6

SETTLING TESTS

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6.1 INTRODUCTION

Settling is an important process in several of the unit operations in wastewater treatment plants (WWTPs). The most commonly known of these unit processes are primary settling tanks (PSTs), which are a treatment units before the biological reactor, and secondary settling tanks (SSTs), which are a clarification step prior to discharge into a receiving water. Moreover, settling also plays an important role in new technologies that are being developed such as granular sludge reactors. Due to the different nature of the settleable components (raw wastewater, activated sludge, and granular sludge) and the concentration at which these compounds occur, these unit processes are characterised by distinctly different settling behaviours. This section presents an overview of the different settling regimes that a particle-liquid suspension can undergo and relates these regimes to the specific settling behaviour observed in SSTs, PSTs and granular sludge reactors.

The settling behaviour of a suspension (e.g. secondary sludge, raw wastewater or granular sludge) is governed by its concentration and flocculation tendency and can be classified into four regimes (Figure 6.1): discrete non-flocculent settling (Class I), discrete flocculent settling (Class II), zone settling or hindered settling (Class III) and compressive settling (Class IV).

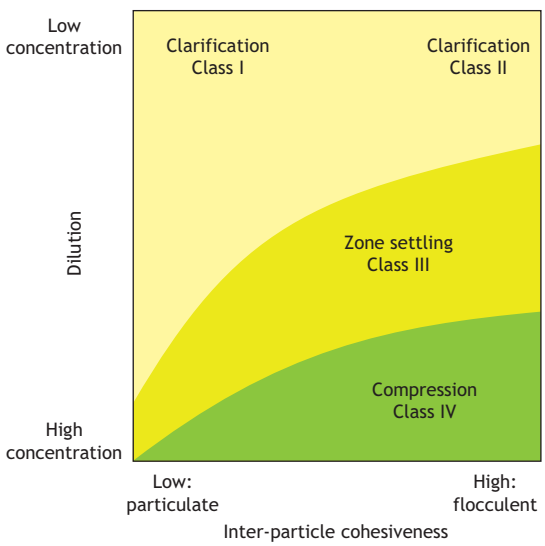

Figure 6.1 Settling regimes (Ekama *et al.,* 1997).

At low concentrations (classes I and II), the particles are completely dispersed, there is no physical contact between them and the concentration is typically too diluted for particles to influence each other's settling behaviour. Each particle settles at its own characteristic terminal velocity, which depends on individual particle properties such as shape, size, porosity and density. If

these dilute particles show no tendency to flocculate (for example, granular sludge), this regime is called discrete non-flocculent settling (Class I). However, certain suspensions (among which raw wastewater solids and activated sludge flocs) have a natural tendency to flocculate even at low concentrations (Ekama *et al.,* 1997). Through subsequent processes of collision and cohesion, larger flocs are formed causing their settling velocity to change over time. This regime is called discrete flocculent settling (Class II). It is important to note that during discrete flocculent settling, the formed flocs will still settle at their own characteristic terminal velocity. Hence, both discrete non-flocculent and discrete flocculent settling undergo basically the same settling dynamics. The difference lies in the fact that, for discrete flocculent settling, an additional flocculation process is occurring simultaneously with the settling process, which alters the particles' individual properties and consequently their terminal settling velocity.

The transition from discrete settling to hindered settling (Class III) occurs if the solid concentration in the tank exceeds a threshold concentration where the particles no longer settle independently of one another. As can be seen from Figure 6.1, this threshold concentration depends on the flocculation state of the sludge. For secondary sludge the transition typically occurs at concentrations of 600 - 700 mg TSS L^{-1} whereas for granular sludge the threshold concentration can go up to 1,600-5,500 mg TSS L^{-1} (depending on the granulation state) (Mancell-Egala *et al.,* 2016). Above this threshold, each particle is hindered by the other particles and the inter-particle forces are sufficiently strong to drag each particle along at the same velocity, irrespective of size and density. In other words, the particles settle collectively as a zone, and therefore this regime is also called zone settling. In this regime, a distinct interface between the clear supernatant and the subsiding particles is formed. When the solids concentration further increases above a critical concentration (5-10 g L^{-1}), the settling behaviour changes to compressive settling (Class IV). The exact transition concentration depends once more on the flocculation state of the particles (De Clercq *et al.,* 2008). At these elevated concentrations, the solids come into physical contact with one another and are subjected to compaction due to the weight of overlying particles. The settling velocity will be much lower than in the hindered settling regime.

Activated sludge with a proper biological make-up shows a natural tendency to flocculate and, depending on its concentration, can cover the entire right side of Figure 6.1. Hence, in an SST, different settling regimes occur simultaneously at different locations throughout the tank. Low concentrations in the upper regions of the SST favour discrete (flocculent) settling whereas the concentrations of the incoming sludge are typically in the range for hindered settling, and sludge thickening inside the sludge blanket is governed by compressive settling. In contrast to this, a specific characteristic of the granular sludge technology is the granules' low tendency to coagulate under reduced hydrodynamic shear (de Kreuk and van Loosdrecht, 2004), thus positioning them on the left side of Figure 6.1. This feature causes granular sludge to undergo discrete (non-flocculent) settling at concentrations where conventional activated sludge undergoes hindered or compression settling. Finally, PSTs are fed by incoming wastewater containing a relatively low concentration of suspended solids (i.e. the upper part Figure 6.1). Hence, the dominant settling regime in these tanks is discrete settling (classes I and II).

As each unit process is characterised by its own settling behaviour, different experimental methods are required to assess their performance. This chapter provides an overview of experimental methods to analyse the settling and flocculation behaviour in secondary settling tanks (Section 6.2 and 6.3), granular sludge reactors (Section 6.4) and primary settling tanks (Section 6.5).

6.2 MEASURING SLUDGE SETTLEABILITY IN SSTs

To evaluate the performance of an SST, it is essential to quantify the settling behaviour of the activated sludge in the system. Batch settling experiments are an interesting information source in this respect since they eliminate the hydraulic influences of in- and outgoing flows on the settling behaviour. Therefore, several methods aim to determine the settling characteristics of the activated sludge by measuring certain properties during the settling of activated sludge in a batch reservoir.

Several types of measurements can be performed by means of a batch settling test. These measurements range from very simple experiments providing a rough indication of the sample's general settleability (Section 6.2.1) to more labour-intensive experiments that measure the specific settling velocity (Section 6.2.2) or even determine a relation between the settling velocity and the sludge concentration (Section 6.2.3). Moreover, some useful recommendations to consider when performing

batch settling experiments are provided in Section 6.2.4 and an overview of recent developments with respect to these type of experiments can be found in Section 6.2.5.

6.2.1 Sludge settleability parameters

6.2.1.1 Goal and application

A number of parameters have been developed to obtain a quantitative measure of the settleability of an activated sludge sample. These Sludge Settleability Parameters (SSPs) are based on the volume that sludge occupies after a fixed period of settling. Among these, the Sludge Volume Index (SVI) (Mohlman, 1934) is the most known. A number of issues have been reported with the SVI as a measure of sludge settleability (Dick and Vesilind, 1969; Ekama *et al.,* 1997) of which the most important one is its dependency on the sludge concentration. Particularly at higher concentrations, measured SVI values can deviate significantly between sludge concentrations (Dick and Vesilind, 1969). Moreover, SVI measurements have been found to be influenced by the dimensions of the settling cylinder. These problems can be significantly reduced by conducting the test under certain prescribed conditions. Hence, a number of modifications have been proposed to the standard SVI test in order to yield more consistent information (Stobbe, 1964; White, 1976, 1975). Stobbe (1964) proposed conducting the SVI test with diluted sludge and called it the Diluted SVI (DSVI). White (1975, 1976) proposed the Stirred Specific Volume Index ($SSVI_{3.5}$) where the sludge sample is stirred during settlement. Although each of these SSPs are described in more detail below, it is important to note that the $SSVI_{3.5}$ is known to provide the most consistent results.

6.2.1.2 Equipment

a. A graduated (minimum resolution 50 mL) cylindrical reservoir with a volume of 1 litre (for SVI and DSVI) or with dimensions specified by White (1975) for SSVI.
b. A digital timer displaying accuracy in seconds.
c. A sludge sample from either the recycle flow of the SST or the feed flow into the SST. The latter can be collected from the bioreactor or the splitter structure.
d. Effluent from the same WWTP (in case dilution is needed).
e. Equipment for the Total Suspended Solids (TSS) test (according to method 2540 D in APHA *et al.,* 2012).
f. A stirrer for the SSVI test.

6.2.1.3 The Sludge Volume Index (SVI)

The Sludge Volume Index (SVI) (Mohlman, 1934) is defined as the volume (in mL) occupied by 1 g of sludge after 30 min settling in a 1 L unstirred cylinder.

- **Protocol**

1. Measure the concentration of the sludge sample with a TSS test according to method 2540 D of Standard Methods (APHA *et al.,* 2012).
2. Fill a 1 L graduated cylinder with the sludge sample and allow the sample to settle.
3. After 30 min of settling, read the volume occupied by the sludge from the graduated cylinder (SV_{30} in mL L^{-1}).
4. Calculate the SVI from Eq. 6.1, with X_{TSS} being the measured concentration of the sample in g L^{-1}:

$$SVI = \frac{SV_{30}}{X_{TSS}} \qquad \text{Eq. 6.1}$$

- **Example**

In this example an SVI test is performed with a sludge sample from the bioreactor in the WWTP at Destelbergen. The concentration of the sample is measured at 2.93 g L^{-1}.

A graduated cylinder is filled with the sludge sample and the sludge is allowed to settle. After 30 min of settling, the sludge occupies a volume of 290 mL (SV_{30}).

Hence the sample has an SVI of:

$$SVI = \frac{290 \text{ mL } L^{-1}}{2.93 \text{ g } L^{-1}} = 99 \text{ mL } g^{-1}$$

This result indicates a sludge with good settling properties. Typical SVI values for AS can be found between 50-400 mL g^{-1} where 50 mL g^{-1} indicates a sample with very good settleability and 400 mL g^{-1} a sample with poor settling properties.

6.2.1.4 The Diluted Sludge Volume Index (DSVI)

The Diluted Sludge Volume Index: DSVI (Stobbe, 1964) differs from the standard SVI by performing an additional dilution step prior to settling. The sludge is hereby diluted with effluent until the settled volume after 30 min is between 150 ml L^{-1} and 250 ml L^{-1}. Note that all the dilutions must be made with effluent (before chemical disinfection) from the plant where the sludge is

obtained to reduce the possibility of foreign substances affecting the settling behaviour.

- **Protocol**

1. Dilute the sludge sample with effluent until the settled volume after 30 min is between 150 mL L^{-1} and 250 mL L^{-1}.
2. Perform steps 1-3 from the standard SVI test.
3. Calculate the DSVI from Eq. 6.1, with X_{TSS} being the concentration of the diluted sample in g L^{-1}.

The advantage of the DSVI lies in its insensitivity to the sludge concentration, allowing for consistent comparison of sludge settleability between different activated sludge plants.

6.2.1.5 The Stirred Specific Volume Index ($SSVI_{3.5}$)

The Stirred Specific Volume Index ($SSVI_{3.5}$) was presented by White (1975, 1976) who found that stirring the sample during settling reduces wall effects, short circuiting and bridge formation effects, thereby creating conditions more closely related to those prevailing in the sludge blanket in SSTs.

The $SSVI_{3.5}$ is determined by performing an SVI test at a specific concentration of 3.5 g L^{-1} while the sludge is gently stirred at a speed of about 1 rpm. To determine the $SSVI_{3.5}$, the sludge concentration is measured with a TSS test and subsequently diluted with effluent to a concentration of 3.5 g L^{-1}. In some cases further concentration of the sample may be necessary if the plant is operating at MLSS values below 3.5 g L^{-1} and sampling from the return activated sludge flow is not possible.

Compared to the DSVI, the $SSVI_{3.5}$ has the additional advantage that it not only overcomes the concentration dependency but is shown to be relatively insensitive to the dimensions of the settling column, provided that it is not smaller than the dimensions specified by White (1976), i.e. a depth to diameter ratio of between 5:1 and 6:1, and a volume of more than 4 L. As the measurement is performed in a larger reservoir, the $SSVI_{3.5}$ cannot be directly calculated from Eq. 6.1 but the volume of the particular column has to be reduced to an equivalent 1 L column. This is done by expressing the settled sludge volume at 30 min as a fraction of the column volume (f_{sv}) and multiplying this fraction by 1,000 mL to obtain the equivalent l L stirred settled volume SSV_{30} (Eq. 6.2).

$$SSV_{30} = f_{sv} \cdot 1{,}000 \qquad \text{Eq. 6.2}$$

This value can then be used in Eq. 6.1 to calculate the $SSVI_{3.5}$ with X_{TSS} = 3.5 g L^{-1}.

Although the $SSVI_{3.5}$ is not as easily executed as the DSVI due to the specified stirring equipment required, it does provide the most consistent results (Ekama *et al.,* 1997; Lee *et al.,* 1983).

- **Protocol**

1. Measure the concentration of the sludge sample with a TSS test according to method 2540 D of Standard Methods (APHA *et al.,* 2012).
2. Dilute the sample with effluent to a concentration of 3.5 g L^{-1}.
3. Fill a graduated cylinder with the minimum dimensions specified by White (1976).
4. After 30 min of settling during which the sample is stirred at a speed of 1 rpm, read off the volume occupied by the sludge from the graduated cylinder and calculate the SSV_{30} from Eq. 6.2.
5. Calculate the $SSVI_{3.5}$ from Eq. 6.1 with $SV_{30} = SSV_{30}$ and X_{TSS} = 3.5 g L^{-1}.

6.2.2 The batch settling curve and hindered settling velocity

6.2.2.1 Goal and application

The sludge settleability parameters presented above provide a low level measurement of the general settleability. However, it should be stressed that they represent only a momentary recording of the settling behaviour. In reality, the volume of a sludge sample after 30 min of settling will depend on both its hindered settling and compression behaviour, which are both influenced by a number of factors such as the composition of the activated sludge (for example, the population of filamentous organisms), floc size distributions, surface properties, rheology, etc. Consequently, two sludge samples with different settling behaviour can result in similar values for the sludge settleability parameters.

More detailed information on the settling behaviour of a sludge sample can be obtained from a batch settling curve which makes it possible to investigate the settling behaviour of sludge at different settling times.

Batch settling curves can serve different purposes. They can be used either qualitatively to determine operational or seasonal trends in the settling behaviour or

they can be used quantitatively to determine the SST's capacity limit. In the former, a simple graduated cylinder can be used (for example, the cylindrical reservoir in Figure 6.2). In the latter, the selection of an appropriate settling reservoir to avoid wall effects during the test is imperative. More information on the optimal shape and size of batch settling reservoirs is provided in Section 6.2.4.1.

6.2.2.2 Equipment

For this test the following equipment is needed:

a. A graduated (minimum resolution 50 mL) cylindrical reservoir.
b. A digital timer displaying accuracy in seconds.
c. A sludge sample from either the recycle flow of the SST or the feed flow into the SST. The latter can be collected from the bioreactor or the splitter structure.
d. Equipment for a TSS test (APHA *et al.,* 2012).
e. Stirring equipment if the results are to be used for quantitative analysis of the SST's capacity.

6.2.2.3 Experimental procedure

To measure a batch settling curve, a reservoir is filled with a sludge sample and a timer is started to keep track of the duration of the experiment. The sludge is allowed to settle and the position of the suspension-liquid interface is measured at different time intervals. This methodology is illustrated in Figure 6.2 where the position of the suspension-liquid interface is indicated by the red arrow. Recording the height of the suspension-liquid interface at several time intervals results in a curve with the evolution of the sludge blanket height over time. Standard measurement times for a batch settling curve are 0, 0.5, 1, 2, 3, 4, 5, 10, 15, 20, 30 and 45 min but these can be adapted depending on the settling dynamics of a specific sludge sample (for more information see Section 6.2.4). At the start of the test, the suspension-liquid interface is typically measured more frequently, as the sludge is settling at a relatively fast pace. Later in the test, the frequency of the measurements is decreased, because the interface is moving more slowly.

- **Protocol**

1. Homogenize the sludge sample. Do not shake the sample vigorously as this will disturb the sludge and alter its settling properties.
2. Fill the cylindrical reservoir with the sample. Pour gently and in a steady flow so as not to disturb the sludge too much nor to allow it to settle again in the container.
3. Start the timer immediately after filling the column.
4. Measure the sludge water interface at the following time intervals: 0, 0.5, 1, 2, 3, 4, 5, 10, 15, 20, 30 and 45 min.

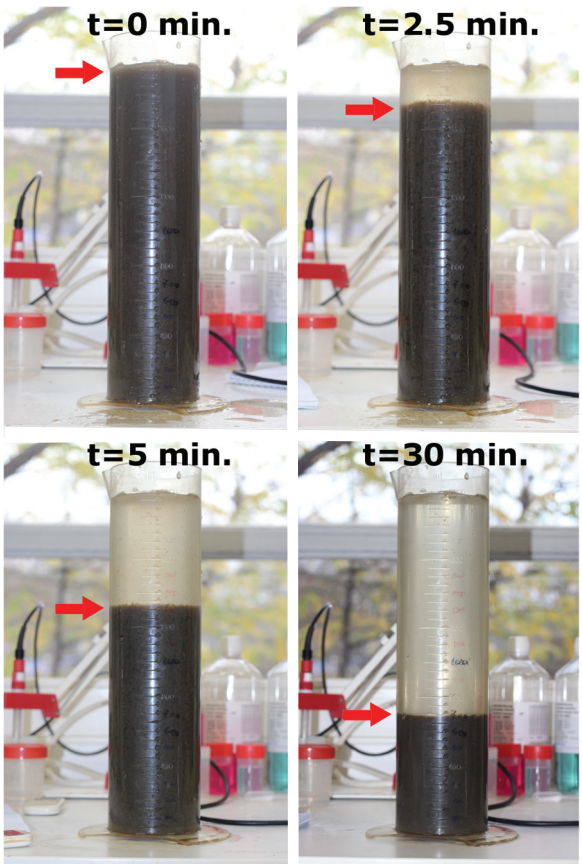

Figure 6.2 Photograph of the batch settling column at different settling times, indicating the suspension-liquid interface (photo: E. Torfs).

- **Example**

A batch settling curve is measured with a sludge sample from the bioreactor in the WWTP at Destelbergen. The concentration of the sample is measured as 2.93 g L^{-1}. The measured heights of the suspension-liquid interface (i.e. the Sludge Blanket Height: SBH) at different settling times are provided in Table 6.2 and the batch curve is shown in Figure 6.3.

Table 6.2 Measured sludge blanket height during a single batch settling test.

Time (min)	SBH (m)
0	0.250
0.5	0.247
1	0.244
2	0.238
3	0.206
4	0.184
5	0.169
10	0.122
15	0.106
20	0.098
30	0.088
45	0.081

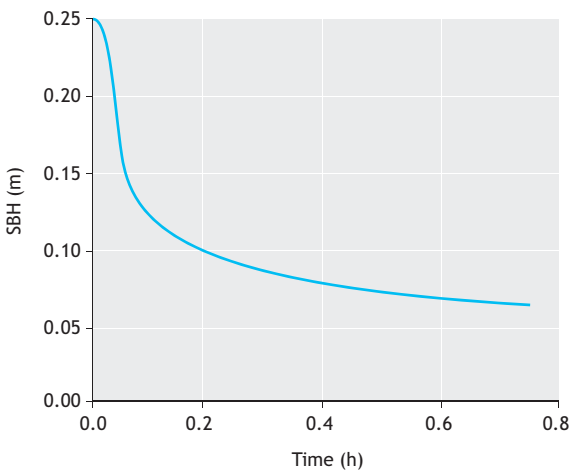

Figure 6.3 Measured batch settling curve.

6.2.2.4 Interpreting a batch settling curve

Typically, four different phases can be observed in a batch settling curve. Each phase marks a change in the settling behaviour at the suspension-liquid interface. Figure 6.4 shows the evolution of the sludge blanket height over time during a batch settling test, indicating the four phases. It is important to note that a batch settling curve only provides information on the settling behaviour at the sludge-water interface. At any specific time, the settling behaviour at different depths throughout the column may differ from the settling behaviour at the interface depending on the local concentrations.

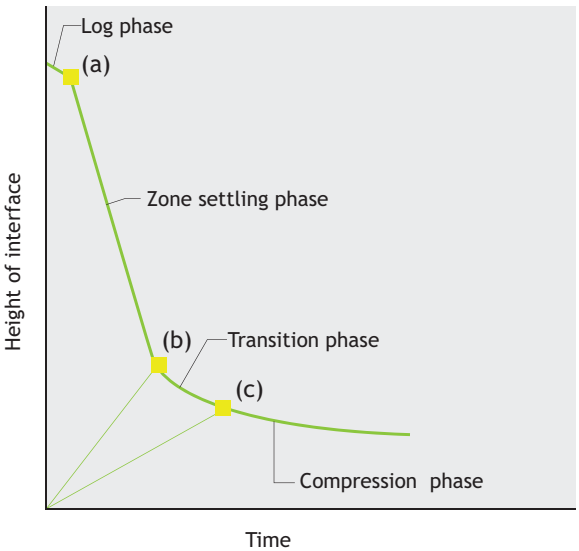

Figure 6.4 Evolution of the sludge blanket height over time, indicating the four phases (Rushton *et al.*, 2000).

Figure 6.5 represents the distribution of settling regions over the depth of the column at different times during a batch settling experiment.

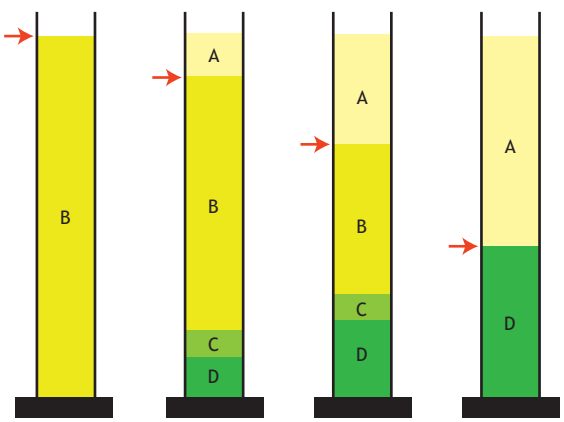

Figure 6.5 Chronological process of a batch settling test (Ekama *et al.*, 1997).

Almost immediately after the start-up of the experiment, four regions are formed at increasing depth. The top region (region A) consists of supernatant. Below region A, regions B, C and D are formed where respectively zone settling, transition settling and compression settling take place (Ekama *et al.,* 1997). The position of the sludge/water interface is indicated with a red arrow. Hence, the phases that are recorded in a batch settling curve occur as the suspension-liquid interface passes through these different settling regions.

From the beginning of the test up to point (a) (Figure 6.4), the suspension-liquid interface is in the lag phase. In this phase, the activated sludge needs to recover from disturbances due to turbulence caused by the filling of the batch column.

During the hindered settling phase or zone settling phase which starts at point (a) and ends at point (b) (Figure 6.4), the interface is located in the hindered settling region (region B). This phase is characterised by a distinct linear decline in the batch curve. An equilibrium between the gravitational forces causing the particles to settle and the hydraulic friction forces resisting this motion results in the same settling velocity for all the particles in the region. If the column is not stirred, then the velocity at which the interface moves downward is called the hindered settling velocity v_{hs} at the inlet solids concentration. If the dimensions and conditions for the batch test were set so as to avoid wall effects (Section 6.2.4.1), the measured settling velocity corresponds with the zone settling velocity in an actual SST.

The transition phase starts (point (b)) when the sludge blanket reaches the transition layer (region C). The transition layer is a layer of constant thickness and is formed by particles coming from the decreasing hindered settling layer and particles coming from the increasing compression layer. Although during this phase the same characteristics exist as in the zone settling regime, the settling velocity decreases because the concentration gradient increases with depth. The transition phase ends when the sludge blanket reaches the compression layer.

The last phase starts at point (c) and is called the compression phase. The time at which the compression phase starts, called the compression point, is difficult to identify. During the compression phase the particles undergo compaction, thus creating an increasing concentration gradient as well as a decreasing settling velocity.

6.2.2.5 Measuring the hindered settling velocity

At moderate sludge concentrations (between approx. 1 g L^{-1} and 6 g L^{-1}), sludge will initially settle according to the zone or hindered settling regime. The slope of the linear part of a batch settling curve corresponds to the hindered settling velocity v_{hs}.

- **Example**

The hindered settling velocity for the data in Figure 6.3 is computed by determining the steepest slope between three consecutive data points (which can be performed in any software). The procedure is illustrated in Figure 6.6 and results in a settling velocity of 1.374 m h^{-1}.

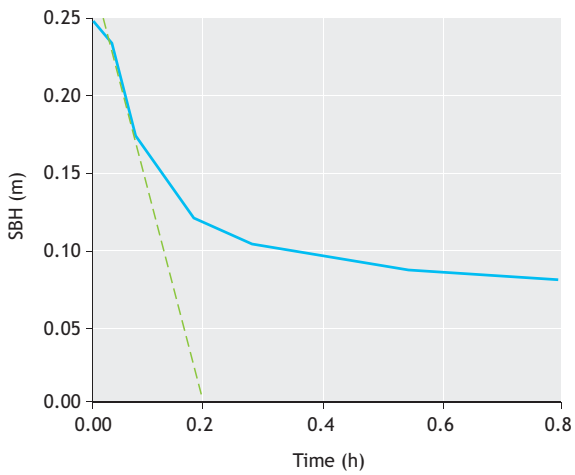

Figure 6.6 Calculation of the steepest slopes from a batch settling curve at a concentration of 2.93 g L^{-1}.

6.2.3 v_{hs}-X relation

6.2.3.1 Goal and application

The concentration in the hindered settling region is uniform and equal to the initial solids concentration of the batch. By calculating the slopes of the linear part of the batch curves for different initial concentrations, the hindered settling velocity can be determined as a function of the solids concentration. The relation between hindered settling velocity and concentration is of particular importance for the design of SSTs as it governs the determination of the limiting flux and thus the SST's surface area. As stated previously, in order to use the v_{hs}-X relation for quantitative calculations such as the determination of the SST's surface area, the dimensions

and conditions of the batch settling test need to be set according to the specifications given in Section 6.2.4.1.

6.2.3.2 Equipment

For this test the following equipment is needed:

a. A graduated (minimum resolution 50 mL) cylindrical reservoir.
b. A digital timer displaying accuracy in seconds.
c. A sludge sample from the recycle flow of the SST.
d. Effluent from the same WWTP (for dilution).
e. Equipment for the TSS test (APHA *et al.,* 2012).
f. Stirring equipment if the results are to be used for quantitative analysis of the SST's capacity.

6.2.3.3 Experimental procedure

To obtain different initial concentrations for the batch experiments, a sludge sample from the recycle flow of the SST is diluted with effluent from the same WWTP. Hence, a dilution series is made with respectively 100, 80, 60, 50, 40 and 20 % of sludge. For example, a 40 % dilution consists of 0.8 L of sludge from the recycle flow and 1.2 L of effluent. For each dilution a batch curve is measured according to the step-wise protocol from Section 6.2.2.3 and the hindered settling velocity is calculated from the slope of the linear part of the batch curve.

In order to obtain reliable results for the v_{hs}-X relation, it is important to have an accurate measure of the initial concentration in each experiment. The concentrations in the dilution series are often determined by measuring the concentration in the recycle flow and then calculating the concentration of the dilution assuming the effluent concentration is negligible. However, this procedure is prone to errors if the recycle flow sample is not fully mixed at any time during the filling of the batch. A more reliable approach is to measure the TSS of each dilution experiment separately. This can be done by mixing up the content of the batch reservoir at the end of each experiment and subsequently taking a sample for the TSS measurement. This approach requires some additional work as more TSS tests need to be performed but it ensures a reliable measurement of the diluted concentration.

- **Protocol**

1. Perform Step 1 from the protocol to measure the batch curves with a sludge sample from the recycle flow.
2. Combine a certain volume of the sludge sample with the effluent until the required dilution is obtained.
3. Perform steps 2 to 4 from the protocol to measure the batch curves.
4. After 45 min of settling, homogenise the sample in the cylindrical reservoir again and take a sample to determine the sludge concentration with a TSS test.

- **Example**

Samples were collected from the recycle flow and the effluent at the WWTP in Destelbergen (Belgium). The sludge/water interface during settling was measured for different initial concentrations (Table 6.3). The resulting settling curves are shown in Figure 6.7.

Table 6.3 Measured sludge blanket height (in m) during batch settling tests at different initial concentrations.

Time	1.37 g L^{-1}	2.37 g L^{-1}	3.42 g L^{-1}	4.10 g L^{-1}	5.46 g L^{-1}	6.83 g L^{-1}
0	0.248	0.248	0.248	0.248	0.248	0.248
0.5	0.243	0.244	0.246	0.248	0.247	0.248
1	0.215	0.236	0.241	0.247	0.246	0.248
2	0.107	0.198	0.214	0.244	0.245	0.248
3	0.074	0.163	0.186	0.242	0.243	0.247
4	0.064	0.144	0.165	0.239	0.243	0.246
5	0.059	0.130	0.149	0.234	0.241	0.245
10	0.046	0.102	0.115	0.195	0.234	0.241
15	0.041	0.091	0.102	0.172	0.227	0.239
20	0.038	0.083	0.092	0.156	0.219	0.236
30	0.033	0.073	0.083	0.132	0.205	0.231
45	0.031	0.064	0.074	0.114	0.182	0.223

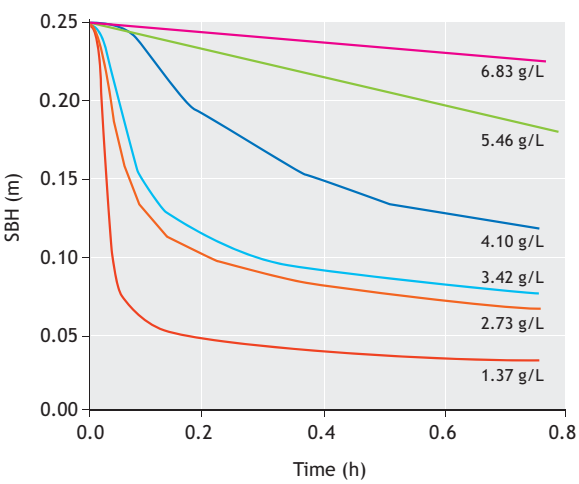

Figure 6.7 Batch settling curves at different initial concentrations.

The hindered settling velocities for the data in Figure 6.7 are computed by determining the steepest slope between three consecutive data points (Figure 6.8A). The resulting velocities are presented in Figure 6.8 B and Table 6.4.

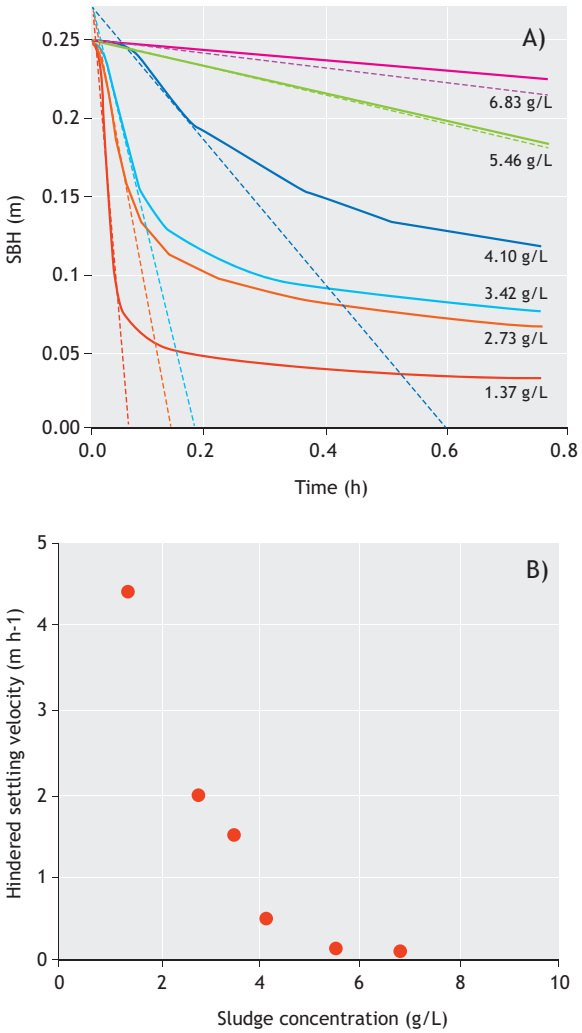

Figure 6.8 (A) Batch settling experiments at different initial solids concentration indicating the maximum slope for each curve. (B) The maximal slope represents a measurement of the hindered settling velocity.

The hindered settling velocity slows down at higher concentrations because the settling particles will be increasingly hindered by surrounding particles. Note that for the concentrations 5.46 g L^{-1} and 6.83 g L^{-1}, it becomes increasingly difficult to determine the steepest slope and the validity of these curves to measure hindered settling may be questioned. More information can be found in Section 6.2.4.3.

Table 6.4 Measured hindered settling velocities at different initial concentrations.

Concentration (g L^{-1})	v_{hs} (m h^{-1})
1.37	4.39
2.73	2.01
3.42	1.53
4.10	0.46
5.46	0.09
6.83	0.05

6.2.3.4 Determination of the zone settling parameters

Mathematically, the relation between the sludge concentration and the zone settling velocity can be described by an exponential decaying function (Eq. 6.3) (Vesilind, 1968). In this equation v_{hs} represents the hindered settling velocity of the sludge, V_0 the maximum settling velocity, X_{TSS} the solids concentration and r_V a model parameter. The parameters V_0 and r_V in this function provide information on the sludge settleability and are frequently used in SST design procedures. More information on design procedures can be found in Ekama *et al.* (1997).

$$v_{hs}(X) = V_0 \cdot e^{-r_V \cdot X_{TSS}} \qquad \text{Eq. 6.3}$$

The parameters V_0 and r_V can be estimated from the experimental data by minimising the Sum of Squared Errors (SSE) in Eq. 6.4 In this equation N is the number of data points, $v_{hs,i}$ the measured hindered settling velocity at concentration i, and $\tilde{v}_{hs,i}$ is the corresponding prediction by the function of Vesilind (1968) for a particular parameter set [V_0 r_V].

$$SSE = \sum_{i=1}^{N} \left(v_{hs,i} - \tilde{v}_{hs,i}(V_0, r_V)\right)^2 \qquad \text{Eq. 6.4}$$

An estimation of the zone settling parameters and calculation of the confidence intervals for these estimates can be performed as explained in Chapter 5. Minimising the SSE from Eq. 6.4 will give more weight to the fit of high settling velocities (i.e. at low concentrations). In order to give equal weight to all the measured settling velocities, a logarithmic fit can be performed.

- **Example**

Table 6.5 provides the initial parameter estimates, the optimal parameters after optimisation and the 95 % confidence intervals for the estimated parameters for the data in Table 6.4. The simulation results of calibrated functions vs. the experimental data points are shown in Figure 6.9.

Table 6.5 Initial values, optimal values and confidence intervals of the estimated parameters for the settling function.

Parameter	Initial value	Optimal value	Confidence interval
V_0 (m h^{-1})	9.647	10.608	$\pm$ 1.265
r_V (L g^{-1})	0.488	0.634	$\pm$ 0.038

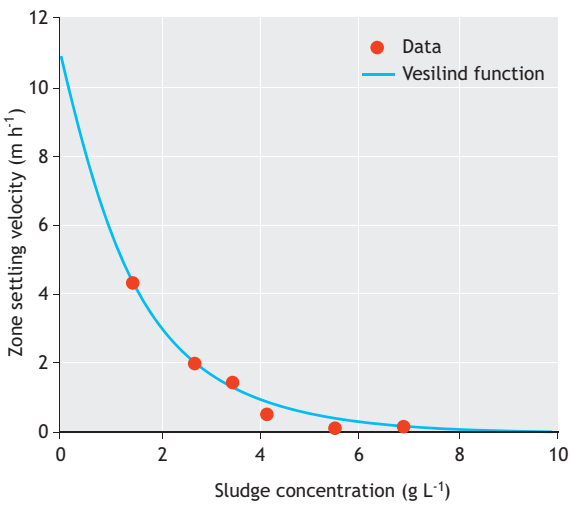

Figure 6.9 Settling velocity as a function of the solids concentration. The circles represent measured settling velocities and the line the calculated settling velocities after calibration of the function by Vesilind (1968).

6.2.3.5 Calibration by empirical relations based on SSPs

As the measurement of batch settling curves is much more time-consuming than the measurements of simple SSPs, several empirical equations have been developed that relate the settling parameters V_0 and r_V to simple measurements of SSPs (Härtel and Pöpel, 1992; Koopman and Cadee, 1983; Pitman, 1984, Daigger and Roper, 1985). Examples of such empirical equations and the resulting parameter estimates are given in Table 6.6.

Figure 6.10 shows that the settling parameters calculated from the empirical equations are not able to accurately describe the measured data from Table 6.4. This could be expected as sludges with a similar SVI may show a different settling behaviour dependent on the sludge properties. Moreover, when using the empirical relations, two parameters are estimated based on only one data point. The SSPs thus provide insufficient information to describe the settling behaviour at different sludge concentrations. For these reasons, the use of empirical relations based on SSPs is not an accurate method to estimate the hindered settling parameters of the settling functions and should be avoided.

Table 6.6 Estimated values for V_0 (m h^{-1}) and r_V (L g^{-1}) by empirical relations based on SSPs.

Reference	Equations	Parameter values	Equation nr.
Härtel and Pöpel (1992)	$V_0 = 17.4\ e^{-0.0113\ SVI}$	$V_0 = 9.647$	Eq. 6.5
	$r_V = -0.9834\ e^{-0.00581\ SVI} + 1.043$	$r_V = 0.488$	Eq. 6.6
Koopman and Cadee (1983)	$\ln(V_0) = 2.605 - 0.00365\ DSVI$	$V_0 = 9.993$	Eq. 6.7
	$r_V = 0.249 + 0.002191\ DSVI$	$r_V = 0.431$	Eq. 6.8
Pitman (1984)	$\frac{V_0}{r_V} = 67.9\ e^{-0.016\ SSVI_{3.5}}$	$V_0 = 5.669$	Eq. 6.9
	$r_V = 0.88 - 0.393 \log\left(\frac{V_0}{r_V}\right)$	$r_V = 0.446$	Eq. 6.10

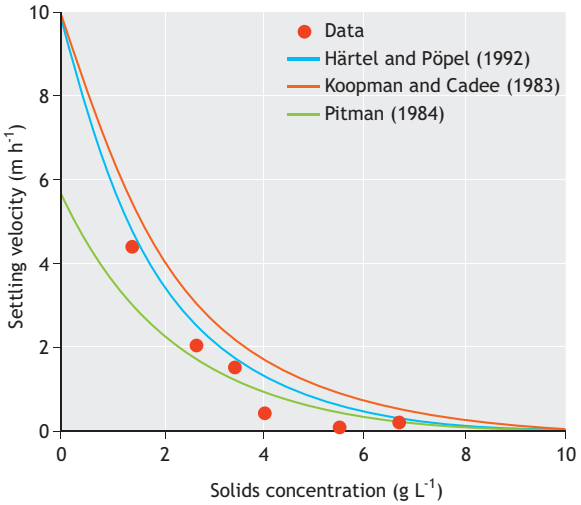

Figure 6.10 Settling velocity as a function of the solids concentration. The circles represent the measured settling velocities from the batch settling tests and the lines represent the settling velocities calculated by the function of Vesilind (1986) with parameter values based on empirical equations.

6.2.4 Recommendations for performing batch settling tests

6.2.4.1 Shape and size of the batch reservoir

It is recommended to use a cylindrical reservoir for batch settling tests. In conical reservoirs, such as Imhoff cones, no zone settling region can be measured because the reducing width of the cross section will inevitably cause a concentration gradient. When the goal of the measurements is to use the zone settling parameters for a qualitative analysis of the limiting flux and the SST's surface area, care should be taken to avoid any influence of the settling reservoir on the settling behaviour (so-called wall effects). This can be achieved by performing the batch test in a column of at least 100 mm in diameter and 1 m deep, and by gently stirring (1 rpm) the sample during settling.

6.2.4.2 Sample handling and transport

Long-distance transport and prolonged storage of the sample should be avoided. Agitation of the sample during transport and biological activity during storage may severely influence the settling behaviour. If possible, perform the measurements on-site at the WWTP immediately after sampling.

6.2.4.3 Concentration range

Hindered settling occurs typically between concentrations of 1 g L^{-1} and 6 g L^{-1}. However, these limits are dependent on the flocculation state of the sludge and may be different between WWTPs. Therefore, there should always be a visual check on whether the recorded batch settling curves are within the hindered settling region. At low initial concentrations, it becomes increasingly difficult to track the solid-liquid interface as the sludge enters the discrete settling regime. If no distinct interface can be observed, this concentration should not be considered in the analysis. On the other hand, at high concentrations, sludge starts to undergo compression. If the initial concentration is high enough for compression to occur at the very beginning of the experiment, the batch curve will no longer show a clear linear descent. If no linear decrease in the batch curve can be seen, the high concentration should also not be considered in the analysis.

6.2.4.4 Measurement frequency

The measurement times and dilution series described in sections 6.2.2 and 6.2.3 can be considered as a minimum set of measurements for a batch curve. However, more frequent measurement times or additional dilutions can be added depending on the case-specific conditions or requirements. For example, if the solid-liquid interface shows a very rapid initial increase then additional measurements can be recorded during the first 2 min of sampling. If the appropriate specialised equipment and experience is available then the solid-liquid interface may even be tracked automatically (Vanderhasselt and Vanrolleghem, 2000). Depending on the concentration of the recycle flow sample, the standard dilution series (100, 80, 60, 50, 40 and 20 %) may not provide sufficient coverage of the concentration range for hindered settling. Additional dilutions such as 55 %, 30 % etc. can be added.

6.2.5 Recent advances in batch settling tests

By measuring a batch curve, the velocity at which the suspension-liquid interface passes through different settling regions can be investigated. However, the drawback of this method is that it only provides information on the suspension-liquid interface. No information on the settling behaviour inside the sludge blanket or the actual build-up of the sludge blanket is recorded. Nor does it allow the settling velocity in the

compression stage to be calculated. More advanced measurement techniques aiming to provide more detailed information on the sludge settling behaviour over time and depth have been presented in dedicated literature. Examples include detailed spatio-temporal solids concentration measurements by means of a radioactive tracer (De Clercq *et al.,* 2005) and velocity measurements throughout the depth of the sludge blanket with an ultrasonic transducer (Locatelli *et al.,* 2015). However, these techniques require specialised equipment and cannot be routinely performed.

6.3 MEASURING FLOCCULATION STATE OF ACTIVATED SLUDGE

As can be seen from Figure 6.1, the settling behaviour of a sludge sample is not only influenced by its concentration but also by its flocculation state. Hence, the ability of an SST to act successfully as a clarifier is highly dependent on the potential of the microorganisms to form a flocculent biomass which settles and compacts well, producing a clear effluent (Das *et al.,* 1993). The aim of the flocculation process is to combine individual flocs into large and dense flocs that settle rapidly and to incorporate discrete particles that normally would not settle alone. If the flocculation process or breakup of flocs fails during the activated sludge process, a fraction of the particles is not incorporated into flocs. They remain in the supernatant of the SST due to lack of sufficient mass and are carried over the effluent weir, reducing the effluent quality. Failure of the settling process can have multiple causes: (*i*) denitrifying sludge, (*ii*) excessively high sludge blankets, (*iii*) poor flocculation, or (*iv*) poor hydrodynamics. In order to take appropriate remedial actions, it is important to be able to pinpoint the cause of the failure. Denitrifying sludge and high sludge blankets can be easily recognised and corrected (Parker *et al.,* 2000). Distinguishing between flocculation problems and poor hydrodynamics is more challenging but can be accomplished by means of a Dispersed Suspended Solids/Flocculated Suspended Solids (DSS/FSS) test (Wahlberg *et al.,* 1995)

6.3.1 DSS/FSS test

6.3.1.1 Goal and application

Wahlberg *et al.* (1995) proposed a procedure that makes it possible to distinguish between hydraulic and flocculation problems in a given SST, the so-called DSS/FSS test. Using the DSS and/or FSS test has been proven to be a useful technique in several studies: it makes it possible (*i*) to assess the flocculation and deflocculation processes in transmission channels (Das *et al.,* 1993; Parker and Stenquist, 1986; Parker *et al.,* 1970), (*ii*) to determine the influence of hydraulic disturbances in the aeration basin on the effluent non-settleable sludge particles (Das *et al.,* 1993; Parker *et al.,* 2000, 1970) and (*iii*) to determine the benefits of a flocculation procedure in decreasing effluent suspended solids in a WWTP (Parker *et al.,* 2000; Wahlberg *et al.,* 1994).

The DSS/FSS test can be divided into three parts: the Effluent Suspended Solids (ESS) test, the Dispersed Suspended Solids (DSS) test, and the Flocculated Suspended Solids (FSS) test. The ESS test consists of a simple TSS test to determine the effluent concentration. The procedures for the DSS and FSS test are provided in sections 6.3.1.3 and 6.3.1.4.

6.3.1.2 Equipment

The following equipment is needed for the execution of ESS, DSS and FSS tests:

General

a. Equipment for the TSS test (APHA *et al.,* 2012).
b. A stopwatch.

ESS test

a. An effluent sample (minimum 0.5 L).

DSS test

b. A Kemmerer sampler.
c. A siphon for supernatant sampling.

FSS test

a. A square flocculation jar of at least 2.0 L.
b. Six paddle stirrers.
c. An activated sludge sample (minimum 1.5 L).

6.3.1.3 DSS test

Dispersed Suspended Solids (DSS) are defined as the concentration of SS remaining in the supernatant after 30 min of settling (Parker *et al.,* 1970). The DSS test thus quantifies an activated sludge's state of flocculation at the moment and location the sample is taken. This is accomplished by the use of a single container (i.e. a Kemmerer sampler) for sampling and settling in order to protect the biological flocs in the sample from any

secondary flocculation or breakup effects caused by an intermediate transfer step.

A Kemmerer sampler is a clear 4.2 L container, 105 mm in diameter and 600 mm tall with upper and lower closures (Figure 6.11). The sample is collected in the Kemmerer sampler and allowed to settle for 30 min, after which time the supernatant is sampled using a siphon and analysed for SS concentration (Wahlberg *et al.,* 1995).

Figure 6.11 A Kemmerer sampler with open upper and lower closures (photo: Royal Eijkelkamp).

The large, settleable flocs settle in the 30 min period, whereas the dispersed, primary particles not incorporated in the settling sludge remain in the supernatant. DSS concentrations have been shown to closely approximate the ESS concentration for a well-designed and operated SST (Parker and Stenquist, 1986). Hence, large deviations between ESS and DSS indicate clarification problems. DSS tests can be performed with samples at several locations in the SST (for example: at the SST inlet, at the outlet of the flocculation well, or near the effluent weir) to analyse where potential problems (and for instance flocculation or breakup) are occurring.

- **Protocol**

1. Immerse the Kemmerer sampler at the desired location in the SST to grab a sample.
2. Allow this sample to settle for 30 min.
3. After 30 min sample 500 mL of the supernatant (be careful not to disturb the settled sludge).
4. Analyse the sampled supernatant for SS concentration.

- **Example**

Parker *et al.* (2000) illustrated the use of a DSS test for SST failure troubleshooting in the case of the Central Marin Sanitation Agency plant in California. A DSS test was performed at the plant as high ESS values were being observed during peak flows. The results of the DSS test are provided in Table 6.7.

Table 6.7 Measured DSS values at the Central Marin Sanitation Agency plant in California (Parker *et al.,* 2000).

DSS (mg L^{-1}) Inlet centre well	DSS (mg L^{-1}) Outlet centre well	DSS (mg L^{-1}) Effluent weir	ESS (mg L^{-1}) Effluent weir
10.4	11.0	3.6	8.5

From these results, it became clear that no flocculation was occurring in the centre well as the DSS values at the inlet and outlet of the centre well are similar. However, the significant reduction in DSS values between the outlet of the centre well and the effluent weir showed that flocculation was occurring in the sedimentation tank. This indicated that the sludge had a good flocculation tendency but merely lacked proper conditions for flocculation in the existing centre well, which was contributing to its small diameter.

Moreover, the DSS results uncovered a significant hydraulic problem in the clarifiers. The high ESS concentration compared to the DSS at the effluent weir signifies the wash out of settleable solids over the effluent weir of the clarifier.

6.3.1.4 FSS test

Wahlberg *et al.* (1995) developed a complimentary test to the DSS test called the Flocculated Suspended Solids (FSS) test. Whereas the DSS test assesses the state of flocculation of a sample at a specific location in the SST at a moment in time, the FSS test quantifies the flocculation potential of an activated sludge sample by flocculating the sample under ideal conditions prior to settling.

For this test, an activated sludge sample is collected in a square flocculation jar (minimum jar volume 2 L). The sample volume should be at least 1.5 L. The sample is gently stirred for 30 min at 50 rpm (Figure 6.12) before it is allowed to settle for 30 min and the concentration in the supernatant is measured. Flocculation is maximized by stirring, and settling is performed in an ideal device (without hydraulic disturbances). Hence, the measured FSS is considered to be the minimal possible ESS.

Because the FSS concentration is measured under conditions of maximum flocculation and ideal settling, it will not change between the aeration basin and the SST. Therefore the activated sludge sample for the FSS test can be collected anywhere between the aeration basin and the SST.

Figure 6.12 Experimental setup for the FSS test. An activated sludge sample is stirred for 30 min in a square flocculation jar (photo: E. Torfs).

- **Protocol**

1. Collect an activated sludge sample of 1.5 L from the WWTP.
2. Pour this sample into a square flocculation jar with a volume of 2 L.
3. Stir the sample for 30 min. at 50 rpm.
4. Stop stirring and allow the sample to settle for 30 min.
5. After 30 min, sample 500 mL of the supernatant (being careful not to disturb the settled sludge).
6. Analyse the sampled supernatant for suspended solids concentration.

6.3.1.5 Interpretation of a DSS/FSS test

A well-functioning SST provides proper conditions for flocculation in order to incorporate small, dispersed solids that do not have sufficient mass to settle in the SST into flocs. Failure of the SST with respect to this function will result in high ESS concentrations. Dispersed suspended solids exist as a result of three possible mechanisms; (*i*) their flocculation is prevented by surface chemistry reactions (i.e. a biological flocculation problem), (*ii*) they are not incorporated into flocs due to insufficient time for flocculation (i.e. a physical flocculation problem), or (*iii*) they have been sheared from a floc particle due to excessive turbulence (i.e. a hydraulic problem). A DSS/FSS test makes it possible to distinguish between these different scenarios in order to take appropriate remedial actions.

A typical DSS/FSS test consists of four measurements: the ESS concentration, the DSS concentration at the inlet of the SST (DSSi), the DSS concentration at the effluent weir (DSSo) and the FSS concentration. If a flocculation well is present, then DSSi should be measured after this structure. Assuming that the system under study is struggling with high ESS concentrations, because DSS/FSS tests are typically performed in the case of clarification failure, four scenarios can be defined. A DSS/FSS troubleshooting matrix which shows the cause of the poor performance under the various testing scenarios is provided in Table 6.8 (Kinnear, 2000).

Table 6.8 DSS/FSS troubleshooting matrix (Kinnear, 2000).

ESS high and:		FSS	
		High	Low
DSS	High	Biological flocculation	Physical flocculation
	Low	Not possible	Hydraulics

The different testing scenarios can be interpreted as follows.

High DSSi - low FSS

This scenario indicates poor flocculation in the SST even though the activated sludge has good flocculating properties. Either the activated sludge is not receiving adequate time to flocculate or significant floc breakup is occurring prior to settlement (for example, by excessive shear in a conveyance structure). The clarification failure can be contributing to a flocculation problem of a physical nature that can be solved by either removing the cause of breakup or by incorporating an additional flocculation step prior to settling.

High DSSi - high FSS

As in the high DSSi - low FSS case, these results indicate poor flocculation in the SST. However, even under the ideal flocculation circumstances provided by the FSS test, the clarification cannot be improved. Hence, additional flocculation will not improve the clarification and the problem is most likely of a biological nature resulting in a sludge with poor flocculation properties. Modifications to the SST will not solve this problem; attention must be directed upstream of the SST.

Low DSSi - low FSS

The low DSSi suggests that the incoming activated sludge is in a well-flocculated state. As the DSSi is already in the same range as the FSS concentration, further flocculation will not improve the SST performance and the problem is most likely a hydraulic one (for example, due to short-circuiting). Comparing the DSSo and ESS concentration can provide further confirmation; if the DSSo concentration is significantly lower than the ESS concentration, then hydraulic scouring of settleable solids from the sludge blanket is indicated. To improve the clarification in this case, the tank's hydrodynamics need to be investigated by means of dye tests and/or 2-3D CFD (computational flow dynamics) modelling.

Low DSSi - high FSS

This outcome is theoretically not possible. Should it occur it is recommended to repeat the test.

- **Example**

A DSS/FSS test was used to assess the performance of the existing clarifiers prior to plant expansion at the Greeley Water Pollution Control Facility in Colorado (Brischke *et al.,* 1997; Parker *et al.,* 2000). The DSS test was performed at two locations i.e. at the inlet to the SST and at the effluent weir. The measured DSS, FSS and ESS values are shown in Table 6.9.

Table 6.9 Measured DSS, FSS and ESS values at the Greeley Water Pollution Control Facility in Colorado (Brischke *et al.,* 1997).

DSSi (mg L^{-1})	DSSo (mg L^{-1})	FSS (mg L^{-1})	ESS (mg L^{-1})
29.2	22.0	8.2	25.5

High DSS values both at the inlet and near the effluent weir indicate that no flocculation is occurring in the tank. The much lower FSS value signifies that the sludge has a high potential to flocculate but lacks appropriate conditions for flocculation in the tank. The high ESS concentrations can thus be attributed to a physical flocculation problem that can be solved by physically modifying the tank in order to provide suitable flocculation conditions. In this specific case this was accomplished through modifications of the centre well.

6.3.2 Recommendations

6.3.2.1 Flocculation conditions

Proper execution of an FSS test requires ideal flocculation conditions. Therefore, it is important to use a square flocculation jar in order to avoid the formation of a vortex during mixing. Moreover, make sure that the sample is completely mixed (i.e. no dead zones at either the top or bottom of the flocculation jar).

6.3.2.2 Temperature influence

The sample volume for the FSS test is relatively small in comparison to the volume of the Kemmerer sampler. Hence, some precautions should be taken to ensure that the samples do not change drastically in temperature during the 1 h it takes to conduct the test. For example: do not perform the test in direct sunlight.

6.3.2.3 Supernatant sampling

Regardless of the specific supernatant sampling technique, care should be taken not to pull any floating debris or settled solids into the supernatant sample as this can severely alter the results. Moreover, as the concentration in effluent and supernatant is generally very low, the sampled volume for the TSS test should be sufficiently large ($\pm$ 500 mL).

6.3.3 Advances in the measurement of the flocculation state

From the above it becomes clear that 'good bioflocculation' of the activated sludge is a prerequisite for 'good sedimentation' and a good effluent quality. Hence, what is the definition of a well-flocculated activated sludge floc? An activated sludge floc is composed of: (*i*) a backbone of filamentous organisms, onto which, (*ii*) microcolonies (i.e. clusters of micro-organisms) can attach and this aggregation of micro-organisms is then embedded in a matrix of (*iii*) extracellular polymer substances (EPS) (Figure 6.13). Whenever one of these components is not in balance with the rest then problems might be encountered.

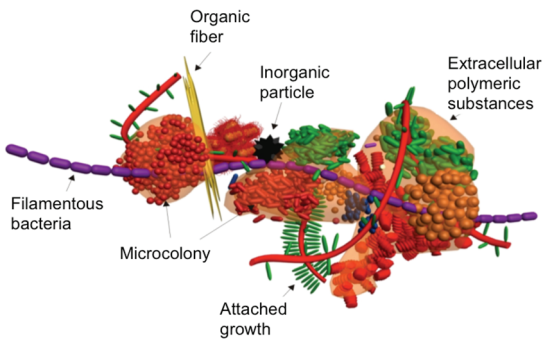

Figure 6.13 The structural makeup of an activated sludge floc: microcolonies attach to filamentous bacteria which form the backbone of the floc, while extracellular polymeric substances constitute the embedding matrix (Nielsen *et al.,* 2012).

One of the most common settling problems is that of filamentous bulking where there is a dominance of filamentous organisms that will make the floc structure very open and will retain a lot of water. Such filaments might even entangle with other protruding filaments so that a network is formed that prevents the sludge from settling. In contrast, when there are not enough filaments to form the backbone, so-called pinpoint flocs are observed. These are small clusters of micro-colonies that do not settle well. Hence, a good floc that settles well should be dense (not open) and sufficiently large.

Such characteristics of activated sludge can be quantified by microscopic image analysis. Changes in the average floc size, filament length, floc roundness, floc fractal dimension etc. can reveal a lot of information on the settling behaviour of sludge. While some research groups have been developing specific image analysis software to infer this information (Amaral and Ferreira, 2005; Da Motta *et al.,* 2001a, 2001b; Jenneé *et al.,* 2004; van Dierdonck *et al.,* 2013), also freeware software is available (such as ImageJ or FIJI (http://fiji.sc/Fiji)) to perform a basic analysis. A comprehensive overview of what is currently available in this image analysis domain is available in dedicated literature (Mesquita *et al.,* 2013). For microscopic monitoring that is more focused on revealing the specific microbial communities present, FISH analysis can be interesting; the reader is referred to the FISH handbook for biological wastewater treatment (Nielsen *et al.,* 2009).

As well as image analysis, three additional bioflocculation-related monitoring tools can be mentioned that are more focused on the forces that hold the floc components together. On the one hand, the global floc strength measurement (Mikkelsen and Keiding, 2002) compares the turbidity of the supernatant before and after shearing of a sludge sample. Lower turbidity after shearing indicates better bioflocculation. On the other hand, relative hydrophobicity and surface charge can be measured. The relative hydrophobicity is related to hydrophobic interactions that prove to be important in keeping the floc aggregated. The value that results from such an analysis (e.g. the MATH test, Chang and Lee, 1998) which assesses the microbial adhesion tendency to hydrocarbons) should not be taken as an absolute value but can be interesting in revealing changes over a certain operational period. The surface charge is related to electrostatic forces; with a more neutral activated sludge surface, the aggregates experience less repulsion resulting in improved coagulation and flocculation. The measurement of surface charge is based on a colloid titration technique (Kawamura and Tanaka, 1966; Kawamura *et al.,* 1967; Morgan *et al.,* 1990).

6.4 MEASURING THE SETTLING BEHAVIOUR OF GRANULAR SLUDGE

6.4.1 Goal and application

In recent years new technologies have been developed to improve the separation of sludge from the treated effluent. One of these technologies is the use of aerobic granular sludge. Aerobic granules are spherical biofilms with a typical shape factor of 0.7 - 0.8 (Beun *et al.,* 2002). Whereas conventional activated sludge is characterized

by settling velocities below 5 m h^{-1} (Vanderhasselt and Vanrolleghem, 2000), granular sludge settles significantly faster with settling velocities in the range of 10 up to 100 m h^{-1} (Bassin *et al.,* 2012; Etterer and Wilderer, 2001; Winkler *et al.,* 2012, 2011a). Moreover, granules show a low flocculating tendency positioning their settling behaviour at the far left side of Figure 6.1. The granules will thus settle independent even at higher concentrations (with almost no hindered and compression regime present) and directly form a compact sludge bed. For this reason the SVI after 5 minutes will be approximately the same as the SVI measured after 30 minutes. Typical SVI values for granules are less than 30 ml/g (de Kreuk and van Loosdrecht, 2004; Liu *et al.,* 2005; Liu and Tay, 2007; Tay *et al.,* 2004).

Granular sludge is developed in Sequencing Batch Reactors (SBR) as these systems fulfil a number of specific requirements for the formation of granules: a feast - famine regime for the selection of appropriate microorganisms (Beun *et al.,* 1999), short settling times to ensure retention of granular biomass and wash-out of flocculent biomass (Qin *et al.,* 2004) and sufficient shear force to ensure an optimal physical granule integrity (Tay *et al.,* 2001).

One of the most important parameters to select for granular sludge is the settling velocity. By applying short settling times in an SBR, only large biomass aggregates that settle well are selected, while flocculent sludge is washed out (Beun *et al.,* 2000). The parameters determining the settling velocity of particles and in turn biomass washout are of crucial importance to granular sludge technology. The balances of forces for the sedimentation of a spherical particle depend on the buoyancy, gravity and drag force (Giancoli, 1995). From this relation, the settling velocity is influenced by the water viscosity, particle size and shape, and the difference between the density of the water and the particles. Hence, the settling velocity of granules is influenced by the density and size of the particles where an increase in diameter affects the settling velocity more severely (Winkler *et al.,* 2012, 2011a). Therefore, this section presents a method to measure the density and size of granules as well as a procedure to calculate the theoretical settling velocity of granules under different temperature conditions.

6.4.2 Equipment

For granular sludge tests the following equipment is needed:

a. A pycnometer.
b. A microbalance.
c. Sieves or a microscope with an image analyser.

6.4.3 Density measurements

Granule densities between 1,036 - 1,048 kg m^{-3} have been reported (Etterer and Wilderer, 2001), which are comparable to densities of conventional activated sludge (1,020-1,060 kg m^{-3}) (Andreadakis, 1993; Dammel and Schroeder, 1991). However, precipitates may form within the granule core (Lee and Chen, 2015; Mañas *et al.,* 2012) and significantly increase the granule density up to 1,300 kg m^{-3} (Juang *et al.,* 2010; Winkler *et al.,* 2013).

The specific biomass density can be measured with a pycnometer (Figure 6.14). A pycnometer is a simple and inexpensive glass flask, with an exactly calibrated volume. The pycnometer flask is closed with a glass stopper that acts as a valve; it contains a small groove, through which excess water is forced out when closed. The pycnometer has a known volume V.

Figure 6.14 Picture of a pycnometer (photo: E. Torfs).

- **Protocol**

1. Measure the weight of the pycnometer (closed with the glass stopper) in a completely dry state (m_0).
2. Fill the pycnometer with water and measure its weight again (m_T). It is very important to dry the pycnometer carefully before the weight is determined in order to remove all excess water from the outside of the pycnometer.

3. Calculate the mass of water in the pycnometer (m_{H2O}) as:

$$m_{H2O} = m_T - m_0 \quad \text{Eq. 6.11}$$

4. Measure the weight of the granule sample for which you want to determine the density (m_s).
5. Place the granule sample inside the pycnometer and determine the weight of the pycnometer together with the inserted sample ($m_0 + m_s$).
6. Fill the pycnometer (containing the solids sample) further with water and weigh its mass again (m_{TS}). As in Step two, make sure that the pycnometer is dried carefully before the weight is determined.
7. Calculate the weight of the added water (m'_{H2O}) as:

$$m'_{H_2O} = m_{TS} - m_o - m_s \quad \text{Eq. 6.12}$$

8. Determine the volume of added water (V'_{H2O}) according to:

$$V'_{H_2O} = \frac{m'_{H_2O}}{\rho_{H_2O}} \quad \text{Eq. 6.13}$$

9. Calculate the volume of measured solids V_s from the difference between the volume of water that fills the empty pycnometer (V) and the previously determined volume of water (V'_{H2O}).

$$V_s = V - V'_{H_2O} = \frac{m_{H_2O} - m'_{H_2O}}{\rho_{H_2O}} \quad \text{Eq. 6.14}$$

10. Finally, calculate the density of the granules ρ_s as:

$$\rho_s = \frac{m_s}{V_s} \quad \text{Eq. 6.15}$$

- **Example**

A pycnometer with a volume (V) of 0.1 L is used to determine the density of a granular sludge sample. All the measurements and calculations are provided in Table 6.10 (the density of water, ρ_{H_2O}, at 20 °C is 998 g L^{-1}).

Table 6.10 Density calculation for a granular sludge sample.

Variable	Symbol	Procedure	Value
Mass empty pycnometer (g)	m_0	Measured	54.51
Mass pycnometer and water (g)	m_T	Measured	153.70
Mass water (g)	m_{H2O}	Calculated	99.19
Mass solids (g)	m_s	Measured	19.56
Mass pycnometer, solids and water (g)	m_{TS}	Measured	154.55
Mass added water (g)	m'_{H2O}	Calculated	80.48
Volume added water (L)	V'_{H2O}	Calculated	0.08
Volume solids (L)	V_s	Calculated	0.02
Density solids (g L^{-1})	ρ_s	Calculated	1,010.40

6.4.4 Granular biomass size determination

Although there is no common consensus on the minimum diameter (Bathe *et al.,* 2005), sieves with a diameter of 0.2 mm have been used to determine the minimum size of granular biomass (Bin *et al.,* 2011; de Kreuk, 2006; Li *et al.,* 2009). The largest diameter reported is 16 mm (Zheng *et al.,* 2006), but typically diameters range between 0.5-3 mm (de Kreuk and van Loosdrecht, 2004; Shi *et al.,* 2009; Winkler *et al.,* 2011b). The size distribution can be measured either by simple sieving tests or by means of an image analyser.

6.4.4.1 Sieving

Granule size can be determined by means of sieves with different mesh sizes. The screening can be performed with sieves with mesh openings of, i.e. 2.0, 1.0, 0.5 and 0.3 mm, making it possible to cover the most common granule size range.

Figure 6.15 Stacked sieves with different mesh openings (photo: Fieldmaster)

- **Protocol**

1. Measure the total wet weight of a sample.
2. Mount the sieves vertically one on top of the other in increasing order of mesh opening (from bottom to top) so that the coarsest mesh is at the top (Figure 6.15).
3. Pour the granule sample onto the sieves.
4. Wash each sieve successively to allow the granules to move from one sieve to the next.
5. Filter the liquid (which has trickled through all the sieves) in order to collect particles smaller than 0.3 mm.
6. Backwash each sieve to retain each granule fraction in a separate beaker.
7. Determine the wet weight, and if needed the dry weight (TSS), the ash content and the VSS of each fraction, which will result in the theoretical percentage of each class size (Laguna *et al.,* 1999).

6.4.4.2 Image analyser

Alternatively, the granule size can be measured by means of an image analyser using the averaged projected surface area of the granules. For this method, a sample is transferred into a petri dish and placed under a stereo microscope with a fixed magnification (e.g. 7.5 × magnification). Each image analysed is recorded by the image analyser. The Petri dish needs to be turned multiple times in order to measure different granules. Different image analysers are available on the market and each requires different handling. An example of granule size distribution data, created during the measuring procedure, is presented in Figure 6.16.

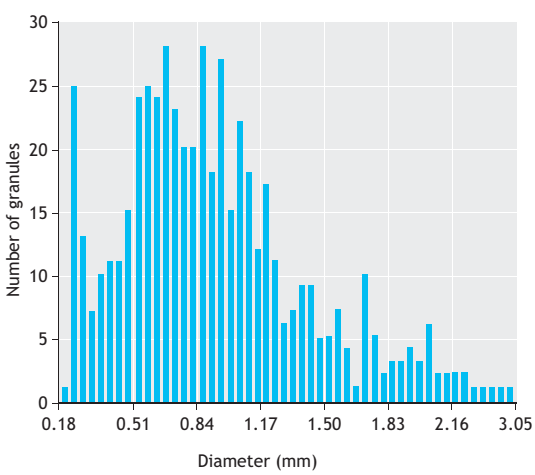

Figure 6.16 Particle size distribution of a granular sludge reactor.

6.4.5 Calculating the settling velocity of granules

- **Protocol**

The measured average density and diameter of granules can be used to calculate the theoretical settling velocity. For particle Reynolds numbers smaller than or equal to 1, Stokes' law can be used to calculate the settling velocity of a particle.

$$v_s = \frac{g}{18} \cdot \frac{\rho_p - \rho_w}{\rho_w} \cdot \frac{d_p^2}{\nu_w} \qquad \text{Eq. 6.16}$$

The Reynolds numbers can be calculated with the following equation:

$$Re = d_p \cdot \frac{v_s}{\nu_w} \qquad \text{Eq. 6.17}$$

Where, v_s is the sedimentation velocity of a single particle (m s^{-1}), d_p is the particle diameter (m), ρ_p is the density of a particle (kg m^{-3}), ρ_w is the density of the fluid (kg m^{-3}), g is the gravitational constant (9.81 m s^{-2}), ν_w is the kinematic viscosity of water (m^2 s^{-1}), and Re_p is the particle Reynolds number.

The density and viscosity of the medium depend on the temperature and the solutes present in the water. With increasing temperature, the viscosity and density of the water decrease. At high temperature, water molecules are more mobile than at low temperature, resulting in a decrease of viscosity by a factor of two between 10 and 40 °C (Podolsky, 2000). A table with density and viscosity values of water at different temperatures can be found elsewhere.

- **Example**

An example of a calculation for the settling velocity of a particle with a diameter of 0.4 mm and 1,010 kg m^{-3} at different temperatures is given in Table 6.11 and plotted in Figure 6.17. Note that the condition of Reynolds numbers smaller than or equal to 1 is not met for every temperature. The implications of this will be discussed further on in this section.

Table 6.11 Settling velocity of a granule with a particle diameter of 0.4 mm and a particle density of 1,010 kg m^{-3} at different temperatures.

T	°C	5	10	15	20	25	30	35	40
ν_w	$m^2 s^{-1}$	1.5e-06	1.3e-06	1.1e-06	1.0e-06	9.4e-07	8.2e-07	7.4e-07	6.6e-07
ρ_w	kg m^{-3}	1,000	1,000	999	998	997	996	994	992
Re	-	0.2	0.2	0.2	0.4	0.5	0.7	1.0	1.5
v_s	m h^{-1}	2.1	2.5	3.0	3.6	4.2	5.5	6.8	8.7

Table 6.12 Settling velocity of a granule with a particle diameter of 0.4 mm and a particle density of 1,050 kg m^{-3} at different temperatures.

T	°C	5	10	15	20	25	30	35	40
ν_w	$m^2 s^{-1}$	1.5e-06	1.3e-06	1.1e-06	1.0e-06	9.4e-07	8.2e-07	7.4e-07	6.6e-07
ρ_w	kg m^{-3}	1,000	1,000	999	998	997	996	994	992
Re	-	0.8	1.0	1.4	1.7	2.1	2.8	3.6	4.7
v_s	m h^{-1}	10.7	12.1	14.0	15.8	17.7	20.9	23.9	27.9

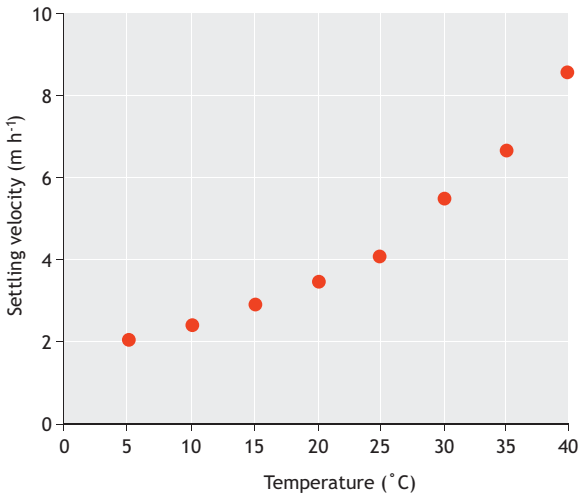

Figure 6.17 Calculated settling velocities at different temperatures for granules with a diameter of 0.4 mm and a density of 1,010 kg m^{-3}.

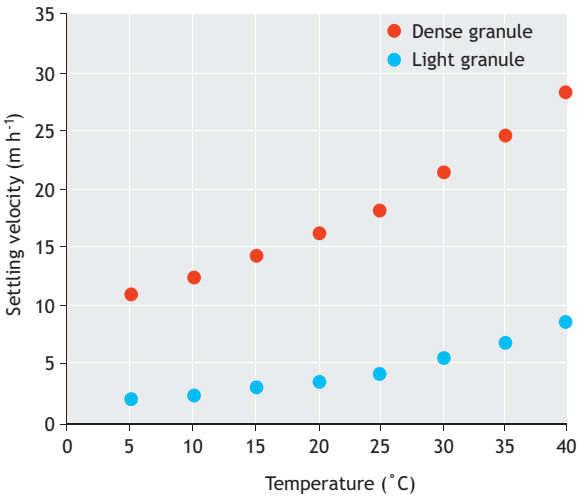

Figure 6.18 Calculated settling velocities at different temperatures for a light (1,010 kg m^{-3}) and dense (1,050 kg m^{-3}) granular sludge particle at a diameter of 0.4 mm.

The example in Figure 6.17 shows that the settling velocity of a small granule with a diameter of 400 µm and a density of 1,010 kg m^{-3} varies between 2 m h^{-1} and 9 m h^{-1} for temperatures ranging between 5 – 40 °C (Winkler *et al.,* 2012). At low temperatures the separation of small and light granules from flocs (with typical settling velocities below ± 5 m h^{-1}) can therefore become troublesome. Earlier research has experimentally proven that a start-up process at cold temperatures is difficult. In addition all microbial processes run slower at low temperatures (Brdjanovic *et al.,* 1997; Kettunen and Rintala, 1997; Lettinga *et al.,* 2001), hence limiting granulation at a lower temperature even further. An increase in density or diameter of the granules will significantly increase the settling velocity and thus facilitate the separation.

- **Example**

Calculated settling velocities for a particle with the same diameter (0.4 mm) but a higher density (1,050 kg m^{-3}) at different temperatures are shown in Table 6.12 and Figure 6.18.

Larger and denser particles result in particle Reynolds numbers larger than 1. For these particles the theoretical settling velocities calculated according to Stokes' law will deviate from the true settling velocities. For most applications, these errors (< 10 %) are acceptable and Stokes' law remains a good approximation. More accurate velocity calculations are possible but require an advanced calculation method including an iterative procedure to determine Re. Illustrations of this approach can be found in Winkler *et al.* (2012).

6.4.6 Recommendations

6.4.6.1 Validation of results

The calculated settling velocities (Section 6.4.5) can be validated experimentally by conducting simple settling tests with granules harvested from sieves with different mesh sizes (Section 6.4.4). The different size fractions can be poured into the reactor column itself or into a

cylinder and the time is measured until the granule reaches the bottom of the reactor in order to express its settling velocity in meters per hour.

6.4.6.2 Application for flocculent sludge

The experimental methods described in sections 6.4.3 and 6.4.4 can also be applied to activated sludge in SSTs (particularly in the top region of SSTs where discrete settling is known to occur). However, this type of application is currently not very common because the high flocculation potential of sludge causes attention to be mainly directed towards hindered settling (as this is the dominant regime at the concentration where activated sludge enters the SST and has been mainly used to determine SSTs' capacity and design). For granular sludge on the other hand, the dominant settling regime is discrete thus causing the focus to shift to size and density measurements.

6.5 MEASURING SETTLING VELOCITY DISTRIBUTION IN PSTs

6.5.1 Introduction

Primary settling tanks (PSTs) are used as a pre-treatment in WWTPs. Measurement campaigns conducted since the early 1970s on urban discharges have clearly shown that many pollutants occur in particulate form. Moreover, particles transported in suspension have also emerged as highly settleable despite their relatively small particle size (30 to 40 µm). Hence, gravitational separation in PSTs can serve as a valuable tool to separate coarse, settleable particles in the raw wastewater prior to further treatment in the biological reactors.

As concentrations of particulate matter in raw wastewater are relatively low compared to concentrations in SSTs, the settling regime in PSTs has a discrete (non-flocculent and flocculent) nature and the settling velocity depends on the individual properties of particles. Given the variety of densities, shapes and sizes of suspended particles in wastewaters (have in mind Stokes' law), it is challenging and time-consuming to calculate the settling velocities of different particles from size and density measurements. Therefore, this section presents a method to directly measure the distribution of settling velocities in representative wastewater samples. This measurement can be performed using the ViCAs protocol, developed by Chebbo and Gromaire (2009). The ViCAs ('Vitesse de Chute en Assainissement', which is French for 'settling velocity in sanitation') is a test to measure the settling velocity of particles in a column under static conditions. The test provides insight into the behaviour of particles present in a wastewater sample in order to obtain an idea about its composition. As such it can serve as important information in different application domains. In primary settling tanks, results from ViCAs experiments can be used as input to primary clarifier models (Bachis *et al.,* 2015) or to study the effect of chemical dosage on the settling velocity distribution in order to improve chemically enhanced primary treatment (CEPT) performance. Furthermore, ViCAs experiments can be applied in the design of combined sewer retention tanks where knowledge of the settling velocity distribution can be used to determine an optimal HRT and corresponding load reduction to the treatment plant.

6.5.2 General principle

A ViCAs test is performed by settling a sample in a ViCAs column (Figure 6.19).

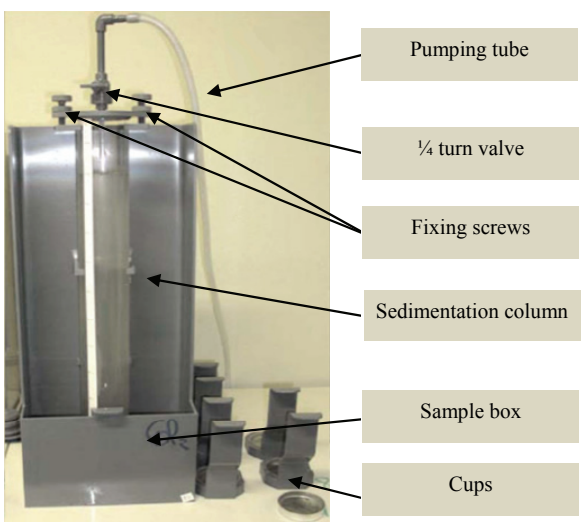

Figure 6.19 The ViCAs test equipment (photo: Chebbo and Gromaire, 2009).

The ViCAs protocol is based on the principle of homogeneous suspensions (Figure 6.20). At the beginning of the measurement, the solids are uniformly distributed over the whole sedimentation height. Then the particles are assumed to settle independently of each

other, without forming aggregates and without diffusion. The solids, after having settled for a predetermined period of time, are recovered at the bottom of the sedimentation column in cups.

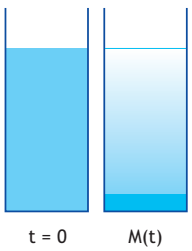

Figure 6.20 Principle of the homogeneous suspension.

Their mass is thus recovered from the cups, which allows the evolution of the cumulative mass M(t) of settled material as a function of time t to be determined (Figure 6.21).

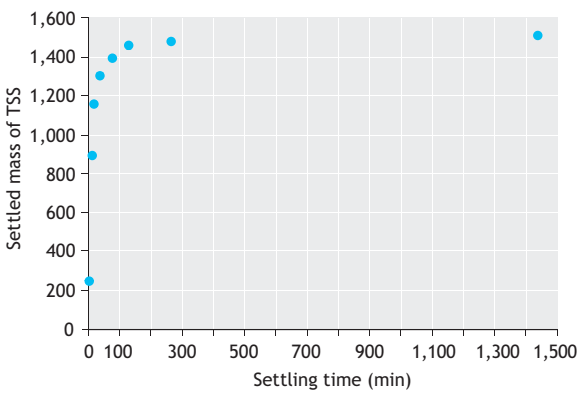

Figure 6.21 Cumulative evolution of the settled mass as a function of time.

In practice, the cumulative curve of settled mass consists of n points ($7 < n < 12$), corresponding to n samples taken after different settling times.

The measurements of the settled mass as a function of time make it possible to calculate the settling velocity distribution $f(v_s)$. Figure 6.22 shows an example of a settling velocity distribution curve for a typical wastewater sample.

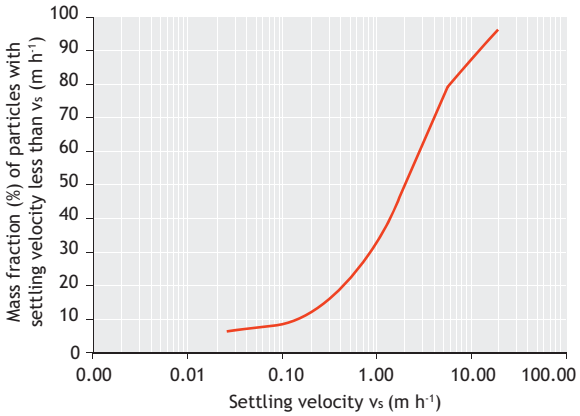

Figure 6.22 Settling velocity distribution curve $f(v_s)$ for a typical wastewater sample.

6.5.3 Sampling and sample preservation

The test can be carried out using a composite sample or by mixing several samples with similar composition (e.g. harvest 5 bottles of 1 L over an interval of 10 min). During sampling, suspended solids that may disturb the test are removed and, if necessary, the sample can be directly filtered with a coarse filter, which does not affect the sample composition. The analyses must be made within a maximum of 24 hours after collection, to avoid flocculation. If the initial concentration exceeds 1,000 mg L^{-1} then it is necessary to dilute the sample. To perform the dilution, the sample is split into two, so as to have 5 L on the one hand, and 15 L on the other. From the 15 L sample, 5 L of supernatant are withdrawn after 24 h of settling, and used to perform the dilution.

6.5.4 Equipment

For the execution of ViCAs protocol following equipment is needed:

1. A ViCAs column with its support and two associated cups.
2. A rubber band to hold the column in place.
3. A timer (to measure the time steps).
4. A beaker of 5 L.
5. A spatula (for homogenisation).
6. A vacuum pump.
7. A plastic connection tube.
8. A filtration Erlenmeyer of 1,000 mL to create a vacuum buffer and protect the pump.

Figure 6.23 shows the ViCAs equipment and the filling system ready for use.

Figure 6.23 ViCAs apparatus with filling equipment (photo: Gromaire M.C. and Chebbo G., ViCAs manual).

6.5.5 Analytical protocol

The sample to be analysed is poured into a sample box (Figure 6.19) and is rapidly aspirated by vacuum within a column. It is then maintained under vacuum for the whole duration of the test, i.e. the sample is hanging in the column. The particles settled during certain periods of time Δt are collected in cups placed under the column; both the cups and column have the same diameter. The cups are first filled with tap water and then each in turn immersed in the sample box and moved under the column. At the end of each particular period Δt, the content of the cups is filtered and the TSS and VSS of the recovered solids are measured.

- **Sample preparation**

a. Homogenize the sample using the spatula and pour 5 L into an appropriate beaker.
b. Stir again and take 1 sample of 500 mL that will be used to determine the initial TSS concentration in the column.

- **Filling the column**

a. Mix the sample of 4.5 L before it is poured quickly into the sample box (Figure 6.24A).
b. Suck the liquid into the column (in 2 to 5 seconds) and then close the valve by a ¼ turn (Figure 6.24B)
c. Stop the vacuum pump.

It is important to note that:

a. The filling phase requires some training because it has to be done very quickly.
b. For a more successful test, it is better to have two operators.
c. An insufficient volume of the sample or closing a valve too late may lead to air leakage into the column. The filling will then have to be repeated.
d. The use of a protective device such as a Woulff bottle is indispensable.

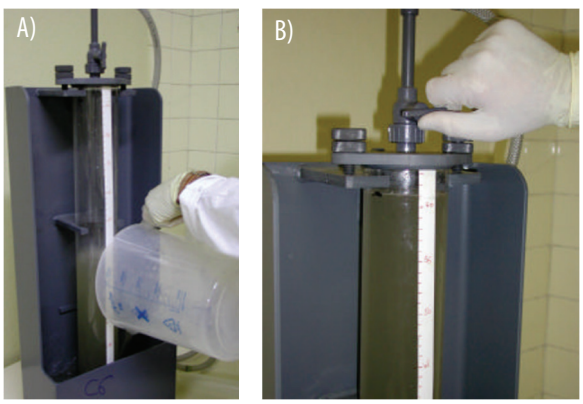

Figure 6.24 Filling the box with a sample (A) and closing the valve towards the vacuum (B) (photo: Gromaire M.C. and Chebbo G., ViCAs manual).

- **Start of the settling test**

a. Immediately after closing the valve with a ¼ turn, slide the first cup under the column: gently place the cup in the sample box, and slide it under the base of the column.
b. Start the timer and disconnect the pumping equipment.
c. Place a piece of adhesive tape to measure the height of water in the column at the end of the test (there may be a variation in water height due to the exchange of cups).

- **Changing the cups**

a. Ten seconds before the time of change, gently introduce a water-filled cup into the groove (Figure 6.25).
b. Slide the two cups gently to the new position and remove the old cup (Figure 6.26).

It is important to note that:

a. For a full analysis the change of cup is carried out at 2 min, 6 min, 14 min, 30 min, 1 h, 2 h, 4 h and over 22 h for a total of 8 cups.
b. This step is tricky because one must not lose the content of the cup and one must minimize any turbulence caused by moving the cups below the column.

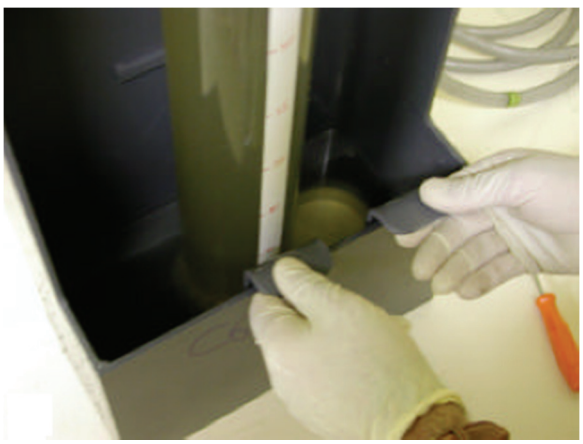

Figure 6.25 Introducing a new water-filled cup (photo: Gromaire M.C. and Chebbo G., ViCAs manual).

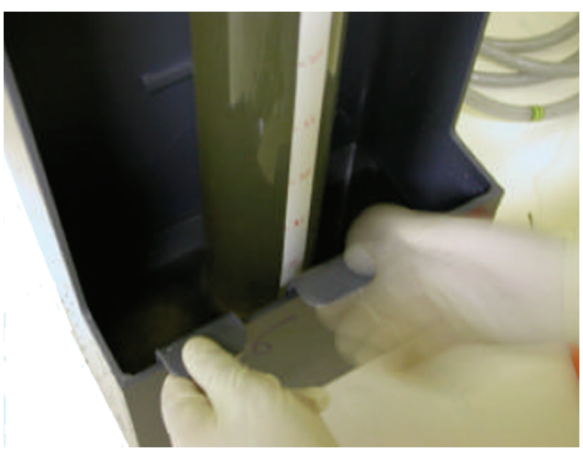

Figure 6.26 Moving the cups to their new positions (photo: Gromaire M.C. and Chebbo G., ViCAs manual).

- **Determination of the final concentration**

The final concentration in the column is determined by collecting the total volume of the column and analysing its content. This step makes it possible to perform a mass balance check, which is essential to determine the validity of the test.

a. After the last sample is taken, plug the end of the column, remove the column from its holder and pour its contents into the 5 L pitcher.
b. Mix the contents thoroughly and take a sample of 500 mL to determine the final TSS concentration.

- **Analysing the TSS and VSS**

The TSS and VSS of the initial, final and cup contents are measured according to methods 2540 D and 2540 E of Standard Methods (APHA *et al.,* 2012).

6.5.6 Calculations and result presentation

6.5.6.1 Mass balance check

A mass balance calculation is performed to estimate losses (or gains) of solids during the experiment and thus to assess the quality of the measurement.

The percentage mass balance error E (%) can be calculated as follows:

$$E = \frac{M_{ini} - (M_{set} + M_{fin})}{M_{ini}} \qquad \text{Eq. 6.18}$$

Where, M_{ini} is the initial mass in the column (mg), M_{fin} is the final mass in the column (mg), and M_{set} is the sum of the masses recovered in the cups (mg).

6.5.6.2 Calculation of the settling velocity distribution

A theoretical analysis (Chebbo and Bachoc, 1992; Chancellor *et al.,* 1998) shows that the cumulative curve M(t) can be written as:

$$M(t) = S(t) + t\frac{dM(t)}{dt} \qquad \text{Eq. 6.19}$$

Where, M(t) is the cumulated mass of particles settled to the bottom of the column between t = 0 and t, S(t) is the mass of particles settled between t = 0 and t that have a settling velocity above H/t, with H the water height in

the column, $t\frac{dM(t)}{dt}$ is the mass of particles settled at time t that have a settling velocity below H/t (and thus initially located at a height in the water column less than H).

In order to obtain the settling velocity distribution of the sample, it is necessary to determine the curve S(t) that can subsequently be transformed into the cumulative settling velocity distribution $f(v_s)$ (Figure 6.22).

Practically, a continuous function M(t) is numerically fitted to the measured values $M(t_i)$, and then used to analytically solve Eq. 6.20.

The following expression can be used for M(t):

$$M(t) = \frac{b}{1 + \left(\frac{c}{t}\right)^d} \qquad \text{Eq. 6.20}$$

Where b, c and d are three numerical parameters that can be determined by the least squares method. The following constraints must be respected: $0 < b \leq M_{init}$, $c > 0$ and $0 < d < 1$. An example of the result of fitting the curve is given in Figure 6.27.

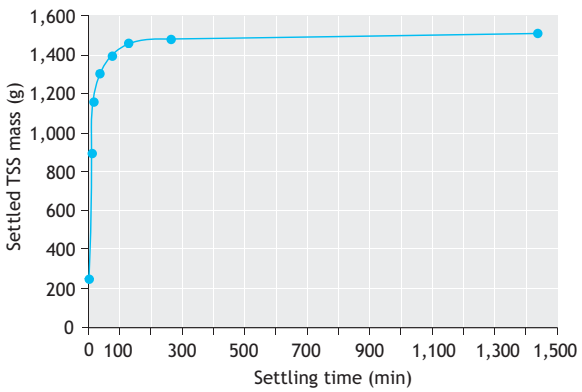

Figure 6.27 Example of a curve fitted to a cumulative series of settled masses.

The fitted curve M(t) can then be used to calculate S(t):

$$S(t) = M(t) - t \cdot \frac{dM(t)}{dt} = \frac{b \cdot \left(1 + (1-d) \cdot \left(\frac{c}{t}\right)^d\right)}{\left(1 + \left(\frac{c}{t}\right)^d\right)^2} \qquad \text{Eq. 6.21}$$

From which follows:

$$f(v_s) = 100 \cdot \left(1 - \frac{S(t)}{M_{set} + M_{fin}}\right) \text{ with } v_s = \frac{H}{t} \qquad \text{Eq. 6.22}$$

6.5.6.3 Recommendations

- **Manipulations**

Careful manipulations will allow mass balance errors below 10% to be achieved. An error exceeding 15 % should lead to an invalidation of the ViCAs analysis.

- **Sample frequency**

The sample times are those generally used to make the TSS fractionation of the sample. They may be subject to change depending on the project and the type of sample. Also, the user is free to change the Δt used, as long as a minimum interval of 7 and a maximum of 15 intervals is respected.

- **Reproducibility**

Reproducibility tests can be performed in order to confirm the results for a single ViCAs test. The success of the test is dependent on the meticulousness of the person in following the protocol and handling the equipment. Especially important is the changing of the cups below the column and the filtration of the recovered masses. Figure 6.28 shows the results of a reproducibility test on primary effluent from the Québec-Est wastewater treatment plant.

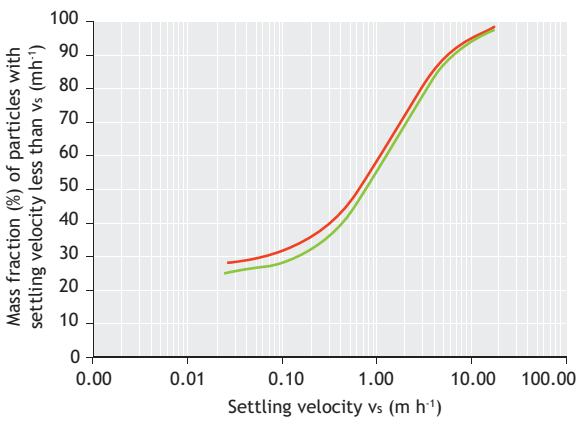

Figure 6.28 ViCAs reproducibility test.

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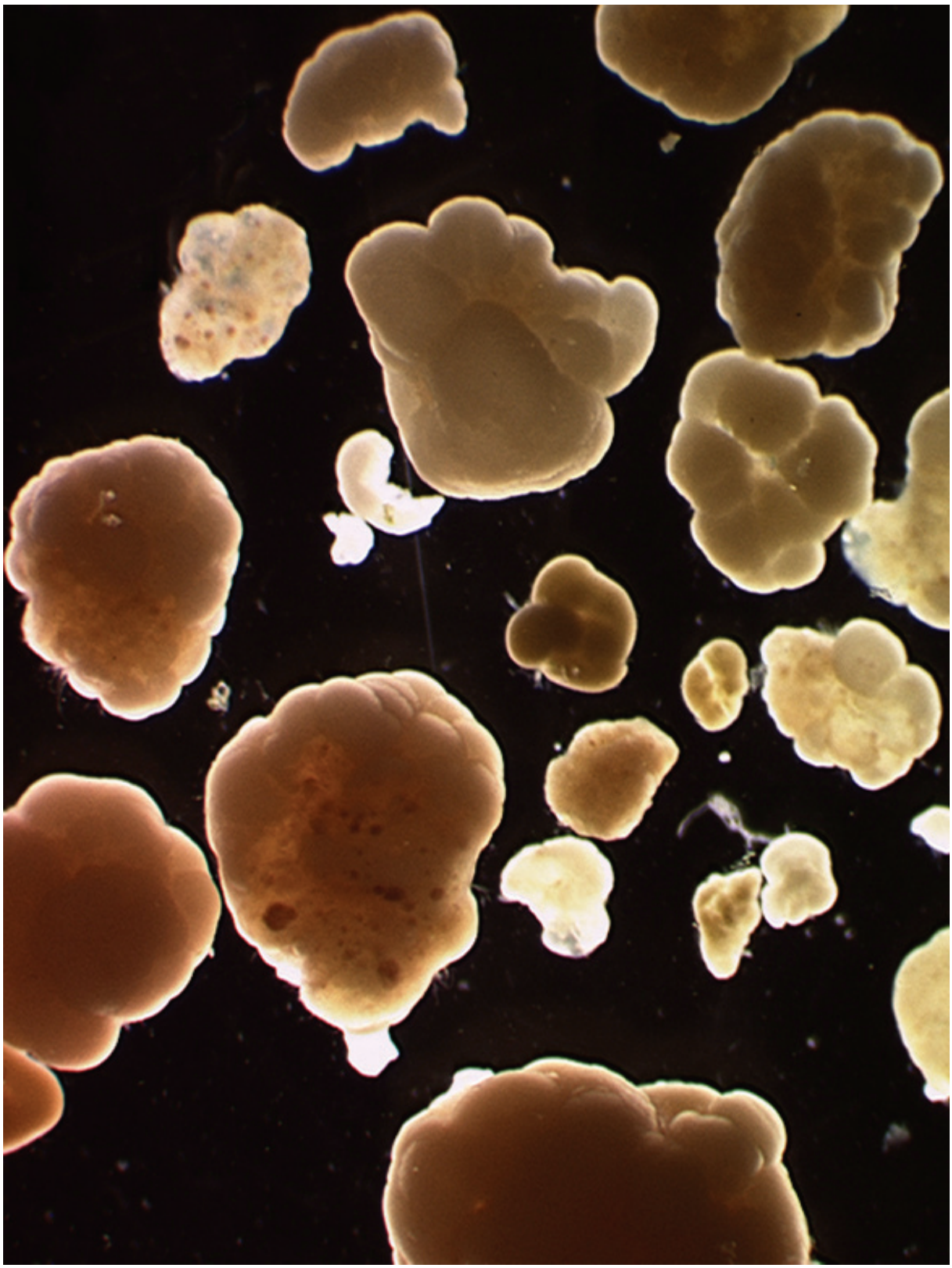

Figure 6.29 Granular sludge (photo: Beun *et al.,* 1999).