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Occurrence of the hairy ark cockle (*Anadara gubernaculum*, Reeve 1844) in Mayangan coastal waters, Subang – Province West Java: new distribution record of Indonesia

Dewi Fitriawati[1]*, Nurlisa Alias Butet[2] and Yusli Wardiatno[2]

[1]*Master Program in Aquatic Resources Management, Graduate School of Bogor Agricultural University, Kampus IPB Dramaga – Bogor 16680, Indonesia*

[2]*Department of Aquatic Resources Management, Faculty of Fisheries and Marine Science, Bogor Agricultural University, Kampus IPB Dramaga – Bogor 16680, Indonesia*

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ABSTRACT

Thirty six specimens of the hairy ark cockle (*Anadara gubernaculum* Reeve, 1844) were collected from Mayangan waters, Subang of Province West Java, during marine biodiversity survey along northern coast of Java Island, Indonesia. Previous records of the cockle showed its occurrence in Cilincing, northern coast of Jakarta, Semarang coastal waters of Province Central Java, and Sidoarjo coastal waters located in northern part of Province East Java. The identification of the species was performed by morphological characters and is supported by molecular analysis. This finding represents a new distribution record of the species in Indonesia.

Keywords: Arcidae, bivalves, first record, mollusc, north Java.

INTRODUCTION

As an aquatic biodiversity hotspot, Indonesia has rich and diverse marine organisms, one of them is the cockles group of Arcidae. The family Arcidae is a large and diverse family of marine bivalve distributed worldwide in warm seas of tropical areas. *Anadara* is one of the genus of Arcidae which widely distributed in coastal areas of Indo-Pasific covering East Africa, Madagascar, Red Sea, Indian Ocean, India and Srilanka, Thailand, China Sea, Taiwan, Malaysia, Indonesia, Philippines, Papua New Guinea, Solomon Islands, New Caladonia, Northern Australia, Western and South-West Pasific, Andaman Sea, Hawaiian Islands, Sulu Sea, and Bay of Bengal (Wye, 1997). One species of cockles which is often used as human food is *Anadara gubernaculum* Reeve 1844.

Distribution of the hairy ark cockle *Anadara gubernaculum* was reported in Vietnam (Evseev & Lutaenko, 1998; Hylleberg, 2000), Porto Novo Africa (Jayabal & Kalyani, 1989), and in the coast of China (Feng *et al.*, 2011). This species lives at depth of 0.8-1.6 m in intertidal zone of Indo-Pacific waters (Evseev & Lutaenko 1998). In Indonesia, some previous studies had recorded the presence of this species in Cilincing, north coast of Jakarta (Dharma, 1998), Sidoarjo muddy beach of Province East Java (Ambarwati & Trijoko, 2011) and in the northewestern coast of Madura (Altena, 1945). Among fisherman communities living along northern coastline of Java Island the hairy cockles is one of target species especially for muddy intertidal fishery activities. The meat is used as food and the shell is for handycraft. This paper reported the first record of A.*gubernaculum* from Mayangan waters, Subang – Province West Java, Indonesia. At the same time it is a new distribution record of the species in Indonesian waters.

MATERIAL AND METHODS

Thirty six specimens of *A. gubernaculum* were collected from Mayangan waters, Subang – Province West Java in June 2015, and were preserved in 96% ethanol. Figure 1 shows the sampling location where the specimens were collected. The collected specimens were transported to the laboratory for identification using morphological characters and molecular analysis. The specimens were deposited in collection room of the Department of Aquatic Resources Management, Bogor Agricultural University, Indonesia. Morphological characters identification was performed using the taxonomic keys from Food and Agriculture Organization of the United Nations (FAO) (1998). One specimen is showed in Figure 2.

To ensure the identification, molecular analysis was conducted using mitochondrial cythochrome subunit I (COI) gene sequences. DNA was extracted from foot muscle tissue, following the protocol of GENE AID DNA Extraction Kit. The DNA product was amplified using Polymerase Chain Reaction (PCR) using the COI primer pair designed by Folmer *et al.* 1994. About 700 base pairs of the COI gene were amplified using reagents from KAPPA 2G Readymix Hotstart. The PCR was performed in a volume 50 µL for each sample with the reagent proportions 9 µL of ddH_2O, 25 µL of Buffer

*Corresponding Author's E-mail: dewi.fitria26@gmail.com

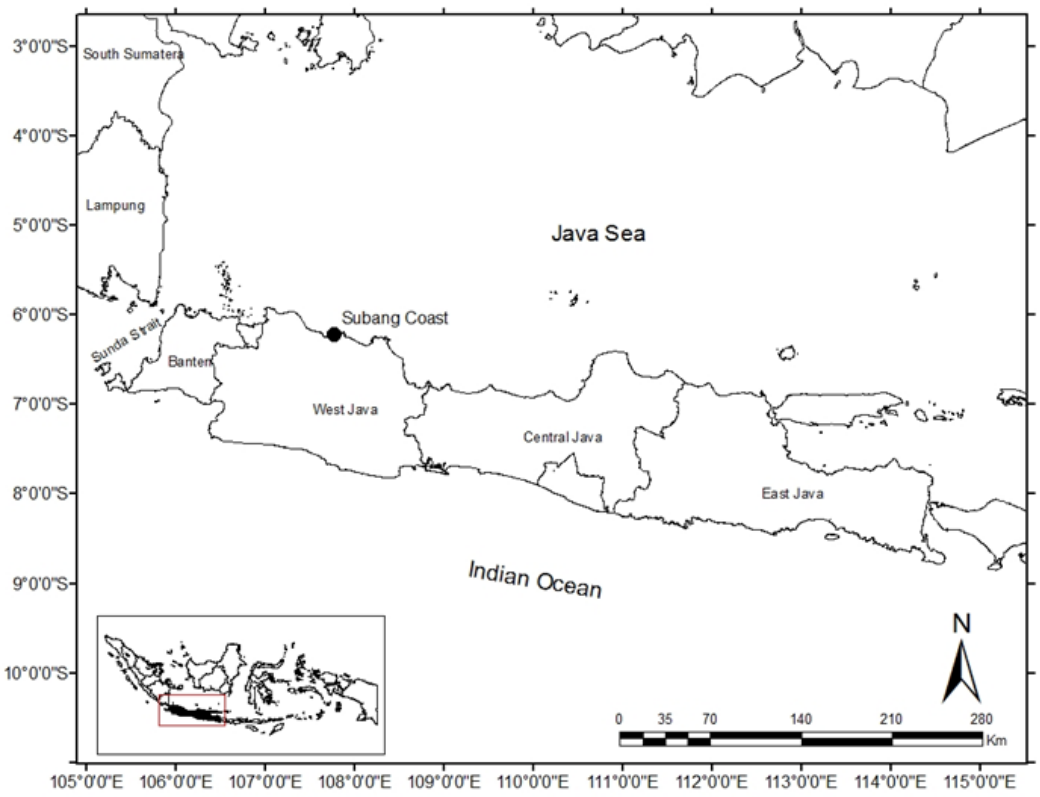

Figure 1. A map of sampling location in Mayangan, Subang – Province West Java as indicated by the black circle.

Taq DNA Polymerase, 1.5 µL of each primer (10 pM), and 10 µL of DNA template. The temperature profile of PCR consist of initial denaturation at 95°C for 3 min, followed by 35 cycles at 95°C for 45s, 50°C for 1 min, 72°C for 1 min, with final extension of 5 min at 72°C. The amplified fragment was visualized on 1.2% agarose gell stained with ethidium bromide. Succesful PCR product was sequenced following standard protocol. The obtained sequences was aligned using MEGA 5 (Tamura *et al.* 2011), followed by BLAST for identification on GenBank. The COI sequences also aligned with other bivalves COI gene from GenBank to construct the phylogenetic tree using Neighbor-Joining method.

Figure 2. A specimen of *Anadara gubernaculum* collected from Mayangan waters, Subang – Province West Java, Indonesia.

RESULTS

Systematics

Class: Bivalvia
Order: Arcida
Family: Arcidae
Subfamily: Anadarinae
Genus: *Anadara*
Species: *Anadara gubernaculum* Reeve 1844

Synonym name: *Scapharca gubernaculums*

Locality: Mayangan Waters, Subang, Indonesia

Description

Shell solid, thick, heavy, laterally compressed and subrectangular in shape, slightly inequivalve, left valve somewhat overlapping the right valve on posteroventral margin. Umbo flat, not tappered, and situated anteriorly. About 22-30 radial ribs at each valve, ribs narrow and flat without granules, wider than the interribs (Fig 3b). Ligament area narrow with a long atraight hinge bearing a row of many toxodont teeth (Fig 3d). Thick periostracum, and there is adventitious hairs derived from periostracum in edge of shell. Colour: outside of shell whitish under blackish brown periostracum, inner side white.

Molecular Identification

The total genomic DNA was succesfully isolated and extracted from foot muscle of *A. gubernaculum.* Total DNA with good quality was using as a template on PCR process. About 700 bp of COI gene of *A. gubernaculum* was succesfully amplified (Figure 4).

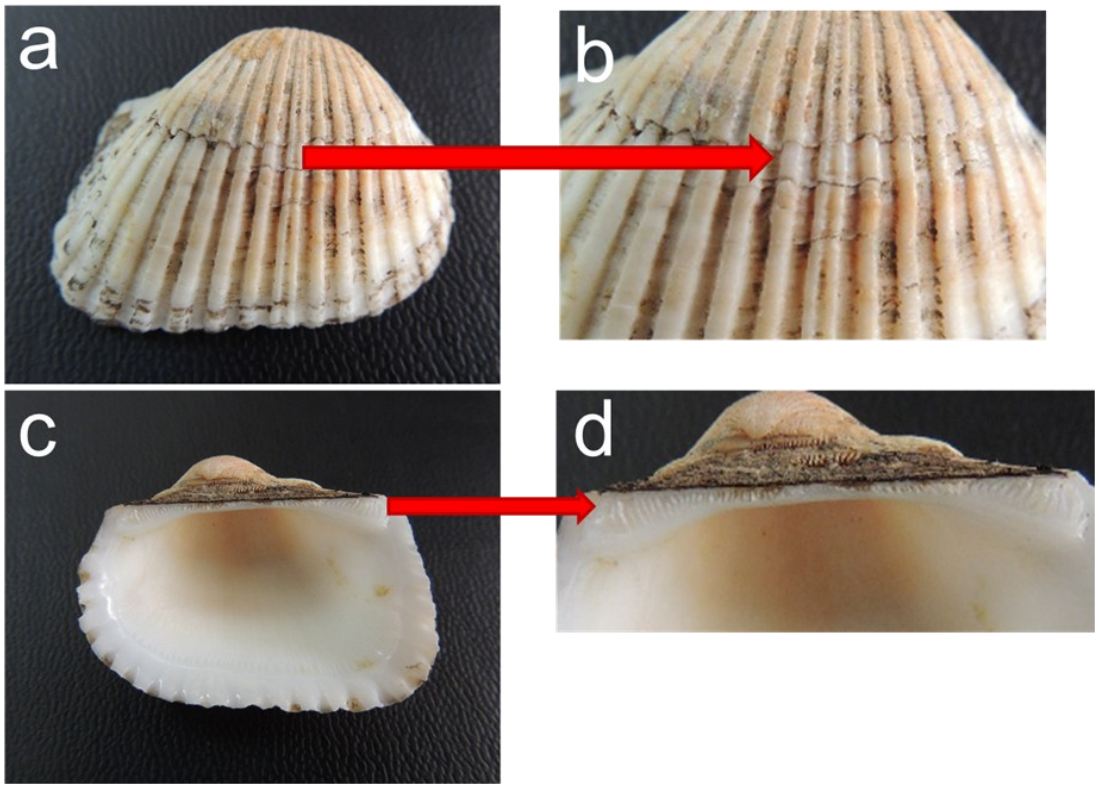

Figure 3. *Scapharca gubernaculum* (Reeve 1844) of Mayangan waters, Subang – Province West Java, Indonesia: a. exterior of shell, b. magnified portion of periostracum, c. interior of shell, and d. hinge area with toxodont teeth.

The sequences result was aligned with another bivalves COI gene sequences for phylogenetic tree analysis. COI gene sequences used in constructing phylogenetic tree obtained from GenBank, consisting of a sequence of the same species (*Scapharca gubernaculum* HQ258857.1), genus *Barbatia*, *Pinctada*, and *Tridacna* (Figure 5). The Neighbor-Joining tree showed that *A. gubernaculum* from this study and *S. gubernaculum* (HQ258857.1) from GenBank has the closest genetic distance so they have the closest branch. There were four subclades in this phylogenetic tree, consist of *Anadara* or *Scapharca* clade, *Barbatia* clade, *Pinctada* clade and the last *Tridacna* clade. Furthermore, genus *Anadara/ Scapharca* and *Barbatia* included in the same big clade, since both of them come from the same family, Arcidae.

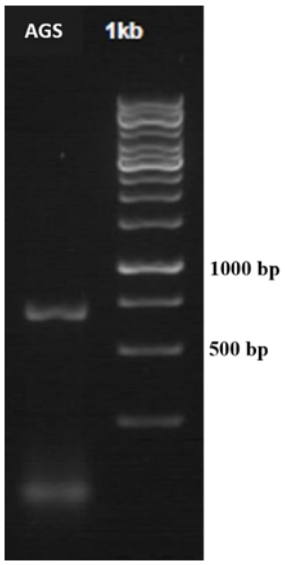

Figure 4. PCR product of COI gene of *Anadara gubernaculum* originated from Mayangan waters, Subang, Indonesia. AGS stands for *A. gubernaculum* Subang.

DISCUSSION

About 300 species are estimated in the order Arcoidea, and is dominated by family Arcidae (Oliver and Holmes 2006). Indo-Pacific is a region which has the highest species richness of this group. There are about 38 species of *Anadara* which live in this region (Lutaenko 2011), consists of *Anadara antiquata* (L.), *A. uropigimelana* (Bory), *A. crebricostata* (Reeve), *A. auriculata* (Lamarck), *A. craticulata* (Nyst), *A. ehrenbergi* (Dunker), *A. ambigua* (Reeve), *A. ferruginea* (Reeve), *A.dautzenbergi* (Lamy), *A. africana* (Sowerby), *A. pygmaea* (H. Adams), *A. consociata* (Smith), *A. vellicata* (Reeve), *A. pilula* (Reeve), *A. binakayanensis* (Faustino), *A. satowi* (Dunker), *A. cornea* (Reeve), *A. kagoshimensis* (Tokunaga), *A. natalensis* (Krauss), *A. troscheli* (Dunker), *A. kafanovi* Lutaenko, *A. gubernaculum* (Reeve), *A. guangdongensis* (Bernard, Cai et Morton), *A. broughtonii* (Schrenck), *A. inaequivalvis* (Bruguiere), *A. rufescens* (Reeve), *A. granosa* (L.), *A. rhombea* (Born), *A. nodifera* (Martens), *A. indica* (Gmelin), *A. erythraeonensis* (Jonas in Philippi), *A. mosambicana* (Bianconi), *A. kikaizimana* (Nomura et Zinbo), *A. deyrollei* (Jousseaume), *A. trapezia* (Deshayes), *A. addita* Iredale, *A.* secticostata (Reeve), A. jurata Iredale.

Indonesia which has a total of 81.000 km of coastline with variation depth providing an ideal habitat

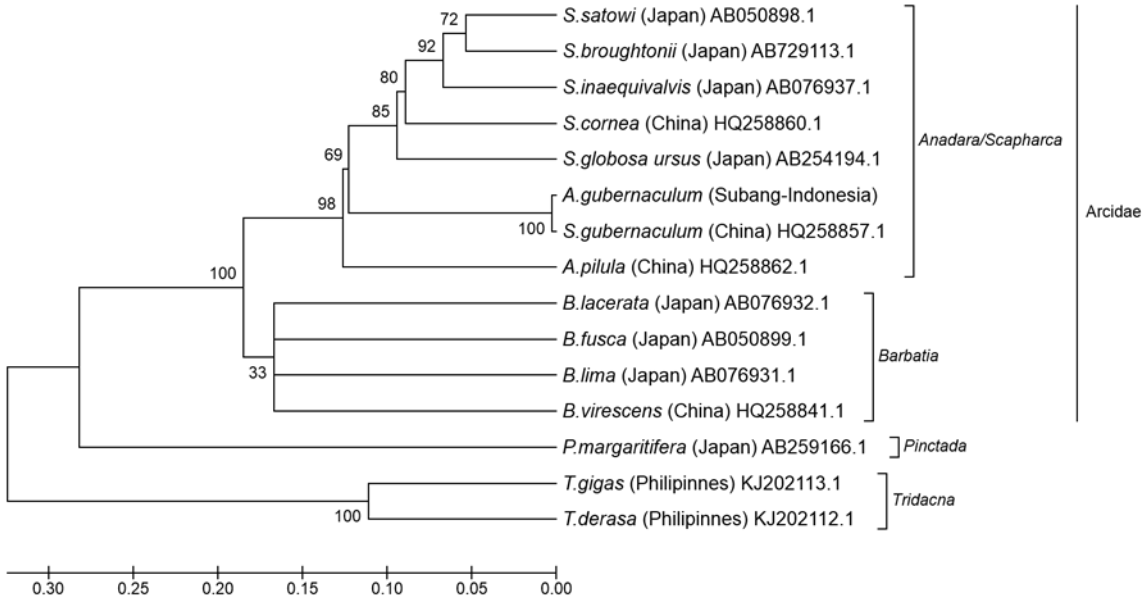

Figure 5. Phylogenetic tree based on COI gene of some bivalves using Neighbor-Joining method.

for a wide variety of molluscs. Indonesian molluscs are compiled in Dharma (1998), and some cockles are included in his publication, such as *Anadara granosa, Perna viridis, Scapharca inequivalvis* and *A. gubernaculum*. Some other species of cockles were reported to occur in Indonesian waters as reported in various researches, e.g. *Clinocardium nuttalli* in Banda Aceh (Husin et al., 2013), *A. antiquata* (Andayani and Sumartono, 2012), *A. pilula* (Qonita et al., 2015), *A. rufescens,* and *A. nodifera* (Ambarwati and Trijoko, 2010).

The biology of cockle has been themes of recent studies in Indonesia, *e.g*. exploration of *actin* gene expression and characteristic from *A. granosa* due to mercury pollution (Butet *et al.*, 2014), depuration ability of *A. granosa* to eliminate microorganisms and toxic chemical contaminants (Ningrum *et al.*, 2016), saxitoxin produced by *A. granosa* and *A. antiquata* (Andayani and Sumartono, 2012), morphological variations of *A. pilula* from different locations (Qonita *et al.*, 2015), etc.

In terms of morphology, there is morphological complexity in cockles species render them look similar inter species (Dixon *et al.*, 1995; Vongpanich, 1996). However, molecular analysis using COI allowed us to identify *A.gubernaculum* clearly. The molecular analysis showed that our specimen matches with *S. gubernaculum* (HQ258857.1) at similarity levels 99% from the collection of GenBank database. As informed above *S. gubernaculum* is a synonym name of *A. gubernaculum*. The phylogenetic tree also showed that *A. gubernaculum* from this study was very close to *S. gubernaculum* from GenBank. The clear separation between different genus (*Anadara*/ *Scapharca*, *Barbatia*, *Pinctada*, and *Tridacna*) also proved that the obtained samples are truly comes from the genus *Anadara*, namely *A. gubernaculum.*

The occurrence of *A. gubernaculum* in Subang waters, Province West Java increases their distribution information in Indonesian waters. This finding enhances and enriches the list of new distribution records of Indonesian marine species. Some new distribution records of Indonesian species were reported, e.g. hippoid crabs: *Albunea symmista* (Mashar et al., 2015), *Hippa marmorata* (Wardiatno et al., 2015), and *Hippa adactyla* (Ardika et al., 2015); palinurid lobster: *Puerulus mesodontus* (Wardiatno et al., 2016a); nephropid lobster: *Metanephrops andamanicus* (Wardiatno et al., 2016b), and scyllarid lobster: *Thenus indicus* and *Scyllarides haanii* (Wardiatno et al., 2016c); etc. These findings are very important in biological conservation point of view. The biological information of *A. gubernaculum* in the study location, e.g. population dynamics, biological reproduction, stock, etc., is needed for future investigation to ensure its sustainable use.

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