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Change of Ultrastructure of the Spermathecal Gland in the Silkmoth, *Bombyx mori*

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Synopsis

Change of ultrastructure of the spermathecal gland in *Bombyx* was observed from the phalate adult stage to termination of the oviposition. The gland consists of the glandular cells and the epithelial cells, and the latter divide into the protective cells producing cuticular intima and the ductule cells. In glandular cells the organelles involved in synthetic processes develop remarkably and a central cavity lined with microvilli exists for accumulation of the secretion.

The secretory activity of the glandular cells begins at the phalate adult stage and reaches the maximum level just before emergence. The secretion discharges into the the lumen of the gland during copulation, but the activity resumes during oviposition. Change of the ultrastructure was described during these processes.

Key words: Spermathecal gland, secretion, ultrastructure, *Bombyx mori*.

Introduction

As to the female reproductive system in *Bombyx*, IKEDA (1913) described the gross morphology and the post embryonic development in detail, and thereafter ŌMURA (1938) observed the structure and function concerning to the mechanism of fertilization and found at first that the spermathecal duct contained two canals and one of them played a role for outgoing sperms from the spermatheca. Recently KATSUNO (1977) clarified in his serial studies behavior of sperms in the internal reproductive organs of female moth.

The present author has carried out ultrastructure of the male and female reproductive system from the viewpoint of ontogeny and as a part of the results obtained ultrastructure of the spermatheca was observed and discussed relating to the problems of fertilization. (MIYA, 1982). This paper deals with ultrastructural change of the spermathecal gland before and after emergence and oviposition.

MATERIAL AND METHOD

A bivoltine race, "Daizo" was used as material. The spermathecal glands were removed from the females at the phalat adult stage (10 days after pupation), just after emergence, 3 hr after copulation, just before oviposition, and after termination of oviposition. The materials were fixed with 2.5% glutaraldehyde in 0.1 M cacodyrate buffer (pH 7.4) at 4°C for 1.5 hr, rinsed with the same buffer and post-fixed with 1% osmium tetroxide in 0.1 M

cacodyrate buffer (pH 7.4) at 4°C for 2 hr. After dehydration with ethanol series, the fixed materials were embedded in Fpon 812 through QY-1. The ultrathin sections were stained with uranyl acetate and lead nitrate.

RESULTS

The spermathecal system of *Bombyx* consists of a spiral spermathecal duct, a two lobed spermatheca and a tubular spermathecal gland. The spermathecal duct starts from the tip of the vestibulum and connects with the lower part of the spermatheca. The spermatheca is composed of a small lobe and a large lobe, and to the latter the spermatheal gland belongs (Fig. 1). The spermathecal gland is not a simple tube, but branches one or more short tubes to indicate polymorphism in different individuals (ŌMURA, 1938).

The ultrastructure of the spermathecal system, especially the spermathecal gland, changes with aging of the female moth. The gland consists of the glandular cells and the epithelial cells, and the latter divide into the protective cells producing cuticular intima and the ductule cells. Fig. 2 represents diagrammatically a cross section of the gland and ultrastructure of a set of the above-mentioned cells in the phalate adult.

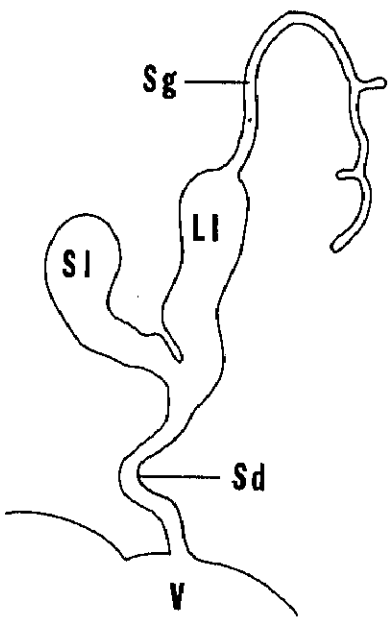


Fig. 1. A drawing of the spermathecal system of a mated female just before oviposition (From Miya, 1982).
Ll : large lobe, Sd : spermathecal duct, Sg : spermathecal gland, Sl : small lobe, V : vestibulum.

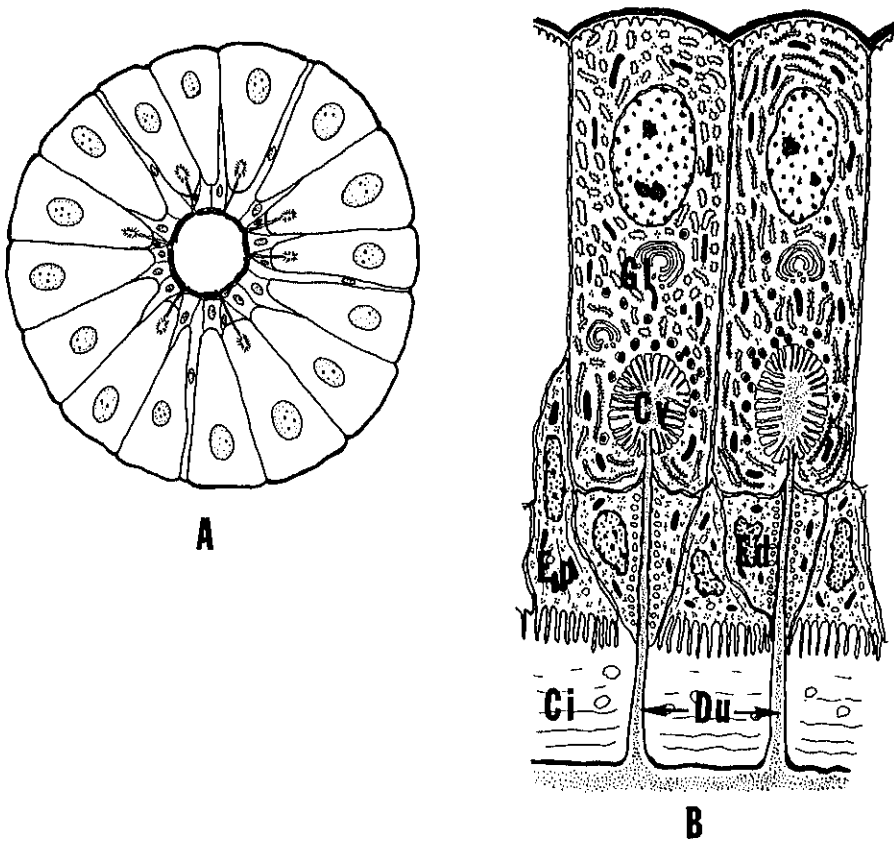
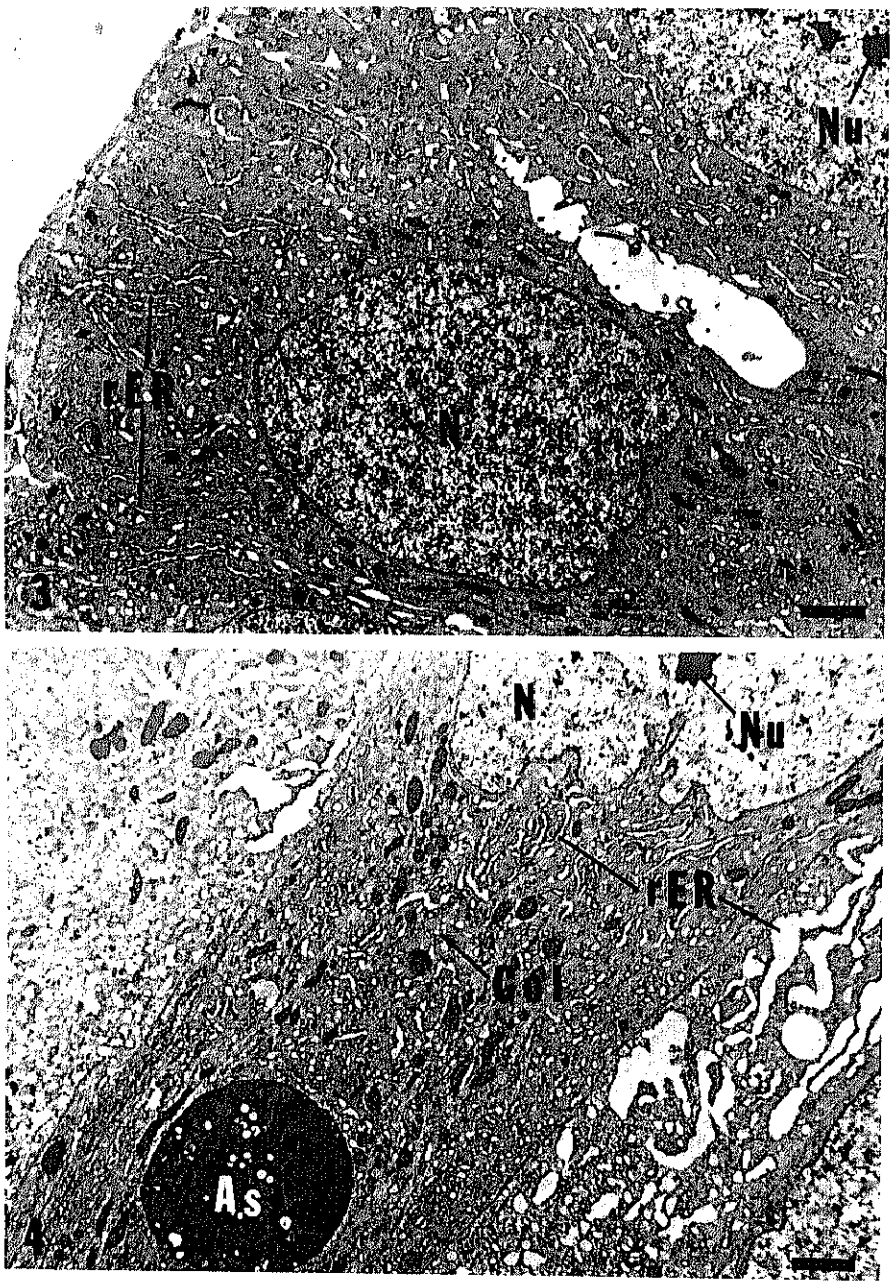
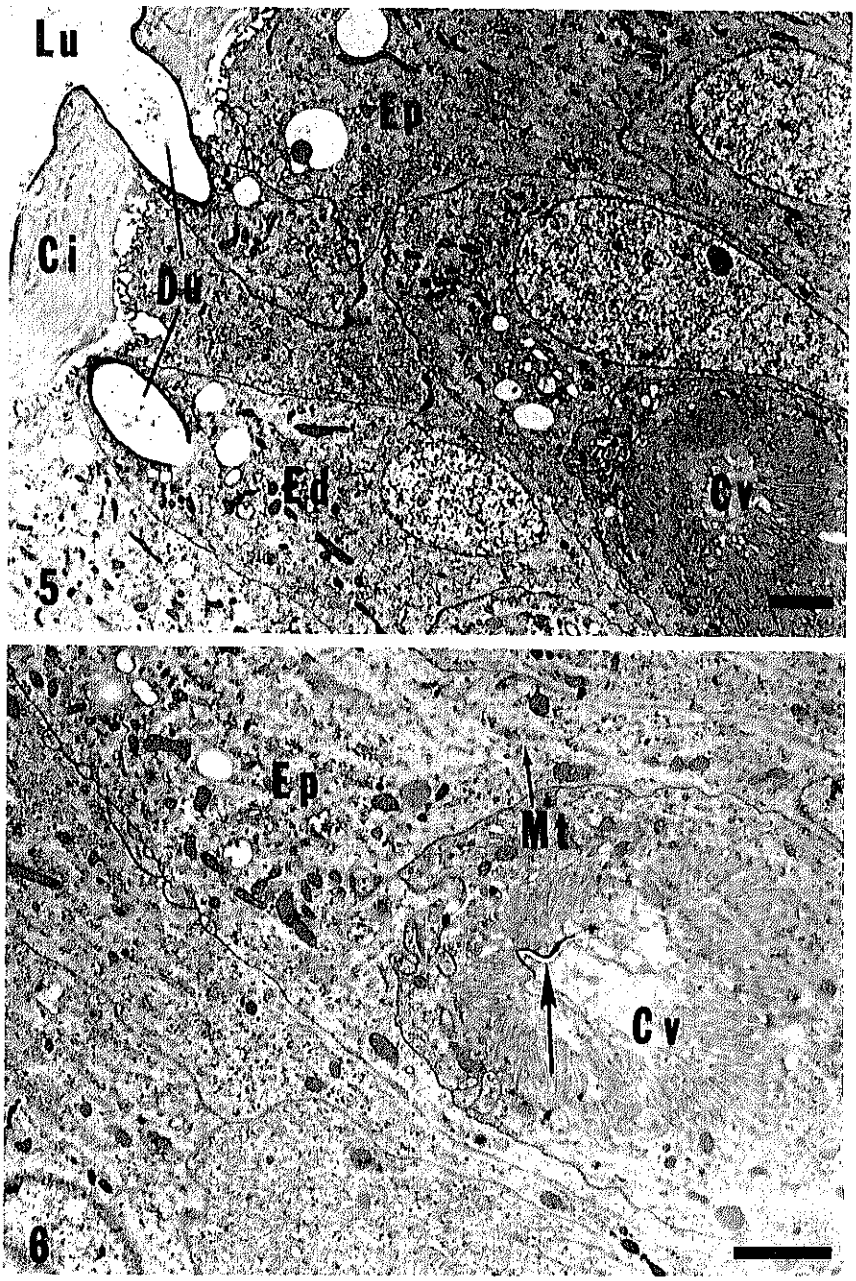


Fig. 2. Diagrammatic figures representing a cross section (A), and ultrastructure of the glandular and epithelical cells (B).
Ci: cuticular intima, Cv: central cavity of the glandular, cell, Du: ductule, Ed: ductule cell, Ep: protective cell, Gl: glandular cell.

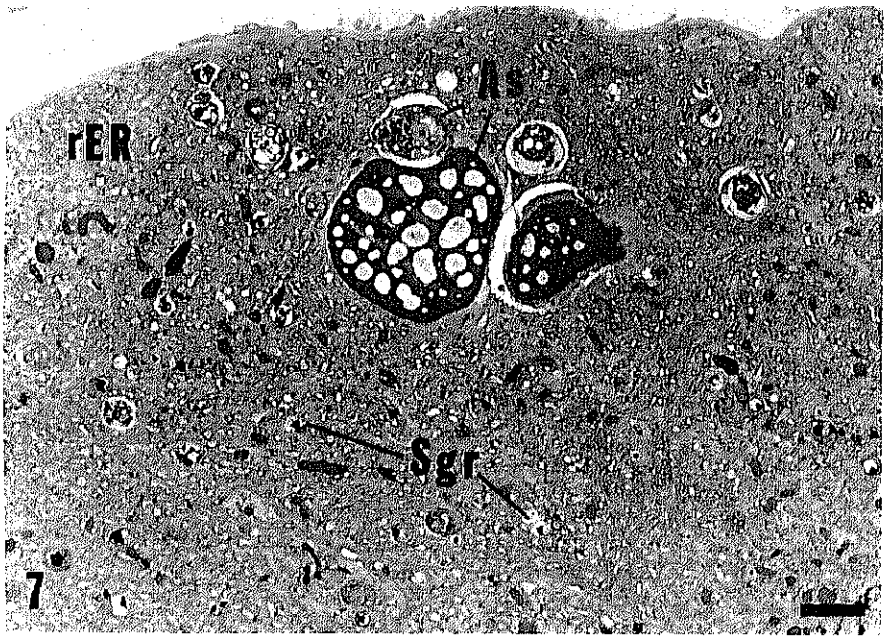
The glandular cells lie in the periphery of the gland and the outside is covered with a thin basement membrane. There is no muscular covering. If the cell is roughly divided into basal and apical halves, the basal half is devoted to organelles involved in synthetic processes. Just under the basement membrane, plasma membrane forms shallow infoldings, and below the plasma membrane well-developed rough-surfaced endoplasmic reticulum (rER) are fouud, interspersed with Golgi bodies, mitochondria, and nucleus. In the nucleus chromatin distributes homogeneously as fine grains and several nucleoli are observed. Sometimes a large autostme-like body is contained in the central region (Figs. 3 and 4). The apical half is dominated by a large central cavity lined with numerous microvilli. At this stage the cavity is relatively small yet (Figs. 5 and 6).

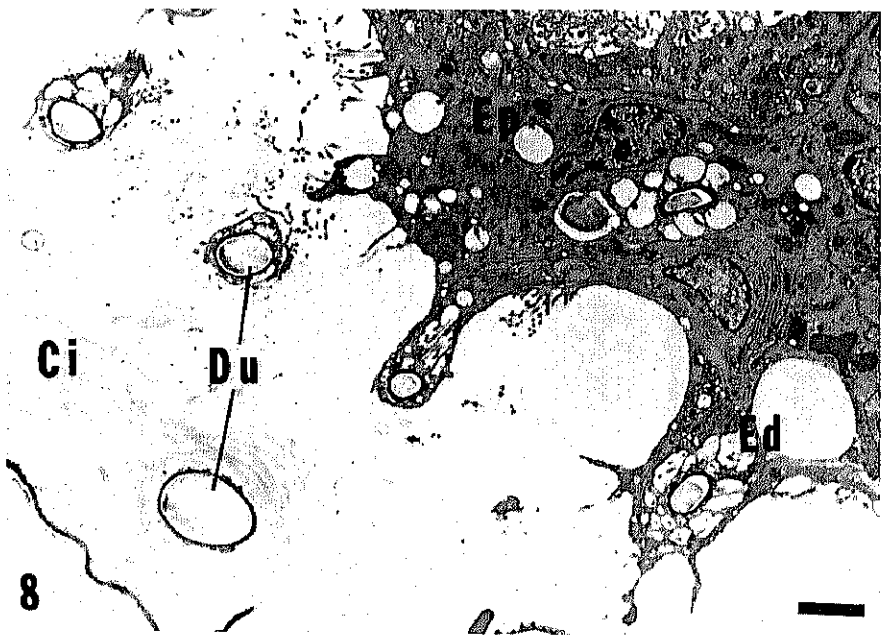


Figs. 3 and 4. Electron micrographs representing the glandular cells at the phalate adult stage. Fig. 3: basal region. Fig. 4: middle region.
As: autosome-like body, Gol: Golgi body, N: nucleus, Nu: nucleolus, rER: rough-surfaced endoplasmic reticulum. Scales: 2 μm.



Figs. 5 and 6. Electron micrographs representing apical region of the cells at the phalate adult stage.
Ci: cuticular intima, Cv: central cavity of the glandular cell, Du: ductule, Ed: ductule cell, Ep: protective cell, Lu: central lumen of the gland, Mt: microtubule, Large arrow: the basal end of the ductule penetrating into the central cavity of the glandular cell. Scales: 2 μm.





Figs. 7 and 8. Electron micrographs representing the glandular cell just after emergence. Fig. 7: basal region, Fig. 8: apical region.
As: autosome-like body, Ci: culticular intima, Du: ductule, Ed: ductule cell, Ep: protevtive cell, rER: rough-surfaced endoplamic reticulum, Sg8: secretion gmanule. Scales: 2 μm.

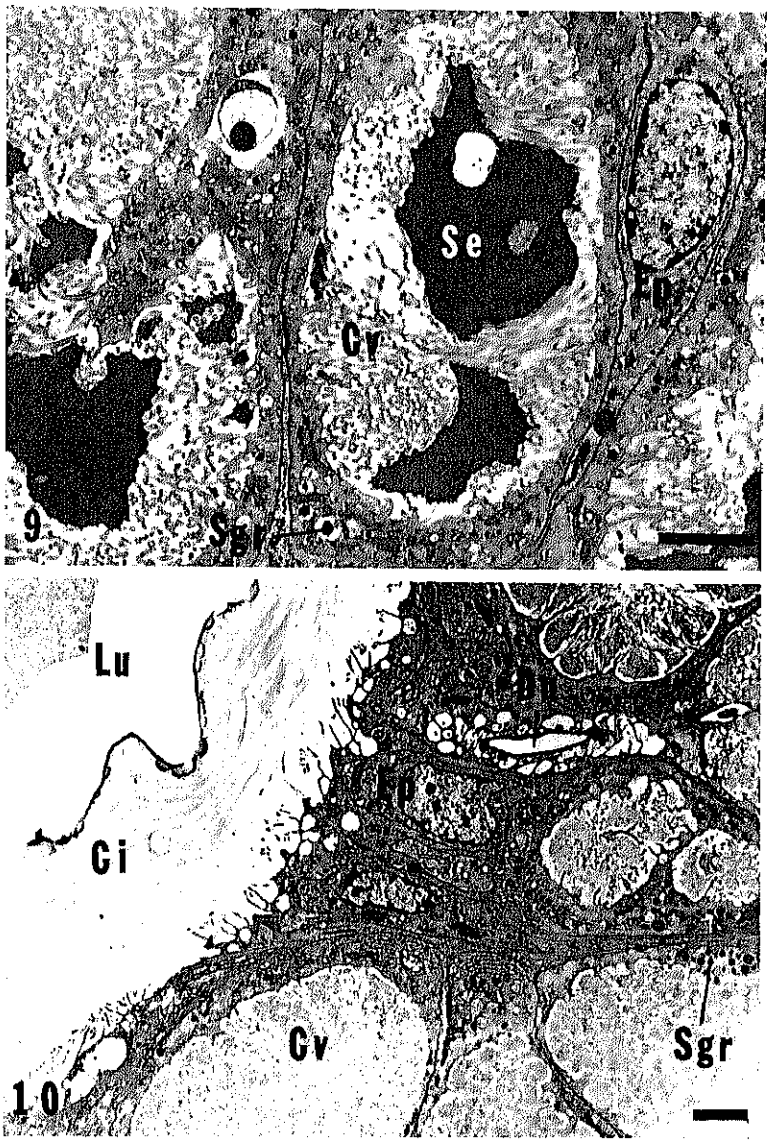


Fig. 9. Electron micrograph representing the central cavity of the glandular cell just after emergence. The cavity is filled with dense masses of the secretion. Cv: the central cavity, Ep: protective cell, Se: dense mass of the secretion, Sgr: secretion granule. Scale: 2 μm.

Fig. 10. Electron micrograph representing apical region of the cells three hours after copulation. The contral cavity is filled with fine secretion grains. Ci: cuticular intima, Cv: central cavity, Du: ductule, Ep: protective cell, Lu: central lumen of the gland, Sgr: secretion glanule. Scale: 2 μm.

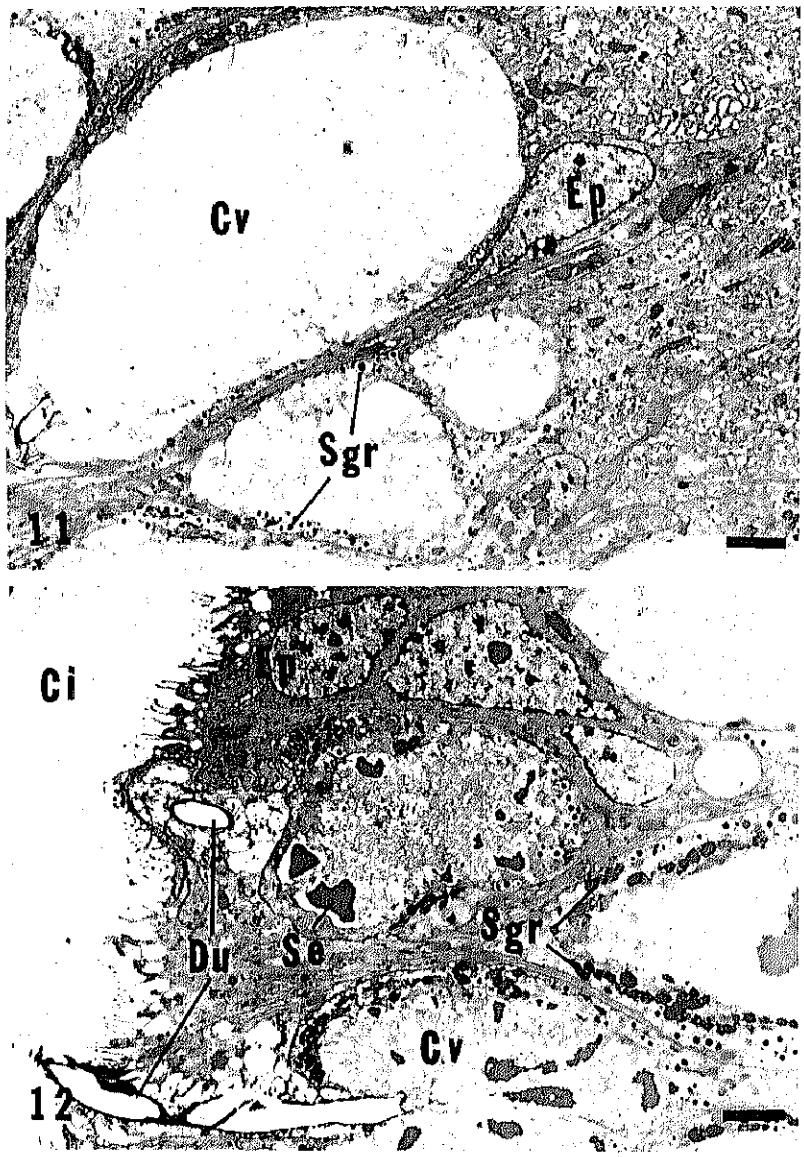


Fig. 11. Electron micrograph representing the central cavity of the glandular cell just before oviposition. The central cavity becomes larger and almost empty because of flowing out of the secretion.
Cv: central cavity, Ep: protective cell, Sgr: secretion granule. Scale: 2 μm.

Fig. 12. Electron micrograph representing apical region of the cells after termination of oviposition. The dense masses and the larger gmanules of the secretion appear again.
Ci: cuticular intima, Cv: central cavity, Du ductule, Ep: protective cell, Se: dense mass of the secretion, Sgr: secretion granule. Scale: 2 μm.

The protective cells line the central lumen of the gland and some of them extend outward among the glandular cells. In the cytoplasm there are few rER, but many microtubules run parallel to the longitudinal axis of the cell. At the apical end of the cell many short microvilli develop and are producing the cuticular intima, which consists of the outer epicuticle and the inner procuticle. The ductule cells attach to the glandular cells and the ductule produced through the cell penetrates into the glandular cell to open in the central cavity and the opposite end opens in the central lumen of the gland. The ductule consists of epicuticle, which connects with that of the cuticular intima (Figs. 5 and 6).

Just before and after emergence, varions changes appear in the spermathecal gland. In the glandular cell the nucleus becomes irregular in shape and fine chromatin grains assemble to form many small masses distributing evenly. The glandular cell indicates active secretory activity and many autosome-like bodies and numerous vesicles containing dense secretion granule occur in the basal half (Fig. 7). The central cevity increases its volume remarkably and several large dense masses of the secretion are accumulated in it. The secretion discharges through the ductule into the central lumen of the gland (Figs. 8 and 9).

According to KATSUNO (1977), the first ejaculation begins 10 minutes after copulation and continues for subsequent 20 minutes. The sperms ejaculated into the copulatory pouch migrate via seminal duct and the spermathecal duct into the spermatheca. Therefore, the spermatheca of the female moth mated for 3 hours is filled with abundant sperms and some of them go up even into the spermathecal gland. The volume of the central cavity of the glandular cell at this stage increases more and more, filled with large amount of fine granules of the secretion, and numerous secretion granules enclose the central cavity (Fig. 10). The secretion flows out continuously into the central lumen of the gland. The size of the epitelial cells decreases remarkably and the cells are pressed to the side of the central lumen of the gland because of hypertrophy of the glandular cells by enlargement of the central cavity.

In the spermathecal gland of the female moth mated for 6 hours, the central cavity of the glandular cell becomes empty because of flowing out of the secretion into the central lumen of the gland (Fig. 11). At the same time, the secretory activity resumes again and many secretory granules are produced in the synthetic organelles. These secretory granules are deposited in the central cavity to form larger dense masses in it during oviposition (Fig. 12). The signiffcane and role of the secondary secretion are unclear.

DISCUSSION

The spermathecal system in *Bombyx* originated from the imaginal disks lying in the 8th abdominal segment (IKEDA, 1913). The spermathecal gland is not a simple tube as described in several lepidopterans, for example, *Pseudaletia unipuncta*, *Peridroma margaritosa* (CALLAHAN and CHOPIN, 1960), and *Chorisoneura fumiferana* (OUTRAM, 1971), but branched into one or more short tubes, resulting in occurrence of polymorphism in

different individuals (ÔMURA, 1938).

The spermathecal gland is ectodermal origin and consists of the glandular cells and the epithelial cells, and the epithelial cells contribute secretion of the cuticular intima and formation of the ductules, as in general epidermal gland (s. NOIROT and OUENNEDET, 1974). Ultrastructure of the glandular cells is similar as in many insect species and these cells have the organelles necessary for synthesis of the secretion and a central cavity lined with numerous microvilli for collecting the secretion. A ductule connects with the central cavity and the secretion flows down through the ductule into the central lumen of the gland. In *Bombyx* also the glandular cells indicate such typical structures, but the collecting apparatus reported in *Anthonomus* (GRODNER, 1979) and *Speonomus* (JUBERTHIE-JUPEAR and CAZALS, 1985) does not develop, but the end of the ductule opens directly into the central cavity.

The epithelial cells in *Bombyx* are divided into 2 types functionally, the one is the protective cells for supporting the glandular cells, which secrete the cuticular intima, and the other the ductule cells for forming the ductule, which extends into the central cavity of the glandular cells and through which the secretion flows down into the lumen of the gland. In the protective cells the microtubules develop and run parallel to the longitudinal axis of the cell the same as in *Periplaneta americana* (GUPTA and SMITH, 1969), *Tenebrio moritor* (HAPP and HAPP, 1970), and so on.

The secretion of the spermathecal gland seems to function as an exogenous nutrient substrate for the stored sperms (OUTRAM, 1971; GRODSER, 1979). Furthermore, the secretion is considered to function as an attractant for swimming up of the sperms in the spermathecal duct, that is, the spermathecal filing of the sperms (GRODNER and STEFFENS, 1978; GRODNER, 1979.).

In *Bombyx* the secretory activity of the spermathecal gland begins at the phalate adult stage and reaches the maximum level just before or after emergence reported already by ÕMURA (1938), and the secretion flows into the spermatheca and seems to play a role as supplier of nutrient for the stored sperms the same as in other insects, but the role as an attractant for sperms is unclear. After the first secretion has flowed out for the following several hours, the secretory activity resumes again during copulation and oviposition, resulting in re-accumulation of large amount of the secretion in the central cavity of the glandular cells after termination of oviposition. The difference between the secretions of the younger and the older female is reported in *Anto nomus* (GRODNER, 1979) and suggests that the secretion must perhaps mature in order to play a role in spermathecal filling. In *Bombyx*, however, the signiffcance of the second secretory act ivity and the nature and role of the secretion are unknown at present.

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