Cyclic sequential batch reactor: nitrogen and phosphorus removal from domestic sewage

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ABSTRACT

Treatment of domestic sewage by sequential batch processes has been shown to be quite attractive, mainly because it allows the maintenance of anaerobic, anoxic and aerobic conditions, which are necessary for the removal of nitrogen and phosphorus, in a single reactor. The aim of this study was to evaluate the behavior of a new reactor known as "cyclic sequential batch reactor" on the removal of organic matter, nitrogen and phosphorus from domestic sewage in a tropical climate. The removal of organic matter, nitrogen and phosphorus reached values of 90.6%, 90.5%, and 89.1%, respectively. Effluent quality of less than 8.3 mg N/L total nitrogen, 4.6 mg N/L ammonia nitrogen, and 0.8 mg P/L of total phosphorus were routinely obtained in reactor conditions as following: temperature about 26°C, a solids retention time of 12 d, a hydraulic retention time of 4 h, organic loading rate of 2.1 g COD/L d, nitrogen loading rate of 0.24 g TKN/L d, phosphorus loading rate of 0.03 g P/L d and food-to-mass ratios of 0.76 g COD/g MLVSS d.

Keywords: Biological nitrogen removal; Biological phosphorus removal; Cyclic sequencing batch reactors (CSBR); Domestic sewage; Eutrophication

1. Introduction

The concern about nitrogen (N) and phosphorus (P) removal from domestic sewage is increasing due to the negative impacts of the eutrophication process on aquatic environments which are caused by undue disposal and poor treatment of this anthropogenic waste [1]. Recently, Brazil introduced restrictive new laws and policies at all levels (federal, state and local) to increase public concern for sewage treatment and water quality. However, many wastewater treatment facilities, especially in developing countries, are only designed for chemical oxygen demand (COD) abatement, given that nitrogen and phosphorus discharge limits are not regulated by local environmental legislation; this is the case of Brazil. Additionally, the reduction of a geographical area for sewage management is driving the development of new systems and technologies. The

removal of N and P can be accomplished through physicalchemical and biological processes. Due to its lower cost and operational simplicity, the biological process is generally chosen for sewage treatment of [1-3]. Among biological processes, the activated sludge (AS) process has been widely used for the treatment of sewage from communities/groups of all sizes, mainly by sequencing batch reactors (SBR) [4-6]. Conventional biological nitrogen removal is accomplished by autotrophic nitrification under aerobic conditions, followed by heterotrophic denitrification under anoxic conditions. However, P removal is achieved by special microbial metabolism under alternative anaerobic and aerobic stages. Such conditions are performed at different stages during the SBR cycle [7-9], which reduces the cost of the treatment system. In order to provide advanced secondary treatment, the SBR reactor could be modified to also perform simultaneous nitrification, denitrification and

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organic matter removal [10-14]. Herein, requirements for different degree of oxygenation and competition for organic substrate among different functional microorganisms bring a great challenge. The nitrogen removal process relies on two steps called nitrification and denitrification. Nitrification is a process in which ammonia is biologically converted to either nitrite or nitrate, while the denitrification process biologically converts nitrite and nitrate to N gas, which is then released into the atmosphere. Nitrification requires the presence of oxygen and a longer solids retention time (SRT). The long SRT has benefit to nitrification, because of the long generation cycles of nitrifying bacteria, rather than grow very quickly. However, denitrification occurs in anoxic conditions (no free oxygen available). Recently, an SBR with anaerobic, anoxic and aerobic conditions was developed without the necessity of changes the phases during the batch. This process is known as a cyclic activated sludge system (CASS) [11,21,22]. This system is based on a compartmentalized reactor containing a bio-selector zone (anaerobic), an anoxic zone and an aerobic zone. As a result, biological sludge treatment with optimal properties has been observed. The presence of the aerobic and anoxic zone guarantees the nitrification and denitrification in the system, it means oxidation of N to nitrate and its reduction consequent to nitrogen gas. Other attractive aspect is the possibility of favoring the growth of phosphorus accumulating organisms in the bio-selector zone, making possible the more efficient removal of this important chemical from the sewage. Thus, CASS performs treatment with excellent technical properties, more compact and economic systems compared to other reactors [23]. However, CASS is a rather recent alternative and the main information comes from researchers in countries with temperate climates. The novelty of this study is to apply the cyclic sequential batch reactor (CSBR) process under different conditions not before developed. First, the higher temperature in tropical climate countries leads to an undesirable low SRT to obtain the nitrification process, and so we need to discover the minimum value when we have a high non-aerated fraction. Also, under high temperatures, the rate of nitrite formation may become higher than the rate a nitrate formation and so it is possible that the treatment system results in the accumulation of nitrite. In addition, the P removal requires low SRT and nitrification demands a high SRT, being simultaneous N and P removal a big challenge in this study. It is highlighted that our study used real sewage with P content of about 5 mg/L, which is a low value if compared to sewage from South Africa and countries in the Northern Hemisphere, where P concentrations in sewage are above 15 mg/L.

2. Materials and methods

2.1. Experimental set-up and reactor operation

Experimental assays were carried out in a pilot-scale. The treatment system was installed at the Polytechnic School of the University of São Paulo (USP, Brazil). Domestic sewage that fed the system came from the USP campus and neighborhood. First, the domestic sewage received a preliminary treatment (screening and grit removal) and then was pumped into the CSBR. Table 1 shows the main characteristics of the domestic sewage used in this study. The pilot-scale CSBR process was constructed of an acrylic aeration tank ($0.81 \text{ m} \times 0.52 \text{ m} \times 0.52 \text{ m}$ with a capacity of 150 L), a metering pump, and an air pump in conjunction with a gas flow. The reactor design adopted in this study was based on Goronszy et al. [21,23], which shows a relation of internal chambers in the order of 1:2:27 for zone 1, 2 and 3, respectively.

The domestic sewage was introduced into zone 1 of the reactor by an intermittent flow metering pump. Then, the domestic sewage was sent by gravity to zone 2, where a mixer was installed to ensure good mixing of the biological sludge. Aeration in zone 3 was done by air diffusers (fine bubbles) located at the bottom of the reactor and connected to an air compressor. The dissolved oxygen (DO) concentration was measured online using a series of 5700 oxygen probes (YSI Inc., Ohio, U.S.A.) connected to DO transmitters and controlled by a Programmable Logic Controller (PLC) coupled to an air solenoid valve. The membranes of these electrodes were changed every 8 weeks. Temperature, pH and oxidation-reduction potential (ORP) were measured online using series SC1000 connected to appropriated sensors (Hach Inc., U.S.A.).

The return of sludge from zone 3 to zone 1 was done by a metering pump, in a continuous flow, to reproduce the traditional CSBR process. Disposal of excess sludge and treated sewage was made automatically by a solenoid valve, controlled with a PLC. The volumetric exchange was 50% of the zone 3 volume. The reactor was operated in sequential batches composed of the feeding, reaction,

Table 1

Table 2

Characterization of the domestic sewage used

Parameters	Amount
pН	7.3 ± 0.5
Alkalinity, mg CaCO ₃ /L	250 ± 49
Chemical oxygen demand, mg COD/L	697 ± 146
Total suspended solids, mg TSS/L	325 ± 55
Total Kjeldahl nitrogen, mg TKN/L	81.6 ± 14.2
Ammonia (N–NH ₄), mg N–NH ₄ /L	66.0 ± 12.0
Nitrate (N–NO ₃), mg N–NO ₃ /L	0.2 ± 0.1
Nitrite (N–NO ₂ ⁻), mg N–NO ₂ ⁻ /L	< 0.3
Total phosphorus, mg P/L	7.5 ± 1.7

* Analyzes were performed in duplicate.

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Operational	conditions	of the c	yclic sec	uential	batch	reactor

Variables	Range and levels
Reactor volume, L	150
Feed flow rate, L/d	450
Recycle ratio, %	100
Solids retention time, d	12
Hydraulic retention time, h	4.0
Airflow rate, mL/min	2.0-3.0
Dissolved oxygen (aeration-zone 3), mg/L	1.0-2.0

sedimentation and effluent disposal phases. Table 2 shows a summary of the operational conditions of the reactor. Additionally, Fig. 1 shows the detailed characteristics of the CSBR experimental system.

2.2. Acclimatization stage

The performance of the CSBR was evaluated over two months of acclimatization (June and July, winter season – Southern Hemisphere) and six months of the experimental investigation stage (August to January, spring and summer seasons – Southern Hemisphere). CSBR went into operational stability in 60 d. Temperature and pH (zone 3) during acclimation were 26.0° C $\pm 2.5^{\circ}$ C and 6.9 ± 0.5 , respectively. After this stage, COD and total Kjeldahl nitrogen (TKN) removals were started.

Biological sludge was used for starting the CSBR: zone 1 was 50% filled with UASB (Upflow Anaerobic Sludge Blanket) and volatile suspended solids (VSS) were about 2,500 mg/L, while zones 2 and 3 were 50% filled with AS and VSS were equal to 3,500 mg/L. Table 3 shows the sludge concentrations maintained after the system reaches the permanent regime. The SRT was controlled daily by the withdrawal of 1/SRT sludge volume from the aeration tank. The solids loss in the final effluent was considered negligible. SRT was maintained during 12 d. Food-to-mass ratio (F/M) was controlled by adjusting the flow rate of the sewage inlet to keep the COD load proportional to the VSS mass present in the reactor. The data were recorded on a computer by a data logger. Table 3 shows the summary of CSBR start-up operating conditions and organic loading rate (OLR), nitrogenous loading rate (NLR) and total phosphorus loading rate (PLR).

2.3. Analytical methods

The analyzed parameters were: temperature, pH, COD concentration, ammonia, nitrite, nitrate, TKN, total phosphorus, alkalinity, total solids and ORP. Laboratory Table 3 Operating conditions of the cyclic sequencing batch reactors

Parameters	Amount
OLR, g COD/L d	2.10 ± 0.20
TKN-NLR, g TKN/L d	0.24 ± 0.10
NH_4^+ – NLR , g NH_4^+/L d	0.20 ± 0.05
PLR, g P/L d	0.03 ± 0.01
F/M, g COD/g MLVSS d	0.76
Anaerobic, mg MLVSS/L	$2,\!807\pm433$
Anoxic, mg MLVSS/L	$1,\!235\pm315$
Aeration, mg MLVSS/L	$2{,}736 \pm 428$
SVI, mL/g	82 ± 12
Anaerobic	$*7.1 \pm 0.5$
Anoxic	7.4 ± 0.4
Aeration	6.9 ± 0.8

* Anaerobic chamber. pH adjusted daily with sodium hydroxide to 7.0.

analyses were performed at the Sanitation Laboratory of the University of São Paulo.

All parameters were determined by protocols in accordance with the Standard Methods for the Examination of Water and Wastewater [24]. Ammonium, nitrite, nitrate and phosphorus were quantified in ion chromatography (Dionex-100, AS4A-SC). The airflow rate was measured by an airflow meter model 101325Pa. All analyzes were performed in duplicates.

3. Results and discussion

3.1. Parameters of the CSBR performance and organic matter removal

Table 4 shows the main chemical and physicochemical properties of the influent and the effluent from the CSBR



Fig. 1. Schematic layout of the cyclic sequential batch reactor used.

pH		Alkalinity	
Anaerobic	$*7.1 \pm 0.5$	Influent (mg CaCO ₃ /L)	250 ± 49
Anoxic	7.4 ± 0.4	Effluent (mg CaCO $_3$ /L)	76 ± 23
Aeration	6.9 ± 0.8	Nitrogen forms	
Influent	7.3 ± 0.5	Ammonia influent (mg NH ₄ -N/L)	66.0 ± 12.0
Effluent	6.9 ± 0.8	Ammonia effluent (mg NH ₄ -N/L)	4.6 ± 2.5
COD		Nitrite influent (mg NO_2^N/L)	< 0.3
Influent (mg COD/L)	697 ± 146	Nitrite effluent (mg NO_2^- –N/L)	< 0.1
Effluent (mg COD/L)	65 ± 23	Nitrate influent (mg NO_3^N/L)	0.2 ± 0.1
Removal efficiency COD (%)	90.6 ± 3	Nitrate effluent (mg NO_3^N/L)	0.9 ± 0.9
TSS		Total phosphorus	
Influent (mg TSS/L)	325 ± 55	Influent (mg P/L)	7.5 ± 1.7
Effluent (mg TSS/L)	19 ± 5	Effluent (mg P/L)	0.8 ± 0.5
Removal efficiency (%)	94.1 ± 3	Removal efficiency (%)	89.1 ± 5.1
ORP		TKN	
Anaerobic (mV)	-80 ± 100	Influent (mg TKN/L)	81.6 ± 14.2
Anoxic (mV)	35 ± 55	Effluent (mg TKN/L)	7.4 ± 3.5
Aeration (mV)	155 ± 76	Removal efficiency (%)	90.5 ± 5.2

Table 4 Main characteristics of the sewage before (influent) and after (effluent) CSBR treatment

* Anaerobic chamber. pH adjusted daily with sodium hydroxide to 7.0.

after six months of treatment. The pH of the influent, the sewage into all zones of the reactor as well as the effluent were stable in values between 6.9 ± 0.8 and 7.4 ± 0.4 . Total suspended solids (TSS) in the treated effluent achieved 94.1% of efficiency (final value of 19 ± 5 mg/L).

Fig. 2 shows the ORP values during the six months of treatment system operation. Literature studies indicate that ORP variation between -400 to -10 mV promotes the formation of volatile organic compounds (VOCs). These values also might imply the occurrence of methanogenesis in reactors [18,25]. The zone 1 (anaerobic) of the reactor showed ORP values about -100 ± 80 mV and probably promoted VOCs generation, which was a goal in this study because it provides a competitive advantage to PAOs. This bacterial population takes up VOCs such as acetate and propionate and store them as intracellular polymers such as poly-β-hydroxybutyrate (PHB). PHB oxidation is used to form poly-P bonds in cell storage so that soluble orthophosphate is removed from the solution and incorporated into poly-P within the bacterial cell. Cell growth also occurs due to PHB utilization and the new biomass with high poly-P storage accounts for phosphorus removal. The formation of VOCs in zone 1 was expected due to the process conduction. Additionally, zone 1 had a bio-selector chamber promoting growth/presence of facultative heterotrophic organisms because the influent exhibited readily biodegradable COD. This condition in zone 1 was maintained with rigorous pH control. In zone 2 (anoxic), ORP was circa of 35 ± 55 mV and in zone 3 (aerobic) was around 155 ± 76 mV. These values are in accordance with ORP for aerobic environments described in the literature, which ranged from 0 to 200 mV [6,25,26]. The aerobic condition in zone 3 was measured and DO concentration range of 1.0 to 2.0 mgO₂/L, ensuring good mixing of the biological sludge and the

development of biochemical processes without damage to obligate aerobic microorganisms.

Fig. 3 shows the monitoring of COD concentrations during six months of CSBR treatment. The average COD concentration in the affluent was 696 mg/L, this value can be considered high for a typical sanitary sewer, this high concentration is due to the frequent disposal of crushed organic waste in the sewer line. COD reached the final value of 65 ± 23 mg/L, being 90.6% of removal efficiency. Goronszy et al. [23] used a cyclic activated sludge system to sewage treatment and obtained a COD efficiency of 96.7%.

Values of VSS along CSBR treatment (anaerobic, anoxic and aeration zones) are shown in Fig. 4. The VSS concentration in the anaerobic zone was 2,807 ± 433 mg/L, in the anoxic zone was $1,235 \pm 315$ mg/L and in the aerobic zone was 2,736 ± 428 mg/L. Considering the COD load of 2.10 g/L d, the total useful volume of 0.150 m³ reactor and the average VSS concentration of 2.75 g/L, the F/M medium was 0.76 g COD/g MLVSS d. Figs. 3 and 4 highlighted that it was possible to maintain the effluent with COD in very low concentrations. This is important because bulking and floating sludge is a big concern in many wastewater treatments. Rezaee et al. [14,27] evaluated the variation of the sludge volume index (SVI) as a function of the mixed liquor suspended solids (MLSS) concentration and hydraulic retention time (HRT) at different aeration modes in an up-flow anaerobic/aerobic/anoxic bioreactor (UAAASB). Rezaee and collaborators [27] also showed that the high values of SVI were obtained at the highest F/M ratio (at the minimum values of MLSS concentration and HRT) and the lowest F/M ratio (the maximum values of MLSS concentration and HRT) independent of the aeration mode. The minimum SVI value obtained was 77.41 mL/g in the UAAASB with mechanical mixing when MLSS concentration, aeration mode, and HRT



Fig. 2. Monitoring of oxidation-reduction potential during the CSBR system, (a) average in time series and (b) box plot for ORP.



Fig. 3. Chemical oxygen demand concentration during the CSBR system, (a) average in time series and (b) box plot for COD.

were 10,000 mg/L, 2, and 6 h, respectively. Asadi et al. [28] evaluated the variation of SVI as a function of HRT at different DO concentrations in a continuous feed and intermittent discharge airlift bioreactor. The minimum SVI value obtained was 96 mL/g with DO of 1.0 mg O_2/L , HRT of 6 h and SRT of 12 d. In our study, the SVI value of 82 ± 12 mL/g was comparable to those values reported in the literature. The results here indicate that the operating conditions (HRT of 4h, DO of 1.0–2.0 mg O_2/L and sludge recycle rate of 100%) had not a negative affect in the sludge sedimentation.

3.2. Phosphorus removal

Fig. 5 shows the total phosphorus concentrations during six months of experimental investigation. P concentration of 0.8 ± 0.5 mg P/L in the effluent was obtained after 4 h of HRT and SRT of 12 d. Consequently, the removal efficiency

was achieved at least 89.1%. Studies in literature with phosphorus loading rates in sequential batch reactors similar to that used here reported removal efficiencies of 85% to 95% [21,23,29]. Although these studies indicated that an SRT less than 12 d is deemed necessary to complete the phosphorus removal, our study needed 12 d to achieve the best phosphorus removal. Thus, the CSBR design enables high efficiency of phosphorus removal, showing better results than those around 25%–30% of phosphorus removal by conventional treatments [15,16]. Li et al. [30] reported that *Candidatus Accumulibacter phosphatis* is the best-known PAOs widely present in full-scale plants for enhanced biological phosphorus removal.

The study by Wang and collaborators [17,19,20] also showed that initial pH of 7.8 favored a high number of PAOs, low number of glycogen accumulating organisms and high enzymatic activities (exopolyphosphatase and



Fig. 4. Volatile suspended solids concentration during the CSBR system, (a) average in time series and (b) box plot for VSS.



Fig. 5. Total phosphorus concentration during the CSBR system, (a) average in time series and (b) box plot for phosphorus.

polyphosphate kinase) if compared to initial pH 6.6. In our study, zone 3 showed pH around 6.1 in the beginning of the operation, which caused a slightly acidic biological sludge due to nitrification and low carbon availability. However, as seen, pH around 6.1 for a short time did not have a negative effect on the BPR process. Although, pH control in a higher range (pH \ge 7.5) would probably improve BPR performance in the CSBR. The pH value in zone 1 (anaerobic) was daily adjusted with sodium hydroxide, resulting in an average amount of 7.1±0.5. This value was adopted based on the literature [31–37].

3.3. Nitrogen removal

The influent and effluent profiles of nitrogenous compounds and alkalinity are shown in Figs. 6 and 7, respectively. It is emphasized that nitrite and nitrate concentrations in the influent were very low. TKN and ammonia had significant removal by CSBR with the operation of NLR 0.24 \pm 0.10 g TKN/L d and OLR 2.10 \pm 0.20 g COD/L d: TKN showed removal efficiency of 90.5%, while ammonia removal efficiency was 92.6 mg N/L. The results show that the C/N ratio was not a limiting factor for N removal. Some studies with organic and nitrogen loading rates in sequential batch reactors, similar to this study, reported TKN and ammonia removal efficiencies between 85% and 95% [23,29].

We observed a reduction in the removal rate of total nitrogen for recycles between 50%–80% and 200%–250% when the maximum removal was 62.6%. However, recycles between 100%–150% promoted removal rates of 88.0% nitrogen and 97.0% COD. Ma and colleagues [22] used a cyclic recirculation system and obtained high removal of ammonia and P when the sludge recirculation rate increased from 50% to 100%. The removal efficiencies of ammonia and total phosphorus were 91.1% and 84.7%, respectively. In this



Fig. 6. Total Kjeldahl nitrogen, ammonia and nitrate concentrations during the CSBR system, (a) average in time series and (b) box plot for TKN, ammonia and nitrate.



Fig. 7. Alkalinity concentration during the CSBR system, (a) average in time series and (b) box plot for alkalinity.

study, the recycling rate was adjusted to 100% recycle in the acclimation phase to obtain the best process performance. This condition of recycling rate resulted in N removal of 90.5% and COD of 90.6%.

Fig. 6 points out sewage nitrification and denitrification occurred efficiently. TKN and ammonia concentrations in the effluent were 7.4 ± 3.5 and 4.6 ± 2.5 mg N/L, respectively, after six months of treatment. Also, throughout the experimental time, nitrate concentrations in the effluent were smaller than 3.2 mg N/L, showing a high efficiency of the denitrification process. It probably occurred due to the sludge recycling rates of 100% in continuous flow (zone 3 to zone 1 and consequently to zone 2). As discussed previously, this recycling rate was the main factor for nitrate removal in zone 1. Thus, the applied organic load was not restrictive for biochemical processes occurrence and the DO concentration in zone 3 was sufficient for nitrogenous compounds oxidation. The DO concentration in zone 3 was established between 1.0 and 2.0 mg O_2/L during the study. More DO concentration in zone 3 was corroborated by increased ORP. The nitrification process leads to a further reduction of pH; but pH reduction may also be associated with a possible return of alkalinity by denitrification process. The effect of ammonification, nitrification and denitrification on alkalinity can be inferred by simple stoichiometric ratios, as demonstrated in the Eqs. (1)–(3) below [26].

$$(\Delta alc/\Delta N)_{am} = 50 \text{ gCaCO}_3/14\text{gN} = 3.57 \text{ mgCaCO}_3/\text{mgN} \qquad (1)$$

 $(\Delta alc/\Delta N)_{\mu} = -100 \text{ gCaCO}_2/14\text{gN} = -7.14 \text{ mgCaCO}_2/\text{mgN}$ (2)

 $(\Delta alc/\Delta N)_d = 50 \text{ gCaCO}_2/14\text{gN} = 3.57 \text{ mgCaCO}_3/\text{mgN}$ (3)

where $(\Delta alc/\Delta N)$: alkalinity change per mg N; am: ammonification; n: nitrification; d: denitrification

Alkalinity concentration in the domestic sewage was 250 ± 49 mg CaCO₃/L and after the CSBR process remained 76 ± 23 mg CaCO₃/L (Fig. 7). The theoretical alkalinity consumption estimated by the combined stoichiometric reactions of ammonification, nitrification and denitrification were 223 mg CaCO₂/L, but the experimental results showed a real consumption of 174 mg CaCO₂/L. The difference between the theoretical and experimental values depends on the ammonia and alkalinity concentrations in the influent. Our strategy was done a supplementation in zone 3 with artificial alkalinizing material to avoid biochemical limitations in the nitrification and denitrification processes. For this purpose, we considered that 1.0 mg NH₄-N/L of ammonium demands an addition of 3.57 mg/L CaCO₂ for well-adjusted alkalinity, promoting the ammonification, nitrification and denitrification processes. Alkalinity in the system without CaCO₃-addition was satisfactory only when nitrite and nitrate were in very low concentrations. Also, we highlighted that the temperature of about $26.0^{\circ}C \pm 2.5^{\circ}C$ in the sludge aeration tank created a favorable condition for the biochemical processes, especially nitrification.

4. Conclusions

This study emphasizes that high removals of 90.6% COD, 90.5% total nitrogen and 89.1% total phosphorus were possible due to CSBR operation as following: OLR of 2.10 g COD/L d, NLR 0.24 g TKN/L d, PLR 0.03 g P/L d, SRT of 12 d and HRT of 4 h. The applied SRT did not allow the nitrite formation rate to exceed the nitrate formation rate, so there was no accumulation of nitrite in the system. More importantly, the CSBR design performed a simultaneous removal of organic matter, nitrogen and phosphorus from domestic sewage. The high removal efficiencies of these compounds show that CSBR is a promising technology under environmental conditions of tropical countries.

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