# EFFECT OF CHLORELLA DENSITY AND TEMPERATURE ON SOMATIC GROWTH AND AGE AT MATURITY OF THE ROTIFER BRACHIONUS PATULUS (MULLER) (ROTIFERA)

#### S. S. S. SARMA

Department of Zoology, University of Delhi, Delhi 110 007, India.

Present address: Department of Animal Physiology, School of Biological Sciences, Madurai Kamaraj University,

Madurai 625 021, India.

#### **ABSTRACT**

The effects of three food levels (1, 2 and  $4 \times 10^6$  cells/ml of Chlorella) and three temperatures (15, 25 and 35°C) on somatic growth and age at maturity of Brachionus patulus were investigated. Both growth rate and maximum body size reached were significantly higher at higher food levels. The effects of temperature alone on the adult body size did not show any clear trend. Higher food and temperature levels significantly reduced the age at maturity.

### INTRODUCTION

Morphological and life history parameters of rotifers, such as body size<sup>1,2</sup> and age at maturity<sup>3</sup>, have been considered as products of natural selection and thus best adapted to their environment. However, the extent to which these parameters vary in rotifers in nature is often difficult to predict. For example, in Keratella cochlearis, when the morphometry was plotted as a function of altitude, marked differences appeared between the populations of the two different localities studied<sup>4</sup>. It is therefore only through experimentation that some conclusions can be drawn. In the present study the effects of food and temperature levels on the somatic growth and age at maturity of the rotifer Brachionus patulus (Muller) have been investigated.

## MATERIALS AND METHODS

The general rotifer culture method was described earlier<sup>5</sup>. In the present study three food levels (1, 2 and  $4 \times 10^6$  cells/ml of *Chlorella* sp.) and three temperatures (15, 25 and 35°C) were employed. For somatic growth studies 25-ml beakers were used as containers. Into each of 18 beakers (3 food levels ×3 temperatures × 2 replicates = 18) containing 20 ml of specified food density were introduced *B. patulus* neonates within 2 h of hatching at a density of 20 individuals/ml. The neonates were obtained as described earlier<sup>6</sup>. Test beakers were maintained in three temperature-controlled water baths set at 15, 25 and 35°C. At the start of the experiment and after

every 12 h 10-15 individuals picked at random were removed from each test beaker and killed with dilute formalin. Morphometric measurements were determined as in earlier studies<sup>6</sup>. The experiment was discontinued when the individuals reached maturity, which was indicated by the appearance of ovigerous females.

To determine age at maturity (= age at first reproduction or AFR) 15-ml glass cavity blocks were used. The food density and temperature combinations used in the somatic study were also employed here. Into each of 27 cavity blocks (3 food levels × 3 temperatures × 3 replicates) 50 neonates were introduced. At the start and after every 2±0.5 h interval each individual in each cavity block was examined and the ovigerous females were counted and removed. The non-ovigerous females were transferred to fresh medium of appropriate food density and temperature after every 12 h. At 15°C, as the time to attainment of maturity was very long (120 h), observations were made at 12 h intervals only.

# **RESULTS AND DISCUSSION**

Many factors appear to influence body size of rotifers. These include genotype<sup>7</sup>, predation<sup>8,9</sup>, salinity<sup>10</sup> and food quality<sup>11</sup>. However, within a clone of rotifer species the most significant factors influencing body size are food density<sup>6,12</sup> and temperature<sup>13,14</sup>. Somatic growth in *B. patulus* occurred only till AFR. No measurable growth in body size (lorica area) occurred beyond AFR regardless of food density and temperature (figure 1).

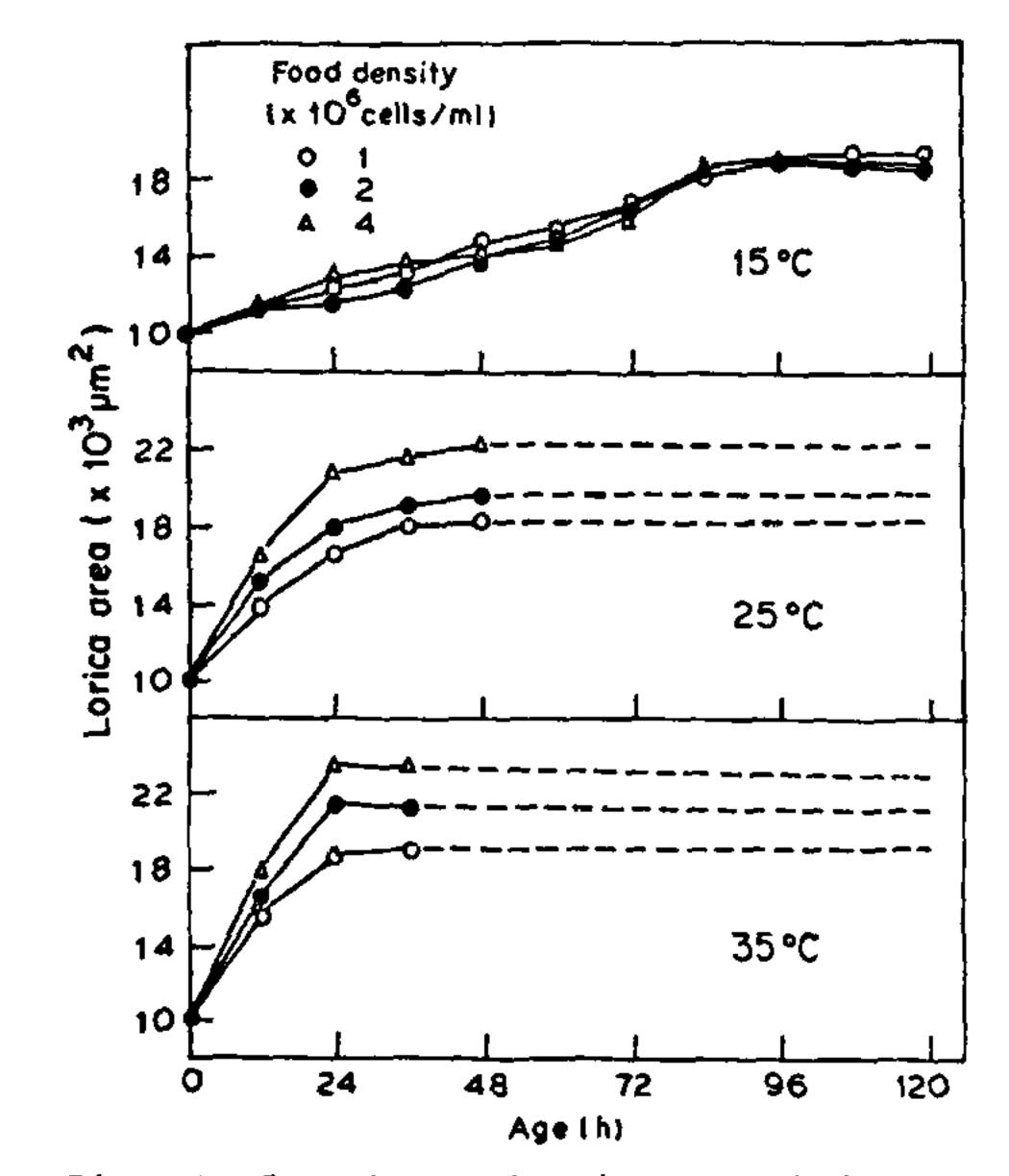


Figure 1. Somatic growth as increase in lorica area of B. patulus in relation to food density and temperature. Each point represents mean of 10-25 measurements from each of the two replicates. Broken lines indicate no measurable growth in lorica area.

The growth rate up to AFR was significantly influenced by both food density and temperature. The effect of food density-temperature interaction was also significant. Both growth rate and maximum body size reached were significantly higher at high food density and temperature. Significant differences in body size of rotifers grown at different food density-temperature combinations were evident as early as 12 h after hatching (table 1). The highest specific growth rate (maximum increase in lorica area/time (h) taken to reach that size) at  $35^{\circ}$ C and  $4 \times 10^{6}$  cells/ml food density was nearly seven times higher than that achieved at  $15^{\circ}$ C at the same food level (figure 2).

The period from hatching to age at maturity is generally subjected to maximum environmental influence<sup>9</sup>. It is only during this period that somatic growth takes place because of active feeding<sup>15</sup>. Since all cell divisions are completed before hatching, the somatic growth of rotifers is mainly because of increase in cell size due to incorporation of assimilated material<sup>16</sup>. It is therefore reasonable to predict the development of larger individuals under optimal food density. The somatic growth rate in rotifers may be measured in terms of lorica area<sup>6</sup> or dry weight<sup>17,18</sup>. The measurement of lorica area as an index of rotifer size may be more reliable as it is independent of the quantity of food present in the

**Table 1** Effect of food density and temperature on somatic growth and age at maturity of B. patulus: Two-way analysis of variance

|                      | Source of                        |    |        |        |                    |
|----------------------|----------------------------------|----|--------|--------|--------------------|
| Parameter            | variation                        | đf | SS     | MS     | F                  |
| Specific growth rate | Food density                     | 2  | 63724  | 31862  | 115.7**            |
|                      | Temperature                      | 2  | 475906 | 237953 | 864**              |
|                      | Food density × temp. interaction | 4  | 40644  | 10161  | 36.9**             |
|                      | Error                            | 9  | 2478   | 275    |                    |
| Growth rate at 12 h  | Food density                     | 2  | 9.75   | 4.87   | 99.47**            |
|                      | Temperature                      | 2  | 96.80  | 48.40  | 987.96**           |
|                      | Food density × temp. interaction | 4  | 5.98   | 1.49   | 30.53**            |
|                      | Error                            | 9  | 0.44   | 0.04   |                    |
| Growth rate at 36 h  | Food density                     | 2  | 23.68  | 11.84  | 95.48**            |
|                      | Temperature                      | 2  | 216.90 | 108.45 | 894.60**           |
|                      | Food density × temp. interaction | 4  | 10.04  | 2.51   | 20.24**            |
|                      | Error                            | 9  | 1.12   | 0.12   |                    |
| Age at maturity      | Food density                     | 2  | 194    | 97     | 8.12*              |
|                      | Temperature                      | 2  | 48375  | 24188  | 2026**             |
|                      | Food density × temp. interaction | 4  | 79     | 19.75  | 1.65 <sup>ns</sup> |
|                      | Error                            | 18 | 215    | 11.94  |                    |

<sup>\*\*</sup>Highly significant P < 0.001; \*Significant P < 0.01; \*Non-significant P > 0.05.

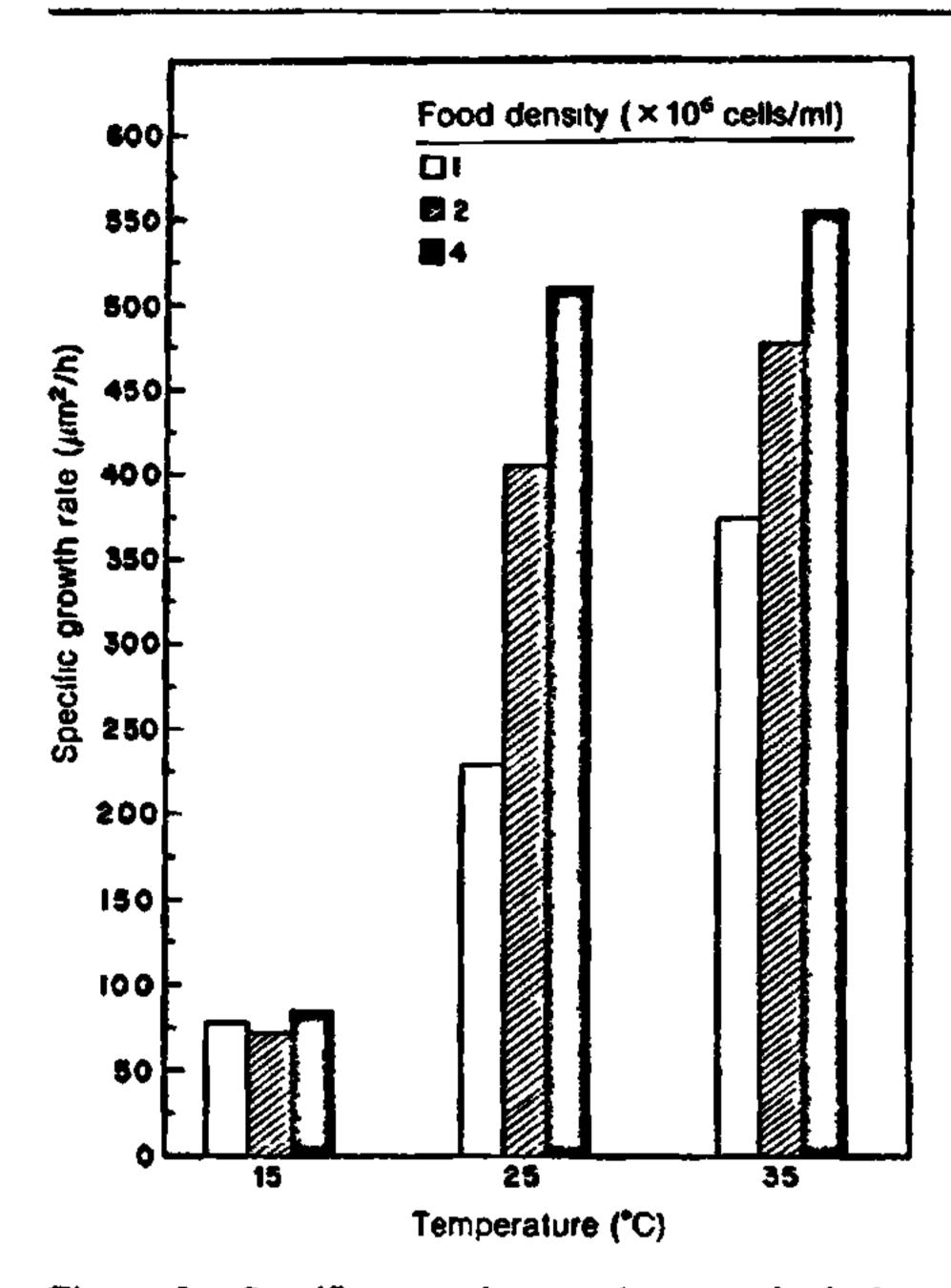


Figure 2. Specific growth rate (increase in lorica area/h) of *B. patulus* under different food density—temperature conditions. Values shown are based on 10-25 measurements from each of the two replicates.

gut. On the other hand, if rotifer body size is measured in terms of dry weight, the physiological state of the animals, such as nutritional status, stage of developing embryo within female body, etc., which are usually difficult to define, should be specified.

The growth patterns recorded in the present study are similar to those reported by others 16.19 in that the maximum possible growth was accomplished soon after hatching (except at 15°C) and increase in food level resulted in increased lorica area 20. The effect of temperature alone on the adult body size was not clear from those results and probably prolonged exposure to a particular temperature may be needed 10. Rotifers collected in winter had larger loricae than those collected in summer 21.22. However, other factor such as predation 9 may also influence rotifer body size in nature. The increase in body size with increase in food density was evident even after age at maturity (figure 1). There is some evidence

that excessive food results in decreased body size in rotifers. Pılarska<sup>12</sup> noticed that the largest juveniles and mature *B. rubens* were those grown at optimal food conditions; there was a general decrease in body size under both low and high food levels.

The differences in growth rates of rotifers kept under different test conditions were also reflected in age at maturity. In general both food level and temperature had significant effect on AFR (table 1). Higher food and temperature levels contributed to reduced AFR, i.e. maturity was reached earlier. The AFR was highest (5 d) at 15°C and low food level and smallest (0.9 d) at 35°C under high food level (figure 3). Reduction in AFR with increasing food availability, as observed in B. patulus, was reported earlier in two other rotifer species, Asplanchna girodi<sup>23</sup> and Euchlanis dilatata<sup>16</sup>. Similarly reduced AFR at higher temperature were also recorded in Asplanchna brightwelli<sup>24</sup>, B. calyciflorus<sup>25</sup> and B. dimidiatus<sup>26</sup>.

The food levels chosen in the present study were within the range used for other rotifers<sup>27</sup>. The

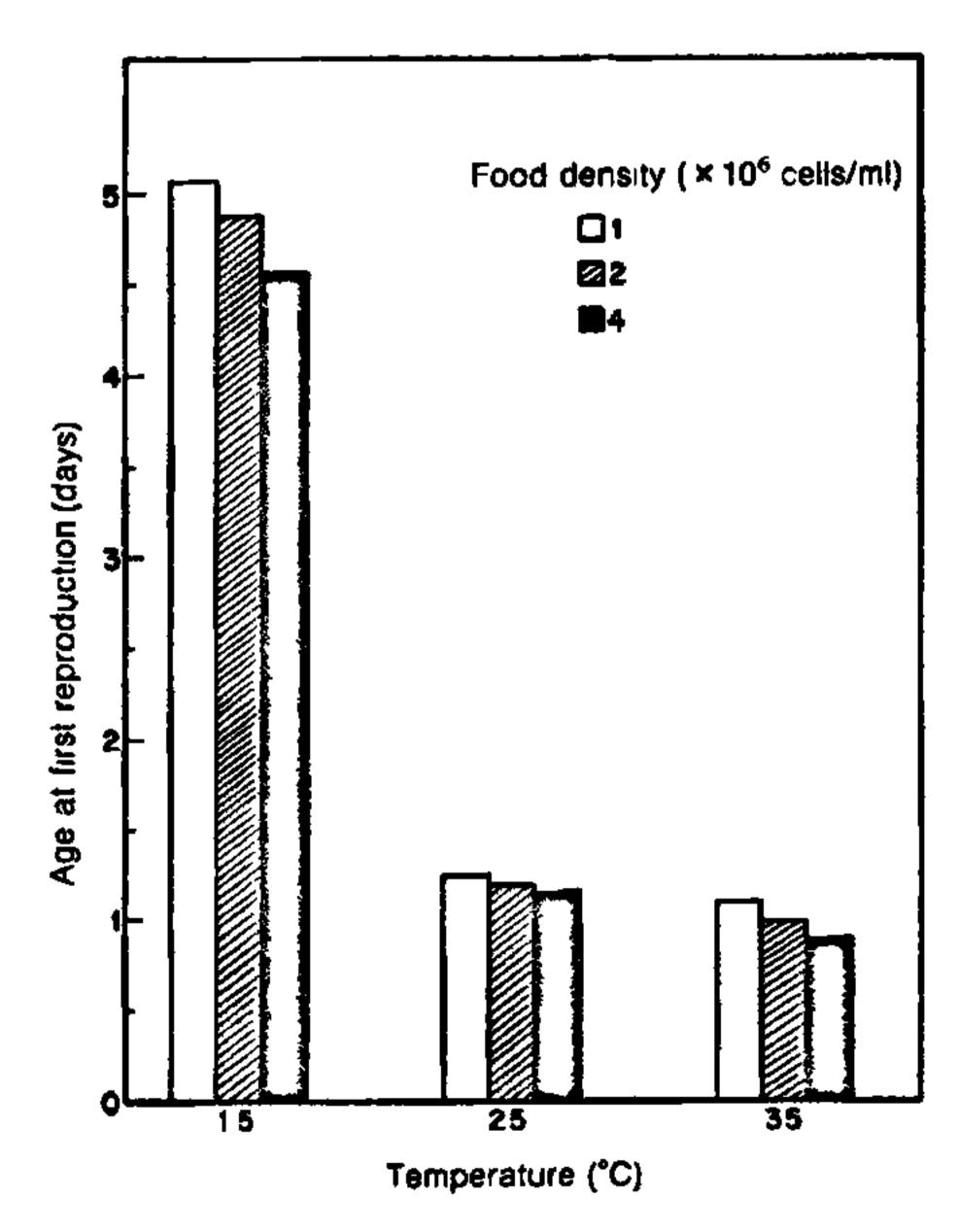


Figure 3. Influence of food density and temperature on age at maturity of *B. patulus*. Fifty neonates were employed for each food density—temperature combination.

temperatures employed here (15-35°C) are in the range that *B. patulus* experiences in nature<sup>28</sup>. However, temperatures higher than 35°C showed marked contrastive effects on rotifers. For example, in *B. calyciflorus* and *B. urceolaris*, increase of temperature to 40°C actually prolonged the juvenile phase, although in no regular manner<sup>27</sup>.

In freshwater ecosystems rotifers in a variety of habitats are just as much successful as cladocerans and copepods. What copepods to a large extent and cladocerans to a certain extent achieve through their high fecundity, rotifers achieve through their quicker maturing (earlier age at first reproduction)<sup>3</sup>. The large seasonal and geographical variations in the dynamics of many rotifer species may be at least partly explained by the effects of food density and temperature on body size and age at maturity.

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- 1. Peiler, B., Hydrobiologia, 1980, 73, 207.
- 2. Bogdan, K. G. and Gilbert, J. J., Proc. Nat. Acad. Sci. USA, 1984, 81, 6427.
- 3. Allan, J. D., Am. Nat., 1976, 110, 165.
- 4. Green, J., Hydrobiologia, 1987, 147, 3.
- 5. Rao, T. R. and Sarma, S. S. S., Curr. Sci., 1985, 54, 499.

- 6. Sarma, S. S. S., Bull, Bot. Soc. Sagar, 1985, 32, 54.
- 7. Sudzuki, M., Hydrobiologia, 1987, 147, 45.
- 8. Pourriot, R., Ann. Hydrobiol., 1974, 5, 43.
- 9. Duncan, A., Hydrobiologia, 1984, 113, 201.
- 10. Serra, M. and Miracle, M. R., *Hydrobiologia*, 1987, 147, 83.
- 11. Yufera, M., Hydrobiologia, 1987, 147, 319.
- 12. Pilarska, J., Pol. Arch. Hydrobiol., 1977, 24, 329.
- 13. Halbach, U., Oecologia, 1970, 4, 262.
- Galkovskaya, G. A., Zh. Obshch. Biol., 1979, 40, 734.
- 15. Pourriot, R., Ann. Hydrobiol., 1973, 4, 103.
- 16. King, C. E., Ecology, 1967, 48, 111.
- 17. Scott, A. P. and Baynes, S. M., Aquaculture, 1978, 14, 247.
- 18. Leimeroth, N., Hydrobiologia, 1980, 73, 195.
- 19. Snell, T. W. and Carrillo, K., Aquaculture, 1984, 37, 359.
- 20. Sarma, S. S. S. and Rao, T. R., Arch. Hydrobiol., 1987, 111, 245.
- 21. Serra, M. and Miracle, M. R., *Hydrobiologia*, 1983, 104, 279.
- 22. Hillbricht-Ilkowska, A., Hydrobiologia, 1983, 104, 297.
- 23. Robertson, J. R. and Salt, G. W., Ecology, 1981, 62, 1585.
- 24. Verdone-Smith, C. and Enesco, H. E., Exp. Gerontol., 1982, 17, 255.
- 25. Halbach, U., Oecologia, 1970, 4, 174.
- 26. Pourriot, R. and Rougier, C., Ann. Limnol., 1975, 11, 125.
- 27. Galkovskaja, G. A., Hydrobiologia, 1987, 147, 307.
- 28. Sarma, S. S. S., Unpublished thesis, University of Delhi, Delhi, 1987.