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Study the Production of Rotifer Culture in Different Concentration of Microalgae with Combination of Manure

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ABSTRACT

The estuarine water containing groundnut oilcake, urea, superphosphate, different combination of manures rice bran (CM1), yeast (CM2), cow dung (CM3) and poultry manure (CM4) with different concentration of chlorella sp., Nannochlopsis sp. and Cyclotella sp. 1, 2, 3, 4, 5×10^5 cells/ml were added as supplementary food along with the culture of Brachionus rotundiformis and Brachionus plicatilis. The aim of the presence study was to optimize the growth of B.rotundiformis and Brachionus plicatilis on different feeds of ricebran, yeast, cowdung and poultry manure and observed their production.

Key words: *B.rotundiformis, B. plicatilis, microalgae, algal cell, culture medium*

INTRODUCTION

Rotifers have been widely used as essential food source in raising freshwater and marine fish larvae due to its unique characteristics[3]. The commonly cultured zooplankton in fish culture is Rotifer. Rotifer is the most dominant zooplankton in all the freshwater aquatic ecosystems and is considered an ideal food for fish larvae[1]. The success in the hatchery production of fish fingerlings for stocking in the grow-out production system is largely dependent on the availability of suitable live food organisms eg rotifer for feeding fish larvae, fry and fingerlings[7].

Brachionus rotundiformis have been used as live food organisms in marine fish-farming centers[6]. The rotifer *Brachionus plicatilis* has become a valuable and, in many cases indispensable, food organism for first feeding of a large variety of cultured marine finfish and crustacean larvae[8].

MATERIALS AND METHODS

Collection of sample

The zooplankton samples were collected from the early morning of Manakudy estuary and are brought to the laboratory for identification. Isolated the rotifers *B.rotundiformis* and *B.plicatilis* were done with the help of light microscope (Fig 1).

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Isolation of algal cells

Microalgae like *Chlorella* sp., *Nanochloropsis* sp., *Cyclotella* sp present in a sample were isolated.

Growth of rotifer in microalgae and combination of manure at different concentration

Two species of *B.rotundiformis* and *B.plicatilis* were collected with 135 µm mesh size plankton net into a plastic bucket containing the estuarine water and brought to the laboratory. The separation of two species and kept in polythene jars of 2 litre capacity were selected for rearing rotifers. Triplicates of experiments were conducted. The room temperature (30±2°C) was maintained during the time of experiment and the concentration of algae was noticed.

In this experiment, estuarine water containing different combination of manure (CM) groundnut oilcake, urea, superphosphate, rice bran (CM1), yeast (CM2), cow dung (CM3) and poultry manure (CM4) were added. Then the different concentration of *Chlorella* sp. 1, 2, 3, 4, 5×10^5 cells/ml, *Nannochloropsis* sp. 1, 2, 3, 4, 5×10^5 cells/ml and *Cyclotella* sp. 1, 2, 3, 4, 5×10^5 cells/ml were added as supplementary food along with the culture of *Brachionus rotundiformis* and *Brachionus plicatilis.* In the first concentration (AC 1×10^5/ml). In the second concentration (AC 2×10^5/ml), third concentration (AC 3×10^5/ml), fourth concentration (AC 4×10^5/ml) and in the fifth concentration (AC 5×10^5/ml). The manure and algae were added at three day intervals. In the same experimental first step contain *Chlorella* sp. (1, 2, 3, 4, 5 × 10^5cells/ml) and the second step *Nannochloropsis* sp. (1, 2, 3, 4, 5× 10^5cells/ml) and third step contain *Cyclotella* sp. (1, 2, 3, 4, 5× 10^5cells/ml). Transparent polythene jars were selected for rearing the species. During the experiment, all the polythene jars were aerated with 2 litre of aerated filtered estuarine water was added along with the culture media containing the various ingredients (Fig 2).

The culture media were inoculated with 100 individuals. In all the culture media the production of rotifers were counted. The final numerical estimates were expressed as no/ml

RESULTS

The observation of the number of organism recorded in various culture media are shown in Tables 1.1 to 1.6. The experiment conducted in algal concentration (1×10^5 cells/ml) exhibited low production of rotifers. It was observed that all the organisms survived in all the media and showed variable production rates.

Growth of rotifer in different concentration of microalgal cell and combination of manure

Rice bran (CM1), Yeast (CM2), Cow dung (CM3) and Poultry manure (CM4) were used to prepare culture media with different concentration of *Chlorella* sp. (1, 2, 3, 4, 5×10^5 cells/ml), *Nannochlopsis* sp. (1, 2, 3, 4, 5×10^5 cells/ml) and *Cyclotella* sp. (1, 2, 3, 4, 5×10^5 cells/ml) as supplementary food. In the present study, 5×10^5/ml of algal cells mixed with all the three media was found as efficient diet in the production of *Brachionus plicatilis* and *B.roundiformis.* The medium with *Nannochloropsis* sp. showed better result than the *Cyclotella* sp. and *Chlorella* sp. in the culture of *Brachionus roundiformis* (880.63 no/ml) (Table 1.2).

When *B.roundiformis* was added into the medium containing *Chlorella* sp., it showed a variable production rates. The production was high (618.75 no/ml) in AC 4 $\times10^5$ /ml and low (227.88 no/ml) in AC 4 $\times10^5$ /ml of cowdung manure, while the next maximum (608.25 no/ml) was obtained in AC 4 $\times10^5$ /ml and low (276.75 no/ml) in AC 1 $\times10^5$ /ml of ricebran. Yeast resulted in high number of rotifer (601.13 no/ml) in AC 4 $\times10^5$/ml and low 213.88 no/ml in AC 1×10^5/ml. The production rate of *Brachionus roundiformis* in poultry manure showed high production of animal (613.38 no/ml) in AC 5×10^5/ml and low (226.25 no/ml) in AC 1×10^5/ml (Table 1.1).

High production of *Brachionus roundiformis* was recorded in all the media with *Nannochloropsis.* Peak production in the *Brachionus roundiformis* was noticed in the cowdung CM3. In this medium, higher production (880.63 no/ml) was noted in AC 5×10^5/ml. Rice bran contain next higher population (798.88 no/ml) in AC 5×10^5/ml. Yeast medium contain next higher population (788.13 no/ml) in AC 5×10^5/ml and the next highest population of *Brachionus roundiformis* in poultry manure (672.13 no/ml) in AC 5×10^5/ml . The low number of production (363.50, 361.88, 375.25 and 369.38 no/ml respectively) was obtained in AC 1×10^5/ml of rice bran, Yeast, Cow dung and Poultry manure (Table 1.2).

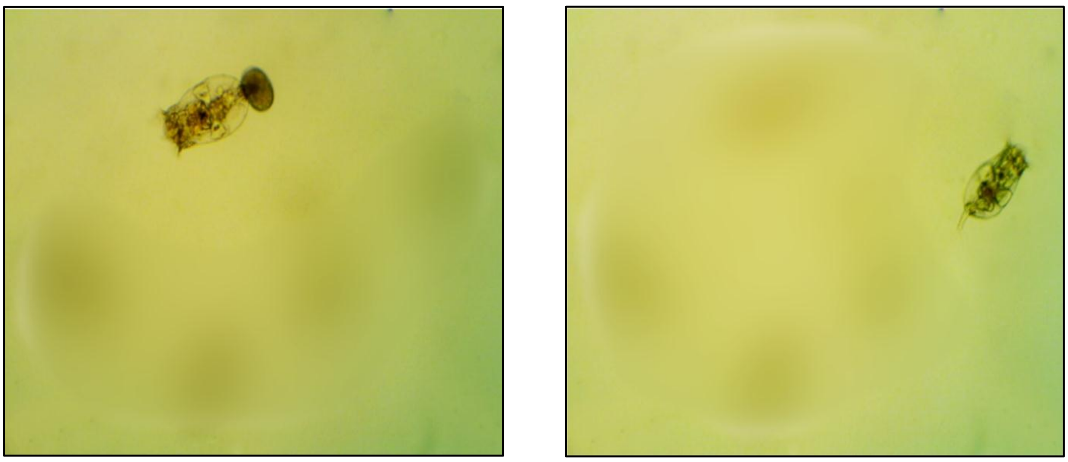
Similarly the density of *Brachionus roundiformis*, showed a maximum population density when *Cyclotella* sp was added to the medium. The high production rate (667.00 no/ml) was noticed in AC 5×10^5/ml in cowdung manure and minimum (344.88no/ml) in AC 1×10^5/ml. It is followed by (630.50 no/ml) with the AC 4×10^5/ml in yeast medium and then low value (357.38no/ml) in AC 1×10^5/ml. In poultry manure, a population density of 604.50 no/ml was obtained in AC 5×10^5/ml and low (331.63 no/10ml) in AC 1×10^5/ml. The rice bran showed the high production (677 no/ml) in AC 5×10^5/ml and low (337.38 no/10ml) in AC 1×10^5/ml (Table 1.3).

When *B. plicatilis* was added to the medium containing *Chlorella* sp, the high number (528.63 no/ml) was attained in AC 4×10^5/ml and low (147.25 no/ml) in AC 1×10^5/ml of ricebran. The peak value (570.63 no/ml) in AC 4×10^5/ml and AC 5×10^5/ml and low (117.50) of yeast manure. In cowdung, the production of *B.plicatilis* was maximum (503.13no/ml) in AC 5×10^5/ml and minimum 151.75/no/10ml of AC 1×10^5/ml. The production was more (528.38 no/ml) in AC 4×10^5/ml and less 197.25/no/ml in AC 1×10^5/ml of poultry manure (Table 1.4).

Similarly, the density of *Brachionus plicatilis* showed a population change when *Nannochloropsis* sp was added to the medium. The production was high (565.63 no/ml) in AC 5×10^5/ml of the yeast medium. A high population of 504.88, 471.38 and 462.38no/ml in AC 5×10^5/ml of ricebran, cowdung and poultry manure respectively. A low production of 134.75, 132.38, 138.13 and 177.75 no/ml respecively in AC 1×10^5/ml of rice bran, Yeast, Cow dung and Poultry manure (Table 1.5).

High production of *Brachionus plicatilis* was recorded in all the media with *Cyclotella* sp. The peak production (568.75 no/ml) was noticed in AC 5×10^5/ml of the yeast manure. Second maximum population (502.50 no/ml) was recorded in AC 5×10^5/ml of ricebran. The poultry manure and the cowdung manure showed the high population of 513.00 and 538.25 no/ml respectively in AC 5×10^5/ml. A low production of 172.13, 136.75, 136.63 and 115.63 no/ml respectively were found in poultry manure, cowdung, yeast and ricebran in AC 1 $\times10^5$/ml (Table 1.6).

Fig. 1: Isolated rotifer *Brachionus rotundiformis and Brachionus plicatilis*



Brachionus rotundiformis *Brachionus plicatilis*

Fig. 2: Experimental setup showing the culture of rotifer in different concentration with culture medium

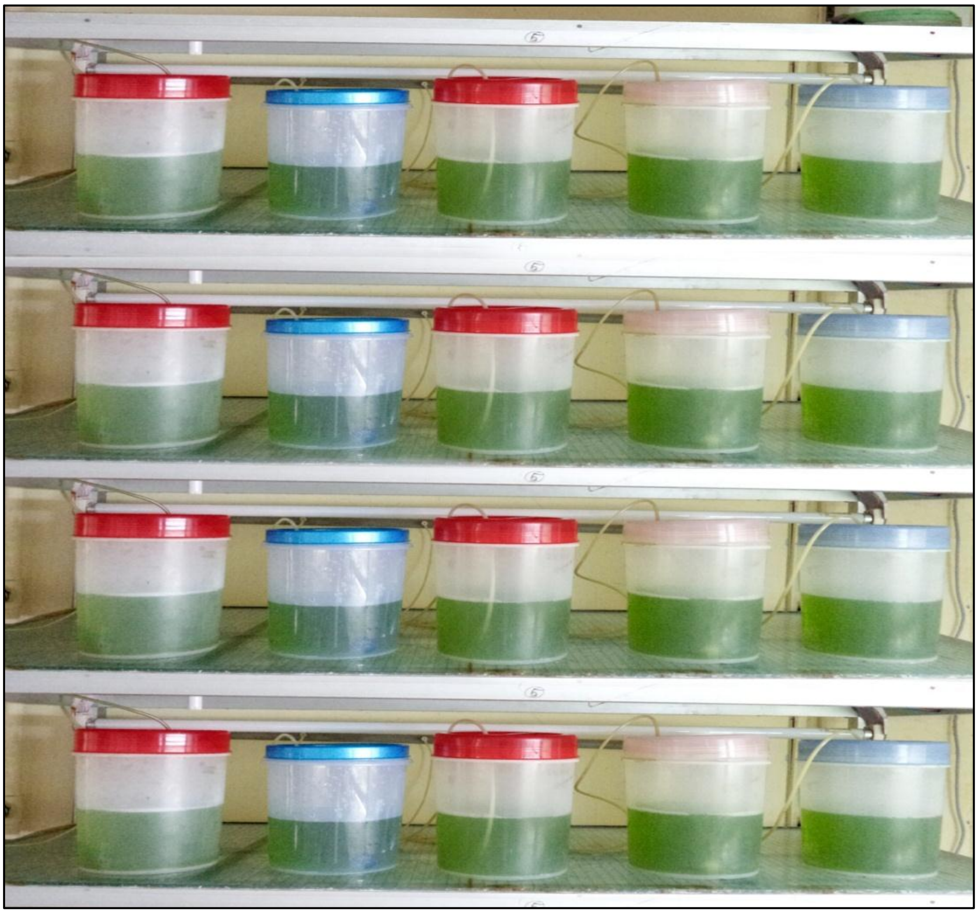


Table 1.1. Production of *Brachionus rotundiformis* in four media with C*hlorella* sp.

Medium	AC 1×10^5/ml	AC 2×10^5/ml	AC 3×10^5/ml	AC 4×10^5/ml	AC 5×10^5/ml
CM1	276.75±22.89	338.25±44.50	467.00±15.84	608.25±25.28	580.50±20.45
CM2	213.88±9.52	342.50±59.40	466.50±24.42	601.13±59.77	571.63±25.31
CM3	227.88±9.20	347.75±23.16	499.75±53.83	618.75±18.42	591.25±59.13
CM4	226.25±8.44	395.63±54.11	450.75±56.83	552.63±24.18	613.38±21.22

Table 1.2. Production of *Brachionus rotundiformis* in four media with *Nannochloropsis* sp.

Medium	AC 1×10^5/ml	AC 2×10^5/ml	AC 3×10^5/ml	AC 4×10^5/ml	AC 5×10^5/ml
CM1	363.50±15.19	484.75±14.75	583.00±18.01	620.88±57.83	798.88±34.86
CM2	361.88±15.09	553.75±50.69	670.38±46.03	713.63±2.97	788.13±9.46
CM3	375.25±19.05	557.25±31.44	613.38±28.70	865.75±16.01	880.63±17.75
CM4	369.38±22.86	554.13±56.50	562.38±32.34	596.63±2.86	672.13±61.78

Table 1.3. Production of *Brachionus rotundiformis* in four media with *Cyclotella* sp.

Medium	AC 1×10^5/ml	AC 2×10^5/ml	AC 3×10^5/ml	AC 4×10^5/ml	AC 5×10^5/ml
CM1	337.38±26.01	383.13±33.43	571.75±38.65	570.50±26.68	677.00±30.50
CM2	357.38±26.01	483.13±33.43	571.75±38.65	630.50±26.68	577.00±30.50
CM3	344.88±30.23	463.63±36.57	547.63±34.44	592.38±55.04	667.00±115.23
CM4	331.63±26.03	484.75±17.29	489.75±33.34	600.63±40.57	604.50±18.50

CM1 - Ricebran CM2 - Yeast
CM3 - Cowdung CM4 - Poultry manure
AC - Algal cell

Table 1.4. Production of *Brachionus plicatilis* in four media with *Chlorella* sp.

Medium	AC 1×10^5/ml	AC 2×10^5/ml	AC 3×10^5/ml	AC 4×10^5/ml	AC 5×10^5/ml
CM1	147.25±24.36	230.38±47.28	385.38±48.10	528.63±48.69	523.88±63.51
CM2	117.50±19.23	215.38±32.28	325.13±36.83	570.63±66.46	570.63±14.77
CM3	151.75±7.46	335.25±17.77	328.75±30.10	408.88±7.86	503.13±18.88
CM4	197.25±10.33	272.88±7.59	338.88±27.60	528.38±26.58	503.88±6.47

Table 1.5. Production of *Brachionus plicatilis* in the four media with *Nannochloropsis* sp.

Medium	AC 1×10^5/ml	AC 2×10^5/ml	AC 3×10^5/ml	AC 4×10^5/ml	AC 5×10^5/ml
CM1	134.75±8.97	306.25±13.55	378.75±25.56	443.75±30.79	504.88±19.19
CM2	132.38±6.14	262.50±15.87	411.75±28.14	521.38±10.31	565.63±52.89
CM3	138.13±4.88	244.75±30.86	342.50±20.54	402.63±12.23	471.38±22.23
CM4	177.75±14.83	334.75±35.86	361.25±33.67	421.63±21.99	462.38±21.99

Table 1.6. Production of *Brachionus plicatilis* in four media with *Cyclotella* sp.

Medium	AC 1×10^5/ml	AC 2×10^5/ml	AC 3×10^5/ml	AC 4×10^5/ml	AC 5×10^5/ml
CM1	115.63±7.25	278.75±25.45	411.25±12.96	444.38±26.54	502.50±7.15
CM2	136.63±15.17	371.25±8.14	488.13±19.16	419.13±53.76	568.75±59.82
CM3	136.75±4.74	219.13±23.58	446.13±14.61	485.00±16.84	538.25±10.63
CM4	172.13±16.64	342.25±23.44	369.13±11.91	483.38±18.44	513.00±24.19

CM1 - Ricebran CM2 - Yeast
CM3 - Cowdung CM4 - Poultry manure
AC - Algal cell

DISCUSSION

The aim of the presence study was to optimize the growth and production of *B.rotundiformis* and *Brachionus plicatilis* on different feeds of ricebran, yeast, cowdung and poultry manure was observed. It is noted that the *Brachionus rotundiformis* thrive well in cowdung manure and their biomass was also high. The greatest potential for rotifer culture resides in the possibility of rearing these animals at very high densities. Densities of 2,000 individuals m^{-1} have been reported by Harita[4]. Even at high densities, the animals reproduce rapidly and can thus contribute to the buildup of large quantities of live food in a very short time. *B.rotundiformis* was maximum in yeast medium and their production rate is low by comparing *Brachionus plicatilis.* Hirata[5] has reported 16789 *Nannochloropsis* cells per marine rotifer, *B. plicatilis*.

Ovie and Ovie[10] on the culture of zooplankton, using organic manure. The result of the study also showed that pig dung, poultry droppings and cow dung can be successfully used to mass produce *Brachionus plicatilis and Brachionus rotundiformis.* Bhat and Moody[2] (1995) have successfully grown rotifers in cowdung manure.

In the presence study, the *B.rotundiformis* size was larger than *Brachionus plicatilis*. So the *B. plicatilis* was more advisiable for the larviculture because of small size. The result indicates the strain variation was one of the major parameters which require detailed investigations. The above said result has also confirmed by the study conducted by Menon and Pillai[9].

CONCLUSION

The local strains of B. *rotundiformis* and *Brachionus plicatilis* are recognized as indispensable live feeds for marine larviculture and grown well in the entire four medium. Therefore these entire medium were used for the culture. Feeding of early larval stages is the major bottleneck for the industrial upscaling of aquaculture of fin and shellfish.

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