

SHORT
COMMUNICATIONS

Transformation of Cellulose Nitroester by the Sulfate-Reducing Bacterium *Desulfovibrio desulfuricans*

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Anaerobic sulfate-reducing bacteria are widespread in various ecological niches and, due to their metabolic flexibility, are involved in the degradation of numerous nonnatural organic compounds. Bacteria of the genus *Desulfovibrio* are known to possess a unique ability to metabolize such nitroaromatic compounds as trinitrotoluene, trinitrobenzene, nitrophenol, tetryl, and others [1–3]. The transformation of nitrogen-containing compounds involves the reduction of the nitro group followed by reductive deamination [3]. In some sulfate-reducing bacteria, active nitrate and nitrite reductases were revealed [4].

This paper presents data on the ability of *Desulfovibrio desulfuricans* to transform nitroester of cellulose (NC), an industrial waste resistant to biodegradation.

Strain *D. desulfuricans* VKM 1388 was obtained from the All-Russia Collection of Microorganisms (Pushchino, Russia). Postgate medium B [5] supplemented with calcium lactate (3.5 g/l) as an organic substrate and Na₂S (100 mg/l) as a reducing agent was used for anaerobic cultivation performed as described earlier [6]. In experimental variants, 12 g/l of NC with a high nitrogen content (11.8%) was added to the medium; in control, bacteria were grown in Postgate medium B without NC. The amount of NC was determined gravimetrically after the extraction from the culture liquid with acetone; nitrates were analyzed by a standard method [7]. Biomass was determined from the protein content, which was measured by a modified Lowry method [8]. The data presented are averages of three replicate experiments, in which every parameter was measured in triplicate.

In medium with NC, the lag phase of *D. desulfuricans* growth somewhat increased (Fig. 1). The specific growth rate of bacteria in the control medium was 1.5-fold greater than that in the presence of NC (0.074 and 0.05 h⁻¹, respectively). The biomass accumulation in the NC-containing medium was comparable to that in the control (160 and 130 mg protein/l, respectively). A decrease in the NC concentration in the medium became noticeable by the 8th day of bacterium growth;

a 10% decrease was observed after 14 days of cultivation. The transformation of NC by *D. desulfuricans* was accompanied by the appearance of nitrates in the culture liquid, whose amount peaked by the 8th day (220 µg/ml) and then decreased sharply (Fig. 2). In the medium without NC, nitrates occurred in trace amounts.

To verify the possibility of spontaneous denitration of NC [9], which could be initiated by hydrogen sulfide, a metabolite formed during the reduction of sulfates by *D. desulfuricans*, we studied the abiotic (chemical) denitration of NC. A three-day culture was autoclaved (0.5 atm, 20 min), supplemented with NC (0.5 g/100 ml), and incubated at 30°C for 20 days. In this case, the nitrate content in the autoclaved liquid was found to increase to 70 µg/ml, whereas NC concentration remained unaltered. Most likely, metabolites produced by sulfate-reducing bacteria changed the level of nitrocellulose esterification but produced no effect on the NC concentration in the medium. Thus, the transformation of NC by living cells revealed in our experiments is of a biogenic nature.

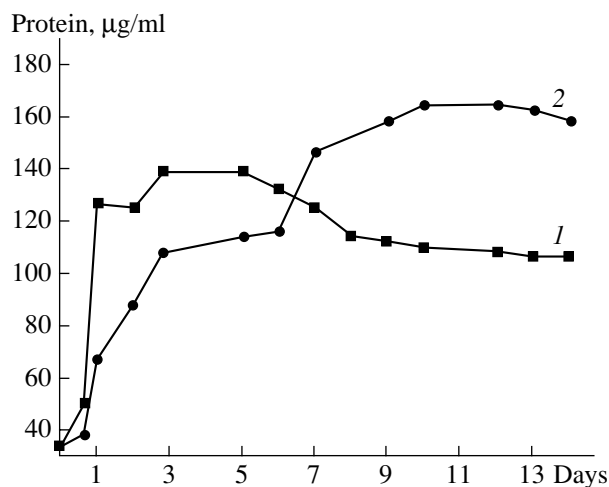


Fig. 1. Curves of growth of *D. desulfuricans* in (1) Postgate medium B and (2) the same medium supplemented with NC.

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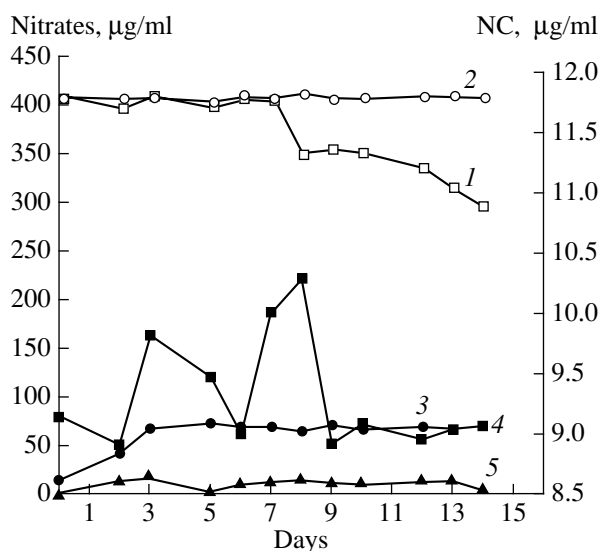


Fig. 2. Time course of NC concentration in Postgate medium B in the presence of (1) growing and (2) killed cells of *D. desulfuricans*; nitrate concentration dynamics in the presence of (3) killed and (4) growing cells. Curve 5 shows nitrate concentration in Postgate medium B without NC.

To conclude, the decrease in the NC concentration and the formation and subsequent disappearance of nitrates during growth of *D. desulfuricans* in NC-containing medium show that this bacterium possesses a biochemical potential for the transformation of nitrocellulose.

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