

Studies on comparative population growth of some species of the rotifer *Lecane* (Rotifera)

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Abstract

We compared the population growth patterns of 5 species of the rotifer genus *Lecane* [*L. quadridentata* (Ehrenberg, 1830), *L. cornuta* (Muller, 1786), *L. papuana* (Murray, 1913), *L. unguitata* (Fadeev, 1925) and *L. pyriformis* (Daday, 1905)] ranging in adult average body size from 30 to 140 μm . All species were cultured under laboratory conditions for 25-30 days using the green alga *Scenedesmus acutus* as the exclusive diet, at a density of 1.0×10^6 cells ml^{-1} at 24°C . Regardless of the species, lecanids reached their peak population densities after 4 weeks. Peak population densities ranged from 15 to 320 ind. ml^{-1} , depending on body size. There was an inverse curvilinear relation between body lengths and peak population abundances (densities) of the *Lecane* species. Egg ratios (eggs per female) for the tested species were <0.6 during the exponential phase but declined to 0.1 (or lower) as the population density increased. The rates of population increase for the lecanids were in general lower (0.10 to 0.21 day^{-1}) than other well-studied rotifer species including members of Brachionidae.

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Introduction

Rotifers of the genus *Lecane* are non-planktonic, inhabiting mostly ponds and lakes. This genus is the second largest among rotifers and with about 160 valid species (Segers, 1995). Like many other non-planktonic herbivorous rotifers such as *Lepadella* and *Euchlanis*, members of *Lecane* feed on bacteria, detritus and green alga such as *Chlorella* and *Scenedesmus* (Gulati *et al.*, 1987; Soto and Sarma, 2009) and form part of the diet of several benthic invertebrates and vertebrates (Wallace *et al.*, 2006), thereby serving as a food link between primary producers and carnivores. They are also used as test organisms in ecotoxicological studies (Snell and Joaquim-Justo, 2007). Field observations have shown that many species of *Lecane* can coexist in a given water body (Ejsmont-Karabin and Kuczynska-Kippen, 2001; Nandini *et al.*, 2005). However, in spite of the fact that all the species of *Lecane* co-occurring in a waterbody have similar availability of food, some species such as *L. closteroerca* (Schmarda, 1859) and *L. inermis* (Bryce, 1892) are usually in high abundances (>10 ind. l^{-1}) while species such as *L. unguolata* (Gosse, 1887) and *L. stenroosi*

(Meissner, 1908) occur in lower numbers (<1 ind. l^{-1}) (Jiménez-Contreras *et al.*, 2009). Most field studies explain this on the basis of relative body sizes of the zooplankton species (Nandini *et al.*, 2008). Usually smaller rotifers are numerically more abundant than larger species under comparable conditions (Nandini *et al.*, 2007). However, quantitative data on patterns of population growth of several species of *Lecane* when cultured under identical conditions are rare (Sarma *et al.*, 2006). This information is needed for understanding the relative abundances of rotifers in nature.

The relative abilities of different lecanid rotifers to feed and grow on a given algal diet depends on many factors: ability to gather the food from the medium, assimilation efficiencies to convert assimilated energy into offspring production, the rate at which offspring are produced and others such as the specific growth rates (Hernández-Rodríguez *et al.*, 2000; Wallace *et al.*, 2006). Population dynamics sums up all such variables and is thus reflected as the difference between birth rate and death rates. Therefore, laboratory studies on the population growth of different zooplankton species are a convenient way to quantify the changes in the death rates and

birth rates (Walz, 1993). Laboratory studies have shown that under food limited conditions rotifers of the genus *Brachionus* reach peak abundances in about two weeks while those of *Lecane* take longer (Sarma et al., 2006; Nandini et al., 2007).

Planktonic rotifers such as *Brachionus* and *Keratella* carry their eggs until hatching at the posterior end of the body. The number of eggs carried by a female reflects fecundity; higher the egg number the higher is the fecundity (Edmondson, 1965). The number of eggs per female, or the egg ratio, declines due to several factors including stress induced by changing abiotic conditions, contaminants or increasing levels of inter- and intra-specific competition (Sarma et al., 2005). In non-planktonic rotifers (except those attached to vegetation e.g., *Sinantherina* and *Ptygura*) the eggs are deposited on a substratum and therefore it is difficult to count (Wallace et al., 2006). In waterbodies where many species of the same genus simultaneously exist, it is difficult to definitively attribute the deposited eggs to any particular species. However, under laboratory conditions since each species is separately cultured, it is possible to derive the egg ratio.

The aim of the present work was to quantify changes in the population abundances of five different lecanid species (*L. quadridentata* (Ehrenberg, 1830), *L. cornuta* (Müller, 1786), *L. papuana* (Murray, 1913), *L. unguitata* (Fadeev, 1925) and *L. pyriformis* (Daday, 1905) when grown on similar algal diet at a concentration of 1×10^6 cells ml^{-1}). In addition, since eggs are deposited in the bottom of the test jars, we also derived the egg ratio.

Materials and Methods

In this study we used *Scenedesmus acutus* Strain no. 72, a green alga (in mostly single-celled stage) to culture the various species of *Lecane*. The original stock of alga was obtained from Texas University cultures (Austin, USA). The alga was batch-cultured on Bold's basal medium (Borowitzka and Borowitzka, 1988) in 2 l transparent bottles under continuous fluorescent illumination and aeration, supplemented with sodium bicarbonate as carbon flow at the rate of 0.5 g l^{-1} every third day. Log phase alga was harvested, centrifuged at 4000 rpm for 5 min and rinsed and re-suspended in distilled water. For the stock rotifer cultures as well as for the experiments we used reconstituted moderately hard water (EPA medium) which was prepared by dissolving 0.9 g of NaHCO_3 , 0.6 g of CaSO_4 , 0.6 g of MgSO_4 and 0.002 g of KCl in 1 l of distilled water (Weber, 1993). Laboratory conditions that consistently resulted

in healthy *Lecane* species in our stock cultures were temperature $24 \pm 1^\circ\text{C}$, continuous but diffused fluorescent light, pH 7.1 -7.3 and the change of medium and replacement of food every alternate day.

The five *Lecane* species (*L. quadridentata*, *L. cornuta*, *L. papuana*, *L. unguitata* and *L. pyriformis*) were mostly isolated from Chimaliapan Lagoon and additional waterbodies in the State of Mexico. Stock cultures for each of these species were separately established starting from a single parthenogenetic female. Rotifers in the stock cultures were fed *S. acutus* at a density of 1×10^6 cells ml^{-1} .

For each rotifer species, population growth experiments were conducted in 50 ml capacity transparent jars, each with 20 ml medium and with an algal density of 1×10^6 cells ml^{-1} of *S. acutus*. For each species we used 4 replicates. Into each test jar containing specified food density and medium, we introduced 20 individuals of one of the five lecanid species. Care was taken to avoid mixing of one species with other in test jars. The test jars were randomly placed in temperature controlled chamber ($24 \pm 1^\circ\text{C}$). Following initiation of growth experiments, we counted daily the number of live individuals and the eggs in each replicate (using whole sample (replicate) count or the 2-3 aliquots of 1 ml each) and transferred them to the new jars containing fresh medium. The growth experiments were terminated on day 30 by when populations in most replicates began to decline.

For each species, the population growth rates were calculated using the formula: $r = \ln N_t - \ln N_0 / t$ where N_0 the initial population density, N_t the population density at time t in days (Krebs, 1985). One way-analysis of variance (ANOVA) was used to quantify differences in the peak population abundances and the growth rates of the tested rotifer species.

Results and Discussion

Population growth curves of *L. quadridentata*, *L. cornuta*, *L. papuana*, *L. unguitata* and *L. pyriformis* and the corresponding egg ratios (eggs per female) are shown in Fig. 1. In general, the 3 of the 5 tested lecanid species (*L. pyriformis*, *L. unguitata* and *L. cornuta*) had a long lag phase of about 10 days, while *L. papuana* and *L. quadridentata* started to grow immediately after initiation of the experiments. All the tested *Lecane* species had a high egg ratio (up to 0.6) but declined to <0.1 , as population densities began to increase. During the exponential phase, the egg ratios were generally lower for *L. papuana* than for the other species.

Table - 1: Results of the one way ANOVA performed on the peak population abundances and the rate of population increase of five species of *Lecane*

Source of Variation	DF	SS	MS	F
<i>Peak population density (ind. ml⁻¹)</i>				
Among the species	4	29696.07	74240.17	49.48***
Error	15	22504.8	1500.32	
<i>Rate of population increase (r) day⁻¹</i>				
Among the species	4	0.032	0.01	121.74***
Error	15	0.001	0.00	

DF: degrees of freedom. SS: Sum of squares; MS: Mean square; F: F-ratio, *** = $p < 0.001$

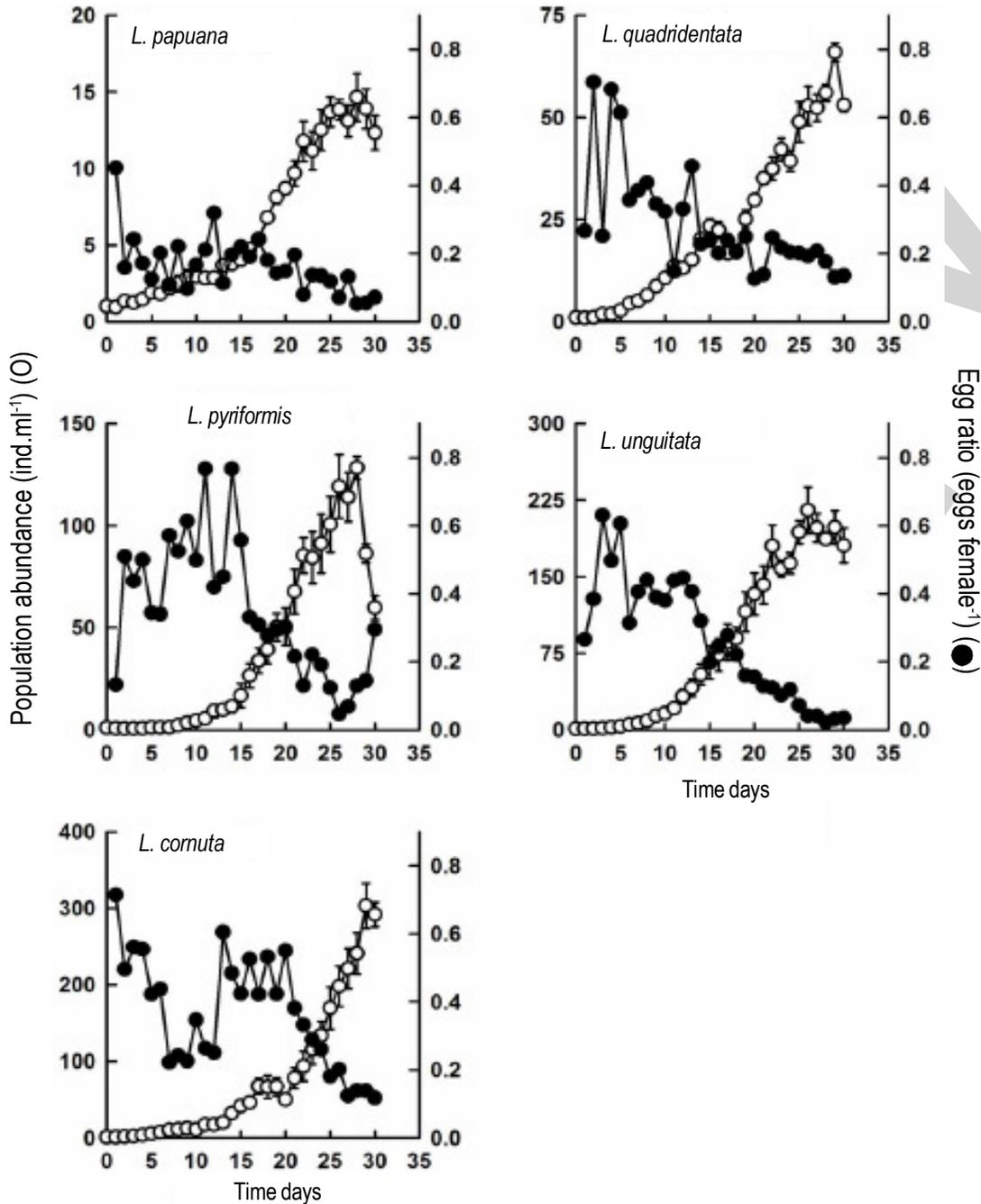


Fig. 1: Population growth curves (open circles) (O) and egg ratio curves (closed circles) (●) of five species of *Lecane* (*L. papuana*, *L. quadridentata*, *L. pyriformis*, *L. unguitata* and *L. cornuta*) in relation to time and cultured at a food level of 1×10^6 cells ml^{-1} of *Scenedemus acutus*. Shown are the mean \pm standard error based on four replicates. Note the differences in the scale on Y-axis

Peak population abundances of the tested lecanids varied from 15 to 320 ind. ml^{-1} and showed an inverse curvilinear relation with the body size (Fig. 2). Thus, smaller species had higher peak abundances compared to larger ones. The rate of population increase for *Lecane* species varied from 0.10 to 0.21 day^{-1} (Fig. 3). The larger species (*L. papuana* and *L. quadridentata*) had lower growth rates compared to the smaller species. Statistically the peak

population abundances and the growth rates of the tested rotifer species differed significantly ($p < 0.05$, ANOVA Table 1).

As far as we know this is the first attempt at determining the comparative population growth of five lecanid species, although similar studies have been conducted for various species of the genus *Brachionus* (Fernández-Araiza *et al.*, 2005). The growth curves of the tested *Lecane* indicated a trend nearly common to all

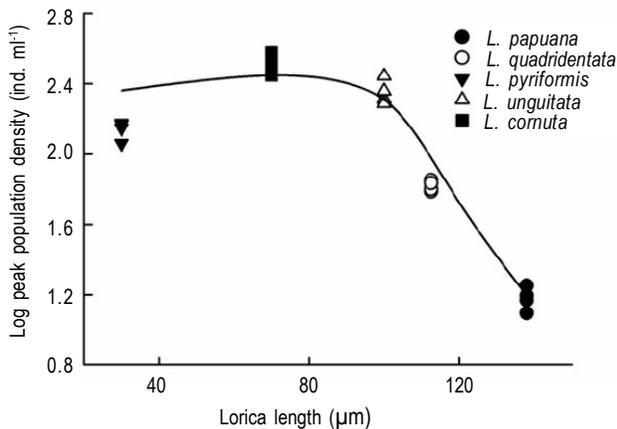


Fig. 2: Relation between peak population density (ind. ml⁻¹) (log₁₀) and the body size (lorica length, μm) of five species of *Lecane* (*L. papuana*, *L. quadridentata*, *L. pyriformis*, *L. unguitata* and *L. cornuta*) ($p < 0.05$). Plotted are the replicates for each species

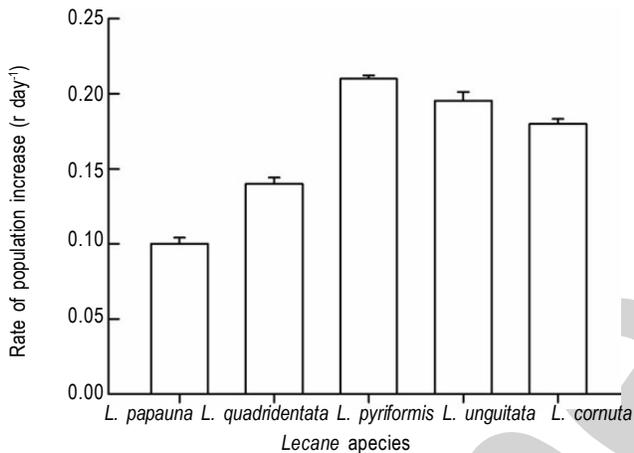


Fig. 3: Rate of population increase (r , day⁻¹) of five species of *Lecane* (*L. papuana*, *L. quadridentata*, *L. pyriformis*, *L. unguitata* and *L. cornuta*). Shown are the mean \pm standard error based on four replicates. Table 1. Results of the one way ANOVA performed on the peak population abundances and the rate of population increase of five species of *Lecane*. DF: degrees of freedom, SS: sum of squares; MS: mean square; F: F-ratio, *** = $p < 0.001$

in that the peak population densities were reached only towards the 4th week. This study confirms earlier observations on *L. quadridentata* (Sarma et al., 2010) where the peak population densities were obtained only after 3 weeks while in most brachionids rotifers, these were reached earlier than 2 weeks (Nandini et al., 2007). The delay in attaining the maximal value later than brachionids is due to the reduced egg output per female and or prolonged egg hatching time (Herzig, 1983).

Edmondson's (1965) egg ratio method was originally developed to measure the population growth rates. However, it is also widely used to interpret the health of rotifer in mass culture tanks (Snell et al., 1987). Usually, during the initial growth, most brachionid species have an egg ratio of 1.0 or more (Sarma et al.,

2005). However, as the populations build up, food limitation sets in leading to reduced egg ratios. For the mass cultures of *Brachionus plicatilis* Müller, 1786, if the egg ratios fall below 0.13, it indicates food-limited conditions which lead to decline of the populations (Snell et al., 1987). Sarma et al. (2005) have reviewed the pattern of egg ratios in planktonic rotifers and reported that under non-stressful conditions, the egg ratios could vary from 0.2 to 2.5 depending on the species and the stage of population growth. In our present work too, there was an increase in the egg ratio during the initial and exponential stage of the population growth. However, as population density began to reach towards the peak abundances, the egg ratios began to fall and in some cases they were close to zero.

The maximum densities reached by aquatic organisms are dependent on the body size (Duarte et al., 1987). It has been shown for cladocerans (Nandini and Sarma, 2003) and for rotifers (Nandini et al., 2007) that the peak population density is inversely related to the body size. In our present work, the body sizes of the tested rotifer species varied from 30 to 130 μm. The relation between the body size and the peak population abundances in this study followed an inverse relation which thus agrees with other published data (Nandini et al., 2007).

The rate of population increase is another important life history variable (Sibly and Hone, 2002). For most rotifer species the r varies from 0.2 to 1.0 and in a few cases up to 2.0 (Miracle and Serra, 1989). In the present work, the r values for the tested rotifer species were on the lower side of this range. Not much information is available on the r values of *Lecane* species (Pérez-Legaspi and Rico-Martínez, 1998). It appears that when cultured under similar food levels, members of Lecanidae in general have lower growth rates (< 0.5 day⁻¹) (Sarma et al., 2010) compared to those in Brachionidae (> 0.5 day⁻¹) (Sarma et al., 2001).

The tested *Lecane* species when cultured on a food density of 1×10^6 cells ml⁻¹ of *Scenedemus acutus*, had peak population abundances of 15 to 320 ind. ml⁻¹, depending on the body size of the rotifer species. There was an inverse curvilinear relation between body length and peak population abundances of *Lecane* species. Egg ratios during the exponential phase of the population were close to 0.6 but declined to nearly zero as the density increased. The rate of population increase (r) of lecanids was nearly one-third of brachionids under comparable test conditions.

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