

Final Report

Brilliant Marine Research Idea 2024

This report should be submitted no later than 28 February 2025 via filantropie@vliz.be and consists of the following documents:

- A final report listing the work done and the problems encountered. This report will be made available online. If any of the tasks has not been completely finished, the report should clearly mention this, including a short explanation. max. 5 pages
- An overview of all expenditures including invoices.
- A set of five pictures (low resolution in this document). The five high resolution pictures should be delivered to VLIZ by email to karen.rappe@vliz.be. Pictures should be free from use - to upload on the VLIZ website and to use in VLIZ communications.

Keep in mind that VLIZ should be mentioned in the acknowledgements of publications following the results of this Brilliant Marine Research Idea.

1. General information

Title of the idea	Can mussel feces reduce turbidity in coastal areas?
Name PhD student	Luz Amadei Martínez
Name supervisor	Koen Sabbe & Wim Vyverman
Flemish University or University College	Ghent University

2. Brilliant Marine Research Idea – Report about the activities

Abstract

Installing offshore structures, such as wind farms and floating solar panels, introduces large amounts of artificial hard substrate into previously soft sediment-dominated areas. These substrates are rapidly colonized by filter-feeding organisms, such as mussels, which have high filtration capacities and extract significant quantities of phytoplankton and suspended particulate matter (SPM). The mussels then egest undigested material as biodeposits (fecal pellets or pseudofeces). In recent decades, the Belgian part of the North Sea has experienced significant changes in SPM dynamics that are poorly understood. Our study hypothesizes that biodeposits from blue mussels (*Mytilus edulis*) contribute to reduced turbidity by promoting particle aggregation and altering floc structure, density, and settling velocity. To test this, we conducted a laboratory experiment to assess the impact of biodeposits on SPM flocculation. Using a custom-made flocculation chamber, we monitored turbidity and particle size distribution in two treatments: (1) kaolinite and seawater, and (2) biodeposits, kaolinite, and seawater. Initially, both treatments were subjected to a high turbulent shear rate (75 s^{-1}) to homogenize the mixture, followed by a lower shear rate (20 s^{-1}) for 120 minutes to promote aggregation. The results indicate that flocs formed from kaolinite combined with biodeposits were larger than those formed by kaolinite alone and that the median floc size increased more rapidly in the presence of biodeposits. However, turbidity decreased at a similar rate for both treatments during the flocculation experiments. Overall, our study demonstrates that biodeposits released by mussels have the potential to significantly enhance sediment aggregation, thereby influencing the dynamics of SPM in areas adjacent to offshore structures.

Intro

As awareness of climate change and its impact on the global economy increases, there is an increasing shift toward sustainable energy production that is fuelling the expansion of the offshore energy industry (Voet et al., 2022). Offshore installations such as wind farms and floating solar panels have the potential to alter marine ecosystems, notably through the introduction of large quantities of artificial hard substrate into areas that were previously dominated by soft sediments (Rezaei et al., 2023; Mavraki et al., 2020). These substrates quickly become colonised by filter-feeding bivalves and other invertebrates (Lindeboom et al., 2011), which filter large volumes of water and remove phytoplankton and suspended particulate matter (SPM), releasing them as biodeposits (Giles & Pilditch, 2004; Cranford et al., 2011).

In recent decades, the Belgian part of the North Sea (BPNS) has experienced notable, yet not fully understood, changes in SPM dynamics (Capuzzo et al., 2015). This knowledge gap hampers our ability to accurately predict SPM transport—a vital process for managing shipping channels, harbours, water quality, and primary production. Particle transport is governed by a cycle that includes suspension, flocculation, settling, deposition, erosion, and resuspension (Eisma, 1993). In particular, the flocculation process is crucial because it determines the size, structure, and density of the suspended particles (Lai et al., 2018; Chassagne et al., 2020).

Previous research has largely concentrated on the ability of filter feeders to sequester particles from the water column and their role in nutrient cycling through the production of biodeposits (Cranford et al., 2011; van Broekhoven et al., 2015). However, there is still a significant gap in our understanding of the fate of fecal pellets after excretion and their specific role in flocculation processes. To address these issues, we performed a novel experimental approach designed to quantify the flocculation potential of mussels's biodeposits. We selected the blue mussel, *Mytilus edulis*, as our study species because of its ecological and commercial relevance in the BPNS (Mavraki et al., 2020; Stechele et al., 2022). This study hypothesizes that biodeposits contribute to turbidity reduction by promoting the aggregation of suspended particles, modifying the structure, density, and settling velocity of flocs, and ultimately influencing the overall transport of SPM.

Material & Methods

Live specimens of *Mytilus edulis* were collected offshore of Westdiep Nieuwpoort (Belgium) on 1st July 2024 and transported to the lab. After an acclimation period of 24 hours to the lab conditions, pseudofeces and fecal pellets of 100 individuals of varied length (41.1 ± 9.4 mm, mean \pm SD) were obtained by vacuuming the accumulated material deposited at the bottom of the tank through a tube for 4 days. These collected materials were then stored at -18 °C. The initial objective of this study was to separate fecal pellets from pseudofeces, however, this was not possible in practice, therefore we combined both types of biodeposits for the experiments. Therefore, we ran the experiment for two treatments: (1) kaolinite in seawater ('kaolinite'), and (2) kaolinite combined with fecal pellets, pseudofeces, and seawater ('kaolinite + biodeposits').

To evaluate the flocculation potential of sediment alone versus sediment combined with biodeposits, we conducted flocculation experiments in a climate-controlled room maintained at 18 °C under dark conditions. First, for the treatment 'kaolinite + biodeposits', biodeposits collected over several days were homogenized in seawater. This homogenized mixture was then divided into four equal portions, with three subsamples used for the flocculation experiments and one reserved for chemical analysis. Before the experiment, the biodeposit subsample was mixed with kaolinite (final concentration of 25 mg L⁻¹), and the mixture was vortexed thoroughly. The prepared mixture was then transferred into the flocculation chamber pre-filled with seawater at room temperature, and the experiment was initiated. For the treatment 'kaolinite', the same procedure was followed, but without including the biodeposits.

The flocculation chamber consists of a 7.05 L mixing chamber equipped with a paddle for precise turbulence control, a high-resolution FLIR® Blackfly® Color Camera fitted with a 100x zoom lens, and a red laser light

source for illumination. During the flocculation experiment, both treatments were subjected to a high turbulent shear rate (75 s^{-1}) to homogenize the mixture for 5 minutes, then flocs were allowed to grow and reach the floc size equilibrium, by keeping the same turbulence (shear rate: 20 s^{-1}) for 120 mins. This shear rate was selected because it enhances flocculation and is representative of turbulent environments such as the BPNS (Zhang et al., 2019; Tran & Strom, 2019). To monitor changes in particle size distribution, 200 images were recorded every 5 minutes during the first 30 minutes—when the most rapid changes are expected (Verney et al., 2009)—and then at 40, 60, 80, 100, and 120 minutes. The particle sizes in these images were determined using the ParChar module in Matlab (Markussen et al., 2016). Concurrently, turbidity changes due to flocculation were continuously measured with a Seapoint Turbidity Meter STM-S, an instrument funded by this grant.

At the end of each experiment, the floc settling velocity was determined by pipetting a range of different-sized flocs into a sinking column connected to a camera. The settling column was placed on a separate table and filled with water two hours in advance to ensure minimal turbulence from chamber manipulation. The released flocs were filmed for 10 minutes, allowing for the calculation of settling velocities from the videos. However, due to the extensive time required to process these images, we were unable to complete the analysis within the expected timeframe. The final goal of this experiment was to analyze the physicochemical composition of biodeposits and compare the differences between fecal pellets and pseudofeces. However, this comparison was not conducted due to the presence of only a single treatment, which prevent any comparative analysis. Nevertheless, a subsample of the biodeposits was retained for potential future analysis.

To analyze changes in flocculation, for each particle size distribution (PSD), we calculated the median floc diameter (D_{50}), using the equivalent spherical diameter (ESD) as the floc size. Later, for each experiment, we fitted an asymptotic regression model to describe the flocculation process. From this model, we extracted several key parameters. The modelled diameter D_{am} has the following equation:

$$D_{am} = D_e - (D_e - D_0)e^{-\frac{t}{t_{ef}}}$$

where D_e represents the value of D_{50} at flocculation equilibrium, D_0 denotes the initial value of D_{50} at time $t_0=0$, and t_{ef} stands for the time taken to reach the equilibrium. Here equilibrium refers to the floc size at a steady state, where aggregation and breakage are balanced, keeping the floc size relatively constant.

A mean aggregation speed (V_f) is computed from the parameters of the model, V_f is then given by:

$$V_f = \frac{(D_e - D_0)}{t_{ef}}$$

Results/Conclusions

This study investigates the flocculation behaviour of kaolinite suspensions both in the absence and presence of biodeposits, providing new insights into the role of organic matter in sediment aggregation.

Overall, the particle size distribution for the flocculation experiments for both kaolinite alone and kaolinite combined with biodeposits show a shift toward larger floc sizes over time. However, in the kaolinite + biodeposits treatment at 120 minutes, this shift toward larger aggregates is even more pronounced. For kaolinite alone, the median floc size increases from $11 \mu\text{m}$ initially to $16 \mu\text{m}$ at 120 minutes, whereas in the presence of biodeposits it grows from $13 \mu\text{m}$ to $37 \mu\text{m}$ over the same period (Fig. 1).

Figure 2a further supports this conclusion, showing the increase in the median floc size during the flocculation experiment increased faster for the experiments with kaolinite and biodepost than for kaolinite alone. Figure 2b

shows that the turbidity during the duration of the flocculation experiments decreased for both treatments at a similar rate. Decreasing 12 NTU in the kaolinite treatment and 13 NTU in the kaolinite + biodeposit treatment.

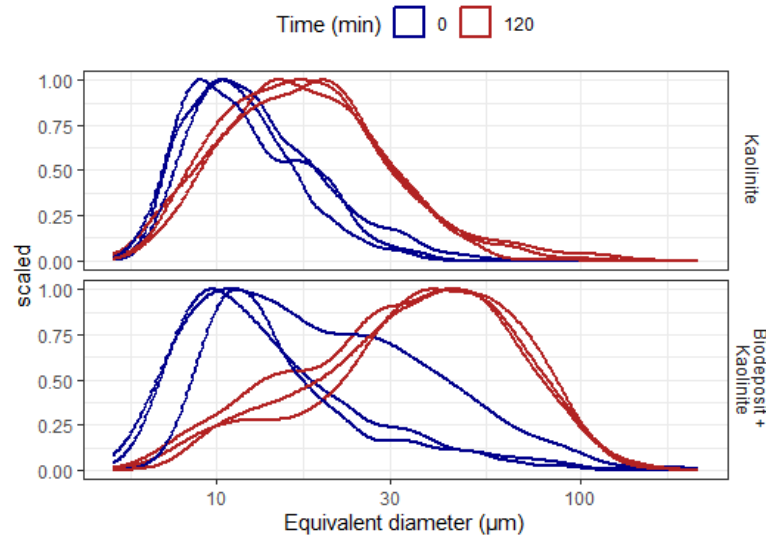


Figure 1 Particle size distributions of kaolinite alone (top) and kaolinite combined with biodeposits (bottom) at 0 min (blue curves) and 120 min (red curves).

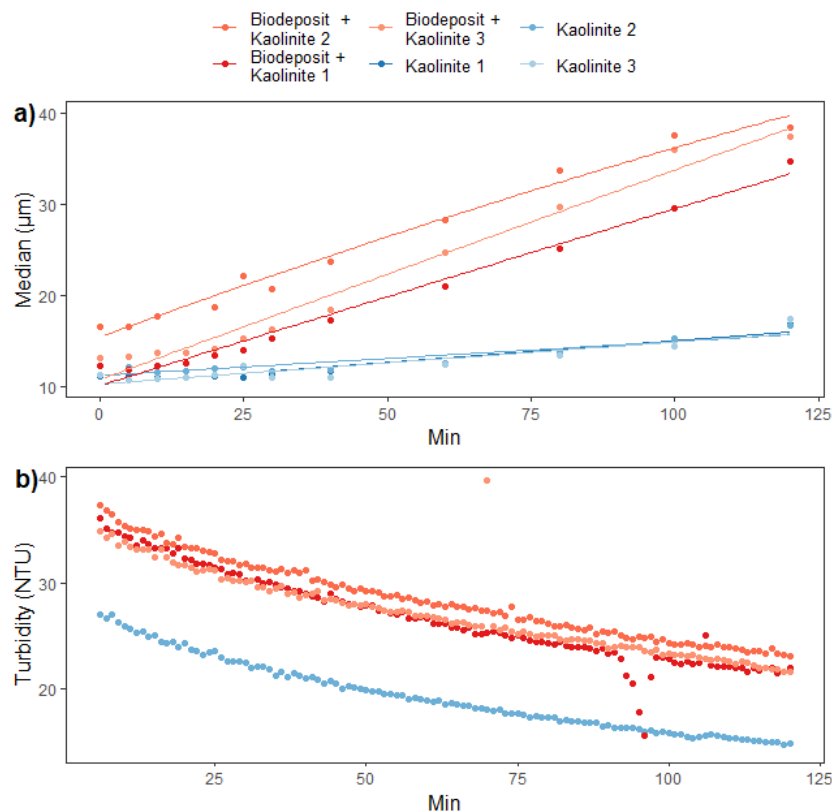


Figure 2 a) Time evolution of the median floc size for kaolinite (blue) and kaolinite with biodeposits (red). Each point set and fitted line corresponds to a different replicate. (b) Corresponding turbidity (NTU) measurements over

the same period.

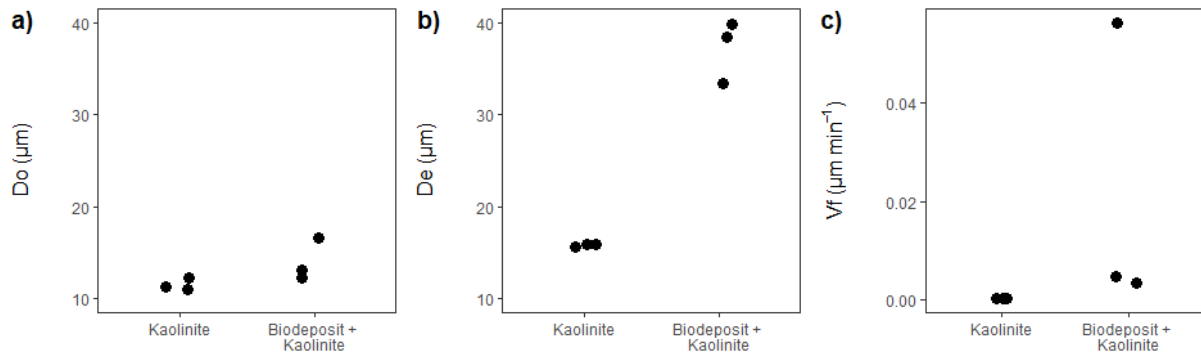


Figure 3 (a) Initial floc diameter (D_0), (b) equilibrium floc diameter (D_e), and (c) flocculation velocity (V_f) for kaolinite and kaolinite with biodeposits.

From the asymptotic regression model (Fig. 2a), we obtained the initial floc size (Fig. 3a), the median floc size at flocculation equilibrium (Fig. 3b), and the flocculation velocity (Fig. 3c). Our results show that adding biodeposits to kaolinite leads to larger flocs and higher flocculation velocities compared to kaolinite alone. Specifically, the initial floc diameter of kaolinite alone was around 11 µm, similar to the kaolinite + biodeposit treatment at 13 µm. After 120 min of flocculation, kaolinite-only flocs remained relatively small at about 15 µm, while kaolinite + biodeposit flocs reached approximately 38 µm. Although the flocculation velocity of kaolinite + biodeposit flocs was higher than that of kaolinite alone, it also showed greater variability.

Overall, the presence of biodeposits greatly enhances floc formation compared to pure kaolinite suspensions.

The next step of our proposal included measuring the settling velocity of the flocs. While we recorded preliminary videos for this purpose, processing them requires more time and expertise than initially anticipated. As a result, unfortunately we do not present definitive settling velocity data at this stage.

To our knowledge, the experiments presented here are quite novel and open up intriguing questions regarding the role of organic matter—in particular, biodeposits—in sediment aggregation. Previous studies on bioflocculation in the open sea have primarily focused on the contribution of phytoplankton to the aggregation of SPM through the production of marine gels (Mari et al., 2017). However, in turbid areas such as the BPNS, it is becoming increasingly evident that we must also consider the influence of organic matter and its degree of mineralization (Fettweis et al., 2022; Schartau et al., 2019).

Our study demonstrates that organic matter produced by mussels, specifically in the form of biodeposits, has the potential to significantly enhance sediment aggregation. Although our laboratory results are compelling, field experiments should be carried out to validate these findings under field conditions. Future investigations should examine the effects of varying turbulence and concentrations of both SPM and fecal pellets. Such comprehensive studies will help to clarify the role of organic matter in sediment dynamics and provide a more complete understanding of bioflocculation processes in turbid environments.

References

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3. Overview of the expenditures

Describe in detail how the requested fund was spent within the implementation period (1 March 2024 and 28 February 2025). Be as specific as possible.

In summary this grant was used to build a settling column with a camera and to add a turbidimeter to the flocculation chamber. More details of the expenses are attached to the additional folder with the expenses.

Expense	Budget
23024 - Vlaams Instituut voor de Zee	5000
20031090/MPICJV/Small Equipment	-133.13
20031090/MPICJV/Small Equipment	-33.45
20031090/JIZFS3/Small Equipment	-36.75
PICO 70124361 Punkt Lasermodul	-27.11
BFS-U3-200S6C-C	-754.25
Seapoint Turbidit Meter STMII	-2627.9
mating connector	-230.29
delivery costs	-135.46
Seapoint Turbidit Meter STMII	-328.37
mating connector	-28.78
delivery costs	-16.93
Rol Parafilm 38mx10cm (4inchx125ft)	-46.19
ecoSHIELD™ Eco Nitrile PF 250, ongepoede	-177.18
microtube 1,5ml met ClearLock snapcap	-66.41
Microtube 2 ml - np Safelock-Cap	-48
Serologische pipet - 10ml - PS, steriel,	-281.4
Pipet serologisch 50 ml, PS, steriel, in	-175.47
Pipet serologisch-25ml - ind. steriel verpakt	-310.06
Pipet serologisch-50ml - ind. steriel verpakt	-175.47
Cell scraper GST L235mm PP/blad in PE 12,5mm	-91.87
AMPHOTERICIN B SOLUTION STERILE-FILTERED	-41.72
	-766.19

4. Pictures

A set of five pictures (low resolution in this document). The five high resolution pictures should be delivered to VLIZ by email to karen.rappe@vliz.be.

