Characterization of microbial nitrate uptake for the treatment of agricultural wastewater

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Although nitrate is an essential nutrient for the growth and reproduction of all plants, the excess nitrate causes serious physiological problems in plant growth, known as 'salt stress'. In order to remove the excess amount of nitrate from cultivating soils, we have isolated soil microorganisms which have a high capacity of nitrate uptake. These microorganisms were found to be also useful for the removal of nitrate from wastewaters. The amount of wastewater, such as household sewage, livestock wastewater, and food wastewater, is increasing every year and regulations of ocean disposal become active at 2012 in Korea. Livestock wastewaters are required to be treated by wastewater treatment plants. To increase the efficiency of wastewater treatment, microorganisms were used for the removal of various chemicals, hydrocarbons, and heavy metals. Microbial applications were also useful for the removal of nitrogen compounds from the livestock wastewaters. In this study, microorganisms were isolated from livestock farms as well as upland soils and their nitrate uptake activities were investigated. We have isolated three microorganisms from a hog farm and four microorganisms from a poultry farm. Nitrate uptake activities of these microorganisms were excellent compared with those of soil microorganisms, *Enterobacter* species and *Bacillus* species. All these soil bacteria have been demonstrated to remove nitrate of 2,000-5,000 ppm. The characteristics of nitrogen metabolism were investigated and it was found that those isolated from wastewaters showed pH-dependence on the growth and nitrate uptake. In the range of pH 5 to 8, both bacterial growth and nitrate uptake were increased from 1,000 to 5,000 ppm. Interestingly, at pH 9, both were maximal but delayed by 2 h. At pH 9, both bacterial growth and nitrate uptake were suppressed for 4 h during the beginning of incubation and then they were rapidly increased to maximal levels after 6 h. This may be due to the precipitation of Mg^{2+} at alkaline pH as $Mg_3(PO_4)_2$, suggesting that nitrate transport enzyme in these bacteria is Mg^{2+}-sensitive. The delayed increase in nitrate uptake may be explained by pH decrease in culture media during cultivation and thus solubilization of precipitate. When the medium was buffered with 20 mM phosphate, the nitrate uptake was completely inhibited at 0 mM Mg^{2+} and was maximal at 2 mM Mg^{2+}. In these bacteria, nitrate uptake is mediated by an enzyme sensitive to pH and Mg^{2+}. The enzyme is inhibited at pH 9 and requires 2 mM Mg^{2+}.