

Zborowska, E., Muszynski, A., Lebowska, M., Podedworna, J., Zubrowska-Sudol, M. Qualitative Determination of Polyphosphate Accumulating Bacteria in Activated Sludge. *Ochrona Srodowiska* 2010, Vol. 32, No. 2, pp. 9–14.

**Abstract:** The aim of this work was to isolate and identify the polyphosphate accumulating organisms (PAOs) that populate a sequencing batch reactor (SBR) designed for the removal of orthophosphates from wastewater under laboratory conditions. In order to isolate bacteria with potential capacity for polyphosphate accumulation, the sludge samples were subjected to aerobic starvation, and the solid media used for cultivation were supplied with dicyclohexylcarbodiimide (DCCD), an ATP synthesis inhibitor. The isolated bacterial strains, as well as the mixed cultures, were then tested in liquid media to confirm the typical PAO phenotype. Each test consisted of five alternate phases – three anaerobic and two aerobic. The nine-month experiment produced the following findings. Gram-negative rods dominated the activated sludge samples, but Gram-positive rods, mainly those of the genus *Arthrobacter* sp., were also in abundance. The micrococci isolated from the sludge were predominantly of the genus *Staphylococcus* sp. Most of the strains that dominated the cultures subjected to aerobic starvation were of the genera *Arthrobacter* sp., *Aeromonas* sp. and *Brevundimonas* sp. The pure bacterial strains that were tested in the liquid media to confirm the typical PAO phenotype showed no capacity either for the uptake of orthophosphates from the wastewater under aerobic conditions, or for the utilization of organic compounds with simultaneous release of orthophosphates under conditions of oxygen deficiency. Seemingly, the absence of other microorganisms with the capacity for synergetic interactions was the underlying cause of the pure strains' failure to share the typical PAO phenotype. The lack of capacity for the uptake and release of orthophosphates was also observed in the mixed cultures. The test performed with activated sludge produced a positive result, typical of the PAO model, which substantiates the presence of PAO in the activated sludge being tested. However, it may as well be assumed that the isolated strains were not PAOs.

**Keywords:** Polyphosphate accumulating organisms (PAO), activated sludge, sequencing batch reactor (SBR), cultivation methods.