STUDY ON SHELL MORPHOLOGY AND INTERNAL SOFT TISSUE ANATOMY OF GOLD-LIPPED PEARL OYSTER *PINCTADA MAXIMA* (JAMESON, 1901) IN MYEIK ARCHIPELAGIC WATERS

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Abstract

Pearl oyster of *Pinctada maxima* is the largest species in genus *Pinctada* and commonly found in Myeik Archipelagic waters. Morphology of *Pinctada maxima* mainly based on shell features such as external scale sculpture, internal features of shell. Its internal organs, especially anal papilla had different shape from other *Pinctada* taxa. Mantle was enclosing all soft tissues and organs. The adductor muscle was strongest and controls the closing of valves.The fundamental function of mantle is to secrete shell valves and ensure their growth. Gills performed the sieving and sorting of food particles. Labial palps accepted the food particles that filtered from gills or released the unnecessary particles as pseudofeces. The adductor muscle is strongest and controls the closing of valves.

Keywords: Nacre boundary, morphology, anal papilla, labial palps, anatomical, pseudofeces.

Introduction

Pearl oysters are members of the Phylum Mollusca and belong to the class Bivalvia. Having two shells (two valves), a soft body with a small foot, a byssal gland and paired gills are distinguished of bivalve mollusks. There are many different species of mollusk that can produce pearls but the pearls which are not made of true pearly nacre have small value except as curiosities (Mar Lar Myo Sein, 1982). Pearl producing molluscs can be divided in two group; those producing nacreous and non-nacreous pearls (as cited in Mamangkey, 2009). Pearl oysters are not closely related to true oysters or edible oysters (Family Oysteridae), being members of a distinct family, the feathered oysters (Family Pteriidae) in the order Pterioida. In family Pteriidae; two genera are cultivated for pearl production, they are *Pinctada* and *Pteria*. In these two genera, the most important is the genus *Pinctada* (Marques & Barbier, 2015). Of the taxa within the genus *Pinctada*, *Pinctada maxima* is the most important in terms of commercial cultured pearl production. The species of *Pinctada maxima* was recorded for the first time in northern Australian waters (as cited in Southgate & Lucas, 2008). It is the largest species in the genus *Pinctada* and it produces the largest and finest pearls. *Pinctada maxima* is differentiated from other species of *Pinctada* by the absence of denticles on the hinge (Humphrey and Norton, 2005).

Pearl oysters are filter feeders; feed on small algae found in the water column. The gills in bivalves are large, and tiny hair-like cilia on the gills are used to trap plankton and particles in the mucus of gill, and from there are transported to the mouth, where they are eaten, digested, and expelled as feces or pseudofeces. Both adults and larvae feed on algae and other small organisms. Clear tropical waters contain limited amounts of algae. Therefore, a large amount of water must be filtered daily in order for the pearl oyster to obtain sufficient food (Maria Haws, 2002). An oyster can filter up to 5 L (1.3 US gal) of water per hour. The commercial harvesting and farming of pearl oysters of the genus *Pinctada* comprises an expanding industry in tropical marine environments worldwide.

Earlier methods of harvesting oysters for their shell and for natural pearls have generally been superseded by farming of world harvested oysters, by the hatchery production of seed stocks and by the cultivation of pearls (Humphrey, Norton, *etal*. 1998). In Myanmar, the natural pearls

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had been produced since the later part of the 19thcentury and the mother of pearls was gathered from natural stock. In 1957, the first cultured pearl were successfully produced from the pioneer pearl culture farm; Myanmar Pearl Enterprise (MPE) at Pale Kyun, Myeik Archipelago, Taninthayi Region. (Nyo Nyo Tun and Htay Aung, 2009). In Myanmar, pearl culture initiated as Japanese-Myanmar joint-venture farm in 1954. The pearl oysters were collected from wild and seeding was implemented by Japanese technicians. In 1987, hatchery trials on *Pinctada maxima* have been lead (Tint Tun, 1998). Today, there are 12 pearl oyster farms extended in Myeik Archipelago. The selective farm for the present study is the Myanmar Oriental Pearl Farm, located on Narr Kho Island in the southern part of Myeik Archipelago. As an introduction to this study, the taxonomy of *Pinctada maxima* (Jameson), the external and internal structures of shell and the anatomy of internal tissues and organs, those are the objectives of this study.

Materials and Methods

Study area

This study was carried out in the pearl farm of Myanmar Oriental Pearl Company located in Narr Kho Island (Lat 10° 42' N and Long 97° 95' E). Narr Kho Island is situated about 80km far from Kawthaung and is located in the southernmost part of Myanmar (Figure 1).

Figure 1 Map showing the study pearl farm area.

Methods for shell morphology and anatomical study

The specimens of different ages and sizes were collected from the long lines. And the external shell morphology of juvenile and adult oysters was studied. To study the internal shell feature, valves of oysters was opened by using openers and the adductor muscle that attached to valves was detached. After removing the adductor muscle, the internal features of shell such as prismatic layer, nacre boundary, nacre layer, adductor muscle scar, pallial muscle scar, retractor muscle scar and ligament were clearly seen. For anatomical study of internal organs, only the left valve of oyster shell was carefully removed not to damage internal organs by detaching the attachment of adductor muscle in the left valve. And then the left lobe of mantle was carefully removed by using dissecting set. Then the internal organs of pearl oyster were dissected and photographed. The filaments of gills and anal papilla were examined under compound microscope.

Results

Classification of *Pinctada maxima*

Phylum – Mollusca

Class – Bivalvia

Order – Pterioida

Family – Pteriidae

Genus – *Pinctada*

Species – *Pinctada maxima* (Jameson 1901)

Description

Pinctada maxima has the large shell up to 30 cm in dorsal ventral measurement, subcircular in outline and typically has pale fawn in colour. The juveniles display the following colours: green, purple-black, yellow, cream, grey, brown and zigzag patterns of purple maroon. Hinge teeth were absent but anodyne (a series of transverse ligament grooves) were strong. Hinge line is long, straight and continues with subtriangular shaped anterior auricle but sometime nearly obsolete in large individuals. The posterior auricle is short, broadly rounded sinuated and it is well developed in young individuals. The internal nacreous layers were highly lustrous, silvery with a variably extended golden border. The anal funnel is flat, relatively long and paddle-shaped: a few narrow at the distal part and end with a blunt tip.

Shell morphology of *Pinctada maxima*

Pinctada maxima possess a pair of valves. Both valves are attached with a ligament in the dorsal hinge region. Each shell valve is composed of three layers: (1) the outer layer is the periostracum or conchiolin layer; (2) the middle layer is the ostracum or prismatic layer; and (3) the inner layer is the hypostracum or nacre (mother of pearl) layer. These structures are embedded within an organic matrix framework composed mainly of protein.

External features of shell

The shell is proscline, typically rounded in outline, with length typically equaling height (Figure 2). The right valve is usually flat to weakly convex and the left valve is more convex. The convexity decreases with age. The exterior colour of the shell is usually pale fawn, sometimes with valves partially green or ochre distally broadening radial rays. However, the juveniles display the following colours: green, purple-black, yellow, cream, grey, brown and zigzag patterns of purple maroon. By the time the oysters are about 120 mm dorsoventral measurement (DVM), the majority of them have brown coloured shells. The exterior shell margins and scales have commarginal (transverse for scales) alternating dark and light bands. The anterior auricles are very small. The byssal notch is small, narrow, slit-like, vertically elongated, and nearly obsolete in the large individuals. The posterior auricles are indistinct from the valves; they are rarely very inconspicuously and broadly sinuated. The sculpture of the exterior surface of the shell consists of commarginal prismatic scales which are most prominent in young individuals and typically abraded in large individuals

4.1.5.3. Internal features of shell

The internal surface of the shell is in direct contact with the external surface of the mantle and is generally covered by nacre. The interior nacreous surface is thick, lustrous, iridescent, silvery, rarely showing any dark tints with a rich golden rim that develops on the periphery of the

nacreous layer (Figure 2). At the margins of the shell, the nacreous layer merges sharply with a zone of dark, semi-soft proteinaceous conchiolin forming the periostracum. Finger-like projections of this material extend beyond the general periphery of the shell. The inner prismatic margin is of the same ground colour as the exterior. The adductor muscle scar is seen as a comma-shaped depression in the center of the shell. The posterior adductor muscle scar is large, wide, very slightly curving, with the dorsal edge narrower than the ventral edge, adjacent to the smaller, narrow, oval shaped, and anteriorly positioned posterior pedo-byssal retractor muscles scar. The pallial muscles are also seen as a discontinuous curve in the anterior side, stretching from the anteroventral extremity of the adductor muscles scar to the dorsoanterior region.

General structure of internal tissues and organs

After removal of one valve, the mantle lobes are covered and enclosed all other tissues and organs. The mouth, foot and byssus are situated in the anterior region. Mouth is surrounded by the labial palps. A visceral mass is located in the very dorsal region of internal tissue. A large part of digestive organs and gonad are enclosed within visceral mass. The large adductor muscle is obvious in a posterior-ventral position. It is separate from the visceral mass, except dorsal part where a portion of free rectum binds it to the digestive mass.The heart, the byssal/pedal retractor muscle and the brown excretory system (running along the anterior part of the branchial axis) are positioned in the central part between the adductor muscle and the foot. The “pearl sac” or nucleus insertion place is located in the posterior end of the foot. The large pigmented gills are connected anteirorly between the labial palps and they run ventrally and end posterior-ventrally at the pallial fold, in front of the anal papilla. Anal papilla is situated adjacent to the posterior-ventrally part of adductor muscle. The diagrammatic and photo views of internal tissues and organs structure of *Pinctada maxima* were shown in figure 3 and figure 4.

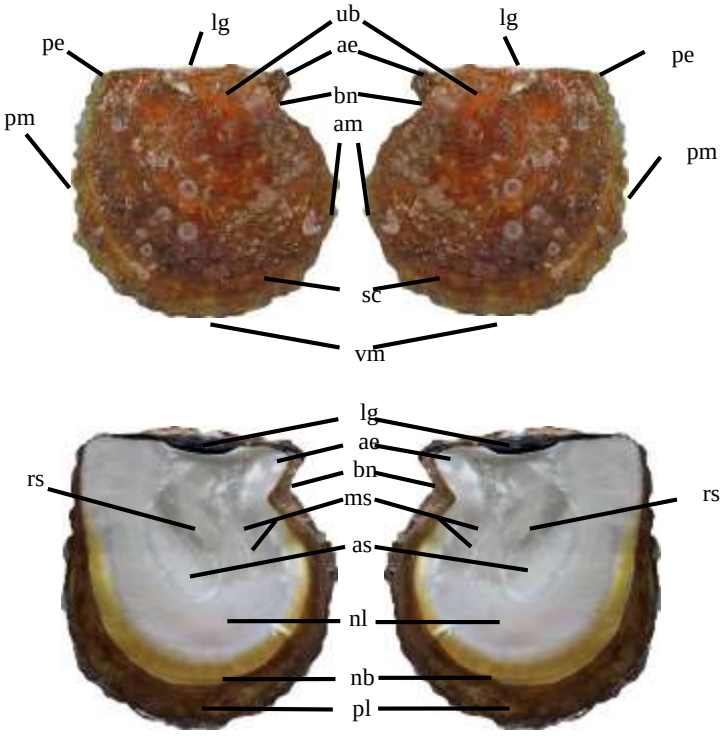

Figure 2 Shell morphology of the exterior and interior surfaces of *Pinctada maxima*. Abbreviations: ae: anterior ear (auricle); am: anterior margin; as: adductor muscle scar; bn: byssal notch; lg: ligament; ms: pallial muscle scar; nb: nacre boundary; nl: nacre layer; pe; posterior ear (auricle); pl: prismatic layer; pm: posterior margin; rs: retractor muscle scar; sc: scales; ub: umbo; vm; ventral margin.

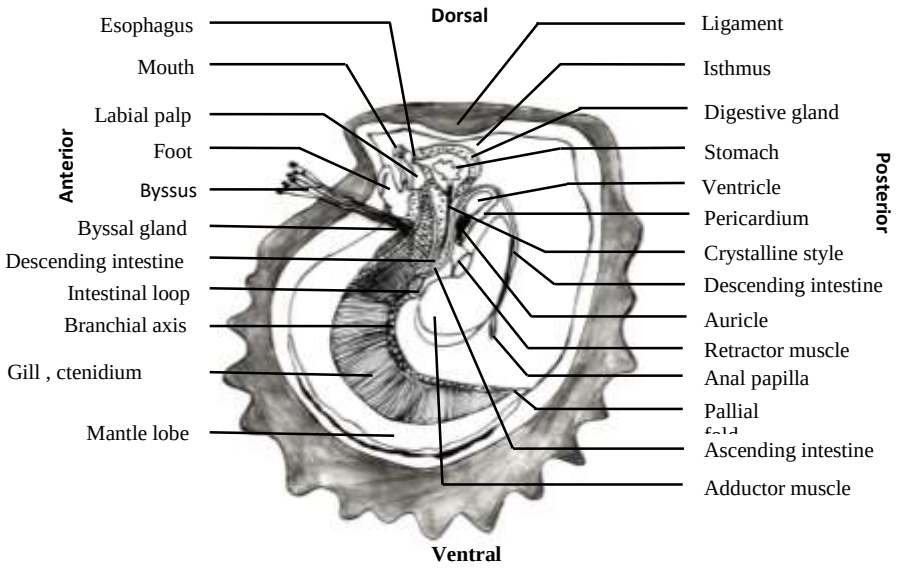

Figure 3 Diagram of internal anatomy of *Pinctada maxima*.

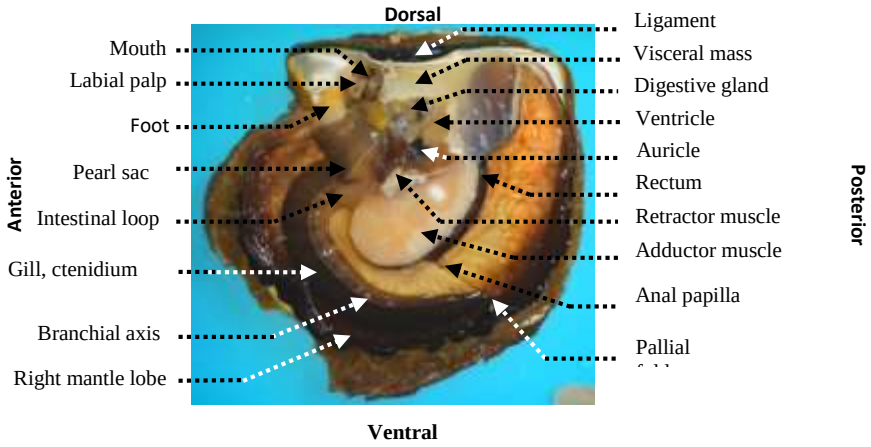

Figure 4 Lateral view of *Pinctada maxima* after removal of left valve, left mantle lobe, left gill.

The mantle

The mantle is a thin, most external and flattened expansive organ, lining the internal surfaces of the shell valves and enclosing all other soft tissues and organs. The fundamental function of the mantel layer is to secrete the shell valves and ensure their growth. The mantle consists of two lobes, with each lining on the inner surface of the shell valve. These two lobes are separated anteriorly, ventrally and posteriorly. But they are fused to the visceral mass and the adductor muscle; they join together at the dorsal portion along the hinge line, forming the isthmus. Each mantle lobe can be divided into four zones: (1) the marginal zone that is thick and pigmented and fringed with branched tentacles, (2) the distal or pallial zones that is attached to the shell, composed primarily of muscular threads used in mantle retraction that are visible to the naked eye (3) the central zone, adhering and covers the soft tissue of visceral mass and adductor muscle, and (4) the isthmus zone of fusing lobes along the hinge dorsally (Figure 5). Along the extremities of the outer margin of the mantle, a distinct pigmented thickening is visible. The outer marginal zone splits into three folds: the outer, middle and inner folds, each with specific roles. The mantle cavity is closed by each side of inner folds, when oyster opens its valves, forms two apertures of inhalant and exhalant to the pallial zone. Tissue from the pallial zone is used as graft tissue in the operation for seeding.

The gills

There are two symmetrical gills that lie between the mantle lobes and enclosed within the mantle cavity, just above the adductor muscle. They are flat, crescent-shaped and filamentous gills, characteristic of bivalves with lamellibranches gills. Each gill is composed of four elongate lamellae and is W-shaped in transverse section. Each gill is fused anteriorly with the visceral mass by its brachial axis and then it is fused with the ventral zone of adductor muscle. The fused gills end at the pallial fold facing the anal papilla, in a posterior-ventral position. The elongate W-shaped lamellae are comprised of ordinary filaments and principle filaments (Figure 6). The ciliated discs connect the ordinary filaments and also join to the principle filaments by long thick and inter-locking cilia. The functions of the gills are respiration and filter feeding as sieving and sorting of food particles. The gills are normally suspended in the water within the shell cavity but collapse on opening of the shell.

The adductor muscle

The adductor muscle is the largest and most important muscle in the body of oyster. It is a conspicuous crescent-shaped mass lies generally central to the shell valve; apposed the gills and the mantle (Figure 3 and 4). The adductor muscle is firmly attached to the inner surface of each shell valve. It is a massive wedge-shaped bundle of muscle fibres and control the closing of the shell valve.

The byssal/pedal retractor muscle

A pair of pedo-byssal rectractor muscle is projecting laterally from the visceral mass. This muscle is V-shaped and originates from the byssal gland and running to posterior and form two large white oval masses adhering to the adductor muscle (Figure 7).

The foot

The foot arises as a protrusive, tongue-like organ that is oval in transverse section. It is located in the anterior part of the body between the mouth and the byssus, extending towards the byssal notch (Figure 3). A pedalgroove runs along the ventral portion of the foot and it extends into the byssal organ. The foot is turgescent muscle and its function as locomotion performs during the early stages before settles as spat.

The byssal gland

The byssal gland is located in the central part of the retractor muscles. Byssal threads are secreted by the byssal gland and pass down the pedal groove which is formed into a tube. Byssal threads are extending toward and through the byssal notch in the shell. The distal end of byssal threads flatten as a disc, which help to anchor with the substrate (Figure 8).

The nucleus insertion place (pearl sac)

The “pearl sac” is located in the posterior end of foot and under the retractor muscle. It is a transparent turgescent organ where the nucleus and mantle tissue are inserted for pearl formation (Figure 8). The intestinal loop comprises in the posterior region of the “pearl sac”. The “pearl sac” is usually recognized as gonad because the gonad invades into it when the animal is sexually matures.

The gonad

The sexes of pearl oysters are indistinguishable from the external appearance of the gonad. The gonads are paired but asymmetrical. They generally occupy as a layer between the connecting tissue and digestive gland and also protruding to antero-ventral part of visceral mass and intestinal loop.

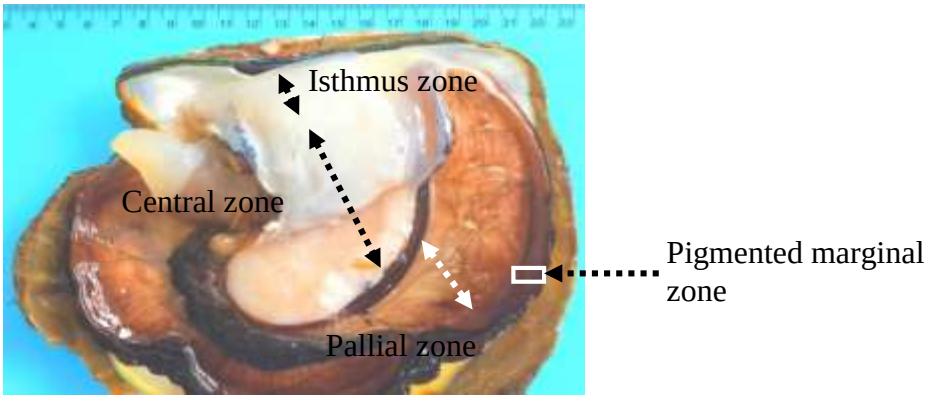

Figure 5 The four zones of mantle lobe of *Pinctada maxima*: (1) marginal zone; (2) pallial zone; (3) central zone; (4) isthmus zone.

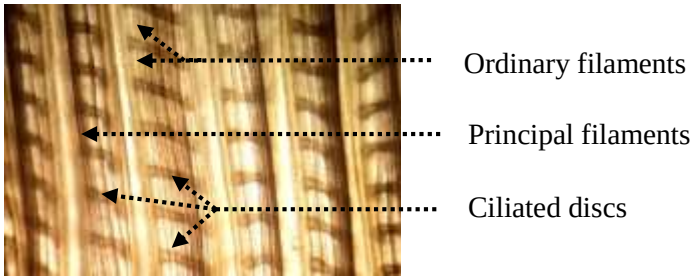

Figure 6 Microscopic view of branchial lamellae of *Pinctada maxima*.

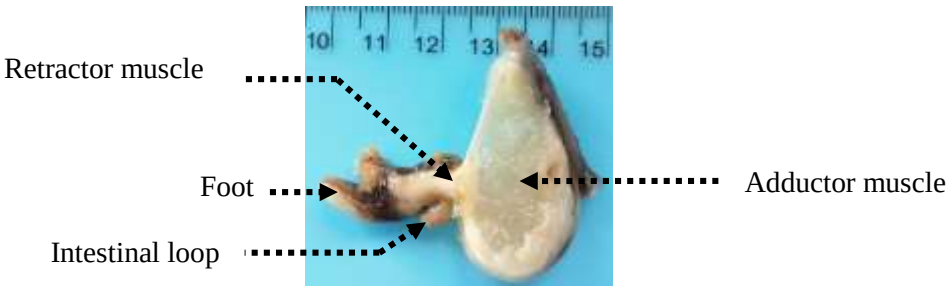

Figure 7 Attachment of retractor muscle, adductor muscle and foot of *Pinctada maxima.*

At the sexual maturity stage, the gonad invades the “pearl sac”. Both sex of the gonad are whitish in the initial stage. In the matured state, gonad of the male is pale creamy in coloured and that of female is yellowish creamy.

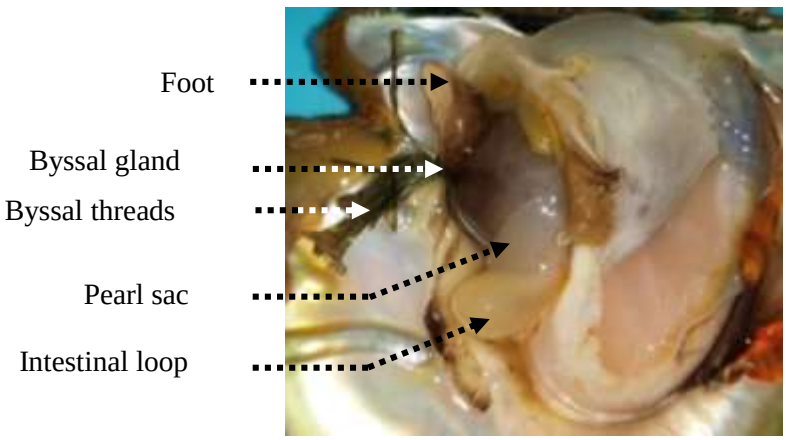

Figure 8 The foot, byssal gland, byssal threads, pearl sac and intestinal loop of *Pinctada maxima*.

The heart

After one of the shells is removed, the heart is seen as a dark mass within a pericardial membrane (Figure 3 and 4). It is located between visceral mass and the adductor muscle. It is limited dorsally by the small portion of free rectum and limited ventrally by the retractor muscles. The heart consists of a single ventricle and a pair of contractile thin walled auricles, one on each side (Figure 9).

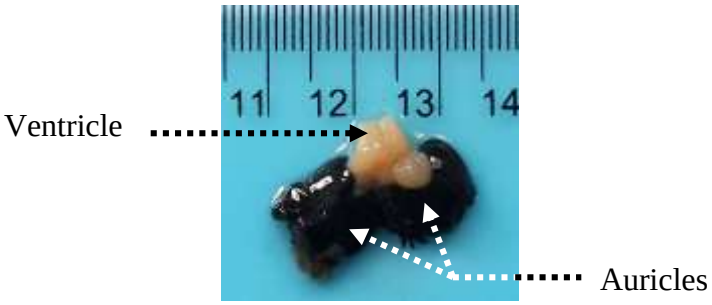

Figure 9 The heart of *Pinctada maxima*: single ventricle and a pair of auricles.

The digestive tract

The visceral mass covers mostly parts of digestive tract of the pearl oyster. The mouth is concealed with two labial palps (Figure 3). These labial palps accept the organic and inorganic particles that are filtered by gills. And also labial palps released unnecessary materials and particles of low nutritional value as pseudofeces. The mouth is connected to short esophagus which opens into the stomach. The stomach has many chambers which are covered by the digestive gland (Figure 10). In the sexually immature stage of pearl oyster, the stomach can be seen through transparent visceral mass tissue. A long semi-transparent crystalline style rod protrudes across the stomach cavity and bears the stomach wall and then into the initial descending intestine (Figure 3 and 10). This crystalline style rod produced gastric fluid.

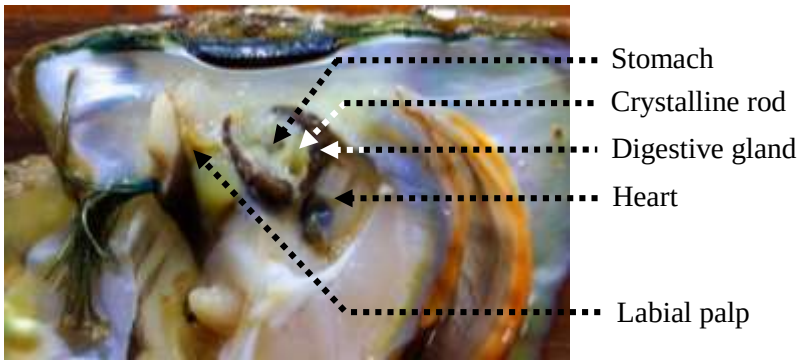

Figure 10 The sagittal section of the digestive gland and stomach in the visceral mass of *Pinctada maxima*.

The intestine of the pearl oyster can be divided into three significant zones: (1) the anterior descending intestine, (2) the ascending intestine and (3) the posterior descending intestine or rectum (Figure 3). The anterior descending intestine emerges from the stomach and runs ventrally between the retractor muscles and extends to the posterior region of the "pearl sac" where the junction between the anterior descending intestine and the ascending intestine is formed as an intestinal loop. The ascending intestine passes around the anterior descending intestine to the left and then suddenly rises and parallel to anterior descending intestine and enter the dorso-posterior part of visceral mass. And then, it emerges from the visceral mass above the heart and curved down

ventrally and then passes around the convex side of the adductor muscle in the median line as the posterior descending intestine or rectum. The rectum terminates as anus and bonds to a paddle-shaped anal papilla which is positioned back to back of the posterior-ventral region of the adductor muscle (Figure 11).

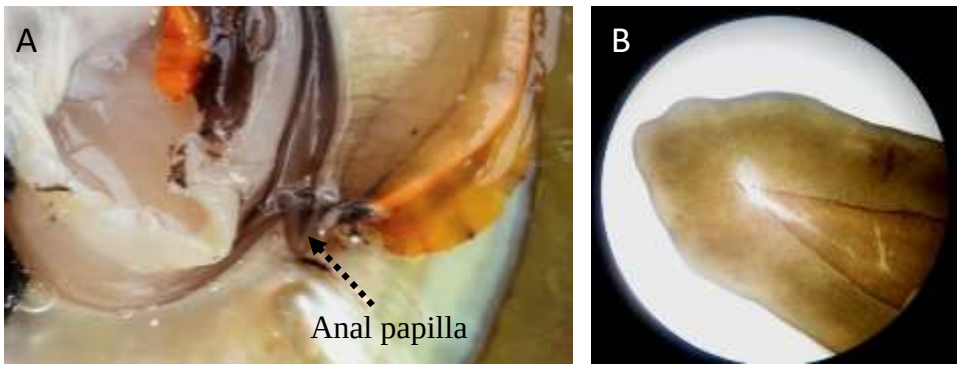

Figure 11 A) The paddle-shaped anal papilla located posterior-ventral part of adductor muscle in *Pinctada maxima*; B) The microscopic view of anal papilla.

Discussion

The pearl oyster *Pinctada maxima* was the largest pearl oyster in the genus *Pinctada*, up to 30 cm in dorsal ventral measurement. According to their nacre boundary, the *Pinctada maxima* can be separated as gold-lipped and silver-lipped. In the present study, oysters with gold-lipped are mostly found and silver-lipped oysters are rarely observed in the pearl farm of Myanmar. They can produce the largest gold and silver south sea pearl. The silver-lipped or gold-lipped pearl oyster, *P. maxima* has no teeth or denticles on the hinge; this character is the difference from other genus. In the study site, another species of genus *Pinctada* such as, *P. margartifera* and *P. fucata* were found. The size of *P. margartifera* reached 20-25 cm in dorsal ventral measurement that is smaller than *P. maxima*. *P. fucata* is the smallest size compared to the previous two because the largest size of *P. fucata* is about 10 cm in dorsal ventral measurement. In general, the colour of external shell of *P. maxima* is pale fawn and has no trace of radial marking. In some cases, the umbonal region is coloured green, dark brown or purple (as cited in Gervis, 1992). The shells of juvenile are green, purple-black, yellow, dark-red, brown and grey colours in the present study. But the external shell colour of *P. margartifera* is dark grayish brown and has lighter striations that are radiating from the umbo. The nacre boundary of *P. margartifera* is black colour and it produced black colour pearl. The external shell colour of *P. fucata* was varying from the cream, brown to bronzes and black. And the colour of internal shell nacre boundary is cream to golden with a hard metallic luster.

The shell valves of *P. maxima* are less convex and hinge lines are longer than *P. margartifera* (Gervis, 1992). In the juvenile stages, the growth processes are slightly convoluted and the distal part is wider than proximal. Their ends are blunt not tapered like *Pinctada fucata*. The dorsal lobe of adductor muscle scar of *P. maxima* is significantly larger and broader compared to *Pinctada margaritifera*. The spat and juvenile stages of *Pinctada maxima* can able to detach their byssal threads and can reattached other suitable place. The byssal attachments are retained up to about three years of age and then byssus are lost they retained on the bottom and kept in position by their shell weights. But the *Pinctada margaritifera* usually attache to the substrate by byssus throughout their life and they can secrete a new byssal threads within a week when they are severed (Gervis, 1992). The anal funnel shapes are also unusual according to different species of pearl oysters. In the *Pinctada maxiam*, the anal funnel is flat, little long and paddle-shaped: a few narrow at the distal part and end with a blunt tip. And the shape of anal funnel of *Pinctada fucata* is thin

and elongated. The distal part is narrow and wider in the middle and end with the elongated tip. But the anal funnel of *Pinctada margaritifera* is wider than others and look like shaped of banyan leaf (Gervis, 1992).

It is very limited scientific researches of pearl oysters *Pinctada maxima* in Myanmar. The present study was mainly emphasized on morphology and anatomy of pearl oyster *Pinctada maxima*. Further study should be carried out for detail process for their histological of gonad and reproduction for the purpose of hatchery, culture and final intension of pearl production.

Conclusion

Pinctada maxima is largest species of oysters and commonly distributed in Myeik Archipelagic waters. It produces the largest and finest gold or silver colour pearls, is also called south sea pearl. It is economical important species because of high price in global market. Water temperature, salinity and nutrition have directly impact on their growth and survivals of pearl oyster. The present study was not complete to express the biological process of *Pinctada maxima*. But it is believed that this study will contribute to promote the fundamental knowledge of morphology and anatomy of pearl oyster *Pinctada maxima*. This could be partly supported for further study of pearl culture.

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References

Gervis, M. H. and Sims, N. A. (1992). *The Biology and Culture of Pearl Oysters (Bivalvia: Pteriidae)*. International Center for Living Aquatic Resources Management, Manila, Philippines. 49 pp.

Humphrey, J. D., Norton, J. H., Barton, M. A., Connell, M. T., Shelley, C. C. and Creeper, J. H. (1998). Pearl Oyster (*Pinctada maxima*) Aquaculture: Health Survey of Northern Territory, Western Australia and Queensland Pearl Oyster Beds and Farms. *Department of Primary Industry*. 1-35.

Humphrey, J. D. and Norton, J. H. (2005). *An Atlas of Functional Anatomy, Pathology and Histopathology*. Department of Primary Industry, Fisheries and Mines. 111 pp.

Mamangkey, N. G. F. (2009). Improving the quality of pearls from *Pinctada maxima*. Unpublished PhD Dissertation. James Cook University, Austualia.

Maria Haws. (2002). *The Basics of Pearl Farming: A layman's Manual*. Pearl Research and Training Program Pacific Aquaculture and Coastal Resources Center University of Hawaii at Hilo Hilo. USA. 79 pp.

Mar Lar Myo Sein. (1982). Taxonomy and Distribution of Burmese Marine Bivalves. Unpublished MSc Thesis. Department of Zoology, University of Yangon, Yangon, Myanmar.

Marques, R. C. & Barbier, E. (2015). Anatomical Differences among Specimens of *Pinctada imbricate* Roding, 1798 from different South American Localities. *Bol. Inst. Pesca. Sao Paulo.* **41**: 751 – 761.

Nyo-Nyo-Tun, Daw, Htay Aung, U. (2009.)Study on the Biology of the Pearl Oyster at Pearl Farming Station, Pale Kyun Waters. *Myeik University Research Journal*. **1**: 47 – 59.

Southgate, P.C. & Lucas, J.S. (2008). *The Pearl Oyster*. Elsevier, Amsterdam. 574 pp.

Tint Tun. (1998). Myanmar pearling: past, present and future. *SPC Pearl Oyster Information Bullentin,* **12**: 1-5