ELEMENTS OF
INSECT ANATOMY

COMSTOCK AND KELLOGG

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THE ELEMENTS

OF

INSECT ANATOMY

AN OUTLINE FOR THE USE OF STUDENTS
IN ENTOMOLOGICAL LABORATORIES

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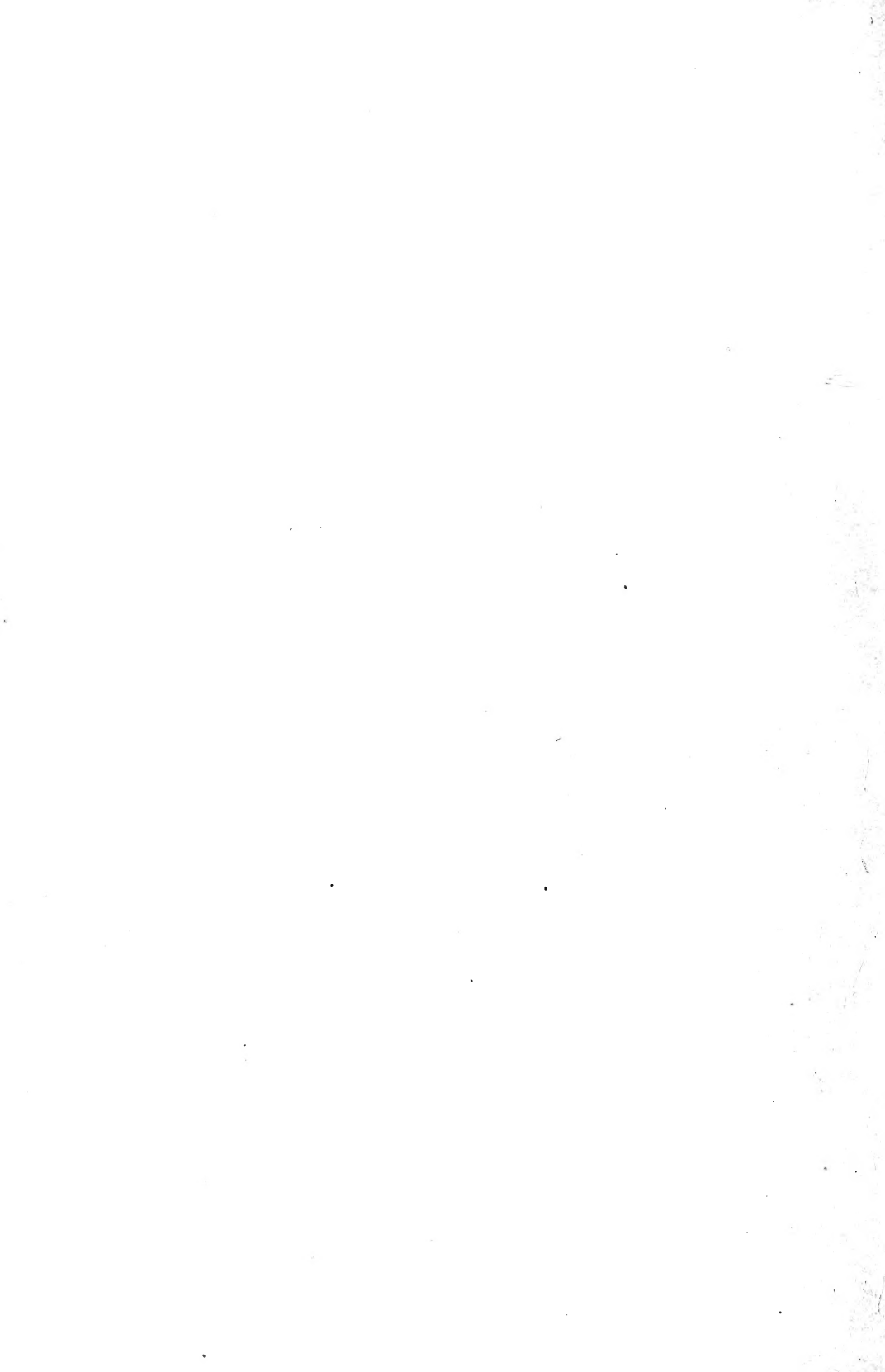Copyright, 1899, by
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PREFACE

The course of study outlined in the following pages is de-signed to enable students to learn the more general features of the structure of insects. It may serve as an introduction to a more extended study of insect morphology.

While the more obvious object of this course is the learn-ing of certain facts, a much more important thing to be gained is a training in methods of careful observation. The student is urged, therefore, to do the work with great care.

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CHAPTER I.

Need of a technical nomenclature.—It has been found that the use of the terms upper, lower, inner, outer, before, behind, anterior, posterior, and similar expressions in the technical descriptions of animals or their parts frequently leads to ambiguity. A great part of the confusion doubtless arises from the fact that the natural position of man differs from that of the lower animals in being erect. Thus, for example, when applied to men, *before* means in the direction indicated by a line drawn from the center of the body to the ventral surface ; in the lower animals it means in the direction indicated by a line drawn from the center of the body to the head. The same difficulty attends the use of the term anterior ; and of the opposite of these terms, behind and posterior.

Another source of confusion in the use of this class of terms is the fact that they are very commonly applied with reference to the plane of the horizon. Thus *above* means towards the zenith ; *below*, towards the nadir ; and *before* and *behind* indicate directions parallel to the plane of the horizon. Consequently whenever the position of an object is changed the terms denoting the relation of its parts must be changed.

In order to avoid these difficulties a special set of terms for designating the position and direction of the parts of animals has been adopted by many writers ; and it is the object of this chapter to define such of these terms as are used in this book.

Construction of the terms used.—Excepting the noun meson, the terms used in this work for denoting the position and direction of parts are either adjectives or adverbs.

The adjectives end in *al*, as dorsal, ventral, and mesal ; the adjectives cephalic and intermediate are exceptions to this rule.

The adverbs are formed by substituting for the adjective ending the ending *ad*, the Latin equivalent of the English suffix *ward*. Thus from the adjectives dorsal, ventral, and mesal, are formed the adverbs dorsad, ventrad, and mesad.

In forming compound words indicating position or direction, the vowel *o* is substituted for the termination of the first member of the compound, as dorso-ventral, caudo-cephalic.

The six cardinal directions.—There are six principal directions to which the position and direction of the parts of a bilaterally symmetrical animal, like an insect, are commonly referred ; these are as follows :

The *cephalic direction* or headward ; this is the direction indicated by a line drawn from the center of the animal to the head.

The *caudal direction* or tailward ; this is the opposite of the cephalic direction.

Two *lateral directions*, or towards the right and towards the left.

The *ventral direction* or bellyward ; this is the direction indicated by a line drawn from the center of the body to the ventral surface and forming a right angle with each of the preceding directions.

The *dorsal direction* or backward ; this is the opposite of the ventral direction.

The adverbial forms of the adjectives *cephalic, caudal, lateral, ventral* and *dorsal* are *cephalad, caudad, laterad, ventrad,* and *dorsad.* Thus a part which extends in a cephalic direction may be said to extend cephalad.

It should be carefully noted that cephalad does not necessarily mean towards the head but *headward;* that is, towards a point which is in a direction indicated by a line drawn from the center of the animal to the head, *but at an infinite distance in that direction.* In other words, these terms must be used in a way analogous to that in which we use right and left.

EXAMPLE.—Take a figure of a Dragon-fly with its wings extended as when at rest. Draw a line from the distal extremity of one of the wings to the head. Although this line extends directly towards the head it does not extend cephalad; but more or less nearly mesad.* A line extending cephalad from the distal extremity of a wing (or from any other part) is parallel to the cephalo-caudal axis of the body.

Differences between the technical and popular uses of cephalic and caudal.—It has just been shown that in the use of these terms it is not the head and tail to which the position and direction of parts are referred, but to two of the cardinal directions which are at right angles to right and left. Thus we can speak of the caudal part of the head or of the cephalic portion of the tail. It will be seen that this does not accord with the popular uses of these terms (as defined in the dictionaries) according to which no part of the body is cephalic except the head ; and of the different parts of the head one is just as much cephalic as another.

Oblique lines.—The position or direction of a part towards a point between two of the cardinal points can be designated by a compound term.

EXAMPLE.—A part which extends in a direction between those directions which are indicated by dextrad and caudad is said to extend *dextrocaudad.*

Meson, mesal, and mesad.—Frequently the position or direction of a part is referred to an imaginary plane di-

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* Mesad is defined later.

viding the body into approximately equal right and left halves. This middle plane is called the *meson* (μέσον, middle). From meson are derived the adjective *mesal* and the adverb *mesad*.

Ectal, ectad, ental, and entad.—It is often necessary, especially in the study of internal anatomy, to compare parts with relation to their nearness to or remoteness from the surface of the body. For this purpose the terms *ectal* (ἐκτός, without) and *ental* (ἐντός, within) are used. The adverbial forms of these terms are *ectad* and *entad*.

EXAMPLE.—The principal muscles of an insect are attached to the *ental* surface of the body-wall, or to parts of the body-wall which project *entad*. The hairs or spines on the body-wall project *ectad*.

Aspects of the body.—In describing animals it is often desirable to specify that part of the body which looks in a certain direction. For this purpose the term *aspect* is used combined with an adjective indicating the direction in which the surface in question looks.

EXAMPLES.—Dorsal aspect, ventral aspect.

Six aspects of the body are recognized ; these are dorsal, ventral, cephalic, caudal and the two lateral. The fact that the outlines of the body of an animal are more or less curved does not interfere with the practical application of the above terms.*

Proximal, distal, proximad, and distad.—In describing appendages of the body (legs, wings, etc.) the position of parts may be referred to the two ends of the appendage by use of the terms *proximal* and *distal*. *Proximal* indicates nearness to the end of the appendage which is attached to the body ; *distal*, to the end which is free. From these ad-

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* Cases occur where it is desirable to speak of an aspect which looks in a direction between two of the cardinal directions. Thus we speak of the lines or spots on the latero-dorsal aspect of a larva.

jectives the adverbs *proximad* (towards the proximal end) and *distad* (towards the distal end) are formed.

EXAMPLES.—The *proximal* segment of the leg of an insect is the coxa. The segments of the leg *distad* of the tibia constitute the tarsus.

Aspects of appendages.—In addition to the two ends of an appendage four aspects are recognized. To these the same terms are applied as to the corresponding aspects of the body : *viz.*, dorsal, ventral, cephalic, and caudal. It is therefore necessary to have a rule by which the correspondence between the aspects of the body and of appendages can be determined. In other words, a definite position must be chosen as the normal position of an appendage. Naturalists are quite well agreed as to what is the normal position of the limbs of the Vertebrates. The following are what we believe to be the analogous positions for the legs and wings of insects.*

(*a.*) *Wings.*—Extended horizontally at right angles to the body as are the wings of a dragon-fly (*Libellula*) when at rest.

(*b.*) *Legs.*—Extended horizontally at right angles to the body so that the convexity of the articulation between the two principal segments of the leg (femur and tibia) shall look dorsad ; and so that the surface of the tarsus (" foot ") which is usually applied to the ground when walking shall look ventrad.

The dorsal, ventral, cephalic, or caudal aspect of a wing or leg is that aspect which, when the wing or leg is in its normal position, looks in the same direction as does the aspect of the body which bears the same name.

First, second, third, etc.—When the members of a series of parts forming a portion or the whole of the trunk

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* The necessity for referring to the aspects of other appendages than the legs and wings will so seldom arise that it does not seem worth while to attempt to determine the normal positions of such appendages.

are indicated by the terms, first, second, third, **etc.,** the cephalic member of the series is the first.

EXAMPLE.—The *first* abdominal segment is the one nearest the head.

When the series forms a part or the whole of an appendage of the body, the first member of that series is the proximal one.

EXAMPLE.—The *first* segment of a leg is the one which is articulated to the body.

The direction of an appendage does *not* modify the above rule.

EXAMPLE.—The *first* segment of an antenna is the one which is articulated with the head; notwithstanding that when the antennæ are directed cephalad, as is usually the case, this segment is the one nearest the caudal end of the body.

Intermediate.—In order to avoid ambiguity the word mesal and its derivatives are used only with reference to the *meson.* The second member of a series of three similar parts is designated as the *intermediate* one.

Limitations to accuracy.—As the body of an animal presents but few plane surfaces or straight lines it is often impossible to describe the position or direction of a part with absolute accuracy. Practically, however, one will meet with but few serious difficulties. Thus in describing the direction of a curved or undulating line on the surface of the body it will rarely be necessary to do more than to give the general direction of that line ; the reader will understand that it follows the sinuosities of the surface of the body.

CHAPTER II.

THE EXTERNAL ANATOMY OF A LOCUST.

(*Melanoplus femur-rubrum.*)

Locusts or short-horned grasshoppers are excellent subjects to use in beginning the study of the external anatomy of insects. They are very common and are comparatively large ; and the parts of the external skeleton in these insects are mostly remarkably distinct.

The species which has been selected as the basis of this outline is the red-legged locust, *Melanoplus femur-rubrum,* which is found in nearly all parts of the United States. Specimens of this insect, preserved in alcohol, will be furnished the student, who will be expected to verify carefully or to correct each statement made in the text.

In order to illustrate certain points not well shown in the locust, comparative studies will be made of parts of a cockroach.

DIVISION OF THE BODY INTO REGIONS.

The body of a locust is composed of a series of more or less ring-like segments. In the caudal part of the body the ring-like nature of the segments is obvious ; in the cephalic part it is less so. These segments are grouped into three regions : *head, thorax*, and *abdomen.*

Head.—The head is the first or cephalic of the three regions of the body. Apparently it consists of a single segment.

Thorax.—The thorax is the second or intermediate region of the body. It is readily distinguished by its appendages; which are three pairs of legs and two pairs of wings. It consists of three segments; but as each segment is composed of several distinct pieces, it requires considerable study to trace the outlines of each segment. We will return to this subject later.

Abdomen.—The abdomen is the third or caudal region of the body. The segments of which it is composed are more simple, distinct, and ring-like than those of the other regions.

STRUCTURE OF THE BODY-WALL.

Chitin.—In studying the anatomy of insects it is found that in the adult stage the greater portion of the body-wall, that part of the insect which corresponds in position to the skin of higher animals, is hard.

This hardness is due to the deposition of a horny substance, called *chitin*, in the membrane which constitutes the body-wall.

Sclerites.—The chitin is not evenly distributed throughout this membrane. Pull the head of a locust so as to separate it from the thorax as far as possible without breaking the skin. Note that the head is joined to the thorax by a soft flexible membrane, in which but little, if any, chitin has been deposited.

Examine the sides of the thorax with a lens and observe that the body-wall appears to be made up of many distinct pieces. The integument is, however, really continuous; and in each case what appears as a distinct piece is simply a portion of the body-wall in which considerable chitin has been deposited. Such a portion of the body-wall is called a *sclerite.**

* The sclerites are analogous to the centers of ossification in the bones of the higher animals.

Sutures.—The sclerites constitute the greater part of the body-wall, the soft membranous portions separating them being in most cases narrow. Usually these narrow portions are mere lines; they are then called *sutures*.

Frequently the sutures become entirely effaced. We are therefore often unable to distinguish certain sclerites in one species of insect which are distinct in another.

In such cases the effaced suture is said to be *obsolete*.

PARTS OF THE HEAD.

The principal portion of the chitinized parts of the head are firmly joined together so as to constitute a box which contains what may be called by analogy the brain of the insect and certain other important organs. To this are articulated a number of movable appendages. The parts of the head may be classed, therefore, under two divisions : first, the fixed parts, or the skull ; second, the appendages.

THE FIXED PARTS OF THE HEAD.

Compound eyes.—The most striking in appearance of tne fixed parts of the head are the *eyes*. These are two large, nearly hemispherical bodies ; one on each side, forming a considerable portion of the latero-dorsal part of the head.

Study one of the eyes with a compound microscope, using a low power. Note the honey-comb-like structure of the eye. If you have difficulty in seeing this, remove a part of one eye with fine-pointed scissors and mount it on a glass slip. Each of the hexagonal divisions of the eye is a cornea of a distinct eye. These large eyes are therefore *compound*, and each of the small eyes of which they are composed is termed an *ommatidium* (plural *ommatidia*).*

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* Formerly the ommatidia were termed *ocelli* (singular *ocellus*) ; but later writers use the term ocelli only to designate the simple eyes.

Make a drawing showing the honey-comb-like structure of the cornea of a compound eye.

NOTE.—The drawings illustrating this course should be made with great care, on good paper. Outline drawings are better than those that are shaded, as shading tends to obscure lines indicating sutures. The drawings should be made first with a pencil, then, after they have been criticized, the lines should be inked.

Simple eyes.—Cephalad of the dorsal half of each compound eye there is a small transparent hemispherical body. These are the *simple eyes*. There is a third simple eye situated in a depression near the center of the cephalic aspect of the head. The simple eyes are termed *ocelli* (singular *ocellus*).

Epicranium.—The larger part of the skull appears to consist of a single sclerite which surrounds the compound eyes, and in the locust, bears the simple eyes. This part is termed the *epicranium*. The cephalic and lateral parts of the epicranium are separated on each side by a suture which extends ventrad from the eye. The ventral ends of these sutures are joined by a very prominent suture which forms the ventral boundary of the cephalic portion of the epicranium.

The epicranium is a compound sclerite, and differs in its extent in different insects.

Remove the head from the thorax and mount it on a slender pin, inserting the pin in the center of the cephalic aspect of the head. The pin will now serve as a handle.

Note the slightly elevated narrow ridge which separates the lateral from the caudal aspect of the head. This ridge marks the position of the suture which constitutes the caudal border of the epicranium. Upon the dorsal aspect of the head this suture is obsolete.

Upon each side joining the ventral end of the suture just described and the ventral end of the one which extends ventrad from the compound eye is a well-marked suture, which

forms the ventral border of the lateral part of the epicranium.

(*a.*) *Vertex.*—The dorsal part of the epicranium is called the *vertex.*

(*b.*) *Front.*—That part of the epicranium which is upon the cephalic aspect of the head is termed the *front.* In many insects the front is a distinct sclerite.

(*c.*) *Genæ.*— The lateral parts of the epicranium are known as the *genæ* or cheeks.

Clypeus.—Examine again the ventral border of that part of the epicranium which is upon the cephalic aspect of the head. Note that the prominent suture bounding this part separates it from a very broad, but short sclerite. This is the *clypeus.*

Labrum.—Articulated to the ventral border of the clypeus is a broad, freely movable flap. This is the upper lip or *labrum.*

Although the labrum is freely movable, it is a part of one of the segments that enter into the make-up of the head-box, and not an appendage, like the jaws. The true appendages of the head hold the same relation to this region that the legs do to the thorax.

Make a drawing of the cephalic aspect of the head ; and name the fixed parts.

Occiput.—Examine a locust's head which has been boiled in caustic potash, cleaned, and mounted with the caudal aspect uppermost. In such a preparation the soft parts have been removed, and the walls of the head bleached, so that the sutures can be more easily traced ; a specimen of this kind will be furnished the student.

Observe the large opening which connects the cavity of the head with that of the thorax. The dorsal half of this opening is bounded by the *occiput.* Each lateral half of the occiput is triangular, with its outline well marked ; but on the dorsal aspect of the head the suture between the occiput

and the epicranium is obsolete. Hence on this aspect there is no indication of the line where the occiput ends and the epicranium begins.

Postgenæ.—On each side ventrad of the occiput and caudad of the gena is a large sclerite. These form the chief portion of the caudal aspect of the fixed parts of the head and may be termed the *postgenæ*.

The postgenæ are separated from the epicranium by the narrow ridge which separates the lateral from the caudal aspect of the head ; a continuation of this ridge marks the position of the suture which separates the occiput from the epicranium on each side of the head.

In many insects the postgenæ and the occiput are not separate. As this is the case in the more generalized insects the occiput may be regarded as a detached portion of the postgenæ.

Tentorium.—Observe the remains of the membrane which connected the head with the thorax. It will be seen that the postgenæ are connected by a strong part extending from side to side, within the head. This is a part of the internal skeleton of the head or *tentorium*.

Make a drawing of the caudal aspect of the head, and name the parts.

Review.—The skull of a locust consists of six sclerites ; three of these, the *occiput* and the two *postgenæ*, pertain to the caudal aspect, one, the *epicranium*, constitutes the greater part of the dorsal, lateral, and cephalic aspects, and two, the *clypeus* and the *labrum*, form the ventral portion of the cephalic aspect. The epicranium consists of the *vertex*, the *front* and the *genæ*.

Cervical sclerites.—In the membrane connecting the prothorax with the head there is on each side a pair of sclerites forming a thickened line from the thorax to the head ; these are the *lateral cervical sclerites*. In some insects there are also ventral and dorsal cervical sclerites. The lateral

Labium.—Remove the head of a locust and pin it with the caudal aspect uppermost to a piece of cork.

Note the freely movable flap which is the caudal part of the mouth-parts. This and the crescent-shaped piece to which it is attached form the lower lip or *labium*.

The labium consists of the following parts :—

Submentum.—The submentum is the proximal part of the labium. It is nearly crescent-shaped, with long tapering points, and is joined to the membrane which connects the head with the thorax.

Mentum.—This is the central portion of the labium ; and is the principal part of that organ. It is articulated to the distal margin of the submentum. To the distal margin of the mentum are joined two movable flaps ; and to each lateral margin is joined a process consisting of three segments.

Labial Palpi.—These are the three-jointed processes of which one is joined to each lateral margin of the mentum.

Palpiger.—The labial palpi are not joined directly to the mentum. There is on each side of the mentum a sclerite which bears the palpus of that side and which is called the *palpiger.* The suture between the palpiger of each side and the mentum is almost obsolete. Its position is indicated by a slight groove which causes the palpiger to appear somewhat like a segment of the palpus.

Ligula.—This is the distal portion of the labium. It consists of two large movable flaps.

Hypopharynx.—If the specimen has become dry so as to be brittle, it should be softened with a little water.

With the specimen pinned as in last section, carefully lift the ligula so as to expose the maxillæ. Note the tongue-like organ which arises from the labium and from between the maxillæ. This is the *hypopharynx.*

Remove the labium and place it on a glass slip in a drop of Canada balsam or glycerine and cover it with a cover

glass. Examine it with a microscope using a low power and reflected, not transmitted, light (*i.e.*, turn the mirror so that the field of the microscope is dark ; and place the microscope so that a strong light falls upon the specimen). Make a drawing of the caudal aspect of the labium, and letter the parts.

Study the distal end of the distal segment of a labial palpus with a higher objective. Observe the sense papillæ with which it is furnished. Make a drawing of this part.

Parts of the maxillæ.—After the removal of the labium it is easy to distinguish the maxillæ, of which there is one on each side between the labium and the mandibles.

Remove a maxilla and mount it in Canada balsam or glycerine, with the caudal aspect uppermost. Examine with a microscope using a low objective, and reflected light.

Make a drawing of a maxilla, and name the parts, which are as follows :—

Cardo.—The cardo or hinge is the proximal part of the maxilla. It consists of two sclerites ; the first is the larger and is triangular in outline.

Stipes.—The stipes or footstalk is the large, quadrangular sclerite which forms the central part of the maxilla.

Lacinia.—Articulated to the distal end of the stipes is a large sclerite, which tapers distad, is curved, and is terminated by strong teeth ; this is known as the *lacinia ;* it is called also the *inner lobe.*

Galea.—Joined to the side of the stipes near its distal end and projecting laterad of the lacinia is a part consisting of two segments. This is the *galea.* The distal segment of the galea is large, spoon-shaped, and covers the inner lobe like a hood ; the proximal segment is constricted in the middle so as to resemble slightly a dumb-bell in outline. The galea is also known as the *outer lobe, upper lobe,* or *superior lobe.*

Palpifer.—Joined to the lateral border of the stipes and

between the cardo and the proximal segment of the galea is a narrow sclerite ; this is the *palpifer*.

Maxillary Palpus.—Articulated to the distal end of the palpifer is a long, slender organ consisting of five segments ; this is the *maxillary palpus*.

After completing the drawing of the maxilla as a whole, study the distal end of the distal segment of the maxillary palpus with a higher objective, and observe the sense papillæ.

Review.—The mouth-parts consist of an upper lip, *labrum ;* an under lip, *labium ;* two pairs of jaws acting laterally between these lips ; and a tongue-like organ, *hypopharynx.* The cephalic pair of jaws is called the *mandibles ;* the caudal pair, the *maxillæ.*

NOTE.—The natural attitude of the head of a locust is such that the *labrum* and *labium* appear to be fore and hind lips respectively; and the *mandibles* and *maxillæ*, fore and hind pairs of jaws. But when the mouth of an insect is in its more usual position, at the cephalic end of the body axis, the labrum is an upper lip, the labium an under lip, the mandibles, the upper jaws, and the maxillæ the lower jaws.

Each maxilla consists of six parts. These are the *cardo, stipes, lacinia, palpifer, galea,* and *maxillary palpus.*

The labium consists of the *submentum, mentum, ligula,* two *palpigers,* and two *labial palpi.*

THE HEAD OF A COCKROACH.

It is desirable to supplement the study of the head of a locust by an examination of the head of a more generalized insect ; for this purpose a cockroach may be used. If practicable one of the larger species should be studied.

Make a drawing of the ventral aspect of the head. Owing to the difference in attitude the ventral aspect of the head of a cockroach corresponds to the cephalic aspect of the head of a locust.

Epicranial suture.—Observe the inverted Y-shaped suture, the stem of which is on the middle line between the compound eyes, and the arms of which extend towards the antennæ ; this is the *epicranial* suture.

The stem of the epicranial suture divides the vertex into two sclerites. The arms of this suture separate the vertex from the front, which in this case is not a part of the epicranium as it is in the locust.

Antennal sclerites.—Surrounding the base of each antenna there is a more or less distinct, ring-like sclerite ; this is the *antennal sclerite.*

The antennal sclerites are much more distinct in the Plecoptera (stone-flies).

Clypeo-frontal suture.—The suture separating the clypeus and the front is obsolete in the cockroaches. Study this suture in the locust, and note the relation of it to the point of articulation of the mandible with the clypeus on each side ; and then indicate the probable position of it in the cockroach by a faint, dotted line in your drawing.

Letter the vertex, front, clypeus, labrum, and genæ.

Make a preparation showing the lateral aspect of the head and neck, and note the following features :—

The suture separating the gena from the postgena.

The absence of a suture separating the occiput from the postgena.

An opening into the head between the postgena and the neck near the point of attachment of the maxilla. This is the mouth of an invagination of the body-wall which forms a part of the tentorium ; *i.e.,* one of the *posterior arms of the tentorium.*

A narrow sclerite bounding the mouth of this invagination on the side towards the neck ; this is one of the lateral sclerites of the *maxillary* segment, the caudal one.

The maxillary segment is one of several segments that enter into the composition of the head, and the one of which the maxillæ are the appendages.

There are two sclerites on each side of the head in this segment, one on each side of the invagination, but the cephalic one is so reduced that it can be recognized only with difficulty.

The articulation of one of the lateral cervical sclerites with the maxillary segment.

Make a drawing of the lateral aspect of the head showing the features mentioned above.

THE MOUTH-PARTS OF THE COCKROACH.

In the cockroach the mouth-parts are very similar to those of the locust with an interesting and important difference in the labium. With the named drawings of the mouth-parts of the locust as guide, the parts of the mouth of the cockroach can be readily determined ; the following brief notes provide additional aid.

Labrum.—Attached to the margin of the clypeus as a movable cuticular flap, is the subquadrangular *labrum* or upper lip, serving as the dorsal or anterior covering of the mouth. Although the position and use of the labrum makes it one of the mouth-parts, it is morphologically different from the other mouth-parts in not being a modified appendage.

Epipharynx.—The *epipharynx* of all insects is a continuation or outgrowth of the upper membranous wall of the pharynx. It is, in the cockroach, as in most insects with biting mouth-parts, simply the ventral or internal membranous wall of the labrum.

A small distal portion of the labrum of the cockroach is rather plainly set off from the rest of the labrum by a transversal suture ; it may be that this distal part is a prolongation of the epipharynx alone (a condition observable among certain other insects). When the labrum and epipharynx are indistinguishably fused the single organ is sometimes called *labrum-epipharynx*.

Mandibles.—Below the labrum-epipharynx are the strongly chitinized prominent *mandibles*. Each mandible is a single sclerite, toothed on its distal or biting surface.

Maxillæ.—Lying next to and below the mandibles are the *maxillæ*. The *cardo* is small and elongate, subtriangular in shape and divided into two parts by a transverse line. The *stipes* is larger, and subquadrangular. Articulating with the stipes are the five-segmented *maxillary palpus*, and the two terminal lobes, the *galea* and the *lacinia*. The galea forms a sort of hood for the smaller triangular lacinia and is composed of two segments. The lacinia ends in a sharp, strong chitinous tooth. Indications, only, of the suture setting off the palpiger can be discovered in the maxilla of the cockroach.

Labium.—The labium is a single flap-like structure, which lies below the maxilla, and forms the floor of the mouth. It is, although an unpaired organ, really formed in all insects by the fusion of two organs similar in character to the maxillæ. The labium is sometimes called, because of this homology of structure, the second maxillæ.

In the cockroach the proximal or articulating sclerite, the *submentum*, is large and shows no indication of its paired origin. The succeeding sclerite, the *mentum*, is smaller and also is without indication of an original bipartite condition. Arising from the mentum, however, are the two four-segmented *labial palpi* and four terminal lobes ; two of these lobes and a palpus belong to each lateral half of the mentum and correspond to the lacinia, galea, and palpus, respectively, of the maxillæ. The inner lobes of the labium are called *glossæ* and the outer ones, *paraglossæ*. The glossæ and paraglossæ are fused so that only their distal portions are free and distinct. This fusion of the terminal lobes of the labium is carried farther among most other insects. Often the paraglossa and glossa of each side will fuse completely so

that there are apparently but two terminal lobes, and these distinct only distally, or all of the terminal lobes may fuse to form a simple flap-like sclerite, as among some of the Neuroptera. Because of this fusing of the bases or of all of the terminal lobes they are often spoken of collectively as the *ligula* (see p. 14. in account of external anatomy of the locust).

Hypopharynx.—The hypopharynx is a fleshy, blunt-pointed, tongue-like lobe, supported by a chitinized framework; this chitinous framework at the basal part of the hypopharynx projects backwards as four slender rods, two dorsal and two ventral, the ventral ones being curved and shorter than the upper. This chitinous framework is called the pharyngeal skeleton.

Make drawings of all the mouth-parts of the cockroach, naming the sclerites.

Cervical sclerites.—Study the neck of the cockroach and note that there are eight cervical sclerites, two dorsal, two ventral and two on each lateral aspect.

THE PARTS OF THE THORAX.

Division into segments.—Return to the study of the locust. The thorax consists of three segments. The cephalic or first segment is named the *prothorax ;* the second or intermediate, the *mesothorax ;* and the third or caudal, the *metathorax.*

These divisions of the thorax can be easily recognized by the appendages they bear. To the prothorax is articulated the first pair of legs ; to the mesothorax are joined the second pair of legs and the first pair of wings ; and to the metathorax, the third pair of legs and the second pair of wings.

PROTHORAX.

Dorsal part (pronotum).—That which may be properly termed the dorsal part of the prothorax is a large sunbonnet-shaped piece which covers the greater portion of the sides as well as the dorsal surface of this segment. This piece is called the *pronotum*.

It is believed that the dorsal part of each thoracic segment consists typically of four sclerites. These are named, beginning with the cephalic, *præscutum, scutum, scutellum*, and *postscutellum*. These sclerites may be distinguished in the dorsal parts of the mesothorax and metathorax (*mesonotum* and *metanotum*) of many insects; but the pronotum consists usually of a single piece. In the insect which we are studying, although the pronotum consists of a single piece, it is crossed by three well-marked sutures, indicating the division into four sclerites, which may be named as indicated above.

On the latero-dorsal aspect of the pronotum the suture between the præscutum and the scutum extends cephalad for a short distance and is then interrupted; the lateral portion of this suture is parallel with and quite near to the cephalic margin of the pronotum. Near the center of each lateral aspect of the pronotum there is a short oblique suture which separates the lateral fourth of the scutellum from the mesal part of that sclerite. None of the sutures extend to the lateral margin of the pronotum.

Make a drawing of the lateral aspect of the pronotum.

Ventral part.—The ventral part of each thoracic segment consists typically of two sclerites, one situated in front of the other; the first is the *sternum*, the second, which has never been named, may be termed the *sternellum*. The sternellum is frequently divided longitudinally into two parts, which may be widely separated.

Prosternum.—On the ventral surface of this segment between the legs there is a sclerite which bears a large tubercle ; this is the sternum of the prothorax or *prosternum.*

Prosternellum.—The sternellum of the prothorax, or the *prosternellum*, is a narrow, transverse sclerite, which is closely united to the following segment.

Lateral parts.—Owing to the great development of the pronotum, which covers the larger portion of the sides as well as the dorsal surface of the prothorax, the lateral parts of this segment are rudimentary. The following named sclerite, however, may be distinguished.

Episternum.—Between the pronotum and the end of the lateral prolongation of the prosternum, which extends on each side of the segment cephalad of the leg, is a conspicuous triangular sclerite ; this is the *episternum.*

Add the episternum to the drawing of the lateral aspect of the pronotum.

<center>MESOTHORAX AND METATHORAX.</center>

Union of these segments. — The second and third thoracic segments are firmly joined together, forming a box to which the two pair of wings and the second and third pairs of legs are joined. Owing to the intimate union of these two segments it will be easier to describe them together than separately.

With fine pointed scissors cut away the caudal border of the pronotum, that part which overlaps the mesonotum ; be careful not to break the membrane connecting the prothorax and mesothorax.

Dorsal part (mesonotum and metanotum). — The dorsal part of the mesothorax is termed the *mesonotum*, that of the metathorax, the *metanotum*. Unlike the pronotum these parts are confined to the dorsal aspect of the body. By cut-

ting away the caudal part of the pronotum as indicated above and spreading the wings laterad, these parts are exposed. Each consists of a nearly square area. To the lateral margins of the mesonotum is articulated the first pair of wings ; and to the lateral margins of the metanotum, the second pair of wings.

The sutures indicating the outlines of the sclerites of which the mesonotum and metanotum are composed, are not well defined ; there is consequently some difficulty in determining the limits of the sclerites ; and a drawing of this part will not be required.

In each of these segments only two of the four dorsal sclerites are well developed ; these are the *scutum* and the *scutellum*. The scutum occupies the cephalic half of the segment, the scutellum the caudal half. The scutellum consists of three parts : a central, shield-shaped part, which in this species is closely united to the scutum ; and on each side a part extending to the base of the wing. The caudal border of the scutellum is thickened and is connected on each side with the caudal border of the base of the wing by a cord-like structure.

NOTE.—In those insects where the præscutum and postscutellum are well developed, they usually extend entad and are often concealed within the thorax. The connection of the scutellum on each side with the caudal border of the base of the wing, is an excellent guide in tracing the homology of the parts of the mesonotum and the metanotum.

Ventral part.—On the ventral surface of the body between the legs of the second thoracic segment is a large sclerite ; this is the sternum and the sternellum of the mesothorax combined. The cephalic margin of this sclerite is nearly straight ; the caudal margin, deeply notched by a large, nearly square incision. The part in front of this notch is the sternum of the mesothorax or *mesosternum*, the two parts, one on each side of this notch, are the widely sep-

arated halves of the sternellum of the mesothorax or *meso-sternellum.*

Caudad of the mesosternum and mesosternellum there is a large sclerite, the mesal part of which is prolonged cephalad so as to accurately fit the notch in the caudal border of the meso-thorax ; this is the sternum of the metathorax or *metasternum.*

The two halves of the sternellum of the metathorax, or *metasternellum*, are widely separated, each being situated near the base of the corresponding leg.

The caudal border of the metathorax is also notched, and the first abdominal segment is dove-tailed into it.

Cephalad of the base of each mesothoracic and meta-thoracic leg there is a crescent-shaped sclerite ; this is the *antecoxal piece.*

Make a drawing of these parts.

Lateral parts (episterna, epimera, and peritremes). —Examine one side of the second and third thoracic seg-ments. Note that it is chiefly composed of four large scle-rites, which extend from the fossæ of the legs dorsocephalad. These sclerites are named as follows :—

Episternum of the mesothorax.—The first or cephalic of these four sclerites is the *episternum of the mesothorax.* The sutures between the episterna and the mesosternum are only faintly indicated in this species.

Epimeron of the mesothorax.—This is the second of this series of sclerites.

Episternum of the metathorax.—This is the third of this series of sclerites ; it is the one which bears the oblique yellow band characteristic of this species.

Epimeron of the metathorax.—This is the caudal member of this series of sclerites.

Spiracles and peritremes.—Between the ventro-caudal angle of the epimeron of the mesothorax and the fossa of the leg is an organ which consists of a slit-like opening guarded by

two fleshy lips; this is one of the openings of the respira-
tory system, these openings are called *spiracles*. When, as
in this case, a spiracle is surrounded by a circular sclerite,
such a sclerite is termed a *peritreme*.

In the membrane connecting the mesothorax with the pro-
thorax there is on each side a spiracle. This spiracle is
covered by the free margin of the pronotum. In this case
the peritreme is developed on the ventral side of the spiracle
into a prominent papilla.

Make a drawing of the lateral parts of the mesothorax and
metathorax.

Review.—The thorax consists of three segments, which
are named, beginning with the cephalic, *prothorax, mesothorax,*
and *metathorax.*

The body-wall of each of these segments is believed to
consist typically of *ten* sclerites. Of these, *four* pertain to the
dorsal part of the segment; *two* to each lateral part; and *two*
to the ventral part.

The dorsal sclerites are named, beginning with the ceph-
alic, *præscutum, scutum, scutellum,* and *postscutellum.*

Of the lateral sclerites, the cephalo-ventral one is the *epi-*
sternum, the caudo-dorsal one, the *epimeron*

The ventral sclerites are known as the *sternum* and the
sternellum. The sterna of the three thoracic segments are
designated as the *prosternum, mesosternum,* and *metasternum*
respectively; and the sternella as the *prosternellum, mesoster-*
nellum, and *metasternellum.*

In most insects the sternella are so reduced in size as to
be indistinguishable.

Sometimes there is also present near the base of the leg a
distinct *antecoxal piece*

The dorsal part of the body-wall of each segment is called
the *tergum.* This name is also applied to the dorsal part of
the three thoracic segments collectively.

The tergum of the prothorax is frequently called the *pro-notum ;* the tergum of the mesothorax, the *mesonotum ;* and that of the metathorax, the *metanotum.*

There are in this insect two pairs of thoracic spiracles.

APPENDAGES OF THE THORAX.

The appendages of the thorax are the legs and the wings ; the number and distribution of these have already been given.

Legs.—Examine the ventral aspect of the first pair of legs. Each leg will be found to consist of the following named parts :—

Coxa.—This is the proximal segment of the leg. It is sub-globular in outline. Examine the cephalic aspect of the coxa, and note the longitudinal suture which traverses this side of it ; this is shown better on the mesothoracic legs.

Trochanter.—This is the second segment of the leg, and is much smaller than the coxa. The ventral aspect of it is much longer than the dorsal.

Femur.—This is the third and principal segment of the leg.

Tibia.—This is the fourth segment of the leg. It nearly equals the femur in length, but is more slender.

Tarsus.—The tarsus includes all of that part of the leg distad of the tibia. It consists in locusts of three segments.

The last segment of the tarsus bears a pair of claws.

On the ventral surface of the tarsus there is a series of cushions , these are called *pulvilli.* The distal segment of the tarsus bears a single pulvillus which projects between the claws , this is often referred to in descriptive works as *the* pulvillus.

NOTE.—In the membrane connecting the coxa with the thorax just ceph-alad of the coxa, is a sclerite, this we believe to be the *trochantin.* The trochantin is a sclerite which is considered to be an appendage of the coxa, and its normal position is between the coxa and the antecoxal piece.

Make a drawing of one of the cephalic legs.

The same parts may be traced on each of the other legs.

Wings.—The wings are plate-like or membraneous expansions of the body-wall. Each wing is traversed by many linear thickened portions ; these are termed *veins* or *nerves*. The principal veins extend proximo-distad. These are joined together by many smaller cross-veins. The thin parts circumscribed by the veins and cross-veins are called *cells*.

The two pairs of wings of a locust differ remarkably in form and texture.

Mesothoracic wings (*tegmina*).—The mesothoracic wings are long, narrow, and of a parchment-like texture. They are termed *tegmina*.

Metathoracic wings.—The metathoracic wings are much larger and of more delicate texture than the first pair of wings. When not in use they are folded in plaits like a fan and concealed by the tegmina. Some writers who designate the mesothoracic wings as tegmina or wing-covers, describe the metathoracic wings simply as *the* wings.

PARTS OF THE ABDOMEN.

Number of segments—There is a difference of opinion as to the number of segments in the body of a locust. The difficulty arises from the complexity of the caudal end of this region, and the fact that some authors have considered the first abdominal sternum as a part of the metathorax. Eight abdominal segments can be readily distinguished in the female , and nine, in the male.* Caudad of the eighth abdominal segment of the female and the ninth of the male

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*If a sufficiently large series of specimens of the red-legged locusts be examined it will be seen that there are two kinds , one. in which the caudal part of the body tapers to the end, and bears four, pointed and curved, horny pieces ; and another, in which the caudal part of the body increases in size caudad and is terminated by a single, large, hood-shaped plate. The former is the female , the latter, the male

are a number of sclerites which are considered by some writers to be merely appendages of the abdomen ; other writers hold that certain of these sclerites represent sterna, and certain others, terga.

It is not within the scope of this work to enter into any discussion of the matter. We shall describe the parts as if there were eleven segments ; but wish the student to understand that the so-called eleventh segment may be merely an appendage of the tenth ; and that what is described here as the ninth and tenth segments have not been considered as such by certain very high authorities.

First abdominal segment.—The dorsal and ventral parts of the first abdominal segment are widely separated by the caudal part of the cavities for the insertion of the third pair of legs.

The ventral part of this segment is dove-tailed into the metasternum, and at first sight would be taken for a part of the thorax.

On each side in the dorso-lateral part of this segment, there is a large opening which is closed by a very delicate membrane ; these are the *auditory organs ;* the membrane is the *tympanum.*

Just cephalad of each auditory organ there is a small opening ; these are the first pair of abdominal spiracles.

Second to eighth abdominal segments.—Each of the abdominal segments, from the second to the eighth inclusive, is ring-like in form, and without appendages.

In each of these segments the lateral margins of the tergum join the pleura * without any suture. Near the cepha-

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* The lateral part of a segment is termed the *pleurum ;* in the same way that the ventral part is called the *sternum ;* and the dorsal part, the *tergum.* By some writers the entire dorsal part of an insect is called the tergum , the lateral part, the pleurum ; and the ventral part, the sternum. These writers apply the terms *tergite, pleurite,* and *sternite* respectively, to the dorsal, lateral, and sternal regions of each segment.

lo-ventral angle of each pleurum there is a spiracle. The sterna are well developed and are separated from the pleura by a narrow, involuted, membraneous part.

Caudal part of abdomen of female.—The most prominent portion of the caudal part of the abdomen of the female is the *ovipositor*. This is an organ consisting of four, strong, curved, and pointed pieces, which form the most caudal part of the body. With this organ the insect makes a hole in the ground in which she lays her eggs. This is done by alternately bringing together and separating the two pairs of pieces, and at the same time pushing the body into the ground. Examine carefully these pieces, and note how well they are adapted to this purpose.

Between the ventral pieces is the *opening of the oviduct*.

Ventrad of this opening and also between the ventral pieces of the ovipositor is a pointed prolongation of the eighth abdominal sternum ; this has been termed the *egg-guide*. Dorsad of the egg-guide there is a forked organ which also is used in placing the eggs.

The ventral pieces of the ovipositor are supported by two pairs of sclerites ; there being a sclerite closely applied to the ventral surface of each pair, and one to the lateral surface of each.

The ninth and tenth abdominal terga are shorter than any of the preceding abdominal terga and are joined together on each side, the lateral parts of the suture separating them being obsolete.

Caudad of the tenth tergum there is on the middle of the back a shield-shaped piece ; this is believed to represent an eleventh segment. It consists of two sclerites, as is indicated by a transverse suture.

On each side, projecting from beneath the caudal border of the tenth tergum, is a pointed appendage · these are the *cerci*.

Each cercus partially covers a much larger, triangular sclerite which extends from the lateral border of the tenth tergum to the caudal apex of the eleventh tergum ; these are the *podical plates*.

By lifting the free end of the eleventh segment, the caudal opening of the alimentary canal, the *anus*, is exposed ; it is situated between the podical plates.

Caudad of the podical plates is the dorsal pair of pieces of the ovipositor.

Make drawings of the dorsal and the lateral aspects of this part of the body ; in the drawing of the lateral aspect represent the entire abdomen.

Caudal part of the abdomen of the male.—In the male ten abdominal sterna are present ; the tenth is a hood-shaped sclerite on the caudal aspect of the body.

As in the female, the ninth and tenth abdominal terga are united on their lateral margins.

Projecting from the caudal margin of the tenth tergum there is on the middle of the back a forked appendage, the *furcula*.

The eleventh tergum is furrowed by three deep longitudinal grooves.

The *cerci* are situated as in the female, but are longer.

The podical plates are nearly as in the female.

Make drawings of the dorsal and the lateral aspects of this part of the body.

CHAPTER III.

THE INTERNAL ANATOMY OF AN INSECT.

Corydalis cornuta.

The larva of *Corydalis cornuta* lives under stones in the bed of swiftly flowing streams ; it is well known to sportsmen under the name of " dobson," and is used extensively as bait for black bass.

As the larval state of *Corydalis* lasts nearly three years, larvæ which are at least two years old may be found at any time. They are most abundant where the water flows swiftest. A good way to obtain them is to hold a dip-net or a wire screen in the stream below some stones, and, lifting the stones with a hoe or garden rake, cause the current to sweep into the net the insects which were under the stones.

This larva is probably as desirable a subject for an elementary study of the internal anatomy of insects as can be found in this country. The species is a large one, being one of our largest insects ; there is a coarseness in its structure, which enables one to study the different viscera with comparative ease ; it is furnished with well-developed organs for aquatic respiration, and at the same time with equally well-developed organs for aërial respiration; and, as already stated, fresh specimens can be easily obtained at any season, even in mid-winter.

Unfortunately, however, the appearance of the insect is very disagreeable to most people. But after a specimen has been opened and pinned upon cork, as is necessary in the study of the viscera, the disagreeable features are not visible ;

and the ease with which the internal organs can be examined
more than counterbalances the unpleasant part of the prepara‑
tion of specimens.

PRESERVATION OF SPECIMENS.

Specimens that are to be used for the study of internal
anatomy should be preserved in an aqueous solution of chlo‑
ral hydrate ; this is made by dissolving one part by weight of
chloral hydrate in twenty parts of water. This liquid pre‑
serves the organs and at the same time leaves them flexible.
After the specimens have been in the solution for one day, a
short, longitudinal slit must be made through the wall of the
abdomen, so as to allow the solution to enter the perivisceral
cavity ; otherwise the viscera will decay. The delay of one
day before making the slit in the body is important ; if the
slit is made too soon, the muscles will contract in such a way
as to distort the specimen and render it worthless. One-half
of the specimens should be slit on the back, the others, on
the ventral side ; for if the specimens are all cut in the same
manner, it will be impracticable to study certain organs.

If a very careful study is to be made of the external anat‑
omy of this larva, one or more specimens should be left for
several days in a warm place, in the chloral hydrate solution,
without being cut. The parts bathed by the solution will be
well-preserved ; the viscera will decay ; and the gases of de‑
composition will so distend the body that the different scle‑
rites will be spread apart.

EXTERNAL CHARACTERISTICS.

In order to understand the internal anatomy of this insect
it is necessary to know the more general features of its ex‑
ternal anatomy. We will not stop to trace out the homol‑
ogies of the different sclerites which enter into the compo‑

sition of the body-wall, but will merely examine the more important external structures.

Pin a larva to the cork or beeswax lining of a dissecting dish, with its ventral aspect uppermost, and cover it with water. Make a drawing of the ventral surface. Name the regions and the appendages of the body shown in this view. The long, tapering appendages on the margins of the abdomen may be termed the *lateral filaments ;* the tufts of hair—like appendages near the bases of the lateral filaments are *tracheal gills ;* and caudad of the ninth abdominal segment is a pair of *prolegs.* These may be termed the *anal prolegs.**

Make a drawing of each of the following parts :—

1. The dorsal aspect of the head, showing the mandibles, the antennæ, and the labrum.

2. One of the tracheal gills. Cut off several of the hair-like branches and mount them in glycerine, using a thin cover-glass. Examine them with a compound microscope, using a high power. The surface of each hair is marked with numerous ridges, which resemble in appearance the fine ridges on the skin of the palm of the human hand. In the center of each hair, and extending nearly its entire length, is a large tube. This is a trachea or air vessel. Tracheæ can be distinguished from other vessels by being marked with transverse lines, which occur at regular and very short intervals. The intimate structure of the tracheæ will be studied later ; but at this point the student should become familiar with the appearance of tracheæ, so that when he dissects the insect he can readily distinguish them from other vessels. Branching from this large central trachea are numerous very

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* Many larvæ bear upon the abdomen locomotive appendages, which resemble legs, and are termed prolegs. This is especially true of caterpillars, which bear from one to five pairs of these appendages. The prolegs are temporary organs, being shed with the skin when the larva transforms to a pupa.

small tracheæ. Carefully trace out the courses of the small tracheæ and represent them in your drawing.

3. A spiracle.

Preliminary work.—Take from the chloral hydrate solution a specimen which was slit on the ventral surface. Immerse the insect in water, with its dorsal surface downwards. With fine scissors extend the slit the whole length of the thorax and abdomen ; in making this slit cut through the wall of the body into the perivisceral cavity ; the body-wall consists of two parts, the external crust of the insect, and, entad of this a wall of muscles; it requires considerable care to cut into the perivisceral cavity and not injure the viscera. Make, on each side, in that part of the body-wall connecting the prothorax with the head, an incision extending from the longitudinal incision to the side of the body.

On the meson just entad of the ventral wall of the body, there are two white cords, extending nearly the whole length of the body. At intervals, which approximate the segments in length, these cords are united ; at the points of union they are greatly enlarged ; from these enlargements there arise numerous, small, white cords, which extend in various directions. The two longitudinal cords, the enlargements upon them, and the numerous cords branching from these enlargements constitute the *nervous system ;* the cords are *nerves* and the enlargements are *ganglia.* The nervous system will be studied later in a specimen which has been opened on the dorsal side.

Cut away from the ganglia the nerves that extend to one side ; do this with the scissors, first placing one blade under the nerves and lifting them away from the other viscera.

Take a strip of sheet-cork a little longer than the insect

and twice as wide, and pin it to the beeswax lining of the dissecting dish, and cover it deeply with water. Place the specimen on the cork and fasten with a pin at each end. Turn laterad each half of the ventral wall and pin it down to the cork, using ribbon-pins.

NOTE.—At the close of the day's work on this subject the student should remove the strip of cork from the dissecting dish and place it with the specimen still spread out upon it in a wide-mouthed bottle of chloral hydrate solution. By doing this the work can be resumed without the necessity of making a new dissection.

Ramifying through all parts of the body are numerous *tracheæ ;* the larger tracheæ are of a dusky color ; but many of the smaller ones contain air, which renders them silvery white. On each side of the body, extending the entire length of the thorax, are two very large tracheæ ; from each side of each abdominal segment except the last there arises a large trachea, which divides and subdivides into numerous branches. Cut a short piece from one of the large abdominal tracheæ, examine it with a compound microscope, and note its characteristic appearance, so as to be able to distinguish tracheæ.

In the dissection of this specimen, the student may cut tracheæ and nerves freely ; but great care should be used not to cut other vessels unless specially directed to do so.

In the center of the perivisceral cavity and extending the whole length of the body, there is a large tube ; this is the *alimentary canal.*

Adipose tissue.—Surrounding the caudal half of the alimentary canal and attached to the lateral and dorsal walls of the abdomen and thorax, there are large, flocculent masses of a white substance ; this is the *adipose tissue* or *fat.*

Examine a preparation of adipose tissue, with a compound microscope, using a high power, and make a drawing showing the minute structure of the tissue.

In the farther dissection of this specimen the adipose tissue may be cut away when necessary to see the parts studied.

Form of the alimentary canal.—Remove the ventral wall of the head so as to expose the alimentary canal throughout its entire length.

Make a drawing of the alimentary canal and label the following parts :—

Pharynx.—The somewhat trumpet-shaped part of the alimentary canal immediately caudad of the mouth is the *pharynx*.

Œsophagus.—That part of the alimentary canal which is immediately caudad of the pharynx and which traverses the caudal part of the head and the cephalic part of the thorax is the *œsophagus*. It is a straight tube of nearly uniform diameter except when some portion of it is distended by food.

Proventriculus.—In the caudal part of the thorax the alimentary canal begins to enlarge. This enlargement increases gradually caudad until, in the first or second abdominal segment, its diameter is twice that of the œsophagus ; then it contracts quite suddenly until its diameter is less than that of the œsophagus ; this enlarged portion is the *proventriculus*. It corresponds in function with the gizzard of birds and is very complicated in structure internally.

Ventriculus.—Caudad of the constriction following the proventriculus, there is a slightly enlarged portion, from the cephalic end of which there project cephalad, four large pouches ; this enlargement is the *ventriculus* or *stomach* and the pouches are the *gastric cæca*. The two cæca of each side are quite closely united.

Malpighian vessels.—Emptying into the caudal end of the ventriculus are several, small, very long, and much convoluted tubes ; these are the *Malpighian vessels ;* they were

named in honor of Malpighi, one of the early anatomists. As uric acid is found in the Malpighian vessels, they are supposed to correspond to the kidneys in function.

Determine the number of Malpighian vessels.

Intestine.—The part of the alimentary canal extending from the ventriculus to the caudal end of the body is the *intestine ;* the part immediately caudad of the ventriculus is the *small intestine ;* following the small intestine is the *large intestine ;* there are two bends in the cephalic part of the large intestine ; the first extends dorsad and cephalad ; the second, dorsad and caudad ; the *rectum* is not a well-defined part of the intestine in this insect.

Attachments of the alimentary canal. — The alimentary canal is attached to the body-wall and thus held in place in various ways. The most obvious attachments are those of the ends of this organ. In addition to these direct connections, the alimentary canal is indirectly connected to the body-wall as follows : —

By tracheæ.—From the lateral wall of each abdominal segment, large tracheæ arise ; many of the minute branches of some of these extend to the walls of the alimentary canal and thus tend to hold it in place.

In connection with the tracheæ, the action of the masses of adipose tissue should be observed. These large masses, which to a great extent are held in place by the tracheæ that extend to the alimentary canal, serve as cushions which tend to keep the organ in place.

By muscles.—A large number of very delicate muscles extend from the ventral wall of the head to the œsophagus. In the specimen which the student is now studying, only the ends of these muscles which are attached to the œsophagus can be observed, as the attachments of these muscles to the wall of the head were cut away in the preparation of the specimen. Large muscles extend caudo-ventrad to the intestine from the line on the dorsal wall of the body between the eighth and ninth abdominal segments. From within the anal prolegs, muscles extend cephalad into the ninth abdominal segment and are attached to the intestine. Other muscles are described in the next section.

By the suspensoria of the viscera.—There are several, long, fine threads that are so attached as to tend to hold the alimentary canal and other viscera in place. These may be termed collectively the *suspensoria of the viscera.*

In *Corydalis* four pairs of suspensoria can be distinguished. There are two pairs of suspensory muscles, a pair of ligaments, and a pair of suspensory nerves.

The suspensory muscles arise from the body-wall in the thorax, and extend caudad into the abdomen, where both pairs are attached to the alimentary canal, and one pair to other viscera also. It is rather difficult to trace out the origins of these threads upon the body-wall; but the threads can be easily seen extending parallel with the œsophagus and proventriculus, after they emerge from the layer of muscles and fat. They can be rendered more easily seen by staining the specimen with hæmatoxylin. Empty the water from the dish containing the specimen, and place a few drops of hæmatoxylin on it; after one or two minutes wash off the stain, and cover the specimen with clean water.

Study first the suspensoria on one side of the specimen, leaving those of the other side for study when the final drawing is made. In the following notes a single member of each pair of suspensoria is described.

The two suspensory muscles may be designated as the simple suspensory muscle and the branched suspensory muscle, respectively.

The *simple suspensory muscle* arises from near the middle of the ental surface of the pronotum, and extends caudad to the gastric cæca, where the fibers of which it is composed spread apart, some going to one cæcum and some to the other.

Make a provisional sketch of this.

With fine-pointed scissors, cut off the tips of the two gastric cæca of this side, and remove them with as long a piece of the suspensory muscle as is practicable, and mount them in glycerine for study with a high power of the microscope. Note the transversely striated appearance of the fibers of this suspensorium. This indicates that it is composed of striated muscular tissue.

Make a careful drawing showing the minute structure of this suspensorium.

The *branched suspensory muscle* arises from the ental surface of the body-wall, on the dorsal side, between the mesothoracic and metathoracic shields, near the lateral margin of the body and extends caudad into the cavity of the abdomen; here it divides into several branches. One branch extends to the ventriculus; one or more to the masses of fat and to the Malpighian vessels; and one joins a suspensorium which extends from a large trachea in the third abdominal segment to the intestine.

Trace out the course of the branches of the branched suspensory muscle, and make a provisional sketch showing their connections.

The *ligament of the viscera* is not attached to the body-wall, but is supported by a large trachea in the third abdominal segment, about which it forms a collar. This suspensorium has three branches; one of these extends caudad to the testis or ovary; one, cephalad, to the heart; and the third, to the intestine. This last branch receives the tendon of one of the branches of the branched suspensory muscle.

Make a drawing representing the alimentary canal in the center, a testis or ovary on each side (these are described in the next section), and the three pairs of suspensoria. While doing this the provisional sketches already made can be utilized, but the observations should be confirmed by a study of the suspensoria of the other side of the specimen.

Cut the trachea supporting the ligament of the viscera, and slip the ligament off from it; cut the ligament extending to the heart as near to the heart as practicable; cut the ligament extending to the intestines between the intestine and the tendon of the branched suspensory muscle; cut the branched suspensory muscle as far cephalad as practicable; cut off the tip of the testis or ovary, leaving it attached to the ligament; mount the preparation thus made in glycerine for study with the microscope, carefully spreading apart the branches of the ligament and the end of the suspensory muscle with a needle before putting on the cover-glass. Study this preparation with a high power of the microscope and note the difference in structure between the suspensory muscle and the ligament of the viscera. Make a drawing showing this.

The *suspensory nerves of the alimentary canal* extend from the small intestine to the last abdominal ganglion. They can be best seen in a specimen opened on the back and will be described later.

The masticatory organs of the proventriculus.—

Remove the alimentary canal, and carefully open the proventriculus by a longitudinal slit. Cover a piece of cork with white paper, spread out the opened proventriculus upon the paper, ental side uppermost, and fasten it in place with ribbon-pins. Fasten the cork bearing the preparation to the beeswax in a dissecting dish, and cover the preparation with water. Study it with a lens and with a compound microscope, using a low power. Write a description of the masticatory organs, and make a drawing illustrating them.

The reproductive organs.—Although there appears to be no external characteristic by which the sexes of the larva of *Corydalis* can be distinguished, the internal reproductive organs differ greatly in appearance ; the testes in this species are long and narrow, while the ovaries are short and broad. These organs are situated one on each side, between the layer of adipose tissue which surrounds the alimentary canal and the layer of the same tissue which is attached to the sides of the wall of the abdomen. The testes extend from near the middle of the third abdominal segment to near the posterior border of the sixth abdominal segment. The ovaries extend from near the middle of the fourth abdominal segment to near the middle of the fifth abdominal segment.

Determine the sex of the specimen that you are studying.

Reproductive Organs of the Male.—Note the shape of a testis, the connection of it with the respiratory system, the groove in the middle of its mesal aspect, and the vessel in the bottom of this groove. This vessel is the seminal duct or *vas deferens* (plural, *vasa deferentia*) and is the outlet of the testis ; it extends caudad to near the caudal end of the body. The cephalic end of the testis is supported by the ligament of the viscera already described.

Make drawings showing the external form of a testis.

Trace out the course of the vas deferens. This will require very careful dissection, especially in the ninth abdominal segment, where the vas deferens passes under a muscle.

The triangular organ into which the two vasa deferentia empty is the seminal vesicle. If the specimen has been opened on the ventral side, the seminal vesicle will lie on the alimentary canal, but if it has been opened on the dorsal side it will be necessary to cut the intestine and turn back the caudal part of it in order to see the seminal vesicle.

The tube extending from the seminal vesicle to the external opening of the reproductive organs is the *ejaculatory duct*.

Make a drawing showing the relations of the testes, vasa deferentia, seminal vesicle, and ejaculatory duct.

Each testis is composed of many, short, pear-shaped tubes, the *testicular follicles*, in which the spermatozoa are developed. These tubes open into

the vas deferens, which, as already described, extends along the groove in the testis. The testicular follicles, however, are enclosed in a thick coat of connective tissue which conceals them.

Reproductive Organs of the Female.—Note the shape of an ovary, the connection of it with the respiratory system, the position of the *oviduct* (which is the vessel extending caudad from the ovary), and the attachment of the ligament of the viscera.

Make a drawing showing these things.

Each oviduct extends caudad from the caudal end of the ovary through masses of fat to the seventh abdominal segment, where, at a point opposite the last ganglion of the nervous system, it passes under the ventral muscles, and terminates in a disc-like enlargement, on the body-wall.

Make a dissection showing this.

NOTE.—In the adult the two oviducts extend caudad to near the caudal end of the body where they empty into a short *vagina*.

Each ovary is composed of many parallel tubes, the *egg-tubes*, in which the eggs are developed, and which open into the oviduct. But as the egg-tubes are enclosed in a thick coat of connective tissue, which envelopes the entire ovary, it is necessary to break this coat in order to see them.

Remove an ovary, place it on a glass slip, tease it apart with needles, and examine it with a microscope.

Make a drawing of an egg-tube.

The reproductive organs of a locust.—As the reproductive organs of the larva of *Corydalis* are in an immature condition, it will be well for the student to study, at this point in his work, the reproductive organs of an adult insect. For this purpose the red-legged locust may be used. If possible use fresh specimens.

The reproductive organs of the male.—Remove the wings and legs from an adult male locust, make a slit in the dorsal wall of the body, extending the whole length of the thorax and abdomen, spread open the specimen, pin it to a piece of cork, and place it under water for dissection.

The *testes* lie upon the stomach, and are enclosed in a common sack-like envelope. This is exceptional; in most in-

sects the two testes are separate, each lying in one side of the body-cavity.

The cephalic end of the united testes is supported by the *ligament of the viscera*, which is a very slender cord, extending to the dorsal wall of the body.

Break the envelope enclosing the testes and note that they are composed of many, long, slender tubes; these are the *testicular follicles*.

Separate the two sets of follicles and observe that those of each side empty into a slender duct; this is the *vas deferens*. The two vasa deferentia pass caudo-ventrad, one on each side of the alimentary canal.

Cut in two the alimentary canal between the testes and remove the caudal portion of it.

Observe the two bundles of tubes which occupy a considerable part of the cavity of the abdomen; these are the *seminal vesicles* and the *accessory glands*. There is on each side a single, long, much convoluted, accessory gland, and a bundle of shorter, tubular seminal vesicles. The seminal vesicles are reservoirs for the products of the reproductive organs; the function of the accessory glands has not been determined, they may secrete the more fluid part of the semen.

Trace out the course of the vasa deferentia, and note that each is joined by the seminal vesicles, and the accessory gland of the same side, and that almost immediately the tubes of the two sides unite, forming a single tube; this is the *ejaculatory duct*.

The ejaculatory duct leads to the *penis ;* owing to the muscles surrounding this organ the determination of its form is quite difficult, and will be omitted in this elementary course.

If fresh specimens are available, kill one in a cyanide bottle, and by pressure of the abdomen force out the caudal end of this organ, and observe the chitinous hooks with which it is armed.

Dissect another specimen, opening it on the ventral side.

Make a diagram representing the relations of the various parts of the reproductive organs of the male locust.

If fresh specimens can be obtained, kill one, remove a seminal vesicle from it, tease apart the vesicle on a glass slip, mount it in glycerine, examine it with a high objective, and observe the bundles of *spermatozoa*. Each bundle consists of a great number of hair-like spermatozoa.

The reproductive organs of the female. — Take an adult female locust, remove the wings and hind legs, and make a slit in the dorsal wall of the body extending the whole length of the thorax and abdomen. Spread open the specimen, pin it to a piece of cork, and place it under water for dissection.

The *ovaries* lie above and on each side of the stomach : the cephalic end of them is supported by the *ligament of the viscera*. The *egg-tubes* of which the ovaries are composed occupy the same relative position as the testicular follicles of the male, but fill a larger proportion of the body-cavity.

Push laterad the egg-tubes of each side so as to expose the alimentary canal ; cut in two the alimentary canal in the thorax, and remove the caudal portion of it ; this will expose two large bundles of muscles connected with the dorsal pair of valves of the ovipositor. Carefully split these apart and fasten them aside with pins. When this is done, the last ganglion of the nervous system will be exposed. Carefully remove this ganglion, cutting such nerves and tracheæ as may be necessary.

Each ovary consists of many egg-tubes which are arranged in two rows, and open into an *oviduct ;* the two oviducts unite near the caudal end of the body and form the common oviduct or *vagina*.

That portion of each oviduct into which the egg-tubes empty is greatly enlarged ; this is termed the *egg-calyx*, and serves as a receptacle for the ripe eggs.

There is a prolongation of each oviduct which extends cephalad of the ovary ; this, we infer is a *colleterial gland.* The colleterial glands are organs which secrete the cement for gluing together the eggs ; the more usual form of them is that of separate glands, like the accessory glands of the male, which open into the vagina, as the accessory glands open into the ejaculatory duct.

Lying dorsad of the vagina, there is a small elongate body, from which there extends a much convoluted tube ; these are the *spermatheca* and its duct.

On each side of the vagina, near the caudal end of the body there is a rounded pouch, which in fresh specimens is of a reddish color. From the cephalic end of each pouch a muscle passes cephalad along the dorsal wall of the vagina, and between the two oviducts, and is attached to the ventral wall of the body. These pouches open below the ventral valves of the ovipositor, one on each side of the opening of the vagina, but a little farther dorsad than that opening. The function of these organs has not been determined ; they are described here on account of their intimate relations with the reproductive organs. They are doubtless eversible glands, as in fresh specimens they can be everted by pressure ; and the muscles are doubtless for withdrawing them after eversion.

If living specimens are available, kill a female, by putting it in a cyanide bottle, and then by pressure of the abdomen evert these glands. Note that they open separately between the last sternum of the abdomen and the ventral valves of the ovipositor.

Take a fresh specimen, or a preserved one, if fresh specimens are not to be had, and spread apart the last sternum of the abdomen and the ovipositor, and observe the elliptical opening between the ventral valves of the ovipositor. This opening is surrounded by narrow bands of chitin, and is

the *opening of the bursa copulatrix.* It is through this opening that the seminal fluid is received.

The *opening of the vagina* is distinct from that of the bursa copulatrix, and ventrad of it. It is on the dorsal aspect of the lobe of the body supported by the last abdominal sternum.

Return now to the study of your dissection.

Trace out the course of the duct leading from the spermatheca and observe that it empties into a pouch ; this is the *bursa copulatrix*, the external opening of which has been examined already.

Trace out the course of the vagina and observe that its opening is distinct from that of the bursa copulatrix, as has been pointed out.

When the eggs are laid they are pushed dorsad past the opening of the bursa copulatrix, in this way spermatozoa coming from the spermatheca can reach them. The egg-shell of insects is perforated at one end by one or more openings, the *micropyle* through which a spermatozoon passes to fertilize the egg.

Dissect another specimen, opening it on the ventral side.

Make a diagram representing the relations of the parts of the reproductive organs of a female locust.

Review.—The following tabular statement will aid the student in making a review of his work on the reproductive organs of a locust, and will serve to indicate the correspondence of the parts in the two sexes. He should bear in mind, however, that the reproductive organs of different insects vary greatly in structure. In most insects each testis or ovary consists of many parallel tubes, as in the locust ; but the structure and position of the reservoirs of the products of these organs and the structure of the accessory glands differ greatly in different insects.

REPRODUCTIVE ORGANS OF A LOCUST.

I. MALE.	II. FEMALE.
1. Two *testes*, each testis consisting of many *testicular follicles.*	1. Two *ovaries*, each ovary consisting of many *egg-tubes.*
2. Two *vasa deferentia.*	2. Two *oviducts.*
3. Several *seminal vesicles*, each an appendage of a vas deferens.	3. Two *egg-calyces*, each calyx an enlarged portion of an oviduct.
4. Two *accessory glands*, each an appendage of a vas deferens.	4. Two *colleterial glands*, each a cephalic prolongation of an oviduct.
5. An *ejaculatory duct*, the united vasa deferentia.	5. A *vagina*, the united oviducts.
6. A *penis.*	6. An *ovipositor.*
	7. A *bursa copulatrix* and a *spermatheca* with a *duct* connecting them.

The respiratory system. — Return to the study of *Corydalis*. Make a diagram showing the arrangement of the larger tracheæ.

The walls of the tracheæ are composed of three layers, which correspond to the layers of the body-wall ; in fact the tracheæ are invaginations of the body-wall. The continuity of the membranes of the tracheæ and body-wall is shown diagrammatically in Figure 1. It should be observed that it is the inner layer of the wall of the tracheæ that corresponds with the outer layer of the wall of the body.

FIG. 1.—Section of trachea and body-wall. *c*, cuticle ; *hy*, hypodermis ; *sp*, spiral thickening of the intima. The third layer is a delicate membrane bounding the ental ends of the hypodermal cells ; this is the basement membrane.

This inner layer of the wall of the trachea, the *intima*, like the cuticle, is chitinous, and is shed from the tracheæ with the cuticle when the insect molts. This layer of the trachea is furnished with thickenings, which

extend spirally and give to tracheæ their characteristic, transversely striated appearance. If a piece of one of the larger tracheæ be pulled apart the intima will tear between the folds of the spiral thickening, and the latter will uncoil from within the trachea like a thread. These spiral thickenings are termed *tænidia* (Greek, little bands). In some insects there are several, parallel thickenings of the intima, so that, when an attempt is made to uncoil the tænidia, a ribbon-like band is produced,which is composed of several parallel tænidia.

Make a preparation and a drawing illustrating the appearance of a trachea of *Corydalis*, with the tænidia partially uncoiled.

The heart and the aorta.—Read the account of the blood-vessels in *Comstock's Manual for the Study of Insects*, pp. 71, 72.

In *Corydalis* the dorsal blood-vessel extends from the eighth abdominal segment to the brain. It is a slender tube, widest in the region of the fourth and fifth abdominal segments. That part of it which lies in the abdomen is the *heart ;* the more slender portion which traverses the thorax and extends into the head is the *aorta*.

The cephalic end of the aorta is attached to the brain, and is trumpet-shaped except that the ventral wall of it is split. In this way a V-shaped opening is formed, through which the blood flows out, into the cavity of the head.

Make a drawing representing the outline of the heart and the aorta.

The valves of the heart and of the aorta.—Usually it is very difficult to determine the position and form of the valves in the dorsal blood-vessel in specimens preserved in chloral hydrate solution ; for a study of these valves specimens which have been prepared especially for this purpose are necessary. On this account this part of the subject may be omitted by students taking a brief course in entomology. For the sake of others the following method of study is indicated :

With a hypodermic syringe inject into the body-cavity of a living larva a solution of lampblack or of carmine.* After about an hour, drop the larva into boiling water and leave it there till killed, which will require from ten to twenty seconds ; then transfer it to cold water. In specimens prepared in this way the heart will be found filled with coagulated blood, which is rendered more easily seen by the presence in it of the lampblack or carmine. The relation of the valves to the segments of the body can be best determined in specimens from which the dorsal body-wall has been removed from over the heart, the specimens being studied from the dorsal aspect.

In *Corydalis* the heart contains eight sets of valves, and the aorta two.

Make figures showing the position and form of the valves of the heart and of the aorta.

The pericardial diaphragm or the "wings of the heart."—Take the specimen which was prepared so as to show the heart and the aorta. Empty the water from the dish containing the specimen, and place a few drops of hæmatoxylin on it ; after one or two minutes wash off the stain, and cover the specimen with clear water. Careful examination will now reveal the dorsal diaphragm, which is so delicate that it is seen with difficulty unless stained.

This diaphragm is attached along its middle line to the ventral surface of the heart. Each lateral edge of the diaphragm is attached to the body-wall in sixteen places, along two lines.

Represent the pericardial diaphragm upon the drawing of the heart and aorta already prepared.

There are differences of opinion as to the function of the pericardial diaphragm.† It seems to us that an important function, if not its chief function, is to protect the heart from the peristaltic movements of the alimentary canal. It also supports the heart ; and may play a part in its expansion.

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*.2 g. lampblack, .2 g. gum arabic, 15 c. c. water; or .4 g. carmine, .2 g. gum arabic, 15 c. c. water.

† See Packard. Text Book of Entomology. pp. 401, 402.

Review.—Take a specimen that has been slit on the *dorsal* surface and make a preparation similar to the one just studied except that it is opened on the back. Review the work on internal anatomy indicated above excepting those parts referring to the nervous system, which will be concealed by the alimentary canal in this specimen, and to the circulatory system, which will be destroyed in the preparation of the specimen.

Note especially the form of the reproductive organs and determine if the specimen is of the same sex as the one previously studied. If it is not, complete the work on the reproductive organs indicated above ; if it is of the same sex, other specimens should be examined after the work on this one is completed.

The suspensory nerves of the alimentary canal.—Gently push the intestine to one side and note the two fine threads extending caudad from the small intestine. Trace out the connection of these threads or nerves with the nervous system. Note the fine branches of these nerves that extend to the caudal part of the intestine.

Make a diagram representing a side view of that part of the alimentary canal caudad of the proventriculus, the last three ganglia of the nervous system, and the nerves just described.

The ventral diaphragm.—Cut the alimentary canal in two between the ventriculus and the first bend in the intestine. Remove that part of the alimentary canal caudad of this point. Cut the tracheæ of one side extending to the remaining part of the alimentary canal, so that it can be pushed away to the other side. Be careful not to injure this part of the alimentary canal till after the vagus nerve has been studied, as indicated in a later paragraph. Empty the water from the dish containing the specimen and put a few drops of hæmatoxylin solution on the muscles and nerves in the abdomen ; after one or two minutes, wash off the stain, and cover the specimen with clear water ; observe the ven-

tral diaphragm. This is a transparent, apparently structure-less membrane, stretched over the floor of the abdominal cavity in such a way as to protect the central part of the nervous system. Owing to the transparency of this membrane, it is very difficult to see it unless it is stained. The ventral diaphragm is attached along each side of the body just laterad of the great ventral muscles ; the points of attachment are on the lines separating the segments of the body. Between the points of attachment, the margins of the membrane curve mesad, giving it the appearance of being strongly stretched at the points of attachment.

This diaphragm has been described as a ventral heart. See Packard, *Text Book of Entomology*, p. 403. We believe its function is to protect the central nervous system from the peristaltic movements of the alimentary canal.

The nervous system.—After removing the alimentary canal from the specimen opened along the back, the central nervous system will be exposed.

Make a diagram showing the disposition of the ganglia and of the principal nerves of the thorax and abdomen. These can be seen better in a specimen which has been opened along the venter. Take for this purpose either a fresh specimen or one that has been slit on the dorsal side, and open it carefully on the ventral side so as to cut through only the body-wall, leaving the nervous system in place.

Make careful dissections of the ganglia and nerves found in the head, and make a diagram showing their arrangement.

The following parts should be observed and figured :—

The supraœsophageal ganglia.—These are two, large, ovoid ganglia, lying above the œsophagus, and connected by a short, thick commissure. They are sometimes termed the brain.

The antennal nerves.—These arise from the latero-cephalic angles of the supraœsophageal ganglia.

The optic nerves.—These arise caudad of the origins of the antennal nerves. Determine the number of divisions of each optic nerve.

The crura cerebri.—These are the two, large cords, one on each side, connecting the supraœsophageal ganglia with the subœsophageal ganglion, and forming with these ganglia the nervous collar of the œsophagus. (The singular form of *crura* is *crus.*)

The vagus nerve.—Just cephalad of the supraœsophageal ganglia there is a minute ganglion, the *frontal ganglion ;* this is connected by an arching nerve on each side with the crura cerebri ; from the frontal ganglion there extends cephalad a small, branching nerve ; from the frontal ganglion there also extend two nerves which pass caudad ; one of these extends to the commissure connecting the supraœsophageal ganglia; the other passes between the supraœsophageal ganglia and the œsophagus, and ventrad of the aorta (which is usually turned to one side in opening the specimen as this one is opened), to a minute ganglion on the middle line of the œsophagus, caudad of the supraœsophageal ganglia. From this minute ganglion two nerves extend, one on each side, to the sides of the alimentary canal, which they follow to the proventriculus, where they divide into many branches. This system of nerves and ganglia is termed the *vagus nerve.*

The subœsophageal ganglion.—This is the large ganglion on the meson, ventrad of the œsophagus. From it two large cords pass caudad to the first thoracic ganglia.

From the subœsophageal ganglia nerves extend to the labium, the maxillæ, the mandibles, and to other parts of the head. The beginning students who are taking a short course in entomology need not trace out these nerves. Other students may use as a work of reference a paper on this subject by Dr. William C. Krauss, published in *"Psyche,"* vol. IV., pp. 179–184.

The muscular system.—In a fresh specimen the muscles appear soft and translucent ; but in specimens that have been kept for a considerable time in a preservative fluid, they are firm and opaque. The greater number of the muscles are attached to the ental surface of the body-wall where they form several layers. This is well shown in the abdomen, where most of the muscles are for moving the segments of the body. In the head and thorax, there are numerous muscles for moving the appendages of the body, and their arrangement is much more complicated.

To attempt to make a detailed study of the muscular system would require much more time than can be devoted to this system in this elementary course. Only the more general features of the structure of the muscles and of their arrangement will be noticed.

Note that the muscular system is composed of an immense number of distinct, isolated, straight fibers, which are not enclosed in tendinous sheaths as they are with vertebrates.

Mount a few of these fibers in glycerine, and study them with a high power of the microscope. Note that the fibers present numerous, transverse striations, like the striped muscles of vertebrates.

Make a figure of a muscular fiber.

In this outline each series or layer of closely parallel fibers is considered as a separate muscle rather than an aggregation of muscles. It complicates the subject unduly to consider each distinct fiber a distinct muscle as has been done by some writers. Thus Lyonet in his " *Traité Anatomique de la Chenille, que range le bois de saule* " describes 1,647 muscles without including the muscles contained in the viscera or those contained in the head.

Take a larva of *Corydalis*, which has been opened on the ventral side and from which the alimentary canal and the

larger masses of fat have been removed, and study the ental layer of muscles of the dorsal wall of the abdomen.

On each side of the heart and ectad of the wings of the heart, there are great bands of longitudinal muscles, occupying the space between the heart and the prominent muscles that extend dorso-ventrad on the sides of the body. Of the longitudinal, dorsal muscles there are two sets on each side. The wider set, which lies near the heart, may be termed the *great-dorsal-recti-muscles ;* the narrower set, which lies between the great-dorsal-recti-muscles and the dorso-ventral muscles of the sides of the body, may be termed the *small-dorsal-recti-muscles.*

Of the dorso-ventral muscles of the sides of the body, referred to above, there are two large bundles on each side of each segment they are situated near the union of the segments.

Between the lateral muscles and the cut edge of the specimen (the dorsimeson) lie the *great - ventral - recti - muscles.* These differ from the great-dorsal-recti-muscles in being somewhat oblique (this is shown better in specimens opened on the back).

Make a drawing of the third, fourth, and fifth abdominal segments representing the muscles mentioned above.

Carefully remove the recti muscles in one or two abdominal segments and note that ectad of them are many muscles extending obliquely in various directions. The study of these oblique muscles will be omitted in this course.

CHAPTER IV.

ANATOMY OF THE LARVA OF THE GIANT CRANE-FLY (HOLORUSIA RUBIGINOSA).

As the larvæ of *Corydalis* are not to be found on the Pacific Coast (and in some other parts of the United States) an account of the anatomy of the larva of a crane-fly (Tipulidæ) is presented for the use of students who may not be able to work with *Corydalis*. The largest Tipulid is the Giant Crane-fly, *Holorusia rubiginosa*, found commonly on the Pacific Coast. The larvæ of this Tipulid attain a length of two to two and one-half inches, and by the absence of fat in the body-cavity, and the ease with which they may be perfectly preserved and readily dissected offer especially available subjects for the study of the internal insect anatomy. As *Holorusia* does not occur elsewhere in the United States than on the Pacific Coast (as far as is known) students in other regions (in which *Corydalis* also does not occur) will have to use the larva of some other Tipulid species. Some rather large Tipulid is common in almost every locality. The account of *Holorusia* will be found to answer as a guide to the dissection of any other Tipulid larva.

EXTERNAL ANATOMY.

Technical note.—Bring a number of the larvæ of *Holorusia* (or other large Tipulid species, to be found in wet moss, vegetable slime or about grass roots in pastures, etc.) alive into the laboratory. Note the various motions and the

locomotion of the body. Kill specimens by dropping into boiling water. After the specimens have straightened out and stiffened, requiring about a minute (death is almost instantaneous) remove to thirty per cent. alcohol. After two or three hours remove to fifty per cent. alcohol, and after three hours into seventy per cent. alcohol. After twelve to twenty-four hours remove the specimens to eighty-five per cent. alcohol, in which keep them.

Structure of body.—(Verify the following statements if *Holorusia* is used : if another Tipulid is used compare conditions with those here described.) The body is composed of thirteen *segments* (exclusive of the head). Retracted into the (apparently) first and second segments is the *head*, with strongly chitinized *skull*. At the anterior end of the head, usually projecting slightly, are the short, cylindrical, unsegmented *antennæ* and the strongly chitinized *mouth-parts*. (The mouth-parts can be better examined after the larva has been dissected and the retracted head wholly exposed.)

There are no *legs* or *tracheal gills* (as there are in the larva of Corydalis). The hindmost body-segment bears terminally on a flat surface two large *spiracles* (breathing openings, see p. 74, Comstock's Manual), surrounded by six backward projecting flexible lobes. On the ventral surface of this segment is the *anal opening* of the alimentary canal on an elevation bearing four large and two smaller flexible processes. The segments of the hinder half of the body have each a median transverse constriction ; those of the anterior half are difficult to distinguish from one another but it is assumed that each pair of the lateral groups of *setæ* of which five pairs may be noted, represents a segment. The absence of all paired appendages back of the head is to be noted.

Make a drawing of the whole body of the larva from a lateral aspect.

INTERNAL ANATOMY.

Technical note.—With fine scissors cut open the body along the median line of the dorsum, cutting through only the body-wall. Put the specimen in a dissecting * dish, pin out the cut edges with ribbon pins and cover with water.

Adipose tissue.—The most conspicuous organ in the opened body-cavity is the alimentary canal extending as a long thick tube longitudinally from head to caudal extremity. It is nearly entirely enclosed in a thin, whitish, perforated sheet of *adipose tissue*, or *fat.* Note the disposition of this sheet. Examine a small piece in water on a slide under the microscope, first under low power and then under higher power. Make a drawing showing the structure of adipose tissue as thus shown.

Alimentary canal and accessory parts.—The alimentary canal is composed of a series of successive regions or parts ; first (foremost) the slender *œsophagus*, embraced by the *circumœsophageal nerve commissures* with the small white *brain lobes* above ; second, an abruptly dilated conical part, the *proventriculus ;* third, a part immediately behind this and not sharply marked off from it, the elongated *ventriculus* bearing at its anterior end four elongated pouches, the *gas-tric cœca.* In the sheet of adipose tissue surrounding the ventriculus several slender convoluted thread-like processes may be seen ; these are the *Malpighian tubules*, the organs of excretion, four in number. They arise from the alimentary canal just back of the ventriculus at a part marked by a pale transverse line. Behind this line is a fourth part of the ali-mentary canal, the *small intestine.* It is of smaller caliber than the ventriculus and opens into a succeeding part of the canal, the *large intestine*, near its anterior end. The large in-

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* For directions for making dissecting dish see pp. 34–35.

testine is largest in front and tapers posteriorly to the very narrow hinder part which is called the *rectum*. That portion of the large intestine in front of the point of entrance of the small intestine may be called the *intestinal cæcum*.

For an account of the functions of the different parts of the alimentary canal see the Cambridge Natural History, Vol. V., pp. 123–127.

Salivary glands.—The salivary glands lie one on each side of the œsophagus. Each is a white, firm U-shaped body, with the two arms much thickened, and the inner reaching a little farther forward than the outer. The *salivary duct* arises from the outer lobe ; the two ducts run forward and unite beneath the œsophagus, the common duct thus formed opening into the mouth-cavity.

Without removing the alimentary canal, make a drawing of it and the salivary glands in position in the body.

Examine a piece of salivary duct in water or glycerine on a slide under the microscope. Note the transversely striated condition. Remove the cover-glass and with needles pull the duct gently apart. Examine again ; the two parts will probably be connected by a spiral thread ; this will be seen to be what formed the transverse striation ; it is really a spiral thickening of the walls, not in a continuous thread, but in a series of short, independent spirals of one or few turns, called *tænidia*.

The respiratory system.—The external openings, *spiracles*, of the respiratory organs, have already been noted in the examination of the external parts of the specimen. Each of these two spiracles opens internally into a main air tube, *trachea*, which runs forward through the whole length of the body. In each of the segments, from the fourth to the tenth inclusive (not counting the head), a large branch trachea is given off from the main longitudinal trunk to the alimentary canal, and a smaller one to the dorsal blood-vessel (see *postea*).

Make a drawing of the tracheal system, tracing the longitudinal and lateral vessels as far as possible.

Cut off a small piece of one of the lateral tracheæ and examine it in water or glycerine on a slide under the microscope. Note the tubular character of the trachea, and note the five transverse striations due to spiral thickenings of the inner wall of the vessel. Pull apart pieces of trachea to show the *tænidia*. See p. 47. Make a drawing of a piece of trachea to show its structure.

For an account of the respiration of insects, see the Cambridge Natural History, Vol. V., pp. 128–132.

The reproductive organs.—The reproductive organs consist in the male of two small, white oval bodies, the *testes*, lying one on each side of the large intestine imbedded in the muscles of the tenth body-segment, and of a delicate duct, the *vas deferens*, running posteriorly from each to the ventral wall of the penultimate segment. In female specimens the white bodies, *ovaries*, are larger and more elongate than the testes of the male, and the *oviducts* (corresponding to the vasa deferentia) are more easily seen.

Make a combined drawing showing the alimentary canal, salivary glands, tracheal trunks, and reproductive organs in position in the body.

For a dissection of the reproductive organs in an adult insect, see pp. 41–46.

The nervous system.—Remove the alimentary canal, cutting across the œsophagus near the anterior end of the proventriculus. The *brain* is composed of two main lobes united posteriorly and lying above the œsophagus. Beneath the brain, just under the œsophagus, lies the *subœsophageal ganglion*, which is connected with the anterior end of the brain-lobes by the *circumœsophageal commissures*, nerve-cords which together with the brain and subœsophageal ganglion form a complete ring around the œsophagus. Back of the subœsophageal

ganglion is a chain of four closely connected ganglia. Leading back from the hindmost of these ganglia are paired, but closely opposed, longitudinal nerve-cords. On these cords in the sixth body-segment is another ganglion, and following this are five others similar to it, each lying over the center of the sternal part of a segment. These ventral ganglia, from the subœsophageal back, and the connecting longitudinal commissures constitute the *ventral nerve chain* which with the brain and circumœsophageal ganglia compose the *central nervous system* of the insect. In addition to this central system there is another smaller system, the *sympathetic nervous system*, which we shall not study in this elementary dissection. Each ganglion of the ventral chain, behind the subœsophageal ganglion, gives off four very conspicuous *nerves;* one on each side arising from the middle of the ganglion going to the muscles of the body-wall, and another arising from the anterior end of the ganglion going to the viscera. The last ganglion lying in the antepenultimate body-segment, in addition to the four lateral trunks, gives off from its posterior part two large divergent ones running backwards to the two following segments. The brain and subœsophageal ganglion give off nerves to the organs of the head.

Make a drawing of the nervous system.

The muscular system.—Along each side of the dorsal and ventral median line of the body is a wide band of longitudinal *muscles*. The most conspicuous fibers reach from the anterior to the posterior border of each segment, but the others reach from either end to the middle, while others extend from the middle of one segment to the middle of the preceding or following segment, while still others are attached to various points of the body-wall between the attachments of the sets already mentioned. Finally, there is an innermost set of lateral transverse muscles in the anterior half of each segment.

Make a drawing showing the complete musculation of two successive segments.

The circulatory system.—Cut a second specimen open longitudinally along the median line of the venter, reserving the first specimen for some later work. Pin out the cut edges. Note again the general disposition of the body organs so far examined. Examine again the reproductive organs ; the specimen may be of the other sex from that previously studied. Remove the alimentary canal.

The *dorsal vessel* or *heart* is a slender, delicate, membranous tube composed of a number of successive parts or chambers lying along the median line of the back. To see this well cut out the median dorsal strip of body-wall carrying the heart and transfer it to a glass slide. Cover with water and examine first under the simple microscope and then under the low power of the compound microscope. It will be distinctly seen that toward the middle of each segment from the last to the third inclusive, the tube becomes dilated, forming a chamber, and in most of these chambers, except the last, a pair of internal valves may be seen. From the most anterior chamber a straight, tapering tube, the *aorta*, runs forward into the head, where it ends by dividing into branches which can be followed for but a short distance, apparently fading out completely. There are no other blood-vessels in the body.

On each side of each chamber may be noted a fan-shaped group of very delicate muscle fibers, apparently attached to the sides of the chamber, and called the *wing muscles* of the heart. The convergent outer ends of these muscles are attached to the body-wall on the line of the median constriction in each segment. These muscles really lie in a thin membrane which forms a sort of *pericardial membrane* enclosing a sinus on each side of the heart and stretching across from these lateral sinuses to the body-wall as a *septum* or

diaphragm. This membrane is attached to the heart on its dorsal and its ventral surface. The membrane and sinuses can be especially well seen in cross sections of the body.

For an account of the circulation in insects see the Cambridge Natural History, Vol. V., pp. 132–136.

The histoblasts of wings and legs.—Returning to the first specimen (that opened along the dorsum) carefully remove the muscle fibers from the body-wall of the three front (thoracic) segments of the body, not including the head. Be careful not to remove certain small whitish bud-like bodies lying between the muscles and the body-wall. In specimens of sufficient age the *histoblasts* (or *imaginal buds* or *imaginal discs* as variously called) of the wings and legs of the adult fly and of the external prothoracic respiratory tubes may be seen as small sac- or bud-like bodies lying against and attached to the inner surface of the body-wall of the thoracic segments. There are two pairs of these imaginal buds in each thoracic segment corresponding respectively to the prothoracic legs of the imago and prothoracic respiratory tubes of the pupa, the meso-thoracic legs and wings of the imago, and the meta-thoracic legs and halteres of the imago. The study of the morphology and development of these imaginal buds is too difficult to be referred to here. It is sufficient to know that the legs and wings of the fly begin thus to develop under the surface of the body and continue this internal development until the larva changes into a pupa. These buds may be seen to be intimately connected with the thoracic skin, and are, in fact, actual invaginated parts of this skin.

The head and its appendages.—After finishing the dissection of the internal organs remove the head entirely from the rest of the specimen and examine with simple microscope. Each of the short *antennæ* arises from a small lobe on the plate covering the top of the head. This plate is long, tapering and decurved behind. It is united along the anterior parts of its

sides with *lateral plates*, while the anterior margin is re-entrant receiving the smaller end of the pear-shaped *labrum*. The distal part of this sclerite is membranous except for two lateral chitinizations. Posteriorly it is fused with the *epicranial plate*. The lateral plates are each oval and shell-shaped, having their anterior lower angles produced forward and united with each other. The single process thus formed projects forward and curves upward between the faces of the posterior jaws. The tips are provided with graduated teeth. In front of each lateral plate is a narrow dorso-ventral sclerite carrying the jaws. The anterior ones, *mandibles*, are large, strong, toothed terminally and provided on the inner side with a large, softer, movable lobe. The posterior jaws, *maxillæ*, are less strongly chitinized than the mandibles. They are flat, and each is provided at its outer angle with several papilla-like processes.

CHAPTER V.

THE EXTERNAL ANATOMY OF A BEETLE.
Pterostichus californicus.

With the knowledge of the external anatomy of the locust as a basis the student may successfully attempt to examine comparatively some of the various conditions of the body exhibited among different orders of insects. With the varied habits of insects there are necessarily correlated various modifications of structure, internal and external. The modifications of the external structure are those taken special cognizance of and used in the present analytical tables and keys for insect classification, and must be studied to some degree before determination of insect forms can be done intelligently. The study of insect anatomy in a comparative way will also give the student an understanding of the significance of homology and specialization.

As an example of simple work of this character which may be undertaken by the student, the external anatomy of a beetle (order Coleoptera) may be studied.

The peculiarly flattened form of many insects, by which the lateral aspects of thorax and abdomen are reduced to a mere ridge or margin, is accompanied by a change in the position of many of the body sclerites, in particular the pleural sclerites of the thorax. This condition is well exemplified among the predaceous ground-beetles (*Carabidæ*) and almost any species may be selected for illustration. We have chosen the species *Pterostichus californicus*, as the representative of a widely spread genus, and the description fol-

lowing applies especially to this form ; however, the notes will serve as a guide for the examination of any member of the genus.

The meso- and metathoracic segments are closely joined to each other and to the abdomen. The prothorax is freely movable and is constricted at its articulation with the meso-thorax, so that it appears to form all of the second or tho-racic region of the body. The insertion of the hinder two pairs of legs, however, shows that part of what at first glance appears to belong entirely to the abdominal region of the body really belongs to the thorax. The body-wall is very strongly chitinized, the body being enclosed in a veri-table coat of armor. The head projects horizontally from the body instead of hanging vertically across the front, as with the locust, and the flattening of the body is evident in all regions, head, thorax, and abdomen.

PARTS OF THE HEAD.

THE FIXED PARTS OF THE HEAD.

The fixed * parts of the head are fused so as to form a strong and rigid box, which is elongated and flattened.

Epicranium.—The *epicranium* bears on its cephalic por-tion two impressed lines which run cephalad until they meet the transversal *clypeal suture*, the suture separating the cly-peus from the epicranium.† Laterad of each of the impressed lines on the epicranium there is, forming the dorso-lateral margin of the head, a sharp ridge called the *frontal ridge*, which runs cephalad from the dorsal margin of the eye.

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* The "head-capsule" of this beetle is very strongly chitinized and deep black in color. To soften and partly bleach the head, so that the fixed parts may be readily determined, the student must boil the head for some time in caustic potash (KOH).

† The epicranium of the beetle, like that of the locust, is a compound sclerite, being composed of the fused true epicranium and front.

The antennæ arise just below this frontal ridge in a short, rounding groove running cephalad from the eye. On the epicranium just above each compound eye are two, long hairs arising from distinct pits. These pits are called *setigerous punctures*. Similar punctures and hairs are found also near the lateral margins of the clypeus.

Clypeus.—The *clypeus* is broader than long, and projects cephalad between the bases of the mandibles. Projecting cephalad from the cephalic margin of the clypeus, is the subquadrangular *labrum*, with its distal margin slightly concave outwardly.

Compound eyes.—The *compound eyes*, on the lateral margins of the head, are comparatively small.

Ocelli.—There are no *ocelli*.

Genæ.—The portions of the epicranium below the eyes and antennæ are the *genæ* and each projects cephalad, lateroventrad of the base of the mandibles, as a thin tapering tongue curving inward slightly at its tip. Below this projecting process, the gena presents a rounded emargination, and, filling in the emargination, may be seen the basal portion of the maxillæ. The genæ form, with portions of the occiput, the lateral portions of the ventral surface of the head.

Occiput.—The *occiput*, although fused with the epicranium to form the firm head-box, is plainly separated from the epicranium on the dorsal and lateral portions of the head by an impressed line, which fades out on the lateral portions of the ventral surface of the head, so that the postgenal and occipital regions cannot here be distinguished.

Gula.—Forming the mesal third of the ventral aspect of the head, slightly widening caudad, and expanding at its cephalic extremity to a narrow, transversal bar, which projects laterad to the genal emargination, is the *gula*, one of the head sclerites, which is wanting or is fused with the sub-

mentum in the head of the locust. The gula is usually a
well developed sclerite among beetles. The sutures separat-
ing it on either side from the contiguous portions of occiput
and epicranium (postgena) are called the *gular sutures.*

Make a drawing of the ventral aspect of the head, show-
ing the skeletal parts described, and also what may be seen
of the mouth-parts *in situ.*

Antennæ.—The *antennæ* are filiform, and 11-segmented ;
the third segment is the longest one. The proximal three
segments are glabrous, and the second and third bear each
two or three longish hairs near their distal end. The re-
maining eight segments are finely pubescent. In addition
each of these eight bears a few longer hairs at its distal end.

Mouth-Parts.

The *mouth-parts* are fitted for biting and are in general
similar to the mouth-parts of the locust.

Labrum.—The labrum has been described already as a
part of the skull.

Mandibles.—The *mandibles* are rather long, and taper
distad to a sharp, curving tooth. They bear on their sharp,
cutting, inner margin, near the base, a few, small, blunt
teeth, and their outer face presents a broad, shallow groove
or furrow called the *mandibular scrobe.* Make a drawing of a
mandible.

Maxillæ.—The *maxillæ* differ especially from those of
the locust by the presence of an additional sclerite, the sub-
galea, by the side of the stipes, as if the stipes were divided
by a longitudinal suture. The *cardo* is large, and broadly
club-shaped ; the median portion of the maxilla is composed
of three sclerites, the *stipes, palpifer* and *subgalea.* These

three sclerites may be distinguished when the maxilla is viewed from the dorsal aspect by the following characters ; the palpifer lies above the distal two-thirds of the stipes, and also overlaps part of the subgalea. It is elongate, sub-triangular in outline with apex directed toward the lacinia. From it arises the four-segmented palpus, and it also bears near the origin of the palpus a single, conspicuous, long bristle. The stipes as seen from above is elongate and rather slender, with the basal extremity widened. It lies laterad of the large subgalea, which forms the inner or mesal half of the median portion of the maxilla. From the sub-galea the two lobes, *galea* and *lacinia*, of the maxilla arise. The galea is slender, elongate, distinctly two-segmented, and palpiform in character. The lacinia is strongly chitinized, and is beset on its inner face with strong and long, curved spines, and on its dorsal aspect with many, weaker hairs. It bears on its distal end a strong tooth or curved tip, known as the *digitus*.

Make a drawing of the dorsal aspect of a maxilla.

Labium.—The *labium* is separated from the cephalic transverse portion of the gula by a straight, transverse suture. The *submentum* is large, strongly chitinized, and with its lateral portions appearing as broad, expanded lobes, separated by a broad and deep, cephalic emargination. Bordering this emargination is the narrow curving *mentum*, with a median forward-projecting, two-pointed portion. From the elongated, subcylindrical, forward-projecting pal-pigers arise the three-segmented *labial palpi*, the first seg-ments of which are very short. Also projecting forward from the mentum is the *ligula*, composed of a dorsal or inner, delicate, transparent, membranous part, which divides at its tip into two, slender, tapering projections, the *para-glossæ*, and a ventral or outer, chitinized, undivided portion, the *glossa*. The tip of the glossa is truncate, and bears two,

long, strong hairs. Each paraglossa lies slightly laterad of the distal half of the glossa. The entire ligula arises, not from the cephalic margin of the mentum, but from its inner or dorsal face, and it may be that its parts are not homologous with the terminal lobe (ligula) of the locust's labium, but that one or perhaps both parts are continuous with the lining of the mouth-cavity ; in which case they would be homologous with the hypopharynx of the locust's mouth.

Make a drawing of the ventral (outer) aspect of the labium.

PARTS OF THE THORAX.

PROTHORAX.

Dorsal aspect.—The *pronotum* is not divided into different sclerites, but appears as a single, firm, convex plate, bearing a median impressed line, and, laterad of this line, on each side near the caudal margin, a short, linear depression. At the acute lateral · margins, the pronotum is inflexed, extending a little distance ventro-mesad on the ventral (pleural *s. str.*) aspect of the body. This inflexed portion is often called the *prothoracic epipleura.*

Ventral aspect.—The *sternum* and the true pleural sclerites or "side pieces," together form the ventral aspect of the prothorax.

Sternum.—The *sternum*, constituting the median region of this aspect, is irregularly saddle-shaped, with a caudad-projecting tongue between the *coxal cavities.* This tongue after reaching the caudal margins of the coxæ bends at right angles and projects dorsad, the end expanding slightly into two dorso-laterad projecting points, which meet a ventro-mesad projecting point of the epimeron on each side, and thus form part of the enclosing, caudal boundaries of the coxal cavities.

Episternum.—The *episternum*, a large rhomboidal sclerite, constitutes most of the body-wall between the sternum and the epipleuræ.

Epimeron.—Separated from the caudal extremity of the episternum by a distinct suture, is the narrow, curving *epimeron*, whose expanded mesal extremity presents two, pointed, curving processes, which enclose the coxal cavity laterad and caudad. The meeting of the epimeron and sternum caudad of the coxal cavity technically *closes* it, or makes it *entire ;* if these two sclerites do not meet, as is often the case among beetles, and the cavity is bounded caudad simply by membrane, the cavity is said to be *open.* If there is no caudal tongue projecting between the cavities, they are said to be *confluent ;* when separated, as in the specimen in hand, by this tongue, they are technically *separate.*

MESOTHORAX.

Dorsal aspect.—When the wing-covers are folded the only part of the mesonotum visible is the small, median, triangular or shield - shaped portion of the *scutellum.* By spreading apart or breaking away the wing-covers, the lateral membranous portions of the scutellum may be seen, as well as the *scutum ;* the median part of the scutum is strongly chitinized and the lateral parts, weakly chitinized. The *postscutellum* also may be distinguished as a narrow, weakly-chitinized, curving bar, running laterad on each side just in front of the caudal apex of the scutellum. The *præscutum* is represented merely by a thin, transversal strip of membrane.

Ventral aspect.—As with the prothorax the ventral aspect of this segment is composed of the sternum and the pleural sclerites.

Mesosternum.—The *mesosternum* is plainly set off by sutures. Its caudal margin has a broad, median tongue,

which projects caudad between the coxal cavities, and is angularly emarginated at its tip. Laterad of this tongue are two rounding emarginations, for the reception of the coxæ. Bounding each coxal cavity laterad is a caudad-projecting portion of the sternum. Cephalad the sternal sclerite tapers somewhat, and the cephalic margin is narrowly truncate. A narrow, collar-like cephalic margin which fits into the prothorax, is separated from the rest of the sternum by a slight carina or elevated line.

Episternum.—The pleural sclerites are distinct ; the *episternum* is large, angularly concave, and does not reach the coxa. It bears near its cephalic margin two, transversal raised lines or carinæ, one of which is a continuation of the collar-making carina of the sternum. Like the sternum, the cephalic margin of the episternum fits into the prothorax. The lateral margin is angularly inflexed along its entire length, as is the case with all the pleural sclerites. The narrow, inflexed portion is covered when the wing-covers are closed by the inflexed, lateral margin of the wing-covers.

Epimeron.—The *epimeron*, lying along the caudal margin of the episternum, is a narrow, transversal sclerite. Its mesal extremity does not reach the coxal cavity, but lies contiguous to the lateral margin of the metasternum.

METATHORAX.

Dorsal aspect.—When the elytra are closed the metanotum is completely covered and invisible. By removal of the elytra, the metanotum is revealed as a narrow transversal bar, on the surface of which a number of sutures and elevated and depressed lines are to be seen. The work of distinguishing the various component sclerites of the metanotum cannot be done satisfactorily by the elementary student. There is a conspicuous depression or groove ex-

tending caudo-cephalad along the middle of the back, with strongly chitinized margins which project slightly mesad over the depression. On the inner surface of the elytra, near the base of the mesal margins, there are two, slight, projecting processes. These small processes have strong, acute margins, which project slightly laterad. When the elytra are closed, the raised processes fit into the groove of the metanotum, and the laterad-projecting margins of the processes lie under the mesad-projecting margins of the groove, the whole structure forming a means for the firm holding of the elytra over the dorsum of the body. The firm holding of the elytra is further aided by the inflexed lateral margins and by the close dovetailing of the mesal margins along the middle of the back.

Ventral aspect.—As in the other thoracic segments this includes both the sternum and the pleural sclerites.

Metasternum.—The *metasternum* is, as the mesosternum, best described as saddle-shaped. It has a rather broad, blunt tongue, projecting cephalad between the mesocoxæ to meet the caudad-projecting tongue of the mesosternum. It presents, also, an acute-angle process, projecting caudad between the cephalic halves of the metacoxæ. The "saddle-flaps," or lateral lobes of the metasternum expand laterad, and their cephalic margins, concavely rounded, form the caudal boundaries of the mesocoxal cavities. There is a line or suture running transversely across the metasternum near the caudal margin which does not reach the lateral margins. That portion of the metasternum caudad of this suture is called the *antecoxal piece* of the metasternum.

Episternum.—The rhomboidal *episternum* is the largest of the pleural sclerites.

Epimeron.—The trapezoidal *epimeron*, though smaller than the episternum, is broader, and more conspicuous than that of either of the other thoracic segments.

Make a drawing of the ventral aspect of the entire thorax, including the legs of the left side.

APPENDAGES OF THE THORAX.

The hind wings are wanting in this beetle. This is exceptional, however, among beetles, the hind wings, membranous and with a few strong veins, being usually well-developed, and differing from the wings of most insects in being folded transversely as well as longitudinally when the insect is at rest.

The fore-wings are very strongly chitinized and thickened, and are called *elytra* or wing-covers. When the beetle is at rest the elytra fit closely over the dorsal aspect of the meso- and metathorax and abdomen, protecting the abdomen and hind wings, which (when present, as in most beetles) lie folded over the metathorax and abdomen and beneath the wing-covers. The elytra are articulated with the body so as to be freely movable, being outspread when the beetle is in flight. The basal or articulating parts of the elytra lie just ventro-laterad of the lateral margins of the scutellum. The expanded flap or wing-like parts present a series of sub-parallel, longitudinal, impressed lines, and the lateral margins, termed *epipleuræ*, are inflexed over the dorso-lateral margins of the body. This inflexed condition of the margins does not extend quite to the tips of the elytra, but disappears at a point where the margin appears to be *interrupted*. On the inner surface of each elytron near the lateral margin there is a distinct longitudinal fold or *plica*.

PARTS OF THE ABDOMEN.

The abdomen is composed of a number of very much flattened segments ; and its dorsal surface or *tergum* is completely covered by the elytra when they are closed. On the

ventral surface, six strongly-chitinized *sterna* may be count-
ed. The first (basal) sternum is completely divided by the
coxal cavities, so that it appears as two triangular pieces,
lying laterad of the coxæ. The cephalic margin of the sec-
ond sternum is emarginated on each side of the meson by
the coxal cavities, so that the mesal part of the cephalic
margin appears as an acute, cephalad-projecting process.
The third, fourth and fifth sterna are of about equal length
(caudo-cephalic), the sixth being longer and having its cau-
dal margin roundly pointed. The first, second and third
sterna are connate (firmly united, not movable on each
other), although the sutural lines are distinct. All of the
sterna have strongly chitinized, lateral margins, which pro-
ject somewhat dorsad, and then are narrowly inflexed over
the dorso-lateral margin of the abdomen. There are seven
terga, the tergal aspect corresponding to the sixth sternum
being unequally divided by a transversal suture, producing
thus an additional small tergum. All of the terga are mem-
branous except the sixth and seventh, which are chitinized,
and are known respectively as the *propygidium* and *pygidium*.
The propygidium is coarsely punctulated, but the surface of
the pygidium is smooth. Each tergum except the seventh
bears a pair of *spiracles*, located near the cephalo-lateral
angles of the terga. Between the lip-like caudal margin of
the pygidium and the sixth sternum there is a transversal
fissure, in which lie the anal and genital openings.

CHAPTER VI.

THE MOUTH-PARTS OF INSECTS.

The study of the mouth-parts of insects is of special interest and importance because of the admirable illustration of the principle of homology presented by the mouth-parts, because in the classification of insects great importance is assigned to the mouth-structure, and finally because of the significance of the structural character of the mouth in the study of the habits and economic relations of insects.

The mouth-parts of the locust, the cockroach, and the beetle have already been studied: they represent the biting type of insectean mouth-parts. The student is, therefore, already acquainted with the component parts of the mouth, and with their character in the biting type. The mouth-parts of many insects, however, are not of the strictly biting type, but are modified to form an elongate tubular beak or proboscis used for sucking liquid food, or are modified to form a combination of the biting and sucking types. This modification of the more generalized biting type varies in the various insect orders possessing "sucking mouth-parts." As a beginning in the comparative study of the sucking type, the mouth-parts of a short series of insects, representing several important orders, may be examined.

In the examination of the mouth-parts dried specimens can be advantageously employed. Remove the head from the dried insect and boil it in dilute potassium hydrate (KOH) to soften and partially bleach the chitinous parts. The length of time of boiling will depend, of course, upon the degree of

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chitinization and coloration of the parts. The head should then be washed in water and removed to a watch glass containing water. The mouth-parts can now be examined *in situ* and finally readily dissected apart by means of dissecting needles and forceps.

The parts can be temporarily mounted in glycerine, or if permanent mounts are desired, the parts can be mounted in glycerine jelly. Carry from the water directly into the jelly. For mounting in Canada balsam the parts must be thoroughly dehydrated by carrying through successive strengths of alcohol up to absolute, and then clearing in xylol or other clearing oil.

BITING AND SUCKING MOUTH-PARTS.

In the Hymenoptera, the mandibles retain their function of biting, while the maxillæ and labium unite to form an apparatus for lapping or sucking liquids. Two examples of this condition of mouth-parts fitted for biting and sucking may be examined : a wasp, in which the maxillæ and labium are somewhat elongated and fitted for lapping ; and a honeybee, in which the maxillæ and labium are much elongated and fitted for sucking.

MOUTH-PARTS OF THE WHITE-FACED HORNET.

Vespa maculata ; order *Hymenoptera.*

Because of the considerable membranous and fleshy tissue of the mouth-parts, bleaching in KOH should not be carried far ; a minute or two of boiling, sufficient for softening, will do. In fact, simply softening in warm water is perhaps safer.

The mouth-parts of the wasps and bees are so thoroughly united at the base by the development of the pharyngeal skeleton and membrane that a considerable tearing of tissue is necessary to separate the parts. The mouth-structure as

a whole should be carefully examined before the parts are dissected out.

In the study of the mouth-parts of insects the student should bear in mind that the labrum, although in position and use actually one of the mouth-parts, differs morphologic-ally from the mandibles, maxillæ and labium in not being a modified pair of head appendages.

Labrum.—The labrum is short and broad, and is immov-ably fused with the clypeus.

Epipharynx.—Directly underneath the labrum is the well-developed elongate strap-like epipharynx. It is fleshy and membranous and bears numerous, spiny hairs.

Mandibles.—The mandibles are large and toothed. When they are closed one overlaps the other to a considerable extent.

Maxillæ.—The maxillæ, although of the generally simple character of the biting type, show some rather confusing modifications. The cardo is strongly chitinized, distinct, rather large and subclavate in shape. The stipes is large, strongly chitinized and is (like the stipes of the beetle's maxilla) partly divided into three sclerites, from one of which rises the small, slender, six-segmented palpus. The terminal lobes assume a peculiar character, the distinctly two-seg-mented galea being large and flat, and bearing many papillar hairs. From the large basal segment of the galea there rises a distinct, flattish lobe also bearing many long papillar hairs. Make a drawing of a maxilla.

Labium.—The labium is a complex organ in which, how-ever, the typical composing parts can be pretty readily dis-tinguished. The submentum and mentum are fused to form a large strongly chitinized, bent, basal sclerite. This scle-rite is bent so as to present a strongly convex under (outer) surface, and a deeply concave upper (inner) surface. This deep concavity or furrow is filled with the fleshy and mem-

branous folds of the hypopharyngeal or ventral lining of the mouth. There is a certain, short, broad, fleshy flap which may be called the *hypopharynx*. From this basal sclerite (fused submentum and mentum) arise the slender four-segmented labial palpi, and the terminal lobes. The paraglossæ are distinct, thin, membranous structures with the basal portion well chitinized and with a peculiar button-like chitinized tip. The glossæ are fused at their base, and show a well-developed basal chitinous skeleton-plate. The membranous portions of the glossæ are also fused for about half their distance; the apical halves, however, are free, and each one bears a button-like, chitinized tip, like the tips of the paraglossæ.

Make a drawing of the labium.

Make a drawing of the cephalic aspect of the head showing the fixed and movable parts.

MOUTH-PARTS OF THE HONEY-BEE.

Apis mellifica ; order *Hymenoptera.*

Most of the bees in alcohol (if an alcohol specimen is used) will be found to have certain of the mouth-parts protruding.

These parts are the maxillæ and labium, united at the base and associated to form a sucking proboscis. Before the detailed examination of these parts is begun the student should discover the labrum and mandibles.

Labrum.—The labrum is small and oblong in shape with its laterocephalic corners rounded. The breadth (lateral) is about three times its length (caudo-cephalic).

Mandibles.—Partially concealed beneath the labrum are the spoon-shaped mandibles. With forceps or needle, press the mandibles apart at their tips (press laterad). Note that the mandibles are not toothed, but are rather paddle or spoon-

like at the tips. Remove a mandible and make a drawing of it.

Remove both mandibles and labrum, and grasp the remaining protruding parts with forceps and carefully pull them loose from the head. Wash while still held in the forceps, and mount in glycerine on a glass slide. Before reading farther the student should endeavor to name the various parts presented before him on the slide.

Make a drawing of the maxillæ and labium, and name the parts, tentatively. Compare the result with the notes following :—

Maxillæ.—The parts of each maxilla present are the cardo, stipes, galea (or lacina ; one of the two is probably wanting), maxillary palpi, and possibly the palpifer.

Cardo.—The *cardo,* or proximal part of the maxilla, is a rather long, slender, strongly chitinized sclerite, somewhat resembling a human femur or thigh-bone in shape. At its proximal end it terminates in two unequal prongs, the point of the larger being bluntly rounded. At its distal end (articulating with the stipes) it expands club-like.

Stipes.—The stipes is an irregular, elongate sclerite, strongly chitinized. Its proximal end is bluntly rounded and swollen. The *stipes* articulates with the proximal segment of the galea (see below) by a long diagonal face.

Galea.—The *galea* (we incline to believe this part homologous with the galea of the locust's maxilla, rather than with the lacinia, because of its two-segmented condition) extends distad from the stipes as a tapering blade-shaped piece. It is composed of two segments. The proximal one is small and triangular, articulating by the entire length of one of its margins with the stipes. The distal segment or sclerite constitutes the real blade-like portion of the maxilla, and nearly equals in length the ligula and labial palpi (see below). Its surface is unequally divided into two portions by a subme-

dian, dark-brown, longitudinal line. (This line may indicate a coalescence of galea and lacinia into this one blade-like compound sclerite.) This line bears several hairs, and there are scattering hairs elsewhere on the sclerite, especially toward the distal end. Near the proximal end of this distal segment of the maxilla, and between the longitudinal line and the outer margin, many two-jointed papillæ (taste organs ?) can be seen with a high power.

Maxillary palpi.—The *maxillary palpi* are minute, exarticulate, outward-projecting pieces, arising from near the outer end of the suture separating the proximal from the distal segment of the galea.

Labium.—The parts of the labium present are the submentum, mentum, glossa, paraglossæ, palpifer, and palpi.

Submentum.—The *submentum* is a small, shield-like piece; its proximal end is connected by two chitinous bands, the *lora*, with the cardo of each maxilla; its distal end articulates with the mentum. The submentum is rather feebly chitinized.

Mentum.—The *mentum* is rather oblong in shape, with rounding corners, and strongly chitinized.

Glossa.—Rising from the distal end of the mentum is the long *glossa*, which terminates in a small transparent lobe or *flabellum*. The glossa should be carefully examined under low and high magnification. Note the reticulated and hairy surface. The visible surface is that of a sheath which encloses a slender flexible rod, the rod being probably concerned with the movements of the organ.

Paraglossæ.—At each side of the glossa, and rising from near its proximal end are two subtransparent lobes or flaps, extending about one-fifth the length of the glossa. These are the paraglossæ.

Palpifers and Labial Palpi.—Lying just laterad of the paraglossæ and rising from the distal end of the mentum are

two, long, flattened processes, composed of several segments. The basal segment, which is the largest and longest of the segments, extends forward for about one-half the length of the ligula, and is the palpifer ; the succeeding three segments compose the labial palpus.

The proximal one of these segments is the largest and appears to be a direct continuation of the palpifer, separated only by a narrow straight suture ; the remaining two segments are very small.

Make a drawing of the cephalic aspect of the head showing the fixed and movable parts.

PIERCING AND SUCKING MOUTH-PARTS.

Among the insects whose mouth-parts are fitted exclusively for piercing and sucking (or sucking alone) the mandibles, if present, are modified to form slender elongate stylets or blades.

This is the condition presented by the Hemiptera, and some of the Diptera ; with most of the Diptera and all of the Lepidoptera (excepting Eriocephala) the mandibles are completely lost, or are so atrophied as to be functionless. As examples of insects with strictly sucking mouth-parts the student may examine a Cicada (Hemiptera), a horse-fly and a house-fly (Diptera, one with and the other without mandibles) and a butterfly (Lepidoptera).

MOUTH-PARTS OF A CICADA.

(*Cicada sp.;* order *Hemiptera.*)

The sucking beak, tapering from base to tip, will be found, in dried specimens, usually appressed to the ventral surface of the body of the Cicada.

Remove the head with the beak from the dried specimen,

and examine the beak before dissection. The long, three-jointed *labium*, forming all of the beak as seen superficially, is specially chitinized (brown) near its distal end. The distal joint is the longest, and its surface is sparsely covered with fine, whitish hairs. The tip is rather blunt than acute. A narrow channel, widest at its proximal end, runs along the upper face of the labium. In this channel, but concealed by the approaching edges of it, lie the mandibles and maxillæ. A glimpse of the mandibles and maxillæ just at the base of the labium can often be had. Above the base of the labium is the minute, acute-angled *labrum* lying just over the entering mandibles and maxillæ.

With a dissecting needle carefully break away the head-wall and muscle near the base of the beak, especially dorsad and laterad. The bases of the *mandibles* and *maxillæ* will be discovered as small, strongly chitinized (brown), terminal dilations of slender, chitin rods, which run forward into the channel of the labium. Note the relative position of the two rods with dilated bases on either side, and decide which is mandible and which maxilla. (The rod lying slightly dorsad and laterad of the other is the mandible ; the mandible is also thicker and larger than the maxilla.) Trace the slender chitin rods or stylets (the mandibles and maxillæ) into the channel of the labium. Here they are all closely appressed, the two maxillæ specially so, so that they can be separated only with difficulty.

Remove the mandibular and maxillar stylets and note the channel in which they naturally lie. The labium is more strongly chitinized along the walls of the channel than elsewhere, except at its tip.

Make a drawing of the mouth-parts from dorsal view, with the mandibles and maxillæ removed from the channel of the labium and spread apart.

MOUTH-PARTS OF THE HORSE-FLY.

(*Tabanus* or *Therioplectes sp.;* order *Diptera.*)

Among the flies with piercing mouth-parts only the females possess mandibles. It is therefore necessary to select a female horse-fly (distinguished from the male by the narrow space between the eyes ; in the males the eyes touch each other for a greater or less distance along the dorsal aspect of the head).

The projecting mouth-parts are conspicuous. On superficial examination there may be noted two, thickened, slightly curving, horn or club-like processes (the maxillary palpi) projecting above a black, thickened stalk or trunk (the labium), on the dorsal surface of which lie several light brown, slender, pointed stylets (mandibles, maxillæ, labrum, epipharynx, and hypopharynx).

For the detailed examination of the mouth-parts, the head of the fly should be removed from the body, a considerable part of the head, laterad and caudad, broken away and the remainder, with mouth-parts attached, boiled in KOH to soften and bleach.

The large *maxillary palpi* are two-segmented ; the distal segment is longer than the basal one, and compressed. The proximal one is subcylindrical, and projects dorso-cephalad, so that the large distal segment is carried above the rest of the mouth-parts.

Lying along the dorsal surface of the large labial trunk are six long, slender, pointed pieces or *stylets.* The uppermost, unpaired, flat piece is the *labrum* (or perhaps labrum and *epipharynx* fused). It is rather bluntly tipped and is the broadest of the stylets. The flat, smooth, sharply pointed *mandibles* lie just above the less strongly chitinized, narrower, and finely marked *maxillæ.* Corresponding somewhat to the labrum, but less broad and strong, is the sixth

stylet, an unpaired slender piece lying below the maxillæ. This is the greatly developed hypopharynx.* These six stylets, labrum, mandibles, maxillæ, and hypopharynx are the instruments with which the female horse-fly pierces the skin of animals to get at the blood ; the male has no piercing stylets, and feeds on flower-pollen.

Beneath the grouped stylets is the long trunk- or proboscis-like *labium*, presenting on its upper surface a shallow furrow in which the stylets may be partially enclosed, and presenting at its distal extremity a conspicuous, expanded, disk-like part called the *labella*. This terminal disk is believed by some entomologists to be composed of the greatly modified *labial palpi*. It is made up of two fleshy lobes or leaves, bearing on the outer or under surface many fine, transversal, subparallel lines or ridges. The two lobes can be closed together like the leaves of a book.

Make a drawing showing all of the mouth-parts from the dorsal view. The stylets can be spread apart laterad, so as to expose the under ones.

In only a few families of Diptera are free mandibles present, and when present they are possessed, as already mentioned, only by the females. In many flies there are no piercing stylets, and as representative of these flies without piercing mouth-parts the common house-fly may be studied.

MOUTH-PARTS OF THE HOUSE-FLY.

(*Musca domestica ;* order *Diptera.*)

In the house-fly the mandibular and maxillar stylets being gone, we find only the trunk-like labium, with the labrum-epipharynx lying closely appressed to, and almost fused with its dorsal surface, and the maxillary palpi.

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* The hypopharynx and the epipharynx (outgrowth from the upper wall of the pharynx) are in most insects small, fleshy, and inconspicuous.

The maxillary palpi are prominent, but are only one-segmented. The labial trunk or proboscis may be described as being made up of three portions, a basal third, the *basiproboscis*, from which arise the maxillary palpi, and in which are imbedded two slender chitinous rods, the "*maxillary tendons*," probably representing the greatly reduced maxillæ; a middle third, the *mediproboscis*, strongly chitinized; and a distal third, the *distiproboscis*, including the disk-like, fleshy *labella*. The labella is like that of the horse-fly, but in the house-fly it is the only organ for obtaining food. With it traversed as it is by transverse, horny, chitinous ridges, the "*pseudotracheæ*," hard food substances may be rasped so that fine particles of food mixed with, or sometimes dissolved in, a salivary secretion, which issues from the ridges, can flow into the mouth, along the dorsal furrow of the labial proboscis.

Make a drawing of the mouth-parts from a lateral view; and also of a portion of the labella, highly magnified, to show disposition of the pseudotracheæ.

MOUTH-PARTS OF THE MONARCH BUTTERFLY.

(Anosia plexippus; order Lepidoptera.)

In the higher Lepidoptera the mouth-parts have undergone a profound modification; mandibles are wholly wanting and the labium is reduced to a small, immovable sclerite, not functional in food getting. The maxillæ, on the other hand, are greatly elongated and are united to form a long, slender, coiling, sucking tube.

In examining the mouth-parts of *Anosia* it will be found necessary to remove carefully the scales and scale-hairs which cover the head before bleaching.

Labrum.—Immovably joined to the clypeus is a very narrow, mostly transversal sclerite, the labrum. It bears two tapering, cephalad-projecting points, the pilifers, rising from

the ends of the transverse portion of the sclerite. Each pilifer bears on its inner margin a row of short bristly hairs, light brown in color. There is also to be made out a very small, triangular piece projecting cephalad from the middle of the transverse portion of the labrum. This is the *epipharynx*.

Mandibles.—The *mandibles* are wanting in *Anosia*. (They are present in an aborted condition in some Lepidoptera; and in one genus of small moths, *Eriocephala*, are present and functional, constituting, with the maxillæ which are not produced into a sucking tube, true biting mouth-parts.)

Maxillæ.—The long, coiling, sucking tube of *Anosia* (as of all the Lepidoptera possessing functional sucking mouth-parts) is composed of the greatly extended, opposed, terminal portions of the *maxillæ*. In addition, there is a fixed basal part of each maxilla, which cannot be divided into *cardo* and *stipes*. This basal part, shining brown, extends caudad and ventrad, partially bounding a cavity lying between it and the labium. The sucking tube consists of two lateral portions, each portion representing a maxilla. These parts are convex outwardly and concave inwardly. By the opposition of the two concave aspects, a complete central tube is formed. The *maxillary palpi* are wanting in *Anosia*, and in most of the butterflies, although present in a one- or two- or even several-segmented condition among most of the moths.

Labium.—The *labium* is a fixed, semi-membranous sclerite, triangular in outline, with its apex projecting cephalad and joining the maxillar proboscis at its base. The *labial palpi* are large, three-jointed, and covered with scales, and normally project cephalo-dorsad. They are inserted on tumid spaces on the base of the triangular labium, and the first joint is pedicellate.

Make a drawing of the cephalic aspect of the head showing the fixed and movable parts.

CHAPTER VII.

THE VENATION OF THE WINGS OF INSECTS.*

INTRODUCTION.

In form an insect's wing is a large, membranous append-age, which is thickened along certain lines. These thick-ened lines are termed the *veins* or *nerves* of the wing ; and their arrangement is described as the *venation* or *neuration* of the wings.

It has been found that the venation of the wings of closely allied insects is very similar, and that great differences in this respect exist between insects remotely connected. Hence, the wings afford excellent characters for use in the classifica-tion of insects. In fact, as slight differences in venation are easily observed, the wings being spread out like an open page, these differences are probably the most available char-acteristics of winged insects for taxonomic work. It is im-portant, therefore, that the student of entomology should learn early in his course the more important facts regarding this subject.

A careful study of the wings of many insects has shown that the fundamental type of venation is the same in all of the orders of winged insects. But this fact is evident only

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* The material for this chapter has been drawn largely from an essay by the writer, entitled *Evolution and Taxonomy*, published in *The Wilder Quarter-Cen-tury Book*, Ithaca, 1893, and from a series of articles entitled *The Wings of In-sects*, by J. H. Comstock and J. G. Needham, published in *The American Naturalist*, vol. xxxii. (1898), and vol. xxxiii. (1899).

when the more primitive or generalized members of different orders are compared with each other. In most of the orders of insects the greater number of species have become so modified or specialized as regards the structure of their wings that it is difficult at first to trace out the primitive type.

NOTE.—The student should have a clear idea of the significance of the terms *generalized* and *specialized*, which are now much used in biology. Generalized indicates a primitive condition, a nearness to ancestral forms. Thus, the most generalized member of a group (as a family or an order) is that member which most clearly resembles the ancient progenitor of that group. Specialized, on the other hand, indicates remoteness from the primitive type, an adaptation to more special conditions of existence. Thus, the most specialized member of a group is the one that departs most widely from the ancient progenitor of that group.

These terms are used in a comparative sense; thus, a highly specialized form may be regarded as generalized when compared with forms that are still more highly specialized.

The specimens indicated for the student to study in the following part of this course have been selected with care to
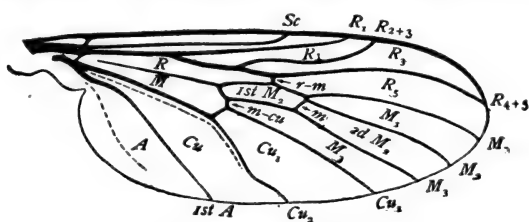
FIG. 2.—Wing of *Rhyphus*.

illustrate gradually increasing degrees of divergence from the primitive type. In the case of each order studied, the work begins with a comparatively generalized form, and passes step by step to those that are more specialized.

The flies of the genus *Rhyphus* afford good examples of comparatively generalized wings. By studying a wing of one of these flies and the accompanying figure (Fig. 2) the

student can gain a good idea of the type of the wings of in-sects belonging to the order Diptera, and have a standard with which to compare wings of insects of other orders.

Longitudinal veins and cross-veins.—The veins can be grouped under two heads : first, *longitudinal veins*, those that normally extend proximo - distad ; and second, *cross-veins*, those that normally extend more or less nearly cephalo-caudad. In Figure 2, three of the cross-veins are indicated by arrows, near the middle of the wing ; two other cross-veins are represented near the the base of the wing, but are not lettered. All other veins represented in this figure are longitudinal veins.

The insertion of the word *normally* in the above definitions is important ; for it is only in comparatively generalized wings that the direction of a vein can be depended upon for determining to which of these two classes it belongs. A little later the student will study wings in which the direction of some of the longitudinal veins has been so modified in the course of specialization that they extend transversely (*i.e.*, cephalo-caudad), and some cross-veins extend in a longitudinal direction (*i.e.*, proximo-distad).

Simple veins and branched veins.—Veins are either simple or branched. The veins lettered *Sc* and *1st A* in Figure 2 are simple veins ; between these there are three branched veins.

In the case of branched veins the entire vein, including all of its branches, is often referred to as a single vein. Thus the third vein in the wing of *Rhyphus*, counting the thickened, cephalic margin of the wing as the first vein, is termed the *radius* or *vein R ;* and by this expression we include both the main stem of the vein and its three divisions. On the other hand, each division of a branched vein is often termed a vein. Thus the first division of the radius, counting from the cephalic margin of the wing, is termed *radius-*

one or *vein R_1*, and the second division, *radius-two* or *vein R_2*, and so on till all are numbered.

NOTE.—In the most generalized flies known to us, the radius is five-branched. But in most flies some of the branches of this vein coalesce so that the number of apparent branches is less than five. In *Rhyphus* veins R_2 and R_3 coalesce so as to appear as a single vein. In order to indicate that this apparently simple vein is composed of two veins, *and in order that homologous veins in different insects shall bear the same designation*, this compound vein is termed *radius-two-plus-three* or *vein R_{2+3}*. In the same way, what appears to be the third branch of the radius in *Rhyphus* is really the fourth and fifth coalesced, and is, therefore, designated as *radius-four-plus-five* or *vein R_{4+5}*. The tracing out of the homologies of the branches of veins is often very difficult; but it is of the greatest importance in determining the relationships of different genera or of families.

Names of the longitudinal wing-veins.—There have been many different sets of names applied to the veins of wings. Not only have the students of each order of insects had a peculiar nomenclature, but in many cases different writers on the same order have used different sets of terms. This condition of affairs was incident to the beginning of the science, the period before the correspondence of the veins in the different orders had been worked out. But now the time has come when it is practicable to apply a uniform nomenclature to the longitudinal wing-veins of all orders; and the following set of terms has been proposed for that purpose:

Costa.—The vein extending along the cephalic or costal margin of the wing is the *costa*.

Subcosta.—Immediately caudad of the costa and extending parallel with it, is a vein, which is usually simple in flies; this is the *subcosta* (Fig. 2, *Sc*).

Radius.—Immediately caudad of the subcosta there is a vein which in generalized insects is always branched; this is the *radius*. In *Rhyphus*, the radius is three-branched (Fig. 2, R_1, R_{2+3}, and R_{4+5}).

Media.—Traversing the middle of the wing there is a longitudinal vein which is always branched in generalized insects ; this is the *media.* In *Rhyphus* the media is three-branched (Fig. 2, M_1, M_2, and M_3).

Cubitus.—The third and last of the branched veins in flies is the *cubitus.* This vein is two-branched in *Rhyphus* (Fig. 2, Cu_1 and Cu_2).

Anal veins.—Caudad of the cubitus there is in *Rhyphus* a single well-developed vein ; this is termed an *anal vein.* As in more generalized insects there are three anal veins, and as this is believed to be the first of the series, it is designated the *first anal vein* (Fig. 2, *1st A*). A rudiment of the second anal vein persists in *Rhyphus ;* this is indicated in the figure by a dotted line, at the left of the letter *A.*

Designation of the longitudinal wing-veins by numbers.—Several writers have designated the longitudinal wing-veins by numbers. In Comstock's *Manual for the Study of Insects* both the names given above and numbers are used. The following table indicates the correspondence of the names and numbers ; and Figure 3 represents the wing of a butterfly, with the veins and cells numbered according to the system used in that manual.

Costa = vein I.	Cubitus = vein VII.
Subcosta = vein II.	1st anal vein = vein VIII. *
Radius = vein III.	2d anal vein = vein IX.
Media = vein V.	3d anal vein = vein X.

It will be observed that in the above table the numbers IV and VI are omitted. At the time the *Manual for the Study of Insects* was published, it was believed that two other longitudinal veins (the so-called *premedia* and *postmedia*) were present in certain orders of insects, and the numbers IV and VI were applied to these veins. It has since been determined that this conclusion was based on an error.

Certain writers number the wing veins without omitting the numbers IV and VI, designating the media as vein IV and the cubitus as vein V.

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* As the anal furrow, which is described later, was supposed to represent the first anal vein, it is numbered VIII in the descriptions of wings of Diptera and Hymenoptera in the work cited, and the true first anal vein is numbered IX.

There are still other writers who do not regard the costa as a true vein, and, therefore, designate the subcosta as vein I.

The result is that there are three distinct systems of numbering the wing-veins, in addition to several old systems which were applied to single orders. It seems better, therefore, to designate the wing-veins by names, and use abbreviations of these names in lettering figures.

Names of the cross-veins.—In the Diptera and in certain other orders of insects there are so few cross-veins that it is practicable to apply names to them ; these are as follows :

The humeral cross-vein.—This is a single cross-vein extending from the subcosta to the costa near the base of the
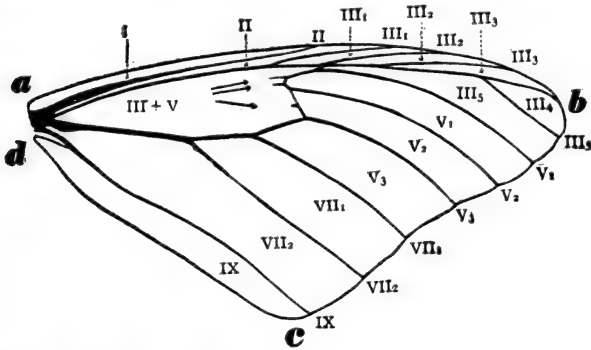
FIG. 3.—Fore wing of a butterfly with the veins and cells numbered.

wing. This is the most constant of all of the cross-veins. It is represented in Figure 2, but is not lettered.

The radial cross-vein.—This is a cross-vein which divides cell R_1. (The cells are defined a little later.) The radial cross-vein is not represented in Figure 2.

The radio-medial cross-vein.—This is a cross-vein extending from the radius to the media, usually near the center of the wing, and is designated by the abbreviation *r-m*. When in its typical position this cross-vein extends from vein R_{4+5} to vein M_{1+2} ; this results in one end being opposite cell R_3 and the other end opposite cell 1st M_2.

The medio-cubital cross-vein.—This is a cross-vein extending from the media to the cubitus, usually near the center of the wing. It is designated by the abbreviation *m-cu*. When in its typical position this cross-vein extends from a point near the base of vein M_{3+4} to a point near the base of vein Cu_1.

The medial cross-vein.—This is a cross-vein extending from vein M_2 to vein M_3 ; it is designated by the abbreviation *m*. The presence or absence of this cross-vein is often a character of considerable taxonomic importance.

The arculus.—In many insects there is what appears to be a cross-vein extending from the radius to the cubitus near the base of the wing. This has been termed the *arculus* by
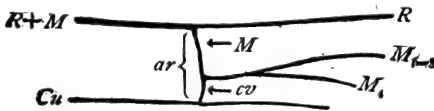
FIG. 4.—The arculus, diagrammatic.

writers on the Odonata, and the use of this term has been extended to all orders in which there is a similar arrangement of the veins in this part of the wing. The arculus is designated by the abbreviation *ar*. Usually when the arculus is present the media appears to arise from it. The fact is, the arculus is compound, being composed of a section of the media and a cross-vein. The structure of this part can be clearly seen in the Odonata (Fig. 4). In *Ryphus* (Fig. 2) the arculus appears as a simple cross-vein extending from the radius to the cubitus, and a part of the base of the media is atrophied.

Designation of the cells of the wing.—The thin spaces of the wings which are bounded by the veins are called *cells*. In descriptions of wings it is often desirable to refer to one or more of the cells. It is necessary, therefore, to have a nomenclature of the cells of the wing, as well as of the wing-veins.

Having named the wing-veins, the simplest possible method of designating the cells of the wing is to apply to

each the abbreviation of the name of the vein that forms its cephalic (front) margin. It should be borne in mind, however, that by modifications of the typical arrangement of the wing-veins, a vein that normally forms the cephalic margin of a cell may apparently bear a very different relation to it ; and this must be taken into account if we are to apply the same term to homologous cells throughout the insect series.

The cells of the wing fall naturally into two groups : first, those on the basal part of the wing ; and second, those nearer the distal end of the wing. The former are bounded by the principal veins ; the latter, by the branches of the forked veins ; a corresponding distinction is made in designating the cells. Thus the cell lying behind the main stem of the radius and on the basal part of the wing is designated as cell R ; while the cell lying behind the radius-one is designated as cell R_1.

It should be remembered that the coalescence of two veins results in the obliteration of the cell that was between them. Thus when veins R_2 and R_3 coalesce, as in *Rhyphus* (Fig. 2), the cell lying behind R_{2+3} is cell R_3, and not cell R_{2+3}, cell R_2 having been obliterated.

When one of these principal cells is divided into two or more parts by one or more cross-veins, the parts may be numbered, beginning with the proximal one. Thus in *Rhyphus* (Fig. 2) cell M_2 is divided by the medial cross-vein into two parts, which are designated as cell *1st* M_2 and cell *2d* M_2 respectively.

The application of this system of naming the cells of the wing is an easy matter in those orders where the wings have few veins ; but in those orders where many secondary veins are developed it is more difficult of application. In the latter case we have to do with *areas* of the wing rather than with separate cells. Thus, for example, it will be seen later that in certain Neuroptera the area R_2 is divided by several

longitudinal veins, which are connected by many cross-veins, the area R_2 (which is strictly homologous with cell R_2) being composed of a large number of secondary cells.

The furrows of the wing.—The wings of comparatively few insects present a flat surface ; in most cases we find that the membrane is thrown into a series of folds or corrugations. This corrugating of the wing in some cases adds greatly to its strength. This is well shown by the wings of dragon flies ; and in most orders the costal margin of the wing is strengthened by a fold between the costa and the radius, the *subcostal fold.* In other cases, the corrugations are the result of a folding of the wing when not in use ; this is well shown in the anal area when this part is broadly expanded.

It rarely happens that there is occasion to refer to individual members of either of these classes of folds, except, perhaps, to the one that has just been designated as the subcostal fold. But there are three other furrows which it is necessary to designate, as they are frequently referred to. These are the anal furrow, the median furrow, and the nodal furrow. Only the first of these is well-marked in the Diptera ; but for sake of completeness all are defined here.

The anal furrow.—This is a longitudinal furrow which is usually between the cubitus and the first anal vein. In the figure of the wing of *Rhyphus* (Fig. 2), it is represented by a dotted line immediately behind the cubitus. In the order Diptera it is always in this position if present.

The median furrow.—This is a longitudinal furrow which is usually between the radius and the media. It is well marked in many of the Hemiptera, where it separates the embolium from the remainder of the corium ; and in the Hymenoptera its course is marked by a series of weak spots (bullæ) in certain veins.

The nodal furrow.—This is a transverse suture beginning at a point in the costal margin of the wing, corresponding to the nodus of the Odonata, and extending towards the inner margin of the wing. It crosses a varying number of veins in different orders of insects.

The furrows of the wing are in no sense homologous or even analogous to veins. More than this, the relative positions of the furrows and of the wing-veins are not constant; for it frequently happens that the course of a vein has been so modified that it crosses the line of a furrow and the relative positions of the two are thus reversed. If this fact had been understood by Adolph we would have been spared his misleading theory of alternating concave and convex veins.*

Margins of wings.—An insect's wing is more or less triangular in outline ; it, therefore, presents three margins ; the *costal margin* (Fig. 3, a-b) ; the *outer margin* (Fig. 3, b-c), and the *inner margin* (Fig. 3, c-d).

Angles of wings.—The angle at the base of the costal margin (Fig. 3, a) is the *humeral angle ;* that between the costal margin and the outer margin (Fig. 3, b) is the *apex of the wing ;* and the angle between the outer margin and the inner margin (Fig. 3, c) is the *anal angle.*

The typical branching of the wing-veins.—In order to determine the homologies of the branches of a forked vein when some of the cells have been obliterated by the coalescence of branches, it is necessary to know what was the primitive type of branching of this vein. For this reason an effort has been made to determine the probable structure of the wing of the primitive winged insect.

In the study of the development of wings, it has been found that in the more generalized insects the cavities of the longitudinal wing-veins are formed about tracheæ, and

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* G. Ernst Adolph, Ueber Insectenflügel. *Nova Acta der ksl. Leop.-Carol-Deutschen Akademie der Naturforscher*, Bd. xli, pp. 215–251. 1879.

that these tracheæ exhibit a striking similarity in their origin and mode of branching in the nymphs and pupæ of the more generalized members of most of the orders of insects. We have been able, therefore, to present a hypothetical type, which we believe represents approximately what was the arrangement of the tracheæ in the nymph of the primitive winged insect (Fig. 5), and, as the wing-veins of the adult are developed about the tracheæ of the wing of the nymph,
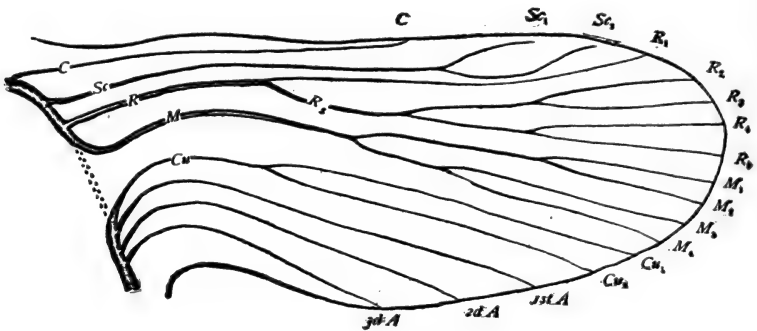
FIG. 5.—Diagram representing the hypothetical arrangement of the tracheæ in a wing of a nymph of the primitive winged insect.

the same figure will serve to indicate the probable mode of branching of the wing-veins in the adult of the primitive winged insect.

The radial sector.—When the radius preserves its primitive mode of branching, it separates at its first fork into two unequal parts ; the first of these is vein R_1 ; the other gives rise to the remaining four branches of the radius (Fig. 5). This second part of the radius, including its branches, is termed the *radial sector* or *vein R_s*.

IDENTIFICATION OF THE WING-VEINS AND OF THE CELLS OF
THE WING IN DIPTERA.

As the chief object of the following work is to give the
student training in tracing the homologies of wing-veins,
comparatively little information will be given directly. The
student will be furnished in each case with the wing to be
studied, and his studies should take the following course :—

Directions for the study of wings.—Make a drawing
of the wing, based upon a careful study of it with a com-
pound microscope, using a low power. The drawing should
be first made with a pencil ; after it has been criticised by
the teacher, the lines should be inked ; ink the lines repre-
senting the media with red ink, use black ink for the re-
mainder of the wing, including the medial cross vein. Make
the drawing on a sufficiently large scale so that each vein
can be represented distinctly ; in most cases the drawings
should be somewhat larger than Figure 3.

Number each vein and cell of the wing.

Note the more important features of its venation, and
especially the more important departures from the primitive
type of the order as indicated by the generalized form first
studied. In the Diptera the wing of *Rhyphus* (Fig. 2) may
be used as a generalized type, although in certain respects
other wings will be found to be more generalized.

The following are some of the more important points to be
noted : Whether the subcosta is simple or forked at the tip ;
the number of the branches of the radius ; in this connection
determine which of the radial cells has been obliterated by
the coalescence of branches of the radius (study Fig. 5) ; the
position of the radio-medial cross-vein ; the number of the
branches of the media (in none of the common Diptera has
the media more than three branches ; and the mode of dis-
appearance of vein M_4 in this order has not been determined

as yet. For this reason the student need not take this vein into consideration in his studies of dipterous wings) ; the division or not of cell M_2 ; the presence or absence of cell M_3 ; the courses of the branches of the cubitus ; the extent of the anal furrow ; and the course of the anal vein.

Wing of a Tabanid.—A specimen of one of the horse-flies, *Tabanus*, will be given the student for examination. Observe the *subcostal fold*, and note that this corrugation stiffens the wing.

Make a drawing of a mounted Tabanid wing, which will be furnished on application to the instructor. Note that in the mounted wing the subcosta is more or less concealed by the radius, although the two veins are distinct, as was seen in the unmounted specimen. Represent these two veins as slightly separated in your drawing.

In the description of this wing, state in what respect it is more generalized than that of *Rhyphus*, and in what respect it is more specialized.

Wing of an Asilid.—A wing of a robber-fly of the genus *Erax* will be used as an example. Note a method of coalescence of veins not exhibited by *Rhyphus*.

Wing of a Bombyliid.—The example used is a wing of *Pantarbes*, one of the bee-flies.

Wing of a Scenopinid.—The wing used is that of a common window-fly, *Scenopinus*.

Wing of an Empidid.—The wing used is that of *Rhamphomyia*, one of the dance-flies.

Wing of a Muscid.—The wing used is that of the common house-fly, *Musca domestica*.

Wings of Dilichopodids.—The wings of two of the long-legged flies will be used. The first belongs to the genus *Psilopus;* the second to the genus *Dolichopus*.

Wing of a Syrphid.—The wings of a fly of the genus *Syrphus* will be used. Note the vein-like structure be-

tween the radius and the media ; this is termed the *spurious vein*.

Wing of Culex.—The wing of a mosquito is used as an example of the venation of the midge-like flies. In these the number of the branches of the radial sector is reduced in a way different from that seen in the families previously stud-ied. Compare with the Asilid, the Bombyliid, and the Sceno-pinid.

THE WINGS OF LEPIDOPTERA.

As the wings of Lepidoptera are covered with scales, it is difficult to determine the nature of their venation without specially preparing them for this purpose. After a student has become familiar with the type of venation characteristic of the order, he can usually determine the course of any particular vein by putting a drop of chloroform on the part of the wing to be examined ; this will render the veins more distinct for a few seconds. Or the scales can be removed from a small part of the wing with a small artist's sable brush. But when a very careful study of the venation of a wing is to be made, it should be bleached and mounted on a card or on a glass slip, in order that it may be studied with a compound microscope. The following is the method of bleaching wings : —

1. Remove the wings carefully so as not to break the fren-ulum if there be one ;* it is well to remove the patagium first.†

2. Dip the wings in alcohol in order to wet them.

3. Immerse them for an instant in hydrochloric acid (muri-atic acid). Use for this purpose dilute acid, one part acid to nine parts water.

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* The *frenulum* is a strong spine or bunch of bristles borne by the hind wing at the humeral angle in most moths.

† The *patagia* are scale-like appendages at the bases of the fore wings.

4. Put them in Labaraque solution with the upper surface of the wings down, and leave them there till the color has been removed from the scales. If a wing bleaches slowly, the process can be hastened by dipping it in the dilute acid and returning it to the Labaraque solution from time to time. This solution can be procured of most druggists. It deteriorates if left exposed in strong sunlight. If it cannot be obtained use an aqueous solution of chloride of lime.

5. When a wing is bleached, put it in alcohol and leave it there till after it floats. This is to wash off the Labaraque solution. The wing can then be mounted on a card. But it is better to mount it as described below.

6. Transfer the wing to a clearing mixture, if it is to be mounted in balsam, and leave it there five or ten minutes. This is to remove any water there may be on it. A good clearing mixture can be made by mixing two parts by measure of carbolic acid crystals and three parts of rectified oil of turpentine.

7. Put the wing on a glass slip with considerable clearing mixture under it to avoid bubbles ; put Canada balsam on top, and cover with a cover glass. In the case of small wings, it is best to transfer them from one solution to another, and to the glass slip by means of a camel's-hair brush.*

Wings bleached and mounted in this way make an important addition to a collection. The slides should be carefully labelled ; and the insect from which the wings were taken should be kept with the slide. It is our practice to remove always the wings from the right side, and then to mount the slide in the collection at the right of the insect from which the wings were taken. Uniformity in this respect adds greatly to the appearance of the collection.

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* In the case of very small wings, as those of Tineids, the very fine veins are more distinct when mounted in glycerine-jelly than when mounted in Canada balsam.

Wings of Hepialus.—Mounted specimens of the two wings of one side of a moth belonging to the genus *Hepialus* will be furnished the student for study. Be very careful of the specimens, as moths of this genus are rare in this country.

The membranous lobe near the base of the inner margin of the fore wing is the *jugum.* This extends under the costal margin of the hind wing, while the greater part of the inner margin of the fore wing overlaps the hind wing. This arrangement assures the acting together of the two wings.

Wings of a Cossid.—Mounted wings of one of the *Cossidæ* will be furnished the student for study.

Wings of the monarch butterfly.—The student will be furnished with specimens of the two wings of one side of the monarch butterfly, *Anosia plexippus.*

Study the fore wing first. Explain the significance of the three short spurs that project into the distal end of the large cell near the middle of the wing.

Wings of frenate moths.—Specimens of moths belonging to the family *Noctuidæ* will be used as illustrations of Lepidoptera in which the frenulum is well developed. Two specimens, a male and a female of the same species, should be studied if practicable.

Remove the wings of the right side of the female, and bleach and mount them.

Figure these wings.

Remove the wings of the right side of the male, and bleach and mount them.

Figure these wings.

Describe the secondary sexual distinction exhibited by the wings of this species.

How could one of these types have been derived from the other?

Study the wings of the male that have not been bleached,

and note the *frenulum hook* on the fore wing, which receives the tip of the frenulum. It may be necessary to remove some of the hairs from the base of the wing. Use a small sable brush for this.

Study the unbleached wings of the female, and note the absence of a frenulum hook.

Compare the hind wing of the monarch butterfly with the hind wings of these moths, and discuss the differences between them.

THE WINGS OF HYMENOPTERA.

The determination of the homologies of the wing-veins of Hymenoptera is very difficult ; as even in the most general-
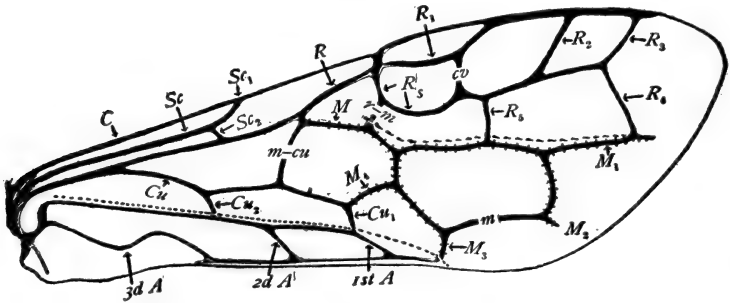
FIG. 6.—The veins of a typical hymenopterous wing.

ized of the living members of the order, the venation of the wings departs widely from the primitive type. For this reason no student should begin his study of the venation of wings with hymenopterous insects. In the following discussion it will be assumed that the student has carefully performed the work on the wings of Diptera outlined in the preceding pages ; and the first step for him to take at this point in his studies is to take his drawings of dipterous wings and review that work.

The method of specialization of wing-veins which has taken place in the Hymenoptera can be most easily seen by a study of the fore wings of certain sawflies. The most useful for this purpose that we have found belong to the genera *Pamphilius* and *Macroxyela*. If we are right in our interpretation of the wings of these insects, there is preserved in each genus all of the primitive wing-veins with a single exception. And, as in each of these genera a different vein is lost, we are able to make a figure of a typical wing from a
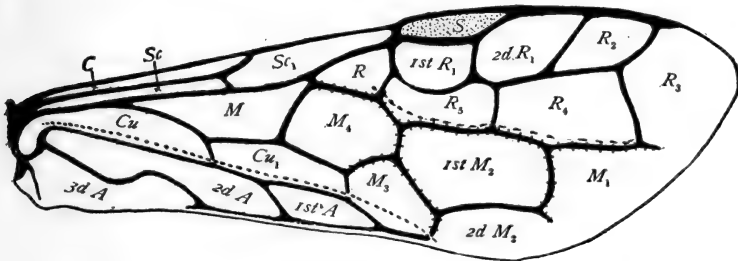
FIG. 7.—The cells of a typical hymenopterous wing.*

study of the two genera. Figures 6 and 7 represent such a wing ; in the former the veins are lettered ; in the latter, the cells.†

In the wings of these sawflies the anal furrow and the median furrow are both well marked, and are in the typical positions ; that is, the anal furrow is immediately in front of the first anal vein, and the median furrow is in front of the

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* The cell lettered S is probably Sc_2. When it is thickened and opaque, as is frequently the case in this order, it is termed the *stigma*.

† Figures 6 and 7 represent the venation of the fore wing of *Pamphilius*, except that vein R_2, which is lacking in this genus, is added. This vein is well preserved in *Macroxyela*, but in *Macroxyela* vein Cu_2 is lost. See Comstock, *Manual for the Study of Insects*, p. 606, for figures of the wings of these two genera.

media. The furrows are represented by dotted lines in the figures.

In the anal area (*i.e.*, that portion of the wing back of the anal furrow) the three typical veins are preserved ; but they coalesce to a considerable extent, both at the base and near the margin of the wing.

In the basal part of the pre-anal area (*i.e.*, that portion of the wing in front of the anal furrow) the stems of the principal veins are as follows : the costa coincides with the costal margin of the wing (Fig. 6, *C*) ; the subcosta (*Sc*) is well preserved and is forked ; back of the subcosta is a strong stem formed by the coalescence of the other three veins ; the cubitus (*Cu*) soon separates from the stem, extending in a curve towards the anal furrow ; while the radius and the media coalesce for about half their length. In order to make these veins more distinct in the figure we have marked the free portion of the media with cross lines.

When we pass from the consideration of the main stems to a study of the branches, we meet a much more complicated problem, a problem which could not have been solved by a study of Hymenoptera alone. But a knowledge of the methods of specialization of the wings of Diptera gives a key to an understanding of the wings of Hymenoptera.

We will study first the branches of the cubitus. Spread out before you your drawings of the wings of the following insects, and arrange them in the order named : a Bombyliid, a Scenopinid, and an Empidid. Now study the figure of a wing of *Rhyphus* (Fig. 2, p. 87) and note that while in *Rhyphus* veins Cu_2 and *1st A* retain their primitive position, in the three wings named above these two veins exhibit varying degrees of coalescence.

A similar method of specialization has taken place in the Hymenoptera, but in this order both branches of the cubitus coalesce with the first anal vein ; and this coalescence has

proceeded so far that both branches cross the anal furrow and end in the anal vein remote from the margin of the wing. (See Fig. 6.)

It should be noted that vein Cu_2 is rarely preserved in this order, even in the more generalized forms. We have found it only in the genus *Pamphilius*. In *Macroxyela** the position of the fork of the cubitus is indicated by a bend in this vein.

If the branches of the media be now examined, it will be seen that vein M_1 (Fig. 6) extends longitudinally near the centre of the distal part of the wing, its primitive course being modified slightly if at all. Vein M_2 follows a course similar to the course of this vein in the Bombyliid ; so also does the medial cross-vein (Fig. 6, *m*). A comparison of the position of cells M_1, *1st* M_2, and *2d* M_2 in the Bombyliid and in the typical hymenopterous wing (Fig. 7) is very instructive.

Returning to *Pamphilius* (Fig. 6), we see that vein M_3 coalesces with the first anal vein, crossing the anal furrow near the margin of the wing. It is evident that the forces that are causing the branches of the cubitus to migrate along the first anal vein and towards the base of the wing are exerting a similar influence on this vein. It is also evident that vein M_4 and Cu_1 coalesce at the tip, and that the migration of the united tips of these veins (marked Cu in the figure) towards the base of the wing has so modified the course of that part of vein M_4 which is still free that this part of this vein extends towards the base of the wing. This change is very similar to the change in the course of vein Cu_2 in the Empidid.

A curious result of this change in the direction of the course of vein M_4 is that the cell M_4 has been closed and

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* Comstock, *loc. cit.*, Fig. 735.

pressed back to the centre of the wing (Fig. 7, M_4), and now lies in front of the free portion of the vein M_4 instead of behind it.*

Let us now consider the courses of the branches of the radius. Here again we can gain help from a study of dipterous wings. Observe in the Bombyliid (*Pantarbes*) the coalescence of the tips of veins R_5 and M_1. In the Hymenoptera a similar coalescence of veins R_5 and M_1 has occurred; but it has proceeded much farther, so that the free portion of vein R_5 in *Pamphilius* (Fig. 6, R_5) is remote from the end of the wing and has the appearance of a cross-vein.

In the Hymenoptera vein R_5 has been followed in its migration along vein M_1 by vein R_4, which has now reached a stage in *Pamphilius* that is quite similar to that reached by vein R_5 in *Pantarbes*. But like vein R_5 it has the appearance of a cross-vein.

From this it will be seen that the vein marked M_1 in Figure 6 is really compound, as it includes the tips of veins R_5 and R_4.

In *Pamphilius* vein R_1 is curved away from the costal margin of the wing to make room for a stigma (Fig. 7, S), and vein R_3 ends in the costal margin a short distance before the apex of the wing (Fig. 6). Vein R_2 has been lost in this genus, but is well preserved in certain closely allied forms,† and is, therefore, represented in the figure.

While the tips of the branches of the radial sector have migrated away from the apex of the wing, the bases of these branches coalesce in the opposite direction; from these

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* At the time that the figures in Comstock's *Manual* were prepared it was believed that the media was typically three-branched. For that reason the vein which we now regard as vein M_4 was believed to be a cross-vein. The interpretation given above accords better with what we have since learned to be the typical form of the media.

† See page 103, footnote.

two causes results the transverse bracing of the radial area of the wing, which is a very characteristic feature of the venation of the wings in this order.

The details of these changes will be made clear by an examination of Figures 8 and 9. The former represents the primitive mode of branching of the radius ; the latter, the radial area of the typical hymenopterous wing (Fig. 6). In the hymenopterous type veins R_{2+3} and R_{4+5} of the primitive
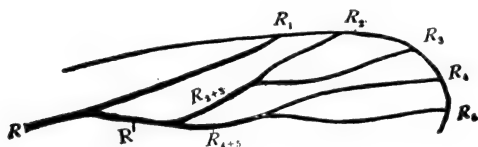
FIG. 8.—The typical radius.
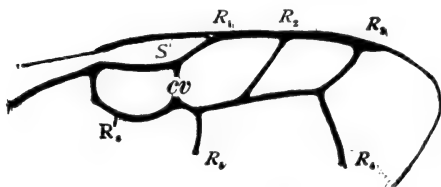
FIG. 9.—The radius in Hymenoptera.

type coalesce so far that the branches of the sector arise from a common stem ; and the tips of all of them have moved away from the apex of the wing, veins R_2 and R_3 following the costal margin of the wing ; and veins R_4 and R_5 following vein M_1.

In the Hymenoptera the radial cross-vein is frequently preserved; it is marked $c\,v$ in figures 6 and 9.

The student who has followed this discussion and has understood it will be prepared to make original investigations of the venation of the wings of Hymenoptera. But no

student should take up the work indicated below before everything in this discussion is clear to him.

Study the fore wings of the insects named below. It is not best to attempt to determine the homologies of the veins of the hind wings at first, owing to the great reduction of wing-veins that has taken place in these wings. The directions for the study of wings given on page 97 will apply here except that Figures 6 and 7 instead of Figure 2 will be used for comparison. Indicate the media with red ink.

The fore wings of Tenthredinids.—The wings of two sawflies will be used as examples of different degrees of specialization :

> (a.) *Macroxyela.*
>
> (b.) *Pteronus.* The currant sawfly.

The fore wing of a Siricid.—The best genus of the Siricidæ for this study is *Sirex.* The more common, *Tremex,* is more difficult.

The fore wing of a Sphecid.—Use for this purpose a wing of the common mud-dauber, *Pelopœus.*

The fore wing of an Ichneumon-fly.—A species of *Ichneumon* will furnish an example.

The fore wing of a Braconid.—Compare with the wing of an Ichneumon-fly. Color vein M_1 red and the medial cross vein black.

The fore wing of the Honey-bee.

THE WINGS OF NEUROPTERA.

The foregoing studies of the wings of Diptera, Lepidoptera, and Hymenoptera have shown that in none of the genera studied has the number of the branches of any vein exceeded that of our hypothetical type (Fig. 5), and that in none of them have all of the branches of all of the veins re-

mained distinct. These wings illustrate, therefore, what has been termed *specialization by reduction.*

'There are, however, several orders of insects in which, as a rule, some of the wing-veins have a greater number of branches than in the hypothetical type. These illustrate what has been termed *specialization by addition.*

Among the orders in which ·specialization of wing-veins by addition has taken place is the Neuroptera ; and several genera of this order will be studied as examples of this method of specialization. We will, however, first point out the more important features of specialization of wing-veins by addition.

In speaking of an increase in the number of veins, reference is made only to a multiplication of the branches of the principal veins. In no case is there an increase in the number of principal veins. And this increase in the number of branches may be confined to one or two of the principal veins in the preanal area, while the number of the branches of some of the other veins may be reduced, the expanding of some parts of the preanal area resulting in a crowding of other parts. In some cases we will find that the multiplication of wing-veins extends to the anal area also ; in others we will find the anal area greatly reduced. But even in those cases where the anal area is reduced, the total result has been the production of a many-veined wing.

In the many-veined wings both the longitudinal veins and the cross-veins are increased in number. In most cases where there are many cross-veins it is impracticable to distinguish from others those particular cross-veins to which we applied special names in describing the few-veined wings. (See page 91.) But in the case of the longitudinal veins it is necessary to distinguish the primitive veins, that is, those of our hypothetical type, from the veins that have been developed in addition to these. For if this is not done it will

be impossible to point out the changes that have taken place in the course of the development of each of the various types of many-veined wings. We therefore apply the term *accessory veins* to these secondarily developed longitudinal veins, and retain the same nomenclature for the primitive veins that we used in describing the few-veined wings.

The development of accessory veins.—Accessory veins may be borne by any of the primitive longitudinal veins ; and they may arise from either of the two sides of such a vein. In most cases it is unnecessary to designate the individual accessory veins, as, usually, it will be sufficient for descriptive purposes to indicate the number of these veins that have been developed upon a particular longitudinal vein. In fact, in certain cases more than this could not well be done, owing to the irregularity of the veins. On the other hand, in many cases the accessory veins borne by a single primitive vein present a high degree of regularity, and it is evident that they have been developed in a regular sequence. Under these circumstances it is practicable to designate them individually ; and we have devised the following method for this purpose.

The accessory veins arising from one side of a single primitive vein are considered as a single set, and to each set of veins a distinct set of numbers is applied, beginning with the oldest (*i.e.*, the first-developed) member of the set.

By this method homologous veins, when a homology exists, will bear the same number. But it should be remembered that as accessory veins have arisen independently in many different groups of insects, it often happens that accessory veins similar in position, and bearing the same number in our system, are merely analogous and not homologous.

In order to apply this system it is necessary to know, in the case of each group of insects studied, the sequence in which the members of the particular set of veins under con-

sideration have been developed. For additions to such a set of veins may be made to the distal end of the series, or to the proximal end, or may be interpolated at some distance from either end.

We will illustrate these principles by a figure of the wing of a nymph of a cockroach (Fig. 10). We do not use for this purpose a figure of a wing of a neuropterous insect, as that would lessen the value, as a means of training, of the study of neuropterous wings which the student is to make. A
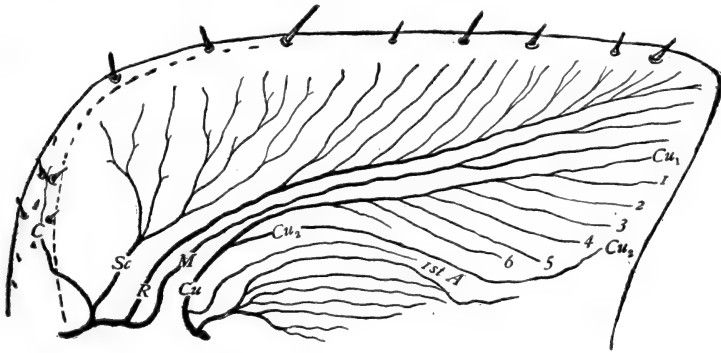
FIG. 10.—Hind wing of a nymph of a cockroach.

nymph is used here, because in the many-veined insects the longitudinal veins, both primitive and accessory, are developed about tracheæ ; and it is much easier to determine the homologies of the tracheæ of an immature wing than it is to determine the homologies of the wing-veins of the adult. And, too, in this way we are able to eliminate the cross-veins which are not preceded by tracheæ in the cockroaches.

Accessory veins added distally.—If the radial trachea of the nymph of a cockroach be examined (Fig. 10, *R*), it will be seen that it bears many branches on its cephalic side ; each of these branches is enclosed by a vein in the adult state. The number of these veins is much greater than that of the

hypothetical type. A comparison of the wings of different species of cockroaches shows that the increase in the number of the branches of the radius has been greater in some than in others ; and a study of the one figured indicates the method of increase, which is doubtless as follows : the branches have been added, one after another, to the tip of the trachea, R, there being a migration of the base of each accessory trachea towards the base of the wing, thus making room for the addition of new branches. In this case the first accessory vein is the proximal one ; but in this particular instance it is impracticable to number the accessory veins, owing to a splitting of some of them which is taking place.

Accessory veins added proximally.—A study of the cubital trachea of the nymph of the cockroach figured above (Fig. 10, *Cu*) shows that accessory veins are being developed on the caudal side of vein Cu_1. It can also be seen that the distal members of the series of accessory tracheæ are well-developed, and that the growth of additional ones is taking place in the disk of the wing at the proximal end of the series. In this case the first accessory vein is the distal one, and the accessory veins should be numbered as indicated in the figure.

Accessory veins interpolated.—Reference has been made above to a splitting of some of the accessory tracheæ developed on the cephalic side of the radial trachea in the cockroach under consideration (Fig. 10). As a vein is developed about each of the divisions of these tracheæ, it is evident that at the same time that accessory veins are being added distally to this series others are being interpolated. In cases of this kind it is impracticable to number the members of a series of accessory veins.

The suppression of the dichotomous branching of veins.—In the more highly specialized of the many-veined

insect wings there exists a type of branching which is very different from that of our hypothetical primitive type. An examination of Figure 5, which represents the latter, will show that in every case the forked veins are branched dichotomously, while in the many-veined wings the more characteristic type of branching results in the formation of pectinate veins; this pectinate type of branching is well shown by the cubitus in a cockroach (Fig. 10 *Cu*).

The changes that take place in the development of the pectinate type of venation from the dichotomous type are of two kinds: first, the development of accessory veins; second, the modification of the primitive veins so that they are no

Fig. 11.—Diagrams of several types of radius.

longer dichotomously branched. The former has been discussed above; we will now briefly refer to the latter. For this purpose we will give a series of diagrams illustrating

several types of branching of the radial sector. Each of these figures represents a condition which exists in some insect.

Figure 11a represents the typical or dichotomously branched radial sector. Figure 11b represents a typical radial sector with the addition of some accessory veins on the caudal side of vein R_2. In this case the radial sector is nearly pectinate, but not quite so, owing to the forked condition of vein R_{4+5}. In the form represented by Figure 11c veins R_4 and R_5 coalesce to the margin of the wing; and in this way the pectinate type is attained. In another insect (Fig. 11d) the pectinate type has been attained by fission instead of coalescence. Here veins R_4 and R_5 have split apart till vein R_5 arises from the main stem of radius.

When many cross-veins are present, the dichotomy of the branching of the sector may be suppressed in still another way, by the transference of the base of vein R_4 to vein R_{2+3}. All stages of this switching of the base of vein R_4 occur in the ant-lions; and two of them are represented by Figures 11e and 11f.

In the genus represented by Figure 11e the base of vein R_4 appears to be forked; one arm of the fork arising from vein R_5, the other from vein R_{2+3}. The former is the true base of vein R_4; the latter is a cross-vein which is assuming the function of a base of this vein. In the genus represented by Figure 11f the switching has been completed, vein R_4 arising from vein R_{2+3}.

In the foregoing illustrations comparisons of allied insects have been made in order to determine the ways in which the wings are being modified; frequently a comparative study of the fore and hind wings of a single insect is equally suggestive, for it often happens that the two pairs of wings exhibit different degrees of the same kind of modification, and thus the course of the change is indicated.

A study of the causes of the changes which we are de-
scribing is beyond our present purpose, which is merely to
determine the homologies of the wing-veins. But we can
gain a hint of the probable reason for the development of
the pectinate type of veins without entering very deeply into
questions of the mechanics of flight.

It is obvious that many styles of flight exist among in-
sects, and that for the different styles of flight different
kinds of wings are required. In *Corydalis*, which will be
studied a little later, it can be seen that the wing is stiffened,
along a line parallel with the costal margin of the wing, by
the subcosta, the main stem of the radius, and veins R_1 and
R_2. Back of this line there is a broad, flexible area, which
bends up during the downward stroke of the wing, forming
an inclined plane, the pressure of which against the air
forces the insect ahead. The flexibility of this area of the
wing is increased by those changes which result in the for-
mation of the pectinate type of branching.

The wings of Sialis.—Among our more common neu-
ropterous insects those of the genus *Sialis* have retained the
most generalized condition of wing-venation. It is sug-
gested, therefore, that the student begin his practice in de-
termining the homologies of the wing-veins of Neuroptera
with a member of this genus. Make a drawing of the wings
and letter the principal veins, and the accessory veins.

The wings of Chauliodes.—

The wings of Corydalis.—In this genus the radial sec-
tor is an example of a high development of the pectinate
type of venation.

The wings of Ant-lions.—Photographs of the wings of
representatives of several genera of the Myrmeleonidæ will
be furnished the student for study. The photographs repre-
sent the wings greatly enlarged and are, therefore, much
more available for study than the actual wings. Each

photograph is labeled with the name of the genus it repre-sents.

Spread out the photographs before you and study the costal area of the wings : the costal area is the area between the costa and the subcosta ; it corresponds to cell C of the few-veined insects. Observe the varying degrees of complexity of the cross-veins ; observe also the stiffening of the area opposite the point where veins Sc and R coalesce, and compare it with the stigma in the Hymenoptera.

Study the radial area of all of the wings, determine the homologies of the branches of the radius in each, letter them, and number the accessory veins. Also indicate with pencil marks the way in which the suppression of the dichotomous branching has been brought about.

That part of the wing of these insects lying caudad of the radius presents difficulties that could hardly be solved by a study of adults alone. A study of the development of the wings of an ant-lion has shown that the second of the two principal branches of the media and vein Cu_1 coalesce. Study the fore wings, which are less modified than the hind wings, and observe that the media appears to be a simple vein for the greater part of its length, and that the cubitus exhibits a well-marked fork before the middle of the wing. This fork is the point of separation of veins Cu_1 and Cu_2. Observe that near this forking of the cubitus there is what appears to be an oblique cross-vein extending from the media to the cubitus ; this is not a cross-vein, but is the beginning of vein M_{3+4}, which coalesces with vein Cu_1 for the greater part of its length. Letter the branches of the media and of the cubitus on each photograph. After this work has been criticised, trace the course of media with red ink and tne courses of radius and cubitus with black ink, omitting tne accessory veins.

THE TRACHEATION OF THE WINGS OF NYMPHS AND OF PUPÆ.

It has been found that, in the course of the development of the wings of the more generalized insects, the tracheæ which traverse the principal veins are developed before the veins appear, and that later the veins are developed about these tracheæ. It is evident, therefore, that much light can be thrown upon questions regarding the homologies of wing-veins by studies of the tracheæ which precede them ; and the following suggestions are given to aid students who wish to make such studies.

If a living pupa or nymph be placed in formol (4%) the tissues of the wings will be rendered translucent in a short time. In the case of very delicate insects only a few hours are required for this, but with larger ones with more opaque wings it is necessary to leave them in the formol for several days, or even for several weeks. While the formol renders the tissues translucent, it does not soon penetrate the tracheæ, which are, therefore, left filled with air, and appear as dark lines when the wing is examined with transmitted light. Just after molting some wings are translucent, but there are few so clear that a short stay in formol will not make them clearer.

In order to study wings prepared in this way, they are removed from the body and mounted in glycerine-jelly, care being taken to cool the mount quickly so that the jelly will not penetrate the tracheæ. In this way most beautiful objects can be prepared, which will show the minutest ramifications of the tracheæ.

Not only can the tracheæ that precede the wing veins be studied in this manner, but, if the wings be taken at the right stage, the forming veins will appear as pale bands when viewed by transmitted light. This is due to the fact

that at this time the veins are merely cavities, filled with lymph, and are more translucent than the spaces between them, which are occupied by tissue.

Unfortunately, however, this distinction is only temporary in most specimens. As a rule, the entire wing becomes transparent in a few hours after it is mounted in the glycerine-jelly. It is necessary, therefore, to make drawings or photo-micrographs promptly, in order to keep a record of the courses of the veins.

On the other hand, the tracheæ, as a rule, stand out more sharply twenty-four hours after mounting, because of the clearing effect of the glycerine-jelly upon the tissue of the wing. But the making of drawings or photo-micrographs of the tracheæ should not be delayed long ; for the tracheæ soon become filled with the jelly, and are then practically invisible.

The preparation of specimens.—Collect living nymphs or pupæ, place them in formol (4%), and leave them for a time, as indicated above. The formol will make the wings of the insects more translucent ; but it will not remove dark colors from chitin. It is well, therefore, to select, at first, the paler species for study.

When ready to mount a wing, spread a drop of melted glycerine-jelly on a slide and allow it to cool.

Dissect off the wing to be studied, taking with it just enough of the thorax to include the basal attachments of the tracheæ. The dissection may be made under water ; but the wing should be removed from the water promptly, so that the tracheæ may not become filled with water.

Place the wing upon the solidified glycerine-jelly on the slide ; and lower upon it a heated cover-glass, which will cause the jelly to melt enough to envelop the wing.

Cool the mount quickly on ice, a marble slab, or some other cold object. Rapid cooling is imperative, for in melted glyc-

erine-jelly the tracheæ soon become filled, and the smaller ones are then invisible.

It is imperative, also, that the wings be handled with care. Being sac-like structures, the tracheæ are almost free within them, and a slight pinch with forceps in the middle of the wing may throw all of its tracheæ out of place. It is better to lift the wing by its thoracic attachments or upon a section lifter.

Not every nymphal wing is fitted for this study. Just before molting, and especially just before the last molting, the wing becomes so crumpled. within its old sheath that the course of its tracheæ can be followed only with difficulty.

The method of study.—So far as is practicable, the studies of the tracheation of the wings of nymphs and of pupæ will be original investigations. For this reason, no particular species is suggested for study. The student will select the most available material, and will endeavor to make an addition to our knowledge of this subject. The series of articles in the *American Naturalist*, already referred to (see page 86, note), may be used as a work of reference. A few suggestions, drawn from the studies upon which those articles were based, are given here :—

Among the more available subjects for the beginner in this line of work, are the pupæ of moths and the nymphs of Orthoptera. The former illustrate specialization by reduction ; the latter specialization by addition. During the winter, when it is difficult to collect Orthoptera, the nymphs of stone-flies (Plecoptera) may be used instead. These can be found under stones in the beds of streams.

In many insects the costal trachea is wanting or is but slightly developed. It does not follow, therefore, that the trachea nearest the costal margin of the wing is the costal trachea. In most cases, the radial trachea can be identified easily, and it will serve as a starting point for the

determination of the homologies of the other principal tracheæ.

In most orders of insects the longitudinal veins can be distinguished from the cross-veins by the fact that the cross-veins are not preceded by tracheæ.

In some of the many-veined insects, as Odonata, the cross-veins, as well as the longitudinal veins, are preceded by tracheæ ; there being, in these insects, a great multiplication of tracheæ.

On the other hand, in the Trichoptera, Diptera, and most Hymenoptera, a great reduction of the tracheal system has taken place. It is not well, therefore, for the student to begin his studies of this subject with members of either of these orders.

In those orders where a specialization of wing-veins by addition has taken place, the accessory longitudinal veins are preceded by tracheæ.

Finally, it should be remembered that it is not safe to base conclusions upon the study of a single insect ; a large series, representing as many genera and families as is practicable, should be investigated.

CHAPTER VIII

METHODS OF INSECT HISTOLOGY.

INTRODUCTION.

The study of the gross anatomy of most insects requires no more in the way of equipment than a simple dissecting outfit,—viz., scalpels, scissors, needles, and dissecting tray; and little more in the way of method than skilful manipulation of these simple tools. But for the study of the anatomy of very small insects, and more especially for the study of the histology of insect organs, the character of the various body-tissues, the histolytic and histogenetic phenomena accompanying the post-embryonal growth of insects with complete metamorphosis, and, finally, for the study of insect embryology, a more elaborate equipment is required, and some knowledge of the special methods of the animal histologist is needed.

Insects present a special problem in histologic method because of the always present covering of chitin which protects their bodies. This chitinous wall hinders very effectively the penetration of the fixing fluids and besides presents very serious difficulties in section cutting. Hence even the student already familiar with the usual methods of animal histology needs some special guiding in beginning the study of insect histology.

The following notes are intended to furnish a guide for beginning students of insect histology and development.

For advanced students, needing a knowledge of more special methods, Lee's *Microtomist's Vade-Mecum* (5th edition, 1900), and the notes of "technic" given in the many papers treating of insect histology and development will furnish valuable suggestions. All of the general methods of killing, fixing, staining, etc., given in the following notes have been personally used by the authors in studying insects. For those few special cases of treatment of particular tissues and organs introduced, which have not been tried by the authors, an authoritative reference to the use of the method is always given.

It is hardly necessary to say that within certain limits there is a great deal of difference in the practice of microtomists in the matter of method. In these brief notes it is intended to outline a simple, straightforward course of procedure which has been found by actual work on insect tissues to give good results in such work. Even this plan has been found difficult to adhere to, because a certain amount of variation is sure to enter into the practice of any worker. The student will find out for himself, however, as he gets more and more familiar with the work, the advantages of variation, and will be able to take advantage of the opportunities for modifying his practice presented by the many (at first glance, confusingly many) suggestions of the standard manuals of microtomic technic. The following three books will be of special value for reference.

Lee, A. B.: *The Microtomist's Vade-Mecum*, 5th edition, 1900.

Whitman, C. O.: *Method of Research in Microscopical Anatomy and Embryology*, 1885.

Gage, S. H.: *The Microscope*, 7th edition, 1901.

For general histology.—Any organs or tissues desired for study should be dissected out of a specimen which has been made insensible by chloroform or ether or cocaine or which has just been killed by some quick process. The point is to get the material in a fresh and perfectly normal condition. The tissue must be alive at the time it goes into the fixing fluid. A certain way of accomplishing this is to kill the insect by dropping it into the fixing fluid. But that the tissue may certainly be reached and affected by the fluid, the specimen, unless small and with very weakly chitinized integument, must be cut open.

For studying anatomy or development.—It is often desirable to make serial sections of the whole body of an insect. In the case of insects too small to dissect this is the only means of studying their anatomy. In the study of the post-embryonal development of insects which have complete metamorphosis, the advanced larval and the pupal stages must be examined by such sections, because of the great breaking down of many of the body-tissues. Specimens should be killed immediately after moulting, when the outer chitin wall is thinnest and most pervious to the fixing liquids. By rearing the specimens, just moulted larvæ, just formed pupæ, and just issued imagines may be obtained, and the use of such specimens will be found to be immensely advantageous. Strongly chitinized specimens, which must be sectioned, may be carefully and cleanly cut into two or three parts, say, head, thorax, and abdomen. This will enable the fixing fluids to penetrate the body cavity, although the difficulty of cutting the firm chitin wall with the microtome knife still remains. Certain methods of softening the chitin are used by some workers, but there is always danger in the practice of injuring the other more delicate tissues.

For embryology.—In obtaining insect eggs it is highly desirable, often imperatively necessary, to know the exact age of the specimens. Hence it is desirable to capture females of the species to be studied, and if possible have the eggs laid in captivity. Thus the exact time of oviposition, and consequently the age of the specimens studied, can be known. For work simply illustrative of the embryologic development of insects, eggs should be chosen which are large, and are not spherical, but of such shape that the specimens can be readily oriented at the time of imbedding or cutting.

PREPARATION OF MATERIAL FOR SECTIONING.

The sequence of treatment.—The fresh, live material (bits of tissue, organs, whole insects, or eggs), which is to be studied by means of sections, must undergo certain treatment necessary to preserve it, and to prepare it for cutting. It has first to be *killed* and *fixed ;* next to be *hardened ;* next to be *cleared ;* and finally to be *infiltrated* with and *imbedded* in *paraffin* (or collodion, see note later). (See also Recapitulation, p. 138.)

Killing and fixing.—Out of the bewildering array of fixing agents, the student of insect histology must choose a few which are characterized by great penetrating power. Many of the commonly used agents for fixing animal tissues in general cannot be used in work with insects (except where dissected-out, non-chitinous organs or tissues are being handled), because of their inability to penetrate the chitinized integument.

In fixing, a relatively large amount of the fixing agent should be used, say at least 50 times the volume of the specimens. The objects should sink, if not at first, then later, in the fixing fluid. If the objects persist in floating they probably contain air, and the tissues adjoining air bub-

bles will not be fixed. The use of heat as suggested in a later note may expel the air.

•It is our practice to divide our material into at least two similar lots, three or even four if there are plenty of specimens, and to use a different fixing agent for each of these lots. For one lot use a sublimate solution, for another a chromic acid solution, and so on. This not only gives a variety of fixation, but insures against total loss in the event of accident to one of the lots.

Boiling water.—Drop the live insects into boiling water, or put them into a glass beaker and pour boiling water over them. Leave the specimens in the hot water, without further heating it, for two to five minutes. Then remove to 30% alcohol for three hours, then to 50% alcohol for three hours, then to 70% alcohol for twelve hours, finally into 85% alcohol for keeping. Or the insects may be killed in hot water and then removed to any one of the fixing agents described in the following paragraphs. This is probably the best way of handling whole insects. The method of primarily fixing by boiling water is especially valuable for eggs and pupæ, which, owing to the impervious character of their chitin-armor, can hardly be fixed in any other way, but we find that it gives excellent results in almost all general work.

Picro-aceto-sublimate.—This useful fixing agent is made by taking of a cold saturated solution of picric acid in 70% alcohol, one part ; of hot saturated aqueous solution of corrosive sublimate, one part; and a few drops (½ to 1%) of glacial acetic acid. The fresh tissue or live insect should be dropped into this fluid and allowed to remain for from twelve to twenty-four hours (incline toward the longer time). Remove to 70% alcohol for twenty-four hours. (The alcohol will slowly remove the picric acid, and should be changed two or three times. If warm alcohol is used the removal of picric acid will be more speedily accomplished.) Then remove to

85% alcohol, changing once or twice if the alcohol becomes discolored. It is not necessary to remove all the picric acid before proceeding to the further preparation of the specimens.

Picro-sulphuric acid.—Distilled water 100 parts, sulphuric acid 2 parts, picric acid as much as will dissolve. Treat specimens as for picro-aceto-sublimate.

Mercuro-nitric mixture.—Distilled water, 400 c.c.; alcohol, 60% strength, 50 c.c.; nitric acid, 46% strength, 7 c.c.; glacial acetic acid, 2 c.c.; corrosive sublimate, 9 grams. Leave specimens in this mixture for from three to twelve hours; twenty-four may be better in some cases. Wash in 70% alcohol and remove to fresh 70% alcohol, to which a small amount of tincture of iodine has been added to hasten the removal of the corrosive sublimate; leave in this solution for two or three days and then transfer to 85% alcohol for indefinite keeping.

Chromo-nitric acid.—Alcohol, 70% strength, 3 parts; nitric acid, 10% strength, 4 parts; chromic acid ½% strength, 3 parts. Leave specimens in this mixture four to eight hours, and wash in 70% alcohol. Leave in 70% alcohol for twenty-four hours, and remove to 85% alcohol for keeping.

Penetration can be more certainly effected if the fixing fluid is used warm. Where the killing is done in the laboratory and the fluid can be readily slightly heated, it is well always to do it.

In addition to the fixing agents of which formulæ have been given, other fixing agents, a host of which are described in chapters IV. and V. of Lee's *Vade-Mecum,* may be used. For cytological work, the osmic acid agents, such as *Flemming's chromo-aceto-osmic acid* (Lee's *Vade-Mecum,* p. 40), and *Hermann's platino-aceto-osmic acid* (Lee's *Vade-Mecum,* p. 45), should be used.

Hardening.—After killing and fixing and washing the specimens are kept in alcohol of 85% strength. The specimens may be kept thus indefinitely. Specimens which are to be prepared for immediate cutting must now be hardened. (If the specimens are to be stained *in toto*, the staining should now be done before removal to the 95% alcohol. For directions, see p. 137.) Remove the specimens to 95% alcohol and leave for twenty-four to forty-eight hours, depending upon size of specimen and degree of chitinization of integument. Then remove to absolute alcohol for from twenty-four to forty-eight hours. The specimens, if properly fixed, will now be thoroughly hardened.

Clearing.—(For theory and practice of clearing, see Lee's *Vade-Mecum*, p. 64 *et seq.*) Remove the specimens from absolute alcohol to a vial which has been half-filled with xylol and then filled with absolute alcohol. Put the specimens into the vial and leave them for twelve to twenty-four hours. Then remove to a vial filled with xylol and leave them until cleared. This will require from twelve to twenty-four hours.

We have used cedar-wood oil a great deal for clearing, as follows: Remove the specimens from absolute alcohol to a mixture of absolute alcohol, one part, cedar-wood oil, one part, for twenty-four hours. Then remove to pure cedar-wood oil for twenty-four hours.

Chloroform, which has been much used, is rather decried by the present-day workers. For insects it should be avoided, as it has little penetrating power.

Infiltrating with paraffin, and imbedding.—After clearing, the specimens are to be infiltrated with melted paraffin. The specimens should be removed from the vial of cedar-wood oil or xylol into a small dish (watch-glass, beaker, evaporating dish, casserole, tin "pattie" dish), into which part of the cedar-wood oil or xylol from the vial is

poured. Place this dish on top of the paraffin* oven (a water-bath oven heated at a constant temperature, a little above the melting point of the paraffin used) and drop into it several small pieces of paraffin. This paraffin will slowly dissolve in the cedar-wood oil or xylol and this mixture will penetrate the specimens. After the paraffin is all dissolved, add more paraffin and remove the dish to the interior of the oven. Leave here for from three to six hours, depending on size, and density, of chitinous integument. Then remove the specimens to a similar small dish of melted pure paraffin and keep in the oven for from six to twenty-four hours. The kind of paraffin to use depends largely upon the character of the specimens. For very delicate specimens, a paraffin of low melting point is desirable. But for most insect work a hard paraffin is desirable, say paraffin of melting point of about 55° Centigrade. In the summer, hard paraffin must be used for the sake of ease in cutting.

Another common method of infiltrating with paraffin is to remove the specimens from pure xylol or cedar-wood oil to a mixture (prepared beforehand) of one-half paraffin and one-half xylol or cedar-wood oil. Leave in this mixture, in the oven, for from three to six hours, and then remove to pure melted paraffin and leave, in the oven, for from six to twenty-four hours.

After the objects are thoroughly infiltrated with pure paraffin they must be imbedded. Make a small oblong paper tray (see Lee's *Vade-Mecum*, p. 91) and pour into it a little melted pure paraffin; when the paraffin has thickened a little by cooling, so that the objects when put into it will

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* The student must get acquainted with the paraffin oven from an inspection of that oven which happens to be in use in the laboratory. With much variation in size, shape, and elaborateness of make, all the different ovens in use are essentially simply water-baths with an automatic thermostat for maintaining a constant temperature.

not sink to the bottom and thus have too thin a layer of paraffin below them, put them in, with more melted paraffin, and with a warmed dissecting needle arrange them in position, *i.e.*, not too near each other, and oriented with regard to the sides of the paraffin block (that is to be). Cool the paraffin quickly by floating the tray on cold water (use ice water in summer), and, as soon as a sufficiently firm film has formed over the top of the paraffin, by plunging the tray into the water. In a minute or two the block will be hard enough to permit of the unfolding and removal of the enveloping paper. Leave the paraffin block in the cold water for ten or fifteen minutes at least, to harden thoroughly. For very small specimens the imbedding can be done in a watch glass.

The specimens are now imbedded, and may be preserved thus indefinitely. On a large piece of paper write a full label, *i.e.*, name of insect and character of specimen, locality and date of capture, fixing agent employed, and other details of preparation that may be valuable to know, and wrap up the paraffin block in this label. Such labelled and wrapped up blocks can be conveniently kept in small wooden or pasteboard boxes, labelled briefly outside with name of the enclosed specimens.

Instead of paraffin, collodion or celloidin is sometimes used for infiltrating and imbedding. It is used especially, however, for very large objects, *i.e.*, objects more than one-half inch in diameter. For small objects paraffin is better because the sections can be cut thinner. For most insect work paraffin is far the better medium. (For details of the collodion method see Lee's *Vade-Mecum*, p. 120 *et seq.*, and Gage, p. 157 *et seq.*)

Section-cutting.—The work of cutting sections with the microtome can best be learned by the student by seeing such work done. The differences in construction of microtomes

with the varying details of the object-carriers, knife-carriers, etc., make it impossible to attempt here to instruct the student in the details of microtome manipulation.

For cutting insect specimens, especially whole bodies with their chitinous outer wall, extreme solidity and rigidity of the microtome in all its parts is absolutely necessary for accurate cutting. The heavy, solidly made, sliding microtomes of the general character of the Thoma-Jung models are, in our opinion, the microtomes best adapted for general insect work. The cutting is not done so rapidly as by the wheel microtomes, but the sureness is an over-balancing item. Short ribbons, as long as the knife is broad, can be cut with the sliding microtomes and transferred directly to the slide.

The specimen is cut as a small block out of the larger paraffin block containing it, and mounted on the object-carrier of the microtome. The block to be cut should be rectangular in outline, and be set with that margin which will first strike the knife parallel with the knife. All screw adjustments, such as those holding the knife and the object, should be tightly made, and the block containing the object should be immovably fastened to the object carrier. The sections should be, for general work, from 5 microns to 10 microns thick. Except in the case of eggs or delicate tissue, it is rarely necessary to cut thinner than 5 microns.

The student will undoubtedly meet with more or less trouble in section-cutting. If it is too warm in the laboratory the sections may stick to the knife ; if too cold, they may roll or break. Rolling of sections is the commonest trouble in cutting. It is often apparently impossible to get that proper temperature or condition which will prevent rolling. There are various expedients for straightening or hindering the rolling of sections that tend to roll. For details and suggestions regarding section-cutting, see **Lee's** *Vade-Mecum*, p. 108 *et seq.*

After the sections are cut, they may, if in ribbons, be kept on sheets of paper for some time before fastening to the slides, but it is always advisable for fear of accident whereby the sections may become lost or disarranged (disarrangement of serial sections is almost as fatal as loss) to fasten them immediately on to the glass mounting slides.

<center>TREATMENT OF SECTIONS.</center>

The sequence of treatment.—The sections have first to be *arranged* and *fastened* on the *slide*, and have the *paraffin removed* from them ; then they have to be *stained*, and finally *cleared* and *mounted*. This is the sequence of treatment for sections which have been cut from a specimen not stained *in toto*. If the specimen has been stained before cutting, the sequence is, (1) arranging and fastening on slide, (2) removal of paraffin, (3) clearing and mounting. (See also Recapitulation, p. 138.)

In the work of removing paraffin, staining, and clearing it is necessary that the sections fastened on the slide be thoroughly bathed by various fluids. The best way of accomplishing this is to have the fluids in small glass jars. These jars should be of a diameter slightly greater than the width of the slides used and of a height slightly greater than the length of the slide. They should have closely fitting covers to prevent evaporation, and should be heavy and firm enough not to be too easily knocked over. The so-called Naples jars, or the Harvard staining jars, or the Stender dishes of various dealers are of the proper kind. Ten or twelve of these jars containing the various necessary fluids, *i.e.*, xylol, absolute alcohol, alcohol of various strengths, staining mixture, clearing mixture, etc., properly labelled, should be arranged on the table in an order corresponding to the successive treatment required by the sections.

Arranging and fastening sections on the slide.—
If serial sections are being cut and the series is long and the
sections of considerable size, larger slides than the ordinary
3 in. by 1 in. slides can be used to advantage. For most
work, however, the smaller slides do very well. The sec-
tions are to be arranged in regular order in successive lines
either parallel with the short or long axis of the slide as
preferred, leaving uncovered only enough room at one end
of the slide for a suitable label. The rows of sections should
be near together, and the sections near together in each
row, *i.e.*, the paraffin block should, before cutting, be·
trimmed as small as is safe. This economizes slides and
covers, and makes the series far more compact and therefore
more readily examined.

Before arranging the sections on the slide, the slide
should be very thinly covered (by rubbing with the finger
tip) with Mayer's albumen fixative (Lee's *Vade-Mecum*, p.
143), which can be bought of dealers in microscopical sup-
plies. Then allow a few drops of distilled water from a
pipette to flow over the slide so that the sections may *float*.
This will straighten out any folded or crumpled sections.
The water must be allowed to evaporate slowly, by putting
the slide on top of the paraffin oven or otherwise heating it
slightly, being careful not to melt the paraffin. After the
sections are entirely dry, the slide should be placed in the
paraffin oven, and allowed to remain until the paraffin is
melted. Or the slide may be gently and carefully warmed
over a gas or alcohol flame. The slide should not be heated
to a temperature higher than required to melt the paraffin.
After the paraffin is melted the slide should be allowed to
cool until the paraffin hardens again.

Removing the paraffin.—The slide should now be put
into a small jar of xylol, which will quickly dissolve the
paraffin. Leave it in the xylol for about ten minutes, even

though the paraffin has apparently been dissolved at the end of tnree or four minutes. (The slide can be left in xylol, if the work must be interrupted, for several hours or even a day without injury.) Turpentine or toluol can be used instead of xylol. From the xylol remove the slide to a jar of absolute alcohol (or 96% alcohol may be used), which will remove the xylol. The slide should remain in the alcohol at least five minutes. It may remain longer without harm. It should then be transferred to a jar of 90% alcohol for a few minutes and then to a jar of 70% alcohol for a few minutes. The sections are now ready for staining.

Staining.—The number of different "stains" described in a manual of technic such as Lee's *Vade-Mecum* is so large, and the recommendations regarding their use often so confusing in the uniformity of commendation, that the beginning student of histology is thoroughly at a loss when it comes to making selection from such an *embarras de richesses*. For general work in insect histology there is little demand for that large and growing series of so-called strictly chromatin stains, so essential in cytological work. A few nuclear stains, in the wider sense of the phrase, a few plasma stains, a stain which will certainly distinguish chitin, and a stain or two especially for staining *in toto*, are all that the beginner needs for most of his work.

Having selected a stain, the student should bring the slide from the 70% alcohol (see above) into the jar of staining fluid. Here the slide must remain until the sections are thoroughly stained (for time required, see in account of each stain). Some workers pour a little of the staining fluid on to the slide, but the use of small jars full of staining fluid is much to be preferred. After the sections are stained, they must be washed in 70% alcohol or water, depending upon the character of the staining solution (see directions in case of each stain). They must then be

dehydrated by bathing with alcohol of successive strengths up to 96%.

The following stains we use commonly and find excellent for most work :—

FOR STAINING SECTIONS.

Mayer's acidulated carmine (Lee's *Vade-Mecum*, p. 174).— Carminic acid, 4 grams, distilled water, 15 c.c., hydrochloric acid, 30 drops. Boil till the carmine is dissolved, and after cooling add 95 c.c. of alcohol of 85% strength (or for staining *in toto* use alcohol of 95% strength), and filter the fluid. After filtering, the now acid solution should be made neutral by slowly adding drops of ammonia until neutralization is effected. This gives a beautiful, deeply-red-colored, and quickly acting stain. Leave the slide in the stain from five to fifteen or twenty minutes. Remove to 70% alcohol for washing and then to 96% alcohol. It will be noted that, in addition to the sections, the thin coating of albumen fixative on the slide has been slightly stained. This can be removed by passing the slide quickly through 96% alcohol containing a very little (1 to 1000) hydrochloric acid. This bath of acidulated alcohol also "differentiates" the stain, *i.e.*, takes out some of the stain from the cytoplasm of the cells, leaving the cytoplasm much more faintly stained than the nuclei. From the acidulated alcohol the slide should be removed to pure 96% alcohol. (For further manipulation see Clearing and Mounting, *postea*.)

Grenacher's alcoholic borax-carmine (Lee, p. 172).—Make a concentrated solution of carmine in borax solution (2 to 3 per cent. carmine to 4 per cent. borax) by boiling for half an hour or more ; dilute it with about an equal volume of 70% alcohol, allow it to stand 24 hours and filter. Add to this solution an equal amount of 70% alcohol ; allow the mixture

to stand for a week and again filter. Especially useful for staining in bulk.

Delafield's hæmatoxylin (Lee, p. 184).—This is one of the long used standard stains and can be bought, ready prepared, of any dealer in microscopical supplies. The formula for its making is as follows : " To 400 c.c. of saturated solution of ammonia alum (ammonia alum dissolves in about 11 parts of water) add 4 grams of hæmatoxylin crystals dissolved in 25 c.c. of strong alcohol. Leave it exposed to the light and air in an unstoppered bottle for three or four days. Filter and add 100 c.c. of glycerin and 100 c.c. of methylic alcohol (CH_4O). Allow the solution to stand until the color is sufficiently dark, then filter and keep in a tightly stoppered bottle. . . . It should be allowed to 'ripen' for at least two months before using it." (Lee.) For staining, some of this stock solution should be considerably diluted with distilled water. We add a few drops of acetic acid (following Bütschli's suggestion) and get a much sharper stain.

The color is blue, but the acetic acid produces a strong reddish tinge. As this is an aqueous stain, the slide should be carried from the 70% alcohol (see p. 133) to a jar of distilled water before putting it into the jar of staining fluid. The stain acts quickly, depending on the amount of dilution, and should be washed out with water. After washing with water, the slide should be carried through 50%, 70%, and into 96% alcohol. It does not need differentiation (if acetic acid has been added) and the albumen coating does not take up the stain.

Mayer's hæmalum (Lee, p. 182).—One gram of hæmatein dissolved with heat in 50 c.c. of 90% alcohol, and added to a solution of 50 gr. of alum in a litre of distilled water. Allow the liquid to cool and settle, and filter if necessary. A quickly acting stain. After staining, the sections should be washed in distilled water.

Picric acid.—If desired, and it will often be advisable, to follow the nuclear stain (any one of the preceding) with a secondary stain, picric acid can be employed to best advantage. It is of special value in insect histology because it clearly distinguishes chitin. A little picric acid should be added to a small jar of 96% alcohol and the slide passed through it before going into the pure 96% alcohol. This bath of picric alcohol must come *after* the bath of acidulated alcohol used in differentiating after Mayer's acidulated carmine.

Heidenhain's iron hæmatoxylin (Lee's *Vade-Mecum*, p. 189). This is a more complicated method of hæmatoxylin staining, which, however, gives excellent and beautiful results and is very helpful in those cases where cellular structure is being studied. The staining is done as follows : Put sections for from one to two hours in a 2% to 4% aqueous solution of ferric alum $(NH_4)_2Fe_2(SO_4)_4$. This mordants them. Then wash the sections in water for from five to ten minutes. Then stain for from one-half to two hours in a $\frac{1}{2}$% aqueous solution of hæmatoxylin (made by taking 3 c.c. of a 16% alcoholic solution of hæmatoxylin, chloral hydrate 2 grams, and distilled water 97 c.c.). Then wash the sections in water, and differentiate by dipping the slide for a few seconds in the mordant (the ferric alum solution) and then rinsing in tap water, repeating this operation until the correct differentiation has been attained as ascertained by examination of the wet slide under the microscope. The chromatin should be deep blue or blue black, the cytoplasm gray or light blue. After differentiating, the slide should be washed in running water for fifteen or twenty minutes to remove completely the ferric alum. Considerable practice is necessary to obtain the best results with this stain. It should not be used with thick sections, *i.e.*, thicker than 10 microns, for it is an opaque stain.

FOR STAINING IN TOTO.

If it is desired to stain the insect specimens before cutting the sections, a custom which used to be very commonly followed, but is being discarded by many present-day workers, a very penetrating stain must be used. Perhaps the most widely used stain of this character is Grenacher's alcoholic borax-carmine (see p. 134) This stain, all ready for use, can be bought of any dealer in microscopical supplies. The specimens, pieces of tissue or organs, or small insects with weakly chitinized integument, are taken from the 70% alcohol in which they have been kept for a day after fixing (see pp. 125, 126) and are put into this stain and left there for one or two days or even longer, and then put into 70% alcohol which has been acidulated with hydrochloric acid (1 HCl to 1000 alcohol) for differentiation. Leave the specimens in the acidulated alcohol until no more coloring matter is being extracted. Then wash in neutral 70% alcohol and harden in 85%, 95%, and absolute alcohol, clear, imbed, cut, and after removing the paraffin from sections in xylol (see p. 132) mount in balsam (see below).

Mayer's acidulated carmine (see p. 134) can also be used for staining in bulk as well as for staining sections.

Clearing and mounting.—The sections after staining and washing and removal to 95% alcohol have yet to be cleared and mounted. Either cedar-wood oil or xylol or other similar oils may be used. We use xylol commonly. The slide should go from the 95% alcohol into a jar of absolute alcohol for ten or fifteen minutes and then into a jar of xylol for as long a time. For mounting use thin balsam and no more than is necessary. The xylol is very volatile and the mounting must be done rapidly to prevent a drying of some of the sections. This is an accident to be carefully

avoided throughout the whole course of manipulation. If the sections dry they are most likely ruined.

After mounting, put the slides into the paraffin oven for several hours to harden the balsam. The label should bear not only the name of specimen but, advisably, the name of stain used and also the name of fixing agent. The date of mounting should be added, so that the sections may serve as tests of the value of the method of fixing and staining. Some stains do not hold well when used after certain fixing agents.

RECAPITULATION.

As a convenient guide for the beginning student the course of procedure described in the preceding pages is here recapitulated in its proper sequence.

FOR MATERIAL TO BE STAINED IN SECTIONS ON SLIDE.

1. Kill and fix fresh material with fixing agent (p. 124).
2. Wash out fixing agent with 70% alcohol (pp. 125, 126).
3. Remove to 85% alcohol for keeping, or for beginning hardening (pp. 125, 126).
4. Harden in 95% alcohol and absolute alcohol (p. 127).
5. Clear in xylol, cedar-wood oil or other clearer (p. 127).
6. Infiltrate with paraffin in paraffin oven (p. 127).
7. Imbed (p. 128).
8. Cut sections (p. 129).
9. Fasten sections on slides with Mayer's albumen and water (p. 132).
10. Remove paraffin from sections with xylol or other agent (p. 132).
11. Remove xylol with absolute alcohol (p. 133).
12. Carry through 90% alcohol and 70% alcohol (p. 133).

13. Stain with Mayer's acidulated carmine, Mayer's hæm-alum, or other stain (p. 133).
14. Wash out stain with 70% alcohol, or water if aqueous stain is used (p. 133).
15. Remove to 95% alcohol (p. 134).
16. Differentiate with acidulated 95% alcohol (p. 134).
17. Remove to neutral 95% alcohol (p. 134).
18. Remove to absolute alcohol (p. 137).
19. Clear sections with xylol, cedar-wood oil, or other clearer (p. 137).
20. Mount in balsam.
21. Label.
22. Harden balsam mounts in paraffin oven.

FOR MATERIAL STAINED IN TOTO.

Vary the preceding course of procedure as follows :

Between 2 and 3 stain *in toto* with alcoholic borax carmine, or other stain (p. 137).

Omit 12 to 18 inclusive. (It is possible to mount directly after removing paraffin with xylol, but the mount is likely to be unclean unless fresh xylol is used for removal of the paraffin.)

INDEX AND GLOSSARY