Effect of hydraulic retention time on continuous biocatalytic calcification reactor

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ABSTRACT

High calcium concentrations in the wastewaters are problematic, because they lead to clogging of pipelines, boilers and heat exchangers through scaling (as carbonate, sulfate or phosphate precipitates), or malfunctioning of aerobic and anaerobic reactors. As a remedy to this problem, the industry typically uses chemical crystallization reactors which are efficient but often require complex monitoring and control and, as a drawback, can give rise to highly alkaline effluents. Biomineralization are emerging as alternative mechanisms for the removal of calcium from aqueous environments. Biocatalytic calcification reactors (BCR) utilize microbial urea hydrolysis by bacteria for the removal of calcium, as calcite, from industrial wastewater. Hydraulic retention time (HRT) effect on calcium removal was studied with a continuous feed BCR reactor treating a simulated pulp paper wastewater. Study showed that HRT is important parameter and HRT of 5–6 h is optimum for calcium removal from calcium-rich wastewaters.

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1. Introduction

Conservation and management of clean water resources have become more significant in the recent years because of the increasing water pollution problems that are caused by the rapid population growth and industrialization. Moreover, the recycling of water produced by the industrial processes has become necessary because of the increase in cost of clean water obtainment. Therefore, the processed water in many different industrial sectors is recycled. The water resources with higher calcium (Ca) content are generally not preferred as potable water. Ca limits the cross section of the pipes by precipitating as $CaCO_3$ Ca in certain conditions (pH, temperature, alkalinity, etc.), and reduces the efficiency the reactor by creating dead spaces, especially when contained in wastewater treated with anaerobic methods. Ca is generally removed from the water by chemical methods. On the other hand, new technologies for Ca removal is expedited since the operation of chemical processes is complex, the chemicals are costly, their effluents is alkali [1], and production of excess sludge in these classical methods. The process of biological mineral precipitation is a good alternative for the removal of water pollutants such as heavy metals, radioactive elements, phosphate and salts from water and wastewater [2]. Calcium removal by urea hydrolysis, which is a natural biological mineral precipitation process, is first used by Hammes et al. [1,3,4] using the name biocatalytic calcification reactor (BCR). The working

principle of this reactor is to precipitate Ca as by $CaCO_3$ by creating high pH and alkalinity with the addition of urea.

Microorganisms have long been known to catalyse the precipitation of $CaCO_3$ in natural environments such as oceans, soils and saline lakes, in a process referred to as microbial carbonate precipitation (MCP) [5,6]. In nature carbonate precipitation may theoretically occur following several known processes: (i) abiotic chemical precipitation from saturated solutions by evaporation, temperature increase and/or pressure decrease; (ii) external or internal skeleton production by eukaryotes; (iii) lowering of CO_2 pressure under effect of autotrophic processes (photosynthesis, methanogenesis); (iv) fungal mediation; (v) heterotrophic bacterial mediation. There are different ways in the $CaCO_3$ production particles through heterotrophic bacterial mediation. (i) The amonification of aminoacids in aerobiosis (i.e. in the presence of gaseous or dissolved oxygen, in the presence of organic matter and calcium); (ii) the dissimilatory reduction of nitrate in anaerobiosis (i.e. in the absence of oxygen) or microaerophily (i.e. in the presence of very low amounts of oxygen, in the presence of organic matter, calcium and nitrate), and (iii) the degradation of urea or uric acid (anaerobiosis, in the presence of organic matter, calcium, and urea or uric acid) [5]. Although there are several MCP pathways and precipitation mechanisms have been recognized in nature, one of the most straightforward examples is microbial urea degradation. In this degradation, urea is hydrolyzed via the urease enzyme which is common in a wide variety of plants, fungi and bacteria. Urea hydrolysis and the mineralizing of organic carbon induce production of carbonate and bicarbonate ions from CO_2, and ammonia, which induces pH increase. These products can

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subsequently lead to precipitate as $CaCO_3$ in the presence of soluble calcium ions [3].

Hydraulic retention time (HRT) is a significant design and operation parameter for biological reactors. Although, in theory, the efficiency of the reactor is determined by SRT, HRT is a necessary parameter for consideration. Suitable HRT durations should be ascertained for biological systems. Since design and operation parameters of BCR are not definitely ascertained, the effect of HRT is researched in the performance of BCR that is operated in fixed SRT and fed with simulated synthetic wastewater simulating domestic and paper factory wastewater in this study.

2. Materials and method

2.1. Wastewater characteristics and BCR for continuous feeding studies

The content of the intended synthetic wastewater that was used for continuous feeding studies was prepared by composing domestic wastewater with average degree of pollutants, as advised by Holakoo et al. [7], and wastewater of a paper factory, which the contents of the water is advised by Kim et al. [8]. The content of the prepared synthetic composition that was used in the studies is given in Table 1. The aim of mixing wastewater composition with the domestic wastewater composition was the combined removal of domestic wastewater caused by the personnel in the paper factories and therefore to prevent the absence of major or minor feeding element in paper factory wastewater. The COD/N/P ratio of wastewater was elicited as 100/28/1. As the first step, wastewater simulation with no containment of Ca, as seen in Table 1, was used for the production of sufficient biomass of urea hydrolysis. BCR had dimensions of 7 cm × 18 cm × 30 cm; and consisted of aeration basin of 1.5 L and sedimentation basin of 0.65 L. Aeration basin and sedimentation unit was separated by a perforated plate that allowed water transition (Fig. 1). The transition of precipitated sludge to aeration unit provided the sludge recycling ratio (r) to be fixed to 1 by the help of a 2–3 cm opening in the bottom of the plate. BCR, with the assumption that there was no biomass in the processed water, was operated by using sludge retention time (SRT) of 5 days. The composition in the reactor was created by using diffused ventilators that could provide sufficient oxygen (>2 mg L^{-1}) in the aeration basin. Simulated wastewater was given to the reactors, from the condition of 4 °C, by peristaltic pumps. Reactor and removal studies were implemented in a fixed temperature of 25 °C. Reactor was operated for 98 days, after it reached stable conditions, by changing the HRT in every 15 days.
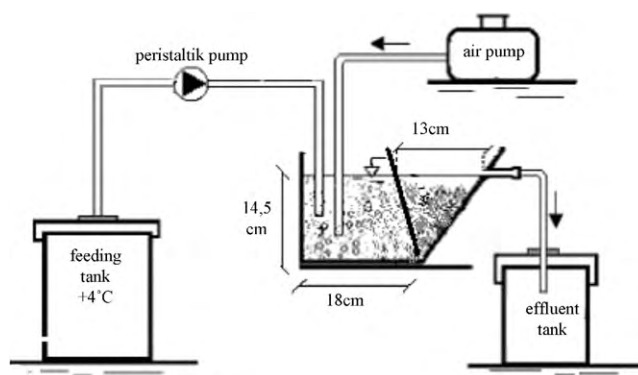

Fig. 1. The schematic configuration of the continuous BCR.

2.2. Analytical methods

All the analyses were implemented by using standard methods [9]. Ammonium was measured by Merc-Spectroquant kits and total nitrogen (TN) measurements were realized by TOC-TN (Shimadzu TOC-VCPN) analysis system.

3. Results and discussion

Although, the only parameter for determining the concentration of the substrate in reactor effluent is theoretically SRT, which is the duration of the containment of the microorganisms in the system, HRT should also be suitable in biological reactors as well. Not all of the organic matters in the wastewaters are in a form that is used by microorganisms. Therefore, the determination of suitable HRT values for biological reactors is significant. For the removal of COD in the treatment process of wastewater containing organic matters with bigger molecules such as cellulose, starch and fat; first of all, these matters should be soluble and transferred to a form that can easily be used by microorganisms [10]. It is a generally advised rule that HRT used in bioreactors should not be less than SRT [11]. Likewise, the organic loading changes with the change in HRT. Therefore, when the SRT, which influences the substrate concentration in the process, is not changed, the biomass concentration should increase or decrease, theoretically, for sustaining the same value in the effluent. When the HRT are reduced, the performance of the system would decrease because the problems of bulking of sludge and insufficient aeration would appear.

COD, TN and Ca^{2+} removal efficiencies in BCR that were operated in fixed SRT and differing HRTs could be seen in Fig. 2. All parameters have significantly low percentages of removal with HRT of 1.58 h whereas all values approaches to optimum with HRT of 5.31 h. 63% of COD, 14% of nitrogen (N) and 72% of Ca removal was

Table 1
Characteristics of simulated wastewater.

| The functions of material used	Material used	Concentration, mg L^{-1}	Target conc., mg L^{-1}
Paper and domestic wastewater	$C_6H_{12}O_6 \cdot H_2O$	1548	1500 mg glucose-COD L^{-1}
Paper and domestic wastewater	$CaCl_2$	1665	601.2 mg Ca L^{-1}
Paper and domestic wastewater and BCR process	$CO(NH_2)_2$(urea)	901	420 mg N L^{-1}
Paper and domestic wastewater	$MgSO_4 \cdot 7H_2O$	1540	600 mg SO$_4$ L^{-1}
Paper and domestic wastewater	Na_2CO_3	477	270 mg CaCO$_3$ L^{-1}
Paper and domestic wastewater	$NaHCO_3$	378	270 mg CaCO$_3$ L^{-1}
Domestic wastewater and trace element	KH_2PO_4	66	15 mg P L^{-1}
Domestic wastewater	$FeCl_2 \cdot 4H_2O$	19	4 mg Fe L^{-1}
Domestic wastewater	$CoCl_2 \cdot 6H_2O$	0.404	0.10 mg Co L^{-1}
Domestic wastewater	$ZnCl_2$	0.229	0.11 mg Zn L^{-1}
Domestic wastewater	$NaMoO_4 \cdot 2H_2O$	0.208	0.08 mg Mo L^{-1}
Domestic wastewater	$CuSO_4 \cdot 5H_2O$	0.118	0.03 mg Cu L^{-1}
Domestic wastewater	$MnSO_4 \cdot H_2O$	0.123	0.04 mg Mn L^{-1}
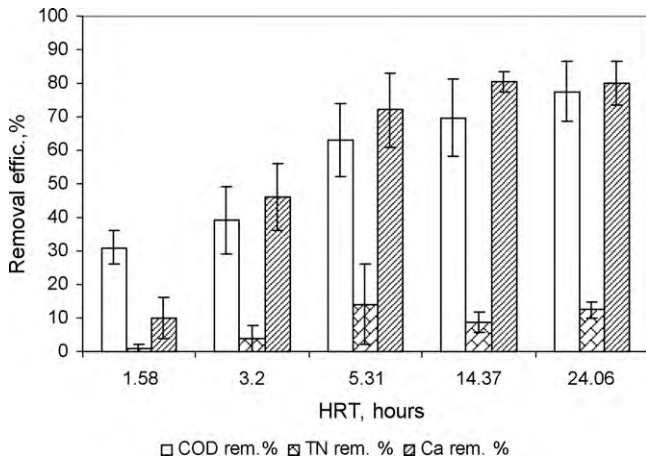
Fig. 2. COD, TN and Ca removal efficiencies of BCR reactor at different HRT.

obtained in this HRT. In the Fig. 2, the decreasing of TN in the effluent shows that N using increased for biomass production with longer HRTs in the reactor. Hammes et al. [4] obtained effluent water containing 695 mg COD L^{-1}, 219 mg NH_4–N L^{-1} and 79 mg Ca L^{-1} by treating anaerobic reactor wastewater of 780–1200 mg of COD L^{-1} and 480–1000 mg of Ca L^{-1} using sequencing batch BCR with a HRT of 8 h and dosage of 1000 mg urea L^{-1}. However, in the study [3] performed by the same group, optimum HRT was reported as 48 h in batch study and this value was used as optimal HRT through semi continuous studies mentioned in the same article. The decreasing of HRT could be attributed evolution of urease activity in the reactors as reported by Hammes et al. (4)

Pastor et al [12] studied the influence of the hydraulic retention times on the struvite precipitation process in the reactor. They found that the precipitation efficiencies obtained working at HRT values of 11.1, 7.1 and 2.1 h were 70.4, 69.8 and 71.4, respectively with showing no effect on the process efficiencies. However the crystal size distribution was affected by a change in HRT. The proportion of particles between 10 and 100 μm increased at low HRT. The decrease in HRT changed the hydraulics of the reactor causing a decrease of the crystal size. In the present study, this effect of lower HRTs may cause decreasing of calcium removal efficiencies due to unsuitable liquid/solid separation.

Fig. 3 shows the increasing of alkalinity, ammonium ion, and pH with increasing of HRTs. COD removal results in CO_2 production leading to alkalinity raising. Furthermore, ammonia and CO_2 production as a res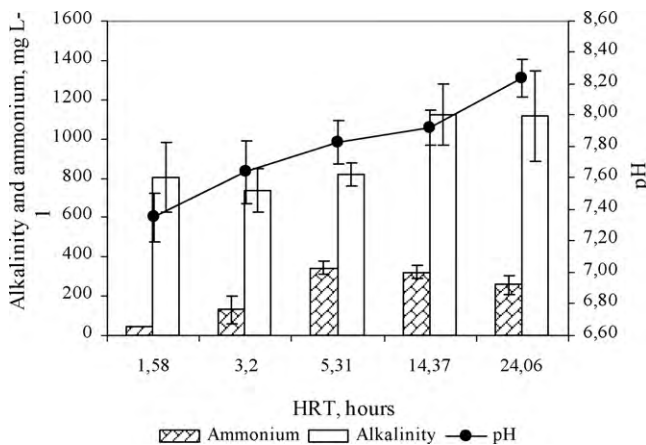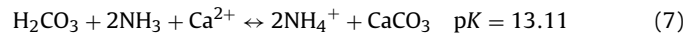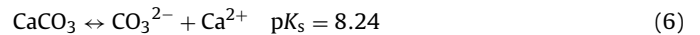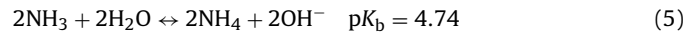ult of urea hydrolysis caused pH and alkalinity increasing. Urea hydrolysis and $CaCO_3$ production are given in equations below.

$$CO(NH_2)_2 + H_2O \rightarrow NH_2COOH + NH_3 \quad (1)$$

$$NH_2COOH + H_2O \rightarrow NH_3 + H_2CO_3 \quad (2)$$

$$H_2CO_3 \leftrightarrow HCO_3^- + H^+ \quad pK_{a1} = 6.37 \quad (3)$$

$$HCO_3^- \leftrightarrow CO_3^{2-} + H^+ \quad pK_{a2} = 10.33 \quad (4)$$

$$2NH_3 + 2H_2O \leftrightarrow 2NH_4 + 2OH^- \quad pK_b = 4.74 \quad (5)$$

$$CaCO_3 \leftrightarrow CO_3^{2-} + Ca^{2+} \quad pK_s = 8.24 \quad (6)$$

$$H_2CO_3 + 2NH_3 + Ca^{2+} \leftrightarrow 2NH_4^+ + CaCO_3 \quad pK = 13.11 \quad (7)$$
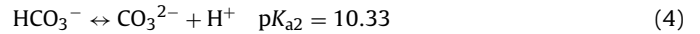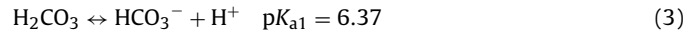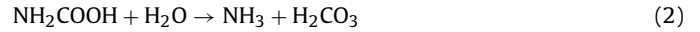
Mixed culture containing urea first hydrolyzed the urea to supply the need for N forming 1 mole of carbamate and 1 mole of NH_3 by reactions numbered 1 and 2; then 1 mole NH_3 and 1 mole of H_2CO_3 was produced from the carbamate. H_2CO_3 is a weak acid and ionized to HCO_3^- as the first step and then HCO_3^- ionized to CO_3^{2-} (Eqs. (3) and (4)) to some degree. Since the increase in pH value of water due to ammonium produced by urea hydrolysis (Eq. (5)) made these transformations more efficient, CO_2 could transform into CO_3^{2-} ion without difficulty. Consequently, great quantity of Ca^{2+} and CO_3^{2-} present in the medium precipitated as $CaCO_3$ in accordance with Eq. (6). General balance Eq. (7) could be attained by reversing and then adding Eqs. 3–6.

Organic matter removal and low urea hydrolysis rate in lesser HRT decreased the pH and alkalinity of the medium. In stable system conditions, HRT of 5.31 were suitable for urea hydrolysis and easier use of substrates. 5.94 kg COD m^{-3} day^{-1} and 4.14 kg urea m^{-3} day^{-1} loading of water in this hydraulic retention time produced Ca^{2+} removal by 72%. Effluent values of ammonium concentration (269 mg NH_4–N L^{-1}) clarified that urea was mostly transformed into ammonium in the process. Although the urea was known to be wholly hydrolyzed in the process, the reason for effluent value being less than 420 mg NH_4–N L^{-1} could be explained by the fact that ammonium was used as nitrogen source in the biomass synthesis and ammonia was volatilized from the medium [1,13]. The measurements of the nitrates (data not shown) stated that there has not been any nitrification. When HRTs were increased to 14.37 and 24.06 h, calcium removal efficiency slightly rose from optimum removal efficiency of calcium. Therefore HRT of 5–6 h was assumed optimum for the process of calcium removal.

Volatile suspended solids (VSS)/total suspended solids (TSS), TSS and sludge volume index (SVI) which are indicators of Ca^{2+} removal could be seen in Fig. 4. VSS/TSS ratio changes between 0.60 and

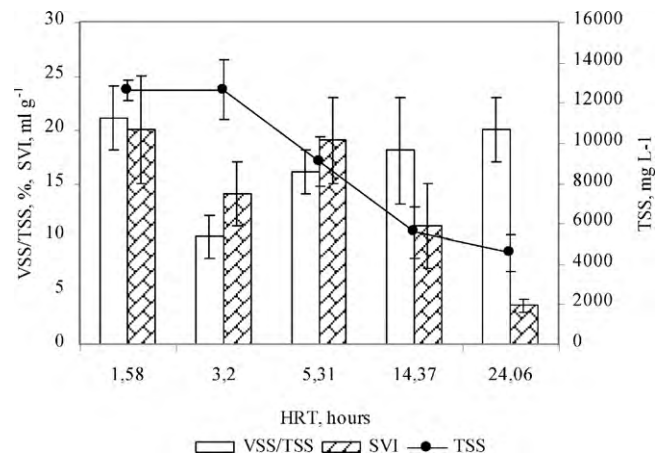**Fig. 3.** Ammonium, alkalinity and pH values in effluent.

Fig. 4. VSS/TSS (%), SVI and TSS values in BCR reactor.

0.80 in normal biomass whereas the lower ratios in sludge samples taken from the reactor, confirmed the formation of $CaCO_3$. SVI measurements showed that $CaCO_3$ in sludge increased the precipitation ratio of the sludge. An occurred calcareous sludge, which functioned both as a niche for the microbial species and as crystal nucleation sites for $CaCO_3$ precipitation, developed in the reactor. A significant increases in TSS in relation to higher organic and calcium loading rates were obtained at low HRTs, while % VSS/TSS remained between 10 and 20 at all HRTs. Low SVI values ($4-20\,\mathrm{mL\,g^{-1}}$) complied with results of a study [4] show that settle ability of this calcareous sludge is better than sludge without calcite.

Calcium was removed from the synthetic wastewater in significant ratios in this study. The other metals, especially Mg, contained in the compound may be prevented the whole precipitation of Ca^{2+}. The potential of mixed culture microorganisms hydrolyzing urea for COD removal in considerable amount is also been clarified in this study.

The disadvantage of the system, however, was the containment of the ammonium nitrogen in noteworthy amount and the necessity of the implementation of nitrogen removal methods before the discharge. The parameters other than HRT (SRT, urea and COD loading, temperature, etc.) should be optimized to investigate the applicability of the system for Ca^{2+} removal.

Acknowledgement

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