DEVELOPING NEW SYSTEMS FOR PERIPHYTON ENHANCEMENT IN FARMERS' PONDS

Production System Design and Best Management Alternatives/Study/16BMA04UM

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ABSTRACT

A field trial was carried out to test four locally available substrates for periphyton enhancement in carp-SIS ponds at Majhui in Chitwan and Seri and Nandapur in Nawalparasi district for seven months. Tested substrates were suggested by farmers at workshops done in each district. Altogether 30 farmers, 15 from Sundardeep women's cooperative in Chitwan and 15 from Mishrit cooperative in Nawalparasi participated in the trial. Five treatments: i) control without substrate, ii) split bamboo, iii) whole bamboo, iv) banana midrib, and v) plastic bottle were introduced to farmers in each district. Farmers were divided into five groups in each district with each group having three farmers adopt one treatment. All farmers stocked 6 carp species at 15,000 fish per hectare and SIS at unrecorded densities. Farmers fed fish with rice bran and mustard oil cake at 1.5% BW/d while grass carp was fed with grass and banana leaves at 50% BW/d. Flood hit both districts in August 2017 and affected the trial. The effect of flooding was greater in Chitwan and pond data varied dramatically. Carp survival and production varied from 13 to 40% and 0.96 to 2.83 tha⁻¹·vr⁻¹, respectively. In Nawalparasi, damage from flooding was less and data were used to determine treatment effects. Combined NFY was 19% higher in ponds using plastic bottles than control ponds, while NFY of SIS was 50% higher in ponds using banana midribs than control, strip bamboo, and plastic bottle treatments. Feed conversion ratio was less than 1.9 in all substrate ponds, indicating substrates have potential for reducing feed cost. FCR was significantly better (P<0.05) in split bamboo ponds than control ponds. Banana midrib decayed fast and required 3-4 replacements during a grow-out while treatments with plastic bottles performed better in terms of production and profit. Water quality was monitored monthly while periphyton abundance and biomass was analyzed three times, in the beginning, middle, and end of trial. Water quality was within an acceptable range for carp. Periphyton abundance and biomass did not differ significantly among substrates. Non-adopting farmers (16 males and 26 females) of Chitwan and Nawalparasi were trained on carp-SIS polyculture in periphyton enhanced systems through a one day workshop.

INTRODUCTION

The government of Nepal has recognized that chronic malnutrition is a major problem in the country. About 41% of children younger than 5 years of age are stunted (UNICEF, 2012) and 48% are anemic (MoHP, 2006). With the nutrition problem, there is a need to develop environmentally sustainable and cost-effective food production systems that function year round to provide adequate nutrients and improve household income for poor rural farmers.

Our research activities in Nepal have targeted local women for improvements in household and larger-scale fish pond production. In Nepal, men from poorer rural areas are often forced to seek employment outside the home (often even outside the country), and women are left to maintain the household and care for the family (Bhujel, 2009). As a result, most ponds developed for household aquaculture are managed by women. Carp-SIS polyculture and carp-SIS polyculture with bamboo substrates managed by Sundardeep women's cooperative have been examples.

In Phase I, we tried to enhance pond production by providing bamboo substrates for colonization of periphyton. As periphyton removes nutrients from the water and adds oxygen as it grows, it also cleans water being discharged from ponds and improves environmental performance. Since rohu, catla, and common carp are periphyton feeders (Rai and Yi, 2012), their growth and production are enhanced in ponds with added substrate for periphyton colonization compared to ponds without substrate (Azim et al., 2002; Rai et al., 2008). We recently completed a series of trials in on-station and on-farm experiments (Rai et al., 2016). These experiments showed dramatic increases in net fish yield (27%) and profit (74%) by adding substrates and reducing feeding rates in on-station experiments. For on-farm studies, total fish production and gross margin were 19.3% and 151% higher in the carp+SIS+substrate treatments with 50% feeding than in carp polyculture with 100% feeding. Reduced feeding that is possible when periphyton is enhanced is not only economically more viable but also enhances environmental performance, as the water quality in ponds is generally higher and effluent released on draining for harvest is not as damaging. However, the on-farm work also identified some problems with our periphyton system. We used fixed rafts of bamboo covering about 1% of the pond area as a substrate for periphyton growth, but culturists believe these structures interfered with harvesting of fish, although on the positive side, they may also have provided hiding places for fish to avoid predation by birds, since survival of some carp species was higher in substrate ponds. Further outreach on this system, including testing of alternative periphyton enhancing substrates that minimize disturbance to their operations, is the main objective of this investigation. Some possible methods might include using portable and floating substrates or ones that could be lifted from the water or pond during management activities. The economic value of periphyton enhancement includes the ability to grow fish faster under similar inputs, as well as the ability to reduce inputs of feed and achieve similar growth rates. However, our previous trials included both periphyton enhancement and feed input reduction together. We have not tested reduced feeding without periphyton enhancement, and thus the gain in profit by reduced feeding has been included in the benefit of periphyton enhancement in our studies to date. We need to also separate these two management activities so we can clearly understand the importance of reduced feeding compared to periphyton enhancement in polyculture systems.

OBJECTIVES

Since previous work using bamboo mats resulted in modifications to ponds that gave better production and economic returns but interfered with pond management, we decided to investigate alternative substrate systems. The purpose of this study is to field test alternative periphyton substrates suggested by farmers in a workshop. The alternative systems were tested on-farm using carps and SIS ponds in two locations of Nepal.

MATERIALS AND METHODS

Periphyton Workshop

Two workshops were held one in Sauraha, Chitwan and one in Kawasoti, Nawalparasi district. The objective of the workshop was to identify possible alternatives for periphyton substrates based on farmers' recommendations. In Chitwan, 32 people participated in the workshop, among them, 16 were farmers (3 male, 13 female), 1 was an executive member of Chitwan Fisheries Entrepreneurial Association, 2 were heads of NGOs, 4 were faculty of Agriculture and Forestry University (AFU), 8 were students (3 male, 5 female), and 1 was an intern from the USA (female). Similarly, 27 people participated the workshop in Nawalparasi, among them, 15 were farmers (13 male and 2 female), 2 were executive members of District Fisheries Entrepreneurial Association, Nawalparasi, 2 were heads of NGOs, 1 was a Fisheries Development Officer, 2 were faculty of AFU, 4 were students of AFU, and 1 was an intern from the USA (female). The participant list is given below in Table 1.

Table 1. Participants in the inception workshop from each district

Participants	Chitwan Nawalpa			Nawalparasi	ırasi	
	Male	Female	Total	Male	Female	Total
Farmer	3	13	16	13	2	15
Fisheries Entrepreneurial						
Association	1		1	2		2
NGO	2		2	2		2
Fisheries Development Office						1
Faculty	4		4			2
Student	3	5	8	2		4
Intern		1	1		1	1
Total	13	19	32	19	3	27

At the workshops, presentations on importance of periphyton substrates to fish, different types of substrates and their uses were made to give knowledge of periphyton enhanced fish production system to non-adopting participants. Farmers that have been involved in the previous periphyton studies along with other participants from the district were divided into 5 discussion groups and asked to propose better alternatives than bamboo mats for periphyton substrates in ponds. Prior to discussion, participants were instructed to consider criteria for alternatives that must be i) environmentally responsible, ii) locally available, and iii) cost effective. Participants suggested the following substrates after group discussion.

Table 2. Periphyton substrates recommended by participants

	Types of substrate	Frequency
1	Split bamboo mat	9
2	Whole bamboo	8
3	Bamboo mat along slope of the dike	1
4	Plastic bottle with sand inside	3
5	Jute sac	1
6	Coconut tree leaf	1
7	Banana leaf midrib	1
8	Boulders	1
9	Galvanized insulated net	1

Based on their recommendations and criteria provided, 4 alternatives including split bamboo, whole bamboo, banana midrib, and plastic bottles were selected for field testing.

On-Farm Trial

An on-farm trial was conducted for 230 days in Majhui of Chitwan district and for 210 days in Seri and Nandapur of Nawalparasi district. Fifteen women farmers from Sundardeep women's fish farmers cooperative in Majhui and same number of farmers (12 male and 3 female) from Mishrit cooperative in Seri and Nandapur participated in the trial. Participant selection was done by respective cooperatives. Lists of farmers in Chitwan and Nawalparasi districts are given below in the Tables 3 and 4. Farmers were divided into five groups including 3 farmers in each group based on substrate types they tested i) control without substrate, ii) split bamboo, iii) whole bamboo, iv) banana midrib and v) plastic bottle.

Table 3. List of farmers used in testing various substrate treatments in Chitwan district

Number of Participants	Gender	Cooperative's name	Substrate type
3	Genuei		Substrate type
3	- 1	Sundardeep women	27 1
	Female	cooperative	No substrate
3		Sundardeep women	
	Female	cooperative	Split bamboo
3		Sundardeep women	
	Female	cooperative	Whole bamboo
3		Sundardeep women	
J	Female	cooperative	Banana midrib
3	1 Ciliaic	Sundardeep women	Danana imario
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	Female	cooperative	Plastic bottle

Table 4. List of farmers used in testing various substrate treatments in Nawalparasi district

Number of Participants	Gender	Cooperative's name	Substrate type
2	Male	Mishrit cooperative	No substrate
1	Female	Mishrit cooperative	No substrate
3	Male	Mishrit cooperative	Split bamboo
3	Male	Mishrit cooperative	Whole bamboo
2	Male	Mishrit cooperative	Banana midrib
1	Female	Mishrit cooperative	Plastic bottle
1	Female	Mishrit cooperative	Banana midrib
2	Male	Mishrit cooperative	Plastic bottle

Farmers prepared ponds by liming at 500 kg/ha. After that they fertilized ponds using urea and DAP at 470 g/100 m² and 350 g/100 m² to stimulate growth of phytoplankton. Prior to stocking, farmers fixed substrates assigned to them in ponds covering 2% of pond surface area. A split bamboo mat was formed by placing bamboo splits one after another over the vertical bamboo splits. The mat was suspended in the water column by using empty plastic bottles (coke, mineral water bottles) tied to upper part of the mat to float the mat while water filled plastic bottles were tied to the bottom part of the mat to give weight to the mat. The number of bamboo splits used in each mat and number of mats per pond was determined by area of bamboo splits. The area of bamboo splits was estimated by measuring length and width of 5 splits and taking their average. On average, 8-10 pieces of bamboo splits were used per mat. Number of bamboo mats per pond varied from 2-19 depending on pond size. Whole bamboo containing side branches were directly kept in the pond without using floats and sinkers. Due to side branches, the bamboo remained suspended in the water column. Area of whole bamboo was estimated by measuring circumference and length of bamboo pieces while area of side branches was not estimated. Number of whole bamboo pieces used per pond varied from 2-19 depending on pond size. Banana midribs were used as rafts due to the buoyancy caused by air space inside the midrib. Area of each midrib, number of midribs in each mat and number of mats per pond was estimated in the same way as bamboo splits. Number of banana midrib rafts per pond varied from 2-17 depending on pond size. For plastic bottle substrates, water filled plastic bottles were tied to a bamboo strip ring using string. Empty plastic bottles were used as floats which were tied at the upper part of each ring. Area estimation process was similar to whole bamboo. Number of bottles per pond varied from 10 to 30 per ring depending on size of bottle because some farmers used mineral water bottles which are relatively small in size while others used large sized Coke and Fanta bottles. Number of rings containing bottles per pond varied from 2 to 6.

Farmers stocked silver carp (Hypophthalmichthys molitrix, 0.7±0.1 g), bighead carp (Aristichthys nobilis, 25.6±4.0 g), grass carp (Ctenopharyngodon idella, 4.5±0.4 g), common carp (Cyprinus carpio, 0.1±0.0 g), rohu (Labeo rohita, 4.3±0.3 g), mrigal (Cirrhinus mrigala, 4.5±0.3 g) at the rate of 15,000/ha. Stocking was started on 10 April and completed on 12 April 2017 in both districts. They fed dough (overall crude protein=24%, crude fat=6%, crude fiber=9%) of rice bran and mustard oil cake mixed at 1:1 to carp at 1.5% of carp biomass per day as defined in the protocol (full feeding was 3% of carp biomass). Farmers also fed grass carp with banana leaves and grass at 50% of body weight per day. Monthly fish growth samplings were done to adjust the feed ration. Farmers in Chitwan stocked SIS (Dedhuwa Esomus danricus and Pothi Puntius sophore) by allowing canal water to enter the pond after the monsoon started whereas farmers in Nawalparasi collected SIS from nearby canals for stocking. Final harvest was started on 10 November and completed on 15 November 2017 in Nawalparasi and started on 30 November and completed on 3 December 2017 in Chitwan. Final harvesting was done by draining the pond as much as possible using pumps. Fish were counted and weighed in batches by species to get the final harvest weight and number. After taking weight, fish were released back to a pond for future use. Tharu people consume and sell many fish during "Maghi," their biggest festival which falls on January 15. A record book was given to each farmer to record the number and weight of fish that were consumed, sold, or died. Fish production includes count and weight of those consumed, sold or died along with final harvest. Farmers did not sell SIS because they consumed all at home.

Gross margin analysis

Gross income from fish sales was calculated from total production, assuming all carp and SIS were sold. Selling price of carp was NRs 270/kg (105 NRs = \$1 US) in Chitwan and NRs 300/kg in Nawalparasi, and that of SIS was estimated NRs. 200 in both districts. Variable costs of carp seed was estimated NRs 3.25/piece, lime NRs 18/kg, urea NRs 20/kg, DAP NRs 50/kg and feed NRs 32.5/kg. SIS was procured free of cost by farm labor. Gross return was calculated assuming all products sold at farm gate prices.

Water quality monitoring and periphyton analysis

Temperature, DO, pH and Sechhi disk visibility of ponds were monitored in situ monthly. Periphyton samples from four different types of substrate were taken from the field and analyzed three times, in the beginning, middle, and end of the trial at the laboratory of the Fisheries Program in AFU. Periphyton genera were identified and their abundance estimated for each substrate. Periphyton attached to a substrate was scraped with a scalpel from a random 1 cm² area of the substrate for periphyton biomass analysis. Dry matter, ash free dry matter, and ash content were determined following APHA (1980).

A one-way analysis of variance (ANOVA) was used to compare periphyton abundance and biomass (dry matter, ash, and ash-free dry matter) among treatments. ANOVA was also used to test differences in fish production and gross return among treatments. Alpha was set at 0.05 for all comparisons. All means are reported with \pm 1 standard error.

RESULTS

Water quality was within an acceptable range in all treatment ponds indicating substrate did not affect water quality adversely. Water quality varied slightly between Chitwan and Nawalparasi districts and between treatments within the district. Mean temperature was slightly lower in all treatments in Chitwan than Nawalparasi. Similarly, mean transparency was lower in all treatments in Nawalparasi than Chitwan.

Table 5. Water quality in ponds in each treatment in Chitwan and Nawalparasi districts

	Control	Split Bamboo	Whole Bamboo	Banana Stem	Bottle	
	Chitwan					
Temperature	28.9±0.5	28.8±0.4	28.5±0.6	28.3±0.4	28.8 ± 0.6	
Transparency	32±6	31±5	29±4	32±4	32±6	
DO	4.5±0.8	4.3±0.7	4.1 ± 0.4	4.1 ± 0.8	4.0±1.0	
рН	8.5±0.2	8.4±0.2	8.3±0.2	8.3±0.3	8.1±0.3	
	Nawalparasi					
Temperature	29.3±0.3	29.8±0.7	30.0 ± 0.8	30.1±0.5	30.1±0.5	
Transparency	24±3	25±4	25±3	22±3	23±3	
DO	3.9 ± 0.7	4.6 ± 0.8	4.3±0.8	4.5±0.5	4.3±0.6	
рН	8.0±0.2	8.2±0.2	8.0±0.2	7.3±0.2	8.1±0.2	

There were no significant differences in periphyton abundance and biomass among substrate treatments. Periphyton biomass was determined in terms of dry matter, ash and ash-free dry matter.

Table 6. Abundance (no./cm²) of periphyton genera in each substrate treatment in Chitwan

Group	Treatment			
	Whole Bamboo	Bottle	Split Bamboo	Banana Stem
Coscinodiscus	4306 ± 1002^{b}	3056 ± 278^{ab}	2083±241b	2083±241 ^b
Cyclotella	9306±773a	3889±501 ^b	5556±1565ab	5833±1339ab
Diatoma	7778±501a	3889±2003a	3750±417a	5833±1502a
Fragillaria	556±278a	0±0a	0 ± 0^a	417±417 ^a
Gomphonema	0 ± 0^a	0±0a	0 ± 0^a	0±0a
Melosira	556±556 ^a	417±417a	0 ± 0^{a}	0±0°a
Navicula	7778 ± 1528^{a}	4722 ± 1806^a	2778 ± 1137^{a}	5694±1389a
Nitzschia	2778 ± 845^{ab}	972±501 ^b	8333±3127 ^b	1111±1111 ^b
Surirella	278±278a	972±972a	972±972a	417±417 ^a
Synedra	6250±1049a	4583±2441a	4028 ± 1187^a	5417±241a
Bacillariophyceae	39583±4455a	22500±7051a	27500±3146a	26806±4654a
Actinastrum	$0\pm0^{\mathrm{a}}$	972±972a	1528±1528 ^a	3056±1547a
Ankistrodesmus	11944±1602ª	5417±2295b	8194±972ab	8472±1806ab
Chlamydomonas	1250±636a	1111±1111 ^a	0 ± 0^a	0 ± 0^{a}
Characium	$0\pm0^{\mathrm{b}}$	$0 \pm 0^{\rm b}$	4444±139a	1667±1667 ^b
Chlorella	12500±5838a	4167±1049a	7361±1002a	11250±2320a
Chodatella	$0\pm0^{\mathrm{a}}$	0±0a	0 ± 0^a	0±0 ^a
Closterium	1667±1667a	0±0a	0 ± 0^{a}	2778±1690a
Coelastrum	0±0a	1667±1667ª	556±556a	0±0 ^a
Cosmarium	3472±1837 ^a	833±636 ^a	139±139 ^a	556±556ª
Crucigenia	4306±3319 ^a	3194±2235a	4167±1049a	2500±2097ª
Gonatozygon	2361±1773 ^a	1111±1111 ^a	833±833ª	0±0a
Mougeotia	0±0a	0±0ª	0 ± 0^{a}	0±0 ^a

Group		Treatment				
	Whole Bamboo	Bottle	Split Bamboo	Banana Stem		
Oocystis	0±0ª	0±0a	0±0ª	0 ± 0^{a}		
Pediastrum	10000 ± 1909^a	4306±2183a	8333 ± 1925^a	9583 ± 636^{a}		
Scenedesmus	4167 ± 3368^a	2222 ± 1325^a	6806 ± 1325^a	7500 ± 2097^{a}		
Selenastrum	556±556a	0 ± 0^a	$694{\pm}694^a$	694±367a		
Sphaerocystis	417±417a	0 ± 0^{a}	0±0a	0 ± 0^{a}		
Staurastrum	5833±1049a	2917±1463 ^a	4722±1137a	4167±1463 ^a		
Tetreedron	556±556a	0 ± 0^{a}	417±241a	833±833 ^a		
Tetraspora	0 ± 0^a	0 ± 0^a	1250±722a	417±417 ^a		
Treubaria	556±556a	972±501a	0±0a	139±139a		
Ulothrix	0±0a	0 ± 0^{a}	0±0ª	0 ± 0^{a}		
Volvox	0±0a	0±0a	0±0ª	0±0a		
Chlorophyceae	59583±13283a	28889±12108a	49444±5195°	53611±6729a		
Anabaena	5972±2639a	3333±1925a	6806±1187a	4167±722a		
Aphanocapsa	0±0a	0±0a	0±0ª	0±0a		
Chroococcus	2639±1565a	2083±1339a	1528±139a	1389±735a		
Gloeocapsa	2500±2500a	0±0a	0±0a	0±0a		
Gomphosphaeria	0 ± 0^a	0±0ª	0±0a	0±0a		
Merismopedia	12500±1273a	5972±2850ab	4306±367 ^b	6806±2434ab		
Microcystis	0±0a	2917±833a	694±694ª	5972±4592°		
Oscillatoria	2222±1470a	2778±1137 ^a	5278±2373a	4306±972a		
Cyanophyceae	25833±7561a	17083±4416 ^a	18611±972a	22639±5540a		
Euglena	7222±1944ª	6250±2097a	8472±1211a	7639±2046a		
Phacus	5000±1684a	3472±1740 ^a	4583±417a	4444±1111°		
Trachalomonas	5417±481ab	9306±2572a	2917±1102 ^b	3056±1806 ^b		
Euglenophyceae	17639±694a	19028±5879a	15972±1470a	15139±3745ª		
Batrachosperium	0±0a	0±0ª	0±0a	0±0a		
Lemanea	0±0a	0±0ª	0±0a	0±0a		
Rhodophyceae	0±0a	0±0ª	0±0a	0±0a		
Oedogonium	10278±6144a	3056±3056a	18889±5725a	9306±5413a		
Nodularia	278±278a	556±556a	0±0ª	278±278a		
Tribonema	0±0a	0±0ª	1667±1667ª	4722±2767a		
Stigeoclonium	8750±4732a	2222±2222ª	1667±1667 ^a	3056 ± 3056^{a}		
Other	19306±9820a	5833±5023 ^a	22222±7097a	17361±5592 ^a		
Total Phytoplankton	161944±14446 ^a	93333±33208 ^a	133750±7504 ^a	135556±17504°		
Difflugia	6389±911 ^a	4722±367 ^a	10139±2900 ^a	6389±911 ^a		
Sarcodina	6389±911 ^a	4722±367 ^a	10139±2900 ^a	6389±911 ^a		
Asplanchna	5139±1450 ^a	3333±241 ^{ab}	1944±735 ^b	3889±733 ^{ab}		
Brachionus	8194±1690 ^a	3472±2183°	3472±845 ^a	5278±845°		
Filinia	0 ± 0^{a}	0 ± 0^{a}	0 ± 0^{a}	0 ± 0^{a}		
	0=0	0-0	0-0	0-0		

Group	Treatment			
	Whole Bamboo	Bottle	Split Bamboo	Banana Stem
Keratella	1528±845a	833±833 ^a	694±367a	1250±722a
Lecane	694±694 ^{ab}	1528±139a	$0\pm0^{\mathrm{b}}$	0±0 ^b
Polyarthra	0 ± 0^{a}	0±0a	0 ± 0^a	0 ± 0^a
Trichocerca	0 ± 0^{a}	0±0a	0 ± 0^a	0 ± 0^a
Rotifera	15556±2373a	9167±3182ab	6111±1602 ^b	10417±636ab
Cyclops	1806±605a	972±139a	694±367a	417±417a
Dioptomus	$0\pm0^{\mathrm{a}}$	0±0ª	0 ± 0^a	278±278a
Daphnia	278±278ab	1250±636a	0 ± 0^{b}	$0 \pm 0^{\rm b}$
Diaphanosoma	$0\pm0^{\mathrm{a}}$	0±0a	0 ± 0^{a}	0 ± 0^{a}
Moina	$0\pm0^{\mathrm{a}}$	0±0a	278±278ª	0 ± 0^a
Nauplius	2639±1002a	1528±367a	694±694ª	972±501a
Crustacea	4722±1410 ^a	3750±636a	1667±833ª	1667±833ª
Total Zooplankton	26667±3368a	17639±4183a	17917±4507ª	18472±1602a

Table 7. Abundance (no./cm²) of periphyton in each substrate treatment in Nawalparasi

	Treatment			
Group	Whole Bamboo	Bottle	Split Bamboo	Banana Stem
Coscinodiscus	3056 ± 1547^{a}	1806 ± 1410^a	3333 ± 1735^a	2500±241a
Cyclotella	5833 ± 636^{a}	4722 ± 1959^a	6250 ± 867^a	8472±2661a
Diatoma	7778 ± 3456^{a}	6389 ± 2504^a	6389±2074	7361 ± 2074^{a}
Fragillaria	$0\pm0^{\mathrm{b}}$	$278{\pm}278^{ab}$	694 ± 367^{a}	0 ± 0^{b}
Gomphonema	0±0a	0±0a	0±0a	0 ± 0^a
Melosira	0 ± 0^a	0 ± 0^{a}	0 ± 0^a	0 ± 0^a
Navicula	6806 ± 3852^a	5694±2410 ^a	10556 ± 4938^a	6111 ± 2434^{a}
Nitzschia	2778 ± 2778^{a}	4167±2295a	2500 ± 1339^a	$2778 {\pm} 2778^a$
Surirella	$0\pm0^{\mathrm{a}}$	0 ± 0^a	0 ± 0^a	417 ± 417^{a}
Synedra	3611 ± 1602^a	3889 ± 1325^a	4538 ± 1502^a	3333 ± 1667^{a}
Bacillariophyceae	29861 ± 6974^{a}	26944 ± 5198^a	34306 ± 12523^a	30972 ± 7500^a
Actinastrum	2500 ± 2500^a	0 ± 0^a	1111 ± 11111^{a}	$278{\pm}278^a$
Ankistrodesmus	6667 ± 1049^a	6944 ± 1637^a	8889 ± 1234^a	8889 ± 773^{a}
Chlamydomonas	694 ± 694^{a}	2222±2222a	2361 ± 2361^{a}	$278{\pm}278^a$
Characium	556 ± 556^{a}	$278{\pm}278^a$	0 ± 0^a	556±556a
Chlorella	9583 ± 4829^a	11111 ± 3546^{a}	5972 ± 2989^a	10556 ± 3729^a
Chodatella	0 ± 0^a	0 ± 0^a	0 ± 0^a	0 ± 0^a
Closterium	0 ± 0^a	0±0a	972±605a	0 ± 0^a
Coelastrum	$0\pm0^{\mathrm{a}}$	0 ± 0^a	0 ± 0^a	0 ± 0^a
Cosmarium	1528±972a	2361±2361a	1806 ± 1085^a	556 ± 556^{a}
Crucigenia	3750 ± 1667^a	3889 ± 2286^a	5278 ± 735^{a}	5694 ± 3046^a
Gonatozygon	0 ± 0^a	2500 ± 2500^a	2639 ± 1869^a	1528 ± 1528^a
Mougeotia	1250±1250a	3056 ± 1547^{a}	1806 ± 1085^a	833±833 ^a
Oocystis	0 ± 0^a	417±417a	0 ± 0^a	0 ± 0^a

	Treatment				
Group	Whole Bamboo	Bottle	Split Bamboo	Banana Stem	
Pediastrum	4861 ± 2572^{a}	6667 ± 1684^a	6528 ± 2504^a	10139±5576a	
Scenedesmus	1667±962a	1667 ± 1667^a	2222 ± 1137^a	3750 ± 833^{a}	
Selenastrum	$694{\pm}694^a$	694 ± 694^{a}	417±417a	0±0a	
Sphaerocystis	0 ± 0^a	0 ± 0^a	0±0a	0±0a	
Staurastrum	694 ± 694^{a}	278±278a	556±556a	833±481a	
Tetreedron	1250±1250a	0 ± 0^a	833±833a	3056 ± 3056^a	
Tetraspora	0 ± 0^a	694±694ª	0±0a	1389±1389a	
Treubaria	0 ± 0^a	0 ± 0^a	0±0a	0±0a	
Ulothrix	0 ± 0^a	1111±1111 ^a	972±972a	0±0a	
Volvox	0 ± 0^a	1111±1111a	0±0a	0±0a	
Chlorophyceae	35694±9293ª	45000±7051a	42361±5684a	48333±13581a	
Anabaena	5000±2927a	5000±3960a	6111±4654a	3056±2457a	
Aphanocapsa	0 ± 0^a	0±0a	0 ± 0^a	0±0a	
Chroococcus	2917±2917a	4861±2457a	3611±2819a	6111±6111ª	
Gloeocapsa	0±0°a	0±0a	0±0a	0±0 ^a	
Gomphosphaeria	0±0a	0±0a	0±0a	0±0a	
Merismopedia	5694±2183ª	2361±1450 ^b	3194±1707 ^{ab}	3750±2774ab	
Microcystis	2500±1735a	556±556a	2222±605a	2361±1187a	
Oscillatoria	3333±1463a	6111±1325 ^a	4444±1806°	4444±1602a	
Cyanophyceae	19444±9343ª	18889±4728ª	19583±8819a	19722±11056a	
Euglena	5417±636a	7778±1325a	8333±1869a	8056±1869a	
Phacus	1111±605 ^a	1111±735a	972±605a	1667±636a	
Trachalomonas	5556±3522a	5139±2102a	4861±1368 ^a	7500±962a	
Euglenophyceae	12083±2546a	14028±1187a	14167±1502a	17222±3266a	
Batrachosperium	0±0a	0±0a	0±0a	0±0a	
Lemanea	0±0ª	0±0ª	0±0a	0±0ª	
Rhodophyceae	0±0ª	0±0ª	0±0a	0±0ª	
Oedogonium	10139±6590a	20556±1038a	25972±17783°	10417±5320 ^a	
Pithophora	0 ± 0^a	0±0a	2083±2083a	0±0a	
Uronema	0±0ª	0±0ª	2222±2222ª	972±972ª	
Other	10139±6590 ^a	20556±10348 ^a	30278±15696 ^a	11389±5700a	
Total Phytoplankton	107222±31753 ^a	125417±19731 ^a	140694±35210 ^a	127639±41629 ^a	
Difflugia	2639±845a	3611±1822a	4722±1707 ^a	2361±1773 ^a	
Sarcodina	2639±845a	3611±1822a	4722±1707 ^a	2361±1773 ^a	
Asplanchna	4167±2295a	3194±1211 ^a	3056±911a	2639±1325 ^a	
Brachionus	3611±2312ab	2917±1102 ^b	6111±1773°	4722±2650ab	
Filinia	0 ± 0^{a}	0 ± 0^{a}	0±0a	0±0a	
Keratella	972±972ª	278±278 ^a	417±417 ^a	556±556 ^a	
Lecane	556±556a	278±278 ^a	556±556a	694±694 ^a	
Polyarthra	0±0a	0±0a	0 ± 0^{a}	0±0a	
Trichocerca	0±0 ^a	0±0 ^a	0±0°	0±0 ^a	
	U±U"	U±U"	U±U"	U±U"	

	Treatment			
Group	Whole Bamboo	Bottle	Split Bamboo	Banana Stem
Rotifera	9306±972a	6667 ± 636^a	10139 ± 1002^a	8611 ± 1470^{a}
Cyclops	972±501a	833±481ª	556±556a	556±556a
Dioptomus	0 ± 0^a	0 ± 0^a	0 ± 0^a	0 ± 0^{a}
Daphnia	139±139a	0 ± 0^a	$278{\pm}278^a$	0±0a
Diaphanosoma	0 ± 0^a	0 ± 0^a	0 ± 0^a	0±0a
Moina	0±0a	0 ± 0^a	0 ± 0^a	0±0a
Nauplius	833±481a	694±367a	556±139a	556±278 ^a
Crustacea	1944±972a	1528 ± 845^{a}	972±972a	1111±735 ^a
Total Zooplankton	13889±911a	11806±2650a	15833±1879a	12083±3014a

Table 8. Periphyton biomass in four substrate treatments

	Whole Bamboo	Bottle	Split Bamboo	Banana Stem		
Chitwan						
Dry matter (g/cm ²)	0.0200 ± 0.0019^a	$0.0260 {\pm} 0.0070^a$	$0.0244{\pm}0.0024^a$	$0.0188 {\pm} 0.0073^a$		
Ash content (g/cm ²)	0.0130 ± 0.0011^a	0.0193 ± 0.0053^a	0.0168 ± 0.0022^a	$0.0124 {\pm} 0.0055^a$		
Ash free dry matter (g/cm ²)	0.0070 ± 0.0010^a	$0.0068 {\pm} 0.0018^a$	0.0076 ± 0.0011^a	0.0063 ± 0.0019^a		
	Nav	walparasi				
Dry matter (g/cm ²)	$0.0271\pm0.0099a$	0.0409±0.0056a	0.0292±0.0068a	$0.0432 \pm 0.0123a$		
Ash content (g/cm ²)	0.0157±0.0071a	$0.0313 \pm 0.0044a$	0.0205±0.0061a	$0.0337 \pm 0.0107a$		
Ash free dry matter (g/cm ²)	0.0114±0.0031a	0.0095±0.0012a	0.0087±0.0010a	$0.0095 \pm 0.0025a$		

Fish production was lower in Chitwan because a flood hit the district in August 2017, when flood water entered the ponds and swept away fish. Dikes also were damaged and fish escaped. Villagers caught pond fish from rice fields and roads later (see picture in Figure 1). On final harvest, 0 to 315 fish were obtained from the ponds in Chitwan. There were no fish in the pond of Ranjita Mahato, while 2, 5, and 9 fish were harvested from ponds of Saraswati Chaudhary, Bijaya Chaudhary, and Gunja Chaudhary, respectively, even though the number of stocked fish ranged from 71 to 824. Highest combined net fish yield was found in control ponds (2.83 t·ha-1·yr-1) and lowest in banana midrib ponds (0.96 t·ha-1·yr-1), corresponding to carp survival of 40% and 26%, respectively (Table 9). Final carp biomass ranged from 6.2 kg/100 m² in banana midrib ponds to 17.4 kg/100 m² in control ponds after 230 days. Final SIS weight ranged from 0.4 kg/100 m² in banana midrib ponds and plastic bottle ponds to 1.0 kg/100 m² in control ponds. Stocked weight of SIS was not accounted because farmers did not weigh SIS while stocking, they just let them enter along canal water through water inlet. Feed conversion ratio varied from 1.7 to 39.4.



Figure 1. Inundated house of Maya Chaudhary and fish caught in the road in front of her house.

Table 9. Yield of carp and SIS $(kg/100 \text{ m}^2)$ in each treatment after 230 days in Chitwan and 210 days in Nawalparasi.

•	Control	Split Bamboo	Whole Bamboo	Banana midrib	Bottle
	Chitwa	n			
Initial Mean Carp Weight (g/fish)	4±0.2	3.9±0.0	3.9±0.0	3.9±0.0	3.9 ± 0.0
Initial Carp Weight (kg/100 m²)	0.6 ± 0.0	0.6 ± 0.0	0.6 ± 0.0	0.6 ± 0.0	0.6 ± 0.0
Initial Total Weight (kg/100 m ²)	0.6 ± 0.0	0.6 ± 0.0	0.6 ± 0.0	0.6 ± 0.0	0.6 ± 0.0
Final mean Carp Weight (g/fish)	258±89	367±79	228±36	180±19	138±70
Final Total Carp Weight (kg/100 m²)	17.4±8.9	10.1±9.5	12.3±0.5	6.2±3.1	10.7±5.6
Final Total SIS Weight (kg/100 m ²)	1.0±0.2	0.8 ± 0.3	0.6 ± 0.3	0.4 ± 0.1	0.4 ± 0.2
Final Combined Total Weight (kg/100 m²)	18.4±0.8	11.0±9.4	12.9±0.5	6.6±3.0	11.0±5.7
Combined Total weight gain (kg/100 m²)	17.8±8.9	10.4±9.4	12.3±0.5	6.0 ± 3.0	10.5±5.7
Survival (%)	40±8	13±16	38±7	26±12	34±17
Combined GFY (t·ha-1·yr-1)	2.93±1.40	1.74±1.49	2.05 ± 0.08	1.05 ± 0.48	1.75±0.91
Combined NFY (t·ha-1·yr-1)	2.83±1.41	1.65±1.49	1.96 ± 0.08	0.96 ± 0.48	1.66±0.91
Feed Conversion Ratio	3.8±1.7	39.4±79.9	1.7±0.3	8.1±3.5	15.8±17.4
	Nawalpar	asi			
Initial Mean Carp Weight (g/fish)	3.9 ± 0.0^{a}	3.9 ± 0.0^{a}	3.9 ± 0.0^{a}	3.9 ± 0.0^{a}	3.9±0.0a
Initial Carp Weight (kg/100 m²)	0.6 ± 0.0^{a}	0.6 ± 0.0^{a}	0.6±0.0 a	0.6 ± 0.0^{a}	0.6 ± 0.0^{a}
Initial Total Weight (kg/100 m²)	0.6 ± 0.0^{a}	0.6 ± 0.0^{a}	0.6±0.0 a	0.6 ± 0.0^{a}	0.6 ± 0.0^{a}
Final Mean Carp Weight (g/fish)	218±36a	229±17 ^a	228 ± 22^{a}	$253{\pm}10^a$	$260{\pm}16^a$

	Control	Split Bamboo	Whole Bamboo	Banana midrib	Bottle
	Chitwa	n			
Final Total Carp Weight (kg/100 m ²)	20.9±2.8a	22.3±1.5a	23.3±4.3 a	24.6 ± 0.7^{a}	24.9±2.1a
Final Total SIS Weight (kg/100 m²)	0.4 ± 0.0^{a}	0.4±0.1a	0.5±0.1a	0.6±0.1a	0.4±0.1a
Final Combined Total Weight (kg/100 m²)	21.3±2.8a	22.6±1.6a	23.9 4.3a	25.2±0.6a	25.4±2.2a
Combined Total weight gain (kg/100 m²)	20.7 ± 2.8^a	22.0±1.6a	23.2±4.3 a	24.5 ± 0.6^{a}	24.7 ± 2.2^a
Survival (%)	67±3ª	67±6a	69±6a	69±4 ^a	69±7a
Combined GFY (t/ha/yr)	3.70 ± 0.4^{a}	3.93 ± 0.28^a	4.15 ± 0.74^{a}	4.38 ± 0.10^{a}	4.41 ± 0.37^a
Combined NFY (t·ha-1·yr-1)	3.59±0.49a	3.82 ± 0.28^{a}	4.03 ± 0.74^{a}	4.27 ± 0.10^{a}	4.30 ± 0.37^{a}
Feed Conversion Ratio	2.0 ± 0.0^{b}	1.5±0.1a	1.7±0.2ab	1.8±0.1ab	1.9±0.2ab

Overall fish production was highest in the treatments with plastic bottles or banana midribs as substrate (Table 9). Unfortunately, the flooding in Chitwan resulted in such a disruption to fish production that we could not determine treatment effects at that location. For Nawalparasi, final total weight of carp and combined NFY was higher in substrate ponds than control ponds, indicating that rohu and common carp utilized periphyton for food. Both final total weight of carp and combined NFY was 19% higher in ponds with plastic bottles than control ponds. Final total weight of SIS in banana midrib ponds was 50% higher than control, strip bamboo, and plastic bottle treatments. Feed conversion ratio was lower than 1.9 in all substrate ponds. FCR was significantly lower (P<0.05) in split bamboo ponds than control ponds but it was similar to values in other substrate ponds.

Gross margin and gross return were highest in the plastic bottle and banana leaf treatments, but due to variability from the flooding, none of these results were statistically significant. Once again, we could not determine these values accurately for Chitwan. Feed cost was less than NRs 866 in substrate treatments in Chitwan because ration was reduced after assessing fish biomass in each pond after flood (Table 10). Related to feed cost, total variable cost was also low in all substrate treatments. Feed cost ranged from NRs 1066 in split bamboo ponds to NRs1489 per 100 m² in 210 days in banana midrib ponds while total variable cost ranged from NRs 2124 in split bamboo ponds to NRs 2509 per 100 m² in 210 days in banana midrib ponds in Nawalparasi. Both gross return and gross margin were higher in substrate ponds than control ponds. Gross return was 19% higher in plastic bottle treatments than controls while gross margin was 30% higher in plastic bottle treatments ponds than controls.

Table 10. Gross margin (Rs/100 m² pond) analysis for each treatment after 230 days in Chitwan and 210 days in Nawalparasi.

	Control	Split bamboo	Whole bamboo	Banana leaf	Bottle
		Chitwan			
Cost					
Carp fingerlings	494±5	487±1	488 ± 0	485±1	487 ± 0
Lime	72±0	72±0	72±0	72 ± 0	72±0
Urea	107±2	112 ± 2	112±1	110±1	112 ± 0
DAP	212±8	217±4	218±1	222±1	220±20
Feed	1164±59	855±70	660±115	866±89	842±124
Total Variable Cost	2049±56	1745 ± 68	1549±115	1756 ± 90	1734±106
Return					
Carp	4697 ± 2408	2738±2559	3315 ± 127	1672 ± 847	2879±1506
SIS	208±38	164 ± 52	125 ± 68	81±19	75±32

	Control	Split bamboo	Whole bamboo	Banana leaf	Bottle
Gross Return	4905±2391	2902±2538	3440±131	1753±829	2954±1537
Gross Margin	2856±2445	1157±2479	1890 ± 103	-2 ± 822	1220±1561
•		Nawalpara	ısi		
Cost		-			
Carp fingerlings	488±0	487±1	487 ± 0	488 ± 0^{a}	488 ± 0
Lime	72±0	72 ± 0	72 ± 0	72±0	72 ± 0
Urea	164±1	166 ± 0	166±0	165±1	166 ± 0
DAP	329±1	333±2	330±1	331±1	245±89
Feed	1368±192 a	1066 ± 75^{a}	1306 ± 288^{a}	1452 ± 117^{a}	1489 ± 25^{a}
Total Variable Cost	2422±193a	2124 ± 74^{a}	2362 ± 287^{a}	2509±115a	2460±111a
Return					
carp	6258 ± 846^{a}	6678±461a	6993±1281a	7386 ± 206^{a}	7485 ± 636^{a}
SIS	83±0a	75±17 ^a	109±26a	115 ± 27^{a}	87±13 ^a
Gross Return	6342 ± 846^{a}	6753±473a	7102 ± 1283^a	7501 ± 180^{a}	7572±643a
Gross Margin	3920±655a	4630±440a	4741 ± 1062^{a}	4992 ± 164^a	5111 ± 749^{a}

DISCUSSION

The four treatments for substrate resulted in considerably higher fish production and gross margin than the control without substrate. Fish production was calculated by summing weight of fish consumed by farmers and weight of fish netted during final harvest. Among substrates used, plastic bottles gave higher fish yield than natural substrates which differed from results obtained in previous work, where natural substrates such as bamboo produced higher fish yield (Van Dam et al., 2002). Most likely differences in the surface area of each substrate type, the exposure to sunlight, and the attraction of algae to the substrate surface made each substrate type a unique environment for production of periphyton and the resulting difference in fish production. Better FCR in substrate ponds than control ponds in Nawalparasi showed that periphyton reduced feed input and feed cost. FCR was much better in the present trial than in the previous field trial as reported by Jha et al. (2018) in the same place.

Fish production was affected by flood in both districts, but the effect of flooding was more serious in Majhui, Chitwan than in Seri and Nandapur, Nawalparasi. There is a stream very close to Majhui that exceeded its banks due to continuous intense rainfall, and water inundated the village. Water entered the ponds and fish escaped. Moreover, the height of dikes for ponds in Majhui was also lower than Nawalparasi, so fish escaped easily. Physical damage also occurred to ponds in Majhui. It took almost three days for flood water to recede in the village. Ponds were netted to assess status of fish nearly one month later. High water level in the pond interfered with netting and fish assessment was not effective. Hence, our data on production and economics does not give real picture of farmers' ponds in Chitwan. The situation was better in Nawalparasi because survival and production of carp was satisfactory and a treatment effect could be seen. But the variation in ponds even there probably resulted in the lack of statistical significance for any of these results. Around 67 to 69% of stocked carp were recovered from ponds there, which is comparable with data from Jha et al. (2018). FCR was very high except for ponds with substrates of whole bamboo in Chitwan, again due to flooding. We could not assess number and weight of fish in ponds to adjust ration after the flood. Due to delay in our fish assessment and farmers putting effort in rehabilitation rather than fish and pond sampling, farmers fed randomly. This hiked feed quantity and FCR in Chitwan.

Gross return and gross margin was very low in all treatments in Chitwan while farmers received good return and margin in Nawalparasi. In Chitwan, 6 farmers (Control-1, split bamboo-2, banana midrib-2, plastic bottle-1) lost money from ponds due to low production caused by the flood. Feed cost and

total variable cost was higher in Nawalparasi than Chitwan because survival of carp was higher in Nawalparasi ponds and feed adjustment was done according to fish weight.

Among substrates used, farmers complained about using banana midrib because they had to replace it 3-4 times during the trial period. Banana midrib decayed in ponds within 2 months which created trouble for farmers. Although banana is easily available from the farm and has multiple uses, replacement effort is important and care should be given on use of it because its decay may cause oxygen depletion in the pond. Comparatively, whole bamboo, split bamboo and plastic bottles are more durable. Based on fish production, profit, and availability, plastic bottles appear to be a reasonable alternative to split bamboo mats for periphyton substrate.

Workshop for Non-Adopting Farmers

A workshop for non-adopting farmers was conducted at Hotel Gangotri, Narayangarh, Chitwan, on 8 January 2018. The objective of the workshop was to disseminate the results of the field trial. Altogether 42 participants including 31 from Chitwan and 11 from Nawalparasi participated in the workshop. Among them, 41 were farmers and 1 was NGO Chair (Rural Integrated Development Society). By gender, 62% of participants were female while 38% were male, while 54% were from Chitwan and 7% from Nawalparasi.

Table 11. Participants of the workshop for non-adopting farmers

Participants	Chitwan			Nawalparasi		
	Male	Female	Total	Male	Female	Total
Farmer	7	23	30	8	3	11
NGO	1		1			
Total	8	23	31	8	3	11

During the workshop, periphyton technology and results of the on-farm trial were presented. The presentation was followed by group discussion on the effectiveness of different substrates and adoption of the technology. Farmers were divided into three discussion groups. After the discussion, each female lead reported on their discussion. Farmers' perceptions on periphyton-based technology and its effectiveness are summarized in Table 12.

Table 12. Outputs of workshop for non-adopting farmers

Particular	Group I	Group II	Group III
Periphyton technology	useful	useful to small scale farmers because this technology helps in reducing pond inputs and increasing production and income	Good for small scale farmers as it helps in reducing feed cost in ponds.
Appropriate substrate	Plastic bottles because they are easy to use and durable for many culture cycles Banana leaves were consumed by grass carp but the midrib could be left in the pond as substrate	Plastic bottles tied in a ring of hollow plastic pipe would be better since this is easier to build and helps in pollution control by using waste material	Whole bamboo with no leaves would be better as it is available, easy to use, and better for periphyton growth. Using whole bamboo with its branches also helps in reducing predation and theft.
Effectiveness/ adoption	Effective and adoptable	Adoptable but needs technical help from AFU	Adoptable but needs technical help
Constraints	No	Exposed parts of substrates may become place for birds to prey on small fish.	Substrates cause problems during partial harvesting of SIS and sampling fish. They may also cause some wounds in fish.
Solution		Gill net may be placed on the ring of pipe to stop birds from using substrate as resting place	

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