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Aditi Rajan Patkar
Department of Aquaculture, College of Fisheries, Mangaluru, Karnataka Veterinary Animal and Fisheries Sciences University, Bidar, Karnataka, India

Manjappa N
Fisheries Research & Information Centre (Inland), Hesaraghatta, Bengaluru, India, Karnataka Veterinary Animal and Fisheries Sciences University, Bidar, Karnataka, India

Harsha Nayak
Fisheries Research & Information Centre (Inland), Hesaraghatta, Bengaluru, India, Karnataka Veterinary Animal and Fisheries Sciences University, Bidar, Karnataka, India

Corresponding Author:
Aditi Rajan Patkar
Department of Aquaculture, College of Fisheries, Mangaluru, Karnataka Veterinary Animal and Fisheries Sciences University, Bidar, Karnataka, India

Dynamics of plankton in substrate based fish & prawn culture

Aditi Rajan Patkar, Manjappa N and Harsha Nayak

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Abstract
A study was conducted to evaluate the effect of periphyton substrate on plankton dynamics. Sugarcane bagasse is used as a substrate for periphyton growth. Three experimental treatments were included; T_0 (Control), T_1 (Substrate + feed) and T_2 (substrate only). Indigenous fishes like *Labeo fimbriatus* (Bloch,1795), *Barbodes carnaticus* (Jerdon,1849) and freshwater prawn, *Macrobrachium rosenbergii* were stocked in tanks in 3:3:4 ratio at 10000/ha, for a period of 180 days. Bagasse substrate influenced the water quality parameters *viz*., pH (6.73 – 8.17), dissolved oxygen (6.70 – 8.50). Low concentration of Ammonia (0.068 – 0.140 mg/l) and Nitrite (0.046 – 0.082) was observed in substrate based ponds. A decreasing trend was observed in periphyton DM, AFDM and ash content value in both the treatment near the termination of experiment, which indicates effective grazing on periphyton by cultured fish species. Also, pigment concentration was also found high in substrate ponds. The result demonstrate that sugarcane bagasse can effectively use as a substrate for enhancing the population of plankton.

Keywords: sugarcane bagasse, water quality, plankton density, periphyton

1. Introduction
There is strong relationship between plankton abundance and fish production, as planktons forms the most abundant food base where fishes are not provided with supplemental feed. Phytoplanktons contain light-sensitive pigments that allow them to convert carbon dioxide and water to simple sugar with the capture of solar energy in the sugar and release of molecular oxygen into the water. Phytoplankton cells use sugar in photosynthesis as a source of energy for respiration. However, these sugar is not used in respiration but biochemically converted to organic compounds such as starch, cellulose, amino acids, proteins, and fats that stored in phytoplankton cells, which are further needed by heterotrophic bacteria and animals that feed on phytoplankton cells or their remains. Diversity, distribution, abundance and variation in the biotic factors provide information of energy turnover in the aquatic systems (Gaikwad *et al.*, 2004) [1]. In these systems, plankton is very important. As we know, periphyton has become a universally accepted term for all organisms attached to a submerged substrate. However, the periphyton quantity and quality depends on abiotic and biotic factors (nutrients, light intensity and quality, temperature, water level, as well as the substrate type and the grazing activity of the fish and invertebrates). The autotrophic organisms of periphyton produce organic material and oxygen by using light energy and absorbing nutrients. The organic material produced in this way can provide valuable nutrition for the zooplankton and other heterotrophic communities in the water. The heterotrophic organisms also use drifting, settling or settled organic material in their metabolic processes (Lakatos, 1997) [2]. Several authors carried out experiments by using different periphyton substrates from various aspects and provided different explanation including improvement in the water quality, addition of the natural food supplement, limitation of disease producing bacteria, provide refuge for prawns to escape any undesirable behavioral interactions and increase living space (Burford *et al.*, 2004; Ballester *et al.*, 2007) [3, 4]. The variability of phytoplankton with the seasonal changes in aquatic environment is strongly needed for the maintenance of water quality and sustainable aquaculture. The qualitative and quantitative abundance of plankton and its relationship with environmental conditions have become a prerequisite for fish production. One of the most important aspects of polyculture is to increase the productivity of water bodies through the effective use of naturally occurring food organisms. Several studies on plankton diversity

made in India and abroad on the ponds, lakes and reservoirs but the data dealing with the effect of substrate on plankton population are scanty. Therefore, the aim of the present work was to i) Study the qualitative and quantitative estimation of planktons as well as periphyton and ii) determine the effects of substrates on diversity of planktons

2. Materials and Methods

2.1 Experimental Design & Site

The experiment was carried out for a period of 180 days at the Fisheries Research Information Centre (Inland), Hesaraghatta (13º8'18.8088"N and 77º28'40.4040" E), Bengaluru, Karnataka, India from Nov-2019 to Apr-2020. The experiment was conducted in cement cisterns, each with size of 20m^2. One control and two treatments of sugarcane bagasse substrates in triplicates were tried: No substrates, substrates and Substrate + feed. Here in called treatments T_0, T_1 and T_2 respectively.

Locally collected Sugarcane bagasse was washed initially and sun dried for a week so as to remove excess molasses from it. Then, bundles of approximately 7.5cm diameter and 1m length were made using nylon rope (Keshavanath, 2001) [5] kept ready aside for installation in T_1 and T_2 treatment ponds.

2.2 Pond Preparation, Stocking & Feeding

Before start of the experiment, all cisterns were dried initially for a week, all aquatic vegetation were removed. Soil base of about 15cm was added & agriculture lime ($CaCO_3$) was applied to pond bottom at a rate of 250 kg/ha. Next day of liming, water was filled in all ponds. Prepared bagasse bundles was hung vertically at 5000kg/ha in ponds of T_1 and T_2 with the regular intervals from thick nylon ropes tied across the pond by maintaining uniform distance of 20 cm between bagasse and tank inner surface as a free border zone. After a week of liming, all ponds were fertilized with semi-decomposed cow dung, urea and triple super phosphate (TSP) at the rates of 3000, 100, and 100 kg/ha respectively & after that same dose of manure were applied fortnightly in order to maintain productivity and buffering capacity of pond water. Water level in the tanks was maintained at 90 ± 2 cm throughout the experimental period, evaporation loss being compensated weekly.

Ponds were allowed for 7 days for the good growth of periphyton on bagasse bundles and planktons in water column. Fingerlings of two indigenous fishes Labeo *fimbriatus* (35.97±0.61mm; 11.89±0.67g), *Barbodes carnaticus* (13.39±0.09mm; 0.69±0.02g) and freshwater prawn larvae of *Macrobrachium rosennbergii* (38.66±0.88; 9.01±0.28g) were stocked at ratio of 3:3:4 respectively, with a total stocking density of 10,000/ha (i. e., 6, 6 & 8 nos. per pond respectively). Pelleted sinking feed of 30% protein were given to the treatment T_0 and T_2 at 5% of body weight in two weeks, there after it was reduced to 2% as per the body weight. The fishes and prawn were fed two times a day during the experimental period.

2.3 Water Quality Monitoring

Water samples were collected on weekly basis & analyzed for parameters like transparency (sechhi disc), temperature (mercury thermometer) and pH. Chemical parameters like total ammonia-N (TAN), nitrite-N (NO_2-N), nitrate-N (NO_3-N) and phosphate-phosphorus (PO_4-P) were determined following APHA (2017) [6].

2.4 Plankton Sampling

Dry matter, ash, AFDM, chlorophyll-a and pheophytin of plankton were determined every week. For plankton analysis, 50L of water sample was taken from five different places of the pond and passed through fine (50µ) mesh plankton net. Filtered samples were taken into a measuring cylinder and carefully made up to a standard volume of 50ml. Then the collected plankton samples were preserved in 5% buffered formalin in small plastic bottles for subsequent studies. For quantitative study, from each 50 ml preserved sample, 1 ml sub-sample was examined using a Sedge Wick-Rafter Cell (S-R Cell) and a binocular microscope (Olympus CH-40). One ml sub sample from each sample was transferred to the cell and then all planktonic organisms present in 100 squares were identified and counted. The quantitative estimation of plankton was done using the following formula:

$$N = (n \times v) / V$$

Where, N= Total number of plankton cells per liter of original water; n= Average number of plankton counted in 100 fields; v= Volume of final concentrate of the sample (ml); V= Volume of total pond water filtered (L). In addition to this, 5L water was passed through 50µ net and filtrate samples were dried and ignited for DM, Ash and AFDM. Chlorophyll-a and phaeophytin analysis of plankton were performed (APHA, 2017) [6].

2.5 Periphyton Sampling

Dry matter, ash, Ash free dry matter, pigment analysis of periphyton was done on weekly basis. Periphyton collection started from 7th day onwards after stocking of substrates. Periphyton was scraped from a 2x2 cm band with the help of scalpel blade from three randomly selected bundles at three depths 25, 50 and 75 cm along the length of the bundle, starting at the water surface in each pond. Care being taken not to remove any of the substrate itself. Samples from different depths and different poles were mixed thoroughly as per individual pond. One third of the mixed sample (pond-wise) was used to determine chlorophyll-a and pheophytin content by following standard methods (APHA 2017) [6]. The remaining periphyton sample was then used for dry matter determination. For this, samples were dried at 100°C to constant weight and the ash content was determined in a muffle furnace (4 h at 550°C). Ash free dry matter (AFDM) was calculated by subtracting the ash value from the dry matter content. Further remaining sample was pooled treatment-wise, used to study the taxonomic composition of periphyton. Taxa of both plankton and periphyton were identified to genus level using keys from Ward and Whipple (1959) [7], Prescott (1962) [8], Belcher and Swale (1976) [9] and Bellinger (1992) [10].

2.6 Statistical Analysis

All statistical analyses were done using SPSS 20.0. Pearson's coefficient of correlation was performed to find correlation among physico-chemical, biological parameters. Significant difference between the treatments were studied by One-way analysis of variance (ANOVA), followed by Duncan's multiple range test at $P < 0.05$.

3. Results

3.1 Water Quality Parameters

For all parameters like transparency (22.07-24.70cm), pH

(6.73 – 8.23), dissolved oxygen (6.07-8.53 mg/l) total alkalinity (120.00-215.67 mg/l) and hardness (73.33-108.33 mg/l), also nutrients such as NH_3-N (0.068-0.167 mg/l), NO_2-N (0.046-0.094 mg/l) NO_3-N (0.069-0.99 mg/l) and PO_4-P (0.019-0.060 mg/l) significant difference between treatments and control ponds ($P<0.05$) was observed except for water temperature (20.03-29.07°C). Physico-chemical characteristics of water recorded in the study are given in Table 1. Initially, low DO concentration was observed and but overall DO concentration was found high in T_1 and T_2 than T_0. However, low water pH and transparency was observed in substrate treatments (T_1 and T_2) than the control. Total alkalinity concentration was found high in substrate treatments compared to control, whereas reverse trend observed for total hardness. The nutrients concentration NH_3-N, NO_2-N and PO_4-P was found low in substrate, however NO_3-N found high in substrate ponds.

Table 1: Water Quality Parameters

Parameters	T_0	T_1	T_2
Temperature (°C)	26.30 ± 0.45	25.60 ± 0.49	25.50 ± 0.48
Transparency (cm)	24.0 ± 0.03	23.50 ± 0.06	23.10 ± 0.14
pH	8.0 ± 0.03	8.7 ± 0.07	8.7 ± 0.08
DO (mg/l)	7.5 ± 0.15	7.5 ± 0.09	8.0 ± 0.09
Tot. alkalinity (mg/l)	160.6 ± 4.70	177.4 ± 5.93	191.7 ± 3.79
Tot. hardness (mg/l)	98.4 ± 1.25	87.2 ± 0.61	82.1 ± 1.26
NH_3-N (mg/l)	0.104 ± 0.003	0.089 ± 0.003	0.092 ± 0.002
NO_2-N (mg/l)	0.075 ± 0.002	0.063 ± 0.002	0.064 ± 0.002
NO_3-N (mg/l)	0.090 ± 0.002	0.093 ± 0.001	0.090 ± 0.002
PO_4-P (mg/l)	0.040 ± 0.002	0.036 ± 0.002	0.035 ± 0.002

*The values of different water quality parameters are mean ± SE of three ponds per treatment

3.2 Plankton Biomass and Composition

The variation in plankton biomass is presented in Table 2. DM, ash and AFDM showed initial increasing trend and it decreased near the termination of the study, there is significant difference found between the treatments except for DM. DM and ash was found slightly high in feed based tanks (T_0 & T_2) than only substrate tank (T_1). Similar trend observed for chlorophyll-a and phaeophytin concentration, it increased up to the 126th day of experiment and later it decreased (Fig 1).

In the present study, after initial fertilization and addition of substrates there was a quick increase in phytoplankton density (fig 3) observed. The mean phytoplankton density was found highest in control (T_0) than in substrate tanks T_1 and T_2 treatment however zooplankton density was found higher in substrate treatment ponds compare to control one. Plankton population in the experimental ponds was found to be of 30 genera. Irrespective of treatments, plankton communities consisted of four groups; *Chlorophyceae* (10 genera), *Bacillariophyceae* (7 genera), *Cyanophyceae* (2 genera), *Euglenophyceae* (3 genera) of phytoplankton & 2 groups of zooplankton; *Rotifera* (4 genera), *Crustacea* (4 genera) were found (Table 4). For entire period of study, *Chlorophyceae* group (30%) was dominant followed by *Bacillariophyceae*. In the zooplankton, rotifers were dominated in T_0, whereas crustacean in substrate ponds (Fig 5A).

Table 2: Plankton Biomass and Pigment Concentration

Parameters	T_0	T_1	T_2
DM (mg/l)	0.89 ± 0.06	0.91 ± 0.06	0.89 ± 0.06
Ash (mg/l)	0.23 ± 0.02	0.24 ± 0.02	0.24 ± 0.01
AFDM (mg/l)	0.66 ± 0.04	0.67 ± 0.05	0.65 ± 0.05
Chlorophyll-a (µg/l)	1.52 ± 0.10	1.65 ± 0.11	1.68 ± 0.12
Phaeophytin (µg/l)	0.88 ± 0.09	0.95 ± 0.10	0.89 ± 0.09

*The values of are mean ± SE of three ponds per treatment

3.3 Periphyton Biomass and Composition

The variation in periphyton biomass is presented in Table 3. DM, ash and AFDM showed initial increasing trend, later it decreased near the end of the experiment, there is no significant difference found between the treatments. DM and ash was found slightly high in feed based substrate tanks than only substrate tanks. Chlorophyll-a and phaeophytin concentration increased up to the 126th day of the experiment and later it decreased (Fig 2).

Phytoperiphyton density was high in T_2 than T_1; however zooperiphyton does not show any difference in two treatments (Fig 4). The highest phytoperiphyton number was observed on 70th day of study. During initial period of study, *Bacillariophyceae* was dominated group and after 70th day *Chlorophyceae* showed dominancy over the other groups. Overall, *Chlorophyceae* group (27%) was dominant in the zooperiphyton, rotifers were dominated followed by crustaceans (Fig 5B). There were almost 25 genera of phytoperiphyton and 8 genera of zooperiphyton found during study period (Table 4).

Table 3: Periphyton Biomass and Pigment Concentration

Parameters	T_1	T_2
DM (mg/cm^2)	1.1 ± 0.07	1.1 ± 0.08
Ash (mg/cm^2)	0.3 ± 0.02	0.3 ± 0.02
AFDM (mg/cm^2)	0.8 ± 0.05	0.8 ± 0.05
Chlorophyll-a (µg/cm^2)	2.9 ± 0.17	2.6 ± 0.19
Phaeophytin (µg/cm^2)	1.0 ± 0.12	0.8 ± 0.10

*The values of are mean ± SE of three ponds per treatment

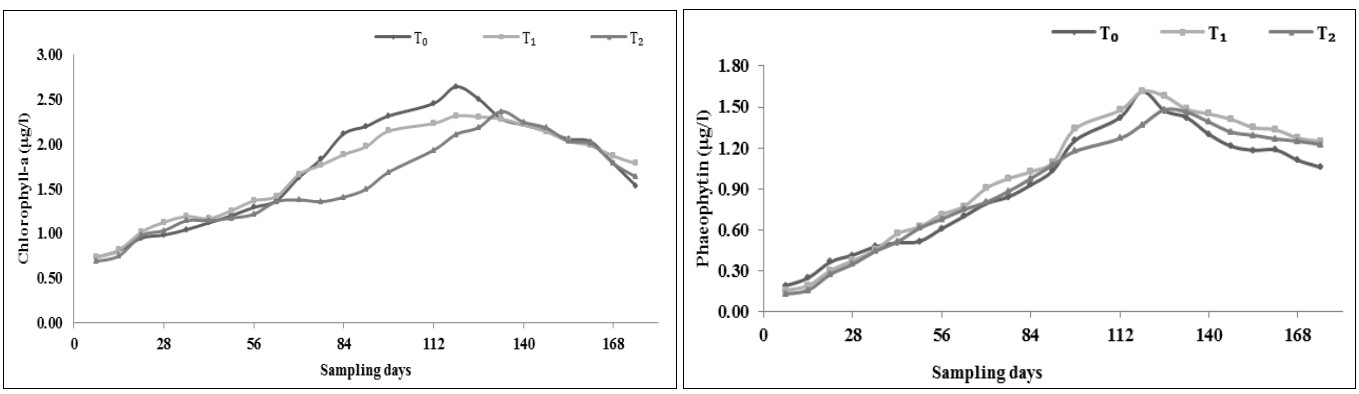

Fig 1: Pigment concentration (µg/l) of Plankton

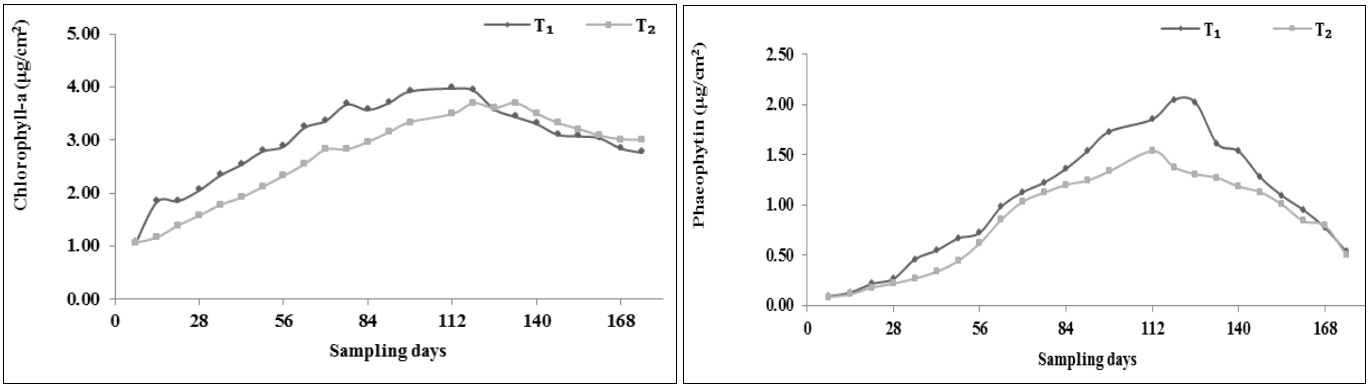

Fig 2: Pigment concentration (µg/cm^2) of Periphyton

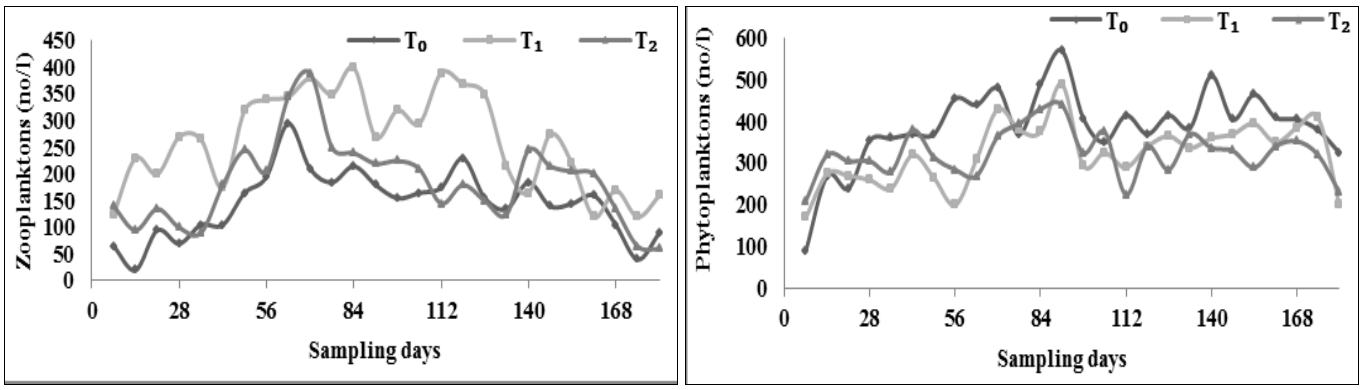

Fig 3: Total phytoplankton and zooplankton density (no/l)

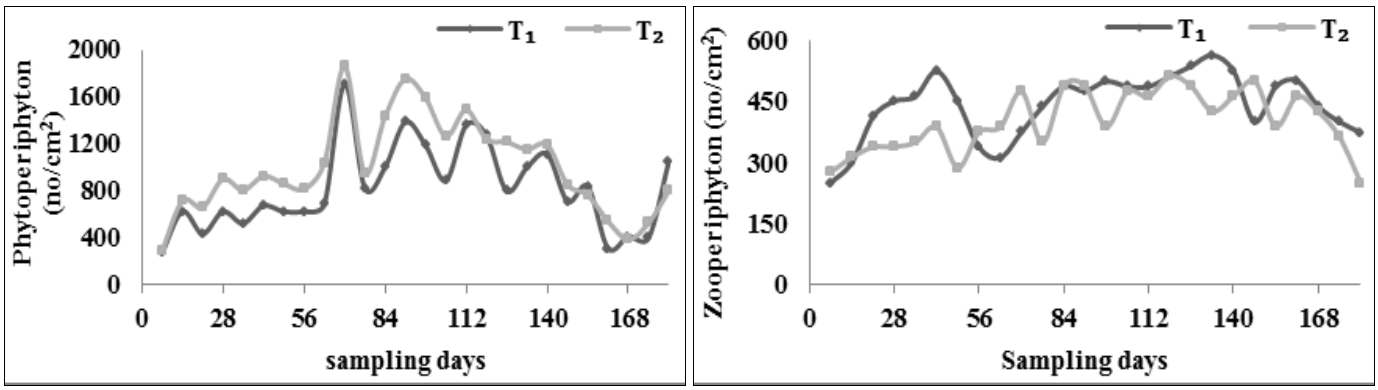

Fig 4: Total phytoperiphyton and zooperiphyton density (no/cm^2)

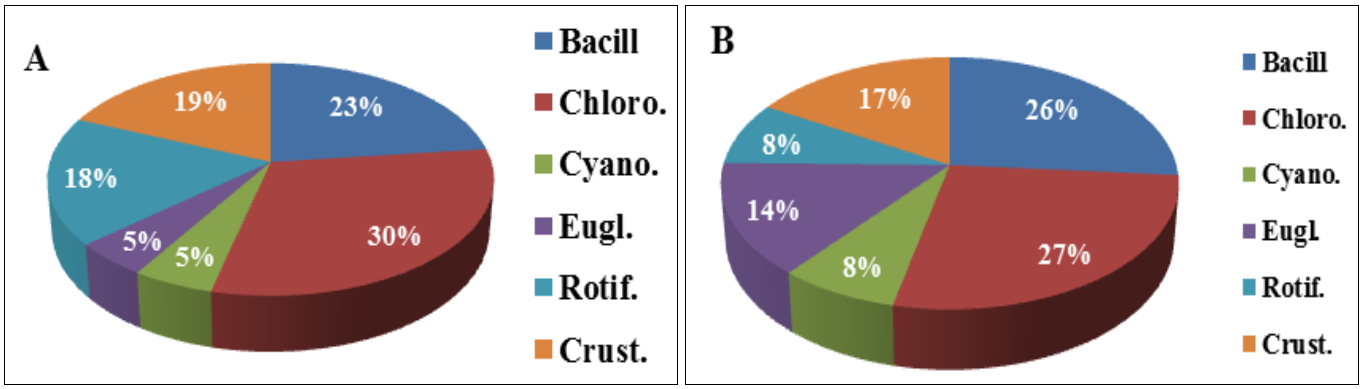

Fig 5: Graphical representation of plankton (A) and periphyton (B) composition recorded during the experiment

Table 4: Generic status of phytoplankton and zooplankton found in the experimental ponds

Chlorophyceae	***Bacillariophyceae***	***Rotifera***
Chlorella sp	*Cymbella sp*	*Asplanchna sp*
Ankistrodesmus sp	*Cyclotella sp*	*Brachionus sp*
Closterium sp	*Melosira sp*	*Keratella sp*
Coelastrum sp	*Fragillaria sp*	*Polyarthra sp*
Microspora sp	*Navicula sp.*	
Spirogyra sp	*Nitzschia sp*	***Crustacea***
Scenedesmus sp	*Gomphonema sp*	*Cyclops sp*
Oocystis sp	*Synedra sp*	*Diaptomus sp*
Ulothrix sp		*Ceriodaphnia sp*
Oedogonium sp	***Euglenophyceae***	*Moina sp*
Pediastrum sp	*Euglena sp*	
	Trachelomonas sp	
Cyanophyceae	*Phacus sp*	
Anabaena sp		
Microcystis sp		
Oscillatoria sp		

4. Discussion

4.1 Water Quality Parameters

The water quality parameters were found within the suitable range of prawn polyculture (Fair and Foftner, 1981) [11]. Though slightly lower water temperature was recorded in tanks with substrate but did not show significant ($p<0.05$) difference from other treatments. Lower temperature in substrate tanks might be the reason for shading effect of substrates (Keshavanath, 2002) [12]. Transparency in substrate ponds were less than that of control. Azim *et al.* (2003) [13] have pointed out inverse relationship between periphyton growth and water transparency. Bergman (1999) [14] was observed that as the concentration of phosphorus and chlorophyll increased, the depth transparency of Secchi decreased significantly. In the present study the mean pH values were recorded within the range of 6.73 – 8.23 slightly alkaline in substrate based tanks, which indicates favourable conditions for biological production (Mridula *et al.* 2003) [15]. DO ranges from 6.07 – 8.53, mostly substrate based ponds show high amount of DO than that of control. Substrates help to maintain good water quality by promoting the absorption of nitrogen compounds and generating high levels of dissolved oxygen related to the production of autotrophic microorganisms (Jesna *et al.*, 2018) [16]. Dissolved oxygen shown positive relationship with pH ($r = 0.643$).

According to Boyd (1982) [17], total alkalinity should be more than 20 ppm in fertilized ponds and total alkalinity was recorded 120.00-215.67 mg/l l in the study ponds seems productive. In terms of oxygen production and photosynthesis, water with higher alkalinity is considered to be more productive, which also shows that the nutrient turnover rate and productivity due to presence of substrate in ponds (Gangadhar *et al.*, 2012) [18]. The positive relationship was found between phytoplankton density and hardness (Jasmine *et al.*, 2013) [19], this might be the reason for high hardness in control group, as phytoplankton density was found more in control rather than substrate ponds. Similar values were also recorded by Gogoi *et al.* (2018) [20]. The overall low ammonia, nitrite concentration was observed in the substrate treatment than in the control group may be due to higher rate of nitrification due to substrates. Significantly higher nitrate level in T_1 and T_2 was attributed with high densities of *Chlorophyceae* (Sipaúba, 2011) [21]. Phosphorus is a key nutrient which regulates the growth of phytoplankton in aquaculture ponds. It mainly stimulates phytoplankton growth and increase the base of the food web. The concentrations of phosphate-phosphorus was found slightly high but later throughout the study period it remained low in substrate ponds compared to control. This suggests an increase in the organic matter's mineralization rate due to the higher heterotrophic bacterial biomass present in the substrate tanks (Baloi, 2013) [22].

4.2 Plankton Composition and Biomass

After first sampling, DM, ash and AFDM increased, become steady and decreased towards the end of the experiment. It showed negative correlation with nitrite and ammonia. Substrate treatment recorded high dry mass and ash of plankton. This attribute the higher grazing on periphyton in the bagasse treatment. However, the amount of plankton DM, ash, AFDM decreases as the experiment goes on which indicates that the fish consume both plankton and phytoplankton (Keshavanath *et al.*, 2015) [23]. Similar results were observed in bagasse ponds by Gangadhar *et al.* (2012) [18].

Both Chlorophyll-a and phaeophytin concentration was found high in substrate group (T_1 and T_2) than control (T_0). This might be due to acceleration in the biochemical cycling of nutrients in the culture system (Moss and Moss, 2004) [24]. Also, the abundance of phytoplankton, which produced dense algal surface film especially, high *Chlorophyceae* leads to enhance chlorophyll-a concentration (Rahman *et al.*, 2016) [25]. In addition to this, low ammonia and nitrite nitrogenous waste might be the reason to enhance pigment content in substrate based ponds (Azim *et al.*, 2002; Mridula *et al.*, 2006) [26, 27]. Similar pattern observed by Jana *et al.* (2006) [28] in inland saline groundwater ponds with bamboo poles as substrate.

Fertilization in aquaculture ponds increases the productivity of phytoplankton, which is the food basis for zooplankton and benthic animals. In the present study, after initial fertilization and addition of substrate, there was a quick increase of phytoplankton density (fig 3) observed. The mean phytoplankton density was found highest in the control (T_0) than in substrate T_1 and T_2 treatment. This was mainly due to predominantly higher periphyton biomass in the substrate treatments would have suppressed phytoplankton population competing for micronutrient. Also, shading effect of substrate leads to the dark colour in the substrate ponds might hide light penetration which otherwise promote phytoplankton growth (Ramesh *et al.*, 1999) [29]. Similar trend were recorded by Langis *et al.* (1988) [30]; Umesh *et al.* (1999) [31].

Phytoplankton showed weekly fluctuations throughout the experiment in all treatments, peak observed on 70th day. The dominant group observed was *chlorophyceae* followed by *Bacillariophyceae*, *Cyanophyceae* and *Euglenophyceae* in all the treatments throughout the experiment. Similarly Bharti *et al.* (2016) [32] reported planktonic communities in fibre reinforced plastic (FRP) tanks from four different substrates namely, paddy straw, sugarcane from four groups *viz*. *Bacillariophyceae* (7 genera), *Chlorophyceae* (10 genera), *Cyanophyceae* (2 genera) and *Euglenophyceae* (1 genus). Present findings are similar with Azim *et al*. (2001, 2002a) [33, 34]. Wahab *et al.* (1999) [35]; Pandey *et al*. (2014) [36]; Gogoi *et al*. (2018) [20], where they recorded *Chlorophyceae* as the most dominant among different plankton groups.

Generally green algae is most abundant in water which is not contaminated with organic matter while, blue green algae, euglenids and diatoms prefer water, which is rich in organic matter (Subrahmanyan, 1959) [37]. High abundance of *Chlorophyceae* generally, attributed to favorable water quality attributes, particularly high levels of total alkalinity observed during the study by Akter *et al*, (2018) [38]. In all three treatments, green algae was the dominant one. Among *Chlorophyceae*, the most dominant taxa were *scenedesmus sp* and *microspore sp*. Chari (1980) [39], stated there was direct relationship observed between nitrate nitrogen and the occurrence of *Chlorophyceae*.

As shown by many studies, zooplanktons are usually the first source of exogenous nutrition for larval fish (Mills, 1986)[40]. Also, Abundance of zooplankton not only depends on phytoplankton availability, but other ecological parameters. In aquatic system, the density of zooplankton is a more reliable index of productivity. In whole study period, zooplankton production showed sustainability, this could be due to the periodical addition of fertilizers and periphyton development on substrate. The zooplankton production was found high in substrate based ponds of T_1 and T_2 than that of control. The higher zooplankton production in substrate treatments might be due to higher periphyton density on a substrate that could have served food for zooplankton. Similar results were obtained by Rai *et al*. (2008) [41]; Ahsan *et al.* (2014) [42]; Brahmchari *et al.* (2018) [43]. In the quantitative study of zooplankton, rotifera and crustacean were most dominantly found in control and substrate ponds respectively. In the present study a total of 8 genera of animal community belonging to rotifera (4), copepoda (2) and cladocera (2) were recorded. Among rotifera, *Keratella sp.*, *Brachionus sp.*, and that of crustacean, *Cyclops sp*., were most frequently occurred. According to the research of Sutela (1997) [44], fish larvae are preyed by rotifers only when there is a shortage of crustaceans. The choice depends on the caloric value of the prey, which is higher in crustaceans than in rotifers. In addition to this, cladocerans were represented by Daphnia and Moina, they generally grow by utilizing organic matter. So this attribute to enhance their population in substrate based tanks.

4.3 Periphyton Composition and Biomass

The biomass of periphyton may differ as per habitat, taxonomic composition, fertilization level and substrate type (Azim and Asaeda 2004b) [45]. The comparative higher DM and ash was observed in feed based treatment than only substrate treatment. Keshavanath *et al*. (2001) [5] observed mixing of bagasse material and periphyton, during scrapping periphyton from bagasse, which leads to enhance the ash content. Also, decreasing content of AFDM indicates appropriate grazing pressure by growing organisms in the pond. Total phytoperiphyton density was found higher in T_2 than T_1. It was mainly due to less grazing by fishes on periphyton in T_2 as there was the availability of supplementary feed as well as the extra nutrients received through the leftover supplementary feed (Mridula *et al*., 2006) [27]. The increase in periphyton biomass indicates that the initial fish has a low overall biomass and low grazing pressure on the surrounding periphyton, while the stable or low periphyton biomass is due to the higher fish biomass at the selected stocking density (Biswas *et al*., 2017) [46].

Chlorophyll-a value is an indicator of pond productivity and has an inverse relationship with water transparency (Ahmed 1993) [47]. The high chlorophyll-a content observed in T_1 indicates the production of phytoplankton, which indicates a positive effect on the nutritional quality of plankton (Azim 2002) [26]. Both pigment concentrations were found to decrease at the end of the study, which attributes to increase in grazing pressure. Similar observation recorded by Garg & Bhatnagar (2000) [48].

The Abu & Saleha (2005) [49] reported an inverse relationship between *Bacillariophyceae* and water temperature. This could be the reason for the high number of *Bacillariophyceae* during the initial period of the study as there was the low temperature initially due to the winter season. Later, in summer temperature increased, So the *Chlorophyceae* showed dominancy over *Bacillariophyceae.* According to Khatoon *et al*., (2007) [50], substrates based ponds, showed high proliferation of *Chlorophyceae* and *Bacillariophyceae* was mainly due to the accumulation of nitrate in the periphytic biofilm formed in the culture system.

5. Conclusion

In periphyton based system, attached algae, zooplankton and invertebrates easily colonize the substrate in the water column, on which fish can graze more efficiently. Moreover, these periphyton acts as nutritious feed for much of the zooplankton. So, the more quantity of zooplankton on substrate helps to improve growth as well as nutrition of cultured species. This study concludes that use of the substrate along with regular fertilization creates.

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7. References

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