



Simultaneous heterotrophic nitrification and aerobic denitrification by bacterium *Rhodococcus* sp. CPZ24

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ABSTRACT

Rhodococcus sp. CPZ24 was isolated from swine wastewater and identified. Batch (0.25 L flask) experiments of nitrogen removal under aerobic growth conditions showed complete removal of 50 mg L⁻¹ ammonium nitrogen within 20 h, while nitrate nitrogen removal reached 67%. A bioreactor (50 L) was used to further assess the heterotrophic nitrification and aerobic denitrification abilities of *Rhodococcus* sp. CPZ24. The results showed that 85% of the ammonium nitrogen (100 mg L⁻¹) was transformed to nitrification products (NO₃⁻-N and NO₂⁻-N) (13%), intracellular nitrogen (24%), and gaseous denitrification products (48%) within 25 h. The ammonium nitrogen removal rate was 3.4 mg L⁻¹ h⁻¹. The results indicate that the strain CPZ24 carries out simultaneous nitrification and denitrification, demonstrating a potential use of the strain for wastewater treatment.

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1. Introduction

Nitrogen (N) is a key nutrient for plants. The N cycle is a critical biogeochemical cycle in aquatic and terrestrial ecosystems. However, a series of environmental consequences has been resulted from the massive increases in fixed N within the aquatic environment (Conley et al., 2009), e.g., eutrophication problems in waterbodies and high nitrate concentrations in drinking water. Those problems are largely caused by livestock and poultry industries (Zhang, 2009). In China, the livestock and poultry industries produce approximately 173 billion tons of manure per year and an estimated 1.6 billion tons of nitrogen per year (Zhou, 2005). Furthermore, most of livestock manure runs out directly into fresh water from livestock houses and so has seriously degraded local water quality. Controlling and reducing nitrogen content in wastewater from livestock farming prior to discharge and developing appropriate nitrogen management technologies are critical for the quality of water systems. Biotreatment is an effective and low-cost technology for reducing the nitrogen content in the wastewater. Biological nitrification–denitrification is one of the most economical processes for nitrogen removal from wastewaters (Gupta and Gupta, 2001).

Inorganic nitrogen can be removed traditionally by both autotrophic nitrifying and heterotrophic denitrifying microorganisms (Fu et al., 2009). Such strategies require separate treatment processes and strict treatment condition control, making these technologies

prohibitively expensive (Khardenavis et al., 2007; Kim et al., 2005). Simultaneous nitrification and denitrification (SND) is an attractive method to treat wastewater. It implies that nitrification and denitrification occur concurrently in the same reaction vessel under identical overall operating conditions (Schmidt et al., 2003). Recent studies showed that most heterotrophic-nitrifying bacteria are capable of aerobic denitrification, including *Alcaligenes faecalis* (Joo et al., 2007), *Pseudomonas stutzeri* (Su et al., 2001), *Microvirgula aerodentificans* (Patureau et al., 2001), *Alcaligenes faecalis* (Joo et al., 2007), *P. putida* (Kim et al., 2008), and *Acinetobacter calcoaceticus* (Zhao et al., 2010a,b).

Rhodococcus species being capable of heterotrophic nitrification have been studied (Zhang et al., 2003). However, little has been examined about the removal of ammonium from wastewater through SND by *Rhodococcus* bacteria. In this study, *Rhodococcus* sp. CPZ24 was isolated from swine wastewater and shown the ability of heterotrophic nitrification and aerobic denitrification not only in batch flasks, but also in bioreactor settings. Moreover, nitrogen gas from the bioreactor was detected to confirm SND by *Rhodococcus* sp. CPZ24.

2. Methods

2.1. Sampling and culture medium preparation

Six wastewater samples were collected from swine wastewater ponds in Xiaoshan County, Zhejiang Province, China in April 2008

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