

Gonad Development and Sexual Maturity of *Gomphina (Macridiscus) veneriformis* (Lamarck, 1818) (Bivalvia: Veneridae) in the East Sea of Korea

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ABSTRACT

Gonad development, the reproductive cycle, first sexual maturity and size at 50% of group sexual maturity (the biological minimum size) of *Gomphina (Macridiscus) veneriformis* were investigated for clams collected from the coastal waters of Donghae City, the East Sea of Korea by histological, and morphometric analysis. Monthly variations of the gonad index showed a pattern similar to that of the reproductive cycle. The reproductive cycle with the gonad developmental stages in female and male *G. (M.) veneriformis* can be classified into five successive stages: early active stage (December to March), late active stage (March to June), ripe stage (June to July), partially spawned stage (June to August), and spent / inactive stage (September to December). The spawning period continued from June to August, with a peak between July and August when the seawater temperature exceeds 20°C. The percentages of first sexual maturities of female and male clams ranging from 25.1 to 30.0 mm were 56.3% in females and 61.1% in males, and for clams over 30.1 mm shell length, it was 100%. Shell lengths at 50% of group sexual maturity (biological minimum size, RM₅₀) were 27.71 mm in females and 26.31 mm in males. Because harvesting clams < 26.31 mm in shell length could potentially cause a drastic reduction in recruitment, a measure indicating a prohibitory fishing size should be taken for adequate fisheries management.

Key words: *Gomphina (Macridiscus) veneriformis*, developmental stage, size at 50% of group sexual maturity

INTRODUCTION

The equilateral venus, *Gomphina (Macridiscus) veneriformis* (Lamarck, 1818), is distributed along the coasts of Korea, China, and Japan (Kweon *et al.*, 1993; Min *et al.*, 2004). More specifically, in Korea, this species is mainly found in silty sand at the subtidal

zone on the east coast of Korea (Min *et al.*, 2004), and is one of the most commercially important edible clams. Because of past over-harvesting, it has been denoted as a fisheries resource that should be managed using a more reasonable fishing regimen. For the propagation and management of living natural resource, it is important that we understand its reproductive ecology with regard to the gonad developmental stage, spawning season, the size at 50% of group sexual maturity, and the prohibitory fishing size.

To date, regarding *Gomphina* spp., there have been several studies on aspects of reproduction, including the spawning season, artificial fertilization and egg development (Lee, 1976), gonadal development and reproductive cycle (Lee *et al.*, 1999), and germ cells and the accessory cells during spermatogenesis (Chung

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et al., 2010), and on aspect of ecology, including culture (Lee, 1976) and age and growth (Hwang and Hwang, 1981; Jung, 2011), and on aspects of toxicology and physiology (Pank and Lee, 2008, 2009, 2010, 2011). Although there are some reproductive ecological studies, there are still gaps in our knowledge on reproductive ecology associated with the gametogenic cycle and the spawning season. In general, it is well-known that the reproductive cycle, spawning periods and size at first sexual maturity of this species vary with latitudinal gradients (locations) of the world. Despite the studies referred above, in particular, the reproductive cycle, spawning period and the size at first sexual maturity that reported in the previous studies, is somewhat inaccurate. In particular, the size at 50% of first sexual maturity in both sexes which is participated in reproduction should be clarified for natural resource management, Park *et al* (2003) reported the size at 100% of first sexual maturity and several reproductive ecology. However, there are many problems to solve. The accurate reproductive cycle of this species according to the seasonal changes should be clarified for the prediction of changes in natural resources in the future.

Little information on the accurate spawning period and the size at 50% of sexual maturity (RM_{50}) (the biological minimum sizes) and accurate fishing size in female and male individuals of this species is available. The knowledge of the reproductive cycle, accurate spawning period in female and male individuals of this species will provide necessary information for the determination of age and recruitment period. Additional information on sizes at the rate (50%) of group sexual maturity (the biological minimum size) and the fishing size of this species would be very useful for propagation, aquaculture and resource management.

In particular, information on the size at which individuals reach 50% of group sexual maturity could be useful in determining a prohibitory measure for adequate natural resource management. Therefore, the purpose of this study is to describe the natural reproductive cycle, spawning period, size at 50% of group sexual maturity and the prohibitory fishing size

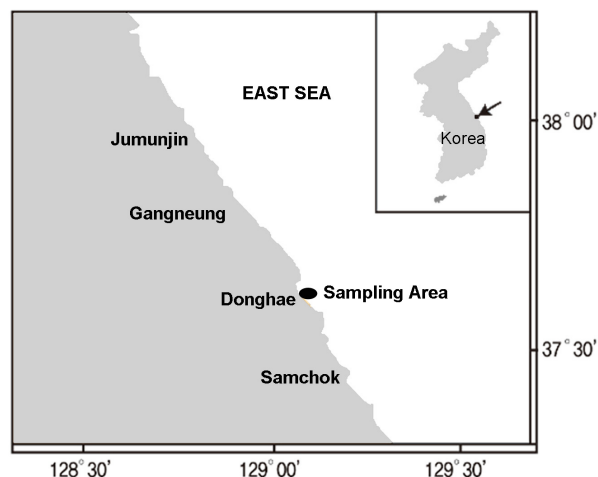


Fig. 1. Map showing the sampling area.

of *G. (M.) veneriformis*, and results will be useful for improved fisheries management of this species.

MATERIALS & METHODS

1. Gonadal Development and Reproductive Cycle by Histological Observations

1) Sampling Methodology

Specimens of the equilateral venus clam, *G. (M.) veneriformis*, were collected monthly by dredging in the subtidal zone of Donghae City, Gangwon-do, the East Sea of Korea, from January to December, 2007 (Fig. 1). A total of 683 clams ranging from 24.0 mm to 58.5 mm lengths were collected during the study. Equilateral venus clams were transported alive to the laboratory and shell length and total weight were measured. Unpublished data of seawater temperatures and salinity measured daily at 10:00 a.m. at East Sea Fisheries Research Institute, National Fisheries Research and Development Institute were used for this study.

2) Histological Preparations of the Gonadal Tissues

For light microscopic examination of histological preparations, a total of 425 individuals were used for histological analysis of the gonads. Gonadal tissues were removed from shells and preserved in Bouin's fixative for 24h. They were then washed with running

tap water for 24h. Tissues were then dehydrated in alcohol and embedded in paraffin molds. Embedded tissues were sectioned at 5-7 μm thickness using a rotary microtome. Sections were mounted on glass slides, stained with Hansen’s hematoxylin-0.5% eosin, and examined using a light microscope (Zeiss Axiovert 10 microscope).

3) Gonad Index (GI)

To explore the spawning season by qualitative analysis (histological observations), a total of 425 clams were calculated for monthly changes in the gonad index (GI). The mean gonad index (GI) in female and male individuals were calculated using a modification of Mann’s method (1979). Each histological section of gonadal tissues was also examined in details to assess the stage of gonadal development. Staging criteria of 1 to 5 were employed for Spent / inactive stage (S1 = 1), Partially spawned stage (S2 = 2), Early active stage (S3 = 3), Late active stage (S4 = 4) Ripe stage (S5 = 5), These categories are only approximations of gonadal development because it is a continuous process and distinctions between stages are not always clear. The monthly gonadal index (GI) for both sexes was determined by multiplying the number of specimens ascribed to each category score, summing all those values and dividing this figure by the total number of clams analyzed.

$$GI = (N \times RVS1) + (N \times RVS2) + (N \times RVS3) + (N \times RVS4) + (N \times RVS5).$$

Total N observed by month

Where, N: number of individuals, RVS: ranking value by stage.

4) Size at the rate (%) of first sexual maturity by Light Microscopical Observation

To clarify the sizes (shell lengths) of approximately 50% and 100% of sexual maturities that are participated in reproduction after sexual maturation, a total of 305 clams (148 female individuals and 157 male individuals) of gonadal histological preparations (17.9-58.5 mm in shell length) were examined the size at the rate (%) of sexual maturity by histological

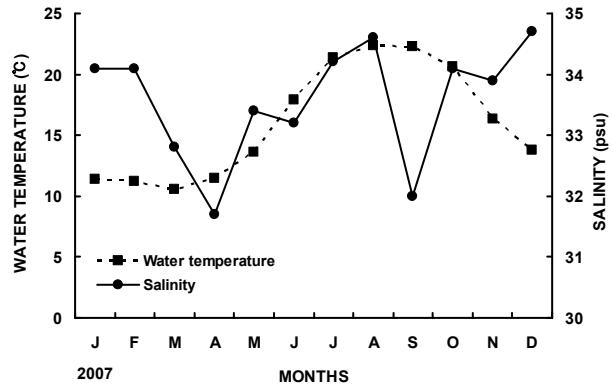


Fig. 2. Monthly changes in seawater temperatures and salinities at the sampling area from January to December 2007.

observations from January to December 2007.

The percentage (%) of sexual maturity = No. of mature individuals x 100 / No. of total individuals. The sizes (shell length (mm)) at approximately 50% and 100% of sexual maturity in female and male sexes were compared respectively by gonadal histological observations.

5) Biological minimum size (Size at 50% of group sexual maturity, RM₅₀)

To calculate the size at the rate (50%) of sexual maturity after fitting the rate of sexual maturity to an exponential equation, the size equivalent to the size at 50% of group sexual maturity was estimated to be the sexually mature length of the population. The exponential equation of the rate of group sexual maturity (exponential equation used by Son & Chung, 2009) is as follows: $RM = 100/1 + \exp^{(a-bx)}$, where, RM: rate of sexual maturity; a,b: constants, x: shelllength.

Results

1. Monthly changes in seawater temperatures and salinities

Monthly changes in seawater temperatures and salinities in Donghae City, in Gangwon-do, Korea are showed in Fig. 2. Monthly changes in seawater temperatures began to gradually increase in April and reached a maximum (22.4°C) in August, thereafter, gradually decreased from September to February.

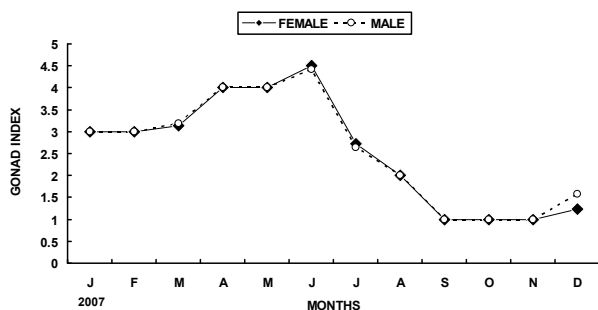


Fig. 3. Monthly changes in the mean gonad index of *Gomphina (Macridiscus) veneriformis* from January to December 2007.

Monthly changes in salinities began to gradually increase in May and reached a maximum (34.7 psu) in December. Thereafter, gradually decreased from January and reached a minimum (31.7 psu) in April. On the whole, monthly changes in salinities (psu) during the year showed relatively small range. In general, seawater temperatures and salinities gradually increased from June to August are closely related with the spawning season of this species.

2. Position and External Morphology of the Gonads

G. (M.) veneriformis, is a dioecious organism. General positions and morphologies of the ovaries and testes of this species are similar to those of other bivalves. The ovary and testis are located between the digestive diverticular and the outer fibromuscular layers, which are compacted by the fibrous connective tissues and muscle fibers. As the ovary and testis matured, the positions of the gonads are occupied from the subregion of mid-intestinal glands in the visceral cavity to the reticular connective tissues of the foot. The ovary is composed of a number of oogenic follicles, and the testis comprises a number of the acini. Mature ovaries were of the same yellowish-white color as mature testes. Therefore, the sex of individuals could not be easily distinguished by external features. However, if the ripe ovary and testis were slightly scratched with a razor, a number of ripe yellowish brown eggs or ripe milky white sperm flowed out readily. Therefore, sex could be easily distinguished by dissection. After spawning and discharging, the ovary and testis degenerated, and they became difficult to

distinguish their sexes by external color or dissection of the gonads.

3. Monthly Changes in the Gonad Index (GI)

To explore the spawning season of *G. (M.) veneriformis* by qualitative analysis (histological observations), we calculated monthly changes in the gonad index during the year of this species. Monthly changes in the gonad index (GI) of in females and males were shown in Fig. 3. The GI values in female and male individuals began to gradually increase in March, and reached a maximum (GI 4.5 in females and 4.41 in males) in June, thereafter, gradually decreased from July to August when spawning occurred. Thereafter, the GI values in both sexes reached a minimum from November to January (GI 1.0 in female and male individuals). Therefore, monthly changes in the GI in both sexes in 2007 showed a similar pattern with gonadal development and the spawning period of this species in Korea showed once a year.

4. Gonadal Development and the Reproductive Cycle

Based on morphological features and sizes of the germ cells and accompanying cells, the reproductive cycle in female and male individuals of this species can be classified into five successive stages: early active, late active, ripe, partially spawned, and spent/inactive stages (Figs. 4, 5). The stages and the criteria used in defining them are as follows (Mann, 1979).

Stage I (early active stage): In females, oogonia and previtellogenic oocytes propagated along the follicular walls of the follicles in the ovary. The oogonia and early vitellogenic oocytes were 9-11 μm and 15-25 μm in diameter, respectively. At this stage, the lumina of the oogenic follicles were empty (Fig. 5A).

In males, the spermatogonia and spermatocytes were 7-8 μm and 6-7 μm in diameter, respectively. They appeared along the acinus wall of acini in the testis (Fig. 6A). Female and male individuals in the stage 1 (early active stage) appeared from December to March when seawater temperatures were relatively low (about 10°C) (Figs. 2, 4).

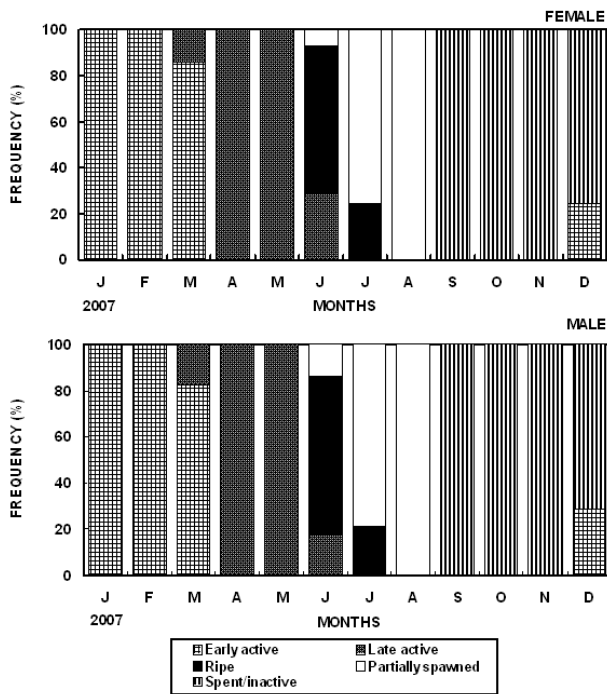


Fig. 4. Frequency of gonadal phases in *Gomphina (Macridiscus) veneriformis* through the gametogenic cycle.

Stage II (late active stage): In females, a number of the early vitellogenic oocytes, ranging from 30 to 40 μm in diameter, appeared in the oogenic follicles. Each early vitellogenic oocyte formed an egg-stalk connected to the follicular wall. When late vitellogenic oocytes grew to 41-50 μm in diameter, they had a large germinal vesicle and egg-stalk attached to the follicular wall (Fig. 5B).

In males, a few spermatogonia and a number of spermatocytes (5-6 μm) and spermatids measuring about 3 μm in diameter appeared in the acini. At this time, a small number of spermatozoa begin to transform into differentiated spermatozoa in the center of the lumen of the acinus (Fig. 6B). Female and male individuals in the stage II (late active stage) were found from March to June when seawater temperatures were gradually increased (Figs. 2, 4).

Stage III (ripe stage): In females, the majority of maturing oocytes grew to 50-60 μm in diameter and

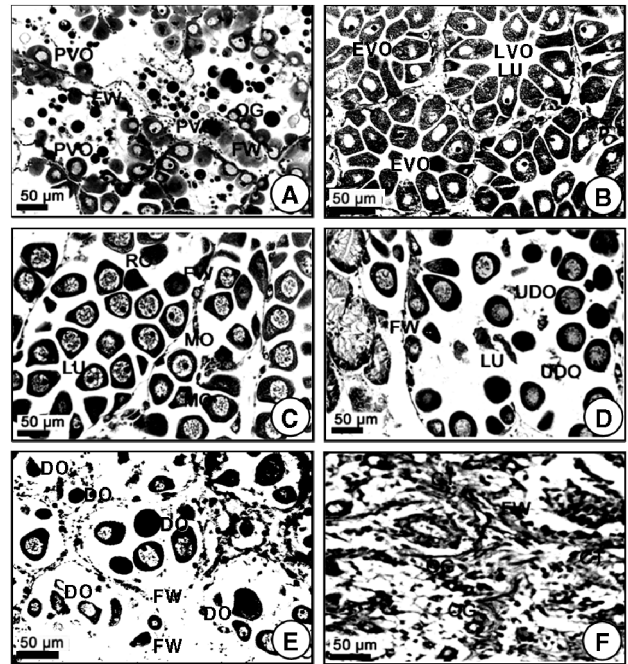


Fig. 5. Photomicrographs of oogenic follicles in various gonadal phases in female *Gomphina (Macridiscus) veneriformis*. **A:** Transverse section of oogenic follicles in the early active stage; **B:** Section of follicles in the late active stage; **C:** Section of follicles in the ripe stage; **D:** Section of follicles in the partially spawned stage; **E:** Section of follicles in the spent stage; **F:** Section of follicles in the inactive stage. Abbreviations: CT, connective tissue; DO, degenerating oocyte; EVO, early vitellogenic oocyte; FOL, follicle; FW, follicular wall; LU, lumen; LVO, late vitellogenic oocyte; MOC, maturing oocyte; OG, oogonium; PVO, previtellogenic oocyte; RO, ripe ovum; UDO, undischarged oocyte

the ripe ova were 60-65 μm in diameter, becoming round or oval in shape. They were located in the center of the lumen. Each ripe ovum contained a large germinal vesicle, and its cytoplasm was filled with a large number of yolk granules. At this time, in particular, Each ripe ovum was surrounded with a thick gelatinous membrane (Fig. 5C).

In males, a number of spermatids began to transform into differentiated spermatozoa in the centre of the lumen, and numerous spermatozoa appeared in the center of the lumen of the acinus (Fig. 6C). Female and male individuals in the stage III (ripe stage) were found between June and July when sea water temperatures were gradually increased over 18°C (Figs. 2, 4).

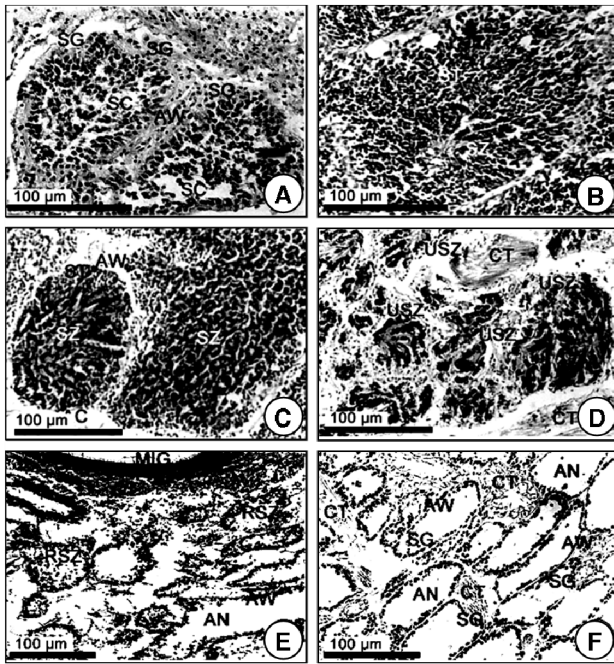


Fig. 6. Photomicrographs of the acini in various gonadal phases in male *Gomphina (Macridiscus) veneriformis*. **A:** Transverse section of the acini in the early active stage; **B:** Section of the acini in the late active stage; **C:** Section of the acini in the ripe stage; **D:** Section of the acini in the partially spawned stage; **E:** Section of the acini in the spent stage; **F:** Section of the acini in the inactive stage. Abbreviations: AN, acinus; AW, acinus wall; CT, connective tissue; DSZ, degenerating spermatozoon; RSZ, residual spermatozoon; SC, spermatocyte; SG, spermatogonium; ST, spermatid; SZ, spermatozoon; USZ, undischarged spermatozoon.

Stage IV (partially spawned stage): In females, most ripe ova were discharged from the oogenic follicles, although a few undischarged ripe ova as well as vitellogenic oocytes remained (Fig. 5D). The lumen of the follicle was empty.

In males, because over 50% of the spermatozoa have been discharged, the lumen of the acini became empty, but undischarged spermatozoa as well as spermatids remain in the lumen of the acinus (Fig. 6D). Female and male individuals in the stage IV (partially spawned stage) appeared from June through August, with the main spawning event occurring from July to August when seawater temperatures were higher than 22°C (Figs. 2, 4).

Stage V (spent / inactive stage): In females, after spawning, each follicle contracted and degenerated, and the undischarged oocytes in the lumen of the oogenic follicle underwent cytolysis. Gamete atresia were resorbed. At the same time, a rearrangement of the connective tissues were observed in the follicle (Figs. 5E, F).

In males, the few remaining spermatozoa and spermatids degenerated and gamete atresia were resorbed, thereafter, a rearrangement of the connective tissue occurred in the acini (Figs. 6E, F).

Female and male individuals in the stage V (spent / inactive stage) appeared from September to December when sea water temperatures were gradually decreased and relatively low (Figs. 2, 4).

5. Size at First Sexual Maturity

A total of 305 (148 females and 157 males) individuals of *G. (M.) veneriformis* were investigated histologically to determine the shell lengths of clams that reach maturation and participate in reproduction from May (before spawning) to late September (after spawning). It was found that the percentages of sexual maturity of smaller female and male individuals ranging from 17.9-20.0 mm in shell length was 0%. The percentages of sexual maturity of female and male individuals ranging from 20.1-25.0 mm in shell length were 28.6% and 31.3%, respectively. The percentages of first sexual maturity of female and male clams ranging from 25.1-30.0 mm in shell length were 56.3% and 61.1%, respectively. These results mentioned above (the percentages of first sexual maturity in female and male clams were 56.3% and 61.1%) were over 50%. And the percentages of first sexual maturity of female and male individuals of shell lengths greater than 30.1 mm were 100%.

6. Biological Minimum Size (Size at the rate (50%) of sexual maturity (RM₅₀))

For the management of living natural resources, we calculated the sizes at 50% of sexual maturities (RM₅₀) of this species in order to understand the biological minimum sizes in female and male individuals. Commonly, we regard the size at the rate (50%) of

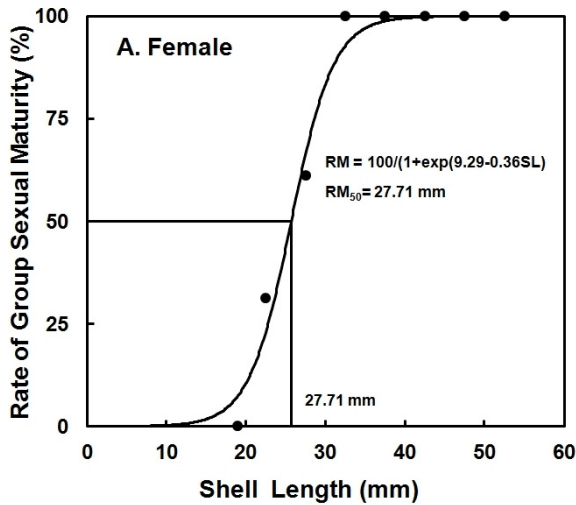


Fig. 7. Relationship between the rate of Group sexual maturities (%) and shell length (mm) in female *Gomphina (Macridiscus.) veneriformis*. A, RM₅₀ in female.

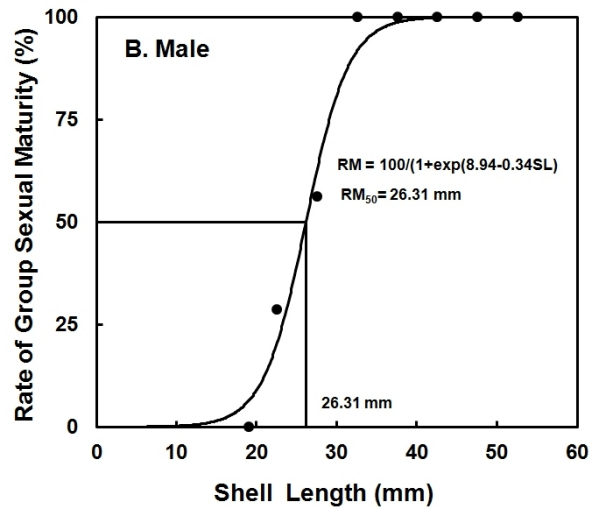


Fig. 8. Relationship between the rate of Group sexual maturities (%) and shell length (mm) in male *Gomphina (Macridiscus.) veneriformis*. B, RM₅₀ in male.

group sexual maturity as the biological minimum size. Sometimes its size (RM₅₀) provided an important information for fishing prohibitory measure as basic data. Shell lengths of sexually mature equilateral venus clam (sizes at 50% of sexual maturity, RM₅₀) that fitted to an exponential equation were 27.71 mm in females (Fig. 7) and 26.31 mm in males (Fig. 8).

DISCUSSION

1. Gonad index

To explore the spawning season of this species by qualitative analysis (histological observations), we calculated monthly changes in the gonad index and checked a unimodal or bimodal cycle during the year of this species. The gonad indice (GI) were monthly calculated based on the gonad developmental stages of histological sections by qualitative reproductive analysis (histological observations). The gonad index (GI) of this species began to increase in spring months and reached a maximum in June when the water temperature rapidly increased. Thereafter, the GI values then showed a gradual decrease because of spawning, with the increase of water temperatures. Chung *et al.* (2007) reported that the high average values of the GI coincided with gonadal maturity, and the minimal average value following high average

values were considered an indication of spawning. Accordingly, variations in the GI showed a close relationship with gonadal development and gonadal activity. In this study, monthly variations in the GI by qualitative analysis showed a maximum showing an unimodal cycle during the year. Therefore, monthly changes in the GI in both sexes in 2007 showed a similar pattern with gonadal development and the spawning period of this species in Korea showed once a year, indicating a unimodal cycle (Fig. 3).

2. Gonadal Development and Maturation

Exogenous factors have recently been suggested as controls for gonadal development and maturation in marine bivalves. Of several exogenous factors, water temperature and food availability seem to be particularly important. Sastry (1966, 1968) stated that these and other factors (salinity, day length, etc) probably interact with endogenous factors (neuroendocrine activity) in a complex manner to control the initiation of gametogenesis. Sastry (1968) stated that sea water temperature acts as a triggering stimulus for the initiation of the germ cell growth phase. The water temperatures required for activating the growth of germ cells at the beginning of oogenesis and spermatogenesis and for attaining maturity

ultimately limit the annual period of gonad activity and gametogenesis in the natural environment.

In this study, gamete differentiation of *G. veneriformis* began in the winter-early spring seasons, and reached maturity in the population between June and July when water temperatures were increased. After basic metabolic requirements are satisfied, gonad activity and gametogenesis of this species occur under temperature conditions that allow nutrients mobilization to the gonads (Sastry, 1966). The periods of food abundance and of gonad development of *G. veneriformis* are nearly coincident gonad growth and gametogenesis in spring coincided with peak food levels, although food concentrations remained high throughout the summer months (Kim, 2005). Therefore, it is assume that if food and temperature criteria are met, growth of germ cells is initiated in conjunction with the transfer of nutrients from digestive diverticular to the gonad. However, it is assumed that the amount of nutrients mobilized for the gonad maturation depends not only on the food level, but also on the water temperature and the basic metabolic requirements of the clams.

In Korean coastal waters, growth and production of bivalves is relatively high from spring to early summer seasons (Chung *et al.*, 1994; Kim, 2005) due to the abundance in phytoplankton. Thus, abundant food supply (e.g., bivalves) is available to *G. veneriformis* during the period of gonadal development and maturation. Therefore, it is suggested that gonadal development and maturation of the Korean *G. veneriformis* is closely related to temperature change and food availability. Fretter (1984) observed that in temperate zones, the seasonal temperature fluctuation associated with changing illumination is a controlling factor in gametogenesis. In consequence, gonadal development and maturation of this species may be retarded under low illumination, due to the decrease in food availability caused by diminished primary production of phytoplankton.

3. Breeding pattern and the spawning period

G. melanaeigis and *G. veneriformis* are mainly found in the same habitats in the coastal waters of Donghae

and Jumunjin, Gangwon-do, Korea. To date, they are well-known that it is hard to distinguish by the external features because the morphologies of two species are very similar. Therefore, above all, it is important to clarify their genetic characteristics and the reproductive cycle (or the spawning period) of two species for classification and the natural resource management.

Booolootian *et al.* (1962) placed mollusks into three large categories: (1) year-round breeders, (2) winter breeders, and (3) summer breeders. We found that *G. veneriformis* and *G. melanaeigis* belong to the summer breeder class because of their reproductive patterns. Rand (1973) reported that breeding strategy vary with latitudinal gradients. Northern climates are characterized by a single synchronous spawning per year, temperate climates by two spawning seasons and tropical ones by year-round spawning. In case of different local populations of the same species, the number of spawning seasons by qualitative reproductive analysis (histological observations) in most bivalves occur once a year in the northern districts of Tokyo Bay, Japan (Kurashige, 1943; Momoyama and Iwamoto, 1979), while twice a year in the southern districts of Tokyo Bay, Japan (Ko, 1957; Tanaka, 1954).

Regarding the spawning period of *Gomphina* species in Korea, recently, Lee *et al.* (1999) and Park *et al.* (2003) reported that the spawning period of *G. melanaeigis* in the coastal waters of Jumunjin, Gangwon-do, Korea was once a year between July and August. Park *et al.* (2003) reported that the spawning period of *G. veneriformis* in the coastal waters of Gangneung, Gangwon-do, Korea was once a year between July and August.

However, in this study, the spawning period of *G. veneriformis* by qualitative analysis (histological observations) was once a year during the period of early June to August, 2007 in the coastal waters of Donghae, Gangwon-do, Korea.

Judging from the results mentioned above, the beginning of spawning of *G. veneriformis* in coastal waters of Donghae, Gangwon-do, Korea was somewhat one month faster than those of *G. melanaeigis* in

Table 1. The shell length at first sexual maturity in *Gomphina (Macridiscus) veneriformis* from May to September, 2007

Shell length (mm)	Female		Male	
	Number (ind.)	Mature (%)	Number (ind.)	Mature (%)
17.9-20.0	3	0.0	5	0.0
20.1-25.0	14	28.6	16	31.3
25.1-30.0	16	56.3	18	61.1
30.1-35.0	24	100.0	26	100.0
35.1-40.0	23	100.0	27	100.0
40.1-45.0	27	100.0	29	100.0
45.1-50.0	25	100.0	24	100.0
50.1-55.0	12	100.0	10	100.0
55.1-58.5	4	100.0	2	100.0
Total	148		157	

*ind. means individuals.

Jumunjin, Gangwon-do, Korea reported by Lee *et al.* (1999) and one month faster than those of *G. veneriformis* in the coastal waters of Gangneung, Gangwon-do by Park *et al.* (2003). Thus, some local variations and timing of spawning of two clams might be related to the geographical differences in the water temperatures, time of the food production (phytoplanktons), and some other environmental factors (Ko, 1957; Momoyama and Iwamoto, 1979).

4. Size at first sexual maturity

As shown in Table 1, it was found that the percentages of first sexual maturity of smaller female and male individuals ranging from 17.9-20.0 mm in shell length was 0%, and those individuals were in the early active stage, characterized by a small number of oogonia and the appearance of previtellogenic oocytes. in the ovary and spermatogonia and spermatocytes in the testis. It is supposed that their sizes at sexual maturities could not have been reached until late August when spawning was completed. In addition, the percentages of first sexual maturity of female and male individuals ranging from 20.1-25.0 mm shell length were 28.6% and 31.3%, respectively. In particular, during the period between June and August, when spawning was observed among older

individuals. however, most younger female and male clams had a small number of undeveloped germ cells. A number of developing germ cells and a small number of mature germ cells were present in the ovaries and testes. It is supposed that small size of clams could not have been reached sexual maturity until late August when spawning of several mature individuals was completed. In addition, the percentages of first sexual maturity of female and male individuals ranging from 25.1-30.0 mm in shell length were 56.3%, and 61.1%, respectively, and those individuals were in the early active, late active, ripe, and partially spawned stages during the breeding season. In contrast, the percentages of first sexual maturity of female and male individuals of shell length greater than 30.1 mm were 100%, and that those individuals were in the late, ripe, partially spawned, and spent / inactive stages. Accordingly, it is assumed that most female and male individuals can reach full maturity by late August if they were larger than 30.1 mm in shell length.

Park *et al.* (2003) studied first sexual maturity of *G. veneriformis* in the coastal waters of Jumunjin, Gangwon-do, Korea from March 2001 to February 2002, A total of 145 clams (74 females and 71 males), ranging from 31.1 mm to 64.9 mm in total length,

were used for the experiments of first sexual maturities in both sexes. However, they could not clarify the size at 50% of first sexual maturities because the sizes of the specimens were too large and over one year of age. They only clarified the sizes at 100% of first sexual maturities. Therefore, it is assumed that they should be clarified their sizes (shell lengths) for the natural resource management.

5. Size at 50% of group sexual maturity (RM₅₀) (= Biological minimum size)

In this study, percentages of first sexual maturity in female and male individuals ranging from 25.1-30.0 mm in shell length were 56.3% and 61.1%. Therefore, we can not know the exact sizes at 50% of sexual maturities in female and male individuals of this species. In general, the size of 50% of sexual maturity (RM₅₀) is regarded as the biological minimum size.

For the measurement of the the sizes at 50% of group sexual maturity (biological minimum size), therefore, we have to calculate the sizes at 50% of first sexual maturity (RM₅₀) which are fitted to von Bertalanffy's equation. In particular, information on the size at 50% of group sexual maturity (size of RM₅₀ = biological minimum size) is very important, and can determine a prohibitory fishing size for adequate natural resources management through determination of size at 50% first sexual maturity. According to data for the size at 50% of group sexual maturity (size of RM₅₀) of *G. (M.) veneriformis*, shell lengths at the rate (50%) of group sexual maturity (RM₅₀) were 27.71 mm in females and 26.31 mm in males, respectively. According to the results of age determination of *G. (M.) veneriformis* reported by Jung (2011), age class (Yr) and shell length (mm) are as follows: 1 Yr = 17.53 mm; 2 Yrs = 29.59 mm; 3Yrs = 38.97 mm; 4 Yrs = 47.60 mm; 5Yrs = 55.44 mm; 6 Yrs = 58.32 mm.

Accordingly, those sizes ranging from 26.31 mm in shell length in males to 27.71 mm in females are considered to be about 2 years of age. Therefore, we assume that both sexes of this population begin reproduction about two years of age. In terms of natural resource management, the present study suggests that harvesting clams less than 26.31 mm in

shell length (< 2 years of age) can potentially lead to a drastic reduction in recruitment. A fishing prohibitory measure should be taken for adequate natural resources management.

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