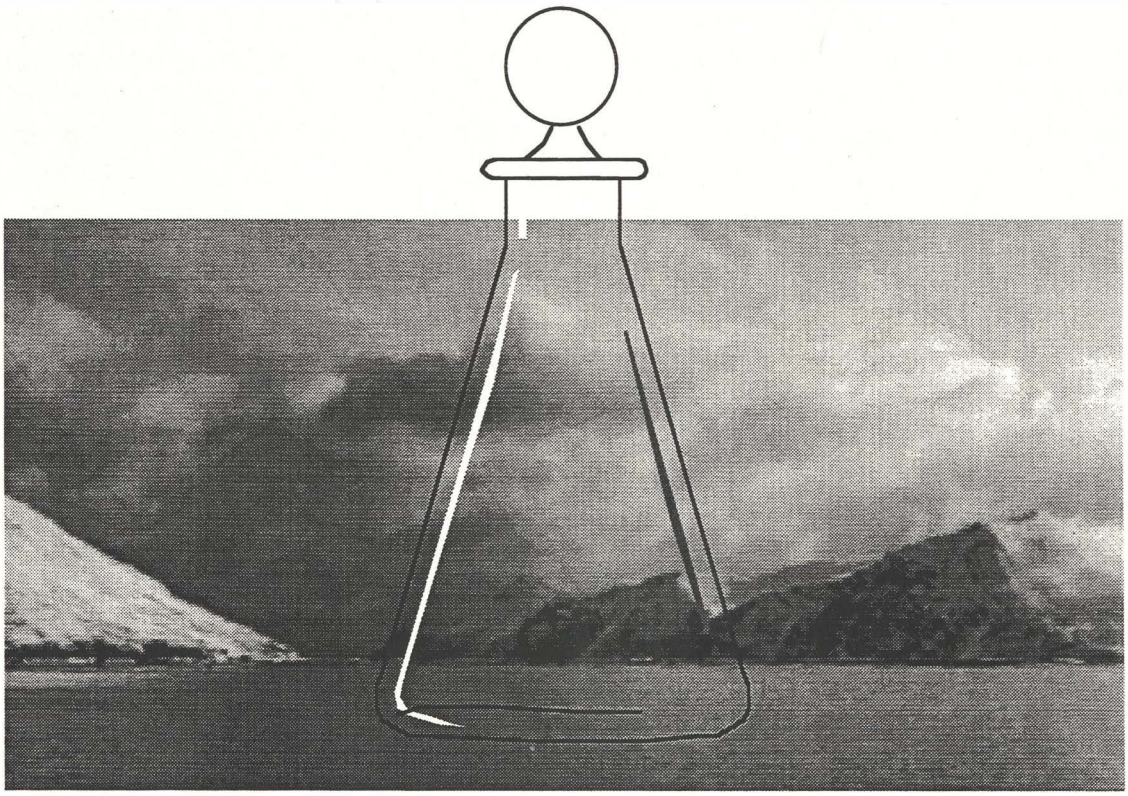
ɔAA Technical Memorandum NOS NCCOS CCMA 143

ıtional Status and Trends Program
r Marine Environmental Quality

NOAA/NRC Intercomparison for Nutrients in Seawater



Dutch Harbor, Aleutian Islands, Alaska, 1953. Rear Admiral H. D. Nygren, NOAA Corps (ret.) (NOAA Photo Collection, NOAA Central Library)

Silver Spring, Maryland
May 2000

US Department of Commerce

noaa NATIONAL OCEANIC AND ATMOSPHERIC ADMINISTRATION

Center for Coastal Monitoring and Assessment
National Centers for Coastal Ocean Science
National Ocean Service

ABSTRACT

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This report, prepared by the National Research Council of Canada (NRC), summarizes the results of an *Intercomparison for Nutrients in Seawater* under the directive of the Center for Coastal Monitoring and Assessment. Thirty participants were included in the exercise, including NOAA, USEPA, University, State and Canadian laboratories. Two 50 ml samples of a stabilized open ocean seawater were sent by NRC to each participant. The analytes to be determined were silicate, phosphate, nitrite and nitrate+nitrite. An assigned mean and confidence interval was calculated for each analyte and laboratory biases were identified. Results were in good agreement with the assigned values.

National Research Council Canada
Conseil national de recherches Canada

Institute for National Measurement Standards
Institut des étalons nationaux de mesure

NRC · CNRC

NOAA/NRC Intercomparison for Nutrients in Seawater

May 2000

Scott Willie and Vincent Clancy

Prepared for the
Center for Coastal Monitoring and Assessment
National Centers for Coastal Ocean Sciences
National Ocean Service
National Oceanic and Atmospheric Administration

Canada

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APPENDICES

1. INTRODUCTION

This is the first intercomparison exercise for nutrients in seawater organized by the National Research Council of Canada (NRC) on behalf of the Center for Coastal Monitoring and Assessment of the National Oceanic and Atmospheric Administration (NOAA), National Centers for Coastal Ocean Sciences (NCCOS). The purpose of this exercise was to assess the capabilities of a number of NOAA and other laboratories to analyse seawater for orthophosphate, dissolved silica, nitrite and total oxidised nitrogen (nitrite + nitrate). The NOAA office invited various laboratories to participate and NRC was asked to coordinate the sample distribution, collect the results, analyze the data and, following distribution of a final report, arrange a means for laboratories to discuss the results.

The test material distributed by NRC was:

> **MOOS-1**, a proposed certified reference material for nutrients in seawater. This water was collected at Lat. 47.062833 °N, Long. 59.982333 °W, off the northern tip of Cape Breton Island. The water was collected from a depth of about 200 meters using a rosette containing 22 Niskin bottles of about 10L each. Two casts were made. The contents of each Niskin were transferred, by means of a peristaltic pump, through a 0.05 µm cartridge filter into 50L carboys. The water was returned to the NRC laboratories in Ottawa, Ontario and homogenized in a 400L tank. Fifty mL subsamples were aliquoted into precleaned plastic bottles, sealed, and gamma irradiated with 25 kGy. The water was collected June 24,1996, bottled July 11 & 12, and irradiated July 16, 1996.

The participating laboratories were each sent two bottles of MOOS-1 and requested to perform duplicate analyses on each of the bottles. The participants were also sent a data file in which to record their results and analytical procedures. NRC also analyzed the sample using two different analytical methods for each measurand.

All concentrations are expressed in micromoles per liter atomic.

In 1997 MOOS-1 served as a supplementary test sample for the European QUASIMEME Laboratory Performance Studies (Quality Assurance of Information for Marine Environmental Monitoring in Europe).

2. RESULTS

The prepared samples were mailed to the thirty-six participating laboratories listed in Appendix A in November 1999 with the deadline for receipt of results set for March 13, 2000. Thirty sets of results were received. Sequential numbers were assigned to each responding laboratory upon receipt of its data. Laboratory numbers 31 and 32 were assigned to NRC. The submitted data are listed in Appendix B.

A copy of the tabulated raw data was sent to each participant that had submitted results by the deadline in order to verify that no errors had been made in the transposition of numbers. The data are listed in the tables as received with respect to significant figures.

This sample material was intended to be a certified reference material for silicate, phosphate, nitrite and nitrite+nitrate. It became apparent, however, from NRC results and evidence from several laboratories that there was a question of interbottle inhomogeneity. This make the bias evaluation difficult and a less stringent judgement using a statistical tolerance interval was adopted. A tolerance interval is determined by multiplying the calculated standard deviation by an expansion factor (between 2 and 3) found in appropriate statistical tables. For this intercomparison, a tolerance interval was constructed such that it will cover at least 95% of the population of bottles with a probability of 95%. This statement does not guarantee that the tolerance interval will include all of the bottles; rather 95% of the time the tolerance interval will include at least 95% of the bottles. The assigned mean and tolerance intervals are listed in Table 1 and were determined using NRC results and the data of several selected laboratories.

Table 1

Analyte	Assigned Mean and Tolerance Interval, µM	Target Standard Deviation
Phosphate	1.67 ± 0.19	0.095
Silicate	25.4 ± 2.5	1.25
Nitrite	2.76 ± 0.58	0.29
Nitrite and Nitrate	23.2 ± 2.4	1.2

It was decided to treat the submitted results from each bottle independently rather than calculate the overall mean of the four replicates. The replicate data are plotted on the graphs with duplicate results from one bottle indicated by the character "x" and the second bottle by "□". One laboratory that required more than 50 mL per determination blended a number of bottles together. The solid horizontal line represents the assigned mean and the shaded area represents the tolerance limits. A histogram for the data is shown in the second graph for each analyte.

The z-scoring system is an accepted method used in intercomparison exercises to assess bias. This is accomplished by comparing the bias estimate for each analyte with a target value for standard deviation. The bias estimate is calculated from the difference between the laboratory mean (x_i) and the accepted (or assigned) mean ($\bar{x}$). The z-score is calculated by dividing the bias estimate by the target value for standard deviation (σ_{target}).

$$z = \frac{(x_i - \bar{x})}{\sigma_{target}}$$

As a consequence of inhomogeneity, σ_{target} was defined as one-half the assigned tolerance interval and treat the submitted results from each bottle separately. For example the results from laboratory 1 are displayed as "1a", the mean from one bottle and "1b", the mean from the second bottle. The z scores are graphed and listed in the tables for each analyte. Conventionally, z scores >3 are unsatisfactory but for this exercise are considered questionable as it is possible the z score for some laboratories is greater than 2 when in fact the lab received an "outlier" bottle.

$\|z\| \leq 2$	satisfactory
$\|z\| \geq 2$	questionable

It is hoped that breaking the results into bottles rather than evaluating the overall mean submitted by a laboratory will be more equitable to a lab that might have received an outlier bottle.

P- scores are used to evaluate the precision of a laboratory. If a number of replicates are performed the p score is defined as the standard deviation of the replicates divided by the target standard deviation. (Note: the target standard deviation may or may not be the same as target standard deviation used to calculate z-scores.) In this exercise two replicates are used to calculate the p score and the equation is modified to become

$$p = \left(\frac{|r_i - r_{i+1}| / \text{assigned value}}{\sigma_p} \right)$$

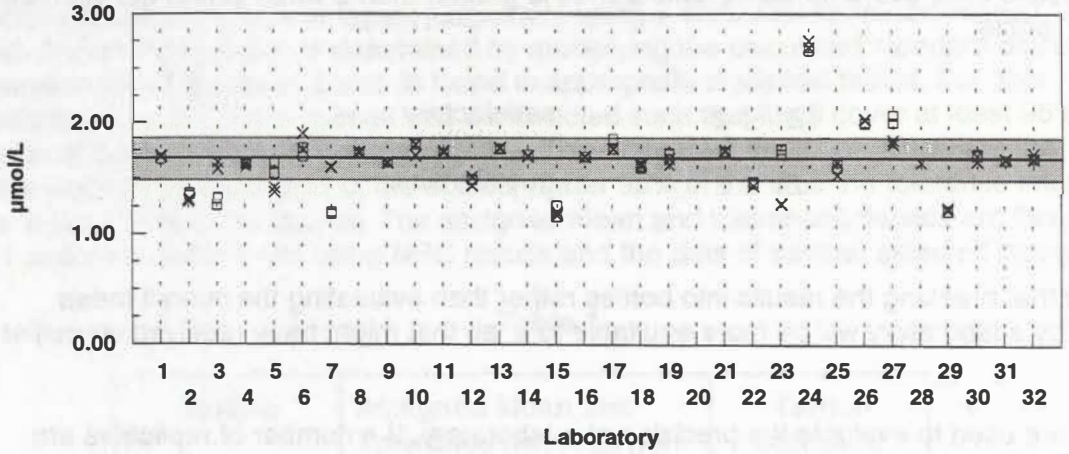
where r_i and r_{i+1} are replicate measurements, the assigned value is listed in Table 1 and σ_p is set at 5%. Calculated p scores are presented in the tables.

Appendix C summarizes the analytical methods as reported by the participating laboratories.

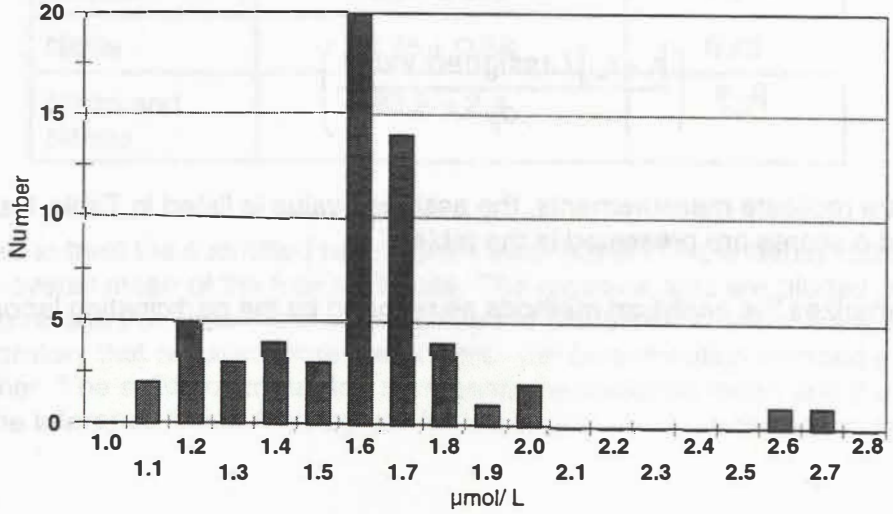
Summary of Phosphate Results

Assigned Tolerance Interval	1.67 ± 0.19 µmol/ L
Target SD	0.095 µmol/ L
Number of Results	31
Consensus mean and SD of all results	1.66 ± 0.28 µmol/ L

Graph of Phosphate Results



Histogram of Phosphate Data



Phosphate z scores

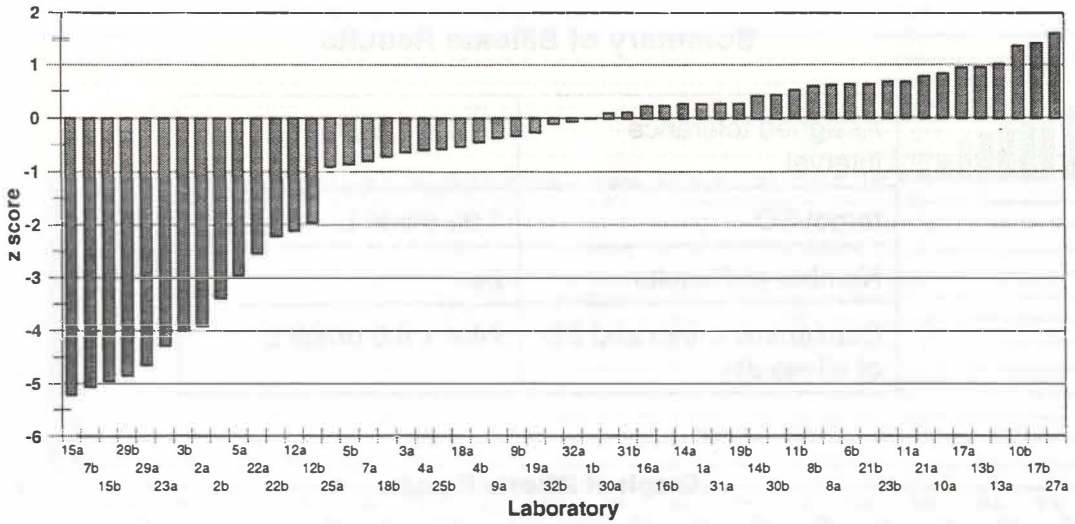


Table of z and p scores for Phosphate

Lab	z	p
1a	0.3	0.1
1b	0.0	0.0
2a	-3.9	0.2
2b	-3.4	0.2
3a	-0.6	0.7
3b	-4.0	0.7
4a	-0.6	0.4
4b	-0.4	0.1
5a	-2.9	0.4
5b	-0.9	1.1
6a	1.6	1.8
6b	0.6	1.0
7a	-0.8	0.1
7b	-5.1	0.2
8a	0.6	0.1
8b	0.6	0.0
9a	-0.4	0.1
9b	-0.3	0.0
10a	0.8	1.2
10b	1.4	0.0

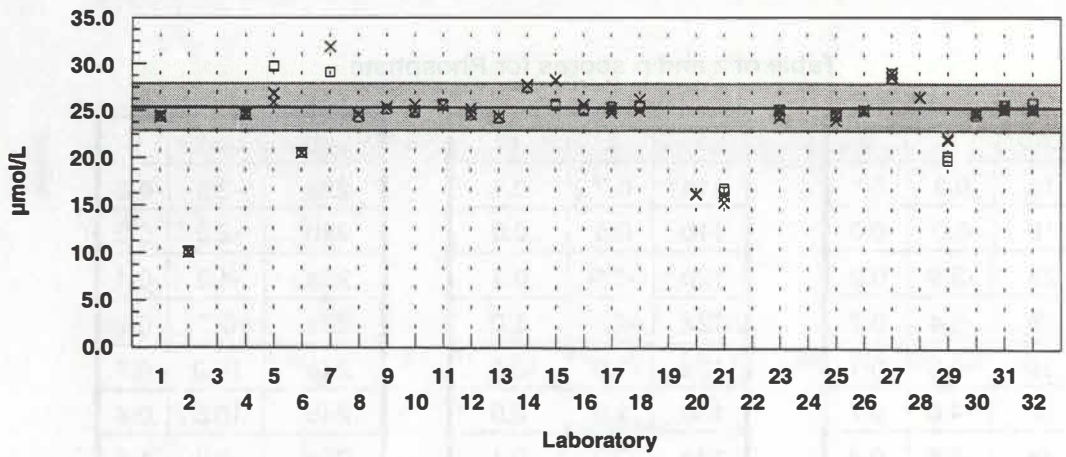
Lab	z	p
11a	0.7	0.1
11b	0.5	0.0
12b	-1.9	0.1
12a	-2.1	1.0
13a	1.0	0.1
13b	1.0	0.0
14a	0.3	0.1
14b	0.4	0.0
15a	-5.2	0.4
15b	-4.9	1.2
16a	0.2	0.0
16b	0.2	0.0
17a	0.9	0.0
17b	1.4	1.1
18a	-0.5	0.2
18b	-0.7	0.2
19a	-0.3	0.6
19b	0.3	0.6
21a	0.8	0.1
21b	0.6	0.2

Lab	z	p
22a	-2.5	0.0
22b	-2.2	0.2
23a	-4.3	0.1
23b	0.7	0.6
24a	10.9	0.7
24b	10.5	0.4
25a	-0.9	1.3
25b	-0.6	0.6
26a	3.7	0.1
26b	3.4	0.1
27a	1.6	0.4
27b	3.8	0.8
29a	-4.6	0.2
29b	-4.8	0.0
30a	0.4	0.1
30b	0.1	0.6
31a	0.3	0.6
31b	-0.1	0.1
32a	0.1	0.0
32b	-0.1	0.0

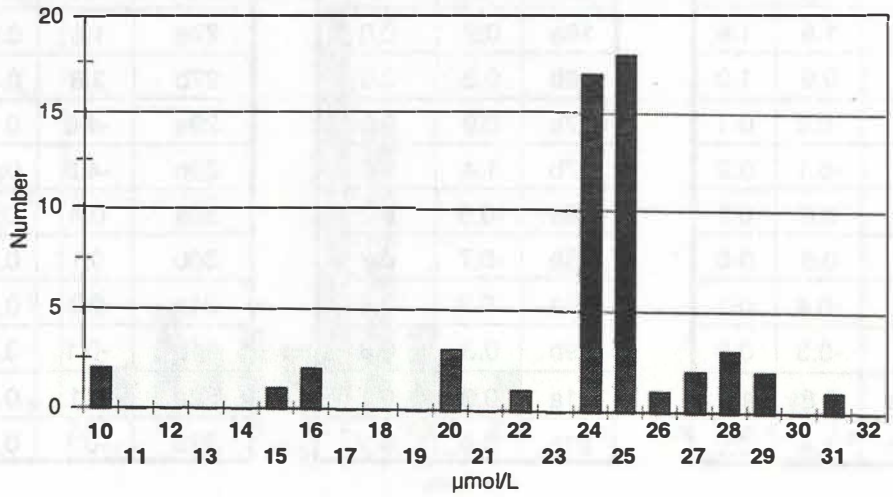
Summary of Silicate Results

Assigned tolerance interval	25.4 ± 2.5 µmol/ L
target SD	1.25 µmol/ L
Number of Results	28
Consensus mean and SD of all results	24.4 ± 4.0 µmol/ L

Graph of Silicate Results



Histogram of Silicate Data



Silicate z scores

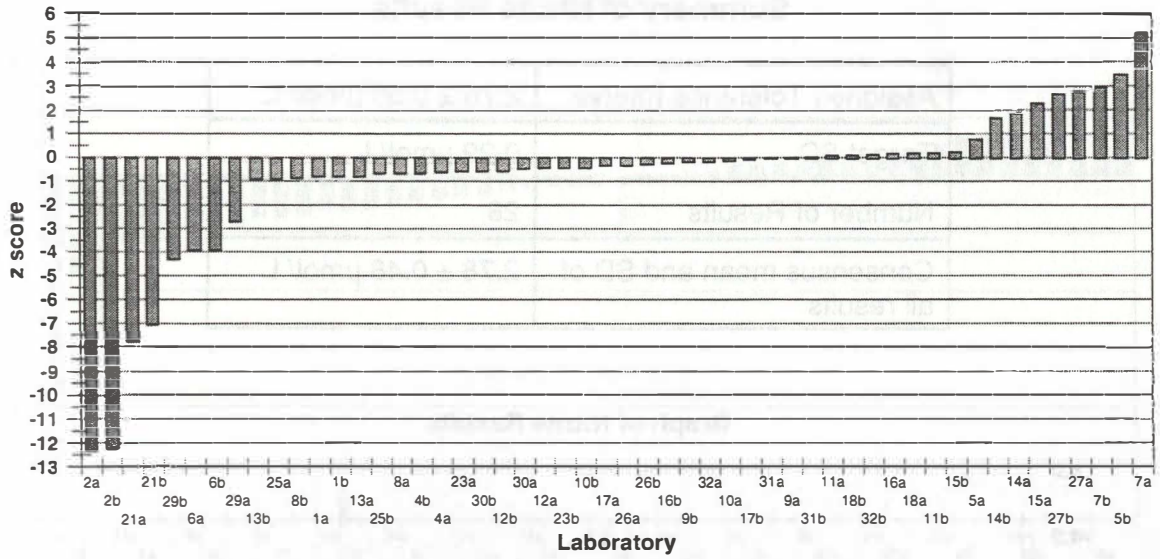


Table of z and p scores for Silicate

Lab	z	p
1a	-0.8	0.2
1b	-0.8	0.0
2a	-12.3	0.0
2b	-12.2	0.1
4a	-0.6	0.1
4b	-0.6	0.1
5a	0.8	0.7
5b	3.5	0.1
6a	-3.9	0.0
6b	-3.9	0.1
7a	5.2	0.0
7b	3.0	0.0
8a	-0.6	0.1
8b	-0.8	0.0
9a	0.0	0.0
9b	-0.2	0.0
10a	-0.1	0.5
10b	-0.4	0.2

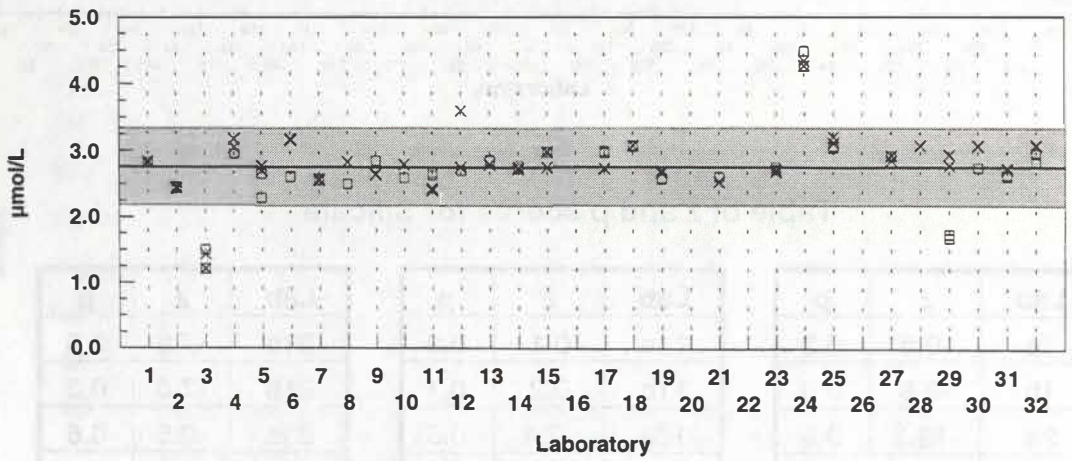
Lab	z	p
11a	0.1	0.0
11b	0.2	0.1
12a	-0.4	0.5
12b	-0.5	0.2
13a	-0.8	0.0
13b	-0.9	0.0
14a	1.8	0.1
14b	1.7	0.0
15a	2.3	0.1
15b	0.2	0.2
16a	0.2	0.1
16b	-0.2	0.2
17a	-0.3	0.4
17b	-0.1	0.2
18a	0.2	0.9
18b	0.1	0.1
20a	-7.4	0.1

Lab	z	p
21a	-7.8	0.6
21b	-7.0	0.2
23a	-0.6	0.6
23b	-0.4	0.5
25a	-0.9	0.4
25b	-0.6	0.0
26a	-0.3	0.1
26b	-0.2	0.0
27a	2.8	0.4
27b	2.7	0.4
29a	-2.7	0.2
29b	-4.3	0.3
30a	-0.4	0.1
30b	-0.6	0.3
31a	0.0	0.3
31b	0.0	0.4
32a	-0.2	0.2
32b	0.1	0.6

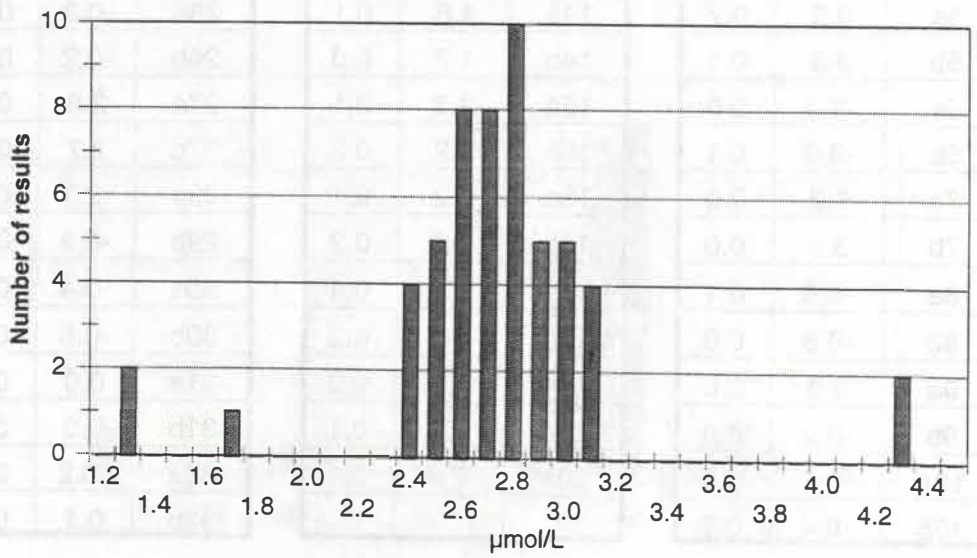
Summary of Nitrite Results

Assigned Tolerance Interval	2.76 ± 0.58 µmol/ L
Target SD	0.29 µmol/ L
Number of Results	28
Consensus mean and SD of all results	2.78 ± 0.48 µmol/ L

Graph of Nitrite Results



Histogram of Nitrite Data



Nitrite z scores

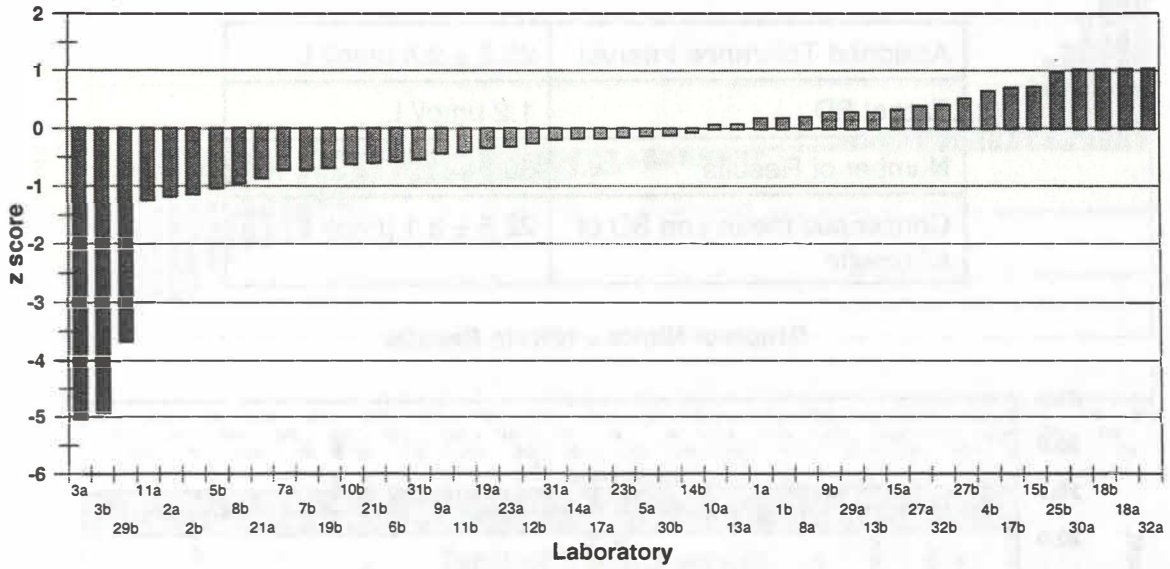


Table of z and p scores

Lab	z	p
1a	0.2	0.1
1b	0.2	0.0
2a	-1.1	0.1
2b	-1.1	0.2
3a	-5.0	1.6
3b	-4.8	2.1
4a	1.3	1.0
4b	0.7	0.0
5a	0.0	0.5
5b	-1.0	2.6
6a	1.4	0.1
6b	-0.5	0.1
7a	-0.7	0.1
7b	-0.7	0.1
8a	0.3	0.0
8b	-0.9	0.0
9a	-0.4	0.1
9b	0.3	0.0

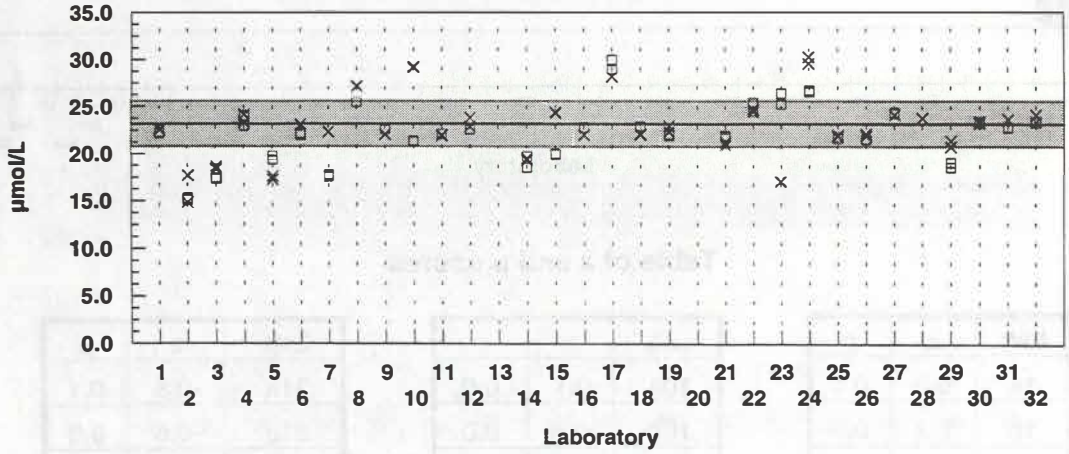
Lab	z	p
10a	0.1	0.0
10b	-0.6	0.0
11a	-1.2	0.2
11b	-0.3	0.3
12a	1.4	6.1
12b	-0.2	0.0
13a	0.1	0.0
13b	0.3	0.1
14a	-0.1	0.1
14b	0.0	0.3
15a	0.4	1.7
15b	0.8	0.0
17a	-0.1	0.1
17b	0.8	0.2
18a	1.1	0.0
18b	1.1	0.0
19a	-0.3	0.2
19b	-0.6	0.1

Lab	z	p
21a	-0.8	0.1
21b	-0.5	0.0
23a	-0.2	0.1
23b	-0.1	0.3
24a	5.4	0.5
24b	5.6	1.6
25a	1.4	0.7
25b	1.0	0.1
27a	0.5	0.5
27b	0.6	0.0
29a	0.3	1.2
29b	-3.6	0.4
30a	1.1	0.1
30b	0.0	0.0
31a	-0.1	0.1
31b	-0.4	0.1
32a	1.1	0.1
32b	0.5	0.9

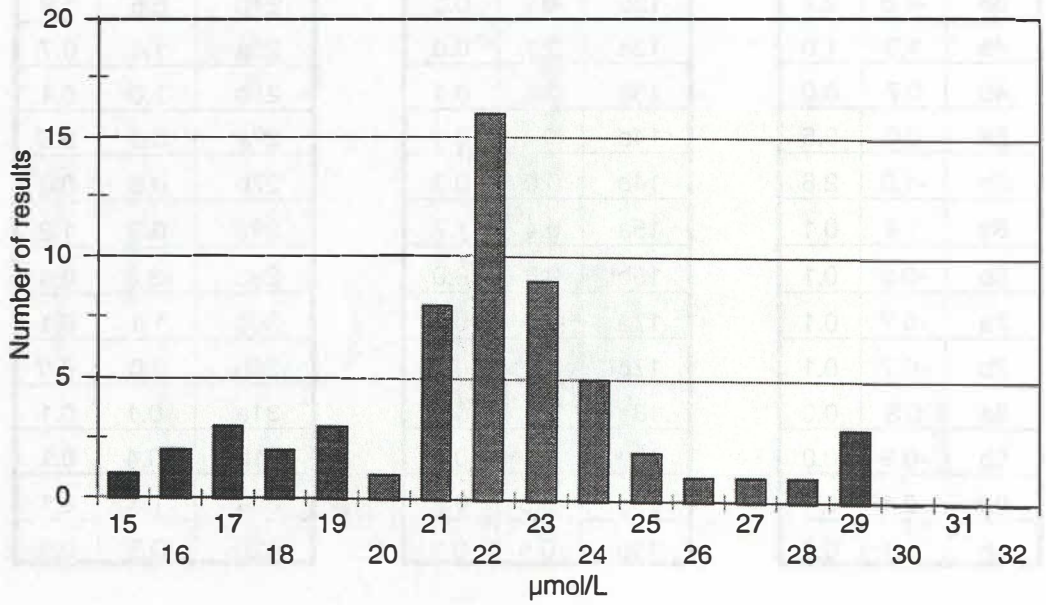
Summary of Nitrite + Nitrate Results

Assigned Tolerance Interval	23.2 ± 2.4 µmol/ L
Target SD	1.2 µmol/ L
Number of Results	30
Consensus mean and SD of all results	22.5 ± 3.1 µmol/ L

Graph of Nitrite + Nitrate Results



Histogram of Nitrite + Nitrate Data



Nitrite + Nitrate z scores

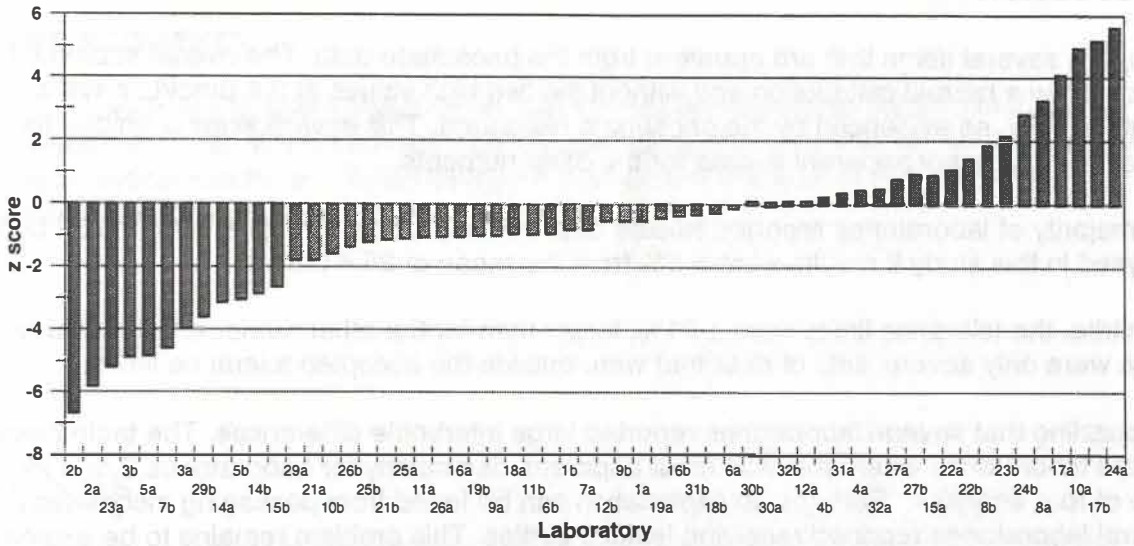


Table of Z and p scores

Lab	z	p
1a	-0.5	0.1
1b	-0.8	0.1
2a	-5.8	2.4
2b	-6.7	0.2
3a	-4.0	0.3
3b	-4.9	0.1
4a	0.5	0.7
4b	0.2	0.9
5a	-4.8	0.3
5b	-3.0	0.4
6a	-0.1	0.0
6b	-0.9	0.2
7a	-0.8	0.0
7b	-4.6	0.2
8a	3.3	0.1
8b	1.9	0.0
9a	-1.0	0.0
9b	-0.5	0.0
10a	5.0	0.1
10b	-1.5	0.1

Lab	z	p
11a	-1.0	0.1
11b	-0.9	0.0
12a	0.1	0.8
12b	-0.5	0.0
14a	-3.1	0.2
14b	-2.8	0.0
15a	1.0	0.1
15b	-2.6	0.1
16a	-1.0	0.0
16b	-0.3	0.0
17a	4.1	0.0
17b	5.2	0.9
18a	-0.9	0.1
18b	-0.2	0.1
19a	-0.5	0.6
19b	-1.0	0.2
21a	-1.7	0.3
21b	-1.1	0.2
22a	1.1	0.3
22b	1.5	0.8

Lab	z	p
23a	-5.2	0.0
23b	2.2	1.0
24a	5.6	0.6
24b	2.8	0.2
25a	-1.1	0.1
25b	-1.2	0.2
26a	-1.0	0.2
26b	-1.3	0.0
27a	0.8	0.0
27b	0.9	0.1
29a	-1.8	0.5
29b	-3.6	0.4
30a	0.1	0.2
30b	0.1	0.2
31a	0.4	0.0
31b	-0.3	0.1
32a	0.5	0.6
32b	0.1	0.0

2. DISCUSSION

There are several items that are apparent from the phosphate data. The overall submitted data do not follow a normal distribution and without the two high values at 2.6 µmol/L, have a negative skew as evidenced by the phosphate histogram. This severe skew is unique for phosphate and is not apparent in data for the other nutrients.

The majority of laboratories reported silicate data in the 24-25 µmol/L range. Of the 52 bottles analysed in this study 8 results were > 5% from the mean of 25.4 µmol/L.

For nitrite, the tolerance limits were ± 21%, larger than for the other nutrients in this study. There were only several sets of data that were outside the accepted tolerance limits.

It is puzzling that several laboratories reported large interbottle differences. The table below lists the laboratories where this was most apparent, particularly for Laboratories 7 and 29 for three of four analytes. Perhaps an explanation can be found from packaging inconsistencies. Several laboratories reported receiving leaking bottles. This problem remains to be examined in more detail.

	Laboratories Reporting Interbottle Differences
phosphate	3, 7, 23, 27
silicate	5, 7, 15, 29
nitrite	6, 8, 9, 10, 11, 17, 21, 29
nitrite + nitrate	5, 7, 8, 9, 10, 15, 19, 23, 24, 29

These interbottle differences were not evident from a recent homogeneity study performed at NRC on forty-eight bottles of MOOS-1. The range of phosphate data was ±0.08 µmol/L from the mean with a RSD of 2%. Similar results were obtained for the other nutrients with nitrite having the largest RSD at 5%.

Concluding Remarks

The results of this intercomparison may, in several respects, have been compromised by the question of homogeneity of the test sample. The target standard deviation for measuring p-scores is too broad and does not reflect the measurement precision that can be attained. An upcoming workshop will present an opportunity to discuss changes to these criteria. The overall results, however, are quite encouraging and it is hoped that these questions can be resolved and that future exercises can benefit from these studies and build on these results.

ACKNOWLEDGMENTS

The authors would like to thank Peter Strain and Pierre Clement of the Bedford Institute of Oceanography for help with the collection and preparation of the sample. Lu Yang of NRC for providing analytical results and Ralph Sturgeon and James McLaren of NRC for helpful discussions.

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Appendix A

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Appendix B

Phosphate Results

Lab	Replicate 1	Replicate 2	Replicate 3	Replicate 4
1	1.69	1.7	1.67	1.67
2	1.29	1.31	1.36	1.34
3	1.58	1.64	1.32	1.26
4	1.60	1.63	1.62	1.63
5	1.37	1.41	1.64	1.54
6	1.9	1.75	1.77	1.69
7	1.6	1.59	1.2	1.18
8	1.723	1.735	1.726	1.728
9	1.63	1.64	1.64	1.64
10	1.7	1.8	1.8	1.8
11	1.73	1.74	1.72	1.72
12	1.51	1.43	1.48	1.49
13	1.772	1.763	1.759	1.762
14	1.7	1.69	1.71	1.71
15	1.16	1.19	1.15	1.25
16	1.689	1.692	1.692	1.692
17	1.76	1.76	1.76	1.85
18	1.61	1.63	1.61	1.59
19	1.62	1.67	1.67	1.72
20	-	-	-	-
21	1.75	1.74	1.72	1.74
22	1.43	1.43	1.45	1.47
23	1.26	1.27	1.71	1.76
24	2.68	2.74	2.65	2.68
25	1.53	1.64	1.64	1.59
26	2.03	2.02	2.00	1.99
27	1.806	1.839	2.000	2.065
28	1.64			
29	1.22	1.24	1.21	1.21
30	1.654	1.703	1.717	1.706
31	1.72	1.67	1.68	1.68
32	1.66	1.67	1.66	1.66

Silicate Results

Lab	Replicate 1	Replicate 2	Replicate 3	Replicate 4
1	24.5	24.3	24.4	24.4
2	10.05	10.03	10.2	10.06
3	-	-	-	-
4	24.58	24.70	24.67	24.58
5	26.8	25.9	29.8	29.7
6	20.49	20.53	20.51	20.6
7	31.9	31.9	29.1	29.1
8	24.67	24.55	24.34	24.36
9	25.41	25.42	25.2	25.19
10	25.6	25.0	25.0	24.8
11	25.5	25.5	25.6	25.7
12	24.6	25.2	24.6	24.9
13	24.458	24.4	24.259	24.276
14	27.76	27.66	27.48	27.48
15	28.3	28.2	25.8	25.6
16	25.66	25.56	25.27	25.07
17	25.28	24.81	25.47	25.19
18	25.03	26.21	25.44	25.62
19	-	-	-	-
20	16.12	16.22	-	-
21	15.3	16.0	16.5	16.8
22	-	-	-	-
23	24.3	25.1	24.6	25.2
24	-	-	-	-
25	24.52	24.02	24.59	24.59
26	25.0	25.1	25.1	25.1
27	29.080	28.600	28.500	29.000
28	26.49			
29	21.94	22.14	20.24	19.84
30	24.77	24.92	24.55	24.87
31	25.16	25.59	25.20	25.66
32	25.3	25.1	25.2	25.9

Nitrite Results

Lab	Replicate 1	Replicate 2	Replicate 3	Replicate 4
1	2.82	2.84	2.83	2.83
2	2.45	2.43	2.47	2.44
3	1.43	1.21	1.21	1.5
4	3.19	3.06	2.97	2.97
5	2.71	2.78	2.66	2.30
6	3.16	3.18	2.62	2.61
7	2.56	2.58	2.56	2.58
8	2.835	2.839	2.509	2.509
9	2.66	2.65	2.86	2.86
10	2.8	2.8	2.6	2.6
11	2.44	2.41	2.68	2.64
12	3.6	2.76	2.71	2.71
13	2.803	2.799	2.857	2.865
14	2.73	2.74	2.74	2.78
15	2.99	2.76	2.99	2.99
16	-	-	-	-
17	2.73	2.74	3	2.97
18	3.08	3.08	3.08	3.07
19	2.67	2.7	2.58	2.59
20	-	-	-	-
21	2.53	2.54	2.61	2.61
22	-	-	-	-
23	2.68	2.7	2.76	2.72
24	4.35	4.28	4.28	4.5
25	3.21	3.12	3.07	3.05
26	-	-	-	-
27	2.857	2.929	2.929	2.929
28	3.08			
29	2.78	2.94	1.75	1.69
30	3.07	3.08	2.75	2.75
31	2.72	2.74	2.62	2.64
32	3.09	3.07	2.96	2.84

Nitrite + Nitrate Results

Lab	Replicate 1	Replicate 2	Replicate 3	Replicate 4
1	22.57	22.66	22.29	22.23
2	14.84	17.65	15.31	15.11
3	18.6	18.3	17.3	17.4
4	23.35	24.14	24.02	22.97
5	17.56	17.22	19.81	19.36
6	23.09	23.06	22.22	22
7	22.3	22.3	17.8	17.6
8	27.11	27.18	25.46	25.45
9	22.04	22.05	22.6	22.61
10	29.2	29.1	21.4	21.3
11	22.0	21.9	22.1	22.1
12	23.8	22.9	22.6	22.6
13	-	-	-	-
14	19.36	19.58	19.84	19.8
15	24.3	24.4	20	20.1
16	21.99	22.03	22.81	22.81
17	28.11	28.14	29.97	28.91
18	22.04	22.14	22.95	22.86
19	22.98	22.34	22.13	21.91
20	-	-	-	-
21	21.27	20.94	21.97	21.73
22	24.4	24.7	25.4	24.5
23	16.98	16.97	25.27	26.39
24	30.3	29.6	26.5	26.7
25	21.97	21.88	21.88	21.65
26	21.9	22.1	21.6	21.6
27	24.143	24.143	24.286	24.357
28	23.79			
29	21.4	20.78	18.64	19.15
30	23.43	23.25	23.42	23.23
31	23.67	23.66	22.79	22.86
32	23.41	24.16	23.35	23.35

Appendix C

Methods used for Determining Phosphate

Lab	Method for Determining Phosphate
1	Phosphate is analyzed using a modification of the Bernhardt and Wilhelms technique. An acidic solution of ammonium molybdate is added to the sample to produce phosphomolybdic acid, then reduced to phosphomolybdous acid (a blue compound) following the addition of dihydrazine sulfate. The reaction product is heated to ~55C to enhance color development. Skalar SanPlus Autoanalyzer Primary standard for phosphate (KH_2PO_4) is obtained from Johnson Matthey Chemical Co. and the supplier reports purity of 99.999%. Bernhardt, H., and Wilhelms, A., "The continuous determination of low level iron, soluble phosphate and total phosphate with the AutoAnalyzer," Technicon Symposia, I, pp.385-389 (1967).
2	Orthophosphate reacts with molybdenum (VI) and antimony (III) in an acid medium to form a phosphoantimonylmolybdenum complex which is reduced by ascorbic acid to a heteropolyblue with a maximum absorbance of 880 nm.
3	EPA method # 365.1, Lachat Quickchem AE method # 10-115-01-1-A
4	Reaction of orthophosphate with molybdenum (VI) and antimony (III) to form a phosphoantimonylmolybdenum complex which is reduced by ascorbic acid to a blue color with a maximum absorbance of 880 nm.
5	For orthophosphate an antimonyphosphomolybdate complex was formed. EnviroFlow 3500 nutrient analyzer (Perstorp Analytical, Wilsonville, OR.)
6	Parsons and Strickland
7	The nutrients were determined using a 3 channel technicon II autoanalyzer system. The methods used were: PO4 - reaction with ammonium molybdate in acidic medium and reduction with hydrazine.(measured at 880 nm)
8	Concentrations of nitrite, nitrate, phosphate and silicic acid were determined using an AlpKem flow solution Auto-Analyzer. The water used for the preparation of standards and wash solution was filtered seawater obtained from the surface of the Gulf Stream. Phosphate in the samples was determined by reacting with molybdenum (VI) and antimony (III) in an acidic medium to form an antimonyphosphomolybdate complex. This complex was subsequently reduced with ascorbic acid to form a heteropoly blue and the absorbance was measured at 710 nm (Zhang et al., 1999).

Lab	Method for Determining Phosphate
9	Analytical methods used for silica, phosphate, nitrite, and nitrate follow the recommendations of Gordon et al. (1993) for the WOCE WHP project. We currently employ an Alpkem RFA II segmented-flow nutrient analyzer equipped with colorimeters and interference filters for wavelength selection. Phosphate is determined by creating the phosphomolybdate heteropoly acid in much the same way as with the silica method. However, its reducing agent is dihydrazine sulfate, after which its transmittance is also measured. A heating bath is required to maximize the color yield. Gordon, L.I., J.C. Jennings, Jr., A.A. Ross, and J.M. Krest. 1993. A Suggested Protocol For Continuous Flow Automated Analysis of Seawater Nutrients. In: WOCE Operation Manual. WHP Office Report 90-1, WOCE Report 77 No. 68/91. 1-52.
10	The analysis of Orthophosphate was performed by combining molybdenum (VI) and antimony (III) in an acid medium to form an antimony-phosphomolybdate complex. This complex is subsequently reduced with ascorbic acid to form a blue color and the absorbance was measured at 660 nm on an autoanalyzer.
11	Nutrients were measured using a Technicon Continuous Flow Analyzer and colorimetric methods. Standards: A stock standard solution was gravimetrically prepared for each nutrient from dry chemicals dissolved in double-Milli-Q (DMQ) water. Phosphate and nitrite concentrations were based on dry-weight measurements prepared from reagents that were 99.5% in purity. Working solutions of silicate and nitrate standards were calibrated with respect to Sagami standard solutions. Four working standards with a concentration range bracketing the anticipated sample concentration range were prepared and analyzed (in duplicate) at the beginning and end of the analyses. The ranges used were: phosphate (0, 1,2,3 umol/l). Working standards were prepared with artifical seawater (3.2% NaCl) which was also used as the baseline/wash solution. All reagents were prepared with (DMQ) water. All glass and plasticware were acid cleaned in 1NHCl, rinsed with Milli-Q water two times and rinsed with DMQ water once. Colorimetric techniques used were: Phosphate: reduced by ascorbic acid to a phospho-molybdenum blue complex. Peaks were recorded digitally and peak heights were measured. A standard curve was calculated using a second order polynomial equation and used to determine sample concentrations.
12	All analyses performed via Flow Injection Analysis using a Lachat Instruments 'QuikChem 8000' analyzer. Phosphate: Lachat Instruments 'QuikChem Method 31-115-01-3-A' Orthophosphate ion reacts with ammonium molybdate and antimony potassium tartrate under acidic conditions to form a yellow complex. This complex is reduced with ascorbic acid to form a blue complex which absorbs light at 880 nm.

Lab	Method for Determining Phosphate
13	1. Four plastic bottles with sea water samples for the NOAA intercomparison exercise were received by December 24, 1999. These samples were kept at 6C until February 11, 2000 and at 16-18C for another week, when the analysis was proceeded. 2. Autoanalyzer II (BRAN+LUEBBE) has been used to perform analyses. Standard methods proposed by BRAN+LUEBBE for AutoAnalyzer applications have been used to analyze the suggested samples for phosphate (G-175-96, MT18), nitrite (G-173-96, MT18) and silicate (G-177-96, MT19).
14	PO4: AAII, hydrazine sulfate and ammonium molybdate
15	PHOSPHATE: EPA Method 365.1 using an Astoria Pacific 300 series Autoanalyzer.
16	Phosphate: Soluble orthophosphate is determined by Technicon Industrial Methods No. 155-71 modified by Brynjolfson (1973). A single reagent stream combining an acidified solution of ammonium molybdate (and a small amount of antimony) and ascorbic acid forms a phosphomolybdenum blue complex. Armstrong, F.A. J., C.R. Stearns, and J.D.H. Strickland. 1967. The measurement of upwelling and subsequent biological processes by means of the Technicon AutoAnalyzer and associated equipment. Deep-Sea Res. 14(3): 381-389.
17	Phosphate and Silica were analyzed following standard colorimetric methods. -Phosphate: Reagents- Ammonium Molybdate, L-Ascorbic acid, H_2SO_4, Potassium antimonyl tartrate Read at 880 nm Concentration= Sample Absorbance/Slope of linear regression
18	Phosphate, silicate and nitrite analyses were performed as per Strickland and Parsons, 1972 Fish. Res. Bull. 10 cm cell for nitrite and phosphate As we used large sample volumes we pooled 14 MOOS-1 sample bottles and then separated these into two replicates.
19	Ortho-phosphate reacts with Ammonium Molybdate and Antimony Potassium Tartrate in acid medium to form an antimony-phospho-molybdate complex. This comples is subsequently reduced with ascorbic acid to form a blue color. The color is proportional to the phosphorus concentration. The developed color is measured at 660nm. EPA method 365.1
20	Not determined

Lab	Method for Determining Phosphate
21	We use a Lachat QC8000 Autoanalyzer made by Zellweger Analytics. All methods described here were written by the Applications Group at Lachat Instruments, Milwaukee, WI: phosphate by Amy Huberty and David Diamond, silica by R. McKnight, nitrite by Scott Schroeder, and total oxidisable nitrogen by David Diamond. The phosphate method is based on reactions specific for the orthophosphate ion (PO43-) and covers a range from 0.03 to 2.00 µM. The PO43- reacts with ammonium molybdate and antimony potassium tartrate under acidic conditions to form a complex. This complex is reduced with ascorbic acid to form a blue complex which absorbs light at 880 nm. The ascorbic acid and molybdate reagents are merged on the chemistry manifold and the reagent stream is then merged with the carrier stream. The sample zone appears at the detector less than 10 sec after injection. The absorbance is proportional to the concentration of PO43- in the sample.
22	Orthophosphate and TOxN (nitrite+nitrate) were determined spectrophotometrically using a Segmented Flow Analyser.
23	The analyses were performed manually on a HP 8453 spectrophotometer with 1 cm cell with about 1.5 ml solution capacity. A 0.7 m NaCl solution was used to prepare the calibration curves and the blanks. The 50 ml sample just allowed us to run the duplicate sample. Since no previous knowledge of the concentration level, the calibration curve for total oxidised nitrogen was outside the linear calibration range thus a polynomial fit was used to derive the concentration. The reagents were prepared according to the recipe of Dr. J-Z Zhang in AOML/NOAA. The recipe was optimazed for flow injection analysis and has been demonstrated to be successful in manual analysis.
24	not available
25	Ascorbic Acid/Molybdate Colorimetric Method
26	Technicon method # 155-71W, January 1973. The automated procedure for the determination of ortho phosphate in seawater depends on the formation of a phosphomolybdenum blue complex which is read colorimetrically at 880 nm.
27	Phosphate- EPA 365.1
28	Technicon Industrial Method #155-71W. The sample is first diluted and then the mixed reagent is added to the stream. This mixture is heated to 36 C after which the absorbance of the phosphomolybdate complex is measured at 820nm in a 50 x 1.5mm flow cell.
29	EPA 365.1 using a four-channel Alpkem RFA-300 (Rapid Flow Analyzer) Nutrient Analyzer
30	Not available

Lab	Method for Determining Phosphate
31	ortho-phosphates - were analyzed by the Technicon AutoAnalyzer II system. Reagents - ascorbic acid, antimony potassium tartrate, ammonium molybdate, sulphuric acid Filter - 880 nm, Standards were made up with 3.7% NaCl; Sample wash was 3.7% NaCl.
32	Phosphate and silica were determined on-line by Ion Exclusion Chromatography - Inductively Coupled Plasma Mass Spectrometry. Eluent used was 50 mM HCl. Standard addition method was used for the calibration. The column was Dionex IonPac ICE-AS1 ion exclusion column.

Methods used for Determining Silicate

Lab	Method for Determining Dissolved Silicate
1	Silicate is analyzed using the technique of Armstrong, 1967. An acidic solution of ammonium molybdate is added to a seawater sample to produce silicomolybdic acid which is then reduced to silicomolybdous acid (a blue compound) following the addition of stannous chloride. Tartaric acid is added to impede PO4 color development. Skalar SanPlus Autoanalyzer. Na_2SiF_6, the silicate primary standard, is obtained from Johnson Matthey Chemical Company and/or Fisher Scientific and is reported by the suppliers to be >98% pure. Armstrong, F.A.J., Stearns, C.A., and Strickland, J.D.H., "The measurement of upwelling and subsequent biological processes by means of the Technicon Autoanalyzer and associated equipment," Deep-Sea Research, 14, pp.381-389 (1967).
2	Silicate Method B-molybdosilicate acid is formed by the reaction of silicate with molybdate at a pH of 1 to 1.8. The B-molybdosilicic acid is reduced by tin (II) to form molybdenum blue with a maximum absorbance of 820 nm.
3	Not determined
4	reaction with molybdate reagent to forma β-molybdo-silicic acid. The complex is reduced by ascorbic acid to form molybdenum blue. The absorbance is measured at 660nm.

Lab	Method for Determining Dissolved Silicate
5	Samples were thawed at room temperature before analysis in an EnviroFlow 3500 nutrient analyzer (Perstorp Analytical, Wilsonville, OR.). They were shaken vigorously and poured into two 4 ml vials and the remaining sample was refrozen. The vials were placed in an autosampler and orthophosphate, silicate and nitrate+nitrite were determined simultaneously. For orthophosphate an antimonyphosphomolybdate complex was formed, for silicate silicomolybdic acid was formed, and the nitrate was reduced to nitrite in a reductor column containing copper coated cadmium. The nitrite was then reacted with an aromatic amine. The nitrite concentration alone in the samples was determined at a later date.
6	Phosphate, Silica, and Nitrite procedures as described by Parsons and Strickland.
7	Type Method Here. The nutrients were determined using a 3 channel technicon II autoanalyzer system. The methods used were: Dissolved silica - Reaction with ammonium molybdate in acidic medium to form silicomolybdic acdi which is reduced to the molybdenum blue with stannous chloride (measured at 630 nm). We had to dilute the samples to fit in our concentration working range (up to 15 uM) The samples were run against low nutrient sea water as baseline and wash.
8	Concentrations of nitrite, nitrate, phosphate and silicic acid were determined using an AlpKem flow solution Auto-Analyzer. The water used for the preparation of standards and wash solution was filtered seawater obtained from the surface of the Gulf Stream. Standardizations were performed prior to analysis with working solutions prepared from purity standards. Silicic Acid: Silicic acid in the sample was reacted with molybdate in a acidic solution to form B-molybdosilicic acid, which was then reduced by ascorbic acid to form the molybdenum blue. Absorbances were measured at 660 nm (Zhang et al., 1997b). References: Zhang, J-Z., and G. A. Berberian, (1997b) Determination of dissolved silicate in estuarine and coastal waters by gas segmented continuous flow colorimetric analysis. EPA's manual " Methods for the determination of Chemical Substances in Marine and Estuarine Environmental Matrices - 2nd Edition". EPA/600/R-97/072, September 1997. Zhang, J-Z., C. Fischer and P. B. Ortner, (1999) Optimization of performance and minimization of silicate interference in continuous flow phosphate analysis. Talanta, 49:293-304.

Lab	Method for Determining Dissolved Silicate
9	Analytical methods used for silica, phosphate, nitrite, and nitrate follow the recommendations of Gordon et al. (1993) for the WOCE WHP project. We currently employ an Alpkem RFA II segmented-flow nutrient analyzer equipped with colorimeters and interference filters for wavelength selection. Silica is determined by forming the heteropoly acid of dissolved orthosilicic acid and ammonium molybdate, then reducing it with stannous chloride, and measuring its optical transmittance. Gordon, L.I., J.C. Jennings, Jr., A.A. Ross, and J.M. Krest. 1993. A Suggested Protocol For Continuous Flow Automated Analysis of Seawater Nutrients. In: WOCE Operation Manual. WHP Office Report 90-1, WOCE Report 77 No. 68/91. 1-52.
10	Dissolved Silica was combined with a molybdate reagent in an acid media to form B-molybdosilicic acid. The complex was reduced by ascorbic acid to form molybdeum blue. The absorbance was measured at 660 nm on an autoanalyzer.
11	Nutrients were measured using a Technicon Continuous Flow Analyzer and colorimetric methods. Standards: A stock standard solution was gravimetrically prepared for each nutrient from dry chemicals dissolved in double-Milli-Q (DMQ) water. Silicate: Technicon Method - reduced by ascorbic acid to a silico-molybdenum blue complex. Working standards were prepared with artifical seawater (3.2% NaCl) which was also used as the baseline/wash solution. Peaks were recorded digitally and peak heights were measured. A standard curve was calculated using a second order polynomial equation and used to determine sample concentrations.
12	Dissolved Silica: Lachat Instruments 'QuikChem Method 31-114-27-1-B' Soluble silica species react with molybdate at 37 degrees centigrade and pH of 1.2 to form a yellow silicamolybdate complex. This complex is subsequently reduced with stannous chloride to form a heteropoly blue complex which has an absorbance maximum at 820 nm. All analyses performed via Flow Injection Analysis using a Lachat Instruments 'QuikChem 8000' analyzer.
13	Autoanalyzer II (BRAN+LUEBBE) has been used to perform analyses. Standard methods proposed by BRAN+LUEBBE for AutoAnalyzer applications have been used to analyze the samples for silicate (G-177-96, MT19).
14	Silica: AAII, ammonium moybdate and stannous chloride
15	EPA Method 370.1 using an Astoria Pacific 300 series Autoanalyzer.

Lab	Method for Determining Dissolved Silicate
16	Dissolved silica is determined by Technicon Industrial Methods No. 186-72W, essentially that of Armstrong et al. (1967). The procedure is based on the reduction of silicomolybdate in acidic solution to molybdenum blue by ascorbic acid. Oxalic acid is introduced to the sample stream before the addition of ascorbic acid to eliminate interference from phosphates. Armstrong, F.A. J., C.R. Stearns, and J.D.H. Strickland. 1967. The measurement of upwelling and subsequent biological processes by means of the Technicon AutoAnalyzer and associated equipment. Deep-Sea Res. 14(3): 381-389.
17	Reagents-Ammonium Molybdate, Metol Sulfite, Oxalic acid, H_2SO_4 Read at 810 nm
18	silicate analyses were performed as per Strickland and Parsons, 1972 Fish. Res. Bull.
19	not determined
20	All reagents and standards were prepared prior to analysis with Milli-Q water. Samples were processed and analyzed following the methodology of Strickland and Parsons (1972). Each duplicate 30 ml sample was added to 1 ml acid molybdate mixed reagent containing ammonium heptamolybdate tetrahydrate and sulfuric acid. The sample was agitated and left to stand for 10 minutes. Next 1 ml of oxalic acid was added to the sample followed by the immediate addition of 0.5 ml of ascorbic acid. The samples were agitated and left to stand for one hour. Samples were then read in a 10 cm cell at 810 nm on an LKB Ultraspec 4050.
21	We use a Lachat QC8000 Autoanalyzer made by Zellweger Analytics. All methods described here were written by the Applications Group at Lachat Instruments, Milwaukee, WI The dissolved silica method analyzes silica as silicon dioxide (SiO2) and covers a range from 0.2 to 2.00 mg SiO2/L (values converted to µM by formula: mg SiO2/L x 1000/MW; MW=60). Soluble silica species react with molybdate under acidic conditions to form a yellow silicamolybdate complex. This complex is subsequently reduced with 1-amino-2-naphthol-4-sulfonic acid (ANSA) and bisulfite to form a heteropoly blue complex which has an absorbance at 820 nm.
22	Not determined
23	The analyses were performed manually on a HP 8453 spectrophotometer with 1 cm cell with about 1.5 ml solution capacity. A 0.7 m NaCl solution was used to prepare the calibration curves and the blanks. The Si and P standards were ordered from Ultra Science. The reagents were prepared according to the recipe of Dr. J-Z Zhang in AOML/NOAA.

Lab	Method for Determining Dissolved Silicate
24	not determined
25	Molybdate blue- Colorimetric Method Oxalic Acid is introduced to eliminate interferences from phosphate
26	Technicon method # 186-72W, March 1973 (with correction for salinity). This automated procedure for the determination of soluble silicates is based on the reduction of a silicomolybdate in acidic solution to "molybdenum blue" by ascorbic acid. Oxalic acid is introduced to the sample stream before the addition of ascorbic acid to eliminate interference from phosphates.
27	Dissolved Silica- EPA 370.1
28	Based on Technicon Industrial Method #186-72W/Tentative. Silica reacts with acid molybdate solutions to form silico-molybdic acids which can be reduced to form an intense heteropoly blue complex which is measured at 820nm in a 50 x 1.5mm flowcell. We use ascorbic acid as the reductant and do not require a heating bath to speed the reaction.
29	Dissolved silica is also determined on the Alpkem RFA using method USGS I2700-85 (a slight modification of EPA method 370.1) using the protocol outlined by Perstorp Analytical Environmental. This modification involves the addition acidified (with sulfuric acid) ammonium molybdate and oxalic acid to the water sample, subsequent reduction with ascorbic acid, and the spectrophotometric measurement of the resulting color development at 660 nm.
30	Not available
31	silicates were analyzed by the Technicon AutoAnalyzer II system. Reagents - ammonium molybdate, oxalic acid, ascorbic acid Filter - 660 nm, Standards were made up by standard additions; Sample wash was 3.7% NaCl.
32	Phosphate and silica were determined on-line by Ion Exclusion Chromatography - Inductively Coupled Plasma Mass Spectrometry. Eluent used was 50 mM HCl. Standard addition method was used for the calibration. The column was Dionex IonPac ICE-AS1 ion exclusion column.

Methods used for Determining Nitrite

Lab	Method for Determining Nitrite
1	A modification of the Armstrong (1967) procedure is used for the analysis of nitrate and nitrite. For the nitrate analysis, the seawater sample is passed through a cadmium reduction column where nitrate is quantitatively reduced to nitrite. Sulfanilamide is introduced to the sample stream followed by N-(1-naphthyl)ethylenediamine dihydrochloride which couples to form a red azo dye. The same technique is employed for nitrite analysis, except the cadmium column is not present. Skalar SanPlus Autoanalyzer. Primary standards for nitrate (KNO_3) and nitrite ($NaNO_2$) are obtained from Johnson Matthey Chemical Co. and the supplier reports purities of 99.999% and 97%, respectively. Armstrong, F.A.J., Steams, C.A., and Strickland, J.D.H., "The measurement of upwelling and subsequent biological processes by means of the Technicon Autoanalyzer and associated equipment," Deep-Sea Research, 14, pp.381-389 (1967).
2	Nitrite is determined as an azo dye with a maximum absorbance of 540 nm following its diazotization with sulfanilamide and subsequent coupling with N-1-Naphthyl-ethylenediame.
3	Nitrate/Nitrite: EPA method # 353.2, Lachat Quickchem AE method # 10-107-04-1-C Nitrite: Same as above without Cd reduction column.
4	Nitrite is reacted with sulfanilamide and NED to form a red azo dye. The absorbance measured at 540nm.
5	Samples were thawed at room temperature before analysis in an EnviroFlow 3500 nutrient analyzer (Perstorp Analytical, Wilsonville, OR.). They were shaken vigorously and poured into two 4 ml vials and the remaining sample was refrozen. The nitrite was then reacted with an aromatic amine. The nitrite concentration alone in the samples was determined at a later date.
6	Nitrite procedures as described by Parsons and Strickland. TOxN performed by chemiluminescence NO-NO2-NOX analyzer.
7	The nutrients were determined using a 3 channel technicon II autoanalyzer system. TOxN - nitrite present in the solution reacts with sulphanilamide to form a diazo compound which then couples with N-1 naphtyl ethylenediamine dihydrochloride to form a reddish purple azo dye (measured at 520 nm) The samples were run against low nutrient sea water as baseline and wash.
8	Concentrations of nitrite, nitrate were determined using an AlpKem flow solution Auto-Analyzer. The water used for the preparation of standards and wash solution was filtered seawater obtained from the surface of the Gulf Stream.

Lab	Method for Determining Nitrite
9	Analytical methods used for silica, phosphate, nitrite, and nitrate follow the recommendations of Gordon et al. (1993) for the WOCE WHP project. We currently employ an Alpkem RFA II segmented-flow nutrient analyzer equipped with colorimeters and interference filters for wavelength selection. Nitrite is determined essentially by the Bendschneider and Robinson (1952) technique in which nitrite is reacted with sulfanilamide (SAN) to form a diazotized derivative that is then reacted with a substituted ethylenediamine compound (NED) to form a rose-pink azo dye which is measured colorimetrically. Gordon, L.I., J.C. Jennings, Jr., A.A. Ross, and J.M. Krest. 1993. A Suggested Protocol For Continuous Flow Automated Analysis of Seawater Nutrients. In: WOCE Operation Manual. WHP Office Report 90-1, WOCE Report 77 No. 68/91. 1-52.
10	Nitrite was determined by combining the sample with sulfanilamide and subsequent coupling with N-1-naphthylene-diamine dihydrochloride, forming an azo dye which was measured at 540 nm on an autoanalyzer.
11	Nitrite: Technicon Method - reacted with sulfanilamide and NED to form a red azo dye. Technicon Continuous Flow Analyzer and colorimetric methods. Working standards were prepared with artifical seawater (3.2% NaCl) which was also used as the baseline/wash solution. Peaks were recorded digitally and peak heights were measured. A standard curve was calculated using a second order polynomial equation and used to determine sample concentrations.
12	Nitrite: Lachat Instruments 'QuikChem Method 31-107-04-1-A' Nitrite is determined by diazotization with sulfanilamide under acidic conditions to form a diazonium ion. The resulting diazonium ion is coupled with N-(1-napthyl)ethylenediamine dihydrochloride. The resulting pink dye absorbs at 520 nm. All analyses performed via Flow Injection Analysis using a Lachat Instruments 'QuikChem 8000' analyzer.
13	Autoanalyzer II (BRAN+LUEBBE) has been used to perform analyses. Standard methods proposed by BRAN+LUEBBE for AutoAnalyzer applications have been used to analyze the suggested samples for nitrite (G-173-96, MT18)
14	AAII, sulfanilamide and N-(1-naphthyl)-ethylenediamine
15	EPA Method 353.2 using a Lachat QuikChem Autoanalyzer.
16	not determined
17	Nitrite and TOxN were analyzed on an Alpkem RFA/2 Rapid Flow Analyzer Reagents- Sulfanilamide, N-1-Naphthylethylenediamine Dihydrochloride Filter= 540nm, Standards were made up with DI water, Sample wash was 35% NaCl.
18	Nitrite analyses were performed as per Strickland and Parsons, 1972 Fish. Res. Bull. 10 cm cell for nitrite

Lab	Method for Determining Nitrite
19	Nitrite (NO_2): the nitrite is determined as an azo dye at 540 nm following its diazotization by sulfanilamide and subsequent coupling with N-1-naphtylethylenediamine dihydrochloride., EPA Method 353.2.
20	Not determined
21	We use a Lachat QC8000 Autoanalyzer made by Zellweger Analytics. All methods described here were written by the Applications Group at Lachat Instruments, Milwaukee, WI: The nitrite method is based on reactions specific for the nitrite ion (NO_2^-) and covers a range from 0.02 to 5.0 µM. Nitrite is determined by diazotization with sulfanilamide under acidic conditions to form a diazonium ion. The diazonium ion is coupled with N-(1-naphthyl)ethylenediamine dihydrochloride. The resulting pink dye absorbs at 520 nm.
22	Not determined
23	The analyses were performed manually on a HP 8453 spectrophotometer with 1 cm cell with about 1.5 ml solution capacity. A 0.7 m NaCl solution was used to prepare the calibration curves and the blanks. The reagents were prepared according to the recipe of Dr. J-Z Zhang in AOML/NOAA. The recipe was optimized for flow injection analysis and has been demonstrated to be successful in manual analysis. Nitrite and nitrate stock solutions were prepared with analytical grade $NaNO_2$ and KNO_3 respectively. Prior use the tubes were first washed with Micro solution followed by soaking in 4N for one week and then rinsed with MilliQ water.
24	not available
25	Sulfanilamide - N-1-napthylenediamine dihydrochloride- Colorimetric method
26	Technicon method # 158-71W/B, August 1979 (with correction for salinity). This automated procedure for the determination of nitrate and nitrite utilizes the procedure whereby nitrate is reduced to nitrite by a copper-cadmium reductor column. The nitrite ion then reacts with sulfanilamide under acidic conditions to form a diazo compound. This compound then couples with N-1-napthylethylenediamine dihydrochloride to form a reddish-purple azo dye.
27	Nitrite- EPA 353.2
28	Technicon Industrial Method #158-71 W/Tentative (1972). The sample is mixed with alkaline ammonium chloride solution. This reacts with acidified sulfanilamide and N-(1-napthyl) ethylenediamine dihydrochloride to form a red azo compound which is measured at 540nm in a 50 x 1.5mm flow cell.
29	EPA 353.2 using a four-channel Alpkem RFA-300 (Rapid Flow Analyzer) Nutrient Analyzer
30	not available

Lab	Method for Determining Nitrite
31	Nitrite and TOxN - were analyzed by the Technicon AutoAnalyzer II system. Reagents - ammonium chloride, sulfanilamide, N-1-naphthylethylenediamine dihydrochloride, phosphoric acid. Filter - 550 nm, Standards were made up with 3.7% NaCl; Sample wash was 3.7% NaCl.
32	Nitrite was analyzed by Ion Chromatography using a Dionex AS10 column with AG10 guard column. Eluant was 80 mM NaCl, Flow was 1 ml/min Detection was by UV @ 225 nm with no suppression

Methods used for Determining Nitrite + Nitrate

Lab	Method for Determining Nitrite + Nitrate
1	A modification of the Armstrong (1967) procedure is used for the analysis of nitrate and nitrite. For the nitrate analysis, the seawater sample is passed through a cadmium reduction column where nitrate is quantitatively reduced to nitrite. Sulfanilamide is introduced to the sample stream followed by N-(1-naphthyl)ethylenediamine dihydrochloride which couples to form a red azo dye. The same technique is employed for nitrite analysis, except the cadmium column is not present.Skalar SanPlus Autoanalyzer.
2	Nitrite is determined as an azo dye with a maximum absorbance of 540 nm following its diazotization with sulfanilamide and subsequent coupling with N-1-Naphthyl-ethylenediame. Nitrate+Nitrite Method Nitrate is reduced quantitatively to nitrite by copperized cadmium in the form of an open tubular cadmium reactor. The nitrite formed plus any originally present in the sample is determined by the same method as nitrite above.
3	Nitrate/Nitrite: EPA method # 353.2, Lachat Quickchem AE method # 10-107-04-1-C
4	Nitrate is converted to nitrite by Cd reduction. Nitrite is reacted with sulfanilamide and NED to form a red azo dye. The absorbance measured at 540nm.
5	Samples were thawed at room temperature before analysis in an EnviroFlow 3500 nutrient analyzer (Perstorp Analytical, Wilsonville, OR.). They were shaken vigorously and poured into two 4 ml vials and the remaining sample was refrozen. The vials were placed in an autosampler and orthophosphate, silicate and nitrate+nitrite were determined simultaneously. The nitrate was reduced to nitrite in a reductor column containing copper coated cadmium. The nitrite was then reacted with an aromatic amine.

Lab	Method for Determining Nitrite + Nitrate
6	Nitrite procedures as described by Parsons and Strickland. TOxN performed by chemiluminescence NO-NO2-NOX analyzer.
7	The nutrients were determined using a 3 channel technicon II autoanalyzer system. TOxN - Nitrate is reduced to nitrite by a copperized Cd column and then the nitrite that was present in the solution and the reduced nitrate react with sulphanilamide to form a diazo compound which then couples with N-1 naphtyl ethylenediamine dihydrochloride to form a reddish purple azo dye (measured at 520 nm) The samples were run against low nutrient sea water as baseline and wash.
8	Concentrations of nitrite and nitrate were determined using an AlpKem flow solution Auto-Analyzer. The water used for the preparation of standards and wash solution was filtered seawater obtained from the surface of the Gulf Stream.
9	Analytical methods used for silica, phosphate, nitrite, and nitrate follow the recommendations of Gordon et al. (1993) for the WOCE WHP project. We currently employ an Alpkem RFA II segmented-flow nutrient analyzer equipped with colorimeters and interference filters for wavelength selection.Nitrite is determined essentially by the Bendschneider and Robinson (1952) technique in which nitrite is reacted with sulfanilamide (SAN) to form a diazotized derivative that is then reacted with a substituted ethylenediamine compound (NED) to form a rose-pink azo dye which is measured colorimetrically. Nitrate is determined by difference after a separate aliquot of a sample is passed through a Cd reduction column to covert its nitrate to nitrite, followed by the measurement of the "augmented" nitrite concentration using the same method as in the nitrite analysis. Gordon, L.I., J.C. Jennings, Jr., A.A. Ross, and J.M. Krest. 1993. A Suggested Protocol For Continuous Flow Automated Analysis of Seawater Nutrients. In: WOCE Operation Manual. WHP Office Report 90-1, WOCE Report 77 No. 68/91. 1-52.
10	Nitrite was determined by combining the sample with sulfanilamide and subsequent coupling with N-1-naphthylene-diamine dihydrochloride, forming an azo dye which was measured at 540 nm on an autoanalyzer.
11	Nitrite: Technicon Method - reacted with sulfanilamide and NED to form a red azo dye. Technicon Continuous Flow Analyzer and colorimetric methods. Working standards were prepared with artifical seawater (3.2% NaCl) which was also used as the baseline/wash solution. Peaks were recorded digitally and peak heights were measured. A standard curve was calculated using a second order polynomial equation and used to determine sample concentrations.

Lab	Method for Determining Nitrite + Nitrate
12	Nitrite: Lachat Instruments 'QuikChem Method 31-107-04-1-A' Nitrite is determined by diazotization with sulfanilamide under acidic conditions to form a diazonium ion. The resulting diazonium ion is coupled with N-(1-napthyl)ethylenediamine dihydrochloride. The resulting pink dye absorbs at 520 nm. All analyses performed via Flow Injection Analysis using a Lachat Instruments 'QuikChem 8000' analyzer.
13	Not determined
14	AAII, sulfanilamide and N-(1-naphthyl)-ethylenediamine
15	EPA Method 353.2 using a Lachat QuikChem Autoanalyzer.
16	not determined
17	Nitrite and TOxN were analyzed on an Alpkem RFA/2 Rapid Flow Analyzer Reagents- Sulfanilamide, N-1-Naphthylethylenediamine Dihydrochloride Filter= 540nm, Standards were made up with DI water, Sample wash was 35% NaCl.
18	Nitrite analyses were performed as per Strickland and Parsons, 1972 Fish. Res. Bull.
19	Nitrate/Nitrite (NOx-N): Nitrate is quantitatively reduced to nitrite by metal Cadmium in the column. Then the nitrite formed by reduction of nitrate plus nitrite originally present is determined as an azo dye at 540 nm following its diazotization by sulfanilamide and subsequent coupling with N-1-naphtylethylenediamine dihydrochloride. Using EPA Method 353.2
20	Not determined
21	We use a Lachat QC8000 Autoanalyzer made by Zellweger Analytics. All methods described here were written by the Applications Group at Lachat Instruments, Milwaukee, WI: TOxN method (nitrate and nitrite together) is based on reactions specific for the nitrate ion (NO3-) and covers a range from 0.03 to 5.0 µM. Nitrate is quantitatively reduced to nitrite by passage of the sample through a copperized cadmium column. The nitrite (reduced nitrate plus the original nitrite) is then determined by diazotization with sulfanilamide under acidic conditions to form a diazonium ion. The resulting diazonium ion is coupled with N-(1-naphthyl)ethylenediamine dihydrochloride. The resulting pink dye absorbs at 520 nm.
22	TOxN (nitrite+nitrate) were determined spectrophotometrically using a Segmented Flow Analyser.

Lab	Method for Determining Nitrite + Nitrate
23	The analyses were performed manually on a HP 8453 spectrophotometer with 1 cm cell with about 1.5 ml solution capacity. A 0.7 m NaCl solution was used to prepare the calibration curves and the blanks. Since no previous knowledge of the concentration level, the calibration curve for TOxN was outside the linear calibration range thus a polynomial fit was used to derive the concentration. The reagents were prepared according to the recipe of Dr. J-Z Zhang in AOML/NOAA. The recipe was optimazed for flow injection analysis and has been demonstrated to be successful in manual analysis. Cd/Cu reducing column was made in a thin Tygon tubing. Solution was pumped through the column with a peri pump. The total space in the tubing is less than 1 ml which allows manual operation with small volume samples. The recovery test with 20 uM of NO3- indicates a recovery of 93%. Thus the final result was corrected. Nitrite and nitrate stock solutions were prepared with analytical grade NaNO2 and KNO3 respectively.
24	not available
25	Sulfanilamide - N-1-napthylenediamine dihydrochloride- Colorimetric method Cadmium Reduction Colorimetric Method
26	Technicon method # 158-71W/B, August 1979 (with correction for salinity).
27	TOxN- EPA 353.2
28	not available
29	EPA 353.2 using a four-channel Alpkem RFA-300 (Rapid Flow Analyzer) Nutrient Analyzer
30	not available
31	Nitrite and TOxN - were analyzed by the Technicon AutoAnalyzer II system. Reagents - ammonium chloride, sulfanilamide, N-1-naphthylethylenediamine dihydrochloride, phosphoric acid. Filter - 550 nm, Standards were made up with 3.7% NaCl; Sample wash was 3.7% NaCl.
32	Nitrate was analyzed by Ion Chromatography using a Dionex AS10 column with an AG10 guard column. Eluant was 200 mM HCl, Flow was 1 ml/min Detection was by UV @ 225 nm with no suppression