Measuring the cost of reproduction

IV. Predation experiments with Daphnia pulex

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Abstract. Gravid and barren Daphnia pulex were exposed to a variety of predators in laboratory aquaria. Small fish (guppies, sticklebacks and shiner fry) consistently preferred the gravid females, establishing the existence of a behavioural cost of reproduction. However, no such cost was associated with predation by more efficient visual predators (sunfish) or by nonvisual predators (hydras), and the excess of gravid females eaten by backswimmers was found to be attributable to their distribution in the water column. Moreover, the cost associated with predation by small fish was observed only when the Daphnia were presented against a light background, and was abolished when a dark background was substituted. In a further series of experiments with guppies we attempted to show that each egg added to the brood caused a decrease in survival; in two such experiments survival rate was related to body size but not to fecundity, while in a third the effect of body size did not appear, and a negative correlation between survival and fecundity could be demonstrated. Although these experiments unambiguously demonstrate a cost of reproduction they also illustrate the elusiveness of the phenomenon and emphasize the need to develop theories which specify the type and magnitude of costs generated by different ways of life.

Introduction

The reproductive cost hypothesis states that any increase in present reproduction should be accompanied by a decrease in future reproduction, since it is only then that reproduction can be optimized through natural selection (Williams 1966). Recently, there have been attempts to identify reproductive cost by measuring the correlations among life history parameters (Snell and King 1977; Rose and Charlesworth 1981; Bell 1983, 1984a, b), or by manipulating the amount of present reproduction by various means and measuring its effect on subsequent survival or fecundity (eg. Dean 1981; Partridge and Farquhar 1982; Tallamy and Denno 1982). It might be argued, however, that the physiological responses of animals raised under favourable conditions in the laboratory do not tell us much about processes in natural populations, where a slight degradation of crypsis or escape ability might be almost inevitably fatal. For example, it has been shown experimentally that pregnant female lizards were preyed on by snake predators more than males of comparable sizes (Shine 1980), and that brightly-colored male fish were preferred by predators to dull-colored males or females (Moodie 1970, cited by Semler 1971; Haas 1976). Such studies are only suggestive; in this paper the survival of gravid Daphnia pulex is compared directly to that of barren females of equal size in simple laboratory experiments.

General methods

All the predation experiments followed the same general pattern. Predators maintained in experimental aquaria were trained to begin foraging as soon as Daphnia were poured slowly out of a small petri dish. They were then allowed to feed until about half the prey had been eaten, as assessed by an observer seated at a distance of about half a metre from the front of the aquarium. All the remaining Daphnia were removed with an aquarium net, washed into a petri dish, and then counted and classified according to the presence or absence of eggs. Female Daphnia carrying assexual eggs (gravid females) were always tested against an equal number of females without eggs (barren females). Size was measured before and after predation in order to correct for any bias due to size-selective predation. The trials were kept fairly brief (from 0.5 to 60 min, according to the predator), so that the likelihood of the animals releasing their eggs or producing a new batch during the experiment was negligible. The numbers of prey available per individual predator in each replicate and the volume of the experimental chamber were adjusted to the size and feeding rate of different predators to allow normal foraging behavior and to ensure that satiation would not occur before the end of the experiment. Predators were fed Daphnia for at least one to two weeks prior to each experiment. A controlled daily feeding regime kept the predators moderately hungry before each experimental session throughout the whole experimental period.

Daphnia pulex originating from a single clone from a S. Ontario stock was used in all the experiments. Since reproduction in these animals is highly dependent upon food availability and the state of population growth, several cultures were maintained so that both reproductive and nonreproductive females of comparable sizes could be obtained at the same time. The cultures were fed with a suspension of green algae (Ankistrodesmus) and continuously aerated. The reproductive condition of the females could
easily be assessed through the transparent carapace. To estimate size, live Daphnia were immobilized by reducing the water volume in a depression slide with a fine pipette, and their length (from base of dorsal spine to top of head) measured to the nearest hundredth of a mm using an ocular micrometer. Water was then immediately restored, taking care that no air was trapped inside the carapace, since this forces Daphnia to float on the water surface.

The results of the predation experiments, unless otherwise stated in the text, were analyzed as follows. All replicates of an experiment were pooled and tested with contingency chi-square against the null hypothesis that size has no effect on the survival of females. Pooling the results of individual trials of an experiment was considered necessary since the sample size of each trial was too small to be analyzed separately. Size effects were also tested on pooled results of experimental replicates with contingency chi-square tests under the null hypothesis that size has no effect on the survival of females. The legitimacy of pooling the results of different replicates was confirmed by performing homogeneity (contingency) chi-square tests. The significance level in all the tests performed was set at 5%.

### Experiments with small fish

(A) **Qualitative effects.** Three different species of small planktivorous fish were used in this group of experiments: guppies (Poecilia reticulata), brook stickleback (Culaea inconstans) and emerald shiner fry (Notropis atherinoides). The guppies came from a stock population kept in a small pool at McGill University for several years while the stickleback and the minnows were collected from a roadside ditch and a small lake close to Montreal respectively. Three different size classes of guppies were tested separately: small immature fish (1.0–1.1 cm in standard length), medium immature fish (1.3–1.4 cm) and adult males (1.7–2.0 cm), in order to see if small differences in the size and capture efficiency of predators affect the occurrence and strength of reproductive cost. The sticklebacks and the emerald shiners used were 1.5–2.0 cm in standard length. Depending on the size of predator, groups of 4–10 fish were tested at a time, in a 6.5 l aquarium coated with light yellow paper on three sides and the bottom. This light background was used as a first step in the experiments since it seemed to contrast well with the darker greenish color of the eggs inside the carapace. A second experiment was carried out with the medium-sized guppies using a darker, brown background in order to see if the nature of the background influenced the results. Prey density was kept constant in all experiments at 50 Daphnia per trial (7,600/m³). Special care was taken to restrict the size range of the prey as much as possible, and to make the size distributions of the different reproductive groups initially presented to the fish as nearly equal as possible, so that size would not be a confounding variable in each trial. The extreme range of body lengths of the Daphnia used in these experiments was 0.68 mm (1.64–2.32 mm), but the range of size in individual replicates of an experiment was less than 0.2 mm in almost all cases.

Table 1 summarizes the results of these predation experiments. All three species of small fish and all three size classes of guppies tested against the light background showed a preference for the gravid females. In all cases except for the shiner fry, the number of gravid females that survived predation was significantly smaller than the number of barren females. Results between replicates were homogeneous and size effects were not significant in any of the above cases. There was no significant difference in the results among different species of small fish nor among the three size classes of guppies tested. When guppies were tested against the dark background, however, no preference for the gravid females was shown. This suggests that the reproductive cost imposed by small fish is due to the increased visual contrast of gravid females against the background.

(B) **Quantitative effects.** To conclude this study an experiment was designed in order to look at the effect on survival of each additional egg carried in the brood-pouch, using medium-sized guppies in a 6.5 l aquarium against a light background. Fifty Daphnia were used in each of ten trials. After measuring the size of each animal the number of eggs it carried inside the brood pouch was estimated. If fewer than six eggs were present, all could be seen and counted directly. If more than five eggs were present, all the eggs that could be seen were counted, and to these the number of eggs that touched the upper surface of the carapace was added, this latter number being assumed to be equal to the number of eggs that were out of view. The accuracy of the estimation procedure was assessed from a sample of 75 individuals in which the number of eggs was estimated and then compared to the number counted after the eggs had been dissected out of the animals. The