

# Morphology and Taxonomic Values of the Sperm in Male *Chlamys (Swiftopecten) swiftii* (Pteriomorphia: Pectinidae) in Western Korea

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## ABSTRACT

The morphology and taxonomic values of the sperm in male *Chlamys (Swiftopecten) swiftii* were investigated by transmission electron microscope observations. The morphologies and ultrastructures of the sperm nucleus and the acrosome of this species are the vase type and long cone shape, respectively. Spermatozoa are approximately 45-50  $\mu\text{m}$  long including a sperm nucleus (approximately 2.60  $\mu\text{m}$  long), an acrosome (about 0.63  $\mu\text{m}$  long), and a tail flagellum (approximately 44-47  $\mu\text{m}$  in long). The axoneme of the sperm tail shows a 9+2 structure. In this study, the right and left basal rings in the acrosomal vesicle of this species show electron opaque part (region), and also the anterior apex part of the acrosomal vesicle shows electron opaque part (region). These characteristics of the acrosomal vesicle were found in Pectinidae and other several families in subclass Pteriomorphia. The number of mitochondria in the midpiece of the sperm of this species are four, as one of common characteristics appear in most species in Pectinidae in subclass Pteriomorphia. In addition, the satellite fibres are found near the distal centriole of this species, as have been reported in other species of Pectinidae in subclass Pteriomorphia. Accordingly, structural characteristics which are found in the acrosomal vesicle, four mitochondria in the sperm midpiece and the appearance of the satellite fibers near the distal centriole of *C. (S.) swiftii* in Pectinidae (subclass Pteriomorphia), can be employed for phylogenetic and taxonomic analyses as taxonomic key or a significant tool.

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**Key words:** *Chlamys (Swiftopecten) swiftii*, sperm morphology

## INTRODUCTION

In Korea, 26 species in Pectinidae in subclass Pteriomorphia have been reported (Min *et al.*, 2004). In general, scallops in Pectinidae comprise one of the more taxonomically perplexing groups of bivalve molluscs, in particular, at the species level (Healy and

Lester, 1991). Aside from their natural resource significance, the scallops in family Pectinidae comprise one of the more taxonomically important group of bivalve molluscs. Recently, spermatogenesis and mature sperm morphology have been documented in many species of bivalve molluscs using electron microscopy (Eckelbarger *et al.*, 1990; Eckelbarger and Davis, 1996; Gaulejac *et al.*, 1995; Chung and Ryou, 2000; Chung *et al.*, 2007, 2010). It is well-known that the ultrastructure of the spermatozoon in the bivalves might be related to the systematics of bivalves (Popham *et al.*, 1974). For that reason, sperm ultrastructure has long been viewed as a tool in assessing phylogenetic relationships in the metazoa

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through the use of spermiocladistic analysis (Jamiesen, 1987, 1991; Franzén, 1970).

Previously, of *Chlamys* sp in Pectinidae, regarding *C. farreri, farreri* there have been several studies on aspects, of reproduction, including the reproductive cycle (Lioa *et al.*, 1983; Yakovlev and Afeichuk, 1995; Chung, 2008), ultrastructural study of oogenesis (Chung, 2008), comparative spermatozoon morphology and bivalve phylogeny (Popham, 1979; Kim, 2001), growth and spawning (Na *et al.*, 1995; Kang and Zhang, 2000), spermatogenesis and sexual maturation (Chung *et al.*, 2005), reproductive ecology and seed production (Lim *et al.*, 1995; Na *et al.*, 1995; Park, 2002), triploid induction (Yang *et al.*, 1999a), on ecological aspects, including distribution, habitat and classification (Whang and Kim, 1973; Yoo, 1976; Kwon *et al.*, 1993; Min *et al.*, 2004), larval growth (Kuang *et al.*, 1997; Yang *et al.*, 1999b), aquaculture experiment (Lim *et al.*, 1995; Sun *et al.*, 1996, 1997). However, regarding *C. (S.) swiftii*, there have been a few studies on reproduction, including comparative spermatogenesis and comparative ultrastructure of spermatozoa (Kim, 2001) and the reproductive cycle (Kim, 2001), and on aspect of ecology, including distribution, habitat and classification (Yoo, 1976; Kwon *et al.*, 1993; Min *et al.*, 2004).

Although a few studies on reproduction and ecology and classification of *C. (S.) swiftii* have been carried out already, there are still gaps in our knowledge on reproductive biology. Little information is available on ultrastructural characteristics of germ cell development during spermatogenesis and its taxonomic values of mature sperm morphology of this species. In particular, the ultrastructural study on spermatogenesis and mature sperm morphology of this species have some differences between genera in Pectinidae. Of sperm ultrastructures, the acrosome of the sperm shows morphological diversity in the bivalve sperm, and hence it may be the most useful structure in assessing phylogenetic relations (Franzén, 1956). Regarding the acrosomal morphology, Healy (1989) reported that different subclasses of bivalves each have unique acrosomal morphologies. Therefore, the acrosomal morphology of the sperm in *C. (S.)*

*swiftii* should be compared with the species of Pectinidae in subclass Pteriomorpha. In addition, the number of mitochondria in the sperm midpiece tend to be stable within any given family or superfamily (Healy, 1989, 1995). Therefore, the number of mitochondria in sperm midpiece of this species should be investigated and compared with the same Genus species in Pectinidae. Beside ultrastructures of germ cells during spermatogenesis, mature sperm morphology should be studied to clarify ultrastructural characteristic in detail. The aim of the present study is the first to describe some taxonomic values of mature sperm morphology of *C. (S.) swiftii*. Therefore, the purpose of the present study is to clarify mature sperm ultrastructural differences between genera species in Pectinidae by taxonomic analyses of *C. (S.) swiftii*.

## MATERIALS AND METHODS

### 1. Sampling

Specimens of *Chlamys (Swiftopecten) swiftii* (Bernardi, 1858) were collected monthly in the subtidal zone of Oryukdo, Busan, Korea, for one year from January to December, 2007. A total of 132 male individuals were used for transmission electron microscope observations.

### 2. Transmission electron microscope observation

For transmission electron microscope observations, excised pieces of the gonads were cut into small pieces and fixed immediately in 2.5% paraformaldehyde-glutaraldehyde in 0.1 M phosphate buffer solution (pH 7.4) for 2 hours at 4°C. After prefixation, the specimens were washed several times in the buffer solution and then postfixated in a 1% osmium tetroxide solution in 0.2 M phosphate buffer (pH 7.4) for 1 hour at 4°C. Specimens then were dehydrated in increasing concentrations of ethanol, cleared in propylene oxide and embedded in an Epon-Araldite mixture. Ultrathin sections of Epon-embedded specimens were cut with glass knives on a Sorvall MT-2 microtome and LKB ultramicrotome at a thickness of about 80-100 nm. Tissue sections were mounted on collodion-coated

copper grids, doubly stained with uranyl acetate followed by lead citrate, and observed with a JEM 100 CX-II (80-KV) electron microscope.

## RESULTS

### 1. Morphology and the Ultrastructures of the Spermatozoon

After spermiogenesis of the spermatid, it becomes a completed sperm. According to transmission electron microscope observations of the spermatozoa in the acinus in the testis, in general, morphology of the spermatozoon is composed of three parts, as have been seen in other bivalve spermatozoa: 1) the head part, 2) the midpiece part, and 3) the tail part. Morphological and ultrastructural characteristics of three parts of the spermatozoon are as follows.

#### 1) Head part of the spermatozoon

The head part of the spermatozoon is composed of an acrosome and a long nucleus. An acrosome lying on the nucleus is composed of acrosomal vesicles (0.63  $\mu\text{m}$  long) and subacrosomal materials existing in a coarsely granular matrix in the subacrosomal space between the invaginated nucleus and the acrosome (Figs. 1a, b, c). The shape of the acrosomal vesicle is long cone in shape, and the acrosome is composed of high electron dense opaque parts (material) from the base to the tip: the base part of the invaginated nucleus (Fig. 1c), the right and left lateral basal rings (Fig. 1b) and the apex part (Fig. 1a) show high electron dense opaque parts (regions), as have been reported in other species in subclass Pteriomorpha. In particular, in case of this species, the subacrosomal materials are filled from the base part of the invaginated nucleus to the half part of the acrosomal vesicle, as have been reported in *Chlamys* spp. In particular, the cross sectioned the acrosomal vesicle and subacrosomal granular materials are shown in Figs. 1b, c. Particularly, subacrosomal materials showed low electron dense granules in the subacrosomal space between the anterior nuclear fossa (the invaginated part) of the nucleus and the middle part of acrosome. There are some gaps between the nucleus and acrosome.

The type of sperm nucleus of this species is the vase in shape (2.60  $\mu\text{m}$  long and 1.41  $\mu\text{m}$  width). At this stage, anteriorly the nucleus is deeply invaginated (Fig. 1c), and the subacrosomal space is occupied by subacrosomal material. Posteriorly, the invaginated posterior nuclear fossa of the sperm nucleus appear near the centrioles. The nuclear electron density is highly electron dense. Electron lucent lacunae are visible in the sperm nucleus in some sections. Therefore, particularly, the vase type and long cone in shape are morphological characteristics of the invaginated nucleus type and the acrosome shape of this species. .

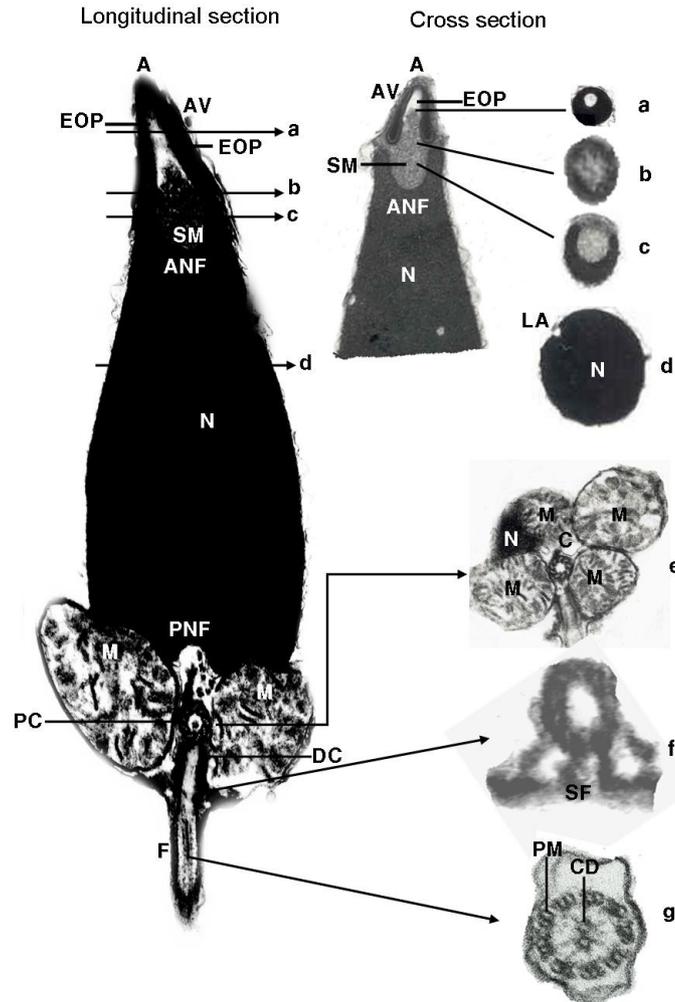
#### 2) Midpiece part of the spermatozoon

Posterior to the nucleus is the sperm midpiece, this region consists of four spherical mitochondria surrounding a pair of triplet substructure centrioles. The cristae of each mitochondrion were randomly arranged, in the cross sectioned distal centriole and sperm midpiece (Fig. 1e). At the sperm midpiece part, the proximal centriole lied at 90° to the sperm longitudinal axis or the distal centriole near the posterior nuclear fossa (basal invagination) of the nucleus. The distal centriole lied parallel to the sperm longitudinal axis and forms the point of origin of flagellar axoneme (Fig. 1e). In particular, in the cross sectioned acrosomal vesicle, the axial rod is not present in the subacrosomal material. However, in particular, the distal centriole of this species appears to possess two lateral satellite fibers because satellite fibers are linked the distal centriole, as have seen in all species of Pectinidae (Fig. 1F).

#### 3) Tail part of the spermatozoon

The sperm tail part of this species is composed of a long flagellum, unlike biflafella have been reported in the natural triploid Corbicullidae species in freshwater bivalves.

A flagellum is composed of a 9+2 substructure axoneme (that is, nine peripheral microtubules surrounding a central doublets) enclosed by the plasma membrane (Fig. 1G) and ensures approximately 43-47  $\mu\text{m}$  long.



**Figs. 1a~1g.** Electron micrographs showing morphologies and ultrastructures of the spermatozoon in male *Chlamys (Swiftopecten) swiftii*.

**Fig. 1a.** Cross sectioned apex part (arrow mark) of the acrosomal vesicle (AV) in the acrosome (A). Note the apex part of the acrosomal vesicle (AV) being composed of an electron high dense opaque part (region) (EOP),

**Fig. 1b.** Cross sectioned right and left lateral basal rings (arrow mark) of the acrosomal vesicle (AV) of the sperm. Note the lateral basal rings of the acrosomal vesicle (AV) showing a high electron dense opaque part (region) (EOP) and subacrosomal materials (SM),

**Fig. 1c.** Cross sectioned base part (arrow mark) of the acrosomal vesicle (AV) and invaginated nucleus (anterior nuclear fossa, ANF) of the sperm. Note the acrosomal vesicle (AV) showing a high electron dense opaque part (region) (EOP) and subacrosomal materials (SM) in the invaginated nucleus ,

**Fig. 1d.** Cross sectioned nucleus (N) (arrow mark) of the sperm. Note the nucleus showing a high electron dense opaque part (region)(EOP) and the lacunae (LA).

**Fig. 1e.** Cross sectioned sperm midpiece region . Note sperm midpiece consisting of four spherical mitochondria (M) surrounding a pair of triplet substructure centrioles (C) and the cross sectioned proximal centriole (PC) and distal centriole (DC) in the sperm midpiece.

**Fig. 1f.** The satellite fibers (SF) in the sperm midpiece. Note. two lateral satellite fibers (SF) near the distal centriole in the sperm midpiece.

**Fig. 1g.** Cross sectioned sperm tail part (arrow mark). Note a flagellum being composed of nine peripheral microtubules (PM) surrounding a central doublets (CD) showing a 9+2 substructure axoneme.

## DISCUSSION

### 1. General morphologies and ultrastructures of the spermatozoa

The morphologies and ultrastructures of the mature sperm after spermiogenesis in *Chlamys (S.) swiftii* are very similar to those of other bivalves that undergo external fertilization (Chung and Ryou, 2000; Kim, 2001; Chung, 2007, 2010; Jun *et al.*, 2009; Kim *et al.*, 2010a,b,c; Kang *et al.*, 2012). However, fine structural differences in molluscan bivalve sperm structures, which were associated with the evolution of the species, are sometimes used as criteria for classification (Popham, 1979). Franzen (1970) divided molluscan sperm morphology into two types: 1) the primitive type found in external fertilization species and 2) the modified type found in internal fertilization species.

However, Verdonk *et al.* (1983) reported that sperm morphology can be divided into four types: 1) primitive, 2) modified, 3) biflagellate, and 4) aflagellate types. In addition to the primitive type and partially modified type of molluscan sperm, a biflagellate type is seen in the triploid *Corbicula fluminea* and *C. leana* in natural populations (Komaru and Konishi, 1996; Komaru *et al.*, 1997). An aflagellate type was also found in a few crustacean (Kim, 2001). *C. (S.) swiftii* undergoes external fertilization and possesses the primitive type of the spermatozoon, unlike the modified type found in most internal fertilization gastropods. The acrosome morphology of the sperm head differs markedly among the species (Popham, 1979).

As shown in Table 1, comparisons of sperm morphologies and structures of four species in Pectinidae in subclass Pteriomorpha are as follows: total lengths of spermatozoa of four species in Pectinidae are almost same (approximately 47–50  $\mu\text{m}$  long), and the nuclear types and acrosomal shapes of four species in Pectinidae are the vase type and cone in shape, respectively, except for the nuclear type (jar in type) and acrosomal shape (cone shape) of *Argopecten irradians*.

However, although total lengths and morphologies

of spermatozoa of four species in Pectinidae were almost same, sizes of the sperm head part (including nucleus length and acrosome length) showed some differences by the species: Of sizes of sperm heads of four species, that of *A. irradians* was a minimum (2.27  $\mu\text{m}$  long) because nuclear length and acrosomal length were the minimum size, however that of *C. (S.) swiftii* showed a maximum (3.81  $\mu\text{m}$  long) because the length of the nucleus was relatively larger than , that of *A. irradians* and acrosomal length was the maximum.

In particular, the axial rod or axial filament, which are existed in the acrosomes or the nucleus, were not found in four species in Pectinidae (subclass Pteriomorpha), unlike the species in other Ostreidae and Mytilidae in subclass Pteriomorpha.

### 2. Taxonomic value of sperm morphology and ultrastructure

In particular, the morphology of spermatozoa appears to be a feature necessary for assessing phylogenetic relationships. Ultrastructures of the spermatozoa in 5 subclasses of the bivalves have some differences in the morphologies and positions of the acrosomes of the sperms (Popham, 1979; Gaulejac *et al.*, 1995). Recently, sperm ultrastructures and acrosomal morphologies of bivalves is considered a valuable tool in assessing taxonomic and phylogenetic problems within the bivalves (Franzen, 1970, 1983; Popham, 1979; Eckelbarger *et al.*, 1990), and it is now widely used in taxonomic analyses by acrosomal morphology and the number of mitochondria in the sperm midpiece (Healy, 1995; Popham, 1979). In general, however, the sizes of sperm nuclei could not be used in taxonomic analyses because morphological characteristics of sperm nuclei were irregular and varied with the species in the same family (Healy, 1995).

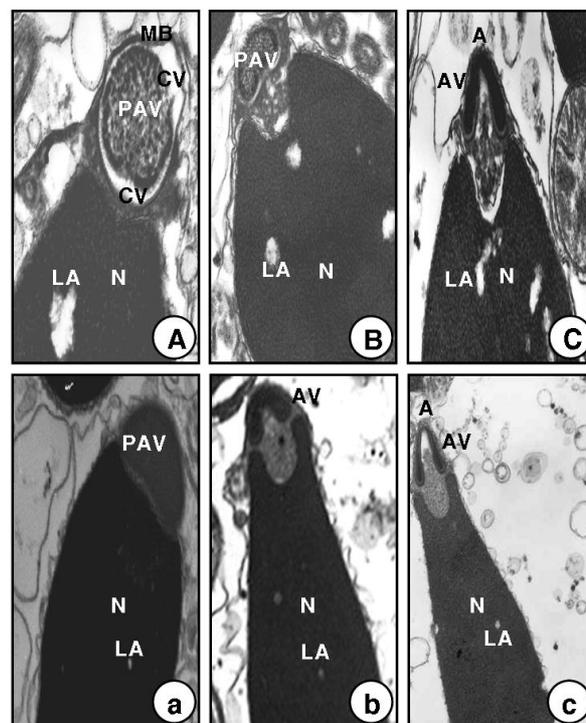
To date, the morphologies of acrosomes in many families in two subclasses Pteriomorpha and Heterodonta have been investigated. We have confirmed that acrosomes can be distinguishable those of genera and families by morphologies and positions of acrosomes. In general, subclass Pteriomorpha in

the bivalves have a common structural characteristics of the acrosomal vesicles showing the cone-like in shape, being composed of electron high dense opaque material (part) from the base to the tip (the apex part and the right and left lateral basal rings) (Hodgson and Bernard, 1986). Taxonomically, Pectinidae belongs to subclass Pteriomorpha. In this study, the ultrastructure and morphology of the acrosomal vesicle of *C. (S.) swiftii* are of long cone shape and it is composed of electron high dense opaque part (material) from the base to the tip, as have been reported by Hodgson and Bernard (1986). Therefore, the same common characteristics mentioned above were confirmed in this species of Pectinidae in subclass Pteriomorpha.

However, Hodgson and Bernard (1986) reported that all species in subclass Heterodonta in the bivalves have a common structural characteristics of the acrosomal vesicles showing the modified cone-like in shape, being composed of high electron dense opaque part (materials), in the base and lateral parts of the basal rings. However, the apex part of the acrosomal vesicle is composed of electron lucent part (materials) at the apex part. Therefore, in case of subclass Heterodonta in the bivalves, structural characteristics of the acrosomal vesicles are composed of high electron dense opaque part (materials) and electron lucent part (materials) at the apex part, as have been reported by some authors (Kim, 2001; Chung *et al.*, 2011). Thus, the acrosomal vesicle of *C. (S.) swiftii* (belongs to the Pectinidae in subclass Pteriomorpha) have some different characteristics, unlike the species of other families in subclass Heterodonta.

Kim (2001) reported that the shapes of sperm nuclei are cylindrical in *Septifer virgatus* and some *Mactra* spp. and *Pernidia venulosa*, global in *Spisula sachalinesis* and *Tersus keenae*, the ovoid shape in the Ostreidae, *Pinctata fucata martensii* and *Atrina pinnata japonica*, the vase shape in *Solen grandis*, and arrow shape in *Corbicula japonica*. Accordingly, the sperm nucleus types vary with the species in families in the subclasses Pteriomorpha and Heterodonta (Chung *et al.*, 2010, 2011).

To distinguish some differences in acrosomal structures by the species, and to clarify the processes of the acrosome formations in the species of Pectinidae, above all, it is needed to investigate the initial processes of the proacrosomal vesicle formation by the species because some differences in proacrosome formations by the species in Pectinidae were found. With reference to the process of the proacrosomal vesicle formation of Pectinidae species, *P. yessoensis* and *A. irradians* have oval proacrosomal vesicles which are filled with relatively coarsed granules surrounded with the outer membrane, and



**Figs. 2A-2c.** Schematic diagrams showing two kinds of the processes of acrosomal vesicle formations in the species of Pectinidae in subclass Pteriomorpha.

**Figs. 2A, B, C:** No.1 process of acrosomal vesicle formation which are found in *Patinopecten yessoensis* and *Argopecten irradians*. Note the inner cavity formed between the granular proacrosomal vesicle and the outer membrane. **Figs. 2a, b, c:** No. 2 process of the acrosome formation which are found in *Chlamys farreri* and *C. (S.) swiftii*. Note lack of the inner cavity between granular proacrosomal vesicle and outer membrane. Abbreviations: A, acrosome; AV, acrosomal vesicle; CV, cavity; LA, lacunae; MB, membrane; N, nucleus; PAV, proacrosomal vesicle.

**Table 1.** Comparisons of sperm morphologies and structures of four species in Pectinidae (Kim, 2001)

Species	Size of sperm head ( $\mu\text{m}$ )	Head part						Middle piece no. of mitochondria	Axial rod or axial filament	Satellite fibers
		Morphology of the sperm nucleus	Nucleus length ( $\mu\text{m}$ )	Nucleus width ( $\mu\text{m}$ )	Acrosome					
					Shape	Length ( $\mu\text{m}$ )	Width ( $\mu\text{m}$ )			
<i>Patinopecten yessoensis</i>	3.48	vase	2.90	1.48	cone	0.60	0.12	4	absent	present
<i>Argopecten irradians</i>	2.27	jar	1.44	1.38	cone	0.33	0.12	5	absent	present
<i>Chlamys farreri farreri</i>	3.68	vase	2.75	1.13	cone	0.50	0.14	4	absent	present
<i>Chlamys swiftii</i> (Present study)	3.81	vase	2.60	1.41	long cone	0.63	0.12	4	absent	present

then a part of the proacrosomal vesicle is uplifted from back to forward.

As shown in Fig. 2, particularly, in the early stage of a large proacrosomal vesicle formation from several proacrosomal granules, there are two different processes as follows: In Figs. 2A, B, C, No. 1 process of acrosome formation are found in *Patinopecten yessoensis* and *Argopecten irradians*. During the formation of the proacrosomal vesicle, the appearance of the inner cavity appears between the proacrosomal vesicle and the outer membrane. However, in Figs. 2a, b, c, No. 2 process of the proacrosomal vesicle formation are commonly found in *Chlamys farreri farreri* and *C. (S.) swiftii*. In this process, particularly, the inner cavity is not formed between proacrosomal vesicle and outer membrane during the proacrosomal vesicle formation. Thus, although the process of proacrosomal vesicle formation in *C. (S.) swiftii* is similar to *C. farreri farreri* (Figs. 2a, b, c), those of *Chlamys* species are very different from those of *P. yessoensis* and *A. irradians* (Figs. 2A, B, C). Therefore, we can conform that the processes of proacrosomal vesicle formation vary with the species in genera.

Kim (2001) reported that the acrosome shape can be classified into four types: cone, cap, elongate modified cone, and modified cap types. Moreover, the sperm nucleus type vary with molluscan species. In the present study, the morphologies of the sperm nucleus type and acrosome shape of *C. farreri farreri*

and other Pectinidae scallops (except for *A. irradians irradians* showing jar shaped) are the vase type and cone type, respectively.

Healy (1995) reported that of sperm ultrastructures of bivalves, the number of mitochondria in the sperm midpiece are now widely used in taxonomic analyses. That is the reason that the number of mitochondria in the sperm midpiece tends to be stable within any given family or superfamily (Healy, 1989, 1995). Recently, some authors (Chung and Ryou, 2000; Kim, 2001; Kim *et al.*, 2010b; Chung *et al.*, 2007, 2010) described that the number of mitochondria at the midpiece of the spermatozoon of bivalve species in many families in subclasses Pteriomorpha and Heterodonta.

Regarding the number of the mitochondria, Kim (2001) described that the number of the mitochondria in the middle piece of the spermatozoon are four in the families Ostreidae, Veneridae, Mactridae, Solenidae and Corbiculidae, while five in the Arcidae, Mytilidae, Pinnidae and Veneridae. The number of the mitochondria in the middle piece of the spermatozoon of *P. yessoensis*, *C. farreri farreri*, and *C. (S.) swiftii* are four. However, *A. irradians irradians* has five mitochondria in the middle piece of the sperm. In particular, the number of the mitochondria in the mid-piece of the sperm of *C. farreri farreri* is four. Accordingly, we can confirm that the number of mitochondria in the sperm midpiece tends to be stable within any given family or superfamily (Healy, 1989,

1995). Therefore, we agree with the opinion suggested by Healy (1989, 1995). However, exceptionally, even though they are the same species and same family, we assume that the number of the mitochondria shows slight differences in number.

In this study, *C. (S.) swiftii* in Pectinidae such as *P. yessoensis*, *C. farreri farreri*, and *A. irradians irradians* has the satellite fibers near the distal centriole in the sperm midpiece. In general, the satellite fibers were found in many species of other families, such as have reported in many species of Ostreidae, Arcidae, Mytilidae in subclass Pteriomorpha. However, to date, the satellite fibers were not found in most species of all families in subclass Heterodonta. Therefore, it is assumed that the appearances of the satellite fibers are a common characteristics of the species in subclass Pteriomorpha.

However, in all species in Pectinidae, the axial rod or axial filament are not found, unlike Ostreidae and Mytilidae species in subclass Pteriomorpha containing the axial rod.

Finally, three kinds of structural characteristics, which are found in the acrosomal vesicle, four mitochondria in the sperm midpiece and the appearance of the satellite fibers near the distal centriole of *C. (S.) swiftii* in Pectinidae (subclass Pteriomorpha), can be employed for phylogenetic and taxonomic analyses as taxonomic key or a significant tool.

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#### REFERENCES

Chung, E.Y. and Ryou, D.K. (2000) Gametogenesis and sexual maturation of the surf clam, *Maetra veneriformis* on the west coast of Korea. *Malacologia*, **42**: 149-163.

Chung, E.Y., Lee, T.Y., Park, K.Y. and Son, P.W. (2005) Ultrastructural study on spermatogenesis and sexual maturation of the male jicon scallop, *Chlamys farreri* on the west coast of Korea, *The Korean Journal of Malacology*, **21**: 95-105.

Chung, E.Y., Kim, E.J., and Park, G.M. (2007) Spermatogenesis and sexual maturation in male *Maetra chinensis* (Bivalvia: Mactridae) of Korea. *Integrative Biosciences*, **11**: 227-234.

Chung, E.Y. (2008) Ultrastructural studies of oogenesis and sexual maturation on female *Chlamys (Azumapecten) farreri farreri* on the western coast of Korea. *Malacologia*, **50**: 282-294.

Chung, E.Y., Chung, C.H., Kim, J.H., Park, S.W., Park, K.H. (2010) Ultrastructures of germ cells and the accessory cells during spermatogenesis in male *Gomphina veneriformis* (Bivalvia: Veneridae) on the east coast of Korea. *The Korean Journal of Malacology*, **26**: 51-62.

Chung, E.Y., Kim, J.H., Kim, S.H., Seo, W.J. (2011) Germ cell development during spermatogenesis and some characteristics of mature sperm morphology in male *Scaphalca subcrenata* (Pteriomorpha: Arcidae) in western Korea. *The Korean Journal of Malacology*, **27**: 121-129.

Eckelbarger, K.J. and Davis, C.V. (1996) Ultrastructure of the gona and gametogenesis in the eastern oyster, *Crassostrea virginica*. II. Testis and spermatogenesis. *Marine Biology*, **127**: 89-96.

Eckelbarger, K.J., Bieler, R. and Mikkelsen, P.M. (1990) Ultrastructure of sperm development and mature sperm morphology in three species of commensal bivalves (Mollusca: Galeommatoidae). *Journal of Morphology*, **205**: 63-75.

Franzén, Å. (1956) On spermatogenesis, morphology of the spermatozoon, and biology of fertilization among invertebrates. *Zoology of Bidr Uppsala*, **31**: 355-482.

Franzén, Å. (1970) *Phylogenetic aspects of the morphology of spermatozoa and spermiogenesis*. In: *Comparative Spermatology*. (ed. by Baccetti, B.) pp. 29-46. Academic Press, New York.

Franzén, Å. (1983) Ultrastructural studies of spermatozoa in three bivalve species with notes on evolution of elongated sperm nucleus in primitive spermatozoa. *Gamete Research*, **7**: 199-214.

Gaulejac, de J., Jenry, M. and Vicente, N. (1995) An ultrastructural study of gametogenesis of the marine bivalve *Pinna nobilis* (Linnaeus, 1758). II. Spermatogenesis. *Journal of Molluscan Study*, **61**: 393-403.

Healy, J.M. (1989) Spermiogenesis and spermatozoa in the relict bivalve genus *Neotrigonia*: relevance to trigonoid relationships, particularly Unionoidea. *Marine Biology*, **103**: 75-85.

Healy, J.M. (1995) Sperm ultrastructure in the marine bivalve families Carditidae and Crassatelloidea and its bearing on unification of the Crassatelloidea with the Carditoidea. *Zoological*

- Science*, **24**: 21-28.
- Healy, J.M. and Lester, R.J.G. (1991) Sperm ultrastructure in the Australian oyster *Saccostrea commercialis* (Iredale and Roughley) (Bivalvia: Ostreidae). *Journal of Molluscan Studies*, **57**: 219-224.
- Hodgson, A.N. and Bernard, R.T.F. (1986) Ultrastructure of the sperm and spermatogenesis of three species of Mytilidae (Mollusca, Bivalvia). *Gamete Research*, **15**: 123-135.
- Jamieson, B.G.M. (1987) *The ultrastructure and phylogeny of insect spermatozoa*. Cambridge University Press, Cambridge
- Jamieson, B.G.M. (1991) *Fish evolution and systematics: evidence from spermatozoa*. Cambridge University Press, Cambridge, pp 181-194.
- Jamieson, B.G.M. (1991) *Fish evolution and systematics: evidence from spermatozoa*. Cambridge University Press, Cambridge. pp.181-194.
- Jun, J.C., Kim, B.S., Chung, E.Y., Kim, J.H., Park, G.M. and Park, S.W. (2009) Spermatogenesis and ultrastructural characteristics of spermatozoa of brackish water diploid clam, *Corbicula japonica* (Bivalvia: Corbiculidae). *Development and Reproduction*, **13**: 115-122.
- Kang, T.G. and Zhang, C.I. (2000) A study on the growth and spawning of Korean scallop (*Chlamys farreri*) around Wando, Korea. *Journal of the Korean Fisheries Society*, **36**: 210-221.
- Kang, H.W., Chung, E.Y., Kim, J.H., Chung, J.S. and Lee, K.Y. (2012) Germ cell differentiation during spermatogenesis and taxonomic values of marine sperm morphology of *Atrina (Servatrina) pectinata* (Bivalvia, Pteriomorpha, Pinnidae). *Development and Reproduction*, **16**: 19-29.
- Kim, J.H. (2001) *Spermatogenesis and comparative ultrastructure of spermatozoa in several species of Korean economic bivalves (13 families, 34 species)*. Ph.D. Thesis, Pukyung National University, 161 pp.
- Kim, J.H., Chung, E.Y., Choi, K.H., Lee, K.Y. and Choi, M.S. (2010a) Ultrastructure of the testis and germ cell development during spermatogenesis in male *Crassostrea gigas* (Bivalvia: Ostreidae) in western Korea. *The Korean Journal of Malacology*, **26**: 235-244.
- Kim, J.H., Chung, E.Y., Choi, K.H., Park, K.H. and Park, S.W. (2010b) Ultrastructure of germ cells during spermatogenesis and some characteristics of sperm morphology in male *Mytilus coruscus* (Bivalvia: Mitilidae) on the west coast of Korea. *The Korean Journal of Malacology*, **26**: 33-43.
- Kim, J.H., Chung, E.Y., Lee, K.Y., Choi, M.S., Seo, W.J. and Kim, S.H. (2010c) Spermatid differentiations during spermatogenesis and mature sperm ultrastructure in male *Crassostrea nipponica* (Seki, 1934, Pteriomorpha: Ostreidae). *The Korean Journal of Malacology*, **26**: 311-316.
- Komaru, A. and Konishi, K. (1996) Ultrastructure of biflagellate spermatozoa in the freshwater clam, *Corbicula leana* (Prime). *Invertebrate Reproduction Division*, **29**: 193-197.
- Komaru, A., Konishi, K., Nakayama, I., Kobayashi, T., Sakai, H. and Kawamaru, K. (1997) Hermaphroditic freshwater clams in the genus *Corbicula* produce non-reductional spermatozoa with somatic DNA content. *Biological Bulletin*, **193**: 320-323.
- Kuang, S., Sun, H., Li, F. and Fang, J. (1997) Feeding and growth of scallop *Chlamys farreri* before and after spawning. *Marine Fisheries Research of China*, **17**: 80-86.
- Kwon, O.K., Park, G.M. and Lee, J.S. (1993) Coloured shells of Korea. 285 pp. Academy Publishing Company, Seoul.
- Lim, H.K., Go, C.S. and Lee, Y.H. (1995) Studies on the technology development for seed production of *Chlamys farreri*. *Technical Report of South Sea Regional Fisheries Research Institute. National Fisheries Research and Development Institute. Busan*. pp. 355-360.
- Liao, C., Xu, Y. and Wang, Y. (1983) Reproductive cycle of the scallop *Chlamys farreri* (Jones and Preston) at Qingdao. *Journal of Fisheries of China*, **7**: 1-13.
- Min, D.K., Lee, J.S., Ko, D.B. and Je, J.G. (2004) Mollusks in Korea. Hanguel Graphics, Busan, Korea. 566pp.
- Na, G.H., Jeong, W.G. and Cho, C.H. (1995) A study on seedling production of Jicon scallop, *Chlamys farreri* 1. Spawning, development and rearing of larvae. *Journal of Aquaculture*, **8**: 307-316.
- Park, K.Y. (2002) Reproductive ecology and seed production of Jicon scallop, *Chlamys farreri*. Ph.D. Thesis, Sooncheonhyang University, 116 pp.
- Popham, J.D. (1974) Comparative morphometrics of the acrosomes of the sperms of externally and internally fertilizing sperms of the sperms of the shipworms (Teredinidae, Bivalvia, Mollusca). *Cell Tissue Research*, **150**: 291-297.
- Popham, J.D. (1979) Comparative spermatozoon morphology and bivalve phylogeny. *Malacological Review*, **12**: 1-20.
- Sun, H., Kuang, S. and Li, F. (1996) Studies on suitable cultures depths and method for scallop in Sanggou Bay. *Journal of Fisheries Science of China*, **3**: 60-65.
- Sun, J., Lin, C., Li, P., Jin, Y. and Zhou, L. (1997) The culture experiment of scallop *Chlamys farreri* in Nanji Islands. *Zhejiang College of Fisheries*, **16**: 247-255.
- Verdonk, N.H., Van Den Biggelaar, J.A.M. and Tompa, A.S. (1983) *The Mollusca*. Vol. 3. Development. pp. 48. Academic Press. New York.
- Whang, H.J. and Kim, M.N. (1973) Study on the distribution and ecology of *Chlamys farreri nipponensis* Kuroda around the Taehuksan Island. *Bulletin of National Fisheries Research and Development Agency*, **11**: 25-35.
- Yakovlev, Y.M. and Afeichuk, L.S. (1995) The reproductive cycle of the scallop *Chlamys farreri* in

- the sea of Japan. Fisheries, Biology and Aquaculture of Pectinids, 8th International Pectinid Workshop, 17: 193-198.
- Yang, A., Wang, Q., Kong, J., Liu, P., Liu, Z., Sun, H., Li, F., Wang, R. and Jiang, M. (1999a) Triploid induction in *Chlamys farreri* by application of 6-dimethyl amino- purine. *Journal of Fisheries of China*, **23**: 241-247.
- Yang, H., Zhang, T., Wang, J., Wang, P., He, Y. and Zhang, F. (1999b) Growth characteristics of *Chlamys farreri* and its relation with environmental factors in the intensive raft-culture areas of Sishiliwan Bay, Yantai, *Journal of Shellfish Research*, **18**: 71-76.
- Yoo, J.S. (1976) *Korean shells in colour*. pp. 48. Ilgisa Co., Seoul.