Chapter

Larval Development and Molting

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Abstract

The term larva applies to the young hatchling which varies from the grown up adult in possessing organs not present in the adult such as sex glands and associated parts. Insect development is of four types namely Ametabolous, Paurometabolous, Hemimetabolous and Holometabolous. The larvae appear in variety of forms and are termed as caterpillars, grubs or maggots in different insects groups. The larval development consists of series of stages in which each stage is separated from the next by a molt. It's a complex process involving hormones, proteins and enzymes. Insects grow in increments. The molting is the process through which insects can routinely cast off their exoskeleton during specific times in their life cycle. The insect form in between two subsequent molts is termed as instar. The number of instars varies from 3 to 40 in different insect orders depending on the surrounding environmental and other conditions such as inheritance, sex, food quality and quantity. The larvae are categorized into four types namely Protopod larva, Polypod larva, Oligopod larva and Apodous larva.

Keywords: instar, insect, larval development, molting

1. Introduction

A larva is a distinct immature developmental form of many animals particularly in insects. The term larva applies to the young hatchling which varies from the grown up adult in lacking some important organs like sex glands and associated parts. The animals such as insects, amphibians and cnidarians with indirect development typically have larval phase in their life cycle. The diet of the larva is considerably distinct from the adult.

The larval forms are often adapted to different environments than of adults. For example, larvae of mosquitoes live almost exclusively in aquatic environment during their developmental stages and live outside water after metamorphosing into adult forms. Such adaptations in distinct environments are for their protection from predators and to avoid competition for resources. During developmental stages, larvae consume more food to fuel up their transition into adult form. In some insect species immature forms are totally dependent on adult forms for feeding such as in social insects of orders Hymenoptera (e.g., bees, wasps, ants) and Isoptera (e.g., termites) and the female workers feed them.

There are several advantages of an embryo developing into larval form instead of growing into an adult directly because it would help the animal to overcome various difficulties. The hatchling may need to obtain food but due to its smaller size is unable to feed itself the same way as an adult does. Further, it would be unable to make an effective use of defense mechanism as done by adult. Thus, the new organization of freshly emerged organism is best suitable to its environment. Further it furnishes a mode of life which is better suited to newly emerged small hatchling. The additional advantage of this corresponding organization is that it enables the larva to exploit an entirely different environment from that of its grown-ups. Thus, a terrestrial adult may have aquatic larval form such as in order Odonata (Dragonflies and Damselflies), a flying adult may have burrowing larvae as in order Diptera (Flies) and an adult may have free-living larvae in order Trichoptera (Caddisflies).

The arthropods cast off their cuticle at regular intervals to undergo a brief period of development before reaching mature size. Post-embryonic development is divided into a series of stages in which each stage is distinct from the next one by a molt. The larval forms usually change in shape during their development and progressive stages are not similar in insects. This change in form is known as metamorphosis. These changes are controlled by a juvenile hormone which is secreted by glands-corpora allata present in the posterior region of insect's head. It is released during each molt and its amount decreases each time. As the concentration of juvenile hormone declines, more adult characters appear and the adult stage is produced. In arthropods, the larval forms move between stages by molting of their exoskeleton. The new exoskeleton develops beneath the old skin. During the formation of new exoskeleton, insect's body gets swelled up due to intake of either air or water until the old exoskeleton breaks down. The newly formed exoskeleton hardens and different tanning agents get deposited onto the surface. After the succession of molts, an insect reaches the final adult form and no further molt takes place. Each developmental stage of an arthropod between molts is termed as Instar. For example, after hatching from egg, the hatchling is said to be first instar. When the insect molts again, it is then a second instar and so on.

2. Patterns of growth and development in insects

There are four patterns of growth and development in insects namely Ametabolous, Paurometabolous, Hemimetabolous and Holometabolous.

2.1 Ametabolous development (simple metamorphosis)

It is the type of insect development in which there is no metamorphosis. The emerged immature stage appears very similar to adult except that it lacks sexual structures. It grows only in size by replacing its old skin through molting. The larva grows bigger and the genitalia develops progressively with each molt. The young one which emerges from egg resembles adult in miniature form, is called nymph. The reproductive organs are undeveloped in nymph and after molting the nymph becomes an adult. Both forms i.e., the nymphs and adults live in the same habitat.

This is the characteristic feature of Apterygotes (e.g., Silverfish-*Lepisma* Linnaeus and Springtail). For instance, the silverfish hatched from egg looks like an adult and undergoes subtle anatomical changes between molts (**Figure 1**). Immature silverfish molts 6–7 times until it reaches sexually mature adult stage. In favorable conditions, silverfish may typically continue to molt during its lifespan and molts 25–66 times [1].

2.2 Paurometabolous development (gradual metamorphosis)

Paurometabolous development is found in less primitive forms like cockroaches, grasshoppers, praying mantis and white ants. In this type of development, the

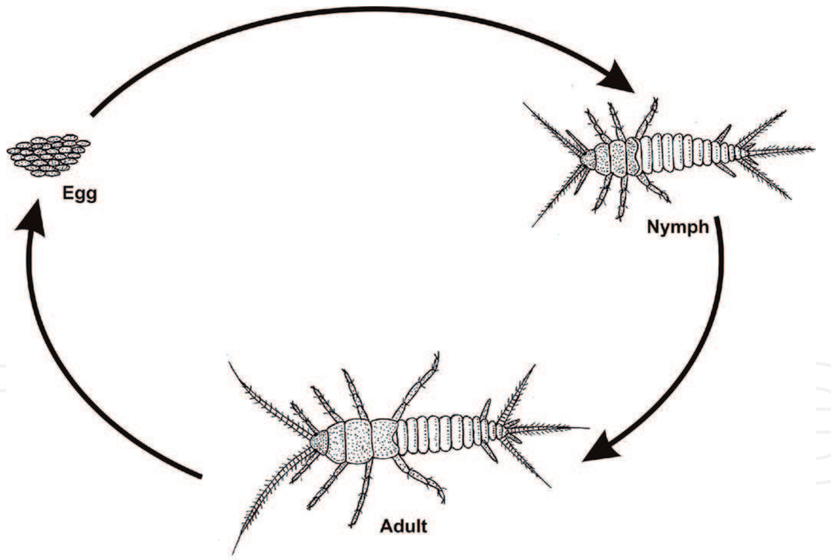

Figure 1.
Ametabolous development in Lepisma.

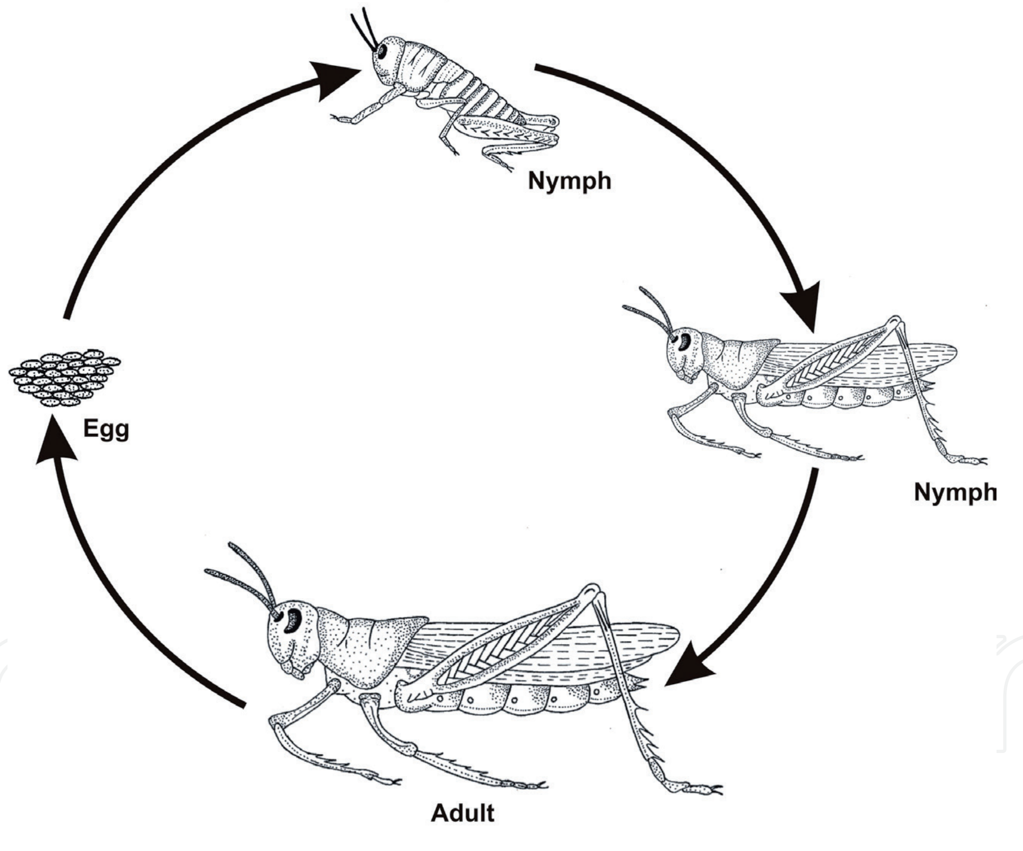

Figure 2.
Paurometabolous development in grass-hopper.

newly emerged young one closely resembles the adult in general body form, habits and habitat but many adult characters like wings and reproductive organs are not developed and their relative proportions of the body also differs. The young forms are termed as nymphs. The wings develop as wing pads on second and third thoracic segments at an early stage and gradually increase in size during each successive molt. The external genitalia also develops gradually after each molt. These nymphs lead an independent life and attain adult features through several molts. There are three stages in the life cycle of these insects i.e., egg, nymph, imago (adult) and no pupal stage is there. For instance in grasshoppers, before becoming adults the nymphs undergo 5–6 molts to change their body form (**Figure 2**). The nymph stage is species specific and lasts for a period of 5–10 days depending upon the weather conditions like temperature and humidity.

2.3 Hemimetabolous development (incomplete metamorphosis)

In this type of development, adult form is attained by gradual morphological changes with successive molts. The hatched larva lacks wings and genitalia but have some other characteristic features which are absent in adult. These features are lost at the final molt. The orders Plecoptera, Ephemeroptera and Odonata (**Figure 3**) have aquatic larval stages. The young forms are known as naiads which are aquatic and respire by external gills but the adults are terrestrial in behavior. Their life cycle also involves three stages: eggs, naiads and adults. When the naiads are ready to transform into adults, they come out of water and adult winged forms are released. The wings and genitalia develop externally but are not fully formed till adulthood. After the formation of wings no further molting takes place, only exception in mayflies where winged forms of aquatic nymphs come out and rest on trees to undergo final molting to become adults.

2.4 Holometabolous development (complete metamorphosis)

Complete metamorphosis is a kind of morphological change during post-embryonic transformation in which larva has no similarity with adult and there is always

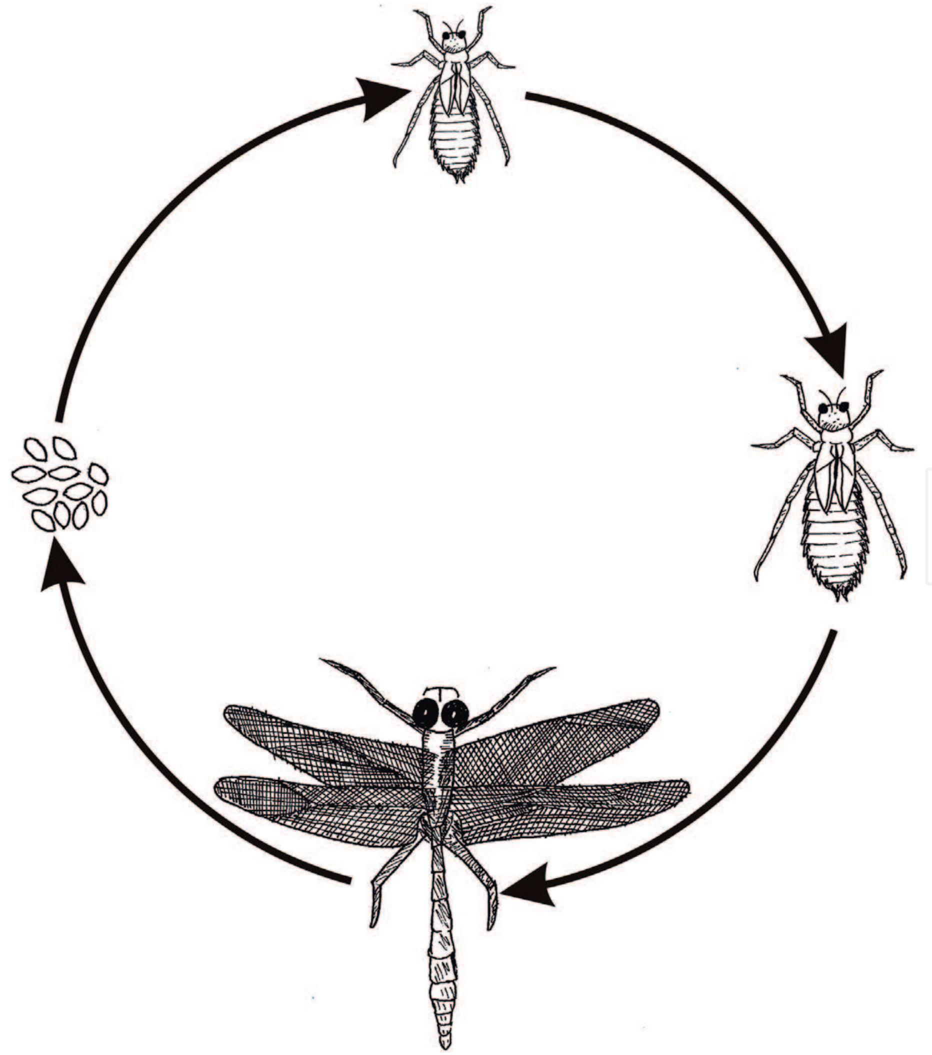

Figure 3.
Hemimetabolous development in dragonfly.

a pupal stage. Complete metamorphosis takes place in orders Coleoptera, Diptera, Hymenoptera and Lepidoptera. Pupal stage is the characteristic of holometabolous development i.e., this stage is present between the last larval stage and the adult.

In Order Lepidoptera (moths and butterflies), the larva is known as Caterpillar (**Figure 4**). It possesses a distinct head with powerful mandibles and three pairs of jointed thoracic legs. The abdomen has four or five pairs of un-jointed, short abdominal legs which are termed as pseudo-legs or prolegs. These caterpillars eat voraciously and grow rapidly with several moltings. After completing four or five molts, the caterpillar is transformed into pupal stage.

In Order Diptera (Houseflies and other flies), the larva is worm-like and devoid of appendages and is known as maggot (**Figure 5**). The mature larva is about 12 mm long. The head is indistinct, with a pair of oral lobes and hooks.

In Order Coleoptera (ground beetles, ladybirds and rove beetles), like adults the larvae referable to many beetle families are predatory in nature. The larval morphology is highly varied among species, with well-developed and sclerotized heads, distinguishable thoracic and abdominal segments and are known as grubs.

In Order Hymenoptera (bees and wasps), the larvae are grub-like with well developed head and mouthparts are of chewing type. Larvae are generally apodous, rarely eruciform with locomotory appendages.

2.5 Types of larvae

The larvae in different orders of insects are known by different names i.e., larvae of butterflies and moths are termed as caterpillars and those of Diptera and

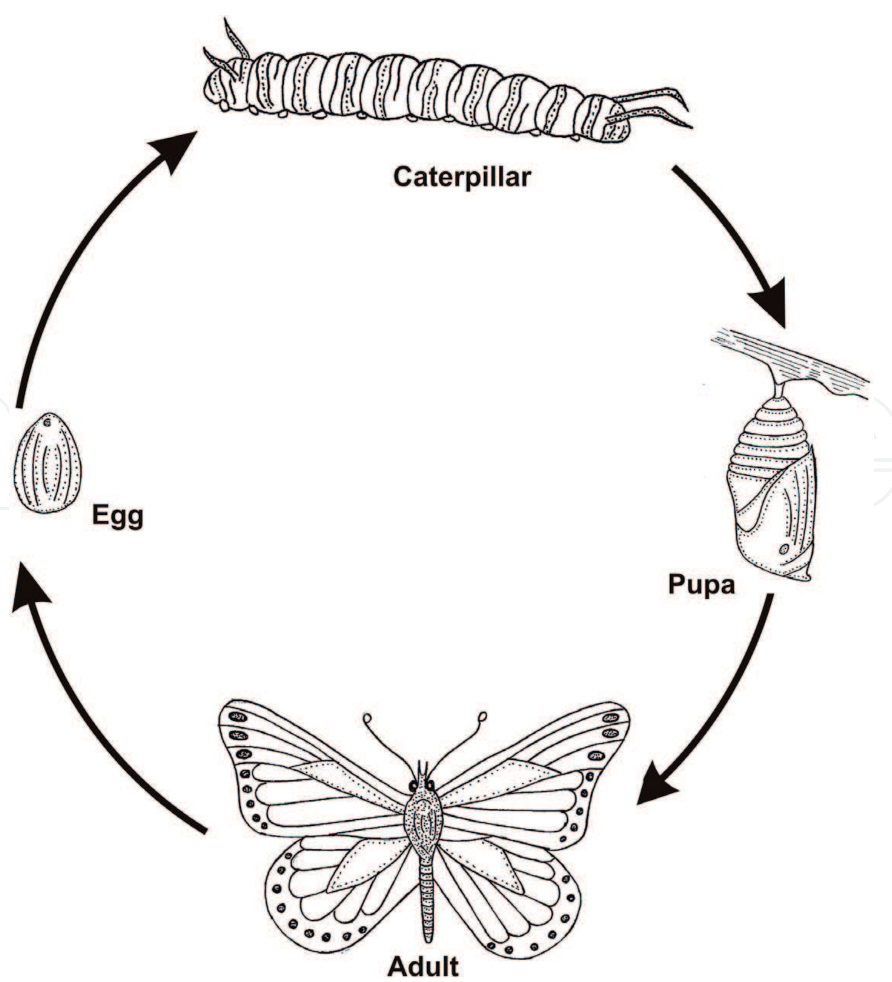

Figure 4.
Holometabolous development in butterfly.

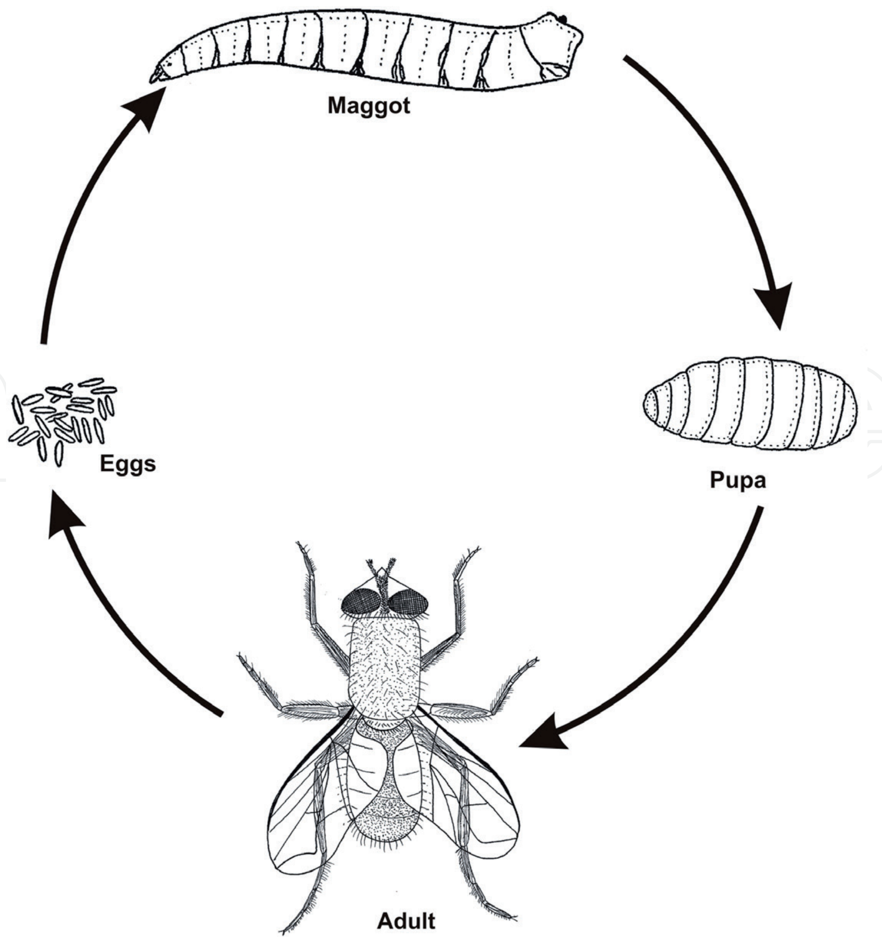

Figure 5.
Holometabolous development in housefly.

Coleoptera are termed as maggots and grubs respectively. The larvae are grouped into four types on the basis of development of appendages (**Figure 6**).

1. **Protopod larva**: In this type, larvae come out from the eggs which contain very little amount of yolk and this happens during the early stages of embryonic development. There is no segmentation on the abdomen. The thoracic appendages and head (cephalic) are primitive in form e.g., endoparasitic larvae of order Hymenoptera.

2. **Polypod larva**: In this type, larvae have three pairs of thoracic legs and two to five pairs of abdominal prolegs. The body of the polypod larva is well segmented and is termed as "eruciform" (cylindrical type). Only prothoracic and abdominal spiracles are open in their respiratory system. Larvae of orders Mecoptera (Scorpion flies and hanging flies) and Lepidoptera (butterflies and moths) are of polypod type.

On the basis of number and location of prologs, the lepidopteran larvae are further classified into three types: caterpillar, semilooper and looper.

a. Caterpillar: Caterpillar is the larval stage in Order Lepidoptera. It has soft body that can grow rapidly between molts. It bears five pairs of prolegs which are present on 3rd, 4th, 5th, 6th and 10th abdominal segments and three pairs of thoracic legs. e.g., larvae of gram pod borer and Lemon butterfly.

b. Semilooper: The semilooper larva bears three pairs of thoracic legs and three pairs of prolegs which are present on 5th, 6th, and 10th abdominal segments e.g., Cotton Semilooper and Castor Semilooper.

c. Looper: The looper larva have three pairs of thoracic legs and two pairs of prolegs present on 6th and 10th abdominal segments e.g., Cabbage looper.

3. **Oligopod larva**: The body of the oligopod larva is well segmented. It have three pairs of thoracic legs and possesses well developed cephalic appendages. The prolegs are absent. In some oligopod larvae, a pair of cerci or similar caudal processes is present. Head capsule is well developed and mouthparts are similar to the adult. On the basis of structure, the oligopod larvae can be further classified into two types viz., Campodeiform type and Scarabaeiform type.

    a. Campodeiform type: The campodeiform larva has dorso-ventrally flattened and well sclerotized body which bears long thoracic legs and a pair of terminal cerci. This type of larvae is found in orders Neuroptera, Trichoptera, Strepsiptera and in some Coleoptera (e.g., Lacewing and Ladybird beetle).

    b. Scarabaeiform type: The larva in this type is fleshy and 'C'-shaped with poorly sclerotized abdomen and thorax. It bears short legs and terminal abdominal processes (cerci) are absent. These larvae are less active and sluggish in nature. Scarabaeiform larvae are mainly in Scarabaeoidea and also in some other Coleopterans (e.g., White grub, Rhinoceros beetle).

4. **Apodous Larva**: Thoracic legs or abdominal prolegs are absent in case of apodous larva and it has poorly sclerotized cuticle (e.g., Honey bee, House fly, Fruit fly). On the basis of degree of development of head, the apodous larvae can be further grouped into the following three types:

    a. Eucephalous Larva: In this type, larva has well sclerotized head capsule with relatively reduced cephalic appendages and is found in Nematocera (Diptera), Cerambycidae (Coleoptera) and Aculeata (Hymenoptera). E.g., Mango stem borer, Mosquito.

    b. Hemicephalous Larva: In this type, larva has reduced head capsule that can be withdrawn within the thorax. It is found in families Tipulidae and Tabanidae of order Diptera. E.g., Crane fly, Horse fly.

    c. Acephalous Larva: This type of larva has no head capsule and cephalic appendages. E.g., Larva of House fly).

Like larvae, the pupae are also of various types (**Figure 7**). These can be grouped according to the presence or absence of functional mandibles which might be used by the adult to emerge from the cocoon or pupal cell. The functional mandibles are present in decticous type of pupa, whereas in the adecticous type, the mandibles are not functional. The latter type can be subdivided into two: exarate and obtect. The exarate pupa has free appendages and the obtect have appendages glued to the rest of the pupal body. An exarate pupa enclosed in a puparium is termed as coarctate whereas the silken protective case of obtect pupa is known as cocoon.

2.6 Heteromorphosis

Heteromorphosis is the type of development characterized by radical change in forms between successive larval instars. The larval instars are pretty much similar in many endopterygotes. However, a larva experiences typical change in morphology

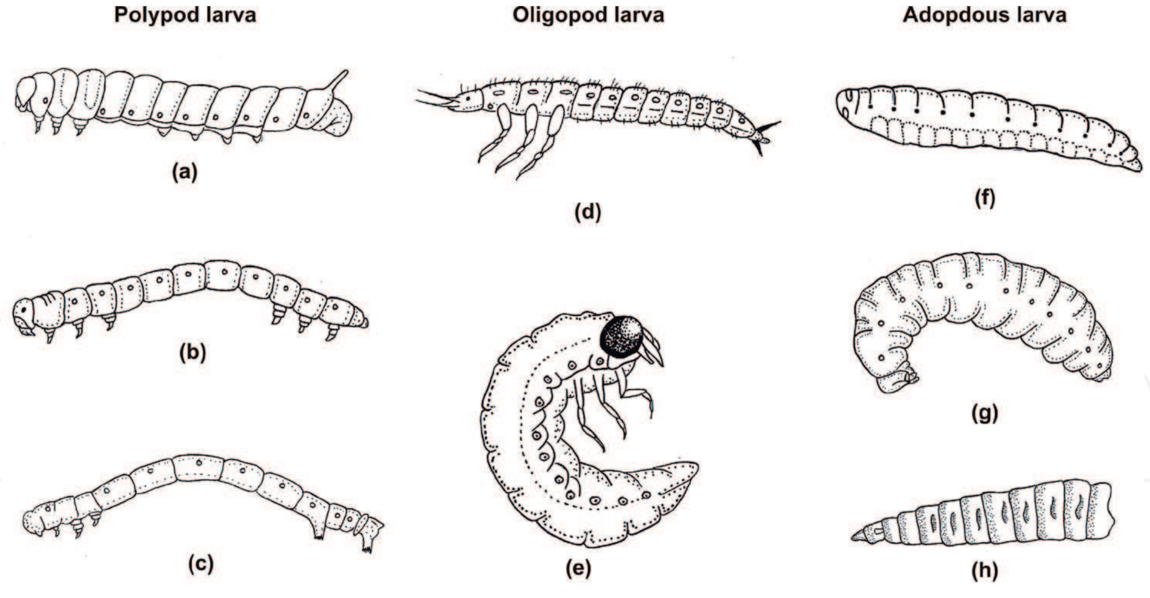

Figure 6.
Different types of insect larvae: (a) Caterpillar, (b) Semilooper, (c) Looper, (d) Campodeiform, (e) Scarabaeiform, (f) Eucephalous larva, (g) Hemicephalous larva, (h) Acephalous larva.

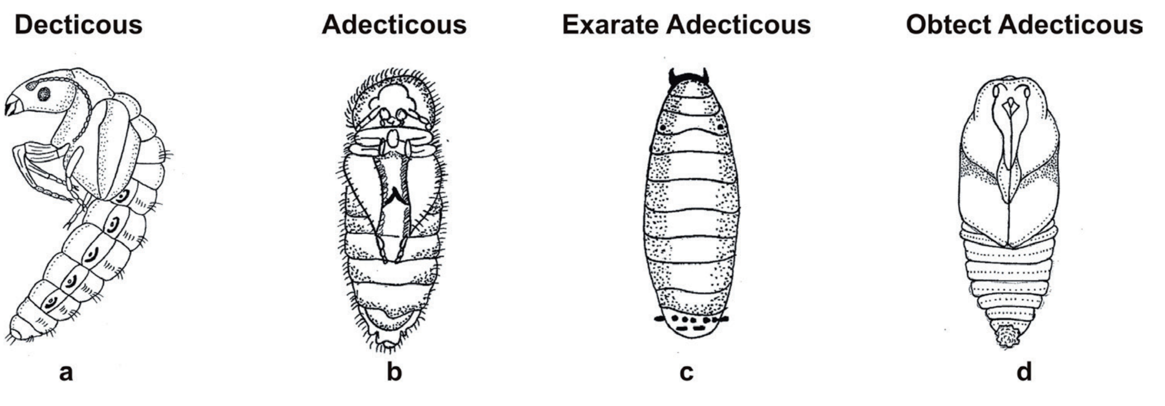

Figure 7.
Different types of insect pupae: (a) Decticous pupa (b) Adecticious pupa, (c) Exarate Adecticous pupa, (d) Obtect Adecticous pupa.

and in habits during development in some families of orders Coleoptera (Meloidae, Ripiphoridae), Diptera, Hymenoptera, Neuroptera and in all Strepsiptera. It is common in parasitic and predaceous insects where change in habit occurs during course of development. This is further of two types:

In first type, eggs are laid in open and the first stage larva searches for its host. In this type, the newly emerged first stage larva is an active campodeiform larva. For example, in Strepsiptera, larva attaches itself to a host often a bee or a sucking bug. When the bee visits a flower in which the larva is lurking, subsequently, it becomes an internal parasite and loses all traces of appendages and a series of dorsal projections starts developing which increases its absorptive area. The cephalothorax develops during sixth and seventh larval stages. Another example is of blister beetles (Meloidae), the larva hatches as free-living campodeiform which can actively search for food. After locating the food source, the larva soon molts to second stage i.e., eruciform (caterpillar like). Further, it has to pass through either two or more additional larval instars, where it may remain as eruciform or become scarabaeiform. A basically similar life history with an active first stage larva followed by inactive parasitic stages occur in Acroceridae (Diptera), Bombyliidae, Epipyropidae (Lepidoptera), Mantispidae (Neuroptera), Nemestrinidae (Diptera), Perilampidae, Eucharidae (Hymenoptera), Meloidae and some Staphylinidae (Coleoptera).

In second type of heteromorphosis, the eggs are laid in or on the host. It occurs in some endoparasitic Diptera and Hymenoptera. Among the parasitic Hymenoptera, the newly emerged larva is of protopod type larva. It has many different forms in different insect species. For instance, the first stage larva of *Helorimorpha* Schmiedeknecht of family Braconidae (Hymenoptera) has a small unsegmented body, a big head and a tapering tail. On the other hand the third stage larva is a fairly typical eucephalous hymenopteran larva. In Platygastridae, the first stage larva is more specialized, with an anterior cephalothorax bearing rudimentary appendages. These larvae hatch from eggs which contain very little amount of yolk.

2.7 Number of instars

During immature development, larvae of insects and other arthropods molt regularly by shedding their exoskeletons. Thus, instar is a developmental stage between two successive molts in the life cycle of an arthropod and the development period of larvae in insects is divided into a few discrete stages. The dissimilarity between instars is often observed in altered body proportions, patterns, colors, number of body segments, head width and appendages. Arthropods shed their exoskeleton in order to grow and to assume a new form. After shedding their exoskeleton, the juvenile insects continue their life cycle till they either pupate or molt again. The first larval instar stage begins at hatching and it ends at the first larval molt. In holometabolous insects, the last instar is a phase from final molt to either prepupal or pupal stage or the eclosion of an imago in hemimetabolous insects. The period of growth is species specific and is fixed for every instar. The larval instars number varies across various insect species.

An insect instar number also depends on the surrounding environmental and other conditions. The most common factors affecting the number of instars are temperature, humidity, photoperiod, physical condition, inheritance, sex, food quality and quantity. Lower temperature and less humidity often slow down the rate of development. In some insects, e.g., in salvinia stem borer moth, the number of instars relies on early larval nutrition. In addition, the presence of injuries has been observed to influence the number of instars in some species. During suitable conditions, the instar number is higher in exceptional species of orders Orthoptera and Coleoptera. Intraspecific variability in the number of larval instars is a widespread phenomenon occurring in most major insect orders, in both hemimetabolous and holometabolous insects. For instance, the hymenopteran egg parasitoid *Trichogramma australicum* (Girault) have only one larval instar [2], whereas 34 larval instars are reported in *Leptophlebia cupida* (Say) [3]. In some phylogenetically older orders like Plecoptera, Ephemeroptera and Odonata larvae have heavily sclerotized, non-expansible exoskeletons and the instar number is usually 10 [4, 5]. The larvae of a notodontid moth have an additional instar even when exposed to artificial rainfall [6].

Apart from other environmental factors, the inheritance and sex are the two factors which most commonly influence the instar number. The number of instars is usually sexually dimorphic and the females in general have a higher number of instars than males. The inherited factors affecting number of instars may be either hereditary or achieved by means of maternal impacts and further may rely upon environmental conditions encountered by a parent. Instar number might be genetically unique in larvae from various populations [7], between genetically determined phenotypes or between the offsprings of different individuals from the same population. Moreover, instar number may also differ genetically between subspecies [8], or between short and long winged individuals [9]. The instar number of progeny is influenced by the prevailing ecological conditions during the oviposition

Orders	Number of larval stages
Siphonaptera	3
Phthiraptera	3–4
Coleoptera	3–5
Hemiptera	3–5
Neuroptera	3–5
Diptera	3–6
Hymenoptera	3–6
Mecoptera	4
Zoraptera	4–5
Dermaptera	4–6
Embioptera	4–7
Lepidoptera	5–6
Thysanoptera	5–6
Trichoptera	5–7
Mantodea	5–9
Isoptera	5–11
Orthoptera	5–11
Psocoptera	6
Blattodea	6–10
Grylloblattodea	8
Phasmida	8–12
Thysanura	9–14
Ephemeroptera	20–40
Plecoptera	22–23

Table 1.
Number of larval stages in different orders of insects.

or larval period of parents. When reared in isolation, the nymphs of locusts namely *Schistocerca gregaria* Forsskal and *Nomadacris septemfasciata* (Serville) have more instars. In case of gypsy moths, the larvae which develop from the last laid smaller eggs of specific females usually have more instars [10]. There are some further factors which may particularly affect the number of instars. In case of termites, the larvae belonging to different castes also have different number of larval instars [11]. Lycaenid butterfly larvae have more instars when they live in association with ants and they have low pupal weight when ants are not present [12].

The larvae enter in a stage of inactivity i.e., remain motionless after final instar and stop feeding. This stage is known as pupal stage (chrysalis in case of butterflies). The larvae begin to resemble adults at the end of this stage due to the anatomical modifications that take place in them and also due to the appearance of new organs and tissues (**Table 1**).

3. Molting during post-embryonic development

In larval forms, when the exoskeleton is outgrown, the insects undergo molting regularly. In insects, the unique process of molting is under hormonal control and thus involves various hormones, proteins and enzymes.

During developmental phase when an immature insect needs a larger exoskeleton, the neurosecretary cells present in the brain are activated. It helps the larva to ward off its old exoskeleton. Thus molting is the phenomenon by which insects develops. Under controlled and protected conditions, it permits the body of the developing insect to expand. In order to increase in size the insect must sheds its skin in favor of new underneath skin. Insect can molt 5–60 times in the total life span depending upon its species. Stadium is the time interval between the two subsequent molts and instar is the form assumed by the insect in any stadium. Each instar ends with molting.

When there is no more space for the insect to expand inside its old exoskeleton, hormone triggers molting which separates the exoskeleton from the underlying epidermis and the molting fluid fills the newly created gap. Proteins are secreted by the epidermal cells to form a new cuticle. Later on, when the new cuticle is in place, the inner layer of the exoskeleton is digested by the enzymes present in the molting fluid. Epidermal cells recycle the chitin and proteins which are then secreted under the new cuticle.

With the formation of new exoskeleton the insects can further start shedding its old exoskeleton. The insect expands its body with the intake of large amount of air and the outer shell is forced to get split, usually down the dorsal side as a result of muscular contractions. The outgrown exoskeleton squeezes out the bud. It is a compulsion for the insect to expand and swell its newly formed cuticle which conclusively makes this new cuticle large enough so it can allow room for any further growth. The newly formed overcoat is much paler in appearance and is soft than that of the older one, however, it starts to become darker and hardens itself within few hours. The appearance of the insect seems like a slightly larger copy of its previous form.

The whole procedure of development of an insect is influenced mainly by three hormones: Prothoracicotropic hormone (PTTH), Ecdysone and Juvenile hormone which are secreted by neuro-secretory cells (NSC) present in the brain, Prothoracic gland (JH) and corpora allata respectively. The signals are sent by the developing body of the insect to the brain and direct it to activate the clusters of neurosecretory cells which then produce PTTH which passes down into neurohemal organ, Corpora Cardiaca (CC) to release stored PTTH into the circulatory system (**Figure 8**). The prothoracic glands get stimulated by this to secrete Ecdysone. The active form of ecdysone triggers a series of physiological events leading to the formation of a new exoskeleton by the process known as apolysis.

Along with serving the purpose of acting as a hormone release site, the Neurohemal organ also synthesizes hormones. It is the responsibility of JH to maintain the insect in its young state and it modifies expression of the molt, acts in conjunction with ecdysone. JH hormone favors the synthesis of larval structures and adult differentiation and thus considered as a modifying agent.

3.1 Regeneration

During larval development some insects are able to regenerate their appendages following their accidental loss. If the loss occurs early in a developmental stage, before the production of molting hormone, the appendage reforms at the next molt. The regeneration occurs in larval forms of Blattodea, Phasmatodea, in some Hemiptera, Orthoptera and holometabolous insects. The regeneration of cuticular structures can only happen at a molt as this is the only time at which new cuticle is produced. Consequently, regeneration of appendages does not occur in adults and is restricted to larval stages only. Regeneration of muscles and parts of the nervous system also occurs during development stages.

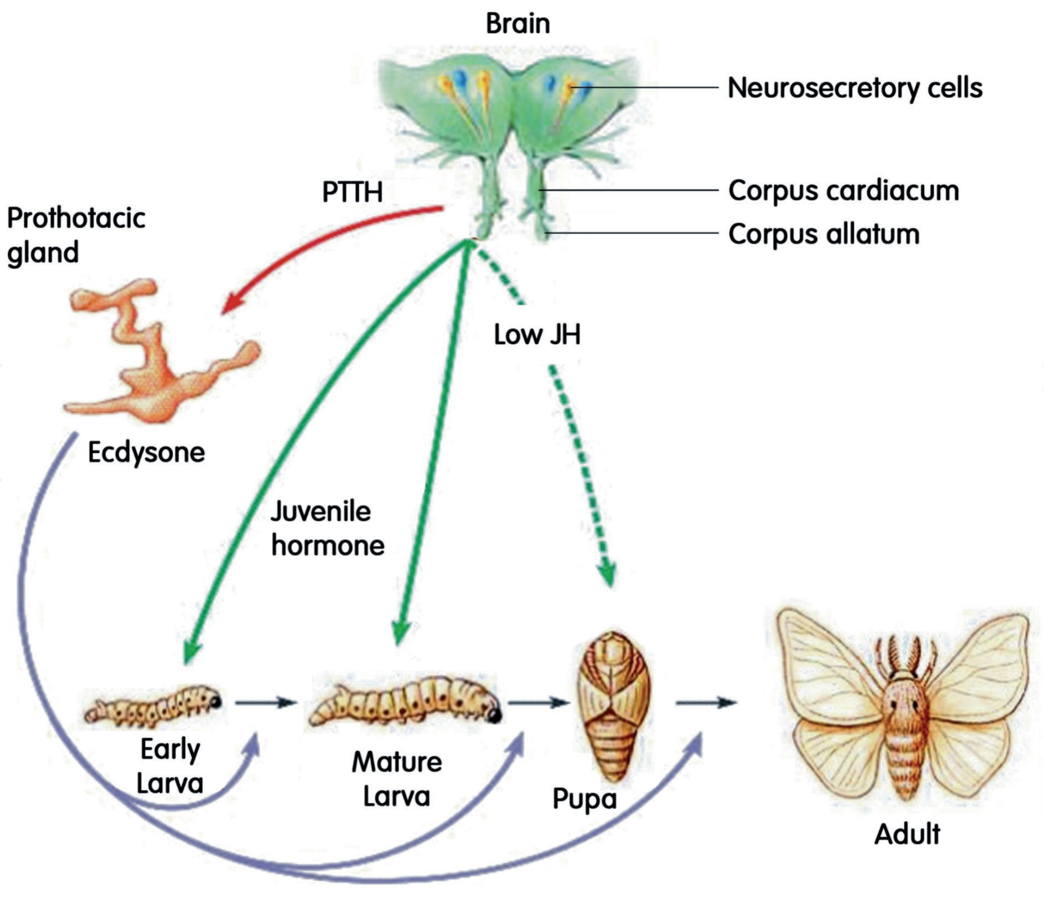

Figure 8.
Hormonal control of insect development.

4. Growth

In an organism, body size is one of the most important life history characters. Its effects on fitness are well documented and have been extensively studied both theoretically and empirically. Gaining weight and eating is the foremost function of the larva which leads to several developmental changes during this phase. The eyes, palpi, proboscis, antennae, reproductive organs and wings starts developing and the larval legs will develop into the adult legs. Prior to pupation, growth of these organs accelerates during the last one or 2 days, hence when pupa is formed the major important changes to the adult form have already been taken place. There is a progressive increase in weight throughout the developmental stages. Body size is flexible i.e., it can change in response to different environmental conditions. For example, insects developing at higher temperatures are generally smaller than those developing at lower temperatures and well fed organisms are typically larger than those fed at poor quality diet.

Normally the weight increases consistently throughout the phase of development and then falls slightly at the time of molting due to the loss of cuticle and some loss of water that is not replaced because the insect is not feeding. After molting, the weight quickly increases above its previous level. Expressed in terms of increase in absolute weight, the growth rate is usually greater in the later stages, but the relative growth rate normally decreases as the organism increases in size. In some aquatic insects before a molt there is no decrease in weight, but, at the same time, there is a sharp increase at the time of ecdysis due to the retention of water, either through the cuticle or by means of alimentary canal. This is utilized to build the volume of the insect, thus splitting the old cuticle.

D'Amico *et al.* analyzed the physiological basis of body size evolution in the tobacco hornworm, *Manduca sexta* (Linnaeus) of family Sphingidae. In this species, the final body size of the adult is determined by five variable factors: the initial size of the last larval instar, growth rate during that instar, critical weight, time delay between achieving critical weight and initiation of prothoracicotropic hormone (PTTH) secretion and timing of photoperiodic rate for PTTH secretion. They demonstrated that in a continuous laboratory culture over a 30 year period (approximately 220 generations) body size of this insect increased by 50%. This evolutionary increase in body size could be accounted for by changes in three of the five factors: growth rate, critical weight, and PTTH delay time (the remaining two factors remained unchanged during this period). The quantitative change in these three factors was shown to account for over 95% of the evolutionary change in body size [13].

In final (fifth) instar larvae of *Manduca sexta* (Linnaeus), somatic growth is causally associated with the timing of number of endocrine events that induce the onset of pupation and metamorphosis [14]. The growth stops and metamorphosis begins, with the secretion of PTTH and ecdysteroids in the final instar. PTTH and ecdysteroid secretions are inhibited by the presence of juvenile hormone (JH) [15]. The level of JH circulating in the hemolymph is high during the first few days of the instar but drops dramatically when the larva attains a specific critical weight. The larval growth stops when the sequence of endocrine and physiological events initiated by the critical weight culminates in the secretion of ecdysteroids [16].

Holometabolous insects acquire their adult biomass during larval growth. In this manner, food consumption is intense and the fat body enlarges amid larval development. However, they do not feed during metamorphosis and simply exploit the nutrients stored during their larval development. During metamorphosis, the fat body is reconstructed through cellular turnover to the degree that when the adult insect emerges, the fat body has been remolded or is completely replaced [17].

The crowding affects the rate of development and also influences the adult size. Insects from crowded conditions are generally smaller than the others developed in isolation. For example in *Aedes aegypti* (Linnaeus), crowding in the habitat generated lighter pupae, longer larval period and increased mortality [18].

4.1 Control of growth

Larval growth is characterized by periodic molts and to some extent the internal changes are correlated with the molting cycle. Larval growth is regulated by ecdysteroid molting hormone which helps in producing larval characters. While hormones exert an overall controlling influence, local factors are also responsible for controlling the form of particular areas in the larval body. For example, epidermal cells often show distinct polarity secreting cuticle in a form giving an obvious anterior–posterior pattern. In the first stage larva of *Schistocerca* Stal, the cuticular plates associated with each epidermal cell on the sides of abdominal sternites are produced into backwardly pointing spines. Similarly in seed bugs, *Oncopeltus* Stal, a row of spines marks the posterior end of the area of cuticle. The scales also grow out with a particular orientation in butterflies and moths. The experimental manipulation shows that the polarity of the cells within a body segment is produced by a gradient of a diffusible substance known as a morphogen.

In addition to having a specific orientation, cuticular structures are dispersed in regular patterns. For example, in assassin bugs (*Rhodnius* Stal), the larvae bear a number of evenly spaced sensilla. Sensilla are the smallest functional units of insect sensory system and form an essential interface between external and internal sensory environments of the insect. At each molt, these increase in number, new sensilla being formed in the biggest gaps between the existing sensilla. This is consistent with the hypothesis that if the sensilla become widely spaced due to the growth of epidermis, the morphogen accumulates between them. If the concentration of morphogen exceeds a certain threshold, the development of a new sensillum is initiated. The development of sensilla on the cuticle in adults of *Oncopeltus* Stal can be accounted for in a similar way. Where two or more integumental features are present in an integrated pattern, they may be controlled by the same substance. For instance, in *Rhodnius* Stal, it is suggested that a differentiating substance- morphogen in high concentration produces the sensilla and the same substance in low concentration initiates the development of dermal glands, which are thus arranged round each sensillum. In *Drosophila* Fallen, and almost certainly in other insects, the boundaries of the para segments are source of signals that organize the patterning and orientation of associated cellular fields [19].

There is relatively little information on control of growth of the integral organs, but some show cyclical activity which coincides with the molt. In *Rhodnius* Stal, the fat body cells exhibit a marked increase in RNA concentration and mitochondrial number just before a molt and the ventral abdominal intersegmental muscles become fully developed only at this time. In some insect orders, Malpighian tubules increase in numbers; mitosis and development of new tubules are phased with respect to each molt.

4.2 Increase in size of the cuticle

Integument is the external layer of tissue that covers the outer surface of insects and the surfaces of the foregut and hindgut. It is composed of epidermis which is a continuous single layered epithelium, an underlying thin basal lamina and the extracellular cuticle that lies on top of the epidermis. An extracellular layer i.e., the cuticle covers the outer surface of the insect's body. It serves dual function. Firstly, it acts as a skeleton for muscle attachment and secondly, as a protective barrier between the insect and its respective environment.

Depending upon insect species, its developmental stage along with the body region, the cuticular layer may vary in thickness which can range from few micrometers to millimeters. It can be as thin as 1 μm in the hindgut and over gills (Ephemeropteran larvae) and as thick as 200 + μm (elytra of large beetles). The cuticle is highly diverse in their mechanical properties and can be divided into two groups: stiff, hard cuticles and soft, pliant (easily bent) cuticles. It also differs in color and in surface sculpturing, but electron microscopy shows that all types of cuticles are built according to a common plan. It is the essential part of the integument which further has the cuticle producing epidermal cells, sense organs and various glands. Epicuticle, procuticle and subcuticle are three distinct layers of the cuticle (**Figure 9**). The epicuticle being the outermost covers the complete cuticular surface. Procuticle comprises the principle part of the cuticle. Subcuticle is present in between the procuticle and the epidermal cells. It is a thin histo-chemically well-defined layer. This layer serves as the accumulation zone where the newly formed cuticular material is assembled and added to the cuticle that already exists and also binds the cuticle and epidermis. In the early stages of the development of the cuticle, before the insect sheds the cuticle of its previous stage, the amount of protein per unit chitin is much less than in the mature developed cuticle [20].

The single celled layer of epidermis constructs the cuticle. The epidermis effectively stores the lamellate endocuticle in those regions where the cuticle is extensible during the intermolt period. At the apical surface of epidermal cells, the plaques of chitin and protein are discharged at the tips of microvilli. Above the plaques in the extracellular space, the cuticle arises by self-assemblage of chitin microfibrils and

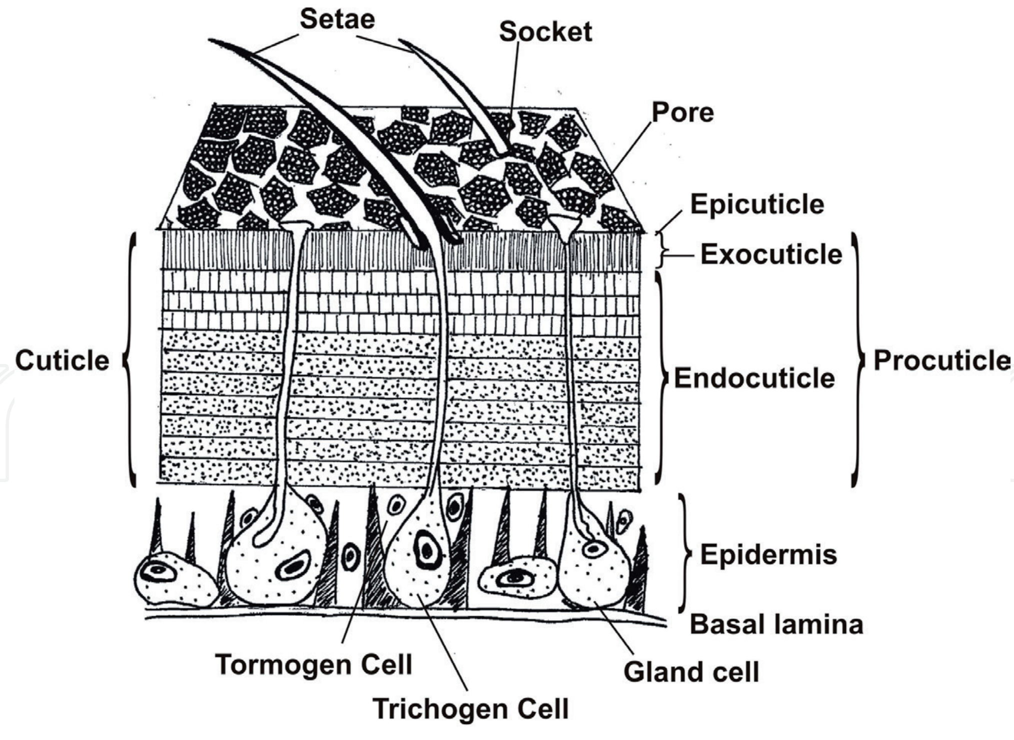

Figure 9.
Structure of insect integument.

secreted proteins. As the larva develops, the epidermal cells beneath the flexible cuticle also grow. The epicuticle in the case of soft-bodied insects e.g., in tobacco hornworm, *Manduca sexta* (Linnaeus) gets deposited in folds to permit development during the succeeding intermolt period. In this case, after ecdysis during the first day, the underlying endocuticle grows by means of apical expansion points created by the discharge of vertical chitin microfilaments. The epidermal cells segregate from the overlying cuticle and experience a burst of RNA synthesis at the beginning of the molt. Endocuticle synthesis comes to an end and then there is secretion of two inactive enzymes namely, chitinolytic and proteolytic. These enzymes form "molting gel" which fills the space in between the apical border of the epidermis and old cuticle. With the activation of these enzymes, the process of digestion takes place in old endocuticle. Then, cuticulin is deposited first at the tips of the plasma membrane plaques followed by deposition between the plaques to form a complete layer. Cuticulin is a structural protein in the cortical layer. It forms the outer part of the epicuticle and provides hardness and waterproofing to cuticle. The epicuticle precursors like polyphenols, lipids and proteins are secreted and congregated on the inner face of the cuticulin layer. Then, the action of phenoloxidases that cross-link the proteins and polyphenols stabilizes the entire structure, followed by the withdrawal of the apical membrane of the epidermal cells from the patterned surface which further starts the formation of procuticle.

4.3 Ecdysis of the old cuticle

Ecdysis is derived from the Greek word *ekdusis*, which means "put off." It describes the process by which arthropods and insects shed/cast off their outer cuticle (exoskeleton). While the new cuticle is formed, the older one remains intact and the muscles stay attached to the same cuticle to make it possible for the insect to move. Towards the end of the molt just before ecdysis, there is secretion of certain proteases into the molting gel. Then, these enzymes work together in the digestion of the chitin and proteins in the old endocuticle down to their components: N-acetyl glucosamine sugars and amino acids. Further, for the production of the next cuticle, the reabsorption of molting fluid takes place into hemolymph which helps these components to be recycled. The process of reabsorption happens in two manners. Firstly, back through new cuticle and epidermis and secondly, through the gut via swallowing and uptake in hindgut.

Close to the end, where molting fluid is reabsorbed the insect undergoes ecdysis i.e., shedding of the old cuticle which takes place in a stereotyped arrangement of behavior. This behavior is characterized by a series of coordinated movements which helps to loosen the muscle attachments with the old cuticle. After this phase, there is a series of peristaltic waves which travel from posterior to anterior and helps to make the insect to rupture the old cuticle anteriorly and to free itself. The cuticle opens at ecdysial sutures. These are the areas of old cuticle which lack exocuticle. In order Lepidoptera, the head capsule has slipped down over the forming mandibles early in the molt to allow the formation of a larger head capsule. The old head capsule isolates from the rest of the old cuticle during ecdysis and falls off as the new larva leaves its old cuticle.

Dermal glands i.e., Verson's glands secretes a waterproofing cement layer on the top of epicuticle. When the larva sheds its old cuticle this layer sprawls over the surface as a result of larval movements under the old cuticle. There are certain cases in which there is a secretion of waxy layer on the top of this layer for the first few days after ecdysis which helps in preventing desiccation. The pore canals transversing through the cuticle from the epidermal cells help in secreting this waxy layer.

4.4 Post-ecdysial expansion and sclerotization

The larva fills its tracheae with air after the process of ecdysis and furthermore swallows air so as to expand the new larger cuticle. After achieving its final size, the new cuticle solidifies. It gets dark or tanned to changing degrees relying upon whether the cuticle is to be rigid or flexible.

Sclerotization is the phenomenon of hardening the exocuticle by cross-linking the proteins together with the chitin to form a balanced structure appropriate for an exoskeleton which helps to anchor the muscles to permit the movement. N-β-alanyldopamine and N-acetyldopamine are the two essential cross-linking agents. The N-β-alanyldopamine is found in tan cuticles of many pupae belonging to order Lepidoptera. The key enzymes for the formation of these compounds are phenoloxidase for conversion of tyrosine to dopa and dopa decarboxylase for conversion of dopa to dopamine.

4.5 Growth of the tissues

The form of the cuticle is determined by the epidermis which may grow either by an increase in cell number or by an expansion in cell size. During developmental period the cell number increases just before molting in larval stages of insects. For instance, the size of the larval forms of *Cyclorrhapha* increases with the increase in the size of epidermal cells.

The growth of central nervous system in hemimetabolous insects does not involve the production of new neurons except in the brain. In the terminal abdominal ganglion of *Acheta* Linnaeus, for example, there are about 2100 neurons at all stages of development. On the other hand, the number of glial cells in the ganglion increases from about 3400 in the first stage larva to 20,000 in the adult and the volume of the ganglion increases 40-fold. There is extensive reconstruction of the nervous system in holometabolous insects during metamorphosis and undifferentiated neuroblasts persist through larval period.

During larval development, the marked changes occur in the sensory system of hemimetabolous insects. At each molt additional mechanoreceptors and chemoreceptors are added to the already present receptors. The ommatidia forming the compound eyes also increase by number. In contrast, the number of sensilla remains constant throughout the larval life in holometabolous insects and compound eyes are only present in the adults.

The musculature in larval forms closely resembles to that of adults in most hemimetabolous insects. In addition, there may be some muscles that are operative only during molting and later disappear after the final molt. During larval development, muscles increase in size and there is no basic change in their arrangement. The muscles grow by an increase in fiber size between molts and by the addition of new fibers at molts.

In case of epidermis, increase in the size of an internal organ results from an increase in cell size or in cell number or sometimes both. The increase in volume of internal structures especially the fat body is limited by the cuticle. In holometabolous insects, larvae with soft, folded cuticle, considerable growth is possible. The extension of the abdomen by unfolding inter-segmental membranes occurs in species with more rigid cuticles. In grasshoppers, and probably in some other insects, some growth of internal organs occurs at the expense of air sacs which become increasingly compressed during each developmental stage. The fat body of larval *Aedes* grows by an increase in cell number. In *Drosophila*, most of the tissues have constant number of cells and grow by cell enlargement. This enlargement is accompanied by endomitosis. In the midgut, both processes occur, the epithelial cells enlarge, but ultimately breakdown during secretion and each is replaced by two or more cells derived from the regenerative cells. In some insects the whole of the midgut epithelium is replaced at intervals by regenerative cells. In general, it appears that tissues which are destroyed at metamorphosis grow by cell enlargement while those that persist in the adult grow by cell multiplication.

The development of Malpighian tubules varies. In Orthopteroid orders, Malpighian tubules increase in number throughout the larval life. The primary tubules arise as diverticula from the proctodeum in the embryo. There are four primary tubules in *Blatta*, some insects have six. Secondary tubules develop later, largely post-embryonically. For example, *Schistocerca* Stal has six primary tubules, but twelve more are added before the larva hatches and more develop in each larval stage up to the adult. Secondary tubules appear as buds at the beginning of each larval stage, but after their initial development they increase in length without further cell division as a result of an increase in cell size. In holometabolous insects, the number of Malpighian tubules remains constant throughout their larval life but tubules increase in length by increasing their cell size and by cell rearrangement.

5. Conclusion

Many animals possess a distinct immature developmental form (e.g.) in case of insects larval forms are distinct during developmental period. The immature forms are much more adapted to environmental conditions than adults and consume more food to undergo the process of transition from immature to adult form. Larval stages undergo metamorphosis in which they usually change in shape, size and organization to form an adult. These changes are triggered and monitored by hormones such as juvenile hormone. Class Insecta is characterized by four different patterns of growth and development i.e., Ametabolous, Paurometabolous, Hemimetabolous and Holometabolous. Each pattern is characterized by specific morphological and hormonal changes. Insect larvae are broadly classified into four groups: Protopod larva, polypod larva, oligopod larva and apodous larva.

During the process of molting, the insect larvae molt with number of times. The number of instars varies across insect species. The environmental conditions like temperature, humidity, photoperiod along with other factors such as sex, inheritance, food quality and quantity affect the number of instars. Some insects can regenerate their lost appendages before the production of molting hormone. Regeneration can be seen in larval forms of Blattodea, Phasmatodea, some hemipterans and orthopterans. Larval development is also marked by significant changes in the sensory system in hemimetabolous insects. Mechanoreceptors, chemoreceptors are added along with the increase in size of muscles.

Larval development is a significant phase in the development history of an insect which molts and physiologically change the insect to adjust in different environmental conditions and habitats.

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