Desalination and Water Treatment
www.deswater.com

51 (2013) 2093–2101
February

doi: 10.1080/19443994.2013.734569

Removal of the metal ions Zn^{2+}, Ni^{2+}, and Cu^{2+} by biogenic sulfide in UASB reactor and speciation studies

Vinicius Farias de Albuquerque[a], Allen Lopes de Barros[b], Alexandre Colzi Lopes[a], André Bezerra dos Santos[a], Ronaldo Ferreira do Nascimento[a,b,]*

[a]*Department of Hydraulic and Environmental Engineering, Federal University of Ceará, Campus doPici—Rua do Contorno, S/N, Bl. 713, Fortaleza, Brazil*
[b]*Department of Analytical Chemistry and Physico-Chemistry, Federal University of Ceará, Campus do Pici—Rua do Contorno, S/N, Bl. 939, CEP 60451-970 Fortaleza, Brazil*
Tel. +55 85 33669958; Fax: +55 85 33669982; email: ronaldo@ufc.br

Received 11 August 2011; Accepted 11 April 2012

ABSTRACT

This work evaluated the effects of the chemical oxygen demand (COD)/SO_4^{2-} ratio on sulfate removal efficiency of an upflow anaerobic sludge blanket reactor inoculated with sludge from an industrial brewery. The removal of nickel (Ni^{2+}), zinc (Zn^{2+}), and copper (Cu^{2+}) by precipitation with the biogenic sulfide was also studied as well as the metals speciation through the reactor height with scanning electron microscopy/energy-dispersive X-ray spectroscopy/X-ray fluorescence (SEM/EDX/XRF) techniques. Ethanol was used as organic carbon source for electron donor, the hydraulic retention time was 18 h, and the organic loading rate was 1.5 kg COD m^{-3} d^{-1}. The experiment was divided in five periods, evaluating the system capacity in terms of sulfate reduction (COD/SO_4^{2-} of 2.26, 1.13, and 0.57) and metals (50 and 100 mg L^{-1}) removal by biogenic sulfide. High removal (higher than 98.5%) of the metals was achieved in the bioreactors, which were directly related to the solubilities solubility product of NiS, ZnS, and CuS. Metals dosage had a positive effect in COD removal by decreasing the toxicity caused by biogenic sulfide. SEM/EDX/XRF analyses indicated that Cu^{2+} was the most concentrated metal at the sludge samples collected at the reactor bottom, which also followed the lowest solubility properties of CuS, and was the least concentrated metal in both mid-height and upper sludge samples.

Keywords: Sulfate reduction; Metals; Speciation; SEM/EDX/XRF analyses

1. Introduction

Sulfate (Na_2SO_4) is the most stable sulfur compound in nature and occurs in municipal wastewater as well as in some industrial residuary waters under variable concentrations. Both direct and indirect disposal of sulfate-containing effluents produced by human activities may cause irreversible damages to water quality as well as to natural sulfur cycle.

Although Na_2SO_4 is present in municipal wastewaters in average concentrations of 50 mg L^{-1} [1], some of the industrial wastewaters may contain several thousands of milligrams of sulfur per liter [2]. Some examples of industrial activities whose effluents contain high amounts of both organic matter and Na_2SO_4 are: fermentation (citric acid and

---

*Corresponding author.

lysine), distillation (sugar cane spirits and wine), paper mill, vegetable oil refining, mining, and leather factories [3].

The anaerobic digestion of organic material in the presence of Na_2SO_4 is a complex process in which different physiological microbial groups mineralize organic matter to ultimately CH_4, CO_2, and H_2S [4]. In this process, sulfate-reducing bacteria (SRB) are able to couple the oxidation of organic compounds and hydrogen to Na_2SO_4 reduction producing H_2S [5,6].

Metabolic interactions between the different physiological microbial groups involved in the anaerobic process are an important research aspect. In anaerobic bioreactors, SRB can compete with methanogens and acetogens for common substrates. Hydrogen constitutes an important intermediate for which different microbes compete. Thermodynamically, the outcome of the competition between SRB and methanogens is clear; methanogens are outcompeted by SRB. This is clearly supported by several works [7,8]. However, despite that acetogenesis is less favorable than Na_2SO_4 reduction, the outcome of the competition between SRB and acetogens is not always clear. In fact, the coexistence of these two bacterial groups has been widely reported. In a bioreactor fed with H_2 and small quantities of CO_2, Weijma [9] observed that Na_2SO_4 reduction was the main process, but acetogenesis occurred as well. Stams [6] proposed that SRB were dependent for growth on acetogens.

Sulfide in the liquid can cause inhibition of the anaerobic organisms. The pH seems to be an important factor, since only the unionized form of sulfide is able to pass through the cell membrane [10]. Also, the different threshold of sulfide toxicity between the different bacterial groups [7,11] and the position of the micro-organisms in the process—e.g. in granular reactors—are important factors. Other problems associated to sulfide either in the effluent or in the biogas are unpleasant odors, corrosiveness, and toxicity.

Driven by this prerequisite, chemical and physical processes used in Na_2SO_4 removal include low-cost approaches, like the precipitation with calcium salts as well as more expensive alternatives, such as reverse osmosis, electrodialysis, and nanofiltration. Due to the amount of reagents employed, chemical precipitation may result in large quantities of residues, which demand proper classification, handling, and disposal, while in membrane techniques special attention on membrane clogging is required [12].

Over the last decade, many biotechnological strategies have been developed aiming the removal of anions containing oxidized sulfur (SO_3^{2-}, HSO_3^-, $S_2O_3^{2-}$, and SO_4^{2-}) from industrial wastewaters. Firstly, sulfur oxides are converted into sulfides ($H_2S_{(g)}$ + $H_2S_{(aq)}$ + $HS^-_{(aq)}$) under anaerobic conditions. The introduction of limited quantities of oxygen/air to anaerobic bioreactors has been considered to lower the levels of sulfide. This technique involves the oxidation of sulfide by sulfur-oxidizing bacteria (SOB) under limiting oxygen conditions. Various studies have demonstrated that, under oxygen limitation, sulfide is mainly converted to elemental sulfur (S^0) and thiosulfate by SOB [13–16].

The presence of heavy metals like nickel (Ni^{2+}), zinc (Zn^{2+}), and copper (Cu^{2+}) is another concern for some industrial wastewaters, such as those from mining, petrochemical, and metallurgical industries, because these elements can contaminate water bodies, damaging both animal and human health [17]. In the sulfate-reducing bioreactors, the sulfide anions can form sparingly soluble precipitates of divalent metals as metal sulfides [18]. The general reactions used to describe the Na_2SO_4 reduction and precipitation metal process are shown in Eqs. (1) and (2):

$$\text{Organic matter (C, H, O)} + SO_4^{2-} \rightarrow HS^- + HCO_3^- \quad (1)$$

$$M^{2+} + HS^- \rightarrow MeS(s) + H^+(M^{2+}) \quad (2)$$

The main advantages of metal precipitation by hydrogen sulfide produced from the biological Na_2SO_4 reduction include: (i) instantaneous metal–sulfide complex formation; (ii) low solubility of the metallic sulfides compared to other metal complex compounds (hydroxides, carbonates, chlorides, etc.); and (iii) low-sludge production [19].

Different configurations of reactors have been designed to maximize the production of biogenic sulfide and the subsequent precipitation of heavy metals [20]. Additionally, there are some investigations regarding Zn^{2+}, Cd^{2+}, Ni^{2+}, and Cu^{2+} removal or formation from aqueous solutions, for instance, by testing different sulfide sources.

However, in most of the studies, the sulfide precipitation was applied as an effluent treatment method, and it was used for heavy metals removal from mine waters and industrial effluents [21].

Sulfide gives the possibility of selective precipitation, due to the different solubility products of the different metal sulfides (Table 1). Having the solubility product defined as $KSP = (Me^{2+})\ (S^{2-})$, it means that different sulfide concentrations (S^{2-} potentials) are required to precipitate different metals.

Table 1
Experimental conditions to evaluate the system capacity in terms of sulfate reduction (chemical oxygen demand [COD]/SO_4^{2-} of 2.26, 1.13, and 0.57) and metals (50 and 100 mg L^{-1}) removal by biogenic sulfide

Period	1		2		3		4		5	
	Influent	Effluent	Influent	Effluent	Influent	Effluent	Influent	Effluent	Influent	Effluent
End of the period (days)	33		62		118		150		170	
Co-substrate (g COD L^{-1})	1.13	–	1.13	–	1.13	–	1.13	–	1.13	–
SO_4^{2-} (mg L^{-1})	500	–	1,000	–	2,000	–	2,000	–	2,000	–
COD/SO_4^{2-}	2.26	–	1.13	–	0.57	–	0.57	–	0.57	–
OLR (kg COD m^{-3} d^{-1})	1.5	–	1.5	–	1.5	–	1.5	–	1.5	–
Ni^{2+}, Zn^{2+}, and Cu^{2+} ions concentrations (mg L^{-1})	–	–	–	–	–	–	50.0	–	150.0	–
pH	7.84–8.28	7.33–7.82	7.40–8.23	7.10–7.78	7.11–8.20	7.03–7.84	7.11–8.12	7.03–7.70	7.11–8.12	7.03–7.70
TA (mg L^{-1} of $CaCO_3$)	–	773 ± 55.4	–	997 ± 104.2	–	995 ± 57.0	–	957 ± 44.5	–	957 ± 50.0
VFA (mg L^{-1} of HAc)	–	125 ± 55.9	–	361 ± 205.8	–	177 ± 99.7	–	156 ± 30.0	–	156 ± 39.5
VFA/TA	–	0.17 ± 0.07	–	0.39 ± 0.12	–	0.13 ± 0.03	–	0.16 ± 0.06	–	0.25 ± 0.01
S^{2-} production (mg L^{-1})	–	49.7 ± 10.5	–	152.3 ± 32.5	–	203.5 ± 61.6	–	80.5 ± 10.9	–	25.8 ± 9.3
COD removal (%)	83 ± 6.9		76 ± 14.3		62 ± 17.2		71 ± 3.5		76 ± 6.7	
SO_4^{2-} removal (%)	67 ± 14.0		69 ± 10.3		43 ± 6.9		39 ± 5.5		65 ± 6.7	

OLR: organic loading rate; TA: total alkalinity; and VFA: volatile fats acids.

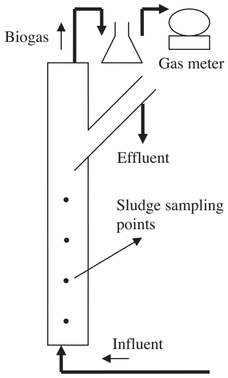

Fig. 1. Schematic of the Y-UASB reactor used in the study.

Nonetheless, Na_2SO_4 reduction is extremely depending upon the sludge source, electron donor used, COD/SO_4^{2-} ratio, reactor design as well as the chemical species to be precipitated.

This work aims to evaluate the effects of the COD/SO_4^{2-} ratio on Na_2SO_4 removal efficiency of an upflow anaerobic sludge blanket (UASB) reactor inoculated with sludge from an industrial brewery. The removal of Ni^{2+}, Zn^{2+}, and Cu^{2+} by precipitation with the biogenic sulfide was also studied as well as the metals speciation through the reactor height was conducted by scanning electron microscopy/energy-dispersive X-ray spectroscopy/X-ray fluorescence (SEM/EDX/XRF) techniques.

2. Materials and methods

2.1. Reactor characteristics

The continuous flow experiments consisted of an UASB reactor to treat a sulfate-containing synthetic effluent in different COD/SO_4^{2-} ratios, to which metals were added in the basal medium during some periods. The experiments were carried out from 5 August 2008 to 12 January 2009.

The reactor (Fig. 1) was made of polyvinyl chloride in Y-form [22], with working volume of 11.2 L. There were four sludge sampling points along the reactor height (Fig. 1).

The UASB reactor was operated at a hydraulic retention time of 18 h and a flow rate of 14.9 L d^{-1} was applied by using diaphragm pumps (Prominent, Brazil). Ethanol was selected as the electron donor (99.8% purity, Dinâmica, Brazil), and the organic loading rate applied was 1.5 kg COD m^{-3} d^{-1}. The influent was kept at 4℃ and the reactors were operated at room temperature of approximately 27℃. The biogas produced was collected and washed in a NaOH solution (0.5 N), after which methane was measured in a gas meter (Model TICO 731, Ritter).

2.2. Inoculation and start-up

The reactor was installed at the Sanitation Laboratory (Labosan) of the Department of Hydraulic and Environmental Engineering at the Federal University of Ceará. After checking for leak, the flow rate was adjusted prior to sludge inoculation. The anaerobic sludge was collected from an UASB reactor located in a brewery (Fortaleza, Brazil). The sludge volume added in the reactor provided a sludge concentration of approximately 30 g VSS/L.

The basal medium consisted of (mg L^{-1}): NH_4Cl (280), K_2HPO_4 (250), $MgSO_4 \cdot 7H_2O$ (100) and $CaCl_2 \cdot 2H_2O$ (10), and 1 mL L^{-1} of trace elements containing (mg L^{-1}): H_3BO_3 (50), $FeCl_2 \cdot 4H_2O$ (2,000), $ZnCl_2$ (50), $MnCl_2 \cdot 4H_2O$ (500), $CuCl_2 \cdot 2H_2O$ (38), $(NH_4)_6Mo_7O_{24} \cdot 4H_2O$ (50), $AlCl_3 \cdot 6H_2O$ (90), $CoCl_2 \cdot 6H_2O$ (2,000), $NiCl_2 \cdot 6H_2O$ (92), $Na_2SeO_3 \cdot 5H_2O$ (162), and EDTA (1,000) and HCl 36% (1). To keep the pH around 7.0, the wastewater was buffered with sodium bicarbonate ($NaHCO_3$) in the proportion of 1 g $NaHCO_3$ to each 1 g COD ethanol.

All chemicals used in the basal medium preparation were from analytical grade weighed in a precision balance and then dissolved in distilled water. The influent was prepared in 50 L container every three days approximately, according to its flow rate.

2.3. Reactor operation

The experiment was divided in five periods (Table 1), evaluating the system capacity in terms of Na_2SO_4 reduction (COD/SO_4^{2-} of 2.26, 1.13, and 0.57) and metals (50 and 100 mg L^{-1}) removal by biogenic sulfide. The influent composition was essentially the same along the experiments: basal medium, ethanol, and $NaHCO_3$, so that the changes consisted of varying the concentrations of Na_2SO_4 and metal ions.

Na_2SO_4 was added in different concentrations (500, 1,000, and 2,000 mg L^{-1}), in order to reach COD/SO_4^{2-} ratios of 2.26, 1.13, and 0.57, for the Periods 1, 2, and 3, respectively. In Period 4, both metal ions Zn^{2+} and Ni^{2+} were added, initially in the concentration of 50 mg L^{-1} each. The same concentration of Cu^{2+} ion was just added from day 16th onwards, because there was a delay in the chemical delivery. In Period 5, the concentration of each metal ion was increased to 150 mg L^{-1} (Table 1).

2.4. Metals speciation studies

Three sludge samples were collected at the end of the experiment in order to verify the metals speciation through the reactor height. Sample A1 was taken from

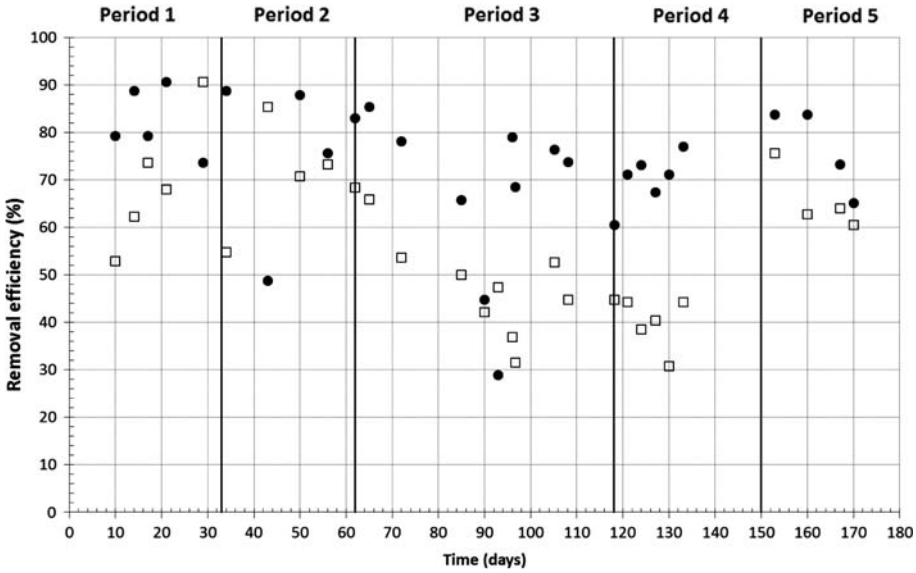

Fig. 2. COD (●) and SO_4^{2-} (□) removal efficiencies during the periods of experiment.

the lowest sampling point, sample A2 was from an intermediate point, and A3 was taken from the highest sampling point. The sludge was dried by using an oven and macerated afterwards in order to be converted into a finely divided powder which was suitable for analysis.

2.5. Analyses

Alkalinity, pH, volatile fat acids (VFA), COD, Na_2SO_4, and sulfide levels were analyzed twice a week in the influent and effluent, according to APHA [23]. Residual concentrations of metal ions Ni^{2+}, Zn^{2+}, and Cu^{2+} were determined by flame atomic absorption spectrometry (model GBC 933 plus).

For COD, pH, and alkalinity analyses, dilution was the only sample treatment. For Na_2SO_4 analyses, vacuum filtration was used (model Q 355 B2, Quimis). Besides vacuum filtration, metal ion analyses required acidification with nitric acid to prevent the formation of suspended particles.

Metals speciation through the reactor height was conducted by SEM, EDX, and XRF techniques (model ZSX Mini II, Rigaku).

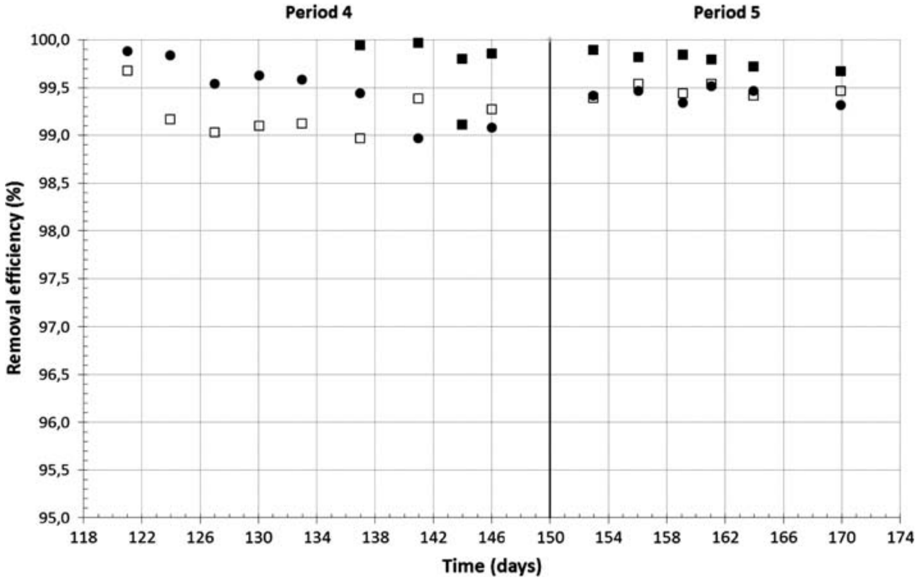

Fig. 3. Removal efficiencies of Zn^{2+} (□), Ni^{2+} (●) and Cu^{2+} (■) during Periods 4 and 5.

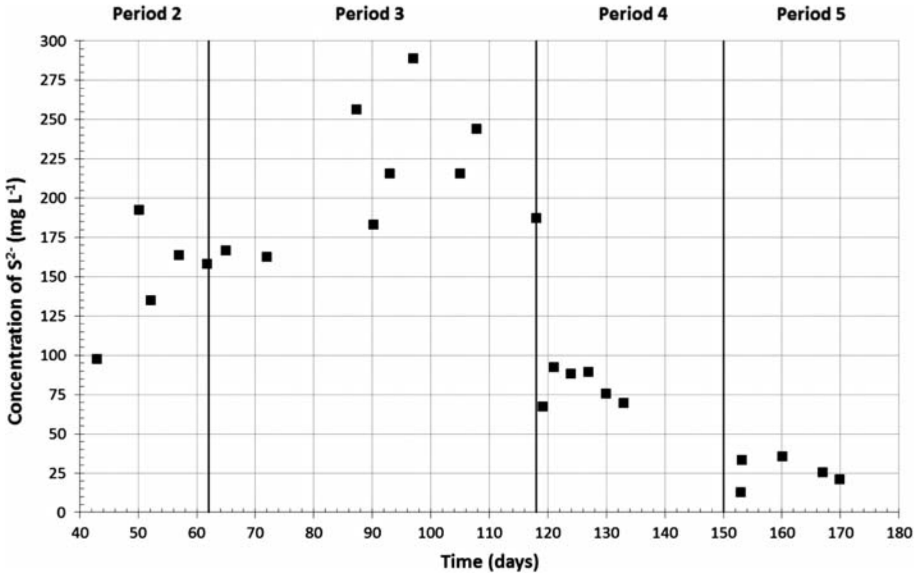

Fig. 4. Sulfide effluent concentration (■) during Periods 2, 3, 4, and 5.

3. Results and discussion

3.1. Effects of the COD/SO_4^{2-} ratio on Na_2SO_4 removal efficiency

In Period 1, Na_2SO_4 concentration in the influent was 500 mg L^{-1}, which resulted in a COD/SO_4^{2-} ratio of 2.26 (Table 1). The average COD removal was 83.3 ± 6.9%, which indicated the consortium was adapted to ethanol as the electron donor. A moderate average Na_2SO_4 reduction capacity of 67.0 ± 14.0% was already found in the system, showing that indeed part of ethanol oxidation verified in terms of COD removal was coupled to Na_2SO_4 reduction.

The effluent pH was in the range of 7.33–7.82, and averages of Total alkalinity (TA) and VFA were 773.1 ± 55.4 and 125.4 ± 55.9 mg L^{-1}, respectively. The VFA/TA ratio was 0.17 ± 0.07. According to Ripley [24], VFA/TA ratios higher than 0.30 indicate disturbances in the anaerobic digestion process. Therefore, the operational and efficiency results of Period 1 show that reactor was stable.

In Period 2, Na_2SO_4 concentration in the influent was increased to 1,000 mg L^{-1} which resulted in a COD/SO_4^{2-} ratio of 1.13. The average COD removal was 76.4 ± 14.3% and Na_2SO_4 removal was 69.5 ± 10.3%. Therefore, comparing to Period 1, no remarkable difference was found in terms of both COD and Na_2SO_4 removal efficiencies. The operational parameters such VFA and VFA/TA ratio show some disturbances in the system, probably due to the higher concentration of sulfide (Table 1).

In Period 3, a COD/SO_4^{2-} ratio of 0.57 was applied, which resulted in COD and Na_2SO_4 removal efficiencies of 62.2 ± 17.2 and 43.8 ± 6.9%, respectively (Table 1). The effluent pH was in the range of 7.03–7.84, and averages of TA and VFA were 995.8 ± 57.0 and 177.3 ± 99.7 mg L^{-1}, respectively. The VFA/TA ratio was 0.13 ± 0.03.

COD and SO_4^{2-} removal efficiencies during the periods of experiment are shown in Fig. 2. It was observed that, lower the COD/SO_4^{2-} ratio, lower the Na_2SO_4 removal efficiency. Cao [19] evaluated COD/SO_4^{2-} ratios of 0.5, 1–3, and 9, and found that the highest Na_2SO_4 removal was obtained during COD/SO_4^{2-} of 3, while the lowest efficiency was observed for COD/SO_4^{2-} ratio of 0.5. According to several authors [8,9,25], when the COD/SO_4^{2-} ratio is higher than 0.6, the organic matter available is expected to be sufficient to complete reduction of Na_2SO_4 anions present.

3.2. Removal of Ni^{2+}, Zn^{2+}, and Cu^{2+} by precipitation with the biogenic sulfide

In Periods 4 and 5, the metal ions were individually added in the concentration of 50 mg L^{-1} (Period 4) and 150 mg L^{-1} (Period 5) (Table 1), in the form of salts $NiCl_2.6H_2O$, $ZnCl_2$, and $CuCl_2.2H_2O$.

In Period 4, initially both Ni^{2+} and Zn^{2+} were dosed being cupper added from day 16th onwards. The efficiencies on both COD and Na_2SO_4 removals were around 71.9 ± 3.5 and 39.6 ± 5.5, respectively, (Table 1, Fig. 2). Comparing to Period 3, an increase

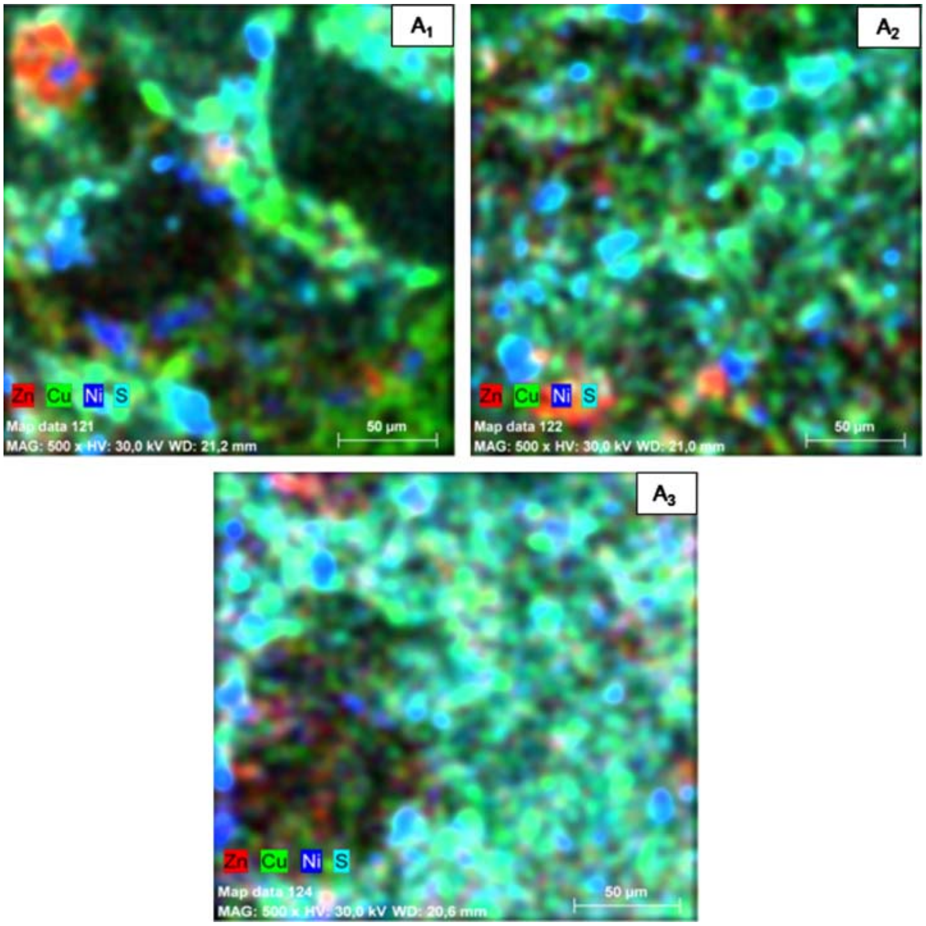

Fig. 5. SEM analyses performed for the presence of metal ions in sludge granules samples (A1, A2, and A3).

in COD removal efficiency was observed, probably due to the partial precipitation of sulfide anions which decreases the sulfide concentration in the reactor. The operational parameters indicated that the reactor remained stable even after the addition of metal ions.

Fig. 3 shows the removal efficiencies of Ni^{2+}, Zn^{2+}, and Cu^{2+}. Metals removal efficiencies were 99.5, 99.2, and 99.9%, respectively, for Ni^{2+}, Zn^{2+} and Cu^{2+}, and the standard deviation was lower than 0.3% in all cases.

In Period 5, the efficiencies on both COD and Na_2SO_4 removals were around 71.9 ± 3.5 and $39.6 \pm 5.5\%$, respectively, (Table 1, Fig. 2). Effluent pH ranged from 7.03 to 7.70, and VFA/TA ratio was 0.25 ± 0.01, which indicated that the reactor stability was not affected by the metal ions addition.

The average removal efficiencies were 99.4, 99.5, and 99.8% for Ni^{2+}, Zn^{2+}, and Cu^{2+}, respectively, (Table 1, Fig. 3). The standard deviation was lower than 0.3% in all cases. These results are similar to those obtained by Jong and Parry [18] by using a combination of SRB in an anaerobic reactor with a sand bed.

Sulfide effluent concentration ($mg\,L^{-1}$) is shown in Fig. 4. Comparing the periods of depletion of metals (Period 3) and presence of metals (Periods 4 and 5) for the same COD/SO_4^{2-} ratio, it can be observed that a decrease in effluent sulfide concentration was attributed to the metals precipitation by biogenic sulfide [7,10,11]. The lowest sulfide concentration was found in Period 5, when metals were present in the highest concentration, probably as a consequence of a higher metal complex formation.

The metals efficiency values presented in Fig. 3 are directly related to the solubilities of NiS (pKs = 27.98), ZnS (pKs = 28.39), and CuS (pKs = 40.94) [26], and are in agreement with the results obtained by Cao [19], Machemer and Wildeman [27] with a fixed bed anaerobic reactor.

3.3. Metals speciation studies (SEM/EDX/XRF analyses)

Both SEM and EDX analyses were performed in order to assess metals speciation through the reactor height. Fig. 5 displays the images obtained in SEM analyses of samples A1, A2, and A3, showing the presence of Cu^{2+}, Ni^{2+}, Zn^{2+} as well as sulfur in the samples. SEM results revealed that in sample A1 (bottom), Cu^{2+} was the most abundant metal, followed by Zn^{2+} and Ni^{2+}. In sample A2 (intermediate), Ni^{2+} was the most concentrated metal, followed by Zn^{2+} and Cu^{2+}. Zn^{2+} concentration was the highest in sample A3 (top), followed by Ni^{2+} and Cu^{2+}.

XRF technique was also used in order to build a both qualitative and quantitative samples profile. The percentages of Cu^{2+}, Ni^{2+}, and Zn^{2+} were, respectively, 28.8, 14.1, and 16.1% in sample A1; 8.6, 21.6, and 18.7% in sample A2; and 11.5, 16.8, and 27.2% in sample A3.

These results are in accordance with those of SEM/EDX analyses, and indicate that in the bottom of the reactor Cu^{2+} was the most concentrated metal, as expected when considering the lower solubility of CuS, which precipitates more easily than the other metals present in this system, thus accumulating near the reactor inlet. Moreover, Cu^{2+} was the least concentrated metal in both mid-height and upper zones of the reactor.

4. Conclusions

High removals (higher than 98.5%) of the metals Ni^{2+}, Zn^{2+}, and Cu^{2+} were achieved in the bioreactors, which were directly related to the solubilities of NiS, ZnS, and CuS.

Metals dosage had a positive effect in COD removal by decreasing the toxicity caused by biogenic sulfide.

SEM/EDX/XRF analyses indicated that Cu^{2+} was the most concentrated metal at the sludge samples collected at the reactor bottom, which also followed the lowest solubility properties of CuS, and was the least concentrated metal in both mid-height and upper sludge samples.

References

[1] G. Tchobanoglous, Wastewater Engineering: Treatment, Disposal and Reuse, Metcalf & Eddy, New York, NY, 1991.

[2] A.J. Silva, M.B. Varesche, E. Foresti, M. Zaiat, Sulphate removal from industrial wastewater using a packed-bed anaerobic reactor, Process Biochem. 37 (2002) 927–935.

[3] D.G. Cirne, F.P. Zee, M. Fernandez-Polanco, F. Fernandez-Polanco, Control of sulphide during anaerobic treatment of S-containing wastewaters by adding limited amounts of oxygen or nitrate, Rev. Environ. Sci. Bio/Technol. 7 (2008) 93–105.

[4] B. Schink, A. Stams, Syntrophism among prokaryotes, in: M. Dworkin, K.-H. Schleifer, E. Stackebrandt (Eds.), The Prokaryotes, Springer Verlag, New york, NY, 2006, pp. 309–335.

[5] H.F. Castro, N.H. Williams, A. Ogram, Phylogeny of sulfate-reducing bacteria(1), FEMS Microbiol. Ecol. 31 (2000) 1–9.

[6] A.J.M. Stams, C.M. Plugge, F.A.M. de Bok, B. van Houten, P. Lens, H. Dijkman, J. Weijma, Metabolic interactions in methanogenic and sulfate-reducing bioreactors, Water Sci. Technol. 52 (2005) 13–20.

[7] V. O'Flaherty, P. Lens, B. Leahy, E. Colleran, Long-term competition between sulphate-reducing and methane-producing bacteria during full-scale anaerobic treatment of citric acid production wastewater, Water Res. 32 (1998) 815–825.

[8] D.Z. Sousa, J.I. Alves, M.M. Alves, H. Smidt, A.J. Stams, Effect of sulfate on methanogenic communities that degrade unsaturated and saturated long-chain fatty acids (LCFA), Environ. Microbiol. 11 (2009) 68–80.

[9] J. Weijma, F. Gubbels, L.W.H. Pol, L.W.H Stams, P. Lens, G. Lettinga, Competition for H_2 between sulfate reducers, methanogens and homoacetogens in a gas-lift reactor
 Anaerobic Digestion IX, Selected Proceedings of the Nineth World Congress on Anaerobic Digestion, Antwerp, Belgium, September 2–6, 2001, Water Sci. Technol. 45 (2002) 75–80.

[10] R.E. Speece, Anaerobic Biotechnology and Odor/Corrosion Control, Archae Press, Nashville, TN, 2008.

[11] H.B. McDonald, G.F. Parkin, Effect of sulfide inhibition and organic shock loading on anaerobic biofilm reactors treating a low-temperature, high-sulfate wastewater, Water Environ. Res. 81 (2009) 265–288.

[12] A. Korchef, I. Kerkeni, M. Ben Amor, S. Galland, F. Persin, Iron removal from aqueous solution by oxidation, precipitation and ultrafiltration, Desalin. Water Treat. 9 (2009) 1–8.

[13] F.P. van der Zee, S. Villaverde, P.A. Garcia, F. Fdz-Polanco, Sulfide removal by moderate oxygenation of anaerobic sludge environments, Bioresour. Technol. 98 (2007) 518–524.

[14] A.J.H. Janssen, G. Lettinga, A. de Keizer, Removal of hydrogen sulphide from wastewater and waste gases by biological conversion to elemental sulphur: Colloidal and interfacial aspects of biologically produced sulphur particles, Colloid Surf. A 151 (1999) 389–397.

[15] A. Sarti, M. Zaiat, Anaerobic treatment of sulfate-rich wastewater in an anaerobic sequential batch reactor (AnSBR) using butanol as the carbon source, J. Environ. Manage. 92 (2011) 1537–1541.

[16] F.P. Ende, J. Meier, H. Gemerden, Syntrophic growth of sulfate-reducing bacteria and colorless sulfur bacteria during oxygen limitation, FEMS Microbiol. Ecol. 23 (2006) 65–80.

[17] B. Johnson, Biological removal of sulfurous compounds from inorganic wastewaters, in: P. Lens, L.H. Pol (Eds.), Environmental Technologies to Treat Sulfur Pollution, IWA, London, 2000, pp. 175–206.

[18] T. Jong, D.L. Parry, Removal of sulfate and heavy metals by sulfate reducing bacteria in short-term bench scale upflow anaerobic packed bed reactor runs, Water Res. 37 (2003) 3379–3389.

[19] A. Velasco, M. Ramirez, T. Volke-Sepulveda, A. Gonzalez-Sanchez, S. Revah, Evaluation of feed COD/sulfate ratio as a control criterion for the biological hydrogen sulfide production and lead precipitation, J. Hazard. Mater. 151 (2008) 407–413.

[20] J. Cao, G. Zhang, Z. Mao, Z. Fang, C. Yang, Precipitation of valuable metals from bioleaching solution by biogenic sulfides, Miner. Eng. 22 (2009) 289–295.

[21] M. Gharabaghi, M. Irannajad, A.R. Azadmehr, Selective sulphide precipitation of heavy metals from acidic polymetallic aqueous solution by thioacetamide, Indus. Eng. Chem. Res. 51 (2012) 954–963.

[22] P.F.F. Cavalcanti, Intregrated application of the UASB reactor and ponds for domestic sewage treatment in tropical regions, Sub-Department of Environmental Technology, Wageningen University, Wageningen, 2003, p. 139.

[23] APHA, Standard Methods for the Examination of Water and Wastewater, 21, American Public Health Association, Washington, DC, 2005.

[24] L.E. Ripley, W.C. Boyle, J.C. Converse, Improved alkalimetric monitoring for anaerobic digestion of high-strength wastes, Journal (Water Pollut. Control Feder.) 58 (1986) 406–411.

[25] A. Rinzema, G. Lettinga, The effect of sulfide on the anaerobic degradation of propionate, Environ. Technol. Lett. 9 (1988) 83–88.

[26] D.M. Ditoro, J.D. Mahony, D.J. Hansen, K.J. Scott, M.B. Hicks, S.M. Mayr, M.S. Redmond, Toxicity of cadmium in sediments: The role of acid volatile sulfide, Environ. Toxicol. Chem. 9 (1990) 1487–1502.

[27] S.D. Machemer, T.R. Wildeman, Adsorption compared with sulfide precipitation as metal removal processes from acid mine drainage in a constructed wetland, J. Contam. Hydrol. 9 (1992) 115–131.