

Developmental and Behavioral Changes of the White Shrimp *Litopenaeus setiferus*

Under the Exposure of Phenylpyrazole Fipronil

Ali A. Al-Badran*, Masami Fujiwara**

Department of Fisheries and Marine Resources, College of Agriculture, University of Basrah, Basrah, Iraq*
Department of Wildlife and Fisheries Sciences, Texas A&M University, College Station, TX, U.S.A.**

* aliabdulameer33@gmail.com, ** fujiwara@tamu.edu



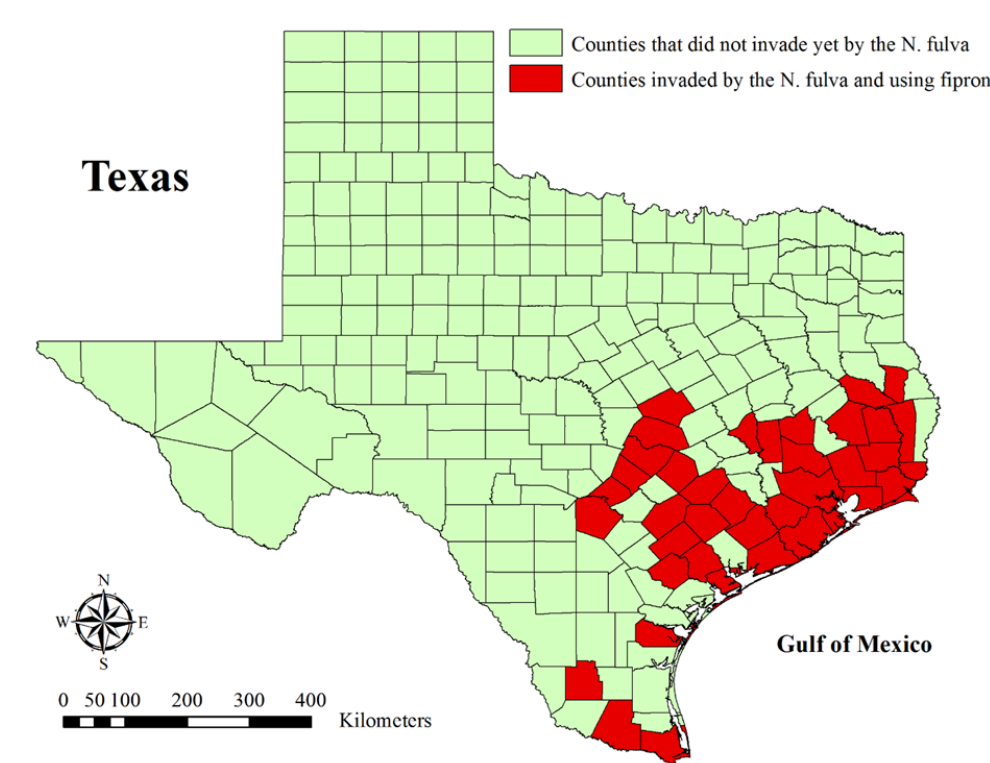
Abstract

Chemical pesticides are commonly used world-wide, and they can flow into estuaries and affect non-targeted organisms. We evaluated the effects of six concentrations of the phenylpyrazole fipronil (0.0, 0.005, 0.01, 0.1, 1.0, and 3.0 $\mu\text{g/L}$), which were previously observed in the environment, on white shrimp *Litopenaeus setiferus* (initially averaging 0.80 ± 0.08 g/shrimp). Compared with the control, survivorship of shrimp over 45 days declined significantly at the higher concentration treatments. Growth was affected at all concentrations, and the percent weight gain decreased significantly. Inter-molt intervals were longer in all treatments. Changes in swimming and feeding behavior of shrimp were observed under all treatments, and change in body color was observed at higher concentration treatments. Lipid content in shrimp decreased significantly while ash content increased with fipronil concentration. Fipronil adversely affected white shrimp under the concentrations observed in the environment and monitoring of fipronil use is needed in coastal areas.

Introduction

- A rapid increase in pesticides use in recent years has resulted in enormous pressure on the ecosystems.
- Pesticides are expected to have a much higher effect on the aquatic environments.
- The majority of ecotoxicological studies focus on a few selected model organisms and neglecting effects on other non-targeted organisms.
- In 2016, the United States Environmental Protection Agency (U.S. EPA) issued a quarantine exemption to the Texas Department of Agriculture, allowing the expanded use of fipronil in southeastern counties of Texas to control tawny crazy ants *Nylanderia fulva*.

- Fipronil can flow into creeks, rivers, and estuaries because it is mobile in soils and soluble in water.
- Fipronil concentrations in some aquatic environments reached up to $12.6 \mu\text{g/L}$, exceeding its acute level ($0.1 \mu\text{g/L}$) in the aquatic life benchmark of the U.S. EPA.
- L. setiferus* is one of the most important commercial fishery species in the U.S.
- They are estuarine-dependent during a juvenile stage; this potentially exposes them to pesticides that are used on land.

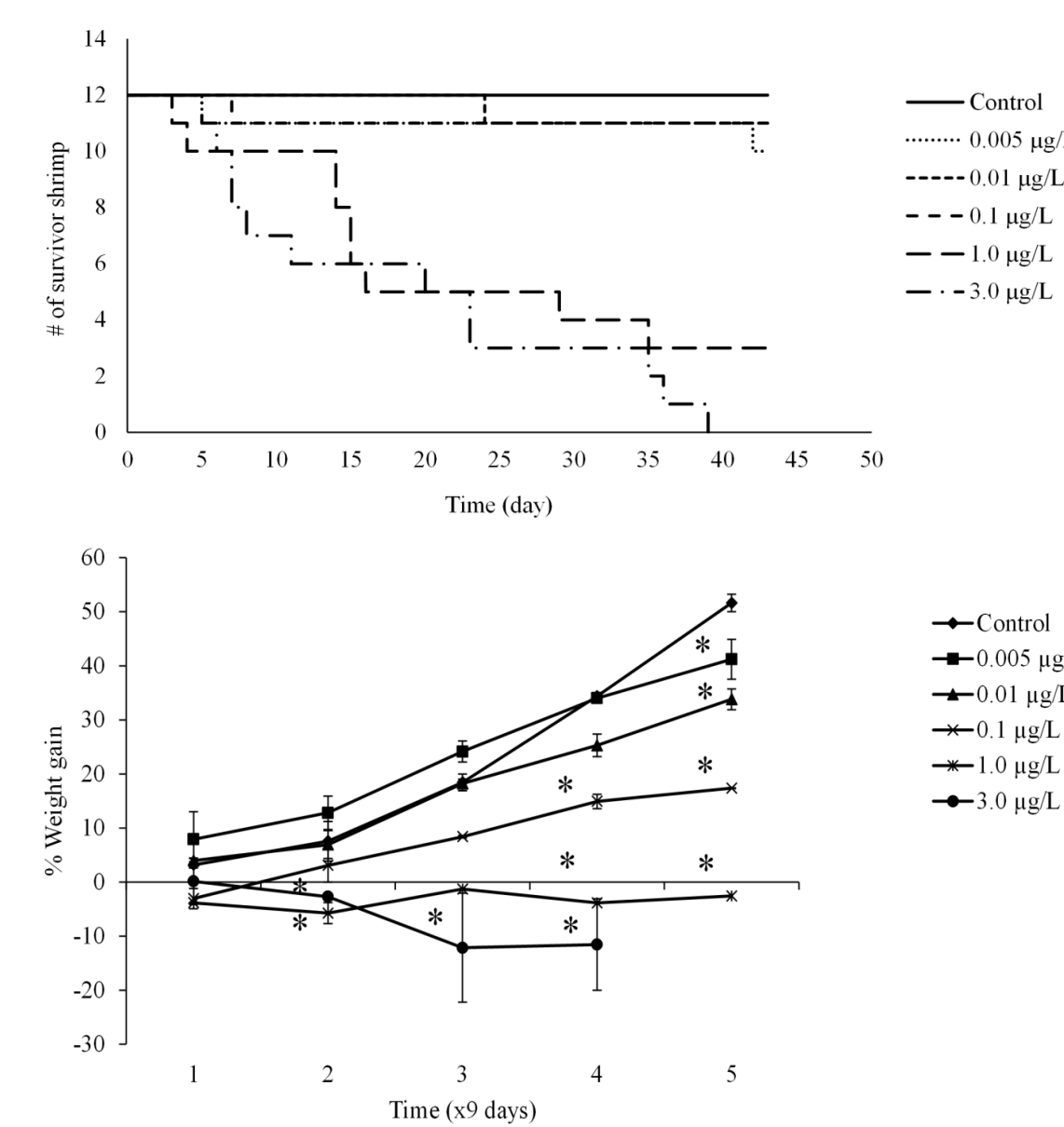


Map of Texas counties. Red color indicate the counties that have been invaded by the tawny crazy ants *N. fulva* and that are using fipronil based on the exemption issued by the U.S. EPA in 2016 to control the invasion.

Objective

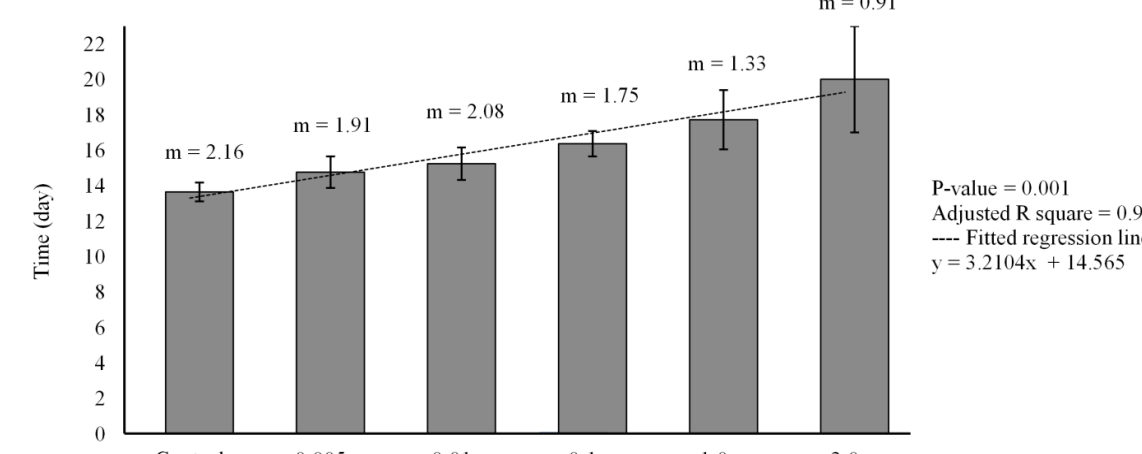
- To investigate lethal and sub-lethal effects of the environmental-related levels of fipronil on the White Shrimp *L. setiferus* using multiple developmental and behavioral endpoints.

Results

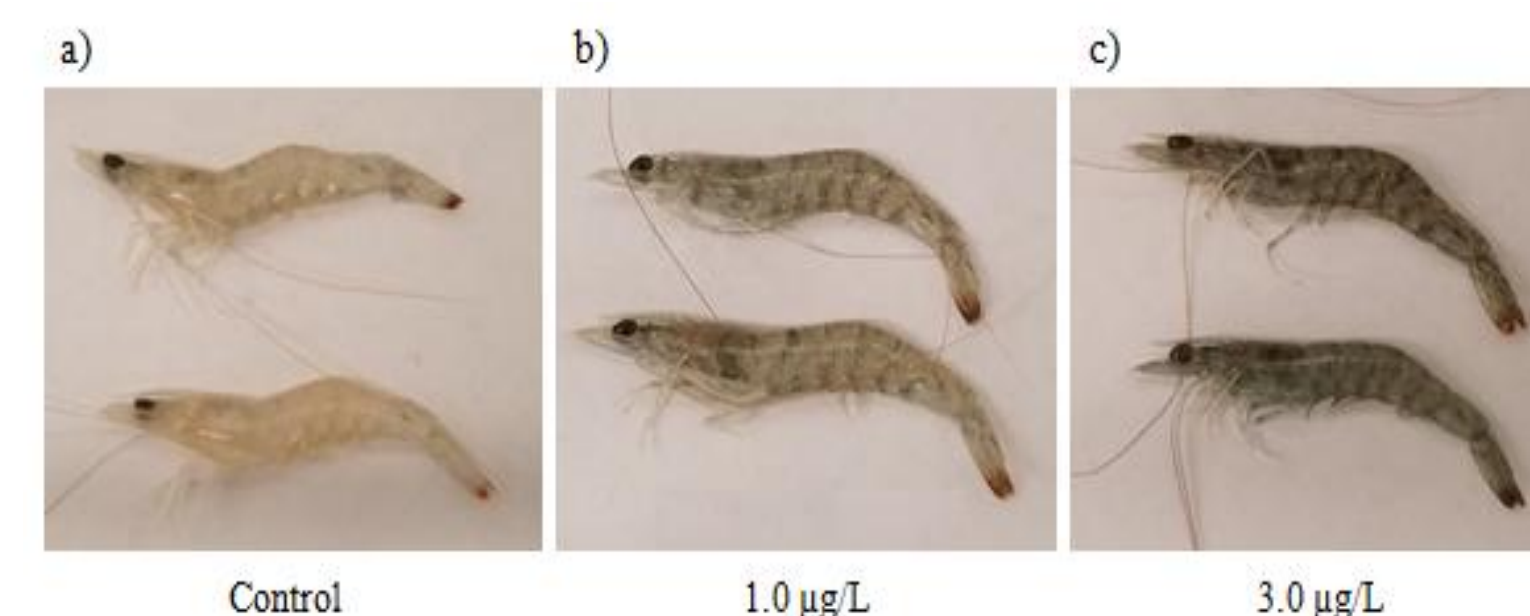


Kaplan-Meier survivorship curves of juvenile white shrimp during 45 days of exposure. Day 1 is 24-h after the beginning of the experiment. Treatments of higher concentrations of fipronil were significantly different from the control (Non-parametric Log-Rank test, $P < 0.0001$).

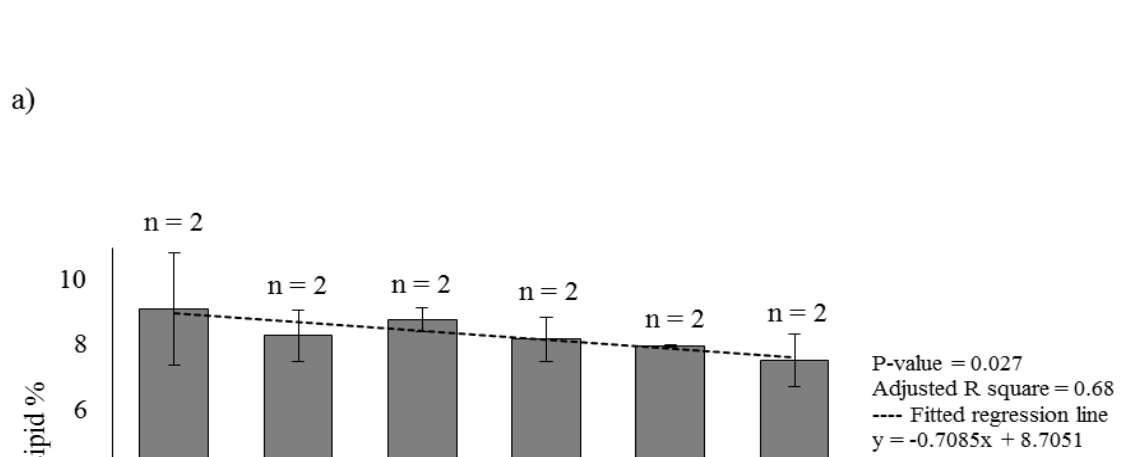
Percent weight gain (per individual) of juvenile white shrimp at multiple time periods (each 9 days). Stars above the curve indicate that fipronil treatment was significantly different from the control at the time of measurement. All treatments were significantly different from the control by the end of the experiment (ANOVA, $P < 0.05$).



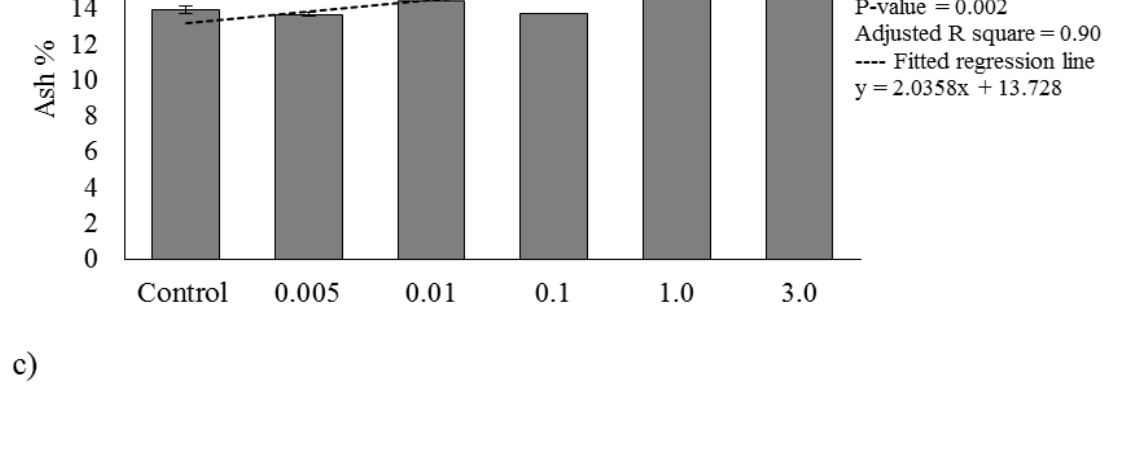
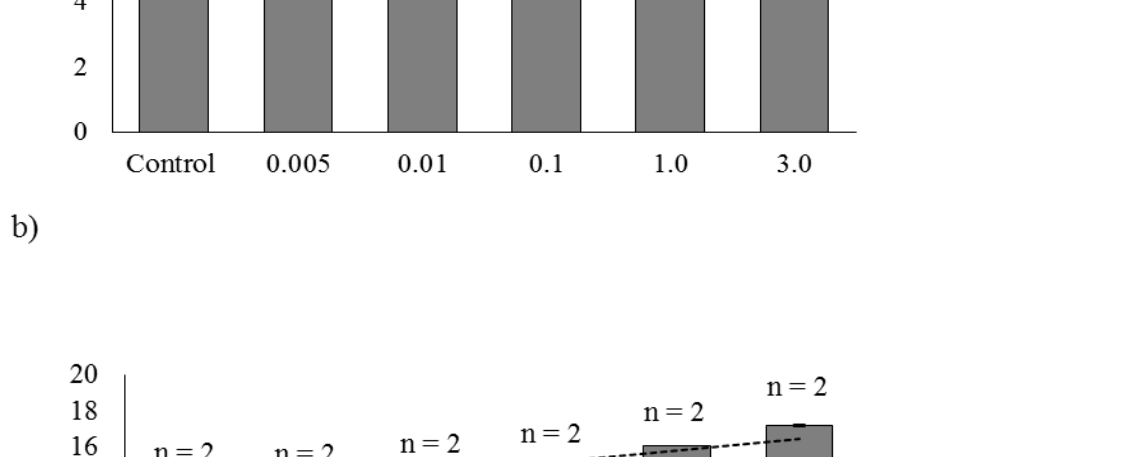
Inter-molt intervals of juvenile white shrimp. m is the average number of molts of individual shrimp in each treatment. Regression analysis showed that inter-molt intervals increased significantly with fipronil concentration ($P = 0.001$).



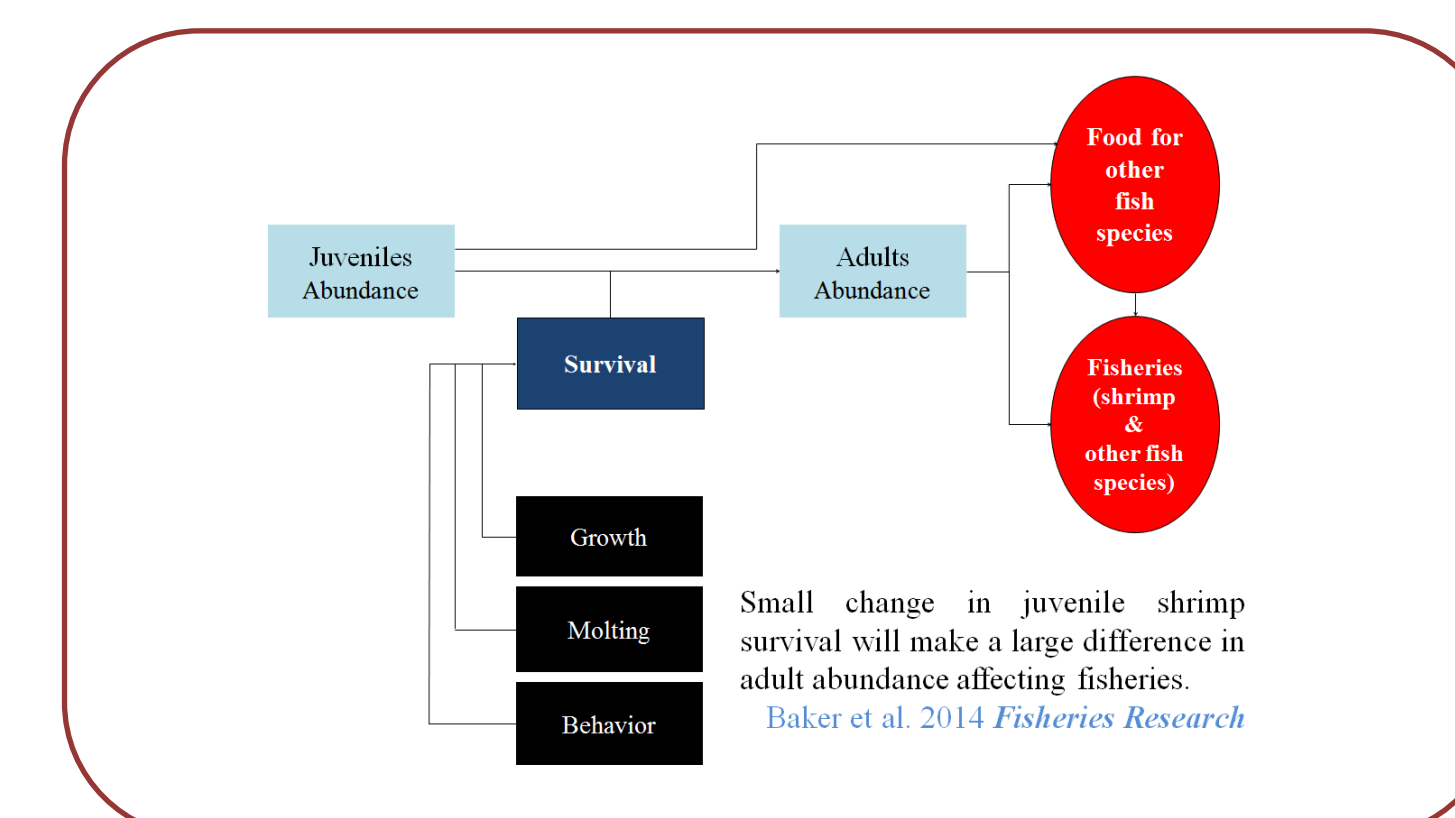
Changes of body color of juvenile white shrimp exposed to the highest concentrations of fipronil ($\mu\text{g/L}$) during the experiment compared to the control. (a) Control $0.0 \mu\text{g/L}$ fipronil, (b) $1.0 \mu\text{g/L}$ fipronil, (c) $3.0 \mu\text{g/L}$ fipronil.



Chemical analysis of body composition of juvenile shrimp. Dashed lines are fitted regression lines. Regression analysis showed that lipid content in shrimp decreased significantly with increased concentration of fipronil ($P = 0.027$), while ash content increased significantly ($P = 0.002$). Error bars indicate the standard errors (n = number of samples analyzed from each treatment).



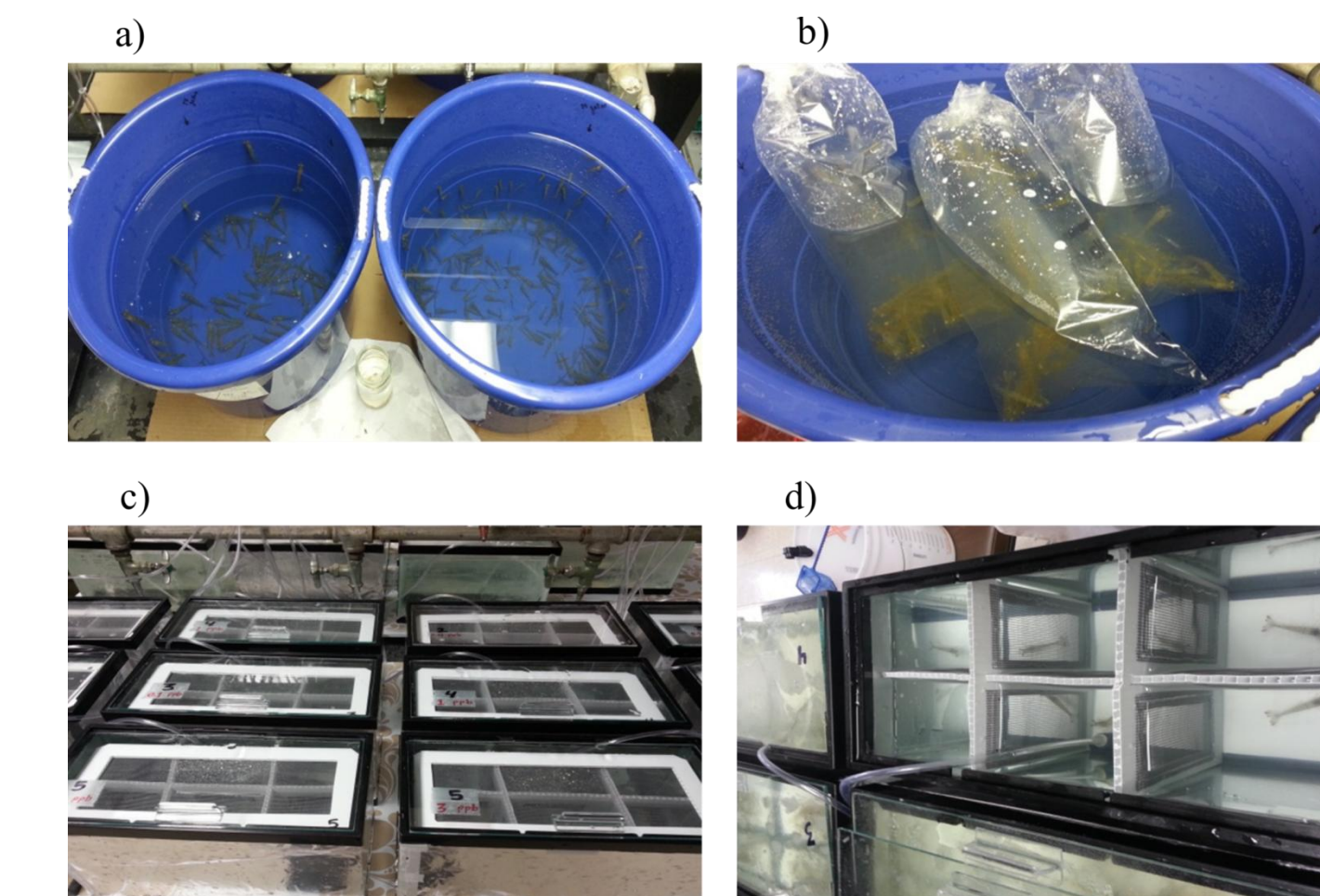
Management and Ecological Implications



We thank Galveston Laboratory of the U.S. National Marine Fisheries Service, especially Mr. Shawn Hillen, for providing shrimp for the experiment. Thanks to the Office of Graduate and Professional Studies at Texas A&M University for the dissertation fellowship awarded to the student while writing this paper. We would also like to thank Drs. Delbert M. Gatlin III, Miguel Mora, and David Wells, who provided comments on the proposal for conducting this research.

Methods

- Juvenile shrimp collected from Galveston Bay, Texas.
- The experimental system consisted of 12 glass aquariums of 9.5 liter.
- Each aquarium was divided equally into six cells.
- Shrimp were fed twice daily.
- Dissolved oxygen (mg/L), salinity (‰), temperature ($^{\circ}\text{C}$), and pH were measured.



Acclimation tanks and experimental system (a) large tanks used for water temperature equilibration and shrimp acclimation before starting the trials; (b) procedure used for moving shrimp to tanks of prepared brackish water for acclimation to laboratory conditions; (c) experimental system, glass aquariums covered with aluminum foil sheets and glass lids; (d) aquariums divided into six separate cells.

Experimental measurements:

- Survival
- Weight gain and growth rate
- Inter-molt intervals
- Behavioral and physical changes
- Analysis of whole-body composition (dry matter, protein, lipids, and ash)

Conclusion

- Environmental-related concentrations of fipronil caused both lethal (acute) and sub-lethal (chronic) effects on *L. setiferus* juveniles.
- Fipronil adversely affected the growth, behavior, and body composition of *L. setiferus* even below the chronic level of the U.S. EPA aquatic life benchmarks.
- Because of the detection of fipronil in estuarine waters and expected increased use of fipronil in areas adjacent to estuarine and coastal areas in the U.S., we recommend the following:

- monitoring fipronil concentration around the coastal regions in and out of the U.S.A.,
- trying to limit the use of fipronil during the peak periods of shrimp migration to estuaries,
- investigating the effects of fipronil on different penaeid species in other countries that are using fipronil,
- conducting further studies of the effects of fipronil and its major metabolites on other non-target organisms using concentrations below chronic levels.

Acknowledgement