Dinoflagellate Cyst Distribution in Relation to the Sediment Composition and Grain Size in the Coastal Area of Pangkajene, South Sulawesi, Indonesia

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Abstract

Dinoflagellate cysts have an important role for their bloom dynamics, which are commonly deposited along fine sediment grains and become the source of the next bloom. This study aimed to describe the cyst banks species composition, and their relationship with the sediment particles size and plankton composition in the coast of Pangkajene, South Sulawesi. Cysts observed in this study were extracted from seabed sediments collected from 9 sites. A potential cyst bank, with a cyst density of 240 cysts.g^{-1} sediment wet weight was found at PK-19, located in proximity to a large harbour complex. Although unusual, cyst density was significantly and positively correlated with the percentage of gravel. In contrast, the diversity of cysts seems to be affected by the percentage of fine sediments, such as silt and clay. None of the sediment composition was found strongly and significantly affecting both cyst density and diversity. A southward increase in cyst density was similar to the trend in the cell density of its planktonic form. Cysts of Protoperidinium spp., Scrippsiella spp., and Pheopolykrikos hartmannii were common and abundant in the sediment. Aside from Protoperidinium spp., most dinoflagellate species found in cyst form were absent from the water column. The occurrence of cysts of harmful dinoflagellates with records of devastating blooms in other coastal areas in Indonesia, such as Margalefidinium polykrikoides, Pyrodinium bahamense, and Gymnodinium catenatum, signifies a need to regularly monitor the area around Pangkajene coast to mitigate impacts of future blooms.

Keywords: *sediment, cyst bank, harmful dinoflagellate*

Introduction

Dinoflagellate blooms are a rising ecological problem in Indonesian coastal waters, with large blooms of harmful species, such as *Margalefidinium polykrikoides* (=*Cochlodinium polykrikoides*), has caused economic and ecosystem damages in Lampung Bay (Thoha *et al.*, 2019). Several cases of paralytic shellfish poisoning (PSP) have also been reported in the coastal area of Cirebon from 2016 to 2018, which were caused by the consumption of shellfish contaminated by saxitoxin produced by *Pyrodinium bahamense* (Nurlina and Liambo, 2018, Rachman *et al.*, 2019). Other blooms of dinoflagellate species, such as *Noctiluca scintillans*, *Prorocentrum minimum*, *Alexandrium affine*, *Gymnodinium* sp., and *Gonyaulax spinifera*, have been reported to cause ocean discolouration and other harmful effects, such as mass fish mortality, in many coastal areas in Indonesia (Praseno *et al.*, 2003). With the ongoing combination effect of climate change and rapid growth of coastal anthropogenic activities, which affects the rainfall pattern, nutrient flux, temperature, and pH level of the ocean, it was expected that the frequency, duration, and negative impacts of harmful algal blooms (HABs) would be increased in the future (Wells *et al.*, 2020).

Some dinoflagellates are known to produce a cyst, which is an immotile form of the resting or dormant stage produced as an adaptation to unfavourable environmental conditions (Anderson, 1989, Genovesi-Giunti *et al.*, 2006, Tian *et al.*, 2017, Sala-Pérez *et al.*, 2020). Cyst production in dinoflagellate species is a part of its survival and dispersion strategy, which ensures its domination during recurrent blooms (Satta *et al.*, 2013). Due to similarity in the mass and hydrodynamic properties, dinoflagellate cyst, or dinocyst, is often distributed and deposited in the sediment alongside the fine-grained sediment particles, such as silt and clay (Anderson *et al.*, 2003, Godhe and Mcquoid, 2003,

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Tian *et al.*, 2017). The accumulation of cysts in the sediment will then form a 'cyst beds', 'cyst bank', or 'seed bank', which often become the starting point of the dinoflagellate blooms (Genovesi-Giunti *et al.*, 2006, Tian *et al.*, 2017, Brosnahan *et al.*, 2020). The settled dinocyst could enter a long period of dormancy and some dinocysts remain viable for more than 150 years, which could be 'revived' from the sediment under a favourable condition (Delebecq *et al.*, 2020). The ability of dinocyst to survive in an extreme condition for a long period could be an important factor for them to be able to perform a jump dispersal via ship ballast water tanks (Hallegraeff, 1998). On the other hand, the species composition in the cyst banks could reflect the past condition in the water column, such as the nutrient level, salinity, temperature, primary productivity, and anthropogenic activities, including industrial pollution (Shin *et al.*, 2010, Satta *et al.*, 2014, Sala-Pérez *et al.*, 2020). Thus, cyst composition is also important to aid in building a mitigation strategy against recurrent or future dinoflagellate blooms in the coastal area.

This study focused on the coastal area of Pangkajene, South Sulawesi, Indonesia. The area was known for its economic potential in various important ecosystems along the coast, particularly the coral reefs and mangrove forests. Those ecosystems have been under threat from unsustainable fishing practices and land conversion from mangrove to shrimp ponds or industrial zones, including harbour and power plants (Noveria *et al.*, 2007). The coastal water of Pangkajene was also influenced heavily by nitrogen (N) and phosphorus (P) input via run-off from the agriculture and aquaculture practices on land (Nasir *et al.*, 2018), which influence the phytoplankton communities in the water. Rashidy *et al.* (2013) reported a case where the dinoflagellate *Ceratium* became a co-dominant genus alongside the diatom *Coscinodiscus* in the coastal area of Tekolabbua, a region of Pangkajene. It indicated that dinoflagellate could replace the commonly dominant diatoms in the coastal area of Pangkajene, which might also produce cysts deposited in the bottom sediment and might fuel the future dinoflagellate blooms. Therefore, this study aimed to determine the location of the dinoflagellate cyst bank, assess its assemblages, and find the relationship between the density and diversity of dinocyst to the sediment characteristics in Pangkajene coastal area.

Materials and Methods

Sample collections and in-situ environmental data measurement

Seabed sediment samples were collected at the coasts of Pangkajene of the southern Sulawesi Peninsula. Nine sites were set up and separated into three zones, (i) northern zone located near Pangkep Island, between Bombang-Talaka, Kanaungan, and Bonto Manai, (ii) middle zone in front of Baccini Baji Harbour, and (iii) southern zone in proximity with Semen Tonasa Harbour (Figure 1.). The climate in the area is wet tropic, including two climate types, C1 type (<2 dry months) and C2 type (2–3 dry months), and both types have a consecutive 5–6 wet months in a year (Anonym, 2017).

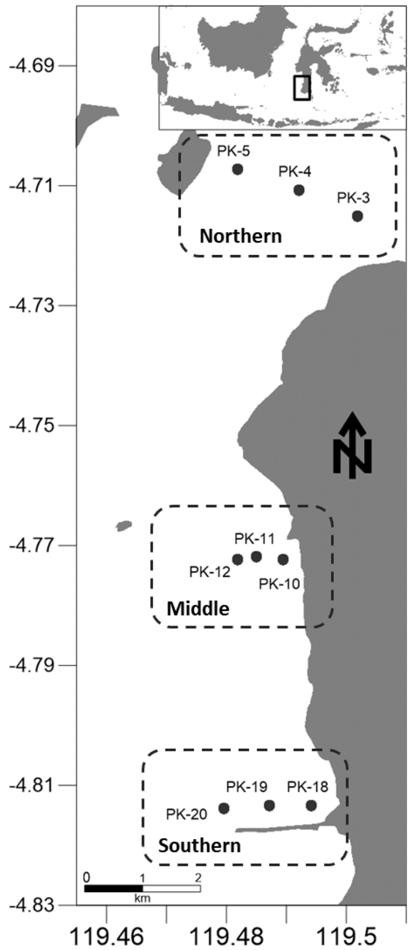

Figure 1. Map of the study site in the coastal area of Pangkajene, South Sulawesi, Indonesia. Nine out of 22 original sampling sites were sampled for their sediment in this study.

The sediment samples were collected using an Ekman grab, placed in a sealed polyethylene box and kept in a cool and dark storage. No preservative was added to the samples. In the laboratory, all sediment samples were kept in the refrigerator at 4°C. Water depth, temperature, pH, and dissolved oxygen (DO) were measured in situ using a YSI 556 Handheld Multi-Parameter tester. Sea surface salinity (±0.5m) was measured using a hand refractometer. Additional information on the weather, sea condition, state of anthropogenic activities, and historical data on algal blooms was also collected by direct observation and interviews with local people.

Dinocyst extraction and sediment analysis

Dinocysts were extracted from 4 g wet sediment using a gradient density method with a mixture of Ludox and sucrose compounds as described in Blanco (1986), Mizushima *et al.* (2007), and Genovesi *et al.* (2009). For grain size analysis, between 50 to 100 g of sediment was sorted using a series of metal sieve with mesh size of 4, 2, 1, 0.5, 0.25, 0.125, and 0.063 mm. The amount of sediment used in the analysis depends on the type of sediment, in which sediment rich with fine grain needs up to 100 g. Sediment type and composition were analysed under a Leica MZ-6 stereo microscope following Wentworth (1922) after an oven drying to remove the water in the sediment.

Analysis of dinocyst assemblages

Dinocysts were observed by an inverted phase-contrast microscope Nikon Diaphot 300 equipped with a digital camera Canon EOS 700D. Dinocyst species were identified based on the cyst morphology described in Al-Yamani and Saburova (2010). Identified dinocysts were counted in a fraction (1.0–1.5 mL) in a Sedgewick-Rafter counting chamber as described in Arinardi (1997) and Legresley and Mcdermott (2010).

Analysis of planktonic phytoplankton assemblages

This study utilized a phytoplankton species dataset from the Plankton Laboratory, Research Center for Oceanography, Indonesian Institute of Sciences (RCO-LIPI; now it merged into RCO-BRIN) (Thoha *et al.*, 2021). The nine sediment sampling sites in this study are a part of 22 sites in Thoha *et al*. (2021). Details on the planktonic phytoplankton sample collection and analysis are described in Thoha *et al.* (2021).

Data analysis

A descriptive analysis of the phytoplankton community, such as the absolute and relative density of phytoplankton genera and dinocysts was done in Microsoft Excel 365 ver 2002 using the formula as described in Cox (1990). Statistical analyses were performed using R Studio (Rstudio Team, 2015). Phytoplankton and dinocyst diversity were described using Shannon-Wiener index (*H'*) and were calculated using a formula based on Krebs (1989). Non-metric multidimensional scaling (nMDS) combined with unweighted pair group method with arithmetic mean (UPGMA) (Zuur *et al.*, 2007; Oksanen, 2010; Borcard *et al.*, 2011) were used to analyse the similarity or dissimilarity between sampling sites based on the dinocyst composition and sediment characteristics (composition or grain size). Bray-Curtis dissimilarity index was used to construct the nMDS plot and UPGMA grouping.

In this study, a Pearson correlogram was constructed using 'corrplot' package in R (Wei *et al.*, 2017) to determine the strength of the relationship between dinocyst and sediment characteristics. Additionally, redundancy analysis (RDA) (Borcard *et al.*, 2011, Zuur *et al.*, 2007) was used to further explore the relationship between each dinocyst species with sediment grain size and composition. The RDA analysis was done in R Studio using 'vegan' package (Oksanen *et al.*, 2015).

Result and Discussion

Mismatch composition of the cyst and planktonic dinoflagellate species

In total, cysts of 20 dinoflagellate species were found in the coastal sediment of Pangkajene. Among those species, *Protoperidinium* spp., *Scrippsiella* spp., and *Pheopolykrikos hartmannii* (= *Polykrikos hartmannii*) were the three most abundant dinocyst in the sediment (Figure 2C.). The cyst density of those species was 207, 113, and 113 cysts.g^{-1} of wet sediment, respectively. The cysts of harmful and toxic dinoflagellates, such as *M. polykrikoides*, *P. bahamense*, and *Gymnodinium catenatum*, were also found in the sediment (Figure 2C). The cyst density of these three harmful species was 40, 33, and 20 cysts.g^{-1} of wet sediment, respectively. The planktonic forms of these harmful species were not found in the water column of Pangkajene (Figure 2B and Figure 2C.). Similarly, based on Thoha *et al.* (2021), the planktonic forms of *Scrippsiella* spp. and *P. hartmannii* were not found in the water column (Figure 2B and Figure 2C).

A mismatch of species compositions between dinocyst assemblages and planktonic dinoflagellates was reported in the coastal areas of Cirebon (Rachman *et al.*, 2019). These mismatch cases could be caused by dormant cysts of many dinoflagellates that cannot undergo germination process due to a lack of triggering factor or unsuitable water conditions to support their planktonic form. In this case, the water condition in Pangkep might only be suitable for the growth of several dinoflagellate species, particularly *Protoperidinium* spp. (Figure 2B and Figure 2C). However, those assumption needs to be properly studied and proved.

Thoha *et al.* (2021) showed that the phytoplankton communities of Pangkajene (Pangkep) were diatom-dominated with dinoflagellate density only consisting of <5% of total phytoplankton cell density (Figure 2A). Six dinoflagellate genera from the Pangkajene waters were also reported by Thoha *et al.* (2021), with

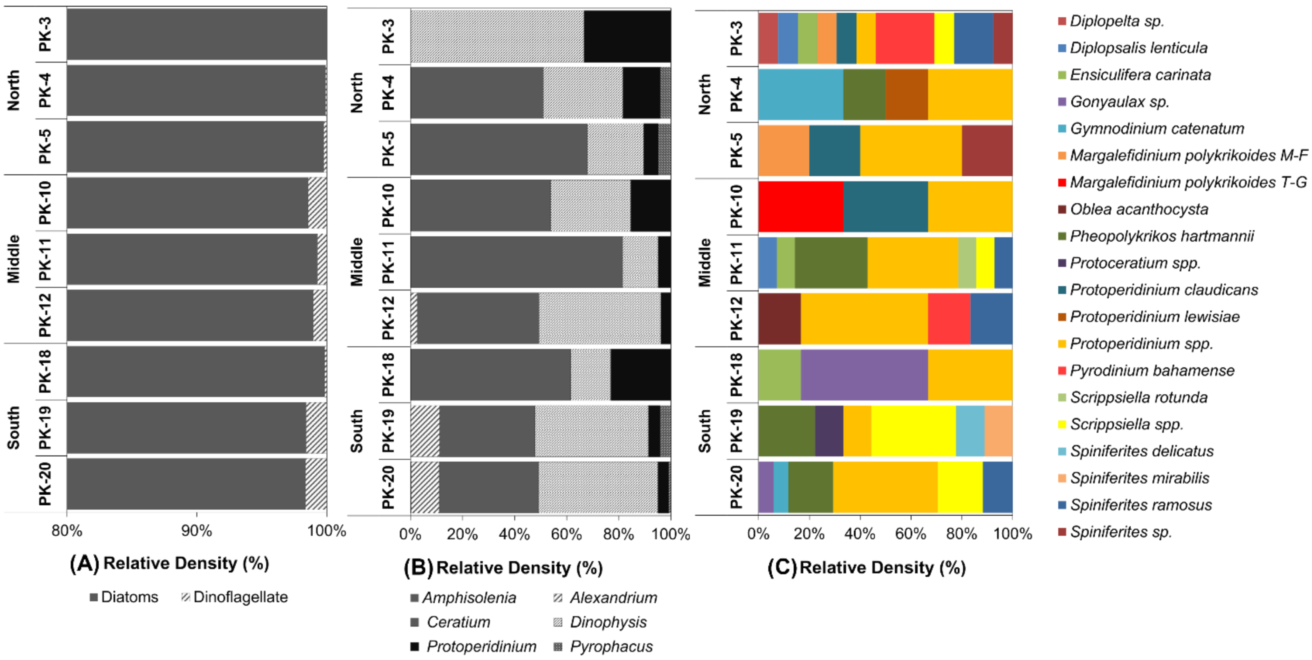

Figure 2. Histogram showing (A) comparison between the density of diatoms and dinoflagellate planktonic cells, (B) genus composition within the dinoflagellate group, and (C) dinocyst species assemblages. Histogram (A) and (B) were constructed based on the data from Thoha *et al.* (2021).

Ceratium and *Dinophysis* as the two most abundant genera (Figure 2B). *Ceratium* is commonly found in eutrophic waters and often used as an indication of eutrophication events in the coastal water (Adnan, 1992, Thoha and Rachman, 2015). On the other hand, *Dinophysis* is a cosmopolitan with ten toxigenic species, *Dinophysis acuta*, *D. miles*, *D. caudata*, *D. acuminata*, *D. fortii*, *D. infundibulum*, *D. ovum*, *D. saccula*, *D. tripos*, and *D. norvegica* (Ajani *et al*., 2016). However, there were no *Dinophysis* cyst was found in the sediment samples of Pangkajene in this study (Figure 2C). Note that the production of cyst in *Dinophysis* genus are still under debate despite *Dinophysis*-cyst like shapes or structures have been reported in some studies (Blanco *et al*., 2005, Reguera *et al*., 2012).

General trends in the dinocyst density and diversity

The dinocysts density in the coastal sediment of Pangkajene was between 20 to 240 cysts.g^{-1} of wet sediment (Figure 3A and 3C). According to McMinn (1991) and Tian *et al.* (2017) classification, the cyst density in Pangkajene was classified between the low (10–100 cysts.g^{-1}) to moderate (100–1000 cysts.g^{-1}) class. The total dinocyst density in Pangkajene was higher than those previously reported in Indonesia, such as Jakarta Bay, Ujung Pandang, and Flores Island (Matsuoka *et al*., 1999). In Pangkajene, a southward increase in the general dinocyst density (Figure 3A) was found in this study, which differs from the trend of dinocyst diversity (Figure 3B.).

The highest dinocyst density in Pangkajene was found at PK-19 and the lowest at PK-18 (Figure 3C.). Those two sites were located next to each other, and the PK-18 was closer to the coastline compared to PK-19 (Figure 1.). A similar low cyst density at the sampling site closest to the shoreline was also reported from *P. bahamense* cyst in Cirebon coastal area in 2017 (Rachman *et al*., 2019). It was suspected that high anthropogenic activities in the waters closest to the shoreline cause stress to the dinoflagellate communities by exerting a higher pressure from anthropogenic pollutants, such as heavy metals from coastal industries (Godhe and Mcquoid, 2003). Additionally, impacts of highly eutrophic and polluted water were suggested to reduce the diversity and richness of dinocyst in the sediment (Mohamed and Al-Shehri, 2011, Satta *et al.*, 2014). That might also be the reason of the lowest dinocyst diversity at PK-18 (Figure 3D.).

Bottom sediment characteristics

The bottom sediments of Pangkajene consisted of finer sediments, mainly silt and clay (Figure 4A.). The silt was more abundant in the middle zone, while the clay was more abundant in the north zone. On the other hand, detrital minerals and rock fragments were the most common components of the bottom sediments in Pangkajene (Figure 4B.). The detrital minerals contributed up to 90% of the total sediment composition, while rock fragments contributed up to 40%. Site-to-site variation in the

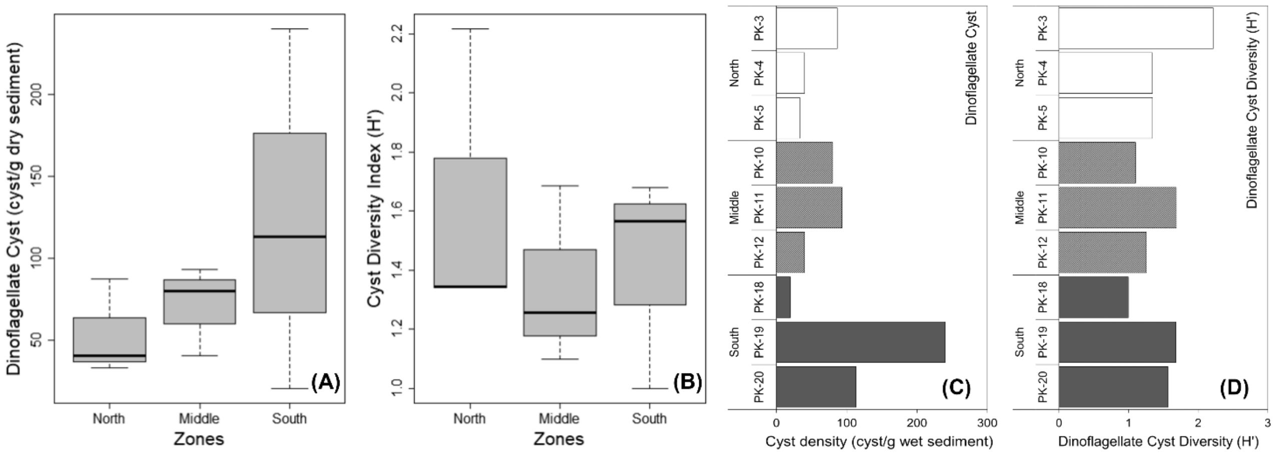

Figure 3. Boxplot showing a general trend of (A) total dinocyst density, and (B) diversity (*H'*) in the North, Middle, and South area of Pangkajene. Histogram showing a more detailed trend in (C) dinocyst density and (D) diversity (*H'*) in each sampling sites of this study.

sediment composition in Pangkajene was high (Figure 4B.), with some sampling sites showing distinct composition related to the distance from the shoreline and anthropogenic activities at the shoreline (Figure 6). Sites located closer to the shoreline had a higher proportion of rock fragments, as heavier materials from the land would sink faster than lighter materials, such as detrital and clay minerals (Figure 4B.). However, an anomaly was found at site PK-18, in which the sediment consisted of mainly detrital minerals and rock fragments (Figure 4B), while the sand was the dominant grain in the sediment of this site (Figure 4A.).

Characterizing sites based on phytoplankton composition, dinocyst composition, grain size, and sediment composition

The nMDS analysis showed that site PK-18 had distinct grain size and sediment composition that separated it from the other sites (Figure 5A and Figure 5B). In addition, PK-18 and PK-19 were found with unique dinocyst species compositions (Figure 5D.). The nMDS analysis also showed that the northern and southern sites [ii] had similar characteristics in the cyst density and grain size, which differed from the sites at the middle zone [i] (Figure 5A). Between cyst density and sediment composition, the sites furthest from the shore [ii] had different characteristics compared to other sites closer to the land [i] (Figure 5B.). On the other hand, most sites in this study had similar dinocyst composition, with an exception in sites PK-18 and PK-19 (Figure 5D). Note that the sites in Pangkajene could be split into two groups based on the planktonic dinoflagellate genus assemblages, which were the [i] site groups closest to the land, and [ii] site groups farther away from the land (Figure 5C.).

Site PK-18 was the only site in which the sediment grain size was dominated by sand (Figure 4). It was generally known that dinoflagellate cysts tend to accumulate in the sediment with finer grain instead of course and sandy sediment (Anderson *et al.*, 2003, Tian *et al.*, 2017). That condition might be related to the low dinocyst density in PK-18. The high proportion of sand in PK-18 suggested that the higher physical process, such as current and wave, might have prevented the deposition of finer materials to the area around the site. Another possible reason was the sandy bottom might be the leftover of the construction of a large harbour, the Semen Tonasa Harbour and that frequent sediment disturbance caused by the movement of ships could prevent the deposition of dinocyst in the sediment. Even so, coastal enclosed areas, such as harbours, often have favourable conditions, such as low hydrodynamic disturbance and high nutrient availability, that could trigger the blooms of dinoflagellate in that area (Tian *et al.*, 2017). On the other hand, the site groupings based on the planktonic dinoflagellate assemblages indicate that the water condition in PK-18 was similar with some sites closer to landmass, such as PK-4 and PK-10 (Figure 5C.).

Relationship between sediment characteristics and dinocysts

The dinoflagellate cyst density was positively correlated to the percentage of gravel, silt, and clay, as well as the benthic foraminifera in the sediment of Pangkajene (Figure 6.). In contrast, dinocyst density was negatively correlated with the percentage of pebble, sand, and detrital minerals (Figure 6.). The analysis also showed cyst diversity was positively correlated with silt, clay, and molluscan shells while being negatively correlated with pebble, sand, and detrital minerals (Figure 6.). Surprisingly, the percentage of gravel was the only sediment parameter that has a significantly strong correlation (p <0.05; R >0.7) with

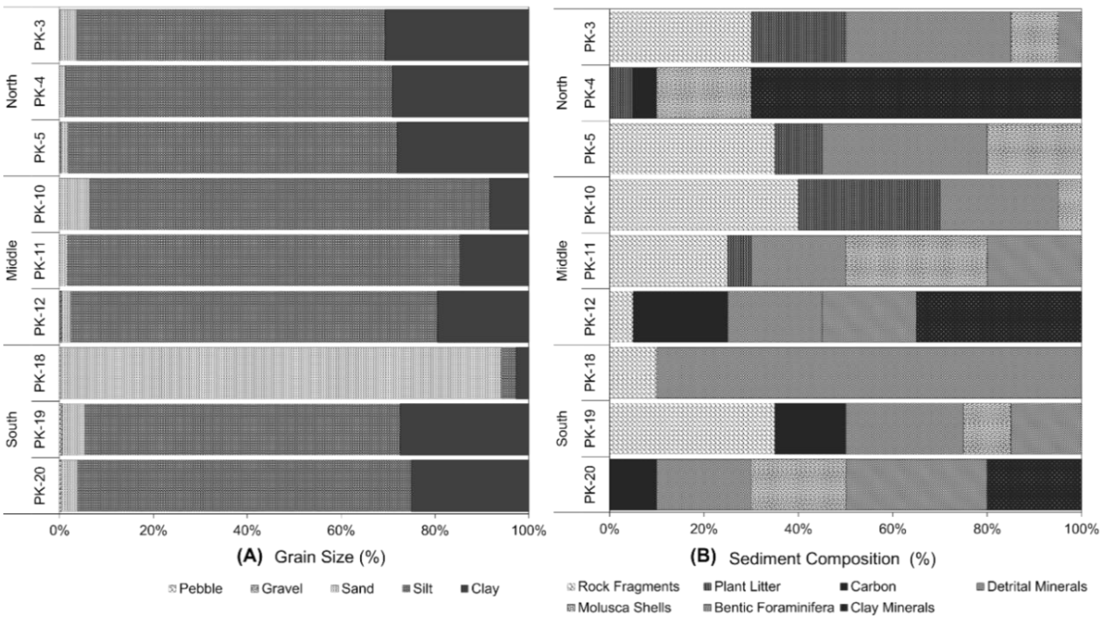

Figure 4. Histogram of (A) Grain size and (B) sediment composition of the bottom sediment in Pangkajene coastal waters.

dinocyst density in this study (Figure 6.). On the other hand, cyst diversity has the strongest correlation with the percentage of finest sediment grain in the sediment, which is clay (Figure 6.).

The general positive correlation between dinocyst with finer sediment, such as silt and clay, was expected. Dinoflagellate cysts were generally smaller or similar size (<63 μm) as silt particles and it often deposited at the same area where silt and clay were deposited (Godhe and Mcquoid, 2003). Furthermore, a high percentage of finer sediment, such as silt and clay, in one area also suggested that the area exhibiting high sediment stability due to weaker disturbance or turbulence effect of wave and currents to the bottom sediment (Godhe and Mcquoid, 2003; Tian *et al.*, 2017). Even so, it was interesting to find that gravel, which is a larger sediment grain, only have a significantly strong and positive correlation (p <0.05; R >0.7) with dinocyst density but not the cyst diversity (Figure 6.). Although unusual, the correlogram (Figure 6.) did show a significantly strong and positive correlation between the proportion of carbon and gravel in the sediment (Figure 6), which might be a reason for the correlation between dinocyst and gravel in this study. As a note, a significantly positive relationship between the total organic carbon (TOC) in the sediment and the total dinocyst abundance was reported by Tian *et al.* (2017). However, the relationship between the dinocyst abundance to the TOC in the sediment were not fully understood (Tian *et al.,* 2017).

On the other hand, RDA analysis showed each dinocyst was affected by a combination of different sediment characteristics (Figure 7.). For example, the cyst density of *P. hartmannii* was weakly influenced by the grain size of sediment in this study (Figure 8A.). However, it can be found at higher density in sites with a higher percentage of carbon and benthic foraminifera in the sediment (Figure 7B). The cysts of two other most abundant species, *Protoperidinium* spp. and *Scrippsiella* spp., were found with contrasting relationship with the sediment grain size, in which *Scrippsiella* spp. was more abundant at sites with sediments with a combination of a higher proportion of gravel, silt, and clay in contrast to *Protoperidinium* spp. (Figure 7A.). The cyst of *Protoperidinium* spp. was more abundant in sites with a higher percentage of sand (Figure 7A.). Both *Scrippsiella* spp. and *Protoperidinium* spp. also had a different relationship with sediment composition, in which *Scrippsiella* spp. show indication of positive correlation with a higher proportion of molluscan shells in the sediment, while *Protoperidinium* spp. were more abundant in sites with a higher percentage of clay minerals in the sediment (Figure 7B.). In the case of *Scrippsiella* spp. the low oxygen concentration in the sediment is known to severely inhibit the germination of some *Scrippsiella* species, such as *Scrippsiella hangoei* (Kremp and Anderson, 2000). Thus, eutrophication in the water that causes oxygen depletion in the bottom layer or the sediment layer, could also limit the density of planktonic *Scrippsiella* spp. in the water column (Kremp and Anderson, 2000), which then limit the number of cysts produced by the species. On the other hand, the increasing abundance of heterotrophic species, particularly *Protoperidinium* spp., could be a sign of rising cultural anthropogenic eutrophication in the water (Azanza *et al.*, 2004; Satta *et al.*, 2014).

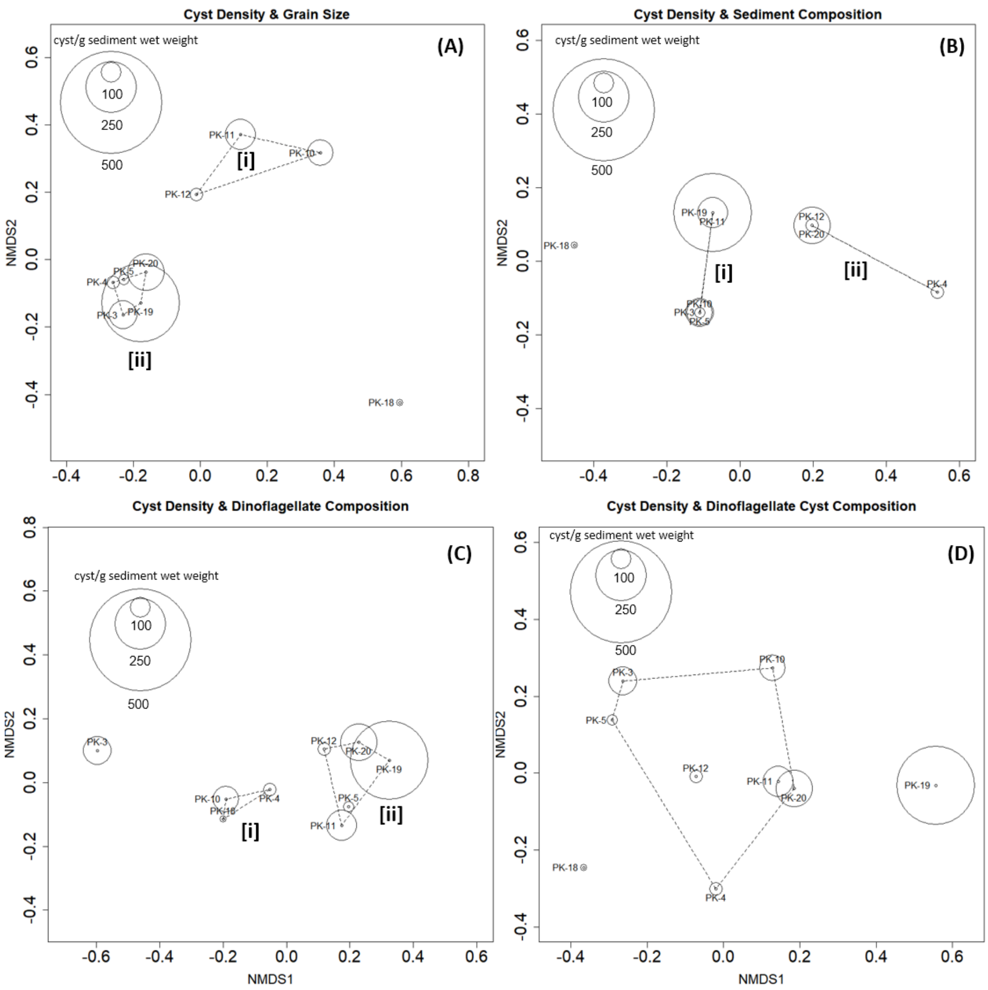

Figure 5. nMDS scatterplot with sites' position and groupings based on Bray-Curtis UPGMA clustering analysis of (A) sediment grain size, group [i] northern and southern sites, [ii] middle sites; (B) sediment composition, group [i] sites closer to the shoreline, [ii] sites furthest from shoreline; (C) Planktonic dinoflagellate composition, group [i] sites closer to the shoreline, [ii] sites further from the shoreline; (D) Dinocyst composition. Circles in the scatterplot represent cyst density in cysts.g^{-1} sediment wet weight. nMDS scatterplot (C) was created using a combination of cyst density data of this study and planktonic dinoflagellate composition data from Thoha *et al.* (2021).

The RDA analysis also showed the cyst density of each harmful or toxic dinoflagellates species was affected by different sediment characteristics (Figure 7.). The cyst density of toxic species, *G. catenatum*, was found at a higher density in sites with a higher proportion of clay mineral (Figure 7B.) but did not seem to be strongly affected by the sediment grain size (Figure 7A.). The cyst density of *P. bahamense* had a strong relationship with a higher proportion of fine sediment, particularly silt (Figure 7A.) and a higher proportion of plant litter in the sediment (Figure 7B.). A study on *P. bahamense* cyst in Cirebon found that silt has a negative correlation with density (Rachman *et al.*, 2019). Note that the positive relationship between the *P. bahamense* cyst density with the proportion of molluscan shells was found both in Pangkajene and Cirebon (Rachman *et al.*, 2019). Unfortunately, the reason behind this relationship was not clear, although there is an indication that abundant molluscan shells are related to well-oxygenated sediment. Even so, due to the lack of sediment oxygen concentration data in this study, it is not possible to prove the hypothesis.

Note that the cyst density of two morphotypes of *M. polykrikoides*, the Matsuoka-Fukuyo-like (M-F) and the Tang-Gobler-like (T-G), had a different relationship with grain size and sediment composition (Figure 7.). The cyst density of *M. polykrikoides* M-F did not seem to be strongly influenced by grain size (Figure 7A.) but could be found in sites with a higher proportion of plant litter and detrital minerals (Figure 7B.). On the other hand, the cyst density of *M. polykrikoides* T-G should be higher at sites with a higher proportion of rock fragment and larger grain size (pebble) (Figure 7A and Figure 7B.). It was not

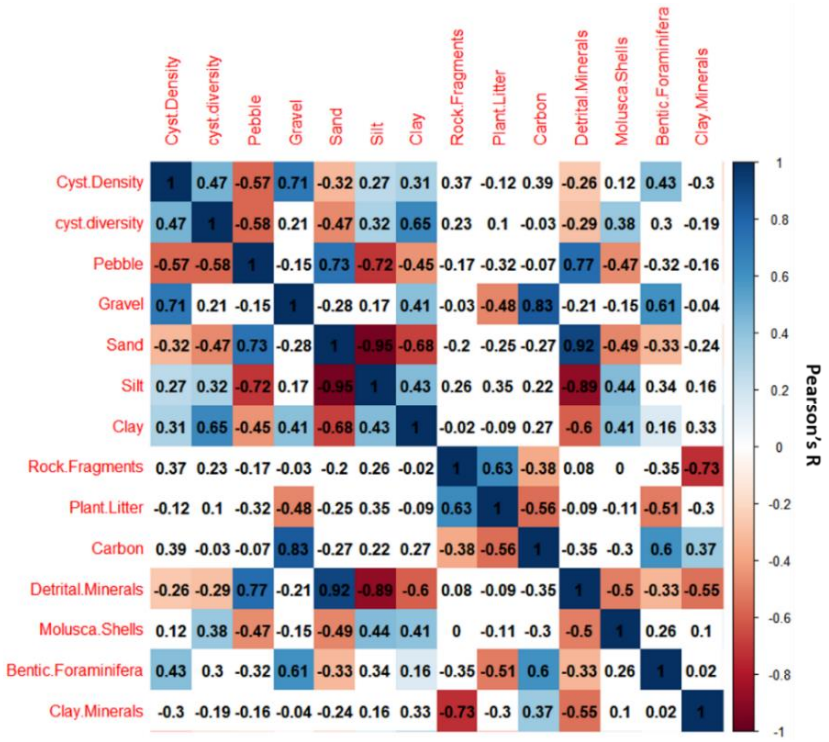

Figure 6. Pearson Correlogram or Correlation Matrix showing correlations between parameters in this study. Blank tiles represent non-significant correlation ($p>0.05$). Strong correlation showed by darker tiles or with Pearson's R-value above 0.7 or -0.7.

known why the two morphotypes of *M. polykrikoides* found in Pangkajene seems to be influenced by different sediment characteristics. These two morphotypes were also found in the sediment of Lampung Bay, and the latter morphotype might be related to the species caused fish-killing blooms (Puspasari *et al.*, 2018; Thoha *et al.*, 2019). As a note, *M. polykrikoides* M-F morphotype was also formed by a recently described unarmored dinoflagellate *Pseudocochlodinium profundisulcus*, and therefore investigation of its motile form is an important topic in Indonesia because the species has caused harmful blooms in China (Hu *et al*., 2021).

The different trend of interaction between the cyst density of each dinoflagellate and sediment grain size (Figure 7A.) could be related to their different sinking ratio. For example, cysts covered with ornamentation such as spines, e.g., *Gonyaulax spinifera*, *G. scrippsae*, *Lingulodinium polyedrum*, and *Protoperidinium conicum*, sink slower compared to the cysts with less ornamentation and composed of heavier element (calcareous cyst), e.g., *Scrippsiella trochoidea* (Anderson *et al.*, 1985). Therefore, the cyst would be accumulated along with the sediment grain with similar sinking properties, which might be the reason why the heavier cyst, such as *Scrippsiella* spp., tends to be accumulated in sites with a higher proportion of larger and heavier grains in this study. Yang *et al.* (2018) showed that the addition of fine sand particles in the culture could significantly increase the cyst production of some dinoflagellate, such as *S. trochoidea*, *Biecheleria brevisulcata* and *Levanderina fissa*. Meanwhile, the cyst production of some other dinoflagellates, such as *P. hartmannii*, *M. polykrikoides*, and *Akashiwo sanguinea*, was barely affected by additional sand particles in their growth (Yang *et al.*, 2018).

Despite the lack of sediment nutrient data in this study, the sediment composition data could act as a proxy to roughly estimate the condition of the sediment in one sampling site. For example, sites with a high percentage of detrital mineral and plant litter, such as PK-3, PK-5, and PK-10, might have nitrogen-rich sediment as the result of the decomposition process of those organic materials. Thus, any disturbances that resuspend the nitrogen-rich sediment and the dinocyst into the water column (Figure 7B.). It was known that the blooms of *M. polykrikoides* were regulated by the concentration ofnitrogen (N) in the water column, and the species have unique nutritional flexibility that allows it to utilize a wide variety of N compounds, such as nitrate, nitrite, ammonium, urea, and amino acids (Gobler *et al.*, 2012). Nitrogen enrichment due to sediment resuspension in the water has been suspected to trigger the first and largest *M. polykrikoides* blooms in Lampung Bay during the end of 2012 (Puspasari *et al.*, 2018). On the other hand, N-enriched water could

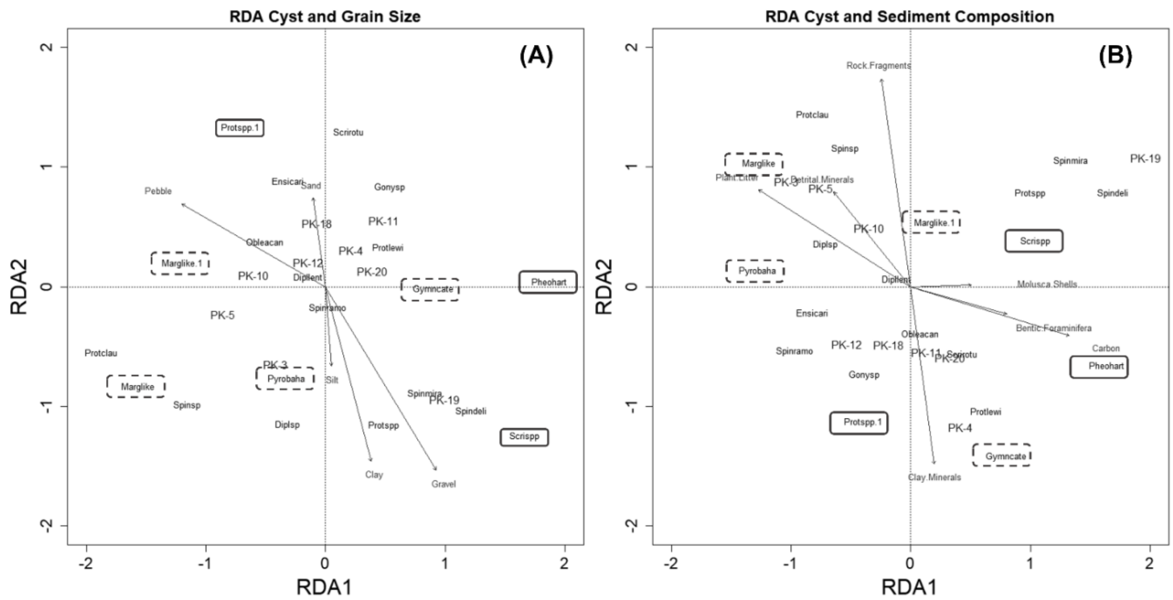

Figure 7. RDA analysis of dinocyst density and (A) grain size and (B) sediment composition. The solid box indicates the most abundant dinoflagellate species, while the dashed box indicates harmful and/or toxic dinoflagellate species. Note: Diplsp = *Diplopelta* sp., Diplent = *Diplopsalis lenticula*, Ensicari = *Ensiculifera carinata*, Gonysp = *Gonyaulax* sp., Gymncate = *Gymnodinium catenatum*, Marglike = *Margalefidinium polykrikoides* – Matsuoka-Fukuyo-like (M-F), Marglike 1 = *Margalefidinium polykrikoides* – Tang-Gobler-like (T-G), Obleacan = *Oblea acanthocysta*, Pheohart = *Pheopolykrikos hartmannii*, Protspp = *Protoceratium* spp., Protclau = *Protoperidinium claudicans*, Protlewi = *Protoperidinium lewisidae*, Protspp.1 = *Protoperidinium* spp., Pyrobaha = *Pyrodinium bahamense*, Scrirotu = *Scrippsiella rotunda*, Scrispp = *Scrippsiella* spp., Spindeli = *Spiniferites delicatus*, Spindeli = *Spiniferites mirabilis*, Spinramo = *Spiniferites ramosus*, Spinsp = *Spiniferites* sp.

as well trigger the rapid growth of other harmful dinoflagellates, particularly *P. bahamense*, which is known to grow rapidly under a high concentration of nitrate and urea in the water (Usup *et al.*, 2012). In this study, *P. bahamense* cyst tends to be abundant in sites with sediment that is richer in plant litter (Figure 7B.). Thus, similar to *M. polykrikoides* M-F, *P. bahamense* also has a chance to grow faster if the sediment resuspension occurred in sites with nitrogen-rich sediments.

The implication to coastal management in Pangkajene waters

The largest cyst bank in Pangkajene was discovered at site PK-19 located near the Semen Tonasa Harbour. The PK-19 also had a distinct dinocyst assemblage characterized by the highest proportion of *Scrippsiella* spp. (Figure 2C.). The cysts of *Protoceratium* spp., *Spiniferites delicatus*, and *S. mirabilis* could be found only from the site (Figure 2C.). With a moderate class cyst density, the site PK-19 might become the source point of blooms of some dinoflagellates, such as *Scrippsiella* spp. The case of cyst bank that became the starting point and sustained a long duration of blooms have been observed in other coastal areas in Southeast Asia, particularly in the recurrent blooms of *P. bahamense* in Manila Bay, Philippines (Azanza *et al.*, 2004, Borja *et al*., 2019, Yniguez *et al*., 2021). On the other hand, the occurrence of *M. polykrikoides* (sites PK-3 and PK-12) and *P. bahamense* cyst (sites PK-10 for T-G type, and PK-5 for M-F type), which have records of devastating blooms in other coastal areas in Indonesia, signify a need to regularly monitor the area around Pangkajene coast. However, it is also important to implement better regulation and coastal management to reduce the nutrient load or concentration in the water. Stronger regulation and better coastal management should help to prevent the emergence of any harmful algal blooms in the Pangkajene coast in the future.

Aside from creating and implementing a mitigation strategy, it is also important to expand the study area on the dinocyst assemblages in Pangkajene coastal waters. As described in this study, there are too many areas with no information about the cyst distribution and assemblages in Pangkajene coastal waters. Even though it is considered as a time-consuming work (Anderson, 1989), any attempt or work to create a detailed and elaborate map of cyst distribution in the Pangkajene coastal water would be a very valuable tool to help the local government to manage the area and mitigate the forthcoming harmful blooms. By knowing the areas of the highest dinocyst accumulation, particularly for the fish-killing or toxigenic species, the local government could develop a strategy to avoid any activities that might disturb the seabed of that cyst bank area.

Conclusion

From this study, the most abundant dinocysts in the sea bottom sediment of Pangkajene were *Protoperidinium* spp., *Scrippsiella* spp., and *P. hartmannii*. Cysts of several harmful or toxin-producing dinoflagellates, such as *M. polykrikoides*, *P. bahamense*, and *G. catenatum* were also found. Aside from *Protoperidinium* spp., most of the dinoflagellate species found in cyst form were absent from the water column. Dinocyst diversity should be higher in the area with a high proportion of clay in the sediment. Although unusual, dinocyst density would be higher at sites with a high proportion of gravel in Pangkajene. Sediment composition did not have a strong relationship with both dinocyst density and diversity. However, each dinoflagellate species did have a specific relationship to either sediment grain size or composition. On the other hand, the largest cyst deposit, or cyst bank, in Pangkajene coast was found in site PK-19, which is in proximity to the Semen Tonasa Harbour. Site PK-19, along with site PK-18, had a distinct dinocyst species composition that is significantly different from the other sites in this study.

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