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Individual and combined effects of nickel and copper on nitrification organisms

Sukru Aslan*, Onur Sozudogru

Department of Environmental Engineering, Cumhuriyet University, 58140 Sivas, Turkey

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ABSTRACT

Individual and combined effects of Ni^{2+} and Cu^{2+} concentrations on the nitrification process were investigated in a submerged biofilter. The activity of nitrification organisms was inhibited by high concentrations of Cu^{2+}. However, low concentrations of Cu^{2+} promoted NOB activity. There were no observable stimulation effects of Ni^{2+}. The presence of 0.2–4.0 mg/L Ni^{2+} caused a decrease in the rate of nitrification. Although the inhibition of Cu^{2+} on the nitrification organisms started at a concentration of 0.5 mg/L, it started at 0.2 mg/L for Ni^{2+}. Ni^{2+} had stronger inhibitory effect on the nitrification organisms than Cu^{2+}. Using a mixture of Ni^{2+} and Cu^{2+}, inhibition level drastically increased to 32% and 49% at concentrations of 2.0 and 3.0 mg/L, respectively. At the same concentrations, individual inhibition by Ni^{2+} and Cu^{2+} was much lower, about 12–15% and 18–24%, respectively.

1. Introduction

Biological nitrification is a widely used process in wastewater treatment facilities. Compared to suspended growth systems, biofilm systems have a longer sludge retention time (SRT), which favors slow growth organisms like nitrifiers. The biofilm system is also less sensitive to toxic compounds such as heavy metals (Kocamemi and Cecen, 2007; Weon et al., 2004).

An aerobic autotrophic bacteria is responsible for nitrification in biological processes. In the process, $NH_4{}^+$-N is first oxidized into NO_2-N by *nitrosomonas* sp. (ammonium oxidizing bacteria, AOB) and then NO_2-N is rapidly oxidized to NO_3-N by *nitrobacter* sp. (nitrite oxidizing bacteria, NOB) (Aslan and Gurbuz, 2011). Nitrification organisms have slow growth rates and they are very sensitive to conditions such as pH, dissolved oxygen (DO) concentrations, temperature, and toxic chemicals.

Wastewater commonly contains heavy metals which usually reduce treatment plants' efficiency (Lester, 1983). However, some heavy metals (like Fe, Cu, Co, Ni, Zn) are considered to be essential elements for microbial growth (Gikas, 2007). Low concentrations of essential heavy metals stimulate the activity of microorganisms and increase the efficiency of wastewater treatment plants (Aslan and Gurbuz, 2011).

Heavy metals in the effluent waters are of great concern due to the toxic effects on the environment. The toxicity of heavy metals on the biological processes depends on the metal speciation, metal concentrations, microbial populations (autotrophic and heterotrophic microorganisms), SRT (in the suspended growth process), and reactor types (suspended or biofilm system) (Ong et al., 2010; Semerci and Cecen, 2006; Stasinakis et al., 2003). Of the operational and environmental conditions, speciation and concentrations of metals are considered the most important factors (Semerci and Cecen, 2006).

Heavy metals are known to inhibit the activity of biological organisms. However, the activity of microorganisms is enhanced by heavy metals at relatively low concentrations (Aslan and Gurbuz, 2011, 2014; Yetis et al., 1999) Gikas (2008) reported that simultaneous interactions between multi-heavy metals and microorganisms might have synergetic, antagonistic or additive effects. Recent experimental studies indicated that acclimatization enhances the tolerance of microorganisms to toxic concentrations of heavy metals (Mertoglu et al., 2008; Ozbelge et al., 2007).

Many experimental results have been published on the effects of single heavy metal to the activity of microorganisms (Dilek and Gokcay, 1996; Sirianuntapiboon and Boonchupleing, 2009; Stasinakis et al., 2002; Yetis et al., 1999). However, very little work has been published on the simultaneous effects of the combination of Ni^{2+} and Cu^{2+} on the performance of nitrification process. Gikas (2008) reported that the combined effects of multi-heavy metals to a microbial population could be greater than the sum of the effects of each heavy metal individually. Nickel and copper are both

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* Corresponding author.
E-mail address: saslan@cumhuriyet.edu.tr (S. Aslan).

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Table 1
Operational conditions of experimental set-up.

Operational conditions of SBFR	
Active height of SBFR(cm)	20
Inner diameter of SBFR (cm)	10
Total volume (L)	1.5
Water volume (L)	1.4
Length of support material for bacterial growth (m)	2.65
Surface area of support material for bacterial growth (m^2)	0.33
Wastewater flow rate (L/day)	6.9
Initial wastewater pH	7.50
Hydraulic retention time (h)	4.9

encountered in untreated wastewater from electroplating, mining, smelting, and metal-finishing industries as well as city wastewater (Weon et al., 2004). In the present study, effects of the individual and combined presence of Ni^{2+} and Cu^{2+} on the activity of nitrification organisms were investigated at various metal concentrations in a submerged biofilter (SBFR).

2. Materials and methods

2.1. Wastewater composition

The synthetic wastewater was used in the experimental study. The synthetic wastewater composition is as follows (in mg/L): NH_4Cl (650), Na_2EDTA (4.83), $CuSO_4$ (0.0046), $ZnSO_4{\cdot}7H_2O$ (0.023), $CoCl_2{\cdot}6H_2O$ (0.0119), $Na_2MoO_4{\cdot}2H_2O$ (0.066), $MgSO_4{\cdot}7H_2O$ (36.97), $NaHCO_3$ (226), $CaCl_2{\cdot}2H_2O$ (36.74), H_3BO_3 (1.0), $FeCl_3{\cdot}6H_2O$ (0.316), and KH_2PO_4 (1920) (Aslan and Gurbuz, 2011). After determining the optimal operating conditions in the SBFR, Ni^{2+} and Cu^{2+} were added to the synthetic wastewater.

2.2. Enrichment of microorganisms

The SBFR was used for about three years for partial nitrification experiments before it was used in this study. In order to reach a state condition, the reactor was fed with synthetic wastewater at an initial pH value of 7.5. Complete nitrification was achieved after operating the SBFR for about two months.

2.3. Reactor set-up and operation

The SBFR was filled with 20 mm-diameter plastic coil pieces that provided about 220 m^2 surface area/m^3 (specific weight of about 50.1 kg/m^3) for bacterial growth. Operational conditions and schematic diagram of the SBFR are summarized in Table 1 and Fig. 1, respectively.

The influent synthetic wastewater was pumped with a peristaltic pump (Watson Marlow, 520S) from the bottom of the SBFR and the effluent water was withdrawn from the top of reactor to an effluent tank. An air diffuser was installed directly in the center of the bottom of the SBFR in order to provide DO for the nitrification organisms. Air was provided continuously to the reactor by an aquarium aeration pump. The DO concentration was controlled at about 1.9 mg/L regularly with an oxygen probe and air flow rates were adjusted if required after changes of the operations. The DO concentration was periodically measured by using a DO meter (Hach Lange HQ40D) from the top of the SBFR. Throughout the experimental study, the reactor was kept at a temperature of 35° C with an electric heating element.

The ammonium-nitrogen loading rate (ALR) was elevated gradually by increasing the NH_4-N concentration of input wastewater. After determining the steady-state condition, experimental studies were carried out at the constant experimental condition by varying the concentrations of Cu^{2+} (in the range of 0.01–5.0 mg/L) and Ni^{2+} (in the range of 0.2–4.0 mg/L) in the influent tank. Combined effects were investigated at various initial concentration ratios of Ni^{2+}/Cu^{2+} (0.2/0.01, 1.0/1.0, 1.5/0.5, 2.0/2.0, and 3.0/3.0 mg/L).

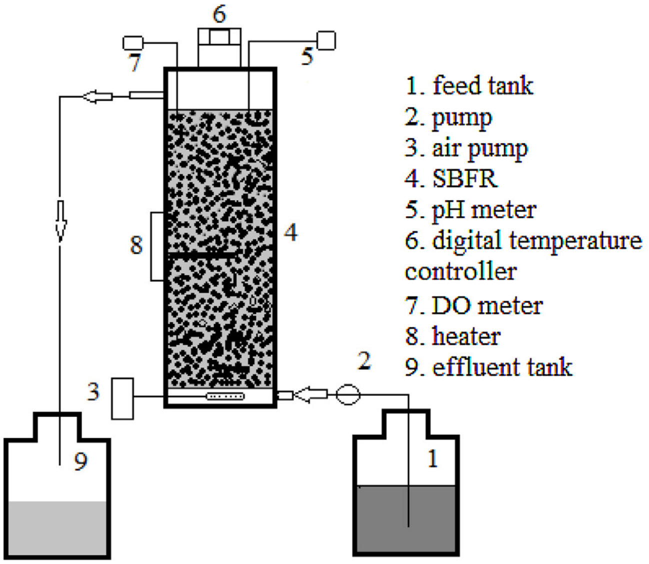

Fig. 1. Schematic diagram of the SBFR.

2.4. Analytical methods

Treated water samples were filtered to remove solids with 0.45 μm, filters with a 47 mm radius. Water samples were tested for NH_4-N, NO_3-N, NO_2-N, Ni^{2+} and Cu^{2+} concentrations using the following analytical kits: NH_4-N (14,752), NO_2-N (14,776), NO_3-N (14,773), Ni^{2+} (14,785), and Cu^{2+} (14,767) with a photometer. Nova 60 (Merck) spectrophotometer with a wavelength range of 340–820 ± 2 nm and spectral band with 10 nm was used to measure concentrations of parameters. Initial concentrations of heavy metals and nitrogen compounds in the feeding wastewater were determined daily. The analyses of samples were carried out at the temperature of 20 ± 2 °C.

3. Results and discussion

3.1. Continues operation of the SBFR

The SBFR was used for about 3 years at various ALR (and surface loading rates, SLR), hydraulic retention times (HRT) and DO concentrations to achieve the highest NH_4-N oxidations. Optimal operating conditions were determined at the initial concentrations for NH_4-N, feed wastewater flow rate, and ALR of 180 mg NH_4-N/L, 6.9 L/day, and 887 g NH_4-N/m^3/day (SLR of 3.73 g NH_4-N/m^2/day), respectively. Concentrations of NO_2-N and NH_4-N increased in the effluent waters when the ALR was higher than 887 g NH_4-N/m^3/day. During the study, about 99% of NH_4-N was converted to NO_3-N and the concentration of NO_2-N was lower than 2.0 mg/L in the effluent waters. The stable NO_3-N production rates of about 880 g/m^3 day (3.72 g NO_3-N/m^2/day) and ammonium removal rates (AARs) of 878 g/m^3/day were obtained under the operational conditions.

In order to evaluate the effects of heavy metal on the nitrifying bacteria, the experimental set-up was with an ALR of 887 g NH_4-N/m^3/day. Experiments were carried out at least two months for each concentration. The data used in the figures were determined