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Mechanism and Influencing Factors of Nitrogen Removal in Subsurface Flow Constructed Wetland

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ABSTRACT

The mechanism of nitrogen removal and the transformation of nitrogen compounds were reviewed. A new way to increase nitrogen removal rate was pointed out. Based on the study of temperature, pH, dissolved organic carbon, dissolved oxygen and other factors, as well as hydraulic retention time, nitrogen load, reflux, water and other configuration patterns outlined, environmental factors and operating conditions for the subsurface effect of nitrogen removal on constructed wetland were explored.

KEYWORDS: subsurface flow constructed wetland; denitrification;

The rapid economic expansion, industrial development, and urbanization have resulted in urban rivers and groundwater pollution, water shortages and a series of environmental problems that threaten human health. Artificial wetland is an effective sewage treatment method. It can be applied to wetland plants, substrates, and microbial mutual role, in an approximate natural way to deal with municipal, agricultural, and industrial wastewater [1-2]. As the water in subsurface flow of artificial wetlands is not exposed to the surface, the water has many good characteristics including good insulation effect, not subject to environmental climate constraints, and good sanitation. Therefore, the wetland system is highly applied and researched internationally [3-5]. For the ability of artificial wetlands to purify pollutants, the degree of nitrogen removal is an important indicator of its effectiveness. Nitrogen compounds are the main components of sewage pollutants, which are also the important indicator of water eutrophication. Within a certain range, it may promote the growth of wetland plants; it can also produce toxic effects on organisms [6-7]. The nitrogen cycle in constructed wetlands is a very complex process. The migration and the transformation of nitrogen in wetland environments involve a lot of physical, chemical and biochemical reactions [8-9]. Therefore, this paper summarizes the current nitrogen removal mechanism in subsurface constructed wetlands and the factors affecting its denitrification, to provide some reference for the researchers of artificial wetland and to prospect the existing problems.

1. Mechanism of Nitrogen removal in Constructed Wetland

Nitrogen is present in both organic and inorganic forms in sewage. Organic nitrogen is present in amino acids (consisting of amino acid chain-forming proteins), urea and uric acid (the simplest form of organic nitrogen in water), and purine nucleoside (the main constituent of DNA). The inorganic nitrogen is present in the form of ammonia nitrogen (NH4 +), nitrite nitrogen (NO2-), nitrate nitrogen (NO3-), nitrogen oxides (N2O) and nitrogen (N2). Figure 1 shows the main removal of nitrogen in subsurface wetlands.

(1) Ammoniation: Ammoniation is the biotransformation process of organic nitrogen to ammonia nitrogen conversion, which is the first step of nitrogen transformation if the sewage is rich in organic nitrogen. This process can be carried out under aerobic or anaerobic conditions, in the dead or decay of cells or tissues to release ammonia nitrogen. This is a complex energy release process with biochemical reactions that are more easily oxidized by amino acids to deoxyglutination to produce NH3. In the wetland system, this effect increases with the depth of the filler layer, which is the most efficient top layer of the wetland.

(2) Nitrification and denitrification: In most of artificial wetland systems, nitrification is the main process for the conversion of ammonia to nitrate nitrogen[10-11]. This is a two-step reaction process involving microbes. The first step is to complete the nitrosomonas, Nitrosococcus and Nitrosospirabacteria to convert NH4-N to NO2-N. The second step

is carried out under the action of nitrobacteria nitrospira and nitrobacter, and further into NO3-N with the participation of oxygen. Denitrification is the main process of total nitrogen removal in wetlands [10,12-13]. Similarly, in the presence of microorganisms (Bacillus, Enterobacter, Micrococcus, Pseudomonas, Spirillum, etc.), oxidized nitrogen acts as an electron acceptor, resulting in nitrogen, N2 O, NO, which is energy consuming [14].

(3) Biological assimilation: This refers to the biological processes that breakdown inorganic nitrogen from a system into organic nitrogen in the nuclear tissue [15]. Generally there are two forms of assimilation which are ammonia nitrogen and nitrate nitrogen, although nitrate nitrogen can also be used by some plants, ammonia nitrogen is more susceptible to the use of wetland plant [16]. If the system is lacking an external carbon source and no ammonia is present, the plant will also use nitrate nitrogen to meet the nutritional requirements.

(4) Nitrate Alienation: This pathway refers to the process of converting NO2-N and NO3-N into NH4-N in wetland systems. This effect generally occurs in carbon-rich and nitrate-limited environments [17]. Generally facultative or obligate anaerobes participate in this transformation process, formate act as the main electron donor, making it easier to reduce nitric acid or nitrite.

(5) Plant uptake: Although the direct absorption of nitrogen on the plant is limited, but the role of the entire denitrification system cannot be ignored. The roots of the plant provide oxygen and carbon for the growth of microorganisms [18]. A comparative studies between plant and non-plant wetlands have shown that, nitrogen removal effect in plant wetlands is relatively significant. The higher the diversity of plants, the better the production of biomass and the retention of nitrogen in the matrix. It is also more conducive to denitrify constructed wetlands [17].

(6) Ammonia volatilization: Ammonia volatilization is a physical process. Non-ionic ammonia is relatively volatile, through the upward diffusion to the surface, and then through the liquid gas surface mass transfer into the atmosphere [17]. Ammonia volatilization may be limited due to the possibility of mass transfer in the subsurface wetlands. Ammonia volatilization requires a pH of water > 9.3 to occur. Water pH of <7.5 will inhibit NH4 + conversion to NH3. In general, the proportion of ammonia volatilization to total nitrogen removal is small.

(7) Adsorption: Artificial wetland adsorption refers to the exchange adsorption between some of the components of the matrix and NH4 + cation, to remove ammonia in the sewage. This adsorption process is reversible, when the concentration of ammonia in water decreases, the ammonia nitrogen ions will be re-resolved to maintain the chemical balance. In the vertical wetland system, the ammonia nitrogen adsorbed on the substrate can be nitrified by the microorganisms on the substrate, as the majority of the matrix is in the aerobic environment. This is due to the principle of the intermittent flow of water into and out of the vertical wetland.

(8) Short-range nitrification and denitrification: This process includes the conversion of NH4-N to NO2-N, and then denitrification of NO2-N to release N2 gas.

$NH_4^- + 1.5O_2 \rightarrow NO_2^- + H_2O + 2H^+$

$NO_2^- + 1/2\ CH_3OH + H^+ \rightarrow 1/2\ N_2 + 1/2CO_2 + 1.5\ H_2O$

From the formula above, we can see that nitrate nitrogen is inhibited due to the low oxygen concentration in the environment [19-20]. In comparison to conventional nitrification and denitrification reactions, the advantage of this pathway is that the demand for oxygen and organic matter is reduced by 25% and 40% respectively. This pathway was recently discovered in the artificial inflow level of subsurface flow wetlands, and the researchers found that NH4-N oxidation occurred apparently at 3/4 of the wetland extension (near the controlled forced aeration system). This process produces a large amount of NO2-N, which has a positive effect on the removal of TN [21].

(9) Anaerobic ammonium oxidation: Anaerobic ammonium oxidation process is occured under anaerobic conditions. NO2- act as an electron acceptor, through this process it oxidized ammonia to N2. Nitrite and ammonium ions that present in the environment can be oxidized through anaerobic ammonium oxidation to produce single molecules. This process can be described as follows:

$NH_4^- + NO_2^- \rightarrow N_2 + 2H_2O$

Furukawa et al. [22] considered that this process was described by the following formula, taking into account all chemical processes including nitrite formation and microbial growth and metabolism requirements:

$NH_3 + 0.85\ O_2 \rightarrow 0.44NO_3^- + 1.43\ H_2O + 0.14H^+$

Anaerobic ammonium oxidation contributes to the removal of nitrogen in the hypoxic or anaerobic regions of the subsurface wetlands. The advantage of conventional nitrification and denitrification is that no additional carbon source required (anaerobic ammonium oxide is autotrophic), lower oxygen demand, and low energy consumption [23].

(10) Full self-sustaining denitrification (CANON): A process that is able to perform short-range nitrification and anaerobic oxidation simultaneously [24]. The main principle of this process relies on the anaerobic ammonium oxide and autotrophic denitrifying of bacteria in a single reactor. The growth of anaerobic bacteria can avoided in the absence

of oxygen environment, at the same time create conditions for short-range nitrification [25]. - The process of CANON described by Sliekers and Van Hulle et al. is as followed [26-27]:

NH4- + 0.9 O2 + 1.11HCO3- → 0.010 3C5 CH7O2N + Ammonia Bacteria + 0.028 CH2O0.5N0.15 (Anaerobic Ammonia Bacteria) + 0.11 NO3- + 0.44 N2 + 1.06CO2 +2 49 H2O

Yuansheng Hua et al. [24] found that under the condition when nitrogen level is above 15 g N / m2 / d, the total inorganic nitrogen in domestic wastewater can be removed by 80% by controlling the contents of oxygen and inorganic carbon.

2. Effects of environmental factors and operating conditions on denitrification

(1) Temperature: Temperature varies greatly from day to night, among seasons and different latitude. It has great influence on the treatment efficiency of wetland. In the wetland system, the processes of ammoniation, nitrification and denitrification are related to temperature; therefore the removal rate of total nitrogen is also highly influenceable by temperature. In addition, plant photosynthesis and transpiration are also closely related to temperature, while plants have a fixed effect on CO2, and the roots can provide a relatively oxygen-enriched environment, and thus can indirectly affect the removal of N [28]. Generally, in the subsurface wetlands, the optimum temperature for nitrification is 16. 5 - 32 ℃, in less than 5 ℃ or higher than 40 ℃ environment is strongly inhibited. Similarly, denitrification is less than 5 ℃ is also very difficult to work [29], and in 20 = 25 ℃ most appropriate.

(2) pH: Wetland water chemistry and biological processes are affected by pH [30]. The appropriate pH range for microorganisms in sewage is 4 - 9. 5, nitrifying bacteria is greater than 7.2, and denitrifying bacteria is 6. 5–7. 5 [17].

(3) Dissolved organic carbon: The lack of carbon sources in sewage is often the limiting factor for organic matter removal, and microbes in wetlands require carbon as sources for nutrition and reproduction [31]. When the NO3 concentration is high in the wetland, the nitrification is dominant, and the organic carbon becomes the main factor restricting the denitrification. On the other hand, when the dissolved organic carbon is sufficient, it will promote the denitrification of the system. At this time, NO3 becomes the main factor controlling denitrification [32-33]. It can be seen in Figure 2 that the action of dissolved organic carbon and denitrification are carried out in the same region. Therefore, addition of carbon into different types of submerged wetlands and sewage according to its carbon and nitrogen ratio can enhance the removal of nitrogen.

(4) Dissolved Oxygen: The distribution of dissolved oxygen in wetlands has a direct effect on the removal of nitrogen. At the same time, the distribution of oxygen is also restricted by the degradation of pollutants [34]. There are two main ways of aerobic following contamination of sewage in wetlands: carbonization and nitrification of oxygen. These occurred mainly in the deposition of scattered oxygen content, respiratory oxygen demand, dissolved organic oxygen demand and dissolved nitrification oxygen demand [35]. Figure 2 shows that the aerobic microenvironment produced around the rhizosphere can guide the nitrification reaction while the denitrification reaction is carried out in an anaerobic environment away from the root zone [36]. The aerobic and anaerobic zones in the constructed wetland system often coexist, and the dissolved oxygen concentration will change with the hydraulic fluctuation. Nitrification is reduced and denitrification is inhibited when DO is <1 mg/L and > 0. 2 mg/L in water respectively [37].

(5) Hydraulic retention time / hydraulic load: Hydraulic retention time and hydraulic load are important factors controlling the removal of pollutants in wetland systems [38-39]. The increase in hydraulic load allows the water flow to pass through the substrate quickly and reduce contact time with the substrate. In generally reduction of the hydraulic load is beneficial to nitrogen removal; however it also depends on the composition of the effluent and other conditions. Developed plant roots can not only provide oxygen for microorganisms, but also can form a buffer for high hydraulic load. By the same token, increased hydraulic retention time can increase the removal efficiency of nitrogen and increase the contact time between contaminants and matrix biofilms [40-42]. However, in engineering applications, the design of excessive residence time will increase the area of wetlands, therefore a reasonable time to adjust the hydraulic retention or load according to the actual situation should be considered.

(6) Nitrogen load: The intensity of nitrogen load in constructed wetlands will have a certain effect on nitrogen removal rate [43-44]. In the confluent wetlands, the nitrogen content is within the tolerance range, and the nitrogen load and its removal rate are generally positively correlated. If the tolerance is exceeded, the wetland plants will be harmed. The study showed that the resistance of Phragmites australis to ammonia nitrogen was not more than 160 mg/L, the duckweed was not more than 80 mg/L, and that of Vallisneria could not exceed 2 mg/L. More than this range could affect plant growth. Therefore, under the high nitrogen load intensity, the removal of nitrogen from plants is no longer significant, and microbes play a major role.

(7) Reflow: In the subsurface wetlands, the effluent is mixed with raw water at a certain rate, which can improve the removal efficiency of pollutants [5, 46]. The results showed that NH4-N removal rate was 10% higher than that of non-refluxed wetland at 1: 1 ratio in the horizontal subsurface flow wetland, and the removal rate of NH4 -N at relux wetland

at 10:1 ratio is 10% higher than that of 1:1 ratio. Cui Lihua et al. [48] showed that the size of the reflux ratio had a significant effect on TN and NO3-N in vertical subsurface wetlands. The return of nitrified effluent back to the original matrix improves the removal of TN and NO3-N degradation, increases the matrix and microbial contact, and plays the role of dilution of raw water.

(8) Water way: Water can directly affect the degree of contact between the sewage and filler, therefore the water supply for the removal of pollutants is very important [49]. In the subsurface flow wetlands, the water way can be divided into continuous flow, intermittent flow, tidal flow, batch water, stage water and so on. Some researchers have found that intermittent influent water in subsurface flow constructed wetlands is slightly better than continuous flow for organic and nitrogen removal [50]. Osorio and García [50] argue that the advantage of intermittent flow is due to: (1) intermittent flow can make the water within the matrix to form a larger turbulence, allow the sewage to go through a better aerobic - anaerobic zone; (2) the high load flow of intermittent flow causes the reactor to form a larger space, allow plant roots to release more oxygen. The flow of tidal streams and batches is similar to the intermittent flow, and the effect is slightly better than the continuous flow [51]. Stefanakis et al. [52] used this method to distribute water at 0, 1/3, 2/3, in the horizontal subsurface wetlands, for better removal of organic matter and nitrogen.

(9) Plant harvesting: Plants can absorb certain contaminants in the effluent, and the roots can provide adhesion and oxygenation to microbes [53-54]. Yin Wei et al. [55] studied the reeds in wetlands and TN absorption of 140 kg / hm2 was obtained, which accounts for about 8% of the total nitrogen removal. Plants are the strongest growers in the summer, and the demand for nutrition is also the highest. Therefore, harvesting plants at the appropriate season can increase nitrogen removal efficiency.

3. Outlook

The artificial wetland purification system is a environment friendly, low energy consumption and high efficiency processing system. It also has good ecological benefit and landscape function. Denitrification mechanism and influencing factors of this system has been explored extensively by many researchers, and various classical denitrification methods have been widely recognized. However, there are still many problems to be solved in further increasing the removal of nitrogen. The issue in optimizing the effectiveness of wetland systems is discussed as followed.

(1) The removal of nitrogen by subsurface flow wetlands is usually achieved by planting aquatic plants, through nitrification and denitrification of microbes in the matrix, to achieve the desired effect. The general conventional denitrification pathway requires conditions such as maintaining a basic state of water, a continuous aerobic-anaerobic environment, and an availability of carbon source, but these conditions are often difficult to achieve. Therefore, these technical problems can be solved by other means, such as short-range nitrification and denitrification, anaerobic ammonium oxidation, full-range autotrophic denitrification and other means. These pathways also exist in subsurface wetlands without the need of organic carbon as an electron donor. However, the optimal environmental parameters of such bioreactors need to be studied, such as pH, dissolved oxygen conditions, temperature and etc; its load conditions need to be further explored, such as influent NH4-N concentration and NO2-N ratio.

(2) The intrinsic relationship between environmental factors and operating conditions affects the removal of nitrogen in wetlands. General temperature and dissolved oxygen often limit the degradation of nitrogenous substances, such as in cold regions, the temperature will inevitably affect the wetland system processing efficiency. So you can take the method of forced aeration, or add moisture insulation for the wetlands to improve the efficiency of treatment. But the former will increase the cost of constructed wetlands while the latter is more economical. Similarly, the removal rate of nitrogen in subsurface wetlands can be increased by controlling the hydraulic load, adjusting the pollutant load and influent mode, and increasing the recirculation facility.

The choice of these intensification measures needs to take into account the effects of the environment, the wetland configuration, the type of sewage, the amount of load, the concentration of pollutants and the geographical location. Most of the above methods can improve the effectiveness of the classical denitrification pathway. Future studies should strengthen and optimize the pathway, according to local conditions to improve the efficiency in nitrogen removal.

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