Investigation into development and preparation of 'artificial' food particles for tropical rock lobster, *Panulirus ornatus*, phyllosoma.

Thesis submitted by

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A late stage phyllosoma of the tropical rock lobster, *Panulirus ornatus*.

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Abstract

This study investigated development of an 'artificial' inert food for tropical rock lobster, *Panulirus ornatus*, phyllosoma, of similar consistency to their natural diet of soft-bodied organisms. Aquaculture hatcheries worldwide rely either partially or totally on production of live foods to rear the larvae of target species. The cultivation of live foods still remains expensive, often unpredictable and, from a nutritional perspective, they maybe sub-optimal. Suitable foods for the culture of rock lobster phyllosoma include *Artemia* during early stages and, mussel gonad and other live zooplankton for later stages of phyllosoma. However, some problems associated with water quality and with tangling of the spiky pereipods of phyllosoma on large fleshy prey, have been reported. Developing an inert food particle could potentially enhance survival and nutrient delivery. Inert food particles have been trialed successfully for a variety of larval crustaceans including penaeid prawns, crabs and freshwater prawns.

Production of a moist microbound diet (MBD) particle using the atomisation method was successfully scaled-up during this study from hand-held glass atomiser to a spraying system capable of producing 1-1.3 kg of moist MBD within 5 minutes. The preferred size ranges of food particles for rock lobster phyllosoma stage 1-3 was 355 – 500 μm and for stages 3-5 were 500 – 850 μm. Size ranges of MBD produced during the study were between 100 and >850 μm, under the conditions trialed however, the size distribution of resulting MBD was broad.

Techniques for preparing multi-walled microcapsules (MC), to encapsulate a broad range of dietary ingredients, were assessed during this study based on similar methods developed by the biomedical and pharmaceutical industries. Multi-walled particles prepared during this study were between 0.5 - 4 mm in diameter. Multi-walled MC allow greater retention of valuable dietary material such as low molecular weight water-soluble nutrients, and allowed for more effective delivery of nutrients to the target species.

The technique for evaluating ingestion of MBD by early *P. ornatus* phyllosoma, stages was determined using a dietary radioisotope tracer (^{14}C). The quantity of ingested MBD significantly increased with progressive stages and ingestion period (up to 4 h). Consumption of MBD by stages 1 and 2 phyllosoma was very low however, consumption by stages 3 and 4 phyllosoma was significantly higher and they consumed 3.6 μg larvae^{-1} and 2.75 μg larvae^{-1} after 4 h ingestion duration, respectively. Stage 4 phyllosoma could potentially be the age at which inert food particles are accepted by phyllosoma however, further research is required to confirm this.

Low ingestion rates of inert food particles are characteristic of the larvae of many marine species, with the exception of penaeid prawns. Feeding strategies of other crustacean larvae, including phyllosoma, involve a series of processes detection (chemical, mechanical and/or visual), seeking and capture strategies to successfully capture prey. Phyllosoma are attracted to the swimming action of *Artemia,* and capture and extract nutrients from them. Ingestion and assimilation of ^{14}C-labelled live *Artemia* metanauplii (1.5 day old) was determined for stage 1 phyllosoma, over a

4 h period. After 4 h, stage 1 phyllosoma consumed approximately one (1.5 day old) *Artemia* metanauplus (8.282 ± 0.501 μg larvae^{-1}), when supplied at a density 1 mL^{-1}. The highest assimilation of live *Artemia* occurred during the 2 h and 3 h feeding periods 0.62 ± 0.21 or 5.31 ± 1.84 μg larvae^{-1} and 0.63 ± 0.343 or 5.35 ± 2.9 μg larvae^{-1} (mean ± SE), respectively; however, these did not differ significantly ($P>0.05$). The second and third hour of feeding also had the highest assimilation efficiency of *Artemia* by stage 1 phyllosoma.

Substitution of live feeds with MBD was investigated for phyllosoma stages 1 and 3. Survival of early stage phyllosoma fed *Artemia* (1 metanauplus mL^{-1}) was shown to be greater than that of phyllosoma fed combinations of *Artemia* and MBD or fed MBD alone. However, both stage 1 and stage 3 phyllosoma fed entirely on MBD survived longer than unfed phyllosoma.

The results of this study allowed a suggested feeding protocol for *P. ornatus* phyllosoma to be proposed using both *Artemia* and inert food particles. Newly hatched *Artemia* should be fed to stage 1 and 2 phyllosoma, while larger metanauplii (500-3500 μm) are suitable in size for later phyllosoma stages. The possible weaning process or introduction of inert food particles could begin at the earliest with stage 4 phyllosoma. Apart from major cost savings associated with reduced live food use, delivery of an adequate formulated food would increase survival of phyllosoma through their long and relative complex larval life. As the nutritional composition of inert food particles can accurately be manipulated, the opportunity to determine the nutritional requirements of phyllosoma could then be achieved, at least for phyllosoma stages which consume such particles.

This study has provided valuable new information relating to the feeding biology and nutrition of *P. ornatus* phyllosoma. The results will allow further development towards production of appropriate hatchery foods for *P. ornatus* phyllosoma and facilitate progress towards more efficient hatchery techniques for this species.

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