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Efficiency of Toxic Substance Removal from Aquaculture Wastewater by Duckweed (*Lemna minor*) and Bacteria (*Bacillus sp.*)

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Abstract

Bioremediation, an approach to reduce hazardous substances in wastewater effluents, was used in the treatment of wastewater collected from African catfish (*Clarias gariepinus*) fish farm in active production in this study. The experiment was designed to evaluate the effectiveness of duckweed, *Lemna minor* (Td) and the bacteria, *Bacillus sp* (Tb) while the negative and positive control treatments were untreated wastewater and fresh well water used in production by sampled fish farm respectively. Fifteen (15) litres of the wastewater was bioremediated ex-situ with duckweeds of an average wet weight of 49.53±0.25g and 15ml of the bacterial inoculums. After 2 weeks of bioremediation, biomass of duckweed increased by 97.51%. There was a significant reduction (P<0.05) in level of phosphate, mg/L; sulphate, mg/L; nitrate, mg/L; ammonia, mg/L; biological oxygen demand, BOD, mg/L; chemical oxygen demand, COD, mg/L; and total soluble solids TSS, mg/L by duckweed, with reduction efficiencies of 70.70%, 90.96%, 40.18%, 77.78%, 96.66%, 96.26% and 36.94%, respectively, compared to the positive control (initial) wastewater (P<0.05). Reduction efficiency (RE) was lower for *Bacilli sp* and negative control compared to duckweed (P<0.05). The pH level in the wastewater increased in all treatments, with the highest values recorded for duckweed. However, the highest dissolved oxygen level (DO) of 4.81±0.03mg/l was recorded in the bacteria treatment, followed by duckweed (4.45±0.06mg/l) and 4.09±0.06mg/l for the untreated wastewater (negative control). The results of this study indicated that *Lemna minor* was more efficient in removal of toxic substances in commercial aquaculture wastewater than *Bacillus sp.* for sustainable aquaculture practice.

Keywords: Aquaculture, Bioremediation, *Bacillus Sp*., Duckweed, Wastewater.

Introduction

The steady growth in the aquaculture sector globally rightly placed it as one of the fastest growing food sector, increasing from 49.9 million tonnes in 2007 to 66.6 million tonnes in 2012 (FAO, 2014). The intensification of aquaculture production will continue to depend on the use of fish meal and plants protein sources, mainly from soya bean (Olsen and Hasan, 2011), which both release large amount undigested phosphate and nitrogen into the aquatic environments (Satoh *et* *al*., 2003; Nwanna *et al*., 2008). Nitrogen and phosphorous constitute the main nutrients in effluents from intensive aquaculture (Haghbayan and Mehrgan, 2015). Excessive phosphate in aquaculture effluent leads to rapid spread of cyanobacteria bloom, resulting in eutrophication in aquatic environments and consequent change in the structure of biodiversity (Kumar *et al*., 2011; Nwanna and Olusola, 2014). Nitrogenous wastes in the form of ammonia, a limiting water quality parameter in intensive aquaculture, are highly toxic to macro-fauna in the open water body (Lazzari and Baldisserotto, 2008). Stephen and Farris (2004) reported that elevated ammonia concentrations could lead to blood ammonia intoxication or autointoxication in fish. Intensification of aquaculture could also result in large amount of waste, including suspended solids, which leads to deoxygenation, clogging of interstitial space, and loss of aquatic biota (Magni *et al*., 2008).

Reducing the outputs of these dissolved wastes is considered to be a key element for the long-term sustainability of aquaculture (Hasan, 2001). Although phytase is used to reduce phytate phosphate levels in plants, their high cost reduce their use in field application (Cao *et al*., 2007). Hence, there is the need to intensify the use of cheap, cost effective and available, traditional techniques in reducing waste produced from aquaculture.

The treatment of hazardous wastes using biological processes renders harmless various contaminants and restores the quality of aquatic environments (Gupta *et al*., 2012; Divya *et al*., 2015; Ugya, 2015). It also provides an alternative for effective management of wastewater for the purpose of reuse thereby reducing pressure on limited freshwater resources. It is a process that uses biological agents, such as yeast, fungi or bacteria to clean up contaminated soil and water. This includes use of important species like *Acromobacter, Alcaligenes, Arthrobacter, Bacillus, Cinetobacter, Corneybacterium, Flavobacterium, Micrococcus, Mycobacterium, Nocardia, Pseudomonas, Vibrio, Rhodococcus* and *Sphingomonas* species (Divya *et al*., 2015). Commercially prepared mixtures of *Bacillus* species mixed into the rearing water are reported to increase fish growth (Queiroz and Boyd, 1998).

However, in addition to time, effort and technical knowhow in isolating specific microorganisms for reducing the organic loading of contaminated sites, microorganisms are not normally present in the required quantities in the water column (Divya *et al*., 2015).

Duckweed *(Lemna minor)* has been used in several studies to absorb nutrients from wastewater arising from aquaculture (Ansal *et al.,* 2010). They are cheap and available, with the potential to permanently remove nutrients after harvest, and hence, reduce nutrient loading of aquatic environment (Gupta and Prakash, 2014). The use of duckweed for treatment and management of wastewater makes them feasible for developing countries in hot climates to provide low-cost treatment of domestic sewage particularly in rural areas (Smith and Moelyowati, 2001).

The study was therefore designed to evaluate the effectiveness of duckweed and bacteria in reducing nutrient load and improve water quality in aquaculture wastewater from catfish farming.

Materials and Methods

Experimental protocol

The experiment was designed to evaluate the effectiveness of duckweed, *Lemna minor* (Td) and bacteria, *Bacillus sp.* in reducing nutrient load and improving water quality of wastewater from African catfish *(Clarias gariepinus)* farming. The wastewater samples were collected from an African catfish farm (N7^{o}35`38.69``, E3$^{o}$85`42.79``) in active production in Ibadan, Nigeria at the point of discharge between 6.30-7.00 a.m. in 25 litres plastic containers. Fifteen (15) litres of the wastewater collected was bioremediated ex-situ for 2 weeks with duckweeds of an average wet weight of 49.53±0.25g and 15ml of the bacterial inoculums. The negative and positive control treatments were untreated wastewater and fresh well water used in production by sampled fish farm respectively. All the treatments in this study were in triplicates. The bioremediation experiment was carried out in the wet laboratory of the Department of Aquaculture and Fisheries Management, University of Ibadan, Nigeria.

Bacteria isolation, culture and identification

The wastewater samples in which the bacteria strain used for the bioremediation experiment were isolated from, were collected using 10ml sterile sampling bottles from the sampled fish farm. One milliliter aliquot of the wastewater serially diluted with 9ml of sterilized distilled water at 10^{-9} and 10^{-10} dilution was pipette into well-labelled sterile petri dish. Nutrient Agar (NA, Oxoid CM3) prepared aseptically according to the instructions of the manufacturer was poured aseptically on the aliquot in the petri dish and incubated for between 18-24 hours at 37°C (Adedeji and Onwenfah, 2013). Distinct bacteria colonies on the incubated plate were sub-cultured on sterile NA plate to obtain pure colonies of the candidate bacteria strain *(Bacillus sp.)*. The bacterial isolates were identified using the morphological, microscopic and biochemical characteristics after 24hours of incubation (Baron and Murray, 1999). The candidate organism was confirmed according to morphological and biochemical description of Bacillus sp. in Berge's Manual of Bacteriology (Breed *et al.,* 1957).

Preparation of the Bacteria Inoculums

Twenty-four hours pure colonies of the confirmed bacteria isolate *(Bacillus sp.)* were transferred aseptically to nutrient broth. The concentration of the bacterial cells in the broth was adjusted to 10^5 colony-forming unit using sterile physiological saline to correspond to 0.5 MacFarland standards. The standard plate count method of bacteria enumeration as described by (Horsely, 1977, APHA, 1995) was used to determine the colony forming unit (CFU) in the inoculums.

Microbial inoculation of wastewater

Fifteen milliliters of the bacterial *(Bacillus sp)* inoculums was introduced into 15litres of wastewater in the experimental aquarium of dimension 0.390m x 0.276m x 0.260m using a sterile needle and syringe. The treatment was repeated in triplicates. The treated wastewaters were screened with mosquito net and thereafter arranged on experimental rack in the laboratory.

Collection of duckweeds and phytoremediation

Fresh, green and lush duckweeds plants *(Lemna minor)* were harvested along with the pond water from the University of Ibadan Botanical Garden using scoop hand-net in clean plastic containers. The plants were collected in the morning between 6:00-7:00 am and were conditioned in plastic bowls with clean well water for three weeks, during which they were exposed to sunlight. Duckweeds of an average weight of 49.53±0.25g (mean wet weight) were assigned to triplicate group of tanks (0.39 m × 0.28 m × 0.26m) containing the wastewater such that the entire surface of the water was covered by a single layer of fronds to prevent direct solar radiation from reaching the water surface (Al-Nozaily, 2001). This was done to prevent proliferation of algae in the bioremediation tanks. The tanks were kept outside the laboratory for direct exposure to sunlight.

Estimation of Waste Removal Efficiency

The waste removal efficiency (RE %) after the bioremediation experiment was estimated according to Umran and Sadettin (2015) procedure using the following equation:

$$\frac{C\text{-}Co}{Co} \times 100 \quad \text{..................................} \quad \text{Eq. 1}$$

Where: *Co* and *C* are respectively, the concentrations of wastewater parameters before and after bioremediation.

Statistical analysis

Significant differences between treatments water quality data were analyzed using ANOVA. Comparison and separation of treatment means were done using Duncan multiple range test (Duncan, 1855) and were declared significant at P <0.05. Correlation and multiple regression analysis were used to establish relationship between variables measured using SPSS software (version 20).

Results

Bacteria Count and Identification

The average number of bacteria counted in the inoculums used in the study was 1.81×10^{11} cfu/L. The microorganism was positive in the Gram staining test and appeared rod-like in shape, when viewed under microscope. In colony morphology test, the microbe colonies were large with undulating circular margins. Bubbles were produced when the microbes were exposed to hydrogen peroxide, indicating the production of oxygen and water due to production of the enzyme catalase. The fermentation of fructose by *Bacillus sp.* was complete. Maltose showed partial fermentation. Other sugars were not fermented as shown in Table 1.

Table 1. Fermentation of different sugars by the microbe used in the experiment

Sugars	Result
Fructose	+
Lactose	–
Mannitol	–
Sucrose	–
Galactose	–
Glucose	–
Maltose	±

+ indicates occurrence of fermentation
– Absence of fermentation
± Partial fermentation

Water Quality Parameters and Nutrient Removal Efficiency

The results of this study indicated that there was an increase in the biomass of duckweed (*Lemna minor*) by 97.51% from an initial average weight of 49.53±0.25g after two weeks of use in bioremediating aquaculture effluent. Presented in Table 2 is the mean concentration of water quality parameters in wastewater samples collected from the sample African catfish farm in Ibadan metropolis, Nigeria. The results of this study indicated that there was significant difference ($P<0.05$) in the mean concentrations of the water quality parameters in the untreated wastewater (Negative control) and wastewater bioremediated with duckweed (*Lemna minor*) and bacteria (*Bacillus sp.*) after two weeks of bioremediation as indicated in Table 2. The least mean concentration of phosphate, sulphate, TAN, DO, pH and TSS of 0.38±0.05mg/L, 0.33±0.06mgL, 1.28±0.05mg/L, 0.06±0.03mg/L, 3.60±0.30mg/L, 6.66±0.18 and 23.39±0.88mg/L respectively were recorded in the untreated well water, water source in the sampled farm (Positive control) after two weeks. Meanwhile, the negative control had the highest mean concentration of phosphate, BOD and

Table 2. Mean concentrations of water quality parameters of wastewater from sampled African catfish farm in Ibadan metropolis, Nigeria and the bioremediated wastewater after two weeks of bioremediation with duckweed (*Lemna minor*) and bacteria (*Bacillus sp.*).

Water Quality Parameters	Raw wastewater	Treatments			
		Positive Control	Negative Control	Bacteria	Duckweed
Phosphate (mg/L)	18.43±0.78	0.38±0.05^{c}	16.57±0.20^{a}	16.47±0.03^{a}	5.40±0.27^{b}
Sulphate (mg/L)	5.53±0.33	0.33±0.06^{d}	3.77±0.11^{b}	4.17±0.08^{a}	0.53±0.03^{c}
Nitrate (mg/L)	9.93±0.36	1.28±0.05^{d}	6.48±0.02^{b}	6.77±0.03^{a}	5.61±0.25^{c}
Ammonia (mg/L)	1.17±0.48	0.06±0.03^{c}	0.15±0.08^{bc}	0.66±0.24^{a}	0.26±0.09^{b}
DO (mg/L)	4.01±0.14	3.60±0.30^{d}	4.09±0.05^{c}	4.81±0.02^{a}	4.45±0.05^{b}
BOD (mg/L)	36.80±1.89	3.52±0.39^{c}	31.90±0.18^{a}	27.27±0.08^{b}	1.23±0.03^{d}
COD (mg/L)	58.81±2.60	32.13±1.75^{c}	58.17±0.17^{a}	52.11±0.26^{b}	2.20±0.05^{d}
pH	6.83±0.66	6.66±0.18^{d}	7.46±0.03^{c}	7.86±0.07^{b}	8.11±0.18^{a}
Temperature (^{o}C)	24.63±0.22	27.71±0.15^{b}	24.38±0.03^{c}	24.26±0.07^{d}	27.93±0.18^{a}
TSS (mg/L)	2136.75±332.37	23.39±0.88^{d}	2015.67±1.71^{b}	2034.00±2.65^{a}	1347.33±1.25^{c}

Note: Mean values on the same row with different alphabet superscripts are significantly different at $\alpha_{0.05}$.

COD of 16.57±0.20mg/L, 3.52±0.39mg/L and 58.17±0.17mg/L respectively. The water quality parameters of *Lemna minor* treated wastewater samples are shown in Table 2. There were significant reduction ($P<0.05$) in concentration of BOD, ammonia, phosphate, COD, nitrate and total *suspended* solids (TSS) in duckweed treated wastewater compared to the bacteria and control treatments.

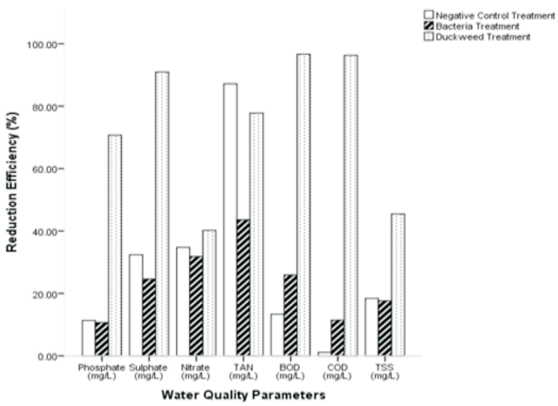

Figure 1. Reduction efficiency of phosphate, sulphate, nitrate and ammonia level in aquaculture wastewater bioremediated with *Lemna minor* and *Bacillus sp.* after two weeks.

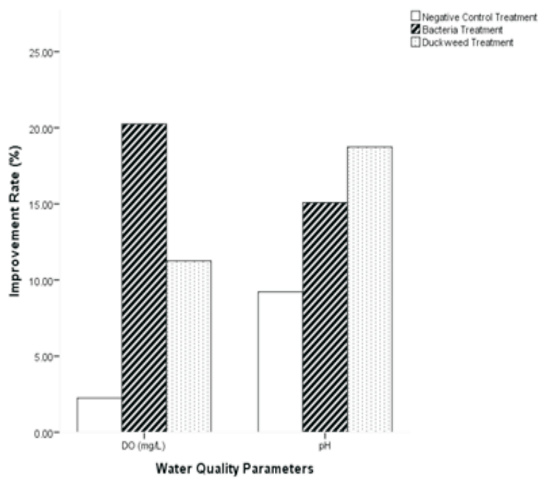

Figure 2. Improvement rate of DO and pH level in aquaculture wastewater bioremediated for two weeks with *Lemna minor* and *Bacillus sp.*

Comparing the water quality parameters of the aquaculture wastewater bioremediated with *Lemna minor* and *Bacillus sp.* for two week, the lowest average concentration of phosphate, sulphate, nitrate, ammonia, BOD, COD, and TSS of 5.40±0.31mg/l, 0.50±0.06mg/l, 5.94±0.47mg/l, 0.15±0.14mg/l, 1.17±0.09mg/l, 2.03±0.19mg/land 1344.42±12.35mg/l, respectively were recorded in the duckweed treated wastewater with reduction efficiency of 70.70%, 90.96%, 40.18%, 77.78% , 96.66%, 96.26% and 36.94% respectively (Figure 1). However, the lowest TAN reduction efficiency of 87.18% was observed in the untreated wastewater after two weeks. The DO and pH level in the wastewater tends to increase across all the treatments after two weeks of bioremediation (Figure 2). Wastewater bioremediated with *Bacillus sp.* had the highest DO value of 4.81±0.02 mg/L with improvement rate of 20.25% followed by 4.45±0.06mg/l at the rate of 11.25% observed in the duckweed treated wastewater. In contrast, duckweed bioremediated wastewater recorded the highest mean pH level of 8.11±0.18 with improvement rate of 18.74% while the negative control had the lowest improvement rate of 9.22%.

Relationship between Water Quality Parameters

The correlation matrix (Table 3) indicated there was a strong positive linear relationship between the physicochemical parameters of the water samples while water temperature had strong negative influence on all the water parameters measured except for DO and pH level on which weak negative relationship was recorded. The multiple regression analysis was used to develop predictive model indicating the relationship between the dependent variables (phosphate, nitrate, sulphate and TAN) and the independent variables (DO, pH and temperature) as presented in Table 4. The model predicting phosphate and sulphate had the highest regression determinant (R^2) values of 93.30% and 90.70% respectively while Nitrate and TAN models had R^2 values of 59.80% and 28.60% at significant level of $P<0.05$. The coefficient (B) value indicated that DO had positive influence on sulphate, nitrate, TAN except phosphate while temperature had negative influence on all the dependent variables. Furthermore, pH had positive influence on phosphate and nitrate while negative relationship with sulphate and TAN.

Table 3. Correlation Matrix Indicating Linear Relationship between Water Quality Parameters of Water Samples Collected

Parameters		Phosphate	Nitrate	Sulphate	BOD	COD	DO	pH	Temp	TAN	TSS
Phosphate	R	1.000									
	Sig.										
Nitrate	R	0.871**	1.000								
	Sig.	0.000									
Sulphate	R	0.964**	0.816**	1.000							
	Sig.	0.000	0.000								
BOD	R	0.949**	0.739**	0.976**	1.000						
	Sig.	0.000	0.000	0.000							
COD	R	0.750**	0.401*	0.843**	0.906**	1.000					
	Sig.	0.000	0.010	0.000	0.000						
DO	R	0.483**	0.544**	0.357*	0.235	-0.029	1.000				
	Sig.	0.002	0.000	0.024	0.144	0.860					
pH	R	0.255	0.404**	0.045	-0.025	-.323*	.765**	1.000			
	Sig.	0.113	0.010	0.782	0.877	0.042	0.000				
Temp	R	-0.948**	-0.686**	-0.951**	-0.968**	-0.890**	-0.374*	-0.076	1.000		
	Sig.	0.000	0.000	0.000	0.000	0.000	0.017	0.641			
TAN	R	0.551**	0.694**	0.635**	0.515**	0.328*	0.375*	0.108	-0.458**	1.000	
	Sig.	0.000	0.000	0.000	0.001	0.039	0.017	0.509	0.003		
TSS	R	0.938**	0.930**	0.834**	0.789**	0.484**	0.642**	0.490**	-0.802**	0.495**	1.000
	Sig.	0.000	0.000	0.000	0.000	0.002	0.000	0.001	0.000	0.001	

** Correlation is significant at the 0.01 level (2-tailed). * Correlation is significant at the 0.05 level (2-tailed).

Table 4. Multiple Regression Models Indicating Relationship between the Dependent Variables (Phosphate, Nitrate, Sulphate and Tan) And The Independent Variables (Do, Ph and Temperature)

Dependent Variables	Independent Variable, Coefficient (B)				R^2	Sig.-value
	Constant	DO	pH	Temp		
Phosphate	96.087	-0.379	2.407	-3.929	0.933	0.000
Sulphate	30.733	0.294	-0.254	-1.063	0.907	0.000
Nitrate	18.672	0.473	1.245	-0.942	0.598	0.000
TAN	1.759	0.406	-0.180	-0.068	0.286	0.006

Discussion

The results indicated that duckweed had the highest nutrient removal efficiency of phosphate, sulphate, nitrate and ammonia (figure 1). The high affinity for nutrient uptake in aquaculture wastewater by duckweed was an indication that nutrients uptake improved biomass production of duckweed and reduced nutrient level and improved DO concentration. Therefore, duckweed has potentials to be an important bioremediation tool in reducing the nutrient content in aquaculture wastewater before discharge into open environment. Compared to the present study, a higher removal efficiency (RE) for TSS (96.3% vs 36.94%), ammonia (82.0% vs 77.78%), nitrate (100% vs 40.18%), but lower values BOD (90.6% vs 96.66%), COD (89.0% vs 96.26%) and phosphate (64.4% vs 70.70%) was reported for duckweed, *Lemna gibba L.* by Abou el- Kheir *et al*., (2007), cited in Chaudhary and Sharma, 2014). The difference between these findings and the report by the authors may be due to species difference and the high absorptive capacity of *Lemna minor* for phosphate, BOD, and COD, hazardous substances that limit water quality in aquaculture and affect fish survival (Akpoilih *et al*., 2015). The reduction and increase in RE of *Lemna minor* for nitrate (40.18%) and phosphate (70.70%), respectively was also supported with a study (Korner and Vermaat, 1998, cited in Gupta and Prakash, 2014) which showed a range of RE for nitrate (73%–97%) and phosphate (63%–99%). Alaerts *et al.* (1996) reported RE of 74%, 77%, and 96% for nitrogen, phosphate, and BOD, respectively in duckweed (*Lemna Gibba L*). The 77% reduction of phosphate reported by the authors is higher than the reported value of 70.70% in the present study (figure 1). This may be due to species differences as mentioned above. However, this value is within the range reported by Körmer and Vermaat (1998). The RE of BOD reported by the authors (Alaerts *et al.*, 1996) is, however, in line with the RE (96.6%) recorded for *Lemna minor*. The high phosphate content and efficiency of its removal in duckweeds makes them suitable for phosphate removal, which significantly reduces the level of eutrophication in aquatic systems (Gupta and Prakash, 2014). High removal efficiency of COD and BOD of the wastewater by duckweed plant after the bioremediation experiment (figure 2) was in agreement with the observation of Chaudhary and Sharma (2014) and Ugya (2015). This suggests the high RE of *Lemna minor* for organic compounds, as well as the decomposition of organic materials by microbes (Zimmon *et al.,* 2005). The low RE for TSS (36.94%) recorded in this study compared to values (60-90%) reported by Ugya (2015) may be due to location and depth of phytoremediation (Gupta and Prakash, 2014). The low RE of TSS could also be as a result of high level of sludge, which could increase the level of total suspended solids in the wastewater (Gupta and Prakash, 2014). The removal of sludge has been reported to enhance the reduction of organic matter in duckweed treatment systems (Iqbal 1999; Smith and Moelyowati 2001, cited in Gupta and Prakash, 2014). Thus, the low RE of TSS (36.94%) may lower RE of ammonia (77.78%), when compared with RE (82.0% and 99%) of duckweed, *Lemna gibba* (Chaudhary and Sharma, 2014; Gupta and Prakash, 2014).

It has been reported that the efficiency of duckweed treatment systems depend on the secondary and tertiary treatment that converts sludge into forms that are available for absorption by duckweeds (Caicedo *et al*. 2000; Smith and Moelyowati 2001; Dalu and Ndamba, 2003, cited Gupta and Prakash, 2014). However, the RE of TSS is higher than values obtained in wastewater treated with *Bacillus sp* and control (figure 2).

Compared to initial wastewater*, Bacillus sp.* exhibited a high efficiency in the removal of ammonia (43.59%) and nitrate (31.82%), and sulphate (24.59%). The result is in line with the study of Bhutto and Dahot (2010) who reported that some *Bacillus sp.* utilized nitrogen from ammonium nitrate, ammonium sulphate, among other sources, in the production of an enzyme called amylase that is of industrial importance. There was also reduction in the sulphate and nitrate level of untreated wastewater after two weeks. This could be as a result of biochemical activities of the indigenous microbes within the wastewater which tend to use up the pollutants in the wastewater. The relatively higher DO in *Bacillus sp* compared to *Lemna minor* (4.81±0.02 mg/L vs 4.45±0.06mg/L, figure 4) can be explained by the fact that while duckweeds inhibit the transfer of oxygen into the water column by forming a mat over the surface, the ease of oxygen circulation in the water may facilitate the bacteria oxidation of organic material (Gupta and Prakash, 2014). The reduction in pH as observed in this study for *Bacillus sp,* when compared with *Lemna minor* (7.86±0.08 vs 8.11±0.21, figure 3), favours microbial degradation of organic substances (Gupta *et al*., 2012). The temperature range measured in the phytoremediation study (27.96±0.26^0 C) is within the optimum range of 10^0 C -34^0 C for the growth of duckweed (Culley *et al.*, 1981). They can survive in outdoor wastewater treatment tanks as reported by Classen *et al.* (2000).

It was observed in the study that there was pronounced differences in the efficiency of removal of pollutants from aquaculture wastewater by *Bacillus sp.* and *Lemna minor*. Duckweed plant was more effective in bioremediation of aquaculture wastewater than *Bacillus sp.* This could be as a result of combined effects of plant uptake and bacteria (endophytic and rhizospheric bacteria) associated with duckweed in phytoremediation process compared with the microbial remediation where only bacteria are involved in the bioremediation of the wastewater (El-Kheir *et al.*, 2007; Farrell, 2012; Gupta and Prakash, 2014). The presence of bacteria enhances the efficient phytoremediation activity, and reduce need for additional fertilization (Afzal *et al.,* 2014). The efficiency of phytoremediation can be enhanced if sufficient amount of sludge, suspended solids and turbidity are reduced mechanically or in combination with other cheap and affordable treatment measures.

Conclusion

This research has demonstrated that *Lemna minor* is an effective biological treatment approach to manage effluents discharged from aquaculture farming, and was more efficient than bacteria treatment using *Bacillus sp*. However, given the fact that this was only a laboratory experiment and the short term duration of the study, field application of the use of this plant in association with other efficient microbes should be carried out.

References

Adedeji, O.B. and Onwenefah, M. (2013). The Antibiotic Resistant Patterns Of Bacterial Flora Of Cultured Catfish Fed With Poultry Hatchery Waste From Selected Farms In Ibadan, Nigeria. *Researcher* 5(9):37-43.

Afzal, M., Khan, Q.M and Sessitsch, A (2014). Endophytic bacteria: Prospects and applications for the phytoremediation of organic pollutants. *Chemosphere.* 117: 232-42.

Akpoilih B.U., Ajani E.K. and Omitoyin B.O (2015). Dietary Phytase Improves Growth and Water Quality Parameters for Juvenile *Clarias gariepinus* Fed Soyabean Meal-based Diets. *International Journal of Aquaculture* 5: 1-20.

Alaerts, G.J., Mahbubar, M.D.R. and Kelderman, P. (1996). Performance analysis of a full-scale duckweed-covered sewage lagoon. *Water Res.* 30(4): 843-852.

Al-Nozaily, F.A. (2001). Performance and process analysis of duckweed-covered sewage lagoons for high strength sewage. Doctoral dissertation. Delft University of technology. International Institute of Hydraulic and Environmental Engineering. Delft-Holland. A. A. Balkema, Rotterdam, Netherlands. Pp. 203. Retrieved on 2nd of June, 2015 from http://repository.tudelft.nl/view/ihe/uuid%3Afa6df21a-6d0e-4815-93d0-17ff2683098b/ .

Ansal, M.D., Dhawan, A and Kaur V. I. (2010). Duckweed based bio-remediation of village ponds: An ecologically and economically viable integrated approach for rural development through aquaculture. *Livestock Research for Rural Development*. 22(7): 22-27.

APHA (1995). Standard Methods for the examination of Water and Wastewater. 19th ed. American Public Health Association.

Baron, E. J. and Murray, P. R. (1999). *Bacteriology.* In: Manual of clinical microbiology 7th Edition P. R. Murray, E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Yolken (eds.). ASM Press, Washington, D.C., pp. 246–831.

Breed, R. S., Murray, E. G.D., and Smith, N. R. (1957). Bergey's Manual of Determinative Bacteriology 7th ed. The Williams & Wilkins Company, Baltimore, USA. Pp. 613-633.

Bhutto M.A and Dahot M.U (2010). Effect of Alternative Carbon and Nitrogen Sources on Production of Alpha-amylase by *Bacillus megaterium*. *World Applied Sciences Journal.* 8: 85-90.

Caicedo, J.R., Van der Steen, N.P., Arce, O and Gijzen, H.J (2000). "Effect of total ammonian nitrogen concentration and pH on growth rates of duckweed (*Spirodela polyrrhiza*)." *Water Research.* 34: 3829–3835.

Chaudhary, E. and Sharma, P (2014).Use of Duckweed in Wastewater Treatment. *International Journal of Innovative Research in Science, Engineering and Technology* 3(6): 13622-13624.

Classen, J. J., Cheng, J., Bergmann, B. A. and Stomp, A. M. (2000). *Lemna gibba* growth and nutrient uptake in response to different nutrient levels. In Animal, Agriculture and Food Processing Waste: Proceedings of 8th International Symposium of American Society of Agricultural Engineers (ASAE), October 9-11, 2000, Des Moines, Iowa. St. Joseph, Michigan. Pp. 40-45.

Culley, D.D., Rejmankova, E., Kvet J. and Frey, J.B (1981). Production, chemical quality and use of duckweeds (Lemnaceae) in aquaculture, waste management and animal feeds. *J. World Maric. Soc.* 12: 27-49.

Dalu, J.M. and Ndamba, J. (2003). Duckweed based wastewater stabilization ponds for wastewater treatment (a low cost technology for small urban areas in Zimbabwe)." Physics and Chemistry of the Earth, Parts A/B/C 28: 1147–1160.

Divya, M., Aanand, S., Srinivasan, A and Ahilan, B (2015). Bioremediation – An eco-friendly tool for effluent treatment: A Review. *International Journal of Applied Research*. 1(12): 530-537.

Duncan, D.B. (1955). New Multiple Range and Multiple Range F-Tests. *Biometrics*. 11:1-42

El-Kheir W.A., Ismail G, El-Nour, A, Tawfik T and Hammad D (2007). Assessment of the efficiency of duckweed (Lemna gibba) in wastewater treatment. *International Journal of Agriculture and Biology* 5: 681-689.

Farrell, J. B. (2012). Duckweed Uptake of Phosphate and Five Pharmaceuticals: Microcosm and Wastewater Lagoon Studies". *All Graduate Theses and Dissertations.* 1212: 57-120.

Food and Agriculture Organization (FAO) (2014). State of the world fisheries. FAO, Rome. 223pp.

Gupta, C. and Prakash, D. (2014). Duckweed: an effective tool for phyto-remediation, *Toxicological and Environmental Chemistry*. DOI: 10.1080/02772248.2013.879309.

Gupta, P., Roy, S. and Mahindrakar, A.B (2012). Treatment of Water Using Water Hyacinth, Water Lettuce and Vetiver Grass - A Review. *Resources and Environment.* 2(5): 202-215.

Haghbayan, S. and Mehrgan, M.S. (2015). The Effect of Replacing Fish Meal in the Diet with Enzyme-Treated Soybean Meal (HP310) on Growth and Body Composition of Rainbow Trout Fry. *Molecules.* 20: 21058–21066.

Iqbal, S. (1999). "Duckweed Aquaculture: Potentials, Possibilities and Limitations for Combined Waste Water Treatment and Animal Feed Production in Developing Countries." SUNDEC Report no. 6/99. Switzerland.

Horsley, R.W. (1977). A review of bacterial Flora of teleosts and elasmobranches including methods for its analysis. *Journal of Fish Biology* 10: 529-533.

Koermer, S. and Vermaat, I.E. (1998). The relative importance of *Lemna gibba* L., bacteria and algae for nitrogen and phosphorous removal in duckweed-covered domestic wastewater. *Water Res*. 33(12): 3651-3661.

Kumar, V., Sinha, A.K., Makkar, H.P.S., De Boeck, G and Becker K (2011). Phytate and phytase in fish nutrition. *J. Anim. Physiol. Anim. Nutr*. 96:335–64.

Magni, P., Rajagapal, S., Vandervelde, G., Perel, G., Kasserberg, J., Vizzini, S., Mazerla, A and Giordanb, G (2008). Sediment features, macroziobathic assemblages and trophic relationship following a dystrophic event with anoxia and sulphide development in the Santa Giuta Lagoon. *Marine Pollution Bulletin*. 57:125 – 136.

Nwanna, L.C., and Olusola S.E., (2014). Effect of supplemental phytase on phosphate digestibility and mineral composition of Nile tilapia (*Oreochromis niloticus*). *International Journal of Aquaculture*. 4 (15): 89-95.

Nwanna, L.C., Kolahsa, M., Eisenreich, R and Schwarz, F.J (2008). Pre-treatment of dietary plant feedstuffs with phytase and its effect on growth and mineral concentration in common carp (*Cyprinus carpio L.*). *Journal of Animal Physiology and Animal Nutrition*. 92: 677–682.

Olsen, R.L. and Hasan, M.R. (2011). A limited supply of fishmeal: Impact on future increases in global aquaculture production. *Trends in Food Science and Technology* 27: 120-128.

Queiroz, J. F. and Boyd, C. E. (1998) Effects of bacterial inoculum in channel catfish ponds. *Journal World Aquaculture Society* 29: 67-73.

Satoh, S., Hernandez, A., Tokoro, T., Morishita, Y., Kiron, V and Watanabe, T (2003). Comparison of phosphate retention efficiency between rainbow trout (*Oncorhynchus mykiss*) fed a commercial diet and a low fish meal based diet. *Aquaculture*. 224: 271-282

Smith, M.D and Moelyowati, I. (2001). Duckweed based wastewater treatment (DWWT). Design guidelines for hot climates. *Water Sci. Technol*. 43(11): 291-299.

Stephen, W.W. and Farris, J.L. (2004). In stream community assessment of aquaculture effluents. *Aquaculture* 231:148-162.

Ugya, Y.A (2015). The Efficiency of *Lemna minor* L. in the Phytoremediation of Romi Stream: A Case Study of Kaduna Refinery and Petrochemical Company Polluted Stream. *J. App. Biol Biotech* 3(1): 011-014.

Umran, T.U. and Sadettin, E.O. (2015). Removal of Heavy Metals (Cd, Cu, Ni) by Electrocoagulation. *International Journal of Environmental Science and Development*. 6(6): 425 – 429.

Zimmon, O.R., Van Der Steen, N.P and Gijzen, H.J (2005). Effect of Organic Surface Load on Process Performance of Pilot Scale Algae and Duckweed Based Waste Stabilization Ponds. *J. Environ. Engr.* 131: 94- 587.

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