PROCEEDINGS

of the

FIRST ANNUAL TROPICAL AND SUBTROPICAL FISHERIES TECHNOLOGICAL CONFERENCE

Volume I

Compiled by

Bryant F. Cobb III and Alexandra B. Stockton

October 1976

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TROPICAL AND SUBTROPICAL FISHERIES TECHNOLOGICAL CONFERENCE

March 8-10, 1976

Corpus Christi, Texas

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IN MEMORY:

Dr. Bryant Franklin Cobb III
November 23, 1935 - August 30, 1976
Associate Professor, Seafood Technology
Department of Animal Science
Texas A&M University

For many years a technological conference on fishery products from the Gulf of Mexico had been discussed by many concerned individuals from industry, education and government. Finally, the First Tropical and Subtropical Fisheries Technological Conference was held in March 1976 in Corpus Christi, Texas. The first meeting was a success and the more than 85 participants from many different states and countries agreed that the concept should be continued.

The meeting was not the result of happenstance; instead it was the result of a dream and a tremendous effort on the part of Dr. Bryant F. Cobb III. Dr. Cobb enjoyed discussing the subject of seafood technology with others working in the same or allied fields and felt that the conference would provide an ideal place for communication and cross-pollination of ideas. It did and will continue to serve that end. For that reason these first proceedings are dedicated to Dr. Cobb.

It is hoped that the professionalism, scientific inquisitiveness and love shown for the seafood industry by Dr. Cobb are reflected in the proceedings, and that they will be an inspiration for generations to come.

ELECTED OFFICERS 1976-77

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E. Spencer Garrett

Executive Council[a]

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Melvin E. Waters - Mississippi

Richard T. Lovell - Alabama

Charles B. Davies - Florida

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Cipriano Reyes-Garza

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Ronald P. DeBenedetto - Bayou Foods - General Mills, Inc., Mobile, Alabama

Gary McCrary - Coastal Freezing, Aransas Pass, Texas

James Pace - Pace Fish Co., Brownsville, Texas

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[a] It was recognized that additional representatives for Mexico, South and Central America, Puerto Rico and the Caribbean area would have to be elected as these areas increase their participation in the conference.

REGISTRATION LIST

for

TROPICAL AND SUBTROPICAL FISHERIES TECHNOLOGICAL CONFERENCE

March 8 - 10, 1976

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KEYNOTE ADDRESS

THE VALUE OF TECHNOLOGY TO THE SEAFOOD INDUSTRY

Roy E. Martin
Director of Science and Technology
National Fisheries Institute
Washington, D.C.

First of all let me add my welcome and that of the National Fisheries Institute to all those attending the First Tropical and Subtropical Fisheries Technological Conference. Your attendance and participation is indication of the importance you placed in establishing this conference. May it grow as your sister conferences - the Atlantic Fisheries and the Pacific Fisheries Technological groups - have done.

I carry personal greetings and best wishes from them to you.

How do you define technology? Webster says it's "A technical method of achieving a practical purpose," or the "means employed to provide objects necessary for human sustenance and comfort."

I prefer to describe the term as <u>progress</u>.

But there are other voices in the land today - best described as anti-technology - doomsday portrayers. At times, from the view in Washington, it has almost seemed as though a lynch-mob mentality has taken over that has banned such products as cyclamate, nitrite, red color #2, the chloroflurocarbons, etc., and put under suspicion anything described as a food additive. This mentality has been anything but scientific.

It has, however, been loud, bold, political, media receptive and quick to institute legal action against the government.

The promulgation of more rules and regulations is the result, which in turn has a negative effect on technological advance. We are slowing, by deliberate action, our forward progress.

If this numbing pessimism continues, future technological progress may turn up stillborn.

Evaluating ourselves technically, we must conclude that we lag considerably behind other segments of the food processing industry. This lag, however, presents this industry with unprecedented opportunities. We did what we had to do to minimally get by. Our products are consumed on an average per capita of only 12 pounds per person per year, and of that total, one half is represented by tuna and shrimp. Compare that to meat consumption of some 120 pounds per person per year.

Mentioning meat is a good point at which to stop for some reflection. There is an industry that has done a superior technological job. Their story indicates a use for everything but the moo of the cow. Their patterns of technological achievement can be patterns and guidelines for us as well.

Our survival and ability to compete will depend on technology. What form that technology takes will depend only on our skill and imagination.

What technology will we need:

- A continued environmental assessment of contaminants - if necessary establishing a national NMFS analytical laboratory with the capacity to adequately monitor the environment.
- One analytical method of decomposition detection and measurement for all seafood. At last count there were 12 methods - none of which agreed statistically with the others.
- A better system of biological determination of stock and resource assessment - when will we be able to know for sure how many fish of what species we can harvest at maximum sustainable yields?
- Redesign of catch vessel holds - compartmentalize to maximize quality of species brought aboard. To have that same vessel bring in all the catch - when will we be able to say that's a fishing vessel - not a shrimp boat, a tuna boat, a lobster boat, a halibut boat, etc. Our present fisheries have no flexibility.
- We will also one day need the technology of block production in the U.S. - we are totally void in this area being dependent entirely on foreign imports.
- Salmonella control and elimination of imported seafoods. Processing methods designed to preserve the products' character, yet simple enough to not add substantially to present costs.

- Continued development of texturizers to generate new product ideas and concepts.
- Adaptation of minced technology to in-line processes as an integral part of multi-product processing operations.
- Aquaculture basics developed to a point where some hard economic analysis can convert good science to profit potential.
- Design and determine the conditions necessary (1) to use thermal effluents from atomic power as a culture medium, (2) determine if this potential can be reduced to a practical business venture.
- What relationships exist between cholesterol, Cd, Hg, Se and the other micro-constituents? What chemical forms do they exist in? Are they toxic in those forms? What seasonal variations exist in the compositional makeup of seafoods? Where is our national grid with all these factors presented so that holes in the available data can be filled on a scientific basis and made available to Sea Grant and the rest of the scientific community to know which parts of the puzzle should be completed?
- When will we as an industry be in a position to use the profitability "all but the flip on the tail?" Little research has been done on by-product recovery - what did our sister industry, the meat processors, do? Consider their by-products:

  (1) glue for furniture

(2) fertilizer for agriculture

(3) leather for shoes

(4) internal organs as a source for drugs

(5) gelatin from pork skin

(6) soaps from fats

(7) lard & shortening for frying

(8) glycerin from tallow

(9) lanolin

(10) cat gut and sausage casings

(11) oleo margerine

(12) cuttings, oils and lubricants

We also need:

- New methods of producing smoked fish products under better controlled conditions.
- Pressure formation and extrusion techniques for new forms and engineered food concepts.
- Film and package development for best protection of finished products.
- Practical development of waste treatment technology that will fit the specific needs of the seafood industry. Except for screening, no technique has come to light that has yet given this industry some hope to solve the 1983 EPA guideline deadline.

What I have just highlighted are some direct technological needs; what about those indirect areas that need pseudo-techno-

logical inputs:

- Nomenclature - a new system of finding consumer names without being deceptive.
- Hazard and critical control point quality control analysis systems.
- Nutritional labeling bases.
- Heavy metal data collection.
- Food inspection legislation response.
- Import detention problems.
- GMP's - fresh & frozen fish.
- Codex standard setting.
- Microgiological standards.
- Pesticide & toxic substances control.
- Metric conversion.
- U.S. fishery product standards & grades -- and more.

We as an industry are not overly-endowed with technology, technical personnel, money for research and development or laboratory facilities. This is not a criticism - but a hard fact. Seafoods, until recently, have never been considered a protein source of national concern. We are a nation of red-meat consumers, and as such, our government has spent its R&D dollars on USDA - not NMFS.

But now that we, as a nation, have experienced exceptional meat price hikes, questions are beginning to be asked about alternative sources of protein - the two most frequently mentioned are seafood and soy protein. This questioning becomes

more significant since we expect a presidential signature on the 200 mile bill before June. A new frontier lies before us and this country if we but put up the resources to do an adequate job. We do not have a "Department of the Oceans" - but we should! We do not have a national ocean policy - but we should! We have not updated old laws and regulations governing our resources. But we should!

The business community is under tremendous federal regulatory pressure from OSHA, EPA, FDA, the National Institute of Occupational Safety and Health, FTC and IRS. When will it stop? I don't think it will - the system has grown too large.

Technology can be a real assist to over-regulation - by finding facts to deal with problem areas - by presenting new opportunities for business potential - by elimination of inefficiency - by being the meeting ground for reasonable negotiation.

You in this audience hold that key, you can and will have the answers to these dilemmas.

What do you face in your technological quest? Let's examine but one case history of many - the consumer activist and FDA:

(1) An act that goes back to 1906 - reworked in 1938 and 1958.

(2) Food and Drug laws are no more nor less responsive to current needs that FDA administrators choose to make them. Don't be misled, I said what <u>administrators</u> choose to make them.

(3) It is current FDA philosophy that it can develop any

regulatory program it thinks is reasonable and consistent with the basic objectives of the act, whether or not the act specifically provides for such action, provided that it is not expressly forbidden. How's that for an open-ended policy?

(4) According to FDA'ers, they have not yet fully begun to explore the reaches of their statutory authority.

(5) In recent years we have seen a significant increase in the number of regulations issued by FDA. Each one, my dear colleagues, effects your technological efforts directly. These regulations have become more and more detailed. Its "how to" regulations take on the appearance of operating manuals, rather than interpretive guidelines. Is this really necessary? Was this the real intent of the act? I don't think so! Personally, I have great difficulty with the concept that FDA should tell industry how to produce and label a product. That's our responsibility and should not be shared by the government - after all, we are the ones that need the repeat sales and satisfaction of the consumer or else we will not be in business for very long.

(6) What are we up against? In 1960, FDA had about 850 people in the field doing enforcement and 650 people in Washington writing rules and determining policy. Today there are 2,600 people in the field and about

3,700 people in D.C. Fifteen years has seen a 300% increase in the field and a 570% increase in Washington.

(7) All this, not to produce one product, or assist the growth of the industry, but to write tickets in the name of consumer protection.

(8) This leads interestingly enough to the subject of consumerists. Can FDA laws be so written and enforced so as to protect everyone against everything? I hardly think so. Even if it could - at what price? Yet in Washington, a vocal few in the name of consumerism are demanding just that. Some reasonable risk must be assumed in order to maintain a proper balance. What are the needs of consumers - notice that I said <u>needs</u> - not wants - <u>all</u> the wants of any group can never be met.

One need is research - research into her needs. However, that research never seems to get done. But who talks to the silent majority? Surely not the regulatory agencies, that would be self-defeating to their power structure.

In the last six years we have seen the professional consumer activist achieve a position of profound power in the U.S., while official Washington will never admit that the activists are getting more attention than is due their constituency.

Do the professional consumerists truly represent the

will and needs of the people? Are the regulatory agencies taking their cues from the right people or should they be making an effort to determine, through objective research, whether the professionals are right? I ask, therefore, what assurances do we have that the consumerists, no matter how well intentioned, truly represent the will and desires of the general public? These needs must be answered economically, as well as as in the name of public safety.

(9) Administration of the Freedom of Information Act under FDA has reached ridiculous heights. FDA has written their interpretations of the Act so broadly that with the exception of trade secrets, FDA will make available to the public any and all information whatsoever it receives from industry, either voluntarily or secured by the agency during an inspection. Yet they will not make public their internal memoranda which shows their decision making process. I thought the law was intended to show how the government works - not how industry works.

(10) There is no such thing anymore as a private meeting with FDA to work out problem areas - next Monday morning you can read all about it in Food Chemical News.

Talk about technological opportunities - listen to these news topic healines:

A. Red # 40 safety questioned.

B. Red # 2 petition rejected by FDA.

C. BHT may be switched from GRAS list.

D. FDA recall procedures revised.

E. PCB ban on Hudson River fish upheld.

F. Hyperkinesis study called too short.

G. Consumer representation hearings boycotted by consumer groups.

H. Vitamin deregulation bill hits snag.

I. Water recycling EPA proposal for food plants sparks FDA concern.

J. Industry GMP's urged as prep for food surveillance law.

K. Bologna should have no nitrite.

L. Folic acid - heart disease effect doubted.

M. Chlorine sanitizing agent use data & safety sought by FDA.

N. Drained WT. proposal endorsed by state consumer departments.

O. Wine labeling proposal to go to public hearings.

- - and on and on.

But what never gets said is that in 1974 only 9% of food-borne illness was attributed to the food processing industry - that, out of the billions and billions of pounds that were pro-

cessed.

You technologists that must deal with the realities of regulatory agencies must properly appraise this type of case study because it has a tremendous impact on your business and professional life.

In summary, the value of technology to the fishing industry is survival - without it, progress stops. We need all your best efforts.

Thank you for your attention. I appreciate this opportunity to present these views to you. May you have a most successful conference, and remember, you have the work technological in you conference title - wear it proudly.

THE FAO PROGRAMME OF COOPERATIVE RESEARCH IN TROPICAL FISH TECHNOLOGY

ABSTRACT

The Fishery Industries Division of FAO is promoting cooperative research in tropical fish technology by encouraging institutes already established in developing countries to pool their limited resources and work on common problems. Input will be required from institutes in developed countries for the transfer and application of low cost technology.

THE FAO PROGRAMME OF COOPERATIVE RESEARCH IN TROPICAL FISH TECHNOLOGY

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There is no question that needy populations in developing countries require additional supplies of animal protein. In many cases fish can contribute more significantly than land animals to closing the protein gap. At present the developing countries, with about half the world's population, consume about one quarter of available fish supplies. This is compared with about half the total supplies consumed by one fifth of the world population in the developed countries.

The most obvious avenues to increasing consumption in the developing countries are increasing production, improving utilization and reducing wastage and post-processing losses. In order to achieve these aims the application of technology by trained and experienced fish technologists is required.

Available resources

FAO publications indicate that a high proportion of the resources, which are classified as more easily exploitable, could be available to developing tropical and subtropical countries. In addition, estimates of more than three million tons have been made

for the by-catch, most of which is currently discarded, from shrimp trawling operations in these waters. However, even assuming that improvements in catching technology and increased fishing effort make it possible to land more fish, the problems would not be solved. In fact increased landings under present conditions would make the situation worse. Spoilage or incipient spoilage between catching and landing is the major constraint to increasing utilization. This is particularly so with small pelagic fish which could perhaps make the highest contribution to increasing fish consumption. If catches reaching the beach are not of sufficiently good quality for distribution to surrounding areas or for processing, then economic return to the fisherman is not possible and there is no incentive to increase catches. There are many complex aspects to this problem; social factors such as traditional market forces and low buying power of the population being particularly important. This means that all inputs of technology which are considered must be evaluated on economic grounds, against the background of what the population can afford because this will determine the extra efforts that the producers can make.

The role of technology

A big question is whether it is necessary to develop specific technology for tropical fisheries. This implies a major research effort, which would have to be centred in the tropics. The spin-off from the research is assumed to be a technology which could be applied in tropical situations. It is conceivable that the establishment of an international tropical fish technology research

institute, along the lines of those for rice and cereals, could make major advances. However, this is a very distant dream and the usefulness of such an institute, before the foundations of tropical fish technology are established, could be questioned. An alternative approach, which is much more reasonable at the present time, is to review improvements in fish technology which have been made in the 70 year history of the subject, and to selectively apply suitable technology in the tropics, to provide a firm base for development. Many of the failures in fishery development projects in developing countries can be ascribed to the indiscriminate introduction of complicated technology, which has arisen in the northern hemisphere specifically for application in large-scale North Atlantic and Pacific fisheries. Among the reasons for failure are the expense of the equipment and its complicated nature, which makes it impossible to service in unsophisticated situations. In addition, the nature of the fishery is completely different. In the tropics numerous small vessels catch a wide range of species, whereas large vessels make predominantly single species catches in northern waters.

Despite these limitations it can be fairly stated that enough technology exists, or can be made available from developed countries, for its selective application to be of benefit in tropical fisheries. What is essential is that it be matched to the economic and practical requirements of the recipients. In following this approach fish technology institutes in developing countries should become centres for the transfer and dissemination of technology; taking it from where it has been developed, to their own industries. Unfortunately there is a tendency, perhaps because of national prestige and the

search for scientific excellence, for developing countries to build up sophisticated research capabilities, the results of which cannot even be applied in wealthier societies. There are a growing number of institutes in developing countries which can serve their industries best by adapting technology to the specific needs.

Acceptance of this approach throws a burden on to institutes in developed countries, i.e., the originators of the technology. Some way must be found to channel this work to where it is required and where it can be applied. The FAO is in an ideal position to act as an effective interface between the developed and the developing world. However, because the Organization cannot contribute new technology it must rely on the originators to make their ideas and experience available for the benefit of the third world. The FAO Programme of cooperative research in fish technology is an attempt to build linkages between fish technology institutes all over the world and to transfer technology through this collaboration.

The success of this venture will ultimately depend on the inputs of technology and money from developed countries and will be measured by improvements in nutrition and living conditions. However, before embarking on too ambitious a programme it is advisable to consider how much technology can be absorbed by the third world and what research is required.

Research requirements

The need for comparing complexity and cost with economic viability has already been expressed. The two most important

conditions for ensuring survival and implementation of advances are: they should be simple and they must be cheap. The search for reducing energy input in fish handling and processing must continue as, especially in the developing world, energy is an increasingly expensive item. The major source of wastage and loss of quality; the period between catching and landing, can to a large extent be eliminated by the proper application of chilling. The fisherman, however, in almost all circumstances, bears the brunt of the cost-price squeeze. If he cannot recover the cost of ice or refrigeration, is there an alternative to the present situation, where much of the fish landed has only a very short residual storage life? This situation applies particularly to the small pelagic species which constitute the bulk of the resources which may be available for exploitation. At present low intrinsic value, coupled with high labour cost for processing, result in lack of incentive or wastage. If chilling is not possible alternatives such as salting on board for drying ashore, or adjustment of the logistics of catching and landing for immediate processing or distribution, should be considered.

Artificial drying is an attractive prospect to achieve high volume throughout, independent of climatic conditions. However, reliance on electric power or oil is energy intensive and other sources of heat should be investigated. New directions in the use of solar energy or the harnessing of waste heat, e.g., from stationary diesel engines, could substantially reduce drying costs.

Another area which urgently requires investigation is the

losses of product, particularly dried fish, from insect infestation in storage. It is generally accepted that at least 30 percent of dried fish is consumed by insects, such as beetles, between the time of production and when it reaches the consumer. As dried fish is the most important processed product available to the low-income group in developing countries, very significant losses of protein result from this wastage. In the long term these losses must be made up by increased catches. The whole area of prevention of waste and increasing the rate of utilization becomes important if one assumes that diversification of fishing effort, to unexploited resources, will be more expensive than fishing the resources currently exploited.

There are numerous other areas of work which could provide a springboard for the practical application of research results. The effect of good sanitation, on fishing vessels and in storage, on the keeping qualities of fish and investigation of the great differences between the microbial flora of tropical and temperate species, are examples. These investigations would not be difficult to carry out but application of the results could prove to be the stumbling block as it has in the past. The above points relate mainly to improvements which could be effected in existing small-scale fisheries. All efforts to promote the development of these fisheries should be made in order to improve employment prospects and encourage economic stability.

Another approach in the face of world food shortages is to use the protein from the sea more wisely by taking a global approach.

- Low cost products from minced fish
- Investigation of microbial spoilage associations during storage of fresh tropical fish
- Ensilage of fish processing waste and surplus fish for animal feeding
- Potential toxicity problems with fish from the IPFC Region.

The institutes involved are almost exclusively in developing countries but it is evident that this programme cannot be sustained without inputs from institutes in developed countries. It is hoped to expand the programme with support, which has been promised, from institutes outside the region. More assistance is however required and FAO is always eager to learn of other institutes which would be prepared to participate. In addition to assisting in direct development action, it is felt that expansion of the programme will promote closer links between fish technology institutes all over the world. In coming years it is hoped to include West Africa and Latin America in the programme. In West Africa work is likely to be focused on handling and processing methods for small pelagic fish. The diversity of resources in Latin America will involve a sub-regional approach. A symposium on merluza handling is planned for the southern countries; while for the Caribbean area it is hoped to start a cooperative programme within the WECAF countries. This will be coordinated with industry, governments and FAO with cooperating institutes to explore promising alternatives for utilizing shrimp by-catches.

Other initiatives which have been taken to improve communication between technologists are the preparation of an International

Directory of Fish Technology Institutes and the planning of a series of information circulars. These would outline directions in development aid and the programmes of various institutes.

In conclusion, it can fairly be said that there is a need for technological improvements to be made available to developing fisheries. In addition to the benefits offered by industry links, such as joint ventures, there is a role for collaborative efforts between institutes. Better communication and closer cooperation at the institute level can be effective in the transfer and application of technology.

THE SPOILAGE OF FISH IN THE TROPICS

ABSTRACT

The special problems associated with the handling and distribution of fresh fish and crustacea in the tropics are reviewed. Attention is drawn to some of the known differences between tropical species and temperate or cold water species. The commercial implications of these differences are considered with special reference to the small scale fisheries situation which occurs in many developing countries. The need for more research on the handling and processing of fish in tropical areas is emphasized.

THE SPOILAGE OF FISH IN THE TROPICS

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Introduction

Fish represents an important source of protein in many developing countries although its importance in the food economy varies from country to country. Furthermore, many of the known underexploited fisheries resources are to be found in tropical areas and the utilization of these, and existing resources, represents a challenge to the fisheries industries in developing countries. An even greater challenge, which must be solved if nutritional levels are to be raised in developing areas, is the need to make maximum use of the catch. For this reason fish utilization, particularly the prevention of waste, is becoming increasingly important, especially in the tropics, where the losses may be considerable. Aid agencies, both multilateral and bilateral, are very concerned about food losses in developing countries and, since the losses are greatest at the rural level, more effort is being directed towards the problems of small scale fisheries.

The development of small scale fisheries in tropical areas

is extremely difficult. Severe economic, social and political constraints are involved as well as the many technological problems which this paper will touch on.

The special problems associated with utilization of fish in the tropics were considered at the FAO Conference held in Tokyo in 1973 (Disney et al., 1973). The known differences between cold water and tropical fish in terms of their handling and processing characteristics were discussed. A plea was also made for much more research on tropical fish to supplement the comparative lack of information on warm water fish.

The present paper will deal largely with the experiences of staff from the Tropical Products Institute, who have worked primarily on fish from Africa and the Far East. The author has no knowledge of the geographic area being considered by this Conference, but information gained in other parts of the tropics may be relevant to the situation in Latin America and the Gulf of Mexico. The paper will deal largely with the handling and distribution of fresh fish and will consider the benefits to be derived from using ice in tropical fisheries. In line with the main experience of the author the discussion will be largely directed towards the problems of the small scale or artisanal fisheries situation which pertain in many developing countries all over the world.

Spoilage at ambient temperature

The rapid spoilage of fish at the high ambient temperatures

in the tropics is a well known phenomenon. Fish spoil within a few hours unless some form of preservation is applied and in certain types of artisanal fisheries, e.g. gill netting, fish are spoiled even before they are cleared from the net (Lima dos Santos, 1973). Although the rapid spoilage is well known, there are very few documented investigations. Ilyas and Nasran (1972) have reported on three species of fish in Indonesia which were rejected by organoleptic and objective tests after approximately 11 hours. African fresh water fish are also considered unacceptable at 24 hours after capture (Hoffman et al., 1974). A further study in West Africa by Kordyl and Karnicki (1969) confirms this view but there are no reports dealing with the causative agents of this rapid spoilage. It is reasonably assumed that the deterioration is due to rapid bacterial growth but the relationship between bacterial enzymes and autolytic enzymes is largely unknown. One experiment in our own laboratories revealed levels of bacteria between 10^3 and 10^6 per g of tissue in Tilapia after 24 hours when the fish were completely spoiled (Disney et al., 1968).

Spoilage in Ice

The most significant improvement in fish preservation in the tropics will probably result from the increased use of ice in the fresh fish trade. However, compared with the wealth of data on the iced storage of cold water species, there is little

information available on the storage life in ice of tropical fish. The information that is available suggests that tropical fish have a much longer shelf life in ice than temperate or cold water species.

A review of this subject referred to earlier (Disney et al., 1973) compared the acceptable shelf life of ten cold water species with twelve tropical species. The cold water fish remain acceptable for 5 to 15 days depending upon the species but the tropical fish kept for 7 to 45 days. A number of subsequent reports have confirmed the longer keeping times of tropical fish when stored in ice (Tambunan and Rijal, 1972; Nair et al., 1974). These reports (some twelve references in all) refer to fresh water and marine fish from Africa, India and South East Asia. A further six marine species have been investigated in West Africa and North Borneo by TPI staff. This data has not been published but the shelf life ranged from 10 to 28 days depending upon the species.

Similar studies may have been carried out in Central and Southern America but the author is unaware of such reports. Nevertheless, it is reasonable to assume that fish from warm waters will have an extended shelf life when compared with colder water species.

The bacterial flora on fish is a function of the environment in which they are caught and subsequently stored. The number and type of bacteria, therefore, vary considerably

between tropical and cold water areas. Tropical species have few psychrophilic types present (unless these are introduced with the ice) and hence have a reduced spoilage potential when stored in ice. The mesophilic organisms on tropical fish are adapted to high ambient temperatures and the large drop in temperature on icing (up to 30°C) has a more profound effect than the smaller temperature drop for cold water species (as little as 4°C) (Disney et al., 1973; WHO, 1975). The reason why some tropical species e.g. pomfret spoil comparatively quickly (7 days) is not known and requires investigation.

The temperature differential and its effect upon bacterial growth is reflected in the lower total viable counts reported for tropical fish. Tilapia, for example, has been found to contain 10^6 organisms per g of flesh after 28 days storage in ice whereas cod is reported to have as many as 10^{10} bacteria per g after 15 days (Disney et al., 1969). Similar quantitative differences have also been reported (Nair et al., 1974; Tampoebolon, 1972) but few qualititive studies on tropical fish have been published. More information is required particularly concerning the relationship between the skin flora and that in the flesh.

The extended shelf life of tropical fish in ice may also apply to crustacea. Despite the commercial importance of shrimp exports from the tropics relatively few studies have been carried out but reports from India and SE Asia suggest that tropical shrimp keep longer in ice than colder water species.

Cann (1973) in his review on tropical shrimp indicated that penaeid shrimp from the Gulf of Thailand remain in acceptable condition for 12-16 days in ice whereas non-tropical shrimp such as Pandalus and Nephrops species are totally spoiled after 8-10 days. This difference was again attributed to the bacterial flora; the mesophilic flora on the shrimp are not active at ice temperatures and little spoilage occurs until a psychrophilic flora develops. The argument was put forward that the degree of spoilage may be related to the degree to which psychrophilic strains are introduced with the ice. This may be particularly relevant to developing fisheries. The comparatively long shelf life of iced shrimp from tropical waters has also been reported by Carroll et al. (1968), Clucas (1971) and Abdurrahman and Abdurrahim (1973).

Tropical shrimp may have a longer iced storage life than other shrimp caught in colder waters but this is conditional upon adequate ice being used. In developing fisheries, unfortunately, this is rarely the case. Shrimp exports from South East Asia to Europe, Japan and the United States have been severely criticised in the recent years largely due to poor microbiological quality. The cause of this poor quality in a country in SE Asia was recently investigated by TPI and two problems were identified. High bacterial levels result primarily from poor handling prior to processing and the contamination by pathogens results from bad processing practices and unhygienic handling in the processing plant. The latter may be remedied

by improved hygiene and better technical management but the former requires a complete revision of the fishing industry. As indicated earlier, small boats are the principal source of shrimp in SE Asia and the large scale use of ice is both impractical and uneconomic in many situations. This is in no way an excuse for the poor microbiological quality of exports from some countries but it is merely an attempt to illustrate the problems and the need for improvement. Where larger vessels with proper facilities and improved transport facilities are available these problems may be solved with relative ease but the small scale fishery has an inbuilt resistance to improvement.

Delays prior to icing

Bad practices in the icing and handling of shrimp have been shown to have serious consequences in the export trade from SE Asia. Similar problems can arise in the fresh fish trade whether it is for local consumption or for export.

Ice should be applied to fish as soon as possible after capture. This is perfectly feasible in larger vessels as indicated above but in small scale fisheries the situation may be very different. Ice may not be available, it may be too expensive or more commonly the size of the boat may prevent the carrying of ice; the latter is very common in the one man/one canoe/artisanal fishery. However, because of the small

size of the boat the catch is usually landed within 6 or 9 hours of capture. Unpublished work carried out by TPI in Ghana and Malawi has shown that delays of up to 9 hours prior to the application of ice are permissible. Two marine species exanined in Ghana were acceptable to a taste panel after 12 and 18 days despite a 9 hour period at ambient temperature prior to icing A shelf life in excess of two weeks, after limited periods at ambient temperatures, has also been reported by Tampoebolon (1972) in Indonesia, Herborg (1971) in Trinidad and in India (CIFT Annual Report, 1969).

Such delays may not represent ideal practice but for domestic consumption in developing countries practices including delays prior to icing may represent a significant advance.

The effect of gutting

Temperate or cold water fish are generally considered to keep better in ice when they are gutted, although some fatty species may be exceptions. Published reports on tropical species are conflicting but a number of reports (Disney et al., 1973; Herborg, 1971; Tambunan et al., 1972) suggest that the benefits of gutting are only marginal. Indeed gutting would not seem to be justified for short term storage and on long term storage the extra shelf life may be as little as two or three days. In many small scale fisheries effective gutting cannot be guaranteed and may do more harm than good. Also in

many situations the consumer demands the whole fish and it is doubtful if gutting should be recommended.

Commercial implications

An attempt has been made to illustrate the differences between tropical and temperate or cold water species in terms of their handling and storage characteristics in ice. These differences can have important commercial implications. In the advanced fishing industries of North America, Japan and Europe the implications may be minimal (although far more research is needed on such aspects as bleeding and rigor mortis). In developing fisheries with widespread use of ice at sea and a well developed distribution system the implications also may be relatively few but in developing fisheries based upon small boats and without an established cold chain for fish distribution the effects could be very significant.

The transition from an artisanal type fishery to a mechanical industrialized fishery may be justified economically but there are many sociological complications, particularly the loss of employment. Aid agencies are currently paying considerable attention to the improvement of small scale fisheries and are attempting to avoid these sociological difficulties. The extended shelf life of tropical fish and the possibility of adding ice some time after capture could considerably assist this type of development. An acceptable shelf life of 2 to 4

weeks, depending upon the species, should be sufficient to permit distribution over considerable distances or make it possible to hold short term buffer stocks against deficiencies caused by day to day fluctuations in catch, which often occurs in the tropics due to rapid changes in the weather. In certain situations the long shelf life may favour the establishment of a distribution network of iced fish rather than the development of a central freezing facility and frozen distribution network. The possibility of storing fish in ice after a period at ambient temperature also permits the provision of ice at centralized landings whilst avoiding the need to take ice on the boat. This could be particularly important in artisanal fisheries, although where possible the immediate application of ice should be encouraged.

The potential benefits of using ice in a small scale fishery may be illustrated by an account of the author's recent experience in North Borneo. The fish supply is caught almost entirely by small inshore boats. Although they all have outboard engines it is impractical to take ice to sea. Conversely the fish are landed within a few hours of capture and the provision of centralized landing facilities including an ice plant will represent a significant improvement. Ice is used in limited quantities in the markets to preserve unsold fish for sale the following morning. At present no fish is kept in ice for longer than 1.5 days and the storage life has never been determined. An experiment was carried out on 5

commercially important species and the minimum shelf life (after 4 hours at ambient temperature) was found to be 10 days for rastrelliger (club mackerel). After 15 days, when the experiment was terminated, it was clear that the other 3 species would remain acceptable for a longer period. At ten days commercial fish traders were invited to inspect the fish, all of which were considered saleable. The refused at first to accept that the fish were 10 days old but subsequently they appreciated the potential benefits of this information. This illustrates the need for on the spot demonstration of the benefits of technology. Fortunately the need for such extension work and the need for advisory notes and information suitable for distribution at the rural level is now being recognized by multilateral and bilateral aid agencies. The benefits of this change in emphasis could do much to improve fish preservation in small scale fisheries in the tropics.

Research requirements

An attempt has been made to illustrate that technological differences exist between tropical and cold water fish in terms of their spoilage characteristics during iced storage. However, relatively few species have been studied and there is an urgent need for more information. The bacteriology and biochemistry of chilled tropical fish and the effect of chilled brine on keeping quality are largely unknown. Two areas in

which almost nothing is known on tropical fish are the effects of bleeding and the development of rigor mortis upon the quality of fresh and processed fish. Blood discolouration is not known to be a major problem in tropical fisheries but information is required on the relative merits of bleeding before icing, particularly in situations involving large fish. The relationship between the quality of fresh tropical fish and that of the products of secondary processing is also an area where little is known.

What information is known about tropical fish suggests that commercial practices, based upon cold water fish, may have to be revised for use in tropical fisheries. Codes of Practice and Codex standards as proposed by FAO and WHO although generally applicable throughout the world may also have to be modified to take into account the technological differences of tropical fish. This complex issue was recently reviewed by Jones et al. (1975).

Conclusions

The research effort required to resolve some of these problems and provide the information required cannot be mounted by the developing countried alone as they do not possess the necessary manpower or facilities. Much of the research required must be carried out by technological laboratories in the developed countries and organizations like FAO are actively promoting regional research programmes in both developed and

developing countries. TPI is also pursuing a limited programme along similar lines.

In this context, national, regional and international meetings of fisheries technologists are particularly relevant. This first annual Tropical and Subtropical Fisheries Technological Conference provides a forum for the presentation of information on tropical fisheries and will hopefully promote research programmes designed to investigate some of the gaps in our knowledge, including those considered above. I congratulate the organizer for arranging this Conference to support the existing Atlantic and Pacific Conferences.

I would also like to take this opportunity of bringing another Conference to the attention of the delegates. An International Conference on the handling, processing and marketing of tropical fish is being organized by the Tropical Products Institute to be held in London in July of this year. This meeing will concentrate upon small scale fisheries and some 68 papers will be presented on the various post-harvest aspects of fisheries development in the tropics. We hope that this Conference will make a valuable contribution to fisheries development in developing countries.

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CELLULAR ASPECTS OF REPRODUCTION IN PENAEID SHRIMP

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INTRODUCTION

With the increased interest in penaeid culture over the past few years a demand has arisen for seed stock. Such demand necessitates the catching of gravid females in their wild habitat. This is true since female shrimp of the genus Penaeus are very reluctant to undergo gonadal maturation and spawning in captivity.

For this reason our laboratory as well as several other groups have been investigating the reproductive processes in penaeids. Our preliminary investigations have demonstrated that penaeid gametes are quite atypical when compared to the gametes of other marine invertebrates. Variations exist not only in the morphology of the germ cells but also in the chemical composition and physiological reactions of these cells.

The purpose of this paper is to review some of the more striking atypical characteristics of both the sperm and eggs.

MATERIALS AND METHODS

Penaeus aztecus and P. setiferus were used for the following study. P. aztecus were caught 80-100 miles south of Galveston, Texas at approximately 95°00' longitude and 28°15' latitude using commercial trawling equipment. P. setiferus were caught from 1-10 miles off the coast of Galveston, Texas at 95°00' longitude and 29°15' latitude. Animals were held in 150-gallon aerated tanks at

15-20° C until transported to the laboratory. Gravid females brought to the laboratory were placed in aerated, inverted carboys for spawning.

Oocytes from spawning females and sperm removed from the terminal ampoules of males were examined under a phase microscope or fixed for light and transmission electron microscopic examination. All cells were fixed in a paraformaldehyde-glutaraldehyde fixative (Karnovsky, 1965) for 30 minutes and post-fixed in 1.0% osmium tetroxide buffered with 0.1 m phosphate (Millonig, 1961) for 15 minutes. Fixed cells were dehydrated in a graded acetone series, embedded in a low viscosity epoxy resin (Spurr, 1969), and sectioned on an ultramicrotome with glass or diamond knives. Thin sections were mounted on uncoated grids, stained with saturated alcoholic uranyl acetate and lead citrate (Reynolds, 1963) and examined with an electron microscope. Thick sections, 0.5-1.0 μ, for light microscopy were stained with 1.0% aqueous methylene blue in 1.0% sodium borate.

For scanning microscopy, cells were fixed in a paraformaldehyde glutaraldehyde fixative (Karnovsky, 1965) and dehydrated in a graded acetone series. Dehydrated cells were critically point dried and coated with carbon and gold in a vacuum evaporator. Specimens were examined with a scanning electron microscope at 5-20 Kv.

RESULTS AND DISCUSSION

Sperm:

At the light microscopic level, the mature sperm looks very much like a "golf ball sitting on a tee" (Fig. 1). That is, the cell is composed of a spherical body with a cap and protruding spike at one pole. The body is 3.6 μ in diameter and the spike is 2.5 μ in length, 0.8 μ in diameter at its base, and 0.3 μ in diameter at its apical end. Live sperm examined with the light microscope exhibit no motility.

For descriptive purposes, the mature sperm will be divided into four gross morphological components: (1) the head; (2) the cap; (3) the base; and (4) the spike (Fig. 2). With the scanning electron microscope, the topographical features of the sperm can be

seen with greater clarity (Fig. 2). The membrane covering the head of the sperm contains numerous pits or pores approximately 700 Å in diameter. This is in contrast to the smooth, unpitted surface membrane covering the cap, base, and spike.

At the fine structural level the head is composed of a central nucleoid-like region surrounded by a cytoplasmic band (Fig. 3). The nucleoid contains a network of fine fibrils 60-180 Å in diameter, which appear closely associated with dense rod-like elements 300 Å in diamater. The cytoplasmic band contains a few mitochondria, some membrane fragments, and numerous vesicular bodies filled with an electron dense substance. In some instances these vesicles appear to be dehiscing (Fig. 3), and may represent the pores seen with the scanning electron microscope; however, this is difficult to substantiate since the membrane continuity is poor around the cytoplasmic band. The cytoplasmic band terminates at the cap and a zone of electron dense granular material is found between the cap and the nucleoid region. The cap contains closely packed dense granules. In the center of the cap is the base of the spike. This base is composed of very fine dense granules and appears to give rise to the filamentous spike. The filaments of the spike are 60 Å in diameter and are tightly packed in longitudinal arrays (Fig. 4).

In view of the above information it becomes readily evident that the penaeid sperm is quite atypical when compared to the more common flagellated sperm. The penaeid sperm is non-motile, lacks any structure common to motile cells (e.g., cilia, flagella, or pseudopodia), contains an uncondensed nucleus, and possesses no mitochondrial aggregate.

However, in spite of these unusual features certain homologies may be drawn between the penaeid sperm and more "typical" sperm common to other animals. We believe the cap represents the acrosome of the sperm. This assumption is based on two characteristics common to acrosomes. First, the cap is PAS positive in nature (Brown, et al., 1976); and second it exhibits an orange fluorescence when stained with acridine orange (Clark, et al., 1973). Extending from the center of the cap is the spike. Pochon-Masson has noted similar spikes in the sperm of Crangon vulgaris (1968)

and Palaemon elegans (1969) and believes them to be the acrosomes per se. We disagree with Pochon-Masson. Since the spike contains dense bundles of microfilamentous material, we feel it may be analogous to a fertilization filament and not unlike the acrosomal processes described in the flagellated sperm of numerous marine invertebrates (Tilney et al., 1973, and Tilney, 1975). This hypothesis is strengthened by the fact that the spermatozoa of penaeids contains significant levels of actin, as determined by tube gel electrophoresis, and its spike exhibits distinct fluorescence when treated with fluorescein labeled actin (Brown et al., 1976).

Eggs:

A mature, unreacted ovum removed from the oviduct of a spawning female is spherical and approximately 250 μ in diameter (Figs. 5 and 8). Large accumulations of yolk platelets are apparent throughout the cytoplasm of the oocyte. The large cortical specializations which reside in the peripheral cytoplasm are the most conspicuous and unusual feature of these eggs (Figs. 8 and 9). These specializations (cortical rods) are club shaped, 40 μ in size, and lie within crypts (Fig. 9) formed when the membranes bounding the rods fuse with the oolemma prior to the initiation of spawning (Duronslet et al., 1976). Closely apposed to the external surface of each egg is a thin investment coat (Fig. 9).

When an egg is spawned and contacts sea water, the cortical rods are rapidly expelled from their crypts and force the investment coat off the surface of the egg. As this expulsion continues the investment coat looses its integrity and falls away from the egg. The cortical rods completely move out of their respective crypts and form a corona around the periphery of the newly spawned ovum (Fig. 6). However, after approximately 8 minutes the corona (composed of expelled rods) dissipates and is lost to the surrounding media (Fig. 7).

In a recent study Lynn and Clark (1975) and Clark et al. (1975) have found that this massive cortical reaction (cortical rod expulsion) is Mg^{++} dependent.

The cortical reactions in the eggs of P. setiferus and P. aztecus are unusual. These unusual features are: (1) the enormous size of the cortical specializations (rods) in the peripheral cytoplasm (2) the rapid expulsion and dissipiation of these cortical rods on contact with seawater; and (3) the Mg^{++} dependency of the reaction.

Only a few examples of such a massive and rapid cortical release as seen in the penaeid shrimp are reported in the literature. A reaction, morphologically similar, to the one displayed in the eggs of penaeid shrimp occurs in the eggs of some annelids (Lillie, 1911; Wilson, 1892; Fallon and Austin, 1967; Novikoss, 1939a and 1939b; and Costello, 1948). A jelly-like material is contained in large alveolar vesicles in a broad band of cytoplasm at the surface of the annelid eggs. The alveolar material is extruded at fertilization and forms a layer around the egg and remains there throughout the early development of the zygote. Teleost eggs also undergo a massive cortical reaction comparable in magnitude to penaeid egg reactions (Yamamoto, 1939). However, the teleost egg retains the extruded jelly-like material beneath a vitelline membrane, and the material released is maintained as a protective coating around the eggs and developing embryos. This is in contrast to the rapid dispersal of the substance released from penaeid eggs.

The Mg^{++} dependent release of the cortical rods from shrimp eggs is quite unlike the Ca^{++} dependent reactions reported in other animals ova to date (Mazia, 1937; Yamamoto, 1939b; Epel, 1975; and Paul, 1975). Vacquier (1975) and Epel (1975) have suggested that enzymes released from cortical specializations in sea urchin eggs are activated by the divalent cation Ca^{++} released intracellularly at fertilization. Mg^{++} may have a similar function in penaeid eggs; that is, it may either activate an enzyme or act as a cofactor for an enzyme released upon egg contact with seawater.

Several possible roles for the cortical reaction in penaeid eggs exist. Some of these are: (1) a block to polyspermy; (2) the formation of an egg jelly to act as mechanical barrier; and (3) a chemical defense mechanism.

The removal of supernumerary sperm from the surface of ova has been demonstrated to be the result of a cortical reaction at the time of fertilization in many animals (Epel, 1957 review and Paul, 1975). Many sperm, attached to the shrimp egg investment coat, are lifted off the surface of the ovum during the expulsion of the rods. However, the removal of supernumerary sperm from the shrimp ovum does not appear to be the primary function of the cortical rod release since the reaction is initiated by contact with seawater and not by sperm penetration.

The formation of a protective layer against mechanical damage may also be a possible role. Such a layer might function, like the egg jelly or investing layers around many other animal ova, as protection against an unstable environment. However, our studies indicate that the cortical rod material rapidly dissipates leaving the egg vulnerable until a hatching membrane is formed during later development.

The development of a chemical defense barrier against microorganisms is a third possible role for the cortical rods. Although morphologically the rods are rapidly broken up, the chemical constituents may remain around the ova for a long period of time. In this form, the chemical constituents could act as an antibacterial agent or as a repellent to other types of microorganisms. Current investigations suggest that such a role is likely.

Our laboratories are continuing to work on this system and hopefully, some of the myriad of unanswered questions will be elucidated in the future.

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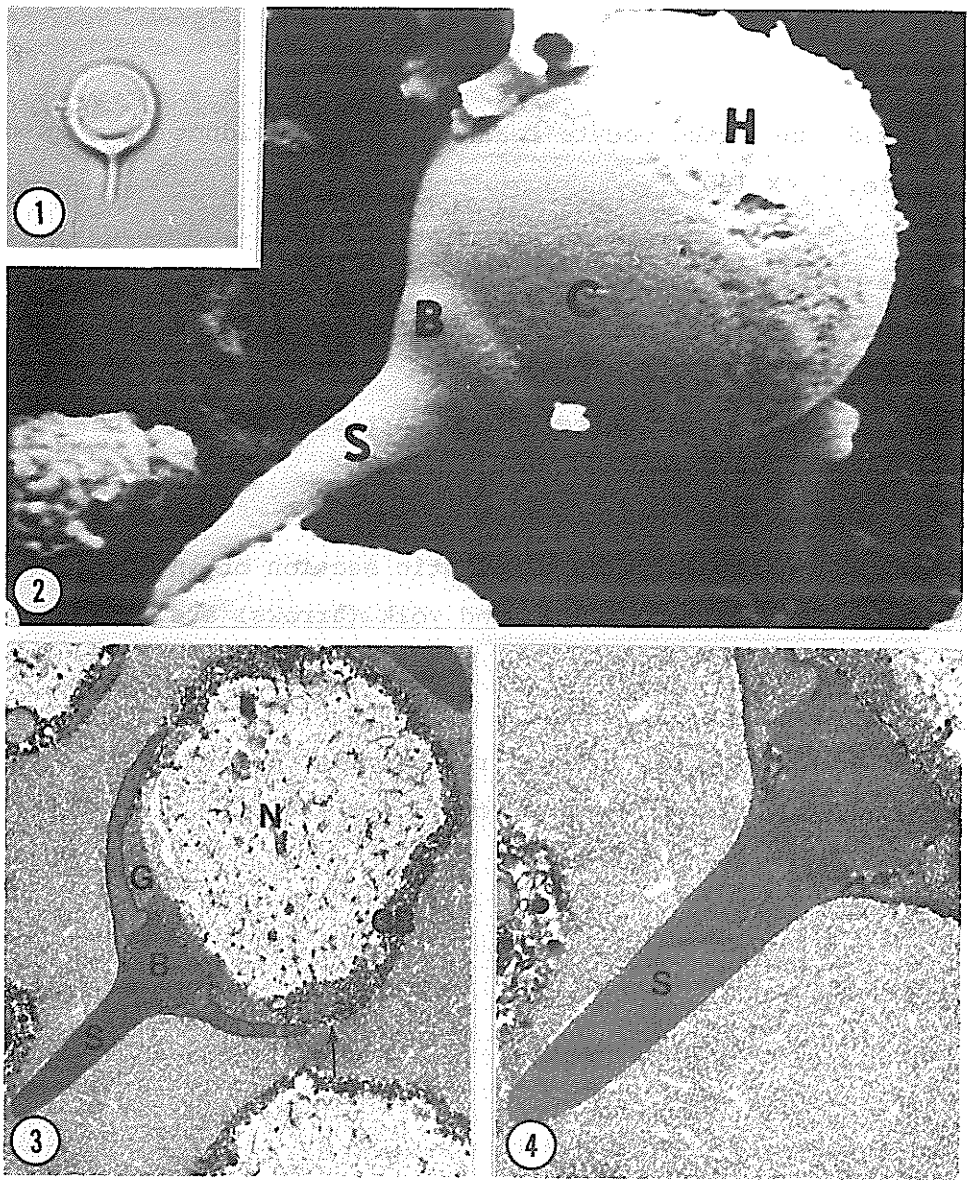
1. A phase micrograph of a mature sperm. X 1,600

2. A scanning electron micrograph of a mature sperm showing head (H), cap (C), base (B), and spike (S). X 15,600

3. An electron micrograph of a mature sperm illustrating the nucleoid region (N), cytoplasmic band (CB), electron dense granular zone (G), base of spike (B), spike (S), and arrow pointing to a dehiscing vesicle. X 9,700

4. A high magnification showing the filamentous substructure of the spike (S). X 19,700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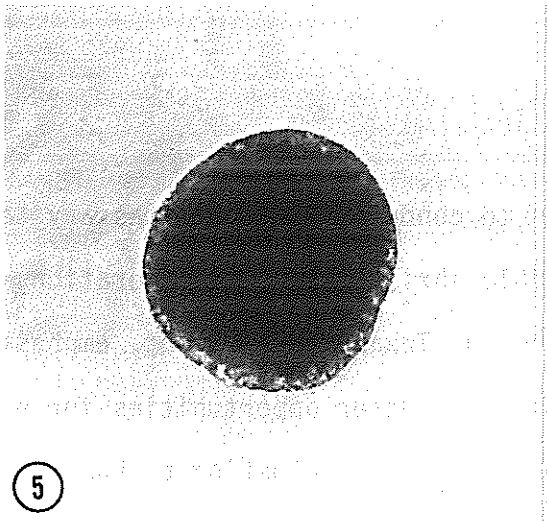
5. Unfixed, unreacted oocyte teased from the oviduct of a spawning female. X 140

6. Phase micrograph of an egg undergoing the cortical reaction. Corona of cortical material is apparent (C). X 110

7. Phase micrograph of an egg whose cortical rods have been expelled and dissipated. X 110

8. Light micrograph of a thick plastic section of an unreacted oocyte. Cortical rods (CR) and yolk (arrows) are apparent. X 380

9. Light micrograph of cortical rod showing typical club-shape (CR) and investment coat (arrow). X 1,000



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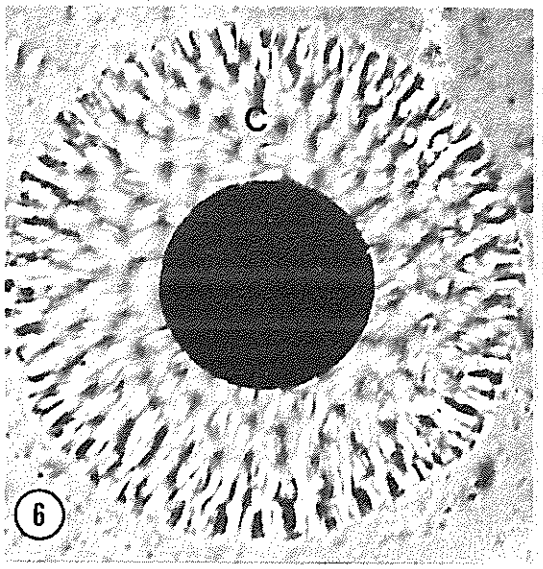

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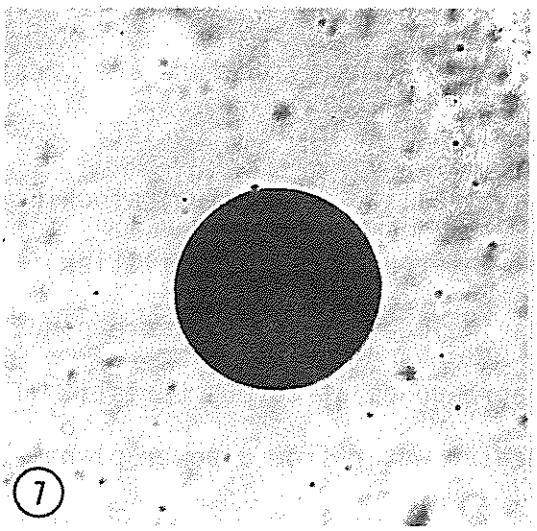

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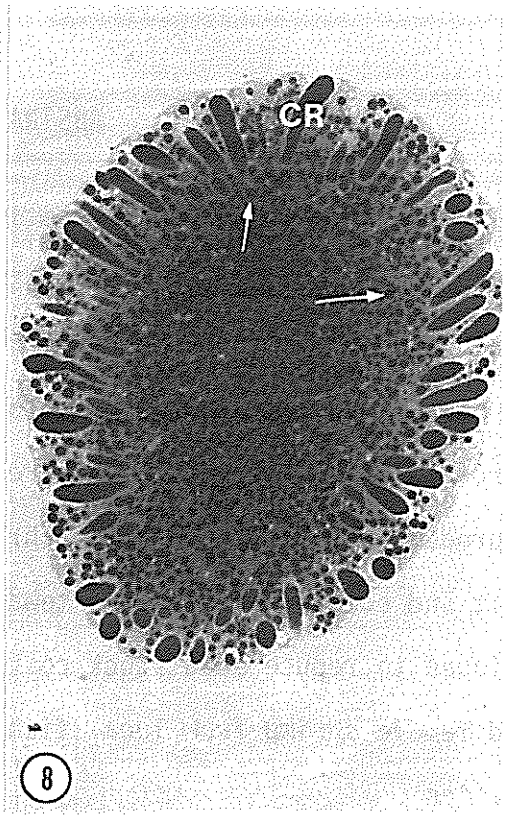

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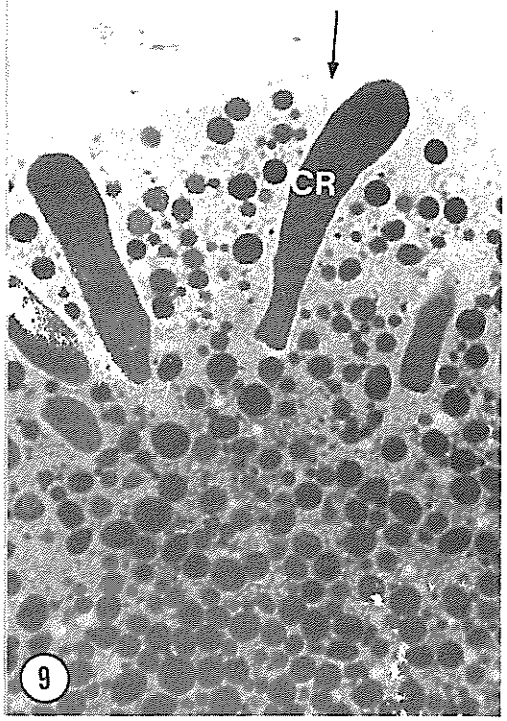

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TEXAS A&M UNIVERSITY SHRIMP MARICULTURE PROGRAM

ABSTRACT

The objective of this program is to conduct research necessary to the development of commercially feasible shrimp production operations designed to capitalize on the resources of Texas. At present, emphasis is on penaeid shrimp production because of better opportunities for early commercialization. Efforts are directed toward evaluating technology for pond production systems to raise shrimp from hatchery-produced postlarvae to bait and edible market size, toward exploring simplified hatchery techniques to reduce seed stock costs, and toward developing sources of gravid female shrimp from local fishing grounds and through controlled maturation.

The program is directed toward those areas where information is seriously lacking and where Texas A&M University can best use its resources and personnel. The intent of the program is to design a production system which will allow the individual farmer to operate both hatchery and growout units.

Pilot production units for low and high density culture are located at the Mariculture Centers in Brazoria County and Corpus Christi. Research is currently directed to determining suitability of species, stocking densities, feeding rates, and rates of water exchange. Facilities in addition to the pilot production units include twenty half-acre ponds in Brazoria County and eighteen quarter-acre ponds at Corpus Christi. The Corpus Christi ponds have access to heated water from a power plant and

are utilized to develop technology for maturing shrimp in captivity. To explore the use of underground salt water, two quarter-acre ponds located near Monahans in West Texas are being utilized. The National Marine Fisheries Service hatchery at Galveston is used to produce seed stock for the grow-out facilities and evaluate hatchery technology and production costs.

TEXAS A&M UNIVERSITY SHRIMP MARICULTURE PROGRAM

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This program is designed to assist in the development of technology needed to establish commercially feasible systems for farming penaeid shrimp in Texas and was initiated through the Texas A&M University Sea Grant Program.

There is more enthusiasm for the commercial culture of penaeid shrimp today than ever before. This is a direct result of improved culture technology, expansion of a world-wide market, attractive market prices and continued financial stress on the fishing industry. Much remains to be done (Giannini, 1972) before profitable culture operations materialize in the United States, but there is little doubt among both researchers and businessmen that farms supplying both bait and edible size shrimp will be a reality in the near future.

The development of this new farming endeavor means an additional use for thousands of acres of land and a significant expansion of the economy.

Because warm waters are required for shrimp growth, early development of aquaculture will be confined primarily to the Southern United States. Pond systems will be utilized for a major part of the grow-out phase and

will necessitate location of facilities in areas where vast acreages of low cost land are available and where there is access to naturally occurring salt water. The rapid growth rate of shrimp makes it possible to produce a marketable crop in about 140 days and allows for the production of a succession of crops from a single production facility. By starting postlarvae shrimp at high densities in heated raceways the growing season can be extended by as much as two months. Raceways utilized in combination with a graduate series of ponds can provide as many as five to seven crops each year between February and November. Considering that production of up to 4,000 lbs per acre per crop has been demonstrated it is realistic to assume that shrimp aquaculture will play a major role in the future development of the shrimp industry. As technology for intensive culture improves heated raceways and tanks may be used throughout the production cycle, thus allowing production possibilities in colder climates and areas without naturally occurring salt water.

It is anticipated that most shrimp farmers will come from the traditional agricultural community, since current technology closely parallels that in other agricultural areas. There is, however, considerable corporate interest which will likely materialize into production systems. Probably the most significant benefit will be realized by processors and consumers, who consistently point up the need for new sources of shrimp.

Contribution of facilities valued at more than $300,000 at Angleton and Corpus Christi by the Brazoria County Commissioner's Court and Central Power and light Company are ample evidence of the interest in shrimp culture along the Texas coast. A grant of $30,000 by Booth Fisheries has provided

for construction of additional pilot systems to encourage interest in shrimp faring and ultimately provide an increased supply of shrimp for Booth's markets. Continued interest by Ralston Purina, which contributed over $20,000 in seed stock this past year, is indicative of the potential foreseen for shrimp farming in Texas. In addition, last year the Texas Legislature appropriated funds to support a full-time mariculture specialist for the Texas Agricultural Extension Service in Corpus Christi and the Brazoria County Commissioner's Court appropriated one-half the salary of the salary of the mariculture specialist located in its area.

PREVIOUS RESEARCH

Hatchery Production

Larvae of the brown shrimp (Penaeus aztecus) were first cultured in the United States in 1963 at the National Marine Fisheries Service Laboratory at Galveston, Texas. The methods used and the basic principles of hatchery design were later described by Cook and Murphy (1966), Cook (1969) and Cook and Murphy (1969). By 1966, the three commercial species of penaeid shrimp indeginous to the Gulf of Mexico (brown shrimp, P. aztecus, white shrimp, P. setiferus and pink shrimp, P. duorarum) had been reared to the post larval stage. Cook and Murphy (1966) described techniques for rearing algae (Skeletonema costatum) and brine shrimp (Artermia sp.) which are the two most widely used feeds for larvae and postlarvae shrimp and Mock and Murphy (1970) described the modern day design of a hatchery to rear shrimp from egg to postlarvae.

Pond Production

G. Robert Lunz pioneered pond-culture research in shrimp in the United States. Based on studies in South Carolina with Penaeus aztecus, P. duorarum and P. setiferus spanning more than ten years (Lunz, 1951; and Lunz and Bearden, 1963), he recognized the potential for ponds located in coastal lowlands and identified many of the problems inherent in site selection, construction and management. His studies were conducted both in ponds built in tidal marshes where flooding was accomplished through flood gates at high tide and on higher land where water was supplied with an irrigation pump. He demonstrated that ponds of these types were suitable for growing shrimp to both bait and edible size and also found that other marine organisms such as fish and crabs prey on shrimp and must be removed to insure good survival.

Broom (1968) reported the findings of a series of pond culture studies conducted at Grand Terre Island, Louisiana, from 1962 through 1968. Using juvenile and postlarvae P. aztecus and P. setiferus collected from nearby bay waters and stocked in one-quarter acre ponds, he determined that stocking rates as high as 20,000 per acre could be achieved with satisfactory survival and growth. He recorded production of edible size shrimp as high as 472 pounds per acre for P. aztecus and 809 pounds per acre for P. setiferus, feeding Purina Catfish Chow. Conversion ratios for this food ranged as low as 2.3:1 (feed to shrimp) for P. aztecus and 3.0:1 for P. setiferus. He also noticed that at comparable densities, P. setiferus achieved higher growth rates than P. aztecus and that harvesting of both species could be greatly simplified if the pond bottom was sloped sufficiently to allow draining into

a catch basin.

Wheeler (1967, 1968), working in ponds at Galveston, Texas, found that the addition of inorganic fertilizer enhanced growth of P. aztecus from postlarvae to large juveniles (length about 4 inches) and concluded that fertilizer increased the production of phytoplankton and hence the biomass of organisms on which young shrimp feed. More (1970) observed, in pond experiments with hatchery reared postlarvae at Palacios, Texas, that P. aztecus grew faster and to a larger size than P. duorarum. Latapie et al.(1972) and Neal and Latapie (1972), continuing Broom's research at Grand Terre, Louisiana, were able to increase production of edible size P. setiferus to 831 pounds per acre and reduce food conversion ratios, using Purina Catfish Chow, as low as 1.1:1. They also noted that P. setiferus grew faster and larger and exhibited lower mortality than P. aztecus in separate ponds treated alike and in ponds where the two species were stocked together.

Parker et al. (1972), working in tidal marsh ponds near West Galveston Bay, Texas, found that removing fish from culture ponds improved the growth, survival, and condition of P. setiferus and concluded that fish not only prey on shrimp, as shown by Lunz and Bearden (1963), but also compete with them for natural and supplemental foods. Parker and Holcomb (1973 and personal communication), confirmed earlier findings that P. setiferus was a better producer than P. aztecus and were able to increase production of edible size P. vannamei to 1,837 pounds per acre over 134 days and P. setiferus to 1,171 pounds per acre over 142 days while obtaining only 547 pounds per acre for P. aztecus over 103 days.

They also demonstrated that protein levels in commercially prepared feeds could be lowered from 45 percent to 20 percent with no reduction in growth and were instrumental in the development of a commercial ration prepared specifically for shrimp. Holcomb and Parker (1973) compared harvest efficiency in ponds using seine and drain techniques and found that draining ponds yielded about 98 percent of the crop while three consecutive seine drags accounted for no more than 80 percent of the crop and required eight times more labor. Parker and Conte (1975) have shown that production levels as high as 4,696 pounds per acre of 40 count (heads-on) P. vannamei can be achieved in modified raceways in 146 days and Parker and Conte (personal communication) have produced up to 1,530 pounds per acre of 15 count (heads-on) P. stylirostris in 139 days in a static system.

Research to date on this and related projects has identified a number of factors important to production and marketing of penaeid shrimp. Critical among these are the desirable species for a particular location; the desirable time of year for rearing; suitable stocking rate for maximum production of an optimum size shrimp; suitable water exchange rates for optimum production; and engineering of production systems for most efficient and economical operation. The complexity of these factors and their combined interaction necessitate continued research if shrimp culture is to assume a productive position in today's agricultural picture. In addition, the improved production obtained with the non-indigenous species, P. vannamei and P. stylirostris, necessitates the development of new supply systems for seed stock if these species are to be utilized in Texas.

THE TEXAS A&M UNIVERSITY PROGRAM

Texas A&M University began its shrimp culture program in 1968 in Brazoria County. In cooperation with the Brazoria County Mosquito Control District, Texaco, Ralston Purina, and Dow Chemical Company, the program expanded into a 22-pond operation that established the foundation for shrimp culture production in Texas. It was determined early that white shrimp (Penaeus setiferus and P. vannamei) provided better yields than brown shrimp (P. aztecus) and as a result of proper species selection and better pond design, production rates increased from 200 to 1,800 pounds per acre. Pond design was improved to facilitate harvest through a drain flume that yields 99 percent of the crop. Cooperating with Ralston Purina, a shrimp ration was developed with a demonstrated food conversion ratio of less than 2:1 and a protein level of only 20 percent.

In 1972 a second mariculture facility was established in Corpus Christi, Texas, in cooperation with Central Power and Light Company and Ralston Purina. Utilizing technology developed at the Brazoria County facility a production module was designed and constructed at the Barney M. Davis Generating Station to demonstrate the feasibility of intensive shrimp culture. Consisting of three adjacent ponds of one-eighth, one-fourth, and one-half acre through which shrimp are rotated as they grow, the tri-pond concept provides better utilization of space than previously experienced with single pond units. The pilot module has demonstrated production capabilities of up to 6,000 pounds of 38-count (heads-on) shrimp (Penaeus vannamei) and up to 3,000 pounds of 15-count (heads-on) shrimp (P. stylirostris) during the six to seven month growing season in Texas.

In 1974 construction was begun on an additional complex of 18 quarter-acre ponds adjacent to the 1,100 acre cooling lake at the Barney M. Davis Generating Station. These ponds compliment the facilities in Brazoria County and allow testing of production techniques under the high salinity conditions characteristic of the Laguna Madre while the Brazoria County facilities evaluate production in the lower salinity waters of Galveston Bay. The new complex, in addition, can utilize either heated water from the cooling lake or water directly from the Laguna Madre at the power plant intake canal.

Facilities at both locations were designed to conduct replicated studies using variations in diet, species, stocking densities, water fertlization, and disease control, among other pond management techniques. Current efforts are directed toward improved pond management. Both facilities are used to compare production capabilities of native white shrimp (Penaeus setiferus) with two white shrimp species imported from the Pacific coast of Central America (P. vannamei and P. stylirostris) and evaluating stocking densities. To compliment the field program campus based research is directed to disease diagnosis and control, reproduction and maturation, product quality, production economics and marketing.

A project to determine the suitability of underground salt water for shrimp mariculture was initiated in 1973 with the County Agricultural Extension agent in Ward County. Two gravel pits containing salt water (15 ppt salt) at two locations about 25 miles apart were stocked with post-larval shrimp (Penaeus stylirostris) to determine whether shrimp could survive and grow. The water to fill these pits came from a strata

less than 25 feet deep. Results from one pit were unsatisfactory because of poor water quality; from two other pits, inconclusive because of inferior experimental design; and from one pit, encouraging because shrimp were recovered which grew at rates comparable to the coastal experiments. Since these shrimp were not fed, the investigators were doubly encouraged since their survival and growth indicated that these waters contained organisms suitable for shrimp food. This is a necessary ingredient in a shrimp farming operation because these organisms must constitute the diet of young shrimp when they are too small to feed on prepared rations.

The following year two quarter-acre ponds were constructed and filled with water from the pit which yielded encouraging results the previous year. These ponds were stocked, but again results were inconclusive. In the first instance only one pond was stocked (P. stylirostris) and it had not been properly aged or fertilized to enhance production of natural foods. It is likely that most of the young shrimp died from starvation. Thirteen shrimp out of 50,000 stocked were recovered and these grew at a rate comparable to shrimp stocked at Corpus Christi. In the second instance each pond was stocked with 5,000 P. setiferus and none survived. These shrimp, however, had been in shipping containers for 18 hours and were in very poor condition when they arrived at Monahans. In fact, more than 50 percent were dead when the containers were opened. It was concluded that these mortalities were the result of stress in shipping rather than factors in the ponds.

This year both ponds were stocked with 10,000 P. setiferus. Growth was better than previously experienced for this species in coastal experiments. After 100 days the two ponds were harvested and yielded an average

of 182 pounds per acre of shrimp measuring 38 count (heads on). These results definitely indicate commercial potential for West Texas and have prompted plans for follow-up studies next year.

The one major factor hindering the development of shrimp farming in Texas is an inadequate supply of brood shrimp to produce the quantity of seed-stock needed for a commercial operation. To remedy this situation, the Texas A&M University has entered into a cooperative program with the National Marine Fisheries Service at Galveston which is intended to develop a supply of brood shrimp from the Gulf of Mexico and to rear brood shrimp to maturity in captivity.

Personnel at the Brazoria County Facility working with the Extension area marine fisheries specialist and county marine agents and with local shrimp fishermen are using shrimp trawlers to collect female shrimp (Penaeus aztecus and P. setiferus) which have mated and are ready to spawn. These gravid females are transported to the National Marine Fisheries Service Hatchery at Galveston where they are spawned and the young are reared to seed-stock size.

The technology involved in this process was developed over the past ten years by the National Marine Fisheries Service and is being demonstrated on a pilot commercial basis using the traditional Extension concepts which have been proven successful many times in agriculture. This year more than one million seed shrimp were produced through the cooperative program. Extension personnel were trained to operate the hatchery and three Texas shrimp fishermen were trained in the complex art of sourcing gravid female shrimp.

Personnel at Corpus Christi are working closely with National Marine Fisheries Service researchers to determine the requirements needed to produce gravid female shrimp from stock reared entirely in captivity. Success to date has been encouraging. In October, 1974, 139-day-old white shrimp (P. stylirostris) six and one-half inches long were harvested from the half-acre pond in the production module at Corpus Christi. About 500 of these were shipped to the National Marine Fisheries Laboratory at Galveston and overwintered in a heated raceway. Since overwintering in a heated facility is necessary to maintain brood stock, researchers were encouraged when this was accomplished with no major problems, particularly since the survival rate was better than 80 percent.

The following April, 100 of these shrimp (50 males and 50 females) were returned to Corpus Christi and stocked in a quarter-acre pond where they could receive heated water from the cooling lake. By June these shrimp had grown from seven to eight and one-half inches and began to show signs of sexual maturity. In August and September a total of five gravid female shrimp were collected from the pond and spawned. In at least two instances fertilized eggs were definitely observed, but none hatched. The failure of the eggs to hatch probably was due to the fact that facilities were inadequate to filter the water properly. As additional gravid females are collected, they will be shipped to the hatchery in Galveston were suitable facilities are available.

Although these results represent a major breakthrough in shrimp reproduction, it may be a few years before technology will be adequate to apply on a commercial scale. Until that time, plans for commercialization much be based on obtaining brood stock from the Gulf of Mexico.

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CURRENT STATUS OF THE CULTURE OF RIVER SHRIMPS OF THE GENUS MACROBRACHIUM

ABSTRACT

Highly esteemed as a luxury food item in its native habitats in Asia, the freshwater shrimp, Macrobrachium rosenbergii, is now cultivated in many parts of the world. With only the rudiments of its biology or its culture presently understood, this species has shown considerable potential for further development as a commercial product. A primary factor contributing to the current success in rearing M. rosenbergii is our ability to manage all stages of its life cycle.

In order to achieve the ultimate potential of Macrobrachium culture, much basic and applied research into this animal's biological functions, physical and social environmental relations and management/culture techniques must be conducted. Texas Agricultural Experiment Station has made a committment to contribute to the development of freshwater shrimp culture through research into these priority areas.

CURRENT STATUS OF THE CULTURE OF RIVER SHRIMPS OF THE GENUS *MACROBRACHIUM*

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Introduction

In many parts of the world, river shrimps of the genus *Macrobrachium* support active fisheries, some of which are large enough to result in international exports. In the early 1960's, Dr. Shao-wen Ling, FAO Regional Fish Culturist in East Asia, sensed the economic potential of being able to culture the locally popular variety of freshwater shrimp rather than suffer the uncertainties of a fishery based on natural populations and therefore began his now well-known studies of *Macrobrachium rosenbergii*.

Dr. Ling found that although adult shrimp did live, mate, bear eggs and produce viable larvae in freshwater, they were not able to produce successive generations in that environment. Ling found that the larvae of this freshwater animal required brackish water for their complete development. Studies of the natural history of this shrimp led to an explanation of this apparent anomaly through an understanding of the pattern of the life cycle: following mating, which normally occurs in the freshwater portions of river systems, the female shrimp begins a migration downstream. During that time, she spawns and carries

the externally attached eggs with her. At about the time she reaches the brackish water of the river mouth, her eggs hatch, releasing larvae to drift out into the estuary. After a period of development in the estuary, the planktonic larvae metamorphose to juveniles which settle to the bottom and begin to seek the freshwater environment of their parents (Ling, 1969a). Subsequent studies have revealed similar life histories for other species of Macrobrachium; in Texas, these are M. carcinus, M. acanthurus, M. ohione and M. olfersii.

With Ling's description of the life history of Macrobrachium rosenbergii and his discovery of successful larval rearing techniques, the life cycle of this animal became subject to control. The term "control" should be used with reservation, for although all phases of the life cycle can be manipulated in culture situations, a lack of basic knowledge of biological functions in this or in virtually any other crustacean does not permit what might properly be called precise control of the animal's life cycle.

The application of Ling's findings to practical culture of M. rosenbergii was initiated at the laboratory of Takeo Fujimura by the Hawaii Division of Fish and Game. Fujimura's group contributed significantly to the early development of hatchery technology and pond management methods (Fujimura, 1966). These efforts resulted in the establishment, in the late 1960's, of the first operational program of commercial freshwater shrimp culture and possibly the world's only commercial program in complete husbandry of any crustacean.

The work of Ling and Fujimura has stimulated a world-wide effort in culturing freshwater shrimp - most of which is directed toward refining the culture of M. rosenbergii or in evaluating the culture potential of several other species of Macrobrachium.

In reviewing the current status of freshwater shrimp culture, I will take illustrations from published accounts and from my own observations during several years in Hawaii. The following comments will refer to M. rosenbergii which is the most promising species at the moment.

Current State of the Art

Producing and Hatching Eggs

The freshwater shrimp breeds and spawns year-round in its natural environment; the same may be said of this shrimp where it is farmed in the Caribbean, in Hawaii and in other tropical and subtropical parts of the world (Goodwin and Hanson, 1975). In temperate regions where reproductive activities are reduced or cease during seasonal low temperatures, egg production and hatching readily continue in environmentally-controlled laboratory or hatchery conditions. The eggs hatch in fresh or brackish water to release free-swimming larvae.

The relative ease with which gonad maturation, mating and egg production occur and the relatively short period (as few as eight months) between successive generations of shrimp have spurred interest in the area of selective breeding and genetic studies (S. Malecha, personal communication). Presently these studies are attempting to evaluate genetic and environmental factors affecting observed

variation in growth rate. The ability to manipulate variability in desired characteristics in a population of animals, distinguishes truly domesticated from wild stocks.

Successful development of larvae to the post-larval or juvenile stage requires brackish water (Fujimura, 1966; Ling, 1969b). The natural food of shrimp larvae is primarily smaller forms of the zooplankton community of which they are a part. The aquaculturist's most widely used substitute for wild zooplankton is the brine shrimp, Artemia salina. Live Artemia nauplii are easily cultured for use in the laboratory or commercial hatchery and have given the most consistently good results of any foods studied in rearing shrimp larvae. Declining production and greatly increased demand for Artemia have recently caused the search for an alternative larval food supply to be intensified. Finely sieved fish flesh is frequently fed to larvae as a supplement to or replacement for (in latter stages) the diet of Artemia (Goodwin and Hanson, 1975).

Rearing Juveniles

Most present grow-out systems for juvenile shrimp are characterized as being extensive in design - relatively low technology, large, earthen pond operations. Fujimura's practice in Hawaii is to stock the rearing ponds 2 or 3 times each year and to harvest weekly, throughout the year. Fujimura's ponds have yielded up to 4000 pounds (live)/acre. In that situation, the fastest growing shrimp reach harvest size of from 5 to 7 live animals per pound (or 10 to 14 headless per pound) in about 7 months from stocking time. Slower growing members of the initial stock continue to enter the harvestable category

for several months after those showing the most rapid growth. In locations where culture may continue year-round, such variation in growth causes no great problems, and, in fact, is utilized in achieving a harvest of the same-sized animals throughout the year. In the temperate zone, however, with its restricted growing season, only a single harvest will be possible in most instances and will, therefore, result in considerable variation in the sizes of animals harvested. Greater uniformity in size of harvested shrimp could be achieved by harvesting smaller animals, before variation in growth had resulted in such a range of sizes. Production of smaller shrimp would result in a more rapid turnover of stock in both hatchery and grow-out phases of the culture system than is presently found in production of "jumbo" size shrimp. Present Macrobrachium hatchery operations would require significantly increased production to support such a program.

Another factor related to size of harvested product is the cost of feed, which during the longest phase of culture, the grow-out of juveniles, has a significant bearing on ultimate profits (Goodwin and Hanson, 1975). To this critical aspect of shrimp culture, that is, feeding the aquatic livestock, today's practitioners can bring but scant knowledge. Any optimum feed must satisfy an animal's nutritional requirements while reducing or avoiding unnecessary components. The rudimentary state of the art of shrimp culture is reflected in our meager knowledge of nutritional requirements of crustaceans and by the variety of feeds currently in use - broiler starter mash, catfish and trout rations and various items formulated

for shrimp. The development of feeds and feeding techniques are areas of, as yet, insufficient research activity.

Other areas of research need or activity include behavioral biology or social requirements, disease control, engineering design, processing and marketing, economics and alternate culture systems to those based on extensive, earthen ponds.

The nature of any future industry based on the culture of Macrobrachium will evolve as decisions are made regarding several outstanding dichotomies: extensive or intensive culture systems, large, industrial scale, or small, family scale operations and systems of monoculture of polyculture. By functioning within a variety of environmental and economic situations, however, and by working toward a variety of goals, elements of a future industry will undoubtedly reflect a similar variety of forms. The present state of freshwater shrimp culture has resulted from a very pragmatic history in which trial and error played a significant role in trying to answer immediately apparent questions. This approach has brought about sufficient development to support the commercial operations of small scale, low technology, extensive monoculture of M. rosenbergii in certain locales. Many areas of the world would benefit today from the establishment of just such operations. Future development of this and other methods of shrimp culture, however, will rely increasingly on more sophisticated, controlled experimentation to provide a more basic level of understanding of the animal and its husbandry.

Texas Agricultural Experiment Station has made a committment

to contribute to the development of freshwater shrimp culture. Our contribution will take the form, initially, at least, of helping to close some of the information gaps in the area of basic biology of the several species of Macrobrachium found in Texas and M. rosenbergii. A more distant objective is to become involved in production schemes.

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CHEMICAL AND NUTRITIVE COMPOSITION OF SHRIMP

ABSTRACT

This paper is a resume of the proximate composition, amino acid profile, fatty acid profile, cholesterol, macro- and micro-element and vitamin content of various species of raw or processed shrimp. The data for the various components in the composition of shrimp were gleaned from literature. The compilation is based on 208 references.

CHEMICAL AND NUTRITIVE COMPOSITION OF SHRIMP

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Introduction

In 1972, 234 million pounds of shrimp, worth nearly a billion dollars, were harvested off the coast of the United States. Over half came from the Gulf of Mexico (Roedel, 1973). During the same year (1972), the United States imported 223 million pounds of shrimp from 69 nations. Mexico led all countries with more than 80 million pounds. India was second with more than 33 million pounds. This accounts for nearly one half of the total shrimp imports (Anonymous, 1973).

In 1972, the American people consumed 400 million pounds of shrimp (Anonymous, 1973). Nearly 90% was sold frozen and approximately three quarters of that went into the institutional trade (Whitaker, 1973). Frozen shrimp consumption is evenly distributed in the various regions of the United States. The United States per capita consumption of shrimp is a little over one and one half pounds in the past year (1975). It is clear that shrimp makes up

an important and vital segment of the total seafood consumption in the United States and plays a key role in our eating pleasures and nutrition.

The objective of this resume is to assemble available data on the chemical and nutritive composition of shrimp. In the future, such data would be useful for guiding further investigations on the composition of shrimp. This kind of information is becoming vitally important in this age of consumerism and nutritional awareness.

Methods and Procedures

The data summarized in this paper were obtained from the Chemical and Nutritional Composition Data Bank being established at Southeast Utilization Research Center (SURC). A description of the data bank and its computer ramifications are described by Sidwell et al. (1974) in an interim report published in Marine Fisheries Review.

Results and Discussion

Proximate composition of various market forms of shrimp is shown in Tables 1 and 2. The values in Table 1 are reported for shrimp or prawns whose species were not identified. In Table 2, the species of shrimp or prawns used in the analyses are characterized by species, but the values for each species are sparse; therefore, the data are grouped according to families. In looking over the bibliography from which the data were gleaned, clearly only

a small portion of the data originated in the United States. Most was obtained from data published by investigators in Australia, England, France, India, Japan, Philippines, Norway, and Venezuela.

Crude protein. Table 1 shows the protein content of raw and processed mixed species of saltwater shrimp. The largest amount of information is available for peeled, raw shrimp. The protein ranges from 14.1 to 25.0 gm with an average of 19.8 gm per 100 gm. This variability in composition may be due to the size of the animal, its physiological status (pre- or post-spawning), geographical area of catch, or the technique used in analyzing the sample. The cooked or canned (wet pack) shrimp contains about the same amount of protein and it is higher for the dry pack. It may be noted from this table that as the protein in the processed shrimp increases, there is a decrease in moisture. Protein of shrimp--dried and salted, and/or fermented--will vary extensively due to variations in the amount of salt added or the degree of dryness. The protein of the shrimp used in easy-to-prepare convenience foods is diluted by the ingredients used to process the product.

In Table 2 it may be noted that the average protein content for the different families of shrimp is very similar. There is more information for the Penaeidae, spp. than for the other groups. Establier (1963) did an extensive study on Parapenaeus longinostris L., caught off the coast of Spanish Morocco and in the Gulf of Cadiz. He observed sex and seasonal differences from 1960 to 1962. He found no protein differences due to sex; the protein content for males ranging from 17.1 to 20.9 gm and for females from 16.7 to 21.4 gm

per 100 gm at the Gulf of Cadiz; males, 17.1 to 20.2 and females, 17.0 to 21.1 gm per 100 gm off the coast of Spanish Morocco. Also, no differences seemed to be present between the two harvesting areas. There appears to be a seasonal variation. Thompson, (1971), over a period of one year, noted that the protein content of P. aztecus harvested from the Gulf of Mexico varied from 19.2 to 21.2 gm per 100 gm. In both studies the size of the animals was held relatively constant.

Crude fat. The fat content (Krzeczkowski, 1970) of whole shrimp is about two times the amount found in the tail muscle, as it may be observed in Table 1. Koga (1970a, 1970b) obtained samples of whole frozen shrimp from the market place at several different times during the year. He assumed that the differences in fat content were due to seasonal variations for the shrimp harvested from Japanese waters. The lowest lipid value he reported, 1.4 gm per 100 gm, was for imported Mexican shrimp.

Raw and peeled shrimp tails contained about half the fat content reported by Koga. The range is quite wide--.1 to 5.8 gm percent. Polansky and Toepfer (1969) found .1 percent in commercially obtained samples. Achard, et al. (1934) reported a high value of 5.8 gm percent in unspecified shrimp from the French market. The values listed in Table 2 for the specific families of shrimp fall within this range. The only indication that the fat content may vary with season is the work done by Thompson (1971) on brown shrimp. For the year under study she found the content to range from .8 to 2.2 gm with an average of 1.4 gm per 100 gm.

In his three year study, Establier (1963) noted a narrower range (.37 to .88 gm per 100 gm) with possible indication that the fat content was higher in the fall.

Phospholipids and sterols make up the greater portion of the crude fat present in shrimp muscle. Cholesterol represents nearly all (95%) of the sterols (Kritchevsky, et al. 1967).

Ash. It is quite difficult to explain the variability in the ash content of whole or raw peeled and of ready-to-prepare shrimp unless processed animals were not wholly deveined. A high ash content can be expected in processed shrimp whether it is canned, salted and/or dried, due to salt added in the manufacturing process.

Amino acids. In Table 3 are the amino acid profiles for the protein in whole egg and Penaeid shrimp muscle. In general, the amino acid content of the protein in shrimp muscle does not differ markedly from that of whole egg protein. Threonine, serine, alanine, valine, isoleucine, and cystine content in shrimp is lower than in whole egg. Even so, the amount in 100 grams of shrimp muscle is sufficient to supply at least one half of man's daily requirement.

The amino acid profile for the mixed species and Penaeidae, spp. is listed in Tables 4 and 5, respectively. With few exceptions, the differences between the raw and the processed shrimp seem to be relatively small; therefore, processing does not appear to affect the amino acids. Savagaon et al. (1972) reported exceptionally high values for aspartic acid, serine, glutamic acid, glycine and alanine in cooked shrimp. Sawat and Magar (1961) reported 19.8 gm and Master and Magar (1954) 18.5 gm of lysine per 100 gm of protein.

The analyses for the whole shrimp caught in Alaska agree quite well with the values obtained by Ackman and Eaton (1969) from the shrimp caught off the coast of Nova Scotia.

The paper published by Bonnet et al. (1974) is probably the most complete resume of lipid analyses on Penaeid shrimp. Table 7 lists the amount of each fatty acid found in the crude fat from shrimp muscle. The amount of crude fat is the same, 1.2 gm per 100 gm. The biggest difference in the fatty acid profile for the white shrimp, P. setiferus, caught in the South Atlantic and in the Gulf of Mexico is the amount of polyunsaturated fatty acids. It is 10% higher in shrimp harvested in the South Atlantic. The brown shrimp, P. aztecus, was more like the white shrimp from the Gulf.

From the limited fatty acid data the following observations may be noted: (1) one fourth of the fatty acids is saturated, of which palmitic acid (16:0) and stearic acid (18:0) make up the greater portion; (2) oleic acid (18:1 9) and hexadecenoic acid (16:1) predominate in the monounsaturated group, and (3) among the polyunsaturates the 20:4, 20:5 and 22:6 respectively prevail.

Cholesterol content of shrimp. Very few values on the cholesterol content of shrimp are reported in literature. The values range from 116 mg (Achard et al., 1934) to 226 mg per 100 gm (Pihl, 1952). Some of the discrepancy may be due to methodology, for the complete saponification of the lipoid material in tissue is extremely important.

Kritchevsky et al. (1967) gives this as a reason for the incorrectness of the 1961 report of 138 mg per 100 gm of shrimp muscle. The value of 200 mg per 100 gm published by the same investigators in 1967 is more consistent with results of other investigators. The degree of variation due to seasonal or species variation is not clearly defined. Koya (1970a) indicated that the difference in the cholesterol content of shrimp harvested in Japan may be due to seasonal variation. His values are 82 mg in October and 245 mg per 100 gm in August. These analyses were performed on the whole animal, and therefore these values were not included in the aforementioned range.

Thompson (1964) reported the cholesterol levels of white and brown shrimp harvested from the Gulf in the winter of 1962. There was no difference between the two species, which contained 157 and 156 mg per 100 gm of muscle, respectively. Okey (1945) and Feeley (1972) reported 150 mg per 100 gm in their compilation of cholesterol levels found in different foods.

How reliable are these data? There are very few values, yet they are the ones used by the medical community to calculate special diets. It is wrong to consider sterol composition alone, without taking into account the effect of other components. Conners (1969), in his review of the research on the effect of dietary lipid and sterols in the sterol balance, commented that the presence of polyunsaturated fatty acids in the diet of normal men reduced the cholesterol level in the blood by increasing the excretion of bile acid salts.

In hyper-cholesterolemic persons, this observation was not noted. The difference may be due to metabolism abnormality. Shrimp has both cholesterol and polyunsaturated fatty acids. The question is how much of the cholesterol is actually absorbed from the gut of the human body.

Macroelements. In Tables 8 and 9 are listed the values for the nutritionally important elements sodium, potassium, calcium, phosphorus, iron and magnesium. Sodium and potassium values are quite diverse. For raw, peeled, mixed species there are four values reported in literature (Table 8) and they range between 84 and 140 mg per 100 gm. For the Penaeidae, spp. the amount of sodium is rather high, 100 to 220 mg per 100 gm, with an average of 178 mg percent. This information is based on the results of 6 investigations (Table 9). More conclusive data are needed for sodium since it is one of the food constituents most often considered by the personnel in the medical community in calculating a special diet.

The values for calcium, phosphorus, iron, and magnesium need to be evaluated more closely. For calcium, there are sixteen values ranging from 50 to 275 mg per 100 gm. Clements and Hutchison (1939) reported the 50 mg and Higashi (1951) 275 mg per 100 gm. The values for the Penaeidae, spp. are still more diverse, ranging from 10 to 305 mg per 100 gm. Gastanaduy (1965) reported the 10 mg in shrimp and Valdehita (1959) the 305 mg per 100 gm. Some of this diversity can be explained by differences in the chemical methods used to measure calcium or by differences in how well the shrimp tails were cleaned.

A great deal of work has been done on the iron content of shrimp muscle. Again, the range of values is wide--1.4 to 135 mg per 100 gm for the Penaeidae, spp. The average and range are far more reasonable for the mixed species; .7 to 3.3 mg percent. If the iron in shrimp is available to the human organism, then shrimp is a good source of nutrient iron. However, the availability of the iron in shrimp is not known.

Magnesium is another important dietary nutrient for it activates the enzyme system which functions in the metabolism of carbohydrates to produce energy. Shrimp is a good source of this element. The range of amounts of magnesium found in shrimp is again quite extensive--40 to 500 mg per 100 gm. It is not known whether the animal accumulates this element in its muscle, or reaches a saturation point, at which point the magnesium content levels off.

Cooked shrimp muscle absorbs some of the salt from brine used in the processing line, or from salted cooking water, thus increasing sodium content. The other elements are concentrated through the loss of moisture due to cooking, but they are not out of line with the raw counterpart. Again, the basis for the elemental content of processed shrimp muscle is quite limited.

Microelements. In Tables 10 and 11 is the resume of the data on trace elements gleaned from literature. The data are by no means conclusive.

Arsenic is of interest because it has a tendency to accumulate in the muscle of shrimp. The amount of arsenic found in shrimp is probably correlated to the amount present in its environment.

The reported values for total arsenic in raw, cooked or canned shrimp range from .08 to 54.0 ppm. Dick and Pugsley (1950) found the very low level .08 ppm in canned shrimp when they surveyed the canned shellfish and crustacea on the grocers' shelves in three populous areas of Canada. Chapman (1926) found, on the average, 54 ppm in shrimp harvested from the Thames estuary. Presently, we do not know in what form arsenic is present is shrimp. Metallic arsenic is more toxic than the arsenic found in shrimp muscle.

Cadmium is another element that is being closely monitored as possibly toxic to the consumer. Peden et al. (1973) reported a cadmium level higher than .1 ppm in shrimp. They obtained samples, which contained 3.9 ppm in the raw shrimp and 2.9 ppm in the cooked, from the polluted waters of the Bristol Channel.

Shrimp contains very little mercury. The FDA has set guidelines for the amount of mercury that can be present in seafoods at .5 ppm. Shrimp falls well within the guidelines. The presence of selenium, .6 to 1.9 ppm, has been shown to interfere with the absorption of methyl mercury, although little is known about this relationship.

Zinc is present in the cells of all living organisms and is especially high in marine organisms. It functions as a component of many enzymatic systems, and therefore has a predominant role in the carbohydrate and protein metabolism. Shrimp contains varying amounts of zinc ranging from 7.3 to 72.0 ppm. The zinc present in the environment has a great influence on the amount of zinc in the shrimp muscle.

Copper is another important element needed for good nutrition. Along with iron, it is necessary for the body's synthesis of hemoglobin. Shrimp contains copper ranging from .1 to 131.0 ppm.

Iodine is necessary for the proper function of the thyroid gland. The values reported in literature range from 0.5 to 1.5 ppm. Again, the amount present in the organism is associated with the amount present in the shrimp's habitat.

Shrimp contains small amounts of other elements, such as vanadium and chromium. Small amounts of vanadium are found in the bones, teeth, and fat of man. There is some evidence that vanadium interferes with the synthesis of certain lipoid materials, that is, cholesterol. Chromium is widely distributed in the tissues of man and functions in glucose tolerance and fat metabolism. There are indications that it reduces the serum cholesterol level in man.

These are but a few benefits that may be attributed to the presence of trace elements in the diet. Shrimp can supply these nutritionally important elements.

The other elements listed in Tables 10 and 11 are represented by too few values for a meaningful conclusion to be drawn.

Vitamins. As may be noted in Tables 12 and 13, very little information is available on the fat-soluble vitamins. Miller and Branthoover (1957) found 60 IU of Vitamin A per 100 gm of raw or canned shrimp. De Ibarra (1964) listed a value of 30 IU per 100 gm in his food composition tables based on a review of literature. Bro-Rasmussen (1958) found no Vitamin A in shrimp bought on the Danish market.

Aschehoug et al. (1939), by rat assay, learned that shrimp muscle contained 150 IU of Vitamin D per 100 gms. There are no values for vitamins E and K.

Eusebio (1966) and Lontoc (1966) analyzed Penaeids for Vitamin A and found 297 IU and 201 IU per 100 gm, respectively. In a review, Pyke and Wright (1941) reported 17 IU. No conclusion can be drawn from these few and diverse values for Penaeidae, spp.

The mean and range of the data clearly reveal that shrimp is a poor to medium source of thiamine and riboflavin. For example, regardless of species, the thiamine content in shrimp muscle ranges from 1 to 100 mcg and the riboflavin contents from 13 to 650 mcg per 100 gm. The 650 mcg per 100 mg was reported by Shaikhmahmud and Magar (1957) for the prawn, Penaeus penicillatus. These authors state that this species also contained more fats, protein, and minerals than the other species, and the possible seasonal variation was not evident. There are indications that thiamine and riboflavin are readily destroyed during processing and storage (Ives, 1944; Thompson, 1944). Novak (1956) found that these vitamins were reduced during frozen storage.

Shrimp is a good source of niacin and pantothenic acid. Again, there is a wide variation in the results obtained by the various investigators. Terri (1957) felt that some of the variation may be due to seasonal variation. It is possible that the age of the animal and the food available in its environment may play an important role in the vitamin content of shrimp muscle.

Shaikhmahmud and Magar (1957, 1961) have reported the presence of ascorbic acid in shrimp muscle. In Penaeidae, spp. they found 0 to 5.0 mg per 100 gm of raw shrimp muscle. Oliveiro (1955) and Leung et al. (1961), in their compilation of food composition tables, claimed that there was no ascorbic acid in Penaeidae, spp.

Vitamin B_{12} is a necessary component in the maintenance of a normal blood pattern. Shrimp can contribute significantly to the daily requirement. Regardless of species, the Vitamin B_{12} content for raw shrimp ranges from 1.0 to 2.5 mcg per 100 gm. There is some evidence that this vitamin is partially destroyed in processing (Tables 12 and 13).

Conclusion

This resume on the composition of shrimp illustrates the lack of conclusive data on its various components. In fact, many conceptions on the chemical and nutritional value of shrimp are formulated on limited evidence. Due to the need for this information for nutritional labeling, it is necessary to fill the gaps and obtain more reliable values for the different compositional data.

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Table 1--Proximate composition of various market forms of mixed species of saltwater shrimp,[1] raw or processed.

	Protein %	Fat %	Ash %	Moisture[2] %
Shrimp				
whole, raw	16.7[3] 10.3 - 23.8[4] 6[5]	2.6[3] 1.4 - 3.5[4] 4[5]	2.1[3] 1.4 - 3.9[4] 5[5]	77.2[3] 69.5 - 82.9[4] 9[5]
peeled, raw	19.8 14.1 - 25.0 28	1.4 0.1 - 5.8 28	2.6 1.0 - 6.8 17	75.1 59.4 - 84.8 31
peeled, cooked	22.6 21.2 - 25.0 4	1.5 0.8 - 2.4 4	3.2 1.0 - 5.4 2	68.6 62.5 - 70.0 4
canned, wet pack	21.6 15.0 - 26.8 15	1.1 0.7 - 1.5 15	4.5 2.6 - 6.5 9	73.4 69.0 - 81.3 12
canned, dry pack	24.5 20.0 - 26.8 3	1.5 1.3 - 1.8 3	5.0 1	66.4 1
dried	70.2 55.4 - 92.3 10		11.8 2.5 - 19.7 4	14.4 1.5 - 24.0 7
dried and salted	50.0 32.2 - 55.4 4	2.0 1.2 - 2.5 4	15.7 13.0 - 20.7 3	28.3 20.4 - 37.7 4

Table 1--Proximate composition of various market forms of mixed species of saltwater shrimp,1/ raw or processed. Cont.

	Protein %	Fat %	Ash %	Moisture %
Shrimp, cont.				
salted and fermented	12.3	2.8	20.5	62.7
	12.2 - 12.5	1.1 - 4.5	14.7 - 26.1	58.4 - 59.9
	3	3	3	3
breaded, not fried	12.0	1.6		65.2
	11.0 - 13.1	1.5 - 1.8		62.7 - 67.6
	2	2		2
breaded, fried frozen	11.8	13.7	2.8	43.6
	1	1	1	1
paste	24.9	7.5	3.5	50.0
	18.4 - 30.0	2.5 - 11.5	3.0 - 4.0	28.0 - 66.2
	4	3	2	4
meal	45.2	12.9	25.8	6.6
	42.3 - 48.6		21.6 - 30.0	5.0 - 8.1
	4	1	2	2
protein concentrate	91.0	0.3	4.5	7.7
	1	1	1	1

Table 1--Proximate composition of various market forms of mixed species of saltwater shrimp,[1/] raw or processed. Cont.

1/ Identified in literature as "shrimp" or "prawns."
2/ Data for this table taken from following references: 2, 4, 5, 6, 7, 14, 15, 19, 26, 29, 32, 35, 39, 42, 49, 50, 54, 57, 62, 67, 77, 78, 80, 81, 90, 91, 93, 97, 98, 99, 102, 103, 104, 109, 110, 119, 119, 121, 125, 131, 135, 136, 138, 142, 143, 145, 149, 152, 155, 157, 170, 174, 187, 190, 200, 203.
3/ Average of the data found in literature.
4/ Range of the data.
5/ Number of figures involved in calculating the statistics.

Table 2--Proximate composition of various species of shrimp,[1] raw or processed.

	Protein %	Fat %	Ash %	Moisture %
Penaeidae spp.				
raw	18.5[2] 10.0 - 25.1[3] 44[4]	1.2[2] 0.2 - 3.6[3] 43[4]	2.0[2] 0.7 - 4.5[3] 43[4]	76.3[2] 67.5 - 82.8[3] 48[4]
canned	21.4 15.0 - 28.2 4	1.2 0.7 - 2.0 4	5.6 5.0 - 6.5 4	73.3 64.6 - 77.3 4
dried	76.2 49.1 - 86.9 12	3.2 2.1 - 3.5 12	7.7 4.8 - 14.1 11	9.3 4.2 - 27.8 12
salted	11.3 10.4 - 13.0 3	1.3 1.2 - 1.7 3	17.7 15.2 - 20.5 3	67.9 67.2 - 69.1 3
salted, fermented	14.9 1	1.0 1	20.6 1	63.3 1
salted, dried	52.3 1	1.9 1	14.6 1	29.6 1
paste	29.8 1	1.2 1	28.0 1	34.5 1
offal	18.4 15.7 - 21.1 2	2.4 1.1 - 3.6 3	3.6 2.1 - 5.3 2	74.3 72.6 - 76.8 3

Table 2--Proximate composition of various species of shrimp,[1/] raw or processed. Cont.

	Protein %	Fat %	Ash %	Moisture %
Pandalidae spp.				
raw	18.1	1.3	1.3	81.3
	17.1 - 19.1			81.2 - 81.6
	2	1	1	2
canned	19.9	1.8	3.4	74.9
	1	1	1	1
Crangonidae spp.				
raw	22.6	0.9		68.8
	1	1		1
Mixed species				
raw	18.1	1.6	4.8	
	10.0 - 22.8	0.6 - 3.2	3.1 - 6.4	
	5	4	2	
dried				
paste	19.5	1.8	34.6	40.7
	1	1	1	1
protein concentrate	78.9	7.0	15.7	
	1	1	1	

Table 2--Proximate composition of various species of shrimp,[1/] raw or processed. Cont.

1/ Data for this table taken from the following references: 3, 10, 12, 20, 30, 31, 32, 38, 40, 42, 45, 55, 56, 58, 71, 74, 92, 105, 106, 113, 132, 133, 136, 138, 147, 148, 155, 156, 171, 172, 173, 174, 183, 188, 191, 193, 196, 198.
2/ Average of the data found in literature.
3/ Range of the data.
4/ Number of figures involved in calculating the statistics.

Table 3--Amino acid profile of the protein of the edible portion of Penaeidae, spp. and of whole egg.

Amino acids (% of protein)	Whole egg 1/	Penaeidae spp. 1/
lysine	6.4	7.8
histidine	2.4	2.3
arginine	6.6	8.8
aspartic acid	7.0	10.4
threonine	5.0	3.9
serine	8.4	3.9
glutamic acid	12.4	15.0
proline	4.2	3.8
glycine	3.5	5.9
alanine	6.5	5.5
valine	7.4	4.8
methionine	3.1	2.8
isoleucine	6.6	4.2
leucine	8.8	8.0
tyrosine	4.3	3.6
phenylalanine	5.8	4.6
tryptophane	1.6	1.2
cystine	2.3	1.2

1/ Data for this table taken from the following references: 51, 55, 70, 78, 87, 92, 106, 108, 116, 120, 186, 196.

Table 4--Amino acid composition of the protein in mixed species of salt water shrimp,[1] raw or processed.[2]

	Raw	Cooked	Canned	Dried	Meal
Lysine	11.6[3] 6.8 - 19.8[4] 8[5]	11.7[3] 8.3 - 13.7[4] 5[5]	12.7[3] 8.1 - 18.7[4] 9[5]	10.0[3] 9.2 - 10.4[4] 4[5]	5.1[3] 4.8 - 5.6[4] 3[5]
Histidine	2.0 1.6 - 2.6 8	2.4 1.8 - 3.0 5	1.9 1.3 - 2.4 9	2.2 2.0 - 2.3 4	1.8 1.7 - 2.0 3
Arginine	8.4 7.2 - 9.3 5	9.6 6.6 - 11.4 4	9.1 8.5 - 9.5 6	9.1 1	5.3 5.3 - 5.3 2
Aspartic acid	9.3 1	17.4 16.3 - 18.4 3	9.3 1	10.0 1	8.5 1
Threonine	4.3 3.8 - 4.7 6	5.9 4.0 - 7.7 5	4.0 2.8 - 4.8 9	4.2 3.8 - 4.4 4	3.1 2.4 - 3.5 3
Serine		7.0 4.0 - 9.3 4		4.8 1	3.9 1
Glutamic acid	15.5 1	26.9 25.5 - 29.7 3	15.5 1	14.8 1	11.6 1
Proline	3.6 1	5.2 5.0 - 5.5 3		3.2 1	4.8 1

Table 4--Amino acid composition of the protein in mixed species of salt water shrimp,[1/] raw or processed.[2/] Cont.

	Raw	Cooked	Canned	Dried	Meal
Glycine	6.8 1	17.3 16.1 - 19.2 3	6.3 1	7.3 1	8.2 1
Alanine	4.6 1	13.2 12.3 - 14.1 3		5.8 1	5.4 1
Valine	4.6 4.1 - 5.0 6	5.8 5.4 - 6.2 4	5.0 4.2 - 5.7 9	5.0 4.8 - 5.1 4	4.5 4.1 - 4.7 3
Methionine	3.6 2.9 - 4.6 7	3.6 3.1 - 4.4 5	3.6 2.6 - 4.4 9	3.0 2.9 - 3.1 4	2.3 1.7 - 3.5 3
Isoleucine	5.4 5.0 - 5.6 6	5.6 5.1 - 5.8 4	5.4 4.6 - 6.0 9	5.5 5.2 - 5.6 4	3.7 3.6 - 3.7 3
Leucine	11.5 7.8 - 14.3 7	11.6 8.9 - 12.9 4	10.1 8.0 - 11.6 9	8.1 7.6 - 8.7 4	5.5 5.4 - 5.6 3
Tyrosine	2.5 1.0 - 3.3 5	3.9 3.6 - 3.9 5	3.6 3.6 - 3.6 3	3.5 3.5 - 3.6 4	5.3 1
Phenylalanine	4.7 3.8 - 6.2 8	4.8 4.6 - 4.9 5	4.5 3.8 - 5.0 9	4.3 4.2 - 4.6 4	4.3 3.7 - 5.5 3

Table 4-- Amino acid composition of the protein in mixed species of salt water shrimp,[1/] raw or processed.[2/] Cont.

	Raw	Cooked	Canned	Dried	Meal
Tryptophan	1.1	1.4	1.4	1.3	1.0
	0.4 - 1.8	1.2 - 1.6	1.0 - 1.6	1.0 - 1.6	1.0 - 1.0
	7	2	5	4	2
Cystine	1.5	1.3	1.6	1.3	
	1.1 - 1.5	1.1 - 1.5	1.6 - 1.6	1.2 - 1.5	
	6	2	3	4	

1/ Identified in literature as "shrimp" or "prawn."
2/ Data for this table taken from following references: 11, 18, 23, 35, 39, 57, 78, 115, 118, 131, 157, 158, 179, 190.
3/ Average of data found in literature.
4/ Range of the data.
5/ Number of figures involved in calculating the statistics.

Table 5—Amino acid composition of the protein in Penaeidae, spp. shrimp,[1] raw or processed.

	Percent of protein			
	Raw	Whole	Canned	Offal
Lysine	7.8[2] 6.6 - 7.4[3] 11[4]	6.4[2] 5.4 - 7.4[3] 2[4]	8.5[2]	5.8[2]
Histidine	2.3 1.5 - 3.8 11	1.7 1.6 - 1.7 2	2.2	2.7
Arginine	8.8 5.9 - 12.9 11	6.4 5.4 - 7.5 2	9.4	6.7
Aspartic acid	10.4 7.0 - 13.7 7	9.7 1		9.1
Threonine	3.9 3.5 - 4.8 7	4.2 1	4.1	4.1
Serine	3.9 3.3 - 4.8 6	4.2 1		4.0
Glutamic acid	15.0 12.9 - 17.5 7	13.4 1		11.2
Proline	3.8 3.5 - 4.5 7	5.7 1		5.3

Table 5--Amino acid composition of the protein in Penaeidae, spp. shrimp,[1/] raw or processed. Cont.

	Percent of protein			
	Raw	Whole	Canned	Offal
Glycine	5.9	6.4		7.2
	3.6 - 8.3			
	6	1		
Alanine	5.5	6.5		7.4
	4.9 - 6.1			
	7	1		
Valine	4.8	5.2	5.1	5.7
	3.4 - 6.2			
	7	1		
Methionine	2.8	2.3	3.4	2.1
	2.2 - 3.3			
	10	1		
Isoleucine	4.2	4.1	5.3	4.5
	3.8 - 5.1			
	5	1		
Leucine	8.0	6.4	8.5	7.3
	6.9 - 9.2			
	6	1		
Tyrosine	3.6	3.6		3.3
	2.3 - 4.9			
	10	1		

Table 5--Amino acid composition of the protein in Penaeidae, spp. shrimp,[1/] raw or processed. Cont.

	Percent of protein			
	Raw	Whole	Canned	Offal
Phenylalanine	4.6	4.3	4.5	4.1
	3.3 - 6.6			
	8	1		
Tryptophan	1.2	0.9	1.0	1.7
	1.0 - 1.6	0.9 - 1.0		
	11	2		
Cystine	1.2	1.0		
	1.1 - 1.8	0.7 - 1.3		
	11	2		

1/ Data for this table taken from the following references: 40, 51, 55, 66, 70, 78, 87, 92, 106 108, 116, 120, 146, 196.
2/ Average of data found in literature.
3/ Range of the data.
4/ Number of figures involved to calculate the statistics.

Table 6--Fatty acid composition of lipid in whole and muscle of pink shrimp, Pandalus borealis.[1/]

	Whole		Muscle	
	% of total fat			
	Nova Scotia	Alaska	Alaska	Maine
Saturated				
10:0	-	.6	.7	-
12:0	.3	.3	.5	-
14:0	2.9	4.1	2.6	1.9
15:0	.5	.5	.6	.5
16:0	14.9	14.3	15.7	10.1
17:0	.3	TR	TR	.5
18:0	2.6	3.0	2.3	1.1
19:0	TR	TR	TR	.3
20:0	TR	TR	TR	.6
24:0	-	TR	TR	-
Total	21.5	22.8	22.4	15.0
Monosaturated				
14:1ω6	.5	TR	TR	.3
15:1ω6	.4	.7	1.0	.2
16:1ω7	14.1	9.6	6.0	6.6
17:1ω8	.4	1.4	1.2	.3
18:1ω9	20.2	20.3	18.6	9.9
19:1	.2	.3	-	-
20:1ω9	4.8	3.6	2.7	.7
22:1ω9	4.9	3.4	1.8	.4
24:1	.3	-	-	-
Total	45.8	39.0	31.3	18.4
Polyunsaturated				
16:2ω4	.5	-	-	-
16:3ω4	.4	-	-	-
16:4ω1	.4	-	-	-
18:2ω6	.8	1.6	1.4	1.4
18:3ω3	.6	.6	1.2	-
18:3ω6	.4	-	-	-
18:4ω3	.9	1.6	1.2	.7
20:2ω6	.2	.7	1.5	.9
20:3ω6	TR	TR	TR	.4
20:4ω6	1.3	1.0	.4	2.3
20:4ω3	.7	TR	1.0	.9

Table 6--Fatty acid composition of lipid in whole and muscle of pink shrimp, Pandalus borealis.[1/] Cont.

	Whole		Muscle	
	% of total fat			
	Nova Scotia	Alaska	Alaska	Maine
Polyunsaturated				
20:5ω3	15.3	17.9	21.1	30.8
21:5ω2	.5	-	-	-
22:2ω6	.1	-	-	-
22:3ω6	-	1.7	2.0	-
22:4ω6	.4	.7	1.5	1.4
22:5ω3	.9	1.0	1.2	2.1
22:5ω6	.3	-	-	1.0
22:6ω3	8.9	11.2	14.9	20.8
Total	32.6	38.0	47.4	62.7
Crude Fat (%)	2.4	2.9	1.4	1.3

1/ Data taken from the following references; 3, 25, 96.

Table 7--Fatty acid composition of lipid in edible portion of raw white, P. setiferus, and brown, P. aztecus shrimp.[1/]

	White (Gulf)	White (So. Atlantic)	Brown
	% of total fat		
Saturated			
14:0	1.1	1.4	1.8
15:0	.7	.7	1.2
16:0	11.9	10.4	10.4
17:0	1.9	1.3	2.4
18:0	9.4	5.0	6.4
19:0	.5	.5	.7
20:0	.2	.2	1.0
24:0	.5	.2	-
Total	26.2	19.7	23.9
Monosaturated			
14:1	.1	.2	.3
15:1	.2	.2	.5
16:1	5.1	6.0	5.1
17:1	.2	.4	-
18:1 ω9	9.7	8.5	7.4
20:1	1.5	1.1	.8
22:1	.2	-	-
Total	17.0	16.4	14.1
Polyunsaturated			
18:2 ω3	1.4	1.4	1.6
18:2 ω6	.7	.6	.4
20:2 ω9	.8	.6	-
20:2 ω6	.3	.5	.9
18:3 ω3	1.0	.9	1.5
20:3 ω9	.3	.3	.3
20:3 ω6	.2	.1	.3
20:4 ω6) 20:3 ω3)	6.7	6.4	7.5
18:4 ω3	.4	.7	.3
20:4 ω3	.7	.8	.4
22:4 ω6	1.5	1.7	3.0

Table 7--Fatty acid composition of lipid in edible portion of raw white, P. setiferus, and brown, P. aztecus shrimp.1/ Cont.

	White (Gulf)	White (So. Atlantic)	Brown
	% of total fat		
20:5 u 3	16.5	25.8	18.4
22:5 u 6	1.5	1.6	1.8
22:5 u 3	2.7	2.6	4.2
22:6 u 3	17.8	18.2	17.3
Total	52.5	62.2	57.9
Crude fat (%)	1.2	1.2	1.2

1/ Data taken from the following reference: 25.

Table 8--Macroelements in mixed species of salt water shrimp,[1] raw or processed.[2]

	Raw	Cooked	Canned	Dried
Sodium, mg.	119[3] 84 - 140[4] 4[5]	2715[3] 1590 - 3840[4] 2[5]	2300[3] 1[5]	
Potassium, mg.	257 140 - 410 3	332 260 - 404 2	163 1	
Calcium, mg.	119 50 - 275 16	232 145 - 320 2	92 18 - 115[4] 7	814[3] 231 - 2306[4] 7[5]
Phosphorus, mg.	221 130 - 400 15	310 270 - 349 2	185 141 - 263 6	662 604 - 779 5
Iron, mg.	2 1 - 3 14	1 1 - 2 2	2 1 - 4 6	9 5 - 21 7
Magnesium, mg.	75 40 - 111 3	74 42 - 105 2	23 1	

1/ Identified in literature as "shrimp" or "prawn."
2/ Data for this table taken from the following references: 5, 19, 21, 34, 42, 44, 54, 69, 83, 93, 99, 102, 103, 104, 117, 121, 124, 136, 138, 142, 149, 152, 174, 184, 185, 200.
3/ Average of the data found in literature.
4/ Range of the data.
5/ Number of figures involved in calculating the statistics.

Table 9--Macroelements in Penaeidae, spp. shrimp, raw or processed[1/].

	Raw	Cooked	Canned	Dried
Sodium, mg.	178[2/] 100 - 220[3/] 6[4/]		197[2/] 1[4/]	3907[2/] 1[4/]
Potassium, mg.	334 220 - 386 7		252 147 - 357[3/] 2	915 1
Calcium, mg.	114 10 - 305 34		76 50 - 126 6	758 236 - 1800[3/] 4
Phosphorus, mg.	207 67 - 397 35		218 149 - 286 6	901 597 - 1100 4
Iron, mg.	48 1 - 135.0 35	40[2/] 1[4/]	23 15 - 35 7	
Magnesium, mg.	250 42 - 500 4			

1/ Data for this table taken from the following references: 10, 33, 38, 40, 42, 51, 56, 65, 84, 106, 132, 133, 136, 138, 171, 172, 173, 174, 194, 196, 198, 202.
2/ Average of the data found in literature.
3/ Range of the data.
4/ Number of figures involved in calculating the statistics.

Table 10--Trace elements in mixed species of salt water shrimp,[1] raw or processed,[2] in ppm.

	Raw	Cooked	Canned
Aluminum			1.00[3] 1.00 - 1.00[4] 2[5]
Arsenic	23.80[3] 1.8 - 54.00[4] 6[5]	3.10[3] 1[5]	6.70 .20 - 20.00 4
Barium			.20 1
Boron			.20 1
Cadmium	.90 .03 - 3.90 5	1.50 .10 - 2.90[4] 2	.10 .02 - .13 2
Chromium	.05 .01 - .12 4		.30 1
Cobalt	.12 1		
Copper	4.70 .10 - 13.00 9	5.50 3.00 - 8.00 2	5.00 1.70 - 13.20 5
Fluorine	.90 1		4.40 1
Iodine	.90 .23 - 1.50 4	.20 .09 - .38 4	.30 .09 - .38 2

Table 10--Trace elements in mixed species of salt water shrimp,[1/] raw or processed,[2/] in ppm. Cont.

	Raw	Cooked	Canned
Lead	.40[3/] .40 - .40[4/] 2[5/]		.40[3/] .30 - .50[4/] 3[5/]
Manganese	1.10 .02 - 2.10 4		.20 1
Mercury	.06 .04 - .09 3		.01 1
Molybdenum	.03 1		
Nickel	.03 1		
Tin			1.80 1
Selenium	.90 .60 - 1.90 2		.30 1
Zinc	18.70 7.30 - 42.00 6	72.30[3/] 1[5/]	19.00 1

1/ Identified in literature as "shrimp" or "prawn."
2/ Data for this table taken from the following references: 5, 14, 19, 24, 36, 37, 41, 42, 48, 49, 54, 68, 69, 73, 82, 83, 85, 93, 99, 102, 103, 104, 110, 117, 119, 121, 122, 124, 125, 126, 128, 136, 138, 139, 140, 141, 142, 143, 149, 152, 154, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, 174, 176, 184, 185, 197, 200, 201, 205.
3/ Average of the data found in literature.
4/ Range of the data.
5/ Number of figures involved in calculating the statistics.

Table 11--Trace elements in Penaeidae, spp. shrimp, raw or processed,[1/] in ppm.

	Raw	Canned
Aluminum		
Arsenic	4.90[2/] 1.30 - 11.50 [3/] 5[4/]	7.00[2/] .08 - 13.90[3/] 2[4/]
Barium		
Boron		
Cadmium	.06 .05 - .08 3	
Chromium	.11 .05 - .19 3	
Cobalt	.22 .03 - .40 2	
Copper	59.20 .10 - 131.00 17	2.80 1
Fluorine	.90 1	
Iodine	.14 .05 - .23 2	.08 1
Lead	.50 .50 - .60 3	.50 1

Table 11--Trace elements in Penaeidae, spp. shrimp, raw or processed,[1/] in ppm. Cont.

	Raw	Canned
Manganese	1.20	
	1	
Mercury	.15	
	.02 - .50	
	19	
Molybd.enum	.49	
	.20 - 1.00	
	13	
Nickel		
Tin	1.80	
	1	
Selenium	.59	
	1	
Zinc	15.50	
	1	

1/ Data for this table taken from the following references: 42, 43, 47, 56, 59, 60, 65, 72, 106, 132, 172, 173, 174, 181, 196, 199, 207
2/ Average of the data found in literature.
3/ Range of the data.
4/ Number of figures involved in calculating the statistics.

Table 12--Vitamin content in mixed species of salt water shrimp,[1] raw or processed,[2] in 100 grams.

		Raw	Cooked	Canned	Dried and/or salted
Fat soluble					
Vitamin A	IU	30[3] 0 - 60[4] 3[5]		60[3] 59 - 60[4] 2[5]	212[3] 210 - 214[4] 2[5]
Vitamin D	IU	150 1			
Vitamin E	mg				
Vitamin K	mg				
Water soluble					
Thiamine	mcg	32 10 - 90 21	30[3] 1[5]	9 5 - 12 5	85 45 - 143 7
Riboflavin	mcg	62 13 - 160 20	75 30 - 120[4] 2	65 30 - 132 10	177 80 - 430 7
Niacin	mg	2.6 0.8 - 5.0 23	3.0 1	1.3 .3 - 2.2 12	4.9 2.2 - 7.2 7

Table 12--Vitamin content in mixed species of salt water shrimp,[1/] raw or processed,[2/] in 100 grams. Cont.

		Raw	Cooked	Canned	Dried and/or salted
Pantothenic acid	mcg	278 165 - 372 15	257 214 - 300 2	243 210 - 350 7	215 180 - 250 2
Pyridoxine	mcg	71 16 - 125 11	100 1	68 40 - 111 4	505 400 - 610 2
Biotin	mcg	1 1			
Folic acid	mcg	5 3 - 7 2	3 2 - 4 2	1.9 1.8 - 2.0 2	17 9 - 24 2
Vitamin B_{12}	mcg	1.5 1 - 2.5 3	.6 .5 - .7 2	.8 1	
Ascorbic acid	mg	1.6 0 - 4.6 8			0 1
Choline	mcg				

Table 12--Vitamin content in mixed species of walt water shrimp,[1/] raw or processed,[2/] in 100 grams. Cont.

1/ Identified in literature as "shrimp" or "prawn."
2/ Data for this table obtained from following references: 13, 14, 16, 17, 26, 28, 54, 63, 64, 75, 86, 89, 99, 100, 101, 103, 104, 107, 121, 123, 124, 130, 134, 136, 137, 138, 145, 151, 152, 158, 169, 177, 184, 187, 189, 190, 192, 200, 204, 208.
3/ Average of the data found in literature.
4/ Range of the data.
5/ Number of figures involved in calculating the statistics.

Table 13--Vitamin content in Penaeidae, spp. shrimp, raw or processed,[1] in 100 grams.

		Raw	Dried	Salted
Fat soluble				
Vitamin A	IU	172[2] 17 - 297[3] 3[4]	9[2] 1[4]	
Vitamin D	IU			
Vitamin E	mg			
Vitamin K	mg			
Water soluble				
Thiamine	mcg	22 1 - 100 21	117 90 - 160[3] 3	20[2] 10 - 30[3] 2[4]
Riboflavin	mcg	191 15 - 650 20	246 97 - 340 3	105 100 - 110 2
Niacin	mg	3.2 1.0 - 5.1 19	6.0 3.4 - 9.5 3	2.5 1.6 - 3.4 2
Pantothenic acid	mcg	210 1	616 1	

Table 13--Vitamin content in Penaeidae, spp. shrimp, raw or processed,— in 100 grams. Cont.

		Raw	Dried	Salted
Pyridoxine	mcg	170	30	
		1	1	
Biotin	mcg		9.3	
			1	
Folic acid	mcg	19.2	73	8.8
		1.8 - 29.3	53 - 89	
		3	4	1
Vitamin B_{12}	mcg	1.7	7.4	
		.9 - 2.5	1.0 - 10.4	
		3	5	
Ascorbic acid	mg	1.7	0	0
		0 - 4.8		
		17	1	1
Choline	mcg		162	
			135 - 218	
			4	

1/ Data for this table taken from the following references: 27, 45, 61, 63, 79, 84, 88, 106, 112, 129, 136, 138, 150, 172, 173, 178.
2/ Average of the data found in literature.
3/ Range of the data.
4/ Number of values involved in calculating the statistics.

BIOCHEMISTRY AND PHYSIOLOGY OF SHRIMP - EFFECT ON USE AS FOOD

ABSTRACT

Biochemical and Physiological factors affecting shrimp composition and quality are discussed. Particular emphasis is placed on the molting cycle and related events and on post-mortem enzymic activity.

BIOCHEMISTRY AND PHYSIOLOGY OF SHRIMP - EFFECT ON USE AS FOOD

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INTRODUCTION

The shrimp fishery is economically one of the most important fisheries in the United States. The present selling price of shrimp places them in the gourmet price range. However, shrimp quality often falls far short of the quality expected for a gourmet product.

Although a large number of investigators have looked at factors affecting shrimp quality, few have attempted to differentiate effects caused by natural enzymes and/or physiology of the shrimp from those caused by postmortem bacterial growth. The shrimp is a member of phylum arthropoda. It resembles an insect in its biochemical and physiological characteristics more closely than it does a teleost fish. However, very few investigators working on technological problems in shrimp have consulted the large volume of insect literature. In this paper we will try to point out some areas where insect work can provide valuable insights into problems which affect the use of shrimp as food.

Biochemistry of Structural Features

The shrimp abdomen (tail) which is the portion most often

consumed consists largely of muscles. Crustacean muscle fibers are crosslinked (Atwood, 1972). Very little is known about shrimp muscle proteins and the subsequent changes they undergo during refrigerated storage. Lightner (1973) has described postmortem changes in shrimp tissue maintained at temperatures ranging from 10-30°C.

The midgut (sand vein) which runs through the abdomen often contains bacteria, partially digested material, digestive enzymes and sand (Figure 1). In the female the midgut is surrounded by ovaries, which may constitute a sizable portion of the shrimp weight. Brown and Patlan (1974) have published a series of photographs which illustrate very vividly the size of penaeid shrimp ovaries. Autodigestion of the midgut results in bacterial invasion, destruction of tissue and undoubtedly promotes spoilage (Carroll et al., 1968).

Near the outer edge of the abdomen (Figure 2) is a tough layer (epidermis) which contains much of the pigment, usually melanin and/or astaxanthin (Katayama et al., 1972).

The cephalothorax (head) of the shrimp which is not used for food in the United States contains a high level of digestive enzymes (as evidenced by rapid autodigestion of the links between the cephalothorax and the abdomen). The cephalothorax also contains about 50 to 80% of the bacterial population of the shrimp (Green, 1949b; Williams et al., 1952). Consequently failure to remove the cephalothorax and to wash the digestive enzymes from the abdomen may result in rapid spoilage. This has been disputed

by Koburger et al. (1974). However, reports by Green (1949a) and yearly experiments by students in my seafood course have indicated that shrimp with heads-on have about a 5 day shelf-life versus 14 or more days shelf-life for deheaded shrimp.

The cephalothorax may also contain high levels of melanin precursor, which if not removed when melanin production occurs, may stain the meat in the abdomen, thus lowering its quality.

Molting Cycle

In order to grow, the shrimp must shed the exocuticle. The molting cycle for shrimp is briefly described in Table 1. The biochemical events of the molting cycle are complex and have not been fully elucidated. However, those that are known suggest that many of the technological problems associated with shrimp m may be the result of various events occuring in the molting cycle.

Probable technological problems associated with the molting cycle in shrimp are listed in Table 2. The relationship between the molting cycle and melanosis has not been recognized by most investigators. During the intermolt period the shrimp builds up melanin precursor, probably a 3-O-phosphate or 3-O-sulfate derivative of n-acetyl dopamine as in the insect (Bodnaryk et al., 1974). Preliminary data indicate that deproteinated shrimp extracts incubated with O-diphenoloxidase (E.C.1.19.3.1) and orthophosphoric monoester phosphohydrolase (E.C.3.1.3.2) tend to develop more melanin pigment than extracts incubated with O-diphenoloxidase alone (Cobb, unpublished). N-acetyl dopamine poly-

merizes and reacts with proteins to form the new sclera (Fig. 3). The buildup or activation of catabolic enzymes prior to molt undoubtedly has technological significance but at present no data is available on its role in quality deterioration.

Recently a 5 pound box of shrimp, opened in our laboratory, illustrated dramatically the problems associated with molting. Approximately one third of the shrimp had old shells. All of these shrimp had a large amount of black discoloration (melanosis). Another third of the shrimp had shells which were beginning to loosen or had already been shed. None of these shrimp had melanin development but those with shells which were just beginning to loosen had a tough unappealing outer surface when cooked and peeled. The soft-shell shrimp in this group were mashed and broken. The final third of the shrimp had new shells and no melanin pigment development but appeared to shrink considerably when cooked.

Increasing evidence suggests that penaeid shrimp molt during the dark of the moon and most shrimp in rapidly growing populations appear to molt at about the same time (Cobb, unpublished). Unfortunately, we probably cannot schedule harvest of wild shrimp at a time during the molting cycle which would produce the best quality product. Also very large shrimp molt only occasionally, making estimation of the proper harvesting time difficult. Shrimp culture operations, however, could take advantage of such knowledge to produce a shrimp of superior quality from rapidly growing populations. This shrimp would have to

be harvested during the intermolt stage. We are presently working on enzyme markers which we feel will eventually allow us to determine the exact time of harvest for mariculture shrimp.

Osmoregulation and Shrimp Quality

A number of investigators have noted the relationship between salinity and free amino acid content in crustaceans. Many of these studies have been recently reviewed by Schoffeniels and Giles (1970). Glycine and proline are the main osmoregulators in penaeid shrimp with alanine playing a less certain role (Cobb et al., 1975). Glycine has been identified as the cause of the sweet taste in shrimp (Hashimoto, 1965; Rajendranathan, Nair and Bose, 1965). Glycine is washed from ice-stored shrimp at a logarithmic rate. Half-life of the free amino acid content of shrimp is dependent on size of the shrimp, method of deheading and bacterial levels (Fig. 4). Simultaneous loss of ammonia and free amino acids has been used to develop a chemical test for shrimp quality (Cobb et al., 1973; Cobb and Vanderzant, 1975). Loss of free amino acids and, hence, flavor to the washing action of melt water is offset by the removal of ammonia, melanin precursor and bacteria.

When the shrimp goes through the molting cycle, it takes in fluid to expand to fill the new shell. Since glycine and proline are mainly responsible for osmoregulation, the shrimp

either has to make available more glycine and proline or else maintain in body fluids at a high salinity which could disrupt muscle proteins. Preliminary evidence suggests that the free amino acid content in shrimp is rapidly increased when molting occurs. Experiments to determine the rate at which the free amino acid content changes when shrimp are challenged with lower salinities were conducted (Cobb and Conte, unpublished data). When intermolt shrimp were challenged with salinity changes, based on a constant shrimp water content, there was a rapid, linear decrease in free amino acid content. However, when the same experiments were conducted with molting shrimp, based on a constant water content, free amino acid levels were very eratic and generally much higher than in intermolt shrimp. This indicated that free amino acid content increased rapidly during molting. An increased free amino acid content relative to other components in newly molted shrimp suggests that the newly molted shrimp might be more flavorable than those in latter stages of molt.

Postmortem Ammonia and Urea Production

During the postmortem ice storage period, ammonia is produced in penaeid shrimp muscle at the rate of approximately one mg/100 g/day (Cobb et al., 1974). Ammonia is also simultaneously leached from shrimp at a logarithmic rate resulting in little apparent increase. Urea is produced by the action of arginase and is leached at approximately the same rate as ammonia. Both urea and ammonia are leached from the shrimp at approximately the same rate as

glycine, allowing development of a series of equations for estimating endogenous enzymic activity in intact shrimp tails.

Enzymic production of ammonia during ice storage causes a postmortem pH change from about 7 to 8 in shrimp muscle (Bailey et al., 1956; Bethea and Ambrose, 1962; Vanderzant and Nickelson, 1971). This in turn causes an increased production of ammonia as pH optimum of the main ammonia producing enzyme(s) is approximately 8.5 (Yeh, C. S. and Cobb, B. F., unpublished).

In working with commercially "fresh" shrimp (probably not freshly caught) urease activity was detected (Cobb and Vanderzant, 1971). Subsequent investigations have indicated that although the enzyme system which produces ammonia in postmortem shrimp has some similar properties to urease, it probably is not urease. The initial urease activity detected may have been from bacteria. Ammonia production from bacterial urease may become important during the latter stages of ice storage, particularly if sufficient urea has not been leached from the shrimp.

Role of Proteolytic Enzymes in Shrimp Quality Deterioration

Cobb et al. (1974) reported both considerable anabolic (amino acid producing) and catabolic (amino acid reducing) activities in postmortem shrimp tails. Eitenmiller (1974) found cathepsin D type activity in P. setiferus muscle. Very little activity was evident in the pH 7-8 range, the normal pH range of postmortem shrimp. Arylamidase activity, which may be responsible for intracellular catabolism of proteins, has also been detected

in P. setiferus muscle (Bauer and Eitenmiller, 1974a,b). Considerable activity was evident in the pH 7-8 range. Tryptic activity in the digestive juices of penaeid shrimp has been studied (DeVillez and Buschlen, 1967; Gates and Travis, 1969, 1973). The pH optimum of shrimp trypsin was rather broad, occurring over the range 7.0-9.5. Digestive enzymes would be important in the midgut and on the anterior portion of the abdomen because of contamination during removal of the cephalothorax. Proteolytic digestive enzymic activity levels have been demonstrated to vary with the stage of the molting cycle and season in Palaemon serratus (Van Wormhoudt et al., 1972, 1973). Presumably, proteolytic digestive enzyme levels would also be higher in feedy than in non-feedy shrimp. Obtaining shrimp at a period of low levels of digestive enzyme activity may have allowed Koburger et al. (1974) to keep heads-on shrimp on ice for longer periods than normally reported by other investigators.

Enzymic and Non-Enzymic Darkening (Melanosis) during Postmortem Storage

The relationship between molting and the buildup of melanin precursor has been briefly examined. The melanin precursor is either dihydroxyphenyalanine (DOPA), a derivative of DOPA such as N-acetyldopamine in which the dihydroxy ring is unaltered, and/or a stable storage form such as a 3-O-phosphate derivative (Fig. 3). The melanin precursor is located near the periphery in the shrimp tail and is rapidly removed by washing (Cobb et al., 1976). As evidenced by rapid darkening, considerable melanin precursor may be stored in the cephalothorax.

It is uncertain whether postmortem melanin formation is due to nonenzymic or enzymic oxidation. It is also uncertain whether one enzyme or several are involved in the oxidation of tyrosine and DOPA or DOPA derivatives. Further confusion is imparted by the variety of names used for the enzyme O-diphenol-oxidase (E.C.1.19.3.1) - tyrosinase, phenolase, phenoloxidase, polyphenoloxidase and catechol oxidase. In addition to O-diphenoloxidase, the various enzymes reported to play a role in the oxidation of phenols in arthropods are tyrosine hydroxylase, laccase, polyphenol peroxidase and tyrosine peroxidase (Neville, 1975).

Monohydric phenols such as tyrosine are fairly stable to chemical oxidation while dihydric phenols such as DOPA are very unstable and readily undergo oxidation, forming colored polymers in the process (Fig. 5). The autooxidation of dihydric phenols is catalyzed by light. DOPA is oxidized to dopaquinone either by enzymic or non-enzymic means and then undergoes rearrangement and ring formation, forming 5,6-dihydroindole which then forms melanin (Neville, 1975). N-acetyldopamine also forms melanin. Sodium bisulfite, a reducing compound, has been employed for years to inhibit melanin development. Bacteria also tend to inhibit melanosis in shrimp (Cobb and Vanderzant, 1971). Quinones or polymers of quinones readily condense with amino groups on proteins in an irreversible tanning process. Once the pigment is bound, it cannot be removed without destruction of the protein.

When the shrimp is near molting or taken from organic rich

waters it may blacken excessively, particularly if the washing action of melted ice is absent (Cobb and Vanderzant, 1971; Vanderzant et al., 1973; Cobb et al., 1976). The tendency for shrimp to darken more readily in organic rich waters has also been reported by fishermen. This phenomenon may be caused by the more abundant food stuffs in such waters which promote more rapid growth and hence more frequent molting.

Postmortem Nucleotide Degradation

Flavor and probably texture of shrimp are influenced by postmortem nucleotide breakdown. Stone (1971) and Flick and Lovell (1972) have demonstrated that the breakdown of nucleotides is as follows: adenosine triphosphate (ATP) is degraded in sequence to adenosine diphosphate (ADP), to adenosine monophosphate (AMP), to inosine monophosphate (IMP), to inosine (Ino), to hypoxanthine (Hx). The exact consequences of the conversion of ADP and IMP to Hx are unknown, but probably result in loss of meaty flavor (Hashimoto, 1965; Jones 1969) and may result in bitterness as observed by Carroll et al. (1968). Whether or not nucleotides are leached from shrimp during ice storage at or near the same rate as amino acids has not been established.

The loss of ATP should bring on rigor in shrimp as has been reported by Lightner (1973). However, Flick and Lovell (1976) reported that rigor was not observable in shrimp. Recent studies in our laboratory have by myograph measurements established that rigor deinitely occurs in shrimp (Wilaichon, 1976). These studies have also indicated that there might be a relationship

between toughening and rigor in shrimp. The low amount of rigor usually observed in shrimp may be caused by the postmortem basic tissue pH.

SUMMARY

As handling techniques for shrimp improve and bacterial degradation is reduced, the biochemical problems associated with shrimp quality deterioration will become more evident. Compared to non-aquatic food animals, we know very little about the postmortem changes in shrimp. For instance, we know very little about the postmortem behavior of shrimp muscle proteins. The establishment of a viable mariculture industry will undoubtedly increase pressure to know more about the biochemistry of the shrimp as it pertains to quality deterioration.

Table 1. Events of possible technological significance occuring in the molting cycle of shrimp.

Stage of Molt	Status of Cuticle	Event
Postmolt	Initially soft becoming increasingly harder	1. Continued water absorption 2. Tissue growth begins
Intermolt	Hard	1. Main tissue growth 2. Accumulation of melanin precursor (storage form of N-acetyldopamine) and other organic reserves
Premolt	Hard but gradually thinned	1. Epidermal and hepatopancreas activation 2. Activation of N-acetyldopamine precursor 3. Epicuticle formation begins utilizing N-acetyldopamine 4. Major portion of skeletal reabsorption 5. Ecdysial sutures open
Molt	Soft	1. Beginning water absorption

Table 2. Possible technological effects of the molting cycle on penaeid shrimp.

Stage of Cycle	Technological Effects
Postmolt	1. Difficulty in sorting and peeling 2. More rapid spoilage 3. Broken or misshapen shrimp 4. Excessive weight loss on cooking 5. Excessive weight loss during processing
Intermolt	1. Difficulty in peeling (early stage) 2. Melanosis (latter stage) 3. Toughness (latter stage)
Premolt	1. Melanosis 2. Toughness
Molt	1. Same as for premolt stage

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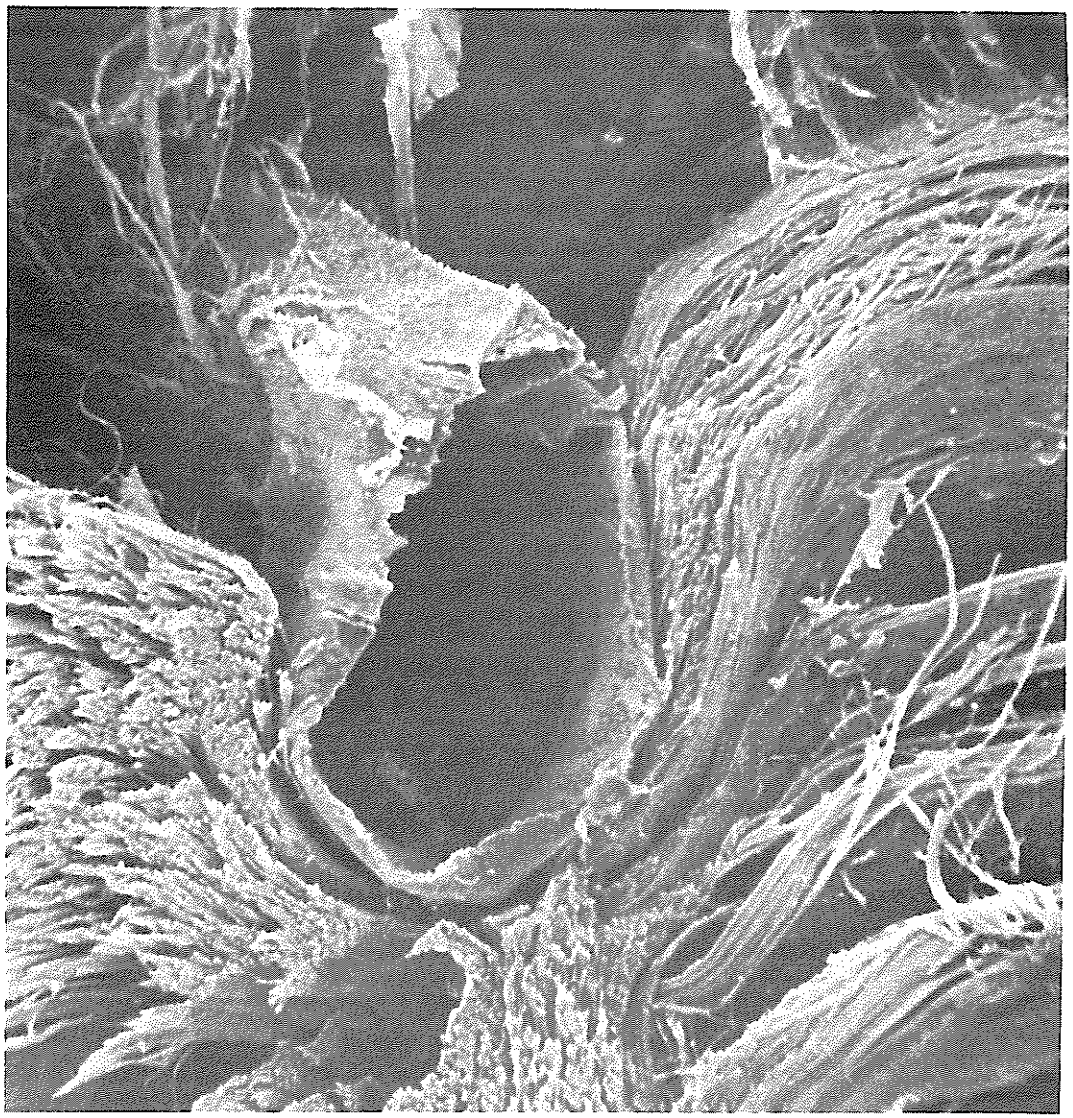


Fig. 1--Scanning electron micrograph (100x) of shrimp midgut.

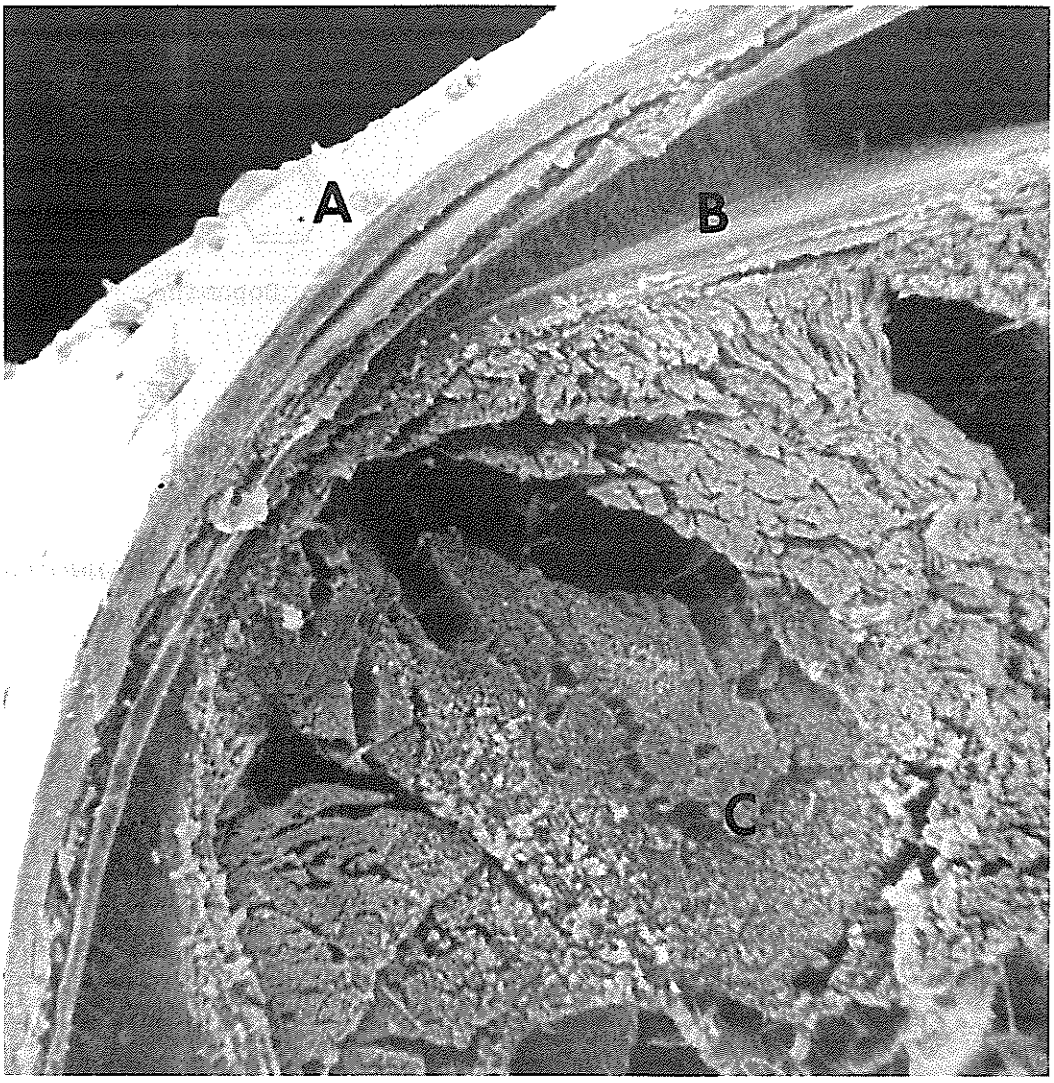
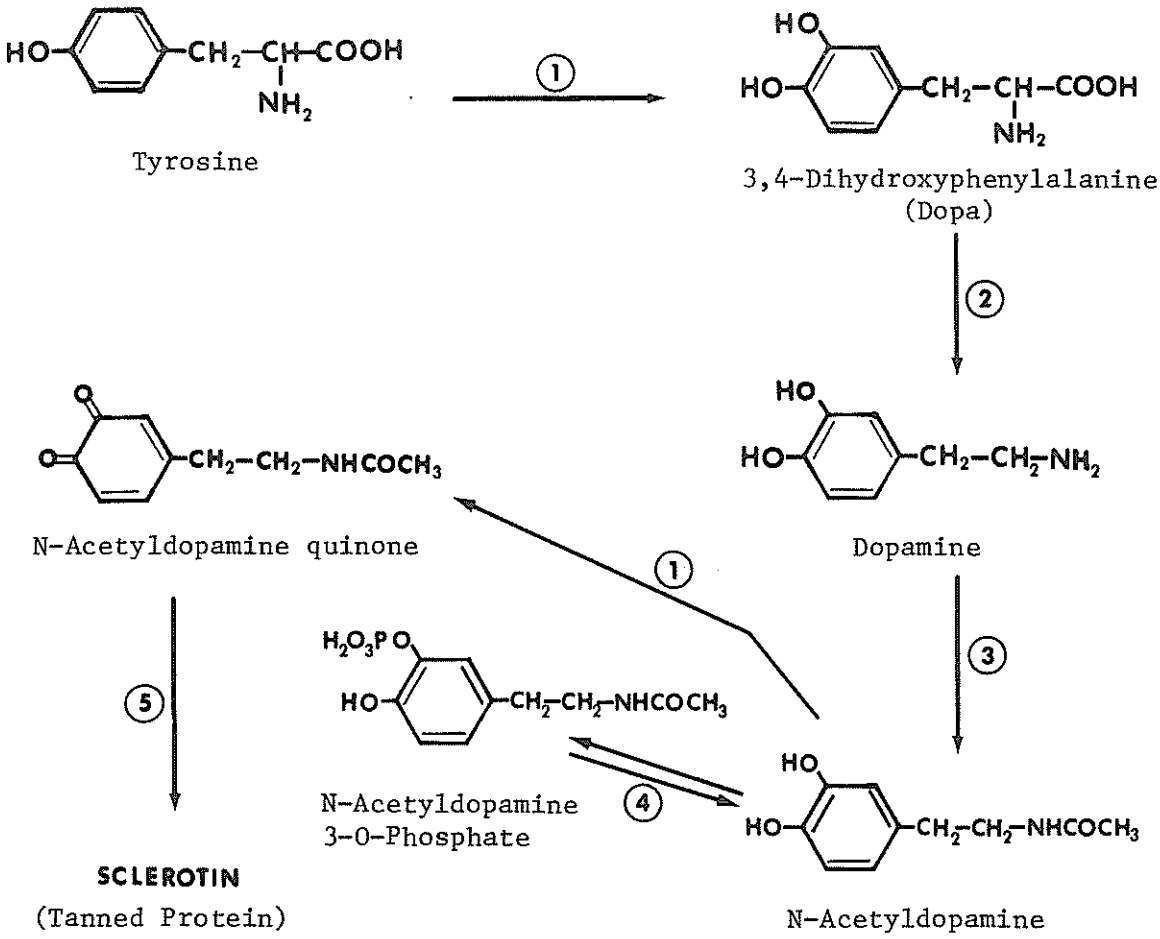


Fig. 2--Scanning electron micrograph (150x) peripheral area of shrimp abdomen. A - cuticle; B - epidermis; C - muscle tissue.



ENZYMES

1. **O-Diphenoloxidase**
2. **Dopadecarboxylase**
3. **Transacetylase**
4. **Phosphatase**
5. **Tanning Enzyme**

Fig. 3--Role of phenols in formation of new cuticle.

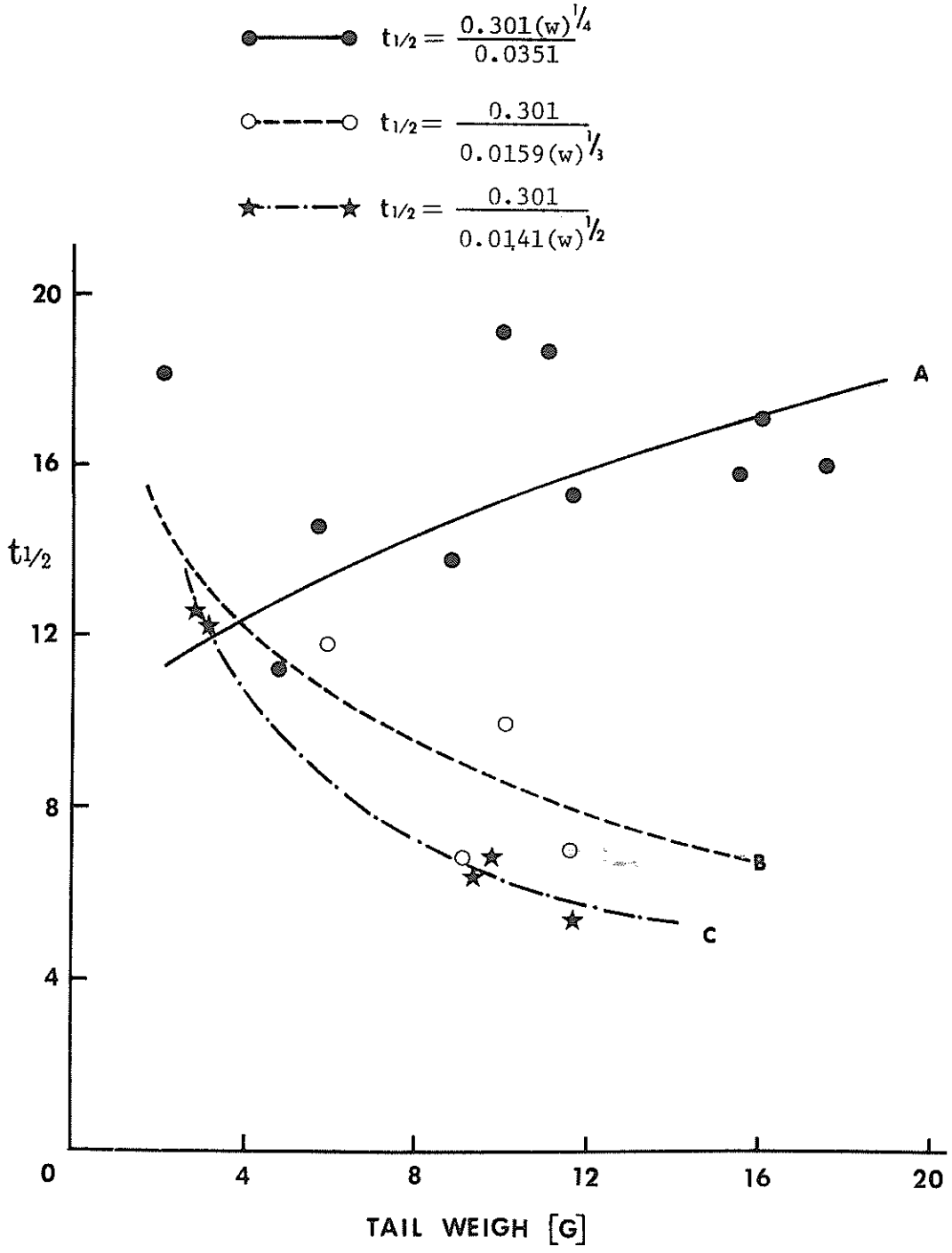
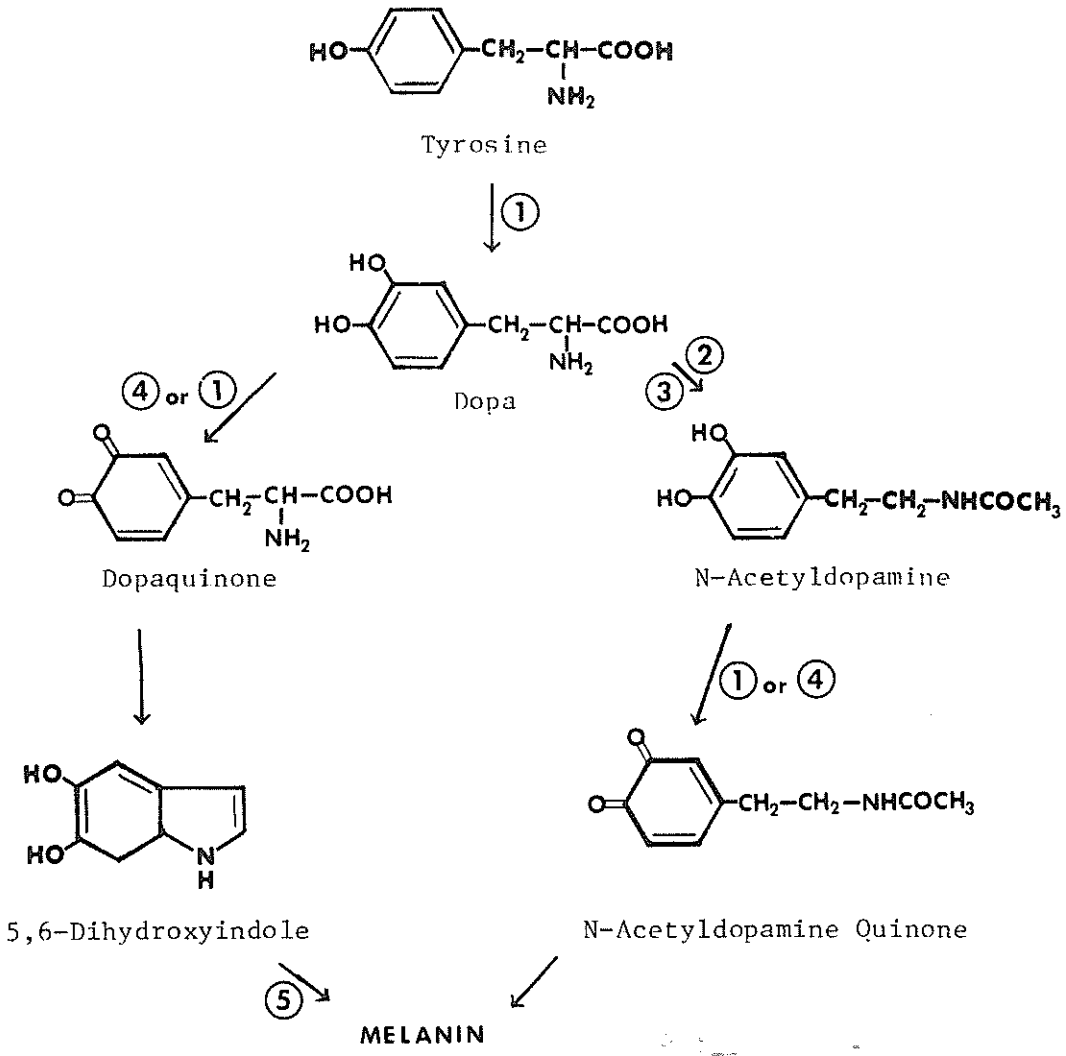


Fig. 4--Half-life of free amino acid content in different groups of shrimp. (A) cephalothorax removed by pinching, bacterial counts $< 10^6$/g; (B) cephalothorax removed by pinching, bacterial counts $> 10^6$/g; (C) cephalothorax snipped off, bacterial counts $> 10^6$/g.



1. O-Diphenoloxidase
2. Dopadecarboxylase
3. Transacetylase
4. Non-enzymic autooxidation
5. Condensation

Fig. 5--Postmortem formation of melanin in shrimp.

VARIATION OF THE CHOLESTEROL CONTENT OF SHRIMP TAILS - Charles Neal, Bryant F. Cobb III, C. W. Dill, Texas A&M University, TAES, and Virginia Sidwell, National Marine Fisheries Service, Fishery Products Technology Laboratory, Southeast Utilization Research Center

ABSTRACT

Reported values of the cholesterol content of shrimp vary considerably. In this study possible factors causing this variation, including method of analysis, season, diet, size, ovarian development, stage of molting cycle and species were surveyed. A reliable method of analysis for cholesterol content in shrimp was established. Shrimp size and state of ovarian development were found not to affect cholesterol content. Species and seasonal differences caused significant variation in the cholesterol content.

VARIATION OF THE CHOLESTEROL CONTENT OF SHRIMP TAILS

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INTRODUCTION

In recent years many investigators have sought to elucidate the relationship between cholesterol consumption and the development of arteriosclerosis in man. The relationship is still not well understood, but most medical authorities recommend that people in the high risk category for heart disease should limit their consumption of cholesterol (Baylor College of Medicine, 1974). Shrimp is one of the foods with reported high levels of cholesterol which has been excluded from low cholesterol diets. However, the diversity of cholesterol values reported for shrimp is very great (Sidwell et al., 1974). Discounting older reports before 1950, which present values for shrimp cholesterol content as low as 40 mg/100g (Renaud, 1949), the range of reported values is 125-226 mg/100 g (Table 1). These differences suggest that the method of analysis employed by some of the investigators may have given erroneous

results and/or biological variables may have affected the cholesterol content of the shrimp. The extent of variation produced by the method of analysis is difficult to evaluate. When colorimetric methods were used, inaccuracy could have resulted from color development by compounds other than cholesterol (Searey and Berquist, 1960).

The object of the present investigation was to analyze various biological variables for possible influence on cholesterol content of the edible flesh of shrimp. Variables investigated for possible influence on cholesterol content are size, ovarian development, species, molting, season and diet.

EXPERIMENTAL

The AOAC (1975) method for determination of cholesterol in eggs was used, except for the following modifications:

1) Samples were prepared by grinding 30-40 grams of peeled-deveined shrimp tails into a paste with a mortar and pestle and taking a 5 gram portion from this paste for each extraction. All analyses involved triplicate extractions.

2) Fifteen ml conc. KOH was added to the sample instead of 10 ml.

3) The sample was heated 45 minutes in a boiling water bath instead of 3 hrs on a steam bath.

4) Ethanol was added to the solution in the separators to disrupt any persistent colloids.

5) After filtration through anhydrous Na_2SO_4 and evaporation of the ether on a steam bath, the residue was transferred to a 25 ml volumetric flask by adding about 5 ml of $CHCl_3$, swirling and

pipeting with a disposable pasteur pipet (4 times).

6) The cholesterol was quantitated by gas chromotography as follows: Squalene was added to each sample as an internal standard and the samples were diluted to 25 ml. A standard solution was prepared with squalene and 99%+ pure cholesterol obtained from Applied Science, Inc. (State College, Pa.). The standard contained the same amount of squalene as each sample. A glass column (1.75 m-length, 5mm-inside diameter) was packed with 3% SE-30 on 80/100 mesh Chromosorb G. The temperature conditions were: injector - 250°C, column - 250°C, detector - 275°C. A Barber-Coleman Series 5000 chromatograph equipped with a Hewlett-Packard 3380A integrator was used.

RESULTS AND DISCUSSION

Recovery of added cholesterol ranged from 89-105% with the average recovery in the first trial being 95% and in the second trial 101% (Table 2). Reproducibility of the method was excellent with standard deviations of about 3% of the total cholesterol content (Table 3).

Shrimp (Penaeus setiferus) of various sizes were analyzed for cholesterol content. The large shrimp (21 g tail weight) averaged about 5% less cholesterol than the small shrimp (1-5 g) (Fig. 1). This variation was not significant and does not account for the large variations reported in the literature.

Shrimp, like many aquatic animals, produce large numbers of offspring in order to ensure survival of a few individuals in a hostile environment. P. setiferus specimens have been

reported to produce 500,000 to 1 million eggs per spawn (Anderson et al., 1949). At maturity the ovaries extend the full length of the tail. The production of so many eggs could cause a considerable drain on the resources of the female's body. Rock shrimp (Sicyonia brevirostris) were used to determine the effects of ovarian development on cholesterol content. No significant difference in cholesterol content was found between samples with developed ovaries and those without ovaries (Table 4).

Some differences in reported cholesterol values could be explained by species differences. Values obtained thus far indicate that samples of P. setiferus and P. aztecus have about the same cholesterol levels, whereas P. duoranum samples have levels about 15% higher. Thompson (1964) also reported that P. setiferus and P. aztecus had about the same cholesterol contents.

The diet of the shrimp varies seasonally as the flora and fauna of the waters vary. Perhaps sterol consumption would vary enough to cause some change in body cholesterol composition. The diet would also vary with location. Feeding studies will be performed with pond-raised shrimp to investigate this factor.

The periodic molting of shrimp could affect cholesterol content. Much tissue is shed and reconstructed in this process, requiring cholesterol. Some cholesterol is also used to synthesize the molting hormone. The shrimp must obtain all required cholesterol from dietary sterols (Kanazawa et al., 1971), therefore the dietary intake might not be adequate to replenish

cholesterol levels when the shrimp is growing rapidly and molting frequently. If this is the case, then when the animal grows most rapidly in the warm months (Lindner and Anderson, 1956; Kutkuhn, 1962) the cholesterol level in the flesh should be lower. Variation of cholesterol content might also be observed during the course of each molt.

Evidence obtained thus far indicates lower cholesterol contents for summer and fall shrimp than winter shrimp (Table 5). This variation is the greatest observed. Possibly dietary differences and differences in molting frequency produced this seasonal variation. However, these two factors must be examined in isolation to determine if this is the case. Investigations of biological factors which may influence the cholesterol content of shrimp are being continued. The effect of season on cholesterol content will be investigated further to determine whether the low values measured during the fall of 1975 represent a general phenomena.

Table 1. Variation of values reported for cholesterol in shrimp.

Cholesterol (mg/100 g)	Investigator
125	USDA Handbook (1963)
138	Kritchevsky and Tepper (1961)
150	Feeley et al. (1972)
157	Thompson (1964)
200	Kritchevsky et al. (1967)
226	Pihl (1952)

Table 2. Recovery determinations for method of analysis.

Trial	Average % recovery of added cholesterol[a]	Range
1	95%	89-99%
2	101%	97-105%

[a] % recovery was determined by subtracting cholesterol content of pooled shrimp sample from that of pooled shrimp sample with added cholesterol.

Table 3. Reproducibility of cholesterol analyses.

Trial	Average Content (mg/100 g)	Standard Deviation
1	187[a]	5.0
2	183[a]	5.5

[a] Average of 15 analyses of shrimp ranging from 1-5 g, taken from the same location during November 1975.

Table 4. Effect of ovarian development on cholesterol content of rock shrimp (Sicyonia brevirostris).

Sample	Average Content
Ovaries present	174 mg/100 g
Ovaries removed	168 mg/100 g
No ovarian development	170 mg/100 g

Each value is an average of three determinations.

Table 5. Effect of season on cholesterol content of *P. setiferus*.

Date Obtained	Cholesterol (mg/100 g)
Aug. 1, 1975	134[a]
Nov. 25, 1975	187
Jan. 7, 1976	183

[a] Average of 5 samples.

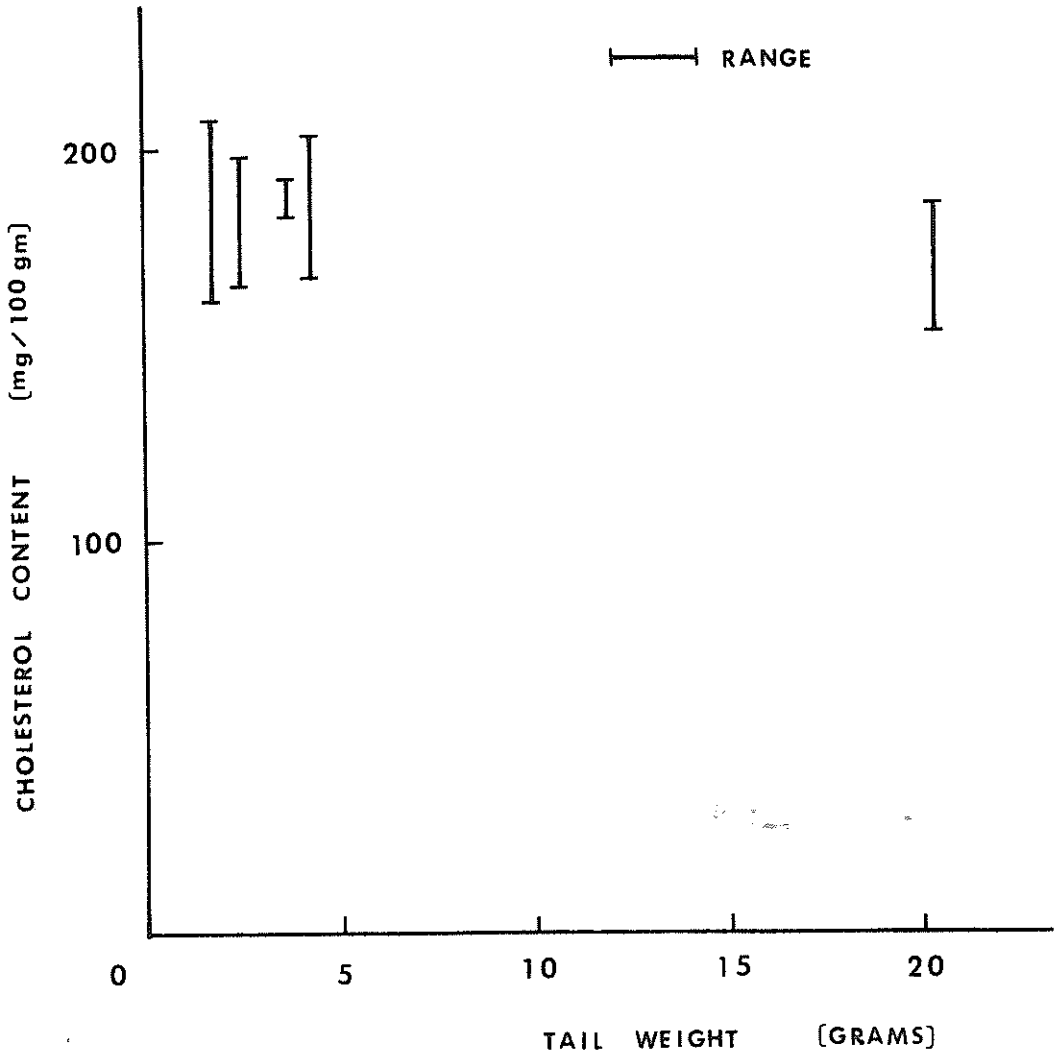

Fig. 1 — Change of cholesterol content with increasing size of P. setiferus

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WATER MIGRATION AND DEHYDRATION IN STORED FROZEN BREADED SHRIMP

ABSTRACT

Moisture migration and dehydration in frozen breaded shrimp were studied with respect to freezing rate, texture and equilibrium temperature of the breading material, and storage time. It was desired to determine the combined effect of these mass transfer mechaisms on the changes in weight of the shrimp and breading material.

Breaded shrimp, consisting of 50% shrimp, were produced by using two freezing rates, two textures of the breading material, and two equilibrium temperatures of the breading material. At one week intervals, samples were removed from the freezer and analyzed. Initial, final, and dry weights of the shrimp and breading material were recorded. These weights were then statistically analyzed in a factorial arrangement of treatments.

Results of the statistical analysis indicated:

1) The fast freezing rate produced a much smaller weight change in the shrimp and breading as compared to the slow freezing rate.

2) For each texture of breading material used the shrimp lost approximately the same amount of weight which was, in turn, gained by the breading material. However, the fine texture lost more moisture to the surroundings than the coarse texture, resulting in a smaller change in weight of the breading material.

3) Equilibrium temperature of the breading material did not alter the moisture gained by the breading material from the shrimp. Less moisture was lost to the surroundings by the breading material

at 40°F than at 82°F, resulting in a larger weight gain for the breading material at 40°F.

4) The initial weight change observed in the shrimp and breading declined as the storage time increased.

Correlation coefficients were calculated for all possible pairs of the four characteristics. Sixteen pairs were significantly correlated.

It was concluded from the results that breaded shrimp produced by using a fast freezing rate, 82°F equilibrium temperature, and fine texture breading material will give smaller changes in weight for the shrimp and breading material and, therefore, smaller gains in percent breading.

WATER MIGRATION AND DEHYDRATION IN STORED FROZEN BREADED SHRIMP

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Introduction

Standards of Identity, section 36.30 (b), of the United States Food and Drug Administration specifies that breaded shrimp, that are produced and marketed in the United States, should contain not less than 50% shrimp material unless it is a composite unit, for which a different standard of identity applies. However, when shrimp are battered, breaded, packaged, and placed in frozen storage several physical, chemical, and microbiological changes occur, one of which is the migration of water molecules from the shrimp to the breading material. Water molecules migrate from the shrimp to the breading material because the breading material used to coat the shrimp has a lower percent moisture than the shrimp meat. This imbalance in percent moisture, that allows water to migrate from the shrimp to the breading material, results in a dehydrational loss of weight in the shrimp component of the breaded product. In addition, weight losses also may occur due to evaporation, sublimation, and the type of equipment used to process the product.

The value and production of breaded shrimp have been increasing yearly. (1). In 1972 the wholesale value of the United States' production of fishery products was 1.575 billon dollars. (2). The value of the breaded shrimp produced in 1972 by the United States was 140.9 million dollars (3, 4, 5). These values indicate that breaded shrimp represented 8.9% of the wholesale value of the United States' fishery products in 1972. In 1973 the value of the breaded shrimp produced in the United States was 174 million dollars (6).

To protect the consumer and maintain quality, the Food and Drug Administration conducts periodic inspections of breaded shrimp to determine the percent composition. If a shipment is found to contain more than 50% breading material the product is deemed to be in violation of the standard of identity and will be removed from the market, regardless of its composition at the time of packaging. The contention of the Food and Drug Administration is that because of a differential in price between the breading material and the shrimp, the breading material normally being cheaper than the shrimp, there is a tendency for the processor to over-bread his product. Confiscation of the product and the time involved in litigation deprives the processor of marketing opportunities and results in heavy financial losses. A review of literature indicated that no experimental results have been reported concerning the causes and prevention of dehydration occurring in breaded shrimp during processing and storage.

Therefore, this investigation was undertaken to determine the causes and prevention of the moisture migration dehydration

occurring in the breaded shrimp. The variables studied were: 1) texture of the breading material, 2) equilibrium temperature of the breading material, 3) freezing rate, and 4) storage time.

MATERIALS AND METHODS

This investigation was undertaken in order to study the effects of four selected variables on the extent of moisture migrating within frozen breaded shrimp. The migration takes place from the shrimp meat into its coating of breading material. Two treatment levels for each of three variables were chosen for the study. These variables were 1) fine and coarse texture of breading material, 2) $40^{o}F$ and $82^{o}F$ as the temperatures at which the batter and breading were applied to the shrimp, 3) slow and fast rates of freezing the breaded shrimp (slow taking about 4 hours; fast taking about 3 to 4 seconds). Together with these variables were five periods of elapsed storage time - 1, 2, 3, 4, and 5 weeks - during which the frozen breaded product was kept at $-6^{o}F$. These levels of texture, temperature, freezing rate, and periods of storage time yielded 40 different groups of frozen breaded shrimp. Since all the operations for each group were performed on five shrimp individually, a total of 200 randomly selected shrimp were prepared and used in studying the effects of the four variables on 1) the weight losses of shrimp meat during frozen storage, and 2) the concomitant weight gains of the breading coatings of these frozen shrimp. The two sets of gravimetric data, i.e. weight-loss of shrimp meat and weight-gain of breading coating, and two corresponding sets of percentage values based on initial weights of

shrimp and breading material, each represented measurements that correspond to those of a 2 x 2 x 2 x 5 factorial arrangement of treatments in a randomized block design with 5 replicates. Each of these four sets of data was subjected to standard analysis of variance.

Sources of Raw Materials

A 20-lb. lot of 40-count size, unpeeled, headed, freshly-frozen shrimp[1] (Penaeus setiferous) was received by air freight from Tampa, Florida. Prior to use in the present study, the shrimp were stored no longer than two weeks in the cold storage room in the Department of Food Science, L.S.U., at -6°F. Commercial breading materials consisting of one 50-lb. bag of batter mix[2] and two 50-lb. bags of breading[2] (one of fine texture, the other coarse) with the ingredient composition of the two being the same, were obtained for preparing the coating of breading material that was applied to the shrimp.

Preparation of Breaded Shrimp

About ten pounds of the frozen shrimp were removed from the cold room and thawed by immersion in tap water. After being thawed, the shrimp were immediately shelled with removal of the terminal segment, deveined, washed in cold water, and placed in a bed of crushed ice to insure no change in quality. After approximately 400 shrimp had been peeled, deveined, and washed, a total

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[1]Supplied by Treasure Island Seafood Company, Tampa, Florida.

[2]Supplied by Modern Maid Food Products, Inc., Ponchatoula, Louisiana.

of 100 shrimp for the slow freezing operation were removed, one at a time at random, from the crushed ice. These were then blotted with absorbent paper to remove excess adhering moisture and weighed. The weighing was done in order to determine the proper amount of breading material necessary in the coating to yield a breaded product that consisted of 50% shrimp and 50% breading material.

The batter had been prepared by mixing two parts batter mix with three parts water and allowing it to come to the proper temperature. Each shrimp, after being weighed, was immediately coated with batter and then dipped into the breading material until the weight of the coating was equal to that of the shrimp. The resulting individually prepared 50% - 50% breaded shrimp was placed in a tared, appropriately identified sample bag of Nasco Whirl-Paks material. These particular bags were used because of their impermeability to gasses and moisture.

For the slow freezing operation, each individual bag containing a single breaded shrimp was placed in the freezer at $-6^{o}F$ immediately after the bag was sealed.

For the fast freezing operation, a total of 100 randomly selected specimens from a second batch of about 400 peeled, deveined, and washed shrimp were battered and breaded in the same manner as described above. Before being placed in its appropriately coded Nasco Whirls-Paks bag, each individually weighed 50% - 50% breaded shrimp was immersed by means of tongs for 3 or 4 seconds in liquid nitrogen for longer than 4 seconds, the texture of the shrimp was damaged and an inferior product resulted.

Gravimetric Measurements

The resulting eight treatment-groups (2 texture x 2 temperature x 2 freezing rate) of breaded shrimp that were stored at -6°F for sampling at weekly intervals were:

Group Number	Breading Material		Freezing Rate
	Texture	Temp. (°F)	
1	Fine	82	Slow
2	Fine	40	Slow
3	Coarse	82	Slow
4	Coarse	40	Slow
5	Fine	82	Fast
6	Fine	40	Fast
7	Coarse	82	Fast
8	Coarse	40	Fast

At the end of each of the five frozen storage periods - 1, 2, 3, 4, and 5 weeks - five shrimp from each of the above eight groups, or a total of 40 specimens weekly, were removed from the freezer. Each of the 40 bags containing a single frozen breaded shrimp was weighed immediately upon removal from the freezer, after which the breaded specimen was removed from its bag, placed in a tared aluminum weighing pan, and weighed to the nearest 1/10 gram. While still in a frozen condition, the breading material was then dislodged from the shrimp with a stainless steel spatula, transferred to another tared weighing pan, and weighed. The weight of the frozen breading material and that of the shrimp were recorded. The pans containing the dislodged breading material, as well as those containing the shrimp meat, were placed in a drying oven at 120°C at atmospheric pressure and dried for 24 hours or until the samples attained constant weight. The pans and contents were then removed from the oven, placed in a desiccator, allowed to cool, and weighed to the nearest 1/10 gram. The dry matter content of

the shrimp and of the breading material were then computed from the gravimetric measurements.

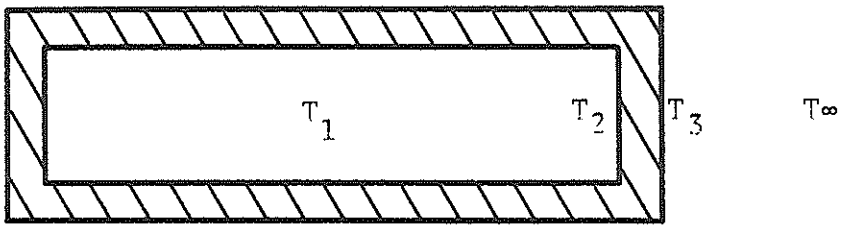
RESULTS AND DISCUSSION

Mathematical Treatment

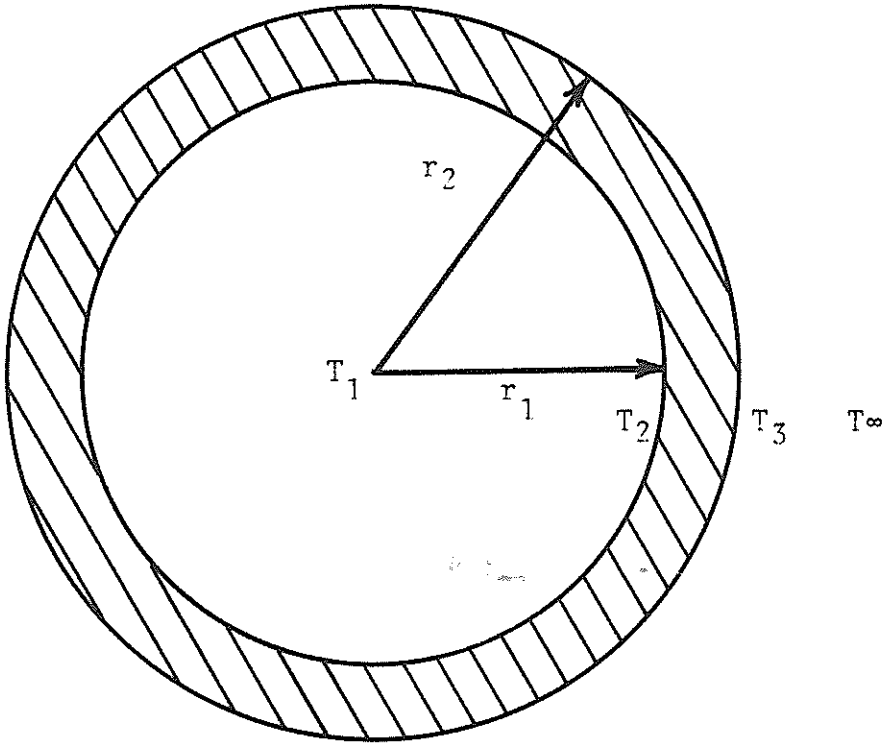
Breaded shrimp are cooled or heated by two mechanisms of heat transfer: convection and conduction. Cooling occurs when the product temperature is above the cold storage temperature and heating occurs when the product temperature is below the cold storage temperature. Convection transfers heat to and from the product's surface and its center. Conduction allows the transfer of heat within the product so as to destroy the temperature difference between the product's surface and its center.

Due to a temperature gradient within the product and a temperature difference between the product's surface and surroundings, various mechanisms of mass transfer occur. The controlling mechanism of mass transfer at a particular point in time depends upon the temperature profile of the product with respect to the temperature of the surroundings. Thus, it is obvious that the temperature profile of the product must be calculated with respect to time.

Certain assumptions must be made in order to facilitate the solution of the heat transfer equations involved. A constant geometric shape must be assumed (Fig. 2). 1) flat-slab, if the product is the butterfly type; 2) cylindrical, if the product is the round-breaded type. End effects and resistance to heat transfer at the shrimp-breading and breading-air interfaces will be



a) Flat Slab



b) Cylindrical

Figure 1. Geometric Shape of Breaded Shrimp

T_1 = Temperature of the product's center
T_2 = Temperature of the shrimp-breading interface
T_3 = Temperature of the breading-air interface
T_∞ = Ambient temperature
r_1 = radius of the shrimp
r_2 = radius of the breaded shrimp

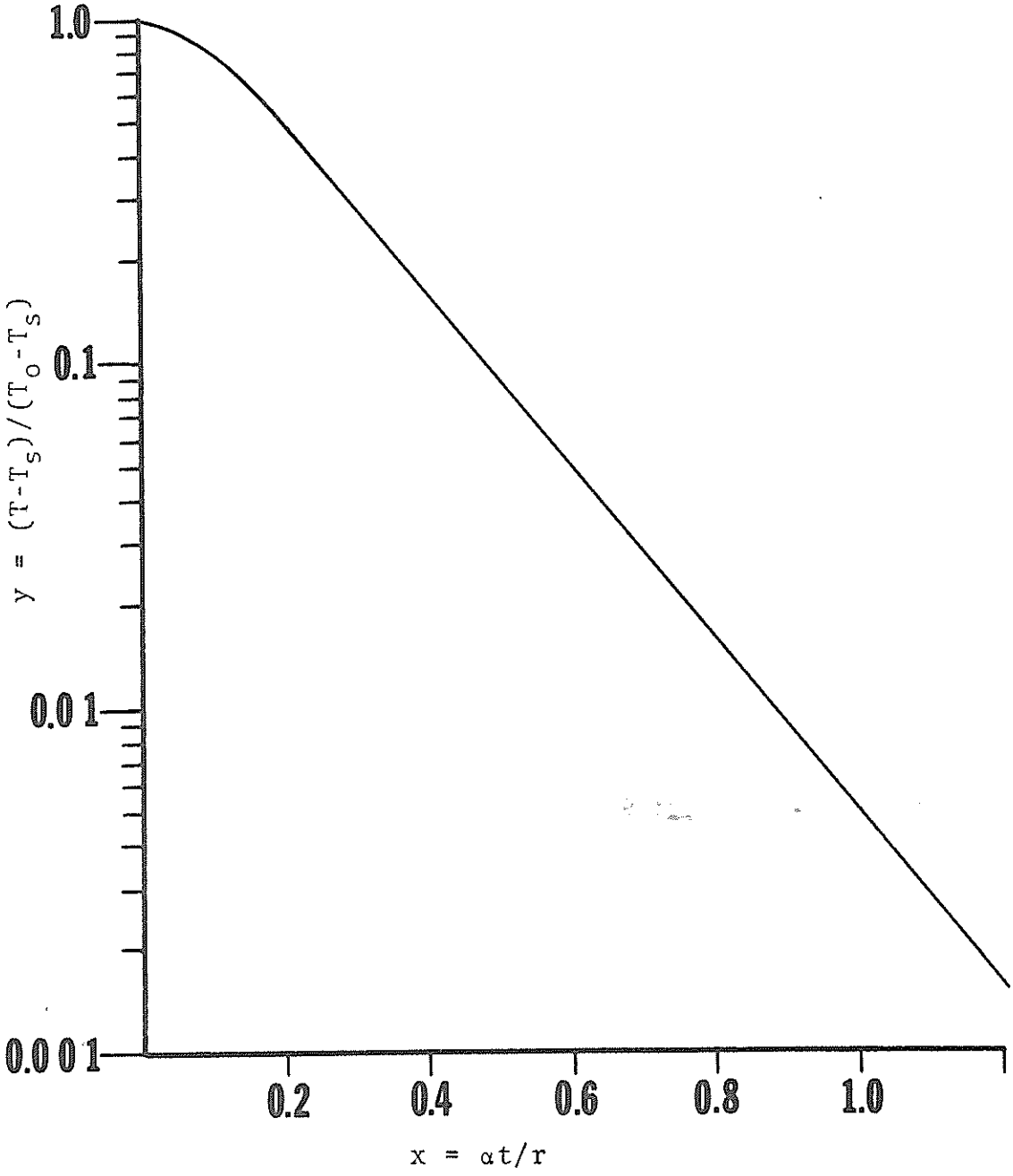


Figure 2. y vs x plot for equation II using cylindrical coordinates

assumed negligible. Round-breaded shrimp with the tails removed were used in this mathematical treatment.

The solution for the temperature history of the shrimp component must satisfy Fourier's Field Equations.

$$\frac{\partial T}{\partial t} = \alpha \nabla^2 T \qquad \text{(I)}$$

where:

T = temperature, ^{o}F
∇ = differential vector operator
t = time, hr.
$\alpha = k/\rho C_p$ = thermal diffusivity, Ft^2/hr
ρ = density, lb/Ft^3
k = thermal conductivity, $BTU/hr \cdot Ft \cdot {}^{o}F$
C_p = heat capacity, $BTU/lb \cdot {}^{o}F$

for cylindrical coordinated, and negligible surface resistance equation I becomes:

$$\frac{\partial T}{\partial t} = \alpha \frac{\partial^2 T}{\partial r^2} + \frac{1}{r}\frac{\partial T}{\partial r} + \frac{1}{r^2}\frac{\partial^2 T}{\partial \Theta^2} + \frac{\partial^2 T}{\partial z^2} \qquad \text{(II)}$$

where:

z = length of the shrimp, Ft
T = temperature, ^{o}F
r = average radius of the shrimp, Ft
Θ = degrees of rotation around the axial length of the product

The solution of equation II, as presented by Welty, Wicks, Wilson ( ), is a plot of y·vs·x, as shown in Figure , where:

$y = (T-T_s)/(T_o-T_s)$
$x = \alpha t/r^s$
T_o = initial uniform temperature, ^{o}F
T_s = surface temperature of the shrimp or temperature of the shrimp-breading interface, ^{o}F
T = center temperature of the shrimp, ^{o}F
α = thermal diffusivity, Ft^2/hr
t = time, hr
r = average radius of the shrimp

A flat slab being cooled or heated from one side (Fig. 3) can be used to simulate the breading layer around the shrimp. The

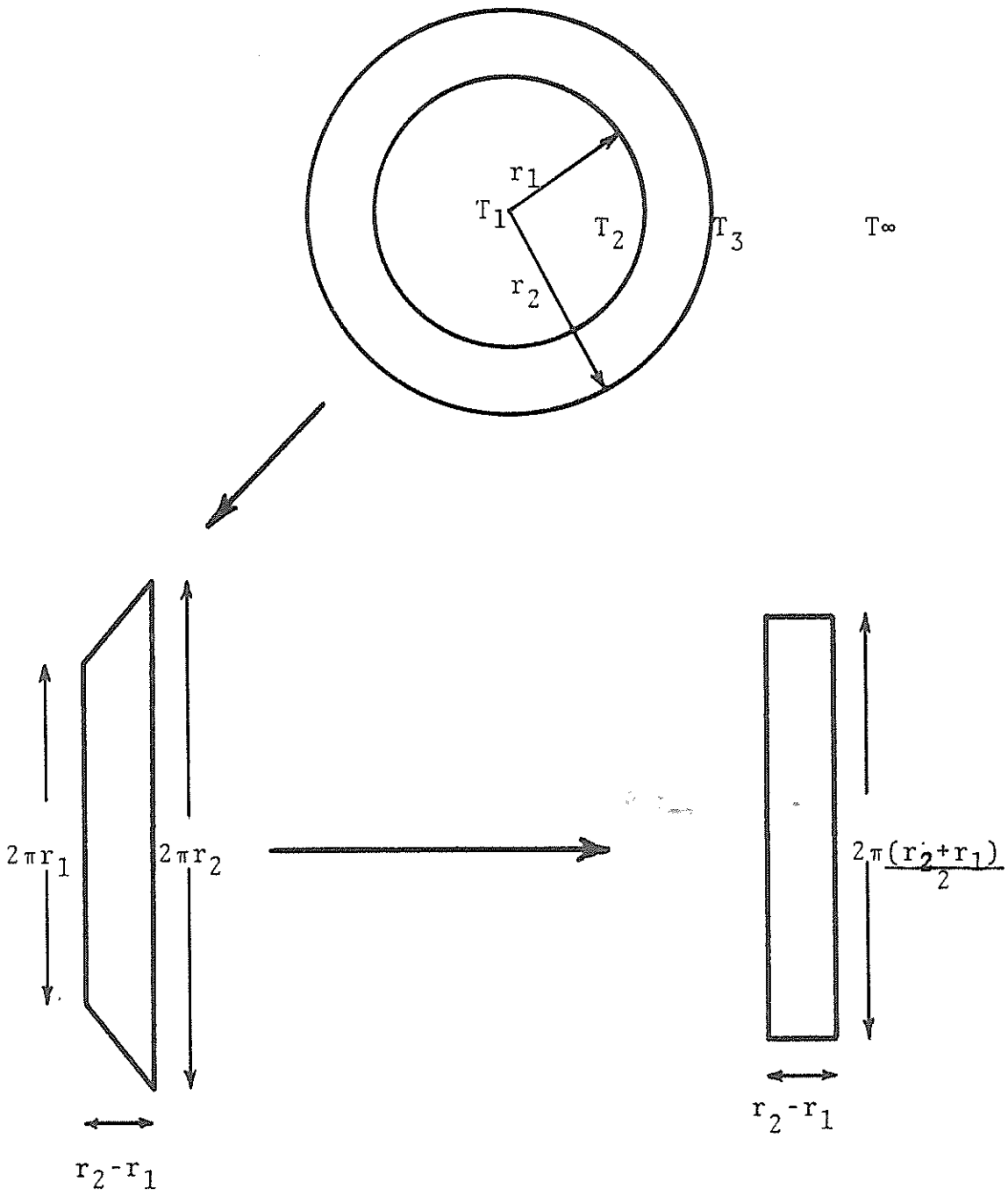


Figure 3. Mathematical Rearrangement of the Breading Layer Shape

solution for the temperature history of the breading must also satisfy Fourier's Field Equation, equation I. Assuming rectangular coordinates and finite surface and internal resistances, equation I becomes:

$$\frac{\partial T}{\partial t} = \alpha \frac{\partial^2 T}{\partial x^2} + \frac{\partial^2 T}{\partial y^2} + \frac{\partial^2 T}{\partial z^2} \qquad \text{(III)}$$

For one-dimensional cooling or heating, equation III becomes:

$$\frac{\partial T}{\partial t} = \alpha \frac{\partial^2 T}{\partial x^2} \qquad \text{(IV)}$$

The solution of equation IV, as presented by Perry's Handbook (8), is a plot of y·vs·x, as shown in Figure 4, where:

$y = (t' - t)/(t' - t)$
$x = k\Theta/\rho C_p r^2 m$
t' = temperature of the surroundings, ^oF
t = temperature at a given point of time, ^oF
t_b = initial temperature, ^oF
k = thermal conductivity, BTU/hr·Ft·^oF
Θ = time, hr
ρ = density, lb/Ft^3
C_p = specific heat, BTU/lb·^oF
r = distance, in the direction of heat conduction from the mid-point or mid-plane of the body to the point under consideration, Ft
$m = k/h_r r_m$
h_r = coefficient of total heat transfer between surroundings on the breading surface, BTU/hr·Ft^2·^oF
r_m = total thickness of a slab being cooled or heated from one face, Ft
$n = r/r_m$

When the physical constants for the shrimp and breading are evaluated, equations II and IV can be solved simultaneously. Pertinent to the calculation of the temperature history is the notation that the ambient temperature, or cold storage temperature, is oscillating above and below a preset temperature. This oscillation of temperature is due to the nature of the temperature

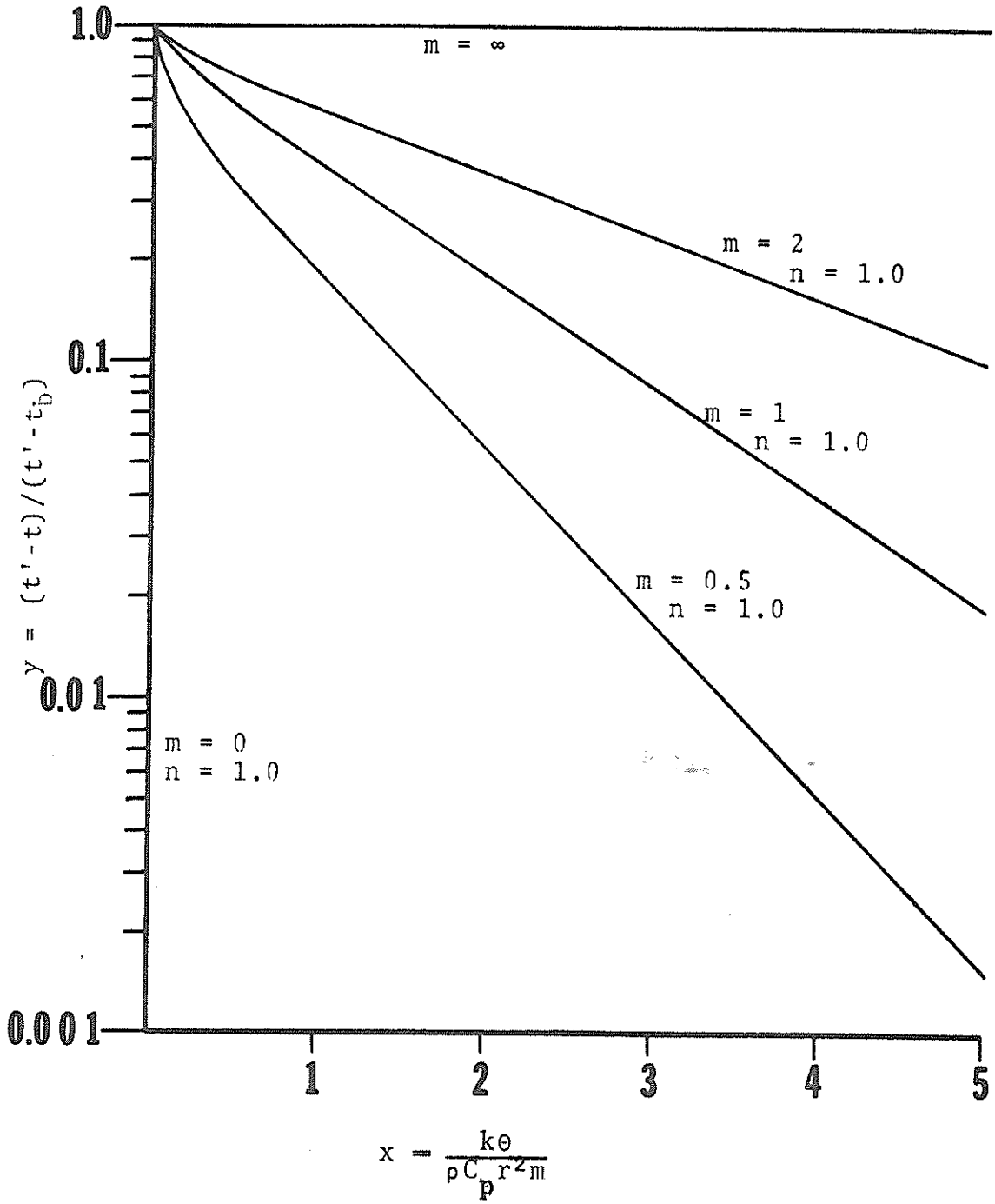


Figure 4.. plot of y·vs·x for the solution of equation IV when heating or cooling a flat slab.

regulating device, ratio of cooling coil surface to the volumn of the cold storage room, and the rate of air circulation in the cold storage room.

As stated previously, the temperature of the shrimp, breading, and the surroundings control the mass transfer mechanisms occurring at a particular point in time. Evaporation, sublimation, and diffusion are the mechanisms of mass transfer involved. Evaporation and sublimation are the means by which the breading loses weight, or moisture, to the surroundings. Diffusion is the mechaism by which the shrimp loses moisture to the breading.

Evaporation is normally associated with the input of heat necessary to boil or vaporize a liquid. This is not so with the case under investigation. A representation of the situation encountered is shown in Fig. 5 which shows that the product is cooled by the loss of sensible heat and latent heat. Latent heat is lost by the evaporation of water from the breading-air interface. The amount of latent heat lost to the air is governed by the rate of evaporation which is dependent upon the difference in the humidities of the breading surface and the surrounding air (Fig. 1). This rate of evaporation (9) can be expressed as:

$$m = M_A k(H_i - H)A \qquad (V)$$

where:

m = rate of evaporation, lb/hr
M_A = molecular weight of air, lb/lb mole
k^A = mass transfer coefficient, lb/moles/ft^2·hr unit mole fraction difference
H_i = humidity of air-breading interface, lb H_2O·lb dry air
H = humidity of air, lb H_2O/lb dry air
A = evaporation area, Ft^{22}

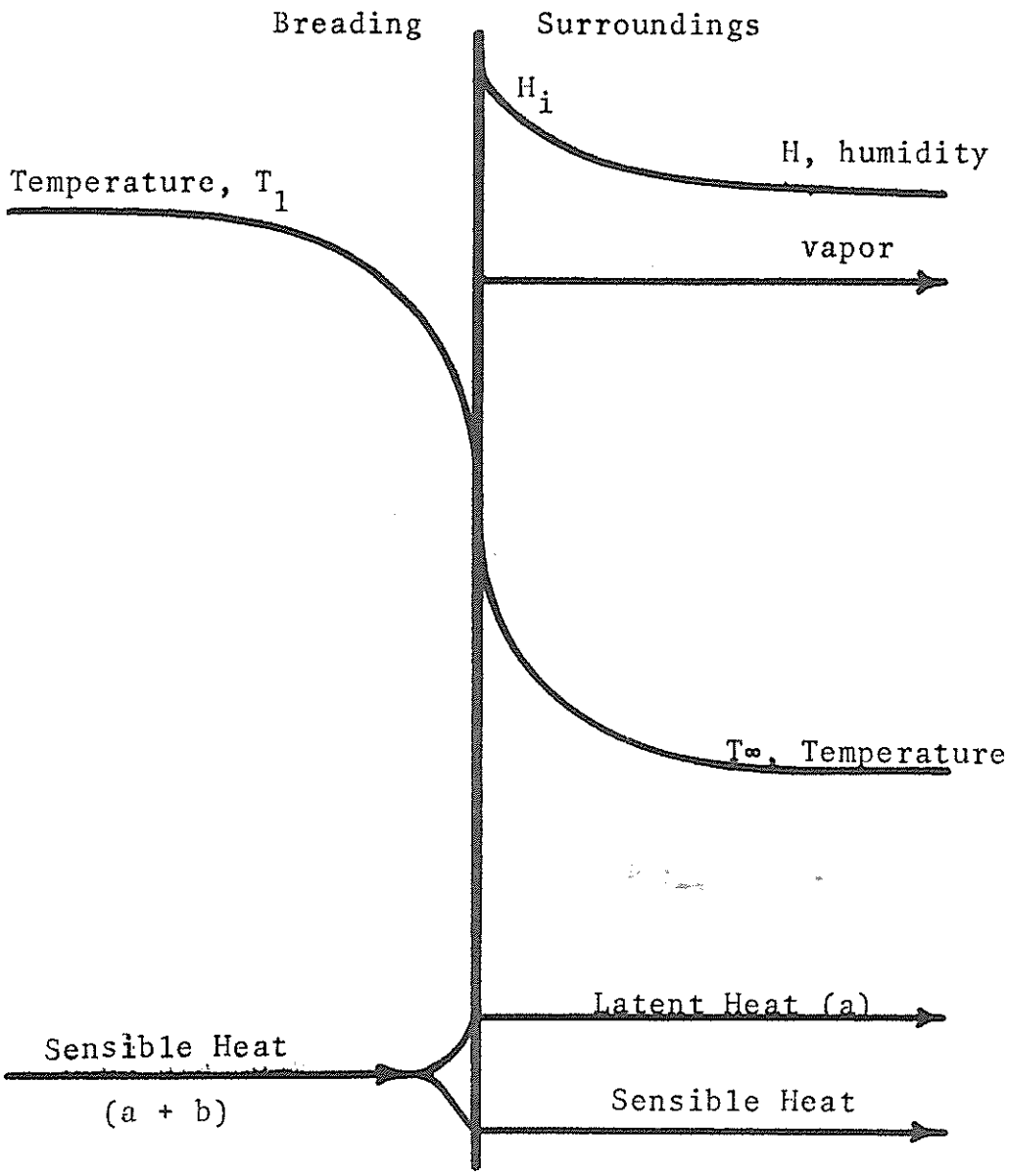


Figure 5. Heat Transfer at the Breading-Surroundings Interface

Evaporation area decreases as the moisture on the breading surface freezes. When the entire breading surface is frozen, the evaporation area is zero and the moisture loss by evaporation ceases.

The temperature of the ice formed on the surface of the breading will now approach the temperature of the cold storage room. During this drop in temperature, the ice will absorb some heat from the product causing small amounts of ice to sublime. There will be periods of time when the ice temperature is lower than the ambient temperature due to the oscillation of the temperature in the cold storage room. It is this situation that allows the surroundings to act as a heat source for the sublimation of ice. The temperature difference between the ice and the surroundings is the driving force for the rate of sublimation, which can be expressed as:

$$\frac{dw}{d\Theta} = \frac{Q}{\lambda_o} = \frac{UA\Delta T}{\lambda_o} \qquad \text{(VI)}$$

where:

$dw/d\Theta$ = rate of sublimation, lb/hr
Q = heat absorbed by the ice, BTU/hr
λ_o = heat sublimation, BTU/lb
U = over-all heat transfer coefficient, BTU/hr·ft^2 ^oF
A = heat transfer area, ft^2
ΔT = temperature difference between the heat source and ice, ^oF

Diffusion of moisture, or moisture migration, is an added problem encountered with breaded shrimp. Shrimp contain a higher percentage of moisture than the breading material and it is this difference in percent moisture that is the driving force for diffusion. Moisture will diffuse from the shrimp to the breading giving the shrimp a negative weight change and the breading a

positive weight change. While the diffusion process is occurring, the breading is losing weight by evaporation and then by sublimation. This loss of moisture by the breading increases the time necessary for the percent moisture of the breading to approach that of the shrimp. Diffusion will continue until the thermal arrest time when the moisture is immobilized by freezing.

The basic equation for the calculation of the diffusion rate can be stated as:

$$\frac{\partial x}{\partial t} = D'_V \frac{\partial^2 x}{\partial z^2} \qquad \text{(VII)}$$

where:

x = average free moisture, lb H_2O/lb dry solid
t = time, hr
D'_V = diffusivity, ft^2/hr
z = distance measured in direction of diffusion, ft

Using cylindrical coordinates, equation VII yields upon integration:

$$\frac{x_T - x^*}{x_{T1} - x^*} = \frac{x}{x_1} = 0.692e^{-5.78\beta} + 0.131e^{-30.5\beta} + 0.0534e^{-74.9\beta} + \ldots \qquad \text{(VIII)}$$

where:

$\beta = D'_V t_T/s$
D'_V = diffusivity of moisture, ft^2/hr
t_V = time, hr
s = radius of the shrimp, ft
x_T = average total moisture content at time t_T
x^* = equilibrium-moisture content
x_{T1} = initial moisture content at start
x = average free moisture content at time t_T
x_1 = initial free moisture content
(units of x are expressed in lb H_2O/lb dry solid)

Solving equation VIII for time t_T, gives:

$$t_T = \frac{s^2}{5.78\ D'_V} \ln \frac{0.692x_1}{x}$$

If the time is plotted against the logarithim of the free-moisture content a straight line should be obtained from which D_v' can be calculated.

Data Analysis

Two hundred breaded shrimp, consisting of 50% by weight shrimp, were produced using 2 freezing rates, 2 textures of breading material, and 2 equilibrium temperatures of the breading material. Freezing rates were denoted as either slow or fast, where the time involved for freezing was about 4 hours at $-6^{o}F$ and 3 to 4 seconds at $-320^{o}F$, respectively. Texture of the breading material was designated as either fine (0.0156 inch in diameter) or coarse (0.0312 inch in diameter). Equilibrium temperature of the breading material, $82^{o}F$ and $40^{o}F$, was the temperature at which the breading material came to thermal equilibrium before being applied to the shrimp.

Four values were derived from the gravimetric measurements for each component (shrimp and breading material) of the 200 individual breaded shrimp used in this study. These values: 1) initial weight, 2) weight after frozen storage, 3) change in weight during frozen storage, and 4) percent change in weight during frozen storage.

The average weight change for the shrimp and breading material and the average percent breading for each group are listed in Table I . It can be seen from Table I that the fast freeze groups had smaller changes in weight for the shrimp and breading material than did the slow freeze groups which are also indicated in the percent breading.

Table I

Average Changes in Weight of the Shrimp and Breading Material and Average Percent Breading

Group No.	Storage Time (wks)	Average Change In Weight (g)		Average Percent Breading
		Shrimp	Breading	
1	1	-0.8	0.5	54.1
	2	-1.1	0.8	55.7
	3	-1.4	0.6	55.8
	4	-0.9	0.5	55.0
	5	-0.8	0.1	52.5
2	1	-1.2	0.6	56.3
	2	-1.2	0.8	56.5
	3	-1.2	0.6	55.5
	4	-1.0	0.6	55.7
	5	-1.1	0.4	54.6
3	1	-1.2	0.7	57.6
	2	-0.9	0.6	55.7
	3	-0.8	0.5	54.4
	4	-0.9	0.5	54.3
	5	-1.0	0.6	54.5
4	1	-0.7	0.4	54.7
	2	-0.8	0.4	53.8
	3	-0.8	0.6	55.1
	4	-1.2	0.8	55.5
	5	-0.8	0.3	53.9

Table I

(continued)

Group No.	Storage Time (wks)	Average Change In Weight (g) Shrimp	Breading	Average Percent Breading
5	1	-0.9	0.0	52.4
	2	-0.6	-0.2	51.1
	3	-0.5	-0.1	51.5
	4	-0.5	-0.1	51.2
	5	-0.3	-0.4	49.9
6	1	-0.8	0.2	53.1
	2	-0.8	0.3	54.0
	3	-0.7	0.0	52.4
	4	-0.6	0.0	52.0
	5	-0.4	-0.1	51.1
7	1	-0.6	0.3	52.6
	2	-0.7	0.4	54.0
	3	-0.5	0.1	52.2
	4	-0.5	0.2	52.3
	5	-0.5	0.1	52.0
8	1	-0.8	0.5	54.1
	2	-0.6	0.1	52.5
	3	-0.5	0.2	53.3
	4	-0.5	0.1	52.0
	5	-0.3	0.1	51.9

Results of an analysis of variance of the data are presented in Table II. The four treatment variables affected the change in weight and the percent change in weight of the shrimp and the breading material as follows:

1) The rate at which the breaded shrimp was frozen had a very significant ($P<0.01$) effect on the amount and the percentage of moisture lost by the shrimp, as well as on the amount and percentage of moisture gained by the breading material.

2) The texture of the breading material very significantly ($P<0.01$) affected the amount of moisture lost by the shrimp, and the amount of moisture gained by the breading material. Also, very significantly ($P<0.01$) affected by the texture of the breading material was the percentage of moisture gained by the breading material, but not significantly ($P>0.05$) affected was the percentage of moisture lost by the shrimp.

3) The equilibrium temperature at which the breading material was applied to the shrimp did not significantly ($P>0.05$) affect either the amount or the percentage of moisture lost by the shrimp to the breading material. However, it did significantly ($P<0.05$) affect both the amount and the percentage gained by the breading material.

4) Differing lengths of time that the breaded shrimp were kept in frozen storage very significantly ($P<0.01$) affected the amount and percentage of moisture lost by the shrimp. Affected, also, were both the amount and percentage of moisture gained by the breading material.

Table II

Analysis of Variance for Four Variables of
50% - 50% Frozen Breaded Shrimp

Source of Variation	D.F.	Mean Squares			
		Weight Loss of Shrimp		Weight Gain of Breading Material	
		Grams	Percent	Grams	Percent
Texture of Breading Material	1	0.7320**	24.10	0.8712**	223.22**
Temperature of Breading Material	1	0.0264	16.96	0.3698*	68.24*
Rate of Freezing	1	8.4460**	1316.01**	10.3968**	1679.32**
Frozen Storage Time	4	0.2963**	60.96**	0.4698**	93.81**
Texture x Temperature	1	0.4140*	102.73**	0.5618**	104.53**
Texture x Freezing Rate	1	0.1300	10.24	0.8192**	124.96**
Texture x Frozen Storage Time	4	0.1954*	14.99	0.1002	15.11
Temperature x Rate of Freezing	1	0.0364	9.82	0.0450	14.10
Temperature x Frozen Storage Time	4	0.0378	2.50	0.0180	2.31
Rate of Freezing x Frozen Storage Time	4	0.1782*	6.02	0.1070	6.63
Texture x Temperature x Rate	1	0.0312	19.94	0.0002	0.47

Table II

(continued)

Source of Variance	D.F.	Mean Squares			
		Weight Loss of Shrimp		Weight Gain of Breading Material	
		Grams	Percent	Grams	Percent
Texture x Temperature x Time	4	0.0884	18.11	0.1486	34.26*
Texture x Rate x Time	4	0.1402	13.80	0.0624	5.45
Temperature x Rate x Time	4	0.0506	13.13	0.0725	7.99
Texture x Temperature x Rate x Time	4	0.2909**	33.54*	0.1242	15.92
Residual Variance (Error Term)	160	0.0639	9.90	0.0806	13.35

**p 0.01

*p 0.05

Main Effects of the Four Treatment Variables

The mean amounts and the corresponding percentage values for the change in weight of the shrimp and the breading material during frozen storage are given in Table III. These amounts and values are attributable to the effects of the four treatmeat variables.

Rate of Freezing

Shrimp frozen slowly (4 hours at -6°F) lost much more moisture during frozen storage than shrimp that had been frozen rapidly (3 to 4 seconds at -320°F). The mean loss in weight associated with the slow freezing rate was 1.00 g, or 12.70%; the loss associated with the fast freezing rate was 0.59 g, or 7.57%. The difference between the two means, 0.41 g, was very highly significant ($P<0.01$) as indicated by a calculated F value of 132.23. This F value was almost identical with the calculated F value, 132.95, for the difference between the two mean percentage values, 5.13.

The mean weight gain of the breading material associated with the slow rate of freezing, 0.55 g, was six-fold greater than that associated with the fast rate of freezing 0.09 g. The difference between the two means, 0.46 g, was highly significant ($P<0.01$) as indicated by a calculated F value of 128.95. This value was similar to the calculated F value of 125.78 for the difference, 5.80, between the two corresponding mean percentage values.

It was noted that the shrimp of the fast freeze samples lost less moisture to the breading material than the shrimp of the slow freeze samples. Similarly, the breading material of the fast

Table III

Mean Values for Changes in Weight During Frozen Storage of Shrimp and Breading Material

Variable	Weight Loss of Shrimp		Weight Gains of Breading Material	
	Grams	Percent	Grams	Percent
Texture of Breading Material*				
Fine	0.86	10.49	0.25	3.09
Coarse	0.74	9.79	0.38	5.21
Temperature of Breading Material				
40°F	0.81	10.43	0.36	4.74
82°F	0.78	9.85	0.28	3.57
Rate of Freezing**				
Slow	1.00	12.70	0.55	7.05
Fast	0.59	7.57	0.09	1.25
Length of Frozen Storage Time (wks)				
1	0.88	11.29	0.41	5.62
2	0.86	11.10	0.40	5.19
3	0.81	10.42	0.32	4.29
4	0.78	9.62	0.32	3.97
5	0.66	8.27	0.14	1.69
Overall Mean	0.80	10.14	0.32	4.15

*Fine = 0.0156 inch particle size
Coarse = 0.0312 inch particle size

**Slow = 4 hrs; Fast = 3 - 4 sec

freeze samples gained less moisture than the breading material of the slow freeze samples. This can be explained by the fact that the moisture in the breaded shrimp is immobilized sooner in the fast freeze samples than in the slow freeze samples. Thus, in the slow freeze samples, water can diffuse from the shrimp to the breading material for a longer period of time resulting in a higher change in weight of the shrimp and breading material. The breading material of the slow freeze samples lost moisture to the surroundings by evaporation before the surface was frozen. This evaporational loss increased the diffusional driving force between the shrimp and breading material. It allowed the shrimp to lose more moisture to the breading material in comparison with the shrimp of the fast freeze samples. Once the breading material was frozen, evaporational losses ceased and sublimation then allowed the breading material to lose moisture to the surroundings. Moisture lost by the breading material via evaporation and sublimation accounted for the breading material not gaining the same amount of moisture lost by the shrimp. From the above it can be concluded that since the slow freeze samples are exposed a longer period of time, more moisture can be lost to the surroundings and transferred within the samples.

Texture of the Breading Material

The mean loss in weight of the shrimp coated with fine-textured (0.0156" diameter) breading was 0.86 g, or 10.49%; the loss associated with the shrimp coated with coarse-textured (0.0312" diameter) breading was 0.74 g, or 9.79%. The difference between the two means, 0.15 g, was highly significant ($P<0.01$) as

indicated by a calculated F value of 10.81. The critical F values at D.F. 1/160 were $F_{0.05}$ = 6.81. These findings suggest that there is an inverse relationship between the surface area per unit weight of the breading material and its moisture absorption capacity, or conversely, the larger the particle size of the breading material, the greater is its capability of retaining moisture from the shrimp meat. The difference between the corresponding mean percentage values for weight gain of the breading material (3.09% for the fine-textured and 5.21% for the coarse-textured was 2.14. This is highly significant as indicated by a calculated F value of 16.72. This finding of a highly significant difference between the effects of the two textures on the mean percentage values for weight gain of the breading material is in sharp contrast to the finding that the mean percentage values for weight loss of the shrimp were not significantly affected by the texture of the breading material.

Being of smaller particle size, the fine-textured breading material will exert a larger capillary attraction on the moisture in the shrimp than the coarse-textured breading material. This increased capillary attraction exerted by the fine textured breading material increases the diffusional rate, as compared to that of the coarse textured, thus causing the fine textured breading to extract more moisture out of the shrimp. The percent change in weight of the shrimp was not significantly affected by the texture of the breading material which indicates that the percent weight loss of the shrimp is approximately the same for the fine or coarse textured breading material. Once the moisture migrated

into the breading material, the fine textured breading material, having a larger surface area, was able to lose moisture to the surroundings by evaporation and sublimation at a faster rate than the coarse textured breading material. From the above it can be seen why the percent weight gain of the breading material is very significantly affected by the texture of the breading material.

Equilibrium Temperature of the Breading Material

The temperature at which the breading material was applied to the shrimp did not significantly ($P>0.05$) affect the amount of moisture lost by the shrimp. The mean loss in weight of shrimp of the shrimp coated with breading material at $40^{o}F$ was 0.81 g, or 10.43%, whereas shrimp coated at $82^{o}F$ lost 0.78 g, or 9.85%. The difference between the mean values, 0.03 g, was not significant, as indicated by an F value of 0.41; the difference between the corresponding percentage values, 0.58, likewise was not significant.

The mean gain in weight of the dislodged breading material that had been applied at $40^{o}F$ was significantly greater ($P<0.05$) than that of the coating applied at $82^{o}F$, where the mean gravimetric values are 0.36 g and 0.28 g, respectively. The calculated F value was 4.59. The difference between the two corresponding percentage values for gain in weight, 4.74% and 3.57%, or 1.17, was also significant ($P<0.05$).

Even though the breading material at the two different equilibrium temperatures extracted approximately the same amount of moisture from the shrimp, the breading material applied at the lower equilibrium temperature had a larger moisture, or weight,

gain. It should be noted that the breading material associated with an equilibrium temperature of 82°F will take longer to cool down and finally freeze, than if it were applied at 40°F. This extended time required to freeze the breading material applied at a higher equilibrium temperature will allow more evaporational loss since the rate of evaporation is faster than the rate of sublimation.

Storage Time

Increasing the length of elapsed time from one to five weeks, during which the breaded shrimp samples were stored at -6°F, resulted in a gradual decline in the amount of moisture lost from the shrimp. The five periods of time during which the breaded samples were kept in frozen storage, 1, 2, 3, 4, and 5 weeks, were associated with the following mean values of weight loss: 0.88 g (11.29%), 0.86 g (11.10%), 0.81 g (10.42%), 0.78 g (9.62%), and 0.66 g (8.27%), respectively. The calculated F values, 4.64 for the mean amounts and 6.16 for the mean percentages, were highly significant ($P<0.01$). The critical F values at D.F. 4/160 were $F_{0.05} = 2.43$ and $F_{0.01} = 3.44$.

A gradual decline in the mean gain of moisture by the breading material, roughly paralleling the decline in the mean losses in weight noted for the shrimp, occurred as the length of time of frozen storage of the breaded shrimp samples increased from one to five weeks. The five periods of time, 1, 2, 3, 4, and 5 weeks, were associated with the following mean values of weight gain of the breading material: 0.41 g (5.62%), 0.40 g (5.19%), 0.32 g (4.29%), 0.32 g (3.97%), and 0.14 g (1.69%), respectively. The

calculated F values, 5.83 for the mean amounts and 7.03 for the mean percentages, were highly significant (P 0.01). The critical F values at D.F. 4/160 were $F_{0.05} = 2.43$ and $F_{0.01} = 3.44$.

The Bivariate Interactions

Freezing Rate x Storage Time

Shrimp that were frozen slowly and stored at -6°F over periods of 1 to 5 weeks lost more moisture than shrimp that were frozen rapidly and stored at -6°F. Mean losses in weight for the shrimp associated with freezing rate and storage times are given in Table IV. The calculated F value of 2.79 for the mean values was significant ($P<0.05$), but the F value of 0.609 for the mean percentages was not significant ($P>0.05$). The critical F values at D.F. 4/160 were $F_{0.05} = 2.43$ and $F_{0.01} = 3.44$. After an initial weight loss for the shrimp of both freezing rates, the shrimp of the rapidly frozen group regained approximately half of the moisture lost but the shrimp of the slowly frozen group remained at approximately the same level of moisture loss. The mean percentage values indicated that the shrimp of both freezing rates began to regain moisture after the initial moisture loss.

Mean gains in weight for the breading material associated with freezing rate and storage times are given in Table IV. The F values for the mean values and mean percentages were 1.33 and 0.50, respectively, and were not significant. After an initial gain in weight the breading material then began to lose moisture. This behavior was also shown in the percentage values.

Rate of Freezing x Texture of the Breading Material

Mean values of moisture lost by the shrimp are given in

Table IV

Effect of Freezing Rate and Storage Time on the Mean Values for Changes in Weight of the Shrimp and Breading Material

Freezing Rate*	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
		Shrimp	Breading	Shrimp	Breading
Slow	1	-0.975	0.555	-13.88	8.14
	2	-1.045	0.655	-12.02	8.19
	3	-1.045	0.575	-13.02	7.37
	4	-1.020	0.600	-12.59	7.40
	5	-0.925	0.345	-11.00	4.15
Fast	1	-0.780	0.260	- 8.71	3.11
	2	-0.670	0.140	- 9.17	2.20
	3	-0.580	0.075	- 7.82	1.20
	4	-0.530	0.045	- 6.64	0.54
	5	-0.395	-0.070	- 5.54	-0.78
F-value		2.79	1.33	0.609	0.496

$F_{0.05} = 2.43$
$F_{0.01} = 3.44$

*Slow = 4 hr; Fast = 3-4 sec

Table V . The calculated F values for the mean values and mean percentages are 2.04 and 1.03, respectively, and are not significant (P>0.05). The critical F values at D.F 1/160 were $F_{0.05}$ = 3.91 and $F_{0.01}$ = 6.81. It was noted that the shrimp lost approximately the same amount of moisture for each of the 2 breading textures. This observation is also evidenced by the percentage values.

The mean values of weight gain for the breading material (Table V) were: 0.544 g (6.78%) for the slow freezing rate and fine texture, 0.548 g (7.31%) for the slow freezing rate and coarse texture, -0.040 g (-0.59%) for the fast freezing rate and fine texture and 0.220 g (3.10%) for the fast freezing rate and coarse texture. Calculated F values for the mean values and mean percentages were 10.16 and 9.36, respectively, and highly significant (P<0.01). It was noted that for the slow freezing rate both the fine and coarse breading material textures gained approximately the same amount of moisture. For the fast freezing rate the fine texture breading material lost moisture, rather than gained moisture, but the coarse breading material gained moisture.

Equilibrium Temperature x Texture of the Breading Material

Mean values of weight loss for the shrimp (Table VI) were: 0.80 g (9.48%) for 82°F equilibrium temperature and fine texture, 0.914 g (11.49%) for 40°F equilibrium temperature and fine texture, 0.770 g (10.22%) for 82°F equilibrium temperature and coarse texture, and 0.702 g (9.36%) for 40°F equilibrium temperature and coarse texture. The calculated F value for the mean values was 6.48 and significant (P<0.05) and calculated F value for the mean

Table V

Effect of Freezing Rate and Texture of the Breading Material on the Mean Values for Changes in Weight of the Shrimp and Breading Material

Freezing Rate *	Texture of the Breading Material**	Mean Weight Change (g)		Mean % Weight Change	
		Shrimp	Breading	Shrimp	Breading
Slow	Fine	-1.088	0.544	-13.28	6.78
	Coarse	-0.916	0.548	-12.13	7.31
Fast	Fine	-0.626	-0.040	- 7.69	-0.59
	Coarse	-0.556	0.220	- 7.45	3.10
F-value		2.04	10.16	1.03	9.36

$F_{0.05} = 3.91$
$F_{0.01} = 6.81$

*Slow = 4 hr; Fast = 3-4 sec

**Fine = 0.0156 inch particle size
Coarse = 0.0312 inch particle size

Table VI

Effect of Texture and Equilibrium Temperature of the Breading Material on the Mean Values for Changes in Weight of the Shrimp and Breading Material

Equilibrium Temperature of the Breading Material	Texture of the Breading Material*	Mean Weight Change (g)		Mean % Weight Change	
		Shrimp	Breading	Shrimp	Breading
82°F	Fine	-0.800	0.156	- 9.48	1.79
	Coarse	-0.770	0.394	-10.22	5.35
40°F	Fine	-0.914	0.348	-11.49	4.40
	Coarse	-0.702	0.374	- 9.36	5.07
F-value		6.48	6.97	10.38	7.83

$F_{0.05} = 3.91$
$F_{0.01} = 6.81$

*Fine = 0.0156 inch particle size
Coarse = 0.0312 inch particle size

percentages was 10.38 and highly significant ($P<0.01$). When the equilibrium temperature of the breading material was decreased from 82°F to 40°F, the loss in moisture of the shrimp coated with the fine texture increased; the loss in moisture slightly decreased when the coarse texture was used. This observation was very noticeable in the percentage values.

The mean values of weight gain for the breading material (Table VI) were: 0.156 g (1.79%) for 82°F equilibrium temperature and fine texture, 0.348 g (4.40%) for 40°F equilibrium temperature and fine texture, 0.394 g (5.35%) for 82°F equilibrium temperature and coarse texture, and 0.374 g (5.07%) for 40°F equilibrium temperature and coarse texture. Calculated F values for the mean values and mean percentages were 6.97 and 7.83, respectively, and were both highly significant ($P<0.01$). When the equilibrium temperature of the breading material was decreased from 82°F to 40°F, the gain in moisture of the fine textured breading material increased; and in regards to the coarse textured breading material, the gain in moisture remained approximately the same.

Storage Time x Texture of the Breading Material

Mean losses in weight for the shrimp associated with the storage times and texture of the breading material are given in Table VII . The F value calculated for the mean values was 3.06 and significant ($P<0.05$), but the calculated F value of 1.13 for the mean percentages was not significant. When the fine textured breading material was used, the shrimp remained approximately at the initial moisture loss level and during the 3rd. week began to regain some of the lost moisture. The shrimp coated with the

Table VII

Effect of Storage Time and Texture of the Breading Material on the Mean Values for Changes in Weight of the Shrimp and Breading Material

Texture of the Breading Material*	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
		Shrimp	Breading	Shrimp	Breading
Fine	1	-0.920	0.320	-10.97	4.39
	2	-0.970	0.400	-12.02	4.80
	3	-0.965	0.280	-11.39	3.21
	4	-0.765	0.255	-10.00	3.36
	5	-0.665	0.005	- 8.05	0.28
Coarse	1	-0.835	0.495	-11.62	6.86
	2	-0.745	0.395	-10.17	5.59
	3	-0.660	0.370	- 9.44	5.37
	4	-0.785	0.390	- 9.23	4.57
	5	-0.655	0.270	- 8.49	3.66
F-value		3.06	1.24	1.13	1.51

$F_{0.05} = 2.43$
$F_{0.01} = 3.44$

*Fine = 0.0156 inch particle size
Coarse = 0.0312 inch particle size

coarse textured breading material immediately began to regain the initially lost moisture. However, from the 3rd. and 4th. weeks there was another loss in moisture followed by the regaining of the moisture lost between the 3rd. and 4th weeks. The mean percentage values showed a similar behavior; after the initial moisture loss, the shrimp coated with either the fine or coarse breading material began to regain moisture. The critical F values at D.F. 4/160 were $F_{0.05} = 2.43$ and $F_{0.01} = 3.44$.

The mean gains in weight for the breading material are given in Table VII . Calculated F values for the mean values and mean percentages were 1.24 and 1.51, respectively, and were not significant ($P>0.05$). Both textures of breading material initially gained moisture and then began to lose moisture.

Storage Time x Equilibrium Temperature of the Breading Material

The mean weight changes for the shrimp and breading material are given in Table VIII. Calculated F values for the mean values and mean percentages for both the shrimp and breading material were not significant ($P>0.05$).

Freezing Rate x Equilibrium Temperature of the Breading Material

The mean weight changes for the shrimp and breading material are given in Table IX . Calculated F values for the mean values and mean percentages for both the shrimp and breading material were not significant ($P>0.05$).

The Trivariate Interactions

Storage Time x Texture of the Breading Material x Equilibrium Temperature of the Breading Material

The mean gravimetric values and mean percent values for weight

Table VIII

Effect of Storage Time and Equilibrium Temperature of the Breading Material on the Mean Values for Changes in Weight of the Shrimp and Breading Material

Equilibrium Temperature of the Breading Material	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
		Shrimp	Breading	Shrimp	Breading
82°F	1	-0.860	0.360	-10.95	5.18
	2	-0.880	0.385	-11.25	4.96
	3	-0.815	0.270	-10.00	3.49
	4	-0.715	0.255	- 9.20	3.22
	5	-0.655	0.105	- 7.83	0.98
40°F	1	-0.895	0.455	-11.63	6.06
	2	-0.835	0.410	-10.94	5.42
	3	-0.810	0.380	-10.83	5.08
	4	-0.835	0.390	-10.03	4.71
	5	-0.665	0.170	- 8.71	2.40
F-value		0.592	0.224	0.253	0.176

$F_{0.05} = 2.43$
$F_{0.01} = 3.44$

Table IX

Effect of Freezing Rate and Equilibrium Temperature of the Breading Material on the Mean Values for Changes in Weight of the Shrimp and Breading Material

Freezing Rate*	Equilibrium Temperature of the Breading Material	Mean Weight Change (g)		Mean % Weight Change	
		Shrimp	Breading	Shrimp	Breading
Slow	82°F	-1.004	0.518	-12.63	6.73
	40°F	-1.000	0.574	-12.77	7.37
Fast	82°F	-0.566	0.032	- 7.06	0.40
	40°F	-0.616	0.148	- 8.08	2.10
F-value		0.571	0.558	0.990	1.056

$F_{0.05} = 3.91$
$F_{0.01} = 6.81$

*Slow = 4 hr; Fast = 3-4 sec

change for the shrimp and breading material are given in Table X . Calculated F values for the mean changes in weight and mean percent values for both the shrimp and breading material were not significant ($P>0.05$) with the exception of the mean percent weight gain of the breading material, for which the calculated F value was 2.56 and was significant ($P<0.05$). The critical F values at D.F. 4/160 were $F_{0.05} = 2.43$ and $F_{0.01} = 3.44$.

Storage Time x Freezing Rate x Equilibrium Temperature of the Breading Material

The mean values and mean percent values for weight change for the shrimp and breading material are given in Table XI . All calculated F values for the mean and mean percent values for both the shrimp and breading material were not significant ($P>0.05$).

Storage Time x Freezing Rate x Texture of the Breading Material

The mean values and mean percent values for weight change for the shrimp and breading material are given in Table XII . The calculated F values for the mean and mean percent values for both the shrimp and breading material were not significant ($P>0.05$).

Freezing Rate x Equilibrium Temperature of the Breading Material x Texture of the Breading Material

The mean values and mean percent values for weight change for the shrimp and breading material are given in Table XIII. The calculated F values for the mean and mean percent values for both shrimp and breading material were not significant ($P>0.05$).

The Quadravariate Interaction

The mean values of weight loss for the shrimp are given in Table XIV. The calculated F value for the mean weight loss of the shrimp was 4.55 and was highly significant ($P<0.01$), and the

Table X

Effect of Equilibrium Temperature and Texture of the Breading Material and Storage Time on the Mean Values for Changes in Weight of the Shrimp and Breading Material

Equilibrium Temperature of the Breading Material	Texture of the Breading Material*	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
			Shrimp	Breading	Shrimp	Breading
82°F	Fine	1	-0.83	0.21	- 9.40	3.12
		2	-0.93	0.27	-10.66	2.61
		3	-0.97	0.25	-10.95	2.94
		4	-0.71	0.18	- 9.53	2.33
		5	-0.56	-0.13	- 6.84	-2.06
	Coarse	1	-0.89	0.51	-12.50	7.24
		2	-0.83	0.50	-11.84	7.32
		3	-0.66	0.29	- 9.05	4.04
		4	-0.72	0.33	- 8.88	4.11
		5	-0.75	0.34	- 8.82	4.02

Table X

(continued)

Equilibrium Temperature of the Breading Material	Texture of the Breading Material*	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
			Shrimp	Breading	Shrimp	Breading
40°F	Fine	1	-1.01	0.43	-12.53	5.65
		2	-1.01	0.53	-13.38	6.99
		3	-0.96	0.31	-11.34	3.47
		4	-0.32	0.33	-10.46	4.49
		5	-0.77	0.11	- 9.26	1.59
	Coarse	1	-0.78	0.48	-10.73	6.48
		2	-0.66	0.29	- 8.51	3.96
		3	-0.66	0.45	- 9.83	6.69
		4	-0.85	0.45	- 9.60	5.93
		5	-0.56	0.29	- 8.16	3.29
F-value			1.38	1.84	1.83	2.56

$F_{0.05} = 2.43$
$F_{0.01} = 3.44$

*Fine = 0.0156 inch particle size; Coarse = 0.0312 inch particle size

Table XI

Effect of Freezing Rate, Storage Time, and Equilibrium Temperature of the Breading Material on the Mean Values for Changes in Weight of the Shrimp and Breading Material

Equilibrium Temperature of the Breading Material	Freezing Rate*	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
			Shrimp	Breading	Shrimp	Breading
82°F	Slow	1	-1.00	0.57	-14.24	8.38
		2	-1.11	0.69	-13.62	3.64
		3	-1.08	0.51	-13.35	6.64
		4	-0.93	0.49	-11.84	6.28
		5	-0.90	0.33	-10.11	3.70
	Fast	1	-0.72	0.15	- 7.67	1.98
		2	-0.65	0.08	- 6.87	1.28
		3	-0.55	0.03	- 6.65	0.34
		4	-0.50	0.02	- 6.56	0.16
		5	-0.41	-0.12	- 5.55	-1.75

Table XI

(Continued)

Equilibrium Temperature of the Breading Material	Freezing Rate*	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
			Shrimp	Breading	Shrimp	Breading
40°F	Slow	1	-0.95	0.54	-13.52	7.89
		2	-0.98	0.62	-12.42	7.73
		3	-1.01	0.64	-12.69	8.10
		4	-1.11	0.71	-13.34	8.51
		5	-0.95	0.36	-11.39	4.60
	Fast	1	-0.84	0.37	- 9.74	4.34
		2	-0.69	0.20	- 9.46	3.12
		3	-0.61	0.12	- 8.93	2.06
		4	-0.56	0.07	- 6.72	0.91
		5	-0.38	-0.02	- 5.53	0.19
F-value			0.792	0.892	1.33	0.599

$F_{0.05} = 2.43$
$F_{0.01} = 3.44$

*Slow = 4 hr; Fast = 3-4 sec

Table XII

Effect of Freezing Rate, Storage Time, and Texture of the Breading Material on the Mean Values for Changes in Weight of the Shrimp and Breading Material

Freezing Rate*	Texture of the Breading Material**	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
			Shrimp	Breading	Shrimp	Breading
Slow	Fine	1	0.08	0.52	-12.88	7.30
		2	-1.24	0.79	-14.55	9.19
		3	-1.29	0.59	-14.87	6.95
		4	-0.99	0.57	-13.17	7.58
		5	-0.94	0.25	-10.91	2.90
	Coarse	1	-0.97	0.59	-14.88	8.97
		2	-0.85	0.52	-11.50	7.18
		3	-0.80	0.56	-11.17	7.80
		4	-1.05	0.63	-12.01	7.21
		5	-0.91	0.44	-11.09	5.41

Table XII

(continued)

Freezing Rate*	Texture of the Breading Material**	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
			Shrimp	Breading	Shrimp	Breading
Fast	Fine	1	-0.86	0.12	-9.06	1.47
		2	-0.70	0.01	-9.48	0.41
		3	-0.64	-0.03	-7.92	-0.53
		4	-0.54	-0.06	-6.82	-0.85
		5	-0.39	-0.24	-5.19	-3.46
	Coarse	1	-0.70	0.40	-8.36	4.75
		2	-0.64	0.27	-8.85	4.00
		3	-0.52	0.18	-7.71	2.93
		4	-0.52	0.15	-6.46	1.92
		5	-0.40	0.10	-5.88	1.00
F-value			2.19	0.774	1.394	0.408

$F_{0.05}$ = 2.43
$F_{0.01}$ = 3.44

*Slow = 4 hr; Fast = 3-4 sec

**Fine = 0.0156 inch particle size
Coarse = 0.0312 inch particle size

Table XIII

Effect of Freezing Rate, Texture, and Equilibrium Temperature of the Breading Material on the Mean Values for Changes in Weight of the Shrimp and Breading Material

Freezing Rate*	Equilibrium Temperature of the Breading Material	Texture of the Breading Material**	Mean Weight Change (g)		Mean % Weight Change	
			Shrimp	Breading	Shrimp	Breading
Slow	82°F	Fine	-1.032	0.464	-12.17	5.69
		Coarse	-0.976	0.572	-13.09	7.77
	40°F	Fine	-1.144	0.624	-14.38	7.87
		Coarse	-0.856	0.524	-11.17	6.86

Table XIII

(continued)

Freezing Rate*	Equilibrium Temperature of the Breading Material	Texture of the Breading Material**	Mean Weight Change (g)		Mean % Weight Change	
			Shrimp	Breading	Shrimp	Breading
Fast	82°F	Fine	-0.568	-0.152	-6.78	-2.12
		Coarse	-0.564	0.216	-7.34	2.92
	40°F	Fine	-0.684	0.072	-8.61	0.93
		Coarse	-0.548	0.224	-7.56	3.28
F-value			0.489	0.002	2.014	0.035

$F_{0.05} = 3.91$
$F_{0.01} = 6.81$

*Slow = 4 hr; Fast = 3-4 sec
**Fine = 0.0156 inch particle size
Coarse = 0.0312 inch particle size

Table XIV

Effect of Equilibrium Temperature and Texture of the Breading Material, Freezing Rate, and Storage Time on the Mean Values for Changes in Weight of the Shrimp and Breading Material

Equilibrium Temperature of the Breading Material	Freezing Rate*	Texture of the Breading Material**	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
				Shrimp	Breading	Shrimp	Breading
82°F	Slow	Fine	1	-0.78	0.40	-10.17	5.76
			2	-1.28	0.78	-13.74	8.37
			3	-1.38	0.56	-15.49	6.71
			4	-0.94	0.48	-12.70	6.52
			5	-0.78	0.10	- 8.77	1.10
		Coarse	1	-1.22	0.74	-18.30	11.00
			2	-0.94	0.60	-13.51	8.91
			3	-0.78	0.46	-11.21	6.58
			4	-0.92	0.50	-10.99	6.04
			5	-1.02	0.56	-11.45	6.30

Table XIV

(continued)

Equilibrium Temperature of the Breading Material	Freezing Rate*	Texture of the Breading Material**	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
				Shrimp	Breading	Shrimp	Breading
82°F	Fast	Fine	1	-0.88	0.02	- 8.64	0.49
			2	-0.58	-0.24	- 7.58	-3.16
			3	-0.56	-0.06	- 6.41	-0.82
			4	-0.48	-0.12	- 6.36	-1.86
			5	-0.34	-0.36	- 4.91	-5.23
		Coarse	1	-0.56	0.28	- 6.70	3.47
			2	-0.72	0.40	-10.16	5.73
			3	-0.54	0.12	- 6.89	1.51
			4	-0.52	0.16	- 6.76	2.18
			5	-0.48	0.12	- 6.18	1.74

Table XIV

(continued)

Equilibrium Temperature of the Breading Material	Freezing Rate*	Texture of the Breading Material**	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
				Shrimp	Breading	Shrimp	Breading
40°F	Slow	Fine	1	-1.18	0.64	-15.60	8.85
			2	-1.20	0.80	-15.36	10.01
			3	-1.20	0.62	-14.25	7.18
			4	-1.04	0.66	-13.64	8.64
			5	-1.10	0.40	-13.04	4.69
		Coarse	1	-0.72	0.44	-11.45	6.94
			2	-0.76	0.44	- 9.49	5.45
			3	-0.82	0.66	-11.13	9.02
			4	-1.18	0.76	-13.04	8.39
			5	-0.80	0.32	-10.73	4.52

Table XIV

(continued)

Equilibrium Temperature of the Breading Material	Freezing Rate*	Texture of the Breading Material**	Storage Time (wks)	Mean Weight Change (g)		Mean % Weight Change	
				Shrimp	Breading	Shrimp	Breading
40°F	Fast	Fine	1	-0.84	0.22	- 9.47	2.45
			2	-0.82	0.26	-11.39	3.98
			3	-0.72	0.00	- 9.42	-0.24
			4	-0.60	0.00	- 7.28	0.16
			5	-0.44	-0.12	- 5.47	-1.69
		Coarse	1	-0.84	0.52	-10.01	6.02
			2	-0.56	0.14	- 7.54	2.26
			3	-0.50	0.24	- 8.53	4.36
			4	-0.52	0.14	- 6.15	1.66
			5	-0.32	0.08	- 5.58	2.07
F-value				4.553	1.540	3.388	1.192

$F_{0.05} = 2.43$
$F_{0.01} = 3.44$

*Slow = 4 hr; Fast = 3-4 sec
**Fine = 0.0156 inch particle size
Coarse = 0.0312 inch particle size

calculated F value for the mean percent weight loss was 3.39 and was significant ($P<0.05$). Shrimp of the slowly frozen groups tended to remain approximately at the same level of moisture loss, whereas the shrimp of the rapidly frozen groups began to regain moisture after the initial moisture loss. Consistent with previous observations, shrimp of the rapidly frozen groups lost less moisture than the shrimp of the slowly frozen groups. The critical F values at D.F. 4/160 were $F_{0.05} = 2.43$ and $F_{0.01} = 3.44$.

The mean values of weight gain for the breading material are given in Table XIV. Calculated F values for the mean and mean percent weight gain of the breading material were 1.54 and 1.19, respectively, and were not significant ($P>0.05$).

Correlations

Significant correlation coefficients for the change in weight and percent change in weight for the shrimp and breading material are listed in Table XV .

Initial moisture of the breading material and the loss in weight of the shrimp are both negatively correlated to the gain in weight of the breading material. These negative correlations indicate: 1) as the initial moisture of the breading decreases, the gain in weight of the breading increases, and 2) as the shrimp loses moisture, or weight, the gain in weight of the breading increases. The gain in weight of the breading material is positively correlated to the dry matter of the breading material, meaning that as the dry matter content of the breading increases, the gain in weight of the breading increases.

The correlations related to the loss in weight of the shrimp

Table XV

Correlations

Change in Weight of:			Correlation Coefficient
Breading Material			
	·vs·	Breading Dry Matter	0.313**
		Breading Initial Moisture	-0.147*
		Shrimp Change in Weight	-0.749**
Shrimp			
	·vs·	Breading Dry Matter	-0.484**
		Breading Initial Moisture	-0.176*
		Breading Change in Weight	-0.749**
		Shrimp Dry Matter	-0.260**
		Shrimp Initial Moisture	-0.306**
Percent Change in Weight of:			
Breading Material			
	·vs·	Breading Initial Moisture	-0.317**
		Breading Change in Weight	0.958**
		Shrimp Change in Weight	-0.652**
		Shrimp % Change in Weight	-0.782**
Shrimp			
	·vs·	Breading Initial Moisture	0.326**
		Breading Change in Weight	-0.741**
		Shrimp Change in Weight	0.838**
		Breading % Change in Weight	-0.782**

*P 5% for 200 observations
**P 1% for 200 observations

are negative. These correlations can be stated as follows:

1) as the dry matter content of the breading increases, the shrimp will lose more weight,

2) as the initial moisture of the breading decreases, the weight loss of the shrimp will increase,

3) as the gain in weight of the breading increases, the weight loss of the shrimp increases,

4) as the dry matter content of the shrimp increases, the weight loss of the shrimp will decrease,

5) as the initial moisture of the shrimp increases, the weight loss of the shrimp increases.

SUMMARY AND CONCLUSIONS

Moisture migration and dehydration in frozen breaded shrimp were studied with respect to freezing rate, texture and equilibrium temperature of the breading material, and storage time. It was desired to determine the combined effect of these mass transfer mechaisms on the changes in weight of the shrimp and breading material.

Breaded shrimp, consisting of 50% shrimp, were produced in eight groups with the following conditions imposed on each group:

Group Number	Freezing Rate	Breading Material	
		Texture	Equilibrium Temperature (^{o}F)
1	Slow	Fine	82
2	Slow	Fine	40
3	Slow	Coarse	82
4	Slow	Coarse	40
5	Fast	Fine	82
6	Fast	Fine	40
7	Fast	Coarse	82
8	Fast	Coarse	40

Freezing rate refers to the time involved to freeze the sample, where the fast freeze involved 3 to 4 seconds at -320^{o}F and the slow freeze involved approximately 4 hours at -6^{o}F. Particle size of the breading material was designated by the texture, where the fine texture was 0.0156" in diameter and the coarse texture was 0.0312" in diameter. Before being applied to the shrimp, the breading material was allowed to come into thermal equilibrium with an ambient temperature. This ambient temperature was referred to as the equilibrium temperature of the breading material. The samples were then stored at -6^{o}F for 1 to 5 weeks. At one week intervals, samples were removed from the freezer and analyzed. Initial and final weights of the shrimp and breading material of the breaded shrimp samples were recorded, as were the dry weights of each. These weights were then statistically analysed in a factorial arrangement of treatments.

Results of the statistical analysis indicated:

1) The freezing rate had a highly significant effect on the mean values and mean percentage values of weight change of the shrimp and breading material. It was found that the fast freezing rate produced much less change in the shrimp and breading as compared to the slow freezing rate.

2) Texture of the breading material did not significantly affect the percent weight loss in the shrimp. There was, however, a significant difference between the mean values of weight gain for the fine and coarse textures. The fine texture lost more moisture to the surroundings than the coarse texture, resulting in a smaller change in weight.

3) Equilibrium temperature of the breading material significantly affected the mean values and mean percent values of weight gain of the breading material. Less moisture was lost by the breading material at $40^{o}F$ than at $82^{o}F$.

4) Storage time had a highly significant effect on the mean values and mean percentage values of weight change for the shrimp and breading material. The initial weight change observed in the shrimp and breading declined as the storage time increased.

Correlation coefficients were calculated for all possible pairs of the four characteristics. Sixteen pairs were significantly correlated.

It was concluded from the results that breaded shrimp produced by using a fast freezing rate, $82^{o}F$ equilibrium temperature, and fine texture breading material will give smaller changes in weight for the shrimp and breading material and, therefore, smaller gains in percent breading.

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INTERNATIONAL STANDARDIZATION OF SHRIMP PRODUCTS - U.S. CONTRIBUTION

ABSTRACT

The paper discusses the formation of the Codex Alimentarius Program which is a joint FAO/WHO endeavor to develop international food commodity trading standards that afford protecting the health of consumers and facilitating international trade in foods. The procedural steps in developing such international food standards through the Codex Alimentarius Commission are discussed, along with the responsibilities of the Governments serving as chairman countries of principal committees. An overview of U.S. activities relating to participation in the Draft International Standard for Shrimps and Prawns is elaborated.

INTERNATIONAL STANDARDIZATION OF SHRIMP PRODUCTS - U.S. CONTRIBUTION

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Most of the developed countries of the world today have complex and sophisticated national food standards. Even so, these countries face a continuing need to revise their regulations to take into account new technological developments. On the other hand, newly independent and developing countries are in the process of writing food laws and introducing systems of food regulation and control for the first time. Such countries are rapidly learning that food standards should safeguard the national interests but should not conflict with the requirements of the world's principal sources of food.

What then has brought about the widespread interest in international food standards over the past few years? One identifiable factor motivating most Governments is the prospect of facilitating international trade in food by the removal of non-economic barriers to trade, particularly in those countries dependent upon agricultural and fisheries exports.

A second factor of equal importance is the need to establish standards to ensure safe and wholesome food in international trade. Hence, the two most significant forces behind the development of international food standards are: (1) the protection of the health of the consumer, and (2) the need to facilitate international trade in foods.

In the past, little progress has been made in the field of international food standards in spite of various attempts and a variety of schemes initiated in Europe, Latin America, and Africa.

The increase in interest to alleviate trade problems was recognized by Member Governments of the Food and Agriculture Organization of the United Nations (FAO) and the World Health Organization (WHO), and a decision was made in the early 1960's to create a forum for international action.

The two international organizations moved rapidly after having decided to undertake the challenge of alleviating trade problems. In 1962, a joint FAO/WHO Conference on Food Standards was held and this conference endorsed the establishment of the Codex Alimentarius Commission and developed certain guidelines for its plan of work.

The purpose of Codex Alimentarius is very clearly set forth by the Commission in a Procedural Manual which states: "The Codex Alimentarius is a collection of in-

ternationally adopted food standards presented in a uniform manner. These food standards aim at protecting consumers' health and ensuring fair practices in the food trade. The Codex Alimentarius also includes provisions of an advisory nature in the form of codes of practice, guidelines, and other recommended measures intended to assist in achieving the purposes of the Codex Alimentarius. The publication of the Codex Alimentarius is intended to guide and promote the elaboration and establishment of definitions and requirements for foods to assist in their harmonization and, in doing so, to facilitate international trade."

The scope of Codex Alimentarius is also set forth by the Commission as follows: "The Codex Alimentarius includes standards for all the principal foods, whether processed, semi-processed or raw, for distribution to the consumer. Materials for further processing into foods should be included to the extent necessary to achieve the purposes of the Codex Alimentarius as defined. The Codex Alimentarius includes provisions in respect to food hygiene, food additives, pesticide residues, contaminants, labeling and presentation, methods of analysis, and sampling. It also includes provisions of an advisory nature in the form of codes of hygiene and technological practice, guidelines and other recommended measures."

The work of the Commission is carried on largely by committees, and each committee is chaired by a country. The chairmanship of a committee really means that the country is responsible for convening experts on the subject of its work and for drawing up standards which are then submitted to the Commission.

The Commission has developed a ten-step procedure for the elaboration of Codex standards. After a draft standard has been prepared by an "author" country and considered by the committees, the procedure allows two rounds of comments by Governments, two examinations by the committees, and two considerations by the Commission. Thereafter, the standard is formally sent to participating Governments for acceptance. This procedure has been deliberately designed to give participating Governments the fullest opportunity to comment on standards while they are still in draft and to allow the Commission to satisfy itself that the standards are being prepared in accordance with its general principles.

The nature of Codex committees can be divided into two types. One group of committees works on general subjects which, when adopted, are applicable to all food standards. Such committees and their chairman countries are as follows:

1. General Principles Committee--France. The purpose of this committee is to set up formats for the adoption of standards and procedures and the format to be used in the preparation of standards.

2. Food Labeling--Canada. The purpose of this committee is to establish provisions on labeling applicable to all foods in international trade.

3. Food Hygiene--United States. The purpose of this committee is to develop basic principles for food plant sanitation and for handling food in international trade.

4. Food Additives--Netherlands. The purpose of this committee is to recommend international tolerances for individual additives in specific food items.

5. Pesticide Residues--Netherlands. The purpose of this committee is to recommend international tolerances for pesticide residues in specific food products.

6. Analysis and Sampling--Federal Republic of Germany. The purpose of this committee is to determine the best method of analysis and sampling for products in which Codex standards are in preparation.

7. Meat Hygiene--New Zealand. The purpose of this committee is to develop basic principles for meat plant sanitation and for handling meat in international trade.

The other group of committees under the Codex Alimentarius is the so-called "commodity committees." Presently, there are nine committees working on standards for specific food products. Of course, this group of committees does not cover all foods, and additional products for standardization are added to their workload from time to time.

The Codex Commodity Committees currently active, along with the country acting as chairman, are as follows:

Cocoa products and chocolate--Switzerland

Sugar--United Kingdom

Processed fruits and vegetables--United States

Fat and oils--United Kingdom

Fish and fishery products--Norway

Dietetic foods--Federal Republic of Germany

Meat and meat products--Federal Republic of Germany

Two other bodies, both of which are Joint ECE/Codex Groups of Experts, are elaborating international standards for fruit juices and frozen vegetables.

The Codex Committee on Fish and Fishery Products, which is of greatest interest to you, functions with Norway as the chairing country. This committee has the responsibility for developing worldwide standards for fresh, frozen, or otherwise processed fish, crustaceans, and mollusks. This committee has met in session ten times and has about fifteen major products for which standards are being developed. Six standards have been completed; these are: Canned Salmon, Frozen Gutted Pacific Salmon, Canned Shrimp, Frozen Fillets of Cod and Haddock, Frozen Fillets of Ocean Perch, and Canned Tuna and Bonito. Countries will be asked to accept these standards in the near future. At its most recent session held October 1975, this committee completed its work on standards for Canned Crab Meat, Frozen Fillets of Flatfish, and Frozen Shrimp and referred the standards

to the Commission for approval. The remaining fishery products for which standards are being elaborated are as follows:

Canned Sardines

Frozen Fillets of Hake

Frozen Fish Blocks (fillets and minced) for further processing

Canned Mackerel in Brine or Oil

Frozen Lobsters

The United States Delegation to the sessions of the Codex Committee on Fish and Fishery Products usually is composed of three Government officials. Additionally, several industry advisors to the Government delegates also attend and participate in the committee sessions. At the Tenth Session of the Codex Committee on Fish and Fishery Products, eight industry advisors to the United States Delegate attended. These advisors serve by invitation and frequently are individuals who are experts in more than one commodity.

In respect to standards for fishery commodities, coordination and liaison with the industry is handled through commodity-oriented trade associations such as the National Fisheries Institute, the Tuna Research Foundation, American Shrimp Canners Association, Maine Sardine Council, National Shrimp Breaders and Producers Association, American Frozen Foods Institute, and others. When needed, meetings are held with the appropriate commodity-oriented industry group in respect to a

specific product standard. In addition to coordination with industry, NMFS utilization research centers and other Federal and State agencies are invited to comment on the standards and, on occasion, to test the practical applicability of selected provisions of standards such as defects tables. Thus, when comments are presented during the developing stages of standards for fishery products on behalf of the United States, the best technical and responsive industry and Government views are presented.

Let us now focus on the Draft Codex Standard for Shrimps and Prawns which is of particular interest to this group. The scope of the standard is very broad covering all styles of frozen raw or cooked shrimp of the four major families of shrimp, i.e., Peneidae, Pandalidae, Crangonidae, and Palaemonidae. It does not cover specialty products where shrimp only constitute a portion of the edible contents.

Many forms of packs covered by the standard are also included:

Whole
Headless
Peeled (tail fans on)
- round
- round and deveined
- fantail (split or cutlet)
- western style

Peeled (tail fans removed)
- peeled
- peeled and deveined

Pieces

As you can see, the standard, as designed, is applicable to all the major forms of pack produced domestically and by foreign countries.

A major effort to develop this international standard for frozen shrimp began at the Fifth Session of the Codex Fish Committee in 1970. At that meeting, individuals knowledgeable of shrimp from the United States, Denmark, Australia, and Brazil laid down some basic points of principle upon which the standard was to be based. The late Ray Robinson, with whom many of you here were acquainted, was one of the U.S. industry advisors who assisted in laying the groundwork. Jim Brooker of our inspection program in Washington also participated in this initial effort.

In the years that followed, the National Fishery Products Inspection and Safety Laboratory in Pascagoula, Mississippi, was assigned the major responsibility for the various critical reviews of the standard and for testing the defects table. Over the four- to five-year period, samples of shrimp of all species, styles, forms of pack, and sizes were tested. The basic point that we kept in mind was to assure that this standard reflected reasonable minimum quality and technical requirements that are desired and could be met by the U.S. industry. To meet this task, we concentrated on the various product defects for the different forms of pack.

Principal defects found from test data on the forms examined were as follows:

Frozen Raw Headless Shrimp (<200/lb.)

1. No. sample units - 671
2. No. lots - 165
3. Total poundage examined - 877,685
4. Percent lots rejected - 17%
5. Principal defects - Black spot (shell and meat)
   Cut, torn, and damaged shrimp
   Pieces (<5 segments)
   Heads and parts of heads

Frozen Raw Peeled Shrimp (<200/lb.)

1. No. sample units - 81
2. No. lots - 6
3. Total poundage examined - 189,000
4. Percent lots rejected - 25%
5. Principal defects - Black spot on meat
   Cut, torn, and damaged or pieces

Frozen Raw Peeled and Deveined Shrimp (<200/lb.)

1. No. sample units - 618
2. No. lots - 150
3. Total poundage examined - 483,008
4. Percent lots rejected - 5%
5. Principal defects - Black spot on meat
   Incomplete deveining
   Cut, torn, and damaged or pieces
   Legs, loose shell, antennae

Frozen Cooked Peeled Shrimp (<200/lb.)

1. No. sample units - 142
2. No. lots - 48

3. Total poundage examined - 212,434

4. Percent lots rejected - 2%

5. Principal defects - Cut, torn, and damaged or pieces

Frozen Cooked Peeled and Deveined Shrimp (<200/lb.)

1. No. sample units - 372

2. No. lots - 85

3. Total poundage examined - 192,842

4. Percent lots rejected - 0

5. Principal defects - Improperly peeled
Incomplete deveining

During the developmental stages of the draft standard, several changes were made to the proposed defect table. In the main these related to black spot and the residuals for sodium bisulfite. During discussions on the draft standard, the Delegates of the United States, Central and South America, Japan, and Australia all spoke of the need to allow the use of sodium bisulfite on shrimp harvested from tropical and semi-tropical waters to prevent "black spot." It was finally decided to allow a tolerance of 100 mg/kg on raw meat or 30 mg/kg in the cooked meat expressed as SO_2 singly or in combination. Prior to the U.S. Delegation accepting the resolution, the committee accepted the U.S. proposal to add an additional defect relating to abnormal coloration to prevent acceptance of excessively bisulfite treated shrimp which were acceptable within chemical residue tolerances. So that, in fact, a product could be rejected on the basis of discoloration

evidencing the misuse of sodium bisulfite even though the end product form might be in compliance with the chemical tolerances.

Some of the key industry members who contributed substantially in the developmental phases included the following:

J. R. Duggan - King Shrimp Co.

Don Toloday - Singleton

R. Martin - NFI

F. Jermann - Alaska King Crab Quality Control Board

John Farquhar - American Frozen Food Institute

Walter King - Red Lobster Inn

Rafael Pedraja - Booth Fisheries

Jack Cofer - Sea Pak

We fully appreciated the help and assistance provided by these and others, particularly our USDC inspectors, in the development of this document.

As I mentioned earlier, this draft standard will be considered by the Codex Alimentarius Commission in April 1976 and, upon approval, it will be sent to Member Governments as a Recommended International Standard for consideration as to acceptance.

United States acceptance of the standard is the next vital step. This means that a petition will be filed to FDA proposing to establish a "Standard of Identity and Minimum Quality" for frozen shrimp based on the International Standard. At that time, all

interested parties will have an opportunity to express their views and comments on the document and this is the time when it will be important for all of us here to carefully consider and move forward to assure that it is accepted.

Thereafter, the trade will have a definite working document on which to trade in shrimp products.

BACTERIOLOGY OF SHRIMP

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The number and types of bacteria on seafoods depend on many factors such as species, area of catch, method of catching, handling on board, further processing and time and temperature of storage. Bacterial counts and types of shrimp vary greatly at the time of landing and at the time of retail sale. An examination of the major steps in catching and processing can help to explain some of these variations.

Fresh Caught - Shrimp are caught by trawl in waters from 10-15 feet deep (bays) to 20-120 or deeper (Gulf). The muscle tissue of freshly caught shrimp is generally regarded as sterile, but the outer surface and intestinal tract contain various bacteria. The number and type of bacteria present depend on water depth (temperature), nearness to shore (estuarine or open water), type of bottom, size of shrimp, time of trawl, and shrimp habitat.

Reports on the number of bacteria found on freshly caught shrimp range from 2.5×10^2 to 2.0×10^6/gram, with Gulf counts averaging from 1.0×10^3 to 1.0×10^4/gram and bay counts averaging from 1.0×10^4 to 1.0×10^5/gram (Campbell and Williams, 1952; Carroll et al., 1968; Cobb et al., 1973; Cook, 1970; Green, 1949;

Vanderzant et al., 1970). Pink shrimp which usually are caught on sand bottoms, have lower counts than brown or white shrimp caught on muddy bottoms (Carroll et al., 1968). Green (1949) showed an inverse relation between the size of the shrimp and the bacterial count. The average count of "15-count" shrimp was 3.1×10^4/gram, "25-count" shrimp 7.5×10^4/gram, and "60-count" 2.1×10^5/gram. She also indicated that longer trawl times (2-3 hours) were more effective in reducing counts by washing than were short trawls (7-15 minutes with "try-net"). Although counts may be reduced by long trawl times, the resulting physical destruction of the shrimp may lead to early bacterial invasion and spoilage. Another factor that influences the number and type of bacteria on a freshly caught shrimp is that white shrimp are caught in daylight hours, whereas brown shrimp are caught at night during cooler ambient temperatures.

Various types of bacteria have been reported on freshly caught shrimp. In the early fifties Campbell and Williams (1952) and Williams et al. (1952) showed species of Achromobacter, Bacillus, Micrococcus, Flavobacterium, and Pseudomonas to be predominant in Gulf Coast shrimp. Vanderzant et al. (1970) reported that the flora consisted of coryneforms, Achromobacter, Flavobacterium, and Bacillus. In Pacific shrimp, Acinetobacter-Moraxella species were predominant (Harrison and Lee, 1968). In a more recent report, Koburger et al. (1975) reported the Flavobacterium-Cytophaga group to represent most organisms isolated from fresh rock shrimp (Sicyonia brevirostris). Some of the differences in the bacterial population of shrimp can probably be attributed to differences in

shrimp species, marine environment or handling on board. The fact that Achromobacter species are not predominant in the more recent studies on shrimp from the Gulf of Mexico and the Pacific is due to a change in the taxonomic status of this organism (Baumann et al., 1968; Buchanan and Gibbons, 1974). Recent investigations (Cobb et al. 1973) indicate that typical spoilage organisms of the genus Pseudomonas are not predominant on freshly caught shrimp. It is not until the shrimp are exposed to handling on board the vessel that this organism becomes more predominant.

Handling on Board - At the completion of the trawl, the "cod-end" or bag of the trawl net is hauled-in and dumped on the deck of the vessel. The shrimp are then separated from the incidental fish catch. If the shrimp are from the Gulf they are headed by the crew at that time. Bay shrimp are brought in heads-on. The separated shrimp are then placed in metal or plastic baskets and washed before icing or freezing. Time on the deck, heading and amount of washing all influence the number and type of bacteria on the shrimp.

Fieger et al. (1958) exposed freshly caught shrimp to air temperatures 79-84°F (26-29°C) for different periods of time before icing. The initial differences in number of bacteria between samples were not significant. However, after storage on ice for 11 days, those exposed 2 and 6 hours at the elevated temperatures had respective counts of 3 and 70-fold greater than the control that had not been exposed. The prolonged exposures also increased the development of melanosis (black discoloration).

Although the head (cephalothorax) accounts for 40% of the total weight of shrimp, it has been reported to contain 50-80% of the total

bacterial load (Carroll et al., 1968; Green, 1949). For many years it has been emphasized that shrimp should be headed as soon as possible and that heads-on shrimp should not be stored on ice for more than 4 days. Heading reduces initial bacterial levels. The intestinal tract which contains some bacteria is often removed with the head and bacterial populations tend to multiply more rapidly on heads-on shrimp than on tails. For rock shrimp the opposite may be true. The tough rigid outer shell seems to offer the flesh some protection from spoilage. Rock shrimp stored with heads-on had lower total counts, maintained higher solids content, and greater organoleptic acceptability than did heads-off shrimp (Bieler et al., 1973).

Adequate washings of shrimp with clean seawater can reduce the bacterial populations associated with surface slime and adhering debris. Green (1949) sampled 0.2 gram of slime from shrimp and found counts of 2.1×10^5/gram. Shrimp from which the slime had been removed yielded 1.5×10^4/gram. These differences in counts show the importance of washing shrimp prior to icing. Carroll et al. (1968) stated that efficient washing may reduce the initial bacterial count by as much as 75%. They also noted that washing had little effect in maintaining lower bacterial counts during the first week of iced storage but had marked effects on bacterial counts during the second week of storage. Result demonstrations conducted aboard various vessels indicate that proper washing before iced or frozen storage is the most important single factor influencing the bacterial numbers on shrimp (Nickelson and Graham, unpublished data). To summarize the changes in bacterial numbers of shrimp as influenced

by handling on board, the following averages have been reported by Green (1949).

	Average
Fresh Caught Shrimp Heads-on	42,000/gram
Fresh Caught Shrimp Tails	10,000/gram
Fresh Caught Shrimp Tails Washed	7,400/gram

Iced Storage - Shrimp are commonly stored aboard vessels in bins located in the hold. An initial layer (6-8 inches) of crushed ice is placed on the floor and sides, then alternate layers of shrimp and ice are placed in the bin. Recommended practices suggest that the ice and shrimp be mixed at a ratio of 2:1 (Carroll et al., 1968). The pile of shrimp and ice is made higher and higher by the addition of "bin-boards" to create the fourth side of the bin. At unloading the shrimp and ice are removed by bucket and hoist or mechanical vacuum equipment and are then dumped into a thaw tank. The shrimp, which sink to the bottom are separated from the ice and removed by conveyor for further processing.

Initially, the microbial flora is a mixture of types from marine and terrestrial environments. During iced storage, the number of bacteria begins to increase and the types shift to a predominantly psychrotrophic (cold temperature) flora. The rate of increase in bacterial number depends on initial numbers of bacteria, handling on deck and amount and quality of ice used. Figure 1 shows the increase in bacteria on shrimp held in crushed ice and was taken from data presented by Campbell and Williams (1952) and is representative of the work reported by other researchers.

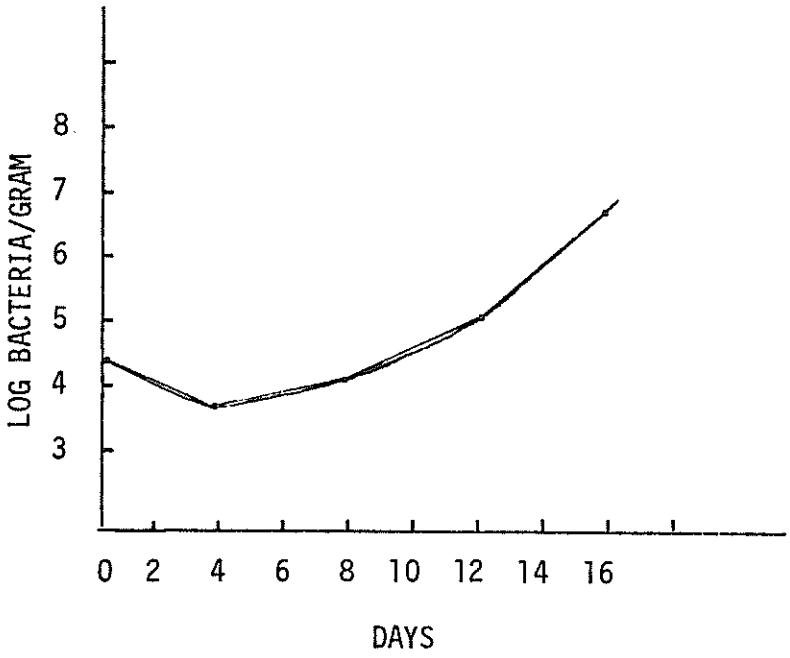

Figure 1. Changes in the bacterial count of shrimp stored on ice.

The initial decrease in numbers is caused by a decrease in certain mesophilic organisms that can not adjust to the colder temperatures and because those capable of growth at refrigeration temperatures are slow in getting started. Bacterial multiplication after that time is usually by the more typical spoilage organisms. These changes in bacterial types have been reported by several investigators. Campbell and Williams (1952) showed Bacillus, Micrococcus, and Flavobacterium to make up over 50% of the flora initially, whereas, the Achromobacter-Pseudomonas group accounted for 98% of the flora after 16 days of storage. In a study on the bacterial spoilage patterns of headless brown shrimp, Cook (1970) noted that there were no consistent changes in bacterial types initially or during decreases in bacterial count. However, as the counts began to rise Pseudomonas species became the predominant species accounting for 80-100% of the bacterial types isolated. Vanderzant et al. (1970) reported that

the predominant bacterial flora of fresh shrimp consisted of coryneforms and that of stored shrimp of Pseudomonas species. Shrimp handled in such a manner as to avoid contamination by Pseudomonas could be stored on ice for 21 to 30 days before quality losses were noted. Cobb et al. (1976) reported a varied microflora of the drip from iced shrimp during the first few days of storage. Pseudomonas and Moraxella-Acinetobacter species predominated in the drip at the end of the storage period. The inability of Vibrio, Flavobacterium and Micrococcus to proliferate may be attributed to temperature, changes in salinity, changes in nutritional properties of shrimp surface and/or interactive microbial activities.

Green (1949) reported that bacterial counts in different layers of shrimp varied. Counts on shrimp from the uppermost layer increased 2-fold whereas those on the bottom layer being inoculated by the melt water increased 1000-fold. The bacterial count of the melt water from the bottom of the bin of 1-day old shrimp was 5.9 x 10^6/ml. She also showed the importance of using and maintaining good quality ice. Unused ice stored for 2 days on one boat contained 8.5 x 10^4/gram and ice from another boat after 7 days contained 2.3 x 10^5 bacteria/gram.

Attempts have been made to prolong the shelflife of iced shrimp by treating the shrimp or the ice with various chemicals or antibiotics. Green (1949) and Fieger et al. (1956) found the following to be no more effective than regular commercial ice: 1 ppm chlortetracycline in ice; acid ice at pH 5.0; 500 ppm tannic acid in ice; 100 ppm sodium bisulfite in ice; and chloramine-T and sodium benzoate in ice. Cook and Bowman (1972) showed that

the treatment of shrimp with 1% sodium bisulfite before icing or ice containing 1% Na_4EDTA (sodium salt of ethylenediaminetetraacetic acid) controlled melanosis and improved the organoleptic quality by retarding spoilage by Pseudomonas species. At this time Na_4EDTA is not cleared for use in ice in which shrimp are stored.

Freezer Boats - In the past few decades, vessels with mechanical refrigeration systems have been developed in an effort to provide a better quality product and to enable the vessel to remain on the fishing grounds for a longer period of time. Shrimp are landed and handled in the conventional manner and are then placed in an open mesh bag (40-50 pounds of shrimp/bag) and immersed in a refrigerated brine solution for rapid freezing. The brine usually consists of 20% NaCl, various concentrations of sugar, molasses or corn syrup solids (to provide a "glaze" on the product), and possibly sodium bisulfite to retard the formation of black spot. The bag of shrimp remains in the brine (at -20C for about 30 minutes) until the shrimp are frozen. They are then removed and stored in the freezer hold. Freezing will destroy as much as 85% of the bacterial population and inhibit the growth of those surviving. Data on the bacteriological changes on shrimp from freezer boats are limited or not available. Factors that will influence the bacterial quality of boat shrimp will include:

1. State of the product prior to freezing
2. Rate of freezing
3. Quality of the brine
4. Storage temperature and history
5. Handling at unloading.

Unloading - The bacteriological quality of shrimp unloaded from a vessel depends on the manner in which it was handled and the time and temperature of storage. Bacterial counts of shrimp being unloaded from vessels vary greatly and have been reported to range from 870-1,300,000 per gram (Vanderzant et al., 1970).

Processing - Shrimp from the Gulf of Mexico reach the consumer in many different forms.

1. Green headless
2. P & D IQF
3. P & D cooked IQF
4. Frozen Raw Breaded Shrimp

Each step in the processing procedure can influence the bacteriology of shrimp and certain processing steps are common to all of the above mentioned forms.

Green-headless (shell-on tails) shrimp usually arrive at the processing plant by truck and are unloaded into a thaw tank to remove ice that may have been placed on the shrimp for transportation. The shrimp are then moved by conveyor to a series of size graders and are separated on the basis of number of shrimp per pound. Each grade size is manually weighed (5 lbs) and placed in a cardboard box with an adequate amount of water to cover the bottom. Upon freezing (usually by plate freezer) the water forms a "glaze" in the bottom of the box. After 12-24 hours the box is removed from the plate freezer, inverted and water is added to glaze the top of the box. Boxes of shrimp are placed into 10 box lots and transferred to cold storage for future shipment to wholesalers, retailers, institutional outlets or other plants for further processing.

Limited data are available on the bacteriological quality of shrimp in a green headless operation. The incoming product will vary depending on its past history. Green (1949) reported that shrimp after transit from the vessel had counts of 3.2×10^7/gram. This was higher than any counts reported on shrimp as they were unloaded and could be attributed to improper refrigeration during transportation. The factors that influence the bacteriological quality of green headless shrimp include:

1. Quality of the water in the thaw tank (some are changed continuously and contain 200ppm chlorine)
2. Cleanliness of grading equipment and conveyors
3. Rapidity of entire process
4. Employee personal hygiene
5. Quality of glaze water
6. Time and temperature of freezing.

Surkiewicz et al. (1967) reported aerobic plate counts of green headless shrimp purchased by raw breaded shrimp processors to range from 3.0×10^4 to 4.0×10^7/gram. Coagulase-positive staphylococci were isolated from 42% of the samples, coliforms from 79%, and Escherichia coli from 14%.

P & D, IQF (peeled, deveined and individually quick frozen) shrimp are graded as to size before processing. The shrimp are then peeled and deveined mechanically or by hand labor. Since most of the bacteria associated with shrimp have been reported to be on the outer shell, one would expect a decrease in bacterial count after peeling and deveining. This is usually true in mechanical operations where large volumes of water are used to flume (transport) shrimp to the

different processing machines. Controlling bacterial counts on shrimp peeled and deveined by hand is more difficult. Repeated increases in total counts, coliform and coagulase-positive staphylococci, were reported by Surkiewicz et al. (1967) in shrimp that had been peeled and deveined by hand. This problem could be eliminated by washing the shrimp before and after peeling and deveining and closer supervision and education of plant personnel.

If the shrimp received by the plant are already graded and frozen, the thawing process prior to processing can greatly influence the bacterial counts of the shrimp. Surkiewicz et al. (1967) noted that thawing processes ranged from placing boxes of frozen shrimp on the floor at room temperature overnight to rapid thawing in cold running water for 2 hours. Plate counts of P & D shrimp which were thawed overnight were 2.0×10^8/gram, whereas, those thawed for 2 hours remained about the same as the original frozen shrimp (6.0×10^5/gram).

After peeling and deveining the shrimp are individually-quick-frozen by liquid nitrogen, freon or other means and glazed before packaging. Freezing and the use of high quality glaze water will reduce or maintain bacterial counts at the prefreezing level.

P & D, cooked, IQF shrimp are processed in the same manner as described for green headless and P & D, IQF with the exception of the cooking process. Harrison and Lee (1968) showed that cooking shrimp for 3 minutes in boiling water caused a 1000-fold reduction in bacterial counts. Re-exposure to human handlers after cooking not

only increased total counts but increased the percentages of gram-positive cocci isolated. Most isolates were coagulase-negative, only one coagulase-positive Staphylococcus was isolated.

Frozen Raw Breaded Shrimp are graded, peeled-deveined as previously described and then conveyed through liquid batter and dry breading material. Depending on the type of product being processed, the batter-breading procedure is repeated until the breading approximates 25-50% of the total weight of the product. Surkiewicz et al. (1967) found that dry batter material (flour, seasoning, nonfat dry milk powder, and dried eggs) contained relatively low levels of bacteria (1.0×10^3 to 6.0×10^4/gram). Plants that did not chill or discard liquid batter were found to have over 1.0×10^6 bacteria per gram of batter in most samples. Liquid batter that was chilled and discarded at least once a day had counts ranging from 5.0×10^3 to 5.0×10^5/gram. Their survey also showed that plants with "good" quality control consistently produced breaded shrimp with bacterial loads lower than the original shrimp.

The bacterial content of the finished frozen raw breaded shrimp depends on the manner in which it has been handled from the vessel to the final product. In the survey conducted by Surkiewicz et al. (1967), the geometric mean for aerobic plate counts on products from plants operating under good sanitary conditions ranged from 4.9×10^4 to 7.0×10^6/gram, that of products from poor plants varied between 4.4×10^5 and 1.4×10^8/gram. Vanderzant et al. (1973) reported finish products counts on 89 samples from a

plant operating under good sanitary conditions to range from 1.1 x 10^4 to 6.8 x 10^6/gram with plate incubation at 35°C and 6.0 x 10^4 to 2.7 x 10^7/gram with plate incubation at 25°C.

Retail samples of frozen raw breaded shrimp have been examined by several investigators. In each case the arbitrary value of 1.0 x 10^6/gram for the aerobic plate count has been used to assess bacterial quality. Silvermann et al. (1961) reported 28% of samples to have aerobic plate counts in excess of 1.0 x 10^6/gram. In a similar, more recent, study (Nickerson and Pollok, 1972) aerobic plate counts were reported to range from 2.4 x 10^4 to 6.0 x 10^7/gram with 65 of 135 samples over 1.0 x 10^6/gram. With plate incubation temperatures of 25°C, Vanderzant et al. (1973) found 52% of all retail samples to contain more than 1.0 x 10^6 per gram. In contrast, Surkiewicz et al. (1967) found only 15% of plant samples with counts exceeding 1.0 x 10^6/gram and Vanderzant et al. (1973) reported 17% of plant samples had counts above 1.0 x 10^6/gram at incubation temperatures of 35°C. This indicated improper handling and temperature control in transportation and in the retail market.

The initial bacterial flora of frozen raw breaded shrimp consisted primarily of Pseudomonas, Achromobacter, Aeromonas, Bacillus, Moraxella, Microbacterium, Micrococcus or coryneform bacteria (Vanderzant et al., 1973). Bacillus, Microbacterium, Micrococcus and coryneform bacteria were predominant in retail samples. The microbial flora of batter and breading which consisted primarily of Bacillus and Microbacterium species, most likely contributed to this condition.

Public Health and Indicator Organisms - Green (1949) found that almost 50% of freshly caught, iced shrimp on board of fishing vessels contained coliforms, but that E. coli was rarely encountered. Most reports on the bacteriological flora of freshly caught shrimp indicate the absence of coliform bacteria (Campbell and Williams, 1952; Harrison and Lee, 1968; and Vanderzant et al., 1970). In shrimp arriving at the processing plant, Surkiewicz (1967) found coliforms and E. coli in 79% and 14% of the samples, respectively. Coliforms and E.coli on shrimp as unloaded from the vessel, may be good indicators of improper handling on board.

The sensitivity of coliforms to frozen storage has led some to the assumption that the more resistant enterococci might be to better sanitary quality index. Vanderzant et al. (1973) noted that no relationship existed between coliform counts of products before shipment and enterococcal counts of products at time of purchase. In the same study, neither Salmonella nor Vibrio parahaemolyticus were isolated from plant processed samples.

Bacteriology of Other Shrimp - Zapatka and Bartolomeo (1973) reported counts of freshly harvested Gulf of Maine shrimp to range from 1.0×10^2 to 1.6×10^3/gram, with a geometric mean of 5.1×10^2/gram. E. coli, coliforms and coagulase-positive staphylococci were absent. Although subsequent storage and improper handling during processing increased total counts and coliforms, the low air temperature (-7.8 to 7.2°C) and large volume of cold water (1.1°C) used inhibited significant bacterial buildup in the finished product.

In a survey of imported shrimp, Surma and Koburger (1972) reported average counts of over 3.0×10^6/gram for frozen green-headless and frozen P & D shrimp. Total counts for domestic shrimp in the same study averaged 8.4×10^5/gram. Since the complete history of the imported products was not known, no speculation could be made as to what an acceptable total count would be.

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TIME AND TEMPERATURE PARAMETERS FOR THE DESTRUCTION OF *SALMONELLA* IN COOKED, PEELED AND DEVEINED SHRIMP

ABSTRACT

The purpose of the study was to determine if the range of time/temperature parameters currently being utilized in U.S. Department of Commerce inspected plants for the production of cooked, peeled, deveined, and individually quick frozen shrimp are sufficient to destroy salmonellae. The conclusion of the study as described in this report is that the current time/temperature processing parameters are sufficient to destroy salmonellae during the cooking process.

TIME AND TEMPERATURE PARAMETERS OF THE DESTRUCTION OF *SALMONELLA* IN COOKED, PEELED, AND DEVEINED SHRIMP

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Introduction

The purpose of the study was to determine if the range of time/temperature parameters currently being utilized in U.S. Department of Commerce inspected plants for the production of cooked, peeled, deveined, and individually quick frozen shrimp are sufficient to destroy salmonellae. The conclusion of the study as described in this report is that the current time/temperature processing parameters are sufficient to destroy salmonellae during the cooking process.

Fresh high-quality shrimp of varying sizes were inoculated with three different species of *Salmonella*, soaked in either tap water, sodium chloride (2%), and sodium chloride (2%) plus sodium tripolyphosphate (4%). After soaking, the shrimp were cooked, the time of cooking varying with the size of the shrimp. Microbiological analyses were conducted with regards to the survival of *Salmonella* in shrimp after soaking, after cooking, and again after 30 days of storage at 0°F.

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Other sample lots of cooked, peeled, and deveined shrimp were inoculated with Salmonella species and stored in heremtically-sealed containers at 0°F for 30 days. Salmonella determinations were conducted prior to storage, and after 1, 3, 5, 10, and 30 days of storage.

Prior to this study, the U.S. Department of Commerce Inspection Service was certifying cooked, peeled, and deveined shrimp as wholesome and safe for human consumption when, quite frankly, little knowledge was available relative to the finished products containing Salmonella organisms. Much of the shrimp raw material is imported from as many as eighty different countries, with little or no knowledge of the initial Salmonella load. Such shrimp may contain high numbers of organisms, and it was believed that processing practices were not sufficient to render such products safe. A comprehensive study was conducted to determine if the current range of U.S. processing parameters for cooked shrimp destroyed the more heat-resistant and more commonly isolated Salmonella organisms.

Materials and Methods

Three species of Salmonellae (Salmonella senftenberg 775 W, S. oranienburg, and S. montevideo) were obtained, and immediately subjected to biochemical and immuniological tests to ascertain purity and correct classification of the strains.

A sufficient quantity of high-quality shrimp, peeled and deveined, of three sizes (21-25/lb, 36-40/lb, and 51-60/lb) were obtained from a Gulf coast commercial concern. A sufficient quantity of a fourth size (over 100/lb) of shrimp, peeled but not

deveined, were obtained from another Gulf coast commercial concern. Immediately after arrival at the laboratory, all shrimp were frozen at 0°F. Approximately 18 hours before subjecting the shrimp to the various tests, the products were transferred to a refrigerator and allowed to thaw at 40°F, whereupon each package of shrimp was tested for total plate count of the indigenous flora and for the possible presence of pre-contaminating Salmonella organisms.

The tests were carried out as follows: Aseptically, 25 g of shrimp were weighed into sterile petri plates and transferred to sterile blending containers containing 225 ml of sterile lactose broth. The samples were blended for two minutes. One ml of the blended shrimp was transferred to sterile phosphate buffered water blanks, and standard serial dilutions were performed to determine the total number of organisms using Bacto-plate count agar as the medium of choice. The inoculated plates (in duplicate) were incubated for 48 hours at 35°C.

The remainder of the blended samples were transferred aseptically to sterile, wide-mouth, screw-cap pint jars and incubated for 24 hours at 35°C. After incubation, the shrimp-lactose broth mixture was gently shaken, 0.5 ml transferred to 10 ml of selenite-cystine broth, and an additional 0.5 ml was transferred to 10 ml of brilliant green tetrathionate broth, and both media incubated for 24 hours at 35°C. A 3 mm loopful of the incubated selenite-cystine broth and brilliant green tetrathionate broth was streaked on selective recovery media of brilliant green agar plates, Salmonella-Shigella agar plates, and bismuth sulfite agar plates. The plates were incubated ro 24 hours at 35°C. with extended incubation

being continued for an additional 24 hours when plates did not contain growth.

All colonies appearing on the recovery agars were transferred to triple sugar iron (TSI) agar slants and incubated for 24 hours at 35°C. A minimum of six TSI cultures were examined for every 25 g sample tested.

Any culture showing TSI reactions was transferred to the following media for biochemical tests: urea broth, lysine decarboxylase broth, phenol red dulcitol broth, phenol red mannitol broth, phenol red adonitol broth, malonate broth, tryptone broth, phenylalanine agar, KCN broth, and MR-VP medium.

With regard to the first phase of the project, e.g., artificial inoculation with three different species of Salmonella, soaking in three different solutions, processing (cooking) according to the preselected time/temperature parameters based on the size of the shrimp, and storage for 30 days, 500 g of shrimp were aseptically transferred to 1,000 ml of soaking solutions, consisting of either tap water, tap water plus sodium chloride (2% NaCl), or tap water plus 2% NaCl plus sodium tripolyphosphate (4% TPP). Each soaking solution was inoculated with 1.5 ml of the respective organisms. The density of the inoculum based upon the cell mass was adjusted to a McFarland #1 standard by using a Bausch and Lomb Spectronic 20 spectrophotometer. A McFarland #1 standard has a percent transmittancy of approximately 56% T, corresponding to approximately 300,000,000 cells/ml. The shrimp were allowed to soak in the inoculated soaking solutions for 30 minutes, whereupon a sample was taken to determine the total plate count after soaking

but before processing (cooking by boiling). The remainder of the shrimp were subjected to processing in boiling tap water, the boiling times corresponding to the particular size of the shrimp samples (Fig. 1). After the cooking process, the shrimp from the samples were removed, hermetically sealed in plastic bags, and stored for 30 days at 0°F, at which time the total plate count of the inoculated shrimp samples were again determined.

Immediately after the proper processing time, another sample of shrimp was taken and subjected to a total plate count to determine the presence of any surviving bacterial organisms.

With regard to the second part of the project--the survival rates during storage of Salmonella organisms--three sample lots of cooked, peeled, and deveined shrimp were exposed to the maximum processing time (21-25/lb, 5 min; 36-40/lb, 4 min; 51-60/lb, 3.5 min) in order to eliminate the indigenous flora of the shrimp. These processed shrimp were submerged in tap water which had previously been inoculated with the above-mentioned Salmonella species at a level of 10^2 per gram for 30 minutes. Following this artificial inoculation, an adequate sample was taken to quantitatively determine the presence of Salmonella species prior to storage of the remainder of the inoculated shrimp in hermetically sealed plastic bags at 0°F. The presence of Salmonella organisms was determined after 1, 3, 5, 10, and 30 days of storage, using plate count agar as the medium of choice.

All experiments indicated in this report were replicated three times.

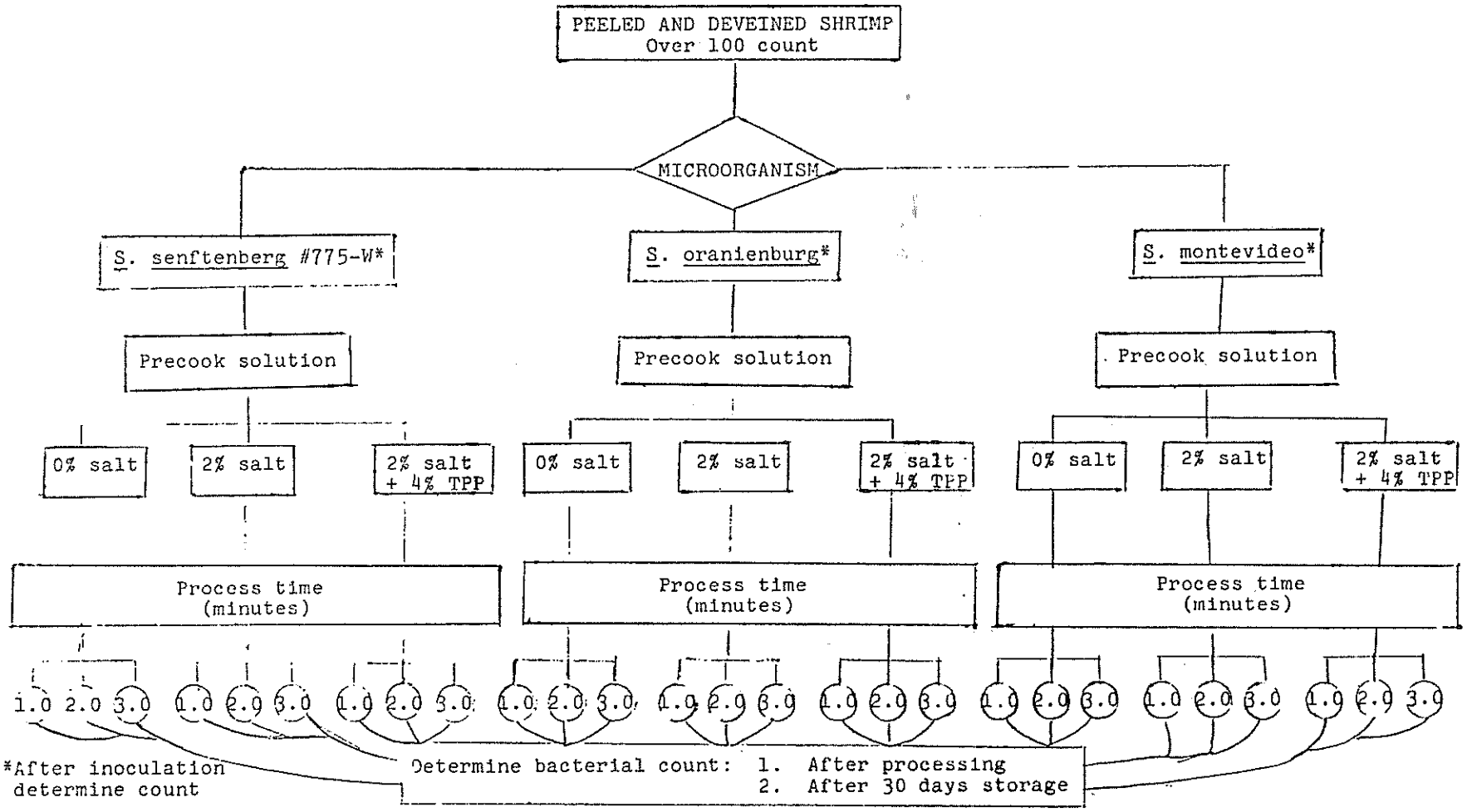

Fig. 1.--Flow chart.

Results

The preselected three Salmonella species are almost identical in their biochemical reactions (Tables 1 and 2). The only differential reaction occurs in triple sugar iron agar (TSI) in that S. montevideo and S. oranienburg are positive for the production of hydrogen sulfide while S. senftenberg 775 W indicates a negative H_2S production. Table 3 clearly demonstrates a vast difference among these three organisms with regard to their somatic and/or flagellar antigens. Therefore, throughout this investigation, whenever possible Salmonella species were suspected, they were inoculated into urea agar and TSI agar slants and after 24 hours incubation were tested using either somatic antigen 1, 3, 6, 7, or 19, and flagellar antigen G, M, S, and T.

Table 4 indicates the indigenous flora of the starting raw material used throughout the various experiments and treatments. It was interesting to note that the shrimp samples counting over 100/lb possessed an initial bacterial load far in excess of that for any other count size category.

Tables 5, 6, and 7 indicate the inoculum load immediately prior to the samples receiving various treatments.

Table 7A indicates that at no time did any of the Salmonella species survive the cooked processing treatment utilized for the various inoculated packs. The significance of this is that the time/temperature parameters currently being utilized in the U.S. Department of Commerce, for the production of cooked, peeled, deveined, and individually quick frozen shrim products are

adequate to destroy massive numbers of Salmonella during the cooking procedure of the process.

Tables 8, 9, and 10 indicate that the indicated species of Salmonella will survive the freezing process for differing periods of time upon a shrimp substrate. It was interesting to note although S. senftenberg 775 W is considered the most heat resistant strain, in this study it appeared to be the most labile strain when subjected to the freezing process.

Table 1.--Biochemical characteristics of three Salmonella species used in this study*.

Test or substrate	Salmonella		
	montevideo	oranienburg	senftenberg 775 W
Urease	-	-	-
Lysine decarboxylase	Alk	Alk	Alk
Phenol red dulcitol	AG	AG	AG
Phenol red mannitol	AG	AG	AG
Phenol red adonitol	-	-	-
Malonate broth	-	-	-
Indole test	-	-	-
Phenylalanine agar	-	-	-
K C N broth	NG	NG	NG
Methyl-red test	+	+	+
Voges-Proskauer test	-	-	-

*All tests according to FDA procedures
- indicates growth but no reaction
Alk indicates alkaline reaction
AG indicates acid and gas
NG indicates no growth

Table 2.--Additional biochemical tests of three Salmonella species used in this study.

Substrate	Salmonella		
	montevideo	oranienburg	senftenberg 775 W
TSI - slant	Alk	Alk	Alk
TSI - butt	AG	AG	AG
TSI - H_2S	+	+	-
Simmon's Citrate	+	+	+
Phenol red lactose	-	-	-
Phenol red sucrose	-	-	-
Phenol red maltose	AG	AG	AG
Phenol red glucose	AG	AG	AG
Phenol red fructose	AG	AG	AG
Phenol red mannose	AG	AG	AG
Phenol red galactose	AG	AG	AG
Phenol red xylose	AG	AG	AG
Phenol red raffinose	-	-	-
Phenol red trehalose	AG	AG	AG
Phenol red arabinose	AG	AG	AG
Phenol red sorbitol	AG	AG	AG
Phenol red inositol	-	-	-
Phenol red glycerol	-	-	-
Phenol red inulin	-	-	-

Alk indicates alkaline reaction
AG indicates acid and gas

Table 3.--Serological characteristics of three Salmonella species.

	Salmonella		
	montevideo	oranienburg	senftenberg 775 W
Somatic antigen			
1	-	-	+
3	-	-	+
6	+	+	-
7	+	+	-
19	-	-	+
Flagellar antigen			
g	+	+	-
m	+	+	-
s	+	-	+
t	-	+	+

Table 4.--The indigenous bacterial flora of the various sizes of shrimp.

Sizes							
21-25/1b	Average	36-40/1b	Average	51-60/1b	Average	Over 100/1b	Average
620,000		360,000		430,000		10,200,000	
630,000		300,000		360,000		9,400,000	
290,000		380,000		520,000		7,500,000	
360,000		900,000		400,000		8,200,000	
740,000	516,000	280,000	593,000	780,000	515,000	7,000,000	6,862,000
650,000		1,200,000		650,000		6,400,000	
390,000		850,000		430,000		2,700,000	
450,000		475,000		550,000		3,500,000	

Table 5.--The bacterial flora of the various sizes of shrimp after soaking for 30 minutes in tap water.

	Sizes of shrimp			
	21-25/lb	36-40/lb	51-60/lb	Over 100/lb
Salmonella senftenberg	340,000	770,000	236,000	370,000
	1,190,000	1,490,000	164,000	910,000
	450,000	480,000	360,000	750,000
Salmonella montevideo	210,000	1,210,000	340,000	490,000
	1,400,000	680,000	405,000	820,000
	700,000	650,000	420,000	780,000
Salmonella oranienburg	350,000	200,000	258,000	725,000
	1,250,000	380,000	265,000	950,000
	390,000	375,000	475,000	620,000

Table 6.--The bacterial flora of the various sizes of shrimp after soaking for 30 minutes in 2% sodium chloride

	Sizes of shrimp			
	21-25/lb	36-40/lb	51-60/lb	Over 100/lb
Salmonella senftenberg	382,000	260,000	164,000	478,000
	640,000	610,000	660,000	320,000
	435,000	510,000	350,000	380,000
Salmonella montevideo	130,000	210,000	360,000	678,000
	1,220,000	570,000	2,080,000	1,400,000
	890,000	410,000	1,200,000	890,000
Salmonella oranienburg	350,000	600,000	1,750,000	320,000
	410,000	650,000	800,000	1,200,000
	750,000	860,000	490,000	775,000

Table 7.--The bacterial flora of the various sizes of shrimp after soaking for 30 minutes in 2% NaCl plus 4% TPP.

	Sizes of shrimp			
	21-25/lb	36-40/lb	51-60/lb	Over 100/lb
Salmonella senftenberg	390,000	200,000	185,000	554,000
	710,000	144,000	430,000	830,000
	690,000	410,000	625,000	1,350,000
Salmonella montevideo	150,000	275,000	750,000	375,000
	620,000	260,000	290,000	580,000
	410,000	380,000	510,000	890,000
Salmonella oranienburg	280,000	185,000	680,000	123,000
	380,000	510,000	940,000	1,400,000
	740,000	625,000	1,310,000	780,000

Table 7A.--Survival rates of Salmonella organisms in processed shrimp.

	Days of storage							
	0	30	0	30	0	30	0	30
	21-25/lb		36-40/lb		51-60/lb		Over 100/lb	
Salmonella senftenberg	NG	NG	NG	NG	NG	NG	NG	NG
Salmonella montevideo	NG	NG	NG	NG	NG	NG	NG	NG
Salmonella oranienburg	NG	NG	NG	NG	NG	NG	NG	NG

NG indicates no growth

Table 8.--Survival rates during storage of Salmonella organisms in non-processed shrimp.

	ACBS	Days of storage					
		0	1	3	5	10	30
		21-25/lb					
Salmonella senftenberg	0	30,000	4,100	3,100	1,410	260	0
	0	65,000	4,900	3,900	1,530	230	0
	0	79,000	5,450	3,700	1,200	310	0
Salmonella montevideo	0	56,000	11,400	9,900	5,100	1,790	440
	0	47,000	12,300	7,500	7,000	2,100	360
	0	39,000	10,500	8,100	6,900	1,900	410
Salmonella oranienburg	0	51,000	34,000	7,900	3,200	1,540	240
	0	48,000	22,000	10,300	2,300	1,790	210
	0	62,000	24,500	11,200	3,700	1,200	320

ACBS indicates after cooking but before soaking

Table 9.--Survival rates during storage of Salmonella organisms in non-processed shrimp.

	ACBS	Days of storage					
		0	1	3	5	10	30
		36-40/lb					
Salmonella senftenberg	0	32,000	4,100	2,800	2,200	510	0
	0	29,000	4,900	3,400	2,800	360	0
	0	34,000	3,750	2,900	2,500	475	0
Salmonella montevideo	0	58,000	11,400	11,100	5,600	3,900	1,510
	0	65,000	12,300	9,300	4,800	4,600	1,890
	0	52,000	14,200	9,800	5,100	3,750	1,250
Salmonella oranienburg	0	59,000	34,000	21,000	10,800	3,500	1,250
	0	53,000	29,000	25,000	12,300	5,100	1,140
	0	48,000	32,500	23,800	12,000	4,900	900

ACBS indicates after cooking but before soaking

Table 10.--Survival rates during storage of Salmonella organisms in non-processed shrimp.

	ACBS	Days of storage					
		0	1	3	5	10	30
		51-60/lb					
Salmonella senftenberg	0	22,000	4,400	2,100	3,000	1,100	200
	0	28,000	4,000	2,900	2,500	930	175
	0	35,500	7,200	4,100	3,900	1,750	320
Salmonella montevideo	0	53,000	46,500	30,300	16,300	7,900	2,700
	0	55,000	42,700	24,500	12,400	6,600	3,500
	0	62,000	49,500	30,500	14,700	9,300	7,400
Salmonella oranienburg	0	54,000	18,500	17,400	14,900	5,900	1,900
	0	44,000	16,100	15,400	12,900	4,700	1,710
	0	52,000	14,200	14,700	13,000	7,200	3,400

ACBS indicates after cooking but before soaking

TRACE ELEMENTS FOUND IN VARIOUS SPECIES OF SHRIMP HARVESTED FROM SELECTED AREAS

ABSTRACT

The levels of 14 trace elements (Hg, Pb, Cd, As, Se, Ag, Cu, Zn, Cr, Ni, Mo, V, Mn and Sn) are reported for 7 species of shrimp (white, brown, pink, pink northern, sidestripe, royal red and ocean) harvested from 1 to 6 sites in each of 5 areas of the coastal United States (N. and S. Atlantic, Gulf of Mexico, Pacific NW and Alaska). In addition to the analytical data, which includes mean values with standard deviations and ranges, for each element, physical and catch data on the shrimp are also summarized. A comparison is made with previously published information on trace element levels in shrimp.

TRACE ELEMENTS FOUND IN VARIOUS SPECIES OF SHRIMP HARVESTED FROM SELECTED AREAS

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Since the mercury scare in Canada in late 1970, there has been a growing concern among consumers, fish processors, fishermen and marine scientists, regarding the impact that possible increases in the concentrations of trace elements in the marine environment and the fishery resource might have on public health, the viability of some marine species and, consequently, the future of the fishing industry.

Therefore, Dr. Robert M. White, Administrator of the National Oceanic and Atmospheric Administration (NOAA), in 1971, directed the National Marine Fisheries Service (NMFS) to undertake a series of surveys of present day trace element levels in fish and shellfish so as to provide baseline elemental composition data on the United States' seafood supply. Most of this research has been carried out through surveys conducted by the Southeast Utilization Research Center (SURC), College Park, Md., and the Pacific Utilization Research Center (PURC), Seattle, Washington. Several surveys have now been completed or are nearing completion. This report covers data so far obtained on fourteen trace elements in various species of shrimp: mercury (Hg), lead (Pb), cadmium (Cd), arsenic (As), selenium (Se), chromium (Cr), silver (Ag), copper (Cu), zinc

(Zn), molybdenum (Mo), nickel (Ni), vanadium (V), manganese (Mn), and tin (Sn). Also included is detailed information on species of shrimp, dates and areas of harvest, size and number of shrimp per analytical sample, the NMFS Laboratories responsible for providing the samples, and miscellaneous data supplied to SURC that could be of value to researchers and others utilizing this report.

Shrimp data from three separate surveys are contained in this report: (1) a product survey, Zook et al, 1976; (2) the Microconstituent Resource Survey, on which an interim report has been issued, SURC Staff, 1975; and (3) a literature survey, Sidwell et al, 1976. Additional unpublished data from the complete Resource Survey also are included.

Mercury, Pb, Cd, Cr and As data from seven sets of shrimp, representing four known species caught in domestic waters, two unknown species imported from Asia and Mexico and a set of mixed species from Alaskan waters, were included among the 29 species of seafoods sampled in the product survey. In general, samples for this survey were limited to ten (each sample containing a minimun of 1 lb edible flesh) from a single area of catch or country of import. Proximate data (protein, moisture, ash, and crude fat), total fat, and the fatty acid profiles on the identical samples were published by Sidwell et al, 1973, and Bonnet et al, 1974.

The Microconstituent Resource Survey includes a much more comprehensive survey of shrimp caught in the coastal waters of the United States. For purposes of data collection and assembly of results, the coastal waters of the United States were divided into

seven areas as shown in Figure 1. Shrimp were harvested by nineteen NMFS laboratories, including SURC and PURC. All samples were prepared for analysis at the College Park and Seattle facilities.

According to the original sampling plan, collectors were to provide enough shrimp of a particular species from each area to yield four sets of ten 2-lb samples of edible tails, each set to be from a distinct site. The shrimp actually collected in the Resource Survey, together with those from other sources, are given in Table 1. A resumé of the magnitude of the surveys reviewed in this report is given below:

Species samples	7
Total samples	406
Total analyses	7,308
Minimum shrimp/sample	50

Shrimp were received as raw tails. Sample preparation consisted of peeling each shrimp, deveining the large animals but not the small, grinding the entire 2 lbs, more or less, of edible tail in a Hobart[1/] Silent Cutter to a uniform paste or blend. Ground samples were then packed in three or more 4-oz plastic containers, frozen and held at -10° until analyzed. All 14 elements were analyzed by atomic absorption spectrophotometry. The bulk of the

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1/ Reference to trade names does not imply endorsement by the National Marine Fisheries Service.

analyses was performed under contract 2/ 3/. Mercury levels in product survey shrimp were determined at SURC. Details of the experimental procedures have been described previously, Zook et al, 1976, SURC/NMFS Staff, 1975.

Tables 2 through 10 show the number of analytical samples per set, all available historical information on the samples, the mean, standard deviation of the mean and the range of each element tested in parts per million (ppm), by individual species and area of catch, under each site of the catch area. Table 11 summarizes the available data on imported shrimp and the trace element levels in shrimp reported by Sidwell et al, 1976, in a literature search covering more than 225 references from 1896 through the first half of 1975.

Figures 2 through 15 graphically present the mean values of the individual element data for all species, in all sites and areas, except for the data presented in Table 11.

In general, the individual element data within areas are more uniform than was found with mollusk data (Seagran, 1976), taken from the Resource Survey. Mean mercury values (Fig. 2) were all below the mean mercury value derived from 14 literature references (0.15 ppm). Only two sets, one of white shrimp from Cape Canaveral, Florida (0.13 ppm) and one of brown shrimp from the western side of the Gulf of Mexico (0.12 ppm) had mean Hg values greater than 0.08 ppm.

2/ Analytical Consulting Services, Inc., Kensington, Md., for product survey samples.

3/ Omni Research, Inc., San German, P.R., for Resource Survey samples.

Mean lead values (Fig. 3) ranged from a low of 0.31 ppm in the same brown shrimp sample from the Gulf of Mexico to a high of 0.89 ppm in pink northern shrimp from Alaska. Twenty-six of the 33 mean lead values were above the 0.46 ppm Pb reported from four literature values.

The eight literature values found for cadmium averaged 0.09 ppm Cd, the range of mean cadmium values (Fig. 4) in the two surveys was from a low of 0.03 ppm Cd in white shrimp harvested in Galveston Bay in 1972 to a high of 0.20 ppm in the pink northern shrimp from Alaska, the same animals with the highest lead content. Seventeen of the mean cadmium values were above 0.09 ppm Cd and twelve of these were among the 13 pink northern shrimp from Alaska and the North Atlantic.

Literature values for arsenic in shrimp taken from eight references, had a mean and standard deviation of 8.71 ± 7.03 ppm. The mean arsenic values (Fig. 5) in the two NMFS surveys ranged from a low of 2.24 ppm in white shrimp from Mobile Bay, Alabama to a high of 16.91 in one set of pink northern shrimp from the North Atlantic. Royal red shrimp were in a class by themselves with 51.75 ± 24.54 ppm As. Only one set was tested so it is impossible to know if this is a true species difference.

Only two literature references listed Se values (1.24 ± 0.92). Selenium was analyzed only in the Resource Survey samples, ranging from 0.26 ppm in sidestripe shrimp from Alaska to 0.86 ppm in brown shrimp from the Gulf side of Florida (Fig. 6).

No literature values were found for silver. With the exception of pink northern shrimp from Alaska and the North Atlantic all Resource Survey values were below 0.1 ppm (Fig. 7). Of the seven sets of pink northern shrimp analyzed, five were above 0.10 ppm while all seven sets were below 0.18 ppm.

Seven chromium literature values were found to average 0.07 ± 0.05 ppm. Only three sets of shrimp, two of white shrimp from the product survey and one of brown shrimp from the Resource Survey were below this mean (Fig. 8). All others ranged from 0.10 ppm to 0.36 ppm, both from sets of brown shrimp caught in the Gulf of Mexico. These values may truly represent the levels of chromium in shrimp. However, the possibility of some contamination from the Ni-Cr wire in the heating coils of the muffle furnace used for ashing the samples prior to analysis, or the scratches and chips found in the bowls and knives of the Hobart Silent Cutters used in sample preparation, cannot be completely eliminated. A controlled experiment with shrimp samples eliminating any known sources of chromium contamination should be considered in the near future to resolve this question.

Nine copper values found in the literature gave a mean and standard deviation of 4.64 ± 3.54 ppm Cu. Only two sets of shrimp from the Resource Survey (Fig. 9) (the only survey reporting copper values) had mean copper values above 4.64 ppm Cu; these were a set of pink northern shrimp from the North Atlantic (6.28 ppm Cu) and a set of sidestripe shrimp from Alaska (5.53 ppm Cu).

Mean zinc values taken from 7 references were 19.29 ± 11.06 ppm. No zinc values (Fig. 10) in the Resource Survey were above 19 ppm Zn, the highest mean value being 15.51 ppm Zn in one set of sidestripe shrimp from Alaska, the lowest being 8.88 ppm Zn in a set of pink northern shrimp from the North Atlantic.

Nickel in shrimp has been reported in only one literature reference. All values (Fig. 11) found in the Resource Survey were considerably higher than this single value of 0.03 ppm Ni. The values ranged from a low of 0.18 ppm Ni to a high of 0.71 ppm Ni. Again, as with the chromium values, a check for possible accidental nickel contamination in the preparation or analyses of the shrimp samples should be investigated.

Molybdenum values in the literature were found to be very sparse, with one reported value of 0.03 ppm. The Mo values for shrimp shown in Fig. 12 are also very low. The mean values range from zero (not detected) to 0.26 ppm, both extreme values being found in white shrimp harvested from the Gulf of Mexico.

A single vanadium value of 1.90 ppm was found in the literature. None of the shrimp tested in the Resource Survey (Fig. 13) approached this level. The highest mean value found was 0.82 ppm V in pink northern shrimp caught in the North Atlantic. Fifteen of the 29 mean values reported were below 0.10 ppm V.

Manganese (Fig. 14) varied considerably among the shrimp analyzed in the Resource Survey. Eight sets of white shrimp had mean values from 0.39 to 1.20 ppm Mn, and five sets of brown shrimp varied from 0.37 to 1.31 ppm Mn. The five sets of pink shrimp

caught in the Gulf of Mexico ranged from 0.17 to 0.74 ppm Mn. The remaining 11 sets of 4 species of shrimp ranged from a low of 0.25 ppm Mn in the ocean shrimp to a high of 0.50 ppm Mn in a set of pink northern shrimp caught in the North Atlantic. Four literature values for Mn gave a mean and standard deviation of 0.73 ± 0.84.

No tin values for shrimp have been reported in the literature. In general, the Sn levels found in the resource survey were fairly uniform (Fig. 15), ranging only from 0.50 to 1.06 ppm Sn.

Values for 14 trace elements in 7 species of shrimp, harvested from 1 to 6 sites in 5 areas of the coastal United States, have been presented. Except for Cu and Ni, trace element levels are similar in magnitude to the relatively few values found in the published literature of the past 75 years.

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Table 1 -- Variables Covered in Shrimp Trace Element Report

Shrimp Name			
Common	Scientific	Area of Catch	Sites/Area
White	Panaeus setiferris	Gulf of Mexico	6
		South Atlantic	4
Brown	Panaeus aztecus	Gulf of Mexico	7
Pink	Panaeus duorarum	Gulf of Mexico	5
Pink Northern	Pandalus borealis	Alaska	2
		North Atlantic	6
Sidestripe	Pandalopsis dispar	Alaska	2
Royal Red	Hymenopanaes robustus	South Atlantic	1
Ocean	Pandalus jordani	Pacific Northwest	1
Imported	Species unknown	Mexico	3
		Asia	3
Mixed	(Pandalus borealis, 1 (Pandalopsis dispar 8 (Pandalus platyeros, 9	Alaska	1
Mixed	Unknown 1 - 12 references		?

Table 2 --Trace element content, ppm, found in raw, peeled tails of white shrimp (Panaeus setiferris) from the Gulf of Mexico

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10 Apalachicola Bay, Fl. 12/16/73[a]/	0.00 ± 0.01[h]/	0.53 ± 0.15	0.08 ± 0.01	2.84 ± 1.10	0.63 ± 0.24	0.05 ± 0.01	0.23 ± 0.11	1.90 ± 0.16	9.35 ± 2.16	0.19 ± 0.04	0.03 ± 0.07	0.04 ± 0.12	0.55 ± 0.13	0.54 ± 0.18
8 ± 1g[b]/ 7 - 10g[c]/ 43 cm[d]/ 102 ± 9[e]/ 1021[f]/ Panama City, Fl.[g]/	0.00 - 0.02[i]/	0.28 - 0.81	0.06 - 0.09	1.53 - 4.50	0.15 - 0.94	0.02 - 0.06	0.13 - 0.44	1.66 - 2.23	3.44 - 10.63	0.13 - 0.26	0.00 - 0.19	0.00 - 0.38	0.31 - 0.72	0.23 - 0.91
10 Mobile Bay, Ala., I 11/28/72[a]/	0.02 ± 0.02	0.57 ± 0.38	0.06 ± 0.05	2.50 ± 1.92	0.56 ± 0.17	0.04 ± 0.02	0.24 ± 0.15	1.75 ± 0.29	10.85 ± 3.93	0.25 ± 0.10	0.26 ± 0.34	0.07 ± 0.10	1.07 ± 0.27	0.64 ± 0.17
21 ± 1g (n=2)[b]/ 86 ± 3 (n=2)[c]/ Miami, Fl.[g]/	0.00 - 0.06	0.23 - 1.25	0.00 - 0.17	0.47 - 6.90	0.03 - 0.87	0.00 - 0.06	0.05 - 0.48	1.43 - 2.36	6.00 - 20.62	0.14 - 0.45	0.00 - 0.88	0.00 - 0.31	0.76 - 1.57	0.45 - 0.88
10 Mobile Bay, Ala.,II 11/ 6 /73[a]/	0.01 ± 0.03	0.72 ± 0.51	0.10 ± 0.04	2.24 ± 1.51	0.52 ± 0.16	0.03 ± 0.01	0.14 ± 0.08	2.80 ± 0.43	12.47 ± 1.93	0.22 ± 0.06	0.03 ± 0.08	0.00	0.41 ± 0.13	0.57 ± 0.14
26 ± 1g[b]/ 23 - 28g[c]/ 71 ± 4[e]/ 709[f]/ Miami, Fl.[g]/	0.00 - 0.10	0.19 - 1.88	0.05 - 0.18	0.45 - 4.50	0.25 - 0.78	0.01 - 0.04	0.00 - 0.32	2.28 - 3.88	9.63 - 16.31	0.10 - 0.30	0.00 - 0.25	0.00	0.26 - 0.73	0.38 - 0.83

Table 2 --Trace element content, ppm, found in raw, peeled tails of white shrimp (Panaeus setiferris) from the Gulf of Mexico, continued.

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10 Mississippi Coast[d]/ 6/17/71[a]/ 10 ± 1g[b]/ Pascagoula, Miss.[e]/	0.07 ± 0.03 0.04 - 0.12	0.56 ± 0.49 0.18 - 1.44	0.08 ± 0.02 0.06 - 0.11	3.00 ± 0.80 1.70 - 4.40			0.05 ± 0.01 0.00 - 0.11							
10 28°53' N 95°10' W,I 6/13/72[a]/ Galveston, Tex.[g]/	0.06 ± 0.04 0.01 - 0.12	0.42 ± 0.30 0.00 - 1.13	0.03 ± 0.03 0.00 - 0.07	6.83 ± 2.53 3.60 - 11.90	0.75 ± 0.13 0.43 - 0.87	0.04 ± 0.06 0.00 - 0.18	0.29 ± 0.20 0.02 - 0.68	3.99 ± 0.82 2.66 - 5.50	13.77 ± 3.95 8.36 - 21.43	0.26 ± 0.22 0.00 - 0.75	0.12 ± 0.25 0.00 - 0.75	0.16 ± 0.28 0.00 - 0.68	0.51 ± 0.09 0.38 - 0.69	0.62 ± 0.20 0.39 - 1.03
9 28°45' N 95°13' W,II 7/25 - 8/15/73[a]/ 25 ± 14g[b]/ 11-47g[c]/ 75 ± 32[a]/ 671[f]/ Calveston, Tex.[g]/	0.08 ± 0.12 0.00 - 0.33	0.73 ± 0.20 0.43 - 1.00	0.14 ± 0.10 0.08 - 0.39	6.05 ± 2.97 2.60 - 10.00	0.58 ± 0.16 0.34 - 0.80	0.05 ± 0.02 0.02 - 0.09	0.23 ± 0.08 0.13 - 0.31	4.51 ± 2.31 1.99 - 8.81	13.26 ± 2.52 8.63 - 17.19	0.25 ± 0.10 0.12 - 0.41	0.00 0.00	0.11 ± 0.12 0.00 - 0.31	0.50 ± 0.19 0.31 - 0.94	0.74 ± 0.16 0.53 - 1.00

Table 2 -- Trace element content, ppm, found in raw, peeled tails of white shrimp (Panaeus setiferris) from the Gulf of Mexico, continued

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn

a/ Date of catch.
b/ Weight, mean with or without standard deviation.
c/ Weight range.
d/ Length, mean with or without standard deviation.
e/ Shrimp per sample with or without standard deviation.
f/ Total number shrimp in set.
g/ NMFS laboratory supplying samples.
h/ Element, mean ± standard deviation.
i/ Element, range
j/ References Sidwell, et al., 1973 and Zook, et al., 1976.

Table 3 --Trace element content, ppm, found in raw, peeled tails of white shrimp (Panaeus setiferris) from the South Atlantic

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10	0.08 ±	0.33 ±	0.05 ±	5.76 ±	0.60 ±	0.06 ±	0.13 ±	2.67 ±	11.38 ±	0.20 ±	0.03 ±	0.09 ±	0.39 ±	0.77 ±
Charleston, S. C.	0.10[e]	0.27	0.04	2.46	0.25	0.08	0.09	0.80	3.98	0.22	0.06	0.14	0.11	0.34
6/14/72[a]														
11g[b]	0.00 –	0.00 –	0.00 –	2.50 –	0.28 –	0.00 –	0.00 –	1.89 –	5.00 –	0.00 –	0.00 –	0.00 –	0.25 –	0.39 –
1.5 yr.[c]	0.31[f]	0.73	0.11	9.00	1.05	0.22	0.32	4.30	19.64	0.61	0.18	0.32	0.53	1.06
Beaufort, N. C.[d]														
10	0.08 ±	0.38 ±	0.05 ±	6.24 ±	0.62 ±	0.03 ±	0.18 ±	2.66 ±	11.75 ±	0.30 ±	0.11 ±	0.29 ±	1.20 ±	0.73 ±
Brunswick, Ga.	0.04	0.14	0.03	2.86	0.30	0.03	0.09	0.44	1.79	0.20	0.13	0.41	0.46	0.23
10/25/72[a]														
Miami, Fla.[d]	0.02 –	0.15 –	0.00 –	3.54 –	0.24 –	0.00 –	0.03 –	2.26 –	8.69 –	0.00 –	0.00 –	0.00 –	0.57 –	0.40 –
	0.14	0.59	0.10	12.63	1.11	0.08	0.31	3.80	13.93	0.64	0.37	1.25	1.86	1.06
10	0.03 ±	0.49 ±	0.07 ±	6.98 ±	0.74 ±	0.04 ±	0.20 ±	1.51 ±	10.35 ±	0.18 ±	0.04 ±	0.13 ±	0.39 ±	0.54 ±
1 mile SE Jekyll Isle, Ga.	0.03	0.31	0.11	3.05	0.35	0.07	0.13	0.58	2.59	0.06	0.05	0.20	0.06	0.11
6/1/72[a]	0.00 –	0.06 –	0.00 –	3.44 –	0.29 –	0.00 –	0.04 –	1.04 –	6.61 –	0.07 –	0.00 –	0.00 –	0.26 –	0.35 –
1.0 Yr.[c]	0.08	1.12.	0.34	11.69	1.37	0.21	0.44	2.87	14.51	0.28	0.11	0.63	0.46	0.70
Beaufort, N. C.[d]														
10	0.13 ±	0.47 ±	0.05 ±	4.60 ±			0.05 ±							
Within 5 miles	0.04	0.33	0.02	1.30			0.01							
Cape Canaveral, Fl.[g]														
6/28/71[a]	0.08 –	0.16 –	0.00 –	2.80 –			0.00 –							
Pascagoula, Miss.[d]	0.18	1.20	0.08	7.70			0.11							

Table 3 --Trace element content, ppm, found in raw, peeled tails of white shrimp (Panaeus setiferris) from the South Atlantic, continued.

a/ Date of catch.
b/ Weight, mean with or without standard deviation.
c/ Age class.
d/ NMFS Laboratory supplying samples.
e/ Element, mean ± standard deviation.
f/ Element, range.
g/ References: Sidwell, et al., 1973 and Zook, et al, 1976.

Table 4 --Trace element content, ppm, found in raw, peeled tails of Brown Shrimp (Penaeus aztecus) from the Gulf of Mexico, Cont.

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10 Mobile Bay, Ala. 11/6/73[a]/ 13.1 ± 0.6g[b]/ 12 - 14g[c]/ 137 ± 7[f]/ 127 - 151[g]/ 1234[h]/ Miami, Fla.[i]/	0.02 ± 0.05 0.00 - 0.15	0.57 ± 0.30 0.25 - 1.16	0.09 ± 0.03 0.06 - 0.14	2.43 ± 1.16 0.66 - 4.00	0.52 ± 0.19 0.23 - 0.93	0.03 ± 0.01 0.03 - 0.31	0.19 ± 0.06 0.13 - 0.31	2.48 ± 0.67 1.54 - 3.94	11.08 ± 3.35 6.50 - 14.94	0.24 ± 0.07 0.16 - 0.38	0.00 0.00	0.08 ± 0.12 0.00 - 0.31	0.73 ± 0.19 0.48 - 0.90	0.59 ± 0.14 0.31 - 0.75
10 Mississippi Sound 11/28/71[a]/ 16.8 ± 0.6 g[b]/ 15.9 - 17.5 g[c]/ 100[f]/ 1000[h]/ Miami, Fla.[i]/	0.04 ± 0.09 0.00 - 0.29	0.53 ± 0.30 0.20 - 1.06	0.05 ± 0.03 0.00 - 0.09	3.31 ± 1.63 1.49 - 6.26	0.59 ± 0.11 0.43 - 0.73	0.04 ± 0.02 0.00 - 0.07	0.06 ± 0.69 0.06 - 0.69	2.30 ± 0.30 1.84 - 2.69	11.53 ± 1.42 8.75 - 13.21	0.35 ± 0.17 0.14 - 0.61	0.06 ± 0.09 0.00 - 0.25	0.08 ± 0.13 0.00 - 0.36	1.31 ± 0.13 1.07 - 1.46	0.57 ± 0.13 0.38 - 0.82
10 SE Sabine Pass, Tx[1]/ 8/11/71[a]/ Pascagoula, Ms[i]/	0.07 ± 0.02 0.04 - 0.10	0.48 ± 0.36 0.10 - 1.19	0.05 ± 0.04 0.00 - 0.12	4.40 ± 0.70 3.10 - 5.20			0.10 ± 0.06 0.00 - 0.19							

Table 4 --Trace element content, ppm, found in raw, peeled tails of brown shrimp (Penaeus aztecus) from the Gulf of Mexico.

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10 St. Andrews Bay, Fla. 7/14-18/72[a]	0.02 ± 0.02[j]	0.50 ± 0.26	0.07 ± 0.06	6.76 ± 3.58	0.58 ± 0.16	0.02 ± 0.02	0.19 ± 0.13	4.32 ± 0.93	11.68 ± 3.20	0.19 ± 0.16	0.11 ± 0.18	0.10 ± 0.11	0.58 ± 0.30	0.72 ± 0.41
21 ± 3g[b] 16 - 25g[c] 13 ± 0.5 cm[d] 13 - 14 cm[e] 62 ± 13[f] 50 - 93[g] 618[h] Panama City, Fla.[i]	0.00 - 0.05[k]	0.13 - 1.03	0.00 - 0.16	1.30 - 12.15	0.28 - 0.80	0.00 - 0.05	0.03 - 0.36	2.87 - 5.71	6.68 - 16.34	0.00 - 0.50	0.00 - 0.44	0.00 - 0.25	0.20 - 1.21	0.25- 1.75
3 West Bay, Fla. 10/9/73[a]	0.00	0.52 ± 0.03	0.07 ± 0.02	4.14 ± 1.40	0.86 ± 0.25	0.04 ± 0.02	0.36 ± 0.24	3.87 ± 0.74	10.87 ± 2.98	0.18 ± 0.09	0.08 ± 0.14	0.00	0.49 ± 0.14	0.65 ± 0.07
16 ± 1g[b] 15 - 17g[c] 11.6 ± 0.006 cm[d] 11.1 - 12.2 cm[e] 97 ± 13[f] 84 - 110[g] 290[h] Panama City, Fla.[i]	0.00	0.50 - 0.56	0.05 - 0.09	3.15 - 5.13	0.57 - 1.01	0.02 - 0.06	0.19 - 0.63	3.02 - 4.38	7.50 - 13.18	0.08 - 0.26	0.00 - 0.25	0.00	0.34 - 0.62	0.63 - 0.66

Table 4 -- Trace element content, ppm, found in raw, peeled tails of Brown Shrimp (Penaeus aztecus) from the Gulf of Mexico, Cont.

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10 28°25'N 94°44'W, I 6/13/72[a]/	0.12 ± 0.04	0.31 ± 0.31	0.04 ± 0.03	14.38 ± 9.03	0.64 ± 0.14	0.04 ± 0.05	0.20 ± 0.15	3.97 ± 0.65	13.54 ± 6.94	0.26 ± 0.20	0.04 ± 0.07	0.21 ± 0.25	0.37 ± 0.20	0.63 ± 0.23
Galveston, Texas[i]/	0.07 - 0.21	0.00 - 1.09	0.00 - 0.08	8.80 - 32.50	0.47 - 0.86	0.00 - 0.18	0.00 - 0.44	2.95 - 5.29	4.93 - 31.07	0.09 - 0.68	0.00 - 0.19	0.00 - 0.60	0.18 - 0.71	0.42 - 1.15
10 28°17 - 45'N 94°49' - 95°13'W, II 8/15-16/73[a]/	0.04 ± 0.04	0.56 ± 0.22	0.09 ± 0.03	7.52 ± 3.32	0.59 ± 0.22	0.04 ± 0.02	0.30 ± 0.16	3.69 ± 0.88	11.57 2.25	0.26 ± 0.08	0.04 ± 0.04	0.17 ± 0.13	0.49 ± 0.16	0.57 ± 0.21
20.2 ± 8.7g[b]/ 14 - 28g[c]/ 71 ± 20 [f]/ 714[h]/ Galveston, Texas[i]/	0.00 - 0.10	0.36 - 1.06	0.07 - 0.14	1.30 - 13.25	0.32 - 0.91	0.02 - 0.06	0.13 - 0.63	2.31 - 4.72	6.56 - 13.63	0.13 - 0.35	0.00 - 0.10	0.00 - 0.31	0.28 - 0.74	0.38 - 0.78

Table 4 — Trace element content, ppm, found in raw, peeled tails of brown shrimp (Penaeus aztecus) from the Gulf of Mexico, Cont.

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn

a/ Date of catch.
b/ Weight, mean with or without standard deviation.
c/ Weight, range.
d/ Length, mean with or without standard deviation.
e/ Length, range.
f/ Number shrimp/sample with or without standard deviation.
g/ Range of shrimp/sample.
h/ Total number shrimp in set.
i/ NMFS laboratory supplying sample.
j/ Element, mean ± standard deviation.
k/ Element, range.
l/ Sidwell et al 1973, Zook et al 1976.

Table 5 — Trace element content, ppm, in raw, peeled tails of pink shrimp (Penaeus du rarum) from the Gulf of Mexico, continued

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10 27°2'N 82°42'W 11/27/72[a] 3.1g[b] 293[f] 2930[h] St. Petersburg, Fla.[i]	0.06 ± 0.07 0.00 – 0.21	0.59 ± 0.36 0.25 – 1.34	0.14 ± 0.05 0.05 – 0.21	7.22 ± 2.22 4.35 – 12.20	0.32 ± 0.14 0.09 – 0.53	0.09 ± 0.04 0.03 – 0.15	0.23 ± 0.24 0.08 – 0.87	2.49 ± 1.10 0.26 – 4.12	9.41 ± 2.15 3.78 – 11.81	0.32 ± 0.09 0.16 – 0.43	0.07 ± 0.06 0.00 – 0.16	0.17 ± 0.21 0.00 – 0.65	0.17 ± 0.06 0.09 – 0.26	0.77 ± 0.37 0.31 – 1.37
10 Dry Tortugas 24°38'N 82°55'W 11/15/72[a] Miami, Fla.[i]	0.01 ± 0.01 0.00 – 0.03	0.57 ± 0.34 0.31 – 1.33	0.14 ± 0.08 0.07 – 0.32	3.72 ± 1.70 1.09 – 6.77	0.48 ± 0.14 0.27 – 0.66	0.03 ± 0.02 0.00 – 0.07	0.19 ± 0.10 0.05 – 0.37	1.19 ± 0.47 0.61 – 1.91	11.41 ± 4.22 7.81 – 14.85	0.19 ± 0.09 0.00 – 0.31	0.03 ± 0.06 0.00 – 0.20	0.10 ± 0.12 0.00 – 0.35	0.18 ± 0.07 0.13 – 0.29	0.65 ± 0.16 0.45 – 0.88
10 Key West, Fla. 10/1/73[a] 7.1 ± 0.7g (n=7)[b] 6 – 8g (n=7)[c] 201 ± 77[f] 157 – 349[g] 2011[h] Miami, Fla.[i]	0.01 ± 0.03 0.00 – 0.08	0.66 ± 0.19 0.38 – 0.94	0.17 ± 0.02 0.14 – 0.21	4.88 ± 1.91 1.35 – 6.93	0.46 ± 0.25 0.21 – 1.10	0.05 ± 0.01 0.04 – 0.06	0.19 ± 0.05 0.13 – 0.29	1.47 ± 0.70 0.74 – 2.91	10.33 ± 2.63 7.88 – 16.25	0.29 ± 0.09 0.21 – 0.48	0.10 ± 0.10 0.00 – 0.25	0.21 ± 0.18 0.00 – 0.63	0.22 ± 0.05 0.15 – 0.33	0.70 ± 0.10 0.54 – 0.88

Table 5 — Trace element content, ppm, in raw, peeled tails of pink shrimp (Penaeus du rarum) from the Gulf of Mexico

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10	0.02 ±	0.72 ±	0.11±	5.38 ±	0.58 ±	0.04 ±	0.21 ±	3.85 ±	11.43 ±	0.41 ±	0.18 ±	0.41 ±	0.63 ±	0.91 ±
St. Andrews Bay, Fl	0.02[j]	0.29	0.07	2.28	0.25	0.04	0.12	1.06	2.42	0.29	0.14	0.43	0.30	0.40
7/18 - 10/4/72[a]	0.00 -	0.40 -	0.00 -	3.10 -	0.22 -	0.00 -	0.08 -	2.86 -	9.64 -	0.17 -	0.00 -	0.00 -	0.31 -	0.35 -
5.0 ± 4.4g[b]	0.04[k]	1.44	0.25	8.46	1.18	0.11	0.49	6.09	13.97	1.21	0.38	1.19	1.22	1.75
2.7 - 17.3g [c]														
9.7 ± 1.2 cm[d]														
8.5 - 12.5 cm[e]														
106 ± 39[f]														
61 - 170[g]														
1061[h]														
Panama City, Fla.[i]														
10														
West Bay, Fla.	0.07 ±	0.48 ±	0.10 ±	3.22 ±	0.71 ±	0.05 ±	0.26 ±	2.56 ±	10.73 ±	0.35 ±	0.01 ±	0.03 ±	0.74 ±	0.64 ±
10/9 - 15/73[a]	0.12	0.06	0.01	1.01	0.13	0.02	0.14	1.01	2.14	0.07	0.03	0.05	0.29	0.15
8.7 ± 1.1g[b]	0.00 -	0.44 -	0.08 -	0.70 -	0.55 -	0.02 -	0.13 -	0.43 -	5.06 -	0.24 -	0.00 -	0.00	0.12 -	0.38 -
8 - 11g[c]	0.30	0.63	0.12	4.23	0.95	0.07	0.50	3.81	12.88	0.48	0.08	0.13	1.07	0.75
9.7 ± 0.1 cm[d]														
9.6 - 9.9 cm[e]														
166 ± 14[f]														
139 - 173[g]														
1662[h]														
Panama City, Fla.[i]														

Table 5 — Trace element content, ppm, in raw, peeled tails of pink shrimp (Penaeus duorarum) from the Gulf of Mexico, continued

No. of samples, site of c[illegible], sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn

a/ Date of capture.
b/ Weight with or without standard deviation.
c/ Weight, range.
d/ Length with or without standard deviation.
e/ Length, range.
f/ Number shrimp/sample with or without standard deviation.
g/ Range of shrimp/sample.
h/ Total number shrimp in lot.
i/ NMFS Laboratory supplying samples.
j/ Element, mean ± standard deviation.
k/ Element, range.

Table 6 — Trace Element content, ppm, in raw, peeled tails of pink northern shrimp (Pandalus borealis) from Alaska.

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10 59°5'N 153°45'W 5/8/73[a]/ Auke Bay, Alaska[b]/	0.01 ± 0.02[c]/ 0.00 – 0.05[d]/	0.55 ± 0.19 0.31 – 0.88	0.10 ± 0.02 0.08 – 0.13	3.35 ± 1.49 1.00 – 6.15	0.26 ± 0.15 0.03 – 0.64	0.09 ± 0.02 0.06 – 0.12	0.12 ± 0.07 0.00 – 0.21	2.43 ± 0.74 1.63 – 4.31	10.71 ± 2.27 9.13 – 16.88	0.28 ± 0.09 0.14 – 0.41	0.03 ± 0.05 0.00 – 0.16	0.06 ± 0.09 0.00 – 0.19	0.31 ± 0.07 0.17 – 0.43	0.70 ± 0.16 0.50 – 0.88
10 Frederick Sound 6/14/73[a]/ Auke Bay, Alaska[b]/	0.03 ± 0.04 0.00 – 0.11	0.89 ± 0.42 0.50 – 1.63	0.20 ± 0.05 0.11 – 0.27	8.89 ± 4.56 4.50– 17.60	0.47 ± 0.13 0.26 – 0.75	0.15 ± 0.04 0.06 – 0.20	0.18 ± 0.10 0.06 – 0.44	4.30 ± 0.96 3.63 – 6.88	12.40 ± 1.18 11.00 – 15.00	0.38 ± 0.15 0.22 – 0.58	0.03 ± 0.06 0.00 – 0.16	0.08 ± 0.14 0.00 – 0.41	0.46 ± 0.07 0.35 – 0.59	0.67 ± 0.71 0.50 – 0.81

a/ Date of capture
b/ NMFS Laboratory supplying sample
c/ Element, mean ± Standard Deviation
d/ Element, range

Table 7 — Trace element content, ppm, found in raw, peeled tails of pink northern shrimp (Pandalus borealis) from the North Atlantic

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10 Gulf of Maine[a] 4/25 - 9/13/71[b] Gloucester, Mass.[c]	0.06 ± 0.01[d] 0.04 - 0.08[e]	0.39 ± 0.15 0.10 - 0.61	0.07 ± 0.07 0.00 - 0.20	6.00± 1.90 3.60 - 9.40			0.10 ± 0.07 0.00 - 0.25							
1 44°40'W 66°30'W 11/16/72[b] Woods Hole, Mass.[c]	0.00	0.38	0.11	13.50	0.41	0.03	0.23	6.28	10.36	0.71	0.00	0.00	0.36	0.50
7 43°12'N 70°13'W, I 5/3 - 14/73[b] 3.1 ± 1.3g[f] 2 - 6g[g] 302[h] 2144[i] Woods Hole, Mass.[c]	0.03 ± 0.03 0.00 - 0.07	0.72 ± 0.31 0.38 - 1.31	0.16 ± 0.03 0.14 - 0.21	16.40 ± 4.50 10.63 - 25.00	0.54 ± 0.13 0.40 - 0.72	0.18 ± 0.03 0.13 - 0.21	0.25± 0.11 0.13 - 0.38	3.38 ± 0.89 2.69 - 5.31	11.23 ± 0.57 10.25 - 11.88	0.36 ± 0.09 0.21 - 0.44	0.09 ± 0.10 0.00 - 0.25	0.82 ± 0.33 0.38 - 1.32	0.50 ± 0.20 0.38 - 0.65	0.94 ± 0.19 0.63 - 1.19

Table 7 -- Trace element content, ppm, found in raw, peeled tails of pink northern shrimp (Pandalus borealis) from the North Atlantic--continued

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
11 41°43' to 43°58'N 65°14' to 68°28'W, II 10/31 - 11/13/73[b] 7.3 ± 1.2g[f] 6 - 9g[g] 161 ± 117[h] 39 - 380[i] 1771[i] Woods Hole, Mass.[c]	0.02 ± 0.02 0.00 - 0.05	0.45 ± 0.12 0.25 - 0.63	0.11 ± 0.02 0.09 - 0.15	16.91 ± 5.87 9.35 - 29.50	0.51 ± 0.16 0.26 - 0.80	0.10 ± 0.04 0.05 - 0.17	0.16 ± 0.06 0.10 - 0.25	2.15 ± 0.56 0.86 - 2.78	9.73 ± 1.68 7.50 - 13.38	0.33 ± 0.09 0.14 - 0.48	0.01 ± 0.03 0.00 - 0.10	0.02 ± 0.04 0.00 - 0.12	0.42 ± 0.20 0.24 - 0.90	0.70 ± 0.19 0.38 - 1.09
14 43°13' to 52'N 68°9' to 45'W, III 11/16/73[b] 3.2 ± 0.9g (n=13)[f] 2 - 4g (n=13)[g] 318 ± 110 (n=13)[h] 116 - 500 (n=13)[i] 4136 (n=13) [i] Woods Hole, Mass.[c]	0.02 ± 0.02 0.00 - 0.07	0.56 ± 0.27 0.25 - 1.25	0.11 ± 0.02 0.08 - 0.18	8.33 ± 3.88 1.15 - 15.23	0.50 ± 0.19 0.03 - 0.97	0.12 ± 0.03 0.09 - 0.19	0.30 ± 0.20 0.06 - 0.75	2.34 ± 1.19 1.31 - 5.63	12.01 ± 5.49 8.50 - 27.34	0.43 ± 0.08 0.29 - 0.56	0.02 ± 0.03 0.00 - 0.08	0.06 ± 0.13 0.00 - 0.47	0.36 ± 0.10 0.23 - 0.62	0.87 ± 0.14 0.64 - 1.13

Table 7 -- Trace element content, ppm, found in raw, peeled tails of pink northern shrimp (Pandalus borealis) from the North Atlantic--continued

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
11	0.02 ±	0.47 ±	0.11 ±	5.93 ±	0.37 ±	0.13 ±	0.20 ±	2.50 ±	8.88 ±	0.38 ±	0.04 ±	0.07 ±	0.31 ±	0.97 ±
42°3'-43°18'N	0.02	0.14	0.02	2.37	0.08	0.03	0.09	0.70	1.57	0.09	0.07	0.17	0.07	0.30
70°11' to 28'W, IV	0.00 -	0.31 -	0.06 -	2.23 -	0.19 -	0.09 -	0.06 -	1.38 -	7.08 -	0.24 -	0.00 -	0.00 -	0.21 -	0.56 -
11/19/73[b]	0.06	0.72	0.15	9.08	0.50	0.19	0.38	3.54	11.88	0.56	0.17	0.57	0.38	1.42
3.5 ± 0.9[f]														
1 - 4[g]														
32 ± 8[h]														
130 - 440[i]														
3230[i]														
Woods Hole, Mass.[c]														

Table 7 — Trace element content, ppm, found in raw, peeled tails of pink northern shrimp (Pandalus borealis) from the North Atlantic--continued

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn

a/ Sidwell et al. 1973; Zook et al. 1976.
b/ Date of catch.
c/ NMFS laboratory supplying samples.
d/ Element, mean ± standard deviation.
e/ Element, range.
f/ Weight, mean with or without standard deviation.
g/ Weight, range
h/ Shrimp per sample with or without standard deviation.
i/ Total number shrimp in set.

Table 8 --- Trace element content, ppm, found in raw, peeled tails of sidestripe shrimp (Pandalopsis dispar) from Alaska

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10 Idaho Inlet 5/25/73[a] Auke Bay, Alaska[b]	0.01 ± 0.02[c] 0.00 – 0.06[d]	0.65 ± 0.22 0.44 – 1.13	0.09 ± 0.04 0.04 – 0.19	4.83 ± 1.53 2.78 – 7.15	0.34 ± 0.15 0.15 – 0.58	0.08 ± 0.04 0.04 – 0.16	0.14 ± 0.07 0.00 – 0.22	5.53 ± 1.78 3.50 – 9.13	15.51 ± 6.71 8.75 – 33.75	0.23 ± 0.05 0.14 – 0.27	0.01 ± 0.03 0.00 – 0.11	0.15 ± 0.24 0.00 – 0.78	0.33 ± 0.06 0.23 – 0.43	1.06 ± 0.30 0.50 – 1.50
7 Kachemak Bay 5/10/73[a] Auke Bay, Alaska[b]	0.04 ± 0.06 0.00 – 0.16	0.73 ± 0.38 0.44 – 1.56	0.15 ± 0.06 0.08 – 0.23	4.20 ± 2.08 2.35 – 8.38	0.26 ± 0.13 0.16 – 0.55	0.07 ± 0.02 0.05 – 0.10	0.19 ± 0.08 0.10 – 0.31	4.40 ± 1.96 2.36 – 8.25	13.68 ± 3.75 9.69 – 20.87	0.31 ± 0.16 0.13 – 0.58	0.02 ± 0.04 0.00 – 0.09	0.09 ± 0.14 0.00 – 0.35	0.38 ± 0.04 0.33 – 0.44	0.73 ± 0.24 0.38 – 1.13

a/ Date of catch.
b/ NMFS Laboratory supplying samples.
c/ Element, mean ± standard deviation.
d/ Element, range.

Table 9 --Trace element content, ppm, found in raw, peeled tails of royal red shrimp (Hymenopanaes robustus) caught off St. Augustine Florida

Number of samples, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
12	0.08 ±	0.66 ±	0.12 ±	51.75 ±	0.75 ±	0.07 ±	0.19 ±	3.07 ±	12.06 ±	0.35 ±	0.07 ±	0.13 ±	0.36 ±	0.87 ±
6/4/73[a]	0.04[e]	0.15	0.02	24.54	0.21	0.02	0.05	0.92	2.06	0.09	0.08	0.15	0.08	0.18
25 ± 5g[b]														
18 - 30 g[c]	0.03 -	0.44 -	0.08 -	17.05 -	0.50 -	0.04 -	0.12 -	1.45 -	9.00-	0.19 -	0.00 -	0.00 -	0.24 -	0.63 -
Miami, Florida[d]	0.15[f]	0.94	0.14	92.50	1.19	0.11	0.31	4.69	15.13	0.50	0.19	0.44	0.49	1.25

a/ Date of catch.
b/ Weight, mean and standard deviation.
c/ Weight range.
d/ NMFS Laboratory supplying samples.
e/ Element mean ± standard deviation.
f/ Element range.

Table 10--Trace element content, ppm, found in raw, peeled tails of ocean shrimp (Pandalus jordani) caught in Coos Bay, Oregon

Number of samples, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10	0.05 ±	0.66 ±	0.16 ±	3.84 ±	0.28 ±	0.07 ±	0.29 ±	1.98 ±	12.81 ±	0.26 ±	0.04 ±	0.03 ±	0.25 ±	0.71 ±
8/28/73[a]	0.09[c]	0.27	0.07	1.89	0.16	0.04	0.25	0.50	4.26	0.08	0.06	0.06	0.15	0.28
Tiburon, CA[b]	0.00 -	0.25 -	0.11 -	1.72 -	0.07 -	0.00 -	0.10 -	1.25 -	9.22 -	0.17 -	0.00 -	0.00 -	0.12 -	0.38 -
	0.21[d]	1.09	0.33	7.23	0.60	0.12	0.81	2.86	23.75	0.42	0.19	0.19	0.63	1.00

a/ Date of catch.
b/ NMFS laboratory supplying samples.
c/ Element mean ± standard deviation.
d/ Element range.

Table 11 -- Trace elements content, ppm, in raw, peeled shrimp tails of unknown species imported into the United States[a], mixed species from Alaska and mixed species from a literature survey[b]

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10 Asia; India, 6; Thailand, 3; Philippines, 1 9.6 ± 6.9g[c] 1.2 - 19.7g[d] Peeled & deveined, 8[e] In shell, 2[e] Packers, 6[e] Seattle, Washington[f]	0.05 ± 0.03g[g] 0.01 - 0.09[h]	0.54 ± 0.41 0.09 - 1.21	0.05 ± 0.02 0.02 - 0.08	1.0 ± 0.8 0.4 - 2.7			0.11 ± 0.06 0.00 - 0.18							
10 Mexico; Gulf of California, 7; no info., 2; Matozian, 1 16.0 ± 3.8g[c] 10.0 - 25.2g[d] Packers, 5[e] Seattle, Washington[f]	0.06 ± 0.03 0.03 - 0.12	0.40 ± 0.36 0.10 - 1.18	0.04 ± 0.03 0.00 - 0.10	1.9 ± 0.8 0.5 - 2.9			0.10 ± 0.07 0.01 - 0.23							

Table 11-- Trace elements content, ppm, in raw, peeled shrimp tails of unknown species imported into the United States[a], mixed species from Alaska and mixed species from a literature survey[b] Cont.

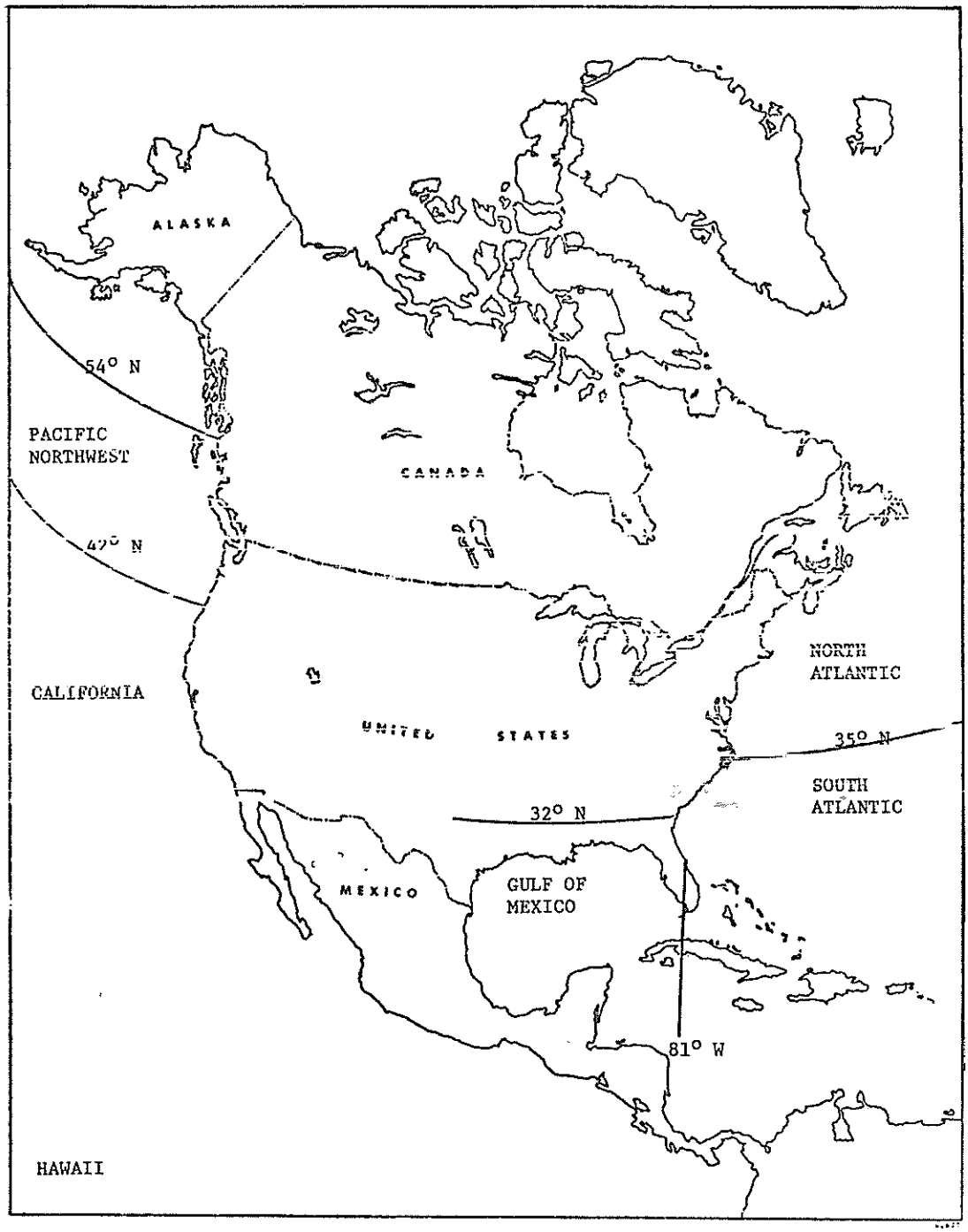
No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn
10 Alaska: S. of Marmot Island, 8; Two Headed Island, 2 5/25-7/14/71[i] Side stripe and coon stripe,9; pink,1; cooked, vacuum packed, Kodiak[e] Seattle, Washington[f]	0.06 ± 0.03 0.01 - 0.11	0.50 ± 0.40 0.17 - 1.24	0.07 ± 0.04 0.02 - 0.11	3.1 ± 1.2 0.9 - 5.2			0.15 ± 0.13 0.09 - 0.19							
Literature values[b]	12[k] 0.15 ± 0.14 0.02 - 0.50	4 0.46 ± 0.06 0.39 - 0.52	8 0.09 ± 0.06 0.04 - 0.24	8 8.71 ± 7.03 1.77 - 23.75	2 1.24 ± 0.92 0.59 - 1.88	0	7 0.07 ± 0.05 0.01 - 0.12	9 4.64 ± 3.54 0.10 - 13.07	7 19.29 ± 11.06 7.39 - 42.00	1 0.03	1 0.03	1 1.90	4 0.73 ± 0.84 0.02 - 1.90	0

Table 11 -- Trace elements content, ppm, in raw, peeled shrimp tails of unknown species imported into the United States[a/], mixed species from Alaska and mixed species from a literature survey[b/] Cont.

No. of samples, site of catch, sample information	Hg	Pb	Cd	As	Se	Ag	Cr	Cu	Zn	Ni	Mo	V	Mn	Sn

a/ References: Sidwell, et al. 1973, and Zook, et al. 1976.
b/ Reference: Sidwell, et al., 1976.
c/ Weight, mean with or without standard deviation.
d/ Weight, range
e/ Inspection service information
f/ NMFS laboratory supplying samples.
g/ Element, mean ± standard deviation
h/ Element, range
i/ Date of capture
j/ Species information
k/ Number of means (one per reference) averaged.

FIGURE 1--RESOURCE SURVEY AREAS:



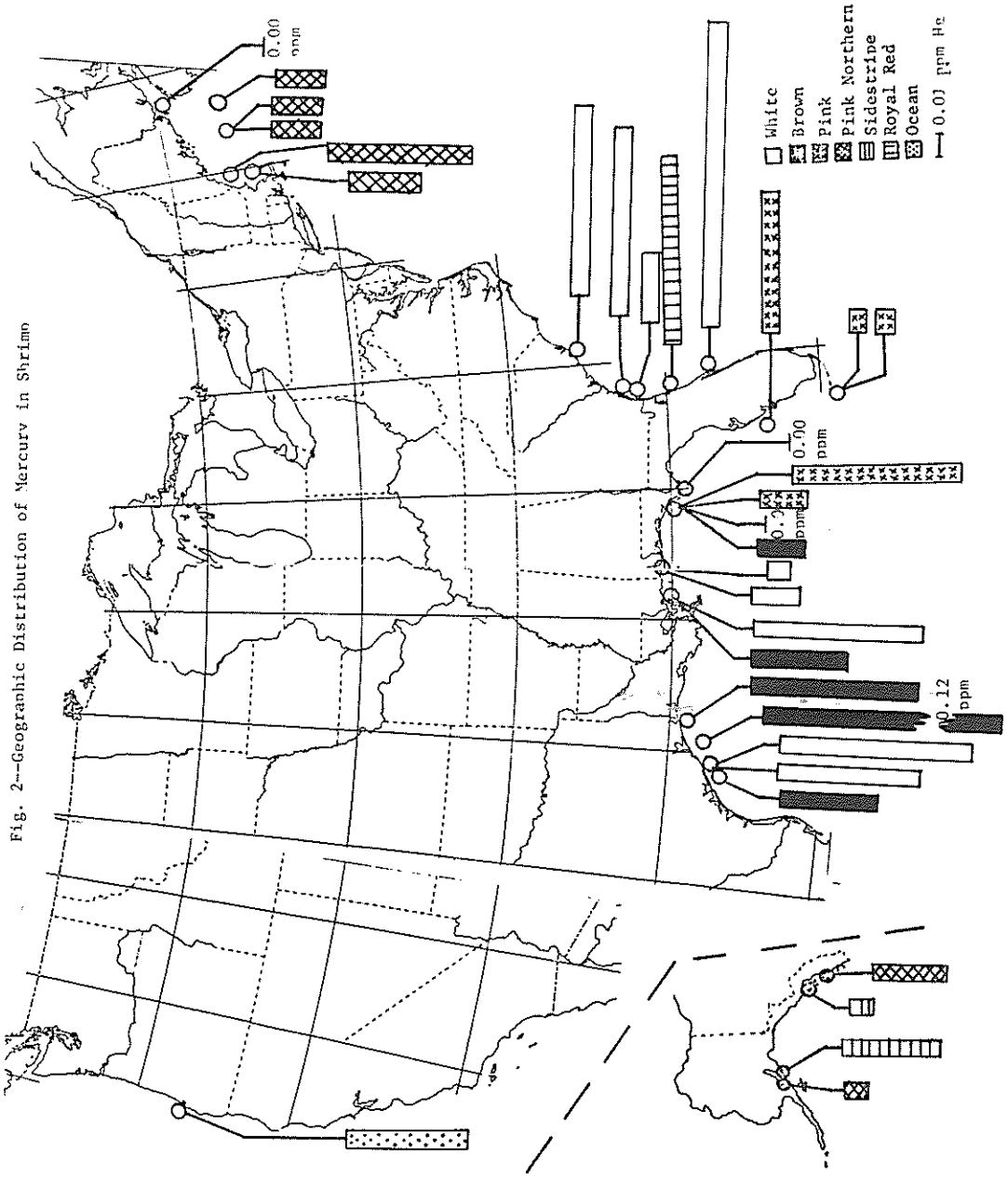
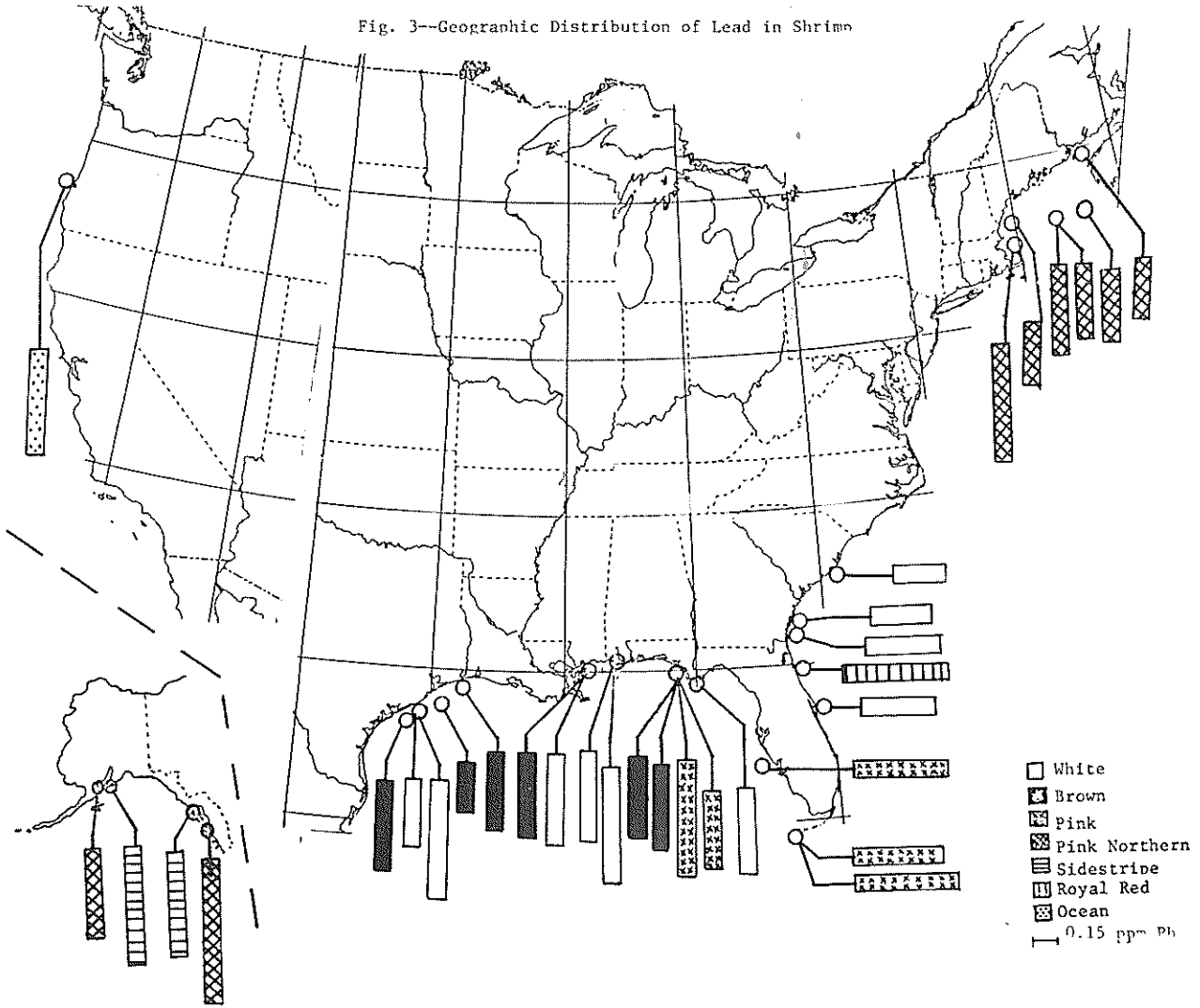


Fig. 2--Geographic Distribution of Mercury in Shrimp

Fig. 3--Geographic Distribution of Lead in Shrimp



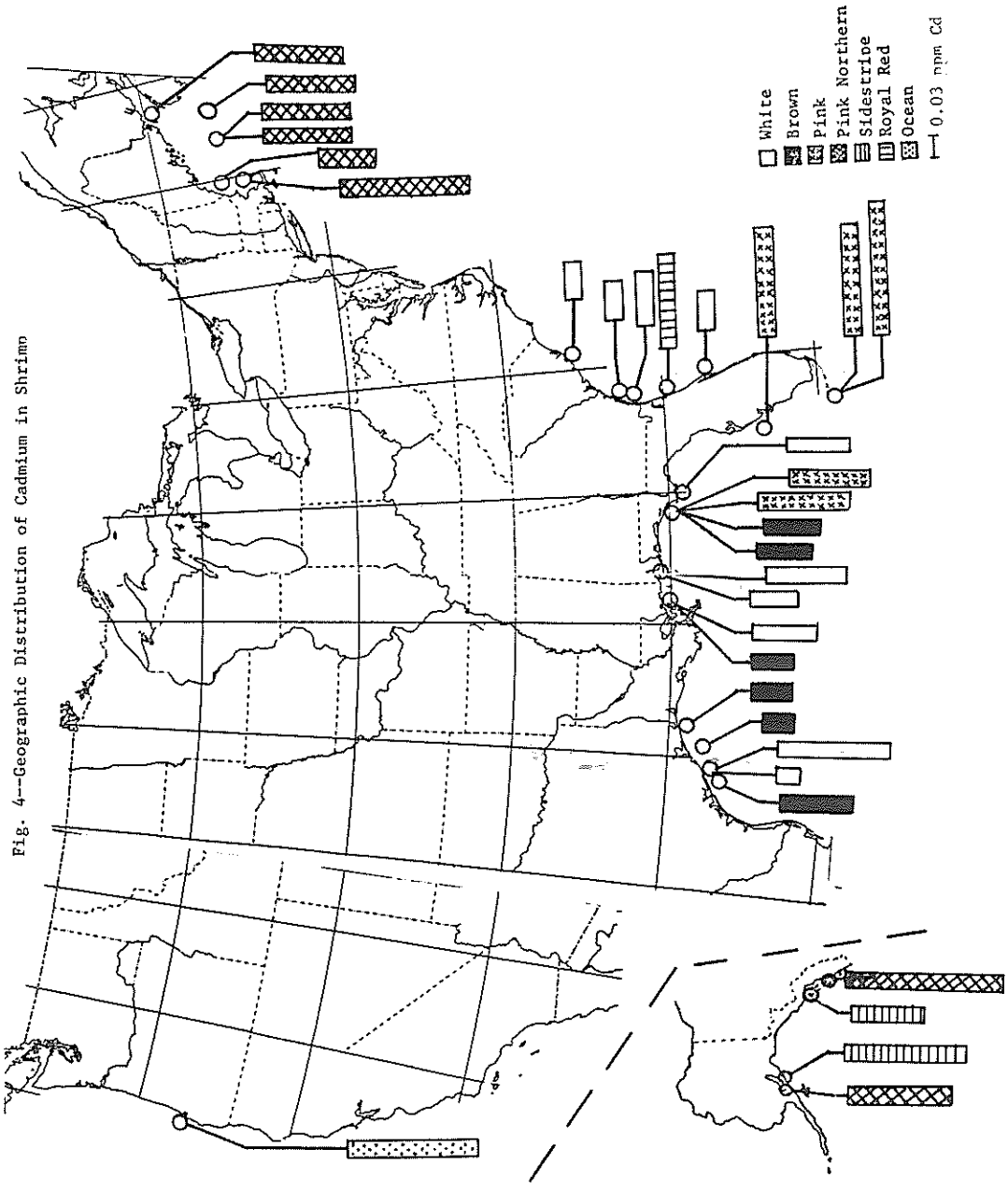
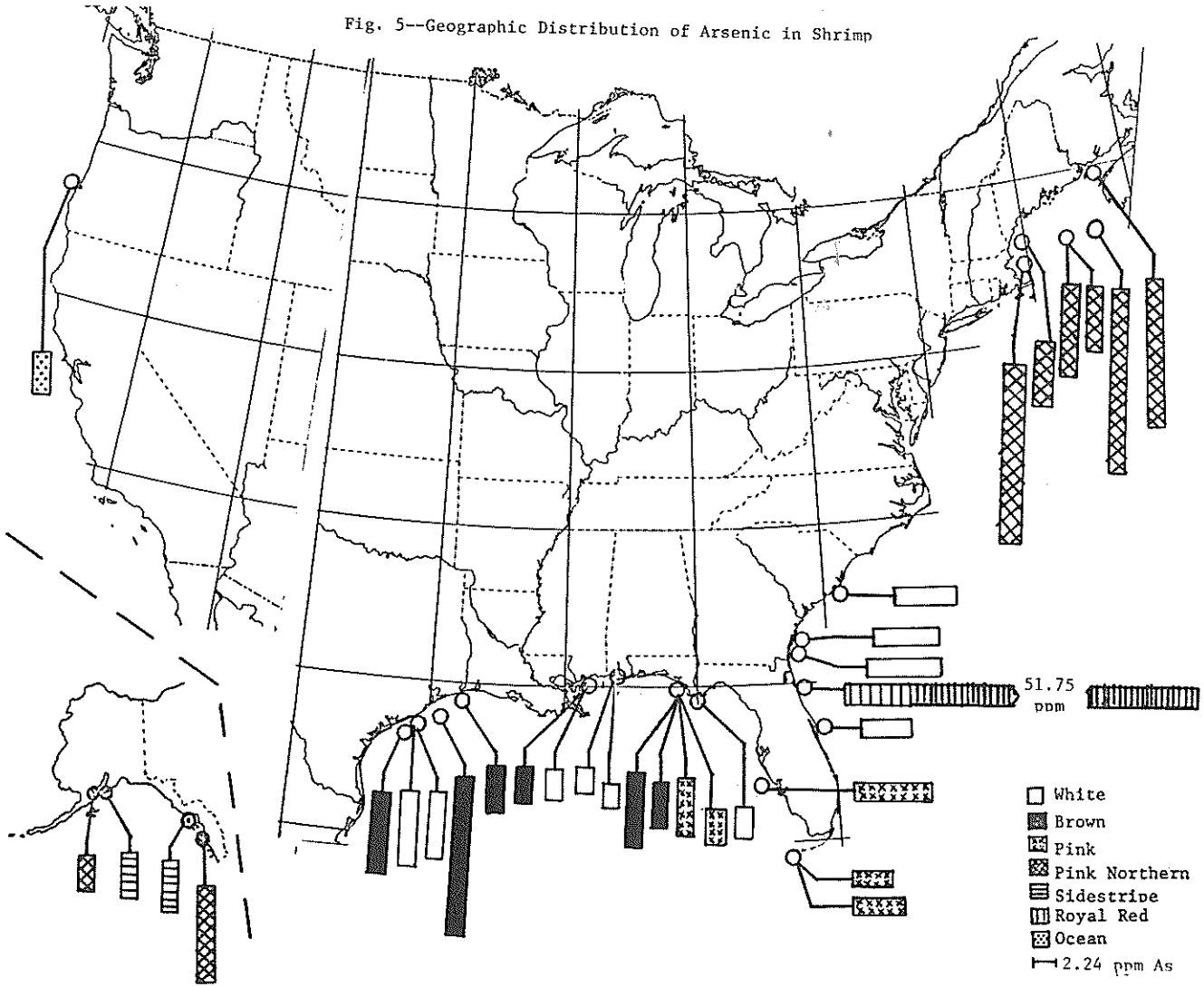


Fig. 4--Geographic Distribution of Cadmium in Shrimp

Fig. 5--Geographic Distribution of Arsenic in Shrimp



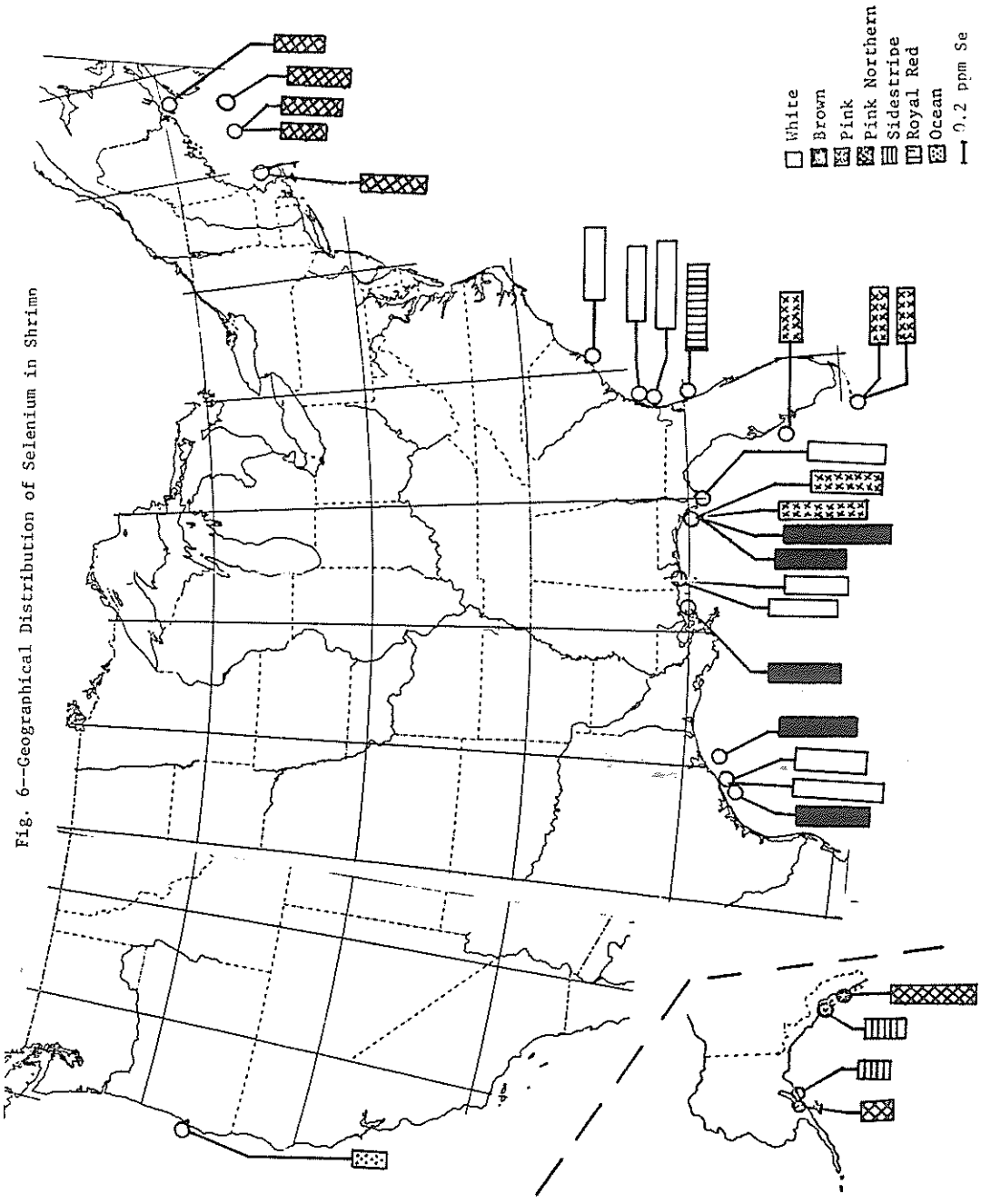
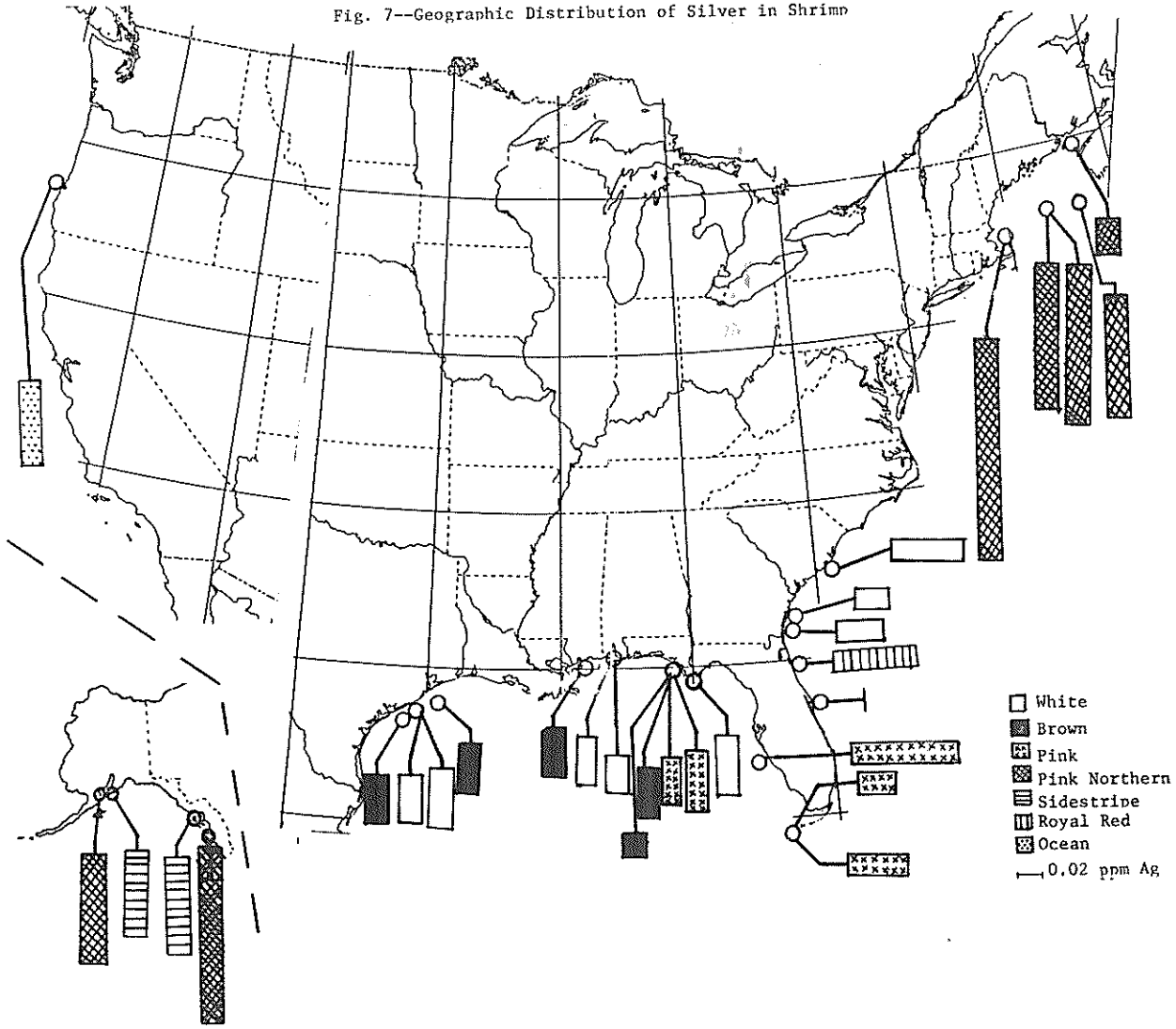


Fig. 6—Geographical Distribution of Selenium in Shrimp

Fig. 7--Geographic Distribution of Silver in Shrimp



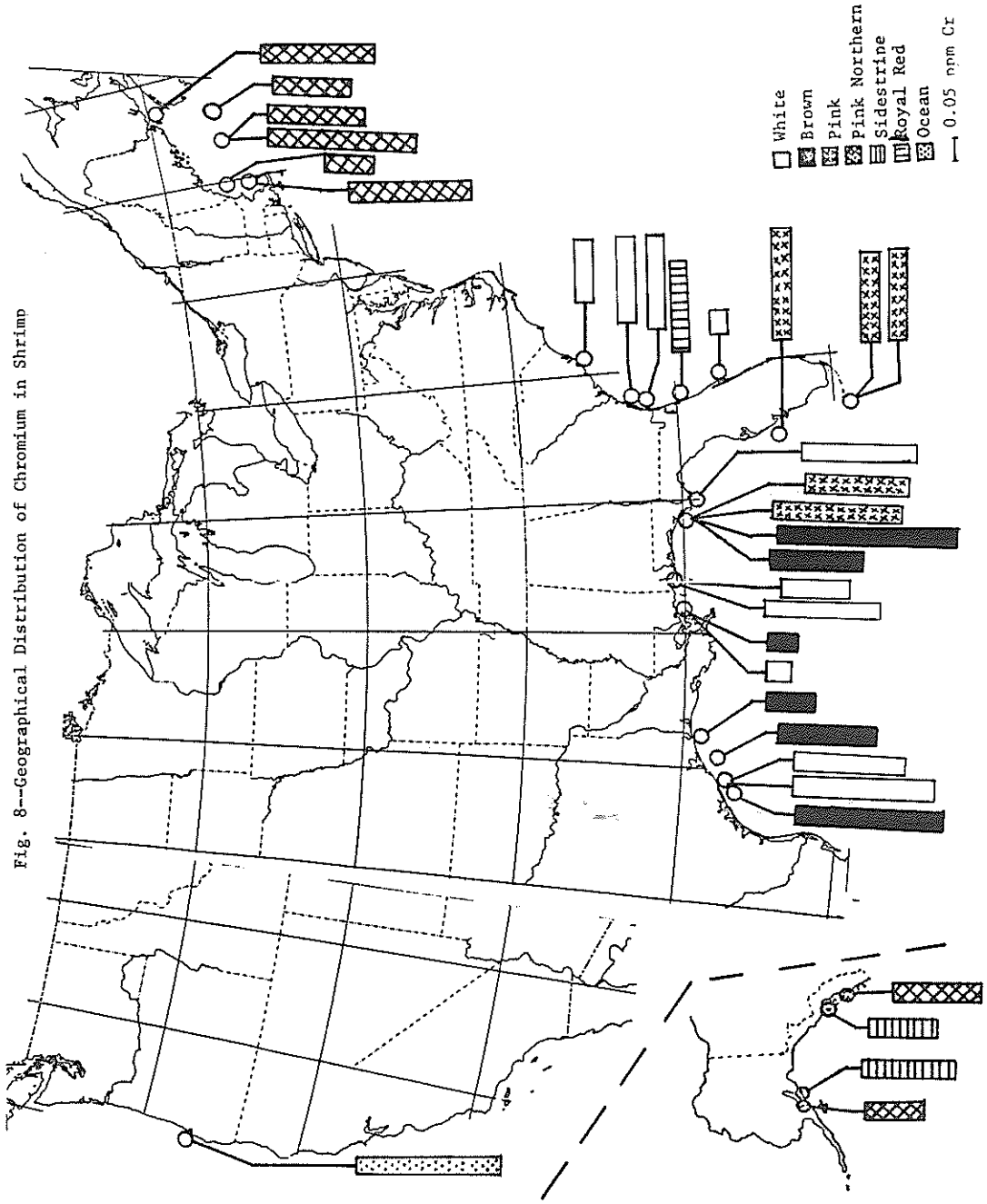
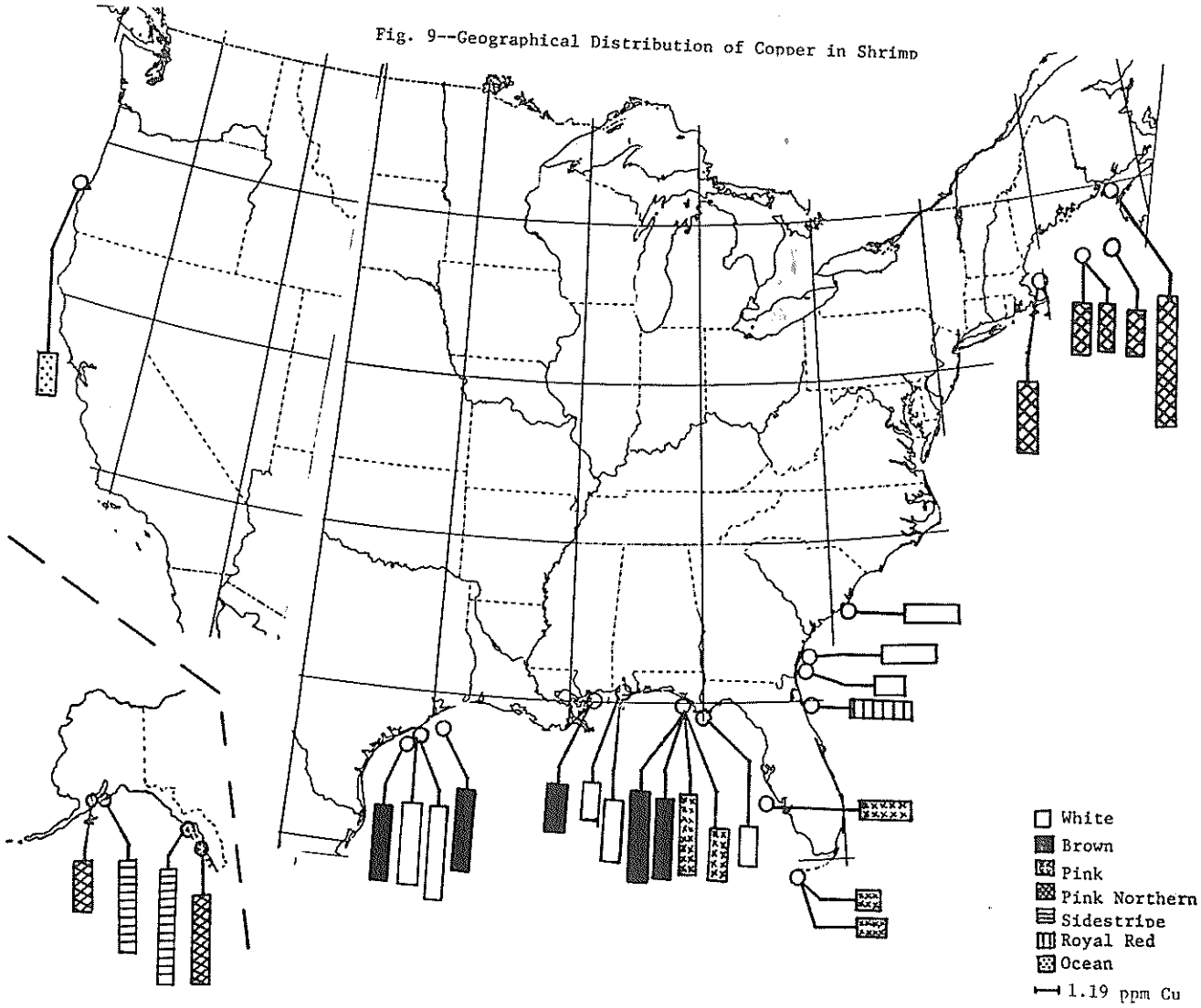


Fig. 8--Geographical Distribution of Chromium in Shrimp

Fig. 9--Geographical Distribution of Copper in Shrimp



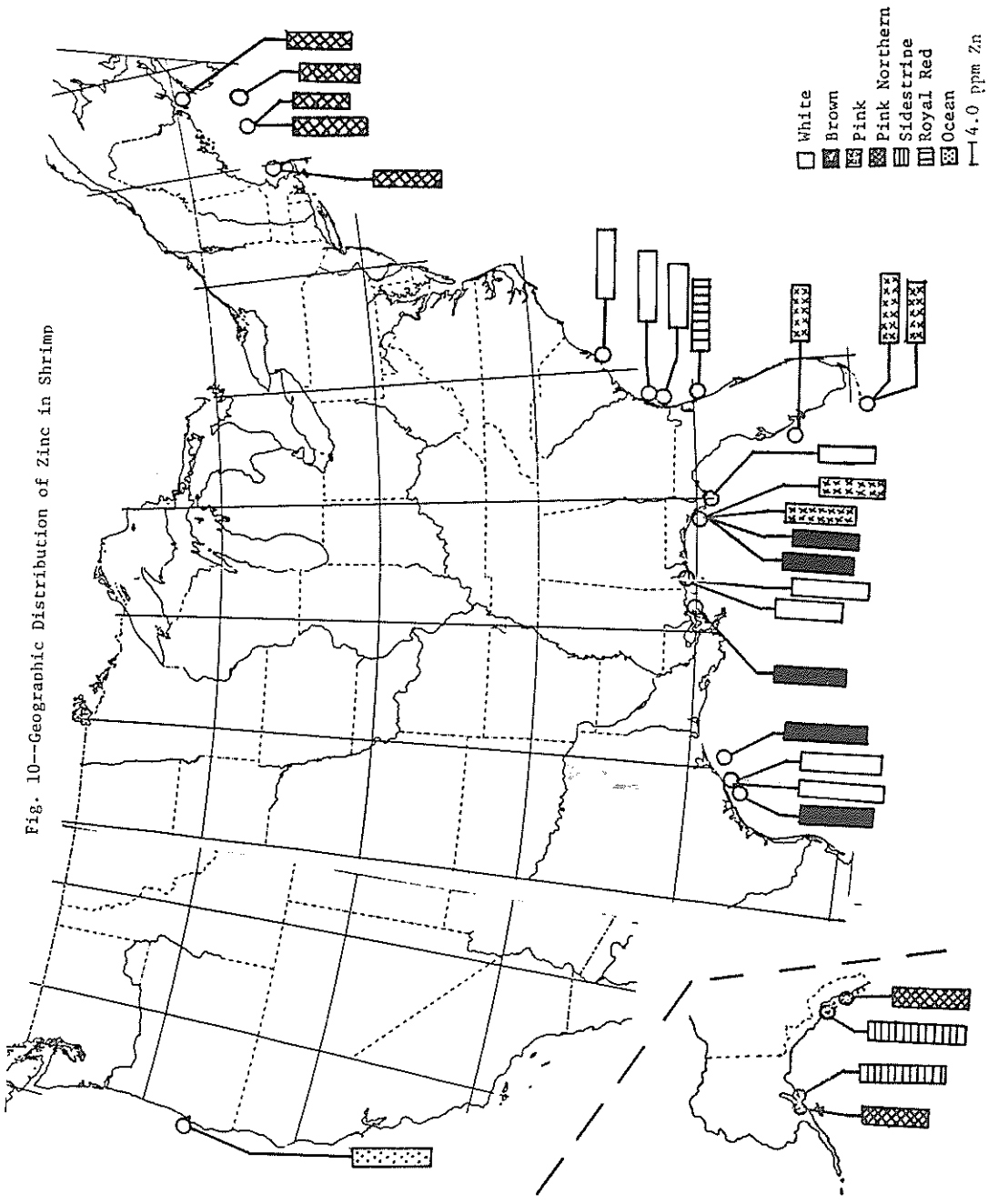
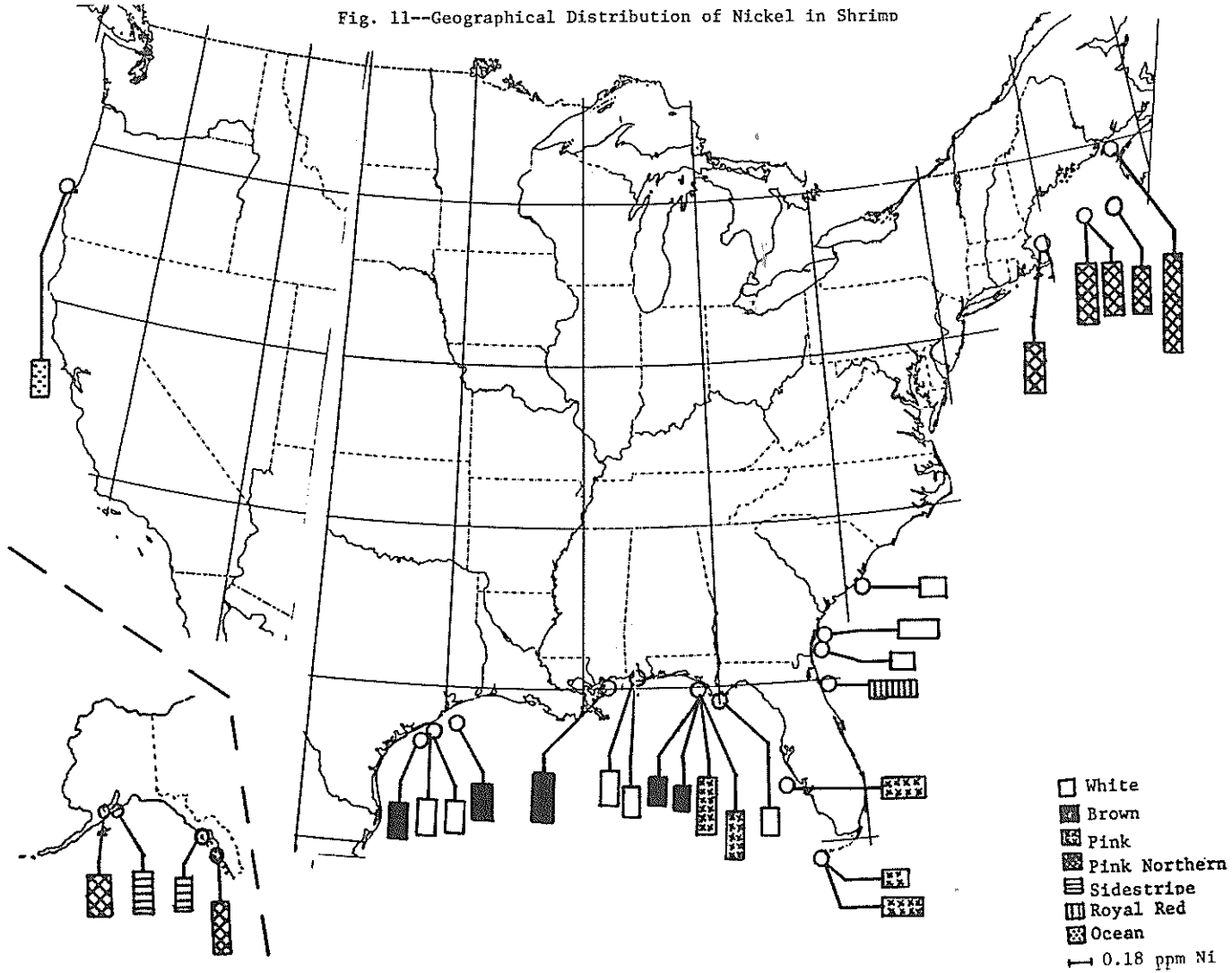


Fig. 10—Geographic Distribution of Zinc in Shrimp

Fig. 11--Geographical Distribution of Nickel in Shrimp



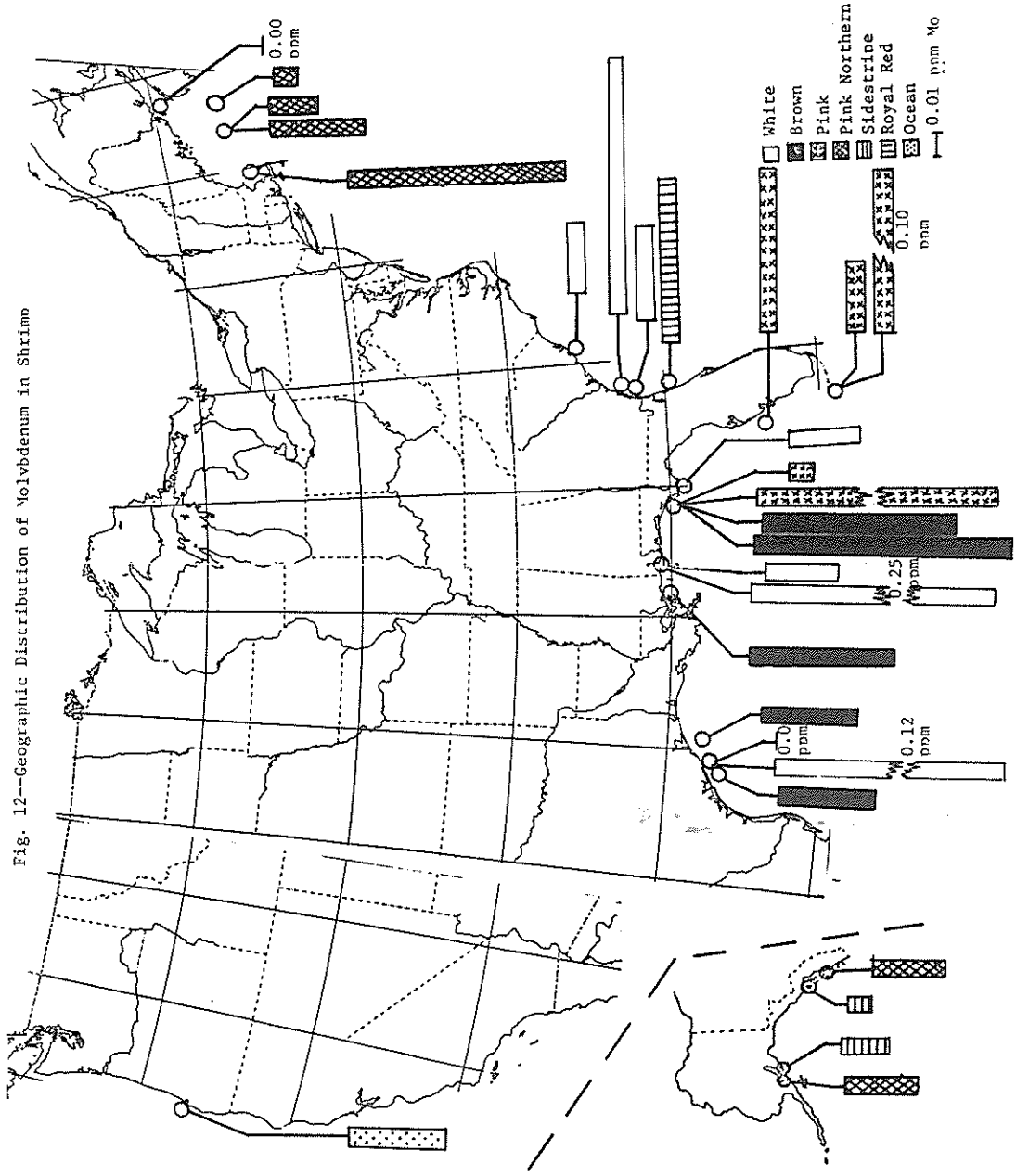


Fig. 12--Geographic Distribution of Molybdenum in Shrimp

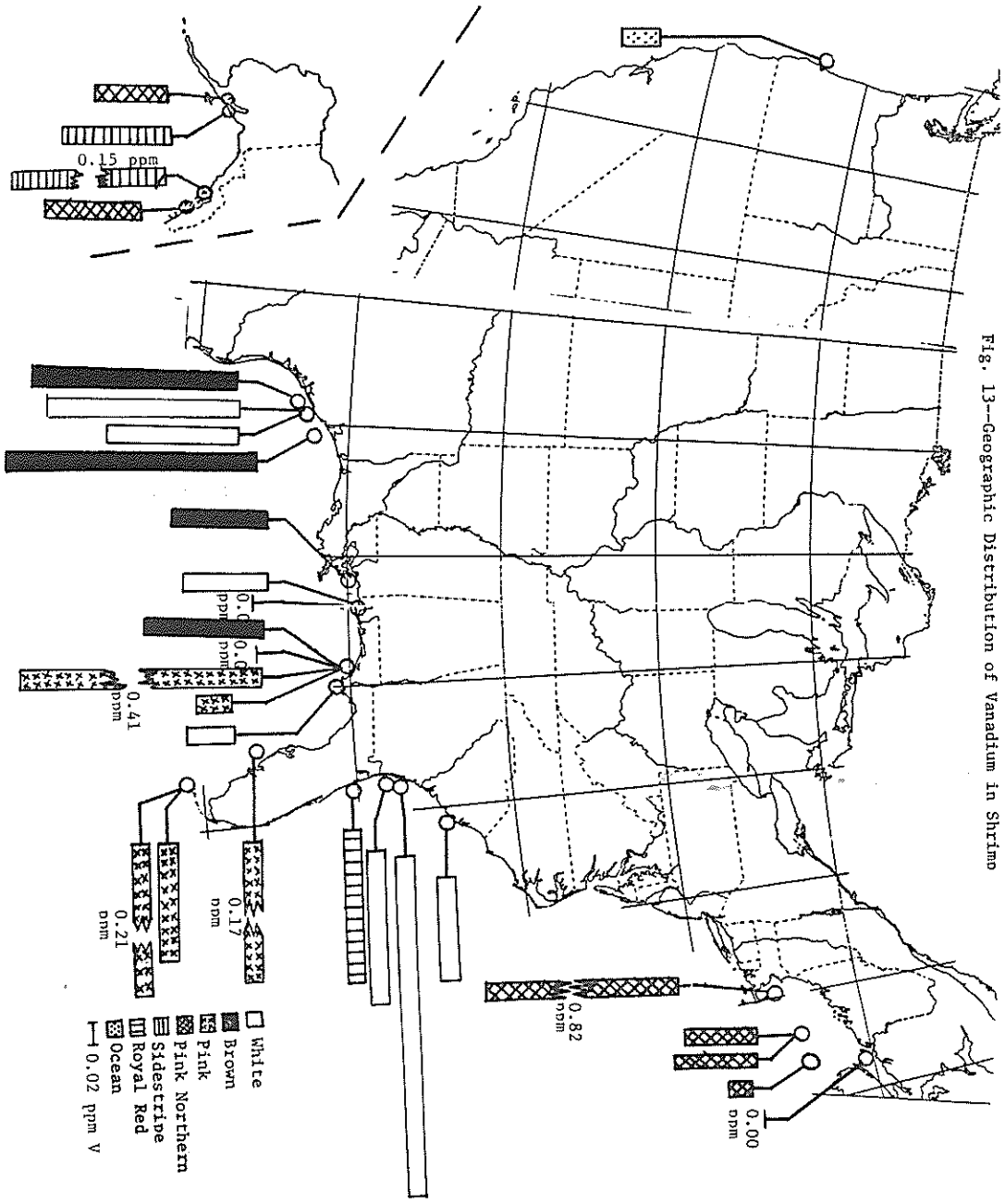


Fig. 13--Geographic Distribution of Vanadium in Shrimp

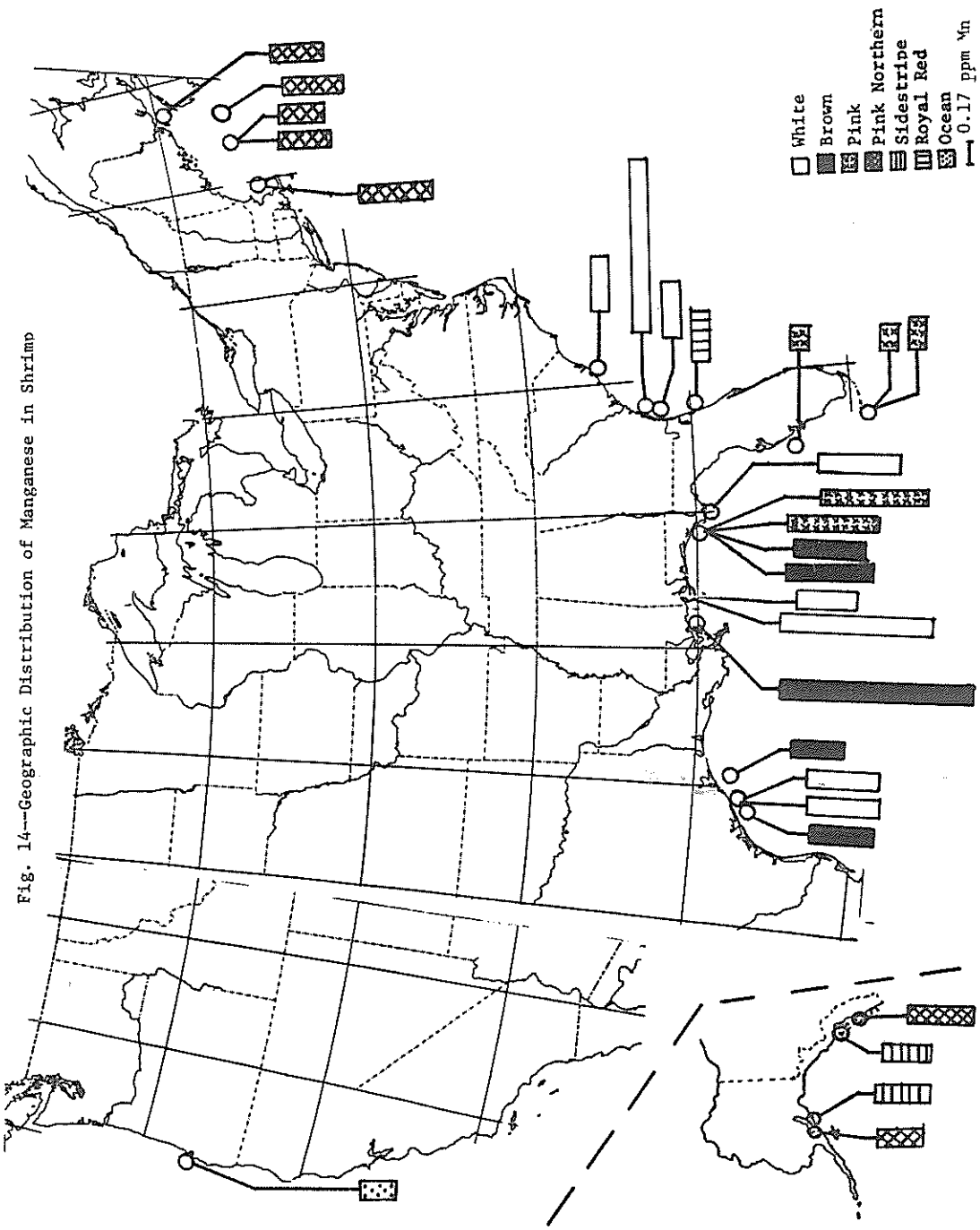


Fig. 14--Geographic Distribution of Manganese in Shrimp

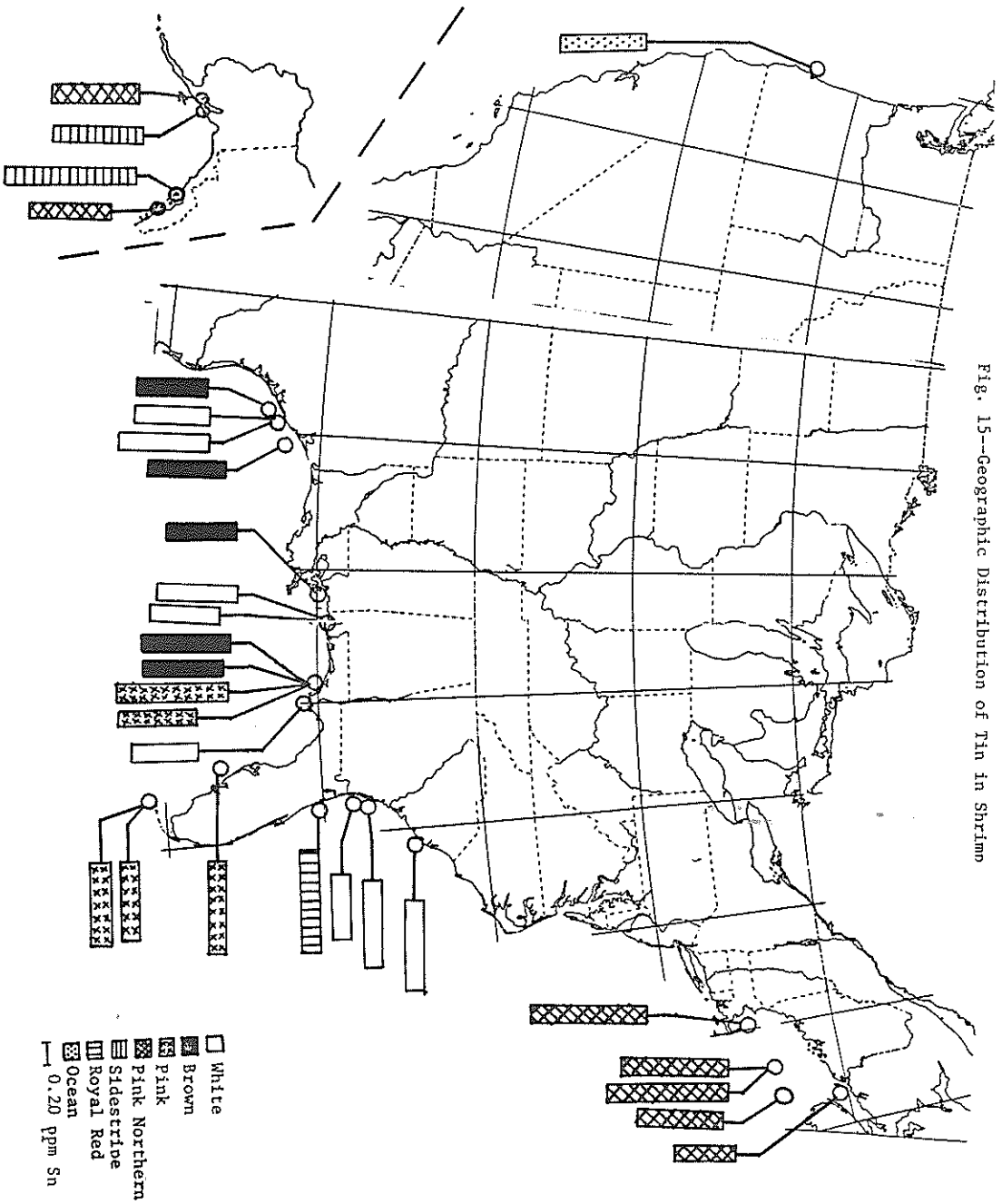


Fig. 15--Geographic Distribution of Tin in Shrimp

POLYCHLORINATED BIPHENYLS IN SHRIMP

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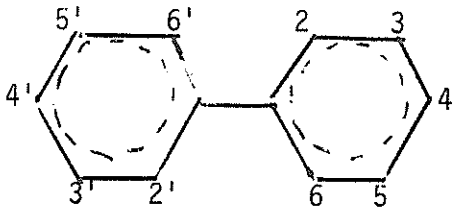
Introduction

Recently, a special class of compounds commonly referred to as PCBs (polychlorinated biphenyls) has attracted world-wide attention of ecologists and scientists. It is paradoxical that although they were widely used commercially during the last forty years, it was only in 1966 that these compounds were reported to be present in environmental samples (Jenson, 1966) and in the following years PCB contamination was found to be almost universal, including human milk, human adipose tissue and brain and liver of small children (Biros et al., 1970).

The generic term PCBs is an abbreviation of a term used to describe the class of compounds referred to collectively as polychlorinated biphenyls, and the commercial products are a family of partially or wholly chlorinated isomers of biphenyl. In a biphenyl molecule are ten positions available for chlorine substitution, and the number of substitution combinations through the chlorination process amounts to some 210 compounds. For example, there will be 3 monochloro-substituted biphenyls; 12 dichloro; 21 trichloro; and so forth as the number of chlorines in the molecule is increased. Industrially, PCBs are produced and sold

as relatively crude mixtures of these isomers.



Structural configuration of PCBs

The nature of chronic toxicities caused by PCBs has led to the study of their extensive distribution in the eco-system, and the routes by which they find their way into the environment. It is interesting to note that the properties which make them industrially indispensable are the same properties which cause them to persist in the environment. These include thermal stability; solubility in a wide range of organic solvents; water insolubility; resistance to oxidation and hydrolysis; high dielectric constant and compatibility with several types of macromolecules. Also, their resistance to oxidation and hydrolysis makes them more stable and persistent than DDT (Gustafson, 1970).

The ubiquity of PCBs can be established from the fact that rainwater in England, brown seals on the coast of Scotland, white-tailed eagles in Sweden, cod in the Baltic Sea, mussels in the Netherlands, Adelic penguin eggs in Antarctica, brown pelican eggs in Panama, Arctic terns, shrimp in Florida, river water in Japan, waters in the Great Lakes, human hair, and human adipose tissue, were all found to contain PCBs, thus making them a class of universally dispersed pollutants.

When a solution or dispersion of PCBs is discharged into a river or lake, the PCBs will accumulate on the sediment because of their low solubility, finally assuming relatively high

concentrations. Subsequently, it is likely to take a long time to flush out a contaminated area.

PCBs were not discovered in environmental samples until very recently, because they were not deliberately distributed around the ecosystem. They demonstrated relatively low acute toxicities and they were difficult to detect analytically. PCBs were discovered rather accidentally because they interfered with the analysis of the organo-chlorine pesticides and they contained enough chlorine atoms to be detected by the electron-capture detectors in the range of tenths or hundredths parts per million (Peakall, 1971).

The incidence of PCBs has been found to be highest in industrialized and urbanized areas. A recent survey reported 32,000 ppm in paddies around an insulator factory in Japan (Japan Times, 1973). Similarly, some samples of mackerel were found to contain up to an amount of 4 ppm in the Tokyo Metropolitan area (Japan Times, 1973). Also, birds whose primary habitat was the San Francisco Bay area showed a larger concentration of PCBs per unit of body weight than those located in Baja California, which is completely rural. The aquatic life around the archipelago of Stockholm, the most industrialized area of Sweden, had a higher concentration of PCBs than fish samples taken from the Western coast of Sweden, a comparatively less developed area. Sediment samples taken from Southern portions of Lake Michigan had a higher concentration than those from other parts of the lake (Gustafson, 1970). Officials of a research institute in Japan believe that the substance has been released into the environment with waste

ink and plastics, and from there found its way into the chain of food cycle and human bodies (Japan Times, 1973).

PCBs have also been detected and analyzed qualitatively in fish and shellfish from Texas and Georgia, and in the Gulf of Mexico. Levels of less than 0.1 ppm were found in fish and blue crabs in estuarine waters at Charleston, South Carolina (Duke et al., 1970). They also monitored the results of an industrial leakage into the Escambia River, which was carried away through the Gulf of Mexico. Maximum levels at the source ranged from a high of 486 ppm in sediment to a low of 20 ppm in fish, crustacea and sediment in the outer bays. The best indicators of the contamination levels were shown to be static organisms like oysters (Duke et al., 1970).

Physico-Chemical Properties of PCBs

There are three characteristic physical properties of PCBs which make them indispensable for industrial applications, which are: (1) high boiling points, (2) low water solubilities, and (3) high dielectric constants. PCBs are miscible with most organic solvents, are compatible with many types of polymers, and usually range from mobile liquids to crystalline materials or hard resins.

PCBs are extremely stable compounds, which are not hydrolyzed by water, acid or alkali, and demonstrate thermal stability to an extent that they are used in fire proofing. They can be heated to 140°C under 260 psi of oxygen pressure without showing any signs of oxidation (Gustafson, 1970), and they are insoluble in water and highly soluble in lipids. Structurally they resemble

chlorinated hydrocarbons such as DDT, are thermoplastic, non-drying, and stable on long heating at 150°C. Electrically, PCBs are non-conducting, do not support combustion when alone above 360°C and are easily soluble in most common solvents and drying oils. It is obvious that the stability of these compounds makes them extremely useful and versatile for numerous formulations and applications.

Nomenclature of PCBs

Structurally, PCBs can be represented as a heterogeneous group of chemicals with varying numbers of chlorine atoms substituted on the biphenyl radical. About 210 different isomers are possible by chlorination of any one or all of the ten available positions. However, it is not known as to which, or how many, of these isomers are involved in food contamination.

In the United States, Monsanto sells PCBs under the trade name of "AROCLOR." The various Aroclors are differentiated by a four-digit number with the last two digits indicating the percentage of chlorine in the mixture. Industrially they are sold as relatively crude mixtures of the isomers.

The average number of chlorines and the numerical code for most of the Aroclor products are as follows:

Aroclor 1200--series represent a biphenyl product, while the last two digits represent the percent chlorine in the product. For example, Aroclor 1254 and 1260 represent PCB formulation containing 54% and 60% chlorine content, respectively.

Aroclor 2565--a blend of biphenyl and triphenyl (75:25).

Aroclor 4465--a blend of biphenyl and triphenyl (60:40).

Aroclor 5442--a chlorinated triphenyl.

Aroclor 1016--a distilled product of 1254, which is more toxic than 1232 or 1242.

Aroclor 1254 and 1260 are the most prevalent forms of PCBs in the environment as well as in foods, and are usually referred to as the standards in research work.

Aroclor 1254. It contains mostly pentachlorobiphenyls with significant but smaller amounts of tetra and hexachlorobiphenyls. Mean chlorine value is 5 atoms per molecule. The most abundant substitutions are: 2,5,--; 3,4,---; 2,3,4,--; 2,3,6,--; 2,4,5,-- and 2,3,4,5,--;

Aroclor 1260. Mainly, it contains hexachlorobiphenyls with a significant level of heptachlorobiphenyls. Mean chlorine value is 6.1 atoms per biphenyl molecule. Most frequent substitution patterns are: 2,5,--; 3,4,--; 2,3,4,--; 2,3,6,--; 2,4,5,--; 2,3,4,5,--; 2,3,5,--; 2,3,4,6,--; and 2,3,5,6,--.

Polychlorinated biphenyls in the environment may constitute a potential health hazard to edible fishery products, which are caught in polluted waters, and/or packaged in materials containing PCBs. Since many state and federal agencies are concerned with the toxicity of PCBs in seafoods, this project was designed to develop physical and chemical methods (ultraviolet radiation and treatment with sodium nitrite) for eliminating these compounds from shrimp.

Experimental

Monsanto Aroclors 1254 and 1260 were used as reference solutions throughout this study, since these two forms of PCBs are widely prevalent in the environment and are used as standards in

PCB studies. Moreover, they provide excellent bases for comparative evaluation for most of the samples.

Hexane was used as solvent for the standard solutions. The stock and working standard solutions were stored at 4°C taking care to protect them from any light source. Stock solutions were prepared afresh after every six months and working solutions were prepared every 2-3 weeks. Before making dilutions or running the standard solutions on gas chromatograph, care was taken to see that they reached the same temperature at which they were prepared. All glassware used in the analysis was thoroughly cleaned with hot water, rinsed with distilled water followed by acetone, and finally with pesticide grade hexane or petroleum ether.

Five pounds of shrimp were peeled, deheaded, deveined and divided into two halves. One half was treated by dipping in 1% sodium nitrite solution for one minute and the other half was left untreated. The sodium nitrite treated shrimp were further subdivided into two groups. One group was dip-coated in 100 ppm solution of Aroclor 1254 and the other group in Aroclor 1260 for exactly one minute. Similarly the untreated shrimp were also dip-coated in Aroclor 1254 and 1260 separately. All samples were placed in different petroleum ether rinsed glass dishes under direct sunlight for 12, 24, 30, 36 and 48 hours. In order to facilitate uniform exposure, the samples were turned every four hours. The sun drying studies were conducted during the months of July, August and September, with temperatures fairly constant at about 90°F. The control samples under each group were not irradiated and kept in dishes covered with aluminum foil, away from any

light source.

On completion of sun-drying 25 g of the fresh sample and 5 g of the sun-dried sample were blended with 100 ml acetonitrile for 2 minutes. The contents were filtered under suction through 12 cm Buchner funnel into a 500 ml suction flask. Residues and filter paper were placed in the same blender jar using another 100 ml acetonitrile and contents were again filtered using the same suction flask. Combined extracts were added to a separatory funnel and 100 ml petroleum ether (60-80°C) was added and vigorously shaken, followed by 10 ml of saturated sodium chloride solution and 350 ml of water. Contents were thoroughly mixed for 30 to 45 seconds. After the separation of layers, the aqueous layer was discarded and the PCB-containing solvent layer was washed with two 250 ml portions of water. The washings were discarded and the solvent layer was transferred to a 100 ml glass stoppered graduated cylinder. Fifteen grams of anhydrous sodium sulfate was added and shaken to absorb any moisture.

The extracted sample was further subjected to cleanup by Florisil column chromatography. Columns were packed with four inches of activated Florisil (Floridin) topped with half inch anhydrous sodium sulfate. Forty to fifty ml petroleum ether was added and eluted to the top of the packing surface. The sample extract was then poured into the column and allowed to elute at about 3-5 ml per minute. On completion, 50 ml of the eluting solvent (6% ethyl ether in petroleum ether) was added and allowed to elute at the rate of 5 ml per minute. After 50 ml of the eluting solvent reached the top of column packing, an additional 150 ml of

solvent was added. The cleaned-up extract was concentrated and used for gas chromatographic quantitation. No further cleanup was necessary since only a comparative evaluation was desired and chances of contamination with other pesticides were minimal.

Gas chromatographic analyses were carried out (Table I) using Varian Aerograph (Model 1200) equipped with an electron capture detector, fitted with a 5 ft by 1/i inch coiled pyrex glass column packed with 10% DC-200 on Chromosorb W (acid washed and silanized) and operated at inlet, column and detector temperatures of 225, 185 (Isothermal) and 210°C respectively. Nitrogen flow rate was adjusted to 55 ml per minute with the chart speed of 1/4 inch per minute. The quantitation was based on the peak areas (peak height x 1/2 base) of the standards and the samples. All calculations were based on solid contents in order to avoid the differences due to varying moisture content during sun-drying of shrimp. Duplicate samples were studied.

Combined gas chromatography/mass spectrometric analyses (Table II) were undertaken to identify different chlorinated biphenyl isomers present in different GC peaks.

Results and Discussion

Gas chromatographs of the two PCB standards (Fig. 1 and 2) indicate the presence of different peaks appearing at different retention time. Aroclor 1260 requires more time which can be attributed to its higher chlorine content (60%) than Aroclor 1254 (54%). A comprehensive study of these two standards is essential

TABLE I

OPERATING CONDITIONS FOR GAS CHROMATOGRAPH
USED FOR MASS SPECTROMETRIC STUDIES
Instrument: Perkin Elmer Model 990

Component		Conditions
1.	Detector	
	a. Source	Flame ionization
	b. Temperature	210°C
2.	Column	
	a. Type	Stainless steel (silanized)
	b. Diameter	1/8 in
	c. Length	6 ft
	d. Packing	OV-1 silicone oil
	e. Temperature	185°C (Isothermal)
3.	Carrier Gas	
	a. Type	Helium
	b. Flow rate	40 ml/minute
4.	Injector	
	a. Temperature	210°C
5.	Recorder	
	a. Chart speed	5 mm/minute

TABLE II

OPERATING CONDITIONS OF
GAS CHROMATOGRAPHY
Instrument: Varian Aerograph Model 1200

Component	Conditions
1. Detector	
a. Source	Tritium Foil
b. Activity	250 mCi (millicuries)
c. Applied voltage	90 volts
d. Temperature	210°C
2. Column	
a. Type	Coiled, Pyrex glass
b. Diameter	1/8 in
c. Length	5 ft
d. Packing - solid support	Chromosorb W
e. Packing - Liquid support	10% DC-200
f. Temperature	185°C
3. Carrier Gas	
a. Type	Nitrogen
b. Flow rate	55 ml/minute
4. Injector	
a. Temperature	225°C
5. Recorder	
a. Type	Honeywell Electronik 19
b. Chart speed	1/4 in/minute
c. Applied voltage	1 mv (millivolt)

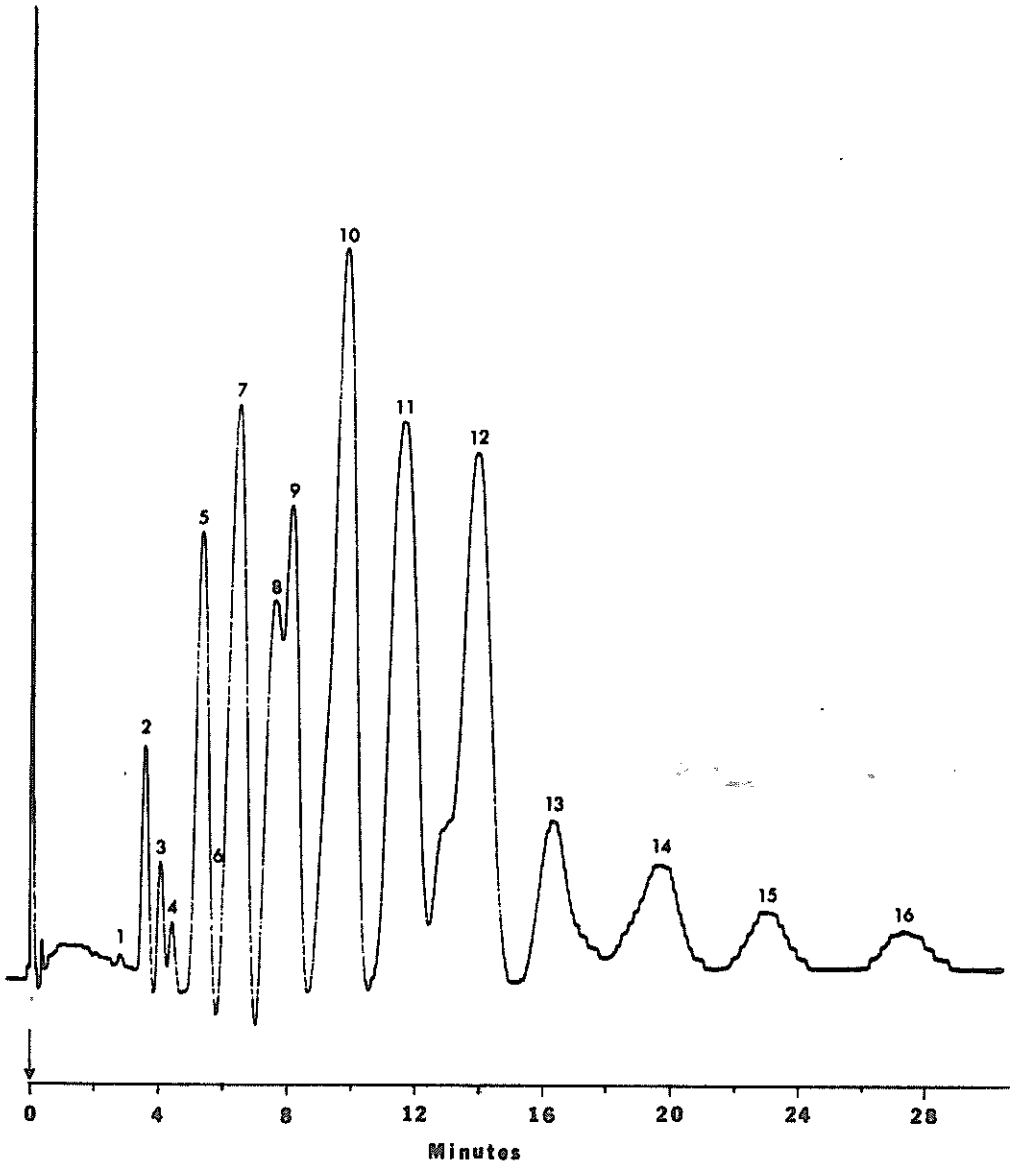


FIGURE 1. Gas chromatograph of Aroclor 1254.

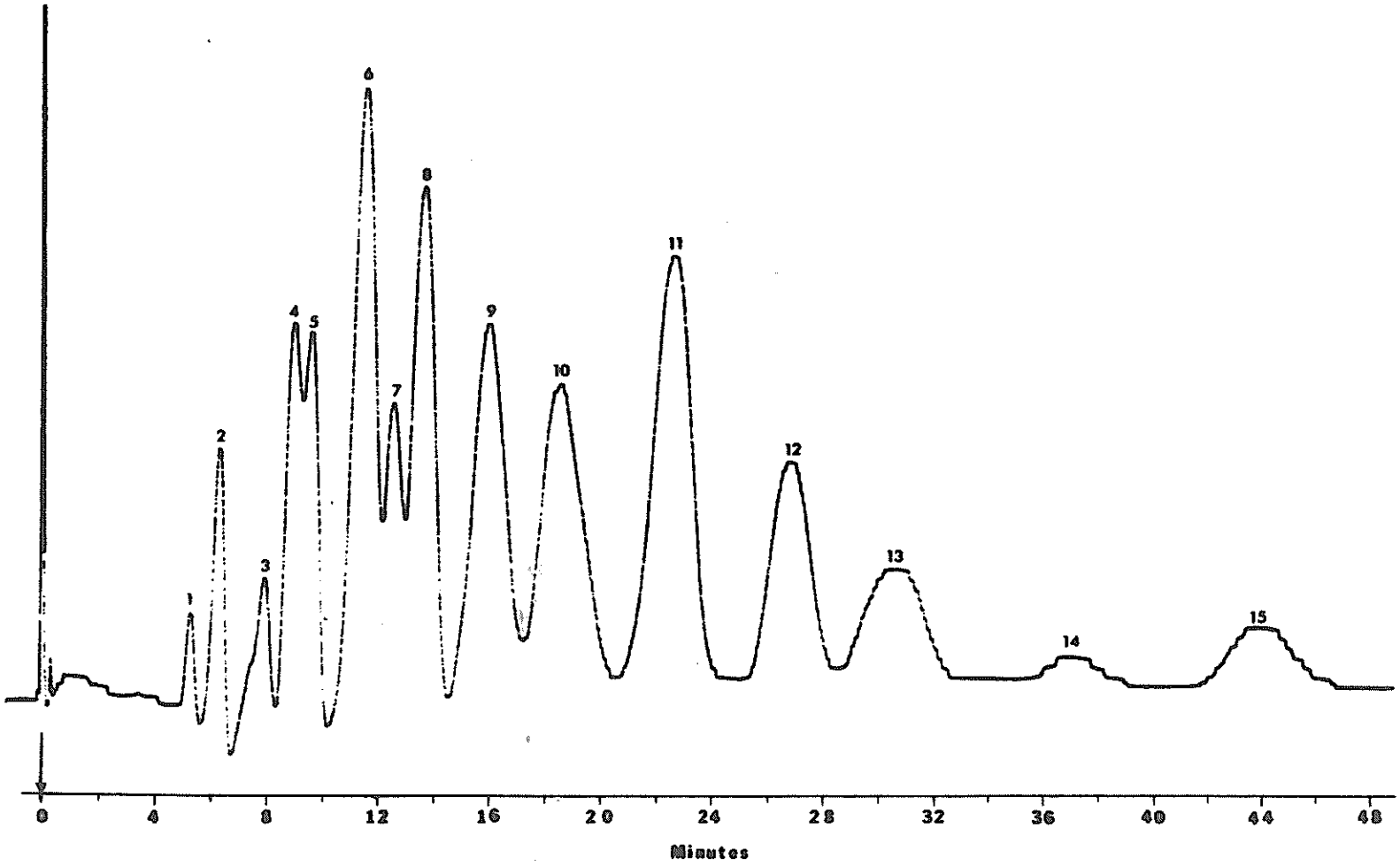


FIGURE 2. Gas chromatograph of Aroclor 1260.

before discussing the results obtained. A combined gas chromatography/mass spectrometric analysis of individual peaks (Figure 3) reveals that each separate peak is representative of a chlorinated biphenyl having a different number of chlorine atoms. An overall combination of these peaks represents complete polychlorinated biphenyls in the mixture.

In the mass spectra of the peaks, relatively intense fragment ions were produced by consecutive loss of chlorine atoms from the parent ion. From all of the confirmatory tests available for PCBs, mass spectrometry represents the most sophisticated and reliable method. The mass spectra for all the major peaks in the gas chromatogram were recorded prior to their comparative evaluation. Relative intensities were calculated on the basis of the most intense peak in the spectrum, which was regarded as the base peak with a value of 100. Doubly charged ions of odd mass values were omitted. Peaks having a relative intensity of 1% or less and those having a m/e ratio less than 100 were also ignored.

Thus interpretation of the mass spectra showed peak 1 in the chromatograph of Aroclor 1254 to be dichlorobiphenyl, peak 2 trichlorobiphenyl, peaks 3 to 5 tetrachlorobiphenyls, peaks 7 and 8 pentachlorobiphenyls, peaks 9 to 13 hexachlorobiphenyls and peaks 14 and 15 heptachlorobiphenyls.

Similarly, in the chromatogram of Aroclor 1260, peak 1 represented trichlorobiphenyl, peaks 2 and 3 tetrachlorobiphenyls, peak 4 pentachlorobiphenyl, peaks 5 and 6 hexachlorobiphenyls, peaks 7 to 12 heptachlorobiphenyls, peak 13 octachlorobiphenyl, peak 14 nonachlorobiphenyl, and peak 15 decachlorobiphenyl.

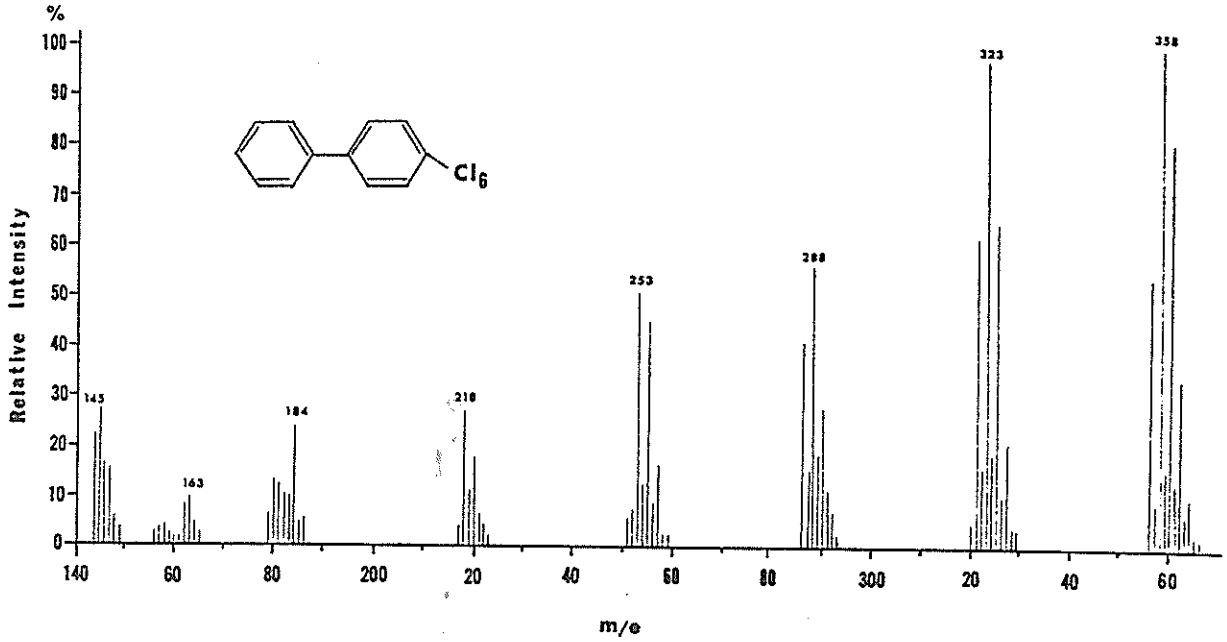


FIGURE 3. Mass spectrum of hexachlorobiphenyl identified in Aroclor 1254 (Peak No. 10).

However, it should be noted that different chlorinated biphenyls vary with the amount of chlorine content, the type of the mixture, and the manufacturing processes involved. In this study, these different chlorinated biphenyls, identified by mass spectrometric analyses, were used for comparative evaluations, as well as to determine the chlorinated biphenyls that are most susceptible or resistant to degradation.

It was noteworthy from our results (Table III and IV) that the complete degradation of Aroclor 1254 in shrimp took place after 30 hours in sodium nitrite treated shrimp and 36 hours in the untreated shrimp. Reduction of Aroclor 1260 required 48 hours of sun-drying for sodium nitrite treated shrimp and 60 hours for the untreated shrimp. Apparently, the sun-drying effects depend upon the percent chlorination of PCBs. In other words, the higher the chlorine content the longer will be the time taken for radiation-induced degradation. Also, the degradation rate was found to be approximately proportional to the time of exposure (Figure 4 and 7) except during the first 12 hours of exposure which showed rapid degradation.

An interesting fact observed was the appearance of several smaller peaks (Figure 5 and 8) upon radiation and the peak height of some peaks increased severalfolds. This led to the conclusion that the lower chlorinated biphenyls are being fromed as a result of breakdown of higher chlorinated biphenyls during the process of radiation. Our assumption agrees with results of Ruzo et al. (1972) who predicted photolysis of 3, 3', 4, 4'--tetrachlorobiphenyl produced a stepwise dechlorination process, that is, tetrachloro

TABLE III
EFFECT OF SUN-DRYING ON PCB (AROCLOR 1254) CONTENT IN SHRIMP

Hours of Exposure	Without Sodium Nitrite Average		With Sodium Nitrite Average	
	µg/g solids	% Reduction	µg/g solids	% Reduction
0	28.9	0	28.8	0
12	24.8	14.1	21.1	26.8
24	12.1	58.2	3.5	87.8
30	1.6	94.4	0	100.0
36	0	100.0	0	100.0

TABLE IV
EFFECT OF SUN-DRYING ON PCB (AROCLOR 1260) CONTENT IN SHRIMP

Hours of Exposure	Without Sodium Nitrite Average		With Sodium Nitrite Average	
	µg/g solids	% Reduction	µg/g solids	% Reduction
0	31.6	0	33.3	0
12	25.6	18.8	22.6	32.2
24	17.2	45.5	12.0	63.9
36	10.0	68.2	9.3	72.2
48	2.7	91.4	0	100.0

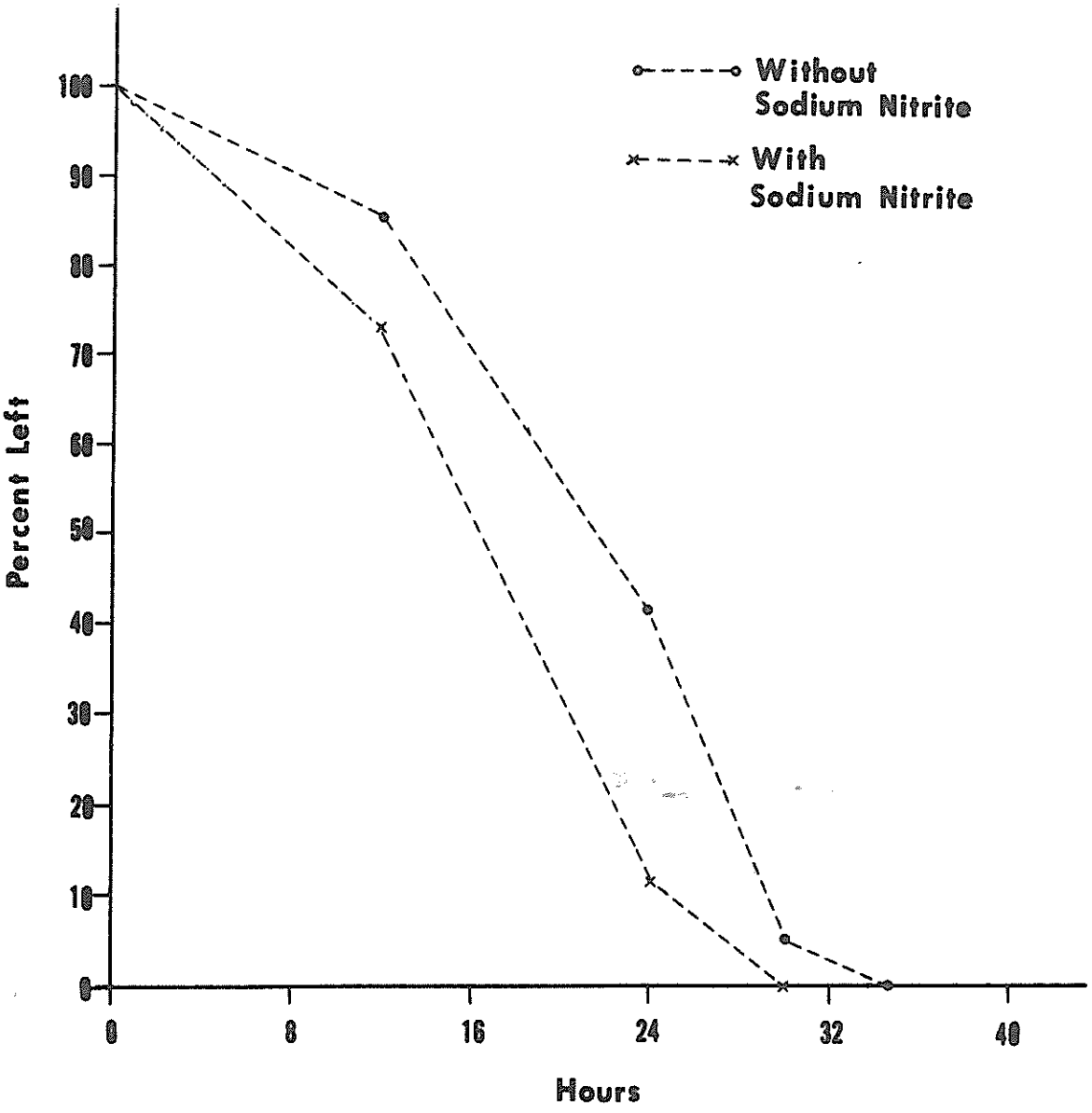


FIGURE 4. Effect of sun-drying on PCB (Aroclor 1254) content in shrimp.

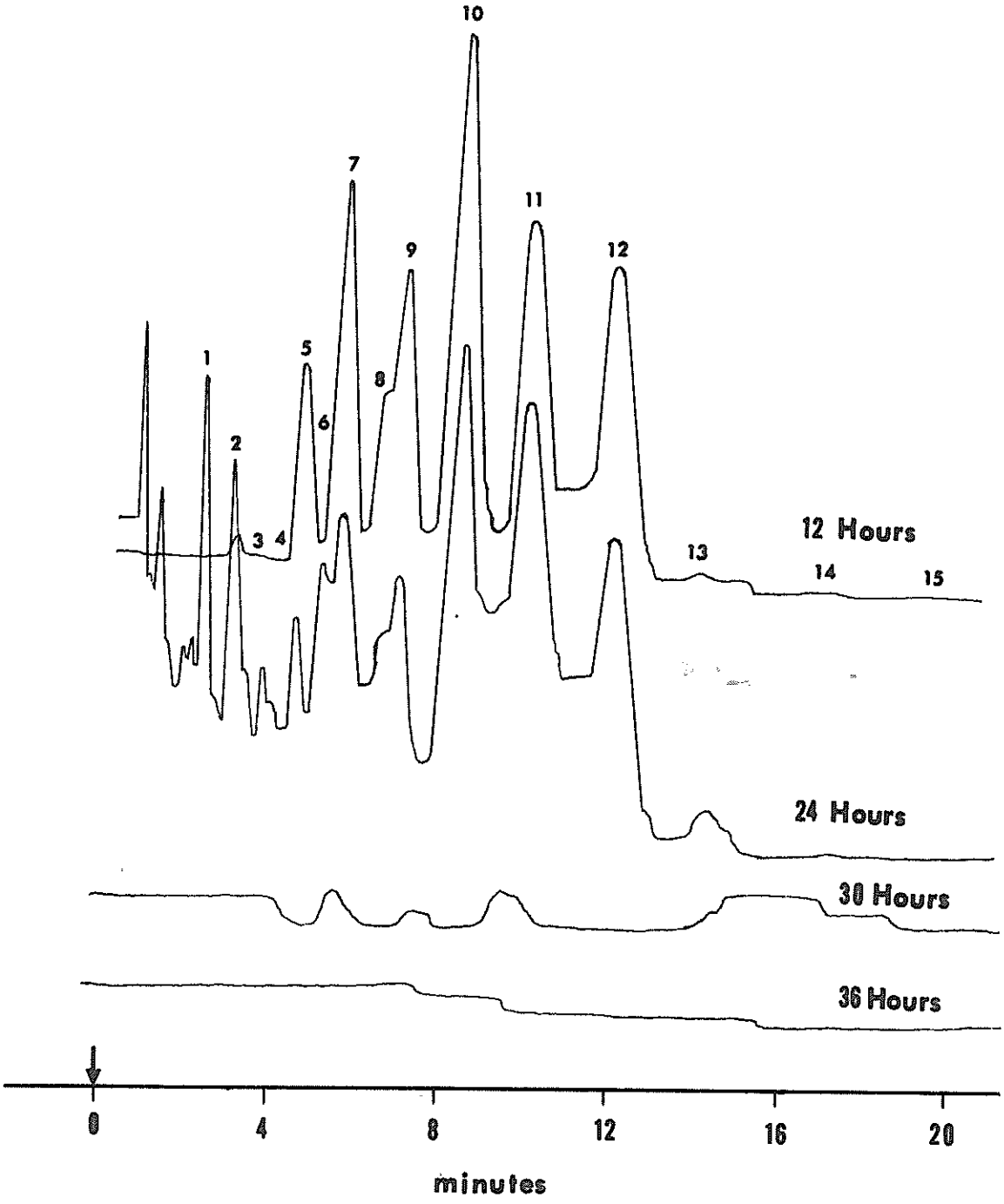


FIGURE 5. Gas chromatographs showing effect of sun-drying on PCB (Aroclor 1254) content in shrimp.

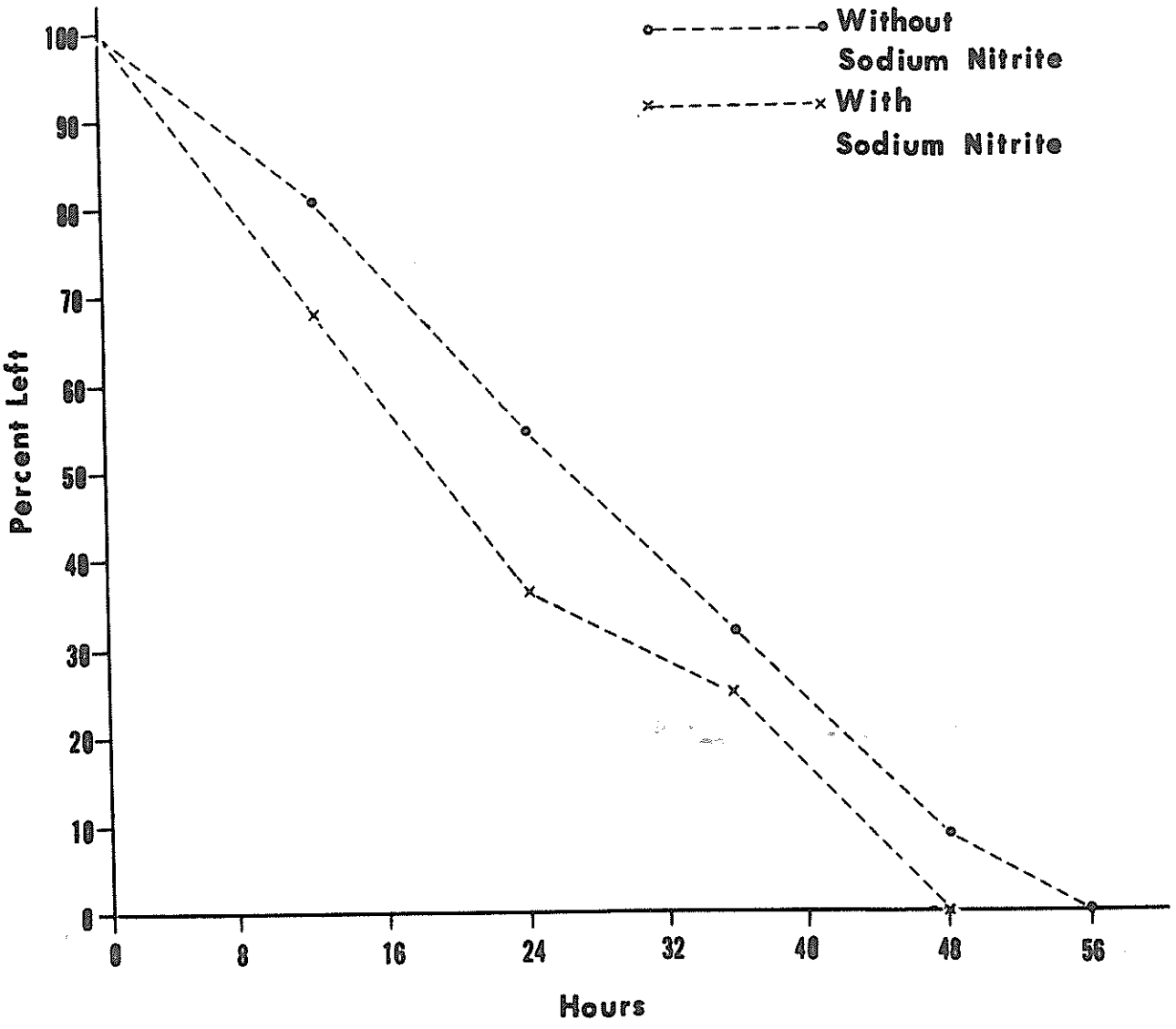


FIGURE 6. Effect of sun-drying on PCB (Aroclor 1620) content in shrimp.

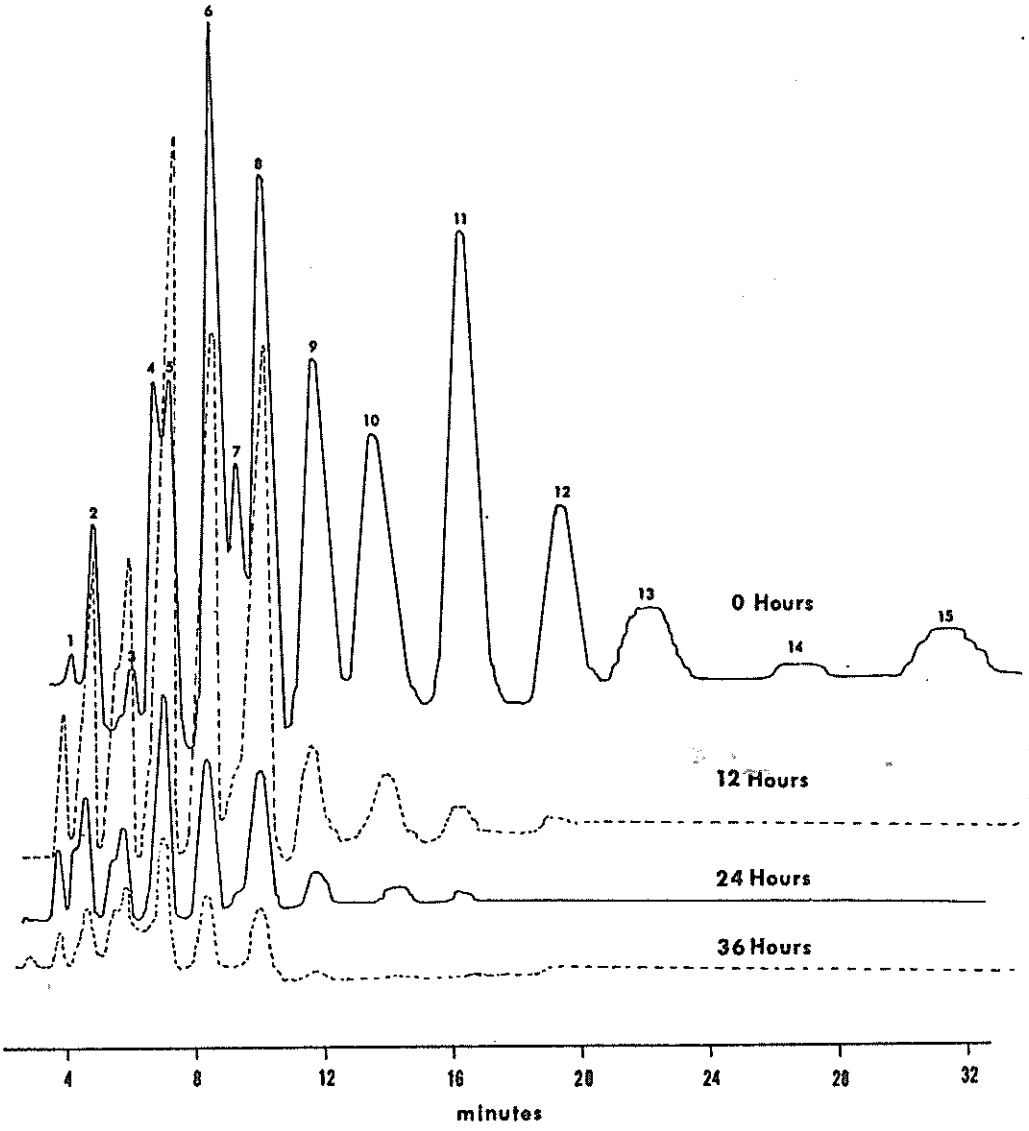


FIGURE 7. Gas chromatographs showing effect of sun-drying on PCB (Aroclor 1260) content in shrimp.

compound led to trichlorobiphenyl which in turn produced dichlorobiphenyl.

After 12 hours of sun-drying, 26.8% and 32.2% reduction of Aroclor 1254 and 1260 respectively was observed for sodium nitrite-treated shrimp compared to 14.1% and 18.8% in untreated shrimp respectively. This amounted to about twice as rapid reduction for treated sample than for the untreated sample during the first 12 hours.

The lower chlorinated biphenyls appeared at early stages followed by higher chlorinated biphenyls in the gas chromatograms of PCBs. Results from Tables V and VI indicated that solar radiation has a two-way impact on PCB degradation. The higher chlorinated biphenyls are broken down to lower chlorinated biphenyls on one hand and on the other, lower chlorinated biphenyls are degraded simultaneously. Also several hundred fold increase in some of the peaks, particularly tri and tetrachlorinated biphenyls indicated that there was an accumulation of breakdown products of higher chlorinated compounds. Peaks representing hexachlorobiphenyls were found to be most resistant to breakdown in Aroclor 1254.

The success of polychlorinated biphenyl degradation or detoxification is based on the dislocation or displacement of chlorine atoms clinging fast to the biphenyl molecules. Irradiation in presence of certain compounds capable of donating hydroxyl or hydrogen ions can partially or completely effect the displacement of chlorine.

It is also probable that PCBs yield "free radical" intermediates as suggested by Ruzo et al. (1974), which in turn explains

TABLE V
PERCENT INDIVIDUAL CHLORINATED BIPHENYLS OF AROCLOR 1254
LEFT AFTER SUN-DRYING OF SHRIMP

Chlorinated Biphenyls	Without Sodium Nitrite				With Sodium Nitrite			
	12	24 (Hours)	30	36	12	24 (Hours)	30	36
Dichloro-Biphenyl	0	0	0	0	0	0	0	0
Trichloro-Biphenyl	434.8	0	0	0	305.5	0	0	0
Tetra-Chloro-Biphenyl	856.0	0	0	0	851.7	0	0	0
Penta-Chloro-Biphenyl	23.7	13.4	0	0	37.0	0	0	0
Hexa-Chloro-Biphenyl	84.9	41.6	5.6	0	71.6	11.1	0	0
Hepta-Chloro-Biphenyl	0	0	0	0	0	0	0	0

TABLE VI
PERCENT INDIVIDUAL CHLORINATED BIPHENYLS OF AROCLOR 1260
LEFT AFTER SUN-DRYING OF SHRIMP

Chlorinated Biphenyls	Without Sodium Nitrite				With Sodium Nitrite			
	12	24	30	36	12	24	30	36
	(Hours)				(Hours)			
Tri-Chloro-Biphenyl	171.5	579.2	348.5	45.1	78.1	115.2	74.7	0
Tetra-Chloro-Biphenyl	176.1	219.4	125.6	19.5	63.9	28.0	39.9	0
Penta-Chloro-Biphenyl	0	0	0	0	0	0	0	0
Hexa-Chloro-Biphenyl	178.8	116.8	77.6	17.6	94.0	66.0	42.6	0

TABLE VI (Contd)
PERCENT INDIVIDUAL CHLORINATED BIPHENYLS OF AROCLOR 1260
LEFT AFTER SUN-DRYING OF SHRIMP

Chlorinated Biphenyls	Without Sodium Nitrite				With Sodium Nitrite			
	12	24	30	36	12	24	30	36
	(Hours)				(Hours)			
Hepta-Chloro-Biphenyl	45.9	17.8	11.0	3.9	30.8	10.1	7.5	0
Octa-Chloro-Biphenyl	0	0	0	0	0	0	0	0
Nano-Chloro-Biphenyl	0	0	0	0	0	0	0	0
Deca-Chloro-Biphenyl	0	0	0	0	0	0	0	0

the occurrence of dechlorination products. The resultant products may be hydrochloric acid or in some cases sodium chloride, although further study is essential to establish the formation of any product or products. Previous work by Crosby and Moilanen (1973) indicate that chlorinated aromatic compounds are reductively dechlorinated in organic solvents under ultra-violet radiation at sunlight wavelengths. Further study under similar conditions revealed that in aqueous media a similar replacement of the chlorine by hydroxyl as well as hydrogen ions takes place. This also accounts for the mechanism of hydroxylated product formation proposed by Safe and Hutzinger (1971). The degradation of PCBs in aqueous suspension further supports our view that the reduction of PCBs during sun-drying of shrimp is possible under polar as well as non-polar solvents and the evaporative aqueous media can provide basis for further reductive dechlorination to a certain extent.

The complete degradation of PCB in shrimp can thus be a phenomena including one or all of the following processes: (a) reductive dechlorination (b) isomerization and (c) polymerization. The chances that the reduction takes place solely by vaporization are minimal since other conventional methods of drying have negligible effect of PCB degradation. However, the possibility exists that PCBs when uniformly distributed in thin layers can be easily degraded under solar radiation. If proved, this phenomena will be of high ecological significance since PCBs may get widely spread on shores and other substrates as a result of deflooding and receding of contaminated waters.

Conclusion

It will be too early to predict what exact role sodium nitrite plays in the accelerated degradation of PCBs. It might have a direct or indirect impact by facilitating absorption of radiation of particular wavelength. But it will definitely prove beneficial considering its advantageous applications at low concentration in food preservation. The concentration of PCBs in foods is usually limited particularly in foods like shrimp which are low in lipid content. Also, the chances of pre-processing contamination are from polluted waters which in turn mostly adhere to the shrimp surface. Since sun-drying of shrimp is practiced in many parts of the world, this process will have an added advantage, as PCBs adhering externally or superficially will get automatically degraded, resulting in relatively safe product. This may prove to be true not only in the case of PCBs but also DDT and other chlorinated pesticides which are closely related to them chemically.

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ANALYSIS OF PETROLEUM HYDROCARBON CONTAMINATION IN SHRIMP

ABSTRACT

A rapid method to determine petroleum hydrocarbon contamination has been adapted for use in shrimp analyses. This method combines standard extraction and isolation procedures with identification and quantitation by gas-liquid-chromatography. Levels of contamination as low as 0.1 ppm can be routinely analyzed. In addition, the type of oil contaminant can usually be determined.

ANALYSIS OF PETROLEUM HYDROCARBON CONTAMINATION IN SHRIMP

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Introduction

The seriousness and potential hazard of oceanic oil pollution has come to the public attention only recently (Ehrhardt and Blumer, 1972). The persistance of biogenic hydrocarbons in the food chain has suggested that oil pollutants might remain in the ocean for long time periods (Blumer et al., 1970), become incorporated into the food chain, and eventually contaminate food products derived from the sea. In addition to possible contamination from pollutants found in the ocean, contamination of marine food products with oil pollutants during harvesting and transport can also occur.

Of the various procedures used to identify and quantitate petroleum hydrocarbons in marine organisms, gas-liquid-chromatography (GLC) has generally been found to be the most useful (Farrington, 1973; Adlard, et al., 1972). In addition to giving a "fingerprint" of the oil pollutant which can be used in determining the source of the contamination, GLC can also be conveniently used to quantitate the amount of hydrocarbon contaminant in a sample.

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This paper describes a rapid method for quantitating and identifying oil pollutants from shrimp, and illustrates the usefulness of this method by describing several studies in which it was used to analyze contaminated shrimp.

Materials and Methods

Extraction and Saponification

Thirty grams of shrimp from each sample were cut into small pieces, sonicated in 100 ml chloroform/methanol (1/2), and extracted by the method of Bligh and Dyer (1959). In some cases, shrimp were shelled and the shells and tissue were analyzed separately. Extracts were filtered through glass wool, rinsed with 100 ml chloroform, and the solvent reduced in volume by rotary evaporation at 40°C. The samples were saponified by refluxing in 20 percent potassium hydroxide in methanol for 4 hr. Fifty ml of water was added to the samples, and the non-saponifiable material was partitioned into hexane using three 50 ml hexane extractions. The solvent was reduced in volume to 5 ml by rotary evaporation at 40°C.

Column Chromatography

Separation of saturated hydrocarbons from aromatic and more polar components was accomplished by column chromatography on neutral alumina (100-200 mesh) over silicic acid (Bio Sil A, 100-200 mesh). Samples were placed on the column in hexane, and the saturated aliphatic and cyclic components were eluted with hexane, while the aromatic components were eluted with hexane:benzene (50/50). Separation of authentic standards by this method was verified by GLC. After elution, samples were concentrated by rotary evaporation, transferred to vials, and reduced in volume to near dryness under nitrogen at 40°C.

Gas-Liquid-Chromatography

Gas-liquid-chromatography was performed on either a 6 ft. x 1/8 in. 3 percent SE-30 column programmed from 150 to 280°C at 8° per min, and/or a 6 ft. x 1/8 in. 3 percent SP2250 column programmed from 100 to 285°C at 8° per min. A flame ionization detector was used, and quantitation was obtained by disc integration and/or planimetry, and compared with known amounts of standards.

All solvents used were redistilled from glass prior to use, and solvent blanks, control shrimp and spiked samples were run with each analysis. The procedures used are presented in a flow diagram in Fig. 1.

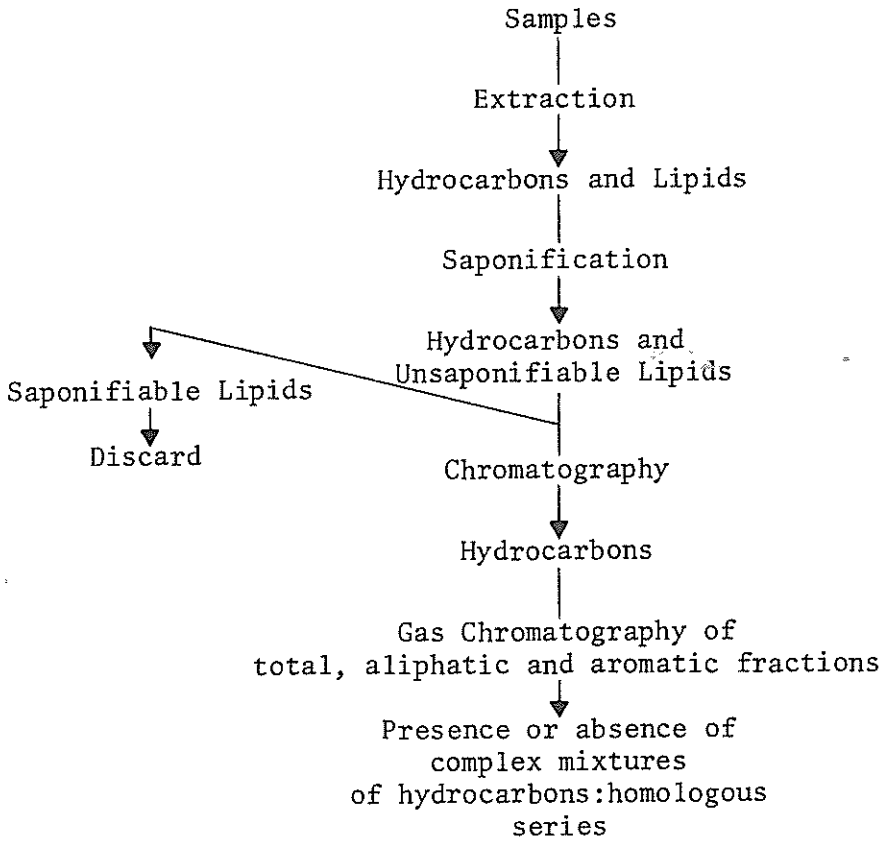

Fig. 1. Flow diagram for analytical techniques used to extract, detect, and analyze petroleum contamination in shrimp.

Results and Discussion

The presence of naturally occuring biogenic hydrocarbons in many marine organisms can interfere with analyses of possible petroleum contamination (Farrington, 1973). However, hydrocarbons from fossil fuels and pollution differ from natural, biogenic hydrocarbons from living organisms (Blumer, et al., 1970), and can be recognized by differences in odd to even carbon chain length ratios, carbon chain length distribution, and other parameters (Farrington, 1973). Results of this study from both control and contaminated shrimp showed that naturally occurring, biogenic hydrocarbons were not present (Fig. 2). In the techniques used, hydrocarbon concentrations as low as 1 µg per gr tissue would have been detected.

A number of different studies on petroleum contaminated shrimp were run, and the results of several of these which illustrate the type of data obtained are described here. One study involved the analyses of three groups of headless shrimp for concentrations of petroleum contamination and identification of the source of the pollutant present. Comparison of GLC traces of the non-saponifiable material from control and suspect shrimp (Fig. 2) showed a large, mostly unresolved background in the C_{21} to C_{35} range in the suspect shrimp that was absent in the control shrimp. A similar GLC trace was obtained from material isolated from the liquid associated with the suspect shrimp.

In order to identify the source of pollution, GLC traces from a number of possible pollutants were obtained (Arab crude oil, empire crude oil, hydraulic oil, crankcase oil, diesel oil). GLC traces of these, as well as a paraffin standard are shown in Fig. 3. The GLC

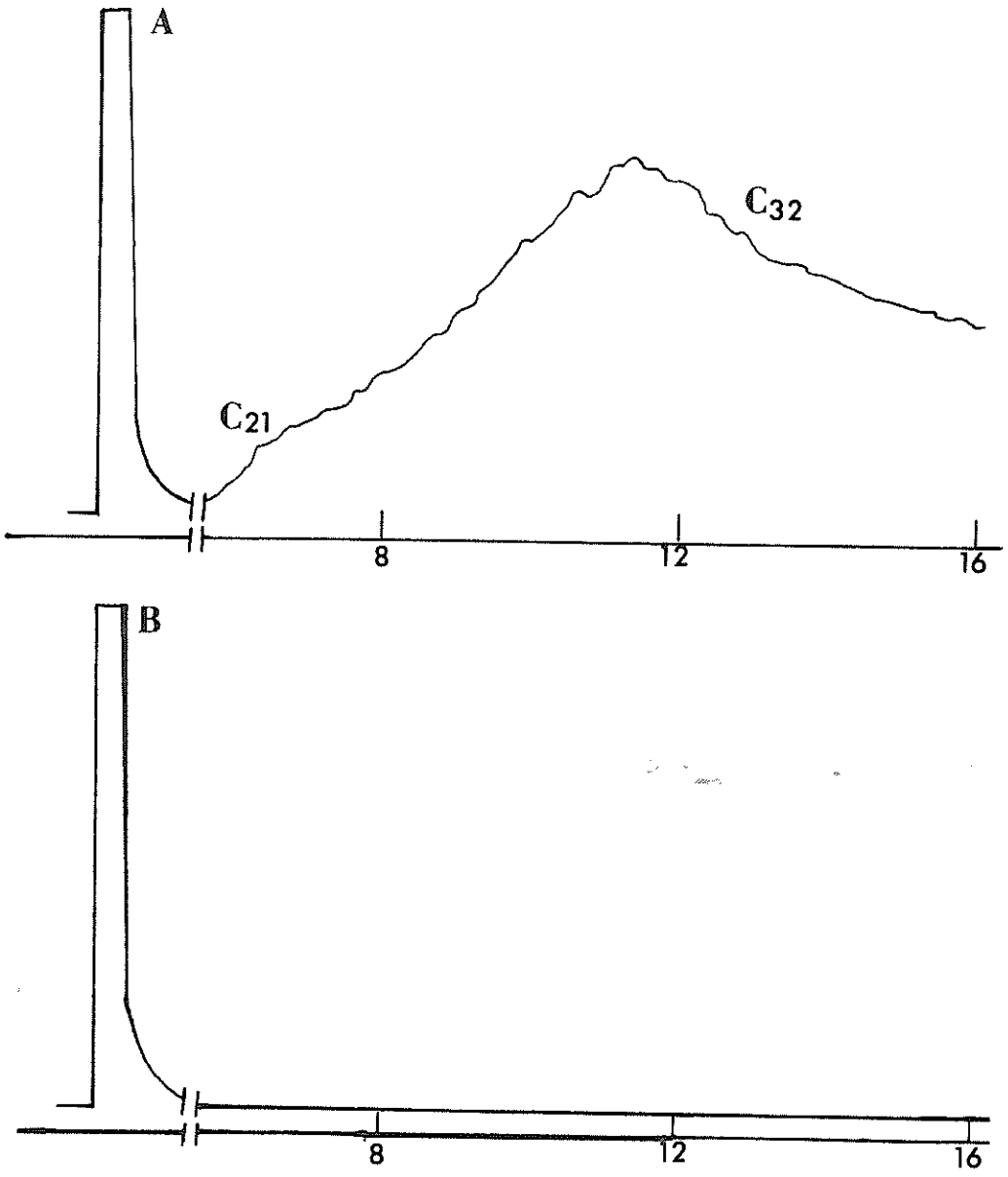

Figure 2. Gas-liquid chromatograms of contaminated (A) and control (B) shrimp.

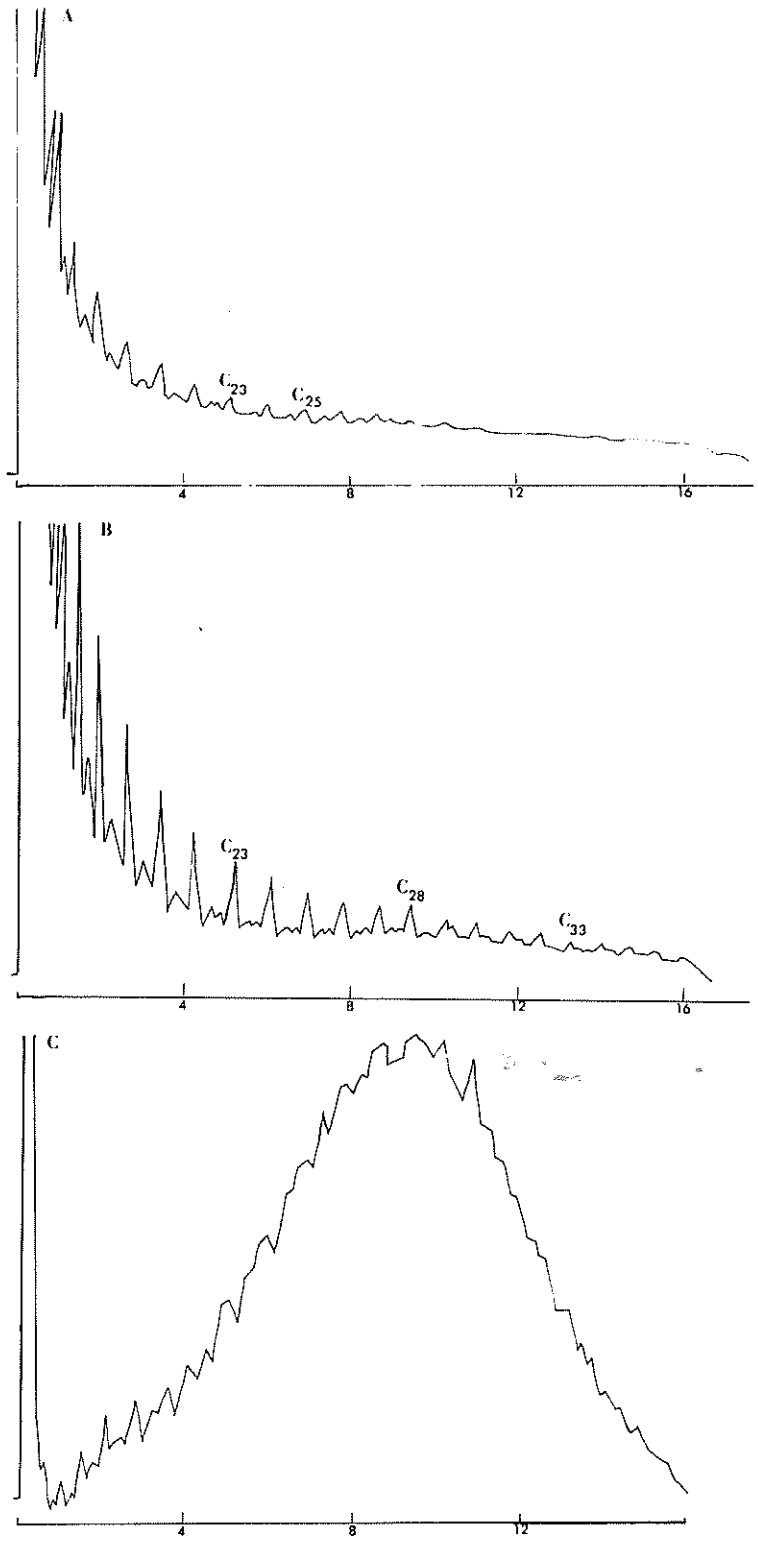

Figure 3. Gas-liquid chromatograms of empire crude oil (A), Arab crude oil (B), hydraulic oil (C), crankcase oil (D), diesel oil (E), and a paraffin standard (F).

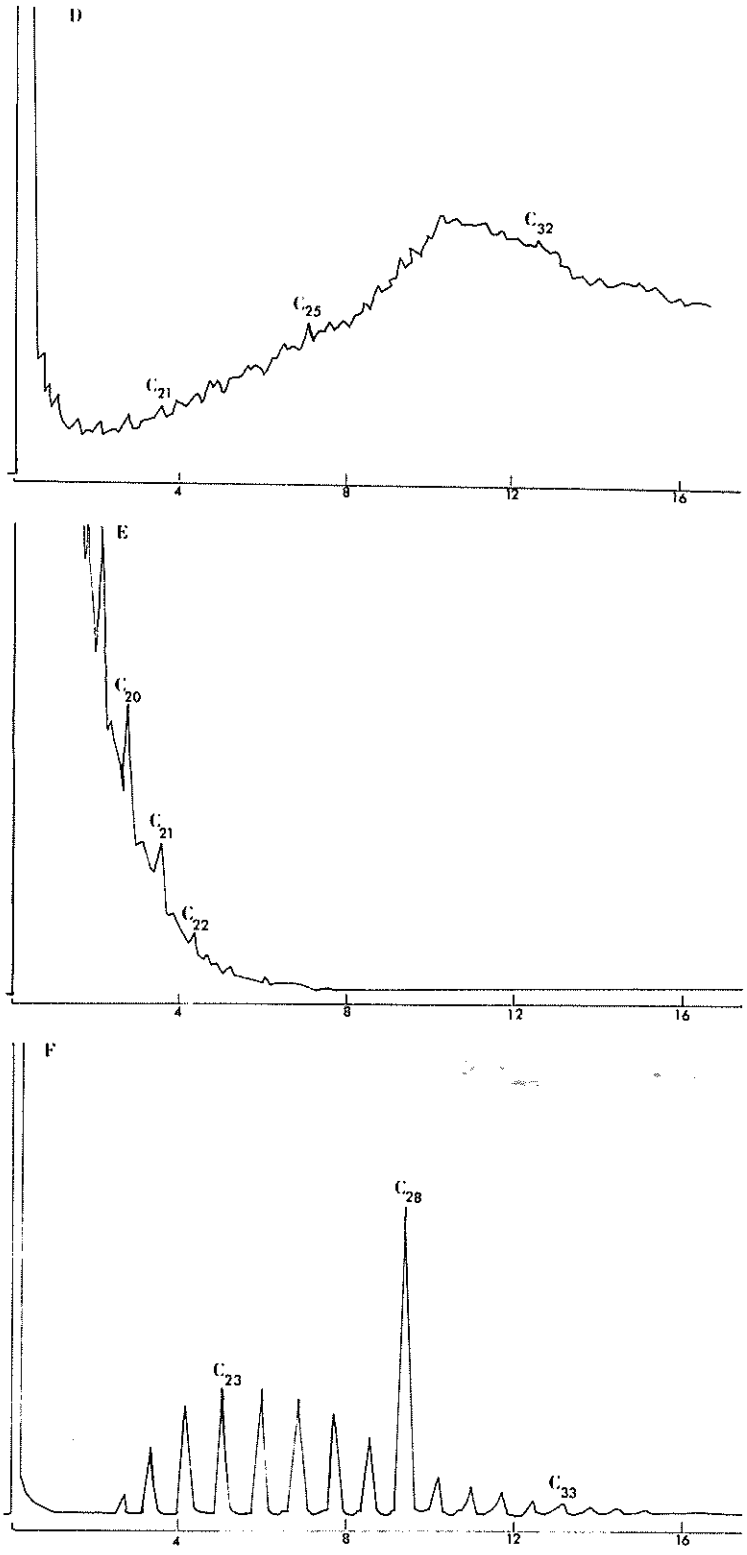

Figure 3. (Cont).

trace from the contaminated shrimp (Fig. 2) was found to be identical to that of crankcase oil. Quantitation by comparison of the area beneath the unresolved envelope with known amounts of crankcase oil indicated that 23 µg of pollutant per gr tissue was present.

Since pollution in the ocean by crankcase oil is seldom encountered, the possibility that contamination occurred during transport was considered. If contamination did occur during transport, it would be expected that much of the contamination would be associated with the cuticle rather than incorporated into the tissues. To determine if this was the case, the shrimp were peeled and the shells and tissue were extracted and analyzed separately. Results showed that almost all of the contamination was associated with the cuticle. Our conclusions that contamination by crankcase oil occurred in transit have since been verified by Glen Kiel (personal communication). The contamination was traced to the spillage of a 5 gallon can of crankcase oil onto shrimp in the hold of a shrimp boat.

In some studies of contaminated shrimp, the presence and amount of oil pollution could be determined, but an absolute source could not be assigned. Fig. 4 shows the GLC trace of a shrimp sample contaminated with aliphatic hydrocarbons in the C_{22} to C_{31} range over an unresolved background of about the same boiling range. These shrimp were caught in an area where a crude oil spill had occurred about a year earlier. Weathering and metabolism of a crude oil yields a GLC trace such as this, and thus probably reflects the fact these shrimp were caught in an area of a previous oil spill. However, a high boiling distilate would also exhibit a similar pattern, and thus the possibility of contamination in transit could not be completely ruled out.

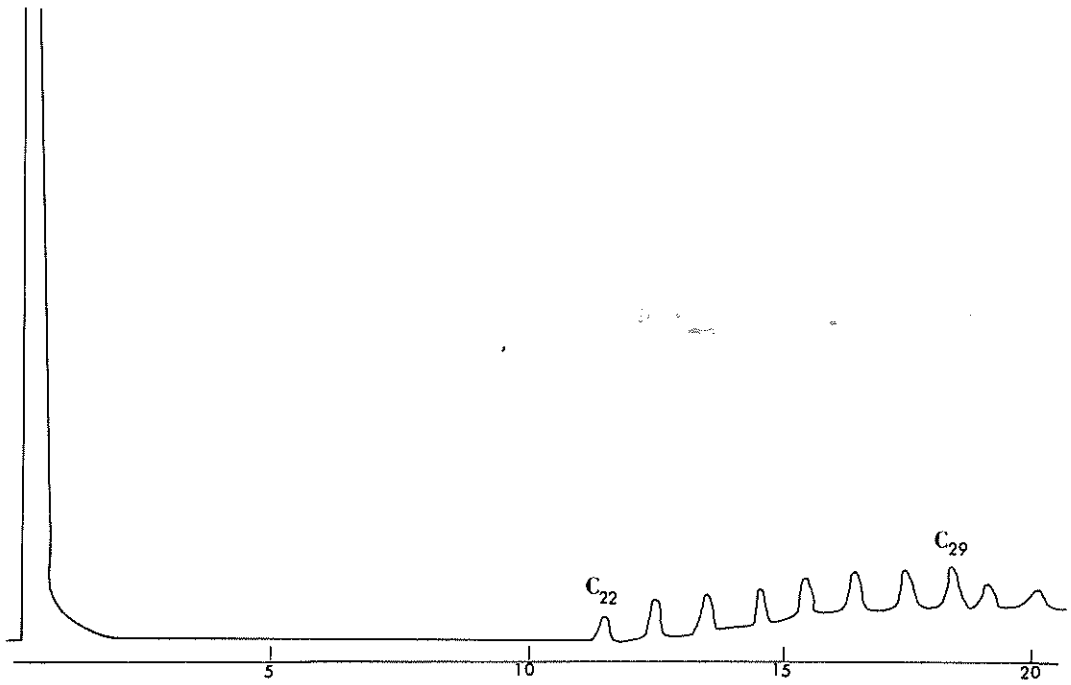

Figure 4. Gas liquid-chromatogram of contaminated shrimp.

Summary

The use of GLC for quantitating and identifying pollutants in marine food products is well documented and its usefulness has been further demonstrated in these studies. In some cases, such as the shrimp polluted by crankcase oil, the source of the pollutant could be readily determined. In other cases, with the absence of samples of the pollutant present in the ocean where the shrimp were caught or other possible contaminants, it is possible to indicate the general type of pollutant present but not possible to absolutely pinpoint the source of the oil contaminant. However, with the increased awareness of the toxicity (Blumer, 1970) of certain types of oil pollutants and the recognition that the aromatic components of oil can act as carcinogens (Miller, 1970), an indication of the general nature of the oil pollutant (i.e. aliphatic, cyclic and branched or aromatic) can certainly help in deciding whether a product is marketable.

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SODIUM BISULFITE AND ITS RESIDUAL USE IN CONTROLLING BLACKSPOT IN SHRIMP

ABSTRACT

The paper describes limited research activities which were undertaken by the National Fishery Products Inspection and Safety Laboratory to determine the impact of a proposed international tolerance or guideline of 30 ppm SO_2 residue in shrimp products on U.S. shrimp producers. Additionally, the paper describes the efficacy of a proposed FAO procedure in application of sodium bisulfite in meeting the tolerance requirements of the proposed international standard. The paper also describes an attempt to develop a more rapid field technique for accurately measuring SO_2 residues in shrimp products utilizing a specific ion electrode approach.

SODIUM BISULFITE AND ITS RESIDUAL USE IN CONTROLLING BLACK SPOT ON SHRIMP

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Introduction

The formation of black spot on raw headless shrimp is a result of the breakdown of tyrosine by the enzyme tyrosinase. Tyrosinase is present in large quantities in the digestive system of the shrimp, and must be removed from the headless shrimp by thorough washing or inactivated through the use of chemicals. The enzyme is spread to the headless shrimp, and, during iced storage, causes black spots to form on the meat and shell.

In 1957, researchers at the University of Miami (Camber et al, 1956) developed a method of treating fresh raw headless shrimp with sodium bisulfite to minimize black spot formation. They recommended and demonstrated the use of this effective chemical. After awhile, however, as we all know, boat crews decided that, "if a little did some good, a lot would do a lot of good." Consequently, misuse forced some buyers to shy away from buying shrimp treated with sodium bisulfite. Excessive treatment with sodium bisulfite discolors the shrimp, severely affecting their appearance.

There are three methods of application:

1. The first method, and most commonly used, is to dip freshly caught, deheaded shrimp in a tank containing a dilute solution of sodium bisulfite in seawater for a short time (about one minute).

2. The second method of common application (which in our judgment should not be used), is to sprinkle sodium bisulfite, as a dry powder, over the shrimp as they are being layered for icing in the vessel hold.

3. The third method is to use ice containing small amounts of sodium bisulfite.

All three methods have the same value in that each will control the enzymatic blackening if administered correctly.

As we have mentioned, the primary purpose for using sodium bisulfite is to allow the boats to stay at sea for an extended period of time and still land with a quality product. In the authors' opinion, however, today, sodium bisulfite is being used not only to control black spot but to mask poor quality shrimp and make them appear to have a higher value in both appearance and odor.

Such practices have caused some importing countries to set tolerances on the amount of total SO_2 residue in shrimp products. Most of these tolerances are for all additives that form SO_2 residues (i.e., sodium bisulfite, sodium sulfite, sodium hyposulfite, and sodium or meta bisulfite). During elaboration of the Codex International Standard for Quick Frozen Shrimps or Prawns (FAO/WHO, 1975), at one point in the procedure, a tolerance for

all of these additives of not more than 30 ppm total SO_2 in shrimp was contemplated.

Additionally, an advisory Codex Code of Practice for Shrimps or Prawns (FAO, 1975), which essentially is an international GMP, under evaluation suggested the use of sodium bisulfite by a dip method for one minute in a 1.25% solution.

To determine the efficacy of these two Codex proposals, as well as to develop a more rapid methodology to determine SO_2, the laboratory performed three limited experiments.

Selected Product for Sodium Bisulfite

Eighty shrimp samples from seven countries, including 40 U.S. domestic samples, were analyzed for SO_2 (Table 1). The samples were divided into shells and peeled meats. In some cases, sample size was limited. The Monier-Williams method of analysis (AOAC, 1975) was used in determining the residues. All samples were conducted in triplicate, and values are expressed as the mean.

Of these samples, only one sample of peeled shrimp and nine samples of shells, contained a residue in excess of the proposed limit of 30 ppm SO_2. One of the shell samples contained a residue of 575 ppm SO_2, and meats extracted from those shells contained an excessive level of 112 ppm SO_2. Another sample of shells had an elevated residue of 325 ppm, but, interestingly enough, no residual SO_2 was found in the corresponding meats. In all other samples having a significant level of SO_2 in the shells, the corresponding meats also contained the additive in lesser amounts.

Table 1.--SO_2 residue in shrimp from seven major shrimp-producing countries.

Sample Number	Country	Shrimp		Sample number	Country	Shrimp	
		Meat	Shells			Meat	Shells
		ppm $NaHSO_3$				ppm $NaHSO_3$	
1	U.S.A.	0	33	41	India	0	-*
2	"	0	34	42	"	0	-*
3	"	0	0	43	"	0	-*
4	"	0	325	44	"	0	-*
5	"	0	0	45	"	0	-*
6	"	0	195	46	"	0	-*
7	"	19	315	47	"	0	-*
8	"	16	208	48	"	0	-*
9	"	15	270	49	"	0	-*
10	"	0	0	50	"	0	-*
11	"	0	0	51	"	0	-*
12	"	112	575	52	"	0	-
13	"	0	-*	53	"	0	-
14	"	0	-*	54	"	0	-
15	"	0	-*	55	"	0	-
16	"	0	-*	56	"	0	-
17	"	0	-*	57	"	0	21
18	"	0	-*	58	"	0	13
19	"	0	-*	59	Kuwait	0	-*
20	"	0	-*	60	"	0	-*
21	"	0	-*	61	"	0	-*
22	"	0	-*	62	"	0	-*
23	"	0	-*	63	"	0	-*
24	"	0	-*	64	Mexico	0	-
25	"	0	-*	65	"	0	-
26	"	0	-*	66	"	0	-
27	"	0	-*	67	Guatemala	0	-
28	"	0	-*	68	"	0	-
29	"	0	-*	69	Trinidad	295	1,752
30	"	0	-*	70	"	159	1,625
31	"	0	-*	71	"	0	55
32	"	0	-*	72	"	33	743
33	"	0	-*	73	"	0	41
34	"	0	-*	74	Honduras	62	-
35	"	0	-*	75	"	16	78
36	"	0	-*	76	"	38	333
37	"	0	-*	77	"	24	82
38	"	0	-*	78	"	58	-
39	"	0	-*	79	"	66	430
40	"	20	152	80	"	27	406

*No shells, peeled and deveined shrimp

Eighteen samples of meats and seven samples of shells from India were analyzed. Two of the shell samples had residues which were significantly below the proposed Codex tolerance. No residues were detected in the meats.

The 10 samples from Kuwait, Guatamala, and Mexico had no detectable SO_2 residues in the meats or shells. It must be pointed out that these sample sizes were limited.

Twelve samples of shells and meats from Trinidad and Honduras had extremely elevated residue levels of SO_2. The shells ranged from 41 to 1,752 ppm with the meats ranging from 0 to 295 ppm. Seven of the meats and 10 of the shell samples had SO_2 residues in excess of the 30 ppm proposed Codex guideline. Admittedly, sample size for these analysis were limited, but it is pointed out that all samples were of a commercial nature and offered for sale.

It should be further pointed out that since the time this specific research was conducted, the Codex Committee on Fish and Fishery Products changed the proposed SO_2 guidelines referenced in this paper from 30 ppm to a limit of 100 ppm on raw meats and 30 ppm in cooked meat products. There is no limitation of shell residue. Therefore, of the total of 80 samples analyzed and discussed in this report, only three exceed the current SO_2 guideline in the latest proposed Codex International Standard for Quick Frozen Shrimps or Prawns.

Effects of Commercial Application of Sodium Bisulfite Residues

Another phase of the study was a limited laboratory project to dip freshly caught shrimp in sodium bisulfite with subsequent storage in a simulated "ice hold", and analyzing them after 0, 4, 7, and 11 days of storage.

The purpose was to determine the relationship between the dipping method (which is being recommended in the Codex Code of Practice for Shrimps or Prawns) and SO_2 residues. A second purpose was to determine the effect of cooking on SO_2 residues.

Ten pounds of raw headless shrimp which were harvested locally and known not to have been treated with sodium bisulfite were obtained. This sample was treated in a 1.25% solution of $NaHSO_3$ as described in the Codex Code of Practice for Shrimps or Prawns. All analysis were conducted by the method described above, performed in triplicate, and all values are expressed as the mean.

On each day of analysis, aliquots of the treated sample were removed from iced storage and divided into three categories for analysis, i.e., shells, peeled raw meat, and peeled cooked meat. For the peeled cooked meat aliquot, the peeled meat was cooked in water heated to a rolling boil for three minutes. Table 2 contains the results.

The shells had a concentration of 884 ppm SO_2 at 0 days storage which was reduced at a fairly constant rate to 159 ppm by the 11th day of iced storage. The raw peeled meats possesed a SO_2 concentration of 218 ppm which dropped sharply to 28 ppm by the end of four days of storage to 5 ppm by the end of the 11 days of iced storage.

Table 2.--SO_2 in freshly caught shrimp dipped in $NaHSO_3$ and stored at laboratory

Day	Shells	Peeled raw meat	Peeled cooked meat
0	884	218	44
4	317	28	36
7	191	23	5
11	159	5	4

In terms of the cooked meat samples, the initial concentration of SO_2 at 0 days was 44 ppm. This SO_2 concentration was sharply reduced by the end of 7 days of iced storage to 5 ppm, and further to 4 ppm by the end of the 11th day of storage.

Rapid Method Development

Inasmuch as the official AOAC method is encumbering and does not lend itself to field application, it was believed that a more simple method could be devised utilizing a specific ion electrode approach. For this an Orion Model 407 specific ion meter with a Model 95-64 SO_2 ion electrode was employed.

Shrimp treatment:--Non-sulphite-treated fresh shrimp were deheaded and immersed for 1/2, 1, and 2 minutes in a 1.25% and a 4% sodium bisulfite solution. These were allowed to drain for 5 minutes and stored at 5°C.

Standards and buffer:--The standard stock solution was 15.8 gm Na_2SO_3 (glycerin added as a preservative) which is 1,000 ppm in SO_2. The stock solution was prepared daily.

The buffer was 235 gm sodium sulfate (anhydrous), 31 ml (57.04 gm) concentrated H_2SO_4, and water added to one liter. The buffer was added to all solutions before measuring the potentials with 5 ml of buffer being added for each 50 ml solution or standards. This buffer in 1:10 dilution adjusted the pH to about 1.7 and the osmotic strength to about 0.5 osmolar.

Preparation of homogenates:--Two differing methods of homogenizing shrimp were tested: (1) the shrimp were blended with the water and (2) the shrimp were blended "dry" requiring care to ensure a properly macerated sample.

In the first method, standards were made by adding appropriate volumes of standard sodium sulfite solutions to 40 gm of shrimp, then adding water to make 200 ml, and blending this mixture for 20 seconds. Five standards of 1, 3, 10, 30, and 100 ppm were used. All samples analyzed had 5 ml of buffer/50 ml sample added to adjust the pH and osmotic strength.

The second method had only one difference--the shrimp were blended "dry", then the water, standard, and buffer were added, in that order.

Measurements at a lower pH:--Diluting the internal electrode solution (IES) 1:10 lowers the detectable concentration of SO_2 (Orion Research Incorp.). This requires using only 0.1 volume of buffer to preserve osmotic strength.

The IES was diluted 1:10 with degassed distilled water and stored under nitrogen. A concentrated solution of NaCl was prepared so that the addition of 0.1 ml of this to the 2.4 ml aliquots of IES yielded an approximately 0.1M salt solution. This restored the osmotic strength of the solution. pH readings were made with a Corning Model 10 pH Meter.

Electrode storage:--The electrode was stored in a buffer of 4.4 gm Na_2SO_3 and 1.5 ml concentrated HCl diluted to 250 ml with water to prevent excessive exposure of the membrane and IFX to air as suggested in a personal communication with the manufacturer (Frant, 1973).

Millivolt readings:--The probe was placed in samples at an angle to avoid trapping air bubbles under the end of the electrode.

The samples were stirred with a magnetic stirrer as rapidly as possible without forming a vortex.

Results:--Analysis of shrimp homogenized in water showed erratic results which are tabulated in Table 3. Perusal of the results indicate little reproducity between the known concentrations of the SO_2 homogenates and the residues as measured by the specific ion electrode. For example, 3 homogenates containing 10 ppm SO_2 gave varying millivolts of 100, 90, and 63, thus making it impossible to plot any type of acceptable curve. Dry-blended homogenates were equally as frustrating. In the authors' opinion, one reason for the disparity of results could be that air is whipped into the shrimp homogenate, rapidly oxidizing the sulfur dioxide and its related compounds to sulfate, thus negating the utility of the SO_2 ion electrode.

Conclusions

The currently proposed Codex guidelines for SO_2 residues in shrimp products should be acceptable to the domestic shrimp-producing industry.

As evidenced from the limited data, foreign producers need to pay particular attention to the application of $NaHSO_3$ in shrimp products.

The dipping method for $NaHSO_3$ application aboard vessels proposed in the Codex Code of Practice for Shrimps or Prawns should present no residue problems.

A more rapid method in analyzing for SO_2 is needed, and the use of a specific ion electrode may be one approach. Before this can be verified, however, considerable research is needed in adapting that method to shrimp homogenates.

Table 3.--Analyses of treated homogenates containing 1 to 100 ppm SO_2

SO_2	MV	SO_2	MV	SO_2	MV
ppm		ppm		ppm	
1	to 0	1	to 0	1	to 0
3	30	3	50	3	40
10	100	10	90	10	63
30	120	30	120	30	86
100	150	100	140	100	100

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UTILIZATION OF RECOVERED SHRIMP PROTEIN AS A PIGMENT SOURCE FOR SALMONIDS

ABSTRACT

A process was developed for the recovery of shrimp protein as a by-product of a chitin recovery operation. The protein was extracted and precipitated from shrimp waste by acid and heat treatment. The coagulum was found to be 80.5% water and 7.6% ash, 10% calcium and 74% protein on a dry weight basis. It was low in bacteriological counts and ordor and compared favorably to reported amino acid profiles for other shrimp meals. The meal and pigment extract of the meal was fed to rainbow trout. After two weeks, significant amounts of astaxanthin and its isomers were incorporated into the flesh and skin of the trout. The results show that the shrimp coagulum could be used as a pigment and protein source for salmonids raised in aquaculture.

UTILIZATION OF RECOVERED SHRIMP PROTEIN AS A PIGMENT SOURCE FOR SALMONIDS

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INTRODUCTION

In terms of dollars, shrimp is an extremely valuable fishery. Current domestic and imported shrimp amount to more than a half billion pounds per year (Meyers and Rutledge, 1971; 1973). In processing shrimp the head and hard carapace are removed during semi-mechanized peeling operations. Waste material accounts for approximately 70% of the whole shrimp and may approach 85% in the processing of other crustaceans such as the Atlantic deep sea red crab. Thus these food processing industries are faced with the problem of disposing of waste products or converting this waste material into economic by-products. These waste products must compete with products that they replace or else create their own market. Shrimp meal is traditionally sold on the basis of its protein content and not on its astaxanthin content. Lambertsen and Braekkan (1971) studied

the astaxanthin content of a number of industrial shrimp meals. It was found that the astaxanthin level was generally low or even absent in commercial shrimp meals. The vacuum-dried meal contained three times more astaxanthin than the best industrial sample.

A number of studies have shown that astaxanthin is the typical pigment in salmonids (cf. Amano et al., 1968; Kanemitsu and Aoe, 1958; Crozier, 1974; Steven, 1947). It was also shown from the stomach contents of salmon (Manzer, 1968) and trout (Needham, 1969) that these fish consume crustaceans, insects and other organisms rich in astaxanthin. It is thus not surprising that a number of authors have shown that shrimp and crab waste can be used to pigment trout and salmon (cf. Spinelli et al., 1974; Steel, 1971: Saito and Regier, 1971; Kuo, 1976).

The limiting factor in the use of crustacean meals as protein supplements has been the large proportion of exoskeleton material in the meal and in most cases, the taste of the product. Although the protein level on a dry weight basis varies from 25-40%, the high calcium level may have a deleterious effect on the overall nutrition.

In the present paper, protein was precipitated from shrimp waste. This coagulum was tested for its amino acid content, bacteriological contamination and calcium and astaxanthin levels. The whole coagulum and a pigment extract of the coagulum were fed to trout to test the bioavailability of the pigments and protein.

MATERIALS AND METHODS

Preparation of shrimp protein coagulum

The mechanical separation of the proteins from shrimp processing waste

was accomplished by an initial grinding to produce a partical size no greater than 10 mm. The resulting slurry was passed through a Baader Fish Bone Separator to remove the shell material. The protein slurry was acidified with acetic acid to the isoelectric point of the protein (pH 4.5), treated with 0.1% TBHQ Tenox (Eastman Chemical) as an anti-oxidant and allowed to coagulate for one hour at 80°C in a stainless steel tank. The coagulum was dewatered by filtering in cloth filter bags or centrifuged in a Sharples P-600 Horizontal Bowl Centrifuge. The coagulum was packaged in air tight bags, frozen and shipped by air freight in dry ice to Rhode Island.

The characteristics of the coagulum were determined by several independent laboratories as well as by MCI and URI. Amino acid analysis was performed by Texas A&M University laboratories (Nickleson, 1976) according to the method of Cobb and Hyder (1972) and employed a fully automatic 120C Spinco Amino Acid Analyzer. Microbial counts were performed by Texas A&M University laboratories (Nickleson, 1976) using the method of Surkiewicz (1966). Moisture, ash and protein were determined by the AOAC methods (1970).

The carotenoids were extracted and identified by the method of Saito and Regier (1971) and Kuo et al. (1976).

Carotenoid extract diet

A commercial trout feed manufactured by Zeigler Bros. Feed Mills, Inc. of Pennsylvania was used to feed the control fish. This diet was found to contain 35.3% crude protein, 6.3% crude lipid, 4.9% fiber, 10.4% ash, 10.0% H_2O and 33.1% nitrogen free extract (Kuo et al., 1976), plus amounts of β-carotene (1μg), α-cryptoxanthin (0.4μg), zeaxanthin (0.1μg) and

lutein (0.8μg). Astaxanthin was not isolated from this diet (Kuo et al., 1976). The fish were fed this feed at the hatchery prior to our purchase and the feed was continued as a control.

The desired amount of carotenoids extracted from the frozen shrimp protein was dissolved in a small amount of petroleum ether (PE) and this was added to the control pellets. The PE was removed under vacuum in a flash evaporator at 40°C. The pigmented diet was kept in a dark refrigerator prior to feeding.

Shrimp protein diet

The shrimp protein was mixed with other nutrients to constitute 10% of the mixture by dry weight. The diet used was based on the Oregon Test Diet (Lee et al., 1967) and is given in Table 1. The procedure was similar to that used by Kuo et al. (1976).

Fish culture

Rainbow trout (Salmo gairdneri) were purchased from American Fish Hatchery, Rhode Island and held in 125 gallon fiberglass tanks (Sims Fiberglass, Corvallis, Oregon) with flowing water (2 gal/min) and supplemental aeration. The water temperature was maintained at 11°C and the O_2 level was maintained at 8 ppm (cf. Lee et al., 1967; Kuo et al., 1976). Sixty fish of 140 g average size were distributed evenly between the four tanks. After 30 days, the fish were fed the diets at the rate of 3% of body weight per day.

Analysis of carotenoids in fish

The fish were sacrificed after the feeding periods. The fish flesh

was cut into pieces and ground in a Waring Blender with acetone. The acetone extract was transferred to PE by the addition of water and dried over anhydrous sodium sulfate. The pigments were separated on Microcel-C column with PE and 1-4% acetone in PE as the developing solvent. Each fraction was saponified by adding an equal amount of 20% KOH in methanol. The mixture was heated on a steam table for 10 minutes in the dark. After saponification the pigments were transferred to diethyl ether by the addition of water. Acetic acid was added to bring astacene to the epiphase. The pigment solution was dried over sodium sulfate and rechromatographed on Microcel-C and MgO:Hyflo Supercel columns. The individual bands were collected and purified on silica gel G sheets. Identification of the pigments was the same as described above (Kuo et al., 1976).

RESULTS

The protein coagulum was found to have the following analysis: moisture 80.5%; ash 1.5%; calcium 0.2%; and protein 14.4% (2.2% N x 6.25 uncorrected for chitin N). Table 2 shows the amino acid profile of the protein coagulum compared to casein and to other crustacean meals. The protein coagulum is marginally different from other shrimp meals. The shrimp protein is higher than casein in arginine, aspartic acid, glycine; lower in valine, glutamic acid and proline; and similar in the other amino acids. Tryptophan and cystine were not analyzed by this method. The protein coagulum gave a calculated PER of 2.709.

Bacteriological analyses on the coagulum were conducted by Texas A&M University laboratories on similarly prepared products. These results are

shown in Table 3.

The carotenoid content of the shrimp meal as received from MCI was determined on the frozen samples and on a tray dried sample (Table 4). The frozen sample contained trace amounts of the typical plant pigments lutein and zeaxanthin. These pigments were the main pigments in the fish as a result of the commercial feed that was used. Other minor pigments were isolated which are characteristic of crustacea (Simpson and Chichester, 1976). Astaxanthin and astaxanthin ester together constituted the main pigments.

A rather large amount of astacene was also isolated presumably as a result of the various handling and processing steps prior to freezing. It can be seen that the tray drying (70°C, 12 hrs.) of the coagulum resulted in a great loss of pigmentation. While the dried protein was red it contained only small amounts of astaxanthin and astacene.

A pigment extract was made of the frozen shrimp meal and this was fed together with a commercial ration. Table 5 shows the results of feeding the shrimp protein extract for two weeks. Astaxanthin, astaxanthin ester and astacene were absorbed in about equal amounts. The higher levels of the feed pigments (lutein and zeaxanthin) found in the fish were unexpected and may represent an increased absorption of pigments due to higher lipid levels.

Table 6 shows the results of feeding a 10% shrimp protein diet for two weeks. This diet contained less than 1/10th the amount of pigment contained in the 20 mg % diet. These results show that the fish also contained somewhat less than 1/10th the amount of pigment. We expected this level to be far lower since some difficulty was experienced in

getting the fish to take either the control or the shrimp meal diet. Initial studies with a diet of corn oil as a lipid source supplemented with ω-3 containing lipids was not eaten by the fish. The results reported here were with a fish diet containing herring oil. The initial mean weight of the fish was 140 g. At the end of the feeding trials the control fish and the fish fed the two pigmented diets were on the average of 174-175 g.

DISCUSSION

The total utilization of crustacean waste has been the subject of the reports of numerous investigators. The waste is composed of calcium carbonate, chitin, protein and carotenoids. Variation in proximate analyses will depend on the species, sex, metabolic state and subsequent processing, handling and storage. The shrimp protein concentrate used in these experiments was prepared by MCI as a by-product of a chitin-chitosan recovery procedure. Normally an alkaline digest is used for the recovery but experiments were conducted with a mild acid coagulation since alkali is known to convert astaxanthin to astacene. The resulting product was found to be low in calcium and bacterial contamination and high in protein quality and astaxanthin ester-astaxanthin content.

The results of these experiments show that the protein or a pigment extract of the protein would be effective in the pigmentation of salmonids.

It has long been assumed that astacene is an artifact produced during extraction of astaxanthin and does not occur as such in trout. The results shown in Table 5 suggest that astacene may be incorporated into trout. Further work is in progress in the preparation and feeding of pure astacene.

Longer studies would have to be conducted to assess the nutritional

problems, if any, of feeding the coagulum. Its amino acid content would appear to be similar to other shrimp meals.

Longer feeding trials would also be necessary in order to obtain a proper level of pigmentation. The results reported here show that the pigment is readily deposited in the flesh and fins. The pigment fed fish were easily distinguished from the control fish after only two weeks of feeding.

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Table 1. Composition of control and 10% shrimp meal diets

	Control %	10% Shrimp Meal %
Casein (vitamin free)	49.4	42.21
Gelatin	8.7	8.70
Dextrin	15.6	15.60
Carboxymethyl cellulose	1.3	1.30
Mineral mixture	4.0	3.30
Vitamin mixture*	2.0	2.00
Marine oil	10.0	10.00
Vitamin E (250 IU/g)	0.6	0.60
α-Cellulose	6.5	3.75
Choline chloride	1.0	1.00
KH_2PO_4		1.54
$CaCO_3$	0.9	
Shrimp meal		10.00

*Vitamin mix was prepared according to Steel (1971).

Table 2. Amino acid profile (as gAA/16g Nitrogen) of freeze-dried shrimp coagulum and other proteins

Amino Acid	Casein[1]	ISP[1]	Sundried Shrimp meal[2]	Spray-dried Shrimp meal[3]	Freeze-dried Coagulum
Arginine	4.07	7.8	6.31	8.06	6.31
Histidine	3.02	2.5	1.90	2.97	2.09
Isoleucine	6.55	4.9	3.26	5.17	4.68
Leucine	10.05	7.7	7.57	8.14	8.10
Lysine	8.01	6.1	6.17	8.34	7.36
Methionine	3.08	1.1	2.84	2.60	2.62
Phenylalanine	5.39	5.4	4.56	5.05	4.01
Threonine	4.28	3.7	4.28	3.91	4.65
Tryptophan	1.33	1.4	1.26	6.73	
Valine	7.39	4.8	4.42	5.62	5.02
Tyrosine	5.82	3.7	3.64	3.61	3.31
Alanine	3.35	3.9	5.29	7.14	7.11
Aspartic Acid	7.39	11.9	10.74	8.63	11.13
Cystine	0.38	1.2	1.59		
Glutamic Acid	23.05	20.5	15.46	17.8	14.38
Glycine	1.99	4.0	4.29	7.52	6.45
Proline	11.75	5.3	3.44	4.54	7.05
Serine	6.65	5.5	4.53	4.69	5.20

[1]Toma (1971)
[2]Meyers and Rutledge (1973)
[3]Johnson and Peniston (1971)

Table 3. Bacterial analyses of wet and dried coagulum*

Culture type	Wet coagulum	Dry coagulum
APC	1.0×10^3 CFU/g	6.0×10^3 CFU/g
Coliforms	< 3/g	< 3/g
E. coli	< 3/g	< 3/g
Staphylococcus	< 3/g	< 3/g
Salmonella	negative	negative

*Test performed by Texas A&M University, College Station, Texas.

Table 4. Composition of carotenoids in shrimp meal before and after drying

Carotenoids	Before drying (μg/g dry basis)	After tray drying
3,3'-Dihydroxy-ε-carotene	1.36	
Echinenone	0.25	
Isocryptoxanthin	0.38	
Canthaxanthin	0.39	
4-Keto-4'-hydroxy-β-carotene	0.74	
Dihydroxypiradixanthin (?)	4.66	
Lutein	trace	
Zeaxanthin	trace	
Astaxanthin ester	66.1	10.3
Astacene	55.4	6.26
Astaxanthin	7.19	trace

Table 5. Composition of carotenoids in fish fed 10% shrimp meal diet (for two weeks)

Carotenoids	Control (µg/g dry basis)	10% Shrimp meal
α-Cryptoxanthin	trace	trace
Canthaxanthin	-	trace
Lutein	1.3	1.76
Zeaxanthin	0.16	0.21
Astaxanthin ester	-	0.075
Astacene	-	trace
Astaxanthin	-	trace

Table 6. Composition of carotenoids in fish fed 20 mg % pigmented diet (for two weeks)

Carotenoids	Control (µg/g dry basis)	20 mg % Pigmented
α-Cryptoxanthin	0.10	0.18
Canthaxanthin	trace	trace
Lutein	1.79	2.04
Zeaxanthin	0.75	0.9
Astaxanthin ester	-	0.30
Astacene	-	0.28
Astaxanthin	-	0.28

THE PRESENT STATUS OF THE SHRIMPING INDUSTRY

John P. Mitchell
National Shrimp Congress
Rockport, Texas

Ladies and Gentlemen, it is a great privilege for me to visit with you today at the First Annual Tropical and Sub-tropical Fisheries Technological Conference. I am certain that the Conference and those to follow will make valuable contributions to the scientific, economic and sociological knowledge so necessary to properly manage our renewable marine resources. Without adequate data in a multitude of diciplines we cannot hope to manage these resources on an optimum yield basis, and depletion of certain fishery stocks will result as it already has in the case of several important commercial and recreational species.

Although commercial fisheries was America's first industry, it was not until the late 1940's that the shrimp industry as we know it today came into being with the discovery of the brown and pink shrimp off the coast of southern Texas and Mexico. Prior to this time the shrimp fishery in the Gulf of Mexico was limited to white shrimp in the bays and the in-shore Gulf near passes. When brown shrimp first appeared on the American market the consumer regarded them as inferior on the basis of coloration. This prejudice was soon overcome and the

fledgling Gulf shrimp industry was underway.

By 1967 the shrimp industry had become the nation's most valuable fishery with landings valued at over 100 million dollars. In 1973, Philip Roedel, Director, NMFS, referred to the American shrimp industry as "A Billion Dollar Industry."

These were encouraging words, but troubled waters lay ahead. The fuel crisis of mid-1973 was to have an enormous impact on all segments of the shrimp industry and in particular the fuel intensive shrimp fisheries. Processors, particularly those with large inventories or frozen shrimp products, also suffered great losses. The shrimp producers in the Gulf of Mexico were caught in an economic crunch unlike any experienced in their history. Not only did fuel increase from 11¢ a gallon to more than 30¢ a gallon, but all equipment and supply items used by the fishermen began escalating at a staggering rate. Consumer resistance in the market place and increased imports soon resulted in an unbalance between supply and demand. The result was a decline in ex-vessel prices paid for shrimp which was to last until mid-1975. During this period most Gulf shrimp producers operated their vessels at a loss. Most of them continued to fish their boats in order to keep their crews working. However, a number of vessels were sold. Some of those sold were converted to other fisheries in the U.S., others were added to the shrimp fishery in foreign waters.

The U.S. imports from 51% to 56% of the shrimp marketed in this country. Mexico is the leading exporter of shrimp to

the U.S. with an annual average of about 80 million pounds. Therefore, it becomes obvious that a weak market in the U.S. is as bad for Mexico as it is for the U.S.

THE SHRIMP RESOURCES IN THE GULF OF MEXICO

Three species of shrimp are presently harvested commercially in the Gulf of Mexico. They are white, pink and brown shrimp with brown shrimp the most abundant and, therefore, the most important. Two other species, the royal red and rock shrimp, are also available. Some commercial utilization is currently being made of rock shrimp. Very little is known about the royal red shrimp. This species is found in very deep water and is not now utilized by the shrimp industry because of the substantially higher costs of production over other species.

SHRIMP UTILIZATION

U.S. landings are published annually by the National Marine Fisheries Service. Landings in 1974 of 369.6 million pounds were worth $177.9 million. The Gulf States accounted for 50% of the total U.S. shrimp landings and 77% of the total value. Landings and value figures are not yet available for 1975, but it is well known in both industry and government circles that landings are down substantially although prices at the dock have improved.

Very little is known about Cuban shrimp landings. For the past two years a fleet of about 30 Cuban trawlers have been observed fishing off the coast of Texas and Mexico and were thought to be exploratory efforts.

FUTURE OF RESOURCE AND UTILIZATION

Under the present 12-mile fisheries zones as recognized by Mexico and the U.S., the shrimp resources in the Gulf of Mexico are in no danger of depletion. Since shrimp are an annual crop and are subjected to a number of environmental influences, their abundance varies from year to year in the different regions of the Gulf. Annual landings and the size of the U.S. shrimp fleet will probably remain at about the present level. Mexico and Cuba are expanding their shrimp fleets. It is not known, however, how many vessels may enter the fishery or what effect such additional pressure will have on the resource.

The unilateral extension of the U.S. and Mexican fishery zone creates a major problem for the U.S. shrimp fleet in the Gulf of Mexico, particularly for those vessels based in the lower part of Texas. Many of these vessels fish almost exclusively in waters between 12 and 20 miles off the Mexican coast. Should they be denied these historical fishing rights and be forced to return to American waters, over-competition for the resource would result with reduced landings per vessel and obviously reduced income to fishermen.

The domestic shrimp producing industry has made determined efforts in 1975 and 1976 to have quotas placed on shrimp imports, such quotas to be on historic basis of 1971 to 1973 imports. This would permit orderly imports which would satisfy consumption demands, but would prevent dumping of foreign shrimp in this country. Such protection is needed to give stability to the industry and will be most important when the industry asks for loans from financial institutions. A small tariff is also being requested, the proceeds from this being channeled to marketing studies and sales promotion of shrimp. Numerous hearings have been held by the International Trade Commission, and their findings will be made known in mid-May.

Of great importance to this industry is the Amendment S961 to HR 200, the 200 mile limit bill. This amendment, known as the Bentsen Amendment, would provide for embargo on specific fishery products from foreign countries which refuse to permit, or bargain in good faith to permit U.S. Flag Vessels from fishing in waters which they have historically and traditionally fished.