Gonadal Histological Study of Oyster (*Crassostrea sp*) Collected from the South-East Coast of Bangladesh

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Registration No: 709

Session: 2019-2020

A Thesis Submitted in the Partial Fulfillment of the Requirements for the Degree of Master of Science in Marine Bioresource Science

Department of Marine Bioresource Science

Faculty of Fisheries

Chattogram Veterinary and Animal Sciences University

Khulshi, Chattogram-4225, Bangladesh

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This is to certify that we have examined the above Master's thesis and have found that it is complete and satisfactory in all respects and that all revisions required by the thesis examination committee have been made

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AUTHORIZATION

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Nasrin Nahar

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The Author

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ABSTRACT

The edible oyster (*Crassostrea* sp.) is one of the world's most promising mariculture shellfish species which is bound in the coastal belt of Bangladesh especially along the Cox's Bazar coast. For development of oyster culture technology, understanding the reproductive biology of *Crassostrea* sp is required. The objective of this study is to use histology to properly identify the dynamic characteristics of gonadal development process. The study was conducted during March 2018 to February 2019 and a total of 240 oysters were taken from the Moheskhali Channel, Cox's Bazar. The collected samples were analyzed for total length, total weight, shell length and width, gonad weight and wet tissue weight. Length-Weight relationships showed negative allometric growth (b<3) GSI (gonadosomatic index) and histological analysis were done to determine the reproductive cycle. GSI value was found to be generally higher in female than male. The percentage of the male was 30% where the female was 70%. The study confirmed that the spawning of *Crassostrea* sp. occurred twice during one year study from March 2018 to February 2019. In case of males, two peaks were reported during June and November, whereas, for females, July and December indicated as peak mature stage. The finding of this study would be beneficial for managing naturally occurring oyster stocks and developing future aquaculture aspects in the coastal belt of Bangladesh.

CHAPTER ONE
INTRODUCTION

1.1 Background of the study

One of the world's biggest deltas is in Bangladesh. The country is located between 20°34" to 26°38" North latitude and 88°01" to 92°41"East longitude. Along its southern boundary, Bangladesh possesses abundant marine and coastal resources. Its 480 km of shoreline runs along the northern and north-eastern shores of the Bay of Bengal. In terms of both local and global growth of economy, the Bay of Bengal has grown in significance. The coastal region of the nation is regarded as one of the most fruitful regions in the world because of its geographic location and climatic characteristics. It is rich in terms of biological richness in addition to its extensive water expanses. The country's social and economic growth as well as the ecological balance of the region have both been significantly influenced by the coastal and marine fisheries. Long-standing commercial exploitation of several significant fish species with substantial export values (BFRSS, 1986). As one of the richest ecosystems in the world, the coastal and marine zone of Bangladesh is distinguished by increased production and distinctive mangrove impacts. The coastal and marine waters of the Bay of Bengal include a total of 475 species of fish that are divided into 133 families (Hussain MA, 1994). Bangladesh is rank third in the world for aquaculture production. Bangladesh's aquaculture industry, accounts for around 2.8 percent of GDP (FAO, 2017).

Molluska is the second largest phylum in the animal kingdom. It belongs to Lophotrochozoa, a clade poorly understood in terms of reproduction. This phylum presents a diversity of organisms with extremely varied sexual reproduction, including gonochorism and functional hermaphroditism (simultaneous and sequential), indicating a variety of underlying sex determination systems. Mollusk are important for food production and the economy, thus understanding how to determine their sex is essential to developing effective methods for controlling their sex in aquaculture (Collin, 2013).

The Bay of Bengal and its surrounding regions are home to a wide diversity of mollusk species. Clams, oysters, scallops, snails, slugs, cuttlefish, squids, octopuses, and other creatures are among them (Islam, 2003). The aquaculture has benefited

significantly from the mollusk fishery. Ahmed (1990) reported that 301 species of marine mollusks have been found in Bangladesh. Very little is known about their biology, prevalence, production, and management (Ahmed, 1990). The small-scale fishermen who reside close to the coast gather them for both personal use and business purposes. Theses' shells are used to manufacture lime and for fish and poultry feed. Ghosh (2004) stated that, among the people of Rakhaing community of Cox's Bazar collect them from local market. They are also eaten in Japan, Malaysia, Thailand and China as their food item. According to Breton et al. (2017), Oysters are a member of the phylum of Mollusca, which offers a wealth of information on the development of sex and sex determination. Mollusk gonad development is often described using a categorical (frequency) scale. However, as they may be connected to biochemical or physiological variables or utilized as a co-variable for analysis, continuous (numerical) scales of maturity are more accurate to represent reproductive capacity in mollusks. A number of quantitative scales based on continuous data have been presented, such as a comparison of the gonad coverage area (GCA) in *Crassostrea gigas* to the visceral mass area (Mori, 1979; Enriquez-Díaz, 2004; Fabioux et al., 2005) or *Crassostrea gigas* males (Lannan, 1980), and also in *Crassostrea* viriginica (Hefferman & Walker, 1989; Barber & Blake, 1991).

According to reports, the edible oyster, pearl oyster, and windowpane oyster may all be found in the Bay of Bengal's coastal waters. *Crassostrea* is the most significant species of edible oyster in Bangladesh. As ecosystem engineers, oysters (*Crassostrea sp.)* employ the structures to create, modify, and sustain habitats and ecological processes. Oysters may be employed as ecosystem engineers to develop organic defenses for coastal areas. Oysters have the ability to create noticeable habitats that have an impact on the dynamics of the local sediment transport, deposition, consolidation, and stabilization processes as well as the tidal flow, wave action, and sediment dynamics in the coastal environment. Bivalve reefs contribute to the food security and way of life of coastal residents while also serving as habitat for a variety of fish, crabs, and other invertebrates. Artificial oyster reefs on a "living shoreline" may be self-sustaining elements for ecosystem goods and services and coastal defense (Hossain et al., 2013).

Due to the Bay of Bengal's strong tidal currents and quantity of plankton, which provide a perfect habitat for oyster *Crassostrea sp*, these organisms are most frequently employed for culture and study. In the littoral and intertidal zones, they

grow on stationary components like as gravel, rocks, or tree roots. They are gathered in the dry season from the intertidal regions along the estuaries (Kamal, 2000).

In 2015, 15.26 million tonnes of mollusks were produced worldwide. Currently, Bangladesh makes up a relatively small portion of global output. But this culture is a low-investment endeavor that yields excellent results. Oysters, mussels, and scallop exports from Bangladesh brought in $5.51 million in revenue during the 2017–2018 fiscal year (EPB, 2018).

The Moheskhali waterway in the Cox's Bazar area was chosen for this study in order to examine oyster gonadal development and maturity. The decision to choose this location was made because it has a variety of marine ecosystems, including mangrove swamps, coral reefs, and sandy, muddy, and rocky seabeds, all of which are potentially suitable for the growth of a shellfish fishery. An optimal habitat for the growth of edible oysters is provided by the high tide amplitude, enough tidal current, lack of pollution, and high phytoplankton richness (*Crassostrea sp).*

1.2 Research objective:

- To observe the gonadal development of oyster (*Crassostrea sp)* through histological process.

CHAPTER TWO
REVIEW OF LITERATURE

Mollusk species in the Bay of Bengal can contribute significantly to our economy. Because of the sandy and rocky land, mangroves, and coral reefs, Bangladesh's coastline environment is ideal for mollusc habitats (Shahabuddin et al., 2010). Although Mollusk receive less attention than finfish and shrimp in discussions about raising aquaculture production, mollusks are one of the most significant aquaculture enterprises. With one of the oldest histories of culture and cultivation on all continents with the exception of Antarctica, oysters are the most prolific molluscan species in terms of productivity (FAO, 2018 ; Günther, 1897). Because of its ability to alternate sex or sequential hermaphroditism depending on the environment, the edible oyster (Crassostrea sp) is farmed all over the world and used as a delectable food item (Galtsoff, 1964; Wakamatsu, 1973). Oysters are belonging of the family of Oysteridae.

2.1 Length Weight Relationship:

Several works has been done on length weight relationship of *Crassostrea sp*. At different parts of the world. The length-weight relationships (LWRs) *of Crassostrea gigas*, a mangrove oyster, were determined throughout a seven-month period from February to August 2010 in tidal ponds under continuous and periodic submergence. Under both cultivation conditions, the length-weight correlations (LWRs) were found to be statistically significant (p 0.001) in Indonesia (Adisa-bolanta et al., 2015). Researchers examined the population structure, age, growth, mortality, and harvest intensity of the oyster *Crassostrea oadrasensis* in Bangladesh's Moheskhali Channel between June 2003 and May 2004. The allometric growth trend was negative (b 3). (Nurul Amin et al., 2008)

2.2 Gonad Development:

Mainly gonadal development was determined by estimating GSI value and histological analysis. Several works has been done worldwide to determine the reproductive cycle of oyster (*Crassostrea* sp.) though histology and GSI.

Seasonal variation in gonad development of *Crassostrea gigas* was investigated at Gosung Bay, Korea by calculating GSI value where gametogenesis began in February

with two spawning peaks in June and August (Ngo et al., 2003). This result was found by calculating GSI value and through histiligical analysis.

2.3 Water quality parameters affecting gametogenic cycle

The salinity influence the gametogenesis process has been verified by Muranaka and Lannan (1984) and different salinity can affect the development or reproductive cycle Hopkins (1931).

*Crassostrea gasa*r's reproductive growth is influenced by increasing warmth and reduced salinity. The development of the oysters' gonadal tissue was impacted by the salinity regime which was analysed by setting a laboratory experiment. A higher salinity of 24 resulted in more reproductive development (Gomes et al., 2014). Oyster's gonadal maturation is also affected by trace metals. Mn showed gametogenetic activity and good positive correlation with the organism's (*Crassostrea corteziensis)* maturation (Frıas et al., 1999). In this study we want to find the development of gonad around one year (March 2018 to February 2019) by histological process of this species.

CHAPTER THREE
MATERIALS AND METHODS

3.1 Study area

Sampling was done at Moheshkhali Channel. It is bounded by Chakaria Upazilla on the north, Cox's Bazar Sadar Upazilla and Bay of Bengal on the south, Chakaria and Cox's Bazar Sadar Upazilla on the east, Kutubdia Upazilla and Bay of Bengal on the west.

Figure 3.1: Sampling site

3.2 Sample collection and transportation

Samples were collected once in every month from bottom settled wild *Crassostrea* sp. from March 2018 to February 2019. Total twenty live samples were collected per month, which were immediately stored in the ice box with ratio of 1:2 samples and ice, transported to the oceanography laboratory, CVASU. Sampling was done during the low tide period following every cycle.

3.3 Morphometric data collection:

Sampled twenty mussels were used to determine the individual total dry weight. Oyste were initially cleaned up with tap water to remove dirt, mud, sand and shell fragments which were attached on oyster.

Figure 3.2: Sample washing with tap water

Biometric measurements such as length (maximum length along the anterior-posterior axis), height (maximum length along the dorsal-ventral axis), and width (maximum length through both valves) were measured individually using a Vernier caliper to the Vernier constant of 0.01. The total weight of each individual was recorded after the inter-valval (or mantle) fluid was drained.

They were than dissected and the wet soft tissue weight and the shell weight were recorded with electronic weight meter (Amin et al. 2005).

3.4 Length-weight relationship:

The relationship of shell length (SL) to total weight (TW), meat weight (MW) and shell weight (SW) were calculated according to the allometric equation (Le Cren, 1951):

$$W = a^* L^b$$

Since weight is a power function of length, the logarithm is taken so that the exponential relationship can be expressed by a linear equation. This equation can be expressed in its linearized form:

$$\log W = \log a + b \log L$$

This equation is related to regression equation**:** $Y = a + bX$

Where, W is the shell weight (total weight, total unshelled meat, shell weight L is the shell length whilst 'a' is the intercept (initial growth coefficient) and 'b' is slope (relative growth rate of variables).

3.5 Condition index (CI):

Dry tissue weight was measured after drying the sample tissue at 105˚C for 12 hours in hot air oven and was cooled in desiccator. Condition Index was calculated following Yep et al. (2003).

$$\text{CI (g/cm}^3) = \frac{\textit{total soft tissue dry weight } (g)}{\textit{shell volume } (cm^3)} \times 1000$$

3.6 Gonadosomatic Index (GSI):

After collection of length, weight and width data, mantle of oyster was opened by knife to collect soft tissue and blotted. After removing the unwanted dirt, digestive parts and intestine, the gonad of the oyster was collected carefully by using forceps. GSI is the ratio of samples gonad weight to body weight, which is particularly helpful to identify the spawning seasons. To determine the spawning period, gonadosomatic index (GSI) plays a major role as there is a cyclic change in gonad weight in relation to total body weight. The GSI was calculated by month-wise and sex-wise using the equation by Vladykov, 1956.

$$\text{GSI} = \frac{\text{Weight of gonad}}{\text{Total weight of mussel}} \times 100$$

3.7 Histological process:

A small part of the gonad from the mesosoma and the mantle lobe from each dissected oyster will be sub-sampled by cutting a transverse section midway along the anteroposterior axis. Then the following steps specified in figure 3.3 was used for histological process.

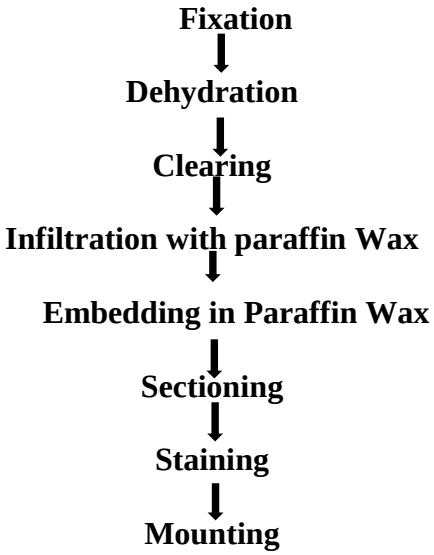

Figure 3.3: Flow chart of steps of histological process

The gonads were fixed in Bouiuns fixative for 24 hours and then post-fixed in 70% alcohol prior to dehydration after washing in 50% alcohol for 2-3 hours. All procedures were followed manually following the procedure described in the table 1.

Table 3.1: Protocol of histological procedure of oyster gonad

STEPS	CHEMICALS	TIME (Hours)
A. Fixation	Bouins Fixative	24
B. Dehydration	I. ethanol 50%	2-3
	II. Ethanol 70%	2-3
	III. Ethanol 80%	2-3
	IV. Ethanol 90%	2-3
	V. Ethanol 95%	2-3
	VI. Ethanol 100%	2-3
	VII. Ethanol 100%	2-3
C. Clearing	1. Alcohol (50%) +Xylene (50%)	overnight
	2 Xylene	2 hours
	3 Xylene	overnight
D. Infiltration	a) Parafin + xylene	2 hours
	b) Paraffin	2 hours
	c) Paraffin	2 hours

E. Embedding

The gonadal tissues were then embedded in paraffin blocks (figure 4a). After embedding the tissues, the paraffin blocks were trimmed to facilitate accurate sectioning.

F. Trimming:

Trimming is a process in which the undesirable wax layers of the embedded blocks are trimmed by knife to obtain suitable blocks. It helps for easy sectioning. After proper embedding, trimming was done to trim undesirable wax layers.

G. Sectioning

The blocks were then sectioned at 5 to 7 µm thickness using microtome (KD-2258) (figure 4b). The sections were then mounted on slides and dried overnight in an incubator at around 40˚ C. Before staining, the sections were dewaxed through immersing them into different bathes of xylene, alcohol, and water.

H. Staining and mounting

Staining was performed using Hematoxylin (Harris Hematoxylin) and Eosin through standard methods (figure 3c) (Bancroft and Stevens, 1996). Upon reaching the desired staining levels, the slides were mounted permanently with D. P. X. mounting media.

.

Figure 3.4: Tissue Embedding, Tissue sectioning, Sectioned tissue in microtome and Staining

The staining schedules are shown in table 2.

Table 3.2: The staining schedule

Sl. No.	Solutions	Time	Process
1	Xylene	10 minutes	Clearing
2	Xylene	10 minutes	
3	Xylene	10 minutes	
4	100% alcohol	5 minutes	Rehydration
5	100% alcohol	5 minutes	
6	90% alcohol	3 minutes	
7	80% alcohol	3 minutes	
8	70% alcohol	3 minutes	
9	50% ethyl alcohol	2 minutes	Staining
10	Distilled water	15 dips	
11	Haematoxylene	3 minutes	
12	Wash in tap water	15 minutes	
13	50% ethyl alcohol	10-15 dips	
14	95% ethyl alcohol	30 seconds	
15	Eosin Y	1 minute	
16	95% ethyl alcohol	2 minutes	Dehydration
17	100% ethyl alcohol	1 minute	
18	100% ethyl alcohol	3 minutes	
19	100% ethyl alcohol	1 minute	
20	Xylene	20 minutes	Clearing
21	Xylene	20 minutes	
22	Drying at room temperature	Overnight	Drying

3.8 Microscopic observation:

The mounted slides were observed under a microscope (OPTIKA, B-192, ITALY), which was connected to computer with Digital camera. By the help of this mechanism several photographs were taken at different magnifications.

3.9 Ranking:

The numerical ranks were valued following Buchanan (2001) and these rankings were used during GI determination. This is done by the stage and different pages of their life style, which is shown in table 3

Table 3.3: Ranking criteria for determining gonad index

Stage	Name	Ranking
1	Resting	1
2-5	Development (A,B,C,D)	2
6	Mature	3
7-9	Spawning (A, B, C)	2
10	Spent	1

3.10 Gonadal index

The maturity stage of each sample was determined by screening the histological slides following Grizel (2003) and the gonad index (GI) was calculated following Buchanan (2001).

$$\mathbf{GI} = \frac{\text{Number in each stage} \times \text{Numerical ranking of that stage}}{\text{Number of animals in the samples}}$$

3.11 Data Analysis

Microsoft excel 2016 was used for data analysis and graphical representation. Calculation and data representation of % gonad development stages, Gonado-somatic index (GSI) and length-weight relationship was calculated by Microsoft excel 2016.

CHAPTER FOUR

RESULT

4.1 Length- weight Relationship

During the study period, 179 samples were taken from the Moheskhali Channel to investigate the association between several shell dimensions (Total length, shell width, and shell height) and weight (Total weight, Soft tissue weight). The morphometric correlations were identified using simple linear regression, as shown in figure 6. The regression analysis' "b" values were compared to 3.0 to see if they were significantly different.

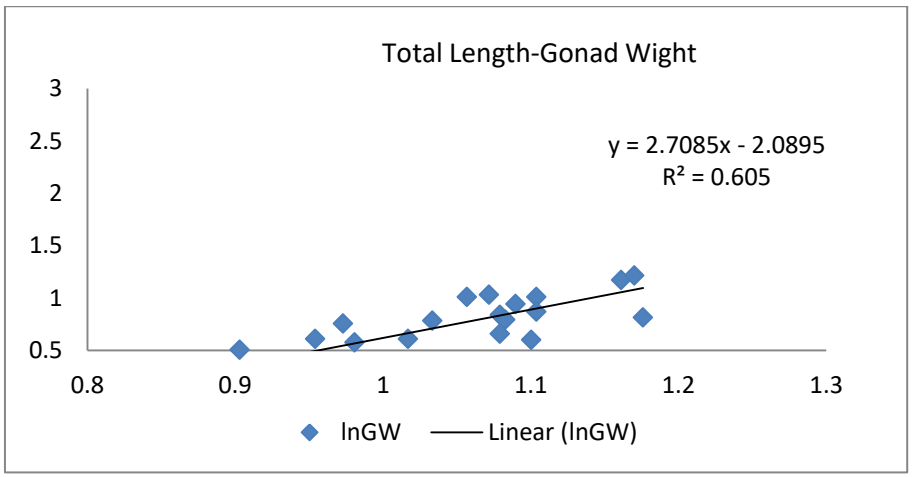

Figure 4.1: Total Length-Gonad Weight

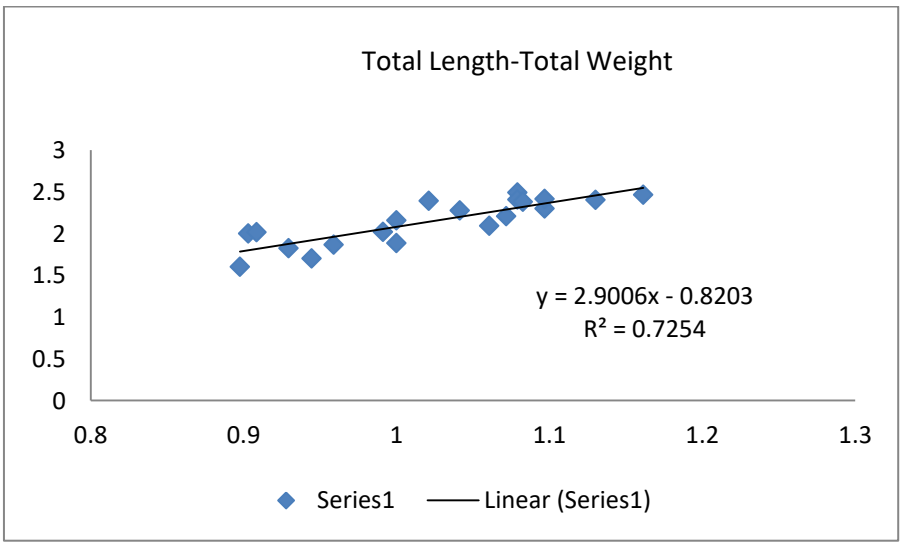

Figure 4.2: Total Length-Total Weight

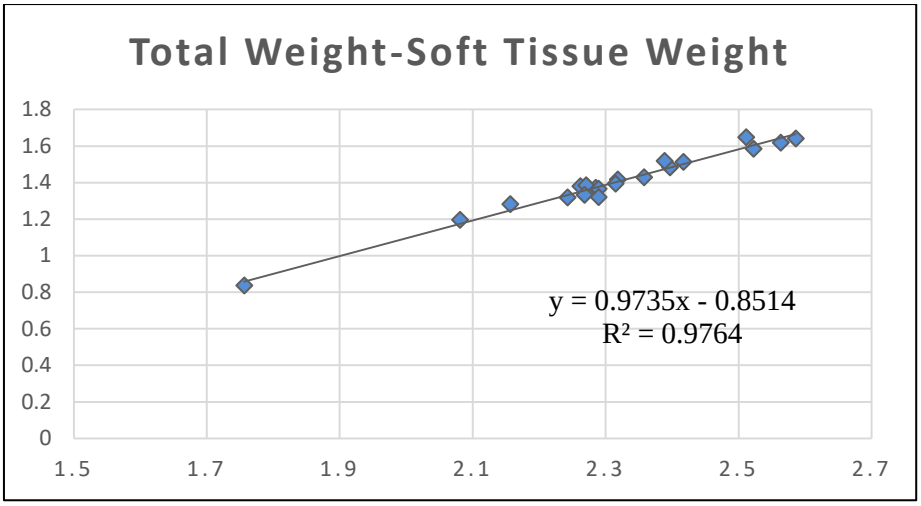

Figure 4.3: Total Weight-Soft Tissue Weight

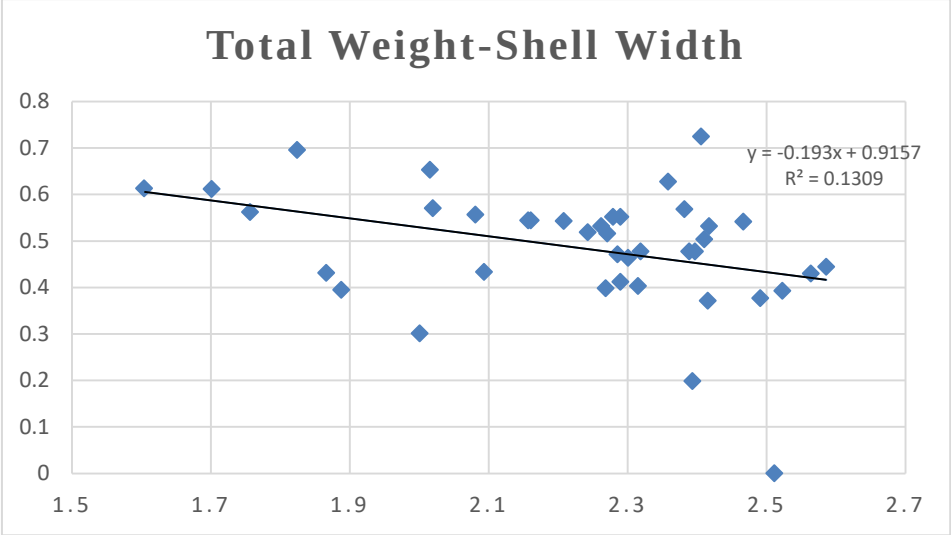

Figure 4.4 : Total Weight-Shell Width

Total length is the indictor of growth of the spices if the value of correction coefficient was close to 1. On the other hand, shell width (SW) and shell height (SH) in contrast of wet tissue weight was far from 1. The statistical analysis showed that, the total weight (TW) and soft tissue weight (STW) increase at a time of the species. But shell length (SL), shell width (SW) and shell height (SH) increases over time.

Table 4.1: Observation of relationship between different lengths dimensions and weight dimension

Variables	R^2	Regression Co-efficient (b)	Growth test	Growth pattern
Total Length-Gonad Wight	0.605	2.7085	b<3	Allometric
Total Length-Total Weight	0.7254	2.9006	b<3	Allometric
Total Weight-Soft Tissue Weight	0.9764	0.9735	b<3	Allometric
Total Weight-Shell Width	0.1309	0.193	b<3	Allometric

4.2 Developmental stages of gonad

1. Resting:

Gonad was largely made up of storage cells without any evidence of sexuality. This stage includes both virgin creatures with a primitive reproductive system and animals that had finished spawning (figure 4.5 A for male and figure 4.6 A for female).

2. Pre-maturation:

Visibility of acini indicated the starting of gametogenesis. Spermatogonia were also observed. Storage connective tissue interspersed between acini. Large quantities of previtellogenesis oogonia and oocytes, as well as a limited number of vitellogenesis oocytes (figure 4.5 B for male and figure 4.6 B for female).

3. In-maturation:

The acini had increased in size containing large number of spermatids and spermatozoa in the lumen. Tails of spermatozoa were directed toward the lumen. There is less interstitial tissue due to follicular expansion and the presence of mature cells in the lumen (figure 4.5 C for male and figure 4.6 C for female).

4. Mature:

With a layer of spermatocytes at the acini's margin and the lumen densely filled with spermatozoa, the gonadal acini attained its maximum size. There was a scarcity of interstitial tissue. The follicles had reached the pinnacle of development and had a large number of mature cells. The oogonia were present in this stage, as they were in all others, although in smaller numbers (figure 4.5 D for male and figure 4.6 D for female).

5. Spawning:

Acini began to be empty. Some redevelopment on the acinus's periphery. Male acini got enlarged, and the boundaries became difficult to detect (figure 4.5 E for male and figure 4.6 E for female).

6. Spent:

With the presence of cells undergoing atresia, the gonadal acini were in regression. Brown cells and haemocyte migration to the tissue reserve were observed. The follicles were smaller and emptier, with some atretic oocytes found (figure 4.5 F for male and figure 4.6 F for female).

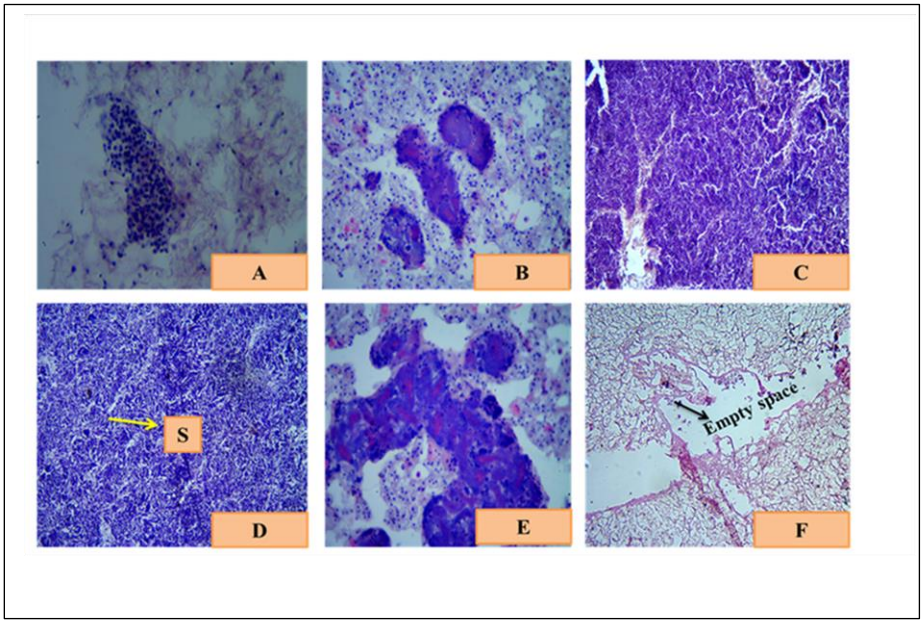

Figure 4.5: Development stages of male gonad
(A=Resting; B= Pre-maturation; C= In-maturation; D=Mature, S=Spermatozoa; E=Spawning; F=Spent)

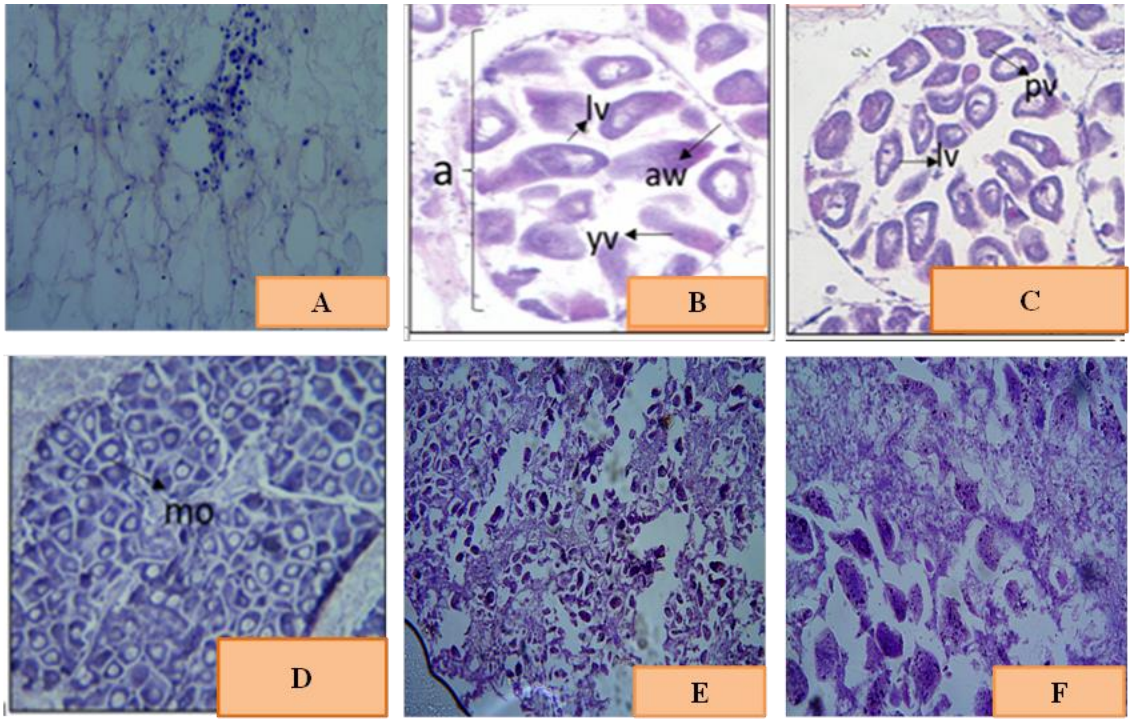

Figure 4.6: Development stages of female (A=Resting; B= Pre maturation; a= acinus, yv= young vitellogenic oocyte, Iv= late vitellogenic oocyte; C= In maturation, pv= Pr-vitellogenic oocyte,D= Mature, mo= mature oocyte,E= Spawning;F= Spent)

4.3 Seasonal variation in gametogenesis

Seasonal variation in gametogenesis of oyster was estimated on the basis of gonadosomatic index (GSI) and histological analysis. The description is given below-

4.4 Monthly gonad development stages

Active gametogenesis of *Crassostrea* sp. in the Moheskhali channel was observed annually from March 2018 to February 2019. During the initial stage of the study, on March'18, resting phase was found for 100% individual. During the month of April'18 small number of male individuals were found at their pre-mature stage which indicates gametogenesis starts at male before female. In May 2018, male were found 40% at their in maturation stage, whereas most females were found 30& at their pre-mature stage and some were at in-maturation stage (30%). In June 2018, mature gonad was spotted first for both male (80%) and female (20%). In July 2018, mature stage were more abundant in female (70%) than male and smaller amount of male (30%) were at their spawning stage. In August 2018 male individuals of spawning

stage were found more than female. In September 2018 spawning females were more abundant than male and some individuals of spent phase were also observed. In October 2018, immature males were observed and spent females were also found. In November 2018, mature males were more abundant than females. In December 2018, mature females were more abundant than males and 20% males were at their spawning stage. In January 2019 spawning females were more abundant than males, whereas spent males were also found at this month. In February 2019, spent stage was observed both for male and female.

Highest percentage of mature males were found at June'18 (80%) and females at July (70%) whereas mature males were also found at Novenber'18 and females at December'18. 100% resting phase was observed at March'18.

4.5 Gonado-somatic index

Calculation indicates gametogenesis occurred twice in a year (March 2018 to February 2019). Two peaks were found both for male and female. Two peaks at June and November for male; July and December for female indicate high GSI value or mature stage of individual. A decreasing trend was found after maturation stage which indicates their spawning stage. In male, decreasing trend was found after June and November which indicates starting of their spawning and in female; it was found after July and December.

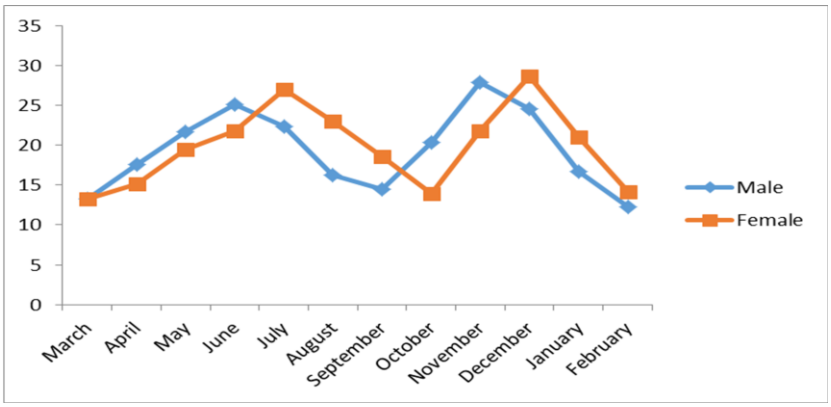

Figure 4.7: Monthly mean gonado-somatic index of male and female

4.6 Egg diameter: Oyster egg were in polygonal shape, not fully round. The egg diameter was estimatedby drawing few lines and then taking average value. Mature egg was considered for calculating egg diameter. The mean egg diameter was 9.46±1.8 µm.

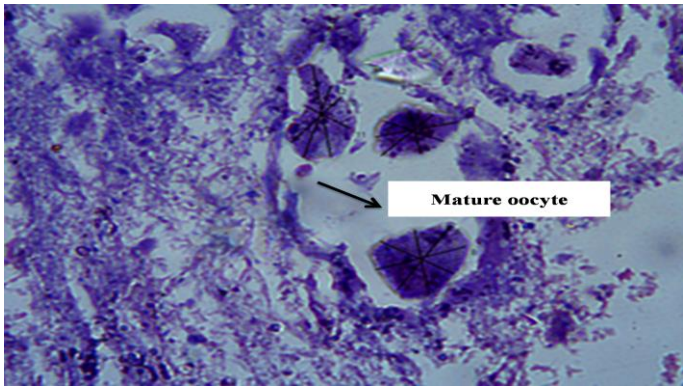

Figure 4.8: Mature oocyte in slide

CHAPTER FIVE
DISCUSSION

This chapter contains the interpretations and descriptions of the research findings. There are additional explanations for the results and the problems that they cause. In this study, the gametogenic cycle is discussed in comparison to earlier research.

5.1 Length - weight relationship

The results showed that the parameter "b" values were significantly different from the isometric value in all circumstances, and that when the value was less than 3 (allometric growth pattern), the weight gain of *Crassosyra* sp. in the Moheskhali Channel was negative. A previous study of Crassostrea madrasensis in Bangladesh's Moheskhali Channel also showed the same growth trend, allometrically negative ($b<3$) (Amin et al., 2008). That means our result matches with the previos study which was conducted at the same place. The results showed that when the shell length, width, and height of the species increased, the total weight and soft tissue weight of oyster samples increased proportionately.

5.2 Gametogenic cycle

Gonadal development of *Crassostrea* sp. was observed through gonado-somatic index (GSI) and histological analysis. Total 240 samples were collected among them 64 were males, 69 were females and 107 were sexually undifferentiated. Maximum number of Males of mature stages was found in June and November. Maximum number of mature females were found at July and December. A previous study conducted at Brazil showed two peaks at June-July and November-December indicating mature stages of Crassostrea brasiliana (Castilho-Westphal et al., 2015). Months of maturation of this study matches with the previous study.

Reproductive cycle and morphology of oyster (Crassostrea sp) which is done by Lamarck (1819), Parana, Brazil, by collecting only 10 adult species at a time in every month. By this they can observe the maturation stage of gonad in which stage female oyster ovarian follicles surrounded by follicular cells and containing germ cells at different stages of development. In this study, we collect all the samples not a time in every month. For this reason the sample size of all species are not same in a month. On the other hand, the oyster take time for maturation and gonadal development. We

take sample for a year, there is no more enough time to find mature female gonad which we want.

CHAPTER SIX

CONCLUSIONS

Crassosstrea sp can be found in a number of locations in the Bay of Bengal's east and southwest shores. *Crassostra* sp. at Moheskhali channel had shown a yearly major gametogenic cycle with two peaks at June-July and November-December indicating mature stage. A true resting phase also observed in March. Gonado-somatic index of female was higher than male. Water quality is also important which can affect the process of gonad development. Mollusk culture, particularly mussels and oysters, is becoming a more urgent choice for coastal aquaculture. During the gathering of oysters, a substantial amount of meat is lost, which can be avoided by better exploitation, transportation, and storage methods. Quality supply at local restaurants can also be ensured with proper processing technologies. Culture development is required to decrease stress on natural oyster, assure shellfish biodiversity protection, and maintain harmony with the future fast-growing business. Culture development will also boost the small-scale fisherman's livelihood by providing job opportunities.

CHAPTER SEVEN

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