

EARLY LIFE STAGE SALINITY TOLERANCE OF WILD AND HATCHERY-REARED JUVENILE PINK SALMON *ONCORHYNCHUS GORBUSCHA*

A.M. Grant, M. Gardner, L. M. Hanson, A. P. Farrell, C. J. Brauner-2010

Journal of Fish Biology 77(6): 1282–1292

Abstract:

Salinity tolerance in wild (Glendale) and hatchery (Quinsam) pink salmon *Oncorhynchus gorbuscha* (average mass 0.2 g) was assessed by measuring whole body $[Na^+]$ and $[Cl^-]$ after 24 or 72 h exposures to fresh water (FW) and 33, 66 or 100% sea water (SW). Gill Na^+ , K^+ -ATPase activity was measured following exposure to FW and 100% SW and increased significantly in both populations after a 24 h exposure to 100% SW. Whole body $[Na^+]$ and whole body $[Cl^-]$ increased significantly in both populations after 24 h in 33, 66 and 100% SW, where whole body $[Cl^-]$ differed significantly between Quinsam and Glendale populations. Extending the seawater exposure to 72 h resulted in no further increases in whole body $[Na^+]$ and whole body $[Cl^-]$ at any salinity, but there was more variability among the responses of the two populations. Per cent whole body water (c. 81%) was maintained in all groups of fish regardless of salinity exposure or population, indicating that the increase in whole body ion levels may have been related to maintaining water balance as no mortality was observed in this study. Thus, both wild and hatchery juvenile *O. gorbuscha* tolerated abrupt salinity changes, which triggered an increase in gill Na^+ , K^+ -ATPase within 24 h. These results are discussed in terms of the preparedness of emerging *O. gorbuscha* for the marine phase of their life cycle.

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OCCURRENCE OF SKELETAL DEFORMITIES AND OSTEOLOGICAL DEVELOPMENT IN RED PORGY *PAGRUS PAGRUS* LARVAE CULTURED UNDER DIFFERENT REARING TECHNIQUES

F. J. Roo, C.-M. Hernández-Cruz, J.-A. Socorro, H. Fernández-Palacios, M.-S. Izquierdo-2010

Journal of Fish Biology 77(6): 1309–1324

Abstract:

The present study describes the osteological development and the occurrence of skeletal deformities in red porgy *Pagrus pagrus* larvae in relation to the intensification of the rearing system. Eggs obtained from natural spawning were cultured under two different rearing systems: intensive (100 eggs/l) in 2000 l and semi-intensive (mesocosm) system (5 eggs/l) in 40 000 l conico -cylindrical tanks. Fish samples were periodically collected along the development from hatching to juveniles at 95 days post hatching (dph). Osteological development, meristic counts and the presence of skeletal deformities were evaluated. Despite the external appearance of the juveniles being similar to wild standards, X-ray studies revealed a high number of fish (semi-intensive: 37.8%; intensive: 45.5%) with skeletal deformities. Regardless of the rearing system, no significant interaction was found between the per cent of the most common deformities, axial deviations (lordosis and presence of fused vertebrae). Cranial deformities and kyphosis incidences, however, were significantly higher in intensively cultured *P. pagrus*. Also, the fused vertebrae in these fish were located mainly in the caudal area instead of pre-haemal area for semi-intensively reared *P. pagrus*. Moreover, a significant interaction was found between the total number of vertebrae and the type of rearing system used; fish from the intensive system showing a higher number of fish with an extra vertebrae (10 abdominal + 15 caudal). Present results suggest a relationship among feeding sequence, osteological development and deformity incidence and location in *P. pagrus* larvae.

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GENETIC EFFECTS OF HATCHERY FISH ON WILD POPULATIONS IN RED SEA BREEM
PAGRUS MAJOR (PERCIFORMES, SPARIDAE) INFERRED FROM A PARTIAL SEQUENCE OF
MITOCHONDRIAL DNA

K. Hamasaki, S. Toriya, H. Shishidou, T. Sugaya, S. Kitada-2010
Journal of Fish Biology 77(9): 2123–2136

Abstract:

Variation in the mitochondrial DNA transcriptional control region sequence was investigated in wild and hatchery-released red sea bream *Pagrus major* from Kagoshima Bay, where an extensive hatchery-release programme has been conducted for >30 years. The programme has successfully augmented commercial catches in the bay (released juveniles have been produced from the captive broodstock, repeatedly used over multiple generations). Samples were also obtained from outside the bay, where limited stocking has occurred. Genetic diversity indices measured as number of haplotypes, haplotype richness, haplotype diversity and nucleotide diversity were lower in hatchery-released fish than in wild fish. Genetic differences in wild fish from the bay, especially in the inner bay, compared with fish from outside the bay were detected in terms of decreased genetic diversity indices and changed haplotype frequencies. Unbiased population pair-wise F_{ST} estimates based on an empirical Bayesian method, however, revealed low genetic differentiation between samples from the bay and its vicinity. Mixed stock identification analyses estimated the proportion of hatchery-released fish in wild populations in the inner and central bays at 39.0 and 8.7%, respectively, although the precision of the estimates was very low because of the small genetic differentiation between populations and relatively small sample sizes. Hence, the long-term extensive hatchery release programme has affected the genetic diversity of wild populations in the bay; however, the genetic effects were low and appeared to remain within the bay.

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ROTIFERS: EXCELLENT SUBJECTS FOR THE STUDY OF MACRO- AND
MICROEVOLUTIONARY CHANGE

Gregor F. Fussmann-2010

Hydrobiologia 662(1): 11-18

From the issue entitled "Rotifera XII: New aspects in rotifer evolution, genetics, reproduction, ecology and biogeography / N. Walz, R. Adrian, J.J. Gilbert, M.T. Monaghan, G. Weithoff & H. Zimmermann-Timm

Abstract:

Rotifers, both as individuals and as a phylogenetic group, are particularly worthwhile subjects for the study of evolution. Over the past decade molecular and experimental work on rotifers has facilitated major progress in three lines of evolutionary research. First, we continue to reveal the phylogenetic relationships within the taxon Rotifera and its placement within the tree of life. Second, we have gained a better understanding of how macroevolutionary transitions occur and how evolutionary strategies can be maintained over millions of years. In the case of rotifers, we are challenged to explain the evolution of obligate asexuality (in the bdelloids) as mode of reproduction and how speciation occurs in the absence of sex. Recent research with bdelloid rotifers has identified novel mechanisms such as horizontal gene transfer and resistance to radiation as factors potentially affecting macroevolutionary change. Third, we are finding that microevolutionary change can be sufficiently rapid to interact with ecological dynamics. Rotifers can be easily cultured, reproduce quickly, and occur at high levels of clonal, genetic diversity in nature. These features make them excellent eukaryotic model systems for the study of eco-evolutionary dynamics.

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GONAD DEVELOPMENT OF THE JAPANESE OYSTER *CRASSOSTREA GIGAS* IN A RECIRCULATING SYSTEM: FIRST STEP TOWARD THE DEVELOPMENT OF CONDITIONING AND MATURATION PROTOCOLS

Roger Frias, Manuel Segovia-2010

Journal of Shellfish Research 29(2): 303-308

Abstract:

This study analyzed the effect of water quality, different diets, and water temperature regimes in the conditioning and maturation of the Pacific oyster *Crassostrea gigas*. A recirculating aquaculture system prototype was used in this experiment and 2 different trials were performed. During the first trial, the effects of 3 different diets using different preserved concentrates of microalgae held at 20°C were tested: D1 (fed with *Tetraselmis* 3600), D2 (fed with *Isochrysis* 1800), and D3 (fed with a 50:50 mix of *Tetraselmis* and *Isochrysis*). During the second trial, 3 different temperature regimes—T1, 16–24°C; T2, 20°C; and T3, 24°C (fed with preserved concentrates of microalgae at a 50:50 *Tetraselmis* and *Isochrysis* food ratio)—were evaluated. For each one of these treatments, 43 organisms were used and every treatment was done in triplicate. Seven samplings were completed in 72 days. During each sampling period, 9 oysters from each treatment were collected, and a gonad sample was obtained for histology analysis to determine gonad development. The results obtained in this study indicate that the recirculation system prototype was able to keep optimum water-quality parameters, and the nitrogenous components stayed below toxic levels (<0.2 mg/L). For the first trial, D3 showed gonad maturation at day 72, which was significantly different from D1 and D2 ($P > 0.05$). In the second trial, no significant differences were detected in gonad development among the 3 temperature treatments. Even when D2, T1, and T3 showed no gonad maturation, a trend was detected. Thus, it can be concluded that the combination of diet and temperature allows the manipulation of the gonad development of *C. gigas*, and that recirculation systems are an efficient tool to maintain appropriate physical—chemical and environmental parameters.

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DEVELOPMENT OF GENOMIC RESOURCES FOR THE PHYLOGENETIC ANALYSIS OF THE BRACHIONUS PLICATILIS SPECIES COMPLEX (ROTIFERA: MONOGONONTA)

Javier Montero-Pau, Africa Gómez-2010

Hydrobiologia 662(1): 43-50

From the issue entitled "Rotifera XII: New aspects in rotifer evolution, genetics, reproduction, ecology and biogeography / N. Walz, R. Adrian, J.J. Gilbert, M.T. Monaghan, G. Weithoff & H. Zimmermann-Timm"

Abstract:

There has been a substantial leap forward in the quantity and quality of genomic resources available for research on rotifers in recent years. We used available genomic and bioinformatics resources to identify variable regions of the genome to design PCR and sequencing primers for the *Brachionus plicatilis* complex. We then tested their suitability for the study of systematics of this group. Eight amplified successfully for members of the *B. plicatilis* complex. We sequenced the amplified products, constructed a concatenated alignment (5,511 bp) and carried out phylogenetic analyses. The resulting tree, based on mitochondrial and nuclear genes for 11 clones of six species, was well supported even at relatively deep nodes, contrasting with results of previous studies using only mitochondrial genes, which provide little phylogenetic information for the deepest nodes. The same procedure could be used to design primers for more conserved regions to be used in a wider range of rotifer groups.

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LARVAL REARING AND SPAT PRODUCTION OF THE RAZOR CLAM *ENSIS SILIQUA* (BIVALVIA: PHARIDAE)

Fiz Da Costa, Dorotea Martínez-Patiño, Justa Ojea, Susana Nóvoa-2010

Abstract:

This work describes for the first time the larval and postlarval development of the razor clam *Ensis siliqua*, and aimed to standardize the techniques for hatchery production of this species' seed. The larvae were obtained from the induced spawning of a group of broodstocks under controlled laboratory conditions. Five larval batches per duplicate were cultured to produce postlarvae for nursery culture. Unfertilized eggs of *E. siliqua* were brown and spherical, and measured from 76.9–99.3 µm in diameter in the different larval batches. Fifteen-day-old larvae settled when a size of 361.5–414.8 µm was reached. Larval survival ranged between 20.0–52.7%, with an average of 39.4%. Mortality was high from settlement until 1 mo of cultivation. The razor clam seed reached lengths of 20.9 ± 2.7 mm and 39.0 ± 4.0 mm at 3 mo and 6 mo, respectively. Substrate experiments during nursery culture showed that better survival was achieved when spat cultures were performed without substratum.

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WORLD ABALONE SUPPLY, MARKETS, AND PRICING

Peter A. Cook, H. Roy Gordon-2010

Journal of Shellfish Research 29(3): 569-571

Abstract:

In recent years, abalone fisheries around the world have shown similar trends, with landings rapidly increasing and then falling as abalone stocks were overfished. A rapid increase in the illegal exploitation of abalone has contributed to population crashes and, sometimes, to the complete decommercialization of abalone fishing industries. Concurrently, however, there has been a rapid increase in abalone farming and, to some extent, farm production has helped to ensure that the overall relationship between supply and demand has not changed too much from what existed during the 1970s. What has changed, however, is the overall availability of abalone on the world market, which has almost doubled from about 20,000 mt during the 1970s to almost 40,000 mt in 2008. Also changed is the species mix of the overall demand, with production and demand in China being a dominant factor but, unfortunately, not necessarily impacting as much as many people expected on overall world demand. The reasons for this will be explored and suggestions will be made on what might be appropriate responses from the abalone farming industry.

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USE OF ALGAL DIETS TO AID EARLY WEANING IN THE ABALONE *HALIOTIS IRIS*

Matt Dyck, Rodney Roberts, Andrew Jeffs-2010

Journal of Shellfish Research 29(3): 613-620

Abstract:

A major bottleneck in the aquaculture of abalone is feeding during the nursery stage of production, from larval settlement to approximately 10 mm in shell length (SL). Most commercial abalone nurseries settle larvae onto vertical plastic plates coated in a film of algae that acts as a settlement cue and an important postsettlement food source. As abalone grow, their food consumption increases exponentially, and it becomes increasingly difficult to maintain an adequate supply of algal food. Even grazing-resistant algae, like *Ulva* spp., are often depleted when abalone reach 3–5 mm SL, requiring nurseries to provide additional algae or to wean the juveniles prematurely onto manufactured diets. Early weaning typically results in slower growth and higher mortality relative to live algal feeds. The current study tested various algal species to aid early weaning of the abalone *Haliotis iris*. Two experiments were conducted pairing nursery tanks of similar environmental conditions to test a total of three algal treatments: (1) the benthic diatom *Nitzschia longissima*, (2) a film of *N. longissima* and *U. lens*, and (3) a film of *N. longissima* and a naturally occurring mixture of green algal species that remained on weaning tank surfaces after a previous cohort of abalone had been harvested from the tank. Algae were established prior to the introduction of juvenile (SL, 2–7 mm) *H. iris* at a density of 15,000–30,000 abalone per tank (2.5 m³ water volume, 13 m² surface area). The abalone were raised to

approximately 15 mm SL with twice-weekly supplementary feeding of suspended *N. longissima* during the first 4 wk, and the addition of manufactured feed 3 times a day for the duration of the experiment. The *N. longissima* films were completely removed by grazing within 4–5 days whereas the green algal films persisted for 60–120 days. Despite the difference in resilience of algal films, there was no significant difference ($P > 0.05$) in abalone performance between treatments 1 (98.7% survival, growth of 85 $\mu\text{m}/\text{day}$) and 2 (98.3% survival, growth of 92 $\mu\text{m}/\text{day}$). In a second experiment, treatment 2 provided slightly greater growth than the mixed algal film of treatment 3 (101 $\mu\text{m}/\text{day}$ vs. 96 $\mu\text{m}/\text{day}$, $P = 0.034$), but survival rates were similar (98.2% vs. 98.3%, $P = 0.86$). All 3 algal treatments offer practical and effective means of weaning *H. iris* onto manufactured feeds at an SL of more than 3 mm. However, both treatments 1 and 3 can be established more rapidly than *U. lens* and are therefore likely to be more efficient for commercial weaning of *H. iris* and potentially for other species of abalone. (Leigh Marine Laboratory, University of Auckland, PO Box 349, Warkworth 0941, New Zealand; email of Rodney Roberts: rodney.roberts@oceanzblue.co.nz)

STABLE ISOTOPE ANALYSES AS A TOOL TO EXAMINE POSTLARVAL DIETS OF HALIOTIS DISCUS HANNAI

Nam-II Won, Tomohiko Kawamura, Hideki Takami, Daisuke Muraoka, Yoshiro Watanabe-2010

Journal of Shellfish Research 29(3): 655-662

Abstract:

Studies on the feeding habits of postlarval abalone in their natural habitats have never been carried out primarily because of the lack of an investigating tool. In this study, carbon and nitrogen stable isotope ratios ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) were tested to detect nutritional sources during the early life stages of the abalone *Haliotis discus hannai* in laboratory experiments to evaluate stable isotope analyses as an appropriate investigative tool for field studies. Fertilized eggs, larvae, and the parent spawner muscle were analyzed to detect a nutritional source during the nonfeeding period. The $\delta^{13}\text{C}$ of fertilized eggs showed slight depletion, about 2‰, compared with that of the parent muscle, and was not detectable after lipid extraction of fertilized eggs. As for the larvae, the isotopic values in postlarvae after settlement and metamorphosis were 2‰ more enriched, become similar to that of the spawner muscle. These results imply that the lipid portion of the eggs is the primary nutritional source during the nonfeeding period of abalone. Laboratory feeding experiments were conducted during which early postlarval abalone (approximately 1 mm in shell length (SL)) were reared for 7 days and supplied with 4 benthic diatoms respectively. The benthic diatom *Cylindrotheca closterium* produced high growth and survival rates for early postlarval abalone, and there was a significant isotopic shift in their $\delta^{13}\text{C}$, approaching that of *C. closterium*. Larger postlarval abalone (SL, >1.3 mm) were fed 1 of 5 benthic diatoms and a gametophyte of a macroalga, *Undaria pinnatifida*, for 21 days. Isotopic changes approaching the $\delta^{13}\text{C}$ of the provided benthic diatoms were detected from postlarvae fed the benthic diatoms *Achnanthes longipes*, *Cocconeis scutellum*, *C. closterium*, and the gametophyte of *U. pinnatifida*, which provided high survival rates and growth rates for the postlarvae. From these results, stable isotope ratios of postlarval abalone are suggested to be useful to infer the origin of the assimilated diets. The current study highlights that stable isotope ratios are a promising tool to investigate the diet composition of postlarval abalone in their natural habitat as well as in abalone hatcheries.

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THE EXPRESSION PATTERN OF DORMANCY-ASSOCIATED GENES IN MULTIPLE LIFE-HISTORY STAGES IN THE ROTIFER BRACHIONUS PLICATILIS

N. Y. Denekamp, R. Reinhardt, M. W. Albrecht, M. Drungowski, M. Kube, E. Lubzens-2010

Hydrobiologia 662(1): 51-63

From the issue entitled "Rotifera XII: New aspects in rotifer evolution, genetics, reproduction, ecology and biogeography / N. Walz, R. Adrian, J.J. Gilbert, M.T. Monaghan, G. Weithoff & H. Zimmermann-Timm"

Abstract:

Rotifer resting eggs retain their viability for several decades in a non-desiccated form and are of interest in discerning the processes associated with dormancy, since in most organisms this phenomenon is linked with desiccation. The expression pattern of candidate genes with biological functions associated with dormancy in several other organisms was examined in rotifers. High-throughput transcriptome profiling revealed three patterns of gene expression in resting eggs: (1) relatively highly expressed genes coding for LEA proteins and putative paralogs of the small heat shock protein family (shsp); (2) genes coding for ferritin (ferr), glutathione-S-transferase (gts) and HSP70, where some of the putative gene paralogs of these families show relatively high expression levels and other putative paralogs show relatively low expression levels in resting eggs; and (3) genes with relatively low expression levels in resting eggs, for trehalose-6-phosphate synthase (tps), fatty-acid binding proteins (fab) and of lipoprotein lipase (lpl) and the aquaporins gene family (aqp). Changes in the expression pattern of some members of putative gene families occurred during the obligatory dormant period of resting eggs. A transition was observed from an expression pattern of diapausing embryos to an expression pattern of amictic females, during hatching. Differences were also found in the expression pattern in the different types of females, especially in those carrying resting eggs, and in males compared with females. These results suggest putative functional significance to genes associated with dormancy in non-desiccated resting eggs. It could also be proposed that their occurrence in resting eggs is developmentally programmed to facilitate survival in case of desiccation.

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MOLECULAR EVOLUTION OF THE MEMBRANE ASSOCIATED PROGESTERONE RECEPTOR IN THE BRACHIONUS PLICATILIS (ROTIFERA, MONOGONONTA) SPECIES COMPLEX

Hilary A. Smith, David B. Mark Welch, Terry W. Snell-2010

Hydrobiologia (2011) 662:99–106

Abstract:

Many studies have investigated physiological roles of the membrane associated progesterone receptor (MAPR), but little is known of its evolution. Marked variations in response to exogenous progesterone have been reported for four brachionid rotifer species, suggesting differences in progesterone signaling and reception. Here we report sequence variation for the MAPR gene in the *Brachionus plicatilis* species complex. Phylogenetic analysis of this receptor is compared with relatedness based on cytochrome c oxidase subunit 1 sequences. Nonsynonymous to synonymous site substitution rate ratios, amino acid divergence, and variations in predicted phosphorylation sites are examined to assess evolution of the MAPR among brachionid clades.

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EFFECT OF FEMALE AGING ON THE MORPHOLOGY AND HATCHABILITY OF RESTING EGGS IN THE ROTIFER BRACHIONUS PLICATILIS MULLER

Hee-Jin Kim, Atsushi Hagiwara-2010

Hydrobiologia 662:107–111

Abstract:

This study examined the morphology and hatchability of *Brachionus plicatilis* resting eggs as a function of the aging of maternal fertilized mictic females. One-hundred twenty fertilized *B. plicatilis* (Australian strain) were individually cultured and monitored daily until death. All cultures were maintained at 25°C, 11 ppt, and fed the micro-algae *Tetraselmis tetraele*. Resting eggs produced by the females were investigated using two parameters: egg morphology, and hatching rate. Under these culture conditions, females normally produce 1–6 (mean \pm SD = 2.7 ± 1.2) resting eggs during their lifetime. However, the number of resting eggs with abnormal morphology increased as a function of maternal age. Among resting eggs with normal morphology ($n = 225$), 82.2% were produced during the first and second spawning, and had hatching rates of more than 60%, while the hatching rates were

below 30% in resting eggs with a spawning order of [2]. Thus, the quality of *B. plicatilis* resting eggs was negatively correlated with maternal age.

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AXENIC CULTURE OF BRACHIONUS PLICATILIS USING ANTIBIOTICS

Koushirou Suga, Yukari Tanaka, Yoshitaka Sakakura, Atsushi Hagiwara-2010

Hydrobiologia 662:113–119

Abstract:

The rotifer *Brachionus plicatilis* culture is composed of complex microcosms including bacteria, protozoans, algae, and fungi. Previous studies reported methods to establish axenic rotifer cultures, but further refinement of these techniques is needed, for molecular biological research which requires pure culture to isolate nucleic acids from rotifers only. In order to render rotifer culture axenic, we tested five antibiotics: ampicillin (Amp), chloramphenicol (Cp), kanamycin (Km), nalidixic acid (Na), and streptomycin (Sm) at 30–100 µg/ml. Except for Cp, which reduces rotifer reproduction, all other antibiotics at the tested concentrations did not affect rotifer reproduction or show any toxic effects. A rotifer disinfection method was finally established by treating the resting eggs with 0.25% (w/v) sodium hypochlorite (NaOCl) for 3 min, washing with sterilized sea water, and then exposing the neonates to an Amp, Km, Na, and Sm mixture. Using four nutrient media, we confirmed that this protocol renders the rotifer culture bacterial and fungus free. The axenic rotifer culture generated here is useful not only for genetic analysis of *Brachionus plicatilis*, but for studying the rotifer life cycle without bacterial influence.

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EFFECT OF EXPERIMENTAL METHODOLOGY ON ESTIMATION OF DENSITY AT SEX INITIATION IN CYCLICALLY PARTHENOGENETIC ROTIFERS

M. J. Carmona, N. Dimas-Flores, J. Montero-Pau, M. Serra-2010

Hydrobiologia 662:131–139

Abstract:

Sexual reproduction is density-dependent in several cyclically parthenogenetic rotifer species and induced by a chemical signal produced by the rotifers. As population density increases, the chemical accumulates in the medium until reaching a threshold that triggers sex. The density threshold for sex initiation is of ecological and evolutionary importance due to the large impact that timing of sex has on fitness in cyclical parthenogenetic rotifers. Here we study the effects of experimental methodology in combination with intrinsic features of the biological system on the estimation of the density required to produce enough of this chemical to initiate sex (i.e., density threshold). We have developed a theoretical model describing the dynamics of both population density and the concentration of the chemical signal. Model analysis showed that the time at which sex is recorded in an experiment, the initial density (i.e., experimental volume) and the population growth rate can affect the estimation of density thresholds. Results from an experiment in which density at sex initiation was tested for 29 clones of *Brachionus plicatilis* at two culture volumes, revealed that density threshold is negatively dependent on the experimental volume. We conclude that in order to estimate density thresholds so that they are comparable across studies, experimental methodology should be standardized.

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REVIEW

ALGAL PHOTOSYNTHESIS AS THE PRIMARY DRIVER FOR A SUSTAINABLE DEVELOPMENT IN ENERGY, FEED, AND FOOD PRODUCTION

Ida G. Anemaet, Martijn Bekker, Klaas J. Hellingwerf-2010

Marine Biotechnology 12(6): 619-629

Abstract:

High oil prices and global warming that accompany the use of fossil fuels are an incentive to find alternative forms of energy supply. Photosynthetic biofuel production represents one of these since for this, one uses renewable resources. Sunlight is used for the conversion of water and CO₂ into biomass. Two strategies are used in parallel: plant-based production via sugar fermentation into ethanol and biodiesel production through transesterification. Both, however, exacerbate other problems, including regional nutrient balancing and the world's food supply, and suffer from the modest efficiency of photosynthesis. Maximizing the efficiency of natural and engineered photosynthesis is therefore of utmost importance. Algal photosynthesis is the system of choice for this particularly for energy applications. Complete conversion of CO₂ into biomass is not necessary for this. Innovative methods of synthetic biology allow one to combine photosynthetic and fermentative metabolism via the so-called Photanol approach to form biofuel directly from Calvin cycle intermediates through use of the naturally transformable cyanobacterium *Synechocystis* sp. PCC 6803. Beyond providing transport energy and chemical feedstocks, photosynthesis will continue to be used for food and feed applications. Also for this application, arguments of efficiency will become more and more important as the size of the world population continues to increase. Photosynthetic cells can be used for food applications in various innovative forms, e.g., as a substitute for the fish proteins in the diet supplied to carnivorous fish or perhaps—after acid hydrolysis—as a complex, animal-free serum for growth of mammalian cells in vitro.

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MAXIMUM PHOTOSYNTHETIC YIELD OF GREEN MICROALGAE IN PHOTOBIOREACTORS

Jan-Willem F. Zijffers, Klaske J. Schippers, Ke Zheng, Marcel Janssen, Johannes Tramper, René H. Wijffels-2010

Marine Biotechnology 12(6): 708-718

Abstract:

The biomass yield on light energy of *Dunaliella tertiolecta* and *Chlorella sorokiniana* was investigated in a 1.25- and 2.15-cm light path panel photobioreactor at constant ingoing photon flux density (930 $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$). At the optimal combination of biomass density and dilution rate, equal biomass yields on light energy were observed for both light paths for both microalgae. The observed biomass yield on light energy appeared to be based on a constant intrinsic biomass yield and a constant maintenance energy requirement per gram biomass. Using the model of Pirt (New Phytol 102:3–37, 1986), a biomass yield on light energy of 0.78 and 0.75 $\text{g mol photons}^{-1}$ and a maintenance requirement of 0.0133 and 0.0068 $\text{mol photons g}^{-1} \text{ h}^{-1}$ were found for *D. tertiolecta* and *C. sorokiniana*, respectively. The observed yield decreases steeply at low light supply rates, and according to this model, this is related to the increase of the amount of useable light energy diverted to biomass maintenance. With this study, we demonstrated that the observed biomass yield on light in short light path bioreactors at high biomass densities decreases because maintenance requirements are relatively high at these conditions. All our experimental data for the two strains tested could be described by the physiological models of Pirt (New Phytol 102:3–37, 1986). Consequently, for the design of a photobioreactor, we should maintain a relatively high specific light supply rate. A process with high biomass densities and high yields at high light intensities can only be obtained in short light path photobioreactors.

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ANTIFREEZE PROTEIN IN ANTARCTIC MARINE DIATOM, *CHAETOCEROS NEOGRACILE*

In Gyu Gwak, Woong sic Jung, Hak Jun Kim, Sung-Ho Kang, EonSeon Jin-2010

Marine Biotechnology 12(6): 630-639

Abstract:

The antifreeze protein gene (Cn-AFP) from the Antarctic marine diatom, *Chaetoceros neogracile* was cloned and characterized. The full-length Cn-AFP cDNA contained an open reading frame of 849 bp and the deduced 282 amino acid peptide chain encodes a 29.2 kDa protein, which includes a signal peptide of 30 amino acids at the N terminus. Both the Cn-AFP coding region with and without the signal sequence were cloned and expressed in *Escherichia coli*. Recombinant Cn-AFPs were shown to display antifreeze activities based on measuring the thermal hysteresis and modified morphology of single ice crystals. Recombinant mature Cn-AFP showed 16-fold higher thermal hysteresis activity than that of pre-mature Cn-AFP at the same concentration. The ice crystal shape changed to an elongated hexagonal shape in the presence of the recombinant mature Cn-AFP, while single ice crystal showed a circular disk shape in absence of Cn-AFP. Northern analysis demonstrated a dramatic accumulation of Cn-AFP transcripts when the cells were subjected to freezing stress. This rapid response to freeze stress, and the antifreeze activity of recombinant Cn-AFPs, indicates that Cn-AFP plays an important role in low temperature adaptation.

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EFFECTS OF EYESTALK ABLATIONS ON GROWTH AND OVARIAN MATURATION OF THE FRESHWATER PRAWN *MACROBRACHIUM LANCHESTERI* (DE MAN)

K.N. Varalakshmi, Ravichandra Reddy-2010

Turkish Journal of Fisheries and Aquatic Sciences 10: 403-410

Abstract:

The effects of unilateral and bilateral eyestalk ablations on growth, ovarian maturation, carbohydrate, protein and energy contents were investigated in *Macrobrachium lanchesteri* (de Man). Daily growth as well as ovarian maturation were accelerated by eyestalk ablation in these prawns. The mortality in ablated prawns was more than that of intact ones. The biochemical components like carbohydrate and protein were altered by eyestalk ablations. The energy content of the different tissues were also showing some variations due to the absence of eyestalk hormones. Eyestalk ablation, especially unilateral eyestalk ablation could be used for inducing rapid growth and spawning in this particular species of freshwater prawns.

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GROWTH OF OVER-WINTERED AND PRE-SEASONALLY PRODUCED POST-LARVAE OF *PENAEUS SEMISULCATUS* IN THE SUBTROPICS

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Abstract:

Growth performance of *Penaeus semisulcatus* under semi-intensive and intensive systems in pond/tank culture conditions with pre-seasonally or over-wintered animals was studied in sub-tropical climate conditions of Turkey. In Pond A, the pre-seasonally produced post-larvae (PL) grew linearly from 0.2 g to 22.6 g in 140 days ($Y = 0.19 X - 2.69$; $R^2 = 0.97$) with DGR (daily growth rate) of 0.06 g day⁻¹. In Pond B, the over-wintered juveniles grew from 3.8 g to 30.5 g with DGR of 0.41 g during the first 30 days (weight gain of 14.42 g) and 0.13 g between 30 and 140 days (weight gain of 14.32 g). Regression equation for growth was calculated to be $Y = 9.62 \text{ Log } (X) + 4.51$ ($R^2 = 0.99$). Compensatory growth due to starvation or over-winter fasting has, for the first time, been demonstrated in this penaeid shrimp species in the present study. Food deprivation during the winter months resulted in growth compensation after refeeding under appropriate culture conditions. During the compensatory growth period (first month after re-feeding), the shrimps gained an average weight of 2.87 g week⁻¹, but after this period, growth rate declined to 0.91 g week⁻¹ (over three-fold less) for the rest of the growth period (between 2nd and 5th months). This preliminary finding is a promising area on which studies should be focused on, as fasting and re-feeding strategy at low temperature can provide a chance to increase marketable size of shrimps or to even produce two crops per year in the sub-tropics. The shrimps (0.2 g)

reached to 13.7 g at 30 PL per m² (DGR 0.10 g), 6.44-7.37 g (DGR 0.05 g) at 40-50 PL per m² in 140 days. The estimated yields were 880-1,150 kg ha⁻¹ in semi-intensive ponds and 1,597-2,673 kg ha⁻¹ in intensive ponds. The shrimps grown in concrete Tanks displayed even poorer growth performance resulting in only 879 kg and 793 kg ha⁻¹ of yield.

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