1. Research project objectives

Recently, other phenomena of anaerobic oxidation of ammonium nitrogen (NH4-N) have been discovered than anammox (Fdz-Polanco, 2001). Anaerobic ammonia oxidizing bacteria (AAOB) have been found to have a more comprehensive metabolism than expected. **In addition to the commonly known electron acceptor in the form of nitrite nitrogen (NO_2-N), there may be others for oxidation of NH_4-N under anaerobic conditions (Zandt et al., 2018). One of them is sulfate (SO_4-S) in the sulfammox process (Bi et al., 2020).** Sulfammox proceeds according to the reaction (Zhang et al., 2019) (1):

$$SO_4^{2-} + 2NH_4^+ \rightarrow S^0 + N_2 + 4H_2O \qquad (1)$$

Compared to the conventional anammox process, sulfammox is easy to handle and the cost of nitriding can be saved because SO_4-S serves as an electron acceptor instead of NO_2-N, it is free from secondary impurities caused by sulfides (S^{2-}) (Zhang et al., 2019). In addition, S^0 is formed in this process, and its recovery is a valuable by-product of the process (Rios-Del Toro and Cervantes, 2019). **The efficiency of simultaneous removal of NH_4-N and SO_4-S from wastewater under anaerobic conditions is still, however, low - 10-55% and 0-80% respectively (Bi et al., 2020, Przywara, 2019, Wang et al., 2017a). Furthermore, the literature provides no reports on the sulfammox process with granular sludge in sequencing batch reactor (SBR). It should be emphasized that none of the authors used the test results to optimize the performance of systems using sulfammox.** Mathematical (mechanistic) modeling and computer simulation are effective and reliable analytical tools to understand both mechanisms driving the dynamics of these systems and the interaction between current groups of microorganisms.

The aim of the project is to identify, characterize and model the sulfammox process compared to the conventional anammox process, as well as their mutual effect on the efficiency of NH_4-N oxidation in the SBR. The research is intended to answer the question whether it is possible to replace the anammox process with the sulfammox process.

2. Research methodology

The tests will be carried out for 3 years (1095 days) on a continuous basis. During the entire test period, the SBR will operate at a constant temperature of 30 (± 1) ° C. The pH will be controlled in the range of 7.5-7.8 by the automatic addition of 6 M hydrochloric acid (HCl). DO concentration in unventilated SBR will not exceed 0.2 mg/l. SBR will be fed with a synthetic substrate according to Dapena-Mora (2004). The most important ingredients, i.e. nitrite, ammonium and sulfate, will be supplied in the form of NH_4Cl, $NaNO_2$ and $MgSO_4$ respectively. The NH_4/SO_4 ratio in the sulfammox process without the addition of NO_2-N will be changed in the range of 0.5 - 3 at a constant C/N ratio (under heterotrophic conditions).

Analysis of the structure of microorganism communities involved in the sulfammox process will be performed using 16S rRNA gene High-throughput Sequencing.

The concentration of COD, NO_3-N, NO_2-N, NH_4-N, SO_4-S compounds was determined using a spectrophotometer, DR 3900 using cuvette tests from Hach Lange GmbH (Dusseldorf, Germany) for this type of analysis. The determination of total sulphur will be performed with the use of microwave plasma atomic emission spectrometer 4210 MP-AES supplied by Agilent.

The latest version of GPS-X (Hydromantis, Canada) will be used as a simulation environment (GPS-X has been used by the GUT research team for 18 years).

3. Justification for tackling specific scientific problems by the proposed project

Sulfammox is a highly efficient and economical alternative to the biological removal of NH_4-N and SO_4-S using wastewater treatment compared to the traditional separate removal of NH_4-N and SO_4-S. NH_4-N in the natural environment may cause eutrophication of water and deterioration of water quality, in turn high concentrations of SO_4-S in wastewater, imbalance in the natural sulfur cycle of water bodies. There is an increasing amount of pollutants such as NH_4-N and SO_4-S (> 1000 mg / l both) in the wastewater (Rikmann et al., 2016). Thus, in the case of wastewater with a high content of N, SO_4-S and organic substances, the simultaneous removal of COD and N can be achieved in anaerobic conditions, while avoiding the accumulation of toxic and harmful to human health and S2- ecosystems. This prevents process disturbance caused by S^{2-} braking.