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# TWO TYPES OF THE NITROGEN FIXATION BY MICROBIAL ORGANISMS IN RIVER WATERS

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### Introduction

The presence of ammonium in natural water stimulates the increase of algae and heterotrophic autotrophic bacteria activities. The ammonia and its derivative environmental amines are physiological products, which are released through a wide range of bacteria catalytic systems and natural redox processes. The dynamics of ammonium oxidation may serve as an indicator of the state and potential of self purification of the aquatic environment. By laboratory simulations two types of fixations by microbial organisms were established: 1) absorption-desorption, the hydromicrobiotic reaction to ammonium (*HMBRA*) at the instantaneous increase of the concentration of ammonium ion in the river water (so-called shock/stress effect); 2) nitrogen fixation stimulated by a certain concentration (0.05mg/L) of 1-naphthylamine (1-NA) and other amines.

The aim of the paper was to investigate two types of the nitrogen fixation by microbial organisms in natural waters.

## Materials and methods

The used physicochemical methods were previously described by ISOs and are based on the sensitivity of the soluble nitrogen species ( $NH_4^+$ ,  $NO_2^-$  and  $NO_3^-$ ) to the influence of the microbial enzymatic system, which, in turn, is a function of the chemical composition of natural water and, in particular, of organic matter from natural water (river, lake, sewage basin, etc.). All parameters of the models were similar (temperature, atmospheric pressure, daylight) to those of the studied aquatic objects from which the water sample was collected for the laboratory model. At the initial stage of the laboratory simulation process a small amount of  $NH_4^+$  was added. The water of the Nistru River at Vadul-lui-Voda section was used.  $NH_4Cl$  solution was added to the river water samples, in order to achieve two concentrations of about 3 mg/L and about 6 mg/L of ammonium ions. The reference sample contained only ammonium ions, while the working samples contained also amines (1-NA, DPA, DEA). Sample testing was completed using the HACH DR/2500 Spectrophotometer and spectroscopy using Perkin Elmer Lambda 25.

## **Results and conclusions**

Two types of the nitrogen fixation by microbial organisms in natural waters, investigated in the model experiments using river waters, were: 1) adding a certain

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amount of  $NH_4^+$  and 2) adding additionally exact amount of 0.05 mg/L of 1-NA to amplify the generation nitrogen-fixing species in river waters.

In the case of the first type of stimulation, the total soluble mineral forms of nitrogen are increasing during the period of nitrogen fixation (starting on second day and continuing on third day) after adding 2 or 3 mg/L. On the other hand, when 6 mg/L of ammonium ions were added, the effect of nitrogen fixation by microbial organisms disappeared thoroughly.

Some microbial populations use ammonium nitrogen compounds for its growing and another use them for maintaining their live through redox process of nitrogen species. The fixation and redox processes of nitrogen-containing compounds take place together, stimulating the selection of microbial population. The analysis of the NH4<sup>+</sup> concentration difference (in mg/L) for various number of days was performed. In all analyzed samples the adsorption of ammonium ions prevailed essentially over other bio-chemical equilibrium during the first day of the experiments. In the second day, the NH<sub>4</sub><sup>+</sup> concentration exceeded the initial value, and then decreased, for the samples of 0.25, 0.1, 0.05 and 0.025 mg/L, which indicated to a drop of the ammonium concentration due to possible adsorption and redox processes. The increase of the NH<sub>4</sub><sup>+</sup> concentration was maximum for 0.05 mg/L of 1-NA sample. Simultaneously. beside the stimulation of nitrogen fixation. the consumption/oxidation of ammonium ions was less significant during the third day comparing to the second day. On the sixth day, the rate of the NH4<sup>+</sup> oxidation/consumption process exceeded that of nitrogen fixation. The ammonium ion concentration dynamics as a function of the 1-NA concentration do point out to the fact that the nitrogen fixation process was still continuing.

The analysis of the natural water model behavior as a function of 1-NA concentration at initial concentration of 6 mg/L NH<sub>4</sub><sup>+</sup> was performed. The determined value of 1-NA concentration, at which it is supposed to stop completely the processes connected to the biochemical production and adsorption of the NH<sub>4</sub><sup>+</sup>, due to its toxicity, was equal to about 0.70 mg/L. The process of decreasing of ammonium concentration on the first day after the initiation of laboratory simulations, and its increasing on the second day, caused by aquatic microorganisms, could be called the hydro-micro-biotic reaction to ammonium (*HMBRA*).

By laboratory simulations, two types of fixations by microbial organisms were distinguished: 1) Ammonium absorption-desorption, e.g. *HMBRA*, at the instantaneous increase in the NH<sub>4</sub><sup>+</sup> concentration in the river waters (so-called shock/stress effect); 2) Nitrogen fixation stimulated by a certain concentration (0.05mg/L) of 1-NA and other analyzed amines. These modifications constituted a sensitive reaction of aquatic microorganisms to environmental changes. A noticeable effect of the attenuation of nitrication processes caused by the toxicity of ammonium derivatives was evidenced.

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