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Sexual Maturation, Sex Ratio and Hermaphroditism of the Pacific Oyster, *Crassostrea gigas*, on the West Coast of Korea

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Monthly changes of the gonad follicle index (GFI), reproductive cycle, egg-diameter composition, first sexual maturity of the Pacific oyster, *Crassostrea gigas*, were studied based on the samples which have been collected from the intertidal zone of Poryong west coast of Korea, from January to December, 1996.

C. gigas, is dioecious, while a few individuals are alternatively hermaphroditic.

Monthly variation of gonad follicle index (GFI) used for determination of spawning period, coincided with the reproductive cycle. GFI increased from April when seawater temperatures gradually increased and reached the maximum in May. And then, GFI sharply decreased from June to September due to spawning.

Reproductive cycle of this species can be divided into five successive stages: in females, early active stage (March to April), late active stage (April to May), ripe stage (May to August), partially spawned stage (June to September) and spent/inactive stage (September to February); in males, early active stage (February to March), late active stage (April to May), ripe stage (May to September), partially spawned stage (June to September) and spent/inactive stage (September to February).

The diameter of fully mature eggs are approximately 50 μm. Spawning occurred from June to September, and two spawning peaks were observed in June and August when the seawater temperature was above 20℃.

Percentages of the first sexual maturity of males of 20.1~25.0 mm in shell height were over 50%, while those of females of 25.1~30.0 mm in shell height were over 50%. All the males of >30.1 mm and all the females of >35.1 mm completed their first sexual maturity. The results suggest that *C. gigas* has a protandry phenomenon.

Sex ratios of 919 oysters observed were 453 females (49.29%), 429 males (46.68%), 16 hermaphrodites (1.74%), and 21 indeterminate individuals (2.29%). In age class I, sex ratio of males were 64.00%, thus, a higher percentage than that of females. It was noted that 64.00 % of the young males (age class I) were more functional than females in age class I, but 2~3 year-old oysters showed higher percentage of females.

Percentages of hemaphrodites in 2~3 year classes were relatively higher than those in other year classes.

Histological pattern of hermaphrodites can be divided into two types: Type I (hermaphrodite having a number of newly formed developing oocytes on the oogenic tissues within a degenerating spermatogenic follicle after discharge of numerous spermatozoa) and Type II (hermaphrodite having two separate follicles in the same gonad).

Key words: *Crassostrea gigas*, gonadal development, reproductive cycle, first sexual maturity, sex ratio, hermaphroditism

Introduction

The Pacific oyster, *Crassostrea gigas*, is present along the coasts of Korea, China, Japan and New Zealand, etc. In Korea, it has been found on the rocks in the intertidal zone of all of Korean coasts (Yoo, 1972, 1979; Yoo et al., 1971; Kwon et al., 19 93), and it is one of the commercially important bivalves in Korea.

So far, there have been many studies on the reproductive cycles, spawning periods and repoductive strategies of *Crassostrea* spp. (Galtsoff, 1964; Dinamani, 1974, 1987) by histological investigation, on

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the aspect of aquaculture (Davies et al., 1986; Paynter and Dimichele, 1990; Roland and Brown, 1990; Figueras, 1991; Goulletquer et al., 1994).

With reference to the sex ratio of the oysters, particularly in *Crassostrea virginica* (Galtsoff, 1964) and *C. glomerata* (Dinamani, 1974), it is well-known that the young individuals (1 year class) are more functionally males than females, but the older classes showed a higher percentage of females, and they showed a protandry phenomenon as a reproductive strategy.

Recently, there have been studied on the aquaculture and reproduction of other oviparous oyster species except for *C. gigas* investigated by many authors (Ford et al., 1990; Choi, 1992; Mann et al., 1994; Brousseau, 1995), but little is known of its reproductive cycle of the Pacific oyster, *C. gigas*, in Korea and Japan.

Particularly, there is no study on gonad development, reproductive cycle, first sexual maturity, sex ratio and hermaphroditism of *C. gigas* using histological methods. Therefore, to get some detailed informations, it is necessary to investigate on some characteristics of natural intertidal population of this species in terms of gonadal development, spawning and sexual maturity.

Above all, understanding the time and duration of spawning, reproductive cycle and first sexual maturity of this species are supposed to be one of the most important tasks for the successful culture of the species.

The main purpose of the present study is to understand gonadal development, reproductive cycle, first sexual maturity, sex ratio and hermaphroditism of the Pacific oyster, *C. gigas*, among natural intertidal populations.

Materials and Methods

Samples of the Pacific oyster, *Crassostrea gigas*, were collected monthly in the intertidal zone of Poryong, Chungcheongnamdo, Korea, from January to December, 1996 (Fig. 1). A total of 919 oysters of 15.4~61.4 mm in shell height were used for the present study. The oysters were transported alive to the laboratory, shell lengths and heights were measured, and total weights were weighted.

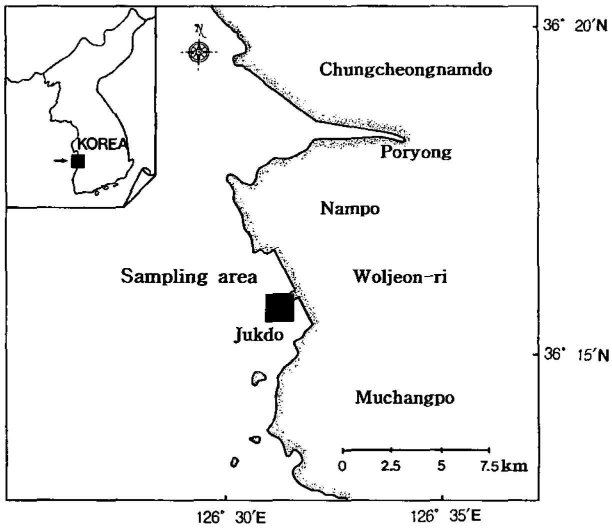

Fig. 1. Map showing the sampling area.

Gonad follicle index (GFI)

To investigate the spawning period, mean gonad follicle index (GFI) was calculated. In each microscopic preparation, representing an anterior-posterior section through the oyster, the percent gonad tissue relative to the non-gonadal tissue, and the average gonad density (ratio of area in follicle to standard area (1 mm^2) in the gonad) were determined in three serial sections, using the count point technique (Tinsman et. al., 1976). To provide a measure of gonadal development the GFI (percent gonad × gonad density) was caculated.

Relative frequency distributions of the egg diameters

To investigate size frequency distributions of egg in the ovary, ca. 1,000 eggs with centrally placed nuclei were measured monthly, and the egg diameter data were expressed as frequency histogram according to Pearse (1965) with some modification.

Gonadal phase and the reproductive cycle by histological observation

For histological study, cross sections were made in the anterior, middle, posterior regions of the visceral mass (gonad, digestive diverticula and gastrointestinal tract) of the oysters. The tissue were then prepared for histological examination (Brousseau, 1995).

Changes in cytological characteristics were used to categorize specimens into one of five arbitary gonadal stages. The five stages used by Holland and Chew (1974) for the manila clam were adopted, but the criteria were modified: one stage, the spent, was modified to spent/inactive stage for sexually undifferentiated specimens (Mann, 1979a). The gonadal phases can be classified into five successive stages: early active stage, late active stage, ripe stage, partially spawned stage, and spent/inactive stage

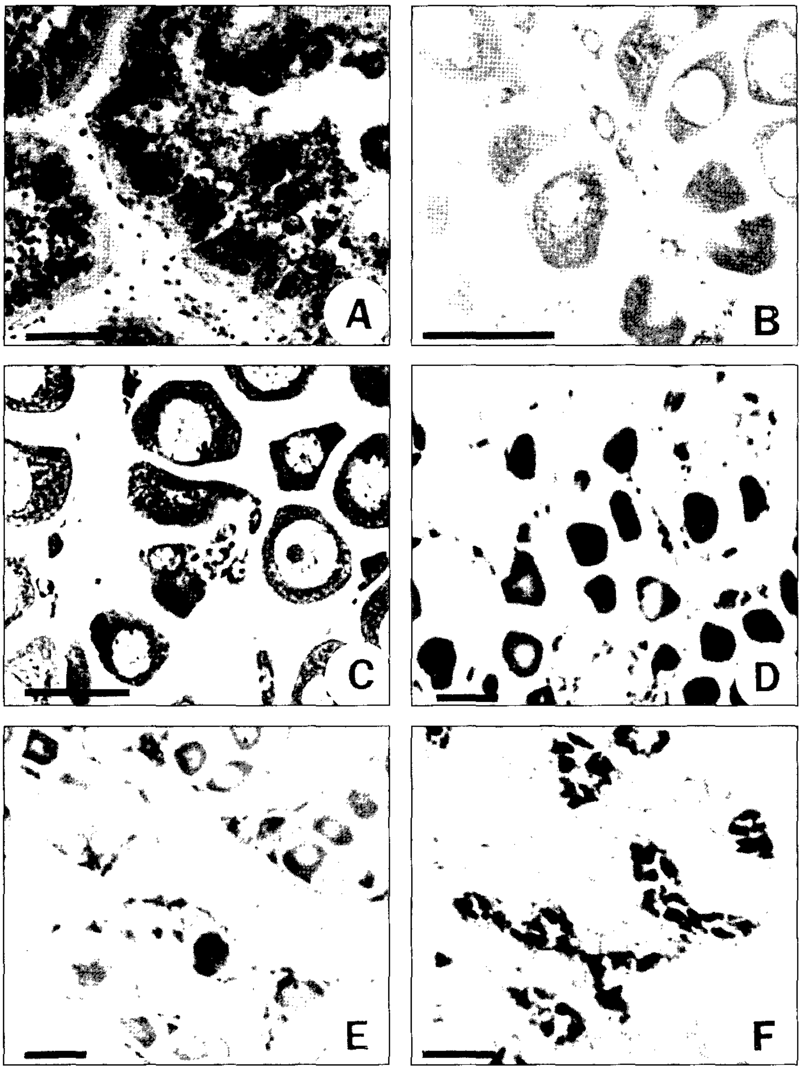

Fig. 2. Photomicrographs of the gonadal phase of female oyster, *Crassostrea gigas*. A, transverse section of the oogenic follicle in the early active stage, scale bar=30 μm; B, section of the oocytes in the late active stage, scale bar=50 μm; C, section of fully ripe oocytes in the ripe stage, scale bar=50 μm; D, section of the oogenic follicles in the partially spawned stage, scale bar=30 μm; E, section of follicles in the spent stage, scale bar=30 μm; F, section of follicles in the inactive stage, scale bar=100 μm.

(Fig. 2 and 3). The criteria used in defining the stages are as follows:

Early active stage

In females, gonad proliferation started, as seen by increasing numbers of discernible oogonia along the follicular wall, and oocytes were still small; mean oocyte diameter was < 20 μm (Fig. 2A).

In males, oyster characterized by the expansion of the follicle and the appearance of well-defined spermatogonia and spermatocytes (Fig. 3A).

Late active stage

In females, the connective tissue is decreased, many young oocytes were attached to the follicular wall and the mean oocyte diameter is between 35 45 μm. Some free oocytes measuring about 50 μm in diameter are present in the lumen (Fig. 2B).

In males, spermatogonia, spermatocytes, spermatids, and spermatozoa coexisted in the spermatogenic follicle (Fig. 3B).

Ripe stage

In females, half or more than half of the oocytes are free in the lumen and had a polygonal configuration. The mean oocyte diameter is about 50 μm (Fig. 2C).

In males, spermatocyte and spermatid layers are less thick than in previous stage. Spermatozoa fill the majority of the lumina and often swirl into the center (Fig. 3C).

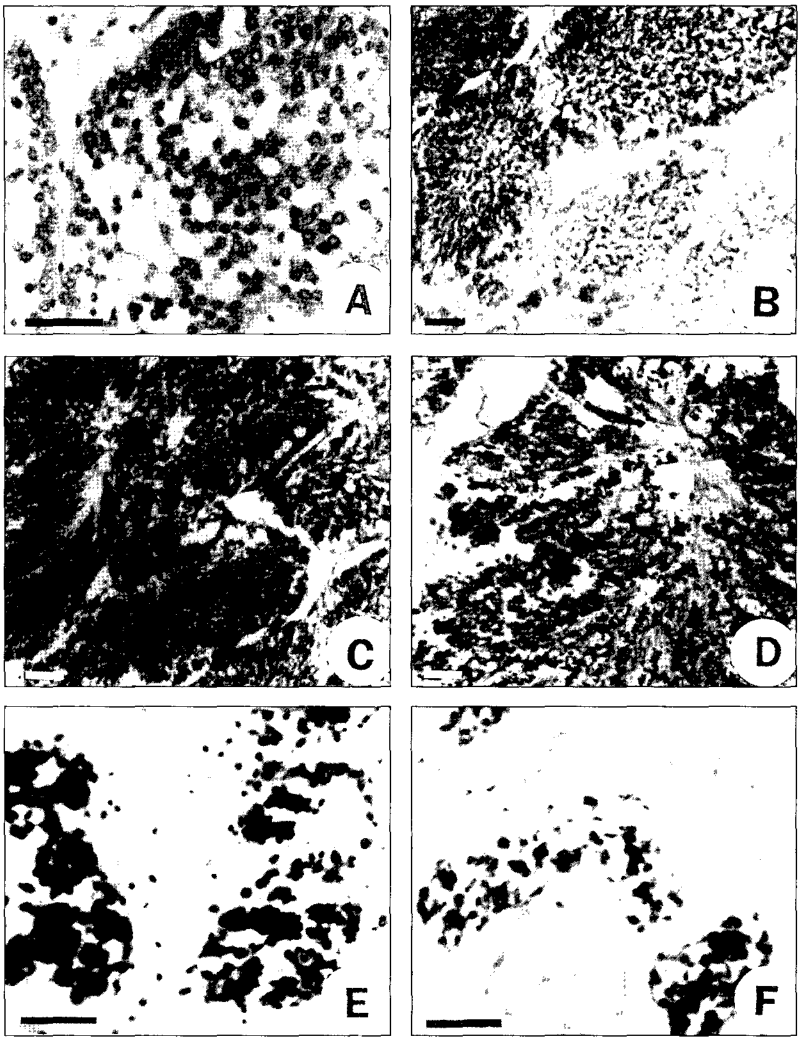

Fig. 3. Photomicrographs of the gonadal phase of male oyster, *Crassostrea gigas*. A, transverse section of the spermatogenic follcle in the early active stage, scale bar=50 μm; B, section of the follicles in the late active stage, scale bar=50 μm; C, section of spermatogenic follicles in the ripe stage, scale bar=50 μm; D, section of the partially spawned stage, scale bar=50 μm; E, section of spermatogenic follicle in the spent stage, scale bar=50 μm; F, section of spermatogenic follicles in the inactive stage, scale bar=50 μm.

Partially spawned stage

In females, number of free oocyte per follicle decrease, empty follicles appear (Fig. 2D).

In males, mature spermatozoa start to release, empty space appear in the center in 30% of the follicles (Fig. 3D).

Spent/Inactive stage

In females, after spawning follicles are shrunk, fused, scattered and disorganized. The gonad is predominantly composed of connective tissue. Thereafter, newly formed oogonia appear along the follicular wall (Figs. 2E, 2F).

In males, only residual spermatozoa can be found in some follicles and connective tissue and phagocytes become incresingly prominent after spawning. Thereafter, newly formed spermatogonia appear along the follicular wall (Figs. 3E, 3F).

First sexual maturity

Percentages of first sexual maturity were histologically investigated with shell heights of specimens participating in reproduction from April to October, 1996. The rate of matured individuals to number of females and males was used to determine first sexual maturity.

Sex ratio

Sex ratio was estimated by with 919 orsters sexually matured individuals from January to December, 1996. It was determined by the photomicroscopic observation. A Chi square, goodness-of-fit was used to test the hypothesis of equal representation of females and males.

Results

Position and external features of the gonads

Most individuals of the Pacific oyster, *C. gigas*, are dioecious, but a few individuals are hermaphrodites. The gonads locate between the digestive diverticula and the outer fibromuscular layers compacted by the fibrous connective tissues and muscle fibers. As the gonads were getting mature, the gonads extended to the lowest part of the muscular layers around the foot. As the gonadal maturation progresses, the external features of the matured ovary and testis became white or milky white in colour. At this time, it was hard to determine their sexes by the external features because of the same colour in both sexes. However, when they were slightly scratched with razor, ripe eggs and milky white sperms were flowed out.

Monthly changes in the gonad follicle index (GFI)

To estimate the spawning period, monthly variations in the GFI were examined from January to December, 1996 (Fig. 4). The GFI values in female and male began to increase between March and April when the mean seawater temperatures gradually increased and reached the maximum (49.97% in female and 55.20% in male) in May. And then, the GFI sharply decreased from June through September when the mean seawater temperatures were relatively high (above 19.65℃) indicating the optimum seawater temperatures for spawning. Therefore, it is assumed that the spawning period is from June to September, when the GFI values in female and male showed a sharp decrease. At this time the optimum seawater temperatures (above 22℃) for spawning were maintained. In the present study, monthly changes in the GFI coincided with gonad of developmental phases.

Monthly changes in relative frequency distributions of the egg diameters

To estimate the spawning period, size frequency disributions of the egg were measured from January to December, 1996 (Fig. 5). Between January and February 1996, relative frequencies of 11~20 μm in egg diameter were over 20%. A number of eggs of 21~30 μm began to appear in March and relative frequency of 31~40 μm (late developing oocyte) in

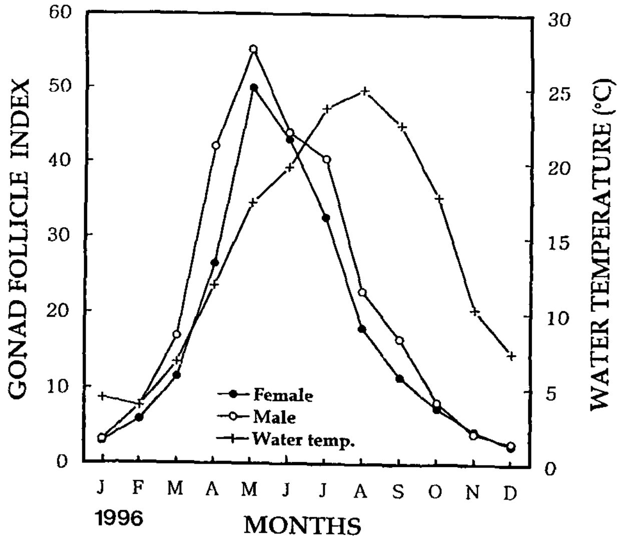

Fig. 4. Monthly changes in gonad follicle index (GFI) of the Pacific oyster, *Crassostrea gigas* and the mean seawater temperature from January to December, 1996.

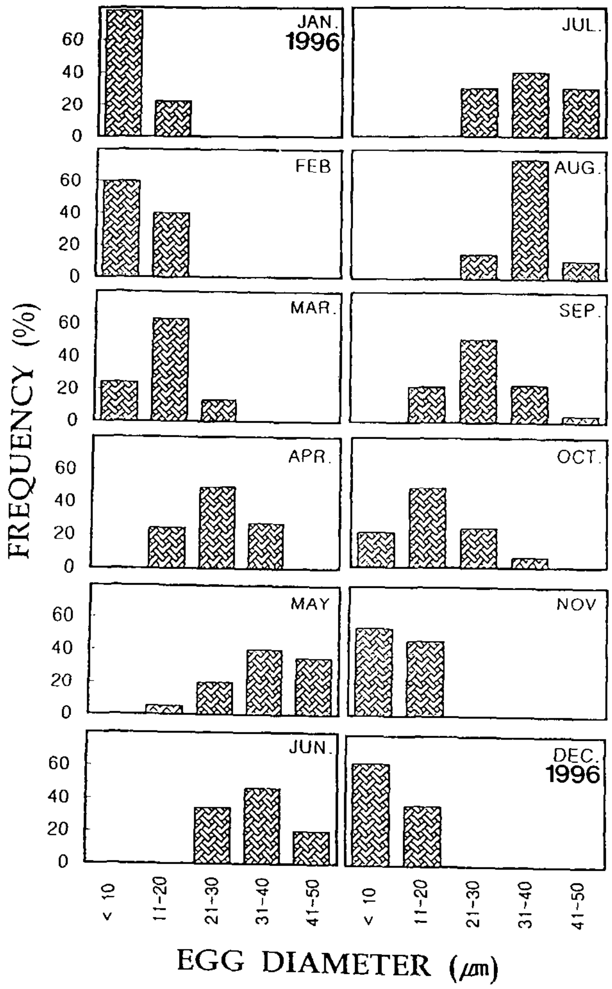

Fig. 5. Relative frequency distribution of the ovarian egg-diameter of *Crassostrea gigas*.

April were about 30%. Percentages of eggs of 41~50 μm (ripe oocyte) in diameter were about 40% in May. Between June and July when spawning began, ripe oocytes of approximately 45~50 μm began to decrease in number because of their discharge. Small number of undischarged large sized oocytes degenerated between August and September. After spawning or gonadal degeneration, newly formed oogonia and oocytes of 11~20 μm in diameter were present during the reproductive cycle, oogenic follicles from September to March.

Reproductive cycle

Frequencies of gonadal phases of the female and male Pacific oyster are shown in Figs. 6, 7. Early active gamete development took place from March (78%) to April (20%) in females and from February (11%) to March (50%) in males. The late active stage were observed between April and May in females and from March to May in males. As a result of oocyte growth and the increase in the number of spermatids, the gonad size gradually increased. And a number of the first ripe female (74%) and male (95%) individuals appeared in May, and then specimens were in the ripe stage from June to September. At this time mature and ripe oocytes (approximately 50 μm) and a number of spermatozoa had full of the follicle. Spawning period appeared from late June to September, and two spawning peaks of both sexes occurred in June (74% in females and 86% in males) and August (71% in females and

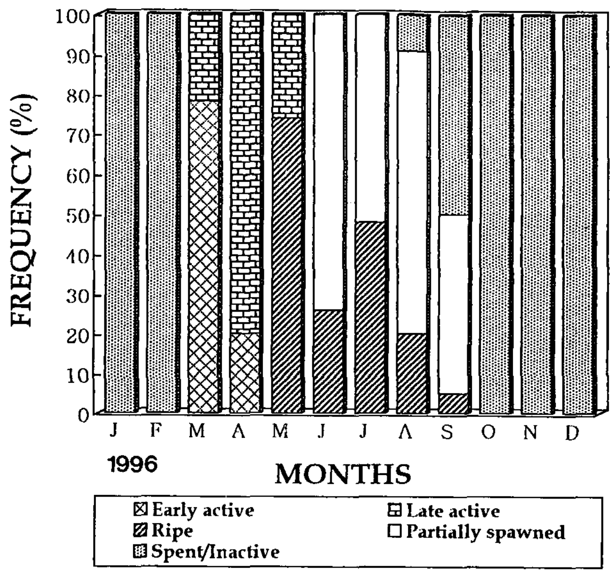

Fig. 6. Frequency of gonadal phases of female oyster, *Crassostrea gigas*, from January to December, 1996.

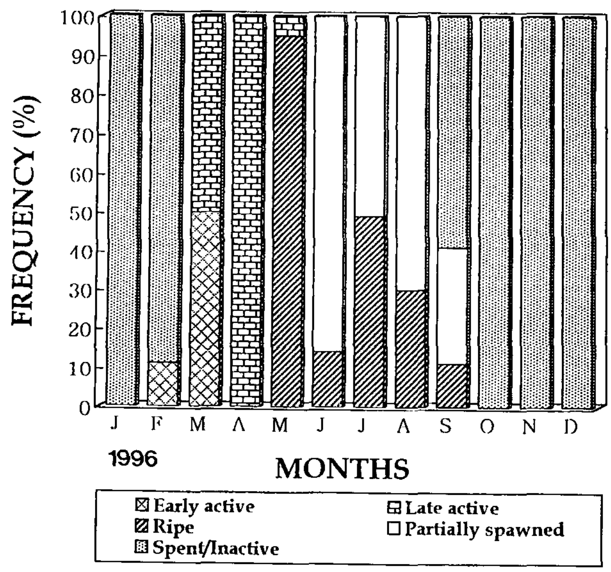

Fig. 7. Frequency of gonadal phases of male oyster, *Crassostrea gigas*, from January to December, 1996.

70% in males). After spawning, the oysters entered the spent stage, and the gametes of both sexes degenerated in most follicles. The spent/inactive stage, in which there was no sign of gonadal activity, appeared from August to March in females and from August to February in males.

First sexual maturity

The first sexual maturity of a total of 474 individuals (242 females and 232 males) of *C. gigas*, of 15.4~61.4 mm in shell height, was investigated histologically in order to find the shell heights that participated in reproduction (Table 1). Percentages of the first sexual maturity of males of 20.1~25.0 mm were over 50%, while females of 25.1~30.0 mm in shell height were over 50%. Those of over 30.1 mm in males, and those over 35.1 mm in shell height in females, were 100%.

Sex ratio

Sex ratio was investigated by histological observation (Table 2). Of 919 oysters investigated, 453 were females and 429 males. The remaining 21 could not be sexed because of having only a few indistinguishable sex cells of various stages, and 16 individuals were hermaphrodite.

There was no significant difference for sex in the numbers of females and males present ($\chi^2=0.653$, $p>0.05$), and monthly comparisons showed no statistical differences in the numbers of female and

Table 1. The shell height of first maturity of the Pacific oyster, *Crassostrea gigas*

Shell height (mm)	Female		Male	
	Number	Mature (%)	Number	Mature (%)
15.4~20.0	22	18.18	26	46.15
20.1~25.0	28	42.85	32	75
25.1~30.0	26	69.23	30	93.33
30.1~35.0	24	83.33	28	100
35.1~40.0	34	100	32	100
40.1~45.0	30	100	28	100
45.1~50.0	32	100	26	100
50.1~55.0	28	100	20	100
55.1~61.4	18	100	12	100
Total	242		232	

male oysters for the twelve-month period.

The sex ratios of individuals were not statistically different from 1 : 1. Therefore, most of the Pacific oysters sampled were dioecious, but it is assumed that possibility of temporary occurrences of hermaphroditism for sexual changes (sex reversal) may exist.

However, when compared the young classes with older ones, the sex ratios were found that 64.00% of the young (1 year class) are more functional for male than female, but 2~3 years old oysters showed a higher percentage of females than that of males (Fig. 8).

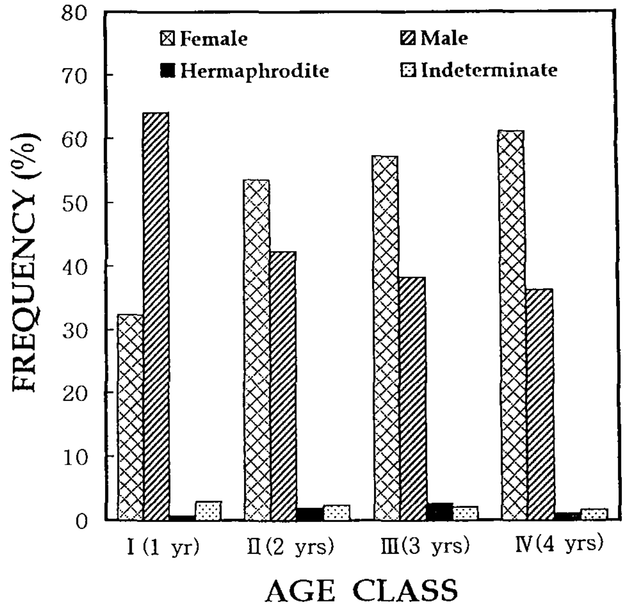

Fig. 8. Frequency of compositions of female, male, hermaphrodite and indeterminate individuals by the age class of shell height of *Crassostrea gigas*, from January to December, 1996.

Hermaphroditism

A total of 919 individuals were used for histological study to investigate the number of hermaphrodites among normal individuals. Of these, 16 hermaphrodites (1.74%) out of 919 individuals were found in the natural oyster population at the intertidal zone on the west coast of Korea (Table 3). The patterns of hermaphrodites can be divided into two types by histological criteria: A, Type I (hermaphrodite having some newly formed developing oocytes on the ovarian tissues within a degenerating spermatogenic follicle after discharge of numerous spermatozoa) and B, Type II (hermaphrodite having two separate follicles in the same gonad); In general, Type I (Fig. 9A) appeared in summer, while Type II (Figs. 9B, 9C) was found in late autumn and winter. Therefore, it is assumed that Type I shows a possibility of sex

Table 2. Monthly variations in sex ratios of the Pacific oyster, *Crassostrea gigas*

Month	Number of samples					Sex ratio [F/(F+M)]	χ^2
	Female	Male	Hemaphrodite	Indeterminate	Total		
Jan. 1996	33	34	1	6	74	0.493	0.015
Feb. 1996	34	35	1	9	79	0.493	0.014
Mar. 1996	36	37	0	0	73	0.493	0.014
Apr. 1996	41	35	0	0	76	0.539	0.474
May. 1996	40	37	2	0	79	0.519	0.117
Jun. 1996	40	36	0	0	76	0.526	0.211
Jul. 1996	41	36	0	0	77	0.532	0.325
Aug. 1996	38	39	0	0	77	0.494	0.013
Sep. 1996	38	33	2	0	73	0.535	0.352
Oct. 1996	39	36	5	0	80	0.520	0.120
Nov. 1996	38	35	3	1	77	0.521	0.123
Dec. 1996	35	36	2	5	78	0.493	0.014
Total	453	429	16	21	919	0.514	0.653

* The critical value for χ^2 goodness of the test of equal numbers of females and males, (1 df) at 95% significance is 3.84.

Table 3. Monthly compositions of sexuality by age class of the oyster, *Crassostrea gigas*

Month		Age class * I (1 yr)				II (2 yrs)				III (3 yrs)				IV (4 yrs)				Total				Total
		F	M	H	I	F	M	H	I	F	M	H	I	F	M	H	I	F	M	H	I	
Jan.	1996	7	13	0	2	9	8	0	1	9	6	1	1	8	7	0	2	33	34	1	6	74
Feb.	1996	8	17	0	3	11	8	0	3	8	6	1	2	7	4	0	1	34	35	1	9	79
Mar.	1996	7	14	0	0	10	9	0	0	10	8	0	0	9	6	0	0	36	37	0	0	73
Apr.	1996	9	13	0	0	12	9	0	0	10	7	0	0	10	6	0	0	41	35	0	0	76
May	1996	8	18	0	0	11	9	2	0	11	5	0	0	10	5	0	0	40	37	2	0	79
Jun.	1996	7	14	0	0	13	11	0	0	11	6	0	0	9	5	0	0	40	36	0	0	76
Jul.	1996	6	13	0	0	12	11	0	0	10	6	0	0	13	6	0	0	41	36	0	0	77
Aug.	1996	7	17	0	0	12	9	0	0	9	6	0	0	10	7	0	0	38	39	0	0	77
Sep.	1996	6	14	0	0	10	6	1	0	10	6	1	0	12	7	0	0	38	33	2	0	73
Oct.	1996	8	15	0	0	12	9	2	0	9	7	2	0	10	5	1	0	39	36	5	0	80
Nov.	1996	9	12	1	1	13	12	1	0	7	6	0	0	9	5	1	0	38	35	3	1	77
Dec.	1996	7	16	1	2	12	9	1	2	8	6	1	2	8	6	0	1	35	36	2	5	78
Total		89	176	2	8	137	110	7	6	112	75	5	4	115	68	2	3	453	429	16	21	919

* Samples divided into 4 age classes: I (1 yr): 15.4~35.0 mm, II (2 yrs): 35.1~45.0 mm, III (3 yrs): 45.1~55.0 mm, IV (4 yrs): <55.1 mm, Total: All the oysters sampled regardless of age class; F: female, M: male, H: hermaphrodite, I: indeterminate.

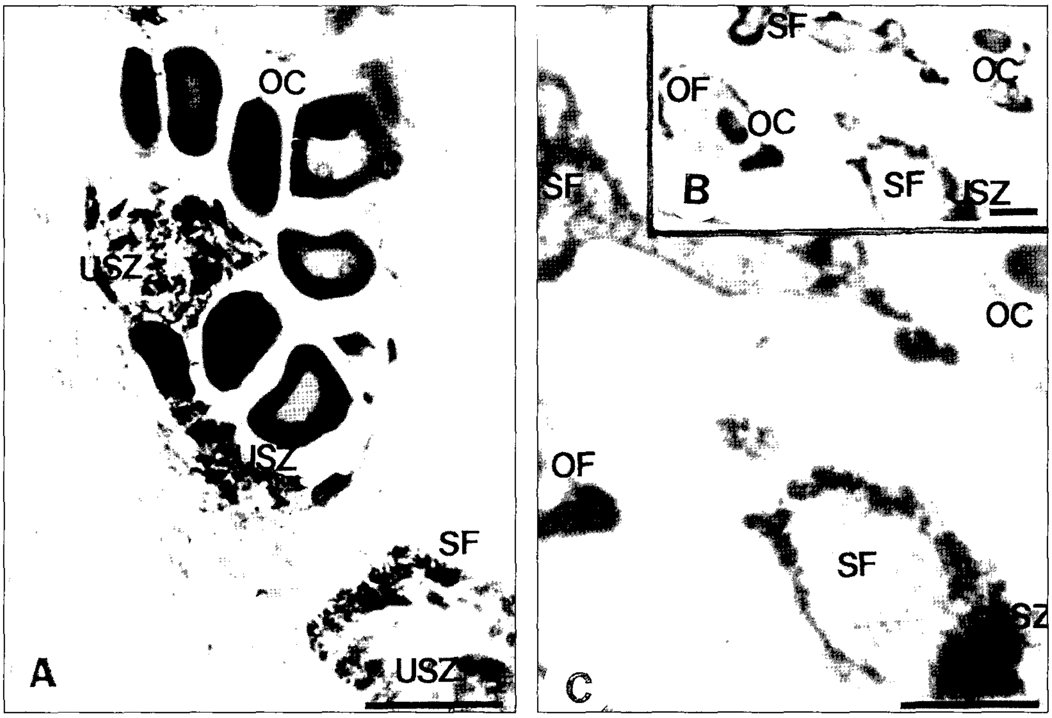

Fig. 9. Hermaphroditic patterns of the gonad of the Pacific oyster, *Crassostrea gigas* A, Type I (hermaphrodite having several newly developing oocytes on the ovarian tissues in degenerating spermatogenic follicle, scale bar=50 μm); B and C; Type II (hermaphrodite having two kinds of separate follicles). C, magnification of Fig. 11B. Abbreviations: OC, oocyte; OF, oogenic follicle; SF, spermatogenic follicle; USZ, undischarged spermatozoa, scale bars=30 μm (Fig. 11B) and 30 μm (Fig. 11C).

change from male to female during summer, when water temperatures are somewhat high and foods are sufficient. Therefore, it was confirmed that this species could not belong to functional and simultaneous hermaphrodite because both germ cells did not show simultaneous developments. This species would rather belong to alternative hermaphrodite because of a possibility of sex changes or sex reversal.

Discussion

With regard to nutritive materials for gamete development, many authors (Lee, 1974; Chang and Lee, 1982; Chung *et al.*, 1986, 1991, 1994) reported that eosinophilic cells and the undifferentiated mesenchymal tissue were abundant in the follicle of the immature stage. Thereafter, these cells and tissues gradually disappeared with the maturation of the gonad, therefore, they could be considered to be a kind of nutritive material. The results of the present study agree with those of others mentioned above.

As shown in Table 4, the seawater temperatures for gonadal maturation of *C. gigas* ranged 18~28℃ (Tanaka, 1954; Bardach et al., 1972; Mann, 1979b; Ventilla, 1984; Dinamani, 1987; Shpigel, 1989). In our study, the seawater temperatures for gonadal maturation of the Pacific oyster ranged 18~25℃.

Therefore, this result is similar to those by several authors. There are some evidences for food level as influencing factors in determining sex in protandric bivalves. Spermatozoa developed at low food level, while oocyte at a higher food level, and oocyte growth may be suppressed if food was insufficient (Sastry 1968).

In the present study, we found hermaphroditism changing their sexualities or protandric phenomena in testicular tissues than development of ovarian tissues in the same follicle after spawning at low food level and lower seawater temperatures. These results coincide with several authors' opinions mentioned above. Several authors (Loosanoff, 1945; Loosanoff and Davis, 1963; Morvan and Ansell, 1988) suggested that gamete production and resorption may be an adaptation phenomenon to environmental factors (e.g., seawater temperature and food availability).

If the energy requirement for the gamete production is too high, nutritive substances may not be enough for all eggs to reach the critical size for spawning. By the processes of degeneration or phagocytosis of the hopeless gametes by the blood cells (Orton, 1927), they may be resorbed to retain energy. Then the energy may be reallocated to developing oocytes or used for other metabolic activities (Dorange and LePennec, 1989; Mortavkine and Varaksine, 1989).

Therefore, the Pacific oyster, may have a mechanism of resorbing and utilizing the high nutritive substances rather than release of undischarged gametes after spawning.

Table 4. Comparison of the optimum seawater temperatures for gonadal maturation of the oyster, *Crassostrea* spp. in different geographic locations

Location	Gonadal maturation	Sources
Ariake Sea (Japan)	20~28℃	Tanaka, 1954
Taiwan, Korea, France, Portugal	19~25℃	Bardach et al., 1972
Woods Hole, U.S.A.	18℃	Mann, 1979b
Hiroshima Bay, Matsushima Bay, Japan	19~25℃	Ventilla, 1984
Northland (New Zealand)	18~25℃	Dinamani, 1987
Gulf of Eilat (Israel)	18~22℃	Shipigel, 1989
Poryong (Korea)	18~25℃	The present study

Griffiths (1977) and Jara-millo and Navarro (1995) suggested that the breeding season of marine bivalves coincided with high concentration of available foods. In general, bivalves spawn when food supply is enough for the planktotrophic larvae and for restoring the energy consumed by adults during spawning (Bayne, 1976). Tanaka (1954) reported that the optimal seawater temperatures for spawning were 18~28℃. In our study, the seawater temperatures ranged from 18℃ to 25℃ during the spawning period (June to September). Therefore, it is assumed that the ranges of seawater temperatures for spawning are 18~28℃.

Tanaka (1954) reported that the spawning season of the Pacific oyster was from early May to early November, and peaks of spawning occurred four times a year in Ariake Sea, Japan. According to Yoo et al. (1971), in Korea spawning season of the Pacific oyster, was from early June to mid-July in natural intertidal population (the intertidal habitat) and from mid-July to early September of the hanging cultured oysters. Yoo et al. (1971) also reported that in spawning season of the Pacific oyster was from early May to early June in Namhae-do, Korea, and the first spawning of the oyster of three years old occurred from late May to early June and the second spawning from late August to early September in Tongyoung in Korea.

The spawning period of the oyster from natural intertidal habitat on the west coast of Korea was from mid-June to late September. The two spawning peaks occurred in June and August when the seawater temperatures were relatively high. Therefore, this result is similar to that of Yoo et al. (1971). Thus, this species belongs to the summer breeder as indicated by Boolootian et al. (1962). The slight differences in the spawning period of the oysters might be related to the geographical differences in the water temperature (Belding, 1930; Chung et al., 1991), time of food production (Chung et al., 1991), and some other environmental factors (Lee et al., 1991). In general, it is supposed that relatively longer spawning seasons are shown in the localities with higher seawater temperatures, while short spawning seasons occur in the regions having lower water temperatures of higher latitude regions. The number of the spawnings in the region of higher latitude are less than in regions of lower latitude. In the present study, the number of spawnings of *C. gigas* are assumed to be at least more than twice because two spawning peaks was appeared in the study period. Therefore, it can be considered that this species is a biannual spawning species.

Percentages of first sexual maturity of males of *C. gigas* with 20.1~25.0 mm in shell height was 75% and that of females with 25.1~30.0 mm in shell height were more than 69%. Males of >30.1 mm and females of >35.1 mm in shell height were 100 %. Accoding to year classes of shell heights using the Petersen's curve, individuals of 20.1~35.0 mm are considered to be one year class, and it is assumed that both sexes of this size begin to reproduce.

According to sex ratio in the American oyster, *C. virginica*, 70% of the 1 year class is functiontionally male than female but 2 year class is nearly an equal proportion of males and females (Galtsoff, 1964). The older classes consist primarily of females as reported in New Zealand rock oyster *C. glomerata* by Dinamani (1974). Amemiya (1929) found 67% males in *C. gigas* of about 1 year class and showed a higher percentage of females in 2 year class. In the sex ratio of *C. gigas* by histological study, these results agree well with the phenomena appeared in the *C. gigas* in Japan (Amemiya, 1929) and other two *Crassostrea* spp. (Galtsoff, 1964; Dinamani, 1974).

Therefore, changes in sex ratios reveal a possibility of sex changes (sex reversal) according to growth or age of the individuals during certain developmental stages. It may be caused by changes of environmental factors (seawater temperature, salinity and food availability, etc.).

Many authors (Ahmed and Sparks, 1967, 1970; Menzel, 1968; Menzel and Menzel, 1965; Wada, 1978) reported that there is no evidence for the existence of sex chromosomes in bivalves. There are many hypotheses on the causes underlying sex change in hermaphrodites (Coe, 1943; Heard, 1975). Coe (1943) stated that certain genes for active or suppressive components, which respond to environmental factors, determine males and females.

Therefore, it is supposed that sexuality of the Pacific oyster, *C. gigas*, may be determined or changed as hermaphroditic state by genetic features, environmental factors (especially, seawater temperature, salinity and food availabity, etc.) and other specific physiological factors.

Coe (1943) and Heard (1975) described that hermaphroditic conditions in pelecypods can be classified into 4 categories mainly based on the sequence of reproductive events: 1) functional hermaphroditism, 2) consecutive sexuality, 3) rhythmical sexuality, 4) alternative sexuality (adults function seasonally as separate sexes). Of hermaphroditic conditions mentioned above, with reference to the alternative hermaphrodite, Galtsoff (1964) stated that "the alternative hermaphrodites tend to have gonads with monoecious acini in which eggs and sperms are produced, and particularly, oviparous oyster, *Crassostrea* species belong to the alternative hermaphrodite."

According to the histologial studies on sexuality, *C. gigas* is dioecious in sex (the sex ratio of female to male=1 : 1). Occassionally, however, 1.74% of total individuals were hermaphrodites, adult gonads function seasonally as separate sexes, and hermaphroditic conditions can be divided into two types: Type I is hermaphrodite showing the sex change of male to female after spermatozoa discharge during the summer or autumn under high seawater temperatures and optimum salinity, Type II is hermaphrodite having two separate follicles, it is considered that this species belongs to an alternative hermaphrodite, and alternative characteristics of sex changes could be easily found in the oysters over 3 years old. Thus, the hermaphroditism of *C. gigas* are similar to that of *C. virginica* studied by Galtsoff (1964).

Coe (1934) found small percentage of hermaphrodites (0.75%) in *C. virginica* on the east coast of the United States. Regarding the rock oyster *C. glomerata*, Dinamani (1974) reported that small percentages (0.70%) were hermaphrodites. With reference to percentages of hermaphrodites in *C. gigas*, Katkansky and Sparks (1966) found a larger proportion (1.01~3.04%) in the Washington area than reported by others. In Japan Amemiya (1929) found about 0.80% of hermaphrodite in *C. gigas*, while Berg (1969) reported 0.26% of hermaphrodite in

California area.

In the present study, percentage of hermaphrodite in *C. gigas* was only 1.74%, and this result is similar to the reports by Katansky and Sparks (1966) and Amemiya (1929).

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