CHARACTERIZATION OF ACTIVATED SLUDGE FLOCS

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ABSTRACT

Sludge characterization is important for an efficient operation of the activated sludge process. Problems with sludge separation occcur frequently related to sludge bulking, foaming and dispersed growth. Different methods are described to measure sludge sedimentation and filtration properties, by use of sedimentation indices, initial sedimentation velocity and capillary suction time (CST). Floc properties may be described by properties as floc strength, specific surface area, surface potential and surface energy. Phosphorus fractionation is important too in understanding the biological phosphorus removal process and possibilities to recover phosphorus from sludges. The different described methods have been used at the laboratory of the division of Water Resources Engineering, KTH.

KEYWORDS

Activated sludge, CST, filterability, floc characterization, phosphorus fractions, sludge indices, sludge properties.

INTRODUCTION

Problems with sedimentation and filamentous bacteria have shown that there is a need to measure other parameters than only sludge volume index (SVI). Different sedimentation parameters are diluted sludge volume index (DSVI), sludge quality index (SQI), stirred specific volume index (SSVI_{3..5}), initial sedimentation velocity (ISV) and modified sludge index (I_{VF}) which is independent of the volatile fraction.

Capillary suction time (CST) measurements describe the filterability of the settled sludge and are used to estimate the floc strength and EDTA-test for flocculation mechanisms. Other important measurements are sludge surface properties as specific surface area, surface potential as negative surface charge and surface energy as internal hydrophobicity. Floc structure may be measured as floc size and the amount of filamentous bacteria or other organisms.

Another important measurement is phosphorus fractionation which is a method to measure how phosphorus is bounded in different inorganic and organic fractions.

The different methods are examples of analytical procedures that have been used at the Division of Water Resources Engineering, KTH, in different research projects.

REVIEW OF METHODS

Sedimentation and filterability of sludge

Sedimentation

Settlebility of sludge may be measured as sludge volume (SV) in a 1000 ml measuring cylinder (height of 36 cm) after 30 minutes sedimentation and is expressed for a known initial sludge concentration as sludge volume index (SVI), sludge quality index (SQI), diluted sludge volume index (DSVI) and index, I_{VF}, which is independent of the volatile fraction. Stirred specific volume (SSV) is measured in a 3.5 l cylinder (10 cm diameter and calibrated with a 0-50 cm scale) with slowly stirring (1r.p.m.) and is expressed as stirred specific volume index (SSVI_{3.5}). These different evaluated sludge indices are described in table 1 (Hultman et al., 1991). When settling time is extended to 60 min for both methods it is possible to calculate the initial sedimentation velocity (ISV).

Sludge index	Apparatus	Calculation of index	Formula No.	Note
Sludge volume index (SVI)	1 litre sed. vessel	$SVI = \frac{SV}{SS}$	(1)	Normally performed with- out stirring in Sweden, although stirring is re- commended by Standard Methods (1981)
Diluted sludge volume index (DSVI)	1 litre sed. vessel	$DSVI = \frac{SV}{SS} \text{ for}$ $SV < 250 \text{ ml/l}$	(2)	Stobbe (1969). Recommen- dated method in FRG.
Sludge quality index (SQI)	1 litre sed vessel	$SQI = \frac{SV}{SS} \text{ for}$ $SV < 300 \text{ ml/l}$ $200 + SV / 3$	(3)	Fitch and Kos (1976). Performed without stirring. Sometimes used in Sweden.
		$SQI = \frac{200 + 5V + 5}{SS}$ for between 300 and 800 ml/l	(4)	
Stirred specific volume index (SSVI _{3.5})	3.5 litre sed. vessel with slowly stirring (1 r.p.m.)	$\mathbf{SSVI}_{3.5} = \frac{SSV}{SS}$	(5)	White (1975 and 1976). Especially used in England (developed at WRC)
Modified sludge index (I _{VF})		$I_{VF} = \frac{1 - VF}{VF} * SQI$	(6)	Hultman and Molina (1987) Modified index independent of the volatile fraction.
Notation:	SV = sludge volume (m SSV = stirred specific vo SS = initial suspended s VF = volatile fraction (s	l/l) after 30 minutes se lume (ml/l) after 30 mi olids concentration (g/ t/l)	dimentation i 1 inutes sedimen 1)	litre sedimentation vessel tation

Table 1. Different sludge indices (Hultman et al., 1991)

Filterability

The filterability of the settled sludge is measured as CST (capillary suction time) as well as floc strength. CST is the time interval needed of a sludge sample to saturate a certain area of a filter paper under the influence of the capillary suction of the paper. A low CST implies good sludge filterability.

A special CST-test apparatus is used.

Procedure:

- 1. A reservoir of either 10 or 18 mm of diameter depending on the sludge filterability is filled with sludge.
- 2. As soon as the cylinder is filled, the filtrate expands under the capillary suction of the filter paper. The velocity of expansion depends also on the filter paper quality but mainly on the filterability of the sludge. Probes are measuring conductivity changes during the expansion.
- 3. The apparatus determines the time (seconds) the filtrate moves from a diameter of 32 mm to 45 mm.

Floc strength by CST-test

CST-test with stirring, the slopes of CST against stirring time as a measure of the floc breakdown. A small (or negative which means better filterability after turbulence) slope means strong flocs. More turbulence for a longer time gives an increasing amount of smaller flocs.

Procedure: Stirring at 500 rpm after which samples are taken for CST measurement in 0, 60 and 120 s.

EDTA-test for flocculation mechanism

EDTA is a chelate and forms complexes with polyvalent metal ions. EDTA-disodium salt (Titriplex III) disperses flocs if the interaction mechanism is based on free metal ions. Titriplex III is effective in complexing calcium and magnesium. EDTA disperses flocs if the interaction mechanism is based on free d-block metal ions. Eriksson and Alm (1991) found that a combination of stirring and addition of EDTA creates larger and more compact sludge flocs.

EDTA-test procedure

- 1. Weigh 0, 75, 750, and 1500 mg of EDTA-disodium salt into 100 ml flasks. Add 120 ml of distillated water.
- 2. Pour 1380 ml of sludge to 2000 ml beaker. Add 120 ml of EDTA solution to 1380 ml of sludge to get end concentrations 0, 50, 500 and 1000 mg EDTA/l.
- 3. Mix for 20 minutes.
- 4. Sedimentation test in 1 litre graduated vessel for 60 minutes.
- 5. Pour carefully away the uppermost 200 ml and take a sample of supernatant on 800-500 ml level for SS.
- 6. CST-test on thickened sludge after 0, 60, and 120 seconds of mixing at 500 rpm.

Filtration velocity

Filtration velocity (the slope of CST against stirring time) reveales flocculation mechanism because it is a measure of the bulk properties of the primary flocs. Increase in the number of small particles causes a decrease in dewaterability.

Floc structure and sludge surface properties

Floc structure may be measured as floc size and the amount of filamentous bacteria or other organisms.

Floc size

Floc size is measured as floc length i.e. maximum dimension of the floc by microscopy described by Li and Ganczarczyk (1985).

Sampling with a pipet with a tip opening as wide as possible. Rapid dilution (3-10-100-500 times have been used depending on the actual MLSS) and agitation.

Micrographs are taken with a Polaroid camera and microadapter (0.55*), ocular magnification 10 and objective magnification 6 and 10. Five different pictures with both magnifications (always two magnifications of the same view). Magnifications are 0.55*10*6=33 and 0.55*10*10=55. Flocs are counted on the micrographs and set in size intervals e.g. <50, 50-250, 250-500, 500-1000 and >1000 μ m. 0.55*10*6=33:1 (33 mm in photo is 1 mm in reality). 0.55*10*10=55:1 (55 mm in photo is 1 mm in reality).

Filament amount

Filament amount classified as described by Li and Ganczarczyk (1985).

Sludge surface properties

Sludge surface properties may be measured as specific surface area, surface potential as negative surface charge and surface energy as internal hydrophobicity.

Specific surface area

The specific surface area of the sludge flocs is determined by a dye adsorption technique (Smith and Coackly, 1983; Andreadakis, 1993). The use of dyes is particularly attractive because it is simple and allows for a rapid colorimetric analysis of the solutions. Most dyes consist of a large organic structure with attached ionic groups. The organic part of the dye molecule is hydrophobic while the ionic components are hydrophilic. This is the reason why dyes tend to form aggregates of a few molecules called micelles in water solution. The adsorption of micelles instead of a monolayer of single dyestuff molecules can be avoided by choosing the right dye. A Naphthalene based dye, known as Lissamine Scarlet 4R or Victoria Scarlet 3R or Acid Red 18 or New Coccine, is suitable for dye adsorption measurements on activated sludge because the dye is stable.

New Coccine from Sigma CI No. 16255 Mol. Form. $C_{20}H_{11}N_2Na_3O_{10}S_3$

F.W. 604.5

Procedure:

- Dye has to be purified from impurities by repeated recrystallisation from 60/40 mixtures of ethanol and water. The purity of the recrystallised material is calculated on the basis of the sulphur composition.
 Because the main point of interest is to compare surface areas between different sludges, purification is not needed.
- 2. A stock solution of 0.29 mmol/l is prepared by weighing 175.3 mg of New Coccine in 1 l of distillated water.
- 3. From stock solution the working solutions are prepared daily.
- 4. A calibration curve is prepared for spectrophotometer measurements with different dye concentrations. The wavelength of maximum adsorption 505 nm is used. The calibration curve should show a linear relationship i.e. adherence to Beer's Law.
- 5. 50 ml of sludge sample of known MLSS¹ is poured/pipetted to a 100 ml flask. pH of the 50 ml sludge sample is lowered to pH 2.5 with 0.5 ml of 5 M HCl, to drop the charge of the sludge surface to zero. This is because the dye molecules are anionic and the sludge surface is negatively charged which causes electrostatic repulsion. Sludge samples did build one or a few big flocs when acid was added.
- 6. 10 ml of working solutions are added into the flasks. 0-sample by adding only 10 ml of distillated water.
- 7. During contact time of 30 minutes the sample is agitated vigorously on rotary table.
- 8. Centrifugation at 3000 rpm for 15 minutes.
- 9. Supernatant is collected and diluted when necessary, before spectrophotometric determination. The wavelength of maximum adsorption 505 nm is used. When dye remaining in solution is in excess of about 180 mmol/kg, the sludge should have reached saturation.
- 10. The specific surface is calculated as

$$S = Y * N * A$$

where

S = specific area (m^2/g) Y = adsorbed dye (mol/g)

N = Avogadro's number = $6.023 * 10^{23}$ molecules/mol

A = area covered by each molecule (m²/molecule) = 196 Å²

¹ There is only little variability of the maximum dye adsorption with sludge concentration (the standard deviation is 2.6 % of the mean). Sludge with 0.25 g SS/l was used.

Surface potential as negative surface charge

Negative surface charge

The negative surface charge can be estimated by the adsorption of cationic molecules such as ruthenium red (RR⁺). Adsorption of RR⁺ (MW = 576.5 g.mol⁻¹) is measured at equilibrium with a constant sludge concentration and increasing dye concentrations. The adsorption data can be approximated by the Freundlich model which relates the adsorbed concentration of the molecule (X) divided by the amount of dry matter (m), to the free concentration (Ce) in the form of X / m = K · Ce^{1/n}. The adsorption constant 1/n shows the non-linearity of the model and K is related to a maximum of adsorption when 1/n is close to zero (Figueroa and Silverstein, 1987; Urbain et al., 1993).

RR⁺-test procedure

- 1. Prepare a RR⁺ stock solution (5 g/l) by dissolving 0.25 g of RR⁺ in 50 ml of distillated water. Solution is slowly heated to 60 °C and cooled before storage in a dark bottle at 4 °C.
- 2. A calibration curve is prepared for spectrophotometer with different dye concentrations. The wavelength 530 nm ($\lambda max RR^+$) is used.
- 3. From stock solution the working solutions are prepared daily.
- 4. 250 ml sample of sludge with constant SS of 0.25 g/l is prepared by diluting with filtered secondary wastewater.
- 5. For 0-sample pore/pipette 18 ml of filtered secondary wastewater into a 100 ml flask and mixed with 2ml of RR⁺ solutions at different concentrations (0.25 1 g/l).
- 6. Pore/pipette 18 ml of sludge sample into a 100 ml flask and mixed with 2 ml of RR⁺ solutions at different concentrations (0.25 1 g/l).
- 7. Put the flasks on a rotary table at 150 rpm at room temperature for 3 h.
- 8. Centrifuge the sludge samples for 10 min at 2500 g.
- 9. Optical density is measured on the supernatants at 530 nm (λ max RR⁺) in a spectrophotometer.
- 10.The adsorbed (mmol/kg SS) and free (mmol/l) concentrations of RR⁺ are calculated from a calibration curve.
- 11. The linear regression analysis on the logarithm of the adsorbed concentrations vs free concentrations of RR⁺ gives the value of l/n (slope) and ln K (y axis intercept).

Surface energy as internal hydrophobicity

Internal hydrophobicity

The salt aggregation test measures relative surface hydrophobicity of flocs. Because hydrophobic surfaces have a natural tendency to avoid hydrated environments, the hydrophobicity of activated sludge flocs must be determined from disaggregated samples (Lindahl et al., 1981; Urbain et al., 1993).

A high value of internal hydrophobicity is equivalent to a large aggregation in response to a small increase in the salt concentration. It can be explained either by a low density of negative charges (hydrophilic groups) or by a high density of hydrophobic sites inside the flocs.

Salting out-test procedure

- 1. Prepare a 0.07 M Na₃PO₄ buffer (pH 7 ± 0.4).
- Weigh 39.6, 99, 198, 297 and 330 mg of (NH₄)₂SO₄ in 100 ml flasks. For positive control one more flask with 330 mg of (NH₄)₂SO₄ is needed. Pipet 10 ml of Na₃PO₄ buffer to the flasks. These give final concentrations of 0.3, 0.75, 1.5, 2.25 and 2.5 M. For negative control(s) do as many flasks as there are different sludges to be tested with only 10 ml of 0.07 M Na₃PO₄ buffer without ammonium sulfate.
- 3. 10 ml sample of sludge with constant SS e.g. 2.5 g/l in 20 ml tubes for sonication (sonicator, 50 W 20 kHz) in an ice bath during two 15 s periods interrupted by a 10 s resting time. An alternative is:

A sludge sample with constant SS (2.5 g/l) is disaggregated by mechanical means. Dispersing instrument at velocity \sim 8300 r p m and two 15 s periods interrupted by a 10 s resting time. Use plastic bottles.

4. Take away from the 100 ml flasks 1 ml of (Na₃PO₄ buffer + (NH₄)₂SO₄) and pipet instead 1 ml of disaggregated sludge sample to the flasks.

- 5. If pH has to be adjusted use NH₄OH.
- 6. Let stand for 3 h at room temperature.
- 7. Stir by hand and measure the amount of produced aggregates as optical density at 600 nm in the spectrophotometer. There are small settling flocs and therefore the optical density value keeps on changing try to catch the first value.
- 8. Because the difference in the optical density the positive control and the test is a linear function of the ammmonium sulfate concentrations.

The Internal Hydrofobicity of the flocs can be expressed by the slope from the linear regression analysis.

Phosphorus fractionation

The first phosphorus fractionation studies were developed for the study of phosphorus forms in soils, and were adapted to the analysis of lake sediments. A method for phosphorus fractionation in lake sediments was suggested by Psenner et al. (1988). A modified phosphorus fractionation method has been applied to activated sludge from waste treatment plants for biological phosphorus removal by Uhlmann et al. (1990), Schönbom (1995) and Aspegren et al. (1995) and for deposited sludge by Rydin (1996). A different fractionation method has been suggested for characterisation of phosphate and poly-phosphate by de Haas (1991).

The phosphorus fractions are determined using a sequential extraction procedure. Inorganic fractions is defined as reactive fractions and organic fractions as non-reactive fractions. A schematic representation of the procedure (Psenner et al., 1988) is given in Figure 1.

Explanations according to figure 1:

A. NH₄Cl-extractable phosphorus at neutral pH

This fraction is often termed labile, loosely bound or adsorbed phophorus. Dissolved reactive phosphorus (DRP) as NH₄Cl-RP, labile phosphorus, loosely bound reactive phosphorus. NH₄Cl dissolutes small amounts of iron- and aluminium-bound phosphorus and affects the solubility of calcium-bound phosphorus.

B. Buffered dithionite extractable phosphorus

DRP as BD-RP, reductant soluble phosphorus. Reductant soluble phosphorus is mainly extracted from iron hydroxide surfaces.

C. NaOH-extractable phosphorus

DRP as NaOH-RP soluble reactive phosphorus. Reactive NaOH-P is phosphate adsorbed to metal oxides (Al_2O_3) and other surfaces, exchangeable against OH⁻, and phosphorus compounds soluble in bases. The difference between total and reactive NaOH-P (= non-reactive phosphorus) is assumed to represent the major part of organic and humic P.

D. HCl-extractable phosphorus

DRP as HCl-RP soluble reactive phosphorus. It represents P bound to carbonates, apatite-P and P released by the dissolution of oxides (not adsorbed to the surface). It may contain traces of hydrolyzed organic phosphorus.

E. Residual phosphorus

It is the difference between TP (determined by digestion methods) and the sum of reactive phosphorus in the fractions A - D. Organic and non degradable phosphorus compounds are included in this fraction.

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1 cm ³ FS	EXTRACTANT	VOLUME	TEMP.	TIME		
	NH ₄ Cl 1 M	25 ml	25°C	2 h		
H ₂ O _{d.} 50ml mixing						
25°C 10min centrif.						
PELLET						
SUPERNATANT	NH ₄ Cl 1 M	25 ml	25°C	2 h		
mixing						
centrif.						
SUPERNATANT VPELLET	NaHCO ₃ 0.11 M	25 ml	25°C	1 h		
filtr.	$Na_2S_2O_4$ 0.11 M					
DRP centrif.						
SUDEDNATANT PELLET		25 ml	25°C	1 h		
	$Na_2S_2O_4 0.11 M$	25 111	25 C	1 11		
mixing						
centrif.						
SUPERNATANT VPELLET						
filtr.	NaOH 1 M	25 ml	25°C	18 h		
mixing		20 111	23 0	10 11		
oxidat centrif.						
V V PELLET						
		251	$25^{\circ}C$	1 L		
SUPERNATANT mixing	NaOH I M	25 mi	25 C	1 n		
centrif.						
SUPERNATANT VPELLET						
filtr		0.5 1	2 - 0 - 0	101		
mixing	HCI 0.5 M	25 ml	25°C	18 h		
neutral. centrif.						
V V PELLET						
DP DRP						
SUPERNATANT mixing	HCl 0.5 M	25 ml	25°C	1 h		
centrif.						
SUPERNATANT						
filtr						
neutral						
♦ PELLET						
DKP						

 $\begin{array}{ll} \mbox{Figure 1. Phosphorus fractionation scheme (Psenner et al., 1988).} \\ \mbox{DP} & \mbox{Dissolved Phosphorus} \\ \mbox{DRP} & \mbox{Dissolved Reactive Phosphorus} \\ \mbox{H}_2O_d & \mbox{deoxygenated distilled water} \end{array}$

SOME STUDIES FOR CHARACTERIZATION OF ACTIVATED SLUDGE FLOCS

Some studies has been done by Piirtola et al. (1998) to determine if some different weighting mineral materials as zeolite, apatite, calcium carbonate and a blend of talc and chlorite had an weighting effect. Tests were performed as batch tests and in a continuous denitrification-nitrification activated sludge pilot plant. The sludge-mineral material interaction was investigated by analysing sedimentation and filterability (CST), floc structure as floc size and amount of filament and sludge surface properties i.e. specific surface area, surface potential and surface energy.

Piirtola et al. (1998) tested two detergent zeolites, Doucil-P and Doucil-A24, in batch tests and in a continuous denitrification-nitrification activated sludge pilot plant. Batch tests as settling tests were done in a 1 l test vessel without stirring to determine if the zeolites were acting as weighting agent, or could cause floc breakdown and reduce the sludge sedimentation due to its ion-exchange capacity. Flocs breakdown can be caused by ion-exchange of metal ions as Ca^{2+} and Mg^{2+} . EDTA-test were done to reveal the importance of metal ions on flocculation. If the interaction mechanism is based on free metal ions EDTA disperses sludge flocs. Capillary suction time (CST) measure the filterability of the settled sludge and to estimate the floc strength. Filtration velocity (the slope of CST against stirring time) gives information about flocculation mechanisms, because it measures the bulk properties of the primary flocs.

Two different sludges were used in the batch tests to determine if there was a dependency of the weighting or floc breaking action, or both, on the sludge characteristics. A well settling sludge was obtained from a full scale plant (SVI 40-86 ml/g) and a bulking sludge was obtained from a pilot plant (SVI 326-414 ml/g). Zeolite concentrations was 10 - 30 mg/l. Suspended solids of the sludge was 2-3 g/l. It was not found any difference between the zeolite-free sludge and the zeolite containing sludge in sludge sedimentation or filterability.

EDTA addition had no influence on the settling for both sludge types. The filterability and floc strength were deteriorated by the addition of EDTA to the well settling sludge, while bulking sludge was not affected. Filterability deteriorated with increasing EDTA dosage in the well settling sludge. A significant difference was observed betweeen the sludge filterability, with and without zeolite addition. Filtration time was more than 40% longer for sludges containing zeolite both with and without EDTA. Flocs were broken down by the addition of EDTA and also zeolite addition had a negative impact on the floc structure by complexing metal ions or by its amorphousness, or both. The floc strength in both sludges was unaffected by the zeolite addition that could show that the reason for deterioration of the filterability is the amorphousness of the zeolite in the sludge matrix.

The pilot tests were performed in two parallel pilot plants, R and B, total volume 120 l and were used in the study of effect of detergent zeolite on the denitrification-nitrification activated sludge process. The flow was 200 l/d. The influent was synthetic wastewater with alkalinity of 2.7 mmol/l, COD 300 mg/l, P_{tot} 6.5 mg/l and N_{tot} 50 mg/l. This carbohydrate-rich favoured the growth of filamentous organisms. The readily biodegradable COD was removed in the pre-denitrification tank and it was also used as an anoxic selector for suppressing filamentous bulking.

Period I in plant R was started with an influent concentration of 10 mg/l Doucil-P. Period II started four weeks later, the dosage was changed to plant B and the dosage was gradually increased (10, 20 and 30 mg/l) during three weeks. Period III was run for three weeks in plant R and with 25 mg/l Doucil-A24. At the end of the test periods, the sludge was analysed for floc size, the amount of filaments, surface area, surface potential and surface energy.

The zeolite addition did not increase the alkalinity or sludge retention time. It could not be observed any differences in the amount of filaments or floc size distribution. After the last test period specific surface area was measured and giving $1.73E+23 \text{ m}^2/\text{g}$ without zeolite addition and $1.77E+23 \text{ m}^2/\text{g}$ with zeolite addition.

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After period II and III the surface potential as negative surface charge and surface energy as internal hydrophobicity was measured. In table 2 the values are shown where, l/n represents the non-linearity of the model and K is related to a maximum adsorption when l/n is zero. The results of internal hydrophobicity (IHB) after period II and III are shown in table 3.

Table 2. Negative surface charge after period II and III.

Negative surface charge (RR ⁺)	II B: 10,20 and 25 mg/l Doucil-P		III R 25 mg/l Doucil-A24	
	R	В	R	В
l/n	85.4	43.3	106.2	107.3
К	601.2	311.3	832.1	873.0

Table 3. Internal hydrophobicity of the flocs after period II and III.

Internal hydrophobicity	II B:10, 20 and 25 mg/l Doucil-P		III R: 25 mg/l Doucil-A24	
	R	В	R	В
IHB*100	8.7	7.9	6.5	5.8

The surface area showed a slight increase and for a negative surface charge a decrease caused by zeolite addition was observed.

CONCLUSIONS

Sludge characterization is necessary for an efficient operation and control of the activated sludge process. Many methods are available for measuring sedimentation and filtration properties, floc properties including floc strength, specific surface area, surface potential and surface energy and how phosphorus is bound to sludges.

Measurements of sludge indices and CST may be performed on a regurlarly basis while other methods as measurements of floc properties or phosphorus fractionation are useful for better understanding of process performance especially during operational disturbances.

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