ANNUAL REPORT
COMPREHENSIVE RESEARCH ON RICE
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PROJECT TITLE: Effective control of tadpole shrimp damage to rice yield: Ground application of copper sulfate and methyl farnesoate-laden pellets

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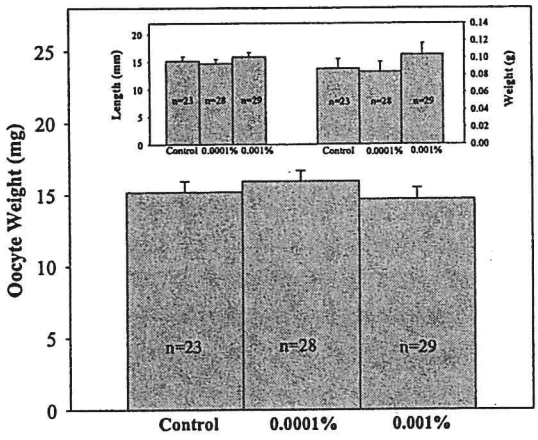
OBJECTIVES AND EXPERIMENTS CONDUCTED BY LOCATION TO ACCOMPLISH OBJECTIVES:

1) Determine the effect of methyl farnesoate pellets on the inhibition of tadpole shrimp reproduction in the field.

2) Determine the efficacy of ground application of copper sulfate on lowering the damage caused by tadpole shrimp in rice yields. Determine the potential cost savings of ground application.

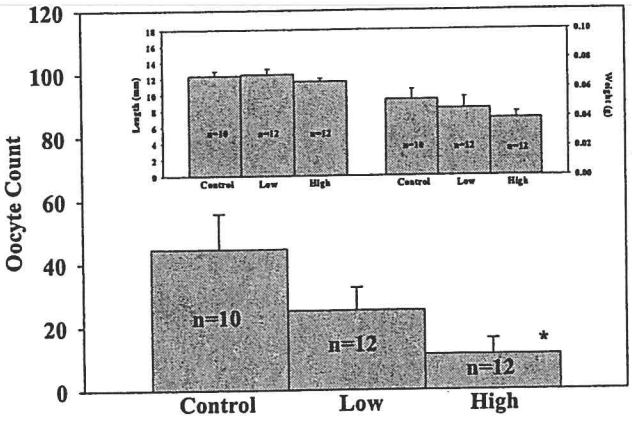
SUMMARY OF 2001 RESEARCH (MAJOR ACCOMPLISHMENTS) BY OBJECTIVE:

1) In an effort to develop a MF-pellet that required one application, we constructed pellets in which MF was directly incorporated into the protein components of the pellets. MF concentrations (0.0001% and 0.001%), shown to be effective in previous trials, were mixed into a protein (lecithin, wheat gluten and casein) mixture, at the Department of Animal Sciences, UC Davis, with the assistance of Dr. E. Chang, Bodega Marine Laboratory, Animal Science Department. These MF-laden pellets had no effect on ovary weight at either day 5 or day 10, whether in the laboratory or in the field (Fig. 1). MF is an organic compound that is readily labile and easily oxidized. These data suggest that the MF should be protected when introduced into pellet form. Because of the labile and oxidation properties, these data also indicate that MF is a good candidate for organic control of tadpole shrimp.



[Figure 1. MF-Laden Pellets Do Not Affect Ovary Weight of *Triops longicaudatus* – In laboratory trials, MF mixed directly into protein mixture has no effect on ovary weight at Day 10. No differences were detected in somatic factors (length and weight) of tadpole shrimp. The absence of effect were similar for Day 5 and for field trials. **Inset**: MF-laden pellets had no effect on somatic indices, length or weight.]

In previous experiments, we demonstrated inhibitory reproductive impacts from MF-coated pellets, but the direct incorporation of MF into pellets was no better than controls. To prevent the possible oxidation of MF, the terpenoid was incorporated into liposomes (Sigma). 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 lecithin, wheat gluten and casein. This feed mixture was consumed in previous and current assays and attracts the carnivorous tadpole shrimp. As the tadpole shrimp devour the pellets, they also consume the liposomes laden with MF. Preliminary data in laboratory tests suggest that this feed effectively reduced the number of oocytes found in developing tadpole shrimp (Fig 2.). We will assay the efficacy of the MF-liposome pellets in the field. Data will be collected and analyzed in the same manner.



[Figure 2. MF-Liposome Pellets Reduce Oocytes in Tadpole Shrimp at Day 5 – In the laboratory, preliminary data indicates that MF-liposome pellets effectively decrease the number of oocytes in tadpole shrimp ovaries. "Low" concentration refers to 0.0001% and "High" refers to 0.001% MF in pellets. * indicates significant difference from control. **Inset**: MF-laden pellets had no effect on somatic indices, length or weight.]

The efficacy of the pellets uses relatively low amounts of MF (0.0001% and 0.001%). We hope to develop pellets that contain micelles of MF, which may provide longer lasting pellets that we could test in the field. In the coming year, we plan to test the hypothesis that pellets laden with MF-filled liposomes will inhibit reproductive processes in tadpole shrimp. The benefit of this inhibitory method is that offspring would not be produced to infect crops in future years. An additional benefit is that MF is an "organic" compound (extracted from plants) that rapidly decomposes when exposed to air. Thus, there are limited projected difficulties associated with regulatory agencies.

Field-testing of the MF-liposome was impaired by the absence of tadpole shrimp in the checks that we established the random block design treatment scheme. The absence of tadpole shrimp the check and in the control rings made difficult the attempt to establish any effect of the MF-liposome pellets. Water temperatures dropped into lethal levels preventing the presence of tadpole shrimp in this check. See arguments below. Water levels in our checks were not stable, carrying from 3-8" in depth. Water managers had difficulty in maintaining constant water heights for our experiments. This was particularly the case for first check where the water entered the field. Future experiments will be run further downstream from the inlet check where water temperature and flow speed are less variable on a day to day basis.

2) Ground application of Bluestone

Random block design was used to assign treatments to 16 rings placed around the checks. Ground application of copper sulfate was performed using appropriate protective gear after the rings were in place. Water temperatures in the check varied dramatically throughout the initial period of the test. During the first 5 days after the initial inundation, water temperatures varied dramatically: June 5 - 24°C; June 7 - 27°C; June 9 - 16°C. Temperatures ranging from 24°C to 30°C are optimal for hatching tadpole shrimp (Riley and Tsukimura, 1998). Colder temperatures, such as 16°C, are reported to be lethal to tadpole shrimp (Fry and Mulla, 1992). Thus, the sudden temperature drop may have eliminated all the tadpole shrimp so that the field used was effectively rid of tadpole shrimp through water temperature control. Because of the absence of tadpole shrimp, no usable data was obtained. This problem can be avoided in the future by using multiple checks (or fields).

PUBLICATIONS OR REPORTS:

Linder, C.J. and B. Tsukimura. Methyl farnesoate inhibits ovarian development in the tadpole shrimp, *Triops longicaudatus*. (in prep)

Presentations

Nelson, W.K., C.J. Linder, and B. Tsukimura. 2002. Effect of methyl farnesoate incorporated into feed on the development of tadpole shrimp, *Triops longicaudatus*. At the Annual meeting of SICB, Anaheim, 2002.

Nelson, W.K., J. Rose and B. Tsukimura. 2002. Reproductive Inhibition By Methyl Farnesoate in the Tadpole Shrimp *Triops longicaudatus*. 83rd Annual Pacific Division meeting of the American Association for the Advancement of Science.

Nelson, W. K. & B. Tsukimura. 2002. Effects of methyl farnesoate on the tadpole shrimp, *Triops longicaudatus*. 23rd Annual Central California Research Symposium.

CONCISE GENERAL SUMMARY OF CURRENT YEAR' S RESULTS:

Methyl farnesoate (MF), when coated to the exterior of pellets, was previously found to inhibit tadpole shrimp reproduction. MF, in concentrations of 0.001% and 0.0001% (by weight) was directly incorporated into protein pellets. These pellets, either in the laboratory or in the field had no effect on tadpole shrimp reproduction. The assumption was that the MF, being a labile and easily oxidized terpenoid, was lost in the production and storage of the pellets prior to application in the experiments. In an additional effort to administer MF to tadpole shrimp, a MF-liposome pellet was developed in which MF, at concentrations of 0.001% and 0.0001% MF (by weight), was incorporated into liposomes. These liposomes, were blended into a protein mixture and formed into pellets. Preliminary data in laboratory tests suggest that this feed effectively reduced the number of oocytes (17 and 28, respectively) found in developing tadpole shrimp when compared to controls (x=43 oocytes). Future tests will assay the efficacy of the MF-liposome pellets in the field. Field tests for both ground application of bluestone and MF-liposome pellets did not produce significant results as none of the fields used for these assays contained tadpole shrimp. Future assays will be conducted on multiple checks and fields to reduce the likelihood of temperature or other catastrophic events from eliminating tadpole shrimp from the entire field.