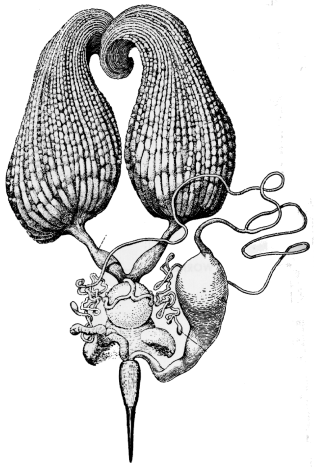

ENTOMOLOGY 322 LAB 20
Female Reproductive System

The female reproductive system consists of paired ovaries (Ov) each with multiple ovarioles (Ovl) Egg maturation occurs gradually as each oocyte passes downward along the ovariole. Oocytes are first formed via meiosis in the germarium (Grm), and progress downward through the vitellarium (Vtl), where the yolk proteins are added to the developing oocyte. The most mature oocytes are located at the bottom of each ovariole, where they are retained until laying. As you might expect, the number of ovarioles is closely related to the rate of egg production. The typical pattern is 4, 6, or 8 ovarioles/ovary, but may be as many as 100-200 ovarioles/ovary occurs in Diptera and eusocial Hymenoptera, and termite queens many have up to 2400 ovarioles/ovary.

Each ovary is connected to a lateral oviduct (Odl), and the paired lateral oviducts meet medially in the common oviduct (Odc). Female insects can store sperm for long periods of time (years) in the spermatheca (Spt), an outgrowth of the common oviduct that is commonly associated with a spermathecal gland (SptGl). Accessory glands (AcGl) may also be present. These glands perform a variety of functions, including producing secretions which hold eggs together in a cluster or glue eggs to substrate, producing the egg case or ootheca in some Orthoptera, providing larval nutrition (as "milk glands") in viviparous Diptera (e.g., *Glossina*), and serving as venom glands in aculeate Hymenoptera.

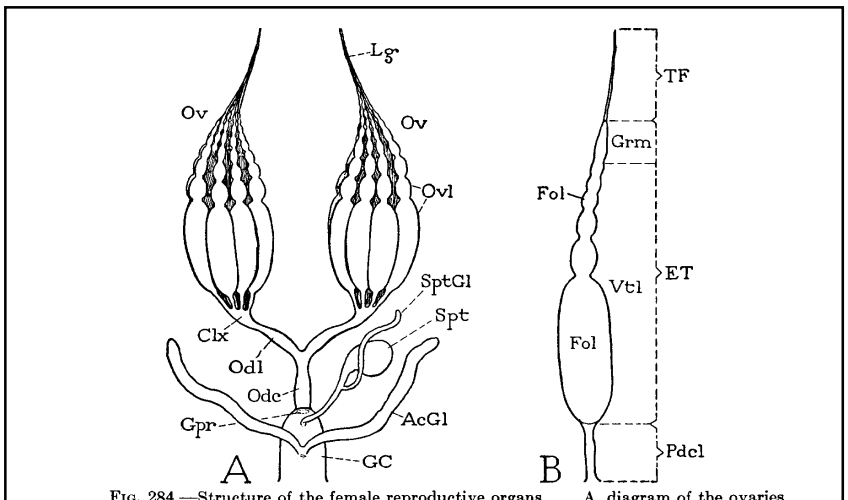

FIG. 284.—Structure of the female reproductive organs. A, diagram of the ovaries, exit ducts, and associated structures. B, diagram of an ovariole. *AcGl*, accessory gland; *Clx*, calyx; *ET*, egg tube; *Fol*, follicle, or egg chamber; *GC*, genital chamber (vagina); *Gpr*, gonopore; *Grm*, germarium; *Lg*, ovarial ligament; *Odc*, oviductus communis; *Odl*, oviductus lateralis; *Ov*, ovary; *Ovl*, ovariole; *Pdcl*, ovariole pedicel; *Spt*, spermatheca; *SptGl*, spermathecal gland; *TF*, terminal filament; *Vtl*, vitellarium.

Figure 20.1 (Snodgrass, 1935)

The location of the female gonopore varies among the insects. Primitively, the gonopore is located behind the 7th sternum. In Ephemeroptera and Dermaptera the lateral oviducts remain separate and open via paired gonopores, while in Apterygotes and some orthopteroids, the lateral oviducts are united in a short median oviduct (Fig. 20.2A). In the majority of insects, the gonopore is shifted caudad, and is located at the posterior magin of the 8th sternum (Fig. 20.2B). In such groups, an additional structure may be formed, the genital chamber (GC), or bursa copulatrix (Fig. 20.2C). In Cicadidae, Panorpida, Trichoptera, Lepidoptera, and Coleoptera, the genital opening is shifted further caudad, and has its opening behind the 9th sternum (Fig. 20.2F). In Lepidoptera (Fig. 20.2E), the reproductive tract has two openings: the genital opening (bursa copulatrix, vulva [Vul]) remains on the 8th segment and opening on the 9th segment serves for the discharge of eggs and is termed the oviporus [Opr in Fig. 20.2E]).

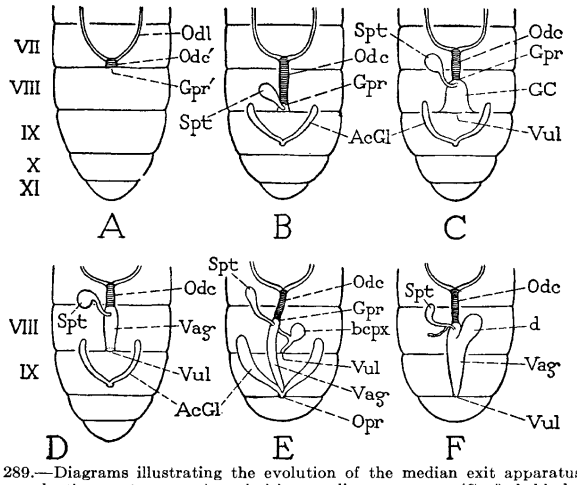

FIG. 289.—Diagrams illustrating the evolution of the median exit apparatus of the female reproductive system. A, primitive median gonopore (*Gpr'*) behind seventh abdominal segment. B, oviduct extended through eighth segment, definitive gonopore (*Gpr*) at end of this segment. C, genital chamber (*GC*) invaginated behind eighth segment. D, genital chamber converted into a vagina (*Vag*). E, vagina extended through ninth segment, but copulatory opening (*Vul*) retained on eighth. F, vagina extended through ninth segment, anterior opening lost, posterior opening becomes the vulva (*Vul*). *AcGl*, accessory gland; *bcpx*, bursa copulatrix; *d*, vaginal pouch; *GC*, genital chamber; *Gpr*, definitive gonopore; *Gpr'*, primitive median gonopore; *Odc*, oviductus communis; *Odc'*, primitive median oviduct; *Odl*, oviductus lateralis; *Opr*, oviporus; *Spt*, spermatheca; *Vag*, vagina (genital chamber); *Vul*, copulatory opening, or vulva.

Figure 20.2 (Snodgrass, 1935)

Fig. 20.3 shows some of the diversity in female reproductive tract morphology.

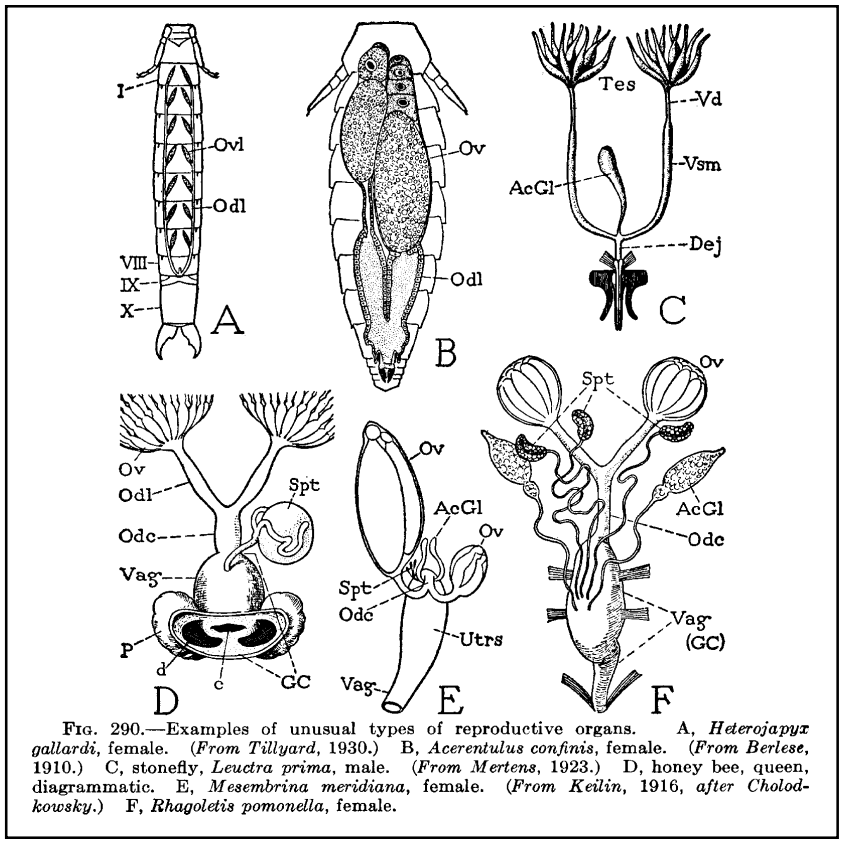

FIG. 290.—Examples of unusual types of reproductive organs. A, *Heterojapyx gallardi*, female. (*From Tillyard*, 1930.) B, *Acerentulus confinis*, female. (*From Berlese*, 1910.) C, stonefly, *Leuctra prima*, male. (*From Mertens*, 1923.) D, honey bee, queen, diagrammatic. E, *Mesembrina meridiana*, female. (*From Keilin*, 1916, *after Cholodkowsky*.) F, *Rhagoletis pomonella*, female.

Figure 20.3 (Snodgrass, 1935)

1.

Observe the demonstration dissection of the female reproductive system of a bristletail (Archeognatha, Machilidae), the most primitive of the insect reproductive systems (Fig. 20.4). Note that the ovarioles are segmentally arranged, with one each in abdominal segments 1 to 6 and another in the thorax. Each ovariole separately joins the lateral oviduct which in turns joins the opposite lateral oviduct to come a common median oviduct which opens in the gonopore behind sternum 7 at the base of the ovipositor. There is no well defined spermatheca or accessory glands; presumably the spermatophore is received in a membranous area at the base of the ovipositor, on segment 8. Note that this type of segmentally arranged reproductive system is also shared by Diplura (Fig. 20.3A).

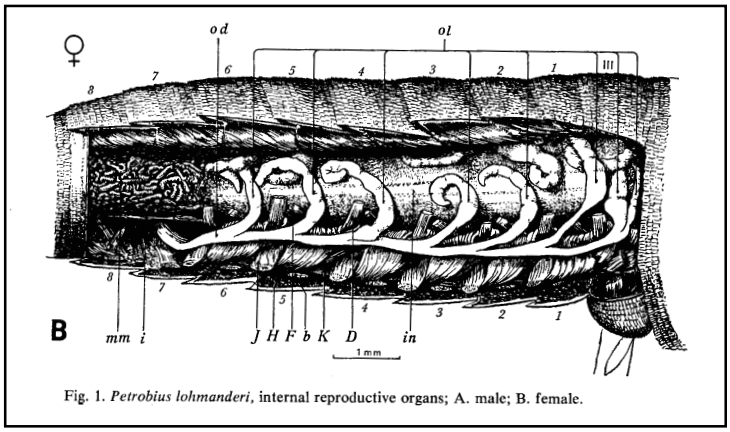

Fig. 1. *Petrobius lohmanderi*, internal reproductive organs; A. male; B. female.

Figure 20.4 (Birket-Smith, 1974. p 7)

2.

Examine the dissection of the female internal reproductive system of the roach *Nauphoeta cinerea* (Fig. 20.5). The pair of ovaries are conspicuous due to enclosed mature oocytes. Locate the following:

ovary
pedical
mature oocyte
median oviduct
young oocyte
ovariole
lateral oviduct

The oviducts are musculated and can be observed to contract in fresh preparations. What might the functions of these contractions be? The median oviduct enters a genital chamber which overlays the yellowish sac-like extension of the vagina called the uterus or brood sac. The eggs are retained here while they develop, as this animal is ovoviviparous and lays newly hatched nymphs.

A conspicuously blue-white mass of tubules lies at the caudal edge of the genital chamber, the accessory glands or colleterial glands. Posterior to these glands and openning on the roof of the genital chamber are a pair of long, translucent, twisted spermathecal glands which are in turn situated posterior to the bases of a pair of shorter, slightly knobbed spermathecae. The external genitalia or ovipositor are hidden beneath the uterus.

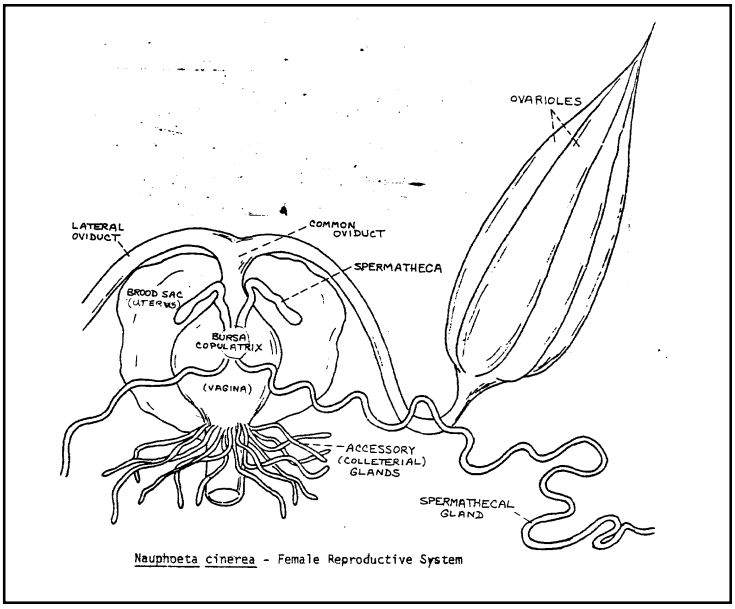

Figure 20.5 *Nauphoeta cinerea*. Female Reproductive tract.

3.

Examine the demonstration of the prepared longitudinal section of the grasshopper (Orthoptera) ovary (Fig. 20.6). Although this is a poor slide, the following structures can be recognized:

ovariole	**oocyte nucleus**
germarium	**oocyte**
vitellarium	**follicular epithelium**

What type of ovariole (panoistic, polytrophic, or telotrophic) does the grasshopper possess?

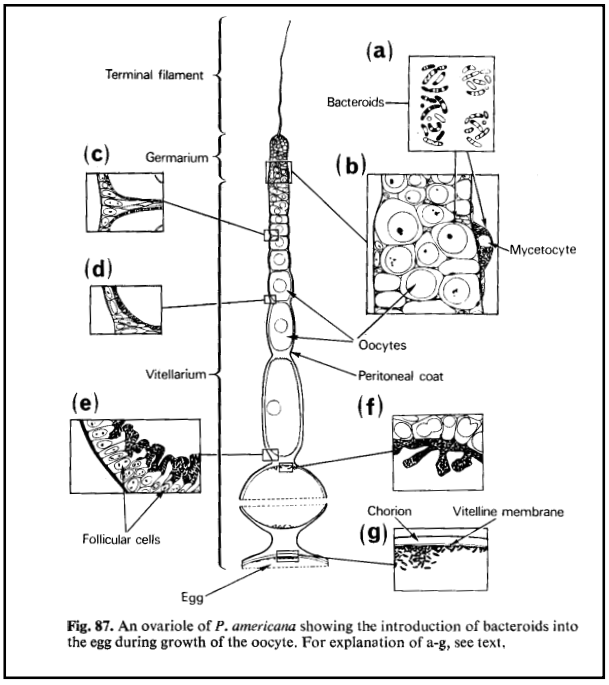

Fig. 87. An ovariole of *P. americana* showing the introduction of bacteroids into the egg during growth of the oocyte. For explanation of a-g, see text.

Figure 20.6 (Cornwell. 1968. p201)

4.

Examine the demonstration dissection of the reproductive system of a mature female black swallowtail butterfly (*Papilio*) (Fig. 20.7). Note that most of the abdomen was filled with the ovaries that contain the mature oocytes, arranged like strings of beads. In the Lepidoptera, the copulatory opening (vulva) is located on segment 8 and is separate from the opening of the vagina, the oviporus on segment 9. In the dissection, locate the vagina which is connected by a duct to the large, muscle-lined bursa copulatrix.

In the demonstration of the exterior of the Lepidopteran abdomen, note that an appendicular ovipositor is lacking. The abdomen ends in two anal pads ("papillae"), modifications of the terga used to manipulate eggs. Locate the vulva and the oviporus.

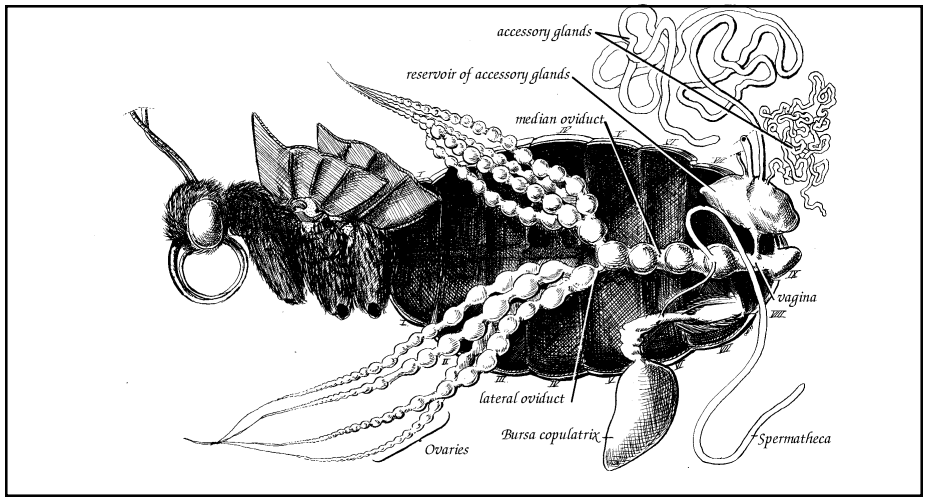

Figure 20.7 *Papilio*. Female reproductive tract

5.

Examine the demonstration dissection of the reproductive system of the worker paper wasp, *Polistes* (or *Vespula*) (Hymenoptera: Vespidae) (Fig. 20.8). As in other aculeate Hymenoptera, the ovipositor is modified to form a sting and the eggs issue from the base of the sting. Note the 2 accessory glands that enter the sting chamber via ducts: the venom gland, with its huge muscle-lined reservoir (venom sac) and pair of tubules that pick up venom precursors from the blood; and the Dufours gland (not visible in the illustration) that may produce pheromones. Note that the ovaries are undeveloped and the spermatheca (connecting dorsally via a duct to the common oviduct) is empty; characteristics of the worker caste in social Hymenoptera.

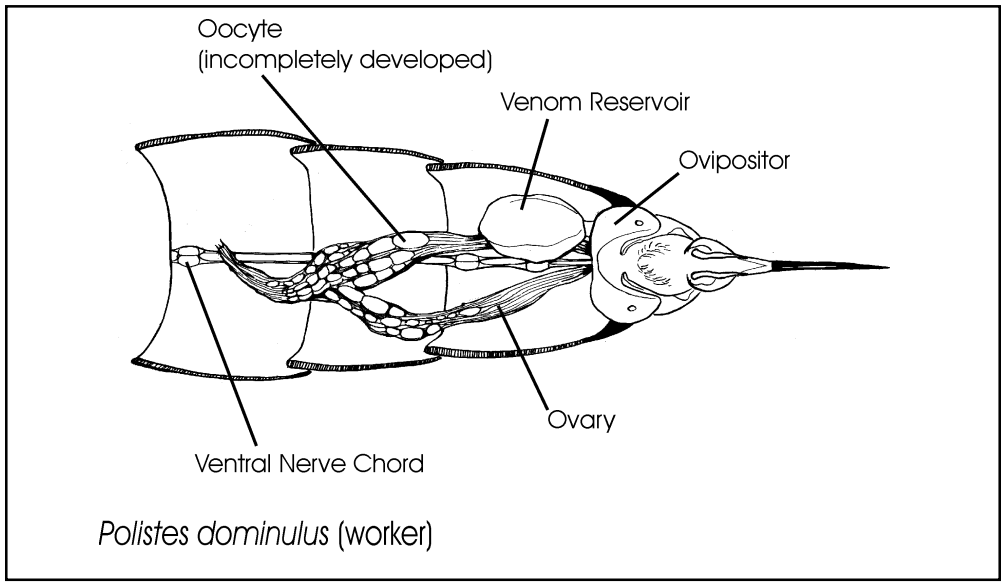

Figure 20.8 *Polistes dominulus*. Female (worker) reproductive tract (A. Burke)

6.

Obtain a female of your insect. Make 2 lateral longitudinal cuts along its abdomen and carefully peel back the terga. Be careful not to destroy the ovaries, which often lie lateral to and dorsal to the gut. Carefully remove the gut and fat body, exposing the reproductive system. The terminal abdominal nerve ganglion and nerves probably overlie the reproductive system and must be removed. The reproductive system may be carefully removed from the adjoining sterna and examined free from the body. Behind which sternum does the vulva open?

Sketch #5: Female reproductive system, dorsal view

Sketch the female reproductive system of your insect from dorsal view. The external genitalia may be roughly indicated in outline or ignored at this time. The sketch should be shaded. Identify if present:

ovary
ovariole
terminal filament
oocyte in vitallarium
pedicel
lateral oviduct
median oviduct
genital chamber or vagina
bursa copulatrix (if distinct) developing
spermatheca
spermathecal gland
accessory glands

7.

Using the above preparation, or a comparable one from your own insect, prepare the whole mount according to the following instructions. The reproductive system should be first "fixed," and then preserved in 70% ethanol. This exercise may be left to the following laboratory period.

ENTOMOLOGY 322
Preparation of Whole Mount

This exercise is designed to give you some experience in one method of preparing cleared, stained, permanently preserved whole mounts of internal organs of insects.

As per previous instructions, prepare the reproductive system, preserved in 70% ethanol. As the final preparation will be somewhat brittle, you should make sure that all crud is removed from the system and that it will lie reasonably flat on a slide. Stender dishes should be used for the below:

DAY 1
1 - transfer to 50% ethanol for 10 minutes.

2 - transfer to Grenacher's alcoholic borax-carmine stain. Leave about 24 hours.

DAY 2

3 - transfer to 50% ethanol for 1-2 minutes.

4 - transfer to 70% acidified ethanol. Leave until specimen assumes bright, translucent color. This may take several hours.

5 - transfer to 80% ethanol for 10 minutes.

6 - transfer to 95% ethanol for 10 minutes.

7 - transfer to second rinse 95% ethanol for 10 minutes.

8 - transfer to 100% ethanol for 10 minutes.

9 - transfer to second rinse 100% ethanol for 15 minutes.

10 - transfer to terpineol-toluene clearing agent for overnight or longer.

DAY 3

11 - mounting: Clean slide and cover slip with soap and water, dry thoroughly. Prepare supports to hold cover slip above specimen from plasticine clay, fragments of cover slip, cut strips of celluloid, or prepared "cells," depending upon thickness of specimen. Place large glob of Canada balsam on center of slide with glass rod. Place specimen in glob and orient it. Place supports to either side. Add more balsam if needed. Place cover slip over preparation. If balsam does not flow to edge of slip, place drop of balsam on slide next to slip and allow to flow under. Final orientation of specimen may be done under edge of slip with minuten probe. Label slide in marking pencil with your initials. Place on warming tray to dry. Clean utensils, hands, etc., with xylene.

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12 - allow to dry several days to a week. Clean off excess balsam with razor blade or xylene. Prepare label as follows:

name of sp., order & family
reproductive system (female)
Grenacher's alc. borax-carmine stain
date of preparation
your name

13 - store slide horizontally.