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EFFECT OF RICE STRAW ON LIMONOLOGICAL PARAMETERS AND BACTERIOLOGICAL COMMUNITIES IN EARTHEN FISH PONDS

Diaa A. Al-Kenawy and Taghred M. Ahmed

[1]*Limnology department, Central Laboratory for Aquaculture Research, Agricultural Research Center, Egypt.*

[2]*Fish Disease department, Central Laboratory for Aquaculture Research, Agricultural Research Center, Egypt.*

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ABSTRACT

The present study carried out in the WorldFish center research station at Abbassa, Egypt, during the period from June to November 2017 to investigate the effect of applying rice straw as periphyton substrates in fertilized earthen fishponds on some water quality parameters and bacteriological communities. Six earthen ponds, of 1000 m^3 average area each, divided into two groups, each of three replicates. Dry rice straw added to one of the two groups in a rate of 45 kg/pond to act as periphyton substrates (T_2), while the other group left without applying rice straw to act as control group (T_1). Some water quality parameters namely water temperature (ºC), dissolved oxygen (DO, mg/l), hydrogen ion concentration (pH), total ammonia nitrogen (TAN, mg/l), unionized ammonia (NH_3, mg/l), total hardness (TH, mg/l as $CaCO_3$), Calcium (Ca, mg/l), Magnesium, (Mg, mg/l), total dissolved solids (TDS, mg/l), total nitrogen (TN, mg/l), and total phosphorous (TP, mg/l) were measured fortnightly in source water, as well as in the two treatments' ponds (T1 and T2). The bacterial communities also monitored and counted in water and fish samples. Most important results obtained summarized as follow: there were no significant ($P < 0.05$) differences in the mean average of water temperature among source water and the two treatments, while the values of dissolved oxygen, pH, TDS, TN, TP in the source water were significantly ($P < 0.05$) lower than those in both (T_1) and (T_2) treatments. Levels of TH and Ca were significantly ($P < 0.05$) higher than those in both (T_1) and (T_2) treatments. Mean values of most investigated parameters were significantly different between the two treatments in some months. However, all measured parameters and the bacteriological communities in the ponds of the two treatments were within the acceptable ranges recommended for successful aquaculture.

INTRODUCTION

Rice straw is relatively low cost and is widely available as crop byproduct in farms. It has been used to mitigate turbidity in fishponds (Yi *et al*., 2003) and to develop bacterial biofilm and periphyton (Ramesh *et al*., 1999) as a source of natural food for fish. Periphyton-based pond culture is a simple and cheap way of producing natural food for herbivorous and omnivorous fishes, such as tilapia and carps. In this culture system, woody branches, bamboo poles or any other hard substrates are planted or fixed in shallow waters such as ponds, lagoons, reservoirs, to enhance the growth of sessile autotrophic and heterotrophic aquatic biota known as periphyton (El-Sayed, 2020). This system enhances natural productivity, which provides natural food for herbivorous and omnivorous fishes in ponds.

Periphyton-based aquaculture is such a low cost technology. In eco-friendly approach in closed water aquaculture, there is possibility for increased energy and nutrient transfer efficiencies with additive effects of periphyton and plankton. There would be more preferable natural food for carps, tilapia and freshwater prawn (Yadav *et al*., 2017). Sakr *et al*. (2015) evaluated periphyton as a natural food source for fingerling Nile tilapia (2.37 g) fed reduced dietary protein levels in cages for 70 days. The results revealed that periphyton provided an additional natural food source for caged fish and positively affected growth performance and feed efficiency. The authors also indicated that periphyton substrates provision, would likely lead to significant reduction in nitrogen load and other particulate nutrients in the surrounding aquatic environment, not to mention the nutrients absorbed directly by the periphyton.

Improving the conversion of nutrients into harvestable products, through adoption of periphyton-based production into existing pond systems, is one solution worth exploring. The results of studies which investigated the impact of applying periphyton substrates in aquaculture ponds, vary from no effect (Shrestha and Knud Hansen, 1994; Faruk-ul-Islam, 1996 and Azim *et al*., 2001a) to a 40–80% increase in fish yield in ponds with artificial substrates

compared to control ponds (Ramesh *et al*., 1999; Wahab *et al*., 1999; Azim *et al*., 2001b; Keshavanath *et al*., 2001 and Sakr *et al*., 2015).

Water chemistry of a pond determines its suitability for fish and deterioration of water quality parameters may harm the cultured species due to the continuous fluctuations in its physicochemical and biological parameters (Shahabuddin *et al*., 2012).

The beneficial bacteria in fish ponds are naturally exist and considerably safe for fish, and people. They are acting to maintain clear healthy water, breaking down organic wastes and ammonia that generated from fish waste, reducing nitrite and nitrate, reducing nutrient load in ponds and balancing the aquatic ecosystem. Aquatic bacteria, through the process of decomposition and as sources of food, play an important role in pond ecosystems and in fish production. Non-beneficial bacteria cause offensive odor in ponds and diseases in fishes. However, fishes have evolved a number of protective adaptations with several unique traits against bacteria and other organisms in their outside coverings including scales, fins and in particular, protective mucous that cover the body (Ajayi and Okoh, 2014).

Variety of biodegradable and non-biodegradable substrates have been used in the past to enhance fish production in periphyton-based aquaculture systems.

The present study conducted to investigate the effect of applying rice straw mats for enhancing periphyton growth on some water quality parameters as well as the bacterial communities in organically fertilized earthen ponds of Nile tilapia.

MATERIALS AND METHODS

Study area and experimental design:

The present study was carried out in earthen fishponds at the WorldFish center at Abbassa, Egypt, during the period from the beginning of June to the end of November 2017, where six 1000 m^3 earthen ponds were divided into two groups, each of three replicates. The first treatment unhanded as control (T_1),

while the second one (T_2) received dry 45 kg rice straw/pond. Rice straw applied in bundles (each of 60 cm in length and 25 cm in diameter). Bundles dangled from vertical pillars that established along the pond sides. Ponds received fresh water from El-Ismailia canal, which occasionally mixed with well water. All ponds fertilized traditionally with sun dried chicken manure at rate of 15 kg/pond/week.

Sampling and analytical techniques:

Fortnightly water samples were collected from the canal of water supply that sourcing the experimental ponds and from the two investigated treatments, control (T_1) and periphyton (T_2) ponds for determining some water quality parameters as well as the bacteriological communities as described hereafter.

1. Water quality parameter

On site measurements:

Water temperature (T, ºC) and dissolved oxygen (DO, mg/l) were measured by oxygen meter (Hana HI 9142). Hydrogen ion concentrations (pH) were measured by pH meter: (EUTECH instruments PCS tester 35). The water transparency measured in ponds and water source using the Secchi Disc. .

Laboratory analysis:

Total hardness (TH, mg/l) and Calcium (Ca, mg/l) were determined by titration method. Total dissolved solids (TDS, mg/l) were determined by weighing the dissolved solids which remained after drying. Total nitrogen values were determined by kjeldahl method using spectrophotometer (model, WPA Linton Cambridge UK).

Total phosphorous (mg/l) was measured by ascorbic acid method using spectrophotometer (WPA Linton Cambridge UK). All previously mentioned methods conducted according to APHA (2000).

1. Bacteriological communities:

Water samples:

Pond water samples for total bacterial counting were collected from the ponds of the two treatment groups using sterilized bottle samples. Study of these samples was carried out in the microbiological laboratory of Central Laboratory for Aquaculture Research (CLAR). Water samples were collected three times during the study period; the initial one was at the beginning of the experiment, while the second one was after applying rice straw in ponds and the third one just before pond harvest.

The water samples were serially diluted and appropriate diluents of 1 ml samples inoculated with pipette into sterile Petri dish; added NA, shaked and incubated in at 30°C for 24-48 h.

Fish samples:

Three fish were collected from each experimental pond twice; the initial one from the fingerlings at ponds stocking while the second time was at harvest. One gram of fish muscles were grinding in sterile nutrient broth, then serial dilution and prepared similar to water samples.

Nutrient agar medium sterilized at 121°C for about 15-30 min in an autoclave. Working surface area sterilized by disinfectant (95% ethanol).

Periphyton Sample preparation:

Pieces of rice straw was cut from three different spots, and then wrapped in aluminum foil until analysis. Each sample was transferred to an Erlenmeyer flask containing 50 ml distilled water, and mechanically shaken for 3 hours to detach periphyton from the straw, then preserved in 6 % formalin until identification.

Statistical analysis:

Prior to statistical analysis, all data were tested for normality of distribution using the Kolmogoro-Smirnov test. The homogeneity of variances

among different treatments was tested using Bartlett's test. Then data subjected to two-way ANOVA to evaluate the effect of rice straw and month on water quality and bacterial communities.

Differences between means were tested at the 5% probability level using Duncan's test as a post hoc test. All statistical analysis were performed using the SPSS program version 20 (SPSS, Richmond) as described by Dytham (2011).

RESULTS AND DISCUSSION

Water temperature:

Water temperature is the degree of hotness or coldness in the body of a living organism either in water or on land (Lucinda and Martin, 1999). Obtained results of the present work revealed that water temperature values ranged between 19˚C and 31.43 ˚C (Table 1). Obtained data revealed that there were no significant ($P< 0.05$) differences in water temperature values among different investigated sites (water supply source and treatments), which revealing that variations in water temperatures were due to corresponding variations in air temperature. Table 1 clarified that the mean average of water temperature values in source, T1, and T2 were 25.75, 26.6 and 25.66 ˚C, respectively.

According to Delince (1992), the recorded water temperature values alongside the study period are considered within the tolerable range for the Nile tilapia.

Hydrogen ion concentration (pH):

Table 1 showed that pH values alongside the study period ranged between 7.2 and 7.97, while total mean in source, T1 and T2 were 7.36, 7.63 and 7.57, respectively. However all recorded values alongside the experimental period could be considered within the optimal range for fish culture. Boyd and Tucker (1998) stated that for most freshwater species, a pH range between 6.5 and 9.0 is ideal.

Table 1 showed that pH values in source water were significantly ($P < 0.05$) lower than those values in both treatments, which could be attributed to the increase of photosynthesis activity by phytoplankton, where pH changes in surface water result from the interaction of various biotic and abiotic processes (Konsowa, 2007).

Ammonia (total ammonia (NH_3+NH_4) and unionized ammonia):

Ammonia originates from fish execration, microbial analysis for nitrogenous compounds, and break down of amino acids. Ammonia usually used as indicator of organic pollution and become toxic to most aquatic species in concentration over 2-5 mg/l. Therefore, ammonia was considered as one of the most important parameters that must be studied in the water ecosystem (Elewa *et al.*, 2001).

The monthly mean of total ammonia values observed during the study reached 0.1, 0.178 and 0.172 mg/l in source, T1and T2, respectively. While that of unionized ammonia were 0.004, 0.018, and 0.014 mg/l in source, T1 and T2, respectively. Table 1 showed that ammonia values in source water were lower than in T1 and T2. Ammonia in fertilized fishponds could be absorbed by the available phytoplankton, which subsequently converts the nitrogenous compounds into amino acids, which depict a much-idealized algal cell as pointed out by Vymazan (1995).

Water transparency (Secchi disc):

Water transparency is a useful and cheap method to assess water quality (Moussa, 2004). The highest reading of the Secchi disc (SD cm) was observed in source water during June with value of 40.0 cm, while the minimum transparency value was found in (T2) during August with value of 18.67 cm. There was a difference in water transparency recorded between treatment ponds and water source; this may attributed to different depths, feeding, and abundance of phytoplankton and soil particles in ponds, Boyd (1990). In addition, the highest SD reading attributed to low phytoplankton growth, while the lowest one due to high primary productivity (Saeed and Mohammed, 2012).

There were a significant spatial and monthly variations in transparency and between treatments and months ($P < 0.05$) (Table 1).

Dissolved oxygen (DO):

Growth, survival, distribution, behavior and physiology of different aquatic organisms; affected mainly by dissolved oxygen values (Solis, 1988). Oxygen depletion in water leads to poor feeding of fish, starvation, reduced growth and more fish mortality, either directly or indirectly (Bhatnagar and Garg, 2000). Table 1 elucidated that DO alongside the experimental period, were between 3.63 and 6.17 mg/l. According to Lloyd (1992), which could be considered within the tolerable range for most fish species specially tilapia.

Obtained results revealed that there were no significant ($P<0.05$) differences in DO between T_1 and T_2 treatments, which indicates that the application of rice straw as substrates in fishponds has no effect on water dissolved oxygen. Similar results were obtained by Rai *et al*. (2012) in spite of higher rates of rice straw mat, significantly decreased dissolved oxygen values than control.

Total dissolved solids (TDS):

Includes those materials dissolved in the water, such as, bicarbonate, sulphate, phosphate, nitrate, calcium, magnesium, sodium, organic ions, and other ions. These ions are important in sustaining aquatic life. However, high concentrations can result to damage in aquatic organism's cell (Mitchell and Stapp, 1992). TDS values that recorded alongside the experimental period, ranged between 196.67 and 470 mg/l. However, obtained TDS values alongside the study period did not exceeded 400 mg/l except during September; where TDS values in S, T1 and T2 were 470, 414 and 387 mg/l, respectively. A maximum of 400 mg/l of total dissolved solids is permissible for diverse fish population (Naeem *et al*., 2011).

Average TDS values that recorded in Source, T1 and T2 samples were 273.33, 271.78 and 281.33 mg/l, respectively. However, there were no significant ($P<0.05$) differences between TDS values in source and T1, while its values in T2 treatment were slightly higher.

Total hardness (TH):

Hardness is the measure of multivalent cations such as calcium and magnesium in an aquatic body along with other ions such as aluminum, iron, manganese, strontium, zinc, and hydrogen ions. Calcium and magnesium are essential to fish for metabolic reactions such as bone and scale formation (Bhatnagar and Devi, 2013). Data in table 2 revealing that total hardness values recorded alongside the experimental period were between 100 and 240 mg/l, which could considered within the acceptable range for fish culture as recommended by many studies. The ideal value of hardness for fish culture is at least 20 ppm, as recommended by Swann (1997), while Santhosh and Singh (2007) recommend a range of 30-180 mg/l.

Table 2 elucidated that the mean average of total hardness values in different investigated sampling sites (Source, T_1 and T_2) over the experimental period were 196.67, 152.78 and 162.78 mg/l, respectively. Total hardness was, however, higher in ponds of T2 than in T1 during the first four months, then declined to be less than in T1 in the last month. It is noticeable that TH value in source water was significantly ($p<0.05$) higher than those values in both investigated treatments. This may be due to a part of total alkalinity was in the form of potassium carbonates which representing the most water soluble form of the carbonates that do not increase total hardness. While a part of calcium carbonates which represent both water hardness and alkalinity, was precipitated due to the increased of water pH. Many studies stated that K content in chicken manure ranged between 1.13 and 1.83 %, dry weight (Kumara *et al*., 2003; Zayed *et al*., 2013 and Eisa, 2016). SRAC (1992) stated that if calcium carbonate is responsible for both hardness and alkalinity, the concentrations will be similar if not identical. However, where sodium or potassium bicarbonate or carbonates are responsible for alkalinity, it is possible to have low hardness and high alkalinity.

Calcium (Ca):

Calcium is a metallic element, fifth in abundance in the earth's crust, of which it forms more than 3%. It occurs abundantly as limestone ($CaCO_3$),

gypsum ($CaSO_4 \cdot 2H_2O$), and fluorite (CaF_2) (CRC, 2005). Calcium is essential in the biological processes of fish (bone and scale formation, blood clotting and other metabolic reactions). Fish can absorb calcium directly from the water or from food. However, calcium is the most important environmental, divalent salt in fish culture water (SRAC, 1992). Table 2 articulated that Ca concentrations that obtained during the present study were between 10 and 80 mg/l. A recommended range of free calcium in culture waters is 25 to 100 mg/l (63 to 250 mg/l $CaCO_3$ hardness). A value of 100 mg/ l (250 mg/L calcium hardness) matches the calcium concentration of fish blood (SRAC, 1992). It is noticeable that Ca had a corresponding trend, as that of total hardness, where Ca beside Mg constitutes the majority of total hardness value (Bhatnagar and Devi, 2013).

Obtained results revealed that the mean average of Ca concentrations which recorded alongside the experimental period were 53.89, 30.83 and 40.83 mg/l in Source, T1 and T2 sampling sites, respectively. The significant ($p<0.05$) highest value were in source water could be attributed to the increased photosynthesis rate in both investigated treatments. This led to subsequent increase in water pH, which in turn led to the conversion of the most soluble calcium from, bicarbonate form, to the barely soluble carbonate form; what led to the precipitation of Ca and adsorbed to the pond sediment (SRAC, 1992).

Magnesium:

Magnesium and calcium are the predominant divalent metals in normal natural waters. Table 2 showed that Mg alongside the experimental period, were between 4.2 and 47.6 mg/l. Data illuminated that the mean average values of Mg in different sampling sites (source, T1, T2) were 20.77, 20.53, 10.93 mg/l, respectively.

Total nitrogen (TN):

Nitrogen is an essential nutrient for plants and animals. However, an excess amount of nitrogen in a waterway may lead to low levels of dissolved oxygen and negatively alter various plant life as well as aquatic organisms. Sources of nitrogen include wastewater treatment plants, runoff from fertilized lawns and croplands, runoff from animal manure and industrial discharges.

Total nitrogen is the sum of total Kjeldahl nitrogen (organic nitrogen and ammonia) and total oxidized nitrogen (nitrate and nitrite nitrogen). An acceptable range of total nitrogen in water is 2 to 6 mg/l (EPA, 2013). Table 2 revealing that TN values were between 0.96 and 2.77 mg/l that fall within the acceptable range.

As shown in Table 2, the mean average of TN in different sampling sites (Source, T1 and T2), were 1.15, 1.40 and 1.57, respectively. TN values in source water were significantly ($P<0.05$) lower than those values in T_1 and T_2 treatments. This is reasonable because of fertilizing the ponds and the existence of fish wastes in the treated ponds. Similarly, Abdel-Warith (2013) stated that nitrogenous compounds values had increased in pond water that received chicken manure as organic fertilizer.

Total phosphorus (TP):

Phosphorus usually found in the form of inorganic and organic phosphates in natural waters. Inorganic phosphates include orthophosphate and polyphosphate while organic forms are those organically bound phosphates. Phosphorus is a limiting nutrient needed for the growth of aquatic plants and algae. However, excess concentrations especially in rivers and lakes can result to algal blooms (Mueller and Helsel, 1999). TP values obtained alongside the present work were between 0.016 and 0.267 mg/l (Table 2). Quality standards on phosphorous levels (in different forms) set by Australia, Malaysia, New Zealand, Norway, Philippines and United States, are between 0.02 and 0.20 mg/l for freshwater (ANZECC, 2000). Furnas (1992) stated that total phosphorus associated with suspended matter in unpolluted tropical rivers normally ranges between 0.62 and 1.86 mg/l.

Table 2 indicated that the mean average of TP values in different investigated sampling sites (Source, T_1 and T_2) were 0.107, 0.103 and 0.134 mg/l, respectively. Despite that these values were pointedly close to each other, but the recorded value in source water was significantly ($P<0.05$) lower than those values in the two investigated treatments, as a normal result of fertilization with chicken manure. Kumara *et al*. (2003) stated that phosphorous

values had increased in pond water that received chicken manure as organic fertilizer.

High rice straw loading rates deteriorated water quality parameters and became unsafe for fish culture (Rai *et al*., 2012). However, rice straw rates used during the present work does not negatively affect water quality aspects. Water quality parameters appeared to be within the acceptable ranges for tilapia culture (Bolivar *et al.* 2004).

Table 1. Monthly mean levels of different water quality parameters (Temperature, pH, Total ammonia, unionized ammonia, transparency, and dissolved oxygen) in the water source and the treated ponds.

Month	Site	Temp (C°)	pH	T. Ammonia (mg/l)	Unionized Ammonia(mg/l)	S. D. (cm)	D.O (mg/l)
Jun	S	27.50± 0.001 ^{f*}	7.50± 0.001 ^{cde}	0.10± 0.001 ^{b}	0.003± 0.0001 ^{e}	40.00± 0.001 ^{a}	7.50± 0.001 ^{a}
	T1	28.73± 0.20 ^{e}	7.97± 0.09 ^{a}	0.10± 0.001 ^{b}	0.009± 0.002 ^{de}	26.00± 1.15 ^{cde}	6.17± 1.30 ^{abcde}
	T2	29.30± 0.001 ^{d}	7.80± 0.17 ^{ab}	0.10± 0.001 ^{b}	0.008± 0.003 ^{de}	25.00± 2.89 ^{de}	5.63± 1.26 ^{defg}
July	S	30.00± 0.001 ^{c}	7.40± 0.001 ^{defg}	0.10± 0.001 ^{b}	0.002± 0.0001 ^{e}	35.00± 0.001 ^{b}	7.40± 0.001 ^{ab}
	T1	31.43± 0.03 ^{a}	7.53± 0.20 ^{cde}	0.17± 0.03 ^{a}	0.010± 0.006 ^{de}	28.33± 0.88 ^{cd}	5.83± 0.54 ^{bcdef}
	T2	31.40± 0.06 ^{a}	7.63± 0.07 ^{bcd}	0.13± 0.03 ^{b}	0.008± 0.001 ^{de}	23.67± 0.88 ^{ef}	5.67± 0.39 ^{cdefg}
August	S	30.00± 0.001 ^{c}	7.20± 0.001 ^{gh}	0.10± 0.001 ^{b}	0.002± 0.0001 ^{e}	35.00± 0.001 ^{b}	7.20± 0.001 ^{abcd}
	T1	30.13± 0.03 ^{bc}	7.47± 0.09 ^{def}	0.20± 0.001 ^{a}	0.008± 0.001 ^{de}	23.33± 2.19 ^{efg}	5.30± 0.40 ^{efg}
	T2	30.23± 0.03 ^{b}	7.83± 0.03 ^{ab}	0.20± 0.001 ^{a}	0.016± 0.002 ^{cd}	18.67± 1.33 ^{hi}	4.90± 0.40 ^{efgh}
Sept	S	26.00± 0.001 ^{h}	7.60± 0.001 ^{bcd}	0.10± 0.001 ^{b}	0.003± 0.0001 ^{e}	35.00± 0.001 ^{b}	7.30± 0.001 ^{abc}
	T1	26.63± 0.03 ^{g}	7.80± 0.06 ^{ab}	0.20± 0.001 ^{a}	0.011± 0.002 ^{de}	22.67± 3.84 ^{efgh}	4.10± 0.12 ^{gh}
	T2	26.67± 0.03 ^{g}	7.73± 0.09 ^{abc}	0.20± 0.001 ^{a}	0.010± 0.002 ^{de}	22.67± 1.20 ^{efgh}	3.63± 0.43 ^{h}
Oct	S	22.00± 0.001 ^{k}	7.10± 0.001 ^{h}	0.10± 0.001 ^{b}	0.009± 0.0001 ^{de}	30.00± 0.001 ^{c}	7.20± 0.001 ^{abcd}
	T1	22.73± 0.13 ^{i}	7.30± 0.06 ^{efgh}	0.20± 0.001 ^{a}	0.028± 0.003 ^{b}	19.00± 1.00 ^{ghi}	4.37± 0.47 ^{fgh}
	T2	22.43± 0.07 ^{j}	7.23± 0.09 ^{fgh}	0.20± 0.001 ^{a}	0.024± 0.004 ^{bc}	18.67± 0.88 ^{hi}	4.37± 0.07 ^{fgh}
Nov	S	19.00± 0.001 ^{m}	7.40± 0.001 ^{defg}	0.10± 0.001 ^{b}	0.003± 0.0001 ^{e}	30.00± 0.001 ^{c}	7.00± 0.001 ^{abcd}
	T1	20.00± 0.10 ^{l}	7.73± 0.07 ^{abc}	0.20± 0.001 ^{a}	0.041± 0.008 ^{a}	15.33± 0.67 ^{i}	5.67± 0.15 ^{cdefg}
	T2	19.93± 0.03 ^{l}	7.20± 0.001 ^{gh}	0.20± 0.001 ^{a}	0.015± 0.0001 ^{cd}	19.67± 0.33 ^{fghi}	5.30± 0.15 ^{efg}
Overall mean	S	25.750	7.367	0.100	0.004	34.167	7.267
	T1	26.608	7.633	0.178	0.018	22.443	5.240
	T2	26.660	7.570	0.172	0.014	21.392	4.917
TWO way ANOVA				*P value*			
Month		0.0001	0.0001	0.0001	0.0001	0.0001	0.008
Site		0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
Month× Site		0.0001	0.0001	0.0001	0.0001	0.055	0.579

* Means with same letters within a column are not significantly different.

Table 2. Monthly mean levels of some water quality parameters (TDS, T.H., Ca, Mg, T.N, and T.P) in the water source and the treated ponds.

Month	site	TDS (mg/l)	T.H (mg/l as $CaCO_3$)	Ca (mg/l)	Mg (mg/l)	T.N (mg/l)	T.P (mg/l)
June	S	220.00±0.	210.00±0.001	63.33±3.33 [c]	9.80±0.00 [i]	1.20±0.001	0.100±0.0001 [fg]
	T1	271.67±3.	193.33±1.67 [e]	40.00±0.001	26.13±0.47 [f]	1.27±0.02 [g]	0.040±0.0001 [j]
	T2	282.67±4.	240.00±0.001	70.00±0.001	18.20±0.00 [g]	1.22±0.02 [g]	0.030±0.0001 [jk]
July	S	220.00±0.	130.00±0.001	70.00±0.001	12.60±0.00 [h]	1.60±0.001	0.110±0.0001 [f]
	T1	256.00±2.	120.00±0.001	30.00±0.001	45.00±0.00 [b]	1.28±0.01 [g]	0.150±0.0001 [e]
	T2	320.33±5.	140.00±0.001	50.00±0.001	4.20±0.00 [l]	1.32±0.02	0.100±0.0001 [fg]
August	S	270.00±0.	200.00±0.001	80.00±0.001	0.0 ±.000 [n]	0.90±0.001	0.070±0.0001 [i]
	T1	251.00±6.	170.00±0.001	50.00±0.001	12.60±0.001	1.00±0.001	0.016±0.0001 [k]
	T2	236.33±8.	180.00±0.001	60.00±0.001	8.40±0.001 [j]	0.95±0.001	0.175±0.0001 [cd]
Sept	S	470.00±0.	220.00±0.001	70.00±0.001	12.60±0.00 [h]	0.90±0.00 [i]	0.100±0.0001 [fg]
	T1	414.00±7.	110.00±0.001	40.00±0.001	2.80±0.001	1.72±0.02 [d]	0.090±0.0001 [gh]
	T2	387.33±2.	100.00±0.001	40.00±0.001	0.0 ± 0.000	2.17±0.03 [b]	0.160±0.0001 [de]
Oct	S	230.00±0.	200.00±0.001	20.00±0.001	42.00±0.001	0.90±0.00 [i]	0.080±0.0001 [hi]
	T1	196.67±8.	180.00±0.001	10.00±0.001	7.00±0.001 [k]	1.07±0.03 [h]	0.090±0.0001 [gh]
	T2	196.67±3.	180.00±0.001	10.00±0.001	7.00±0.001 [k]	0.96±0.01	0.070±0.0001 [i]
Nov	S	230.00±0.	220.00±0.001	20.00±0.001	47.60±0.001	1.40±0.00 [f]	0.180±0.0001 [c]
	T1	241.33±1.	143.33±3.33 [h]	15.00±0.001	29.63±0.93 [d]	2.03±0.03 [c]	0.233±0.017 [b]
	T2	264.67±2.	136.67±3.33 [i]	15.00±0.001	27.77±0.93 [e]	2.77±0.15 [a]	0.267±0.017 [a]
Overall mean	S	273.33	196.67	53.89	20.77	1.15	0.107
	T1	271.78	152.78	30.83	20.53	1.40	0.103
	T2	281.33	162.78	40.83	10.93	1.57	0.134
TWO way ANOVA		*P value*					
Month		0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
Site		0.001	0.0001	0.0001	0.0001	0.0001	0.0001
Month× Site		0.0001	0.0001	0.0001	0.0001	0.0001	0.0001

* Means with same letters within a column are not significantly different.

Total bacterial count in water samples of the examined ponds:

Figure 1 showed the total bacterial count in water of the examined ponds before and after adding the rice straw in water. Total bacterial count performed three times throughout the experimental period; the initial sample collected before adding rice straw and fertilizers to ponds, the second sample made few days after adding rice straw and fertilizers, while the final sample collected at the end of the experiment, just before harvesting the fish.

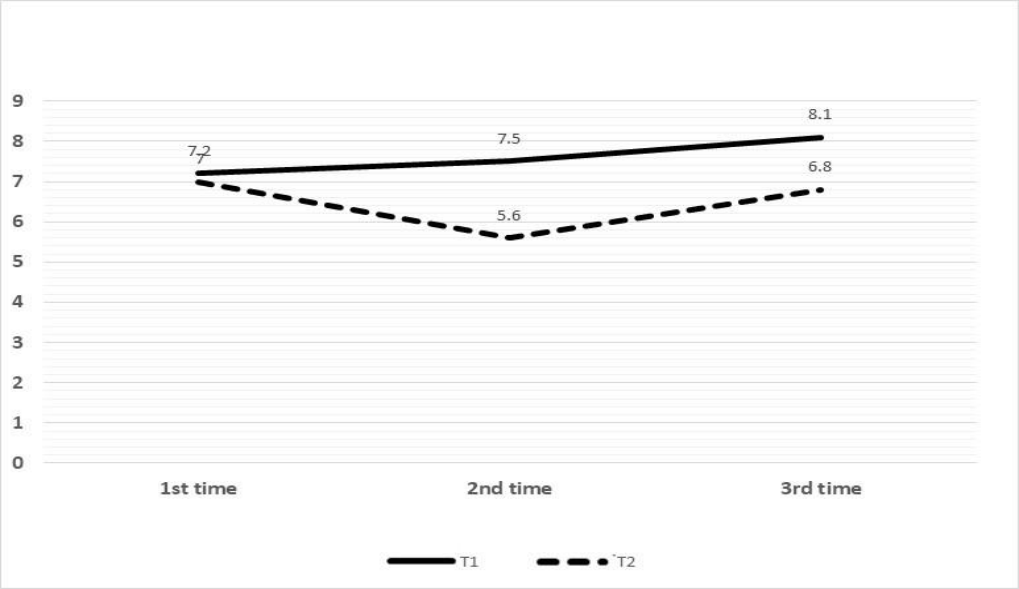

Figure 1. Total bacterial count ($x \times 10^3$ CFU/ml) in water of the two treatments (T1 and T2).

Obtained results revealed that the bacterial count was almost similar in the initial water samples (before adding rice straw to ponds) of both treatments in (7.2 and 7.0×10^3 CFU/ml in T1 and T2), then decrease in second sample of T2, where ponds treated with rice straw, to reach 5.6×10^3 CFU/ml. While in the control ponds, T1, it remained higher and even increased to level of 7.5×10^3 CFU/ml. The bacterial counts then increased in both treatments after stocking ponds with fish to reach 8.1×10^3 and 6.8×10^3 CFU/ml in T1 and T2, respectively. It is clear enough that application of rice straw in fishponds as a periphyton substrate has advantage of reducing bacterial abundance in water.

Total bacterial count on fish samples from the examined ponds:

Figure 2 shows the total bacterial count on fish from the examined ponds. The initial sample taken from the fish at stocking, while the second one was at harvest.

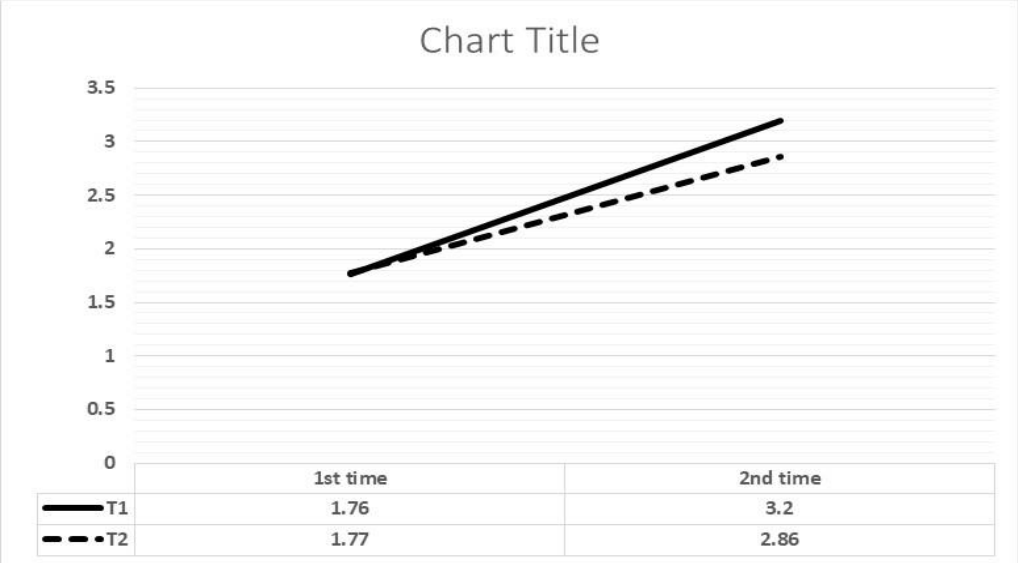

Figure 2. Total bacterial count ($x \times 10^3$ CFU/ml) on fish from the two treatments (T1 & T2).

The results of total bacterial count of fish muscles from treated ponds revealed that it's count increase in both treatments after adding fertilizers and rice straw to reach 3.2×10^3 CFU/ml in T1 (control) and $2.86 \times {}^{103}$ CFU/ml in T2 (ponds with rice straw). Again, and similar to findings in ambient water samples, it is clear that the application of rice straw has reduced the bacterial count in fish body. However, levels of bacterial count in both treatments remained under permissible limits in cultured fish.

Similar results indicated by Kumar *et al*. (2015) who reported that there was significant difference ($p<0.05$) in total bacterial count (TBC) in water of substrate based ponds compared with control ponds with no substrate.

For both types of the tested water, applying of rice straw as periphyton substrate lowered the count of bacteria. The re-increase in total bacterial count

in water of the two treatments could be attributed to the fact that agricultural wastes are energy rich substrates; so, it has been employed for the cultivation of microorganisms as reported by Ezejiofor *et al*. (2014).

Odeyemi and Agunbiade (2012) mentioned that values of total bacterial count in water of fishponds and river in Ila-Orangun, Osun State, Nigeria were between 6.4 and 7.4 x 10^4 CFU/ml, respectively. Saha *et al*. (2012) reported that the maximum aerobic heterotrophic bacterial count of four pond water samples of Dhaka Metropolis ranged between 6.92×10^4 and 1.72×10^6 CFU/ml.

Microbial community developed over submerged substrate in ponds provides high quality natural food, as well as improving the water quality through nitrification (Ramesh *et al.*, 1999; Thompson *et al.*, 2002; Anand *et al.*, 2013).

CONCLUSION

It could concluded from this study that the growth of algae could be promoted by the application of rice straw as mats in fishponds, which acts as additional food surfaces for fish. The examined application does not negatively altered the investigated water quality parameters, where all examined water characteristics and microbial communities that recorded alongside the study period were within the acceptable limits for fish culture.

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تأثير إستخدام قش الأرز على الخواص الليمنولوجية والمجتمعات البكتيرية فى أحواض الأسماك الترابية

ضياء عبدالرحيم القناوي[١]، تغريد أحمد محمد [٢]

[١] قسم الليمنولوجي– المعمل المركزي لبحوث الثروة السمكية– مركز البحوث الزراعية– مصر.

[٢] قسم أمراض الأسماك– المعمل المركزي لبحوث الثروة السمكية– مركز البحوث الزراعية– مصر.

الملخـــــــص العربـــــــي

أجريت هذه الدراسة بأحواض المركز الدولي للأسماك بالعباسة خلال الفترة من أول شهر يونيو إلي نهاية شهر نوفمبر من عام ٢٠١٧، لإختبار تأثير إستخدام قش الأرز كركائز لتنمية الهائمات الملتصقة في أحواض الأسماك الترابية المسمدة علي بعض خصائص جودة المياة بها وكذلك المحتوي البكتيري بالمياه والأسماك. تم تسميد جميع الأحواض تسميدا عضويا بإستخدام زرق الدواجن بمعدل ١٥ كجم/حوض/أسبوعيا.

تم إستخدام ستة أحواض ترابية مساحة كل منها ١٠٠٠ م^{٢} حيث قسمت إلي مجموعتان كل منها ذات ثلاث تكرارات. تركت أحواض المجموعة الأولي بدون إضافة القش لتعمل كمجموعة ضابطة للمقارنة، بينما تم إضافة قش الأرز الجاف بمعدل ٤٥ كجم/حوض إلي ثلاث أحواض تمثل المعاملة الثانية لتعمل كركائز توفر سطح جاذب لتنمية الهائمات الملتصقة من الطحالب.

تم قياس بعض عوامل جودة المياة وهي: درجة حرارة المياه، الأكسجين الذائب، تركيز أيون الهيدروجين (pH)، العسر الكلي، تركيز أيون الكالسيوم و أيون الماغنيسوم، الأملاح الكلية الذائبة، النيتروجين الكلي، والفوسفور الكلي، وذلك بصفة أسبوعية لبعض العناصرونصف شهرية لبقية العناصر في مياه مصدر الري وفي أحواض كل من المجموعة الضابطة وفي أحواض المعاملة بقش الأرز. أيضا تم قياس تعدادات المجتمعات البكتيرية في مياه الأحواض والأسماك من كلا المعاملتين. أوضحت النتائج المتحصل عليها في نهاية الدراسة عدم وجود فروق ذات دلالة إحصائية في متوسطات درجة الحرارة بين مياه المصدر عنها في أحواض كلا المعاملتين. بينت النتائج أيضا أن متوسطات قيم كل من الأكسجين المذاب وتركيز أيون الهيدروجين والأملاح الكلية الذائبة والنيتروجين الكلي والفوسفور الكلي في مياه المصدر كانت أقل منها في مياه الأحواض، بصورة ذات دلالة إحصائية، بينما كانت متوسطات قيم كل من العسر الكلي وأيون الكالسيوم في مياه المصدر أعلى من متوسطات تلك القيم في مياه أحواض التجربة، بصورة ذات دلالة إحصائية. على كل الأحوال، كانت كل قيم خصائص جودة المياه محل الدراسة في الحدود المسموح بها والموصي بها من الهيئات الدولية ذات

الاختصاص. أوضحت النتائج أيضا أن وجود قش الأرز بمياه الأحواض له تأثير مباشر في تقليل الأعداد الكلية للتجمعات البكتيرية في مياه الحوض وكذلك في جسم الأسماك.

بناءا علي النتائج المتحصل عليها في نهاية فترة الدراسة، يمكن التوصية بإمكانية إستخدام قش الأرز في أحواض الأسماك الترابية المسمدة بالكميات المستخدمة في الدراسة، لتنمية الهائمات الملتصقة، والتي تعمل كمصدر إضافي لتغذية الأسماك' دون أن يؤثر ذلك علي خصائص جودة المياه بتلك الأحواض.