Effect of culture duration on layer thickness and pearl quality of winged pearl oyster *Pteria penguin* (Bivalvia: Pteriidae) in Palabusa waters, Buton Strait, Southeast Sulawesi

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Abstract. The aim of this study was to determine the layer thickness and the pearl quality of winged pearl oyster *Pteria penguin* under different culture duration in Palabusa waters, Buton Strait, Southeast Sulawesi. A total of 120 pearl oysters with dorso-ventral height of 95.70±9.42 mm, width of 115.45±10.05 mm width, thickness of 31.92±3.15 mm, and weight of 105.71±17.21 g were implanted with a hemispherical plastic nuclei (12 mm diameter and 10 mm height) in the left valve by using a glue without anaeshetized and then cultivated for 6 months. The pearls were harvested at different culture duration after implantation; 90 days (treatment A), 120 days (treatment B), 150 days (treatment C) and 180 days (treatment D). The thickest top layer was obtained in the treatment D (0.264±0.013 mm) while the thinnest top layer was obtained in the treatment A (0.156±0.011 mm). The thickest base layer was also obtained in the treatment D (0.543±0.011 mm) and the thinnest base layers was obtained in treatment A (0.199±0.015 mm). The highest quality of pearls at AAA grade was obtained from the treatment D (28.6%), followed by treatment C (13.0%), treatment B (4.5%), and treatment A (0%). The lowest grade, non commercial grade (NC), was highest in treatment A of 37.0% followed by treatments B 18.2% and treatment C of 17.4%, and the lowest in treatment A by 4.8%. This study suggests that the culture duration of 6 months increases the thickness and quality of winged pearl oyster *P. penguin* cultured in the Palabusa waters.
Key Words: aragonite layers, cultivation period, mabe pearls, *Pteria penguin*.

Introduction. Pearl oyster from the genus *Pteria* (*Pteria penguin*) are commonly cultivated by communities both in groups and individuals to produce semi-round pearls or commonly called "mabe" and handicrafts made from raw shells in the form of blisters with varied forms (Ellis & Haws 1999; Mohamed et al 2005; Haws et al 2006; Teitelbaum et al 2008; Chand et al 2011). The method for producing mabe pearls is easier than round pearls. Also the cultivation of mabe pearl can be easily taught and adopted by rural communities compared to cultivation of round pearls that is difficult and costly. In addition, the harvesting of mabe pearls after implantation was relatively short compared to round pearls (Mohamed et al 2005; Haws et al 2006).

The successful pearl production including mabe pearl depends on the percentage of survival during harvesting, the number of implanted nuclei and the quality of pearls (Cartier et al 2012; Demmer et al 2015; Jagadis et al 2015). The quality of pearls depends on the surface layer of the nacre. Nacre is formed from hundreds to thousands of aragonite crystals (Zhang & Xu 2013) which grew in a polygonal form (generally hexagonal) with an average diameter of 0.5 µm and covered the nucleus (Taylor & Starck 2008) with a thickness of 0.29-0.6 µm (Alagarswami 1991), where each layer consists of 3-10 nm of crystal particles (Rosseau et al 2005).

Pearl quality is graded based on several criteria including shape, luster, surface contour, color, size and thickness of nacre (Alagarswami & Dharmaraj 1984; Goebel & Dirlam 1989; James et al 1992; Victor 1995; Wada & Komaru 1996; Chellam et al 1998; Ellis & Haws 1999; Haws 2002; Victor 2005; Taylor & Starck 2008; Cariño & Monteforte 2009). Criteria for pearl size and shape are depending on the size and shape of the nuclei that is implanted into the body (Victor 2005). Pearls that are larger, rounder, smoother and brighter are those that have a high value, while color is a subjective indicator depending on the species of pearl oyster (Taylor & Starck 2008). Pearl grading systems vary depending on the company that classifies them (Taylor & Starck 2008). The assessment of pearl quality is grouped alphabetically in four categories, namely AAA, AA, A, and B. Those categories can be used to assess the quality of mabe pearls (Ruiz-Rubio et al 2006; Kishore et al 2015). The thickness and quality of mabe pearls are influenced by several factors including the culture duration (Ruiz-Rubio et al 2006; Kishore et al 2015; Gordon et al 2018). Short cultured duration can affect pearl quality because the thickness of the nacre is thin and easy to crack while delaying the harvest of the pearl can be caused excessive deposition of nacre so that the mabe pearl will lose the shape (Haws et al 2006).

Palabusa waters located in the Buton Strait is one the best location of mabe pearls cultivation in Southeast Sulawesi, Indonesia. Pearl cultivation has been done by the local community for many years in this area with short harvesting period of 3-4 months after implantation. The aim of this study was to determine the mabe pearl thickness, the number and thickness of aragonite layer, and the quality of mabe pearl at different culture duration.

Material and Method

Rearing and implantation procedures. This study was conducted from July 2017 to January 2018 in the Palabusa waters located in the Buton Strait, Southeast Sulawesi. The seed of *P. penguin* was collected from the wild and cultured for three months at the same raft culture of 1 m depth. A total of 120 shells with dorso-ventral height of 95.70±9.42 mm, 115.45±10.05 mm width, 31.92±3.15 mm thickness, and 105.71±17.21 g weight were implanted with one hemispherical plastic nuclei with 12 mm diameter and 10 mm height in the left valve using a glue without anaeshetized (Gordon et al 2018), by one professional technician to get relatively the same results (Ky et al 2013; Ky & Le Moullac 2017; Ky et al 2018; Blay et al 2018). To minimize the stress of the oyster during implantation, the implantation procedure was carried out very quickly in less than two minutes (Kishore et al 2015). After implantation, the oysters were hunging vertically 20 cm under the water surface, with 10 individuals per rope at a distance between individual of about 10 cm, and the distance between two ropes of about 50 cm. The mabe pearls were harvested at 90 (A), 120 (B), 150 (C) and 180 days (D), by removing them from the raft culture. The valve of the shells was open and the adductor was cut using a knife to separate the two shell valves. The mabe pearl formed in the shell was released using an electrical drill following the methods of Ruiz-Rubio et al (2006); Kishore et al (2015); Gordon et al (2018). The thickness of the mabe from the top and base of nacre was measured and the mabe pearl quality was classified following the modified classification from Ruiz-Rubio et al (2006) and Kishore et al (2015) (Table 1).

Layer thickness. The thickness of pearl layers were measured by millimeter screws (0.01 mm) on the top and base of layers (Ky et al 2013; Kishore et al 2015). The thickness of the pearl layer in each treatment was analyzed statistically by One Way ANOVA.

Aragonite thickness. The numbers of layers and the thickness of the aragonite were observed using the scanning electron microscopy (SEM) method (Lumenta et al 2017, 2019). Observations were made in the Physics laboratory of the Faculty of Mathematics and Natural Sciences, Halu Oleo University. Pearl samples observed were randomly selected according and the observed layer was the layer formed on the outside which is assumed to be the layer formed at the end of the pearl layer.

Pearl quality. Assessment of pearl quality was based on several criteria including thickness, surface, lustre, shape and colors. Determination of quality grade pearl in this study refers to the *Tahitian/South Sea Pearl System,* namely AAA, AA, A, B, and NC, (Table 1) (modified from Ruiz-Rubio et al 2006; Kishore et al 2015).

Table 1

Criteria for evaluating pearl quality

Grade	*Characteristics*	*Category*
AAA	Very high luster, quite symmetrical, smooth surface and even.	The highest quality
AA	High luster, smooth but uneven surface, no surface defects, symmetry.	High quality
A	Good luster, uneven surface slightly deformed, less symmetry.	Medium quality
B	The sparkle is quite a bit blurry, the surface is uneven, not symmetry.	Sufficiently
N/C	Bad luster, thin, speckled, not symmetrical defects.	Not commercial

Statistical analysis. Significant difference of the pearl layer and aragonit thickness was analyzed statistically by One Way ANOVA. Least Square Difference test (LDS test) was then used to test the differences between each treatment.

Results

Pearl layer thickness. The measurements of the average pearls thickness in the top and base sections are shown in Figure 1. The thickest top layer was obtained in treatment D while the thinnest was obtained in treatment A (0.264±0.013 mm, 0.156±0.011 mm respectively). The thickest base layer was obtained in treatment D (0.543±0.011 mm) while the thinnest base layer was obtained in treatment A (0.199±0.015 mm).

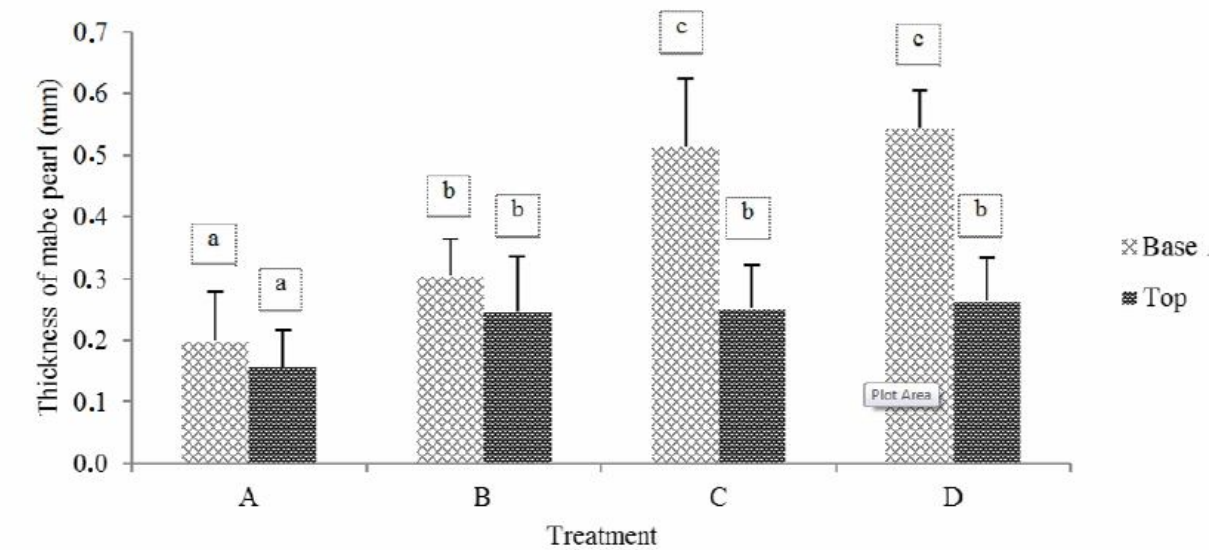

Figure 1. The average of pearl thickness (mm).

The results of the statistical analysis showed that there was a significant difference in the thickness layer of the pearl in the top and base sections in each treatment ($p < 0.05$). Based on the Least Square Difference test (LSD test), the thickness of the top layer in treatment A was significantly different from treatment B, C and D, but there was no significant difference in the thichkness top layer between treatments B, C and D. The base layer thickness in treatment A was significantly different from treatment B, C and D.

Treatment B was significantly different from treatment C and D but treatments C and D were not significantly different.

Aragonite layer. Scanning electron microscopy (SEM) images of the pearl layer during the study period showed variations in thickness of the aragonite layer on the top and base ranging from 0.21 to 0.46 µm and 0.29 to 0.37 µm, respectively (Figure 2).

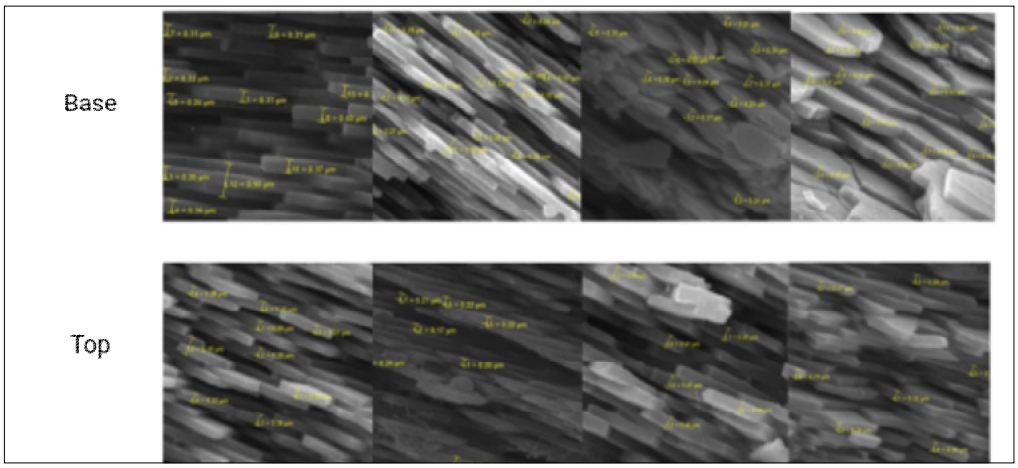

Figure 2. SEM images of aragonite layer thickness (µm).

The number of aragonite layers on pearl formed varied between treatments. The number of top and base aragonite layers in treatment A were 515 and 628 layers, respectively. The top and base layers of treatment B were 811 and 960 layers. Treatment C has 830 layers at the top and 1619 layers at the base, whereas in treatment D, there were 870 top layers and 1730 base layers (Figure 3).

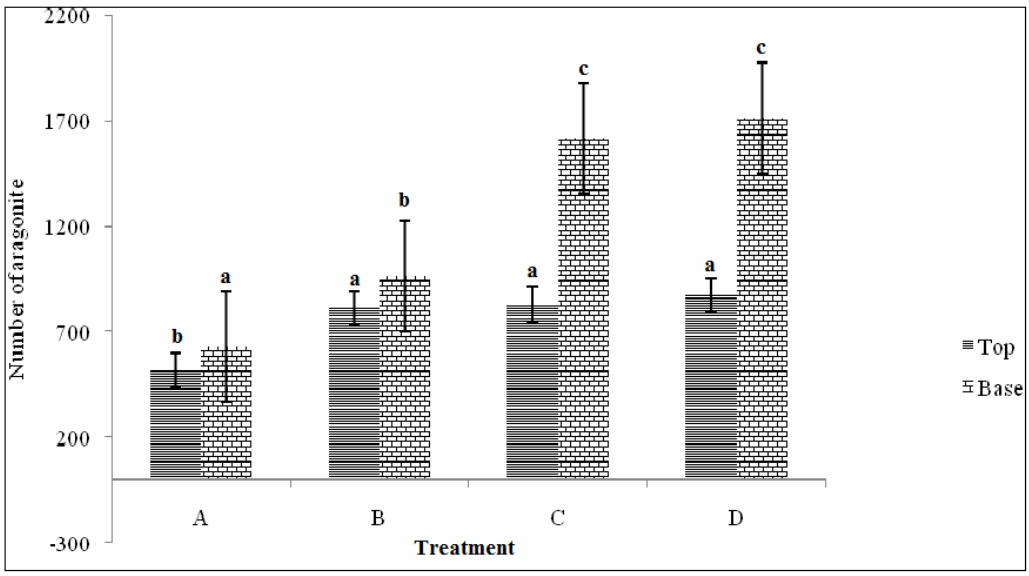

Figure 3. The number of aragonite layers in each treatment.

Pearl quality. The percentage of pearl quality obtained during the study period is presented in Table 2. The highest quality of pearls (AAA grade) was obtained in treatment D (28.6%), followed by treatment C (13.0%) and treatment B (4.5%) whereas treatment A has none AAA grade pearl (0%). The highest grade AA pearls were obtained in treatment D (28.6%), followed by treatment B (22.7%), treatments C (17.4%) and treatment A (11.2%). The grade A pearl was highest in treatment B (50%), followed by treatment C (26.1%), treatment D (19.0%) and A (14.8%). The highest grade B and NC pearls were obtained in treatment A (both at 37%).

Table 2

Pearl quality percentage (%) at different culture period

Grade	*Treatment*			
	A	*B*	*C*	*D*
AAA	0.0	4.5	13.0	28.6
AA	11.2	22.7	17.4	28.6
A	14.8	50.0	26.1	19.0
B	37.0	4.6	26.1	19.0
NC	37.0	18.2	17.4	4.8
Total (%)	100	100	100	100

Information: AAA (highest quality); AA (good quality); A (medium quality); B sufficient); NC (bad/non-commercial).

Discussion. The thickness layer and the quality of the pearl oyster are affected by the culture duration. In this study we found that the longer the culture period after implantation, the higher the thickness of both top and base layers of the pearls and also the quality of pearls. Increasing the thickness of the pearl layer is closely related to the rate of deposition of the nacre in the shell. During the growth process, the mantle organ will continuously secrete calcium carbonate ($CaCO_3$) in the form of aragonite crystals in the nacre layer.

This study showed that the thickness of the pearl layers affects the quality of the pearls in which the thicker the layers the higher the quality of the pearls. This is presumably because the luster of pearls is increasing due to the growing number of aragonite layers. In contrast, the decrease in the pearl quality is generally due to the lack of thickness of the pearl layer, not due to changes in shape caused by excessive nacre deposition. The research done by Ruiz-Rubio et al (2006) and Kishore et al (2015) found that the pearls began to show an increase in layer thickness 5-6 months after implantation.

The quality of mabe pearls is determined by the thickness of the nacre layers that surrounds the nucleus where the thickness of the layer is affected by the increasing number of aragonite layers. The number of aragonite layers can vary between individual pearl, however the desired aragonite layer is a thinner layer with a large numbers of layers. According to Muhammad et al (2017) the thickness of the pearl layer affects the quality of the pearl, while Kripa et al (2007) states the thickness can affect the luster of the pearl. Alagarswami (1987, 1991), states that a thinner layer of aragonite, especially at the top, can add to the luster of pearls, while Taylor & Starck (2008) state that an inner pearl aragonite layer affects pearl resistance. Furthermore, Muhammad et al (2017) states that aragonite pieces are the causative agent for the mixing of colorful pearls.

Mabe pearls that are marketed both locally and out of the area are generally obtained from pearl oyster that are kept for 4 months after implantation with a thickness of 0.25-0.35 mm. The thickness of the pearl still looks transparent which allows the craftsmen to add various colors inside the pearl cubes before further processing. The colors used generally vary, namely red, pink, blue, cream and white which are adjusted to consumer demand and trend. High thickness causes the pearl layer to become non-transparent so that the craftsmen have difficulty in providing additional color to process pearls. According to Gordon et al (2019), the minimum thickness of mabe pearls suitable for mabe pearl industry was 0.25 mm.

Based on the experience of the pearl farmers at the study site, pearl harvesting was generally carried out at 4 months after implantation resulted in 20-30% of AAA grade of mabe pearls while the remaining 40-50% were unprocessed or had no economic value due to insufficient thickness, uneven coating, imperfect surface and the presence of defects on the surface of the pearl as well as an asymmetrical shape (Laode Mahmud, 2018 personal communication). From this study, we found that harvesting at 6 months after implantation resulted in 28.6% of AAA grade pearls, 28.6% of AA grade pearls, 19% of grade A pearls and 19% grade B pearls. Hence, it is more economically valuable

to harvest at 6 months after implantation as almost all pearls produced have economic values.

Conclusions. Culture duration affects aragonite layer thickness, number of aragonite layers and the pearl quality of winged pearl oyster (*Pteria penguin*). Treatment D (6 months culture duration) has the thickest aragonite layers (top and base layers) whereas treatment A (3 months culture duration) has the thinnes aragonite layers. The highest number of aragonite layers obtained in the treatment D and the lowest number obtained in treatment A. The highest quality of pearls (AAA grade) obtained in treatment D followed by treatment C, treatment B and treatment A.

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