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Use of swimming speed and egg ratio as predictors of the status of rotifer cultures in aquaculture

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Key words: Aquaculture, *Brachionus plicatilis*, rotifera, swimming speed, egg ratio, culture quality

Abstract

This study evaluated the use of egg ratio (eggs rotifer^{-1}) and swimming speed (mm min^{-1}) as prediction criteria for production and culture quality in mass cultures of the rotifer *Brachionus plicatilis*. Egg ratio was determined to be a suitable predictor of rotifer growth and production in the cultures. Low egg ratios (i.e., 0–0.17 eggs rotifer^{-1}) indicate reduced rotifer population over time (i.e., negative net population growth rates). However, at this time egg ratio dynamics are not suitably understood to predict in advance a sudden population collapse.

Swimming speed of reproductive, egg-carrying females in the exponential growth phase was 40–45 mm min^{-1}. During exponential growth swimming speed was independent of the food used. Lower swimming speeds were obtained in late stationary phase (10–25 mm min^{-1}) when yeast was used as a food source. Both environmental factors (e.g., accumulating metabolites) and changes in nutritional state of the rotifers may have affected the swimming speed, but environmental factors appear to be the most important. We believe that swimming speed has the potential of becoming an accurate predictor of culture quality in mass cultures of rotifers.

Introduction

During the last thirty years the marine aquaculture industry has expanded from raising species of fish which can be easily cultured on artificial food (e.g., salmon) to growing more demanding species which require live food during their early stages (e.g., halibut and turbot). Most farms mass-produce the live food needed for the early feeding period. For example, turbot *(Scophthalmus maximus)*, which have a relatively small yolk sac, are normally fed the rotifer *Brachionus plicatilis* from the time when the mouth first opens (about 2 days after hatching) until they are large enough to ingest *Artemia* nauplii (Cunha & Planas, 1993; Reitan *et al.*, 1994). Because of the importance of having enough high quality live food on hand to raise marine fish, aquaculture farms devote considerable resources to maintain healthy rotifer cultures. As a hedge against disaster, commercial operations usually harvest their rotifers well before a potential crash, sometimes after only two days of growth (Attramadal & Sandvand, 1993). Those farms that prolong the period before harvesting normally take daily samples of the rotifers and base their predictions of the quality of the culture on rotifer density and on the number of eggs per female (egg ratio).

Snell *et al.* (1987) proposed measurement of swimming speed as another criterion to assess the healthiness of rotifer cultures. In this method, individual animals are taken from the culture, placed on a microscope slide in a drop of the culture fluid, and observed for a short time (0.5–2 min). Under those conditions healthy animals are reported to swim faster than unhealthy ones (Snell *et al.*, 1987; Janssen *et al.*, 1993). Here we report a study in which replicate rotifer cultures were followed from inoculation to early decline to evaluate use of egg ratio and swimming speed as production and quality criteria for mass cultures of *B. plicatilis*.

Materials and methods

Six rotifer cultures (3 treatments × 2 replicates each) were inoculated with approximately 3 ind ml^{-1} and daily fed one of the following foods at 10–15 mg C l^{-1}: Baker's yeast plus Super Selco (10:1 ratio by wet weight; Artemia Systems S. A., Belgium – hereafter referred to as yeast), the microalga *Isochrysis galbana* (Prymnesiophyceae), or the microalga *Tetraselmis* sp. (Prasinophyceae). Algal cultures were grown semi-continuously in f/2 media (Guillard, 1975) under constant illumination and were harvested by daily removal of c. 30% of the culture volume with addition of new media and seawater. The rotifer cultures were kept at a volume of 100 l in 250 l conical tanks by draining from the bottom an equal volume of the culture just before adding new food each day. Other rotifer culture conditions maintained throughout the 35 day period of this study were as follows: 20 °C, strong water aeration, 20‰ salinity, and low light (one 60 watt bulb, 50 cm over each tank).

Daily measurements were made of number of rotifers per ml, egg ratio, and swimming speed. Swimming speed was measured by a modification of the method described by Snell *et al.* (1987). Individual females with one egg were carefully pipetted into a microscope depression slide (depression 15 mm diameter and 1 mm deep) with 150 µl water. The slide was transferred to a dissecting microscope and the observations were done under conditions of low substage illumination at room temperature (21–22 °C). A gridded piece of paper (1 mm squares) was placed under the slide and the number of millimeters that the animal moved in one minute was recorded. Average from five replicate females was used in our statistical analysis. If a rotifer attached and thus stopped swimming for > 5 sec, the female was replaced by a new one.

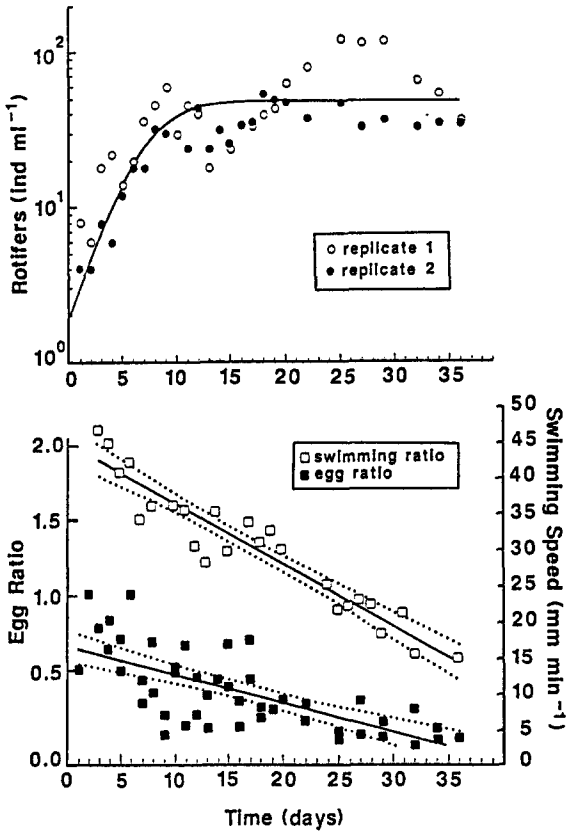

Fig. 1. Changes in numbers of individuals, egg ratio and swimming speed with time for rotifers fed yeast + Super Selco. The data for numbers of rotifers were fitted to the equation: $y = a(1 + ((a - N_0)/N_0)e^{\mu\,max})^{-1}$, where N_0 is the initial rotifer density and μ_{max} is the specific growth rate at 20 °C and 20% salinity at food saturated conditions (Olsen *et al.*, 1993). Data for changes in egg ratio and swimming speed were fitted by a linear regression; dotted lines show the 95% limits of confidence.

Results

The numbers of rotifers increased exponentially with time in the early phase in all cultures and reached an apparent constant level (saturation) after two weeks of cultivation (Figs 1–3). Saturation was different for the 3 diets used (53 ind ml^{-1} for yeast, 86 ind ml^{-1} for *Isochrysis*, 138 ind ml^{-1} for *Tetraselmis*) because population growth is dependent on the food ration used.

Egg ratio decreased steadily with time during the various phases of growth. This decrease was faster in the early exponential phase than in the later phases in some of the cultures. In each replicate, egg ratio approach zero by the end of the experiment (Figs 1–3). Short term variation in the egg ratio was often pronounced during the exponential phase of growth. This is commonly observed in mass cultivation, and is probably a result of partial population synchronism in the egg production rather than uncertainty during sampling and counting. The extent of synchronism, which is believed to be a result of the changes in conditions during inoculation, will theoretically fade off in the early stationary phase.

Swimming speed decreased linearly with time in all cultures ($P < 0.05$ for yeast and *Tetraselmis*, $P < 0.10$ for *Isochrysis*). The values obtained for yeast-fed

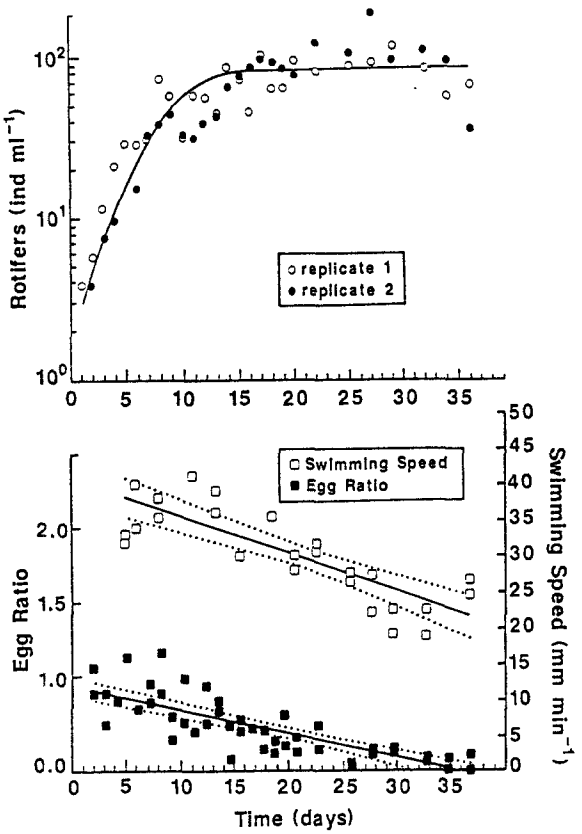

Fig. 2. Changes in numbers of individuals, egg ratio, and swimming speed with time for rotifers fed *Isochrysis*. (See legend in Fig. 1).

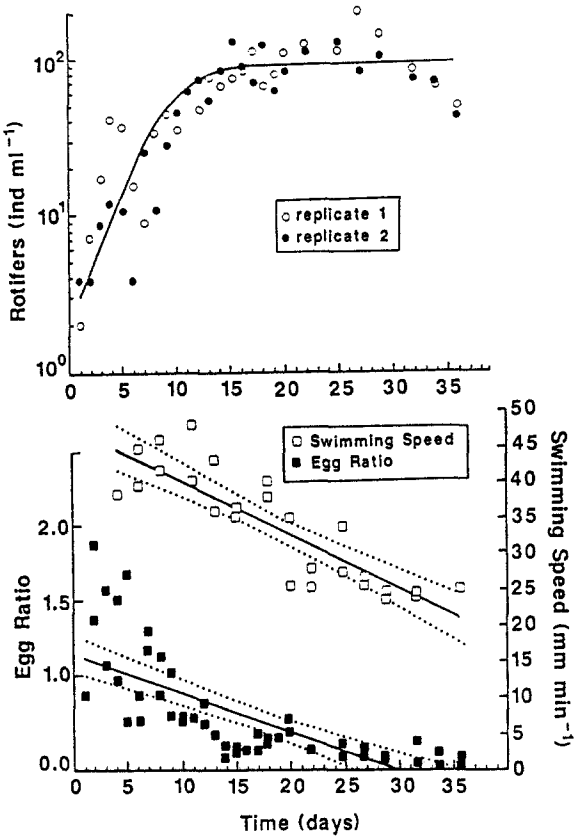

Fig. 3. Changes in numbers of individuals, egg ratio, and swimming speed with time for rotifers fed *Tetraselmis*. (See legend in Fig. 1).

rotifers were slightly lower than corresponding values obtained for algal-fed rotifers, but the general pattern of decrease with time was identical for all cultures.

The relationship between egg ratio and swimming speed for all cultures and diets are summarized in Fig. 4. The data exhibited pronounced scatter, but these traits were positively correlated ($P < 0.10$). Simple correlation analysis revealed that 35% of the observed variation in swimming speed could be explained by variation in egg ratio. The data suggest that algal-fed rotifers showed a slightly higher swimming speed than yeast-fed rotifers at given egg ratios (Fig. 4), but more data are needed to confirm this finding.

Discussion

Our method of measuring swimming speed differed slightly from the one of Snell *et al.* (1987) because the total distance that the rotifer moved was recorded and not just the number of squares entered. The methods are probably equivalent, and are both well suited for use in fish farms. Use of video camera with computer-assisted motion detector would, on the other hand, be beneficial for research purposes.

Most of our cultures showed declining rotifer densities after 4 weeks of cultivation. The egg ratio was < 0.17 eggs rotifer^{-1} after 3–4 weeks of cultivation in most cultures, and reached values close to zero by the end of the experimental period. Birth will balance mortality in cultures when the egg ratio is 0.17 eggs rotifer^{-1} (Reitan *et al.*, 1994), but the population is expected to decrease at lower egg ratios. Therefore, we concluded that egg ratio is a suitable predictor of population growth and production in the cultures. The potential of the egg ratio to predict a sudden collapse of the population in advance cannot be evaluated, because no such events occurred. Values in the range of 0–0.17

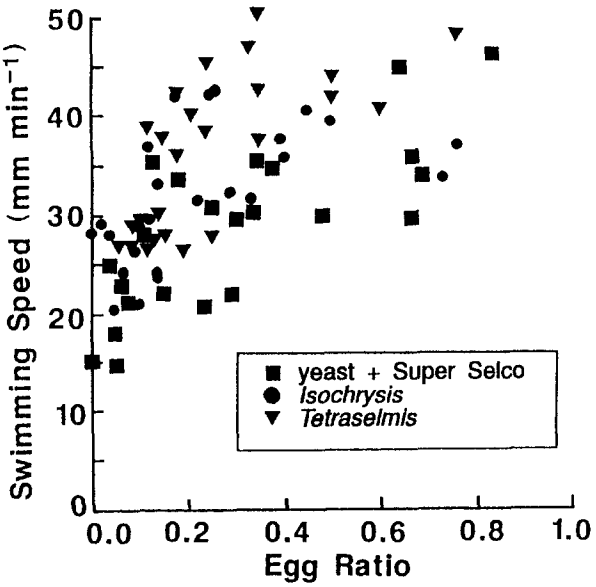

Fig. 4. Relation between egg ratio and swimming speed for rotifers fed yeast + Super Selco, *Icochrysis*, and *Tetraselmis*.

eggs rotifer^{-1} will, however, indicate reduced population with time or negative net growth of the rotifer population.

The swimming speed of reproductive rotifer females carrying one egg during exponential phase of growth was 40–45 mm min^{-1}. During exponential growth swimming speed was independent of the type of food used. The value decreased linearly with time and after 35 days reached slightly lower values for yeast-fed rotifers (10–15 mm min^{-1}) than for algal-fed rotifers (20–25 mm min^{-1}). Swimming speed was inversely related to rotifer numbers and positively related to the population growth rate (slope of curves, Figs 1–3) and egg ratio (i.e., birth rate). Both environmental factors (e.g., accumulating metabolites) and changes in the nutritional state of the rotifers with time may have affected swimming speed.

Swimming speed cannot replace egg ratio as a predictor for growth during the subsequent 24 h period in mass cultures. The fact that the swimming speed of egg-carrying females decreased steadily during the stationary phase, when egg ratio and growth rate remained fairly constant with time, may suggest that swimming speed depends more on environmental factors than on nutritional factors. If so, swimming speed may turn out to be a useful predictor of culture quality. However, our data do not allow speculation of possible mechanisms, and further studies are needed to make final conclusions.

Because swimming speed was independent of the food used, we conclude that it has the potential of becoming a predictor of culture quality in mass cultures of rotifers. However, at this time the protocol for its use must be further refined.

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References

Attramadal, Y. G. & M. Sandvand, 1993. A rotifer cultivating method resulting in high production and stability. In Reinertsen, H., L. A. Dahle, L. Jørgensen & K. Tvinnereim (eds), Fish Farming Technology, Proceedings of the First International Conference on Fish Farming Technology, Trondheim, Norway, 9–12 August 1993: 135–139.

Charoy, C. & P. Clement, 1993. Foraging behavior of *Brachionus calyciflorus* (Pallas): variations in the swimming path according to presence or absence of algal food *(Chlorella)*. Hydrobiologia 255/256 (Dev. Hydrobiol. 83): 95–100.

Cunha, I. & M. Planas, 1993. A study on the maximization of the ingestion rates of turbot larvae using different sized live prey. ICES 1993 Symposium on Mass Rearing of Juvenile Fish/No. 29: 11 pp.

Guillard, R. R. L., 1975. Culture of phytoplankton for feeding marine invertebrates. In Smith, W. L. & M. H. Chanley (eds), Culture of Marine Invertebrate Animals. Plenum Press, New York: 29–60.

Janssen, C. R., M. D. Ferrando Rodrigo & G. Persoone, 1993. Ecotoxicological studies with the freshwater rotifer *Brachionus calyciflorus*. Hydrobiologia 255/256 (Dev. Hydrobiol. 83): 21–32.

Olsen, Y., K. I. Reitan & O. Vadstein, 1993. Dependence of temperature on loss rates of rotifers, lipids, and 3 fatty acids in starved *Brachionus plicatilis* cultures. Hydrobiologia 255/256 (Dev. Hydrobiol. 83): 13–20.

Reitan, K. I., J. R. Rainuzzo & Y. Olsen, 1994. Influence of lipid composition of live feed on growth, survival and pigmentation of turbot larvae. Aquacult. Int. 2: 1–16.

Snell, T. W., M. J. Childress & E. M. Boyer, 1987. Assessing the status of rotifer cultures. J. World Aquacult. Soc. 18: 270–277.