International Journal of Fisheries and Aquatic Studies 2014; 1(4): 12-19

ISSN: 2347-5129
IJFAS 2014; 1(4): 12-19

www.fisheriesjournal.com
Received: 01-02-2014
Accepted: 15-02-2014

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Management of Euglenophytes Bloom in Fish Pond at Rajshahi, North-west Part of Bangladesh

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ABSTRACT
An attempt was made to study the management of euglenophytes bloom in fish ponds by application of duckweed and lime. The study was carried out for five months in twelve fish ponds at Rajshahi, Bangladesh under four treatments viz., T1 (the ponds with duckweed, *Lemna* sp.), T2 (The ponds with lime, CaO), T3 (The ponds with both duckweed and lime) and T4 (The ponds without duckweed and lime as control). The study ponds were stocked with *Labeo rohita*, *Catla catla*, *Hypophthalmichthys molitrix*, *Puntius gonionotus* and *Cirrhina mrigala* at 60 juveniles/dec. Water quality parameters, algal community and density, and growth performances of fish were monitored by using standard methods. The results showed that the application of duckweed and lime in the ponds had positive effects on water quality, euglenophytes density and fish growth. Better water quality, lower density of euglenophytes and higher growth of fish were recorded in T3 as compared to other treatments ($P<0.05$). The present study indicates that application of duckweed and lime in combination is better for management of euglenophytes bloom in fish pond.

Keywords: Management, Euglenophytes, bloom, Fish pond, Duckweed, Lime, Combination.

1. Introduction

Eutrophication is considered as one of the most pressing environmental problems in both the developed and the developing countries [44, 19]. Decomposition of organic wastes and unutilized feeds plus direct application of fertilizers are the major causes of eutrophication which leads to thick algal bloom in fish ponds of Bangladesh. Although, algal bloom indicates high productivity of the water body concerned [7] but thick algal bloom causing serious economic losses to aquaculture [18]. Euglenophytes bloom is the common phenomenon in fish ponds of Bangladesh which often leads to water quality degradation and hampered growth of fish [53, 40] even causes mass mortality of fish [39]. Therefore, it is imperative to develop management techniques to control this algal bloom in fish pond.

Several chemical methods are employed to control algal bloom in tropical water bodies [54, 26] but, they are either expensive or have residual effects in the aquatic food chain. Algaecides destroy water quality and add new toxic sediment to the bottom which interfere bacterial decomposition of sediments and fish growth [31]. On the other hand, most of the herbicides are known to have negative effects on fish growth and are not environment friendly [35]. Though, filter-feeding fish are selective algae grazers that can suppress algae through ingestion but they can also be enhanced algal density indirectly by suppressing herbivorous zooplankton and by increasing nutrients availability [12]. Moreover, if the algal bloom is controlled without eliminating the cause (excess nutrients), the algal bloom will quickly return. Thus, it is necessary to find out effective and environment friendly management techniques to control algal bloom.

Development of euglenophytes bloom in the fish ponds is related to higher concentrations of nitrate and phosphate nutrients [28, 40], higher concentrations of heavy metals [22, 13] and acidic pH [37, 40]. Thus, the bloom of these algae can be controlled by reducing nutrients and heavy metals, and by increasing pH of water. For this purpose, duckweeds (*Lemna* spp.) and lime can be applied in the ponds because duckweeds have shown potential usefulness to reduce the nutrients [30, 15] and heavy metals [36, 45] from waste water systems and lime can increase pH to a level suitable for pond productivity [8, 46].

However, concerning the management and control of algal bloom, a numbers of research works have been reported in different countries of the world [11, 34, 32, 33]. But, researches on the management of euglenophytes bloom in farmers' managed fish pond at Rajshahi, Bangladesh are scare. Therefore, the present study was conducted to investigate the management of euglenophytes bloom in fish ponds by application of duckweed and lime.

2. Materials and methods

2.1 Location and design of the study

The study was conducted in the fish ponds at Raighati, Mohanpur Upazila, Rajshahi, North-west part of Bangladesh for five months form August to December 2011. The study was conducted in twelve fish ponds (where the bloom of euglenophytes is developed) under four treatments viz., T1, T2, T3 and T4 (three replicates in each treatment) among which T1 was assigned to the ponds treated with duckweed (*Lemna* sp.); T2 assigned to the ponds treated with lime (CaO); T3 assigned to the ponds treated with both duckweed and lime; and T4 assigned to the ponds treated as control (without duckweed and lime).

2.2 Pond management

Initially, all study ponds were treated with lime at the rate of 1kg/decimal. One fourth of the water surface in the ponds of T1 and T3 was covered with duckweed and maintained it throughout study period. Periodic liming in the ponds of T2 and T3 was done at the rate of 250 g/dec./15 days. The study ponds were stocked with *Labeo rohita* (rohu), *Catla catla* (catla), *Hypophthalmichthys molitrix* (silver carp), *Puntius gonionotus* (silver barb) and *Cirrhina mrigala* (mrigel) at the rate of 60 juveniles/dec. with the ratio of 12:7:13:18:10.

2.3 Monitoring of water quality

The water quality parameters viz., water temperature, pH, dissolved oxygen (DO), nitrate-nitrogen (NO_3-N), ammonium-nitrogen (NH_4-N), phosphate-phosphorus (PO_4-P), iron (Fe), zinc (Zn), manganese (Mn) and copper (Cu) were monitored fortnightly. Water temperature, DO and pH were determined by Celsius thermometer, HACH kit (HANNA, HI-9142) and pH meter (Jenway, 3020 UK), respectively. The concentrations of NO_3-N, NH_4-N and PO_4-P were determined by using HACH kit (DR/2010). The concentrations of Fe, Zn, Mn and Cu were determined by Atomic Absorption Spectrophotometer (Model-3310).

2.4 Study of planktonic algae

Water samples (15 Liters) were collected from different depth of each pond and passed through the plankton net (25 µm mesh size). The concentrated algae samples were preserved with 5% buffered formalin. Then the concentrated samples were examined using Sedgewick-Rafter counting cell (S-R cell) under a compound microscope (NOVEX). Identification of algae was performed according to APHA [2] and Bellinger [6]. The enumeration of algae was done according to Stirling [48].

2.5 Monitoring of fish growth

For monitoring of growth, about 10% fish from each pond were sampled and weighted monthly. The growth performance in terms of mean weight gain (MWG), average daily weight gain (ADWG) and specific growth rate (SGR) were evaluated by using following formulae.

MWG (g) = Mean final weight (g) – Mean initial weight (g).

ADWG (gbwd^{-1}) = {Mean final weight – Mean initial weight}/Number of days

$$\text{SGR (\% bwd}^{-1}\text{)} = \frac{\log_e W_2 - \log_e W_1}{T_2 - T_1} \times 100$$

[W_1 = Initial body weight (g) at T_1 (day), W_2 = Initial body weight (g) at T_2 (day)]

2.6 Statistical analysis

For the statistical analysis of collected data, one way analysis of variance (ANOVA) was performed using software SPSS (Statistical Package for Social Science, version 16.0). The mean values were compared to see the significant difference from the DMRT (Duncan Multiple Range Test). Significance was assigned at the 0.05 level.

3. Results

3.1 Water quality parameters

The monitored data of water quality parameters in the study ponds under four treatments are shown in Table 1. The water temperatures among the treatments did not show any significant difference. It was over 32.0 ^{0}C in mid of August and below 19.5 ^{0}C in end of December. The mean concentration of DO was relatively high in T1 and T3 followed by T2 whereas in T4, it was significantly low. The mean value of pH was significantly high in T2 and T3 followed by T1 whereas in T4, it was significantly low. The mean concentrations of NO_3-N, NH_4-N and PO_4-P) were significantly high in T4 followed by T2 whereas in T1 and T3, these nutrients were significantly low. Like nitrogen and phosphorus nutrients, the mean concentrations of heavy metals (Fe, Zn, Mn and Zn) were significantly high in T4 followed by T2 whereas in T1 and T3, these heavy metals were relatively low.

Table 1: Water quality parameters in the study ponds under four treatments

Parameters	Treatments			
	T1	T2	T3	T4
Temperature (^{0}C)	28.37±4.39^a	28.38±4.43^a	28.37±4.44^a	28.36±4.43^a
DO (mg/l)	5.54±0.44^a	5.39±0.43^a	5.53±0.35^a	4.72±0.51^b
pH	7.08±0.26^b	7.49±0.34^a	7.69±0.41^a	6.21±0.34^c
NO_3-N (mg/l)	0.74±0.19^c	1.13±0.17^b	0.71±0.24^c	1.41±0.26^a
NH_4-N (mg/l)	0.57±0.23^c	0.87±0.18^b	0.54±0.25^c	1.34±0.18^a
PO_4-P (mg/l))	0.76±0.20^c	1.14±0.22^b	0.69±0.25^c	1.61±0.39^a
Fe (mg/l)	0.29±0.09^c	0.36±0.07^b	0.24±0.10^c	0.69±0.08^a
Zn (mg/l)	0.14±0.05^c	0.23±0.05^b	0.15±0.04^c	0.33±0.06^a
Mn (mg/l))	0.15±0.05^c	0.20 ±0.04^b	0.14±0.05^c	0.29±0.05^a
Cu (mg/l)	0.13±0.03^c	0.17±0.04^b	0.12±0.05^b	0.26±0.03^a

* T1: The ponds with duckweed, T2: The ponds with lime, T3: The ponds with duckweed and lime, and T4: The ponds without duckweed and lime (Control ponds).

* Values of water quality parameters are mean of triplicate determination. Values in the same row with different superscripts are significantly different (P<0.05).

3.2 Community of planktonic algae

Total 29 genera of planktonic algae belonging to euglenophytes, cyanophytes, chlorophytes and bacillariophytes were recorded during the study period (Table 2). There was no significant difference in the number of planktonic algal genera in T1, T2 and T3 but relatively lower number was recorded in T4. The genera numbers were varied from 23-29, 22-27, 23-28 and 14-21 in T1, T2, T3 and T4, respectively. Among the algal groups, chlorophytes had the maximum number of genera (11) followed by cyanophytes (8) and bacillariophytes (7) whereas euglenophytes had the minimum number of genera (3) in all the treatments.

Table 2: Planktonic algal genera found in the study ponds under four treatments

Group of algae	Genera under each group
Euglenophytes	*Euglena*, *Phacus* and *Trachelomonas*
Cyanophytes	*Anabaena, Anabaenopsis, Aphanizomenon, Aphanocapsa, Chroococcus, Gomphosphaeria, Oscillatoria* and *Microcystis*
Chlorophytes	*Chlorella, Closterium, Coelastrum, Pediastrum, Scenedesmus, Spirogyra*, *Staurastrum*, *Teraedon, Ulothrix*, *Volvox* and *Zygnema*
Bacillariophytes	*Asterionella, Cyclotella, Fragilaria, Navicula, Nitzschia*, *Synedra* and *Tabellaria*

3.3 Density of euglenophytes

During the study tenure, the density of euglenophytes was found to be ranged from 8.31 to 9.49, 7.12 to 9.61, 4.02 to 9.49 and 11.14 to 29.22 x 10^4 cells/l in T1, T2, T3 and T4, respectively. Significantly higher mean density of these algae was recorded in T4 followed by T1 and T2 whereas the lower mean density was recorded in T3 (Table 3). The average percent contributions of these algae in the total algae were 49.64, 49.24, 37.25 and 69.54% in T1, T2, T3 and T4, respectively.

3.4 Density of cyanophytes, chlorophytes and chlorophytes

The density of cyanophytes showed no significant difference among the treatments but the mean density was relatively high in T3 followed by T1 and T2 whereas in T4, the density of these algae were relatively low (Table 3). The mean density of chlorophytes showed no significant difference in T1, T2 and T3 but, it was significantly low in T4. Significantly higher mean density of bacillariophytes was recorded in T1 followed by T3 and T2 whereas the mean density was relatively low in T4 (Table 3).

Table 3: Mean density of different groups of planktonic algae in four treatments

Algal group (x 10^4 cells/l)	Treatment			
	T1	T2	T3	T4
Euglenophytes	8.87±1.58^b	8.15±2.23^b	5.40±2.13^c	18.97±6.78^a
Cyanophytes	5.21±1.92^a	5.07±1.54^a	5.43±1.83^a	5.04±1.57^a
Chlorophytes	3.30±0.61^a	2.96±0.36^a	3.11±0.64^a	2.33±0.37^b
Bacillariophytes	0.60±0.11^a	0.48±0.09^b	0.55±0.15^{ab}	0.21±0.08^c
Total algae	17.99±2.25^b	16.66± 2.97^{bc}	14.49±2.89^c	26.55±5.65^a

* T1: The ponds with duckweed, T2: The ponds with lime, T3: The ponds with duckweed and lime, and T4: The ponds without duckweed and lime (Control ponds).

* Values of algal density are mean of triplicate determination. Values in the same row with different superscripts are significantly different (P<0.05).

3.5 Growth performance of fish

The results of growth performances of the fish are shown in Table 4. Significantly higher value of MWG, ADWG and SGR were recorded in T3 for all experimental fish species followed by T1 and T2 whereas in T4, the values of these parameters were quietly low. Among the fish species, the ADWG value of silver carp was relatively high but it was relatively low for silver barb. Silver barb showed the maximum SGR in T1 and T3 whereas rohu showed the minimum SGR in T4 and T2. There was no significant difference in survival rates of the fishes in T1, T2 and T3 but survival rate was significantly low in T4. The values of survival rate were 90, 89, 93 and 82% in T1, T2, T3 and T4, respectively.

Table 4: Growth parameters of the fish species in four treatments

Parameter	Fish species	Treatment			
		T1	T2	T3	T4
Initial weight (g)	Rohu	25.54±6.25	25.50±7.13	25.53±7.10	25.56±6.84
	Catla	29.25±7.14	29.27 ±6.28	29.31±7.11	29.29±7.16
	Mrigal	15.70±4.21	15.68 ±5.02	17.71±4.52	17.69 ±4.94
	Silver carp	17.85±4.03	17.80 ±3.44	17.82±4.12	17.83±3.17
	Silver barb	4.25±1.02	4.20 ±1.12	4.19±1.09	4.23±1.13
Final weight (g)	Rohu	209.02±15.84	196.96 ±17.25	233.71±18.95	176.67±18.69
	Catla	271.51±18.70	261.09 ±19.77	301.31±22.36	220.10±16.41
	Mrigal	199.06±12.43	192.03 ±13.61	218.77±15.73	172.60±18.55
	Silver carp	393.36±18.14	376.53 ±19.91	401.11±23.10	351.09±18.45
	Silver barb	159.51±15.46	141.93 ±18.49	166.32±15.16	127.40±17.54
Weight gain (g)	Rohu	183.38±19.24^b	171.32±18.25^b	208.07±21.67^a	151.03±19.47^c
	Catla	242.24±20.32^b	231.82±18.52^b	272.04±21.15^a	190.83±20.13^c
	Mrigal	183.36±20.21^b	176.33±19.78^b	203.07±17.25^a	156.90±21.25^c
	Silver carp	375.54±25.14^b	358.71±27.25^b	391.29±26.78^a	333.27±24.18^c
	Silver barb	157.31±16.25^b	139.73±22.10^b	164.12±19.25^a	125.20±18.39^c
Daily weight gain ($gbwd^{-1}$)	Rohu	1.22 ±0.04^b	1.14±0.05^b	1.39±0.06^a	1.01±0.06^c
	Catla	1.61±0.05^b	1.55±0.04^b	1.81±0.07^a	1.27±0.04^c
	Mrigal	1.22±0.06^b	1.18±0.02^b	1.35±0.04^a	1.05±0.06^c
	Silver carp	2.50±0.08^b	2.39±0.04^b	2.61±0.07^a	2.22±0.05^c
	Silver barb	1.04±0.04^b	0.92±0.06^b	1.08±0.03^a	0.82±0.05^c
SGR (% bwd^{-1})	Rohu	1.40±0.02^b	1.36±0.02^b	1.47±0.03^a	1.29±0.03^c
	Catla	1.48±0.02^b	1.46±0.02^b	1.55±0.02^a	1.34±0.02^c
	Mrigal	1.69±0.03^b	1.67±0.01^b	1.76±0.02^a	1.60±0.03^c
	Silver carp	2.06 ±0.02^{ab}	2.03±0.01^b	2.09±0.02^a	1.99±0.01^c
	Silver barb	2.42±0.02^a	2.35±0.04^b	2.45±0.02^a	2.27±0.03^c

* Values of growth parameters are mean of triplicate determination. Values with different superscripts in the same row varied significantly (P<0.05).

4. Discussion

4.1 Water quality parameters

Thick algal bloom is known to negatively affect water quality [3, 40]. Concurrently, duckweed and lime have potential usefulness to improve water quality [46, 51]. In the present study, a considerable variation was observed in the water quality parameters (except water temperature) in response to the application of duckweed and lime (separately or in combination) in the bloom ponds.

During this study, there were no significant differences in water temperature among the treatments which indicated that application of duckweed and lime had no significant effect on water temperature. But, it was found to vary from 19.25 to 32.5 °C which might be due to the changes of weather condition from summer to winter season. The water temperatures in four treatments were within the productive range according to the report of Jhingran [25] who reported that water temperature as 18.5 to 37.5 °C is favorable for ponds productivity.

Dissolved oxygen, 5.0 to 7.0 mg/l is considered as fair or good in respect of productivity [41, 42]. Significantly higher DO concentrations were recorded in T1, T2 and T3 as compared to T4. This might be due to addition of oxygen through duckweed, and to the reduction of BOD and COD through duckweed and lime. This assumption is supported by the pervious reports that duckweed supplied additional oxygen in water [30] whereas duckweed and lime reduced BOD and COD [7, 17]. On the other hand, significantly lower DO was recorded in T4 which might be due to the bloom of euglenophytes. This finding is in conformity with the previous report [39] that euglenophytes bloom caused serious oxygen depletion in fish pond.

pH is considered as an important factor for aquaculture [8]. In the present study, significantly higher pH recorded in T2 and T3 which might be due to the regular application of lime. This finding is supported by Boyd and Tucker [7] who stated that lime improves water quality by raising pH. The pH in T1 was also within the acceptable range. This might be due to the application of duckweed which increased pH through utilization of free carbon dioxide in water. On the other hand, the pH values were almost below 6.5 in T4 which might be due to lower dissolved oxygen and higher carbon dioxide concentrations. Tucker [50] reported that pH in water has a direct relation with dissolved oxygen and an inverse relation with free carbon dioxide concentration. This report is supportive to the present study.

Duckweed may improve water quality in eutrophic ponds through absorbing nutrients [14]. During the study tenure, significantly lower concentrations of NO_3-N, NH_4-N and PO_4-P were recorded in T1 and T3 as compared to T2 and T4. This might be due to absorption of these nutrients by duckweed. This assumption is supported by the previous reports [24, 4] that duckweeds have potential usefulness in absorbing nutrients from waste water systems. Ferdoushi *et al*. [15] reported that aquatic macrophytes (*Lemna* sp. and *Azolla* sp.) appeared as a nutrient filter for absorption of nitrogen and phosphorus. Perniel *et al*. [38] reported that *Lemna* consistently removed the largest amount of ammonia and phosphorus from eutrophic storm water. According to previous reports and the result of the present study, it can be stated that application of duckweed in euglenophytes bloom ponds reduced excess nutrients.

Aquatic macrophytes found to be the potential scavengers of heavy metals from aquatic environment [1, 27]. In the present study, Fe, Zn, Mn and Cu concentrations were significantly lower in T1 and T3 as compared to T2 and T4 which might be due to absorption of these heavy metals by duckweeds. Supporting evidence to this assumption can be drawn from previous studies that duckweed can be used to remove heavy metals [14, 10]. Moreover, application of lime might contributed to decrease heavy metals ions from water as confirmed by lower concentrations of heavy metals in the lime treated ponds (T2) than the control ponds (T4). Sipauba-Tavares *et al*. [46] reported that liming reduced heavy metals in the pond sediment and water. This report is supportive to the present study.

4.2 Planktonic algal community

Planktonic algal community structure is regulated by environmental factors, growth rate of algal species and specific rate of loss attributed to grazing, sedimentation and dilution [16]. The present study showed that the number of algal genera in T4 were relatively low as compared to T1, T2 and T3. This might be due to the variation in ambient environmental factors. This assumption is consistent with the earlier findings that aquatic environment is subject to high temporal variation with frequent reorganization of algal communities, as a result of interaction among physical, chemical and biological factors [43].

4.3 Density of euglenophytes

The present study showed that the density and percent contribution of euglenophytes were significantly high in T4 followed by T1 and T2 but the density and percent contribution of these algae were significantly low in T3. Significantly higher density of euglenophytes in T4 might be due to acidic pH, higher nutrients and heavy metal concentrations as confirmed by the water quality parameters of that treatment. This assumption is supported by the previous reports that the density of euglenophytes is increased in acidic pH [37, 55] with higher nutrients [40, 29] and heavy metal concentrations [13, 22]. On the contrary, lower density of euglenophytes in T3 might be due to alkaline pH (>7.0) and lower concentrations of nutrients and heavy metals which might hamper the growth of euglenophytes. This speculation is supported by the previous studies [13, 40] that decreasing concentration of nutrients and heavy metals, and alkaline pH collapsed the luxurious growth of euglenophytes. The application of both duckweed and lime in T3 contributed to reduce nutrients and heavy metals concentrations and to increase pH as confirmed by the water quality parameters. Moreover, duckweeds might be disturbed the ratio of nitrogen to phosphorous through ammonium reduction which might also contributed to reduce euglenophytes density.

During this study, it was also observed that nutrients and heavy metals concentrations in T1 were relatively low due to application of duckweed, but relatively lower pH values in that treatment might increase density of euglenophytes than that of T3. On the other hand, pH values were relatively high (>7.0) in T2 due to application of lime, but relatively higher nutrients and heavy metal concentrations in that treatment might also increase density of these algae than that of T3. Thus the results of this study revealed that application of duckweed and lime (in combination) was better to reduce euglenophytes density as compared to application of duckweed and lime separately.

4.4 Density of cyanophytes, chlorophytes and chlorophytes

The results of the present study showed that mean densities of cyanophytes, chlorophytes and bacillariophytes were found to be high in T3 which might be due to favourable environmental factors whereas the lower density of these algae found in T4 which might be due to the bloom of euglenophytes and unfavourable environmental conditions. Rahman *et al.* [40] reported that euglenophytes bloom in the fish ponds reduced the density of chlorophytes and bacillariophytes. Hosmani [20] also reported that the blooms of *Euglena elastica, E. gracilis* and *Trachelomonas charkoweinis* have a significant effect in reducing the number of other algal species. These reports are supportive to the present study.

4.5 Growth performance

A variety of factors such as water quality, algal bloom, culture technique, genetic condition etc. controlled the growth rate of fish. In the present study, growth of the fishes in terms of MWG, ADWG and SGR were relatively higher in T1, T2 and T3 as compared to T4. But, the highest growth was recorded in T3 as compared to other treatments which might be due to the application of both duckweed and lime which might provide better water quality as confirmed by the water quality parameters of that treatment (Table 1). Several references indicated that duckweeds improve water quality by reducing nutrients [9, 24] and heavy metals [10, 36] whereas lime improves water quality by raising pH in acidic water [8, 23]. In addition, duckweed enhanced degradation of organic matter through additional oxygen supply in water [30] and lime reduced organic matters in sediment [46] which might be helpful for fish growth. More to the point, application of duckweed and lime reduced the density of euglenophytes by absorbing nutrients and heavy metals, and by increasing pH which might contribute to better growth of fish. Moreover, duckweed might be enhanced the growth through use as fish food. There is strong supporting evidence that duckweed enhanced the growth of fish in polyculture system [47, 52].

On the other hand, lower growth and survival rate of the fish in T4 might be due to the thick bloom of euglenophytes, lower DO and acidic pH. Supporting evidence to this assumption can be drawn from the previous report [40] that euglenophytes form bloom in acidic pH and nutrients rich environment leads to DO depletion which hampered the growth of beneficial algae and fish. Bloom makes a problem through DO deficiency which hampered the growth of fish as water with DO below 5.0 mg/l is to be unproductive [5, 49]. Furthering, acidic pH in the bloom ponds affect fish growth as neutral or almost alkaline waters (pH 7.0 to 8.00) are the most important for fish growth [21].

5. Conclusion

The presents study revealed that the application of duckweed and lime in combination had a more positive bearing to improve water quality and to decrease density of euglenophytes in fish pond by reducing nutrients and heavy metals, and by increasing pH. Thus the present study concluded that the application of duckweed and lime in combination is better for managing euglenophytes bloom as well as for increasing growth of fish. Further long-term studies are necessary to study the ecological consequences of the management measures applied into euglenophytes bloom infested fish ponds.

6. Acknowledgement

Authors are thankful to the Department of Fisheries, University of Rajshahi, Bangladesh and to the Soil Resource Development Institute (SRDI) Rajshahi, Bangladesh for providing the laboratory facilities.

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