

STRONG ENVIRONMENTAL TOLERANCE OF ARTEMIA UNDER VERY HIGH PRESSURE

K. Minami, F. Ono, Y. Mori, K. Takarabe, M. Saigusa, Y. Matsushima, N.I. Saini, M. Yamashita-2010
Journal of Physics: Conference Series 215: 1-4

Abstract:

It was shown by the present authors group that a tardigrade in its tun-state can survive after exposed to 7.5 GPa for 13 hours. We have extended this experiment to other tiny animals searching for lives under extreme conditions of high hydrostatic pressure. Artemia, a kind of planktons, in its dried egg-state have strong environmental tolerance. Dozens of Artemia eggs were sealed in a small Teflon capsule together with a liquid pressure medium, and exposed to the high hydrostatic pressure of 7.5 GPa. After the pressure was released, they were soaked in seawater to observe hatching rate. It was proved that 80-90% of the Artemia eggs were alive and hatched into nauplii after exposed to the maximum pressure of 7.5 GPa for up to 48 hours. Comparing with Tardigrades, Artemia are four times stronger against high pressure.

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DIAPAUSE TERMINATION AND DEVELOPMENT OF ENCYSTED ARTEMIA EMBRYOS: ROLES FOR NITRIC OXIDE AND HYDROGEN PEROXIDE

The Journal of Experimental Biology 213: 1464-1470

Heather M. Robbins, Gilbert Van Stappen, Patrick Sorgeloos, Yeong Yik Sung, Thomas H. MacRae, Peter Bossier-2010

SUMMARY:

Encysted embryos (cysts) of the brine shrimp Artemia undergo diapause, a state of profound dormancy and enhanced stress tolerance. Upon exposure to the appropriate physical stimulus diapause terminates and embryos resume development. The regulation of diapause termination and post-diapause development is poorly understood at the molecular level, prompting this study on the capacity of hydrogen peroxide (H₂O₂) and nitric oxide (NO) to control these processes. Exposure to H₂O₂ and NO, the latter generated by the use of three NO generators, promoted cyst development, emergence and hatching, effects nullified by catalase and the NO scavenger 2-phenyl-4,4,5,5-tetramethylimidazoline-1-oxyl 3-oxide (PTIO). The maximal effect of NO and H₂O₂ on cyst development was achieved by 4 h of exposure to either chemical. NO was effective at a lower concentration than H₂O₂ but more cysts developed in response to H₂O₂. Promotion of development varied with incubation conditions, indicating for the first time a population of Artemia cysts potentially arrested in post-diapause and whose development was activated by either H₂O₂ or NO. A second cyst sub-population, refractory to hatching after prolonged incubation, was considered to be in diapause, a condition broken by H₂O₂ but not NO. These observations provide clues to the molecular mechanisms of diapause termination and development in Artemia, while enhancing the organism's value in aquaculture by affording a greater understanding of its growth and physiology.

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REVIEW ARTICLE**A GLOBAL REVIEW OF SEAHORSE AQUACULTURE**

Heather J. Koldewey, Keith M. Martin-Smith-2010

Aquaculture 302(3-4): 131-152

Abstract:

Seahorses (*Hippocampus* spp.) are flagship species for many issues in marine conservation including overexploitation, incidental bycatch and habitat loss. Aquaculture has been proposed as one solution to address unsustainable trade for traditional medicine, aquarium fishes and curios. Here we review historical and current information on global seahorse aquaculture including characteristics of

aquaculture operations, species in culture, contribution to international trade and technical issues associated with raising seahorses in captivity. We found that prior to the 1990s, seahorse aquaculture was plagued by problems with disease and feeding. In the late 1990s and early 2000s there was considerable expansion in the number and size of aquaculture operations and the number of species in culture. This was reflected in an increasing contribution of captive-bred seahorses to the aquarium trade but not in the larger traditional medicine market. Currently, the majority of seahorse aquaculture involves small-scale operations in developed countries, employing relatively few personnel and selling live animals for the home aquarium market. Although, there are still considerable technical problems with diseases and with breeding and raising some species, others are performing successfully in aquaculture. There are currently at least 13 species in commercial culture or under research for their culture potential. However, economic viability remains a concern to many current aquaculture operations including price competition with wild-caught animals. Large-scale aquaculture to supply the traditional medicine market or as a livelihood venture has not yet been demonstrated to be commercially viable, although it is being actively researched.

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MONITORING OF THE BIOENCAPSULATION OF A PROBIOTIC PHAEOBACTER STRAIN IN THE ROTIFER BRACHIONUS PLICATILIS USING DENATURING GRADIENT GEL ELECTROPHORESIS

José Pintado, María Pérez-Lorenzo, Antonio Luna-González, Carmen G. Sotelo, María J. Prol, Miquel Planas-2010

Aquaculture 302(3-): 182-194

Abstract:

The bioencapsulation of the probiotic bacteria *Phaeobacter* 27-4 in the rotifer *Brachionus plicatilis* was monitored by culture methods and denaturing gradient gel electrophoresis (DGGE) of PCR-amplified 16 S rDNA.

In a first experiment, the permanence of the probiotic bacteria in clear water and green water was studied. *Phaeobacter* 27-4 added to the water of the tanks (107 CFU ml⁻¹) remained at levels around 106 CFU ml⁻¹ for 72 h and was not affected by the presence of the algae added (*Isochrysis galbana*, 105 cells ml⁻¹). The DGGE fingerprints showed a temporal predominance of the probiont in the water and the presence of bacteria belonging to the Flavobacteria, γ -proteobacteria, and Sphingobacteria groups. A *Tenacibaculum* strain became predominant when *Phaeobacter* 27-4 decline, and at the end of the experiment, bacterial profiles became similar to the initial ones with predominance of bacteria belonging to the Oceanospirillaceae family.

Three different ways of bioencapsulation of the probiont in the rotifer were assayed: E24, addition of *Phaeobacter* 27-4 for 24 h during the enrichment with *I. galbana*; E3, addition of *Phaeobacter* 27-4 during the last 3 h of the enrichment with *I. galbana* and E3+, with the bioencapsulation done in a separated step, after the 24 h enrichment with *I. galbana*, being the rotifers filtered, washed and transferred into tanks containing *Phaeobacter* 27-4 in seawater, and maintained for 3 h.

The result showed that the presence of the algae was not determinant in the effectiveness of the bioencapsulation and the probiont was bioencapsulated in all cases in the first 3 h to a level of 102 cfu rotifer⁻¹. When the rotifers with the bacteria bioencapsulated were transferred to green-water tanks and kept in the conditions used in turbot larvae rearing, *Phaeobacter* 27-4 maintained levels close to 102 CFU rotifer⁻¹ for 48 h in the case of E24 and E3, and for 24 h in the case of E3+, a period of time sufficient to the larvae to graze on them and to incorporate the probiotic. The E24 protocol was selected for the simplicity of the procedure. DGGE fingerprints showed the incorporation of the probiotic and a temporal colonization of the rotifers. Predominant bands identified in the rotifers correspond to γ -proteobacteria as *Pseudoalteromonas*.

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TEMPORAL PATTERNS OF ARRIVAL OF BEACHCAST GREEN-LIPPED MUSSEL (*PERNA CANALICULUS*) SPAT HARVESTED FOR AQUACULTURE IN NEW ZEALAND AND ITS RELATIONSHIP WITH HYDRODYNAMIC AND METEOROLOGICAL CONDITIONS

Andrea C. Alfaro, Brian McArdle, Andrew G. Jeffs-2010

Aquaculture 302(3-4): 208-218

Abstract:

The substantial Greenshell mussel aquaculture industry in New Zealand is heavily reliant on one major source of wild spat at Ninety Mile Beach, at the northern end of the country. The spat of *Perna canaliculus* arrives intermittently at the beach attached to seaweed and other debris whereupon it is harvested for seeding mussel farms around the country. Periods of low or non-existent spat arrival at the beach have caused major disruption to the aquaculture production of this species. In an attempt to better understand the daily, monthly, and inter-annual patterns in the arrival of spat at Ninety Mile Beach, the harvesting records of spat harvesters for 1990 to 1999 were analyzed in relation to historical records of wind speed and direction, tidal range, water temperature, and modeled swell height and direction. For the long-term data set, spatfall events and the amount of spatfall increased markedly with strong offshore winds. On days with high tidal range, there tended to be an increase in the amount of spatfall, but this trend was not significant statistically. Daily and seasonal water temperature records did not show a significant effect on the timing or the scale of spatfall events. However, low swell height in the onshore direction was associated with a significant increase in spatfall events and amounts. Within the 9 year data set, storm events (wind speeds > 20 m s⁻¹) were most frequent during May to October. An average lag time of 4 months was found between peak storm events and the subsequent peak in spatfall events and amounts of spatfall occurring in September to October. Years with a greater number of storm events were also associated with significantly higher number of spatfall events and amounts of spatfall. Storminess and water temperature are associated with El Niño/La Niña episodes, which greatly influence the wind climate of New Zealand. During El Niño periods mussel farm managers could greatly reduce their risk of a shortfall in natural spat supply interrupting mussel production by securing sufficient spat to stock their farms from the large but less frequent spatfall events. Overall, the results provide valuable insight into possible ecological and oceanographic processes involved in spat arrival and will help with better utilization of the spat resource for this major mussel aquaculture industry.

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SELECTION OF BACTERIA AND THE EFFECTS OF BACTERIAL TREATMENT OF ATLANTIC HALIBUT (*HIPPOGLOSSUS HIPPOGLOSSUS* L.) EGGS AND LARVAE

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Aquaculture 302(3-4): 219-227

Abstract:

Bacteria dominating the cultivable gut community of overall successful first feeding halibut (*Hippoglossus hippoglossus* L.) larvae were tested for their in vitro growth inhibition activity against selected fish pathogenic bacteria and isolates dominating the cultivable gut community of larvae with an overall poor success. A mixture containing equal numbers of three isolates was selected for the treatment of halibut eggs through repeated bathing, and larvae through grazing of live prey in a mixture of the selected isolates prior to offering to larvae. The isolates were found as a part of the dominating bacterial community of treated eggs and treatment was not found to affect egg survival. Improved larval survival was observed as a result of offering bacteria-treated live prey to larvae, and improved larval growth was observed in one of the two experiments that were carried out in commercial size production units. The bacterial community structure of the live prey, analysed using PCR and denaturing gradient gel electrophoresis, was only partly reflected in larvae after one week in feeding. A successful colonization of fertilized eggs by the isolates used for treatment entails the possibility to establish a favourable bacterial environment already prior to hatching.

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THE EFFECT OF DIETARY DOCOSAHEXAENOIC ACID (DHA) ON GROWTH, SURVIVAL AND PIGMENTATION OF CALIFORNIA HALIBUT *PARALICHTHYS CALIFORNICUS* LARVAE (AYRES, 1810)

Verónica Vizcaíno-Ochoa, Juan P. Lazo, Benjamín Barón-Sevilla, Mark A. Drawbridge-2010
Aquaculture 302(3-4): 228-234

Abstract:

The California halibut (*Paralichthys californicus*) is a good candidate for aquaculture due to its good growth, survival and high commercial value. Several farms in the Western coast of North America are currently evaluating the potential of this species under commercial conditions. However, one of the main problems in the production of juveniles for commercial purposes is the high percentage of malpigmented fish obtained after metamorphosis (up to 80%). This problem seems to be related, among other things, to nutritional deficiencies during the larval period, in particular to the quantities and proportions of highly unsaturated fatty acids (HUFAs) in the diet. As a first approach to reduce malpigmentation, improve growth and determine the requirement for DHA in California halibut late larvae, we evaluated the effect of four levels of dietary DHA (0, 1, 2, and 4% of total fatty acids in the diet) on growth, survival, weaning success and pigmentation. DHA was administered to the larvae through enriched *Artemia metanauplii*. Larvae standard length and wet weight were taken during the initial stage of *Artemia metanauplii* supplementation (18 days post hatch, dph); at the beginning of the weaning period (50 dph); and at the end of the experiment (75 dph). We quantified the amounts of total fatty acids in 18 and 50 dph larvae. No significant differences on growth, survival and pigmentation as a result of increasing dietary DHA levels were found in 50 dph recently settled juveniles. However, larvae fed the highest DHA level resulted in the highest growth and survival at the end of the experiment (75 dph). Additionally, highest weaning success was achieved with this treatment. Significantly higher numbers of normally pigmented fish (ca., 33%) were obtained with the highest DHA level at 75 dph compared to the low DHA levels (0 and 5%). However, since this treatment resulted in the highest survival, part of the population had abnormal pigmentation (ca., 30% of the population). Based on a second order polynomial regression, the recommended DHA level in the diet for pre-metamorphic larvae to attain adequate growth and survival as estimated here for recently settled California halibut at 50 dph was 1.21% DHA of total fatty acids (TFA). However, for post-metamorphosis fish (75 dph) highest pigmentation rates, growth and survival are obtained with 2.40% DHA of TFA in the diet during the *Artemia* feeding period.

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SPERM COMPETITION BETWEEN ALTERNATIVE REPRODUCTIVE TACTICS OF THE ATLANTIC SALMON IN VITRO

Tomislav Vladić, Lars A. Forsberg, Torbjörn Järvi-2010

Aquaculture 302(3-4): 265-269

Abstract:

The maintenance of brood stock in appropriate conditions is an important requirement for the production of high quality offspring. In this study, we investigated fertility of the two life history forms of Atlantic salmon males, precocious parr, brought up in breeding tanks in the hatchery and anadromous, migratory sea-ranched males, caught when returning to the home river. The sperm quality was assessed by experiments between equal amount of sperm from one adult and one parr male in competition to fertilize eggs of a single female. The paternity was determined by a microsatellite analysis. Parr males achieved greater reproductive success than anadromous males under competition, and anadromous adults had greater fertility in controls as compared to the sperm competition situation. In total, parr males achieved 3.6 times greater fertilization success than anadromous males. Sperm ATP content contributed significantly to male fertility. Our results provide evidence that ejaculates of

precociously mature Atlantic salmon parr are of increased quality as an adaptation to high sperm competition intensity due to better maintenance in the fish farm than in the wild.

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EFFECTS OF DIETARY VITAMIN A ON BROODSTOCK PERFORMANCE, EGG QUALITY, EARLY GROWTH AND RETINOID NUCLEAR RECEPTOR EXPRESSION IN RAINBOW TROUT (*ONCORHYNCHUS MYKISS*)

Stéphanie Fontagné-Dicharry, Emilie Lataillade, Anne Surget, Jeannine Brèque, José-Luis Zambonino-Infante, Sadasivam J. Kaushik-2010

Aquaculture 303(1-4): 40-49

Abstract:

The objective of the present study was to investigate the effects of dietary vitamin A supplementation on reproduction and fry quality of rainbow trout (*Oncorhynchus mykiss*). Two feeding trials were conducted with rainbow trout broodstock at a water temperature of 7 °C over a 6-month period prior to spawning. Three practical diets based on fish meal and fish oil (trial 1) and on plant ingredients (trial 2) were used. Diets 1A0, 1A20 and 1A700 were supplemented with 0, 20 and 700 IU/g vitamin A supplied as retinyl acetate resulting in dietary vitamin A contents of 45, 59 and 627 IU/g in trial 1; whereas, diets 2A0, 2A20 and 2A200 supplemented with 0, 20 and 200 IU/g vitamin A displayed total vitamin A contents of 21, 39 and 194 IU/g diet in trial 2.

In both trials, gonadosomatic indices were not significantly different among dietary groups. Egg size remained unaffected by the levels of vitamin A in the broodstock diets. In both feeding trials, the levels of retinyl palmitate and retinol, the two main storage forms of vitamin A, were significantly higher in oocytes from 1A700 and 2A200 groups compared to other dietary groups; whereas, the level of retinoic acid remained unaffected. In trial 2, broodstock females fed diet 2A200 showed better fecundity than females fed diets 2A0 and 2A20. A significant decline in survival from the eyed stage onwards was noticeable in group 1A700 in trial 1 but no differences were found in trial 2. At the swim-up stage, trout fry from broodstock fed diet 1A20 displayed significantly improved growth rates compared to other dietary groups in trial 1; whereas, in trial 2, trout fry from 2A200 group exhibited the best growth performance. The expression of genes coding for retinoid receptors and other associated nuclear receptors was not significantly different among dietary groups at any developmental stage considered and no skeletal malformation was recorded in offspring.

The results indicate that feeding rainbow trout broodstock a high level of vitamin A induced mortality at embryonic stages but was not teratogenic as no skeletal malformation was recorded at late developmental stages. However, quite high levels of vitamin A (60–200 IU/g) were necessary to sustain fecundity of broodstock and early growth of offspring.

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ULTRASOUND ENHANCED PERMEATION OF METHANOL INTO ZEBRAFISH, *DANIO RERIO*, EMBRYOS

Sawitri Silakes, Amrit N. Bart-2010

Aquaculture 303(1-4): 71-76

Abstract:

Only vitrification is likely to result in successful cryopreservation of teleost embryos, requiring a high level of cryoprotectant penetration into the cell. This study determined the effect of ultrasound on methanol (MeOH) penetration in three types (dechorionated, soft chorion and intact embryos) and three stages (90% epiboly, bud and 4-somite stages) of zebrafish (*Danio rerio*) embryos. Three types of embryos were first exposed to varying levels of methanol and optimized for survival. Dechorionated embryos were then exposed to 20%, soft chorion embryos were exposed to 30% and the intact embryos were exposed to 40% methanol for 3 min with or without ultrasound treatment. High performance

liquid chromatography (HPLC) was used to measure methanol levels in the treated embryos. The total MeOH recovery from zebrafish embryo samples spiked with MeOH was $100.62 \pm 3.99\%$. The overall penetration of methanol into zebrafish embryo ranged from $85.3 \pm 8.1 \mu\text{mol}$ (in soft chorion embryos at 90% epiboly stage treated with ultrasound) to $3.1 \pm 1.0 \mu\text{mol}$ (in dechorionated embryos at 90% epiboly stage treated without ultrasound). Ultrasound clearly helped to improve the permeation level of methanol into zebrafish embryos ($p < 0.05$). A high level of methanol was found in soft chorion embryos. Dechorionated embryos had the highest relative difference in methanol levels between ultrasound and controls. Although, ultrasound significantly affected survival ($p < 0.05$), survival was generally high ($64 \pm 2\%$ to 100%). Even the highest level of methanol ($85.3 \pm 8.1 \mu\text{mol}$) in the best treatment was far lower than that needed for vitrification. Further studies using novel and non-invasive methods are needed to achieve greater methanol penetration for successful vitrification.

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OUT-OF-SEASON GAMETE PRODUCTION IN STRONGYLOCENTROTUS DROEBACHIENSIS: PHOTOPERIOD AND TEMPERATURE MANIPULATION

Nicole T. Kirchoff, Stephen Eddy, Nicholas P. Brown-2010

Aquaculture 303(1-4): 77-85

Abstract:

The natural spawning of *Strongylocentrotus droebachiensis* is limited to late winter into early spring. In order to reduce the cost of urchin production, out-of-season gamete production is necessary. This study attempted to condition broodstock for out-of-season gamete production through the use of two different photoperiod and temperature regimes: a Constant Spring and an Advanced year; hypothesized to induce maturation through suspension or advancement of the gametogenic cycle, respectively. Viable gametes were successfully produced out-of-season with fertilization and hatch rates equal to or surpassing the levels recorded for wild urchin's in-season. In addition, specific fecundity was significantly higher in out-of-season conditioned urchins compared to the in-season wild urchins. Histological techniques proved that gametogenesis was both suspended and advanced through manipulation of the photoperiod and temperature. In addition, several important observations were made regarding reproductive physiology in the green sea urchin, the implications of which are important to the economic development of the green sea urchin aquaculture industry and management of wild stocks.

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REPRODUCTIVE PERFORMANCE AND OFFSPRING QUALITY OF THE FIRST AND THE SECOND BROOD OF FEMALE SWIMMING CRAB, PORTUNUS TRITUBERCULATUS

Xugan Wu, Yongxu Cheng, Chaoshu Zeng, Chunlin Wang, Zhaoxia Cui-2010

Aquaculture 303(1-4): 94-100

Abstract:

Although the swimming crab *Portunus trituberculatus* females can spawn up to 5 times during a reproductive season, only larvae hatched from the first brood are normally used for larval culture in commercial hatcheries. With the increasing price of swimming crab broodstock and decreasing availability of wild stock, there are increasing interests in the possible utilization of the second brood for larval culture. However, to date, no detailed study has been conducted to assess the reproductive performance and offspring quality from the second brood of female *P. trituberculatus*. The present study was conducted to compare reproductive performance and offspring quality of the first (1st) and the second brood (2nd) produced by female *P. trituberculatus* during a reproductive season. No significant differences in several important reproductive parameters, including percentage of females spawned, percentage of berried females successfully hatched, egg production per female and fecundity per unit body weight, was found between the first and the second brood. However, the first brood had significantly higher individual egg wet weight (1st = $40.54 \mu\text{g}$; 2nd = $31.40 \mu\text{g}$), egg dry weight (1st = $13.51 \mu\text{g}$; 2nd = $10.55 \mu\text{g}$) and hatchability (1st = 78.41% ; 2nd = 66.04%), and significantly higher

survival (1st = 88.31%; 2nd = 65.38%) and faster development (1st = 3.82 days; 2nd = 4.34 days) from zoea I to zoea II and showed better starvation resistance in newly hatched larvae (1st = 5.03 days; 2nd = 2.98 days) than the second brood ($P < 0.05$). Furthermore, the newly hatched zoea I from the first brood also had significantly higher total lipids, 16:1n- 7, 18:1n- 9, threonine (Thr) and arginine (Arg) contents, but lower levels of 18:0 and histidine (His) ($P < 0.05$) as compared to those of the second brood. These biochemical differences are likely linked to different larval quality of the first and the second brood.

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THE EFFECT OF DIETARY PHOSPHATIDYLCHOLINE/PHOSPHATIDYLINOSITOL RATIO ON MALFORMATION IN LARVAE AND JUVENILE GILTHEAD SEA BREEM (*SPARUS AURATA*)

E. Sandel, O. Nixon, S. Lutzky, B. Ginsbourg, A. Tandler, Z. Uni, W. Koven-2010

Aquaculture 304(1-4) : 42-48

Abstract:

Malformation in commercially raised fish, such as cranial, vertebral and gill cover deformities is a major factor reducing their market value. Although these deformities are most apparent in the juvenile and adult stages they may originate from suboptimal nutrition during the critical larval rearing stage. Previous research hypothesized that dietary phosphatidylinositol (PI) was more effective in reducing deformities than the main membrane phospholipid, phosphatidylcholine (PC). Consequently, the aim of this study was to test the effect of different dietary ratios of PC and PI fed to the gilthead sea bream (*Sparus aurata*) larvae, on developmental performances in juvenile fish in terms of survival, growth and malformation rate.

Four microdiet (MD) treatments, that differed in their PC/PI ratio and replaced 75% of the Artemia ration (wt/wt), were fed to 20–34 dph (days post hatching) sea bream larvae. In addition to the high PC/PI or low PI containing MD control, a commercial reference treatment (100% Artemia ration) was also given. At 40 dph, the larvae were graded in all treatments into small (< 1.3 mg dry wt larva⁻¹) and large (> 2.9 mg dry wt larva⁻¹) larvae, in order to test if growth rate influenced treatment effect throughout development to 141 dph.

There was no marked ($P > 0.05$) treatment effect on growth rate in 40 dph larvae. On the other hand in later juvenile development (67 dph), decreasing dietary PC/PI ratio contributed to significantly ($P < 0.05$) better growth and ($P > 0.05$) higher survival. Moreover, reducing dietary PI markedly ($P < 0.05$) increased jaw (cranial) deformity in both size groups at 67 dph which may have adversely affected juvenile feeding on a dry hard starter feed. Conversely, increasing dietary PI (reducing PC/PI ratio) showed a non-significant trend of increased skeletal deformity which was markedly ($P < 0.05$) higher in faster growing larvae in all MD treatments. Although there was no clear effect of PC/PI ratio on gill cover deformity rate, there was a size dependent susceptibility to this deformity where smaller larvae showed the highest incidence of this malformation. Osteocalcin (BGP) mRNA levels correlated well ($R^2 = 0.964$) with development in the faster growing fry fed the high PI diet. Higher production of BGP may have reduced ($P < 0.05$) the jaw deformity while tending to cause over-mineralization and deformity of the skeleton. The results suggest an effective dietary PC/PI ratio of 1.28 for sea bream larvae during culture.

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EFFECT OF DIETARY BACILLUS SPP. AND MANNAN OLIGOSACCHARIDES (MOS) ON EUROPEAN LOBSTER (*HOMARUS GAMMARUS* L.) LARVAE GROWTH PERFORMANCE, GUT MORPHOLOGY AND GUT MICROBIOTA

Carly L. Daniels, Daniel L. Merrifield, Dominic P. Boothroyd, Simon J. Davies, Jan R. Factor, Katie E. Arnold-2010

Abstract:

The effect of dietary application of a commercial probiotic (*Bacillus* spp.) and mannan oligosaccharides (MOS), used singularly and combined, on the survival, growth performance and feed cost-benefit of larval *Homarus gammarus* was assessed. Un-supplemented *Artemia* (control) or *Artemia* enriched with probiotics (100 mg l⁻¹ *Bacillus* spp.), MOS (12 mg l⁻¹) or probiotics + MOS (100 mg l⁻¹ *Bacillus* spp. + 12 mg l⁻¹ MOS) was fed to four replicate groups of zoeal I lobsters for 30 days. Carapace length and weight of five *H. gammarus* from each replicate was recorded on 1, 3, 9, 13, 18 days post hatch (dph) and for post-larval condition at 18 dph. Additionally, moulting success was recorded from 14 to 30 dph with survival calculated at 30 dph. Morphological analysis of the posterior intestine was also conducted on larval and post-larval *H. gammarus* using light and electron microscopy. In a secondary experiment the effects on gut microbiota were assessed using both culture-dependent and culture-independent methods. After 18 dph, larval *H. gammarus* fed a diet containing *Bacillus* spp + MOS had significantly ($P < 0.01$) improved weight gain, carapace length, weight to carapace length ratio, specific growth rate (SGR), food conversion ratio (FCR) and post-larval condition, compared to all other groups. The individual supplementation of either *Bacillus* or MOS also significantly improved growth parameters, survival and post-larval condition compared to the control group, but to a lesser extent. Survival of all groups was significantly ($P < 0.01$) elevated after 30 days compared to the control group. Light microscopy demonstrated no significant increases in gut absorptive surface area in larvae or post-larvae receiving biotic supplemented diets. However, electron microscopy revealed significant increases in microvilli length and density in larval and post-larval *H. gammarus* fed biotic supplemented diets compared to the control group ($P < 0.05$). Culture-based analysis of gut microbiota demonstrated probiotic *Bacillus* spp. colonisation in *Bacillus* and *Bacillus* + MOS fed larvae. Denaturing gradient gel electrophoresis of PCR-amplified 16S rRNA revealed that microbial species richness and diversity was reduced in *Bacillus* + MOS fed post-larval lobsters. Subsequently the microbial profiles of *Bacillus* + MOS were the most dissimilar to the control group. Improvements seen in the present study appear to be an amalgamation of effects highlighted with the individual use of *Bacillus* and MOS, and so are probably additive rather than synergistic in nature.

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REPRODUCTIVE EFFORT OF PACIFIC OYSTERS: A TRAIT ASSOCIATED WITH SUSCEPTIBILITY TO SUMMER MORTALITY

Arnaud Huvet, Julien Normand, Elodie Fleury, Virgile Quillien, Caroline Fabioux, Pierre Boudry-2010
Aquaculture 304(1-4) : 95-99

Abstract:

Summer mortality of the Pacific oyster *Crassostrea gigas* is the result of a complex interaction between oysters, their environment and their pathogens. The physiological status of an oyster, especially its reproductive status, is suspected to play a significant role in the outcome of this interaction. As genetic variability exists for susceptibility to summer mortality, resistant (R) and susceptible (S) oyster lines were produced using a divergent selection scheme. The present paper reports a histological study on gonad area, which is representative of reproductive effort, in randomly chosen five R and five S oyster lines. The R lines showed a significantly lower gonad area than the S lines ($P < 0.001$), with an estimated mean difference of 12.5%, whereas, taken together, R and S lines showed a similar distribution of gametogenic stages when sampled. Considering the lines separately, the significant difference in gonad area went up to 24% between R and S lines. The present data confirm and strengthen the negative correlation between reproductive effort and resistance to summer mortality observed in previous studies. Summer mortality of *C. gigas* in France could, therefore, be partly due to a physiological disorder and metabolic disturbance in oysters associated with their reproductive effort. This does not, however, imply a direct link between the cost of reproduction and mortality because other causal factors, such as pathogenic agents, could be the primary causal factors.

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OPTIMISATION OF LARVAL CULTURE OF THE MUSSEL MYTILUSEDULIS (L.)

Thomas H. Galley, Frederico M. Batista, Richard Braithwaite, Jon King, Andy R. Beaumont-2010
Aquaculture International 18(3): 315-325

Abstract:

The blue mussel *Mytilus edulis* is a commercially important species whose fishery and culture generally relies on natural spat collection. Hatchery-production could provide an alternative source of seed, enabling reliable expansion of the industry. Mussel spawning and larval rearing trials were carried out to optimise elements of hatchery production. Culturing fertilised eggs at low density (20–200 eggs cm⁻²) rather than high density (400–720 eggs cm⁻²) significantly improved the quality of first veliger larvae and differences in this improvement were evident between the eggs from different females (maternal effects). Veliger larval growth at 17 or 21°C was significantly faster than growth at 14°C. Feeding veliger larvae an identical total ration either daily or at 2–3 day intervals did not significantly affect their growth. Different microalgal diets (1: *Isochrysis* sp. (clone T-ISO), 2: *Chaetoceros calcitrans* forma *pumilus*, 3: *C. muelleri*, 4: mixed *Isochrysis* sp. (clone T-ISO) and *C. calcitrans* f. *pumilus*, and 5: mixed *Isochrysis* sp. (clone T-ISO) and *C. muelleri*) were tested on veliger larval growth and mixed diets outperformed single-species diets.

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STOCKING DENSITY FOR THE INTENSIVE REARING OF JUVENILE CRAYFISH, PACIFASTACUS LENIUSCULUS (ASTACIDAE), USING ARTEMIA NAUPLII TO SUPPLEMENT A DRY DIET FROM THE ONSET OF EXOGENOUS FEEDING

R. González, J. D. Celada, A. González, V. García, J. M. Carral, M. Sáez-Royuela-2010
Aquaculture International 18(3): 371-378

Abstract:

Recent advances in intensive rearing of astacid juvenile crayfish have greatly improved the results. This challenges the current application possibilities of the studies performed previously, and new research on density is required. A 100-day experiment was carried out under controlled conditions to evaluate density effects on survival and growth rates of juvenile crayfish in optimal conditions of feeding. Juvenile stage 2 *Pacifastacus leniusculus* were stocked in fibreglass tanks (1 m², 200 l water) at 20 ± 1°C and fed a dry diet for salmonids supplemented with restricted amounts of *Artemia nauplii*. Stocking densities were 100, 300, 600 and 1,000 crayfish m⁻². Mean survival rate was reduced significantly with increased stocking density, ranging from 86.33% (100 m⁻²) to 39.13% (1,000 m⁻²). All checks showed that at the lowest initial density (100 m⁻²) animals grew significantly faster those at higher densities, recording a final carapace length of 15.28 mm and weight of 1.08 g. Among the treatments of 300, 600 and 1,000 m⁻² no differences were found either in carapace length or in weight throughout the experimental period, with a final mean growth of 14 mm carapace length and 0.72 g weight. The final proportion of animals with chelae autotomy rose significantly with increasing stocking density, ranging from 14.44% (100 m⁻²) to 41.45% (1,000 m⁻²). This study shows that diet is a decisive factor for stocking successfully high densities under controlled conditions and provides useful information to set adequate densities in accordance with the production objectives.

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HERBAL BIOMEDICINES: A NEW OPPORTUNITY FOR AQUACULTURE INDUSTRY

Thavasimuthu Citarasu-2010
Aquaculture International 18(3): 403-414

Abstract :

Hormones, antibiotics, vitamins and several other chemicals have been tested in aquaculture operations for various remedies. Even though they give positive effects, they cannot be recommended due to their residual and other side effects. The alternative herbal bio-medicinal products in the aquacultural operations, that have the characteristics of growth promoting ability and tonic to improve the immune

system, act as appetite stimulators. They increase consumption, induce maturation, and have antimicrobial capability and also antistress characteristics that will be of immense use in the culture of shrimps and other fin fishes without any environmental and hazardous problems. Herbal compounds such as phenolics, polyphenols, alkaloids, quinones, terpenoids, lectines and polypeptides have been shown to be very effective alternatives to antibiotics and other synthetic compounds. The present paper is presented after a careful review of more than 50 herbal plants for their biological effects such as growth promotion, immunostimulation, antistress, antibacterial, antifungal, antivirals, appetite stimulators and aphrodisiac.

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EFFECTS OF STOCKING DENSITY, TEMPERATURE, AND SALINITY ON LARVAL SURVIVAL AND GROWTH OF THE RED RACE OF THE SEA CUCUMBER APOSTICHOPUS JAPONICUS (SELENKA)

Li Li, Qi Li-2010

Aquaculture International 18(3): 447-460

Abstract:

The red race of the sea cucumber *Apostichopus japonicus* was introduced into China from Japan for large-scale seed production because of its economic value. This paper reports the effects of stocking density, temperature, and salinity on survival and growth of early larvae before and after feeding, in order to establish conditions for optimal larval growth and production. To maximize the yield per unit of space, densities of 0.5–1 larvae/ml are recommended for non-feeding larvae, while 0.1–0.2 larvae/ml are best for feeding larvae. Higher survival and growth values were obtained for both non-feeding and feeding larvae at temperature ranges from 21 to 24°C. Larvae reared at a salinity of 30‰ always showed maximum growth and survival. Based on results of this study, a temperature range from 21 to 24°C and a salinity of 30 are considered optimal for early development of the red *A. japonicus*.

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WEANING OF THE WEDGE SOLE DICOLOGOGLOSSA CUNEATA (MOREAU): INFLUENCE OF INITIAL SIZE ON SURVIVAL AND GROWTH

Marcelino Herrera, Ismael Hachero-Cruzado, Catarina Oliveira, José F. Ferrer, José M. Márquez, Montserrat Rosano, Jose I. Navas-2010

Aquaculture International 18(3): 475-485

Abstract:

The weaning phase can be decisive in fish-culture viability. In this work, the relationship between the initial size and weaning success has been studied in wedge sole (*Dicologoglossa cuneata*). For each age (30, 50, and 70 days after hatching, DAH), two to three sizes were selected, and all were put on the same feeding schedule for 20 days. Each batch (three replicates) was sampled at 1, 10, and 20 days. Specific growth rate (SGR) and survival were compared at the end of the co-feeding period, after 10 days on dry feed only. The best results for survival and growth were found with the smallest larvae, and vice versa. The SGRs and survival rates recorded during the co-feeding period were higher (0.8–15.6 day⁻¹ and 68.3–97.8%) than those from the dry-food phase (0.9–4.7 day⁻¹ and 56.3–66.7%). Successful weaning (survival = 65% and SGR = 9.3 day⁻¹) is possible with 30 DAH larvae (7.6–8.1 mm and 3.9–4.6 mg). In conclusion, the most effective weaning would be possible at 30 DAH, implying significant *Artemia* savings (25–50%).

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PENAEUS MONODON LARVAE CAN BE PROTECTED FROM VIBRIO HARVEYI INFECTION BY PRE-EMPTIVE TREATMENT OF A REARING SYSTEM WITH ANTAGONISTIC OR NON-ANTAGONISTIC BACTERIAL PROBIOTICS

Srinivas Somnath Pai, Abdulaziz Anas, Natamai Subramaniam Jayaprakash, Prabhakaran Priyaja, Balachandran Sreelakshmi, Radhakrishnan Preetha, Rosamma Philip, Ambat Mohandas, Isaac Sarogeni Bright Singh-2010

Aquaculture Research 41(6): 847 - 860

Abstract:

This study shows that the disease resistance and survival rate of *Penaeus monodon* in a larval rearing systems can be enhanced by supplementing with antagonistic or non-antagonistic probiotics. The antagonistic mode of action of *Pseudomonas* MCCB 102 and MCCB 103 against vibrios was demonstrated in larval mesocosm with cultures having sufficient concentration of antagonistic compounds in their culture supernatant. Investigations on the antagonistic properties of *Bacillus* MCCB 101, *Pseudomonas* MCCB 102 and MCCB 103 and *Arthrobacter* MCCB 104 against *Vibrio harveyi* MCCB 111 under in vitro conditions revealed that *Pseudomonas* MCCB 102 and MCCB 103 were inhibitory to the pathogen. These inhibitory properties were further confirmed in the larval rearing systems of *P. monodon*. All these four probionts significantly improved larval survival in long-term treatments as well as when challenged with a pathogenic strain of *V. harveyi* MCCB 111. We could demonstrate that *Pseudomonas* MCCB 102 and MCCB 103 accorded disease resistance and a higher survival rate in *P. monodon* larval rearing systems through active antagonism of vibrios, whereas *Bacillus* MCCB 101 and *Arthrobacter* MCCB 104 functioned as probiotics through immunostimulatory and digestive enzyme-supporting modes of action.

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LHRHA-INDUCED OVULATION OF THE ENDANGERED-CASPIAN BROWN TROUT (*SALMO TRUTTA CASPIUS*) AND ITS EFFECT ON EGG QUALITY AND TWO SEX STEROIDS: TESTOSTERONE AND 17 α -HYDROXYPROGESTRONE

Ahmad Noori, Bagher Mojazi Amiri, Alireza Mirvaghefi, Daniel W. Baker-2010

Aquaculture Research 41(6): 871 - 877

Abstract:

To induce synchronized ovulation, migrating wild Caspian brown trout (*Salmo trutta caspius*) females were treated with two interperitoneal injections of Des-Gly10, d-Ala6 LHRH (LHRHa), given 3 days apart. Two injections of 100 $\mu\text{g kg}^{-1}$ body weight of this hormone effectively induced ovulation. Within 27 days from the second injection, all fish injected with 100 $\mu\text{g kg}^{-1}$ LHRHa had ovulated compared with 54.5% of the controls. The mean time to ovulation was reduced significantly ($P < 0.05$) from 31.67 \pm 4.84 days in control fish and 28.83 \pm 7.31 days in sham-treated fish to 16.36 \pm 1.61 days in fish injected with 100 $\mu\text{g kg}^{-1}$ LHRHa. The fertilization rate in 50 and 100 $\mu\text{g kg}^{-1}$ LHRHa-injected fish was significantly lower than that in the control fish ($P < 0.05$). In fish injected with 50 and 100 $\mu\text{g kg}^{-1}$ LHRHa, significant ($P < 0.05$) changes in testosterone (T) and 17 α -hydroxyprogesterone (OHP) levels were observed. After the second LHRHa injection, the fish injected with 100 $\mu\text{g kg}^{-1}$ showed the highest serum levels of testosterone and OHP. These results demonstrate that the use of LHRHa can effectively reduce the mean time to ovulation and induce synchronized ovulation in Caspian brown trout.

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ENCAPSULATION CAPACITY OF ARTEMIA NAUPLII WITH CUSTOMIZED PROBIOTICS FOR USE IN THE CULTIVATION OF WESTERN KING PRAWNS (*PENAEUS LATUSULCATUS* KISHINOUE, 1896)

Ngo Van Hai, Nicky Buller, Ravi Fotedar-2010

Aquaculture Research 41(6): 893 - 903

Abstract:

The encapsulation capacity of *Artemia nauplii* with customized probiotics *Pseudomonas synxantha* and *Pseudomonas aeruginosa* for use in the cultivation of western king prawns (*Penaeus latisulcatus*) was investigated. Seven trials were conducted to investigate this encapsulation capacity in terms of *Artemia* survival and probiotic load in *Artemia*. Newly hatched *Artemia nauplii* at 250 nauplii mL⁻¹ were fed individual probiotics at 0, 10³, 10⁵ and 10⁷ colony-forming units (CFU) per millilitre, and mixtures of these two probiotics (10⁵ CFU mL⁻¹) at 30:70, 50:50 or 70:30 v/v in a medium of ozonated water (OW), tryptone soya broth (TSB), and a mixture of these media. The appropriate medium for encapsulation of probiotics by *Artemia nauplii* was the mixture of OW and TSB at 75:25 v/v; whereas, the use of OW or TSB alone was not effective. *Artemia nauplii* most effectively encapsulated the customized probiotics at 10⁵ CFU mL⁻¹. The results indicates that the encapsulation of *Artemia nauplii* is optimized by using a combination of *P. synxantha* and *P. aeruginosa* at 50:50 v/v in a media mixture of OW and TSB at 75: 25 v/v. *Artemia* should be harvested at 48 h when survival is still high (78%) and the probiotic load in *Artemia* is high (3×10^4 CFU nauplius⁻¹).

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