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PROGRESS REPORT

Ecological and Radioecological Studies in the Columbia River Estuary and Adjacent Pacific Ocean

Robert L. Holton, William G. Pearcy
and
Norman H. Cutshall
Investigators

Edited by
Karla J. McMechan

Submitted to
U.S. Energy Research and Development
Administration

Contract E(45-1)-2227, Modification 7 to
Task Agreement 12
RLO-2227-T12-63

Reference 76-2

1 April 1975 through 31 March 1976

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Principal Investigator

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Karla J. McMechan

School of Oceanography
Oregon State University
Corvallis, Oregon

Reference 76-2
April 1976

John V. Byrne
Dean

ACKNOWLEDGMENTS

A major expense in oceanographic research is "time at sea." Operations of the research fleet, the R/V YAQUINA, R/V CAYUSE, R/V PAIUTE, and R/V SACAJAWEA, were funded by several agencies, with the bulk coming from the National Science Foundation and Office of Naval Research. Certain special cruises of radiochemical or radioecological import were funded by the U.S. Energy Research and Development Administration, as was much of the equipment for radioanalysis and stable element analysis. We gratefully acknowledge the role of these agencies in support of the research reported in the following pages.

We also wish to express our thanks to the numerous students and staff who contributed to the preparation of this progress report. The text was typed by Mrs. Gerri Riley.

NOTICE

The progress report that follows includes research results ranging from unproved ideas to scientific papers published during the tenure of this contract. The end of the contract year finds several facets of our work in various stages of preparation, therefore the reader is cautioned that all except the published papers are subject to revision.

PRINCIPAL INVESTIGATOR

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CO-INVESTIGATORS

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ABSTRACTS OF THESES COMPLETED

RESEARCH IN PROGRESS

RADIOECOLOGICAL STUDIES IN THE COLUMBIA RIVER AND ESTUARY

INTRODUCTION

This report represents the progress in "Ecological and Radioecological Studies in the Columbia River Estuary and Adjacent Pacific Ocean" for the period 1 April 1975 through 31 March 1976. This research has been supported with funds from the Division of Biomedical and Environmental Research, U.S. Energy Research and Development Administration through Contract AT(45-1)-2227, Task Agreement 12.

Our current research activities are dealt with in a descriptive fashion in the section on Research in Progress. The Research Completed section of the report includes manuscripts which have been submitted for publication during the reporting period, as well as reprints of papers published during this same period.

Dr. Norman H. Cutshall has resigned his position at Oregon State University, effective 31 January 1976. However he will continue to serve as the major professor for his students who are well along in their thesis research programs. He has accepted a position at Oak Ridge National Laboratory, Oak Ridge, Tennessee and is currently at that address. We are searching for a replacement for Dr. Cutshall and expect to have someone hired and on the job sometime during the summer of 1976.

SCIENTIFIC BACKGROUND

The oceans and, particularly, the nearshore zone and the continental shelf are destined to be the site of increasingly intense development of energy generation facilities. Nuclear facilities seem particularly adaptable to the advantages of coastal sites and the Outer Continental Shelf (OCS) oil leasing program is rapidly becoming a reality. It is imperative that this imminent development proceed with maximum possible understanding of how the nearshore and shelf ecosystems function.

It has been apparent for some years now that location of electrical generating plants, especially nuclear plants, may be increasingly oriented toward the coastal ocean. Not only are many power users, i.e. industrial and population centers, already sited in the coastal zone, but the access to marine transportation and to abundant coolant water are of compelling importance. Offshore sites on artificial islands or floating platforms, and even submerged sites, have gained prime consideration during recent years. By moving large generating facilities offshore, dose interaction with people is lessened with relatively little diminution of the qualities that make the coastal zone attractive for such facilities.

Great force drives this move to the coastal zone and offshore. Increase of energy demand has brought 1960's forecasts of impending shortages to fact even in the mid-1970's. Already it appears that both the rate of increase of energy generating (converting) capability and the rate of shift toward the coastal zone are increasing. The oil crisis must cause a greater share of our energy to come from alternative sources such as nuclear fission and, eventually, fusion. The scale and scope of man's activities in the nearshore ocean during the remainder of the twentieth century will be substantially greater than it has been to date.

The OCS oil leasing program presents another spectrum of potential problems for the coastal marine environment that must be examined in the next very few years to enable us to make the reasonable decisions necessary for the adequate protection of this environment.

It is important that this development be undertaken with the greatest possible insight into the operation of coastal ecosystems. Not only is there a potential for alteration of natural community structures and loss of economic species, but the very success of installed facilities may also be at stake. Our program probes into the mechanics of coastal marine ecosystems. It is directed research because we seek to make it as responsive as possible to forseeable needs for knowledge arising as coastal zone development progresses. It is basic research because we are seeking to learn what fundamental processes control the coastal ecosystem. Our efforts are focused upon learning rates and routes of material transport and transfer. We have seen that "artificially" introduced materials are often useful in studying these processes. We have found radionuclides from the Columbia River and from atmospheric fallout to be especially useful as tracers because of their characteristic time constants and because they can be measured at extremely low levels.

Our studies have also extended as far inland in the Columbia River as Hanford because operations there have provided a major source of radioisotope tracers. In other rivers our efforts terminate where oceanic salt water is lacking. We emphasize estuarine and continental shelf problems in our studies. We have a limited effort underway in laboratory studies. Our program has always included a component of research oriented toward the open ocean, far from land and we hope to maintain such a component. Large-scale oceanic systems are thought to operate relatively slowly compared to coastal systems. Thus, although their remoteness from most of our daily lives and their vast scale lessen concern for acute contamination problems, the relative permanence of problems once they are created in the open ocean requires careful attention to "blue water" processes. The scope of this program allows us to conduct only limited open ocean research. However, we seek to conduct limited sampling programs designed to answer questions we think are significant to the environmental quality of the open ocean system.

RELATED PROGRAMS

A recent study has been conducted under the sponsorship of the Port of Astoria to assess the standing crop of benthic infauna in a proposed fill site adjacent to Pier 3. The entire estuary has also been surveyed during this study to allow us to compare and contrast the productivity of the proposed fill site with that of the estuary in general. This study is the first attempt to quantitatively assess the benthic secondary production for the Columbia River estuary.

Dr. Holton is currently working with the Pacific Northwest River Basins Commission to design a Columbia River estuary study plan. The plan will designate research required in coming years to provide information needed for making sound management decisions. The product of this effort should be useful to all agencies conducting research in the estuary.

In addition Drs. Holton and Cutshall are completing a study with Drs. L.F. Small and L.I. Gordon of the impact of open ocean disposal of dredged materials. This program, sponsored by the U.S. Army Corps of Engineers, is concentrated on disposal off the Columbia River Mouth and is, therefore, geographically akin to previous ERDA research. Its focus is more narrow, however, in terms of goals and extent.

Dr. Holton has conducted a study of thermal effects on certain coastal invertebrate animals. These studies have been conducted on the larvae of the Dungeness crab, *Cancer magister* and on mixed species of copepods. These studies are currently continuing, but will be completed during the summer of 1976. This work was supported by three Oregon electrical utility companies.

FACILITIES

Counting Laboratory

Several hardware additions have been made to our gamma-ray spectrometry system, as reported in the last progress report. Among these is a lithium-drifted germanium detector (85 cm^3) purchased from Princeton Gamma Tech in May 1975. This new detector and associated electronic hardware provides us with additional gamma-ray counting capability. The more essential specifications of this detector are listed below.

Efficiency:	16.9% @ 25 cm source-to-detector distance (relative to a 3 x 3 inch NaI(Tl) detector)
Resolution:	0.769 KeV FWHM @ 0.122 MeV
	1.75 KeV FWHM @ 1.33 MeV
	3.26 KeV FW.1M @ 1.33 MeV
	(FW=full width, HM=at half maximum, .1M=at 0.1 maximum.)

Peak/Compton Ratio: 46.2 @ 1.33 MeV

An RK-11 Disc Pack unit [Digital Equipment Corporation (DEC)] has been added to the system. Many advantages are offered by this addition, most notably the shorter time involved with acquiring and loading into memory the software programs used in gamma-ray data reduction and the added capability available during editing of new or existing software programs. The disc pack is capable of storing 1.2 million words.

Other additions of hardware include twelve thousand words of core memory and an expansion box to extend the effective main frame capacity of the central processor unit (BK-11, DEC). Also a DEC Writer-II was added, which replaced the teletype (ASR 33) and increased the typed output rate by a factor of three.

Future plans include the purchase of a high-speed punched paper tape unit to be used for obtaining hard-copy software program documentation that can be read back into the computer for reprogramming should the disc unit information become destroyed.

Low-level environmental radioactive samples are currently being analyzed by use of the two lithium-drifted germanium detectors, each surrounded by 10 cm of lead and each coupled to a 4096 multichannel analyzer. Sample counting time typically ranges between 300 to 1000 minutes duration. Upon completion of a count, background radioactivity is subtracted electronically from the sample counts after peak integration utilizing the PDP-11 computer system. Data output is by a line printer and contains peak-energy identification and radionuclide concentrations determined from calibrated known sources.

Two sample container configurations or geometries are presently being utilized. Large sample sizes are placed into Marinelli beakers (volume: 550 ml); small sample sizes are placed into 4 ounce jars (volume: 125 ml).

Table 1 lists lower limits of detection for the specified radionuclides. The lower level of detection is defined as the minimum amount of radionuclide which must be present in order to report a value greater than zero approximately 95% of the time. These values depend upon the background present in the counting system, as well as the counting time, and were calculated by the method described by Altshuler and Pasternack (1963) and Pasternack and Harley (1971).

To compare our system with a known standard we conducted the following evaluation. A liquid solution of known amounts of added radionuclides was obtained from the Oregon State Board of Health through the courtesy of Mr. George L. Toombs, Chief, Environmental Radiation Surveillance Program. This solution was originally distributed by the National Environmental Research Center, Las Vegas, Nevada. The known amounts of gamma-emitting radionuclides added to the standard were compared with the amounts detected by our system. The results were decay corrected to 18 April 1975, the reference date of the standard, and are given in Table 2.

Table 1. Lower levels of detection (LLD)* for various radionuclides in the given geometric volume per 1000-minute counting time. MB=marinelli beaker, 550 ml; 4-oz=4 ounce jar, 125 ml.

Radionuclide	KeV	Background Counts	LLD (pCi)	Minimum Detectable Sample Concentration (pCi/ml)
^{137}Cs	661.6	MB = 129	3.5	0.0064
		4-oz = 120	3.6	0.029
^{54}Mn	834.8	MB = 65	2.6	0.0047
		4-oz = 78	3.0	0.024
^{65}Zn	1115.5	MB = 59	5.9	0.011
		4-oz = 54	5.9	0.047
^{60}Co	1332.5	MB = 41	2.9	0.0053
		4-oz = 37	2.9	0.023
^{40}K	1460.8	MB = 238	67	0.12
		4-oz = 239	71	0.57

*LLD = $Q(K_a + K_b)\sqrt{(2)(BG)}$, where Q = appropriate conversion factor for converting counting rate to picocuries; K_a, K_b both @ 95% confidence level 1 tail-*t* test = 1.645; BG = Background counts per 1000 minutes. For a 1000-minute (16.7 h) counting time the concentration of the above radionuclides must be present in the given geometric volume in order to produce a value greater than zero approximately 95% of the time.

Table 2. Amounts of known radionuclides in standard compared to amounts detected by our system.

Radionuclide	Amount Added (pCi/l ± SD)	Amount Determined (pCi/l ± SD)
^{134}Cs	400 ± 20	377 ± 30
^{137}Cs	450 ± 23	469 ± 13
^{65}Zn	497 ± 25	461 ± 48
^{60}Co	425 ± 21	391 ± 16
^{106}Ru	497 ± 25	526 ± 110

Analytical Laboratory

We have now installed and tested the solid-state electronic interface and timer sequence controller (referred to as DACU: Data Acquisition and Control Unit), as described in our 1974-75 progress report. The unit has been in constant use with remarkably little trouble. It has been connected to all three analytical instruments (Atomic Absorption Spectrophotometer, UV-VIS Spectrophotometer and the pH electrometer) and has performed even better than originally anticipated. Since the unit was installed in July 1975, approximately thirteen thousand analyses have been made using the unit.

Other equipment purchased for the Atomic Absorption Spectrophotometer (AAS) include a Background Corrector Unit (Varian Techtron Model BC-6), a motorized wavelength-drive unit and an emission-chopper unit (Varian Techtron). The Background Corrector allows us to perform simutaneous background correction. This is especially important for removing the non-atomic signal contribution when analyzing the elements lead and cadmium in complex matrices. Previously it was necessary to make these measurements separately, which required additional sample solution and more time. Under this system there was always the possibility of over or under correcting the desired signal, since instrument parameters would change slightly during a subsequent analysis (i.e. atomization efficiency changes caused by heating or cooling of the burner, etc.). The emission chopper and wavelength scanner now facilitate analysis of a sample for multiple elements such as calcium, sodium, potassium and magnesium in a single matrix all at once (i.e. single scan). Use of the emission chopper gives us the capability to analyze elements for which we do not have spectral line sources (required for atomic absorption analysis). It also provides lower limits of detection than with the AAS for a few specific elements, notably europium.

Computer (PDP-11/05). Due to the upgrading of our computer from a cassette-based operating system to that of a disc-based (RT-11) system, our former computer AAS data-reduction software programs required some limited modifications. These were made and the programs are now resident in disc storage rather than cassette tape storage. Access speed to programs by the PDP-11 processor has reduced the total time for data reduction of AAS data file by at least twenty-five to thirty percent. Future modification of the AAS software programs is intended to include a lower limit of detection calculation for each element analyzed based upon data generated from a least-squares linear regression analysis of AAS standards and a confidence interval based on linear regression applications (Larsen and Wagner, 1975).

Aquarium Room

The marine aquarium room at the OSU Radiation Center is now fully in operation. The large (1000 liter) batch-culture phytoplankton experiments are under way. We are also using the laboratory for other experiments using radioactive tracers.

Marine Science Center Wet Laboratory

We are now operating a functional wet laboratory at the OSU Marine Science Center in Newport, Oregon. The laboratory is supplied both with flowing seawater and freshwater supplies. The laboratory was originally designed to house thermal experiments, but it has proved effective for other pollutant-associated research. This area is not currently cleared for radiotracer work.

REFERENCES

Altshuler, B., and B. Pasternack. 1963. Statistical measures of the lower limit of detection of a radioactivity counter. *Health Phys.* 9:293-298.

Larsen, I.L., and J.J. Wagner. 1975. Linear instrument calibration with statistical application. *J. Chem. Ed.* 52:215-218.

Pasternack, B., and N.H. Harley. 1971. Detection limits for radionuclides in the analysis of multi-component gamma ray spectrometric data. *Nuclear Instruments and Methods* 91(3):533-540.

STUDENT PARTICIPATION

Students contribute substantially to our research program. During the past year one student completed a master's degree. An abstract of his thesis is printed in the following section. The name of each person's thesis advisor is given in parentheses.

DEGREES COMPLETED

David Stein, B.A. (1969) Humboldt State; M.S. (Fisheries) OSU

Mr. Stein, an OSU Oceanography staff member since 1969, completed his thesis, "A Review of the Deep Water Liparidae from the Coast of Oregon and Adjacent Waters," in October 1975 in Fisheries. His work is included here because it was supported in part on this ERDA contract. He is continuing his studies on deepwater benthic fishes. (Carl Bond, Fisheries)

Ph.D. CANDIDATES

David W. Evans, B.S. (Chemistry) U.C.L.A.; M.S.(Oceanography) OSU.

Mr. Evans spent two and one-half years at the Atlantic Estuarine Fisheries Center of the National Oceanic and Atmospheric Agency in Beaufort, North Carolina. During that time he performed research on the chemical reactions of transition metals in estuarine waters which will form the basis of his dissertation. He returned to Corvallis in May 1975 and has completed his additional course work since that time. He is currently writing his thesis with completion expected during the summer of 1976. (Cutshall)

Arthur Russell Flegal, Jr., B.A. (Zoology) U.C.S.B.; M.A. (Biology) University of California-Hayward.

Mr. Flegal is now preparing to start his thesis research work. He is interested in the flux of trace elements from suspended sediments to particulate organics, including detritus, phytoplankton, and zooplankton in the estuary. (Cutshall)

Vernon G. Johnson, B.S. (General Science) OSU; M.S. (Oceanography) OSU.

Mr. Johnson is currently working on his thesis on radionuclide transport in the Columbia River. Part of this work was presented at the Fourth National Symposium on Radioecology. In addition to working on his thesis, Mr. Johnson has worked for us on the Miller Sands project in the Columbia River estuary during the past year. (Cutshall)

MASTER OF SCIENCE CANDIDATES

John Steven Davis, B.A. (Biology) University of California-Riverside.

Mr. Davis spent the summer and early fall of 1975 collecting data in the Columbia River estuary that will form the basis for his thesis. His research will increase knowledge of the behavioral strategies employed by various species of amphipods in this ecosystem. These amphipods constitute an important link in the food chain of commercially important fish species. He is currently supported by other funds. (Holton)

Katsuo A. Nishikawa, K., B.S. (Oceanography) Escuela Ciencias Marinas, Enseñada, B.C. MEXICO

Mr. Nishikawa has completed his course work and his research. He is currently analyzing his data and starting to write his thesis. He is studying the partitioning of various metals upon injestion by the pacific oyster, *Crassostrea gigas*. He is currently supported by the Mexican government. (Cutshall)

Gerhardt F. Riedel, B.S. (Oceanography), A.B. (Biology) Humboldt State

Mr. Riedel is currently conducting his thesis research. His study of the uptake of various chemical forms of metal by phytoplankton and mixed phytoplankton-zooplankton cultures will increase our understanding of the cycling of metals in marine systems. (Cutshall)

Thomas J. Roffe, B.S. (Biology) University of California-Santa Cruz.

Mr. Roffe has been preparing ashed samples of pelagic animals for atomic absorption spectrometry and analyzing data on trace element concentrations of zinc, manganese and cesium. He plans to conduct thesis research on marine mammals with Dr. Bruce Mate. (Pearcy)

Daniel L. Simmons, B.S. (Zoology) Arizona State

Mr. Simmons is currently completing his thesis research at the Marine Science Center in Newport, Oregon. His study of the effects of thermal insults on marine copepods will increase our understanding of the interaction of these species with the physical environment. He is currently supported by other funds. (Holton)

ABSTRACTS OF THESES COMPLETED

RLO-2227-T12-55

A REVIEW OF THE DEEP WATER LIPARIDAE FROM THE COAST OF OREGON AND ADJACENT WATERS

M.S. Thesis by David L. Stein*

Descriptions and distributions are given of 28 species of Liparidae occurring or possibly occurring below 200 m between San Francisco and northern Vancouver Island, with keys for their identifications.

Nine genera are included: *Careproctus, Elassodiscus, Lipariscus, Nectoliparis, Rhinoliparis, Acantholiparis, Paraliparis,* and two genera are described, *Odontoliparis* and *Osteodiscus*. Eight new species are described: *Careproctus filamentosus, C. microstomus, C. oregonensis, Osteodiscus cascadiae, Odontoliparis ferox, Paraliparis paucidens, P. megalopus, and P. pectoralis.* One generic change is made: *Elassodiscus caudatus* is removed from *Paraliparis*. Second records of four species are reported: *Careproctus longifilis, C. ovigerum, Paraliparis latifrons,* and *Acantholiparis caecus*. The occurrence of four known species previously unreported from Oregon is recorded: *Careproctus longifilis, C. ovigerum, Paraliparis latifrons,* and *P. rosaceus*. Previously doubtful Oregon occurrence of two species, *Paraliparis dactylosus* and *P. ulochir*, is verified. Two rare species were collected: *Paraliparis cephalus* and *P. mento*. Four species are included as possibly occurring off Oregon: *Careproctus cypselurus, Rhinoliparis barbulifer, Paraliparis deani,* and *P. melanobranchus*. Other species present are *Careproctus melanurus, C. gilberti,* and *Acantholiparis opercularis*.

Of the 28 species from the study area, four are considered to be pelagic: *Nectoliparis pelagicus, Lipariscus nanus, Rhinoliparis attenuatus,* and *Rhinoliparis barbulifer*. The benthic species are shown to be divisible into two groups based on depth distributions. The shallower group contains 13 species from 2200-3600 m.

Morphological characters are related to the depth distributions of the species possessing them. Shallower species tend to have more pectoral and caudal rays, and more pyloric caeca than deep living species; they have pale skin, and darkly pigmented stomachs; the opposite is true in deeper species. The more primitive genera of deep water liparids do not occur shallower in the study area than more derived genera, although the more primitive species within those genera do.

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*Major Professor: Carl Bond, Ph.D., Fisheries, Oregon State University.

MEETINGS ATTENDED

John J. Bolen

*Fourth National Symposium on Radioecology
Oregon State University, Corvallis
May 12-14, 1975

Norman H. Cutshall

American Chemical Society
109th National Meeting
Philadelphia, Pennsylvania
April 7-11, 1975

*Fourth National Symposium on Radioecology
Oregon State University, Corvallis
May 12-14, 1975

Hanford Life Sciences Symposium
"Biological Implications of Metals in the Environment"
Richland, Washington
September 29-October 1, 1975

*Workshop on Standard Reference Materials in Marine Science
U.S. National Bureau of Standards
Santa Catalina, California
October 8-9, 1975

Priscilla J. Harney

*Fourth National Symposium on Radioecology
Oregon State University, Corvallis
May 12-14, 1975

Robert L. Holton

American Institute of Biological Sciences
Annual Meeting
Corvallis, Oregon
August 17-22, 1975

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*Abstracts or manuscripts of papers presented are included in the following section.

Fourth National Symposium on Radioecology
Oregon State University, Corvallis
May 12-14, 1975

Vernon G. Johnson

*Fourth National Symposium on Radioecology
Oregon State University, Corvallis
May 12-14, 1975

Earl E. Krygier

*Fourth National Symposium on Radioecology
Oregon State University, Corvallis
May 12-14, 1975

William G. Pearcy

*†American Institute of Biological Sciences Annual Meeting
Oregon State University, Corvallis
August 18-22, 1975

*Fourth National Symposium on Radioecology
Oregon State University, Corvallis
May 12-14, 1975

Walter H. Pearson

†Third International Estuarine Research Conference
Estuarine Research Federation
Galveston, Texas
October 6-9, 1975

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*Abstracts or manuscripts of papers presented are included in the following section.

†Paper presented is included in the "Research Completed" section.

PAPERS PRESENTED

ACCUMULATION AND CYCLING OF HANFORD PRODUCED ^{32}P BY THE JUVENILE STARRY FLOUNDER *PLATICHTHYS STELLATUS* (PALLAS) IN THE COLUMBIA RIVER ESTUARY[1]

John J. Bolen[2] and William C. Renfro[3]

Seasonal variations in concentration, turnover and mode of accumulation of ^{32}P by the juvenile starry flounder was investigated during 1969-70 in Alder Slough, a small segment of the Columbia River Estuary. Concentrations of ^{32}P and specific activity in flounder followed a similar annual cycle. Radioactivity was maximum in the spring and summer and minimum in fall and winter. Concentration factors for flounder ranged from about 600 in winter to near 200,000 in summer. Retention studies using caged fish, and turnover estimates from free-living flounder showed effective half-lives of ^{32}P on the order of 10-14 days. These results suggest that ^{32}P elimination from flounder occurred at about the same rate as physical decay of the radionuclide. The amounts of ^{32}P accumulated by flounder from food, water or labeled sediment was determined. Flounder fed labeled food or labeled food and sediment mixtures accumulated respectively 3.3 and 3.7 times the activity of flounder receiving ^{32}P from water alone. Radioactive sediment did not appear to make a direct contribution to the ^{32}P body burden of the fish.

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[1] Presented at the Fourth National Symposium on Radioecology, Corvallis, Oregon, May 12-14, 1975.

[2] 2900 N.W. Ashwood Drive, Corvallis, Oregon

[3] Environmental Programs, Northeast Utilities Co., Hartford, Connecticut

DECLINE OF ^{65}ZN, ^{54}MN, ^{60}CO LEVELS IN COLUMBIA RIVER SEDIMENT: 1971-1974[1]

Norman H. Cutshall and Vernon G. Johnson

Surface sediments collected in the reservoir behind McNary Dam have been analyzed for radionuclide content. Decline of radionuclide content in sediments following closure of reactors at Hanford was more rapid than expected from radioactive decay. Three additional processes were considered to account for this decline: chemical exchange with overlying waters, erosion of surface layers of sediment, burial by new deposits of lower radioactivity. The last process appears to be most important.

New inputs of ^{54}Mn and ^{60}Co into McNary reservoir sediments occurred in late 1972 and during 1973.

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[1] Presented at the Fourth National Symposium on Radioecology, Corvallis, Oregon, May 12-14, 1975.

USE AND MISUSE OF STANDARD REFERENCE MATERIALS IN MARINE SCIENCE[1]

Norman H. Cutshall

Standard Reference Materials (SRMs) are distributed by the National Bureau of Standards with the intention that they be "...used to calibrate measurement systems and to provide a central basis for uniformity and accuracy of measurement" [1]. In addition to these vital and important purposes, other uses of SRMs evolve in the user community. It is my purpose here to explore these other uses, to identify some problems with such uses and to seek means for solving the problems. I shall focus upon SRMs for marine science with emphasis on trace materials.

During recent years increased attention has been focused upon trace constituents in samples collected from the marine environment. Concern for pollution problems, often involving trace materials, has brought many analysts into the marine field. At the same time, more rapid and widespread communication of results has highlighted certain problems.

In examining the current literature it is frequently seen that values for trace concentrations are unexpectedly divergent. All too often a prominent hypothesis for the explanation of the divergence is that one or more of the measurement systems is at fault. This situation is not too

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[1] Presented at the Workshop on Standard Reference Materials in Marine Science, U.S. National Bureau of Standards, Santa Catalina, California, October 8-9, 1975.

surprising, however, because measurement systems (defined as the entire set of operations and tools involved in acquisition, processing and analysis of samples) for marine samples are highly complex. Measurement systems involve many steps which must be carried out under conditions which are not always ideal for preservation of sample integrity. Indeed the opportunities for contamination of marine samples are almost incredibly numerous. The scientific community engaged in trace analysis of marine samples has looked to SRMs as a means for assuring consistency among the different laboratories.

What benefit to the community can SRMs offer? Obviously SRMs provide no test of sample collection, preservation or storage segments of a measurement system. Only the sample preparation and analysis procedures are tested. Analysis of SRMs does not automatically confer validity upon the data of a community. Rather community benefit must be derived from individual laboratory benefit. Ideally, the SRM would be analyzed using methods which are standard in the laboratory making the analysis. It is pointless to develop methods only for SRM analysis. Problems in analyses are indicated to the individual laboratory when their results for an SRM fail to match the certified values. At this signal it is incumbent upon the individual laboratory to locate and eliminate their source of error. At least in the case of trace materials in marine samples it is not adequate to simply rely upon SRMs as primary standards against which measurement systems are calibrated. That is, the SRM results ought not be used as a fudge factor to simply "adjust" laboratory output data values. (There are situations where primary standardization against SRMs is appropriate; i.e. isotopic standards.) The diversity of causes for error in trace analysis of marine samples is so great that it is not feasible to rely upon SRMs to provide corrections for yields, matrix interferences and instrument response parameters. Rather a mismatch between the individual laboratory result and the standardized value should be taken to indicate that something is amiss. Further action to uncover and eliminate the source of error is imperative. This effort at laboratory level probably provides the greatest community benefit as individual problems are eliminated and overall data quality improves.

Although Standard Reference Materials do not provide verification of the quality of data from an individual laboratory, the results of SRM analyses often appear to be offered as proof of quality. The user community is desperately in need of improved means of data quality verification, and it is unfortunate that a more reliable test for quality is not available. Perhaps SRMs might be used to develop such tests. I shall explore two possible applications. Each of these is designed to minimize the impact of foreknowledge of the "correct" answer during analysis. (Table 1 demonstrates that results of analysis of "knowns" are more precise than results of analysis of "unknowns.") The two approaches might be called the "neo-SRM" approach and the "blended-SRM" approach. The neo-SRM approach is applicable during the time before standardization is completed. Analyses made during this time are of necessity for "unknowns" and are therefore unquestionably free from the influence of foreknowledge of the "correct" result. The neo-SRM approach would require wide advertisement and distribution of SRMs during their development stages, before any standardized values are available. Laboratories taking part in the neo-SRM analyses would receive advance distribution of material and would submit their results to a central holding office, perhaps but not necessarily at NBS. Results identified as neo-SRM results would soon receive special recognition in the user community.

Table 1

Results of Analysis of "Known" (Orchard Leaves) and "Unknown" (Bovine Liver) for Copper and Zinc. Values are reported as fractions of NBS values.

Laboratory	Copper		Zinc	
	Known	Unknown	Known	Unknown
A	0.93	1.05	1.09	1.00
B	1.00	1.02	0.98	0.92
C	1.01	0.99	1.11	1.05
D	0.83	0.62	1.06	0.61
E	0.98	0.97	1.06	1.33
F	0.96	0.94	0.99	1.32
G	0.99	0.95	1.10	1.12
H			1.16	1.05
mean	0.96	0.93	1.07	1.05
standard deviation	0.06	0.14	0.06	0.23
coefficient of variation	6.5%	15.4%	5.7%	21.9%
number of laboratories	7	7	8	8

(Data adapted from Reference 2)

The blended-SRM approach would rely upon accurate mixing of SRM portions from more than one source. The analyzing laboratory would not be informed of the relative proportions of the mixture and therefore would have foreknowledge only of the range of possible values. A number of SRM components would have to be individually standardized. So long as the potential number of components in a mixture exceeded the number of standardized values for each component, it would be mathematically impossible to calculate a result for one element from another. This might require several components and the difficulty of administering the approach would increase substantially as more and more elements were standardized. Practically, however, four or five components would probably assure that the difficulty in computing results would exceed the difficulty of measuring them. Again each laboratory would report its results to a central office such as NBS and would gain increased recognition and respect from results identified with blended-SRM analysis.

The neo-SRM approach offers the advantage of simplicity of administration, although its continued use would require continued development of new SRMs. Potential participants would be forced to the schedule of development of new SRMs. The qualities which accrue to a long established SRM, very high user-community confidence and widespread familiarity, would be lacking. On the other hand the blended-SRM approach would be based upon older SRMs and would be available upon a short term, unscheduled time basis. Means for accurate blending of several components would be needed. Administration of the blended-SRM approach would be more difficult.

In summary, Standard Reference Materials for trace constituents in marine samples are useful primarily to individual laboratories for testing routine laboratory procedures. The value of SRMs to the scientific community for external verification of results might be improved if analyses could be made in the absence of knowledge of the standardized values. This end might be accomplished through early distribution of periodically developed new SRMs or by a program of mixing of SRMs prior to distribution.

References

[1] Standard Reference Materials: 1973 Catalog, National Bureau of Standards Publication 260, U.S. Government Printing Office, Washington, D.C. 20402 (1973).

[2] Baseline Studies of Pollutants in the Marine Environment and Research Recommendations: The IDOE Baseline Conference, May 24-26, 1972, New York (1972).

SOME OBSERVATIONS OF RADIONUCLIDES IN WATER AND SEDIMENT OF THE LOWER COLUMBIA RIVER AND ESTUARY FOLLOWING CLOSURE OF KE REACTOR[1]

Vernon G. Johnson, Norman H. Cutshall, and Jerome J. Wagner

Periodic measurements of radioactivity and selected trace metals in water and sediment were initiated shortly after final shutdown of the last Hanford plutonium production reactor in January 1971. This effort was undertaken with the hope of taking advantage of the enhanced sensitivity provided by the terminated radionuclide input for observing sediment-water transfer and exchange. The salient features of the data from this study will be presented and its relevance to the general question of recycling of sediment bound trace metals and radionuclides in fluvial and estuarine ecosystems will be discussed.

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[1] Presented at the Fourth National Symposium on Radioecology, Oregon State University, Corvallis, May 12-14, 1975.

COBALT-60 IN ALBACORE TUNA OFF THE WEST COAST OF NORTH AMERICA[1]

E.E. Krygier

Radioactive cobalt-60 (^{60}Co), found in livers of albacore tuna collected in 1963-1969 from the west coast of North America (Washington to Baja California), exhibit geographical, annual and within-season variations. Variations are related to the two possible sources of ^{60}Co, the Columbia River and atmospheric fallout, the latter being more important. Highest concentrations of ^{60}Co in tuna were found to occur at latitudes from 42° to 43°N during the first part of summer; thereafter a steady decline in radioactivity is observed through the remaining summer months. Annual variations were noted to differ between waters of Oregon and Southern California, which lead to speculation on migration patterns by study of the radioactive cobalt "tag". A rough estimate of assimilation efficiency, 17.9%, of ^{60}Co is determined with pertinent data.

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[1] Presented at the Fourth National Symposium on Radioecology, Oregon State University, Corvallis, May 12-14, 1975.

DISTRIBUTION PATTERNS OF FISH LARVAE IN OCEAN WATERS OFF YAQUINA BAY, OREGON--AN UPWELLING AREA[1]

Sally L. Richardson and William G. Pearcy

A one and one-half year survey of planktonic fish larvae from 2 to 111 km (1 to 60 mi) off the Oregon coast yielded 287 samples which contained 23,578 individuals in 86 taxonomic groups (76 at the species level). Twenty-six families were represented with the Cottidae and Pleuronectidae being the most speciose.

Sampling with large mouth (0.7 m) bongos having 0.571 mm mesh nets was conducted monthly from January 1971 to August 1972 at 12 stations along an east-west transect off Yaquina Bay, Oregon.

During 1971, 93% of all larvae were taken during the 6 month period from February to July.

Two distinct faunal assemblages were found--"coastal" (2 to 28 km offshore) and "offshore" (46 to 111 km offshore). The "coastal" group was dominated by osmerids, *Parophrys vetulus*, *Isopsetta isolepsis*, and *Microgadus proximus*. Distributional patterns within the coastal group can be separated into three components--1) estuarine, 2) coastal before upwelling, 3) coastal during upwelling. The "offshore" group was dominated by *Sebastes* spp., *Stenobrachius leucopsarus*, *Tarletonbeania crenularis*, and *Lyopsetta exilis*. Distributional patterns within the "offshore" group may be separated into 1) outer-shelf, demersal origin, 2) oceanic, pelagic origin, 3) Columbia River Plume associated.

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[1] Presented at the American Institute of Biological Sciences Annual Meeting, Oregon State University, Corvallis, August 18-22, 1975.

VERTICAL TRANSPORT OF ^{65}ZN INTO THE DEEP SEA[1]

William G. Pearcy

Data on ^{65}Zn activity of oceanic animals captured from the surface (neuston) to 2800 m (abyssobenthics) have been tabulated for winter and summer seasons, for three time periods (1962-1970) and for four taxa (fishes, crustacea, cephalopods and medusae), punched on computer cards and regressed against depth of capture or depth of maximum nighttime abundance for each species. The slope of the lines fitted to the log ^{65}Zn data therefore estimates the rate of change of radioactivity with depth. This can be interpreted as a transport rate from surface waters into the deep-sea. Although the variability of the data is large, the slopes of the regressions are often similar.

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[1] Presented at the Fourth National Symposium on Radioecology, Oregon State University, Corvallis, May 12-14, 1975.

RESEARCH IN PROGRESS

RADIOECOLOGICAL STUDIES IN THE COLUMBIA RIVER AND ESTUARY

POST-SHUTDOWN RADIONUCLIDE TRANSPORT STUDY

Vernon G. Johnson, Norman H. Cutshall, Jerome J. Wagner,

I. Lauren Larsen and Robert L. Holton

This study was initiated in February 1971 when operation of the last "once through" plutonium production reactor was terminated. The objective of the project was to use the residual, Hanford-produced radioactivity in riverbed sediment for a field study of relationships between sediment-water transfer or exchange processes and dispersion of sediment-bound radionuclides in a fluvial and estuarine section of the Columbia River system. By taking the "tracer" view of the radioisotopes in the system it was expected that information would be gained concerning mechanisms and dynamics of trace metal recycling from bottom sediment in a major aquatic ecosystem. Highlights of this work were presented in two papers given at the Fourth National Symposium on Radioecology in May 1975 (Abstracts of these papers are included in the "Papers Presented" section of this report.) Mr. Johnson is preparing a final account of this project, part of which will serve as the basis of his Ph.D. dissertation. A modest effort is planned to follow the rate of decline for as long as detectable activity is present.

Related Studies

Two projects were undertaken in the 1974-75 period which made use of some of the ideas and methods developed in the above study. Data acquired from these related projects thus serve as supplemental data for the above study. One of these projects was a geochemical baseline investigation of bottom sediment in Youngs Bay for a proposed aluminum reduction facility and the other was a physical-chemical baseline study at Miller Sands Island in the upper reach of the estuary. A final report for the Youngs Bay study was issued in May 1975 and a final report for the Miller Sands study is in final stages of preparation.

Youngs Bay Geochemical Baseline. This study (Johnson and Cutshall, 1975) involved the analysis of 85 surface and 131 subsurface (cores) sediment samples for radionuclides, fluoride, iron, manganese, zinc, cadmium, copper, lead, nickel, chromium and cobalt. Selected samples were also processed for total sulfide. Radionuclide data was used to estimate sedimentation rate (cobalt-60/europium-152 ratios) and to distinguish between contemporary and pre-Hanford age strata. These estimates were helpful in assigning a time scale to sediment profiles.

Fluoride concentrations were found to be controlled by sediment texture (fine sediment content) which was related to mineralogy (i.e. clay minerals are enriched in the fine fraction of sediment deposits and are known to be high in total fluoride).

Trace metals were also related to particle size, being generally higher in fine textured sediments than in coarse. The exception to this distribution was manganese which showed surface enrichment in several core profiles where sediment texture was invariant. This was attributed to reduction in deeper layers and upward migration in the interstitial water followed by entrapment in the surface oxidized layer.

Miller Sands Study. This project involved physical and chemical characterization of water, sediment and soil on and in the vicinity of Miller Sands Island (located at river mile 25 in the upper reach of the Columbia River estuary). This work was part of the Habitat Development Program managed by the Waterways Experiment Station, Army Corps of Engineers, Vicksburg, Miss. This program is part of a broad, national program designed to evaluate beneficial uses of dredged material as an alternative to current disposal practices.

Measurements made included: 1) soluble and particulate trace metals and nutrients in water, 2) total and interstitial trace metals and nutrients in sections of sediment cores, and 3) trace metals and fertility parameters in island soil samples. Selected core sections were analyzed for radionuclide content in order to estimate relative age and sedimentation rate. In addition to providing baseline data, some of the results of this study provide field evidence for proposed mechanisms by which trace metals (and radionuclides) may be mobilized in and from bottom sediment (Cutshall, In Preparation).

Information gained in both of these related studies will be helpful in evaluating mechanisms and rates of material transfer/exchange between water and bottom sediment in the Columbia River system. It should also be noted that the radionuclide measurements made in the above studies serve a dual purpose, because they provide additional radioactivity baseline data for nuclear operations upstream from the estuary (The Trojan Nuclear Power Plant, located approximately 50 miles upstream from the estuary, began operation in December 1975).

SEDIMENT-ORGANISM CYCLING OF TRACE RADIONUCLIDES: UPTAKE AND LOSS OF ZINC-65 AND MANGANESE-54 BY THE FRESHWATER CLAM *ANODONTA*

Priscilla Harney, Robert L. Holton and Norman H. Cutshall

Loss of ^{65}Zn and ^{54}Mn from the freshwater clam *Anodonta* was examined to determine whether uptake of these nuclides occurred from sediments, the only significant radionuclide source remaining after production reactor shutdown at Hanford, Washington, in January 1971.

A comparative field loss study was performed to determine whether uptake of ^{65}Zn and ^{54}Mn by *Anodonta* occurred after shutdown. Loss rates of these two nuclides were measured in organisms transferred to the nonradioactive Willamette River and were compared to loss rates found in organisms remaining *in situ* in the Columbia River. Half-lives were time-averaged after four months of study. The resulting ^{65}Zn half-lives of 103 ± 5 days and 136 ± 15 days for transfer and *in situ* groups, respectively, indicated that significant uptake took place after shutdown. Time-averaged results of periodic sacrifice collections of *Anodonta* from McNary Reservoir and the estuary further confirmed this finding. Large variation in the *in situ* ^{54}Mn data precluded a meaningful comparison of time-averaged loss rates, although sacrifice collections yielded ecological half-lives that were not significantly different from the pooled effective half-life.

Examinations of *in situ* loss rates calculated on a monthly basis compared to time-averaged transfer loss rates indicated that uptake of both nuclides took place during restricted periods and that uptake and loss was relatively rapid. This suggests that uptake processes were important during confined periods, occupying about one-third of the total study time. For the majority of the study period, the Columbia River group was losing ^{65}Zn and ^{54}Mn at virtually the same rate as the transfer group, that is, at the effective loss rate, as though no radioactivity was present in the environment.

Specific activity data from several clams transferred in the opposite direction, from the Willamette River to the Columbia River at McNary Reservoir, support the observation that the uptake of ^{65}Zn from sediments is significant. A comparison of specific activity data was made between clams after they had spent at least a year at McNary Reservoir and bottom sediments taken from the same area of the reservoir. It is clear that uptake of ^{65}Zn available on sediments was significant; the clams attained from 22 to 51% of the ^{65}Zn specific activity of the sediments. Thus it appears that sediments contribute a significant fraction of zinc to these clams. Analysis of ^{54}Mn uptake was complicated by an apparent new input of ^{54}Mn some two years after reactor shutdown.

A mathematical model was formulated to describe the specific activity of a clam at any time after reactor shutdown under conditions where the clam was simultaneously losing activity via physical decay and biological turnover, as well as taking up activity from a still-active source which was itself losing activity via physical decay and turnover. Sediments were assumed to be the sole source of activity for the clams, and at various times after shutdown specific activities were calculated using field-determined loss-rate constants. These were compared to specific activities measured in resident clams collected at the same time. Comparison of observed and calculated ^{65}Zn specific activities indicated that, on the average, observed values were 32.1% of calculated values (range: 26.0-37.8%), with sediments taken to be the only ^{65}Zn source to the clams. This provides additional evidence to indicate that sediments act, if not as the predominating, at least as a highly significant radionuclide-trace metal source to these freshwater clams.

ACCUMULATION AND CYCLING OF HANFORD PRODUCED ^{32}P BY THE JUVENILE STARRY FLOUNDER *PLATICHTHYS STELLATUS* (PALLAS) IN THE COLUMBIA RIVER ESTUARY

John J. Bolen[1] and William C. Renfro[2]

Seasonal variations in concentration, and mode of accumulation of ^{32}P by the juvenile starry flounder was investigated during 1969-70 in Alder Slough, a small segment of the Columbia River estuary. Concentrations of ^{32}P and specific activity in flounder followed a similar annual cycle. Radioactivity was maximum in the spring and summer and minimum in fall and winter. Concentration factors for flounder ranged from about 600 in winter to near 200,000 in summer. The amounts of ^{32}P accumulated by flounder from food, water or labeled sediment was determined. Flounder fed labeled food or labeled food and sediment mixtures accumulated respectively 3.3 and 3.7 times the activity of flounder receiving ^{32}P from water alone. Radioactive sediment did not appear to make a direct contribution to the ^{32}P body burden of the fish.

A manuscript is now under preparation.

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ZINC-65 SPECIFIC ACTIVITY IN MUSSELS AND BARNACLES FROM THE OREGON COAST

I. Lauren Larsen, Jerome J. Wagner, and Robert L. Holton

The zinc-65 specific activity in mussels *Mytilus californianus* from along the Oregon Coast has been studied since closure of the single pass water cooled plutonium reactors at Hanford, Washington, from late January 1971 to the present. A conceptual model has been developed relating uptake, retention, and loss of zinc in these organisms. Environmental observations as well as laboratory growth rate studies have been initiated in order to evaluate effects of growth upon zinc turnover. A manuscript is now in preparation.

Studies of the specific activity of ^{65}Zn in the barnacle *Pollicipes polymerus* are also being continued.

SPECIFIC ACTIVITIES OF ZINC-65 AND VERTICAL BIOLOGICAL TRANSPORT INTO THE DEEP SEA.

William G. Pearcy, Earl E. Krygier, and Norman H. Cutshall

Data on ^{65}Zn radioactivities and specific activities of oceanic animals were regressed against depth of capture for three periods between 1962-1969 and for winter and summer seasons. Seasonal differences in slopes were not statistically significant.

Time differences required to explain differences of specific activities among depths were estimated from the physical decay of ^{65}Zn. Our calculations suggest that an atom of ^{65}Zn takes an average of two years to be transported from epipelagic animals to abyssobenthic fishes. Transport rates are slowest from the surface to 100 m depth, suggesting that ^{65}Zn is retained in biological systems near the surface.

TROJAN POWER PLANT STUDIES

I. Lauren Larsen and Robert L. Holton

A study program has been developed to assess the radiological conditions near the Trojan Power Plant site downstream from Goble, Oregon, on the Columbia River. Levels of radioactivity in water, sediments and in the fresh water clam *Anodonta* were established by sampling before the plant began operation. Current efforts will determine the radioactivity in each of these compartments during the first several years of plant operation.

We have previously found the clam, *Anodonta*, to be an excellent biological integrator of several radioisotopes. The lack of natural clam beds immediately below the plant has made it necessary to transport clams to this location. We are fortunate in being able to use the National Marine Fisheries Service laboratory at Prescott, Oregon, for culturing the clams. This lab is less than one-half mile below the power plant and can supply the culture tanks with a constant flow of unfiltered river water.

We are currently analyzing samples for gamma activity only, but will supplement this with a counting for alpha particles as soon as our alpha counting system is functional.

We are currently expanding this program to include sampling in the Willamette River, where only fall-out should contribute to the radioactivity of samples. The Willamette samples should make it possible to distinguish the source of any radioactitivy detected, when examined in conjunction with samples taken above Trojan, where fallout and activity from Hanford may be observed, and the samples from below Trojan, which would integrate radioactivity from all three potential sources.

DIEL AND TIDAL ACTIVITY PATTERNS OF ESTUARINE INVERTEBRATES IN THE COLUMBIA RIVER, WITH SPECIAL REFERENCE TO THE GAMMARID AMPHIPOD *COROPHIUM SALMONIS*

John S. Davis and Robert L. Holton

Preliminary investigation in Youngs Bay has revealed that *Corophium salmonis*, a tube-dwelling gammarid amphipod, is the numerically dominant intertidal and subtidal infaunal resident. This dominance is reflected in the prevalence with which *Corophium* is found in fish stomachs.

An investigation was carried out that sampled the near-surface, mid-water and epibenthic regions at the mouth of Youngs Bay over complete tidal and diel periods in September 1975. *Corophium*, and its faunal associates, *Anisogammarus*, *Neomysis*, *Eohaustorius* and *Crangon* were found in surface water only during periods of darkness. Further correlation with environmental variables, such as tide stage, will attempt to discern an additional activity pattern of tidal period. Further studies will compare the size, sex and reproductive state of the swimming animals relative to those remaining in the substrate and will investigate persistent endogeneous rhythms for individuals kept under constant environmental conditions in the laboratory.

CYCLING OF METALS IN NATURAL SYSTEMS

CYCLING OF Mn, Zn, Cu, AND Fe IN THE NEWPORT RIVER, NORTH CAROLINA

David W. Evans, Norman H. Cutshall, Ford A Cross, Douglas A. Wolfe

Field research on the cycling of these trace metals has been completed and data analysis is nearing completion.

Manganese cycling is characterized by transfers from bed sediments to solution occurring in the upper estuary and subsequent precipitation in the lower estuary. Apparent upstream recycling and subsequent deposition of particulate manganese (Mn) maintains an approximate steady-state dissolved Mn maximum in the upper estuary. Reducing conditions in bottom sediments probably provide the means of solubilizing particulate Mn, and oxidation in the water column probably precipitates insoluble MnO_2 in the lower estuary. A full discussion of the behavior of Mn is given in a manuscript submitted for publication and included in this report.

Dissolved iron (Fe) is conservative within the estuary, and passes from the river through the estuary to the ocean without undergoing any net exchange with particulate forms. About 95% of the dissolved Fe in Newport River water is bound to organic matter. Organic matter, measured by absorption in the water of ultraviolet light in the range 250-350 nm, is also conservative. It is postulated that Fe is maintained in solution by chelation to dissolved organic matter. This contrasts with the behavior of iron in other estuarine systems where dissolved Fe is precipitated and flocculated downstream from the initial mixing of fresh and saline waters.

Zinc (Zn) and copper (Cu) show less consistent behavior. On most occasions, Zn and Cu show maxima in dissolved concentrations in the same region as the dissolved Mn maximum. A postulated bottom sediment source for the added dissolved Zn and Cu is not supported, however, by experiments in which bottom sediments were resuspended into the water column. These same experiments had shown bottom sediments capable of supplying dissolved Mn to the overlying water. At other times Zn and Cu appeared to be approximately conservative within the estuary. Their behavior follows no readily acceptable model.

Results will appear in final form in a dissertation to be completed during 1976.

CULTURING SOME YOUNG CLAM SPECIES OF OREGON

John B. Morgan and Robert L. Holton

Seven clam species ranging in age from less than one to three years are being cultured in water tables filled with selected sediments and flowing sea water from Yaquina Bay. Species being cultured presently include the soft shell clam (*Mya arenaria*); native littleneck clam (*Protothaca staminea*); heart cockle (*Clincardium nuttalli*); bent-nosed clam (*Macoma nasuta*); gaper clam (*Schizothacrus nuttalli*); butter clam (*Saxidomus giganteus*); freshwater clam (*Anodonta*). All clams are cultured in sediments similar to those from which they were collected.

Monthly growth rates are determined by measurement of total wet weight and length of ten to sixteen animals of each species. We will incidentally be determining which species are most amenable to laboratory culture conditions and are hence most useful for future studies. We hope to be able to handle *Anodonta*, a fresh water genus, *Mya*, a genus found in relatively low salinity waters, and one other genus from the more saline parts of the estuary for various studies of metal uptake and retention.

GROWTH RATE STUDIES OF THE MUSSEL *MYTILUS CALIFORNIANUS*

John B. Morgan, I. Lauren Larsen, and Robert L. Holton

Laboratory growth rates of mussels collected from Yaquina Head, Agate Beach, Oregon, are being determined. Mussels of various sizes are being cultured in aquariums under simulated day light illumination and a normal tidal cycle. A tidal device allows water levels to be synchronized with the tidal cycle at Yaquina Head.

Monthly growth rates (change in length and wet weight versus time) of laboratory cultured mussels are being compared with rates recorded from mussels at the same site during the same period of time.

Growth rate studies of mussels which have been submerged in a continuous water flow system for one year are being continued.

Knowledge of the growth rate of mussels is required for interpretation of the results of our radioactive turnover studies of mussels collected in the field.

TRACE ELEMENT CONCENTRATIONS IN OCEANIC ANIMALS

Thomas J. Roffe, William G. Pearcy, Earl E. Krygier

Concentrations of zinc, manganese, and cesium in midwater fishes, shrimps and euphausiids have been determined from ash samples of organisms collected during 1962-1964. Zinc had the highest average levels and highest variability, cesium was next highest, and manganese had the lowest concentration of these three trace elements in all animal groups. No difference was found in element concentrations between animals collected in deep ($\geq$ 450 m) and shallow ($<$ 450 m) waters.

Cesium concentrations were significantly higher in fishes than crustaceans, the difference being primarily a result of the difference between fish and shrimp in both seasons. Zinc concentrations were higher in crustaceans than fishes during the summer season, and higher in euphausiids than fishes during the winter season. Mean values, of all elements and for all three animal groups, excepting cesium concentration in fishes, <u>increased</u> from summer (March-September) to winter (October-April). Zinc concentrations in fishes increased by a factor of two between these two seasons.

TRACE METAL UPTAKE BY PHYTOPLANKTON AND ZOOPLANKTON

Gerhardt F. Riedel, Robert L. Holton and Norman H. Cutshall

A seawater laboratory is now in operation in the Radiation Center on the OSU campus. A study of the uptake of various chemical forms, organically bound and ionic, of zinc (Zn), manganese (Mn) and iron (Fe) is currently underway. Six 1200 liter tanks contain cultures of phytoplankton and copepods. The cultures were allowed to develop for several months with only the stable forms of the metals present. Recently ^{65}Zn, ^{54}Mn, and ^{59}Fe in ionic form were added to the cultures.

A regular sampling program has been established, and samples for radioactivity analysis, stable element analysis, carbon, hydrogen and nitrogen analysis have been taken. The radioactivity samples are currently being analyzed, and the rest are being stored for analysis later.

A STUDY OF THE ELEMENTAL FLUX FROM SUSPENDED SEDIMENTS TO ORGANIC PARTICULATES IN THE ESTUARINE ZONE

A. Russell Flegal, Norman H. Cutshall, and Robert L. Holton

The objective of this research is to further define the flux of selected trace elements from the suspended sediments to the particulate organics (detritus, phytoplankton and zooplankton) in the estuarine zone. This has been identified as a poorly understood, yet critical, pathway of environmental transfer and has been given a high priority for future research needed to assess potential ocean pollutants (NAS, 1975).

Samples are being collected in the field to determine ambient or background conditions in a relatively unpolluted system. Additional laboratory studies will be conducted to support the *in situ* data and to simulate potentially polluted conditions.

A series of samples are being taken from the Alsea River, where the only industries are logging activities and fishing and the sole wastewater outfall is located at Waldport. The seston is collected with a continuous flow centrifuge and separated into its major constituents by density gradient centrifugation, using a modification of the methods developed by Lammers (1968). Dissolved metals in filtered water (0.2 μm) are concentrated with Chelex resin. The trace element analyses are made by atomic absorption spectrophotometry and neutron activation. Other measurements are being made of the C, H, N contents and total surface area of the particulates and physical/chemical characteristics of the water. Sampling is done along the length of the transition zone of the estuary to determine the spatial flux, and will be repeated several times to determine the magnitude of diurnal and seasonal variations.

Because of the presumed absence of anthropogenic sources of additional trace elements along the Alsea, an effort will be made to model the flux of trace elements from the suspended sediments to the organic particulates using a point source input.

The model will be tested in laboratory studies in which suspended sediments, in dialysis bags, will be activated and placed in phytoplankton cultures of varying salinities and pH. Then, after a defined period, the elemental flux into the phytoplankton and solute may be measured. By increasing the initial amount of a trace element(s) adsorbed on the sediment, it may be possible to simulate supra-background conditions to determine variations in the metal's exchange ratio and associated effects on phytoplankton productivity.

The study is primarily oriented toward the flux of manganese, which is mobile in the transition zone (Evans and Cutshall, 1973) and small amounts of its oxides on particulates may significantly affect the sorption of transition series metals (Jenne, 1970).

CONCENTRATION OF CADMIUM, ZINC, COPPER, MANGANESE AND IRON IN THE SOFT TISSUES OF THE MUSSEL *MYTILUS CALIFORNIANUS* FROM BAJA CALIFORNIA, MEXICO

Katsuo Nishikawa, Jerome J. Wagner and Robert L. Holton

During the summer of 1974 California mussels (*Mytilus californianus*) were collected at two locations: one within the Southern California Bight (Punta Banda, Lat. 30° 43.5'N, Long. 116° 43.5'W) and the other just southeast of Punta Eugenia (Punta Abreojos, Lat. 26° 43.5'N, Long. 113° 36'W). Punta Banda is located close to the south edge of the bight, and is an area where intense human activity is taking place. In contrast, Punta Abreojos is 650 km south of Punta Banda and has no major human settlement.

From each location we collected 28 to 50 mussels of which we determined the metal concentration by atomic absorption spectrophotometry. Of the five metals (cadmium, zinc, copper, manganese, and iron) analyzed, the concentrations of Cd and Zn were especially interesting. In the Punta Abreojos mussels we found an anomalously high concentration of Cd, whereas in the Punta Banda samples the Zn concentration was anomalously high.

The elevated Cd levels in Punta Abreojos are in agreement with the concentrations reported by Martin and Broenkow (1975) on plankton samples collected offshore of this area. However, the high Zn concentrations in Punta Banda are probably related to the local input of the metal through the underwater hot springs located at about 15 fathoms, about 200 m offshore from the collection area.

The results of this work will be presented in Spanish at the Primera Reunion Latinoamericana sobre Ciencia y Tecnologia de los Oceanos, Veracruz, Mexico, 26 May to 1 June 1976.

CONCENTRATIONS OF CADMIUM, ZINC, COPPER, MANGANESE AND IRON IN THE BIODEPOSITS OF *CRASSOSTREA GIGAS*

Katsuo Nishikawa and Norman H. Cutshall

The biodeposits of pacific oysters, *Crassostrea gigas,* were collected by using a specially designed apparatus. This instrument separates the fecal material from the pseudofeces and has the capability of working continuously.

From the middle of spring to midfall of 1975 we collected 19 samples of biodeposits and 14 samples of settleable solids on which the oysters feed. Each sample was analyzed for Cd, Zn, Cu, Mn and Fe by atomic absorption spectrophotometry.

The results are being analyzed to determine patterns in metal concentrations with time and to find the relationship between samples (feces, pseudofeces and settleable solids) and between metals and organic carbon and nitrogen in each of the phases.

PACIFIC OCEANIC STUDIES

DEEP-SEA BIOLOGY

William G. Pearcy, David Stein, Earl E. Krygier, Shelly Johnson

The large epifauna of the Cascadia Abyssal Plain has been intensively sampled during recent years with bottom trawls. The data on the catches of fishes in 168 of these collections has been entered on computer cards to facilitate a study of the distribution and the biology of deep-sea fauna. Changes in the abundance of individual species and species associations will be related to depth and distance from shore. The distributional features of the macrourid fishes (rattails or grenadiers) will receive special attention since they are the dominant family of fishes by weight, and often by numbers, in the trawl catches.

The Zoarcidae are an important benthic component of the Oregon coastal ichthyofauna occurring below 400 meters. Numerically, zoarcids are the third most abundant deep-sea family of fishes on the continental slope, and third most abundant on the abyssal plain. Twelve species have been collected by OSU off Oregon and identified by Shelly Johnson of Pasadena City College. Four (*Embryx crotalina, Lycenchelys camchaticus?, Lycenchelys* "A", and *Lycodes diapterus*) occur on the slope, and eight (*Bothrocara brunneum, B. molle, Lycenchelys* "B", "C", "D", and "E", *Maynea bulbiceps,* and *Taranetzella lyoderma*) occur on the abyssal plain. A total of 242 specimens representing the above listed species were examined; 54 represented the five new *Lycenchelys* species; the majority (159) were *E. crotalina* and *L. diapterus*. Apparently the slope species are much more abundant, but there are more species on the plain.

The systematics and biology of deep-sea octopods, a very poorly known group of animals, are being studied by Dr. G.L. Voss of the University of Miami, and by W. Pearcy. Four genera have been identified from trawl contents obtained on the Cascadia and Tufts Abyssal Plains off Oregon.

They include new species of *Graneledone, Benthoctopus* and *Grimpoteuthis*. A paper describing the abyssobenthic octopods off Oregon is in progress.

We are continuing our studies of the systematics, biology and vertical distributions of bathypelagic organisms collected in opening-closing mid-water trawl samples off Oregon between depths of 1000 and 2400 m. Crustaceans dominate these collections. Fishes are rare. Some bathypelagic animals have broad vertical distributions, extending into mesopelagic waters, (see paper by Pearcy et al., in "Research Completed" section). Abyssal benthic fishes have been captured in bathypelagic waters far above the sea floor (see paper by Pearcy in "Research Completed" section). Hence, migrations of animals inhabiting bathypelagic depths may accelerate vertical transport of materials into the deep sea.

A new species of *Acanthephyra* shrimp from bathypelagic depths is being described by Earl Krygier and Carl Forss.

CARBON-14 AND DEEP-SEA FOOD WEBS

William G. Pearcy and M. Stuiver

We are continuing our cooperative research with Dr. Stuiver at the University of Washington on Carbon-14 content of deep-sea animals in order to obtain estimates of pathways through the food web and transit times of food sources from surface waters. At present, samples are being analyzed to supplement those analyzed earlier.

STUDIES OF CESIUM-137 IN NORTH PACIFIC OCEAN SEDIMENTS

Robert L. Holton

The recent completion of our low level gamma ray counting system has allowed us to begin counting samples collected previously for the study of cesium-137 in North Pacific sediment. A major cruise will be taken in August 1976 to obtain additional samples for this work. We expect to obtain samples from stations on a transect from Alaska to Hawaii on this cruise as well as additional stations while steaming to reach this transect.

We currently must solve some software problems in data reduction to enable us to obtain better results in counting our cesium-137 samples. We expect to make these adjustments in the near future.

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RESEARCH COMPLETED*

RLO-2227-T12-39

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Linear Instrument Calibration with Statistical Application

Quantitative determinations involving instrumentation are often accomplished by establishing a calibration curve. Readings from the particular instrument are plotted against known concentrations of standards and the concentration of the analyte in the sample evaluated by the underlying relationship of the calibration line. If the calibration is linear, or nearly linear such that a straight line provides a reasonable approximation, the concentration of the sample can be readily determined if it falls within the range of the standards. The equation for a straight line is given by

$$Y = M \cdot X + I \tag{1}$$

where the Y value (instrument response) is a function of the concentration (X value); M is the slope of the calibration line, and I is the ordinate intercept (vertical axis). It may happen that the calibration is not linear. If this occurs several options are available. The unknown concentration can be determined graphically if sufficient reproducible data points are obtained from the appropriate standards. Curve fitting techniques, facilitated by use of a computer, may also be used. The application of transformations such as Beer's Law *(1)* to photometric absorption data, within a limited range, usually produces an adequate linear calibration line. Acton *(2)* discusses the use of transformations and some simple suggestions are given in reference *(3)*.

After establishment of the calibration line the analyst proceeds to analyze his samples. Usually this is done by taking repetitive measurements upon a sample, converting these determinations to concentration units via the calibration line, and then reporting a mean and error term based upon the repetitive measurements. However, as stressed by Linning and Mandel *(4)* this replication error may not provide a realistic estimate of the precision for the method. Why? Because the overall precision in establishing the calibration line may be poorer than that indicated by the sample replication error. The scatter of the data points establishing the calibration line provides a measure of the expected error in analyzing the sample and may be larger than that indicated by sample replication error *(4)*. The statistical uncertainty in establishing the calibration line must be considered.

Statistical Application

For a first approximation, a straight line drawn through the data points may be sufficient to evaluate the concentration in the sample. For quantitative analysis the application of linear least squares produces an unbiased calibration line. The best line which can be fitted is the one which minimizes the sum of the squares of the vertical distance between the data points and the fitted line *(5)*. This is referred to as the regression of Y on X, X being the independent variable (concentration) and Y, the dependent variable (instrument response). The assumptions underlying linear regression require that the data means at each concentration lie on the line $\dot{Y} = M \cdot X + I$ with the errors associated with the X values small in comparison to those associated with the Y values; these Y values are normally distributed with equality of variance at each corresponding X value, and are statistically independent *(2)*.

In order to facilitate writing computational formulas the symbols as given in Table 1 will be used to designate the required calculations (after ref. *(2)*). The slope for the equation of a straight line as given by eqn. (1) is

$$M = \frac{Sxy}{Sxx} \tag{2}$$

The intercept of the calibration line with the vertical axis which represents a background signal contribution in the absence of the analyte, is given by

$$I = \overline{Y} - M \cdot \overline{X} \tag{3}$$

Table 1. Symbols Indicating Designated Computations

$Sxx = \Sigma(X - \bar{X})^2$	$= \Sigma X^2 - \frac{(\Sigma X)^2}{N}$	$= \frac{N \cdot \Sigma X^2 - (\Sigma X)^2}{N}$
$Syy = \Sigma(Y - \bar{Y})^2$	$= \Sigma Y^2 - \frac{(\Sigma Y)^2}{N}$	$= \frac{N \cdot \Sigma Y^2 - (\Sigma Y)^2}{N}$
$Sxy = \Sigma(X - \bar{X})(Y - \bar{Y})$	$= \Sigma XY - \frac{\Sigma X \cdot \Sigma Y}{N}$	$= \frac{N \cdot \Sigma XY - \Sigma X \cdot \Sigma Y}{N}$
$\bar{X} = \frac{\Sigma X}{N}$		
$\bar{Y} = \frac{\Sigma Y}{N}$		
N = number of readings obtained		
Using weighted values		
$Swxx = \Sigma W(X - \overline{Xw})^2$	$= \Sigma WX^2 - \frac{(\Sigma WX)^2}{\Sigma W}$	$= \frac{\Sigma W \cdot \Sigma WX^2 - (\Sigma WX)^2}{\Sigma W}$
$Swyy = \Sigma W(Y - \overline{Yw})^2$	$= \Sigma WY^2 - \frac{(\Sigma WY)^2}{\Sigma W}$	$= \frac{\Sigma W \cdot \Sigma WY^2 - (\Sigma WY)^2}{\Sigma W}$
$Swxy = \Sigma W(X - \overline{Xw})(Y - \overline{Yw})$	$= \Sigma WXY - \frac{\Sigma WX \cdot \Sigma WY}{\Sigma W}$	$= \frac{\Sigma W \cdot \Sigma WXY - \Sigma WX \cdot \Sigma WY}{\Sigma W}$
$\overline{Xw} = \frac{\Sigma WX}{\Sigma W}$		
$\overline{Yw} = \frac{\Sigma WY}{\Sigma W}$		
W = appropriate weighting factor		

For statistical purposes computations using these formulas should be made to more than the required significant number of figures with the final desired results rounded to the significant number of figures.

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*Arranged by RLO number.

Table 2. Example of Data and Calculations

Standards & Blanks (X values) arbitrary units	Instrument Response (Y values)	
0	0.019	$N = 15$, Student $t_{(N-2,\ 0.95)} = 2.160$
0	0.024	$\Sigma X^2 = 22.50$
0	0.021	$\Sigma X = 15.00$
0.5	0.498	$(\Sigma X)^2 = 225.00$
0.5	0.521	$\bar{X} = 1.00$
0.5	0.511	$\Sigma XY = 22.4575$
1.0	0.980	$\Sigma X \Sigma Y = 225.8400$
1.0	1.014	$\Sigma Y^2 = 22.419366$
1.0	1.002	$\Sigma Y = 15.0560$
1.5	1.482	$(\Sigma Y)^2 = 226.683136$
1.5	1.498	$\bar{Y} = 1.003733$
1.5	1.491	$Sxx = 7.500$
2.0	1.972	$Syy = 7.307157$
2.0	2.025	$Sxy = 7.401500$
2.0	1.998	Slope (eqn. (2)), $M = 0.987$
sample reading		Intercept (eqn. (3)), $I = 0.0169$
$Y' = 0.770$		Standard error regression (eqn. (4))
		$Sr = 0.0148$
		Sample concentration (eqn. (5)), $X' = 0.763$
		95% confidence level for one observation (eqn. (6))
		$C.L. = \pm 0.0337$
		$\% \ C.L. = \frac{(0.0337)(100)}{0.763} = \pm 4.42\%$

Table 3. Example of Data and Calculated Values for Replicate Readings Upon the Same Sample Using Calibration Line Data Given in Table 2

Independent Sample readings, Y'	Corresponding Concentration eqn. (5), X'
0.770	0.763
0.778	0.771
0.761	0.754
0.765	0.758
0.773	0.766
0.777	0.770
0.772	0.765
0.768	0.761
0.773	0.766
0.763	0.756
$\bar{Y}' = 0.770$,	$\bar{X}' = 0.763$

Standard deviation, $S.D. = \sqrt{\frac{S'xx}{N-1}} = 5.72 \times 10^{-3}$

Standard error, $S.E. = \frac{S.D.}{\sqrt{10}} = 1.81 \times 10^{-3}$

95% $C.L. = (S.E.)(t) = (1.81 \times 10^{-3})(2.262) = 4.09 \times 10^{-3}$

$\% \ C.L. = \frac{(C.L.)(100)}{\bar{X}'} = \frac{(4.09 \times 10^{-3})(100)}{0.763} = 0.536\%$

95% confidence level for $Q (Q = 10)$ observations eqn. (7), $C.L. = \pm 0.0136$

$\% \ C.L. = \frac{(0.0136)(100)}{0.763} = \pm 1.78\%$

An estimate of the scatter in fitting the straight line, referred to as the standard error of regression is obtained by subtracting the fitted regression line, $\hat{Y}$, from each data point, Y, summing the squared deviations, dividing by the degrees of freedom ($N - 2$) and extracting the square root. Thus, the standard error of regression, Sr, is calculated by

$$Sr = \sqrt{\frac{\Sigma(Y - \hat{Y})^2}{N - 2}} = \sqrt{\frac{Syy - \frac{(Sxy)^2}{Sxx}}{N - 2}} \tag{4}$$

Manipulation of eqn. (1) allows the calculation of the amount of unknown in the sample, in terms of concentration units, to be made. After establishment of the calibration line, a sample instrument reading, Y', will have a corresponding concentration

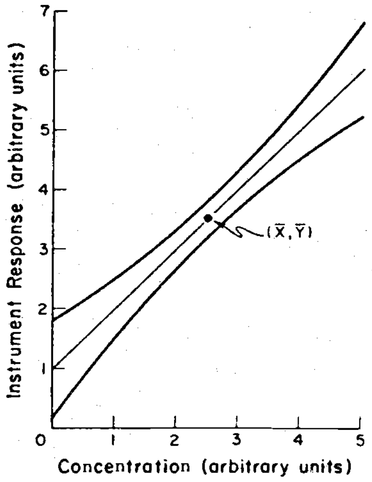
$$X' = \frac{Y' - I}{M} \tag{5}$$

An error term, expressed as a confidence level for this value X' is approximated *(3, 4, 6)* by

$$C.L. = \frac{t \cdot Sr}{M} \sqrt{1 + \frac{1}{N} + \frac{(X' - \bar{X})^2}{Sxx}} \tag{6}$$

Table 4. Comparison of the 95% Confidence Levels (Expressed As a Percent of the Mean) Obtained by Different Procedures

Replication within sample ($N = 10$)	Using calibration line $Q = 1$	$Q = 10$
0.536%	4.42%	1.78%



Calibration Line with confidence intervals.

where t is the Student t value for $N - 2$ degrees of freedom and the desired confidence level. The other values are as previously defined. In some instances a mean value composed of more than one reading of the sample may be obtained, and in this case the value 1 under the square root sign becomes $1/Q$, where Q represents the actual number of independent determinations performed on that particular sample *(6, 7)*. An estimated confidence level for a mean of Q independent determinations is thus approximated by

$$C.L. = \frac{t \cdot Sr}{M} \sqrt{\frac{1}{Q} + \frac{1}{N} + \frac{(\bar{X}' - \bar{X})^2}{Sxx}} \tag{7}$$

where $\bar{X}'$ is the mean concentration corresponding to the mean instrument readings ($\bar{Y}'$) and evaluated similar to eqn. (5). Thus, a greater degree of confidence (resulting in a narrower confidence interval) can be placed on a mean of Q determinations than for a single observation.

Tables 2 and 3 provide examples of the calculations for estimating the confidence interval for a single sample observation and for a mean of 10 ($Q = 10$) independent measurements on the same sample. Table 4 compares these resulting confidence intervals (expressed as a percent of the mean). As indicated, errors other than those associated between sample readings must be considered: the calibration line scatter must be included.

When there is a strong *a priori* knowledge that the relationship between the instrument response and concentration is linear, and the required regression assumptions fulfilled, the confidence interval can be minimized by maximizing $\Sigma(X - \bar{X})^2$; since the X values are assumed to be fixed, the choice of the X values is often available *(3)*. The value of $\Sigma(X - \bar{X})^2$ is maximized when the X observations are divided equally at the two extreme points of the desired range of the X values. Also, the narrowest confidence interval will result if the sample has a concentration near the means ($\bar{X}$, $\bar{Y}$) of the calibration line data.

Examination of the figure indicates the increase in the confidence interval as values move along the calibration line away from the mean values, $\bar{X}$ and $\bar{Y}$. Wider confidence intervals result when extrapolating beyond the range of the data. For this reason the rule of thumb "it is safer to interpolate than to extrapolate" applies to calibration line evaluations.

Calibration Lines Using Average Values

Occasionally an excessive amount of calibration data may be obtained and the analyst decides to use average instrument values to construct the calibration line rather than using total data. This may facilitate the handling of the data but results in a loss in the degrees of freedom. It is the averages of the data points which then establish the scatter about the calibration line rather than the total individual points. Since the number of averages is less than the total number of data points this will result in the selection of a larger Student *t* value and increases the confidence interval.

In using averages the appropriate weighting factors must be applied to properly establish the calibration line. These weighting factors are generally the number of data points used in computing the average for the particular instrumental readings *(2)*.

Table 5 gives numerical values obtained for a calibration line using the total data available and Table 6 presents computations for the same calibration line data based on using weighted means. A comparison of the resulting data is presented in Table 7. The values obtained using weighted means are essentially those obtained by using the total data (slight discrepancies exist due to rounding of data) with the exception of the confidence interval which depends upon the appropriate Student *t* value.

Table 5. Calibration Line Data With Calculated Values Using Total Data

Concentration X values	Instrument Response Y values	
Arbitrary units		
0	0.019	$N = 21$. $t_{(N-2,\,0.95)} = 2.093$
0	0.024	$\Sigma X^2 = 24.00$
0	0.021	$\Sigma X = 17.00$
0	0.023	$(\Sigma X)^2 = 289.00$
0	0.020	$\bar{X} = 0.809524$
0	0.021	$\Sigma Y^2 = 23.959155$
0.5	0.498	$\Sigma Y = 17.1530$
0.5	0.521	$(\Sigma Y)^2 = 294.225409$
0.5	0.511	$\bar{Y} = 0.816810$
0.5	0.513	$\Sigma XY = 23.97650$
0.5	0.515	$\Sigma X \Sigma Y = 291.601$
1.0	0.980	$S_{xx} = 10.238095$
1.0	1.014	$S_{yy} = 9.948421$
1.0	1.002	$S_{xy} = 10.090738$
1.0	1.005	Slope (eqn. (2)), $M = 0.9856$
1.5	1.498	Intercept (eqn. (3)), $I = 0.0189$
1.5	1.491	Standard error of regression (eqn. (4)),
1.5	1.482	$S_r = 0.0124$
2.0	1.972	Sample concentration (eqn. (5)), $X' = 0.833$
2.0	2.025	95% confidence level (eqn. (6)), $C.L. =$
2.0	1.998	± 0.0269
sample reading, $Y' = 0.840$		

Calibration Line Through the Origin

Occasionally the analyst may decide that the calibration line should pass through the origin of his axes; i.e., with a zero intercept. The equation for a line through the origin is given by

$$Y = M \cdot X \tag{8}$$

where the slope

$$M = \frac{\Sigma XY}{\Sigma X^2} \tag{9}$$

and an estimate of the variability, S, about the line is given by

$$S = \sqrt{\frac{\Sigma Y^2 - \dfrac{(\Sigma XY)^2}{\Sigma X^2}}{N - 1}} \tag{10}$$

The concentration of the sample can be obtained by manipulation of eqn. (8). For a sample reading, Y', the corresponding concentration, X', is

$$X' = \frac{Y'}{M} \tag{11}$$

and a confidence level estimate for X' is given approximately by *(3)*

$$C.L. = \frac{t \cdot S}{M} \sqrt{1 + \frac{(X')^2}{\Sigma X^2}} \tag{12}$$

where t is selected for the appropriate confidence level with $N - 1$ degrees of freedom.

The construction of this line may not actually pass through the mass center of the data points as it is based upon the deviations of the data from the origin *(2)*. Careful examination of the data should be made before adopting this model. In many instrumental analyses, signal contributions arising from blanks or instrumental noise

Table 7. Summary of Data Obtained From the Two Calibration Lines

	From total data	From weighted means
Sample concentration, X'	0.833	0.833
Degrees of freedom	19	3
Student t $(N - 2, 0.95)$	2.093	3.182
Standard error of regression	0.0124	0.0124
95% confidence level	0.0269	0.0438

Table 6. Mean Calibration Line Data and Calculations

Mean Concentration $\bar{X}$	Mean Instrument Response $\bar{Y}$	W	$W\bar{X}$	$W\bar{X}^2$	$W\bar{Y}$	$W\bar{Y}^2$	$W\bar{X}\bar{Y}$
0	0.021333	6	0	0	0.127998	0.002731	0
0.5	0.511600	5	2.5	1.25	2.558000	1.308673	1.2790
1.0	1.000250	4	4.0	4.0	4.001000	4.002000	4.0010
1.5	1.490333	3	4.5	6.75	4.470999	6.663277	6.706498
2.0	1.998333	3	6.0	12.00	5.994999	11.980004	11.989998
		$\Sigma = 21$	17.00	24.00	17.152996	23.956685	23.976496

Number of means, $N = 5$; $t_{(N-2,\,0.95)} = 3.182$
$\overline{X_w} = 0.809524$
$\overline{Y_w} = 0.816809$
$S_{wxx} = 10.238095$
$S_{wyy} = 9.945958$
$S_{wxy} = 10.090737$
Slope, $M = S_{wxy}/S_{wxx} = 0.9856$
Intercept, $I = \overline{Y_w} - M \cdot \overline{X_w} = 0.0189$

Standard error of regression, $S_r = \sqrt{\dfrac{S_{wyy} - \dfrac{(S_{wxy})^2}{S_{wxx}}}{N - 2}} = 0.0124$

Sample concentration ($Y' = 0.840$), $X' = 0.833$

95% confidence level, $C.L. = \dfrac{t \cdot S_r}{M}\sqrt{1 + \dfrac{1}{N} + \dfrac{(X' - \overline{X_w})^2}{S_{wxx}}} = \pm 0.0438$

(8) produce a background signal in the absence of the analyte which will result in a non-zero intercept for the calibration line.

Acknowledgment

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ISOTOPIC DILUTION OF ZINC-65 IN OREGON COASTAL WATERS

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Abstract—Specific activity of zinc-65 (^{65}Zn/Zn) observed in mussels, *Mytilus californianus*, collected along the Oregon coast is compared to values predicted, based on a simple model for isotopic dilution of ^{65}Zn in the Columbia River. The observed values are very similar to those computed from the model. Our analysis provides no evidence that chemical speciation of zinc in coastal waters affects the specific activity of mussels.

INTRODUCTION

THE "SPECIFIC ACTIVITY APPROACH" (NAS–NRC, 1962) was originally proposed as a simple guideline for determining acceptable rates of discharge of radionuclides into the environment. The approach assumes that ecosystems do not function as isotope separators and that specific activities (ratios of radioisotopes to total element) cannot be increased by ecological processes. In fact, specific activities are lowered in time by the natural processes of radioactive decay and isotopic dilution during mixing into the receptor medium. It was recognized that certain elements are present in more than one physical/chemical form and that isotopic dilution might not be instantaneous. Were "... such elements as ^{3}H, C, S, V, Fe, Co, Cu and Zn ... introduced into the environment in stable organic complexes," they might not be diluted by the common and abundant form of the stable element (NAS–NRC, 1962:23). Discrimination among chemical forms is expected in ecological processes. For purposes of release guidance an arbitrary safety factor of 10 was introduced in the estimation of permissible release rates for the above elements.

Zinc, in particular, has been identified as an element for which isotopic dilution may be slow in marine ecosystems. BERNHARD and ZATTERA (1969) found that uptake rates and steady-state concentration factors for ionic zinc and organic complexes of zinc were not the same in a marine alga. PIRO *et al.* (1973) reported that zinc occurs in seawater in at least three chemical forms and that added ionic ^{65}Zn did not equilibrate rapidly with the other forms. CROSS *et al.* (1971) found that phytoplankton cultures did not attain ^{65}Zn/Zn isotopic equilibrium after even 12 months. SMALL *et al.* (1974) found that a chemical form of ^{65}Zn, similar to that identified by PIRO *et al.* (1973) as "chelated," was released by zooplankton which had been labelled with the isotope. In the natural environment fewer clear-cut examples are available. ROBERTSON (1971) found higher ^{65}Zn specific activity in fishes than in the water from which they were collected. LARSEN *et al.* (1973) reported that ^{65}Zn specific activities in mussels along the Oregon coast seemed surprisingly high. It seems appropriate to consider isotopic dilution more carefully in order to determine if selective assimilation and retention of ^{65}Zn in organisms is occurring.

The Columbia River has been identified as the primary source of ^{65}Zn in the coastal system off Oregon and Washington (OSTERBERG *et al.*, 1964; CUTSHALL *et al.*, 1973) (Fig. 1). The dynamics of the Columbia River plume have been discussed by OSTERBERG *et al.* (1964), CONOMOS *et al.* (1972) and BARNES *et al.* (1972). In general, the plume flows northward along the Washington shoreline during winter months and southwesterly off Oregon during the summer. Short term variations in plume properties have been discussed by DUXBURY (1972). Seasonal variations in Columbia River discharge and seasonality of biological processes have been shown by RENFRO *et al.* (1972) to produce cyclic variations of ^{65}Zn specific

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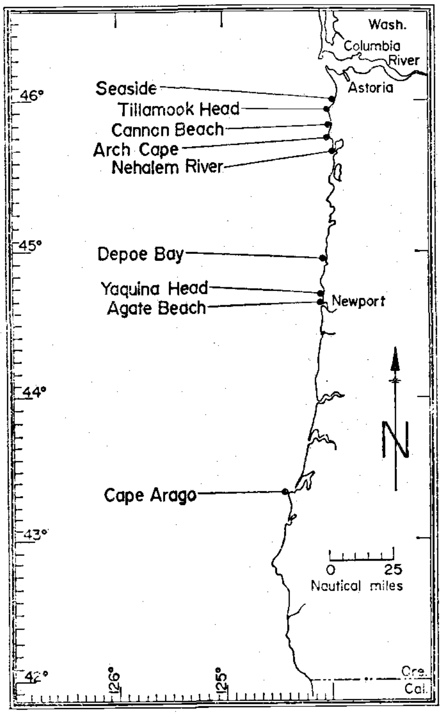

FIG. 1. Map of Oregon coastline showing collection stations and Columbia River mouth.

activities in Columbia River estuarine biota. Annual specific activity maxima are roughly 1.5 times annual minima. It is clear from these studies that instantaneous conditions are inevitably transitory. Our intention here is to employ sufficient quantities of input data to average out such variations. Alternatively, it would be possible to model each of the known sources of variation.

ISOTOPIC DILUTION IN ESTUARINE MIXING

If all properties involved are conserved during mixing the specific activity of mixed estuarine or plume waters is given by:

$$A_m = \frac{A_r}{1 + \dfrac{Z_s}{Z_r} \cdot \left(\dfrac{1}{\dfrac{S_s}{S_m} - 1} \right)}, \qquad (1)$$

where A is specific activity, Z is zinc concentration and S is salinity and the subscripts r, m and s denote river water, mixed water and seawater, respectively. It has been assumed that seawater contains no radiozinc. This simple, conservative model will predict a higher specific activity than observed values because it neglects radioactive decay and changes in salinity owing to rainfall or non-radioactive riverwater. Only an unknown additional source of radioactive zinc or chemical fractionation of radioactive and stable zinc could cause the observed specific activity to be higher than predicted. The model requires five input data which are selected by averaging reported values as discussed below.

The concentration of zinc in Columbia River water was reported by SILKER (1964) for samples collected within the Hanford reservation during 1962. For 21 analyses the average zinc concentration was 18.2 with a standard deviation of 9.9 μg/kg (Table 1). KOPP and KRONER (*ca.* 1968) reported zinc concentrations from several Columbia River stations over a 5-yr time interval (1962–1967). For 68 results they found a range of 4–239 μg/kg and a mean of 29μg/kg. For purposes of calculation the average of the above values, weighted according to number of analyses reported, is used here. That value is 26.5 μg/kg. For the zinc concentration in seawater we select 5.75 μg/kg, the mean of 14 analyses in surface and 10 m waters collected off Newport, Oregon, in the summer and fall of 1968 (WAGNER, 1974). ZIRINO and HEALY (1971) reported 4.7 μg/kg in a sample from the same region. The ratio Z_s/Z_r is then $5.75/26.5 = 0.217$.

The specific activity in the Columbia River, A_r, is taken to be the average of ^{65}Zn specific activities reported by RENFRO *et al.* (1972) for juvenile starry flounders (*Platichthys stellatus*) of less than 100 mm standard length over the time interval of September 1964 through July 1968. The flounders were taken from Harrington Point, the upper reach of salt water incursion in the Columbia River estuary, and are considered to represent an essentially pure river source of zinc and ^{65}Zn. The specific activity of small flounders is probably lower than river water itself, owing to isotopic dilution within the organisms. Biological isotopic dilution would also cause specific activities in mussels to be lower than calculated. The degree of this error

Table 1. Calculation of input parameters

a. Parameters independent of station

Parameter	Number of Observations	Range	Mean	Std. Dev.	Value Used	Reference
Zinc in Columbia River water μg/kg	21	5.2-35.4	18.2	9.9	26.5	(1)
Zinc in Columbia River water μg/kg	68	4.0-239	29.0			(2)
Zinc in Seawater μg/kg	14	2.6-9.8	5.75	2.32	5.75	(3)
^{65}Zn Specific Activity in fish nCi/g	27	400-2380	1310	440	1310	(4)
^{65}Zn in Columbia River water pCi/l	54	1.5-86	34.4			(5)
Salinity of Seawater o/oo		32.6-33.4			33.0	(6)

b. Salinity values at shore stations

Station	Distance from Columbia River Mouth, km	Number of Analyses	Average of monthly mean daily salinity o/oo values	Reference
Seaside Aquarium	27	745	29.06	(7)
Arch Cape	49	382	30.94	(7)
Depoe Bay Aquarium	160	613	31.70	(7)
Cape Arago	335	816	32.46	(7)

[1] Silker, 1964
[2] Kopp and Kroner, 1968?
[3] Wagner, 1974
[4] Renfro et al., 1972
[5] Toombs and Culter, 1968; Toombs, 1973
[6] Conomos and Gross, 1968
[7] Bourke et al., 1971

depends upon the biological turnover rate of zinc in the organism (CUTSHALL, 1974). Were the turnover times for flounders and mussels identical, then the errors would exactly compensate. For 27 analyses the average specific activity was 1310 nCi/g with a standard deviation of 440 nCi/g. The specific activity can be estimated in another way. Averaging of ^{65}Zn concentrations in Columbia River water from the years 1963–1968 (TOOMBS and CULTER, 1968; TOOMBS, 1973) yields 34.4 pCi/l for 54 analyses. If this is divided by the average zinc concentration, a specific activity of 1300 nCi/g is calculated. The agreement is gratifying.

Finally, the value used for the salinity of seawater is the midpoint of the range reported by CONOMOS and GROSS (1968) for the seawater component of the Columbia River mixing region (32.6–33.4‰). The values selected as input parameters for our calculation are summarized in Table 1a.

The remaining parameter required is the salinity of the mixture of seawater and river water. Oregon coastal salinity data have been reported by BOURKE *et al.* (1971). They tabulated monthly averages of mean daily salinities for several stations (Fig. 1). Their monthly values are averaged and given in Table 1b.

RESULTS AND DISCUSSION

Using input data from Table 1 the isotope dilution equation, equation (1) was used to compute ^{65}Zn specific activities expected at stations where salinity data were available. The results are shown as a function of distance from the river mouth in Fig. 2. When a least-squares regression line is fitted through the points, the following expression for natural logarithm of specific activity results:

$$\ln (A_m) = (6.16 \pm 0.15) - (0.00495 \pm 0.00080)X, \quad (2)$$

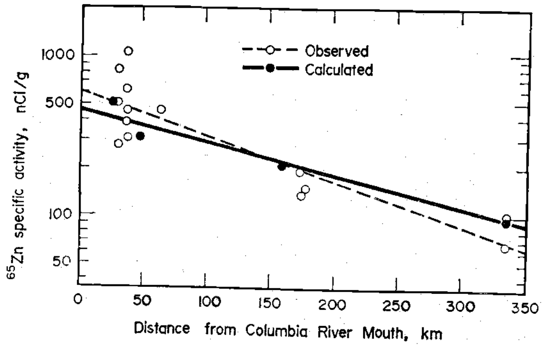

FIG. 2. Specific activities of zinc-65 calculated from isotope dilution equation and observed in mussels.

where X is distance in km. (Errors indicated are standard errors.) There are two degrees of freedom. The line corresponding to this equation is drawn through the points.

Observed ^{65}Zn specific activities in mussels (*Mytilus californianus*) are tabulated in Table 2 and plotted in Fig. 2. Least-squares regression analysis of observed values yields:

$$\ln (A_m) = (6.43 \pm 0.15) - (0.00655 \pm 0.00101)X. \quad (3)$$

This line has also been drawn in Fig. 2. There are 12 degrees of freedom.

The slopes for the lines fitted to observed values and to calculated values do not differ within the errors of the regression analyses; the difference is 0.00162 ± 0.00182. Provided that the data are linear the error indicated is valid for significance testing. However, neither the line fitted to the observed values nor the one fitted to the calculated values produces a reasonable intercept value. [The intercept should be 7.18 = ln (1310).] This failure presumably indicates that the logarithm of specific activity versus distance is not linear over the entire range of values. Values near the river mouth fall above the line. We have no reason to expect the data to be linear (or log-linear) with distance; the logarithmic plot was used for simplicity.

Within the available data it does not appear

Table 2. Zinc-65 specific activities of Mytilus californianus

Location	Distance from River mouth km	Specific Activity nCi/g	Date	Reference
		Calculated Values		
Seaside Aquarium	27	504		
Arch Cape	49	308		
Depoe Bay Aquarium	160	208		
Cape Arago	335	93		
		Observed Values		
Tillamook Head	32	837	July, 1967	(1)
Tillamook Head	32	506	March, 1964	(5)
Tillamook Head	32	278	August, 1964	(2)
Cannon Beach	38	308	July, 1969	(3)
Cannon Beach	38	1080	July, 1965	(4)
Cannon Beach	38	620	Sept., 1965	(4)
Cannon Beach	38	380	Jan., 1966	(4)
Cannon Beach	38	470	April, 1966	(4)
Nehalem River Jetty	65	466	July, 1969	(3)
Yaquina Head	175	192	July, 1967	(1)
Yaquina Head	175	135	March, 1964	(5)
Agate Beach	176	147	August, 1969	(3)
Cape Arago	335	97	June, 1967	(1)
Cape Arago	335	67	March, 1964	(5)

[1] Larsen, 1971

[2] Alexander and Rowland, 1966.

[3] Toombs, 1973.

[4] Seymour, (1972) kindly provided ^{65}Zn data and museum samples. Zn was measured at CSU.

[5] Larsen *et al.*, 1973.

necessary to reject the isotopic dilution model. That is, the specific activity of ^{65}Zn in mussels along the Oregon coast appears to follow the isotopic dilution pattern expected from the model. The seemingly high specific activity of Yaquina Head mussels noted by Larsen *et al.* (1973) can be attributed to the greater zinc concentration in Columbia River water than in seawater. Our analysis does not confirm or conflict with anomalous point observations of deviations from the isotopic dilution model (Robertson, 1971), nor does it refute the laboratory results (Bernhardt and Zattera, 1969; Cross *et al.*, 1971). It does, however, show that in a very large system, actually contaminated over a long time period, the "Specific Activity Approach" (NAS–NRC 1962) works reasonably well.

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RLO-2227-T12-51

DESCRIPTION AND BIOLOGY OF A NEW SPECIES OF PELAGIC PENAEID SHRIMP, *BENTHEOGENNEMA BURKENROADI*, FROM THE NORTHEASTERN PACIFIC[1]

EARL E. KRYGIER[2] AND ROBERT A. WASMER[3]

ABSTRACT

The new species of pelagic penaeid shrimp lacks the richly plumose arthrobranch described for the genus and has a single pair of terminal spines on the telson. It is found mainly in transitional water of the North Pacific between 500 and 1,000 m by day and 150 and 1,000 m at night. Examination of testes and ovaries, and the structures of the petasma and thelycum, indicates a 4-5 mo spawning season and an equal male to female sex ratio. Generation time was estimated to be 2 yr.

This paper describes the systematics and biology of a new species of pelagic penaeid shrimp of the genus *Bentheogennema*. Since 1961, studies of the fauna and ecology of the mesopelagic waters off the coast of Oregon have been conducted by members of the School of Oceanography, Oregon State University. Several unusual species of macrurous decapod Crustacea have been obtained. The discovery and identification of this new species of *Bentheogennema* was by Carl Forss, who entrusted his material to the authors. Subsequent sampling with mid-water trawls has provided detailed information on the distribution and biologyof this shrimp, as well as abundant material for taxonomic description.

METHODS AND MATERIALS

Material for the zoogeographic distribution was collected in Isaacs-Kidd Mid-water Trawls (IKMT) from the research vessels *Yaquina*, *Endeavor*, *John R. Manning*, and *Hugh M. Smith* in the northeastern Pacific, normally within 320 m of the surface (Wasmer 1972). Information on vertical distribution, reproductive biology, and growth of this species was obtained from samples taken on five cruises aboard RV *Yaquina* at a single sampling station 65 nautical miles (120 km) off the central Oregon coast (NH 65–lat. 44°35′N, long. 125°25′W) in 1972-73. Samples at this station were taken both day and night, using an 8-foot IKMT with a five net opening-closing cod end section similar to the one described by Pearcy and Mesecar (1971).

All samples were preserved at sea in 10% buffered Formalin.[4] The samples were later sorted, identified, sexed when possible, and measured. Carapace length (measured from the postorbital margin to the median posterior edge of the carapace) was used as an indication of size. All figures were drawn with the aid of a camera lucida.

In males, sexual maturity was based on three characteristics: 1) petasmata joined; 2) well-developed accessory lobe on anterior surface of the petasma; 3) and dilated vas deferens with large terminal ampoule (indicative of developed spermatophore) at the base of the fifth pereiopod. The combined characteristics of fully developed thelycum and the posterior lateral lobe of the ovary swollen with eggs at the base of the fifth pereiopod were used as signs of sexual maturity in females. Estimates of growth are presented from analysis of length-frequency data.

Section Penaeidea
Family Penaeidae Bate
Subfamily Aristaeinae Alcock
Series Benthesicymae Bouvier
Bentheogennema burkenroadi n. sp.

Types.–Holotype (USNM 150835), male, carapace length (c.l.) 18 mm, from Station lat.

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[4] Reference to trade names does not imply endorsement by the National Marine Fisheries Service, NOAA.

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51°26′N and long. 138°28′W, Mid-water Trawl No. 857 (MT 857); Allotype (USNM 150836), female, 14.5 mm c.l., station Newport Hydrographic Line 65 nautical miles (120 km) offshore (NH 65), MT 2130; Paratypes: 1 male (USNM 150837), 15 mm c.l., from NH 265, MT604; 2 males (USNM 150838), 8 and 6.5 mm c.l., NH 65, MT 2088; 1 male (USNM 150839), 14 mm c.l., NH 65, MT 2131; 1 male (USNM 150840), 13.5 mm c.l., NH 65, MT 2130; 4 females (USNM 150841), 14.5, 14, 10, 18 mm c.l., NH 65, MT 2130; 4 males (USNM 150842), 12.5 13.0, 13.2, 17.5 mm c.l., NH 65, MT 2130; 1 male (USNM 150843), 9.0 mm c.l., NH 50, MT 570; 1 male (USNM 150844), 9.5 mm c.l., lat. 40°28′, long. 133°46′, MT 613; 3 females (USNM 150845), 13.3, 14.0, 15 mm c.l., NH 65, MT 2121 Net #5; 1 female (USNM 150846), 20.0 mm c.l., NH 65, MT 2133 Net #1; 2 females (USNM 150847), 7 and 10 mm c.l., NH 65, MT 2070 Net #5; 2 males, 1 female (BMNH 1975:10), 14.2, 16.4, 13.2 mm c.l., NH 65, MT 2175 Net #5; 1 female (BMNH 1975:10), 15.7 mm c.l., NH 65, MT 2178 Net #4; 1 female (BMNH 1975:10), 12.6 mm c.l., NH 65, MT 2302 Net #4; 1 male (BMNH 1975:10), 11.5 mm c.l., NH 65, MT 2301 Net #1. Other, nonparatype, material deposited at Los Angeles County Museum; Fisheries Research Board of Canada Biological Station, Nanaimo, British Columbia; and School of Oceanography, Oregon State University, Corvallis, Oreg.

Diagnosis.—Benthesicymae with podobranch on second maxilliped to third pereiopod inclusive; first maxilliped with single rudimentary arthrobranch; only sixth abdominal somite with middorsal carina; telson distally truncate, usually with single pair of mobile terminal lateral spinules. Accessory lobe of petasma characterized by large upturned terminal hook. Plate of thelycum on sixth thoracic sternite triangular and elevated, projecting ventrally in strong ridge; plate on eighth thoracic sternite pentagonal with anteriormost angle concave and anterolateral margins bearing spines.

Description.—Rostrum extending to level of eye tubercle, well elevated above middorsal carina of carapace (Figure 1). Margin between rostral tip (apex) and dorsal spine with usual setal fringe (although broken in type). Middorsal carina of carapace bearing minute tubercle posterior to

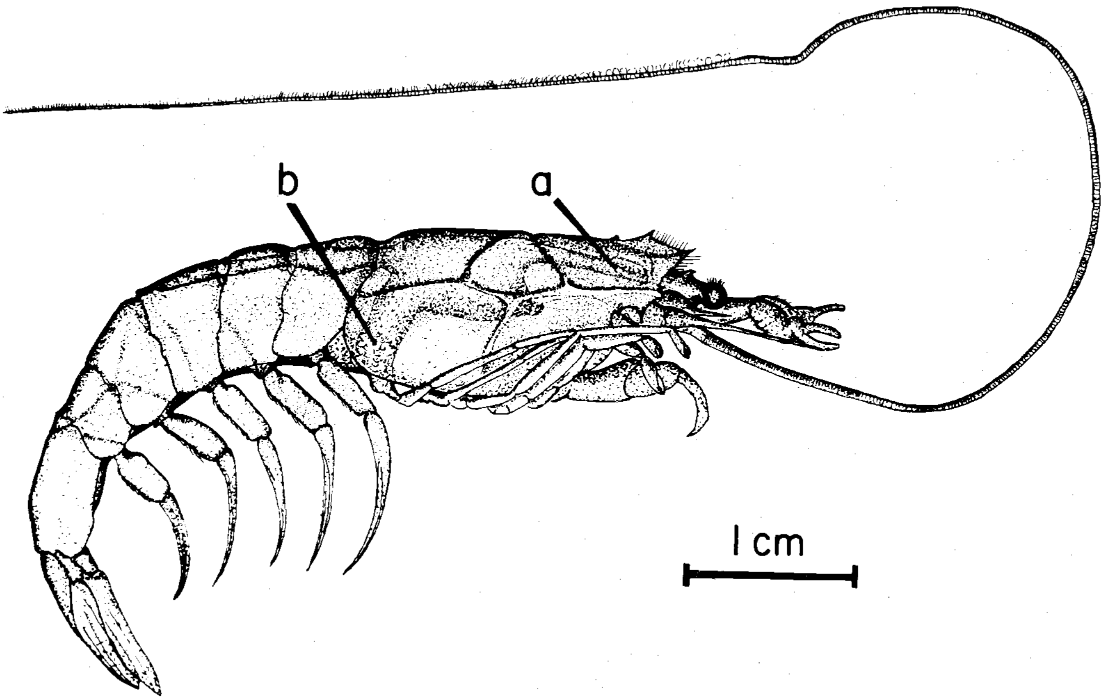

FIGURE 1.—Mature female *Bentheogennema burkenroadi* n. sp. illustrating the (a) anterior and (b) posterolateral lobes of the ovary.

dorsal spine; carina absent between well-defined cervical and postercervical sulci and on posteriormost portion of the carapace. Mid-lateral longitudinal carina consisting of strong antennal carina continuous with hepatic and branchial carinae. Branchiostegal spine small and branchiostegal carina distinct; hepatic sulci continuous from branchiostegal spine towards lower margin of carapace; post-hepatic carina orientated dorsoventrally from longitudinal hepatic carina toward inferior margin of carapace. Antennal angle obtuse and infra-antennal angle acute (Figure 2a).

Only sixth abdominal somite with middorsal carina; second through fifth abdominal somites with weak lateral ridges in approximately dorsoventral position extending from mid-lateral to ventrolateral edge of the pleuron. Fourth, fifth, and sixth abdominal somites with prominent, roughly transversal, lateral ridges which together form "half moon" area (Figure 1). Fourth and fifth abdominal somites bearing small mid-lateral tooth on posterior margins.

Antennal flagellum (Figure 1) similar to *Gennadas* (Foxton 1969), having proximal and distal sections divided by short series of annuli forming kink in flagellum; proximal section rigid, bearing scattered short nonplumose setae; distal section bearing paired arched plumose setae with small plumose setae perpendicular to flagellum at irregular intervals between bases of some arched pairs. Second element of antennular peduncle, along dorsal midline, 0.7 ultimate element (Figure 2b). Antennal scale (Figure 2c) little less than 3 times as long as greatest width; distinct spine (outer margin of scale), slightly convex, terminal end free, not extending beyond narrow apex of blade.

Mandible (Figure 2d) with two segmented palp; palp thickly covered with setae on medial and lateral margins, distal element not quite as long as widest portion of basal element. Endopod of first maxilla (Figure 2e) distally narrow, with tip rounded; proximal gnathobasic lacinia (endite of coxa) subequal in width to distal lacinia (endite of basis), both terminating in strong spines among setae fringe. Anterior lobe of proximal lacinia (endite of coxa) of second maxilla (Figure 2f) strongly constricted behind apex, not broader than posterior lobe of distal lacinia (endite of basis); anterior lobe of distal lacinia very broad; endopod distally long and narrow, with two (sometimes three) curved spines at base of apical portion.

Endopod of first maxilliped (Figure 2g) reaching beyond endite of basis but falling short of exopod; endopod of four elements, third less than twice second; fourth extremely minute; first element bearing usual compliment of three curved spines on distomesial margin. Exopod bladelike, without constricted, segmented distal portion. Merus of second maxilliped (Figure 2h), including anterior prolongation, 1.9 times as long as wide; dactylus with single strong apical spine surrounded by medium and small spines back to proximal end of propodus; merus and carpus with numerous spines and setae; podobranch present. Third maxilliped (Figure 2i) reaching to, or beyond, middle of ultimate joint of antennal peduncle; ischium nearly 3 times as long as greatest width; merus usually twice as long as greatest width; carpus slightly longer than propodus; dactylus with long slender terminal spine; podobranch present.

Merus of first pereiopod (Figure 3a) 1.4 times length of carpus and 1.7 ischium; fingers slightly setose. In second pereiopod (Figure 3b), carpus 1.2 times length of propodus; merus 1.2 carpus and 1.5 propodus; chela with heavy tufts of bristles. Merus and carpus of third pereiopod (Figure 3c) of equal length, each twice ischium; fingers of chela similar to those of second pereiopod. Carpus and propodus of fourth pereiopod nearly equal, each approximating two-thirds of merus which is 2.4 times ischium. Propodus of fifth pereiopod subequal to carpus which is subequal to merus; ischium slightly more than one-third of merus.

Outer scale of appendix masculina (Figure 3d) longer than inner; proximal half of lateral margin expanding slightly then tapering toward base. Inner scale broadly rounded distally; spines on distomesial margin (few to many) long and thin, spines on distal margin smaller, stronger, and of uniform length.

Telson with single pair of mobile terminal-lateral spines (Figure 3e) fringed with setae on terminal and distal two-thirds of lateral margins (of the large number of specimens inspected, only two mature males had any indication of more than one pair of mobile spines (USNM 150839, 150840), each with two pair of mobile spines on terminal edge of telson). No mobile nonterminal-lateral spines present on telson. Lateral margins of lateral rami of uropods (Figure 3f) bearing spine at 0.78 total length. Mesial rami about 0.73 lateral rami.

Each half of petasma (Figure 3g, h), distally divided into three lobes (external, median, and in-

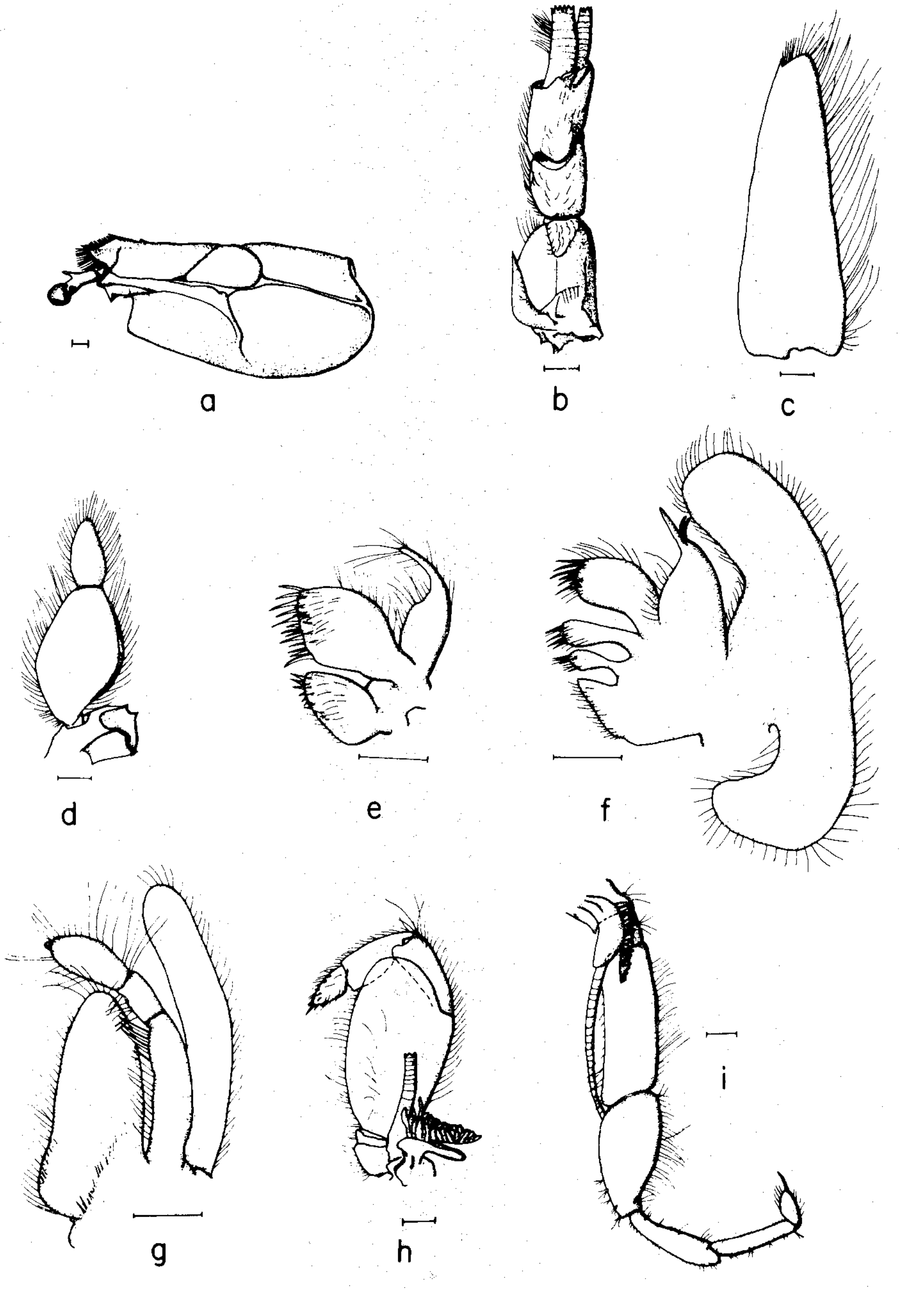

FIGURE 2.—*Bentheogennema burkenroadi* n. sp. (Holotype, male 18 mm carapace length) a, carapace; b, antennular peduncle; c, antennal scale; d, mandible; e, first maxilla; f, second maxilla; g, first maxilliped; h, second maxilliped; i, third maxilliped. Scale equals 1 mm.

ternal (Balss 1927) which are equivalent to Burkenroad's (1936) distoventral, distolateral, and distomedian lobes). External lobe bipartite; lateral part elongate projection with minute terminal teethlike protuberances distally; mesial part curving inward with apex directed toward median lobe. Median lobe broadly rounded; subdistally, accessory lobe on anterior face of petasma, characterized by large upturned terminal hook (Figure 3g) with free margin attaching to base of

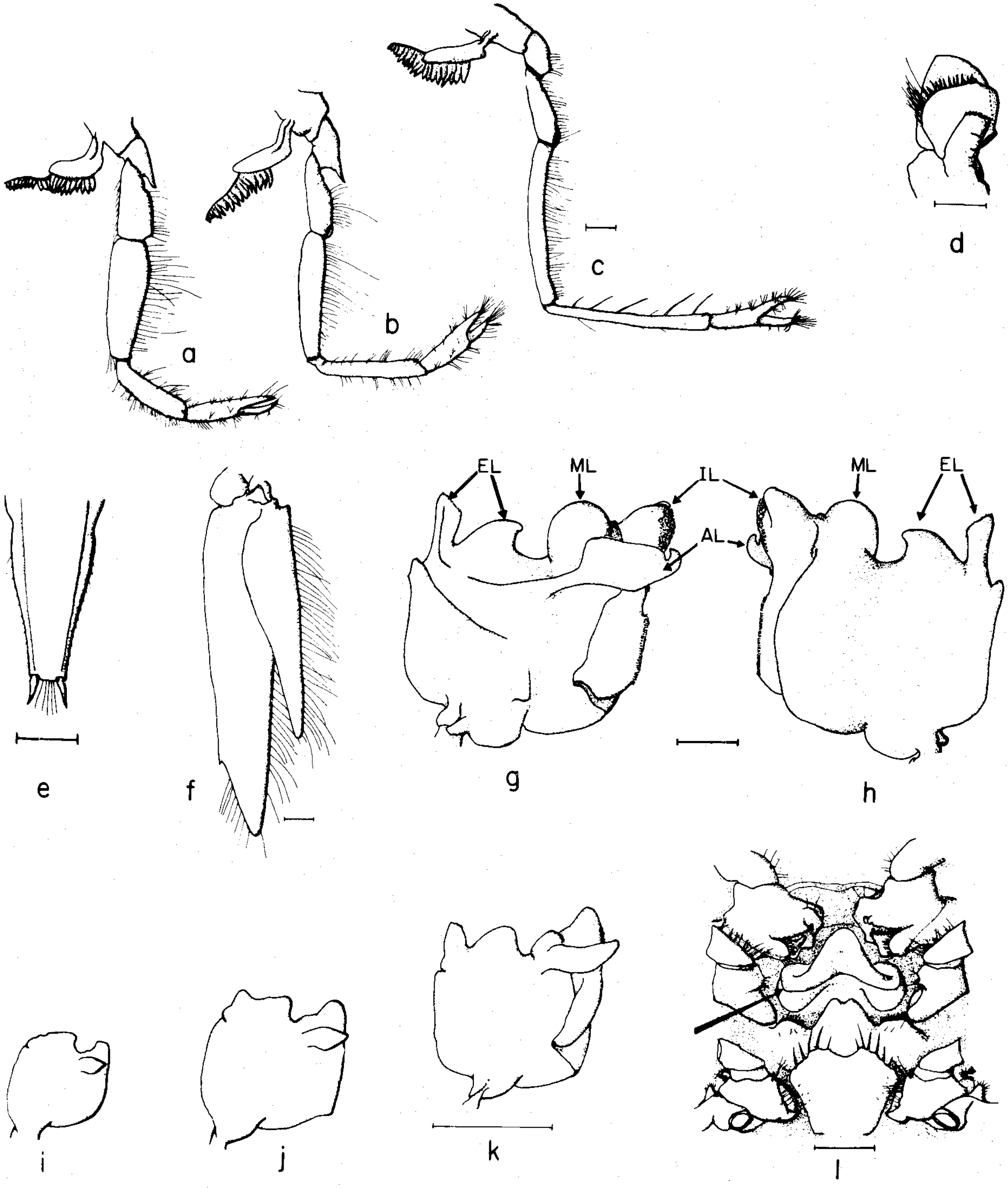

FIGURE 3.–*Bentheogennema burkenroadi* n. sp. (Holotype, male 18 mm carapace length) a, first pereiopod; b, second pereiopod; c, third pereiopod; d, appendix masculina; e, distal half of telson; f, uropod; g, anterior view of petasma (EL = external lobe, ML = median lobe, IL = internal lobe, AL = accessory lobe); h, posterior view of petasma; i, anterior view of petasma from young male (8.0 mm c.l.); j, anterior view of petasma from young male (9.0 mm c.l.); k, anterior view of petasma from young male (9.5 mm c.l.); l, thelycum of female (17 mm c.l.), arrow pointing to right sperm receptacle. Scale equals 1 mm.

median lobe, attachment area distinguishable to level of elongate projection of external lobe. Internal lobe undivided, bearing rigid hooks continuous with row of cincinnuni, holding two halves of petasma together. The accessory lobe develops early in the juvenile stage (Figure 3i, j, k) and together with characteristic spination of telson and presence of podobranchs behind second maxilliped, young of this species were discernable to a size of 6 mm c.l. (the smallest size captured).

Thelycum (Figure 3l) with plate on eighth thoracic sternite pentagonal, with anteriormost angle concave, anterolateral margin bearing long spines (this plate exhibits greatest variation during growth, being more rectangular in young females, changing to the pentagonal shape of maturity but becoming almost bilobed in very large females). Plate of seventh thoracic sternite bearing three anterior directed projections; lateral pair, shortest, bearing short spines; center projection exhibits varying amount of concavity after maturity such that distolateral margins may appear as raised wings. Elevated plate on sixth thoracic sternite triangular and inverted "V" shaped, with apex pointing anteriorly; apex not reaching anterior limit of sternite. Sperm receptacles located toward lateral edges near bases of inverted V.

Coloration at time of capture varying from deep red over entire body to medium red on cephalothorax and lighter on abdomen. Black pigment fleck on distolateral edge of ocular peduncle just below corneal region (Figure 2a). Other small flecks of purple pigmentation often observed on carpus and propodus of third maxilliped and first and second pereiopods, on carpus of third pereiopod, and on ventral surface of abdominal somites just anterior to lateral edge of base of each pleopod.

Remarks.—Burkenroad (1936) proposed the genus *Bentheogennema* for those species of *Gennadas* Bate which possesses podobranchs on the second maxilliped to third pereiopods inclusive. Other generic characters he included were: arthrobranch of first maxilliped large and richly plumose; exopod of first maxilliped without a constricted, segmented distal portion; dorsal carina on sixth abdominal somite only; telson with truncated apex and more than a single pair of mobile lateral spinules.

As is often the case, the addition of a new species changes the generic formula for that group. The new species is similar to *Gennadas* in the armature of the telson but more closely resembles *Bentheogennema* with podobranchs on the second maxilliped through third pereiopod. We agree with Kemp (1909) and Burkenroad (1936) that the presence of podobranchs, a primitive characteristic, is a more important generic trait than the number of pairs of spines on the telson.

We found that the two species of *Bentheogennema*—*B. borealis* (Rathbun) and *B. burkenroadi* n. sp.—from the Oregon coast lack the large, richly plumose arthrobranch on the first maxilliped that Burkenroad (1936) included as a generic characteristic. Both have small rudimentary arthrobranchs similar to *Gennadas*. We assume that Burkenroad (1936) did not have samples of *B. borealis* but included this arthrobranch structure as a generic characteristic from samples of *B. intermedium* (Bate) and *B. pasithea* (Man).

Although Tirmizi (1959) stated that the endopod of the first maxilliped is five-segmented in *Gennadas* and apparently only four-segmented in *Bentheogennema*, we have found that *Gennadas propinquus* Rathbun off the Oregon coast has a four-segmented endopod. Hence these characters are not reliable to distinguish these two genera.

Bentheogennema burkenroadi can be separated from *B. borealis*, *B. intermedium*, *B. pasithea*, and *B. stephenseni* (Burkenroad) by the armature of the telson, and the structures of the petasma and thelycum. The telson of *B. burkenroadi* typically possesses only a single pair of terminal-lateral spines, whereas the other members of this genus possess two or more pairs of lateral spines: *B. borealis* and *B. stephenseni* two pairs; *B. pasithea* three pairs; *B. intermedium* (as described by Tirmizi 1959) four pairs. The number of spines present on the telson should not be held as an invariable characteristic, there is undoubtedly a small percentage of variation as exemplified by the two males of *B. burkenroadi* (USNM 150839, 150840) which possess two pairs of terminal spines. It is possible that one of the two specimens of *Gennadas calmani* (Kemp) (synonymy: *B. borealis*), which Kemp (1909) illustrated with two pairs of terminal spines is also an example of such variation.

The petasma of this new species is unique and easily distinguishable from that of other members of the genus. The combined structures of the accessory lobe with its mode of attachment, its large size, and its terminal hook (present in mature individuals) and the shape of the bipartate external lobes make identification, even of the juvenile stages (Figure 3g-k), possible.

The thelycum differs from that seen in other species by the pentagonal shape of the plate on the eighth thoracic sternite and the elevated triangular plate on the sixth sternite (Figure 3l).

We have named *B. burkenroadi* after Martin D. Burkenroad, whose work on Crustacea, especially the Penaeidae, is well known.

GEOGRAPHICAL AND VERTICAL DISTRIBUTION

Shrimps were examined from mid-water trawl collections taken over much of the North Pacific (Figure 4). *Bentheogennema burkenroadi* was found only in collections from the northeastern sectors (lat. 52-34°N and east of long. 142°W) (Wasmer 1972). Pearcy and Forss (1966, 1969) observed *B. burkenroadi* off the coast of Oregon, as close as 28 km to the northern end of the coast and occurring ≥92 km off the central and south coast. Wasmer (1972) found the greatest concentration in the Transitional Water Mass (Figure 5), with a few individuals occurring in the Pacific Subarctic and eastern North Pacific Central Water Masses. It is assumed to be a transitional species, although as is the case for many shrimps, it is not totally confined to a single physicochemically definited water mass (Wasmer 1972). Since *B. burkenroadi* is a deep mesopelagic species and most of the available geographical collections were from shallow depths, the known geographic range will undoubtedly be increased by more systematic deep trawls in the eastern Pacific.

This species was captured in opening-closing nets from the surface to 1,000-m depth. It apparently demonstrates a diel vertical migration. The depth distribution is, with few exceptions, below 500 m during the day and below 100 m at night (Table 1). Neither day nor night distributions are confined to a narrow depth stratum but are diffused in concentration over a broad range. The nocturnal migration into the upper waters appears to entail only a small segment of the population with the main concentration remaining at depth. Those migrating above 500 m included both sexes, though the immature shrimp (<11-mm

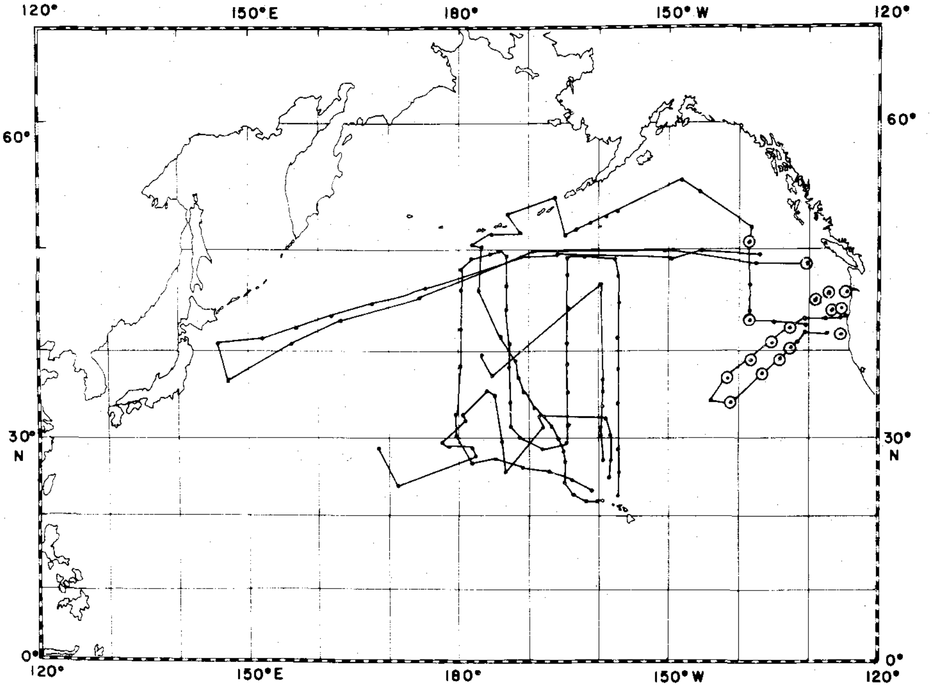

FIGURE 4.—Cruise tracks of the North Pacific from which shrimp were enumerated, indicating mid-water trawl stations (solid dots). *Bentheogennema burkenroadi* n. sp. was collected at stations where dot is encircled.

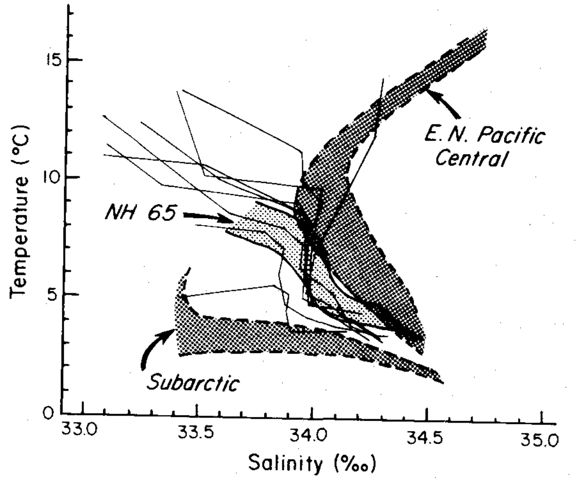

FIGURE 5.–Zoogeographical temperature-salinity (T-S) capture diagram for *Bentheogennema burkenroadi* n. sp. Relevant water masses in darkest bands; medium band in T-S envelope for the station 65 nautical miles off Newport, Oreg. (NH 65); single lines indicate T-S diagram for sampling stations other than NH 65 where this shrimp was captured.

males and <12-mm females) were not observed to migrate as high in the water column as adults. Vinogradov (1968) earlier stated that the intensity of diurnal migration of zooplankters increases with age, and migration may be absent in early stages of development, and our observations concur with this.

More shrimp were caught in nighttime than daytime tows. This may be explained by enhanced visual avoidance of the net during the day (Pearcy and Laurs 1966). However, the lack of obvious differences in size structure between day and night caught shrimp (Figure 6) argues against increased daytime avoidance, as larger more mobile animals should be preferentially sampled at night. Another explanation for the increased nighttime catch, as suggested for *Acanthephyra purpurea* Milne-Edwards and *Gennadas valens* (Smith) by Foxton (1970a, b), is migration up from below our maximum sampling depth of 1,000 m. Such a migration is indicated by the high concentrations between 600 and 1,000 m both day and

TABLE 1.–Seasonal diel vertical distribution to a depth of 1,000 m of mature males (≥11 mm c.l.), mature females (≥12 mm c.l.), and sexually immature male and female *Bentheogennema burkenroadi* at a sampling location 65 nautical miles (120 km) off Newport, Oreg. (lat. 44°35′N–long. 125°30′W).

Time of year	Size group	Depth (m)											
		0-50	50-100	100-150	150-200	200-300	300-400	400-500	500-600	600-700	700-800	800-900	900-1,000
							DAY						
June 1972	Mature males									3	4	2	2
	Mature females									3	2	3	2
	Immatures								1	5	6	2	2
Sept. 1972	Mature males								1	2	6	4	1
	Mature females								2		8	2	4
	Immatures										2	1	1
Nov. 1972	Mature males			1				1	2	2	4	5	
	Mature females				1				2	3	3		1
	Immatures									1	1		1
Mar. 1973	Mature males										[1]3		
	Mature females										[1]1		
	Immatures						1		1		[1]1		
Total number				1	1		1	1	9	19	41	19	14
Total volume in 1,000 m³		103.5	149.9	103.2	94.7	474.7	497.9	310.9	259.6	132.6	182.3	183.3	162.4
Number/1,000 m³ (day)		0	0	0.0097	0.0106	0	0.0020	0.0032	0.0347	0.1433	0.2249	0.1037	0.0862
							NIGHT						
June 1972	Mature males	1				1	3				1		
	Mature females						1	3		1			
	Immatures						2	2					
Sept. 1972	Mature males	[2]2			1	1	1	4	5	[2]1	2	1	1
	Mature females			1	2		5	2	2		3	1	4
	Immatures						3	1	1	1	5		
Nov. 1972	Mature males					1	5	2		7	3		5
	Mature females				[2]1	4	7	2	1	5	1	1	4
	Immatures					1		1		1	3		1
Mar. 1973	Mature males				3	3	1	1	1	1	1		
	Mature females			1	1	4	2	1	1	2			
	Immatures					2			1	1			
Total number		3		2	8	17	30	19	12	20	19	3	15
Total volume in 1,000 m³		111.8	530.6	184.8	292.8	251.0	435.9	288.6	261.6	147.2	170.7	132.3	129.3
Number/1,000 m³ (night)		0.0268	0	0.0108	0.0273	0.0677	0.0688	0.0658	0.0459	0.1359	0.1113	0.0227	0.1160

[1] Twilight 1 h after sunset.
[2] Twilight 1 h before sunrise.

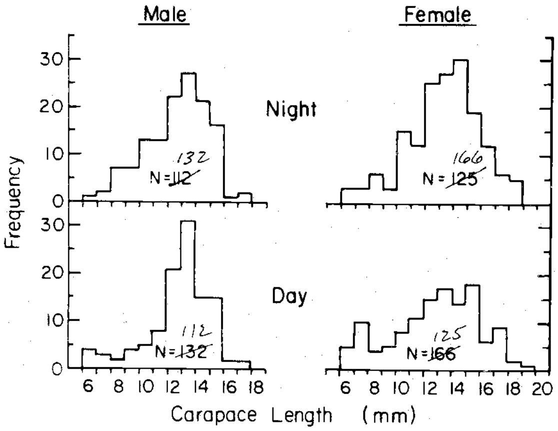

FIGURE 6.–Day and night length-frequency distributions of male and female *Bentheogennema burkenroadi* n. sp.

night with no progressive drop in concentration with increased depth to 1,000 m (Table 1).

The slight upward movement of this species may be related to its morphology. Vinogradov (1968) considers reduced musculature and a thin integument, which we observed in *Bentheogennema burkenroadi*, to be a means of achieving buoyancy. Because of weak swimming musculature, they may swim too slowly to keep pace with the upward movement of the stimulating isolume, resulting in broad day and night distributions (Donaldson 1973).

REPRODUCTION

Since penaeid shrimp do not brood their eggs, a description of the breeding cycle must rely on anatomical changes, especially in the development of the ovary and ova. The female reproductive system consists of a bilaterally symmetrical ovary and paired oviducts internally, and externally of a thelycum. Each half of the mature ovary has an anterior lobe angling from the cervical sulcus and almost reaching the base of the eye, and then folding back along itself (Figure 1a). The anterolateral lobe lies over the hepatopancreas extending approximately one-half the way down the body wall. The posterolateral lobe, of such a mature ovary, will have visible distinct ova, measuring up to 240-288 μm crossectional diameter, and will extend ventrally, making a pouchlike structure at the base of each of the fifth pereiopods (Figure 1b). The posterior lobes extend beneath the dorsal abdominal muscle bands, becoming swollen in the first abdominal segment and then extending on toward the end of the third segment. Females were considered to have reached maturity after attaining a size of 12 mm c.l. and males at a size of 11 mm c.l.

The reproductive cycle, as judged from the sexual condition of the testis and ovary, appears to consist of a 4- to 6-mo spawning season and a 6- to 8-mo resting phase. Based on samples collected in 1972 and 1973, the carapace of females in June is fairly rigid, though the ovaries are not ripe. Some males, from external observation, appear to be ready to release sperm, though most display only partial swelling of the terminal ampoule and vas deferens or lack swelling at all. By fall, females exhibit developing ovaries (two females were in spawning condition), and the carapace is correspondingly rigid. Most males have full, ripe looking testes and dilated terminal ampoules. By the end of November, spawning is in evidence. Most females are mature with readily distinct ova; some mature females have evidently spawned as the thoracic cavity appears empty; the carapace is correspondingly nonrigid, due to the spent ovary which had crowded much of the other organs; others have developing ovaries distended by small diameter ova. All males at this time have ripe testes and dilated terminal ampoules. By February, 50% of all females exhibit signs of spawning activity; the rest have probably spawned because their thoracic cavities appear empty and the carapace nonrigid due to the flaccid ovary. Most males still exhibit ripe testes and enlarged terminal ampoules.

The sex ratio for adult males to females (N = 440), when all tows are included, was: 1:1, 1:1.08, 1:1, 1:1.02, and 1.03:1 for the respective cruises. This approximate 1:1 sex ratio, if it applies to all ages, indicates that there is no selective mortality by sex for this species (Geise 1959).

GROWTH

If spawning occurs from November through February and young (6-7 mm c.l.) enter the population April through June (Figure 7), the intervening egg and larval stages must take 3 to 5 mo. Based on size frequency diagnosis, about 12 additional months are required to reach maturity (11-12 mm c.l.) and another 5 to 6 mo are required before spawning commences Thus the generation time is estimated to be about 2 yr. The largest

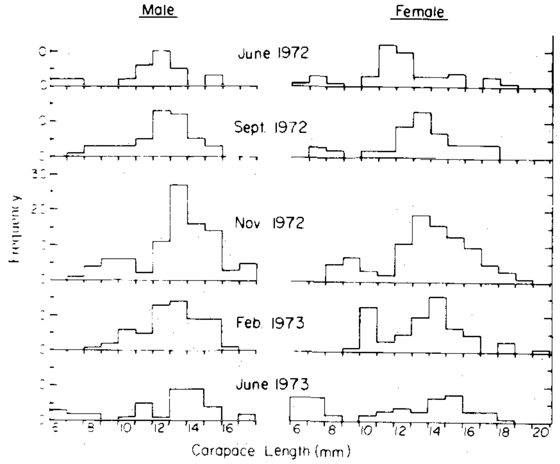

FIGURE 7.–Length-frequency histograms of *Bentheogennema burkenroadi* n. sp., from the five cruises (1972-73) off the Oregon coast.

shrimp captured were a 17-mm c.l. male and a 20-mm c.l. female.

Since the mesh size of the net liner was small enough to retain the young (6-7 mm c.l.) and we assume equal chance of capture of young and adults, then adults apparently live more than a year or two after first spawning because the number of adults captured is greater than the number of immature. In fact, the 12- to 15-mm mode must consist of greater than one age class since by itself it exceeds the juveniles in number. This overlap of age classes at >12 mm c.l. indicates that growth slows after maturity is reached.

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PCB AND THE ACTIVITIES BUDGET OF THE CRAB *HEMIGRAPSUS OREGONENSIS*[1]

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ABSTRACT

In order to understand the adaptive processes of an estuarine organism under stress from a pollutant the behavior of the crab *Hemigrapsus oregonensis* was assessed with and without exposure to polychlorinated biphenyls (PCB's). Activities budgets were developed from observations of the time crabs spent in 20 categories of shelter use, posture, and activity while held in habitat models complete with tides. Females spent more time sheltered and feeding but less time displaying than males. For both sexes certain activities predominated at certain tidal stages.

Multivariate discriminant analysis was used to explore differences in activities budgets between ordinary conditions and stressing conditions when PCB-contaminated sand was present. Discriminant functions containing all 20 activities did not clearly distinguish the budgets of stressed and unstressed crabs. Stepwise discriminant analyses, used to choose the most parsimonious functions, indicated where the budgets differed. For both sexes and different trials, feeding consistently appeared in the multivariate discriminate functions and decreased under PCB exposure.

The discriminant analysis suggested that the consequences one would predict for a pollutant-induced stress are functions of the paradigm under which one observes. In a search to predict the consequences of pollution one should be alert not only to the character of the organism's failures, but also to the nature of its successes.

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INTRODUCTION

Although predictions of the consequences of various courses of action that will alter estuaries are presently being sought, current biological theory is less than adequate to indicate the fate of populations of estuarine organisms exposed to pollutants and other environmental alterations (13, 14, 17). Up to now the scientist has examined what he could with available techniques and often rested with the demonstration of a pollutant effect. Now the need is for explication of the meaning of these demonstrated consequences in light of developing knowledge of ecosystem structure and function. This study is an attempt to explore not only the kinds and magnitudes of any pollutant-induced change in the behavior of the yellow shore crab, *Hemigrapsus oregonensis*, but also changes of ecological import.

Moving beyond conventional toxicity testing is not simply finding a new response criterion, such as behavioral disruption (10) or reproductive failure (7), but abandoning the implicit view that the living organism is little more than a reactive machine driven to responses by stimuli (8, 19). We much prefer to see the animal as a goal-directed general open system (8), primarily active rather than reactive (12), and capable of considerable plasticity in reaching its goals (2). Such a broader view of the animal's capabilities leads to methods of observation and experimental designs in which, hopefully, we avoid making robots of our subjects.

Because one productive way to investigate the ecological meaning of behavior is to ask where and how individual crabs spend their time (20, 21), time budgets for the crab's activities were used to assess the behavior of *Hemigrapsus oregonensis* with and without exposure to polychlorinated biphenyls (PCB's). Our initial hypothesis was that an ecologically meaningful behavioral change induced by PCB exposure would become evident as a shift in the activities budget. As we tried to be alert not only to possible disruption of ordinary behavior, but also to innovative behavior that brings adaption to environmental challenge (8, 14), we expected not quantitative predictions of damaging levels of PCB, but the discerning of patterns of events flowing from the crabs' attempt to adapt to the pollutant.

MATERIALS AND METHODS

In hopes of observing ordinary behavior in the laboratory, we constructed two banks of four 40-liter observation and holding chambers, each of which was designed to simulate the crab's mud-rock habitat. A tide generator of composite design (3, 6, 18) maintained a tidal period of 12.4 hours in the habitat replicas. The tide generators and replicas were oriented to give a four-hour period of air exposure and a 20-cm water height at high tide. The timing of the replica tides matched that in Yaquina Bay, Oregon, where the crabs were collected for this study. Daylight-type flourescent lights lit the replicas on a natural photoperiod, and dark-red safe-lights provided illumination for night observations. Fine sand, crushed and whole oyster shell, and barnacled rocks with algal growth provided natural substrate and shelter. Three crabs of each sex were held in each replica and fed commercial shrimp pellets, small blue mussels, and isopods. Before and during the experiments one gram of crushed shrimp pellet was sprinkled over the substrate at low tide every 37 or 50 hours.

A behavioral repertoire (Table 1) developed from several months of observation defined the activities to be timed for the budgets. The amount of time individual marked crabs spent in 56 behavioral units was measured using an electronic metronome (21) that gives an audible tone every five seconds. We watched a crab and, upon hearing a tone, recorded letter codes for the behavioral units seen during the five-second interval. Summing the number of letter codes in a given observational period gives the time spent in the various behaviors.

Because knowledge of an organism's rhythmicity should lead to more appropriate designs for behavioral observations (5), we followed an observation schedule based on the lunar day (24.8 hours). During each 62 minute lunar hour the six crabs in a habitat replica were observed in a random order, and their activities timed for seven minutes. These hourly observation periods began exactly at low tide and continued for two full tidal cycles. The order in which the replicas were watched was also randomized, but the schedule was such that at the end of eight lunar days each crab in every replica had been observed at every hour of the lunar day. Dividing the number of five-second intervals spent in each behavioral unit by the total number of intervals spent in each behavioral unit by the total number of intervals a crab was observed over the eight lunar day sampling period estimates the proportion of the lunar day the crab spends in each behavior. The observations on the 56 behavioral units were grouped into 20 higher-level categories of shelter use, posture, and activities as indicated in Table 1, and a listing of the time spent by the crabs in each of these 20 categories constitutes the activities budget.

Because *Hemigrapsus oregonensis* readily ingests sand and concentrates PCB from contaminated sand, we thought placing one kilogram of PCB-contaminated sand into the habitat replicas to be a natural mode of exposing the crabs to the pollutant. Eight lunar days' observation gave the time measurements for activities budgets of crabs under control and exposed conditions in two experiments. PCB's (Aroclor® 1260) were sorbed to fine sand by mixing oven-dried sand and an acetone solution of PCB for several days and then evaporating the acetone. In the first experiment the sorption of the PCB to sand varied so that the two highest concentrations could not be distinguished. Consequently the first experiment had two control replicas, two replicas receiving low exposure averaging 30 ppm PCB in the sand, and four replicas receiving moderate exposure averaging 260 ppm. The second experiment, in which the PCB sorbed to sand more consistently, had four control replicas and four

Table 1. Behavioral repertoire of *Hemigrapsus oregonensis* with the 20 activities categories into which the behavioral units were grouped.

BEHAVIORAL UNIT	ACTIVITY CATEGORY
Shelter use	
1 Unsheltered	1 Unsheltered
2 Sheltered	2 Sheltered
3 Perched	3 Perched
4 Buried	4 Buried
Posture	
5 Mating	5 Mating
6 Display	6 High
7 High	6 High
8 Low	7 Low
9 Huddled	8 Huddled
Locomotion	
10 Standing	9 Standing
11 Walking	10 Walking
12 Climbing	11 Climbing
13 Running	12 Fast Movement
14 Fleeing	12 Fast Movement
15 Chasing	12 Fast Movement
16 Swimming	*
Social Behavior	
17 High Intensity Lateral Merus Displaying	13 Displaying
18 Mod Intensity Lateral Merus Displaying	13 Displaying
19 Low Intensity Lateral Merus Displaying	13 Displaying
20 Claw Shuddering	13 Displaying
21 *Hemigrapsus* Displaying	13 Displaying
22 Walking Leg Contact	14 Walking Leg Contact
23 Piling Up	*
24 Lunging	15 Fighting
25 Striking	15 Fighting
26 Pinching	15 Fighting
27 Pushing	15 Fighting
Prefeeding	
28 Claw Probing	16 Prefeeding
29 Dactyl Searching	16 Prefeeding
30 Maxilliped Sweeping Out	16 Prefeeding

Table 1. (Continued)

Feeding

31 Maxilliped Sweeping Substrate	17 Feeding
32 Claw Feeding	17 Feeding
33 Scraping Feeding	17 Feeding
34 Claw Tearing	17 Feeding
35 Spitting	*

Environmental Manipulation

36 Digging	18 Digging
37 Claw Shovelling	18 Digging
38 Bulldozing	18 Digging
39 Sand Scraping	18 Digging
40 Lifting	18 Digging
41 Ice Tonging	*

Sexual Behavior

42 Grabbing	19 Sex
43 Embracing	19 Sex
44 Struggling	19 Sex
45 Copulating	19 Sex

Body Maintenance

46 Eye Bobbing	20 Cleaning
47 Claw Picking	20 Cleaning
48 Dactyl Picking	20 Cleaning
49 Claw Rubbing	20 Cleaning
50 Leg Rubbing	20 Cleaning
51 Claw Sweeping	20 Cleaning
52 Eye Sweeping	20 Cleaning
53 Antennule Sweeping	20 Cleaning
54 Mouthpart Mashing	20 Cleaning
55 Abdomen Flapping	20 Cleaning
56 Egg Ventilating	20 Cleaning
57 Egg Grooming	20 Cleaning
58 Foam Bathing	20 Cleaning
59 Antennule Flicking	*
60 Scaphognathite Beating	*
61 Molting	*
62 Defecating	*

* These behavioral units were identified for this repertoire but were not used in the construction of the activities budgets.

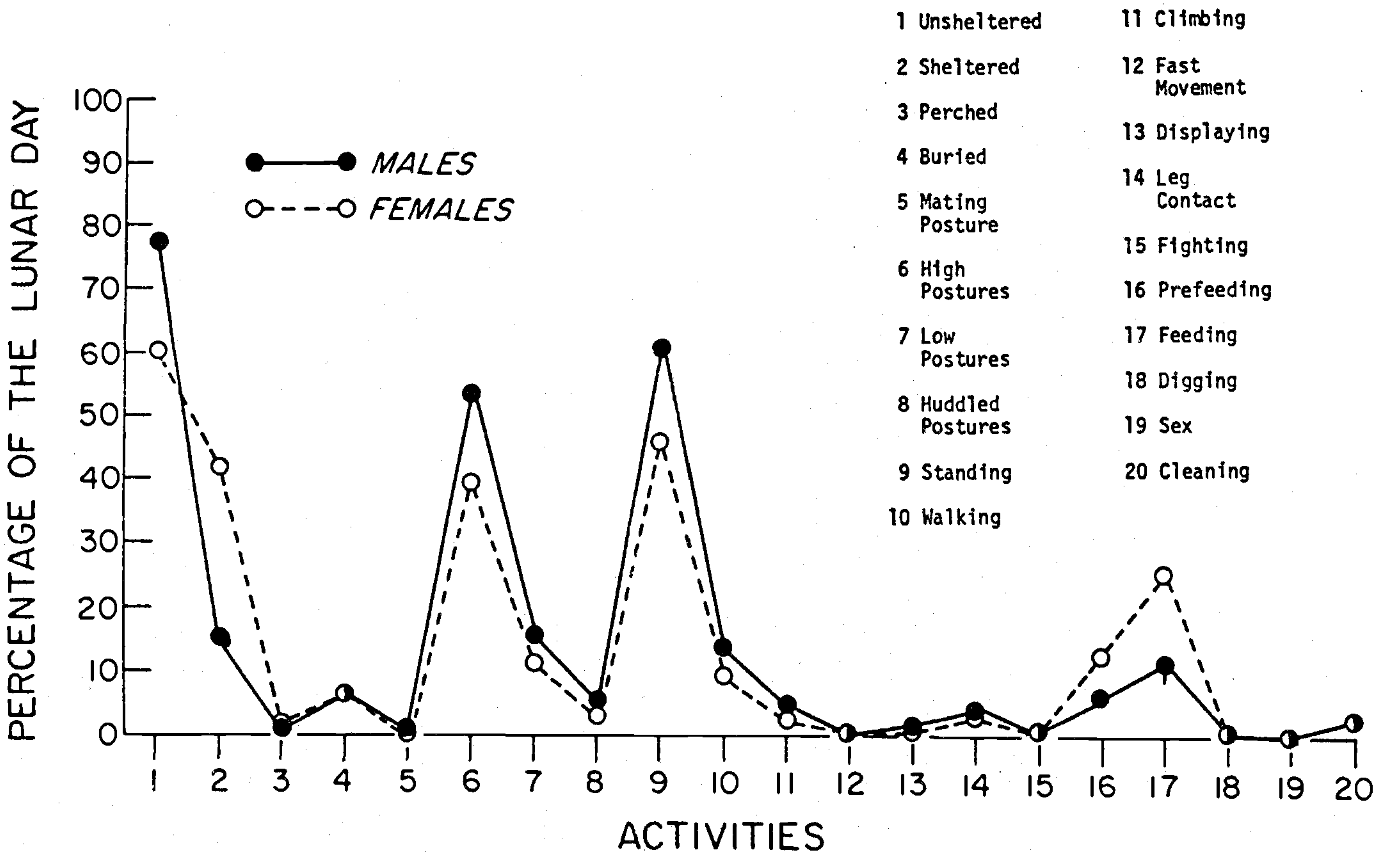

Figure 1. Activities Budgets of Male and Female Crabs under Ordinary Conditions.

PCB-exposure replicas with sand containing an average of 442 ppm. In the second experiment the observer did not know which tanks received the PCB-contanimated sand.

RESULTS AND DISCUSSION

Comparison of the average activities budgets of male and female crabs observed under the eight lunar day schedule (Figure 1) shows that under ordinary conditions with no PCB present in the replicas female crabs spend considerably more time sheltered and feeding than males. The male crabs exhibit slightly more social interaction, displaying, walking, leg contacting, and fighting, than the females. The profile for male crabs through the posture and locomotion categories (activities 5 to 12 in Figure 1) is somewhat above that for females and reflects the males' being more unsheltered, but both sexes having profiles of the same shape indicates that female crabs budget their unsheltered time among postures and locomotion in the same proportion as males.

The points in Figure 1 to 3 represent the mean of the replica averages of the percentages of the lunar day spent in the activities. The replica average is the mean of the three male or female crabs and is the basic measurement for the figures and following analysis because the replica is the experimental unit in the statistical sense (3). The variability among replicas is less than the variability among individual crabs within a replica because within a replica a hierarchy based on size develops. One consequence of this hierarchy is that the dominant crab shelters itself less than the others (Table 2).

Male crabs not only spend a larger share of time displaying than females, but also give different kinds of displays. Although both sexes commonly give lateral merus displays, in accord with others (15) we observed only the male giving the claw shuddering display. Females typically give the *Hemigrapsus* display described elsewhere (22), but males rarely do.

For both sexes certain activities predominate at certain tide stages. On a rising tide crabs emerge from shelter and feed but are quiet at high tide. General movement, shelter maintenance and displays increase with the falling tide, and the crabs seek shelter during exposure at low tide. At night the crabs are more active, with general movement and feeding occurring from submersion through the high tide and into the falling tide. During the nocturnal low tides crabs often do not shelter themselves but stand quietly.

Similar to fiddler crabs (9, 11), *Hemigrapsus oregonensis* concentrates PCB's from contaminated sediment, and male crabs have the lower whole body burdens. After 31 days at the low exposure in the first experiment, whole body concentrations of PCB averaged 13 ppm for males and 59 ppm for females. At the moderate exposure males concentrated an average of 15 ppm PCB and females 57 ppm. In the second experiment

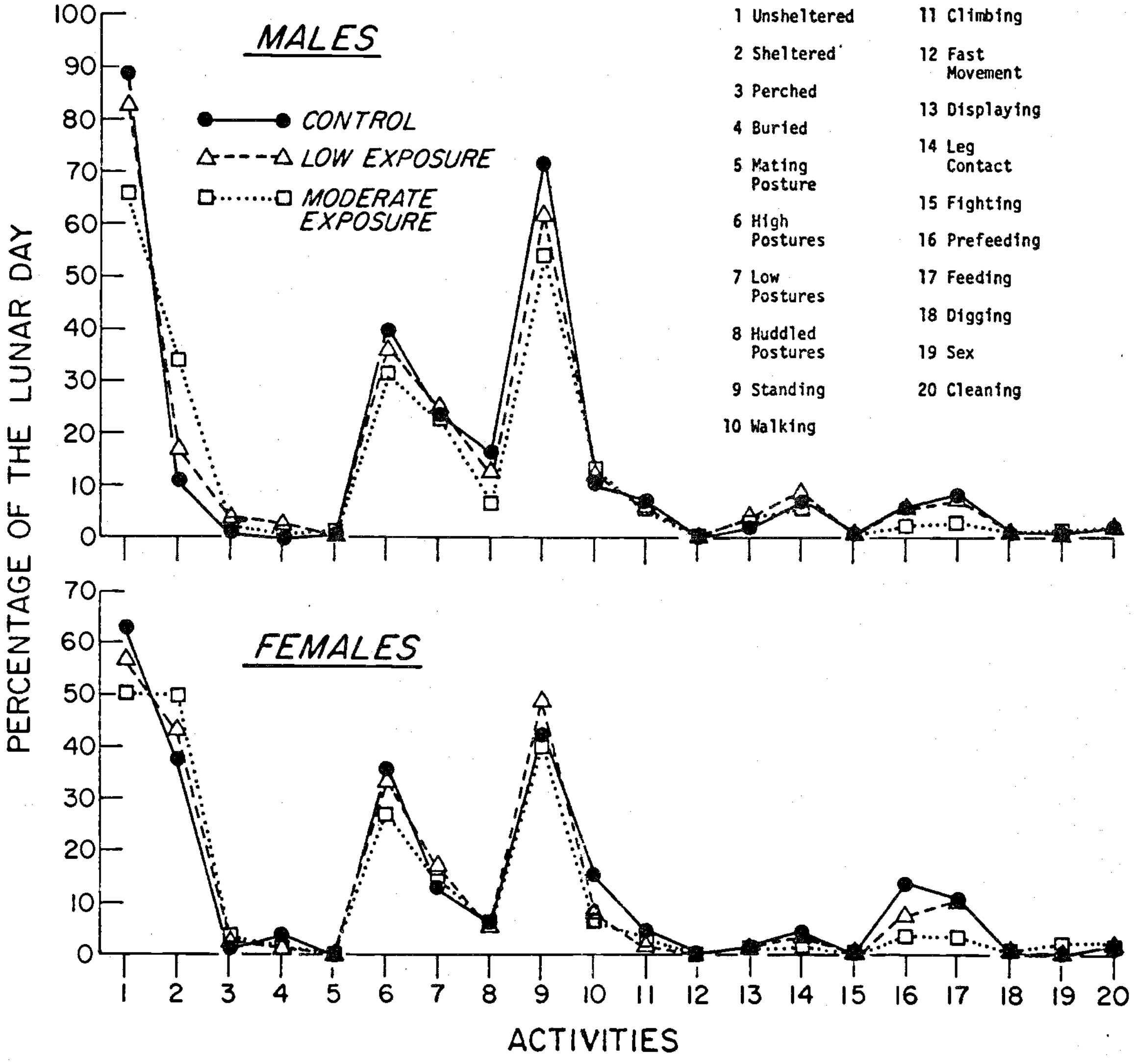

Figure 2. Activities Budgets of Male and Female Crabs in the First Experiment.

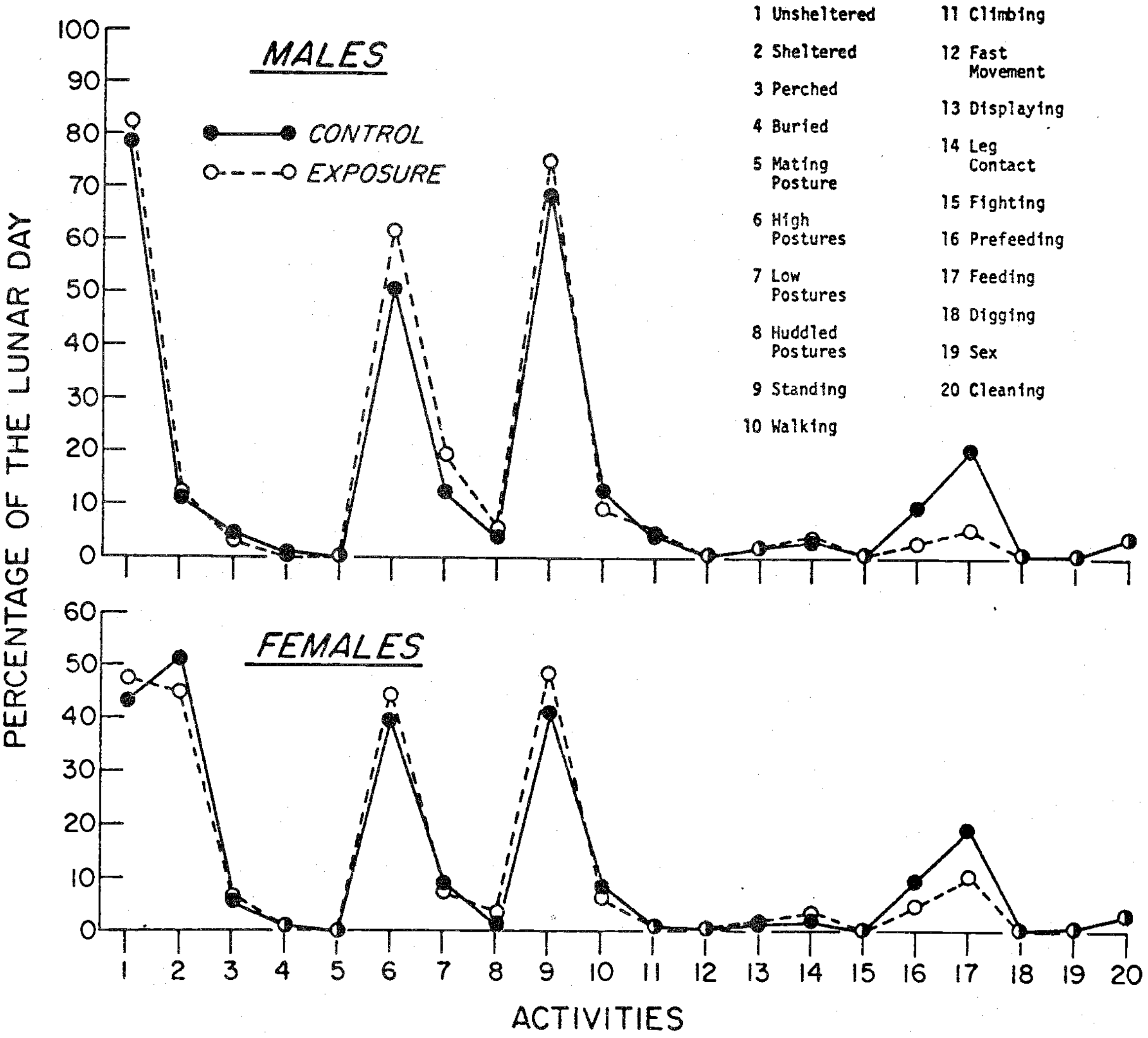

Figure 3. Activities Budgets of Male and Female Crabs in the Second Experiment.

Table 2. The Proportion of the Lunar Day Male and Female Crabs Spent Unsheltered

Males			Females		
Proportion of time unsheltered		Carapace width	Proportion of time unsheltered		Carapace width
Individual	Replica Average	mm	Individual	Replica Average	mm
.885		28.4	.739		25.5
.870	.868	24.0	.577	.589	21.2
.849		25.1	.451		21.1
1.000		28.7	.654		21.5***
.629	.739	24.4*	.458	.491	23.9
.589		23.6	.360		19.1
.958		30.5	.662		24.1
.907	.829	24.1	.473	.532	20.5
.623		24.4	.461		20.6
.998		30.0	.479		20.5
.875	.892	24.7	.462	.407	22.0
.803		24.6	.281		21.5
.868		29.5	.476		19.5
.642	.700	24.5	.326	.371	24.5
.591		26.0	.310		21.4
.753		23.6	.912		23.5
.745	.708	25.5	.626	.623	20.5
.626		28.5**	.330		21.4***
.916		25.5	.796		23.0
.756	.774	26.0	.638	.642	20.1
.649		24.6	.494		22.5
.766		24.2	.540		20.0
.668	.680	26.8	.405	.357	21.5
.606		24.2**	.127		21.5
Mean .774	.774	25.9	.502	.502	21.7
STD DEV .136	.081	2.2	.178	.113	1.6

* Crab lost both claws.
** Crab lost one claw.
*** Female crab became ovigerous.

males had a body burden of 36 ppm PCB after 23 days while females averaged 79 ppm. A female exhibited the maximum concentration observed in either experiment, 190 ppm PCB.

Examination of Figure 2, the average activities budgets for male and female crabs in the first experiment, reveals that the budgets for control and lowly exposed conditions differ little except that females spend less time buried and prefeeding under PCB exposure. Under moderate exposure both sexes considerably decrease the time spent feeding, and females spend more time sheltered but less buried.

The time consumed in feeding by both sexes is also markedly lessened under exposed conditions in the second experiment (Figure 3). Like the first, the second experiment finds the female crabs buried for a lesser time when exposed to PCB, but, unlike the first, sheltering by females does not increase.

To explore further the differences in activities budgets between ordinary and stressing conditions we performed multivariate discriminant analyses. When all 20 activities categories are used as variables (MANOVA Program of OSU Computer Center), discriminant functions for both sexes in each experiment do not clearly distinguish the budgets of the two conditions.

Next we performed stepwise discriminant analysis (BIOMED Program of OSU Computer Center), which is designed to choose the most parsimonious discriminant function. This discriminant function, in turn, can be used to indicate in which activities the budgets for ordinary and stressing conditions differ as well as to classify newly measured budgets as belonging to ordinary or PCB-exposed conditions. Essentially the BIOMED Program answers the question: If one were to pick n number of variables from the 20 available for a discriminant function, which particular activities would combine to give the discriminant function? The program selects variables stepwise, that is, one at a time, on the basis of the variables with the largest F-value. To enter the discriminant function a variable needed an F-value exceeding that for the 0.01 significance level.

The activities categories selected by the BIOMED Program for stepwise discriminant analysis are listed in Table 3 in the order in which they were selected. The repeated selection of a feeding category as the first variable in the discriminant function for both sexes and both experiments indicates that feeding behavior is consistently influenced by PCB exposure. A posture category also repeatedly appears in the four functions. For the males, locomotion is influenced in both experiments. Female crabs in both experiments show differences between the two budgets in shelter use, digging into the substrate, and cleaning.

At least two perspectives on the discriminant analyses exist. First, the inability to clearly distinguish the budgets when all 20

Table 3. Discriminant functions resulting from stepwise analyses with the activities categories given with their final coefficients and in the order of their selection.

	ORDER OF SELECTION					
	1	2	3	4	5	6
First Experiment						
Males	Feeding	Climbing	Mating Posture	Huddled Posture	Fast Movement	
	+3030	+53	-1076	-545	-10232	
Females	Prefeeding	Buried	High Posture	Cleaning	Sheltered	
	-1915	-2391	+214	+848	-113	
Second Experiment						
Males	Feeding	Walking	Cleaning	Huddled Posture	Displaying	Perched
	-254	-2055	-3796	+352	+476	+263
Females	Prefeeding	Low Posture	Digging	Cleaning	Feeding	Unsheltered
	+0	+57	-1856	-79	+56	-0.31

variables rather than five or six are used suggests that a small change in behavior, in our case in feeding, allows the crab to maintain its general activities. Qualitative observations not reflected in the quantitative data encourage this suggestion. The crabs shifted from feeding on sediment and particles in the sediment under ordinary conditions to feeding by scraping the algal growth from the chamber walls and rocks under exposed conditions. We plan more experiments to refine these observations. Such a shift from feeding on contaminated sand to other apparently less available food sources may serve to lessen the received dose of PCB and has implications for the cycling of this pollutant in the estuarine ecosystem.

Although such pollutant-induced shifts in the kind of feeding behavior found here have not been observed elsewhere, other crustaceans do exhibit depression of feeding in the presence of crude oil (1, 16). Exposure of *Pachygrapsus crassipes* to crude oil extracts inhibits the feeding responses (16) which include the similar prefeeding behaviors of *Hemigrapsus oregonensis* found in Table 1. The time which lobsters, *Homarus americanus*, use to find food is increased in the presence of crude oil extracts (1).

The other view of multivariate analyses has implications for the methodology of future pollutant-behavioral studies. If we had initially chosen to observe just the five of six categories selected by the stepwise procedure, we would have observed a considerable shift in behavior. Alternatively, if we had chosen to observe any of the other categories, we might very well have missed observation of a behavioral effect of PCB exposure and our predictions from the exact same experimental events would be far different. In behavioral studies such as this one, one should initially look at the animal in the broadest terms and resist the temptation to focus one's vision down to a few aspects of behavior.

CONCLUSIONS

Under ordinary conditions male and female shore crabs, *Hemigrapsus oregonensis*, differ in their activities budgets. Under stressing conditions when PCB-contaminated sand is present in the habitat replicas both sexes consistently show a decrease in the time spent feeding but differ in other points of the activities budget where stressinduced shifts occur.

ACKNOWLEDGEMENTS

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SMALL-SCALE HORIZONTAL DISTRIBUTION OF COASTAL COPEPODS[1]

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ABSTRACT

The horizontal spatial distribution of four species of coastal copepods was investigated by pumping two continuous series of samples and subjecting the data collected to statistical analysis. Sampling was conducted 1.5 km offshore of Moolack Beach, Oregon, at a depth of 7 m. Two small boats, each equipped with a pump of 280 liter min^{-1} capacity, were anchored 40 m apart and collected a series of 50 consecutive, paired zooplankton samples. Current speed and direction were measured at the beginning of each sampling interval. The amplitude and frequency of the fluctuations in copepod densities indicate large changes in density occurred over linear distances of 30 m to a few hun dreds of meters. Peaks of abundance occurred simultaneously for most categories of copepods counted. Time series analysis showed that a lag of 12 min (equivalent to 159 m) occurred between observation of abundance peaks at the seaward boat and their observation at the shoreward boat. This suggests that the variations in zooplankton density occurred in a pattern which is oblique to the direction of the current.

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INTRODUCTION

Since the beginning of zooplankton sampling there has been interest in the spatial distribution of these small animals. Knowledge of distributional patterns is vital for understanding basic interrelationships between the zooplankton and the environment. Historically, distributional studies involved the use of nets towed over hundreds of meters with comparison of samples taken many kilometers apart. More recently, interest has progressed to smaller scales of spatial distribution--tens to hundreds of meters. This paper presents the results of a study of zooplankton distribution on these smaller scales by a new combination of familiar techniques. Statistical methods for the analysis of time series have been applied to extended series of samples taken sequentially with plankton pumps. It has been found that the power of this combination can be enhanced by the use of pumps on several vessels to take parallel, simultaneous sample series.

A number of previous studies have suggested this combination of methods. Cassie (1958) suggested that small-scale spatial distribution could be determined by anchoring a boat and pumping samples continuously as the current flowed past the intake. In a second paper (Cassie, 1959) he reported on two individual series of samples pumped from a pier in Wellington Harbour, New Zealand, but his results were inconclusive with respect to characterization of smallscale density variations. Wiebe (1970) towed a modified Longhurst-Hardy plankton recorder (LHPR) to obtain a series of 550 m long horizontal records in the lee of Guadalupe Island, Baja California. He estimated plankton patch size by assuming patches were circular, and all of the same size, both of which are improbable *a priori*. Fasham, *et al*. (1974) have made three LHPR tows of 1800 to 4560 m length at 550 m depth in the Atlantic. They fitted the data for numbers of animals per sample with a statistical distribution, the parameters of which are supposed to represent patch dimensions and density characteristics. To do that, they had to assume circular patches with constant numbers of animals per patch, Gaussian distribution of density around the patch centers, and random distribution of patches. All of these assumptions are unsupported by data or *a priori* logic.

In light of the results of Wiebe and of Fasham, *et al*., it is likely that the notion of actually identifying plankton patches should be dropped. Zooplankton are certainly patchy, but attempts to statistically discern patches as such lead to untenable assumptions. It will be more productive to attempt to characterize small-scale distributions directly and to seek correlations between density variations and variations in components of the environment such as temperature. It is also unlikely that fully convincing data can be extracted from LHPR records until the instrumental problems pointed out by Haury (1973) have been solved.

Several recent studies have attempted to correlate small-scale distribution of phytoplankton with physical parameters (Denman and

Platt; Kamykowski, 1974; Platt, Dickie, and Trites, 1970; and Platt, 1972). These studies are based upon spectral analysis of variations in phytoplankton density, measured as chlorophyll, along extremely long sample series. Many implications are arising which will help to answer questions related to small-scale zooplankton distribution patterns. For example, Platt (1972) has shown that the spatial power spectrum of variations in chlorophyll density is very close to that expected for a homogeneously turbulent fluid.

We have used two boats anchored 40 m from each other to pump two series of samples simultaneously. The boats were anchored offshore in Oregon coastal waters to take advantage of the extremely high population densities available there in the summer and fall months. The paired sample series allows a definitive determination of whether abundance variations actually occurred in the water or were an artifact produced by the pump or some other feature of the sampling regime. The pumping system provided a small but adequate sample within a two minute interval, allowing refined resolution of changes in abundance over short distances. Because samples in the two series were pumped simultaneously, they can be paired. This allows application of a cross-correlation analysis to determine the most probable spatial interrelation between changes of abundance observed along the two sampling lines. The result is a new approach to studying the smallscale horizontal spatial distribution of zooplankton.

METHODS AND MATERIALS

Sampling was conducted on 5 October 1973 between 0900 and 1400 hrs Pacific Standard Time at a permanent buoy positioned in 18 m of water 1.5 km off Moolack Beach, Oregon. Headlands are present to the north and south of the sampling station. Winds were from the southwest at a mean speed of 2.7 m sec^{-1} with partly cloudy skies and calm seas. A 35-foot boat, the PAIUTE, and an inboard-engined dory were each equipped with identical pumping systems: a Jabsco centrifugal pump, model 10490, with a neoprene impellor run by an hydraulic motor. The pumps were fitted with 5.1-cm (inside diameter) intake and discharge hoses and a rapid filter changing system. Organisms passed through this system with little or no damage. The pumping rate was 280 liters min^{-1} (determined from the time required to fill a container of known volume) with an estimated fluctuation of less than ± 15 liters min^{-1}. This rate was constant over the entire depth range at the sampling station. The flexible intake hose was attached to a weighted line to hold it vertical in the water.

The filtering system (similar to that of O'Connell and Leong, 1963) was designed to reduce the water flow rate before actual filtration. It consisted of a funnel with 0.233 mm Nitex® mesh inserts glued over three openings cut in the funnel's frame. The exhaust of the funnel was threaded (a jar with its bottom removed), and was closed by a jar cap with a 38 mm hole in its center. This cap held a 50-mm diameter disc of 0.233 mm Nitex® mesh. A rubber gasket between the cap and the mesh disc supplied pressure to hold the mesh taut when the cap was screwed onto the exhaust. Two of these filtering systems were used on each boat, permitting continuous filtration. While removing the sample from one funnel, water was filtered through the other. Samples were preserved by placing the mesh disc in a sample jar with ten percent formalin During sampling, water swirled around the funnel, continually flushing it. Although not washed down between samples, organisms did not accumulate on the funnel mesh.

The sampling proceeded in the following sequence: the PAIUTE was tied to the permanent buoy. By pumping two minute samples at varying depths it was determined that the depth of maximum abundance of organisms was 7 m. A Hydroproducts® Savonius rotor current meter with directional vane and on-deck readout module was positioned at 7 m to determine current direction. Because the current was from north to south, the dory anchored 40 m due west of the PAIUTE. Current velocities were not measured from the dory. Sampling times on both boats were coordinated by means of citizen's band radio. Sampling began at 1157 hours with a five second countdown to ensure sample changes at the same instant on both boats. Each two minute sample represented a 0.56 m^3 of water filtered. After 10-15 samples another countdown was made to check that both boats were changing samples simultaneously. After pumping 50 pairs of samples at 7 m depth, a vertically stratified sample series was collected. Replicate samples, pumped continuously, were taken at five depths on each boat: just below the surface (0), 3, 7, 10, and 13 m. The pump was run for five minutes at each new depth before sampling to prevent contamination of samples between depths.

In the lab, three samples taken at random throughout the series were counted to determine species composition and densities. A total of 22 species of zooplankton were observed in those samples. Nine categories were selected for enumeration:

Acartia clausi Giesbrect females
A. clausi males
Acartia longiremis (Lilljeborg) females
A. longiremis males
Pseudocalanus sp. females
Pseudocalanus sp. males
Pseudocalanus copepodites
Oithana similis Claus females
O. similis males

These were chosen on the basis of abundance and specificity of identification. Small subsamples were sequentially counted under a binocular microscope in a ruled petri dish until the entire sample had been counted. Organisms trapped on the filter mesh were also identified and counted. Subsampling was not employed because it was felt it would introduce more variability than was expected between samples. This turned out to have been overcautious.

The current speed for each sampling interval was recorded at the beginning of the interval. When multiplied by the sampling interval (2 min) an estimate of the linear movement of water past the intake during each sampling interval was obtained, the "nominal linear dimension." The mean current of 0.22 m sec^{-1} flowing nearly directly from north to south resulted in a mean nominal linear dimension of 26.47 m. The range of velocities was 0.16 to 0.29 m sec^{-1}, equivalent to 20 to 35 nominal linear meters per sample.

The spatial changes in density of the nine categories were studied by application of simple techniques for the analysis of time series. An auto-correlation function was used to determine if the sample interval was sufficiently small to provide a reasonable representation of the dominant frequencies of density variation. A cross-correlation function was used to quantify the similarity of abundance variations between the two boats and between categories. A product-moment correlation coefficient was calculated in these methods for all pairs of samples at a given distance (lag) from each other in the same (auto) or different (cross) sample series. The results are presented as tables or graphs of correlation vs. lag. All computations were performed by computer using the ARAND program system of Ochs _et al_. (1971). The cross-correlation functions were computed from data filtered by conversion to a weighted (1-2-1) running mean.

RESULTS

Abundance plots for four representative categories are presented in Fig. 1. Distinct variability in the numbers of organisms observed per sample is apparent for both sample series. The density at the peaks is three to ten times as large as the density in the valleys. Two levels of fluctuation are observed in the graphs: overall trends with several samples in each peak or valley, and smaller fluctuations within the larger trends. The latter are considered to be the result of density variations on a scale smaller than the sampling interval.

The dory samples generally had a higher density than the PAIUTE samples. Means of 2143 and 1427 copepods were counted in the 50 dory and 50 PAIUTE samples respectively. We have no explanation for this. It is probably the result of an undetected instrumental error, but it may represent a real feature of the onshore-offshore distribution gradient. There is a relationship between the position and shape of the abundance peaks on the two boats, with the PAIUTE pattern displaced upstream. When each boat is considered separately and the categories are compared, the multiple-sample peaks of abundance occur at about the same sample number for every category except _O. similis_ male and female.

Table I presents the auto-correlation functions for all nine categories on both boats. Fig. 2 shows a graph of the auto-correlation

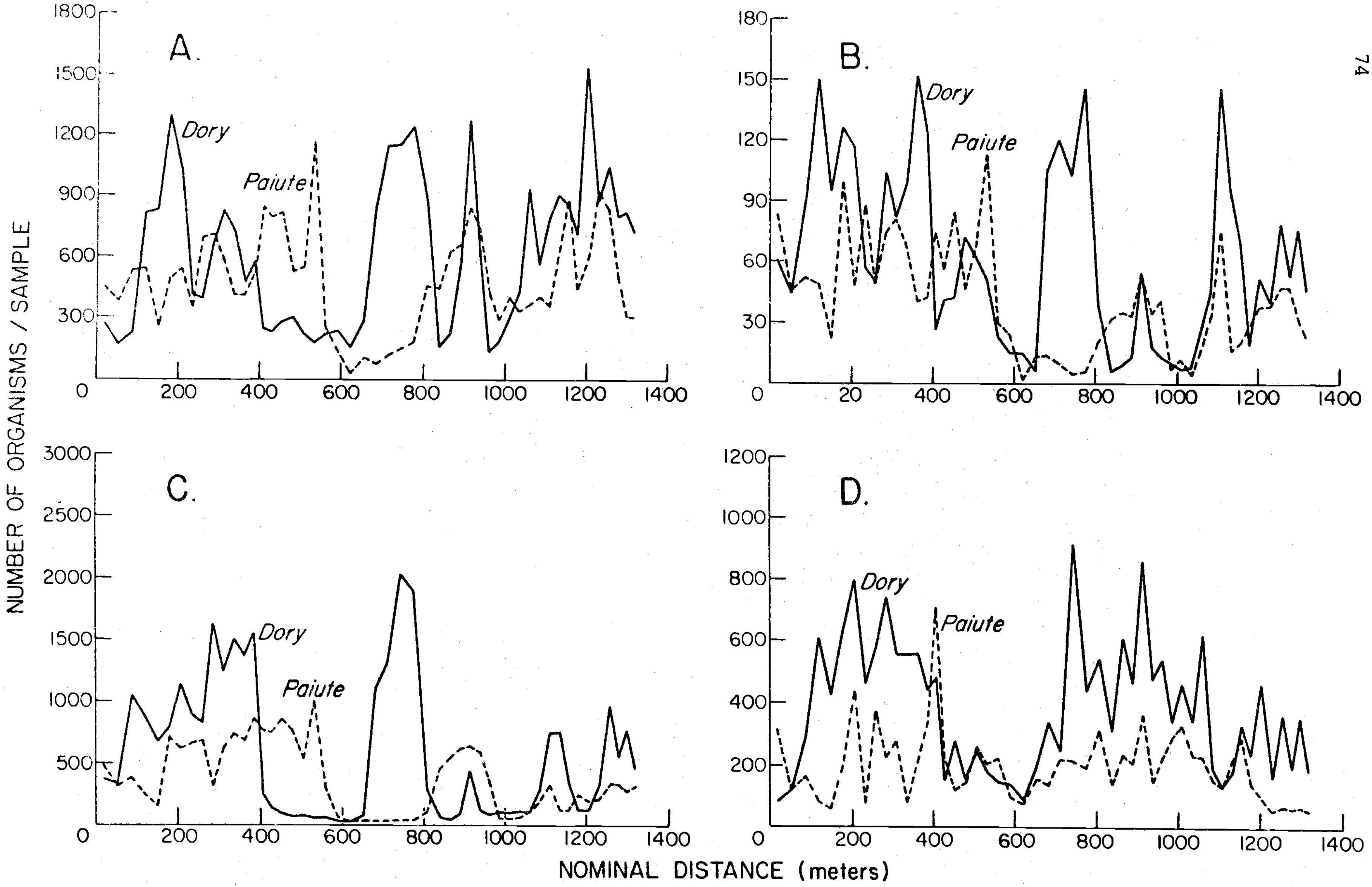

Figure 1. Abundance plots of copepods sampled by dory and PAIUTE. A) Acartia clausi male, B) Acartia longiremis female, C) Pseudocalanus sp. copepodite, D) Oithona similis female.

Table I. Biased auto-correlation function for each category of copepods on each boat at several lag steps.

DORY					
	LAG				
Species	1	2	3	4	5
Acartia clausi female	0.70	0.41	0.19	0.13	0.15
" " male	0.58	0.23	-0.02	-0.03	0.01
Acartia longiremis female	0.56	0.24	0.02	-0.13	-0.10
" " male	0.63	0.34	0.17	0.05	-0.03
Pseudocalanus sp. female	0.39	0.16	0.08	0.21	0.16
" " male	0.49	0.41	0.35	0.26	0.27
" " Copepodites	0.70	0.40	0.14	-0.03	-0.10
Oithona similis female	0.41	0.50	0.17	0.25	-0.01
" " male	0.38	0.35	0.08	0.24	0.06
TOTAL	0.63	0.36	0.03	-0.07	-0.16

PAIUTE					
Acartia clausi female	0.75	0.50	0.29	0.11	-0.05
" " male	0.53	0.24	0.17	0.05	-0.09
Acartia longiremis female	0.43	0.43	0.26	0.24	0.29
" " male	0.52	0.44	0.35	0.14	0.16
Pseudocalanus sp. female	0.64	0.49	0.37	0.26	0.28
" " male	0.52	0.37	0.18	0.10	-0.01
" " Copepodites	0.75	0.59	0.46	0.23	0.15
Oithona similis female	0.22	0.09	-0.13	0.17	0.03
" " male	0.32	0.29	0.10	0.11	0.12
TOTAL	0.67	0.47	0.29	0.09	0.00

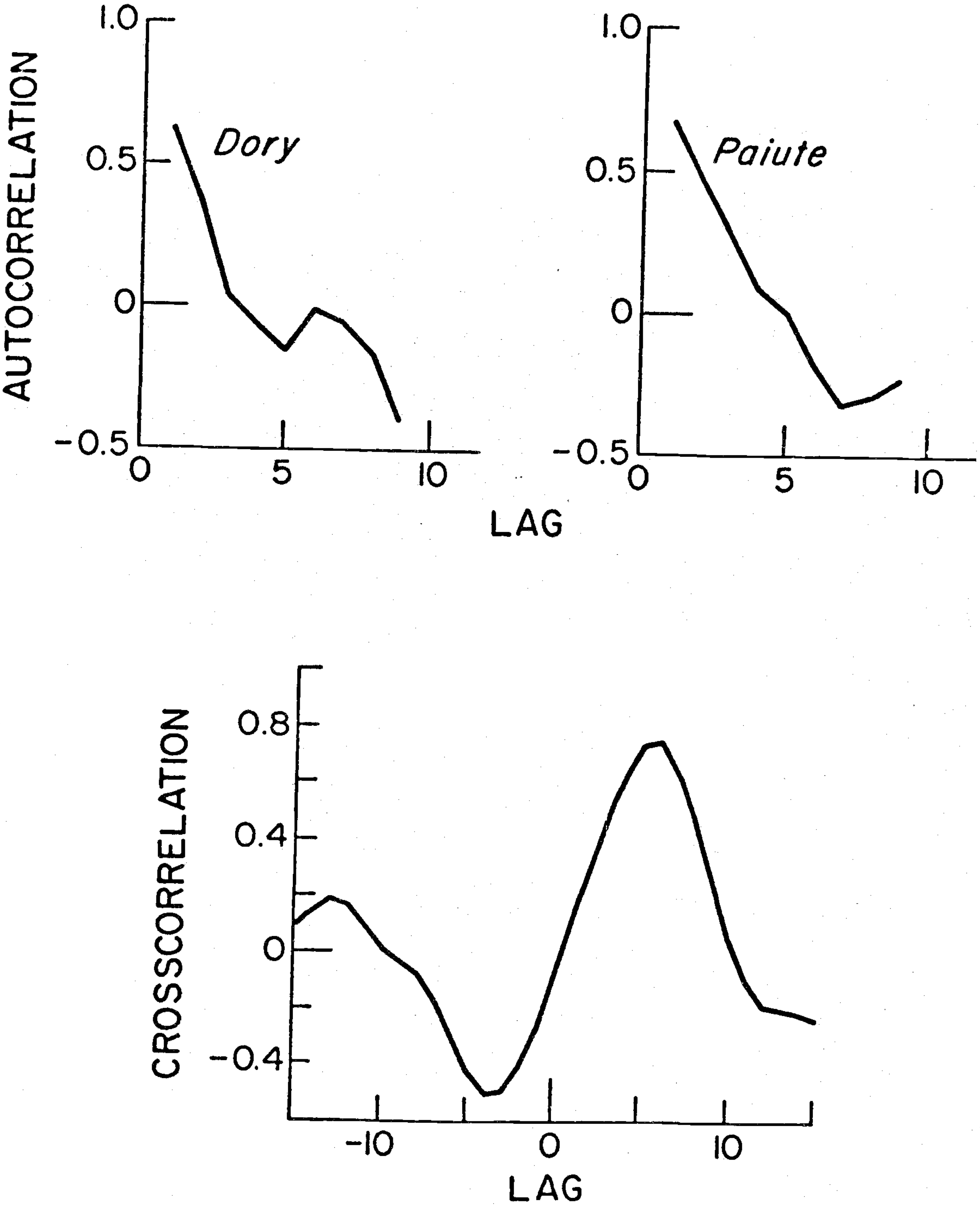

Figure 2. Auto- and cross-correlation functions for total number of organisms.

function for the total of all nine categories. It is clear that neighboring abundance estimates are substantially correlated (the 95% significance level for a single lag step is r = 0.235) to a distance of two to three samples, but not beyond that point. Therefore, the sample series should adequately represent fluctuations with "periods" greater than 26 m, the approximate sample length. In most cases the auto-correlation in the PAIUTE series extends over more lag steps than in the dory series. None of the correlations at greater lags is significant (although no probablistic tatistical test is applicable in this type of analysis), which implies that no distince periodicity was present in the density fluctuation. The series are too short for such a determination to be meaningful. The auto-correlation values at small lags are lower for Oithona similis than for the other forms. Presumably this is because more of its heteorogeneity in distribution is at smaller scales.

Table II presents the cross-correlation functions for all nine categories. The functions indicate that the highest correlation between dory and PAIUTE samples for total numbers of organisms occurs at a positive lag (PAIUTE series displaced upstream from the dory series) of six samples, and that for the other categories the lag varies between four and six samples. Total abundance estimates for the dory and PAIUTE are plotted in Fig. 3 with a lag of six samples. This displacement is equivalent to 159 m. It implies that the sequence of density variations is at an angle of approximately 14° to the direction of current flow, with density features passing the offshore station first.

Simple correlation coefficients between categories are presented in Table III. All are positive and highly significant, except those between Oithona similis male and female categories and other categories. Those two categories are, however, highly correlated with each other.

DISCUSSION

Results of this study indicate that copepods in the very nearshore zone of Oregon are highly aggregated on spatial scales of a few tens of meters. This is in good agreement with previous work (Cassie, 1963). It was hoped that this study could define characteristic dimensions of the density fluctuations in space, and perhaps characteristic amplitudes as well. It must be concluded that a series of 50 samples, equivalent to a total of 1300 m, is not adequate for that purpose. However, the existence of significant auto-correlation between samples taken within 50 m and 75 m indicates that fluctuations of great amplitude on very small scales are rare enough that longer series should allow this characterization.

The graphs of abundance show the same peaks (distinguished by position and shape) to be present in both data series for all categories, with the exception that Oithona similis male and female did not exhibit the same peaks as the other categories, although their own

Table II. Cross-correlation function between boats for each category of copepods with the spatial series lagged in each direction. Positive lag steps move the PAIUTE series upstream relative to the DORY series.

Species	LAG -10	-9	-8	-7	-6	-5	-4	-3	-2	-1	0	1	2	3	4	5	6	7	8	9	10
Acartia clausi ♀	0.11	0.13	0.13	0.09	0.03	-0.02	-0.05	-0.03	0.02	0.09	0.18	0.28	0.38	0.48	0.55	0.54	0.43	0.28	0.15	0.06	-0.01
" " ♂	0.19	0.15	0.08	-0.05	-0.22	-0.36	-0.44	-0.41	-0.31	-0.19	-0.05	0.07	0.19	0.32	0.45	0.50	0.43	0.31	0.23	0.18	0.07
Acartia longiremis ♀	0.24	0.35	0.44	0.40	0.26	0.10	0.01	0.03	0.14	0.23	0.28	0.31	0.37	0.44	0.53	0.62	0.66	0.58	0.37	0.14	-0.00
" " ♂	0.11	0.21	0.26	0.22	0.10	-0.02	-0.10	-0.09	-0.01	0.09	0.17	0.27	0.39	0.53	0.66	0.77	0.79	0.67	0.47	0.29	0.16
Pseudocalanus sp. ♀	0.07	0.11	0.17	0.21	0.22	0.18	0.09	0.03	0.09	0.24	0.37	0.43	0.46	0.51	0.61	0.72	0.72	0.61	0.45	0.34	0.27
" sp. ♂	0.07	0.00	-0.06	-0.12	-0.19	-0.26	-0.28	-0.23	-0.10	0.07	0.17	0.19	0.30	0.53	0.73	0.82	0.80	0.66	0.47	0.33	0.21
" sp. C	0.16	0.17	0.17	0.10	-0.03	-0.17	-0.26	-0.26	-0.18	-0.08	0.02	0.16	0.34	0.54	0.71	0.83	0.85	0.73	0.51	0.26	0.04
Oithona similis ♀	-0.40	-0.47	-0.45	-0.39	-0.35	-0.31	-0.25	-0.14	0.00	0.20	0.40	0.48	0.52	0.54	0.52	0.48	0.38	0.26	0.20	0.14	0.00
" " ♂	-0.04	-0.15	-0.20	-0.17	-0.13	-0.12	-0.13	-0.07	0.08	0.21	0.29	0.38	0.42	0.50	0.51	0.45	0.32	0.20	0.10	0.00	-0.20
TOTAL	0.01	-0.03	-0.08	-0.17	-0.30	-0.43	-0.51	-0.49	-0.41	-0.26	-0.08	0.11	0.30	0.47	0.62	0.74	0.75	0.64	0.46	0.25	0.05

Table III. Product-moment correlation coefficients between species in the two sample series. PAIUTE abundances are compared above the diagonal, DORY abundances are compared below. Values greater than 0.33 are significant at the 0.01 level (N=50).

Species		1	2	3	4	5	6	7	8	9
		PAIUTE								
Oithona similis ♀	(1)	***	0.64	0.09	0.28	0.23	0.20	0.26	0.10	0.12
" " ♂	(2)	0.85	***	-0.03	0.25	0.45	0.32	0.54	0.42	0.40
Acartia clausi ♀	(3)	0.29	0.18	***	0.83	0.31	0.26	0.39	0.54	0.47
" " ♂	(4)	0.44	0.37	0.83	***	0.48	0.41	0.61	0.63	0.57
Pseudocalanus sp. ♀	(5)	0.35	0.32	0.24	0.26	***	0.56	0.84	0.78	0.79
" " ♂	(6)	0.42	0.39	0.41	0.32	0.72	***	0.74	0.54	0.48
" " C	(7)	0.46	0.52	0.49	0.49	0.60	0.75	***	0.87	0.78
Acartia longiremis ♀	(8)	0.39	0.40	0.63	0.56	0.64	0.75	0.88	***	0.92
" " ♂	(9)	0.26	0.28	0.59	0.53	0.55	0.67	0.82	0.92	***
		DORY								

DORY

PAIUTE

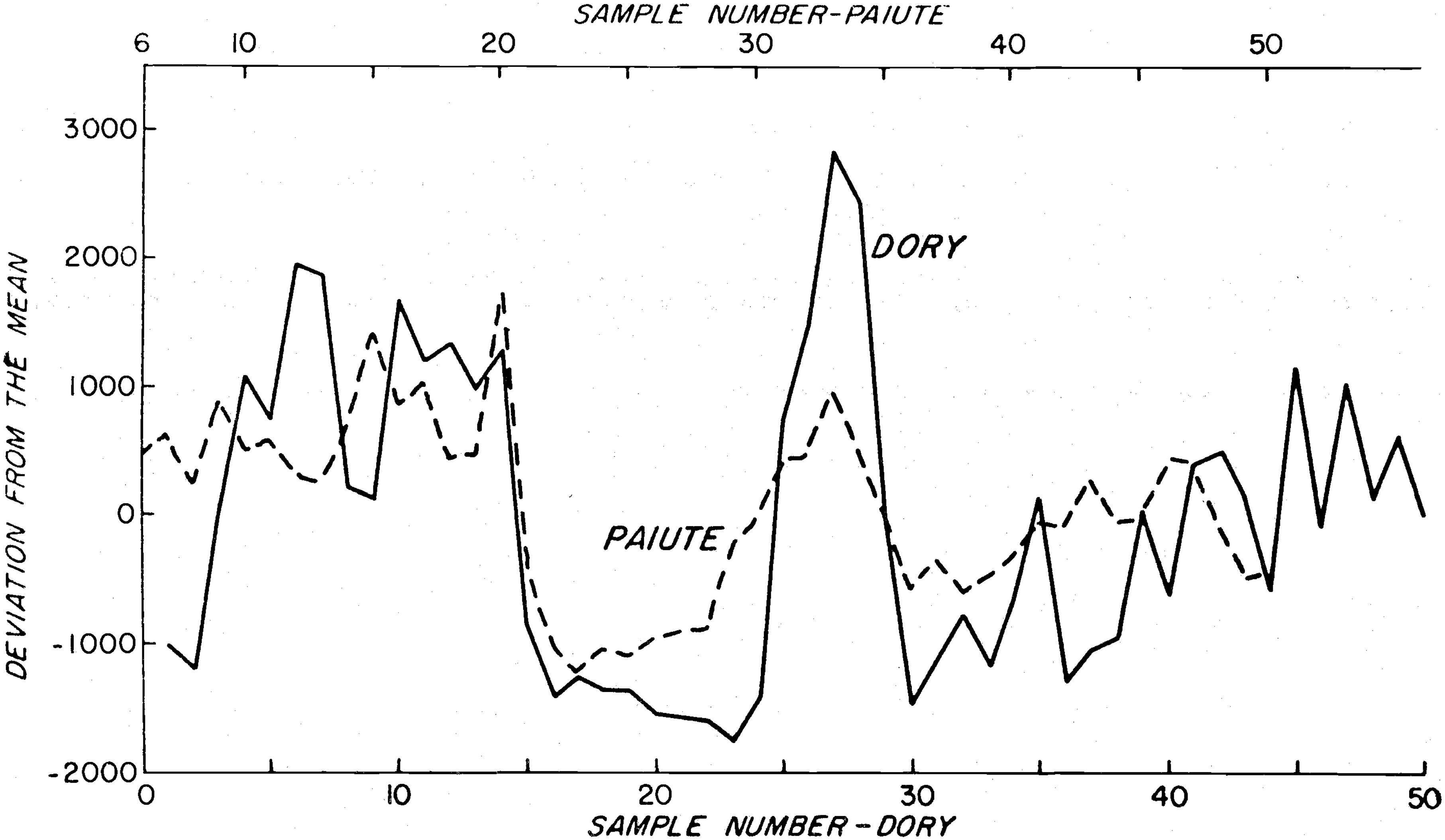

Figure 3. Deviation from the mean of total numbers of copepods with a time lag of twelve minutes (159 m) between dory and PAIUTE samples.

fluctuation patterns were similar. This coincidence of peaks indicates a strong multispecies nature for the small-scale distribution of the copepods. The high correlation of the categories (Table III) confirms that this common spatial distribution pattern is more than statistical artifact. It is improbable that species as different in their overall biology as *Pseudocalanus* sp., *Acartia clausi*, and *Acartia longiremis* would select identical zones in which to accumulate and from which to migrate. It is much more likely that they are being similarly affected by turbulent and other processes because they are alike in their simplest characteristics as particles. The discrepant species, *Oithona similis*, is nearly an order of magnifude smaller in body size. It may behave differently as a particle for that reason. It is possible also, that differences in size are the most important determinants of ecologyc processes which could induce aggregated distributions, such as predation. More data from more places on the concordance of distribution patterns between species will help resolve this problem, perhaps by suggesting different hypotheses.

Examination of possible sources of error in the pumping system is necessary to determine if such errors could have affected the results. The consistently higher numbers of organisms in dory samples and larger fluctuations on that boat in numbers of organisms in adjacent samples can most easily be explained by differences in volumes pumped. A flow meter at the discharge would permit much more accurate abundance estimates. Leong (1969) and Singarajah (1969) indicate sources of error for pumping systems, but only a few are factors in this study. Error due to organism damage was minimized by considering only copepods less than 2 mm long and by using a pump with a neoprene impellor. Most individuals emerge from the pumps in better condition than is typical for net samples. Avoidance is a major source of probable error whose magnitude cannot be determined. Fleminger and Clutter (1965) have shown that avoidance of nets tends to be influenced by density in copepods, the rate decreasing with increasing density. It is likely, therefore, that density fluctuations in a study like the present one will be differentially affected by avoidance. Zones of lesser density may appear to be relatively less than they actually are in the field. However, according to the criteria developed by Fraser (quoted by Wiborg, 1948), our sampling should have resulted in minimal avoidance because pumping rates were well in excess of his suggested minimum of 200 liters min^{-1}.

There is no strong explanation for the apparent angular orientation of the variations in copepod density to the direction of the flow. It is to be expected that diffusive processes in the coastal zone would make the fluctuation patterns elongate in the direction of flow (Seligman, 1956; Bowles *et al.*, 1958). With a pair of records as short as this, it is impossible to test whether the coherence obtained between the records at lags equivalent to about 160 m would persist in a very long record. The small number of major density oscillations sampled suggests that it could be a statistical artifact. It may be real, on the other hand, without having the nearby headland could very well have included an without having any general significance. The complexities in circulation induced by the nearby headland could very well have included an acceleration of the offshore water relative to the inshore water at some point in its transport along the coast, thus skewing the density field with respect to the residual current downstream.

This study has indicated the requirements for the sort of study which will enable us to determine the dominant features of the small-scale distribution of zooplankton. Pumps equipped with flow meters will be the best sampling method. They obtain small samples of known volume in a short period of time from a very limited area. The strong multi-species character in the distribution suggests the influence of a physical parameter. Therefore, current meter readings, sensitive continuous temperature readings, salinity, and chlorophyll measurements should be taken along with the zooplankton samples to give indication of the environmental correlatives of the spatial distribution. An attempt to do that for temperature in the present study failed because of equipment breakdown. Sample series must be very long, on the order of 500 samples. This requirement should not be prohibitive if samples are kept small enough and a limited number of categories are enumerated in each field experiment.

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MANGANESE CYCLING IN THE NEWPORT RIVER ESTUARY, NORTH CAROLINA

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and Douglas A. Wolfe[2]

Abstract

Dissolved Mn was measured in the Newport River estuary, North Carolina, in October 1974, February 1975, and April 1975 by extraction on Chelex-100 resin and subsequent analysis by flameless atomic absorption spectrophotometry. Dissolved Mn concentrations at intermediate salinities were in excess of those predicted by conservative mixing of inflowing river and ocean water, with maxima between 4‰ and 14‰ salinity.

Reduction and solubilization of Mn in bottom sediments in the upper estuary probably supplies much of the added dissolved Mn. Fixation of dissolved Mn on particles takes place in the lower estuary with subsequent upstream recycling.

The export of dissolved Mn from the estuary in excess of that input by the river is probably balanced by continuous traction flows of river-derived bed sediment or pulse inputs of suspended sediment in storm runoff followed by partial solubilization.

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Introduction

Manganese (Mn) is one of the more biogeochemically active transition metals in aquatic environments. It is readily transferred between solution and solid phases (sediments and suspended matter) in response to changes in environmental conditions such as pH, Eh, and the composition and relative concentrations of dissolved and particulate phases (Hem, 1963).

Estuaries are zones of transition between freshwater and marine environments and as such contain concentration gradients and other variations in geochemical conditions. This has suggested to many (Murata, 1939; Johnson *et al.*, 1967; Kharkar *et al.*, 1968) that estuaries are likely sites of transfer of Mn and other trace metals between solution and sediment phases.

We have sought to detect transfer of Mn between dissolved and particulate phases in the Newport River estuary, North Carolina, and to relate observed concentrations of Mn to possible reaction processes.

If the estuary is a system in steady state with inflowing river water and seawater, the concentration of a conservative dissolved substance is a linear combination of the concentrations in river water and seawater. Deviations from linearity are the result of processes which add or remove the substance from solution. This approach was used by Evans and Cutshall (1973) to observe nonconservation of radioactive ^{54}Mn in the Columbia River estuary. They concluded that observed positive deviations from linear dilution resulted from desorption from suspended and bottom sediments. More recently, Boyle *et al*. (1974) have developed the mathematics of nonconservative mixing models more rigorously.

Our sampling was carried out in the Newport River estuary, North Carolina (Figure 1). This is a shallow "drowned river" estuary with extensive intertidal areas, often populated by the marsh grass *Spartina alterniflora*. The mean depth at low tide is about one meter, with an intertidal range of 0.7 meters. A more thorough description of the region is given by Wolfe *et al*. (1973). Of particular interest to our study is the region between Cross Rock and 2.2 kilometers (km) upstream where the river proper widens into the estuary. This is the region of maximum salinity gradient where river water mixes with brackish estuarine water. On the flood tide brackish water is swept up the river as much as 8-10 km during periods of low river flow. The Cross Rock region is also important in being a deltaic area where fine sediments are deposited.

Methods and Materials

Sampling for dissolved and particulate Mn was done during three different periods: October 1974, February 1975, and April

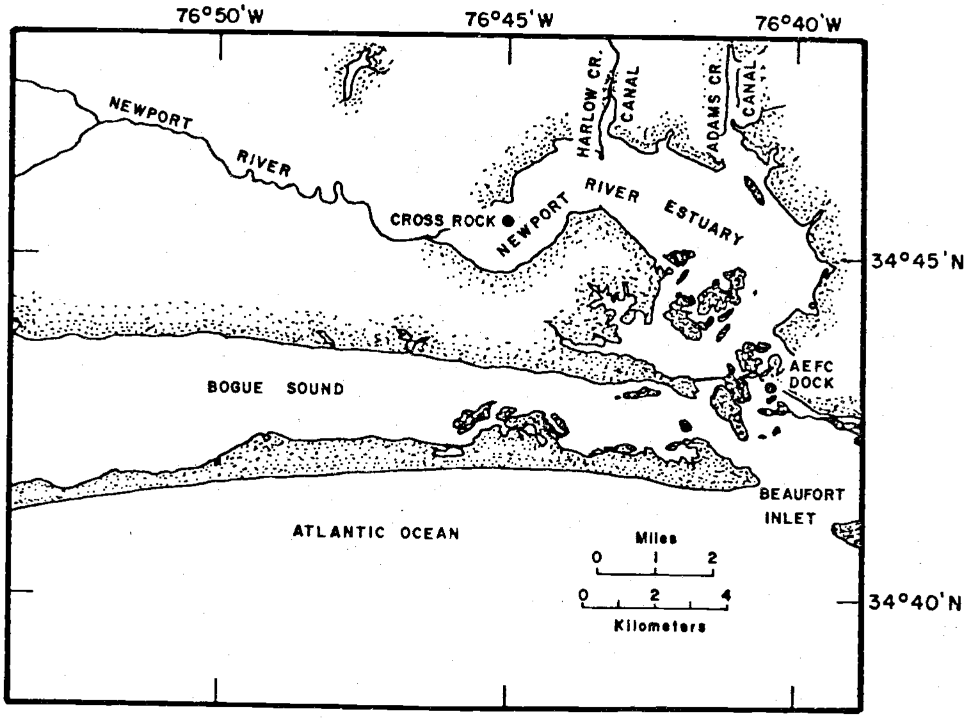

Figure 1. The Newport River estuary, North Carolina, USA.

1975. Longitudinal sample transects from salinity 0‰ to greater than 30‰ were taken within two hours of high tide on October 30 and 31, February 4, and April 14, and within two hours of low tide on February 5. Water was sampled periodically from the Newport River (0‰ salinity) and from the Atlantic Estuarine Fisheries Center dock (salinities greater than 30‰) during the three weeks preceding both the October 1974 and February 1975 estuary sampling transects. This was done to test the assumption of constant end member input concentrations required by the steady state model. This was not done for the April 1975 period.

During the entire October 1974 period approximately 4.52 cm of rain fell in the river's drainage basin (computed as the average rainfall of Morehead City and New Bern, N.C.). February, on the other hand, was an unusually rainy period. During the three weeks prior to these transects, 15.91 cm of rain fell with 4.45 cm falling during the night of February 4-5. This was sufficient to induce stratification behind Cross Rock at slack low water. Zero salinity was observed at the surface and 14‰ salinity was found at the bottom, only 0.5 meters deeper. During the three weeks prior to the April 1975 transect 8.15 cm of rain fell.

Duplicate water samples were collected from the upstream side of a 5-meter aluminum boat into a cylindrical 400-ml plexiglass pressure vessel. The samples were immediately pressure filtered with 30 psig N_2 directly through Nuclepore filters (47 mm diam by 0.4 μm pore size) held in acid-washed Nuclepore filter holders. This provided an operational separation of dissolved and suspended particulate fractions. The filters had been previously weighed so that suspended particulate concentrations could be determined later. Salinities were measured in the field with a Beckman RS5-3 inductive salinometer and later on the water samples themselves with an Aminco Cotlove chloride titrator to an accuracy of ±0.3‰.

Filtration was carried out until the filters clogged. This provided samples from about 50 ml in freshwater to 400 ml in seawater. Two filters were often required to provide a minimum 50-ml sample at the low salinity stations. Blanks were run identically using 300 ml of deionized distilled water (DDW). The filtrates were collected directly in acid-washed 500-ml polyethylene bottles and acidified with 0.1 ml redistilled HNO_3.

When returned to the laboratory the filtrates were frozen until time for analysis, and the filters were carefully washed free of sea salts with 15 ml DDW. The filters were then dried in a desiccator for 48 hours before being reweighed.

In preparation for analysis the filtrates were thawed and weighed. Five ml of 5% $K_2S_2O_8$ solution (previously purified by passage through a column of Chelex-100 resin) was added to the samples, still in their original polyethylene bottles. The caps were screwed tight and the samples allowed to digest for 16 hours at 90°C in a drying oven. This procedure oxidized dissolved organic matter which might otherwise bind Mn and prevent its subsequent extraction (Noakes and Hood, 1961).

Because sea salts interfere with Mn analysis, Mn was separated by the chelating exchange resin Chelex-100 (BioRad Labs, 50-100 wet mesh). The cooled and digested samples were brought to pH 6-7 with 5 ml of previously purified 8% $NaHCO_3$ solution. They were then allowed to flow through 0.7 cm diam by 4 cm polypropylene columns (BioRad) containing Chelex-100 resin in the ammonium form. The (uncontrolled) flow rate was about 2.7 ml/min. The columns were subsequently flushed with 30 ml of DDW to remove interstitial sea salts. Elution with 15 ml 2N HNO_3 stripped the collected Mn from the resin. The eluate was collected in 30-ml FEP Teflon beakers, evaporated to dryness at 70°C and then redissolved in DDW to which 0.05 ml HNO_3 was added. The extracts were diluted to 10 ml with DDW in cleaned 15-ml polypropylene test tubes.

The extracts were subsequently analyzed for Mn by flameless atomic absorption spectrophotometry by a Perkin Elmer HGA 2000 Graphite Furnace and Model 403 atomic absorption spectrophotometer. Twenty microliters of the extracts were injected into the graphite tube with an Eppendorf pipet. The furnace was programmed to dry the sample at 100°C for 30 seconds, ash it at 1100°C for 10 seconds, and atomize it at 2400°C for 12 seconds. Gas interrupt was employed and Deuterium arc background correction was used to correct for non-atomic absorption. Output was in absorbance units to a Perkin Elmer Model 56 strip chart recorder. If necessary, 1:10 dilutions were made to bring concentrations into an analyzable range.

Standards of 0, 1, 2, and 3 μg/l of Mn in DDW and in seawater were extracted by this procedure. Only 83% ± 2% (based on the slope and standard error of the resulting least squares calibration lines) of the added Mn in both sets of extracts was detected by the flameless atomic absorption spectrophotometric analysis. Recovery of a ^{54}Mn tracer carried through the same extraction, however, was quantitative. The apparent less than complete recovery probably results from suppression of atomization of Mn in the furnace by salts of Na, Mg, Ca and K which were not completely removed by the extraction procedure. Concentrations of field samples are therefore calculated on the assumption of 83% recovery.

Manganese also was determined on the suspended particulate material. The filters, with associated particulate matter, were digested with 0.1 ml HNO_3 and 0.1 ml HF for 16 hours at 90°C in tightly capped, cleaned polypropylene test tubes. The digests were brought up to 10 ml volume with DDW and analyzed by flameless atomic absorption spectrophotometry as described above for the filtrates.

Results and Discussion

In Figures 2a, 2b and 2c dissolved Mn concentrations are plotted for the three sampling periods, October 1974, February 1975 and April 1975. Included in Figures 2a and 2b are the concentrations of dissolved Mn flowing into the estuary from the river and ocean during the three weeks prior to the estuary transects. The flushing time of the Newport River estuary varies from 4 to 10 days depending on river flow (Jennings *et al.*, 1970). If the steady state model is to be strictly valid, input concentrations should not vary during the time period of several flushing times prior to estuary sampling. As can be seen from the figures, river input concentrations of dissolved Mn are not constant. They do not vary enough, however, to explain the dissolved Mn variations within the estuary which are discussed below.

It is obvious from Figures 2a, 2b and 2c that dissolved Mn concentrations within the estuary are not merely the result of conservative (linear) mixing of Newport River water and seawater. Dissolved Mn concentrations at intermediate salinities lie well above the line predicted for conservative mixing. (This

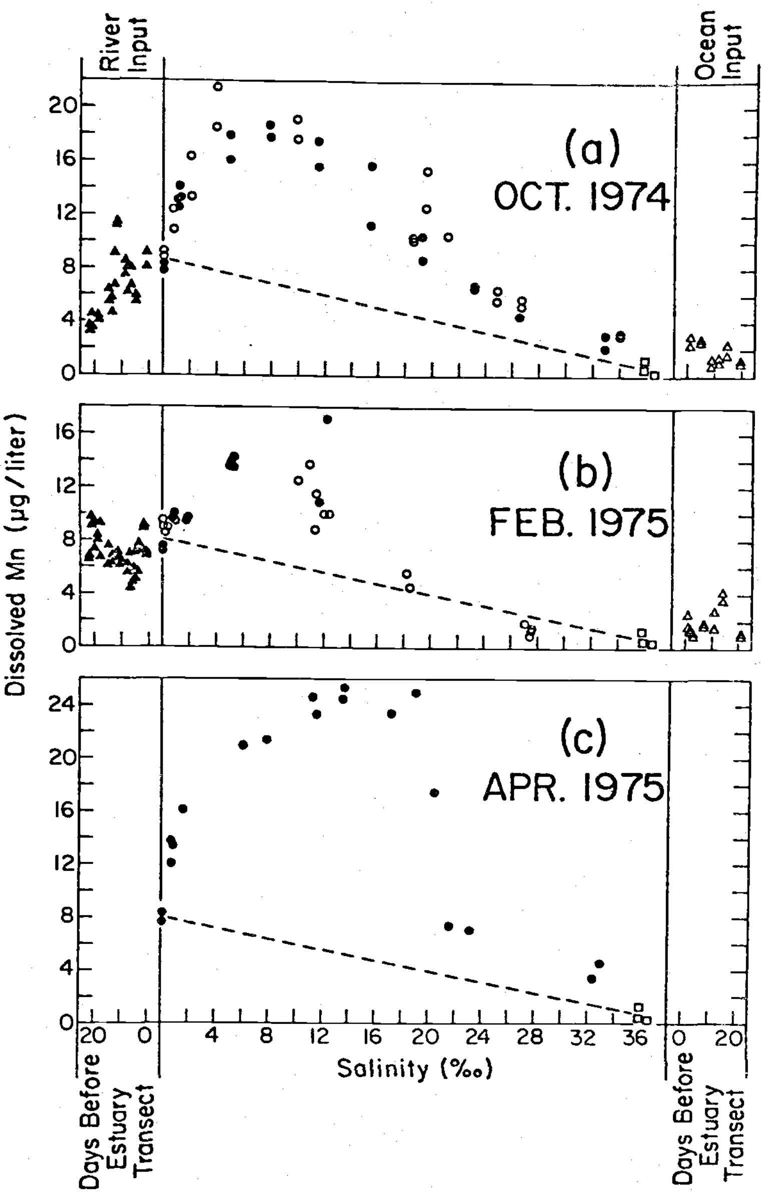

Figure 2. Dissolved manganese (µg/l) in the Newport River estuary as a function of salinity: (a) ●, October 30, 1974; o, October 31, 1974, (b) ●, February 4, 1975; o, February 5, 1975, (c) o, April 14, 1975. A dashed line connects the mean dissolved Mn concentration in the river on the days of estuarine sampling with the concentration in offshore ocean water sampled December 3, 1974 (presumably representing the oceanic water mixing in the estuary). This line represents the theoretical, conservative mixing concentrations. Also shown in (a) and (b) are the river, ▲, and ocean, △, (at AEFC dock) input concentrations during the three weeks prior to estuary sampling.

line connects the zero salinity river input on the day of the transect, and offshore ocean water, 7 km from the estuary mouth sampled December 3, 1974, which is assumed to represent a constant ocean end member in concentration.) Concentration maxima lie in the range of 4‰ to 14‰ salinity, which is the zone of initial mixing of fresh and salt water. The general similarity of the dissolved Mn/salinity relation during three disparate sampling periods strongly suggests that the nonconservation of dissolved Mn in the Newport River estuary is a general, long-term phenomenon. This, together with near identity of the relationship on consecutive days (Figures 2a and 2b), lends credence to the steady state assumption.

The dissolved Mn concentrations above those predicted by conservative mixing can only result from the addition of dissolved Mn. Because there are no obvious external sources of dissolved Mn (e.g. industrial wastes, atmospheric fallout, or other freshwater runoff) local to this area, the added Mn must result from *in situ* dissolution of particulate Mn.

In such a steady state, the only significant original supply of particulate Mn is that carried in suspension or as traction load by the river and deposited in or transported through the estuary. Kharkar et *al.*(1968) showed that clay mineral analogs of river suspended matter were capable of adsorbing certain trace metal radioisotopes in freshwater and partially desorbing them upon contact with seawater. Evans and Cutshall (1973) have shown that Columbia River suspended matter released 30% to 60% of its ^{54}Mn upon contact with seawater. Johnson et *al*. (1967) and Lentsch et *al*. (1973) have shown that bottom sediments will release large fractions of their bound ^{54}Mn and stable Mn if exposed to seawater or to salt solutions of the cations Mg, Na, and Ca which are abundant in seawater. All of these results seem explainable in terms of simple ion exchange displacement.

This process cannot, however, explain the magnitude of the dissolved Mn maxima if they represent long-term steady state phenomena. For the October 30 and 31 transects, the difference between the observed dissolved Mn concentration at the maximum at 4‰ salinity and that predicted by conservative mixing is about 13 μg/l or about 14 μg/l of river-derived water. The concentration of suspended particulate Mn flowing into the estuary from the river rarely exceeded 3 μg/l during the three weeks prior (see Figure 3). Thus, even if complete desorption of Mn from suspended particulate matter were to take place, it would supply only a small fraction of the excess dissolved Mn that must have been added. The February and April 1975 samples also show that suspended particulate matter cannot supply the added dissolved Mn. Other mechanisms must be postulated to maintain a steady state dissolved Mn maximum.

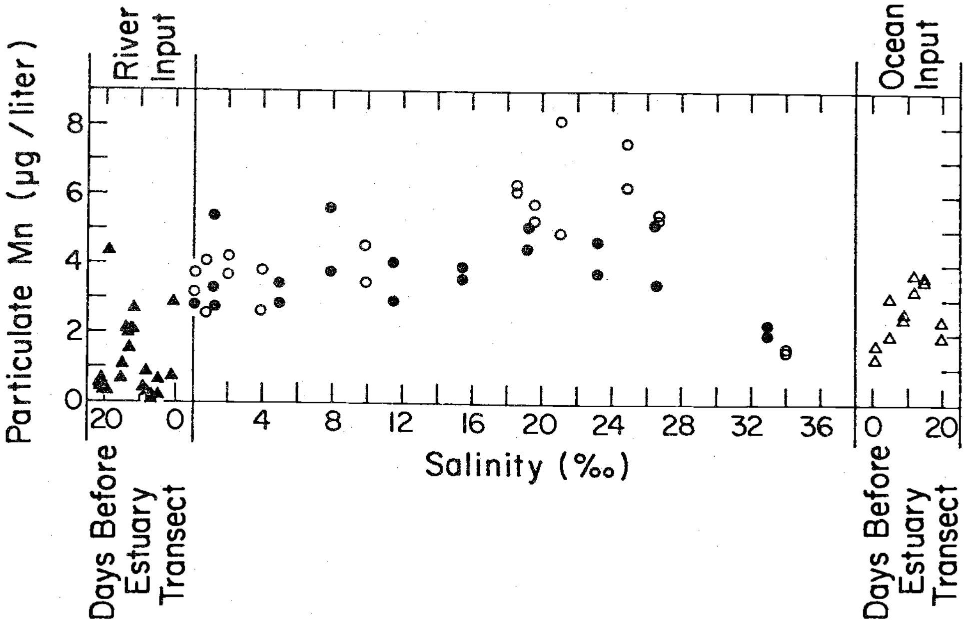

Figure 3. Suspended particulate manganese (μg/l) in the Newport River estuary October 30 (●) and 31 (o), 1974. Also shown are river input concentrations (▲) and ocean input (at AEFC dock) concentrations (Δ) during the three weeks prior to estuary sampling.

As mentioned earlier, Mn associated with bottom sediments can be desorbed by contact with seawater. Other mechanisms exist by which particle-bound Mn can be solubilized. Disruption of living cells due to changes in salinity or decomposition of Mn-containing organic matter could release dissolved Mn. Furthermore, in areas of rapid sediment deposition or slow replacement of dissolved oxygen, sediments are likely to become anoxic,and reducing conditions can develop. Thus in the sediment deposition region near Cross Rock, sediments below the top two or three centimeters are reducing. Under such conditions, oxidized Mn (as MnO_2) on particles is reduced to soluble Mn^{++} or MnS, which is then free to diffuse through the interstitial water to the overlying water. Calvert and Price (1972), among many others, have shown evidence for this process in vertical profiles of sediment Mn which is depleted at depth. Interstitial water Mn was found by them to be higher in concentration than dissolved Mn in overlying water, indicating a flux of dissolved Mn out of the sediments.

To show that bottom sediments from the Newport River estuary can act as a source of dissolved Mn to overlying waters, six sediment cores of 20 cm length and 8 cm diameter were collected with about 30 cm of overlying water on April 17, 1975, from Cross Rock. The salinity was measured at 14.0‰. Three cores were inverted, resuspending the top 5-10 cm of sediment, which was then allowed to settle for one hour. Three other cores were allowed

Table I. Dissolved manganese in water overlying sediment cores from Cross Rock April 17, 1975. Control contained no sediment. Inverted had top 5 cm of sediment mixed into the 30-cm water column. Uninverted had the core remain upright without being mixed into the water column. Contact time was one hour.

Sample	Dissolved Mn concentration (μg/l)	
Control	18.8	mean 18.6
	18.8	
	19.0	
	17.9	
Uninverted	28.8	mean 26.2
	21.1	
	28.8	
Inverted	27.4	mean 37.0
	46.9	
	36.7	

to remain upright for one hour. The overlying water was sampled in quadruplicate at the same time. Allowed to stand for one hour in coring tubes without sediment, they served as controls. The overlying water in each sample was filtered after the one hour period and analyzed for dissolved Mn as previously described. Table I shows the analytical results. For cores that were not inverted dissolved Mn increased in the overlying water by 7 μg/l, from 19 μg/l to 26 μg/l. For cores in which the sediment was resuspended dissolved Mn increased in the overlying water by 18 μg/l. In a crude way, this shows that bottom sediments can act as sources of dissolved Mn to the overlying water in this region. It is not clear from this experiment whether the mechanism is ion exchange desorption, reduction/solubilization or decomposition of organic matter, and this would require more rigorous experiments to differentiate.

It is therefore possible that bottom sediments in the region of the dissolved Mn maximum can act as a source of the excess dissolved Mn if a mechanism can be found to deposit particulate Mn there in excess of that carried in suspension in the river at the time of sampling. This could be done either by recycling particulate Mn from the lower estuary or by depositing riverborne particulate Mn in large amounts during times when the river suspended particulate Mn was not sampled (i.e. during storm runoff when riverbed sediments are scoured out and carried downstream).

Cutshall (in press) has proposed a model in which the dissolved Mn maximum is maintained by a recycling mechanism, much like the well known estuarine nutrient trap. In his model, dissolved Mn as Mn^{++} is carried downstream from the maximum by the net downstream flow of water in the estuary. In the lower estuary, it is oxidized to insoluble MnO_2. The new particulate Mn is carried upstream and deposited in the upper estuary where it is reduced and solubilized to complete the cycle. Postma (1967) discussed mechanisms for upstream flow of particulate matter in tidal estuaries which can operate counter to the net downstream flow of water,even in unstratified estuaries such as the Newport River estuary. The coincidence of the dissolved Mn maxima with the region of sediment deposition supports this model. It is entirely analogous to the model used by Spencer and Brewer (1971) to explain the vertical Mn maximum at mid-depth in the Black Sea. It differs conceptually only in being horizontal rather than vertical, with Mn reduction/solubilization occurring in the bottom sediments rather than in the water column.

It also seems possible that Mn could be biologically cycled in the same way as nutrients in the classical nutrient trap. Thus biological uptake of Mn in the lower estuary combined with release in the upper estuary might account for our observations. Our experiments do not differentiate between the chemical and biological alternatives.

If Mn is fixed in the lower estuary as predicted by this model, the concentration of Mn in the suspended matter should be higher in the lower estuary than the upper. Of course, this assumes that non-manganese particulate matter is not created in proportion. Figure 4 shows Mn per gram of suspended particulate matter for the October 1974 period. It increases with the downstream direction in the estuary and drops suddenly near the oceanic end. This is entirely consistent with the recycling model which predicts the creation of high Mn suspended particulate matter in the lower estuary which is recycled upstream.

Complementary to the increase in particulate Mn concentrations at higher salinity in the estuary, dissolved Mn should decrease. If there is a loss of Mn from solution in the lower estuary, the Mn:salinity plot should show a curvature with concavity upward. (See Boyle *et al.*, 1974 for the mathematical derivation of this conclusion.) Figures 2a, 2b, and especially 2c show such a curvature. Loss of Mn from solution does appear to be taking place in the lower Newport River estuary.

A further testable consequence of the model is that, in steady state, no more dissolved Mn should exit the estuary at the ocean end than enters at the river end. In a plot of dissolved Mn against salinity, this implies that Mn concentrations near the ocean end member should asymptotically approach the original

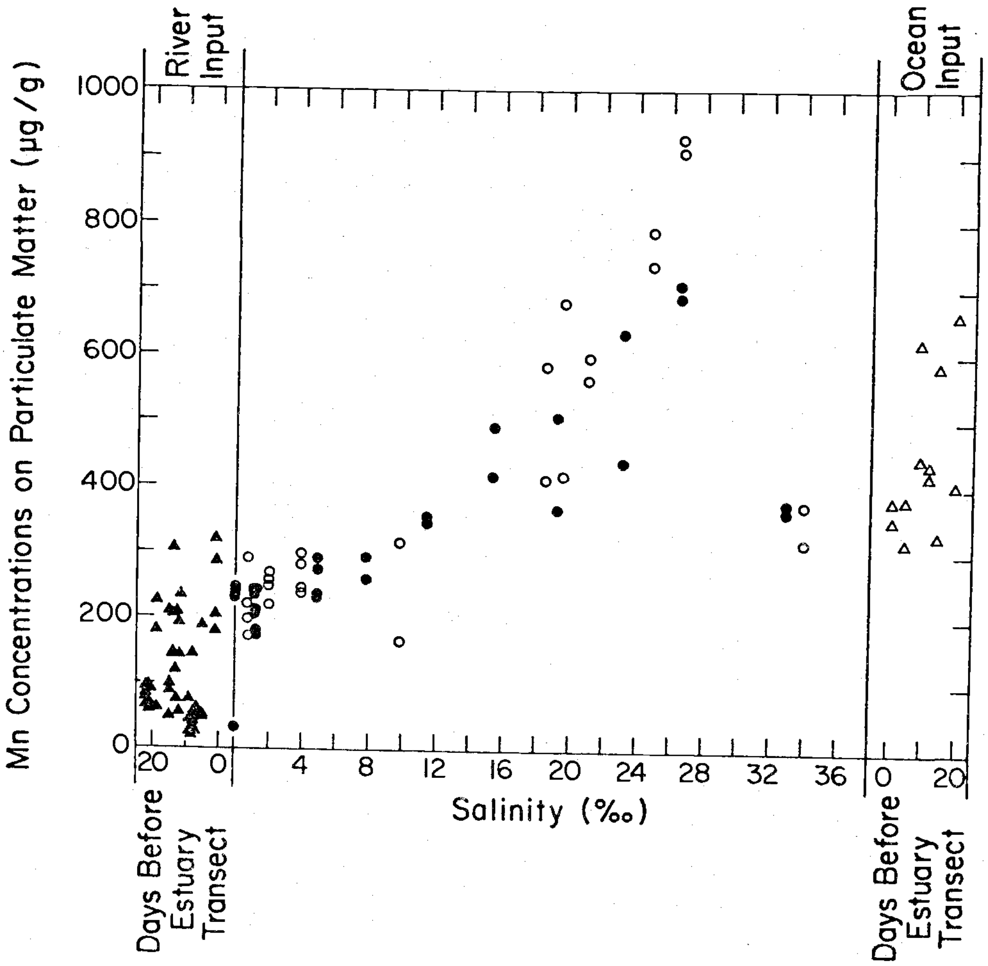

Figure 4. Manganese concentrations on suspended particulate matter (g/g) October 30 (●) and 31 (o), 1974. Also shown are river input concentrations (▲) and ocean input (at AEFC dock) concentrations (Δ) during the three weeks prior to estuary sampling.

conservative mixing curve. Both October 1974 and April 1975 transects show dissolved Mn concentrations at salinities of greater than 20‰ which do not appear to approach the original conservative mixing line, but rather to lie above it. This means that the system is exporting more dissolved Mn to the ocean than it imports from the river during part of the year.

Since the estuary seems to be exporting more dissolved Mn than it imports in solution or suspended matter from the river under quasi-steady state conditions, it is likely that occasional large inputs of particulate Mn are deposited in the upper estuary during periods of heavy river runoff. Alternatively, bed or traction loads of river borne sediments (which have not been measured) could arrive nearly continuously. Reduction/solubilization of the sedimentary Mn as described above would provide continuous inputs of dissolved Mn to the estuary waters at intermediate salinities. The postulated periodic inputs of particulate Mn have not been measured directly.

Wolfe *et al*. (1973) have estimated deposition rates of sediment in the Newport River estuary at 1 mm to 4 mm per year, basing their estimate on work by Biggs (1967) in Chesapeake Bay. The upper estimate of 4 mm per year would be representative of the Cross Rock region. If these new sediments have a water free density of 0.5 g/cm^3 and a Mn concentration we have measured by HNO_3-HF digestion at 150 μg/g, then 30 $\mu g/cm^2$ of Mn is supplied each year. If all of this Mn is solubilized, the concentration of dissolved Mn in a one-meter column of water above the sediments would be raised by 300 μg/l each year. If the estuary is flushed every six days, or 60 times a year, the water column at steady state could be raised by 5.0 μg/l of dissolved Mn. This is still far less than that observed in this region. Better estimates of sedimentation rate might refine this estimate, but since it was assumed that all deposited Mn was released to solution, it probably represents an upper limit on the estimate.

Conclusions

None of the postulated sources accounts by itself for the observed added dissolved Mn concentrations at steady state. It is probable that they all operate in conjunction. The alternative would require the postulation of a new, as yet unknown, source. The steady state assumption might be a close approximation of average conditions: however, it does not represent temporal variations, especially transient phenomena, such as pulse additions or removals during heavy river runoff. Therefore, this assumption might require modification as well. Nevertheless, there appears to be strong evidence in support of the recycling mechanism in maintaining the dissolved Mn maximum at intermediate salinities.

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Pelagic Capture of Abyssobenthic Macrourid Fish

by

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The macrourid fishes (rattails or grenadiers) are important components of deep-sea communities. They are often the dominant fishes caught in bottom trawls on continental slopes and adjoining abyssal plains (see Pearcy and Ambler, 1974 for references), and they may have key roles in the benthic food-web (Dayton and Hessler, 1972).

Most kinds of macrourids swim or hover close to the sea floor (see Marshall, 1964; Marshall and Bourne, 1964; Dayton and Hessler, 1972). Some slope-dwelling macrourids, however, are known to migrate off the bottom into the water column (Pechenik and Troyanovskii, 1970; Iwamoto and Stein, 1974). Other species are known to be bathypelagic (Marshall, 1964). Although vertical migration of abyssal macrourids has been suspected based on the occurrence of pelagic prey in their stomachs (Haedrich and Henderson, 1974; Pearcy and Ambler, 1974) this is the first direct evidence of "abyssobenthic" rattails residing several hundred meters above the bottom.

Six Isaacs-Kidd midwater trawl tows were made to maximal depths of 2000-2200 m on May 24-28, 1975, off the central Oregon coast over the Cascadia Abyssal Plain where the depth of water was 2700 to 2800 m, 8-65 km from the slope base. The net had a 5.4 m^2 mouth area with a 1-m^2 codend device that sequentially opened and closed five separate nets (see Pearcy and Mesecar, 1971). The first codend net fished from the surface to 2000 or 2200 m; subsequent codend nets fished horizontally at 2000 or 2200 m, or from 2000 m to

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1000 m in 250 m depth increments. Depth was monitored through electrical cable by a pressure sensor on the trawl. The total trawling time was 87 hours, mostly between depths of 2200 and 1000 m.

Four Coryphaenoides filifer (Gilbert, 1895), the second most common macrourid in bottom trawl collections on the abyssal plain off Oregon, were captured in the midwater trawl. They were 536-580 mm long (standard length). Two were captured in codend nets that fished horizontally at about 2000 m. The other two, both non-ripe females, were caught in nets that fished from the sea surface to 2000 or 2200 m. Based on the known depth of the trawl and the bottom topography at the time of capture, the minimum depths that these fish were swimming above the sea floor were 543 m, 725 m, 816, and 853 m. Since fish of this size readily avoid and are hardly ever captured in our small trawl, the actual number of macrourids at these depths is undoubtedly much larger than indicated by these catches.

Pearcy and Ambler (1974) found that pelagic animals were important in the diet of large macrourids. Large (500-700 mm) C. filifer caught in bottom trawls fed mainly on cephalopods, pelagic crustaceans and fishes. On a weight basis, over 75% of their stomach contents consisted of pelagic animals, including a few species common to mesopelagic waters. They proposed four possible explanations of how deep-sea fishes obtained their pelagic food; viz., (1) extensive feeding migrations into overlying waters, (2) vertical distributions of pelagic prey into near-bottom waters, (3) sinking of carcasses to the bottom where they are consumed, and (4) net feeding. Although not precluding other explanations, the capture of the four C. filifer in bathypelagic waters is support for the explanation that abyssal macrourids undertake migrations into overlying waters. The presence of red pelagic shrimp remains and a squid beak in the one uneverted stomach of these rattails is circumstantial evidence for feeding in midwater. Such feeding forays into the water column may accelerate the vertical transport of energy and elements to the deep-ocean floor.

Acknowledgments

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VERTICAL DISTRIBUTION AND MIGRATION OF OCEANIC MICRONEKTON OFF OREGON

W.G. Pearcy[1], E. Krygier[1], R. Mesecar[1] and F. Ramsey[2]

ABSTRACT

An Isaacs-Kidd midwater trawl system that opens and closes codend nets and monitors depth and water velocity was used to collect micronekton from 12 contiguous depths in the upper 1000 m. Analysis of variance of the catches of common species indicated significant vertical migrations for five of seven fishes and two of seven shrimp and mysid species. Season had little effect on the pattern of vertical migrations, average depth distributions or overall variations in abundance. None of the common species was appreciably more numerous in night than day catches; thus there was no evidence for day-night differences in avoidance of the trawl.

Diel vertical migrations, as evidenced by distinct depth separation of day and night modes of abundance, were evident for only 32% of all the species of fishes, 13% of the shrimps and mysids and 40% of the cephalopods. The range of diel migrations was generally 200 to 400 m. The four most common migrant fishes and the most abundant migrant shrimp numerically dominated upper mesopelagic waters by day and epipelagic waters by night. Lower mesopelagic waters were inhabited by a larger number of generally less abundant species which exhibited little, if any, diel vertical migration.

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The population of Stenobrachius leucopsarus, the most common fish, was composed of migratory and non-migratory individuals producing nocturnal peaks of abundance at both mid-depths and in near-surface waters. Both groups included a broad range of individual sizes. Individual fish probably do not migrate into surface waters on every night.

The depth range of all species during either day or night was large, extending over several hundred meters and several decades of irradiance. As a result, vertical ranges often overlapped broadly. Common migratory species usually occupied depths of 0-50 m at night and 300-500 m during daytime. Size segregation by depth was not a pronounced feature.

INTRODUCTION

Knowledge of vertical distributions and how they vary with time is necessary for a real understanding of the structure and function of pelagic ecosystems. Data on where organisms are in the water column are requisite to describing (a) physical, chemical and biological features of their habitat, which often change radically with depth, (b) the degree of bathymetric overlap between predators and prey and competitors, (c) the role of vertical migrants in transporting energy and materials through the water column, and (d) correlation between animal abundances and acoustical scattering layers.

Vertically migrating micronekton (small fishes, squids, shrimps, etc.) are important components of oceanic ecosystems and consume much of the zooplankton produced (Clarke, 1973). Although numerous papers have dealt with the vertical distributions and diel vertical migrations of oceanic micronekton, few have been intensive site-oriented studies that included repeated collections made in a consistent, quantitative manner with reliable opening-closing nets (Badcock, 1970; Atsatt and Seapy, 1974). Some of the most comprehensive studies to date are those of Badcock (1970), Foxton (1970a and b), Clarke (1973), Donaldson (1975), and Roper and Young (1975). These investigators used either non-closing nets or nets with inherent contamination problems. Midwater trawling studies that provide data on vertical distributions of micronekton in the northeastern Pacific include Pearcy (1964), Pearcy and Forss (1966), Taylor (1968), Paxton (1967), and DeWitt (1972). Using opening-closing nets, Pearcy and Laurs (1966) found that the common mesopelagic fishes off Oregon had similar vertical distributions, but they only sampled three broad depth intervals in the upper 1000 m.

In this paper we provide detailed information on the catches of midwater fishes, crustaceans and cephalopods obtained from multiple opening-closing net samples within 12 depth strata at one station for a series of cruises. We pose the following questions relevant to the ecology of these animals:

(1) Are depth, diel period, cruises and seasons important variables in explaining variations in abundance?

(2) Are patterns of vertical distribution and migration similar to those found by Pearcy and Laurs (1966), or are species actually segregated by depths, thereby minimizing spatial overlap?

(3) What percent of the animals migrate on a diel basis, and between what depths?

(4) Does the extent of vertical migration differ among ages or sizes of a species?

METHODS

The midwater trawl system shown in Figure 1 consisted of a 2.4 m (5.4 m^2 mouth opening) Isaacs-Kidd Midwater trawl (IKMT) with a 1 m^2 Multiple Plankton Sampler (MPS) with five codend nets. An 11 mm armored coaxial cable was used for towing the trawl system and conducting electronic commands and data. This cable terminated at a commutator swivel. A two-conductor armored cable extended along the bridle and safety lines from the swivel to a pressure housing on top of the MPS. Data on sampling depths, temperatures, and water flow through the MPS were displayed aboard ship on a frequency meter and a strip chart recorder (see Pearcy and Mesecar, 1971, for further details). The MPS nets were opened and closed on command and positive actuation of net release was confirmed by a change in frequency. Upon command from the

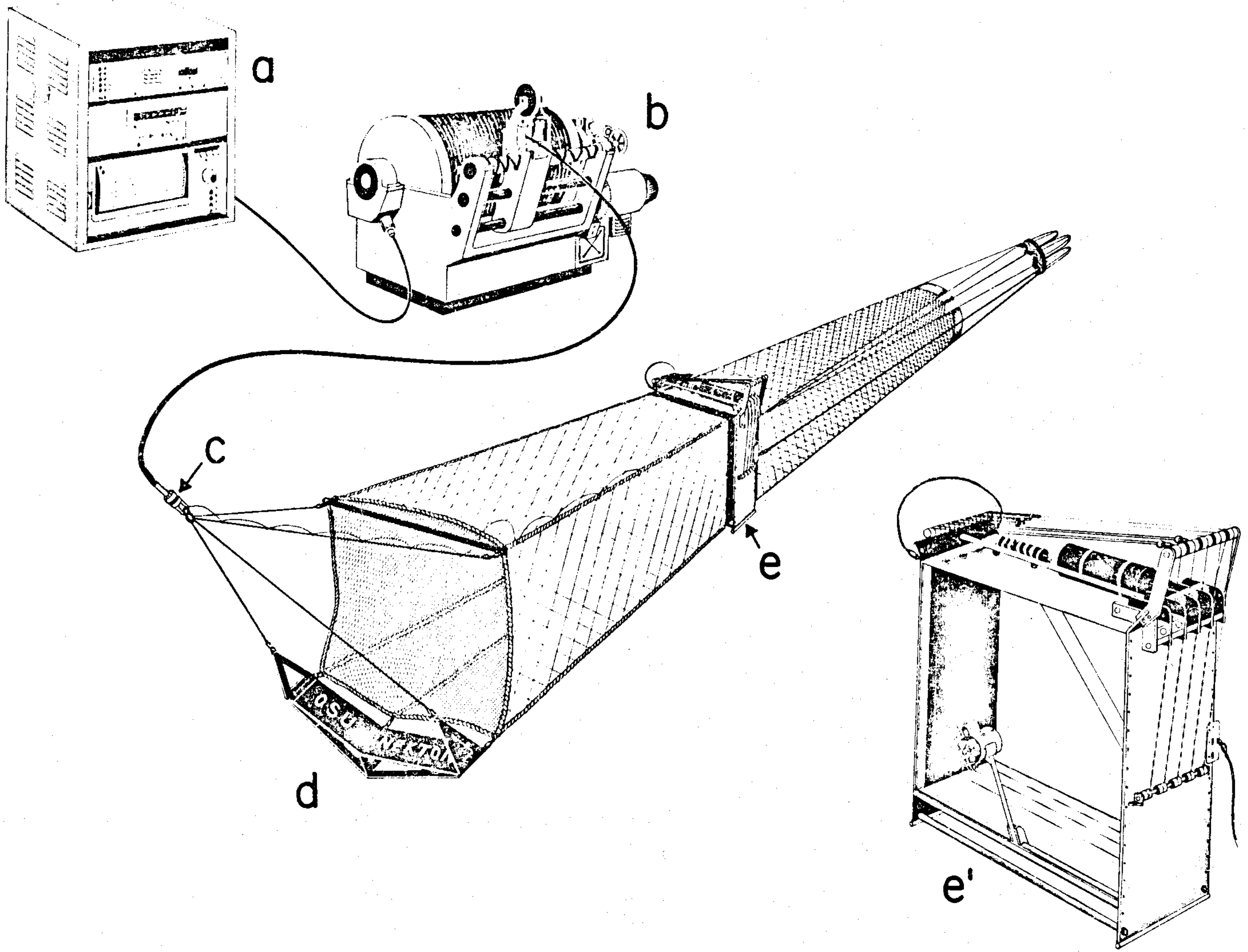

Fig. 1. Midwater trawl system components: a) shipboard data recording and command unit, b) shipboard winch with 5,000 m of 11 mm double armor coaxial conductor tow cable, c) commutator swivel, d) 2.4 m IKMT, e) five-net Multiple Plankton Sampler, e^1) enlarged view of the MPS illustrating the location of the electronic package and the flow meter.

surface an electric motor was actuated in the pressure housing which caused external shaft to rotate one revolution. This shaft was coupled to a series of five cams which sequentially released hinged bars under tension from elastic shock cords. With the release of each bar, one codend net was closed and another opened.

The IKMT net preceding the MPS was 4 m long; the five nets aft of the MPS were 4.6 m long. A 5 mm (bar measure) knotless nylon was used for the cylindrical portions of the MPS nets and for a liner in the IKMT net. The terminal 1.8 m sections of the MPS nets were made of 0.57 mm mesh nylon netting. They were conical, with 22 cm diameter collecting buckets of 0.57 mm nylon mesh. These buckets were mounted along the inside of a 0.8 m diameter aluminum ring which was connected to the MPS by safety cables (Fig. 1). The length of the safety lines were adjusted to relieve strain on the net but to prevent the net from bagging.

The MPS box (Fig. 2), constructed from 6 mm thick aluminum, was 101 cm wide, 101 cm tall and 38 cm deep. Stainless steel rods 2 cm in diameter are used for the net bars. Fully assembled, excluding nets, the MPS weighs 41 kilograms in air.

The mechanics for closing the MPS nets (see Bé, 1962) were improved by a new release system that eliminated the external arms of the rotating bars and maintained a constant tension on the wires which passed through the leading edges of both open and closed nets (Fig. 2b). These wires kept all net margins tightly closed so that water and animals could not flow into the closed nets or around the edges of the open net and cause contamination. To provide the seal needed for this large a codend unit, a 1.5 mm diameter

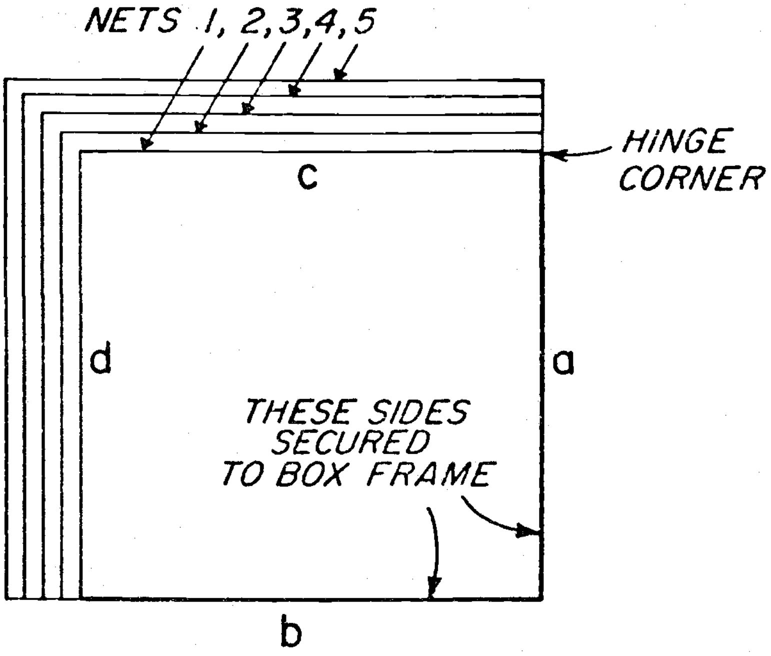

Fig. 2a. Schematic illustrations of the codend multiple net opening and closing sequence. Sides a and b are secured to sides of the MPS box. All nets on side c are sewn around pivoting solid rods. Side d is held in place by the rods and the fixed side b. When a bar rotates 90°, side c pivots to lay flat on side a and side d onto side b. The time to close one net and open the next is less than three seconds.

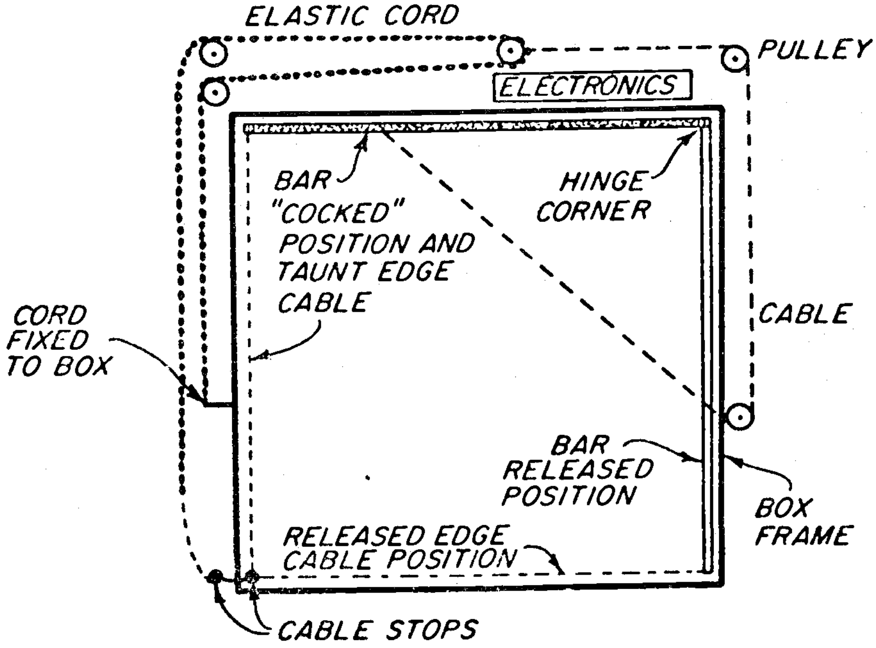

Fig. 2b. This illustration shows the incorporated use of elastic cord, pulleys and cable to open and close the codend nets and to keep their leading edges taut. The pulley above the electronic package is the only one that is not fixed in position.

steel cable was sewn into one moving edge of each net collar (identified as side d in Figure 2a); the rotating bar [side c] was the other moving edge. As shown in Figure 2b, the cable is an integral part of the net opening and closing mechanism and is tensioned by elastic cord. During the bar transition from a cocked to released position, the cable momentarily goes slack. While this is happening two small cable stops prevent the cable from changing its length. The taut cable length is self adjusting and eliminates the need to depend on the dimensions of the fabric collar remaining stable to maintain the sealed edges.

A flowmeter, with a reed switch which produced a signal every 1000 revolutions of the impeller, was mounted near the center of the MPS frame and connected by electrical cable to the electronic unit on top of the MPS. During several tows, a flowmeter was mounted on a stanchion of the trawl depressor as well as in the MPS opening. The distance recorded by the MPS flowmeter was 77.4% (s = 6.7%, n = 7) of the distance recorded by the meter in front of the mouth of the trawl. Therefore volumes filtered were calculated as $(\frac{1}{.774})$ (MPS flowmeter distance [m]) x (5.4 m^2).

A total of 342 samples were obtained on a series of six cruises between July 1971 and June 1973 at approximately 120 km off Newport, Oregon (44°40'N, 125°35'W), where the depth of the bottom is about 2800 m. Most tows were made so the trawl sampled obliquely through standard depth strata of 50 m in the upper 200 m and strata of 100 m between 200 and 1000 m. Thus 12 contiguous strata were sampled throughout the entire 1000 m column. Contiguous oblique tows may provide better estimates of abundance within a water column than non-contiguous horizontal tows since horizontal tows may sample only a fraction of the depth stratum they presumably represent.

The trawl was deployed so that it descended to below the maximum desired depth of the tow; wire was retrieved; and, as the trawl passed the lower depth of the deepest stratum, the first net was closed and the second opened. The second net was closed and the third opened at the top of the first stratum (the bottom of the second), etc., until all five nets were fished. The MPS ascended open (with no nets fishing) except for tows from 200 m to the surface when the last net was closed at the surface. In order to sample all depths equally, cable was retrieved continuously during the ascent of the trawl at about 5 m/min for 0-200 m tows and 10 m/min for deeper tows. MPS nets fished about 30 min each within the upper 200 m and about one hour below 200 m. The average volumes filtered and standard deviation for each depth are given in Table 1. Generally each depth stratum was sampled more than once during both day and night periods of each cruise. Tows on one cruise were made horizontally at one depth and were assigned to one of the 12 strata. Nets that sampled between 1 hour after sunset to 1 hour before sunrise were considered to be during nighttime; nets that sampled between sunrise and sunset were considered to be during daytime. Collections that were mainly during twilight periods (dawn-1 hr., dusk + 1 hr.) were not included in this study.

In any net where the opening-closing device is located in the codend, animals caught may be delayed in the main net and not pass immediately through the opening-closing device. If this flushing time of the net is long relative to the open-to-close time of codend nets, contamination may be serious, resulting in animals caught by the net at one time ending up in a codend net that actually fished later, possibly at a different depth. Evidence for this type of net flushing problem was noted by Foxton (1970a), Badcock (1970), and Donaldson

Table 1. Number of IKMT-MPS tows at different depths, with average and standard deviation of volumes filtered for each depth.

		Volume Filtered	
DAY	#Tows	Average	Std. Dev.
0-50	14	12.84	5.87
50-100	12	14.88	6.24
100-150	13	12.15	2.90
150-200	10	11.96	2.17
200-300	26	22.15	7.36
300-400	26	22.06	8.74
400-500	21	20.97	4.77
500-600	19	19.65	5.27
600-700	10	20.14	7.72
700-800	7	21.33	11.26
800-900	8	23.51	14.86
900-1000	8	21.61	12.37
NIGHT			
0-50	13	12.17	5.01
50-100	30	17.97	6.90
100-150	18	13.74	4.58
150-200	15	18.08	8.22
200-300	15	20.83	9.06
300-400	17	26.71	10.79
400-500	20	20.90	6.62
500-600	13	21.96	9.45
600-700	6	20.48	6.25
700-800	6	28.45	9.93
800-900	6	22.05	5.64
900-1000	6	21.55	12.47

(1975) who used IKMT's with small, codend closing devices. They found suspiciously large catches in the last nets fished and concluded that it was caused by inadequate flushing of the net.

To evaluate the flushing of the 2.4 m IKMT-1 m^2 MPS system, we used a time-actuated release device that ejected "contaminants" from a chamber by means of a plunger (Fig. 3). The contaminants were the shrimp *Pandalus jordani* and mesopelagic fishes (mainly myctophids that were marked by removal of their tails). Both types of animals were preserved and had specific gravities higher than live midwater animals. Four tows were made using this device suspended in front of the trawl mouth.

The type of tows made were: oblique tows sampling 50 m strata on the ascent; and horizontal tows at one depth (Table 2). They were different in that the codend nets 2-5 were fished short periods of time, about 15 min/net, except for the last tow listed where nets fished 40-70 minutes. On the first three tows, 96% of all contaminants were recovered in net 1 which fished during the release. In the fourth trial, the timer apparently released just before net 1 closed, as all but 5 contaminants were recovered in net 2 and none were present in nets 3, 4, or 5. A few animals were found in nets that fished an appreciable time after release of contaminants. A few were also found caught in the mesh of the main net at the end of the trawl. This was particularly evident in the first three trials where shrimp were used. Their long, serrated rostrum sometimes entangled in the netting. Based on these results, we conclude the animals can hang up in the net and later be flushed into the wrong codend net. The extent of this contamination is relatively minor, but may be significant if numbers are low or animals are prone to entangle in the netting.

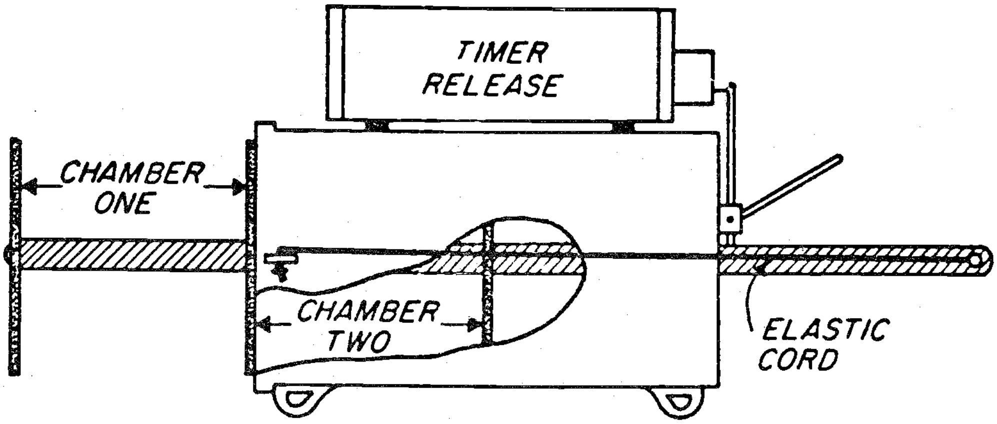

Fig. 3. This is a cut-a-way illustration of the "contaminants" release unit. It has two 8-liter chambers that are released sequentially by a time-controlled release. An electronic timer actuates a small electric motor which releases a flip bar restraining an elastic cord which in turn ejects the sample chamber from its housing. The sample chamber is mechanically restrained from moving more than its length outside the housing. The timer releases the second chamber in the same manner. Release times are selectable from minutes to hours. The release unit is 20 cm in diameter, 105 cm in length, and weighs 7 kilograms in air without a sample in it.

Table 2. Results of tests to evaluate contamination and flushing of the IKMT-MPS system by release of animals in the mouth of the net.

	Type Tow	Minutes after release that net 1 fished	No. Contaminants in Net No.:				
			1	2	3	4	5
1)	Oblique 200-0 m	16	112	1	1	0	5
2)	Oblique 250-750	18	148	0	3	0	2
3)	Horizontal	10	80	0	1	0	1
4)	Horizontal	?	5	72	0	0	0

Samples were preserved with formalin at sea. In the laboratory they were sorted into major micronektonic groups (fishes, shrimps, cephalopods) and species were identified, counted and measured (standard length [SL] for fishes, carapace length [CL] for shrimps, and dorsal mantle length [ML] for squids.

Casts were made with salinity-temperature-depth (STD) probes several times during each cruise, usually to 1000 m depth.

Using a linear regression model, an analysis of variance was performed on the data for common species of fishes, shrimps and mysids after a logarithmic transformation to log (numbers per $10^3 m^3 + 1$). The purpose of this transformation was to make the variation in the observations uniform, because variation in estimated densities increased substantially with the actual densities. One problem encountered with our data was the occurrence of zero densities for some depth and diel periods. The overall effect of this is to underestimate natural variability. And although the transformation we used helps to reduce this effect, we recognize that it by no means yields observations whose distributions conform strictly to the ANOVA model requirements. For this reason, and because of the large number of tests performed, we only considered probability levels less than 0.01 as statistically significant.

Analysis of Variance Model

The analysis of variance (ANOVA) model estimates the average transformed density, as

$$(1) \qquad \mu + \gamma_1 s + \sum_{i=1}^{2} \sum_{j=1}^{m_i} \nu_{ij} c_j^{(i)} + \gamma_2 t + \gamma_3 st + \sum_{k=1}^{11} \{\delta_k d_k + \alpha_k d_k s + \beta_k d_k t + \theta_k d_k st\}$$

We assumed that the residuals had uniform variability. The variables, $\{s, c_j^{(i)}, t, d_k\}$, are simple indicators of when and where observations were recorded. Thus s (= -1, 1) indicates season; $\{c_j^{(i)}, (i=1, 2; j=1, \ldots, m_i)\}$ indicates cruise within season; t indicates diel period (day or night); and $\{d_k, (k=1, \ldots, 11)\}$ indicates depth. (Depth level 12' is indicated by $d_k = -1$ for $k = 1, 2, \ldots, 11$.) The unknown parameters in the model measure the influences of the following factors on the average log-density.

μ = overall mean value

γ_1 = seasonal influence

ν_{ij} = effect of the j th cruise in season i

γ_2 = day-night influence

γ_3 = effect of season x day-night interaction

δ_k = abundance of an animal at depth k

α_k = season x depth interaction

β_k = day-night x depth interaction

Θ_k = depth x season x day-night interaction

In the ANOVA the term labeled "error" therefore includes unaccounted variation of two types: (a) all interactions between cruises-in-season with the other factors (day-night and depth); and (b) replication. Tows were replicated so few times on the same cruise, however, that most of the error is from (a)

Because each depth was not sampled the same number of times during each day or night period on each cruise, each sum of squares in the analysis was produced by comparing the residual sum of squares from the "full model" (equation 1) with the residual sum of squares from the fit of a "reduced" model with appropriate terms in equation 1 eliminated (Scheffe, 1959).

Equation (1) was fit to the data by ordinary least squares procedures. The estimates of the coefficients which were obtained were then used to estimate the average log-density at any required depth in any season or day-night period by suitable definition of the indicator variables. In addition, standard errors for such estimates were estimated. Estimates of the averages and corresponding 95% confidence intervals can be converted by exponentiation to estimates of average densities with appropriate confidence intervals.

We assumed that the residuals from the model equation are normally distributed and that they are homoscedastic. Neither assumption is strictly justifiable with our data and certain anomalies appear as a result, especially with respect to the confidence intervals. Thus large confidence intervals may result from a poor fit of data by the model, from the sampling pattern, or from variability in the actual data. However, the mean value estimates are robust, and the use of the model approach to the analysis produces average density estimates which are relatively free of effects from imbalance in the sampling pattern.

RESULTS

Profiles of temperature and salinity, plotted for each cruise in Fig. 4, show some of the hydrographic features of the study area. These waters are modified Subarctic, characterized by a permanent halocline in the upper 150 m and low salinities and temperatures in the upper 500 m.

The six cruises were grouped into two seasons, summer and winter, on the basis of hydrographic differences. During the June, July and September cruises strong thermoclines were present in the upper 50 m and a surface halocline usually occurred due to the presence of the Columbia River plume water. During the November and February cruises, mixed layer depths were deeper, surface temperatures were cooler and salinities were higher than

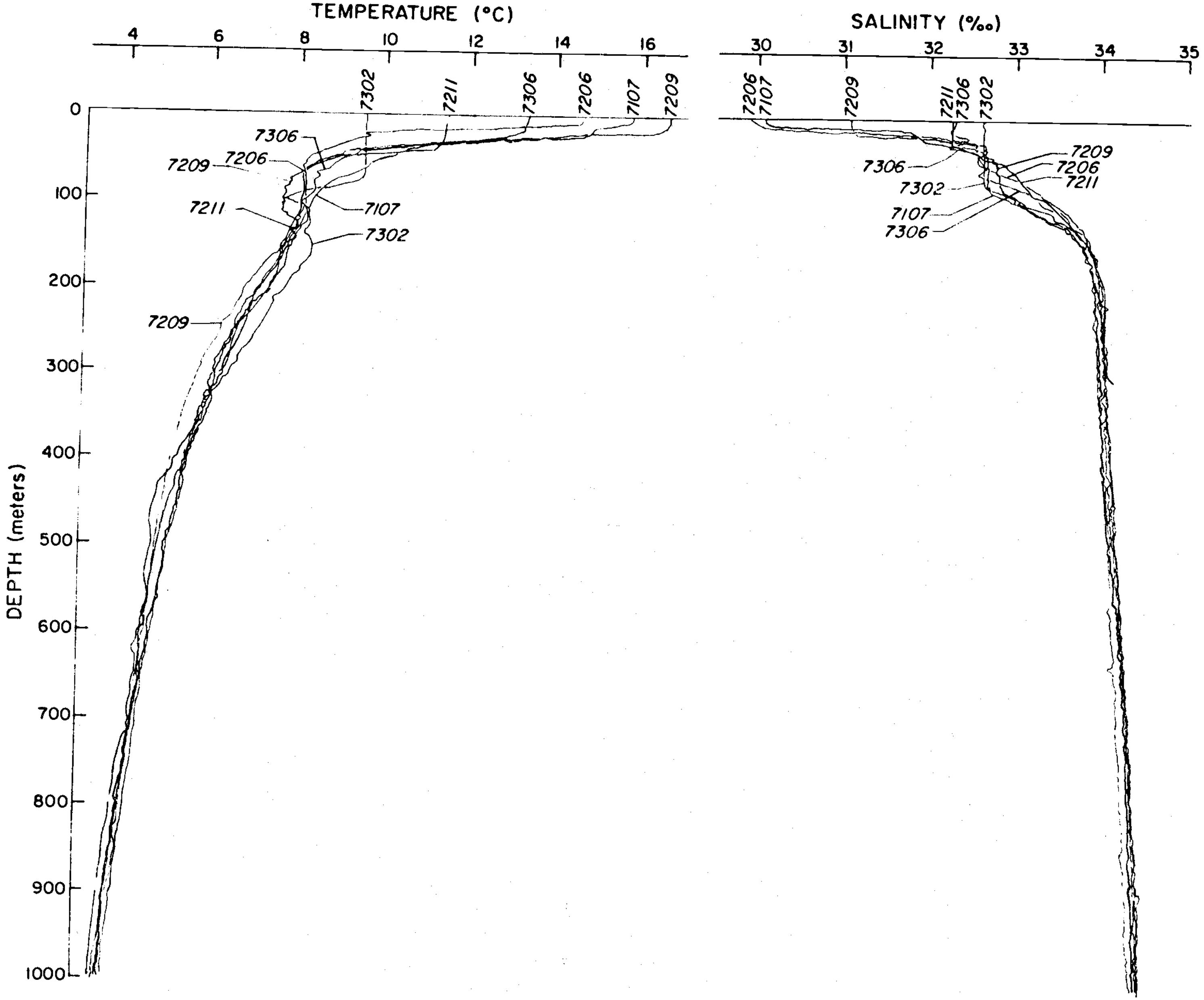

Fig. 4. Representative temperature-salinity profiles for each of the six cruises.

during the summer. The summer season is also the period of upwelling, whereas the winter is the period of downwelling off Oregon.

To illustrate the variability of temperature and salinity during a single cruise, four of seven T-S profiles for the February cruise are shown in Fig. 5. These were taken over a six day period within a 7 x 25 nautical mile area. Variations in salinity were slight. Temperatures were fairly uniform within the upper-mixed layer, but the variability between 100-300 m was large, especially at the depth of the temperature inversion. In fact, variations of temperature here were of the same magnitude as the variations among different cruises (Fig. 4).

Analysis of Variance - Common Species

The results of the analysis of variance model, summarized in Table 3, are as follows:

Season

Statistically significant ($P < 0.01$) seasonal differences were found for only the most abundant fish in our collections, a lanternfish - *Stenobrachius leucopsarus*. Calculation of average numbers per m^2 in a 1000 m column, after correcting for the differences in the depth of strata sampled between 0-200 m and 200-1000 m, showed that the catches of large (>30 mm SL) individuals of this species were almost twice as large during the summer as during the winter. Pearcy (1964) and Pearcy and Laurs (1966) also reported larger catches of mesopelagic fishes off Oregon during summer months, probably in large part due to variations of numerically dominant *S. leucopsarus*. Catches of small *S. leucopsarus* (<30 mm SL) were also larger, by about three times, during summer than winter but variability was large and differences were significant

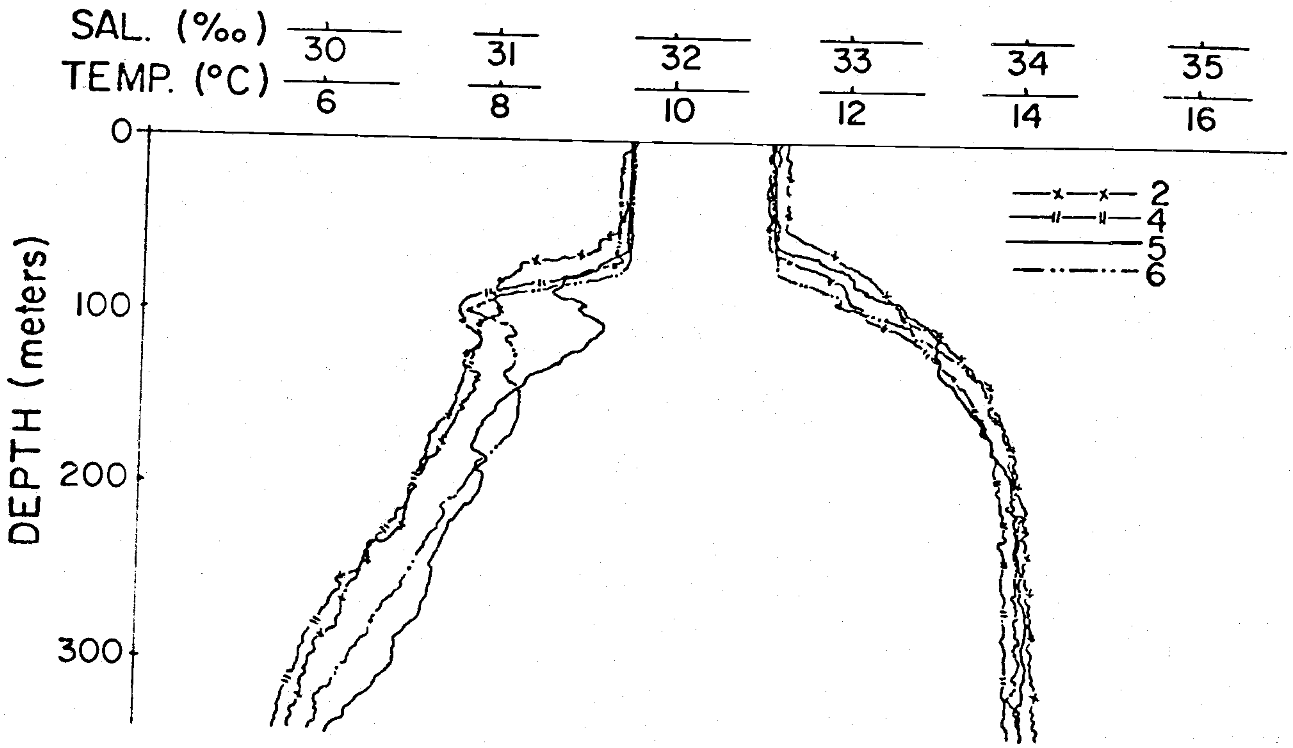

Fig. 5. Four temperature-salinity profiles from the February 1973 cruise.

Table 3. "F" values resulting from the analysis of variance for the common animals using log (catch/$10^3 m^3$ +1).

	Season	Cruises within Winter	Cruises within Summer	Diel Period	Season x Diel P.	Depth	Season x Depth	Diel P.x Depth	Season x Diel P.x Depth
A. Fishes d.f. 278									
Tarletonbeania crenularis	1.8	0.1	3.0	<0.1	0.4	2.2*	0.8	6.2***	0.4
Diaphus theta	<0.1	0.8	0.8	5.8	0.1	7.2***	0.7	11.3***	1.0
Stenobrachius leucopsarus <30 mm	5.4	0.2	4.0*	0.1	0.2	4.1***	1.6	3.9***	0.6
Stenobrachius leucopsarus >30 mm	7.4*	12.2***	1.5	3.1	0.2	5.8***	1.8	6.4***	1.4
Protomyctophum thompsoni	5.4	2.6	1.6	0.3	<0.1	11.2***	1.2	1.1	1.0
Tactostoma macropus	1.9	3.9	2.8	3.8	0.2	2.8**	1.0	5.8***	0.9
Chauliodus macouni	0.2	0.1	2.3	0.3	0.7	8.2***	0.6	0.2	0.6
Lampanyctus ritteri	<0.1	0.7	4.1*	4.9	<0.1	1.8	0.2	5.1***	0.3
B. Shrimps - mysids d.f. 249									
Sergestes similis <10 mm	12.4***	1.3	7.3	0.5	<0.1	5.0***	1.7	4.4***	0.5
Sergestes similis >10 mm	0.8	18.5***	1.7	5.7	2.5	3.6***	1.3	6.4***	1.5
Bentheogennema burkenroadi	2.1	2.0	0.7	0.9	1.4	9.7***	1.3	3.3***	1.5
Petalidium suspiriosum	27.2***	0.4	1.6	2.4	0.6	34.9***	4.5***	0.9	0.3
Hymenodora frontalis	1.1	1.8	3.5	0.9	10.4**	62.0***	2.1	1.2	1.5
Eucopia unguiculata[1]	0.6	4.0	5.6**	3.0	<0.1	86.2***	4.1***	1.8	1.2
Boreomysis californica	4.4	0.3	3.4	3.2	3.7	37.5***	1.9	1.1	2.3*
Boreomysis rostrata	<0.1	0.4	7.6***	0.9	1.0	5.2***	1.8	1.4	0.3

P <0.01* <0.005** <0.001***

[1] The taxonomy of E. unguiculata - group is uncertain. Thus identification of this mysid is tentative.

only at the 95% level. Smoker and Pearcy (1970) reported the largest proportion of small S. leucopsarus (20-25 mm SL fish) in 0-200 m night tows during the winter. This was also true in this study, despote the large catches of young during the summer.

Seasonal differences (P <0.001) were obvious in catches of two species of penaeid shrimps. Catches of Sergestes similis (<10 mm SL) were 3.5 times larger in summer than winter, mainly a result of recruitment of young shrimp. Pearcy and Forss (1966) also found largest catches of S. similis, mainly young individuals, in offshore waters during the summer. Catches of Petalidium suspiriosum were 2.3 times larger in summer than winter, possibly because a larger portion of the population inhabited depths below 1000 m in the winter.

Cruises within Seasons

The fact that cruises within a season gave significant differences in catch more frequently than cruises between the two seasons indicates that variability among cruises was just as large or larger than seasonal variations. Within-season effects were most pronounced for the decapod crustacea, especially during the summer season. Sampling variability and biological and advective changes presumably all contributed to these variations in numbers of a species collected during one season at one station. Differences among temperature profiles within one cruise (Fig. 5) are indicative of variability in physical parameters and perhaps the history of water within one area, but how significant these differences are to the distribution of the animals is unknown.

Diel Period - Avoidance

No significant day-night differences were observed for catches averaged over all depths. Thus evidence for enhanced visual avoidance of the net during daylight hours is lacking in our data. This was surprising since Pearcy and Laurs (1966) found larger night than day catches of mesopelagic fishes (as a group) and of the four common species (S. leucopsarus, D. theta, T. crenularis and T. macropus) averaged for the 0-1000 m water column in the same region. They ascribed these differences to changes in the vulnerability of animals to capture caused by enhanced visual avoidance of the trawl during daylight hours. Vertical migration as well as avoidance could explain their increased catches in the upper 150 m at night. The reason for the disparity in catches between these two studies is probably related to different methods. Pearcy and Laurs (1966) used a IKMT with a 2.9 m^2 cross-sectional area which fished at 4-5 knots while descending. We used a 5.4 m^2 IKMT which fished at about 3 knots while ascending. Neither Clarke (1973) nor Atsatt and Seapy (1974), who used 7.8 m^2 IKMT's towed at 3.5 and 2.5 knots, captured larger numbers of mesopelagic fishes at night than during day.

Furthermore, we found little evidence in this study for capture of larger animals by night than day which would be indicative of enhanced daytime avoidance. The size-frequency distributions, as exemplified by the three common migratory myctophids (Table 4) and one species of shrimp (Krygier and Wasmer, 1975), reveal no obvious day-night differences.

Season x Diel Period

This interaction, significant only for the carid shrimp Hymenodora frontalis, suggests that season of the year influences diel differences in catches. Highest catches of this shrimp were during daytime periods in the summer, but during nighttime periods in the winter.

Depth

All the other sources of variability in Table 3 not yet discussed include the effects of depth. All species of fishes (except Lampanyctus ritteri) and

Table 4. Comparisons of sizes of three myctophids caught during day and night periods, expressed as a percent of total catch for each species for each diel period. D= day, N = night.

Standard Length	*D. theta*		*S. leucopsarus*		*T. crenularis*	
	D	N	D	N	D	N
10-15	1.0	.2	3.6	.4		
15-20	1.5	1.9	10.6	4.2	7.9	4.5
20-25	3.0	3.1	4.6	6.6	26.1	41.4
25-30	4.0	4.8	18.7	11.6	28.4	17.1
30-35	8.0	7.4	16.9	14.5	11.5	8.1
35-40	11.0	8.3	6.0	10.6	7.9	1.8
40-45	14.0	18.3	4.8	7.2	6.7	2.7
45-50	13.0	20.5	5.0	5.5	3.6	3.6
50-55	16.0	10.9	5.0	7.1	5.4	5.4
55-60	17.0	16.2	4.3	5.7	3.6	6.3
60-65	6.0	5.7	4.7	7.1		4.5
65-70	3.0	1.9	3.9	6.7	2.4	1.8
70-75	.2		4.4	4.7		0.9
75-80			4.2	4.4		1.8
80-85			2.1	3.3		
85-90	.5			.1		
90-95			.1	.1		
95-100			.3	.2		
100-105			.4			
105-110			.2	.1		
Total Number	394	420	913	837	165	111

all crustacea demonstrated significant variations in abundance among depths that were independent of diel effects. We are aware that the mean effect of depth may not be as pertinent when accompanied by significant interactions of depth. Nevertheless it is obvious from Figs. 6-10 that even if day and night distributions are averaged, equal numbers would not occur at all depths sampled. Often catches of a species were low in either shallow or deep water during both day and night periods.

Season x Depth

This interaction, which is significant for the penaeid *P. suspiriosum* and the abundant mysid *Eucopia unguiculata*, indicates a change in depth distribution with seasons. Both of these animals inhabited depths mainly below 500 m. Peaks in abundance occurred at shallower depths in winter than summer (Fig. 6), possibly the result of a slight seasonal migration. Vertical distributions of two other crustaceans are shown for the two seasons in Fig. 6. Though not statistically significant, peak abundance of *Hymenodora frontalis* also appears to be deeper in the summer than winter, whereas this trend is not evident for *Boreomysis rostrata.*

Significant seasonal differences in depth distribution were absent for species that undertake diel migrations (significant diel period x depth interaction). Species migrating in large numbers into the upper 50 m at night include *S. leucopsarus*, *D. theta*, *T. macropus*, *T. crenularis* and *S. similis* (Fig. 7-9). They had highest nighttime abundance within the upper 50 m on all individual cruises. Thus the pronounced summer thermocline and halocline within this depth zone had no ascertainable influence on their depth distributions. Similarly, temperature did not appear to affect the distributions of vertically migrating sergestid shrimps off Bermuda (Donaldson, 1975).

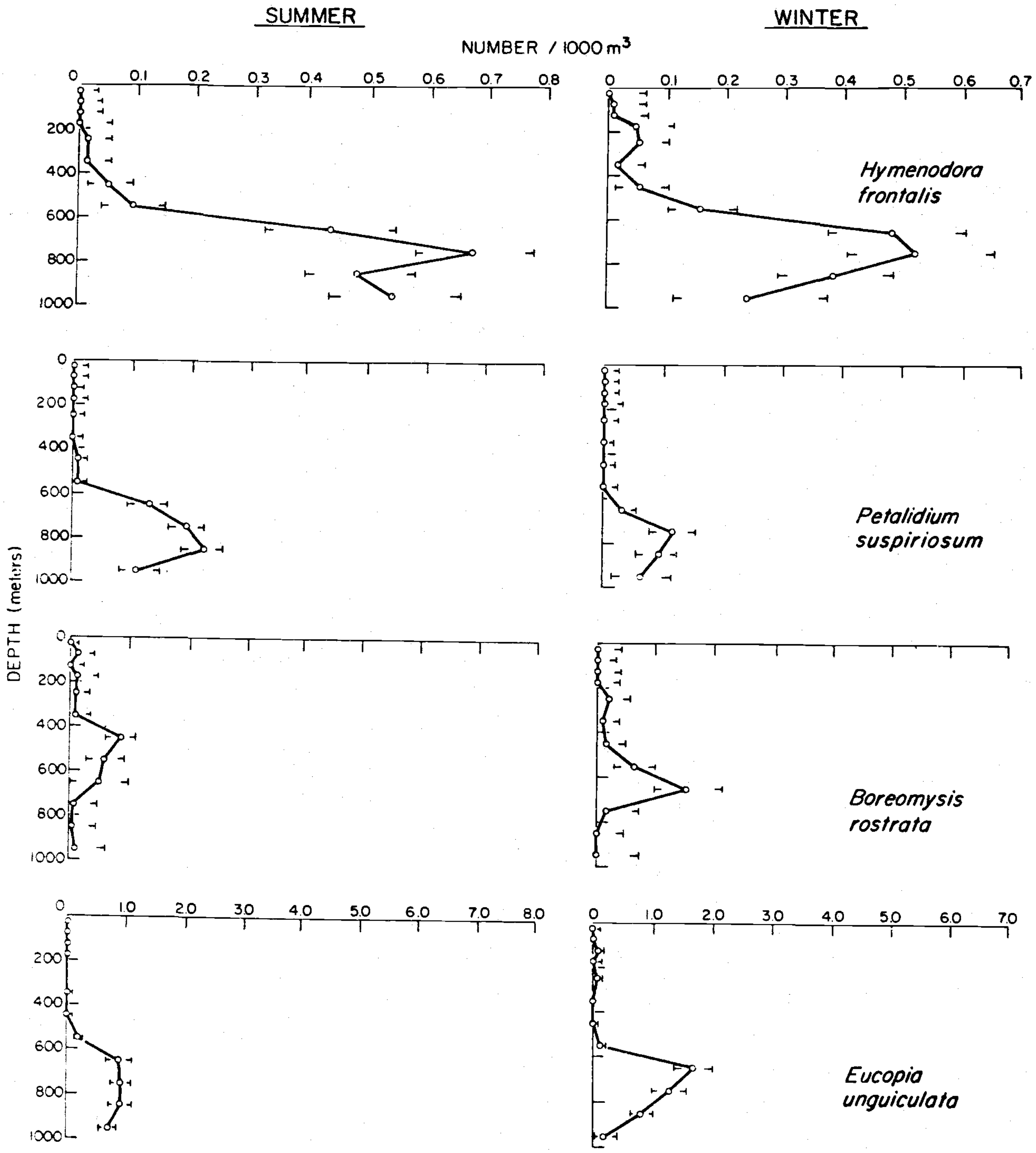

Fig. 6. Average number per 1000 m^3 ± 2 standard deviation units of some common shrimps and mysids collected at 12 depths, 0-1000 m, on the six cruises, summer vs. winter seasons.

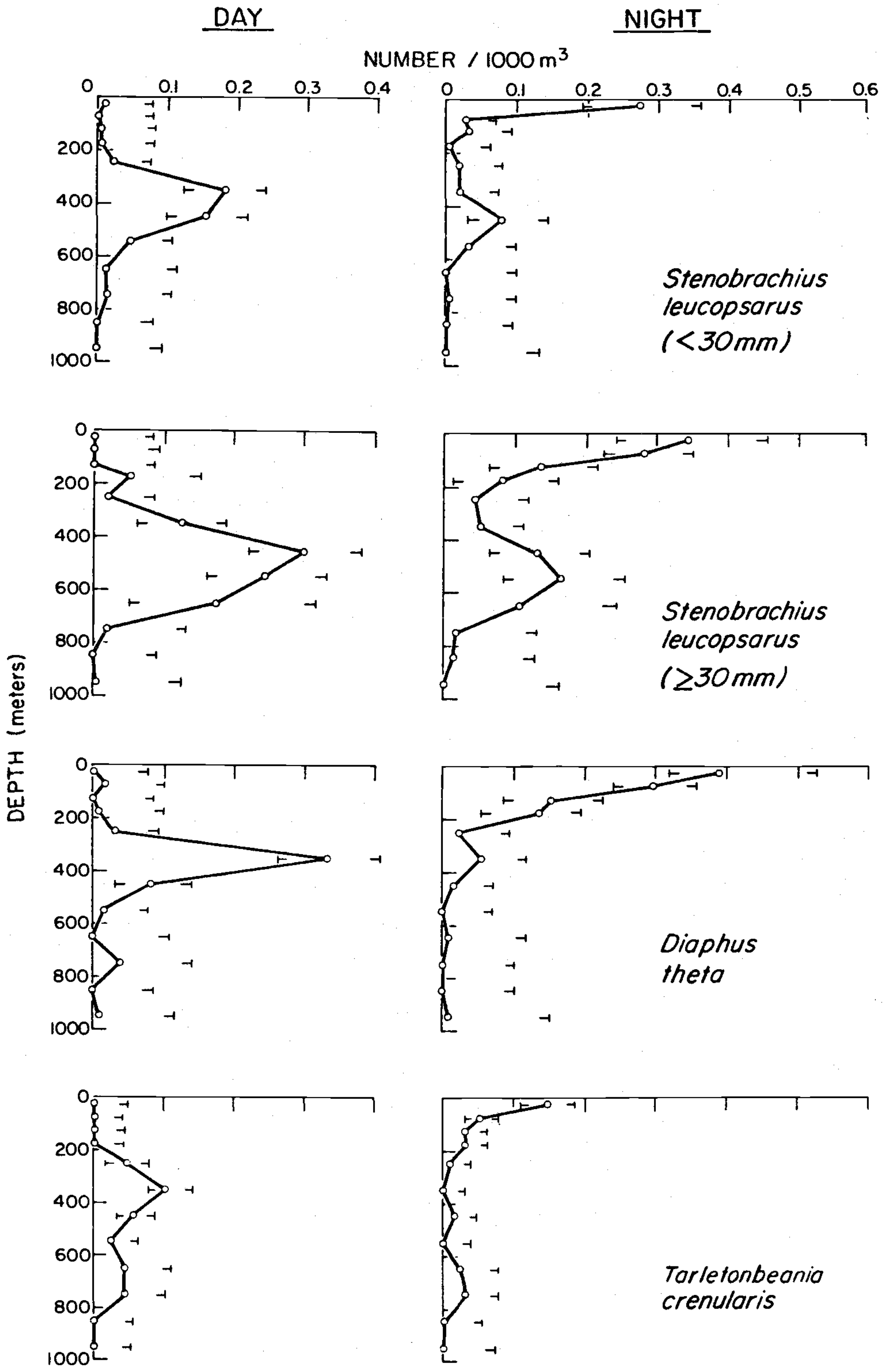

Fig. 7. Average vertical distributions of common mesopelagic fishes during day and night periods. Number per 1000 m^3 ± 2 standard deviation units.

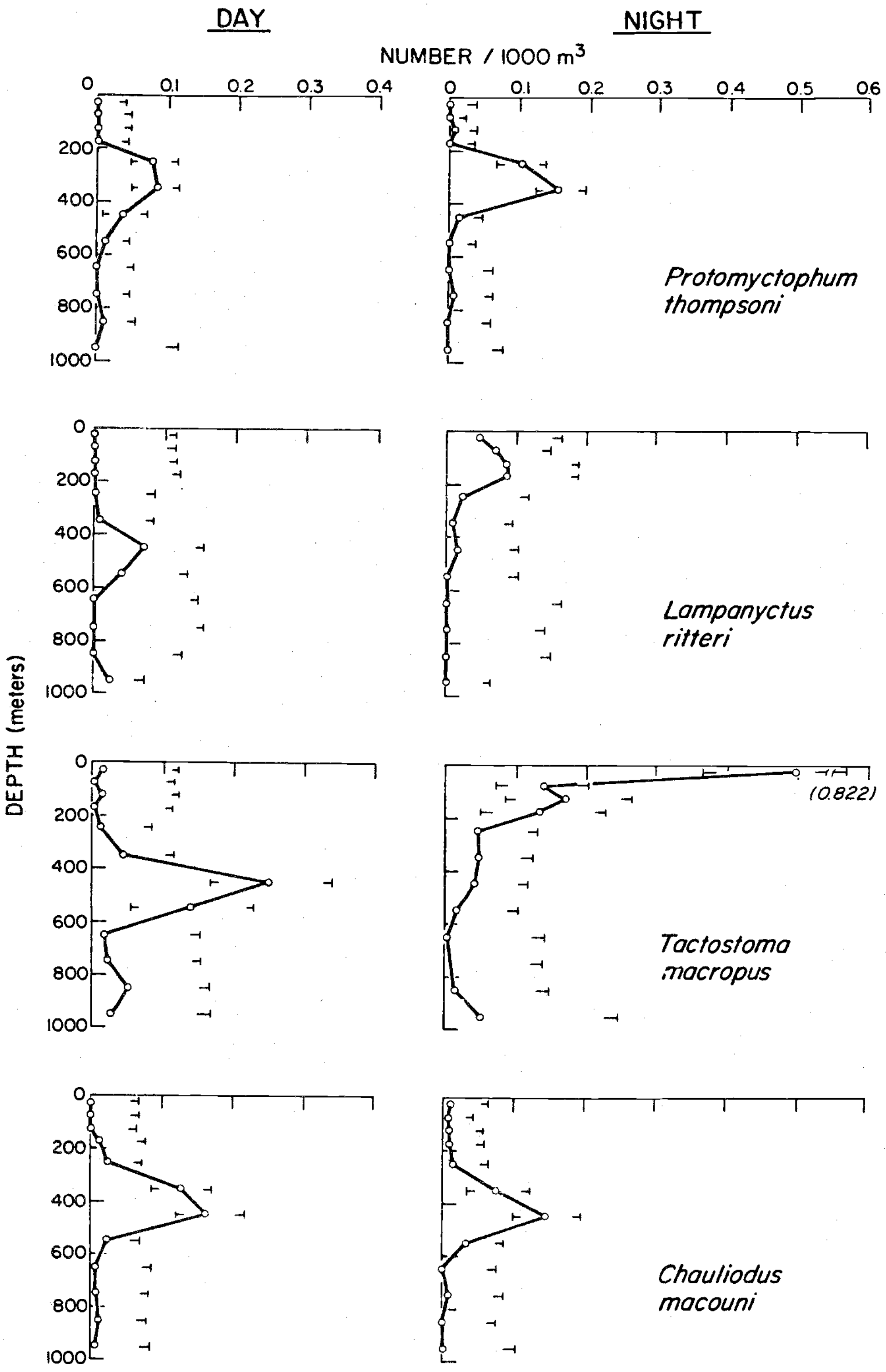

Fig. 8. Average vertical distributions of common mesopelagic fishes during day and night periods. Number per 1000 m^3 ± 2 standard deviation units.

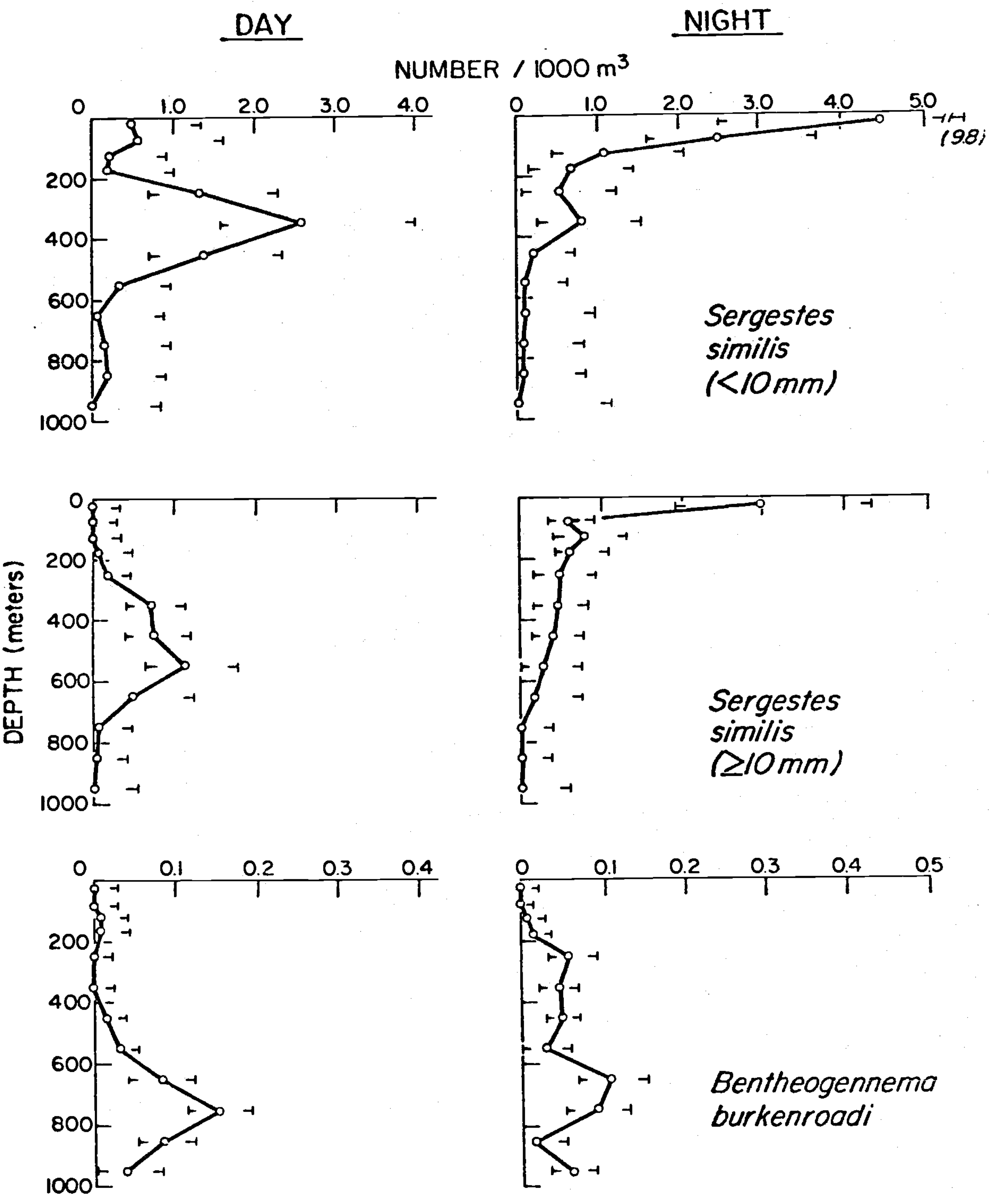

Fig. 9. Average vertical distribution of common pelagic shrimps during day and night periods. Average number per 1000 m³ ± 2 standard deviation units.

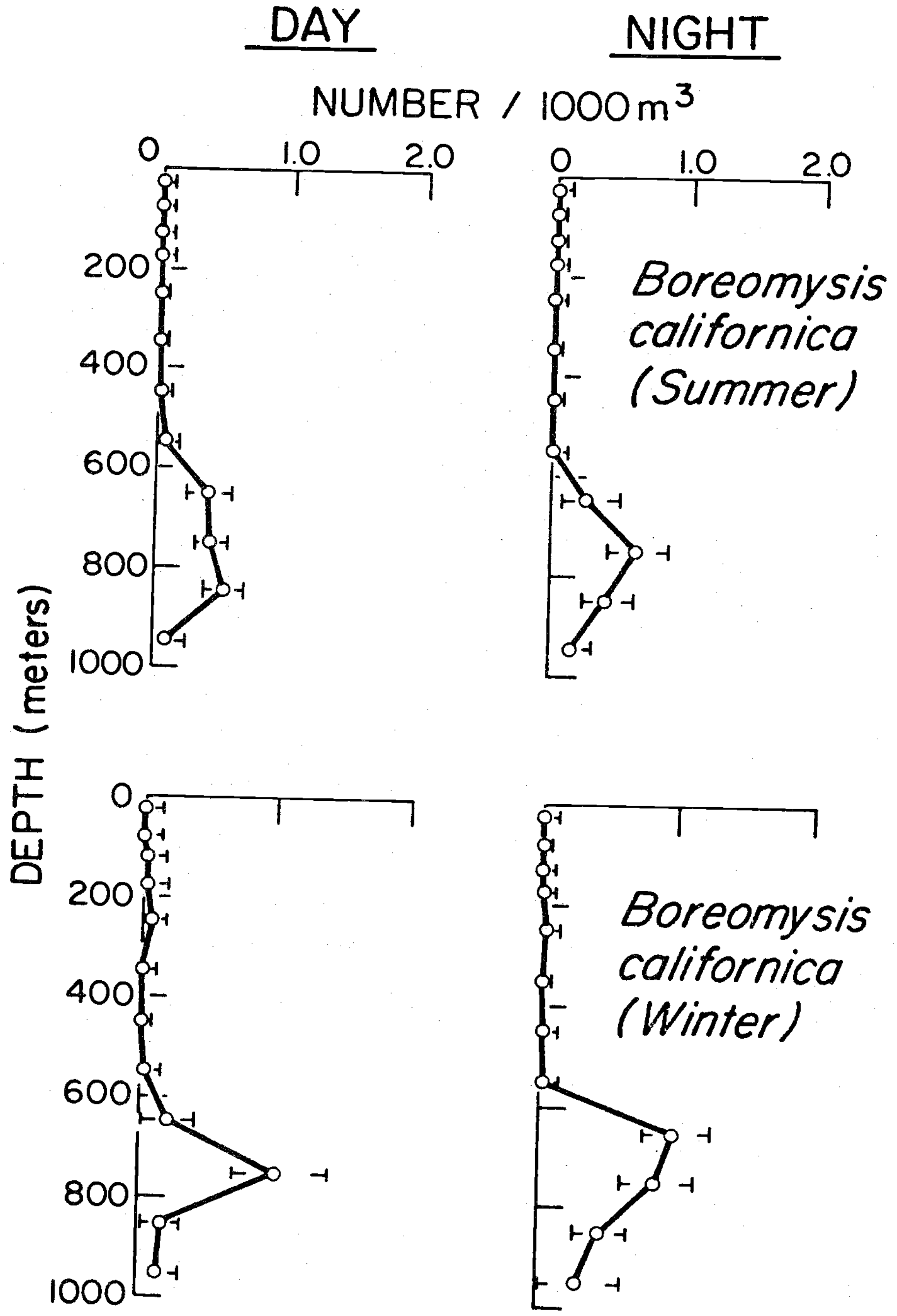

Fig. 10. Average vertical distributions of Boreomysis californica during day and night periods for summer and winter. Average number per 1000 m^3 ± 2 standard deviation units.

Diel Period x Depth

This interaction reflects the difference in vertical distribution during day and night periods; i.e., vertical migration. It was highly significant (P <0.001) for all fishes except *Protomyctophum thompsoni* and *Chauliodus macouni*. This agrees with Taylor (1968), who found little evidence of vertical migration for *P. thompsoni*, and with Pearcy and Laurs (1966).

Of the decapod crustacea, *Sergestes similis* and *Bentheogennema burkenroadi* migrated vertically (see also Pearcy and Forss, 1966; Omori, Kawamura and Aizawa, 1972; and Krygier and Wasmer, 1975). The other four common species apparently are weak migrators or non-migrators.

Season x Diel Period x Depth

This term, which measures seasonal changes in patterns of vertical migration, was significant only for *Boreomysis californica* (Fig. 10). This was the third most abundant animal in our study. Both species of *Boreomysis* were undersampled because of their small size and net escapement. *Boreomysis californica* appears to exhibit a slight vertical migration during the winter; with a peak at 700-800 m during the day and at 600-700 m during the night. Vertical migration is not evident during the summer, and this evidently obscures any trend for vertical migration from the data combined for both seasons (P >0.01, Table 3).

Vertical Distributions

The vertical distributions of each of the common species of fishes and crustaceans, shown in Figures 6 to 10, extend over a range of several hundred meters during both day and night periods. None of the species captured was confined to a single depth stratum, either day or night. Variability of the catches within a depth, as indicated by the 2 standard deviations (≐ 95% confidence interval) is large relative to the mean for most species of fishes, but is lower for the crustacea. Apparently catches of the common species of crustaceans were less variable and/or most the variability in their numbers could be explained by the ANOVA model.

Several patterns of vertical distribution and migration are apparent for these species:

I. Non-diel migrants

  A. Peaks in abundance 500 m or above (upper mesopelagic)
    Protomyctophum thompsoni
    Chauliodus macouni

  B. Peaks in abundance below 500 m (lower mesopelagic)
    Petalidium suspiriosum
    Hymenodora frontalis
    Boreomysis californica
    Eucopia unguiculata
    Boreomysis rostrata

II. Diel Migrants

  A. Peak abundance in near-surface waters (0-50 m) at night, 300-600 m by day
    Stenobrachius leucopsarus
    Diaphus theta
    Tarletonbeania crenularis
    Tactostoma macropus
    Sergestes similis

  B. Peak abundance in sub-surface waters (100-200 m) at night, 400-500 m by day
    Lampanyctus ritteri

  C. Peak abundance in lower mesopelagic waters, spreads upwards at night
    Bentheogennema burkenroadi

All the common migratory species of fishes were upper mesopelagic in distribution. Of those, all but *L. ritteri* had peak abundance in surface waters at night. With the exception of the abundant *S. similis*, the common shrimps and mysids are predominantly lower mesopelagic in distribution and, with the exception of *B. burkenroadi*, are non-migratory.

Clarke (1973) and Badcock (1970) found that size of fishes may be a function of depth, with average size usually increasing with increasing depth. Some changes in the size composition with depth were noted in our collections for both fishes and crustaceans, but distinct segregation by size was not evident. The vertical distribution of large and small *S. leucopsarus* in Figure 7 shows that both size groups occupied similarly broad depth ranges; increased abundance in near-surface waters at night was indicative of vertical migration of both groups. Small (<30 mm SL) *S. leucopsarus* were abundant at depths of 0-50 at night, but not 50-150 m, whereas large (>30 mm SL) *S. leucopsarus* were common at all depths from 0-150 m at night. The mid-depths peaks of abundance of small *S. leucopsarus* were also shallower than those for large *S. leucopsarus* during both day and night periods. The size-frequency distributions of *S. leucopsarus* captured in the surface and sub-surface modes at night were also not grossly different (Fig. 11).

Slight trends for increases in average size with depth are illustrated in Tables 5 and 6 for *D. theta* and *S. similis*.

Uncommon Species

The catches of species of fishes, large crustaceans and cephalopods that were captured less frequently (but whose total numbers were greater than 10) are listed in Table 7. Large numbers of *Cyclothone* spp., (mainly *C. signata* and *C. pacifica*) were collected, but because of their propensity to entangle in the net and the poor condition of many specimens, we did not feel that the data warranted statistical analysis. *Symbolophorus californiense*, *Bathylagus ochotensis* and *Lestidium ringens* are apparently diel vertical migrants. There is little evidence for migration of other fishes, most of which are lower mesopelagic in distribution.

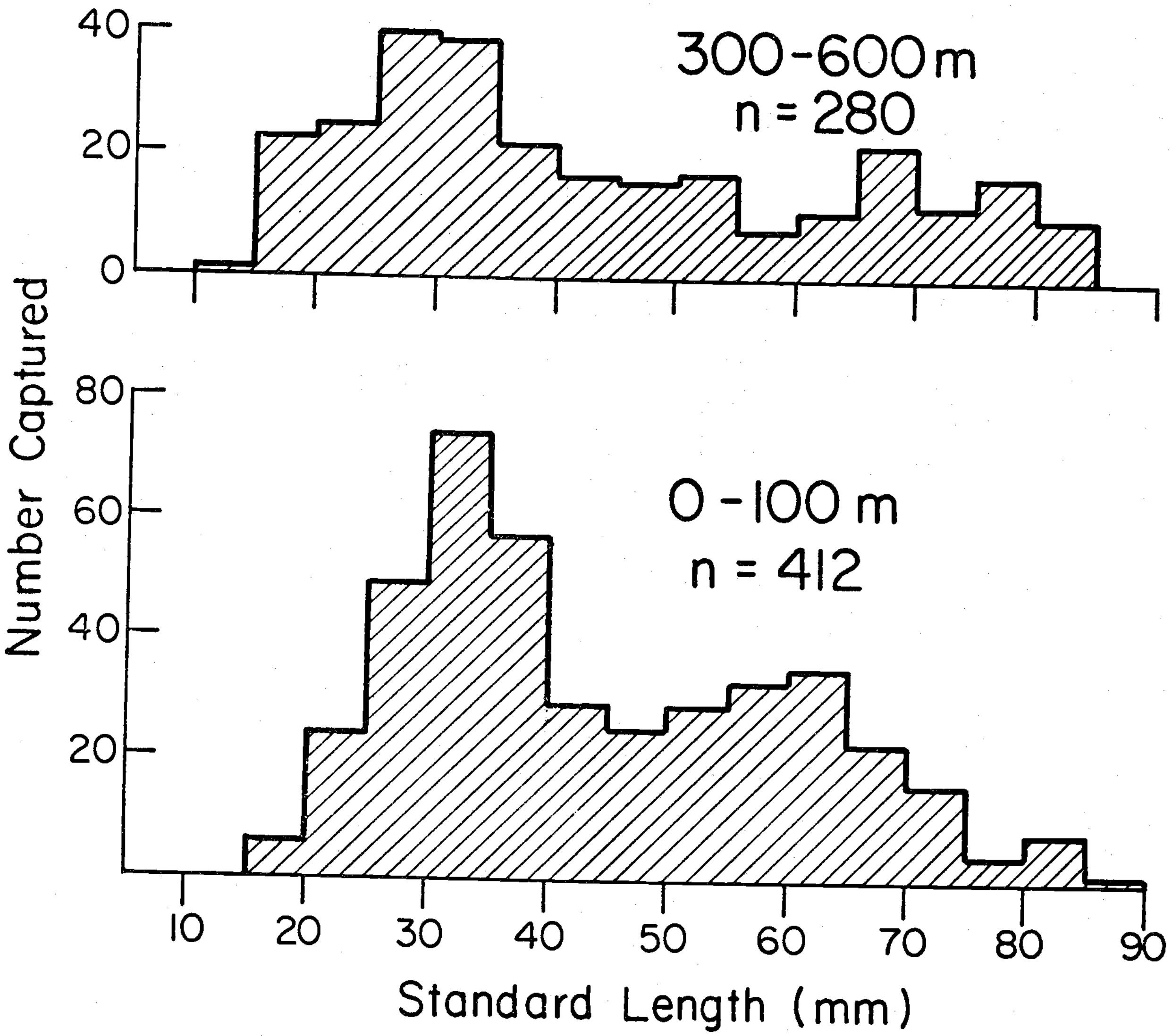

Fig. 11. Size-frequency histograms for catches of Stenobrachius leucopsarus at depths of 0-100 m and 300-600 m. All cruises.

Table 5. Numbers of different sizes of *Diaphus theta* captured at different depths during the night, all cruises.

Standard Length (mm)	5	10	15	20	25	30	35	40	45	50	55	60	65	70	75	80	85	90	95	100
0-50			4	6	15	17	16	9	9	8	15	6	3							
50-100		2	3	2	1	9	10	51	54	21	28	10	3							
100-150	1	1				1	8	11	11	9	7	2	1							
150-200			1	4	4	1	1	5	7	4	11	3								
200-300						1			1	1	3	1	1							
300-400						2			1	1	1	1								
400-500				1				1	1	1		1								
500-600																				
600-700										1										
700-800																				
800-900																				
900-1000									2		3									

Table 6. Numbers of different sizes of *Sergestes similis* captured at different depths during the day. All cruises.

Carapace Length (mm)	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
0-50	7	5	48	60	48	88	8	2		3	3		1		
50-100	2	1	38	272	275	125	25	3	1	3					
100-150		4	3	15	14	5	2	2	2				1		
150-200		5	4	5	10	14			1				1		
200-300	1	93	394	989	4318	3715	307	37	43	34	17	9	3		
300-400		9	133	702	1085	774	275	161	141	207	144	40	4		
400-500		2	21	136	280	294	152	109	100	83	62	28	17	5	1
500-600		2	24	16	30	42	22	58	133	100	76	37	34	6	1
600-700			3	13	9	6	6	7	22	28	16	23	11	8	1
700-800		1	5	20	7	7	8	1	1	8	3	2	2	2	
800-900			4	7	6	11	3	2		2	1	1			
900-1000				4	8	9	4	4	8	6	2	1			

Table 7. Numbers of uncommon fishes, shrimps, mysids and cephalopods captured at the various depths, summarized for all cruises.

	FISHES													CRUSTACEANS				CEPHALOPODS									
	Protomyctophum crockeri	Stenobrachius nannochir	Lampanyctus regalis	Symbolophorus californiense	Arygropelecus sladeni	Cyclothone signata[1]	Cyclothone pacificus[2]	Bathylagus milleri	Bathylagus pacificus	Bathylagus octotensis	Poromitra crassiceps	Benthalbella dentata	Lestidium ringens	Acanthephyra curtirostris	Systellaspis braueri	Bentheogennema borealis	Gnathophausia gigas	Gonatus pyros	Gonatus onyx	Gonatopsis borealis	Gonatus spp. (larvae)	Taonius pavo - Taonius sp.	Abraliopsis felis	Octopoteuthis deletron	Chiroteuthis calyx[3]	Japetella heathi	Octopus spp.
DAY																											
0-50	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	63	6	7	8	48	1	2
50-100	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	13	0	3	1	2	0	10	3	0
100-150	0	0	0	0	0	6	9	0	1	0	0	0	0	0	0	0	0	0	1	0	6	0	0	1	6	16	1
150-200	1	0	0	0	0	12	3	0	0	0	0	0	1	0	0	0	1	0	9	0	3	0	0	0	6	14	2
200-300	4	0	6	1	4	21	8	1	0	2	0	5	0	0	0	0	0	1	1	3	18	7	1	1	22	14	14
300-400	7	0	0	4	2	69	2	0	0	0	0	8	1	0	0	0	0	4	37	1	7	9	5	0	10	7	0
400-500	0	3	7	0	0	285	44	4	12	19	4	2	0	4	5	1	1	5	19	3	9	33	5	1	20	26	2
500-600	1	9	4	3	0	69	35	2	12	11	8	2	0	3	5	1	2	0	0	0	1	3	0	1	3	17	1
600-700	0	13	2	0	0	57	37	4	4	7	2	1	0	5	4	1	3	0	0	0	0	0	0	0	0	6	0
700-800	0	4	0	0	0	2	32	1	4	4	4	0	0	10	3	3	0	0	0	0	0	1	0	0	1	1	0
800-900	0	3	1	0	1	5	31	1	3	0	7	0	0	6	4	0	2	0	0	0	2	0	0	0	0	1	0
900-1000	0	0	1	0	1	4	17	0	1	1	2	0	0	2	5	1	0	0	0	0	0	0	0	0	1	2	0
NIGHT																											
0-50	0	0	1	3	0	0	0	0	0	40	0	0	6	0	0	0	0	1	8	1	19	2	4	5	18	2	1
50-100	0	0	1	2	0	2	3	0	0	4	0	0	2	0	0	0	0	0	14	0	7	4	1	4	69	7	1
100-150	2	0	0	0	2	2	2	1	0	5	0	0	0	0	0	1	1	5	8	1	7	6	0	2	19	11	7
150-200	3	0	0	1	6	0	2	0	0	1	0	0	1	0	0	0	0	2	4	0	6	3	1	0	10	12	4
200-300	3	0	3	1	3	4	6	5	0	3	0	5	0	0	0	0	1	4	3	1	1	4	0	0	1	13	2
300-400	2	0	3	1	0	23	3	12	3	1	0	2	0	1	0	0	0	0	1	0	1	13	0	2	1	4	1
400-500	0	0	6	0	1	191	28	4	4	2	0	2	1	0	0	1	0	0	0	0	1	2	0	0	1	6	0
500-600	0	1	2	0	0	27	8	7	3	3	1	2	0	2	0	0	1	0	0	0	1	0	0	0	0	8	0
600-700	0	6	1	0	0	14	17	0	3	0	2	1	0	3	1	2	0	0	0	0	0	0	0	0	0	3	0
700-800	1	3	2	0	0	5	24	1	3	0	4	1	0	6	7	3	0	0	0	0	1	0	0	1	0	4	0
800-900	0	1	0	0	0	1	15	0	2	0	5	1	0	3	4	0	1	0	0	0	0	0	0	0	0	3	0
900-1000	0	3	0	0	0	5	19	0	4	0	4	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0

[1] may include a few C. pseudopallida

[2] may include a few C. acclinidens and C. atraria

[3] mainly "doratopsis" stage

Using a non-closing IKMT, Pearcy (1964) reported larger numbers of *Bathylagus pacificus*, *Stenobrachius* (*Lampanyctus*) *nannochir*, *Poromitra crassiceps* and *Cyclothone microdon* (= C. pacificus) in 0-1000 m tows than 0-200 or 0-500 m tows; *Bathylagus milleri* and *Lampanyctus regalis* were about equally common in both 0-500 m and 0-1000 m tows. Our results with the opening-closing IKMT thus corroborate these earlier conclusions.

The oplophorid shrimps *Acanthephyra curtirostris* and *Systellaspis braueri* are also lower mesopelagic in distribution (see Pearcy and Forss, 1966); neither shows evidence for diel vertical migration.

Most of the cephalopods, which were mainly juvenile or larval forms, were captured in largest numbers in upper mesopelagic or epipelagic waters. Vertical migrations are suggested for *Gonatus pyros*, *Gonatus onyx*, *Gonatopsis borealis* and *Abraliopsis felis.* Roper and Young (1975) also reported that these species migrated vertically off Southern California. During the day, an interesting bimodal distribution, with highest catches near the surface and again at 200-400 m, and low catches in between, occurred for some species. At night, largest catches were generally within the upper 200 m. Most cephalopods were rarely captured in lower mesopelagic waters. An exception was *Japetella heathi*, a pelagic octopod, that was caught throughout the 1000 m water column.

The sizes of *Gonatus onyx* and *Taonius* caught (Table 8) in the upper 300 m during the day were small compared to those caught from 0-300 m at night or from 300-1000 m during the day. The same trend obtained for *Octopoteuthis deletron* and *A. felis* suggesting that small individuals are less migratory than large individuals.

Table 8. The numbers of different sizes of squids caught from 0-300 and 300-1000 m, day and night, all cruises.

	Mantle Length (mm)	5		10		15		20		25		30		35		40		45		50		55		60		65		70		75		80		85	110		115
Taonius pavo - Taonius sp.	Day 0-300		1		8		4		1		0		1		0		0		0		0		0		0		0		0		0		0			0	
	Night 0-300		4		5		4		0		0		0		0		0		0		0		0		1		0		1		0		3			1	
	Day 300-1000		0		8		23		10		0		2		0		0		0		0		2		0		0		1		0		0			0	
	Night 300-1000		0		1		8		3		2		0		0		0		0		0		0		0		0		0		0		0			0	
Gonatus onyx	Day 0-300		7		15		2		0		0		0		0		0																				
	Night 0-300		4		2		8		14		6		2		1		0																				
	Day 300-1000		0		6		19		17		11		2		0		1																				
	Night 300-1000		0		0		0		0		0		1		0		0																				

DISCUSSION

Most of the micronektonic fishes and crustaceans within the upper 1000 m off Oregon are included in two groups based on depth distribution, vertical migration and abundance. The first includes the numerically dominant species of fishes (S. leucopsarus, D. theta, T. crenularis and T. macropus which comprised 55% of all the fishes collected) and the most abundant decapod crustacean (S. similis which comprised 86% of all the shrimps and mysids). All of these species undertake extensive vertical migrations. They numerically dominate the catches within the upper 100 m at night (see also Pearcy, 1964) and within upper mesopelagic waters at depths of 300-500 m by day.

The other group consists of a large number of species of lower mesopelagic animals that demonstrate little if any vertical migration. The species in this group are in general less abundant than the migratory upper mesopelagic animals. All of the common crustaceans, except S. similis, and most of the uncommon species of fishes and shrimps belong in this category. Thus the number of species of non-migrants appears to out-number the number of species of diel vertical migrants off Oregon, while the migrants predominate on the basis of number of individuals in the upper 1000 m.

Migratory and non-migratory portions of the population of the numerically abundant S. leucopsarus are evidenced by two maxima in depth distribution at night--one in the upper 100 m and the other in the vicinity of the daytime peak of abundance. Diaphus theta and small S. similis had smaller nighttime maxima at depth, suggestive of a similar behavioral pattern. A fraction of these populations remain at daytime depths and apparently do not migrate. The fact that the residual peaks of abundance at night are deeper than the daytime peaks suggests that migratory animals reside at the shallower depths of the daytime abundance peak. It is not known whether individual fish are either migrants or non-migrants, constituting two distinct groups of the population of S. leucopsarus, or whether some individuals may migrate to the surface on some night but not others. Zusser (1958) suggested that satiated animals may not respond to the stimulus of decreasing light intensity by migrating upward at night.

The non-migrants are not simply small or juvenile fishes, as both small (<30 mm SL) and large (>30 mm SL) S. leucopsarus were found at depths of the surface and mid-depth peaks of abundance (Fig. 7). Size-frequency histograms for S. leucopsarus captured at depths of nighttime peaks in abundance at 300-600 m and 0-100 m (Fig. 11) indicate a similar size range in these two strata. Apparently a broad size range of individuals are both non-migratory and migratory.

Our documentation of two different migration patterns for S. leucopsarus confirms the results of Pearcy and Mesecar (1971). Using an opening-closing IKMT they captured small S. leucopsarus during both day and night periods within a deep-scattering layer at 350-420 m and in the upper 50 m at night. Clarke (1973) also reported that large fractions of the populations of several species of myctophids off Hawaii remained at day depths during the night. He could find no differences in the size composition, sex ratios or percentage of ripe females between migrants and non-migrants. Paxton (1967) presented data that suggested this behavior for S. leucopsarus in San Pedro Basin, California, and Tucker (1951) showed a residual nocturnal maximum at depth in the abundance of myctophids in San Diego Trough. The existence of the non-migratory population could be questioned in all of these latter studies, however, because they used non-closing nets, and large catches from nets towed mainly in deep water at night could conceivably result as the nets descended or ascended through dense concentrations of fishes in near-surface waters. Nevertheless, all these studies lend support to the idea that some mesopelagic fishes include both migratory and non-migratory individuals.

The vertical distributions of all the animals, given an adequate size sample, were broad, and encompassed most of the 1000 m water column during both day or night periods. This conclusion that species were not confined to discrete, narrow layers is not simply a result of averaging discrete vertical distributions that varied from cruise to cruise, since broad distributions occurred for individual species on individual cruises. The few effects of seasons on both depth distributions or depth and diel period interactions also support this conclusion.

It follows from the ranges of vertical distributions that individual species inhabit depths associated with a wide range of light intensities. Based on surface irradiance measurements and depth-attenuation curves (unpublished data), the modes of abundance for common species varied over several orders of magnitude of light intensity between day and night periods on individual cruises. An example of irradiance-depth envelopes is given in Fig. 12 for common fishes captured on the November 1972 cruise. During the day, peaks in abundance of _S. leucopsarus_, _D. theta_, _T. macropus_ and _S. similis_ were found at 300-400 or 500-600 m, about 10^{-3}-10^{-6} and 10^{-7}-10^{-10} $\mu W/cm^2$ respectively. All these species were most abundant in the upper 50 m at night where irradiance was 10^{-3}-10^{-5} $\mu W/cm^2$. Secondary peaks in the abundance

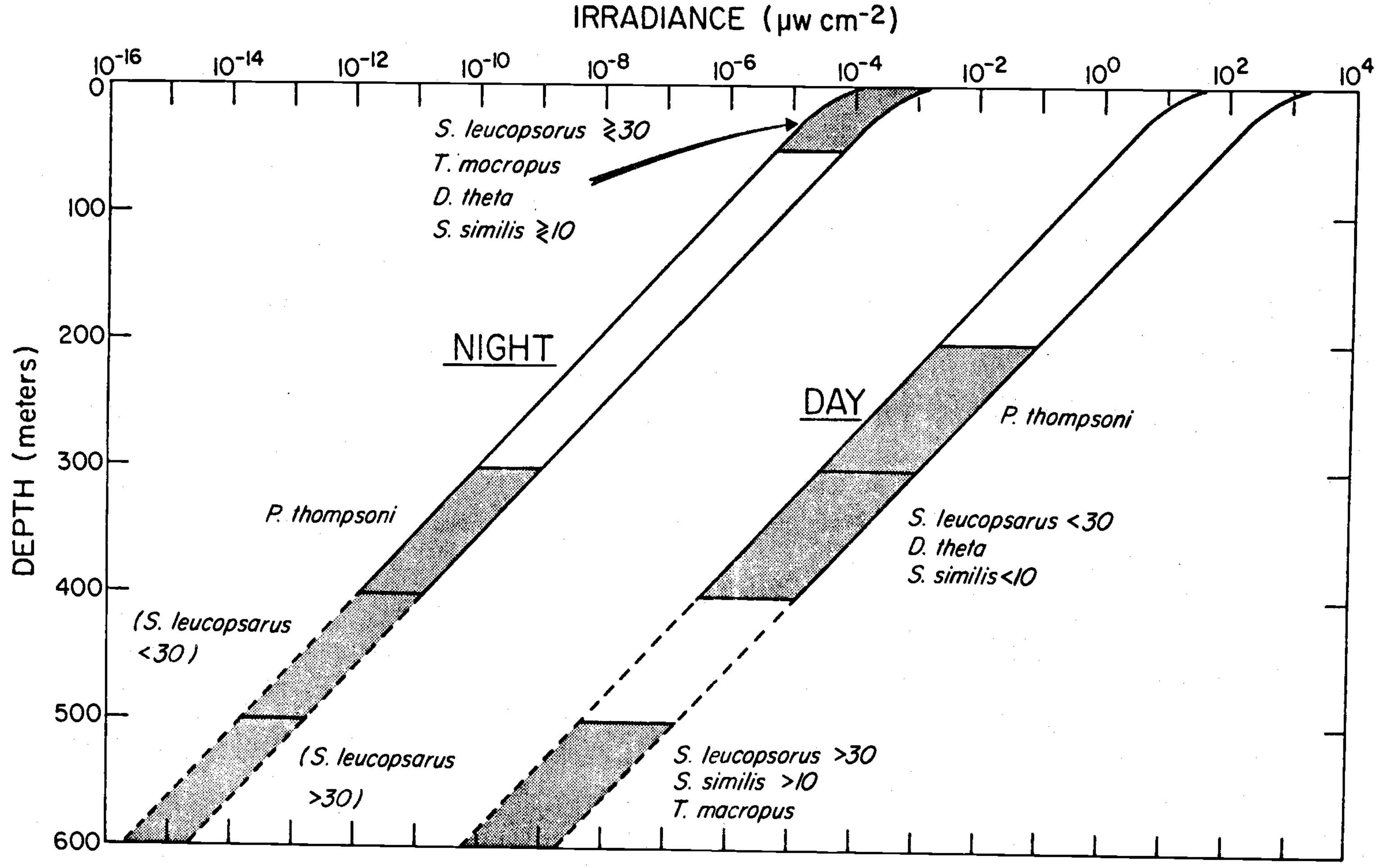

Fig. 12. Irradiance-depth envelopes for common species during the November 1972 cruise. Light intensities at depths of peak (and secondary peak) abundances are shown. The range of irradiance represents the variation encountered during this cruise.

of S. leucopsarus remained at depths of 400-500 m at night, at intensities far below those experienced during the day, and 10^6 to 10^{12} less than experienced by individuals that migrate into surface waters at night. Protomyctophum thompsoni had maximal abundance at intensities at least 10^5 lower at night than day. On the other hand, small S. leucopsarus, D. theta and small S. similis were about the same intensities both day and night. Therefore individual species populations, and in some instances individual animals, may inhabit grossly different photic environments. Our results do not contradict or confirm Boden and Kampa's (1967) theory of isolume following by individual species, since different individuals of a species may follow different isolumes during one diel period, or the same individual may follow different isolumes during different diel periods.

Because of the broad and common patterns of vertical distribution, extensive overlap occurs in the depth distribution of the species. Most of the strong migrants, for example, reside at 300-500 m by day and in the upper 50 or 100 m at night. Most of the lower mesopelagic species have maximal abundance at 600 or 700 m. Thus we have little evidence for clear-cut partitioning of depths within species groups. It is conceivable, but unlikely, that different species occupy different narrow depth ranges within the 50 or 100 m depth strata sampled by our gear. Pearcy (1964) has shown that the three common lanternfishes that migrate may have different distributions within the upper 100 m at night, but Tyler and Pearcy (1975) reported that the food habits of these three myctophids (S. leucopsarus, D. theta and T. crenularis) were similar and broadly overlapped. Vertical niche segregation of species that has evolved from interspecific competition is thus not obvious from our data on the midwater community of micronekton off Oregon.

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ZINC-65 SPECIFIC ACTIVITIES IN THE MIGRATORY PACIFIC HAKE *MERLUCCIUS PRODUCTUS* (AYRES)

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ABSTRACT

Pacific hake (*Merluccius productus*), migrating northward along the California coastline during summer, encounter the southwesterly flowing plume of the Columbia River. Radioactive ^{65}Zn associated with the plume waters is taken up by the migrating hake. Smaller fish reflect this contamination earlier than larger fish. Zinc-65 specific activities increase toward the north as far as 46°N, off the mouth of the Columbia River, and then decline northward along the coast of Washington.

INTRODUCTION

Zinc-65 specific activities in Pacific hake (*Merluccius productus*) vary with latitude and with the size of individual hake. Latitudinal variations are clearly related to the Columbia River plume. Other migratory fishes such as albacore (Pearcy and Osterberg, 1968) and salmon (Kujala, 1966) have been found to accumulate ^{65}Zn after entering and feeding in waters off Oregon and Washington. The extent of accumulation in these fishes depended upon many environmental and biological variables: geographic position in the Columbia River plume, time spent in the area, food habits, age, and sex, in addition to each species' propensity for zinc accumulation.

Many radioecological studies have demonstrated the widespread distribution and ubiquitous occurrence of radioactive zinc-65 (^{65}Zn) in the biota off the coast of Oregon and Washington (Osterberg, 1963; Pruter and Alverson, 1972; Carey, Pearcy, and Osterberg, 1966). Zinc-65 (and other radionuclides) were neutron induced from constituents dissolved in Columbia River water used to cool plutonium production reactors at Hanford, Washington. When the cooling waters were returned to the Columbia River, the entire river system was labelled with radioactivity. Osterberg (1963) demonstrated that this radioactivity was readily detectable in organisms collected off the coast, even hundreds of kilometers from the Columbia River mouth. Osterberg, Pattullo, and Pearcy

(1964) were able to relate the ^{65}Zn contents of euphausiids to the seaward extension of the Columbia River. They reported seasonal variations of ^{65}Zn in euphausiids which, although significant, were rather less than might have been expected from the known seasonal distribution of the Columbia River plume. Both physical and biological factors influenced the overall distribution of this radionuclide. Thus, while most of the total inventory of ^{65}Zn carried by the Columbia River into the Pacific Ocean could be accounted for in shelf sediments (Cutshall *et al.*, 1973), that portion which entered the biosphere appeared to be retained within the food chain (Osterberg, Pearcy, and Curl, 1964). This biotic influence appears to retard the downward mixing of ^{65}Zn out of the near surface zone, but once ^{65}Zn has escaped to deeper waters, its downward transport appears to be accelerated (unpublished data).

The Pacific hake is an abundant migratory fish whose biology is fairly well known (Alverson and Larkins, 1969; U.S. Fish and Wildlife Service, 1970). Apparently one population ranges from the Gulf of Alaska to the Gulf of California, with the greatest abundance concentrated between Vancouver Island in the north and Baja California in the south (Nelson and Larkins, 1970). Another nonmigratory population apparently lives in Puget Sound. Mature animals of the oceanic population (over 3-4 years of age) spawn in the winter season in the southern part of their range where most juveniles apparently remain. Adults, and a few juveniles (1- and 2- year olds), migrate northward during summer (Figure 1).

Euphausiids comprised over half of the weight of stomach contents of adult hake collected off Oregon and Washington with fishes and pandalid shrimps making up most of the remainder (Alton and Nelson, 1970). In addition to euphausiids, stomach contents include flounder, small hake, anchovies, shrimp, clams, and squid, as well as a variety of other prey (Best, 1963).

During their northward migration hake encounter the plume of the Columbia River which is carried southerly and southwesterly off the coastline of Oregon and northern California by the prevailing summer winds from the northwest (Figure 1). Euphausiids in the entire region generally contained their highest ^{65}Zn content (except for spring maximim very near the Columbia River mouth) during the summer season (Osterberg, Pattullo, and Pearcy, 1964).

Therefore, hake would be expected to gradually increase ^{65}Zn content and specific activity during their northward migration at least until they pass through the Columbia River plume. Then ^{65}Zn should decline.

PROCEDURES

Pacific hake were collected over a broad latitudinal range (32°52'N to 48°28'N) along the western U.S. coastline during the "summer" months (May-October) of 1969-

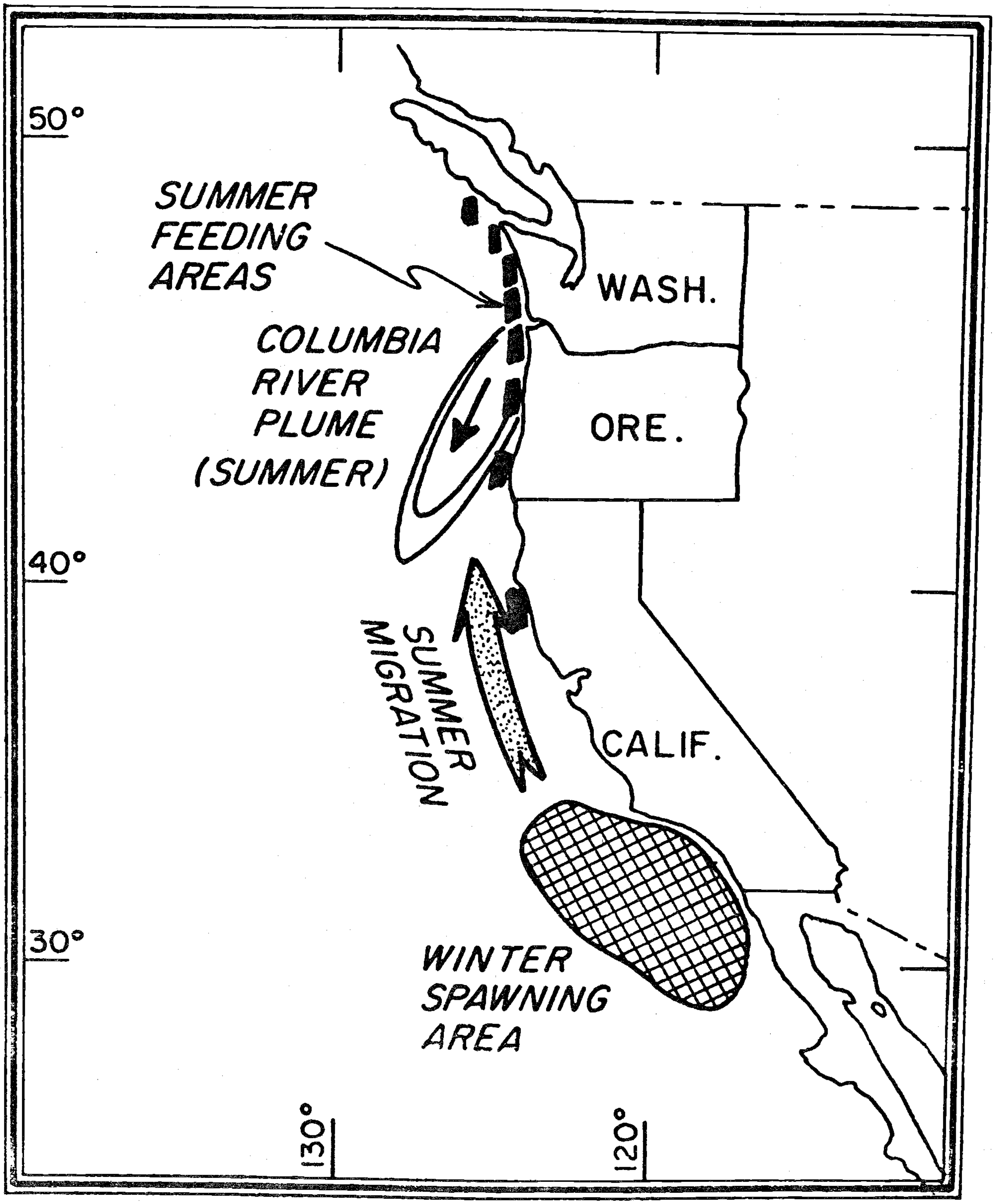

Fig. 1. Chart showing relationship of hake (Merluccius productus) migration path to Columbia River plume during summer.

1970. The great majority of fish were taken during 1969 by Soviet scientists from the research vessel PROFESSOR DERYUGIN in collaboration with the U.S. Bureau of Commercial Fisheries (now National Marine Fisheries Service, NMFS). Additional samples were collected from NMFS research vessels (JOHN N. COBB, BARON, COMMANDO) and by Oregon State University vessels (CAYUSE and YAQUINA). Russian and NMFS fish were provided to us by the Exploratory Fishing and Gear Research Base of NMFS in Seattle, Washington.

We have determined the ^{65}Zn specific activity (^{65}Zn/total Zn) of over 500 samples. By determining specific activity, as compared to ^{65}Zn concentration, the effects of variations in zinc contents of individuals is minimized. In addition the specific activity is thought to be related to turnover time of zinc in the animals (Cutshall, 1974) and we hoped to test this hypothesis.

About 44 percent of the samples were entire fish, and the remainder consisted of muscle tissue (flesh) only. For the samples where muscle only was analyzed, total wet weights were not measured. Therefore, we have determined a weight-length relationship for the whole fish samples and have used that relationship to estimate wet weights for the fish whose muscle was analyzed. The relationship calculated by linear regression was:

$$\log \text{(wet weight, g)} = -14.5 + 3.445 \log \text{(length, mm)}.$$

We checked this relationship by computing wet weights for the whole fish samples and comparing the results to the measured values. Linear regression using 228 pairs of data yielded:

computed weight, g = -18.8 + 1.03 (measured weight, g).

Pearson's correlation coefficient for the regression was 0.9820. Since the ideal regression coefficient (slope) would be 1.00 instead of 1.03, we may introduce systematic errors into the computed weight values on the order of 3 percent. This is comparable to the precision of the best of our specific activities, so that such errors are acceptable.

Fish were individually packaged in plastic bags and frozen immediately after collection. They were stored frozen. For analysis, the fish were thawed and their lengths measured. Samples to be analyzed whole were weighed and the stomach contents removed. Samples for muscle analysis were filleted. All samples were dried to constant weight in beakers at 90°C. After the samples were weighed, they were then ashed at 400°C for 40 hours to produce a grey-white ash. Gorsuch (1959) showed that loss of Zn under 600°C was negligible. The ash was packed into 13-cc plastic vials for analysis by gamma-ray spectrometry.

Gamma-ray radiations from the samples were detected using a 12.7 by 12.7 cm NaI(Tl) well-type detector and a 512-channel multichannel analyzer. Spectral data were reduced using a non-linear least squares computer program.

After gamma-ray analysis a portion of the ash was dissolved in nitric acid, evaporated to dryness and taken up in 0.36 N hydrochloric acid (HCl) for total Zn analysis by atomic absorption spectrophotometry. Total zinc analyses were made by aspirating the HCl solutions in the flame of an atomic absorption spectrophotometer which had previously been calibrated using known solutions of Zn. More thorough descriptions of analytical procedures and total listings of results have been given by Naidu (1974) and Naidu and Cutshall (1974). These same samples were also analyzed for cadmium and mercury.

RESULTS

Specific activities of ^{65}Zn are plotted as a function of total wet weight for various latitudes (Figure 2). Zinc-65 was below our limits of detection in hake collected (in the spawning area) off southern California. Even though it is not obvious in the figures, the specific activities do appear to increase slightly with increasing latitude from 32°52'N to 39°16'N, but all samples from that range are uncomfortably near our limit of detection for the radioisotope and we draw no conclusions regarding this apparent increase.

At 40°08'N latitude, however, definitely elevated ^{65}Zn specific activities are found, particularly in animals of less than 400 grams (g) wet weight. The specific activity/wet-weight plot is not linear but rather is a smooth concave-upward curve at weights

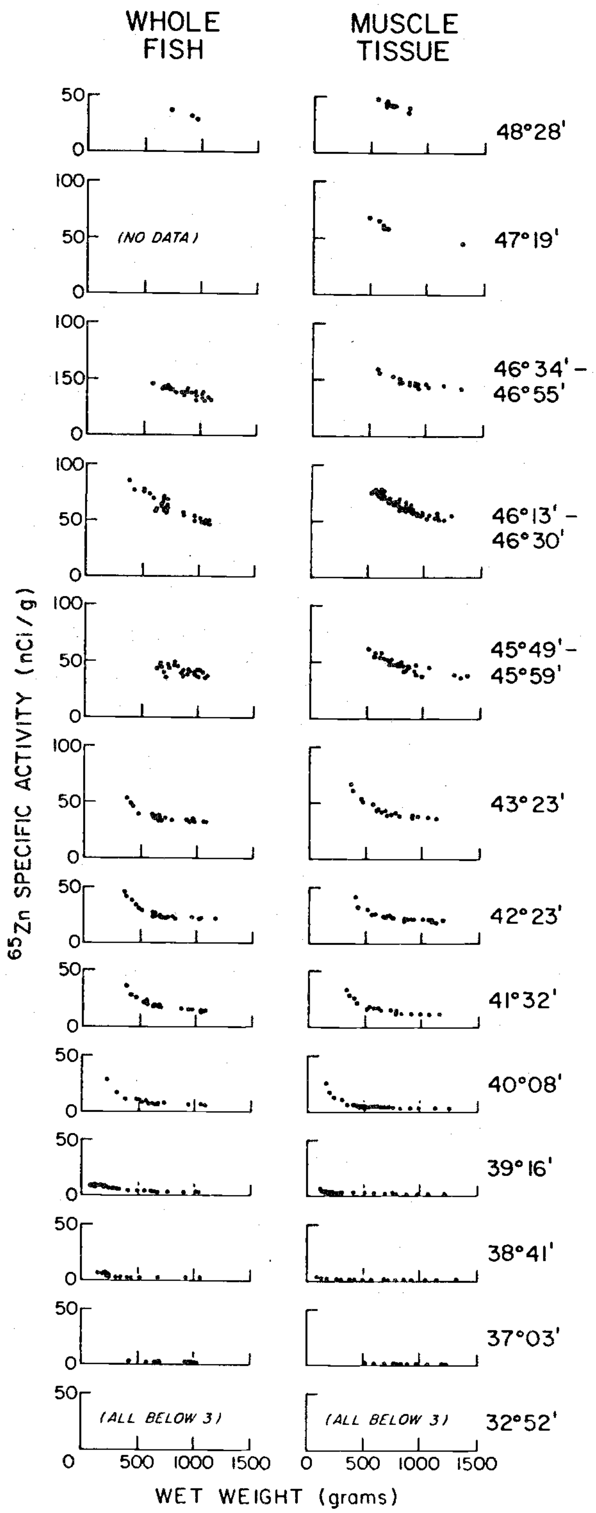

Figure 2. Specific activities of ^{65}Zn in hake (Merluccius productus) muscle and whole fish as a function of total wet weight and latitude.

below 400 g. Above 400 g little curvature is obvious. As latitude increases through 41°32'N, 42°23N, and 43°23'N, ^{65}Zn specific activities increase progressively for all body weights. The smaller fish, however, continue to increase more rapidly than the larger fish. Curvature of the specific activity/wet-weight plots involves fish of progressively greater weight at more northern sites until at 43°23'N no portion of the plot appears linear.

Off the northern Oregon and southern Washington coast (latitudes 45°49'N through 46°30'N) specific activities are even higher than at southern sites but the specific activity/wet-weight plots are only slightly concave upward. Off northern Washington (46°34'N through 48°28'N), where we unfortunately have few samples, specific activities are lower than nearer the Columbia River mouth, presumably reflecting the fact that fish from the northernmost latitudes have passed through the zone of greatest exposure to ^{65}Zn and have begun to excrete their body burden of the radionuclide. Puget Sound hake (not shown), apparently a separate non-migratory population of the Pacific hake, did not contain enough ^{65}Zn to be detected. Their specific activity was therefore less than 3 nCi/g.

DISCUSSION

Several features of the ^{65}Zn specific activity data are striking. The abrupt increase in specific activity from 39°16'N to 40°08'N by threefold in whole 250-g fish and by twofold in their muscle must reflect a substantial change in exposure to Columbia River ^{65}Zn. These latitudes also correspond to a transition in the coastline from predominantly north-south to a more northwesterly-southeasterly trend. These two locations were sampled on successive days (July 30 and 31, 1969) so that the difference must be spatial rather than temporal. Based on current measurements and return of drift bottles, surface currents flow southward off Oregon during the summer (Huyer, Pillsbury, and Smith, 1975; Wyatt, Burt, and Pattullo, 1972). Geostrophic flow at the surface is also toward the south off California (Schwartzlose and Reid, 1972). Perhaps Columbia River waters advected toward the south during the summer are transported progressively farther offshore by the offshore component of upwelled waters. This combined with the change from a northwest-southeast coastline south of Cape Mendocino to a predominantly north-south coastline north of Cape Mendocino may transport Columbia River waters beyond the shelf. Then, the hake migrating northward along the California coastline might be exposed only to much diluted Columbia River waters which have broken off the California Current (Young and Folsom, 1973). In the vicinity of Cape Mendocino (about 40°N), however, they may encounter the less dilute waters of the Columbia River plume and their specific activities would rise as is seen.

Enhanced specific activities are most obvious in small hake, producing the sharply curved line of data points in Figure 2. As specific activities generally increase with latitude, the plots become less curved, suggesting progressive changes in the rate of accumulation with size of fish and latitude. Smaller hake would be expected to reflect the newly encountered radionuclide sooner than larger hake because: (1) the smaller hake probably have a higher zinc turnover rate; and (2) the total content of zinc in smaller hake is lower (Naidu, 1974) and uptake of a given amount of ^{65}Zn has a greater impact on the specific activity of the animal.

This same reasoning pertains to the northernmost samples where uptake of ^{65}Zn is apparently exceeded by loss and the specific activity declines. It would be expected that smaller hake would decline in specific activity sooner than larger hake. While we observe that the concave-upward specific activity/wet-weight relationship disappears off northern Washington, no concave downward curves are seen. Relatively few northern samples were analyzed, however, and practically no small hake are taken at the northern end of the range, so that our data are not particularly well suited for observing the suspected more rapid loss of ^{65}Zn in smaller hake.

Specific activities in muscle tissue are consistently higher than specific activities in whole fish. In part this may result from the fact that concentrations of total zinc (3-4 μg/g wet) in muscle are substantially lower than zinc in the whole animals (10-12 μg/g wet). In addition, it is likely that the turnover of zinc in muscle tissue may be more rapid than the turnover in some of the organs known to contain high concentrations of zinc such as eyes, liver and kidneys, gonads, and spleen (see Table 1).

In January 1971, the last of the reactors which exposed Columbia River water to a high neutron flux and then released the contaminated product was shut down. Input of ^{65}Zn to the river virtually ceased and input into the Pacific Ocean was derived principally from the sediment inventory of the nuclide on the riverbed. Apparently very little of the riverbed ^{65}Zn was remobilized (Cutshall and Johnson, 1975) so that the Pacific Ocean received a markedly lowered supply. As would be expected, hake collected in the summers of 1971 and 1972 off northern Oregon and southern Washington were much lower in ^{65}Zn and its specific activity than samples taken in 1969-70 (Naidu, 1974). Similarly, other species of fishes collected off Oregon demonstrated as much as an order of magnitude decrease in ^{65}Zn specific activity between 1965 and 1970 (Pearcy and Vanderploeg, 1973).

Current operating regulations and practice within the nuclear industry prevent radioactive contamination on the scale previously seen in the Columbia River system. Levels released to the Columbia during this study

Table 1. ^{65}Zn concentrations and specific activities in hake (*Merluccius productus*) organs.

Tissue	^{65}Zn pCi/g ash	Total Zn µg/g ash	Specific Activity nCi ^{65}Zn/g
1. Skin	14.2	1470	9.8
2. Bones - vertebrate and skull	1.6	150	10.7
3. Liver	51.8	4290	12.1
4. Scales and mucous	11.7	700	16.1
5. Eyes	41.6	2440	17.0
6. Fins	7.8	370	20.7
7. Gills	12.2	420	29.1
8. Flesh	8.7	300	29.2
9. Heart	32.3	1030	31.2
10. GI Tract	37.8	1130	32.2
11. Gonads (ovary)	110.5	3060	36.2
12. Spleen	145.5	1580	92.5

exceeded the releases from 1976 sources by many orders of magnitude. While deleterious environmental or health effects related to those earlier releases have not been demonstrated, it seems prudent to avoid such contamination of the environment if at all possible. The Columbia River "experiment" has been illustrative, however. Not only has it been possible to use the man-made radioactivity in the system as a tracer and timer of ecological processes, but this contamination underscores an obvious fact which may otherwise be overlooked. That is, substances cast into the ocean do not disappear forever. Whereas it might have been acknowledged that the Columbia River does carry trace elements into the ocean for long distances, ^{65}Zn contamination of hake up to 650 kilometers away from the river mouth probably would not have been foreseen a few years past. The impact of a large river and its contaminants on oceanic biota can extend well beyond our capability to recognize the river output even in cases where unique tracers such as ^{65}Zn are not involved.

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CYCLING OF ^{55}Fe AND ^{65}Zn IN COLUMBIA RIVER CARP FOLLOWING REACTOR SHUTDOWN*

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Abstract

CYCLING OF ^{55}Fe AND ^{65}Zn IN COLUMBIA RIVER CARP FOLLOWING REACTOR SHUTDOWN.

The decline of ^{55}Fe and ^{65}Zn has been observed in the tissues of Columbia River carp following shutdown of the plutonium production reactors at Hanford, Washington. A model for the decrease in specific activity (activity per gram total element) is tested for carp flesh. The predictions of the model are slightly higher than the specific activities observed, probably because of stable element dilution. Rate of loss of ^{65}Zn appears to follow the order skin > flesh > liver, eggs > heart, while the rate of loss of ^{55}Fe follows the order heart > eggs > flesh > liver, underscoring the fact that each tissue may treat each element in a characteristic manner. Therefore the ability of an organism to free itself of radionuclide contamination will depend on what portion of the organism and what radionuclide is involved.

Introduction

The Columbia River served as a source of coolant water for single pass plutonium production reactors at Hanford, Washington, from 1944 to 1971. Neutron activation of stable trace elements contained in the river water resulted in many radionuclides, including ^{55}Fe and ^{65}Zn. The coolant water, following a brief holding time to allow for decay of shortlived radionuclides, was returned to the river resulting in concentrations of radio activity in the river ecosystem below Hanford [1] and in the Pacific Ocean off Oregon and Washington [2, 3, 4]. Since the shutdown of the last production reactor in January 1971, however, radioactivity in the Columbia River and adjacent Pacific Ocean has been declining. It is in this environment of declining radioactivity that our study of the cycling of ^{55}Fe and ^{65}Zn in the common carp, Cyprinius carpio, was undertaken.

In the absence of reactor effluents most of the radioactivity in the Columbia River system is found in bottom sediments and ^{55}Fe and ^{65}Zn make up most of the photon-emitting radioactivity. Robertson et al. [1] reported 64 percent of the photon-emitting radioactivity of the sediments to be from ^{55}Fe and 14 percent to be from ^{65}Zn with ^{46}Sc and ^{155}Eu, the only other major radionuclides, contributing 7 percent each. The radioactivity that enters the water column does so through resuspension of radioactive sediments or through desorption of radionuclides from sediments. The radioactivity was 70 to 90 percent in particulate form when the reactors were discharging effluent into the river and this tendency is enhanced now that reactor operation has ceased [1].

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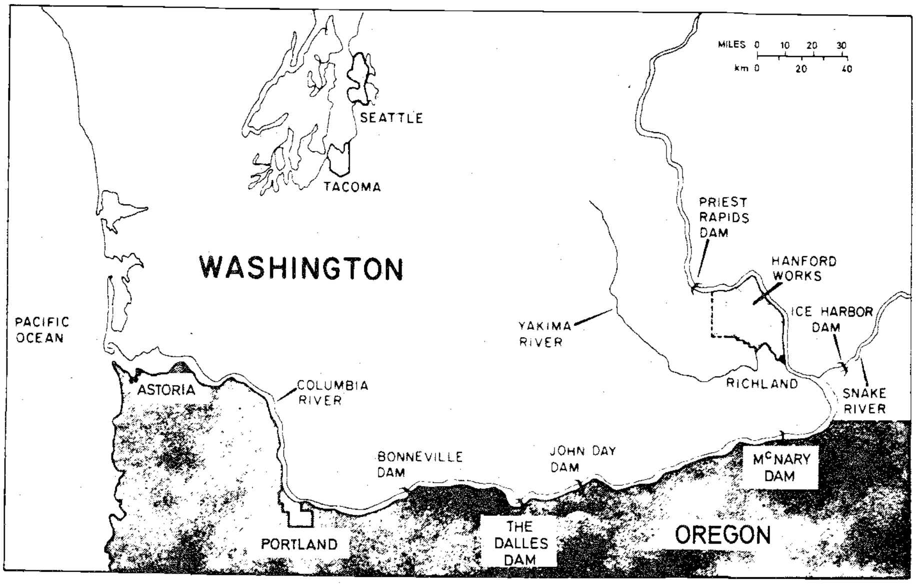

FIG. 1. Geographical relationship of the Columbia River to the Hanford Works.

Carp are well suited for studying the decline of radioactivity in the Columbia River because they live and feed in the sediments, the primary site of radioactivity. The diet of carp consists of up to 85 percent algae and detritus [5] which they get by rooting around in soft bottoms [6], sucking up mud and other materials from the bottom, ejecting it, and then selecting food when it is suspended in the water [7]. Small silt- and clay-sized particles contain a much greater concentration of radionuclides than do coarser particles [8], so carp ingest the most highly-radioactive material. Also, carp do not appear to migrate so that a series of samples from the same location should result in fish that reflect the changes in radioactivity at that location.

1. Sampling Program and Procedures

Carp collected for this study were taken from the reservoir behind McNary Dam, the first slack water downstream from Hanford (Figure 1) which contains the largest sediment deposits to have been contaminated by Hanford reactors. Collection began on July 17, 1970 and continued to May 18, 1973 with a total of seven samplings being made. After collection, the fish were placed in plastic bags and kept in a cooler with dry ice until they could be put into a freezer. Methods of collection included beach seine, gill net, and bow and arrow.

Prior to dissection, the fish were thawed overnight, washed with deionized water, and weighed. Stainless steel surgical instruments were used for dissection; new knife blades were used with each group. Separate samples of flesh and gonads were collected from each fish, while each of the remaining tissues samples was a composite of all the fish caught on the same day in order to have large enough samples for analysis.

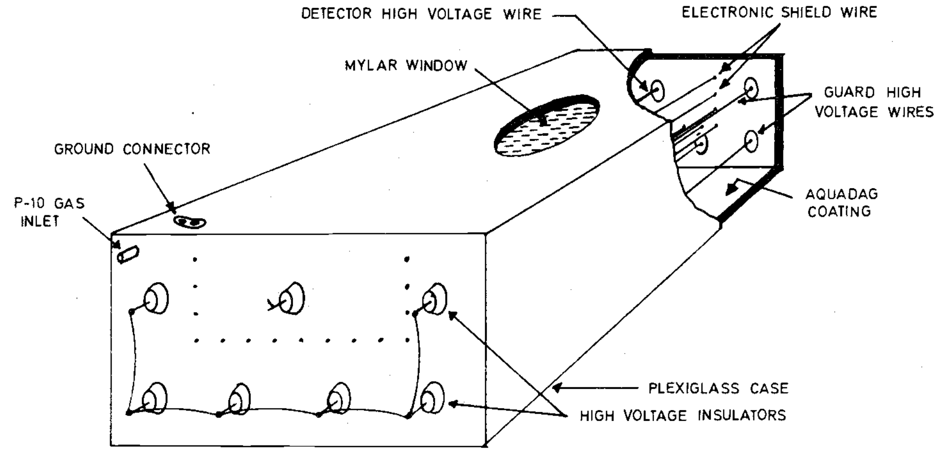

FIG. 2. Anticoincidence-shielded gas-flow proportional counter.

Samples were dried, ashed and then analyzed for ^{65}Zn on a gamma ray spectrometer. The ashed samples were dissolved in 6 M hydrochloric acid; an aliquot was removed for stable element analysis and the remainder of the sample was used to measure ^{55}Fe.

Iron-55 was measured by first extracting iron from the dissolved sample with 20% Alamine-336 in xylene, electroplating the iron by the method of Maletskos and Irvine [9], and counting the k x rays of the ^{55}Mn daughter of ^{55}Fe in an anticoincidence-shielded, gas-flow proportional counter (Figure 2). This method has been described in detail elsewhere [10].

Stable iron and zinc were measured by atomic absorption spectrophotometry.

2. A Model for Loss of Radioactivity

As a first estimate of the cycling of ^{55}Fe and ^{65}Zn in carp, we considered that a carp in steady state would have its blood in isotopic equilibrium with the environment through contact with the intestine, the site of uptake and loss of radionuclides from the environment (see Figure 3). The exchange of radionuclide with each body tissue occurs by exchange from the blood and the change in activity with time is just the uptake rate less the excretion and decay rates.

If the specific activity (activity of radionuclide (pCi)/total amount of element g) of the source is changing, as was the case in this study, the equation predicting radioactivity in carp can be found by first deriving an equation for no uptake and then modifying the equation to account for uptake from a changing source. For the condition of no uptake the change in activity for a tissue would equal losses by excretion and physical decay or symbolically:

$$\frac{dA_{T,t}}{dt} = -\beta A_{T,0} - \lambda A_{T,0} \qquad (1)$$

$$= -(\lambda + \beta) A_{T,0} \qquad (2)$$

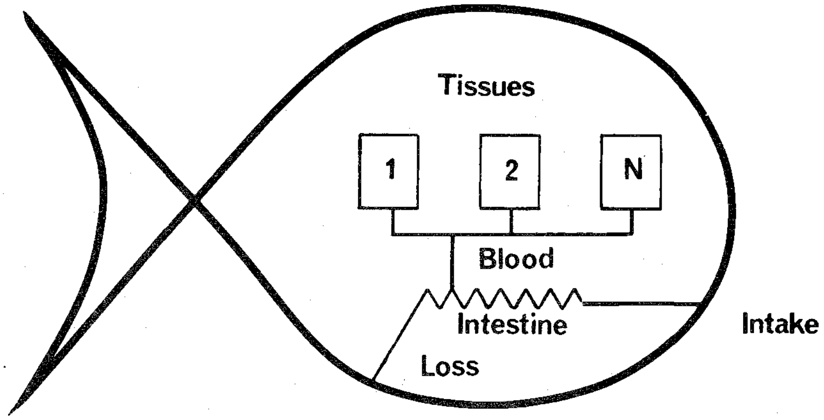

FIG. 3. Proposed model for the uptake and loss of a radionuclide from carp.

where $A_{T,t}$= activity of tissue at time t, β= turnover constant, and λ= decay constant giving

$$A_{T,t} = A_{T,0}e^{-(\lambda+\beta)t}, \text{ or} \tag{3}$$

in terms of specific activity

$$S_{T,t} = S_{T,0}e^{-(\lambda+\beta)t} \tag{4}$$

To this specific activity we must add the increment of specific activity that the tissue will gain from uptake from an environment with declining activity.

$$\text{Tissue Activity from Uptake} = \left[\frac{\text{Original Source specific activity} \times \text{Rate of radionuclide uptake}}{\text{Biological Turnover}}\right] \times \left[\text{Physical decay of radionuclide} - \text{Interaction of biological and physical decay rates}\right]$$

Or symbolically:

$$A_{T,t} = \frac{RS_{S,0}}{\beta}\, e^{-\ t} - e^{-(\lambda+\beta)t} \tag{5}$$

But since the concentration of stable element $C_T = R/\beta$,

$$S_{T,t} = S_{S,0}(e^{-\lambda t} - e^{-(\lambda+\beta)t}) \tag{6}$$

Adding equations (4) and (6) we have the specific activity of a tissue from an environment with declining activity:

$$S_{T,t} = S_{S,0}(e^{-\lambda t}-e^{-(\lambda+\beta)t}) + S_{T,0}e^{-(\lambda+\beta)t} \tag{7}$$

TABLE I. CALCULATED FLESH SPECIFIC ACTIVITIES FROM PREDICTION MODEL

$S_{s,0}$ (pCi/g)	$S_{t,0}$ (pCi/g)	$Biol_{t1/2}$ (days)	Elapsed Time (days)	Calculated $S_{t,t}$(pCi/g)	Observed $S_{t,t}$ (pCi/g)
^{65}Zn					
$1.26X10^5$	$1.22X10^5$	519	644	$2.01X10^4$	$3.79X10^3$
^{55}Fe					
$1.50X10^5$	$1.50X10^5$	516	644	$9.37X10^3$	$5.39X10^3$

In order to test whether the model can predict what actually happened in the ecosystem, the model was tested for carp flesh over a 644 day period and in the case of ^{65}Zn sediment specific activity was used as the source (sediments were assumed to be the source of radionuclides for carp). No information on the specific activity of ^{55}Fe in sediments was available, but since the specific activity of ^{65}Zn was nearly the same in flesh and sediments, it was assumed that the similarity would hold for ^{55}Fe as well.

A comparison of the calculated and observed specific activities (Table I) shows that the predictions of the model are slightly higher than the observed specific activities, about two times higher for ^{55}Fe and five times higher for ^{65}Zn. It is not surprising that the predictions of the model are slightly high because no consideration is given to the addition of stable elements through sedimentation which has the effect of diluting the radionuclides. Nevertheless, the model predicts reasonably well the specific activities of carp flesh even though errors occur because of stable element dilution, difficulties in choosing time zero, and difficulty in estimating biological turnover rates.

Loss Rates of ^{65}Zn and ^{55}Fe

Loss rates of ^{65}Zn from flesh, eggs, liver, heart, and skin are recorded in Figure 4. The order of declining specific activities suggested is:

$$\text{skin} > \text{flesh} > \text{liver, eggs} > \text{heart}$$

However, the similarity in loss rates amongst the tissues is readily apparent and, indeed, when an "F" test was performed, no significant differences were found for the loss rates ($F = .6347$, $F_{3,74}(\alpha) = 4.04$ at 95 percent confidence).[1] Because of the lack of statistical differences amongst the loss rates of the individual tissues, a common loss rate was derived with an ecological half-life of 177 days.

Furthermore, since the ecological half-lives were not significantly different from each other, a "t" test was performed to see if any of the tissue loss rates differed from the physical decay rate of ^{65}Zn. Only flesh differed significantly ($t=5.32$, $t_{35}(\alpha/2) = 3.59$ at 99 percent confidence).[2] Small numbers of samples and large variations in the activities of the samples may have masked significant differences for the other tissues.

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[1] F = calculated F-value; $F_{df}(\alpha)$ = tabulated F-value [11]; df = degrees of freedom.

[2] t = calculated t-value; $t_{df}(\alpha/2)$ = tabulated t-value, two-tailed [11].

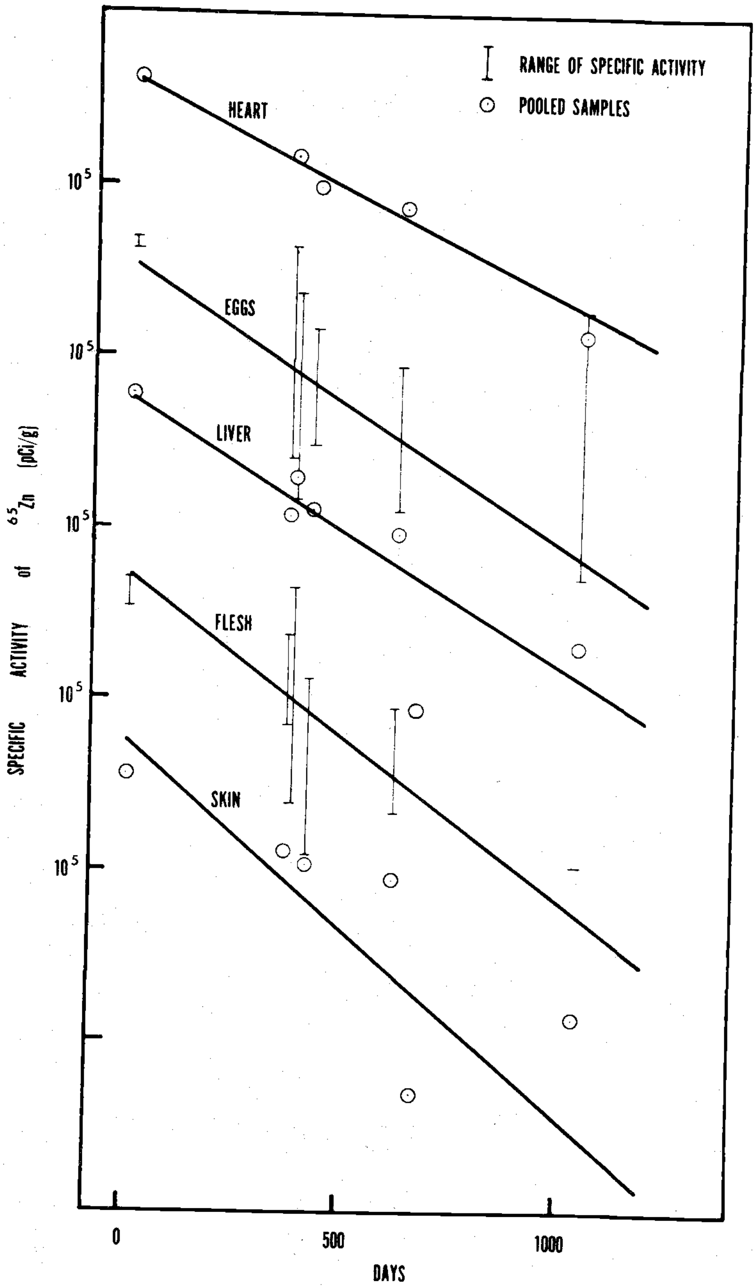

FIG. 4. Loss rates of ^{65}Zn specific activity from several tissues of carp in the Columbia River. The lines were fitted to the data by least-squares analysis.

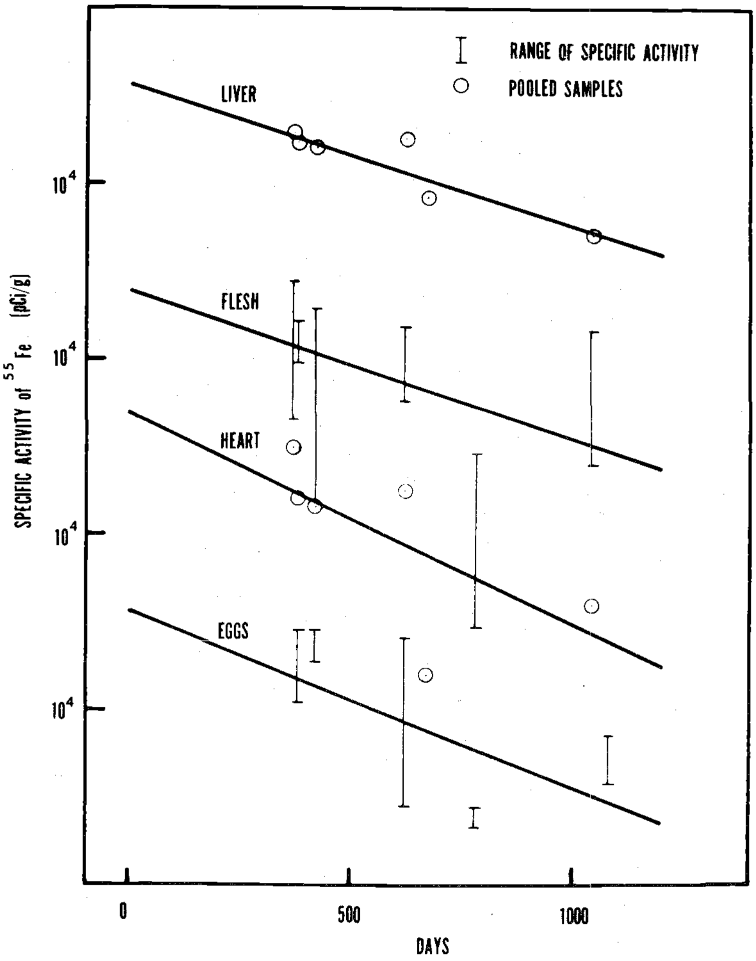

FIG. 5. Loss rates of ^{55}Fe specific activity from several tissues of carp in the Columbia River. The lines were fitted to the data by least-squares analysis.

Although no differences were noted in the loss rates of ^{65}Zn in individual tissues, differences were apparent for ^{55}Fe. The loss rates of ^{55}Fe from flesh, eggs, livers and hearts are recorded in Figure 5. The loss rates were shown to be different from each other in an "F" test (F=4.80, $F_{3,54}$( ) = 4.16 at 95 percent confidence). The data suggest the rate of decline of ^{55}Fe specific activities to follow the order:

heart > eggs > flesh > liver

The suggested order seems logical if one assumes that the liver is a storage center for iron and that the heart (which had not been drained of blood) may be rich in blood which may contain up to 60 percent of the body's iron [12]. Since blood was assumed earlier to be in isotopic equilibrium with the environment, it is reasonable to assume that the blood would respond most rapidly to an environmental change in radioactivity. The eggs would also tend to respond rapidly to an environmental change because they are produced yearly while the flesh is probably not renewed at that rate.

None of the tissues showed their ecological half-lives to be significantly different from the physical decay rate of ^{55}Fe when subjected to a "t" test. Small numbers of samples and large variances may again have masked significant differences. However, if the lack of a difference is accepted, one might conclude that the body either basically conserves and recycles iron or that the source of iron-55 was declining at the same rate as the tissues. Furthermore, there may be a storage pool, such as the liver where iron is stored but at any given time an atom of iron (stable or radioactive) may be transported to any given tissue of the body, dependent on the body needs at that moment [12, 13].

Conclusions

It is evident from this study that each tissue may treat each radionuclide and, hence, each element, in a characteristic manner, i.e., the loss rate of ^{55}Fe from carp hearts was faster than the other tissues while the loss rate of ^{65}Zn from hearts appeared slowest (see Figures 4 and 5). Therefore the ability of an organism to free itself of radionuclide contamination will depend on what portion of the organism and what radionuclide is involved.

Statistical tests for significant differences in tissue ecological half-lives indicate that:

1) The loss rates of ^{55}Fe do vary from one tissue to another. The following order is suggested:

heart > eggs > flesh > liver

The order seems reasonable when it is recalled that the hearts contained undrained blood which reacts to ecological changes most rapidly (the body fluids being in isotopic equilibrium with the source of radioactivity). Eggs, being reproduced once a season, would also be expected to react rapidly. The liver, considered to be a storage center and not subject to removal or renewal as in the case of eggs, should have the longest turnover time.

2) Zinc-65, on the other hand, showed no statistically significant difference in rates of loss between tissues. A trend, however, was suggested following the order:

skin > flesh > eggs-liver > heart

The suggested trend differs markedly from that of ^{55}Fe which points up the fact that zinc's role as an enzyme activator allows it a different mode of body usage than iron, perhaps a more general mode. On the other hand, the nonsignificance of loss rates between the tissues may have been primarily due to small numbers of samples and large variations in the observed activities, and/or radionuclide uptake by tissues with rapid turnover may have caused their ecological half-lives to appear longer and approach the half-lives of those which would have picked up smaller amounts of the element.

The model developed in the study can be used as a tool for estimating tissue radioactivity in an environment of declining radioactivity. However, complicating factors, such as radionuclide dilution by stable element addition as in sedimentation and the history of the environmental contamination itself are known to influence its reliability. Furthermore, computation of biological half-lives are necessary for the working of the model. This is a real limitation because a no-uptake situation must be assumed in the face of knowledge that uptake from a declining environment, such as the carp/sediment relationship in the Columbia River, is probably a major factor in the amount of radionuclides present in the carp.

Finally, in spite of the movement of carp which present the possibility of sampling more than one population, the carp appear to be very useful in fresh water ecological studies of this type because of their size and ease of capture and abundance.

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DISCUSSION

A. BAYER: Referring to Eq.(6) in your paper, I see that the influence of the 'declining environment' is described by λ, the radioactivity decay constant. The 'declining environment' might also be influenced by the entrainment of resuspended particles containing radioactive material. In such a case it should be replaced by a λ_{eff}, which is a larger number than λ. You might find that the discrepancies in your results were then somewhat reduced.

C.D. JENNINGS: Thank you for your comment. We shall certainly look into that possibility.

O.J. VAN DER BORGHT: Perhaps I could just comment that the relative extent of Zn+Fe uptake from the water, on one hand, and from the sediments, on the other, could provide a sound physiological basis for assessing the exchanges of Zn+Fe.

C.D. JENNINGS: I agree that studies using a physiological rather than mathematical model would be very useful.

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