A review on promising strategy to decrease sludge production: Oxic-settling-anaerobic/anoxic process

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ABSTRACT

Recently, as environmental regulation for the removal of nutrients and excess sludge produced through wastewater treatment has become more restricted, many wastewater treatment plants face serious challenges in terms of waste production. Nowadays, the issue of excess sludge production has received considerable critical attention. Recent developments in sludge treatment technologies have heightened the need for more promising strategies to reduce sludge levels in a cost-effective and environmentally friendly manner. The purpose of this paper is to review recent research into the oxic-settling-anaerobic/anoxic (OSA) technology for sludge minimization. The OSA process is a modification of a conventional activated sludge system with the addition of interchange bioreactor parallel to recycled activated sludge line. The OSA process seems to be a revolutionary and cost-effective alternative for sludge reduction approach in the future. It is hoped that this research will contribute to a deeper understanding of the OSA process in terms of sludge reduction efficiency, carbon and nutrient removal, operational parameters, possible reduction mechanisms and microbial community changes after the implementation of the OSA system and applied in the treatment of real wastewater at full-scale.

Keywords: Activated sludge, OSA process, sludge reduction

1. INTRODUCTION

In addition to demographic growth and increased sludge generation in sewage facilities, considerable attention has been paid to sludge management and new sludge reduction technologies developed in recent years to minimize sludge production in wastewater treatment plants (WWTPs). Sewage sludge arises as a by-product of wastewater treatment. The increase of the annual sewage sludge worldwide production is expecting to exceed 13 million tons of Dried Solids (DS) in 2020 [1]. As has been previously reported in the literature, European Union targets to reduce final waste disposal by 50% by the year 2050 comparing it to the amount of the sludge to be wasted in 2000 [2]. Treatment and disposal of excess sludge require significant amounts of energy and chemical agents, which result in significant increases in the carbon footprint and resource consumption of the wastewater treatment process [3]. The waste sludge treatment and disposal represent one of the major operating costs in a WWTP, which can vary depending on local conditions and size of the treatment plant. It is well known that land use of sewage sludge can greatly reduce the cost of disposal of sludge, but because it can contain high concentrations of metals, pathogens and trace organic pollutants and due to restricted land usage, tightening goals have been set to reduce landfilling [4]. For instance, Europe, North America, and East Asia are the main sludge producers in the world. The previous study by Kelessidis and Stasinakis [1] has emphasized that the European Union alone is generating around 50 million tons of sewage sludge annually. Moreover, Turkey has the most wasteful residents in Europe, actively disposing 32.3 million tons of household and industrial waste straight into landfills, therefore national waste management strategies should be improved and strengthened to protect environmental and human health. For example, as there are many health and environmental issues presented by landfill waste, land application as the major route for the use of sewage sludge has now been banned in several countries (Germany, Netherlands).
Various technologies can be applied in the wastewater treatment chain to reduce net partition of carbon in sludge, including physical (mechanical, thermal, electrical), chemical (adding oxidizer or uncoupler) [5], [6], and biological (with bacterial predator) [7] methods. A major issue of these processes is the high cost that should be very precisely considered during the implementation of one of these technologies. Even though anaerobic digestion is the most commonly used sludge stabilization method, it is generally limited by the poor biodegradability of waste activated sludge [8].

As the amount of excess sludge produced continues to rise throughout the world and has become a major problem for many wastewater treatment plants, a new approach is needed to develop more promising strategies to reduce sludge levels in a cost-effective and environmentally friendly manner. Minimization of sludge production in the wastewater treatment is better than the post-treatment of the sludge produced to solve sludge-associated problems [9]. An ideal approach to overcoming the excess sludge problems would be to reduce excess sludge in existing wastewater treatment plants. One of the possible solutions to create a feasible engineering approach to this problem is the modification of a conventional activated sludge process - an oxic-settling - anoxic/anaerobic process (OSA). Although this process was firstly discovered in 1964 by Westgarth et al. [10] it can be considered as new technology as we know from the recent laboratory-scale works and a few full-scale applications in the United States of America and only one in Turkey. Commercially OSA process is called the Cannibal process. As an example, the sludge reduction is up to 80 % in Rock Springs WWTP (USA) [11], in Oak Lodge Sanitary District (USA) the plant with a capacity of 40000 m³ d⁻¹ reduces sludge production by 65% [12] with Cannibal implementation. This process, in which Siemens Water Technologies’ system of OSA is used (Inegöl, Turkey), the amount of excess sludge is also significantly reduced. This revolutionary technology reduces excess sludge in a significant way, offers great saving possibilities based on operational cost aspects in the wastewater treatment plant, has a simple design, flexible operation and presents no risk to the environment. From an engineering perspective, sludge can be substantially reduced with minimal management process configuration in the conventional activated sludge process.

The goal of this research study is to develop a more rigorous understanding of a new technique – oxic-settling-anoxic/anaerobic (OSA) process, that significantly minimizes the amount of produced sludge from municipal/industrial wastewaters. Starting with a description of the oxic-settling-anaerobic/anoxic process (OSA), following by operating parameters influencing the performance, different explanations for the sludge reduction mechanisms that are still very unclear between the researchers, pollutant removal, and microbial community is going to be presented in this review paper. This paper ends with directions for the research that must be considered in the future.

2. CONCEPT OF OSA PROCESS

The OSA process is a modification of a conventional activated sludge system with the addition of interchange bioreactor parallel to recycled activated sludge line. In this system solids that would be normally wasted from the conventional system are sent to anaerobic/anoxic sludge interchange bioreactor inserted in the return activated sludge loop to minimize sludge generation and increase process reliability as shown in Fig.1.

Fig 1. Schematic representation of OSA process

Once sludge is settled, a required volume of it is being sent to interchange anaerobic/anoxic reactor and held under a unique conditioning environment (no oxygen and substrate supply) for specified detention time. Because there is no oxygen supply in the interchange reactor, the conditions can range from anoxic to anaerobic. The same volume of sludge is then recirculated back to the main aerobic bioreactor. Normally about 50% of return activated sludge passes through solid separation unit (containing ultra-fine mesh screens and hydrocyclones) in full-scale Cannibal process to remove grit, trash, and inert solids content. This content typically constitutes up 20% to 25% of the mixed liquor solids. OSA cycles equal volume of sludge between rich conditions (aeration tank) and deficient (external anoxic/anaerobic reactor/s) in oxygen and substrate [13]. During this process, greater solids destruction is achieved as the overall observed biomass yield is reduced.

There have been various configurations of OSA applications including the attachment of interchange bioreactor to the main conventional activated sludge (CAS) [21], [33], membrane (MBR) [14] or sequencing batch reactors (SBR) [19], [34] reactors presented in the literature. SBR-OSA configuration has advantages over the CAS-OSA or MBR-OSA applications due to the lower space requirement in the waterline due to the absence of the secondary settling and the intermittent sludge cycling.

2.1. The potential of the OSA process in terms of observed sludge yield

Many researchers use the observed yield (Fig. 2), and it has been recognized as an effective marker for sludge reduction [14]-[19].
Observed yield is a ratio of the amount of biomass produced to the amount of substrate consumed [20]. The slope of the linear regression is used to determine \( Y_{\text{obs}} \), then the ratio of observed sludge yield values of reference and OSA systems are used to calculate sludge reduction using the equation below:

\[
\text{Sludge reduction}(\%) = \left( \frac{Y_{\text{obs-ref}} - Y_{\text{obs-OSA}}}{Y_{\text{obs-ref}}} \right) \times 100
\]  

Previous studies have emphasized that application of the OSA process can reduce solids up to 87% more than the conventional activated sludge process [14], [21]-[25]. For instance, the following study conducted by Chudoba et al. [22] demonstrated that observed yield in the SBR-OSA system was 3 times lower, which resulted in 60% less biosolids production compared to SBR. This view has also been supported by Novak et al. [21], who reported observed yield in OSA system to be lower (0.13 to 0.29 gVSS/gCOD) than that in a conventional activated sludge process (0.28 to 0.47 gVSS/gCOD) and achieved 39% of sludge reduction of sludge yield. Comparative research by Ye et al. [23] showed biosolids reduction of 14–33% with shorter sludge age (5.5–11.5h) compared to ones applied in the study of Saby et al. [14]. In 2010, Semblante et al. [25] by increasing sludge cycling from once to four times in a day, reported a sludge reduction of 53-77%, whereas Semblante et al. [25] noted that sludge yield of OSA system was 24.8% lower than in reference system without any FeCl₃ addition to the systems at a sludge age of 10 days. Other observations indicated that sludge yield can be reduced up to 87% in the OSA system (SRT=10 days) when the highest tested ferric iron concentration of 16.05 mg L⁻¹ was present in the influent [26]. In the study of Rodriguez-Perez and Fermoño [27], the OSA system (SRT=14 days) achieved a 51.7% reduction of sludge yield, reducing the excess sludge production by 52.9% compared with the control system.

However, previous studies mentioned above can only be considered the first step towards a more profound understanding of the OSA process, because these studies reported the feasibility of an OSA process related to synthetic wastewater. This has been previously assessed only to a very limited extent because synthetic wastewater strongly differs from real wastewater due to the lack of inert organics and non-volatile solids. Recently, there has been a slight increase in studies focusing on the OSA process using real wastewater. Coma et al. [28] obtained a maximum reduction of 18.3% of the observed yield treating the whole sludge return line treating real wastewater in a UCT pilot plant. For instance, 35% of sludge reduction at the sludge age of 20 days was achieved by using real wastewater by reference [18] and his co-workers. Vitanza et al. [29] evaluated 16 months of performance of an OSA pilot plant fed with real municipal wastewater. The observed yield varied between 0.112 and 0.465 gTSS/gCOD, showing that the excess sludge reduction due to the insertion of the sludge holding tank was 49.6 ± 20.7% compared to the conventional CAS system. Apart from this, a recent study by Karlikanovaite-Balikci and Yagci [19] evaluated simultaneous sludge reduction and in an oxic-settling-anoxic (OSA) system fed with real domestic wastewater at different sludge ages. The greatest corresponding sludge reduction was achieved as 58% operated at an interchange ratio of 7.7% (1/13) in the OSA system. Recent work by Sodhi et al. [30] demonstrated the mechanism of excess sludge disruption from real tannery wastewater feed. The oxic-settling an aerobic coupled conventional activated sludge configuration confirmed around 52% of biosludge reduction. These results demonstrate a strong effect of the use of real wastewater, since it is more complex, therefore it showed lower sludge reduction compared with a study including synthetic wastewater (Table 1).

No less important is the manipulation of operational parameters that have a very strong impact on OSA process optimization and performance in terms of solid destruction and pollutant removal.

### 2.2. Carbon and nutrient removal efficiency in the OSA process

The OSA process has an impact not only on reducing sludge but also on carbon and nutrients removal (Table 1). Unfortunately, a lot of wastewater treatment plants do not meet the discharge requirements of 10 mg TN and 1-2mg/L TP and are facing real problems because of tighter discharge regulations. Simultaneous nutrient removal and excess sludge in biological wastewater treatment processes are closely related to the microbial population composition in treatment processes [31]. Most studies indicated that the phase of OSA does not adversely affect the removal of COD and total nitrogen performance [19], [30], [32], [33]. For example, data from the 2019 study by Karlikanovaite-Balikci and Yagci [19] showed that efficiency in the removal of nitrogen was approximately the same in SBR and SBR-OSA control systems with a slightly smaller concentration of oxidized nitrogen in the OSA process. The efficiency in COD removal was around 85% in CAS systems and slightly higher in OSA systems, approximately 90%. The findings are consistent with findings of past studies by Datta et al. [32], where ammonia (100%) and phosphorus (90%) removal efficiencies were found to be nearly the same as in control system and as in [30] in which COD removal efficiencies were very close in control An-CAS and CAS-OSA systems and slightly higher TN removal in CAS-OSA (74%) compared to An-CAS (81%) was found. Khursheed et al. [34] and Ye et al. [23] have also found nearly equal efficiencies in terms of COD and TN removal.
Table 1. The operating conditions and the removal of pollutants in OSA applications

| Process     | Wastewater type | SRT (d) in the whole system | ORP (mV)       | Temperature (°C) | IR (%) | Y_{obs} | Sludge reduction (%) | COD removal (%) | TN removal (%) | TP removal (%) | Reference                        |
|-------------|----------------|-----------------------------|----------------|------------------|--------|---------|----------------------|----------------|--------------|---------------|-----------------------------------|
| CAS-OSA     | synthetic      | 5                           | -250           | 18±5 20±2        | 100    | 0.21 gTSS/gCOD       | no reduction       | 82-99         | n.a            | 19-42\(^1\) Chudoba et al., 1992 |
| MBR-OSA     | synthetic      | 19.5 25.9 30.4              | +100 -100 -250 | 20               | 100    | 0.29 gTSS/gCOD       | 28                 | 93-98         | n.a            | 28-63\(^2\) Saby et al., 2003     |
| SBR-OSA     | synthetic      | 80                          | n.a            | 10 7 4           | 0.11 gVSS/gCOD | 60     | 96-97               | n.a            | n.a           | Novak et al., 2007                  |
| CAS-OSA     | urban          | 37-406 -248±133 11.5-27.7   | n.a            | 0.112-0.465 gTSS/gCOD | 49.7±20.7 | 84.9  | 63.8 ± 11.4\(^4\)   | n.a            | n.a           | Vitanza et al., 2019                |
| CAS-OSA     | synthetic      | n.a                         | -128±10        | 25±5             | 200    | 0.20 gTSS/gCOD       | 51.7              | n.a            | n.a            | Rodrigues-Perez et al., 2016       |
| SBR-OSA     | synthetic      | n.a                         | n.a            | 12.5 25 37.5 50  | 0.193-0.267 gVSS/gCOD | 3-51 | 96.4-96.9 | 70.9-80.6\(^3\) | 42.6-76.1\(^4\) Khursheed et al., 2015 |
| SBR-OSA     | a mixture of urban and glucose | n.a | n.a | 7.7 5.9 5 | 0.104 gVSS/gCOD | 52     | 90.95 | 85.2\(^1\) | 90.4\(^2\) | 92.1\(^1\) | n.a Karlikanovaite and Yagci, 2019 |
| CAS-OSA     | urban          | 60                          | -150 to -100   | n.a              | 100    | 0.212 gTSS/gCOD      | 30.4              | 85             | 88\(^1\) 51.4\(^4\) | 33.6\(^4\) Zhou et al., 2015 |
| SBR-OSA     | synthetic      | n.a                         | n.a            | 10               | 0.13 gVSS/gCOD | 38–87 | n.a               | n.a            | n.a            | Yagci et al., 2015                  |
| CAS-OSA     | synthetic      | n.a                         | 25±1           | 100              | n.a    | 14-33              | 91                | 28-30\(^1\)   | 49-58\(^4\) | Ye et al., 2008                     |
| SBR-OSA     | synthetic      | 100                         | n.a            | 10               | 0.17 gTSS/gCOD | 63     | -                 | 100\(^2\)      | 90\(^3\)      | Datta et al., 2009                  |
| CAS-OSA     | synthetic      | n.a                         | 15-35 100      | 0.25 gTSS/gCOD   | 45-80  | 70-99              | 50-85\(^5\)       | 60\(^5\)       | Corsino et al., 2020               |
| CAS-OSA     | industrial     | 23-36 -246 and +72         | n.a            | 12-15            | 0.42-0.87 gVSS/gCOD | 40.2-52.3 | 91.7 | 81\(^3\) n.a      | Sodhi et al. 2020                   |

\(^1\) TN removal; \(^2\) NH\(_3\)-N removal; \(^3\) NH\(_4\)-N removal; \(^4\) TP removal; \(^5\) PO\(_4\)-P removal
However, Saby et al. [14] observed smaller COD concentrations in the effluent of the OSA system stating that the insertion of anoxic tank favors higher COD removal. Zhou et al. [33] and Cantekin et al. [35] also clarified that due to more carbon source present, a high COD/N ratio resulted in greater TN removal output in OSA systems compared to control system. The presence of higher temperatures in the anoxic/anaerobic interchange reactor also can improve COD removal efficiency [36].

In regards to phosphorus removal, Chudoba et al. [21] pointed out that phosphate removal in the OSA type system could not be expected to exceed 50%. At the same time, Vitanza et al. [29] also found phosphorus removal at very low rate 5.1% whereas [33] also reported phosphorus removal to be lower in A+OSA 33.6% than in the control system 43.9%. Temperatures over 30°C also lowers the metabolic activity of PAO [38], therefore no PO₄-P release was observed in the anaerobic reactor and PO₄-P removal entirely due to the heterotrophic biomass synthesis resulted in poor removal efficiency close to 60% throughout experiments in the study carried out by [36]. This view is also supported by Corsino et al. [36] who stated that SBR-OSA should not be considered a good candidate for phosphorus removal. These findings suggest that, in general, the addition of interchange bioreactor to recycled activated sludge line can worsen phosphorus removal. On the other hand, these studies cannot be considered as conclusive because further evidence against [21], may lie in the findings of [34], who reported TP removal to be 76.1% and 30% higher than control reactor, when recirculation ratio was increased from 0 to 6.4 and then to 8.2gVSS/reclculated/gVSS present at average C/P ratio around 50. Phosphorus removal enhancement (48-58%) over the control reactor (48.9%) was also noticed in the study of [23] with higher substrate loading.

More information on the OSA effect on phosphorus removal would help us to establish a greater degree of accuracy on this matter. A better understanding of optimization of operational conditions that could enhance phosphorus removal needs to be developed in the future.

2.3. Operational parameters associated with OSA efficiency

Sludge interchange ratio

The sludge interchange ratio (IR) is defined to be a critical key design parameter, which strongly influences the sludge reduction mechanism and operational costs. Sludge interchange rate is the number of volumetric interchanges per day between the main and interchange (OSA) reactor.

Many studies [22], [24], [26], [38]-[40] applied the interchange ratio of 10% of biomass per day for OSA process, which was defined as the most optimal by Novak et al. [22]. Sun et al. [24] achieved an enhanced sludge reduction (from 53% to 77%) by increasing the frequency of return from once per day to four times per day while maintaining the IR between an SBR and an external anaerobic reactor at 10%. Semblante et al. [18] investigated the impact of IR on sludge reduction by the OSA process using unsettled and settled sewage. IR was varied from 0% to 22% and showed the highest sludge yield reduction (53%) in OSA-SBR comparing to reference SBR at an IR of 11%. Conceptually similar work has also been carried out by [19] in which it was found out that IR of 8% was the most optimum level resulting in 50% sludge reduction treating real domestic sewage. IRs of 9.3% and 5% revealed lower sludge reduction of 37% and 35%, respectively. This study seems to validate the view that IR of 8%, which is lower than in most studies with 10% IR, is an excellent fit for the OSA process thereby more cost-efficient. Although the above investigations examined the effect of IR varying 0%-22%, and mostly 10%, few studies ([22], [33] and 50% [36]) in the literature systematically used IR of 100%. According to Zhou et al. [33], 30.4% of sludge reduction was observed when IR was 100%, whereas 14%-33% sludge reduction was found by Ye et al. [23] and 80% was observed in Corsino et al.’s [36] study, where OSA process was combined with thermal treatment. In our view, a lot of interesting results have been reported regarding IR and this operational parameter should still be of central importance, as it is very uncertain if 10% of IR is the most optimal one.

Redox potential

Oxidation-reduction potential (ORP) is a measure of the ability of the solution to oxidize or reduce another solution. It is a widely used parameter for the on-line monitoring of characteristics reflecting many chemical and biological oxidation processes [41]. In biological wastewater treatment systems, it is often necessary to know the ORP of the various treatment basins to optimize the system. Redox potentials of less than −150 mV indicate anaerobic environments, while values greater than 100 mV indicate aerobic environments [42]. It has been suggested that the ORP level below −100 mV stimulates excess sludge reduction in the anaerobic/anoxic tank [43]. A study investigating the ORP effect on sludge reduction in the OSA process has been carried out by [14]. Authors have applied +100mV, −100mV and −250 mV of ORP values. Anaerobic ORP value of −250mV resulted in the lowest Yobs value of 0.18 gMLSS/gCOD. Sludge reduction in the OSA system can be explained by sludge decay, which is accelerated effectively under low oxidation–reduction potentials (ORP) in the anoxic/anaerobic tank [14]. Li et al. [44] suggested that proper regulation of ORP from -120 to −250 mV can effectively reduce sludge by 30-60%. The findings by [16] are in contrast with the previous studies where two ranges of ORP were established for an equal period: firstly, from −400mV to −200 mV and secondly, from −200 to +50mV. Despite prior shreds of evidence, this study resulted that alternation redox conditions from anaerobic and anaerobic caused remarkable low observed growth yield of 0.13 kgTSS/kgCOD, which was 45% lower from the yield of 0.24 kgTSS/kgCOD found by [21] and 27% lower than the one (0.18 kgTSS/kgCOD) found by [14] where ORP of −250 was kept constant. A research finding by [10] also pointed out that intermediate ORP range (−50 mV) can facilitate sludge reduction.
Therefore, it is reasonable to expect that alternation anoxic and anaerobic phases are more favorable than ORP at a certain value.

### Sludge retention time

Sludge retention time is also another very important key parameter affecting the efficiency of sludge reduction in the main and the interchange OSA bioreactors. The sludge age of the system is defined as the SRT of the main reactor. The sludge retention time in the anaerobic/anoxic reactor depends on the volume fraction interchanged between the main reactor and side-stream reactor and not associated with the total SRT of the system [19]. Semblante et al. [13] reported the sludge yield values as a function of SRT. The same researcher and his colleagues in 2016, determined the effect of SRTs varying from 10 to 40 days on OSA interchange bioreactor. Novak et al. [22] operated an SBR-OSA configuration with an SRT nearly 80 days, achieving a 60% percent reduction in sludge, but also stated that the solid loss in the SBR-OSA system was not due to the high SRT of the entire system. It has conclusively been shown that SRT varying from 10-20 days favored the destruction of the solid whereas SRT of 40 days was not effective in terms of sludge reduction. SRT of 20 days showed the highest sludge minimization with more than 35%. It is consistent with literature stating that employing long SRT in biological wastewater treatment systems can negatively impact the aerobic digestibility of activated sludge by increasing the fraction of non-biodegradable sludge [45]. The study carried out by [14] concentrated on the influence of sludge age in the MBR-OSA reactor interchange over a period of 11.1-17.4d. and there was an interchange over a period of 11.1-17.4d. and there was an 80% percent reduction in excess sludge compared to the MBR control system. Ye et al. [23] tested different values of SRT (of 5.5 h, 7.6 h, and 11.5 h) in the ASSR and contrasted the CAS-OSA system’s Y_{obs} with a CAS control system. The lowest sludge production was achieved with HRT of 7.6 h.

### Temperature

Most processes of biological wastewater treatment are temperature sensitive and higher process temperatures are more effective in reducing sludge. Temperature can influence the overall rate of hydrolysis in the reactions [46] and the increase in temperature allows for greater biomass activity. However, studies on temperature effects on the OSA process are rare to find in literature. Most of the experimental studies regarding OSA process were operated under room temperature [22], [26], [28], [29], [35]. The recent study [36] explored the impact of temperature on sludge stabilization, where the OSA process was coupled with thermic treatment at moderate temperature (35°C) resulting in high sludge reduction (80%), as well as improving sludge settling properties. Realizing the gap in the extant literature, more research is needed for exploring temperature levels and its’ impact on the OSA sludge reduction process.

#### 3. POTENTIAL SLUDGE REDUCTION CAUSES IN OSA PROCESS

The mechanisms of this biological sludge reduction method remain unclear. The key mechanism that induces sludge reduction in the OSA process remains highly controversial among researchers in the current literature. The mechanisms involved in reducing sludge yield are linked to uncoupling metabolism, enhanced endogenous decay, the domination of slow-growing microorganisms and destruction of EPS [13].

##### 3.1. Enhanced biomass endogenous decay

Conventionally endogenous decay is used to account for the cell biomass loss that is due to the oxidation of internally stored products for energy, cell death, and predation [20]. The endogenous decay phenomena result in the release of free energy from the biomass which itself becomes substrate [47]. Biomass is concentrated when it is recycled from the clarifier which contributes to the starvation, death, and lysis of some microorganisms therefore starved conditions that encourage cannibalism are created.

Karlikanovaite-Balikci and Yagci [48] used a modified version of the ASM1 model to compare and stoichiometric and kinetic coefficients with control systems to investigate the key mechanism contributing to sludge reduction in OSA systems. After a series of respirometric tests and model calibration results, it was found that the decay rate was the most vital kinetic parameter showing a significant increase after introducing the side-stream reactor into a conventional activated sludge system. The higher sludge decay coefficient in the anoxic/anaerobic OSA system suggests that low sludge production in the OSA system was due to the increased sludge decay rate in the anoxic/anaerobic OSA tank. Moreover, since not all biomass present in the reactor is active biomass, the active biomass ratio in the control SBR reactor was found to be around 75%, whereas in the side-stream reactor it was nearly twice lower. All the findings have brought a point that the OSA process is encouraging endogenous decay, ultimately decreases the viability of the biomass in the reactor and ensures excess sludge reduction in the system. This is supported by [43] who also evaluated four different (energy uncoupling, the domination of slow growers, soluble microbial products (SMPs) effect and sludge decay) possible scenarios for sludge reduction mechanism responsible for sludge reduction in MBR - OSA process. They compared the number of total bacteria and respiring bacteria before and after anaerobic treatment. The main findings from this study indicated that active biomass was reduced by sludge decay processes and it was determined to be the main cause of the sludge reduction. This kinetic coefficient was observed to be accelerating in the MBR- OSA interchange bioreactor under ORP levels lower than -100mV. Wang et al. [49] showed that cell decay contributed to 66.7% of sludge reduction in the OSA process.
3.2. Uncoupling metabolism/spilling

Energy uncoupling/spilling is characterized as a discrepancy in the energy balance between catabolism and anabolism [15]. Uncoupling metabolism can be accomplished by different methods: by the addition of chemical uncouplers [50]-[52], high So/SoX [53] or oxic-settling-anoxic/anaerobic (OSA) process [43]. Ye and Li [51] investigated the potential of the oxic-settling-anoxic (OSA) process with the addition of 3,3,4,5-tetrachloro salicylic anilide (TCS) to reduce excess sludge production. Although a higher dosage of TCS resulted in a higher reduction rate of excess sludge production, 3,3,4,5-tetrachloro salicylic anilide is proven to be xenobiotic organic matter and toxic to the microorganism and environment therefore it should be used with caution. Low sludge production can be achieved with a high So/SoX ratio, but in this case, additional treatment of organic pollutants would be required to meet effluent discharges.

Cycling from anoxic/anaerobic to an aerobic environment, microorganisms are exposed to stress conditions, facilitating the uncoupling of catabolism and anabolism [54]. The uncoupling approach is to improve the energy difference (ATP) between catabolism and anabolism to limit energy supply to anabolism. Consequently, the obtained growth yield of biomass is decreased therefore when the energy uncoupling takes place. Consumption energy for anabolism without decreasing the removal efficiency of organic pollutants in biological wastewater treatment may accordingly supply a direct method for minimizing sludge generation [55]. The phenomenon of uncoupling metabolism in CAS- OSA systems was detected by [49] although it was not significant (7.5%). With the same objective, Chen et al. [43] performed numerical experiments on possible sludge reduction mechanisms and concluded that energy uncoupling was not the case leading to sludge minimization.

3.3. Domination of slow-growing organisms

Few authors have controversial views as to whether slow-growing bacteria could influence the process of sludge reduction. Slow growth rate and high maintenance energy requirement can result in low biomass yield. The early study by [21] stated that around 60% of the total bacterial population was low yield having PAO’s (phosphorus accumulating organisms) in the CAS-OSA system. However, interestingly, this is contrary to a study conducted by [43], who studied the mechanism of selection of slow growers and reported that sludge reduction cannot be due to slow growers’ dominance.

3.4. Destruction of EPS

A complementary sludge fraction to the active cellular biomass is the extracellular polymeric substances (EPS). Extracellular polymeric substances (EPS) are a complex mixture of high molecular weight polymers, consisting of protein, polysaccharides, humic acids, lipids, and nucleic acids [56], [57] that serve as the structural framework of sludge flocs. Among them, 70%-80% of EPSs are proteins and polysaccharides [58]. The structural framework of EPS is responsible for intercellular adhesion, communication, and propagation. EPS provides physical protection from bactericides and physical stresses [59]. For instance, it is proposed that the destruction of extracellular polymeric substances (EPS) under anaerobic conditions eventually lead to sludge reduction [60]. The remnants of EPS could also serve as a substrate in the aerobic reactor, which further minimizes sludge yield [55]. In that case, EPS destruction and aerobic endogenous decay contribute to sludge reduction. The mechanism of EPS degradation is unclear, but the findings of [61] showed that the addition of α-amylase and β-glucanase improved the hydrolysis of EPS, which led to floc destruction [62]. Extracellular enzymes could play an important role in the degradation of particulate biomass and especially for EPS degradation since hydrolysis of particulate organic matter is usually the rate-limiting step in sludge degradation processes [22].

Some research offer suggestions that sludge reduction in sludge cycling schemes is rooted in the anaerobically-driven degradation of EPS into smaller forms, which are easily degraded when sludge is recycled back to the aerobic reactor [22]. Analysis of protein concentration and OUR tests on control and OSA systems was carried out by [22] resulting in higher OUR profiles in the OSA system due to higher content of readily biodegradable material.

As being a possible explanation of sludge reduction in MBR-OSA system, effects of soluble microbial products (SMP) which are a soluble portion of EPS was also studied by [43] and it was concluded that SMPs could not be the reason of sludge reduction due to no variations in Y_{s}/S found between the MBR-OSA and CAS systems.

Novak et al. [22] also showed moderate concentrations of soluble proteins (81 mg L\(^{-1}\)) in an anaerobic SSR. However, the estimation of EPS solubilization rate in the anaerobic SSR based on the soluble protein measurement is very unprecise, because the concentration of proteins is affected not only by the solubilization process but also by translocation and transportation processes. Sludge degradation extent under anaerobic conditions was increased for increased ratios of iron/sodium [60]. Park and Novak [63] showed that EPS extractable with base, thus presumably EPS attached to iron, were more solubilized than other EPS fractions during anaerobic digestion.

4. VARIATION OF MICROBIAL CULTURE POPULATION IN OSA PROCESS

Various specific molecular methodologies have been applied to document and compare the microbial culture structure and population dynamics in control and OSA process systems [21], [26], [33], [36], [64], [65]. Microbial communities can be strongly affected by diverse factors [66]. Alternating anoxic/anaerobic and aerobic environments play an important role in microbial community composition and shifts. The first serious discussion and analyses of microbial community emerged during the 1990s when [21]...
stated that OSA biomass contained 50-60% polyphosphate-accumulating bacteria. Despite prior evidence, a decade later, in 2003, [43] pointed out that slow-growing bacteria may not be the reason for solid destruction in the OSA process. In 2015, [33] and co-workers applied 454-high throughput pyrosequencing to investigate microbial community structure in A+OSA and AO systems. They identified that Proteobacteria, Bacteroidetes, Chloroflexi, Planctomycetes, and Actinobacteria were the dominant phyla. During this research, that has conclusively been shown that classes such as Anaerolineae and Actinobacteria played a major part in sludge minimization in the A+OSA process. Similarly, using the DGGE fingerprint technique it was shown that Proteobacteria and Bacteroidetes species were the most abundant in the OSA process [66]. The finding is consistent with findings of the later study by [64] using Next Generation Sequencing (NGS) of bacterial 16S rRNA gene amplicons, which showed that Proteobacteria, Actinobacteria, Bacteroidetes, and Chloroflexi were also predominant phyla in OSA systems in which their proportions in the microbial community distinguished due to the different IRs applied during the operation period. But surprisingly, Thiobrixaceae (phylum: Proteobacteria) species were not detected in the seed sludge sample, the majority of the total sequences were represented by Thiobrixaceae at the family level in all OSA systems and the genus level, Thiobrix was the predominant one. Thiobrix species are filamentous bacteria usually found in wastewater treatment plants associated with low bulking problems, but during this study bulking problems were not faced. The dominance of Thiobrix species possibly could be caused by glucose addition to the influence of real domestic wastewater. A study carried out by [25] determined the microbial community structure in SBRcontrol and SBROSA systems conducting Illumina sequencing analysis. Proteobacteria, Bacteroidetes was the most dominant phylum and γ-, β-, and α-Proteobacteria, Sphingobacteria as the predominant classes in both systems. Predatory (e.g., orders Myxobacterales and Bdellovibrion), fermentative (e.g., orders OP8, Firmicutes, WS3, and Spirochaetae) and hydrolyzing (e.g. phyla Bacteroidetes and Chloroflexi) bacteria enriched in SBRcontrol reactors. This observation indicated that these bacterial species were very likely responsible for lower sludge production. Proteobacteria members can contribute to release intracellular compounds and then Bacteroidetes may use the secondary substrate produced by those species for hydrolytic fermentation to boost their abundance [67].

Moreover, during the OSA process sludge settleability is improved [14]. Rodriguez-Perez and Fermosto [27] investigated the impact of the OSA process on protozoa diversity and filamentous bacteria. Based on the results, while the increase in floc-forming bacteria was detected in the control reactor, there was none in the OSA process. The study has shown that an improvement in a decrease in protozoa diversity, stable development of filamentous bacteria and better sludge settling can be achieved by the OSA process. Corsino et al. [36] investigated the feasibility of couple a of conventional OSA process with a thermic treatment at moderate temperature using acetate-based synthetic wastewater. When the OSA process was operated at room temperature, the amount of filamentous bacteria Thiobrix and Type 0914 significantly increased compared to seed sludge. When the temperature was increased to 35°C, the amount of these types of bacteria was significantly suppressed and better sludge settling properties detected.

5. CONCLUSIONS

The study has gone some way towards enhancing our understanding of sludge reduction by simply modifying a conventional activated sludge system with the insertion of interchange bioreactor into a sludge return line. To summarise, this review paper has shown the overall performance of the OSA program from various aspects: sludge reduction, pollutant removal, microbial population changes and dominance, operational parameters associated with OSA efficiency and future research directions. The OSA process seems to be a revolutionary and cost-effective alternative for sludge reduction approach in the future. There are only a few real full-scale applications right now in the World and limited data in the literature, future research must be undertaken to understand the mechanism and modeling of the process using real wastewater. Besides, the fate and removal efficiencies of some pollutants such as nitrogen, phosphate, and sulfate are needed to be investigated. The information provided in this paper could be helpful for evaluating different possibilities for the realization and management of a wastewater treatment plant's entire strategy, in estimating costs and besides, environmental impacts and benefits.

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