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Mineral composition of the scallop, *Placuna placenta* shell from the eastern part of East Java Waters, Indonesia

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ABSTRACT
The shell of bivalve such as *Placuna placenta* is one of many biomasses that is commonly used as bio-inspired and biomimetic material. Different habitat may influence the living of organism, including the biomineralization of *P. placenta* shell affecting their mineral composition. This study was aimed to know the mineral composition of the *P. placenta* shell collected from Ujung Pangkah-Grasik and Junganyar Socah-Bangkalan, Indonesia. Observation was carried out on the shell (near umbo) using SEM-EDX (Scanning Electron Microscope-Energy Dispersive X-ray Spectroscopy). The result showed that the shell collected from Junganyar Socah-Bangkalan have larger element quantities (9 types) than that from Ujung Pangkah-Gresik (6 types). The elements found on the shell collected from Junganyar Socah-Bangkalan were: C, O, Na, Mg, Si, Ca, S, Al, and Cl, while from Ujung Pangkah-Gresik were: C, O, Na, Mg, Ca, Zn, and S. The Na/Ca ratio of the shell from Ujung Pangkah-Gresik was higher than that from Junganyar Socah-Bangkalan. The Mg/Ca and S/Ca ratio of the shell from Junganyar Socah-Bangkalan was higher than that from Ujung Pangkah-Gresik.

Keywords: Mineral composition, Scallop, Ujung Pangkah, and Junganyar Socah
Article type: Research Article.

INTRODUCTION
Scallop, *Placuna placenta* is a marine organism that is living in the bottom or sediment of aquatic environments (Yonvitner *et al.* 2011). This organism belongs to Phylum Mollusca, Class of Bivalvia (Swennen *et al.* 2001). It lives in the organic material-rich substrate, and digs holes using its long foot (Dharmaraj *et al.* 2004; Anti *et al.* 2014). The foot is used to prevent mud getting into the gills and the other organs during its movement (Allan 1962). *P. placenta* has a transparent shell, ranging from 3 to 8 cm (Dharmaraj *et al.* 2004; Hasriyadi 2017). The adult scallop is characterized by a thick shell and color gradations like a rainbow, while the young one shows thin purple color in the upper part of the shell (Swennen *et al.* 2001). *P. placenta* is known to have commercial value for the people living in the coastal area of East Java, Indonesia. This organism is one of the most popular marine commodities that are consumed by human in this area (Kurniawan & Wonoseputro 2018). It is used as the material of a unique food, namely Griting Simping (Yaqin and Fachruddin 2018). All parts of *P. placenta* are safe to be consumed by human. Their meat can be used as the high nutrition source, while their shell can be utilized for ornamental accessories, window glass, medical treatments, and animal feeds (Widowati *et al.* 2008; Tongchan *et al.* 2009; Nisra *et al.* 2014; Anti *et al.* 2014; Yaqin & Fachruddin 2018). Another study by Achuthankutty *et al.* (1979) showed that *P. placenta* from Goa India can obtain pearl up to 2 mm. The shell of *P. placenta* contains a lot of essential mineral, such as: Calcium carbonate and Calcite (Li 2013, 2014). The content of calcite on its shell

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is known to have higher energy dissipation than that in the nature (Li 2014). Therefore, it is potential to be used as a filler in several industries, such as: production of fiberglass ship and high technology of polymer manufacturer (Mufidun & Abtokhi 2016). It can also be used as bio-inspired and biomimetic material in the environment. Environmental condition is known to influence the metabolism of living organisms (Zinchenko *et al.* 2021; Al-Asaadi 2022), including bio mineralization process on the bivalve. As a result, it causes variation on its mineral compositions. The investigation on the geochemical compositions of bivalve shells may give information about the environmental change of this species during its life, including in the scallop (Yan *et al*. 2014). Scallops (*P. placenta*) are categorized as a filter feeder (Mondal *et al.* 2020). This species could absorb several minerals existed in the environment, and then accumulate the minerals at its shells and mussel (Yaqin & Fachruddin 2018). This study aimed to know the mineral compositions and ratio in the shell of scallops collected from two different fishing grounds in East Java Waters, Indonesia.

MATERIALS AND METHODS

Study area

This study was carried out in the eastern part of East Java Waters, Indonesia, from February to March 2019. A purposive sampling method was used to collect the specimen in this study. The sampling site was determined based on the highest population of *P. placenta* in the East Java area. Site 1 was located at Junganyar Socah (Bangkalan District; 07° 05' 32.12" S; 112° 41' 00.58" E), while Site 2 was located at Ujung Pangkah (Gresik District; 06° 53' 405 "S; 112° 31' 397" E; Fig. 1). The *P. placenta* sample was placed in a plastic bag, and kept in the laboratory until further analysis.

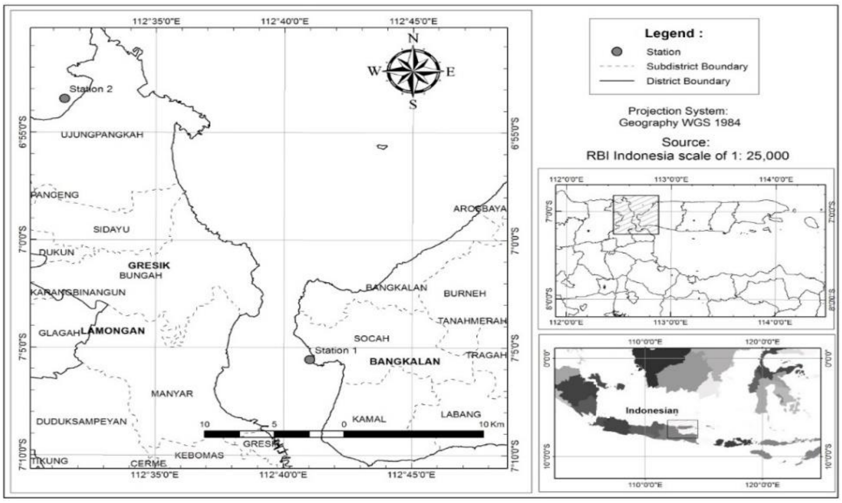

Fig. 1. Site of sampling in the eastern past of East Java waters: station 1 Junganyar Socah (Bangkalan District; 07° 05' 32.12" S; 112° 41' 00.58 "E) and station 2 Ujung Pangkah (Gresik District; 06° 53' 405" S; 112° 31' 397" E).

Procedures

Sample preparation

P. placenta samples were cleaned using tap water, and allowed to air dried. Sample was cleaned up to remove the attached organisms and detritus on its shell. Next, samples were labeled based on their site locations. A small piece of shell (near Umbo) was used for mineral type and concentration analysis.

SEM-EDX analysis

Incubation of the sample was carried out using 3% Glutaraldehyde solution for 30 min, and then was resuspended. Next, sample was cleaned up using 3× Phosphate Buffer Saline (PBS) solution, and was centrifuged three times at 2000 g for 5 min. The pellet yielded from the centrifugation process was then placed on a cover glass, and allowed to dry above the Bunsen burner. Then, a gradual dehydration was carried out using alcohol in different concentration (30%, 50%, 70%, 80%, 90%, and 96%). The last step included coating the sample using gold, and observation in the SEM-EDX equipment. Analysis of existing element in the Bivalve shells using EDX during this study was according to the study by Lakhsmana *et al.* (2017).

RESULTS AND DISCUSSION

Characteristics of *P. placenta* shell

The result showed that *P. placenta* was round in shape. It exhibited large shell size with symmetrical shape between the left and right valve. The shell was transparent and showed color gradation when exposed to the light. The outer part of the shell is known to be used as attaching medium by the attached organisms and detritus. The average size of *P. placenta* shell living in the Ujung Pangkah-Gresik and Junganyar Socah-Bangkalan was about 9 to 12 cm (Fig. 2).

Fig. 2. Shells of scallop (*P. placenta*) collected from the Ujungpangkah Gresik (Left) and Junganyar Socah Bangkalan (Right) Waters.

Characterization of element on the shell

Observation on the Scallop shells from both locations showed several mineral layers. The structure and layer of the scallops shell collected from the site location is shown at Figs. 3 - 4. The mineral structure of scallops collected from UjungPangkah-Gresik showed an agglomeration of several particles (Fig. 3), while the particle of those collected from Junganyar Socah-Bangkalan were spread to the edge of the shell (Fig. 4). The agglomeration and the spread of the particle may explain the quantities of the mineral to be observed.

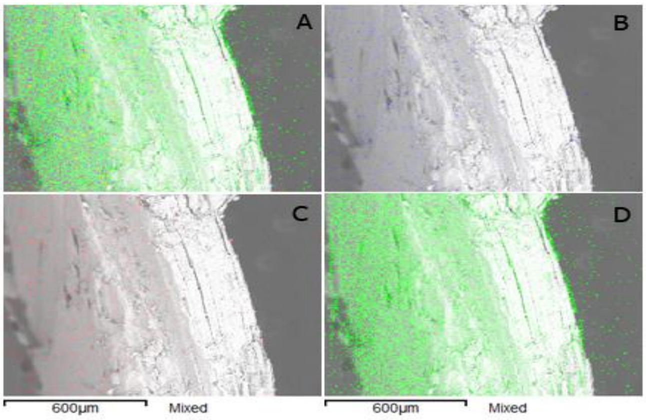

Fig. 3. SEM Micrograph and element surface distribution of Composite (A), Sulfur (B), Magnesium (C), and Calcium (D) on the scallops (*P. placenta*) shell collected from Ujung Pangkah-Gresik.

The type of minerals on the scallops shell was shown in different colors. In the case of the scallops collected from Ujungpangkah Gresik, calcium (Ca) was represented by green color, magnesium (Mg) by red, and sulfur (S) by blue. On the other hand, the minerals in the shell of scallops collected from Junganyar Socah-Bangkalan was represented by blue color for magnesium (Mg), red for aluminum (Al), and green for sulfur (S). The different dots in the shell show the composition of minerals in it. Magnesium, calcium, and sulfur are mainly known as macro minerals (Arifin 2008). Magnesium is the major mineral of sea water which is ionized into Mg^{2+} and present as magnesium chloride crystal (Abdel-Aal *et al.* 2017). The other mineral detected in the shell of scallop was calcium. The present of calcium in the scallop shell might be related to the calcium carbonate which is the main mineral composing the scallops shell (Kalesaran *et al.* 2018).

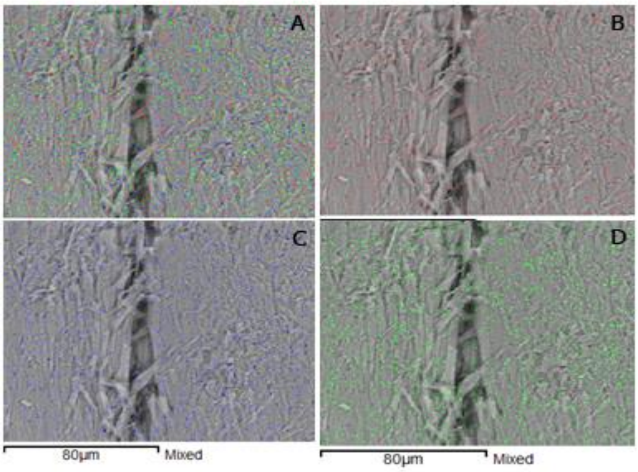

Fig. 4. SEM Micrograph and element surface distribution of composite (A), aluminum (B), magnesium (C), and sulfur (D) on the scallops (*P. placenta*) shell collected from Junganyar Socah-Bangkalan.

Element composition on the shell of *P. placenta* collected from Ujung Pangkah-Gresik and Junganyar Socah-Bangkalan

Element composition analysis on the bivalve shells may be determined by observing the transition result of X ray between the electron of certain energy level in an atom using voltage from 20 to 25 KeV (Lakhsmana *et al.* 2017). Observation on the mineral composition of the scallops (at 20 KeV) living in the eastern part of East Java Province showed that the highest intensity was attributed to calcium at X Mα = 3.80 keV (Fig. 5), then carbon at Mα = 3.70 keV, oxygen at X Mα = 5.98 keV, sodium at X Mα = 3.70 keV, magnesium at X Mα = 3.70 keV, aluminum at X Mα = 3.70 keV, silicate at X Mα = 3.70 keV, sulfur at X Mα = 3,70 keV, and chloride at X Mα = 3.70 keV (Fig. 5). The mineral composition of bivalve shells is known to be influenced by several factors, such as: species, habitat, age, size, region, and the environmental condition (Yan *et al.* 2014). The mineral content on the bivalves may be influenced by the water quality on their habitat (Nurjanah *et al.* 2013).

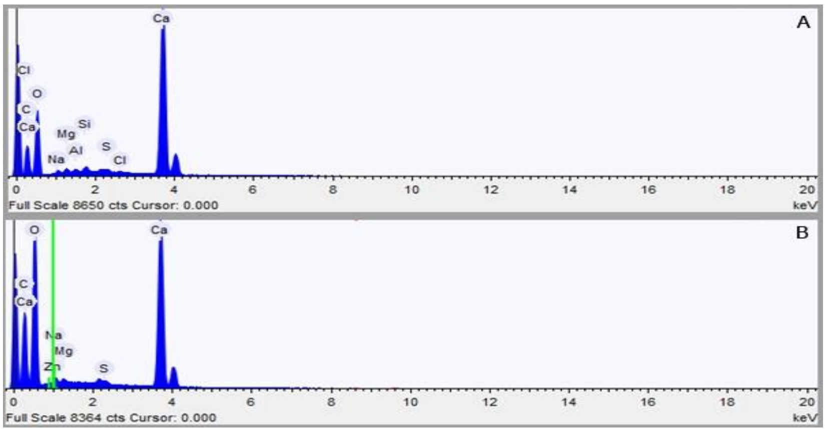

Fig. 5. Element spectrum of *P. placenta* shell collected from Ujung Pangkah-Gresik (A) and Junganyar Socah-Bangkalan (B).

The results showed that calcium (Ca) is the most abundant element on the *P. placenta* shell (Fig. 5). The shell of bivalve is commonly known to be composed of calcium carbonate ($CaCO_3$) and organic protein matrix. The most dominant mineral composing calcium carbonate are calcite and aragonite (Southgate & Lucas 2008). These minerals are grouped into essential mineral that plays important role on the formation of tissue and organ (Kalesaran *et al.* 2018). Calcium mineral on the shell could also be found in the form of calcium phosphate, that is commonly used to remediate wastes due to its adsorbent capacity (Onoda & Nakanishi 2012). The element composition on the *P. placenta* shell collected from the location showed different type and quantities. The results showed that there were 7 types of element on the shells collected from Ujung Pangkah-Gresik (carbon, oxygen, sodium, magnesium, calsium, zinc, and sulfur; Table 1). The highest mass weights were attributed carbon (11.911-18.083%), calcium (20.275-37.692%), and oxygen (49.511-60.689%). The high percentage of these element was obtained from the percent atomic value during analysis. The percentage of atom on the carbon, calcium, and oxygen were 19.610-25.768%, 8.658-18.597%, and 61.198-64.924%, respectively. In contrast to the element quantities in the Ujung Pangkah-Gresik, those from Junganyar Socah-Bangkalan was more abundant than those from the Ujung Pangkah-Gresik. There were 9 types of elements found on the shell, such as: carbon, oxygen, sodium, magnesium, calsium, silicon, sulfur, alumunium, and chlorine (Table 2). The highest percentages of element weight were attributed to carbon (11.432-14.037%), calcium (31.106-33.491%), and oxygen (51.584-53.863%), while the highest rates (%) of element atom were attributed to carbon (18.393-21.840%), calcium (14.504-16.269%), and oxygen (61.894-63.827%).

Table 1. Element composition of *P.placenta* shell collected from Ujung Pangkah-Gresik.

Result	Replication	Element						
		C	O	Na	Mg	Ca	Zn	S
	I	11.911	49,511	0,308	0,305	37,692	0,273	0,000
Weight (%)	II	17.705	59,122	0,446	0,263	22,359	0,000	0,105
	III	18.083	60,689	0,494	0,295	20,275	0,045	0,119
Average								
	I	0.250	0.379	0.077	0.043	0.301	0.161	0.000
Weight (%) □	II	0.220	0.250	0.047	0.033	0.148	0.000	0.029
	III	0.199	0.234	0,059	0.030	0.125	0.125	0.026
Average								
	I	19.610	61.198	0.265	0.248	18.597	0.082	0.000
Atomic (%)	II	25.589	64.146	0.337	0.188	9.684	0.000	0.057
	III	25.768	64.924	0.368	0.207	8.658	0.012	0.064
Average								

The element composition of *P. placenta* shell from both locations can be used as bio-inspired and biomimetic material in the environment. The element composition on the shell may describe the environmental condition, especially the water characteristic of the location where the species is living in (Anna *et al.* 2017). One of the ways is by calculating the element ratio on the shell of *P. placenta*.

Table 2. Element composition of P.placenta shell collected from Junganyar Socah-Bangkalan

Hasil	Replication	Element								
		C	O	Na	Mg	Ca	Si	S	Al	Cl
	I	11.432	52.843	0.504	0.505	33.491	0.561	0.144	0.295	0.225
Weight (%)	II	12.957	51.584	0.536	0.416	33.967	0.340	0.200	0.000	0.000
	III	14.037	53.863	0.411	0.357	31.106	0.000	0.226	0.000	0.000
Average										
	I	0.297	0.327	0.054	0.042	0.241	0.038	0.038	0.035	0.036
Weight (%) □	II	0.236	0.323	0.055	0.043	0.242	0.036	0.040	0.000	0.000
	III	0.228	0.300	0.054	0.041	0.216	0.000	0.038	0.000	0.000
Average										
	I	18.393	63.827	0.424	0.402	16.147	0.386	0.087	0.211	0.123
Atomic (%)	II	20.709	61.894	0.447	0.329	16.269	0.232	0.120	0.000	0.000
	III	21.840	62.916	0.334	0.275	14.504	0.000	0.132	0.000	0.000
Average										

The analysis showed that Na/Ca ratio on the *P. placenta* shells collected from Ujungpangkah were higher than those from Junganyar Socah. On the contrary, the Mg/Ca and S/Ca ratio of those collected from Junganyar Socah were higher than those from Ujungpangkah. Na/Ca ratio on the shell can be used as indicator to describe the salinity of the water that play important role on the shell crystallization phase (Dalbeck 2008). Thus, Mg/Ca and S/Ca ratio can be used as the indicator of temperature in the water.

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