# Food limitation, spination, and reproduction in *Brachionus calyciflorus*

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*Abstract*

The *Brachionus calyciflorus* polymorphism involves both a chemical secreted by the predatory rotifer *Asplanchna* and food concentration. Food concentration accounted for 30–37% of the explained variance in spine length in treatments containing *Asplanchna* factor. The longest spines developed only in low-food conditions containing the *Asplanchna* chemical, indicating that the factors were additive. Newborns exposed only to low concentrations of food during the first 3 d after hatching developed relatively long spines and smaller body sizes than their high-food-adapted mothers.

The population growth rate of the *Asplanchna*-induced phenotype was significantly higher than that of the short-spined controls over a wide range of food concentrations (0.4–15.0 μg ml⁻¹ dry mass of algae). Most importantly, elongated spines were associated with a reduced threshold food concentration for reproduction, suggesting that they return an energetic benefit exceeding their developmental and maintenance costs.

The extensive variation in spination and body size of the rotifer *Brachionus calyciflorus* was noted as early as 1850 (Hudson and Gosse 1886) and has since been the focus of numerous morphological and ecological studies (*see* Gilbert 1967; Halbach 1970). Explanations of the adaptive significance of the elongation or de novo development of posterolateral spines in the species have included decreased settling velocity (Wesenberg-Lund 1930), enhanced filtration rates (Erman 1962), and predator defense (Beauchamp 1952; Gilbert 1966; Halbach 1970).

It is now well accepted that a chemical secreted by the predatory rotifer *Asplanchna* induces *Brachionus* mothers to produce offspring with elongated movable posterolateral spines and, often, larger body sizes. These features prevent or greatly inhibit capture and ingestion by *Asplanchna* (e.g. Beauchamp 1952; Gilbert 1967, 1980*b*; Halbach 1971; Clément 1987), but the role of spines in this polymorphism is only partially understood. Before the *Asplanchna*-

factor discovery, Schneider (1937) reported that starvation or low concentrations of food induced a lengthening of posterolateral spines. Although the effect of starvation was recognized in Europe (Buchner et al. 1957; Erman 1962; Rauh 1963; Halbach 1970), its ecological significance was not understood. Interest in chemical induction and predation, furthermore, overshadowed the starvation-induced phenomenon. With rare exception (Dodson and Havel 1988), the effect of food concentration has been neglected in most subsequent examples of chemically induced polymorphism in zooplankton (Havel 1987).

The elongation of spines during starvation seems maladaptive if spines incur an energetic or reproductive cost and is, therefore, difficult to explain. Low concentrations of food, however, could be a cue of harmful interaction with *Asplanchna*; low intake of food by *Asplanchna* might reduce the amount of the chemical secreted. This argument assumes that the longer spines in *Brachionus* populations acclimated to low food protect against *Asplanchna* predation. Because spines become longer when *Asplanchna* factor is present (Halbach 1970) they must not be long enough to provide complete protection against predation. If long spines are otherwise beneficial during starvation, such as by enhancing feeding rates or reducing swimming costs, then we

should expect the defensive (long spined) phenotype irrespective of the presence of *Asplanchna* factor and food level. This expectation clearly is not met in nature or in the laboratory, suggesting that elongated spines may be beneficial in low-food conditions in the presence of *Asplanchna* but disadvantageous in high-food conditions when *Asplanchna* is absent.

The historically important experimental studies on the *Brachionus* polymorphism have been published in four languages, spanned six decades, and encompassed a variety of culture techniques—often making results difficult to compare and interpret. In light of the long history of study on this well-known polymorphism, I was motivated to redo and expand on key experiments using a single clone of *Brachionus* and a well-defined culture method. I first tested Schneider's (1937) finding that low-food conditions induced spine elongation and that low food and *Asplanchna*-factor together enhanced the degree of spine elongation as reported by Halbach (1970). Analysis of variance of these factors affecting spine length would indicate if they were additive or synergistic and would reveal their relative importance in the polymorphism. Second, I determined whether food concentration operated on spine development before or after hatching from the egg. Third, using a similar cohort life-table technique, I tested Gilbert's (1980a) conclusion that the defensive phenotype incurred no reproductive cost relative to the basic (short spined) one. Because the former study used high concentrations of food (10 and 100 $\mu g$ $ml^{-1}$), its experimental design did not actually evaluate the effect of low concentrations of food on reproduction and did not include statistical tests to support the growth-rate results. Fourth, I determined respiration rates for the phenotypes to determine whether a maintenance cost was associated with the defensive phenotype. Fifth, I assessed the threshold food requirement for reproduction between the two morphs, i.e. where the intrinsic rate of natural increase, $r_m$, is zero (Stemberger and Gilbert 1985, 1987a). If a reduced reproductive rate were associated with the defensive phenotype, it should be reflected in a higher threshold food

requirement for reproduction (Stemberger and Gilbert 1985, 1987a). On the other hand, if the threshold food requirement of the defensive phenotype is the same or lower than that of the short-spined one, then spines provide an energetic benefit that exceeds their development and maintenance costs. Lastly, I determined whether the larger bodied, defensive phenotype had a higher maximal population growth rate than the smaller, short-spined one. This relationship is predicted by the positive allometric relationship between $r_{max}$ and body size in rotifers (Stemberger and Gilbert 1985). A higher $r_{max}$ would more likely occur if the cost of producing or maintaining the spines were small.

## Methods

*Life tables*—A clone of *B. calyciflorus* was isolated from a plankton sample from the Potomac River. The phenotypes of this clone were influenced by food concentration and *Asplanchna* factor. The basic phenotype had short posterolateral spines when cultured at high levels of food without *Asplanchna* factor. A small percentage (<2%) of individuals, however, in these high-food cultures did not have posterolateral spines. *Brachionus* was fed *Cryptomonas erosa* var. *reflexa* and cultured in modified WC medium diluted 1:1 (vol:vol) with double-distilled water (Stemberger 1981; Stemberger and Gilbert 1987b). The defensive phenotype was induced with medium from *Asplanchna brightwelli* cultures containing ~1 *Asplanchna* $ml^{-1}$ (Stemberger and Gilbert 1987b). These cultures were filtered with Whatman 934-AH glass-fiber filters and the filtrate was diluted 3:1 (vol:vol) with inorganic medium.

Population growth rates were done simultaneously for the two phenotypes at each of five food concentrations (0.4, 0.6, 1.5, 8.0, and 15.0 $\mu g$ $ml^{-1}$ dry mass of algae) with a cohort life-table method (Stemberger 1988). About 75 egg-carrying females of each phenotype from high-food cultures were isolated in 5-ml Petri dishes in medium containing food at the experimental density without predator factor. Twenty-four offspring from these mothers were collected within 5 h of birth and placed into 2-ml

wells of 24-well polystyrene tissue-culture plates containing 2 ml of factor-free medium and the experimental cell density. The offspring from mothers having the defensive phenotype when raised with predator factor had relatively well-developed posterolateral spines, whereas those from mothers having the basic phenotype when cultured without the predator factor had short spines. Offspring were incubated at 20°C with a 14:10 L/D cycle. Every 24 h the mothers were transferred into fresh medium and algae. The number of newborns from each mother was recorded until all mothers of the cohort died. A potential maternal effect from well-fed mothers could increase offspring survivorship and fecundity in low food. Maternal influence should not affect life-table comparisons at a given food concentration, however, because mothers of both phenotypes were treated identically.

Population growth rates ($r_m$, $d^{-1}$) from the life-table experiments were computed iteratively using a half-interval iteration for the approximation (Euler equation) and mid-point method of Birch (1948). The mean and variance of the population growth rates were based on 200 bootstrap samples from 17 to 24 individual survivorship and fecundity schedules (Meyer et al. 1986; Stemberger 1988). Survivorship curves were analyzed with a Mann-Whitney $U$-test (Pyke and Thompson 1986).

*Morphometry*—Two sets of experiments were done to assess the effect of food concentration and predator-factor on morphology of the phenotypes. In the first experiment I tested the effect of food concentration on morphology of offspring collected from mothers cultured in predator factor and high food (6.0 $\mu$g ml$^{-1}$). After collecting 20 offspring (<3-h old), I preserved the mothers in 4% Formalin. Each newborn was placed in a 2-ml well of a 24-well polystyrene culture plate containing low food (0.6 $\mu$g ml$^{-1}$) and medium without *Asplanchna* factor. These individuals were transferred daily to fresh food and medium until they produced their first offspring, usually on the third day of life. At this time they were preserved in 4% Formalin. Measurements of the original mothers and their adult offspring were taken at 250× with a compound microscope. Student's $t$-tests were done on the means of the various body features. As adults these offspring should have a similar phenotype to that used in low-food, life-table experiments.

A second experiment was done to assess the effect of both food concentration and predator factor on morphology of *Brachionus* under four treatments—high (6.0 $\mu$g ml$^{-1}$) and low food (0.6 $\mu$g ml$^{-1}$) with and without *Asplanchna* factor. The *Asplanchna* factor was prepared in the same manner as in the life-table experiments. Ten newborns of each phenotype were placed into 15-ml polystyrene Petri dishes for each treatment. Every 2 d the population in each of the treatments was transferred to fresh medium and algae, and after 10 d the populations were preserved in 4% Formalin. Measurements were made on individuals identified as adults on the basis of an attached egg or large body size.

The morphological measurements were analyzed with a two-way ANOVA and Student-Newman-Keuls multiple range test for treatment differences (SAS Inst. 1985). The treatment factors were not correlated, as verified with Pearson correlation coefficients (SAS Inst. 1985). Therefore, each factor's contribution to the coefficient of determination ($r^2$) from the ANOVA should be about equal to the fraction of the sum-of-squares of each treatment factor divided by the total sum-of-squares. This relationship was further verified with simple linear regression on each factor to determine whether the coefficient of determination of the single factor was the same as calculated from the procedure described above: the values were in most instances identical or only slightly different.

*Respiration rates*—Oxygen consumption rates of the two morphs were done with a Clark-style oxygen electrode and a model 102B oxygen electrode amplifier modified with high-precision resistors (Instech Laboratories, Inc.). The probe was connected to a 600-$\mu$l, airtight respiration chamber maintained at 20°±0.05°C by a Brinkman and Lauda RC20 water bath. Medium used in the experiments was kept in a flask and kept in the water bath for at least 5 h before
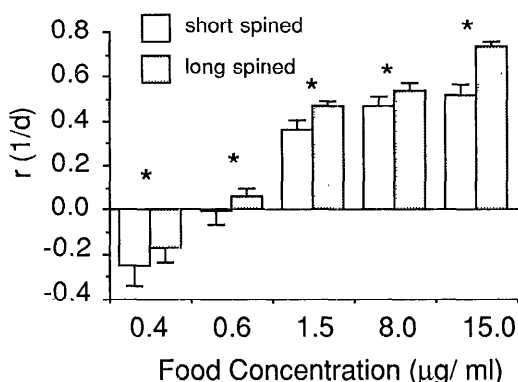
Fig. 1. Intrinsic rates of population growth ($r$, d$^{-1}$) of short- and long-spined (*Asplanchna*-induced) phenotypes of *Brachionus calyciflorus* vs. food concentration ($\mu$g ml$^{-1}$ dry mass of *Cryptomonas erosa* v. *reflexa*). Vertical bars represent $\pm 1$ SE of the mean of 200 bootstrap replicates of $r$. SE = $(S_r^2)^{1/2}$ where $S_r^2$ is the variance of the 200 bootstrap $r$-values.

the experiments. This preconditioning allowed the water to achieve the partial pressure of atmospheric oxygen. Between 30 and 45 well-fed, nonovigerous, similarly sized adult females of each phenotype from high-food cultures were placed in 5-ml, gastight glass syringes with 1.5–2.0 ml of medium and no food. Three to four control syringes without rotifers were also run. A total of 4–5 replicates was completed for each phenotype. The experimental and control treatments were submerged directly into the water bath and after 1.5–2 h of incubation the samples were injected into the respiration chamber, and the change in partial pressure from control and experimental treatments was determined. A small magnetic stirring bar in the 600-$\mu$l respiration chamber maintained a constant oxygen diffusion rate across the polyethylene membrane of the electrode. All oxygen consumption calculations were corrected to STP from ambient barometric pressure, water vapor, and altitude (Lampert 1984). Dry masses of the phenotypes were measured on a Cahn electrobalance (Stemberger and Gilbert 1985).

## Results

*Life tables*—The intrinsic rate of population growth ($r_m$) of the long-spined morph was significantly higher than that of the short-spined one in all paired life-table ex-

periments (Fig. 1). The maximal population growth rate (mean$\pm$SE) of the long-spined phenotype (0.74$\pm$0.020 d$^{-1}$, $n = 17$) was greater than that of the short-spined one (0.52$\pm$0.052 d$^{-1}$, $n = 24$) at 15 $\mu$g ml$^{-1}$ of dry mass of food. At near-threshold food concentrations (0.6 $\mu$g ml$^{-1}$), population growth rate of the induced morph (0.06$\pm$0.034 d$^{-1}$, $n = 18$) was still significantly higher ($t = -3.815$, df $= 39$, $P < 0.01$) than that of the short-spined one ($-0.004\pm0.064$ d$^{-1}$, $n = 23$) (Fig. 1). Even at the lowest concentration of food (0.4 $\mu$g ml$^{-1}$)—well below threshold levels for reproduction—the population growth rate of the long-spined phenotype ($-0.169\pm0.073$ d$^{-1}$, $n = 20$) was higher than that of the short-spined morph ($-0.253\pm0.091$ d$^{-1}$, $n = 21$) ($t = 5.752$, df $= 39$, $P < 0.01$) (Fig. 1).

The defensive morph had higher age-specific fecundity at all food levels, but there were no discernible or consistent differences in age-specific survivorship schedules ($P > 0.05$, Mann-Whitney $U$-test) (Fig. 2) between the morphs. Therefore, the higher fecundity of the spined morph was the primary factor contributing to its higher reproductive rate (Fig. 3).

*Morphometry*—After 3 d in low concentrations of food, the offspring of well-fed *Asplanchna*-induced mothers showed significant changes in body dimensions from those of their parents (Fig. 4). The posterolateral spines were ~30% longer despite the absence of predator factor in the culture medium. Furthermore, body width and length of the adult offspring were ~21 and 11% smaller than those of the mothers; therefore, as adults these offspring had considerably smaller body sizes than their mothers.

Both phenotypes also showed pronounced differences in body dimensions in experiments involving high- and low-food treatments with and without predator factor. The ANOVA of each of the morphological characteristics between the two phenotypes were all highly significant ($P < 0.001$), but the interaction term was not ($P \gg 0.05$) (Table 1). Over 50% of the total variation in the measured characters of *Brachionus* was explained by the two factors (Table 1). Food concentration explained a
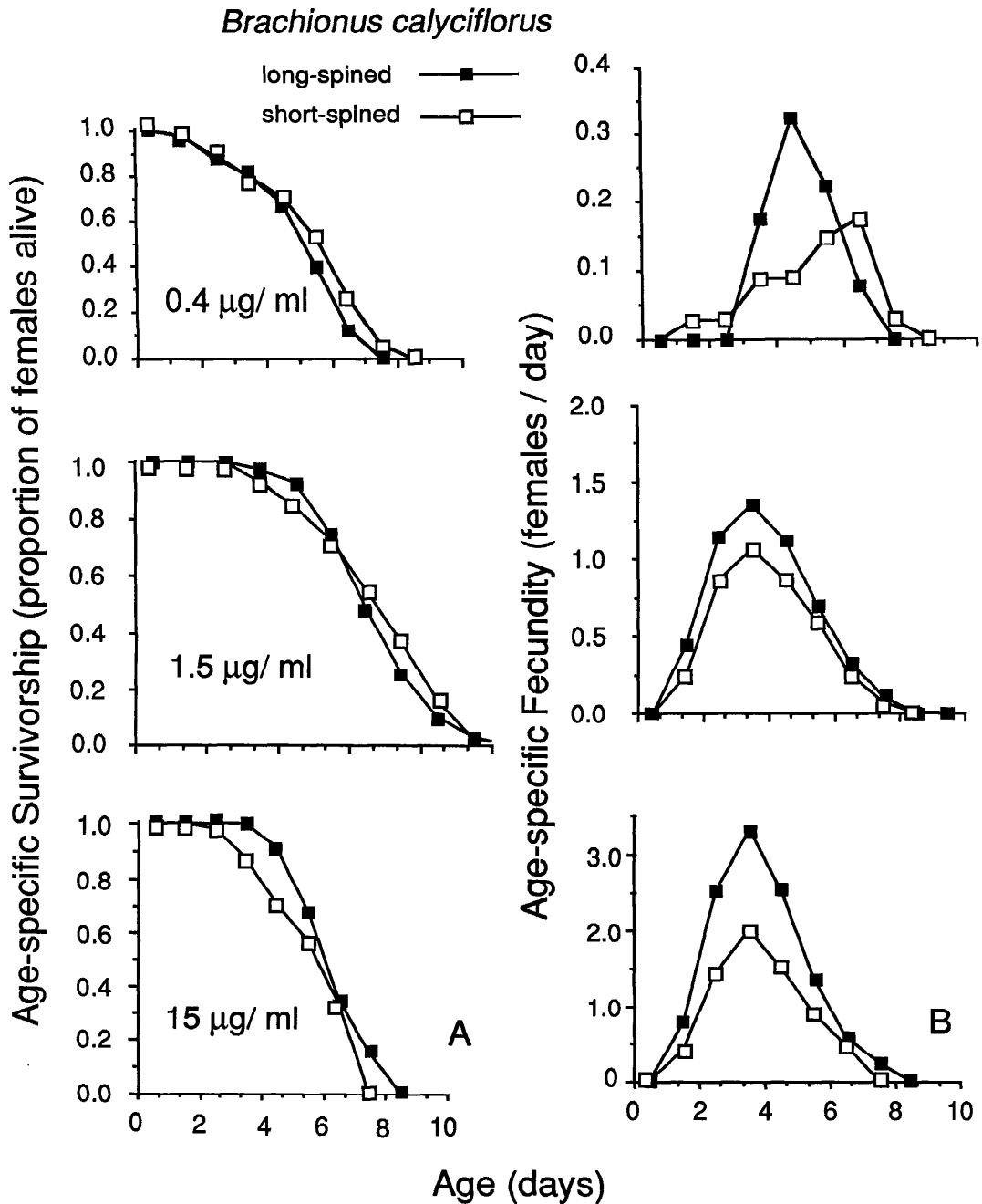
## *Brachionus calyciflorus*



Fig. 2. A. Age-specific survivorship (proportion of females alive) of *Brachionus calyciflorus* phenotypes for two food concentrations ($\mu$g ml$^{-1}$ dry mass of *Cryptomonas erosa* v. *reflexa*) vs. age (d). B. Age-specific fecundity (females d$^{-1}$) vs. age (d) of both phenotypes for the two food concentrations.

significant fraction of variation in length of posterolateral spines (Table 1). Both phenotypes had larger bodies when cultured in high-food conditions (Fig. 5). Dry masses (mean ±1 SD) of well-fed, adult, short-spined and *Asplanchna*-induced phenotypes were 0.342±0.005 and 0.519±0.029 $\mu$g ($t = -8.098$, df = 4, $P < 0.01$). Low
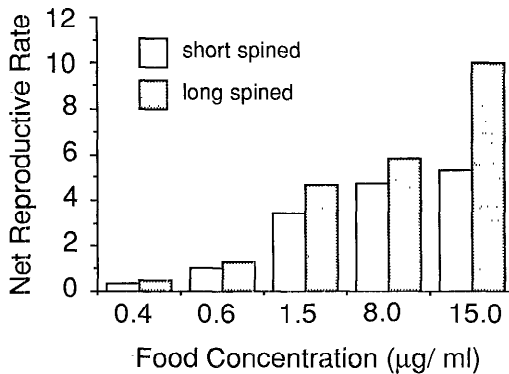
Fig. 3.   Net reproductive rate (total No. of offspring per female) of the two phenotypes for five food concentrations ($\mu$g ml$^{-1}$ dry mass of *Cryptomonas erosa* v. *reflexa*).



Fig. 4.   The length of body features of *Asplanchna*-induced mothers raised in high concentrations of food (6.0 $\mu$g ml$^{-1}$ dry mass of *Cryptomonas erosa* v. *reflexa*) and those of their adult offspring in low concentrations of food (0.6 $\mu$g ml$^{-1}$) without *Asplanchna* factor. Total length—TL; body length—BL; body width—BW; left posterolateral spine—LPS; right posterolateral spine—RPS; anteromedian spine—AMS; anterolateral spine—ALS; posteromedian spines—PMS. Vertical bars—SD of the mean ($N = 8$); NS—nonsignificant ($P > 0.05$) Student's $t$-tests of means; asterisk—$P < 0.05$.

food, with or without predator factor, produced individuals having significantly smaller body widths (a factor of ~15–20%)—a result consistent with experiment 1 (Figs. 5, 6) and supported by the large contribution (~69%) of this factor to the coefficient of determination (Table 1). Body length of the *Asplanchna*-induced phenotype, however, did not significantly decrease in low food as it did in experiment 1 (Figs. 4, 5); despite the difference levels, the relative decrease was similar in both experiments—11 vs. 8% reduction in body length for experiments 1 and 2. Food concentration contributed 23% of the explained variance in body length but did not significantly affect anterolateral, anteromedian, and posteromedian spine lengths (Table 1, Fig. 5).

The posterolateral spines of both phenotypes showed very striking changes in length as a function of food concentration and predator factor. The greatest absolute lengths of posterolateral spines occurred in cultures having predator factor and low food. The right and left posterolateral spines were 31 and 35% longer in low food with predator factor than in high food with the factor. The most pronounced relative changes in spine lengths occurred in treatments without predator factor—low food produced phenotypes having right and left posterolateral spine lengths 56 and 63% longer than those in high food.

There was considerable variation in the posterolateral spines of the short-spined
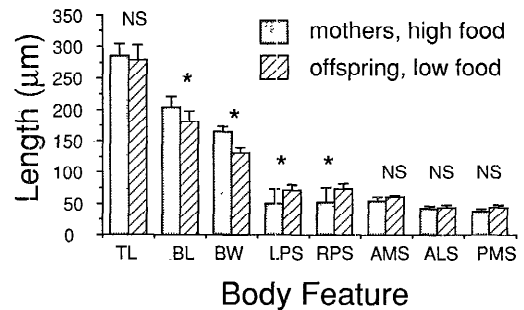
phenotype. Some individuals living in high-food cultures without predator factor lacked posterolateral spines altogether. All individuals living in low food without predator factor, however, had posterolateral spines. Despite the relatively large increase in posterolateral spine length, total length decreased ~14%. This decrease is partially explained by the significant decrease in body length and anteromedian spine length in low food without predator factor (Fig. 5).

*Respiration rates*—The respiration rate (mean ±1 SD) of the *Asplanchna*-induced

Table 1.   The contribution of food concentration (F) and *Asplanchna* (A) factor to the coefficient of determination ($r^2$). Factor contribution was calculated as the fraction of each treatment factor's sum-of-squares divided by the total sums-of-squares in the two-way ANOVA. All factors are significant at $P < 0.01$ unless indicated otherwise and are uncorrelated. (ns—$P > 0.1$.)

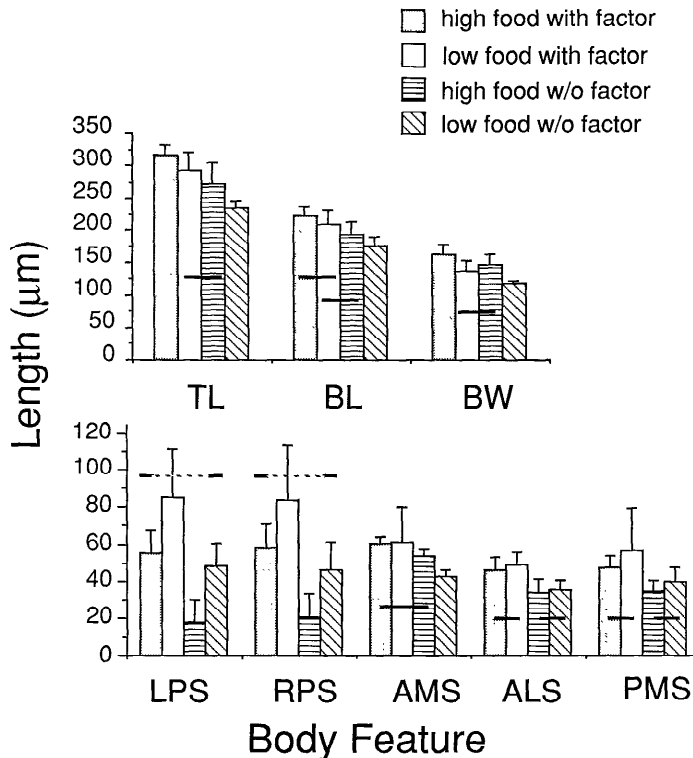| Body feature* | F | A | $\Sigma r^2$ | Relative contribution of F to $\Sigma r^2$ (%) |
|---|---|---|---|---|
| BW | 0.40 | 0.18 | 0.58 | 68.9 |
| LPS | 0.24 | 0.40 | 0.64 | 37.4 |
| RPS | 0.17 | 0.40 | 0.57 | 29.8 |
| TL | 0.18 | 0.46 | 0.64 | 28.1 |
| BL | 0.12 | 0.40 | 0.52 | 23.1 |
| ALS | 0.01 ns | 0.49 | 0.50 | 2.0 |
| AMS | 0.07 ns | 0.41 | 0.58 | 12.0 |
| PMS | 0.04 ns | 0.26 | 0.32 | 15.3 |

* Symbols same as Fig. 4.

Fig. 5. The mean length of body features ($N$ = 10) of populations of short-spined and *Asplanchna*-induced phenotypes. Shaded and open bars—high- (6.0 $\mu$g ml$^{-1}$) and low-food (0.6 $\mu$g ml$^{-1}$) treatments of cultures containing *Asplanchna* factor; striped and hatchet bars—high- and low-food treatments without *Asplanchna* factor. Symbols as in Fig. 4. Histograms connected by horizontal lines are not significantly different at $P < 0.05$ with Student-Neuman-Keuls multiple range test.

phenotype (2.33±0.43 $\mu$l $O_2$ ind$^{-1}$h$^{-1}$, $n$ = 5) was 24.3% higher than that of the short-spined one (1.76±0.57 $\mu$l $O_2$ ind$^{-1}$h$^{-1}$, $n$ = 6) ($t$ = $-1.886$, df = 9, $P < 0.05$). These rates are not unusual and are similar to rates reported for rotifers having similar body sizes (Starkweather 1987).

*Discussion*

This study clarifies and elaborates upon earlier studies and presents new findings on the effect of food concentration on spination and reproduction in a polymorphic clone of *B. calyciflorus*. Several lines of evidence support the beneficial effects of long posterolateral spines in low-food conditions. The threshold food concentration for reproduction of the long-spined phenotype was reduced relative to the short-spined one, suggesting that spines return an energetic benefit that exceeds their developmental and

maintenance costs. The higher reproductive rate of the long-spined phenotype in high-food conditions was attributed to its larger body size. Low food induced significant spine elongation even in the absence of predator factor, and the induction occurred during the first 3 d of life in short-spined offspring hatched from well-fed, short-spined mothers. The phenotype of the adult of these offspring was clearly different from that of the mothers. The consistency and rapidity of these responses establish low concentrations of food, in addition to the *Asplanchna* chemical, as a major factor in the evolution and maintenance of this polymorphism. About 30–37% of the explained variance in posterolateral spine lengths was attributed to food concentration alone—a very striking result that confirms the observations of earlier European investigators. The effect of low food may also explain the
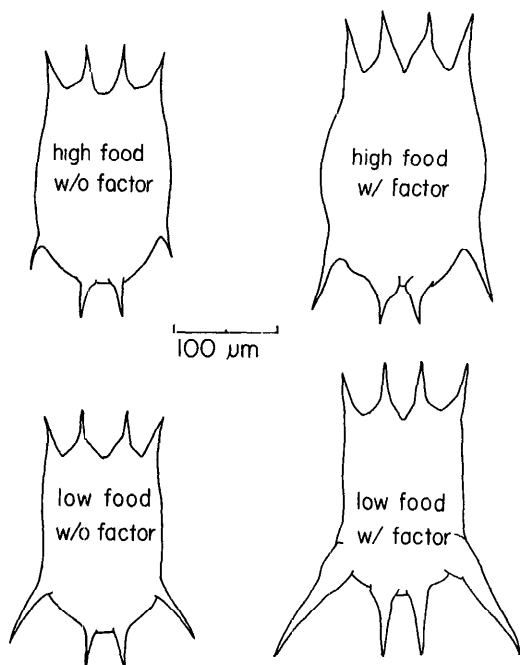
Fig. 6. Dorsal view of typical morphs produced from experimental treatments designated in Fig. 5 from high- and low-food cultures with and without *Asplanchna* factor.

apparent spontaneous development of long-spined forms from the short-spined phenotype in old laboratory cultures without the *Asplanchna* chemical (Stemberger pers. obs.).

*Life tables*—The higher reproductive rate of the *Asplanchna*-induced phenotype at all food levels (Fig. 1) was attributed to its higher fecundity and not to differences in survivorship relative to the short-spined morph (Fig. 2). The maximal population growth rate of the larger bodied defensive phenotype (~34% greater dry mass) at high concentrations of food was consistent with the positive allometric relationship between $r_{max}$ and body size in rotifers (Fig. 1) (Stemberger and Gilbert 1985) and consistent with the population growth-rate trends reported by Gilbert (1980a) for the species in high-food conditions.

The lower threshold food requirement for reproduction of the *Asplanchna*-induced morph was opposite to that predicted from the relationship between threshold food concentration and body size in rotifers

(Stemberger and Gilbert 1985). These results may be explained if body size of the defensive morph were reduced relative to the short-spined one—the defensive phenotype was considerably larger (Fig. 5)—or, alternatively, the posterolateral spines improved hydrodynamic characteristics, such as reducing energy costs for swimming or increasing food intakes rates. Any hydrodynamic benefits must exceed the developmental and maintenance costs of producing spines. The oxygen consumption rates of phenotypes adapted to high food, furthermore, suggest that spines are not costly to maintain.

The reproductive rate of the defensive phenotype of the polymorphic species *Keratella testudo* was indistinguishable from the unspined one near threshold food concentrations (Stemberger 1988). At high concentrations of food, however, its reproductive rate was only half that of the unspined one. The spined phenotype also had a higher (~27%) coefficient of skin friction (i.e. the ratio of the sinking rate of a sphere having the same volume and specific gravity as that of the rotifer to the sinking rate of the rotifer) than the unspined one in low- but not in high-food conditions. Because *K. testudo* has a rigid and thicker lorica than *Brachionus*, its denser spines should have a greater developmental or maintenance cost. Consequently, differences in reproductive rates of the phenotypes in low-food conditions were not as large as those observed in *Brachionus*.

*Morphological responses*—The effect of low food on morphology of newborns was striking and occurred in the first 3 d of life: young adults had much longer posterolateral spines and smaller body sizes than their well-fed mothers. This new finding on the juvenile development contrasts sharply with the maternal effect on development in the presence of *Asplanchna* factor (Gilbert 1967). This study clearly shows that both factors are independent and additive with respect to spine elongation.

Other body spines were significantly affected by the chemical factor but not by food concentration (Fig. 5, Table 1). The lower threshold food requirement of the predator-induced morph coincided with elongation

of the posterolateral spines and concomitant reduction in body size. Food concentration had the strongest effect on body width (Table 1) but may represent distention of the body by the volume of food in the gut, growing eggs, and storage products in the germovitellarium, as well as actual changes in body dimensions (Stemberger 1988).

The hydrodynamic effect of spines—Spines differentially influence hydrodynamic characteristics depending on food conditions. Longer, more massive spines, for example, increase sinking rates in several ways. They reduce swimming speeds by increasing drag, increase body mass and specific gravity, and shift the center of gravity posteriorly (Stemberger 1988). These effects should be most pronounced in hard-bodied rotifers because the lorica has relatively high mass and high specific gravity. High concentrations of food also increase specific gravity, shift the center of gravity posteriorly, increase mass, and change the orientation of the body during sinking (Hutchinson 1967; Stemberger 1988). These factors tend to increase sinking rates and reduce swimming speeds. The spined phenotype of K. testudo, for example, has a larger body size than the unspined form and a heavier lorica due to the dense posterior spines (Stemberger 1988). In high-food conditions these weight-dependent factors exceed the effect that the drag of long spines has on reducing sinking rates. Sinking rates, however, are similar for both phenotypes in low food where food-dependent effects on mass and density are less (Stemberger 1988). All factors tending to increase sinking rates should be very important because of the limited power output from the cilia used for swimming and because of the apparently high energetic costs associated with ciliary locomotion in the transformation of chemical energy into kinetic energy (Epp and Lewis 1984).

Erman (1962) observed an inverse relationship between spine length and food concentration and hypothesized that the longer spines in low-food conditions increased drag, allowing the rotifer to filter a greater volume of water than it could if it were to swim at a faster rate without spines. He did not, however, propose a clear mechanism. Subsequent studies did not support his hypothesis (see Bogdan and Gilbert 1982). Bogdan and Gilbert (1982) also refuted Erman's hypothesis; they used well-fed rotifers from high-food cultures and acclimated to low cell densities for 2.5 h or less. Differences in sinking rates and swimming speeds could not be detected between phenotypes of K. testudo for at least 6 h of acclimation in low-food conditions (Stemberger 1988). Therefore, differences in clearance rates attributable to the spines of Brachionus would not be easily detected without a longer acclimation period. Consequently, Erman's hypothesis still provides a viable explanation for the growth-rate advantage observed for the long-spined phenotype. The effect may also explain the similarity of reproductive rates of phenotypes of K. testudo in low-food treatments (Stemberger 1988).

One might expect posterolateral spines to be longest in high-food conditions, particularly if a developmental or maintenance cost is associated with them. The inverse relation between spine length and food concentration suggests that spines may be highly beneficial for reasons other than protection against predation. Elongated spines cannot be so costly to produce and maintain because they are associated with higher reproductive rates in low-food conditions. The respiration rates for well-fed phenotypes cannot be used to infer respiration rate differences at threshold food concentrations. They suggest, however, that the soft, flexible spines of Brachionus, at least, are not costly to maintain.

A reduction of posterolateral spines in high-food conditions is also contradictory with respect to potential hydrodynamic benefits: long spines should increase drag and reduce sinking rates. This paradox may involve a tradeoff between drag and motility. In high-food conditions swimming speeds are substantially reduced because of increased body mass and specific gravity (Stemberger 1988). The limited power produced by the coronal cilia may be insufficient to offset the additional drag of long spines, causing further and substantial reductions of swimming speed. Consequently, a well-fed, long-spined rotifer may not have adequate swimming speed to maintain depth position, making it prone to sinking out of

the phototrophic zone where adverse conditions may exist. Natural selection should favor morphology which preserves motility.

Females carrying attached eggs sink faster than females without them (Stemberger 1988). In high-food conditions natural selection should favor features permitting high reproductive rates (large body size and high egg ratio) even when they contribute to faster sinking rates. On the other hand, features that may increase sinking rates but do not contribute to reproduction, such as large posterior spines having high specific gravity, should be selected against. In contrast, in low-food environments morphological features reducing sinking rates should be favored because they help reduce the threshold food requirement for reproduction. Short-spined phenotypes carrying one or two attached eggs provide protection against *Asplanchna* predation over those without eggs (Gilbert 1980*b*). This benefit may represent an additional evolutionary factor favoring reduced posterolateral spine lengths in high-food conditions where the probability of carrying one or more eggs is high.

At high concentrations of food, the larger body sizes of both phenotypes permit higher population growth rates (Stemberger and Gilbert 1985). The shorter posterolateral spines of the predator-induced phenotype in high-food treatments are probably sufficiently long to protect against *Asplanchna* predation and, simultaneously, small enough to minimize their effect on increasing sinking rates due to weight and drag factors. Also, body size of *Brachionus* significantly increases in high-food conditions when *Asplanchna* factor is present. Consequently, large body size should provide additional protection against *Asplanchna* predation and further reduce the need for long spines. Natural selection should favor spine lengths not exceeding those required for protection against predation. Therefore, reduced spine lengths in high-food conditions also are consistent with a hydrodynamic explanation.

*Implications for rotifer community structure*—The inverse relationship between food concentration and posterolateral spine lengths in *Brachionus* helps interpret patterns of spination and body design of other species. The potential effect of faster sinking rates of spined forms in high-food conditions may explain why *Keratella cochlearis* often lacks a posterior spine (f. *tecta*) in eutrophic environments whereas in mesotrophic and oligotrophic waters the spined form (f. *typica*) prevails (Hillbricht-Ilkowska 1972; Gannon and Stemberger 1978; Eloranta 1982). *Kellicottia longispina* has the longest body spines of any planktonic rotifer and is an important species of oligotrophic lakes in temperate regions (Gannon and Stemberger 1978). The very slow sinking rate ($\sim 0.05$ mm s$^{-1}$, 20°C) of this species is attributed to the high drag of the spines. For comparison, *K. cochlearis* has only $\frac{1}{10}$ the body mass but nearly twice the sinking speed ($\sim 0.09$ mm s$^{-1}$) of the former (Stemberger unpubl.).

Other dominant patterns of body design that occur among planktonic rotifers may also provide beneficial hydrodynamic effects. For example, the dorsoventrally flattened bodies of *Notholca* have high surface-to-volume ratios which should increase drag and decrease sinking rates. *Notholca foliacea*, a prominent rotifer of oligotrophic temperate lakes (Stemberger 1976), has a keellike midrib projection along the ventral lorica plate which could increase drag and stabilize the center of gravity. The mucus sheaths of *Ascomorpha, Collotheca, Conochiloides,* and *Conochilus* may reduce specific gravity and improve suspension. Other features such as the gelatinous spiral tube extending from the toe of *Trichocerca multicrinis* may influence both drag and buoyancy.

In conclusion, spine development in this clone of *B. calyciflorus* is mediated by both chemical and resource concentration factors. The first provides protection against predators and the second reduces the threshold food concentration for reproduction—an important advantage in food-limited environments. Food concentration influences this polymorphism during the juvenile growth stages. Spines may reduce threshold food concentration by reducing sinking rates—an energetic benefit related to reduced swimming costs—or by increasing filtration rates. The latter benefit might involve a direct increase in the volume of

water filtered or an increase in the efficiency of particle collection. Long spines did not develop in high food for several possible reasons. The potential effect of long spines on enhancing filtration rates is irrelevant or trivial when food is abundant. The large body size and attached eggs of the *Asplanchna*-induced, high-food-adapted phenotype give adequate protection against *Asplanchna* predation so there is no selection for long spines. Long spines may be detrimental in high concentrations of food because the power output of the locomotory apparatus, already taxed by high sinking rates, cannot compensate for the additional drag. Diminished swimming speed and loss of motility might cause the rotifer to sink out of food-rich strata or impair its ability to locate new food patches.

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