International Journal of Aquatic Biology (2014) 2(6): 299-304
ISSN: 2322-5270; P-ISSN: 2383-0956
Journal homepage: www.NPAJournals.com

Original Article

Spawning season and larval occurrence of blood cockle (*Anadara granosa*) off the Selangor coast, Peninsular Malaysia

Tatsuya Yurimoto*[1], Faizul Mohd Kassim[2], Alias Man[3], Reiko Fuseya[4]

[1]Japan International Research Center for Agricultural Sciences, Tsukuba, Ibaraki, Japan.
[2]Penang Office of Japan International Research Center for Agricultural Sciences, Penang, Malaysia.
[3]Fisheries Research Institute, Department of Fisheries, Penang, Malaysia.
[4]National Research Institute of Fisheries Engineering, Fisheries Research Agency, Kamisu, Ibaraki, Japan.

Abstract: We performed a plankton survey of blood cockle larvae, *Anadara* spp., and histological observations of the gonads of adult blood cockle, *Anadara granosa*, in the coastal waters off Sungai Buloh, Peninsular Malaysia from October 2010 to April 2011. The histological observations indicated the main spawning period in both sexes was from November to March. In addition, the occurrence of blood cockle larvae peaked in November and March. Furthermore, a comparison of the distribution between umbo-stage and full-grown larvae suggested the umbo-stage larvae diffused offshore and return to coastal sites where they settle and mature into full-grown larvae.

Article history:
Received 20 April 2014
Accepted 25 October 2014
Available online 25 December 2014

Keywords: Blood cockle, Spawning season, Larva, *Anadara granosa*, Malaysia.

Introduction

Blood cockle, *Anadara granosa*, is one of the most popular bivalves for sowing aquaculture in Southeast Asia and the Selangor coast off Peninsular Malaysia (Pathansali and Song, 1958; Broom, 1985). Regional fishermen collect natural spats of blood cockle in tidal flats of the Selangor coast and cultivate them by sowing culture. Therefore, elucidating the current status of blood cockle reproduction in the Selangor coast is important for ensuring the sustainable production of this species in this region. However, information about the relationship between larval occurrence and the spawning period of blood cockle is insufficient, preventing fisheries management from achieving sustainable production. Therefore, this study evaluated the main spawning season of blood cockle, performed histological observations of the gonads, and monitored the density of bivalve larvae including blood cockle larvae at 4 stations of the Selangor coast. The results will help establish a monitoring system for blood cockle larvae.

Materials and methods

Field survey: A total of 4 sampling surveys at Sungai Buloh off the west coast of Peninsular Malaysia were carried from October 2010 to April 2011 (Fig. 1). Four survey stations (stations 1-4) were set along the coast and the adult blood cockles were collected with a dredge of 1.5 cm mesh. At these 4 stations, bivalve larvae samples were collected with a zooplankton net (diameter: 30 cm (0.07 m^2), side length: 100 cm, netting: NXX13 (0.1 mm mesh opening)) by vertical hauling from the seabed to surface. The average±SD depth at stations 1-4 during the survey period was 2.3 ± 1.1, 4.0 ± 1.5, 7.4 ± 1.4, and 5.2 ± 1.4 m, respectively. Water temperature, salinity, the dissolved oxygen concentration in bottom-layer water (~20 cm above the seabed), and the average concentration of chlorophyll *a* in the water column of station 1 were measured with a water quality

* Corresponding author: Tatsuya Yurimoto
E-mail address: yurimoto@outlook.com

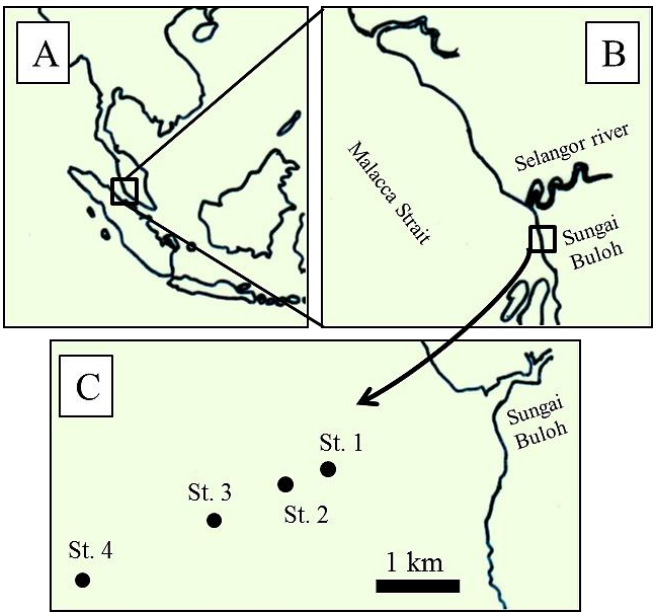

Figure 1. Sampling location of adult blood cockle and zooplankton at (C) Sungai Buloh (B) off the Selangor coast, (A) Peninsular Malaysia. Station 1, adult cockle sampling site; Stations 1–4, zooplankton sampling sites.

sensor (AAQ-RINKO, JFE Advantech Co., Ltd., Japan). In addition, during the surveys in November and March, when blood cockle larvae peaked, the vertical gradients of water temperature and salinity in 4 stations (except station 2 in November) were measured to determine the relationship between water mass and the distribution of blood cockle larvae.

Gonad observation: Adult blood cockles (*A. granosa*) collected from the survey field were transferred to a temporary laboratory. Then the shell length measurement and gonadal tissue was removed. Tissue blocks fixed in 10% seawater formalin solution were substituted in 100% lemosol after alcohol dehydration and embedded in paraffin. Sections 7-10 μm thick were observed under a light microscope after hematoxylin–eosin staining. Histological observations of the gonads were performed to determine sex and maturity according to 5 stages: the immature, developing, mature, spawning, and spent stages—to determine the main spawning period. These stages were determined on the basis of gonad observation in blood cockle, *A. granosa* (Yurimoto et al., 2014).

Larval observation: The morphology of fresh larvae samples collected from the field was identified based on Yoshida (1964) and Muthiah et al. (1992), and the numbers of individuals were counted under a light microscope. The items counted were the number of total bivalve larvae, bivalve larvae excluding blood cockle larvae, blood cockle larvae (*Anadara* spp.; individuals between the umbo and full-grown stages at the genus level), and umbo-stage and full-grown blood cockle larvae (*Anadara* spp.). In addition, the density of individuals in the water column, sampled using a zooplankton net, was determined.

Results

Environment: Water temperature at station 1 ranged from 29.8-30.9°C (Fig. 2A). The temperature increased gradually after a nadir at the early of February. On the other hand, salinity decreased from 30.3 to 29.1 PSU during October and November (Fig. 2B) and subsequently increased to 29.7 PSU in April. The concentration of dissolved oxygen remained at 2.7 mg/L throughout the survey period (Fig. 2B). Meanwhile, the concentration of chlorophyll *a* increased to 10 μg/L over on average in November and March.

Gonad condition: The average shell length of the blood cockles collected during the survey period was 29.1 ± 3.5 mm (n = 92), and the male:female ratio was 0.47 : 0.53. Regarding sexual maturation, males were mainly classified into the developing and mature stages in October, the mature and spawning stages from November to March, and the spawning and spent stages in April (Table 1). On the other hand, females were mainly classified into the mature stage from October to November; the spawning stage from February to March; and the developing, spawning, and spent stages in April.

Larval occurrence: The greatest density of bivalve larvae occurred at station 2, ranging from 425-431/m^3 during November and February; the density peaked at 1,351/m^3 in March and decreased rapidly to 381/m^3 in April (Fig. 3A). Station 3 had the next highest density, ranging from 245-624/m^3. In addition, the densities at stations 1 and 4 fluctuated but remained low; the peak densities of stations 1 and

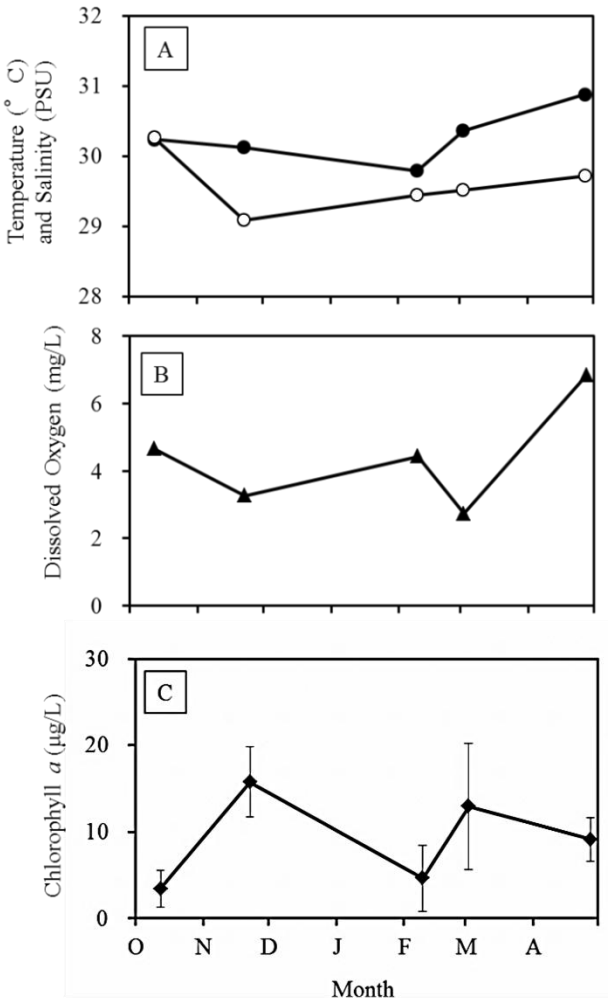

Figure 2. Environmental changes at station 1, Sungai Buloh. (A, ●) water temperature, (A, ○) salinity, (B, ▲) dissolved oxygen in the bottom layer (~20 cm from the bottom), and (C, ◆) average concentration of chlorophyll *a* in the water column; error bars indicate standard deviation.

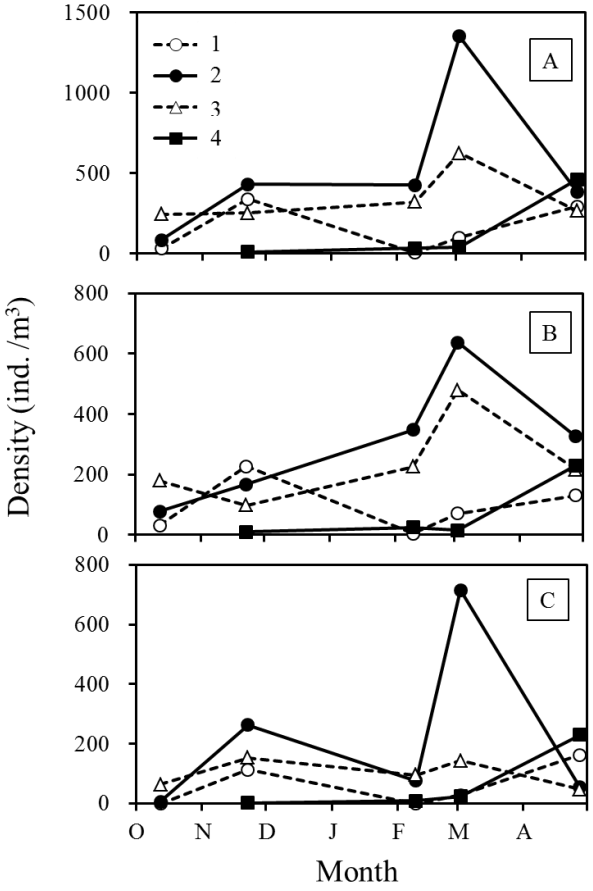

Figure 3. Changes in the density of (A) bivalve larvae, (B) bivalve larvae except blood cockle larvae, and (C) blood cockle larvae (*Anadara* spp.) from the umbo stage to the full-grown stage.

4 were 340/m^3 in November and 461/m^3 in April, respectively. The greatest density of bivalve larvae excluding blood cockle larvae occurred at station 2, peaking at 637/m^3 in March. This was followed by 480/m^3 at station 3 in March (Fig. 3B). Similar to block cockle larvae, the densities at stations 1 and 4 fluctuated but remained low, showing small peaks in November and April, respectively. In addition, high densities of blood cockle larvae occurred at station 2, increasing from 6/m^3 in October to 264/m^3 in November; the low density in February increased again to 714/m^3 in March and decreased to 54/m^3 in April (Fig. 3C). The densities at stations 1, 3, and 4 fluctuated but remained low: 0-163, 48-154, and 2–230/m^3, respectively. In addition, blood cockle larvae were divided into 2 stages - umbo-stage and full-grown larvae - to determine the differences in the distributions of each stage (Fig. 4).

The highest density of umbo-stage larvae occurred at station 2, peaking at 225 and 669/m^3 in November and March, respectively. The densities at the other stations fluctuated below 216/m^3 (Fig. 4A). In addition, the density of full-grown larvae increased at stations 1, 2, and 3 in November and station 2 in March; the densities at those sites ranged from 14-47/m^3 (Fig. 4B). Next, we calculated the mean ± SE densities of umbo-stage and full-grown larvae at each station. The mean densities of umbo-stage larvae at stations 1, 2, 3, and 4 were 49 ± 28 (n = 5), 206 ± 108 (n = 5), 90 ± 17 (n = 5), and 61 ± 45/m^3 (n = 4), respectively (Fig. 5). In contrast, densities of full-grown larvae at stations 1, 2, 3, and 4 were 12 ±

Table 1. Sexual maturation of blood cockle (*Anadara granosa*) collected at station 1 off the Selangor coast, Peninsular Malaysia.

Month	*n*	Unknown	Male				Female			
		I	II	III	IV	V	II	III	IV	V
October	18		2	7				9		
November	20			8	2			10		
February	18			3	4				11	
March	18			3	5				10	
April	18			1	6	2	2		2	5

n, number of samples per month; I, immature stage; II, developing stage; III, mature stage; IV, spawning stage; V, spent stage.

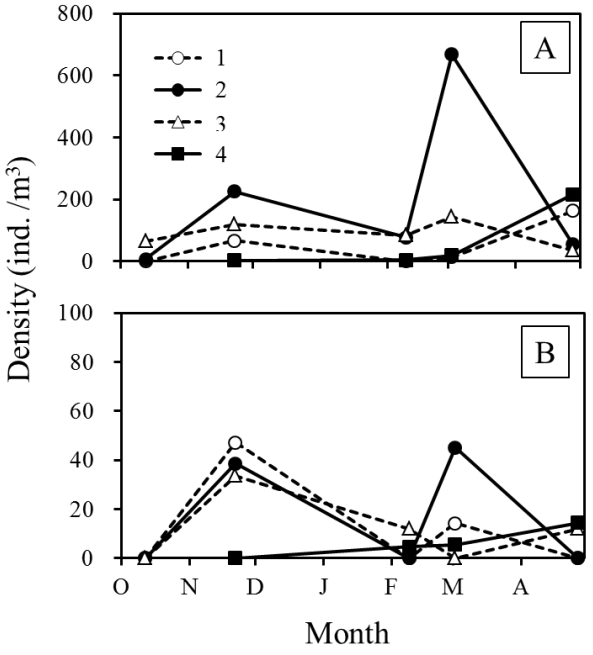

Figure 4. Density changes of (A) umbo-stage and (B) full-grown larvae of blood cockle (*Anadara* spp.).

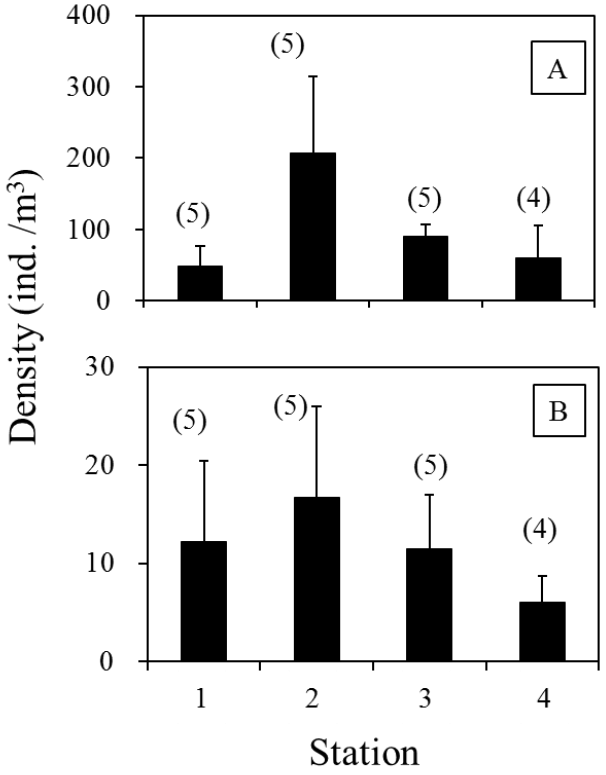

Figure 5. Comparison of average densities of (A) umbo-stage and (B) full-grown blood cockle larvae at each station in Sungai Buloh. Error bars indicate standard error; numbers in parentheses indicate numbers of samples.

8 (n = 5), 17 ± 9 (n = 5), 12 ± 5 (n = 5), and 6 ± 3/m^3 (n = 4), respectively; the highest and lowest densities were at stations 2 and 4, which were coastal and offshore sites, respectively.

Finally, we analyzed vertical gradients of water temperature and salinity in November and March, when the density of blood cockle larvae was high. However, no thermocline or halocline was observed in either month. Water temperature and salinity decreased gradually with increasing depth at the coastal and offshore sites (Fig. 6A-D).

Discussion

Environment and sexual maturation: In culture experiments, *A. granosa* grew well in Kakinada Bay, India, in which water temperature and salinity ranged from 27.8-33.5°C and 13.69-34.40 PSU, respectively, and dissolved oxygen levels were kept at 4.45 mg/L (Narashimham, 1983). In addition, salinity affects the survival of blood cockle at <20 PSU in the laboratory (Davenport and Wong, 1986). However, little is known about how low dissolved oxygen concentrations affect blood cockle. The LC_{50} of many bivalves is <2 mg/L (Raquel and Carlos, 2008). In the present survey, water temperature and salinity ranged from 29.8-30.9°C and 29.0-30.3

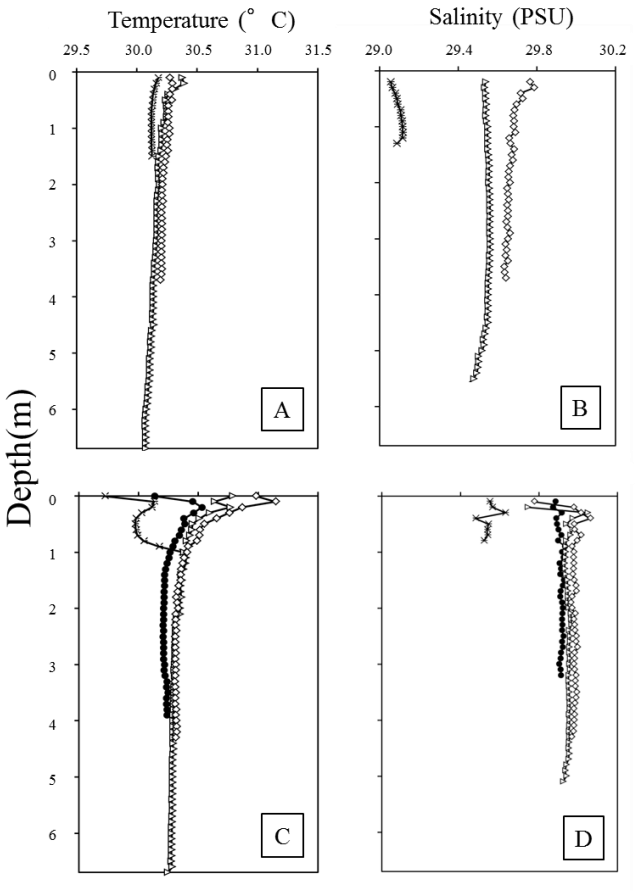

Figure 6. Vertical gradients of (A, C) water temperature and (B, D) salinity in the survey stations in (A, B) November and (C, D) March. station 1, ×; station 2, ●; station 3, △; station 4, ◇.

PSU, respectively, and the dissolved oxygen concentration exceeded 2.7 mg/L. Therefore, the environment of the present survey area is considered suitable for blood cockle culture.

Mature blood cockle mainly occurs along the Selangor coast from September to April (Broom, 1983). In the present study, mature cockles of both sexes were observed in October and spawning-stage individuals occurred from November to March; thus, the spawning period of blood cockle is long, similar to that reported by Broom (1983).

In addition, the chlorophyll *a* concentration in the water column at station 1 increased in November and March. The densities of blood cockle larvae and the number of adults in the mature and spawning stages increased simultaneously. These results suggest food availability is closely related to the larval survival and sexual maturation of adult blood cockle.

Planktonic larva: The occurrence of bivalve larvae was similar to that of blood cockle larvae. The density of bivalve larvae increased at stations 2 and 3 in March, and that of blood cockle larvae also increased at station 2 from February to March. These results suggest the spawning of other kinds of bivalves as well as blood cockle peak in March off the Selangor coast. In addition, a high density of blood cockle larvae was observed in November. These results show blood cockle larvae occur for a long period in this region. Histological observations of adults showed the gonads of many individuals had reached the mature and spawning stages from November to March corresponding well with the occurrence of blood cockle larvae. Furthermore, when blood cockle larvae were divided into the umbo and full-grown stages, umbo-stage larvae composed more half of all larvae. However, as the numbers of full-grown larvae peaked in November and March, blood cockle may have multiple settlement peaks in a year.

In an experiment, the planktonic period of a blood cockle larva from hatching to settlement was approximately 18 days; furthermore, it took 14-16 days for them to become full-grown larvae (Muthiah et al., 1992). In the present study, umbo-stage larvae were distributed mainly at stations 2 and 3, and were distributed more in offshore areas than station 1, where more adult cockles were found. On the other hand, low density full-grown larvae were collected from station 4, an offshore site, because the center of distribution was at stations 1 and 2. These results indicate that after spawning at the coastal sites, blood cockle larvae are distributed to offshore sites before returning to coastal sites where they become full-grown larvae. Then, the blood cockle larvae settle around muddy tidal flats where they live as adults. The present results support this notion, as the total average density at each station during the survey period showed there were high densities of umbo-stage larvae at stations 2 and 3, and full-grown larvae at stations 1 and 2. Regarding water mass structure in November and March, when blood cockle larvae peaked, larvae were mainly distributed in mixing zones between fresh water and offshore water. Thus,

these results suggest blood cockle larvae are mainly distributed in water masses influenced by fresh waters (i.e., low-temperature and/or low-salinity water) to prevent improper dispersion due to influence of offshore water. The relationship between water mass and the distribution of bivalve larvae, such those of clams and oysters, is well-known (Matsumura et al., 2001; Hofmann et al., 2004; Sakurai and Nakao, 1996). In the case of blood cockle larvae living in brackish waters, the present results also suggest an association between water mass structure and larval distribution.

References

Broom M.J. (1983). Gonad development and spawning in *Anadara granosa* (L.)(Bivalvia: Arcidae). Aquaculture, 30: 211-219.

Broom M.J. (1985). The biology and culture of marine bivalve molluscs of the genus *Anadara*. WorldFish, ICLARM Studies and Reviews, 12: pp. 37.

Davenport J., Wong T.M. (1986). Response of the blood cockle *Anadara granosa* (L.) (Bivalvia: Arcidae) to salinity, hypoxia and aerial exposure. Aquaculture, 56: 151-162.

Hofmann E.E., Powell E. N., Bochenek E. A., Klinck J.M. (2004). A modelling study of the influence of environment and food supply on survival of *Crassostrea gigas* larvae. ICES Journal of Marine Science, 61: 596-616.

Matsumura T., Okamoto S., Kuroda N., Hamaguchi M. (2001). Temporal and spatial distributions of planktonic larvae of the clam *Ruditapes philippinarum* in Mikawa Bay; application of an immunofluorescence identification method. Japanese Journal of Benthology, 56: 1-8.

Muthiah P., Narashimhan K.A., Gopinathan C.P., Sundarajan D. (1992). Larval rearing spat production and juvenile growth of the blood clam *Anadara granosa*. Journal of the Marine Biological Association of India, 34: 138-143.

Narasimham K.A. (1983). Experimental culture of the blood clam *Anadara granosa* (Linnaeuas) in Kakinada Bay. Proceedings of the Symposium on Coastal Aquaculture, Part 2: pp. 551-556.

Pathansali D., Song M.K. (1958). Some aspects of cockle (*Anadara grunosa* L.) culture in Malaya. Proceedings of the Indo-Pacific Fisheries, pp. 26-31.

Raquel V.S., Carlos M.D. (2008). Thresholds of hypoxia for marine biodiversity. Proceedings of the National Academy of Sciences, 105: 15452-15457.

Sakurai I., Nakao S. (1996). Distribution of planktonic larvae of the Japanese surf clam, *Pseudocardium sachalinensis* at Tomakomai, Hokkaido. Suisanzoshoku, 44: 17-23.

Yoshida H. (1964). Seedling of shellfish, Hokuryu-Kan co., ltd. Tokyo, pp.221.

Yurimoto T., Mohd Kassim F., Man A. (2014). Sexual maturation of the blood cockle, *Anadara granosa*, in Matang mangrove estuary, peninsular Malaysia. International Journal of Aquatic Biology, 2: 115-123.