USE OF MODULAR ESTUARINE MESOCOSMS TO ASSESS THE EFFECTS OF AN ACUTE PESTICIDE EXPOSURE.

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ABSTRACT

The effects of acute endosulfan exposure were assessed in modular estuarine mesocosms. Mesocosm test systems were constructed in a greenhouse at the NOS Charleston Laboratory (CCEHBR). Each system was designed to allow for tidal flux and consisted of four sections that included a stream channel and three elevated marsh components. Sediments, flora, fauna, and seawater were collected from a historically pristine site on Wadmalaw Island, SC and established in the mesocosms. Mesocosm systems were dosed at a range of endosulfan concentrations (0.367, 1.102, to 3.34 ☞g/L) with a control. Endpoints measured included: survival of fish (*Fundulus heteroclitus*), grass shrimp (*Palaemonetes pugio*), oysters (*Crassostrea virginica*) and fiddler crabs (*Uca pugilator*), as well as chlorophyll *a* and nutrient concentrations. Endosulfan exposure had no effect on the survivorship of oysters and only a slight effect on fiddler crab survivorship. Endosulfan was acutely toxic to fish (LC_{50} = 2.2 ☞g/L; 95% CI = 1.9, 2.6 ☞g/L) and grass shrimp (LC_{50} = 1.08 ☞g/L; 95% CI = 0.08, 1.4 ☞g/L). Ammonia concentrations were significantly higher than controls after 96 hours in the highest treatment (3.34 ☞g/L). Six weeks after the exposure, chlorophyll *a* concentrations were significantly lower in all treatments when compared to controls.

INTRODUCTION

Mesocosms have been in use for approximately the last 50 years. The majority of work done with mesocosms within the realm of ecotoxicology has been with freshwater systems. Lauth et al. [2] described a modular estuarine mesocosm that incorporated estuarine flora and fauna with sediment and water in a system that has tidal flux and mixing. The systems were further modified by Pennington et al. (unpubl.) to allow for independent and replicated systems. A series of experiments were performed to test the effectiveness of using these mesocosms in ecotoxicological studies, including assessing direct and indirect effects of acute and chronic pesticide exposure as well as fate and transport modeling. This poster represents a portion of one of the studies conducted with endosulfan, a commonly-used agricultural insecticide. The purpose of this study was to evaluate the use of the modular estuarine mesocosm to examine if tidal creek organisms respond to pesticides in a similar manner to that observed under laboratory conditions.

METHODS

- Mesocosm test systems were constructed in a greenhouse at the NOS Charleston Laboratory (CCEHBR).
- Modular estuarine mesocosms were used to represent an endosulfan field exposure. The design for the mesocosms was modified from Lauth et al. [2]. Rather than the flow through design, a replicated closed system design was employed.
- Sediments, flora, fauna (Table 1), and seawater were collected from Leadenwah Creek, a historically pristine site, on Wadmalaw Island, SC and established in the mesocosms.
- Twelve mesocosm systems were dosed daily at three endosulfan concentrations (0.367, 1.102, to 3.34 ☞g/L) and a control, with three replicates each.
- Endpoints reported in this poster include: 96 hour survival of fish (*Fundulus heteroclitus*), grass shrimp (*Palaemonetes pugio*), oysters (*Crassostrea virginica*) and fiddler crabs (*Uca pugilator*), as well as chlorophyll *a* and nutrient concentrations.
- Statistical treatment comparisons made using ANOVA and Dunnett's procedure for multiple comparison.
- LC_{50}s were calculated using the Trimmed Spearman-Karber approach.

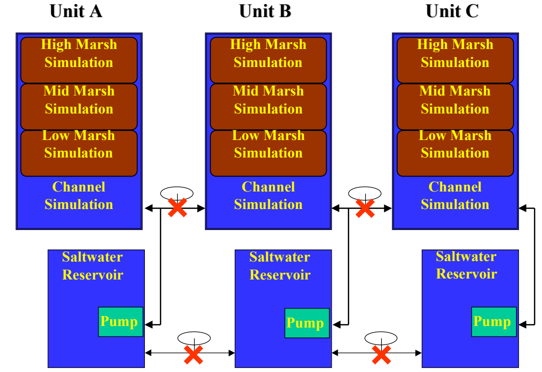
METHODS (Continued)

Table 1: Numbers of animals in each tank at time = 0 hours.

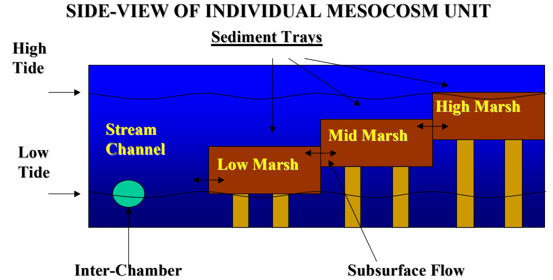
Treatment	Conc.	Rep.	#Fish	#Shrimp	#Oysters	#Crabs
Control	0 (µg/L)	1	10	10	16	10
		2	10	10	17	10
		3	10	10	17	10
Low Dose	0.367 (µg/L)	1	10	10	14	10
		2	10	10	17	10
		3	10	10	12	10
Mid Dose	1.102 (µg/L)	1	10	10	17	10
		2	10	10	15	10
		3	10	10	15	10
High Dose	3.34 (µg/L)	1	10	10	14	10
		2	10	10	13	10
		3	10	10	13	10

- http://www.chbr.noaa.gov/public/MesocosmWebPage.html

TOP-VIEW OF REPLICATED MESOCOSM DESIGN



SIDE-VIEW OF INDIVIDUAL MESOCOSM UNIT



RESULTS

- Endosulfan 96 hour LC_{50} for fish (*F. heteroclitus*):
  - 2.2 ☞g/L; 95% CI = 1.9, 2.6 ☞g/L (Figure 1).
- Endosulfan 96 hour LC_{50} for shrimp (*P. pugio*):
  - 1.08 ☞g/L; 95% CI = 0.08, 1.4 ☞g/L (Figure 2).
- Endosulfan had no effect ($p > 0.05$) on the oyster *C. virginica* (Figure 3).
- Endosulfan had a slight, but non-significant effect ($p > 0.05$) on the crab (*U. pugilator*) at all concentrations tested (Figure 4).
- Community derived endosulfan 96 hour LC_{50} (all four species):
  - 3.2 µg/L; 95% CI = 2.2, 4.8 µg/L
- Chlorophyll *a* concentrations were low and not significantly different between treatments at Time = 0 hours. All treatment tanks (Figure 5) were higher at time = 6 weeks than time = 0 hours, but endosulfan treatments were significantly lower than the controls at time = 6 weeks ($p = 0.005$).
- Nitrate/nitrite (NO_3-NO_2), ammonia (NH_3), and phosphate (PO_4) were not significantly different ($p > 0.05$) between treatments at Time = 0 hours; however, after 96 hours (Figure 6) ammonia was significantly greater ($p = 0.0007$) than the controls at the highest concentration tested.

Figure 1: Survivorship of the Fish *Fundulus heteroclitus* after 96 hours

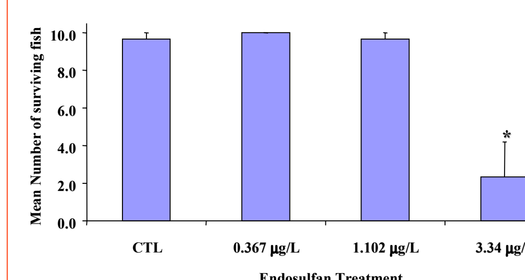


Figure 3: Survivorship of the Oyster *Crassostrea virginica* after 96 hours

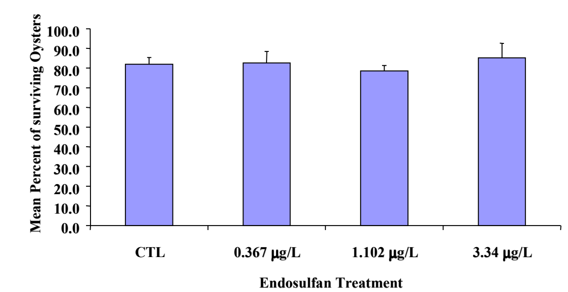


Figure 2: Survivorship of the Grass Shrimp *Palaemonetes pugio* after 96 hours

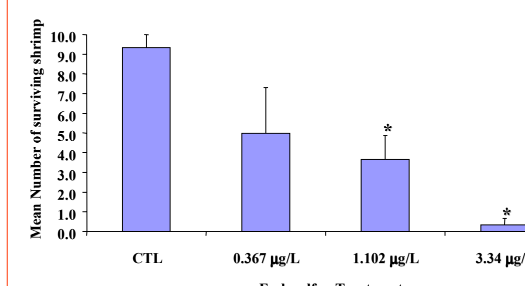


Figure 4: Survivorship of the Fiddler Crab *Uca pugilator* after 96 hours

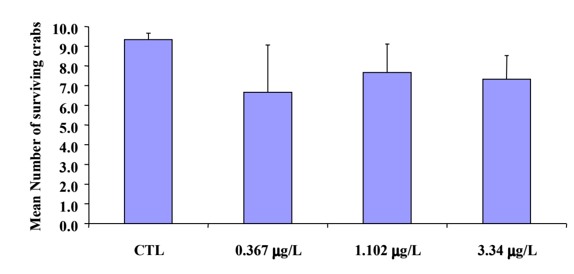


Figure 5 :Chlorophyll *a* Concentration at T = 0 hours and Six Weeks Post-Exposure

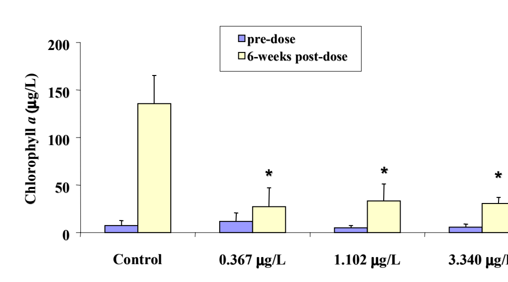
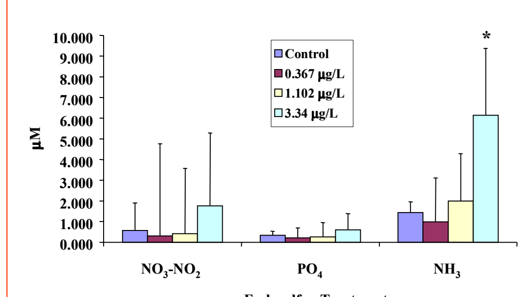


Figure 6: Nutrient Concentrations at T = 96 hours



* = significantly ($p < 0.05$) different from control

CONCLUSIONS

- The acute toxicity of endosulfan to the grass shrimp *P. pugio* in mesocosms was very similar to reported literature values for other types of bioassays.
  - This Study: mesocosm with tidal flux
    1.08 µg/L (95% CI = 0.08, 1.4 ☞g/L)

Reference	Bioassay Type		LC_{50}
[4]	flow-through		1.31 µg/L
[5]	static renewal	high salinity	1.01 µg/L
[5]	static renewal	low salinity	0.62 µg/L
[8]	static renewal		0.62 µg/L
[3]	static		0.657 µg/L
[1]	static renewal		0.25 µg/L
[5]	field derived		0.28 µg/L

- Endosulfan was less toxic to the fish *F. heteroclitus* in mesocosms when compared to reported literature values for laboratory based bioassays.
  - This Study: mesocosm with tidal flux
    2.2 ☞g/L (95% CI = 1.9, 2.6 ☞g/L)

Reference	Bioassay Type	LC_{50}
[7]	static	1.45 µg/L
[6]	static	1.15 µg/L
[5]	field derived	> 0.998 µg/L

- Endosulfan was not acutely toxic to oysters or crabs at the concentrations tested.
- Ammonia levels increased significantly in the highest treatment after 96 hours most likely due to adverse effects imparted to the microbial community.
- Reduced chlorophyll *a* concentrations in all treatments after six weeks (well beyond the 96 hour exposure period) indicated that endosulfan may pose chronic toxicity to microalgae at the concentrations tested.

REFERENCES

1. Baughman DS, Moore DW, Scott GI. 1989. A comparison and evaluation of field and laboratory toxicity tests with fenvalerate on an estuarine crustacean. Environ. Contam. Toxic. 8:417-429.
2. Lauth JR, Scott GI, Cherry DS, Buikema AL. 1996. A Modular Estuarine Mesocosm. Environ Toxicol Chem 15(5):630-637.
3. Moore DW. 1988. An integrated laboratory and field study of nonpoint source agricultural pesticide runoff and its effect on the grass shrimp *Palaemonetes pugio* (holthius). Doctoral Dissertation: University of South Carolina,
4. Schimmel SC, Patrick JM, Wilson AJ. 1977. Acute toxicity to and bioconcentration of endosulfan by estuarine animals. Aquatic Toxicology and Hazard Evaluation, ASTM STP 634, FL Mayer and JL Hamelink (eds.), American Society for Testing and Materials, 1977. Pp. 241-252.
5. Scott GI, Fulton MH, Crosby M, Key PB, Daugomah JW, Waldren J, Strozier ES, Louden C, Chandler GT, Bidleman TF, Jackson K, Hampton T, Huffman T, Schulz A, and Bradford M. 1992. Agricultural insecticide runoff effects on estuarine organisms; correlating laboratory and field toxicity test, ecophysiology bioassays and ecotoxicological biomonitoring (Final Report). U.S. EPA, Gulf Breeze, FL.
6. Trim A. 1985. The acute toxicity of insecticides present in non-point source, agricultural runoff on the mummichog, *Fundulus heteroclitus*, with an emphasis on the sublethal effects of endosulfan. Master's Thesis: University of South Carolina.
7. Williams L. 1988. A comparative toxicological assessment of several agricultural and mosquito control insecticides with an emphasis on time/dose relationships, low slinity and sublethal effects in the mummichog, *Fundulus heteroclitus.* Master's Thesis: University of South Carolina.
8. Wirth EF, Lund SA, Fulton MH, Scott GI. (submitted). Determination of acute mortality in adults and sublethal embryo responses of *Palaemonetes pugio* to endosulfan and methoprene exposure. Aquatic Toxicology.

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