

UNIT 2: BIOLOGY OF SILKWORM- LIFE CYCLE OF BOMBYX-MORI STRUCTURE OF SILK GLAND AND SECRETION OF SILK

Systematic Position

Class: Insecta

Order: Lepidoptera

Family: Bombycidae

Genus: Bombyx

Species: B. mori

The Silkworm, *Bombyx mori* produces the silk of commercial importance. It is the caterpillar of a moth whose cocoon is used to make silk. This insect is also called the silkworm-moth and the mulberry silkworm. Male and female moths are flightless and lack functional mouth parts. The moths differ in morphological features. The female has a larger abdomen whereas the male has a much larger pair of antennae.

The life cycle of *Bombyx mori* demonstrates the most advanced form of metamorphosis. The serial progressions of four distinct stages of development complete one generation of the species; egg (ova), larvae, pupa and imago.

Ova:

Egg is the first stage of a silkworm's life cycle. The female moth lays an egg about the size of an ink dot during summer or the early fall. The egg remains in dormant stage until spring arrives. The warmth of the spring stimulates the egg to hatch. The egg of *Bombyx mori* is a very small and hard structure; about the size of a pin head and resembling a poppy seed. The egg shell provides a protective covering for embryonic development. When first laid, an egg light yellow. A fertile ovum darkens to a blue-gray within a few days.

Larva:

The larva is the vegetative stage where growth takes place. The larva of *Bombyx mori*, commonly called a silkworm, is host specific to mulberry. During growth, the larva molts 4 times. The period between successive molts is called an instar. The silk worm, upon hatching, is about 1/8th of an inch and extremely hairy.

Young silkworms can only feed on tender mulberry leaves. However, during the growth phase they can eat tougher mulberry leaves as well. The larval stage lasts for about 27 days and the silkworm goes through five growth stages called instars, during this time. During the first molting, the silkworm sheds all its hair and gains a smooth skin.

Pupa:

As the silkworm prepares to pupate, it spins a protective cocoon. About the size and color of a cotton ball, the cocoon is constructed from one continuous strand of silk, perhaps 1.5 km long (nearly a mile). The silk cocoon serves as protection for the pupa. Cocoons are shades of white, cream and yellow depending on silkworm genetics. After a final molt inside the cocoon, the larva develops into the brown, chitin covered structure called the pupa. Metamorphic changes of the pupa result in an emerging moth

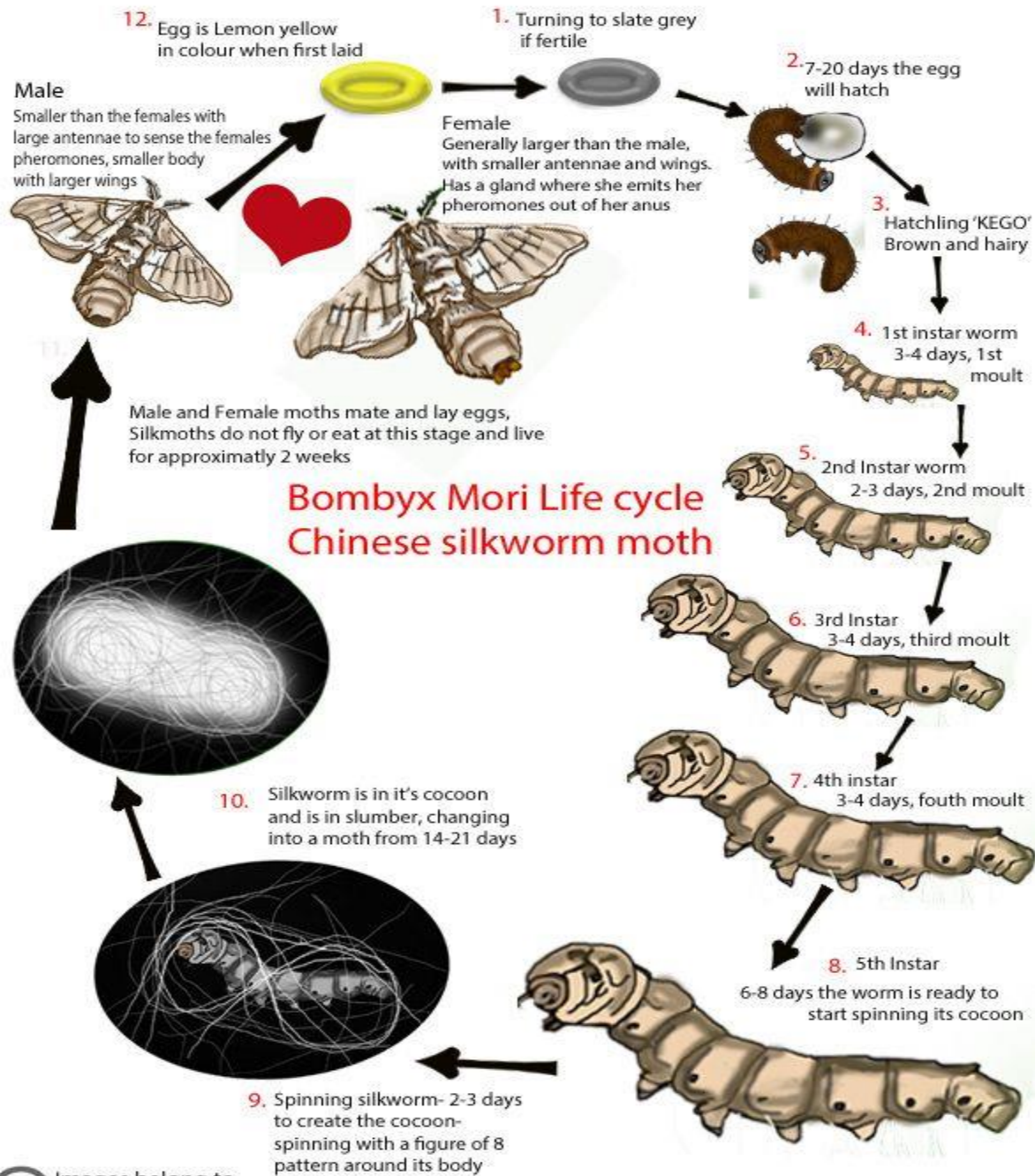
If the silkworms are allowed to mature and break through the cocoon, the silk would be rendered useless for commercial purposes. So the encased insect is plunged into boiling water to kill the inhabitant and dissolve the glue holding the cocoon together. The end of the silk is then located and the cocoon unwound onto a spindle to be made into thread.

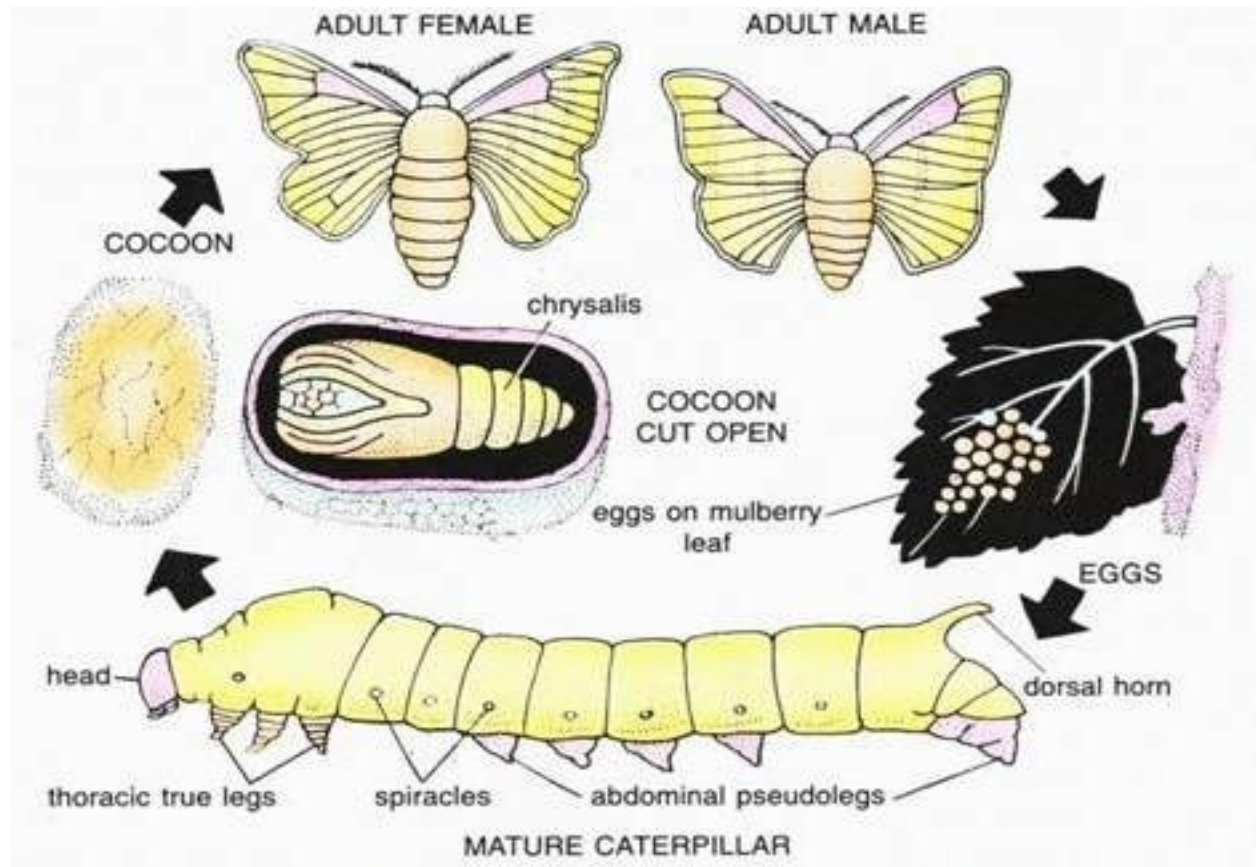
Cocoon:

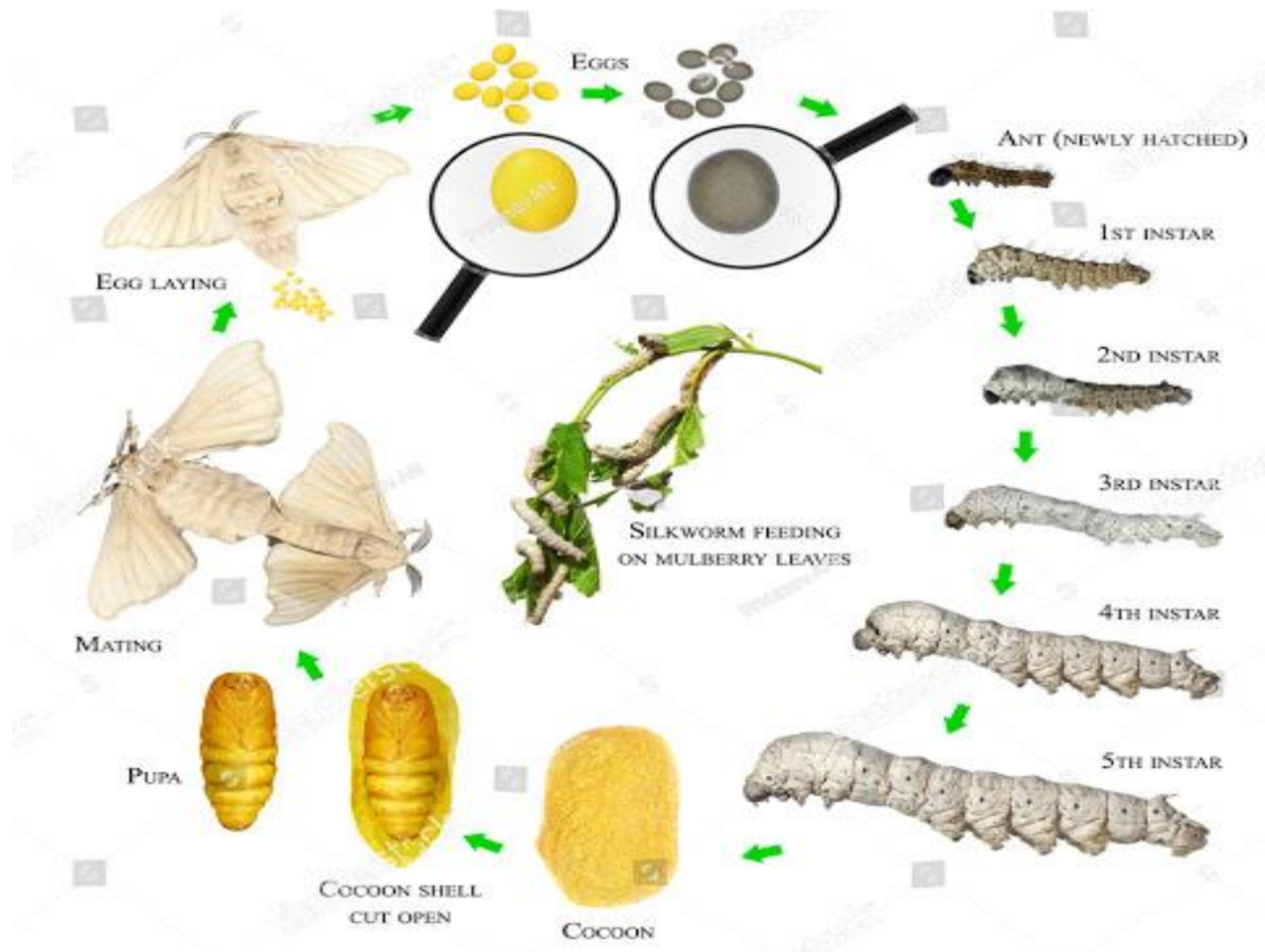
Cocoon is the stage in which the larva spins silk threads around it, to protect itself from its predators. The larva traps itself inside the cocoon in order to pupate. The color of the cocoon varies, depending upon what the silkworm eats. It can range from white to golden yellow. The second molting occurs inside the cocoon, when the larva turns into a brown pupa. It takes about 2-3 weeks for the pupa to metamorphose into an adult moth.

Imago:

The adult stage completes the life cycle of *Bombyx mori*. It is the reproductive stage where adults mate and females lay eggs. Moths are flightless and lack functional mouth parts, so are unable to consume the food/nutrition.

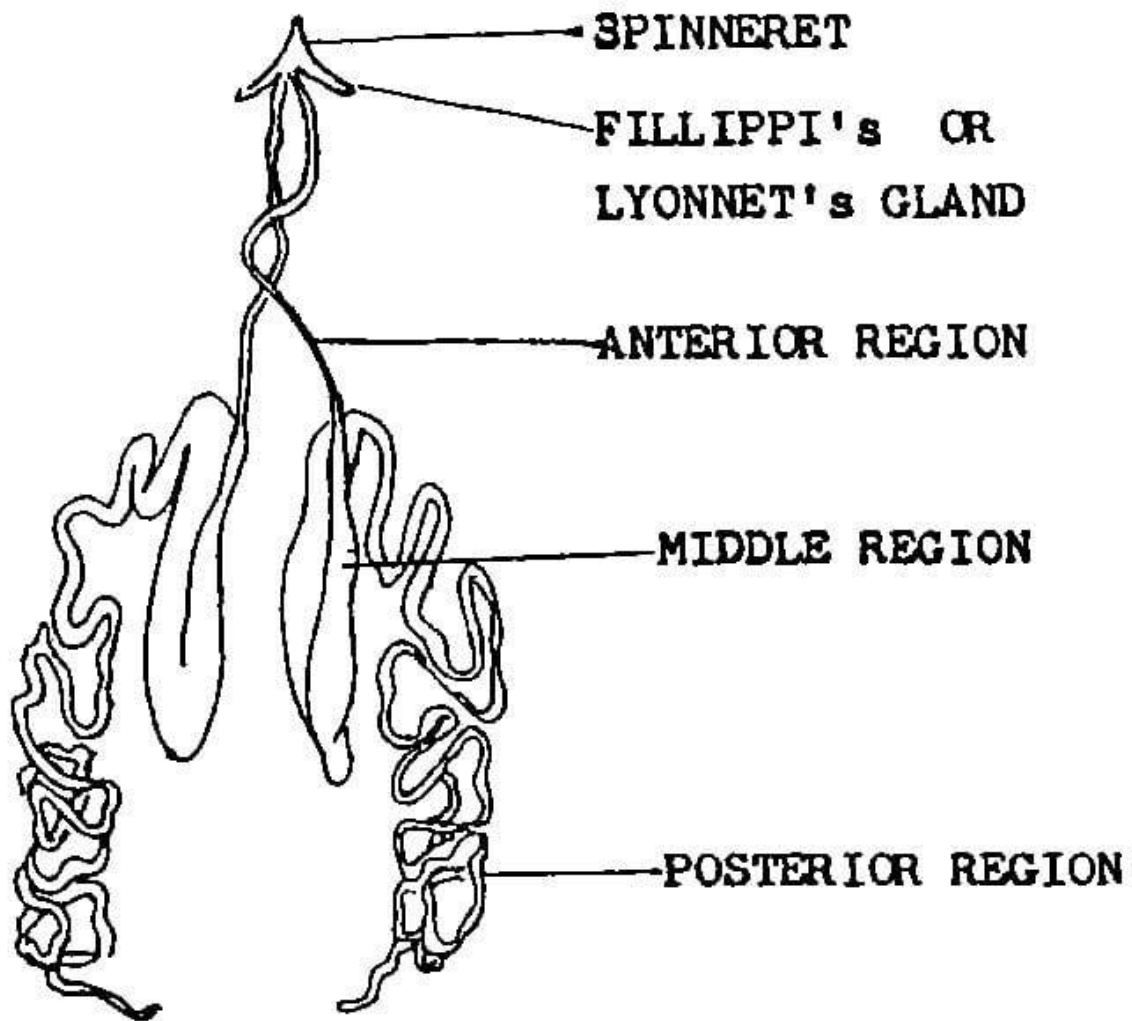






SILK GLAND

The silk thread of the cocoon is secreted by a pair of silk glands which are actually modified labial glands. These dermal glands are well differentiated in the fourth and fifth instar larvae. They lie below the alimentary canal and are so large in the fully grown final instar larva that it occupies most of the body cavity ventral to the alimentary canal and accounts for about 50% of the weight of the larva.



The silk glands are tubular in nature and the width of the tube varies in different regions of the gland. The entire gland is formed of three layers. The outer tunica propria of uniform thickness, the middle glandular layer and the inner tunica intima of varying thickness. The inner tunica intima is very thick in the anterior region and is shed at each moult and in the other regions it is thin and not shed at each moult. The number of cells in the glandular layer is fixed in the first instar larva itself and during the growth of the gland through the different instars, there is only growth of the cells and no increase in cell number. The number of gland cells in the gland varies in the different races. European races have the maximum number of cells. There is a gradual decrease in the number of cells in the different races in the following order - Japanese Univoltine, Japanese Bivoltine, Chinese Bivoltine, Chinese Multivoltine and the Oriental race. The gland cells have

a characteristic branched nucleus in the mature condition but in the young worms, the nuclei are round in shape.

There are three distinct regions in the silk gland differing in structure and function. They are the **posterior region, the middle region and the anterior region.**

The posterior-most region is highly folded and the folds lie in the midst of the dermo-visceral muscles and are attached to the tracheal of the region. They secrete the major protein of the silk, namely fibroin. It is secreted as a precursor fibrinogen which on extrusion from the gland becomes denatured to fibroin.

The middle region is the most prominent region of the gland and is also the widest. It is folded into a W-shaped structure and hence has three limbs, the posterior, the middle and the anterior limbs. The middle region acts as a reservoir of the fibroin secreted by the posterior region and fibroin matures in this region during the storage period. The layer of sericin secreted by the posterior limb of the middle region is called sericin I, that added around sericin I by the middle limb is sericin n and that added around sericin II by the anterior limb is sericin III.

The anterior region is of uniform thickness and is very thin. It does not secrete any material and serves only to conduct the silk fibre assembled in the middle region to the spinneret. The anterior region of the two sides open at the base of the median projection in the labium called spinneret which draws out silk in the form of a fine filament. The silk is moulded to a thread as it passes through the silk press which resembles a typical salivary pump. The threads of the two sides are called the brins and the sericin layer of the two bind them together into a single filament or bave.

FILIPPI'S GLAND OR LYONNET'S GLAND

This is a pair of glands situated in the head region. They open into the anterior part of the silk gland near its opening into the spinneret. Its function is not established but it is assumed that it produces a waxy material which coats the silk threads. Some consider that its secretion lubricates the tube through which silk passes.

The silk is a continuous filament comprising fibroin protein, secreted from two salivary glands in the head of each worm, and a gum called sericin,

which cements the filaments. The sericin is removed by placing the cocoons in hot water, which frees the silk filaments and readies them for reeling.

The silk of silkworms is secreted by a pair of labial gland, known as silk glands. The silk glands lie ventral to the alimentary canal. In full grown larvae, these occupy most of the body cavity. The silk glands are tubular in shape with different diameters in different regions. Each gland has 3 distinct regions

(1) Posterior region:

Blunt, highly folded tubular posterior regions of both glands remain attached to tracheal bushes of silkworm. This part secretes fibroin as fibrinogen which converted to fibroin upon extrusion.

(2) Middle region:

Most prominent and widest part of silk gland. It remains folded in a W-shaped structure and thus has 3 limbs — posterior, middle and anterior limbs. The posterior arm secretes sericin-I. It gets surrounded by sericin-II secreted from the middle limb

This sericin again gets surrounded by sericin- III secreted from the anterior limb. The middle region of silk gland also acts as the reservoir of fibroin where the later gets mature during the storage period.

(3) Anterior region:

The thin anterior region of silk gland has no secretory role and only transports the assembled silk to the spinneret.

Spinneret:

It is a projection of the median part of the labium, which draws the silk out in the form of fine filament. The secreted silk comes out as a thread or filament as it passes through silk press which resembles a typical salivary pump. The two filaments coming out of two sides are called brins. The sericin (gum) layer of the two brins then bind together into a single filament or bave.

Histologically the entire gland has 3 layers:

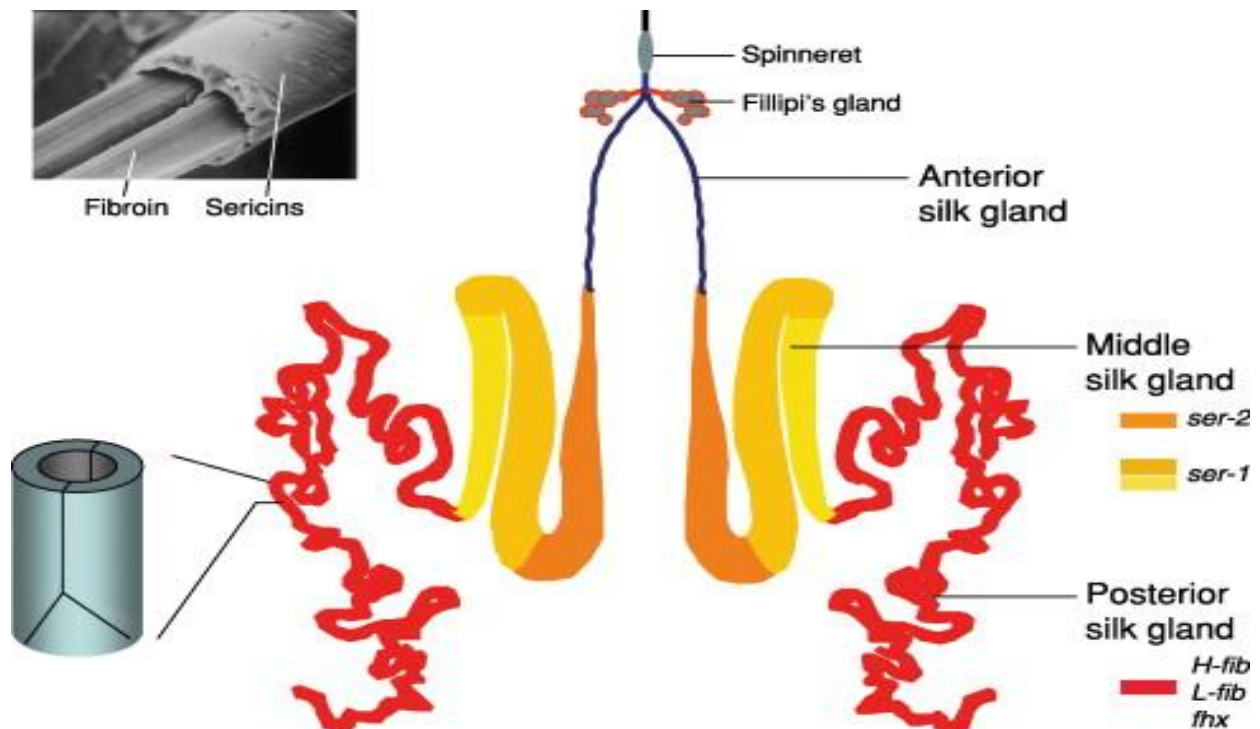
- (1) The **outer tunica propria** with uniform thickness;
- (2) The **middle glandular layer** with gland cells which increase in size during later instar stages of larval development and
- (3) The **inner tunica intima**: It has varying thickness.

In the anterior region of the gland, this layer is very thick and is shed at each moult. In other regions of silk gland, it is thin and not shed at each moult.

Filippi's gland or Lyonnet's gland:

In the head region of the larvae, a pair of glands is situated which open into the anterior part of silk gland near its opening into the spinneret. It is thought that these glands contribute some waxy materials to the silk thread or lubricate the passage of silk while coming out.

In the anterior region of the gland, this layer is very thick and is shed at each moult. In other regions of silk gland, it is thin and not shed at each moult.



SECRETION OF SILK

The silk gland is the only organ where silk proteins are synthesized and secreted in the silkworm, *Bombyx mori*. Silk proteins are stored in the lumen of the silk gland for around eight days during the fifth instar. Determining their dynamic changes is helpful for clarifying the secretion mechanism of silk proteins. Here, we identified the proteome in the silk gland lumen using liquid chromatography–tandem mass spectrometry and demonstrated its changes during two key stages. From day 5 of the fifth instar to day 1 of wandering, the abundances of fibroins, sericin, seroin and proteins of unknown functions increased significantly in different compartments of the silk gland lumen. As a result, these accumulated proteins constituted the major cocoon components. In contrast, the abundances of enzymes and extracellular matrix proteins decreased in the silk gland lumen, suggesting that they were not the structural constituents of silk. Twenty-five enzymes may be involved in the regulation of hormone metabolism for proper silk gland function. In addition, the metabolism of other non-proteinous components such as chitin and pigment were also discussed in this study.