Effects of Aquamats™ on the Water Quality and Periphyton Growth in Nile Tilapia (*Oreochromis niloticus*) Fry Ponds

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Abstract

Attempts were made to assess the effects of an artificial substrate named AquaMats™ on periphyton growth and water quality in Nile tilapia fry ponds, located at the Asian Institute of Technology, Thailand. The experiments were conducted in twelve 330 m³ ponds with randomized complete block design using sex reversed all-male Nile tilapia fry between 1.58-1.73 g in size. The stocking density was 30.3 fish m⁻² in all the ponds. There were four treatments with three replicates each: (a) Non-feeding without AquaMats™, (b) Feeding without AquaMats™, (c) Non-feeding with AquaMats™, (d) Feeding with AquaMats™.

Sixty-two periphyton species were identified on AquaMats™, among which 14 species belonged to Cyanophyta (blue green algae); one to Xanthophyta, 17 to Chlorophyta (green algae), 28 to Bacillariophyta (diatoms) and, two to Euglenophyta (euglenoids). Bacillariophyta were the dominant group with the largest number of species, followed by Chlorophyta, Cyanophyta, Euglenophyta and Xanthophyta, towards the end of the experiment. Total fouling solids increased towards the end of the culture period, whilst chlorophyll-*a* concentration on AquaMats™ decreased drastically towards the end of the experiment. At the end of the experimental period it was observed that AquaMats™ were fully covered with thick mud in all experimental ponds, indicating that it attracted the mud particles from the water column. Periphyton growth suppressed due to the deposition of mud particles on AquaMats™ and it appears that AquaMats™ cannot be used effectively in earthen ponds with Nile tilapia fry to enhance periphyton growth. The results revealed that the AquaMats™ had no significant effect on water quality, both in feeding and non-feeding treatments ($P > 0.05$).

Introduction

Periphyton is defined as the total assemblage of sessile or attached organisms on any substrate. Periphyton may contain protozoans, bacteria, fungi, algae, rotifers, annelids, insect larvae and crustaceans (Van Dam et al. 2001). Periphyton has the same functions of oxygen and feed production as phytoplankton, but may be more stable and can be utilized more efficiently by fish. Substrates provide sites for the growth of periphyton and subsequently these periphyton are eaten by fish food organisms and fish. Hard substrates, which are required by periphyton for attachment, are usually absent in fish ponds (Keshavanath et al. 2001). More recently, a number of novel periphyton-based aquaculture systems have emerged (Welcomme & Bartley 1998). Recent trials with various types of substrates for periphyton growth have shown significant increases in fish production. Periphyton growth has been promoted by introducing substrates such as tree branches, bamboo poles, plastic baffles, sugarcane bagasse, paddy straw and dried *Eichhornia* (Shrestha & Knud-Hansen 1994; Ramesh et al. 1999).

AquaMats™ is an artificial substrate, produced by a commercial establishment in the USA. The manufacturer claims that the use of AquaMats™ in tilapia culture may improve the water quality and enhance the periphyton growth. Furthermore, it is claimed to reduce the production cost, improve the immune system, provide shelter, act as a bio-filter, increase dissolved oxygen in water without aeration and increase the stocking density of fish (Anon. 1999). Because of these features it was hypothesized that AquaMats™ may be beneficial for Nile tilapia fry production. The purpose of this study was to identify the periphyton species growing on these artificial substrates and to investigate on the effects of AquaMats™ on the water quality.

Materials and Methods

The experiment was conducted at the Asian Institute of Technology (AIT) Thailand from 16 January 2001 to 21 April 2001. There were four combinations (treatments) with three replicates each: (a) non-feeding without AquaMats™, (b) feeding without AquaMats™, (c) non-feeding with AquaMats™ and (d) feeding with AquaMats™.

Twelve earthen ponds of approximately 330 m² in surface area were used in the experiment. Experimental ponds were grouped into three blocks, and treatments were randomly allocated to the ponds in each block. Sex-reversed all-male Nile tilapia fry, produced using 17 α -methyl testosterone at AIT Tilapia Hatchery averaging 1.58-1.73 g in weight, were stocked at 30.3 fish m⁻² in each pond. AquaMats™ (Meridian Applied Technology Systems Ltd., USA) were 2 m² (1x2m) in

area. Eight pieces of AquaMatsTM were vertically suspended in two rows apart equally in each pond of the treatments with AquaMatsTM. A sand bag was hung on one lower corner of each piece of AquaMatsTM to swing it around the heavy end.

All ponds were fertilized weekly with urea and triple super phosphate (TSP) at rates of 28 kg N and 7 kg P per ha (2.26 kg urea per pond per week and 1.4 kg TSP per pond per week) respectively. The initial fertilization took place one week prior to stocking tilapia fry.

Tilapia in the two treatments with supplemental feeding were fed with commercial pelleted feed (AT feed, 35% crude protein, Cargills Co.Ltd.) at 30%, 6.5%, 6%, 4.4% and 4.2% body weight per day (BW/d) during the first, third, fifth, seventh and ninth fortnight respectively. The feed ration was adjusted biweekly based on fish standing crop, which was estimated using the sample weight, assuming 100% survival rate.

Prior to the start of the experiment, all ponds were drained completely, dried for one week, and then filled with canal water. Water depths in all ponds were maintained at approximately 100 cm by adding water weekly to compensate for the water loss resulted due to evaporation and seepage. Sodium bicarbonate was added weekly to maintain the minimum total alkalinity at 75 mg/l as CaCO₃, based on weekly measurement of total alkalinity in pond water.

Column water samples were taken near the center of each pond at approximately 0900 h weekly for the determination of total alkalinity, and biweekly for the determination of the amount of total ammonia nitrogen (TAN), nitrate-N, nitrite-N, total phosphorus (TP), chlorophyll-a, total suspended solids (TSS) and total volatile solids (TVS). Dissolved oxygen content (DO), temperature and pH were also measured biweekly *in situ* at 25, 50 and 75 cm below the water surface at 0630-0830 h. Secchi disk visibility was measured biweekly at approximately 0900 h before taking water samples for other analyses. Standard Methods for Water and Waste Water Examination (APHA, 1985) were employed for water quality analysis.

Soil samples were taken from each pond before filling the water and just after harvesting fish for the determination of total nitrogen (TN) and total phosphorous (TP) contents using standard methods (Boyd, 1995).

AquaMatsTM were sampled monthly to identify the different kinds of food organisms attached to AquaMatsTM and measure the chlorophyll-a content. Eight stripes, one from each AquaMatsTM piece, were randomly selected and a sample of an area of 4 cm² was taken at 25, 50 and 75 cm depth from each stripe. The samples from the same depth in each pond were pooled into BOD bottles with 200 ml of distilled water and kept for 3 hours for incubation. The incubated bottles were kept in an agitator for 24 hours

to separate the attached organisms from the stripes. After the separation, 5 ml of the suspension were filtered for determining chlorophyll-*a* content and the rest was fixed with Lugol's solution (3 drops per each sample) for periphyton identification according to Bellinger (1997).

The rest of the AquaMatsTM stripes (after taking 4 cm² samples) were squeezed in 1000 ml of distilled water. Fifty ml of the suspension were transferred to a pre-weighed porcelain and dried in the oven (103°C) for 48 hours and the weight of total fouling solids per unit volume of sample was calculated.

Data were analyzed statistically using three-way ANOVA and Least Significant Difference (LSD) test with SPSS (version 9.0) statistical software package.

Results

The mean values $\pm$ SE for the water quality parameters measured are given in Table 1. Dissolved oxygen concentration (DO) decreased steadily to around 1 mg/l over the experimental period. There was no significant difference in mean DO concentrations among all treatments ($P>0.05$, Table 1).

There was no prominent temperature stratification in all experimental ponds. There were no significant differences in mean values of temperature and pH among all treatments ($P>0.05$, Table 1).

Mean concentration of alkalinity was significantly higher in the ponds with feeding than that in the ponds without feeding ($P<0.05$) while there was no significant difference between the treatments with and without AquaMatsTM in both non-feeding and feeding ponds ($P>0.05$, Table 1).

Concentrations of TAN, nitrite-N and nitrate-N fluctuated throughout the experimental period. TAN concentrations were higher in feeding ponds than those in non-feeding ponds, however, there was no significant difference among treatments ($P>0.05$). There was also no significant difference in nitrite-N and nitrate-N concentrations among treatments (Table 1). TP concentrations also fluctuated throughout the experimental period without significant differences among treatments ($P>0.05$, Table 1).

Secchi disk visibility decreased towards the end of the culture period in all treatments without significant differences among treatments ($P>0.05$, Table 1). The concentration of chlorophyll-*a* fluctuated over the entire experimental period. Significantly higher concentration of chlorophyll-*a* was observed in the feeding ponds than that in the non-feeding ponds at the end of the experiment ($P<0.05$).

TSS concentration increased towards the end of the experimental period in all treatments while TVS concentration fluctuated throughout the

entire experimental period. Both TVS and TSS concentrations were higher in feeding treatments than in non-feeding treatments at the end of the experimental period, with a significant difference in final and mean concentrations of TVS among treatments ($P < 0.05$). The low final and mean TVS concentrations were observed in the non-feeding treatments.

Table 1. Overall mean values $\pm$ SE of the water quality parameters measured over the experimental period. DO – Dissolved oxygen content at early morning; T- Temperature; Alk – Alkalinity; TAN – Total ammonia nitrogen; TP – Total Phosphorous; Chl-*a* – Chlorophyll-*a*; SDD – Secchi disc visibility; TSS – Total suspended solids; TVS – Total volatile solids. Values with different superscripts in the same row are significantly different ($p < 0.05$).

Parameters	Non-feeding		Feeding	
	No AquaMats	AquaMats	No AquaMats	AquaMats
DO (mg l ⁻¹)	2.13 $\pm$ 0.45	1.37 $\pm$ 0.28	1.41 $\pm$ 0.39	1.88 $\pm$ 0.74
PH	6.97 $\pm$ 0.31	6.50 $\pm$ 0.24	7.07 $\pm$ 0.06	6.86 $\pm$ 0.18
T (°C)	27.7 $\pm$ 0.2	27.9 $\pm$ 0.13	27.5 $\pm$ 0.07	27.5 $\pm$ 0.22
Alk (mg CaCO ₃ l ⁻¹)	70.15 ^a $\pm$ 17.43	61.47 ^a $\pm$ 7.93	116.23 ^c $\pm$ 7.3	125.25 ^c $\pm$ 11.55
TAN (mg l ⁻¹)	1.69 $\pm$ 0.44	1.46 $\pm$ 0.39	1.76 $\pm$ 0.28	2.03 $\pm$ 0.15
Nitrite-N (mg l ⁻¹)	0.21 $\pm$ 0.11	0.21 $\pm$ 0.08	0.17 $\pm$ 0.05	0.23 $\pm$ 0.16
Nitrate-N (mg l ⁻¹)	1.24 $\pm$ 0.08	1.31 $\pm$ 0.11	1.13 $\pm$ 0.17	1.05 $\pm$ 0.03
TP (mg l ⁻¹)	0.49 $\pm$ 0.12	0.64 $\pm$ 0.29	0.96 $\pm$ 0.26	0.53 $\pm$ 0.04
Chl- <i>a</i> (mg m ⁻³)	26 $\pm$ 9.18	27 $\pm$ 7.62	26 $\pm$ 8.86	19 $\pm$ 6.34
SDD (cm)	39 $\pm$ 11.7	32 $\pm$ 6.2	27 $\pm$ 4.7	38 $\pm$ 1.8
TSS (mg l ⁻¹)	46 $\pm$ 11.64	46 $\pm$ 10.46	65 $\pm$ 10.22	47 $\pm$ 8.73
TVS (mg l ⁻¹)	30 ^a $\pm$ 3.73	30 ^a $\pm$ 3.25	38 ^b $\pm$ 4.09	36 ^b $\pm$ 5.21

Sixty-two periphyton species were identified on AquaMatsTM, among which were 14 species of Cyanophyta (blue green algae) one species of Xanthophyta, 17 species of Chlorophyta (green algae), 28 species of Bacillariophyta (diatoms), and two species of Euglenophyta (euglenoids) (Table 2). Bacillariophyta was the dominated group with the largest number of species followed by Chlorophyta, Cyanophyta, Euglenophyta and

Xanthophyta (Table 2). Some mollusc species were also found attached to AquaMats™.

Table 2. Periphyton species recorded on the AquaMats™ that were installed in Nile tilapia fry ponds.

Cyanophyta

Anabaena spiroides
Coelosphaerium sp.
Lyngbya versicolor
Lyngbya coerulea
Microcystis aeruginosa
Microcystis flosaquae
Naegelianum sp.
Nostoc piscinale
Oscillatoria limnetica
Oscillatoria limosa
Oscillatoria rubescens
Oscillatoria tenuis
Stigonema sp.
Tolypothrix sp.

Xanthophyta

Tribonema affine

Chlorophyta

Carteria sp.
Chlorogonium sp.
Cladophora sp.
Closterium aciculare
Hormidium sp.
Mougeotia sp.
Oocystis natans
Scenedesmus armatus
Scenedesmus quadricauda
Sphaerocystis sp.
Spirogyra inflata
Spirogyra weberi
Spirogyra condensate
Spondylosium planum
Stigeoclonium sp.
Ulothrix aequalis
Ulothrix zonata

Bacillariophyta

Achnanthes lanceolata
Achnanthes rutilans
Amphipleura pellucida
Amphora ovalis
Anomoeoneis exilis
Anomoeoneis sphaerophora
Asterionella Formosa
Caloneis amphisbaena
Cyclotella glomerata
Cyclotella operaculata
Diatoma elongatum
Diatoma vulgare
Epithemia turgida
Fragilaria capucina
Fragilaria virescens
Frustulia rhomboids
Gyrosigma spencerii
Navicula dubium
Navicula lanceolata
Navicula phyllepta
Navicula subtilissima
Neidium affine
Neidium placenta
Pleurosigma aestuarii
Rhizosolenia eriensis
Sarirella biserita
Tabellaria fenestrata
Tabellaria flocculosa

Euglenophyta

Euglena pisciformis
Euglena viridis

The estimated periphyton standing crop expressed as chlorophyll-*a* concentration increased linearly in the first two months in both treatments with AquaMats™, but during the second month, in the non-feeding treatment with AquaMats™, the rate of increase was lower than that of feeding treatment. The Chlorophyll-*a* concentration decreased drastically in both treatments in the third month towards the end of the experiment. The chlorophyll-*a* concentrations were higher in the feeding ponds than those in the non-feeding ponds in the second half of the experimental period, but varied largely within treatments, and there was no significant difference between the treatments (Table 3).

Total fouling solids increased linearly in the first two months in both AquaMats™ treatments, i.e., with and without feeding. It continued to increase but at a slower rate in the non-feeding treatment and then decreased in the treatment with feeding during the last month. There was no significant difference in total fouling solid concentrations between AquaMats™ treatments with and without feeding over the 90-day culture period (Table 3).

At the end of the experiment, significant amounts of total nitrogen and total phosphorus were accumulated in the pond sediments in all treatments ($P < 0.05$, Table 4). In both non-feeding and feeding ponds, total nitrogen and total phosphorus contents accumulated in pond sediment during the experimental period tended to be lower in the AquaMats™ treatments, however, no significant differences were observed ($P > 0.05$, Table 4).

Discussion

At the end of the experimental period, AquaMats™ in all ponds were fully covered with mud particles. This might be due to the attraction of clay particles by AquaMats™. This has probably occurred due to resuspension of bottom mud particles as a result of biweekly sampling of fish by seining and also due to grazing and movements of fish. The activity of AquaMats™ may have retarded as a result of clogging of the surface area by mud particles. AquaMats™ would be effectively used for shrimp culture as they do not graze on the habitat. Further, these could be effectively used in tanks and ponds with sandy bottoms.

Water quality measurements made during the experiment indicated that there were no statistically significant differences between treatments with and without AquaMats™. Therefore, it could be concluded that AquaMats™ were not effectively activated and did not improve the water quality in earthen ponds. Moreover, there are locally available substrates that might be used for this type of aquaculture (Keshavanath et al. 2001).

Table 3. Mean $\pm$ SE values for chlorophyll-*a* and total fouling solids over the experimental period in AquaMats™ treatment. Values with the same superscripts in the same row are not significantly different ($P>0.05$).

Parameters		Depth (cm)	AquaMats™ treatments	
			Non-feeding	Feeding
Chlorophyll- <i>a</i> ($\mu\text{g cm}^{-2}$)	Initial		0	0
	Day 30	25	0.478 ^a $\pm$ 0.147	0.223 ^a $\pm$ 0.084
		50	0.392 ^a $\pm$ 0.1	0.343 ^a $\pm$ 0.022
		75	0.518 ^a $\pm$ 0.227	0.765 ^a $\pm$ 0.255
	Day 60	25	0.360 ^a $\pm$ 0.153	0.885 ^a $\pm$ 0.617
		50	0.294 ^a $\pm$ 0.162	0.819 ^a $\pm$ 0.583
		75	0.943 ^a $\pm$ 0.689	1.253 ^a $\pm$ 0.257
	Day 90	25	0.053 ^a $\pm$ 0.016	0.393 ^a $\pm$ 0.192
		50	0.051 ^a $\pm$ 0.008	0.312 ^a $\pm$ 0.126
		75	0.081 ^a $\pm$ 0.049	0.252 ^a $\pm$ 0.121
Fouling solids ($\mu\text{g cm}^{-2}$)	Initial		0	0
	Day 30		0.788 ^a $\pm$ 0.16	0.760 ^a $\pm$ 0.06
	Day 60		2.111 ^a $\pm$ 0.92	2.407 ^a $\pm$ 0.54
	Day 90		2.562 ^a $\pm$ 0.73	2.238 ^a $\pm$ 0.44

Table 4. Accumulation of nutrients in pond sediments in different treatments during the experimental period. Values with the same superscripts in the same row are not significantly different ($P>0.05$).

	Non feeding		Feeding	
	No AquaMats™	AquaMats™	No AquaMats™	AquaMats™
Total N				
At the beginning	2.1 ^p $\pm$ 0.2	2.0 ^p $\pm$ 0.2	1.9 ^p $\pm$ 0.3	2.1 ^p $\pm$ 0.3
At the end	2.4 ^q $\pm$ 0.4	2.2 ^q $\pm$ 0.2	2.4 ^q $\pm$ 0.08	2.4 ^q $\pm$ 0.2
Accumulation	0.3 ^r $\pm$ 0.16	0.2 ^r $\pm$ 0.1	0.5 ^r $\pm$ 0.1	0.3 ^r $\pm$ 0.0
Total P				
At the beginning	0.472 ^a $\pm$ 0.04	0.418 ^a $\pm$ 0.03	0.486 ^a $\pm$ 0.04	0.467 ^a $\pm$ 0.04
At the end	0.537 ^b $\pm$ 0.04	0.47 ^b $\pm$ 0.02	0.573 ^b $\pm$ 0.03	0.548 ^b $\pm$ 0.03
Accumulation	0.065 ^r $\pm$ 0.03	0.052 ^r $\pm$ 0.018	0.087 ^r $\pm$ 0.009	0.081 ^r $\pm$ 0.009

Chlorophyll-*a* concentrations in periphyton tend to decrease, while the total fouling solids tend to increase towards the end of the culture period. The possible reason for these would be the clogging of the surface of AquaMats™ by mud particles due to resuspension of bottom sediments.

Among the periphyton species identified, diatoms were the dominated group on AquaMats™. Huchette et al. (2000) identified about 32 species of diatoms as periphyton along with other microorganisms, animals and plants growing on artificial substrates in tilapia cages.

The amounts of total nitrogen and total phosphorus accumulated in pond sediments increased towards the end of the experiment. This probably has resulted due to addition of urea and TSP weekly to all ponds. Feeding treatments had higher values than non-feeding treatments probably due to the accumulation of uneaten feeds in the bottom soil. The high nutrient contents in the treatments without AquaMats™ than in those with AquaMats™ may be due to the adsorption of the nutrients by AquaMats™ from the bottom sediment.

From the results of the present experiment, it can be concluded that AquaMats™ did not improve the water quality in earthen ponds stocked with Nile tilapia fry. AquaMats™ in earthen ponds did not facilitate a significantly higher periphyton growth as it attracted and adsorbed non-living particles specially clay particles on to the surface which resulted in a low periphyton growth.

Similar studies have to be carried out in earthen ponds without sampling fish by seining to avoid disturbing of pond bottom, which may reduce the amount of clay particles attached on AquaMats™. Further studies are also needed to compare effects of AquaMats™ on water quality between ponds with different substrates. Comparative studies should also be undertaken between AquaMats™ and the other natural substrates such as sugarcane baggase, bamboo and tree branches to develop effective periphyton based culture systems.

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