

A STUDY ON THE FEEDING RESPONSES OF A FILTER - FEEDING *CYCLOPS* SP. ON VARIOUS CONCENTRATIONS OF *CHLORELLA VULGARIS*

M M M Alam ^a, M I Miah ^a and M A B Habib ^{b*}

^aDepartment of Fisheries Management, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh

^bDepartment of Aquaculture, Faculty of Fisheries, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh

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The feeding responses of *Cyclops* sp. fed on *Chlorella vulgaris* at various concentrations were studied. The filtration rate of *Cyclops* ranged from 3.0 to 6.43 $\mu\text{l/h}$ /individual. The range of food concentrations was 515 to 2039 cells/ μl . The critical food concentration was 1435 cells/ μl . The ingestion rate was recorded a maximum of 9971 cells/h/individual at the concentration of 771 cells/ μl . This study also relates filtration rate with pH and dissolved oxygen.

Key words: *Cyclops* sp., *Chlorella vulgaris*, Feeding responses.

Introduction

The filter feeders especially rotifers are playing significant role as live food organisms in the food chain of fishes starting as starter food (Lubzens 1987; Tidwell *et al* 1997). These rotifers depend on the microalgae for their proper growth, development and reproduction (Maruyama 1993; Schwarz *et al* 1995; Habib 1998). Microalgae can biosynthesize these essential nutrients from the inorganic nutrient media or natural environment (Habib *et al* 1997; Vazhappilly and Chen 1998) which may be taken by rotifers after feeding them (Bennet and Boraas 1988; Hirayama *et al* 1989; Maruyama 1993; Habib 1998). Among microalgae, *Chlorella* sp., *Ankistrodesmus* sp., *Selenastrum* sp. etc. can successfully biosynthesize essential nutrients from the media (Duncan 1989) which are available in the ponds of Bangladesh Agricultural University Farm, Mymensingh. Among zooplankton, *Brachionus* sp., *Cyclops* sp., *Keratella* sp. etc. are found dominant in these farm ponds. These microalgae are smaller in size than others (Thompson *et al* 1988) and may be used to feed above zooplanktons after isolation. Therefore, the present work was undertaken to study the feeding responses of *Cyclops* sp. to different concentrations of *Chlorella* sp.

Materials and Methods

The samples of phytoplankton were collected from the ponds of Bangladesh Agricultural University (BAU) fish farm by plankton net of mesh size 30 μm . The cells of *Chlorella vulgaris* (isolate no. DAQ001) were isolated with the help of microcapillary pipette through different dilutions under microscope. Isolated microalgae was cultured in modified bold

basal medium (MBBM) (Thompson *et al* 1988). Pure culture of isolated algae was done through repeated isolation and culture. Particle free medium (PFM), stock culture of *C. vulgaris*, design of experiment and mass culture were performed according to Miah (1993).

The samples of zooplankton were collected from the collected samples of BAU farm ponds with the help of plankton net of mesh size 55 μm . The dominant species of zooplankton as *Cyclops* sp. was isolated using pasteur soft micropipette under microscope. Pure culture of *Cyclops* sp. was performed through repeated isolation. It was fed with *C. vulgaris* cultured in the laboratory. Filtration and ingestion rates were calculated following the formula of Nimura (1980). In the evening before the day of experiment, zooplankton in stock culture was washed by sieving through a net of 50 μm mesh size by reverse filtration. This filtration was repeated 3 - 5 times for the experiment in higher food concentration while 4 - 8 times in lower food concentration. Washed zooplankton and algal suspension (both with aeration) were kept in the laboratory over night at $26 \pm 2^\circ\text{C}$ for acclimatization.

Just before the experiment, the algal suspension was diluted with PFM to have desired concentration. To the dilute algal suspension, the PFM was added in the ratio of 20:7 over control. The mixture was delivered to two control bottles, each containing 200 ml PFM. To the diluted algal suspension, the washed zooplankton were mixed in the ratio of 20:7. The mixture was delivered to four bottles each containing 200 ml water. Fifty individuals of zooplankton were released as initial density. All the bottles were kept in the laboratory at $26 \pm 2^\circ\text{C}$ under dim light condition. These bottles were slowly aerated to agitate the suspension according to Omori and Ikeda

*Author for correspondence; E.mail:ahsan@royalten.net.bd

(1984). Sub sampling was performed at every 90 min for five times in each series including the initial one. The initial sampling was done from each bottle using a plastic syringe after gently mixing the medium. During sampling, dissolved oxygen and pH were measured with oxygen (Yellow Springs Instrument, Model 58) and pH meters (Jenway, Model 3020), respectively. The sample was used to count algae and zooplankton. All the samples were fixed with 5% particle free formalin after sampling immediately. The microalgae and zooplankton were counted using haemocytometer and Sedgewick Rafter counting chamber, respectively. Statistical analysis was done using excel of microsoft win 6.0.

The quantitative study of phytoplankton and zooplankton of samples was done by Sedgewick Rafter (SR) counting cell, which was 50 mm long, 20 mm wide and one mm deep. The volume of the chamber was one ml (1000 mm^3 or 1 cc). The counting chamber is equally divided into 1000 fields. Each field had a capacity of one $1 \mu\text{m}$. From the concentrated volume of the plankton samples, one ml was taken by a dropper and then put on the SR cell. The counting chamber was covered with a cover slip so as to eliminate the air bubbles and then it was placed under microscope for further analysis. Phytoplankton was counted from 30 random fields (units) out of the fields of the SR counting cell with the help of binocular microscope. The cell/l of cultured samples was calculated according to Ward and Whipple (1959). Identification of phytoplankton was done following the keys of Whitford and Schumacher (1973) and Pontin (1978).

Results and Discussion

A series of feeding experiments of *Cyclops* sp. was carried out with *Chlorella vulgaris* at $26 \pm 2^\circ\text{C}$ for getting filtration and ingestion rates. The filtration rate in *Cyclops* sp. ranged from 3.00 to 6.43 $\mu\text{l}/\text{h}/\text{individual}$. Fig 1 shows the filtration rate in *Cyclops* sp. relating to food concentrations. It was attained a plateau. The range of food concentration was 515 to 2039 cells/ μl . The critical food concentration was 1435 cells/ μl . The filtering rate was obtained scattered widely in respect to various food concentrations. One of the problem in measuring actual rate was the failure in keeping the same initial food concentrations in the control and the experimental, because food organisms were usually attached with zooplankton in spite of several washings especially in the case of low food concentrations. The contribution due to growth of food organisms and grazing was assumed as identical on the specific growth rate. The number of grazers in respect to time was little bit changed. The data for analysis were chosen from those which showed less than 33% mortality. Hirayama and Ogawa (1972) reported the effect of hunger on the filtration

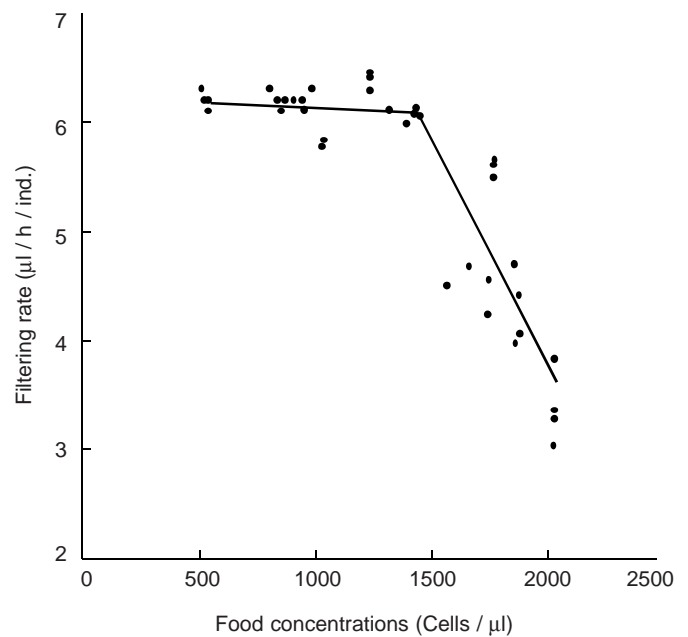


Fig 1. Filtering rate of *Cyclops* sp. in response to various food (*Chlorella* sp.) concentrations.

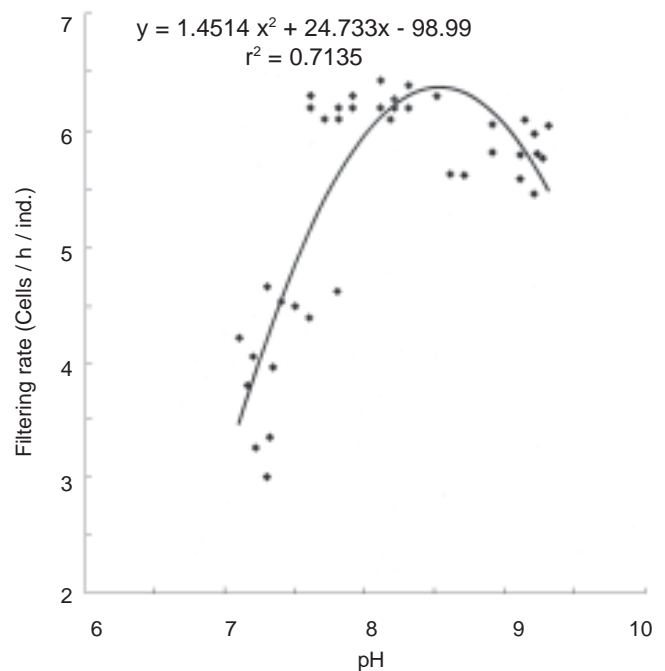


Fig 2. Filtering rate of *Cyclops* sp. in relation to pH.

rate in *Brachionus plicatilis*. A similar phenomenon was observed during the experiment. In low concentration, the initial data in filtering rate seemed to be larger than those obtained in later measurement. Hirayama and Ogawa (1972) obtained similar results during 60 min experiment with large food concentration. Rothhaupt (1990) considered more than 10 min for food to pass through the gut of *Brachionus*

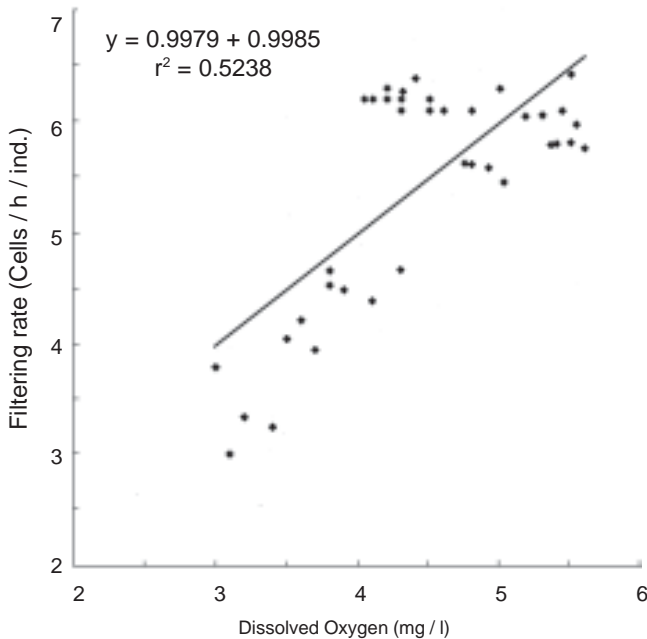


Fig 3. Filtering rate of *Cyclops* sp. in relation to dissolved oxygen.

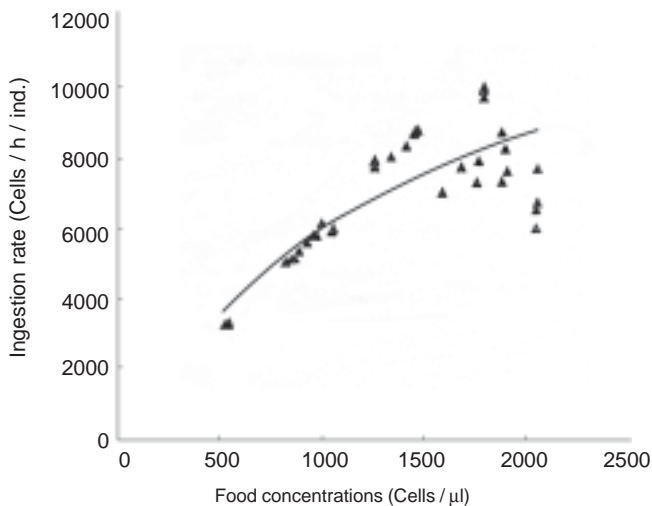


Fig 4. Ingestion rate of *Cyclops* sp. in response to various food (*Chlorella* sp.) concentrations.

calyciflorus. However, more or less similar results of present findings were reported by Hirayama and Ogawa (1972) and Chotiyaputta and Hirayama (1978).

The filtering rate of *Cyclops* sp. was positively related and highly significant ($p < 0.01$) with pH ($r^2 = 0.714$) (Fig 2). But it was observed from the graph that the filtering rate was decreased from pH 8.50. The filtering rate of *Cyclops* sp. was directly and significantly ($p < 0.01$, $df = 38$) related with dissolved oxygen ($r^2 = 0.524$) (Fig 3). There was no trend of decrease of filtering rate due to increase of dissolved oxy-

gen. At present there is no comparable data regarding this aspect. The estimated critical food concentration in *Cyclops* sp. was 1435 cells/ μ l. This was very high in concentration with that of Chotiyaputta and Hirayama (1978) and Schlosser and Anger (1982) except in some cases of rotifers reported by Rothhaupt (1990). However, the slope obtained was usually larger than 1.0 which meant that the ingestion or food uptake rate increased above the critical food concentration. It was very difficult to know the reason for the difference in the critical concentration from others at this moment. No comparable data on critical food concentration in *Cyclops* sp. was available.

Figure 4 shows that the food uptake rate (ingestion rate) of *Cyclops* sp. is related to the food concentration. The ingestion rate attained a maximum of 9971 cells/h/individual at the concentration of 771 cells/ μ l. The ingestion rate increased with food concentration tested throughout. The minimal rate obtained was 3245 cells/h/individual at a concentration of 515 cells/ μ l. The maximal ingestion rate in *Brachionus plicatilis* reported by Doohan (1973) and Schlosser and Anger (1982) ranged from 0.04 to 0.29 μ l/h/rotifer in the temperature ranged from 20 to 25°C. Rothhaupt (1990) reported that ingestion rate in *B. calyciflorus* and *B. rubens* ranged from 0.019 to 0.198 μ l/h/rotifer at 20°C. The corresponding data of the experiment was 3245 - 9971 cells/h/*Cyclops* at $26 \pm 2^\circ\text{C}$ which was larger than the above reports. It was not formed a plateau. This may be due to the size and condition of food organisms.

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