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### Initiative Area "LARVI" - Larviculture and Juvenile Production of Fish and Shellfish

Introduced by Björn Myrseth, CEO Marine Farms ASA, Norway at the 1st EATP Stakeholder meeting in Brussels on March 22, 2007

Over the past two decades Europe has contributed to significant developments in the industrial production of fry, seed and post-larvae of different fish and shellfish species. Successful introduction of hatchery techniques was at the origin of several industrial successes with sea bream, sea bass, turbot, halibut, cod, etc. In the Mediterranean for example, hatchery production of bass and bream is expected to reach the turning point of 1 billion fry in 2007.

The fish and shellfish larviculture industry in Europe is valued at several hundred million Euro. This sector is expanding very rapidly on other continents often with European products and technology.

Although profitable today, the marine fish hatchery industry is open for much improvement and increased profitability:

- better understanding of brood-fish development is vital in order to produce eggs of the highest quality, equal to eggs produced in nature; i.e. basic understanding of environmental factors (light, space, temperature and water quality) as well as nutritional requirements and hormone regulation; large scale research facilities and close cooperation between industry and research are required; in addition to improving rearing of present species it would open up R&D possibilities for new species
- survival rates from egg to fry for stocking are mostly under 20 % and still very variable, which means that an increase in survival up to and beyond 50 % could have significant effects on production costs
- egg quality is very variable and remains difficult to assess, as a result of which hatchery outputs are often unpredictable and/or need to be aborted shortly after larval stocking in the hatchery
- live food still plays a critical role and furthermore is cause of concerns with regard to availability, nutritional quality and stability, microbial contamination, production cost, etc. The long-term goals should be complete substitution of live food with dry feed, first of Artemia followed by substitution of rotifers
- optimisation of the microbial environment to the benefit of the larvae
- fry quality is much more a concern than ever before as deformities, stress sensitivity and other defects compromise later production yields as well as market value of the end product
- limited knowledge of the onset and dynamics of the larval digestive physiology hamper further improvements in feeding regimes
- many disease problems cannot be treated adequately because of incomplete understanding of the onset and dynamics of the larval immune system(s)
- limited exploration so far to increase automation, process control and intensification of production methods and use of rearing tanks/systems
- reduction of energy consumption in the larviculture and juvenile production
- the nursery phase requires more attention as automation and improvement of rearing juveniles up to 5-10 grams are required to reduce costs, e.g. better feeds and feeding methods, improved bio-security, innovation in grading, counting and quality control systems

We invite the production sector in the first place as well as the research community to join in this initiative and help identifying short term as well as long term research priorities that could help the larviculture & juvenile production sector to increase its commercial competitiveness at both the European as well as the world level.

For more information, please contact EATP initiative area LARVI  
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#### MICROBIAL DIVERSITY WITHIN EARLY-STAGE CULTURED PANULIRUS ORNATUS PHYLLOSOMAS

Matthew S. Payne, Mike R. Hall, Lindsay Sly, David G. Bourne-2007

Applied and Environmental Microbiology 73(6): 1940-1951

Abstract:

A thorough understanding of the microorganisms and pathogens associated with the larval stage of the tropical ornate rock lobster, *Panulirus ornatus*, is required to overcome disease outbreaks that currently block aquaculture attempts. This study used microscopy in addition to culture and molecularly based microbiological techniques to characterize the bacterial community associated with cultured, developmental stage PI to PII *P. ornatus* phyllosomas. Scanning electron microscopy demonstrated colonization of phyllosomas by filamentous, rod-shaped, and coccus-shaped bacteria. A clone library constructed from dead phyllosomas sampled from the larval rearing tank on day 10 was dominated by *Thiothrix*-affiliated sequences (56% of clones). A comparable library from live phyllosomas also contained *Thiothrix*-affiliated sequences, though these only represented 19% of clones within the library. Fluorescent in situ hybridization (FISH) confirmed identification of the filamentous bacteria as *Thiothrix* sp., being present on dead phyllosomas. FISH also identified *Leucothrix* sp. and *Vibrio* sp., as well as a range of other rod- and coccus-shaped bacteria, colonizing both live and dead phyllosomas. The development of the microbial community associated with phyllosomas was monitored through a standard larval rearing run using denaturing gradient gel electrophoresis (DGGE). *Vibrio* sp.-affiliated bands dominated the profiles of live animals through the rearing period and dead phyllosomas sampled on selected days. The population of *Vibrio* sp. associated with phyllosomas was monitored with culture-based analysis on selective media and demonstrated to increase significantly on day 7, coinciding with the beginning of the larval molt. An isolated *Vibrio harveyi* strain demonstrated an identical 16S rRNA sequence with retrieved DGGE and clone library sequences. Colonization of phyllosomas with filamentous bacterial species potentially hinders the ability of the animals to molt and, combined with the added stress of the molt process, likely results in reduced immune function, allowing opportunistic pathogenic *Vibrio* sp. to cause larval mortalities.

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#### SOLAR HEATING SYSTEMS FOR RECIRCULATION AQUACULTURE

R.J. Fuller-2007

Aquacultural Engineering 36(3): 250-260

Abstract:

The literature over the past 25 years indicates that there has been a continued interest in using passive and active solar technologies to reduce the conventional energy required to maintain water temperatures in small recirculation aquaculture systems. Although all of the experimental systems reviewed report favourable results, there is little information available to guide system designers. This paper describes the use of a simulation model to predict the annual conventional energy consumption of a 10.6 m<sup>3</sup> RAS enclosed in a double layer polyethylene greenhouse in two different climates. The water was maintained at 22.5 °C and the recirculation rate was 10% of tank volume per day. Simple unglazed solar collectors have also been combined with the greenhouse to further reduce energy consumption. The effect of increasing collector area on the solar fraction and utilization of useful energy was predicted. Finally, the model was used to investigate the relationship between the occurrence of condensation on the inner cover, ventilation rates and energy use.

It was found that in a hot dry climate, the greenhouse alone was sufficient to reduce the conventional energy requirements by 87%; while in the cooler temperate climate reductions of 66% were possible. When solar collectors were added to the system, conventional energy requirements were reduced further and depended on the area of collector used. For example, in the temperate climate location, conventional energy requirements were reduced to 23% of a RAS enclosed in a non-solar building when 26 m<sup>2</sup> of solar collector inclined at the optimum angle for winter energy collection were used. Although condensation could be successfully reduced by ventilation of the greenhouse, this increased conventional energy requirements because the potential for evaporation was increased. Covering the tanks at night was found to be a more effective strategy because it reduced condensation and conventional energy use simultaneously.

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#### USING A MACROALGAE ULVA PERTUSA BIOFILTER IN A RECIRCULATING SYSTEM FOR PRODUCTION OF JUVENILE SEA CUCUMBER APOSTICHOPUS JAPONICUS

Hua Wang, Chang-Fa Liu, Chuan-Xin Qin, Shu-Qing Cao, Jun Ding-2007

Aquacultural Engineering 36(3): 217-224

##### Abstract:

A simple indoor recirculating system for production of juvenile sea cucumber (*Apostichopus japonicus*) was operated on a commercial scale for 90 days during winter. The system consists of three 70 m<sup>3</sup> sea cucumber rearing tanks and one biofilter tank where macroalgae (*Ulva pertusa*) was used as a biofilter in order to reduce water requirements. Effluent from the sea cucumber tanks drained into the macroalgae biofilter tank and were then returned to the sea cucumber tanks by a discontinuous-flow recirculation system. Survival and growth rates in the sea cucumber culture tanks were similar to those in the control tank (with one water exchange per day). The survival rate averaged about 87%. The average body weight increased from 3.5 ± 0.3 g to 8.1 ± 0.8 g and total sea cucumber biomass production over the experimental period was 745 g m<sup>-2</sup> after initial stocking densities of 375 g m<sup>-2</sup>. The growth rate of *U. pertusa* was 3.3% day<sup>-1</sup>. *U. pertusa* was efficient in removing toxic ammonia and in maintaining the water quality within acceptable levels for sea cucumber culture; there were only small daily variations of temperature, pH and DO. The *U. pertusa* tank removed 68% of the TAN (total ammonia-nitrogen) and 26% of the orthophosphate from the sea cucumber culture effluent; the macroalgae biofilter removed ammonia at an average rate of 0.459 g N m<sup>-2</sup> day<sup>-1</sup>. It would be efficient to use the *U. pertusa* biofilter in a recirculating system for production of *A. japonicus* juveniles in winter.

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#### USE OF THE GENERIC NAME PENAEUS

D.J. Alderman, B.A. Costa-Pierce, E.M. Donaldson, G. Hulata, R.P. Wilson-2007

Aquaculture 264(1-4): 1

##### EDITORIAL

The editors wish to draw to the attention of authors to the publication in this issue of *Aquaculture* of a paper by T.W. Flegel entitled "The right to refuse revision in the genus *Penaeus*". This paper discusses the effects of the publication by Pérez Farfante and Kensley (1997) of a monograph of shrimps and prawns of the world which included a proposed taxonomic revision raising former subgenera in the genus *Penaeus* to generic rank.

The editors of *Aquaculture* have for some time felt that this revision had resulted in some confusion with some authors and journals using the new binomials of Pérez Farfante and Kensley (1997) and others remaining with the traditional *Penaeus* binomials.

The editors are in agreement with the arguments put forward in T.W. Flegel's paper and have agreed that Aquaculture will prefer that authors of future submissions to this journal that concern species of Penaeid shrimp use the generic epithet *Penaeus*, preferably qualified at the first mention by Flegel's proposal, namely to follow the rules of zoological nomenclature by placing the sub-genus names in brackets between the traditional genus name *Penaeus* and the relevant species name at first mention [e.g., *Penaeus* (*Fenneropenaeus*) *chinensis*].

The editors do not however wish to be “coercive” in this matter and so authors who feel strongly supportive of the revised Pérez Farfante and Kensley (1997) generic names should use Flegel's alternative at the first mention of the genus to indicate that another name is also used [e.g., *Fenneropenaeus chinensis* (also called *Penaeus chinensis*)].

References:

Pérez Farfante and Kensley, 1997 I. Pérez Farfante and B. Kensley, Penaeoid and sergestoid shrimps and prawns of the world (keys and diagnoses for the families and genera), Muséum National d'Histoire Naturelle, Paris (1997).

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## THE RIGHT TO REFUSE REVISION IN THE GENUS *PENAEUS*

T.W. Flegel-2007

*Aquaculture* 264(1-4): 2-8

Refers to: Use of the generic name *Penaeus*, *Aquaculture*, 264(1-4) 1, by D.J. Alderman, B.A. Costa-Pierce, E.M. Donaldson, G. Hulata and R.P. Wilson

Abstract:

In 1997 Pérez Farfante and Kensley published a monograph of the penaeoid and sergestoid shrimps and prawns of the world. It incorporated a proposed taxonomic revision by raising former sub-genera in the genus *Penaeus* to generic rank. This would result in replacement of the 27 traditional penaeid shrimp binomials with an almost completely new set. Due to general unfamiliarity regarding the rules of zoological nomenclature, non-specialists in the shrimp industry and even scientists in related academic fields felt that they were obliged by taxonomic rules to follow the changes embodied in the monograph, whether they agreed with them or not. Others more familiar with their rights (including myself) continued to use the traditional binomials. The result has been some confusion in shrimp nomenclature in the succeeding 9 years. The purpose of this review is to argue that the revisions embodied in the Pérez Farfante and Kensley monograph are extremely disruptive to communication amongst practitioners in the shrimp fishery and the shrimp aquaculture industry and to scientists and students who study shrimp. This feature alone is counter to the goal of stability embodied in the zoological code of nomenclature and can alone be sufficient justification to consider the proposed revisions unacceptable. Indeed, the success of proposed taxonomic revisions does not fall under the zoological code, since the code is concerned with issues of priority. Instead, revisions survive or die depending on the majority action of the whole impacted community acting as individuals to accept them by use or reject them by disuse. Apart from arguments based on nomenclatural stability, I will attempt to show that sufficient new genetic information on penaeid shrimp has been accumulated in the past 9 years to show that there is no compelling reason to accept the revisions. Should readers accept my arguments and wish to carry on with the use of traditional penaeid shrimp binomials, I would like to propose a transitional, compromise practice to improve communications. This would involve acceptance of the sub-genus names by including them in brackets between the genus name *Penaeus* and the relevant species names, as is recommended by the rules of zoological nomenclature [e.g., *Penaeus* (*Fenneropenaeus*) *chinensis*]. Unfortunately for those who support the proposed revision, the reverse cannot be done because *Penaeus* is not a sub-genus name. In that case, the only viable option is to include a statement in brackets after the new binomial the first time it is mentioned [e.g., *Fenneropenaeus chinensis* (also called *Penaeus chinensis*)].

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GENETIC ASPECT IN BROODSTOCK MANAGEMENT OF THE CRITICALLY ENDANGERED MEKONG GIANT CATFISH, PANGASIANODON GIGAS IN THAILAND

Kednapat Sriphairoj, Wongpathom Kamonrat, Uthairat Na-Nakorn-2007

Aquaculture 264(1-4): 36-46

Abstract:

A broodstock management plan was designed for the world's biggest catfish, the critically endangered Mekong giant catfish (MGC), *Pangasianodon gigas*, based on multilocus genotypes of seven microsatellite loci. The broodstock included a total of 129 pit-tagged mature MGC from seven farms in Thailand covering approximately five year-classes. The genetic variation within stocks was relatively low (number of alleles/locus =  $2.29 \pm 0.76$ – $4.00 \pm 1.83$ ; effective number of alleles/locus =  $1.84 \pm 0.51$ – $3.04 \pm 1.04$ ;  $H_o = 0.58 \pm 0.34$ – $0.80 \pm 0.12$ ;  $H_e = 0.43 \pm 0.21$ – $0.66 \pm 0.11$ ). The genetic relatedness ( $r_{xy}$ : [Ritland, K., 1996. Estimators for pairwise relatedness and individual inbreeding coefficients. *Genet. Res.* 67, 175–185.]) was used to identify the unrelated pairs wherein a minimum value of  $r_{xy}$  for half-sib family ( $r_{xy} = 0.07$ ) was considered as a critical value of unrelatedness. The average  $r_{xy}$  within farm ( $0.21 \pm 0.13$ – $0.31 \pm 0.07$ ) was relatively high compared to the overall average  $r_{xy}$  ( $0.20 \pm 0.01$ ), suggesting collaborative broodstock management among farms should be carried out in future. In the short-term six scenarios were proposed based on the 28 and 63 mature male and female brooders, respectively. The results suggest that the best strategy for maintaining the genetic diversity while minimizing inbreeding of MGC was to select mating pairs based on the  $r_{xy}$ . A long-term management plan simulation using the BOTTLESIM program showed that if a random mating scheme is adopted  $N_e$  should be kept at 100 in order to preserve > 90% A for four generations (120 years). Applying the mk (minimal kinship) selection for only the first generation can reduce  $N_e$  to > 30 individuals and can retain > 90% A throughout the same period.

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EFFECTIVENESS OF TREATMENTS AGAINST EGGS, AND ADULTS OF HALIOTREMA SP. AND EURYHALIOTREMA SP. (MONOGENEA: ANCYROCEPHALINAE) INFECTING RED SNAPPER, LUTJANUS GUTTATUS

E.J. Fajer-Ávila, S.P. Velásquez-Medina, M. Betancourt-Lozano-2007

Aquaculture 264(1-4): 66-72

Abstract:

Ancyrocephaline monogeneans such as *Haliotrema* sp. and *Euryhaliotrema* sp. are common parasites infecting gills on the red snapper, *Lutjanus guttatus*, an important potential fish species for aquaculture in Mexico. The effectiveness of different treatments against these monogeneans using in vitro and in vivo assessments on the viability of eggs and adults was evaluated. Eggs from both monogenean species were in vitro exposed to sodium hypochlorite, drying, formalin, and freshwater for 3 h, and returned to seawater to determine post-treatment hatching. Adults of both parasites were also in vitro exposed to formalin, freshwater, and caprylic acid for periods ranging from 10 to 60 min using seawater as control. In vivo tests were carried out with freshwater, formalin, and praziquantel (active ingredient of Drontal™ Plus and Vermiplex™ Plus) on snappers infected with both parasites. After 3 h exposure, egg hatching was completely suppressed at 120 mg l<sup>-1</sup> of available chlorine, while drying, formalin (83 mg l<sup>-1</sup>) and freshwater showed eggs viabilities of 10, 23, and 10% respectively. In vitro freshwater exposure significantly reduced the mean intensity of ancyrocephaline adults (99% at 30 min), followed by formalin at 51 mg l<sup>-1</sup> (72% at 60 min) and caprylic acid at 334 mg l<sup>-1</sup> (5% at 30 min). In vivo freshwater baths for 30 min removed 100% of ancyrocephalines, although this method was stressful for the fish, while formalin (60 min at 51 mg l<sup>-1</sup>), Drontal™ Plus (12-h at 4.5 mg l<sup>-1</sup>) and Vermiplex™ Plus (24-h at 3.5–4.5 mg l<sup>-1</sup>) removed 72, 100 and 100% respectively. This study indicates that praziquantel, freshwater, and formalin baths were effective against ancyrocephaline adults from red snapper, and sodium hypochlorite would be useful to prevent horizontal infection with eggs on fish-rearing facilities.

(Centro de Investigación en Alimentación y Desarrollo, A. C., Unidad Mazatlán en Acuicultura y Manejo Ambiental. Av. Sábalo Cerritos s/n, C.P. 82010, A.P. 711, Mazatlán, Sinaloa, México ; email of E.J. Fajer-Ávila : [efajer@victoria.ciad.mx](mailto:efajer@victoria.ciad.mx))

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#### EFFECT OF TANK COLOUR ON ARTEMIA INGESTION, GROWTH AND SURVIVAL IN CULTURED EARLY JUVENILE POT-BELLIED SEAHORSES (HIPPOCAMPUS ABDOMINALIS)

Leonardo Martinez-Cardenas, G. John Purser-2007

Aquaculture 264(1-4): 92-100

Abstract:

The effect of the background (tank) colour on feeding activity, growth and survival of early juvenile seahorses *Hippocampus abdominalis* was investigated in two experiments. In experiment one, two sets of background colour (Set 1: Clear, white, red, blue, black; Set 2: Clear, white, yellow, orange, green) were used to quantify short-term Artemia ingestion in 42-day-old fish, and 7-day-old fish (Set 1 only). No statistical difference was observed between data. In the second experiment 3-day-old seahorses were cultured over 6 weeks in one of six coloured tanks (clear, white, yellow, red, blue, black) in a temperature and photoperiod controlled recirculation system to determine the effect of the background colour on growth, survival and conditioning to tank colour. At the end of the experiment there were no significant differences between any of the parameters tested. Early juvenile seahorses under the described experimental conditions were able to feed, grow and survive in a similar manner in any of the tank colours tested. Seahorses did not appear to display any conditioning to the tank colour in which they were cultured.

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#### SAFETY OF ELECTROLYZED SEAWATER FOR USE IN AQUACULTURE

Masahiko Katayose, Kyoichiro Yoshida, Nobuo Achiwa, Mitsuru Eguchi-2007

Aquaculture 264(1)4): 119-129

Abstract:

The safety of electrolyzed seawater was evaluated by measuring the production rate of organic halogen compounds and the occurrence of reverse mutations. Aquaculture feedwater and wastewater were collected from a fish-culturing facility, and available chlorine of approximately 1.0 mg/L was generated to ensure a disinfectant effect. More than 90% of the generated organic halogen compounds were bromoform. The amount of bromoform was far less than the reference values for drinking water standards in Japan and the U.S., provided that the electrolyzation was performed within the range of normal use. The reverse mutation assay of electrolyzed seawater showed no mutagenicity. Electrolyzed seawater with available chlorine at an adequate level for disinfection can be used safely and effectively in various aspects of aquaculture.

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#### MIXOTROPHIC CULTIVATION OF MICROALGA SPIRULINA PLATENSIS USING MOLASSES AS ORGANIC SUBSTRATE

Michele R. Andrade, Jorge A.V. Costa-2007,

Aquaculture 264(1-4): 130-134

Abstract:

*Spirulina* is a microalga rich in proteins, vitamins and polyunsaturated fatty acids. This microorganism grows photosynthetically but an organic substrate can stimulate its growth. Molasses is a by-product from sugar industry, containing more than 50% of sugar, and potentially useful as substrate to microalgae culture. On the other hand, light is needed to the photosynthetic fixation of

CO<sub>2</sub>. So, we determined the effects of molasses concentration and light levels on mixotrophic biomass production by *Spirulina platensis*. Molasses concentration was the main factor influencing maximum biomass concentration ( $X_{max}$ , g L<sup>-1</sup>) and maximum specific growth rate ( $\mu_{max}$ , d<sup>-1</sup>), although light intensity also influenced both parameters after 11 days.  $X_{max}$  reached 2.94 g L<sup>-1</sup> and  $\mu_{max}$  0.147 d<sup>-1</sup> while the average maximum productivity ( $P_{max}$ , g L<sup>-1</sup> d<sup>-1</sup>) of 0.32 g L<sup>-1</sup> d<sup>-1</sup> occurred within the first few days and was not significantly affected either by the molasses concentration or light. Biomass production was stimulated by molasses, suggesting that this industrial by-product could be used as a low-cost supplement for the growth of *Spirulina platensis*.

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DEVELOPMENT OF MUSSEL (*MYTILUS GALLOPROVINCIALIS*) SEED FROM TWO DIFFERENT ORIGINS IN A SEMI-ENCLOSED MEDITERRANEAN BAY (N.E. SPAIN) M. Ramón, M. Fernández, E. Galimany-2007

Aquaculture 264(1-4): 148-159

Abstract:

Growth and mortality of mussel seed from two different Mediterranean ecosystems, a small semi-enclosed bay (Fangar) and an open sea area (Masnou), were investigated in Fangar Bay (Ebro Delta) using standard procedures. Mussel sampling was performed on a monthly basis from October 2002 to August 2003, in conjunction with monitoring of hydrobiological parameters. Environmental parameters varied over a broad range and their fluctuations showed a similar pattern at the three depths studied (0, 1 and 3 m). Seston quantity (total particulate matter, TPM) ranged between 2.7 and 14.95 mg l<sup>-1</sup>, whereas the percentage organic matter in the TPM varied between 11.56% (November 2002) and 60.98% (August 2003). Growth results showed that mussels developed from bay seed maintained a greater length than those from open waters from October 2002 to February 2003, but there were no differences in length between both groups from March 2003 until the end of the experiment. Although there was no significant difference in final length for the two seed sources, better performance was observed for bay seed with respect to weight (both shells and soft tissues) and mortality. There was no correlation between the seasonal pattern of mussel growth in Fangar Bay and primary production, temperature or seston, probably due to the wide-ranging variation of these parameters. Seasonal growth patterns were coincident between shell and soft tissues, with the exception of August, when shell weight was maintained and flesh weight diminished. The results of yield in weight and mortality obtained in this study showed the benefits of using mussel seed from the same area where the cultures are carried out.

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EFFECT OF REARING DENSITY ON GROWTH, SURVIVAL AND REPRODUCTIVE PERFORMANCE OF DOMESTICATED *PENAEUS MONODON*

G.J. Coman, S.J. Arnold, M.J. Jones, N.P. Preston-2007

Aquaculture 264(1-4): 175-183

Abstract:

The growth, survival and reproductive performance of tank-reared, 2nd generation domesticated *Penaeus monodon*, originating from founder stocks collected from a population off the coast of Weipa (12°48', 141°32'), Gulf of Carpentaria, Queensland, Australia, were compared when reared at a standard density for maturation (SD) (approx. 3 individuals m<sup>-2</sup>) and a low density for maturation (LD) (approx. 1 individual m<sup>-2</sup>). Growth and survival of the shrimp reared at the two densities in 10·000 L tanks (3.6 m dia.; 10 m<sup>2</sup> tank-bottom area) were assessed from 5.5 months of age through to 10.5 months of age. Reproductive performance of shrimp in the two density treatments was assessed from 10.5 months of age for two moult period's post-ablation under standardized conditions. The weight of the female shrimp at 10.5 months of age (mean ± S.E.) was significantly greater at LD

(131.2 ± 4.9 g) than at SD (120.3 ± 2.5 g). No difference in 10.5 month weight was found between males at LD (90.9 ± 2.2 g) and SD (87.4 ± 1.5 g). Significantly more spawnings were produced at LD (2.2 ± 0.2 ablated female<sup>-1</sup>; 3.4 ± 0.6 spawning female<sup>-1</sup>) than at SD (1.3 ± 0.2 ablated female<sup>-1</sup>; 2.4 ± 0.6 spawning female<sup>-1</sup>). The percentage of spawnings that hatched (LD 87.2 ± 5.1%; SD 61.8 ± 6.6%), eggs per spawning (× 103) (LD 231 ± 21; SD 148 ± 16) and nauplii per spawning (× 103) (LD 39 ± 8; SD 16 ± 3) were also significantly higher at LD. Hatch rates per spawning were low and did not differ between LD (19.0 ± 2.7%) and SD (16.8 ± 2.4%). No significant differences in spermatophore weight (LD 0.19 ± 0.02 g; SD 0.20 ± 0.01 g) or sperm count (× 106) (LD 6.25 ± 0.77; SD 8.44 ± 1.15) were found between males at the two densities. While egg and nauplii production (× 103) were significantly higher at LD than SD when expressed per female (LD 434 ± 115 eggs, 73 ± 26 nauplii; SD: 160 ± 44 eggs, 17 ± 7 nauplii), no significant difference was found when expressed per m<sup>2</sup> tank-bottom area of the maturation tanks (LD 272 ± 67 eggs, 46 ± 21 nauplii; SD 271 ± 109 eggs, 29 ± 15 nauplii). Our results found that lowering the maturation density of tank reared *P. monodon* stocks from approx. 3 individuals m<sup>-2</sup> to 1 individual m<sup>-2</sup>, over a 5 months maturation period, improved growth of females and reproductive performance when expressed per spawning or per female. However, no significant increases in egg and nauplii production were obtained by reducing the maturation density when expressed per tank-bottom area.

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#### EFFECT OF WEANING AGE AND DIET ON PIKEPERCH LARVICULTURE

Patrick Kestemont, Xu Xueliang, Neïla Hamza, Jean Maboudou, Ibrahim Imorou Toko-2007  
*Aquaculture* 264(1-4): 197-204

##### Abstract:

The effects of weaning age and diets on survival, cannibalism, deformity and growth were investigated in pikeperch post-larvae *Sander lucioperca*. In a first experiment, fish were fed *Artemia* nauplii after hatching and then divided, on days 12, 19 and 26 post-hatch, into groups receiving *Artemia* nauplii as control or artificial diet. Significant differences were observed on survival, cannibalism, deformity and growth performance between the different weaning age and control groups. The best growth (mean weight gain = 380.8 mg) with the lowest mortality (48.1%) and lowest deformity rate (11.9%) but high cannibalism (36.7%) was obtained in larvae weaned at day 19 post-hatch, whereas the lowest growth (mean weight gain = 218.9 mg) with the highest mortality (68.6%), high deformity (22.3%) and low cannibalism (13.8%) was obtained in fish weaned at day 12 post-hatch. The highest survival but with highest deformity, mainly incomplete mouth development, was observed in control group that was fed *Artemia* nauplii only. In a second experiment, 19-day old post-larvae were fed different larval feeds (formulated for marine or freshwater fish species) during 18 days and performances (survival, growth, cannibalism, resistance to osmotic stress test) were compared to fish fed standard or HUFA + vitamin C enriched *Artemia* metanauplii. The best growth was obtained in fish fed *Artemia* metanauplii (final mean weight = 301.5 and 372.7 mg, in standard and enriched *Artemia* groups, respectively), while the feeds formulated for freshwater fish species resulted in better growth (final mean weight = 176.6 and 230.8 mg) than those formulated for marine species (final mean weight = 123.8 and 144.5 mg). The highest survival, growth and resistance to saline stress as well as the lowest deformity were found in fish fed HUFA + vitamin C enriched *Artemia* metanauplii. Significant differences of ascorbic acid content in fish were observed among the dietary treatments, ranging from 17.5 ± 6.4 to 62.5 ± 8.3 µg fish dry matter<sup>-1</sup>. Highly significant correlations were achieved between dietary ascorbic acid content and the ascorbic acid content in larval carcass (R<sup>2</sup> = 0.91) or the reduction of larval deformity (R<sup>2</sup> = 0.75).

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#### GROWTH OF JUVENILE COBIA, *RACHYCENTRON CANADUM*, AT THREE DIFFERENT DENSITIES IN A RECIRCULATING AQUACULTURE SYSTEM



Kenneth A. Webb, Jr., Glenn M. Hitzfelder, Cynthia K. Faulk, G. Joan Holt-2007  
Aquaculture 264(1-4): 223-227

Abstract:

Cobia (*Rachycentron canadum*) is an excellent aquaculture candidate and culture of this species continues to develop worldwide. Current culture practices generally include larviculture and production of early juveniles on land with final growout occurring in ocean cages. Data indicate that production and/or growout of juveniles in land based recirculating systems may be hampered by growth depression in fish held at even relatively low densities. The current study investigated the responses of early cobia juveniles cultured at three different stocking densities (0.04, 0.22, and 0.44 g of fish/L) over a 10-week period in a recirculating aquaculture system. Water temperature was maintained at  $27 \pm 1$  C and salinity was  $32.0 \pm 3.0$  ppt. Fluorescent light banks were used to maintain a light/dark cycle of 14 h light/10 h dark. Early juveniles used in this trial had an average initial weight of  $6.7 \pm 0.2$  g and were stocked at 3, 10, or 20 per tank in order to reach target stocking densities. Fish were fed to satiation twice daily with a commercial diet and the amount fed was measured to determine feed efficiency. Fish from each tank were counted and weighed collectively each week until the end of the trial to monitor growth and survival. In addition, fish from the control (0.04 g/L) tanks were individually marked via clipping of the pectoral fins and weighed individually each week. Survival was high ( $\geq 96\%$  in all treatments) and there were no significant differences among treatments. Growth was also not significantly different among treatments with weight gains between 2523 and 2747% of initial weight (SGR ranging from 5.18 to 5.29). Feed efficiency of the control ( $0.96 \pm 0.02$ ) was significantly lower than that of the 0.22 g/L treatment ( $1.04 \pm 0.03$ ).

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THE EFFECT OF DIET TYPE ON GROWTH AND FATTY-ACID COMPOSITION OF SEA URCHIN LARVAE, I. PARACENTROTUS LIVIDUS (LAMARCK, 1816) (ECHINODERMATA)  
Hui Liu, Maeve S. Kelly, Elizabeth J. Cook, Kenneth Black, Heather Orra, Jian Xin Zhu, Shuang Lin Dong-2007

Aquaculture 264(1-4): 247-262

Abstract:

This study investigated the growth and fatty acid composition of the sea urchin larvae *Paracentrotus lividus* fed on four types of diets: the microalgae *Dunaliella tertiolecta*, two types of microencapsulated formulated feeds and a concentrated algal paste. The larvae were successfully raised to metamorphosis on three diets, including the microalgae and the two formulated feeds. No significant difference was found in survival rate and metamorphosis rate for larvae fed with microalgae, formulated feeds, or microalgae plus formulated feeds. The fastest growth rate was obtained for larvae fed with microalgae, while normal growth and survival were only achieved for larvae fed with formulated feeds at a high frequency feeding rate. Normal larval development was not supported by either of the formulated feeds at a low feeding rate. A concentrated algal paste was also used for the low ration experiment, but failed to support any larval growth. Post-larval development was better for larvae fed on the formulated feeds, presumably reflecting the nutritional value of these feeds. *P. lividus* larvae grew well on *D. tertiolecta* which had a relatively low content of protein (37% dry weight) and lipid (7% dry weight). The larvae could assimilate highly unsaturated fatty acids (HUFAs) such as DHA, EPA and ARA from the diet, or synthesize them from linoleic acid 18:2 n-6 and linolenic acid 18:3 n-3. They could also biosynthesize non-methylene-interrupted-dienoic fatty acid NMID 20:2 de novo when feed on *D. tertiolecta* and the formulated feeds. An accumulation of PUFA n-3 may improve larval growth. *P. lividus* larvae showed the tendency of food selection towards *D. tertiolecta*, and a better formulated feed with additives or attractants will give a better performance in larval growth.

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THE EFFECT OF DIET TYPE ON GROWTH AND FATTY ACID COMPOSITION OF THE SEA URCHIN LARVAE, II. PSAMMECHINUS MILIARIS (GMELIN)

Hui Liu, Maeve S. Kelly, Elizabeth J. Cook, Kenneth Black, Heather Orr, Jian Xin Zhu, Shuang Lin Dong-2007

Aquaculture 264(1-4): 263-278

Abstract:

This study investigated the growth and fatty acid composition of the sea urchin larvae *Psammechinus miliaris* fed on four types of diets: the microalgae *Dunaliella tertiolecta*, two types of microencapsulated formulated feed (L and F) and a concentrated algal paste. The fastest growth rate was obtained from *D. tertiolecta*, but there was no significant difference in larval survivorship between treatments. Larvae fed the formulated feeds showed good growth and survival, although those fed on feed L performed better than those fed on feed F. The metamorphosis rate of competent larvae fed on *D. tertiolecta* and feed L was not significantly different, but higher than those in the other treatments. Similar post-settlement survival was obtained for larvae fed on microalgae and both formulated feeds, yet 10 d post-settlement juveniles from larvae fed on the formulated feeds were significantly larger. The concentrated algal paste did not support normal growth of the larvae. *P. miliaris* larvae could grow well on relatively low protein (37% dry weight) and lipid (7% dry weight) *D. tertiolecta* diet, but high levels of dietary protein and lipid of the formulated feeds may benefit the post-settlement growth of newly metamorphosed juveniles. Dietary fatty acid had a strong influence on lipid composition of the larval tissue. The larvae could incorporate dietary highly unsaturated fatty acids (HUFAs) directly from the formulated feeds, or obtain ARA, EPA and DHA through the elongation of linoleic acid 18:2 n-6 and linolenic acid 18:3 n-3 when fed the HUFA-deficient *D. tertiolecta*. Higher levels of HUFAs did not seem to act as a growth promoter for the larvae, but a higher concentration of DHA in the formulated feeds may have improved post-settlement growth. An accumulation of PUFA n-3, especially 18:3 n-3, 18:4 n-3, 20:3 n-3 and 20:4 n-3 may promote larval growth. The shortage of PUFA n-3 and PUFA n-6, and especially the lack of non-methylene-interrupted-dienoic fatty acid NMID 20:2 may have impeded the growth of larvae fed on the algal paste. It is suggested that a better formulated feed with improved buoyancy and a more even particle size distribution would improve larval growth.

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MODELLING SURVIVAL AND GROWTH OF MITHRACULUS FORCEPS' LARVAE AND JUVENILES (A. MILNE EDWARDS, 1875) (DECAPODA: BRACHYURA: MAJIDAE) IN AQUACULTURE

Gil Penha-Lopes, Joana Figueiredo, Luís Narciso-2007

Aquaculture 264(1-4) : 285-296

Abstract:

In the present study, by using accumulated information of *Mithraculus forceps* larval and juvenile rearing, survival, growth/size and productivity models were developed, allowing for optimization and improvement of the culture methodology of this species. By taking into account the productivity, we estimated larval optimal conditions as: 60 larvae.L<sup>-1</sup> and 10 *Artemia* nauplii.mL<sup>-1</sup>. For juveniles we estimated optimal conditions as: 12 900 crabs.m<sup>-2</sup> and feed juveniles with *Artemia* nauplii, if possible supplemented with *Amphora* microalgae.

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NITRITE-OXIDIZING BACTERIA, NITROSPIRA, DISTRIBUTION IN THE OUTER LAYER OF THE BIOFILM FROM FILTER MATERIALS OF A RECIRCULATING WATER SYSTEM FOR THE GOLDFISH *CARRASSIUS AURATUS*

Shiro Itoi, Noriaki Ebihara, Sayaka Washio, Haruo Sugita-2007

Aquaculture 264(1-4): 297-308

Abstract:

We used a clone library method to investigate changes in the biofilm microflora associated with the conditioning of filter materials in a recirculating water system for the culture of goldfish *Carassius auratus*. The microbial density was higher in the outer layer of biofilm from filter materials ( $1.7 \times 10^{10}$ – $3.0 \times 10^{10}$  cells/g) than in the inner layer ( $1.5 \times 10^9$ – $2.4 \times 10^9$  cells/g) throughout the experimental period. The clone library method using bacterial 16S rRNA genes collected from the outer layer of filter material yielded sequences from four (day 8), nine (day 15), twelve (day 22) and nine (day 64) taxonomic categories of bacteria including Acidobacteria, Actinobacteria, Bacilli, Bacteroidetes, Flavobacteria, Fusobacteria, Nitrospira,  $\alpha$ -Proteobacteria,  $\beta$ -Proteobacteria,  $\gamma$ -Proteobacteria, Sphingobacteria, Verrucomicrobia and unclassified bacteria. The inner layer yielded sequences from six (day 8), eight (day 15), five (day 22) and five (day 64) taxonomic categories of bacteria including Acidobacteria, Actinobacteria, Bacilli, Flavobacteria,  $\alpha$ -Proteobacteria,  $\beta$ -Proteobacteria,  $\gamma$ -Proteobacteria, Sphingobacteria, Verrucomicrobia and unclassified bacteria. Bacteria in the outer layer of biofilm belonged predominantly to the genera *Acinetobacter*, *Cetobacterium*, *Comamonas*, *Flectobacillus*, *Flavobacterium*, *Ideonella* and *Pseudomonas*, whereas those in the inner layer were predominantly members of the genera *Flavobacterium*, *Flexibacter*, *Ideonella*, *Janthinobacterium*, *Pedobacter* and *Pseudomonas*. As the filter materials became conditioned, the population of nitrite-oxidizing bacteria related to *Nitrospira* was restricted to the outer layer of the biofilm. In addition, phylogenetic analysis indicated the presence of both an indigenous *Nitrospira* and a distinct type of *Nitrospira* that appeared after conditioning had begun.

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THE EFFECT OF IONIZING IRRADIATION OF POST-LARVAE ON SUBSEQUENT SURVIVAL AND REPRODUCTIVE PERFORMANCE IN THE KURUMA SHRIMP, *PENAEUS (MARSUPENAEUS) JAPONICUS* (BATE)

Melony J. Sellars, Greg J. Coman, Tamera R. Callaghan, Stuart J. Arnold, Jan Wakeling, Bernard M. Degnan, Nigel P. Preston-2007

Aquaculture 264(1-4): 309-322

Abstract:

Considerable genetic advances in the Kuruma shrimp, *Penaeus (Marsupenaeus) japonicus*, have been achieved in the last decade, creating the demand for a technique to provide genetic copyright and prevent escapees from farm ponds genetically contributing to natural fishery populations. The induction of sexual sterility is one technique that may provide such protection. This study evaluated the potential of sterilizing reproductive age (10 month old) *P. japonicus* by ionizing irradiation (IR) treatment at an early life-history stage. To achieve this, a lethal dose curve was established for post-larval stage 15 (PL15) shrimp by treating with different doses of IR. From this curve, six irradiation levels — all below the 100% mortality treatment — were trialed, ranging from 0 to 30 Gray (Gy). Another group of PL15 *P. japonicus* were subsequently exposed to the six IR treatments and then reared until sexual maturity, at which time their survival and reproductive performance were assessed. At 8 months of age, females and males from each of the 0, 10, 15 and 20 Gy treatment groups were reciprocally crossed to give 16 mating combinations, whilst females and males from each of the 25 and 30 Gy treatment groups were directly crossed with 0 Gy shrimp of the opposite gender to give four direct mating crosses. At 10 months of age, all females had their reproductive performance assessed in a standardized 30 day trial. The percentage of females maturing and spawning in the 0 Gy treatment group ( $80.97 \pm 5.5\%$  and  $69.9 \pm 5.77\%$ ) were significantly higher ( $P < 0.05$ ) than for IR treated females in the 10, 15 and 20 Gy treatment groups ( $63.93 \pm 5.34\%$ ,  $61.68 \pm 5.77\%$  and  $43.66 \pm 6.03\%$  for percentage maturations and  $51.44 \pm 5.5\%$ ,  $49.69 \pm 6.04\%$  and  $38.94 \pm 6.31\%$  for percentage spawnings). Similarly, the number of maturations and spawnings per female in the 0 Gy treatment group ( $1.50 \pm 0.13$  and  $1.24 \pm 0.12$ ) were significantly higher ( $P < 0.05$ ) than for IR treated females in the 10, 15, and 20 Gy treatment groups ( $1.13 \pm 0.12$ ,  $1.04 \pm 0.13$  and  $0.76 \pm 0.14$  for the number of maturations and  $0.88 \pm 0.12$ ,  $0.84 \pm 0.13$  and  $0.67 \pm 0.13$  for the number of spawnings). The effects of IR on protozoal metamorphosis varied. No differences in fecundity, hatch rates,

gonadal somatic indices or gonadal histology were found between control and irradiated shrimp. These results indicate that IR of PL15 *P. japonicus* had some effect on ovarian maturation and spawning, however IR was not able to confer sterility in shrimp. Treatment of PL15s with IR that does not result in death (i.e. between 10 and 30 Gy) is therefore not effective at preventing the production of some viable offspring.

(CSIRO Food Futures National Research Flagship, CSIRO Marine and Atmospheric Research, 233 Middle Street, Cleveland, Qld. 4163, Australia; email of Melony J. Sellars: [Melony.Sellars@csiro.au](mailto:Melony.Sellars@csiro.au))

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#### INFLUENCE OF TEMPERATURE ON GONADAL DEVELOPMENT OF *Ruditapes philippinarum* (ADAMS AND REEVE, 1850) WITH SPECIAL REFERENCE TO INGESTED FOOD AND ENERGY BALANCE

Marina Delgado, Alejandro Pérez Camacho-2007

Aquaculture 264(1-4): 398-407

Abstract:

This study evaluated the influence of temperature on gonadal development in *Ruditapes philippinarum* under controlled dietary conditions, in order to distinguish between the effect of temperature alone and the associated effect of temperature on ingestion. The experiment was therefore divided into two groups: group L (at temperatures of 14 °C and 18 °C, with a daily ingestion rate of 470–550 µg organic weight of phytoplankton per g clam live weight) and group H (at temperatures of 18 °C and 22 °C, with a daily ingestion rate of 1000–1100 µg organic weight of phytoplankton per g clam live weight). Both ingestion and respiration rates were calculated for each of the diets and temperatures. Gonadal development of the different groups of clams was monitored over the 70-day experimental period by means of image analysis techniques and calculation of the soft tissue dry weight of a standard clam.

Experimental results show that when the amount of food ingested is similar a difference of temperature between 14 °C and 18 °C, or between 18 °C and 22 °C, has no significant effect on the rate of gonadal development, the effect of temperature possibly being masked by the differences in the respective energy balances.

When a higher temperature coincides with a higher ingestion rate, as is often the case with naturally-occurring populations of bivalves, then gonadal development is greater, and faster, at the higher temperature, although this is mainly due to the greater amount of food ingested.

When, in experimental (or environmental) conditions, a high temperature (18 °C) is associated with a low ingestion rate, a situation of negative energy balance arises, associated with a slow rate of gonadal development that takes place at the cost of the animal's energy reserves. When the temperature is reduced to 14 °C, there is a decrease in the ingestion rate and lower energy consumption, leading to a situation of positive energy balance. In these conditions gonadal development is slow and the surplus of available energy is, presumably, used to accumulate energy reserves, as is often the case during the stage of growth prior to the onset of gonadal development in many bivalves that inhabit geographical areas with noticeable seasonal variations.

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#### DIETARY N-3 HUFA DEFICIENCY INDUCES A REDUCED VISUAL RESPONSE IN GILTHEAD SEABREAM *SPARUS AURATA* LARVAE

T. Benítez-Santana, R. Masuda, E. Juárez Carrillo, E. Ganuza, A. Valencia, C.M. Hernández-Cruz, M.S. Izquierdo-2007

Aquaculture 264(1-4): 408-417

Abstract:

Developmental changes in swimming speed were analysed in the seabream (*Sparus aurata*) larvae. Four feeding regimes using live preys (rotifer *Brachionus plicatilis*) enriched with fish oil, soybean oil, linseed oil and rapeseed oil, differing in fatty acid profile, were tested during the first weeks of larval life. There was an increase in burst swimming speed and cruise swimming speed during the visual stimulus experiment at day 16th of life in the present study in agreement with the better eye development in larvae of this age. Swimming activity before stimulus was significantly reduced when larvae were fed rotifers enriched with vegetable oils. Larvae fed with rotifers enriched with fish oil reacted with a higher burst swimming speed after a visual stimulus than after the sound stimulus (159.5 SL/s vs. 18.30 SL/s) denoting the importance of the vision during this period of development not only for predation but also for the burst. The reduction in dietary essential fatty acid contents, by the enrichment with vegetable oils, delays the appearance of response to visual stimulus, in agreement with the minor DHA content in eyes and brains of these larvae and suggesting a delay in the functional development of brain and vision.

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#### SWIMMING ACTIVITY AND FEEDING BEHAVIOUR OF LARVAL EUROPEAN SEA BASS (*DICENTRARCHUS LABRAX* L): EFFECTS OF ONTOGENY AND INCREASING FOOD DENSITY

V. Georgalas, S. Malavasi, P. Franzo, P. Torricelli-2007

Aquaculture 264(1-4): 418-427

Abstract:

The behaviour of European sea bass larvae reared with the mesocosm technique was analysed in relation to ontogeny and increasing food density, with a particular focus on swimming and feeding activities. The behaviour of groups of larvae (20 animals each) belonging to three age classes (10, 20 and 30 days post-hatching) was video-recorded in experimental tanks under controlled laboratory conditions. In each replicate, three different food densities, in terms of number of naupliar *Artemia*/l, were obtained. The videotapes analysis allowed the identification of 6 behavioural units (larval MAPs) and their quantification in terms of either frequency (number/min), or duration (percent of time spent in that activity with respect to the total time of observation). Behavioural variation was analysed in relation both to age and food density. Results revealed that swimming activity increased significantly with age, especially between the 10 and the 20 days post-hatching, whereas resting activities decreased with age and, in some cases, also with food density. Frequency of feeding behaviours decreased with age if they were analysed singly, but when the association between aiming posture (Sigmoid-posture) and attacks was considered, results showed that these behaviours increased with both age and food density. Further, the increase of feeding efficiency with age was also confirmed by the increase in the number of *Artemia* nauplii/individual across the three age groups. Results are discussed in the light of both a comparative analysis conducted with the available data on the larval behaviour of other marine fish species, and their potential use for the assessment of the quality of the reared fish, also in relation to the rearing techniques.

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#### DEVELOPMENT AND CHARACTERISATION OF SOME INTESTINAL ENZYMES IN ATLANTIC COD (*GADUS MORHUA* L.) AND ATLANTIC HALIBUT (*HIPPOGLOSSUS HIPPOGLOSSUS* L.) LARVAE

A. Kvåle, A. Mangor-Jensen, M. Morena, M. Espe, K. Hamre-2007

Aquaculture 264(1-4): 457-468

Abstract:

To determine the status of development of the intestine, the activities of intestinal leucine-alanine peptidase (Leu-ala), brush border membrane (bbm)-bound alkaline phosphatase (AP) and bbm-bound leucine aminopeptidase N (LAP) were analysed from first feeding to passed weaning in Atlantic

halibut (*Hippoglossus hippoglossus*; 1–78 days post first feeding; dpff) and Atlantic cod (*Gadus morhua*; 0–72 dpff). Before conducting the ontogenetic studies, the enzyme assays used were optimised in relation to temperature and pH (only AP and LAP), and the Michaelis constant (KM; All enzymes) determined, in order to ensure valid data and to maximise the activity potentials. Leu–ala was used as a marker of intracellular digestion which was expected to decrease during the experimental periods. AP and LAP were used as markers of development of the bbm and achievement of an adult mode of digestion. In halibut, Leu–ala increased 11-folds ( $P = 0.0002$ ) in specific activity ( $\text{mU (mg protein)}^{-1}$ ) from first feeding to 34 dpff and thereafter a small decrease (0.65-fold;  $P = 0.006$ ) in activity was observed. In cod, the specific activity of Leu–ala varied randomly during the experiment. Distinct increases in specific activities of AP and LAP were observed from 29 to 52 dpff in halibut (fivefold;  $P < 0.002$ ) and from 29 to 37 dpff in cod (two–threefold;  $P < 0.02$ ). The bbm-bound activities of AP and LAP in percent of total intestinal activity of the respective enzyme increased from approximately 2 to 15% between 34 and 43 dpff in halibut ( $P < 0.0003$ ), confirming the results of specific activities. In cod, a similar result with AP was found between 29 and 51 dpff ( $P = 0.0003$ ). The increase in percentage of bbm-bound activity of LAP was later, increasing exponentially from constituting 5 to 31% of total individual activity between 37 and 72 dpff ( $P = 0.0003$ ). The experiments indicated that the digestion in halibut had advanced towards the adult mode at 40 to 50 dpff, while time of reaching the same maturational level in cod was less certain due to the different activity profiles of AP and LAP. As AP is well recognised as a bbm differentiation marker, it can be assumed that the intestine in cod matured when AP activity had reached an elevated level at approximately 40 to 50 dpff, whereas LAP possibly was induced later. Ceasing of the larval mode of digestion, expected to be indicated by reduced activity of Leu–ala, was not detected. (National Institute of Nutrition and Seafood Research (NIFES), P.O. Box 2029 Nordnes, 5817 Bergen, Norway; email of A. Kvåle: [audbrufl@online.no](mailto:audbrufl@online.no))

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#### IN VITRO MANIPULATION OF EGG ACTIVATION IN THE OPEN THELYCUM SHRIMP LITOPENAEUS

Ericka Rojas, Jorge Alfaro-2007

Aquaculture 264(1-4): 469-474

Abstract:

This research was designed to develop a basis for the in vitro manipulation of egg activation in *Litopenaeus*. The timing and sequence of events associated with egg activation of three species of *Litopenaeus*: *L. occidentalis*, *L. stylirostris*, and *L. vannamei* are described. Spawning in cold seawater (21.5 °C) induces a significant delay in cortical rod release as well as in hatching envelope formation. Magnesium concentration at 318.8 and 5.2 mg l<sup>-1</sup> did not induce any detectable variation in egg activation. In vitro availability of unfertilized eggs was accomplished by dissecting ovaries at the time of spawning; a latency period in dry conditions from 1 to 10 min induced no detrimental effect on initiation of egg activation. In addition, an in vitro assay for evaluating interspecific and intraspecific primary binding among species of *Litopenaeus* is presented.

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