

Denitrifying activity of activated sludge in suspension and in biofilm

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Denitrification consists in the reduction of oxidized nitrogen compounds like nitrite or nitrate to gaseous nitrogen compounds. Most of biological denitrification processes rely on heterotrophic microorganisms and therefore the use of methanol, ethanol, glucose, citrate or acetate as a carbon source is needed. Activated sludge is currently the most widely used process for the treatment of both domestic and industrial wastewaters. Attached to a support sludge microorganisms develop an active thin layer known as biofilm, which has many advantages like high biomass concentration and resistance to short-term toxic loads when compared with suspended growth processes. The aim of the present work is to compare the denitrifying activity of an adapted consortium of activated sludge in the form of planktonic cells with its biofilm form.

A volume of concentrated sludge was collected from an activated sludge tank from a wastewater treatment plant and was acclimatized in a denitrification medium as reported by Beaubien et al. (1995), using acetate as a carbon source, a ratio of C/N=2 and a phosphorus concentration of 10 mg P/L, during 3 weeks, at room temperature. The adapted activated sludge inoculum was grown in biofilm form in an anoxic rotating biological contactor (AnRBC). The AnRBC single-stage system consisted of 8 polymethylmethacrylate discs (diameter = 130 mm, thickness = 3 mm, 20 mm interspace) mounted on a horizontal shaft, having a working volume of 2.5 L. The submergence of the discs was 70%. The rotational speed was 4 rpm and the temperature was maintained at 28 °C by means of a heating jacket. The reactor was inoculated with 2.5 L of the adapted consortium of sludge in batch mode for 4 days. After this period, the reactor was fed continuously with the synthetic denitrifying medium using acetate as carbon source at a ratio C/N=2 and a phosphorus concentration of 10 mg P/L. To allow biofilm formation the hydraulic retention time (τ) was gradually reduced to a final value of 5.68 h and the organic and nitrate loads were doubled at the 8th day of operation. The total period of operation was 25 days. Denitrifying activity tests were performed in 160 mL serum flasks, containing 90 mL of denitrifying medium. In sludge activity tests, each flask was inoculated with 6 mL of adapted biomass (3.0±0.3 g TSS/L). The biofilm activity assays were performed with 1 g of biofilm (wet weight) added to each flask. The biofilm samples were collected from the discs of the AnRBC at the end of operation. To obtain anoxic conditions, the flasks were flushed with helium gas. Finally flasks were incubated at 28 °C at 150 rpm and aliquots of 2.5 mL were collected every hour and immediately analysed for various parameters. Solids were measured on a spectrophotometer at 660 nm. Samples were then filtered over a 0.2 µm membrane filter and used for nitrite, acetate and nitrate quantification according to the *Standard Methods of Analysis* (APHA, 1992). All the assays were done in triplicate.

The activity of the adapted consortium of activated sludge when grown in suspension and in biofilm form is expressed as nitrate and acetate uptake rate and presented in Table 1, as well as biomass yield. It has to be noted that the determined values for biofilm are an average of the values obtained for biofilm samples collected along the reactor.

Table 1

Biomass yield, specific acetate and nitrate consumption rates for planktonic and biofilm cells of activated sludge.

Parameter	Inoculum	Planktonic cells	Biofilm
Biomass yield (g TSS/g acet)		0.6381± 0.0697	0.0793±0.0150
Specific acetate consumption rate (g CH ₃ COO ⁻ /g TSS.h)		0.0298±0.0036	0.1375±0.0126
Specific nitrate consumption rate (g NO ₃ ⁻ /g TSS.h)		0.0510±0.0077	0.1284±0.0147

These results show that specific activity in terms of acetate and nitrate consumption rate was significantly higher for cells in biofilm form. This is in accordance with the results of several authors who have reported that bacteria associated with a solid surface exhibit higher uptake rates. This behaviour can be justified based upon the fact that bacteria adhering to a surface establish strong relationships between them and with the support developing “self-defences”. Considering the biomass yield, free-living cells reveal higher growth. This is also in accordance with expected results and one reason for this is that fixed-cells direct the substratum for the production of extracellular polymeric substances in detriment of the production of cells.

This elicits the conclusion that a biofilm based process can be more efficient in the treatment of wastewater with high nitrate loads.

References

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