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Nitrogen removal and microbiology of wood chip bioreactors in treatment of aquaculture effluents

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There is an essential need for simple, low cost and low maintenance technologies to remove nitrate from aquaculture effluent in order to improve sustainable development of recirculating fish farming. Heterotrophic denitrification reactors are typical treatment units applied in the purification process to mitigate the nitrate concentration in aquaculture effluent. However, due to their dependency on readily available organic carbon sources, they face practical and economic limitations. In this study, the purification performance of bioreactors packed with different carbon sources treating aquaculture effluent under design hydraulic retention time of 48-h and temperature of 15.4$\pm$ 0.3˚C was investigated. Four clear acrylic vertical columns (diameter = 10 cm, height = 32 cm) with upward flow direction were packed with birch wood chips (BR1), and mixtures of those woodchips with three different carbon sources: potato peels from industrial process (BR2), dried *Sphagnum* sp. moss material (BR3) and biochar (BR4). Nutrients, organic carbon fractions and other reactor parameters (e.g. ORP, BOD and COD) in bioreactor inflow and outflow were evaluated weekly. The results clearly showed instant NO_3^--N reduction in all bioreactors and stable removal efficiency after one week. The influent mean nitrate concentration of 35.6 mg NO_3^--N L^{-1} reduced to $<$ 10 mg NO_3^--N L^{-1} in all bioreactors, which corresponded to a 78 % removal efficiency and $\sim$18 g N $m^{-3}d^{-1}$ removal rate. The average NO_3^--N removal rate ranged from 31 to 38 g N $m^{-3}d^{-1}$. Reported removal rates ranked BR2$>$BR1$>$BR3$>$BR4; respectively. As much as 99.6% of the NO_3^--N was removed from aquaculture effluent with 69.5 mg NO_3^--N L^{-1} concentration, in bioreactor BR2, corresponding to daily rate of 38 g N $m^{-3}d^{-1}$. The bioreactor BR2 revealed also more stable removal efficacy over time, while removal rates declined in the other bioreactors after 151 days. Start-up leaching of organic carbon and ammonia was observed in all the bioreactors, but with higher rate in bioreactor BR2. Continuous production of ammonia in bioreactor BR2 indicated dissimilatory reduction of nitrate to ammonia (DRNA). After 69 days of operation, the microbial community composition on the woodchip reactors consisted of .*Proteobacteria* (58% of sequences), *Bacteroidetes* (17%),and, *Firmicutes* (8%), determined by IonTorrent PGM sequencing of the 16S rRNA genes. In bioreactor BR2, the diversity and richness indicators of the microbial community were much lower than in the other three bioreactors. Moreover, the amount of OTUs shared between bioreactor BR2 and the other bioreactors was low. In bioreactor BR2, the relative abundance of *Firmicutes* and *Epsilon-Proteobacteria* was higher than in the other bioreactors, while the relative abundance of *Alpha-, Beta-,* and *Gamma-Proteobacteria* as well as *Actinobacteria* was lower. This indicates that the functional potential in the bioreactor BR2 differed from that in the other bioreactors, resulting in the increased NO_3^--N removal.