Journal of Applied & Environmental Microbiology, 2015, Vol. 3, No. 2, 31-37
Available online at http://pubs.sciepub.com/jaem/3/2/1

DOI:10.12691/jaem-3-2-1

Effect of Dissolved Oxygen Concentration on Morphology and Settleability of Activated Sludge Flocs

Melvin-Guy Adonadaga[1,2,*]

[1]Department of Earth and Environmental Science, Faculty of Applied Sciences, University for Development Studies, Box 24 Navrongo, Ghana
[2]Department of Biotechnology for Water Treatment, Faculty of Environmental Sciences and Process Engineering, Brandenburg University of Technology, Cottbus 03046, Germany
*Corresponding author: madonadaga@gmail.com

Received March 01, 2015; Revised March 22, 2015; Accepted March 25, 2015

Abstract Efficient solids-liquid separation of the activated sludge wastewater treatment method depends on the formation of flocs in the biological step, and their rapid settling and proper compaction in the secondary clarifier. The effect of dissolved oxygen (DO) concentration on sludge floc morphology and settleability in three full-scale conventional activated sludge plants treating industrial and municipal wastewater was studied. Floc structure was considered in terms of size, shape, strength and filament index, while settleability was determined based on the sludge volume index (SVI). Generally, higher DO concentrations produced more round and compact flocs, while irregular and open flocs were associated with low DO concentrations. There was only a trend towards larger flocs at higher DO levels. SVI values did not correlate well with DO concentrations, with municipal plants operated at lower DO levels achieving better settling compared to industrial plants operated at higher DO levels. However, there was a positive relationship between SVI and levels of suspended solids in the effluent. Additionally, flocs with the same filament index but from different plants were morphologically different. These results indicate that activated sludge floc morphology and settleability are affected by other plant operating conditions, not DO exclusively.

Keywords: *activated sludge process, dissolved oxygen concentration, floc morphology and settleability*

Cite This Article: Melvin-Guy Adonadaga, “Effect of Dissolved Oxygen Concentration on Morphology and Settleability of Activated Sludge Flocs.” *Journal of Applied & Environmental Microbiology*, vol. 3, no. 2 (2015): 31-37. doi: 10.12691/jaem-3-2-1.

1. Introduction

The conventional activated sludge process consists of an aeration tank followed by a sedimentation tank (secondary clarifier) in which the solids-liquid separation occurs. This settling stage should produce an effluent with concentrations of suspended solids low enough to satisfy discharge standards, and a thickened sludge to maintain the required concentration of activated sludge in the aeration tank [1]. The properties of the activated sludge flocs formed in the aeration tank are dependent on plant operation conditions such as organic loading, dissolved oxygen (DO) and solids retention time (SRT) [2]. These properties determine the settleability of the flocs, measured as sludge volume index (SVI). A sludge with an SVI greater than 150ml/g is considered a bulking sludge as it does not settle properly [3]. Compact, round and firm flocs are desirable since they ensure good settling and dewatering, with the subsequent effect of an effluent low in suspended solids produced [4]. On the contrary, large, irregular shaped and weak flocs have low settling velocity and poor compaction, due in part to their large surface area and also the large amount of bound water trapped within them. The resultant effect is a reduction in the capacity of the sedimentation tank as well as a highly turbid effluent.

Accordingly, considerable efforts have been made over the years to understand how different factors affect floc structure in the light of its importance on sludge settleability [5,6]. Dissolved oxygen is indispensable to the activated sludge method since it is an aerobic process. According to [7], the dissolved oxygen level in the aeration tank should be maintained at 2mg/l or higher to ensure a balance between the consortium of microorganisms in the aeration tank. Based on the substrate diffusion limitation theory, [8] argued that irregular floc morphology should be associated with dissolved oxygen limitation, while more round and smooth flocs should abound when oxygen gradient is not important. Also, [1] reported that low DO concentration resulted in the production of porous flocs with poor settling, and that there was no clear effect of DO concentration on floc size. Similarly, [9] reported that decreasing the DO concentration in the bulk liquid had a strong negative effect on sludge settleability.

SRT, also referred to as mean cell residence time or sludge age, is the average amount of time that microorganisms are maintained in the aeration tank. This parameter is calculated for each treatment plant based on the sludge loading level, and maintained by removing a calculated amount of biomass (typically new growth) from the system in the form of wasted activated sludge [10]. Based on microscopic observations, [5] found out that lower SRTs resulted in production of irregular and cylindrical shaped flocs, whereas flocs were more compact and round at higher SRTs.

Additionally, filamentous bacteria, considered as providing a skeletal matrix to which flocs adhere [11], have been reported to be affected by DO concentration. For instance, a decrease in DO negatively affected solids-liquid separation in a sequencing batch reactor (SBR) due mainly to dominance of slow growing filaments [9]. Similarly, low DO concentration has been reported to result in excessive growth of filamentous bacteria, affecting settling of flocs [1,14]. An increase in filament numbers can result in a decrease in settling velocity of the flocs, causing the SVI to deteriorate [12,13].

However, most of these studies were either carried out on laboratory scale, or on plants with different configurations [1,5,9]. In addition, reports on comparative studies of DO effects on sludge structure and settleability in completely mixed industrial and municipal plants in different geographic regions are limited. Therefore, the purpose of this study was to 1) investigate how DO concentration affects the morphology and characteristics of activated sludge flocs in full-scale WWTPs, and 2) determine the effects of floc morphology on sludge settleability and effluent suspended solids levels.

2. Materials and Methods

2.1. Sample Collection

Mixed liquor and effluent samples were collected from three full-scale activated sludge wastewater treatment plants (WWTPs) in Accra, Ghana. Two of these plants treat wastewater from hotels, while the other treats wastewater from a food and beverages processing factory. Interviews with plant operators showed that all the plants operate at high SRTs; the industrial plant at about 30 days and the municipal plants even higher. Each treatment plant was sampled monthly from May to July 2013. Representative grab samples were collected from areas of good mixing during aeration using long-handled aluminum dipper attached to a wooden handle. Effluent samples were collected from the outlet pipe of the secondary clarifier. Samples were stored in pre-sterile plastic containers which were appropriately labeled using waterproof marker. Samples were stored in an ice chest at a temperature below 4°C in order to reduce any change to their characteristics, and transported to the laboratory within 2 hours of sampling.

2.2. On-site Measurements

Dissolved oxygen concentration and sludge settleability of the mixed liquor were determined on-site. Dissolved oxygen level was measured using a portable Hach dissolved oxygen/pH meter (Model HQ 20), which was calibrated before measurements were taken. Settleability of sludge was measured as SVI, according to standard methods [15]. 1000ml of freshly sampled mixed liquor was poured into a 1L graduated cylinder, allowed to settle under quiescent conditions for 30 minutes, and the volume occupied by the settled sludge recorded. The SVI was subsequently calculated based on the obtained mixed liquor suspended solids value.

2.3. Mixed Liquor Suspended Solids and Effluent Suspended Solids Determination

Mixed liquor suspended solids (MLSS) and effluent suspended solids (ESS) were analyzed according to standard methods [15]. An aluminum dish was placed on a dry balance and the balance reading set to zero. 5ml of well agitated sample was placed on the dish and the initial reading recorded. The sample was then dried at 103-105°C till the reading stabilized, indicating that all the moisture content has evaporated. The suspended solids value was then calculated from the differences between the initial and final readings.

2.4. Floc Characterization and Filament Index Determination

Visual characterization of activated sludge samples was done according to [13], using phase-contrast and bright-field illumination. Wet mounts of sludge was done to determine the general characteristics such as size (small, medium and large), shape and strength (round or irregular, compact or diffuse, firm or weak) of the flocs. These were examined under phase-contrast illumination at 100× magnification with a Nikon Eclipse LV100 (Japan). Floc size was determined based on the following categorization by [13]: small ≤ 150µm, medium 150-500µm, large ≥500µm. Overall filamentous organism abundance was rated using the subjective scoring technique outlined by [13]. Filament index (FI) was scored on a scale between 0 and 6 (integer scores had the following meanings: 0 = none; 1 = few; 2 = some; 3 = common; 4 = very common; 5 = abundant; and 6 = excessive). The abundance of filamentous bacteria was determined using phase-contrast microscopy at 200× and 400× magnification; the transparent nature of filamentous bacteria under bright-field hinders their visualization.

3. Results

Dissolved oxygen levels in all the treatment plants were below the minimum concentration of 2mg/l required for optimum performance of the activated sludge process. The industrial plant operated at relatively higher values (1.3-1.7mg/l) compared to municipal plants (0.4-0.9mg/l).

Table 1. SVI, DO and ESS values

	May			June			July		
	SVI	DO	ESS	SVI	DO	ESS	SVI	DO	ESS
Plant 1	90	1.3	1.1	107	1.7	2.8	147	1.6	17
Plant 2	104	0.5	1.6	168	0.4	9.7	86	0.6	0.6
Plant 3	134	0.7	7.8	115	0.9	6.2	98	0.7	0.4

SVI = ml/g, DO = mg/l, ESS = g/l
Plant 1 = Industrial plant, Plants 2 and 3 = Municipal plants.

There was no clear relation between DO concentration and SVI values. For example, SVI was 147ml/g at a DO of 1.6mg/l in the industrial plant, while SVI of 86ml/g was recorded at a DO of 0.6mg/l in the municipal plant (Table 1). However, there was a trend towards lower SVI values at higher DO concentration, especially in the municipal plants (Figure 1-Figure 3).

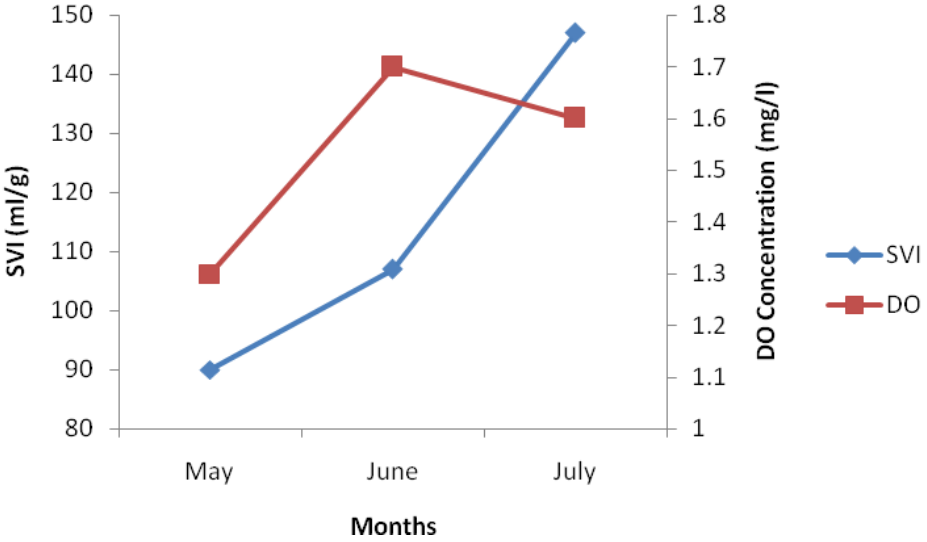

Figure 1. Effect of DO concentration on SVI in Plant 1

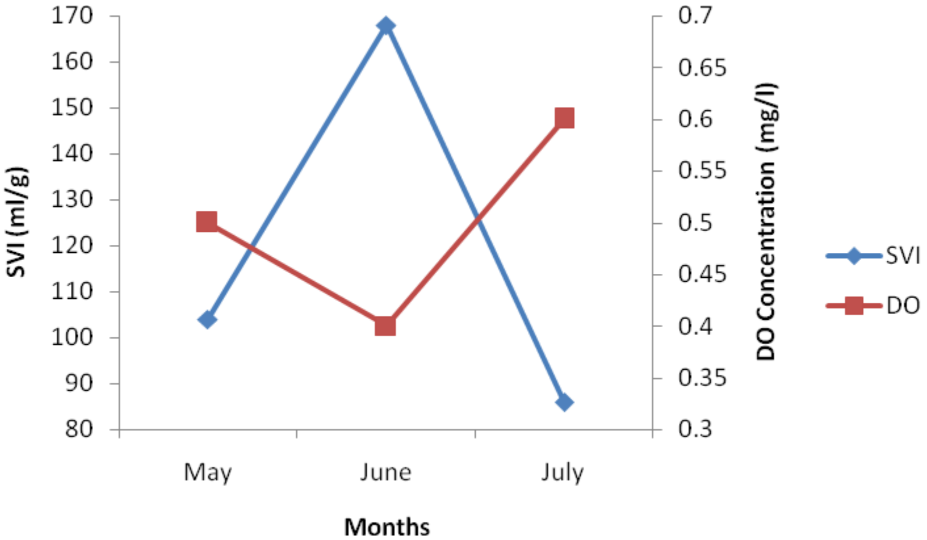

Figure 2. Effect of DO concentration on SVI in Plant 2

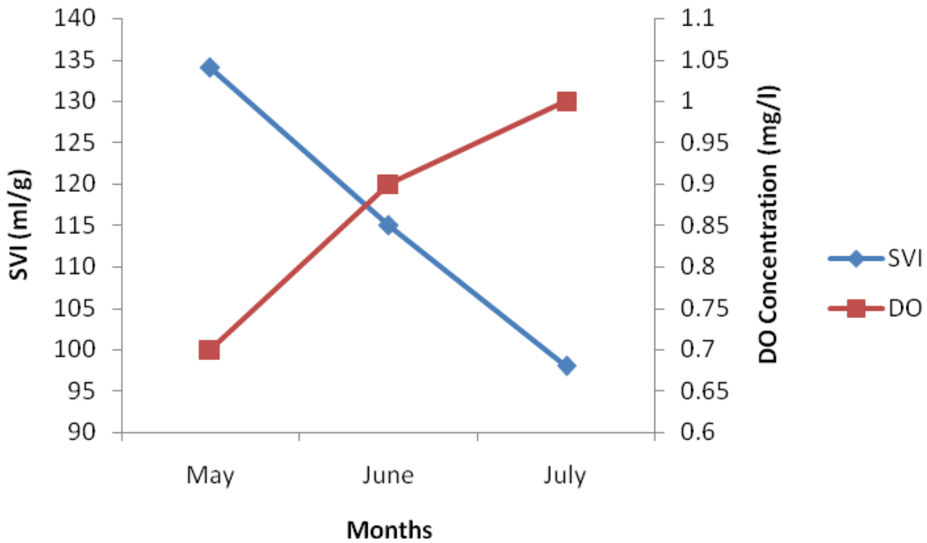

Figure 3. Effect of DO concentration on SVI in Plant 3

The amount of suspended solids in the effluent gives an indication of the solids separation efficiency in the secondary clarifier, and effectively the quality of the effluent. As expected, SVI values correlated very well with ESS levels for all plants (Figure 4-Figure 6). The lowest ESS value of 0.4g/l was recorded in municipal plant, while the highest value of 17g/l was recorded in industrial plant. However, all effluent suspended solids values were generally very high, exceeding the discharge guideline values of 50mg/l set by the Environmental Protection Agency of Ghana. No clear relationship between SRT and ESS was observed in this study.

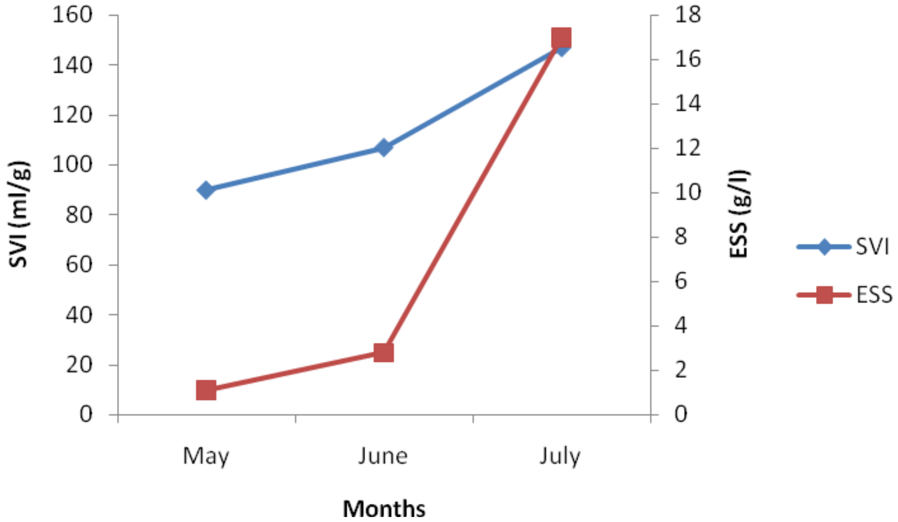

Figure 4. Relationship between SVI and ESS in plant 1

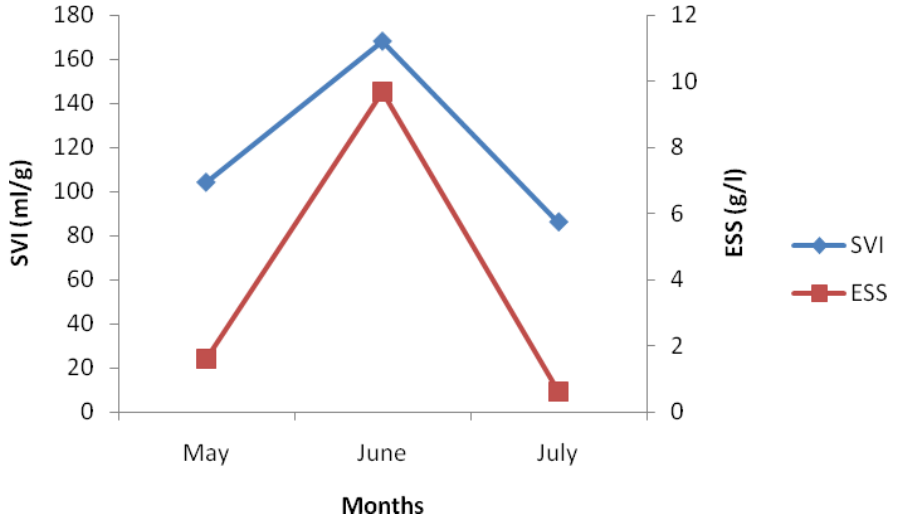

Figure 5. Relationship between SVI and ESS in plant 2

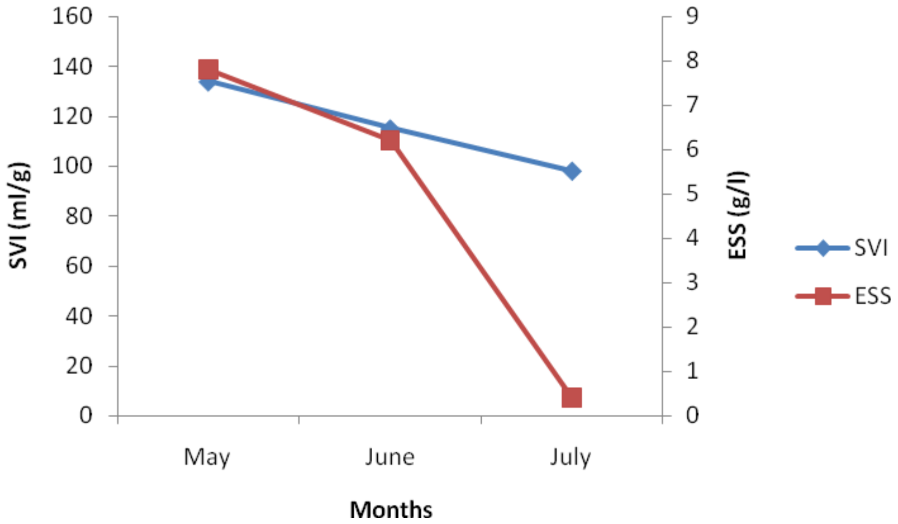

Figure 6. Relationship between SVI and ESS in plant 3

Generally, floc morphology was unique and stable in each plant throughout the study period. For the industrial plant (Plant 1), round, compact and medium sized flocs (average diameter of 185µm) were observed. Filaments could be observed occurring mostly within the floc, with a few protruding from the flocs. The overall filament index was 2 and remained unchanged (Pictures A and B). In the first municipal plant (Plant 2), pin point flocs with average diameter of 35µm were observed. Filamentous bacteria were hardly observed (Pictures C and D). This plant suffered a dysfunction with the aeration system during the study. In the second municipal plant (Plant 3), open and irregular flocs with a lot of bound water were observed. These flocs were medium size with average floc diameter of 250µm. An overall filament index of 2 was also recorded in this plant throughout the study. The filaments were observed to provide a frame to which most of the flocs attached; hence the shape of the flocs was determined largely by the filaments.

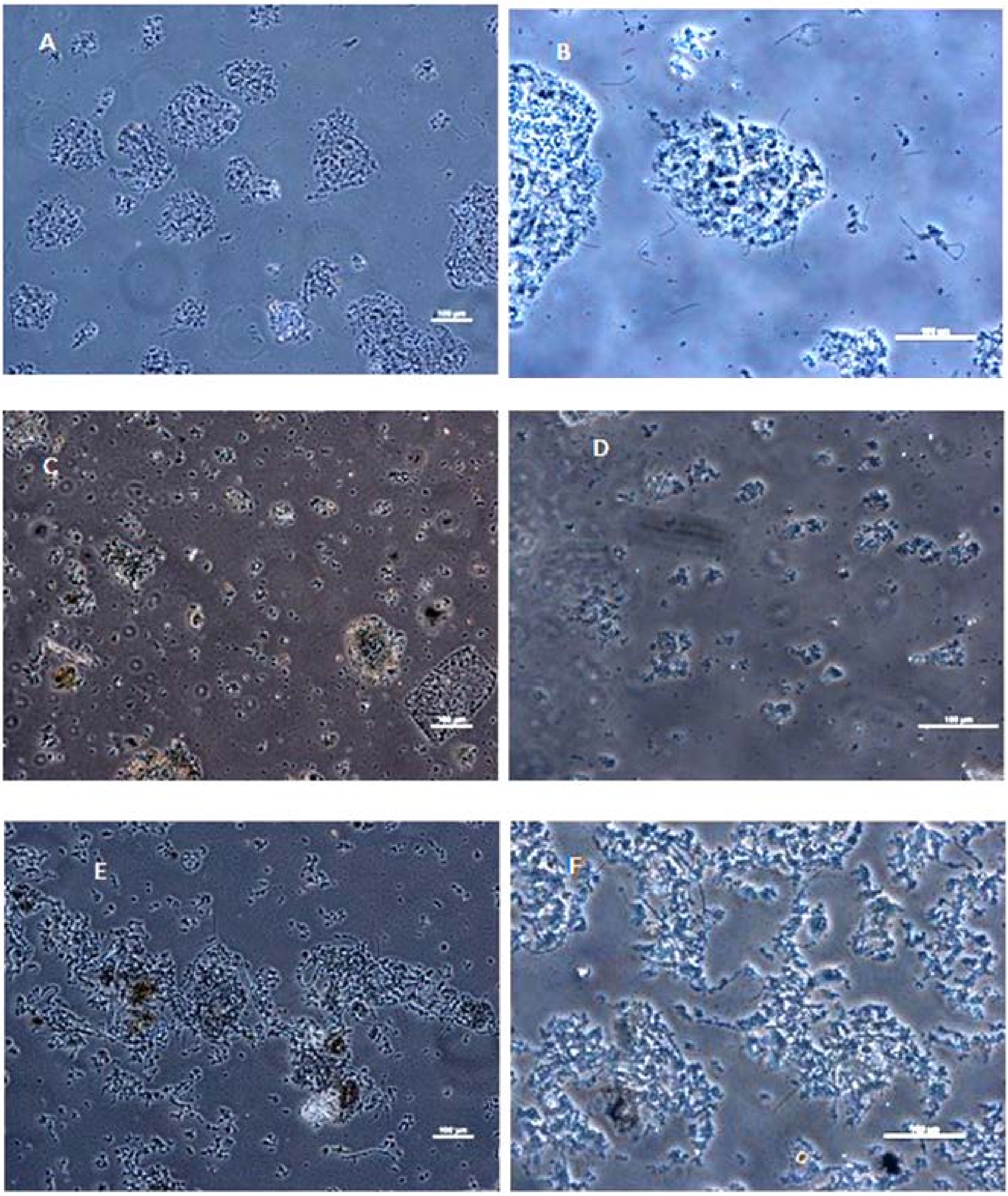

Figure 7. Photomicrographs showing flocs shape, size and filaments: (A and B) round and compact flocs containing filaments from Plant 1, (C and D) pin point flocs with no filaments from plant 2, and (E and F) open and irregular flocs with filaments from plant 3

4. Discussion

Activated sludge flocs have been described as particulate aggregates whose stability and settling properties are influenced by several factors such as amount and composition of the extracellular polymeric substances (EPS), organic and colloidal particles, cations, and the interaction forces between them [4,16]. The trend towards better settleability at higher DO concentrations is consistent with findings by [17]. However, the fact that there was no clear relationship between SVI and DO suggests that sludge settleability is affected by other factors, not DO exclusively. Complex interactions between measured individual parameters make it sometimes difficult to find clear relationships between them [18].

The positive relationship between SVI and ESS confirms the fact in the literature that good settling properties of flocs are necessary for efficient solids-liquid separation in the activated sludge process. Hence, operating parameters in the biological step should be optimized on plant by plant basis [13]. A study conducted on laboratory-scale SBR found higher ESS concentration at lower SRTs than that at high SRTs [5], which is inconsistent with the results of the current study. These contradictions further point to the differences in studies conducted under controlled and uncontrolled conditions, and the need for *in situ* studies to be preferentially considered.

Based on observations of large variations in floc morphology between sludge from different treatment plants, [4] concluded that plant design as well as changes and differences in process operating conditions have significant effects on floc morphology. For instance, [19] found that under high DO conditions, compact, robust flocs with rounded shape were produced. Similarly, [20] reported that low DO concentration in a SBR resulted in the production of porous and irregularly shaped flocs. The differences in DO concentrations in the various plants should therefore partly account for the distinct appearance of flocs from each plant in this study. The low DO in plant B should account for the general lack of filaments in the flocs, hence their pin point nature. Although DO concentration and floc size did not correlate properly, there was a trend towards larger flocs at higher DO concentrations, which is consistent with results by [1] and [17].

According to [5], “famine” conditions at high SRTs produce more stable biomass, whereas “feast” conditions at lower SRTs lead to breakage of flocs into smaller ones. This conclusion, however, is inconsistent with the results of this study. Other studies have reported that high SRT favors the growth of slow growing filamentous bacteria [21], which tend to grow outwards into the bulk solution when there is an oxygen deficiency within the floc. This is consistent with the diffusion limitation theory [11]. Therefore, the low DO concentration, coupled with the high SRT, should account for the open nature of the flocs in plant 3. It has been argued based on the filament backbone theory [11] that flocs with a high FI should be stronger than those with low FI [8]. Also, the presence of filamentous bacteria should produce stronger flocs due to the tensile strength of the filaments [22]. However, the results of this study indicate that flocs from the various plants were morphologically different, despite having the same FI. Hence, floc strength cannot be explained by only filamentous organism abundance. Other research has proved that not just filament numbers, but also filament types, affect floc structure [23]. This is because different filaments occur at different locations in the floc [4], and hence influence floc shape differently. Also, different filamentous bacteria have been found to have different surface properties which affect floc formation and structure [24]. Obviously, filamentous bacteria are important in sludge floc characteristics, hence detailed studies on how they are affected by DO concentration need to be conducted.

5. Conclusions

This research investigated the effect of DO concentration on characteristics and settleability of activated sludge flocs in full-scale municipal and industrial WWTPs. The following conclusions are made:

DO has an effect on floc morphology since more round and compact flocs were observed in the industrial plant operated at relatively high DO concentrations as compared to open and irregular flocs in municipal plants operated at low DO concentrations. However, the effect of DO on floc settleability could not be established because of the low SVI values recorded in the municipal plants operated at low DO levels. The role of other factors on floc settleability should be studied, since the investigated plants treat different wastewater types under different operating conditions. For instance, the results suggest that higher SRT has a positive impact on sludge floc settleability irrespective of the DO level. As such, operating at a suitable SRT can improve solids-liquid separation. However, further research is needed to confirm this.

Although DO concentration has an effect on filamentous bacteria abundance, filament index alone cannot be used to determine morphology and settling properties of flocs, since flocs with the same filament index exhibited different shapes and settleability. The effect of DO on different filamentous bacteria, and how this correlates with floc structure and settleability should be investigated.

SVI is a good indicator of levels of effluent suspended solids as these parameters correlated very well for all plants.

Acknowledgement

The author would like to acknowledge Ansah Emmanuel with Ecolab, University of Ghana for assistance with field sampling and general laboratory assistance.

Statement of Competing Interests

The author has no competing interests.

References

[1] B.-M. Wilén and P. Balmér, “The effect of dissolved oxygen concentration on the structure, size and size distribution of activated sludge flocs,” *Water Res.*, vol. 33, no. 2, pp. 391-400, 1999.

[2] J. D. and P. D. S. Palm J. C., “Relationship between organic loading, dissolved oxygen concentration and sludge settleability in the completely mixed activated sludge process.,” *J. Wat. Poll. Contr. Fed.*, vol. 52, no. 19, pp. 2484-2506, 1980.

[3] D. H. Eikelboom, *Process control of activated sludge plants by microscopic investigation*. IWA Publishing, 2000, p. 156.

[4] B. M. Wilén, B. Jin, and P. Lant, “Impacts of structural characteristics on activated sludge floc stability,” *Water Res.*, vol. 37, pp. 3632-3645, 2003.

[5] B. Q. Liao, I. G. Droppo, G. G. Leppard, and S. N. Liss, “Effect of solids retention time on structure and characteristics of sludge flocs in sequencing batch reactors,” *Water Res.*, vol. 40, pp. 2583-2591, 2006.

[6] Starkey J. E. and Karr J. E., “Effect of low dissolved oxygen concentration on effluent turbidity. J. Wat. Poll. Contr. Fed. 56(7), 837±843.,” *J. Wat. Poll. Contr. Fed.*, vol. 56, no. 7, pp. 837-843., 1984.

[7] J. Chudoba, “Control of activated sludge filamentous bulking. VI: Formulation of basic principles.,” *Water Res.*, vol. 19, pp. 1017-1022, 1985.

[8] A. M. P. Martins, K. Pagilla, J. J. Heijnen, and M. C. M. van Loosdrecht, “Filamentous bulking sludge--a critical review.,” *Water Res.*, vol. 38, no. 4, pp. 793-817, Feb. 2004.

[9] A.M.P. Martins, J.J. Heijnen, M.C.M. van Loosdrecht, “Effect of dissolved oxygen concentration on sludge settleability,” *Appl. Microbiol. Biotechnol.*, vol. 62, no. 5-6, pp. 586-593, 2003.

[10] J. M. Carley, “The Effects of Temperature and Solids Retention Time on Activated Sludge Treatment Performance. " Master’s Thesis,” University of Tennessee., 2003.

[11] J. D. and P. D. S. Sezgin M., “A unifed theory of filamentous activated sludge bulking.,” *J. Water Pollut. Control Fed.*, vol. 50, pp. 362-381, 1978.

[12] B. Eikelboom, D H & Geurkink, “Filamentous micro-organisms observed in industrial activated sludge plants,” *Water Sci. Technol.*, vol. 46, pp. 535-542, 2002.

[13] G. T. D. David Jenkins, Michael G. Richards, *Manual on the causes and control of activated sludge bulking, foaming, and other solids separation problems*, Third edit. florida: lewis publishers, 2004.

[14] T. Ramothokang, G. Drysdale, and F. Bux, “Isolation and cultivation of filamentous bacteria implicated in activated sludge bulking,” *Water Sa*, vol. 29, no. 4, pp. 405-410, 2004.

[15] APHA, *Standard Methods for the Examination of Water and Wastewater*, Twentienth. Washington, 1998.

[16] G. G. Liss, S.N., Droppo, I.G., Flannigan, D.T., Leppard, “Floc architecture in wastewater and natural riverine systems.,” *Environ. Sci. Technol.*, vol. 30, no. 2, pp. 680-686, 1996.

[17] J. Guo, Y. Peng, S. Wang, Y. Zheng, H. Huang, and Z. Wang, “Long-term effect of dissolved oxygen on partial nitrification performance and microbial community structure.,” *Bioresour. Technol.*, vol. 100, no. 11, pp. 2796-802, Jun. 2009.

[18] T. V. Bugge, P. Larsen, A. M. Saunders, C. Kragelund, L. Wybrandt, K. Keiding, M. L. Christensen, and P. H. Nielsen, “Filtration properties of activated sludge in municipal MBR wastewater treatment plants are related to microbial community structure,” *Water Res.*, vol. 47, no. 17, pp. 6719-6730, Nov. 2013.

[19] J. H. Guo, Y. Z. Peng, C. Y. Peng, S. Y. Wang, Y. Chen, H. J. Huang, and Z. R. Sun, “Energy saving achieved by limited filamentous bulking sludge under low dissolved oxygen,” *Bioresour. Technol.*, vol. 101, no. 4, pp. 1120-1126, 2010.

[20] a M. P. Martins, J. J. Heijnen, and M. C. M. van Loosdrecht, “Effect of dissolved oxygen concentration on sludge settleability.,” *Appl. Microbiol. Biotechnol.*, vol. 62, no. 5-6, pp. 586-93, Oct. 2003.

[21] K. Kohno, T., Be, And K. Mori, “Characterization of type 1851 organism isolated from activated sludge samples.,” *WATER SCI TECHNOL*, vol. 46, no. 1-2, pp. 111-114, 2002.

[22] J. D. Parker DS, Kaufman WJ, “Physical conditioning of activated sludge flocs.,” *J Water Pollut Control Fed*, vol. 43, pp. 1817-33, 1971.

[23] A. T. Mielczarek, C. Kragelund, P. S. Eriksen, and P. H. Nielsen, “Population dynamics of filamentous bacteria in Danish wastewater treatment plants with nutrient removal.,” *Water Res.*, vol. 46, no. 12, pp. 3781-95, Aug. 2012.

[24] G. Thompson and C. Forster, “Bulking in activated sludge plants treating paper mill wastewaters.,” *Water Res.*, vol. 37, no. 11, pp. 2636-44, Jun. 2003.