-body temperature. At an ambient temperature of 31°C, the measured water loss was 58 per cent of the maximal possible value.

Total evaporative water loss. Figure 5 presents data for the measured total evaporative water loss from the three sea lions at air temperatures between 13 and 27°C. The increase in total evaporative water loss, with increasing air temperature over this range, was small.

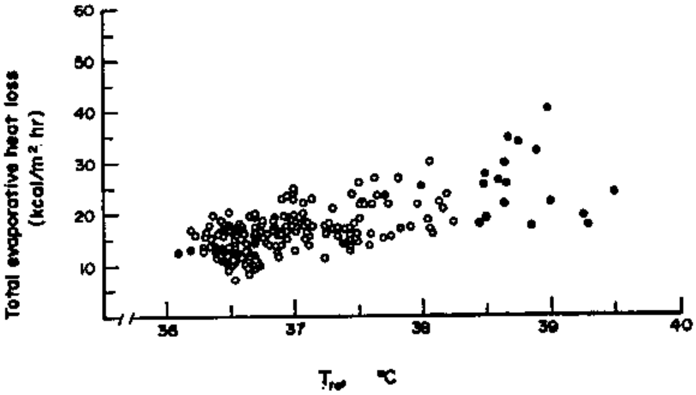

FIG. 6. Comparison between direct measurements of total evaporative heat loss (○), and estimates derived from separate sweat capsule measurements and determinations of respiratory evaporative heat loss (●). T_{re}, rectal temperature.

Figure 6 displays the measured total evaporative water loss, expressed as $kcal/m^2$ per hr, and plotted against rectal temperature. Also included in Fig. 6 are estimates of the total evaporative heat loss derived from the measured respiratory evaporative heat loss and cutaneous evaporative heat loss, the latter determined by the ventilated sweat cup. The estimates of cutaneous evaporative heat loss included appropriate weightings for the area of the flippers and torso (Matsuura, 1972). It was assumed that the rate of evaporation from the entire torso was similar to that measured on the dorsal surface of the animal. Rates of water loss were converted to heat loss units assuming that the heat of vaporization of water is 0·58 kcal/g. The agreement between the measured and derived values was good, justifying the assumptions made in the derivations. Unfortunately, a comparison between the measured and estimated values could not be made at rectal temperatures higher than approximately 38·5°C, because the animals urinated and the direct measurements were no longer valid. If the estimated values accurately reflect the total evaporative heat loss, then Fig. 6 shows that the total evaporative heat loss increased approximately twofold as the sea lions became hyperthermic.

Partition of evaporative heat loss. Information on the heat production of the sea lions, reported elsewhere (Matsuura & Whittow, 1973), together with the data obtained in the present investigation, permit the respiratory and cutaneous evaporative heat loss to be expressed as a percentage of the amount of heat produced by the animals. It may be seen from Fig. 7 that, in a cool environment (15°C), the respiratory and cutaneous components of evaporative loss represented 3·1 and 9·9 per cent, respectively, of the heat production of the sea lions. In a warm environment (30°C), in which the sea lions became hyperthermic (T_{re} = 39°C), approximately 2·5 per cent of the heat production was lost from the respiratory

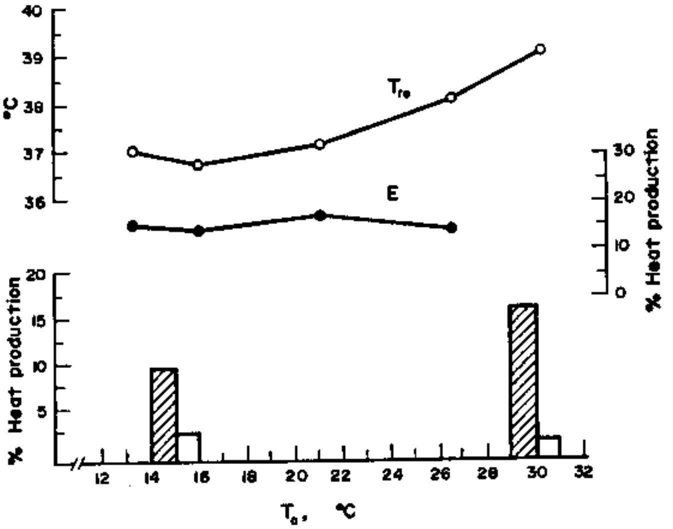

FIG. 7. Total evaporative heat loss (E), cutaneous evaporative heat loss (■) and respiratory evaporative heat loss (□), expressed as a percentage of heat production at different air temperatures (T_a). Rectal temperatures (T_{re}), obtained by telemetry, are also shown. Data from three California Sea Lions. Each point represents the mean of six measurements.

tract and 16·0 per cent by cutaneous evaporation. Consequently, the total evaporative heat loss of the hyperthermic animals represented only approximately 19 per cent of the heat that they produced.

Young animal. Only measurements of total evaporative heat loss were made on the young animal. At air temperatures of 15, 20, 26 and 31°C, the total evaporative heat loss was 22·8, 18·5, 16·4 and 29·5 per cent respectively, of the heat production. At an air temperature of 31°C, the animal salivated a great deal and evaporation of saliva from the skin on the throat may have contributed to the value of 29·5 per cent.

Harbor Seal

The data obtained from the Harbor Seal are shown in Fig. 8. The Harbor Seal became hyperthermic only at an air temperature of 35°C. The respiratory frequency at this air temperature was actually less than at lower air temperatures.

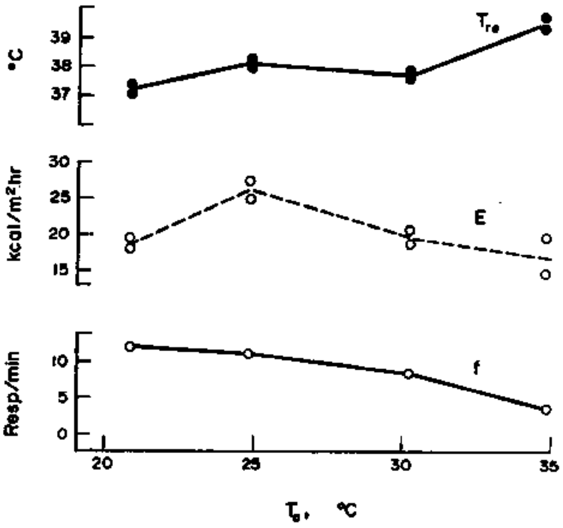

FIG. 8. Rectal temperature (T_{re}), total evaporative heat loss (E) and respiratory frequency (f) in a Harbor Seal at different air temperatures (T_a). Each point represents a single measurement.

Total evaporative water loss did not increase either, when the seal became hyperthermic. In a separate series of experiments, the cutaneous evaporative water loss was measured from the dorsal surface of the animal, using the ventilated sweat capsule. The evaporative water loss at air temperatures of 17, 28 and 36°C was 13·67, 17·31 and 42·73 g/m² per hr, respectively.

DISCUSSION

The magnitude of evaporative water loss from the haired dorsal surface of the torso of the sea lions is not compatible with active secretion of sweat by the sweat glands (Ingram, 1964). This conclusion conforms with the failure to detect sweat gland activity by means of the starch-iodide technique in an earlier study (Whittow *et al.*, 1972). Although only a single area on the dorsal surface of the trunk was examined in the present study, it is unlikely that, if active sweating occurs on other regions of the torso, it can be of any great physiological significance, because of the relatively low values for total evaporative water loss obtained in the present experiments. It would be of great interest to know if thermal sweating occurs on the haired surface of other pinnipeds. The data obtained on the single Harbor Seal used in the present study suggests that it does not, but clearly further investigations are required on other species.

Evidence for active sweating on the flippers in response to heat is of interest, because terrestrial members of the Carnivora, to which order pinnipeds also belong, do not sweat on the haired surface of the trunk but they do sweat on the foot pads during exposure to heat (see Adams, 1971). The pattern of sweating in sea lions may, therefore, be characteristic of the Carnivora, rather than representing a special feature of sea lions. Measurements of evaporative water loss from the flippers of the Harbor Seal were not made. However, the small size and hairy integument of the flippers, in most seals, lead to the conclusion that sweating on the flippers is of functional significance only in sea lions, and possibly in fur seals, which also have large flippers (Bartholomew & Wilke, 1956). Preliminary histological studies in the California Sea Lion indicate much larger, and possibly more numerous, sweat glands in the flippers than in the skin on the trunk (Jenkinson, Szekerczes and Whittow, unpublished data). The functional differences in sweat gland activity in different regions appear, therefore, to be reflected in structural differences in the skin.

The absence of thermal polypnea in sea lions, although confirming earlier reports (Whittow *et al.*, 1970, 1971, 1972) is decidedly not characteristic of the terrestrial Carnivora, which include some of the most proficient panting mammals (Adams, 1971). Sea lions breathe through their mouths when they are hyperthermic, and it was postulated in a previous report (Whittow *et al.*, 1972) that they might accomplish an increased evaporative heat loss in this way, by virtue of a diminished cooling of expired air (Schmidt-Nielsen *et al.*, 1970). This did occur to a limited extent but, nevertheless, the total respiratory evaporative heat loss was extremely small. Although respiratory evaporative water loss was not measured directly in the Harbor Seal, the absence of thermal polypnea and the low values for total evaporative water loss suggest that in this species also respiratory evaporative heat loss is of little significance physiologically. The significance of reports in the literature that Northern Fur Seals (Bartholomew & Wilke, 1956) and Harbor Seals (Harrison & Kooyman, 1968) "pant" may hinge on the definition of "panting" (Whittow, 1966). If open-mouth breathing is construed as panting, then sea lions do this also, but in terms of heat loss it is relatively ineffective.

The total evaporative water loss from the sea lions in a warm environment could account for the dissipation of less than 20 per cent of the heat that the animals were producing. In contrast, many terrestrial mammals and birds are able to lose heat equivalent to their entire heat production by evaporation of moisture (Dawson & Hudson, 1970; Hart, 1971; Dawson, 1973). If the minor sweating responses of pinnipeds are the legacy of their carnivore ancestry (see above), then the ineffectiveness of evaporative cooling mechanisms largely represents the absence of panting or of saliva spreading, in pinnipeds as opposed to terrestrial carnivores. Sea lions regularly urinate during exposure to heat and the evaporation of urine from the skin must result in some heat loss (Gentry, 1973) but this is not sufficient to prevent hyperthermia (Whittow *et al.*, 1971). The absence of effective evaporative cooling mechanisms in sea lions has been discussed elsewhere (Whittow *et al.*, 1972; Whittow, 1973). Teleologically, sea lions may attempt to conserve water rather than to maintain a constant body temperature. In the course of their adaptation to the sea, dehydration may have had a role in suppressing the evaporative cooling mechanisms that ancestral pinnipeds may have possessed.

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Key Word Index—California Sea Lions; Harbor Seal; evaporative heat loss; sweating; effects of heat; temperature regulation; respiratory evaporative heat loss; *Zalophus californianus*; *Phoca vitulina*.

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BODY TEMPERATURE OF THREE SPECIES OF WHALES

The available information on the body temperature of whales has been tabulated by Morrison (1962), Ridgway (1972), and Irving (1973). The data were obtained from either dead whales or animals subjected to some degree of restraint necessary for the insertion of a rectal probe. Nothing is known about the body temperature of free-swimming whales. The present investigation was prompted by the opportunity to obtain data from three species of odontocete whales, and by the availability of a technique for measuring the body temperature, in the unrestrained animal, by telemetry. The study was performed on animals in tropical waters, and interest in it was enhanced by the possibility that whales, with a high level of heat production and a thick layer of insulating blubber (Irving, 1973), might encounter problems of heat dissipation in warm water, especially during activity.

Measurements were made on a male pilot whale (*Globicephala scammoni*), "Morgan," approximately 4.1 meters (m) long and weighing 637 kilograms (kg), and on a male killer whale (*Orcinus orca*), "Ahab," approximately 5.3 m long, with a body weight of 2727 kg. The two animals were kept in separate sea water pens at the Naval Undersea Center, Kailua, Oahu, Hawaii. The pens were fenced-off enclosures of the sea, 30.5 m by 30.5 m, with an average depth of 6.1 m.

The third animal studied was a male, false killer whale (*Pseudorca crassidens*), "Ola," 3.25 m long and weighing approximately 300 kg. Ola was kept in a circular concrete sea-water tank at Sea Life Park, Makapuu, Oahu, Hawaii. The diameter of the tank was 12.2 m and its maximal depth was 3.1 m.

Each animal was fed a fish containing a "radio-pill" (Fox *et al.*, 1962). The radio pill was a small plexiglass capsule, approximately 2.5 centimeters long, containing a temperature-sensitive transmitter. The signal from the transmitter was detected by an antenna placed in the water, and a receiver kept at the side of the tank. The sensitivity of the system was such that a change of temperature of 0.1° C could be detected. Measurements of body temperature were made at intervals of 30 minutes until the pill was passed. Respiratory frequency was counted over a 5 minute period, every 30 minutes. The temperature of the sea water was measured by means of a Yellow Springs Instrument Company (YSI) thermistor probe (no. 409) connected to a YSI telethermometer (no. TUC 46). Special efforts were made to ensure that the animals followed their normal daily pattern of behavior and a careful record of the behavior was kept.

The most complete data were obtained from the false killer whale, and these are shown in Fig. 1. The body temperature was measured continuously for 56 hours. The initial increase in body temperature, after the pill was fed to the animal, represents the warming of the pill, and the fish containing it, to body temperature. The highest temperatures recorded from the animal (a, c, and e, Fig. 1) coincided with periods when the whale was swimming actively and leaping out of water. Conversely, the lowest body temperatures occurred when the whale was resting quietly and appeared to be asleep (b, d, Fig. 1). A diminution in the temperature transmitted by the pill occurred each time that the animal was fed. It is believed that these decreases represent cooling of the pill by the ingested food. The decrease in temperature during the last feeding session is especially interesting. The pill must have left the stomach by that time because, shortly afterwards, it was ejected. Conceivably, the pill was in a segment of the large intestine adjacent to the stomach. The possibility that the pill remained in the stomach and that it was regurgitated cannot be completely excluded, but the animal was not seen to regurgitate at any time. The respiratory frequency of the whale varied from less than one breath per minute to seven breaths per minute. In general, it was lowest while the animal appeared to be sleeping.

The data from the pilot whale were similar to those from the false killer whale, both with regard to the range of body temperature (Table 1) and its variation with activity. The pilot whale ejected the radio pill twenty-two hours after it had been fed to the animal.

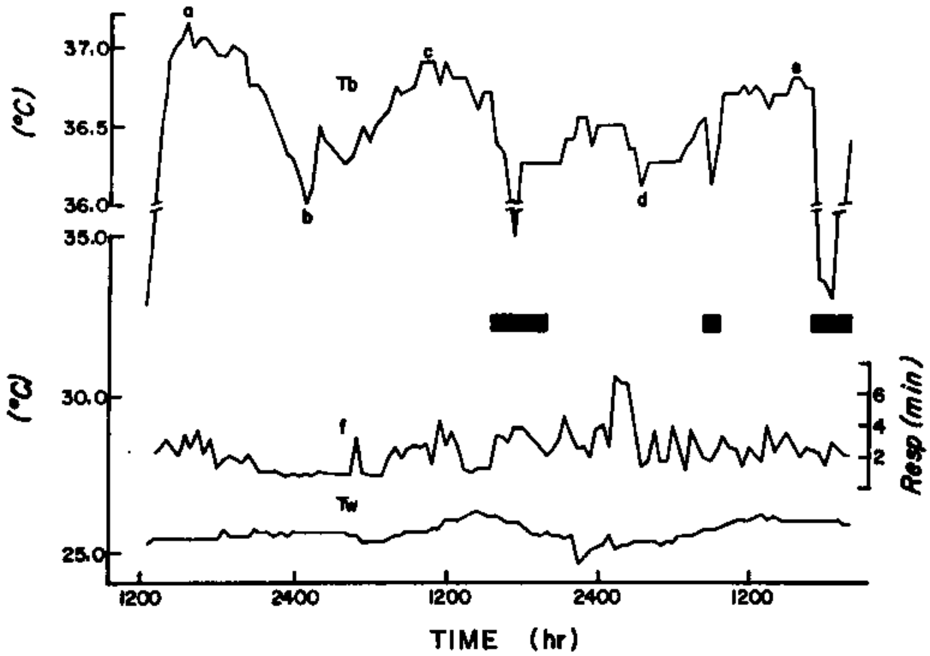

FIG. 1.—Deep-body temperature (T_b), respiratory frequency (f), and water temperature (T_w) for a male false killer whale, "Ola," over a period of 56 hours. The horizontal bars indicate the feeding periods. The small letters (a-e) are identified in the text.

Radio pills were fed to the killer whale on two separate occasions. The first trial indicated that the body temperature of the killer whale was higher than that of either the pilot whale or the false killer whale (Table 1). The temperature increased when the whale swam and diminished when it was inactive. The body temperature of the whale was measured continuously for 25.5 hours, when recordings were discontinued. On the second occasion, six months later, the body temperature was higher but so also was the water temperature (Table 1). The animal was very inactive, but, in spite of this, there was a detectable variation of body temperature with the temperature increasing during the day and decreasing at night. The pill was ejected 60 hours after it had been fed to the animal.

It seems reasonable to conclude from these experiments that the body temperature of odontocete whales, up to 637 kg in body weight, is within the range of 36.0 to 37.2° C. This is similar to the body temperature of a large atlantic bottlenose dolphin (*Tursiops truncatus*) tested under similar circumstances (Hampton *et al.*, 1971). Ridgway (1972) determined that the rectal temperature of a pilot whale was 36.5° C. The present data appear to be the first recorded measurements of the deep-body temperature of a false killer whale. It is conceivable that the body temperature of the animals would be higher under open-ocean conditions, in which higher swimming speeds might be attained. However, this does not necessarily follow because, in the ocean, the whales would probably dive to greater depths and for longer periods. In so doing, they might encounter water at a lower temperature and also experience a reduction in metabolic rate during the dive (Irving, 1973). Both factors might conspire to lower their body temperature. A reasonable speculation, therefore, might be that in the open ocean, the range of body temperature would be greater, but that the average temperature might be similar to the values obtained in the present investigation.

TABLE 1.—*Body temperature (T_b) and respiratory frequency (f) of three whales. The number of measurements is in parentheses. T_w = water temperature. Excluded from the data are periods in which the temperature of the radio pill was directly influenced by the presence of food in the gut.*

Species	T_b (°C)		f (respirations/minute)		T_w (°C)
	Mean	Range	Mean	Range	Range
False Killer whale (*Pseudorca crassidens*)	36.6 (83)	36.0–37.2	2.3 (84)	0.8–7.1	24.6–26.3
Pilot whale (*Globicephala scammoni*)	36.9 (30)	36.4–37.2	2.1 (31)	0.9–5.9	26.2–26.9
Killer whale (*Orcinus orca*)	37.5 (48)	37.1–38.0	1.3 (38)	0 –3.0*	24.0–24.7
	38.0 (96)	37.5–38.5	1.6 (103)	0.8–3.7	26.8–27.5

* Measured over a one-minute period; 0 indicates spontaneous breath-holding.

The higher body temperatures of the killer whale are more difficult to interpret. They were certainly not due to a greater level of activity on the part of the killer whale; it was, in fact, less active than either the pilot whale or the false killer whale. The higher body temperature of the killer whale on the second occasion that it was measured could have been related to the higher temperature of the sea water (Table 1). This suggests that the whale, because of its large size, and, consequently, its relatively small surface area to body mass ratio, was experiencing difficulty in dissipating heat to the environment. Ridgway (1972) reported that the rectal temperature of a killer whale in the cooler waters off the California coast was 36.4° C. However, the conclusion that killer whales have a higher body temperature in tropical waters, because of the higher temperature of the water, *per se*, has to be tempered by the fact that the killer whale studied in the present investigation was probably not a healthy specimen, particularly at the time of the second trial. In fact, the animal died approximately one week after the second series of measurements. An autopsy conducted by one of us (Allen) revealed that the cause of death was disseminated nocardiosis. Therefore, the body temperatures recorded on the second occasion may represent a fever. Little is known about the dimensions of fever in mammals such as cetaceans, that are continuously in the water. However, it is particularly interesting that Ridgway (1972) reported body temperatures of 38 to 39° C in cetaceans with manifest symptoms of infectious disease. In the febrile state, terrestrial mammals regulate their deep-body temperature at a higher level, but with a similar circadian range of temperature, than in the absence of fever (MacPherson, 1959). Inspection of Table 1 reveals a similar situation in the killer whale. It is also interesting that, if the killer whale was indeed displaying a fever, this was not associated with a notably increased respiratory frequency, indicating that respiratory frequency cannot be used as an easily measured correlate of fever in whales.

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TEMPERATURE REGULATION IN THE CALIFORNIA SEA LION (ZALOPHUS CALIFORNIANUS)[1]

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INTRODUCTION

Under natural conditions, California Sea Lions are found as far south as Mexico, with a subspecies on the Galapagos Islands (Scheffer 1958). When they are on land, they may therefore encounter high air temperatures and high levels of solar radiation. It is uncertain whether they respond to these conditions by behavioral thermoregulation, as the field observations of Peterson and Bartholomew (1967) would suggest, or by physiological means. The experiments reported in this paper were designed to investigate the latter possibility—that these animals are physiologically equipped to deal with a hot environment, as some other species of pinnipeds appear to be (Bartholomew and Wilke 1956).

MATERIAL AND METHODS

Experiments were performed on three California sea lions approximately 1 year old, their body weight ranging from 38 to 45 kg. The animals were kept in pens out of doors and in the shade. Each pen was provided with a tub (6 ft long × 3 ft wide × 1 ft deep) of fresh running water, and each sea lion was fed 8% of its body weight a day, of frozen smelt, supplemented with vitamin and mineral capsules. As the animals were always fed after an experiment and at the end of the day, they had fasted for approximately 16 hr before the experiment began.

Each animal was exposed, on a separate occasion, to each of four different air temperatures, approximately 36, 30, 18, and 10 C, in a temperature-controlled room. The relative humidity of the air could not be controlled, and in different experiments it varied within the range 46%–58%. However, in any one experiment, the humidity did not change by more than ±2%. The air movement was approximately 50 ft/minute. Throughout the exposure, which lasted for at least 3 hr, measurements were made at 20-min intervals of rectal temperature, skin temperature, hair surface temperature, heart rate, and respiratory rate. In addition, measurements of heat flow from the skin and tests for sweating were made after the animals had been in the climatic room for approximately 2½ hr. Skin temperature measurements were made on a small area of shaved skin on the back, approximately 35 cm rostral to the tail, on an area of bare skin on one of the front flippers and on a similar area on one of the hind flippers. Measurements of heat flow and tests for the presence of sweating were also made on the area of shaved skin on the back of the animal. The data for heat flow (H) were used in conjunction with the values for rectal temperature (Tr) and skin tempera-

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ture (Ts) to compute the tissue insulation (I), in degrees Celsius, square meter, hour, per kilocalorie of heat flow, in accordance with the following relationship:

$$I = \frac{(Tr - Ts)\ ^\circ\mathrm{C}}{H\ (\mathrm{kcal/m^2\,hr})}.$$

The "thermal circulation index" (TCI) for the front flipper was computed as follows:

$$TCI = \frac{Ts - Ta}{Tr - Ts},$$

where Tr, Ts, and Ta are the rectal temperature, skin temperature of the flipper, and air temperature, respectively (Burton and Edholm 1955). Hair surface temperatures were made contralaterally to the shaved area of skin on the back.

During the experiments, the animals wore a lightweight harness which was tethered to the sides of a specially designed pen. Although the animals could make small postural movements, they could not turn completely around within the pen. The head and neck of the animal were free. If the animal was wet before the experiment, sufficient time was allowed for it to dry before it entered the temperature-controlled room.

Measurements of rectal temperature were made by a Yellow Springs Instrument (YSI) thermistor probe (no. 401) inserted into the rectum to a depth of 12 cm. Skin and hair surface temperatures were measured by a YSI surface probe (no. 402). The probes were connected to a YSI Telethermometer (no. 46). Respiratory rates and heart rates were recorded by needle electrodes connected to a Physiograph impedance pneumograph and Hi-Gain preamplifier. Heat-flow determinations were made with a Hatfield heat-flow disk (Hatfield 1950) connected to a Turner microvoltmeter. Two methods were used to detect the presence of sweating: the starch iodide technique (Randall 1946) and by the application of quinizarin 2-6-disulfonic acid to the skin (Guttman 1942). Surface areas were estimated from linear measurements made on the animal; the trunk was treated as two cones and the flippers as triangles. The results of the experiments were examined statistically by the student t-test for two small samples (Bailey 1959).

RESULTS

FIELD OBSERVATIONS

When the animals were exposed to direct sunlight on land, they remained reasonably quiet until their coats had dried. Following this, their activity increased and small areas of shade or moisture were sought. Shallow burrows were excavated in the sand and the animals frequently lay on their backs and waved their front flippers. Each animal urinated under these conditions, and it was able, in this way, to keep the ventral surface of its abdomen and thorax moist. The respiratory rate did not appear to increase during exposure to solar radiation, but the three sea lions adopted an open-mouthed, gasping type of breathing when they were clearly heat stressed.

LABORATORY STUDIES

Rectal temperature.—At air temperatures of approximately 36 and 30 C, the animals were unable to achieve thermal equilibrium and their rectal temperatures increased throughout the exposure (fig. 1). At the higher of these two environmental temperatures (36 C), the animals were in fact removed

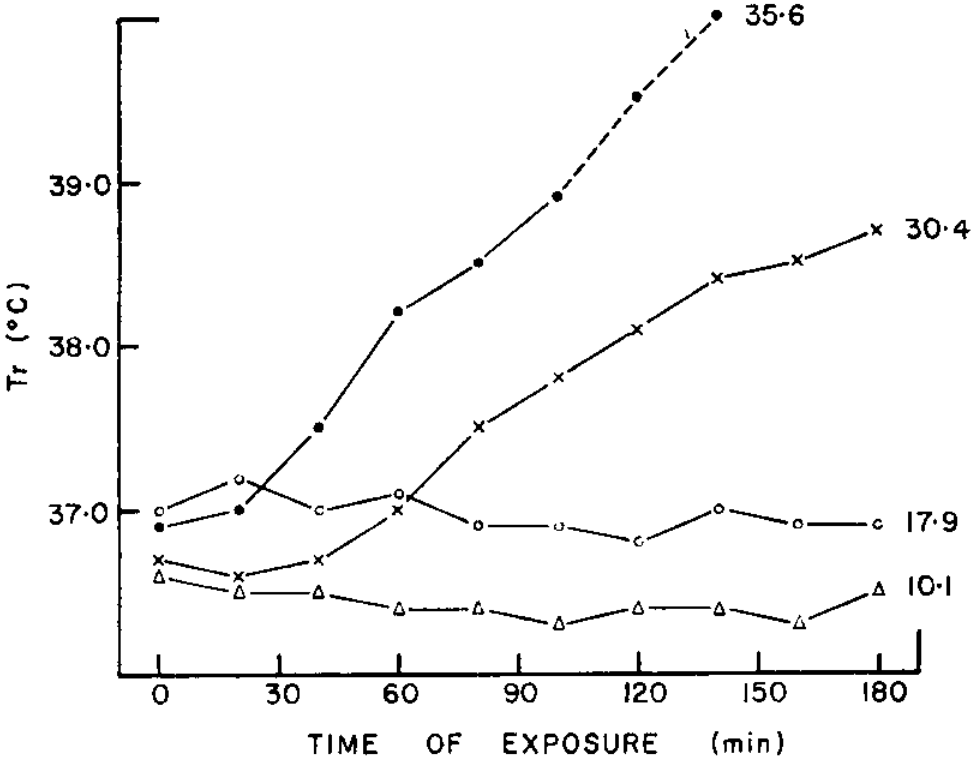

FIG. 1.—Rectal temperatures (*Tr*) of three California sea lions during exposure to the air temperatures indicated to the right of the figure. Each point represents the mean value for the three animals, with the exception of the points connected by a broken line, which include data for two animals only.

from the climatic room when their rectal temperatures had reached 40 C, after 100–140 min. At air temperatures of 18 and 10 C, rectal temperatures were well maintained.

Skin temperatures and tissue insulation.—The skin temperature on the back of the animal increased in the two hotter environments (36 and 30 C). At air temperatures of 18 and 10 C, the skin temperatures tended to decrease with time, but periods of activity on the part of the animal resulted in increases in skin temperature (fig. 2). Comparison of the skin temperatures at the different air temperatures suggested that a decrease in the blood flow to the skin occurred between air temperatures of 18 and 10 C. Some support for this suggestion is provided by the data presented in figure 3, which shows the tissue insulation at different air temperatures. The data are rather fragmentary, but they do indicate that at air temperatures above approximately 15 C the tissue insulation is low but that at temperatures below 15 C the insulation increases rather abruptly.

Flipper skin temperatures.—The skin temperature of the flippers also increased in the two hotter environments and tended to decrease at air temperatures of 18 and 10 C (fig. 4). The behavior of the flipper skin temperature was similar to that of the trunk in that the temperature increased when the animal became active. An additional source of variation in the skin temperature of the flipper was provided

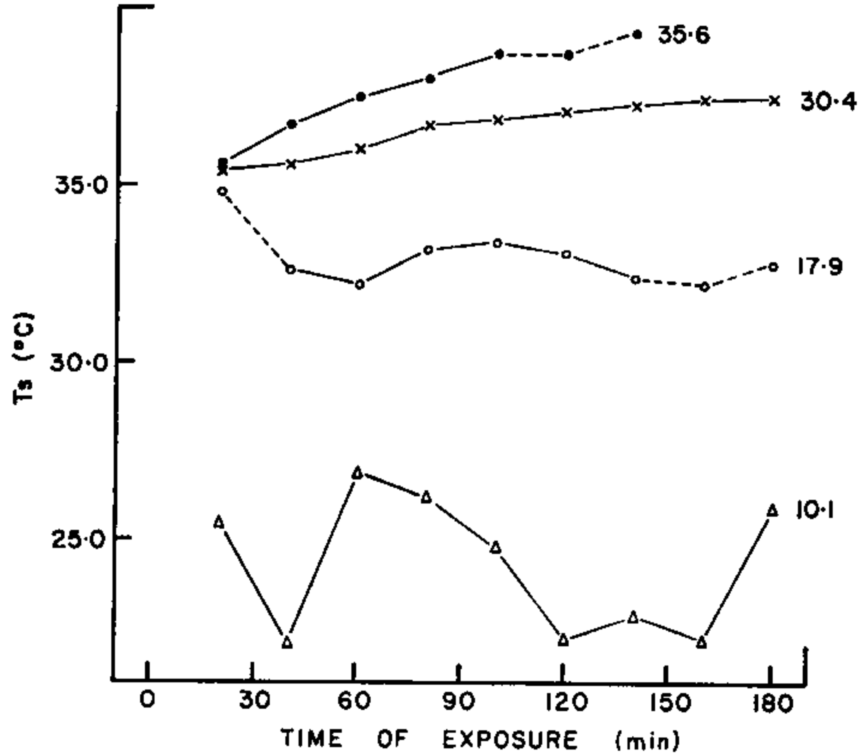

FIG. 2.—Skin temperature (*Ts*) on the back of three California sea lions during exposure to different air temperatures. Notations as in fig. 1.

by the animal tucking its flipper under its body. When this occurred, the flipper temperature increased. It was difficult to decide from the data for flipper temperatures whether there was an underlying change in blood flow at the different air temperatures. In order to provide more information on this point, the thermal circulation index (*TCI*) was computed for the front flipper after the animals had been exposed to each of the four air temperatures for 100 min. The results are shown in figure 5. The main point illustrated by figure 5 is that the *TCI* was significantly lower ($t = 89.7$; $.02 > P > .01$) at an air temperature of 18 C than at 30 C.

The surface area of the flippers represented 31% (30%–33%) of the total surface area of the animals.

Hair temperature.—In general, the surface temperature of the coat varied in a manner similar to that of the skin temperature of the shaved area on the back. At the lowest air temperatures used, however, it was observed that the temperature of the hair was higher than that of the shaved skin, which suggested that the skin temperature under the hair was even higher.

Respiratory rates.—The mean respiratory rates of the three animals at the four air temperatures are presented in figure 6. The pattern of breathing was irregular and the respiratory rates were low. There was no evidence of thermal polypnea at the higher air temperatures, but the three animals did adopt open-mouthed breathing when they were hyperthermic and they salivated profusely.

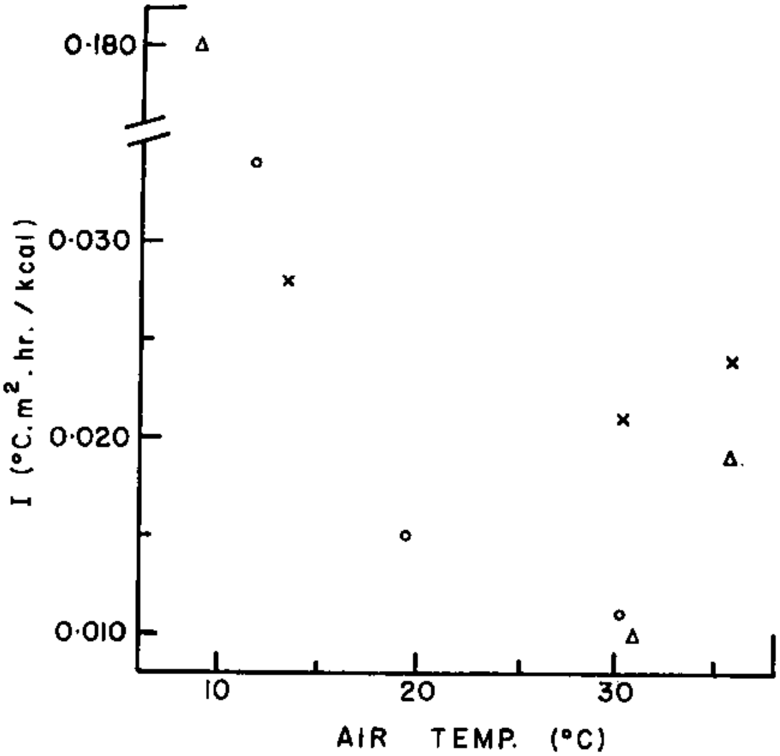

FIG. 3.—Tissue insulation (*I*) on the back of the California sea lion at different air temperatures. Each point represents a single measurement on one of the animals. The different symbols identify the three animals.

Cutaneous moisture loss.—Neither of the techniques used indicated that sweating occurred when the animals were hyperthermic.

Heart rates.—The heart rates were irregular, varying with each respiration and with the activity of the animals. These variations tended to obscure any effect of temperature on the heart rate. In the few instances in which the animals did not struggle unduly, the heart rate increased by 10–15 beats/minute when the animals became hyperthermic.

Shivering.—One animal shivered at an air temperature of 9 C, when its rectal temperature was 36.7 C, the temperature of the shaved skin was 19.9 C, and the temperature on the surface of the coat was 23.3 C.

Other responses.—When the animals became hyperthermic, at air temperatures of 30 and 36 C, they urinated at a deep body temperature of approximately 38.5 C. When the rectal temperature exceeded 39 C, the animals defecated frequently.

DISCUSSION

Under the conditions prevailing in these experiments, the sea lions were unable to attain thermal equilibrium at air temperatures of 30 or 36 C. An air temperature of 30 C is not unlikely in the Galapagos Islands or off the Mexican coast. With an added heat load from solar radiation, sea lions would not be able to endure such conditions continuously for more than a

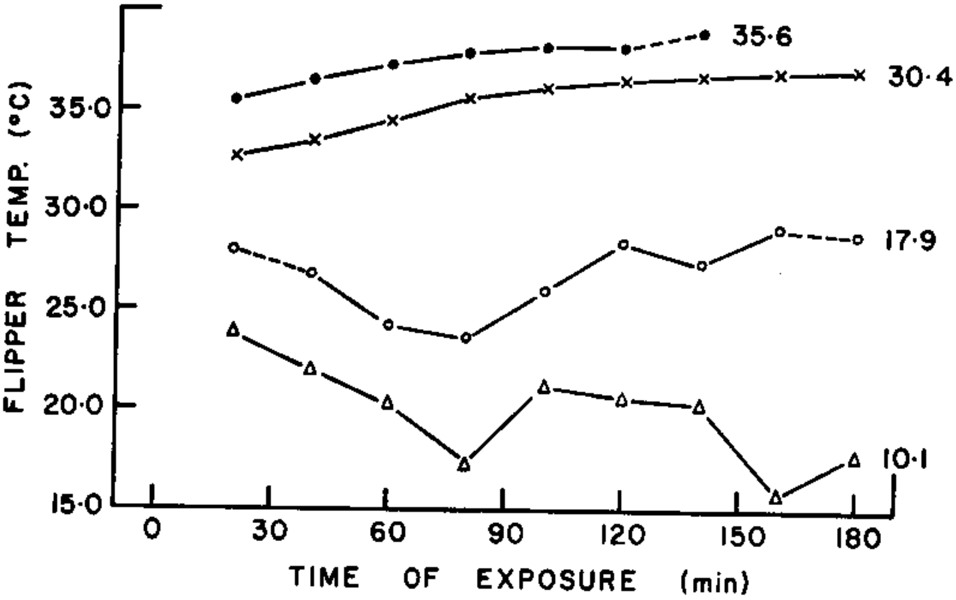

FIG. 4.—Skin temperature on the front flippers of three California sea lions at various air temperatures. Notations as in fig. 1.

few hours. This conclusion conforms with the field observations on the California sea lion made by Peterson and Bartholomew (1967).

The diminution in the skin temperature of the back and the increase in tissue insulation between air temperatures of 18 and 10 C suggest that a decrease in the blood flow to the skin occurred between these temperatures. In this respect, the sea lion differs from terrestrial haired mammals such as the ox (Whittow 1962), in which the skin temperature of the trunk decreases uniformly with environmental temperature. On the other hand, it resembles relatively hairless animals such as the pig (Ingram 1964) and another marine mammal, the walrus (Ray and Fay 1968). There is evidence of a similar phenomenon in the Weddell seal (Ray and Smith 1968) and also in the harbor seal (Hart and Irving 1959). The air temperature at which a large diminution in skin temperature occurs varies, however, in different species. This conclusion has to be qualified, in the case of the sea lions, in the light of the relatively high hair temperature at low air temperatures. This may mean that the blood flow through the skin under the coat does not diminish until air temperatures lower than those used in the present investigation are reached. It would be interesting to know whether the skin blood flow of a species such as the northern fur seal, which has a thick waterproof coat, behaves in a manner similar to that of species with a short wettable coat.

Evidence that the blood flow through the flippers diminishes between air temperatures of 30 and 18 C, that is, at a higher level of air temperature than that of the skin of the back, is to be expected. The flippers have a higher ratio of surface area to volume than does the trunk of the animal, and they should lose heat more rapidly. A thermoregulatory function for the

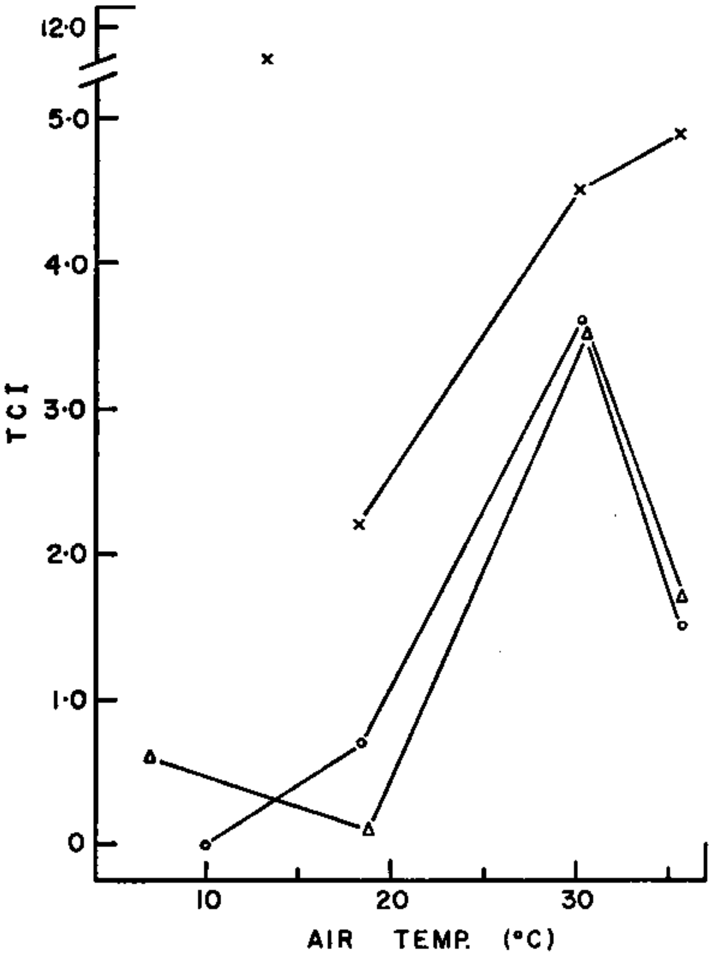

FIG. 5.—Thermal circulation index (*TCI*) of the front flippers of three California sea lions at various air temperatures. Notations as in fig. 3. The highest point (*X*) is not connected to its fellows because in this instance the flipper was tucked under the body of the animal.

flippers has been claimed in the harbor seal (Irving and Hart 1957; Hart and Irving 1959), the northern fur seal (Irving et al. 1962), the walrus (Ray and Fay 1968), and the Weddell seal (Ray and Smith 1968).

Although changes in the insulation, *TCI,* and skin temperature of the back and flippers have been interpreted in terms of variations in blood flow, they could equally well be explained by a countercurrent heat-exchange mechanism between arterial and venous blood, operating with or without a change in blood flow (Scholander and Krog 1957). There is no anatomical or histological evidence for this in the California sea lion, but the arrangement of the blood vessels in the flippers of the northern fur seal and harbor seal provides the basis for such a mechanism (Tarasoff and Fisher 1970). In view of the high proportion of the total surface area represented by the flippers, they are probably of considerable importance in the regulation of heat loss in sea lions, particularly under cold conditions.

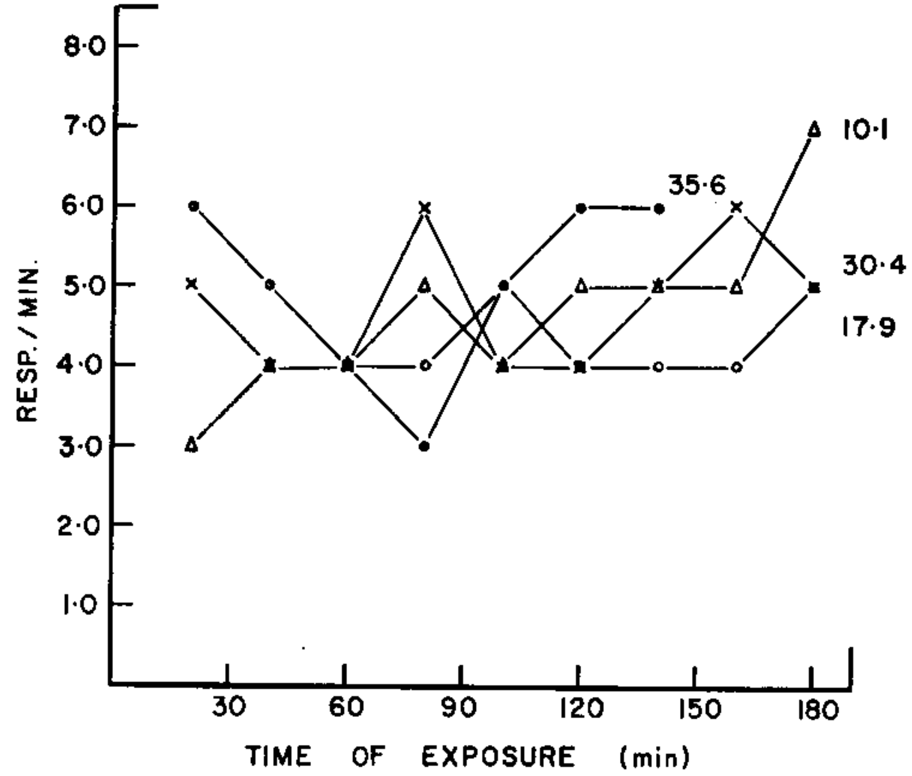

FIG. 6.—Respiratory rates of the three sea lions at different air temperatures. Notations as in fig. 1.

At an air temperature of 36 C, the tissue insulation on the trunk of the animal was higher while the *TCI* in the flipper tended to be lower, than at an air temperature of 30 C. Although the differences are not statistically significant, they do suggest that at elevated levels of body temperature the blood flow to the skin may actually decrease, possibly associated with a diminution of cardiac output (Whittow, Sturkie, and Stein 1964).

The absence of thermal polypnea in sea lions, even at a rectal temperature higher than 40 C, was surprizing in view of reports that both the northern fur seal (Bartholomew and Wilke 1956) and the harbor seal (Harrison and Kooyman 1968) pant when they are hot. On the other hand, Peterson and Bartholomew (1967) did not observe panting in the California sea lion rookery on San Nicholas Island, even in bulls engaged in vigorous fighting. Furthermore, panting did not occur in either the Weddell seal (Ray and Smith 1968) or the walrus (Ray and Fay 1968). The absence of thermal polypnea does not necessarily mean that sea lions are not able to increase their respiratory evaporative water loss during exposure to heat. The open-mouthed breathing observed during hyperthermia might result in a significantly greater moisture loss per breath, in spite of the fact that the respiratory rate did not increase. This could be accomplished by a greater tidal volume or by a diminished cooling of the expired air when the animal breathed through its mouth (Schmidt-Nielsen, Bretz, and Taylor 1970).

The failure to detect sweating during exposure to heat was unexpected in the light of reports that sweating occurs in the northern fur seal (Bartholomew and Wilke 1956), albeit from the flipper. Preliminary histological studies on the skin of the California sea lion indicate that there are sweat glands in the skin. Their apparent failure to respond to heat is analogous to the absence of thermal sweating in the pig (Ingram 1964) and the dog (Aoki and Wada 1951).

The sea lion appears to be one of the very few mammals that neither sweats nor pants. The only water available to marine mammals is the water in their food, and seawater. There is little evidence that seals drink seawater or that their kidneys are especially equipped to deal with it (Harrison and Kooyman 1968; Depocas, Hart, and Fisher 1969). It is possible therefore that in marine mammals considerations of water economy take precedence over the regulation of body temperature.

Although measurements of heat production were not made in this study, it is of interest that one animal shivered when its skin temperature was close to 20 C. Hart and Irving (1959) noted that the heat production of the harbor seal increased, at a similar skin temperature, when the animals were tested during the summer.

During this investigation, evidence was obtained that the California sea lion has a strongly developed pattern of thermoregulatory behavior, which is described in more detail in another report (Whittow, Ohata, and Matsuura 1971). This would accord with the field observations of Peterson and Bartholomew (1967), and with the conclusion which has emerged from the present study, that sea lions are not very well equipped physiologically to deal with a hot environment.

SUMMARY

The physiological responses of three California sea lions to different environmental temperatures were studied, principally in order to determine whether this species is physiologically equipped to deal with a hot environment.

None of the animals was able to achieve thermal equilibrium at an air temperature of 30 or 36 C. At 36 C, the animals were removed from the climatic room when their rectal temperatures had increased to 40 C.

Evidence was obtained that sea lions respond to these conditions by increasing the blood flow to the skin both on the trunk and in the extremities.

Neither thermal polypnea nor sweating was observed during exposure to heat, indicating that physiological evaporative cooling mechanisms are relatively unimportant in this species.

The sea lions appeared to show a strongly developed pattern of thermoregulatory behavior when exposed to heat, which suggests that behavioral mechanisms of temperature regulation may be more important than physiological temperature regulation.

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chapter 3

BUBBLE FORMATION DURING DECOMPRESSION

Chapter 3 is probably the best possible wedding between physical and biological science; the physics of bubble formation and the attendant implications of basic principles to the applied problem of decompression sickness and embolism.

A different technique is presented here, one of which allows bubble "trapping." Thus, one can more quantitatively measure appearance and subsequent growth of bubbles.

Based on these data, experiments to prevent or reverse bubble formation point strongly toward possible modification of existing decompression tables. These studies are a perfect example of Fenn's maxim.

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Bubble formation in gelatin: implications for prevention of decompression sickness

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Strauss, R. H. 1974. Bubble formation in gelatin: implications for prevention of decompression sickness. Undersea Biomed. Res. 1(2):169-174.–Gelatin exposed to N_2 at differing pressures was decompressed to form bubbles. Findings are consistent with the existence in gelatin of a spectrum of stable gas nuclei which can be compressed or transformed into bubbles. Results suggest that the number of bubbles and their total volume can be decreased, and decompression time shortened, if the gas supersaturation pressure (i.e. the difference in pressure between dissolved gas and environment) remains constant for decompression of a given tissue.

bubbles
nitrogen
gas elimination
decompression sickness
supersaturation
tissues

Decompression sickness is associated with diving, work at increased pressures in tunnels and caissons, and flying at high altitudes in unpressurized aircraft. The disease follows a reduction in ambient pressure and is generally thought to be a result of bubble formation and growth in blood or tissues (Harvey 1951; Buckles 1968). Manifestations may include paralysis or joint pain (the bends). The object of decompression in diving is to permit a transition from high to low pressures in the least possible time consistent with the avoidance of decompression sickness.

In 1908 J. S. Haldane and coworkers enunciated many of the principles presently utilized to decompress divers and avoid decompression sickness (Boycott, Damant, and Haldane 1908). One hypothesis, sometimes called the Haldane ratio principle, is: "In decompressing men or animals from high pressures the first part should consist in rapidly halving the absolute pressure: subsequently the rate of decompression must become slower and slower, so that the nitrogen pressure in no part of the body ever becomes more than about twice that of the air." Haldane assumed that increasing the ratio (tissue gas pressure)/(ambient pressure) above a critical limit at any time during decompression would lead to bubble formation and growth. This article describes a series of decompression tests in which gelatin is used to simulate animal tissue. The study examines primarily the characteristics of initial bubble formation rather than the growth of existing bubbles by diffusion of gas. The principal finding is that Haldane's method is not optimal for avoiding bubble formation and growth in gelatin.

METHOD

Bubble formation and growth have been studied in various media (Harvey 1951; Buckles 1968; Epstein and Plesset 1950; Bateman and Lang 1944). Nevertheless, the initial process of bubble formation, which could be critical in decompression, is poorly understood. This lack of information is largely a result of the methods of observation and the nature of the systems studied: bubbles which form within liquids generally rise when still too small to be observed; bubbles which form in opaque tissues cannot be seen. The decompression of transparent gelatin avoids these difficulties and yields bubbles which are stationary and can be counted and measured.[1]

A single batch of gelatin was made by dissolving 127 g Knox gelatin crystals in 5 liters (l) of water and then freezing 10 ml aliquots. For each experiment, gelatin was thawed at 40°C and pipetted into two slightly curved glass counting chambers of approximate horizontal cross section 6 mm by 27 mm diameter. The counting chambers were filled to a depth of 4 mm with liquid gelatin and placed in ice water for 10 minutes to speed gelation. Two chambers were then placed in a 21°C water bath within a small pressure chamber. Pressure was increased at 13.6 atm/min to some maximum pressure P_m by adding N_2, and then maintained at P_m for 20 minutes or longer. The gelatin could be viewed and photographed at any pressure with a microscope through a window in the pressure chamber. No bubbles were visible prior to decompression.

Saturation of the gelatin by N_2 took place at pressure P_s, which was set equal to or less than P_m. The time spent at saturation pressure P_s was 5.25 hours unless otherwise noted. According to calculations by the method of Crank (1957), and assuming a diffusion coefficient of N_2 in water of 11×10^{-4} cm^2/min (Bartels 1971), the bottom of a 4-mm gel layer would be greater than 97% saturated after 5.25 hours. Saturation time was also investigated experimentally by varying time at P_s from 1 second to 24 hours ($P_s = 73$ psig). The maximum number of bubbles was reached at saturation times between 5 and 7 hours.

Decompression was achieved by lowering the pressure from P_s to a final pressure P_f in 10 seconds, during which there was little time for diffusion of gas from within the gelatin to the atmosphere. Within seconds after decompression, bubbles became visible and grew rapidly. After a few minutes no additional bubbles appeared, and growth slowed and eventually ceased. The system was then stable for hours. Bubbles occurred throughout the gelatin and were approximately equal in diameter in the lower 3 mm, which corresponded to a gelatin volume of about 0.372 ml/counting chamber. The bubbles in the lower 3 mm were counted using a 7-power microscope. The upper 1 mm and meniscus were disregarded because bubbles near the surface appeared more numerous and smaller than elsewhere.

RESULTS

Supersaturation pressure P_{ss} is the amount by which dissolved gas pressure exceeds ambient pressure. Thus, immediately following decompression and before significant diffusion occurs, P_{ss} is given by $P_{ss} = P_s - P_f$. It was hypothesized that N_b, the number of bubbles appearing, was a function of P_m, P_s, and P_f. Briefly, the maximum pressure P_m represents a pressure which apparently decreases the size of gas nuclei, making bubble

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[1] Cavitation studies in which a glass rod was suddenly torn out of gelatin are mentioned by Harvey (1951). More recently, extensive decompression tests have been carried out on gelatin by LeMessurier (1972). I am grateful to Carl Edmonds, M.D., for calling this work to my attention (private communication of August, 1973).

formation more difficult, and the supersaturation pressure P_{ss} promotes the formation and growth of bubbles.

The dependence of bubble formation upon supersaturation pressure was investigated (Fig. 1). In this experiment $P_m = P_s = 21.4$ atmospheres absolute (ATA), and P_f was allowed to vary, producing P_{ss} of 8.2 to 20.4 atmospheres (atm). The number of bubbles and their size increased with increasing P_{ss}. Similar results have been observed in other media (Hills 1967; Gent and Tompkins 1969).

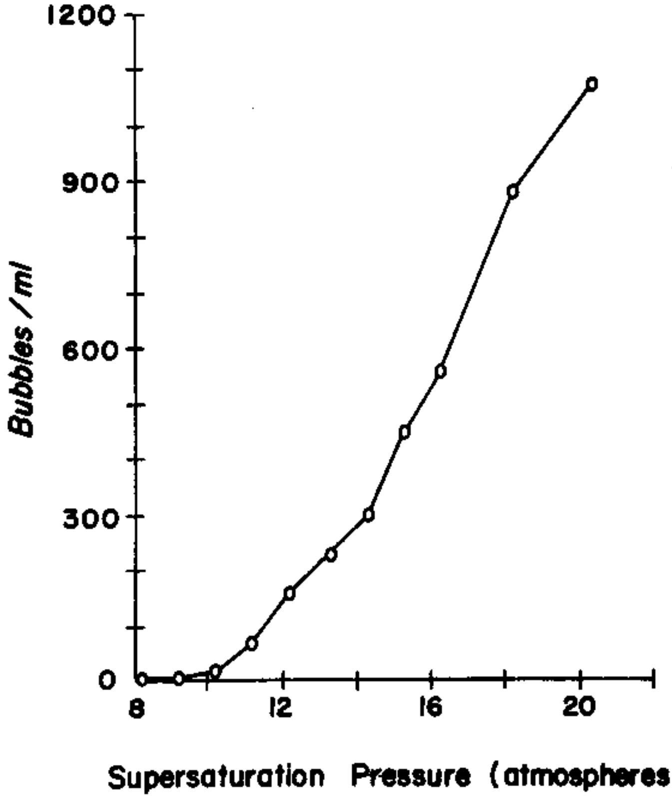

Fig. 1. The number of bubbles appearing in gelatin increased as supersaturation pressure increased. Maximum and saturation pressures were 21.4 ATA. Two chambers were counted at each pressure.

Bubble formation can be suppressed in water (Hemmingsen 1970) and shrimp (Evans and Walder 1969) by raising the maximum hydrostatic pressure at some time before decompression. This point was confirmed in experiments A and B (Table 1) by varying only the maximum pressure P_m. The greater P_m was maintained for 20 minutes at the beginning of experiment B and resulted in fewer bubbles ($P < 0.001$). Earlier experiments showed that the effect of P_m increased with time and was complete within 20 minutes. Any diffusion of gas into the gelatin during the time spent at the elevated P_m would tend to cause increased bubble formation rather than the decreased bubble formation which was observed. These findings are consistent with the existence in gelatin of nuclei of gas which can be compressed in a manner which is not entirely reversible. The effect of P_m upon bubble formation was not an artifact of the gel state since results were similar when P_m was applied to the sol (liquid) state before it became a gel.

In these experiments P_s/P_f immediately following decompression corresponds to the Haldane ratio (tissue gas pressure)/(ambient pressure). In order to test the effect of P_s/P_f upon bubble formation it is necessary to hold both P_m and P_{ss} constant because, as has been shown, each greatly affects bubble formation. This was done in experiments B and C (Table 1). P_s and P_f were varied. The larger ratio P_s/P_f resulted in fewer bubbles ($P < 0.05$) which is opposite to the prediction of the Haldane ratio hypothesis. These findings suggest an alternative basis for decompression. In terms of staged decompression (by steps), they imply that if pressure is reduced repeatedly by a constant value following sufficient time at each pressure for gas equilibration, bubbles will form only following the first pressure drop. Such was found to be essentially the case when ambient pressure was reduced successively from 21.4 ATA to 11.2 ATA to 1.0 ATA following 5.25 hours at each pressure for gas equilibration. In 5 trials, using 2 counting chambers in each trial, the first pressure drop of 10.2 atm resulted in a total of 208 bubbles formed. The second pressure drop of 10.2 ATA resulted in the growth of existing bubbles and the formation of a total of 7 additional bubbles. This increment of 3.4% could be due to small technical variations.

TABLE 1
Number of bubbles N_b as a function of pressure variations

Experiment	P_m ATA	P_s ATA	P_f ATA	P_{ss} atm	P_s/P_f	N_b bubbles/ml
A	11.2	11.2	1.0	10.2	11.2	1380.0 ± 40.0
B	21.4	11.2	1.0	10.2	11.2	40.8 ± 2.4
C	21.4	21.4	11.2	10.2	1.9	51.1 ± 4.0

The above results suggest *a.* that the initial pressure drop of decompression should be sufficiently small to prevent or minimize bubble formation; and *b.* that during subsequent decompression, supersaturation pressure could be maintained constant without additional bubble formation. The advantage of maintaining supersaturation pressure is that the diffusion gradient of gas from tissue is maintained, thus speeding decompression. However, the large initial supersaturation pressure of the Haldane method would foster early formation of bubbles into which gas would diffuse during the remaining decompression as long as supersaturation existed. Haldane's method of decreasing the supersaturation pressure as ambient pressure decreases may well be an empirical attempt to control the size of bubbles which formed much earlier. The above principles can be applied to either continuous or staged decompression.

Standard U.S. Navy decompression schedules are based upon the Haldane ratio hypothesis (Workman 1969). In the final experiment a U.S. Navy decompression schedule (U.S. Navy 1970) and an experimental decompression schedule based upon the present results were compared using gelatin 2 mm deep. All bubbles which formed were counted and measured. Figure 2 presents the pressure-time profile for the two schedules. Bubbles became visible following the large initial decompression of the Navy schedule. In the experimental schedule, the initial decompression was considerably less and was followed by linear decompression (which does not necessarily lead to a constant supersaturation pressure).

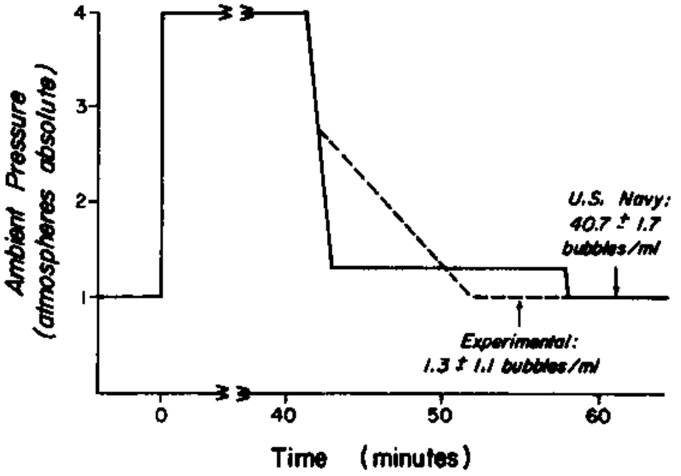

Fig. 2. Comparison in gelatin of a standard U.S. Navy decompression schedule (solid line) (U.S. Navy 1970) and an experimental decompression schedule (broken line). The latter was shorter and resulted in fewer bubbles.

Following decompression in 16 minutes 40 seconds by the U.S. Navy schedule there were 40.7 ± 1.7 bubbles/ml with a total volume of 4.79 ± 0.52 mm^3/ml. Following decompression in 10 minutes 40 seconds by the experimental schedule there were 1.3 ± 1.1 bubbles/ml with a total volume of 0.11 ± 0.04 mm^3/ml. The experimental schedule was shorter than the Navy schedule and resulted in fewer bubbles ($P < 0.001$) with a lesser total gas volume ($P < 0.001$).

The relationship between bubble formation in gelatin and decompression sickness is unknown. The gelatin experiments appear to reflect the response of gas nuclei to pressure changes. Nuclei within animals, either preformed or resulting from motion, have been proposed as a critical factor in bubble formation (Harvey 1951; Evans and Walder 1969). The above findings are consistent with this notion and suggest that the supersaturation pressure of a given tissue may remain constant during decompression. This important result implies that a nontraditional approach is needed to the problem of decompressing persons as safely and quickly as possible.

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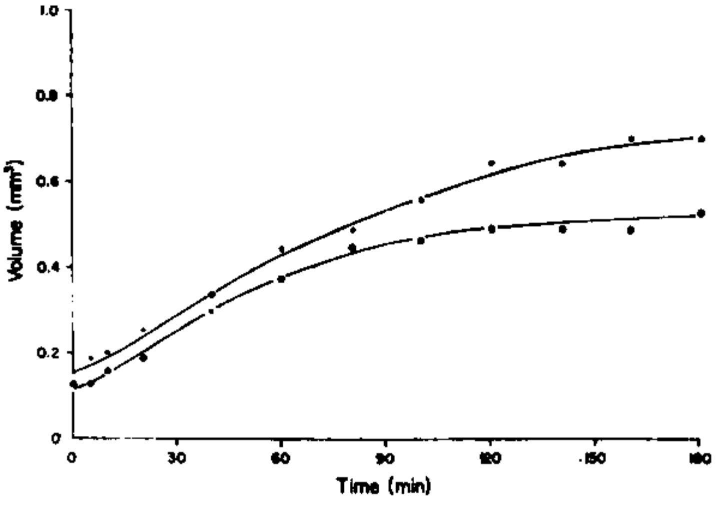
Isobaric Bubble Growth: A Consequence of Altering Atmospheric Gas

Abstract. *During certain treatments of decompression sickness following dives made with compressed air, the U.S. Navy advocates breathing helium-oxygen mixtures. However, stable nitrogen bubbles created within gelatin by decompression have been found to enlarge when the atmosphere was switched from nitrogen to helium without changing ambient pressure. This suggests that decompression sickness would be worsened by switching from nitrogen to helium in the breathing gas mixture.*

Decompression sickness affects divers whose tissues have become supersaturated with gas. The primary cause is generally thought to be the formation and growth of bubbles within tissues and blood (*1*). The treatment of decompression sickness involves (i) increasing the ambient pressure in order to reduce bubble size and (ii) breathing oxygen in order to increase the gradient for inert gas loss from the body and to increase tissue oxygenation. Under certain conditions, such as very high ambient pressures, inert gas as well as oxygen must be breathed in order to avoid oxygen toxicity. The *U.S. Navy Diving Manual* (*2*) advocates that the inert gas breathed be helium, even when decompression sickness has occurred after diving with air or other nitrogen-oxygen mixtures. The rationale appears to be to increase the net rate of loss of nitrogen from the body (*3*). However, switching to helium may be the wrong thing to do. There is considerable evidence that the body is saturated more rapidly by helium than it is desaturated by nitrogen (*3, 4*). During this transition period, bubbles within the body are expected to grow, since helium would diffuse into them faster than nitrogen would diffuse out. We have demonstrated this in a model in which stable bubbles were created in gelatin by decompression. The model simulates the body since diffusion calculations show that the gelatin is saturated by helium more rapidly than it is desaturated by nitrogen.

Experiments were performed in a small pressure chamber having a window in one end through which gelatin and bubbles could be seen and photographed by microscope at any pressure. Gelatin was prepared in a single batch by dissolving 127 g of Knox gelatin crystals in 5 liters of water. Portions were frozen and subsequently thawed at 40°C for each experiment. Thawed gelatin was pipetted into three rectangular glass chambers of horizontal cross section 6 mm by 27 mm, the gelatin depth at the meniscus being 4 mm. Before compression, the gelatin was converted from sol to gel by partly immersing the glass chambers in ice water for 10 minutes. The glass chambers were then placed in a water bath at 21°C inside the pressure chamber, which was pressurized at 13.6 atm/min by adding N_2. The chamber pressure was held at 21.4 atm absolute for 5.25 hours for the gelatin to become saturated with N_2. Pressure was then decreased to 11.2 atm absolute in 10 seconds, following which bubbles ap-



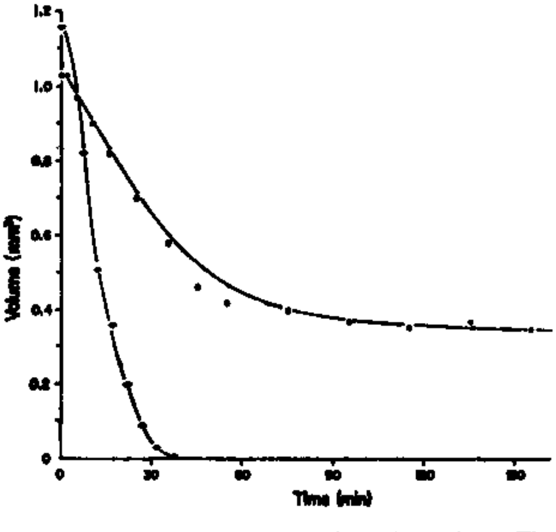


Fig. 1 (left). Switching atmospheric gas from N_2 to He causes bubbles in gelatin to grow. Ambient pressure is unchanged. Fig. 2 (right). Switching atmospheric gas from He to N_2 causes bubbles to shrink or disappear.

peared within seconds and grew for a number of minutes. There were 17.3 ± 1.2 bubbles in the lower 3 mm of gel in each glass chamber. Bubbles within 1 mm of the surface were not counted because they were smaller, more numerous, and difficult to see through the meniscus. The system was allowed to stabilize overnight. The next morning the chamber was flushed with helium for several minutes without altering ambient pressure. Photographs of bubbles were taken serially before and after flushing.

Before the pressure chamber was flushed with helium, there was no change in bubble size. Soon after it was flushed all the bubbles grew, and subsequently stopped growing, as indicated for two bubbles in Fig. 1. The change in bubble volume appeared to depend on the initial bubble volume and the bubble's distance from the gel surface and from other bubbles. The growth of five bubbles in three separate experiments was measured with similar results. These data suggest that switching from breathing nitrogen-oxygen to breathing helium-oxygen during the treatment of decompression sickness would make the disease worse rather than better. This conclusion is also supported by studies of the change in size of subcutaneous pockets containing various gases in rats (*5*). Other related experiments in animals have involved venous air embolism (*6*), pneumothorax (*7*), and bowel obstruction (*7*).

The converse experiment was performed, in which the initial saturation was by helium and the chamber atmosphere was subsequently changed to nitrogen. Bubbles near the surface became smaller and those deep in the gel disappeared. The histories of two bubbles are shown in Fig. 2. A total of four bubbles were measured in three experiments with similar results. These data support work by Keller and Buhlmann (*8*), in which the decompression time in humans was shortened by switching from helium-oxygen to nitrogen-oxygen breathing during decompression.

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30 May 1974

chapter 4

HYPERBARIC PHARMACOLOGY and EFFECTS OF HELIUM BREATHING

How inert is an inert gas? Does hyperbarism, particularly the thermal and CNS narcotic effects of hyperbarism, alter responses to CNS-active drugs or to antipyretics?

Since helium is the inert gas of choice, replacing nitrogen, in deep or prolonged dives, the obvious question arises whether observed effects of high pressure, i.e., bradycardia, might be due to possible pharmacologic effects of helium.

We have concentrated here on cardiovascular function and its autonomic control, since alteration of these systems is readily observed during helium dives. The investigation of the ANS is particularly thorough (Nicholas et al.).

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The antipyretic effects of hyperbaric air and salicylate on rats

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Hart, J. L. 1974. The antipyretic effects of hyperbaric air and salicylate on rats. Undersea Biomed. Res. 1(1):83-89.–Rectal temperatures were recorded in yeast-fevered rats exposed to normobaric (1 ATA) and hyperbaric (3, 6, 9 ATA) air, and the antipyretic actions of sodium salicylate (15, 30, 100 mg/kg) were compared in the normobaric and hyperbaric conditions. Rats exposed to the hyperbaric conditions showed decreases in body temperatures that were highly correlated with pressure, and probably due to the increased heat loss to the hyperbaric environments. The temperatures of salicylate-treated rats in hyperbaric air showed dose-dependent decreases below control rats in the same environments. The magnitudes of these decreases were not significantly different from those of salicylate-treated rats in normobaric air. The total temperature drop of salicylate-treated rats exposed to hyperbaric air was the sum of the antipyretic action of the salicylate and the temperature-lowering effects of the hyperbaric environments. It is concluded that hyperbaric air does not interfere with the antipyretic action of salicylate, and that the temperature-lowering effects of salicylate add to, but do not potentiate, the temperature-lowering effects of hyperbaric air environments.

salicylate antipyresis
hyperbaric air
temperature
heat loss
drugs

Gaining an understanding of the safe and effective use of pharmacological agents under hyperbaric conditions is becoming more important as diving for sport, scientific, and industrial purposes increases in frequency. Bennett (1972) has recently reviewed efforts to identify drugs which prevent or decrease the major diving hazards of inert gas narcosis, oxygen toxicity, and decompression sickness. However, less effort has been invested in determining how hyperbaric conditions might influence the efficacy and/or therapeutic effects of drugs. Of particular interest are the readily-available *over-the-counter* drugs, the most common of which is aspirin. Bennett (1962) reported that aspirin was effective in ameliorating nitrogen narcosis in rats as determined by the minimal stimulus required to elicit a tail twitch. In addition, Serril et al. (1971) observed that aspirin increased the sensitivity of mice to oxygen toxicity. However, studies of the therapeutic effects of aspirin under hyperbaric conditions have not been reported.

The present study was undertaken to investigate the antipyretic action of salicylate under hyperbaric air conditions. The temperature-lowering effects of *therapeutic* doses of sodium salicylate (Loux et al. 1972) were observed in rats in air at 1, 3, 6, and 9 ATA–the practical limit of compressed air diving (260 fsw).

METHODS

Male Wistar rats (Simonsen, California) ranging in weight from 220 to 275 grams were maintained in an air-conditioned room (21-23°C) for 6 to 13 days prior to their use. At 0930 hours on Day 1, rats were placed in wire-mesh restraining cages. Thermistor probes (Yellow Springs Instrument Co., Series 400 or 423) inserted 5 cm into the rectum and secured to the tail with tape were used to measure temperatures, which were read every 30 min on a Tele-Thermometer (Yellow Springs Instrument Co., Series 43). From 10-12 rats were used simultaneously. Temperatures fell initially but did not fluctuate significantly after 90 min in the restraining cages. At 1230 hours, nonfevered control temperatures were recorded. The average (± SE) temperature of the 138 rats studied was 37.21 ± .10°C at this time. Fever was then induced by the subcutaneous injection of 20 ml/kg of a 20% aqueous suspension of brewer's yeast (Mead Johnson), according to the method described by Loux et al. (1972). At the same time, rats that were to serve as nonfevered controls received equal volumes of the vehicle (distilled water). The rats were then returned to their housing cages (3/cage) and fasted for the duration of the experiment. Water was available ad libitum.

At 0830 hours (20 hours post-yeast) on Day 2, the rats were returned to the restraining cages where rectal temperatures were recorded every 30 min until 1400 hours. As on Day 1, temperatures fell initially but did not fluctuate significantly between 0900 and 1000. The temperatures at 1000 hours (21.5 hours after yeast administration) were used as the *post-yeast control* temperatures. The average (± SE) increase in temperature of the 114 fevered rats at this time was 1.69 ± .07°C, which was similar to the increase observed by Loux et al. (1972). The temperatures of the group of 15 fevered control rats did not vary significantly 21.5 to 24 hours following yeast administration. There was a significant decrease in temperatures during the last 90 min of the experiment (24.5 to 26 hours post-yeast) (.42°C at 26 hours post-yeast); however, data for analysis were taken from the 21.5- to 24-hour period when the control temperatures were constant. The temperatures of nonfevered control rats were not different on Day 2 from the previous day, and did not fluctuate significantly during the remaining 4 hours of the experiment.

Sodium salicylate (Baker and Adamson Products) injections were given intraperitoneally at 1000 hours, immediately following post-yeast control temperature readings. The salicylate was dissolved in 0.9% sodium chloride solution in various concentrations so that equal volumes (3 ml/kg) were injected for all doses tested. Nonfevered rats and fevered control rats received equal volumes of 0.9% sodium chloride solution only.

Following the salicylate or sodium chloride injections, rats to be exposed to hyperbaric conditions were placed in a 24-ft^3 Galeazzi Recompression Chamber. Compression to 3, 6, or 9 ATA with medical-grade compressed air was begun at 1015 hours at a rate of 30 ft/min. Throughout all experiments, the chamber was ventilated every 30 min with 9 to 11 ft^3 of compressed air. Decompression was begun at 1400 hours. Chamber temperature was recorded during the dives. Predive temperatures were between 21 and 23°C. Temperatures increased to 26-28°C during compression, but within 10 to 15 min following the completion of compression had returned to predive levels.

Doses of 10, 15, 30, 60, and 100 mg/kg of salicylate were given to groups of 6 rats at 1 ATA. Doses of 15, 30, and 100 mg/kg were given to groups of 6 rats at 3 and 6 ATA, and to groups of 3 to 4 rats at 9 ATA. Untreated fevered and nonfevered groups of at least 6 rats were also run at all pressures. The positions of the rectal thermistor probes were checked at the end of all experiments, and data were not used from rats in which the probes had slipped out more than 1.5 cm.

Results were analyzed by Student's *t* test and regression analysis. Differences with $P < .05$ were considered to be significant.

RESULTS

During exposure to all of the hyperbaric conditions studied (3, 6, and 9 ATA), rectal temperatures of fevered and nonfevered rats not treated with salicylate decreased significantly below those of untreated fevered and nonfevered rats at 1 ATA. The magnitudes of these decreases at the end of the observation periods (1400 hours) are plotted in Fig. 1. Decreases in temperature were positively correlated with pressure in both the fevered and nonfevered rats. In all cases, the magnitude of the decrease in temperature was greater for the fevered rats than for the nonfevered rats at the same pressure. The slope of the regression line for the fevered rats was significantly greater than for the nonfevered rats.

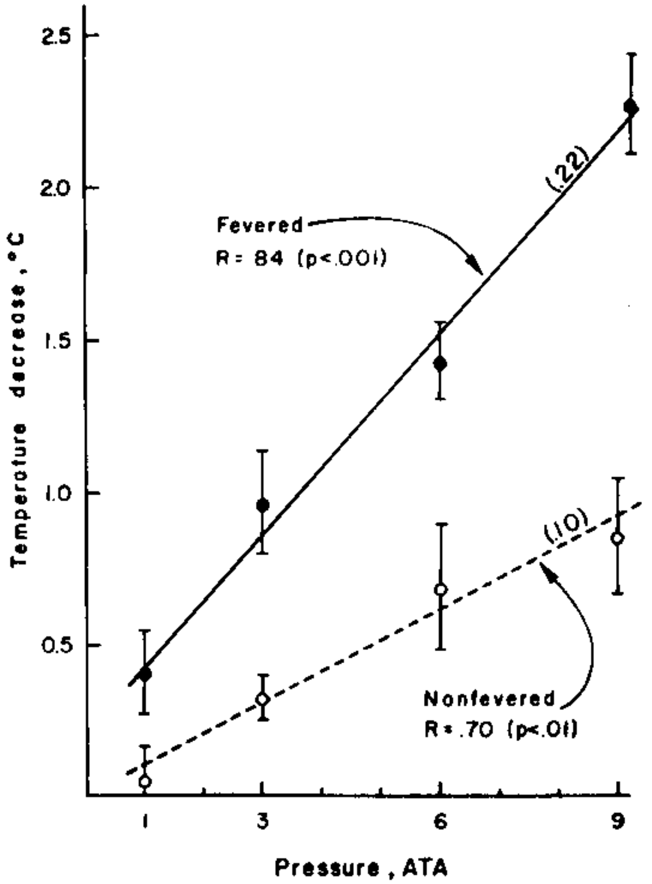

Fig. 1. The relationship between atmospheric pressure and decreases in body temperatures of fevered and nonfevered rats. The abcissa is the pressure in atmospheres absolute; the ordinate is the decrease in temperature below pre-pressure levels after 3.5 hours at pressure. The calculated least squares regression lines are shown, with slopes in parentheses. The points are means ± SE of the temperature decreases of the groups of rats exposed to each pressure.

R = correlation coefficient.

The log-dose effect curve for the antipyretic effect of sodium salicylate at 1 ATA is graphed in Fig. 2. This is the regression line of the *maximum* antipyretic effect. The maximum effect was taken as the difference between the mean temperature of the untreated, fevered group and that of the salicylate-treated, fevered group at the time when the greatest difference occurred. These maximum effects occurred at 60 min after salicylate injections for 10, 15, and 30 mg/kg, and at 120 min after injection for 60 and 100 mg/kg.

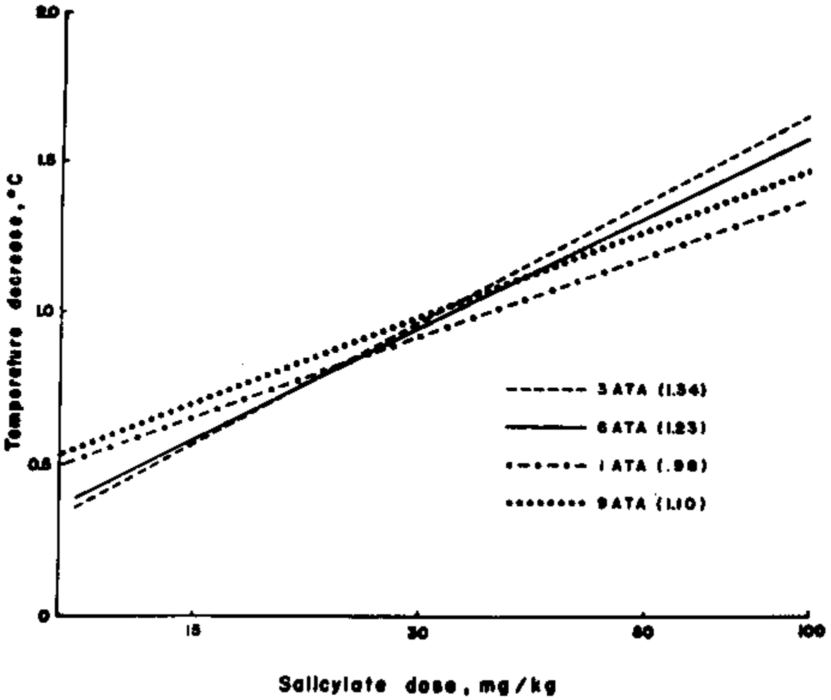

Fig. 2. The relationship between salicylate dose and temperature decrease in rats exposed to 1, 3, 6, and 9 ATA air pressure. The abcissa is the dose of salicylate on a log scale. The ordinate is the temperature decrease below fevered rats exposed to the same pressure. The calculated least squares regression lines are shown, with the slope of the lines in parentheses.

The three doses of salicylate (15, 30, and 100 mg/kg) tested in rats in hyperbaric environments all caused significant decreases in temperatures below untreated, fevered rats in the same hyperbaric environment. The maximum antipyretic effect in the hyperbaric conditions occurred at the same times as in the normobaric environment. For all doses, the actual body temperatures reached were lower in hyperbaric environments than for the equivalent doses at 1 ATA, and were progressively lower with increasing pressure. This is illustrated in Fig. 3, where the actual temperatures (mean ± SE) of the groups of rats receiving 100 mg/kg of salicylate are plotted for the 4-hour post-injection period. However, when the log-dose effect lines were plotted for the maximum salicylate-dependent temperature depressions (i.e.—the difference between untreated, fevered rats and salicylate-treated, fevered rats at the same pressure), there was no difference between the regression lines for the hyperbaric and normobaric conditions, or among the lines for the hyperbaric conditions (Fig. 2).

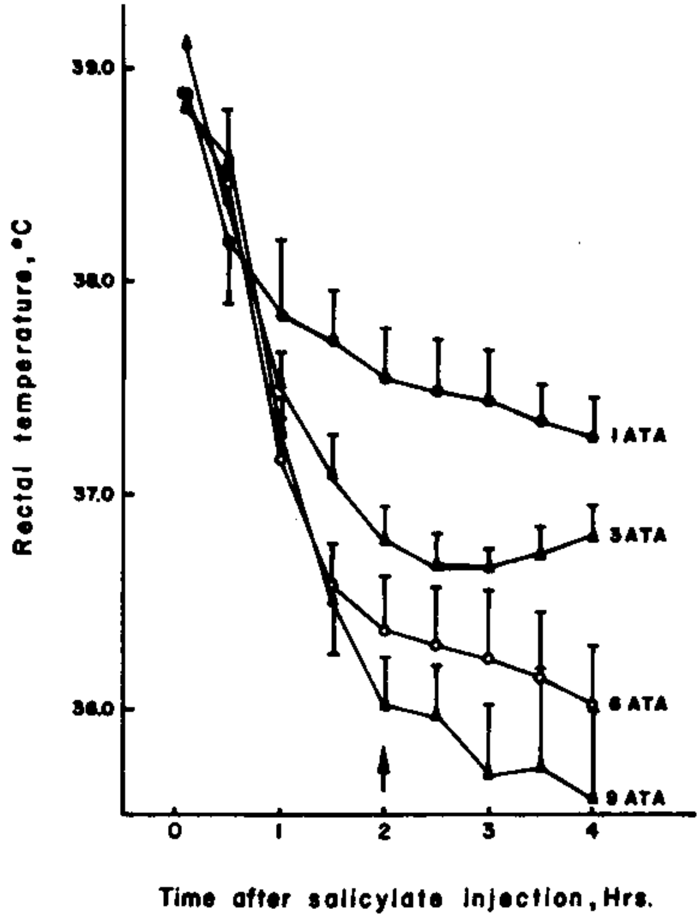

Fig. 3. The combined effects of salicylate (100 mg/kg) and hyperbaric air (1, 3, 6, and 9 ATA) on the temperatures of fevered rats. The values plotted are means ± SE. The arrow indicates the time of the maximum effect of the salicylate (see text for explanation).

DISCUSSION

Both hyperbaric air and salicylate had temperature-lowering effects on pyretic rats. The temperature decreases observed in fevered rats exposed to hyperbaric air were highly correlated with the magnitude of the pressure (Fig. 1). The temperature-lowering effects of salicylate were additive with the temperature effects of the hyperbaric environments. Dose-effect curves for the antipyretic effect of salicylate in the hyperbaric conditions based on temperature decreases below untreated, fevered rats in the *same* hyperbaric condition were not significantly different from the dose-effect curve at 1 ATA (Fig. 2), nor were the times of maximum effects of salicylate different in the hyperbaric conditions. The magnitude and time course of the antipyretic effects of salicylate appeared to be unaffected by the hyperbaric conditions studied here.

Pressure-related decreases in body temperatures observed in fevered and nonfevered control rats were probably due to the thermal effects of the hyperbaric air. The convective conductance of gas environments increases with increasing pressure, which results in greater loss of body heat (Raymond 1967). According to Raymond's calculations, convective heat loss in air by natural convection alone is almost 4 times greater at 9 ATA than at 1 ATA. The rate of heat loss at a given pressure is also dependent on the difference between skin and ambient temperatures—the greater the temperature difference, the faster heat is lost. This could explain the different depressions of body temperature observed in the untreated-fevered and untreated-nonfevered rats (Fig. 1). Whereas the ambient temperatures were the same, the body temperatures of the fevered rats were over 1.6°C higher than the nonfevered rats, just prior to exposure to hyperbaric conditions.

There have been no reports that the body temperatures of humans drop from exposure to the relatively mild heat-loss stress of the hyperbaric air conditions studied here. It is not unreasonable, however, that the same heat-loss stress would result in decreases in body temperatures of rats, since it is recognized that the body temperatures of small animals are more labile than the temperatures of humans, in part because of the higher surface area to body weight ratio of small animals (Fuhrman and Fuhrman 1961). Stetzner and DeBoer (1972) have also reported decreases in body temperatures of rats in hyperbaric air. Although the narcotic effects of nitrogen prohibit air breathing by humans much above 9 ATA (Bennett 1966), there are other diving conditions encountered by humans in which heat loss is a major consideration; examples of these are open-water diving (Hong 1973), and helium exposure (Moore et al. *in press*), where the additive temperature-lowering effects of salicylate could be of consequence.

The fact that the antipyretic effect of salicylate did not potentiate the temperature-lowering effect of the hyperbaric air is consistent with the findings of other investigators that salicylate did not interfere with normal temperature regulation in humans (Downey and Darling 1962; Rosendorff and Cranston 1968). In contrast, Satinoff (1972) reported that the normal temperature-lowering effects of large doses of salicylate in rats potentiated the body-temperature drop of 5°C cold-air exposure.

In conclusion, the present experiments indicate that hyperbaric air up to 9 ATA does not interfere with the antipyretic action of sodium salicylate, and that the temperature-lowering effects of salicylate add to, but do not potentiate, the temperature-lowering effects of hyperbaric air environments.

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Effects of Hyperbaric Conditions on the Responses of Animals To Central Nervous System Stimulants and Depressants

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Abstract—Awakening times were observed in animals in normobaric and hyperbaric air following administration of the CNS depressant pentobarbital. Awakening times of mice following doses of 50 and 75 mg/kg of pentobarbital were significantly longer at 2, 4, 6, and 8 ATA than those at 1 ATA. However, hyperbaric conditions did not significantly alter the awakening times of cats, guinea-pigs, or rats.

In 6 ATA air, the LD50 for the CNS stimulant picrotoxin in mice was significantly increased more than twofold above that at 1 ATA; the LD50 for caffeine was unaffected. The protective effect of hyperbaric conditions was due to the elevated P_{O_2}, for if the P_{O_2} was kept constant and the P_{N_2} was increased, the LD50 for picrotoxin was unaltered.

Introduction

As increasing numbers of persons participate in sport diving and in deeper excursions of longer duration, it is increasingly important to investigate responses to drugs in hyperbaric environments and to ascertain if the responses are altered by such conditions. Bennett (1) has recently reviewed the limited literature in this area.

One problem in gathering information in this area is that experimental animals appear to have greater resistance than man to the narcotic effects of inert gases. Whereas man exhibits psychomotor deficits during brief exposures to air at about 4 atmospheres absolute pressure (ATA) and rapidly becomes unconscious between 8 and 10 ATA (2, 3), animals are much more resistant, at least to the totally incapacitating effects of high nitrogen pressures. Marshall (4) has reported that more than 90 min exposure at 10 ATA is required to produce

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narcosis in mice (as indicated by loss of righting reflex), and rats appear "normal" after 4 hr at 8 ATA. However, the recent work of Walsh *et al.* (5) indicates that rats may be almost as susceptible as man to nitrogen narcosis, if careful psychomotor measurements are made.

Since the central nervous system (CNS) is thought to be the site of the narcotic action of nitrogen at high pressure, it was postulated that the effects of drugs acting on the CNS might be altered in experimental animals, at pressures which produce no obvious behavioral effects. Therefore, the effects of high pressures on the response to the CNS depressant (pentobarbital) and CNS stimulants (picrotoxin and caffeine) were investigated.

Methods

Pentobarbital.

Pentobarbital (¹) awakening times were studied in mice, rats, guinea-pigs, and cats under normobaric and hyperbaric conditions. Animals were exposed to hyperbaric conditions in a 24 cubic foot Galeazzi Recompression Chamber, the volume of which was reduced to approximately 20 cubic feet with water-filled carboys. Compression was carried out at the rate of 0.7 to 1 foot per sec with medical-grade compressed air. Although preliminary tests showed that changes in oxygen and carbon dioxide were insignificant over the time course of these experiments, the chamber was ventilated with 5 cubic feet of compressed air every 30 min for mice, rats, and guinea-pigs and every 15 min for cats. Decompression was carried out in stages over periods ranging from 25 min to 3 hr, depending on the size of the animal used. Apart from some pawing at ears and noses during compression, untreated animals exposed to experimental conditions showed no unusual overt behavior during compression, exposure, or decompression.

The awakening times of groups of 10 white mice (Swiss) were recorded in air at 1, 2, 4, 6, and 8 ATA following i.p. injections of 50, 75, and 100 mg/kg of pentobarbital (in 0.9 % NaCl solution). The awakening time was the time from injection until the resumption of walking. This end point is quite precise in mice and was recorded rather than the usual sleeping times, because the regaining of the righting reflex (the end point for sleeping times) could not be determined when animals were sealed in the hyperbaric chamber. It was necessary to use some end point that could be observed visually. Since awakening time (as well as sleeping time) is a variable, control groups were always studied simultaneously with pressurized groups, and groups were matched for weight and sex. Up to 3 groups were pressurized at one time, the highest doses being given first in a series of injections. All animals were housed 5 per cage during experiments. The pressurization period lasted 4 hr; the decompression lasted

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(¹) Nembutal®.

25 min. Student's *t*-test was used to analyze these results and results of pentobarbital studies in other animals. The criterion for significance was $p < .05$.

Rectal temperatures as well as pentobarbital (50 mg/kg) awakening times were recorded in 2 groups of 8 mice—one group in air at 1 ATA and the other in air at 6 ATA. Prior to the pentobarbital injections, control temperatures were taken in the hand-restrained mice with a YSI thermistor probe inserted 3 cm. Following the injections, the thermistor probes were reinserted and taped in place, and temperatures recorded every 5 min until the animals were awake. The difference between the control temperature and the lowest temperature reached was determined for each animal, and the means of these decreases in temperature were compared between the pressurized and non-pressurized groups.

The 11 cats (1.76 ± .1 kg) used in this study were given pentobarbital (20 mg/kg, i.p.) with sterile needles and syringes, since each cat was used 3 times. Each cat received pentobarbital injections at 5- to 8-day intervals. Immediately following one of the injections, the cat was placed in the hyperbaric chamber (4 cats/run) and pressurized to 5 ATA for 2 hr, before being decompressed for a period of 3 hr. Since the cats were restrained, the awakening time was when the cat lifted its head and righted its front quarters. This end point was not very precise in most cases, but was more consistent than any of the other signs observed. For analysis, the 2 control awakening times in air at 1 ATA were averaged and compared to the 5 ATA awakening time.

Twenty white rats (Wistar) (545 ± 12 g) were given i.p. doses of pentobarbital (30 mg/kg). Awakening times (resumption of walking used as the end point) were observed in 10 rats at 1 ATA and 10 rats at 6 ATA.

Groups of 10 guinea-pigs (Hartley) (170 ± 22 g) were given pentobarbital (35, 25, or 15 mg/kg, i.p.). Awakening times for each of the doses were recorded in 5 animals at 1 ATA and 5 animals at 6 ATA. The awakening end point for guinea-pigs was similar to that for cats, i.e., raising the head and righting the front quarters.

Central nervous system stimulants.

White mice (Swiss) were used in all of these studies. Drugs—caffeine and picrotoxin—were dissolved in 0.9 % NaCl solution. Injections were given s.c. in the mid-back region in volumes of 0.4 ml per 25 g of body weight. The LD50, potency ratios, and similarity of log dose-effect curves were determined by the method of Litchfield and Wilcoxon (6). Doses were administered to groups of 10 animals, and 4 or 5 doses were used to determine each dose-effect curve at 1 and 6 ATA.

Two groups of 10 mice were usually exposed to hyperbaric conditions at the same time. Injections of drugs required 2 to 3 min and compression to 6 ATA required 3 to 4 min (0.7 to 0.9 ft/sec). The total time from the first injection to the completion of compression was from 5 to 7 min. Dose-effect curves for mice treated with caffeine and picrotoxin were determined at 1 ATA and at 6 ATA of compressed air. For this hyperbaric series, the chamber was pressurized with medical-grade compressed air and ventilated with compressed air every 30 min during the 2 hr observation period. The partial pressure of oxygen in these conditions was 900 mm Hg, 6 times as great as the oxygen pressure at 1 ATA.

A dose-effect curve was also determined for picrotoxin-treated mice in "normoxic" hyperbaric conditions (6 ATA). For this series, pure nitrogen was used for compression, and a 3.5 % oxygen, 96.5 % nitrogen mixture was used for ventilating the chamber every 15 min. The partial pressure of oxygen under these condition was the same as under normobaric (1 ATA) conditions—150 mm Hg.

Decompression was carried out in stages over a period of 25 min; however, decompression did not influence the results of these studies, since all data were collected prior to beginning decompression. Control animals, which were exposed to the conditions identical to those of the treated animals, showed no adverse effects to compression, exposure, or decompression procedures with either gas mixture.

Results

Pentobarbital.

The results of the pentobarbital studies in mice are listed in Table I. The awakening times of mice receiving 50 mg/kg doses at pressures of 4, 6, 7, and 8 ATA were significantly greater than controls (1 ATA) receiving 50 mg/kg; there was no difference at 2 ATA. At a dose level of 75 mg/kg there were significant increases in awakening times when the mice were exposed to 2, 6, or 8 ATA; there were no significant differences at 4 or 7 ATA. At a dose of 100 mg/kg, there was a significant increase in awakening time only at 6 ATA. However, a number of problems were associated with this dose that make the interpretation of the results difficult. This dose of pentobarbital (100 mg/kg) is a "threshold" lethal dose, and there were occasional deaths in most of the control and pressurized groups. In the 8 ATA pressurized group there were 5 deaths out of 10 animals. (There were no deaths in the corresponding control group at 1 ATA.) Untreated animals showed no unusual signs at this pressure. High-pressure oxygen is known to be especially toxic to rats and mice, and van den Brenk and Jamieson (7) have reported that the toxic effects were greatly increased (as indicated by CNS damage and paralysis) in rats by a dose of 38 mg/kg of sodium pentobarbital. Although in the present experiments the oxygen pressure was below the toxic levels reported by van den Brenk and Jamieson, the dose of pentobarbital was higher (100 mg/kg), so that the deaths at this dose may have been due to the potentiation of the toxic effects of the high-pressure oxygen.

In the groups of mice in which rectal temperatures were measured following pentobarbital, there was a significant increase in awakening time in the pressurized group, but no difference in the temperature drops observed. The mean temperature drop in the control group was 5.46 ± .63° C, and in the pressurized group the drop was 5.14 ± .22° C.

TABLE I

Pentobarbital Awakening in Mice

Pento-barbital Dose	Pressure for Experimental Group (atmospheres)	Control (1 ATA) Awakening Time (1) y	Pressurized Awakening Time (1) x	Per cent Difference $\frac{x-y}{y}$
50 mg/kg	2	68.9 ± 2.8	65.8 ± 2.1	−4.5
	4	68.8 ± 7.1	89.8 ± 8.2	30.5 (4)
	6	52.9 ± 2.9	59.0 ± 7.7	11.5 (4)
	6	52.1 ± 2.7	61.6 ± 3.5	18.2 (4)
	7	46.8 ± 2.2	85.2 ± 6.4	82.0 (4)
	8	59.9 ± 2.8	78.6 ± 9.4	31.0 (4)
75 mg/kg	2	115.6 ± 11.5	154.0 ± 15.4	32.6 (4)
	4	124.1 ± 13.4	153.1 ± 18.6	23.4
	6	88.8 ± 5.6	107.1 ± 7.1	20.6 (4)
	7	113.5 ± 7.6	110.5 ± 7.0	−2.7
	8	102.6 ± 9.2	165.0 ± 7.0	60.5 (4)
100 mg/kg	4	221.1 ± 13.3	221.5 ± 22.7	0
	6	168.4 ± 9.7 (2)	237.5 ± 24.2 (2)	41.0 (4)
	7	180.8 ± 19.1 (3)	202.5 ± 8.7 (2)	11.5
	8	158.6 ± 10.8	206.8 ± 26.6 (3)	30.4

(1) Awakening times are shown in min, as means ± SE. There were 10 mice per group except as indicated by (2) or (3). — (2) Indicates mean of 9. — (3) Indicates mean of 5. — (4) Significant at $p < .05$.

The results of the pentobarbital studies in cats are shown in Table II. There were no significant differences in the awakening times at 1 AYA and 5 ATA. There was an increase in awakening times in 5 of the cats, and a decrease in 6. As can be seen from the data, there was a wide variation in control times among cats (54–267 min) as well as between the control times for some of the individual cats (167–299 min, and 133–286 min). One contributing factor to this was uncertainty regarding the awakening end point in some of the cats.

There was not a significant difference between the awakening time for rats in air at 1 ATA (65.3 ± 6.3 min) and in air at 6 ATA (65.4 ± 4.1 min). The

TABLE II

Pentobarbital Awakening Times in Cats at 1 ATA and 5 ATA

Cat No.	Treatment Order C = 1 ATA P = 5 ATA	Control Awakening Times at 1 ATA (1)		Average Awakening Time at 1 ATA (1)	Awakening Time at 5 ATA (1)	Difference Between Awakening Times at 1 ATA & 5 ATA (1)
		C_1	C_2	$\frac{C_1 + C_2}{2}$	P	$P - \frac{C_1 + C_2}{2}$
599	P-C-C	198	218	208.0	122	− 86.0
600	P-C-C	133	286	209.5	343	+ 133.5
594	C-P-C	272	262	267.0	192	− 75.0
581	C-P-C	237	186	211.5	238	+ 26.5
589	C-C-P	188	208	198.0	157	− 41.0
464	C-C-P	232	169	200.5	133	− 67.5
467	C-C-P	159	273	216.0	180	− 36.0
591	C-C-P	122	113	117.5	180	+ 62.5
593	C-C-P	40	68	54.0	147	+ 93.0
574	C-C-P	299	167	233.0	163	− 70.0
522	C-P-C	88	131	109.5	194	+ 84.5
Mean ± SE				184.0 ± 19.0	186.3 ± 18.4	

(1) Awakening times are shown in min.

awakening times of guinea-pigs at 1 ATA and 6 ATA were not significantly different at any of the doses tested (Table III).

TABLE III

Pentobarbital Awakening Times in Guinea-Pigs At 1 ATA and 6 ATA

Pentobarbital Dose (mg/kg)	Awakening Times (¹)	
	at 1 ATA	at 6 ATA
15	82 ± 3.2	78 ± 2.8
25	220 ± 10.1	243 ± 28.5
35	312 ± 34.6	268 ± 25.4

(¹) Awakening times are shown in min. Values are means ± S.E. There were 10 animals per group.

Central nervous system stimulants.

The LD50 for picrotoxin in mice was 5.3 mg/kg at 1 ATA in air, which increased to 14 mg/kg when the pressure was increased to 6 ATA. The dose-effect curves at 1 and 6 ATA were parallel and the potency ratio was 2.64 ($p < 0.05$). The "protective" effect of high pressure was absent when the mice were exposed to normoxic conditions in which oxygen pressure remained at 150 mm Hg: the LD50 was 5.8 mg/kg and not significantly different from that in air at 1 ATA (Fig. 1).

The LD50 for caffeine in air at 1 ATA was 300 mg/kg and in air at 6 ATA was 340 mg/kg. Although there was an increase in the LD50 with hyperbaric conditions, it was not a statistically significant difference ($p > .05$).

Discussion

The anesthetic action of pentobarbital was prolonged during hyperbaric conditions in mice but not in rats, guinea-pigs, or cats. The prolongation of awakening times in mice occurred at pressures as low as 2 ATA and there was some indication that awakening time was directly related to pressure. Under the hyperbaric conditions studied, both oxygen and nitrogen pressures were elevated; however, it is unlikely that the elevated oxygen pressure played a part in prolonging the effect of the pentobarbital, since it has been reported (8, 9) that hyperbaric oxygen alone shortens, rather than lengthens, sleeping times in mice.

There is no apparent narcotic effect of nitrogen in mice at the pressures tested, but it has been reported (4) that nitrogen is narcotic in mice at higher

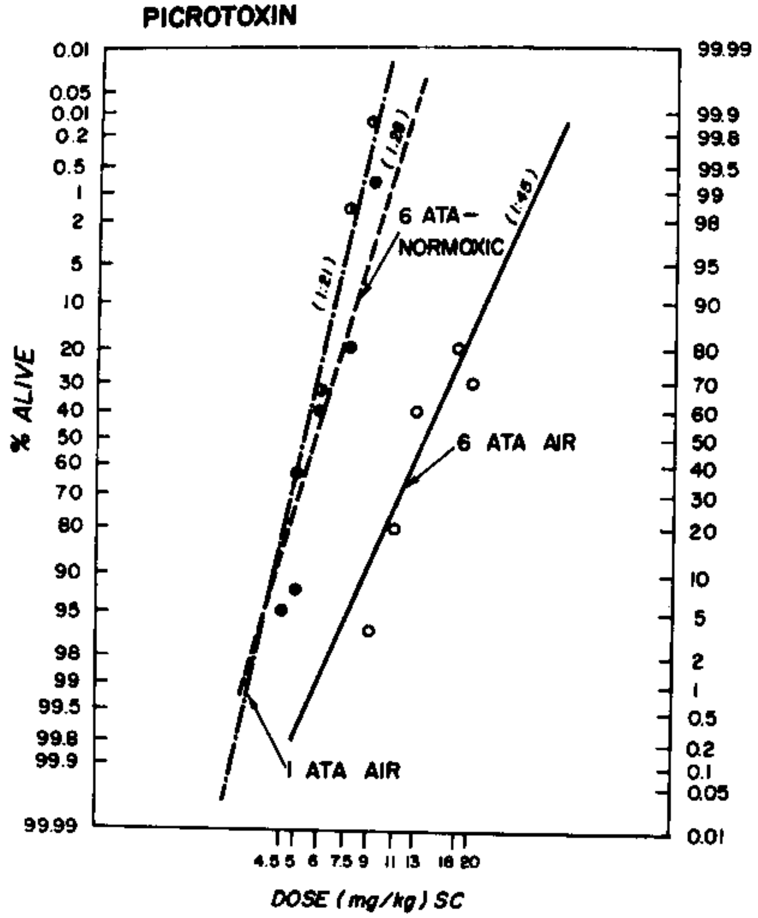

FIG. 1

Picrotoxin dose-effect curves for mice in 1 ATA air, 6 ATA air, and 6 ATA normoxic-3.5 % O_2, 96.5 % N_2. Data are plotted according to method of Litchfield and Wilcoxon (6). Numerals in parentheses are slopes of lines.

pressures. Therefore, it could be postulated that nitrogen is depressing the CNS at pressures lower than those at which obvious signs of narcosis appear, and that this is enough to prolong the depressant effects of pentobarbital. One might further postulate that the CNS of mice is more sensitive to the depressing effects of nitrogen than that of cats, rats, or guinea-pigs.

The possible role of body temperature on this selective effect of hyperbaric conditions on mice was considered. Lowered body temperature is known to prolong sleep produced by barbiturates (10). It is also recognized that the smaller the animal, the more labile its body temperature, and that this is probably dependent on the high metabolic rate and high ratio of surface area to body weight in small animals. In addition, CNS depressants such as pentobarbital impair the ability of animals to thermoregulate (11). The density of air in a hyperbaric environment is increased, which results in increased heat loss from an animal. Although this is normally a minor effect at the pressures used in these experiments, it could be significant in a small animal such as a mouse, with its thermoregulatory mechanisms compromised by the pentobarbital. However, the decrease in rectal temperature in the pentobarbital-treated mice was not significantly different in the control and pressurized groups. Further possible causes of the increased pentobarbital awakening times of mice exposed to hyperbaric conditions have not been investigated.

The LD50 of the CNS stimulant picrotoxin was significantly increased in mice in hyperbaric air, whereas that of caffeine was unchanged. The protective effect of high pressure in the case of picrotoxin was eliminated if oxygen pressure was maintained near control levels.

It was originally postulated that although the CNS-depressing effect of high nitrogen pressure was not apparent in the overt behavior of mice exposed to the pressures used in these experiments, it might be expressed as a lessening of the CNS stimulant and/or lethal effects of convulsants. If nitrogen has such an effect at the pressures used, it did not antagonize the toxic effects of all CNS stimulants, since there was no protection evident in the case of caffeine. Furthermore, the high oxygen pressure of the compressed air rather than the elevated nitrogen pressure appeared to be the critical factor in the increased resistance to the lethal effects of picrotoxin, since at normal oxygen pressure there was no longer any protection with the hyperbaric conditions. High oxygen pressure per se can cause convulsions, particularly in mice (8). However, in the present experiment, the levels of oxygen reached (900 mm Hg) were below the reported toxic levels in mice. Also, control mice exposed to 8 ATA compressed air (1200 mm Hg oxygen) for up to 4 hr showed no signs of oxygen toxicity. It is thus likely that the elevated oxygen provided "protection" against picrotoxin toxicity by increasing the oxygen content of the blood, thereby preventing lethal hypoxia during maximal extensor convulsions. Such convulsions usually immediately preceded death at 1 ATA. During these convulsions, respiration ceases because of full contraction of the respiratory muscles. Therefore, the actual cause of death at 1 ATA was probably hypoxia during the maximal convulsions. At 6 ATA, the additional oxygen dissolved in the blood due to the high oxygen pressure (900 mm Hg) could have been the critical factor in the animals surviving the periods of respiratory paralysis.

Summary

In general agreement with previous work, there were no obvious signs of nitrogen narcosis in the animals studied (mice, rats, cats, and guinea-pigs) in hyperbaric air up to 8 ATA. Nor was there any consistent evidence that the

effects of drugs which interact with the CNS were altered significantly by high nitrogen pressures studied. Pentobarbital awakening times were prolonged only in mice. The LD50 of picrotoxin, but not that of caffeine, was significantly elevated in mice in hyperbaric air (6 ATA); however, in normoxic environments of the same pressure, there was no protection from the lethal effects of picrotoxin. If, indeed, experimental animals are influenced by elevated nitrogen pressures in the range studied, more sensitive behavioral tests may be required to assess changes in responses to drugs in these animals.

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Effects of helium gas on heart rate and oxygen consumption in unanesthetized rats

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Lin, Y. C., and E. N. Kato. 1974. Effects of helium gas on heart rate and oxygen consumption in unanesthetized rats. Undersea Biomed. Res. 1(3):281-290.—The nonthermogenic effect of $He\text{-}O_2$ (80% helium and 20% oxygen) was studied in unanesthetized male Sprague-Dawley rats weighing 250-350 g. Blood pressure and heart rate (f_h) were recorded from a chronically implanted catheter located in the left carotid artery. Oxygen consumption ($\dot{V}_{O_2}$, $ml \cdot min^{-1} \cdot kg^{-1}$, STPD) was measured by a closed ventilated system. Ambient temperature was adjusted to between 15 and 35°C for $He\text{-}O_2$ and 10 and 35°C for air to alter $\dot{V}_{O_2}$. Rectal temperature and arterial blood pressure were not statistically different between air and $He\text{-}O_2$ environments. The relationship between simultaneously obtained f_h and $\dot{V}_{O_2}$ in 17 rats can be described by $f_h = 6.2\ \dot{V}_{O_2} + 158$ in air and $f_h = 4.9\ \dot{V}_{O_2} + 124$ in $He\text{-}O_2$ within the range studied. The intercepts ($P < 0.01$) but not the slopes ($P > 0.05$) were significantly different between air and $He\text{-}O_2$ environments, indicating that f_h was significantly lower in $He\text{-}O_2$ than in air at any given $\dot{V}_{O_2}$. At similar $\dot{V}_{O_2}$, elevated vagal and depressed sympathetic activities were observed in $He\text{-}O_2$, as elucidated by selective blockade of vagal and sympathetic activity by atropine and propranolol.

helium
depression effect of helium
heart rate and oxygen consumption
heart rate and ambient temperature

Helium at normobaric conditions is known, or assumed, to have no direct pharmacological potency. Physiological effects thus far reported have largely been attributed to its high thermal-conductivity characteristics, especially under high-density conditions (Webb 1970), or to its low specific density (Maio and Farhi 1967). Alterations in heart rate and/or energy expenditure under normobaric helium environments (Rhoades, Wright, Hiatt, and Weiss 1970; Clarkson, Shatte, and Jordon 1972; Stetzner and De Boer 1972; Raymond, Bell, Bond, and Lindberg 1968) and hyperbaric helium environments (Stetzner and De Boer 1972; Hamilton 1967; Salzano, Raysch, and Saltzman 1970; Raymond et al. 1968) are attributable to the indirect effect of the high thermal conductivity of helium. On the other hand, in experiments conducted in *thermal neutral* normobaric ambients, the breathing gas containing helium has been shown to exhibit antiarrhythmic activity following coronary artery ligation in the anesthetized dog (Pifarre, Cox, Jasuja, and Neville 1966, 1967; Raymond, Weiskopf, Halsey, Goldfien, Eger, and Severinhaus 1972; Toltzis and Scott 1972); to increase plasma growth hormone concentration in man during exercise (Raymond, Sode, Langworthy, Blosser, and Johnsonbaugh 1974); to suppress circulating catecholamine concentration in the anesthetized dog (Raymond et al. 1972); to potentiate bradycardia response during apnea in man (Hong, Moore, Lally, and Morlock 1973); to elevate ventricular fibrillation threshold (Leon and Harris 1973); to alter electroencephalographic activity (Hu and Russo 1972); to cause hemodilution and to increase CO_2 production without altering oxygen uptake, suggesting changes in metabolic pattern (Schatte, Jordon, Phillips, Clarkson, and Simons 1973); all indicating that helium may exert some primary pharmacological activities.

It is now recognized that in the determination of such primary actions, it is paramount to account for all the physical factors involved. In the present study, ambient as well as body temperatures were kept comparable in both He-O_2 (80% helium and 20% oxygen) and air (80% nitrogen and 20% oxygen). However, since the thermal neutral zone is greatly abbreviated in the helium environment (Leon and Cook 1960; Rhoades et al. 1967) and the thermal factor remains significant, it is important that metabolic rate itself be used as the basis for comparing cardiovascular parameters. In this way, cardiovascular burden of transporting oxygen can be equated in both environments.

Evidence suggesting the possible role of the autonomic nervous system in helium-effect mediation has been presented (Raymond et al. 1972). We have evaluated this problem by using sympathetic and parasympathetic blocking agents in the unanesthetized rat.

METHOD

Male, albino Sprague-Dawley rats ranging from 250 to 350 grams were used in the present study. They were individually caged and fed standard laboratory chow and water *ad libitum*, without fasting prior to experimentation. They were used either intact, with the exception of cutaneous electrode attachment for electrocardiographic recording, or surgically prepared as follows:

The carotid artery and jugular vein were cannulated with polyethylene tubing (PE. 50) under pentobarbital anesthesia. The tubings were threaded subcutaneously, brought out on the back of the neck between the ears, and secured using purse-string sutures. The tubes were filled with heparin-saline solution, heat sealed, and securely fastened with masking tape. The dorsal incision was closed using silk suture and allowed to heal for 2 to 3 days.

Blood pressure was monitored with a Statham P23Db pressure transducer and a Beckman Type R Dynograph via the carotid artery. Heart rate was obtained from ECG recordings or from the blood pressure trace. Rectal temperature was obtained by using a telethermometer and thermistor probe (#402, Yellow Springs Instruments Co., Inc.). They were calibrated with a precision mercury thermometer and a constant-temperature bath.

OXYGEN CONSUMPTION

Oxygen consumption was measured employing a closed-ventilation system (Fig. 1). The animal chamber measured 22 × 16 × 18 cm^3 and was sealed with a top plate which was equipped with air-tight penetrations for measurements of blood pressure, rectal temperature (T_r), and chamber ambient temperature (T_a) and for gas entrance and gas exit. T_a and the entrant gas temperature were controlled by submerging the animal chamber and a long coiled tygon tube in the same water bath. The flow rate was measured at room temperature (24-25°C) before entering the animal chamber by a float-type flowmeter (Gilmont Instruments, Inc.) interjected between the compressed gas source and the chamber. The flowmeter was calibrated for each mixture by using a Colin spirometer. Based on this calibration, the flow rate was set at 1,000 ml/min and 1,050 ml/min (same marking on the flowmeter), for air and He-O_2 mixture (80% helium, 20% oxygen), respectively. These flow rates provided inflow-to-outflow oxygen concentration differences of around 1%. Com-

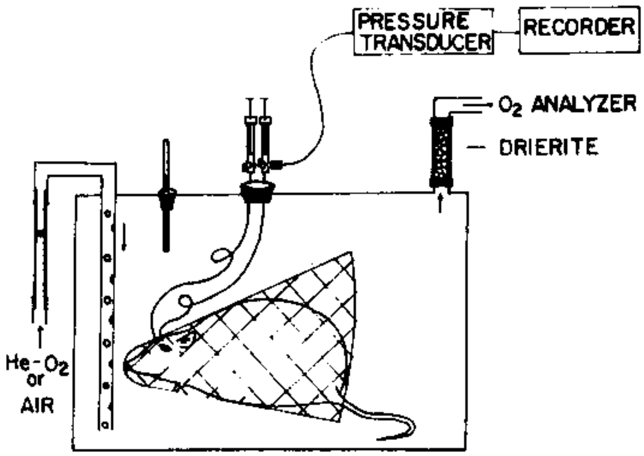

Fig. 1. Experimental set-up for measurements of oxygen consumption, blood pressure, and heart rate. The rat, confined in a wire mesh cone, was placed in a temperature-regulated Lucite chamber, which was ventilated with a known concentration of gas mixtures. The exhaust gases were monitored for oxygen. Blood pressure and heart rate were measured via the carotid catheter, while the jugular catheter was used for drug introduction. The entire animal chamber was submerged in a temperature-regulated water bath (not shown) for controlling ambient temperature in the animal chamber.

pressed sources of air and helium-oxygen mixture provided constant concentrations of oxygen in the inflow gas. The inflow O_2 concentrations of both gases were determined at the beginning of each experiment by passing the compressed gas through the flowmeter, the animal chamber without the animal, and a dessicator. Then the exhaust gas was measured for oxygen concentration by a Beckman OM-11 oxygen analyzer (Beckman Instruments). The oxygen analyzer heated surroundings of the oxygen sensor and the exhaust gas to 39°C before the gas reached the oxygen sensor in order to minimize the temperature and humidity effect on the diffusion process of oxygen through the membrane covering the oxygen sensor. The oxygen analyzer was calibrated with room air before each measurement was taken. The oxygen consumption ($\dot{V}_{O_2}$) was then represented by the product of flow rate and oxygen concentration difference between the gases entering and leaving the animal chamber. All $\dot{V}_{O_2}$ values were converted to $ml \cdot min^{-1} \cdot kg^{-1}$, STPD, from room temperature, at which the flow was measured, rather than animal-chamber temperature.

AUTONOMIC NERVOUS CONTROL OF HEART RATE

Atropine sulfate (1 mg/kg, Eli Lilly and Co.) or propranolol hydrochloride (8 mg/kg, Sigma Chemical Co.) was injected via the jugular vein catheter and peak responses were recorded. The dosages used were those established by Lin and Horvath (1972).

PROTOCOL

Experiment I

In the first series of experiments with six rats, oxygen consumption was not measured. The rat was contained in a plexiglas cylinder 15 cm in length and 7.5 cm in diameter, equipped with air-tight penetrations for the ECG cable and ports for gas entrance and gas exit. Entrant gas temperatures were 25°C for air and 35°C for He-O_2. Air and He-O_2 were alternated for 1 hour each for a total of 4 hours, and heart rate was monitored from the ECG records. The sequence of gas-mixture applications was randomized. The paired *t* test was used for comparisons of results obtained in air and He-O_2 environments. Although temperature control represented an attempt to compensate for the difference in thermal-conductivity characteristics of the two gas mixtures, there is no assurance that the thermal balance is the same, and heart rate comparisons in these two environments may be biased. The following experiments were therefore made.

Experiment II

Seventeen surgically prepared rats were individually placed in the animal chamber (Fig. 1). Heart rate (f_h) and oxygen consumption ($\dot{V}_{O_2}$) were monitored continuously and sampled at 5-minute intervals. Gas mixtures and/or T_a were changed every 30 min to alter $\dot{V}_{O_2}$ of the animal. The temperature range was 10 to 35°C for air and 15 to 35°C for He-O_2. Not all animals were subjected to the same temperature and gas-mixture combinations, but the number of observations were noted (see RESULTS). Group *t* tests were used for comparison of results obtained in the air and He-O_2 environments.

Experiment III

Another group of eight rats were treated, five with atropine and three with propranolol. Prior to drug injection, T_a was adjusted so that $\dot{V}_{O_2}$ in the He-O_2 environment matched $\dot{V}_{O_2}$ in the air environment.

RESULTS

EXPERIMENT I

Repetitive alteration of gas mixture (25°C for air, 35°C for He-O_2) in six unanesthetized rats caused repetitive changes in f_h. Paired *t* tests showed that f_h during 30-60 min of He-O_2 exposure were significantly lowered ($P < 0.05$) from the f_h in air just prior to each change to He-O_2 mixture. The overall f_h in the period of 4 hours continued to decrease, but f_h changes were reversible following each gas change, indicating a suppression effect of helium gas on the heart rate. The suppression, however, was small, in the order of less than 10% of the pre-He-O_2 value (Fig. 2).

EXPERIMENT II

Effect of T_a on $\dot{V}_{O_2}$ and f_h

A representative response of an unanesthetized rat to changes in ambient temperature and/or gas mixture is shown in Fig. 3. Heart rate and $\dot{V}_{O_2}$ were recorded every 5 minutes. Except for the first 5 minutes, f_h and $\dot{V}_{O_2}$ were relatively stable throughout the half-hour periods, and the last five values obtained were averaged to represent f_h and $\dot{V}_{O_2}$ in a specific gas media and T_a. Measurements of $\dot{V}_{O_2}$ were 42.1 ±0.5 (16), 26.6 ±1.08 (16), and 25.7 ±0.96 (17) for 10, 25, and 35°C, respectively, in air; 43.9 ±1.9 (13), 36.4 ±3.3 (14), 29.6 ±1.0 (13), and 35.6 ±3.1 (9) for 15, 25, 33, and 35°C, respectively, in He-O_2 (Fig. 4). The number of observations from 17 rats are indicated in parentheses. Heart rate was lowest in regions concurrent with lowest $\dot{V}_{O_2}$. Heart rates were as follows: 492 ±10.0, 359 ±8.9, and 352 ±8.7 for 10, 25, and 35°C , respectively, in air; 507 ±7.6, 413 ±11, 311 ±10.9, and 356 ±8.3 for 15, 25, 33, and 35°C, respectively, in He-O_2 (Fig. 4). The number of observations from 17 rats are as indicated for $\dot{V}_{O_2}$.

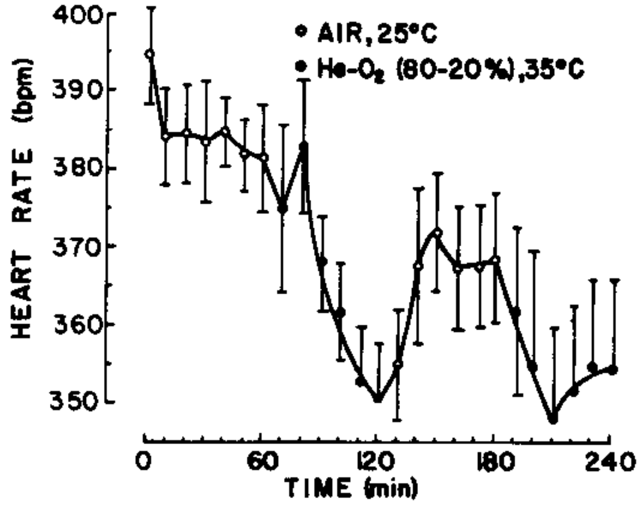

Fig. 2. Effect of alternating gas mixtures on the heart rate of six unanesthetized rats. Vertical bars are ± S.E. Heart rates during 30 to 60 min of He-O_2 exposure were significantly lowered ($P < 0.05$) from the heart rate in air just prior to each change to He-O_2 mixture, by paired *t* test.

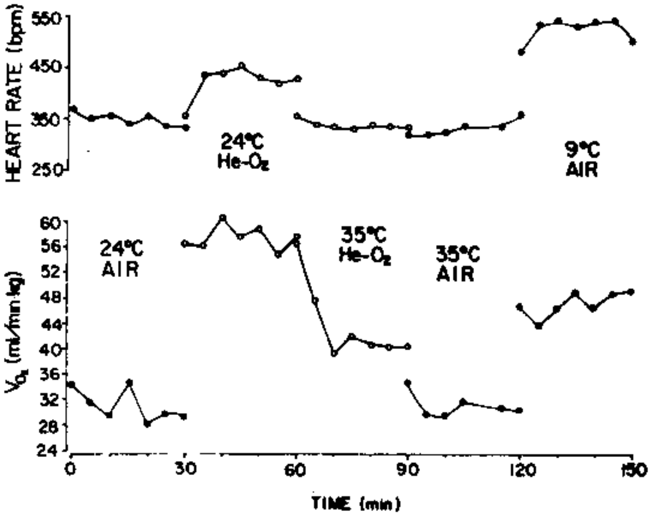

Fig. 3. Typical time course of heart rate and oxygen consumption ($\dot{V}_{O_2}$) responses to ambient temperature (T_a) and/or gas mixture changes. All except the first reading were averaged to represent the heart rate and $\dot{V}_{O_2}$ in a given T_a and gas mixture environment.

Relationship between f_h and $\dot{V}_{O_2}$

When the individual observations (listed previously) of f_h were plotted as a function of $\dot{V}_{O_2}$, it was found that f_h was linearly correlated with $\dot{V}_{O_2}$ at a *P* value of less than 0.001 both in the air and in the helium environments (Fig. 5). The intercepts ($P < 0.01$), but not

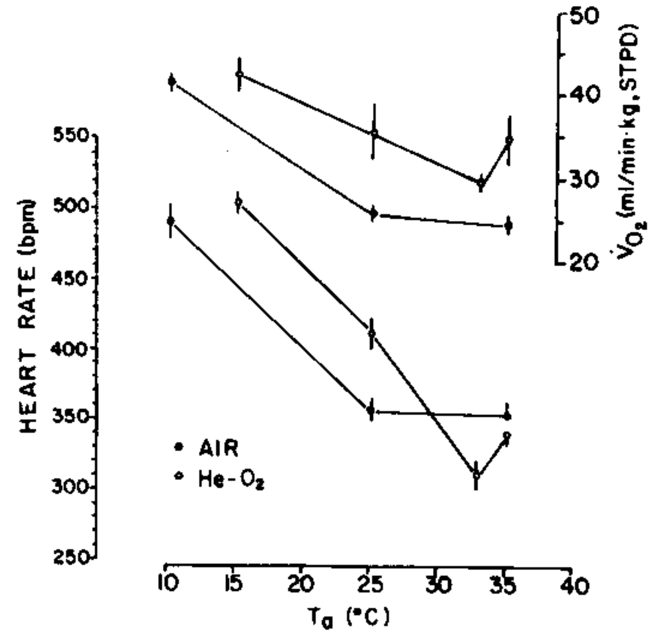

Fig. 4. Effect of temperature (T_a) on oxygen consumption ($\dot{V}_{O_2}$) and heart rate of the unanesthetized rat. Vertical bars indicate ± 1 standard error of the mean. The number of oberservations from 17 rats were 16, 16, and 17 in 10°C, 25°C and 35°C air, respectively; and 13, 14, 13, and 9 in 15°C, 25°C, 33°C and 35°C He-O_2, respectively.

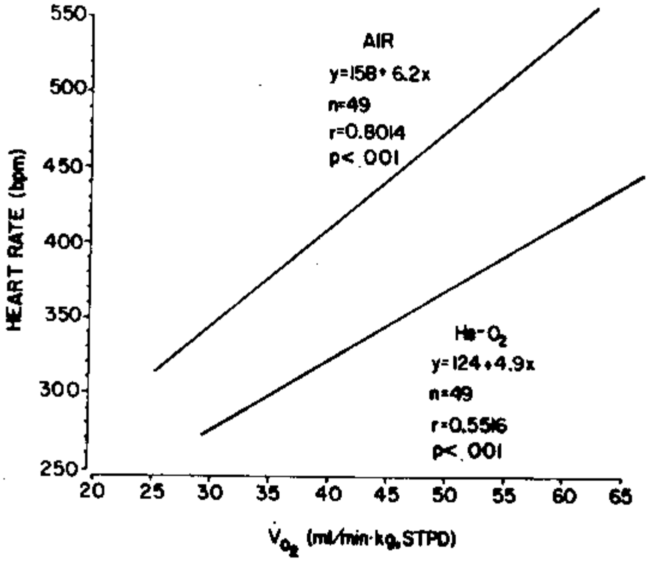

Fig. 5. Correlation between heart rate and oxygen consumption ($\dot{V}_{O_2}$). A total of 49 observations in each environment were made in 17 rats.

the slopes ($P > 0.05$) for the two regression lines were statistically different, indicating that heart rate was lower in the He-O_2 environment than in air at any given $\dot{V}_{O_2}$ level. Heart rates differed by more than 100 beats per min at $\dot{V}_{O_2}$ values ranging from 40 to 60 ml/kg of body weight.

Relative contribution of sympathetic and parasympathetic nervous control over f_h

The peak increase in f_h following atropine treatment and maximal depression in f_h following propranolol treatment are summarized in Table 1. The f_h was elevated to a greater degree in He-O_2 than in air following atropine treatment ($P < 0.05$), indicating increased vagal influence on the heart in the He-O_2 environment. Propranolol effect on f_h was greater in air than in He-O_2 ($P < 0.01$), indicating reduced sympathetic influence on the heart in He-O_2. Comparison of the pretreatment f_h values showed that the f_h was lower in He-O_2 than in air (Table 1).

TABLE 1

Effect of atropine sulfate (1 mg/kg) and propranolol hydrochloride (8 mg/kg) treatment on the heart rate of the unanesthetized rat

Treatment group	Media	T_a (°C)	n	MBP (mm Hg)	T_{re} (°C)	$\dot{V}_{O_2}$ (ml/min/kg)	Heart rate Pretreatment	Heart rate Treated
Atropine	Air	25-26	5	118 ± 8	39.9 ± 0.16	29.1 ± 2.4	421 ± 11	+57 ± 10
	Helium	33-34	5	124 ± 5	39.6 ± 0.21	31.6 ± 1.9	399 ± 10*	+106 ± 16*
Propranolol	Air	25-26	3	106 ± 10	38.0 ± 0.08	32.6 ± 6.7	468 ± 32	−253 ± 23
	Helium	33-34	5	111 ± 10	37.9 ± 0.09	36.9 ± 5.6	395 ± 26*	−124 ± 24†

Values are mean ± S.E., n is number of rats, T_a is ambient temperature.

MBP, T_{re}, and $\dot{V}_{O_2}$ are pretreatment values of mean arterial blood pressure, rectal temperature, and oxygen consumption, respectively.

* $p < 0.05$, † $p < 0.01$, for paired comparisons between air and helium media.

Blood pressure and body temperature

The mean blood pressure and rectal temperature were not affected by air (25–26°C) to He-O_2 (33–34°C) transitions in both atropine and propranolol testing groups (Table 1).

DISCUSSION

Due to its low narcotic potency and chemical inertness, helium has been considered an ideal diluent for lowering gas densities during deep-sea diving operations, with the provision that its thermal effects are compensated for. The premise that helium lacks direct biological effect may require revision as nonthermogenic effects of helium are recognized in purified enzymes (Doebbler, Mohlke, McMullan, and Schreiner 1967), in tissue slices (Maio and Neville 1967; Rinfret and Doebbler 1961), in the mold (Schreiner 1968), in the chick embryo (Weiss et al. 1965), and in the whole organism (see INTRODUCTION, and Lin 1973). It is fortunate, however, that no direct adverse effects have been reported in man or other species.

The indirect thermogenic effect of helium gas has been widely demonstrated and has even been utilized to accelerate cooling of small homeotherms (Fisher and Musacchia 1968; Musacchia 1972; Musacchia and Jacobs 1973) and to induce maximal oxygen uptake (Rosenmann and Morrison 1974) without the use of extreme low temperatures. In the present study, the lower critical temperature of the rat at 1 ATA increased from 25°C in air to 33°C in He-O_2 (Fig. 4), in agreement with the studies of Rhoades et al. (1967) and Clarkson et al. (1972). Leon and Cook (1960) and Herrington (1940) have also shown elevation of the lower critical temperature, but with slightly lower values than the present study.

Depressed f_h in He-O_2 as compared with that in air at a comparable $\dot{V}_{O_2}$ (observed in this study) has been recorded previously. Rhoades et al. (1967) has shown that, in the rat, f_h in 33°C He-O_2 is lower than that in 23, 28, and 33°C air environments. Oxygen consumption in 33°C He-O_2 was not statistically different (mean value was, however, still higher in He-O_2) from that in 33°C air, and yet, f_h in He-O_2 manifested a definite decrease. Since cardiac output ($\dot{Q}$) can be represented by:

$$\dot{Q} = f_h \cdot V_s \tag{1}$$

and

$$\dot{Q} = \dot{V}_{O_2} \cdot (Ca_{O_2} - C\bar{v}_{O_2})^{-1} \tag{2}$$

where V_s and (Ca_{O_2} -- $C\bar{v}_{O_2}$) are stroke volume and oxygen content difference between arterial and mixed venous blood, respectively. Combining (1) and (2), we have

$$f_h = \dot{V}_{O_2}\,(V_s)^{-1}\,(Ca_{O_2} - C\bar{v}_{O_2})^{-1} \tag{3}$$

Therefore, at a given $\dot{V}_{O_2}$ and a lowered f_h in He-O_2, the following conditions may exist: *a.* increased stroke volume, *b.* increased arterial-venous oxygen-content difference, or *c.* a combination of *a.* and *b.* These possibilities are currently under investigation in our laboratory.

Our blockade data indicated that combined elevation of vagal and depression of sympathetic influences on the heart may be responsible for the lowered heart rate in He-O_2 as compared to that in air at a given $\dot{V}_{O_2}$. In conjunction with the study of autonomic influences, the possible role of this system in the observed depression of circulating catecholamine concentrations in the anesthetized dog breathing He-O_2 (Raymond et al. 1972) should be investigated further. The f_h-depression effect of normobaric, normoxic helium mixture on the unanesthetized rat is also of significant interest to cardiologists following the discovery (Pifarre et al. 1966), confirmation (Pifarre et al. 1970, Raymond et al. 1972), and denial (Holland, Wolfe, and Kylstra 1973) of the unexpected antiarrhythmic and antifibrillatory effect of helium. The altered autonomic nervous activities during He-O_2 respiration may be a mechanism by which helium effect is mediated. In this context, it is significant that Nicholas, Hart, and Kim (1974) reported an identical rate of turnover of cardiac norepinephrine in rats breathing He-O_2 and those breathing air, precluding a possible effect via sympathetic outflow to the heart; and that Raymond et al. (1972) found an unaltered cardiac sensitivity to exogenous epinephrine in the anesthetized dog, precluding the possible effect via the altered cardiac response to an identical sympathetic outflow to the heart. However, it is possible that the elevated vagal activities in He-O_2 completely mask the effects mediated by sympathetic activities, a possibility which has not been investigated.

Heart rate increased concurrently with $\dot{V}_{O_2}$ elevation at a T_a below the upper critical temperature. Thus, f_h may be higher, lower, or the same in He-O_2 as in air depending on the T_a under consideration (Fig. 4). It follows that comparisons based on T_a without knowledge of $\dot{V}_{O_2}$ may yield erroneous conclusions concerning the existence of a primary effect of helium on the heart, as it may be masked completely or reversed by its indirect thermogenic effect.

The slopes for the linear relationships between f_h and $\dot{V}_{O_2}$ reported here are 6.2 and 4.9 units f_h per unit $\dot{V}_{O_2}$ change in air and He-O_2, respectively. These values are in close agreement with the findings of Rhoades et al. (1967) in a study of the f_h and $\dot{V}_{O_2}$ relationship in both the acutely (3 hours) and chronically (10 days) He-O_2 exposed rat. They reported the slope values of 5.8–5.9 for this relationship. It should be cautioned, however, that this linearity applies only to the $\dot{V}_{O_2}$ range studied.

In conclusion, a lower heart rate is observed in the rat in He-O_2 than that in air at any given $\dot{V}_{O_2}$ within the range studied. The lowered f_h appeared to result from combined elevation of vagal and depressed sympathetic activities in He-O_2.

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EFFECT OF BREATHING HELIUM ON SYMPATHETIC NERVOUS AND CARDIOVASCULAR FUNCTIONS

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Nicholas, T. E., J. L. Hart, and P. A. Kim. 1974. Effect of breathing helium on sympathetic nervous and cardiovascular functions. Undersea Biomed. Res. 1(3):271-280.—Following the suggestion that the antiarrhythmic action of helium is mediated by changes in sympathetic activity, we investigated the effect of He-O_2 (80:20) on sympathetic function. The unanesthetized rat with indwelling cannulae and the pithed rat both responded to norepinephrine (NE), tyramine, and dimethylphenylpiperazinium similarly in He-O_2 (31°C) as in air (26°C). Furthermore, He-O_2 failed to alter the depressor response to a 45° tail-down tilt in either rats or guinea pigs. Likewise, He-O_2 did not alter the response of the cat nictitating membrane to preganglionic stimulation. Finally, we examined the effect of He-O_2 on the turnover of cardiac NE. Following injection of ^{3}H-L-NE, the turnover of NE, as measured by the rate of fall of tritium and the endogenous level of NE, was identical in rats exposed to He-O_2 (31°C) and to air (26°C). We conclude that helium does not affect sympathetic nervous or cardiovascular function in the species studied.

sympathetic nervous system
pressor responses
norepinephrine
cardiac catecholamine turnover
nictitating membrane
thermal neutrality
animals

Although exposure to gas mixtures containing helium (He) may affect the growth rate of mold (Schreiner, Gregoire, and Lawrie 1962), the metamorphosis of fruitfly (Cook 1950), and the respiratory enzymes (South and Cook 1953), the high thermal conductivity of this gas may account for many of the reported effects of helium on mammals at 1 ATA (Leon and Cook 1960; Rhoades, Wright, Hiatt, and Weiss 1967; Schatte, Jordan, Phillips, Clarkson, and Simmons 1973). Therefore, it was of considerable interest when Pifarré, Cox, Jasuja, and Neville (1969) reported that the presence of helium in the inspired gas depressed the incidence of cardiac arrhythmias following ligation of the left coronary circumflex artery in dogs anesthetized with sodium pentobarbital. Subsequently, Pifarré, Raghunath, Vanecko, Chua, Balis, and Neville (1970); Raymond, Weiskopf, Halsey, Goldfein, Eger, and Severinghaus (1972); and Toltzis and Scott (1972) repeated this finding. In contrast, Holland, Wolfe, and Kylstra (1973) failed to find any evidence of antiarrhythmic action of helium in a series of experiments performed in an apparently identical fashion. There seems to be no immediate explanation for this discrepancy, for in most cases, body temperature appears adequately controlled. Finally, Leon and Harris (1973) reported that breathing helium increased the minimal directly applied electrical stimulus to produce ventricular fibrillation in dogs.

An antiarrhythmic action of helium is of two-fold importance. First, since helium is the gas of choice to replace nitrogen (N_2) during deep sea diving, any pharmacological property of this gas is of potential significance. Second, any treatment effective in reducing the incidence of arrhythmias is of possible clinical significance.

The role of the sympathetic nervous system in the induction of cardiac arrhythmias is well documented (Manning and Cotten 1962; Raper and Wale 1969; Gillis 1971). Furthermore, Raymond et al. (1972) have reported that breathing helium depresses the plasma levels of catecholamines in anesthetized dogs. Hence, the sympathetic nervous system appears to be a likely site for a pharmacological action of helium. The purpose of the present research was to explore for possible effects of breathing helium on the sympathetic nervous system.

METHODS AND PROCEDURES

CARDIOVASCULAR EFFECTS (RATS, GUINEA PIGS)

Pressor responses in unanesthetized rats

Indwelling cannulae were implanted in Sprague-Dawley rats (200-300 g) in the following manner. The rat was anesthetized with the ultrashort-acting barbiturate sodium methohexital (Eli Lilly, Ind.) (intraperitoneal: 40 mg/kg) and polyethylene cannulae (Intramedic #50) were placed in the left carotid artery and the right jugular vein. These cannulae were passed under the skin to re-emerge in the dorsal-cervical region, where they were threaded through a tightly coiled steel spring (6 inches x 1/8 inch), the end of which was implanted under the skin. The rat, complete with cannulae and protecting spring, was placed in a black Plexiglas container (6-inch diameter x 6-inch height), which in turn was immersed in a temperature-controlled water bath (Haake type FE circulator). Gases were introduced through the bottom of the container via a submerged Tygon coil, to exit through a tube at the top of the container. The cannulae and protecting spring also emerged through this latter tube, so that upon awakening the rat could move about the container while being lightly restrained by the spring. The ambient temperature of the container was routinely checked during calibration of the system. All drugs were dissolved in 0.9% NaCl solution and injected as 0.025-0.05 ml volumes. Although dose-effect curves had been constructed previously, the submaximal dose of pressor drug used was verified for each rat. Each dose of drug was flushed from the cannula with 0.03 ml of 0.9% NaCl solution. The rats were allowed to recover for at least 2 hours following surgery, or until the mean arterial pressure and heart rate had stabilized. Arterial pressure was monitored with a Grass Model 7B Polygraph via a Statham transducer (P 23AC). Following recovery, 100 ng doses of norepinephrine (NE) were injected until the pressor response was constant, following which NE tyramine (Tyr) and dimethylphenylpiperazinium (DMPP) were injected in the form of a 3 x 3 Latin square. The rat was equilibrated for at least 30 min following introduction of a new gas before the pressor agents were again administered.

Pressor responses in pithed rats

The rat was anesthetized and carotid artery and jugular vein cannulae inserted as above. The trachea was cannulated with a glass T-piece connected to a humidifier and a Harvard

Rodent Respirator (model 680) and, finally, the rat was pithed by the orbital approach of Shipley and Tilden (1947). The rat was respired 60 times/min with a tidal volume of 2.5 ml and an end expiratory pressure of 2 cm H_2O. The body temperature was maintained at 37°C by an infrared lamp and heating pad controlled by the feedback from a rectal probe to a Thermistemp Temperature Controller (Yellow Springs Instrument Co., Inc.). This heating system was used in all experiments involving pithed or anesthetized animals.

Depressor response to tilt in rats and guinea pigs

Rats and guinea pigs used for tail-down tilt experiments were anesthetized with urethane (intraperitoneal: 1.2 g/kg) and a carotid artery cannula inserted. Again, the trachea was cannulated with a glass T-piece through which the desired gas could be circulated; the animal was allowed to breathe spontaneously. The animal was strapped to a tilt board in the supine position and the arterial cannula was connected to a Statham transducer fastened to the board at heart level. The board was tilted manually and every attempt was made to standardize the rate of tilt. The animal was maintained in the 45° tail-down position until the blood pressure stabilized or commenced to compensate and the maximum depressor response was recorded.

NICTITATING MEMBRANE PREPARATION (CATS)

The nictitating membrane-superior cervical ganglion preparation was set up according to the method of Paton and Perry (1953), modified so that the nictitating membrane was connected to a Grass force-displacement transducer (model FT .03), which was connected to a Grass Model 7B Polygraph. A Grass Stimulator Model S6 was used. The trachea was cannulated with a glass T-piece and the cat respired with a Harvard model 607 respirator.

PLASMA CATECHOLAMINES (RABBITS)

Plasma catecholamine levels were monitored by superfusing blood from the animal over a rat fundic strip, according to the method of Vane (1957). The fundic strip was removed from the rat and placed in a gut bath partially submerged in a water bath maintained at 37 ± 0.05°C. Initially, the tissue was superfused with Krebs bicarbonate solution (in mM: NaCl 118, KCl 4.75, KH_2PO_4 1.19, $MgSO_4$ 1.19, $CaCl_2$ 2.54, $NaHCO_3$ 25 and glucose 5), equilibrated with 95% O_2-5% CO_2 at 37°C. The rabbit was anesthetized with urethane (intraperitoneal: 1.4 g/kg); a carotid artery, a femoral artery, and a jugular vein were cannulated with polyethylene tubing (Intramedic #60) and 3000 units of heparin injected intravenously. The trachea was cannulated with a glass T-piece through which the desired humidified gas could be circulated while the rabbit breathed spontaneously. Blood was withdrawn from the carotid artery through one chamber of the Holter roller pump (model P0001) and cascaded at 10 ml/min over the fundic strip, to collect at the bottom of the gut bath. From there it was withdrawn through a second chamber of the pump and infused back into the rabbit via the jugular vein. The tension of the fundic strip was monitored with a Grass force-displacement transducer (model FT .03) connected to a Grass Model 7B Polygraph. In each experiment, the sensitivity of the fundic strip was tested by infusing known concentrations of epinephrine into the perfusing medium. During superfusion with Krebs bicarbonate solution, a concentration of epinephrine as low as 0.5 ng/ml was sufficient to induce a consistent relaxation; however, during superfusion with blood the sensitivity decreased to approximately 1.5 ng/ml.

TURNOVER OF CARDIAC NE (RATS)

The cardiac turnover of NE was determined by injecting 33.3 Ci/kg of ^{3}H-L-NE (Amersham/Searle, Ill.), specific activity 9.2 Ci/mmol into a caudal vein of male Sprague-Dawley rats of 150-200 g. The purity of the ^{3}H-L-NE was checked using the thin layer chromatographic method of Fleming and Clark (1970). Twenty of these rats were placed in a temperature-controlled unit (Hot Pack, Philadelphia) with an internal volume reduced to 12 ft^3, maintained at 31 ± 0.4°C and ventilated with He-O_2 (80:20) at 3 liters/min. Another group of 20 rats was placed in a Plexiglas container of comparable volume, through which air was circulated at 3 liters/min at 26 ± 0.6°C. The level of CO_2 was continually monitored and an attempt was made to standardize lighting conditions. At various times following the injection of ^{3}H-L-NE, rats were removed from the containers, killed by cervical dislocation, the hearts weighed and then homogenized in 0.4 M perchloric acid in a glass homogenizer at 4°C. Duplicates of the homogenate (0.2 ml) were removed for counting in a Packard Tri-Carb Liquid Scintillation Spectrometer in a solution of Aquasol (New England Nuclear, MA). Quenching was determined by using internal standards. The homogenate was then centrifuged at 10,000 g for 30 min at 4°C, the supernatant removed, and the levels of endogenous NE determined by the method of Anton and Sayre (1962). Results were corrected for a recovery of 72 ± 1.2% (mean ±S.E.).

Drugs used in the following experiments were obtained from commercial sources. Norepinephrine solution was prepared freshly each day in 0.9% NaCl containing 0.05% ascorbic acid and kept at 3°C until immediately prior to injection, when it was warmed to 37°C.

RESULTS

Changing the gas environment from air to He-O_2 (80:20) at room temperature (23°C) significantly and reversibly depressed the pressor response to 100 ng NE ($P < 0.001$), did not alter the mean arterial pressure, and reversibly increased heart rate ($P < 0.001$) in unanesthetized, lightly restrained rats (Table 1). However, if the ambient temperature was decreased from 26°C (thermal neutral) to 18°C in air, a similar pattern emerged (Table 2). In contrast, if thermoneutral temperatures were observed, no differences emerged in pressor

TABLE 1

Effect of a helium-oxygen (80:20) environment at 23°C on mean arterial pressure, heart rate, and pressor response to norepinephrine of unanesthetized, lightly restrained rats

	Air	He-O_2	Air	*N*
Mean arterial pressure (mm Hg)	123 ± 5.1	128 ± 4.8	122 ± 4.6	8
Heart rate/minute	322 ± 6.5	438 ± 18.1 ***	305 ± 11.2	8
Response to 100 ng	46 ± 2.8	34 ± 1.6 ***	42 ± 2.0	8
Norepinephrine (mm Hg)				

Significantly different from initial value in air: *** $P < 0.001$. *N* is the number of rats. Results expressed as mean ± S.E. Data were subjected to a paired *t* test.

TABLE 2

Effect of ambient air temperature on the mean arterial pressure, heart rate, and pressor response to norepinephrine of unanesthetized, lightly restrained rats

	26°C	18°C	26°C	N
Mean arterial pressure (mm Hg)	130 ± 3.4	139 ± 2.8 **	137 ± 4.0	11
Heart rate/minute	365 ± 23.5	413 ± 24.3 *	352 ± 21.7	5
Response to 100 ng Norepinephrine (mm Hg)	47 ± 2.6	38 ± 2.3 ***	46 ± 2.1	11

Significantly different from initial value at 26°C: * $P < 0.05$, ** $P < 0.01$; *** $P < 0.001$. N is the number of rats. Results expressed as mean ± S.E. Data were subjected to paired *t* test.

responses to NE, Tyr, and DMPP, mean arterial pressure, and heart rate between rats in air (at 26°C) and rats in He-O_2 (at 31°C) (Table 3). Similarly, in a pithed rat maintained at a rectal temperature of 37°C, paired *t* tests revealed no significant differences in pressor responses, mean arterial pressure, and heart rate between rats breathing air or He-O_2 (Table 4). Likewise, no significant differences in depressor responses elicited by subjecting rats and guinea pigs to tail-down tilting emerged between animals breathing air or He-O_2 (Table 5).

We next examined the effects of He-O_2 on the contractile response of the cat nictitating membrane to presuperior cervical ganglion stimulation (pulse duration 0.7 msec, stimulus strength 4 volts, period of stimulation 10 sec, and frequency of stimulation 4/sec). In these experiments the protocol was arranged so that initially the animal was respired with either air or He-O_2 and, after at least eight responses, the gas was changed for the other mixture. The cat was allowed to equilibrate for at least 30 min following introduction of the new gas. The mean of the eight responses was determined on each gas mixture in a total of five cats. During air breathing the contraction was 724 ± 51.9 mg (mean ± S.E.) and during He-O_2 breathing the contraction was 770 ± 51.5 mg. Although the response of the nictitating

TABLE 3

Effect of a helium-oxygen (80:20) environment at thermoneutral temperature on mean arterial pressure, heart rate, and responses to pressor agents of unanesthetized, lightly restrained rats

	Air 26°C	He-O_2 31°C	Air 26°C
Mean arterial pressure (mm Hg)	140 ± 4.6 (14)	141 ± 4.8 (14)	137 ± 4.4 (11)
Heart rate/minute	344 ± 5.7 (8)	342 ± 7.6 (8)	353 ± 7.8 (8)
Pressor response (mm Hg):			
norepinephrine (100 ng)	42 ± 2.8 (14)	41 ± 1.9 (14)	43 ± 2.6 (11)
tyramine (60 μg)	28 ± 3.1 (10)	27 ± 2.2 (10)	29 ± 3.9 (6)
DMPP (10 μg)	42 ± 4.3 (8)	44 ± 2.6 (8)	42 ± 3.1 (6)

Results expressed as mean ± S.E. No differences emerged when data were subjected to paired *t* test.

TABLE 4

Effect of breathing helium-oxygen (80:20) on mean arterial pressure, heart rate, and responses to pressor agents of pithed rats

	Air 26°C	He-O_2 31°C	N
Mean arterial pressure (mm Hg)	60 ± 3.9	63 ± 4.8	5
Heart rate/minute	370 ± 21.5	377 ± 26.4	5
Pressor responses (mm Hg)			5
norepinephrine (50 ng)	40 ± 3.3	47 ± 4.7	
tyramine (15 μg)	31 ± 2.7	33 ± 2.5	
DMPP (5 μg)	43 ± 2.2	45 ± 4.5	

Results expressed as mean ± S.E. No differences emerged when data were subjected to paired *t* test.

TABLE 5

Effect of breathing helium-oxygen (80:20) on the depressor response to a 45° tail-down tilt in anesthetized rats and guinea pigs

	Air	He-O_2	N
Rats	15.4 ± 1.9	15.1 ± 1.5	9
Guinea pigs	12.9 ± 1.4	13.9 ± 1.6	6

Results expressed in mm Hg as mean ± S.E. No differences emerged when the data were subjected to paired *t* test.

membrane to preganglionic stimulation varied greatly between cats, the response in the same cat was remarkably constant regardless of whether the animal was breathing air or He-O_2. This constancy was reflected in the lack of significant difference revealed by a paired *t* test.

Figure 1 is a tracing from one of five experiments in which a superfusion assay was used to monitor plasma levels of catecholamines in a rabbit breathing either air or He-O_2. Since

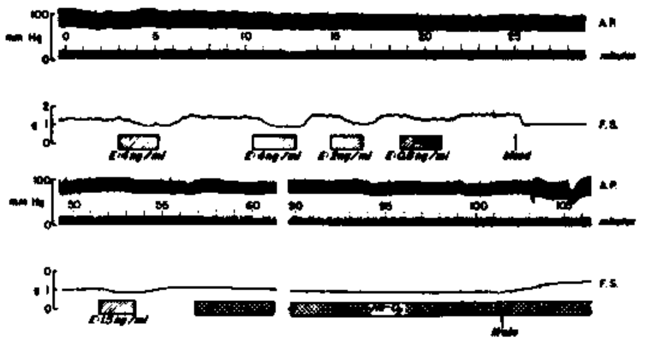

Fig. 1. The effect of breathing He-O_2 (80:20) on the plasma catecholamine levels of an anesthetized rabbit, as monitored by the tension (grams:g) of a superfused fundic strip (F.S.) from a rat. Epinephrine (E) was infused into the perfusion circuit immediately prior to the fundic strip, as a test of the sensitivity of the tissue to catecholamines. A.P. is the arterial pressure.

catecholamines relax the rat fundic strip, a decrease in plasma catecholamine levels should be reflected in a contraction of the tissue and an increase in baseline; this was not observed, even after breathing He-O_2 for 1 hour. The fall in baseline following the transition from Krebs bicarbonate solution to blood probably reflects the levels of endogenous catecholamines in blood, a change in P_{O_2}, or a change in the ionic composition of the superfusate. In all preparations, there was a small depression of baseline over the period of the experiment (50-300 mg in 5 experiments); however, when Krebs bicarbonate solution again replaced blood as the superfusate the baseline inevitably increased, thus confirming the viability of the tissue.

The decline of radioactivity in 40 rat hearts over the 23 hours following injection of ^{3}H-L-NE is illustrated in Fig. 2. As there were no significant differences between the level of radioactivity in the hearts of the remaining 15 rats maintained in air (at 26°C) and 15 rats maintained in He-O_2 (at 31°C) at 6, 12, and 23 hours following injection, a single curve has been constructed. In addition, because there was no difference in the levels of endogenous NE between the two groups, the values at 6, 12, and 23 hours have been combined and are also presented in Fig. 2.

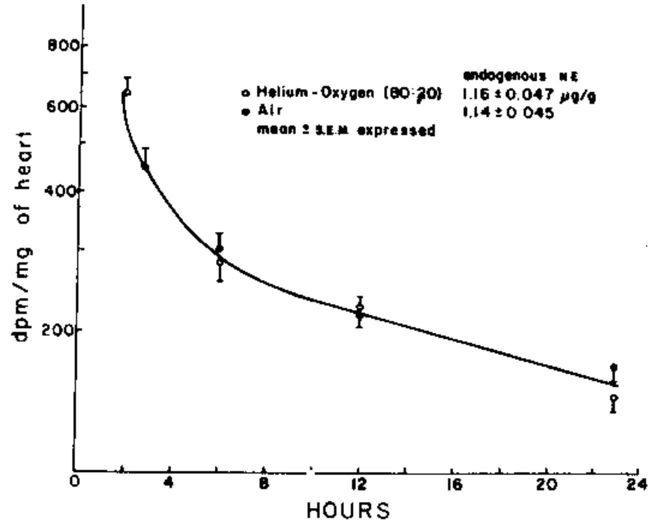

Fig. 2. A plot of disintegrations per minute per mg of heart (as a log function) versus time in hours. Rats were injected intravenously with 33.3 µCi/kg ^{3}H-L-NE and the tritium content and endogenous levels of NE determined at various times following. One group of 20 rats was maintained at 31°C in an environment of He-O_2, while the other group of 20 rats was maintained at 26°C in air.

DISCUSSION

Our present experiments lend no credence to the hypothesis that breathing helium mixtures affects the sympathetic nervous system, or more particularly, the peripheral sympathetic system—even when the highest possible percent of helium was used (He-O_2 80:20). Whenever possible we have utilized unanesthetized animals to avoid the complication of anesthetics and, hopefully, to create a more natural physiological situation. Raymond et al. (1972) mentioned that breathing helium did not alter the response of mean blood pressure to injected epinephrine in anesthetized dogs, but he failed to provide data to substantiate this statement. We have extensively examined the cardiovascular response of rats to the sympathetic postganglionic transmitter, NE, to evaluate whether helium alters receptor sensitivity or possibly the NE re-uptake mechanism. In addition, we examined the effect of helium on the pressor response to the indirectly acting amine tyramine as an index of the readily releasable stores of NE. Finally, we determined whether helium affected the pressor response elicited by the ganglion-stimulating agent, DMPP, in case the gas interfered with the nicotinic receptors in the ganglion. At thermoneutral temperatures, which had been determined in our laboratory and also by Clarkson, Schatte, and Jordan (1972), we could find no evidence that breathing helium altered the response to these three agents. This experiment was repeated in pithed rats, which are more sensitive to pressor agents and have very stable arterial pressures. Once again, helium failed to alter the responses to NE, Tyr, and DMPP. The importance of thermoneutral temperatures cannot be overemphasized, as illustrated by our findings in the different gases at room temperature and the effects of ambient temperature of air. The reversible depression of response to NE found either when He-O_2 replaced air at room temperature or when the ambient temperature of air was decreased possibly reflects an increase in the endogenous levels of plasma catecholamines (Leduc 1961) and a subsequent tolerance to the amine. This depressed reactivity to NE is distinct from the increased sensitivity found in more prolonged adaption to cold (Leblanc 1960).

The lack of effect of helium on the postganglionic sympathetic fiber and effector cells was confirmed using the cat superior cervical ganglion-nictitating membrane preparation as a model. Use of this preparation would reveal also any alteration in ganglionic transmission attributable to breathing helium: no differences emerged. The possibility still existed that breathing helium altered autonomic reflexes, but because of the difficulties in testing this in unanesthetized animals we had to resort to examining the cardiovascular response of anesthetized rats to tail-down tilt. Although these experiments clearly revealed no differences in the rate of compensation between rats breathing He-O_2 and air, the possibility remains that an effect was masked by the presence of the anesthetic.

An action of helium on the sympathetic system possibly would be reflected in a change in either heart rate or arterial pressure. Surprisingly, neither Pifarré et al. (1969, 1970) nor Holland et al. (1973) mentioned these important variables; however, Raymond et al. (1972) and Leon and Harris (1973) showed that there was no difference in mean arterial pressure between anesthetized dogs breathing He-O_2 and another group breathing air, but any small difference would have been masked by the large variation between dogs. Raymond et al. (1972) also reported that breathing He-O_2 resulted in a decrease in heart rate of a few beats per minute; this was not confirmed by Leon and Harris (1973). Our results clearly demonstrate that respiring helium had no effect on either heart rate or mean arterial pressure when the same rat was used as its own control; however, the minute-to-minute variation would have precluded detecting changes of only a few beats. Although Lin (1973) had reported that at the same oxygen consumption rats breathing He-O_2 had lower heart rates than those breathing air, examination of his curves revealed that one would not expect to see different heart rates under the conditions we have used (air at 26°C and He-O_2 at 31°C).

Using the superfused rat fundic strip, we were unable to demonstrate a change in plasma catecholamines attributable to breathing helium; however, a change of below 1 ng/ml would have escaped detection in our preparation. The possibility also exists that a fall in plasma catecholamine levels was masked by a concomitant fall in the baseline tension of the fundic strip due to the preparation failing; however, the baseline fell similarly whether the rabbit was breathing He-O_2 or air. Alternatively, a contraction may have been masked by a fall in the plasma levels of serotonin or angiotensin, both of which contract a fundic strip. The fall in plasma levels of epinephrine and norepinephrine reported by Raymond et al. (1972) as consequent of helium breathing is far from convincing, for the plasma levels continued to fall in four of the six dogs when air breathing was resumed. In only one dog did the plasma catecholamines change in a manner to suggest that helium in fact caused a depression. Recently Raymond, Sode, Langworthy, Blosser, and Johnsonbaugh (1974) reported that breathing He-O_2 failed to influence either plasma or urinary catecholamines of humans. Any action helium may have to inhibit either dopamine-β-hydroxylase (as suggested by these workers), or any other enzyme involved in the biosynthesis of NE would have become apparent in our study of the turnover of cardiac catecholamines in unanesthetized rats at thermoneutral temperatures: there was no difference in the rate of turnover between rats in a He-O_2 environment and those in air. A criticism of our method could be our failure to measure helium levels during this experiment; however, we feel that a flow rate of 3 liters of He-O_2 (80:20) was excessive for twenty 150-g rats.

In conclusion, we could find no evidence that breathing helium affected sympathetic nervous or cardiovascular functions in the species tested. The possibility remains that the enhanced sensitivity of the infarcted heart to the arrhythmic action of catecholamines (Maling and Moran 1957) is depressed by helium; this has not yet been tested.

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INABILITY OF HELIUM TO INFLUENCE THE OCCURRENCE OF ARRHYTHMIAS IN CATS, RATS AND MICE

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ABSTRACT

In view of recent reports that breathing helium-oxygen (He-O_2) mixtures tended to inhibit the occurrence of arrhythmias and ventricular fibrillation following left circumflex coronary artery occlusion in anesthetized dogs, we have examined the effects of helium on the occurrence of arrhythmias induced pharmacologically. The total intravenous dose of ouabain required to induce continuous arrhythmias in anesthetized rats, or to flatten the EKG in anesthetized mice, was identical whether the animal was breathing He-O_2 (80-20) or air. Similarly, the presence of He in the inspired air did not alter either the intravenous dose of deslanoside required to induce continuous arrhythmias, or the duration of arrhythmias induced by intravenous epinephrine, in anesthetized cats. In light of these negative results, we examined finally whether respiring He would affect the occurrence of arrhythmias following left coronary artery occlusion in anesthetized cats: we found that He affected neither the frequency of arrhythmias nor the incidence of ventricular fibrillation. In conclusion, we could find no evidence that He possessed antiarrhythmic properties in the species studied.

Key words: Helium, pharmacological arrhythmias, ouabain, deslanoside, epinephrine, ventricular fibrillation.

INTRODUCTION

Because helium at one atmosphere is generally regarded as devoid of pharmacological activity and many of the earlier reports to the contrary were possibly consequent of the high thermal conductivity of the gas (Leon and Cook, 1960), it was of considerable interest when Pifarre et al., (1969) reported that breathing helium reduced the incidence of cardiac arrhythmias induced in anesthetized dogs by left circumflex coronary artery ligation. This observation was repeated by Raymond et al. (1974), who further suggested that the action may involve a change in sympathetic activity. In contrast, Holland et al. (1973) could find no evidence of an antiarrhythmic action attributable to He, in apparently identical experiments.

Such an action of He would be important, first, because of the possible clinical application and, second, because any demonstrable physiological action of breathing He would have ramifications in deep sea diving, where He is the gas of choice to replace nitrogen in the respired gas.

With one exception, the action of He has been explored only in arrhythmias induced in dogs by ligation of the left coronary circumflex artery: Leon and Harris (1973) have reported that breathing He increased the minimal electrical current required to induce fibrillation when directly applied to the right ventricle of dogs. In view of the well documented involvement of the sympathetic nervous system in induction of arrhythmias (Manning and Cotten, 1962; Raper and Wale, 1969) and the suggestion by Raymond et al. (1972) that the action of He be mediated by the sympathetic nervous system, we have examined the effect of He on arrhythmia induction following infusion of epinephrine, deslanoside and ouabain; these two cardiac glycosides appear to exert their arrhythmic action via the sympathetic system (Gillis et al., 1972). In addition, we wished to ascertain whether He could influence the course of arrhythmias induced by coronary artery occlusion in a species other than the dog.

METHODS

Ouabain induced arrhythmias in rats and mice

The toxic dose of ouabain was determined in animals anesthetized with urethane (intraperitoneal: 1.2 g/kg) and breathing spontaneously through a T-shaped glass tracheal cannula, through which the desired, humidified gas mixture was circulated. Ouabain was infused through a cannula attached to a No. 28 gage needle inserted into a lateral caudal vein. EKG was monitored via standard lead II connected to a model 7B Grass Polygraph. In rats, the arterial pressure was monitored (Statham transducer P23 AC). Body temperature was maintained at 37°C by the feedback from a rectal probe to a Thermistemp temperature controller (Yellow Springs Instrument Company, Inc.), which in turn controlled an infrared lamp and a heating pad. The endpoint of infusion in mice was the appearance of a flat EKG (Small, et al., 1971), whereas the endpoint in rats was the induction of continuous arrhythmias. The dose of ouabain required is expressed as µg/g of body weight. The animals breathed the gas for at least 30 minutes before ouabain infusion commenced.

Induction of arrhythmias in cats

The cat was anesthetized with sodium pentobarbital (intraperitoneal: 30 mg/kg) and a femoral artery and vein cannulated (polyethylene intramedic No. 50) for the measurement of arterial pressure and the injection of drugs, respectively. Pressure was monitored by a Statham transducer (PAC 23) connected to a Grass model 7B Polygraph. The trachea was cannulated with a glass T-piece connected to a Harvard model 607 respirator and a humidifier. Animals were respired at 20 breaths/minute, each of 12 ml/kg. A standard lead II EKG was used to monitor cardiac events. Body temperature was maintained at 37°C as in first method above.

Epinephrine arrhythmias were induced by rapid intravenous injection of epinephrine (50 µg/kg of a solution containing 500 µg/ml), at 30 minute intervals throughout the experiment. At least two injections were administered while the cat was breathing air, and then, fifteen minutes before the next injection of epinephrine, the cat was switched to breathing $He-O_2$ (80:20). Following two additional injections of E, the respired gas reverted to air. The duration of the arrhythmias was determined from the EKG tracing.

Deslanoside arrhythmias were induced by injections of deslanoside (100 µg/ml; 25 µg/kg) every fifteen minutes until continuous ventricular arrhythmias of at least 10 minutes duration occurred. The total dose of deslanoside required to induce this condition was then determined.

Coronary artery occlusion was performed by making a mediosternal incision, separating the pericardium and then rotating the heart until the left coronary artery could be isolated. The cat was ventilated with He-O_2 for at least 30 minutes prior to occlusion of the artery with a small clamp.

Drugs used in the following experiments were obtained from commercial sources.

RESULTS

Table 1 demonstrates that breathing He had no effect either on the heart rate of anesthetized rats or mice, or on the mean arterial pressure of rats. In addition, the total dose of ouabain required to induce continuous arrhythmias in rats or to flatten the EKG tracing in mice, was not altered whether the animals were breathing air or He-O_2. Likewise, breathing He-O_2 did not significantly alter the heart rate of mean arterial pressure of anesthetized cats (Table 2). The table also shows that the same dose of deslanoside was required to induce continuous arrhythmias whether the cat was breathing air or He-O_2. One rat examined demonstrated spontaneous arrhythmias characteristic of a two-, or three-to-one atrio-ventricular block. Changing from the air to He-O_2 breathing had no effect on the frequency of these arrhythmias.

		Heart rate per minute	M.A.P. mm Hg	Total dose of ouabain (µg/g)	Duration of infusion (minutes)	N
Rats	air	344 ± 16	97 ± 5.6	12.7 ± 0.63	18.2 ± 1.9	16
	He-O_2	305 ± 15	92 ± 5.2	12.3 ± 1.20	18.6 ± 1.2	16
Mice	air	494 ± 44		1.6 ± 0.24	6.8 ± 1.3	5
	He-O_2	478 ± 32		1.4 ± 0.12	6.6 ± 0.8	5

Table 1. Effect of breathing He-O_2 (80:20) on the toxic intravenous dose of ouabain in rats and mice anesthetized with urethane.

Results expressed as mean ± S.E. N is the number of animals. The heart rate and mean arterial pressure (M.A.P.) were measured immediately prior to infusion of ouabain.

Gas	Sex	Weight	Heart Rate	Mean arterial B.P.	Dose of Deslanoside
N_2O_2	3M,3F	3.2 ± 0.3 kg	197 ± 7.7	125 ± 11.2	150 ± 6.5 µg/kg
HeO_2	4M,3F	3.0 ± 0.2 kg	203 ± 8.8	112 ± 11.5	158 ± 10.5 µg/kg
			beats/min.	mm of Hg	
Value of t:			0.617	0.821	1.227

Table 2. Effect of breathing helium-oxygen (80:20) on the dose of deslanoside required to induce continuous cardiac arrhythmias in cats anesthetized with pentobarbital.

Results expressed as mean ± SEM. M: male, F: female. Heart rate and mean arterial pressure were measured immediately prior to the first injection of deslanoside.

Table 3 reveals that there was no difference in the duration of arrhythmias induced by injecting epinephrine, whether the cat was breathing air or He-O_2. Even though only four animals were used in this experiment, each acted as its own control, so that paired t-tests could be applied to the data.

Cat No.	Weight	Sex	Duration of arrhythmias (seconds)				
			N_2O_2		HeO_2		N_2O_2
1	5.0 kg	M	225	156	155	195	208
2	4.8	F	130	128	140	132	147
3	4.4	F	252	238	281	234	245
4	4.1	M	139	175	161	157	177
Mean ± SEM			186 ±31	174 ±23	184 ±33	180 ±22	194 ±21
Paired t-tests:					0.254		1.332

Table 3. Duration of epinephrine induced arrhythmias in anesthetized cats during nitrogen-oxygen (80:20) and helium-oxygen (80:20).

We also attempted to use each cat as its own control in experiments involving ligation of the left coronary artery, however, the great variability in the time taken for ventricular fibrillation made this difficult (Table 4). Finally, we resorted to having one group of cats breathe air and another group breathe He-O_2 and compared both the incidence of ventricular fibrillation and the frequency of arrhythmias in the two groups following ligation of the left coronary artery. Following coronary artery constriction, we consistently observed a fall in both mean arterial pressure and heart rate. Table 5 shows that breathing He had no effect on either the incidence of ventricular fibrillation or the frequency of arrhythmias.

	Cat No. 2			Cat No. 4		
Gas	Period of exposure to gas	Duration of occlusion for VF		Period of exposure to gas	Duration of occlusion for VF	
N_2O_2		2 min.	5 sec.		10 min.	55 sec.
		2	0		5	5
		2	17		6	20
					16	30
					8	40
					1	35
					1	37
					1	40
HeO_2	10 min.	2	26	4 min.	0	45
	23	1	55	17	0	25
	35	3	0	23	1	15
	56	2	0	30	0	55
N_2O_2	7	1	45	5	0	50
	16	1	48	11	0	45
	32	1	52	17	0	45
				27	0	55

Table 4. Effect of breathing helium-oxygen (80:20) on the rate of induction of ventricular fibrillation in cats following coronary artery occlusion.

Gas	Sex	Weight kg	Incidence of V.F.	Time for V.F. (min.)	Arrhythmias per hour
N_2O_2	7M, 3F	3.1 ± 0.4	60%	7.5 ± 2.4	34 ± 15.2
HeO_2	3M, 6F	3.2 ± 0.4	56%	7.5 ± 4.1	65 ± 11.5

Table 5. Effect of breathing helium-oxygen (80:20) on the occurrence of arrhythmias in cats following left coronary artery occlusion.

Mean ± SEM V.F.: ventricular fibrillation

DISCUSSION

Our present results strongly support the conclusion of Holland et al. (1973) that the presence of He in the inspired gas does not affect the induction of arrhythmias due to coronary artery occlusion; in addition, we have demonstrated that breathing He does not influence the induction of arrhythmias due to cardiac glycosides or epinephrine. Our present research is open to criticism on two main counts. First, anesthetized animals were used throughout. A more physiological approach may be to use animals having spontaneous arrhythmias, such as the one which we have reported. Unfortunately, it is impossible to draw conclusions from results with one rat, but it would probably be feasible to screen a rat colony for animals with spontaneous arrhythmias. A second criticism relates to the use of open-chest animals, which is far removed from a physiological situation and the results gained must reflect to a large extent the surgical expertise of the experimentors. A better approach appears to be the use of a Swan-Ganz catheter to occlude the coronary artery, without thoracic surgery (Toltzis et al., 1974). Unfortunately, due to the size of the cat, we found it impossible to approach the heart via an intercostal space and had to resort to a mediosternal incision, which is far more traumatic an operation. This, or the fact that we used cats rather than dogs, could possibly account for our inability to show an anti-arrhythmic action of He, however, it does not explain why Holland et al. (1973) arrived at the same conclusion. The pattern of events which we observed to follow coronary artery occlusion, closely resembled those reported by Gillis (1971); these include a consistent fall in both mean arterial pressure and heart rate.

Breathing He-O_2 clearly did not affect the arrhythmogenic action of either the cardiac glycosides, or epinephrine, agents which appear to exert their effect largely via an action on the sympathetic nervous system. This finding tends to confirm our previous findings (manuscript accepted for publication by Undersea Biomedical Research) that breathing mixtures containing He failed to influence either the autonomic nervous or cardiovascular systems. In experiments involving the toxic effect of ouabain in mice, we used cessation of electrical activity as our endpoint, for it occurred soon after the onset of electrical disturbances and was far more consistent an indicator. The considerably larger dose of ouabain required for a toxic action in rats than required in mice, is consistent with the finding that rats are particularly resistant to the Na, K-ATPase inhibiting action of ouabain (Repke, et al., 1965). Our results confirm those of Small, et al. (1971) who found that hyperbaric He did not alter the toxicity of cardiac glycosides in rats, guinea pigs, and cats.

From our experiments the possibility remains that the "antiarrhythmic action" of He is specific for arrhythmias induced by left circumflex coronary artery occlusion in dogs; this is currently being investigated in our department.

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chapter 5

PSYCHOSENSORY FUNCTION

To a physiologist, depressed performance might be anything from dyspnea to decreased cardiac output and would be described in terms of systems or symptomology. However, the proper integrative finctions of the CNS constitute a special kind of performance which is also affected by the hyperbaric environment. Therefore, we include papers within from the point of view of the psychologist. The data include sensory and cognitive function as well as investigations of adaptive phenomena.

This volume is by no means exhaustive, nor does it imply that we feel we have answers to the scientific questions addressed to the hyperbaric condition. As this is written, further experiments are being performed; new questions being asked. .We regard this compilation not as a final report of a research effort, but rather as a beginning.

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PERFORMANCE DECREMENTS UNDER HYPERBARIC He-O_2

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ABSTRACT

Six subjects participated in a saturation chamber dive to 16.1 ATA, to investigate the effects of hyperbaric He-O_2 on human performance. Five of the six subjects had no previous saturation experience. Systematic behavioral assessment disclosed performance decrements directly related to degree of task complexity. Significant reductions in long-term memory and cognitive function were noted. It was postulated that increased arousal enhanced the possibility of helium-induced narcosis while improving effectiveness on psychomotor and reaction-time tasks. The results indicate that, unless subjects are adapted to the environment and familiar with required tasks, performance under hyperbaric He-O_2 may be significantly impaired.

Key words: Behavioral assessment, diving, He-O_2, human performance, hyperbaric, narcosis, saturation.

INTRODUCTION

It is generally recognized that the narcotic or intoxicating effect attendant with breathing inert gases under pressure is a primary cause of observable behavioral decrements in psychomotor and cognitive performance during exposure to compressed air beyond 4 ATA. While there exists some evidence of narcosis with He-O_2 at pressures as low as 7 ATA (Baddeley and Flemming, 1967), most investigators have reported no appreciable performance loss with He-O_2 at pressures to 36 ATA.

However, since many studies have concentrated on physiological and technological considerations as new limits for depth and endurance were established, psychological investigations have been relatively restricted, and have failed to provide for systematic interpretation or adequate standardization essential for cross-study comparisons. Additionally, the necessity of employing highly experienced divers on these pioneer ventures may limit the ability to generalize from published findings.

In an attempt to clarify the functional effects of helium narcosis, the present study sought to apply systematic behavioral methodology to the analysis of performance under He-O_2. Based on observations with hyperbaric air, a direct relationship exists between level of task complexity and degree of performance impairment (e.g., Baddeley et al., 1968; Kiessling and Maag, 1962). There is less evidence that increased levels of helium narcosis are similarly related to performance loss, but recent use of sensitive behavior schedules with lower animals has provided evidence of complex performance loss with He-O_2 at 7 to 10 ATA (Thomas and Bachrach, 1971; Thomas, Walsh and Bachrach, 1971). Loss of complex functioning is consistent with the assumption that narcosis selectively interrupts synaptic transmission of the CNS (Albano, 1970; Bennett, 1966).

To assist in comparing diverse measures, a predictive model was adopted which classified various tasks by the degree of neurological complexity thought necessary to maintain adequate performance. As information-processing capability is the functional contribution of a diver to an ocean system, stages of task complexity were superimposed on a crude information processing model, comprised of five arbitrarily-labeled processing stages: sensory transduction, arousal state, motor skills, memory and decision making. Representative tasks from the existing literature were selected for each of five corresponding response categories (sensory, reaction time, psychomotor, memory and cognitive functioning), and tested to determine the degree of performance interruption experienced under He-O_2 saturation conditions.

METHODS

Dive history. A 72 feet long, 3 chamber habitat (the Aegir) was moored at the Makai Range pier (Makapuu Point, Oahu, Hawaii) and remained surfaced throughout the study. Six diver-technician volunteers were selected, having an average of 31.2 years, and a range from 22 to 42. Only one subject had prior saturation experience.

The dive simulation occupied ten days (Figure 1), with initial compression to 8.6 ATA. After 17 hours, the habitat reached a maximum internal pressure of 16.1 ATA. This pressure was maintained for 43 hours, until decompression commenced on Day 5. Control testing occurred on two days prior to the dive, and approximately two weeks following its termination.

Figure 1. Aegir internal pressure by day, measured in atmospheres absolute (ATA).

Sensory testing. Although there is little evidence that inert gas narcosis directly influences sensory functioning (Bieraner, 1972; Elcombe and Teeter, 1973), audiometric testing was initiated on Day 3 (16.1 ATA) to verify pressure-related hearing loss noted with hyperbaric air (Fluur and Adolfson, 1966). Sound level meter (SLM) readings were taken at periodic intervals throughout the dive.

Reaction time. Studies with hyperbaric air have disclosed significant reaction time and speed changes (e.g., Frankenhauser et al., 1963; Kiessling and Maag, 1962). A generalized measure of choice reaction time was selected for the present study, providing sequential two-choice discriminations of visual (blue/yellow) and auditory (high/low) stimuli and requiring appropriate left-right movement of a hand-held switch to terminate the signal; timing was in hundredths of a second. Visual-only, auditory-only, and a combined series provided a total of 100 trials on Days 0, 3, and 7. On days 1 and 2, only the visual series of 25 trials was presented.

Psychomotor. Two measures of voluntary motor coordination were included: the first, the Minnesota Rate of Manipulation Test (MRMT), was selected for its standardized procedures, its factor weightings on gross motor manual dexterity and its prior use underwater (Ono and O'Reilly, 1971). Using only the preferred hand, subjects completed two trials on each of 3 MRMT subtasks (Placing, Turning, Displacing) on Days 3, 7, and 0' (post-test control); time to completion was recorded.

The second test was a measure of fine-motor coordination, the screwplate test, found to be significantly influenced by He-O_2 and open-ocean environments (Baddeley et al., 1968; Baddeley and Flemming, 1967). Obtained scores were total time to shift 16 bolts and nuts from one array to another, number of components dropped, and number of loose nuts (defined as loose 1/4 turn). The screwplate test was performed on Days 0, 2, 4, 7, 8, and 9.

Memory. Studies reporting significant narcosis-related memory loss (e.g., Adolfson, 1965; Biersner, 1972) have disclosed selective differences between two components of memory, with short-term memory (STM) not as affected as consolidation into long-term memory (LTM) (Fowler, 1973). In the present study STM was tested by number recall performance on a three part dichotic listening tape, with 2, 3, and 4 digits presented to each ear. Three scoring techniques were followed: (1) Total correct - any number which had been presented and recalled was scored as correct, irrespective of its position in the recalled sequence; (2) Ear order - responses were scored as correct if they appeared in the proper relative position within each ear series, or half-span; (3) Other systematic strategies - alternative recall strategies that presented a logical response pattern were included, such as "attempted ear order" and "temporal order". Testing was accomplished on Day 0 and 4.

A standard associative memory test (the Picture-Number Test, or Ma-1, from the KIT OF REFERENCE TESTS FOR COGNITIVE FACTORS by French, Ekstrom and Price, 1963), found to be sensitive to N_2O narcosis (Dickson et al., 1971), was included on Days 3 and 0' as a measure of LTM.

Cognitive functioning. In the absence of reliable objective physiological correlates of cortical functioning, use of factorially distinct paper-and-pencil psychological tests, which previously exhibited sensitivity to narcosis (Dickson et al., 1971), were adapted for use in the present study. The following standardized tests, reflecting various cognitive factors, were chosen from the KIT OF REFERENCE TESTS FOR COGNITIVE FACTORS, and administered on the days noted:

Perceptual Speed	P-3 (Identical Pictures)	Days 0,2
Spatial Orientation	S-1 (Card Rotation)	Days 0',2
Flexibility of Closure	Cf-2 (Hidden Patterns)	Days 0,2

Discussion of several ancillary measures, collected as part of the psychological protocol, has been presented elsewhere (O'Reilly, 1973).

RESULTS

Sensory testing. Audiometric testing, initiated on Day 3 at 16.1 ATA in the living chamber with all life support equipment functioning, disclosed a substantial decline in hearing ability at the low frequencies (500 and 1000 Hz), and some improvement above 1000 Hz (Table 1). SLM readings, collected coincident with audiometry testing, identified an appreciable low-frequency noise level in the habitat, even with the principal noise-emitting machinery shut down (Table 2).

Frequencies:	500	1000	2000	3000	4000	6000	Hz
Right Ear	31.7	35.8	-11.7	-15.8	-19.2	-21.7	dB
Left Ear	31.7	35.0	-10.8	-16.7	-20.8	-24.2	dB

Table 1. Group mean difference scores for audiometric testing at 1 and 16.1 ATA ($n = 6$)[a].

[a] A positive score indicates increased threshold shifts to sound at a given frequency. A negative score identifies improved hearing over sea level controls.

Reaction time. A series of Test by Subject Analyses of Variance on choice reaction time performance speeds determined that no significant differences existed for the various retests on the auditory or combined stimulus conditions. However, a possible difference in reaction time to visual stimuli was noted ($F = 2.87$, df = 4, 20, $.20 > p > .05$). Range testing indicated that Day 2 (first testing at 16.1 ATA) reactions were significantly faster than those on Day 3 (second testing at 16.1 ATA).

Total errors (summed from all three stimulus series) were found to increase significantly across replications ($F = 8.86$, df = 2, 10, $.025 > p > .01$), as

	Readings Under Three Conditions[a]		
Meter Scale[b]	All Equipment On	Dehumidifiers Off Scrubbers On	Dehumidifiers Off Scrubbers Off
A	64 dB	62 dB	60 dB
B	85	78+	75
C	105+	98+	95+

Table 2. Sound-level readings in Aegir living area, SLM mounted.

[a] Readings taken at 3:00 p.m. on Day 3, 16.1 ATA.
[b] All octave band filter readings were 70 to 90 dB.

did errors on stimuli presented in the combined condition ($F = 7.65$, df = 2, 10, $.05 > p > .01$). Total errors on Day 7 did not differ from those on Day 3, but combined errors on Day 7 were significantly greater than on Day 3; Day 3 total and combined series errors were significantly higher than control scores.

When all trials with auditory-only stimuli were considered (including trials from the combined series), there was a significant increase in errors noted across replications ($F = 11.28$, df = 2, 10, $.025 > p > .01$); with Day 3 and Day 7 errors significantly greater than sea levels controls, but not different from each other. Similar significance was not reached on trials requiring responses to visual only stimuli. No significant differences were found between the two stimulus modes (visual and auditory) on either speed or accuracy.

Psychomotor. Four of the six subjects completed all three testings of the MRMT. Time to completion (speed) was not significantly altered across testings for all three subtasks; although there was a strong trend toward slower completion times for Placing and Turning on Day 3 over control or Day 7 testings.

The functional screwplate test provided evidence of significant performance improvement across presentations for the four replications completed by all six subjects (Days 0, 2, 7, 9) ($F = 5.30$, df = 3, 15, $.10 > p > .025$) performance on Day 9 was significantly improved over earlier testings.

Significantly more components were lost on Days 2 and 9 than on Days 0 and 7 ($F = 8.54$, df = 1, 15, $.025 > p > .01$). No significant difference was noted on analyzing the frequency of loose screws across replications.

Memory. Analysis failed to disclose significant performance change on the 2-digits and 4-digits series of the dichotic listening test, but identified a significant effect on the 3-digits series ($F = 9.38$, df = 1, 5, $p < .05$), reflecting improved recall on the second half-span across testings. While the various scoring techniques (total correct, ear order, other strategies) differed significantly from each other, none proved to be selectively sensitive to hyperbaric effects, although a significant interaction was found between the latter two techniques and level of He-O$_2$ on the 4-digits series ($F = 15.68$, df = 1, 4, $p < .025$) (Figure 2).

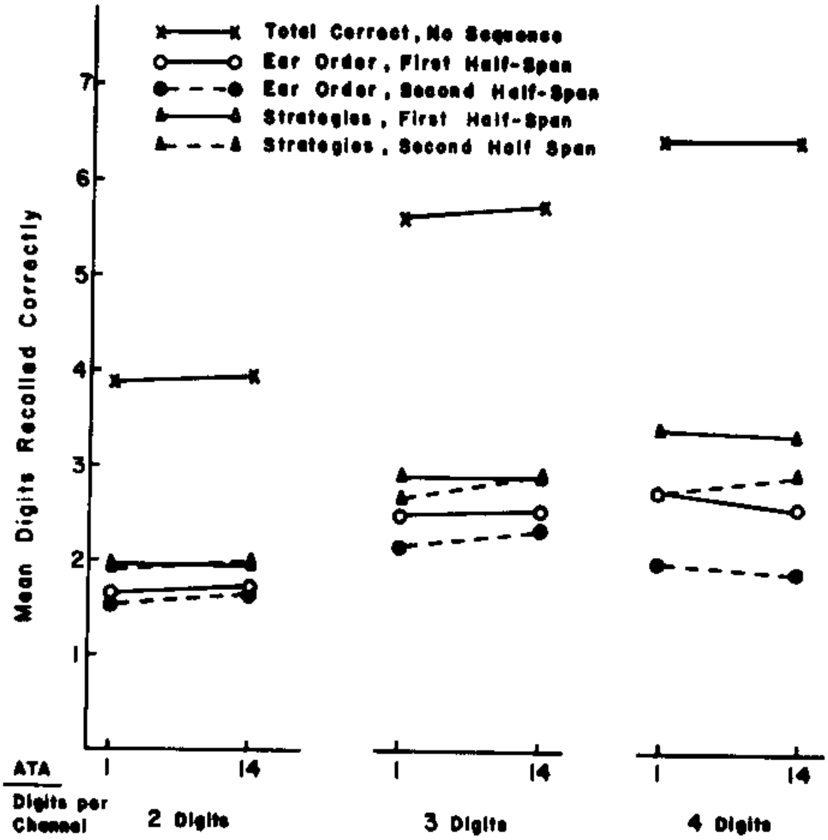

Figure 2. Mean number of digits recalled by half-span and scoring technique over two presentations of the dichotic listening test ($n = 5$).

Associative memory performance on the Ma-1 test demonstrated a significant 25% decrement from sea level control to testing at 16.1 ATA ($t = 2.60$, df = 5 $p < .05$).

<u>Cognitive functioning</u>. Performance under hyperbaric He-O$_2$ conditions was significantly below sea level controls on Perceptual Speed (P-3) ($t = 3.41$, df = 5, $p < .02$), Flexibility of Closure (Cf-2) ($t = 3.64$, df = 4, $p < .05$), and Spatial Orientation (S-1) ($t = 3.06$, df = 5, $p < .05$) (Figure 3).

DISCUSSION

Results of the present study indicate that human performance can be impaired under He-O$_2$ saturation conditions. A significant reduction in performance on relatively complex information processing tasks was observed concurrent with moderate improvement in less complex behaviors.

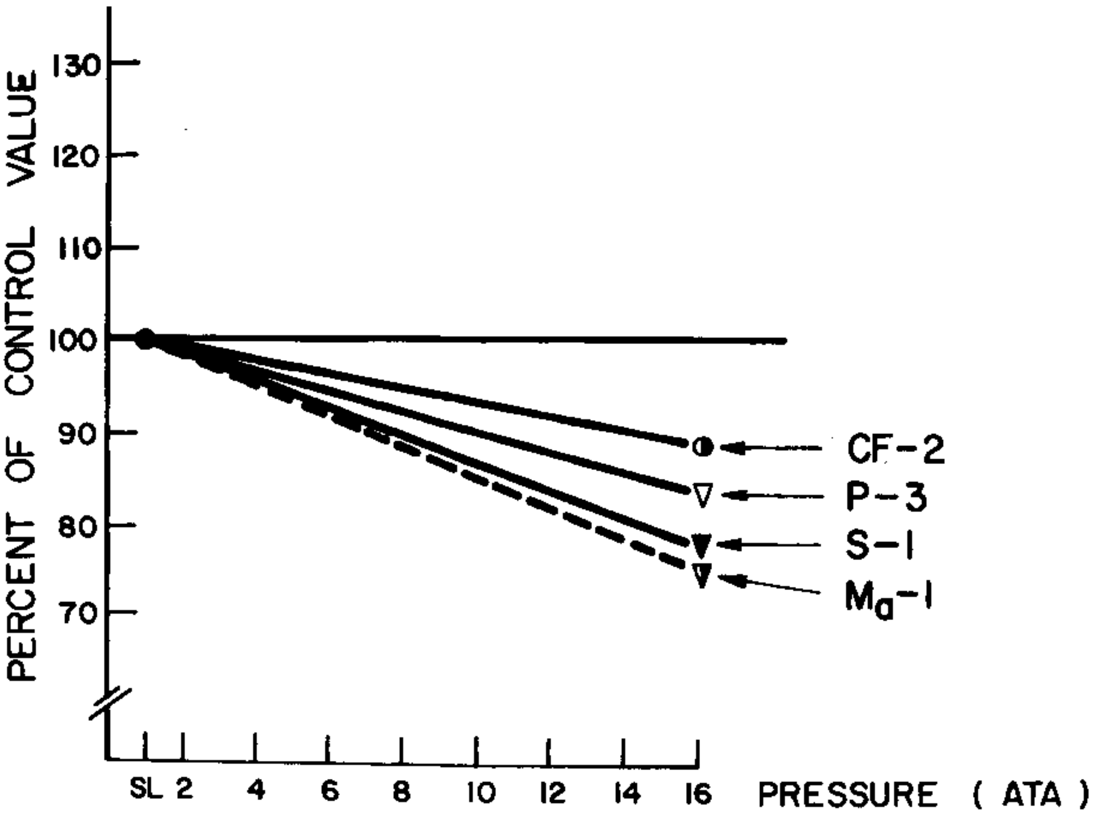

Figure 3. Group average performance on cognitive and Associative Memory tests. Results are listed as a percent of sea level control values. (Cf-2 completed by 5 subjects, all others had n = 6).

Present audiometry results suggest a pressure-related threshold shift at lower frequencies similar to that noted in hyperbaric air. An abrupt transition beyond 1000 Hz produced a slight hearing improvement in the high frequency ranges. While the degree of instrument-related loss from pressure and humidity is not known, high ambient noise from life-support machinery and its magnification due to the reverberation characteristics of the steel hull could contribute to sensitivity loss in the lower ranges. Design of audio warning devices and communications equipment for similar environments should reflect consideration of anticipated revisible hearing loss.

Maintenance of reaction time speed indicates that helium at 16 ATA does not appear to increase reticular system inhibition during perceptual-motor processing. The significant increase in incorrect responses deserves further comment, however.

There was a tendency for errors to increase throughout the dive, rather than remain a direct function of ambient pressure. This increasing rate of incorrect responses on Day 7, at the relatively less narcotic pressure of 6 ATA, suggests the existence of additional factors influencing response behavior. Loss of subject motivation after a week of repeated testing would be an appropriate expectation, but the consistency of response times, as well as sustained performance on the psychomotor measures, indicates that the subjects' interest, or general operant level, did not alter appreciably. Similar performance maintenance commonly has been noted during isolation or sensory-deprivation studies.

An alternative explanation might postulate a cumulative effect of helium breathing, with inert-gas tension increasing over time, maintaining an appreciable level of narcosis. There is no corroborating evidence for this hypothesis; indeed, there is ample evidence to indicate adaptation to narcosis over continuous exposures (Adolfson, 1965; Elcombe and Teeter, 1973; Kiessling and Maag, 1962; Walsh and Bachrach, 1971).

A more plausible explanation for the decrease in response would be an increase in perceived stress at the time of testing. Although modified somewhat by personality variables, it appears that increased arousal improves performance to a point, but excessive anxiety can interfere with efficiency. There is also evidence that anxiety disrupts complex behaviors more than simple activities (e.g., Martin, 1961).

Responses to daily anxiety scales and post-test debriefings indicated that anxiety increased markedly during decompression. All subjects considered the decompression period commencing around 5 ATA (Day 8) as the area of high flammability, as well as the region of increased susceptibility to decompression sickness. Such high perceived stress should increase arousal, possibly improving performance on the reaction time and psychomotor tasks, while interfering with decision-making ability.

Bennett (1966) noted that stress directly influenced the severity of N_2 narcosis. Bennett et al., (1967) speculated that increased levels of arousal, encountered during their chamber studies, influenced the narcotic effect of inert gases, and hypothesized that this increased arousal caused subjects to work faster (at sorting cards) at depth, but make increased errors -- a finding analogous to that of the present study. Thus, high anxiety during decompression may be thought to increase narcosis and impair decision-making performance.

The significant performance decrements noted by Baddeley and Flemming (1967) with He-O_2 at 7 ATA undoubtedly reflect such stress-induced losses. Others (Weltman, Christianson and Egstrom, 1970; Weltman, Smith and Egstrom, 1971) have noted similar performance changes attributed to psychological stress concomitant with chamber dives and dive simulations. The confounding effects of stress, learning and narcosis on complex human performance in hyperbaric environments need to be more clearly defined.

In addition to anxiety effects, psychomotor improvement during the later phase of the dive also may reflect characteristics of a learning function plus increased motivation as competitive aspects of the task increased motivation as competitive aspects of the task increased during the less demanding decompressing schedule.

The two tasks thought to measure different aspects of human memory processing provided some indication to the affected level of neurological organization. The dichotic listening task did not identify significant pressure-related perceptual or STM storage losses. Although responses on the 4-digits/ear series tended to confirm previously reported differences between first and

and second half-span recall, STM storage loss was not significant and did not decline at 14 ATA. While the three scoring techniques employed disclosed significant absolute differences, no advantage of one method over another was perceived.

Performance on the Associated Memory test, representative of a LTM task, was significantly impaired in the He-O_2 condition. The present results support similar findings with nitrogen narcosis (Fowler, 1973), and indicate a disruption of storage capability in LTM rather than STM. Although a 25% decrement in LTM was noted under hyperbaric conditions learning was still possible but less efficient. Interference with LTM may have influenced performance on the cognitive tasks, and possibly exaggerated deficits in such measures as choice reaction time.

Several tests purporting to measure cognitive ability disclosed significant decrements from control to hyperbaric administrations. However, performance impairment was not sufficient to incapacitate the subjects; eye-hand coordination, gross and fine-motor skills, and reaction speeds all appeared minimally disturbed at high pressures. As long as the behaviors required of a saturation diver are highly practiced skills, or minimally complex, performance losses appear to be minimal.

While it is evident that complex information processing was disrupted in the present study, He-O_2 narcosis remains just one of the several possible explanations for this effect. Selection of subjects lacking in He-O_2 saturation experience is an important consideration when contrasting the present results with earlier studies reporting no significant loss of memory or cognitive functioning. Minimum prior exposure should augment any potential for high arousal states engendered by the unfamiliar hyperbaric conditions. It is possible that the significant effects noted would diminish with continued saturation experience, although considerably more research is needed to identify the existence and course of He-O_2 adaptation, and to clarify the interaction of perceived stress and narcosis. Such knowledge would be essential for defining the limitation to be placed on users of treatment, research, commercial and recreational hyperbaric facilities to insure maximum safety and effectiveness.

ACKNOWLEDGMENTS

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SHIFTING ADAPTIVE SYSTEMS WITH VISUALLY DISTORTED INPUT

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ABSTRACT

Modes of adaptive recorrelation were investigated, with sixteen novice and sixteen experienced divers tested for distance estimation errors after working 12, 24, 30 and 36 minutes at various underwater tasks. Visual adaptation, defined as significant performance changes with working and nonworking hands, was evidenced by experienced divers, while novice divers demonstrated proprioceptive adaptation, as defined by performance change only with the working hand. Experienced divers showed greater adaptation to underwater distance distortion than did novices, for targets set within arms' reach.

INTRODUCTION

Adaptation to classical prism-induced optical distortion has been well documented, with initially large reaching and pointing errors decreasing with continued exposure. A return to veridical viewing after prism removal provides temporary overcompensatory movements, or after-effect, reflecting adaptation within the viewer's visual-proprioceptive system (Epstein, 1967). Similar reaching errors, accompanied by indications of adaptive correction, also have been reported when viewing through an air-glass-water interface such as that encountered while viewing underwater (Kinney et al., 1968; Ono and O'Reilly, 1971; O'Reilly, 1975).

The conditions necessary for the onset of such adaptation to optical distortion have not been fully resolved. Numerous investigators have affirmed the necessity of self-induced (reafferent) movement for adaptation to occur (Held, 1963, 1965; Held and Hein, 1963; Held and Rekosh, 1963). Ono and O'Reilly (1971) demonstrated that adaptation to underwater distance distortion is directly related to reafferent movement, and unaffected by passive viewing of underwater activity. However, reafferent movement alone may not be a sufficient condition for adaptation; learned readiness to respond also has been found necessary for adaptation (Festinger et al., 1967).

Since the recorrelation of information on effector commands issued to the body musculature and the visual feedback of resulting efferent responses are thought by many to be the basis of adaptation, development of appropriate correlated movements becomes critical. It has been suggested that, following practice with active movements in a distorted environment, subsequent adaptive movement is initiated based on newly stored correlations different from those required for efferent movement in the original visual field.

In investigating which afferent system adapts to conflicting information produced by visually distorted stimuli, several investigators (e.g., Harris, 1965) concluded that a proprioceptive shift in felt-hand position relative to the body is basic to optical-displacement adaptation. However, the typical exposure times used in these studies have been of short duration. Long duration exposures (144 hours and 42 days) used by Hay and Pick (1966) provided evidence of visual-system adaptation. Others have since reported adaptation in both visual and proprioceptive systems, with proprioceptive adaptation occurring first (Festinger et al., 1967; Rierdan and Wapner, 1966; Rock, 1966). Visual adaptation was found to occur as a long-term effect after sufficient nonvisual stimuli, including body exposure, were placed in conflict with vision.

It is recognized that visual information determines what is perceived when visual and tactile sensory inputs are in conflict (Hay et al., 1965; Rock and Harris, 1967). Kinney and Luria (1969) confirmed this phenomenon of "visual capture" of the proprioceptive system in underwater viewing. While some investigators suggest that modified eye posture or other visual cues can lead to adaptation of the visual system (Hay and Pick, 1966; McLaughlin and Rifkin, 1965), the actual mechanism for visual adaptation has not been clearly identified. Ono, et al. (1970) propose that oculomotor adjustment through convergence and accommodation may provide appropriate visual cues when adapting to underwater distance distortion.

Optical distortion investigations have disclosed that exposure conditions effective in producing proprioceptive adaptation do not cause these adaptive shifts to transfer to the contralateral, non-working hand (Efstathiou, et al., 1967; Hamilton, 1964; Harris, 1963, 1965; Mikaelian and Held, 1964; Rock and Harris, 1967). On the contrary, discrepancy between working and non-working hands has been accepted as a measure of proprioceptive adaptation in the working hand; while adaptive responding in the non-working hand has been taken as indication of visual recorrelation (Hay and Pick, 1966; Harris, 1965; McLaughlin and Bower, 1965; Rock and Harris, 1967). The present study adopted this adaptation transfer convention as a method for differentiating visual from proprioceptive adaptation.

Use of experienced divers, with long exposure to a naturally "distorted" environment, may provide a unique comparison to the typically short-term studies dominating adaptation literature, and more closely approximate the few extensive prism-exposure studies. If experienced divers have established visual-motor recorrelation contingent on the environment in which they function, then a brief exposure to water-distorted distances should provide a situation analogous to long-term prism exposure. Thus, the present study sought to define the course of adaptation to underwater distortion, and determine the mode of adaptation occurring for divers with varying underwater viewing experience.

METHODS

Subjects. Sixteen male experienced divers were selected from sport-diving volunteers; their average age was 29 years, and median SCUBA experience was over 200 hours (range of 40 to 1200 hours). Sixteen male novice divers were solicited from diving courses; the only stipulation was familiarity with skin diving and/or SCUBA diving. Average age for the novice group was 26, and median SCUBA experience was 2.25 hours (range of 0 to 10 hours).

Testing apparatus. Two adaptation measures were taken (working and non-working hands) on a device designed to measure over-reaching error, for after-effect, occassioned when a visual-proprioceptive coordinating system adapted to the water environment was used in making distance estimates in air. Use of the after-effect response as a dependent measure is consistent with most adaptation studies, and has the advantage of controlling for subjects' conscious correction.

This after-effect testing device (the A-ETD) was patterned after one used in earlier research (Ono et al., 1970; Ono and O'Reilly, 1971). Basically, it was a painted plywood tank, 48 inches long by 30 inches high by 16 inches wide, fitted with a standard facemask and containing a moveable, five cm, black disk stimulus (Figure 1). Target distances used were 7 and 14 inches from the facemask plate; the distances were presented in a block randomized design. The A-ETD width was sufficiently narrow to allow subjects to indicate apparent stimulus distance with either hand when viewing through the facemask into the tank interior. The A-ETD was stationed in shallow water, with the tank and the subject's head and shoulders out of water.

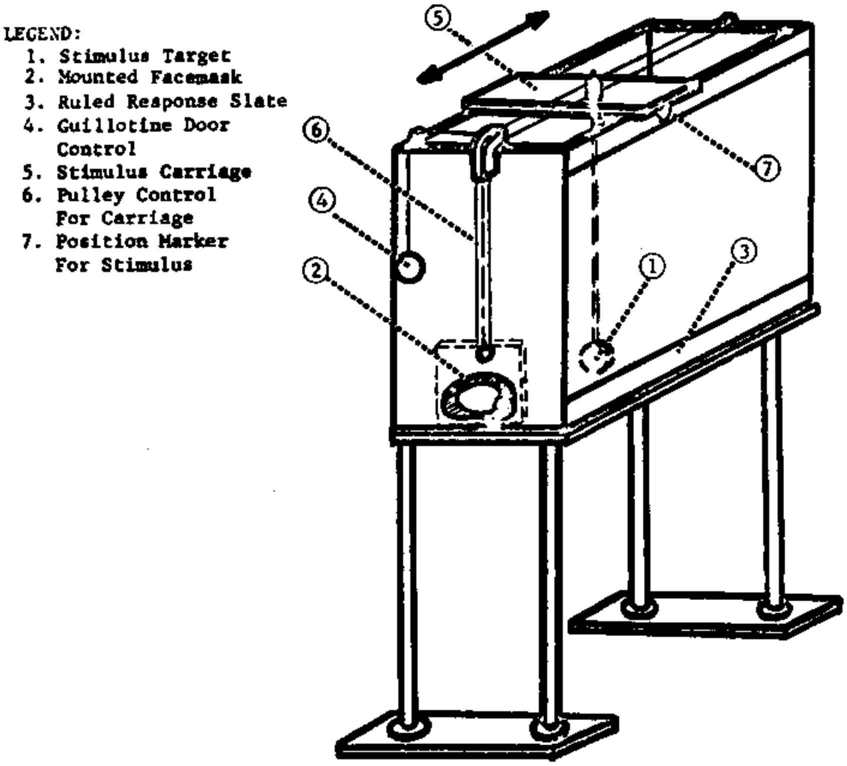

Figure 1. Schematic drawing of the After-effect Testing Device (A-ETD).

Adaptation tasks. All subjects received identical tasks for the first 24 minutes underwater. Two tasks, earlier found to be effective in inducing adaptation (Ono and O'Reilly, 1971), were administered to all subjects in a balanced design. One task, a modified version of the Minnesota Rate of Manipulation Test, provided the opportunity for ballistic movements when reaching for targets within close distances (6 to 24 inches). The second task, the Underwater Distance Learning Apparatus, was constructed to maximize new afferent-efferent associations by providing discrepancy feedback for hand movements within arms' reach which lacked visual guidance. Subjects worked at each task for 12 minutes in a water depth of seven feet while breathing from a SCUBA tank.

Feedback tasks. After 24 minutes of work on the adaptation tasks (interrupted by above water measurements), subjects were assigned to one of two conditions designed to provide similar distance reaching tasks while varying the quality and quantity of feedback placed in conflict with visually-distorted input. Half of each experience group was assigned high feedback tasks thought to place visually-distorted input in conflict with high reference cues such as views of one's body and veridical measures of surround. The remaining subjects received low feedback tasks, equivalent in amount and type of muscular activity and opportunity for ballistic reaching movement, but without equivalent body or surround referent feedback. The tasks were:

High Feedback:

Body Reference - The seated diver first looked at various indicated body parts (left and right thighs, knees, ankles, toes), then reached quickly for the part indicated.

Ground Reference - The standing diver first estimated the distance from a reference point to one of several targets spread across a table. After entering a written estimate, the diver retrieved the target, deposited it in a container, and then measured the actual distance with a ruler, entering this figure opposite his first estimate.

Low Feedback:

N-Body Reference - Similar to the body reference task, except that the diver worked at an underwater table, reaching for one of eight lines placed at distances equivalent to those in the high feedback condition.

N-Ground Reference - Similar to the experimental condition except that no distance measurements were performed. The diver made a second estimate after removing the target.

Procedure. Subjects first were provided with a detailed description of each task and a brief practice session on the adaptation tasks. Instruction slates were prepared as reminders for underwater use. Subjects were randomly assigned to use their preferred or non-preferred hand, and provided with a glove for the non-working hand, as a reminder not to use or view that hand at any time. They were also provided with wet suits, weight belts, SCUBA tanks and facemasks.

All subjects were pretested on the A-ETD, with twenty distance estimations taken (five trials for each of two target settings and two hands). They then swam to the underwater work site, located at a depth of seven feet in a large, heated, lightly chlorinated, freshwater pool. After working for 12 minutes on one of the adaptation tasks the subject returned to the A-ETD for twelve distance estimates (three trials at each target setting for each hand). Following this testing he returned for 12 additional minutes underwater working on the second adaptation task, followed by an additional twelve A-ETD measures.

The feedback conditions were administered at the end of 24 minutes of underwater activity, with subjects working on the first of their two conditions for six minutes, followed by twelve A-ETD distance estimations. A final six minute work period was followed with twenty A-ETD measures.

Total underwater working time was 36 minutes, with five A-ETD testings: a pretest before entering the water, and a test at the end of 12, 24, 30 and 36 minutes of work underwater. Time underwater was chosen as the task variable rather than number of trials on the individual tasks, since it has been shown that, over time, cold adversely affects performance underwater (Bowen, 1968), and subsequent adaptive after-effect performance (Ono and O'Reilly, 1971).

RESULTS

Mean scores were derived from three (or five) trials at each stimulus distance for each hand. The after-effect data were expressed as difference scores, calculated as change from initial pretest measurements. Positive values represent over-reach from pretest estimations. No significant difference was found between 7 and 14 inch stimulus distance scores for any group or condition, allowing these after-effect measures to be averaged across distances to simplify presentation and increase score stability. All analyses were performed with mean difference scores averaged across the two distances. Separate analyses were performed on the results from the first two adaptation periods (after 12 and 24 minutes of underwater work), and those from the feedback conditions (the 30 and 36 minute testings).

For the adaptation data, a 2 (diving experience) by 2 (hand) by 2 (time underwater) analysis of variance, with repeated measures on the last two factors, indicated a significant difference in level of adaptation exhibited by the experienced subjects compared to novice subjects ($F(1.39) = 7.29$, $p < .025$). Highly significant differences also were noted for working versus non-working hand ($F(1.30) = 18.05$, $p < .001$) and time underwater (12 minutes versus 24 minutes) ($F(1.30) = 14.39$, $p < .001$). A significant experience by time interaction was also found ($F(1.30) = 8.79$, $p < .01$) (Figure 2).

After twelve minutes of underwater work there was no difference between the after-effect exhibited by the working hand of the novice and that found for the working and non-working hands of the experienced subjects. After 24 minutes, experienced subjects exhibited significantly more adaptation with their working hand than did the novices ($t(30) = 2.68$, $p < .02$).

Additional mixed model designs disclosed a significant difference between working and non-working hands for novices ($F(1.14) = 14.17$, $p < .01$), but not for experienced subjects. Significant changes in adaptation over time, as

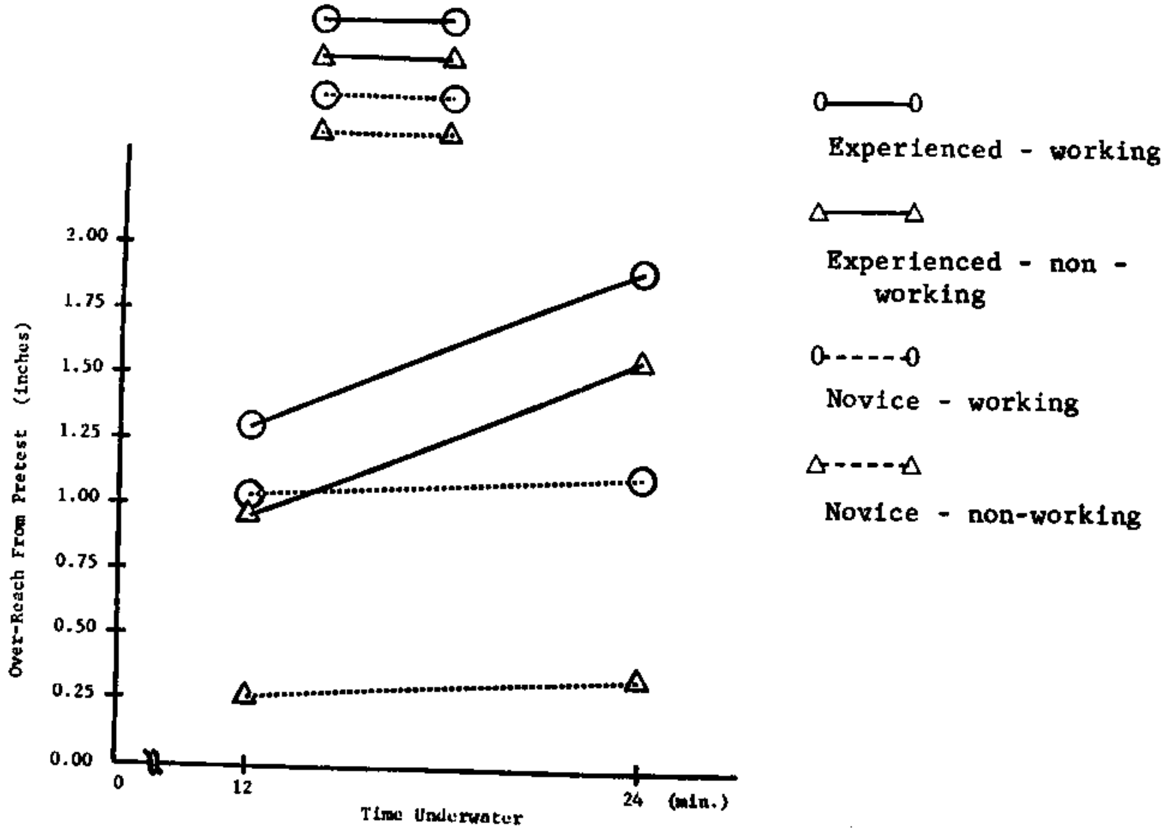

Figure 2. Comparison of amount of adaptation by working and non-working hands for experienced and novice Ss during adaptation phase.

measured by increasing after-effect scores, occurred for experienced subjects (F(1.14) = 18.41, $p < .001$), but not for novices.

To investigate changes in adaptation occurring during the last two testing sessions (30 and 36 minute tests), data for this period were analyzed as difference scores from the after-effect measurements registered at the 24 minute A-ETD testing. Novice subjects generally exhibited increasing adaptation during these sessions (Figure 3). Only the non-working hand of the high feedback experienced subjects demonstrated increased adaptation during this same time (Figure 4). Analysis of the experienced subjects' performance disclosed a significant hand effect (F(1.14) = 9.99, $p < .01$), and feedback by hand interaction (F(1.14) = 6.19, $p < .05$); reflecting significant improvement for the non-working hand of the high feedback subjects only.

A significant feedback by time interaction was noted within the novice group (F(1.14) = 5.43, $p < .05$), partially reflected in improved adaptative responding over time for the low feedback subjects concomitant with a slight decline in average the high feedback subjects' responding. This interaction was also affected by a significant difference in non-working hand performance between novice feedback groups (t(14) = 2.53, $p < .05$) at the 30 minute testing, which was not evident at the 36 minute testing.

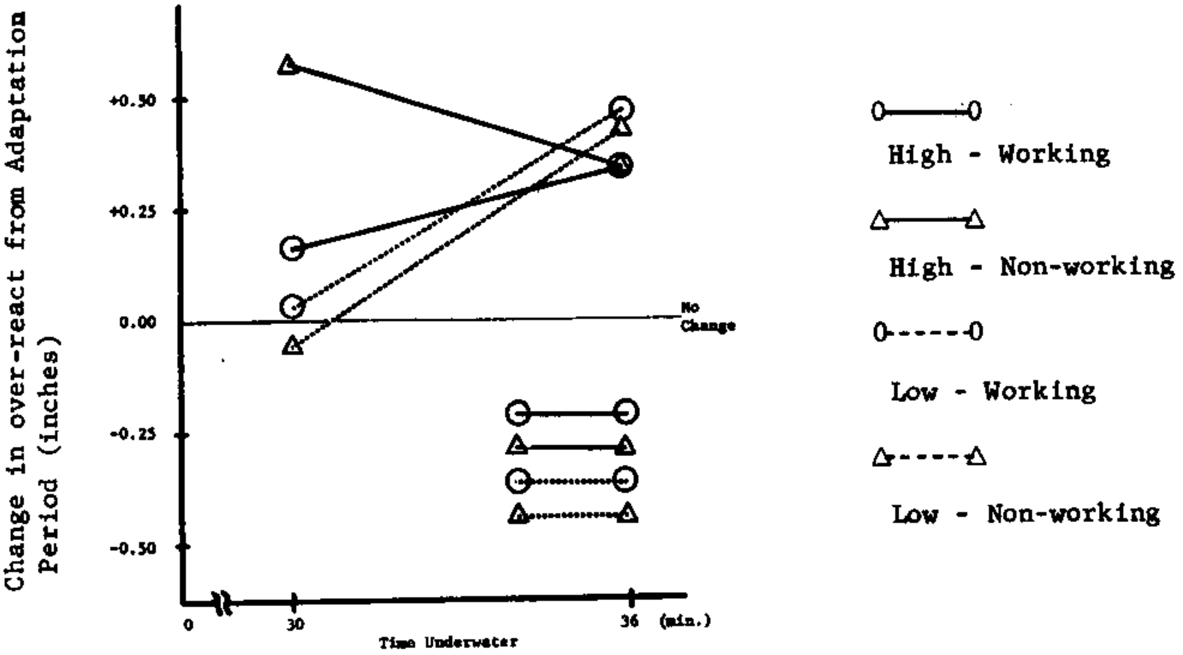

Figure 3. Comparison of change in after-effect by working and non-working hands. High and low feedback subjects novice group.

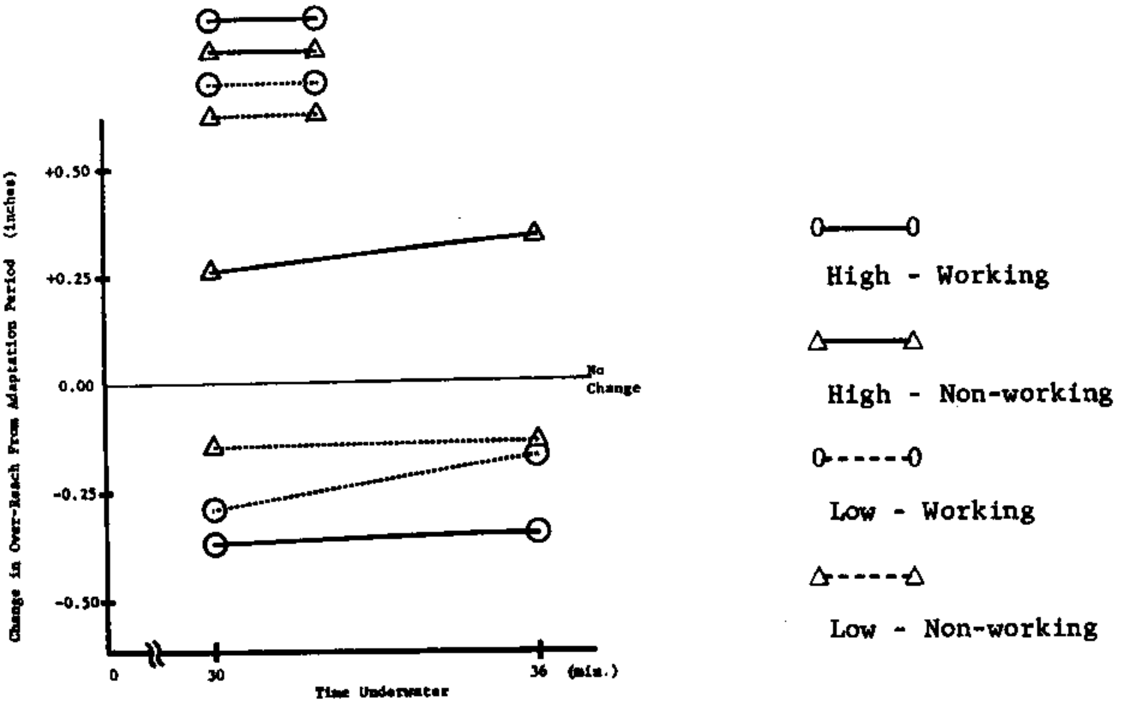

Figure 4. Comparison of change in after-effect by working and non-working hands. High and low feedback subjects experienced group.

The 30 minute responses for high feedback novices were significantly above their 24 minute level ($t(7) = 2.65$, $p < .05$), while a significant decrease in distance estimation error was noted for the high feedback experienced subjects at their 30 minute testing ($t(7) = 2.48$, $p < .05$). Other after-effect changes were not significantly different.

DISCUSSION

The present results demonstrate a further instance of plasticity in human sensing capabilities, with the data indicating that novice and experienced subjects utilized different adaptive methods in adjusting to distance distortion through a water-glass-air interface. Experienced subjects exhibited visual adaptation, as defined by similar after-effect responses produced by both working and non-working hands. Inexperienced subjects showed proprioceptive adaptation to the underwater optical distortion, as defined by a difference in after-effect responding for working and non-working hands.

Experienced divers exhibited greater over-reach (reflecting increased adaptation) over time than did the novices; approaching 30% error for the closest target distance. Continued adaptation over time was found for the experienced subjects, while novices demonstrated a lower, constant level of adaptation, peaking after the first 12 minutes of underwater work. As has been found in prism-distortion research, proprioceptive adaptation occurred rapidly, within the first 12 minutes for novices. The experienced subjects, utilizing visual adaptation, adapted to this same level initially, but over-reaching errors (or adaptation) continued to increase significantly after the second underwater work session.

The feedback tasks were implemented to increase the possibility of adaptation through visual restructuring, due to the selective presentation of input stimuli in conflict with veridical referents. As experienced subjects appeared to function from the outset with a change in visual coding, no dramatic performance alteration could be expected due to the feedback conditions. On the other hand, novices appeared to function through the first 24 minutes with an altered felt-position of the hand, or change in the more plastic proprioceptive sense. Based on their lower adaptation level, novices had the best opportunity to improve performance during the high feedback condition. Significant improvement in non-working hand responses for high feedback novices, and a slight general increase in adaptive responding, are consistent with the introduction of more efficient adaptive processing occurring over time, but do not provide unequivocal evidence that the high feedback conditions elicited a shift from proprioceptive to visual restructuring.

Experienced subjects demonstrated a generally insignificant, but consistent loss in adaptation after 30 and 36 minutes of water exposure. Confounding factors such as fatigue, boredom or chill may have influenced performance; a similar decline has been noted earlier for divers passively observing underwater work (Ono and O'Reilly, 1971). As the experienced divers appeared to have approached a ceiling level of adaptation they may have been more susceptible to demonstrating the effects of competing environmental factors.

The failure of targets at different distances to enhance the absolute magnitude of after-effect responding was inconsistent with the optical principles underlying the distortion effect, which would predict that absolute adaptation for the 14 inch placement should be twice that observed for the 7 inch placement. Rather than a linear increase in adaptation, as had been observed in a separate study utilizing underwater measurements (O'Reilly, 1975), a relatively constant limit of absolute adaptation was noted for target placements in air (at 7 inches, average error was 1.88 inches, or 30% of the initial setting; at 14 inches, average error was 1.93 inches, or 14% of the initial setting).

The existence of an absolute level of over-reach may be evidence of competition between the newly acquired (or reacquired) afferent-efferent coordinating system, and that already established for the normal air environment (in which the measurements were taken). One might speculate that the relatively less accurate proprioceptive sense allows a margin of flexibility in position sensing before higher-order integrating centers override errant positioning responses dictated by the newly acquired coordinating system.

While performance of the experienced divers was indicative of visual adaptative responding, alternate explanations for the acquisition of non-working hand adaptation should be considered. Proprioceptive adaptation in a non-working hand would require intermanual transfer; however, numerous investigators have reported no such transfer under conditions designed for proprioceptive adaptation. Additionally, the inexperienced subjects exhibited no adaptive transfer in the present study, although receiving the same visual distortion and psychomotor tasks as the experienced subjects.

The possibility of experienced subjects exhibiting reestablishment of conditional proprioceptive adaptation, independently acquired by each hand during previous visual-motor performances underwater, is not consistent with studies reporting no adaptive shift in distance estimations for experienced divers when viewing, but not working, underwater (Ono and O'Reilly, 1971), or data supporting limited conditional adaptation to distance estimations when divers were tested immediately after entering the water (O'Reilly, 1975). Since adaptation appeared only after working in the visually-distorted environment, and accepting that proprioceptive adaptation does not transfer, establishment of learned movements based on efferent sets activated by newly recoded visual input is the most parsimonious explanation.

Exactly how the various sensory systems recode information in establishing new afferent-efferent correlations cannot be determined from this investigation. The availability of muscular cues from increased convergence and accommodation due to optical displacement of the virtual image underwater might serve a function analagous to that postulated for prism-induced changes in "felt-position" of the eye. While there is some controversy surrounding the actual functioning of these muscular changes (e.g., Richards and Miller, 1969), oculomotor processes are generally considered as effective cues for close distances, and may underly adaptation to underwater distortion. In addition to loss of accommodation and convergence as distance cues, other variables such as illumination, turbidity, contrasts and object surroundings must be considered when target distances differ. For close distances additional factors must be considered, particularly thickness of the glass interface and distance from target to interface.

Consideration of adaptive changes in visual-motor performance of divers may be important in understanding human performance in the sea. Divers can be expected to exhibit underestimation of distances within arms' reach. Diving experience is related to severity of error and mode of adaptive restructuring. Adaptation, or decrease in error of estimation, occurs over time for both novices and experts, indicating possible advantages in introducing "warm-up" tasks prior to critical underwater operations. Tasks requiring ballistic reach, or proximal-distal movements without close visual guidance, should be avoided immediately on entering the water or after emerging from the water.

In general, results from this study illustrate the advantage of utilizing a natural source of visual distortion, and subjects experienced in functioning within this environment. Utilization of the natural semi-weightlessness and decorrelation conditions unique to the water environment should be encouraged to further extend knowledge of human sensory functioning.

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ADAPTATION TO UNDERWATER DISTANCE DISTORTION

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ABSTRACT

Thirteen novice and thirteen experienced SCUBA divers were tested for accuracy in matching target distances set within arm's reach (6, 12, 18 inches). Experienced divers demonstrated greater accuracy than novices for initial underwater testings. Working on selected tasks while underwater significantly improved distance judgments for both groups, with novices approaching the accuracy of experienced divers after 36 minutes of underwater activity. Both groups failed to reach the same level of accuracy while underwater that they achieved during above water testing, indicating less than complete adaptation to underwater distance distortion.

INTRODUCTION

While it is recognized that visual input is distorted when viewed through an air-glass-water interface, such as a diver's facemask, little is known regarding the functional alterations in apparent distance occurring in water environments. On the basis of physical laws of refraction and optical cues derived from changing convergence and accommodation, it has been estimated that objects viewed underwater should appear approximately twenty-five percent closer than the same distance would appear if viewed in air (Ono and O'Reilly, 1970).

While seventy-three percent of professional Navy divers interviewed perceived objects as appearing closer in water than in air (Kent, 1966), experimental investigations into distance judgments under water have tended to be in variance with these subjective reports. Kent (1966) reported overestimation of actual distance for underwater stimuli placed further than three feet from the diver, but little distance estimation error below three feet. Overestimation of apparent distance relative to actual distance also has been noted for target distances between four and fifteen feet (Luria, Kinney, and Weisman, 1967). Other studies have disclosed distance underestimation both in air and water, with the water estimates less at near distances and markedly greater than air estimates beyond forty to fifty feet (Ross, 1965, 1967).

The degree of water turbidity appears to affect distance estimation, with underestimation occurring only under ideal conditions of clear water and distances within arm's reach (Kinney, Luria, and Weitzman, 1969). Overestimation of distances has been found to be related to a reduction in target contrast, lack of specific distance cues, and homogeneity of stimulation (Luria, et al., 1967; Luria and Kinney, 1968).

Studies utilizing targets within arm's reach have demonstrated that both novice and experienced divers produce apparent distance measures closely predicted by underlying optical principles (Ono, O'Reilly, and Herman, 1970; Ono and O'Reilly, 1971). Practicing specific visual-motor tasks in the water environment produced significant adaptation to underwater distance displacement, when measured by a surface-administered after-effect test. However, the existence of conditional adaptation for experienced divers, and differential rates of adaptation relative to prior underwater viewing experience were not determined.

Kinney, Luria, and Weitzman (1968) investigated eye-hand coordination and adaptation over time for divers estimating distances within arm's reach. They reported complete adaptation to distance distortion occurring immediately for experienced divers; novice divers tended to underestimate apparent distances initially, reaching complete adaptation within fifteen minutes after working on various underwater tasks. While this study discloses the existence of complete conditional adaptation for individuals experienced in underwater viewing, the limited sample size (two experts, four novice divers, and two completely inexperienced subjects) and arbitrary definition of experience serve to limit the applicability of their findings.

Accordingly, the present study attempted to identify the degree of distances estimation error occurring for novice and experienced divers immediately upon entering the water, and the degree and rate of adaptation to underwater distortion occurring over time as a function of prior diving experience.

METHOD

Thirteen novice and thirteen experienced divers were selected from volunteers from local diving clubs and courses. The novice subjects had an average age of 27 years, and a median SCUBA experience of one hour, with a range from 0 to 10. Experienced subjects averaged 29 years of age, with a median SCUBA experience of 100 hours, ranging from 40 to over 1200 hours. Half of each group were instructed to use their preferred hand, half their non-preferred hand, only this working hand was used for testing.

Testing was accomplished in an outdoor, lightly chlorinated, heated pool, with scheduling controlled for similar light and weather conditions across groups. All tasks were administered in seven feet of water. The contents of the pool were continually filtered and closely monitored, with resulting visibility being high and approximately constant throughout the experiment. Masks and SCUBA gear were standardized across subjects.

Distance measurements were obtained first above water (A/W pretest), followed by an underwater (U/W) pretest, then additional tests at the end of three twelve-minute work periods (12, 24 and 36-minute testings).

All subjects received the same tasks for the first twenty-four minutes underwater, with presentation balanced across tasks. These tasks were: 1) a modified version of the Minnesota Rate of Manipulation Test, and 2) an eye-hand task thought to maximize ballistic movement and provide meaningful proprioceptive feedback. Both of these tasks previously had been found to be effective in facilitating adaptation (Ono and O'Reilly, 1971). After twenty-four minutes, half of the subjects in each experience group received practice on tasks designed to further increase adaptation; the remaining subjects worked on tasks providing identical reaching experiences with different degrees of visual feedback (O'Reilly, 1969). Since no significant differences were found within experience groups due to these latter tasks, only analyses relating to the total pooled-sample experience groups will be considered.

The testing apparatus consisted of a rectangular plyboard assembly, covered with parallel lines one inch apart, and supporting two movable red plexiglas markers on slide tracks. One marker (the target), controlled by the experimenter, was visible to the subject. The second marker, controlled by the subject, was concealed from his view, but visible to the experimenter. The subject was required to match the setting of the visible marker by reaching underneath the device and placing his marker immediately adjacent to the target. Positioning accuracy was measured to the nearest tenth of an inch. Three distances were presented: 6, 12, and 18 inches from the edge of the apparatus facing the subject. The stimulus distances were presented in randomized block design with two sets of three distances presented on each administration.

RESULTS

The dependent measures were the means of two trials at each of three stimulus distances for five administrations. Underwater pretests exhibited consistent underestimations of target distances relative to A/W pretests. Figure 1 details the initial underestimation and subsequent improvement for all stimulus distances for novice and experienced divers. The two experience groups did not differ significantly in mean distance estimates on the A/W pretests. A series of treatment-by-subject analyses of variance was performed, with subsequent application of Newman-Keuls range tests.

For the 6-inch target setting, the experienced and novice groups both demonstrated a significant effect across the five test administrations ($F(4.48) = 3.08$, $p < .05$, experienced group; $F(4.48) = 4.64$, $p < .01$, novice group). For the experienced group, the only differences were between the U/W pretest and the 24 and 36-minute testings. For the 6-inch target distance only, the A/W and U/W pretests were not significantly different, indicating accurate matching at the closest distance. Distance estimates by the novices followed those of the experienced subjects, with the exception that the initial U/W pretest was significantly lower than the corresponding A/W setting. Significant improvement in accuracy was noted for the 12, 24 and 36-minute testings, relative to the U/W pretest. No significant increase in accuracy of estimation was noted for either group after the 12-minute test.

Significant improvements in accuracy were found for both groups on the 12-inch target placement ($F(4.48) = 7.07$, $p < .001$, experienced; $F(4.48) + 18.31$, $p < .001$, novice). Both groups demonstrated significant losses in accuracy from air to water pretests. For the novice group, all underwater testings were significantly underestimated relative to the air testing. For experienced subjects, the 36-minute testing provided the only underwater measurement not significantly different from the A/W pretest for this target distance.

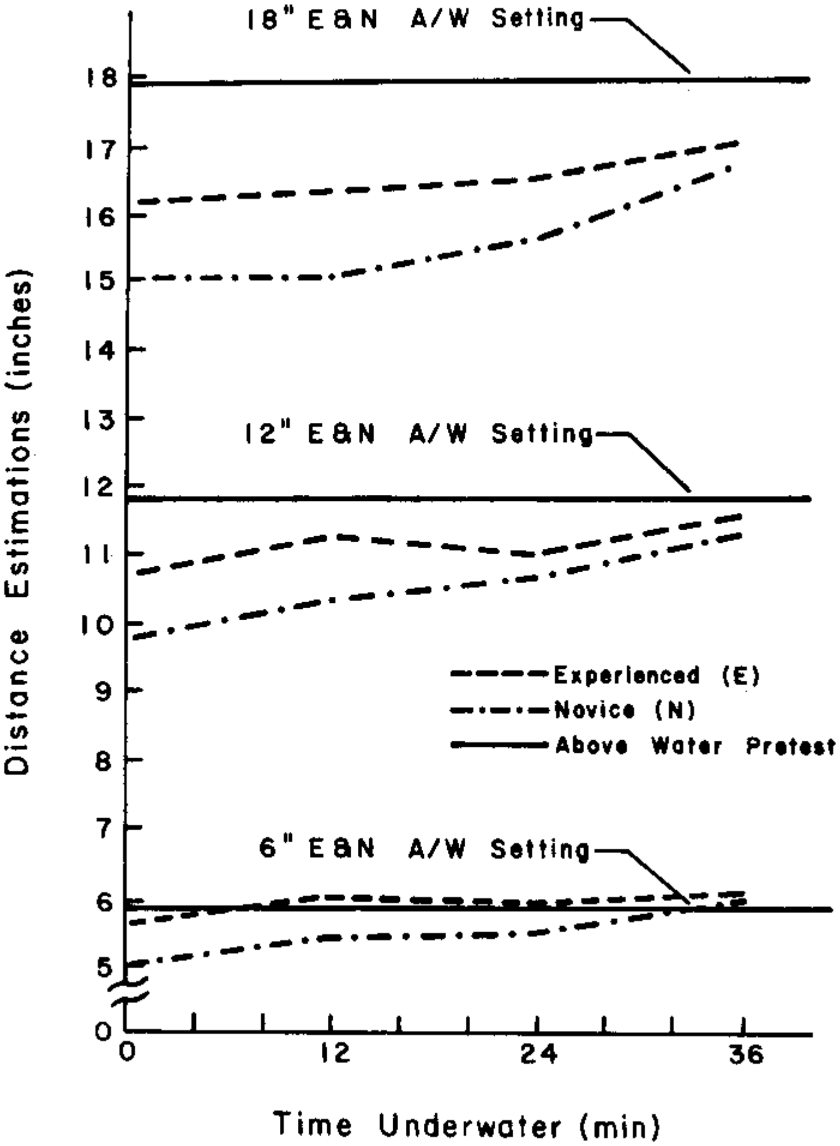

Figure 1. Accuracy of distance matching by experience group by target distance.

Very significant losses in accuracy of distance matching occurred from the A/W pretest to the U/W pretest, for responses to the target set at 18-inches ($F(4.48) = 16.20$, $p < .001$, experienced; $F(4.48) = 33.26$, $p < .001$, novice). All U/W measurements for both experience groups were significantly different from the air and 36-minute testings, with distance judgments from this latter testing also significantly underestimated relative to the A/W distance.

Group comparisons of U/W distance-estimation accuracy relative to A/W scores were accomplished by a series of t-tests. No significant difference in accuracy was found for the closest target placement (6-inches) ($t = 0.78$, $df = 12$, $p =$ n.s.). At greater target distances novice subjects exhibited significantly greater underestimation from A/W pretest to U/W pretest than did experienced subjects ($t = 2.59$, $df = 12$, $p < .05$, 12-inch placement; $t = 2.42$, $df = 12$, $p < .05$, 18-inch placement).

In contrasting U/W pretests to A/W pretests by target distances, it was noted that the absolute value of distance underestimation increased as a function of target distance, but the percentage error was constant for the greater distances. Distance underestimation for novice subjects averaged 0.59 inches (10.5%), 2.01 inches (17.0%) and 2.83 inches (15.8%) for the 6, 12 and 18-inch settings respectively. Experienced subjects averaged underestimations from A/W pretest of 0.40 inches (6.7%), 1.10 inches (9.3%), and 1.71 inches (9.6%) for the 6, 12 and 18-inch settings.

DISCUSSION

A significant increase in accuracy of distance matching occurred over time for both experienced and novice subjects. Novices demonstrated a greater rate of adaptation to underwater distance distortion, starting significantly less accurate than the experienced divers at the two greater target distances, and reaching approximately the same level of performance after 36 minutes of underwater activity.

Generally, experienced subjects failed to reach a level of accuracy equivalent to above water results. This finding casts doubt on the Kinney et al. (1968) study reporting rapid or complete adaptation after fifteen to thirty minutes of underwater activity. Although there was a difference in adaptation procedures between the two studies, tasks chosen for the present study were specifically designed and tested to facilitate adaptation; while Kinney et al. merely instructed their subjects to be as active as possible in diving for objects, playing checkers, etc. -- all movements capable of complete visual guidance. Additionally, they also tested a more limited range of target distances (6 to 10 inches) than did the present study (6 to 18 inches), which may have reflected a tendency for more accurate responding at closer distances. The present study disclosed that underestimation was increased from the 6-inch condition to the 12-inch condition, but within each experience group the percent error remained constant for the 12-inch and 18-inch settings, perhaps reflecting some limitation dictated by optical principles interacting with proprioceptive feedback.

Results of the present study indicate that significant underestimation of distance for targets within arm's reach can be expected for both novice and experienced subjects upon first entering the water. Significant increases in accuracy occurred over time, although considerable error was still evident after 36 minutes of underwater work. This improvement in accuracy of distance estimation could be considered evidence of adaptation to underwater visual distortion, although the present research design does not entirely control for conscious correction. Evidence of conditional adaptation was suggested with initial underwater responses of experienced subjects significantly more accurate than novice scores at the greater target distances.

Implications can be extended to the design and sequencing of operational tasks to minimize the possibility of reaching errors, and the need to control for differential levels of experience-related adaptations.

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