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PROJECT TITLE: Inhibition of reproductive processes in tadpole shrimp using liposome carriers: Potential decrease in future damage to rice crops

Project Leader:

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OBJECTIVES AND EXPERIMENTS CONDUCTED BY LOCATION TO ACCOMPLISH OBJECTIVES:

- 1) Complete the determination of the efficient order of ingredients for MF-liposome pellets;
- 2) Determine the effective range of doses of liposome pellets that reduce tadpole shrimp fecundity;
- 3) Complete the assessment of tadpole shrimp capacity to synthesize MF;
- 4) Assess the reproductive productivity of tadpole shrimp offspring treated with MF.

SUMMARY OF 2006 RESEARCH (MAJOR ACCOMPLISHMENTS) BY OBJECTIVE:

1) Complete the determination of the efficient order of ingredients for MF-liposome pellets

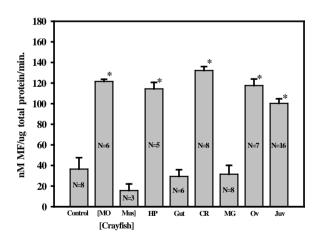
In previous experiments, we demonstrated inhibitory reproductive impacts from MF-liposome pellets. To prevent the oxidation of MF, it was incorporated into liposomes. These liposomes, at concentrations of 0.001% and 0.0001% MF (by weight), were blended into a protein mixture similar to that used above. These pellets contain a standard crustacean feed mixture of casein, lecithin, wheat gluten and albumin. We tested previous pellets and determined that they contained significantly less MF than previously thought. For instance, 10 $\mu g/g$ MF pellets contained 3.8 $\mu g/g$ MF, 1 $\mu g/g$ pellets contained 0.75 $\mu g/g$ MF, and 100 ng/g pellets MF content was below detection limits or 0.1 $\mu g/g$ MF. This information shows us that the MF is affective at lower concentrations than previously thought.

2) Methyl Determine the effective range of doses of liposome pellets that reduce tadpole shrimp fecundity

We have started preliminary studies looking at the inhibitory effects of these new pellets under laboratory conditions. Completion of pellet synthesis came after the rice growing season. We have had some trouble identifying soil with tadpole shrimp cysts, thus studies examining the effectiveness of these MF liposome pellets has been difficult.

3) Complete the assessment of tadpole shrimp capacity to synthesize MF

We have determined that MF is a native compound of *Triops longicaudatus* (Nelson, 2005). Biochemical analysis of the synthetic capacity of the tadpole shrimp indicate that the hepatopancreas (HP), cephalic region (CR), and ovarian (Ov) tissue homogenates, and whole body homogenates of juveniles of the tadpole shrimp are capable of synthesizing MF (Fig. 1).



[Figure 1. Radiochemical Synthesis Assay for Farnesoic Acid O-Methyl Transferase (FAOMeT) Activity in *T. longicaudatus*. Hepatopancreas (HP), cephalic region (CR), and ovarian (Ov) tissue homogenates, and juveniles whole body homogenates, incubated in farnesoic acid and [³H-methyl] SAM indicated MF synthetic capacity. Gut and mandibular gland had no activity. Crayfish (*P. clarkii*) mandibular organ (MO) was used as a positive control and muscle as a negative control.]

4) Assess the reproductive productivity of tadpole shrimp offspring treated with MF

This objective was also delayed, as we are trying to identify soil with significant tadpole shrimp cysts. These are currently ongoing studies within the laboratory.

PUBLICATIONS OR REPORTS:

Tsukimura, B., W.K. Nelson and C.J. Linder. 2006. Inhibition of ovarian development in by methyl farnesoate in the tadpole shrimp, *Triops longicaudatus*. Comp. Biochem. Physiol. A 144:135-144.

Presentations

Tsukimura, B., and W.K. Nelson. 2006. Ovarian Development Inhibition by Methyl Farnesoate in the Tadpole Shrimp, *Triops longicaudatus*. Central California Agricultural BioTechnology Conference. Fish Camp, CA., March, 2006.

CONCISE GENERAL SUMMARY OF CURRENT YEAR 'S RESULTS:

As the potential for increased copper sulfate regulation looms, we are developing a method of treating tadpole shrimp using an organic hormone, methyl farnesoate. This terpenoid compound has been successfully incorporated into liposomes, which protects MF from oxidation. We have developed a methodology that protects the liposomes during the production of the MF-liposome pellets. Over the last year we have succeeded in keeping MF within the pellets. These studies came after the rice growing season, and we hope to test these pellets in the upcoming 2007 growing season. These pellets have been shown to be effective inhibitors of gonad development in juveniles, both in preliminary studies in the laboratory. Data from earlier studies have been successfully published in a refereed journal. Support from the Rice Research Board is used as matching funds for my grant with California Agriculture and Technology Institute – Agricultural Research Initiative.