
INFORMATION OF INTEREST

Aquaculture Twin Event, Patras, Greece, June 11-12, 2007: copy of presentations at <http://www.aquaculture-event.eu>

Regional Review on Aquaculture Development: Western-European Region – 2005: for pdf copy see <http://www.thesalmonsite.com/articles/312/regional-review-on-aquaculture-development-6-westerneuropean-region-2005>

First International Workshop on Advanced Techniques in Sturgeon Fish Larviculture" March 12-14, 2007 in Urmia, Iran: Workshop Recommendations

"Asian - Pacific Aquaculture 2007" The annual meeting of the Asian Pacific Chapter of the World Aquaculture Society, August 5 – 8 2007, Hanoi, Vietnam.

"Approaches to linking producers to markets" A review of experiences to date, Agricultural Management, Marketing And Finance Occasional Paper No 13, FAO, Rome, Italy.

Use of fishery resources as feed inputs to aquaculture development: Trends and policy implications, FAO Fisheries Circular No. 1018

Aquaculture: changing the face of the waters; meeting the promise and challenge of sustainable aquaculture: World Bank Report No. 36622 (2006)

INDUCED OUT-OF-SEASON AND SEASONAL TANK SPAWNING AND STRIPPING OF PIKE PERCH (SANDER LUCIOPERCA L.)

András Rónyai-2007

Aquaculture Research 38(11): 1144-1151

Abstract:

Nine trials were carried out on the induced propagation of pike perch in out-of-season as well as in the natural reproduction period. From January till May, periodically, 16–20 pairs were transported to the hatchery, and acclimated to 15–16°C. The following hormonal treatments were used to induce reproduction: carp pituitary homogenate (CP) and human chorionic gonadotropin (hCG) alone or in combination; gonadotrop-releasing hormone 'Ovurelin' (D-Phe6-LH-RH) alone or with metaclopramid (MTC); and 'Ovopel' [containing des-Gly10-(D-Ala6)-LH-RH-ethylamide and MTC]. The effects of daylight regime, water temperature and the length of the pre-spawning conditioning period on the reproduction performance were determined. From the 130 hormonally treated pairs, 122 either spawned in tanks or were stripped. There were no differences in the ovulation rate between out-of-season (93%) and seasonal (91%) propagation. The developmental stage of the eggs correlated with the latency period, which significantly declined from January till April. The latency period was the shortest in fish treated with hCG and, secondly, with CP. The germinal vesicle migration did not proceed without hormonal treatment, although in some trials breeders were kept at the spawning temperature for 1–2 weeks. The results demonstrate that the pike perch can be successfully induced to spawn about 3 months earlier than its natural spawning season, which allows a significantly prolonged yearly supply of fry.

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EFFECTS OF PHOTOPERIOD ON GROWTH AND SPAWNING EFFICIENCY OF NILE TILAPIA (*OREOCHROMIS NILOTICUS* L.) BROODSTOCK IN A RECYCLING SYSTEM

Abdel-Fattah M. El-Sayed, Mamdouh Kawanna-2007

Aquaculture Research 38 (12): 1242–1247

Abstract:

This study was conducted to evaluate the effects of photoperiod on the growth, feed conversion ratio (FCR) and spawning performance of Nile tilapia (*Oreochromis niloticus*) broodstock. Duplicate groups of two males (66 ± 3.04 g) and six females (50.5 ± 1.58 g) were stocked in 0.4 m³ fibreglass tanks in a recirculating water system at a male:female ratio of 1:3. The fish were subjected to four photoperiod treatments: 24:0 light:dark (L:D), 18L:6D, 12L:12D and 6L:18D. Light intensity was kept constant at about 2500 lx throughout the study. The fish were fed a commercial tilapia diet (35% crude protein and 16.6 MJ GE kg⁻¹) at a daily rate of 2–3% of tank biomass, twice a day, for 130 days. Males grew significantly faster than females under all photoperiod regimes. The best growth rate and FCR were attained at 18L:6D, followed by 24L:0D, 12L:12D and 6L:18D respectively. The number of eggs per female, number of eggs per spawn and number of spawnings per female were all significantly higher in the 12L:12D treatment than in all other photoperiod cycles. Interspawning intervals and days elapsed per spawn were also shorter in the 12L:12D treatment. The time to first spawning was slightly longer in the 24L:0D and 6L:18D than in 12L:12D and 18L:6D light phases. The 18L:6D and 6L:18D photoperiods produced the lowest spawning performance. It is concluded that a 12L:12D photoperiod regime should be adopted for maximum fecundity, seed production and spawning frequencies of Nile tilapia broodstock reared in intensive, recirculating systems. If maximum reproduction is desired, a near-natural day length photoperiod should be used.

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VIABILITY OF DIPLOID AND TRIPLOID LARVAE OF SIBERIAN STURGEON AND BESTER HYBRIDS

Dorota Fopp-Bayat, Malgorzata Jankun, Pawel Woznicki, Ryszard Kolman-2007

Aquaculture Research 38 (12): 1301–1304.

Abstract:

The aim of the present study was to produce *Acipenser baeri* × (*Huso huso* × *Acipenser ruthenus*) hybrids in a diploid and triploid state and to study their viability in comparison with the *A. baeri* from the fish farm stock. A heat shock (37°C) in the 18th minute after fertilization was applied to induce triploidy. The survival rate and the ploidy level of the hybrids obtained were studied. The mortality of triploid hybrids was approximately twice as high as the mortality of diploid hybrids. No significant difference in the survival rate between Siberian sturgeon and their diploid hybrid with bester was noted. Cytogenetic analysis was performed by preparing chromosomes from the gill epithelium. The results showed that all studied fish from the heat-shocked group were triploid.

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EFFECTS OF DIFFERENT DIETS AND SEAWATER SYSTEMS ON EGG PRODUCTION AND QUALITY OF THE BROODSTOCK *BABYLONIA AREOLATA* L. UNDER HATCHERY CONDITIONS

Sirusa Kritsanapuntu, Nilnaj Chaitanawisuti, Yutaka Natsukari-2007

Aquaculture Research 38 (12): 1311–1316.

Abstract:

The broodstock of spotted Babylon snail, *Babylonia areolata*, were conditioned in indoor tanks under two seawater systems (static system and flow-through system) and five types of foods (fish, shrimp, squid, green mussel and formulated diet). High survival and good feeding was found from broodstock conditioned in all treatments of seawater systems and diets. All broodstock showed active behaviour with no signs of stress as exhibited by active movement and feeding, protrusion of siphonal tube and constant egg laying throughout the experiment. Two-way analysis of variance showed that there were no significant differences ($P>0.05$) in the total yield of egg capsules, size of egg capsules, size of newly hatched larvae and growth of larvae among broodstock *B. areolata* conditioned in the seawater system and diet treatments, but there were significant differences ($P>0.05$) in the frequency of egg capsule laying, number of fertilized eggs/embryos per capsule, total yield of fertilized eggs/embryos and final survival of larvae among broodstock *B. areolata* conditioned in the diet treatments. We present initial evidence that the seawater system had a stronger influence on the egg production and quality of broodstock *B. areolata* than conditioning diets.

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LIPID-BASED TRANSFECTION AS A METHOD FOR GENE DELIVERY IN ZEBRAFISH (*DANIO RERIO*) EMBRYOS

Vanesa Robles, M Leonor Cancela-2007

Aquaculture Research 38 (12): 1317–1322

Abstract:

A major challenge to the widespread production of transgenic, knockout and knockdown zebrafish has been the absence of a simple and effective procedure for introducing macromolecules into the fertilized egg. None of the existing techniques for gene transfer in fish embryos has proven to be a major advance over cytoplasm microinjection, which is a technically demanding and time-consuming procedure. This report addresses this need, considering that the development of protocols for lipid-based transfection with fish embryos would considerably simplify gene transfer in this complex biological model. In this study, lipid-based transfection with two different reporter vectors was carried out in zebrafish embryos at different developmental stages. The parameters tested included different plasmid/transfection reagent ratios as well as the influence of an added transfection enhancer reagent. When embryos were transfected in the blastula stage with a pEGFP-N1 vector, more than 35% successfully incorporated the plasmid and expressed the fluorescent protein 24 h after transfection. The transfection enhancer did not show any significant effect in our experiments. This work presents an approach to implement this technique as a faster, cheaper and more practical alternative than microinjection.

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EFFECTS OF LIGHT QUALITY ON THE REPRODUCTION AND MORPHOLOGY OF SPOROPHYTES OF *LAMINARIA JAPONICA* (PHAEOPHYCEAE)

Hiroyuki Mizuta, Tomoki Kai, Katsuhiko Tabuchi, Hajime Yasui-2007

Aquaculture Research 38 (12): 1323–1329

Abstract:

Responses of the sporophytes of *Laminaria japonica* to blue, red and white lights were investigated using segments, discs and whole plants. The relative growth rate with regard to the blade's length and width showed no difference among the light conditions during 2 weeks of culturing. On the contrary, the elongation rates of the stipe and the holdfast were 1.7–2.5 times higher under red light than that under white light as a control. Callus formation in the segments was also promoted more under red light conditions than under white or blue light. Blue light irradiation resulted in a 48% and 12% decrease in the relative growth rate of the

stipe and the holdfast, respectively, in comparison with the control. When sporophyte discs were cultured for 10 weeks, all discs formed zoosporangial sori 2–3 weeks earlier under blue light than under white light. However, red light inhibited the formation of zoosporangia. Indoleacetic acid (IAA) oxidase activity was about two-fold higher under blue light than under red light, suggesting that the difference in response between the blue and red light conditions is related to the internal IAA level of the tissues. The developmental, reproductive and morphological effects of blue and red lights will be useful for the artificial control of the life cycle of *Laminaria* plants and for the development of more efficient cultivation techniques.

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DO DIETARY AMINO ACID PROFILES AFFECT PERFORMANCE OF LARVAL GILTHEAD SEABREAM?

Cláudia Aragão, Luís E.C. Conceição, Marc Lacuisse, Manuel Yúfera, Maria Teresa Dinis-2007

Aquat. Living Resour. 20:155-161

Abstract :

Live preys commonly used in fish larval rearing seem to be imbalanced in terms of amino acids. Manipulation of their amino acid composition is difficult, but the use of microencapsulated diets allows this manipulation. This study analysed the effect of amino acid supplementation, in order to compensate for dietary amino acid imbalances, on growth and survival of gilthead seabream (*Sparus aurata*) larvae. Larvae were reared until 32 days after hatching, in a closed recirculating water system (19 °C), using only live food (rotifers and *Artemia*). Thereafter, larvae were fed *Artemia* or one of the experimental microencapsulated diets: supplemented with indispensable amino acids (IAAsup diet), or supplemented with dispensable amino acids (DAAsup diet). Experiment lasted for 10 days. Dietary supplementation with indispensable amino acids resulted in A/E ratios [(each indispensable amino acid) (total indispensable amino acids)-1 1000] more similar to the ones of larval seabream and in higher IAA:DAA ratios than in the DAAsup diet. Survival was similar in larvae fed the IAAsup diet (75%) or *Artemia* (87%), but was significantly lower in larvae fed the DAAsup diet (52%). Larvae from all treatments more than doubled their average dry weight during the experimental period. Final dry weight was similar in larvae fed both microcapsules, but these were lighter than larvae fed with *Artemia*. Relative growth rate (RGR) and total biomass production tended to be higher in larvae fed the IAAsup (RGR = 9% day⁻¹) than the DAAsup diet (RGR = 7.5% day⁻¹) and only in this last treatment these parameters were significantly lower than in larvae fed with *Artemia*. Therefore, dietary supplementation with indispensable amino acids resulted in a more balanced dietary amino acid profile, which significantly increased survival. Further studies introducing microdiets earlier in the development seem necessary in order to optimise growth.

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EFFECTS OF SHORT-TIME ARTEMIA SPP. FEEDING IN LARVAE AND DIFFERENT REARING ENVIRONMENTS IN JUVENILES OF COMMON BARBEL (*BARBUS BARBUS*) ON THEIR GROWTH AND SURVIVAL UNDER INTENSIVE CONTROLLED CONDITIONS

Tomás Polícar, Pavel Kozák, Jitka Hamácková, Andrea Lepicová, Jirí Musil, Jan Kouril - 2007

Aquat. Living Resour. 20: 175-183

Abstract:

The effect of short-time *Artemia* spp. feeding on growth performance and cumulative survival rate of barbel (*Barbus barbus*) larvae were studied under controlled aquaria conditions during the 21-day larval period. Three different diets (presenting reduced *Artemia* feeding) were tested for first exogenous nutrition of larvae (since 13 days post hatch): (1) artificial feed (Asta); (2) *Artemia* nauplii for 7 days followed by artificial feed; (3) *Artemia* nauplii for 14 days followed by artificial feed. The longer period of live food statistically improved growth of larvae ($W = 174 \pm 20$ mg and $SGR = 14.5 \pm 0.5\%$ d⁻¹). The artificial dry food Asta without the addition of *Artemia* nauplii caused statistically decreased growth ($W = 135 \pm 22$ mg and $SGR = 13.1 \pm 0.7\%$ d⁻¹). However, the growth of larvae with the short period of *Artemia* nauplii ($W = 153 \pm 25$ mg and $SGR = 13.8 \pm 0.7\%$ d⁻¹) did not differ compared to either group. All used feeding diets did not have a significant effect on the cumulative survival rate of larvae ranging from 73.1% to 74.1% at the end of the larval rearing period. The effects of the rearing environment on growth performance and survival rate of juveniles were tested under intensive controlled conditions in aquaria and troughs for 84 days following the larval period (from 34 to 118 dph). The environment of the troughs caused significantly decreased growth ($W = 2079 \pm 433$ mg and $SGR = 3.1 \pm 0.05\%$ d⁻¹) of juveniles compared to ones reared in aquaria ($W = 3236 \pm 264$ mg and $SGR = 3.6 \pm 0.1\%$ d⁻¹) at the end of the juvenile rearing period. Nevertheless, rearing environment did not have a significant influence on the cumulative survival rate of barbels (= 90.4% and 81.3% in aquaria and troughs, respectively). (University of South Bohemia, Research Institute of Fish Culture and Hydrobiology, Department of Aquaculture and Hydrobiology, Zátisí 728/II, 389 25 Vodnany, Czech Republic; email of Tomáš Polícar: policar@vurh.jcu.cz)

TWO IMPORTANT TECHNIQUES FOR ISOLATION OF MICROALGAE

M. Parvin, M.N. Zannat, M.A.B. Habib-2007

Asian Fisheries Science 20(1)

Abstract:

Microalgae (phytoplankton) plays a very important role as live food for aquatic organisms. However, out of the 80,000 species of microalgae, only 50-60 species are commercially important as live food and for other nutritional purposes. Moreover, these microalgae are available in freshwater as well as marine water which should be isolated first and then used for monoculture or production for use as live food or for other commercial purposes. There are two important cosmopolitan methods worldwide for the isolation of microalgae and then pure culture. These two methods are: Micropipette Washing Technique and Centrifuge Washing and Streak Plating Technique which are described in detail for the isolation of microalgae in the Materials and Methods section, to make these methods available to researchers. So far, there is no available published literature about a reliable, easily understandable technique for the isolation of phytoplankton and or microalgae in our library nor in our country. Researchers as well as students are having a hard time obtaining the same from their localities. Therefore, with the aim in view of producing a manual discussing these two important techniques to isolate microalgae using clear and simple language and easily available, this paper is prepared.

GROWTH PERFORMANCE AND NUTRITIONAL VALUE OF CHLORELLA ELLIPSOIDEA IN FERTILIZER FACTORY EFFLUENT MEDIA

M.A. Toyub, S.R. Ahmet, M.I. Miah, M.A.B. Habib-2007

Asian Fisheries Science 20(1): 65-79

Abstract:

The growth performance of green alga, *Chlorella ellipsoidea* was studied in a laboratory in different concentrations of fertilizer factory effluent media (FFEM). Five different concentrations viz. 40, 45, 50, 55 and 60% of FFEM and bold basal medium (BBM) (control) were used with three replications for a period of three months. Each trial was done for a

period of 16 days. The initial cell density of *C. ellipsoidea* was 2.5×10^5 cells.ml⁻¹ which attained a maximum density of 198.49×10^5 cells.ml⁻¹ in BBM followed by 182.07, 157.41, 142.34, 137.57 and 121.35 ($\times 10^5$ cells. ml⁻¹) in 50, 55, 45, 60 and 40 % FFEM, respectively on the 10th day of culture. A similar trend was observed in the case of chlorophyll a content and the range was 5.85 to 9.39 mg.l⁻¹. The specific growth rate (SGR, μ .day⁻¹) on the basis of cell number and chlorophyll a were found at 0.40 to 0.44 and 0.41 to 0.45, respectively, and was significantly ($p < 0.05$) higher in BBM. The total biomass was found at 629.13 and 541.36 mg.l⁻¹ in BBM and 50% FFEM, respectively. The overall growth performance of *C. ellipsoidea* was significantly ($p < 0.05$) higher in 50% FFEM than in other concentrations of FFEM. The cultured microalga was found nutritionally rich.

NUTRITIONAL VALUE OF BINARY MICROALGAL DIETS FOR LARVAE OF THE BLACKLIP PEARL OYSTER PINCTADA MARGARITIFERA (L.)

I. Teajoro, I. Arabua, J. Whitford, P.C. Southgate-2007

Asian Fisheries Science 20(1): 55-64

Abstract:

This paper reports on two experiments to assess the nutritional value of cultured microalgae for larvae of the blacklip pearl oyster, *Pinctada margaritifera*. In the first experiment, two binary diets were assessed and *Isochrysis* sp. (T-ISO) was administered in combination with either *Pavlova salina* or the diatom *Chaetoceros muelleri*. The second experiment assessed the nutritional value of the individual species of microalgae from the best binary diet used in the first experiment (T-ISO and *C. muelleri*). Larvae fed T-ISO/*C. muelleri* in the first experiment showed significantly better growth and survival ($P < 0.05$) than those fed T-ISO/*P. salina* after 20 days of feeding. Administering the microalgae from the best binary diet separately, showed that *C. muelleri* alone is unsuitable for rearing D-stage *P. margaritifera* larvae. Larvae fed with T-ISO showed significantly better growth and survival ($P < 0.05$) than those fed *C. muelleri* at the end of 9 days of feeding and the slower growth rate of larvae fed *C. muelleri* was apparent from day 5. The mean antero-posterior shell measurement (APM) of larvae fed *C. muelleri* at day 5 ($88.33 \pm 1.95 \mu\text{m}$) was significantly smaller ($P < 0.05$) than that of the larvae fed T-ISO ($94.33 \pm 4.47 \mu\text{m}$). The data generated in this study will be valuable in the further refinement of hatchery culture techniques for *P. margaritifera* throughout the Indo-Pacific region.

EVALUATION OF DIFFERENT LIVE FOODS ON GROWTH AND SURVIVAL OF SEAHORSE FISH (HIPPOCAMPUS KUDA, SIGNATHIDAE) PONIES

H.B. Dhamagaye, N.D. Chogale, R.D. Bondre, V.R. Bhatkar, S.T. Indulkar, B.P. Bhosale, S.G. Belsare-2007

Asian Fisheries Science 20(1): 1-6

Abstract:

Seahorse (*Hippocampus kuda*, Signathidae) ponies were reared for 15 days at the laboratory of Marine Biological Research Station, Ratnagiri (India) using three types of live food viz *Artemia nauplii*, rotifers (*Branchionus plicatilis*) and a combination of both. The experiment was conducted in all glass aquaria (0.30 m x 0.30 m x 0.30 m). Seahorse ponies (10 + 0.27 mm) were stocked at 2 L⁻¹ and fed with nutritional live food ad libitum. After 15 days of rearing, the ponies were counted and their individual lengths and weights were recorded. Significantly higher ($P < 0.05$) weight gain percentage ($128.92 + 1.27\%$) of seahorse ponies, length gain percentage ($57.4 + 0.82\%$), SGR percentage per day ($5.53 + 0.04\%$ day⁻¹) and survival percentage (100%) of seahorse ponies were observed in the combination of *Artemia nauplii* and rotifers as food compared with *Artemia nauplii* or rotifer alone. Rotifers as live food resulted in poor growth and survival of seahorse ponies under this experimental condition.

EXPRESSION OF IMMUNE-RELATED GENES IN LARVAL STAGES OF THE GIANT TIGER SHRIMP, *PENAEUS MONODON*

Pikul Jiravanichpaisal, Narongsak Puanglarp, Sasithon Petkon, Seri Donnue, Irene Söderhäll, Kenneth Söderhäll-2007

Fish & Shellfish Immunology 23(4): 815-824

Abstract:

Shrimp undergo several morphologically different stages during development and therefore the expression of some immune-related genes such as prophenoloxidase (proPO), peroxinectin (Prx), crustin (Crus), penaeidin (Pen), transglutaminase (TGase), haemocyanin (Hc) and astakine (Ak) were determined during larval development of the shrimp (*Penaeus monodon*), i.e. nauplius 4 (N4), protozoa 1 and 3 (Z1 and 3), mysis 3 (My 3), post-larvae 3 (PL3) and also in haemocytes of juveniles. Semi-quantitative RT-PCR analysis showed that all transcripts were already present in the early larval stage of N4 but at different levels. The transcript of proPO was found to be extremely low or even absent at N4, whereas Prx, Crus, Pen, TGase, Hc and Ak were significantly expressed at all larval stages. Up to now expression of proPO and Prx has only been reported from haemocytes in crustaceans and in this study Prx also appeared to be expressed in stages which appear to lack haemocytes. Thus, this may suggest that Prx is expressed in other cells than haemocytes. It is well known among invertebrates that the proPO system plays a crucial role as an immune effector molecule against microbes. However, in this study, the transcript of proPO was low during the larval stages and hardly present at all at N4. This might indicate that the development of immune-competent haemocytes during the larval stages is not completed and as a consequence they are likely to be more susceptible to infectious diseases during these stages.

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SWIMMING BEHAVIOUR OF *BRACHIONUS PLICATILIS* IN RELATION TO FOOD CONCENTRATION AND FEEDING RATES

M. Yúfera-2007

Hydrobiologia 593: 13-18

Advances in Rotifer Research

Abstract:

We examined feeding rates and swimming speed in amictic females of *Brachionus plicatilis* over algal cell concentrations ranging from 15×10^3 to 30×10^6 cell ml⁻¹, to determine to what extent filtration rate is a consequence of a real modulation of swimming speed in response to food availability. Swimming rates were measured using an automated motion analysis system via video recording. The results showed that swimming speed changed as a function of food density. Swimming speed increased from the lowest tested concentration of algae to reach a maximum at 6×10^6 cell ml⁻¹. Above this density, swimming speed declined slightly and then remained constant at a mean speed of 0.45 mm s⁻¹. Filtration and ingestion rates changed as cell concentration increased, following patterns consistent with those generally described for suspension feeders. However, the observed swimming pattern did not explain the recorded changes in clearance rate. These results suggest that filtration, and therefore ingestion, is mainly regulated by modifying particle retention efficiency.

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AN INDIVIDUAL-BASED POPULATION MODEL FOR THE PREDICTION OF ROTIFER POPULATION DYNAMICS AND RESTING EGG PRODUCTION

M. O. Alver, A. Hagiwara-2007

Hydrobiologia 593:19-26

Advances in Rotifer Research

Abstract:

Most species of rotifers have a combination of sexual and asexual reproduction, with sexual reproduction resulting in resting eggs, which can lay dormant for long periods. The occurrence of sexual reproduction affects population dynamics through the temporary presence of male rotifers, and a reduction in the growth of the number of female rotifers. A previously published, individual-based model used dynamic energy budget theory to describe rotifer food intake, growth, egg production, and mortality, but assumed asexual reproduction only. In the current study, we have expanded the model to describe the entire reproductive cycle of the rotifers, making it usable for investigating relationships, such as those between the signal triggering mictic egg production, and the timing and number of resting eggs produced. The model is intended for use in predicting the specific future development of cultures, for instance, as a process model in rotifer or resting egg production for aquaculture.

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INDUCIBLE DEFENSES AND ROTIFER FOOD CHAIN DYNAMICS

Irene Van der Stap, Matthijs Vos, Wolf M. Mooij-2007

Hydrobiologia 593:103-110

Advances in rotifer research

Abstract:

Theoretical studies have predicted that inducible defenses affect food chain dynamics and persistence. Here we review and evaluate laboratory experiments that tested hypotheses developed from these theoretical studies. This review specifically focuses on the effects of inducible defenses in phytoplankton-rotifer food chain dynamics. First, we describe the occurrence of colony formation within different strains of green algae (Scenedesmaceae) in response to infochemicals released during grazing by the herbivorous rotifer *Brachionus calyciflorus*. Then we examined the effects of inducible defenses on the population dynamics of this planktonic system in which algal strains that differed in their defense strategies were used. Simple food chains were composed of green algae (Scenedesmaceae), herbivorous rotifers (*Brachionus calyciflorus*) and carnivorous rotifers (*Asplanchna brightwellii*). In this system *B. calyciflorus* exhibits an inducible defense against predation by developing long postero-lateral spines. Experimental studies showed that inducible defenses, as opposed to their absence, could prevent high-amplitude population fluctuations. We discuss the dual effects of induced defenses on extinction probabilities and consider the fit of a theoretical model to experimental data to understand the mechanisms that underlie the observed dynamics.

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TRANSFECTION OF SIRNA INTO BRACHIONUS PLICATILIS (ROTIFERA)

Tonya L. Shearer, Terry W. Snell-2007

Hydrobiologia 593: 141-150

Advances in Rotifer Research

Abstract:

Transfection experiments have played a prominent role in functional genomics, being used to probe phenotypes with reporters and knockouts with techniques like RNAi, gene over-expression studies, and the creation of transgenic animals. Rotifers are not amenable to many classic genetic manipulations, hence have not been a model system in the genomics revolution. However, no one has explored whether rotifers are susceptible to gene expression knock-out via transfection, and with the establishment of the rotifer transcriptome sequencing project, an increasing amount of sequence data is available to define target genes for manipulation. As a first step towards transfection of rotifers, we describe our attempt to transfect double-stranded, fluorescently-labeled siRNA into resting eggs, hatched rotifers and amictic eggs with varying levels of success. Transfection was successful when hatched rotifers (rather than resting eggs or amictic eggs) were directly treated, and possibly successful when treated as resting eggs. Highest levels of transfection (as determined by significant fluorescence of the siRNA in the pseudocoelom) were observed in fed rotifers that were 2 h old at the time of treatment, which involved combining lipofection and electroporation methods of transfection. Successful transfection of siRNA into these animals now provides a possible mechanism for the exploration of rotifer gene function.

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ROTIFER DIGESTIVE ENZYMES: DIRECT DETECTION USING THE ELF TECHNIQUE

Martina Štrojsová, Jaroslav Vrba-2007

Hydrobiologia 593:159-165

Advances in rotifer research

Abstract:

Hydrolytic enzymes involved in rotifer digestive processes were investigated directly at the sites of enzyme action using the ELF (Enzyme Labelled Fluorescence) technique. After enzymatic hydrolysis of an artificial ELF substrate, the fluorescent product ELF alcohol (ELFA) marked the sites of enzyme action. The time development of ELFA labelling was studied at different incubation times. Phosphatases, β -N-acetylhexosaminidases and lipases were examined in *Brachionus angularis*, *B. calyciflorus*, *Keratella cochlearis* and *Lecane closterocerca* from fed-batch cultures. We detected activities of all studied enzymes mostly in the stomach and intestine of rotifers. *L. closterocerca* was the only species showing enzyme activity at the mastax. Lipase activity was observed in the stomach and intestine of all species and in the mastax of *L. closterocerca*. Phosphatases were frequently located at the corona of *B. calyciflorus*. In other cases, both phosphatases and β -N-acetylhexosaminidases were rarely detected at the corona, and on the lorica and epidermis of some species.

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WORKSHOP ON BARCODED DNA: APPLICATION TO ROTIFER PHYLOGENY, EVOLUTION, AND SYSTEMATICS

C. William Birky Jr.-2007

Hydrobiologia 593: 175-183

Advances in Rotifer Research

Abstract:

DNA barcoding is the use of segments of gene sequences to assign individual organisms to species. Thus it can be used to define species and to identify specimens. Barcoding has been applied as an aid to systematics with little controversy in both monogonont and bdelloid rotifers, and also in environmental sequencing projects designed to determine the diversity of microscopic organisms. In contrast, a great deal of controversy has arisen over the creation of

the Consortium for the Barcode of Life, a major initiative to barcode all the species in several major groups of animals, with the long-range goal of barcoding all species of organisms. This is a very brief review of DNA barcoding, especially as applied to rotifers, and a summary of the results of a workshop held at the 11th International Workshop on Rotifera.

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INDIVIDUAL- AND POPULATION-LEVEL EFFECTS OF ANTIBIOTICS ON THE ROTIFERS, BRACHIONUS CALYCIFLORUS AND B. PLICATILIS

Adriana Araujo, James N. McNair-2007

Hydrobiologia 593:185-199

Advances in Rotifer Research

Abstract:

The toxicity of three common antibiotics (streptomycin sulfate, tetracycline hydrochloride, and tylosin tartrate) to the freshwater rotifer *Brachionus calyciflorus* and brackish-water rotifer *B. plicatilis* was investigated using full-lifespan exposure durations. Effects of each antibiotic on lifespan, lifetime reproduction, and Malthusian parameter were assessed at seven nominal concentrations (ranging from 5.6 mg l⁻¹ to 2,000 mg l⁻¹) and a negative control. Lowest Observed Effect Concentrations (LOECs) were determined for reproduction and lifespan, while 1%, 10%, 25%, and 50% Inhibitory Concentrations (IC1, IC10, IC25, IC50) and 95% confidence intervals were estimated for all three endpoints. LOECs ranged from 5.6 mg l⁻¹ to 90 mg l⁻¹, with all LOECs less than 90 mg l⁻¹ occurring in *B. calyciflorus*. The lowest IC1 concentrations were 3.91 mg l⁻¹ for the effect of tetracycline on lifetime reproduction in *B. calyciflorus* and 4.06 mg l⁻¹ for the effect of tylosin on lifetime reproduction in *B. plicatilis*. Overall, lifetime reproduction was the most sensitive endpoint and the Malthusian parameter was the least sensitive. IC1 values for lifetime reproduction were roughly one to two orders of magnitude lower than the corresponding IC50 values. (Center for Environmental Research, Academy of Natural Sciences of Philadelphia, 1900 Benjamin Franklin Parkway, Philadelphia, PA 19103, USA; email of James N. McNair: mcnair@acnatsci.org)

EFFECT OF DIAZINON ON LIFE STAGES AND RESTING EGG HATCHABILITY OF ROTIFER BRACHIONUS PLICATILIS

Helen S. Marcia, Atsushi Hagiwara-2007

Hydrobiologia 593: 219-225

Advances in rotifer research

Abstract:

The effects of organophosphate pesticide, diazinon, on life history parameters and hatchability of resting eggs of rotifer *Brachionus plicatilis* were assessed. Newly hatched (<1 h-old) neonates were individually cultured in six varying concentrations (0/control, 0.1, 1.0, 2.5, 5.0 and 10.0 mg/l) of diazinon. The life history parameters such as time (h) the rotifers bear first egg and release first neonate, reproductive period, net reproductive rate, mixis, intrinsic rate of population increase, and life span were evaluated. Results showed that among the life history parameters, the time the rotifers took to release neonates is the most sensitive, giving the lowest EC50 value of 1.24 mg/l. The fecundity of maternal females, amictic and mictic daughters was also investigated. Rotifers exposed to 10.0 mg/l produced significantly fewer amictic daughters, and at this concentration, rotifers did not produce any mictic daughter. At 5.0 mg/l, the number of male offspring was significantly lower than the control. Furthermore, the hatchability of resting eggs produced by the rotifers was evaluated when exposed to diazinon: from birth until they produced resting eggs (early development); during late developmental stage of resting eggs (before diapause); and during diapausing stage. The

hatchability of the resting eggs was not affected when exposure was timed at late developmental and diapausing stages. Overall results showed that even though amictic females reproduced normally in the presence of low-concentration of diazinon, sexual reproduction is severely affected, especially the hatchability of resting eggs when the exposure was timed on its early developmental stages.

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WORKSHOP ON ROTIFERS IN ECOTOXICOLOGY

Terry W. Snell, Célia Joaquim-Justo-2007

Hydrobiologia 593: 227-232

Advances in rotifer research

Abstract:

The aim of the workshop on rotifers in ecotoxicology was to stimulate discussions on new developments in the field. Discussions about the use of biomolecular tools indicate that gene expression analysis with rotifers should be available in the next few years. Such analyses will be a great asset as they enable ecotoxicologists to study molecular mechanisms of toxicity. Rotifers also appear as useful tools in the risk assessment of pharmaceuticals and their metabolites that find their way into aquatic ecosystems because their sensitivity to some of these substances is higher than that of cladocerans and algae. The nature and extent of the impact of potential endocrine disruptors on aquatic invertebrates is another poorly resolved issue for which rotifers are a promising tool. Indeed, rotifers seem to be particularly sensitive to androgenic and anti-androgenic substances, whereas copepods and cladocerans are typically more affected by estrogens and juvenile hormone-like compounds. Besides their usefulness in these emerging fields of aquatic ecotoxicology, it was emphasized that research with rotifers on basic issues like, e.g., toxicant interference with predation, competition, or interspecific and interclonal variation in ecotoxicological tests is still needed.

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ANALYSIS OF THE AGGLUTINATING ACTIVITY FROM UNICELLULAR ALGAE

Chun-Yao Chu, Rang Huang, Lian-Ping Lin-2007

Journal of Applied Phycology 19(5): 401-408

Abstract:

Agglutinating activity often varies both between and within the algal species assayed. However, it is difficult to interpret such variation without further analysis. We report a statistical analysis of agglutinating activities against human, cow, sheep, and pig erythrocytes, using cell extracts from 43 taxa (strains) of freshwater microalgae. Most of the extracts agglutinated erythrocytes from at least one of the sources, but pig erythrocytes appeared to be most suitable for the detection of agglutination reactions. *Chlorella* cell extracts preferentially agglutinated human erythrocytes, whereas extracts of other taxa were less active against mammalian erythrocytes. Cluster analysis generated four distinct subclusters of taxa, characterized by different specificities for antigens or carbohydrate receptors on the erythrocytes. Principal component analysis further separated the agglutination characteristics of *Chlamydomonas* from *Chlorella* on the first two components. Specificity for pig erythrocytes accounted for most of the clustering or grouping of algal taxa in multivariate analysis. However, clustering or grouping patterns of *Chlorella* species on haemagglutinating activity resembled that based on DNA sequences, revealing a possible genetic connection of agglutinins and their biochemical characteristics in algal cells. Variability of agglutination reactions among the algae investigated is simplified and interpreted most easily using multivariate analysis.

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REMOVAL OF NITROGEN AND PHOSPHORUS FROM WASTEWATER USING MICROALGAE IMMOBILIZED ON TWIN LAYERS: AN EXPERIMENTAL STUDY

Jing Shi, Björn Podola, Michael Melkonian-2007

Journal of Applied Phycology 19(5): 417-423

Abstract:

Removal of nitrogen and phosphorus from wastewater by two green microalgae (*Chlorella vulgaris* and *Scenedesmus rubescens*) was investigated using a novel method of algal cell immobilization, the twin-layer system. In the twin-layer system, microalgae are immobilized by self-adhesion on a wet, microporous, ultrathin substrate (the substrate layer). Subtending the substrate layer, a second layer, consisting of a macroporous fibrous tissue (the source layer), provides the growth medium. Twin-layers effectively separate microalgae from the bulk of their growth medium, yet allow diffusion of nutrients. In the twin-layer system, algae remain 100% immobilized, which compares favourably with gel entrapment methods for cell immobilization. Both microalgae removed nitrate efficiently from municipal wastewater. Using secondary, synthetic wastewater, the two algae also removed phosphate, ammonium and nitrate to less than 10% of their initial concentration within 9 days. It is concluded that immobilization of *C. vulgaris* and *S. rubescens* on twin-layers is an effective means to reduce nitrogen and phosphorus levels in wastewater.

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PALATABILITY AND FATALITY OF THE DINOFLAGELLATE PROROCENTRUM LIMA TO ARTEMIA SALINA

C. C. Ajuzie-2007

Journal of Applied Phycology 19(5): 513-519

Abstract:

Prorocentrum lima is a toxic alga that produces both intra-cellular and extra-cellular toxins, including okadaic acid (OA) and dinophysistoxins (DTXs). Nauplii of the brine shrimp *Artemia salina* were exposed to both the cell and cell-free culture medium of *P. lima* in order to test the hypotheses that the extra-cellular medium is toxic to brine shrimp and that the *P. lima* cell is palatable but fatal to it. *Artemia* cysts incubated in the cell-free medium hatched, but mortalities were recorded for nauplii that hatched in, and metanauplii exposed to, test solutions (autoclaved filtered seawater + cell-free medium) that contained at least 50% of the cell-free medium. Animals exposed to cells of *P. lima* readily fed on the cells. Some, especially among the Day 1 nauplii, ingested only one cell before dying, while others ingested more than one cell, up to six cells in the case of Day 3 nauplii, before dying. Day 3 nauplii were readily and heavily impacted by the *P. lima* cells. Survival analysis was used to evaluate survivorship of Day 1 to Day 3 nauplii exposed to cells of *P. lima*. Estimates were made of tD50s for the different age groups. Comparisons of the tD50s showed that the tD50s for Day 1 and Day 2 nauplii did not vary significantly, but they each varied significantly from the tD50 for the Day 3 nauplii. The possible ecological implications of the findings are discussed. (Laboratoire d'Océanographie Biologique et Aquacultures, Université Libre de Bruxelles (ULB), CP 160/19, Av. Roosevelt 50, 1050 Bruxelles, Belgium ; email of C. C. Ajuzie : cajuzie@ulb.ac.be)

THE TAXONOMY OF THE GENUS DUNALIELLA (CHLOROPHYTA, DUNALIELLALES) WITH EMPHASIS ON THE MARINE AND HALOPHILIC SPECIES

Michael A. Borowitzka, Christopher J. Siva-2007

Journal of Applied Phycology 93(5): 567-590

Abstract:

The taxonomy of the green algal genus *Dunaliella* is often seen as confusing and the names associated with species in culture collections are sometimes suspect. This paper evaluates and reviews the current taxonomy based on morphological and biochemical characters of this genus. The life history of *Dunaliella* is also presented. The variability, stability and usefulness of the main characters used to characterise the taxa are evaluated, based both on the literature and on a detailed examination of the *Dunaliella* strains in the Murdoch University Microalgae Culture Collection. A detailed updated description and key to the 22 species and a number of varieties and forms of the marine and halophilic species of *Dunaliella* currently recognised is given to allow researchers to identify their strains.

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IMAGE ANALYSIS - A SIMPLE METHOD OF ALGAL CULTURE GROWTH ASSESSMENT

Tomáš Hauer, Lukáš Jirka-2007

Journal of Applied Phycology 93(5): 599-601

Short Communication

Abstract:

A non-destructive method for the relative growth quantification of algae or cyanobacteria in the culture has been developed. It is based on image analysis and does not require any special equipment. Results provided by this method were compared with those provided by a chlorophyll a assessment, with a correlation factor of over 88%.

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