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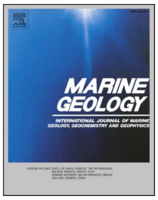

Research Article

A laboratory study on the behavior of estuarine sediment flocculation as function of salinity, EPS and living algae

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ABSTRACT

The interactions between organic and inorganic particles in the context of flocculation is an on-going topic of research. Most current researches do not distinguish between the effects of EPS (produced by microorganisms) and living microorganisms (like algae). In this study, the effect of salinity, EPS and living algae on sediment flocculation are investigated separately. Several types of measurements were performed, which can be divided into the following categories: sediment at different salinities, sediment in the presence of EPS at different salinities, sediment in the presence of living algae at a given salinity. Results show that increasing salinity enhances slightly sediment flocculation. In the presence of EPS there was hardly any flocculation in demi-water, but the flocculation was significant in saline water. The living algae cells were shown to flocculate with themselves and form large flocs. These algae flocs can bind to sediment particles to form larger flocs, both in demi-water and sea water. Size-wise algae-sediment flocs were largest, EPS-sediment flocs came second, and salt-sediment flocs were smallest.

1. Introduction

Sediment that is found in natural estuarine aquatic environment is usually transported as part of entities called flocs (Droppo and Ongley, 1994; Droppo and Ongley, 1992). The process of flocculation requires to be studied as flocculation, influenced by hydrodynamic and biogeochemical estuarine conditions, will modify sediment erosion, settling and transportation (Bianchi, 2007; Geyer et al., 2000; Jay et al., 2000).

In recent years, more and more researches focused on the biological processes related to flocculation. Flocculation between minerals and algae has been reported (Verspagen et al., 2006). Microorganisms can attach to the sediment surface in combination with biological activities, such as growth, metabolism and decay (Grossart et al., 2006). At the same time they can affect the flocculation processes due to their excretion of extracellular polymeric substances (EPS). EPS are typically polyelectrolytes (polysaccharides), usually anionic like galacturonic acid (Plude et al., 1991). Divalent cations (e.g. $MgCl_2$ and $CaCl_2$) can increase the EPS effects on flocculation (Park et al., 2010; Tan et al., 2008, 2014). Bridging by cationic ions is enhanced when divalent cations are used (instead of monovalent ions like NaCl) (Mietta et al., 2009a).

Cohesive sediment organic matter mainly consists of EPS (Winterwerp and Van Kesteren, 2004). EPS changes the properties of the sediment particle's surface hereby affecting the flocculation processes. Note that the effects of living organisms like (micro)algae on flocculation and the action of EPS on flocculation are generally not dissociated in researches on the topic. Some algae species can form large flocs due to their large cells size or cells chain length (de Lucas Pardo et al., 2013). They can combine with sediment particles and form bio-sediment flocs which consist of sediment particles and algae particles (Deng et al., 2021; Deng et al., 2019). The aim of this article is to distinguish between the effect of EPS in the absence of microalgae and the effect of microalgae on flocculation. To this end, a series of laboratory measurements were performed, where the effects of salinity, EPS and living algae on the estuarine sediment flocculation are reported. Three types of salts (NaCl, $MgCl_2$, $CaCl_2$) were used, at three different concentrations (defined as

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"low", "medium" and "high"). First, we investigated the variation of floc size and particle size distributions with different cation concentrations, then sediment added to EPS and mixed at different salinities was studied. Finally, we report the variation in particle size and particle size distribution of sediment with (micro)algae.

2. Methodology

2.1. Samples

The sediment samples used in the experiments were bed surface sediments collected from the maximum turbidity zone in the South Passage of the Changjiang estuary (the site where in-situ observations were conducted as reported in Deng et al. (2021)).

The sediment samples were collected during the in-situ observations, and they were kept at 4 °C to avoid changes in sediment properties. Before the experiment, the samples were taken out and placed at room temperature (at 20 ± 2 °C). The organic matter content of each sediment sample was measured by calcination method, and the organic matter content was found to be less than 3% in mass.

As *Skeletonema costatum* represents over 90% of the algae species in Changjiang Estuary, in particular at the study field station (He and Sun, 2009; Wu, 2015), this species was used in the laboratory tests. The algae were bought from the company Roem van Yerseke B.V. (The Netherlands) and used within a few days following the purchase. The approximate number of cells in the batch suspensions was given by the company. During the experiments, the concentration and number of algae were obtained according to the volume of the algae solution added. Finally, to ensure the survival of the algae, each experiment usually lasted no more than three hours. Every two weeks a fresh batch of algae was used to keep the bulk concentration constant.

The EPS used in this article was obtained from municipal waste water extractions (provided in-kind by the sanitary engineering section of the TUD) and contained 200 mg l^{-1} proteins, 40 mg l^{-1} humic acids and 110 mg l^{-1} polysaccharides.

The salt used ($NaCl$, $MgCl_2$ and $CaCl_2$) were obtained from Merck (Darmstadt, Germany). Artificial sea salt used in some experiment consisted of the four major components of sea salt: $NaCl$, $MgCl_2$, $CaCl_2$ and KCl (Kester et al., 1967). To test the effect of concentration on flocculation, different salinity ranges were used for each cation: $Na+$—0.1, 0.2, 0.3 mol l^{-1}, Mg^{2+}—0.01, 0.05, 0.1 mol l^{-1} and Ca^{2+}—0.005, 0.01, 0.02 mol l^{-1}. These three concentrations are defined as "low", "medium" and "high". All measurements were carried out at room temperature and the water temperature was 20 ± 2 °C.

2.2. Static Light Scattering

The static light scattering (SLS) experiments were performed using a Malvern Mastersizer (Mietta, 2010). The experimental device consists of mixing jar, pipes, paddle, pump and Malvern Mastersizer (Fig. 1). The principles of the LISST equipment used in-situ and the Malvern MasterSizer are quite similar (Filippa et al., 2011). From the SLS measurements a full particle size distribution (size range 2 nm–2 mm in 100 log-spaced bins) was recorded every 30 s, enabling to follow flocculation in time. Sediment and algae sample were added to a mixing jar and stirred by a paddle at the lowest speed possible to keep particles suspended. The

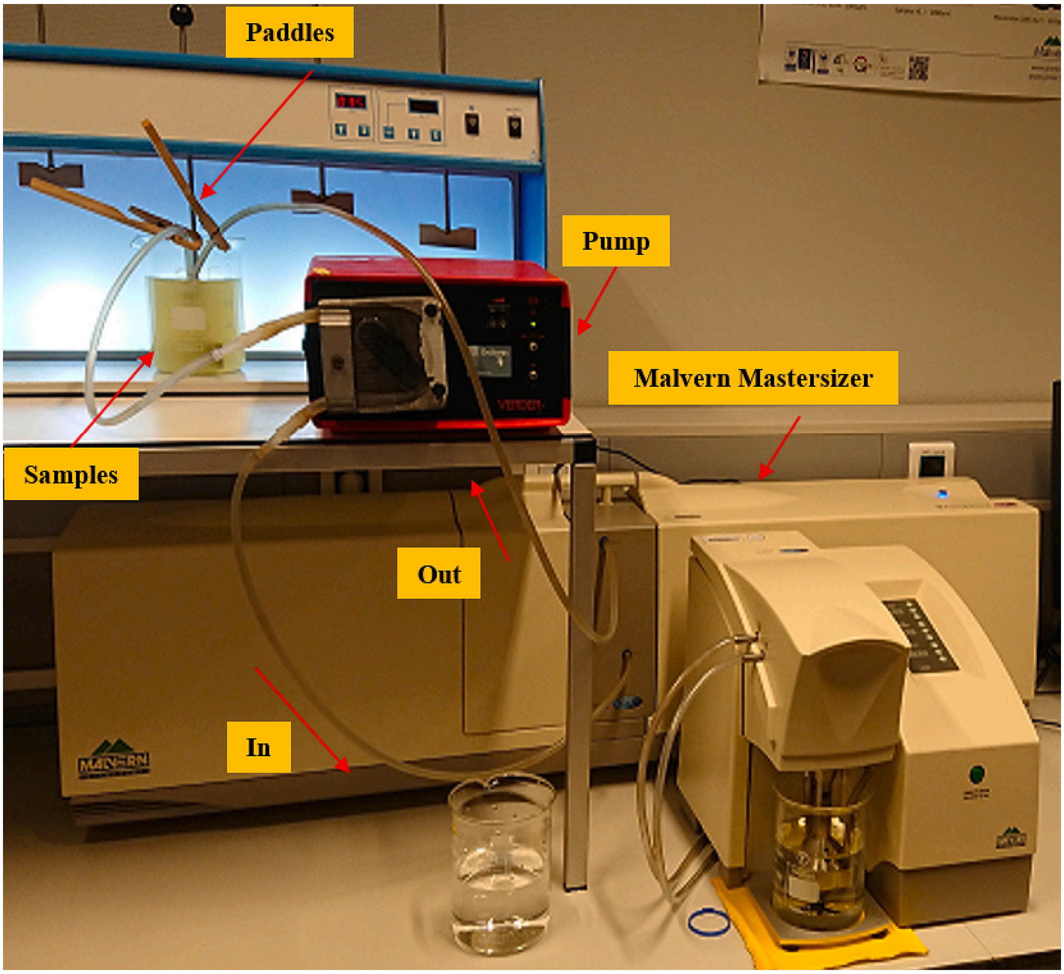

Fig. 1. The Static Light Scattering device (MasterSizer) with connected pump and jar used for the flocculation experiments.

samples were pumped into instrument and back to mixing jar continuously through two pipes of diameter 6 mm. The mixing jar was 0.125 m wide and 0.185 m high.

In previous studies, the shear rate in the jar was evaluated using$logG = -0.849 + 1.5\,log\,(60RPS)$, where RPS is the stirring rotations per second (de Lucas Pardo, 2014; Mietta, 2010). In the experiments, flocculation is not only affected by the shear forces in the jar, but also affected by the shear rate in the tubes. The shear rates in the tubes are estimated using:

$$\boldsymbol{G = 4Q/\pi r^3} \tag{1}$$

where Q is the discharge ($m^3\ s^{-1}$) and r is the radius of the pipe (3 mm).

The pump speed was kept to 10 rotations per minute (RPM). This limit was imposed by the fact that the pump would sometimes stop working when the pump speed was lower than 10 RPM. 10 RPM corresponds roughly to 90 s^{-1}.

The speed of the paddle was adjusted to 30 RPM to keep the samples in suspension. Although there were two different shear rate distribution in jar and tubes, it has been shown that the shear rate distribution in the jar affects the shape of the floc size distribution but not the mean floc size (Bouyer et al., 2004).

The minimum shear rate used in the experiments was therefore 90 s^{-1} which is usually higher than in situ observations, implying that smaller flocs could be created in laboratory experiments compared to in situ. Another difference is the residence time in the jar: as the particles are kept in suspension, their collision probability and frequency will be higher than in-situ.

2.3. Settling columns

The settling column measurements were performed in 5 cylindrical glass columns (1 l). Each cylinder contained different concentrations of sediment and fresh algae samples in demi-water, as given in Table 1. In that table, CC stands for "Chlorophyll a Concentration" that is used as a proxy to estimate the amount of microalgae in the water. The ratio CC/SSC was introduced in Deng et al. (2019) and will be used in the discussion. The samples were mixed thoroughly by inverting the columns ten times within 10 s to ensure an adequate mixing and prevent their significant breakage (Shakeel et al., 2020). The columns were then left to stand for about 24 h. The water/suspension interface was recorded by camera (canon 70D) every 2 min.

3. Results and discussions

3.1. Sediment, EPS and algae flocs

In a first set of experiments, the time-dependence of sediment, EPS and algae flocs were investigated separately in demi-water (Fig. 2).

The sediment flocculation over a period of 120 min was limited as the D_{50} varied from 8 to 10.5 μm. The PSD at start was a log-normal distribution and after 120 min a second peak appeared, one at about 6 μm (about the D_{50} size of the primary particles) and the other at 20 μm. This indicated that sediment flocculation in demi-water is very slow and incomplete.

The SLS measurement of EPS is shown to be unreliable. This is due to the fact that EPS is composed of flexible elongated polymeric chains with a very large aspect ratio. These features lead to multiple peaks in the PSD, as the instrument is calibrated for the measurement of spherical particles.

The D_{50} of the 1% algae suspension fluctuated between 70 and 120 μm as the sample remained very polydisperse. The D_{50} of the initial 10% algae sample is about 40 μm, which is much larger than the D_{50} of the original sediment particles. This D_{50} is however smaller than the D50 of the 1% algae suspension. From the PSD's, it can be observed that at 10% algae there is a large relative volume amount in the range 10–50 μm which is much smaller in the 1% algae PSD. The reason for this is at this stage unclear, but we make the hypothesis that this could be attributed to (1) the different conformation adopted by the algae strains at high concentrations and/or (2) a laser diffraction artefact due to the fact that the particles are anisotropic and have a not well-defined refractive index. This last effect could be enhanced at high particle concentration.

Algae cells are shown to aggregate over time and form larger algae flocs, even in demi-water. It should be noted that the shear rate used in the measurement is usually higher than the ones experienced by algae in their natural environment. The algae flocculate faster than sediment particles, reaching a maximum D_{50} value within 30 min. The change in the small and large peaks size indicate that the small algae cells tend to aggregate and form large flocs of the order of 70–100 μm in size. The rather unimodal PSDs found at the end of the experiment seems to indicate that these flocs are rather spherical in shape. This sphericity is due to the constant shear experienced by these flocs during the measurements (Shakeel et al., 2020). This type of shear is not often encountered in-situ where long-chain algae colonies are usually observed.

3.2. Salinity and EPS effects

3.2.1. Evolution of sediment flocculation with salinity

The changes in the floc size and particle size distribution as a function of time for sediment flocculation with different salts and salt concentration are shown in Fig. 3.

The floc size increased slowly with NaCl (ranging from 5.3 μm to 8.5 μm) at 0.1 mol l^{-1}, and higher NaCl concentrations led to higher flocculation rates and larger flocs. For the highest concentration used (0.3 mol l^{-1}) the D_{50} varied from 6.3 μm to 11.2 μm.

The $MgCl_2$ led to faster flocculation than NaCl. The results show that the sediment particle size reached steady-state within 10 min. Contrary to expectation, and as is usually observed, see Mietta et al. (2009b), higher $MgCl_2$ concentrations led to smaller steady-state sizes. A similar effect is observed with the other divalent salt used ($CaCl_2$). This peculiar behavior indicates that flocculation with divalent salts is different from the classical DLVO theory. Most probably chemical bindings are involved, but further analysis would be required to confirm this.

For flocculation with divalent salts, one can observe that the PSDs stays multimodal with time: a tail is observed around 1 μm, a main peak at about 10 μm and a small fraction of the flocs has a size of about 100 μm. There can be two explanations for the apparition of the peak at 100 μm: (1) there are indeed such large flocs forming in the presence of divalent salt or (2) some air bubbles became entrapped in the measurement chamber. Unfortunately, it was not possible to repeat the measurements to confirm or disprove these hypotheses.

Although the concentration of $CaCl_2$ was lower than $MgCl_2$, the flocs grew faster than with $MgCl_2$ leading to larger mean sizes of flocs. This indicates that besides cation concentration, the type of cation also influences the sediment flocculation. This is another indication that the

Table 1
Composition of the settling columns.

Group name	Suspended sediment concentration (SSC)	Algae concentration			CC/SSC
	g l^{-1}	Volume	cells l^{-1}	(Estimated CC) μg l^{-1}	μg g^{-1}
A	0.7	1%	5×10^4	1.25	1.8
B	0.7	10%	5×10^5	12.5	17.9
C	0.7	–	–	–	–
D	–	1%	5×10^4	1.25	–
E	–	10%	5×10^5	12.5	–

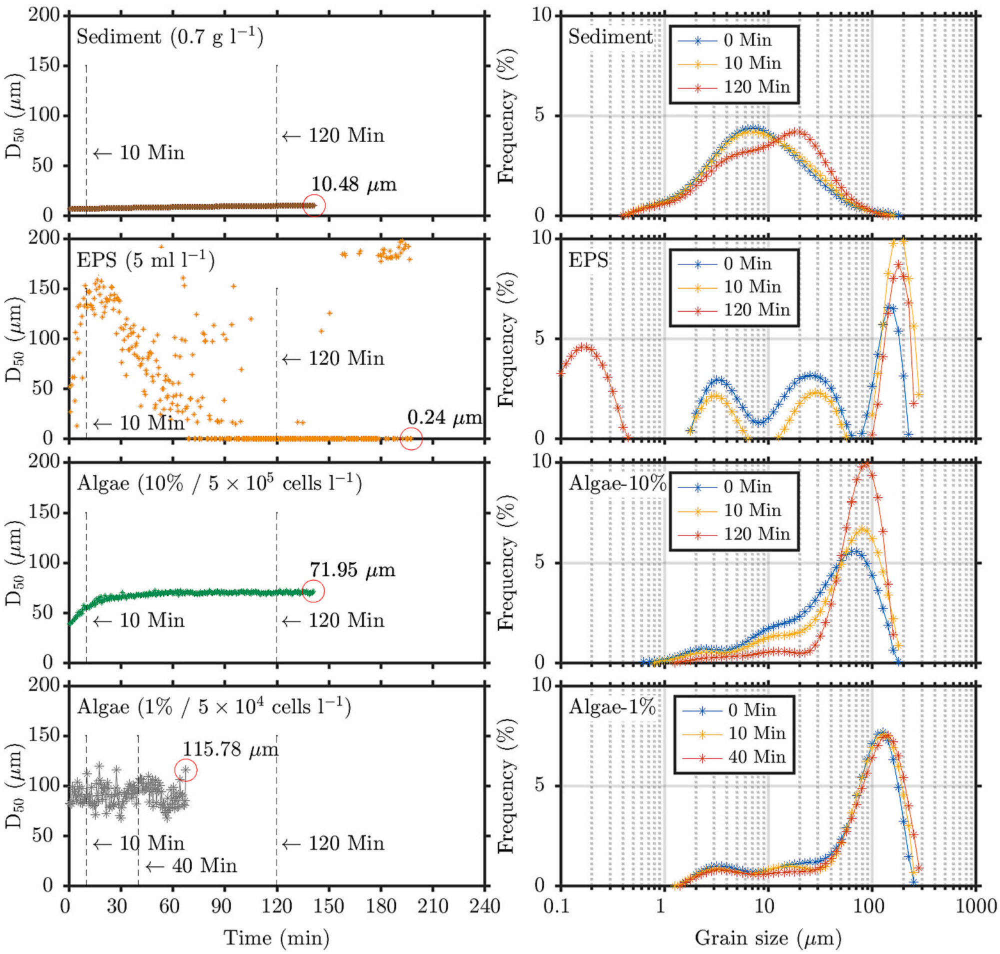

Fig. 2. Floc size time evolution and distribution of sediment suspensions, EPS and living algae. The concentration of sediment was 0.7 g l^{-1}, EPS 5 ml l^{-1}, algae 10% and algae 1%, all of them in demi-water.

flocculation occurring with divalent salts does not obey the classical DLVO theory. Following DLVO theory, for a same concentration and same valence of ions, flocculation should be the same.

3.2.2. Evolution of sediment flocculation with EPS and salinity

In this section, the combined effect of EPS and salinity on flocculation is investigated. A fixed concentration of 5 ml l^{-1} EPS was added to all samples and the salt type and concentration was varied. The results are shown in Fig. 4.

The flocculation of sediment with EPS and salt led to larger steady-state floc sizes than in the experiments with only added salt. In the presence of sediment, for the shear rate used, EPS binds to the sediment to create flocs that are in good approximation spherical as can be deduced from the rather unimodal PSDs found.

The trends as function of salt type and concentration were similar to the ones observed in Fig. 3, when no EPS was added: the flocs growth was slower with NaCl than with $MgCl_2$ and $CaCl_2$. In contrast to what was observed in Fig. 3, the steady-state floc size decreases with increasing salinity for all salts (hence: also with the monovalent salt NaCl). The reason for this behavior is attributed to the decrease in the radius of gyration of the EPS, as the salt ions screen the electrostatic repulsions between the charged groups of the polyelectrolyte. As a consequence, the capture of sediment by EPS becomes less favourable. For the divalent salts, it is found that the time to reach the steady-state floc size is highest for the lowest salinity.

3.2.3. Experiments with sea water

In this section, the results presented in the two previous sections are compared with the results obtained from experiments done in artificial sea water.

Fig. 5 and Fig. 6 show the growth of floc size at different salt conditions. The horizontal bars represent the mean floc size (D_{50}), the ends of the boxes D_{25} and D_{75} and the ends of the vertical lines are the D_{10} and D_{90} of PSDs. The results displayed with EPS at different salt types and concentrations are the same as the ones presented in the three previous sections.

Note that in demi-water, the floc size reached its steady-state only after 2 h (Fig. 2), while for all the other measurements it was within 30 min (Fig. 3 and Fig. 4). Therefore all PSDs are shown at 30 min after the start of the experiments but the one for demi-water are at steady-state.

It is found that the mean floc size at steady-state is largest in the case of experiments done with sea salt. The cationic composition of sea water is roughly 0.5 mol l^{-1} Na^+, 0.05 mol l^{-1} Mg^{2+} and 0.01 mol l^{-1} Ca^{2+}. It would therefore appear, in light of the experiments shown in Figs. 4–6, that, in the presence of EPS, the kinetics of flocculation with sea salt is dependent on the flocculation with Na^+. Indeed, there is a marked difference in particle size between 10 and 30 min after the start of the experiments for both experiments with sea salt and with NaCl (much less with the other ions). On the other hand, the suspension with EPS in sea water has a wide spread in size.

Fig. 7 shows the different initial flocculation rates obtained at different experimental conditions. The flocculation rate is defined as the slope of the D_{50} size as function of time in the time interval [0, 10 min]. Flocculation with $CaCl_2$ is found to be the fastest, for any concentration when no EPS is present. The flocculation rate with sea salt is second highest when no EPS is present. In the presence of EPS, on the other

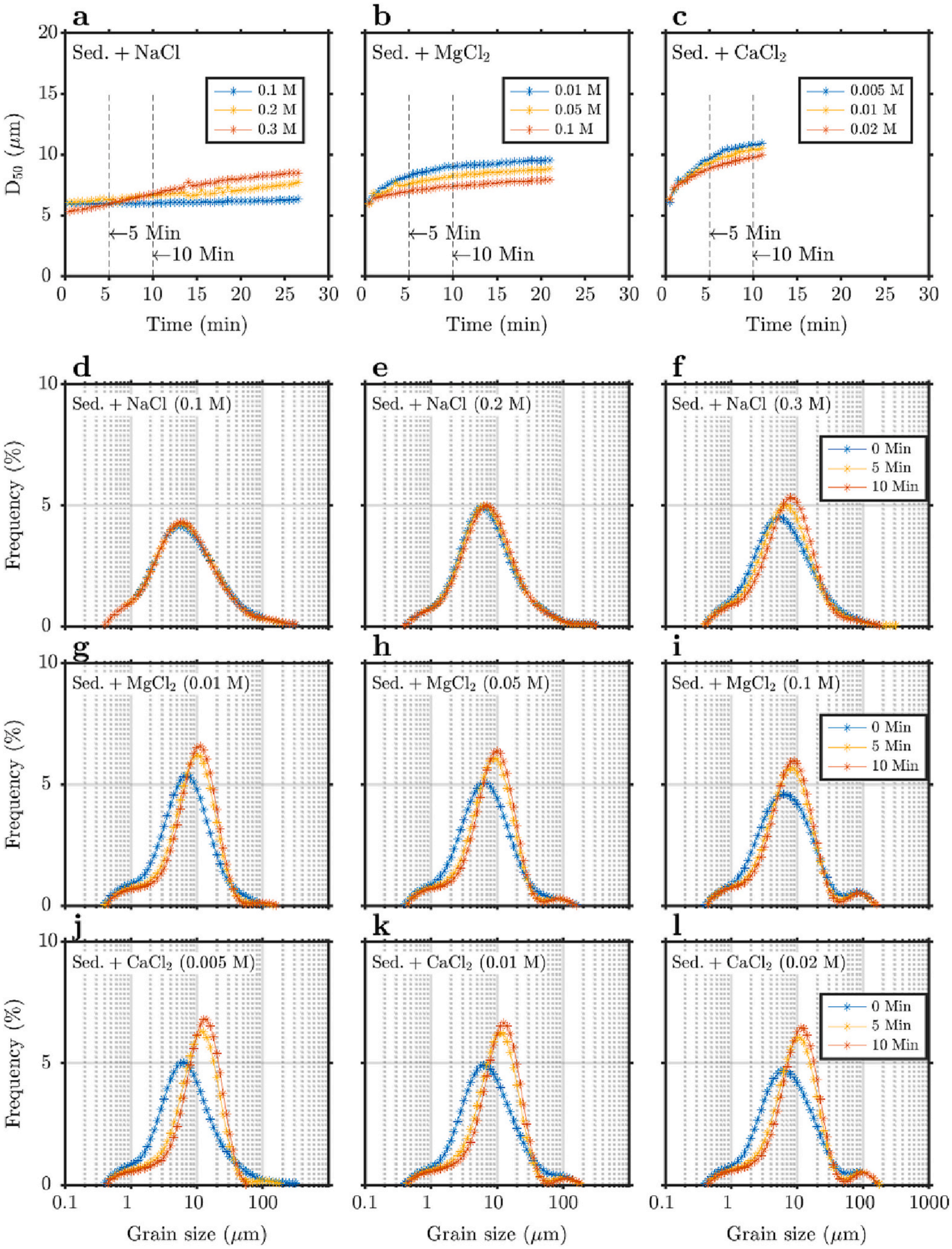

Fig. 3. Effect of NaCl, $MgCl_2$ and $CaCl_2$ on floc size time evolution and distribution of sediment suspensions.

hand, the flocculation rate with monovalent and divalent salts follows the trends discussed in section 3.2.2 and flocculation with sea salt is found the highest.

3.3. *Experiments with living algae*

3.3.1. *Flocculation with living algae in demi-water*

In the estuarine environment, the sediment PSD is affected by the ratio of algae concentration and sediment concentration (CC/SSC), see Deng et al. (2021, 2019). The symbol CC stands for Chlorophyll Concentration (which is linearly proportional to algae concentration) and SSC for Suspended Sediment Concentration. To remain as close as possible to in-situ experiments, flocculation was triggered in settling columns (see section 2.3). The measurements performed in these settling columns were used to discuss the effects of different living algae concentrations on flocculation/settling.

Fig. 8 shows that the sediment suspension is hardly settling in demi-water, and the water kept turbid even after 24 h. This result is in line with the results reported in section 3.2.2, where it was discussed that the depletion of ions in the water led to larger repulsion between particles. The living algae seemed to be settling little due to the fact that they tend to move to the surface for respiration and photosynthesis. In suspensions made of both sediment and algae a fast settling of particles is observed. Most of the sediment was settled after 5 h with 10% living algae, and the sediment was settled after 24 h with 1% living algae, though the living algae concentration (and hence CC/SSC) was very low.

SLS experiments were performed on four samples composed of different sediment and algae concentration (Table 2 and Fig. 9). For two of these experiments (group 1 and group 2) the settling column experiments are displayed in Fig. 8. The sample corresponding to group 1 has the lowest CC/SSC ratio (of about 1.8 μg g^{-1}) and the lowest flocculation rate as the particle size did not reach its maximum value after 30 min.

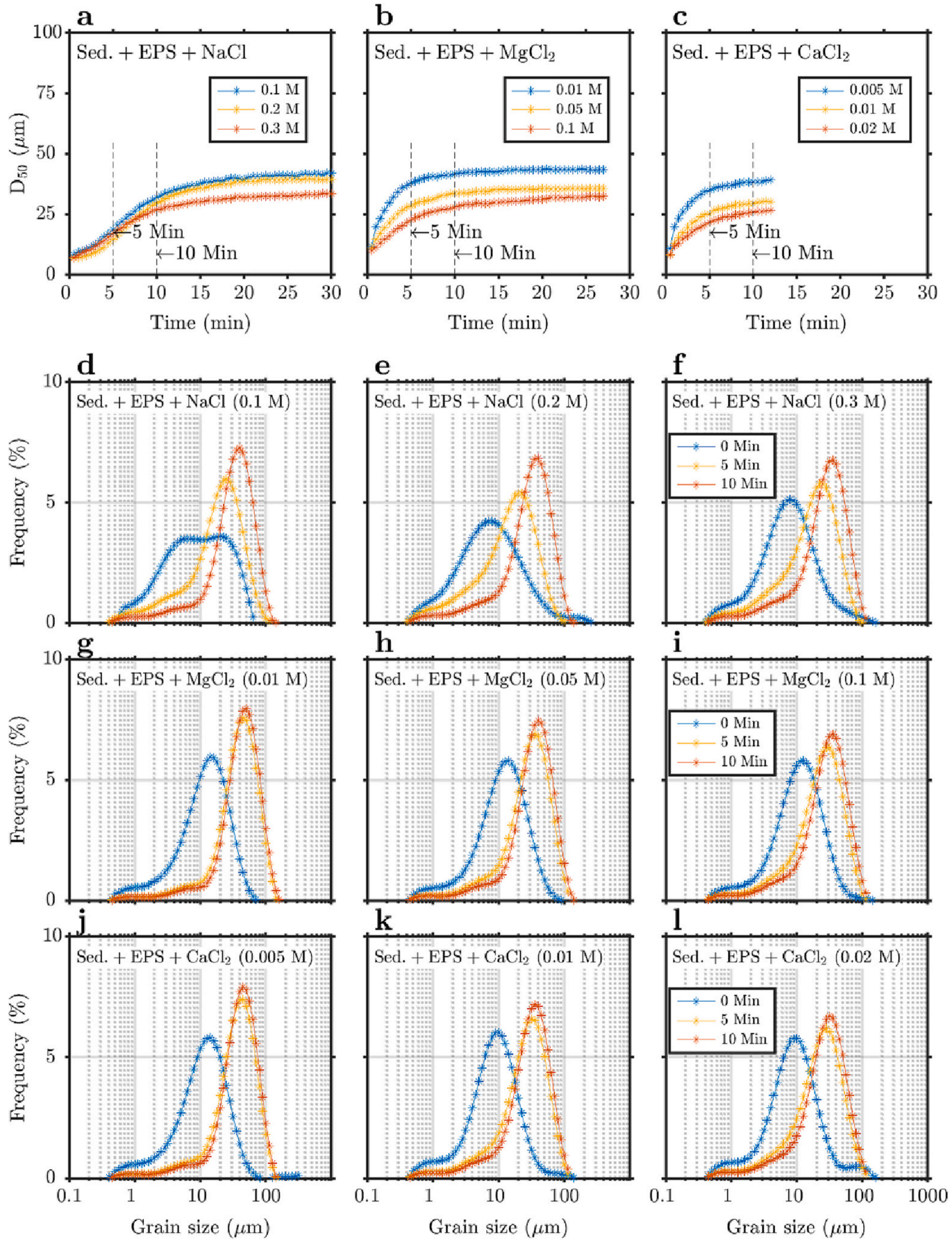

Fig. 4. Effect of EPS combined with NaCl, $MgCl_2$ and $CaCl_2$ on floc size time evolution and distribution of sediment suspensions.

This is in agreement with what was observed in the settling column. For group 2, of higher CC/SSC ratio (CC/SSC = 17.9 μg g^{-1}), the flocculation rate is much higher than for group 1 and the D_{50} reaches a value of 71 μm within 10 min. Keeping the same CC/SSC ratio, but decreasing both CC and SSC 10 times (group 3) both flocculation rate and maximum D_{50} value were also higher than group 1, but less than for group 2. For group 4, of highest CC/SSC ratio (having the same CC as group 2 but much less suspended sediment), the maximum D_{50} value was higher than any other group but the flocculation rate was smaller than the one of group 2. The evolution of PSDs (sediment + algae) show different flocculation processes:

At high CC (10% algae), a substantial increase in D_{50} is observed as a consequence of flocculation. The flocculation rate is highest for the sample with lowest CC/SSC ratio (group 2) but the largest floc size is obtained with the sample with highest CC/SSC ratio (group 4).

At low CC (1% algae), two behaviors are observed:

(1) at low CC/SSC (group 1), not much flocculation is found, the D_{50} remains low. This is in line with the results found with 1% algae suspension (see Fig. 2).
(2) at high CC/SSC (group 3), a significant increase in D_{50} is observed, which is the result of flocculation between algae and sediment.

These results confirm that a relevant parameter for studying flocculation is the CC/SSC ratio: when the CC/SSC ratio is high, large flocs are formed and vice-versa (irrespective of CC or SSC concentrations). The flocculation rate (group 2 vs group 4) is however highest for the sample with lowest CC/SSC (group 2). There are two reasons for this behavior: 1) algae do not aggregate at low concentration (see Fig. 2); 2) high SSC lead to high collision frequency, so the flocculation is fastest when SSC is high. The SSC of group 2 is 10 times larger than the SSC of group 4.

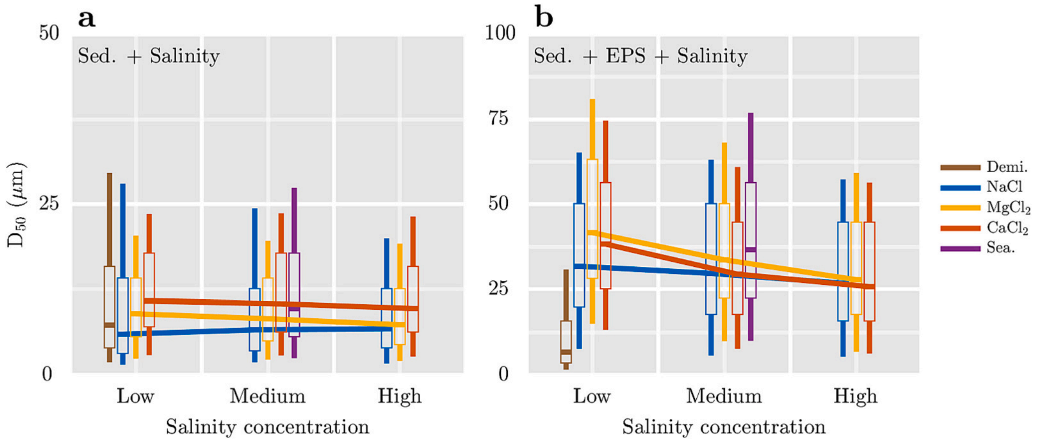

Fig. 5. Floc size of sediment and sediment with EPS suspensions on the effect of salinity 10 min after the start of the experiments. Note that the data for demi-water is plotted in the "low" column.

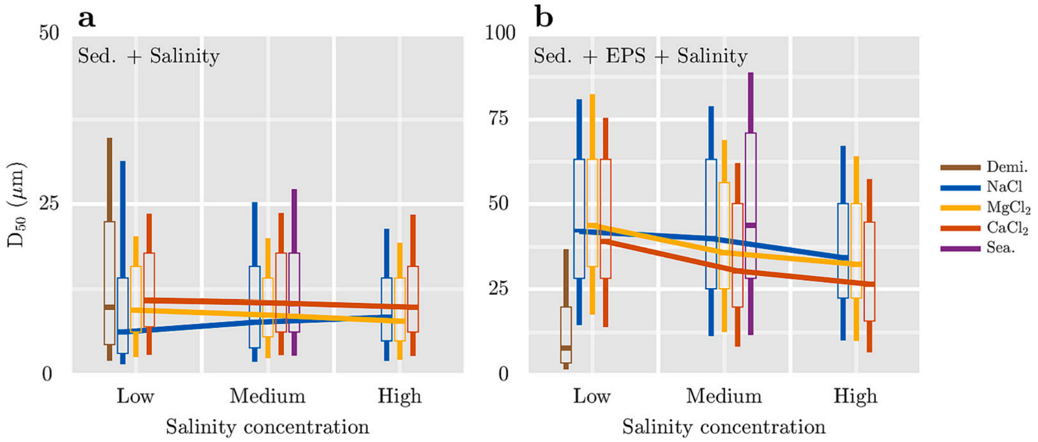

Fig. 6. Floc size of sediment and sediment with EPS suspensions on the effect of salinity 30 min after the start of the experiments. Note that the data for demi-water is plotted in the "low" column.

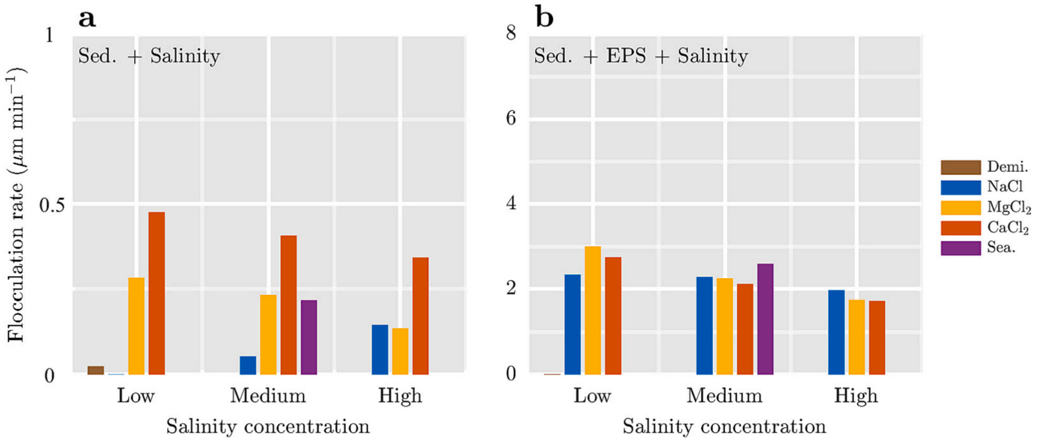

Fig. 7. Flocculation rate of sediment and sediment with EPS suspensions at different salinities 10 min after the start of the experiments. Note that the data for demi-water is plotted in the "low" column.

The results are summarized in Fig. 10.

It is found that the steady-state floc size of group 1 (1.8 μg g^{-1} CC/SSC) is smaller than all the others, confirming that the maximum D$_{50}$ value was limited by low CC/SSC despite the high SSC. The D90 of group 2 (17.9 μg g^{-1} CC/SSC) and group 4 (178.6 μg g^{-1} CC/SSC) were larger than group 1 (1.8 μg g^{-1} CC/SSC) and group 3 (17.9 μg g^{-1} CC/SSC), indicating that higher concentrations of living algae lead to larger particle sizes.

The flocculation rate of group 2 (17.9 μg g^{-1} CC/SSC) was much higher than the other groups. The flocculation rate of group 4 (178.6 μg

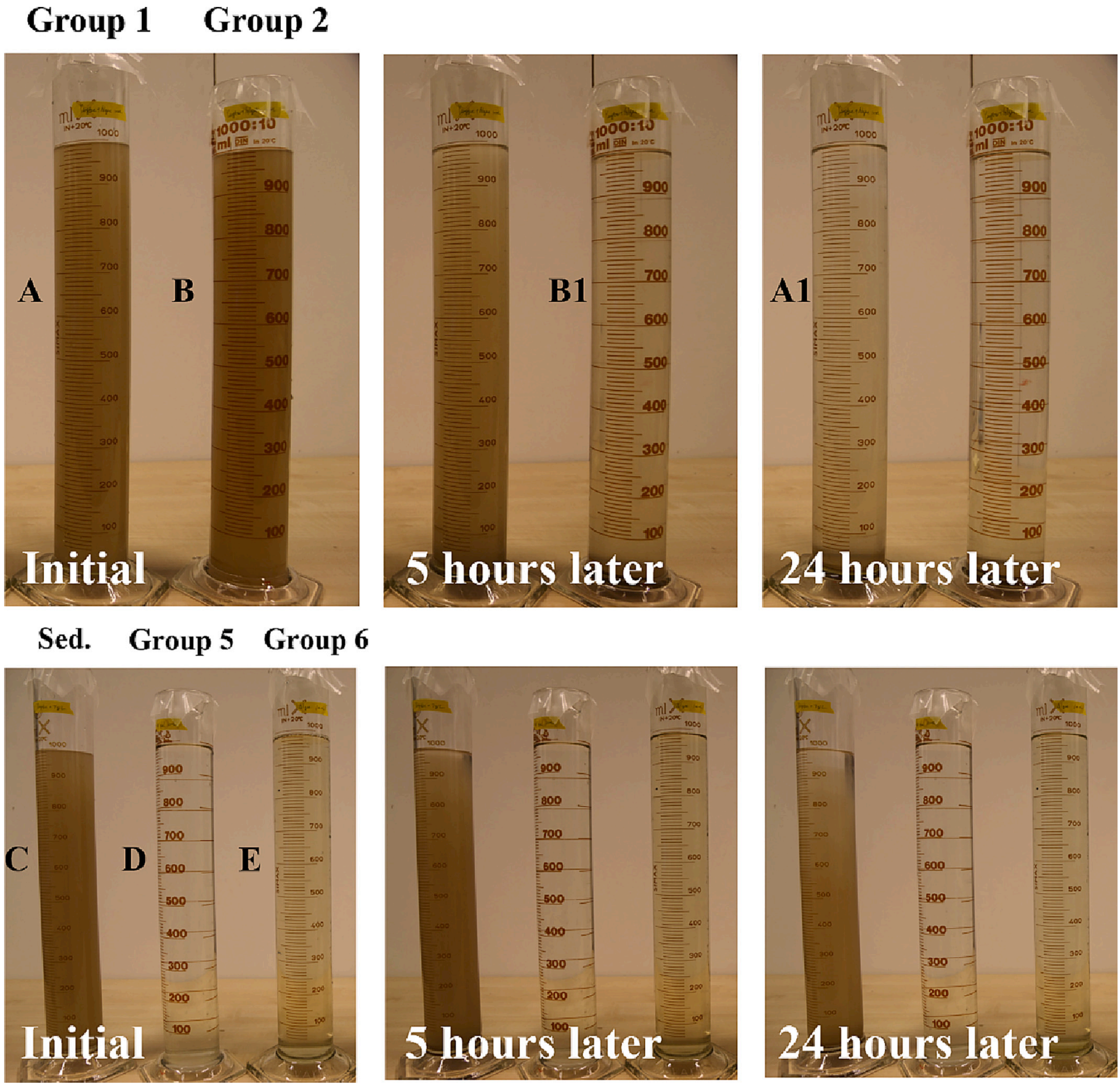

Fig. 8. Settling column measurements of suspensions with different sediment and algae concentrations in demi-water.
(A.0.7 g l^{-1} sediment with 1% living algae (Group 1), B. 0.7 g l^{-1} sediment with 10% living algae (Group 2), B1. 0.7 g l^{-1} sediment with 10% living algae 5 h later, A1. 0.7 g l^{-1} sediment with 1% living algae 24 h later, C. 0.7 g l^{-1} sediment, D. 1% living algae (group 5), E. 10% living algae (group 6)). The groups refer to Table 1 and Table 2.

g^{-1} CC/SSC) is higher than group 3 (17.9 μg g^{-1} CC/SSC). The flocculation rate of group 1 (1.8 μg g^{-1} CC/SSC) was lowest. The flocculation rate was higher with higher particles concentration (SSC), except for group 1 (1.8 μg g^{-1} CC/SSC), as said above. This highlight the fact that algae is the trigger for flocculation and that below a certain algae concentration neither algae cells with themselves, nor algae cells and sediment will flocculate.

Overall, it can be concluded that the maximum D_{50} value is controlled by the CC/SSC, while the flocculation rate is controlled by SSC (above a certain CC threshold).

3.3.2. *Flocculation with living algae in salt water*

Fig. 11 shows the time evolution of flocs with different SSC and different algae concentrations (CC/SSC) in NaCl (0.5 M, equal to approximately seawater NaCl concentration). In the absence of algae, the highest SSC leads to the largest floc sizes with NaCl (purple lines). In contrast to demi-water experiments, the high salinity ensures that the surface charge of the sediment particles is fully screened, making flocculation possible (Mietta, 2010). In the presence of algae, it is found, as was discussed in the previous subsection, that the largest flocs are obtained for the suspensions with the highest CC/SSC (implying, for a given CC/algae concentration for samples with the lowest SSC). The flocculation rate, on the other hand is proportional to SSC: the highest the SSC, the fastest the flocculation.

The highest floc sizes are observed 30 min after the start of the experiment, when a maximum D_{50} value of about 60–80 μm is reached.

Table 2
Set-ups of the SLS measurements.

Group name	Suspended sediment concentration (SSC)	Algae concentration			CC/SSC
	$g\ l^{-1}$	Volume	cells l^{-1}	(Estimated CC) $\mu g\ l^{-1}$	$\mu g\ g^{-1}$
Group 1	0.7	1%	5×10^4	1.25	1.8
Group 2	0.7	10%	5×10^5	12.5	17.9
Group 3	0.07	1%	5×10^4	1.25	17.9
Group 4	0.07	10%	5×10^5	12.5	178.6
Group 5	–	1%	5×10^4	1.25	–
Group 6	–	10%	5×10^5	12.5	–

Higher algae concentrations do not lead to higher floc sizes and the floc size even decreases with algae concentration at longer times. The reason is most probably the reconformation of algae chains under shear, the algae chains being extended at the beginning of the experiments while they tend to bind together while times passes and the concentration is increased. This effect is also visible on the experiments presented in Fig. 12. When the algae concentration reached to 20% (10^6 cells l^{-1}), the flocs had nearly a same size for two different SSC (green lines), which indicates that the flocs are algae-dominated when the algae concentration is high.

Experiments realized in artificial sea water are shown in Fig. 12. In Fig. 12a, the time evolution of the D_{50} of different suspensions is shown, for a sediment concentration of 0.7 g l^{-1}. In contrast to flocculation with NaCl, flocculation with sea salt produces smaller flocs. One reason could be that due to the presence of divalent ions in sea salt, flocs get a denser structure than with monovalent NaCl. Additional measurements with algae in presence of divalent salt should help to confirm this. The D_{50} of the suspension with 30% algae shows that over time the mean particle size decreases at long times. This phenomenon is attributed to the shape of the flocs at this high algae concentration. The anisotropy of flocs is discussed in more detail in the Supplementary Material file.

3.3.3. *Comparisons between EPS and living algae influences on flocculation*

The effects of EPS and living algae are usually grouped under the generic term of "organic matter effects". To distinguish between the two, several samples made in demi-water and sea water were compared. These samples were made of: (a) sediment (no added algae nor EPS), (b) algae (no added sediment nor EPS), (c) sediment with EPS and (d) sediment with algae. The results are shown in Fig. 13.

In demi-water, the sediment and sediment with EPS suspensions (a and c) display no significant flocculation, while the sediment with algae and the pure algae suspensions (b and d) display similar flocculation kinetics and floc size. Note that the flocs formed by aggregation of algae cells reach a constant steady-state value whereas flocs formed by combining sediment and algae display a long-term time-dependent floc size. In sea water, all samples displayed flocculation except the pure sediment samples. Although the floc size of sediment did not significantly change from demi-water to sea water, the peak of sediment PSD in sea water was shaper than that in demi-water, which could indicate a slight flocculation effect in sea water. The fact that flocculation with EPS improved in seawater is consistent with section 3.2. The PSD of the samples with algae (green curves) shows that these samples are multimodal indicating the presence of some single algae cells or short-chains.

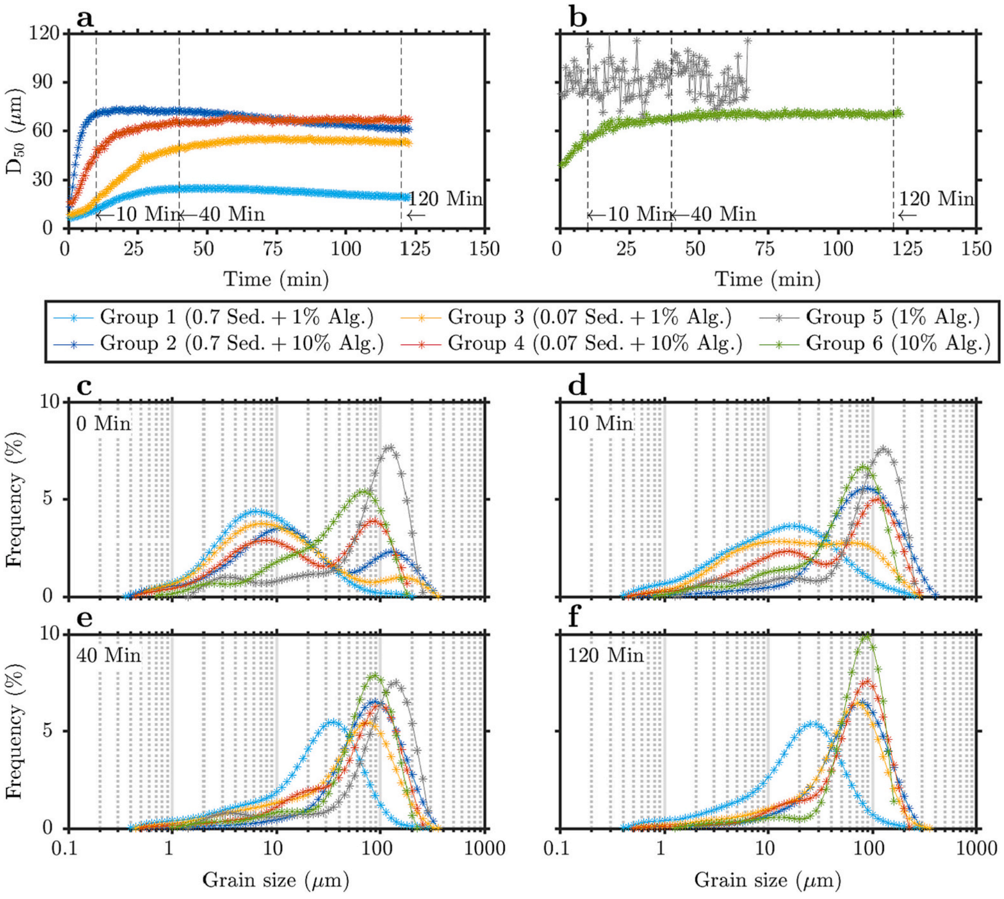

Fig. 9. Floc size time evolution and distribution of suspensions with different sediment and algae concentrations in demi-water.

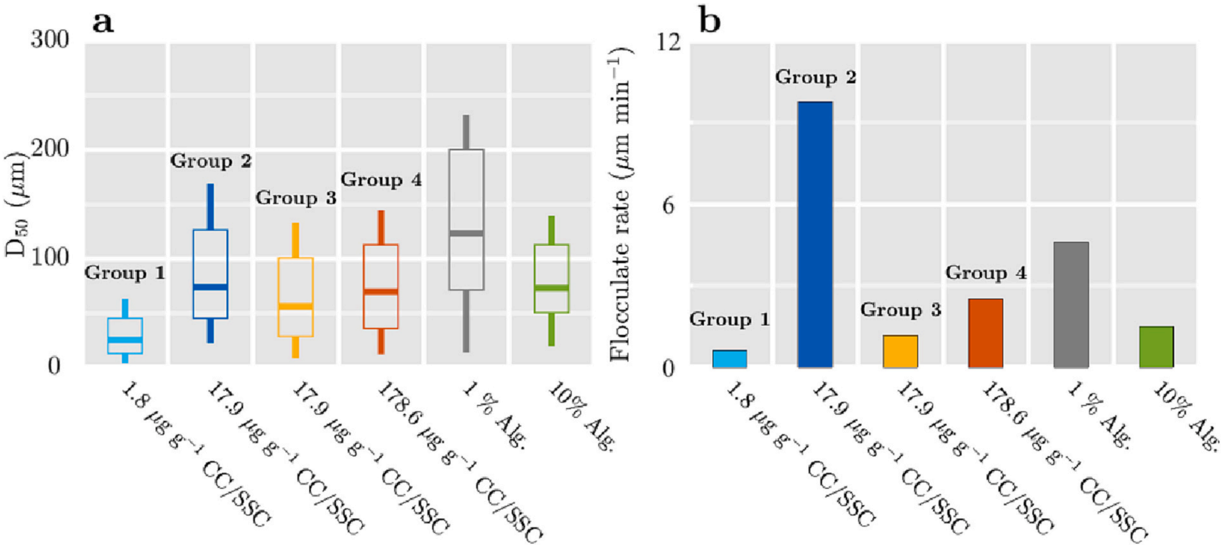

Fig. 10. Mean floc size (D_{50}) and flocculation rate of different suspensions in demi-water.

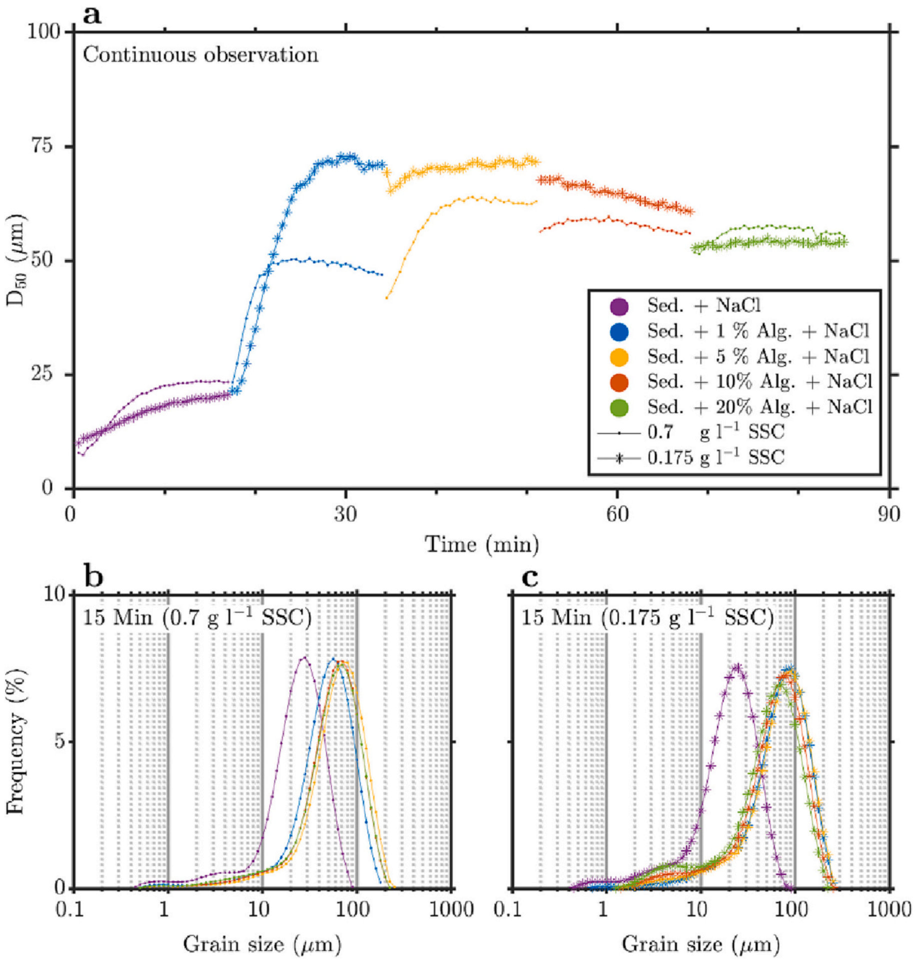

Fig. 11. Floc size time evolution and distribution of suspensions having different sediment and algae concentrations in 0.5 M NaCl.

The small peaks disappear when the algae is mixed with sediment particles (red curves) and the main peak moves from 25 µm to 100 µm over time, which indicates that flocculation occurs. The D_{50} in sea water is much larger than the one in demi-water for algae suspensions and larger for the sediment/algae suspensions, which indicates that sea water is especially beneficial to floc formation for algae cells.

4. Conclusions

The laboratory experiments presented in this article confirmed that

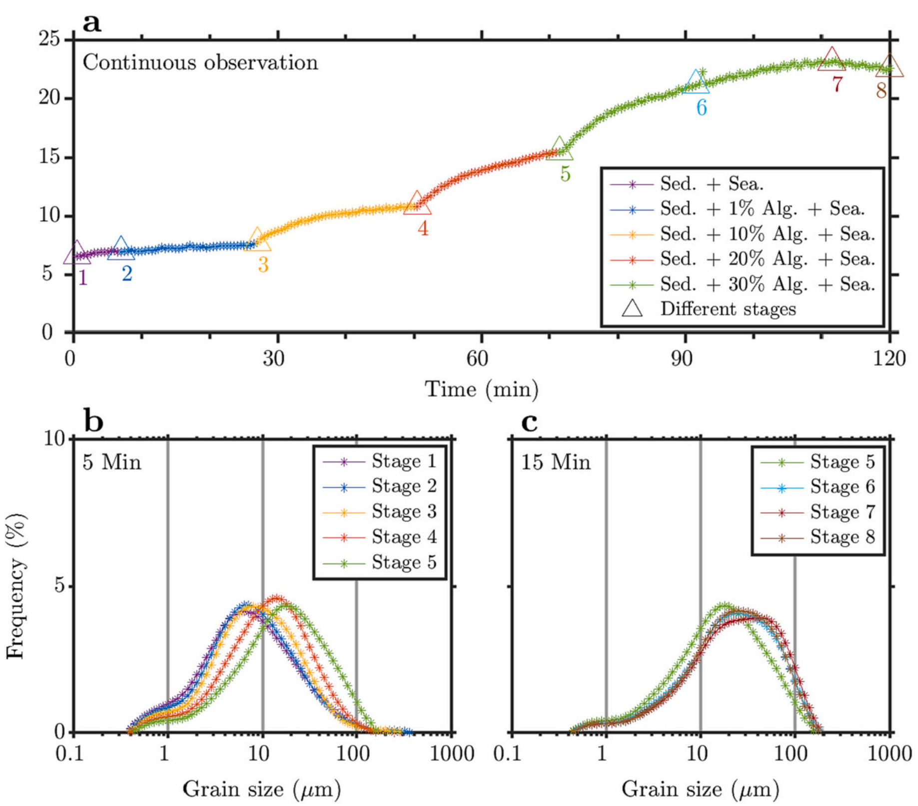

Fig. 12. Floc size time evolution and distribution of suspensions having different sediment and algae concentrations in sea water. Figures b and c represent the PSD of suspensions given in a.

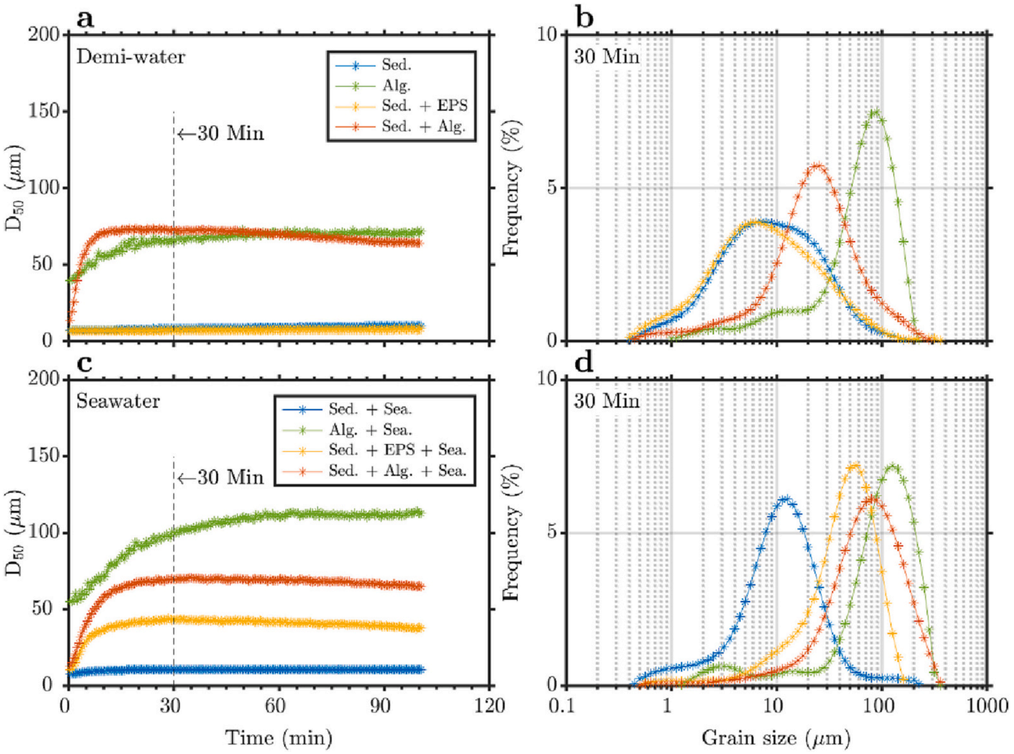

Fig. 13. Floc size time evolution and distribution of sediment (0.7 g l^{-1}), sediment (0.7 g l^{-1}) with EPS (5 ml l^{-1}) and sediment (0.7 g l^{-1}) with algae (10%, yielding CC/SSC =17.9 μg l^{-1}) suspensions in demi-water (top panel) and sea water (bottom panel).

the production of large flocs is correlated with the presence of EPS and living microalgae. Usually no distinction is made when organic matter effects are discussed. We show that that while EPS, being a polyelectrolyte, acts as a traditional flocculant, microalgae can form large flocs as the microalgae cells aggregate with each other and also bind to sediment. This is in line with what has been found by analyzing in-situ monitoring data (Deng et al., 2021; Deng et al., 2019), where the ratio CC/SSC was introduced. We confirm with the laboratory study that the ratio algae concentration (which is linearly proportional to CC) to sediment concentration (SSC) is driving floc properties. A continuous addition of algae to a suspension of sediment + algae that has reached a steady-state size results in a new flocculation. Flocs made of relative large amounts of algae are elongated. It should be noted that the algae effect is limited by sediment concentration, as high sediment concentration restricts sediment flocculation, which can also be related to the CC/SSC ratio. The flocculation rate is shown to be of the order of a few μm/min and a steady-state floc size (at constant shear rate) is obtained within 30 min for all experiments realized with organic matter.

In this study, the shear rate used was usually higher than the one encountered in natural environment conditions. This could lead to the fact that the effects of salinity (sediment + salt experiments) were smaller than the ones reported in other laboratory measurements (Mietta et al., 2009a), where different shear rates were used. The combination of cations with organic matter (EPS or algae) requires further investigation. We showed in particular that the combined action of salt and EPS does not follow the classical DLVO theory as EPS is also undergoing steric interactions, but that the presence of cations is required to promote flocculation.

Overall, salinity, EPS and living algae are parameters that all present in natural aquatic environments and their combined action makes flocculation processes complex. Further study is required to analyze the precise role of each parameter in combination with the others.

Declaration of Competing Interest

The authors declared that they have no conflicts of interest to this work.

Data availability

Due to the confidentiality agreement of relevant projects, more data that support the findings of this study are available from the corresponding author, [Q.], upon reasonable request.

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