# THE ROLE OF SPARTINA ALTERNIFLORA IN

# THE TRANSFER OF MERCURY IN A SALT MARSH ENVIRONMENT

A THESIS

22

Presented to

The Faculty of the Division of Graduate

Studies and Research

¢

122

by William R. Rahn, Jr.

15

In Partial Fulfillment

of the Requirements for the Degree

Master of Science in Geophysical Sciences

Georgia Institute of Technology

July, 1973

.

# THE ROLE OF SPARTINA ALTERNIFLORA IN

# THE TRANSFER OF MERCURY IN A SALT MARSH ENVIRONMENT

-

83

Approved:

Herbert L. Windom, Chairman

1 - 1 Charles E. Weaver 11 c = c

12

Kevin C. Beck

32

Date approved by Chairman: <u>9/20/73</u>

# TABLE OF CONTENTS

		Page								
ACKN	OWLEDGMENTS	Iv								
LIST C	OF TABLES	v								
LIST C	DF FIGURES	vi								
SUMM	ARY	viii								
Chapte	er									
I. INTRODUCTION										
II.	METHODS	6								
<ul> <li>Experimental Approach</li> <li>Detritus Experiments</li> <li>Metal Uptake by Detritus Vs. Decomposition of Detritus</li> <li>Uptake Vs. Time</li> <li>Uptake Vs. Weight</li> <li>Uptake Vs. Concentration Changes of Hg or Methyl Hg</li> <li>Uptake as a Function of Capillary Action</li> <li>Live Plant Experiments</li> <li>Uptake by Roots</li> <li>Water in Lower Compartment Only</li> <li>Water in Lower and Upper Compartments</li> <li>Uptake by Leaves and Stalks</li> <li>Water in Lower and Upper Compartments</li> <li>Water in Lower and Upper Compartments</li> <li>Uptake by Roots (Special Upper Chambers)</li> <li>Uptake by Roots Vs. Increasing Concentrations of Hg or Methyl Hg</li> </ul>										
111.	RESULTS Detritus Experiments Metal Uptake by Detritus Vs. Decomposition Uptake Vs. Time	17								

28

23

## TABLE OF CONTENTS (Continued)

### III. RESULTS (Continued)

32

Detritus Experiments (Continued) Uptake Vs. Weight Uptake Vs. Concentration Change of Hg or Methyl Hg Uptake as a Function of Capillary Action Live Plant Experiments Uptake by Roots Water in Lower Compartment Only Water in Lower and Upper Compartments Uptake by Leaves and Stalks Water in Upper and Lower Compartments Water in Upper Compartment Only Uptake by Roots Using Special Upper Chambers' Uptake by Roots Vs. Increasing Concentrations of Hg or Methyl Hg Marsh Box Experiments

IV.	CONCLUS	SIC	)N	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	5	56
BIBLIC	OGRAPHY			•															•			•	•								6	30

Page

### ACKNOWLEDGMENTS

I wish to express my sincere appreciation to Drs. C. E. Weaver and K. C. Beck for reviewing the manuscript and offering advice. My special appreciation and thanks are extended to my thesis advisor, Dr. Herbert L. Windom, whose guidance and patience were essential to the completion of this study. Thanks are especially in order for all the faculty and staff of Skidaway Institute of Oceanography whose friendship will long be remembered. The International Decade of Ocean Exploration provided funds for this project under grant number GX-33615.

Finally, to my wife Dahlia, I would like to express many thanks for patience and cheerfulness during all phases of my study.

٠.

# LIST OF TABLES

	Table		Page
	1.	Percent CH <sub>3</sub> HgCl Uptake: Plexiglass Boxes with Water in Upper and Lower Compartments (Lower Loaded)	39
a.	2.	Percent HgCl <sub>2</sub> Uptake: Plexiglass Boxes with Water in Upper and Lower Compartments (Lower Loaded)	40
	3.	Percent CH <sub>3</sub> HgCl Uptake: Plexiglass Boxes with Water in Upper and Lower Compartments (Upper Loaded)	43
	4.	Percent HgCl_ Uptake: Plexiglass Boxes with Water in Upper and Lower Compartments (Upper Loaded)	44
2	5.	Percent CH <sub>3</sub> HgCl Uptake: Plexiglass Boxes with Special 1 Liter Upper Chambers (Lower Loaded)	46
	6.	Percent HgCl <sub>2</sub> Uptake: Plexiglass Boxes with Special 1 Liter Upper Chambers (Lower Loaded)	47
	7.	Percent HgCl <sub>2</sub> Uptake: Marsh Box Experiments	52
	8.	Mercury Budget	58

# LIST OF FIGURES

Figure		Page
1.	Spartina's Influence on Hg Transfer in an Estuarine System	. 3
2.	Plexiglass Box	. 7
3.	Marsh-Tidal Simulator	. 9
4.	Photographs of Plexiglass Boxes with <u>Spartina</u> in Place	. 13
5.	Special One Liter Upper Chambers in Place in Plexiglass Box	. 15
6.	Detritus: Uptake of Methyl Hg at Different Concentrations Vs. Time	• 18
7.	Detritus: Uptake of Hg at Different Concentrations Vs. Time	. 19
8.	Detritus: Equilibrated Uptake Rate of Methyl Hg for 5 Month Period	. 20
9.	Detritus: Equilibrated Uptake Rate of Hg for 5 Month Period	. 21
10.	Fresh Detritus: Uptake of Hg Vs. Time for Sections of Spartina .	. 22
11.	Fresh Detritus: Uptake of Methyl Hg Vs. Time for Sections of Spartina	. 23
12.	Fresh Detritus: Uptake of Hg for Different Weights of Spartina	. 25
13.	Fresh Detritus: Uptake of Methyl Hg for Different Weights of <u>Spartina</u> Sections Vs. Time	• 26
14.	Detritus: Uptake of Methyl Hg at Different Concentrations of Methyl Hg for Sections of <u>Spartina</u> Vs. Time	. 27
15.	Detritus: Uptake of Hg at Different Concentrations of Hg for Sections of Spartina Vs. Time	• 28
16.	Uptake of Hg as a Function of Capillary Action	. 30

# LIST OF FIGURES (Continued)

Figure		Page
17.	Live Plants: Uptake of Methyl Hg Vs. Time (Lower Com- partment Spiked; No Water in Upper)	31
18.	Live Plants: Uptake of Hg Vs. Time (Lower Compartment Spiked; No Water in Upper)	32
19.	Live Plants: Uptake of Methyl Hg Vs. Time (Lower Com- partment Spiked; No Water in Upper)	33
20.	Live Plants: Uptake of Hg Vs. Time (Lower Compartment Spiked; No Water in Upper)	34
21.	Average Loss of Radioactive Methyl Hg from Uptake Solutions ···	36
22.	Average Loss of Radioactive Hg from Uptake Solutions	37
23.	Live Plants: Uptake of Methyl Hg and Hg Vs. Time (Lower Compartment Spiked; Water Also in Upper Compartment)	38
24.	Live Plants: Uptake of Methyl Hg and Hg Vs. Time (Upper Compartment Spiked; Water Also in Lower Compartment)	42
25.	Live Plants: Uptake of Methyl Hg and Hg Vs. Time (Upper Compartment Spiked; No Water in Lower Compartment)	45
26.	Live Plants: Uptake of Methyl Hg and Hg Vs. Time (Special 1 Liter Upper Chambers Used)	48
27.	Live Plants: Uptake of Methyl Hg at Different Concentrations of Methyl Hg for Sections of <u>Spartina</u> Vs. Time (Lower Com- partment Spiked)	50
28.	Live Plants: Uptake of Hg at Different Concentrations of Hg for Sections of <u>Spartina</u> Vs. Time (Lower Compartment Spiked)	51
29.	Injection Sites for Hg in the Marsh Box	54

15

ľ

### SUMMARY

The smooth cord marsh grass <u>Spartina alterniflora</u> was shown to be an effective agent in the uptake and transfer of the pollutant heavy metal mercury. Experiments were conducted as to the uptake, accumulation, and release of inorganic and organic mercury. These experiments were conducted using tagged Mercury 203 and Methylmercury 203.

Uptake rates of plant detritus as well as live plants were established. Mercury 203 and Methylmercury 203 were shown to be taken up at extremely rapid rates and usually concentrated in the plant root system. Results show that approximately 20-25% of available Hg 203 and MeHg 203 was taken up by <u>Spartina</u> root systems at equilibrium. Transfer to other portions of plants was noted and release rates of the tagged metals from plant leaves to surrounding waters were established. Five percent of the mercury available for uptake by <u>Spartina</u>, at equilibrium, was released to surrounding water by its leaves. The estimated total annual Hg uptake by <u>Spartina</u> is approximately 0.7 mg/m<sup>2</sup> yr. These results would imply that at least an additional 35 ug/m<sup>2</sup> yr would be taken up by the <u>Spartina</u> which is released to surrounding water.

Remobilization of mercury by the root systems of <u>Spartina</u> was shown to be an effective way of transferring mercury into the food web.

• •

### CHAPTER I

## INTRODUCTION

Because mercury is highly toxic to biological systems and is being introduced into the environment naturally as well as artificially it is important to identify processes which determine its fate (Windom, 1973). Mercury is ultimately discharged into the marine environment through estuarine systems (Cranston and Buckley, 1972); (Huckabee and Blaylock, 1972); so processes acting here are extremely important to the final disposition of this metal. Estuarine sediments can act as sinks for certain heavy metals (Windom, 1973). If this is the case for mercury, its uptake by <u>Spartina alterniflora</u> could be a significant process leading to its remobilization.

Many low trophic level organisms concentrate most heavy metals (i.e., Hg, Cd, As, Zn and Pb) (Galtsoff, 1960). If the uptake of metals at low trophic levels is great, their transfer to higher organisms in the food chain may lead to toxic conditions in these higher organisms (Jernelov and Lann, 1971). It is therefore important to understand metal transfer at the lower trophic levels of ecosystems such as the salt marshes of Georgia.

The most abundant primary producer in the estuaries of Georgia is the smooth cord grass, <u>Spartina alterniflora</u>. Because of the importance of this plant to the salt marsh ecosystem, the present study was initiated to determine

its role in the transfer of mercury through the salt marsh system. This study included experiments designed to establish rate of uptake and release of inorganic and methylmercury and their concentration in Spartina.

Heavy metals are of considerable importance because of their toxic nature to organisms (Saha, 1971). These organisms, and ultimately man, are being subjected to higher and higher concentrations of heavy metals due to the way organisms in food chains tend to convey the metals that they have concentrated to their preditors (Knauer and Martin, 1972). This study, using a highly productive primary producer, provided information on rates of availability of mercury which is possibly available to the next members of the food chain. Subsequent studies should be performed to obtain concentration rates of intermediate food chain members. This study, however, dealt only with an initial link between the salt marsh food web, <u>Spartina alterniflora</u>, and its environment.

Toxic metals are introduced into estuarine systems as schematically shown in Figure 1 which emphasizes <u>Spartina</u>. Input processes are shown as river input, atmospheric input, and ocean input. What happens to mercury upon entering the estuarine system is of major concern in the present study. Metals entering the system subsequently follow several pathways through it. They can be flushed directly out of the estuary to the sea, or be taken up directly from the water by organisms, both plant and animal. Also they may be deposited in the sediments where they may or may not be remobilized by organisms. It was the purpose of the present study to investigate only one part

 $\mathbf{2}$ 

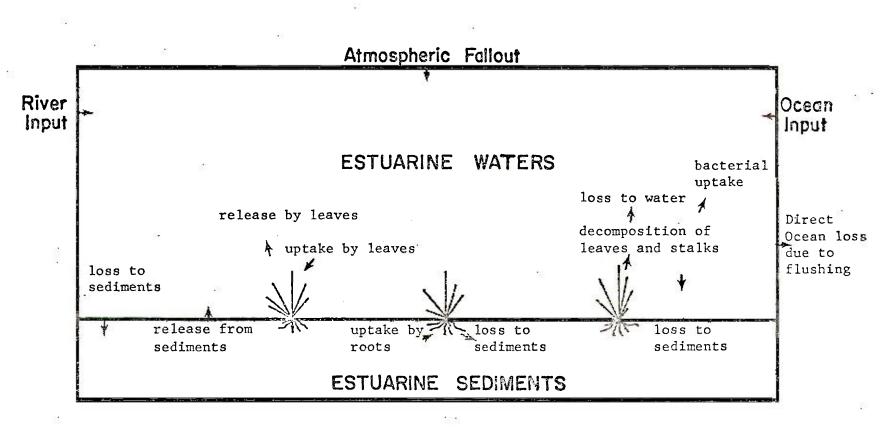


Figure 1. Spartina's influence on Hg. transfer in an estuarine system.

÷.,

cc

of this large system (Fig. 1) to elucidate the role of <u>Spartina alterniflora</u> in transferring the mercury. The choice of mercury for study is due to its high toxicity to animals and plant populations as well (Fishbein, 1971) (Gluoschenko, 1969) (Harris, White and Macfarlane, 1970).

The importance of <u>Spartina alterniflora</u> to the estuarine system of Georgia was eluded to above. Although <u>Spartina</u> is highly productive and occurs in dense stands, it probably does not serve as a direct food source to animals until it has undergone decomposition. Bacterial action predominantly accounts for the availability of <u>Spartina</u> as a food source (Pomeroy, <u>et al</u>, 1969). This study deals with the various sections of <u>Spartina alterniflora</u>. These sections are defined as 1) leaves (only the leaf blades are considered), 2) stalks, (the main stem of the plant), 3) Roots (all underground parts). This includes the underground stem (known as rhizomes) and root hairs (defined as the smallest sections of the root).

Since root haris are the most important uptake site for metals by vascular plants, the physiology of the root system plays a major role in the initial uptake of metals (Bristow and Whitcombe, 1971). Like other halophytes, <u>Spartina</u> is capable of maintaining high osmotic pressures. These pressures usually cause difficulty in the absorption of water; however, osmosis is not a factor in the absorption of inorganic substances by the roots. The absorption of inorganic substances usually in the form of ions is an exceedingly complex process which is not yet well understood. Ions diffuse into the root hairs independently of the movements of water. Diffusion is apparently the major factor controlling metal absorption; however, the basic laws of diffusion cannot alone explain their uptake since many are concentrated against a gradient (e.g., <u>Spartina</u> roots concentrate Hg at higher levels than the surrounding water). This phenomenon is not unique to <u>Spartina</u>. For example the freshwater algae <u>Nitetta</u> concentrates chlorine to two orders of magnitude higher in concentration than its support medium. Another more important factor influencing absorption of inorganic substances is the metabolic activity of the absorbing cells. This may produce the energy required for the diffusion of metals against the concentration gradient.

Once absorbed into the root system, metals may diffuse through plant cells until they reach the xylem (the interior conducting tissue of water and other nutrients) where they are carried to all parts of the plant.

The studies described in this report were designed to determine answers to the following questions:

- 1) What are the rates of Mercury uptake by <u>Spartina</u>?
  - a) by root systems?
  - b) by leaves and stalks?
- 2) What are the rates of Mercury release from <u>Spartina</u> to surrounding water?

3) What sections of <u>Spartina</u> accumulate most mercury?

#### CHAPTER II

### METHODS

Radioactive mercury 203 and methylmercury 203 were used to determine rates of uptake, release rates and concentrations in plants used in the laboratory. The amounts of radioactive tracer used in the experiments varied from  $0.1 \mu$ C to 50  $\mu$ C depending upon dilution factors relating to different amounts of water needed to conduct the experiment. Instrument detection problems were overcome in this manner. All activity was counted using an Ortec single channel scintillation counter with a Na-Iodide Well detector. Instrument efficiency was determined to be 1/10 of 1%.

Plexiglass boxes were made which allowed two compartment access to the plant systems. Using these boxes, radioactive tracer studies were performed on <u>Spartina alterniflora</u>. Figure 2 shows the box with <u>Spartina</u> in place. This is similar to the approach of McCroy and Barsdate (1968) who used a radio-active tracer study with eelgrass, <u>Zostera marina</u>. Containers were construct-ed which allowed the roots to be isolated from leaves. Access to this two compartment container was obtained through ports in the container. The plexiglass boxes were constructed of  $\frac{1}{4}$ " material cut and glued to size so that a top compartment could fit inside a lower compartment. Dimensions of the boxes are shown in Figure 2. Top compartments housed stems and leaves while the lower compartments housed roots and rhizomes. Each compartment remained sealed

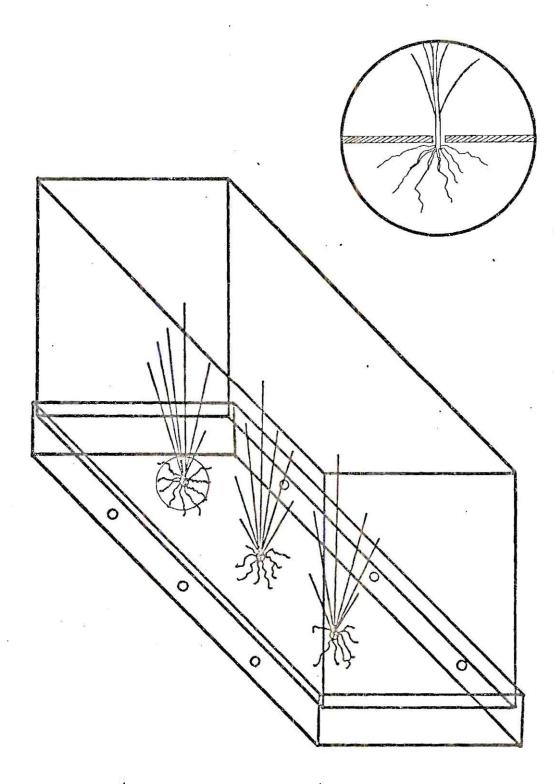


Figure 2.Plexiglass Box Capacities and Dimensions:<br/>Bottom Compartment:8 liters, 24 x 12 x 5 inches<br/>30 liters, 23½ x 11½ x 18 inchesTop Compartment:30 liters, 23½ x 11½ x 18 inches

from the other compartment while the experiments were in progress. The plants were positioned through holes in the bottom of the upper compartment and sealed with a rubber septum and silicone sealant. This allowed complete isolation of upper and lower water with the only channel for metal migration between the two compartments being the plant. Holes were placed in the sides of the lower compartment where rubber septum could be positioned for easy loading of the radioactive tracers. Filtered sea water  $(0.45 \,\mu)$  was generally used as a medium.

An existing outdoor marsh box was also employed (Figure 3). This box simulated actual marsh conditions in which periodic inudation of <u>Spartina</u> reflected tidal action. These boxes are approximately 6 x 4 feet with a 2 feet depth of substrate.

## Experimental Approach

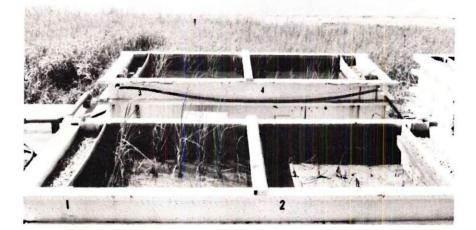
### Detritus Experiments

۰.

<u>Spartina</u> detritus was used in uptake experiments to obtain information on its ability to take up mercury from surrounding waters by absorption rather than by root systems. Since the physiological processes of the plant had been destroyed by the action of cutting the stalks and leaves into small (2-5 mm) sections the only uptake function available would be that of absorption. All detritus experiments except the decomposition experiments were run in duplicate. The values used for presentation are means of the replicates.

The detritus uptake experiments were of several types. Descriptions of

R.



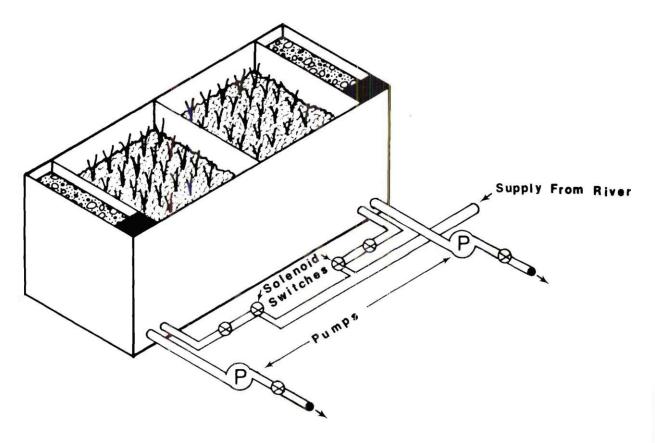


Figure 3. Marsh-Tidal Simulator

these are as follows:

<u>Metal Uptake by Detritus Versus Decomposition</u>. <u>Spartina</u> stalks and leaves were cut into 2-5 mm sections and placed in plastic mesh bags and allowed to decompose in the Skidaway River for a period of five months. At one month intervals during this period, samples were retrieved and subjected to uptake experiments. Equal amounts of the decomposed detritus were placed in beakers containing varying concentrations of mercury equilibrated with radioisotope in filtered sea water solutions. At 30 minute intervals the detritus was taken from its beaker, rinsed in distilled water, paper towel-dried and counted for radioactivity then replaced in the water. Counts were then converted to percent uptake and to actual uptake.

<u>Uptake Vs. Time</u>. <u>Spartina</u> stalks, leaves and roots were cut into sections and separately weighed into equal portions and placed in beakers containing spiked filtered sea water. At 15 minute intervals the detritus was taken from each beaker, rinsed in distilled water, towel-dried and counted for radioactivity then replaced in the water. The counts were converted to percent uptake of available Hg 203 or MeHg 203.

<u>Uptake Vs. Weight.</u> <u>Spartina</u> stalks and leaves (mixed) and <u>Spartina</u> roots were cut into sections and weighed out into varying amounts (0.1 g, 0.5 g, 1.0 g and 2.0 g). These amounts of detritus were then placed in beakers containing equal amounts of spikes. At 30 minute intervals the samples were taken from the water, rinsed, towel-dried and counted, then returned to the beakers for additional soaking. The counts were converted to percent uptake of available Hg 203 or MeHg 203.

<u>Uptake Vs. Concentration Changes of Hg or MeHg.</u> Spartina roots were cut into 2-5 mm sections, weighed and placed in three beakers containing equal amounts of either Hg 203 or MeHg 203 and equilibrated with normal Hg or MeHg at levels of 10, 100 and 1000 ppb, respectively. At 15 minute intervals the roots were taken from their respective beakers, rinsed, towel-dried and counted for radioactive uptake. These counts were then converted to percent uptake. This experiment was repeated with stalks and leaves of Spartina as well.

<u>Uptake as a Function of Capillary Action</u>. In order to investigate capillary action in <u>Spartina</u>, sections of <u>Spartina</u> leaves measuring approximately 30 mm were placed vertically in approximately five mm of Hg 203 spiked water. Periodically the leaves were removed from the solutions, rinsed, towel-dried and cut into five mm sections. Each five mm section was then counted for radioactivity and the counts converted to percent of available Hg 203 in each section. MeHg 203 was not available for use in the experiments.

## Live Plant Experiments

These experiments were performed using the plexiglass boxes described previously. <u>Spartina</u> plants were dug from an undisturbed marsh area so that root and rhizome systems would remain intact. These plants were then washed free of all mud. Leaf scars which usually remain on the lower stalks of <u>Spartina alterniflora</u> were also removed. These leaf scars are the residue of dead leaves that remain attached to the <u>Spartina</u> at the leaf base. The <u>Spartina</u> plants were then inserted into rubber septa and the rubber septa were placed in the holes in the bottom of the upper compartment of a plexiglass box. Around the septa silicone sealant was used to finalize the seal. Photographs of the plants in place are shown in Figure 4. After being fixed in the plexiglass boxes the plants were then ready for uptake and transfer experiments. All water used in these experiments was filtered sea water, unless otherwise specified. Three plants were always used in each plexiglass box experiment. Duplicates were run in all cases to check for accuracy of experimentation. Tables and plots are taken as the mean of a set of replicates.

Uptake by Roots (1 - Water in Lower Compartments Only). Experiments were performed using both Hg 203 and MeHg 203 in which the lower compartment of the plexiglass boxes was flooded and spiked. The upper compartment held no water. At intervals of time entire plants were taken from the box, rinsed in distilled water, towel-dried and cut into sections (i.e., roots, leaves and stalks). These sections were counted for radioactivity and the counts converted into percent uptake of Hg 203 or MeHg 203 per gram weight of Spartina.

Similar experiments were also conducted in which the <u>Spartina</u> was placed in 400 ml beakers in such a way that the root system was submerged in spiked sea water. These plants were also periodically taken from the beakers, rinsed, towel-dried, cut into sections, counted and weighed.

Uptake by Roots (2 - Water in Lower and Upper Compartments). Water was used in the upper and lower compartments in these experiments. The Hg 203 or MeHg 203 was injected into the lower compartment and uptake and transfer was noted by the movement of Hg 203 or MeHg 203 in the plants. In these

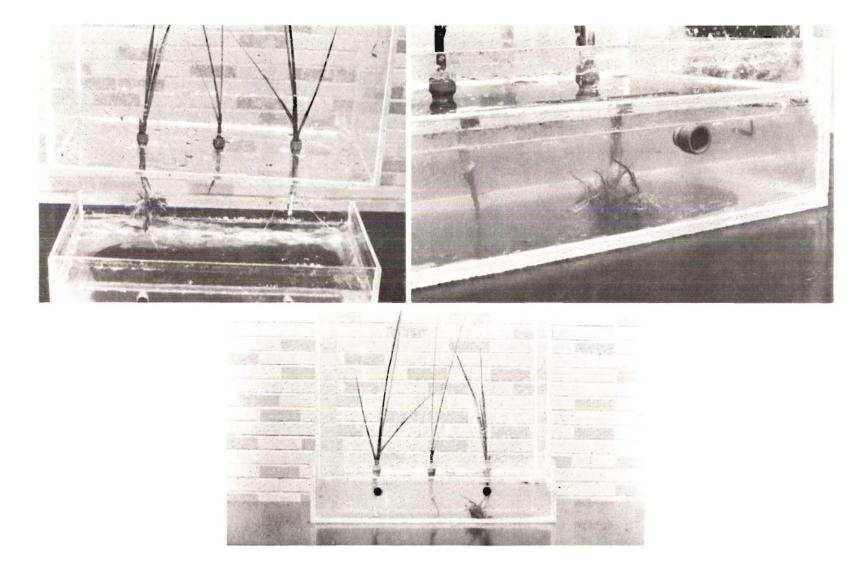


Figure 4. Photographs of Plexiglass Boxes with Spartina in place.

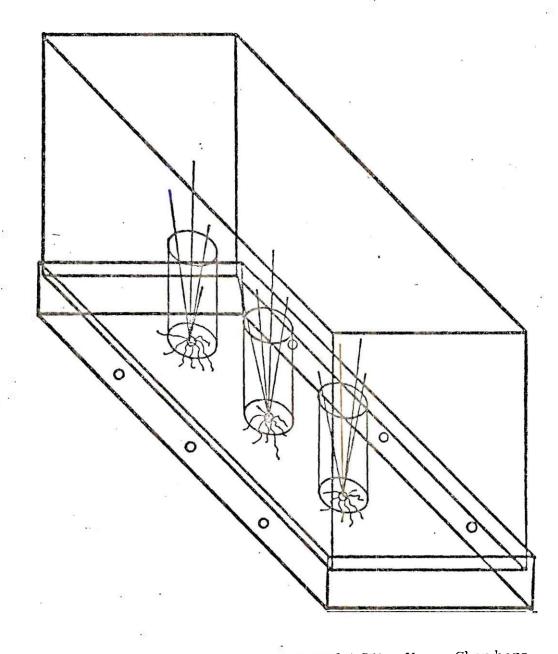
experiments the water in the upper compartment as well as lower was monitored for radioactivity. The detection of upper compartment mercury activity would reflect a release from the leaves or stalks of <u>Spartina</u>. As in the previous experiments the plants were taken from the boxes at time intervals, weighed and counted. Percentages of Hg 203 and MeHg 203 uptake were calculated per gram weight of Spartina.

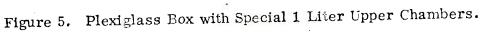
<u>Uptake by Leaves and Stalks (1 - Water in Lower and Upper Compartment</u>). The Hg 203 or MeHg 203 spike in these experiments was added to the upper compartment so that transfer from leaves and stalks to roots and rhizomes could be detected. The water in the lower compartment was monitored to reflect a loss of metal from the roots to surrounding water. The <u>Spartina</u> plants were periodically taken, rinsed, dried, counted and weighed. Counts were converted to percent of mercury available as was done in previous experiments.

<u>Uptake by Leaves and Stalks (2 - Water in Upper Compartment Only)</u>. Same as above but no water was contained in lower compartment. The experiments were otherwise duplicates of the above.

<u>Uptake by Roots (Special Upper Chambers)</u>. A set of experiments similar to above was conducted, but instead of the upper compartment being used, smaller one liter chambers were fitted so that detection of radioactive Hg 203 or MeHg 203 could be made easier (Figure 5). Release from leaves and stalks could be detected at lower levels in this manner.

<u>Uptake by Roots Vs. Increasing Concentrations of Hg 203 or MeHg 203.</u> Spartina plants were placed in a set of three 400 ml beakers with approximately





200 ml filtered sea water covering the root systems. The concentration of mercury in each of three beakers was raised to 10 ppb, 100 ppb and 1000 ppb, respectively. Both Hg 203 and MeHg 203 were used in identical experiments. After these spikes were added to the solutions, individual plants were periodically taken out, rinsed, dried, cut into sections, counted and weighed. The counts were then converted to percent uptake of the metal per gram weight of Spartina.

## Marsh Box Experiments

Experimental boxes which simulated actual marsh tidal conditions were used to culture <u>Spartina alterniflora</u> (Figure 3). Sand was used as the substrate in which the <u>Spartina</u> grew. In these experiments 0.4 mC Hg 203 was injected at eight locations in the box all at 10 cm below the surface, so that each spiked area received 0.05 mC Hg 203. <u>Spartina</u> plants were taken from the boxes at different time intervals for five months. Each plant was rinsed, dried, cut into sections, counted, and weighed so that uptake of Hg 203 could be followed. The metal uptake by the plants was monitored in relation to proximity to injection site.

## CHAPTER III

### RESULTS

### Detritus Experiments

## Metal Uptake by Detritus Versus Decomposition

Figures 6 and 7 show uptake rates in ng/g of fresh <u>Spartina</u> detritus. These results suggest that the initial mercury uptake rate increases as the concentration of Hg in solution increases. After equilibration or saturation (approximately 24 hours) the same percentage of the mercury available in the medium was taken up by the plant detritus regardless of the concentration in the medium. This was true for both Hg and MeHg and amounted to approximately 10% of the total available mercury for the weight of detritus used (5 g/liter).

As the <u>Spartina</u> detritus decomposes (Fig. 8 and 9) from month to month, there is a total increase in the equilibrated uptake rate for each concentration level. This equilibrated uptake rate is defined as the concentration of mercury per weight of plant after equilibrium with the medium. This increased uptake may be due to bacterial colonization of the detritus which also has an increased surface area. A maximum is generally reached around the third month of decomposition.

### Uptake Vs. Time

۰.

Figures 10 and 11 show uptake curves for stalks, leaves, and roots of

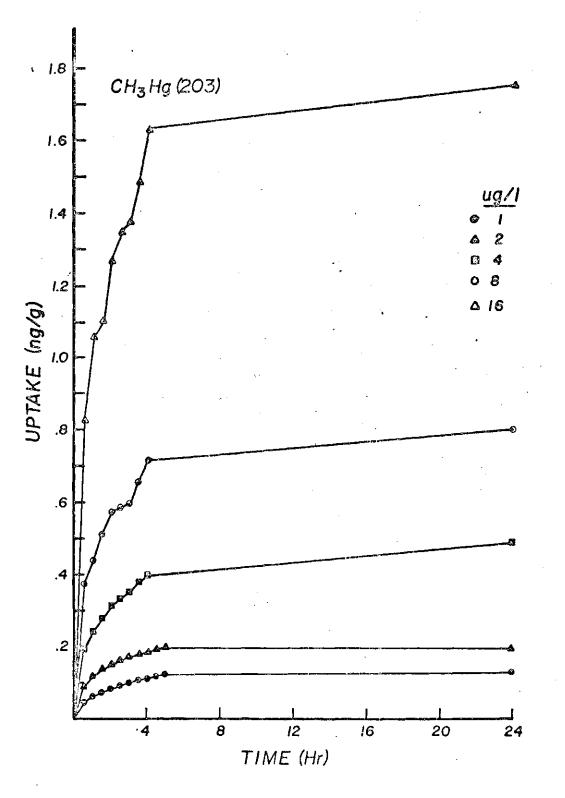


Figure 6. Detritus: Uptake of MeHg at Different Concentrations Vs. Time

18

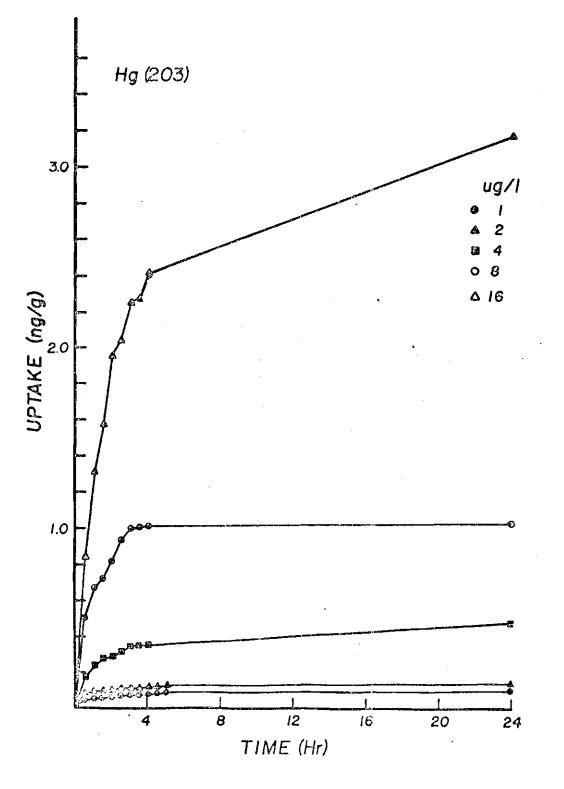
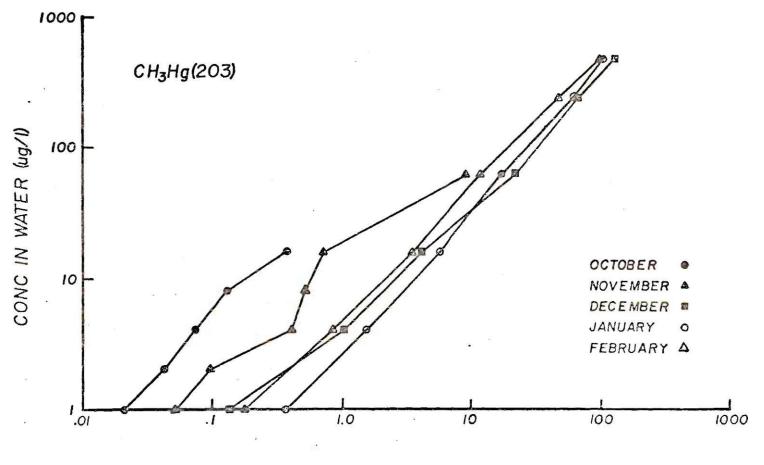


Figure 7. Detritus: Uptake of Hg at Different Concentrations Vs. Time



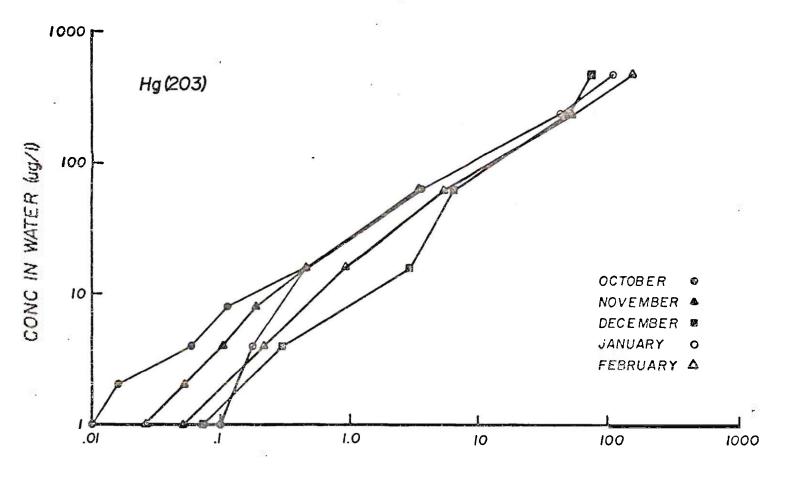
1

RATE OF UPTAKE (ng/g detritus/hr)

Figure 8. Detritus: Equilibrated Uptake Rate of MeHg for 5-Month Period

20

•



RATE OF UPTAKE (ng/g detritus/hr)

Figure 9. Detritus: Equilibrated Uptake Rate of Hg for 5-Month Period

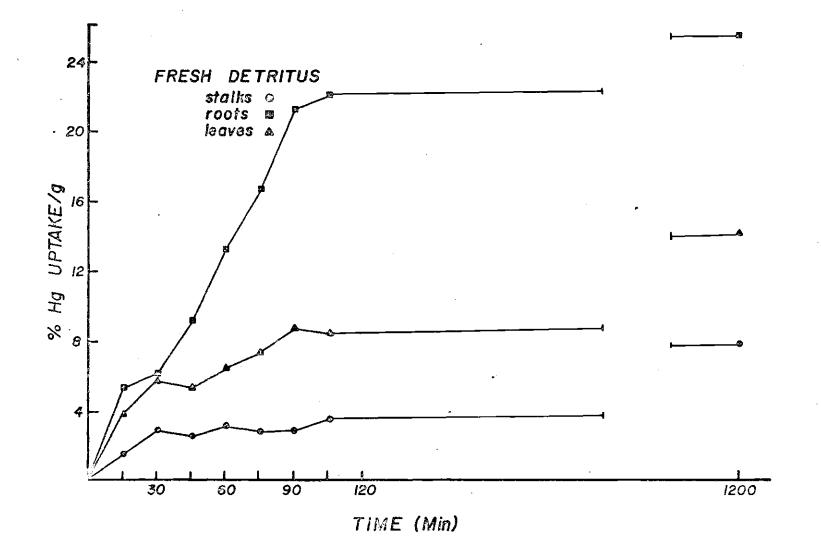


Figure 10. Fresh Detritus: Uptake of Hg Vs. Time for Sections of Spartina

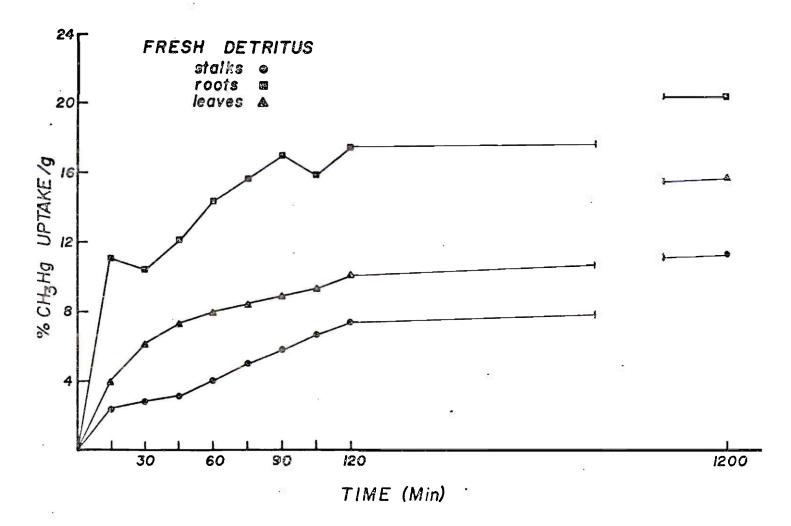


Figure 11. Fresh Detritus: Uptake of MeHg Vs. Time for Sections of Spartina

<u>Spartina.</u> It can be seen clearly that uptake by roots is far more rapid than uptake by stalks and leaves. Maximum uptake is usually reached in 4 hours for all sections of Spartina.

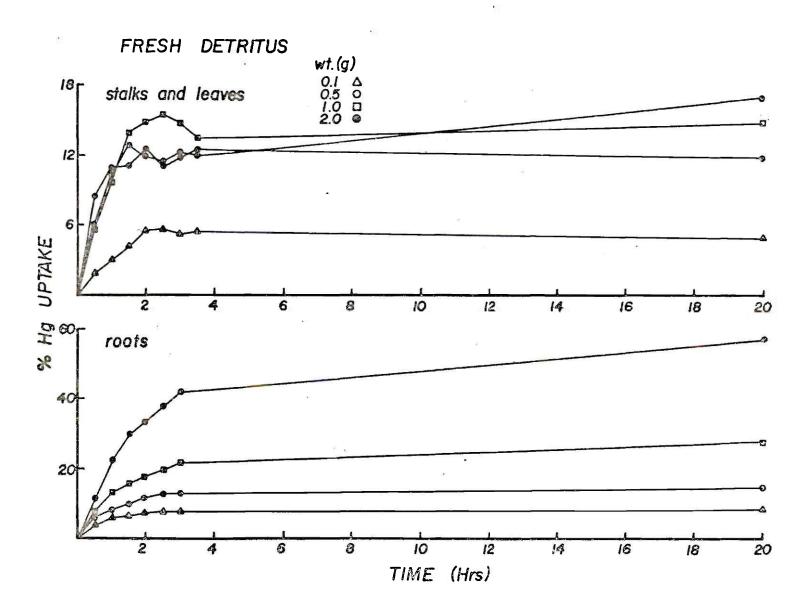
## Uptake Vs. Weight

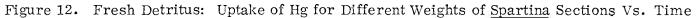
It was originally thought that to a certain point, the uptake of mercury would be directly proportional to the weight of <u>Spartina</u>. These experiments, however, show this is not necessarily true for stalks and leaves (Figure 12 and 13). A more important criteria for uptake here would be the surface area available for adsorption. Increased uptake does, however, take place with increased weight of root hairs. Although the surface areas for leaves and stalks were probably similar, at least superficially to the root samples, the enhanced mercury uptake by the latter apparently reflects the extremely different nature of the texture and composition of the two types of materials. This conclusion follows for both Hg and MeHg.

These uptake rates as will be true for all following figures (except where otherwise specified) are plotted as percent of available Hg taken up per gram weight of <u>Spartina</u> in 4 liters of medium. This appears to be the most reasonable approach to data presentation since uptake was shown to be a function of total concentration of Hg in the solution.

# Uptake Vs. Concentration Change of Hg or MeHg

Figures 14 and 15 show results of experiments where attempts were made to find saturation points of mercury uptake or the concentration of mercury in the medium at which no further uptake is observed. As can be seen, very little





• •

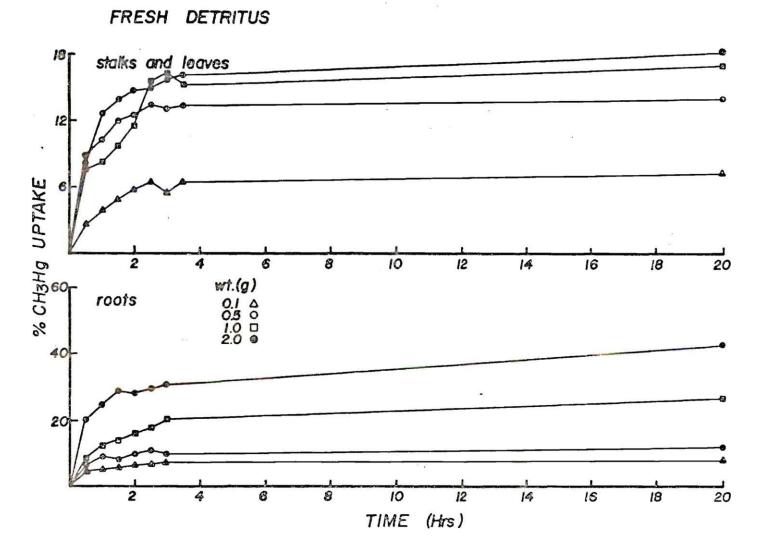


Figure 13. Fresh Detritus: Uptake of MeHg for Different Weights of Spartina Sections Vs. Time

...

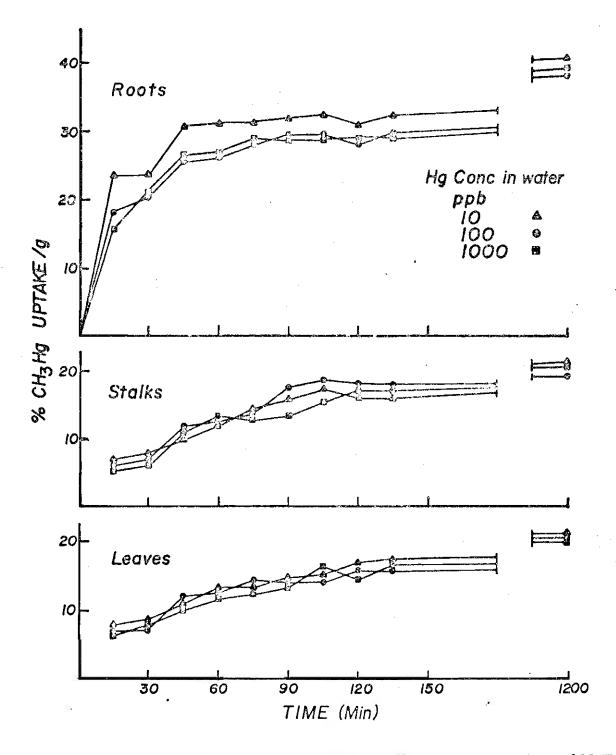


Figure 14. Detritus: Uptake of MeHg at Different Concentrations of MeHg for Sections of <u>Spartina</u> Vs. Time

 $\mathbf{27}$ 

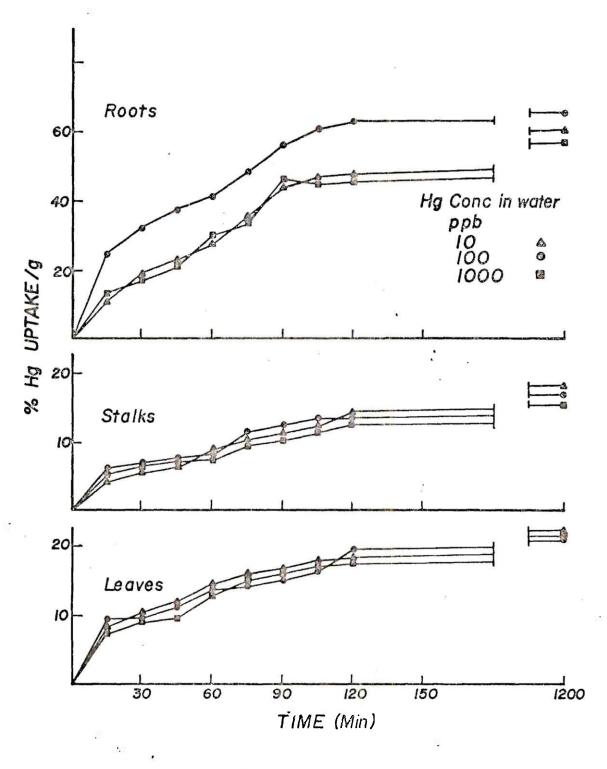


Figure 15. Detritus: Uptake of Hg at Different Concentrations of Hg for Sections of Spartina Vs. Time

change in uptake percentages was observed when the concentrations of mercury in the uptake solution was 10, 100 or 1000 ppb. This is again evidence that <u>Spartina</u> takes up mercury at high concentrations as rapidly as it does at low concentrations. A saturation point where no further mercury is taken up is apparently above any reasonable level that would be found in the natural environment.

### Uptake as a Function of Capillary Action

Results of these experiments show that no significant uptake occurs due to capillary action (Fig. 16). Slight uptake (less than 0.5% of the available Hg) did occur after 68 hours but only in the section nearest to the surface of the spiked solution.

### Live Plant Experiments

The uptake of Hg 203 and MeHg 203 by live <u>Spartina</u> plants is similar to the uptake by detritus in that the most rapid uptake occurs at the beginning of each experiment. In all cases a greater proportion of the available Hg 203 was taken up as compared with MeHg 203.

### Uptake by Roots (1 - Water in Lower Compartment Only)

۰.

The MeHg 203 and Hg 203 taken up by live plants (Figures 17 and 18) in a 120 hour period is almost all confined to the root systems. Although uptake by roots was rapid, no transfer to the upper parts of the plants was recorded in this period of time for any of these experiments. In later experiments, using longer uptake periods and higher concentrations of mercury spike, transfer to leaves was detectable (Fig. 19 and 20).

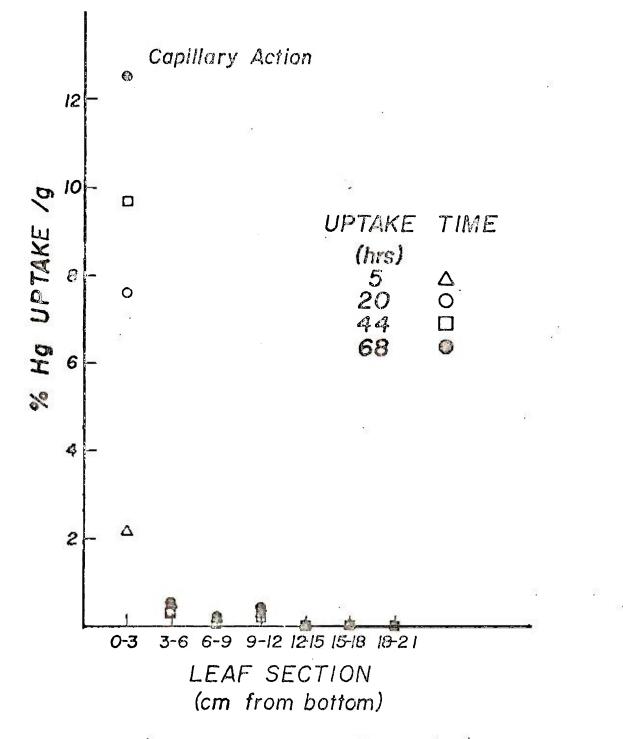
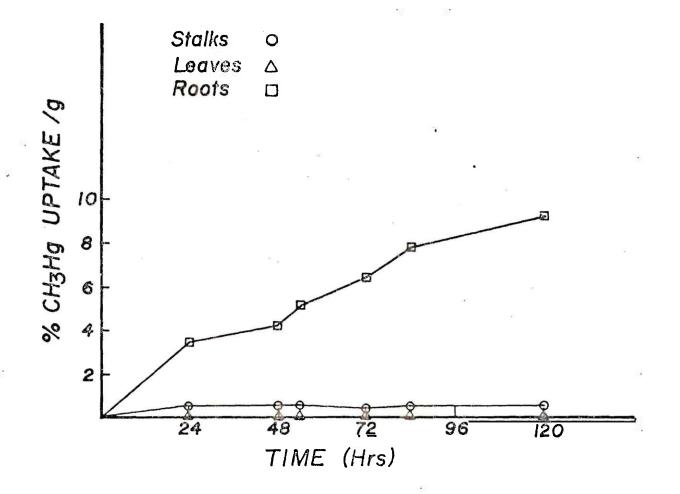
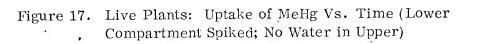
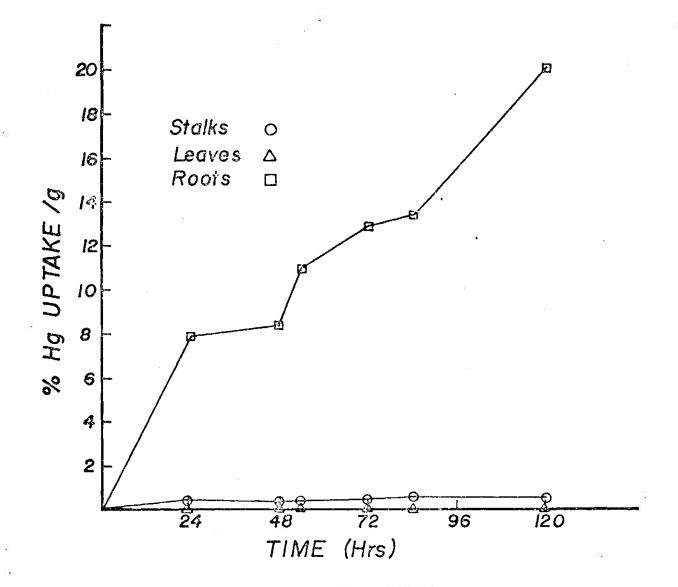
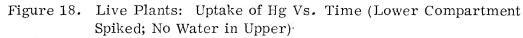


Figure 16. Uptake of Hg as a Function of Capillary Action









;

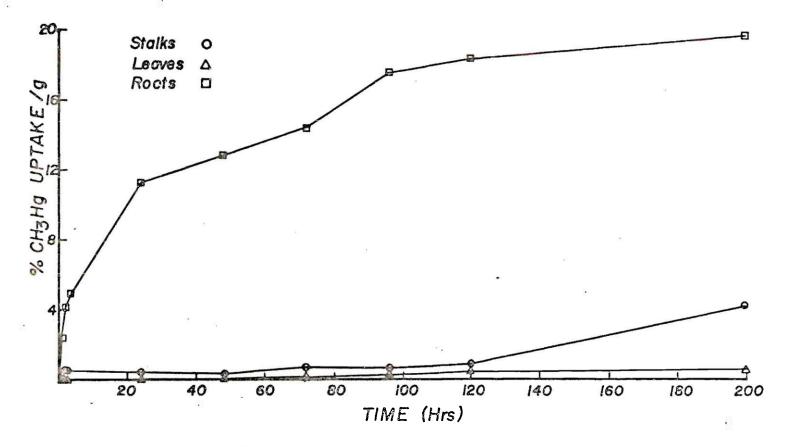


Figure 19. Live Plants: Uptake of MeHg Vs. Time (Lower Compartment Spiked; No Water in Upper Compartment)

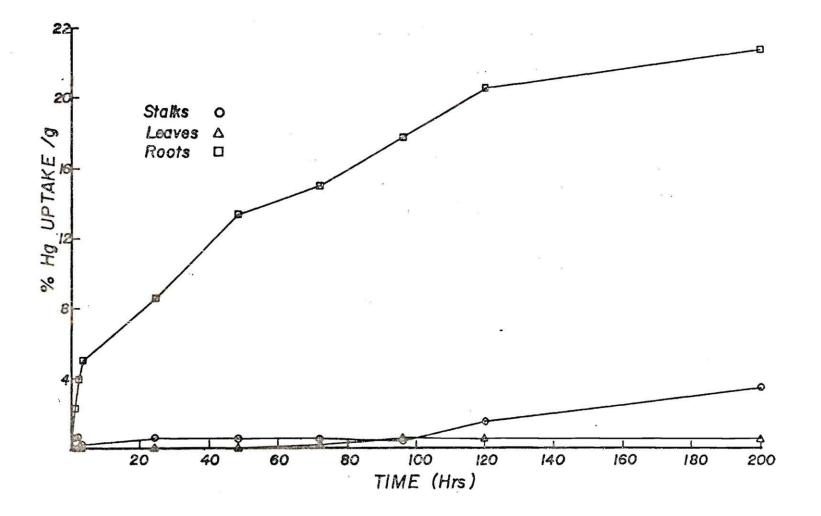


Figure 20. Live Plants: Uptake of Hg Vs. Time (Lower Compartment Spiked; No Water in Upper)

Figure 19 shows that for MeHg 203 about 25% of the total available mercury can be taken up. Of this 25%, 4% is transferred to stalks and approximately 1% is transferred to leaves. The total uptake of Hg 203 is similar to MeHg 203 or approximately 27% (Fig. 20). Twenty-three percent remained in the root system, while 1% went to the leaves and 3% went to the stalks, again similar to MeHg 203.

The tendency of MeHg 203 and Hg 203 to concentrate in the roots of <u>Spar-</u> <u>tina</u> is evident by the fact that 83 to 85% of the MeHg 203 and Hg 203 taken up by the plant remained here.

Results shown in Figures 21 and 22 indicate that the average loss of radioactivity from the uptake solutions versus time for Hg 203 and MeHg 203 in the root uptake experiments is similar whether fresh or salt water is used. Uptake percentages in the plants and percentages remaining in solution for any given time do not add up to be 100% due to adsorption to beaker and box walls; loss due to rinsing the plants after uptake periods and adsorption to particles of plant matter settled in the container bottoms. Differences in counting efficiencies account for some of the deviation as well.

### Uptake by Roots (2 - Water in Lower and Upper Compartments)

As already mentioned the purpose of this set of experiments was to show whether or not mercury could be taken up by roots, transferred to the leaves and subsequently released to surrounding waters. Results shown in Figure 23 indicates that very small amounts of mercury actually reached the leaves and from Tables 1 and 2, it can be seen that no radioactivity was noted in the upper

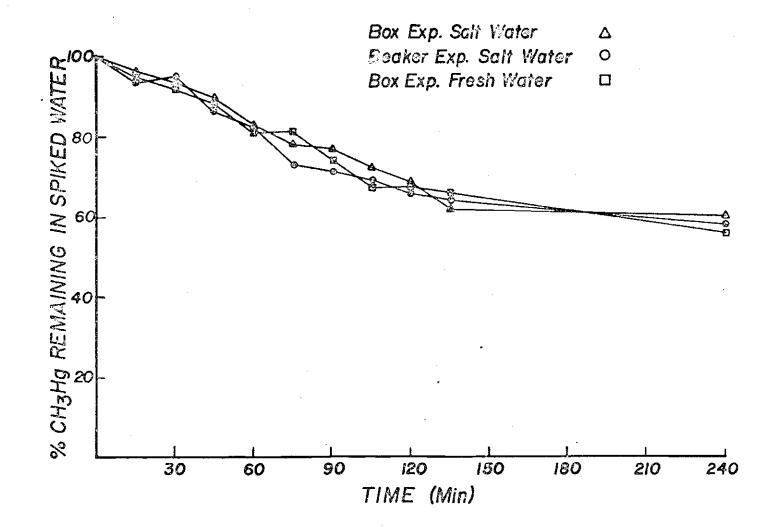


Figure 21. Average Loss of Radioactive MeHg from Uptake Solutions.

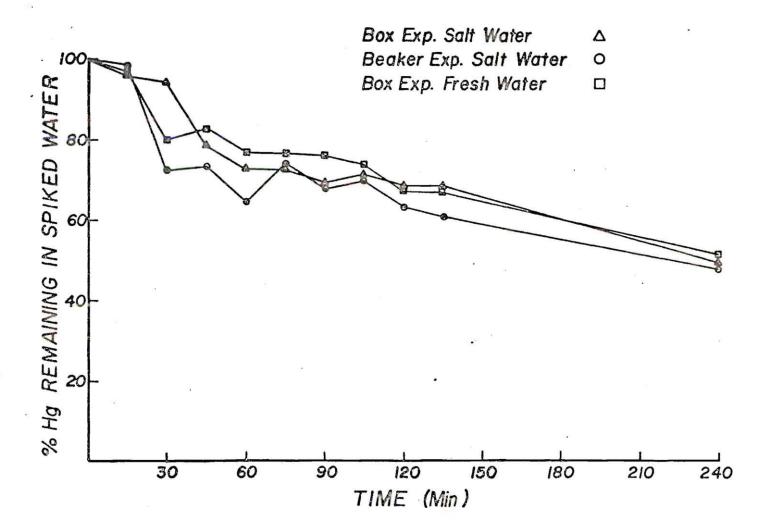


Figure 22. Average Loss of Radioactive Hg from Uptake Solutions

\$

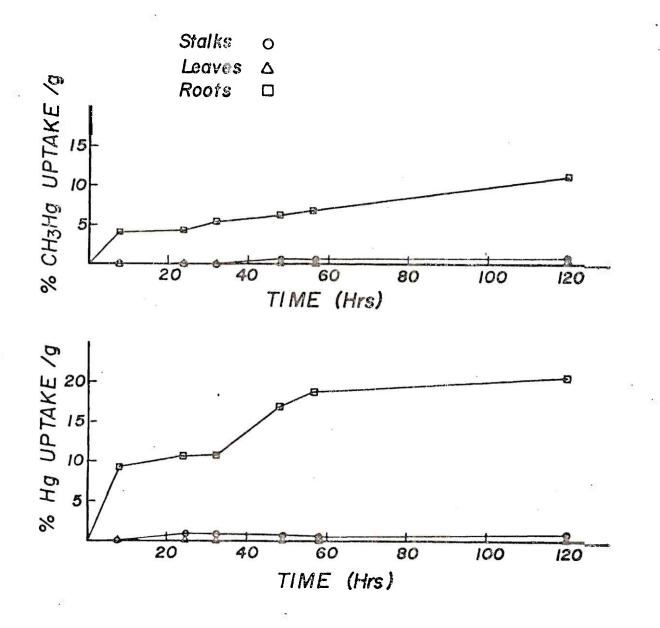


Figure 23. Live Plants: Uptake of MeHg and Hg Vs. Time (Lower Compartment Spiked; Water Also in Upper Compartment)

Time	Roots	Stalks	Leaves	%CH <sub>3</sub> Hg	%CH <sub>3</sub> Hg
(hrs) -	27	20 BF	1007	in Upper	in Lower
8	4.3	<u></u>	-	_	68.0
24	4.6	<del></del>	-	-	60.0
24	4.7	-	-	-	60.0
32	5.2	-	-	-	58 <b>. 7</b>
48	6.2	.01	-	-	56.2
48	6.4	.04	.04	-	56.2
56	6.8	.1	.02	_	50.1
120	11.4	• 2	.06	-	31.2
120	11.6	• 2	.06	-	31.2

.

۰.

5.5

### Table 1. % CH<sub>3</sub>HgCl Uptake/g: Plexiglass Boxes with Water in Upper and Lower Compartment (Lower Loaded)

Time (hrs)	Roots	Stalks	Leaves -	%HgCl <sub>2</sub> In Upper	%HgCl <sub>2</sub> In Lower
8	9.0	_		1 <u>1</u> 10	72.0
24	11.0	_	-	<del></del>	60.2
24	11.0		_	<u>-</u>	60.2
32	11.0	-	-		57.6
48	17.0	-	-	-	52.2
48	16.0	-	-	<u>-</u> 1	52.2
56	18.0	.1	.01	<b></b>	48.1
120	21.0	.2	.01	-	32.6
120	20.0	.2	.01	-	32.6

Table 2.	% HgCl <sub>2</sub> Uptake/g: Plexiglass Boxes with Water	
	in Upper and Lower Compartments (Lower Loaded	)

water surrounding the leaves (below detectability). This inability to detect the release of mercury by leaves was due to two factors: 1) insufficient mercury was transferred to the leaves and 2) the upper compartment contained too great a volume of water to detect very small amounts of radioactivity. This latter problem was remedied in later experiments which employed the use of special upper chambers (Fig. 5) which were placed around each plant.

### Uptake by Leaves and Stalks (1 - Water in Upper and Lower Compartments)

Results of these experiments show that uptake of Hg 203 and MeHg 203 by leaves of live <u>Spartina</u> plants is slow (Figure 24). Hg 203 appears to be taken up by the leaves better and transferred more efficiently to the stalks than MeHg 203. Results shown in Tables 3 and 4 indicate Hg or MeHg is not transferred to the lower water compartment.

### Uptake by Leaves and Stalks (2 - Water in Upper Compartment Only)

Results shown in Figure 25 are similar to those in the above (Figure 24) experiments. Again Hg 203 seems to be taken up by leaves and transferred to stalks more efficiently than MeHg 203.

### Uptake by Roots (Using Special Upper Chambers)

Results of experiments where the special upper chambers were used are given in Tables 5 and 6 and Figure 26. In this case the lower chambers were spiked. Uptake of Hg 203 and MeHg 203 by the root systems is again 20-25%, but in these experiments sufficient amounts of mercury transferred to the leaves was released to the water in the smaller volume containers surrounding the plants.

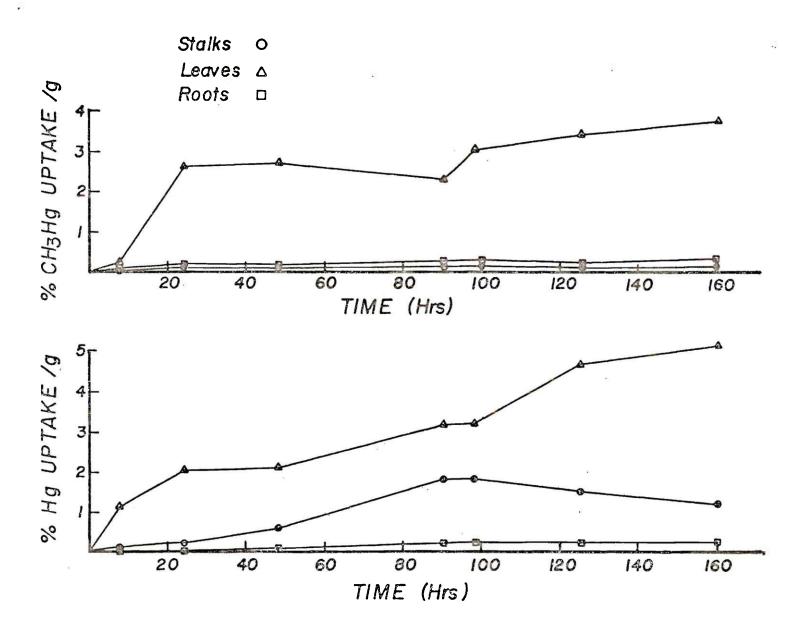


Figure 24. Live Plants: Uptake of MeHg and Hg Vs. Time (Upper Compartment Spiked; Water Also in Lower Compartment)

Time (hrs)	Roots	Stalks	Leaves	%CH <sub>3</sub> Hg In Upper	%CH <sub>3</sub> Hg In Lower
8	.04	. 05	.2	64.2	
24	.08	.14	2.6	52.2	-
48	.13	.12	2.7	42.2	3. <del>44</del> )
90	• 2	.10	2.3	33.6	17. <u>111</u> 1
98	• 2	.30	3.0	33.0	1.55
125	• 2	1.2	3.4	N.R.	N.R.
160	.2	.12	3.8	32.3	1000
	5				

25

Table 3. % CH<sub>3</sub>HgCl Uptake/g: Plexiglass Boxes with Water in Upper and Lower Compartments (Upper Loaded)

N.R. = Not Recorded

۰.

.

Time (hrs)	Roots	Stalks	Leaves	%HgCl <sub>2</sub> In Upper	%HgCl <sub>2</sub> In Lower
8		.07	1.1	41.6	
<b>24</b>	-	.18	2.0	38.6	(7 <del>11)</del>
48	.1	.6	2.1	31.1	-
90	• 2	1.8	3.4	32.6	-
<b>9</b> 8	.2	1.8	3.2	30.1	-
125	• 2	1.5	4.6	N.R.	N.R.
160	• 2	1.2	5.2	28.7	(7 <u>14</u> )

Table 4.	%HgCl <sub>2</sub> Uptake/g: Plexiglass Boxes with Water
	in Upper and Lower Compartments (Upper Loaded)

N.R. = Not Recorded

·

.

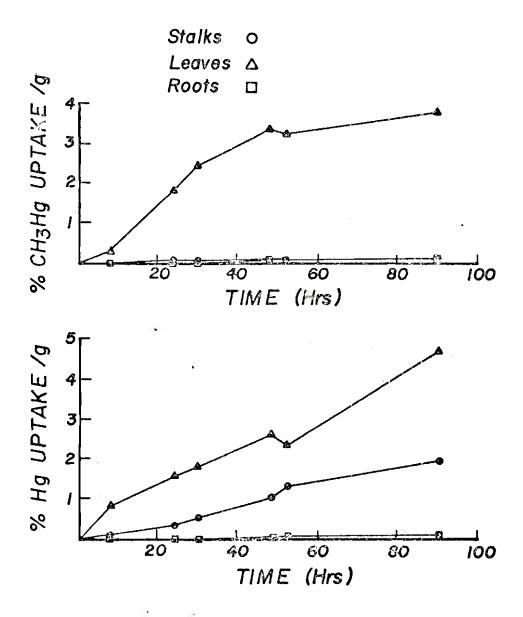


Figure 25. Live Plants: Uptake of MeHg and Hg Vs. Time (Upper Compartment Spiked; No Water in Lower Compartment)

45

Time (hrs)	Roots	Stalks	Leaves	%CH <sub>3</sub> HgCl in Water in Upper	%CH <sub>3</sub> HgCl in Water in Lower (Adjusted)
96	10.2	_	_	_	48.6
192	12.2	1.0	.2	.3	28.4
264	18.1	2.4	• 5	. 1.1	23.0

### Table 5. % CH<sub>3</sub>HgCl Uptake/g: Plexiglass Boxes with Special 1 Liter Upper Chambers (Lower Loaded)

Time (hrs)	Roots	Stalks	Leaves	%HgCl <sub>2</sub> in Water in Upper	%HgCl <sub>2</sub> in Water in Lower (Adjusted)
96	8.7	.1	÷	-	50.6
192	14.6	•1	.1	.2	33.3
264	21.8	1.1	•4	.9	16.1

•

•

Table 6.	%HgCl2 Uptake/g: Plexiglass Boxes with Special 1
	Liter Upper Chambers (Lower Loaded)

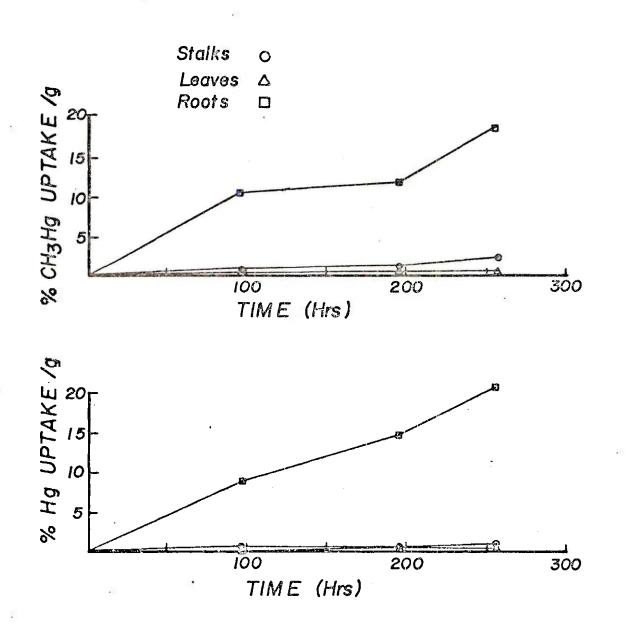


Figure 26. Live Plants: Uptake of MeHg and Hg Vs. Time (Special 1 Liter Upper Chambers Used)

i.

The amounts of Hg 203 and MeHg 203 which are released from the leaves is approximately 1% of the total mercury available for uptake and approximately 5% of the total taken up by the plants.

### Uptake by Roots Vs. Increasing Concentrations of Hg 203 and MeHg 203

The purpose of these experiments was to establish saturation levels of mercury uptake. The results are shown in Figures 27 and 28. Although concentrations of mercury in the uptake media were raised to 1 ppm there was essentially no change in the uptake rate. As with the detritus, the plant is capable of accumulating the same proportion of the available mercury over a range of concentrations exceeding what might be environmentally significant (Windom, 1973). Differences in the uptake of MeHg 203 and Hg 203 were again very slight.

### Marsh Box Experiment

Table 7 gives the results of uptake by plants in the marsh box experiments. A, B, and C indicate distance from injection sites (Figure 29). The proximity of the plants to actual injection sites were recorded as A-Nearest (5-10 cm), B-Midway (10-15 cm), C - Farthest away (15-25 cm). The eight injection sites were numbered 1-8. The numeral suffixes represent different injection sites. The uptake percentages were calculated on the basis of total Hg 203 injected at one site. Only Hg 203 was used since only one marsh box was available which had suitable grass growth and was not already involved in other experiments.

Sand samples were taken randomly throughout the box during the first two months of the experiment. Radioactivity was noted in the sediments only during the first two days after injection.

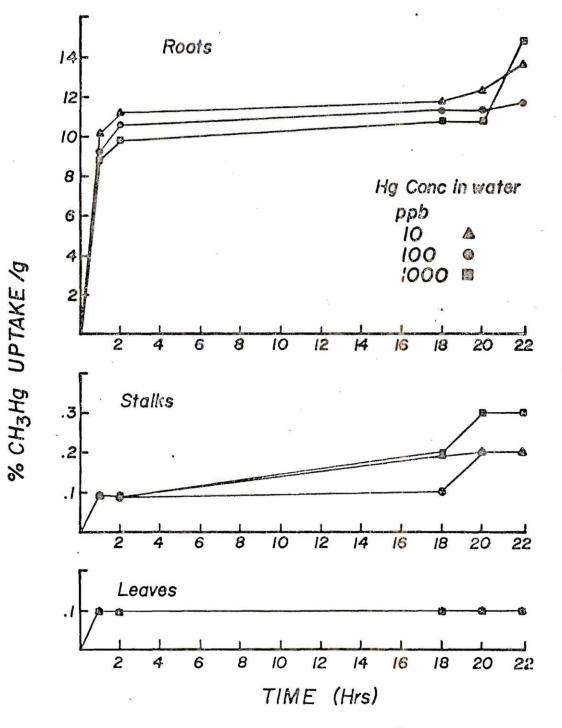
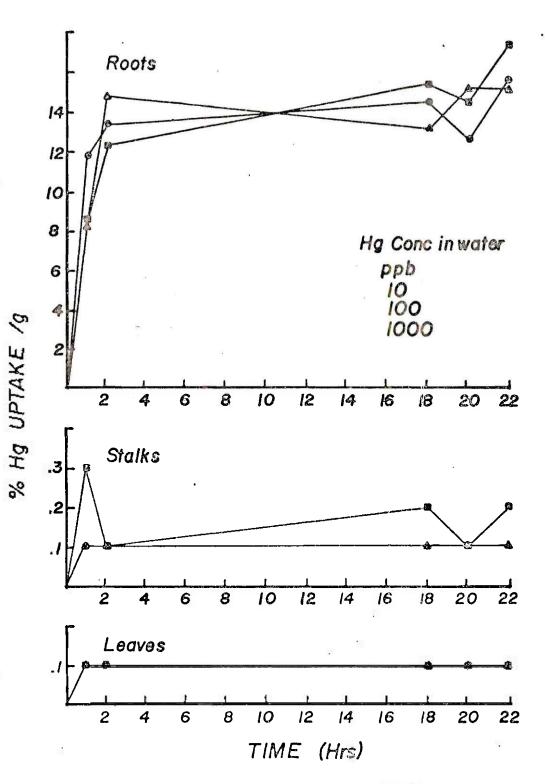


Figure 27. Live Plants: Uptake of MeHg at Different Concentrations of MeHg for Sections of <u>Spartina</u> Vs. Time (Lower Compartment Spiked)



٠.

Figure 28. Live Plants: Uptake of Hg at Different Concentrations of Hg for Sections of <u>Spartina</u> Vs. Time (Lower Compartment Spiked)

Time (days)	Locality	Roots	Stalks	Leaves
1	A-1	< 1%	-	-
1	B-1	_	-	-
1	C-1	-	-	-
2	A-2	< 1%	-	-
2	В-2		_	-
2	C-2	-	-	—
3	A-3	8%	-	-
3	В-3	1%	-	-
3	C-3	3%		-
4	A-4	7%	-	
4	B-4	2%	-	-
4	C-4	-	-	-
5	A-5	5%	< 1%	< 1%
5	В-5	1%	-	-
5	C-5	-	-	-
8	A-6	8%	1%	<1%
8	В-6	-	-	-
8	C-6	1%	-	-
9	A-7	5%	1%	< 1%
9	B-7	1%	-	-
9	C-7	1%	-	-
11	A-8	19%	1%	1%
11	B-8	-	-	-
11	C-8	-	-	-
12	A-1	6%	-	-
12	B-1	-	80 <b>-</b>	-
12	C-1	1%	_	-
30	A-2	14%	1%	1%
30	В-2	2%	-	-
30	C-2	-		-
45	A-3	6%	1%	1%
45	В-3	1%	-	
45	C-3	1%	-	-
60	A-4	4%	1%	1%
60	В-4	<b></b> ·	. <b></b>	-

## Table 7. % Uptake of $HgCl_2/g$ : Marsh Box Experiment

1.5

Time	Locality	Roots	Stalks	Leaves
(days)		12		
			2011 - 201	300
60	C-4	() <del>.</del>		
90	A-6	2%	1%	1%
90	B-6	2%	1%	1%
90	C-6	-		-
120	A-7	1%		-
120	B-7	1%	-	-
120	C-7	-		-
150	A-8	< 1% 🗈		-
150	В-8	-	-	-
150	C-8		-	-

Table	7	(Continued)
Table	4	(Continueu)

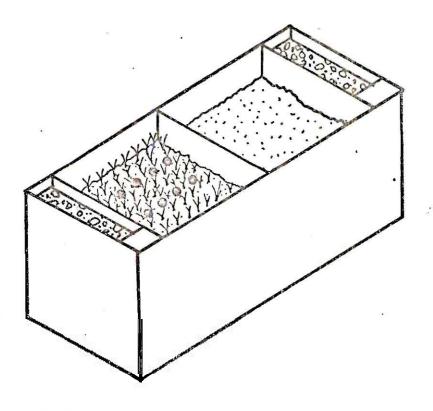


Figure 29. Marsh Box Experiment Injection Sites for Hg<sup>203</sup>. Dots Represent Injection Sites 1-8 (Top Left to Bottom Right). As would be expected, plants nearer to the injection sites had generally the higher uptake values. Root systems, however, are so extensive that a plant 15 cm away from the injection site could have roots near the site. This explains how some plants at a distance of 20 cm from an injection site have a high uptake percentage.

23

.

.

8

٩.

82

.

### CHAPTER IV

### CONCLUSIONS

The importance of <u>Spartina alterniflora</u> in the transfer of mercury in an estuarine-nearshore environment is clearly evident from the above results. The ecological significance of this is implied by results of other studies.

Although mercury can be taken up directly by high trophic level organisms such as finfish (Jernelov, 1970) it is more likely to enter an organism more readily in its food and in turn be transferred through food chains by feeding (Huckabee and Blaylock, 1972) (Jernelov and Lann, 1971) (Knauer and Martin, 1972). In ecosystems like in the Georgia estu**ar**ies, <u>Spartina</u>, being the major primary producer, may represent the most efficient pathway of introduction of mercury into the food chain. Huckabee and Blaylock (1972), in experiments involving a simple food chain composed of green algae (<u>Chlorella</u>) and the freshwater bass (<u>Micropterus</u>) have shown that at least for a one step food chain, mercury accumulation due to consumption of plants is a significant contributor to the body burden of this metal in fish.

Williams and Murdoch (1969) suggested that <u>Spartina</u> is an important agent in the transfer of heavy metals in estuarine food chains. Results of the present study, showing that mercury is taken up rapidly by <u>Spartina</u>, suggests that <u>Spartina</u> indeed plays a significant role in the transfer of this metal.

٠.

The movements of phosphorous through Spartina was studied by Reimold

(1970), who demonstrated the release of phosphorous from leaves to the surrounding water. Although the toxic heavy metal mercury is quite unlike phosphorous, which is essential to plant life, it apparently experiences a similar transport mechanism.

Much of the mercury introduced into the marine environment as a result of industry is in the inorganic form. Mercury methylation can subsequently occur in marine bottom sediments where it is then available to rooted plants. The pathways of Hg and MeHg in <u>Spartina</u> as implied by the results of this study are very similar. The uptake rates of both Hg and MeHg also appear to be quite similar, with the uptake of Hg being consistantly higher by only a small margin. This is contrary to results of experiments on snap beans (<u>Phascolus vulgaris</u>) by Huckabee and Blaylock (1972) who showed that this plant concentrated methylmercury to an order of magnitude higher than inorganic mercury. Physiological differences in root uptake mechanics probably accounts for this variation.

Results generally show that once methylmercury is taken up by <u>Spartina</u> it is transferred to the leaves and stalks more efficiently than inorganic mercury. This being the case, MeHg is more likely to be transferred to higher organisms in Spartina detritus.

The total Hg taken up by <u>Spartina</u> in Georgia estuaries has been estimated to be approximately  $0.7 \text{ mg/m}^2 \text{ yr}$  (Windom, 1973). The basis for this estimate is shown in Table 8. In addition to this amount of mercury taken up by <u>Spartina</u> an additional amount may be taken up which is subsequently lost through the leaves. In fact results of the present study indicated that about 5% of the mer-

# Table 8. Mercury Budget for Georgia EstuariesConcerning Spartina alterniflora

\$3

Input		Uptake by Plants	Release via Leaves
In Solution:	1.5 mg/m <sup>2</sup> yr	$0.7 \text{ mg/m}^2 \text{ yr}$	$35 \text{ug/m}^2 \text{ yr}$
In Suspended Sed.	$2.3 \times 10^{-1} \text{ mg/m}^2 \text{ yr}$		

Based on:	1)	Annual production of Sparting at 700 g/m <sup>2</sup> yr
	2)	Total marsh area at $1.6 \times 10^8$ m <sup>2</sup>

,<sup>e</sup>

2

53

3) Concentration of 0.1 ppb in water, and 0.4 ppm in Sus. Sed.

32

35

.

cury available for uptake by <u>Spartina</u>, at equilibrium, was released to the surrounding water by its leaves. This implies that at least an additional  $35 \,\mu g/m^2$  of mercury can be annually taken up by <u>Spartina</u> to be released by its leaves to surrounding water.

The adsorption of mercury and methylmercury by <u>Spartina</u> detritus is another important influence on the transfer of mercury in an estuarine-nearshore environment. Not only do the live plants accumulate mercury but detritus experiments show that when the plants die the resulting detritus is quite efficient in the absorption of mercury from surrounding waters. It is also able to concentrate mercury from solutions containing environmentally absurd levels (1000 ppb) with equal efficiency.

Results of the present study indicate that there are essentially two processes by which mercury can be conveyed to the food chain via <u>Spartina alter-</u> <u>niflora:</u> 1) the direct uptake of Hg or MeHg from estuarine sediments or surrounding waters, and 2) the direct uptake of mercury by adsorption on plant detritus.

#### BIBLIOGRAPHY

- Bristow, J. M. and Whitcombe, M., 1971. The Role of Roots in the Nutrition of Aquatic Vascular Plants. Amer. J. Bot. 58(1): 8-13.
- Cranston, R. E. and Buckley, D. E., 1972. Mercury Pathways in a River and Estuary. Environmental Science and Technology. 6(3): 274-278.
- Fishbein, L., 1971. Chromatographic and Biological Aspects of Inorganic Mercury. Chromatogr. Rev. 15: 195-238.
- Galtsoff, P.S., 1960. Environmental Requirements of Oysters in Relation to Pollution. <u>IN</u>: Biological Problems in Water Pollution. Trans. 1959. Seminar Robert A. Taft Sanitary Eng. Center Tech. Rept. W60-31: 128-133.
- Glooschenko, W. A., 1969. Accumulation of Hg<sup>203</sup> by the Marine Diatom Chaetoceros costatum. J. Phycol. 5: 224-226.
- Harriss, R. C., White, D. B., and Macfarlane, R. B., 1970. Mercury Compounds Reduce Photosynthesis by Plankton. Science. 170(3959): 736-737.
- Huckabee, J. W. and Blaylock, B. G., 1972. Transfer of Mercury and Cadmium from Terrestrial to Aquatic Ecosystems. <u>IN:</u> CSUI-ANL Symposium, "Role of Metal Ions in Biological Systems". Chicago, Illinois.
- Jernelov, A., 1970. Release of Methylmercury from Sediments with Layers Containing Inorganic Mercury at Different Depths. Limnol. Oceanog. 15: 958-960.
- Jernelov, A. and Lann, H., 1971. Mercury Accumulation in Food Chains. Oikos 22: 403-406.
- Knauer, G. A. and Martin, J. H., 1972. Mercury in a Marine Pelagic Food Chain. Limnology and Oceanography. 17(6): 868-876.

McRoy, C. P. and Barsdate, R. J., 1968. Phosphate Absorption in Eelgrass. 6-13.

.

### BIBLIOGRAPHY (Continued)

- Pomeroy, L. R., and Johannes, R. E., Odum, E. P., Roffman, B., 1969.
  The Phosphorus and Zine Cycles and Productivity of a Salt Marsh.
  412-419. IN: D. J. Nelson and F. C. Evans (Editors) Proc. 2nd
  National Sym. on Radioecology. N.B.S. Springfield, Virginia.
- Reimold, R. J., 1970. The Movement of Phosphorous through the Salt Marsh Cord Grass, Spartina alterniflora. Loisel, ---.
- Saha, J. G., 1972. Significance of Mercury in the Environment. Residue Reviews. 42: 103-163.
- Williams, R. B. and Murdoch, M. B., 1969. The Potential Importance of Spartina alterniflora in Conveying Zinc, Manganese, and Iron into Estuarine Food Chains, pp. 431-439. <u>IN</u>: D.J. Nelson and J. C. Evans (Editors) Proc. 2nd National Sym. on Radioecology. N.B.S. Springfield, Virginia.
- Windom, H. L., 1973. Mercury Distribution in Estuarine-Nearshore Environments. Journal of the Waterways, Harbors and Coastal Engineering Division, ASCE, No. WW2, Proc. Paper 9753. 99: 257-264.

۰.