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The role of extracellular polymeric substances on carbon capture in a high rate activated sludge A-stage system

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HIGHLIGHTS

- Domestic wastewater was treated in an A-stage system with varying SRT, HRT and DO.
- The effect of EPS production on bioflocculation and settling was measured.
- Highest TSS, tCOD, pCOD and cCOD removal was at 0.56 day SRT and 1.0 mg/L DO.
- EPS fractions and components did not influence COD capture and redirection.

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ABSTRACT

This paper quantifies the effect of varying solids retention time (SRT), hydraulic retention time (HRT) and dissolved oxygen (DO) concentrations on extracellular polymeric substances (EPS) production and subsequently effluent quality, carbon capture (bioflocculation) and carbon redirection (settling) in a high rate activated sludge A-stage system treating domestic wastewater. Two pilot-scale A-stage reactors were set-up with HRTs of 30 and 60 min. Cascade DO control was used to maintain 3 DO set-points of 0.5, 1.0 and 1.5 mg/L. A mixed liquor suspended solids (MLSS) concentration of 3000 mg/L was maintained and the waste activated sludge (WAS) flow was varied to achieve SRTs of 0.28 and 0.56 day. EPS fractions and the protein and polysaccharide concentrations of the mixed liquor were measured. Operation at the 0.56 day SRT and 1.0 mg/L DO resulted in the highest total suspended solids (TSS), total COD (tCOD), particulate COD (pCOD), and colloidal (cCOD) removal. The best overall performance in terms of bioflocculation (cCOD removal) and carbon capture (percent COD in the WAS) occurred at the 0.56 day SRT and coincided with decreasing total EPS concentrations but the settling characteristics of the sludge were better at the 0.28 day SRT. Overall, low correlations were found between EPS production and system performance. It is likely that at the high loading rate of the A-stage system, EPS production did not play a major role compared to the influence of operating parameters on effluent quality, carbon capture and redirection.

1. Introduction

Aerobic biological treatment is necessary to remove the organic carbon present in the raw wastewater to meet the minimum National Pollutant Discharge Elimination System (NPDES) 30 mg BOD_5/L and 30 mg TSS/L limits. In 2011, water resource recovery facilities (WRRF) in the United States consumed approximately 0.8% (30.2 billion kWh/year) of the nation's electricity and 50% of this energy was attributed to aeration [1]. This paradigm needs to shift from energy consumption through oxidation of organic carbon to energy recovery by capturing the organic carbon in the raw wastewater and redirecting to an anaerobic digester for energy production. There are two main fractions of organics in the wastewater: readily and slowly biodegradable COD (rbCOD and

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sbCOD). rbCOD is easily oxidized and harder to capture compared to sbCOD which consists of colloidal COD (cCOD) and particulate COD) (pCOD) [2]. One of the primary mechanisms for sbCOD capture is through the enmeshment of this fraction into the activated sludge floc matrix. Extracellular polymeric substances (EPS) assist with this enmeshment and also act as a buffer against oxidation of the captured cCOD and pCOD [2]. EPS are microbial secretions and products of cellular lysis and are mainly composed of carbohydrates, proteins, humics, uronic acids and DNA [3]. They are negatively charged and constitute about 50–80% of the organic fraction in activated sludge [4,5]. EPS serve as a microbial aggregate, structural backbone of the floc, and survival mechanism for microbes against turbulent conditions, dehydration, nutrient deficiency and toxic substances [6].

One technology that has been used to promote energy recovery in WRRF is the adsorption high rate activated sludge (HRAS) process. This process was developed by Böhnke and Diering [7] and is also known as the A-stage process. It is typically operated at a hydraulic retention time (HRT) of about 30 min, solids retention time (SRT) between 0.2 and 1 day, dissolved oxygen (DO) concentration below 1 mg/L and a food to microorganism (F/M) ratio of 2 to 10 g BOD/g VSS-day [8,9]. Operation at this high loading rate and short SRT may result in minimal sbCOD oxidation by restricting the rate of hydrolysis in the floc matrix and minimal decay of the active biomass [2]. Once these sbCOD fractions are captured, the sludge settling characteristics determine the separation of the solids from the liquid and the sbCOD rich solids can then be redirected to anaerobic digestion for increased energy production. However, to promote bioflocculation, capture of pCOD and cCOD and to produce biomass with good settling characteristics, A-stage operating parameters and consequently EPS production and composition have to be considered.

The EPS structure is divided into three fractions; soluble EPS (S-EPS), loosely bound EPS (LB-EPS) and tightly bound EPS (TB-EPS) [6,10–12]. S-EPS which can also be considered as soluble microbial products (SMP) are produced by microorganisms during cell growth and lysis [6]. Due to its short SRT, S-EPS in the A-stage may be associated with the influent based on microbial activity in the collection system. TB-EPS surrounds the cell while LB-EPS diffuses from this fraction and provides the primary surface for cell attachment [11].

Since bioflocculation plays a key role in achieving high quality effluent, previous researchers have investigated the relationship between the EPS production and operating parameters such as DO concentration, temperature, SRT and HRT. Jimenez et al. [2] operated a HRAS pilot-scale process with SRTs of 1, 2, 3, 5, and 10 days and found that increasing the SRT above 3 days did not improve bioflocculation or effluent quality. However, Li and Yang [11] operating a bench-scale system with glucose and acetate feed demonstrated that increasing SRT from 5 to 20 days improved bioflocculation. This same study also noted that as the LB-EPS concentration increased, settleability and bioflocculation decreased as a result of LB-EPS fraction increasing the amount of bound water between aggregates and weakening the attachment between cells. At a DO concentration of 1 mg/L, Jimenez et al. [13] reported increased EPS production from 50 to 105 mgCOD/gVSS and cCOD and pCOD removal efficiencies from 22 to 65% as the SRT increased from 0.3 to 1 day. Besides EPS concentration, the components and surface properties of EPS also affect settling and bioflocculation. Approximately 70–80% of EPS is associated with proteins and polysaccharides [4]. Increased concentration of divalent cations have shown to influence bioflocculation, floc strength, shear resistance and bound water content by binding with negatively charged hydrophobic proteins [10,14]. Polysaccharides alone have not shown much correlation with settling and bioflocculation; the ratio of protein to polysaccharides however, influences these mechanisms. High concentrations of polysaccharides that occupy a large volume of the EPS matrix may limit the influence of proteins that would enhance bioflocculation via interparticle forces [10]. Treating synthetic wastewater in a sequencing batch reactor (SBR), Liao et al. [10] reported that the ratio of proteins to polysaccharides increased from 1.3 to 5.0 as the SRT increased from 4 to 12 days. As this ratio increased (increase in total EPS concentration), settleability of the sludge decreased. Although prior research has characterized EPS production in conventional activated sludge and HRAS systems, more research is needed to explicitly investigate the effect of SRTs < 1 day, HRTs and DO concentrations in an A-stage system.

In addition to understanding how operating parameters impact carbon capture through EPS production, control strategies are needed to maximize this capture through the use of instrumentation, and automation. One common control strategy is the use of DO control which involves adjusting the aeration to maintain a DO set-point; this set-point determines if the microbes have sufficient DO for various metabolisms and keeps the biomass in suspension [15]. DO concentrations cannot be viewed individually; aerobic SRT and temperature also influence performance. Maintaining the SRT by manually controlling the waste activated sludge (WAS) flow based on the mixed liquor suspended solids (MLSS) concentration has also been successfully used in full-scale WRRF. DO and SRT control strategies are well documented for activated sludge systems with SRT > 1 day [15,16]. However, their use in an A-stage process operated with the goal of maximizing carbon capture has not been studied.

Therefore, this study investigated the effect of DO, HRT and SRT on the capture of pCOD and cCOD in 20 °C A-stage system treating municipal wastewater in a pilot-scale system through experimental studies of EPS production. This study has two novelties: the first is the use of cascade DO and WAS/mixed-liquor suspended solids (MLSS) control strategies using *in situ* online sensors with the goal of maximizing carbon capture. Second, this study is novel because it demonstrates the relationship between operating parameters and EPS production, how these variables affect carbon capture (bioflocculation), carbon redirection (settling) and effluent quality in a HRAS A-stage system.

2. Materials and methods

2.1. A-stage pilot and control

A HRAS pilot-scale A-stage system treating raw municipal wastewater consisted of 2 identical trains each with three bioreactors in series followed by a separate clarifier. The system was fed with screened (2–3 mm openings) and degritted wastewater and the temperature was adjusted to 20 °C using submersible heaters (OEM OTS, Minneapolis, MN) or a water chiller (Aqualogic MT-9, San Diego, CA). Each train (3 bioreactors) had a total volume of 511 L with a side water depth of 3.4 m. One train was operated at a HRT of 30 min while the other was maintained at 60 min. The clarifier was a 1700 L cone bottom tank with a scraper for solids removal and a surface loading rate of 16.6 and 8.3 m^3/m^2-day for the 30 and 60 min HRT, respectively. The influent and return activated sludge (RAS) flow rates were maintained equal and at 24.6 (30 min HRT) and 12.3 m^3/day (60 min HRT) using variable frequency drives (VFDs) and magnetic flow meters (Rosemount 8705, Houston, TX). Aeration in the bioreactors was provided through 17.8 cm diameter fine-pore membrane disc diffusers mounted on the bottom of each bioreactor. The DO concentration was measured using optical sensors (InsiteIG Model 10, Slidell, LA) and the DO set-point (0.5, 1.0 and 1.5 mg/L) was maintained by proportional-integral-derivative (PID) controls varying

the airflow through a mechanically operated valve (MOV; v-notch valve) receiving compressed air. Airflow was measured using a precision gas flow meter (Cole-Parmer, Vernon Hills, IL). The average influent pH was 6.79 ± 0.20 and was measured using a handheld pH meter (Beckman Coulter Φ 400 Series, Brea, CA).

In an A-stage process with low SRTs (<1 day), maintaining a constant wasting rate would result in diurnally varying MLSS concentration, even with the constant feed flow that was applied in this study, due to the changing COD loading that resulted from normal diurnal variations in COD concentration. For constant waste rate operation, the MLSS concentration would decrease during periods of low COD load, and when COD subsequently increases, the system would be effectively biomass limited with respect to the new COD load. In a conventional activated sludge system by maintaining a constant wasting rate, the change in MLSS concentration due to COD loading is not as rapid (in comparison with the SRT), therefore, even though the MLSS concentration changes, the biomass is able to accommodate the change in COD loading due to the long SRT (3–15 days). A-stage is quite different, because the process SRT is substantially less than the period of diurnal COD load variation. In the A-stage, Miller et al. (in review) determined that controlling based on a constant MLSS concentration by automatic variation of wasting rate minimizes biomass limitations as the COD loading changes over diurnal periods, wet weather, seasonal variation, etc. This is also quite different than various methods of automated SRT control, which adjust waste rate to achieve a constant SRT set-point. Again, Miller et al. [17] determined that constant MLSS control was more appropriate than constant SRT control for A-stage, providing stabilization of COD removal and more consistent effluent quality. This is because constant SRT control makes the system inherently unstable with periods of COD variability at a longer time scale than the SRT itself, making the process biomass limited in periods of high COD load and biomass rich (and their higher oxygen demand and COD removal) during periods of low COD load. A conventional constant SRT control system would therefore be effectively impossible to tune in an A-stage process without sophisticated machine learning techniques that make some prediction of normal COD load variation. Therefore, for this study, the MLSS concentration was maintained using PID controls by adjusting the WAS flow rate using a peristaltic pump (Masterflex L/S, Vernon Hills, IL). This resulted in the SRT being maintained between 0.1 and 0.5 days. The MLSS concentration set-point was 3000 mg/L and was measured using optical MLSS sensors (s::can, Vienna, Austria). A-stage systems are typically operated at MLSS concentrations of 1000 to 5000 mg/L. For this study, the 3000 mg/L MLSS concentration set-point was chosen based on previous studies at this pilot system for two reasons; (1) DO, SRT, OUR and MLSS are interrelated and affected bulking of the sludge and (2) COD removal efficiency did not increase beyond this MLSS concentration [18].

A schematic of the A-stage bioreactors and control logic is shown in Fig. 1. Influent, effluent, mixed liquor and WAS samples were collected on weekdays and measurements of COD, total suspended solids (TSS) and VSS were performed as described below. Influent characteristics shown in Table 1 represent averages of 3 weeks' sample analysis at each SRT and DO condition collected over 9 weeks of operation.

2.2. EPS extraction and analysis

Twenty-four-hour composite samples of the mixed liquor and WAS samples were collected once a week and analyzed for S-EPS, LB-EPS and TB-EPS. A heat extraction method was adapted from Li and Yang [11], with modifications. Briefly, the phosphate buffer saline (PBS) solution was diluted with tap water to match the wastewater conductivity and the centrifugation speed was

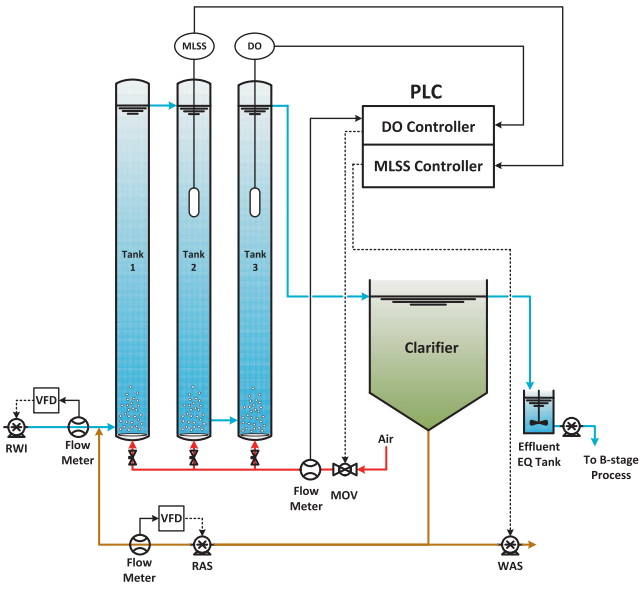

Fig. 1. A-stage bioreactors and control logic.

increased to 6000 G s^{-1} for 10 min for all extractions (S, LB and TB) to ensure a tight sludge pellet. All EPS fractions were analyzed for COD [19], proteins [20] and polysaccharides [21].

2.3. Analytical methods

Twenty-four-hour composite samples of the influent, effluent, mixed liquor and WAS samples were collected every weekday. Standard methods [19] were used to measure TSS and VSS (2540 D), sludge volume index (SVI) (2710 D), total COD (5200 B) and sCOD (filtered through a 1.5 μm glass microfiber filter). pCOD was calculated as the difference between total COD and sCOD. Flocculated and filtered COD (ffCOD) was measured by taking 100 mL of sample and adding 1 mL of $ZnSO_4$ and 25% NaOH to bring the pH to 10.5. The sample was filtered through a 0.45 μm cellulose membrane filter soaked in deionized water for at least 24 h. The cCOD concentration was calculated as the difference between ffCOD and sCOD. During the SVI_{30} analysis, the supernatant was analyzed for settleometer decant TSS (TSS_{sd}). The TSS_{sd} results provide sludge bioflocculation characteristics. Mixed liquor samples were analyzed for filamentous organisms using the methods described in Jenkins et al. [22].

2.4. Data analysis

The percent COD in the WAS, effluent and mineralized was calculated by performing a COD mass balance in which the influent tCOD concentration was considered equal to the sum of effluent tCOD, WAS COD and mineralized COD. WAS COD was calculated by multiplying the WAS VSS concentration by 1.5 based on an average calculated pCOD/VSS ratio of 1.5 ± 0.34 for this A-stage system over the various operating conditions. Influent and effluent tCOD were measured as described in Section 2.3. Statistical analysis was performed using SigmaPlot 12.5 (Systat Software, San Jose, CA). Statistical significance was assessed by using the one-tailed *t*-test in which parameters were deemed significantly different if the resulting p-value was < 0.05. Correlations between independent and dependent variables were assessed using Pearson Product Moment Correlation in which the correlation coefficient ranged from −1 to 1. Values closer to −1 represent a strong negative

Table 1
Average raw wastewater influent (RWI) characteristics (n = 15).

Parameter	Units	RWI	RWI	RWI
DO set-point	mg/L	0.5	1.0	1.5
tCOD	mg/L	575 ± 67	635 ± 63	595 ± 54
ffCOD	mg/L	149 ± 18	138 ± 5	155 ± 16
pCOD	mg/L	345 ± 3	417 ± 68	370 ± 45
cCOD	mg/L	85 ± 20	75 ± 12	74 ± 18
TSS	mg/L	211 ± 26	250 ± 55	206 ± 30
VSS	mg/L	197 ± 29	226 ± 48	188 ± 26

relationship and values closer to 1 represent a strong positive relationship.

3. Results and discussion

3.1. A-stage process control

One of the objectives of this study was to establish control strategies for DO and MLSS using *in situ* online sensors and automation to achieve maximum carbon capture. The MLSS set point was 3000 mg/L. The HRT was set to 30 and 60 min, and 3 DO set points (0.5, 1.0 and 1.5 mg/L). Table 2 provides the average volumetric loading rate (VLR), SRT, airflow, DO and MLSS concentrations achieved in this study. The average SRT of the 30 min and 60 min HRT across all DO concentrations was 0.28 ± 0.05 days and 0.56 ± 0.11 days, respectively. The MLSS concentrations were relatively stable for each operating condition as all conditions produced a variance of <400 mg/L. The lower MLSS concentrations found at the DO set point of 0.5 mg/L may have been due to sensor calibration issues which were fixed.

Airflow was bound between 20 and 90 standard liters per minute (SLPM) to maintain well mixed reactors and mimic full-scale aeration oxygen transfer rates (OTR) which are typically limited to 150 mg/L-h. Each DO set-point was maintained within the bounded airflow range with the exception of when the system was operated at the 0.28 day SRT with a DO set-point of 1.5 mg/L. During this scenario the maximum airflow rate was adjusted to 120 SLPM to maintain the DO set-point. Due to the high loading rate of this system, the change of fouling factor with time was monitored to ensure sufficient oxygen transfer efficiency (OTE). The diffusers were also cleaned weekly to prevent fouling. At the 0.28 day SRT, the airflow rates for DO set-point of 0.5 and 1.0 mg/L were similar ($p = 0.5$) (Table 2) and this may have been attributed to DO sensor fouling. In addition to air-blast cleaning that occurred every 10 min, daily manual cleaning of the sensors was performed to prevent biofouling.

3.2. Overall performance

The overall performance for each operating condition for the A-stage HRAS pilot was evaluated in terms of TSS and COD removal, fate of COD, bioflocculation and settleability. Bioflocculation was characterized as cCOD removal efficiency and TSS_{sd} concentration. Settleability is characterized by SVI_{30}. The average performance of each operating condition is shown in Fig. 2. A COD mass balance was performed to evaluate the fate of COD; the results are illustrated in Fig. 3. From these results three conclusions can be made.

First, operating at a longer SRT resulted in slightly better COD removal efficiencies but settling characteristics of the sludge (SVI) deteriorated with increasing SRT. An average filament abundance of 3.2 ± 0.4 was found when operating at the 0.56 day SRT which was significantly higher ($p < 0.001$) than 1.9 ± 0.5 when operated at the 0.28 day SRT. *Thiothrix* sp. I was the most dominant filamentous bacteria found when operating at the longer SRT whereas operating at the shorter SRT varied primarily between *Thiothrix* sp. II and Type 1863. The highest SVI values were reported at an SRT of 0.56 day and DO concentration of 1.0 mg/L. According to Jenkins et al. [22] *Thiothrix* sp. I presence in activated sludge is caused by septicity and nitrogen deficiency. Since, the process was not nitrogen deficient (data not shown), the septic environment of the sewer system may have resulted in the presence of filamentous organisms. However, the shorter SRT may have promoted washout of filamentous organisms, resulting in lower SVI values compared to the higher SRT. Different types of filamentous organisms prefer to grow under low DO concentrations, therefore, by increasing the DO concentration to 1.5 mg/L at the 0.56 day SRT the SVI values decreased from 155 ± 50.5 mL/gTSS to 109.5 ± 9.58 (Fig. 2).

Second, TSS_{sd} concentrations decreased while cCOD removal efficiency increased for both SRTs as the DO concentration increased from 0.5 to 1.0 mg/L but remained stable as the DO further increased to 1.5 mg/L. The highest average percentage cCOD removal (60.2% ± 15.8) was observed at an SRT of 0.56 day and a DO of 1.5 mg/L. These results are similar to other studies that showed cCOD removal increased as the SRT increased [13,23]. Li and Yang [11] also reported decreasing TSS_{sd} concentrations as the SRT increased from 5 to 20 days. Providing a longer SRT may provide additional time for the particles and flocs to interact. However, the longer SRT may accommodate filamentous organisms' growth that can act as a structural backbone for flocs resulting in increased floc diameter [24]. This increased floc diameter may act as a sweep floc capturing dispersed particulate and colloidal matter in the clarifier, hence increasing bioflocculation (decreased cCOD in the effluent and TSS_{sd} concentrations in the supernatant). Unfortunately, the elongated and rigid structure of the filamentous organisms deteriorates the ability to produce a compact sludge blanket resulting in increased SVI values.

Lastly, results from the COD mass balance performed for each scenario indicated that the highest average percent COD in the WAS was observed at an SRT of 0.56 day and a DO of 1.0 mg/L. However, the differences in% COD captured in the WAS were not statistically significant between the 3 DO set-points ($p = 0.15$). These results indicate that operating at the longer SRT promoted COD capture through bioflocculation. These results differ from those reported by Jimenez et al. [13] who reported that increasing the SRT from 0.3 days to 2 days at a DO concentration of 1 mg/L

Table 2
Average A-stage HRAS operating parameters (n = 15).

Parameter	Units	DO set-point = 0.5 mg/L		DO set-point = 1.0 mg/L		DO set-point = 1.5 mg/L	
HRT	min	30	60	30	60	30	60
DO	mg/L	0.50 ± 0.04	0.52 ± 0.06	0.99 ± 0.05	1.01 ± 0.03	1.49 ± 0.07	1.49 ± 0.06
Airflow	SLPM	77.9 ± 7.4	37.9 ± 5.4	76.4 ± 7.8	43.7 ± 8.0	87.9 ± 10.7	62.8 ± 12.5
SRT[a]	Days	0.33 ± 0.06	0.52 ± 0.11	0.25 ± 0.04	0.52 ± 0.11	0.26 ± 0.04	0.61 ± 0.09
MLSS	mg/L	2679 ± 304	2571 ± 194	3514 ± 219	3540 ± 156	3409 ± 393	3383 ± 288
VLR	kg COD/m^3-d	27.7 ± 3.7	13.7 ± 1.6	30.8 ± 2.5	15.2 ± 1.5	28.3 ± 2.6	14.3 ± 1.2

[a] The specific SRTs discussed in the manuscript are average SRTs at 30 and 60 min HRTs.

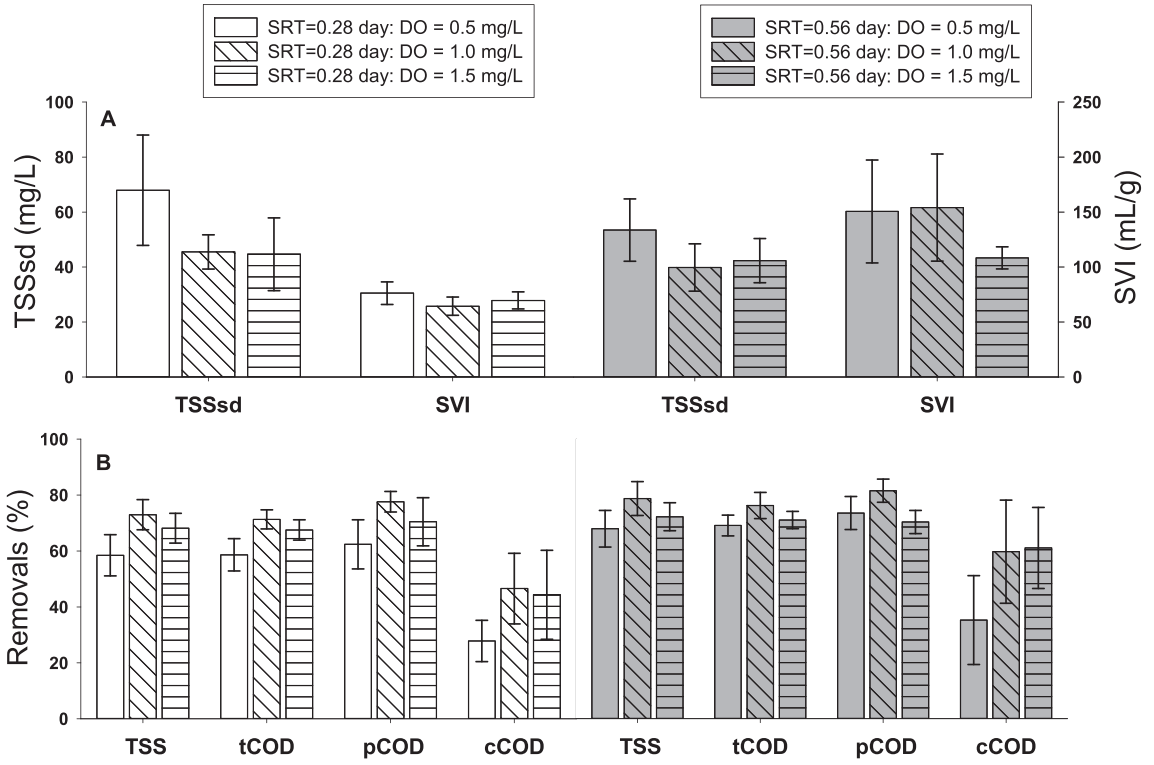

Fig. 2. Average performance of the A-stage HRAS pilot. Error bars represent the standard deviation (n = 15 per scenario).

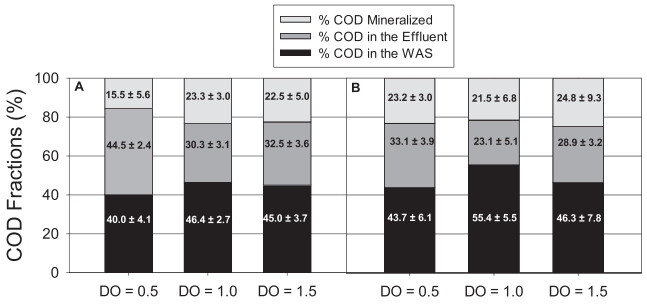

Fig. 3. Percent COD in the WAS, effluent and mineralized (n = 15 per scenario).

resulted in a decrease in the percent COD captured in the WAS. This difference in results could be attributed to different MLSS concentration and influent characteristics. Operating at the shorter SRT resulted in higher percent COD in the effluent (Fig. 3) indicating that this SRT may not have been sufficient for the microbes to utilize the influent COD. The differences in the average percent COD mineralized for each scenario were not statistically significant ($p = 0.43$). This indicates that at these operating parameters, controlling sCOD removal is difficult and other mechanisms of capturing sCOD are required such as production of intracellular storage polymers.

3.3. Effect of SRT and DO on EPS production

The average EPS component concentrations of COD, proteins, polysaccharides and the ratio of proteins to polysaccharides for the mixed liquor are shown in Fig. 4. Proteins and polysaccharides were measured as mg/L as BCA and polysaccharides but are reported as mg COD/L, assuming 1.5 mol COD per mole of BCA and 1.07 mol of COD per mole of polysaccharides.

First, at the 0.56 day SRT, increasing the DO concentration resulted in decreased proteins, polysaccharides and COD concentrations for each EPS fraction (S-EPS, LB-EPS and TB-EPS). At the 0.28 day SRT, this trend was only observed for the TB-EPS fraction and for S-EPS and LB-EPS as the DO increased from 0.5 to 1.0 mg/L. At the lower SRT as the DO increased from 1.0 to 1.5 mg/L, the proteins, polysaccharides and COD concentrations in the LB-EPS fraction increased significantly. Comparing the influence of each SRT applied in this study, across all DO concentrations, showed that operating at a longer SRT showed strong negative correlations between the DO concentration and all EPS components. At the longer SRT increasing the DO concentration negatively correlated with the protein and polysaccharide concentrations in the S-EPS (−0.83 and −0.79), LB-EPS (−0.87 and −0.75), and TB-EPS (−0.91 and −0.82) fractions. At 0.28 day SRT, the protein and polysaccharide correlation to the DO concentration was negative and below −0.60. Interestingly, the opposite occurred in relation to the ratio of proteins to polysaccharides in which all EPS fractions were positively but weakly correlated with DO concentrations. In a SBR treating synthetic wastewater at a 7 day SRT, Shin et al. [25] observed that at airflow rates of 2 and 4 L/min the protein content in the EPS did not change but polysaccharide concentrations increased. At a 0.8 L/min airflow rate (low DO), the polysaccharide and protein concentrations did not change. For this present study, the correlation data indicates that when operating at a 0.56 day SRT, DO concentrations played a more significant role on EPS production compared to operation at a shorter SRT.

Second, at the 0.28 day SRT, total COD concentrations of the extracted EPS decreased from 319.6 ± 0.1 to 227 ± 13 mgCOD/gVSS when the DO increased from 0.5 to 1.0 mg/L and from 293.3 ± 5.6 to 199.8 ± 1.6 mgCOD/gVSS at the 0.56 day SRT. Interestingly, total COD concentrations of the extracted EPS further decreased to 165.2 ± 10.9 mgCOD/gVSS at 0.56 day SRT and DO of 1.5 mg/L but increased to 245.6 ± 30.9 mgCOD/gVSS at the shorter SRT. Apart from the 0.56 day SRT and DO of 1.5 mg/L scenario, the total COD

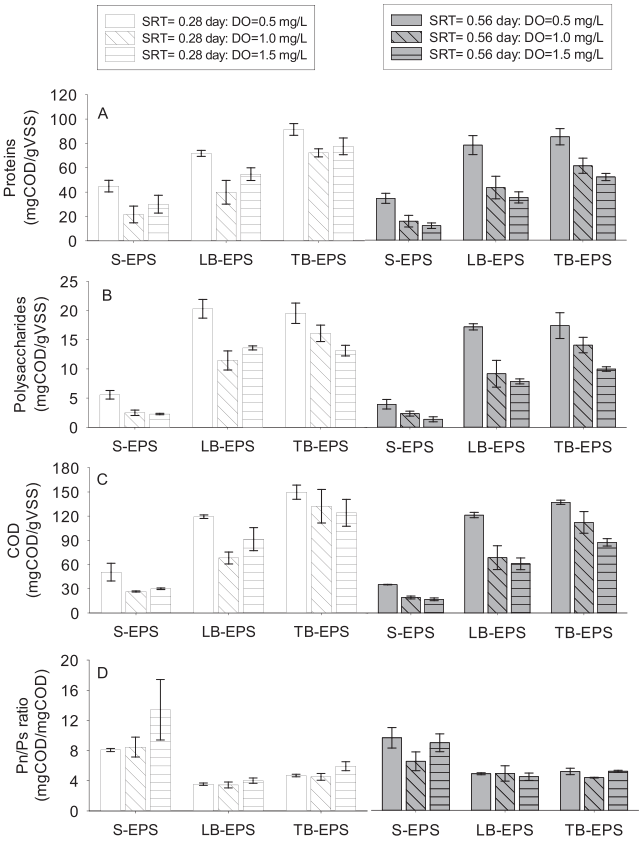

Fig. 4. Average EPS concentrations (n = 3 per scenario).

content of extracted EPS was significantly lower when the SRT was increased ($p < 0.03$). This was in contrast to findings from Jimenez et al. [13] who showed that increasing the SRT from 0.3 to 1.0 day resulted in increased EPS production from 50 ± 25 mgCOD/gVSS to 105 ± 16 mgCOD/gVSS, respectively. Liao et al. [10] did not see a clear trend in total COD content of EPS when the SRT was increased from 4 to 20 days but the ratio of proteins to polysaccharides increased as the SRT increased. The differences in trends could have been due to differences in the SRT range, wastewater characteristics and EPS extraction methods. This study used a modified heat extraction method while Liao et al. [10] and Jimenez et al. [13] used a cation exchange resin to extract EPS.

Third, for this A-stage system, the LB-EPS concentration made up 24–42% of the total EPS which was higher than the 10–20% reported in conventional activated sludge [11]. This significantly higher LB-EPS concentration could be associated with bacteria producing EPS but due to the short SRT not having sufficient time to utilize it while at longer SRTs bacteria may produce less EPS or have more time to degrade EPS for cell maintenance [23]. This may have resulted in lower total EPS concentrations for the 0.56 day SRT compared to the 0.28 day SRT.

3.4. Influence of EPS production on overall performance

The objective of this research was to understand the relationship between operating parameters, specifically, SRT, HRT and DO and EPS production and how this affects carbon capture (bioflocculation), carbon redirection (settling) and effluent quality. Previous researchers have postulated that operating activated sludge systems at SRTs < 1 day would deteriorate bioflocculation and effluent quality due to high production of LB-EPS [10,11,26]. This can be attributed to the fact that LB-EPS contains a high water content which can block binding sites on the floc [10,27]. For this present study, the LB-EPS fraction did not follow the same trend between the two SRTs investigated; therefore, performance was compared to the total EPS concentration (Fig. 5). The best overall performance in terms of bioflocculation (cCOD removal and TSSsd concentration) and carbon capture (Fig. 3) occurred at the 0.56 day SRT and coincided with decreasing total EPS concentrations (Fig. 5). However, the settling characteristic of the sludge was better at the shorter SRT. As previously discussed, decreasing the EPS concentrations would result in decreased bound water content, thus enhancing bioflocculation and carbon capture by allowing easier access to adsorption sites for pCOD and cCOD but would also deteriorate settling. Yang and Li [28] reported that increasing the LB-EPS content weakened cell attachment and resulted in an increase in SVI values. For this study the correlation between LB proteins and SVI was low at 0.49. As previously discussed poor settling at the longer SRT may have been a factor of filamentous organisms in the influent and not EPS.

The best performance at each SRT was observed at the 1.0 mg/L DO set-point which corresponded with the lowest LB/TB ratio (Fig. 5). As the DO further increased to 1.5 mg/L, the protein, COD and polysaccharide LB/TB ratio also increased and coincided with decreased TSS, tCOD and pCOD removal efficiencies. Since the only common factor associated with EPS between the two SRTs at the 1.5 mg/L DO set-point was a decrease in polysaccharides and COD in the TB-EPS, it may be that TSS and pCOD removal was associated with the TB-EPS. Conversely, bioflocculation (cCOD removal and TSSsd) remained stable as the DO concentrations increased from 1.0 to 1.5 mg/L indicating that neither operating conditions nor EPS production may have further influenced bioflocculation as the DO increased. Therefore, it is likely that operating at a DO concentration of 1.0 mg/L maximized bioflocculation, adsorption sites were fully saturated and further bioflocculation was limited by the hydrolysis rate of the adsorbed organic matter [13]. Therefore, carbon capture was likely enhanced by a low LB/TB ratio and maximized at a DO concentration of 1.0 mg/L.

Previous literature has suggested that increasing the protein content of EPS would result in better bioflocculation by decreasing the bound water content [10,14,29]. However, for this study EPS production and components may not have been the primary

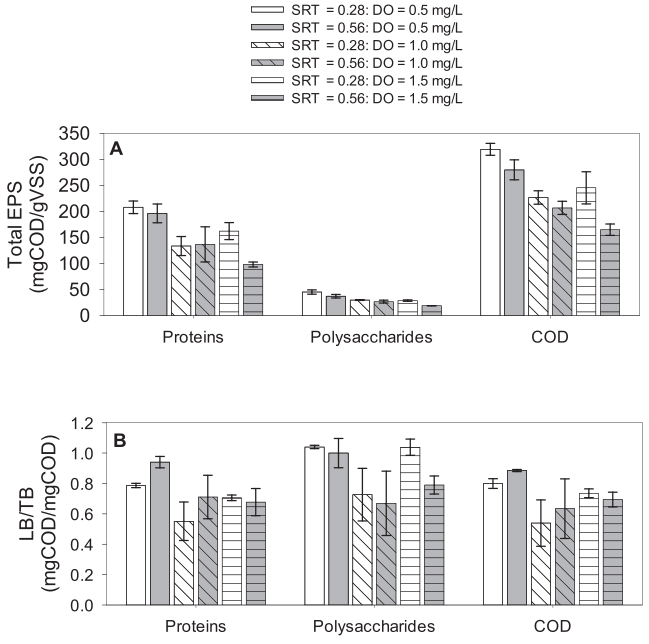

Fig. 5. Average total EPS concentrations, protein/polysaccharide (Pn/Ps) ratios and LB/TB ratios (n = 3 per scenario).

factors affecting bioflocculation but seemed to play a more significant role in the removal of TSS, tCOD and pCOD, especially when operating at the shorter SRT. Low correlations were found between EPS production and system performance, therefore it is likely that at the high loading rate of the A-stage system EPS production played a supplemental role compared to the influence of operating parameters on carbon capture and redirection.

4. Conclusions

This study combined research and development to investigate carbon capture and redirection through EPS production by varying the SRT, HRT and DO. Although cascade DO and WAS/MLSS control strategies were used successfully in maintaining the DO set-points and controlling the SRT < 1 day, EPS production did not seem to have a significant influence on bioflocculation and settling due to the short SRT. There were low correlations between EPS production at the various scenarios and performance. Since only one MLSS concentration was evaluated, future work should evaluate if varying the MLSS concentration would influence the effect of EPS on carbon capture. Operating parameters played a major role in controlling sbCOD removal and SVI values. Operation at 0.56 day SRT resulted better bioflocculation but worse settling.

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