Denitrification rates determination in the process of removing nitrogen from wastewater

M Y Dyagelev¹, V G Isakov¹ and E V Grakhova²

¹Department “Water supply and water treatment”, Kalashnikov Izhevsk State Technical University, Studencheskaya 7, Izhevsk, 426069, Russia
²Department “Industrial and civil engineering” Kalashnikov Izhevsk State Technical University, Studencheskaya 7, Izhevsk, 426069, Russia

E-mail: mdyagelev@yandex.ru

Abstract. The purpose of the study was to determine denitrification rates during aeration tanks converting to the aero-mode of the nitr-denitrifier with the nitrate recycling. Studies were conducted on an experimental facility with wastewater and activated sludge from one of the sewage treatment facilities of the Udmurt Republic. At the beginning of the experiment, the activated sludge was under anoxic conditions (the mixture was mechanically mixed without aeration), while periodically the mixture was analyzed for nitrate content. Thus, the rate of denitrification during endogenous respiration was determined. Further, wastewater was added to the sludge (anoxic conditions were preserved) - simulation of nitrate recycling. According to the results of experimental studies, the calculated denitrification rates were 

\[ r_{ds} = 0.71 \pm 0.04 \text{ mg N-NO}_3 / \text{mg of active heterotrophic microorganisms per day on easily oxidizable substrate}, \]

\[ r_{de} = 0.12 \pm 0.02 \text{ mg N-NO}_3 / \text{mg of active heterotrophic microorganisms per day on difficult oxidizable substrate}, \]

\[ r_{dp} = 0.12 \pm 0.01 \text{ mg N-NO}_3 / \text{mg of active heterotrophic microorganisms per day}. \]

1. Introduction

In large cities, the pollution problem of natural waters with biogenic elements becomes more crucial. Increasing an amount of phytoplankton in water bodies, which is the first step of water pollution, as well as the process of water bodies eutrophication, is associated with this problem [1]. A significant part of biogenic elements, including nitrogen compounds, go into reservoirs with wastewater. Nowadays, nitrogen removing is one of the most important tasks of wastewater treatment [2,3].

2. Theoretical part

Nitrogen removal from wastewater occurs as a result of the biological oxidation of ammonium nitrogen to nitrates and then the oxidation of nitrates to nitrites (nitrification processes), followed by denitrification, that is, the reduction of nitrites and nitrates to nitrogen gas [4]. Part of the nitrogen consumed in the synthesis of bacteria cell. In parallel, the processes of lysis and self-oxidation of bacterial cells take place, as a result of which organic nitrogen of microbial cells is transferred to ammonium nitrogen. But the main methods of nitrogen removal from wastewater are the nitrification and denitrification processes, which require the creation of specific conditions directly in biological treatment facilities. [1,4-6].
In the process of vital activity of microorganisms, nitrates can be reduced to ammonia or molecular nitrogen. It is called nitrate reduction. There are assimilation nitrate reduction and dissimilation nitrate reduction [7]. Assimilation nitrate reduction is carried out by prokaryotes and eukaryotes by consuming nitrate as a source of nitrogen for cell synthesis:

\[
\text{NO}_3^- \rightarrow \text{NO}_2^- \rightarrow x \rightarrow \text{NH}_2\text{OH} \rightarrow \text{organic nitrogen}
\]  

(1)

This process can take place both in aerobic and anoxic conditions. Dissimilatory nitrate reduction (denitrification) is carried out in anoxic conditions by denitrifying bacteria. Denitrification is a microbiological process of reduction of oxidized nitrogen compounds (nitrates, nitrates) to molecular nitrogen (in some cases to \(N_2O\) [8]) and the oxidation of organic matter to carbon dioxide. The denitrification process is carried out by a large group of heterotrophic optional anaerobic bacteria [9-12], many of which can be used as other oxidized nitrogen forms, \(NO_2\), \(NO\), and \(N_2O\), as electron acceptors.

The denitrification process (dissimilative nitrate reduction) is described by the following scheme:

\[
\text{NO}_3^- \rightarrow \text{NO}_2^- \rightarrow \text{NO}^- \rightarrow \text{N}_2\text{O} \rightarrow \text{N}_2
\]  

(2)

Optional heterotrophic bacteria implement the denitrification process during biological wastewater treatment [10]. Although these microorganisms prefer aerobic respiration, and in case of a lack or absence of dissolved oxygen, they use nitrates or nitrates in the presence of a sufficient amount of an organic substrate [5,13].

The enzymatic apparatus of the denitrification process is formed only in the absence of dissolved oxygen or its low concentration. During the process of denitrification, dissolved oxygen is an inhibitor of this process, and under aerobic conditions, all bacteria, which are capable of denitrification, prefer oxygen respiration [14].

For urban wastewater, the denitrification process can be described by the following summary equation [5,15]:

\[
\frac{1}{70} \text{C}_{18}\text{H}_{19}\text{O}_9\text{N}^+ + \frac{1}{5} \text{NO}_3^- + \frac{1}{5} \text{H}^+ \rightarrow \frac{1}{10} \text{N}_2 + \frac{17}{70} \text{HCO}_3^- + \frac{1}{70} \text{NH}_4^+ + \frac{1}{5} \text{H}_2\text{O}
\]  

(3)

However, it should be taken in mind that the composition of urban wastewater may vary widely, and the formula of integral organic matter used in reaction (3) may also have a different appearance.

In theory, the COD of any organic substance described structural formula \(C_xH_yO_z\) determined from oxidation and recovery reactions:

**Oxidation reaction** [5]:

\[
\text{C}_x\text{H}_y\text{O}_z + (2x-z)\text{H}_2\text{O} \rightarrow x\text{CO}_2 + (4x+y-2z)\text{H}^+ + (4x+y-2z)e^-
\]  

(4)

**Recovery reaction:**

\[
\frac{1}{4} \text{O}_2 + \text{H}^+ + e^- \rightarrow \frac{1}{2} \text{H}_2\text{O}
\]  

(5)

According to reactions (4) and (5), we determine:

\[
\text{C}_x\text{H}_y\text{O}_z + \frac{1}{4}(4x+y-2z)\text{O}_2 \rightarrow x\text{CO}_2 + \frac{3}{2} \text{H}_2\text{O}
\]  

(6)

3. Materials and methods

The main problem of the ideal realization of the denitrification process in real facilities is to provide strictly anoxic conditions (the complete absence of dissolved oxygen) in the denitrification zone [1, 16].
Dissolved oxygen enters the anoxic zone, both with recycled activated sludge recycling and with “nitrate” recycling. In this case, part of the organic compounds entering the anoxic zone with wastewater will be oxidized with dissolved oxygen coming from recycling. First of all, easily oxidized organic substances will be oxidized.

Nowadays there are a number of recognized and approved models for describing denitrification processes, which are used both in the design of structures and in their operation. The most common of these are the dependencies of the type of Mono reactions, as well as calculations based on the load of organic matter on activated sludge, and reactions of kinetics of zero order in the concentration of nitrates.

The Refling-Stensla equation describes the pace of the denitrification process, which depend on the organic load:

\[ r_{\text{anox}} = 0.03 \frac{S_S}{X} + 0.029 \]  

where \( X \) – total mass of microorganisms in the system; \( S_S \) – concentration of biodegradable substrate in the incoming waste water.

This equation was obtained from pilot studies [5] of specific denitrification rates as a function of the load on active sludge. However, this equation is a statistical model, which is based on the determination of an empirical relationship between denitrification rate of the load of organic substances in the activated sludge, based on a number of the experiments [16].

The analysis of a sufficient data set under specific conditions (time of year, load on the structure, technological parameters of the process) allows to obtain the minimum deviation of the calculated and real data in statistical models. However, this class of models allows to predict the results only for specific facilities operating under the existing conditions. Thus, to use equation (7) when calculating the denitrification process, it is necessary to conduct a series of experiments to determine the dependence of the denitrification rate on the load on activated sludge by organic matter and to obtain constants for specific wastewater [17].

4. Results

The denitrification rates of sludge were determined on an experimental equipment when conducting a series of experiments, where the source of the substrate was real wastewater entering the aerotanks of one of the sewage treatment facilities in the Udmurt Republic.

The rates of reduction of nitrate nitrogen were determined indirectly, by analytically controlling the concentration of the substance in the experimental reactor over time. At the same time, in the experimental reactor, before starting the experiment, the initial waste water was added to the sludge mixture (with a known concentration of nitrates). After that, the concentration of nitrate nitrogen in the filtered mixture was analyzed for several tens of minutes. The difference in concentrations, reduced to a unit of time and concentration of heterotrophic biomass (concentration in a series of experiments remained constant – 0.428 kg/m³), determined the desired value:

\[ r_{\text{anox}} = \frac{C_{2,\text{NO}_3} - C_{3,\text{NO}_3}}{(t_2 - t_1) \cdot X_v} \]  

where \( X_v \) – organic part of activated sludge.

Before the start of the experiment, the sludge from the experimental equipment was blown off for several hours in order to complete the oxidation of adsorbed contaminants. At the end of the blowing off, the concentration of nitrate nitrogen was determined in the mixture [18, 19].

At the beginning of the experiment, the sludge was under anoxic conditions (the mixture was mechanically mixed without aeration), while the mixture was periodically analyzed for nitrate content. Thus, the rate of denitrification during endogenous respiration was determined (see Fig. 1).
Figure 1. Denitrification rate with endogenous respiration.

Further, waste water was added to the sludge (anoxic conditions were preserved), while there was a sharp increase in the concentration of nitrates [20], as can be seen in fig. 2. In the process of observation, samples of the sludge mixture were also taken, which were analyzed for nitrate content.

Figure 2. Denitrification rate after reconstitution of nitrate reduction.

5. Conclusions
According to the results of the determinations, the estimated denitrification rates were:
- on easily oxidizable substrate $r_{ds} = 0.71 \pm 0.04$ mg \textit{N-NO}_3/mg active heterotrophic microorganisms per day;
- on endogenous respiration $r_{de} = 0.12 \pm 0.02 \text{ mg N-NO}_3/\text{mg active heterotrophic microorganisms per day;}$
- on a non-oxidizable substrate $r_{dp} = 0.12 \pm 0.01 \text{ mg N-NO}_3/\text{mg active heterotrophic microorganisms per day.}$

The obtained denitrification rates for these wastewaters will allow us to calculate the volumes of anoxic and aerobic structures. (during increasing time of the sludge mixture in anoxic conditions, it becomes necessary to proportionally increase the volume of aerobic structures, in order to maintain the growth rate of autotrophs).

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