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INVESTIGATING THE EFFECT OF SALINITY ON THE PERFORMANCE OF DENITRIFICATION BEDS TO REMOVE NITRATE FROM AGRICULTURAL DRAINAGE WATER

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ABSTRACT

Contamination of groundwater and surface water with nitrate in the world is considered to be a serious problem. Finding a strategy to remove nitrate from agricultural drainage water before entering the river can play an important role in reducing river water pollution. One possible strategy for nitrate removal is to use denitrification beds. The aim of this study was to investigate the effect of salinity on the performance of denitrification beds to remove nitrate from water. Therefore, Therefore, four denitrification beds with a rectangular cross-section were constructed and sesame straw was poured into them. Input nitrate concentration in the denitrification beds in this study, approximately 50 mg per liter. In order to simulate the inflow, four salinity levels of about 1, 4, 7 and 10 ds/m were used. The experiment was performed in 6 months. The nitrate concentration of the samples was measured with 220 nm wavelength of spectrophotometer. In this study, Excel 2016 software was used to show the effect of salinity on the amount and efficiency of nitrate removal in each denitrification bed. The results showed that in constant hydraulic conditions, beds in higher salinity had lower performance for optimal nitrate removal; However, the performance of the beds designed in salinity levels 1 and 4 ds/m, were similar. According to the results of this study, these bioreactors can be used to remove of drainage water nitrate up to salinity of 4 ds/m. However, it is predicted that there should be a threshold limit value for the salinity level. In general, this study shows that beds designed by sesame straw can be used as optimal beds for nitrate. removal.

***Keywords*:** Nitrate, Denitrification beds, Salinity, Sesame straw, Constant hydraulic conditions

Introduction

Nitrate is an anion that is not present enough in soil. Therefore, farmers have to use nitrogen fertilizers to meet the needs of plants. Nitrate leaching is from the soil very quickly and enters surface water through a subsurface drainage system (Jackson *et al.*, 1973). With the entrance of nitrogen compounds into rivers, problems such as accumulation of nitrates in aquatic creatures' tissues (especially fish) and disturbance in their reproduction, incidence of diseases and endangering human health, feeding the region's waters and algae growth may occur (Fei, 2004; Shimura *et al.*, 2002). Therefore, it is necessary to target nutrient management in order to increase the environmental sustainability of agricultural systems (Christianson *et al.*, 2010). There are several methods for removing nitrates from water, and biological removal (biological denitrification) is one of the most suitable methods for removing nitrates from the drainage water of farm subsurface drainage (Soares, 2000). One of the possible strategies of biological denitrification is the use of denitrification beds. Denitrification beds are an affordable technology for removing nitrate from agricultural drainage water (Cameron and Schipper, 2011). Denitrification beds are trenches filled with organic matter and act as a source of energy for bacteria that convert nitrate to nitrogen gas (Warneke *et al.*, 2011). The determinant key of nitrate removal is access to carbon by denitrifying bacteria; and any process that removes denitrifying bacteria from competition to access carbon will reduce the rate of nitrate removal in denitrification bioreactors (Schipper *et al.*, 2010). High salinity is a key problem that affects the wastewater biological treatment process (Chen *et al*., 2016). In recent years, several articles have been conducted on the effect of salinity on the activity of microorganisms in denitrification bioreactors. These studies show that high salinity in sewage can somehow lead to a destruction of the balance of decomposing enzymes and also lead to a decrease in cellular activity and cell plasmosis (Rietz and Haynes, 2003). Bassin *et al*. (2012) reported that salinity stress can affect the performance of denitrification by altering the physical and chemical properties of microbial populations. Wu *et al*. (2008) evaluated the denitrification potential at different salinity levels on treatment of municipal wastewater by constructed mangrove wetland microcosms. The results of their research showed that the denitrification potential has decreased at the highest salinity level. Numerous studies conducted in swampy soils have shown that increasing salinity leads to lower nitrogen mineralization rates. In other words, salinity leads to biological stress in microbial communities and ultimately leads to a decrease in microbial population and poor performance of existing microbial communities (Jackson and Vallaire, 2009). Jafari *et al*. (2015) examined the performance of a rotary biological reactor to remove nitrate at several salinity levels. The researchers observed that at the beginning of the experiment, denitrification was a function of salinity, so that with increasing salinity, the efficiency of nitrate removal decreased and after a few days of experimentation, it again approached stable conditions (100% reduction in nitrate). The results of this study show that the denitrifying bacteria in the reactor are resistant to salinity stress over a period of time and are able to repeat the metabolic conditions for the biological denitrification process. Di Capua *et al*. (2015) investigated the effect of sodium chloride salt concentration simultaneously on heterotrophic and autotrophic denitrification based on sulfur and methanol as an organic carbon source. The researchers observed a complete elimination of nitrate of up to 3.5% of sodium chloride salt and reported that increasing the salinity concentration to 4% and 5%, respectively, led to a decrease in nitrate removal efficiency to 78% and 48%, respectively. Kristensen and Jepsen (1991) the occurrence of denitrification process in the presence of 30 g/l of sodium chloride salt (NaCl) have reported to be successful. In addition, the reduction in C: N ratio in soils maintained under higher salinity has been reported by Yuan *et al*. (2007). So far, little research has been done on the effect of salinity on denitrification performance, and there is still no general agreement (Zhou *et al*., 2013). Based on the research background, it seems that a comprehensive study on the effect of salinity on the performance of denitrification beds for nitrate removal has not been performed. Therefore, the aim of this study was to investigate the effect of salinity on the performance of denitrification beds for nitrate removal.

Materials and Methods

In this study, four beds with a rectangular cross section (length 2 m, width 1 m, depth 1 m) were constructed. After the beds were formed, in order to prevent water from penetrating into the ground, they were cemented and then bituminous waterproofing. After this step, they were filled with sesame straw. After filling all the beds with sesame straw, it was covered with plastic on all the beds and then on plastics, soil was dumped. The concentration of nitrate required to enter the beds is 50 mg/l. Potassium nitrate was used to make nitrate solution. To prepare the 50 mg/l nitrate solution, 82.26 g of potassium nitrate was weighed first and dissolved in a bucket with some water for each tank and then poured into the tanks. In this study, four 1000-liter tank were used to supply water to the beds, and the salinity of each tank was different with else tank (The salinity of the water used in the tanks was 1 ds/m. According to the research topic, NaCl salt was used to adjust the salinity of the tanks according to the four salinity levels studied.). To simulate the inflow, four salinity levels of 1, 4, 7 and 10 ds/m were used. However, it should be noted that the water used to fill the tanks is not purified water and contains some nitrate (40 mg per liter). The experiment was performed in 6 months. Sampling was conducted from beds on a daily basis. Each day after sampling, the samples were transferred to the refrigerator and after the end of the sampling period, qualitative analysis of the samples was performed. Nitrate measurement of samples was performed by spectrophotometer. To measure the nitrate of samples by spectrophotometer, you must first find an equation between the concentration of soluble nitrate and the absorption number; then read the adsorption number of the samples and put these numbers in the equation to get the nitrate concentration of the samples. For this purpose, nitrate standard solutions with a concentration of zero (distilled water), 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 mg per liter were made. After preparing the standard solutions, the nitrate absorption number of the solutions was read with spectrophotometer in a wavelength of 220nm. To find the relationship between nitrate concentration and absorption number, the data were transferred to Excel software and by fitting the data, the linear equation y = 19.676x + 2.7786 was obtained with an R^2=0.99(Coefficient of Determination); in which x is the absorption number and y is the nitrate concentration (Figure 1).

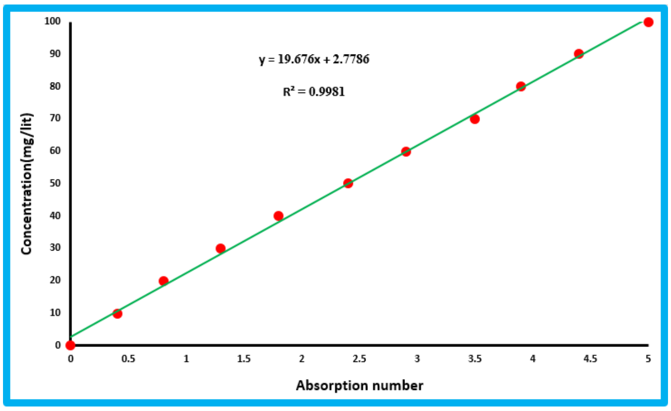

Fig. 1 : Nitrate standard data fit chart; and the result of the relationship between the absorption number and nitrate concentration

The efficiency of nitrate removal from the relationship reported by Ghane *et al*. (2015) was calculated (Relationship 1).

$$R\ \% = (C_i - C_{ef})/C_i \times 100 \qquad ...(1)$$

In this relationship, C_i and C_{ef} are the inflow and outflow nitrate concentrations from the beds in milligrams per liter (mg/l), respectively.

In this study, Excel 2016 software was used to show the effect of salinity on nitrate removal efficiency in each denitrification bed.

Results and Discussion

Figures 2, 3, 4, and 5 show the efficiency of nitrate removal in 180 days by sesame straw at salinity levels of 1, 4, 7, and 10 ds/m. Based on Figure 2, it can be said that the efficiency of nitrate removal in this period varies between 30.95 and 89.53. In Figure 3, the nitrate removal efficiency changes in 180 days between 30.86 and 89.26. The efficiency of nitrate removal at a salinity level of 7 ds/m varies from 24.69 to 71.41 (Figure 4). Figure 5 shows the efficiency of nitrate removal at a salinity level of 10 ds/m and varies between 17.02 and 49.24 in 180 days. The rate of nitrate removal efficiency has an inverse relationship to the nitrate concentration of the outflow from the denitrification beds. This means that the higher the nitrate removal efficiency, the outflow nitrate concentration from the denitrification beds decreases. Therefore, it can be said that in all four denitrification beds, the nitrate concentration of the outflow from the denitrification beds has a decreasing trend compared to the nitrate concentration of the inflow to the denitrification beds. Therefore, it can be said that sesame straw is a suitable carbon material for reducing the nitrate of aqueous solutions. Also, according to Figures 2, 3, 4 and 5, it can be said that the process of decreasing the nitrate concentration of the outflow (increasing the efficiency of nitrate removal) compared to time in all four denitrification beds is an increasing trend. In the early days of the experiment, the rate of nitrate removal is lower than in other days because the environment of the denitrification bed is still unsaturated. By creating a saturated environment, the activity of anaerobic microorganisms, especially heterotrophic bacteria, increases. Due to the presence of sesame straw in denitrification beds with rectangular cross-sectional area, the growth rate and activity of this type of bacteria increases extremely. These bacteria perform denitrification to gain energy, and as oxygen levels fall below a certain threshold, synthesis of denitrificator enzymes begins. In the absence of oxygen, these bacteria supply the oxygen they need from the nitrate in the water and convert the nitrate into nitrogen gases, including NO, N_2O and N_2.

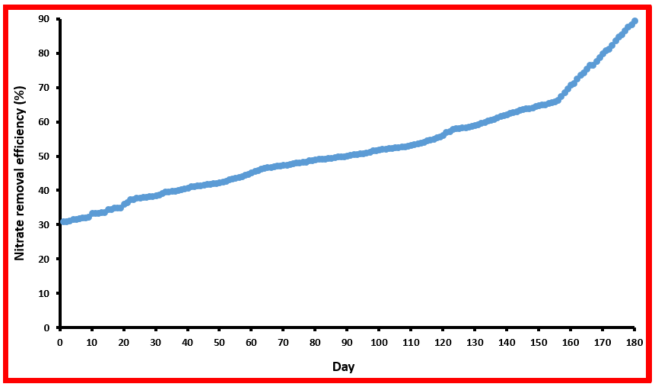

Fig. 2 : The efficiency of nitrate removal at the salinity level of 1 ds/m

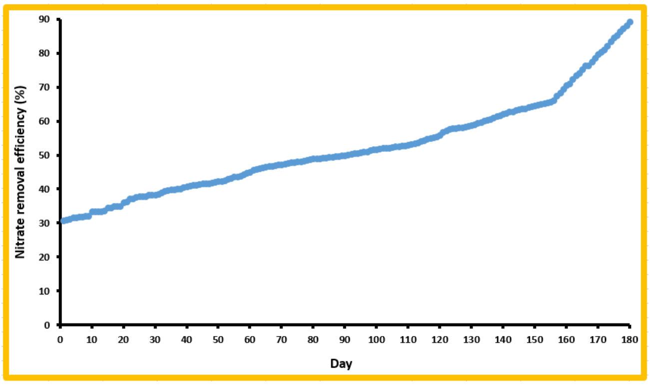

Fig. 3 : The efficiency of nitrate removal at the salinity level of 4 ds/m

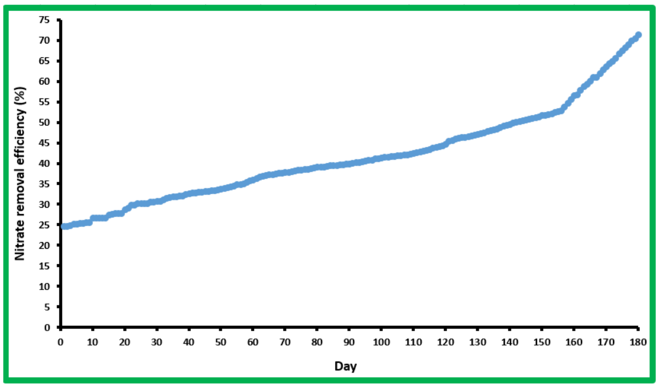

Fig. 4 : The efficiency of nitrate removal at the salinity level of 7 ds/m

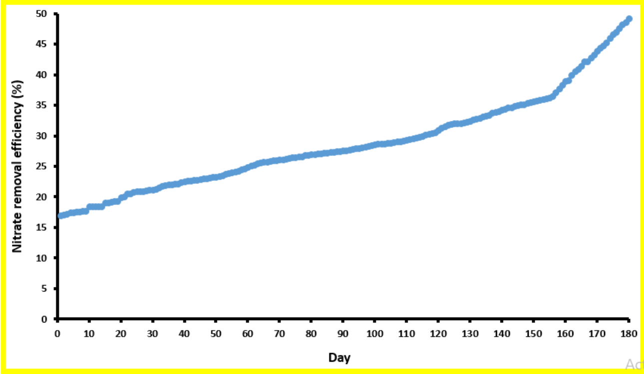

Fig. 5 : The efficiency of nitrate removal at the salinity level of 10 ds/m

Figures 2, 3, 4, and 5 show the salinity relationship with nitrate removal efficiency. As can be seen in these figures, the efficiency of nitrate removal decreases with increasing electrical conductivity. That is, in constant hydraulic conditions, beds with higher salinity have lower performance for the ideal removal of nitrate. However, the performance of the beds designed in salinity levels 1 and 4 ds/m were similar. In general, declining nitrate removal efficiency with increasing salinity indicates that bacteria in denitrification beds are under stress and, despite having sufficient carbon source, are unable to remove nitrate at the optimal level. Therefore, it can be said that the results of this study confirm the results of researchers such as Jafari *et al*. (2015) and Di Capua *et al*. (2015). These researchers reported that increasing salinity in microbial communities caused biological stress so that the metabolism of these communities increased and eventually the function of the denitrification bed decreased. Lefebvre *et al* (2012) also reported that salinity has a negative effect on the physiology of anaerobic bacteria and reduces denitrification. According to the results of this study, these bioreactors can be used in the removal of nitrate from drainage water with salinity up to 4 ds/m. However, it is predicted that there should be a threshold limit for the salinity level. In general, this study recommends beds designed with sesame straw as beds with optimal efficiency for nitrate removal. According to the results of this research, the following suggestions are provided for further research.

1) Evaluation of the performance of denitrification beds in inflow nitrate concentrations of more than 50 mg/l
2) Evaluation of the performance of denitrification beds in salinity (electrical conductivity) of more than 10 ds/m
3) Evaluation of the performance of denitrification beds for a longer period of time (more than 6 months)
4) Investigating the performance of other carbon materials for use in denitrification beds

Conclusion

The results of this study showed that the nitrate concentration of the outflow from the denitrification beds has a decreasing trend compared to the nitrate concentration of the inflow to the denitrification beds. Therefore, it can be said that sesame straw is a suitable carbon material for removing nitrate from water. The trend of decreasing nitrate over time in all four denitrification beds is an increasing trend. This means that in the early days of the test, nitrate removal is lower than in other days because it is not a lot of time from start of the experiment ago. With the start of the experiment and with the creation of a saturated environment, the activity of anaerobic microorganisms, especially heterotrophic bacteria, increases. Due to the presence of sesame straw in denitrification beds, the growth rate and activity of this type of bacteria increases sharply. These bacteria perform denitrification operation to obtain energy, and because the oxygen content is less than a threshold value, synthesis of denitrifying enzymes begins. In the absence of oxygen, these bacteria supply the oxygen they need from the nitrate in the water and convert the nitrate into nitrogen gases, including NO, N_2O and N_2. The results also showed that salinity is a factor influencing the denitrification process that overshadows the performance of denitrification beds. According to the results of this study, with increasing salinity in denitrification beds, the rate and efficiency of nitrate removal decreases. Because the bacteria in the denitrification beds are under stress, and despite having enough carbon source, they are not able to remove nitrate to the desired level.

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