

THE DISTRIBUTION OF TOXIC METALS IN MARINE ECOSYSTEMS  
AS A RESULT OF SEWAGE DISPOSAL  
AND NATURAL PROCESSES

by

Patrick J. Whaling, Richard T. Barber,

and Jacqueline C. Paul

Duke University Marine Laboratory

Beaufort, North Carolina 28516

The work upon which this publication is based was supported in part by funds provided by the Office of Water Research and Technology of the U.S. Department of the Interior, as authorized under the Water Resources Research Act of 1964, as amended, the Water Resources Research Institute of The University of North Carolina, and by Duke University.

Project No. B-071-NC

Agreement No. 14-31-0001-4115

December 1976

## ABSTRACT

The serious environmental crisis resulting from the industrial discharge of wastes containing high concentrations of mercury and other toxic metals has largely been abated by strong federal and state regulation. The recovery of the polluted ecological systems is taking longer. The problem of chronic, low concentration discharges of toxic metals in sewage effluent and sludge continues to be an environmental problem that is poorly defined, very widespread and one for which there is no simple solution, since sewage treatment is a necessary part of our way of life. In addition, the natural accumulation of threatening levels of mercury and cadmium in unpolluted marine food chains is an aspect of environmental concern that must be understood to protect human health. The role of sewage disposal and natural food chain processes in the distribution of toxic metals in marine ecosystems was examined in several small eastern North Carolina estuaries receiving different inputs. On the basis of metal distribution, estuaries receiving treated sewage effluent were clearly distinguishable from those not receiving this discharge. The magnitude and spatial extent of the elevated toxic metal distributions was very limited in both sediments and organisms. While some organisms did have elevated metal concentrations, the economically important oysters did not, nor did the marsh grass, Spartina alterniflora. The large and consistent variation in the amount of mercury and other trace metals discharged to the surrounding estuaries by various eastern North Carolina towns was traced to variations in the metal content of inflow to the plant and not to operational variations between the plants. It was not possible to account for the origin of the metal variations in wastewaters of these small towns which have no known commercial activities using these metals. Experiments with fish and shrimp fed natural foods containing wide variations in toxic metal strongly suggest that ingestion is the process responsible for the observed species specific variation in metal concentrations between organisms inhabiting the same environment.

## PREFACE AND ACKNOWLEDGMENTS

The primary intent of this report is to provide environmental managers and the responsible government authorities with the necessary information to make informed decisions on the chronic release of toxic trace metals to our surrounding estuaries via sewage disposal practices. Unlike the acute, high-concentration discharges involved in intentional industrial disposal, the release of toxic metals via routine sewage treatment practices is an environmental problem that is geographically widespread and not easily solved by government regulations. The responsibility for this problem rests with all of us and cannot be assigned to a remote entity, such as a corporation. To achieve the intent of providing information for decision making, we elected to include in the report the complete set of observations from which our conclusions and recommendations were drawn, making it possible for each reader to reach his own conclusions in the event he disagrees with those we reached.

A secondary use we intend for this report is to record a baseline set of environmental determinations of toxic metal distributions in eastern North Carolina estuaries and ecosystems so that future changes can be evaluated and interpreted with confidence. Perhaps the largest social benefit of this work will be derived when these data are reexamined in twenty-five or fifty years and our followers determine the progress made in maintaining the environmental quality of coastal North Carolina.

The authors acknowledge financial support from the Water Resources Research Institute of The University of North Carolina under an OWRT Matching Grants Project, No. B-071-NC, and Duke University. A large number of sewage treatment plant operators in eastern North Carolina and throughout the United States provided considerable assistance in the study of their treatment facilities; in particular, we thank Joe Clayton of the Morehead City sewage treatment plant and Ben Odham of the New Bern sewage treatment plant for their generous help. For guidance on analytical methodology and intercalibration, we thank Ford Cross of the Beaufort Laboratory of the National Marine Fisheries Service, Beaufort, North Carolina, and Wayne Willford of the Great Lakes Fishery Laboratory, Ann Arbor, Michigan. William Kirby-Smith and Elaine Barber provided editorial assistance. The authors acknowledge a special debt of gratitude to Sylvia Springle for her continued patience and expert typing skills. On the eve of David H. Howells' retirement as Director of the Water Resources Research Institute of The University of North Carolina, we express our thanks for his initial encouragement and continuing support. North Carolina has been fortunate to have a scientific administrator with the vision and vigor of David H. Howells.

## TABLE OF CONTENTS

	<u>Page</u>
ABSTRACT . . . . .	ii
PREFACE AND ACKNOWLEDGMENTS. . . . .	iii
TABLE OF CONTENTS. . . . .	iv
LIST OF FIGURES. . . . .	v
LIST OF TABLES . . . . .	viii
SUMMARY, CONCLUSIONS AND RECOMMENDATIONS . . . . .	xii
INTRODUCTION . . . . .	1
Background. . . . .	1
Objectives. . . . .	2
Strategy. . . . .	3
METHODS. . . . .	11
Mercury . . . . .	11
Other Metals. . . . .	12
Sediment Organic Content. . . . .	13
Intercalibration. . . . .	14
Sampling in Calico Creek. . . . .	18
RESULTS AND DISCUSSION . . . . .	22
Calico Creek. . . . .	22
Turner Creek. . . . .	67
Other North Carolina Estuaries. . . . .	93
Sewage Treatment Plants . . . . .	103
Feeding Experiments . . . . .	115
Exposure Experiments. . . . .	127
REFERENCES . . . . .	130

## LIST OF FIGURES

	<u>Page</u>
1. Map of eastern Carteret County showing the study areas . . . . .	4
2. Diagram of Calico Creek showing area sampled . . . . .	5
3. Exposed mud flats in Calico Creek . . . . .	6
4. Diagram of Turner Creek showing area sampled . . . . .	7
5. Plan of the Morehead City Sewage treatment plant . . . . .	8
6. Total mercury found in Calico Creek during 1972 . . . . .	24
7. Percent loss on ignition in Calico Creek for 1974 . . . . .	28
8. Total mercury found in Calico Creek during 1974 . . . . .	29
9. Cadmium found in Calico Creek during 1974 . . . . .	30
10. Chromium and copper found in Calico Creek during 1974 . . . . .	31
11. Iron and manganese found in Calico Creek during 1974 . . . . .	32
12. Lead found in Calico Creek during 1974 . . . . .	33
13. Zinc found in Calico Creek during 1974 . . . . .	34
14. Percent loss on ignition in Calico Creek during 1975 . . . . .	38
15. Total mercury found in Calico Creek during 1975 . . . . .	39
16. Cadmium found in Calico Creek during 1975 . . . . .	40
17. Chromium found in Calico Creek during 1975 . . . . .	41
18. Copper found in Calico Creek during 1975 . . . . .	42
19. Iron found in Calico Creek during 1975 . . . . .	43
20. Lead found in Calico Creek during 1975 . . . . .	44
21. Manganese found in Calico Creek during 1975 . . . . .	45
22. Zinc found in Calico Creek during 1975 . . . . .	46
23. Percent loss on ignition in Calico Creek for 1976 . . . . .	49
24. Total mercury found in Calico Creek during 1976. . . . .	50
25. Cadmium found in Calico Creek during 1976 . . . . .	51

26.	Chromium found in Calico Creek during 1976 . . . . .	52
27.	Copper found in Calico Creek during 1976 . . . . .	53
28.	Iron found in Calico Creek during 1976 . . . . .	54
29.	Lead found in Calico Creek during 1976 . . . . .	55
30.	Manganese found in Calico Creek during 1976 . . . . .	56
31.	Zinc found in Calico Creek during 1976 . . . . .	57
32.	<u>Spartina alterniflora</u> heavy metal stations in Calico Creek . . . . .	60
33.	Locations of sediment samples on transects from high marsh to subtidal. . . . .	69
34.	Total mercury found in Turner Creek during 1972 . . . . .	71
35.	Percent weight lost on ignition of sediments in Turner Creek 1974. .	73
36.	Total mercury found in Turner Creek 1974 . . . . .	74
37.	Cadmium and chromium found in Turner Creek for 1974. . . . .	75
38.	Copper and iron found in Turner Creek 1974 . . . . .	76
39.	Lead and manganese found in Turner Creek during 1974 . . . . .	77
40.	Zinc found in Turner Creek during 1974 . . . . .	78
41.	Heavy metal stations for oysters in Turner Creek during 1975 . . . . .	80
42.	Percent weight lost on ignition of sediments in Turner Creek during 1975 . . . . .	81
43.	Total mercury found in Turner Creek during 1975. . . . .	82
44.	Cadmium found in Turner Creek during 1975 . . . . .	83
45.	Chromium found in Turner Creek during 1975 . . . . .	84
46.	Copper found in Turner Creek during 1975 . . . . .	85
47.	Iron found in Turner Creek during 1975 . . . . .	86
48.	Lead found in Turner Creek during 1975 . . . . .	87
49.	Manganese found in Turner Creek during 1975 . . . . .	88

50.	Zinc found in Turner Creek during 1975 . . . . .	89
51.	<u>Spartina alterniflora</u> heavy metal stations in Turner Creek . . . .	95
52.	A comparison of mercury concentrations in sediment cores from Calico Creek and Turner Creek . . . . .	97
53.	Metal concentrations in sediment located below the Wrightsville Beach sewage treatment plant . . . . .	100
54.	Map of the Newport River showing sampling stations for heavy metals . . . . .	102
55.	Map of South River showing sampling stations for heavy metals . .	104
56.	Diel mercury and particulate pattern in Morehead City effluent . .	108
57.	Accumulation of mercury by pinfish fed various mercury diets . . .	119
58.	Mercury accumulation by pinfish fed high and low metal diets . . .	123
59.	Mercury, manganese, zinc, and chromium accumulation by pinfish fed high and low metal diets . . . . .	124
60.	Cadmium, iron, copper, and lead accumulation by pinfish fed high and low metal diets . . . . .	125
61.	Mercury accumulation in various organs of pinfish fed high and low metal diets . . . . .	126

LIST OF TABLES

	<u>Page</u>
1. Analytical conditions and detection limits of the atomic absorption spectrophotometer used in this study . . . . .	.13
2. EPA mercury in fish intercalibration . . . . .	.14
3. EPA mercury in sediments intercalibration . . . . .	.14
4. Trace metal intercalibration in EPA water reference samples . . . . .	.15
5. National Bureau of Standards reference sample intercalibration . . . . .	.16
6. National Bureau of Standards gold stabilized reference sample intercalibration . . . . .	.15
7. Duke University Marine Laboratory and NMFS Beaufort Lab blue marlin <u>Makaira nigricans</u> muscle sample intercalibration . . . . .	.17
8. Duke University Marine Laboratory and NMFS Beaufort Lab intercalibration of mercury in blue hake, <u>Antimora rostrata</u> , axial muscle . . . . .	.17
9. Duke University Marine Laboratory and Northern States Fisheries Laboratory intercalibration of mercury in total fish . . . . .	.18
10. International Atomic Energy Agency heavy metals reference sample intercalibration . . . . .	.19
11. Mercury concentrations in surface sediment, the snail and the oyster from stations above and below the outfall in Calico Creek-May 1972 . . . . .	.23
12. Heavy metal concentrations and percent loss on ignition values in surface sediment from stations above the outfall in Calico Creek - May 1974 . . . . .	.25
13. Heavy metal concentrations in the snail at individual stations above and below the outfall in Calico Creek - May 1974 . . . . .	.27
14. Heavy metal concentrations and percent loss on ignition values in surface sediment from stations above and below the outfall in Calico Creek - July 1975 . . . . .	.35
15. Heavy metal concentrations in the snail at stations above and below the outfall in Calico Creek - July 1975 . . . . .	.36
16. Heavy metal concentrations in the oyster at stations above and below the outfall in Calico Creek - July 1975 . . . . .	.37

17.	Heavy metal concentrations and percent loss on ignition values in surface sediment from stations above and below the outfall in Calico Creek - January 1976 . . . . .	47
18.	Heavy metals concentrations in the snail at stations above and below the outfall in Calico Creek - January 1976 . . . . .	48
19.	Heavy metals in the marsh grass <u>Spartina alterniflora</u> compared to total and leached metal data for the same stations above and below the outfall in Calico Creek . . . . .	61
20.	Heavy metal concentrations in eight surface sediment samples taken 1/2 m apart 800 m below the outfall in Calico Creek in July 1975 . . . . .	63
21.	Heavy metal concentrations in eight surface sediment samples taken 1/2 m apart 800 m below the outfall in Calico Creek in January 1976 . . . . .	63
22.	Heavy metal concentrations in six surface sediment samples taken 1/2 m apart at the same station 10 m above the outfall in Calico Creek . . . . .	64
23.	Leachable heavy metals in surface sediments in Calico Creek . . . . .	65
24.	Heavy metal concentrations in leached sediment samples from below the outfall in Calico Creek in July 1975 . . . . .	66
25.	Heavy metal concentrations of sediments in transects across Calico Creek . . . . .	68
26.	Mercury concentrations found in surface sediments, snails, and oysters from stations in Turner Creek - May 1972 . . . . .	70
27.	Heavy metal concentrations and percent loss on ignition values in surface sediment from stations in Turner Creek - May 1974 . . . . .	72
28.	Heavy metal concentrations and percent loss on ignition values in the surface sediment from individual stations in Turner Creek - July 1975 . . . . .	79
29.	Heavy metal concentrations in snails in Turner Creek - July 1975 . . . . .	90
30.	Heavy metal concentrations in the oyster <u>Crassostrea virginica</u> at individual stations in Turner Creek - July 1975 . . . . .	91
31.	Heavy metal concentrations in eight surface sediment samples taken 1/2 m apart in Turner Creek . . . . .	92

32.	Heavy metal concentrations in <u>Spartina alterniflora</u> compared to total and leached metal data for the same station in Turner Creek . . . . .	94
33.	Heavy metal concentrations of sediments in transects across Turner Creek . . . . .	96
34.	Comparison of mercury concentrations in sediment cores from Calico Creek and Turner Creek . . . . .	98
35.	Heavy metal concentrations and percent loss on ignition values in the surface sediment from stations below the Wrightsville Beach sewage treatment plant . . . . .	99
36.	Heavy metal concentrations and percent loss on ignition values in the surface sediment below the Newport sewage treatment plant. .	101
37.	Heavy metal concentrations and percent loss on ignition values in surface sediment below the West Carteret High School sewage treatment plant . . . . .	103
38.	Heavy metal concentrations and percent loss on ignition in surface sediments in South River . . . . .	103
39.	Heavy metal concentrations and percent loss on ignition in a Newport River estuary transect . . . . .	105
40.	Temporal variation of mercury in the influent of sewage treatment plants . . . . .	106
41.	Mercury concentration at the various stages of treatment throughout the Morehead City and New Bern sewage treatment plants . . . . .	106
42.	Temporal variation of mercury concentrations in Morehead City sewage treatment plant effluent . . . . .	107
43.	Mercury concentrations in effluent of the Morehead City sewage treatment plant taken during a 24-h sampling period . . . . .	107
44.	Kilograms of mercury dumped annually based on mercury in effluent and average daily flow rate of various sewage treatment plants . .	109
45.	Geographic variations in mercury concentrations of sewage treatment plant effluent . . . . .	111
46.	Chronological listing of metal concentrations in sludge . . . . .	112
47.	Heavy metal concentrations found in sludge from various cities . .	113

48.	Concentrations of metals at three stages of sludge processing in the Morehead City sewage treatment plant . . . . .	.110
49.	Heavy metal concentrations in repeated samplings of the Beaufort and Newport sludge beds . . . . .	.114
50.	Mercury accumulation in <u>Penaeus sp.</u> muscle tissue after a 30-day feeding experiment of four diets . . . . .	.117
51.	Mercury concentration in <u>Lagodon rhomboides</u> using adequate and deficient diets with both high and low mercury concentrations . . .	.118
52.	Mean weights (g) of <u>Lagodon rhomboides</u> during a 60-day feeding experiment. . . . .	.120
53.	Mercury concentrations in gill tissue of pinfish fed a high and low metal diet. . . . .	.122
54.	Heavy metal concentrations in high and low metal diets . . . . .	.122
55.	Mercury concentrations in oysters held over sludge. . . . .	.127
56.	Mercury concentrations in animals held in running seawater in contact with sediments. . . . .	.128
57.	Mercury in <u>Cyrius carpio</u> taken from the Durham Sandy Creek sewage treatment plant holding lagoons, and published values . . . . .	.129

## SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

### Summary and Conclusions

The small tidal creeks in Morehead City and Wrightsville Beach that received treated effluent had elevated concentrations of toxic metals, especially mercury, whose distributional pattern was clearly related to the point source discharge of effluent from nearby municipal sewage treatment plants. The spatial and long-term temporal pattern of quantitative distribution of metals within the Calico Creek system at Morehead City was complex but during every sampling period of our three-year study an association between the site effluent discharge and elevated metal concentrations in the sediments was present. The element most strongly and consistently associated with the effluent discharge was mercury, the most serious toxic metal polluting marine ecosystems. For human health and economic considerations the major question is, "How much of the elevated toxic metals enter the marine food chain?" In Calico Creek the spatial pattern of mercury distribution in periwinkle snails living throughout the creek reflected the outfall-determined pattern that was present in the sediment. In one transect the concentrations of mercury in snails were  $0.61 \pm 0.07$  ppm (wet weight) 30 m below the outfall, decreasing to a value of about  $0.09 \pm 0.03$  ppm 900 m below the outfall. On this transect (January 1976) mercury content in the snails correlated with the distance to the outfall with a coefficient of 0.87 and the sediment mercury content correlated with snail mercury content with a coefficient of 0.718. It seems clear that the mercury entering the estuary via sewage effluent is entering the food chain. The mercury concentration observed in snails on this transect 30 m from the outfall,  $0.61 \pm 0.07$  ppm (wet weight), exceeds the Food and Drug Administration limit of 0.5 ppm (wet weight) set for human consumption. On the other hand, the periwinkle is not consumed by man, at least not in North Carolina. Therefore, this concentration of mercury, while exceeding slightly the federal guideline, is not a threat to human health.

Oysters were not abundant along the Calico Creek transect and fewer analyses were possible, but the available information indicates that oysters did not show a pattern of mercury enrichment related to the effluent discharge and that mean mercury concentrations in oysters in the creek with an outfall and in a "control" creek receiving no point source discharges were the same. The oyster, while growing on contaminated sediments, is isolated from metal contamination by its filter feeding mode. The important estuarine primary producer, Spartina alterniflora, which grows throughout the creek, illustrates the biological complexity of pollutant transfer phenomena. The stems and leaves of Spartina growing in sediments with elevated mercury did not show elevated mercury concentrations, but the roots reflected the sediment pattern. The correlation coefficient of root mercury concentration with sediment mercury was 0.758 and the correlation coefficient of root mercury concentration with distance to the outfall was 0.855. While the pattern of elevation in Spartina roots was definite, the magnitude of elevation was not great. The mean mercury concentration in Spartina roots in Calico Creek was  $0.14 \pm 0.09$  ppm, while in the "control" estuary it was  $0.13 \pm 0.08$  ppm.

Laboratory experiments in which organisms were held in running seawater

for 90 to 150 days over trays of sludge sediment containing up to 8 ppm (dry weight) of mercury confirmed the field observations in considerable detail. Oysters exposed for 120 days to sludge sediment having mercury concentrations elevated approximately 100-fold relative to the natural control sediment showed no increase in mercury content. Exposure for 150 days to sludge resulted in an increase in mercury concentration in oysters but the weight of the oyster meat decreased in this experiment so that the total body burden of mercury for each oyster remained the same or dropped during five months of exposure to a highly contaminated sediment. A parallel 90-day experiment with shrimp, scallops and mud snails provided further confirmation of the pattern of mercury partitioning among the organisms which was observed in the estuary receiving sewage effluent. The concentration of mercury in snails increased two and one-half times while the scallop and shrimp concentrations did not change during the three-month exposure to the contaminated sediment. Our lab and field observations indicate that physical exposure to contaminated water or sediments does not lead to increased mercury concentrations, and that only direct ingestion of the contaminated material will alter the organisms' metal content. This prosaic observation is interesting since the current dogma in metal uptake work is that mercury and other trace metals enter marine organisms directly from the water rather than via food. The uptake route was examined in detail by maintaining fish and shrimp in running seawater and feeding them diets that had widely varying, but natural, mercury content. Blue marlin containing about 10 ppm (wet weight) of mercury and bluefish containing 8 ppm of mercury were used as food that was naturally high in mercury. These experiments clearly indicated that this food was the major source of the mercury found in the shrimp and fish tissues.

Studies of various sewage treatment plants in eastern North Carolina having effluent mercury concentrations that differ by a factor of eight indicated that the differences in concentrations were not related to the operational practices of the various plants. All of the plants examined reduced the mercury content of the incoming wastewater by differentially partitioning the mercury into sludge; the efficiency of partitioning varied slightly but not enough to account for the variations in effluent mercury concentrations. The major cause for higher effluent concentrations was higher concentrations in the wastewater entering the treatment facility. The reason why two towns as similar as Morehead City and Beaufort should have mercury concentrations in their wastewaters that differ by a factor of eight remains an interesting mystery.

On the basis of this study we can confidently conclude that any coastal zone municipality discharging effluent into a small tidal estuary will produce a zone of elevated toxic trace metal concentration in the sediment and in certain organisms. It seems clear that this environmental alteration violates the Federal Water Pollution Control Act as amended in 1972 (Public Law 92-500), but there seems to be a "gentlemen's agreement" in the United States not to strictly enforce Public Law 92-500 in regard to municipal effluent alterations of the environment. Only two suits have been filed in federal courts attempting to force municipalities to comply strictly with Public Law 92-500; the second legal action of this nature was taken against the town of Wrightsville Beach and the data presented in this report were a

portion of the evidence indicating that the town was indeed violating federal statutes. As a result of the mid-trial plea bargaining, the outcome of that court action set no precedent, but the town did admit to unwittingly violating Public Law 92-500.

### Recommendations

It is clear that discharging municipal effluent into small tidal estuaries causes elevated accumulations of toxic metals, especially mercury, and that this environmental alteration, because of the extreme toxicity of mercury, is a form of environmental degradation prohibited by federal statute. But the authors of this report feel that major modifications of our waste treatment practices are not justified on the basis of the toxic metal alterations generated by the current practices. The changes we observed are easily detected but do not pose a serious threat to human health, economic potential or ecological integrity. (We address here only the toxic metal consequences of effluent discharges; the bacterial contamination from these discharges certainly levy a high economic toll by causing large areas of the estuarine waters to be closed to shellfishing.) The impacted zones created by the outfalls are small in horizontal extent and the magnitude of the elevated concentrations in sediments and organisms is low relative to the concentrations resulting from intentional industrial disposal. Our value judgment is that the ecological consequences of current effluent practices are acceptable in view of the social utility of these practices. Certainly, the elevated concentrations of mercury that we observed would not justify the expense of tertiary treatment of the effluent for mercury.

The important recommendations from this study deal with the future and the decisions to be made regarding population growth and new waste treatment initiatives. We strongly concur with the first recommendation on sewage given in the current EPA Handbook of Water Quality Criteria that "Untreated or treated municipal sewage discharges should be recognized as a major source of toxic substances. Recommendations for these constituents will limit the amount of sewage effluent that can be dispersed into estuaries."

To implement this recommendation new waste treatment initiatives will have to consider the total physical and biological character of the proposed receiving system in relation to the magnitude of the discharge. While in the past design considerations involving toxic metals were associated only with industrial discharges, it is now known that municipal facilities are significant sources of these dangerous elements.

An additional recommendation is that the mercury "budget" of small towns without metal-using industry and the budget of their associated waste treatment facilities must be better known. It is impossible to design and plan logical treatment facilities in total ignorance of the sources of an important constituent of the waste being treated. A classical biogeochemical flux analysis of a small town would solve this problem.

## INTRODUCTION

### Background

Considerable research effort has been expended during the past half decade to determine man's impact on the biogeochemical cycling of toxic trace metals, especially mercury and cadmium. The early work was stimulated by events involving death and permanent impairment resulting from the human consumption of marine or aquatic organisms that were contaminated by intentional industrial discharges (Lofroth, 1970; Goldwater, 1971). Logically, early research effort focused on the threat to man via aquatic and marine food chains contaminated by the discharge of industrial wastes containing very high concentrations of the toxic metals, especially mercury and cadmium. By the mid-1970's the initial environmental crisis and health threat had been considerably abated by fast action on the part of federal and state agencies, halting the discharge of these toxic waste materials by strict regulations and standards. Having dealt with the acute problem of toxic metal contamination of marine ecosystems, it was then appropriate to turn research effort towards the less serious problem of chronic discharge of municipal sewage or sewage treatment products containing mercury and other toxic metals. The important question was, "What is the impact of these widespread and continual inputs of trace metals via sewage treatment practices on the structure and function of the receiving marine ecosystems?" It has been established that the natural global flux of mercury is large and, in fact, quantitatively overwhelms the cultural inputs to the world's oceans (Weiss *et al.*, 1971). However, on the local estuarine scale the chronic input of sewage effluent containing mercury and other toxic metals appears to be capable of altering the biotic community of the receiving system. Studies focusing on the large effluent and sludge discharges from the New York metropolitan area and the southern California region have clearly demonstrated that effluent and sludge disposal causes elevation of toxic metal content of sediments and organisms around the outfall and in areas of sludge dumping (NOAA, 1975; NAS, 1976). Furthermore, it is clear that the presence of toxic metal ions in the water and sediments is responsible for functional disorders in the form of diseases or necrotic lesions on fish and invertebrates (Young and Pearce, 1975; SCCWRP, 1975, 1976). The large magnitude discharges from the two largest metropolitan areas in the United States are the extremes of man's intervention in marine ecosystems and it is not surprising that in both cases there has been considerable loss of environmental quality and some threat to human health. The question this study was designed to address is conceptually related to the large metropolitan discharges but differs in scale in two ways. The concentration of toxic metals in treated effluent and sludge is much lower than in industrial wastes and the magnitude of effluent discharge is much less than the discharges by New York or Los Angeles so the flux of metals to a given area of the marine environment is many times lower than in the previously mentioned situations. On the other hand, the discharges to the estuarine environment pervade the entire coastal region; each municipality having an organized sewage system creates a point source input of toxic metals. It seems appropriate to establish clearly the ecological consequences of these inputs. The potential importance of sewage derived toxic metals was recognized in the current EPA Handbook of Water Quality Criteria (NAS, 1973). The

first recommendation regarding sewage was:

- "1. Untreated or treated municipal sewage discharges should be recognized as a major source of toxic substances. Recommendations for these constituents will limit the amount of sewage effluent that can be dispersed into estuaries."

### Objectives

The first objective of this study was to determine the extent and importance of mercury and other toxic metal contamination on the eastern North Carolina estuarine environment by the routine discharge of treated sewage effluent and sludge. To achieve this objective, the distribution of metals was examined in a variety of small estuarine systems, some of which received point source effluent and sludge discharges from nearby towns and some of which did not. The spatial pattern of metal distribution within the small estuarine creek was examined in relation to the point of discharge and this pattern was compared with the distribution in comparable estuarine creeks without point source inputs of effluent or sludge. The metal concentration as a function of space and over a period of three years was analyzed in sediments, in two common estuarine animals (the periwinkle snail, Littorina irrorata, and the oyster, Crassostrea virginica) and in the marsh grass, Spartina alterniflora.

Pilot studies on the effluent from a number of small eastern North Carolina towns indicated that there were significant and consistent differences in the mercury content of effluent and sludge from different towns; the second objective of this study evolved from this observation and sought to determine the source of the variation in mercury concentrations that existed in various sewage treatment systems. Understanding this local variation would obviously be useful in predicting the ecological consequences of a particular sewage treatment plant's discharges and it was hoped that an understanding of this variation would provide a basis for helping sewage treatment plant operators to improve their operating procedures. The investigation of variations between sewage treatment plants led to studies on the mercury budgets of various plants and on the temporal patterns of mercury flux in municipal wastewaters.

A third objective of the study focused on the character of the uptake mechanism involved in the flux of toxic trace metals into marine organisms. Pursuit of this objective led to exposure experiments where organisms were maintained in proximity to sludge, and to feeding experiments where a variety of foods containing naturally occurring mercury was provided. The feeding experiments, using foods having widely varying concentrations but naturally occurring toxic metals, provided interesting insights on the dynamics of metals in marine ecosystems and received additional emphasis as the study progressed.

Primary focus in terms of the metals analyzed was on mercury with secondary emphasis on the analysis of cadmium, chromium, copper, iron, lead,

manganese and zinc. While the study was originally designed to investigate mercury alone, it became apparent as technique progressed that considerable efficiency in terms of the information gained for each unit of analytical effort could be achieved by expanding the sample processing procedure to include the other metals. An additional 50% in sample processing effort provided information on seven additional elements including environmentally critical elements, cadmium, chromium and lead. These metals have been clearly and unequivocally implicated in ecological degradation or in human health considerations (Nomiya, 1975; Bascom, 1976) but it is appropriate to justify the primary emphasis on mercury by citing this element's involvement in the largest human health event, the Minamata Bay disaster (Goldwater, 1971) and its status as the "most critical" element in marine pollution according to the Ketchum index of criticality (Ketchum et al., 1975).

### Strategy

The basic strategy of this study was to determine the effects of the discharge of effluent on estuarine ecosystems by comparing different areas that had different relationships to the known point sources. Most of the analytical effort was focused on two small tidal embayments in Carteret County, North Carolina. The two creeks are small tributaries of larger embayments. They are shallow with much of the area exposed at low tide. Neither creek has significant freshwater input and both are bounded by Spartina and Juncus marsh. The creeks are about three miles from Beaufort Inlet and are very similar in size and configuration (Figure 1). Calico Creek, the system receiving treated effluent from the Morehead City sewage treatment plant, empties into the Newport River and is surrounded by residential dwellings on the north shore, both above and below the outfall (Figure 2). A former sanitary landfill is located opposite the outfall and the 20th Street bridge (800 m) that crosses Calico Creek is a major traffic artery in Morehead City. Rain runoff from the street plus the dumping of various materials such as asphalt are additional sources of metals. Other major street drains enter the creek at 1200 and 1500 m below the outfall. A large agricultural area has a ditch emptying into the creek 550 m above the outfall. The average depth of Calico Creek at high tide is one meter and at low tide, only about 15 cm. The average salinity is 5 ppt and 16-20 ppt 1800 m below the outfall. Two representative areas of Calico Creek are shown in Figure 3.

Turner Creek (Figure 4) was selected as a comparison study area because it receives no point source discharges but is similar in many aspects to Calico Creek. Turner Creek has fewer residential dwellings on its shore. About fifty percent of the area is surrounded by agricultural land (cattle and horse grazing) and an area of about 400 m of shoreline used to be the municipal landfill for the Town of Beaufort.

The Morehead City sewage treatment plant (Figure 5) is a trickling filter plant that handles an average flow of 1.04 million gallons per day. The plant empties its effluent into a small tidal creek about 0.8 km upstream from the 20th Street bridge in Morehead City. Sludge from the sludge beds is trucked off by area farmers or dumped at the local sanitary dump. A typical summer influent consists of 1060 mg/l total solids and 233 mg/l total suspended

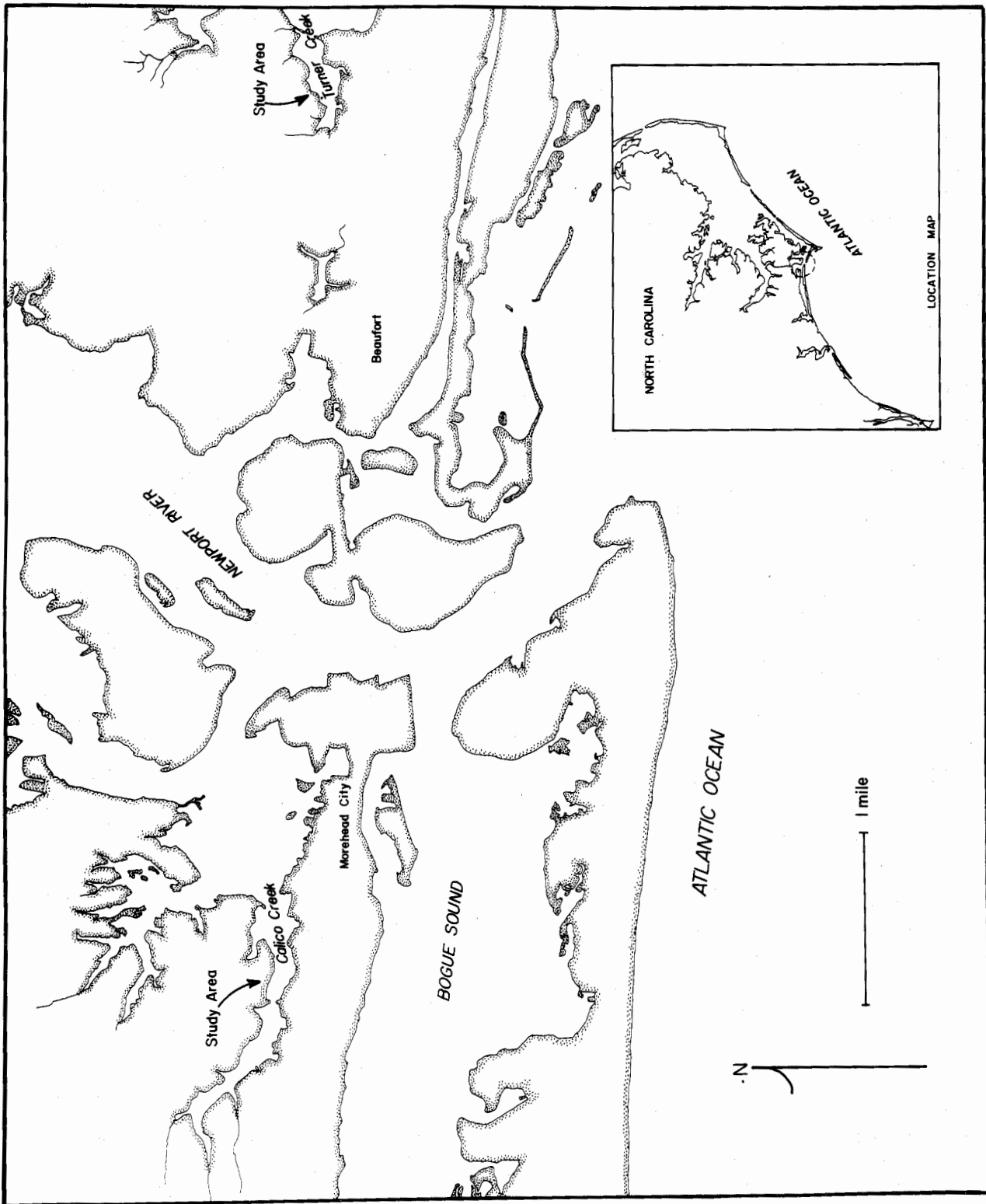


Fig. 1. Map of eastern Carteret County showing the study areas

5

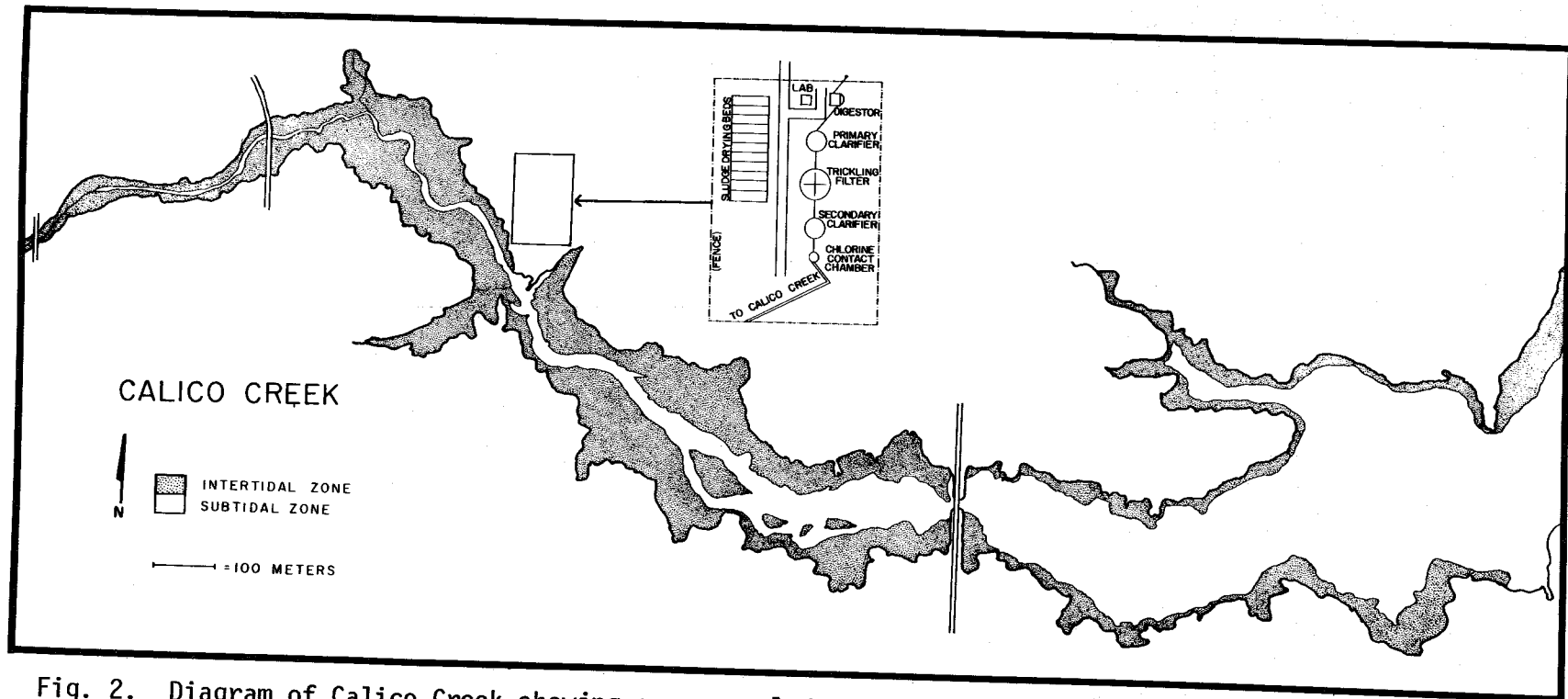


Fig. 2. Diagram of Calico Creek showing area sampled



(A)



(B)

Fig. 3. (A) The exposed mud flats of Calico Creek from 600 meters to 800 meters above the outfall at low tide looking away from outfall. (B) The exposed mud flats and oyster beds from 1600 meters below the outfall looking towards the outfall.

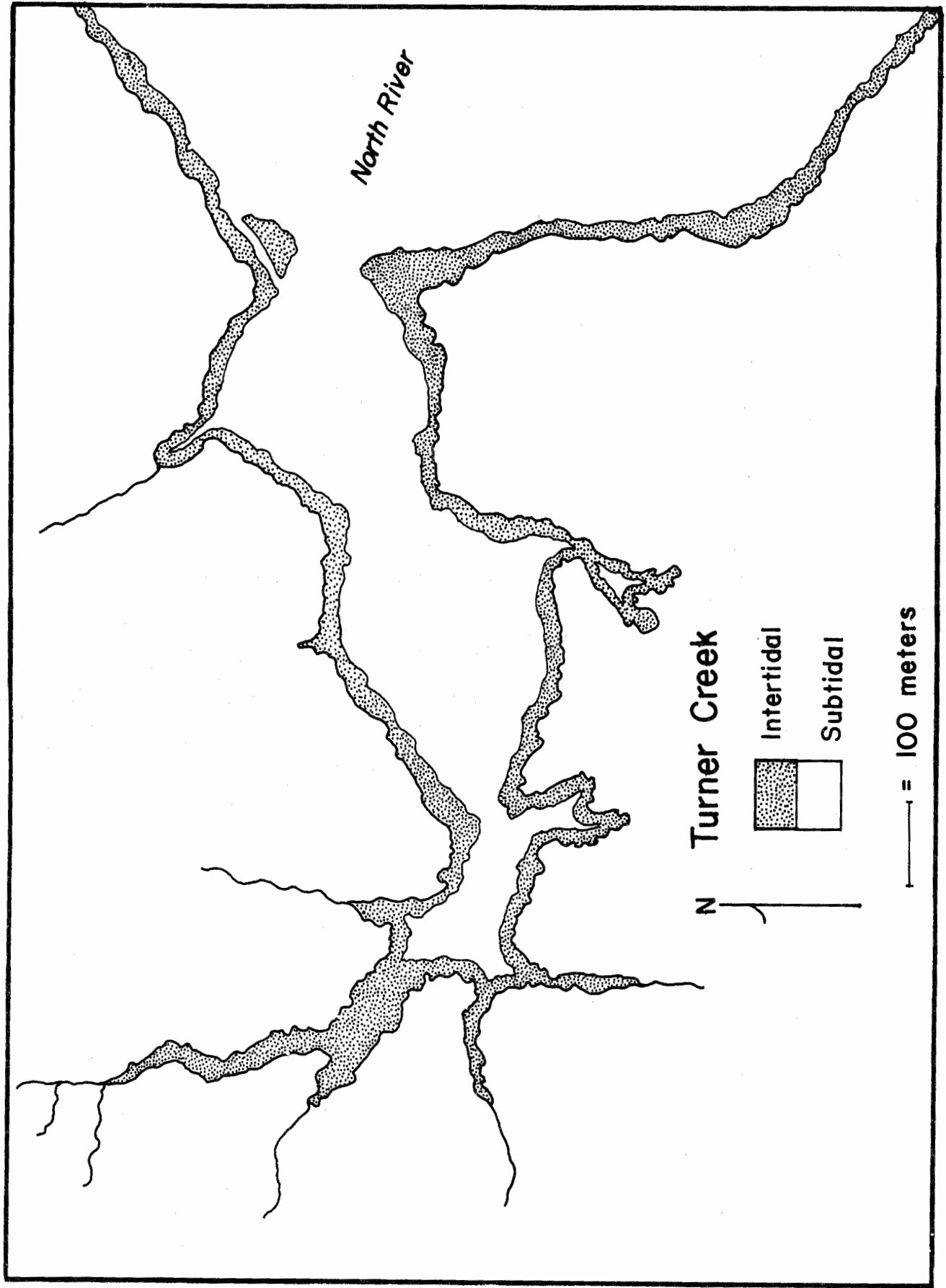
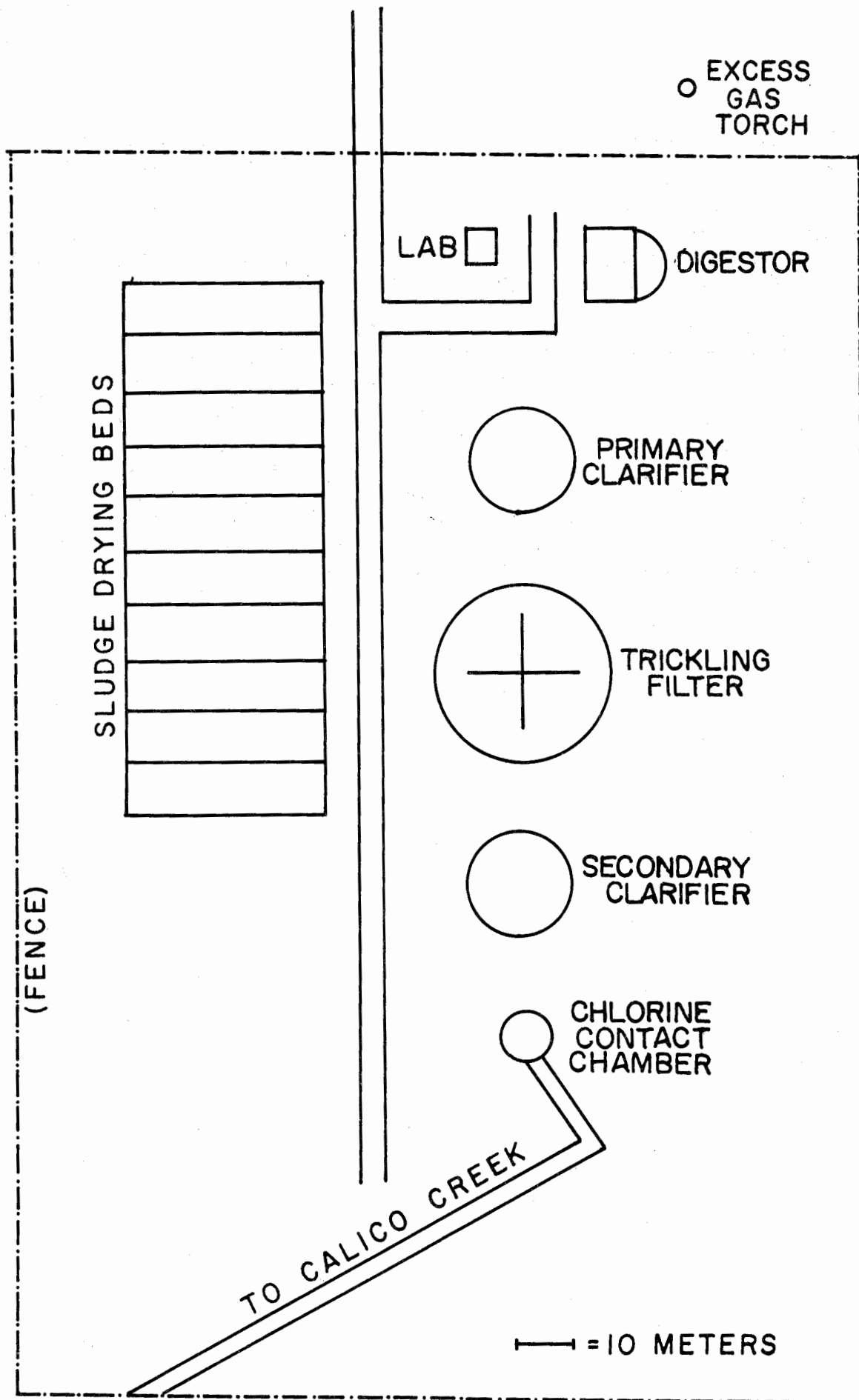


Fig. 4. Diagram of Turner Creek showing area sampled

Fig. 5. Plan of the Morehead City Sewage Treatment Plant. Raw sewage enters the primary clarifier where solids and liquids are separated. Solids are then passed to the digester and finally to sludge beds to dry. Liquid wastes are transferred from primary to secondary clarifiers after which they may move into the primary clarifier again or go directly to the trickling filter. From the trickling filter, liquids move into the chlorine contact chamber and then into Calico Creek at the outfall. Samples were collected from the sludge drying beds, the primary clarifier and the chlorine contact chambers.



solids; after a retention time of about four hours in the plant the effluent is discharged containing 1090 mg/l total solids and 32 mg/l total suspended solids. When this study was begun in 1972 the plant had problems with infiltration from the storm sewers, infiltration at high tide from Calico Creek, and major breakdowns. Since 1972 the plant has been upgraded considerably and appears to be doing an adequate job of treating the sewage it receives before the effluent is discharged into Calico Creek.

Beaufort sewage treatment plant is a complete mix activated sludge design that handles an average summer flow of 0.75 million gallons per day. A typical influent consists of 3.9 ml/l settled solids with only a trace remaining in the effluent. This effluent is dumped into Taylor's Creek which has a much greater flushing action than Calico Creek. The sludge is dumped at a sanitary landfill well removed from the "control" creek. The Newport sewage plant is a contact stabilization activated sludge plant handling an average of 0.11 million gallons per day. A typical influent has 516 mg/l of total solids and 150 mg/l total suspended solids. The effluent has 39 mg/l of total solids and 9 mg/l total suspended solids and is discharged into the Newport River, a medium-sized coastal plain river with considerable freshwater flow.

## METHODS

### Mercury

Total mercury was determined by the Hatch and Ott (1968) procedure with the detailed modifications suggested by the Environmental Protection Agency (1972). A detailed study of the mercury procedure per se is reported in Eganhouse (1975). The following format was used to determine total mercury.

#### Reagents

1. Concentrated sulfuric acid
2. Concentrated nitric acid
3. Potassium permanganate
4. Potassium persulfate (10% W/V solution)
5. Hydroxylamine hydrochloride
6. Stannous chloride (100 g/18 ml H<sub>2</sub>SO<sub>4</sub> brought to one-liter volume)

#### Procedure

1. Weigh sample and place in 300 ml BOD
2. Add 5 ml of HNO<sub>3</sub> and 8 ml of H<sub>2</sub>SO<sub>4</sub>
3. Heat to 90°C for minimum of 18 hr
4. Dilute sample to 100 ml with deionized H<sub>2</sub>O
5. Add KMnO<sub>4</sub> until purple color persists
6. Add one ml of K<sub>2</sub>S<sub>2</sub>O<sub>8</sub> and allow to oxidize for 24 hr at 80°C
7. Add hydroxylamine hydrochloride until solution is clear
8. Add 5 ml of stannous chloride and immediately attach to bubbler system
9. Using a Coleman MAS-50 mercury analyzer record peak height with memory on and find the amount (µg of Hg) on calibration curve

Various aliquots of Coleman 1000 ppm Mercury Standard were added to 100 ml of deionized water and were run blindly with the samples to determine a calibration curve. Only the linear portion of the scale was used. If a sample did not fall on the linear portion of the scale, a dilution of the sample was rerun.

Investigators have pointed out that 50% stannous chloride solution which is several days old does not effectively vaporize Hg<sup>++</sup> (Eganhouse, 1975). We found that after two days the stannous chloride solution is ineffective and, therefore, fresh stannous chloride solution was made up daily. The EPA procedure (EPA, 1972) suggested that hydrochloric acid be used to prevent the stannous chloride from being cloudy; we found this change decreased resolution considerably. The blank values for hydrochloric acid stannous chloride solution were twice as great as for stannous chloride dissolved in nitric acid. To prevent cloudiness the dispenser bottle was placed on a magnetic stirrer with a Teflon magnetic bar added to the solution. If the stannous chloride did not rapidly dissolve with the addition of the concentrated nitric acid, it was heated over a Bunsen burner for 2 min. The absorbent, magnesium perchlorate, was changed every 20-30 min (about every 15 samples) or, if moisture could be seen in the absorbent cell, the magnesium perchlorate was changed. The biggest analytical problem which was encountered was clouding of the

quartz windows in the absorption cell of the Coleman MAS-50. The clouding usually occurred on the end of the cell next to the lamp and away from the stannous chloride. Until June 1975 the only commercially available windows were thin and lasted only a few weeks. After June 1975 thicker windows became available and a single set has lasted for the remainder of the project. Windows that looked clean could still cause a considerable amount of detection variation. If two windows, a new and a used, are held side by side, the operator should be able to read detailed print clearly through both of them; when even a slight haze develops the windows must be changed. Volume flow of the sweep pump was monitored regularly. This is often overlooked by investigators, although personal communication with the Coleman service people indicated this is usually the first thing that must be corrected when a machine needs repair. A size #12 Gilmont flowmeter was used to ensure a uniform one liter per minute flow.

### Other Metals

A dry ash technique described by Henderick (1968) was used to analyze for cadmium, chromium, copper, iron, lead, manganese and zinc. To determine these metals subsamples were combusted, reweighed, dissolved in acid, and analyzed using a Varian Flame Atomic Absorption Spectrophotometer Model AA6.

#### Reagents

1. Concentrated nitric acid

#### Procedure

1. Weigh 5 g of sample and place in 125 ml Erlenmeyer flask
2. Combust at 500°C
3. Reweigh
4. Add 5 ml of acid and heat to near dryness
5. Filter with glass fiber filter and 0.45  $\mu\text{m}$  Millipore filter and bring filtrate to volume
6. Aspirate sample into AA6 and record the absorption

To estimate the amount of metal that was biologically available to organisms (as opposed to the total metal content) a preferential leaching technique described by Smith and Windom (1972) was used.

#### Reagents

1. Acetic acid 35% (v/v) solution
2. Hydroxylamine hydrochloride 25% (v/v) solution
3. Mixed acid-reducing reagent (150 ml of 25% hydroxylamine hydrochloride mixed with 350 ml of 35% acetic acid)
4. Concentrated nitric acid

#### Procedure

1. Place 5 g of powdered, dried sediment in a 150 ml Erlenmeyer flask with fitted ground glass stopper
2. Add 10 ml of the mixed reagent and stopper the flask
3. Shake the flask on a mechanical shaker for 4 hr
4. Filter the solution through a glass fiber filter paper

5. Add 5 ml of concentrated HNO<sub>3</sub> to the filtrate and heat to near dryness
6. Bring filtrate to volume
7. Aspirate sample into AA6 and record the absorption

Table 1 gives the wavelengths (nm), spectral band pass (nm), and lamp current (ma) specified by Varian for Model AA6. Detection limits (ppm) are for the solution aspirated into the atomizer and are defined as the concentration of an element in solution which gives a signal equal to twice the standard deviation of a series of measurements near blank level. Standards were made up daily from the 1000 ppm stocks and blanks were run with the appropriate amounts of acids added. All glassware was washed with concentrated nitric acid, and rinsed five times with tap water and once with deionized water.

Table 1. Analytical conditions and detection limits of the atomic absorption spectrophotometer used in this study

<u>Metal</u>	<u>Wave Length (nm)</u>	<u>Spectral Band Pass (nm)</u>	<u>Lamp Current (ma)</u>	<u>Detection Limit (ppm)</u>
Mercury	253.7	x	x	.0005
Cadmium	228.8	.5	3	.01
Chromium	357.9	.2	5	.05
Copper	324.7	.5	3	.05
	327.4	.2		.1
Iron	248.3	.2	5	.1
	372.0	.2		1.0
	386.0	.2		10.0
	394.0	.2		50.0
Lead	217.0	1.0	5	.05
Manganese	279.5	.2	5	.05
	403.1	.2		1.0
Zinc	213.9	.5	5	.01

#### Sediment Organic Content

Percent loss on ignition was used to determine the quantity of organic matter present in sediments. The sediment is ashed at 500°C. Values obtained from anhydrous weighing before and after combustion were used to calculate the percentage of weight lost. One problem with this method is that carbonate begins to decompose at 500°C; this problem had to be balanced against the need to combust at a temperature high enough to ensure complete oxidation of the organic matter.

## Intercalibration

Several U.S. and international agencies provide intercalibration standards for comparison of procedures, testing methods and monitoring consistency. This study was intercalibrated with 1) U.S. Environmental Protection Agency, Athens, Georgia; 2) International Atomic Energy Agency, Vienna, Austria; 3) National Oceanographic and Atmospheric Administration Labs - Great Lakes Fishery Lab, Ann Arbor, Michigan; 4) National Marine Fisheries Service, Beaufort, North Carolina; and 5) National Bureau of Standards, Washington, D. C.

Environmental Protection Agency. In early 1973 the U.S. Environmental Protection Agency (EPA) carried out a dry, homogenized fish intercalibration in which Duke University Marine Lab participated. All concentrations (using 0.5 g samples) were fairly high and our first determinations were low because the values fell on the plateau portion of the concentration vs absorption curve. With recalibration of the mercury analyzer and a much smaller sample the values reported in Table 2 were determined.

Table 2. EPA mercury in fish intercalibration. (Means for all labs, all methods and this study.) (ppm dry weight)

<u>Sample Number</u>	<u>Mean all Labs All Methods</u>	<u>This Study DUML</u>
72C1222	2.06 ± .84	2.30 ± .10
72C1223	5.75 ± 1.62	3.47 ± .38
72C1224	7.23 ± 2.02	6.55 ± 1.00

The number of replicates per sample for this study was twelve. The mean values for this study are in good agreement with sample numbers 72C1222 and 72C1224 but fall low of the mean for all methods with sample 72C1223. In 1972 EPA distributed dry sediment samples as an intercalibration and the results are reported in Table 3.

Table 3. EPA mercury in sediments intercalibration. (Means for all labs, all methods and this study.) (ppm dry weight)

<u>Sample Number</u>	<u>Mean all Labs All Methods</u>	<u>This Study DUML</u>
72C5644	43.9 ± 9.13	40 ± 2
72C5645	.22 ± .38	.09 ± .02

The number of replicates for samples 72C5644 and 72C5645 was fifteen and the sample sizes were 0.05 g and 1.0 g respectively. There was good agreement of this study with the values reported for all labs and all methods. EPA provided us with three water sample concentrates that had certified values

for ten metals of which only mercury, chromium, copper, lead and zinc were analyzed in this study. The data given in Table 4 report our results.

Table 4. Trace metal intercalibration in EPA water reference samples (ppb wet weight)

	<u>1</u>		<u>2</u>		<u>3</u>	
	EPA	DUML	EPA	DUML	EPA	DUML
Mercury	.42	.48±.05	2.4	2.9±.1	7.0	11.1±.1
Chromium	9	9	83	70	73	72
Copper	9	6	67	66	314	>100
Lead	28	23	92	84	350	>100
Zinc	10	11	76	73	367	350

National Bureau of Standards. Our results on the NBS standards (Table 5) agreed very well with the NBS certified values. NBS reference powdered coal dust standard analyzed in this study did not have a matrix similar to the matrix encountered in this study but did provide a check on the methods used.

Table 6 lists the results of twenty analyses of a NBS gold stabilized mercury-in-water reference sample. This standard was used in the procedure for the analysis of effluent samples. Spikes of the reference sample were added to effluent samples and their recovery was 100%.

Table 6. National Bureau of Standards gold stabilized reference sample intercalibration

	Mercury (ppm)	
	<u>NBS</u>	<u>DUML</u>
$\bar{N}$		20
Mean	1.5	1.8
St.Dev.	.1	.2
Co.Var.	7	11

National Marine Fisheries Service. In 1974 a set of eleven digested and processed blue marlin, *Makaira nigricans*, axial muscle samples previously analyzed at Duke were analyzed by the Beaufort Lab of the National Marine Fisheries Service (Table 7). A Perkin-Elmer atomic absorption system was used with the parameters described by Cross et al. (1973). The means and standard deviations for the four metals analyzed by the two labs agree well.

Table 5. National Bureau of Standards reference samples (bovine liver, orchard leaves, powdered coal dust) (certified values and this study's values) (ppm dry weight)

	<u>Bovine Liver</u>															
	Hg		Cd		Cr		Cu		Fe		Pb		Mn		Zn	
	NBS	DUML	NBS	DUML	NBS	DUML	NBS	DUML	NBS	DUML	NBS	DUML	NBS	DUML	NBS	DUML
$\bar{N}$		12		6		3		1		5		6		6		3
Mean	.02	.02	.27	.32	.28 <sup>a</sup>	.31	193	179	270	248	.34	.43	10	8	130	119
St.Dev.	.005	.005	.04	.04	x	.07	10	x	20	3	.08	.01	1	2	10	18
Co.Var.	25	25	15	12	x	21	5	x	7	1	24	23	10	20	8	15
	<u>Orchard Leaves</u>															
$\bar{N}$		12		4		6		7		6		7		7		7
Mean	.16	.12	.11	.11	1.7 <sup>a</sup>	1.6	12	13	300	220	45	36	91	77	25	28
St.Dev.	.02	.01	.02	.04	x	.5	1	2	20	52	3	2	4	10	3	10
Co.Var.	13	8	18	36	x	30	8	15	7	24	7	6	4	13	12	36
	<u>Powdered Coal Dust</u>															
$\bar{N}$		12		4		6		4		5		4		4		4
Mean	.12	.07	.19	.22	20	10	18	10	8.7 <sup>b</sup>	3.6 <sup>b</sup>	30	22	40	30	37	30
St.Dev.	.02	.005	.03	.04	.5	4	2	4	3.0 <sup>b</sup>	.8 <sup>b</sup>	9	3	3	4	4	2
Co.Var.	17	7	16	21	3	39	11	39	34	23	30	16	8	15	11	5

<sup>a</sup> not a certified number

<sup>b</sup> parts per thousand

x not available

Table 7. Duke University Marine Laboratory and NMFS Beaufort Lab blue marlin Makaira nigricans muscle sample intercalibration (ppm wet weight)

	Cu		Fe		Mn		Zn	
	DUML	NMFS	DUML	NMFS	DUML	NMFS	DUML	NMFS
1.	.22	.26	1.2	1.5	.03	.09	3.1	2.6
2.	.12	.21	2.1	1.4	.06	.06	2.1	2.1
3.	.23	.29	1.5	2.1	.01	.04	3.3	2.8
4.	x	.26	3.8	3.2	.02	.02	3.0	2.2
5.	.23	.25	4.6	3.8	.05	.09	4.0	4.1
6.	.07	.03	1.9	1.7	.03	.03	1.4	1.2
7.	.25	.17	3.4	3.5	.10	.07	5.4	5.1
8.	.07	.10	2.6	2.9	.07	.09	6.1	5.4
9.	.21	.14	4.0	4.3	.11	.15	4.3	4.6
10.	.23	.07	1.2	1.6	.05	.06	2.8	2.3
11.	.11	.03	1.9	1.9	.01	.05	1.4	1.2
Mean	.17	.16	2.6	2.5	.05	.07	3.4	3.1
St.Dev.	.07	.10	1.2	1.0	.03	.04	1.5	1.5
Co.Var.	42	59	47	41	69	53	45	49

The results of the mercury intercalibration experiments between Duke University Marine Laboratory and the National Marine Fisheries Service (Table 8) indicate good agreement between the two sets of data.

Table 8. Duke University Marine Laboratory and NMFS intercalibration of mercury in blue hake, Antimora rostrata, axial muscle

	Mercury (ppm)	
	DUML	NMFS
	.34 ± .02	.45 ± .02
	.63 ± .03	.71 ± .09
	.76 ± .02	.74 ± .02
	.63 ± .06	.53 ± .07
	.64 ± .06	.61 ± .09
	.42 ± .04	.49 ± .04
	.52 ± .12	.56 ± .08
	.51 ± .04	.59 ± .05
	.55 ± .05	.66 ± .12
	.55 ± .05	.68 ± .13
	.73 ± .05	.82 ± .05
	.80 ± .17	.81 ± .05
	.91 ± .05	.82 ± .13
Mean	.61	.65
St.Dev.	.16	.13
Co.Var.	26	19

Great Lakes Fishery Lab. The Great Lakes Fishery Lab provided us with frozen whole homogenized fish tissue samples which they use in every mercury analysis. The concentration ranges were ideal for quality control of our study; the fish had one low (0.1 ppm), two medium (0.4 and 0.7 ppm), and one high concentration (2.6 ppm). Considering our small sample size of 4 to 12 analyses and a large sample size of 65-290 analyses for the Great Lakes Fishery Lab, there is good agreement with the means for all four fish (Table 9).

Table 9. Duke University Marine Laboratory and Northern States Fisheries Laboratory intercalibration of mercury in total fish. Mercury shown in ppm (wet weight).

<u>#I Lake Michigan Coho Salmon</u>						
DUML	N	7	Range	.066 - .202	Mean	.12 ± .05
NSFL	N	290	Range	.056 - .141	Mean	.09 ± .01
<u>#II Lake St. Clair White Bass</u>						
DUML	N	12	Range	1.87 - 3.27	Mean	2.6 ± .71
NSFL	N	243	Range	2.24 - 3.20	Mean	2.8 ± .20
<u>#IV Ludington Lake Trout</u>						
DUML	N	12	Range	.282 - .510	Mean	.36 ± .06
NSFL	N	76	Range	.310 - .567	Mean	.40 ± .05
<u>#VI Lake Erie White Bass</u>						
DUML	N	4	Range	.650 - .844	Mean	.74 ± .10
NSFL	N	65	Range	.596 - 1.09	Mean	.77 ± .07

International Atomic Energy Agency. In Table 10 a comparison is made between data from our laboratory and data supplied by the International Atomic Energy Agency for metal concentrations in reference samples of fish solubles, calcinated animal bone and oysters. In general there was good agreement between the two sets of data.

#### Sampling in Calico Creek

Influent. Influent samples were taken from a catwalk (from 1974-76) over the middle of the primary filter of the sewage treatment plant. This material is fairly consistent because at this point the liquid is bubbling up from the bottom of the tank and it is very well mixed. At all other points within the tank settling is taking place. Influent samples taken in 1973-74 were taken in the final pump station south of Calico Creek. Because of the difficulty of obtaining samples this way, the practice was abandoned. Samples

Table 10. International Atomic Energy Agency heavy metals reference samples (fish solubles, calcinated animal bone and oysters) (ppm) intercalibration

		<u>Fish Solubles</u>															
		Hg		Cd		Cr		Cu		Fe		Pb		Mn		Zn	
		IAEA	DUML	IAEA	DUML	IAEA	DUML	IAEA	DUML	IAEA	DUML	IAEA	DUML	IAEA	DUML	IAEA	DUML
$\bar{N}$			12		6		3		9		4		4		5		5
Mean		.07	.07	X	.03	.70	.88	5	5	565	464	X	.5	5	4	19	17
St. Dev.		.02	<.005		.08	.29	.09	1	2	95	33		.1	1	1	3	3
Co. Var.		29	7		28	41	10	20	29	17	7		20	20	15	16	17
		<u>Calcinated Animal Bone</u>															
$\bar{N}$			6		4				6		5		3		4		1
Mean		X	<.05	X	.08	683	188	7	6	15 <sup>b</sup>	.9 <sup>b</sup>	7	4	32	22	183	142
St. Dev.			<.02		.04	241	20	2	1	.4 <sup>b</sup>	.2 <sup>b</sup>	1	2	5	5	12	X
Co. Var.					49	35	11	29	21	27	21	14	50	16	22	7	X
		<u>Oysters<sup>a</sup></u>															
$\bar{N}$			12		7		4		3		4		5				4
Mean		.21	.16	2.5	2.3	1.4	1.3	334	303	300	244	2.7	1.3	70	55	2.9 <sup>b</sup>	2.4 <sup>b</sup>
St. Dev.		.02	.01	.2	.2	.2	.1	5	35	10	35	.5	.3	2	5	.04 <sup>b</sup>	.1
Co. Var.		10	6	8	8	14	8	1	12	3	14	19	33	3	13	1	2

<sup>a</sup> Not a certified number

<sup>b</sup> Parts per thousand

were taken 20 cm from the surface with a 300 ml BOD bottle. The rest of the process is the same as with an effluent sample.

Effluent. Effluent water samples were taken from 1972-74 at the end of the conduit pipe that dumped the effluent into Calico Creek. Since the pipe was submerged at high tide, only low tide samples could be taken. From 1974 all effluent samples were taken inside the sewage treatment plant in the mouth of the conduit pipe connected to the chlorine contact chamber. A 300 ml BOD bottle was submerged to the depth of about 20 cm and allowed to fill. Upon return to the lab, within an hour, three 100 ml samples were subdivided from each sample and 5 ml of  $\text{HNO}_3$  and 5 ml of concentrated  $\text{H}_2\text{SO}_4$  were added to each BOD bottle. The bottles were then placed in the water bath, oxidized and vaporized as previously described. The X5 scale on the Coleman MAS-50 was used because of the low quantities of mercury present.

Sludge. Samples of the sludge "cake" were taken 0.5 m from the edge of the cement retaining wall holding the sludge. The samples were taken about 20 cm below the dry hard crust formed on the surface. This material was always spongy. The sun-dried crust was always avoided because of the possibility of losing mercury due to volatilization. Samples were transported in plastic, dried at  $70^\circ\text{C}$  and analyzed as previously stated for sediments.

Sediment. Sediment samples were collected from the top 3 cm of exposed mud at low tide (unless otherwise stated) from the various study areas. The sediment samples were scraped up and held in glass petri dishes and immediately returned to the lab for processing. The samples were then dried at  $65^\circ\text{C}$ , ground with mortar and pestle, redried at  $65^\circ\text{C}$  and weighed. Three 0.5 g and one 5 g subsamples were weighed for mercury and the other metals respectively. The samples were then analyzed as described above.

Organisms. Periwinkles, Littorina irrorata, were collected from the base of the stalks of marsh grass, Spartina alterniflora. This was done to ensure the smallest amount of sediment being taken into the apex of the shell when the animal closed its operculum after removal from the stalk. Animals were transported back to the lab in plastic bags and frozen or immediately analyzed. Shells and operculums were washed and lightly scrubbed to remove any sediment or algal growth adhering to the shell. The shells of the animal were broken using pliers, and the entire animal with the operculum was carefully removed. The operculum was left attached to avoid rupturing the tissues. Prior to weighing, the animals were placed on paper towels and blotted dry. After weighing, one whole animal was placed in a 300 ml BOD bottle. Three replicate samples were analyzed for mercury. The results reported are the means and standard deviation of three animals collected within 0.5 square meter of each other. Other metals were determined using five individuals taken from the same 0.5 square meter. Each value for metals other than mercury represents one determination for the five combined animals from a single station. Animals were dried to constant weight at  $90^\circ\text{C}$  for at least 48 hr prior to ashing at  $500^\circ\text{C}$ . The addition of 5 ml of concentrated  $\text{HNO}_3$ , heating, filtering and storage of samples prior to analysis was the same as the method described for sediment.

Oysters, Crassostrea virginica, were collected and analyzed in the same manner as described for Littorina irrorata. The soft tissues of oysters were removed from their shells and analyzed. Values reported for mercury are the means of the analyses of three replicates. Concentrations of the other metals are based on a single analysis of five oysters from an individual sample station.

Marsh grass, Spartina alterniflora, was collected within the study area in June 1976. Plants were washed with deionized water to remove external material, separated into two sections (stems and roots) and ground in a Wiley Mill. Five grams of stems and two grams of roots were weighed, combusted, digested and analyzed as previously described.

## RESULTS AND DISCUSSION

### Calico Creek

Surface sediment samples and organisms were collected from 1972 to 1976 in Calico Creek to determine the quantity and distribution of metals. In May 1972 eight stations above the outfall and seven stations below the outfall were sampled for total mercury. Results are reported in Table 11 and Figure 6. In May 1974 a more extensive sampling took place. Analyses were carried out for cadmium, chromium, copper, iron, lead, manganese and zinc in addition to mercury; the results are presented in Tables 12 and 13 and Figures 7 through 13. In July 1975 surface sediments and organisms were analyzed from seven stations above the outfall and eight stations below the outfall (Tables 14-16 and Figures 14 through 22). The final surface sediment and animal samples were taken in January 1976. Eleven stations were sampled above the outfall and twelve below the outfall. The results of these analyses are given in Tables 17 and 18 and Figures 23 through 31.

The concentrations of mercury in the top 3 cm of sediment in the 1972 stations in Calico Creek are listed in Table 11. The highest concentrations of mercury occurred 25 m above (1.4 ppm) and 25 m below (1.7 ppm) the outfall. There was a good correlation between mercury concentration and distance above ( $r=-0.953$ ) and below ( $r=-0.868$ ) the outfall, establishing that the effluent discharge is the most probable source of the "excess" mercury in the sediments. The mean value of 0.89 ppm for the stations above the outfall is higher than the mean value of 0.71 for the stations below the outfall, reflecting the tidal influences. The common periwinkle snail, Littorina irrorata, had a maximum mercury concentration of 0.56 ppm wet weight 25 m above and 0.62 ppm 10 m below the outfall (Table 11). These stations also had elevated mercury in the sediments. The correlations of mercury in L. irrorata with distance above and below the outfall ( $r=-0.726$  and  $r=-0.926$ , respectively) were similar to the sediment vs distance correlation. There was a correlation between L. irrorata mercury and sediment mercury of  $r=0.718$  above and  $r=0.979$  below the outfall. L. irrorata was the only organism that was consistently common in the study area. L. irrorata was collected from the stems of Spartina alterniflora to eliminate the possibility of mercury-rich mud being trapped in the digestive tract of the gastropod. Investigation showed this not to be a problem; twelve L. irrorata feeding on the exposed mud had a mean mercury concentration of  $0.24 \pm 0.07$  ppm, not differing from twelve L. irrorata collected from the stems of S. alterniflora having a mean of  $0.22 \pm 0.04$  ppm.

Mercury concentrations in oysters, Crassostrea virginica, collected in 1972 below the outfall had a strong correlation with distance from the outfall ( $r=-0.917$ ) (Table 11); however, the highest concentration ( $0.13 \pm 0.05$  ppm at 10 m) is only 20% of the highest value in the periwinkle. C. virginica mercury concentrations were strongly correlated with mercury content in the sediment ( $r=0.912$ ).

The organic content of the sediment increased with distance ( $r=0.674$ ) above the outfall in the 1974 sampling (Table 12 and Figure 7). All metals decreased with distance above the outfall. Heavy metal patterns below the

Table 11. Mercury concentrations found in the top 3 cm of surface sediment (ppm dry weight), in the snail *Littorina irrorata* (ppm wet weight) and in the oyster *Crassostrea virginica* (ppm wet weight) from stations ranging from 900 meters above the outfall to 1450 meters below the outfall in Calico Creek - May 1972. Each sediment value is the mean of three replicates.

<u>Meters above the outfall</u>	<u>Sediment</u>	<u>Littorina</u>	<u>Crassostrea</u>
900	.08 ± .04	.24 ± .08	
400	.68 ± .19	.21 ± .03	
350	.90 ± .18	.25 ± .07	
300	.85 ± .04	.26 ± .06	
225	1.1 ± .08	.30 ± .02	
150	.90 ± .18	X	
25	1.42 ± .17	.56 ± .04	
10	1.16 ± .08	.54 ± .03	
Mean	.89	.34	
St.Dev.	.40	.15	
Co.Var.	45	44	
r(distance)	-.953	-.726	
		r(sed vs <u>Lit</u> )	.718
<u>Meters below the outfall</u>			
10	1.60 ± 10	.62 ± .05	.13 ± .05
25	1.66 ± .21	.57 ± .02	
350	.56 ± .04	.33 ± .05	.09 ± .02
800	.35 ± 11	.24 ± .09	
900	.33 ± .05	.20 ± .05	
1400	.24 ± .03	.12 ± .04	.07 ± .02
1450	.26 ± .01	.18 ± .01	.04 ± .02
Mean	.71	.32	.08
St.Dev.	.63	.20	.04
Co.Var.	89	61	46
r(distance)	-.868	-.926	-.917
		r(sed vs <u>Lit</u> )	.979
		r(sed vs <u>Crass</u> )	.912

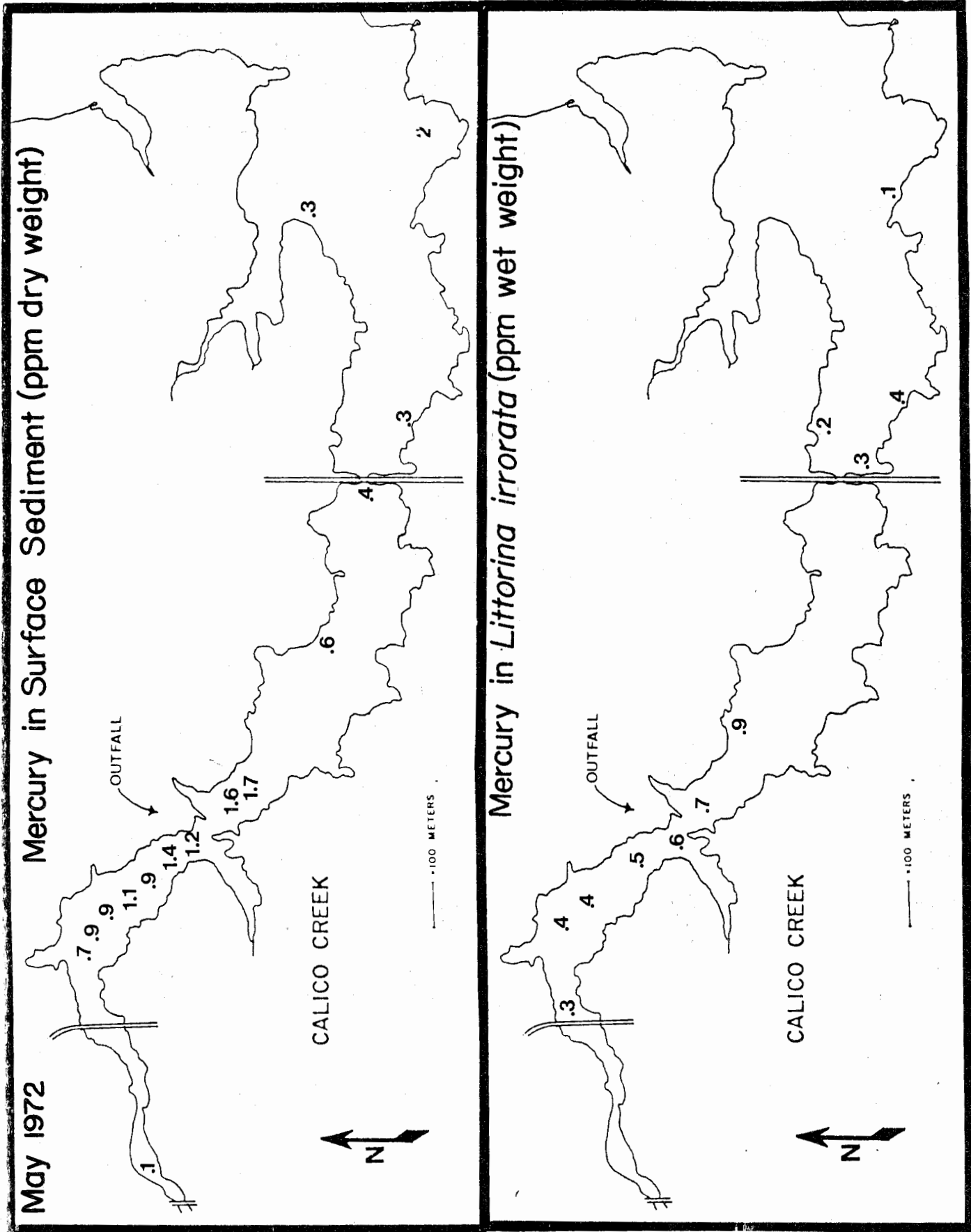


Fig. 6. Total mercury found in Calico Creek during 1972

Table 12. Heavy metal concentrations (ppm dry weight) and percent loss on ignition values (an estimate of organic matter) in the top 3 cm of surface sediment from stations above and below the outfall in Calico Creek - May 1974.

Meters above Outfall	% Loss on Ignition	Hg	Cd	Cr	Cu	Fe (X1000)	Pb	Mn	Zn
985	31	.10 ± .01	.3	8	7	11.0	11	11	64
875	33	.14 ± .18	1.3	9	17	20.0	10	x	x
770	25	x	1.6	20	26	21.6	55	104	149
765	26	.40 ± <.00	1.2	30	47	4.0	51	21	71
700	19	.45 ± .04	.9	22	43	x	20	9	190
655	19	.45 ± .07	.9	11	43	x	42	x	167
605	21	.28 ± .04	.8	37	38	15.7	42	56	144
595	23	.29 ± .03	1.3	5	43	9.0	53	75	122
555	22	.38 ± .04	1.2	15	42	9.8	14	13	115
550	13	.18 ± .02	.3	29	32	11.8	52	60	177
540	14	x	2.4	28	22	21.0	52	118	186
525	19	.40 ± .01	1.1	30	36	x	52	x	170
475	20	.39 ± .02	.8	26	28	11.5	43	x	166
180	20	.50 ± .05	1.3	47	55	9.0	32	22	180
120	18	.99 ± .02	1.4	28	32	11.0	56	102	176
90	19	.54 ± .02	.6	34	46	20.5	35	133	164
80	15	.85 ± .11	1.9	19	35	10.6	67	46	188
70	18	.66 ± .02	.8	55	48	23.5	46	130	180
25	16	.80 ± .04	1.8	12	43	6.6	59	74	141
10	17	.40 ± .05	1.0	47	44	26.9	40	132	193
Mean	20	.46	1.1	26	36	14.3	42	69	155
St. Dev.	5	.24	.5	14	12	6.6	17	46	38
Co. Var.	25	53	45	54	32	46	40	67	24
r(distance)	.674	-.769	-.208	-.542	-.554	-.148	-.452	-.531	-.572
r(% Loss vs metal)		-.568	-.210	-.408	-.427	-.068	-.531	-.417	-.825

Table 12. (continued)

Meters below Outfall	% Loss on Ignition	Hg	Cd	Cr	Cu	Fe (X1000)	Pb	Mn	Zn
30	20	.55 ± .06	1.2	25	30	18.3	21	97	101
100	19	.50 ± .03	1.4	14	40	20.0	30	55	291
200	15	x	1.2	11	34	13.7	42	115	120
430	x	.31 ± .09	.9	14	27	6.5	33	99	125
500	14	.34 ± .01	1.3	45	31	24.0	41	135	259
600	14	.51 ± .10	1.4	x	22	6.6	8	x	151
700	14	.31 ± .08	.6	6	6	20.0	20	70	92
720	14	.35 ± .04	.5	5	8	42.2	x	27	60
755	13	.18 ± .01	.7	8	16	6.8	x	15	111
800	12	.14 ± .03	.6	13	18	12.3	21	13	115
810	14	.16 ± .02	.6	32	18	15.0	30	8	129
815	14	x	.7	x	15	8.0	23	159	175
845	8	.40 ± .08	1.2	5	21	14.6	50	77	82
950	17	.27 ± .06	1.3	25	20	x	25	64	101
1000	12	.15 ± .02	.6	20	18	4.8	6	77	100
1050	13	.21 ± .02	.4	21	21	8.5	13	16	85
1050	14	.31 ± .04	1.1	7	18	9.5	30	37	105
1200	12	.08 ± .02	.7	20	22	16.0	26	123	90
1320	10	.20 ± .04	.6	15	17	12.0	27	14	185
1320	11	.23 ± .09	.7	2	12	3.6	6	11	74
1400	9	.10 ± .05	.7	9	18	4.5	25	42	82
1550	9	.07 ± .03	.4	13	17	6.3	20	21	90
1800	1	.03 ± .01	.3	3	12	5.1	10	44	44
Mean	13	.26	.8	15	20	12.7	24	60	120
St.Dev.	4	.15	.3	11	8	9.0	12	45	59
Co.Var.	31	58	42	71	41	70	49	76	49
r(distance)	-.828	-.822	-.667	-.315	-.631	-.437	-.412	-.421	-.499
r(% Loss vs metal)		.692	.584	.422	.496	.431	.168	.260	.455

Table 13. Heavy metal concentrations in the snail Littorina irrorata (ppm wet weight) at individual stations above and below the outfall in Calico Creek - May 1974

<u>Meters above</u>		<u>Hg</u>	<u>Cd</u>	<u>Pb</u>	<u>Zn</u>
10	.6	.56 ± .18	.5	<.9	76
120	.6	.50 ± .09	3.2	<.2	50
180	.5	.38 ± .01	1.1	3.7	60
475	.6	.35 ± .01	.4	10.0	14
555	.7	.28 ± .05	.1	X	8
Mean	.6	.41	1.1		42
St.Dev.	.1	.11	1.3		30
Co.Var.	12	28	118		71
r(distance)		-.924	-.500		-.976
r(metal vs <u>Littorina</u> )		.427	.671		.840
<u>Meters below</u>					
30	.5	.68 ± .07	.3	<1.0	66
200	.6	.89 ± .10	.8	X	79
845	.6	.23 ± .04	1.5	9.0	44
950	.6	.25 ± .07	.5	<1.0	64
1000	.5	.38 ± .05	.5	11.0	64
1550	.4	.11 ± .05	.56	9.0	49
Mean	.5	.42	.7		61
St.Dev.	.1	.30	.4		13
Co.Var.	15	71	62		21
r(distance)		-.898	.090		-.659
r(metal vs <u>Littorina</u> )		.734	.222		.980

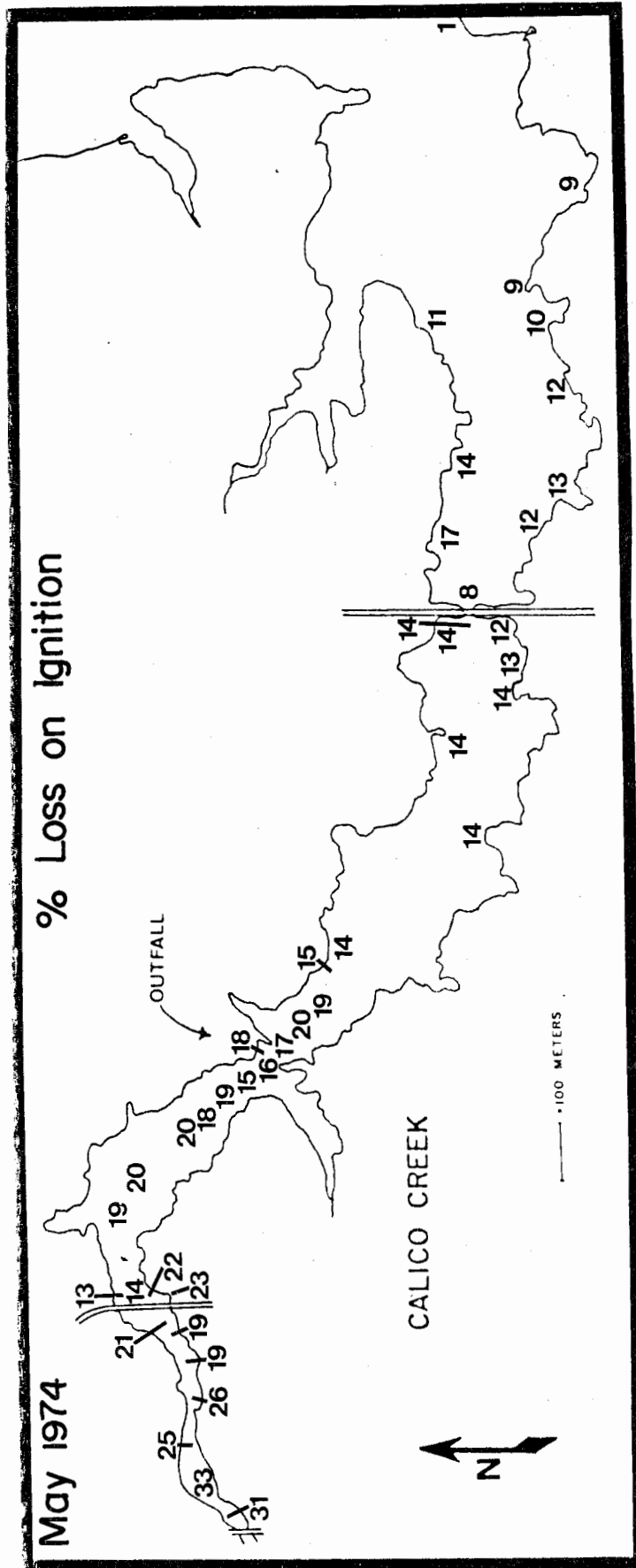


Fig. 7. Percent loss on ignition in Calico Creek for 1974



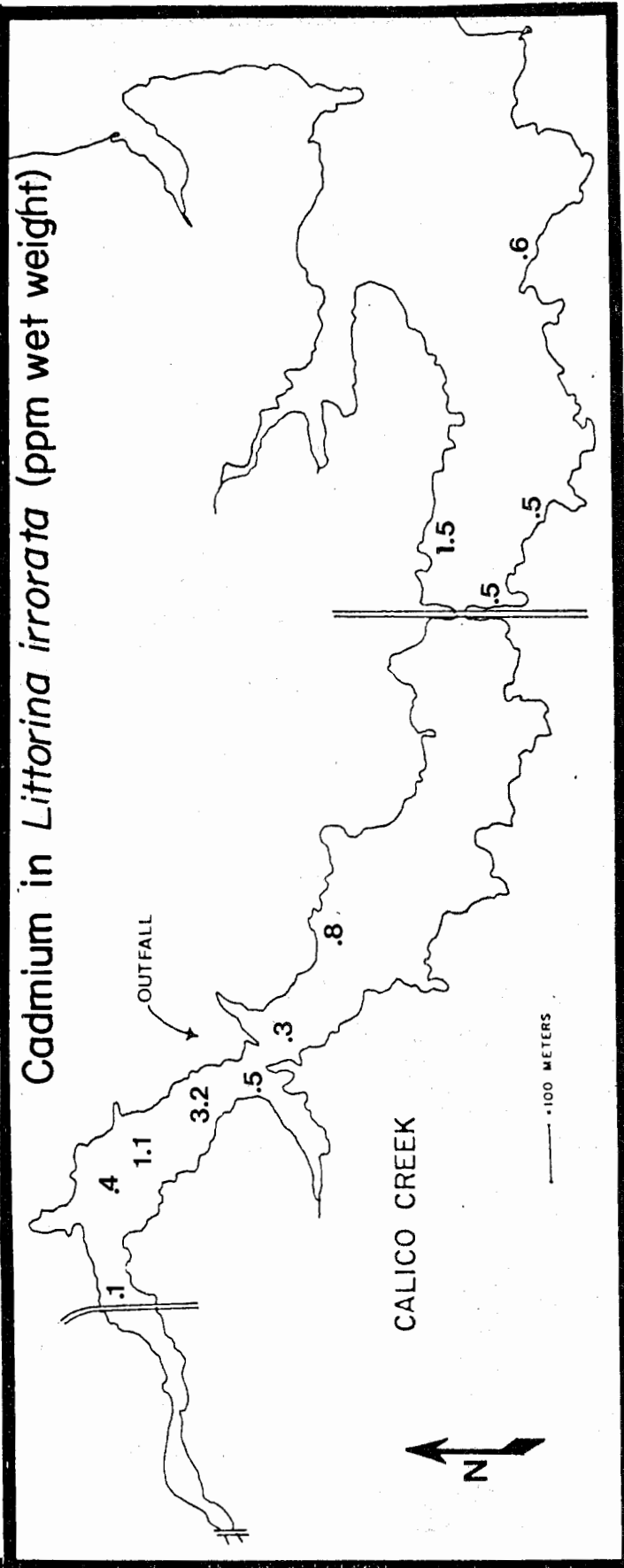
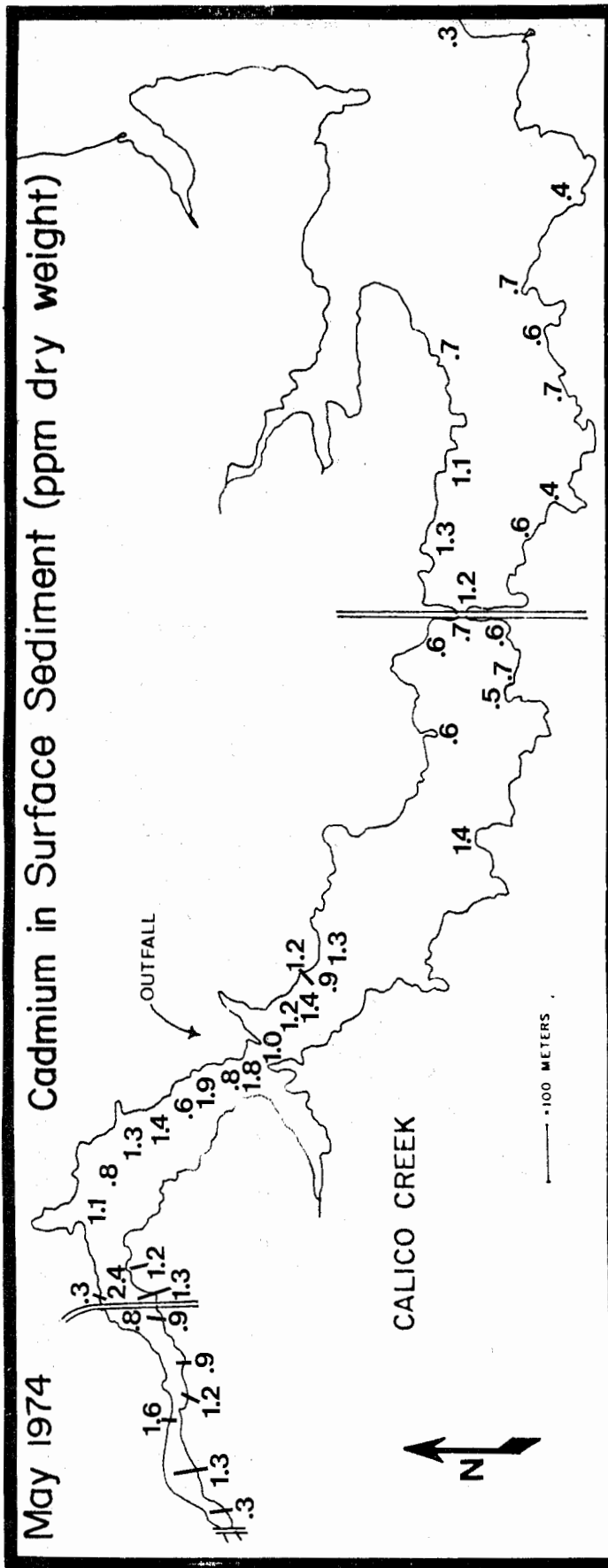


Fig. 9. Cadmium found in Calico Creek during 1974

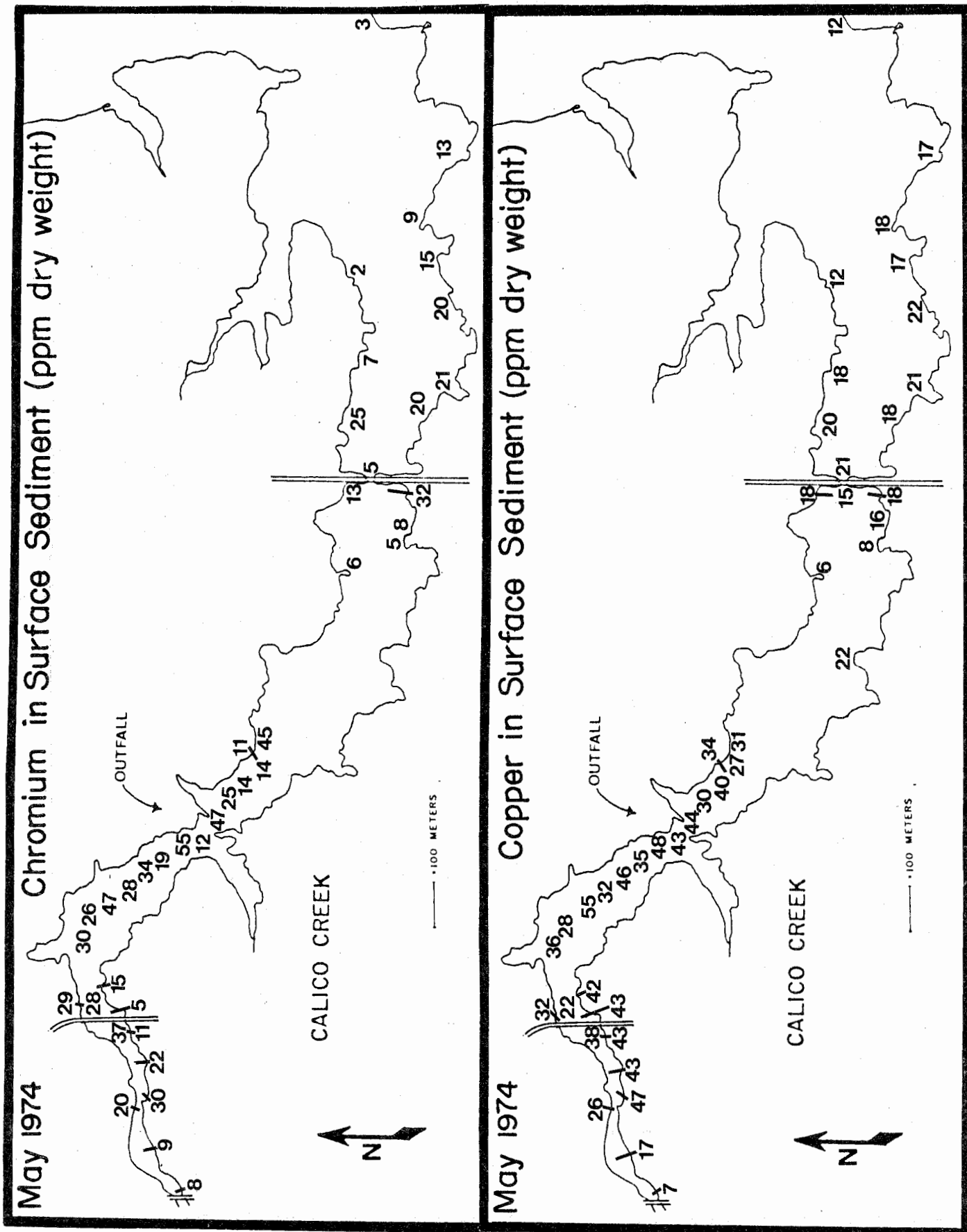
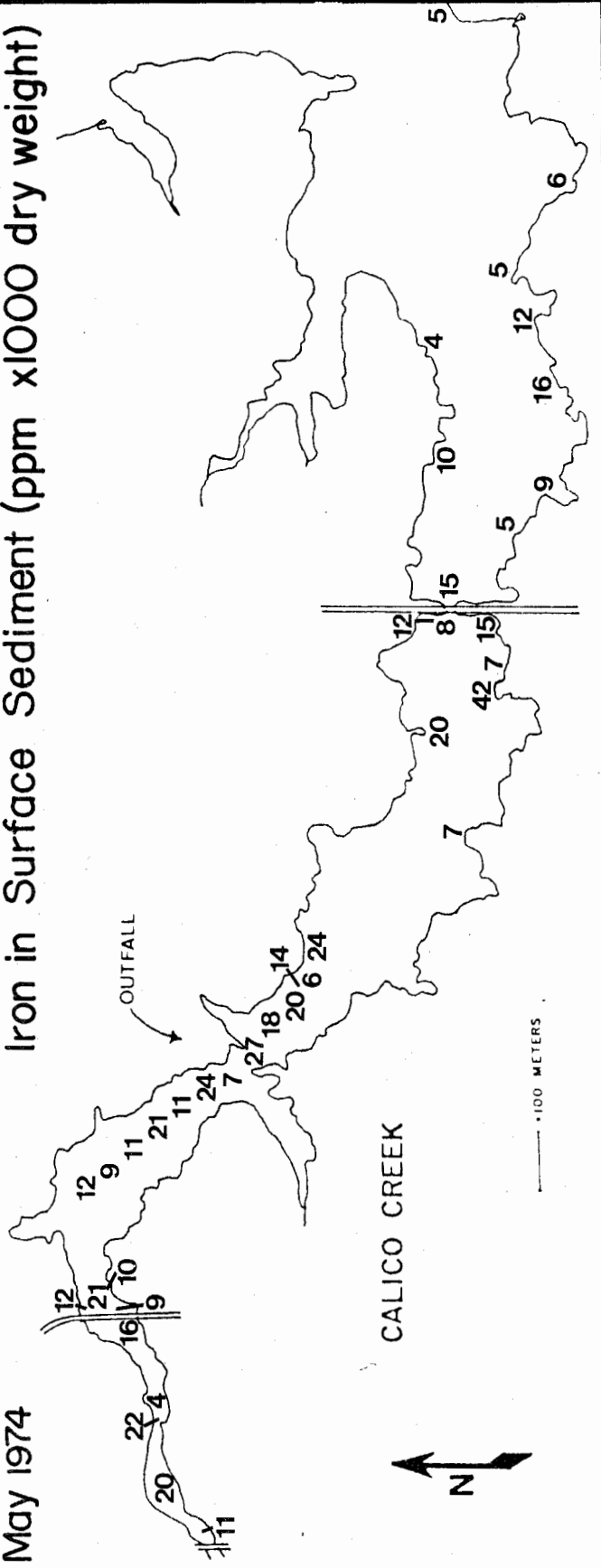


Fig. 10. Chromium and copper found in Calico Creek during 1974

Iron in Surface Sediment (ppm x1000 dry weight)

May 1974



Manganese in Surface Sediment (ppm dry weight)

May 1974

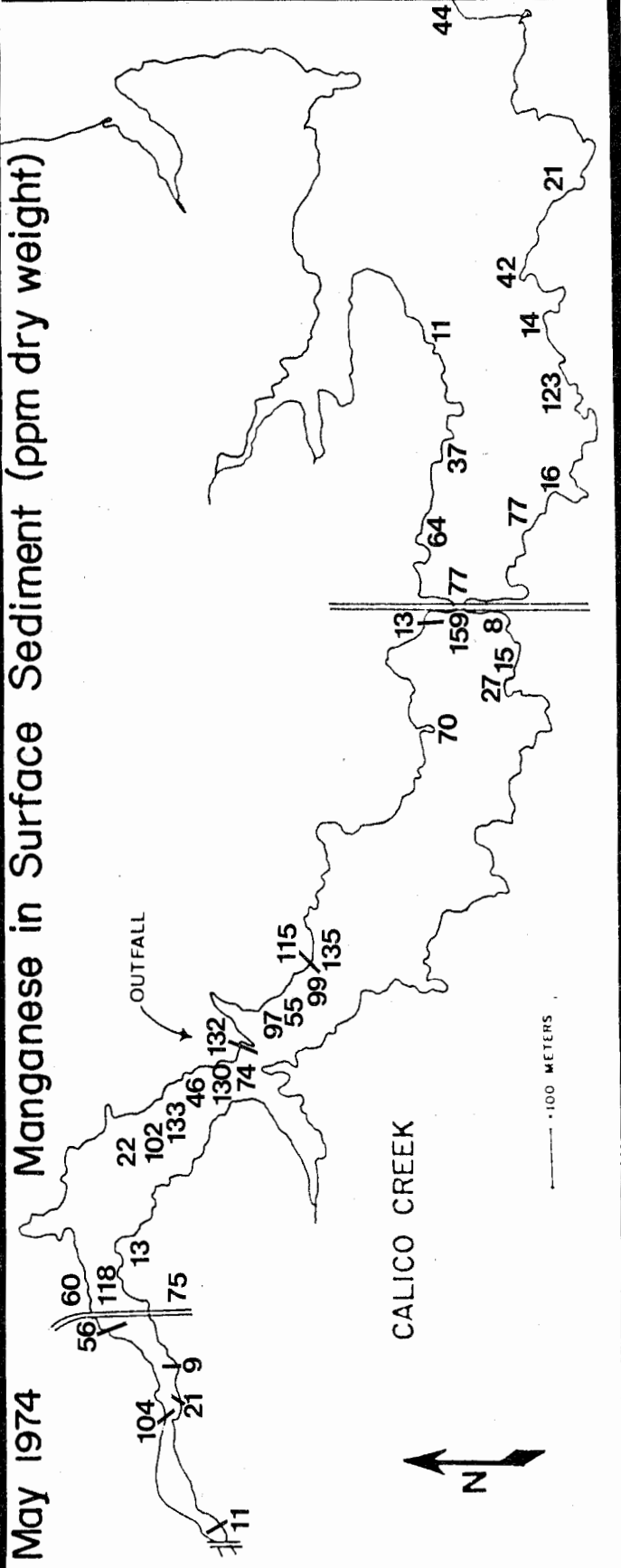


Fig. 11. Iron and manganese found in Calico Creek during 1974

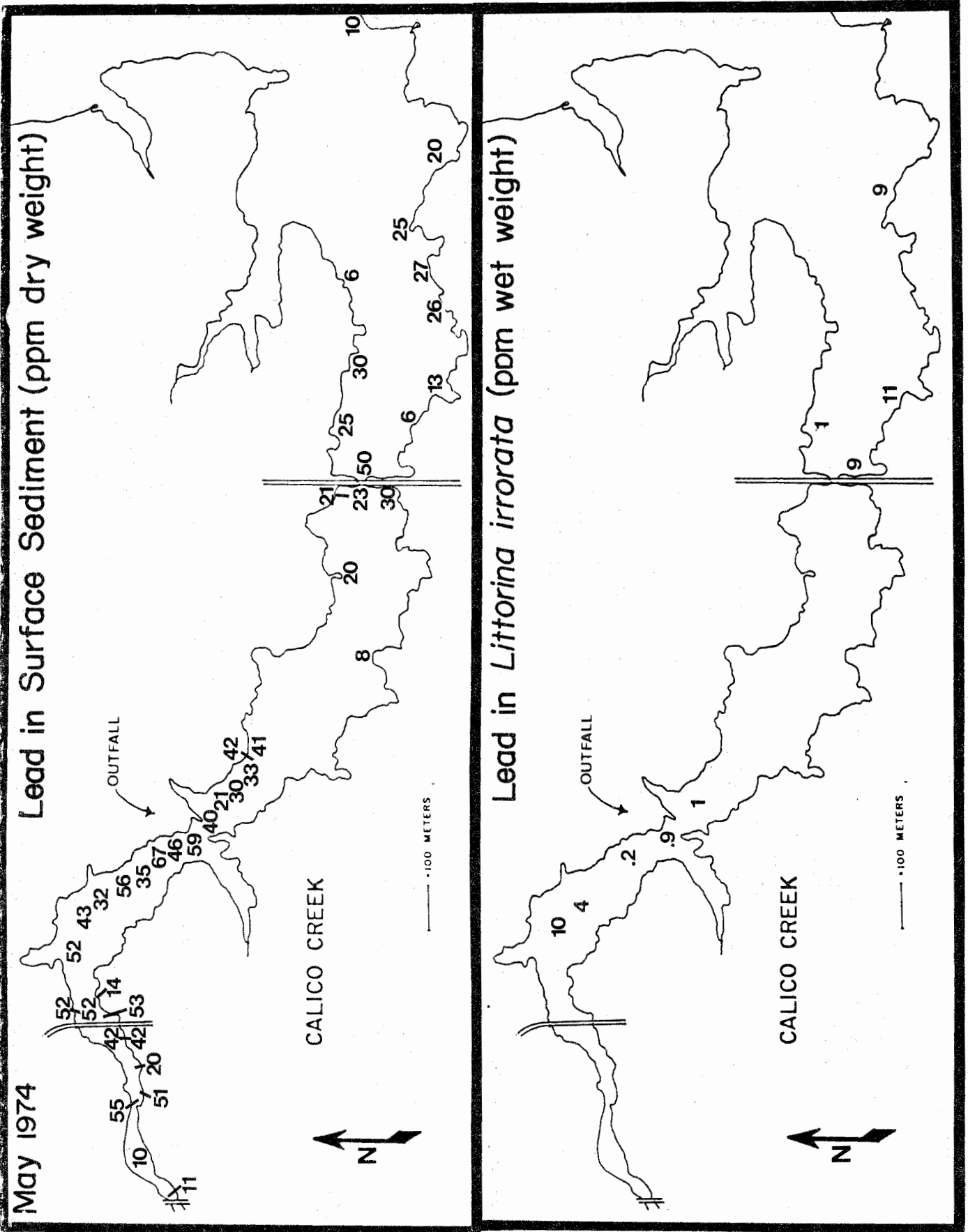


Fig. 12. Lead found in Calico Creek during 1974

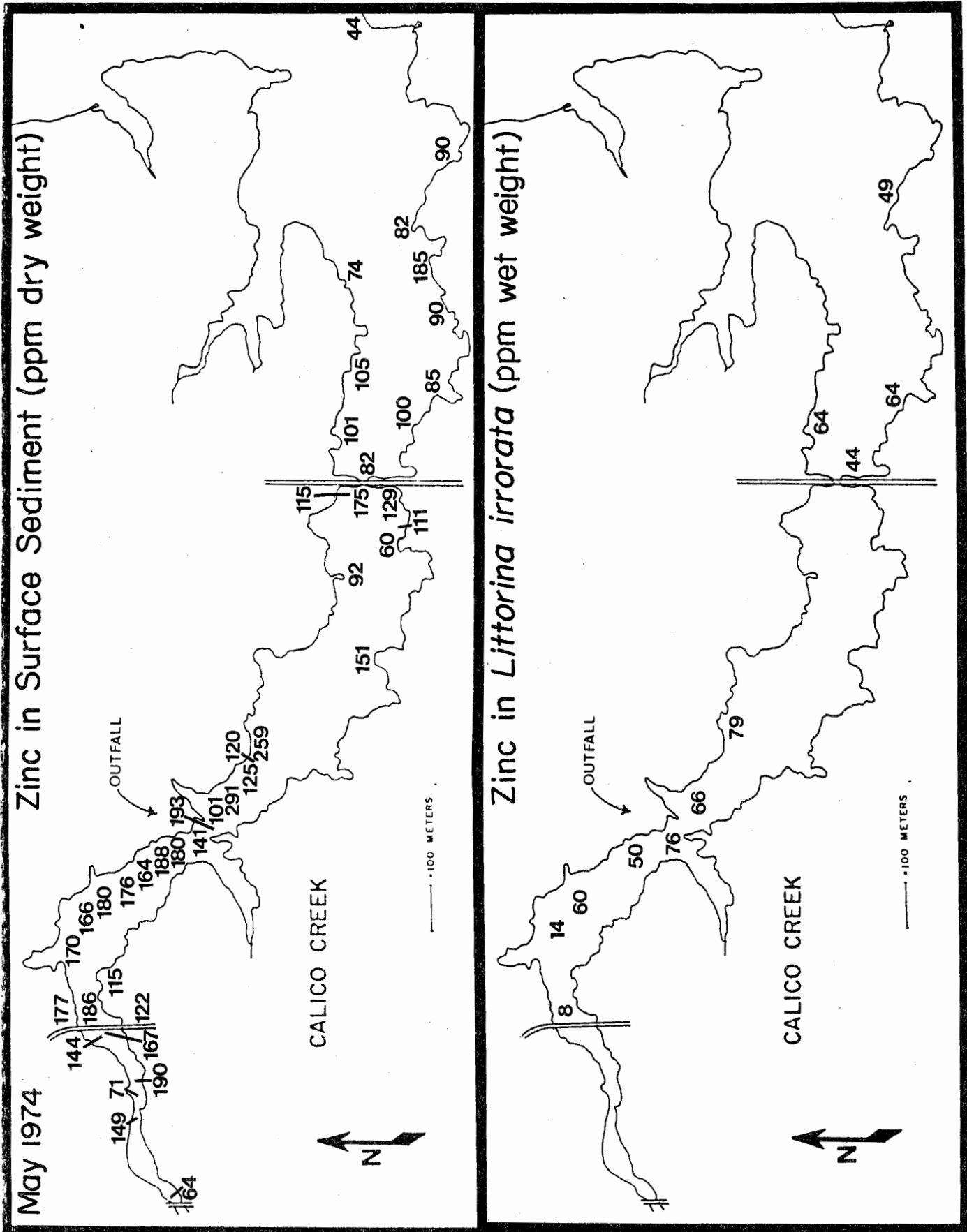


Fig. 13. Zinc found in Calico Creek during 1974

Table 14. Heavy metal concentrations (ppm dry weight) and percent loss on ignition values (an estimate of the organic matter) in the top 3 cm of surface sediment from individual stations above and below the outfall in Calico Creek - July 1975

Meters above Outfall	% Loss on Ignition	Hg	Cd	Cr	Cu	Fe X1000	Pb	Mn	Zn
10	18	.31 ± .02	1.0	6	16	2.9	7	13	115
300	X	.48 ± .01	.9	19	43	5.5	15	85	170
350	15	.55 ± .08	.9	20	18	X	15	84	139
425	11	.49 ± .02	1.1	26	37	X	25	X	150
475	20	.38 ± .10	1.0	12	31	6.5	19	58	152
540	20	.36 ± .08	.8	16	36	8.2	19	77	117
605	21	.39 ± .09	.8	19	32	5.5	10	84	149
Mean	18	.42	.9	17	30	5.7	16	67	142
St.Dev.	4	.09	.1	6	10	1.9	6	28	20
Co.Var.	22	20	12	38	33	34	38	42	14
r(Distance)	.253	.148	-.445	.538	.536	.806	.461	.746	.288
r(% loss vs metal)		-.696	-.481	-.599	.011	.654	-.527	.495	-.219
Meters below Outfall									
30	19	.35 ± .06	.7	5	18	.9	2	40	150
100	15	.29 ± .02	.7	6	25	7.2	9	36	125
200	17	.25 ± .01	1.3	7	14	5.9	4	16	104
650	13	.23 ± .02	.6	12	19	11.5	13	48	80
800	29	.18 ± .01	.6	7	16	5.0	13	78	80
860	13	.17 ± .03	.6	13	16	X	4	89	70
900	17	.15 ± .02	.6	20	22	12.0	6	64	101
1050	10	.15 ± .03	.5	5	10	2.7	3	27	89
Mean	17	.22	.7	9	18	6.5	7	49	100
St.Dev.	6	.07	.3	5	5	4.2	4	26	27
Co.Var.	35	33	36	56	27	65	65	54	27
r(distance)	-.136	-.951	-.582	.469	-.402	.284	.131	.561	-.664
r(% loss vs metal)		.101	.035	-.117	.109	-.159	.573	.349	.081

Table 15. Heavy metal concentrations in the snail *Littorina irrorata* (ppm wet weight) at individual stations above and below the outfall in Calico Creek - July 1975

<u>Meters Above Outfall</u>	<u>Ave Wt</u>	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe</u>	<u>Pb</u>	<u>Mn</u>	<u>Zn</u>
10	.9	.29 ± .04	.5	.3	105	174	.6	11	53
300	1.4	.14 ± .06	.4	.7	91	375	1.3	10	23
350	1.4	.20 ± .09	.3	.4	83	115	.8	9	23
425	1.5	.19 ± .10	.4	.8	126	356	1.3	12	29
475	1.1	.19 ± .02	.6	1.6	221	315	2.5	6	50
540	1.1	.25 ± .03	6.3	.6	60	292	6.5	4	36
605	1.2	.15 ± .03	.5	1.7	94	293	.8	9	63
Mean	1.2	.20	1.3	.9	111	274	2.0	9	40
St.Dev.	.2	.05	2	.6	52	95	2.1	3	16
Co.Var.	17	26	172	64	47	35	106	32	40
r(distance)		-.532	.350	.681	.061	.431	.437	-.531	.111
r(sed vs <i>L. irrorata</i> )		-.565	-.483	.101	.011	.591	.401	-.305	-.267
<u>Meters Below Outfall</u>									
30	1.2	.20 ± .03	.5	.8	185	322	.6	11	48
100	1.0	.31 ± .08	.5	.4	111	305	1.0	12	34
200	.8	.30 ± .20	.3	.6	67	303	1.8	14	26
650	1.0	.15 ± .05	.4	.4	57	358	.6	25	35
800	.9	.38 ± .20	.7	4.1	81	338	2.3	10	46
860	.8	.26 ± .05	.4	1.5	693	134	.6	11	28
900	1.0	.22 ± .07	.5	.4	78	228	.7	2	19
1050	.8	.11 ± .03	.7	.8	459	225	3.5	10	36
Mean	.9	.24	.5	1.1	216	277	1.4	12	34
St.Dev.	.1	.09	.1	1.3	234	75	1.1	6	10
Co.Var.	15	37	28	112	108	27	77	50	29
r(distance)		-.270	-.511	.258	.435	-.517	.451	-.233	-.272
r(sed vs <i>L. irrorata</i> )		.099	-.645	-.190	-.455	-.001	-.083	-.243	.302

Table 16. Heavy metal concentrations (ppm wet weight) in the oyster Crassostrea virginica at individual stations above and below the outfall in Calico Creek - July 1975

	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe</u>	<u>Pb</u>	<u>Mn</u>
10 m above	X	1.3	11	27	81	1	4
150m below	.02 ± .01	.1	.1	9	39	.2	2
650m below	<.0005 ± <.0005	.2	<1	5	46	1	2
870m below	.01 ± .01	.1	.2	8	53	.2	2
1050m below	.01 ± .01	X	.2	6	59	.3	2
1320m below	.01 ± .01	.1	.2	9	41	.1	2
Mean	.01	.4	X	11	53	.5	2.3
St.Dev.	.01	.5	X	8	16	.4	.8
Co.Var.	100	146	X	77	29	90	35
r(distance)	-.623	-.623	X	-.630	-.416	-.541	-.636
r(metal vs <u>Crass</u> )	-.586	-.945	X	-.030	-.583	-.539	-.459

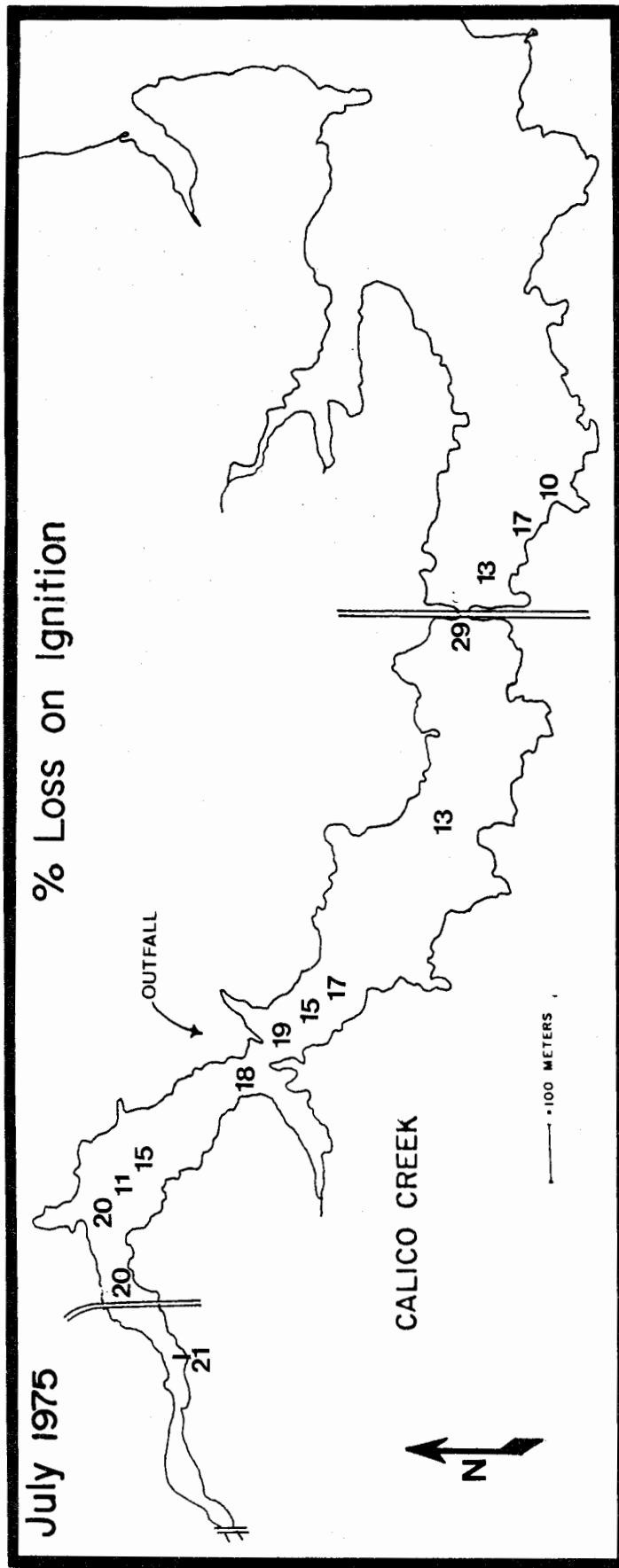


Fig. 14. Percent loss on ignition in Calico Creek during 1975

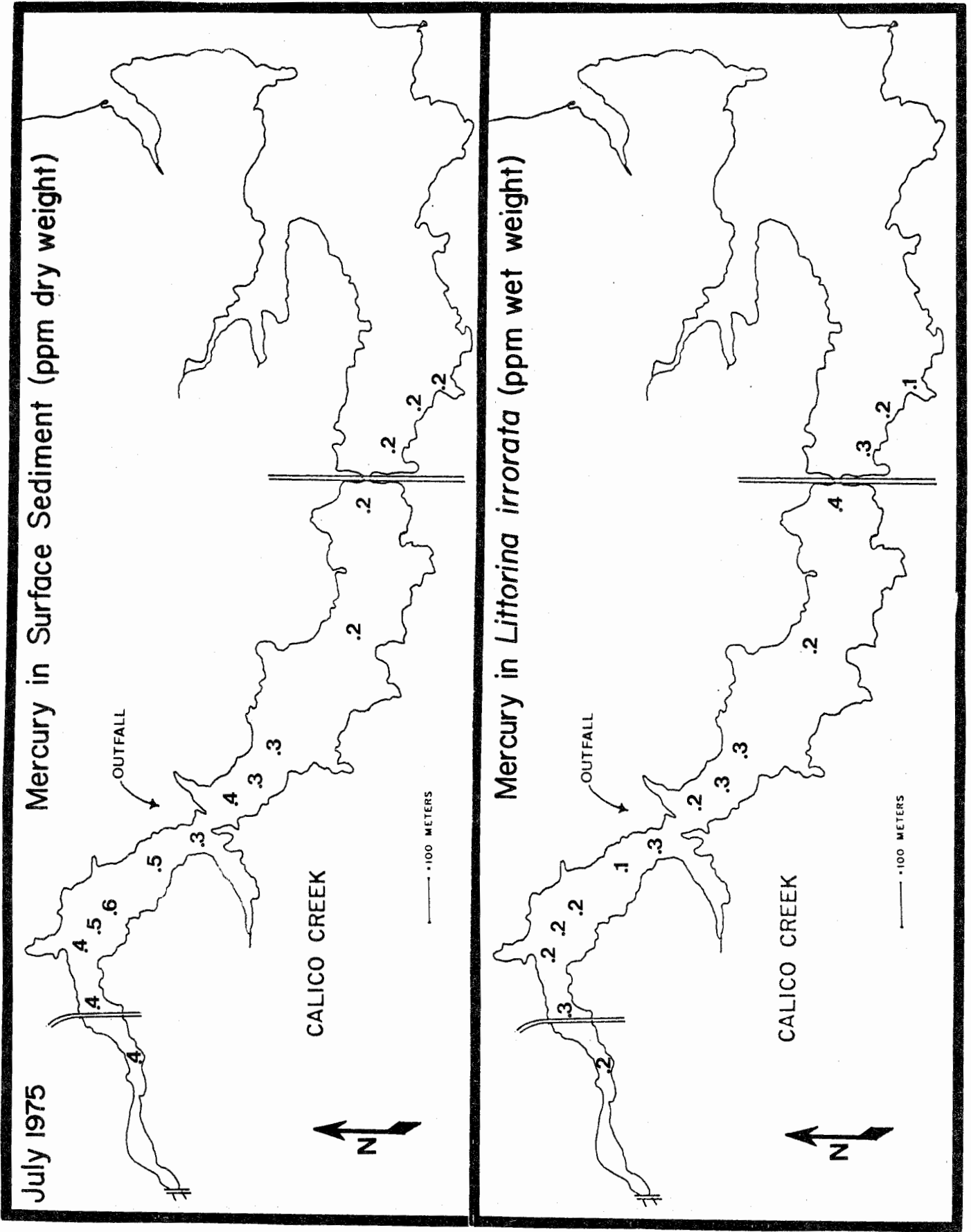


Fig. 15. Total mercury found in Calico Creek during 1975

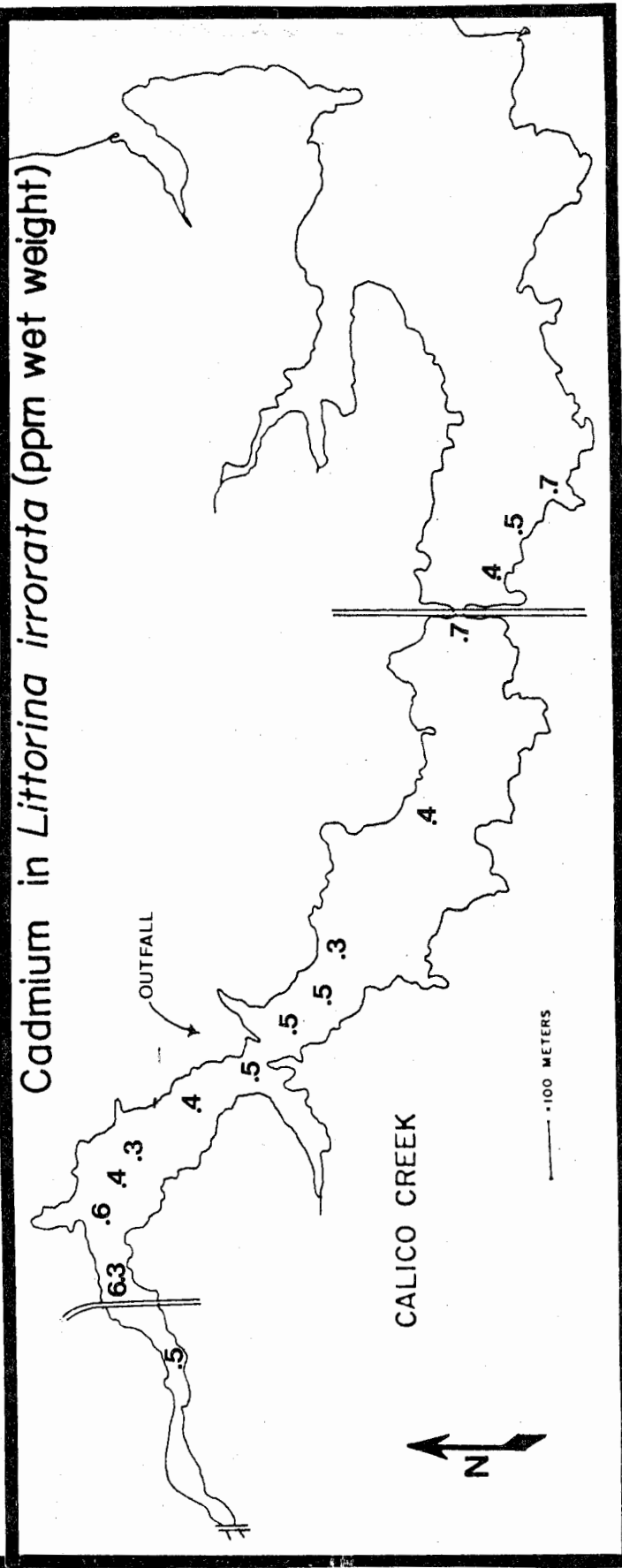
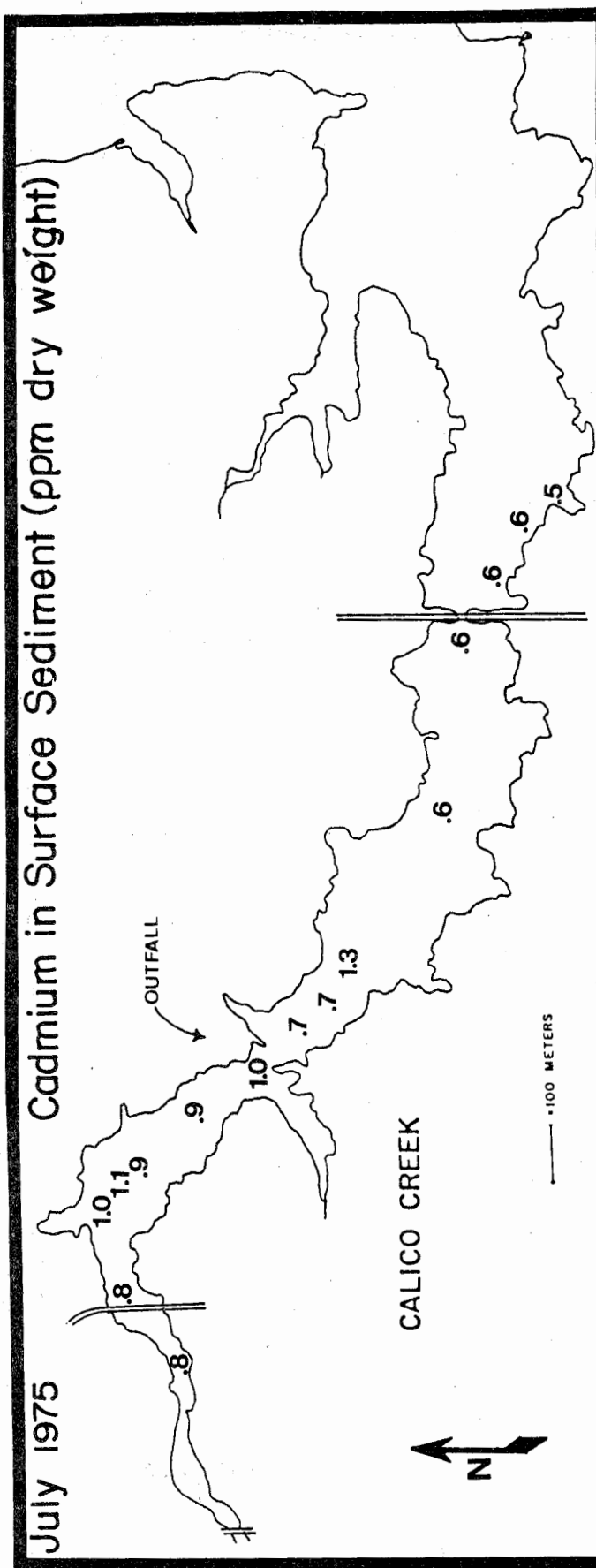
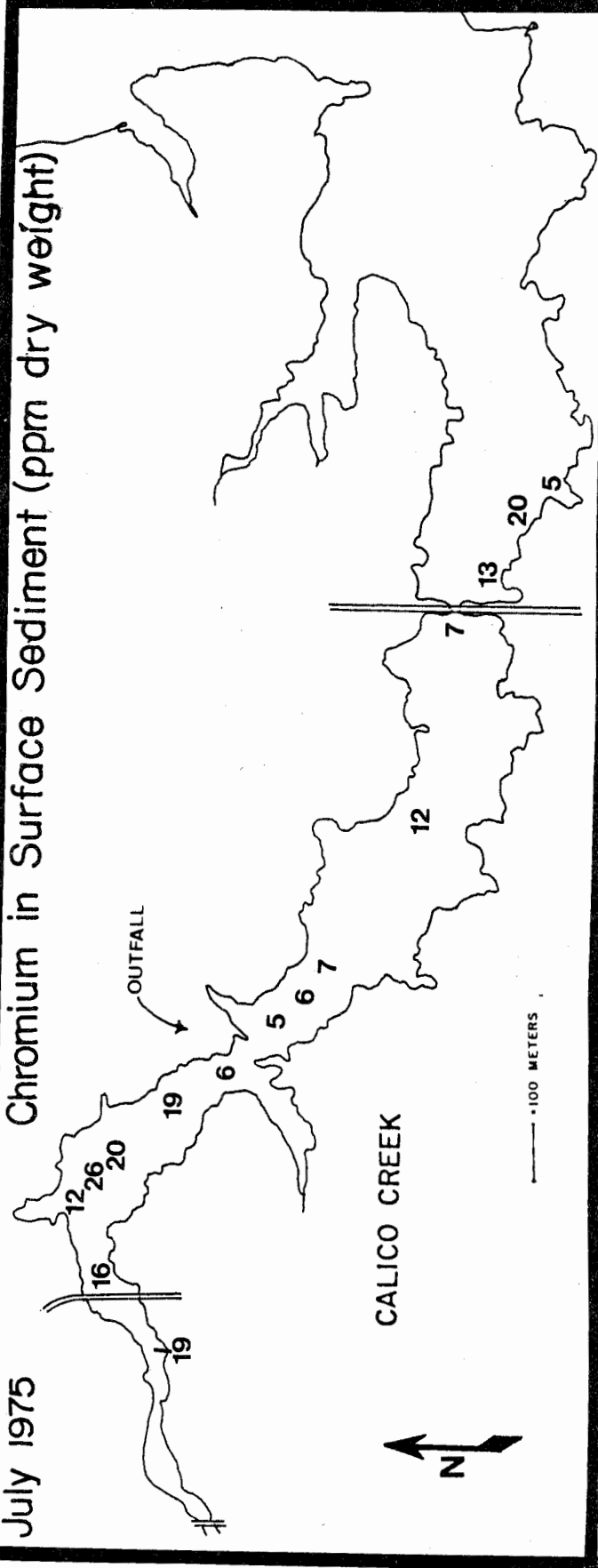


Fig. 16. Cadmium found in Calico Creek during 1975

July 1975

Chromium in Surface Sediment (ppm dry weight)



Chromium in *Littorina irrorata* (ppm wet weight)

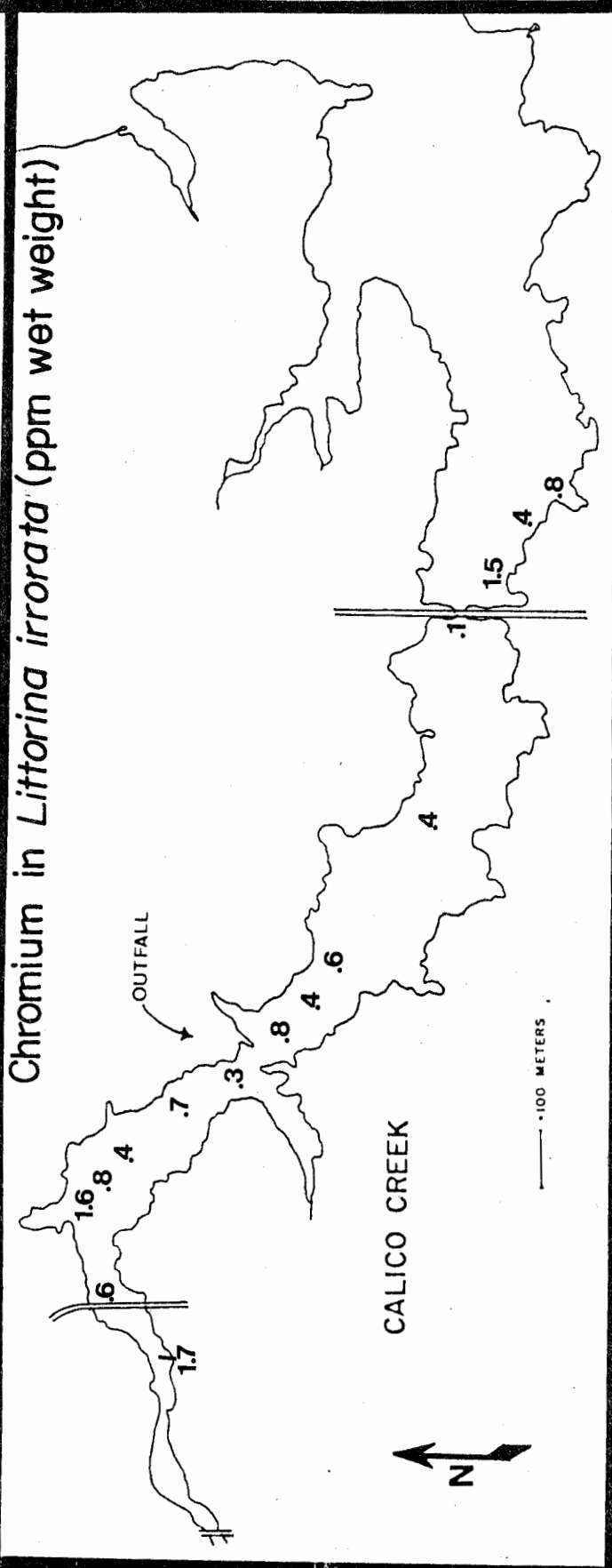


Fig. 17. Chromium found in Calico Creek during 1975

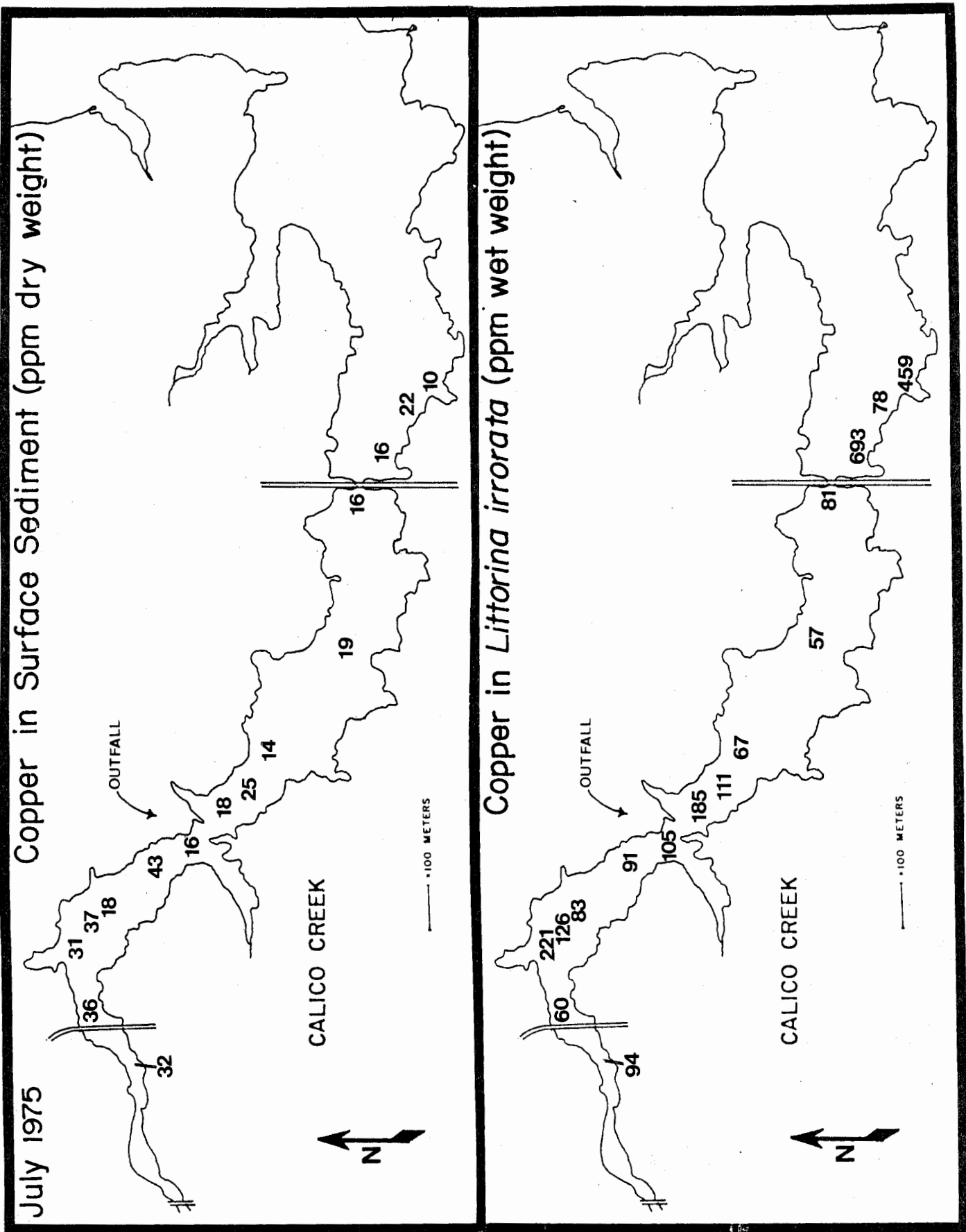
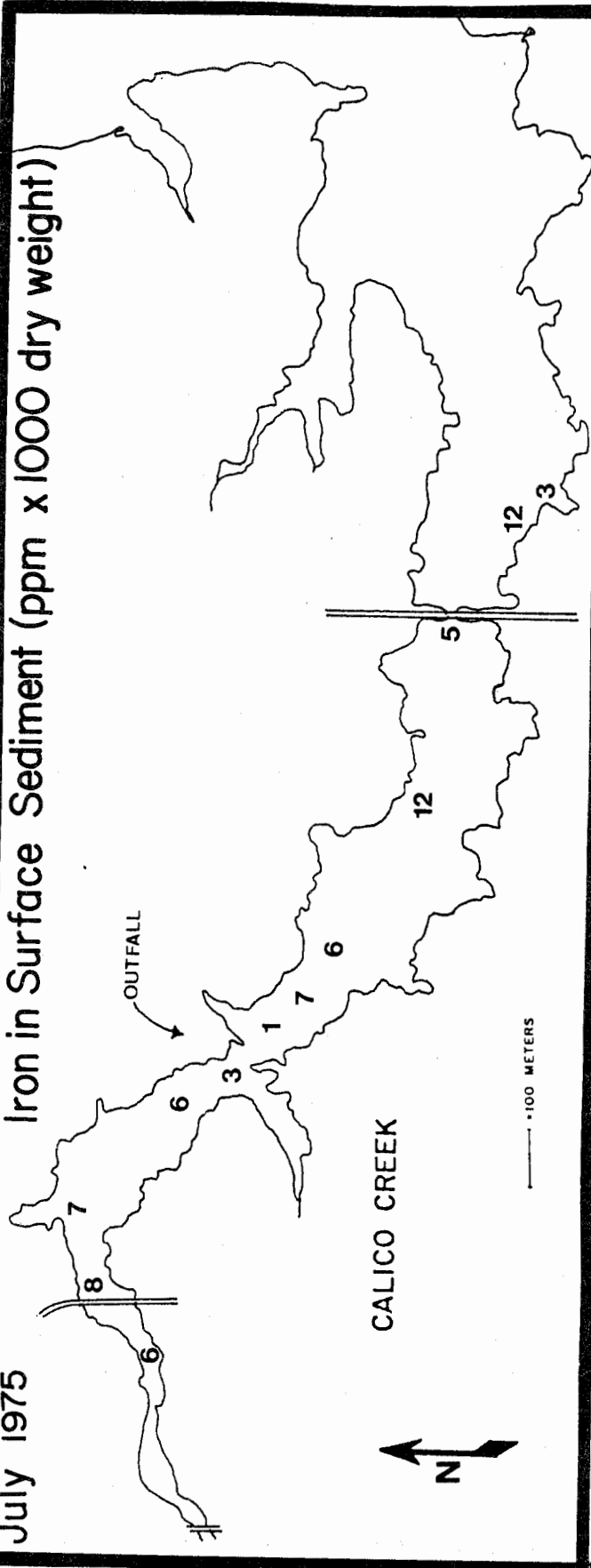


Fig. 18. Copper found in Calico Creek during 1975

July 1975

Iron in Surface Sediment (ppm x 1000 dry weight)



Iron in *Littorina irrorata* (ppm wet weight)

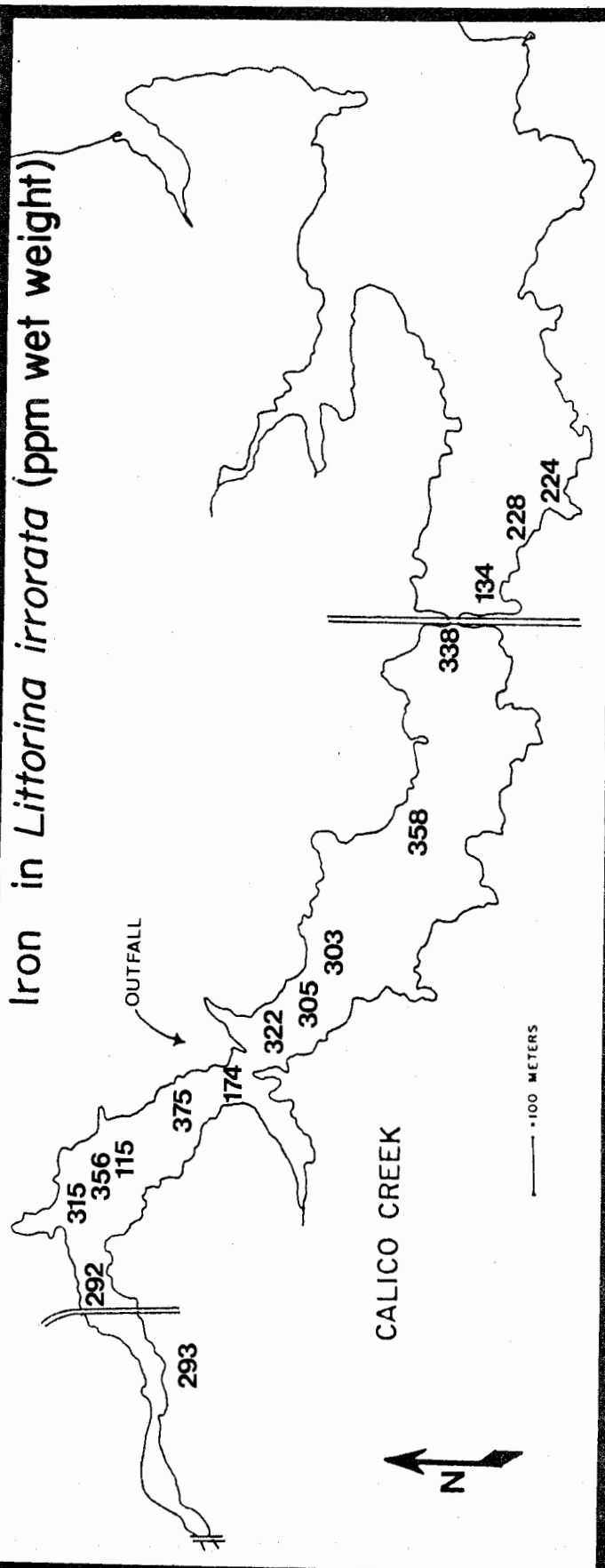


Fig. 19. Iron found in Calico Creek during 1975

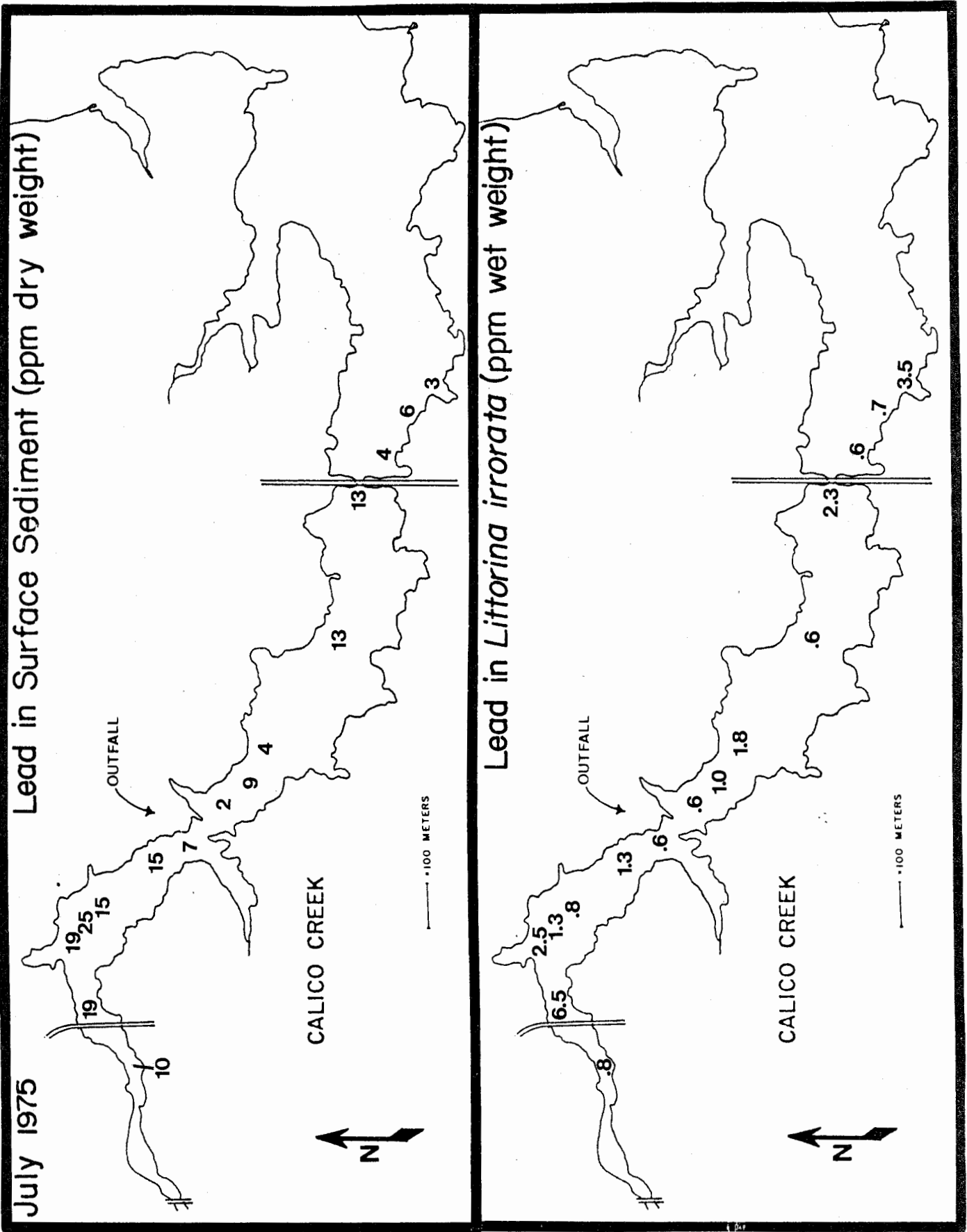
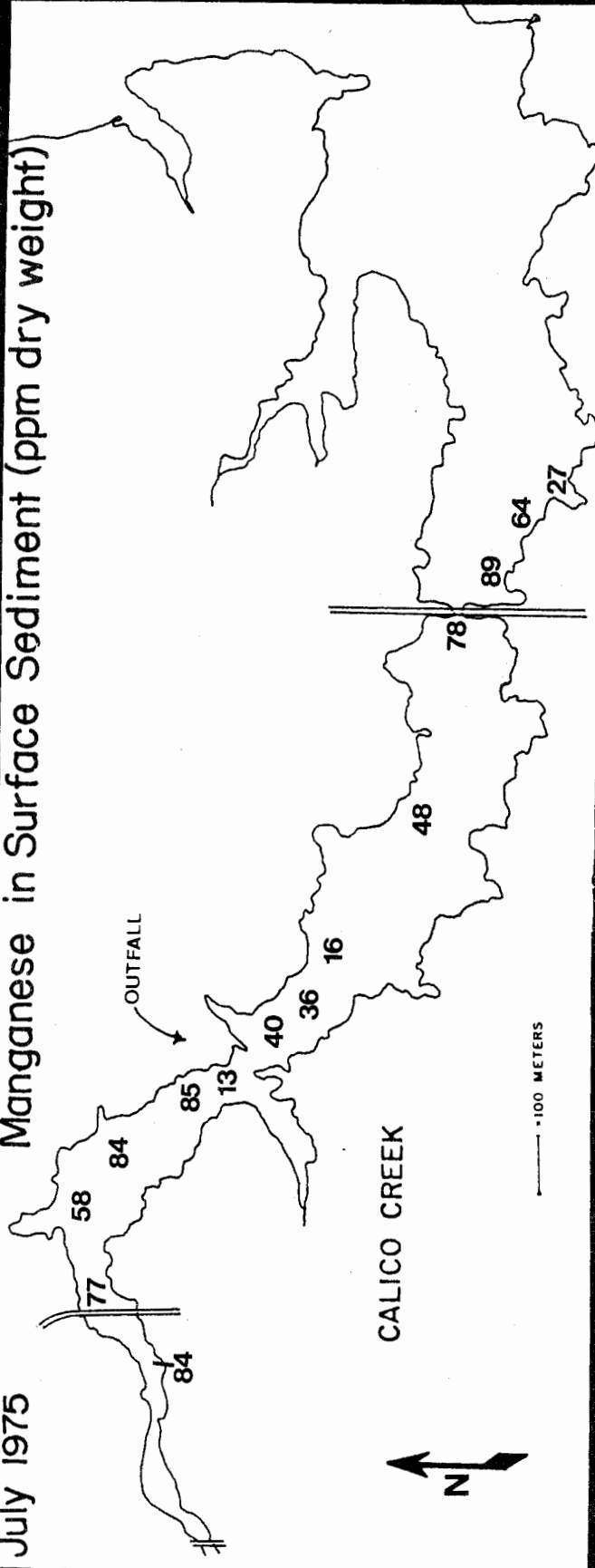


Fig. 20. Lead found in Calico Creek during 1975

Manganese in Surface Sediment (ppm dry weight)

July 1975



Manganese in *Littorina irrorata* (ppm wet weight)

July 1975

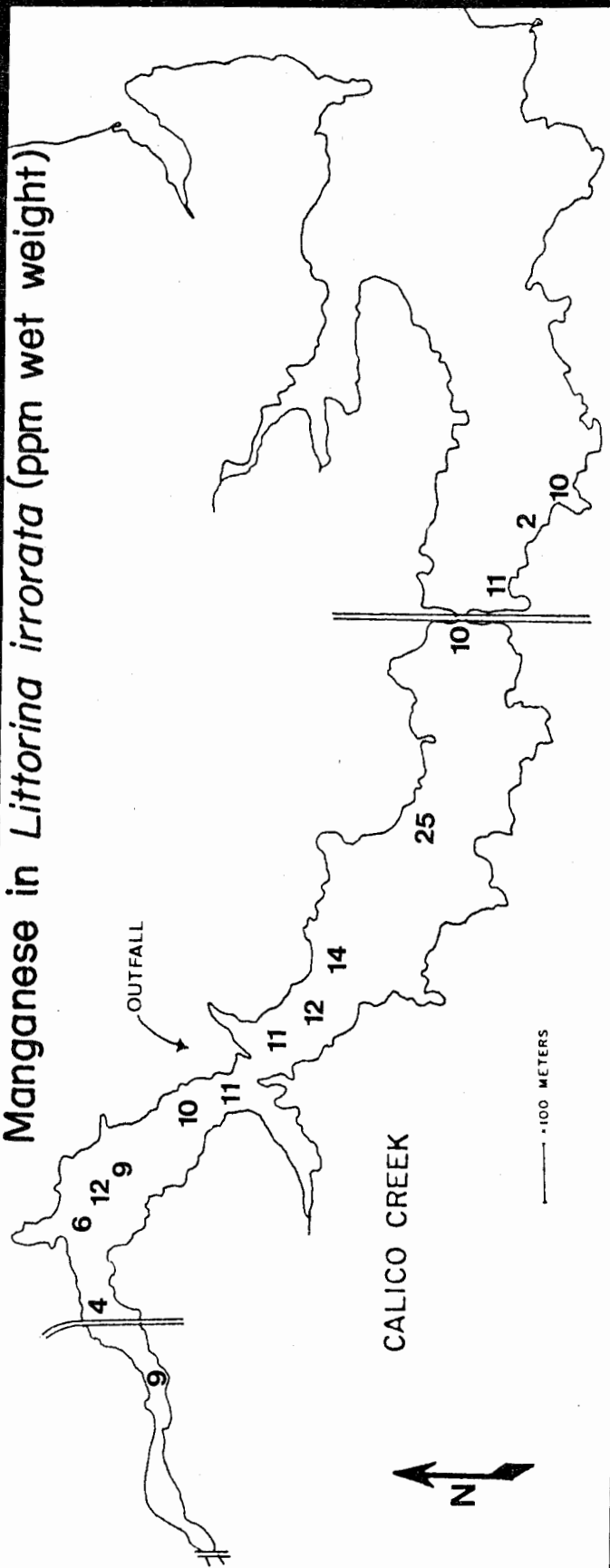


Fig. 21. Manganese found in Calico Creek during 1975

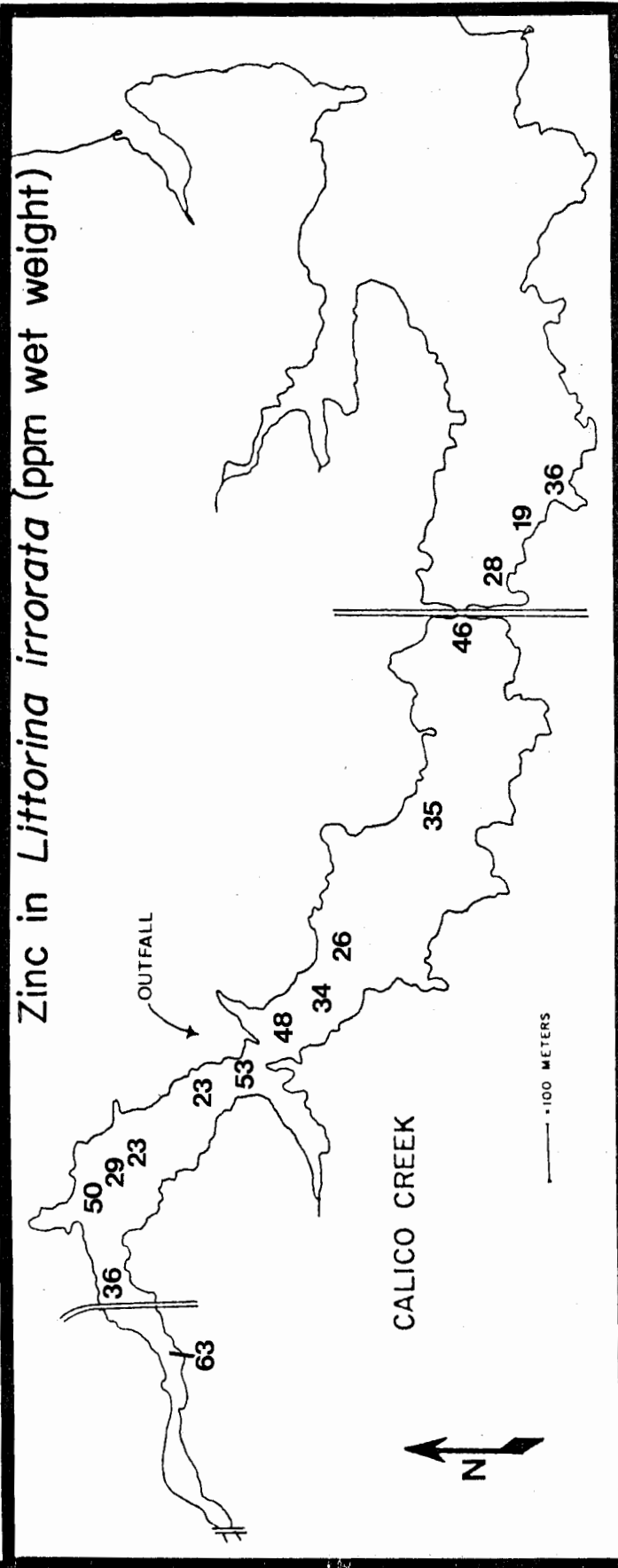
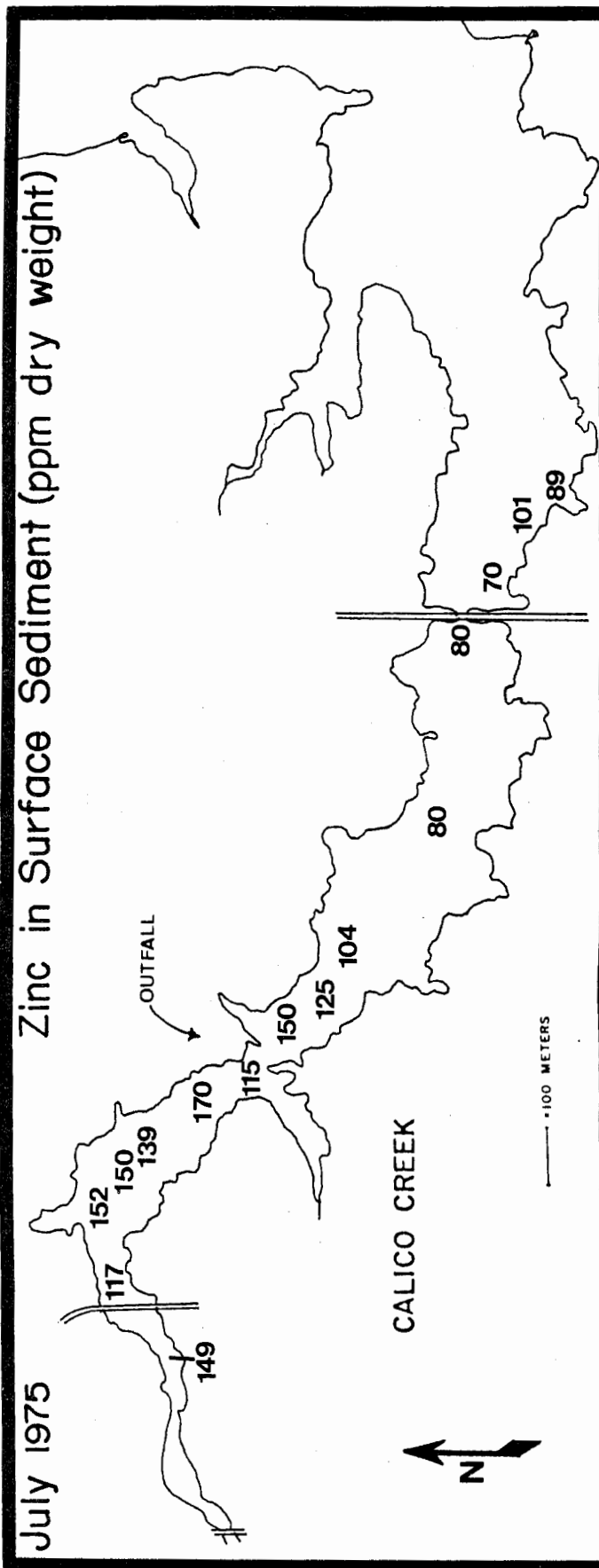


Fig. 22. Zinc found in Calico Creek during 1975

Table 17. Heavy metal concentrations (ppm dry weight) and percent loss on ignition values (an estimate of the organic matter) in the top 3 cm of surface sediment from individual stations above and below the outfall in Calico Creek - January 1976

Meters Above Outfall	%Loss on Ignition	Hg	Cd	Cr	Cu	Fe x 1000	Pb	Mn	Zn
10	17	.37 ± .07	.3	8	25	6.0	20.0	30	106
70	17	.58 ± .09	.2	4	20	3.4	7.0	11	96
120	18	.57 ± .02	.4	6	22	4.2	14.0	13	102
300	19	.49 ± .02	.3	4	18	2.1	x	12	92
350	20	.36 ± .09	.4	7	27	5.7	28.0	23	122
425	18	.54 ± .05	.4	10	15	4.3	77.5	34	88
475	18	.40 ± .08	.4	5	9	3.6	7.5	13	78
525	16	.39 ± .06	.5	7	7	3.1	2.4	16	94
540	8	.15 ± .01	.5	7	13	4.6	38.8	25	65
590	6	.06 ± .02	.2	1	6	3.8	41.7	10	29
605	16	.36 ± .01	.3	7	16	4.3	88.2	20	93
Mean	16	.39	.4	6	16	4.1	32	19	88
St.Dev.	4	.16	.1	2	7	1.1	30	8	24
Co.Var.	29	42	29	40	44	27	91	44	28
r(Distance)	-.489	-.605	.295	-.082	-.741	-.219	.469	-.037	-.561
r(%Loss vs metal)	x	.846	.100	.425	.579	.004	-.160	.123	.873
Meters Below Outfall									
20	17	.35 ± .08	.3	6	24	4.9	28.0	16	102
30	18	.65 ± .05	1.0	8	19	5.0	27.0	35	99
200	18	.58 ± .02	.3	7	20	4.7	31.0	25	96
340	18	.21 ± .05	1.2	4	12	3.8	39.0	20	72
600	12	.16 ± .05	.5	3	6	3.2	38.8	17	70
845	12	.17 ± .07	.5	10	14	5.1	56.3	21	95
900	12	.15 ± .06	1.0	5	6	4.1	3.2	49	69
1050	12	.12 ± .02	.3	13	11	5.0	57.6	59	74
1100	14	.10 ± .03	.3	4	7	3.2	7.5	13	75
1200	11	.15 ± .02	.4	5	13	4.2	2.3	24	103
1320	12	.13 ± .02	.3	5	7	3.4	4.7	10	63
1400	8	.08 ± .005	.2	7	8	4.4	.1	54	53
Mean	14	.24	.5	6	12	4.3	25	29	81
St.Dev.	3	.19	.3	3	6	.7	21	17	17
Co.Var.	24	80	65	44	49	17	84	57	21
r(Distance)	-.885	-.817	-.427	.034	-.757	-.359	-.427	.216	-.581
r(%Loss vs metal)	x	.777	.434	-.090	.683	.195	.318	-.370	.528

Table 18. Heavy metals concentrations in the snail *Littorina irrorata* (ppm wet weight) at individual stations above and below the outfall in Calico Creek - January 1976

Meters above Outfall	Av Wt g	Hg	Cd	Cr	Cu	Fe	Pb	Mn	Zn
300	.8	.32 ± .15	.2	.5	120	336	1.0	8	30
350	1.1	.38 ± .09	.2	.3	157	164	1.0	4	25
425	.9	.17 ± .02	.2	.5	102	115	2.6	4	26
540	1.0	.21 ± .07	.2	.2	137	152	1.5	6	19
605	.9	.12 ± .06	.3	1.1	92	224	1.8	5	22
Mean	.9	.24	.22	.5	122	198	1.6	5	24
St.Dev.	.1	.11	.04	.3	26	86	.7	2	4
Co.Var.	12	45	20	67	22	44	42	31	17
r(distance)		-.830	.706	.486	-.449	-.348	.432	-.261	-.847
r(sed vs <i>Littorina</i> )		.063	-.535	.000	.629	-.807	.788	-.779	.459
Meters below Outfall									
30	.7	.61 ± .07	.3	.4	256	198	2.4	10	34
340	.8	.39 ± .03	.2	.4	90	201	1.0	10	27
600	.6	.57 ± .08	.2	.5	151	178	3.9	5	18
900	.8	.09 ± .03	.2	.4	69	229	3.0	5	19
1050	.8	.10 ± .04	.1	.4	67	152	1.9	11	19
1100	.7	.12 ± .02	.1	.7	65	286	2.7	10	24
1200	.4	.16 ± .08	.2	.4	328	173	1.0	7	26
1320	.7	.17 ± .03	.3	.4	139	132	2.5	9	21
1400	.6	.13 ± .08	.2	.5	43	183	3.4	8	23
Mean	.7	.26	.2	.5	134	192	2.4	8	23
St.Dev.	.1	.21	.07	.1	98	45	1	2	5
Co.Var.	19	80	35	22	73	23	41	27	22
r(distance)		-.847	-.304	.388	-.262	-.171	.120	-.112	-.559
r(sed vs <i>Littorina</i> )		.718	.326	-.324	.620	-.270	-.106	.022	.632

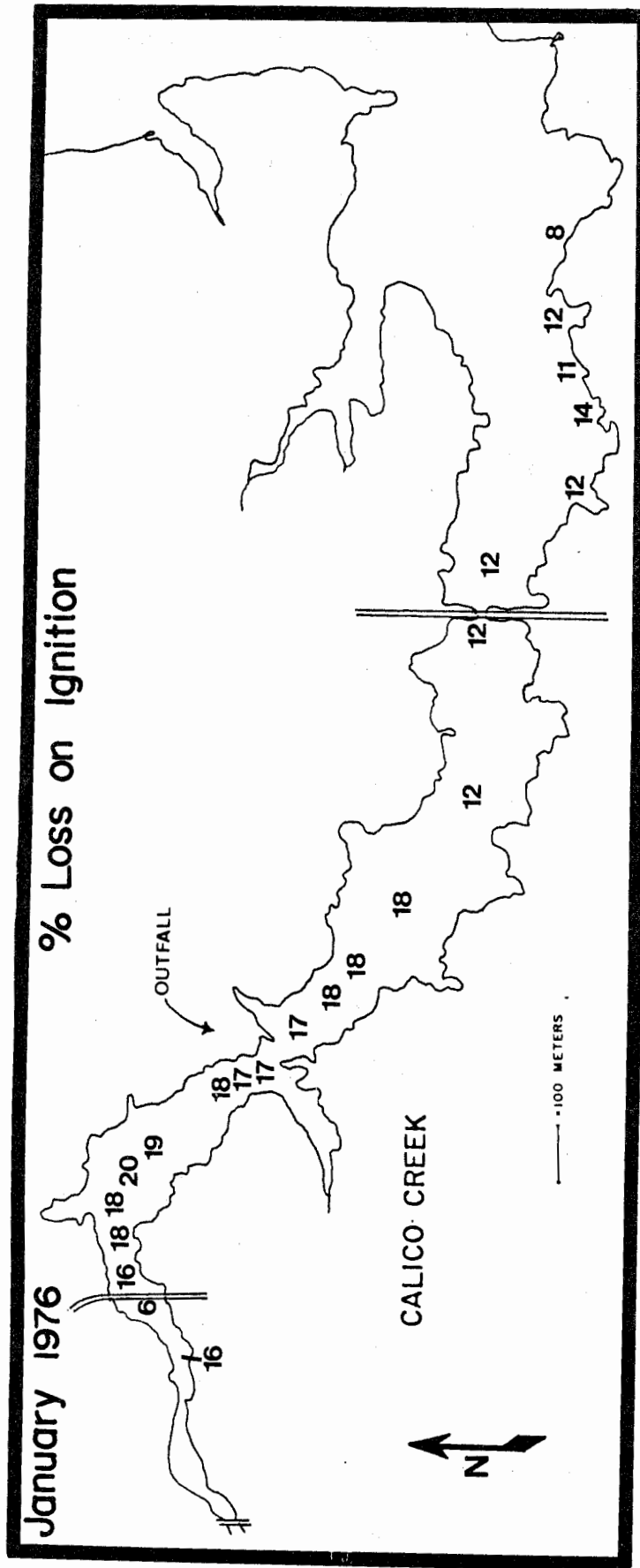


Fig. 23. Percent loss on ignition in Calico Creek for 1976

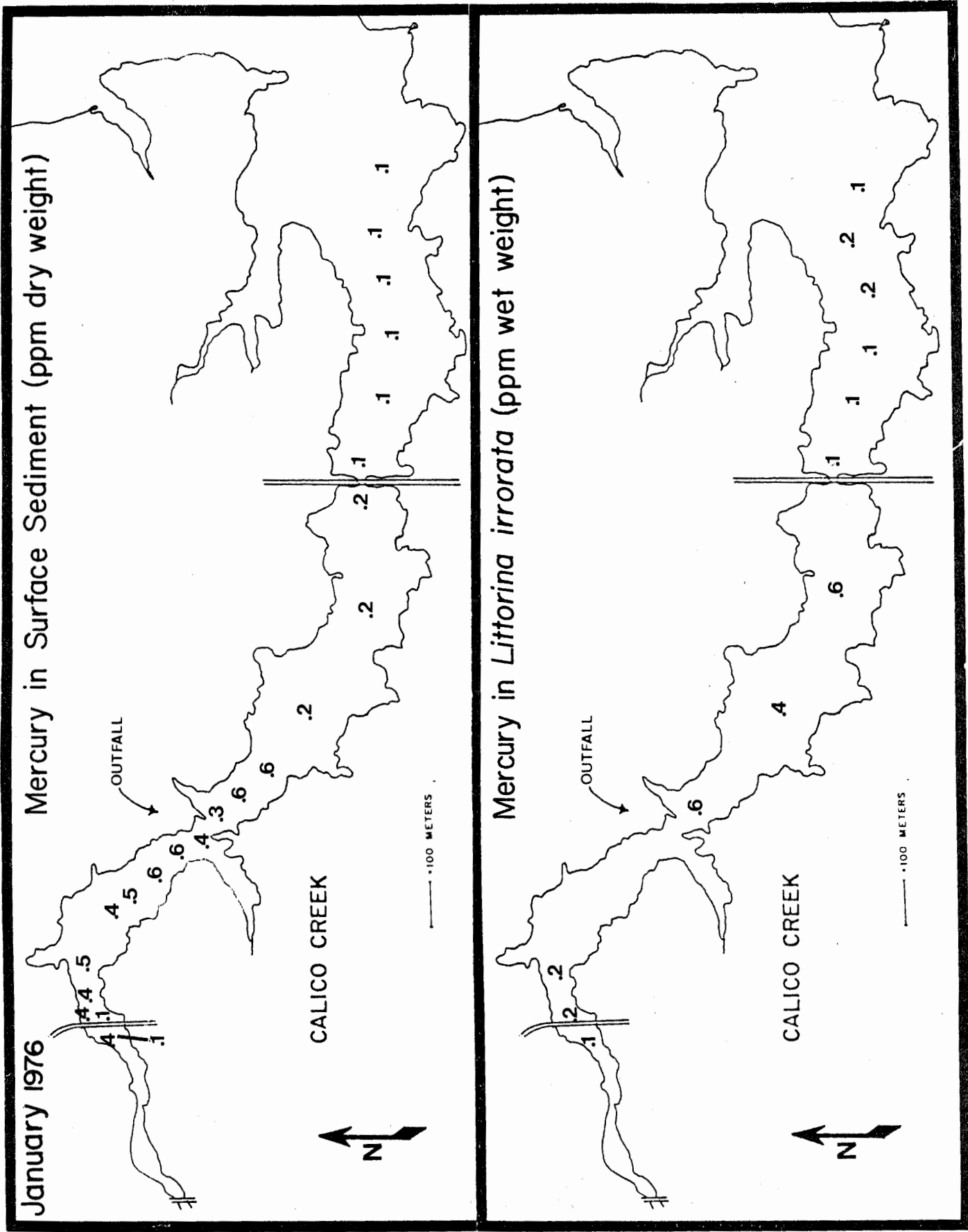


Fig. 24. Total mercury found in Calico Creek during 1976



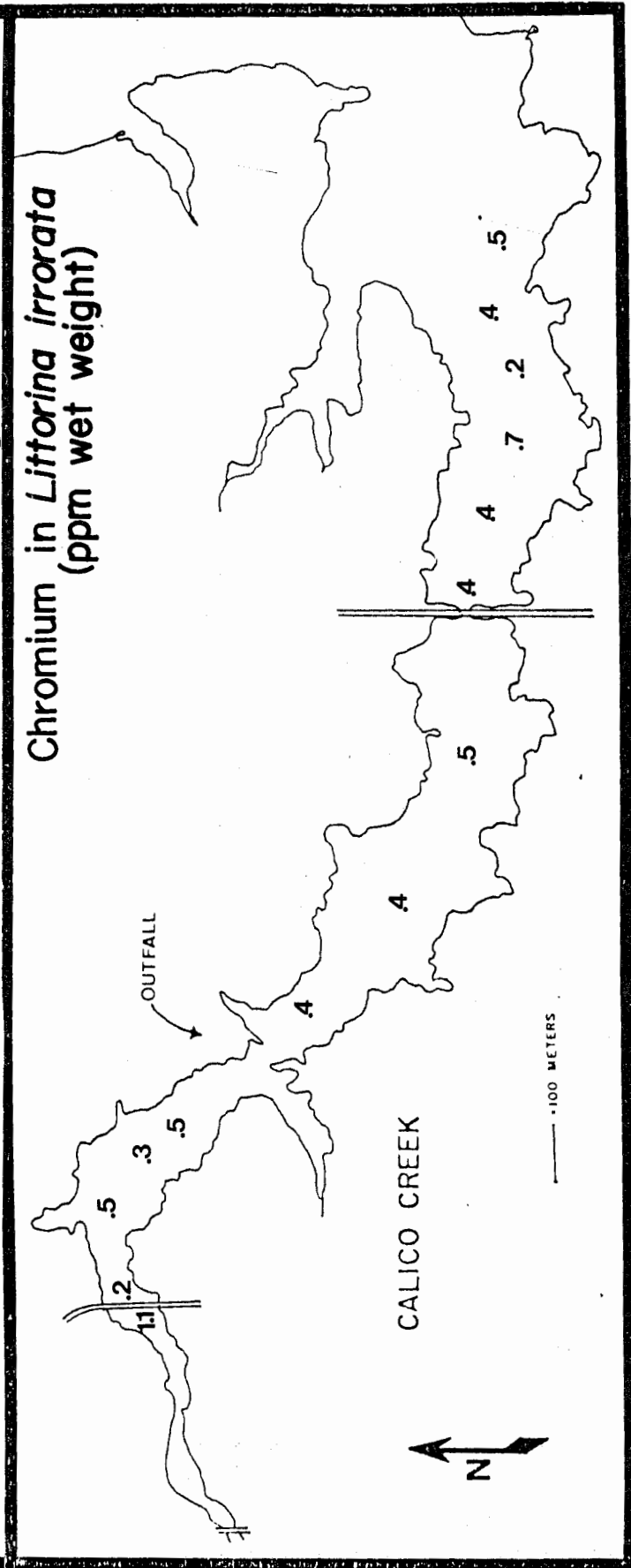
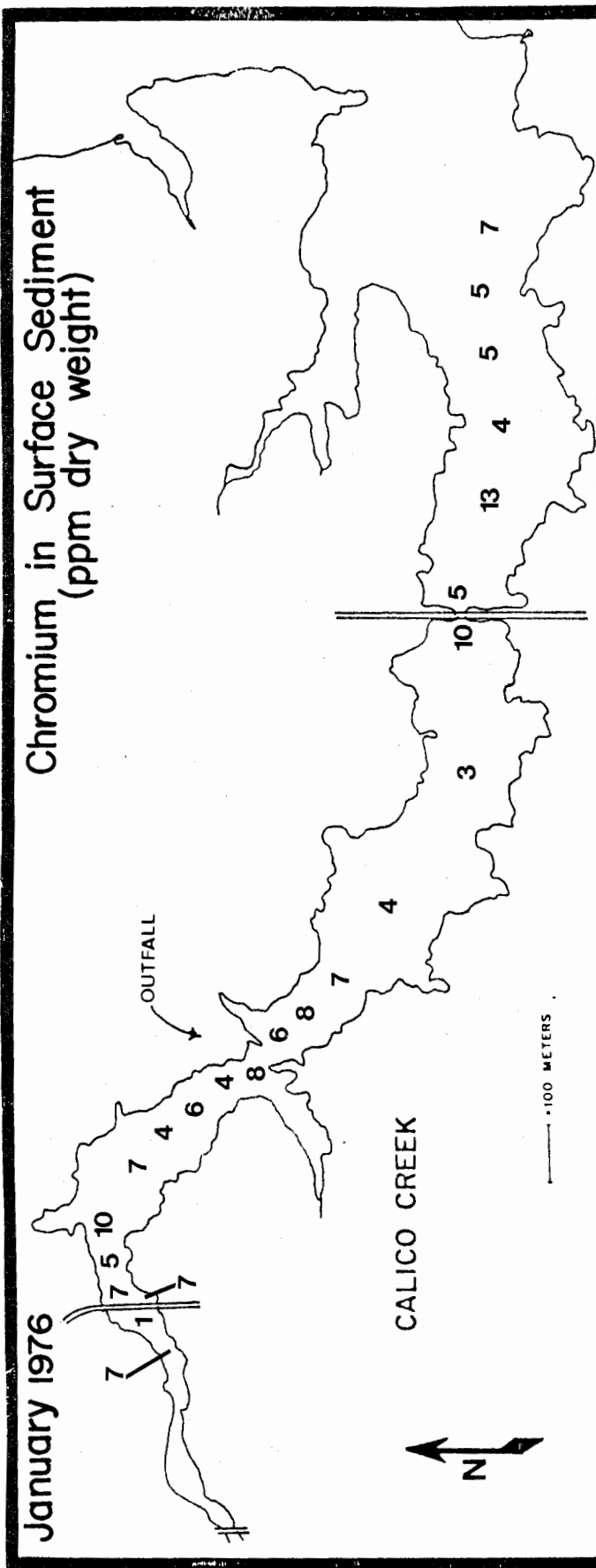


Fig. 26. Chromium found in Calico Creek during 1976

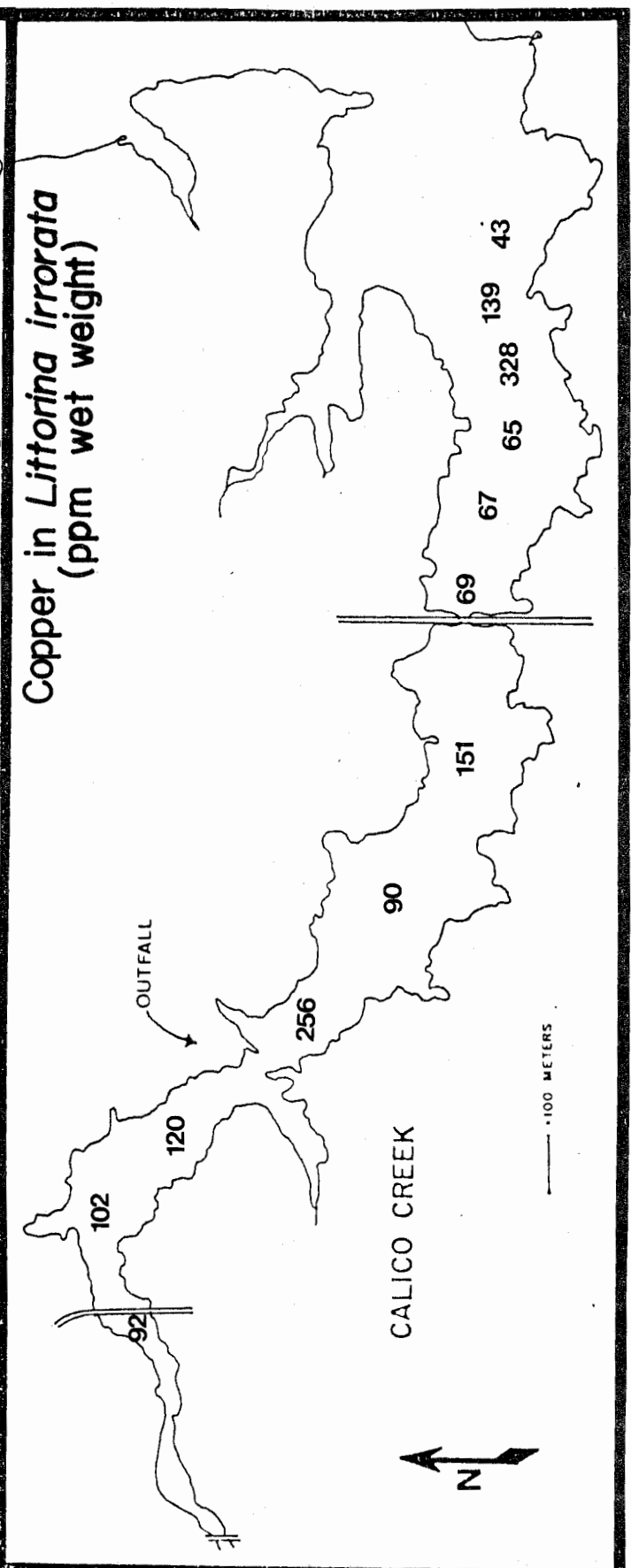
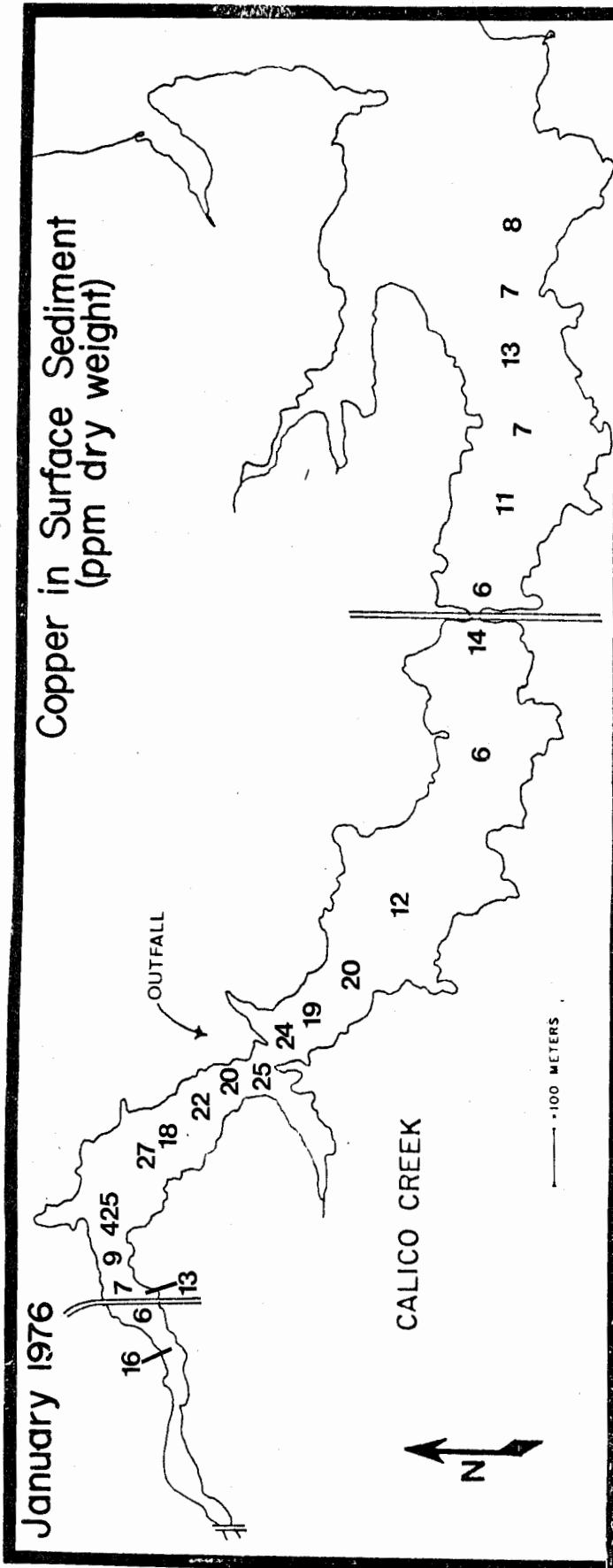


Fig. 27. Copper found in Calico Creek during 1976

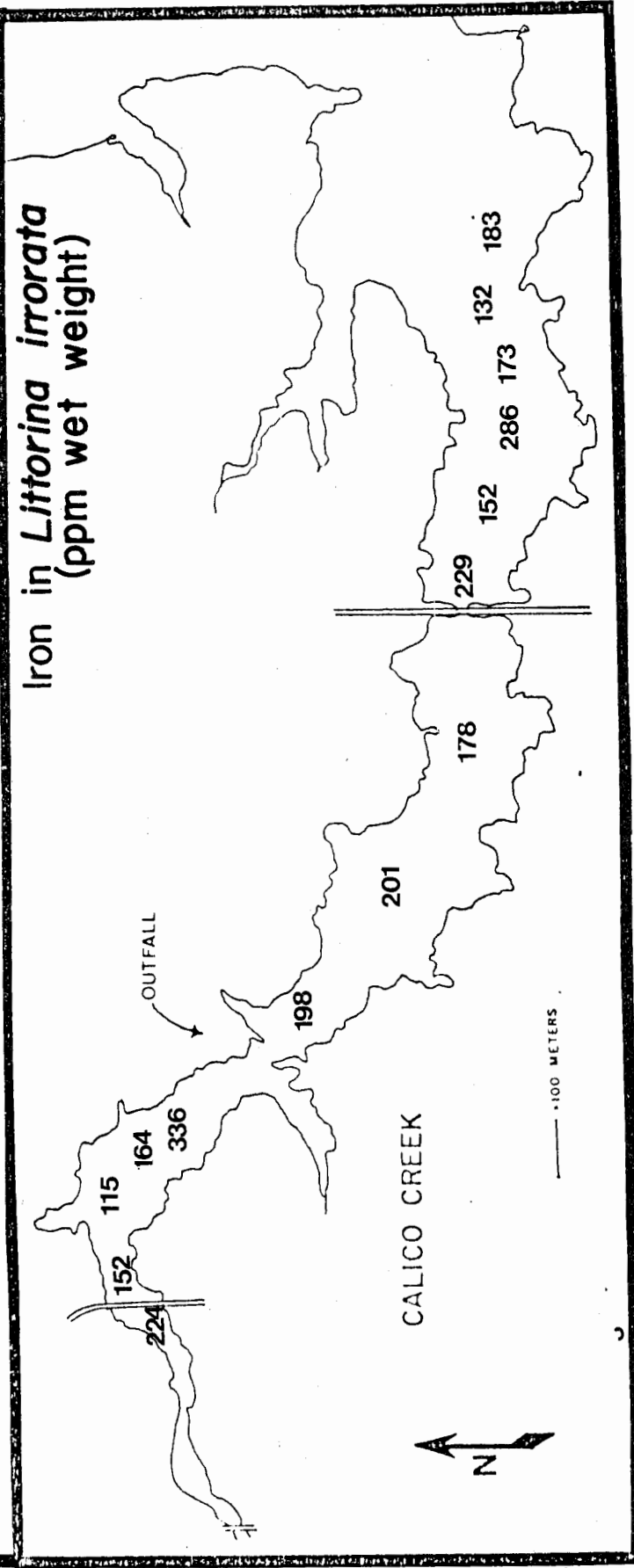
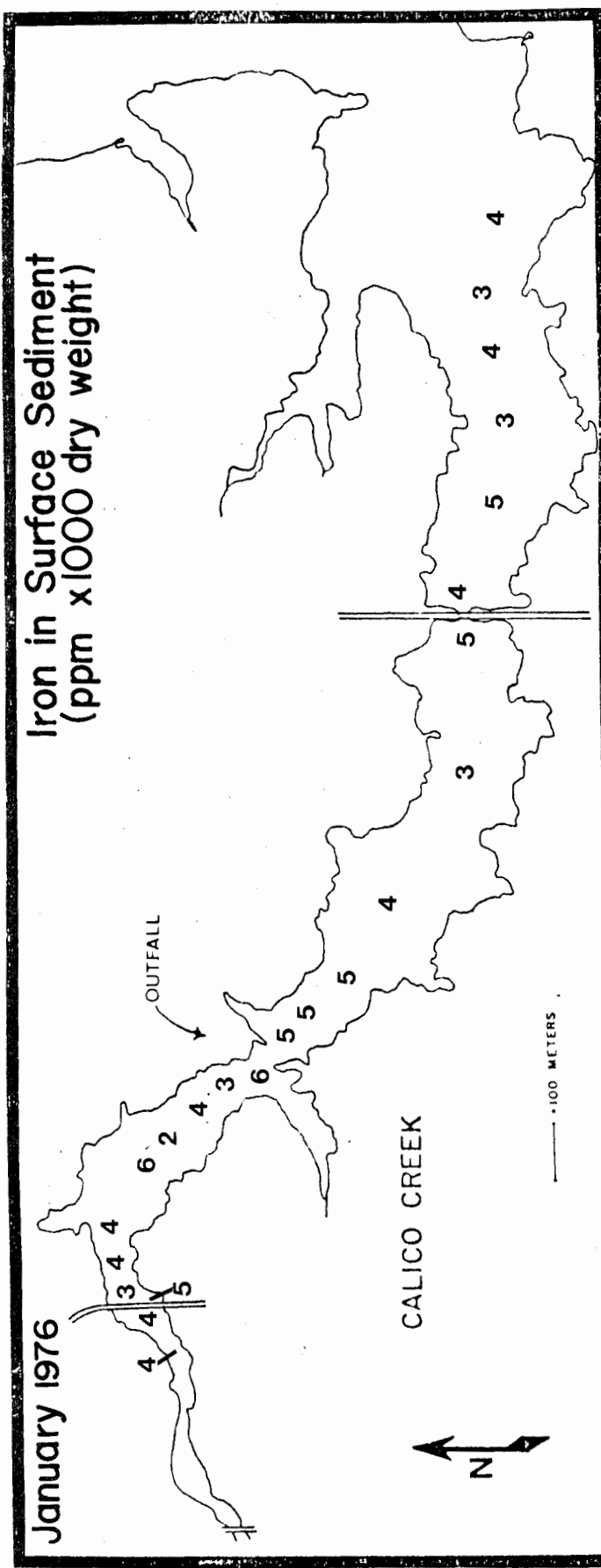


Fig. 28. Iron found in Calico Creek during 1976

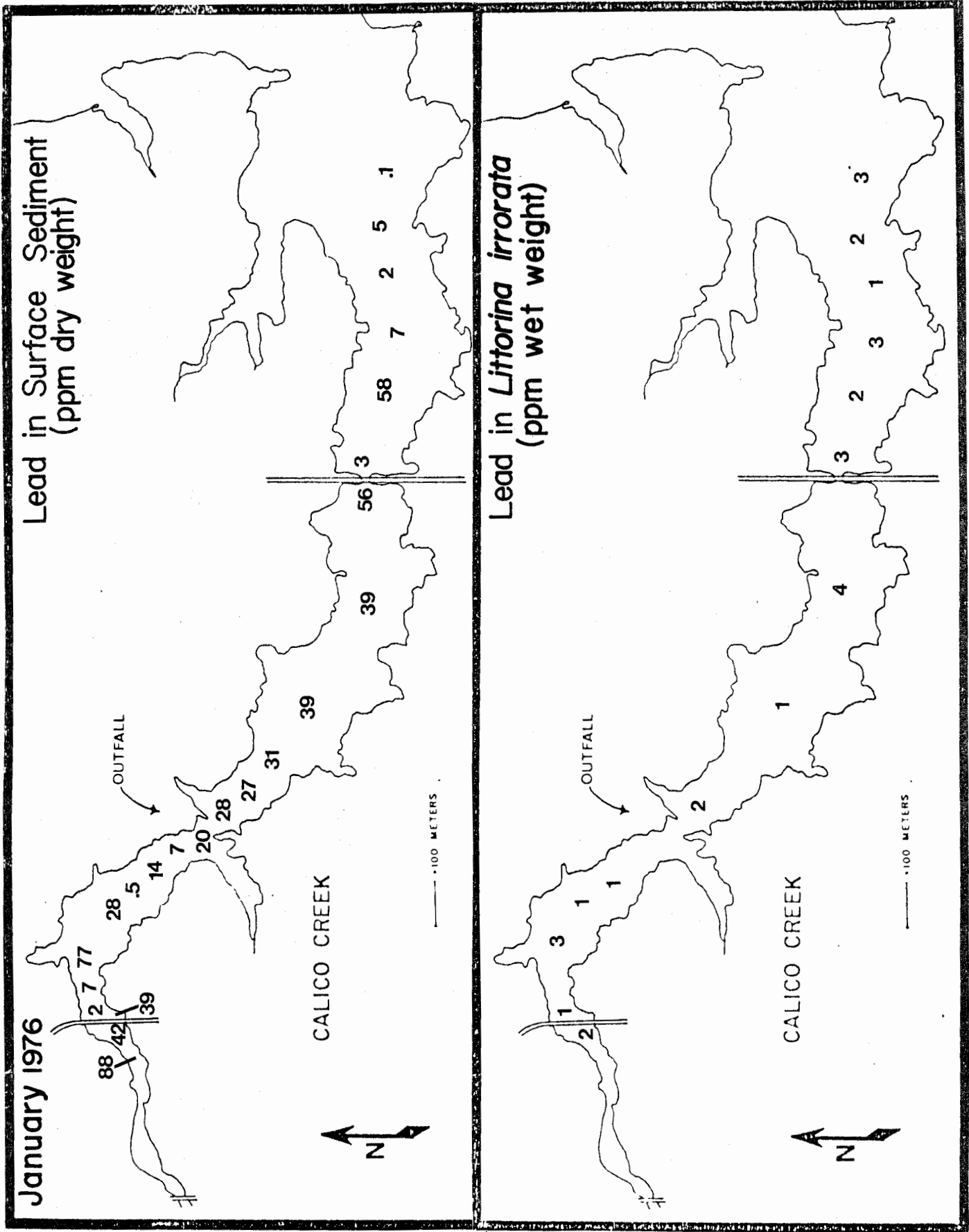


Fig. 29. Lead found in Calico Creek during 1976

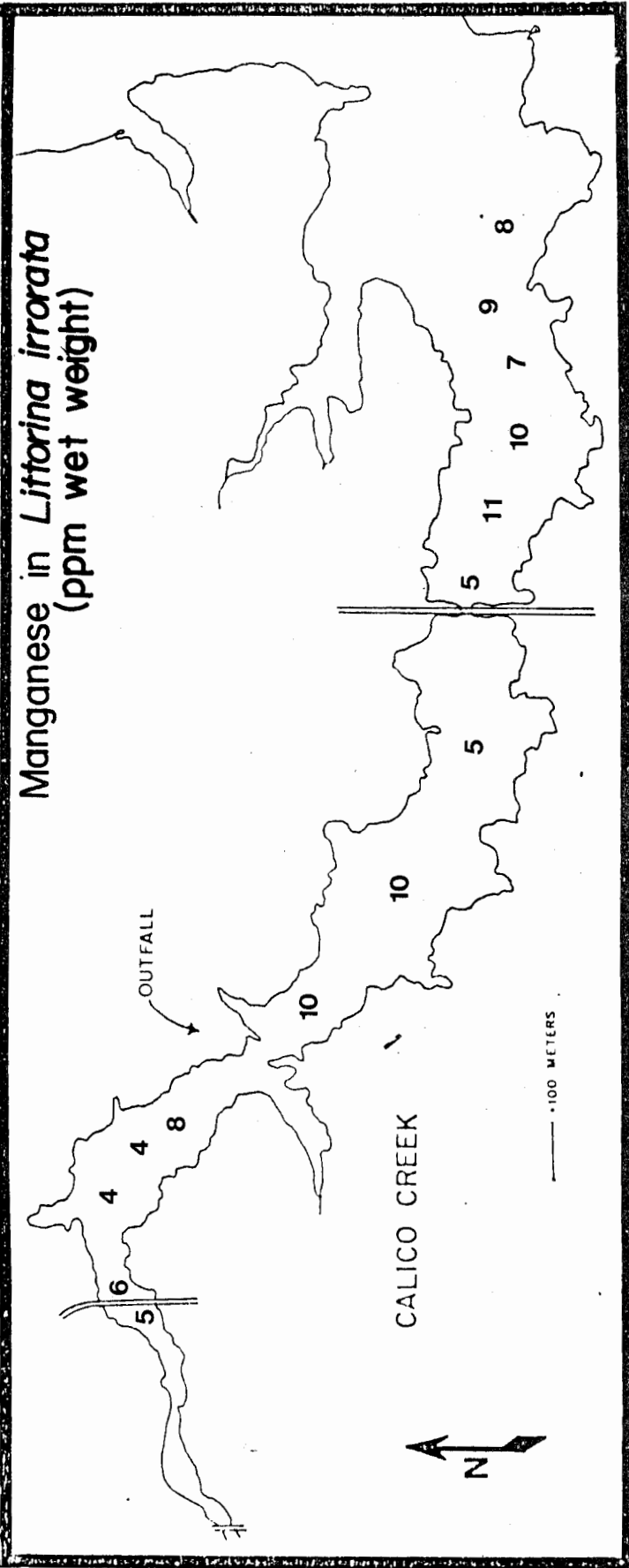
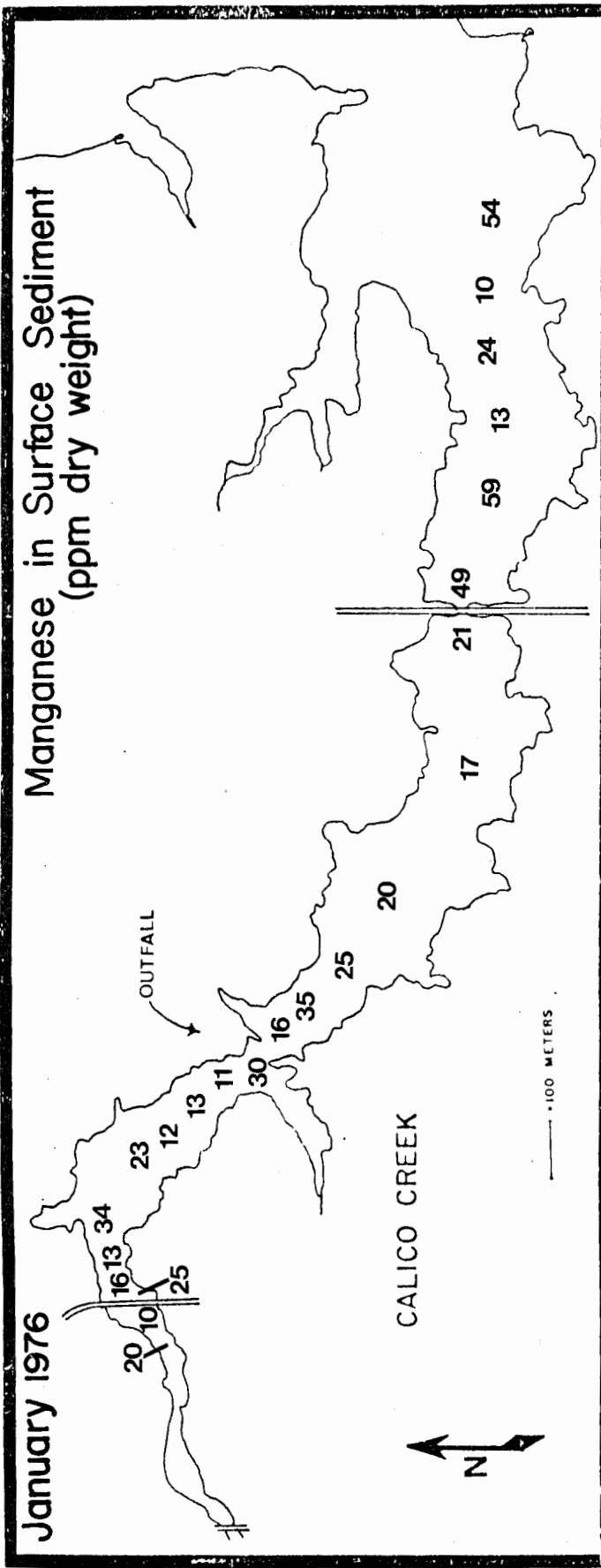


Fig. 30. Manganese found in Calico Creek during 1976

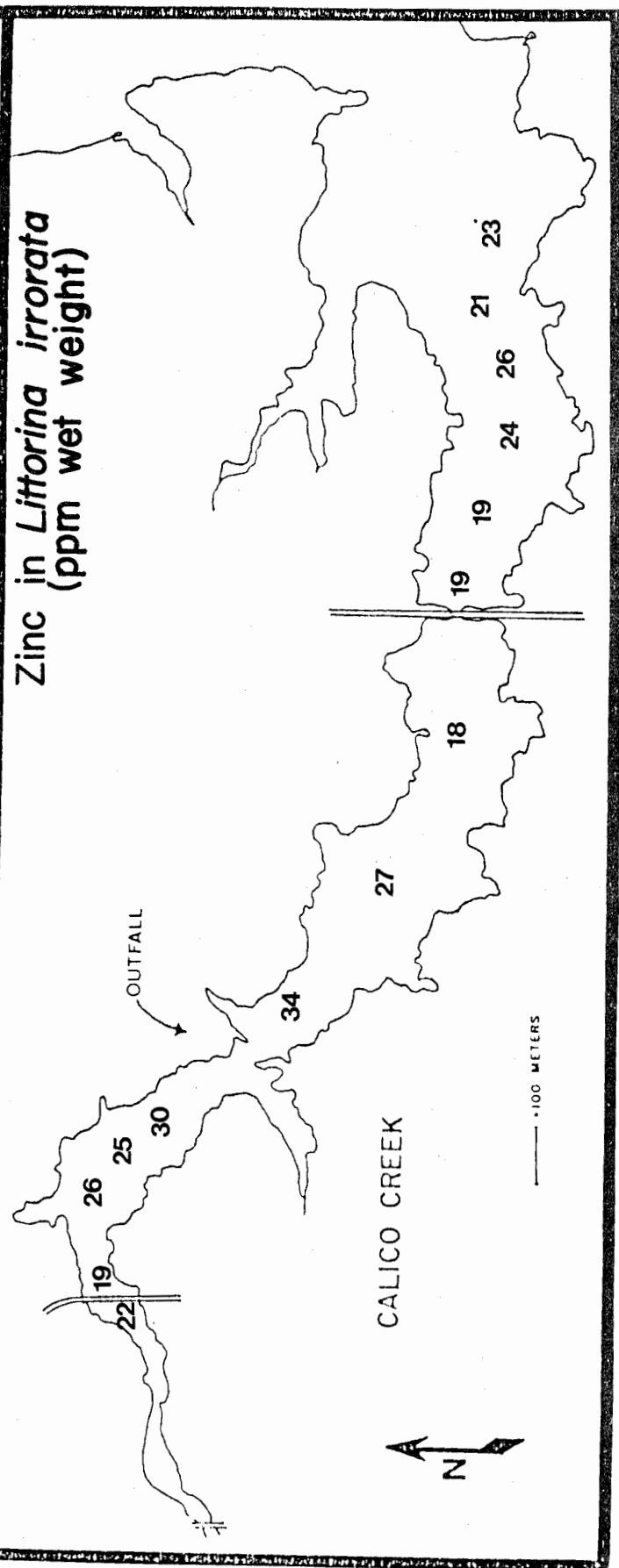
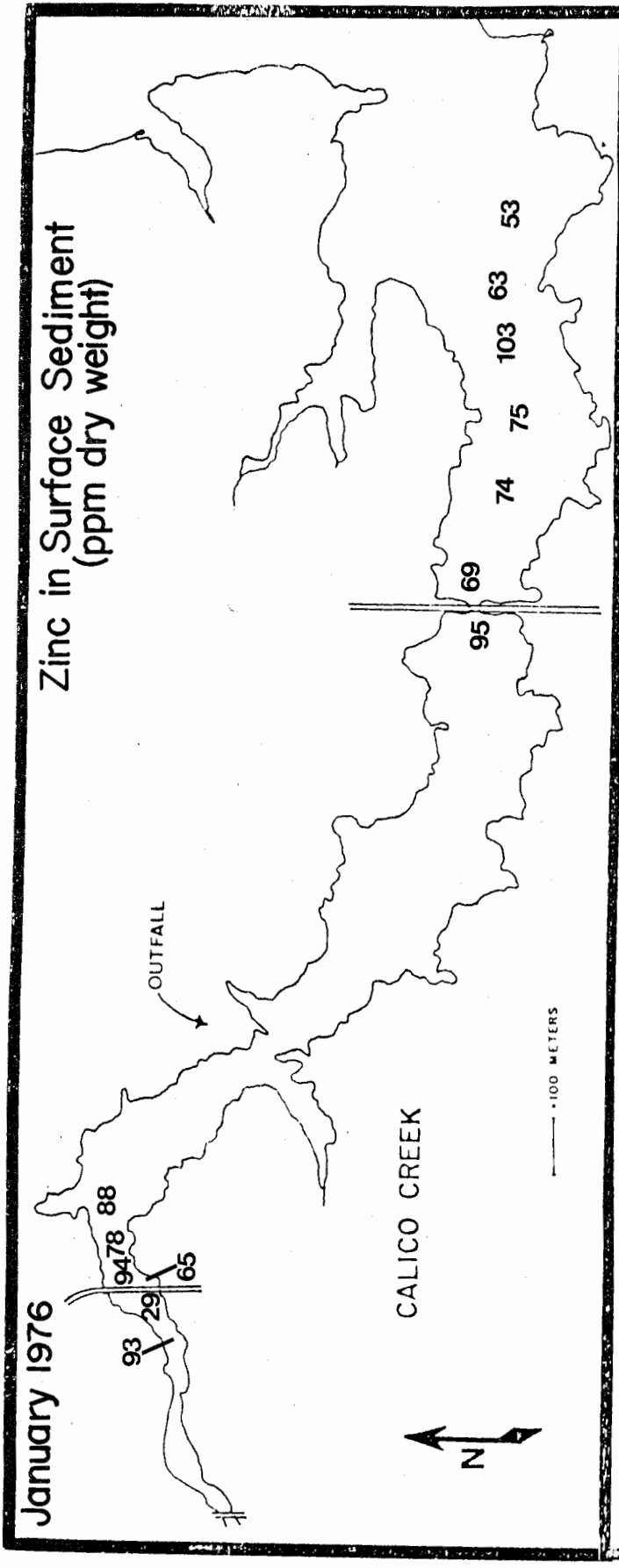


Fig. 31. Zinc found in Calico Creek during 1976

outfall in Calico Creek were similar to those above the outfall in the 1974 surface sediment sampling (Table 12). All metal concentrations decreased with distance from the outfall. The percent loss on ignition decreased significantly below the outfall. From Figure 7 it can be seen that the outfall had little or no influence upon the organic content of the sediment. The closer to the mouth of the creek, the lower the concentration of organic matter in the sediments. In contrast to organic content, the concentrations in sediments for all the metals analyzed fell off more or less symmetrically above and below the outfall with increasing distance, reflecting the strong tidal influence on point source discharge.

In 1975 the concentration of mercury in the surface sediments (Table 14 and Figure 14) had decreased to 0.3 ppm at 10 m above the outfall. The mean concentration in 1975 for the Calico Creek sediments above the outfall was  $0.42 \pm 0.09$  compared to  $0.89 \pm 0.40$  in 1972 and  $0.46 \pm 0.24$  in 1974. The only other metal concentration that was lower in 1975 was lead, at  $16 \pm 6$  ppm, compared to  $42 \pm 17$  ppm in 1974. In 1975 the previously strong negative correlation of mercury concentration with distance above the outfall had disappeared. Chromium, copper, iron, lead, manganese, and zinc all increased slightly with distance above the outfall, exactly reversing the trend of the previous year when concentration decreased slightly with distance above the outfall. Cadmium was the only metal that continued its 1974 pattern.

Lead was the only metal in which the mean sediment concentration below the outfall varied greatly from 1974 ( $24 \pm 12$  ppm) to 1975 ( $7 \pm 4$  ppm). The mean concentration for all other metals (Table 14) was within the standard deviations for the two years. The highest concentrations of mercury ( $0.35 \pm 0.06$  ppm) and zinc (150 ppm) were at 30 m below the outfall; all other metals had maximum concentrations at a distance of 900 m below. Mercury, cadmium, copper and zinc decreased with distance below the outfall in 1975 as did the samples taken in 1974. Chromium, lead and manganese increased slightly with distance below the outfall in contrast to the data from 1974. These differences are similar to those found above the outfall in 1975. The percent loss on ignition for both years was the same.

In 1975 mercury and manganese concentrations in L. irrorata decreased with distance above the outfall (Table 15 and Figures 15 through 22), while cadmium, chromium, copper, iron, lead and zinc all increased slightly with distance above the outfall. Table 15 emphasizes that the metal content of Littorina is relatively independent of the sediment concentration of metals. The concentrations of mercury, cadmium, iron, manganese and zinc in L. irrorata decreased with distance below the outfall. The remaining metals, chromium, copper and lead, increased slightly with distance. All metal concentrations in C. virginica (Table 16) decreased with distance from the outfall.

The mean concentrations in January 1976 surface sediments (Table 17 and Figures 24 through 31) decreased for cadmium, manganese and zinc from 1975 means, while the remaining metals did not differ. Mercury was the only metal found to be highest near the outfall ( $0.58 \pm 0.09$  ppm at 70 m). Mercury, chromium, copper, iron, manganese and zinc decreased with distance above the outfall while cadmium and lead increased. Below the outfall mercury, cadmium,

copper, iron, lead, and zinc decreased with distance, while chromium and manganese increased. Mercury, copper and zinc above the outfall, and mercury and copper below the outfall were positively correlated with the organic content of the sediments. Below the outfall zinc was negatively correlated with organic content.

The marsh grass, *Spartina alterniflora*, was analyzed above and below the outfall (Figure 32). As shown by the data in Table 19, mercury and zinc were the only metals in the stems and leaves of *S. alterniflora* that significantly decreased with distance from the outfall. In *Spartina* roots cadmium, chromium, copper, iron, lead, manganese and zinc all significantly decreased with distance above the outfall. Mercury in roots, however, increased significantly with distance from the outfall in the stations above the outfall. This may be related to the increase in organic content with distance from the outfall. Metals in the roots of *S. alterniflora* correlated with the increase in sediment metal concentrations in stations above the outfall (Table 19). Mercury, chromium, iron, lead and zinc decreased with distance below the outfall (Table 19) on Calico Creek in the roots of *S. alterniflora* indicating that the plants were taking up these metals near the outfall.

To determine the precision of the metal determination used in this study, eight samples 0.5 m apart at a single station were taken in 1975 (Table 20) and 1976 (Table 21). The coefficient of variance ranged from 1% for cadmium to 34% for lead in 1975. The coefficient of variance was higher in 1976 subsamples with 14% for mercury and 66% for chromium. The mean concentration of mercury, cadmium, manganese and zinc did not change significantly from 1975 to 1976. Chromium, copper, iron, and lead showed a significant decrease. A station 10 m above the outfall was also sampled six times 0.5 m apart in 1976 (Table 22). The coefficient of variance (19%-71%) was very high at this station for all metals; if we assume that the data in Tables 20 and 21 establish the precision of the analyses, then the variance in Table 22 can be assigned to small scale spatial heterogeneity associated with the proximity of the outfall.

Sediments were also analyzed using a preferential leaching technique with hydroxylamine hydrochloride and acetic acid to determine the concentration of metals readily available for uptake by organisms. There is a decrease in leachable metal concentrations with distance above the outfall (Table 23) with the exception of manganese; while below the outfall cadmium, copper, lead and zinc decreased with distance from the outfall. Leachable metals had a higher coefficient of variance in the eight 0.5 m apart samples (Table 24) than did the total metals.

Mercury was the only metal that was consistently and significantly elevated near the Morehead City sewage treatment plant outfall on Calico Creek. Ranges of total mercury near the outfall were 1.2 ppm dry weight to 0.1 ppm dry weight for the stations 1000 m away from the outfall. All other metal concentrations varied randomly in relation to the location of the outfall. With the exception of lead, all the metal concentrations in the sediment have decreased in Calico Creek during the period 1973 to 1976. There remained throughout the study period a correlation between metal and the

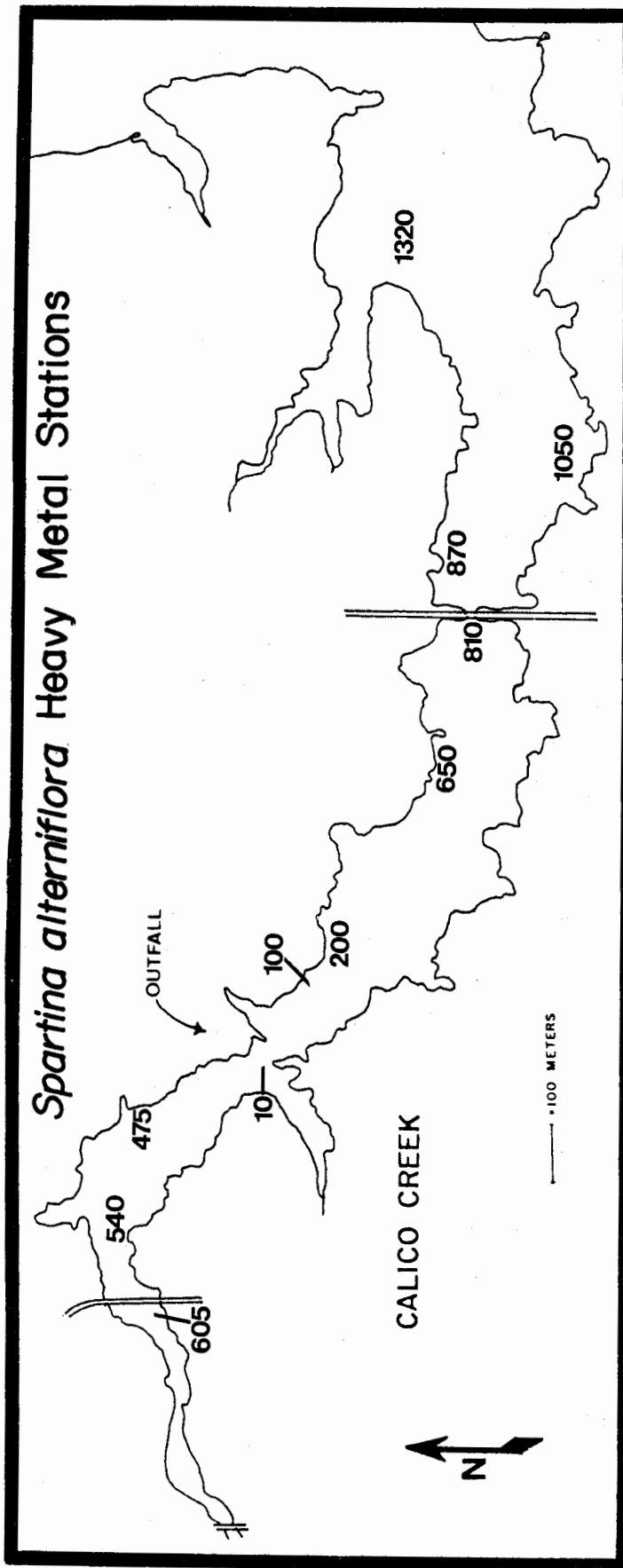


Fig. 32. *Spartina alterniflora* heavy metal stations in Calico Creek (distance from outfall in meters)

Table 19. Heavy metals (ppm dry weight) in the stems, leaves, and roots of the marsh grass *Spartina alterniflora* compared to total and leached metal data for stations above and below the outfall in Calico Creek

Distance Above Outfall (m)	% Loss	Hg	Cd	Cr	Cu	Fe X1000	Pb	Mn	Zn
605 St & Lea		.07± .05	.2	2	6	.8	5	11	24
Root		.22± .06	.4	11	45	.6	16	23	113
Total Sed	21	.39± .09	.8	19	32	5.5	10	84	149
Leach Sed		x	.7	2	6	1.6	39	9	116
540 St & Lea		.10± .04	.1	4	6	1.2	2	16	25
Root		.20± .08	.3	8	19	.6	15	20	63
Total Sed	20	.36± .08	.8	16	36	8.2	19	77	117
Leach Sed		x	.7	2	4	1.4	36	10	101
475 St & Lea		.03± .02	.1	3	10	.9	4	13	24
Root		.19± .01	.4	6	24	6.0	13	21	65
Total Sed	20	.38± .10	1.0	12	31	6.5	19	58	152
Leach Sed		x	.6	1	5	1.5	35	7	97
10 St & Lea		.16± <.005	.1	3	8	.8	3	15	28
Root		.18± .01	.9	>75	55	8.1	20	27	119
Total Sed	18	.31± .02	1.0	6	16	2.9	7	13	115
Leach Sed		x	.6	2	4	1.0	33	7	85
St & Lea mean±StDev		.09± .05	.1±.1	3±.8	8±2	.9±.2	4±1	14±2	25±2
Root mean±StDev		.20± .02	.5±.3	9±2	36±17	3.8±3.8	16±3	23±3	90±30
Total Sed mean±StDev		.36± .04	.9±.1	13±6	29±9	6±2	14±6	58±32	133±20
Leach Sed mean±StDev		x	.7±.1	1.8±.5	5±1	1.4±.3	36±3	8±2	100±13
r(St & Lea vs Total Sed)		-.906	-.577	-.218	-.328	.828	-.313	-.340	-.787
r(Root vs Total Sed)		.768	.640	-.048	.809	.694	.879	.799	.109
r(St & Lea vs Leach Sed)		x	.577	.000	-.091	.151	.579	.125	-.795
r(Root vs Leach Sed)		x	-.640	.816	.138	-.003	-.408	-.556	-.016
r(St & Lea vs Distance)		-.777	.487	-.098	-.338	.389	.315	-.441	-.950
r(Root vs Distance)		.811	-.966	.187	-.638	-.845	-.806	-.846	-.501

Table 19. (continued)

Distance Below Outfall (m)		% Loss	Hg	Cd	Cr	Cu	Fe X1000	Pb	Mn	Zn
100	St & Lea		.08± .02	.1	3	7	1.2	2	x	20
	Root		.25± .03	.6	9	25	7.5	14	24	110
	Total Sed	19	.50± .03	1.4	14	40	20.0	30	55	291
	Leach Sed		x	.6	6	13	2.8	42	19	106
200	St & Lea		.15± .04	.1	3	8	1.6	5	22	25
	Root		.23± .03	x	x	x	x	x	x	x
	Total Sed	17	.25± .01	1.3	7	14	5.9	4	16	104
	Leach Sed		x	.4	1	3	1.1	21	13	56
650	St & Lea		.07± .03	.1	2	7	.8	3	12	21
	Root		.18± .08	.5	30	29	8.3	15	32	95
	Total Sed	13	.23± .02	.6	12	19	11.5	13	48	80
	Leach Sed		x	.4	2	3	1.1	19	10	50
810	St & Lea		.18±<.005	.1	3	10	1.2	3	14	22
	Root		.08± .02	.5	10	24	5.8	17	21	90
	Total Sed	29	.18± .01	.6	7	16	5.0	13	78	80
	Leach Sed		x	.4	1	4	1.5	21	24	58
870	St & Lea		.15± .04	.1	3	5	1.5	5	18	18
	Root		.07± .01	.3	14	19	5.3	13	28	65
	Total Sed	13	.17± .03	.6	13	16	x	4	89	70
	Leach Sed		x	.3	1	2	1.2	20	10	50
1050	St & Lea		.15± .03	<.05	2	5	.8	2	14	18
	Root		.17± .01	.9	9	24	4.8	13	30	107
	Total Sed	10	.15± .03	.5	5	10	2.7	3	27	49
	Leach Sed		x	.3	1	2	.9	18	11	46
1320	St & Lea		.01±<.005	.2	2	3	.4	2	15	19
	Root		.01±<.005	.4	6	25	2.1	5	16	63
	Total Sed	8	.10± .01	.3	18	9	9.8	11	83	49
	Leach Sed		x	.2	1	1	1.1	10	17	23
St & Lea mean±StDev			.11± .06	.1±.04	2.5±.5	6±2	1.1±.4	3±1	16±4	20±3
Root mean±StDev			.14± .09	.5±.2	13±9	24±3	5.6±2.1	13±4	25±6	88±20
Total Sed mean±StDev			.23± .13	.8±.4	11±5	18±10	9.2±6.2	11±9	57±28	103±85
Leach Sed mean±StDev			x	.4±.1	2±2	4±4	1.4±.6	22±10	15±5	56±25
r(St & Lea vs Total Sed)			-.070	-.290	-.162	.312	-.014	-.516	-.276	.128
r(Root vs Total Sed)			.758	.189	.021	.151	.438	.162	-.608	.532
r(St & Lea vs Leach Sed)			x	-.401	.263	.337	.221	-.121	-.150	.171
r(Root vs Leach Sed)			x	.190	-.034	.150	.442	.479	-.738	.661
r(St & Lea vs Distance)			-.211	.369	-.622	-.599	-.668	-.294	-.569	-.610
r(Root vs Distance)			-.855	-.085	-.242	-.194	-.837	-.603	-.291	-.629

Table 20. Heavy metal concentrations (ppm dry weight) in eight surface sediment samples taken 0.5 m apart 800 m below the outfall in Calico Creek in July 1975

	<u>% Loss</u>	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe</u> <u>X1000</u>	<u>Pb</u>	<u>Mn</u>	<u>Zn</u>
A	11.2	.17 ± .05	.7	16	17	9.7	12	93	90
B	10.8	.18 ± .01	.6	19	16	8.9	13	90	78
C	10.3	.11 ± .02	.7	17	16	10.4	14	85	99
D	10.7	.18 ± .01	.5	26	21	10.2	30	40	79
E	10.2	.25 ± .04	.7	14	14	7.2	21	93	100
F	10.5	.14 ± .03	.8	28	17	13.9	28	92	77
G	10.8	.17 ± .01	.6	20	20	10.0	24	86	100
H	10.5	.16 ± .04	.7	33	18	11.0	27	88	110
Mean	11	.17	.7	22	17	10.2	21	83	90
St.Dev.	.3	.04	.1	7	2	1.9	7	18	10
Co.Var.	3	24	14	30	13	19	34	21	11

Table 21. Heavy metal concentrations (ppm dry weight) in eight surface sediment samples taken 0.5 m apart 800 m below the outfall in Calico Creek in January 1976

	<u>% Loss</u>	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe</u> <u>X1000</u>	<u>Pb</u>	<u>Mn</u>	<u>Zn</u>
A	12	.15 ± .06	1.0	5	6	4.1	3.2	49	69
B	11	.13 ± .02	.8	4	6	3.3	2.1	73	62
C	10	.12 ± .05	.4	13	12	4.9	3.3	74	88
D	11	.09 ± .03	.3	6	12	4.7	3.3	66	81
E	11	.13 ± .01	.3	3	5	2.8	2.2	10	54
F	10	.13 ± .02	.3	3	6	3.2	1.3	49	68
G	11	.12 ± .01	.3	4	6	3.7	1.8	16	59
H	10	.14 ± .02	.4	13	12	4.6	1.4	66	72
Mean	11	.13	.5	6	8	3.9	2.3	50	69
St.Dev.	.6	.02	.3	4	3	.8	.8	25	11
Co.Var.	6	14	56	66	40	20	36	50	16

Table 22. Heavy metal concentrations (ppm dry weight) in six surface sediment samples taken 1/2 m apart at the same station 10 m above the outfall in Calico Creek

	<u>% Loss on Ignition</u>	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe X1000</u>	<u>Pb</u>	<u>Mn</u>	<u>Zn</u>
A	17	.47 ± .07	.3	8	25	6.0	20	30	106
B	16	.44 ± .07	.5	4	15	3.6	25	19	74
C	17	.59 ± .01	1.6	6	19	5.1	25	12	95
D	17	.69 ± .15	.3	4	14	3.9	24	13	76
E	17	.65 ± .12	.6	4	12	2.7	10	6	68
F	16	.83 ± .06	1.2	5	25	4.6	8	27	104
Mean	17	.612	.75	5.2	18	4.3	19	18	87
St.Dev.	.52	.145	.53	1.6	5.6	1.2	7.7	9.3	16.5
Co.Var.	3	23	71	31	31	27	41	52	19

Table 23. Acetic acid hydroxylamine hydrochloride leachable heavy metals (ppm dry weight) in the top 3 cm of surface sediment at individual stations above and below the outfall in Calico Creek

<u>ABOVE OUTFALL</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe X1000</u>	<u>Pb</u>	<u>Mn</u>	<u>Zn</u>
10	.6	2	10	1.6	34	6	90
70	.5	4	11	1.6	36	13	89
300	.6	2	3	1.5	43	6	94
475	.6	1	5	1.5	35	7	97
525	.6	2	3	.8	32	4	88
540	.7	2	4	1.4	36	10	101
595	.8	2	9	.9	55	4	128
605	.7	2	6	1.6	39	9	116
875	.5	1	1	1.9	30	12	57
985	.2	<.5	1	.9	17	24	32
Mean	.58	1.9	5.3	1.4	36	10	89
St.Dev.	.16	.9	3.6	.4	10	6	27
Co.Var.	28	51	68	27	36	63	31
r(distance vs metal)	-.344	-.737	-.759	-.293	-.366	.496	-.449
<u>BELOW OUTFALL</u>							
30	.5	2	4	1.1	30	4	82
100	.5	2	3	.9	26	8	70
200	.4	1	3	1.1	21	13	56
720	.4	6	2	2.0	24	19	63
750	.4	2	3	1.1	19	10	50
800	.4	1	4	1.5	21	24	58
860	.3	1	2	1.2	20	10	50
950	.4	3	2	2.1	22	14	52
1050	.3	1	2	.9	18	11	46
1200	.4	1	2	1.0	25	11	54
1420	.2	1	1	1.1	10	17	23
Mean	.38	1.9	2.5	1.3	21	13	55
St.Dev.	.08	1.5	.9	.4	5	6	15
Co.Var.	23	79	37	33	24	43	27
r(distance vs metal)	-.799	-.141	-.739	-.060	-.694	.446	-.832

Table 24. Heavy metal concentrations in eight acetic acid hydroxylamine hydrochloride leached sediment samples taken 1/2 meter apart 800 meters below the outfall in Calico Creek in 1975

	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe</u> <u>X1000</u>	<u>Pb</u>	<u>Mn</u>	<u>Zn</u>
A	.3 ( 75)	.9 ( 6)	1.0 ( 6)	.9 ( 9)	19 (158)	8 ( 9)	45 (50)
B	.4 ( 50)	1.2 ( 6)	1.2 ( 8)	.9 (10)	22 (169)	8 ( 9)	54 (69)
C	.3 ( 75)	1.0 ( 6)	1.0 ( 7)	.9 ( 9)	21 (150)	8 ( 9)	49 (49)
D	.4 ( 25)	1.2 ( 5)	1.8 ( 9)	1.0 (10)	24 ( 80)	9 (22)	55 (69)
E	.4 ( 75)	2.1 (15)	2.6 (23)	1.1 (15)	26 (124)	9 (10)	58 (58)
F	.5 ( 50)	1.3 ( 5)	1.0 ( 6)	1.0 ( 7)	23 ( 82)	9 (10)	55 (71)
G	2.0 (100)	1.0 ( 5)	.8 ( 4)	.9 ( 9)	20 ( 83)	8 ( 9)	49 (49)
H	.3 ( 75)	1.2 ( 4)	1.2 ( 7)	.9 ( 8)	22 ( 81)	8 ( 9)	53 (48)
Mean	.6 ( 66)	1.2 ( 6)	1.3 ( 7)	1.0 (10)	22.3 (106)	8.4 (10)	52 (58)
St.Dev.	.6	.4	.59	.1	2.3	.5	4
Co.Var.	100	33	44.4	10	9	6	8

( ) % of total metal that is available

amount of organic matter (percent loss on ignition) in the surface sediment. The results of Calico Creek sediment samples are similar to other studies (Klein and Goldberg, 1970; Applequist *et al.*, 1972) in that the elevated mercury concentrations were found nearest to the sewage outfall.

Sediment samples were collected from a series of transects across Calico Creek and analyzed for heavy metals (Table 25 and Figure 33). The results of those analyses show that there is a predictable decrease in organic matter in sediments from high intertidal to subtidal but that the concentrations of heavy metals followed no predictable pattern. The concentration of mercury was relatively constant across the transect; however, samples with low concentrations of organic matter had low concentrations of mercury.

### Turner Creek

Surface sediment samples were taken in Turner Creek at ten stations in May 1972 (Table 26 and Figure 34), twenty stations in May 1974 (Table 27 and Figures 35 through 40), and twenty stations in July 1975 (Table 28 and Figures 41 through 50). As with the Calico Creek samples, each value reported is the mean of three replicates of a single sample.

The mercury concentrations throughout the length of Turner Creek in 1972 (Table 26 and Figure 34) were lower than values in Calico Creek. The mean concentration of 0.14 ppm in Turner Creek corresponds with a mean concentration of 0.80 ppm in Calico Creek. The pattern was repeated in 1974 and 1975; the mean mercury was 0.09 ppm in Turner Creek (1974) compared to a mean of 0.36 ppm for Calico Creek and in 1975 the value in Turner Creek was 0.09 ppm compared to 0.32 ppm in Calico Creek. The mean concentrations for metals other than mercury in sediments listed below are averages of 1974 and 1975 determinations. Lead was the only metal with even a slightly higher mean concentration in Turner Creek.

	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe</u> <u>X1000</u>	<u>Pb</u>	<u>Mn</u>	<u>Zn</u>
Turner Creek	0.5	10	14	6.7	26	20	115
Calico Creek	0.9	17	26	9.8	23	64	130

The sediment organic content in Turner Creek (Figures 35 and 42) ranged from 1%-27% with a mean of 9% for the two-year period compared with a range of 1%-33% with a mean of 17% for Calico Creek 1974-75. Mercury, cadmium and chromium in Turner Creek in 1974 (Table 27) were correlated with the organic content of the sediment. Copper, iron, lead, manganese and zinc showed little or no correlation with organic matter. In 1975 (Table 28) cadmium, mercury and zinc were correlated with organic matter.

Mercury concentration in L. irrorata remained relatively constant in Turner Creek (Tables 26 and 29 and Figure 36). The values were lower than the values reported for L. irrorata from Calico Creek (Tables 11, 13, 15 and

Table 25. Heavy metal concentrations (ppm dry weight) of sediments in transects across Calico Creek

<u>Sediment Location</u>		<u>Distance Apart (meters)</u>	<u>% Loss on Ignition</u>	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe X1000</u>	<u>Pb</u>	<u>Mn</u>	<u>Zn</u>
1-A	<u>J. roemerians</u>	0	31	.24 ± .04	1.2	44	19	6.3	49	106	70
B	<u>S. alterniflora</u>	4	24	.34 ± .09	.9	34	18	15.4	25	102	84
B/C	<u>Just Below S. alterniflora</u>	7	18	.31 ± .04	.8	4	18	3.7	16	13	109
C	<u>Below Oyster Zone</u>	10	14	.34 ± <.005	1.1	6	17	18.6	17	37	105
D	<u>Exposed Mud</u>	12	14	.23 ± .01	.7	1	15	23.0	12	150	105
D	<u>Exposed Mud</u>	14	14	.41 ± .16	.7	17	20	9.5	30	22	100
E	<u>Subtidal</u>	3	5	.05 ± .01	.1	6	1	4.5	2	11	14
2-A	<u>J. roemerians</u>	0	22	.30 ± .01	.4	15	6	10.3	23	36	53
B	<u>S. alterniflora</u>	.5	8	.08 ± .01	.3	8	2	3.0	5	10	12
3-A	<u>S. alterniflora</u>		31	.41 ± .03	.6	28	55	X	18	35	170
C	<u>Exposed Mud</u>		15	.49 ± .02	.9	20	18	7.2	15	30	150
D	<u>Exposed Mud</u>		11	.47 ± .04	1.1	26	37	X	15	84	139
E	<u>Subtidal</u>		11	.28 ± .14	1.1	X	18	X	25	X	150

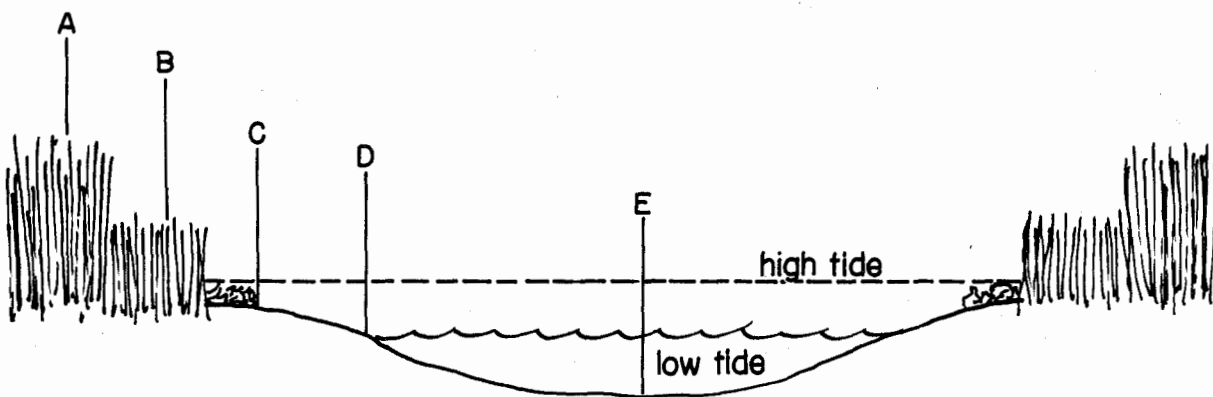


Fig. 33. Locations of sediment samples on transects from high marsh to subtidal. (A) Juncus roemerianus; (B) Spartina alterniflora; (C) Exposed mud just below oyster zone; (D) Exposed mud; (E) Subtidal. The data in Tables 25, 33 and 39 were collected according to this plan.

Table 26. Mercury concentrations found in the top 3 cm of surface sediment (ppm dry weight), in the snail Littorina irrorata (ppm wet weight) and in the oyster Crassostrea virginica (ppm wet weight) from stations in Turner Creek May 1972

	<u>Sediment</u>	<u>Littorina</u>	<u>Crassostrea</u>
	.14 ± .05	.21	.07 ± .01
	.24 ± .10	.19	.04 ± .03
	.16 ± .01	.18	x
	.16 ± .03	.22	x
	.16 ± .08	x	.02 ± .02
	.08 ± .01	x	x
	.03 ± .03	.20	x
	.15 ± .05	.16	.09 ± .01
	.21 ± .13	.15	.04 ± .02
	.11 ± .01	.11	x
Mean	.14	.18	.05
St.Dev.	.06	.04	.03
Co.Var.	.42	20	53

May 1972

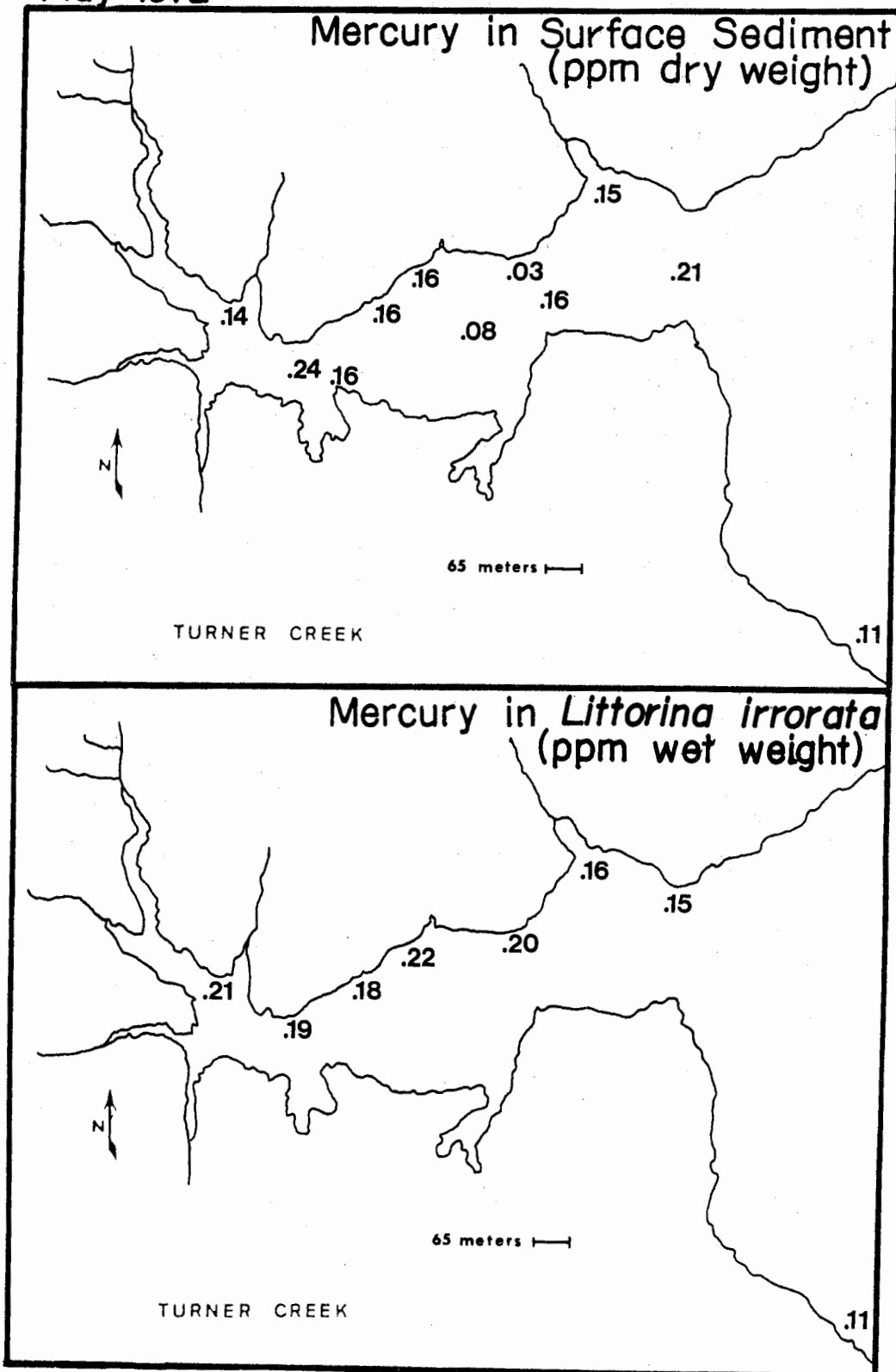


Fig. 34. Total mercury found in Turner Creek during 1972

Table 27. Heavy metal concentrations (ppm dry weight) and percent loss on ignition values (estimate of organic matter) in the top 3 cm of surface sediment from stations in Turner Creek - May 1974

%Loss on Ignition	Hg	Cd	Cr	Cu	Fe X1000	Pb	Mn	Zn	
5	.09 ± .01	.3	10	7	38.0	22	8	55	
16	.10 ± .02	.7	16	8	16.1	9	7	117	
14	.16 ± .02	.6	9	6	5.3	2	8	76	
16	.07 ± .005	.6	12	10	8.5	27	13	95	
18	.21 ± .03	.6	19	9	4.2	35	47	118	
9	.08 ± .02	.6	16	6	8.1	8	12	90	
14	.12 ± .02	.8	8	13	3.6	2	10	127	
5	.05 ± .02	.3	6	9	3.8	14	8	73	
13	.09 ± .01	1.2	8	27	5.3	13	10	250	
7	.14 ± .01	.4	8	50	9.7	102	40	441	
4	.07 ± .01	.2	5	4	7.0	14	38	82	
6	.06 ± .005	.7	11	22	4.5	8	24	290	
5	.08 ± .03	.3	13	4	7.8	10	13	23	
4	.09 ± .01	.2	5	4	7.1	10	39	75	
5	.08 ± .03	.1	5	3	5.2	7	32	50	
4	.03 ± .02	.2	10	6	5.5	10	6	67	
5	.08 ± .01	.4	11	7	4.9	5	11	48	
2	.02 ± .005	.3	7	4	2.2	8	7	11	
1	.02 ± .01	.2	3	1	.9	5	7	3	
1	.07 ± .01	.4	1	X	X	4	X	17	
Mean	8	.09	.5	9	11	7.8	16	18	105
St. Dev.	5	.05	.3	5	12	8.0	22	14	107
Co. Var.	70	54	59	50	109	103	139	78	101
r(%Loss vs metal)		.695	.715	.664	.188	.027	.129	.041	.291

May 1974

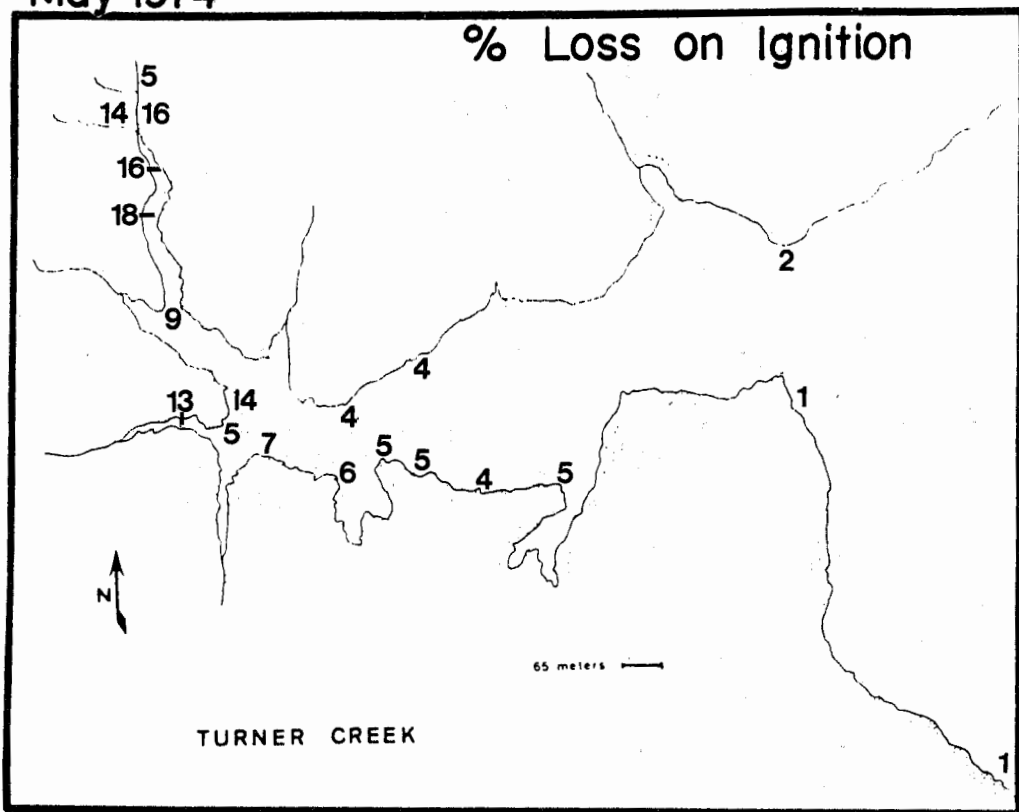


Fig. 35. Percent weight lost on ignition of sediments in Turner Creek 1974

May 1974

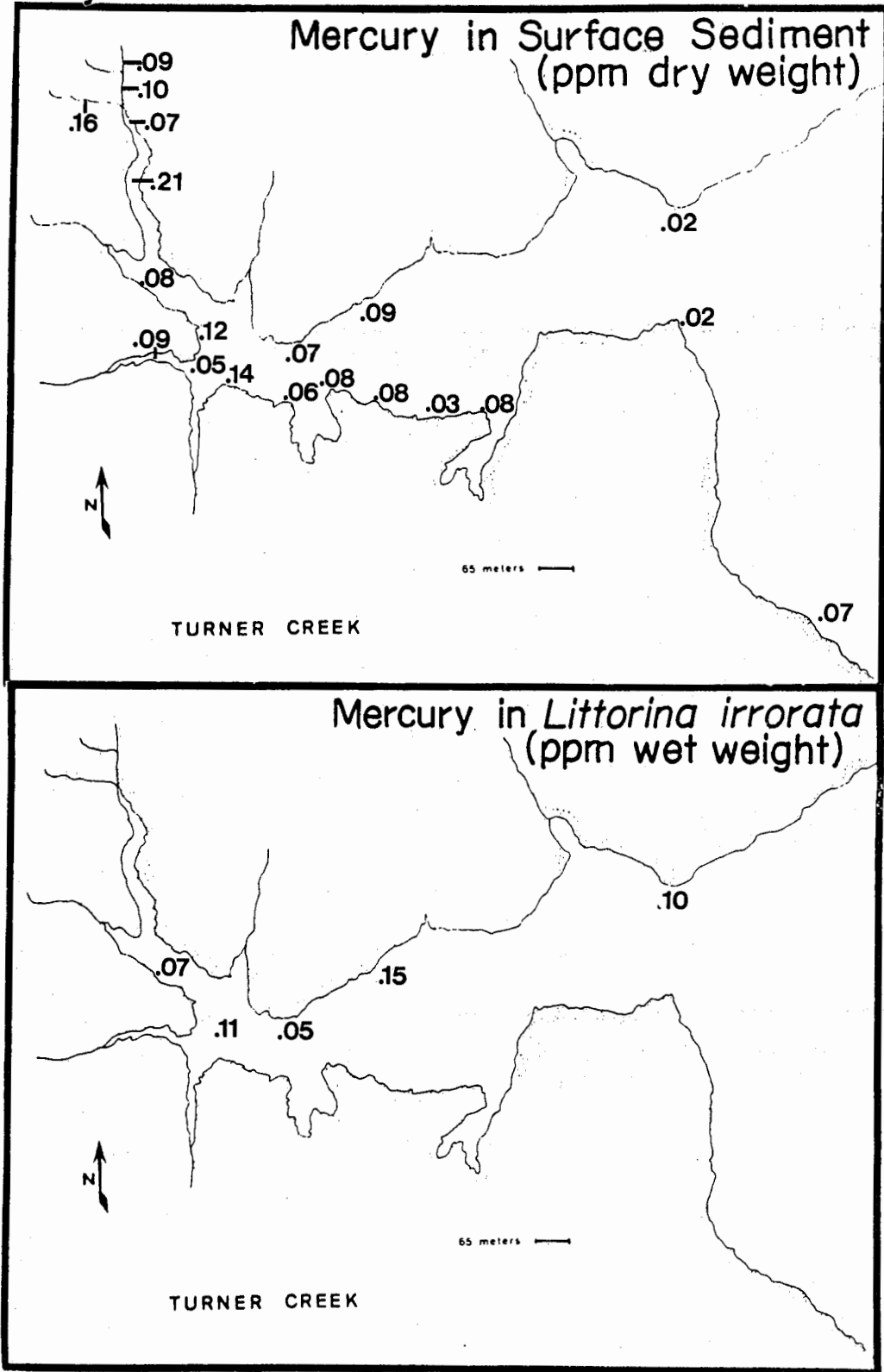


Fig. 36. Total mercury found in Turner Creek 1974

May 1974

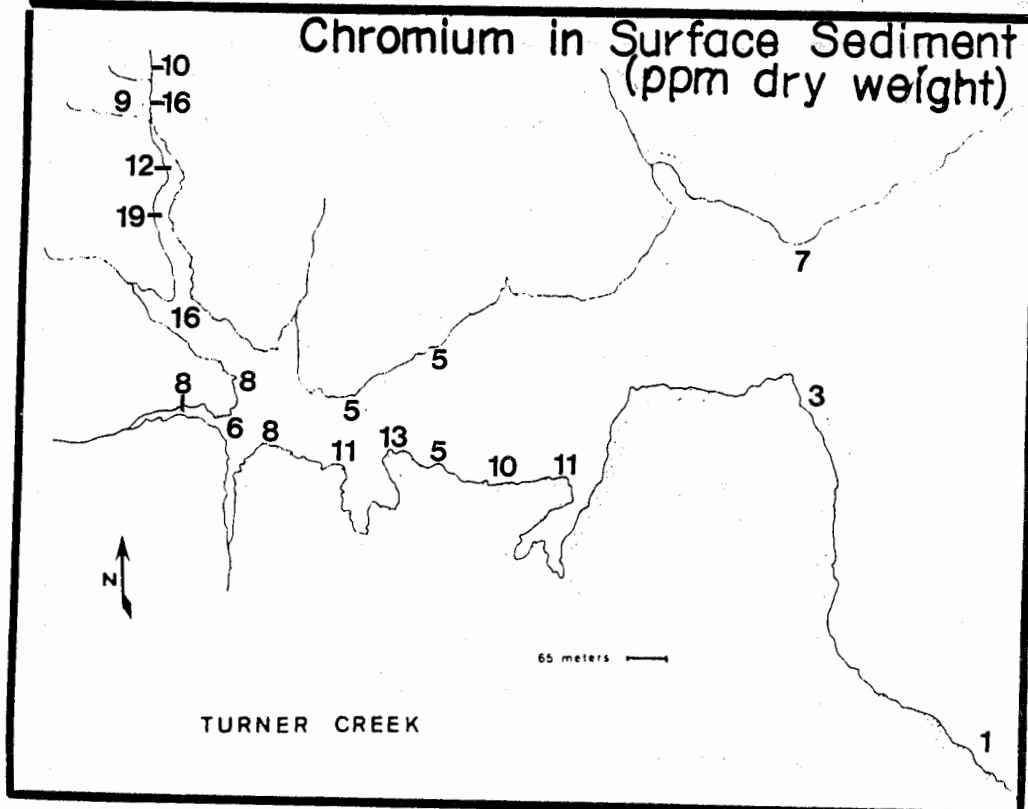
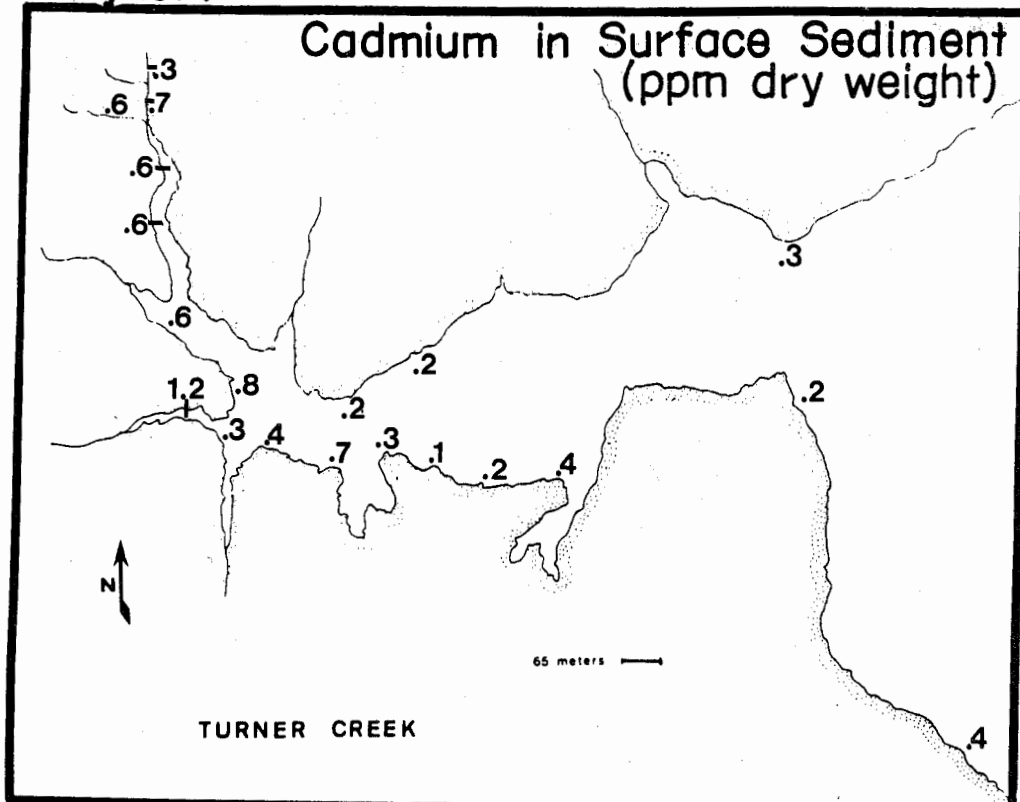


Fig. 37. Cadmium and chromium found in Turner Creek for 1974

May 1974

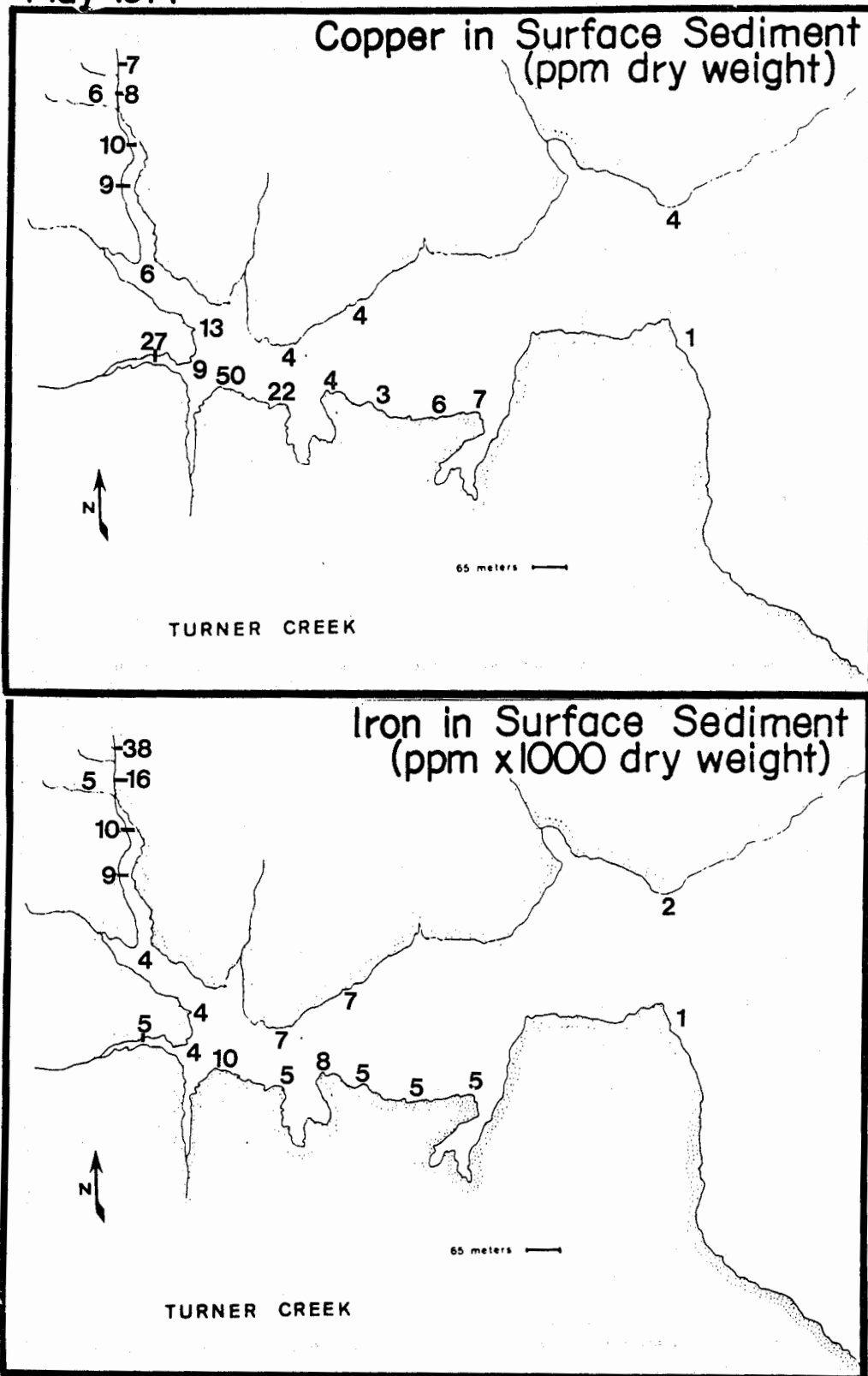


Fig. 38. Copper and iron found in Turner Creek 1974

May 1974

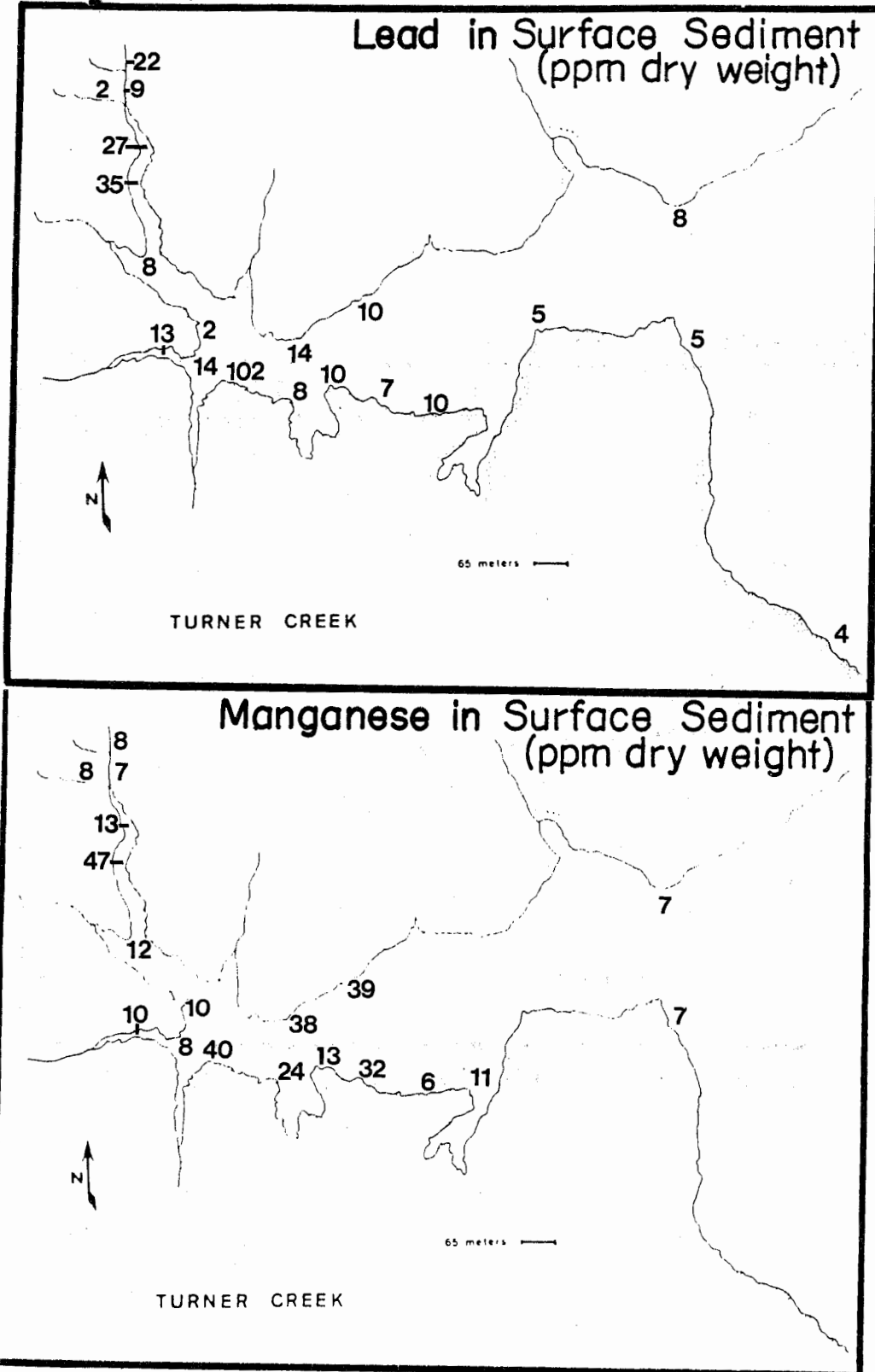


Fig. 39. Lead and manganese found in Turner Creek during 1974

May 1974

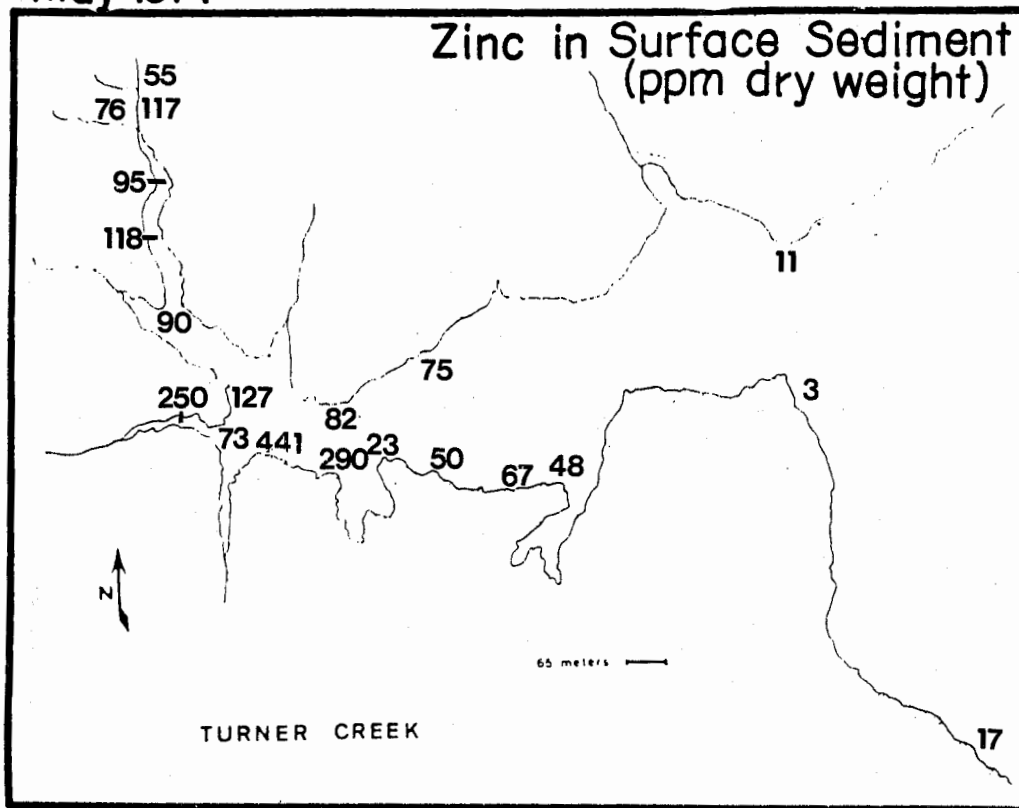


Fig. 40. Zinc found in Turner Creek during 1974

Table 28. Heavy metal concentrations (ppm dry weight) and percent loss on ignition values (an estimate of the organic matter) in the top 3 cm of surface sediment from individual stations in Turner Creek - July 1975

%Loss on Ignition	Hg	Cd	Cr	Cu	Fe X1000	Pb	Mn	Zn	
13	.09 ± .03	.7	20	12	8.5	20	56	122	
16	.09 ± .02	.9	17	11	11.8	19	67	128	
12	.24 ± .01	1.2	24	78	10.5	260	80	>500	
27	.21 ± .01	1.9	3	16	3.6	7	4	270	
18	.27 ± .01	1.1	21	16	10.6	38	76	>340	
15	.18 ± .01	.7	19	19	>6.8	41	X	133	
19	.14 ± .03	1.0	5	23	1.8	7	12	221	
16	.19 ± .05	.8	6	21	3.0	X	8	182	
5	.05 ± .005	.4	10	7	5.4	54	38	41	
12	.14 ± .03	.8	10	29	8.8	130	20	295	
3	.02 ± .005	.3	11	5	4.4	10	20	27	
4	.03 ± .01	.2	11	5	6.2	10	35	35	
3	.03 ± .01	.6	8	2	4.3	47	25	12	
2	.02 ± .01	.1	12	4	4.9	5	28	15	
3	.04 ± .03	.4	10	6	5.8	5	X	15	
2	.03 ± .01	1.0	7	4	4.9	6	28	14	
1	.02 ± .01	.4	14	4	2.2	4	16	7	
4	.03 ± .03	.4	8	4	5.8	6	X	17	
1	.02 ± .01	.2	3	1	.9	5	7	3	
3	.04 ± .01	.5	10	17	.9	14	7	X	
Mean	9	.09	.5	11	14	5.6	36	31	125
St.Dev.	8	.08	.5	6	17	3.2	62	25	142
Co. Var.	85	89	96	52	120	57	171	79	114
r(%Loss vs metal)		.849	.929	.142	.419	.330	.182	.187	.725

1975

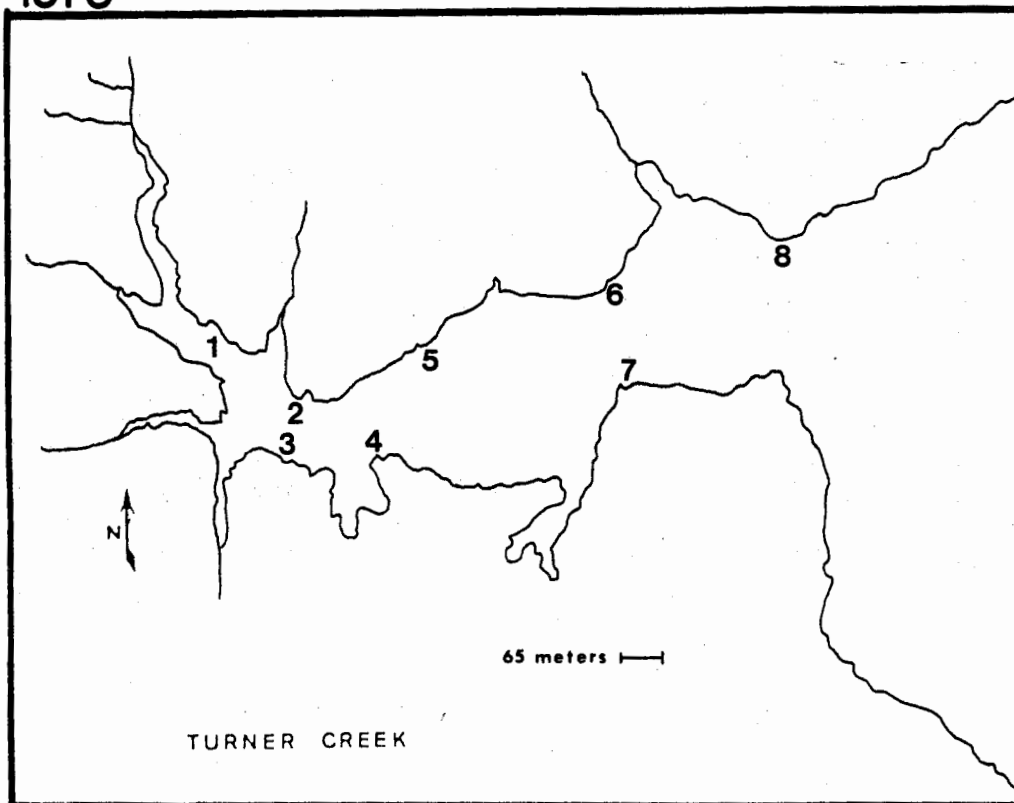


Fig. 41. Heavy metal stations for oysters in Turner Creek during 1975

July 1975

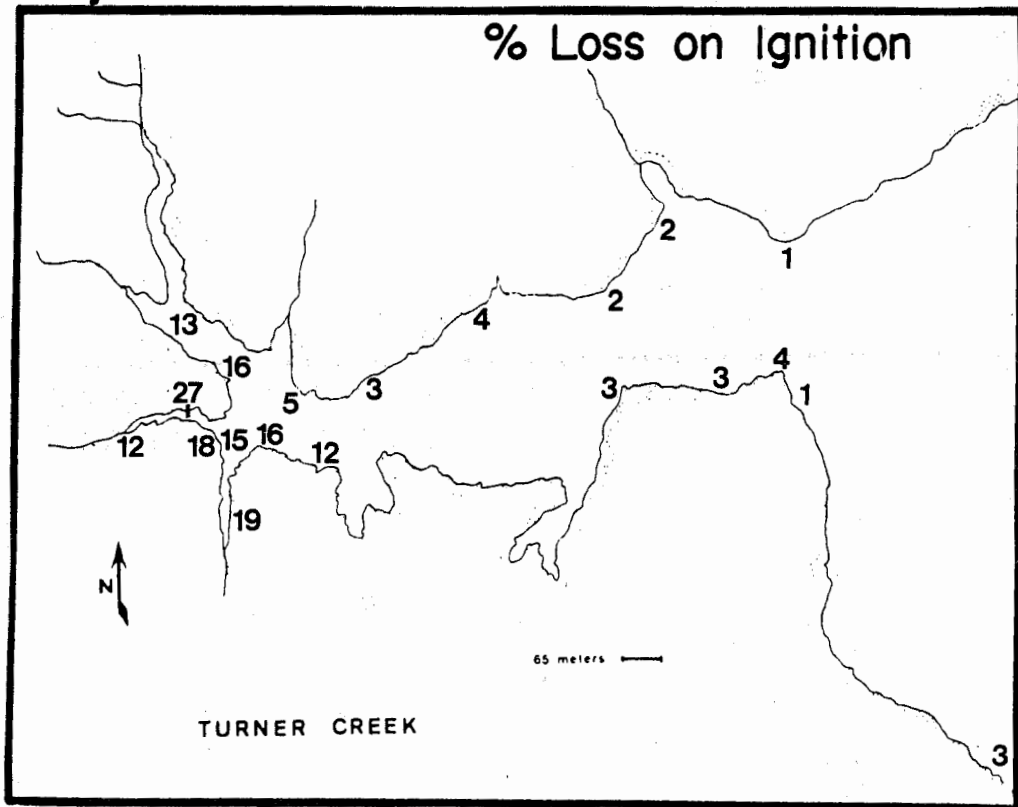


Fig. 42. Percent weight lost on ignition of sediments in Turner Creek 1975

July 1975

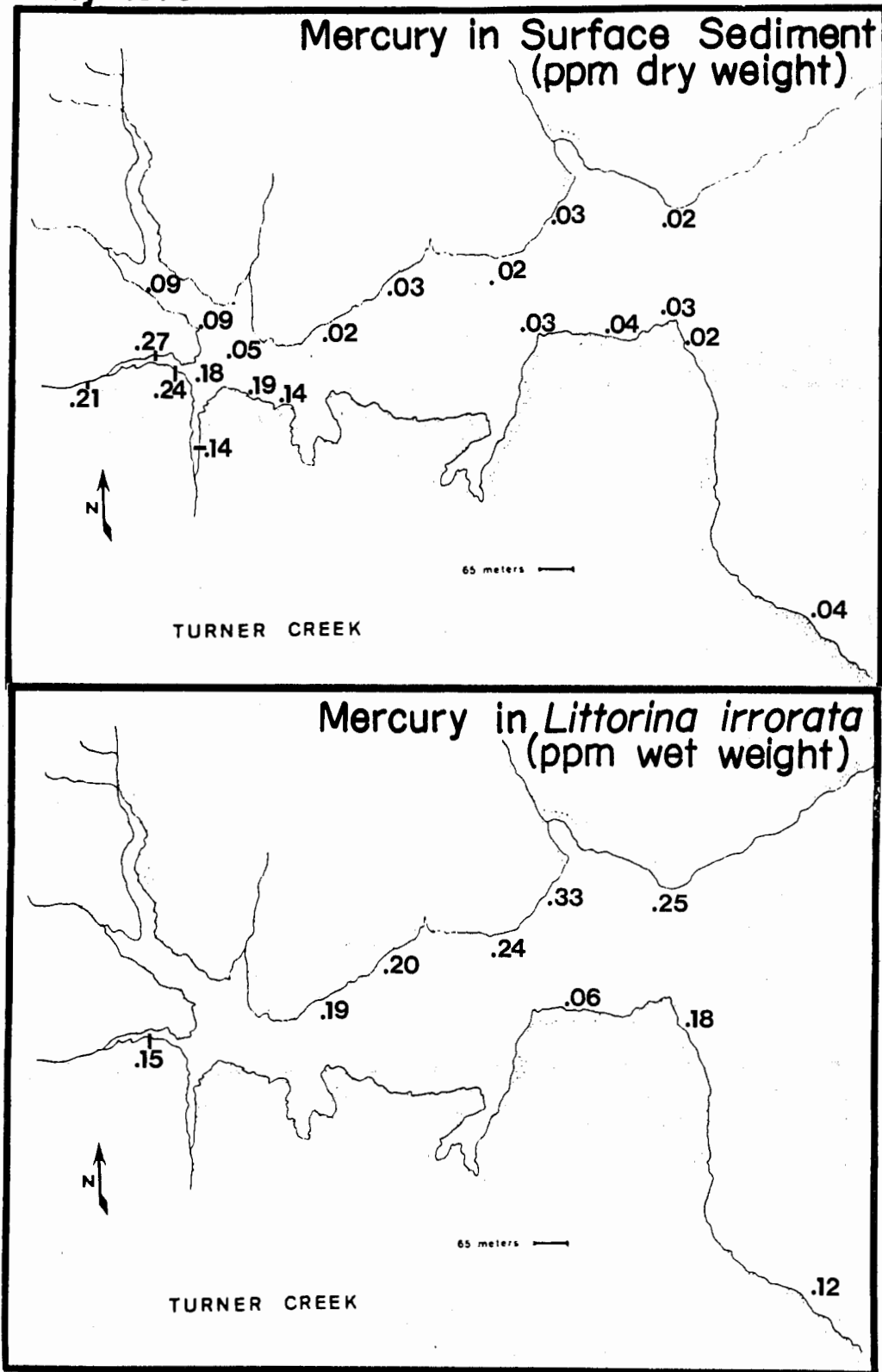


Fig. 43. Total mercury found in Turner Creek during 1975

July 1975

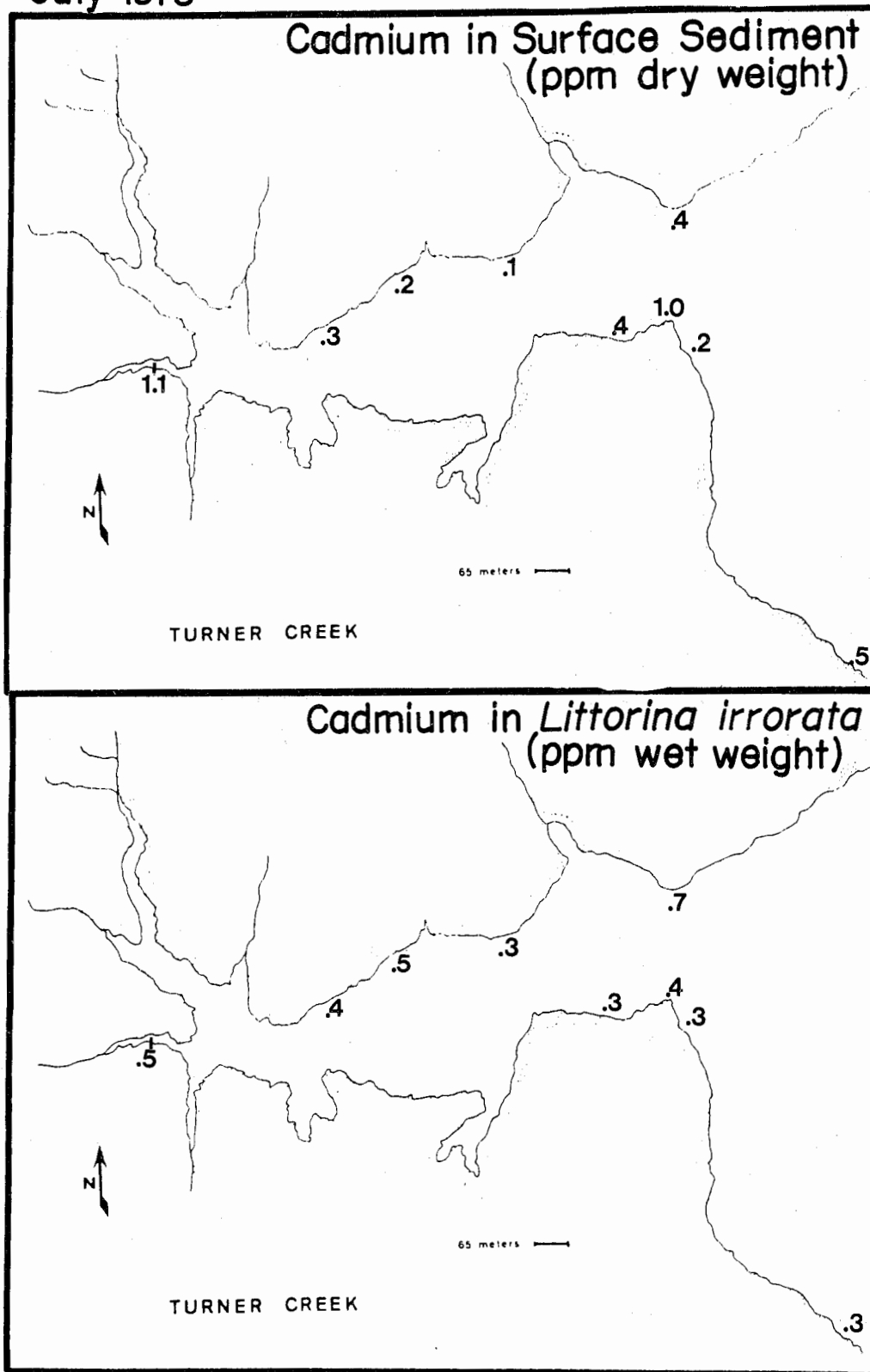


Fig. 44. Cadmium found in Turner Creek during 1975

July 1975

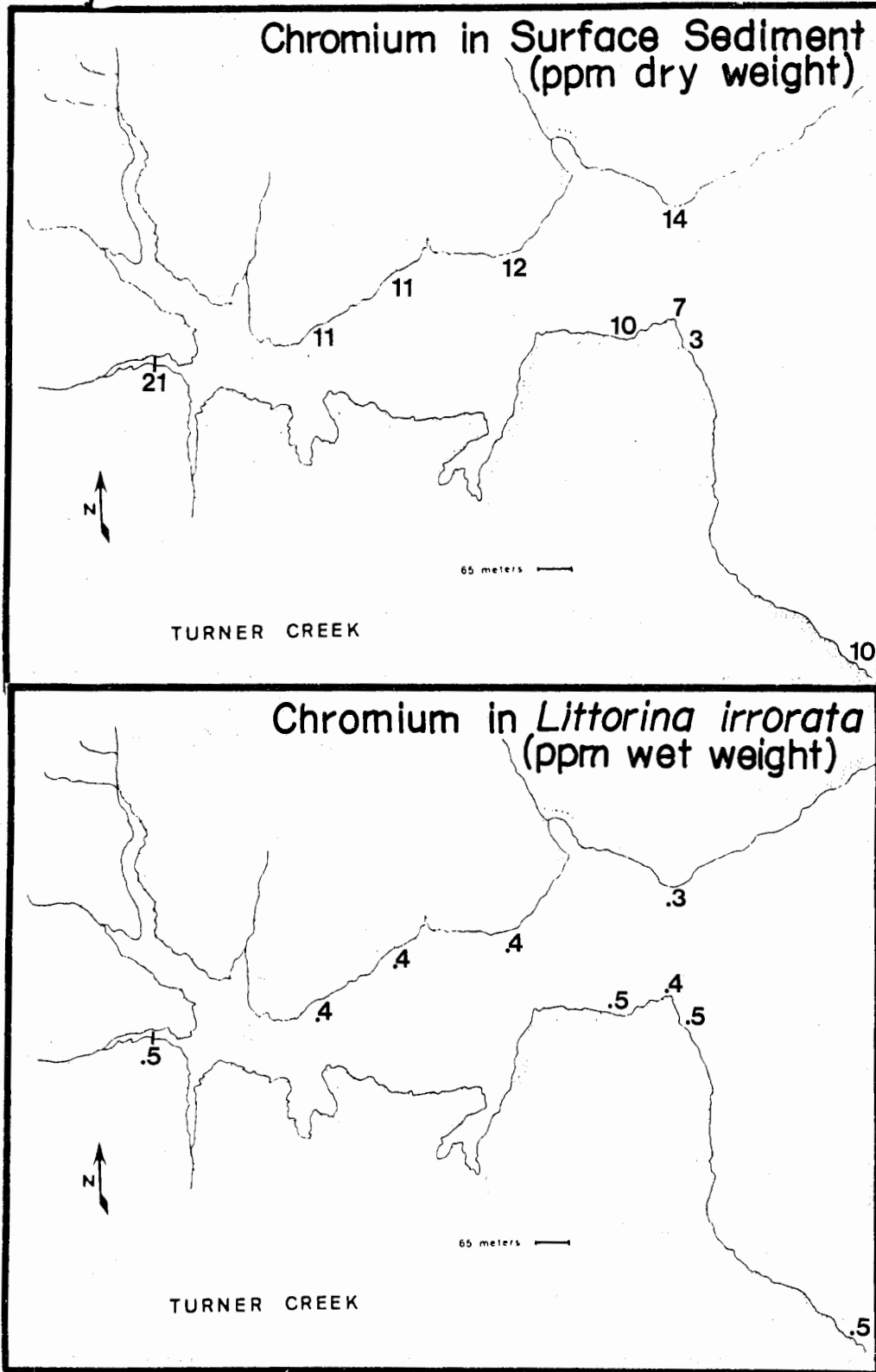


Fig. 45. Chromium found in Turner Creek during 1975

July 1975

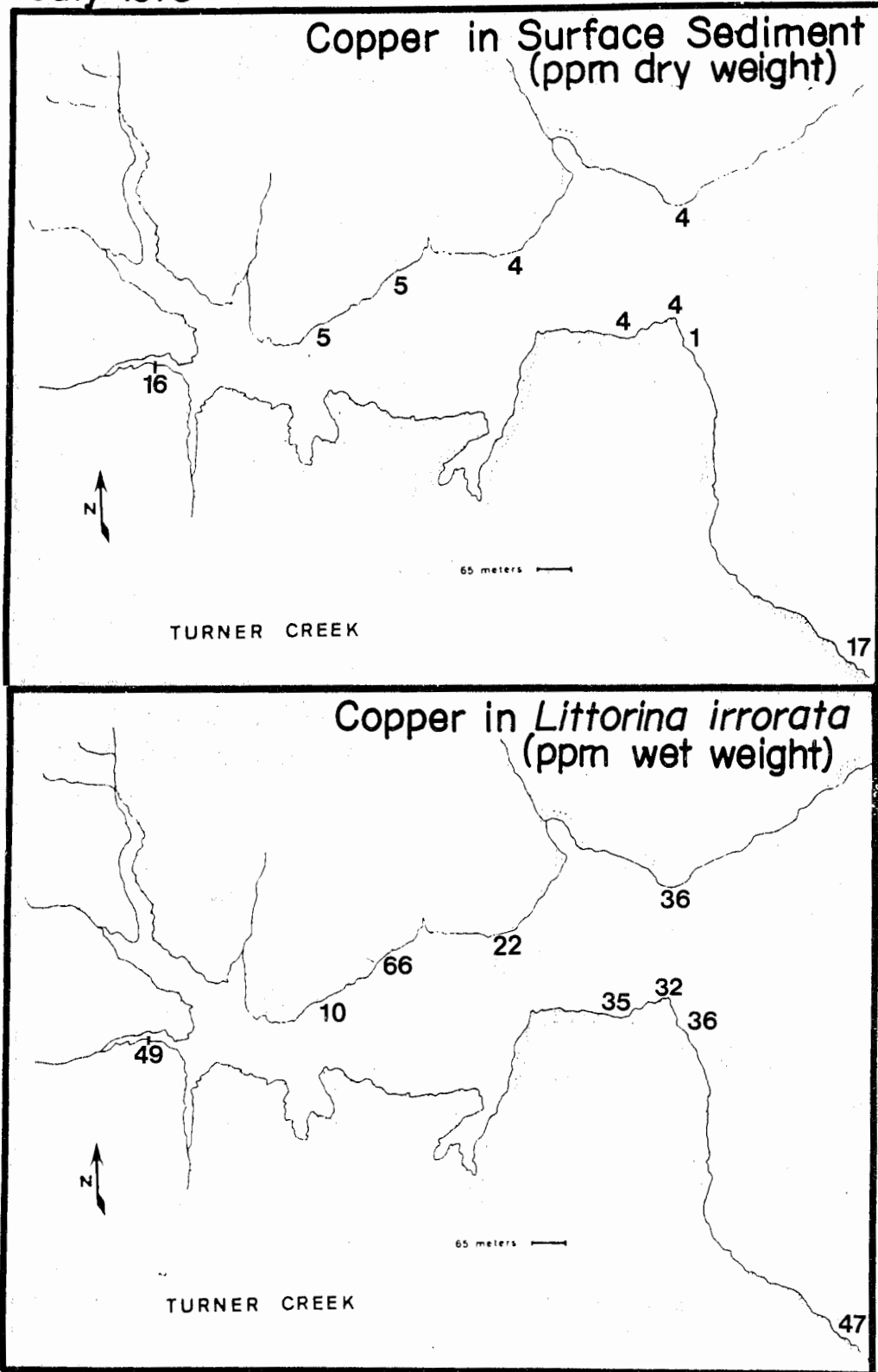


Fig. 46. Copper found in Turner Creek during 1975

July 1975

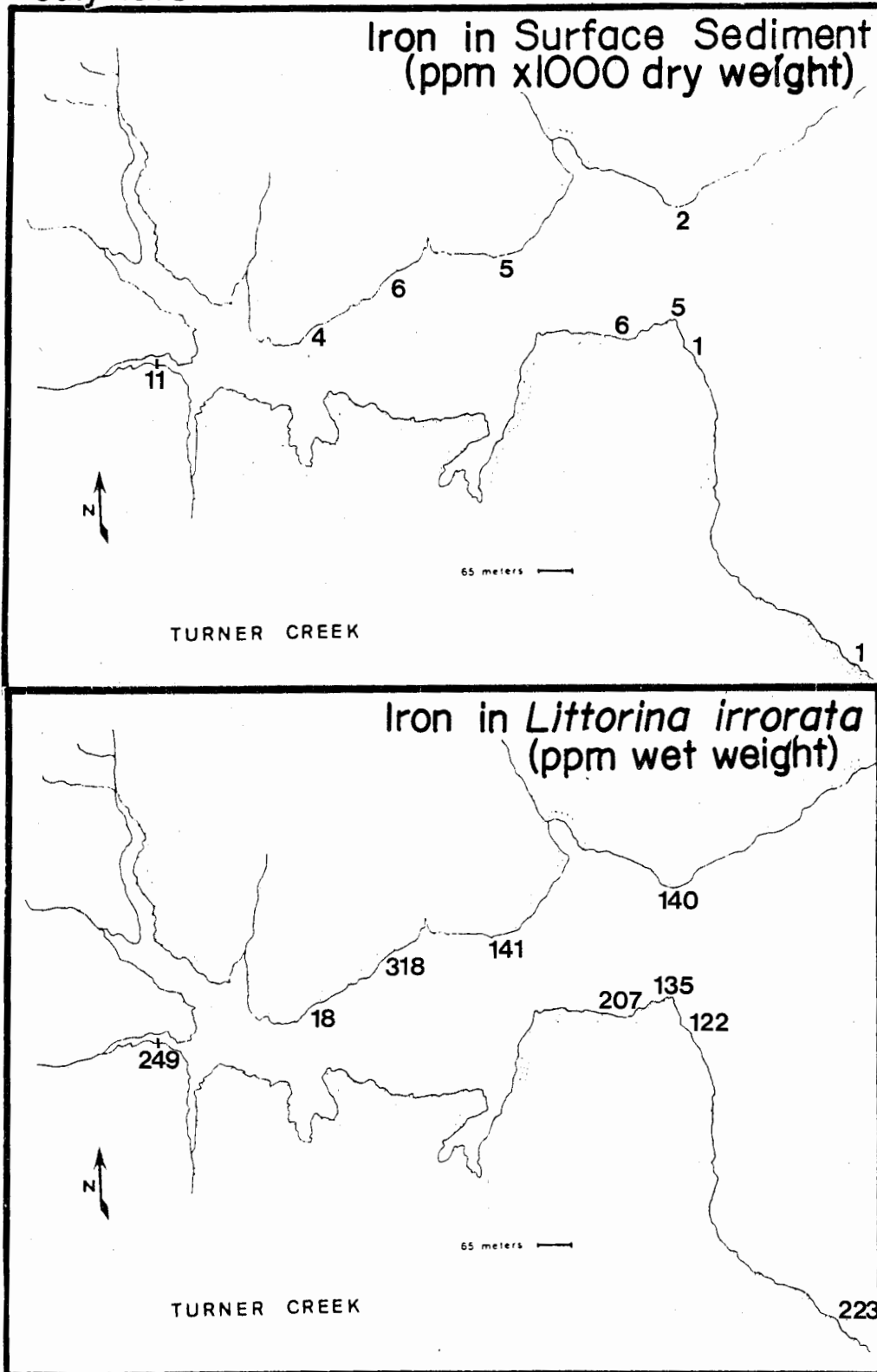


Fig. 47. Iron found in Turner Creek during 1975.

July 1975

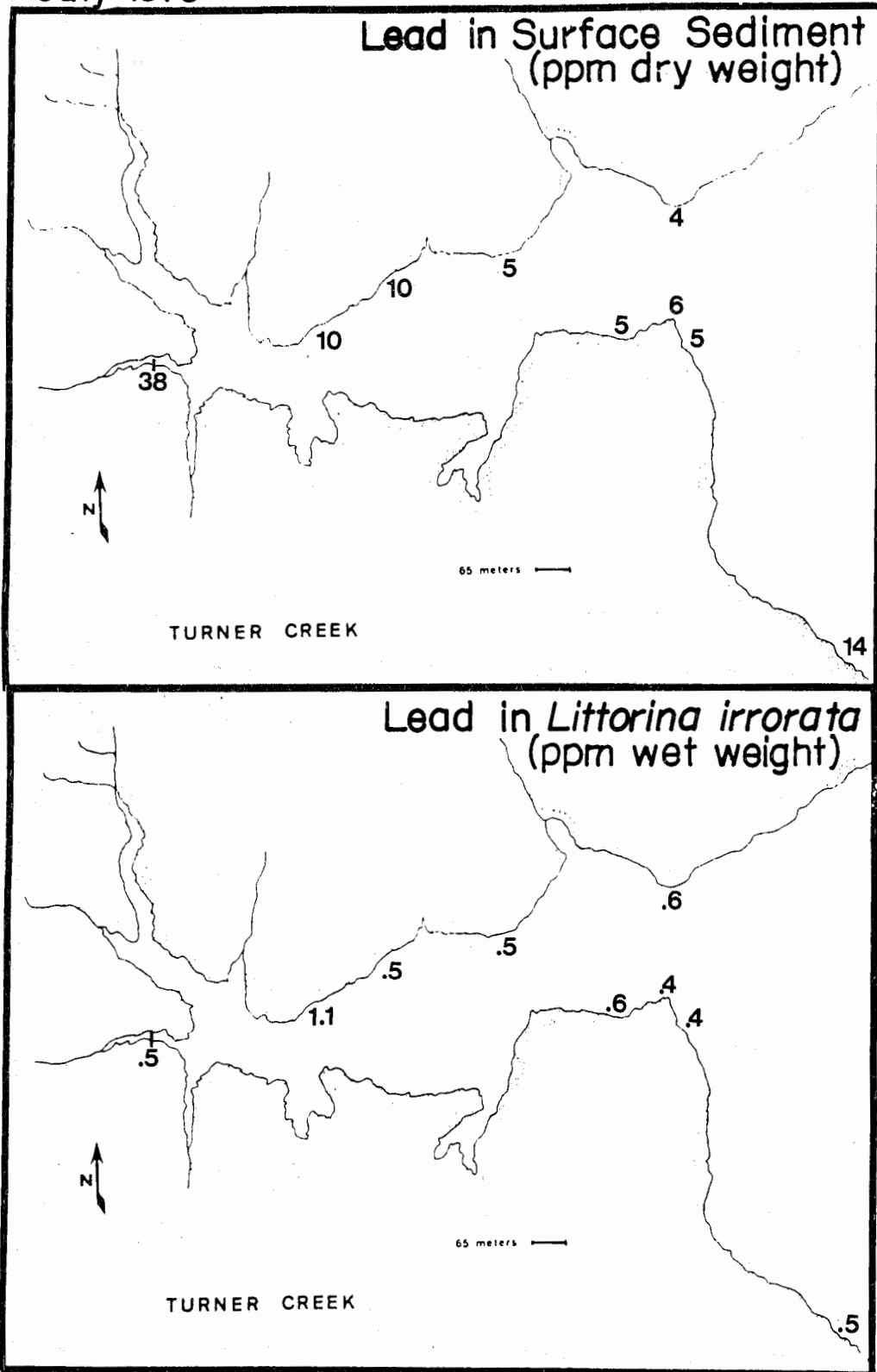


Fig. 48. Lead found in Turner Creek during 1975

July 1975

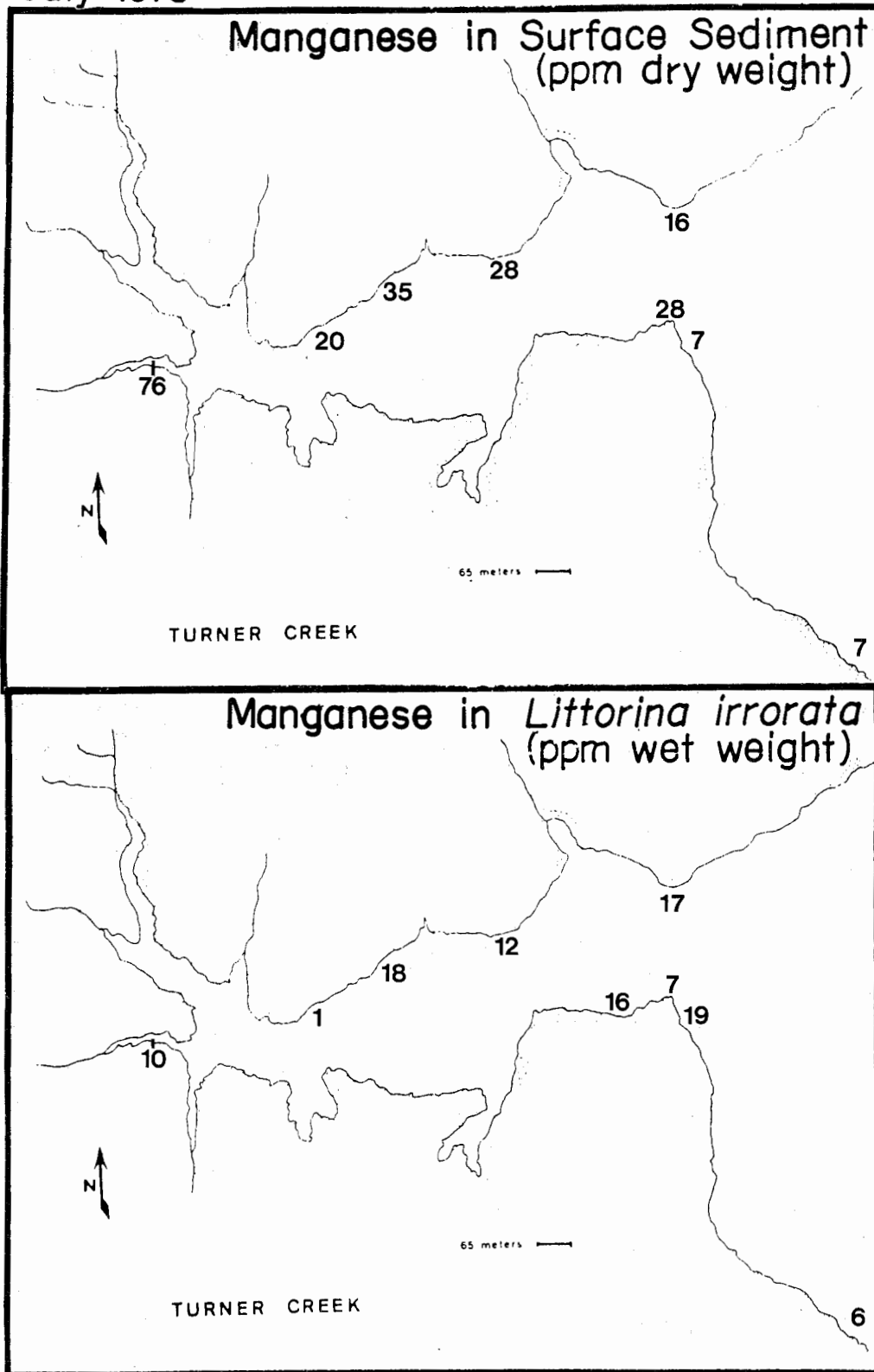


Fig. 49. Manganese found in Turner Creek during 1975

July 1975

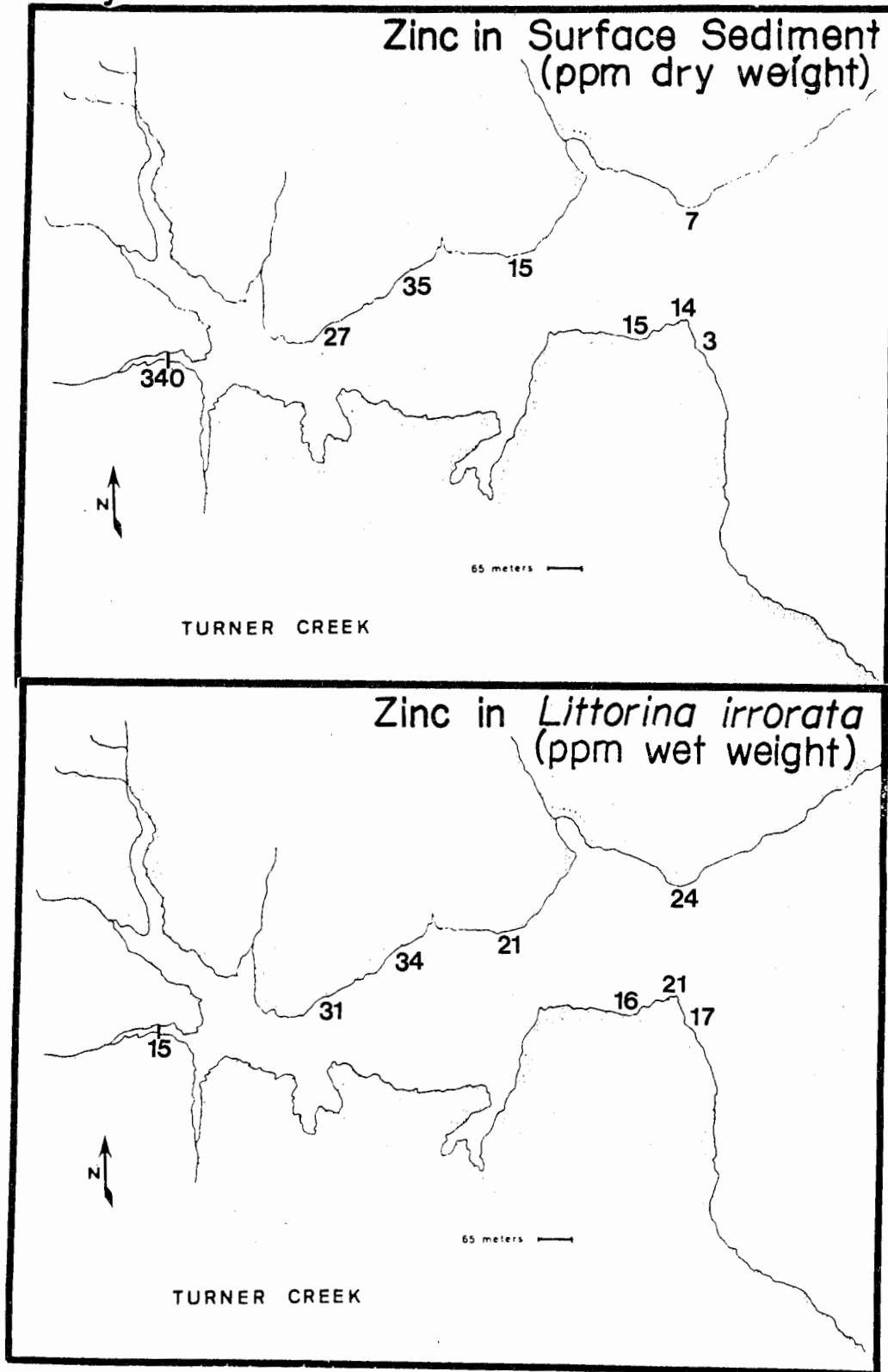


Fig. 50. Zinc found in Turner Creek during 1975

Table 29. Heavy metal concentrations in the snail Littorina irrorata (ppm wet weight) at individual stations in Turner Creek - July 1975

<u>Station Location</u>	<u>Av Wt g</u>	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe</u>	<u>Pb</u>	<u>Mn</u>	<u>Zn</u>
A	1.1	.15 ± .04	.5	.5	49	249	.5	10	15
B	.9	.19 ± .08	.4	.4	10	118	1.1		31
C	.9	.20 ± .09	.5	.4	66	318	.5	18	34
D	.9	.24 ± .03	.3	.4	22	141	.5	12	21
E	.9	.06 ± .02	.3	.5	35	207	.6	16	16
F	.8	.33 ± .21	.4	.4	32	135	.4	7	21
G	.9	.25 ± .02	.7	.3	36	140	.6	17	24
H	.8	.18 ± .09	.3	.5	36	122	.4	19	17
I	.7	.12 ± .09	.3	.5	47	223	.5	6	X
Mean	.9	.19	.4	.4	37	184	.6	12	22
St.Dev.	.1	.07	.1	.1	17	69	.2	6	7
Co.Var.	17	36	25	18	46	38	35	50	32
r(Metal vs <u>Littorina</u> )		-.252	.216	-.108	.363	.480	-.037	-.517	-.349

06

Table 30. Heavy metal concentrations (ppm wet weight) in the oyster Crassostrea virginica at stations in Turner Creek - July 1975

<u>Stations</u>	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe</u>	<u>Pb</u>	<u>Mn</u>
1	.01 ± .01	.3	<1	15	15	<.5	2
2	.01 ± <.005	.5	6	15	83	<.5	4
3	X	.2	<1	19	67	.3	3
4	X	.3	.1	20	53	.3	2
5	.01 ± <.005	.3	1	34	45	<.5	2
6	.02 ± .01	.3	7	25	57	<.5	2
7	X	.3	<1	20	61	.6	2
8	.02 ± .01	.4	<1	9	25	<.5	2
Mean	.01	.3	X	20	51	<.5	2.4
St. Dev.	.01	.08	X	7	22	X	.7
Co. Var.	39	27	X	38	44	X	31

18). There was no significant correlation between increasing mercury in L. irrorata and increasing mercury in sediment. Also, no metal in L. irrorata significantly increased with increased sediment concentrations of the same metal. Manganese concentration in L. irrorata was the only metal that significantly decreased with increasing metal in sediment. Mean values for metal concentrations in L. irrorata from Calico Creek in 1975 were higher than the parallel concentrations reported in Turner Creek.

The oyster C. virginica in Turner Creek in 1972 (Table 26) had a mercury concentration of  $0.05 \pm 0.03$  ppm and the oysters in Calico Creek (Table 11) had a similar concentration ( $0.08 \pm 0.04$  ppm). The mercury concentration in Turner Creek oysters decreased in 1975 (Table 30 and Figure 41) to  $0.01 \pm 0.01$  ppm; during the same period mercury concentrations in Calico Creek oysters decreased to the same value of  $0.01 \pm 0.01$  ppm (Table 16).

A Turner Creek station that was sampled every 0.5 m was located in an exposed flat at low tide; the relatively sandy condition at this site is evident in the low percent loss on ignition of 8.5% compared to 17% for the Calico Creek subsampled site (Table 31).

Table 31. Heavy metal concentrations (ppm dry weight) in eight surface sediment samples taken 0.5 m apart in Turner Creek

	<u>%Loss</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe</u> <u>X1000</u>	<u>Pb</u>	<u>Mn</u>	<u>Zn</u>
O	5	.4	11	11	4.9	7	47	48
A	9	.6	16	8	13.8	7	48	45
B	8	.6	9	17	4.1	1	42	84
C	9	1.1	12	17	6.8	7	46	80
D	8	1.0	10	12	9.3	10	20	75
E	9	.5	14	18	4.1	15	34	69
Mean	8.5	.7	12	12	7.2	8	40	67
St.Dev.	.5	.3	3	6	3.8	5	11	17
Co.Var.	6	40	25	51	54	59	27	25

No accumulation of mercury occurred in the S. alterniflora stems, leaves and roots in Turner Creek. There was, however, a significant ( $r=0.686$ ) correlation of mercury in stems and leaves with increasing mercury concentrations in sediments. Roots had only a slight correlation ( $r=0.399$ ) with increasing mercury in sediment. There is a slight decrease in mercury in the stems, leaves and roots downstream in Turner Creek (Table 32 and Figure 51). Copper is concentrated in roots of S. alterniflora while the stems and leaves did not significantly differ from the sediment. Copper concentration in stems and leaves is correlated ( $r=0.962$ ) with increased concentration in the sediment.

Table 33 presents data on concentrations of heavy metal in sediments from transects across the intertidal zone in Turner Creek. There was no pattern

of heavy metal distribution which related to the intertidal position of the sediment samples. Piston cores were taken 10, 400, and 1400 m from the outfall in Calico Creek and at two stations in Turner Creek to determine the vertical distribution of metals. The cores were extruded onto parafilm, split down the center, subsampled at 5 and 10 cm intervals to 50 cm, then analyzed as previously described. The top 20 cm in the 10-meter station in Calico Creek is a very fine sediment that is compressed somewhat in the coring operation; therefore, values reported in panel "a" of Figure 52 have some negative vertical distortion near the top. Mercury concentrations in the surface sediments (Table 34 and Figure 52) in Calico Creek were 1.16 ppm, 0.68 ppm, 0.24 ppm for 10 m upstream, 400 m upstream, and 1400 m downstream, respectively. The sediment in Turner Creek is much sandier and there was no compaction in the surface sediment. All cores had similar mercury concentrations of less than 0.1 ppm at a depth of 10 cm and deeper. In Turner Creek there was only 0.16 ppm and 0.34 ppm of total mercury in the surface sediment and then the concentration became undetectable (<0.005) at 10 cm depth and deeper. The only core that had mercury enrichment in other than the surface was the 10-m upstream station in Calico Creek. This is possibly the result of years of mercury rich particulate matter discharged from the sewage treatment plant.

The surface layer of all the cores has more mercury than the lower layers of the cores, suggesting that the mercury in both creeks has come from human activities.

#### Other North Carolina Estuaries

Wrightsville Beach. The concentrations of metals in the sediments below the outfall of the Wrightsville Beach sewage treatment plant (Table 35 and Figure 53) decreased with increasing distance from the outfall. Within 30 m of the outfall metal concentrations are at natural levels. The isolated increase in all metals at 60 m is unexplained; however, it occurred in all the analyses and is probably a real phenomenon. With the exception of manganese all metals have a very high correlation with percent organic matter in the sediment.

Newport River. A series of samples to determine the concentrations of metals in sediments was collected in the vicinity of the Newport sewage treatment plant in January 1976 (Table 36 and Figure 54). Samples were taken both above and below the outfall. Unlike the Wrightsville Beach transect, there was no regular decrease in concentration of metals with distance above or below the outfall. The low mercury values found in the sediment near this outfall (0.07 and 0.05 ppm) agree well with the low mercury values in the effluent (0.1 ppm) from this plant.

West Carteret High School, Morehead City. Three samples of sediment were collected below the outfall at West Carteret High School in May 1975 and analyzed for the concentration of metals (Table 37). The sediments around this outfall were very sandy (2% organic matter) and had no visible dark organic matter present.

Table 32. Heavy metal concentrations (ppm dry weight) in the stems, leaves, and roots of the marsh grass *Spartina alterniflora* compared to total and leached metal data for stations in Turner Creek

Distance downstream (m)	% Loss	Hg	Cd	Cr	Cu	Fe X1000	Pb	Mn	Zn
0 St & Lea		.16± .02	.2	1	5	.1	1	6	30
Root		.16± .04	1.5	3	22	1.3	5	8	89
Total Sed	18	.21± .03	.6	19	9	4.2	35	47	118
Leach Sed		x	.3	.6	1	1.0	32	7	60
130 St & Lea		.07± .01	.2	2	9	1.1	3	12	15
Root		.21± .01	.4	4	16	1.5	12	11	56
Total Sed	16	.09± .02	.9	17	11	11.8	19	67	128
Leach Sed		x	.6	2	1	1.7	28	9	84
290 St & Lea		.05± .01	.2	4	3	1.0	3	86	19
Root		.02± <.005	.8	13	14	1.3	7	23	88
Total Sed	3	.02± .01	.3	11	5	4.4	10	20	27
Leach Sed		x	.2	.6	2	.6	6	3	24
425 St & Lea		.05± .02	.1	2	3	.5	1	12	15
Root		.16± .04	.2	2	8	1.0	1	11	56
Total Sed	4	.03± .01	.2	11	5	6.2	10	35	35
Leach Sed		x	.1	.7	1	1.1	10	8	23
650 St & Lea		.13± .05	.2	1	2	.1	1	20	21
Root		.18± .02	.8	12	56	5.0	5	41	139
Total Sed	2	.02± .01	.1	12	4	4.9	5	28	15
Leach Sed		x	.1	.4	.4	.6	4	5	9
975 St & Lea		.07± .05	.1	1	2	.2	1	14	13
Root		.05± .03	.6	3	10	1.6	4	25	64
Total Sed	1	.02± .01	.4	14	4	2.2	4	16	7
Leach Sed		x	.6	.1	.2	.1	1	2	2
St & Lea mean±StDev		.08± .05	.2±.1	2±1	4±3	.5±.5	2±1	25±30	19±16
Root mean±StDev		.13± .08	.7±.4	6±5	21±18	2.0±1.5	6±4	20±12	72±38
Total Sed mean±StDev		.07± .08	.4±.3	14±3	6±3	5.6±3.3	14±12	36±19	55±54
Leach Sed mean±StDev		x	.3±.2	.7±.7	.8±.7	.9±.5	14±13	6±3	34±32
r(St & Lea vs Total Sed)		.686	-.046	-.165	.962	.669	.044	-.459	.414
r(Root vs Total Sed)		.399	.279	-.224	-.190	-.105	.251	-.606	.297
r(St & Lea vs Leach Sed)		x	-.111	.217	.214	.787	.206	-.514	.275
r(Root vs Leach Sed)		x	-.099	-.187	-.343	-.256	.468	-.612	-.346
r(St & Lea vs Distance)		-.252	-.625	-.303	-.691	-.408	-.437	-.065	-.573
r(Root vs Distance)		-.397	-.369	.072	.120	.368	-.432	.664	.120

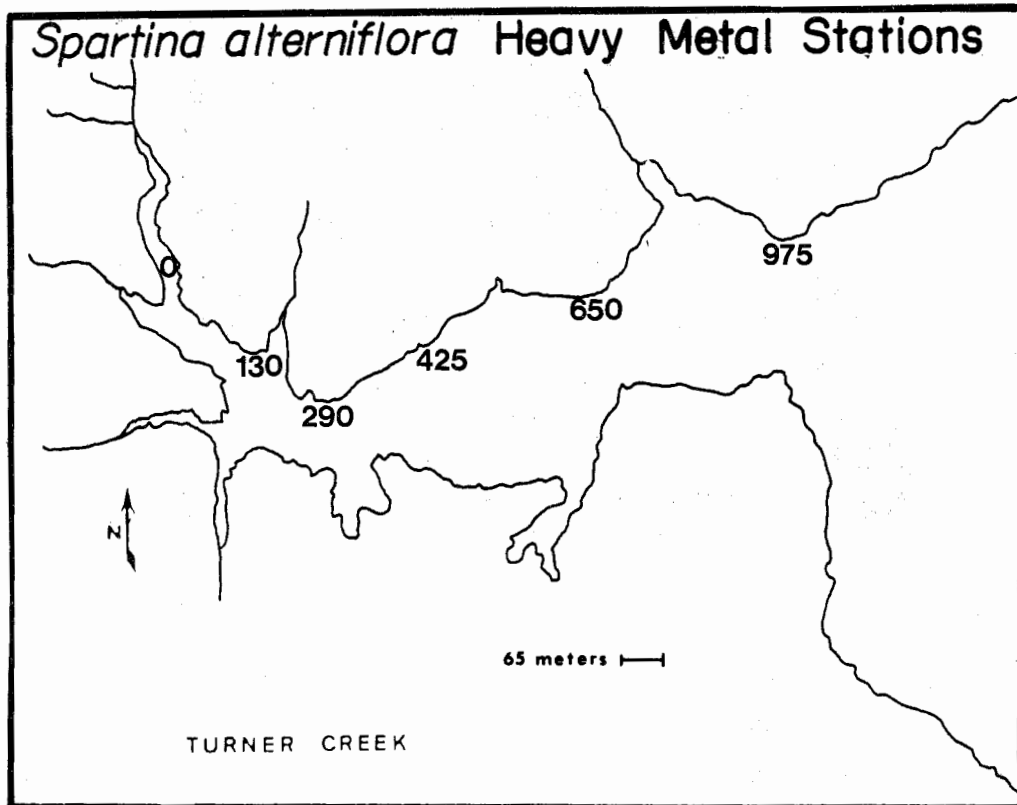


Fig. 51. *Spartina alterniflora* heavy metal stations in Turner Creek

Table 33. Heavy metal concentrations (ppm dry weight) of sediments in transects across Turner Creek

<u>Sediment Location</u>		<u>Distance Apart (meters)</u>	<u>% Loss on Ignition</u>	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Pb</u>	<u>Mn</u>	<u>Zn</u>
1-A/B	<u>S. alterniflora/J. roemerians</u>	0	16	.10 ± .2	X	28	18	36	59	17
C	Exposed Mud	1	9	.08 ± .02	.6	16	6	8	12	90
D	Exposed Mud	3	16	.14 ± .01	1.9	54	17	21	66	162
E	Subtidal	5	15	.16 ± .04	.7	16	10	X	X	120
2-C	Exposed Mud	0	20	.08 ± .02	.7	12	8	10	38	110
D	Exposed Mud	1	15	.10 ± .06	.7	16	8	9	7	117
E	Subtidal	.5	9	.09 ± .01	.4	X	4	11	18	54
3-B	<u>S. alterniflora</u>	5	5	.06 ± .03	1.0	9	4	7	22	30
C	Exposed Mud	0	3	.04 ± .03	.4	10	6	5	X	15
D	Exposed Mud	.5	3	.03 ± .01	1.5	8	9	12	27	29
D	Exposed Mud	1	4	.01 ± .01	.6	5	8	7	X	25
4-B	<u>S. alterniflora</u>	2	3	.04 ± .01	.5	9	17	14	64	17
C	Exposed Mud	0	4	.07 ± .04	.4	10	11	9	54	23
5-B	<u>S. alterniflora</u>	0	18	.27 ± .01	1.1	21	16	38	76	145
C	Exposed Mud	2	26	.16 ± <.005	1.6	18	8	28	76	129

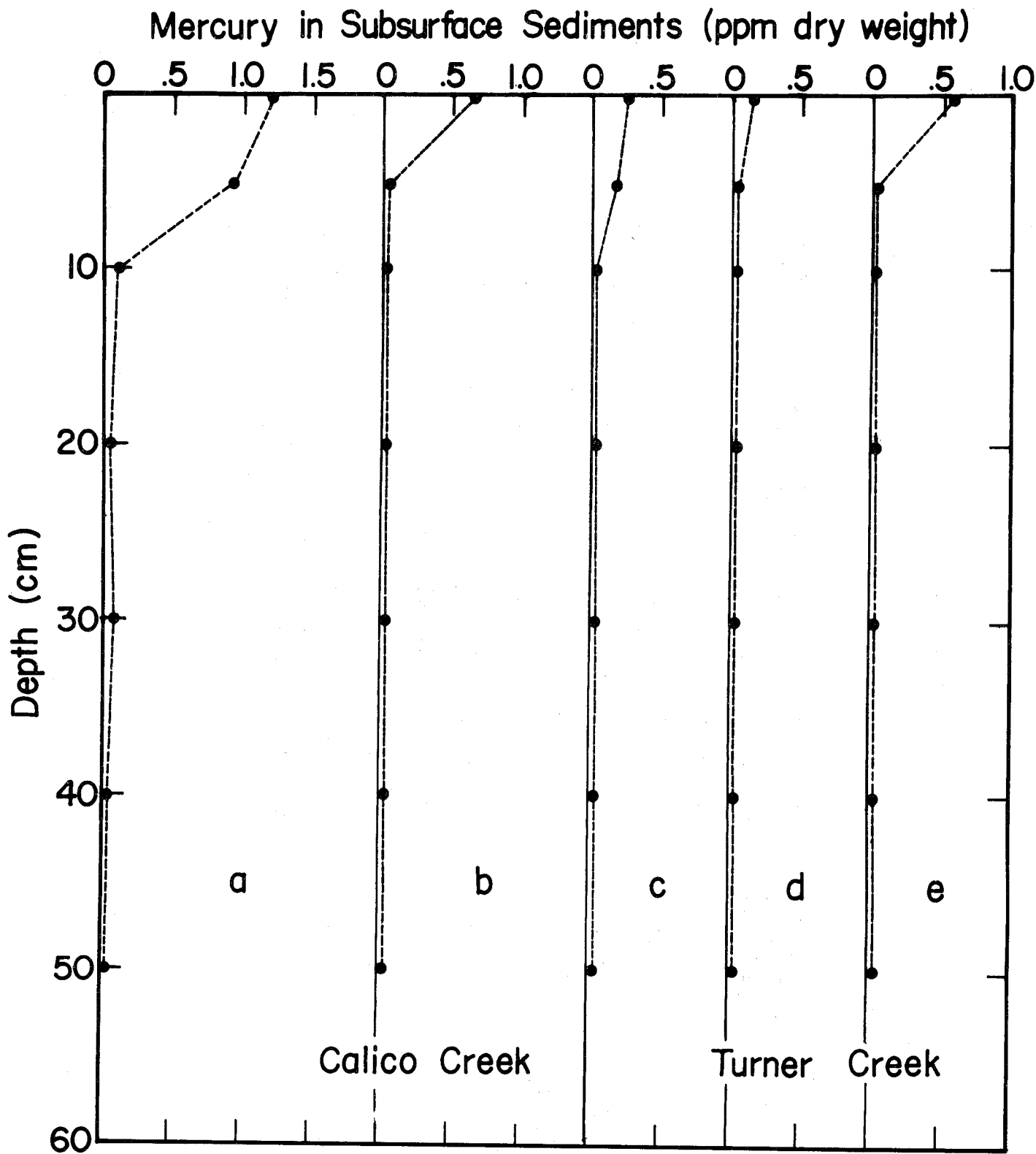


Fig. 52. A comparison of mercury concentrations in sediment cores from Calico Creek (a,b,c) and Turner Creek (d,e). The locations of the cores are: (a) 10 meters above outfall; (b) 400 meters above outfall; (c) 1400 meters below outfall; (d) 660 meters below origin and (e) 120 meters below origin. The numerical values for the points are given in Table 34.

Table 34. Comparison of mercury concentrations in sediment cores from Calico Creek and Turner Creek. These data are presented graphically in the same format in Figure 52.

<u>Depth</u>	<u>Calico Creek</u>			<u>Turner Creek</u>	
	<u>(a)</u> <u>10m above</u> <u>outfall</u>	<u>(b)</u> <u>400m above</u> <u>outfall</u>	<u>(c)</u> <u>1400m below</u> <u>outfall</u>	<u>660m below</u> <u>origin</u>	<u>120m below</u> <u>origin</u>
Surface	1.16±.08	.68±.19	.24±.02	.16±.02	.34±.04
5	.84±.06	.02±.02	.19±.03	<.005	.02±.02
10	.07±.01	.02±.02	.04±.01	<.005	<.005
20	.05±.02	.04±.01	.02±.01	<.005	<.005
30	.09±.05	.02±.01	<.005	<.005	<.005
40	.01±.02	<.005	.01±.01	<.005	<.005
50	.01±.01	.01±.01	.005	<.005	<.005

Table 35. Heavy metal concentrations (ppm dry weight) and percent loss on ignition values (estimate of organic matter) in the top 3 cm of surface sediment from stations to 155 meters below the Wrightsville Beach sewage treatment plant

<u>Meters from Effluent</u>	<u>% Loss on Ignition</u>	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe X1000</u>	<u>Pb</u>	<u>Mn</u>	<u>Zn</u>
10	23	2.10 ± .24	2.3	30	545	9.1	66	11	636
15	7 <sup>1</sup>	.40 ± .01	.6	11	125	2.6	20	9	159
20		1.00 ± .10							
25	15	.95 ± .11	1.2	9	195	2.4	29	8	275
30	13	x	3.6	13	168	3.3	24	8	246
40		.12 ± .08							
55	1	.07 ± .06	.1	4	3	.5	2	6	6
60	3	.08 ± .02	.2	15	7	2.9	7	20	21
63		.20 ± .02							
65	1	.07 ± .06	.1	8	6	.7	5	4	12
68		.10 ± .02							
70	1	.07 ± .03	.2	6	12	1.3	8	8	30
75	1	.07 ± .03	.1	4	2	.6	2	5	7
100	1	.06 ± .02	.1	3	1	.5	2	2	2
110	1	.11 ± .08	.1	3	1	.5	3	6	4
150	1	.09 ± .01	.1	4	1	.5	1	4	4
155	2	.07 ± .01	.1	10	1	2.3	2	16	8
r(distance vs con)	-.670 <sup>1</sup>	-.590	-.576	-.522	-.619	-.519	-.673	-.673	-.648
r(% Loss vs Metal)	x	.979	.813	.958	.950	.879	.966	.211	.978

<sup>1</sup>extremely sandy sediment - this station was located in the turbulence created by the effluent.

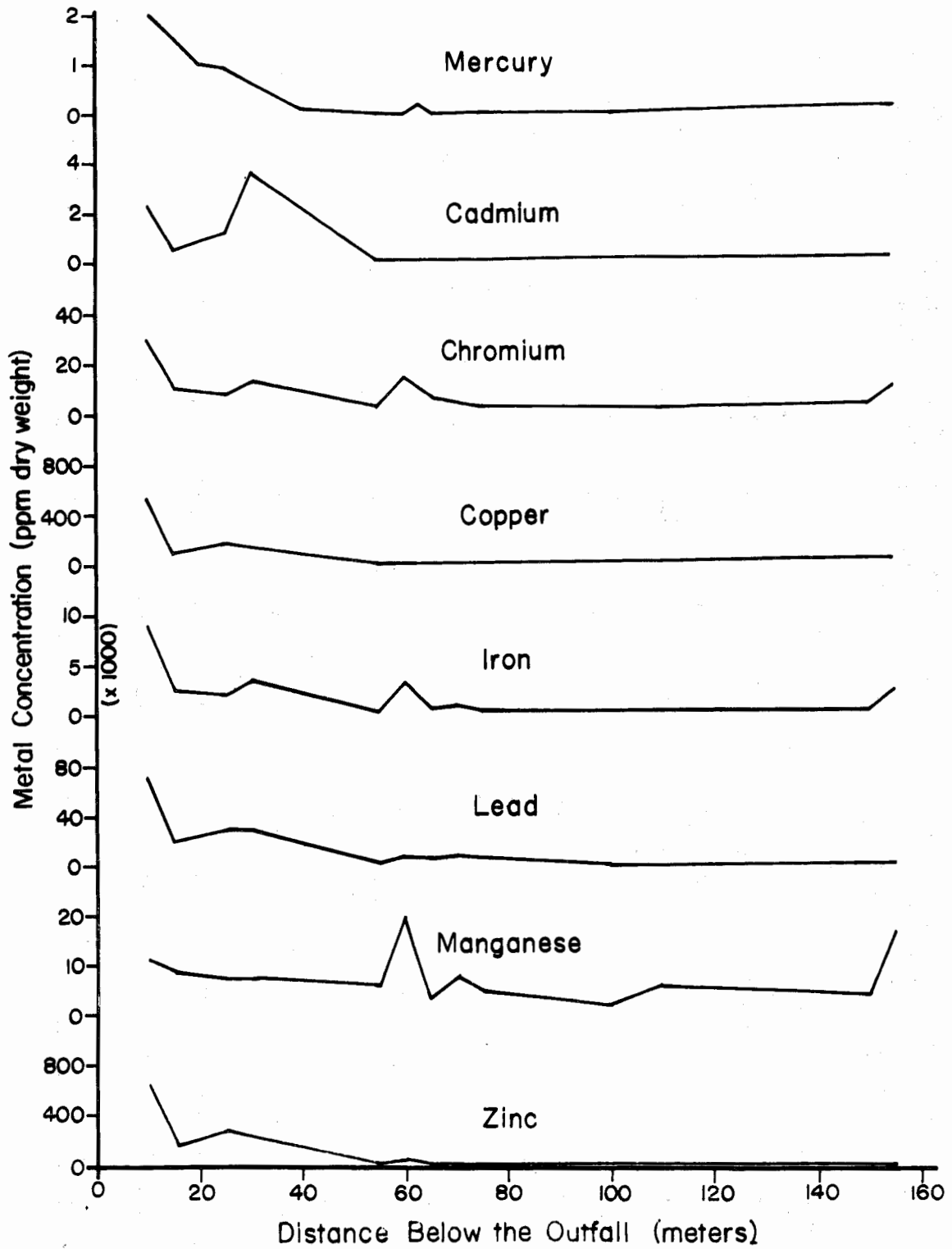


Fig. 53. Metal concentrations in sediment located below the Wrightsville Beach sewage treatment plant

Table 36. Heavy metal concentrations (ppm dry weight) and percent loss on ignition values (estimate of organic matter) in the top 3 cm of surface sediment from stations to 300 meters below the Newport sewage treatment plant. These numerical values are graphically represented in Figure 54.

101

Station Nos.	Meters from Outfall	% Loss on Ignition	Hg	Cd	Cr	Cu	Fe X1000	Pb	Mn	Zn	
1	10	10	.07 ± .01	.1	4	2	4.6	6	13	25	
2	15	11	.05 ± .01	.1	5	3	5.8	16	18	28	
3	25	36	.23 ± .01	.2	14	2	2.6	24	33	58	
4	50	37	.27 ± .03	.3	9	5	7.0	11	44	71	
5	70	37	.14 ± .04	.3	18	3	9.0	27	53	80	
6	200	37	.11 ± .03	.2	18	3	8.5	25	64	52	
7	235	25	.15 ± .01	.1	19	3	3.9	24	81	71	
8	300	33	.19 ± .04	.3	16	3	10.0	21	33	106	
r(distance vs con)			.345	.129	-.040	.700	.088	.465	.476	.591	.570
r(% loss vs metal)			.742	.806	.783	.370	.396	.608	.537	.710	

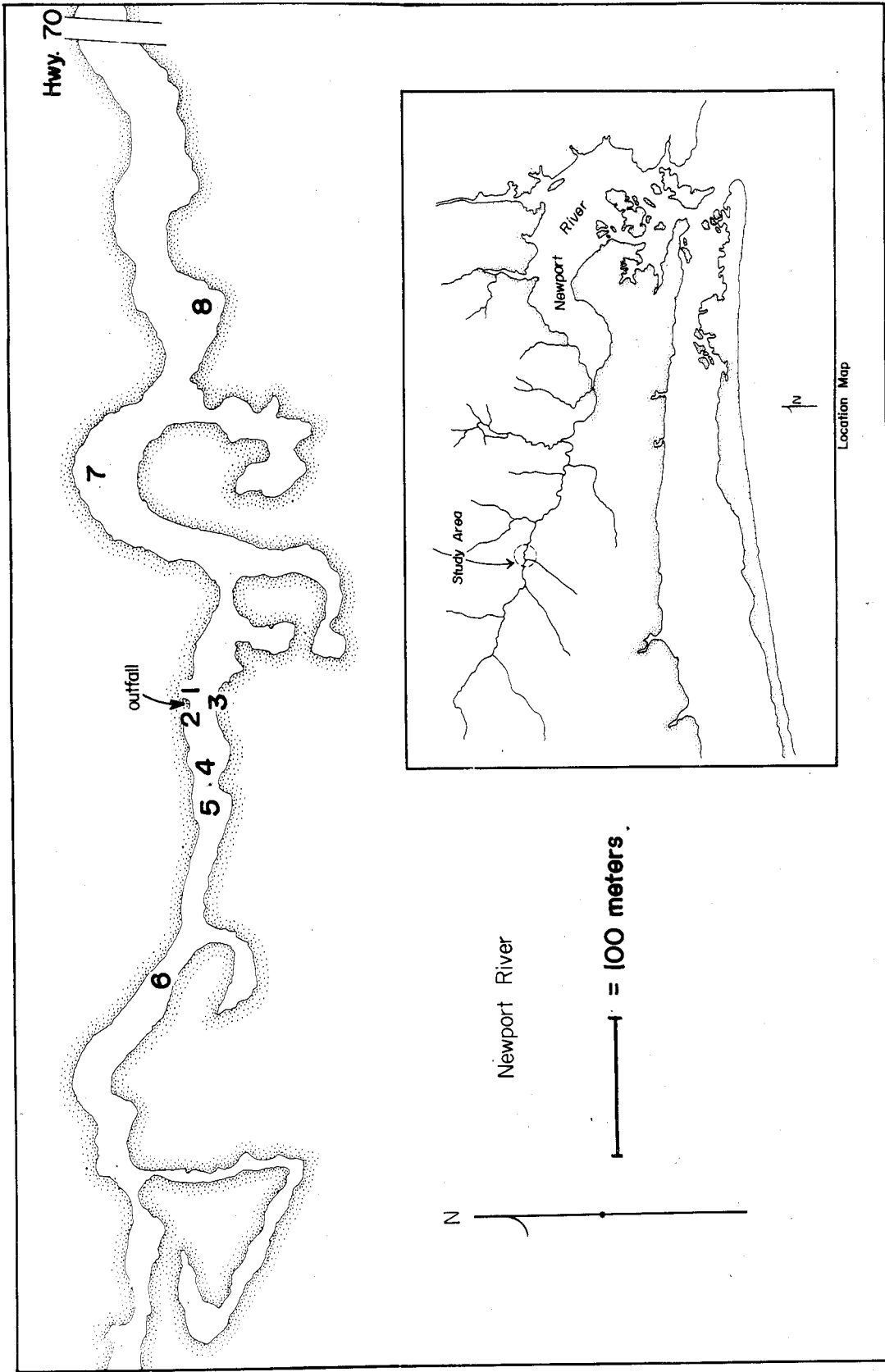


Fig. 54. Map of the Newport River showing sampling stations for heavy metals

Table 37. Heavy metal concentrations (ppm dry weight) and percent loss on ignition values (estimate of organic matter) in the top 3 cm of surface sediment from stations to 15 m below the West Carteret High School sewage treatment plant

Meters Below Outfall	% Loss on Ignition	Hg	Cd	Cr	Cu	Fe X1000	Pb	Mn	Zn
0	2	.11 ± .01	.06	2	50	1.0	13	7	62
10	2	.28 ± .05	.01	4	16	2.2	43	10	26
15	2	.23 ± .02	1.25	3	14	1.7	36	11	54

South River. The South River is a tributary of the Neuse River, located in Carteret County, which receives no point source input of metal-rich wastes. Samples were collected and analyzed to determine a baseline of values for this estuary. Table 38 and Figure 55 present the data obtained.

Table 38. Heavy metal concentrations (ppm dry weight) and percent loss on ignition in the top 3 cm of surface sediments in South River

	% Loss on Ignition	Hg	Cd	Cr	Cu	Fe X1000	Pb	Mn	Zn
1		.04 ± .005							
2	2	.09 ± .02							
3	69	.25 ± .09	.8	4	1	4.9	39	10	35
4	48	.09 ± .02	.1	12	11	1.7	3	27	15
5	24	.08 ± .01	.3	16	12	5.5	28	150	38
6		.08 ± .01							

Newport River Estuary. A site 3.5 km from the Morehead City outfall in the middle of the Newport River estuary was sampled as being representative of an area that receives no point source input of metals (Table 39). The values are very similar to those found 1400 m below the outfall in Calico Creek suggesting that at that location the Calico Creek metal concentrations are unaffected by the Morehead City sewage treatment plant.

#### Sewage Treatment Plants

The range of mercury concentrations entering sewage treatment plants varies temporally within individual plants and varies spatially between plants. Morehead City (Table 40) had a 3.3 ppb mercury content in the influent at the beginning of this study and in April 1976 an 11.0 ppb mercury load. This must be the result of a new mercury source using the system or a general increase in mercury disposal in the Morehead City area; both explanations seem unlikely when it is noted that no parallel increase occurred in

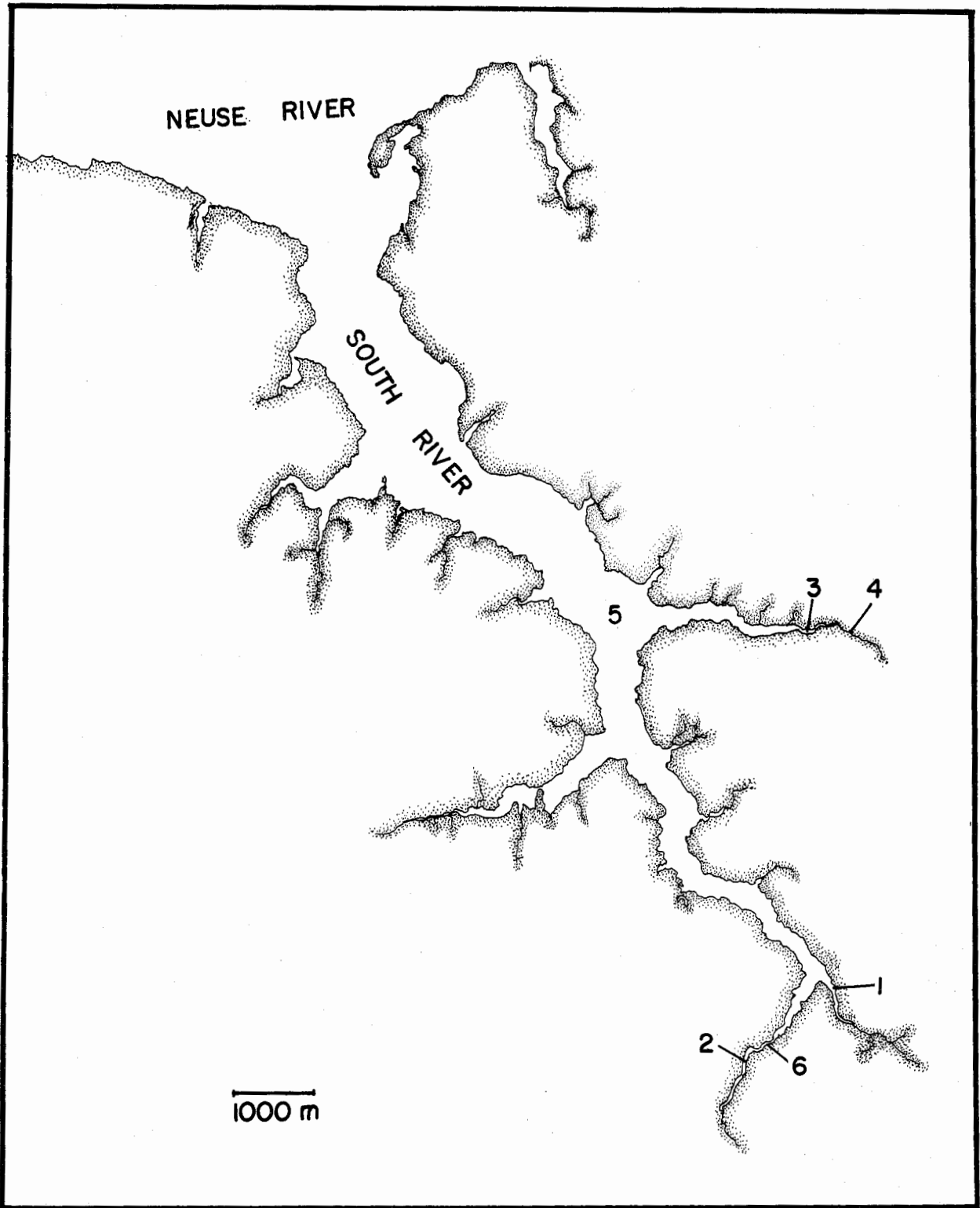


Fig. 55. Map of South River showing sampling stations for heavy metals

Table 39. Heavy metal concentrations (ppm dry weight) and percent loss on ignition (as an estimate of organic matter) in a Newport River estuary transect

Sample Location	Meters	% Loss		Cd	Cr	Cu	Fe X1000	Pb	Mn	Zn
		on Ignition	Hg							
NPR-1 Exposed Mud	10	2	.03 ± .01	.6	1	2	2.6	24	25	11
2 Below <u>S. alterniflora</u>	6	1	.03 ± .01	X	X	X	X	X	X	X
3 <u>S. alterniflora</u>	3	3	.02 ± .01	.4	1	2	4.1	7	30	13
4 <u>S. alterniflora</u> / <u>J. roemerians</u>	0	7	.02 ± .60	.6	9	5	9.8	14	70	29
5 Exposed Mud		10	.04 ± <.005	.6	14	10	6.9	20	17	99
6 Exposed Mud		8	.08 ± .01	X	15	6	5.0	8	44	52
7 Exposed Mud		X	X	X	13	6	X	12	X	77
8 Exposed Mud		12	.04 ± .01	.3	17	10	7.0	15	46	47
Mean			.04	.5	10	6	5.9	14	39	47
St.Dev.			.02	.1	7	3	2.5	6	19	33
Co.Var.			50	28	66	56	43	43	49	70
r(% Loss vs Metal)			.408	-.327	.962	.969	.631	-.039	.217	.748

Beaufort or Newport in the same time period. The source of these inputs remains a mystery; the "suspected" broken thermometers, fungicides, slimicides, pharmaceuticals and cosmetics are simply guesses.

Table 40. Temporal variation of mercury in the influent of sewage treatment plants (ppb wet weight)

<u>Morehead City</u>		<u>Beaufort</u>		<u>New Bern</u>		<u>Newport</u>	
5/72	3.3	5/72	1.7 ± .1	5/72	3.3	5/72	2.7
5/72	2.5	8/75	.7 ± .4	6/72	2.5	7/73	1.4 ± .1
7/73	.7 ± <.05	1/76	1.0 ± .3	7/72	1.4	4/74	1.4 ± .4
4/74	1.4 ± .4	4/76	.8 ± .2	7/73	.5 ± .1	8/75	.8 ± .2
5/74	1.1 ± .5			4/74	3.3 ± .7	1/76	.8 ± .5
8/75	10.0 ± .5					4/76	1.7 ± .1
1/76	10.2 ± 1.1						
4/76	11.0 ± 1.0						

Samples of sewage were taken at both the Morehead City and New Bern sewage plants throughout the treatment process. Table 41 indicates that a considerable amount of mercury is removed in the secondary clarifier and trickling filter.

Table 41. Mercury concentrations (ppb) at the various stages of treatment throughout the Morehead City and New Bern sewage treatment plants. Two sets of data were obtained for each plant.

<u>Morehead City</u>		<u>New Bern</u>		
3.3	2.5	1.4	2.5	influent
2.8	2.4	x	2.4	after primary treatment
2.2	1.9	.9	1.8	after secondary clarifier
1.6	.9	.4	.9	after trickling filter
.6	.5	.4	.4	after chlorine contact chamber (effluent)

It has been suggested that a possible source of mercury in sewage treatment plants per se could be faulty mercury seals in the trickling filter stage of the sewage treatment. North Carolina passed a law making this kind of seal illegal after July 30, 1972, and the Morehead City sewage plant complied with the law in August of 1972 by removing the seal. Mercury concentrations in the effluent after removal (Table 42) were the same as the values before the seals were removed.

Table 42. Temporal variation of mercury concentrations in Morehead City sewage treatment plant effluent (ppb) before and after removal of the mercury seal

2/72	.5 ± .2	1/73	.5 ± <.05	5/74	.5 ± .2
3/72	.4 ± <.05	6/73	.7 ± <.05	7/74	.5 ± <.05
4/72	.4 ± .2	7/73	.4 ± .1	1/75	.4 ± .1
5/72	.9 ± .4	12/73	.2 ± <.05	6/75	.3 ± <.05
Seal Removed		3/74	.5 ± .1	8/75	.7 ± .1
8/72	.5 ± .2	4/74	.5 ± <.05	1/76	.6 ± .2
11/72	.4 ± .2	4/74	.3 ± .1	4/76	.5 ± <.05
12/72	.6 ± <.05				

Samples were taken to investigate the temporal fluctuations (Table 43 and Figure 56). Periods of high flow with high mercury concentrations alternate with periods of low flow with low mercury concentrations (Figure 56). High flow is indicated by high particulate load (Figure 56). Two effluent samples having  $0.50 \pm 0.10$  ppb and  $0.55 \pm <0.05$  ppb mercury were filtered. These samples contained  $0.16 \pm <0.05$  ppb and  $0.17 \pm <0.05$  ppb mercury, respectively, in the filtrate indicating that a majority of the mercury is in the suspended particulate fraction.

Table 43. Mercury concentrations (ppb) in effluent of the Morehead City sewage treatment plant taken during a 24-hr sampling period

1300 =	.8 ± .1	0200 =	.6 ± .1
1330 =	.9 ± .1	0400 =	.5 ± .1
1400 =	1.0 ± .2	0600 =	.3 ± .1
1600 =	.7 ± .2	0630 =	.4 ± .1
1800 =	.7 ± .1	0700 =	.5 ± .1
1830 =	.6 ± .1	0800 =	.5 ± .1
1900 =	.8 ± .1	0930 =	.9 ± .1
2000 =	.7 ± .1	1000 =	.7 ± .2
2200 =	.7 ± .1	1130 =	.8 ± .2
2400 =	.8 ± .1	1145 =	.8 ± .1
0030 =	.6 ± .1	1200 =	.9 ± .1
0100 =	.6 ± .1	Mean =	.8 ± .1

The formula to determine the amount of mercury discharged annually by a sewage treatment plant is:

$$\text{Flux (kg/yr)} = \text{concentration (ppb or } \mu\text{g/kg)} \times \text{flow (gal/day)} \times K$$

$$K = 1.38 \times 10^{-6} \text{ kg} \cdot \text{da/ppb} \cdot \text{gal} \cdot \text{yr}$$

Using this formula Table 44 gives the amount of mercury being discharged by various sewage treatment systems annually. Assuming a mercury load and using the flux formula, the total world annual mercury flux from human sewage can

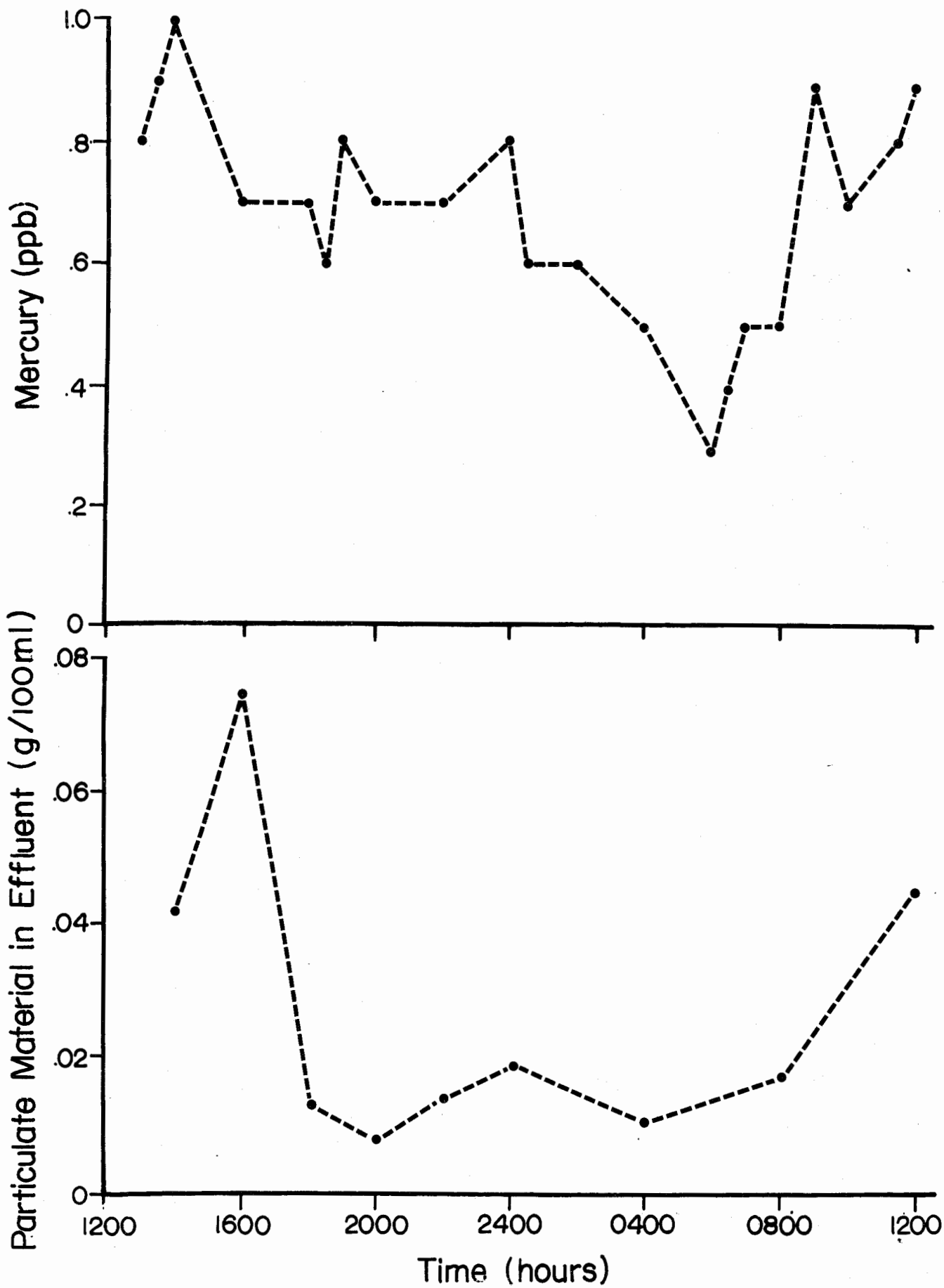


Fig. 56. Diel mercury and particulate pattern in Morehead City effluent. Numerical values for these points are found in Table 43.

Table 44. Kilograms of mercury dumped annually based on mercury in effluent and average daily flow rate of various sewage treatment plants

	Mean Mercury in Effluent (ppb)	Mean Daily Flow (gal. x 10 <sup>6</sup> )	Mercury Dumped Annually (kg)
North Carolina			
Beaufort	.1	1.9	.26
Durham (Sandy Creek)	.1	1.9	.26
Greensboro	.6	40.0	33.0
Morehead City	.5	4.6	3.2
New Bern	.4	10.0	5.5
Newport	.1	.4	<.1
West Carteret High School	.3	<.1 <sup>1</sup>	<.1
Winston-Salem	.5	91.0	62.8
Atlanta, Georgia			
Atlanta, Georgia	1.6	152.0	336.0
Corvallis, Oregon	.3	27.0	11.1
Palm Beach Gardens, Florida	.3	3.0	1.2
South Charleston, West Virginia	.3	42.0	14.0
Richmond, Virginia	1.0	190.0	262.0
Toledo, Oregon	.1	2.7	.37

<sup>1</sup>flow rate 5 days a week during the school year

be estimated. Given a population of  $3.0 \times 10^9$  people and 100 gal. of sewage effluent per person per day (Clark and Viessman, 1965) and 0.5 ppb mercury per liter of effluent, the flux would be  $2.1 \times 10^2$  metric tons. This figure compares to an annual industrial flux of  $3 \times 10^3$  and natural geological degassing rate of  $7 \times 10^3$  metric tons annually. This flux of mercury through sewage systems is an order of magnitude lower ( $2.1 \times 10^2$  vs  $3 \times 10^3$ ) than the industrial flux but is an important flux to consider in environmental management because it is all discharged to the environment.

Table 45 lists geographic variations in mercury concentrations in sewage treatment plant effluents. Cities with larger populations and industrial influences did not have higher mercury loads in effluents. Newport, Oregon, with the highest value reported (2.1 ppb), is a considerably smaller city than Atlanta, Georgia (1.6 ppb) or Richmond, Virginia (1.0 ppb). There is no major industry in Newport, Oregon. Oneonta, New York (<0.05 ppb) and East Jefferson, Louisiana (<0.05 ppb) are both areas of high industrial activity and large populations but have low mercury effluent.

Table 46 indicates that there has been a significant increase in the amount of mercury removed by the Morehead City sewage treatment plant and partitioned into sludge (1972 mean of 4.1 ppm vs 1976 mean of 18.9 ppm). Beaufort had a mean of 2.7 ppm in 1972 while in 1976 it still had 2.3 ppm. This difference may be due to a new source of mercury in Morehead City or to the improvements at the Morehead City plant, resulting in a cleaner effluent which disperses more metal to the sludge phase.

Samples from various stages of sludge processing at the Morehead City sewage treatment plant (Table 48) show that the digestion process results in a net increase in metal concentration.

Table 48. Concentrations of metals (ppm dry weight) at three stages of sludge processing in the Morehead City sewage treatment plant

	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Pb</u>	<u>Zn</u>
Undigested	6.59 ± 1.34	3.1	11	>150	70	>350
Digested	16.60 ± 2.10	9.1	25	>150	x	>350
Supernatant	14.33 ± 1.36	>5.1	x	>150	>40	>350

Van Loon *et al.* (1973) reported heavy metal inhomogeneity in sludge samples and concluded that grab samples were not representative of the entire sludge. Weaver *et al.* (1974) also noted a variation of not more than 20% in a given sludge bed and suggested combined grab samples would be more representative. The homogeneity of heavy metals in two local sludges was determined (Table 49). Beaufort had the highest inhomogeneity in zinc content (61%) and the lowest in copper (7%). The remaining metal variations were all less than 20%. The highest variability in the Newport sludge was found in mercury (27%), the remaining all being lower than 20% with copper (4%) again being the lowest.

Table 45. Geographic variations in mercury concentrations (ppb) of sewage treatment plant effluent

<u>Location</u>	<u>Effluent</u>	<u>Date</u>	<u>Time</u>
New Haven, Connecticut	.6 ± .1	9-10-72	x
Palm Beach, Florida	.3 ± <.05	5-24-73	1000
Atlanta, Georgia	1.6 ± .5	3-3-72	1200
East Jefferson, Louisiana	<.05	1-7-73	x
Cherry Point, North Carolina	.2 ± <.05	6-6 -72	x
Durham, N. C. (Sandy Creek)	.1 ± .1	3-10-73	1100
Greensboro, North Carolina	.7 ± .2	1-23-73	1600
Wilmington, North Carolina	.4 ± .1	2-7-74	1200
Winston-Salem, North Carolina	.5 ± .1	10-3-72	1600
Wrightsville Beach, N. C.	.5 ± .1	5-27-74	1130
Oneonta, New York	<.05	9-3-72	x
Corvallis, Oregon	.3 ± <.005	6-13-73	1700
Cottage Grove, Oregon	.3 ± .1	6-13-73	1400
Newport, Oregon	2.1 ± .7	6-12-73	1300
Toledo, Oregon	.1 ± .2	6-12-73	1100
Brownsville, Virginia	.2 ± .1	5-15-73	x
Richmond, Virginia	1.0 ± .3	5-29-72	1330
South Charleston, West Virginia	.3 ± .1	12-30-72	1100
Havelock, North Carolina	.3 ± <.05	7-6-73	1100

Table 46. Chronological listing of metal concentrations in sludge (ppm dry weight) in Morehead City and Beaufort sludges

<u>Location</u>	<u>Date</u>	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe</u> <u>X1000</u>	<u>Pb</u>	<u>Zn</u>
Morehead City, NC	May 72	4.1 ± 1.1						
	Aug 72	1.1 ± .1						
	Jun 73	4.68 ± .61	9				440	
	Aug 74	8.6 ± 1.0						
	Jan 75	16.6 ± 1.3	9	25	>150	x	x	
	Jun 75	7.6 ± 1.7	12	10	676	2.1	50	>300
	Jan 76	18.9 ± .7						
Beaufort, NC	Aug 72	2.7 ± .6						
	Jun 73	.9 ± .3						
	Aug 74	2.7 ± .1						
	Jun 75	.5 ± .1	13	3	268	x	184	55
	Jan 76	2.3 ± .4	10	36	526	x	221	1400

Table 47. Heavy metal concentrations (ppb) found in sludge from various cities

<u>Reference</u>	<u>Location</u>	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Fe</u> <u>X1000</u>	<u>Pb</u>	<u>Zn</u>
1. Van Loon, J. C., <u>et al.</u> , 1973	Toronto	15	44	400	120	7.8	140	600
2. _____, 1974	Sweden	6	10	125	100	X	150	200
3. Weaver, J. N., <u>et al.</u> , 1974	North Carolina	19	X	1189	X	17.0	X	X
4. Oliver, B. G., and Cosgrove, E. G., 1974	Ontario	.7	2.1	127	159	.5	90	1205
5. Bloomfield, C. and Pruden, G., 1975	Great Britain	X	64	3300	X	X	1200	6500
6. Boswell, F. C., 1975	Atlanta	X	165	1754	636	X	2748	11812
7. Lagerwerff, J. V., <u>et al.</u> , 1976	Baltimore Washington, D. C.	X X	15 12	X X	1050 648	X X	600 420	1914 1619
8. Cunningham, J. D., <u>et al.</u> , 1975	Wisconsin	9	194	9200	1200	32.9	940	5100
9. This study	Morehead City, NC	8	10	17	670	2.1	183	>400
	Beaufort, NC	2	11	20	400	X	190	X
	Cherry Point, NC	4	78	X	254	X	390	500
	East Carteret HS, NC	.9	7	10	167	2.5	155	>500
	New Bern, NC	8	10	X	>200	X	400	>400
	Newport, NC	3	12	23	198	1.6	X	1250
	West Carteret HS, NC	.6	62	11	>200	5.3	45	>500
	Wilmington, NC	12	X	X	X	X	X	X
	Athens, GA	12	5	61	250	>3.0	350	>1000
	Richmond, VA	2	12	X	760	X	124	1800
	Tunis, Tunisia, Africa	2	32		2300			
	Toledo, OR	11						
	Cottage Grove, OR	3						

Table 49. Heavy metal concentrations (ppm dry weight) in repeated samplings of the Beaufort and Newport sludge beds

	<u>% Loss</u>	<u>Hg</u>	<u>Cd</u>	<u>Cr</u>	<u>Cu</u>	<u>Pb</u>	<u>Zn</u> <u>X1000</u>
A Beaufort	55	2.31 ± .44	10	36	526	221	1.4
B Beaufort	57	2.77 ± .36	9	23	521	267	1.2
C Beaufort	51	2.69 ± .59	8	32	448	260	1.2
D Beaufort	50	3.11 ± .18	8	33	500	210	1.3
Mean	53	2.72	9	31	499	240	1.3
St.Dev.	3.3	.46	.7	6	36	28	.8
Co.Var.	6	17	8	19	7	12	61
A Newport		1.55 ± .24	10	22	213	107	1.3
B Newport		.91 ± .11	8	23	203	99	1.3
C Newport		1.15 ± .14	9	21	200	103	1.5
D Newport		1.71 ± .10	11	26	219	121	1.8
Mean		1.33	9	23	209	107	1.5
St.Dev.		.36	1.2	2	9	10	.2
Co.Var.		27	13	9	4	9	15

## Feeding Experiments

Previous studies on the uptake and retention of mercury in fish have shown that absorption of mercury from the water by the gill is a very important source of this metal to the fish (Hannez, 1968). There has also been much work dealing with the methylation of mercury within a fish (Jensen and Jernelov, 1969; Johnels, 1967). In the study reported here a food source naturally high in metals was fed to shrimp (Penaeus sp.) and pinfish (Lagodon rhomboides). The concentration of selenium, which detoxifies or makes mercury inaccessible to an organism, was also measured (Ganther et al., 1972). The most important aspect of these experiments was that these were natural mercury food sources and not the classic radioactive or mercury chloride spiked diets.

The first feeding experiment involved shrimp (Penaeus sp.) fed on four different diets. Four groups (20/group) of adult shrimp were fed the following diets of 2 g per day per shrimp:

- Group 1 - squid (0.2 ppm Hg)
- Group 2 - bluefish (0.8 ppm Hg)
- Group 3 - blue marlin (9.8 ppm Hg)
- Group 4 - sludge/oysters (0.7 ppm Hg)

The mantle and tentacles were the only parts of the squid used in experiments. Only the axial muscle tissue of the bluefish and blue marlin were used as food and the sludge/oyster diet was made up by blending (by weight) about 5 parts powdered sludge (dry) to one part oyster (wet). This produced a solid material which was frozen and then fed to the shrimp. The groups of shrimp were held in separate tanks of running seawater for 30 days at a mean temperature of 26°C. During the experiment deaths occurred in all four tanks. Five shrimp died in the squid-fed group, twelve in the bluefish-fed group, twenty in the marlin-fed group, and ten in the sludge/oyster-fed group. Prior to the deaths no visible behavioral abnormalities, such as impaired or erratic swimming, were observed in any of the four groups.

Table 50 shows the results of mercury analysis of five shrimp from each group taken on day 30. In the case of the marlin-fed group the shrimp were the last five which were alive on day 29 but dead by day 30. The deaths of all shrimp in the marlin-fed group ended the experiment. Shrimp that died prior to day 30 were consumed by the other shrimp in the tank and thus no data could be obtained from shrimp which died during the experiment.

The squid, bluefish, and blue marlin-fed groups increased in mercury concentration by 5, 9, and 27 times, respectively, over the initial group. The sludge/oyster-fed group increased by only 1.8 times. In addition to having the greatest mercury concentration the blue marlin-fed group also had the greatest death rate.

Groups fed squid, blue marlin and, to the greatest extent, sludge/oyster lost weight. The bluefish diet was the only group that gained weight. The

weight loss by the sludge/oyster group is evidence that the diet was not being assimilated by the shrimp.

The sludge/oyster diet (0.7 ppm Hg) and the bluefish diet (0.8 ppm Hg) were similar in mercury concentrations; however, the shrimp in the two groups had very different accumulations of mercury. One explanation for the difference might be that the mercury in the bluefish was organic. In marine fish organic mercury accumulation rate is higher and its excretion rate slower (Johnels et al., 1967; Karnps et al., 1972) than inorganic mercury.

The second feeding experiment involved pinfish (Lagodon rhomboides) which were fed adequate and deficient diets with both high and low mercury concentrations. Four groups of ten pinfish per group were held in running seawater tanks and maintained on the following diets:

- Group 1 - deficient - low Hg (0.3 g/day/fish of oysters with 0.1 ppm Hg, wet weight)
- Group 2 - adequate - low Hg (3.6 g/day/fish of oysters with 0.1 ppm Hg, wet weight)
- Group 3 - deficient - high Hg (0.3 g/day/fish of bluefish with 0.8 ppm Hg, wet weight)
- Group 4 - adequate - high Hg (3.6 g/day/fish of bluefish with 0.8 ppm Hg, wet weight)

The estimate of what constituted an "adequate" diet was based upon data from Peters and Kjelson (1975) which indicated 8% per day of pinfish body weight as the maximum rate of food consumption. The average pinfish weight was 40 g, thus the "adequate" diet was calculated to be 3.6 g/day/fish. The underfed groups (deficient diet) were fed just enough to survive. Pinfish were weighed on day 0, day 30, and day 60 (termination) and frozen for subsequent analyses. A fish that increased in weight should have had its food supply increased proportionally to keep the food to body mass constant throughout the experiment. However, in these experiments the diets were kept constant. The mean temperature throughout the experiment was 27°C and feeding lasted 60 days. Several water samples were analyzed for mercury during the experiment and undetectable (<0.05 ppb) mercury levels recorded for all samples.

The fish fed the adequate and deficient diets of oysters did not accumulate mercury in either axial muscle, liver or brain when compared to fish from the initial group sacrificed on day 0 (Table 51 and Figure 57). There is no statistical difference among these three groups when comparing data for muscle, liver, and brain between groups. On the other hand, fish fed the adequate diet of bluefish had a mean mercury concentration of 1.42 ppm in axial muscle, 1.42 ppm in liver, and 0.95 ppm in the brain at the end of 60 days. These values are considerably greater than those from the initial group. The fish fed the deficient diet of bluefish had a mean mercury concentration of 0.31 ppm in the axial muscle tissue, a value considerably lower than that of the adequate group (1.42 ppm). The mean concentration in the liver of the deficient group was 1.24 ppm, similar to the liver concentration of the 1.42 ppm found in the adequate group, suggesting that mercury concentrations in the liver are dependent upon concentrations in the food and not upon absolute

Table 50. Mercury accumulation in Penaeus sp. muscle tissue after a 30-day feeding experiment of four diets

	<u>Weight (g)</u>	<u>µgHg/g (wet weight)</u>
Initial		
	6.4	.05 ± .03
	7.9	.06 ± .04
	8.7	.15 ± .04
	11.1	.06 ± .04
	11.6	.13 ± .01
	mean 9.14	mean .09
Group #1	Squid diet - 2 grams/day containing 0.2 ppm Hg	
	5.8	.21 ± .01
	7.5	.23 ± .01
	8.1	.65 ± .13
	9.4	.13 ± .06
	9.5	.90 ± .15
	mean 8.06      1.08g loss	mean .42
Group #2	Bluefish diet - 2 grams/day containing 0.8 ppm Hg	
	10.4	.57 ± .20
	10.8	1.51 ± .11
	11.0	.87 ± .03
	11.2	1.05 ± .08
	15.2	.28 ± .05
	mean 11.72      2.58g gain	mean .86
Group #3	Blue marlin diet - 2 grams/day containing 9.8 ppm Hg	
	4.9	2.35 ± .11
	6.0	1.88 ± .47
	6.0	2.53 ± .31
	9.9	2.36 ± .24
	10.5	3.02 ± .51
	mean 7.46      1.68g loss	mean 2.43
Group #4	Sludge/Oyster diet - 2 grams/day containing 0.7 ppm Hg	
	1.5	.11 ± .02
	2.1	.32
	3.7	.18 ± .01
	4.5	.13
	4.6	.20
	6.9	.08 ± .01
	mean 3.88      5.26g loss	mean .17

Table 51. Mercury concentrations (ppm) in Lagodon rhomboides axial muscle, liver and brain tissue after 60-day experiment using adequate and deficient diets with both high and low mercury concentrations

<u>WEIGHT(g)</u>	<u>MUSCLE</u>	<u>LIVER</u>	<u>BRAIN</u>	<u>WEIGHT(g)</u>	<u>MUSCLE</u>	<u>LIVER</u>	<u>BRAIN</u>
Day 0 ( $\mu\text{gHg/g}$ wet weight)							
19.5	.11 $\pm$ .03	.08	.13	44.1	.12 $\pm$ .02	.21	.03
24.0	.10 $\pm$ .01	.23	.07	44.5	.06 $\pm$ .01	1.11	.32
33.4	.05 $\pm$ .01	.11	.17	47.9	.11 $\pm$ .01	.07	.005
33.5	.11 $\pm$ .02	.17	.14	48.1	.09 $\pm$ .02	.17	.21
37.5	.06 $\pm$ .01	x	.40	66.3	.19 $\pm$ .01	.26 $\pm$ .04	.11
Mean - Muscle .10 $\pm$ .04, liver .27 $\pm$ .30, brain .16 $\pm$ .12							
Group #1 Adequate diet (3.6 grams/day/fish) of oysters (.07 $\pm$ .03 $\mu\text{gHg/g}$ )							
32.5	.12 $\pm$ .01	.11	.005	66.7	.17 $\pm$ .04	.15 $\pm$ .01	.06
52.3	.11 $\pm$ .01	x	.17	80.1	.04 $\pm$ .06	.04 $\pm$ .06	.02
57.9	.15 $\pm$ .03	.24 $\pm$ .02	.24	80.2	.13 $\pm$ .02	.07 $\pm$ .02	.04
58.9	.02 $\pm$ .03	x	.04	81.0	.10 $\pm$ .01	.17 $\pm$ .01	.16
65.1	.09 $\pm$ .01	.11 $\pm$ .01	.04	82.3	.14 $\pm$ .01	.23 $\pm$ .05	.14
Mean - Muscle .11 $\pm$ .05, liver .14 $\pm$ .08, brain .09 $\pm$ .08							
Group #2 Deficient diet (.3 grams/day/fish) of oysters (.07 $\pm$ .03 $\mu\text{gHg/g}$ )							
11.0	.04 $\pm$ .01	.80	.005	34.2	.11 $\pm$ .03	.005	.15
13.6	.07 $\pm$ .10	.005	.005	37.6	.07 $\pm$ .01	.04	.005
16.5	.04 $\pm$ .01	.005	.15	40.6	.15 $\pm$ .04	.01	.19
22.6	.05 $\pm$ .05	.005	.005	42.2	.09 $\pm$ .03	.03	.005
29.0	.03 $\pm$ .04	.25	.005	43.0	.23 $\pm$ .08	.23	.06
Mean - Muscle .09 $\pm$ .06, liver .14 $\pm$ .25, brain .06 $\pm$ .08							
Group #3 Adequate Diet (3.6 grams/day/fish) of bluefish (.76 $\pm$ .17 $\mu\text{gHg/g}$ )							
50.4	1.69 $\pm$ .13	.79	1.40	59.7	1.83 $\pm$ .10	.64 $\pm$ .15	.40
50.6	1.98 $\pm$ .22	1.69	x	69.3	.96 $\pm$ .18	2.61 $\pm$ .53	1.65
55.2	.99 $\pm$ .02	1.63 $\pm$ .08	.89	69.6	1.59 $\pm$ .25	.42 $\pm$ .36	.23
57.6	1.40 $\pm$ .34	x	1.29	70.9	1.87 $\pm$ .47	1.28 $\pm$ .04	.35
59.6	.80 $\pm$ .05	2.09 $\pm$ .42	1.49	104.8	1.05 $\pm$ .31	1.59 $\pm$ .03	.88
Mean - Muscle 1.42 $\pm$ .44, liver 1.42 $\pm$ .71, brain .95 $\pm$ .53							
Group #4 Deficient diet (.3 grams/day/fish) of bluefish (.76 $\pm$ .17 $\mu\text{gHg/g}$ )							
16.0	.38 $\pm$ .02	2.9	.79	22.9	.37 $\pm$ .04	.44	.19
16.4	.27 $\pm$ .04	.95	.50	23.7	.44 $\pm$ .02	2.17	.99
20.0	.26 $\pm$ .107	.90	.34	27.7	.34 $\pm$ .10	1.23	.94
21.1	.17 $\pm$ .03	1.5	.40	43.5	.09 $\pm$ .01	.66	x
21.4	.29 $\pm$ .14	1.3	.09	44.8	.30 $\pm$ .09	.48 $\pm$ .22	.08
22.6	.26 $\pm$ .15	1.7	.40	49.9	.38 $\pm$ .08	.69	.17
Mean - Muscle .31 $\pm$ .07, liver 1.24 $\pm$ .74, brain .44 $\pm$ .33							

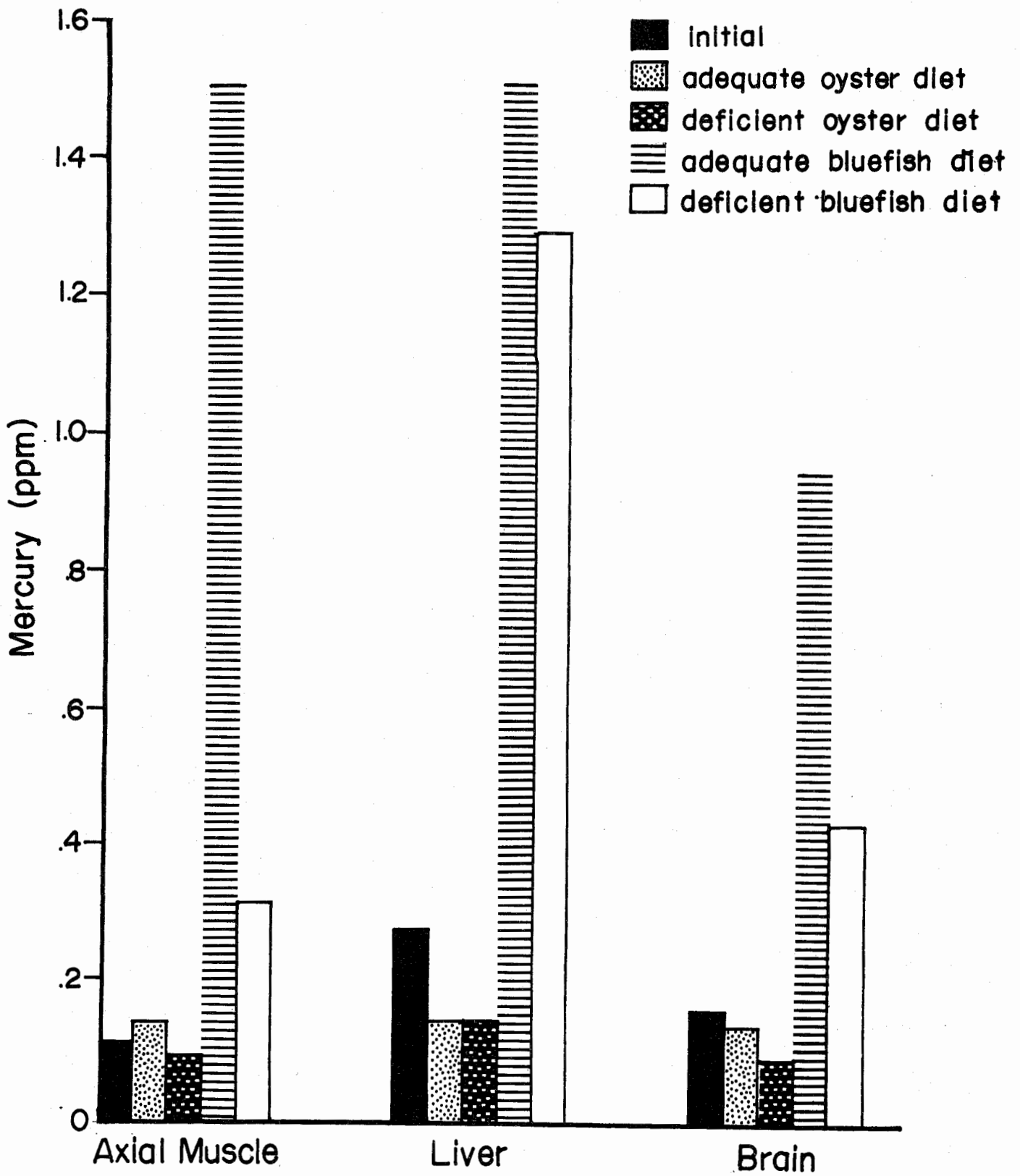


Fig. 57. Accumulation of mercury by pinfish fed various mercury diets

amounts of mercury consumed per unit time. The concentration of mercury in the brains of the fish fed a deficient diet of bluefish (0.44 ppm) is less than the group fed an adequate diet of bluefish (0.95 ppm). The range of mercury values of the brains of these two groups as well as those of the initial group indicates the large variability in individual pinfish of the mercury entering and/or remaining in brain tissue.

No deaths occurred in any of the four groups and the diet with the maximum amount of mercury (3.6 g of 0.8 ppm mercury per day per fish) was not enough of a stress to visibly affect the behavior of the pinfish.

Pinfish were weighed on day 0, day 30, and day 60. As seen in Table 52, groups 1 and 3 lost weight while groups 2 and 4 gained weight.

Table 52. Mean weights (g) of Lagodon rhomboides during a 60-day feeding experiment

	<u>Day 0</u>	<u>Day 30</u>	<u>Day 60</u>	<u>Change</u>
Group 1	38.1	34.0	28.5	25% decrease
Group 2	42.4	53.8	67.8	25% increase
Group 3	33.8	37.2	27.4	19% decrease
Group 4	43.7	36.5	65.2	29% increase

In a third feeding experiment which lasted 160 days 25 juvenile pinfish (Lagodon rhomboides) with an average weight of 20 g were held in running (3.5 liters per minute) seawater tanks at ambient temperatures (19° to 26°C). Each day the fish were fed 4 g wet weight of axial muscle tissue of blue marlin (Makaria nigricans). The blue marlin tissue had the following metal concentrations: 15 ppm total mercury (10 ppm organic), 0.04 ppm cadmium, 0.25 ppm chromium, 0.2 ppm copper, 2 ppm iron, 0.1 ppm lead, 0.1 ppm manganese, and 3 ppm zinc. Another group of 25 fish was fed oysters (4 g/fish/day) which had metal concentrations of 0.1 ppm mercury, 2 ppm cadmium, 0.21 ppm chromium, 120 ppm copper, 46 ppm iron, 0.1 ppm lead, 6 ppm manganese, and 400 ppm zinc. The concentrations of chromium and lead were about the same in the two diets. The concentration of mercury was much greater in the blue marlin; however, the concentration of other metals was much greater in the oyster tissue.

A group of 10 pinfish were killed at the start of the experiment and analyzed to provide data on initial concentrations of metals in their tissues.

All fish were dissected for axial muscle, liver, brain, heart, and kidney. Some fish were sampled for skin and lateral muscle tissue. Mercury concentrations for axial muscle and liver are reported with standard deviations based upon three replicate analyses of the tissue. Due to the small size of the tissues all other data are from only one analysis. Axial muscle tissue was also analyzed for other metals. Samples that were analyzed for selenium were low temperature ashed with Technico 300-1 Plasma Excitor and analyzed following

the procedure of Inhat and Weslerby (1974) with the graphite furnace of the Varian atomic absorption spectrophotometer.

The appearance and behavior of pinfish fed the two diets was very different. One of the first differences noticed in the marlin-fed group was the appearance of a blood embolism at the base of the dorsal and caudal fins. Embolisms first appeared about day 10 and remained in all marlin-fed fish until the end of the experiments. The marlin-fed fish were very irritable when compared to oyster-fed fish and they exhibited frantic and erratic swimming behavior when disturbed. Deaths occurred in the marlin-fed tank after any stress. From time to time the running seawater system would malfunction or a missed feeding or overfeeding occurred and in these instances there would be mortality in the marlin-fed tank. In the process of dying the symptoms were always the same. The red blood embolisms became more apparent. The fish's swimming speed became much slower. Eventually the fish was unable to swim upright and would swim at a 45° angle. The fish appeared to have limited use of the pectoral fins. Eventually all swimming ceased and the fish would float to the surface. This whole process would take only a few hours. There were no deaths in the oyster-fed pinfish group.

During the experiment the oyster-fed group gained an average of 10 g in body weight. The pinfish which survived in the marlin-fed group gained an average of 26 g weight, 2.5 times that of the oyster-fed animals. These data suggest that the nutritional value of marlin was greater than that of the oyster even though the much greater mercury content of the marlin produced pathological symptoms in that group of pinfish.

Mercury accumulations in the various tissues of the oyster-fed and marlin-fed pinfish are illustrated in Figures 58A-D and 59A and accumulations of the other metals in muscle tissue are given in Figures 59B-D and 60A-D. These graphs indicate that there was no accumulation of mercury in the tissues of the oyster-fed pinfish. In the marlin-fed pinfish mercury concentrations increased continuously during the course of the experiment. The variability in mercury concentrations also increased suggesting an extreme degree of individuality among the fish in rates of accumulation. Figure 61 is a graph of the regression lines for rates of mercury accumulation in the various tissues. These rates were greatest for liver, followed in order by kidney, brain, heart, and muscle. The correlation coefficients suggest that mercury concentrations were least variable in muscle tissue ( $r=0.941$ ) and most variable in the kidney ( $r=0.739$ ). Interpretation of these results would depend upon biochemical explanations which are beyond the scope of this report.

In the past the mercury concentrations in the gill tissues of fish have been analyzed and high values have been interpreted as an indication that fish were absorbing mercury from the water in their environment. The data in Table 53 suggest that diet alone can result in significant increases in mercury concentration in the gills of fish.

Table 53. Mercury concentrations (ppm wet weight) in the gill tissue of pinfish fed a high and low metal diet for 160 days

Oyster-fed			Marlin-fed		
Day	Wt	Hg (ppm)	Day	Wt	Hg (ppm)
120	39	.04 ± .04	119	57	3.00 ± .28
120	44	.03 ± .05	120	58	2.81 ± .09
160	37	.05 ± .03	160	63	2.04 ± .06
160	41	.01 ± <.005	160	67	2.04 ± .42
160	45	.01 ± <.005	160	71	2.18 ± .31

The concentrations of other metals found in the diets of pinfish are presented in Table 54. In comparison with the marlin diet the oyster diet was enriched in cadmium, copper, iron, manganese and zinc. Chromium and lead concentrations were the same for both diets. The concentrations of these metals in muscle tissue of pinfish (Figures 59B-D and 60A-D) indicate a complex pattern of metal uptake and loss which is dependent upon both metal concentration in the food and the type of food given. Individual fish were highly variable in concentrations of these metals and there were no strong correlations (positive or negative) of concentration with time.

Table 54. Heavy metal concentrations (ppm wet weight) found in the axial muscle of blue marlin and in oysters fed to pinfish

	Cd	Cr	Cu	Fe	Pb	Mn	Zn
Marlin	.04	.3	.2	2	.1	.1	3
Oyster	2	.2	120	46	.1	6	400

Recently, attention has focused on the relationship between mercury and selenium; Ganther *et al.* (1972) found that quail fed a diet containing both mercury and selenium (1:1 to 1:2 atomic ratio) were protected against mercury poisoning. The marlin tissue fed to the pinfish had a selenium concentration of 2.0 ppm. Selenium concentrations of 0.2, 0.3, and 0.3 ppm were found in the axial muscle tissue of pinfish before being fed the marlin. Concentrations of 0.3, 0.7 and 1.8 ppm were found in marlin-fed pinfish sacrificed on day 160. These data suggest a slight increase in selenium in pinfish muscle during the course of the experiment. Based upon observations of pathology and behavior there was no "selenium protection" even though the concentration of selenium in the diet of the pinfish was within 1:1 to 1:2 atomic ratios of mercury to selenium.

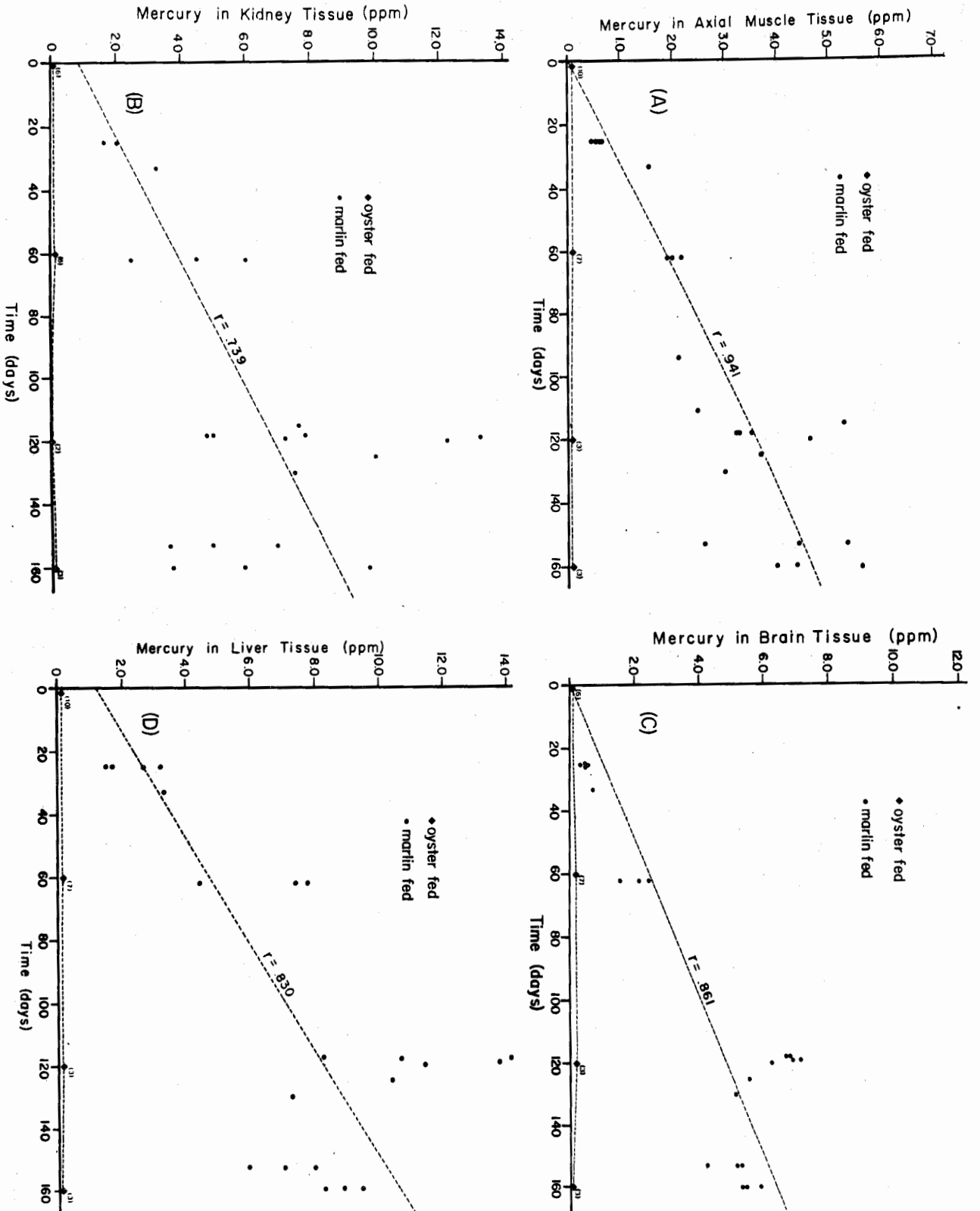


Fig. 58. Mercury accumulation by pinfish fed high and low metal diets

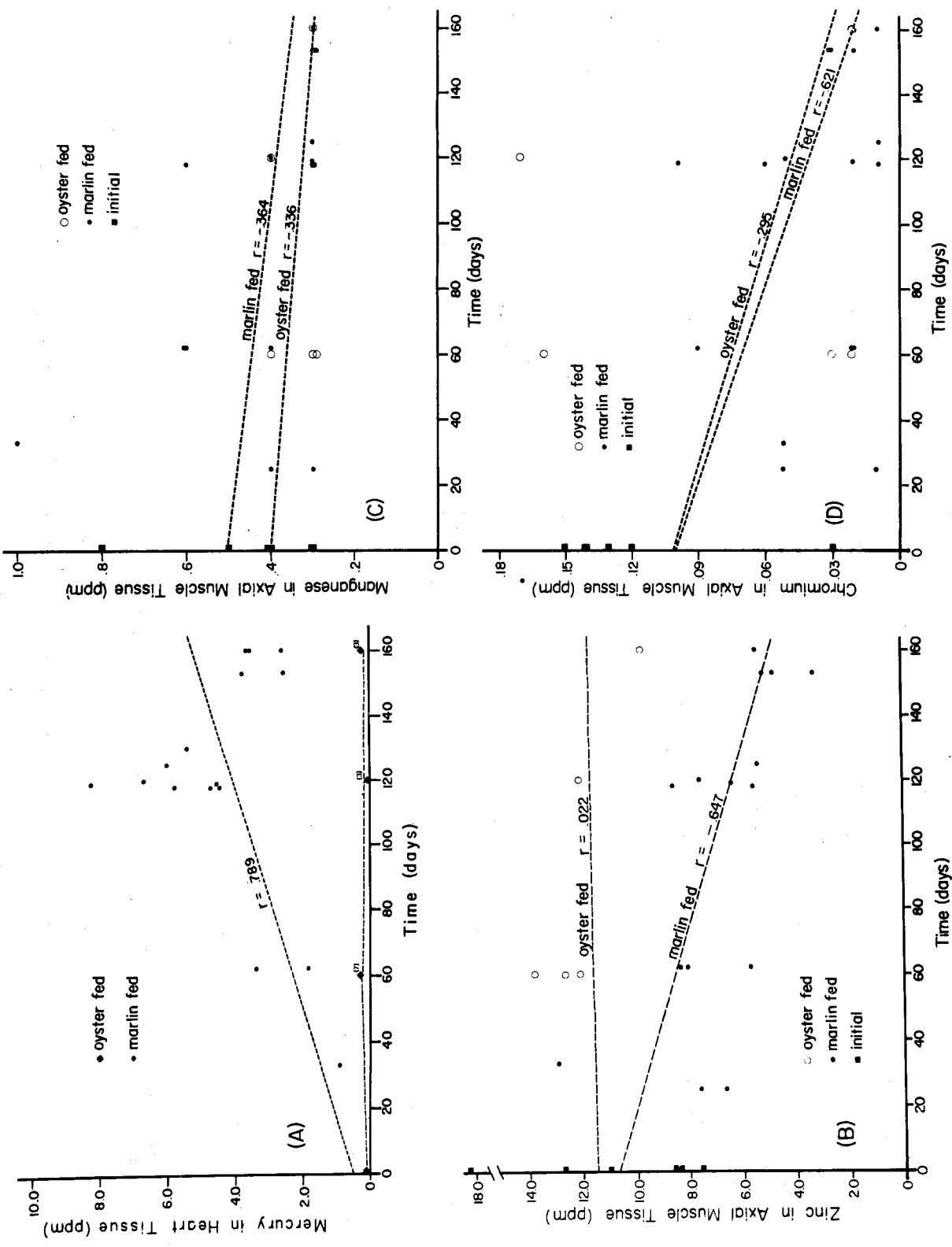


Fig. 59. Heavy metal accumulation by pinfish fed high and low metal diets

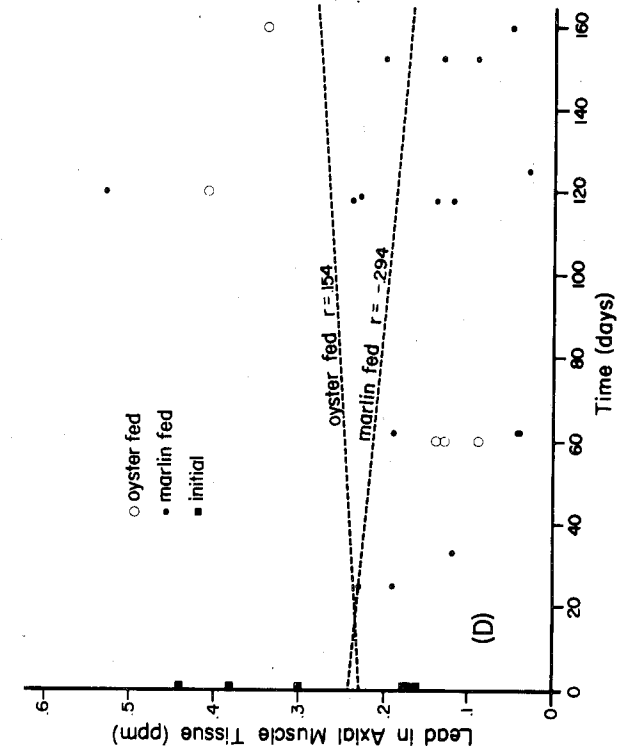
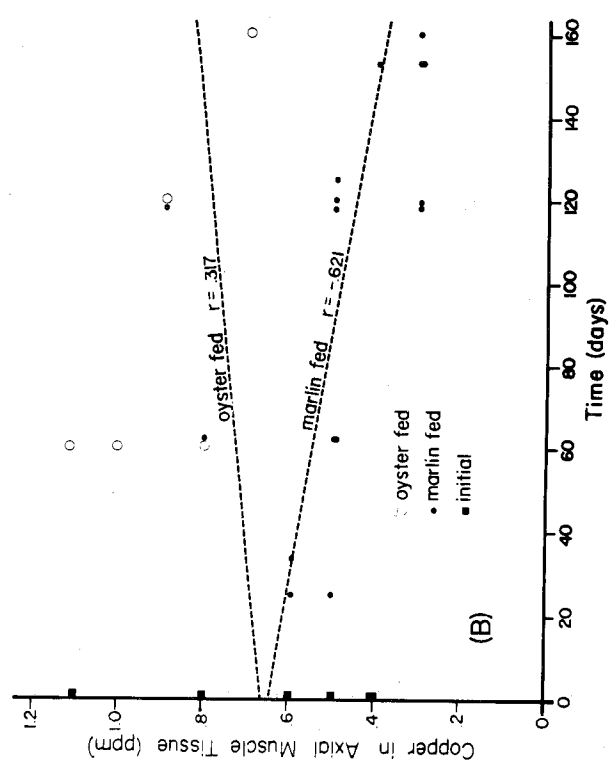
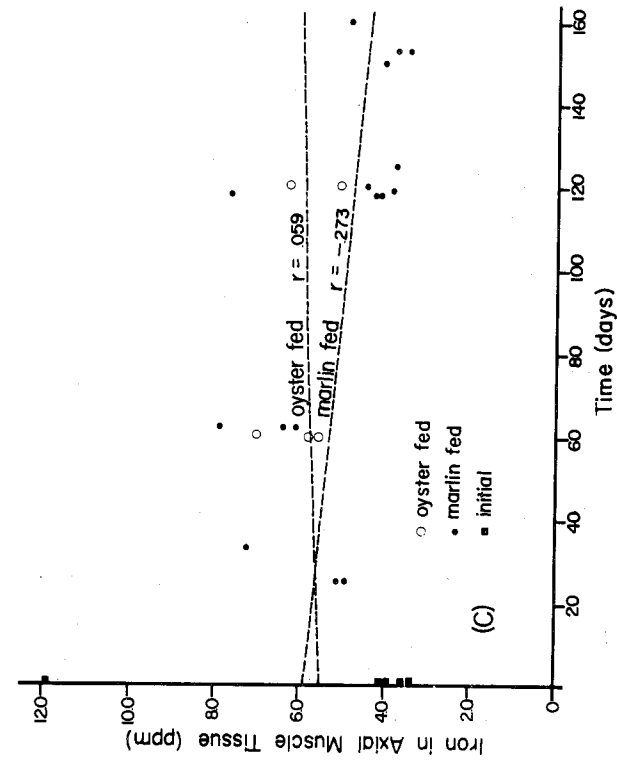
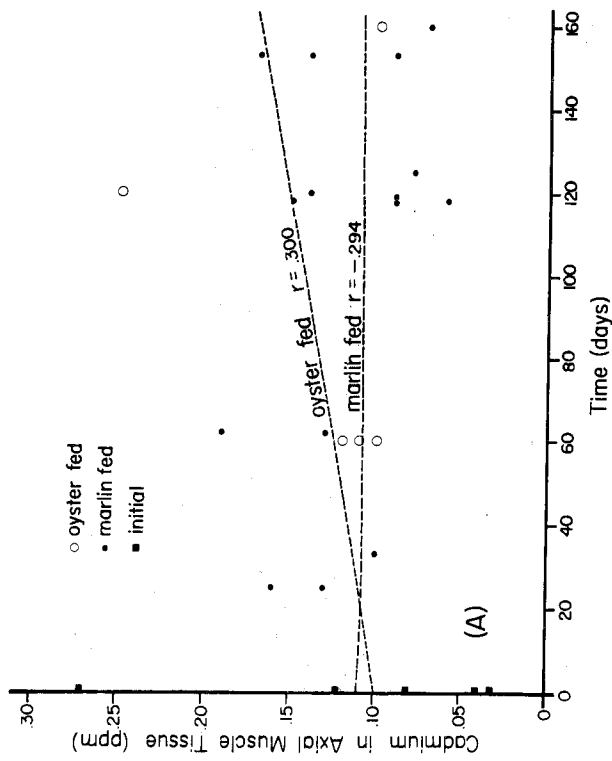


Fig. 60. Heavy metal accumulation by pinfish fed high and low metal diets

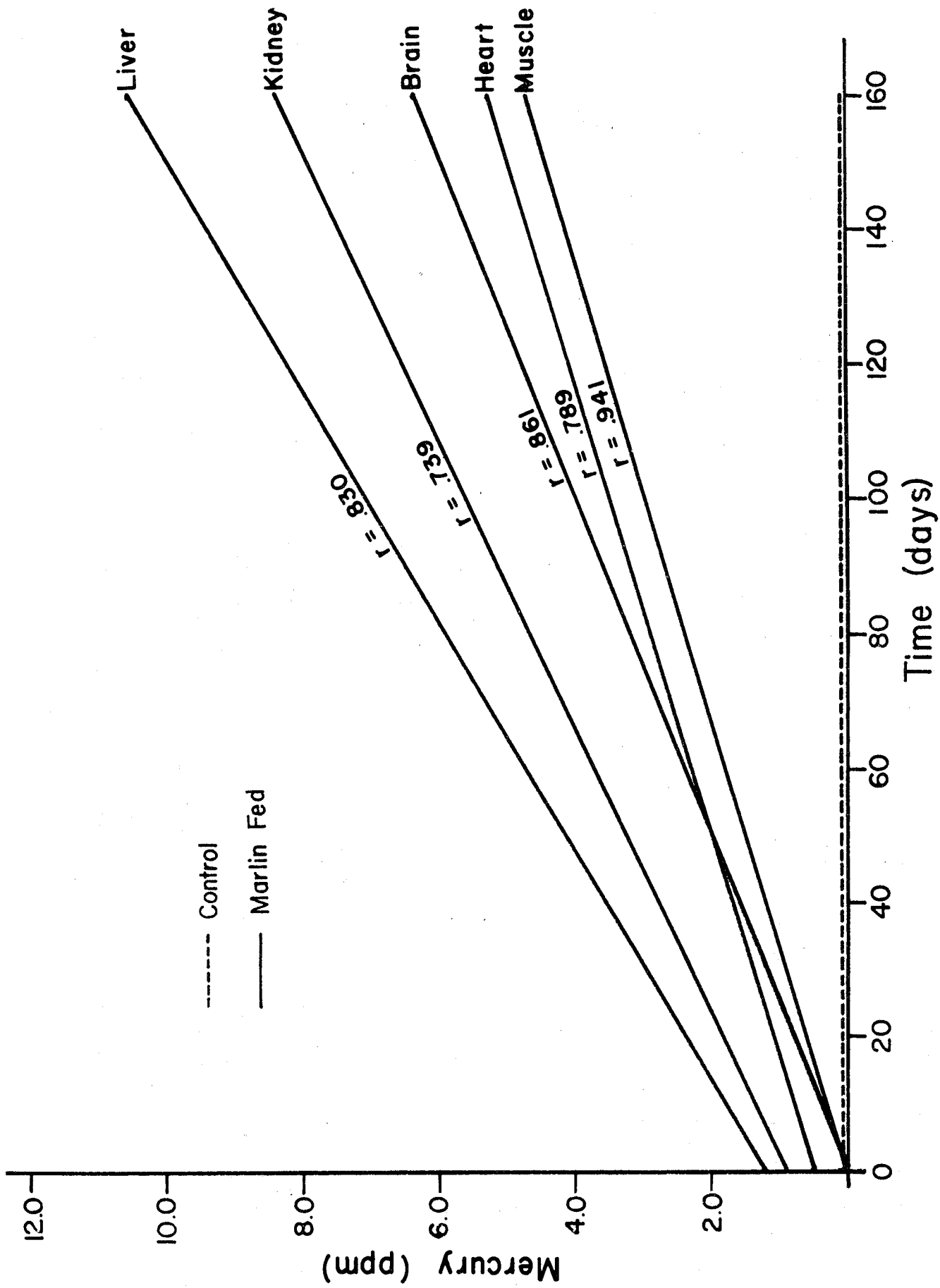


Fig. 61. Mercury accumulation in various organs of pinfish fed high and low metal diets

## Exposure Experiments

In the first experiment two groups of spat oysters (Crassostrea virginica) were held in trays 5 cm above two different substrates: the control sediment was organic-rich mud from a nearby Newport River marsh which had 0.1 ppm total mercury; the other sediment was the same marsh mud with domestic sewage sludge from the Morehead City sewage treatment plant added to yield a sediment of 8 ppm total mercury. The trays were placed in running seawater tanks for 150 days. A temperature of 20°C was maintained throughout the experiment. Table 55 lists the results of this 150-day exposure experiment.

Table 55. Mercury concentrations (ppm) in oysters held over sludge

	<u>Control Sediment</u>	<u>Sewage Sludge Enriched Sediment</u>
Day 0	.07 ± .03	.07 ± .03
Day 120	.05 ± .02	.05 ± .02
Day 150	.11 ± .05	.21 ± .06

During the 150-day experiment the average weight of the oysters dropped from 14 g to 2 g. The data suggest a slight accumulation of mercury. However, since the animals decreased in weight, it is probable that the increased concentration in the tissue was a result of lack of mercury excretion during weight loss.

A second experiment was designed to investigate possible mercury accumulation in animals living in direct contact with sewage sludge. The species used in the experiment were shrimp (Penaeus sp.), which feed by scavenging, scallops (Argopecten irradians), which filter feed, and mud snails (Ilyanassa obsoletus), which are browsers. Animals were placed in two tanks with running seawater. The bottom of one tank was covered with mud (0.1 ppm Hg) while the other bottom was covered with sludge (3.8 ppm Hg). After 90 days the animals were killed and their tissue was analyzed for mercury. The results of this experiment are given in Table 56. The snails were the only group of animals which had a significantly higher mercury concentration in sludge as compared to the control sediment, and the snails were the only species likely to ingest the sediment in the process of feeding. The other two species would have been exposed to any mercury being leached from the sediments; however, neither of the two would be likely to ingest large quantities of the sediment.

In a third exposure experiment freshwater carp (Cyrius carpio) were taken from the settling lagoons at the primary treatment plant of the Durham, North Carolina, Sandy Creek sewage treatment plant in January 1973. Carp browse over the organic rich sediments to derive their food. The Sandy Creek effluent in which the carp were swimming had a mercury concentration of 500 ppm; thus the situation was ideal to study increased metal uptake from sewage. The data on mercury concentrations in the carp are presented in Table 57 along with previously published values for comparison. The mean value of 0.17 ppm

in axial muscle is not high compared to published values listed. Concentrations in the liver (0.15 ppm), kidney (0.15 ppm) and gill (0.10 ppm) do not seem elevated either, although there are no published values for comparison.

In conclusion, these exposure studies along with the results of feeding experiments support the hypothesis that aquatic animals derive the mercury found in their tissue from their food and do not absorb mercury directly from water or sediments.

Table 56. Mercury concentrations (ppm) in animals held for 90 days in tanks of running seawater with two kinds of sediment

	<u>Control Sediment</u> (0.1 ppm Hg)	<u>Sludge Sediment</u> (3.8 ppm Hg)
Shrimp	.09 ± .01	.10 ± .03
	.09 ± .01	.13 ± .01
	.07 ± .02	.06 ± .01
	.03 ± <.005	.07 ± .03
	.05 ± .01	.07 ± .03
	.05 ± .02	.10 ± .03
	.04 ± .01	.04 ± .02
Mean	.06 ± .02	.08 ± .03
Scallops	.15 ± .04	.11 ± .05
	.13 ± .06	.14 ± .09
	.08 ± .02	.09 ± .06
	Mean	.12 ± .04
Snail	.35	1.09
	.36	.51
	.26	.25
	Mean	.32 ± .06

Table 57. Mercury (ppm) in carp (Cyrius carpio) taken from the Durham Sandy Creek sewage treatment plant holding lagoons, and published values (ppm wet weight)

<u>Weight</u> <u>(kg)</u>	<u>Axial</u>	<u>Liver</u>	<u>Kidney</u>	<u>Gill</u>
2.6	.15 ± .06	.06 ± .02	x	.23 ± .08
2.8	.14 ± .06	x	x	.05 ± .02
2.8	.11 ± .01	.05 ± .02	.10 ± .02	.06 ± .02
.1	.10 ± .04	x	.11 ± .01	.07 ± .02
.2	.10 ± .03	.16 ± .05	.15 ± .01	.06 ± .02
.7	.42 ± .01	.31 ± .12	.22 ± .03	.10 ± .02
Mean	.17	.15	.15	.10
St.Dev.	.12	.12	.05	.07
Co.Var.	73	83	38	72

Published values

<u>Axial</u>	<u>Location</u>	<u>Reference</u>
3.86-10.72	Ball Lake, Canada	Annett, C. S. <u>et al.</u> , 1975
.25	Lake Erie, Ohio	Aronson, J. L. <u>et al.</u> , 1976
.04-.22	Mississippi River	Hartung, R., 1974
.10-.43	South Dakota	Walter, C. M. <u>et al.</u> , 1973

## REFERENCES

- Annett, C. S., F. M. D'Itri, J. R. Ford and H. H. Prince, "Mercury in fish and waterfowl from Ball Lake, Ontario," Journal of Environmental Quality, 4(2):219-222 (1975).
- Applequist, M. D., A. Katz and K. K. Turekian, "Distribution of mercury in the sediments of New Haven (Conn.) Harbor," Environmental Science and Technology, 6(3):1123-1124 (1972).
- Aronson, J. L., M. Spiesman and A. K. Aronson, "Note on the distribution of mercury in fish species in three Ohio lakes," Environmental Bulletin, 10:1-7 (1976).
- Bascom, W., "How they do it in southern California," Marine Pollution Bulletin, 7(5):81-83 (1976).
- Bloomfield, C. and G. Pruden, "The effects of aerobic and anaerobic incubation on the extractabilities of heavy metals in digested sewage sludge," Environmental Pollution 8: 217-237 (1975).
- Boswell, F. C., "Municipal sewage sludge and selected element applications to soil: effect on soil and fescue," Journal of Environmental Quality, 4 (2): 267-273 (1975).
- Clark, J. W. and W. Viessman, Water Supply and Pollution Control, International Textbook Company, Scranton, Pennsylvania, 575 pp. (1965).
- Cross, F. A., L. H. Hardy, N. Y. Jones and R. T. Barber, "Relation between total body weight and concentrations of manganese, iron, copper, zinc, and mercury in white muscle of bluefish (Pomatomus saltatrix) and a bathyl-demersal fish (Antimora rostrata)," Journal of the Fisheries Research Board of Canada 30:1287-1291 (1973).
- Cunningham, J. D., Keeney, D. F., and J. A. Ryan, "Yield and metal composition of corn and rye grown on sewage sludge-amended soil," Journal of Environmental Quality, 4 (4): 448-454 (1975).
- Eganhouse, R. P., "The measurement of total and organic mercury in marine sediments, organisms, and waters," TM221, Southern California Coastal Water Research Project, El Segundo, California (1975).
- Environmental Protection Agency, "Mercury in sediment (cold vapor technique)," #759-396/101 (1972).
- Ganther, H. E., C. Boudie, M. L. Sunde, M. J. Kopecky, P. Wagner, S. H. Ott and W. G. Hoekstra, "Selenium: Relation to decreased toxicity of methylmercury added to diets containing tuna," Science 175:1122-1124 (1972).
- Goldwater, L. J., "Mercury in the environment," Scientific American 224(5): 15-21 (1971).

- Hannez (1968) For complete reference contact authors of this report.
- Hartung, R., "Heavy metals in the lower Mississippi," Proceedings of the International Conference on the Transition of Persistent Chemicals in Aquatic Ecosystems, Ottawa, Canada, p. I 93-98 (1974).
- Hatch, W. R. and W. L. Ott, "Determination of submicrogram quantities of mercury by atomic absorption spectrophotometry," Analytical Chemistry 40:2085 (1968).
- Henderick (1968) For complete reference contact authors of this report.
- Inhat, M. and R. J. Weslerby, "Application of flameless atomization to the atomic absorption determination of selenium in biological samples," Analytical Letters 7(4):257-265 (1974).
- Jensen, S. and A. Jernelev, "Biological methylation of mercury in aquatic organisms," Nature 223(5207):753-754 (1969).
- Johnels, A., et al., "Pike Esox lucius and some other aquatic organisms as indicators of mercury contamination in the environment," Oikos 18:323-333 (1967).
- Karnps, L. R., R. Carr and H. Miller, "Total mercury-monomethylmercury content of several species of fish," Bulletin of Environmental Contamination and Toxicology 8:273-279 (1972).
- Klein, D. H. and E. D. Goldberg, "Mercury in the Environment", Environmental Science and Technology, 4: 765-768 (1970).
- Ketchum, B. H., V. Zitko and D. Saward, "Aspects of heavy metal and organo-halogen pollution in aquatic ecosystems," in Ecological Toxicology Research, C. F. Mills and A. D. McIntyre, editors, Plenum Press, pp. 75-90 (1975).
- Langerwerff, J. V., Biersdorf, G. T., and D. L. Brower, "Retention of metals in sewage sludge 1: constituent heavy metals," Journal of Environmental Quality, 5 (1):19-22 (1976).
- Löfroth, G., "Methylmercury, a review of health hazards and side effects associated with the emission of mercury compounds into natural systems," Natural Science Research Council, Ecology Research Commission, Stockholm, Sweden, Bulletin 4:1-56 (1970).
- NAS, "Water quality criteria," National Research Council, Committee on Water Quality Criteria, Washington, D. C.: National Academy of Sciences (1973).
- NAS, "Disposal in the Marine Environment," Ocean Disposal Study Steering Committee, Washington, D. C.: National Academy of Sciences (1976).
- NOAA, "Ocean dumping in the New York Bight," NOAA Technical Report ERL 321-MESA 2, 78 pp., NOAA/ERL, U. S. Department of Commerce (1975).

- Nomiyama, K., "Toxicity of cadmium - mechanism and diagnosis," in Heavy Metals in the Aquatic Environment, P. A. Krenkel, editor, Pergamon Press, pp. 15-23 (1975).
- Oliver, B. G., and E. G. Cosgrove, "The efficiency of heavy metal removal by a conventional activated sludge treatment plant", Water Research, 9 (11):869-874 (1974).
- Peters, D. S., and M. Kjelson, "Consumption and utilization of food by various postlarval and juvenile fishes of North Carolina estuaries," Estuarine Research 1:447-472 (1975).
- SCCWRP, "The 1975 annual report," El Segundo, California (1975).
- SCCWRP, "The 1976 annual report," El Segundo, California (1976).
- Smith, R. G., and H. L. Windom, "Analytical handbook for the determination of arsenic, cadmium, cobalt, copper, iron, lead, manganese, mercury, nickel, silver and zinc in marine and estuarine environments," Marine Science Program of the University of Georgia Technical Report Series #72-6, Georgia Marine Science Center, Skidaway Island, Georgia, 61 pp. (1972).
- Van Loon, J. C., "Mercury input to the environment resulting from products and effluents from municipal sewage treatment plants," Environmental Pollution 7:141-147 (1974).
- Van Loon, J. C., J. Lichwa, D. Rutlan, and J. Kinrade, "The determination of heavy metals in domestic sewage treatment plant wastes," Water, Air, and Soil Pollution 2:473-482 (1973).
- Walter, C. M., F. C. June, and H. G. Brown, "Mercury in fish, sediments, and water in Lake Oahe, South Dakota," Journal of the Water Pollution Control Federation 45(10):2203-2210 (1973).
- Weaver, J. N., A. Hanson, J. McGaughey, and F. J. Steinkruger, "Determination of heavy metals in municipal sewage plant sludges by neutron activation analysis," Water, Air, and Soil Pollution 3:327-335 (1974).
- Weiss, H. V., M. Koide, and E. D. Goldberg, "Mercury in a Greenland ice sheet: Evidence of recent input by man," Science 174:692-694 (1971).
- Young, J. S., and J. B. Pearce, "Shell disease in crabs and lobsters from New York Bight," Marine Pollution Bulletin (in press) (1975).