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EVALUATION OF POLYPROPYLENE AND PALM KERNEL SHELL AS BIOFILTER MEDIA FOR DENITRIFICATION OF FISH CULTURE WASTEWATER

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ABSTRACT

Nitrification removes ammonia and Nitrite-Nitrogen ($N0_2$-N) from aquaculture wastewater but lead to increase in the amount of Nitrate-Nitrogen (NO_3-N) in the system. NO_3-N though not toxic to fish at low concentration, affects growth and yield at higher concentration thus making further denitrification of the wastewater a necessity. This study investigated Polypropylene bio-block (PP) and Palm kernel shell (PK) as biofilter media for the denitrification of fish farming wastewater. Pairs of biofilter units comprising nitrification and denitrification columns were used. Wastewater from African catfish (Clarias gariepinus) monoculture facility was treated using PP and PK as biofilter media. Selected water quality parameters were measured in effluent wastewater and biofilters filtrates to determine the change in water quality. The denitrification efficiency was determined using Percentage NO_3-N removed (PNR) and Volumetric NO_3-N conversion rate (VNR). Data obtained were analyzed using t-test. The selected water quality parameters in the filtrates were within the range recommended for aquaculture water reuse and discharge into the environment. NO_3-N in filtrates was reduced to 45 mg/L in PP and 25 mg/L in PK in the denitrification system. The PNR recorded by the two media was not statistically significant ($p > 0.05$) though PK had higher PNR of 72.22% compared to 59.09% in PP. The difference in VNR between the two media was significantly different ($P<0.05$) with PP having higher VNR of 58775.51±7068.43 mgNO_3-N/m^3d compared with 31084.03±13478.05 mgNO_3-N by PK biofilter.

Keywords: Biofiltration, Denitrification, Effluents, Palm Kernel Shell, Polypropylene

INTRODUCTION

The aquaculture industry, though supplies food and provides livelihood to many people, it is a bane to aquatic environments, due to the heavy pollutant load that results from aquaculture wastewater. Recirculating aquaculture systems are used to rear fish at high densities, employs water treatment and conservation techniques by continuously recycling the culture water (Dauda *et al*., 2014). The remarkably high productivity and energy efficiency of the system have minimal effect on the reduction in pollutant load, this is due to the frequency of changing rearing water per day which varies from 5 to 10% (Blancheton, 2000). The vital form of water treatment in most recirculating systems includes; removal of sludge through sedimentation or mechanical filtration, removal of total ammonia (TAN) through nitrification and exchange of water (van Rijn *et al*., 1996). Biofilters are predominantly used in recirculating aquaculture systems to achieve nitrification, but over time it may lead to nitrates accumulation especially when combined with decrease in water exchange. High concentration of nitrate may be attained in recirculating systems that uses nitrifying biofilters for removal of ammonia. Unlike ammonia and nitrite, nitrate is relatively non-toxic to aquatic organisms. However, high nitrate concentrations can affect the growth and performance of commercially cultured aquatic organisms (Kamstra and van der Heul, 1998). Ajani *et al*. (2011) noted that nitrate above 250 mg/L will affect the growth of tropical fish species such as *Clarias gariepinus*. Biological denitrification has been experimented as an effective means to remove nitrates from recirculating aquaculture system waters. Most of the biofilter media used in the denitrification system are same as the one used in the nitrification medium. They are expensive materials mainly packaged from Europe (Akinwole, 2005). Palm kernel shell, a local agricultural waste product was experimented as biofilter media in the nitrification column by Akinwole and Dauda (2014) and it performed effectively. Therefore this study focused on the use of palm kernel shell, as biofilter media in denitrification column while synthetic polypropylene bio-block was used as control for comparison.

MATERIALS AND METHODS

Biofilter Units and Media

The preparation of biofilter units and placement of polypropylene bio-block were done in line with descriptions of Dauda *et al*. (2014).

The palm kernel (PK) shells are resultant waste products of palm fruit processing. The shells were sieved to remove sand particles and later put in basket in batches, thoroughly washed and soaked in water for three days to remove impurities. The PK shells were later sundried for two days and kept in water proof sacks before loading into the filter housing up to 600 mm height. The physical properties of the selected biofilter media that are considered important for pilot scale studies in line with Colt *et al*. (2006) were measured, and their values are shown in Table 1. Each unit of biofilter comprises of two biofilter columns, therefore each media has six biofilter columns for their three replicates making a total of 12 biofilter columns for the experiment.

Aquaculture Wastewater

Fish culture effluent was collected between 0800 and 0900 hours from a commercial African catfish facility in Ibadan metropolis, Oyo State, Nigeria. The farm operates monoculture system and use concrete tank as the fish holding structure. The culture condition of the holding facility during the period of the wastewater collection is shown in Table 2.

Experimental Procedure and Water Quality Analysis

The experimental procedure and water quality analysis was carried out as described by Dauda *et al*. (2014) but unlike the procedure of Dauda *et al*. (2014), the system was left for 144 hours after which the analysis was commenced.

Biofilter Media Performance Assessment

Performance of the biofilters were evaluated using degree of change in the selected water quality parameters while denitrification efficiency of the biofilters was determined using adapted formulae from that of nitrification efficiency stated by Colt *et al*. (2006).

The adapted formulae were for percentage nitrate-nitrogen removed (PNR) and volumetric nitrate-nitrogen conversion rate (VNR).

$$PNR = \frac{(NO_3\text{-}N_{in} - NO_3\text{-}N_{out}) \times 100}{NO_3\text{-}N_{in}}$$

$$VNR\ (mgNO_3\text{-}N/m^3d) = \frac{86{,}400Q\ (NO_3\text{-}N_{in} - NO_3\text{-}N_{out})}{V}$$

Where:

NO_3-N_{in} is the Nitrate-nitrogen in the nitrification filtrate

NO_3-N_{out} is the Nitrate-nitrogen in the denitrification filtrate

V is the volume (m^3) of the media,

Q is the biofilter flow (m^3/day),

86, 400 is a conversion factor from seconds to day (60x60x24).

Statistical Analysis of data

Mean and standard of the selected water quality parameters, change in water quality parameters, residence time and biofiltration rate were determined. T-test was used to determine if the difference in data values between the two media for the change in selected water quality parameters, residence time, biofilters flow, PNR and VNR were significant at $P < 0.05$. The statistical analysis was done using IBM SPSS version 20.

RESULTS AND DISCUSSION

The filtrates from the polypropylene columns and palm kernel shell columns are represented by values under PP and PK respectively. The wastewater collected from the fish rearing tank (in Table 2) is the effluent for the nitrification columns while the filtrate from the nitrification columns serves as the effluent into the denitrification columns.

Water quality parameters

Temperature was higher in PP filtrates for both Nitrification (28.67±0.29^{o}C) and denitrification column (28.83±0.28) compared to 28.83±0.28^{o}C recorded in the two columns for PK filtrates (Table 3), but the different was not significant ($P > 0.05$). The change in temperature between the influent and the effluent for the PP filtrates were negative in both nitrification column (-0.60 %) and denitrification column (-0.59%) while PK filtrates had a positive value (0.60%) in the nitrification column and no marked change (0.00%) in the denitrification column (table 4). The difference in change in temperature between PP and PK filtrates was significant ($P < 0.05$) in both nitrification and denitrification column. The temperature in the columns for the two media was suitable for warm water fish culture as it is within the recommended level of 20 – 30^{o}C (Ajani *et al*., 2011) and also safe for discharge into the environment as stated by FEPA 1988(< 40^{o}C). PP had negative value for both nitrification and denitrification column, this indicated that the filtrate temperature was higher than that of the influent; the increase in the temperature in the filtrate may be attributed to the metabolic activities of the bacteria in the media. The low

percentage change in values of temperature between the influent and the filtrate for the two media showed that temperature the systems did not experience wide variation and hence the culture organism will not experience sudden change in temperature and shock that may arise from it (Le'Morvan and Deschaed 1995).

Table 3 also revealed that pH was higher in PP filtrates, 7.47±0.32 and 7.57±0.25 in the nitrification and denitrification columns respectively but the difference was not significant (P >0.05) between PP and PK filtrates. The change in pH was positive in PK filtrates for both nitrification (0.93%) and denitrification column (1.75%), PP filtrates also recorded a positive change in the nitrification column (0.40%) but a negative change (-1.34%) in the denitrification column (Table 4). All the pH values were within the recommended range (6.5-8.5) for warm water fish culture (Akinwole 2005) and range recommended (6.0 – 9.0) for discharge to the environment (FEPA 1988). The pH in the columns for the two media was within the range of 7.0 – 8.0 regarded as optimum for nitrifying bacteria (Michael *et al*., 1995) and denitrifying bacteria (Glass *et al*., 1997). The percentage change was very small in all the columns, this indicated a stable system.

The Dissolved oxygen (DO) for the nitrification columns was higher in PK filtrate (3.53±1.36 mg/L), but in the denitrification columns, PP filtrate recorded the higher DO (3.43±0.38 mg/L) (Table 3). The change in DO between the influent and the effluent for PP was positive (6.45%) in the nitrification column and negative (-18.28%) in the denitrification column while for PK it was negative (-13.87%) in the nitrification column and positive (5.67%) denitrification column (Table 4). T-test did not show a significant difference between the two media in both nitrification and denitrification column. Though all the dissolved oxygen values were very low and below the >4 mg/L recommended for warm water fish culture by Ajani *et al*., (2011), they are still within the safe range (> 2 mg/L) for discharge into surface water (FEPA 1988). DO in both media can still support the metabolic activities of the nitrifying bacteria, this in line with Malone (1995), who stated that DO below 2 mg/L will limit the activities of Nitrobacter and Nitrosomonas in biological filters. While in the denitrification column, DO can be regarded to be high because the column is suppose to be anaerobic column (Abeysinghe *et al*., 1996) and DO above 2mg/L can still encourage further nitrification. The general low DO in the filtrates should not be regarded as a negative development in RAS facilities or any aquaculture water reuse system (Akinwole, 2005), filtrates are to be re aerated before its return to fish rearing tanks. In the nitrification column, the negative values recorded in the PK filtrate indicated that the filtrates DO is higher than that of the influents, this is contrary to observation of Akinwole (2005), who recorded lower values for the filtrates compared to the influents in biofilters with sharp sand media. This can be attributed to the higher void ratio of PK compared to that of sand used as biofilter media by Akinwole (2005), the cascading movement of the wastewater in the media may also account for the relatively elevated DO. The denitrifying column is an anaerobic column and oxygen is not expected to be consumed, but the positive value in PK reflected that filtrate had lesser values than influent and this may be attributed to high amount of DO in the influents that encouraged some nitrifying bacteria to still thrive in the columns and that will likely reduce the denitrification efficiency.

According to Table 3, PP and PK filtrates had same TAN in both nitrification (1.60±0.00 mg/L) and denitrification columns (1.33±0.46 mg/L). The change in TAN for the PP and PK filtrates was also 30.43% in the nitrification column and, 16.88% in the denitrification column. All the TAN reported were still within the recommended level (< 8.8 mg/L) for warm water fish culture (Akinwole 2005). Although the TAN for the two media was higher than 0.9 2mg/L reported by Al-Hafedh *et al*. (2003) and 0.02 mg/L reported by Ridha and Cruz (2001) but it was lower than 2.98 mg/L reported by Akinwole, (2005). The positive values for the change in TAN in the denitrification column indicated that there was further nitrification in the system and this tends to affect the denitrification performance of the column, the systems were able to nitrify further because of the DO that was above 2 mg/L that can still support nitrifying bacteria in the columns, as stated by Malone (1995), that the DO values can only limit the activities of nitrifying bacteria when it is below 2mg/L. All the nitrite nitrogen values were within the level recommended (<0.25 mg/L) for warm water fish culture (Ajani *et al*., 2011).

The NO_2-N values were higher in the PP filtrates than PK filtrates, 0.15±0.00 mg/L was recorded in both the nitrification column and the denitrification column for the PP filtrates, while PK filtrates had 0.13±0.04 mg/L in the nitrification column and 0.11±0.03 mg/L in the denitrification column (Table 3). The difference in NO_2-N between the two media was significant (P<0.05) in both nitrification and denitrification column. The change in NO_2-N was negative for both the PP filtrate and PK filtrate in the nitrification column while in the denitrification column, no change (0.00%) was observed in PP filtrates but a positive change in (15.39%) in the PK filtrate (Table 4). T-test showed a significant difference in changes between the two media in both columns. The PP filtrates had slightly higher NO_2-N

than the PK in both nitrification and denitrification column, this

Table 1: Physical properties of the two biofilter media evaluated

Media properties	PK	PP
Nature &Type	Natural , Crushed shell	Synthetic, Injection-moulded
Specific Surface area (m^2/m^3)	166.54	166.67
Mass density of dry media (kg/m^3)	833	Not applicable
Bulk density (kg/m^3)	800	400
Void ratio (%)	28	92
Volume of the media (m^3)	0.00977	0.00924

Table 2: Operating condition of the culture system used for wastewater sampling.

Parameters	Description
Organism cultured	*Clarias gariepinus*
Age of the fish (Weeks)	24
Feed type	Floating pellet
Feeding frequency/day	Twice
% Crude Protein in Feed	42
Type of water holding facilities	Concrete
Water Source	Deep well
Water exchange rate	weekly
No of Fish stocked	1,100
Volume of water in the holding structure (m^3)	11.4

Table 3: Mean values of selected water quality parameters in the nitrification and denitrification columns

Parameters	Influent	PP	PK
Nitrification column			
Temperature (°C)	28.17±.0.29	28.67±0.29	28.33±0.76
Dissolved Oxygen (mg/L)	3.10±0.00	2.90±0.66	3.53±1.36
pH	7.50±0.00	7.47±0.32	7.43±0.21
NH_4-N (mg/L)	2.30±0.00	1.60±0.00	1.60±0.00
NO_2-N (mg/L)	0.00±0.00	0.15±0.00[a]	0.13±0.04[b]
NO_3-N (mg/L)	0.00±0.00	110.00±0.00[a]	90.00±34.64[b]
Denitrification column			
Temperature (°C)	28.83±0.28	28.33±0.76	
Dissolved Oxygen (mg/L)	3.43±0.38	3.33±0.15	
pH	7.57±0.25	7.30±0.17	
NH_4-N (mg/L)	1.33±0.46	1.33±0.46	
NO_2-N (mg/L)	0.15±0.00[a]	0.11±0.03[b]	
NO_3-N (mg/L)	45.00±8.66	25.00±22.91	

Values (mean ±standard deviation) in the same row with different superscripts are significantly different ($P < 0.05$)

indicated that PK is slightly better in terms of NO_2-N. This could be attributed to higher biofilter flow in PP than PK, Abeysinghe *et al*. (1996), also noted that increase in bio-filtration rate will result in NO_2-N accumulation. The negative value indicated in the nitrification column indicated that nitrification did occur, since nitrite is an intermediate product in the oxidation of ammonia to nitrate (Philip 1997). So also the positive values in the denitrification columns established there was denitrification.

The NO_3-N in the PP filtrates was higher in the nitrification column (110.00±0.00 mg/L) and in the denitrification column (45.00±8.66 mg/L) and the difference in NO_3-N values recorded by the two media biofilters was significant ($P<0.05$) in the nitrification column but not in the denitrification column (Table 3). The change in NO_3-N between the influents and the filtrates was negative in the nitrification column for both PP and PK, with a difference that is significant between the two media. While in the denitrification column the change was

positive for both PP (59.09%) and PK (72.22%) but the difference between the two media was not significant (Table 4). The negative values in the nitrification columns indicated that the filtrate had higher NO_3-N than the influent wastewater, establishing the fact that nitrification did occur in both media. The positive value in the denitrification columns showed that denitrification did occur in the two media. PK is relatively better in terms of denitrification, though the value recorded here can still be regarded to be low in the systems compared to 100% removal achieved by Abeysinghe *et al*. (1996) but, it is in line with that reported by Suzuki *et al*. (2003).

Table 4: Change in selected water quality parameters in the nitrification and denitrification columns

Parameters	PP (%)	PK (%)
	Nitrification column	
Temperature (°C)	-0.50±0.00[a] (-0.60)	0.17±0.58[b] (0.60)
Dissolved oxygen (mg/L)	0.20±0.65 (6.45)	-0.43±1.36 (-13.87)
pH	0.03±0.32 (0.4)	0.07±0.21 (0.93)
NH_4-N (mg/L)	0.70±0.00 (30.43)	0.70±0.00 (30.43)
NO_2-N(mg/L)	-0.15±0.00[a] (NA)	-0.13±0.03[b] (NA)
NO_3-N(mg/L)	-110.00±0.00[b] (NA)	-90.00±34.64[b] (NA)
	Denitrification column	
Temperature (°C)	-0.17±0.29[a] (-0.59)	0.00±0.00[b] (0.00)
Dissolved oxygen (mg/L)	-0.53±0.31 (-18.28)	0.20±1.37 (5.67)
pH	-0.10±0.10 (-1.34)	0.13±0.06 (1.75)
NH_4-N (mg/L)	0.27±0.46 (16.88)	0.27±0.46 (16.88)
NO_2-N(mg/L)	0.00±0.00[a] (0.00)	0.02±0.03[b] (15.39)
NO_3-N(mg/L)	65.00±8.66 (59.09)	65.00±22.91 (72.22)

Values (mean ±standard deviation) in the same row with different superscripts are significantly different ($P < 0.05$). NA means not applicable

Table 5: Denitrification performance variables for Polypropylene bio block and Palm kernel shell biofilters

Parameters	PP	PK
Residence time (s)	31.34±1.15	62.01±12.82
Biofilter flow m^3/s	9.68±0.27	5.01±0.79
PNR (%)	59.09±7.87	72.22±18.56
VNR ($mgNO_3$-N/m^3d)	58775.51±7068.43	31084.03±13478.05

Values (mean±standard deviation) in the same row with different superscripts are significantly different ($P < 0.05$)

Performance of the Biofilter Media

Water residence time (res. time) is naturally dependent on the void ratio of the media. The PP had res. time of 31.34±1.15s while the PK had 62.01±12.82s (Table 5). The biofilter flow for the PP was 9.68±0.27 $x10^{-5}m^3/s$ while PK has 5.01±0.79 $x10^{-5}m^3/s$ (Table 5). Though, there was difference between the two media but the difference was not significant ($P>0.05$).

As shown in table 5, PNR was higher in PK (72.22±18.56%) than PP (59.09±7.87%) but for the VNR, PP had higher conversion rate (58775.51±7068.43mg/m^3d) compared to PK (31084.03±13478.05mg/m^3d) but the differences in both cases between the two media was not significant

(P>0.05). The res time is higher in PK and this tends to positively influence the performance of the media because the wastewater spent more time in the column for treatment. That can be attributed to its lower void ratio compared to PP. Biofilter flow is depended on the residence time and nature (void volume) of the media and is far higher in PP than PK and this can be attributed to the high void ratio in PP. Denitrification efficiency is based on reduction in NO_3-N concentration and it was measured using Percentage NO_3-N removed (PNR) and Volumetric NO_3-N conversion rate VNR in NO_3-N mg/m^3d as adapted from estimation of nitrification efficiency suggested by Colt *et al.*(2006). The PK performed relatively better than the PP and it was able to reduce NO_3-N from 90 mg/L to a level of as low as 25 mg/L (72.22% reduction). This is similar to the observation of Suzuki *et al.*(2003) and that of Menasveta (2001). Menasveta (2001) also noted that compacted media performed better in denitrification of wastewater, therefore the better performance noted with the PK can be attributed to it low void ratio compared to that of PP. The higher VNR recorded in PP biofilter despite low PNR compared to PK can be attributed to higher biofilter flow in the PP which allows for higher conversion rate of NO_3-N per unit time. PK is locally available at a very cheap price and its usage as biofilter media is highly economical compared to the imported and expensive polypropylene bio-block (Akinwole and Dauda, 2014). Findings of this study have established denitrification to be essential in water treatment systems for fish culture in addition to the usual nitrification of culture wastewater. This will not only prevent fish culture system from excessive accumulation of nitrate that may end up leading to stress and reduce the fish growth and yield but also assure that the environment is relatively safe of eutrophication due to discharge of high concentration of nitrate into the environment.

CONCLUSION

Palm kernel shell exhibited good potential as a biofilter media for denitrification of fish farm wastewater and it performed favourably compared to the expensive and imported polypropylene bio-block. The media produced reduction in polluting wastewater parameters to levels that are within the limit for discharge into the environment and also safe for reuse in fish culture. Its availability locally at a relatively cheaper rate compared to synthetic polypropylene bio-block, is an added advantage for its use to be highly encouraged.

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