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Optimum Fertilizer Media and Plant Substrates: (*Musa paradisiaca* L. Peels, *Carica papaya* L. Peels and *Brassica oleracea var. capitata* L. Leaves) for the Growth of Freshwater Rotifer (*Brachionus calyciflorus* Pallas, 1766)

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Authors' contributions

This work was carried out in collaboration among all authors. Author OFO designed the study, wrote the protocol and wrote the first draft of the manuscript. Author LEI performed the statistical analysis and managed the analyses of the study. Author PCO managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

The aim of this study was to search for a cheap source for the mass production of *Brachionus calyciflorus* Pallas, 1766 which is a good substitute for artemia in the culture of *C. gariepinus hatchlings.* Poultry manure (1.25 g/L), soya bean (1.25 g/L), groundnut cake (1.25 g/L) and single super phosphate (0.04 g/l) were used exclusively and in combinations as fertilizer media in the culturing of *Brachionus calyciflorus* Pallas, 1766 used in feeding *C. gariepinus* hatchlings. The four fertilizer media were combined to give four treatments with two replicates. Three fresh plant substrate (*Carica papaya L.* Peels, *Musa paradisiaca* L. peels and *Brassica oleracea* var. capitata

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L. leaves) were introduced into treatments labeled A (Poultry manure, soya bean, groundnut cake, single super phosphate and banana pills), B (Poultry manure, soya bean, groundnut cake, single super phosphate and *Carica papaya L.* Peels), C (Poultry manure, soya bean, groundnut cake, single super phosphate and cabbage slice) with D(Poultry manure, soya bean, groundnut cake, single super phosphate) as the control. A total of 20 Individuals/mL of *Brachionus calyciflorus Pallas,* 1766 were inoculated into each treatment and the culturing lasted for 12 days. At the end of the culturing period, the best doubling time (t_D) were observed in treatment A and B with a doubling time of 2.89 on the 10^{th} day.
Result of the study showed medium B and C to be sig. different ($P< 0.05$) and performing better than A and D in the early stages of the study. There was an initial lull in production using the C medium, as it had the least performance. By day eight, it had picked up and there was no sig. difference ($P>0.05$) in production of *B. calyciflorus* Pallas, 1776 using the four culture media. Thus any of the media can be used for mass production of *B. calyciflorus*. Use of media B and C is however, recommended because they had a higher carrying capacity and can sustain production over a longer period using a giving quantity compared to the other two media.

Keywords: Brachionus calyciflorus Pallas 1766; Musa paradisiaca L. peels; Carica papaya L. peels and Brassica oleracea var. capitata L. leaves.

1. INTRODUCTION

Zooplankton is a collective term used for a variety of freshwater and marine animals. The word zooplankton is derived from two Greek words “zoon” meaning animals and “plankton” which means drifters or wanderers. They are microscopic with low morbidity and also have high amino acid content [1]. Some are large and visible to the eye e.g. jelly fish [2]. Although they are primarily transported by ambient water current, many have locomotion organs with which they use to avoid predators [3]. They feed on variety of food which includes bacterioplankon [3].

Zooplankton plays a very important role in the aquatic food web, as they serve as an ideal starter food sources for consumers on the higher tropic levels. Rotifers improved the growth and survival of gourami larvae [4]. A single rotifer can become thousands, in a few days when the environmental conditions are favorable [1]. With this, a female *B. calyciflorus* can produce up to 7 batches of eggs simultaneously without any genetic input from the male and these eggs hatch in 12 hours and by 18 hours from the hatching time, the new *B. calyciflorus* is ready to start reproducing [1]. Although, a female *B. calyciflorus* can produce ten generations of offspring before they eventually die [5], their availability in good quality and quantity serves as a good factor for the successful production of *Clarias gariepinus* fry [6,1]. This study was aimed at finding the optimum fertilizer media and plant substrates, which will be the best in the growth and survival of fresh water rotifer (*Brachionus calyciflorus Pallas,* 1766). The fresh water rotifer *B. calyciflorus Pallas, 1766* has been used successfully for the culture of the African Catfish *Clarias gariepinus* [7].

2. MATERIALS AND METHODS

2.1 Preparation of Fertilizer Media and Plants Substrates

Poultry manure (1.25 g/l), soya bean (1.25 g/l), groundnut cake (1.25 g/l) and single super phosphate (0.04g/l) were obtained locally from the market and afterwards sun dried for 3 days. All ingredients were ground into powdery form using a corn mill. The milled ingredients were then sieved with a mosquito net to separate the fine powder from the remaining fiber. They were then soaked in water inside a 2 Liter plastic container covered with a mosquito net and allowed to stand for 2 days this was done as per the method described by [8]. The *Musa paradisiaca L.* peels, *Carica papaya L. Peels and Brassica oleracea var. capitata L. leaves* were freshly plucked from their branches on the ground of Collage of Forestry, Jos, Plateau Sate, Nigeria. The leaf stalks were removed by hand. The leaves were soaked in water for 3 days and afterwards sundried for another 3days.

2.2 Screening and Sampling of *Brachionus calyciflorus Pallas,* 1766 Feeding

Pure culture of *Brachionus calyciflorus* were used and obtained from the fisheries unity, Faculty of Agriculture, Nasarawa State University, Keffi. Nigeria. The culture was

collected using a modified standard of Clarke-Bumpus zooplankton sample fixed with a straining net and collection bottle according to [2,9]. They were transferred to the site of the experiment in the in the automated thermo-controlled fish hatchery unit of global Aquaculture and Allied Ventures (GAAV) in Jos-south, Plateau State, Nigeria.

2.3 Experimental Set Up

The experiments were carried out in four 10 Liter plastic containers with water volume maintained at 8 liters. The banana pills, pawpaw pills and cabbage leaves were allotted at the rate of 20g per pills and leaves into each of the experimental plastic containers labeled A (*Musa paradisiaca L. Peels*), [B] (*Carica papaya L.* Peels*).* [C] (*Brassica oleracea var. capitata L.* leaves) with D serving as the control. Each experimental plastic container has 2 other replicates. A total of 20 individuals/ml *B. calyciflorus Pallas, 1766* were inoculated into each treatment. The *B. calyciflorus Pallas, 1766* were fed twice daily, between 9.00-10.00am and 15.00-16.00pm at 2% body weight throughout the experiment using the fertilizer media at room temperature. Yield evaluation started two days after inoculation by collecting water from the plastic containers using an automated pipette and centrifuging for 2mins. The automated pipette was also used to place 2 drops of the water sample on a glass slide and was mounted on a microscope. The sample was observed in x10 objective of the wild binocular BH2 Vanox 7 microscope as reported by [10]. The experiment lasted for 12 days.

2.4 Data Collection and Analysis

Data on *B. calyciflorus Pallas, 1766* instantaneous growth rate and doubling time were recorded and calculated at every 2 days as follows:

Instantaneous Growth Rate (K) = $\frac{[\log N_t - \log N_0]}{T}$

where

N_0 = Number of rotifer in the inocula (10)
Nt = Final number of rotifer after time (t) in days

Doubling Time t = $\frac{\log_2}{K}$

Where K is Instantaneous Growth Rate (10)

2.5 Water Quality

Water quality parameters were measured during each sampling. Temperature and dissolved oxygen were measured in *situ*, with a WTM, OxiCal-SL portable electronic probe. The water pH was also measured with Suntex model SP-701 pH meter.

2.6 Statistical Analysis

All data obtained were analyzed using Statistical Package for Social Scientists (SPSS 15.0 2006) to determine their significant difference. The differences were checked at p = 0.05 level of significance. A 2-way analysis of variance (ANOVA) and Duncan's multiple range tests were used to determine significant differences in yields of rotifer from the different fertilizer media.

3. RESULTS

The results of the mean values of *B. calyciflorus Pallas, 1766* yield culture media over 12 days is presented in Table 1 and Fig. 1 respectively. The result of this study showed that media B and C were significantly different (P< 0.05) and performing better than A and D in the early stages of this study. There was an initial lull in production using the C medium, as it had the least performance. By the sixth day, it had picked up and there was no sig. difference (P>0.05) in production of *B. calyciflorus* Pallas, 1776 using the four culture media.

There was a significant difference (P< 0.05) amongst treatments although, all the fertilizer media and their substrate supported increase of *B. calyciflorus Pallas, 1766* yield for the period of 12 days. The result showed that the highest yield (268 Individuals/mL) was obtained in fertilizer media A [Poultry manure + Groundnut cake + Soya bean cake + SSP + *Musa paradisiaca L.* Peels] on the 10^{th} day of culturing, followed by B [Poultry manure + Groundnut cake + Soya bean cake + SSP] with 232 individuals/ml then, fertilizer media C [Poultry manure + Groundnut cake + Soya bean cake + SSP + *Brassica oleracea var. capitata L.leaves*] with 224 individuals/ml and the least was observed in fertilizer media D [Poultry manure + Groundnut cake + Soya bean cake + SSP + *Carica papaya L. Peels*] with 211 individuals/ml. The growth and doubling time performance of *B. calyciflorus* pallas, 1766 cultured in fertilizer media is presented in (Table 2). The doubling time for treatment B and C was 2.89 while, that of treatment A and D were 2.67 and 2.77 respectively. Anova results indicated a significant difference amongst treatment but there was no significant difference between treatment B and C.

Table 1. Mean values of *B. calyciflorus Pallas,* 1766 yield culture media over 12 days

Fertilizer media	Rotifer mean yield period of culture (days)						
	0	**2**	**4**	**6**	**8**	**10**	**12**
A	20	29	52	110	222	268	10
B	20	26	44	102	150	211	54
C	20	24	32	90	128	224	20
D	20	22	34	96	132	232	0

A [Musa paradisiaca L. peels], B [Carica papaya L. Peels], C [Brassica oleracea var. capitata L. leaves.] with D serving as the control

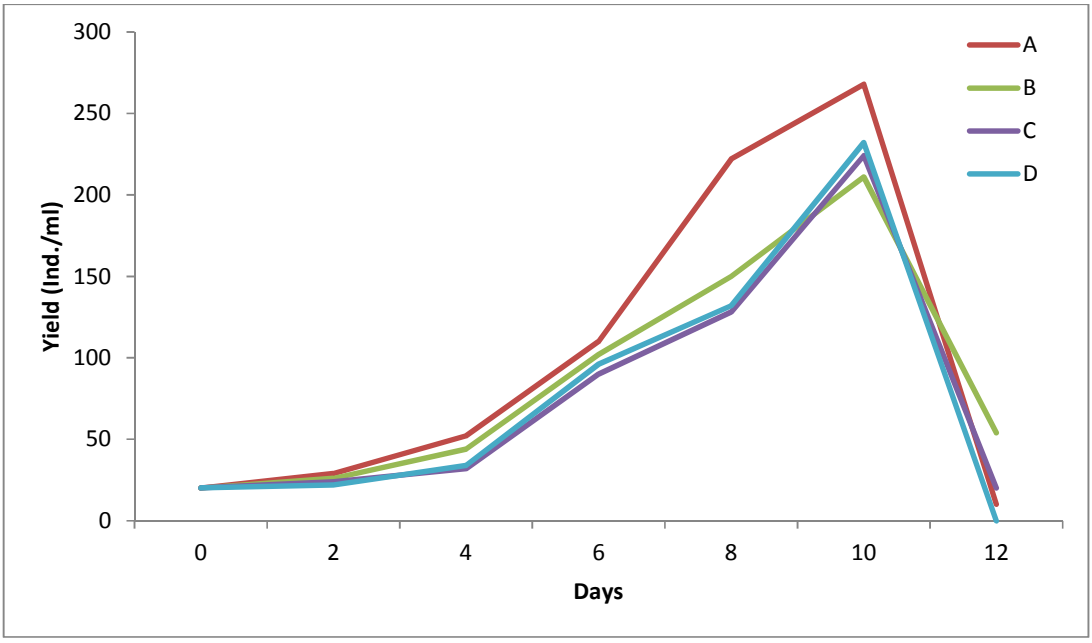

Fig. 1. Mean daily yield of *B. calyciflorus Pallas*, 1766 in different culture media

Table 2. Instantaneous growth rate and best doubling time of *B. calyciflorus Pallas*, 1766

Fertilizer media	Initial no. of *B. calyciflorus*	Finial no. of *B. calyciflorus*	Time (days)	Instantaneous growth rate	Doubling time
A	20	268	10	0..26	2.67
B	20	211	10	0.24	2.89
C	20	224	10	0.24	2.89
D	20	232	10	0.25	2.77

4. DISCUSSION

This study revealed that the highest rotifer yield (268 individuals/mL) was attained on the 10th day of culturing using poultry manure, soya bean cake, groundnut cake, single super phosphate and *Musa paradisiaca L.* leaves. This is in agreement with [11] who obtained a peak of 230 individuals/mL on the 9th day after inoculating 10 ind/mL of *Dunaliella* species fed with *Brachionus Plicatilis*. Though the yield in this study differs slightly from theirs, which could be as a result of the type of species used and also the number inoculated.

This study indicates that there was an initial lull in production using the C medium, as it had the least performance. By the sixth day, it had picked up and there was no sig. difference (P>0.05) in production of *B. calyciflorus* Pallas, 1776 using the four culture media. Thus any of the media

can be used for mass production of *B. calyciflorus* Pallas, 1776.

This difference could be explained by the nature of the various fertilizers utilized showing that media B and C might be rich in nutrient for the production of phytoplankton which are foods for rotifers than those of others. This is in agreement with [12] who opined that poultry manure is superior to cattle dung and press mud, considering the biomass and plankton biomass. Also [13] reported that the fertilized media offered the best phytoplankton biomass and the best zooplankton maximum density. These results are comparable to those obtained by [14] with pig dung and with those obtained by [15] with poultry dropping. [16] also reported a very positive result with poultry dropping in the mass culture of *Ceriodaphnia reticulata* (Jurine) as a live fish feed.

5. CONCLUSION

This study suggests that any of the media can be used for mass production of *B. calyciflorus*. However, the use of media B and C are recommended because they had a higher carrying capacity and can sustain production over a longer period using a giving quantity compared to the other two media (A and D).

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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