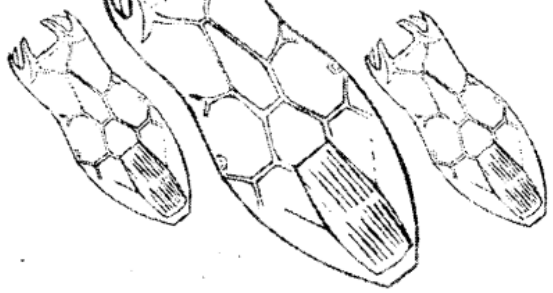
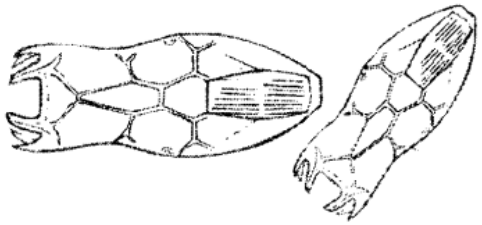
ROTIFERA

MEXICO CITY, MEXICO
11 – 18 MARCH 2006



ABSTRACTS



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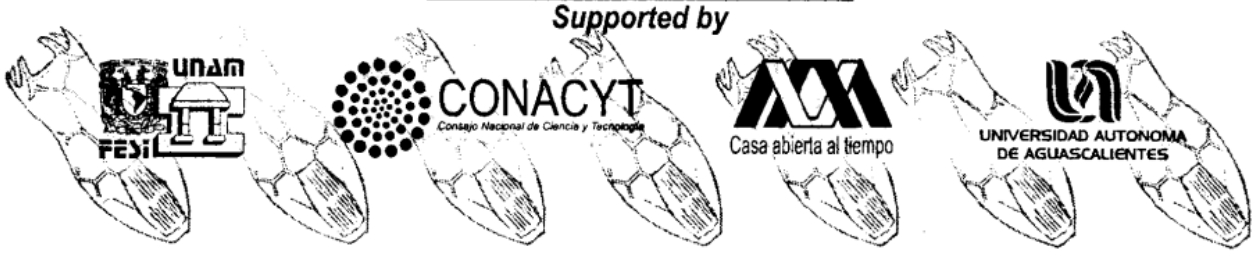

ROTIFERA

XI International Symposium on Rotifers

Mexico City, Mexico

11th – 18th March

2006

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Abstracts

Preparation of single specimens or parts of Rotifera and other Meiofauna for SEM

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Because of their small size, meiofauna organisms are easily lost when dried and mounted for SEM. This is why drying and mounting have to be optimised. Traditionally, rotifer specimens have been dried in balzers® microporous teflon containers or in self-made tubes covered with gauze. These methods are appropriate when larger specimens are prepared or if specimens are available in high numbers. If, however, specimens are very small (<30 µm in diameter), they pass through the wall of the microporous containers. Additionally, it is difficult to re-trieve single specimens out of the drying tube in order to mount them. To overcome these problems, we have developed a little teflon container (4 mm height, 2 mm in diameter) covered on either side with grids normally used for TEM. Using this teflon container, it is possible to prepare single specimens of rotifers. Having dried the specimens, they are subsequently retrieved from the container using an eyelash attached to a toothpick. Next, they are mounted on an aluminium stub. This is usually done by gluing the specimens or parts of them on stubs covered with double-sided sticky tape using impinging-light because the aluminium stub doesn't allow light to pass through. Rotifers, however, are very small which makes it difficult to properly handle them with this kind of illumination technique. Furthermore, the specimens mounted are prone to sinking into the glue on the sticky tape. Attempting to overcome these problems, we here present a method for mounting rotifers using transmitted light. Additionally, we are now able to carefully control the strength of the layer of glue. Our simple solution is to use round coverslips matching the diameter of the aluminium stubs and covered by a defined layer of the glue. Specimens can be mounted using a toothpick with an eyelash attached. Finally, the coverslips are mounted on the aluminium stub and sputtered with gold.

Challenges and opportunities of different techniques of preparation of rotifer specimens

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Rotifers have traditionally been considered a group of animals difficult to study. This is probably because of their minuteness and sensitivity towards disturbance. Furthermore, taxonomically and phylogenetically important features such as the rotatory organ and the trophi display a highly degree of complexity. What is essential are techniques which are specifically adapted to the small size of rotifers and, additionally, accurately visualize their complex structures. Here I present a host of techniques useful for specimen preparation for both light- and electron microscopy. Specifically, techniques of anaesthetisation, fixation, and preparation of permanent mounts, documentation, serial sections and subsequent three-dimensional reconstructions are given. Moreover, optimized methods for SEM preparations are presented. All these techniques are demonstrated based on instructive examples.

The labium of the Rotifera – a morphologically and phylogenetically interesting structure of the rotatory organ

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Females of several species of Rotifera have been observed by SEM and their external morphology compared. Here we report the existence of a characteristic morphological structure as part of the rotatory organ which has widely been overlooked as such. We have termed this structure labium in analogy to a structure similar in form and function to that in insects. The labium is derived from a ventral duplication of the rotifer head. It consists of an unpaired proximal plate and an attached pair of distal plates. The rounded distal plates converge medially forming a tapering rim. This tapering rim reaches the proximal plate covering the mouth. Both distal plates are subdivided by a distinct fold. This labium is present in several taxa of Monogononta, e.g. Lepadellidae, Mytilinidae, Brachionidae and Euchlanidae but is lacking e.g. in Dicranophoridae, Synchaetidae, Asplanchnidae and Notommatidae. Interestingly, the labium is also present in Seison and possibly Bdelloidea. These findings are potentially important from a phylogenetic perspective. First, given that the labium evolved in the stem species of Syndermata, its absence in several taxa of Monongononta is apomorphic. Second, if the absence of a labium in monogonont taxa is plesiomorphic, the labium must have evolved within Monogononta

Effect of mixed diets (*Microcystis* and *Chlorella*) on competition between the rotifers *Brachionus calyciflorus* and *Brachionus havanaensis*

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Many freshwater bodies in México are generally shallow and eutrophic with toxic cyanobacteria such as *Microcystis aeruginosa* blooming for several months in a year. In these waterbodies, the species divesity of *Brachionus* is often limited to fewer than 4 species. *B. havanaensis* is one of the most common planktonic rotifers sometimes found together with *B. calyciflorus* in *Microcystis*-infected waterbodied. The aim of the present work was to evaluate the impact of mixed diets (0, 25, 50, 75 and 100% *Microcystis* or *Chlorella*) on the competition between *B. calyciflorus* and *B. havanaensis*. In general, increased proportion of *M. aeruginosa* in the diet resulted in decreased population growth rate both the rotifer species. When grown separately or together, *B. havanaensis* was always numerically more abundant than *B. calyciflorus*, regardless of the proportion of *Microcystis* in the diet. However, both the rotifer species were grown together, the impact of increased proportion of *M. aeruginosa* in the diet had more adverse effect on *B. calyciflorus* than on *B. havanensis*. When grown alone, both the rotifer species showed better population growth on a diet of 75% *Chlorella* and 25 % *Microcystis* than either 100% *Chlorella* or *Microcystis*.

Individual-based modelling for the prediction of rotifer population dynamics

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An individual-based model has been constructed based on dynamic energy budget theory to describe rotifer feed intake, growth, egg production and mortality. In an ensemble simulation, the model can be generalized to describe rotifer populations. The ensemble model allows for simulating an arbitrary number of actual individuals using a limited number of parallel model instances. Each instance has independent state values, and the ensemble model can therefore represent populations with different age structures and growth rates. This simulation structure makes the model equally applicable for describing stationary conditions as for a variety of transient scenarios. The model describes the entire reproductive cycle of rotifers, and can be used to investigate relationships such as that between the signal triggering mictic egg production and the timing and number of resting eggs produced. The model can be used for predicting the specific future development of cultures such as the process in rotifer resting egg production for aquaculture.

Individual- and population level effects of three antibiotics on the rotifers *Brachionus calyciflorus* and *B. plicatilis*

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The toxicity of three common antibiotics (tetracycline, tylosin tartrate, and streptomycin sulfate) to the marine rotifer *Brachionus plicatilis* and freshwater rotifer *B. calyciflorus* was investigated. Effects of each antibiotic on reproduction, lifespan, and several demographic parameters (Malthusian parameter, net reproduction rate, and three key properties of the net maternity function) were assessed at five nominal concentrations (from 5.6 to 720 mg L^{-1}) and a control (without antibiotics). Test animals were obtained by hatching resting eggs and culturing them individually in wells with 100 µL test medium and at chosen food concentrations. The number of offspring from each test animal was counted daily, and lifespan was recorded at death. Lowest Observed Effect Concentrations (LOECs) were determined for reproduction and lifespan; 1% and 10% Inhibitory Concentrations (IC_1, IC_{10}) and 95% confidence intervals were estimated for all endpoints. All *B. plicatilis* LOECs were 90 mg L^{-1} (reproduction and lifespan for all three antibiotics). For *B. calyciflorus*, tylosin LOECs were 90 mg L^{-1} for reproduction and lifespan, tetracycline LOECs were 5.6 mg L^{-1} for reproduction and 90 mg L^{-1} for lifespan, and streptomycin LOECs were 5.6 mg L^{-1} for both reproduction and lifespan. All IC_1 and IC_{10} estimates were well above 1.0 µg L^{-1}, which is roughly the maximum antibiotic concentration likely to occur in natural water bodies.

Distribution of rotifers in the family Brachionidae in Thailand

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Biodiversity and distribution of rotifers in the Family Brachionidae in Thailand were investigated. Five hundred and seven samples were collected from 294 sampling sites in two occasions during dry and rainy seasons from April 1998 to January 2004. Twenty seven species and 9 forms belonging to 5 genera were identified. *Brachionus* was the most diverse genus, comprising of 17 species and 9 forms while *Plationus* and *Platyias* were the least diverse genera, only one species was found. The most frequently encountered species were *Brachionus falcatus* Zacharias (51.02% of the sampled localities), *Keratella cochlearis* (Gosse) (50.68% of the sampled localities), *B. angularis* Gosse (50.00% of the sampled localities), *K. tropica* (Apstein) (46.60% of the sampled localities), *B. forficula* Wierzejski (45.92% of the sampled localities) and *Anuraeopsis fissa* (Gosse) (35.37% of the sampled localities). The infrequently encountered species were *Brachionus durgae* Dhanapathi, *B. sericus* Rousselet and *B. urceolaris* (Müller). These species were restricted to a single site only. The highest species diversity of 15 species was recorded in Nong Changpeuak and Bueng Srithat in Udonthani province, northeast Thailand. The correlation analysis shows that temperature and pH tend to be positively related to species diversity while conductivity and salinity tend to be negatively related to species diversity.

Ecology and ethology of Colonial Rotifera

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We recorded from different parts of North Eastern India seven species of colonial Rotifera: *Lacinularia elliptica* Shephard, 1897, *L. flosculosa* (Müller, 1758), *Sinantherina socialis* (L., 1758), *S. procera* (Thorpe, 1893), *S. semibullata* (Thorpe, 1889), *S. spinosa* (Thorpe, 1893) and *Conochilus unicornis* (Rousselet, 1892). Interestingly, these colonial forms possess some specific ethological characteristics. During the food collecting process, contraction and relaxation of body of these rotifers occurred. In response to external stimuli, body contraction was observed. These species, however, differ in their selection of substrate and ambient physico-chemical characteristics of water. Interestingly, the shape and size of the colony differ in different species. For example, the colony of *L. elliptica* is very large, showing up to 192 individuals at a time with variation in age of the individuals. The elliptical shape of the colony possesses broader end with larger number of adult individuals, whereas the narrower end contains mostly larvae and pre adults. On the other hand, in *L. flosculosa*, the individuals form a true colony only during their adult stages.

Impact of rotifers on growth potential of a rare fish *Nandus nandus* with reference to environmental temperature

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The impact of mass production of planktonic rotifers has been observed over growth potential of fingerlings of the freshwater fish *Nandus nandus* under different climatic conditions. The results showed that during summer period (30 ± 3^0C) total calories of food (rotifers) consumed by fingerlings in a month was 22.3 ± 3.4 Kcal, while in winter (16 ± 2^0C), the consumption was only 5.4±0.4 Kcal. It indicated that the rate of food consumption was larger during summer months. The percent of total calories lost (24±2%) through faecal matter in winter was much greater (17±1) than during the summer. Further, the caloric value of faecal matter in winter was 7.5 ± 0.7 Kcal g^{-1}, while in summer it is much less (4.891 ± 0.292 Kcal/gm). Therefore, it appeared that the climatic parameter not only affected the rate of prey consumption but also affected the magnitude of loss of faecal matter. During summer months the caloric value of fish tissue was 5.7 ± 0.4 Kcal g^{-1}, while in winter it was 4.0±0.7 Kcal g^{-1}. Further, in summer total gain in terms of dry weight was 0.81±0.32 g, whereas in winter there was a loss in (-0.18±0.01 g). Although rotifers may be largely acceptable as growth promoting food organisms for the fishes, climatic conditions, mainly temperature, could play an effective role.

The hydrographical and anthropological environments as rotifer habitats

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On the territory of Upper Silesia (Southern Poland) there are many anthropological hydrographic environments which have originated as a result of industrial mining activity. Some of them are localized on old mine dumps of coal wastes which are the sources of fluvial rivers. In such cases, the anthropological water sources are found flowing in the dumps. Other kind of specific artificial aquatic environments are the old galleries, serving as gravitational drainage passages for excavating silver and lead. The water flowing from the galleries forms rivers, some with strong, low flow or stagnant. Rotifers inhabiting these extreme habitats are the evidence of their great ability for adaptation to these conditions. Three artificial flowing sources in coal pits and two flowing from galleries were studied. The water flowing from a coal mine gallery was always characterized by a constant temperature and a high conductivity caused by the presence of ionized sulphates, chlorides and soda. In general, in artificial sources in dumps there were 6 or 7 species of rotifers, and 9 to 17 species were found in the outflow from galleries. The mean abundance of rotifers in this artificial microhabitat was very low and fluctuated between 3 – 16 ind. dm^3 water. The greatest numerical abundance (56 ind. dm^3) was attained by the rotifer *Proales theodora* in the "Fryderyk" gallery followed by *Colurella adriatica* (51 ind. dm^3) in the Mikołów drain off.

Genetic Diversity in sexual and asexual rotifers

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At the Rotifera X Meeting (Ilmitz, Austria), I described a species concept for asexual organisms, the Evolutionary Genetic Species Concept, which identifies inclusive populations that are independent arenas for the evolutionary forces of mutation, selection and drift. I showed how such species can be detected in bdelloid rotifers using phylogenetic trees of mitochondrial *cox1* gene sequences using the 4-times rule. The species defined thus are comparable in many ways to species in Monogononta and other sexual organisms as described by the Biological or Genealogical Species Concepts. In particular, we can compare the genetic diversity within asexual species in the bdelloids with that in sexual species in the monogononts. We measured nucleotide diversity (mean pairwise sequence difference) in the *cox1* genes of a sample of more than 37 species of bdelloids and 10 species of monogononts. Diversity in both monogononts and bdelloids is of the same order of magnitude as that in other invertebrates. Our sample may underestimate diversity but not an order of magnitude. Unless the mutation rate is remarkably low in rotifers, this result contradicts the suggestion that microscopic organisms have extremely large effective population sizes. Diversity in the bdelloids was slightly less than half as high as that in the monogononts; qualitatively, this is compatible with more frequent periodic selection in the strictly asexual bdelloids.

Genetic and ecological differentiation among rotifer populations in Eastern Spain

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Natural selection may play a strong role in the genetic differentiation of populations by promoting local adaptation. This role will be even more marked in cyclically parthenogenetic zooplankton, which combines parthenogenesis and sexual reproduction. Natural selection can act very efficiently against the less-adapted genotypes during the parthenogenetic phase of the life cycle. Previous studies have reported patterns of genetic differentiation among rotifer populations inhabiting Eastern Spain. However, it is unknown if this differentiation occurs in ecologically relevant traits. Thus, the relative roles of natural selection and genetic drift on this differentiation remain unexplored. The latter issue can be addressed by comparing between-population genetic differentiation in neutral markers and in ecologically relevant, quantitative traits. Here, we report on both ecologically relevant trait variation and neutral genetic differentiation among populations of the monogonont rotifer *Brachionus plicatilis*. One hundred and eighty clones from six populations differing in salinity (30 clones per population) were tested at four environmental conditions in a common-garden experiment by measuring several life-history traits. Populations responded differentially to experimental conditions, showing genetic and ecological differentiation among rotifer populations within the same species and suggesting local adaptation related to salinity in at least one population. Moreover, we compared levels of differentiation in the quantitative traits with those found in a genetic marker, the mitochondrial gene cytochrome oxidase subunit I.

Rotifers of Coyuca de Bbenitez, Guerrero, a coastal lagoon of Mexico

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The Coyuca de Benitez coastal lagoon is located to the northwest of Acapulco (Guerrero State, Mexico) (17°27'36" - 16°46'48" N and 100°28'48" - 99°49'12" W) at 20 m asl with a total surface of 34 km^2. The principal tributary is the Coyuca river which flows all through the year. This lagoon has an ephemeral inlet, the mouth of approximatively 500 m wide is usually open only for a month or two in September and/or October every year. Two main economic activities realized in this lagoon are tourism and fisheries. The present study was aimed at determining the rotifer diversity of this. The samples were collected at 14 sampling sites at different depths and littoral vegetation. The ranges of physical and chemical parameters of the water were: salinity 0 to 22 g L^{-1}, water temperature 26-35°C, dissolved oxygen 1.04 - 6.75 mg L^{-1}, transparency 0.18 - 0.90 m, and the total depth of about 15 m. We found a total of 93 species representing 30 genera and 23 families. We recorded 13 new records to the Mexican fauna: *Keratella cruciformis, Keratella mixta, Cephalodella hiulca, Conochilus hippocrepis, Encentrum putorius, Epiphanes brachionus, Lecane donneri, Lecane kluchor, Squatinella donneri, Lindia tecusa, Ptygura barbata, Floscularia rigens* and *Testudinella elliptica*.

A morphometrical study on two natural populations of *Brachionus angularis* (Rotifera: Brachionidae)

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We quantified morphometric variations of natural populations of *Brachionus angularis* from two different water bodies, Xochimilco Lake (Mexico City) and Coyuca de Benitez coastal lagoon (Guerrero State, Mexico). Using digital image analysis, we measured the body length and width, corona width and distance between frontal spines of *B. angularis*. Statistical analysis showed significant differences in all the measured characters of the two populations. One important morphological difference was that the specimens from Coyuca de Benitez coastal lagoon had a pair of short anterolateral spines while those from Xochimilco Lake did not have.

Abundance, distribution and body size of *Keratella americana* (Ploimida: Brachionidae) in two dissolution lakes of Yucatan Peninsula

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Effect of biotic (vertebrate and invertebrate predation) and abiotic factors (dissolved oxygen, water temperature, pH, conductivity, chlorophyll *a* and transparency) on the abundance, distribution and body size of the limnetic rotifer *Keratella americana*, were evaluated in two closed, dissolution lakes: "El Padre" (EP) and "Minicenote" (M) in Quintana Roo, Mexico, on annual cycle (June 2003-May 2004) using a Forward Selection Procedure. The model showed that the spatial distribution and abundance of *K. americana* in EP is governed by environmental limitations [$y=-0.9+(0.51\bullet x_1)-(0.85\bullet x_2)-(5.82\bullet x_3)$; x_1=pH, $x_2=PO^{-}_4$, x_3= transparency], as a result of the mesotrophic conditions and anoxic layers in the bottom. In M, the population responds to thermal features, and variables related with production [$y=1.06+(0.85\bullet x_1)+(1.78\bullet x_2)-(0.77\bullet x_3)$; x_1=temperature, x_2=oxygen x_3= transparency], probably as consequence of oligotrophic characteristics. In both systems, *K. americana* was more abundant in the dry and rainy seasons; in concordance with the greater primary production. Therefore, the temporal variation of *K. americana* is related to food availability. On the other hand, the body size of *K. americana* in EP, is larger in winter storm, than it is during dry season. The gut content analysis of nekton, showed its highest predation pressure on *K. americana*, which happened when it displayed the highest abundance. In contrast, differences in body size by season in M were not found, probably by the lower fish abundance in the system. Finally, the larvae of the phantom midge *Chaoborus* sp. (invertebrate predation), did not affect the rotifer community structure in the ecosystms studied. *Chaoborus* did not prefer *Keratella*, and hence the population structure of the rotifer was not related to the abundance of the larvae. These results have been discussed in terms of dynamics of tropical zooplankton communities.

Are deep, oligotrophic lakes inhospitable environments for rotifers?

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Resource competition is an important factor influencing the dynamics of zooplankton populations and the structure of zooplankton communities. Rotifer populations are often extremely dynamic, exhibiting large fluctuations in abundance on very short time scales. Experimental studies during the last decade have lead to the general conclusion that herbivorous planktonic rotifers are usually food limited, despite the trophic status of the lake. In addition, diel vertical movements of rotifers are normally restricted to a few meters; they are never as dramatic as those typical of competing microcrustaceans. This restriction could be another disadvantage in a deep (>60m) oligotrophic lake Alchichica in which food has a strong vertical and temporal heterogeneous distribution, with inedible algae composing most of the phytoplanktonic biomass, and with persistent a calanoid copepod populations. We studied the food limitation (food supplementation experiments) and vertical migration (migration traps) of two rotifer species, *Brachionus plicatilis* complex and *Hexarthra jenkinae*. We found that most of the times that rotifers were present; they were food limited, irrespective of the season. In addition, their vertical movements were restricted to the epilimnion and were almost negligible in the context of a water column of 60 m. We suggest that the observed irregular time course of rotifers in this lake is consequence of the interplay between harsh conditions such as scarce food availability and predation risk from planktivorous fish on the one hand, and the quick exploitation of opportunities, like sudden food availability or recruitment from the resting egg bank, on the other hand.

Rotifers inhabiting shells of testate amoebae (Protozoa), with description of new taxa

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Rotifers are known to live in close association, whether synoecious, commensal or parasitic, with a large and diverse number of animal taxa. Among these are the testate amoebae, a group of protozoans possessing a shell enclosing the body and provided with an aperture for the pseudopodia. Empty shells of *Nebela* and *Difflugia* are known to be inhabited by the bdelloids *Habrotrocha annulata* (Murray) and *H. angusticollis* (Murray), whereas *H. incola* Bartoš lives in empty tests of *Centropyxis aerophila* Defl. To date, only one monogonont, *Dicranophorus difflugiarum* Penard, has been reported parasitizing testate amoebae (*Difflugia acuminata* Ehrbg.). Two new species, inhabiting the shells of live *Arcella* and *Difflugia* respectively are presented here. The taxonomic position of the taxa is discussed.

Filtration activity of tree-hole bdelloid rotifer *Habrotrocha thienemanni* Hauer, 1924

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Habrotrocha thienemanni is a common, bdelloid rotifer that inhabits tree-holes where it can achieve high densities by feeding on bacteria. Under the laboratory conditions, I examined the feeding activity using inert fluorescent particles of 0.5 μm in diameter. Two kinds of particles were used: those coated in BSA protein and those not coated. Both particles were readily ingested, with clearance rates varying between 1.65 and 3.79 μl ind.$^{-1}$ h^{-1}. Particle uptake was related to body size of the animal. However, uptake of protein-coated particles was significantly higher than those that were not coated. Using the natural population density of *H. thienemanni* in tree hole of 56800 ind. l^{-1}, the clearance rates of the whole population were estimated to be between 981 and 5170 ml^{-1} d^{-1}.

Seasonal development and vertical distribution of soil rotifer populations in South-Bohemian beech forest

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The soil rotifer community of a climax beech forest in South Bohemia was investigated during 2005 with reference to seasonal and vertical distribution changes. The samples were divided into 5 well developed layers, which were processed separately. In all 34 rotifer species were identified during the investigation: 9 monogononts and 25 bdelloids. The most important species were *Encentrum arvicola* and *Wierzejskiella vagneri* among monogononts and *Adineta steineri*, *Ceratotrocha cornigera*, *Habrotrocha filum*, *H. ligula*, *Macrotrachela plicata*, *Mniobia tentans*, *M. incrasata* and *M. granulosa* among bdelloids. Mean Shannon diversity index per square unit varied from 1.99 to 2.63. the total rotifer abundance (mean±standard deviation) varied from 212±63 $X10^3$ to 513±127 $X10^3$ ind. m^{-2}, with the highest numbers being present in May and the lowest in July. Most of the community was concentrated in the upper (fresh litter), with the second in the partially decomposed litter layers. However, some species such as *Wierzejskiella vagneri* or *Mniobia incrasata* occured in deeper strata in some part of the season at least. The highest density (43 ind. g^{-1}) of rotifers was found in the upper layer in May.

Rotifers from tropical high-mountain lakes in Mexico

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The lakes El Sol and La Luna are located at 4,170 m asl, inside the crater of the Nevado de Toluca volcano (State of Mexico, Mexico). Both lakes are shallow (i.e., maximum depths = 15 m and 10 m, respectively), warm polymictic (water temperature = 5 to 11°C), and oligotrophic. To study the seasonal changes in the rotifer composition, montly samples were collected along the water column at the central and deepest part of the lakes. In all 29 rotifer taxa were identified (22 in La Luna and 21 in El Sol). The density of the zooplankton in La Luna was dominated by the rotifers all throughout the study period while in El Sol the density was higher during the second half of the sampling period. The zooplankton density was four times higher in El Sol than in La Luna. Maximum densities were observed in El Sol in November and December and in La Luna in July and September. Bdelloidea was dominant in La Luna and *Polyarthra dolichoptera* in El Sol. The rotifers were mostly found close to the bottom of the lakes probably as a mechanism to avoid the high U.V. radiation common in high-mountain lakes. It is noteworthy that the rotifer species richness found in the Nevado de Toluca lakes is unusually high when comparing to temperate as well as other tropical high mountain lakes. This study also added *Aspelta psitta*, *Hexarthra bulgarica*, *Cephalodella delicata* new to the Mexican fauna.

Preliminary analysis of spatial and seasonal variability in hatching of rotifers egg bank of a deep tropical lake in Central Mexico

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We present preliminary results of the analysis of dormant structures of rotifers in a deep (Z_{max}= 64 m), tropical lake, Alchichica (Central Mexico). We address the following questions: (1) Are there differences of species composition and diversity in the horizontal distribution (littoral vs. pelagic zone) of the lake? (2) Are emergence rates and species diversity different along the year cycle? (3) Are there differences in hatching rate and diversity of eggs from different ages? We obtained sediment samples from both the littoral and the deepest zones on the lake. A sediment core from the deepest zone, sliced at 1-cm intervals, as well as Eckman dredge samples of recent sediments (upper 5 cm) were explored. We analyzed emergence of eggs under two combinations of temperature, photoperiod and light intensity, simulating the coldest and warmest periods in the lake. Up to now, only two rotifer species have hatched in all treatments. The most frequent is a species of the *Brachionus plicatilis* complex, and rarely *Hexarthra jenkinae*. Results on *B. plicatilis* complex showed that hatching in the deep zone is higher than in the littoral zone. Hatching percent was about 22-30% of viable eggs, and no significant differences were observed among the combinations of tested emergence cues. We observed emergence just in the upper 4 cm of the sediments, and emergence rate was significantly lower in deeper strata. We have discussed our results considering the cost of resting egg production in deep lakes with sediments in permanent darkness and anoxia.

The effects of three potential endocrine disruptors on the sexual reproduction of the rotifer *Brachionus calyciflorus*

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Cyclical parthenogens such as monogonont rotifers require ephemeral periods of successful production of resting eggs. Failure to successfully produce resting eggs has severe consequences. These periods allow for genetic recombination and future establishment of rotifer populations. We investigated the disruption of sexual reproduction in *Brachionus calyciflorus* by three putative endocrine disrupting chemicals. Preston et al. (2000) documented potential disrupting chemicals on asexual and sexual reproduction in a 96-hour life cycle study. This research demonstrated that three chemicals adversely affected sexual reproduction without affecting the asexual reproduction. The chemicals that demonstrated effects were flutamide, nonylphenol, and testosterone at 10 µg L^{-1}, 50 µg L^{-1} and 10 µg L^{-1}, respectively. These concentrations are at least an order of magnitude lower than previously published no-observed-effect-concentrations (NOEC). We used these three chemicals at the reported concentrations to investigate if the male or female caused the disruption of resting egg production. We report here the results of cross-mating experiments, which combined the treated and untreated males and females, to reveal which sex was affected by the three chemicals. We also report data on the effects of algal concentrations on these potential endocrine disrupting chemicals, as well as provide data on the morphology of treated vs. untreated male sperm.

Combined effects of algal food (*Chlorella vulgaris*) concentration and temperature on the population dynamics of *Brachionus angularis* (Rotifera: Brachionidae)

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B. angularis is one of the common brachionid rotifers in many freshwater bodies in Mexico. We studied the population growth of *B. angularis* at two temperatures (22 and 32±1°C) and three algal food levels ($0.5X10^6$, $1.0X10^6$ and $2.0X10^6$ cells ml^{-1} of *Chlorella*) for 15 days. For each treatment, we maintained four replicates. Regardless of food level, increased temperature resulted in higher population growth rates (r). Similarly, regardless of temperature, increase in food level resulted in increased population abundances of *B.angularis*. Statistically significant food x temperature interaction was evident for peak abundances. Morphometric data on the egg bearing adults of each treatment were also presented. The results were discussed in light of food and temperature interactions in nature.

Rotifer abundance and diversity in benthic microhabitats of a littoral zone in Lake Mikołajskie (North-eastern Poland)

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Microzonation and seasonal dynamics of hydropsammon rotifers were studied in psammolittoral of Lake Mikołajskie (North-eastern Poland) in 2005. The aim of the study was to test the hypothesis that epihydroarenal plays a role of a transitory microzone between adjacent water layer and endohydroarenal. Ninety-four monogonont species were found with endopsammon community being the richest in species. Of these species, 15% were restricted to adjacent water, 20% to endohydropsammon and only 5% to epihydropsammon. Twenty-six (28%) species were observed in all three zones. From 57 species of epipsammon 52 (91%) were common with neighboring microzones. Rotifer densities were highest in the endohydroarenal zone if expressed in area units. However, the rotifer densities in epipsammon if expressed in the same volume of occupied environment were ca 40 times higher than in adjacent water and about twice of those noted in endohydroarenal. This may suggest that epihydroarenal plays a role of both a transitory zone and a refugium for animals from adjacent water layer.

Life history strategies of *Brachionus havanaensis* subject to vertebrate (*Ambystoma mexicanum*) and invertebrate (*Megacyclops* sp.) predation

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Several brachionids are known to show changes in their life history strategies and develop spines in response to predation. Here we evaluated the different strategies of the rotifer *Brachionus havanaensis* in the presence of vertebrate or invertebrate predators. We used conditioned-water in which the salamander (axolotl, *Ambystoma mexicanum*) and the copepod (*Megacyclops* sp.) were maintained separately for 72 h prior to conduct the experiments. Population growth and life table demography of *B. havanasensis* were studied at 22±2ºC using two food (*Chlorella vulgaris*) concentrations ($0.5X10^6$ and $1.0X10^6$ cells ml^{-1}). In all there were 24 jars (3 test waters, including controls X 2 food levels X 4 replicates) for either experimental design. Experiments were initiated with cohort or populations of 20 individuals in 20 ml of test medium. Morphometric information of *B. havanaensis* was also obtained. Our results indicated that growth rates were higher in the presence of predator infochemicals than in the controls. Data have been discussed with emphasis on the implications of these findings in nature.

Rotifer fauna of the flooded area of the nature park Kopački rit (Croatia) during treatment with *Bacillus thuingiensis israelensis*

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Natural Park Kopački rit is flooded area of the rivers Drava and Danube. It is situated at the north-east part of Croatia and is one of the most preserved swamps in the Europe. During the high water-levels in the Danube River, water overflows to lower area where many small ponds retain it for the next 45 to 90 days. Mosquito larvae which develop in these ponds each year are treated with *Bacillus thuingiensis israelensis* (*B.t.i.*). We aimed to test if *B.t.i.* has any impact on zooplankton community in these ponds. Samples of zooplankton and benthos community were collected before and after treatment with *B.t.i.* by standard methods on four sites in April, May and June. Standard physical, chemical and bacteriological parameters were also measured. Most numerous group in zooplankton community was the Rotifera represented by 20 species, followed by three species of Cladocera; Copepoda were represented only by copepodite forms and naupliar stages. Before treatment the rotifer density was < 30 ind. L^{-1}, which was probably due to competition for food from the mosquito larvae and the short duration of water in the ponds. During this short period rotifers (*Brachionus angularis*, *B. qudridentatus, Filinia longiseta, Kellicottia longispina*, *Keratella cochlearis* and *K. quadrata*) were dominant. After treatment with the *B.t.i.* in April the number of rotifer desity was smaller than before treatment. It was probably due to the consequence of successions which depended on water retention time rather than the effect of *B.t.i.* which degrades very quickly. The rotifer density increased again in May and June but as a result of the organic matter inflow. The dominant species were *Keratella cochlearis* (and its form *tecta*) and *K. quadrata* as well as representatives of higher trophic levels.

Morphology of *Hexarthra mira* (Hudson, 1871) using SEM

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Hexarthra mira is a characteristic pelagic rotifer found in eutrophic ponds and lakes. We here present a study based on our refined SEM technique. *H. mira* is characterised by six strongly bristled arms, a distinctive rotatory organ and a pair of caudal appendages. The corona is conspicuously positioned at the top of the head. It consists of a circumapical band and a ciliated oral field. The circumapical band encloses a field without locomotory cilia. There are, however, tufts of sensory cilia. The circumapical band is interrupted dorsally. This is where the trochus turns into the cingulum. The mouth region can be divided into a praeoral and a postoral ciliary field prolonged into a lip-shaped outgrowth. Whereas the dorsal and the ventral arms are unpaired, both the dorso-lateral and ventro-lateral arms are paired. The lateral antennae are positioned on the ventro-lateral arms just above the point of insertion of the skipping bristles. The unpaired dorsal antenna is positioned on a hump-shaped protrusion of the dorsal body wall. This example of *Hexarthra mira* demonstrated that with carefully prepared specimens scanning electron microscopy could reveal a broad range of morphological structures.

A new *Cephalodella* species (Notommatidae, Rotifera) from North-West Germany

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We here report a putative new rotifer species from North-West Germany. This species was found in a pH-neutral ditch near Leer (Lower Saxony, Germany) in November 2005. The body is elongate, cylindrical with a soft hyaline lorica and ill-defined plates. The short claw-bearing toes, usually sligthly retracted, are approximately 1/5 of the total length and bent dorsally. The convex corona is nearly terminal without lips. Eyespots are missing. A small RCO and subcerebral glands are present. The trophi is of type D with asymmetrical unci. There are three pairs of salivary glands; one lateral, one ventral and one dorsal. The present species resembles *Cephalodella tenuiseta*, especially its variation *americana*. This variation has indepently been described by three different authors with some variation in trophi, toes and body shape. The new species can be distinquished from *C. tenuiseta* by its shorter and thicker, claw-bearing toes which are fairly wide apart. Additionally, the trophi of the new *Cephalodella* differs from that in *C. tenuiseta* in having asymetrical unci with two uncinal teeth on the left and one on the right uncus. The inner margins of the rami are sligthly different as well. Observations were carried out using light- and electron microscopy.

Incomplete induction of mixis in *Brachionus calyciflorus*: patterns of reproduction at the individual level

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In the rotifer *Brachionus calyciflorus* mictic-female production is density-dependent and induced by a chemical produced by the females themselves. Even at the highest densities, however, populations never become entirely mictic, i.e. some amictic females continue to be produced. Surprisingly, the phenomenon also occurs in clonal laboratory populations with genetically identical individuals. Here we studied how this ecologically adaptive phenomenon was generated at the level of individual reproducing females. In a life-history experiment we subjected 120 amictic females of a clone of *B. calyciflorus* to a daily stimulus of culture medium conditioned at a density of 30 females ml^{-1}. For each of these mothers we isolated the life-time offspring individually and recorded whether these females were amictic or mictic. Mothers produced on average 16 offspring but none of the mothers produced 100% mictic offspring; the average proportion of mictic females was 30%, despite the extremely strong stimulus. The distribution of amictic vs. mictic offspring was not uniform over the mothers' lifetime. Early and late offspring had a low probability of being mictic whereas mid-aged mothers produced the highest proportion of mictic daughters (up to 56%). We conclude that not all oocytes of *B. calyciflorus* can be turned into mictic females, even if the mictic-female-inducing stimulus is extremely high. Moreover, the propensity to become a mictic female depends on the rank of an egg within a female's offspring production.

Effect of pulsed exposure to heavy metals (Cu and Cd) on some population variables of *Brachionus calyciflorus* Pallas (Rotifera: Brachionidae)

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Heavy metals such as cadmium and copper, which are used for both industrial and domestic purposes, may eventually reach freshwater bodies affecting different groups of zooplankton including rotifers. In this study, we evaluated separately the impact of different concentrations of two heavy metals (0.0375, 0.075, 0.15 mg L^{-1} for $CuSO_4$ and 0.025, 0.05, 0.1 mg L^{-1} for $CdCl_2$) and with different durations of exposure (3, 6, 12 and 24h) on the population growth of *B. calyciflorus* at one algal food density ($1X10^6$ cells ml^{-1} of *Chlorella vulgaris*). Controls contained only alga. For each heavy metal concentration and exposure time, we maintained 4 replicates. At each treatment, the egg-bearing adults were used for morphometry and eggs for hatching success. Regardless of exposure time, at the highest concentrations, both the heavy metals reduced the population growth of *B. calyciflorus*. For a given heavy metal, generally increase in the toxicant concentration and the exposure period had an adverse effect on the population growth rate (*r*) of *B. calyciflorus*. The response of rotifers was similar for both the heavy metals; however, due to Cd the magnitude of impact was more severe. Data on the hatching success showed a reduction in the viability of the parthenogenetic eggs with increasing metal concentration and the duration of exposure.

Influence of heavy metal (Cu and Cd) exposure time and concentration on the predator's (*Asplanchna brightwelli*) population growth

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Prey-predator interactions are the most common and the major linking forces regulating plankton communities in freshwater ecosystems. In nature these interactions are affected by seasonally varying factors such as temperature, pH and food availability. However, anthropogenic pollutants including heavy metals such as copper and cadmium may eventaully reach aquatic ecosystems affecting the planktonic communities through their influence on the prey-predator interactions. In this work, we evaluated the combined effects of heavy metal (Cu and Cd, separately) concentration (0.1 and 0.2 mg L^{-1}) and exposure time (6 and 24h) on the population growth of the predatory rotifer *Asplanchna brightwelli* (predator) grown on the prey *Brachionus calyciflorus*. In general, at the concentrations used in this study, copper had no significant influence on the population growth of *A. brightwelli*. However, under longer exposure period and higher Cu concentration, the predator´s population became extinct after the day 7. The trends on population growth of *A. brightwelli* under the influence of Cd were similar to those under Cu, but the magnitude of negative impact was higher. The period of exposure had far greater negative influence than the concentration of the heavy metals on the population growth rates of *A. brightwelli*.

Combined effects of sediment and lead (Pb) on the demography of *Brachionus patulus* (Rotifera: Brachionidae)

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Rotifers in nature are subject to stress due to several factors including pollutants, the impact of which is generally reflected in their demography. In Mexico, several lakes that receive inflows of pollutants are shallow with high turbidity levels almost throughout the year. Sediment is known to mitigate the adverse effects of toxicants to zooplankton such as cladocerans. Here we studied the response of *Brachionus patulus* to different concentrations of the heavy metal lead in the presence and absence of sediments. We conducted acute (LC_{50}) and chronic (life table demography and population growth) toxicity tests using sediment levels of 0, 30 and 280 mg L^{-1} and Pb concentrations of 0, 0.06 and 0.6 mg L^{-1}. For each of the two chronic toxicity tests, we used 36 experimental jars (3 turbidity levels by 3 Pb concentrations by 4 replicates). Experiments were conducted at 20±1ºC on a horizontal shaker to maintain the sediment in suspension. *B. patulus* was introduced into 50 ml glass jars, each with 30 ml of test medium of desired sediment level and Pb concentration and with the algal food *Chlorella vulgaris* at $1x10^6$ cells ml^{-1}. We observed an adverse effect of high sediment levels and lead concentrations on the survivorship and growth rates of the test species. The impact of our findings has been discussed in relation to waste discharge from industries into natural waterbodies.

Rotifer diapausing egg banks: past studies and recent findings

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In this contribution we review studies on the demography of rotifer diapausing egg banks. We focus on the abundance, viability, age, and vertical distribution of diapausing eggs in the sediments, as well as optimal patterns of diapausing egg hatching in relation to habitat conditions. Despite their importance for rotifer population dynamics and evolution, rotifer diapausing egg banks have been little studied if compared to other zooplankton groups. Moreover, until recently most zooplankton egg bank studies were typically restricted to deep waterbodies, whereas temporary, shallow ponds were practically ignored. Rotifer studies report that viable diapausing eggs in surface sediments may be quite abundant (up to 4000 eggs cm^{-2}), and the maximum egg age ever reported is 100 years. A recent research project performed on diapausing egg banks in 15 shallow ponds suggests that interannual production of diapausing eggs is highly variable in these habitats and that most of the viable eggs are located in the uppermost 2 cm sediment layer. Accordingly, compared to maximum survival times for other zooplankton species, rotifer egg banks seem to be younger. Recent studies show that the health of diapausing eggs can be recognized in the sediments ('healthy-looking', 'deteriorated' and 'hatched' eggs). The abundances of these egg categories, if interpreted by using mathematical dynamic models, give information on the processes in the sediment. A striking finding is that egg deterioration rates are high (about 90% 'deteriorated' eggs) suggesting that resting in the sediment is not as safe as previously thought. By finding hints for deterioration, hatching and production of diapause eggs, these processes can be related to habitat features, such as sediment harshness or unpredictability of water column.

Phylogenetic relationships among syndermata inferred from nuclear and mitochondrial gene sequences

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Phylogenetic relationships among Syndermata have been extensively debated, mainly because the sister-group of the Acanthocephala has been unclear from analyses of morphological and molecular data. Herein we sampled the 4 classes of Acanthocephala (Archiacanthocephala, Eoacanthocephala, Polyacanthocephala and Palaeacanthocephala) and the 3 rotifer classes (Bdelloidea, Monogononta and Seisonidea) and inferred phylogenetic relationships using small-subunit (SSU) and large-subunit (LSU) ribosomal DNA, and cytochrome *c* oxidase subunit 1 (*cox*1) sequences. These nuclear and mitochondrial DNA sequences were obtained for 27 acanthocephalans, 9 rotifers, and representatives of 6 phyla that were used as outgroups. Maximum parsimony (MP), maximum likelihood (ML) and Bayesian analyses were inferred from nuclear rDNA (SSU + LSU) and from the combined sequence dataset (SSU + LSU + *cox*1 genes). Phylogenetic analyses of the combined rDNA and *cox* 1 data uniformly provided strong support for a clade including rotifers plus acanthocephalans (Syndermata); this group was evident with strong support regardless of inference method. Strong support was also found for monophyly of Acanthocephala in analyses of the combined dataset or rDNA sequences alone. Within the Acanthocephala each class was strongly supported as monophyletic; relationships among acanthocephalans yielded Archiacanthocephala as the sister group to the remaining taxa. Analyses of the combined dataset revealed a sister group relationship between Acanthocephala and Bdelloidea by parsimony, likelihood and Bayesian methods. The reliability of this clade was generally strong, and alternative topologies depicting a different rotifer sister-group of Acanthocephala (or monophyly of Rotifera) were significantly low. In this paraphyletic assemblage of rotifers, the relative positions of Seisonidea and Monogononta to the clade Bdelloidea + Acanthocephala were inconsistent among trees based on different inference methods. These results indicate that Bdelloidea is the free-living sister group to acanthocephalans, which should prove key for comparative investigations of the morphological, molecular, and ecological changes accompanying the evolution of parasitism.

Rotifers diversity in Centro de Investigaciones Biológicas y Acuícolas de Cuemanco (CIBAC), Xochimilco (Mexico)

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CIBAC is situated at 19°15'7.2" N and 98°58'26" W besides Cuemanco Chanel of Xochimilco Lake. CIBAC has experimental ponds of different sizes and one rustic canal for aquaculture, all of which were sampled in order to evaluate the diversity of rotifers that developed there. The samples were collected in 2005 using a zooplankton net of 30 µm mesh size, and selected physico-chemical variables were also measured. A total of 58 species representing 23 genera and 18 families were found. *Asplanchna brightwelli*, *Brachionus angularis*, *Brachionus calyciflorus*, *Keratella americana*, *Keratella tropica*, *Lecane bulla*, *Lecane closterocerca,* and *Filinia longiseta* were the more abundant species. *Albertia crystallina,* new record to the Mexican fauna, was also observed.

Rotifers of Mexico

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This work was done in order to give a taxonomic listing of all reported species of Mexican rotifers, as well as the localities in which were they were found and the references. The information reviewed provides 386 valid species representing 71 genera and 28 families.

Intraclonal variation for propensity to produce mictic daughters in the rotifer *Brachionus*: variation among replicate females and with maternal age

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In 12 experiments with three species of *Brachionus*, females cultured singly in small volumes from birth through most of their reproductive period produced high proportions of mictic daughters. These data are analyzed to test two hypotheses regarding variation in the propensity of replicate females to produce mictic daughters. As predicted, significant heterogeneity among females (χ^2) occurred in seven of nine experiments with clones of *B. calyciflorus* from Florida, Georgia, Texas, Spain and Australia, and in one experiment with a clone of *B. angularis* from Argentina. Possible causes, and ecological effects, of this heterogeneity are discussed. There was no significant heterogeneity among replicate females in two clones of *B. calyciflorus* from Florida and in a clone of *B. variabilis* from Spain. Little support was provided for the hypothesis that the propensity of a female to produce mictic daughters changes with her age. In six experiments with *B. calyciflorus* using six different clones from four strains, only one experiment showed significant variation in the proportion of mictic daughters produced in three or four successive one-day periods.

Morphological variation of *Keratella cochlearis* in Myanmar in relation to zooplankton community structure

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Plankton samples from 35 waterbodies in Myanmar (Burma) showed *Keratella cochlearis* present in 26, and most abundant in 10. Measurements of lorica length and posterior spine length were made from 20 localities. The length of the posterior spine varied with lorica length, but also with the composition of the crustacean zooplankton. Long spines were associated with dominance by the copepod *Heliodiaptomus.* The shortest spines were found in samples dominated by cladocerans or cyclopoid copepods. Quantitative estimates of diaptomid copepod abundance were made for 10 localities. Posterior spine length was positively correlated with the number of diaptomids per unit volume. Forms without posterior spines were found in 17 localities. The lorica lengths of these spineless forms were generally similar to those with spines, but in a few samples the loricas of the spineless forms were significantly larger. These samples were dominated by cladocerans or cyclopoid copepods. In a single locality spineless forms were found in the absence of spined forms. The crustacean zooplankton in this locality was also dominated by cladocerans. These results have been discussed in relation to known variation in *K. cochlearis* elsewhere in the world.

Ultrastructural comparison of the trophi among the loricate species of the genus *Brachionus* Pallas, 1766 fro the State of Aguascalientes (Mexico)

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Taxonomic studies from the State of Aguascalientes have revealed the occurrence of 11 species of *Brachionus* so far. However, most of the studies were based on light microscopic observations. Using ultrastructural analysis by SEM JEOL LV 5900, we noticed finer details in the trophi of different species of *Brachionus*. Based on these differences, it is possible to distinguish different *Brachionus* species and thus taxonomy of this genus becomes easy.

The relationship between trophi and body size of rotifer *Brachionus plicatilis*

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Rotifers form a food resource for many fish species during larval stage. Most of them are digested quickly on reaching the stomach of fish larvae, but their trophi remain undigested. If one can predict the rotifer size based on the trophi size, food-size preference of different fish species at different life stages may be inferred. We used one strain each of *Brachionus plicatilis*, *B. ibericus* and *B. rotundiformis*. Although the shapes of the trophi appear very similar among the three rotifer species, the fulcrum of the last two last species appear similar (like two sticks) but is different from *B. plicatilis* (cone-shaped). The size differences between the three types of rotifer trophi are clear. For all the three species, lorica length increased significantly from 0 to 12 h after hatching, but not later than 18 h. All these showed positive correlationship between the trophi size and the lorica length. The best correlation was found for trophi width, ramus width and ramus length.

HUFA enrichment of the rotifer *Brachionus plicatilis* with different commercial products for use in fish larval culture

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The enrichment and retention of highly unsaturated fatty acids (HUFA, particularly EPA and DHA) was investigated in the rotifer *Brachionus plicatilis* using several commercial enrichment products. Aqualene (Takeda-Kagakushiryo), DHAce (Oriental Yeast Co.), Protein Selco (INVE), Super-rotifer (Higashimaru) and microcapsules made with squid liver oil, were used to enrich the rotifers for 12 h. The enrichment process was made according to the manufacturers' instructions in triplicate 200-L tanks for each enrichment product. Samples were obtained at every 3, 6, 9 and 12 h for evaluating the rotifer density, contents of total lipid, EPA and DHA. At the same time, ammonia and dissolved oxygen levels in the enrichment media were also measured. Rotifer fatty acid profiles for each treatment were determined by gas chromatography, after extraction of total lipids and lipid class determination. Rotifer density in the different products did not show changes during the enrichment process, excepting those enriched with Aqualene, which decreased in 50%. The maximum contents of DHA-EPA were observed in rotifers enriched with the squid liver oil microcapsules after 6 h of enrichment (DHA= 73.2, EPA=46.2 mg g^{-1} of dry weight), the DHAce enriched rotifers (DHA= 64.8, EPA=46.3 mg g^{-1} of dry weight) after 12 h of enrichment, Protein-Selco (DHA= 65.7, EPA=24. mg g^{-1} of dry weight) after 3 h of enrichment, Super-rotifer (DHA= 32.2, EPA=11.2 mg g^{-1} of dry weight) after 3 h and Aqualene (DHA= 7.3, EPA=2.7 mg g^{-1} of dry weight) after 12 h of enrichment. From these results, it is concluded that best enrichment products that may be used in larviculture are the squid liver oil microcapsules and the Protein Selco, due to the high contents of EPA and DHA in the rotifers after a short period of enrichment.

The long-term aspect of the winter rotifer community of Neusiedler See (Austria), with special reference to the development of *Rhinoglena fertöensis*

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Zooplankton densities and biomass and the population parameters of the dominant species from Neusiedler See, a shallow lake in central Europe, were monitored for 35 years. Here, I focus upon the winter rotifer plankton community, particularly *Rhinoglena fertöensis*. Between 1968 and 2003, mean winter rotifer densities varied between <10 ind. L^{-1} and 468 ind. L^{-1}. High winter rotifer densities (maximum about 2000 ind. L^{-1}) were recorded in years when *Rhinoglena* developed very well, and represented nearly 100% of the rotifer community. Only in a few years with high winter densities of *R. fertöensis*, was *Synchaeta* sp. (*tremula-oblonga* group) or *Keratella quadrata* dominant. *Rhinoglena fertöensis* is a cold stenothermal species that occurrs mainly in the winter months. In Neusiedler See, it is a typical planktonic species, which is restricted to temperatures below 15°C, and reaches its maximum when the lake is covered by ice. Population dynamics can be described in 3 phases: first phase is the hatching of resting eggs in autumn at the time of the steepest temperature decline; on the long-term average the first individuals appear at 12°C at the beginning of October. Hatching is scattered over a 4-6 week-period. The second phase resembles the exponential growth, which happens on average 60 days later; and peak densities are reached in the first half of March. At this time sexual reproduction begins, but at least a fraction of the population continues to reproduce by parthenogenesis. The third phase is a rapid decrease in numbers; during this period the percentage of mictic females carrying resting eggs is highest. The mean day of disappearance is the 5th of May, when the mean temperature is 15.5°C. The mean total period of occurrence is 211 days. The success of the *Rhinoglena* population is determined by temperatures during autumn and spring, by the food conditions during the exponential growth phase, and by the number of resting eggs produced.

3-D Cerebral Architecture and innervation in *Asplanchna* (Rotifera: Asplanchnidae)

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New information of the structure and circuitry of the cerebral ganglion is presented for the rotifers *Asplanchna brightwellii* and *A. priodonta*. Immunoreactivity to the neurotransmitters serotonin, FMRFamide, and SCPb, and the structural proteins tubulin and neurofilament, is described using confocal scanning laser microscopy and advanced imaging software. Novel three-dimensional reconstructions of the brain of each species reveal unambiguous differences in the quantity and distribution of immunoreactive neurons. *Asplanchna brightwellii* is characterized by the presence of an extensive serotoninergic network in the cerebral ganglion with several decussatory pathways; the number of serotoninergic neurons in *A. priodonta* is considerably lower and the brain appears to lack decussating neurons. The neurotransmitters FMRFamide and SCPb are present throughout the cerebral ganglion of both species and are found in peripheral neurons that innervate both the gastric glands and the cingulum. Specifically, the cingulum is innervated by a pair of nerves that extend out laterally from each side of the brain and encircle the apical field; these nerves coalesce below the mouth margin and form two large ganglionic masses. Immunoreactivity to tubulin and neurofilament also reveals novelties in cerebral architecture and organ system innervation. Anti-tubulin staining exposes the gross morphology of the brain and its innervation of sensory receptors in the apical field. Anti-neurofilament exposes additional aspects of cerebral circuitry and the innervation of sensory receptors in the trunk region. It is suggested that detailed wiring diagrams of the rotifer brain be incorporated into future studies of rotifer ethology and physiology to better understand how differences in circuitry may help explain the evolution of both behavior and lifestyle.

Diversity and zoogeography of Mongolian rotifers

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A three-year sampling program has recently been started to prepare an inventory of aquatic micrometazoa in Mongolia, a biologically rich but inadequately studied region in central Asia. Some of the most pristine freshwater environments in the world were sampled: many of these are now affected by global warming (permafrost thaw) and human activity (overgrazing). First results from boreal north and arid central parts suggest that rotifers comprise a large portion of biological diversity in both freshwater and inland saline habitats. Numerous records represent disjunct populations of species otherwise known only from West Palearctic or other parts of the Holarctic Region; others may be endemic to central Asia. A database is currently being developed to accommodate comprehensive species-level information for future morphologically-based phylogenetic analyses. It includes survey and literature-based distribution records, locality and habitat classification and specimen images. Setting up a local reference collection and training of local students will help to build in-country expertise for future biodiversity studies.

Rotifer (Monogononta) diversity from selected sites in the canals of Xochimilco Lake (Mexico City, Mexico)

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Lake Xochimilco and its associated canals are the remnants of a vast, ancient water body that once covered 125,000 ha in the Valley of Mexico. It has now been reduced to a shallow (depth 1.5 m) water body consisting of lakes and interconnecting canals, covering 190 ha in the south of Mexico City. In this study, we analyzed the density and diversity of monogonont rotifers collected from four different sites on a monthly basis over a calendar year. The physicochemical parameters analyzed were temperature, pH, conductivity, depth, dissolved oxygen, biological oxygen demand, alkalinity, hardness, phosphorus, orthophosphates, nitrogen, nitrates and chlorophyll *a*. We collected the rotifers by filtering 80 L of water from each site through a 50 µm sieve. The rotifers were identified to species and quantified using a Sedgewick-Rafter cell. The sampled sites were shallow and eutrophic with BOD_5 values ranging from 20 mg L^{-1} to 30 mg L^{-1}, and N:P ratios between 1 to 5. We found 65 rotifer species representing 16 families of which Brachionidae and Lecanidae were most dominant. In addition to the common species such as *Brachionus budapestinensis*, *B. calyciflorus*, *B. havanaensis*, *Keratella cochlearis*, *Keratella tropica* and *Polyarthra vulgaris*, we also found several interesting taxa including *Brachinonus durgae*, *Lecane aegania*, *L. venusta*, *L. tenuiseta*, *Trichocerca ruttneri* and *Filinia cornuta*. Shannon-Weiner species diversity index ranged from 1.9 to 3.8. The data have been discussed in relation to the importance of rotifer biodiversity studies in Mexico.

Effect of anti-androgenic substances on the sexual reproduction of the rotifer *Brachionus calyciflorus*

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Many substances released in the aquatic environment have been reported to have disruptive effects on endocrine systems of vertebrates and invertebrates. In rotifers, some of these chemicals have been shown to depress sexual reproduction at concentrations one to two orders of magnitude below those that affect asexual reproduction. The anti-androgen flutamide impacts sexual reproduction of rotifers at particularly low concentrations. We studied the effect of different types of putative endocrine disruptors on sexual reproduction of *Brachionus calyciflorus*. Contaminants tested were cyproterone acetate, an anti-androgen with a steroid-like structure, the pesticides fenitrothion and linuron which both have high affinity to the androgen receptor in vertebrates, although they are nonsteroidal molecules, and lindane a widely used pesticide that binds weakly to the androgen receptor in vertebrates. A series of endpoints relative to different steps of sexual reproduction were monitored to define critical functions affected and to get information on mechanisms of action. Fenitrothion, had the strongest impact on the sexual reproduction of rotifers. We observed a decrease in the proportion of mictic females among the ovigerous individuals and in the production of fertilized eggs per female that was proportional to the dose. The most sensitive parameter was the number of males produced per mictic female which was considerably higher in treated animals even at concentrations 10 times lower than the NOEC of asexual reproduction.

Effect of cadmium and zinc (separately and together) on the population growth of *Brachionus havanaensis* (Rotifera: Brachionidae)

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Among the heavy metal frequently used, cadmium and zinc are important. While Cd is a non-essential metal while Zn is necessary for all organsims. Both these metals occur together in nature. In this work we evaluated the impact of Cd and Zn separately and together on the population growth of the common Mexican brachionid rotifer, *B. havanaensis*. The concentrations of Cd used were 0, 0.05, 0.10, 0.21 and 0.42 and for Zn: 0, 0.07, 0.14, 0.28 and 0.57 mg L^{-1}. The LC_{50} 24h bioassay was 0.42±0.01 mg L^{-1} for Cd and 2.27±0.40 mg L^{-1} for Zn. In controls the peak population density was about 400 ind. ml^{-1}, while under heavy metal treatments, it was strongly reduced (5 ind. ml^{-1}). The rate of population increase varied from 0.01 (under heavy metal treatment) to 0.41 d^{-1} (in controls). The population of *B. havanaensis* failed grow when Cd concentration was 0.21 mg l^{-1} and Zn was 0.28 mg l^{-1}, but at other lower concentrations, there was a reduction in the rotifer population growth in relation to increase in heavy metal concentration, together or separately.

Species diversity of rotifers from different freshwater ecosystems of Kerala State (India), with comments on new records

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The present study is based on 314 plankton samples collected from different freshwater bodies, representing 110 localities in the Kerala State. This study revealed the occurrence of 108 species of Rotifera representing 16 families, and 30 genera. Of these, 64 are new records to the Kerala State, 40 are new records from South India and five species, viz. *Keratella tecta* (Gosse, 1851), *Lecane tenuiseta* Harring, 1914, *Macrochaetus danneli* Koste & Sheil, 1983, *Filinia camascela* Myers, 1938 and *Brachionus bidentatus* f. *inermis* (Rousselet, 1906) are reported first time from India. The family Lecanidae is represented by 30 species, Brachionidae by 22 species and Colurellidae by 13 species. Genus-wise Lecane, Brachionus and Lepadella were the most abundant groups.

Habitat choice in rotifer communities of three shallow lakes: impact of macrophyte substratum and season

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The distribution of rotifer community structure between emergent (*Typha angustifolia*) and submerged (*Chara tomentosa*) vegetation and comparatively open water zone was compared during the spring, summer and autumn seasons at three macrophyte-dominated lakes. Out of the total 107 rotifer species, 58% taxa was common for three examined lakes. Rotifer densities were positively related to macrophyte stem length, chlorophyll *a* and pH. Stoneworts due to their complex spatial and morphological structure, supported higher rotifer densities. The distribution of pelagic species did not differ between particular stations, which may have reflected the behaviour requirements of those rotifers. Some of them remained in the open water zone while some, looking for anti-predator refuge, gathered within macrophyte stands during the daytime. Moreover, there were 14 *Chara*-associated species and only 1 *Typha*-associated. The similarity of rotifer communities within the examined habitats was strongly influenced by particular habitats, and by lake or season to a lesser extent.

Studies on the cryopreservation of a freshwater rotifer, *Brachionus calyciflorus* Pallas

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Cryobiological studies on neonates and adults of *Brachionus calyciflorus* were attempted. After screening a variety of cryoprotectants, a protocol for the cryopreservation has been achieved using cryoprotectants like DMSO and methanol. There exists an associated depression in the freezing points of the rotifer on equilibration with these two cryoprotectants. Cryoprotection using 10% DMSO and 15% methanol was seen to promote various degrees of dehydration of the rotifer on external freezing compared to freezing in a freshwater medium. Low molecular weight cryoprotectants like methanol seem to permeate into embryos, neonates and adults more efficiently. Cryomicroscopic observations revealed variations in the occurrence and locations of internal freezing depending on the cryoprotectants used. The pattern of ice formation was different; with DMSO ice formation was unidirectional and blade-like, while with methanol the crystals were blunt posing less threat of the ice crystals penetrating the animal. Using 10% DMSO as a cryoprotectant and a cooling rate of -1°C min^{-1} permitted a recovery of 20% of viable amictic females when subjected to -30°C. At the same time, 15% methanol as a cryoprotectant under similar conditions gave 70% recovery of viable amictic females. The results suggest that a low molecular weight cryoprotectant like methanol, offers better cryoprotection than the widely used DMSO in cryopreserving *B. calyciflorus.*

Seasonal dynamics of the planktonic rotifer assemblage in the euphotic zone of a Mexican saline lake

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Saline lakes contain low numbers of planktonic rotifer species since salinity is an important limiting factor. The simplicity of the planktonic rotifer assemblage gives a good opportunity to understand the influence of the environment on seasonal variations in rotifer populations. Here we studied the composition and temporal fluctuation of the planktonic rotifer assemblage in the euphotic zone of the warm monomictic lake Alchichica, a deep (64 m) saline crater-lake in Mexico. Selected environmental variables, such as temperature, dissolved oxygen, chlorophyll *a* and food availability were also measured. A monthly sampling program was conducted during four years (1998-2001). Samples were taken in 5 depths along the euphotic zone (0-40 m) in a sampling point located in the center of the lake. Five liters were sampled from each depth using a Niskin sampler and filtered using a 63 µm net. Rotifers were identified and counted using a Sedgwick-Rafter counting chamber. Only two rotifer species were observed in the samples: *Brachionus rotundiformis* and *Hexarthra jenkinae*, both of which are typical to saline waters. In Alchichica they inhabited the upper part (5-20 m) of the euphotic layer. The higher mean densities were observed between January and March (when lake overturn occurred) or at the end of the stratification period (October-December). *B. rotundiformis* was observed in each of the four studied years but *H. jenkinae* was completely absent in 2000 and during the warm season (May-August) of the other years. The interannual dynamics of rotifer assemblages in the lake Alchichica showed strong differences. Food availability for rotifers, mainly expressed as picophytoplankton densities, seemed to be the principal factor governing the temporal fluctuations of planktonic rotifers in this lake.

Colonial life, oligotrophication and invertebrate predation: *Conochilus*, *Daphnia* and *Bythotrephes* in Lake Maggiore (Northern Italy)

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The oligotrophication of Lake Maggiore was accompanied by a decrease in *Daphnia* numbers due to the exponential increase in *Bythotrephes longimanus*, observed at stable population densities of *Letodora kindtii*, which lead to a general increase in invertebrate predation pressure. Colonial rotifers, namely *Conochilus* (*unicornis-hippocrepis* group) became an important component of Lake Maggiore pelagic food web, and were able to grow exponentially, and replace, along the seasons, the large filter feeder *Daphnia*. We report here the results of a study in which the long-term and the seasonal dynamics of *Conochilus* were analyzed with a distinction of the different organization levels exploited by these organisms, namely spherical colonies, conical fragments, as well as individuals, of which we recorded changes in abundance and size, in addition to those in the number of individuals/colony. An analysis of the changes we observed along the seasons and through the years, allows an understanding on the role of oligotrophication and invertebrate predation for the success of colonial life.

Effect of diazinon on life stages and resting egg hatchability of the rotifer *Brachionus plicatilis*

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The effects of organophosphate pesticide on life history parameters and hatchability of resting eggs of rotifer *Brachionus plicalitis* were assessed using diazinon. Ten newly hatched (<1h-old) neonates were individually cultured in five varying concentrations (0.1, 1.0, 2.5, 5.0 and 10.0 mg L^{-1}) of diazinon, and life history parameters such as time (h) the rotifers bear first egg and release first neonate, reproductive period, net reproductive rate, intrinsic rate of population increase, and lifespan were evaluated. Results showed that the time the rotifers took to release neonates was the most sensitive, giving the lowest EC_{50} value of 1.24 mg L^{-1}. The maternal females were further reared until they produced mictic daughters, and the number of offspring produced by amictic and mictic daughter was investigated. Result showed that rotifers exposed to 10.0 mg L^{-1} of diazinon produced significantly fewer amictic daughters, and at this concentration, rotifers did not produce any mictic daughter. At 5.0 mg L^{-1}, the number of male offspring was significantly lower than the control. Furthermore, the hatchability of the resting eggs produced by the rotifers when exposure was timed from birth until they produced resting eggs (early development), during late developmental stage of resting eggs (before diapause), as well as during diapausing stage was investigated. Rotifers exposed to 5.0 and 10.0 mg L^{-1} starting from birth, did not produce resting eggs, and at 1.0 and 2.5 mg L^{-1}, the hatchability of resting eggs was significantly reduced. 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 rotifers 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. This provides further evidence that the resting eggs should be produced under conditions of less environmental stress.

Water loss and morphological changes during desiccation in *Macrotrachela quadricornifera* (Rotifera, Bdelloidea)

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Bdelloid rotifers are aquatic microinvertebrates able to cope with the loss of environmental water and thus can live in temporal habitats. When water is evaporating, bdelloids contract into 'tuns', silence metabolism and lose water from the body. This condition is known as anhydrobiosis. Under controlled conditions, the bdelloid rotifer *Macrotrachela quadricornifera* was made anhydrobiotic and the variation of body volume, of metabolic activity and the adjustment of the internal structures were recorded and compared in hydrated and dry rotifers. Rotifers were observed by light (LM and CLSM) and electron microscopy (SEM and TEM), to know the body volume and the spatial distribution of the internal structures (muscles, tissues, cavities). The volume of the body was estimated comparing images at SEM and CLSM, and the loss of volume at desiccation was found to be about 50%. The dry condition of the rotifer is characterised by a very compact organization of its anatomy and also by condensation of the fine structure of its tissues. Images at LM and TEM did not show cavities in the dry rotifer body, while cavities were visible in the hydrated animal. We hypothesize that the volume loss is mostly due to the clearing of body cavities. We found also that the tun shape is attained by the contraction of longitudinal muscles, whereas the same muscles seem loose during anhydrobiosis suggesting that the tun shape is not maintained by muscle contraction. This agrees well with the apparent shutdown of the bdelloid metabolism during anhydrobiosis as observed in previous experiments.

Morphological and molecular data on *Brachionus* sp. 'Austria': preliminary results

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Brachionus plicatilis is a complex of cryptic species, clustered into three morphotypes (L Large, M Medium and S Small). Here we focus on biotypes within the L-morphotype, which have not yet been described morphologically and about which information exits from molecular studies. Failure to identify species boundaries in this complex may have a negative impact on studies of biodiversity, speciation and ecology. Furthermore, the coexistence of cryptic species may result in erroneous interpretation of ecological processes. This might be the case for *Brachionus* sp. 'Austria' and *Brachionus plicatilis* sensu stricto both belonging to the *B. plicatilis* species complex. We have isolated individuals from wild populations of *B*. sp. 'Austria' (Lake Koronia, Greece; Tianjin Province, China). Cultured populations of *B. plicatilis* s.s. were used as reference material. The genetic identification of the samples was performed using the method of Restriction Fragment Length Polymorphism (RFLP) and also sequencing of COI and 16S rRNA mitochondrial regions. In particular, selected endonucleases (*Ban*II & *Ava*II for COI and *Dra*I & *Taq*^{a}I for 16S rRNA) and sequencing data were used to identify the *B. plicatilis* s.s. species and the *B*. sp. 'Austria' biotype. Nine lorica measurements were taken to determine characters that would permit the morphological separation of these species. Discriminant analysis performed on log-transformed data showed that variables describing lorica shape (i/a: head aperture/lorica length) and the shape of the anterior spines (d/e: distance between central spines/dorsal sinus depth, g/f: medial spine length/distance between central and medial spines) were the main discriminating characters. [Part of this work was funded by a European Union project (ROTIGEN, Q5RS-2002-01302).]

The future of rotifer taxonomy: Genetics or Morphometrics? A cautionary tale from the *Brachionus plicatilis* species complex

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The importance of correct taxonomic classification cannot be understated, for the reductionism of science allowing us to characterize and hence investigate a small part of nature underpins the cognitive process by which we come to an understanding of how nature works as a whole. With the rise of molecular genetics we have a powerful tool with which to classify our world, a tool that is after all morphology but on a much finer scale. With the popularization of PCR and DNA sequencing techniques an impassioned debate has arisen over the use of genetic markers to "barcode life". Proponents of this technique advocate at one extreme the analysis of a single mitochondrial gene, COI, as the basis of a DNA identification system for animals. This suggestion has lead to staunch criticism most fervently voiced by the practitioners of classical taxonomy namely that DNA barcoding is complementary to, not a replacement for, traditional taxonomy. To inform this debate I investigated four members of the *B. plicatilis* "cryptic" species complex, including two species from the southwest of Western Australia that are new to science. True cryptic species exhibit hidden species diversity having their taxonomic identities masked by a significant degree of morphological stasis. With the aid of genetic markers, COI and ITS1, and a more traditional morphological approach species have been identified both within and between lineages with concordance between the two methods. This suggests that the *B. plicatilis* species complex may not be "cryptic" after all. To date only 3 of the 15 genetically identified species have been classified. While genetic technologies may one day hold the key to the rapid identification of species in the field facilitating large scale ecological studies, I suggest that it would be prudent to first describe what these species look like.

Where on earth is the *Brachionus plicatilis* species complex?

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A review of 138 papers indicated 307 sites from around the globe as the habitat of the *B. plicatilis* species complex. Members of the *B. plicatilis* species complex are found in estuaries, lagoons, saline lakes and coastal waters with no known records of these organisms occurring in the open ocean away from the coast. They have a cosmopolitan distribution with members being found as far north as Yukon Territory, Canada (≈ 62.8º N) and as far south as Lake Victoria in Christchurch, New Zealand (≈ 43.5º S). The salinity range over which *B. plicatilis* species complex members were found extended from 0.05 to 310 ‰. Such extremes were rare, however, and questionable, with a more probable range of 1-20 ‰ implicated. While members of the species complex are found in acidic waters, on the whole they inhabit saline waters with a pH of greater than 7, mostly likely attributable to the underlying chemistry of saline lakes which are generally alkaline. The distribution of the species complex is truly global, with some members having a cosmopolitan distribution. However, there is evidence to suggest that some members of the species complex form isolated populations, for example, *B. spatiosus*, *B. magadiensis* and *B. colongulaciensis*. Further investigation is needed to clarify this point. This review is accompanied by the original species illustrations indicating type localities. Phylogenetic studies suggest that the species complex may contain 15 species; the reviewed taxonomic literature indicates the presence of at least 13 species, more if miscellaneous descriptions are included. By mapping out the global distribution of this species complex and reviewing its taxonomy a valuable tool has been forged for the selection of specimens to enhance our understanding of its phylogeny and biogeography.

Fish and nutrient effects on rotifers in a shallow lake. A mesocosm experiment

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A mesocosm experiment was conducted "in situ" in a *Chara* dominated shallow lake near Valencia (Spain) to study top-down and bottom-up effects on rotifers. Both processes were important in determining rotifer abundance, biomass and diversity. A total of 36 mesocoms was established with treatment combinations of three fish levels (from no fish to 45 individuals of *Gambusia holbrooki* males) and 4 nutrient enrichment levels (from no additions to 10 mg L^{-1} nitrate-N and mg L^{-1} phosphate-P) and three replicates. The main effect was a notable increase of planktonic and plant-associated rotifers with fish density. This was due to the removal of crustaceans by the zooplanktivorous mosquitofish, which caused an immediate elimination of cladocerans and drastic reductions of predatory cyclopoids in the mesocosms. Planktonic and plant-associated rotifers increased with nutrients in the mesocosms with the highest fish densities, but not in the absence of fish or at the low fish densities. This was explained by the reduction of predatory pressure of copepods on rotifers when fish was controlling cyclopoid abundances. The results showed a marked negative relationship between rotifer and cyclopoid abundances. In the absence of fish or low fish level, predatory copepods increased in the mesocosms with high nutrient loadings, specially at the second level of enrichment (5 mg L^{-1} N; 0.5 mg L^{-1} P) and prevented rotifer increase. The effects of nutrients and mosquitofish were also analysed for each of the rotifer species inhabiting the lake. High level nutrient additions induced a switch to a turbid state with macrophyte disappearance. Abundance of plant-associated as well as planktonic rotifers diminished when the turbid state was well established. The changes in rotifer species composition after the switch from a clear to a turbid water state were also described.

Testing the monopolization hypothesis in rotifer populations from a salt lake basin

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Continental zooplankton are thought to have high dispersal capacity trough their diapausing eggs. However, according to the results of molecular studies, zooplankton populations, even at a local scale, are highly genetically differentiated. The monopolization hypothesis (MH) intents to explain this paradox, proposing that the high population growth rates, the large diapausing egg banks and a rapid process of local adaptation result in a persistent founder effect and a monopolization of the resources by a small number of colonizing genotypes. Some studies in rotifers and *Daphnia* have found a correlation between genetic differentiation and geographic distance and this seemed to counteract the MH, as this correlation is usually attributed to genetic-drift equilibrium populations following the isolation by distance model. However, recent studies have shown that such correlation can arise from the history of population colonization due to a process of serial founder effects, and this would fit with the persistent founder effects expected under the MH. Here we tested the occurrence of serial founder events in rotifer populations by reconstructing the history of neighboring populations. We made a large sampling effort in a set of 14 salt lakes in a small endorheic area in Eastern Spain. From the pond sediment samples, we isolated about 2000 diapausing eggs of *Brachionus plicatilis*. From each egg, we were able to amplify and analyse mitochondrial genes. In order to process our large amount of samples, we optimized a high throughput technique, single strain conformation polymorphism (SSCP). This technique reduced sequencing effort by identifying identical genes in different eggs. Preliminary results of the project have been presented.

Combined effects of temperature and heavy metal (Pb) on the population growth of the rotifers *Brachionus havanaensis* and *Brachionus rubens*

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Among the brachionid rotifers, *B. havanaensis* and *B. rubens* are frequently found in many freshwater bodies in Mexico. Lead is one of the non-essential heavy metals with high industrial applications. In this study, we evaluated the combined effects of temperature (22 and 32°C) and the concentration of Pb (0, 0.05, 0.1 and 0.2 mg L^{-1}) on the population growth of the two rotifer species. For the experiments we used one algal food density ($1X10^6$ cells ml^{-1} of *Chlorella vulgaris*). For each treatment we maintained 3 replicates and the duration of growth experiments was 16 days. Regardless of species, increase in Pb concentration resulted in decreased population growth of both the rotifers. Regardless of toxicant concentration and the rotifer species, higher temperature accelerated the population growth. Regardless of Pb concentration and the temperature level, *B. havanaensis* was always numerically more abundant than *B. rubens*. In controls *B. havanaensis* reached peak densities of about 225 ind. ml^{-1}. At comparable conditions, *B. rubens* reached less than half of this density. Rate of population increase (*r*) and the peak population densities of both the rotifer species were significantly affected by the metal concentration in the medium.

Seasonal and depth-related zooplankton distribution in the reservoir Valle de Bravo (State of Mexico, Mexico) with emphasis on rotifers

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Valle de Bravo is one of largest drinking water reservoirs serving nearly 12% of Mexico City's 20 million population. However, due to anthropogenic eutrophication, the water quality has steadily been deteriorating. In an effort towards a better management, we studied the monthly variations in the zooplankton density and diversity for one year (November 2004 to October 2005) at 5 different depths (2, 4, 8, 12 and 20 m) at 5 different sites of the reservoir. The zooplankton mostly composed of rotifers particularly, *Keratella cochlearis, Polyarthra vulgaris, Trichocerca similis, T. capucina* and *Ascomorpha ovalis*. *Kellicottia bostoniensis* was occasionally dominant at lower (4-20 m) depths. The diversity of both cladocerans and copepods was lower. Occasional blooms of *Daphnia laevis* and *Chydorus sphaericus* were observed; *Bosmina longirostris* was consistently present in almost all the sites and throughout the study period. The data have been discussed with emphasis on effective management of the reservoir.

Combined effects of temperature, food (*Chlorella vulgaris*) concentration and predation (*Asplanchna girodi*) on the morphology of *Brachionus havanaensis* (Rotifera)

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The combined effect of temperature, food level and the presence of a predator on the body size of the brachionid rotifer *B. havanaensis* was tested in this study. *B. havanaensis* was cultured at 15, 20 and 25°C under 3 different *Chlorella* levels ($05X10^6$, $1.0X10^6$ and $2.0X10^6$ cells ml^{-1}) in the presence and in the absence of *A. girodi*. For each treatment we maintained 3 replicates and constant (0.4 ind. ml^{-1}) population density of *B. havanaensis*. In treatments containing *Asplanchna*, the predator was separated from the prey by a mesh (pore size 50 µm). On the last day of the experiment, a portion of the *B. havanaensis* population was sampled for morphometric measurements (adult lorica length, width, posterior spine length, and the egg volume). Size measurements were done by drawing the specimens using calibrated camera lucida. Statistically significant impact of temperature, food concentration and the presence of predator on the lorica length and the spine length of *B. havanaensis* was observed. Egg volume was also influenced by these factors. Regardless of the treatments, there was a direct positive correlation between lorica length and width. However, spine length varied depending on the treatment. Egg volume was linearly related to the adult size. Notably long posterior spines were observed in treatments containing the presence of *A. girodi*.

Influence of vertebrate and invertebrate infochemicals on the population dynamics and epizoic tendency of *Brachionus rubens* (Ehrenberg) (Rotifera: Brachionidae)

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Brachionus rubens is often epizoic on large cladocerans. While this behaviour confers advantages in the presence of invertebrate predators, it is detrimental in the presence of vertebrate predators. The aim of this study was to determine whether the epizoic tendency of *B. rubens* varies in the presence of infochemicals from veterbrates and invertebrates. For the invertebrate predators we used *Asplanchna brightwelli* (Rotifera) and *Megacyclops* sp. (Copepoda) and for the vertebrate predator, the zebra fish (*Danio rerio*). We studied the population dynamics, morphometry and egg ratio of *B. rubens* in the medium in which the predators were maintained for 24h and this was compared to controls (water without infochemicals). Experiments were conducted using 50 ml of test medium containing the algal food *Chlorella vulgaris* at a density of $1X10^6$ cells ml^{-1}. We introduced 5 individuals of *Daphnia pulex* into each container in order to quantify the number of epizoic individuals of *B. rubens*. In all there were 20 test vessels (5 treatments by 4 replicates). Experiments were conducted at 23±1ºC and daily population growth data were collected for 15 days. The results showed that *B. rubens* suffered considerable exploitative competition from *Daphnia pulex*. Growth rates however did not vary significantly due to the presence of vertebrate or invertebrate infochemicals. In the presence of *Daphnia* more than 68% of *B. rubens* population was epizoic. The data have been discussed with special emphasis on the protective value of epizoic behaviour against predation pressure in nature.

Exocytotic membrane docking proteins in *Brachionus calyciflorus* (Rotifera: Monogononta)

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We performed the determination of exocytotic membrane docking proteins SNAP-25, SNAP-23, Syntaxin-1, and Syntaxin-4 in the freshwater rotifer *Brachionus calyciflorus*, which are involved in the exocytosis of neurotransmitters like acethylcholine. The presence of these proteins was determined by immunohistochemistry and immunoblot analysis using antibodies against SNAP-25, SNAP-23, Syntaxin-1, and Syntaxin-4. The immuno-staining techniques on these organisms showed that the antibodies located a conspicuous region at the neuromuscular junctions of the rotifer.

Density dynamics of *Notholca squamula salina* Focke in Antarctic freshwater Lake Wujka (South Shetlands, King George Island, Polish Antarctic Arctowski Station)

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Rotifers were collected during the XXVIII Antarctic Expedition in 2003-2004 organized by Polish Academy of Sciences to the Henryk Arctowski Antarctic Station located at King George Island (Southern Shetlands). Rotifers inhabiting Lake Wujka, one of the largest freshwater basins on the terrain of the Polish Antarctic Station, were investigated. This lake, located near the sea, is shallow (maximum depth, 138 cm). Rotifer were sampled at three-day intervals from December 2003 to June 2004 at two different locations SP1 and SP3. The physicochemical parameters (pH, temperature, salinity, conductivity, TDS - total dissolved solids) of the water were also studied to assess their impact on the density dynamics of *Notholca squamula salina* in the unstable Antarctic aquatic habitat. The maximum density (114 ind. L^{-1}) of *N. squamula salina* was noted in June at SP1, but the two highest density peaks were noted in January (80 ind. L^{-1}) and May (150 ind. L^{-1}) at SP 3. Spearman's nonparametric correlation indicates that TDS (r_s = -0,415; p = 0.004) and pH (r_s = -0,314, p = 0.021) at SP1 and temperature (r_s = -0.437, p = 0.001), salinity (r_s = 0,442; p = 0.001) and pH (r_s = -0.506, p < 0.001) at SP3 had significant influence on the density dynamics of *N. squamula salina* in Lake Wujka.

Bacterivory by brachionid rotifers: implications for wastewater management

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Several rotifer species are efficient bacterivores. Since bacterial contamination in natural waterbodies as well as in wastewater treatment plants is high, we considered it important to compare the efficacy of common rotifer taxa in reducing bacterial loads. *Vibrio cholerae* is one of the most common bacteria in wastewaters in Mexico. We isolated this from the channels of Xochimilco and cultured on TSBS medium. We compared the efficacy of *Brachionus calyciflorus*, *B. patulus*, *B. rubens* and *B. havanaensis* in reducing the bacterial load from wastewaters. We also studied the functional response of these rotifer species on *V. cholerae*. We found that *Brachionus calyciflorus*, *B. patulus* and *B. rubens* could reduce the total bacterial densities by 40% in 30 minutes but *B. havanaensis* by only 15 %. The data have been discussed with reference to the use of rotifers in reducing bacterial densities in wastewater treatment plants.

Combined effects of heavy metal (Hg) concentration and algal (*Chlorella vulgaris*) food density on the population growth rate of *Brachionus calyciflorus* (Rotifera: Brachionidae)

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The genus *Brachionus*, especially *Brachionus calyciflorus*, has been extensively used as bioassay organism in aquatic toxicology. Among heavy metals, mercury is highly toxic to zooplankton even in very low concentrations. In this study we evaluated the combined effects of two food levels ($0.5X10^6$ and $1.5X10^6$) and five concentrations (0, 0.000625, 0.00125, 0.0025, 0.005 mg L^{-1} $HgCl_2$) on the population growth of *B. calyciflorus*. The growth experiments were conducted for two weeks at 23±1°C under continuous fluorescent illumination. For each food level – heavy metal combination, we maintained 3 replicates. Our data showed that regardless of food level, increase in the heavy metal concentration in the medium resulted in decreased population growth of *B. calyciflorus*. At any given heavy metal concentration, *B. calyciflorus* fed higher food levels had higher population abundance. The rate of population increase was significantly influenced by both the heavy metal concentration and the algal level. The results of this study were discussed in relation to the protective role of algal density against heavy metal toxicity.

Ecological differentiation of two sympatric species of the L-morphotype in the *Brachionus plicatilis* complex

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Sibling or cryptic species (i.e., with little or no morphological divergence) have proven to be widespread in the zooplankton, sometimes involving sympatry. Examination of genetic and ecological relationships within cryptic species complexes can provide insights into species diversity and speciation processes. The taxon *Brachionus plicatilis* (Monogononta; Rotifera) has been recently recognised as an ancient cryptic species complex with three major, deeply diverged clades. *Brachionus plicatilis*, *sensu stricto*, and *B. ´Manjavacas´* are two, closely related species in the complex, which belong to the L- (large) morphotype often co-occur in the same ponds in the Iberian Peninsula. Despite the fact that they are clearly genetically differentiated, multivariate morphometry showed only minor but highly overlapped morphology differentiation. The purpose of this study was to elucidate whether ecological differentiation exists between these two cryptic rotifer species. A factorial experiment was designed to test for inter- and intra-specific differences in population growth rates at different salinity levels, representing the range over which the L-morphotype is found in its natural environment. Although the results did not show different optimal salinity values for species, *B.´Manjavacas´* grew better at high salinity than *B. plicatilis*, suggesting that seasonal variation in salinity plays a role in stabilizing sympatry. Our results showed also within-species variation in salinity response. These results also suggest that within-population variation in *B.* '*Manjavacas*' and in *B. plicatilis*, s.s., are correlated when co-occurring populations are compared.

Feeding and reproduction in the rotifer *Asplanchnopus hyalinus*

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Relative to that on *Asplanchna*, published information on the biology of the predatory and predominantly periphytonic rotifer *Asplanchnopus* is limited. I present here some aspects of the feeding and reproduction in *Asplanchnopus hyalinus*, based laboratory studies. The rotifer was originally isolated from zooplankton collected from the backwaters of Yamuna River in Delhi. Cloned cultures of *A. hyalinus* were maintained on brachionid rotifers and chydorid cladocerans. *A. hyalinus* is a voracious eater, an adult female being able to consume upto 20 individuals of *B. rubens* or up to 4 *Chydorus* in a 30-minute period. Prey handling times were recorded in relation to predator-prey size ratios. For a given ratio, handling times with *Chydorus* were lower than with *Brachionus calyciflorus*. At high food levels, the female started reproducing within 30 h after hatching and produced 19 ± 4 amictic eggs in her lifetime (3-6 days). The developmental time for amictic eggs was 24.3 ± 0.8 h at 20°C and 17.3±1.0 h at 25°C. Sexual reproduction was frequent in laboratory cultures, but the factors triggering a switch to mictic reproduction were not identified. A single male could mate as many as 4 times with a female, the duration of each mating being 62.4 ± 16.1s. Mated female produced 4.5 ± 1.0 resting eggs. Efforts to hatch resting eggs of different ages were unsuccessful. The range of population growth rates (0.3-1.3 d^{-1}) achieved by *A. hyalinus* was a function of the type and concentration of prey offered.

Rotifer research in India: An overview

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The generally made claim that India has the third largest scientific manpower in the world is not reflected in the proportional number of rotifer researchers in the country. The total number of rotifer species recorded from India so far (350+), is substantially lower than the number one would expect from a country with a large geographical area and high habitat diversity. It reflects, besides inadequate exploration, a scarcity of taxonomic expertise, particularly for the bdelloid group. Search of the data bases BIOSIS and Aquatic Sciences and Fisheries Abstracts (ASFA) for Indian publications on Rotifera reveals the following trends: 1. Approximately 37% of the contributions are on routine limnological-zooplankton studies in which the species composition and abundance of rotifers are recorded along with those of the other taxonomic groups. 2. A large proportion of papers (~44%) concern rotifer species records, range extensions, and seasonal variation in relative abundances, and many of them highlight the importance of rotifers as bioindicators of water quality. 4. Aquaculture-related papers on rotifers (<5%) and those on experimental studies (laboratory and field) (~9%) are limited, but the latter address some interesting questions in population growth, life history strategies and predation. 5. Conspicuously missing are specific studies on rotifer species interactions and their role in the structure and organization of food webs in tropical and subtropical aquatic ecosystems typical of India. General problems faced by limnological research laboratories in India in conducting high-impact research on rotifers are discussed.

Differences in population growth rates, morphometry and *Cox1* sequence among strains of *Brachionus calyciflorus* from Mexico City (Mexico)

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Obvious variations in the body size of *B. calyciflorus* collected in different waterbodies in Mexico City prompted us to investigate other morphological, physiological and ecological differences. Strains of *B. calyciflorus* were obtained from Tezozomoc pond, Chapultepec Lake and Xochimilco Lake. Cultures under similar test conditions were developed for the three strains at 23°C, under continuous and diffused fluorescent illumination and daily fed with the green alga *Chlorella vulgaris* at a density of $1X10^6$ cells ml^{-1}. At the end of test period, samples (20-40 egg-bearing individuals) from each strain were used for morphometric measurements and the rest of the population for isolating the total genomic DNA. The DNA samples were then used to amplify *Cox1* coding sequences using specific oligonucleotide primers. After purification of the PCR products, DNA fragments were sequenced with an ABI PRISM 310 Genetic Analyzer, using dye terminator sequencing kits with two M13 universal primers. Individuals in one of the three strains were up to 42% smaller than others (mean body length: 180 vs 270 µm), and showed growth rates (*r*) close to 0.5 d^{-1}. An attempt was made to interpret these ecological and morphometric data based on the molecular analysis.

Novel approaches to an old technique: Rotifer specimens mounted as permanent slides and digital photomicrography using image stacks

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Rotifer research has traditionally been plagued by a lack of type material. The literature on rotifer taxonomy is scattered with species, subspecies and varieties, whose validity is doubtful from a phylogenetic point of view. Many rotifers have been found only once and our knowledge about them is based on often inadequate descriptions. If type material was available, both re-examination of species already known and the identification of species new to science would be considerably facilitated. Type material, however, presupposes properly narcotised and fixed specimens that can be mounted as permanent slides. Here we present a reliable and uncomplicated way of permanently mounting narcotised and fixed rotifer specimens. Prior to fixation, rotifer specimens are narcotised and rapidly killed off using Osmiumtetroxide (1% OsO_4). After fixation, specimens are washed in H_2O_{bidest} and transferred to a 1:10 glycerol / water solution. The water is allowed to evaporate for a few days. The specimens are subsequently mounted as permanent slides embedded in glycerol, surrounded by a mixture of paraffin and beeswax and sealed off with a sealing agent. Computer programmes (e.g. Helicon Focus® or CombineZ) allow stacking of digital images, that is of adding up different focal planes. Based on this technique, it is possible to overcome the problem of limited focal depth inherent to light - microscopy. Examples of the potential of this technique are given.

A quick and reliable method for obtaining fully extended rotifer specimens fixed in a life-like state

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One of the most characteristic features of rotifers is a complex ciliary apparatus in the head region, the rotatory organ, from which the whole taxon derives its name. This ciliary apparatus is a potentially valuable character in reconstructing rotifer phylogeny. Unfortunately, most rotifers are sensitive towards any kind of disturbance and immediately react by retracting the corona. This problem is often particularly acute with soft-bodied, bottom dwelling species such as many Dicranophoridae and Notommatidae. In some species of the latter group, lateral ciliary tufts (auricles) are a characteristic feature displayed only by undisturbed individuals. As soon as fixatives or inappropriate narcotics are added to the sample, the animals predictably react by a complete withdrawal of the rotatory organ. Here we present a quick and reliable strategy to overcome this problem. In a two-step procedure prior to fixation, it is possible to obtain fully extended specimens. First, individual specimens are isolated from the sample under a stereomicroscope using finely drawn capillary pipettes. Next, soda water is added very carefully again using a very fine capillary pipette. In case the specimen should have retracted, the water enriched with carbolic acid can be carefully removed and the process repeated. At some point, the animal will be fully extended and the motion of the cilia (visible under the stereomicroscope at higher magnifications, preferably darkfield) have ceased. Now a very little drop of 1% Osmiumtetroxide (OsO_4) is to be added immediately killing off the specimen in a life-like state. OsO_4 has the additional benefit of staining the inner organs which is particularly useful if permanent slides are to be prepared. Finally, the specimen is washed in H_2O_{bidest} and fixative solution (PAF, 240 mOsm) is added. Specimens fixed in this way can now be used either for SEM or permanently mounted on slides. Examples of both are given.

Life history responses of a rotifer (*Platious patulus*) mixtures of arsenic and heavy metals

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While many studies have determined toxicity of single heavy metals few studies have addressed the combined effects of multiple elements. In order to estimate how metal mixtures affect population dynamics of freshwater rotifers, we exposed *Plationus patulus* to environmentally relevant combinations and concentrations of arsenic and five heavy metals (Cr, Cu, Ni, Pb and Zn) for 7 days. Life history parameters were used as end-points. Cu affected population growth and mortality, while Ni affected rotifer reproductive mode and seemed to be a teratogenic agent. Cr affected both reproductive mode and population growth. Population growth was negatively affected by Cr. Lead also negatively affected population growth parameters (intrinsic rate of increase (*r*), reproductive cumulative ratio, RCR). Zn decreases resting egg production and population growth (RCR). The observed effects of multi-elements interactions were more diverse than those produced by individual metals. The interaction of the six elements at different concentrations caused a decrease of 62-92% in *r* as the number of elements present at their highest concentration increased. RCR and the number of diapausing eggs also decreased under these conditions while mortality increased. Responses of this basal consumer to heavy metal contamination would provide insights into how aquatic food webs and their constituent organisms can be altered by anthropogenic inputs such as metal mixtures in aquatic ecosystems.

Characterization of Peruvian strains of eurihaline *Brachionus* species used in aquaculture

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Rotifers of *Brachionus plicatili*s species complex have been used in Peru as live food for the first larvae stages of *Paralichthys adspersus* (fine flounder) and *Cryphiops caementarius* (prawn river of Peru). The culture of *B. plicatilis* is done using the batch technique, obtaining densities of 60 – 120 ind. ml^{-1}, by feeding on *Tetraselmis suecica*, *Nannochloris maculata* and *Nannochloropsis oculata*. However, these cultures are unstable and unpredictable. The rotifer strains used in these cultures have been isolated from several biotopes of Peru (Puerto Viejo, Chilca and Puerto Chicama) or have been introduced from Japan and Mexico. Nevertheless the taxonomical identities of the native strains have not been studied and hence have limited the standardization of mass culture techniques. Morphometric characterization of a native strain (Bocapan) of *Brachionus* is being made and strains of *B. plicatilis* (LI) and *B. rotundiformis* (SS2) from the Valencia University (Spain) are used as a reference patron. Preliminary molecular analysis based in the mitochondrial COI gene indicated that 3 of Peruvian strains (Chilca, Puerto Viejo, Ventanilla) are grouped in the same cluster with *Brachionus ibericus* while Agraria University strain is grouped with *B. plicatilis sensu stricto*. In order to allow further characterizations, SS2, L1, Bocapan and other strains isolated from Chilca, Ventanilla, Puerto Viejo and two commercial strains from Agraria University (all located in Peru) and Baja California University (Mexico) are kept in the national rotifer stock-culture in the Peruvian Marine Institute (IMARPE).

Determination of lead in zooplankton, water and sediments from El Niágara reservoir in Aguascalientes (Mexico)

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The city of Aguascalientes with a population of 700,000 inhabitants and more than 500 industries is located in Central Mexico. The drinking water system consists primarily of wells. Agricultural, domestic and industrial wastewater is dumped at the San Pedro River which eventually ends up at El Niágara reservoir. Our study consisted of 6 collections from February 1999 to February 2000 to determine the levels of lead in water, sediments and zooplankton samples collected from this reservoir. The highest concentration of lead was found in the sediments (mean = 5.07 ppm). However, this value does not exceed the permitted levels for agricultural soils. In contrast the mean values (15.57 ppb) of lead in the water column exceed the limits for drinking water. We found evidence for bioconcentration, and perhaps biomagnification, of lead in different trophic levels of the zooplankton. We determined lead in two species of herviborous cladocerans; *Moina affinis*, and *Daphnia similis*. We also determined lead levels for two predator species; the rotifer *Asplanchna brightwelli* and dipteran larvae of the genus *Culex*. Our data represent the first report of a bioconcentration factor (BF = 22,000) for rotifers.

Do we have sex or not?: Ecological implications of subitaneous eggs of *Brachionus* passing trough fish gut in a deep tropical lake

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Diapause is a life-history trait that is found in the life cycle of monogonont rotifers. Up to now, several biotic factors have been proved to induce sexual reproduction in rotifers, including resource quality and quantity and chemical exudates by conspecifics, competitors, or even predators. We analyzed a rotifer belonging to the *Brachionus plicatilis* species complex inhabiting a karstic, monomictic, hyposaline and deep lake, Alchichica (Central Mexico). Previous results indicated that conspecific crowding chemicals induced sexual reproduction in this species, and that different strains have significant differences in response to resource limitation, some have high mixis ratios when resources became scarce while others not. We studied here for a non-mixis-inducing clone the effect of two combined factors: a) resource limitation and, b) fish-predator chemicals. Our results confirmed previous findings; this clone had a positive response in mixis induction related to food concentration. However, fish kairomones failed to induce mixis, although we observed differences between the first and the second generation in this parameter. Considering the relatively low mixis induction by both resource limitation and fish, we evaluated the effect of direct predation on subitaneous eggs. Unexpectedly, over 90% of *Brachionus* eggs that passed through the fish gut were morphologically intact in fish feces, and almost all of them released healthy rotifer neonates. We discuss our results considering, a) high variation within a single population, b) the cost of resting egg production in a deep waterbody where emergence cues are not easily attained, and c) a relatively reduced resting egg bank.

An Analisis of the factors Influencing the presence of males in the freshwater rotifer *Lecane quadridentata* (Rotifera: Monogononta)

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We studied several factors (light:dark cycle, food concentration, temperature and strain type and their combinations) influencing the presence / absence of males in two strains of *L. quadridentata* for 7 days. Statistical analsysis showed that the mean female density was significantly affected by all factors and their combinations. However, male density was only affected by photoperiod and strain type, and their combination, the combination of food concentration, photoperiod and strain type and the combination of all factors. Males were present in only one strain type during the treatments, despite being previously observed in cultures of both strains. After 7-d period, and from the 5 initial neonates, the cultures in the highest food concentration reached mean female densities of up to 414 females ml^{-1}. In contrast in the best cultures males only reached mean density values of 1.6 males ml^{-1}. These results and the factors affecting presence of males in other monogonont rotifer species are discussed.

Small prey size is an effective deterrent against predation: a case study on two species of *Asplanchna* and three brachionid prey

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Many species of *Asplanchna* are predatory feeding on prey of different body sizes. Brachionid rotifers armed with strong posterior spines are usually large (>180 µm) and therefore resist *Asplanchna* predation. Extremely small (<80 µm) rotifers such as *Anuraeopsis* may also escape from predation, particularly at the densities in which they are found in nature. Here, we tested this hypothesis with two predator (smaller *Asplanchna brightwelli*: 500 µm and larger *A. sieboldi*: 800 µm) and three prey rotifer species (smaller: *Anuraeopsis fissa* (70 µm); larger: *Brachionus calyciflorus* (200 µm) and intermediate: *B. patulus* (120 µm)) using population growth, functional response, life table and prey preference studies. For both the predator species, best survivorship and reproduction related performance was observed on *B. calyciflorus*, while those raised on *A. fissa* showed lower survival rates and population growth rates. *Brachionus* were also consumed in greater numbers than *Anuraeopsis* by both the predators. Similar trends were observed in the prey preference studies. Our study shows that small species, by virtue of their size alone, can effectively escape predation, especially under low densities.

Cryptic speciation in the cosmopolitan *Epiphanes senta* complex (Monogononta, Rotifera)

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Many rotifers are considered to be cosmopolitan species. One of these species is *Epiphanes senta* which has been reported from all continents except Africa and Antarctica. Morphology and mating behavior were studied in populations in a tropical alpine lake on Mauna Kea (Hawaii), in temporary rock pools of the Chihuahuan Desert (Texas) and in floodplain habitats of the Oder River (Germany). Morphological differences among these populations are restricted to the surface structure of resting eggs. Males of the European and the Chihuahuan desert populations display a unique mate guarding behavior which is not found in the Hawaiian population: males attend female eggs and mate with the hatching female. In mate choice experiments males of the floodplain population differentiated between female eggs from their own and from other populations, attending almost exclusively those from their own population (99.9% of their mate guarding behavior displayed in 20 tests was directed towards eggs of their own population). Males of the Chihuahuan desert population made no distinction among eggs from different populations. They spent 53% of the time attending eggs from their own population and 47% of the time attending eggs from another population. In cross mating experiments Chihuahuan desert males mated with hatching females from the floodplain population. However, juvenile mortality among these fertilized females reached 41% and was significantly higher than juvenile mortality after intrapopulational mating (2%). Surviving females did not produce viable resting eggs. Hawaiian males, although not attending eggs, readily mated with newborn females from the Chihuahuan desert which then produced viable offspring. Partial reproductive isolation among the studied populations demonstrates that the cosmopolitan *Epiphanes senta* is actually a species complex.

A Global Assessment of rotifer diversity in continental waters

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There is a need to monitor the status and trends of freshwater biodiversity in order to quantify anthropogenic impacts on freshwater systems and to improve sustainable management of freshwater biodiversity. Current projects carrying assessment of freshwater biodiversity focus mainly on leading- better-known groups such as fish, or identify keystone species and/or endemic freshwater systems for conservation purposes. The purpose of the project "A global assessment of Animal Diversity in Continental Waters", which is an extension of a previous preliminary literature study is to complete these existing projects by providing quantitative estimates of species numbers for all freshwater groups on each continent and/or major biogeographical regions. Here I present a preliminary overview of the results obtained for Rotifera within the framework of the project.

High rotifer diversity in a Laotian rice paddy and adjacent pond: is there potential for the conservation of threatened freshwater biodiversity?

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During a short visit of Northern Laos in August 1999, we sampled a number of aquatic habitats in order to increase the Rotifera record for that country. To our surprise, two samples collected in an otherwise nondescript rice paddy and adjacent roadside pond near Vientiane yielded an astonishingly rich diversity of Rotifera: over 134 species including at least 5 new species and several globally rare ones were found, albeit all in low numbers. A rotifer fauna as diverse as observed here is unrecorded for such habitats, even comparing with the abundant information on the rotifers of systems of the same nature and origin in the Thai part of the floodplain of River Mekong and tributaries. Differences in agricultural practices between Laos and Thailand may offer a possible hypothesis accounting for the difference in species richness: whereas rice culture in Thailand is intensive, with abundant use of fertilizers and pesticides, this is probably much less the case in the economically challenged Laotian rice culture. Similarly diverse rotifer faunas are known to occur in natural floodplain systems only. This, however, is less surprising if rice fields are seen as the artificially flooded, hence floodplain, habitats they essentially are. Our observation offers potential for the conservation of freshwater biodiversity, if confirmed by more detailed research. In this context, it may be rewarding to study the effects on rotifer diversity of fertilizer and pesticide use, and to examine the impact of organic farming.

Transfection and reporter gene expression in *Brachionus*

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Transfection transfers foreign genes into a host to investigate their expression. This method is critical in functional genomics to probe phenotypes using reporter genes, knockouts, and overexpression studies. We describe how this powerful tool can be applied to rotifers, surveying a variety of transfection methods, including lipid and polyamine chemical reagents, and electroporation of rotifers and resting eggs. Electroporation proved most reliable and was used to transfect a plasmid containing a green fluorescent protein (GFP) gene into resting eggs of *Brachionus plicatilis*. Hatchlings expressed this gene and fluorescence was quantified in tissues by epiflourescence microscopy. Transfection efficiency was relatively high, with more than 80% of hatchlings expressing GFP. The time course of gene expression, as well as the expression intensity and tissue distribution have been described.

One new species of *Brachionus* from Aguascalientes (Mexico)

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In Aguascalientes State (Mexico), we collected a new species of *Brachionus*. It is a large loricate brachionid rotifer, closely related to *Brachionus bidentatus* Anderson, 1889. The presence of two strong postero-lateral spines on the dorsal lorica distinguishes it from *B. bidentatus*.

Comparison of the predation rates of *Paramecium multimicronucleatum and Rotaria rotatoria* under different enterobacterial densities

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Pathogenic microorganisms are commonly present in the domestic wastewaters and they can be removed by disinfections process. Chlorination is the most effective and economic disinfection method used for enteric bacteria elimination. However, different dangerous by-products are generated during this process. Biological disinfection using bacterivores microorganisms is an ecological alternative. The aim of the present work was to measure and compare the predation rate between *Paramecium multimicronucleatum* and *Rotaria rotatoria* at various enterobacterial densities. The feeding rates were calculated using counting plate and selective culture media, for *Escherichia coli*, *Shigella sonnei* and *Salmonella typhi*. Initial concentrations tested were 10^5, 10^7 and 10^9 bacteria ml^{-1} suspended in tap water were offered during 15 min. Based on equivalent body weights of predators, at an initial inoculation density of 10^5 bacteria ml^{-1}, for *E. coli*, S*. sonnei* and *S. typhi*, separately, the feeding rates of *R. rotatoria* were 3.6, 4.2 and 1.3 times higher than *P. multimicronucleatum*, while at 10^7 bacteria ml^{-1}, these were 5.3, 7.7 and 1.2 times higher, respectively. The results are discussed with respect to the bacterial strain and its abundance.

Mate choice in *Brachionus*: Male ability to discriminate female fitness

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Encounters between *Brachionus* conspecific males and females often result in the initiation of mating. It seems to be the male's choice whether to mate and is based on his perception of glycoproteins on the body surface of females. Chemoreceptors in the male corona provide information regarding the potential mate's species, sex, age, and reproductive status. Mate choice is a critical decision affecting male fitness because they are short lived and can perform only about 10 inseminations before their sperm are exhausted. We performed a variety of experiments to probe the sensory ability of males. Males reliably discriminated conspecifics from heterospecifics, males from females, and young from old females. Males did not discriminate amictic from mictic females, virgin from inseminated females, or resting egg hatchlings from amictic egg hatchlings. This is despite the fact that amictic females and resting egg hatchlings are not fertilizable, so inseminating them conveys zero fitness. Whether females have a role in mate choice has been unclear. We observed female resistance to male circling by acceleration and foot flipping. Both these types of behavior tend to knock off males attempting to copulate. Female resistance may erect a barrier to copulation so that only the most agile and persistent males succeed.

The Rotifer Biocomplexity Project: A Biochemical, Genetic, and Genomic Investigation of the Evolution and Ecology of Sexual Reproduction

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Many animal species can give rise to populations that reproduce only asexually, and these asexual populations can rapidly expand at the expense of their sexual progenitors. However, most species and nearly all higher-order taxa maintain sexual reproduction, implying that the abandonment of sex eventually leads to extinction. This is particularly striking in cyclical parthenogens such as monogonont rotifers. Populations of cyclically parthenogens readily lose the ability to undergo meiosis, becoming obligately asexual and enjoying a short-term advantage, yet both sexual and asexual reproduction are maintained at the species level. This suggests a dynamic interaction between reproductive mode, fitness, and population ecology. Of central importance to understanding this dynamic is the evolution and ecological role of the genes that determine the frequency and prevalence of sex in cyclically parthenogenetic populations. Ultimately, these genes influence genomic diversity, rates of adaptation and speciation, and levels of biocomplexity in ecosystems. We have recently been awarded a 5 year Biocomplexity in the Environment grant from the Emerging Frontiers Program at US NSF to study the evolution and population dynamics of the genes that regulate sex and how they correlate with genome variation and ecological change in the monogonont *Brachionus plicatilis*. We are using a combination of biochemical, genetic, genomic, bioinformatic, and modeling approaches to investigate natural and laboratory populations of *B.plicatilis* that have lost or maintained sex under a variety of ecological conditions. Here we review our initial results and discuss genomic tools that will be developed under this grant that will benefit the rotifer community.

Rotifer phylogeny inferred from a combined approach of four molecular loci and morphology

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The phylogeny of selected members of Rotifera is examined based on analyses under parsimony direct optimization and Bayesian inference of phylogeny. Species of the higher metazoan lineages Acanthocephala and Micrognathozoa and potential outgroups are included to test rotiferan monophyly. The data include 74 morphological characters combined with DNA sequence data from four molecular loci, including the nuclear 18S rRNA, 28S rRNA, histone H3 and the mitochondrial cytochrome *c* oxidase subunit I. The combined analyses of the morphological and molecular data sets support the inclusion of Acanthocephala as a rotiferan ingroup, whereas Micrognathozoa is not. Within Rotifera, Acanthocephala, Seisonidea and Bdelloidea form a clade, sister group to the monophyletic Monogononta, but the relationship among the former three clades, remains ambiguously resolved. Within Monogononta, Gnesiotrocha and Ploima are also supported by the data. The relationships within Ploima also remain unclear, but the analyses showed that monophyly was questionable for the families Dicranophoridae, Notommatidae and Brachionidae, and for the genus *Proales*. Otherwise monophyly was generally supported for the represented ploimid families and genera.

Rotifères du sel: salinity and rotifer ecology

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Rotifers are known from habitats of greatly varying salinity, from the most pristine fresh waters to marine settings and hypersaline desert endorheic lakes. The structure of rotifer communities is associated with both the quantity and quality of ions in solution, with certain halophilic species associated with up to 3-times seawater (~3000 mOsmol total salts) and one or more of carbonate-, sulfate- or chloride-dominated waters. In general, the higher the salt content, the lower the overall contribution of rotifers to species richness, although well-known (and expanding) euryhaline multi-species taxa such as the *Brachionus plicatilis* complex, can thrive over a wide range of ionic conditions. Other groups, perhaps typified by *Hexarthra jenkinae*, demographically respond to seasonal and interannual variation in salinity in hypersaline inland lakes. Salinity has striking influence on several life history measures in halophilic rotifers, including diapause termination, juvenile development, survival, reproductive success and induction of mixis. Accordingly, saline rotifer communities have continuous dynamics driven by the halo-tolerances of competitors, predators and prey. These and other factors, including plastic ecophysiological responses to ion quantity and quality, likely have substantial effects on the long- and short-term distribution of halophilic rotifers in natural ecosystems, as well as on their utility in aquaculture and ecotoxicology.

Unilateral induction of sex between freshwater and saline *Brachionus* spp. (Monogononta, Rotifera)

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In many rotifer species, chemicals that accumulate during population crowding can induce sexual reproduction (mixis). The specificity of this crowding response has been recently investigated in two cryptic species complexes of the monogonont rotifer *Brachionus*. In the freshwater rotifer *Brachionus calyciflorus*, mixis induction is highly species-specific. In contrast, the saline *Brachionus plicatilis* does not show any signs of diversification in the mixis signal, as all of its species that have been investigated so far could cross-induce mixis. In this study we tested whether cross-induction was possible between members of these two species complexes, using conditioned media for mixis induction. We demonstrate that cross-induction is possible, but only in one direction: conditioned water from *B. calyciflorus* induced mixis in *B. plicatilis*, but not vice versa.

Rotifer digestive enzymes: Progress in their direct detection using the ELF method

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The enzyme-labelled-fluorescence (ELF) method was used for direct visualization of the location of enzyme activity within the rotifer body. Three groups of enzymes: phosphatases, β-N-acetylhexosaminidases and lipases were examined in rotifers from cultures (*Brachionus angularis*, *B. calyciflorus*, *Keratella cochlearis* and *Lecane lunaris*). We used three artificial fluorogenic substrates, ELF97 phosphate, ELF97 N-acetyl- β-D-glucosaminide, and ELF97 palmiate. After enzymatic hydrolysis, the non-fluorescent substrates change to fluorescent ELF97 alcohol, tagging the sites of enzyme activity. After incubation (30 min. – 3 h) in the test tubes, samples were filtered over polycarbonate filters. Almost six thousand rotifers were examined for presence of enzymatic activity in an epifluorescence microscope. Secretions of all three enzymes listened beyond were detected always in the mastax, stomach and intestine. These activities were connected with the digestive processes. Activities of phosphatases and β-N-acetylhexosaminidases were detected also at the corona, on the lorica and epidermis of rotifers. The role of these extracellular enzyme activities has not been clarified yet.

Analysis of expressed sequence tags (ESTs) of the rotifer *Brachionus plicatilis*

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Rotifers are important live food for the initial stage of larval rearing of marine and freshwater fishes and widely used as model animal in ecotoxicological and population dynamic studies. To acquire the genetic information of *Brachionus*, we have started to construct ESTs from cDNA library. The generation of ESTs has enabled us to have a rapid and efficient approach for discovery of novel gene functions and definition of gene expression profiles. In order to make ESTs of *Brachionus*, we constructed cDNA library from poly $(A)^+$ RNA of axenically cultured *Brachionus plicatilis* (NH1L strain) using a phage vector lambda ZAPII. We randomly selected cDNA clones from the cDNA library and sequenced them either from 5' or 3' end of 2366 clones. There were 512 unique sequences from sequencing clones. These sequences were individually compared to other organisms in the database using BLAST search, 381 (74%) clones matched to the known genes, while 131 (26%) clones had no significant database matches and thus potentially represent *B. plicatilis*-specific genes. Among the 381 former clones, 368 matched to functional genes and 13 matched with ribosomal proteins or RNA genes. Highly abundant clones are 16S ribosomal RNA (5.7%), catepsin L (2.5%), beta tubulin (1.9%) and ribosomal protein L23 (1.6%). The EST data generated in this study will be a valuable resource in further studies on the molecular mechanisms underlying the life history parameters of *B. plicatilis*.

Inheritance of mitochondrial DNA in the rotifer *Brachionus plicatilis*

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Mitochondrial DNA (mtDNA) has been widely studied in various groups of animals, and its inheritance is predominantly maternal. However, paternal inheritance does occur in a few animals and plants. The mode of inheritance of rotifer mtDNA in molecular basis is still unknown. In this study, we investigated the mode of inheritance of mtDNA in *Brachionus plicatilis* using hybrid rotifers. By crossing *B. plicatilis* NH1L and German strains, we obtained two types of hybrids, NH1L (female) × German (male) and German (female) × NH1L (male). Each genomic DNA was isolated from NH1L, German and the two hybrid strains. First, to confirm the crossing of the two hybrid strains on genetic level, random amplified polymorphic DNA (RAPD)-PCR analysis using 10 kinds of primers (10-12 mer) was carried out. Some amplified DNA fragments from RAPD of both hybrid strains showed mixed patterns of NH1L and German strains, confirming that both hybrids are crossbreeds of NH1L and German strains. Second, to determine the mode of mitochondrial inheritance, the full length mtDNA of the four strains were amplified by PCR and double digested with *Bgl*II and *Sal*I restriction enzymes to obtain restriction fragment length polymorphism (RFLP) patterns. Both hybrid strains had the same RFLP patterns as their female parents. These results showed that the mode of mitochondrial inheritance of rotifers is maternal.

Differences in rotifer communities in two freshwater bodies (Lake Ohrid and Lake Dojran, Macedonia) of different trophic status

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This study was conducted to investigate the littoral and pelagic rotifers in two natural Macedonian lakes (Lake Ohrid and Lake Dojran) that differ greatly in regard to time of origin, size and trophic state. These lakes have considerable recreation value and economic importance. A main characteristic of Lake Ohrid ecosystem is the scarcity of nutrients and hence low level of primary production. The lake is rich in biodiversity and especially in relic and endemic species. Lake Dojran represents a typical eutrophic lake. In the last two decades the process of natural aging of the Lake is accelerated due to unreasonable and irresponsible human activities. Due to imprudent exploitation of water and the drought years, there has been a rapid decrease of water level. Depth is reduced to less than 5 m and total destruction of habitats in littoral region is also evident. The Lake Dojran has been brought to the ecological catastrophe. Qualitative analysis of rotifer composition in the Lake Ohrid showed the presence of 66 (11 pelagic and 55 littoral) taxa, whereas 43 (15 pelagic and 28 littoral) taxa were recorded in the Lake Dojran. The structure of the rotifer communities and the similarity index (Jaccard index = 0.2) showed great differences between the lakes. The species in common showed not only morphological but also behavioral differences. The results suggest that trophic status of the lakes is important in determining distribution of rotifer communities.

Implementation of an Acute Toxicity Test for Zinc Using the Freshwater Rotifer *Lecane quadridentata* (Rotifera: Monogononta)

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We implemented an acute toxicity test for zinc using 24-h old neonates of the freshwater rotifer *Lecane quadridentata*. The final test used a range of concentrations of 0.03125, 0.0625, 0.25, 0.5 to 2.0 mg L^{-1} of zinc. A total of five replicates were obtained for the control and each concentration. We used 1-ml test volume in each well of a 24-well polystyrene plate. The CL_{50} value at 48-h was 0.444 mg L^{-1} (95% C.L. = 0.248 – 640 mg L^{-1}). The NOEC = 0.03125 mg L^{-1} and the LOEC = 0.0625 mg L^{-1}. These results are part of a multidisciplinary effort to study the levels of toxicity and contamination of the Río San Pedro watershed in the state of Aguascalientes. These data together with toxicity test on field samples have been used to estimate the potential contribution of zinc to the total toxicity found in each sampling point.

Effects of inbreeding in a rotifer population

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Inbreeding depression -a decrease of fitness due to an increase in genome homozygosity- can be an important evolutionary factor, particularly in rotifers when intraclonal reproduction is expected because a few immigrants colonize a new habitat. If inbreeding depression is important, monopolization of the habitat by first immigrants can be difficult because new immigrants would have colonization chances due to decreased inbreeding. By contrast, rotifer inbreeding depression might be low because rotifer males are haploid and might act as a purge for deleterious recessive alleles. However, rotifers are the only major zooplanktonic group where inbreeding depression has not been studied yet. We studied the effects of inbreeding in *Brachionus plicatilis sensu stricto*. Six clones of the same population were selected as parental clones. The 36 possible crosses between these clones were done, resulting in 6 homogamic (male and female from the same clone) and 30 heterogamic. A total of 1609 sexually produced diapausing eggs were obtained, and several life cycle fitness components in both the eggs and the clones funded from their hatching were determined. We found that fertilization rate was not dependent on the type of cross. However, hatching was significantly lower in homogamic diapausing eggs than in the heterogamic ones. Clone viability, as assessed seven days after hatching, did not show differences, but the viable clones from homogamic diapausing eggs tended to have lower growth rates, lower mixis ratio and much lower diapausing egg production (F2 sexual generation) than clones from the heterogamic ones.

Doing it differently: the bdelloid rotifer's approach to anhydrobiosis

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Bdelloid rotifers are able to withstand desiccation by entering a state of suspended animation. In this ametabolic condition (anhydrobiosis), they can remain viable for extended periods, perhaps decades, but resume normal activities on rehydration. In other desiccation tolerant organisms, e.g. some nematodes, brine shrimp cysts and bakers' yeast, a non-reducing disaccharide, trehalose, is thought to be important for protection against water stress. However, literature shows that bdelloids do not produce this sugar and, therefore, we are looking for other biochemical adaptations associated with bdelloid anhydrobiosis, specifically in *Adineta ricciae*, which is well suited to laboratory culture. A suppressive subtractive hybridisation approach has been used to identify genes upregulated by desiccation and has resulted in the characterisation of approximately 100 genes. Induction of a number of these genes by dehydration has been further confirmed by quantitative PCR. Whereas many of these sequences appear to be novel, others share significant similarity with genes identified in other organisms. One example encodes a protein related to Group 3 LEA (late embryogenesis abundant) proteins associated with desiccation tolerance in nematodes and plants. Further analysis by PCR and Southern blotting has shown that there are probably two related LEA protein genes, which encode similar proteins of ~45 kDa (AriLEA1) and ~40 kDa (AriLEA2). These proteins exhibit a number of unusual properties: for example, unlike all other LEA proteins described so far, both bdelloid proteins contain candidate N-terminal signal peptides, suggesting they are secreted. To provide additional information about rotifer AriLEA1 and AriLEA2, we have produced recombinant forms of the proteins, and further details of the structure and function of the bdelloid LEA proteins have been presented.

Inducible defenses and community dynamics

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Theoretical studies have shown that inducible defenses affect food web dynamics and persistence. We performed laboratory experiments to test the hypotheses of these theoretical studies. First, we studied the occurrence of colony formation within different strains of green algae Scenedesmaceae in response to grazing-released infochemicals from the herbivorous rotifer *Brachionus calyciflorus*. Then, we observed the effect of inducible defenses on the population dynamics of a freshwater planktonic system using algal strains with different defense strategies and rotifers. Simple food webs were composed of green algae (Scenedesmaceae), herbivorous rotifers (*Brachionus calyciflorus* and/or *Brachionus rubens*) and carnivorous rotifers (*Asplanchna brightwelli*). In this system *Brachionus calyciflorus* exhibits an inducible defense against predation by developing long posterolateral spines, while *Brachionus rubens* does not exhibit a defense in presence of *Asplanchna*. Our experimental results showed that inducible defenses, as opposed to the absence of defenses, can prevent population fluctuations and decrease the strength of trophic cascades. Competition experiments between the two herbivorous rotifers under predation by *Asplanchna* indicated that inducible defenses may promote coexistence. As a final step, we fitted the theoretical model to the experimental data to understand the mechanisms underlying the observed dynamics.

Rotifer communities in different types of pools

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Zooplankton investigation in pools is very complicated because of the difficulty in determining the input water into these small ecosystems. Five different pools in the Odra River floodplain were studied. The main aim of the study was to monitor and analyze the factors that mainly affect the formation of the rotifer community, especially the influence of a flood. Qualitative and quantitative analyses of water samples were done fortnightly from spring 2001 to autumn 2005, and monthly in the cold season. Almost one hundred rotifer taxa were determined in the samples. It was needed to measure biomass as a determinant in some cases. Original data of volume biomass were obtained by image analyses (program Lucia). The obtained data do not indicate any explicit patterns in seasonal succession of the rotifer community, but show periods with stronger influence of some biotic factors.

Cultivation experiments with sediments from pools

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A sediment of the different back-water resevoir types may contain resting eggs of many animal species including rotifers and crustaceans. Five pools in the Odra River floodplain were chosen for the experiments because of consequential research. Samples of wet and dry sediment were collected during the year. Sampling area for dry sediment was situated from presently wetted perimeter to the site where water level can reach the maximum (mixed sample from three subsamples). Mixed dry sediment samples were taken by tube from five sites located in different depths. Eggs were not extracted from the sediment because of the separation problems related with various egg and sediment type. Sediments were incubated at 15°C for eight weeks, controlled every day during the first two weeks and twice a week for the rest of the cultivation. Winter and early spring samples were most befitting to cultivation. First rotifers and cladocerans appeared at dry sediment samples after 72 hours. Copepods (nauplii) were identified for the first time during the third and the fourth week. In case of wet sedimets the situation was different. It was impossible to separate cyclopoids resting stages and littoral rotifers from fine sediment that is why these animals had been in samples since the beginning of the experiment. Concurrely with experiments, the zooplankton community succession was recorded. More than twenty rotifer taxa were determined during the experiment.

Genetic Variation in *Plationus patulus* and its relationship to brachionid rotifers

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Recent research suggests the taxonomic placement of *Plationus patulus* needs to be reexamined. During 1993, using SEM, the morphological characteristics of trophi in *P. patulus* were compared to those of several brachionids and considered sufficient to merit the placement of the then known *Brachionus patulus* into a new genus, *Plationus*. However, recent genetic analyses of nuclear ITS and 18S rRNA shows a close genetic relatedness between *P. patulus* and several brachionid species. Moreover, crossmating tests showed that that reproductive isolation is not complete between *Brachionus calyciforus* and *P. patulus*, further supporting the close genetic relationship between the two genera. We analyzed the mitochondrial COI gene and the nuclear internal transcribed spacer region (ITS) of six geographically separated strains of *P. patulus* and eight brachionid species, *Brachionus angularis, B. calyciflorus, B. variabilis, B. quadridentatus, B. bidentatus, B. plicatilis, B. caudatus, and B. budapestinensis*, *Keratella americana, Notholca acuminata* and *Platyias quadricornis*. We discuss genetic variation within populations of *P. patulus* and their phylogenetic placement relative to the brachionid species.

Rotifera of Big Bend National Park, Texas (USA): species richness, turnover, and interannual variation among selected sites

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While exceedingly valuable for their contributions to our knowledge of biogeography, samples taken as part of expeditions to remote habitats are somewhat limited because they usually represent a single picture of the habitat, even when the sampling is comprehensive. Here we examine interannual variation of rotifer species richness (alpha diversity) from 10 aquatic systems, of four habitats—springs, rock pools (*tinajas*), cattle tanks, and ponds—from Big Bend National Park (Texas, USA). At each site, plankton, littoral, and benthic samples were collected at about the same time each summer over the course of five years (2001–2005). Our survey yielded 15 monogonont families comprising 30 genera and 84 species. Two bdelloid taxa also were recognized. Species richness varied widely among these four habitats: (range 1–32; mean (±1SD) = 11.2 ±8.0), with total richness in the four habitats summed over the five summers also varying considerably: springs = 54 taxa > ponds = 35 taxa > *tinajas* =19 taxa > and cattle tanks = 15 taxa. Sessile species comprised ≈13% of the taxa in our samples. Species Turnover Indices of these systems indicated low relatedness: mean (±1S.D.) = 85±7%. The Relative Frequency of Encounter (RFE) of most taxa in the four systems was low, with 79 taxa (≈92%) with values ≤2%. Two bdelloid taxa possessed the highest RFE values (6 and 13%). The singleton rate in the four habitats was quite high, ranging from 47 to 71%, with an overall rate of ≈65%. We recommend that whenever possible researchers use repetitive sampling strategies to provide confidence that a full species inventory has been achieved. Moreover, additional effort is needed to identify bdelloids, which may be significant members of rotifer community structure.

Community composition and phylogeography of selected invertebrates in Chihuahuan desert springs in USA and Mexico

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The Chihuahuan desert is a unique and biologically diverse region. Like other arid ecosystems, the Chihuahuan desert is at risk from a variety of anthropogenic activities including development, overgrazing, and aquifer depletion. Although the Chihuahuan desert is known to to possess a high degree of local endemism, relatively few studies have been focused on its aquatic systems. Moreover, previous works on the Chichuahuan desert have been carried out on larger forms neglecting smaller invertebrates: those that are important in nutrient cycling and which comprise food for invertebrate predators and small fishes. Thus, while these unique freshwater habitats potentially represent an important ecological sentinel for overall environmental health of the Chihuahuan desert, our knowledge of its aquatic fauna is incomplete. There are two components to present investigation: (A) to develop an inventory of the biodiversity of aquatic invertebrates with emphasis on rotifers and small crustaceans. Besides Insecta, these microinvertebrates are likely to represent a large faunistic component of these systems and this study would thus serve as an essential foundation for describing overall large-scale, heterotrophic biodiversity of these ecosystems. An essential component in accomplishing this includes the characterization of the springs and ephemeral waters in terms of their physical structure, water chemistry, and aquatic vegetation. (B) To address other conceptual issues including: assessing the biogeographic history of the target taxa, examining the extent that community structure is regulated by ecological characteristics, and comparing the phylogeographic patterns of multiple, co-distributed taxonomic groups.

Chaoborus predation on zooplankton: no preference for rotifers

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Chaoborus predation was studied on zooplankton in a mesotrophic lake. General consumption rates were recorded using a bioenergetic approach. Gut contents of *Chaoborus* were analysed for all zooplankton groups. Together this information allowed the measurement of individual consumption rates for each species of all groups (cladocerans, copepods and rotifers). Using zooplankton densities, the selectivity indices and loss rates could be calculated. In contrast to the expectation, rotifers were less eaten than cladocerans and copepods. The same result was shown also by the selectivity indices when the abundances data in the plankton were taken into account. Rotifers were mostly avoided and some smaller cladoceran were preferred. *Chaoborus* could not cause high loss rate of the rotifers. As the consumed cladocerans are of the same size as the rotifers, it remains to be tested which factor is responsible for rotifer avoidance.

The clone and primary analysis for meiosis related genes (BpDMC1) of the rotifer *Brachionus plicatlis*

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Monogonont rotifers produce offspring by parthenogenesis or by sexual reproduction (i.e. heterogenesis). It is known that many environmental factors, such as population density, environmental temperature, pH, salinity, DO, osmotic pressure, illumination, the type, size and density of food etc., may induce sexual reproduction in the rotifers. However, different researchers have reported different results even when used the same factor. We believe that the meiosis is the key point in these changes from parthenogenesis to sexual reproduction. Hence, the study of meiosis related genes can help to identify the reasons for change in reproduction methods of the rotifers. In this study, the degenerate primers were designed according to the conservative domain of other (biology) meiosis related genes DMC1/RAD51/RecA. The meiosis related gene analogy fragments were amplified from genomic DNA and cDNA of *Brachinorus plicatilis* by PCR. These fragments were cloned and sequenced. The main results are as follows: 1. The domain of amino acid identity in DMC1/RAD51/RecA genes was used to design two degenerate primers. The fragments of 504bp and 417bp were respectively amplified by PCR and RT-PCR from *Brachinorus plicatilis*. 2. Seven recombinants of PCR and RT-PCR were sequenced. These fragments have been analyzed by BLAST on internet and aligned by DNassist 2.0 and Dnastar. One of DNA and two of cDNA got some homology score at nucleotide level and high homology score at amino acid level (64.5-86.2%) with the meiosis related genes DMC1/RAD51/RecA. The phylogenic relationship of BpDMC1 and other DMC1/RAD51/RecA amino acid fragments of seven organisms was studied. The genetic distance between *Brachinorus plicatilis* and *Dictyostelium discoideum* is the nearest and that between *Brachinorus plicatilis* and *Caenorhabditis elegans* is farthest. 3. To get the 5' and 3' sequences of BpDMC1 requires further research.

The mode of nutrition of mixotrophic flagellates determines their food quality for rotifers

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Mixotrophic flagellates are widespread in pelagic ecosystems and serve as a potential food source for rotifers. Depending on environmental conditions, mixotrophic flagellates acquire carbon either by photosynthesis or by the uptake of dissolved / particulate matter or both. Consequently, the mode of nutrition influences their biochemical composition and thus, their food quality for consumers. We investigated the food quality of the osmo-mixotrophic flagellate *Chlamydomonas acidophila* grown auto-, mixo- and heterotrophically for three rotifer species, *Cephalodella hoodi*, *Elosa worallii* and *Brachionus sericus*. Using life table experiments, we found species-specific differences in the lifespan and the fecundity of the species. Furthermore, we determined the fatty acid composition of the flagellate and the rotifers to get a better insight in potential limiting chemical compounds for reproduction and / or lifespan.

Phylogenetics and ribosomal gene evolution in Bdelloidea

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Bdelloid rotifers may be the oldest successful group of asexual metazoans. As such their phylogeny and evolution is of great interest. Recent examination of bdelloid genomes, combined with studies of the extreme desiccation tolerance and radiation resistance, suggest that bdelloid nuclear genomes are subject to several phenomena that could confound phylogenetic analysis using nuclear markers. Here I present the results of gene surveys of the 18S small subunit ribosomal RNA gene in bdelloids and Bayesian phylogenetic analyses using bdelloid 18S genes collected from databases, gene surveys, and sequencing of genomic library clones. The resulting gene trees are clearly at odds with bdelloid systematics and suggest that no nuclear markers are appropriate for phylogenetic analysis within Bdelloidea. I discuss how the gene trees do show patterns of duplication, divergence, loss, and conversion consistent with expectations for these unusual genomes.

Morphology and taxonomy of Mytilinidae Bartos, 1959

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In 1959 Bartos combined the two genera *Lophocharis* Ehrenberg and *Mytilina* Bory de St. Vincent and erected the taxon Mytilinidae which now contains about thirty species. Although the first *Mytilina*-species was described as early as in 1773 (*M. mucronata* Müller, 1773) and several authors have stressed the need for detailed examinations of Mytilinidae, the whole taxon is still poorly known. Furthermore, there is a lack of standardised documentations. Species determination is often doubtful because of an unclear taxonomy and incomplete knowledge of the morphology. So, redescriptions are urgently needed. When examining seven species using both light and scanning electron microscopic techniques, many important and new species-specific characters were obtained. Microscopic observations of living *Mytilina*-species, for example, showed variations of depth and width of their dorsal sulcus. Contrary to previous descriptions, the closing and widening of the dorsal sulcus does not passively follow head retraction, but is achieved actively and independently of body contraction and feeding condition. Given the importance of the shape of the dorsal sulcus in traditional taxonomy, our observations call into question the diagnostic features traditionally used in species determination.

A computer aided 3D- reconstruction of a rotifer mastax

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Understanding the morphology and function of the rotifer mastax has always played an important role in rotifer taxonomy and phylogeny. A three dimensional reconstruction of the mastax greatly facilitates both our understanding of its function and is also helpful for comparisons among different rotifer species. The present computer-aided approach overcomes the shortcomings of SEM, because a three dimensional reconstruction makes it possible to rotate the trophi in virtual space thus visualising them from all sides. This approach is applied here to the mastax of a dicranophorid rotifer. In order to reconstruct the mastax, a complete series of ultrathin section has to be photographed. In the computer programme SolidWorks®, each image is placed on a layer. The distances between the single layers are proportional to the section thickness of the ultrathin sections. With these images in the background, the contours of the relevant structures of the mastax are drawn on each layer using digital drawing tools. The resulting stack of sil-houettes is then exported to the programme 3DStudioMax® which combines the single draw-ings to complete objects. Having done so, details in shape and surface structure can be modified with different tools.

Effects of DDT, dicofol and estradiol on the life history characteristics of freshwater rotifer *Brachionus calyciflorus*

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The effects of DDT, dicofol and estradiol at concentrations of 0.08, 0.16, 032, 0.64 and 1.28 mg L^{-1}, respectively, on the life history characteristics of freshwater rotifer *Brachionus calyciflorus* were studied at 25°C and using 3.0×10^6 cells ml^{-1} of *Scenedesmus obliquus* as food. Compared with the control, both DDT and dicofol at concentrations >0.32 mg L^{-1} significantly prolonged the juvenile period of the rotifers; and DDT at >0.32 mg L^{-1} and dicofol at >0.64 mg L^{-1} significantly decreased the survival and fecundity, and the life table demographic parameters of the rotifers including net reproduction rate, life expectancy at birth and intrinsic rate of population increase except the generation time. Estradiol at concentrations of 0.08-1.28 mg L^{-1} did not significantly affect the duration of juvenile period and the demographic parameters of the rotifers.

The effect of vitamin E on density dynamics of the rotifer *Brachionus calyciflorus* at different temperatures

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The population density changes of the rotifer *Brachionus calyciflorus* cultured at different concentrations (20, 40, 80 and 100 ng ml^{-1}) of vitamin E were studied at three (15, 25 and 30°C) temperatures. At 15°C, there was a positive correlation between the population density and culture time for the treatments receiving 20, 40 and 80 ng ml^{-1} and the corresponding regression equations were $y=e^{(-0.330+0.544X)}$, $y=e^{(-0.230+0.500X)}$ and $y=e^{(-0.370+0.369X)}$, respectively. The peak rotifer densities in treatments receiving 20 and 40 ng ml^{-1} of vitamin E were higher than those in the other treatments at this temperature. At 25°C, the peak population densities were 68, 122, 124, 168 and 236 ind. ml^{-1}, respectively for control and those receiving vitamin E of increasing levels. The positive relation between the population abundance and the culture time in treatments receiving vitamin E of 80 and 100 ng ml-1, was expressed as $Y=e^{(1.361+0.325X)}$ and $Y=e^{(1.017+0.354X)}$. At 30°C, the peak population abundances of B. calyciflorus were 114, 121, 123, and 151 ind. ml^{-1}, respectively in treatments receiving increasing levels of vitamin E and the corresponding regression equations: $Y=e^{(-0.815+0.520X)}$, $Y=e^{(0.456+0.463X)}$, $Y=e^{(-0.148+0.854X)}$ and $Y=e^{(1.028+0.424X)}$. The significant promoting effects of vitamin E on rotifer densities existed between the experimental groups and in control.

The fatty acid analysis of *Monodus subterraneous* cultured in marine and freshwaters and the effects of enrichment on the rotifer *Brachionus plicatilis*

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The composition of fatty acids, which were extracted from the alga *Monodus subterraneous* cultured in freshwater medium BG-11 and in seawater f/2 medium after being acclimatized gradually from 0.05‰ to 20‰ and from the rotifer *Brachionus plicatilis* enriched with *Monodus subterraneous* for 12, 24, 72 hours and 7 days respectively, was analyzed. The results showed that the fatty acid composition was similar from the freshwater medium and that from the seawater-adapted. Both of them were dominated by C18:3, C18:1n-9, C16:0 and C16:1n-9. The fatty acids C20:1n-9 and C20:4n-6 were present in algae cultured in seawater medium but were absent in freshwater medium. The proportion of C16: 2n-4, C16:3, C18:0 and C18: 1n-9 increased when the alga cultured in seawater than that in freshwater medium; on the contrary, the proportion of C14:0, C16:0 and C16: 1n-9 decreased. The fatty acids were dominated by C16:0, C16: 1n-9, C16:2n-4, C18:1n-9, C18:2n-6 and C18:3 in *Brachionus plicatilis* enriched with alga of different durations. The highest proportion of eicosapentaenoic acid (7.82%) and docosahexaenoic acid (1.8%) appeared at 72 hours after enriched with alga.

Swimming behaviour of *Brachionus plicatilis* in response to food concentration and its relation to feeding rates

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Filtration and ingestion rates are considered as indicative of the feeding behaviour exhibited by suspension feeding organisms under different environmental conditions. In free swimming rotifers, the search for food and subsequent particle uptake is obviously related to swimming activity. The objective of this study was to determine to what extent filtration rate, measured from algal cells removed from water, is a consequence of a real modulation of the swimming speed in response to the food availability. With this purpose, this study examines the swimming speed in amictic females of *Brachionus plicatilis* at a wide range of algal cell concentrations in the laboratory cultures. The swimming rates were measured using an automated motion analysis system over video recording. Results show that swimming speed actually changes as a function of food density, showing constant values at high cells concentration and a decreasing trend at progressively lower concentrations. The observed pattern is linked to predation efforts but it does not fully explain the changes in filtration rate. The swimming activity seems to be limited by the energy available from continuous cell uptake.

Spatial Dynamics of Rotifers in a Large Lowland River, the Elbe, Germany - How important are retentive shoreline habitats for the plankton community?

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The longitudinal dynamics of rotifers in the potamal region of the River Elbe between Dresden and Geesthacht (Germany) was investigated on five surveys in 1999 and 2000. All surveys included transversal sampling (left bank, main channel, right bank) as well as sampling of five major tributaries and twelve connected backwaters to evaluate lateral impacts on the zooplankton community. A simple model of longitudinal development for the dominant species *Trichocerca pusilla* was calculated, based on the results of in situ incubations and on literature data, and compared with the dynamics observed on the lagrangian surveys. To characterize the influence of shoreline structure on the main channel zooplankton community, additional lateral sampling programmes were conducted in selected regions of the river. The zooplankton community of the River Elbe was clearly dominated by rotifers, with 5000–18000 ind. L^{-1}. A distinct increase of rotifers in a downstream direction was observed on all longitudinal surveys. In addition to demonstrating the lateral influences due to tributaries, the results of this study indicate that the longitudinal development of rotifers can be explained predominantly by reproduction during downstream transport. This means that the influence of retentive shoreline habitats seems to be negligible for the development of the rotifer community in large rivers.

Bdelloid rotifers Join the 'Odd Splicing Club'

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Analysis of full-length cDNA libraries from bdelloid rotifers *Adineta ricciae* and *Philodina* sp. revealed that at least 50-65% of their genes feature a common 23 nucleotide sequence at their 5' ends. This is diagnostic of spliced leader (SL) RNA-mediated trans-splicing - an unusual mechanism for the generation of mature messenger RNAs, where 5' ends of precursor mRNAs are replaced by a short exon from a separate small SL RNA. We characterised the small RNA encoding SL in Rotifera. Like other known spliced leader RNAs, rotifer SL RNA comprises the SL 23 nt exon itself, a conserved splice donor site and an intron containing a putative spliceosome binding motif. We also demonstrated that this conserved 5' end sequence could be used as an anchor in production of full-length rotifer cDNA libraries and provided additional evidence for *trans*-splicing in bdelloids by analysing a number of transcribed and genomic copies of an *A. ricciae hsp82* gene. SL RNA-mediated *trans*-splicing has not been found in vertebrates, insects, plants or yeast and its current phylogenetic range in metazoan phyla includes nematodes, cnidarians, platyhelminths, primitive chordates and now bdelloid rotifers. Interestingly, where *trans*-splicing occurs, no significant nucleotide sequence conservation of SL RNAs between phyla has been observed. The lack of SL RNA homology and the apparently sporadic phylogenetic distribution have led to opposing hypotheses on its evolution, involving either an ancient origin followed by loss in multiple lineages, or independent acquisition in several taxa. However, the description of SL-mediated *trans*-splicing in Rotifera extends its representation to at least five metazoan phyla, making it increasingly probable that this is a phylogenetically widespread and therefore ancient phenomenon.

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