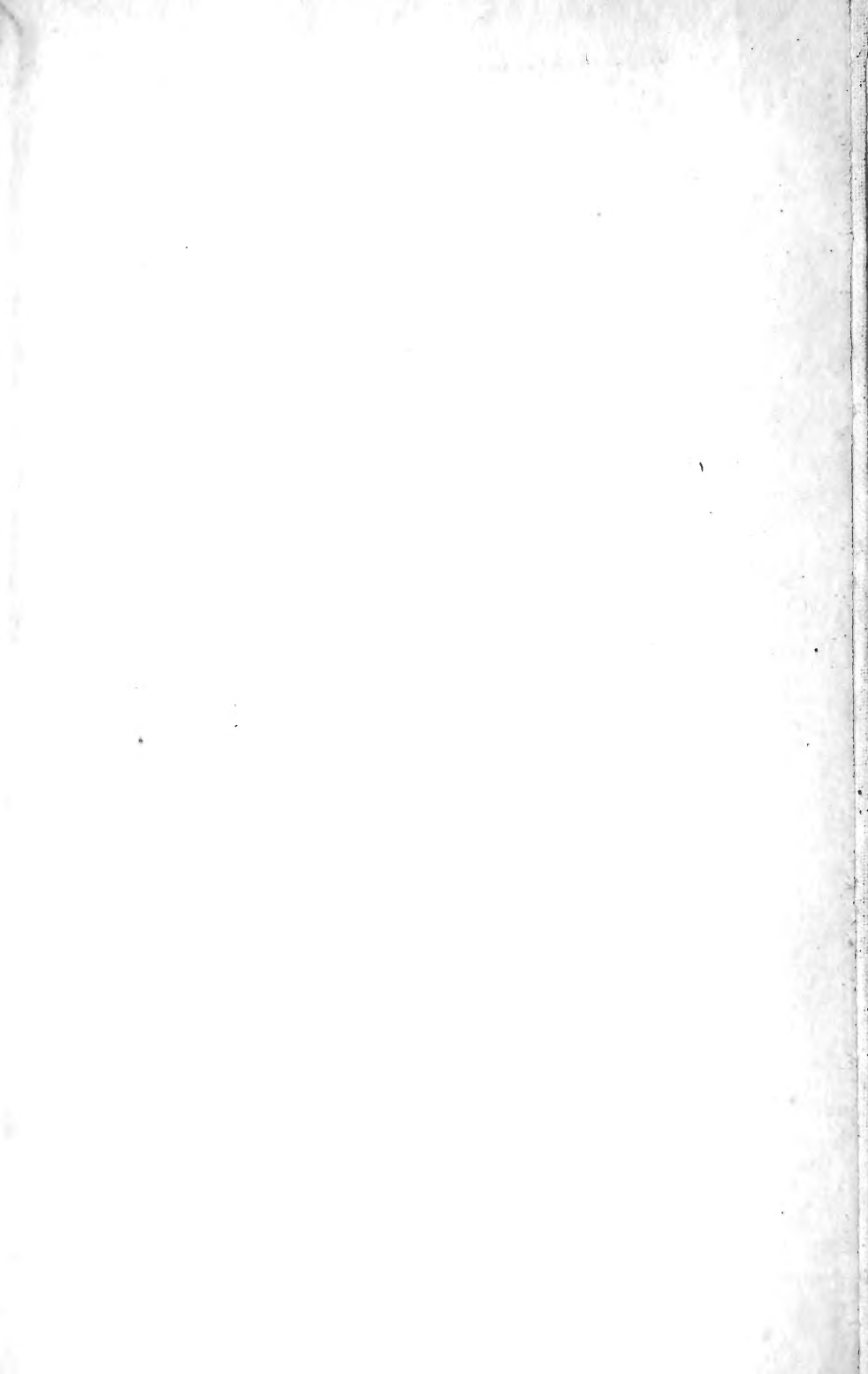
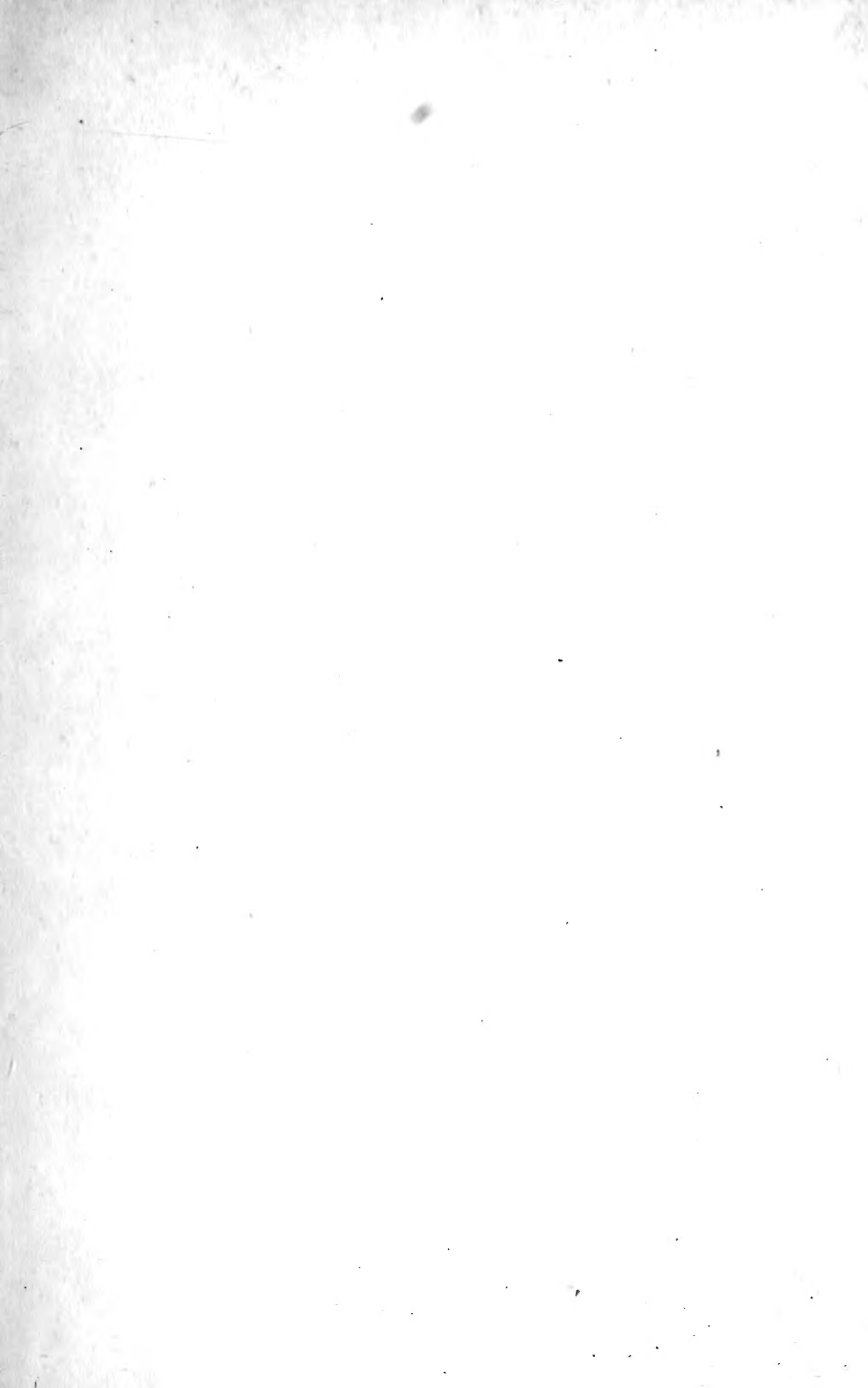


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TRANSACTIONS

OF THE

American Microscopical Society

VOLUME XXXV

1916

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SPENCER-TOLLES MEMORIAL

This Society holds a Fund, accumulated in honor of these pioneer microscope makers and now amounting to nearly \$5,000. The income from this fund is devoted to stimulating research in any field of microscopy.

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TRANSACTIONS OF American Microscopical Society

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Vol. XXXV

JANUARY, 1916

No 1

MASONRY BASES FOR THE INSTALLATION OF MICROSCOPES AND THEIR ACCESSORIES, INCLUDING THE CAMERA LUCIDA AND THE MICROSCOPE CAMERA

By N. A. COBB

MASONRY AS A BASE

It has long been customary in the best laboratories to mount instruments of precision upon heavy pillars having foundations located in wells in the ground and passing upward through the floors without contact, the object being to prevent the tremors of the building from being transmitted to the instruments. The earth receives and nullifies the tremors.

The microscope has not often* received such special attention, notwithstanding the fact that whenever high powers are used, and especially when photomicrographs or high power camera lucida drawings are being prepared, vibration is objectionable. For many years I have had microscopes mounted on solid bases and strongly favor this method of support.

THE USE OF GIRDERS

One such installation was carried out in cement and steel as shown in Fig. 1.** Three girders, two approximately eight inches in each transverse dimension, and between them a third smaller one,

*The author knows of only about twenty of the massive installations such as are described in the following pages.

**The illustrations were made under the personal supervision of the author by Mr. E. M. Grosse and Mr. W. E. Chambers. They show so much detail, so nearly to scale, that any good mechanic can build from them.

are imbedded vertically to the depth of several feet in a block of cement weighing many tons located under the building. The middle short girder, extending 18 inches above the floor, carries the microscope and certain accessories connected with illumination. The two tall, paired girders extend to within eighteen inches of the ceiling, projecting upward into the room about eleven feet. The wooden floor was laid tightly about the girders after they had been set in the cement and everything was then given a few months in which to settle into permanent position, after which an ordinary key-hole saw was run through the floor entirely around the contour of each girder, so that each cleared the flooring and floor-covering by the thickness of a saw blade. See 3, 43, etc., Fig. 1.

The girders, which clear the wall of the room by an inch or two, carry the accessory apparatus, which is attached to them in some instances by means of sliding one-sixteenth-inch sheet-metal sleeves that may be clamped at any desired height, in other instances by other means. All parts are dead black. The sleeve of the small central pillar projects outward at the top, that is, toward the observer, so as to form a microscope shelf one to two times larger than the base of the microscope. The sleeve carrying the microscope is clamped to its pillar by set-screws, and can be set high or low to suit different operators and different classes of work. When photographing it is better to set the microscope near the floor, so as to bring the camera (19, Fig. 1) low enough to make it unnecessary to use a step-ladder in focusing. By placing the microscope high and the drawing table low, one can obtain for camera lucida work a distance of two and one-half feet between the level of the eye-piece of the microscope and that of the drawing table.

The sleeve carrying the microscope carries also a large vertical wooden front, two feet wide and as long as the microscope window is wide. This wooden front or screen slides up and down with the microscope, and has in it two apertures, one in front of the microscope mirror, designed to allow light from the sky, or from an illuminating screen, to strike the mirror and pass thence through the microscope; and the second to secure a correct illumination of the drawing board when the camera lucida is in use. See Fig. 1. This latter aperture is much the larger, is glazed with ground

glass, and is opened and closed at will by means of a light, suspended, opaque slide worked up and down by foot-power.

The microscope window faces the sun, and is fitted with two light-proof roller blinds, one just behind the other, so that the sun's light may be shut off or allowed full access. The roller blinds move in lateral grooves ten inches deep, a depth sufficient to prevent the blinds from troubling by bellying on windy days. As a further provision against wind action, tight wires may be strung horizontally on the window frame inside the blinds. The blinds may be of any opaque material, and if long, should be thin. Ordinary opaque window blinds can be sized black so as to become practically light-proof, and since it is advisable to have two blinds, such sizing wholly excludes the light. Ordinary spring rollers are used and are boxed in at the top in a light-tight manner.

CARRIERS FOR THE ACCESSORIES

The right-hand lower sleeve carries a leg-of-mutton shaped shelf or table for use in making camera lucida drawings. The sleeve, like all the other supports is balanced with a sash-weight, so as to move freely up or down through a range of several feet. The shape of the shelf has been evolved from years of use, and gives the investigator free play for hands and body. See 33, 34, Fig. 1.

At the left is a similar sleeve and shelf used for a different purpose,—although, as it is the mate to the camera lucida table, it could, in the case of a left-handed operator, be used as the right-hand table would be used by a right-handed operator. The usual position for the left-hand shelf is, however, about on a level with the microscope stage; first, because that is about ordinary table height and is convenient for supporting the dissecting microscope, which has a special illumination of its own (See Fig. 1); and second, because preparations may then be moved on and off the stage of the microscope with the least danger and with the greatest facility; a third reason is that in this position the left forearm finds it a most convenient rest in working the fine adjustment screw of the microscope, as shown in Fig. 3.

The two long girders also carry two strong vertically adjustable cross-pieces for the attachment of accessories above the microscope. Set-screws are provided for clamping the cross-pieces. Arms extend upward from the cross-pieces to carry anti-friction pulleys travelling on edges of the pillar. The microscope camera (See 19, Fig. 1,) hangs above the microscope in readiness for instant use, and is of vertical pattern, carrying the photographic plate in a horizontal position. In focusing, the cross-piece carrying the camera is moved upward or downward; a scale on the pillars gives the various magnifications. The operator loosens two hooks and the camera front drops instantly into position on the end of the microscope barrel. The whole is ready for use in a few seconds' time. If the exposure is long, one leaves the instrument during the exposure with the greatest confidence that nothing can disturb it; tremors in the building will not be received either by the microscope or the photographic plate.

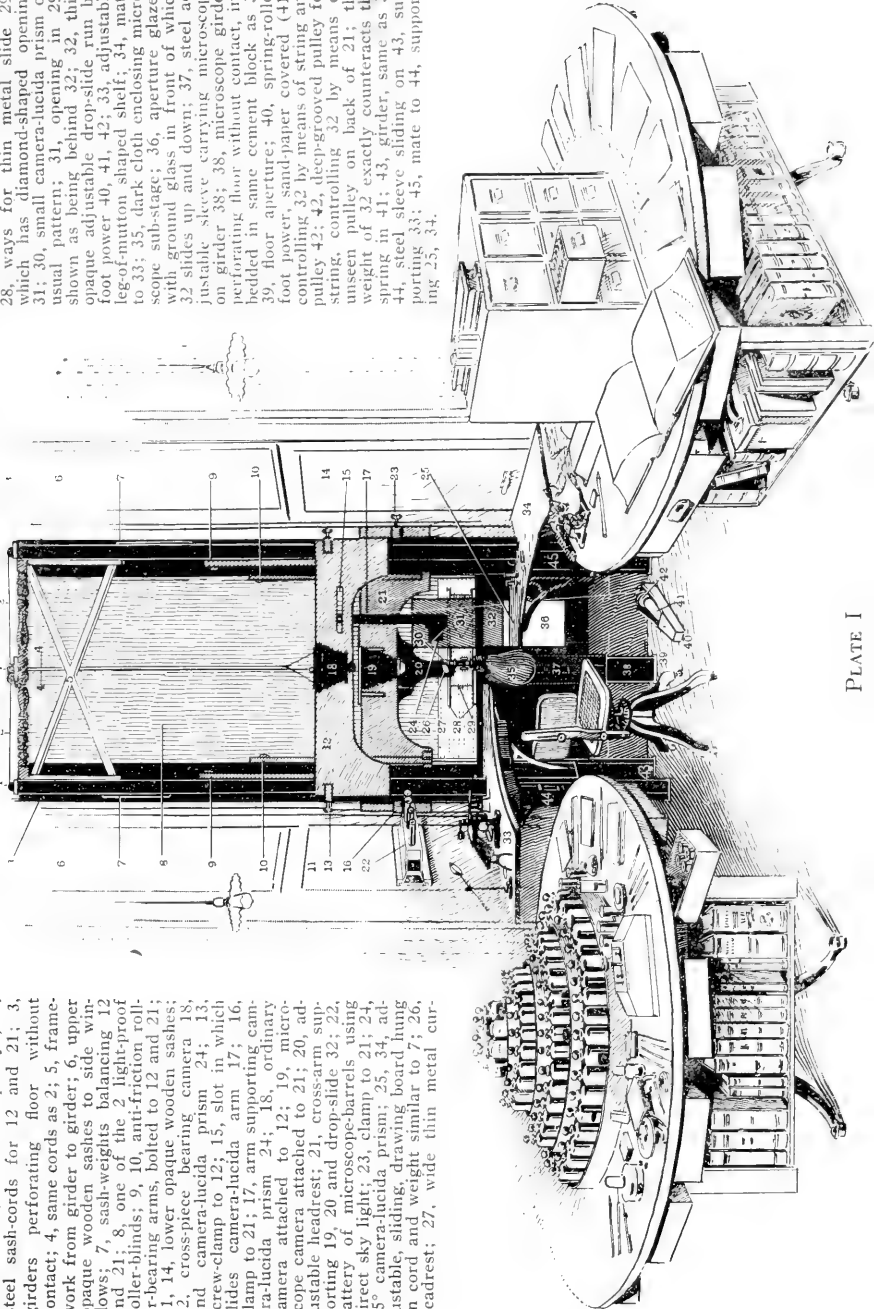
CAMERA LUCIDA

A second attachment of prime importance in producing illustrations is the peculiar camera lucida. Fig. 1, 17, 24.

The history of the camera lucida is a very interesting one. It is impossible to go into details here, but nothing is clearer than that this instrument is one of great importance to the microscopist, and its history is in accordance with this fact. The utmost ingenuity has been exercised to produce an instrument by means of which sketches of small objects can be made with the aid of the microscope. The necessity for this class of work is very great. The photographic camera is inadequate for most objects. Only in the case of smears or exceedingly thin sections, or natural objects of great thinness, is a photomicrograph satisfactory. In all other cases, in order fully to elucidate the structure by means of an illustration, it is necessary to represent the appearances at different depths in the preparation. This can be done only by focusing the microscope for each particular depth. This fact, thus hastily explained, is what makes it absolutely necessary to use a camera lucida for the proper representation of most microscopic objects. This fundamental necessity is what has given rise to the many

Fig. 1.—1, sash-cord pulleys; 2, steel sash-cords for 12 and 21; 3, girders perforating floor without contact; 4, same cords as 2; 5, frame-work from girder to girder; 6, upper opaque wooden sashes to side windows; 7, sash-weights balancing 12 and 21; 8, one of the 2 light-proof roller-blinds; 9, 10, anti-friction roller-bearing arms, bolted to 12 and 21; 11, 14, lower opaque wooden sashes; 12, cross-piece bearing camera 18, and camera-lucida prism 24; 13, screw-clamp to 12; 15, slot in which slides camera-lucida arm 17; 16, clamp to 21; 17, arm supporting camera-lucida prism 24; 18, ordinary camera attached to 12; 19, microscope camera attached to 21; 20, adjustable headrest; 21, cross-arm supporting 19, 20 and drop-slide 32; 22, battery of microscope-barrels using direct sky light; 23, clamp to 21; 24, 45° camera-lucida prism; 25, 34, adjustable sliding drawing board hung on cord and weight similar to 7; 26, headrest; 27, wide thin metal cur-

tain-stick to inside roller-blind 8; 28, ways for thin metal slide 29, which has diamond-shaped opening 31; 30, small camera-lucida prism of usual pattern; 31, opening in 29 shown as being behind 32; 32, thin opaque adjustable drop-slide run by foot power 40, 41, 42; 33, adjustable leg-of-mutton shaped shelf; 34, mate to 33; 35, dark cloth enclosing microscope sub-stage; 36, aperture micro- with ground glass in front of which 32 slides up and down; 37, steel adjustable sleeve carrying microscope on girder 38; 38, microscope girder perforating floor without contact, imbedded in same cement block as 3; 39, floor aperture; 40, spring-roller foot power, sand-paper covered (41) controlling 32 by means of string and pulley 42; 42, deep-grooved pulley for string, controlling 32 by means of unseen pulley on back of 21; the weight of 32 exactly counteracts the spring in 41; 43, girder, same as 3; 44, steel sleeve sliding on 43, supporting 33; 45, mate to 44, supporting 25, 34.



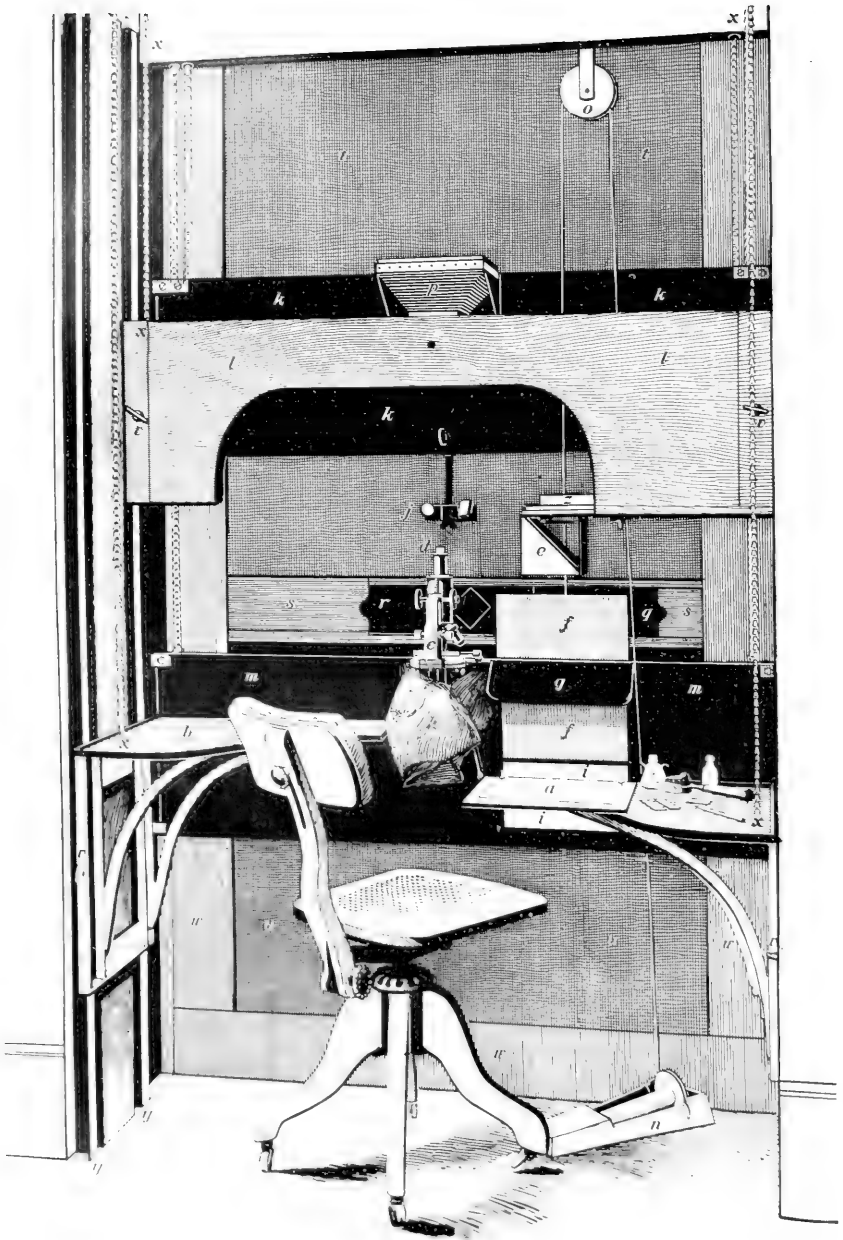


FIG. 5

pinned to right-hand shelf; *b*, left-hand shelf; *e*, microscope; *ism*; *c*, large camera lucida prism; *f*, opaque slide, moving in by means of the foot power, *n*, *c*, hinged flap to prevent direct view of the operator, *h*, cloth surrounding the base and sub-stage of the camera lucida, with ground glass; *f*, adjustable camera lucida prism; *p*, *l*, cross-piece supporting prism *c*; *n*, foot power which, by means of a string pass-

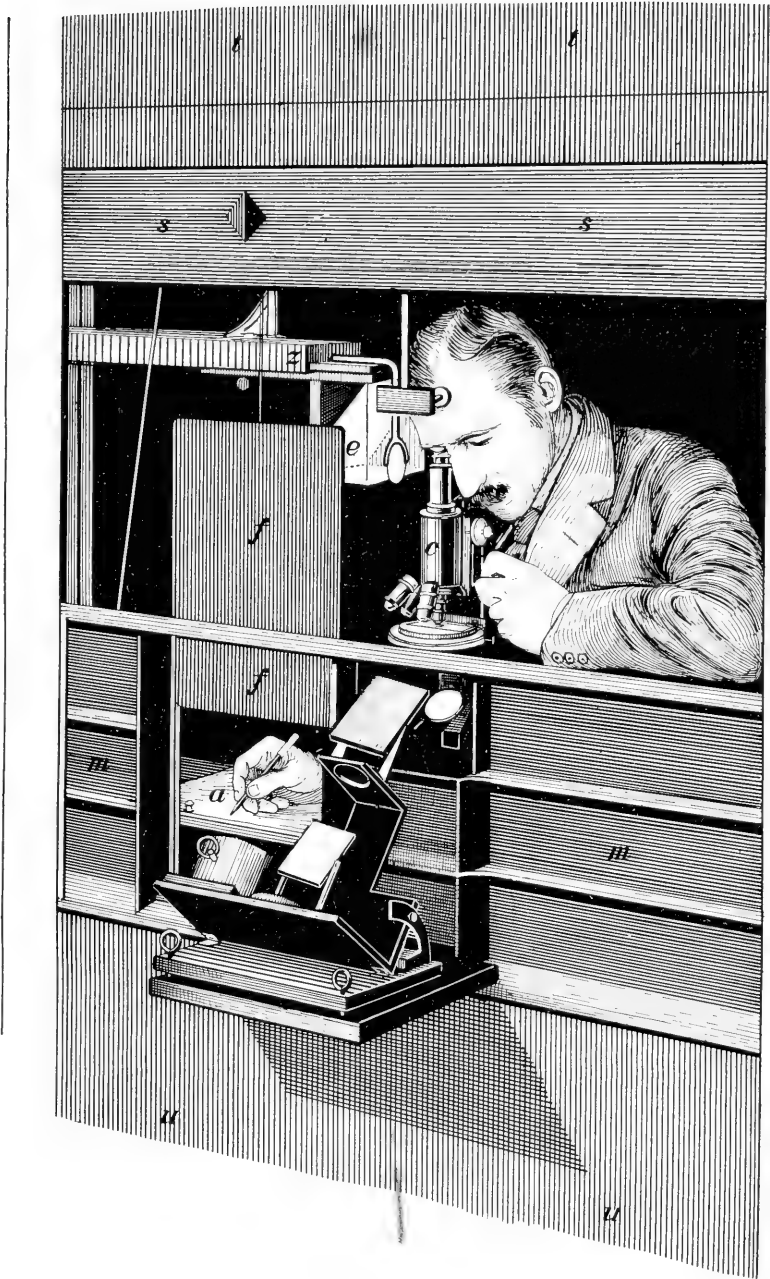


FIG. 3

ing over the pulley *c*, works the slide *f*; *o*, pulley; *f*, micro-camera; *q*, *r*, thin opaque slides with diamond-shaped openings sliding in front of a glazed aperture in the wide, thin curtain stick *s*; *t*, opaque blind rolling upward carrying with it *q*, *r*, *s*; *u*, roller blind rolling downward carrying with it *g*, *h*, *m*; *v*, set-screws for the shelves *a* and *b*, as well as for the cross-pieces above; *w*, ten-inch grooves forming light traps for the blinds *t* and *u*; *x*, chains passing over pulleys to sash weights by which the various sliding parts are counterbalanced; *y*, *z*-shaped girders bolted to masonry of the building.

PLATE IV

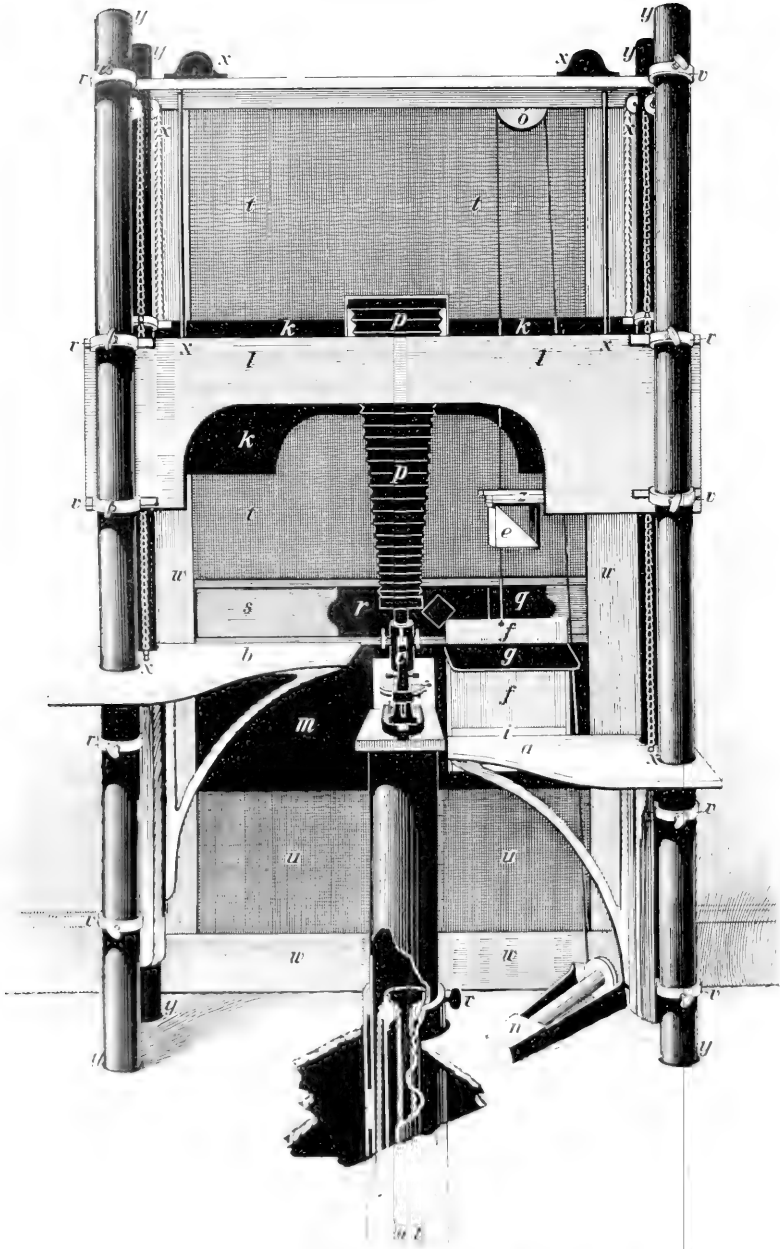


Fig. 1. A perspective view of the printing press, showing the rollers, gears, and frame.

patterns which the camera lucida has taken on during its development. The first instrument was an extremely simple one. From time to time improvements and additions have been made until at the present time the instruments issued by the best makers are marvels of ingenuity and workmanship. In fact, in the writer's opinion, they are almost too ingenious, for it appears to him that the various additions instrument-makers have made to the camera lucida during recent years, while they do accomplish the object aimed at, do so in an unsatisfactory manner.

To produce a good camera lucida drawing, it is necessary to have such light passing through the microscope as will enable the operator to see the object with the greatest possible clearness; and it is equally necessary so to control the light from the drawing board as to enable him to see his pencil-point at all times with the greatest possible clearness. With most objects it is impossible to secure this adjustment once for all, and for all portions of the drawing. Different portions of the object transmit or reflect different amounts of light, and, as this light varies it is necessary, in order that the drawing may be made with the greatest ease and precision, that the light from the drawing board should be modified accordingly. This end has been sought in a variety of ways, and more than any other one thing the effort to achieve this end has added to the complexity of the modern camera lucida. When the modern instrument is in good order, it may, it must be admitted, in a way accomplish its object; the difficulty is that it is complex and easily thrown out of adjustment, and some of its parts easily become soiled and dusty so as to be a hindrance rather than a help. Again, no very suitable device has yet been furnished by manufacturers for modifying the light from the drawing paper, except by a series of steps. It is usual to interpose tinted glasses, until the right balance of light has been secured, but it often happens that the most desirable shade cannot be secured, and in any case by this method there is always being inserted between the object and the eye, or the pencil and the eye, or between both and the eye, various pieces of apparatus that must be regarded as necessary evils, objectionable from a number of points of view.

A second defect presented by many camera lucidas is the "double" reflection due to a silvered glass mirror. This defect can be tolerated for a short time, but after several hours the double reflection of the pencil point becomes very tiresome to the eye.

Any form of camera lucida hitherto introduced is trying to the eye-sight. During many years of experience the writer has endeavored to reduce the risk of injury to eye-sight due to use of the camera lucida, and the following suggestions, embodied in the outfits here described, are the result of his experiments. First, he has substituted for the ordinary mirror a large, and therefore necessarily heavy, 45 degree glass prism mounted on a support separate from the microscope. (24, Fig. 1 and e Figs. 2, 3 and 4). The advantages of this substitution are: 1. As the prism is mounted separately, it may be of any desired size, and may be placed at a considerable distance from the eye-piece of the microscope, thus increasing the magnification of the drawing; the advisability of this increased magnification will be dwelt upon later. 2. There are no double or multiple images and less light is lost by reflection. The light passes from the drawing-point through the lower, i. e. horizontal face of the prism in a nearly perpendicular direction with little loss. It is then "totally" reflected from the oblique face, again with very little loss. 3. A third advantage of considerable importance is the stability of the apparatus; it rarely gets out of register. Forty-five degree prisms are made in about three qualities. The most perfectly corrected ones are very expensive; the second or even third grade prisms of the best manufacturers are suitable for camera lucida work.

ILLUMINATION OF THE DRAWING-PAPER

The second modification is that referred to on a previous page as the suspended slide or blind worked by foot-power. (Fig. 1, 32 and f Figs. 2, 3 and 4). This slide enables the operator to illuminate the drawing with almost any degree of light at an instant's notice without disturbing the adjustment of any part of the microscope or camera lucida. This is highly important in the rapid production of good camera lucida sketches. Especially with high powers, the light coming through

the microscope is often so faint that it is only by almost completely shutting off the light from the drawing, that the investigator can see at the same time both his pencil and the details of the structures to be sketched. With the foot-power arrangement the operator modifies the light in a second without disturbing the position of his body or his drawing-point, and instantly brings about that adjustment which is most favorable for any particular part of the sketch. Briefly, we may say that the operator's left arm rests on the left-hand "leg-of-mutton" shaped table on a level with the fine adjustment of the microscope, with his left hand in position to work the fine adjustment screw with the greatest ease and accuracy. His right hand, carrying the drawing-point, rests on the drawing board and is engaged in producing the sketch. As the light required for the different portions of the sketch varies he can effect the necessary change in illumination of the drawing paper by a slight movement of his right foot, which disturbs neither his hands nor the equilibrium of the instruments.

METHOD OF TRACING CAMERA-LUCIDA DRAWINGS

As will be at once conceded by any one who makes a trial, black paper is best adapted for camera lucida drawing; a white drawing point should be used. This is an improvement over a pencil used on white paper. The best combination is a thin black tissue-paper, blued on the under side by rubbing on dry prussian blue powder. A piece of drawing paper or enameled board of suitable size for the drawing is pinned to the drawing board,—i. e., the right-hand leg-of-mutton shaped table,—and is then covered with the black tissue-paper, blue side down, pinned at its back edge only. The sketch, a blue tracing, is now made with a fine white ivory or bone point. This blue sketch is put aside for further reference, or for the production of a finished drawing later on, or it may be inked in at once. The object aimed at is a satisfactory representation of the object to be illustrated, of sufficient size to admit of liberal reduction when the ink drawing is photographed on metal preparatory to etching. If it is desired to publish an illustration having a magnification of say 250 diameters, it is advisable to produce a blue sketch of at least 1,000 to 2,000 diameters.

Highly magnified sketches are easily obtained with the apparatus just described, for by placing the prism reflector at a considerable horizontal distance from the eye-piece of the microscope, say one foot, and lowering the right-hand leg-of-mutton shaped table, magnifications of 5,000 diameters and upward are easily secured. Not infrequently the production of a large coarse drawing is an easier matter than the production of the same drawing on a smaller scale. The conversion of the blue sketch into a pen and ink drawing presents no special peculiarities, but perhaps it ought to be mentioned that the object of using blue is to avoid trouble arising from alterations that may become necessary in finishing the drawing. Any light blue lines which are left on the drawing paper need not be removed, as they do not affect the photographic film sufficiently to cause any inconvenience in the production of an etched block. The black tissue paper mentioned is produced by inking ordinary tissue. The ordinary blue carbon paper gives too dark a blue to meet the requirements. A good quality of blue tracing paper may be made from black typewriter carbon-paper that has been exhausted by the typewriter,—the blue powder being rubbed onto the clean side of the carbon paper.

DARKENING THE ROOM CONTAINING THE INSTALLATION

In addition to blackening all accessories, arrangements are made to darken the room itself, in fact, to make it convertible into a photographic dark-room at will. All the window-blind connections are light-tight. The oblong aperture, about five inches by eight inches, through which the microscope mirror receives its light is screened by means of several thicknesses of flexible black cloth made into the form of a sleeve. This cloth sleeve, attached to the perimeter of the beveled aperture, is slit above and made to surround the microscope just beneath the stage, and the margins overlap and button to one of the screws at the back of the microscope; the lower part of the microscope is thus located within the sleeve. No light reaches the observer's eye except that which comes through the instrument.

If, now, the slide in front of the large glazed aperture be closed and all direct light shut out, the operator sits in darkness. Any one who has had experience with a photographic dark-room must have observed how after a period of from five to ten minutes therein the eye becomes accustomed to the darkness of the room and is able to distinguish objects much more readily than at first. This is a principle which can be utilized to advantage in connection with high-power microscope work. In fact, it appears to be the relation between the external and the internal illumination which leads so many operators to use artificial light, and even in some cases to prefer working in the evening. If the surrounding light is dim and the eye is allowed to adjust itself to this dimness, then on looking through the properly adjusted microscope, certain details may be seen more clearly than in any other way. It is sometimes painful to witness the unconscious efforts of microscopists to bring about this condition as fully as possible by means of awkward attitudes and facial expression. It is not at all uncommon to see the microscope placed in a glare with strong light beating on the top of the preparation being examined and thence reflected confusingly up through the microscope, and to see the operator sitting in a cramped position, bending his head over the top of the instrument so as to shade his eye-piece as much as possible, and thus prevent eye-piece reflections. All this painful effort is simply an attempt to give the eye the benefit of a weak extraneous light and to prevent confusing reflections from the top of the mount and the eye-piece. With the apparatus here described these difficulties are minimized. The room is darkened. All light which could reach the operator's eye is excluded, except that which comes through the microscope. There is no light falling upon the top of the object, to cause confusing reflections inside the microscope, nor can rays of light be thrown into the eye from the top of the eye-piece, or from high lights on the stand. The image to be examined is as clear as it can be made and the eye is given every facility to see it, and is distracted by no others.

The advantages of this system of using a microscope are not confined to high-powers. It is well known that the central portions of microscope lenses act more perfectly than the peripheral portions.

By shutting out these latter, better optical results are secured, but the illumination is considerably diminished. The low degree of illumination is less objectionable when the observer is in darkness or semi-darkness. I believe this is due to some extent to relaxation of the iris muscles.

It might at first be thought that this would result in a less perfect image on the retina of the eye, and no doubt this is true if we consider the entire image on the retina. The fact is, however, that the observer concentrates his attention upon a small portion of the retina image and this portion may be focused as accurately as his eye admits. In respect to perfection of image, a comparison of the human eye to an ordinary camera may be very misleading.

CAMERA-LUCIDA DRAWINGS OF DIFFICULT OPAQUE OBJECTS

Camera lucida drawings of opaque objects present numerous difficulties, prominent among which is the small amount of light coming through the microscope. By illuminating the object with a strong light, sunlight if necessary, and reducing to a minimum the amount of light coming from the drawing paper, it is not at all difficult with this apparatus to produce satisfactory drawings of these difficult objects.

Needless to say the apparatus is a daylight apparatus. It hardly seems necessary to argue that as daylight is the light that has developed the human eye it is probably the light to which it is best adapted. This seems a sufficient argument for the use of daylight and a sufficient explanation of its superiority to every other light for the average run of microscopic work. Using the installations here described, monochromatic light of superlative quality is easily obtained by interposing colored glasses or liquids. However, when all this is said, it is not possible always to secure and control daylight so as to get the best results. The following contrivances are such as experience has shown to be very useful for this purpose, especially in sunny climates.

SOURCE OF LIGHT FOR THE MICROSCOPE

Outside of the microscope window a universally adjustable three-by-five-foot white screen is placed in a sunny position, preferably about ten feet away. The surface of this screen should be

smooth but not shiny and may be of any fine-grained white material. It can be made of wood, painted white, or lined with plaster of Paris; or, what is better, be a plain wooden screen covered with sheet metal and over all several thicknesses of bleached cotton cloth. Whitewash makes a cheap and very excellent white surface for this purpose, and may be applied over cloth. If a small mirror be attached to one corner of the screen, it will indicate the position of the screen that will reflect to the microscope a maximum of white light. Place the screen so that the flash of sunlight from the mirror strikes in the vicinity of the microscope; then the whole screen will be in corresponding position and reflect a maximum of light to the microscope. It is better if this screen can be adjusted from the interior of the microscope room, but this is not essential. After the screen is set the light from it remains for an hour or more practically constant, so that while an adjustment by cords or other mechanism from the interior is a convenience, it is not a necessity. If an adjustable screen is not available it may be possible to arrange two fixed screens, one screen for morning and the other for afternoon.

Blue sky is not a satisfactory source of light. A white cloud gives a very good light, but clouds are so changeable that it is not wise to rely upon them. It is therefore much preferable to construct, as described, an adjustable white screen that will be available whenever the sun shines. When the sun does not shine the sky may serve, or if the day is too dull, one resorts to artificial light.

MICROSCOPE INSTALLATION BOLTED TO THE MASONRY OF THE BUILDING

The installation just described can be bolted to the masonry of the building by means of small vertical I-shaped or Z-shaped girders as shown in Fig. 2. The installation is in most respects similar to that just described, except that the microscope is carried on a heavy cast-iron cross-piece, *m*, sliding up and down on a pair of L-shaped or Z-shaped girders, and carrying with it an opaque roller blind, *u*, which passes around a roller at the bottom of the window near the floor.

In Fig. 2, a number of details are more clearly shown than in Fig. 1, for instance, the details of the headrest, *j*, which consists of two oblique cork-covered pads, one circular, the other square, each carried on a horizontal screw so that they can be rotated and at the same time moved inward or outward. These pads screw into a cross-piece, which in turn is fastened securely to a vertical rod which slides up and down and rotates, and is clamped by the set-screw shown near *k*. The great adjustability of these pads makes it easy to fit them to the head of any operator. They serve to keep the observer's eye in register, as well as to decrease fatigue, and actually improve his observing power.

Owing to the action of the heart and lungs, and to the unsteady action of the muscles of the neck, the head, when unsupported,

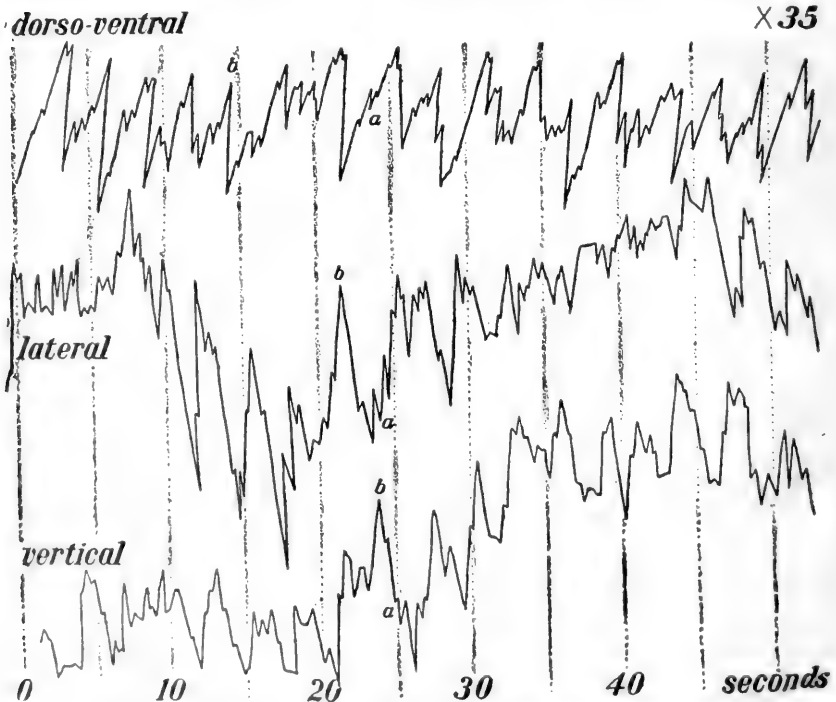


FIG. 5. Three successive graphs showing the nature and extent of the motions of the head of an observer while looking through a microscope. These graphs were obtained by recording the trace of a beam of light from a small mirror actuated by the head of the observer at the microscope.

is in constant motion, and the image of any stationary object formed meanwhile on the retina of the eye is moving in a corresponding way. The movements of the microscopist's head are shown in the accompanying three graphs (Fig. 5). Both the nature and extent of the rotations about three coordinate axes, at right angles to each other, are shown at a magnification of 35 diameters. These graphs were obtained by recording the trace of a beam of light reflected from a small mirror actuated by the head of the observer at the microscope. The smallest motions indicated in the graphs, a, a, a, are due to the action of the heart; each tiny irregularity represents a heart-beat. The larger irregularities, such as those shown at b, b, b, are due to lung action; the head rises and falls with each breath. Moreover, these two irregularities, those due to the heart's action and those due to the action of the lungs, are distributed on a great curve, which I suppose to be a curve of fatigue connected with the action of the muscles of the neck. These graphs show how complicated are the movements of the image on the retina. Keeness of vision is a function, among other things, of the steadiness of the retina. This is a matter of personal experience, but arguments in favor of a steady retina may be suggested somewhat as follows: It is a matter of common observation that all organisms possessing well-developed eyes, hold the head as steady as possible when looking intently. If while reading from a printed page one wags the head perceptibly, even at so slow a rate as that of the heart's beat, vision is very considerably impaired; the less of this motion, the keener the vision. What is the limit of this improvement in vision due to increased steadiness of the retina? Theoretically it would seem to be reached when the retina is absolutely steady. When we consider the minuteness of the elements in the retina having to do with vision,—the rods and cones and other elements—it seems a very reasonable supposition that vision can be increased in keenness beyond the degree attained by holding the head as steadily as possible by means of the cervical muscles. Whether these arguments are valid or not, the writer is convinced, from years of experience with mechanical headrests, that their use adds to the acuteness of vision. Moreover, they diminish fatigue, making it possible for the observer

to use the microscope hour after hour continuously with a smaller expenditure of energy.

The attachment of the large prism is also more clearly shown in Fig. 2. It is mounted on a triangular sheet-metal frame, which revolves on a vertical axis, and is clamped in any position by means of the lever shown in the midst of the triangle.

Fig. 3 is an outside view of the installation shown in Fig. 2, and is correspondingly lettered. Note the headrest in use, and also the spectacle lens between the operators' eye and the prism, e. This spectacle lens is one adapted to the operator's eye, and in case he is accustomed to wear ordinary spectacles for reading or drawing, he is able to dispense with them, which is an advantage, as they would be a hindrance in camera lucida work. The substituted spectacle lens should be made by the optician who makes the operator's spectacles, and according to much the same formula; this lens enables the operator to see his pencil point and drawing clearly.

In this illustration is also shown on a shelf in front, a small heliostat, actuated by the small alarm clock, the top of which is to be seen in the figure. The lower reflector of the heliostat is a mirror, the upper a piece of finely matt white card-board. This heliostat may be used instead of the large reflector previously described, and gives a very good light.

INSTALLATION CARRIED OUT IN STEEL TUBING

The installation shown in Figs. 1, 2, and 3 may be carried out in steel tubing as shown in Fig. 4. This form of installation has not yet been thoroughly tried out, but appears to offer a number of advantages over the other forms at an expense but little, if any, greater. The peculiarities of the construction are well set forth in Fig. 4, and as the lettering is the same as in the preceding figures, little need be added. It will be noted that the sash weights are for the most part suspended inside the tubes, and that the various accessories slide up and down on the tubes through the mediation of cast iron rings, malleable castings, turned to fit the tubing somewhat loosely. The microscope is supported on a vertical tube descending

into another tube of slightly larger size embedded in the cement below, and is counterpoised by a weight which slides in a third smaller tube, also embedded in the cement below. The details are shown in section in the illustration. Collars threaded into the outside tube are turned to fit the tube carrying the microscope. The cross-piece, 1, is suspended on metal straps passing into the spring-pulleys, xx. In all other respects the installation is almost identical with that shown in previous illustrations.

Ordinary rough tubing may be used if its outer surface is smoothed by filing. With such a rough finish, however, the apparatus is liable to be noisy during the adjustment of the various slides. This noise can be eliminated by turning the tubes in a lathe having a long bed, and carefully fitting the cast iron rings to the tubes. With good fitting the apparatus is then practically noiseless. This turning and fitting, however, adds materially to the expense. This installation is neat in appearance, and easy to work. The tubing used is from $3\frac{1}{2}$ inches to 6 inches internal diameter.

This installation differs from those shown in Figs. 2 and 3 in respect to the screen in front of the microscope through which light is admitted to the mirror of the microscope and to the drawing. In the present installation this screen is attached to the tube that carries the microscope, and therefore slides up and down with this tube and with the microscope substantially as shown in Fig. 1. In the tubular installation, therefore, this screen is of very light construction,—made of wood, and very thin,—and its ends, as in Fig. 1, extend deeply into the 10-inch groove at either side of the window.

In Fig. 4 the cloth sleeve that surrounds the microscope has been removed, so as to show the size and nature of the aperture through which the light is admitted to the mirror of the microscope. It will be seen that in this aperture light-filters in the form of glass tanks or colored glasses can be installed *ad libitum*. This method of using tanks and glass filters is a very convenient one. Those of large size can be used.

The shelf at the top of the installation, carrying the spring-pulleys, x, x, and clamped in position by the higher set-screws, v, v, and supporting the pulley, o, has for its main object to keep the four cylindrical pillars in register,—that is, parallel to one another,—and to prevent vibration.

MORPHOLOGY OF ADULT AND LARVAL CESTODES FROM POULTRY*

JOHN E. GUTBERLET

During the course of experimental studies on the life history of certain chicken cestodes described in the succeeding section of this work (Gutberlet, 1916) it was necessary to determine exactly the morphological features of the species, which to be sure, had been studied by others but were only partially and imperfectly known. In the former paper I recorded experiments which demonstrated the intermediate stage of *Choanotænia infundibuliformis* to be in the common housefly, *Musca domestica*, and discussed the symptoms of infection and methods of control for tapeworm diseases in chickens.

In this paper are taken up the structure of the adult and cysticercus of *Choanotænia infundibuliformis* (Goeze), and also the adult form of four other species occurring in chickens of this country.

As described more fully in the preceding paper the worms were removed from the intestine under water. The use of normal salt solution was avoided since it was found to be injurious. The cestodes were killed in a corrosive-acetic solution and preserved in 70% alcohol and glycerine. Best results were secured by staining in Delafield's or Ehrlich's acid hæmatoxylin and destaining in acid alcohol.

The five species discussed here were collected at two widely separated points, a farm at Hardy, Nebraska, and the poultry farm at the University of Illinois. These morphological studies were carried on at the Zoological Laboratory of the University of Illinois. This work was taken up at the suggestion of Dr. Henry B. Ward, to whom I am greatly indebted.

*Contributions from the Zoological Laboratory of the University of Illinois under the Direction of Henry B. Ward, No. 57.

STRUCTURE OF ADULT AND LARVA (CYSTICERCUS)

A. ADULT

Choanotænia infundibuliformis (Goeze 1782) Railliet 1896

1. Diagnosis: Length 50 to 200 mm. Scolex (Fig. 2) small, rounded, or conoidal, about 0.4 mm. wide. Rostellum (Fig. 2, 3, *r*) 60 to 70 μ in diameter, armed with a single row of 16 to 20 hooks (Fig. 8) 25 to 30 μ long, with long dorsal root and short ventral root. Suckers prominent, elongated antero-posteriorly, length 180 to 210 μ ; breadth 135 to 175 μ between the extreme outer edges. Neck short and unsegmented, somewhat narrower than broad. In specimens well extended neck much narrower than head. Anterior proglottids very short and as they become older funnel-shaped, much narrower at anterior than at posterior margins; posterior segments 1.5 to 2.5 mm. broad and 1.5 to 3 mm. long according to amount of contraction, with convex lateral borders, nearly as wide at anterior as at posterior margin. Genital pores irregularly alternating, situated one in each segment in the anterior third of the lateral margin, usually under cover of the backward projecting border of the preceding segment. Vas deferens (Fig. 14, *vd*) and vagina pass between excretory canals and dorsal to nerve trunk.

Male Reproductive Organs: Testicles (Fig. 14, *t*) 25 to 40 or more, 60 in some cases, in posterior half of proglottid, posterior and lateral to large yolk gland, within limits of excretory canals. Vas deferens passes forward and in anterior third of proglottid forms a mass of coils between ovary and excretory vessels from which it extends outward as a convoluted tube to base of cirrus pouch. Cirrus pouch (Fig. 14, 15, *cp*) ovoid in shape, 75 to 95 μ in long diameter. Portion of vas deferens in cirrus pouch is much coiled. Cirrus 50 to 65 μ long, armed with spines; outer surface of cirrus pouch forms base of deep genital cloaca.

Female Reproductive Organs: Vaginal opening in genital cloaca posterior to cirrus. Vagina posterior to cirrus pouch, after crossing ventral excretory canal dilated to form ovoid seminal receptacle, posterior and ventral to vas deferens, extending to well developed shell gland, 40 to 50 μ in diameter located in front of middle of proglottid. Transversely elongated ovary (Fig. 14, *o*)

occupies anterior portion of middle field of proglottid in front of shell gland. Large yolk gland posterior to ovary and shell gland, irregular in shape, elongated transversely, with convex ventral surface and concave dorsal surface. Uterus (Fig. 16, *u*) developed as tube between anterior and ventral lobes of ovary. Gravid uterus fills up most of proglottid, extending beyond excretory canals on each side. Eggs oval (Fig. 7), with very thin membrane next embryo, followed by thick, smooth membrane 40 by 32 μ to 45 by 36 μ in diameter, and one or two outer membranes, very thin and wrinkled in preserved material. Diameter of outer membrane 65 by 40 μ to 60 by 45 μ ; at each pole of outer membrane a delicate appendage. Embryonal hooks 18 μ long. Embryo 32 by 22 μ in diameter.

2. *Morphology*: The scolex of the living worm shows up very prominently and can be used as a distinguishing feature. When first removed from the intestinal wall the suckers appear distinct and the neck is much narrower than the scolex. Soon after the removal it often contracts and takes on the appearance of a flattened bulb which includes the neck and anterior segments (Fig. 1). This feature is characteristic of this species and is a factor which alone assists very materially in distinguishing it from others that occur in chickens.

The rostrum or crown of the scolex is somewhat pointed when the rostellum is enclosed within its sheath (Fig. 2). The rostellum is an ovoid structure with a bulbous expansion at its anterior end. It has a length of 140 μ and a breadth of 60 to 65 μ at its anterior end. A crown of 18 hooks is arranged in a single row around the bulbular anterior end. The structure of the wall is of a fibrous nature and presents a transversely striated appearance due to contraction. In the interior of the rostellum the structure is a connective tissue mass with few cells, some of which possess long processes. The hooks (Fig. 8) are 30 μ in length with a long dorsal root and a short ventral root.

The rostellar sheath or sac (Fig. 3, *rs*) into which the rostellum is withdrawn is oval in shape and 230 to 240 μ in length by 80 to 90 μ in width at its broadest point. Histologically, the structure is that of a fibrous connective tissue type with spherical and spindle-shaped cells. The cells coming in contact with the rostellum, as

well as those on the outer edge of the sac, bear long processes. The outer layer of the rostellar sac is composed of longitudinal and oblique fibers of a muscular nature which probably have for their function the movement of the rostellum.

The four excretory canals, that have extended forward through the entire length of the body, unite in the scolex to form a ring (Fig. 3, *ex*), which lies in the tissue of the rostellar sac around the body of the rostellum.

The suckers are prominent. They are oval in shape and in preserved specimens measure 180 to 210 μ in length and from 135 to 175 μ in extreme breadth. In the center of each sucker there is a depression or an acetabulum, 30 to 40 μ in diameter. The entire inner surface of the suckers possesses minute hooklets or spines (Fig. 4) 1.5 to 2 μ long. These hooklets not only line the suckers but also extend over the entire surface of the scolex (Figs. 3, 5) and down onto the neck region; they disappear before reaching the first segment. They appear more distinctly on scolices that are somewhat contracted than on those that are well extended. These hooklets can be seen only in sections as they are too small to be distinguished readily in whole mounts.

Musculature: The longitudinal muscle fibers are arranged in bundles which are scattered, forming a loose irregular layer. The bundles are numerous and nearly of a uniform size. There are no transverse muscle fibers present except a few minute oblique fibers which connect some of the longitudinal fibers near the ends of the proglottids. Some dorso-ventral fibers are present, but they are not abundant.

Nervous System: The longitudinal nerve fibers are arranged in fiber tracts which approach the structure of a nerve cord. The individual fibers do not form a compact mass, but are more or less free in the tract. Nerve cells have no definite arrangement, but are situated irregularly along the fiber tract (Fig. 6). The nerve cells are somewhat spindle-shaped and quite large, being from 20 to 25 μ long by 6 to 8 μ wide with large nuclei. Transverse nerves are composed of individual cells with long processes extending transversely from the lateral fiber tracts. The transverse fibers are much scattered and have no definite arrangement except that they

are more numerous near the ends of the proglottids. Peripheral nerve cells are widely and irregularly distributed. They are more numerous at the anterior end of the proglottids, especially on the portion that is covered by the backward extension of the preceding segment.

Excretory System: The excretory system is fairly well developed in this form. The ventral canal (Fig. 14, *v ex*) is the larger, and has a diameter of 28 to 30 μ . A transverse canal unites the two longitudinal canals in each segment. The dorsal canals (Fig. 14, *d ex*) are much smaller, having a diameter of 6 to 8 μ and are not united by transverse connections. The four longitudinal canals extend anteriorly to the scolex where they unite to form a ring which lies in the rostellar sheath around the body of the rostellum. The vas deferens and vagina pass between the dorsal and ventral excretory canals.

Male Reproductive Organs: The testes vary in number, usually from 25 to 40, but in a few cases the number is much greater, being as high as 55 or 60. The testes are quite large, being from 40 to 55 μ in diameter, and are located in the posterior half of the proglottid (Fig. 14, *t*), posterior and lateral to the yolk gland. The testes are not arranged in layers, but are grouped in a more or less compact mass almost entirely within the limits of the excretory canals. The vas deferens (Fig. 14, *vd*) in the anterior third of the proglottid forms a coiled mass at the side of the ovary, from whence it passes laterad to the cirrus pouch as a convoluted tube. The portion of the vas deferens inside the cirrus pouch is coiled, varying in extent in different specimens (Figs. 14, 15). The vas deferens passes into the cirrus. There is no seminal vesicle formed by the vas deferens in the cirrus pouch nor are there any accumulations of sperm cells. The cirrus pouch (Fig. 15) is ovoid in shape and is from 75 to 90 μ in diameter. The wall is made up of layers of fibers which are both circular and oblique, forming a basket-like network which incloses the cirrus and a portion of the vas deferens. The outer wall of the cirrus pouch forms the inner wall of the deep genital cloaca. The cirrus is a compact structure from 50 to 65 μ long and lined with spines. It is a slightly curved structure passing from the cirrus pouch and curving posteriorly

toward the vagina which is directly posterior to it. The cirrus was not observed extending from the genital cloaca, but was noted in some specimens curving toward the vagina, though not passing into it. A few sperm cells were present in the vas deferens, also in the vagina and the seminal receptacle.

Female Reproductive Organs: The large ovary (Fig. 14, *o*) lies in the anterior third of the proglottid and extends transversely across the segment. It has a length of 300μ and a breadth of about 75 or 80μ at its broadest point. It is irregular in shape, being composed of a number of lobes. The end which is nearest the genital pore is smaller than the other, allowing room for the mass of coils of the vas deferens, the vagina, and the seminal receptacle. The ovary is concave on the dorsal surface and convex on the ventral. On the dorsal surface of the end nearest the genital pore is located the seminal receptacle and the vagina. The ova are large and very distinctly shown in the ovary (Fig. 16). Posterior to the ovary is the large yolk gland (Fig. 14, 16, *y*) which lies about the middle of the proglottid. It is irregularly elongate in shape and extends transversely across the segment, having a length of from 120 to 130μ and a breadth of from 35 to 50μ . Immediately in front of and dorsal to the yolk gland and posterior to the ovary is the shell gland (Fig. 14, *sg*) which is slightly ovoid in shape, 40 to 50μ in diameter. A small duct, the vitelline duct (Fig. 16, *v*), passes from the yolk gland through the shell gland from which it receives a duct. The combined ducts after passing through the shell gland unite with the oviduct (Fig. 16, *ov*) which appears as a curved tube leading from the ovary. These united tubes or ducts pass anteriorly and slightly ventrad into the uterus which develops as a blind tube in the region of the ventral lobes of the ovary. This blind tube (Fig. 16, *u*) grows in size and extends transversely across the segment. As it becomes larger the tube forms pockets which extend anteriorly and posteriorly and also dorsally, until it takes up the entire mass of the proglottid between the excretory canals. In gravid segments it even extends beyond the excretory canals. A small tube or duct, which is really the end of the vagina, connects the seminal receptacle with the yolk-shell gland duct and oviduct. This tube serves to carry the sperm to the eggs in the oviduct for fertilization. The

seminal receptacle (Fig. 16, *sr*) is a dilation of the vagina into an oval shaped structure which is about 50μ long and from 25 to 30μ in breadth at the widest part. From the seminal receptacle the vagina passes laterad, lying posterior to the cirrus pouch, and unites with the genital cloaca. The genital cloaca has its pore on the lateral margin near the anterior end of the proglottid. The pore is usually covered by the backward projection of the segment anterior to it. The vas deferens and vagina pass between the dorsal and ventral excretory canals and dorsal to the nerve tract. The vas deferens is dorsal and anterior to the vagina.

In the mature segments the uterus becomes filled with ova and it increases in size until it occupies the entire area between the excretory canals, even extending beyond the canals in the gravid proglottids. The uterus finally breaks up into compartments, each containing a single embryo. The embryos (Fig. 7) are about 32 by 22μ in diameter with onchospheric hooks 18μ long. Usually three membranes, but often four, enclose the embryo. The inner membrane is thin and closely surrounds the embryo; the next is heavy, being from 1.5 to 2μ thick, composed of fibrous layers with a few cells present. This layer is variable in thickness, depending considerably upon the amount of contraction of the segment, as it ranges in size from 40 to 32μ to 50 by 36μ , or it may be even slightly larger. Usually one (Fig. 7) and sometimes two thin membranes are found on the outside of the thick layer. These are often wrinkled and bear at each end an appendage formed from the outer membrane by which it is attached to the wall of the capsule or compartment of the uterus.

In this species the oldest proglottids drop off from the worm before they are fully mature. The embryos from the oldest segments on the worm do not show the characteristics of entirely mature ones, and there are distinct differences between them and those that have been separated from the worm for some time. Single proglottids that have separated from the worm are quite active and remain in the intestine for some time before passing out with the feces. Proof of this is furnished by the fact that a large number of the free proglottids are found in the intestine at any time. Even tho only a few worms are present in the intestine of a bird there is

usually a large number of free proglottids. If they did not remain in the intestine for a considerable length of time there would not be nearly as many. Further proof is furnished by the fact that the free proglottids have embryos which are mature, showing the oncospheric characteristics, while the oldest segments that are still attached to the worm have embryos that are not entirely mature. This same condition has been observed in *Davainea proglottina* as Blanchard (1891:435) states that the oldest proglottids separate from the others and remain in the intestine to become mature before passing out. The proglottids do not always separate from the worm singly, but may drop off in groups of three or four.

The fact that the proglottids separate from the worm before they are entirely mature is one of great importance in taking up experimental work for infection of intermediate hosts. If the embryos are fed to insects or other invertebrates before they are mature they will be digested, and thus infection cannot be produced.

B. CYSTICERCUS

The cysticercus of *Choanotænia infundibuliformis* was found in the abdominal region of the body cavity in the common house fly, *Musca domestica*. The flies had been fed on embryos from ripe proglottids of this species of worm, and at the end of twelve days were killed. The cysticerci appear to be nearly ripe or ready for transmission into the adult host. The time for the development of the cysticercoid varies with different species and under different conditions. Grassi and Rovelli (1892:85) found that *Davainea proglottina* developed from the onchosphere into a ripe cysticercus in less than twenty days. Schmidt (1894:9) found that the development of the cysticercoid of *Drepanidotænia anatina* (Krabbe) varied with the time of the year and the influence of the temperature. In the summer the embryo developed in an ostracod, *Cypris ovata*, into ripe cysticercoids in two weeks.

The cyst proper (Figs. 11, 12, *c*) containing the scolex is oval in shape, 220μ long and 120μ in diameter.

The bladder (Fig. 12, *b*) or tail, which is also oval in shape, is located against one side of the cyst and is somewhat flattened on that side. It is 220 to 230μ long and from 116 to 120μ in breadth.

The scolex is 80μ in breadth and 120μ in length; neck is 40μ in diameter and 30 to 35μ long; suckers are 55 to 60μ in diameter. The rostellum is 60μ long and 20μ in breadth, armed with a crown of 18 hooks arranged in a single row. These hooks (Fig. 9) are 30μ long with a long dorsal root and a short ventral root. The suckers are lined with numerous minute hooklets or spines 1.5 to 2μ long which extend over the edges of the suckers and also over the greater part of the surface of the scolex, including a part of the neck region. Schmidt (1894: 16) described cuticular hooklets on the suckers of *Drepanidotænia anatina*.

The size of the scolex may be somewhat variable as shown by those in the cysticercoids of *Drepanidotænia anatina* by Schmidt (1894: 10). In that species the intermediate host could be one of two or more species of crustaceans and the size of the cysticercoid varied with the size of the host in which it was parasitic.

The head of the rostellum is conical in shape, bearing a bluntly pointed apex anterior to the end of the dorsal roots of the hooks (Fig. 10, *r*). This part of the rostellum is composed of minute muscle fibers which are both circular and oblique. The rostellum is slightly broader below the circle of hooks as it is an oval shaped body.

The rostellar sac (Fig. 10, *rs*) is a deeply stained structure 10 to 12μ thick. It extends from 10μ below the hindermost part of the rostellum to the anterior extremity of the scolex, forming an oval shaped sac or sheath. It is composed of parenchymatous tissue with large heavily stained oval or spindle shaped cells which bear processes. The outer part of the sac is composed of a thin layer of fine fibers which help to give it a definite shape. At the lower edges of the sac the fibers are connected or associated to some extent with similar fibers that form the inner layer of the suckers. The anterior region of the rostellar sac, which forms the sheath for the free head portions of the rostellum, is constructed of an inner layer of fine fibers and an outer layer of large spindle-shaped cells, the most of which bear fibrous processes at one or both ends.

The suckers are composed of large spindle-shaped cells which are arranged perpendicular to the edge. These are heavily stained

and form a compact layer. The inner boundary of the suckers is composed of a layer of fibers which are both circular and oblique. Some of these at the upper edges are associated with similar fibers in connection with the rostellar sac.

The cyst is composed of two cell layers with an irregular cavity between them. The cells are large and irregular in shape with no special arrangement in the layer. Large intercellular spaces lie between the cells, thus forming a loose network structure, except at the base of the neck. At this point where the neck is attached to the inner layer of the cyst the cells are smaller and are in a compact mass. There is no definite boundary to the outer part of the inner layer as well as to the inner part of the outer layer of the cyst. Few cells with long connective processes extend across the cavity from one layer to the other. This then forms an irregular cavity (Fig. 11 *ca*) 2 to 20 μ in width between the two layers of the cyst. This is the primitive cavity of Grassi and Rovelli (1889: 373). The two layers of the cyst are formed apparently by a fold which extends upward and inward from the base of the neck, forming the gastrula cavity of Grassi and Rovelli (1889: 402, *g*) and enclosing the scolex. This cavity varies in width from 3 to 10 or 15 μ .

The bladder, an oval shaped structure, is located at one side of the cyst and is attached to it at the posterior end by a narrow connection (Fig. 12, *cn*). The posterior end of the cyst or the region caudad of the base of the neck is somewhat drawn out (Fig. 12). From this point is given off the attachment to the bladder or tail portion of the cysticeroid. The fact that this bladder is really a tail, even though it possesses a cavity, is shown by the presence of the onchospheric hooks, which are located at the end of the bladder opposite to that of the attachment of the cyst (Fig. 12, *oh*).

The order of arrangement of the onchospheric hooks is individual. In some specimens they are situated at the end of the bladder, while in others they are at the side. In some the arrangement is in a group, while in others they are in pairs. Some of my specimens show a pair of embryonic hooks in the layers of

the cyst between the base of the neck and the attachment of the bladder, while the other two pairs of hooks are located in the bladder.

The cavity of the bladder is formed apparently by a splitting or hollowing out of the cells of the tail, because the wall is continuous and of the same histological structure. The wall of the bladder is constructed of two layers, an inner cell layer and an outer cuticular layer. The outer cuticular layer is more or less striated on account of minute fibrils uniting it with the inner cell layer. Histologically, the structure of the inner layer is constructed of somewhat granular substance arranged in fibers forming a network which encloses clear spherical cells with large nuclei (Fig. 13). Outside of the cuticular layer is located the peritoneum of the host which lies upon the bladder and surrounds it as well as the cyst.

C. COMPARISON OF ADULT AND CYSTICERCUS

A comparative study of the adult and the cysticeroid shows the likeness which exists between them. The presence of the same number of hooks, having exactly the same size and shape as seen by comparing Figures 8 and 9. Minute hooklets of the same size are present in both cysticeroid and adult lining the suckers, the entire surface of the scolex and a part of the neck region. Rosseter (1891: 365) shows that the hooks on the rostellum and suckers of *Echinocotylus Rosseteri* undergo no changes during the act of transition from cysticeroid to adult stage. The rostellar sac is of the same general shape in both. The head of the rostellum is not expanded in the cysticeroid as in the adult because it has not functioned as yet. This corresponds to figures as shown by Schmidt (1894, Pl. VI, Fig. A) of the cysticeroid and Krabbe (1869, Pl. VI, Fig. 114) of the adult of *Drepanidotenia anatina*, and by Grassi and Rovelli (1892, Pl. IV, Fig. 7, 8) of the cysticeroid and Blanchard (1891: 16) of the scolex of *Davainea proglottina*. No measurements are given for the rostellum of either the cysticeroid or the adult by the above authors.

There is a great deal of difference in the size of the scolex between the cysticeroid and the adult. In my specimens the

scolex of the adult is between four and five times as large as that of the cysticeroid. The scolex of the cysticeroid has as yet not functioned so that the musculature of the organs is not developed as in the adult, consequently is not nearly as massive. The cells also are smaller than those of the adult.

Schmidt (1894: 10, 44) shows that the adult scolex of *Drepanidotænia anatina* is about three times as large as that of the cysticeroid. He also states that the size of the cysticeroid may vary with the size of its host.

Different forms become modified in changing from the intermediate to the adult hosts as shown by Schmidt (1894) in *Drepanidotænia anatina*, Rosseter (1891) in *Echinocotylus Rosseteri*, and Grassi and Rovelli (1892) in *Davainea proglottina*.

Onchospheric hooks in the wall of the tail are the same size (18μ) and shape as those of the embryos found in the mature proglottids.

A consideration of these factors of morphological significance which demonstrate the resemblances between the cysticeroid and adult, indicates clearly that this cysticeroid is the intermediate stage of *Choanotænia infundibuliformis*.

OTHER CHICKEN CESTODES IN THE UNITED STATES

1. *Davainea tetragona* (Molin 1858) Blanchard 1891

Diagnosis: Length 10 to 250 mm. by 1 to 2.5 mm. in breadth, varying with state of contraction. Scolex (Fig. 19) 175 to 215 μ in diameter, with retractile rostellum 25 to 50 μ in diameter, armed with single row of about 100 hooks. Rostellar hooks (Fig. 20) 6 to 9 μ long through longest axis, hammer-shaped, with long ventral root and short dorsal root, prong short and recurved. Suckers oval, 60 to 110 μ in diameter, armed with 8 to 10 rows of small hooks of various sizes. Acetabular hooks (Fig. 21) range in size from 4 to 8 μ through longest axis, having thorn-like prong, short dorsal root, and longer flattened ventral root, which is shorter than prong. Neck long and slender, but often as broad as head. Segments trapezoidal and imbricate, edges of strobila serrate. Oldest segments usually longer than broad, often bell-

shaped. Genital pores usually unilateral, situated one in each segment, at or in front of middle of lateral margin, frequently marked off by papilla. Male and female canals pass on dorsal side of nerve and excretory vessels.

Male Reproductive Organs: Testes 20 to 30 in median field surrounding female organs, most of them lying on aporose side of latter. Vas deferens situated in anterior third of segment, beginning near median line, and extending in much convoluted course laterally to base of cirrus pouch which it enters and, after a few coils in basal portion of latter, passes into cirrus. Cirrus pouch pyriform, 75 to 100 μ in length. Basal portion surrounded by prominent layer of longitudinal muscle fibers, neck with thick layer of transverse fibers. Cirrus without apparent spines.

Female Reproductive Organs: Ovary in middle of segment. Yolk gland posterior to ovary, irregularly reniform, slightly longer in its transverse axis, about 100 μ in diameter. Shell gland prominent, 50 μ in diameter, immediately in front of yolk gland. Vagina begins at genital pore, posterior to opening of cirrus pouch, at first very slender but at distance of 15 to 25 μ from genital pore swells out into thick-walled tube, functioning as seminal receptacle. This extends transversely across segment and joins oviduct on dorsal side of ovary near median line. Oviduct, after being joined in shell gland by vitelline duct, proceeds forward and ends on dorsal side of ovary. Definite and persistent uterus not developed. Eggs pass from distal end of oviduct, become imbedded in fibrous and granular or gelatinous mass which fills up most of segment. This mass divides into 50 to 100 portions to form egg capsules, each surrounded by membrane and containing 6 to 12 or more eggs. Egg is surrounded by three envelopes,—inner, close to onchosphere, often scarcely visible; middle layer or envelope much folded, giving appearance of network between inner and outer membranes; and smooth outer envelope. The onchosphere measures 10 to 15 μ in diameter; the outer envelope measures from 25 to 50 μ in diameter.

One point noted here that has not been mentioned before by other authors is that the genital pores are irregularly alternate.

They are usually unilateral. The existence of this irregularly alternate occurrence of the genital pores may be an anomaly, but it is rather frequent for such a condition.

2. *Davainea echinobothrida* (Mégnin 1880) Blanchard 1891

Diagnosis: Length up to 250mm; width 1 to 4 mm. Head (Fig. 22) 0.25 to 0.45 mm. in diameter, with retractile rostellum 100 to 150 μ in diameter, armed with crown of about 200 hooks arranged in two rows. Suckers round or oval, 90 to 200 μ in diameter, armed with 8 to 10 rows of hooks. Rostellar hooks (Fig. 23) similar to those of *Davainea tetragona*, but larger, measuring 10 to 13 μ in length. Acetabular hooks (Fig. 24) likewise similar to those of *D. tetragona*, but also larger; size variable, smallest being 7 or 8 μ in length and largest measuring from 14 to 16 μ . Neck thicker and generally shorter than *D. tetragona*, nearly equal to width of head. Strobila resembling that of *D. tetragona*, but serrate border more pronounced. Oldest segments in preserved specimens also differ from those of *D. tetragona*, being less elongate and frequently marked by median constriction. Owing to this constriction adjacent borders of most posterior segments pull apart in median line and remain joined only at sides, giving rise to median series of openings through posterior portion of strobila. Genital pores irregularly alternate, or sometimes almost entirely unilateral, situated one in each segment posterior to middle of lateral margin. Male and female canals pass on dorsal side of nerve and excretory vessels.

Male Reproductive Organs: Testes 20 to 30, arranged in median field surrounding female glands as in *D. tetragona*. Vas deferens lies in anterior third of segment much as in *D. tetragona*. Cirrus pouch flask-shaped, 130 to 180 μ in length. Basal portion globular or ovoid, surrounded by layer, about 10 μ thick, of longitudinal muscle fibers inside of which is a layer about 12 μ thick of transverse fibers. Neck of pouch measures 50 μ to 75 μ in length by 15 to 20 μ in diameter, surrounded by layer of transverse fibers thickened at distal end to form sphincter. According to Mégnin, the cirrus is armed with minute spines.

Female Reproductive Organs: Female organs same as in *Davainea tetragona*, and onchospheres (Fig. 25) are also similar

in structure and size, 14 to 15 μ in diameter. Onchospheric hooks 6 to 7 μ long. Egg capsules in groups of 6 to 12 or more, embedded in a fibrous gelatinous mass.

In the living specimens very little difference can be noticed except in size of the species *D. tetragona* and *D. echinobothrida*. They are both quite transparent and appear much alike in every respect in external appearance, except that *D. tetragona* is slightly more transparent, while the oldest segments of *D. echinobothrida* have very distinct median constrictions between them, appearing almost as a series of openings.

The chief differences between *D. tetragona* and *D. echinobothrida* are that in the latter the animal is larger, the hooks are more numerous and larger, and the structure and size of the cirrus pouches show a very distinct difference. There is also a difference in the pathological effect of these spiny-suckered forms. *D. echinobothrida* produces large nodules or ulcers in the intestinal wall. The scolex bores through the mucosa of the intestine and in some cases nearly through the muscular coats. This disease in fowls is termed "nodular tæniasis", as described by Moore (1895: 1), and is often mistaken for other diseases.

3. *Davainea cesticillus* (Molin 1858) Blanchard 1891

Diagnosis: Length 10 to 125 mm. Maximum width 1.5 to 3 mm. Head cylindrical (Fig. 28), sometimes spheroidal, 0.3 to 0.6 mm. wide and 0.2 to 0.4 mm. long. Suckers unarmed, about 100 μ in diameter. Rostellum broad and flat or hemispherical, 0.25 to 0.35 mm. wide, armed with a crown of 200 to 300 hooks which are very unstable and easily lost, arranged in two ranks. Hooks (Fig. 29) 8 to 12 μ long with short dorsal root and long ventral root. Neck very short. Anterior segments three to five times as broad as long; the following increase in size until they become equal in length and breadth and finally even longer than broad; borders overlapping. Genital pores irregularly alternate, one in each segment, somewhat in front of middle of lateral margin in young segments and nearer the middle in older segments. Vagina and cirrus pouch pass dorsal of the two excretory canals and nerve.

Male Reproductive Organs: Testes (Fig. 17, *t*) 20 to 30 in number in posterior portion of segment. Vas deferens much coiled before entering base of cirrus pouch, also coiled within latter. Cirrus pouch ellipsoidal, 120 to 150 μ long by 55 to 70 μ wide. Cirrus when protracted 10 μ in diameter, armed with minute spines, and with bulbous enlargement 20 μ in diameter at its base, where it becomes continuous with cirrus pouch.

Female Reproductive Organs: Vagina enlarged before reaching median line into small seminal receptacle (Fig. 17, *sr*). Ovary occupies middle field in front of testes. Yolk gland and shell gland posterior to ovary, ventral and dorsal, respectively, in relative position. Uterus at first in front of ovary as cord of cells; gradually increasing in size, finally occupies most of segment and frequently extends laterally beyond excretory canals. In oldest proglottids it becomes divided into compartments, or capsules, each containing a single egg. Embryo (Fig. 30) 36 by 27 μ in diameter, with very thin membrane closely adherent to surface. Embryo further enveloped by thicker, smooth fibrous membrane, oval in shape, 45 to 40 μ in diameter, with filament at each pole attaching to thin outer wrinkled membrane about 35 by 50 μ in diameter: finally egg is surrounded by capsule composed of outer and inner membrane, latter closely adherent to or fused with outer egg membrane; and former more or less widely separated from latter and connected with it by number of septa.

One of the principal points noted here that is not mentioned by other authors is the size of the rostellar hooks. In my specimens they seem to be somewhat larger than those described by others. They have been described as being 8 to 10 μ long, while my forms show many of them to be distinctly 12 μ in length. A second point noted here is the method of the development of the uterus. The uterus develops in front of the ovary. It first appears as a solid cord of cells connected with the united ducts of the ovary, shell gland, and yolk gland. The solid cord of cells which later gives rise to the uterus becomes hollow and appears as a blind sac or tube. This then grows in size, forming pockets, and finally fills up the entire proglottid.

This form is one of the most common chicken tapeworms and is the most easily recognized. It can be identified by the head with its broad, flat rostellum which shows up very prominently; the width of the most anterior segments is usually equal to or greater than the width of the head, and the eggs are distributed in individual egg capsules in mature proglottids.

4. *Hymenolepis carioca* (Magalhaes 1898) Ransom 1902

Diagnosis: Length 30 to 80 mm. Breadth at neck 75 to 150 μ , at posterior end 0.5 to 0.7 mm. Segments three to five times or more broader than long throughout strobila. Head (Fig. 26) flattened dorso-ventrally, 140 to 160 μ long, 150 to 215 μ wide and 100 to 140 μ thick. Suckers shallow, 70 to 90 μ in diameter, unarmed. Rostellum unarmed; in retracted position 25 to 40 μ in diameter and 90 to 100 μ in length, with small pocket opening to exterior in anterior position. Unsegmented neck portion of strobila 0.6 to 1.5 mm. long. Genital pores almost entirely unilateral, a single pore being located in each segment slightly in front of middle of right-hand margin.

Male Reproductive Organs: Testicles three in number, normally two on left and one on right of median line. On dorsal side of inner end of cirrus pouch vas deferens is swollen into prominent seminal vesicle (Fig. 18, *sv*) which may attain a size of 70 by 50 μ . Cirrus pouch (Fig. 18, *cp*) in sexually mature segments 120 to 175 μ long by 15 to 18 μ in diameter; almost cylindrical, slightly curved toward ventral surface of segment; on outer surface about 20 longitudinal muscle bands, 2 to 3 μ in thickness, very prominent in cross section; vas deferens enlarged within cirrus pouch to form small seminal reservoir occupying proximal two-thirds of pouch; distal third of portion of vas deferens within pouch very slender, about 1 μ in diameter and functions as cirrus. Genital cloaca 12 to 36 μ deep.

Female Reproductive Organs: Opening of vagina in floor of genital cloaca, ventral and posterior to cirrus opening. First portion of vagina very narrow, 1 μ in diameter. Small vaginal sphincter 8 to 10 μ from vaginal opening. On inner side of sphincter vagina gradually increases in diameter, and in sexually mature

segments swollen into prominent seminal receptacle (Fig. 18, *sr*) which extends forward to anterior border of segment and inward considerable distance beyond proximal end of cirrus pouch. Ovary faintly bilobed or trilobed in posterior half of proglottid. Yolk gland spherical or ovoid, 30 to 40 μ in diameter, situated near median line of segment, posterior and dorsal of ovary. Uterus at first solid cord of cells extending transversely across segment along anterior border of ovary; becomes hollowed out and grows backward on dorsal side of ovary; in gravid segments occupies nearly entire segment and filled with eggs. Eggs (Fig. 27) in gravid uterus spherical or oval, with four thin membranes, the two middle membranes often approximate to form thick layer which shows somewhat of a cellular or coarse granular structure. Diameter of outer membrane 36 by 36 μ to 75 by 70 μ , of outer middle membrane 30 by 30 μ to 65 by 60 μ , of inner middle membrane 26 by 26 μ to 40 by 35 μ , of inner membrane 24 by 16 μ to 29 by 21 μ . This membrane often lies so close to onchosphere that it can scarcely be distinguished from edge of embryo. Onchosphere is 18 by 14 to 27 by 19 μ in diameter; length of embryonal hooks 10 to 12 μ .

This form is thread-like and usually occurs in great numbers. It is very delicate and fragile and can be recognized by that fact alone, as it is the most fragile of the chicken forms known.

SUMMARY

1. By morphological comparison of the cysticercoids produced experimentally in flies and adult of *Choanatenia infundibuliformis* they are shown to be identical.

2. Morphological points noted are the presence of minute hooklets on the suckers and entire surface of scolex in *Choanatenia infundibuliformis*. The manner of development of uterus in the same species is by means of a blind tube which grows in size, forming pockets, and later breaks up into small compartments. In *Davainea tetragona* the genital pores were found to occur irregularly alternate in the proglottids. The hooks on the rostellum of *Davainea cesticillus* were found to vary in length from 8 to 12 μ . The uterus in development first appears as a solid cord of cells which becomes hollow and in growing forms pockets, filling the entire proglottid.

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EXPLANATION OF PLATES

Unless otherwise stated all drawing were made with the aid of a camera lucida.

ABBREVIATIONS

<i>b</i> —bladder	<i>rs</i> —rostellar sac
<i>c</i> —cyst	<i>sg</i> —shell gland
<i>ca</i> —primitive cavity	<i>sr</i> —seminal receptacle
<i>cn</i> —connection of bladder with cyst	<i>sv</i> —seminal vesicle
<i>cp</i> —cirrus pouch	<i>t</i> —testes
<i>dex</i> —dorsal excretory canal	<i>u</i> —uterus
<i>ex</i> —excretory ring in scolex	<i>v</i> —vitelline duct
<i>o</i> —ovary	<i>va</i> —vagina
<i>oh</i> —onchospheric hooks	<i>vd</i> —vas deferens
<i>ov</i> —oviduct	<i>vex</i> —ventral excretory canal
<i>r</i> —rostellum	<i>y</i> —yolk gland

PLATE V

CHOANOTAENIA INFUNDIBULIFORMIS

- Fig. 1. Scolex much contracted. x40
 Fig. 2. Scolex normal extension. x145
 Fig. 3. Longitudinal section of scolex, showing rostellum and rostellar sac. x425
 Fig. 4. Section of portion of sucker, showing hooklets. x425
 Fig. 5. Section of portion of wall of scolex, showing hooklets. x425
 Fig. 6. Longitudinal nerve tract, showing nerve cells with processes. x650

PLATE VI

- Fig. 7. A, B, C, D. Embryos from mature proglottid. x425
 Fig. 8. Hooks from rostellum of adult. x425

CYSTICERCUS OF CHOANOTAENIA INFUNDIBULIFORMIS

- Fig. 9. Hooks from rostellum of cysticercus. x425
 Fig. 10. Section through scolex, showing rostellum with hooks and rostellar sac. x425
 Fig. 11. Section through scolex and cyst, showing suckers with hooklets, structure of cyst and primitive cavity between layers of cyst. x425
 Fig. 12. Reconstruction of cysticercus with cyst and bladder or tail, showing scolex in cyst and onchospheric hooks in bladder. x145
 Fig. 13. Section of wall of bladder, showing histological structure and peritoneum of host. x425

PLATE VII

- Fig. 14. *Choanotaenia infundibuliformis*. Reconstruction of mature proglottid, showing reproductive organs, excretory vessels, and nerve. x145
- Fig. 15. *C. infundibuliformis*. Reconstruction of cirrus pouch showing cirrus and vas deferens, also part of vagina in connection with cloaca. x310
- Fig. 16. *C. infundibuliformis*. Reconstruction of female reproductive organs, showing part of ovary, yolk gland, shell gland, oviduct, vitelline duct, uterus, and connection of ducts with uterus and seminal receptacle. x310
- Fig. 17. *Davainea cesticillus*. Reconstruction of mature proglottid, showing reproductive organs and excretory vessels. x145
- Fig. 18. *Hymenolepis carioca*. Reconstruction of mature proglottids, showing reproductive organs from ventral view. x145

PLATE VIII

- Fig. 19. Scolex of *Davainea tetragona*. x145
- Fig. 20. Hooks from rostellum of *D. tetragona*. x425
- Fig. 21. Hooks from suckers of *D. tetragona*. x425
- Fig. 22. Scolex of *Davainea echinobothrida*. x145
- Fig. 23. Hooks from rostellum of *D. echinobothrida*. x425
- Fig. 24. Hooks from suckers of *D. echinobothrida*. x425
- Fig. 25. Embryos of *D. echinobothrida*, showing capsule and fibrous gelatinous mass in which it is embedded. x425
- Fig. 26. Scolex of *Hymenolepis carioca*, after Ransom.
- Fig. 27. A, B, C, D. Embryos of *Hymenolepis carioca*, showing enveloping membranes. x425
- Fig. 28. Scolex of *Davainea cesticillus*. Free-hand drawing of living specimen well extended, showing rostellum.
- Fig. 29. Hooks from rostellum of *D. cesticillus*. x425
- Fig. 30. A, B, C, D. Embryos of *D. cesticillus*, showing enveloping membranes. x425

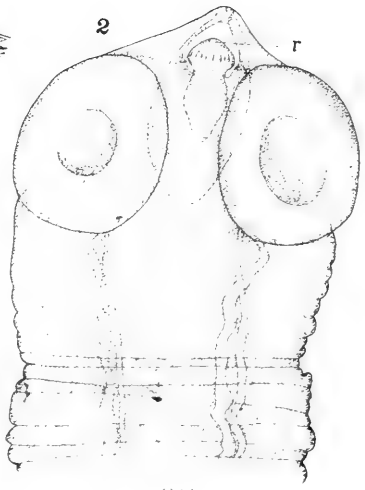
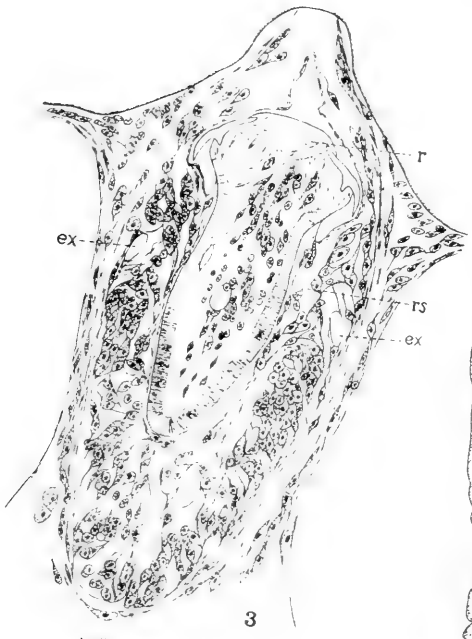
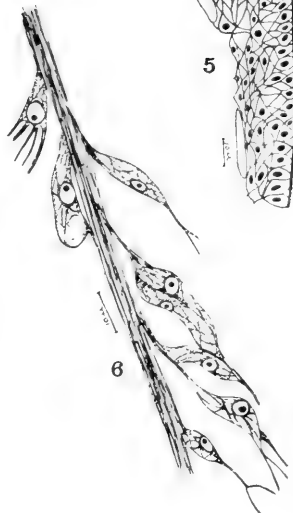
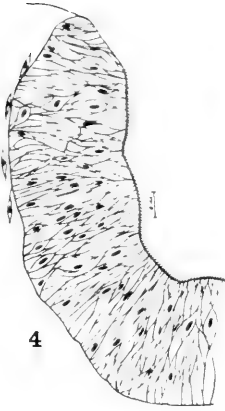
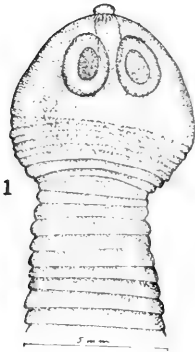


PLATE V

100 μ

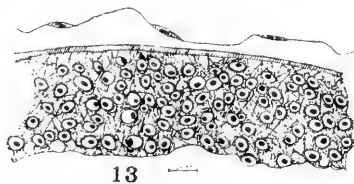
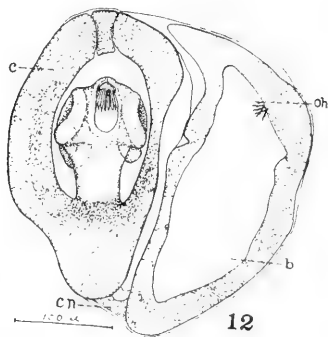
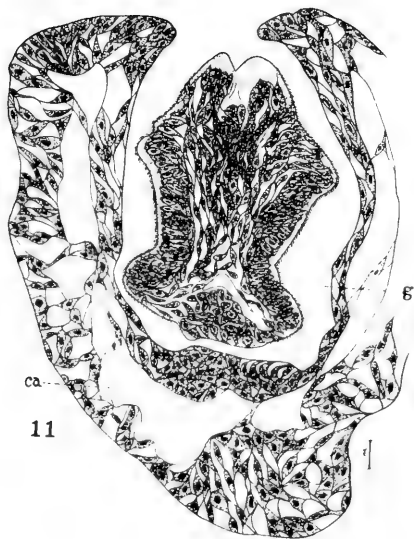
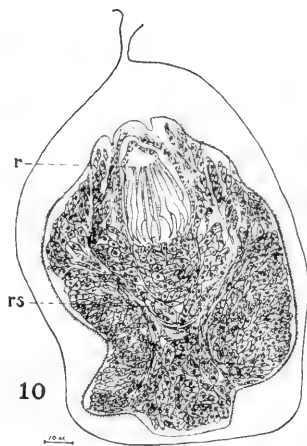
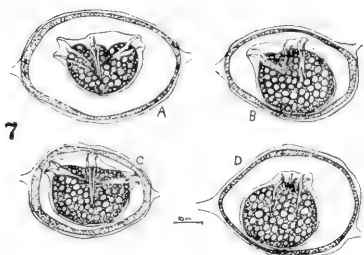


PLATE VI

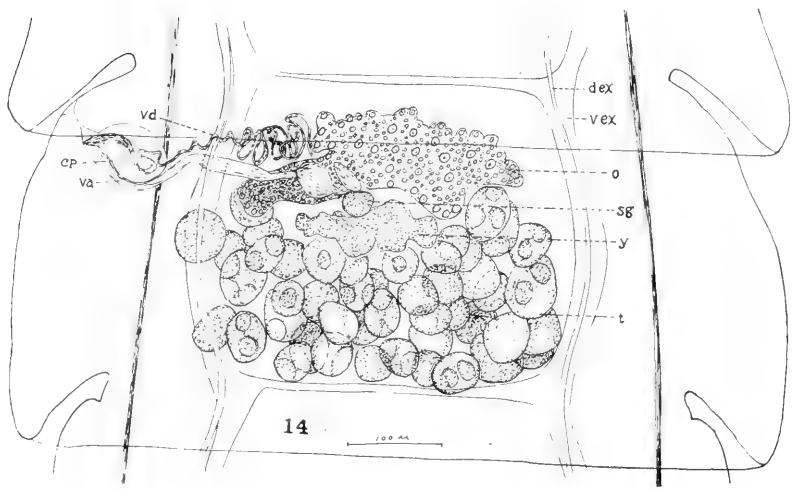
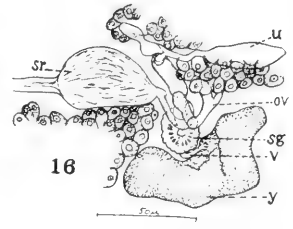
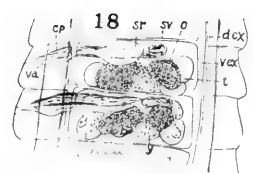
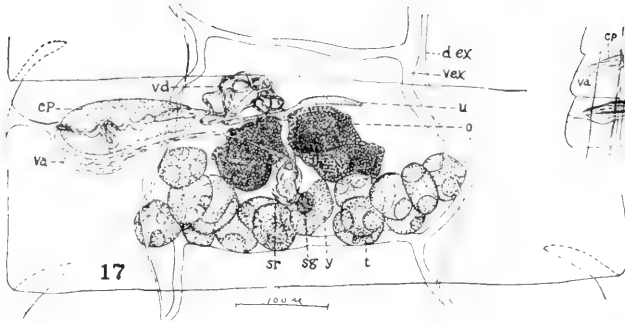


PLATE VII



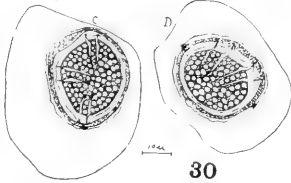
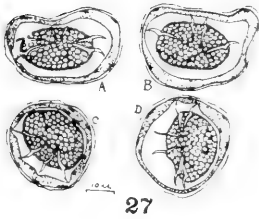
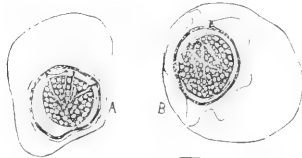
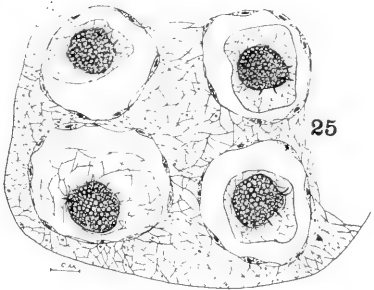
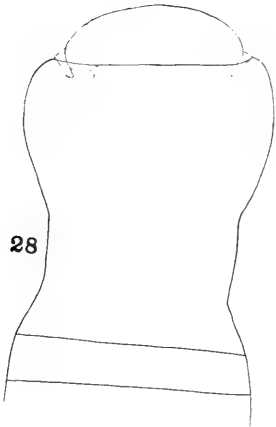
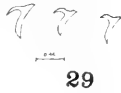
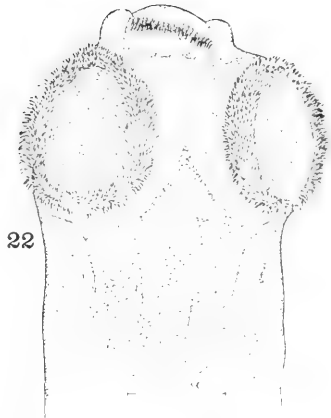


PLATE VIII



A PRELIMINARY STUDY OF THE SPERMATOGENESIS OF *BELOSTOMA* (ZAITHA) FLUMINEA

By A. M. CHICKERING

The study of *Belostoma fluminea* was begun during the summer of 1914 at the University of Wisconsin under the direction of Prof. M. F. Guyer to whose help and criticism the writer owes much. I also wish to take this opportunity to express my gratitude to Prof. H. D. Densmore of Beloit College for the loan of expensive and indispensable apparatus.

MATERIAL AND METHODS

Belostoma fluminea is the common, giant water bug of our shallow ponds and slowly moving streams. This form is very common in the region around Madison, Wisconsin and has been taken in large numbers, especially during the month of July.

Nymphs and adult males, which are often taken bearing the egg-cluster on their backs, were used to obtain the necessary material. Some of the adults show the whole general history of the germ cells from spermatogonia to mature spermatozoa with great clearness. By careful study the seriation is placed beyond reasonable doubt by virtue of the arrangement of the cells in cysts in a nearly progressive series from one end of the testes to the other.

On account of their extreme delicacy and transparency some trouble was experienced at the beginning in removing the testes from their position in the abdominal cavity to the fixing fluid. Later, however, this was overcome by squirting the fixing fluid directly on to the testes after removal of the dorsal abdominal wall. After a few moments the organs stood out sharply and could be transferred easily to the final fixing fluid without injury.

The best results in fixing were obtained with Gilson's and Bouin's fluids.

The iron-hæmatoxylin method of staining has proven very satisfactory for this work. Other stains have been tried with

varying degrees of success but all the slides used in preparing this paper were stained with Haidenhain's iron-haematoxylin. Sections were cut from four to six μ in thickness.

Smears have been almost uniformly unsuccessful. In a few cases smear slides of fair quality were obtained but as yet no careful study has been made of them. The method of preparation as described by Morse ('09) gave better results than any other. The testes are simply removed to a slide and the cysts pricked or teased gently. This allows the contents to run evenly over the slide which is afterwards dried and stained without the use of any fixing fluid.

The works cited at the end of this paper are only those which have been especially helpful to the writer in preparing this brief paper. No attempt has been made to append a complete list of papers on this subject.

SPERMATOGENESIS

I. Spermatogonial stages

The spermatogonial divisions are very numerous in this material, particularly so in the testes taken from nymphs. But usually the chromosomes are so crowded together in these division stages as to prevent any accurate counting or study of the separate chromosomes. In fact the writer has been able to find only four metaphase plates in which the chromosomes could be effectively studied. These were all found in the testes of a single specimen and are probably undergoing final division before synapsis.

The two cells shown in figures 1 and 2, have the chromosomes arranged in a very flat equatorial plate in a plane almost parallel to the plane of the section. Both show twenty-four chromosomes, and in each case there are four large bean-shaped chromosomes, eighteen of intermediate size and spherical or ellipsoid in shape, and two very small ones. Nearly all are connected to one or more of the others by means of delicate filaments.

The other two cells mentioned above show, respectively, twenty-three and twenty-two chromosomes. In the first case one of the small pair is missing and in the second both are missing. These are so small and the chromosomes so numerous that they

may be covered up easily and so escape detection. From the number and behavior of the chromosomes in the maturation divisions one would expect the spermatogonial or unreduced number to be twenty-four. The writer considers it fair to conclude from the evidence at hand that such is the case.

It is quite evident from inspection of figures 1 and 2, that three pairs of chromosomes may be identified at this stage. The xy-pair, which becomes so apparent in the second maturation division, is not distinguishable nor so far has it been possible to recognize more pairs of chromosomes in this stage.

II. *Synaptic and Post-Synaptic Stages*

It has thus far been found impossible to obtain satisfactory results with *Belostoma* in attempts to work out the history of the chromosomes during synapsis and the early prophase stages of the primary spermatocytes; hence no attempt will be made to describe these in any detail, only a brief outline being given. I hope, however, to obtain suitable smear material at a later date and make a careful study of the stages necessarily slighted now.

Immediately following the last spermatogonial division the chromosomes loosen up and flow together into a rather lightly staining, confused network (Fig. 3). This stage is of short duration, passing quickly into what is probably a spireme stage but since no suitable material is available no drawings have been made from this point to synizesis (Fig. 4). The threads making up the synaptic knot are so crowded and tangled that usually only the ends of the threads can be seen projecting beyond the heavily stained mass. The knot is usually placed at one pole of the nucleus and often a plasmosome is visible in these stages.

After the synizesis stage the threads quickly spread apart throughout the nucleus. They now appear very thick and heavy, and stain more deeply than in the preceding stages (Fig. 5-6). The plasmosome attains its maximum size during this stage and gradually diminishes and finally disappears during the early prophases.

In the later part of this period some nuclei appear to show a divided condition of the separate threads even in sections and in the best smears the threads are distinctly divided by a longitudinal cleft (Fig. 7). Usually in the sections there are shown

two threads which are much longer and broader than any of the rest, presumably corresponding to the two large chromosomes of the first division and made up of the four bean-shaped spermatogonial chromosomes.

No evidence has yet been adduced to show that the sex-chromosomes in *Belostoma* are present in the form of chromatic nucleoli during the growth period as described by Wilson ('12) in the spermatogenesis of *Lygæus* and *Oncopeltus* although the general history of the chromosomes seems to be very similar to that of the two forms mentioned.

Before the prophase stages begin there is interpolated a "confused" stage which results from a fading out and ultimate disappearance of the heavy, divided threads. This process gives rise to a nucleus which is traversed throughout by a loose network of chromatin material consisting of much finer, irregular threads the boundaries of which can not be made out. This goes on from Figure 8 until the network is reduced to an exceedingly confused condition. In this condition the threads stain so lightly and are so interlaced as to make it impossible to see the separate elements at all. This "confused" stage is of relatively long duration, but finally condensation starts and the network is reorganized into the tetrad rods, rings and V's (Fig. 9-9A).

III. Chromatoid Bodies

In the confused stage are *clearly* seen for the first time (further work seems likely to disclose their presence earlier) bodies which have been called the chromatoid bodies (Wilson '13 and Fasten '14).

The larger of these bodies which becomes very prominent in the division of the primary spermatocyte attracted my attention when the work on *Belostoma* was first started. It then seemed to be an x-chromosome and for a time the work was carried on with that idea. The reader will readily see the resemblance of figures 14-23 to several already published by numerous workers demonstrating the presence of an odd chromosome. In the case here presented this idea must be abandoned. The chromatoid bodies are seen in Fig. 8 outside of the nuclear membrane and therefore

not directly connected with the chromatin elements. They are never found within the spindle although they are sometimes so close to the group of chromosomes that they are indistinguishable in polar views of metaphase plates, and in this way sometimes are very confusing in taking chromosome counts. They do not occur in all cells and when present behave irregularly. With iron-hematoxylin they are stained like the chromosomes.

IV. Primary Spermatocytes

In the early tetrad stages of the chromosomes two rings are very large and prominent. They are in practically every cell and seem to give rise to the two large chromosomes of the mature spermatocyte by a decrease in the diameter and a closing up process. There are other smaller rings sometimes showing but they do not seem to be constant. The rods seem to shorten and thicken to form the final chromosomes; the same process probably takes place with the Vs, and the rings apparently close up to form such tetrads as shown in Figs. 10-11. Such figures are very common in my material and may be favorable for working out the details of the changes taking place here.

All of these figures finally condense into dumb-bell shaped bodies (Fig. 12). This shape is retained throughout the first division, the constriction marking the plane of cleavage.

Very shortly after the stage shown in the last figure the nuclear membrane begins to break down, the spindle forms at the two poles, the chromosomes take up a position in the equatorial plane and the cell is ready for division.

The chromatoid bodies remain of about the same size until the spindle begins to be formed and then a decided increase in the size of at least one of them is plainly seen (Fig. 13-15.)

The stages from this point onward show with diagrammatic clearness. There are thirteen chromosomes in the equatorial plate of the primary spermatocyte. This number is one more than half the spermatogonial number. The arrangement of chromosomes is inconstant, no two plates showing the same placing with respect to each other. There are two large, ten intermediate and one very

small chromosomes, they all divide equally in this division, carrying over thirteen in every case to the secondary spermatocytes (Figs. 14-24).

These stages have been carefully searched to find the sex-chromosomes and to determine their behavior but without success.

The chromatoid bodies are seen here (Figs. 14-24) at their maximum size. Usually there is only one present, but quite often there are two and more rarely three. In a few cases four have been seen. Occasionally one or more of the number has an irregular shape (Figs. 14, 20 and 22).

When division takes place these bodies are not apportioned evenly but may all be retained in a single secondary spermatocyte, or one or more may go into each. No regularity seems to obtain in the distribution of these bodies. Given three or four of different sizes and a large number of types of secondary spermatocytes may be derived if classified on the basis of the kind and number of chromatoid bodies they possess.

V. *The Interkinesis*

It seems to be a well established fact that in the interkinesis there is no more than a slight pause between the telophase of the first and the prophase of the second divisions. The chromosomes are much crowded together, but enough is evident to show that they retain their individuality and are not in any degree reorganized into a nucleus (Fig. 25). The centrosomes also divide in late anaphase and are already well moved apart, accompanied by small spindles, in the late telophase.

Whether the grouping of chromosomes is changed in this stage is uncertain, but the probability is that the new grouping assumed in the next metaphase takes place during the prophases of the same division.

VI. *Secondary spermatocytes*

In the prophases of this division the spindle is very rapidly built up while the chromosomes which were so crowded before now spread apart and assume a *grouping very different from that in the first division*. Eleven of the chromosomes are arranged in a circle at the equator of the spindle while two remain in the center

of the circle forming an xy-pair of sex-chromosomes. In polar views only twelve chromosomes are usually visible (Figs. 26-28). Lateral views of the same stage show however, that what appears to be a single chromosome in polar views is in reality the xy-pair, the members of which are united end to end. (Fig. 29).

All of the stages through inter-kinesis leading up to the metaphase in which the sex-chromosomes are seen united have been studied to see how complete the conjugation between the two is effected, and such a process seems to be limited to a simple end to end union as described above. This union exists only for a short time before the final separation and seems to take place while the chromosomes are rearranging themselves in their new grouping during the later prophase stages.

By a comparative study of Figs. 23-25, and 29-30, it will be seen readily that a rotation of each chromosome through about ninety degrees takes place while the spindle is being formed, so that the long axis of each becomes parallel instead of perpendicular to the long axis of the spindle. A rotation of the entire group may also take place as described for *Oncopeltus* and *Lygaeus* (Wilson '10) although it seems more probable that the virtual rotation is accomplished by the relative change in position of the separate chromosomes as they assume their new grouping.

In the latter part of this period the sex-chromosomes have separated and are on their way to the opposite poles of the spindle before the others have even started to divide at all (Fig. 30). This aptitude for the sex-chromosomes seems to be a common phenomenon and has been described by numerous workers.

The chromatoid bodies present about the same appearance as in the primary spermatocytes. There are many cells without any chromatoid body, many more with only one large one, and others with two or three.

VII. *The Spermatids*

When division of the secondary spermatocytes takes place twelve chromosomes are delivered to each spermatid, but one-half of them receives eleven ordinary chromosomes plus the x-chromo-

somes while the other half receives eleven plus the y-chromosome (Figs. 32-34).

In studying the cysts containing spermatids it is very evident that those containing no chromatoid bodies are most numerous, those with one fairly common and those with two or more are much less common. In general, too, the size of these bodies is somewhat less than in the primary spermatocyte although occasionally one large one seems to be retained throughout at its maximum size (Figs. 35-38).

VIII. Summary

1. The spermatogonial number of chromosomes in *Belostoma (Zaitha) fluminea* is twenty-four.

2. Only general facts have been determined in regard to synapsis and the post-synaptic stages. During the post-synaptic period a double nature of the chromosome threads is evident.

3. There is a "confused" stage just previous to the prophases of the first division.

4. The chromatoid bodies appear in this "confused" stage for the first time. Proof that they originate from the cytoplasm is not lacking because they are plainly seen outside of the nuclear membrane in this stage.

5. Tetrads appear in the form of rings, Vs, and rods, and all become dumb-bell shaped, by continued condensation, at the time that they enter the spindle.

6. The first maturation division is an equational division. Polar views show thirteen chromosomes, which number is one-half the spermatogonial number plus one.

7. The chromatoid bodies are at their maximum size in this and the following division and generally grow smaller from that point onward.

8. The interkinesis is of short duration. No nuclear vacuole is formed, the chromosomes maintaining their individuality throughout.

9. When the chromosomes arrange themselves in the metaphase of the second division an entirely new arrangement is assumed and an xy-pair of sex-chromosomes can be identified.

10. Twelve chromosomes are delivered to each spermatid in the second division, one-half receive in addition to the eleven ordinary chromosomes an x- and the other half a y-chromosome.

11. The chromatoid bodies behave irregularly all along. Some spermatids have none, others have one and still others in decreasing proportions have two or three.

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EXPLANATION OF PLATES

All figures are from *Belostoma (Zaitha) fluminea* are drawn with a camera lucida and are magnified about 1350 diameters. Unless otherwise stated the figures are not intended to show *all* the nuclear components.

PLATE IX

1-2. Polar views of spermatogonial metaphases, showing twenty-four chromosomes.

3. Spermatogonial telophase (probably the last spermatogonial division).

4. Synzinesis stage with a plasmosome.

5-6. Post-synaptic nuclei showing the heavy undivided chromosome threads and plasmosomes.

7. A little later stage showing the divided threads. From a smear-preparation. The cell is somewhat distorted.

8. The confused stage, showing three chromatoid bodies outside of the nuclear membrane.

9-9A. About middle prophase of the first spermatocyte division, showing the various forms of the chromosomes during condensation, and the chromatoid bodies.

10. Later prophase of the same division, showing the tetrads and only one chromatoid body.

11. A very clear tetrad.

PLATE X

12. A late prophase of the first division showing the chromosomes organized into dumb-bell forms.

13. Very late prophase, showing all the chromosomes and two chromatoid bodies.

14-17. Polar views of metaphases of the first division, showing thirteen chromosomes and either one, two, or no chromatoid bodies.

18-21. Lateral views of the same stage with a varying number of chromatoid bodies.

22-23. Anaphases of the first division illustrating the irregularity in the behavior of the chromatoid bodies during division of the cell.

PLATE XI

24. Telophase of the first division. Here the two secondary spermatocytes have each received one chromatoid body. The four small bodies are the centrosomes already divided preparatory to the next division.

25. Polar view of the same, showing the crowded condition of the chromosomes.

26-28. Polar views of secondary spermatocytes. All the ordinary chromosomes are here arranged in a circle with the sex-chromosomes in the center and chromatoid bodies without the circle.

29-30. Lateral views of the same, showing the xy-pair of sex-chromosomes in the center.

31. Same stage without the sex-chromosomes.

32. Anaphase of the second division, showing the x- and y- chromosomes going into different spermatoids and slightly in advance of the others.

33-34. Polar views of the same, showing the small y- and the larger x- chromosomes in different spermatids. These cells were somewhat tilted so that in drawing the sex-chromosomes are made to appear displaced from their normal position.

35. Polar view of a spermatid with the chromosomes crowded in telophase, and two chromatoid bodies.

36. Lateral view of the same stage showing one spermatid receiving three chromatoid bodies and the other, none.

37-38. Later views of spermatids just before metamorphosis starts.

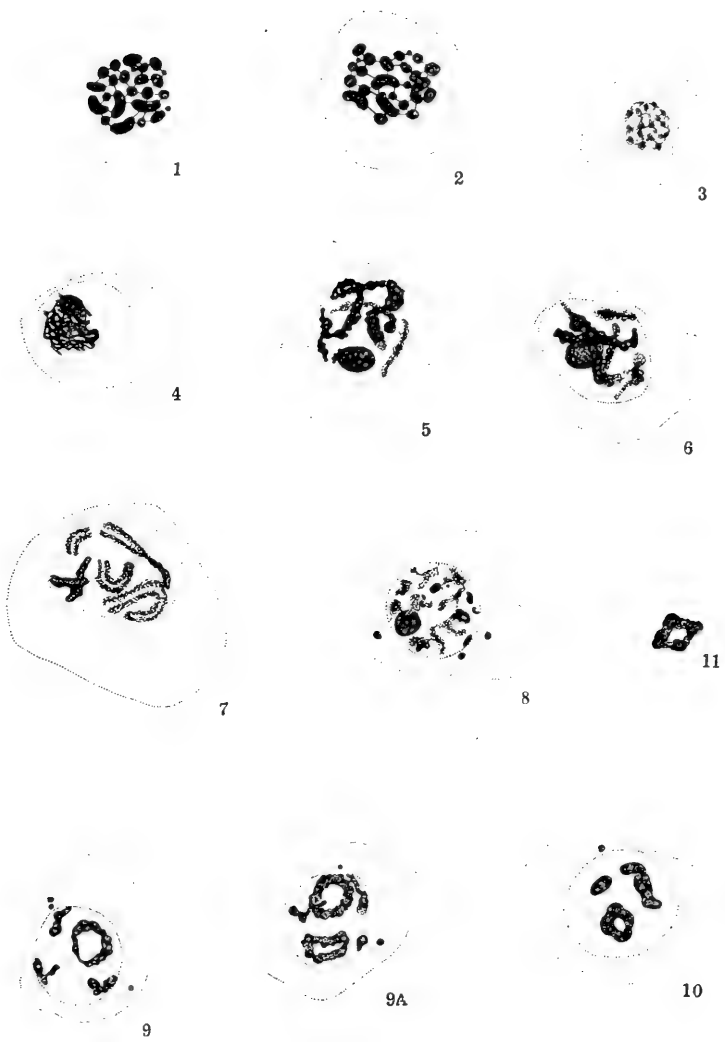


PLATE IX



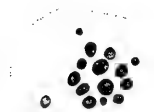
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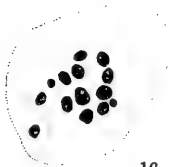
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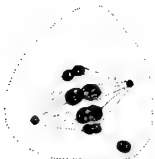
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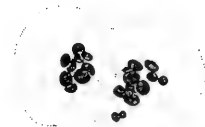
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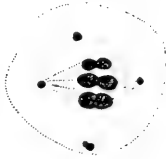
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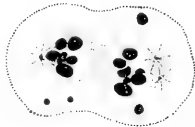
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PLATE X



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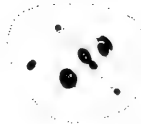
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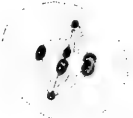
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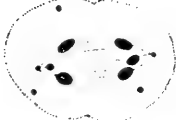
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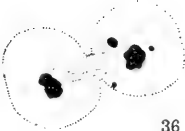
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PLATE XI

DEPARTMENT OF NOTES, REVIEWS, ETC.

It is the purpose, in this department, to present from time to time brief original notes, both of methods of work and of results, by members of the Society. All members are invited to submit such items. In the absence of these there will be given a few brief abstracts of recent work of more general interest to students and teachers. There will be no attempt to make these abstracts exhaustive. They will illustrate progress without attempting to define it, and will thus give to the teacher current illustrations, and to the isolated student suggestions of suitable fields of investigation.—[Editor.]

A SYSTEM FOR RECORDING CYTOLOGICAL MATERIAL, SLIDES AND LOCATIONS ON THE SLIDES

The following contribution is offered in full recognition of the fact that many cytologists already have in use excellent methods of recording their material and slides. Indeed many of the most essential details of the present system of recording slides have been taken over from a method in use by McClung, for which privilege the writer acknowledges his indebtedness. The system to be described has stood the test of the writer's use in all particulars and it is offered here in hope that it may serve as a suggestion for cytologists, who as yet have no recording method, on which to base a system serving their own particular needs. If the scheme here outlined is impractical for certain workers this note will at least serve to indicate the requirements that a cytological recording method must meet to be really efficient.

Two sets of cards are used, one to record the gross material and the other the slides and locations on the slides. On the former card (Fig. 1) are all the notes concerning the material from the fresh condition until it is embedded. On this card is to be found:

1. the serial lot number.
2. the material, the animal or plant from which it was taken, the age and other notes of possible interest.
3. place of collection and condition of obtaining the material.
4. dates of fixation.
5. fixing fluid and the temperature the fluid was used at.
6. time in fluid.
7. washing and dehydration.
8. clearing methods used.
9. embedding methods and materials.
10. location of embedded material.

Under "Dehydration" in Fig. 1 the time the material remains in each grade of alcohol is recorded beneath that grade. In case the more recent practice is used (not yet published) of displacing the water with alcohol drop by drop an arrow is drawn, as indicated, to the percentage of alcohol the material is in at the end of the displacement. If the tissue was preserved in 70% alcohol then an arrow would be drawn to "70%" and later when the dehydration is continued a second arrow would be drawn to the grade at the

<u>LOT</u> 507 Testes of Cat - Half grown Solid tissue														
<u>Collected</u> U. of Penna. Castrated under ether	<u>Dehydration.</u>													
	<table border="1"> <tr> <td>Water</td> <td>35%</td> <td>50%</td> <td>70%</td> <td>80%</td> <td>95%</td> <td>100%</td> </tr> <tr> <td>running 20 hrs.</td> <td></td> <td></td> <td></td> <td></td> <td></td> <td>12 hrs.</td> </tr> </table>	Water	35%	50%	70%	80%	95%	100%	running 20 hrs.					
Water	35%	50%	70%	80%	95%	100%								
running 20 hrs.						12 hrs.								
<u>Killed</u> Nov. 1 - 1915 10 ⁰⁰ AM	<u>Drop Method</u> →													
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Flaming + urea used at 5° C.	<table border="1"> <tr> <td><u>Cleared.</u> Aniline Wintergreen</td> <td><u>Embedded.</u> 2 changes of para Rubber Paraffin.</td> </tr> <tr> <td></td> <td><u>Location</u> Box #1.</td> </tr> </table>	<u>Cleared.</u> Aniline Wintergreen	<u>Embedded.</u> 2 changes of para Rubber Paraffin.		<u>Location</u> Box #1.									
<u>Cleared.</u> Aniline Wintergreen	<u>Embedded.</u> 2 changes of para Rubber Paraffin.													
	<u>Location</u> Box #1.													
<u>Time in fluid</u> 24 hrs.														

Fig. 1

end of the series. The writer embeds his material immediately as in the long run it saves considerable time and tissue in paraffin is much easier to carry about the country than in bottles. The material under each lot number is usually embedded in a petri dish (the lot number on a small piece of paper is embedded with the material). The disk of paraffin after removal from the petri dish is wrapped in wax paper and filed in a 3x5 cardboard filing case behind an index

card bearing the lot number. In this way the material is compact and easy to get at.

On the second set of cards is to be found:

1. lot number.
2. slide number.
3. number of box in which slide is located.
4. stain used.
5. what is to be found on the slide.
6. thickness of sections.
7. condition of slide (i. e. good fixation or stain).
8. location of favorable areas on the slide.
9. notes concerning certain locations.
10. areas that have been drawn.
11. areas that have been photographed.
12. areas that have been drawn or photographed and used for publication.

The number is scratched on each slide of the series. If spermatogenesis is being worked upon one card is devoted to a single phase in the process found on a particular slide. Similar phases on other slides have their own cards. This card is labeled as shown in Fig. 2 under "Shows". The same slide may therefore have several cards devoted to it should it show more than a single phase. These cards are filed first behind an index card bearing the lot number and then in numerical order behind index cards bearing the phase name of the particular stage they happen to represent. When the observations do not deal with spermatogenesis then, of course, the cards are classified according to the special need.

As can be seen in Fig. 2 there is a place for forty readings. The right hand reading of the mechanical stage is placed above the short line, the horizontal reading is put beneath it. The slide is first searched with a low power lens and readings of apparently favorable locations are put down. Afterwards these locations are tested with the oil immersion lens and either crossed out or drawn. When the figure is drawn the location is circled as shown in Fig. 2. Any notes that are to be made are indicated by the figures in the space to the right of the readings. These numbers refer to corresponding numbers on the back of the cards under which the notes are written. Small sketches may also be put in these spaces

to recall what the reading is of. When the plates have been prepared for publication the figure number is entered in the square opposite the reading. When the cell has been photographed this information is also placed here with the number of the photo-


<u>Lot</u>	<u>Slide</u>	<u>Box</u>	<u>Stain</u>
507	6	30	Iron haematoxylin
<u>Shows</u>	<u>Thickness</u>	<u>Condition</u>	
Spermatozoa & <u>meta.</u>	8.		
<u>LOCATIONS.</u>			
78.2 48.7			
76.8 60.7	1.		
(51.3) 20.6	 Fig. 13		
70.8 50.8			
(127.4) 81.5	2. Photo 56.		

Fig. 2

graph. With these records should the plates be lost or when the original of a figure is to be examined the location on the slide may readily be found.

With the records on these cards before him the investigator has all his data well in hand for the preparation of his paper.

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ROBERT T. HANCE.

A MINIATURE DARK ROOM FOR USE WITH THE MICROSCOPE

All microscopists prefer to work either at night or in a darkened room. Using the microscope under such conditions does away with the strain to which both the observing and the unused eye are subjected by the side light—i. e., light coming from sources

other than through the tube. When working in darkened surroundings the effect is that of looking at a picture on a screen. The image appears brighter and objects become clear that under the usual conditions are scarcely visible.

For several years the writer has been trying to devise some method to control the light perfectly and to do this without necessitating the darkening the whole room. It is desirable that any apparatus for the purpose should weigh little and (for ease in carrying from one place to another), it should be simple to take apart. It should, of course, be adaptable to every condition. For further convenience of the worker definite places should be present in such an apparatus for the usual microscopical accessories—pens, pencils, drawing and memorandum cards and lens paper.

The following description is of a miniature dark room for use with the microscope fulfilling these requirements. It was designed and made by the writer last fall and, after a year's use, he has found it to be exceedingly practical in eliminating all the strain that results when the eye is unshielded. In this darkened enclosure the eye not in use is at perfect rest. Moreover for drawing the light may be controlled so that it is possible always to have light of the same intensity directed on the drawing paper.

Description

Figure 1.

A. Base— $\frac{1}{4}$ -inch white pine 12x18 inches with a binder of the same wood across each end to prevent warping.

B. Uprights—dowel sticks 1 inch in diameter cut to 18 inches in length.

C-C'. Rods—common telescoping curtain rods. Each of the rods C' is cut 8 inches from the end that ordinarily would be used to fasten it to the window. C is formed of the remainder, of the part between the ends.

D. Wire—a piece of annealed wire $\frac{1}{8}$ inch in diameter about 4 $\frac{1}{2}$ feet long bent as shown.

To assemble:—one two inch screw fastens each upright to the base. The upright on the right can be seen to have two angle irons aiding in its support but this is only necessary when the fan is

added. Holes are drilled in both uprights to correspond to the diameter of C which is inserted in them. The rods C' are attached by one end to the tops of the uprights by a screw through the eyelet in the rod. Through the eyelet at the opposite end a small rod is passed as shown to prevent the curtains from slipping off. The wire D is fastened to the outer sides of the uprights by means of a single round head screw passed through each flattened end. All the wood and metal work is painted a dead black.

For many valuable suggestions on the design of the curtains and for the excellence of their construction I am indebted to my mother. (See figure 2).

The curtains suspended from the rods C and C' are in four parts, all overlapping each other and fastening together with spring snaps. They are made of the heaviest grade of black sateen doubled. On the right hand curtain are pockets for pencils and cards. On the left side is a pocket for lens paper. The pocket is provided with a flap to exclude the dust. The upper curtain carried on the wire D is of single thickness. The central curtain is in two parts so that they may be separated to permit light to fall on the drawing board. The left hand curtain of the central set has a rectangle 1 inch wide by 5 inches high cut from the center of the basal portion. Across the top of this aperture is stitched a flap of double thickness, $3\frac{1}{2}$ inches wide by $5\frac{1}{2}$ inches in length. To one corner of the loose end of the flap is attached a tape which passes around the tube of the microscope and fastens to the other corner of the flap by means of a spring snap.

With the microscope surrounded by these curtains it is impossible to read the figures on the mechanical stage and so the small light (fig. 1 E) was installed. This can be adjusted by means of sliding rods locked with winged nuts to hang directly over the stage. The lamp arm is attached to the right hand upright by means of a collar made of two pieces of brass stripping fastened on either side of the pillar with a thumb screw. The lamp is a small tungsten bulb set in a porcelain socket. The shade or reflector, shown in the photograph, was taken from an old tubular flash light. A small three-cell pocket battery furnishes the current which is

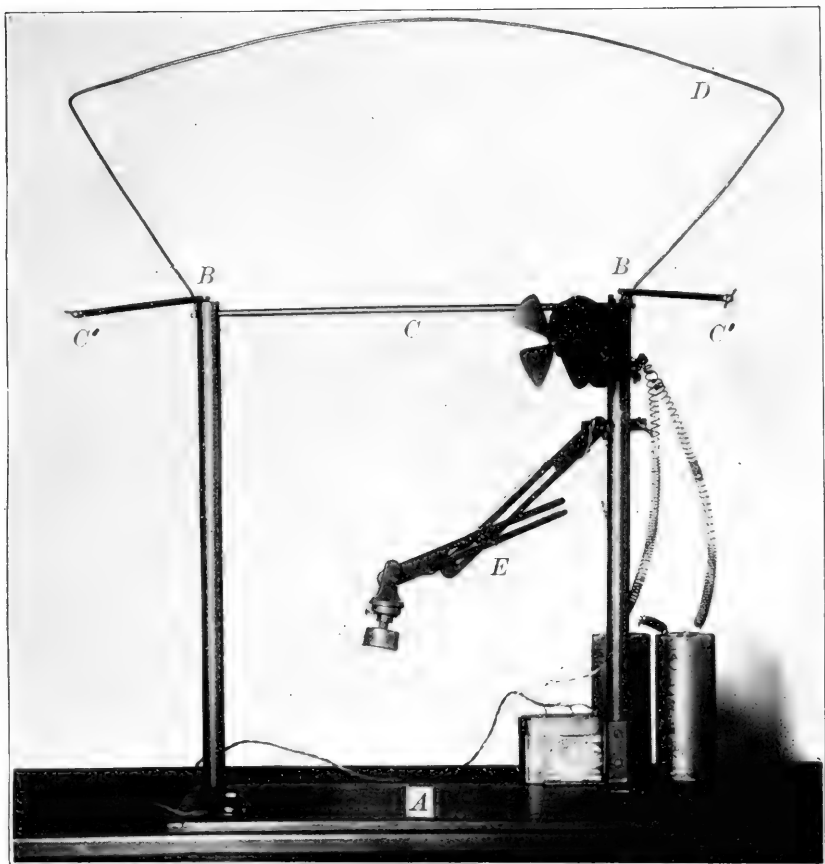


PLATE XII

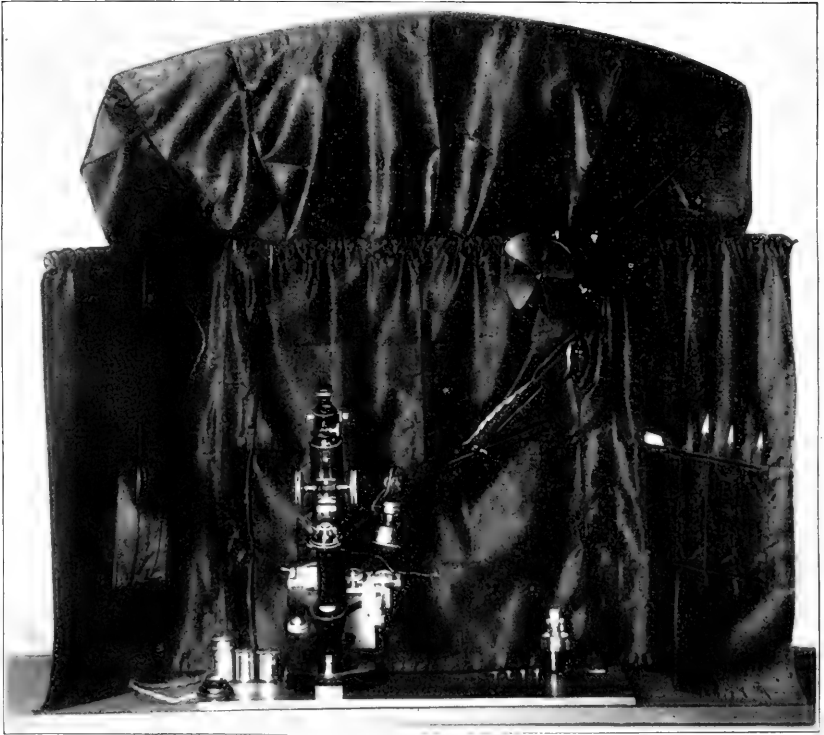


PLATE XIII

controlled by a push button at the left of the microscope. The same battery has lasted for very nearly a year now without visible signs of weakening.

The fan shown in both photographs is a toy motor equipped with a $4\frac{1}{2}$ inch blade. The motor is operated on two dry cells. It is fastened to a wooden base that is inserted in a slot in the upright and clamped tight by means of a winged nut. This fastening permits the fan to be tilted up and down while the single screw securing the fan to the base allows a left and right rotation. The air current may thus be directed on any spot desired.

Operation

For microscope illumination with this dark room a concentrated filament mazda frosted globe is used. This globe is placed behind the slit in the central curtain and the microscope is put in position on the opposite side. The flap covering the slit is then snapped about the tube of the microscope just above the nose piece. The slit through which the light comes is so narrow that the stage of the microscope effectively shields the eye from the light coming through the lower part of the slit while the flap takes care of all other dispersion.

In the average room having windows on only one wall the side curtains can be left wide apart. In places where the worker is almost surrounded by windows it is of advantage to draw the side curtains so close that there is just room for the observer's head to enter. The telescoping rods supporting the side curtains permit these to be narrowed or widened to suit the circumstances. The top curtain works to or from the microscopist and is frequently convenient in cutting out the light from the upper parts of the windows.

Light on the drawing paper is obtained by separating the lower portions of the central curtains from each other and fastening them back. The bulb illuminating the microscope then throws its light over the right hand side of the base. A constant intensity of illumination is in this way assured.

The fan is a luxury—possibly an unnecessary one, but in very warm weather or on days when a few flies persist in maintaining

their position at all hazards on top of the writer's head he has not been at all skeptical as to whether the luxury was unnecessary or not.

*Zoological Laboratory,
University of Pennsylvania.*

ROBERT T. HANCE.

NOTES ON A NEW SPECIES OF LOXODES (EHRBG.)?

In the course of work upon the distribution of fresh-water protozoa in the southeastern part of Massachusetts many species were found which could not be named according to the available classifications. This is true of various species of the genus *Loxodes*.

Loxodes belongs to the class of Infusoria and to the sub-class Ciliata; that is, the protozoon is provided with cilia or setæ during all of its stages, but is free of flagella. This sub-class is divided into a number of orders, *Loxodes* falling under the order of Holotricha. This order includes the ciliata which possess but one kind of cilia and show the anus and mouth conspicuously. The members of the genus *Loxodes* show a hook-like projection on the anterior end which is bent to the left, and cilia cover nearly the entire body. The body is flattened, slightly elongated and possesses a well defined outer envelope of the cell or ectoplasm which is constant in form. The dorsal surface is free from cilia, smoothed and curved. The ventral surface is flat and well ciliated, with a mouth on the left anterior edge which is at the bottom of a slit-like peristome. Some writers claim this leads into a pharynx, the existence of which I have not been able to see. The animal is a free swimmer and shows nuclei clearly.

The species under consideration has an average length of 60 microns and width of 16 microns. It is found in great numbers among *Oscillaria*, associated with *Nassula*, *Paramecium* and rotifers. There was no evidence of its feeding upon the algæ. Its food consisted principally of small paramecia.

They may be narcotized for study by a .47% solution of cocaine hydrochloride; though after about two minutes the animal slowly assumes an oval shape, then becomes round and all evidence of life ceases. The action of the narcotic was not so pronounced up-

on the paramecia and rotifers, for at the end of ten minutes locomotion was evident in these, although not so rapid as in the clear water.

During swimming these ciliata have a spiral motion which is to the right. Trichocysts were noted in the larger specimens, this being limited to the posterior region. Some of them seemed to possess contractile vacuoles while others did not. The anterior end is hook-like but not a rostrum. In this respect it differs from *Loxodes rostrum* (Mull.); the anterior end is also more blunt, and the dorsal surface was more curved than in *L. rostrum*.

Further study upon this species and the genus is in progress.
Chadwick, N. Y. ELTON R. DARLING.

ENTOMOLOGICAL NOTES

Variation in Spermatozoa.—Zeleny and Senay ('15, Journ. Exp. Zool., 19:505-514) report results of studies on variation in head length of spermatozoa in insects. This work is a continuation of earlier studies by Zeleny and Faust, an abstract of which appears in this journal (34:191). The following insects were studied: *Corizus lateralis*, *Leptocoris trivittatus*, *Reduviolus ferus*, *Euschistus variolarius*, *Cosmopepla carnifex*, *Passalus cornutus*, and *Berosus striatus*. With the exception of *Passalus cornutus*, all gave distinctly bimodal curves indicating the existence of two distinct size groups of spermatozoa. The one exception yielded a unimodal curve and is interpreted as indicating lack of distinction among the spermatozoa or else as having two groups which differ so slightly from each other that a unimodal result is produced. Additional support is thus given to the belief that dimorphism in size of spermatozoa is common among animals having two chromosomal classes of spermatids, due to quantitative differences in chromosomal content.

Bibliographies.—The Journal of Animal Behavior (5:407-461) has issued its annual lists of literature pertaining to the behavior of animals. Turner (pp. 415-445) has given a brief analysis of the more important results which have appeared in the literature of 1914 and lists one hundred seven papers from American and foreign sources which treat of the behavior of spiders and insects other

than ants. The analysis of subject matter is very useful and the list of titles, while not entirely complete, forms an important source of information.

Differential Incidence of Bruchus.—Harris ('15, Journ. N. Y. Ent. Soc., 23:242-253) reports the results of a statistical study on "differential incidence of the beetle *Bruchus*" in which the purpose was to discover whether the characters of the bean pod, on which the eggs of *Bruchus* are laid, determine, to any degree, the frequency of infestation ("parasitization"). Examination of over fourteen thousand pods, producing almost forty thousand seeds, showed that the relative number of infested seeds is greater in pods with larger numbers of ovules. Increase in percentage of infestation accompanies increase in the number of seeds matured per pod. No relationship between the position of the seed in the pod and liability of infestation was discovered. The hypothesis offered is that since young pod size is correlated with the number of ovules and the number of developing seeds, thus resulting, in the larger pods, increased ease of foothold and additional facility for oviposition are made possible.

Respiration in Zygopterous Larvæ.—Calvert ('15, Ent. News, 26:435-447) summarizes the literature on the subject of the respiration of zygopterous larvæ (Odonata). From compiled and original data it appears that the oxygen demands of the animals are satisfied, at least in part, by several modes of respiration. The general body-surface, the caudal processes, the rectal epithelium, certain spiracles, and, in some species, the lateral external tracheal gills, all form functional parts of the respiratory system.

Behavior of the Ant-lion.—Turner ('15, Biol. Bull., 29:277-307) has studied the behavior of the ant-lion and finds, among numerous other things, that the characteristic pits, which may be found in any kind of dry friable soil in protected situations, are constructed either by furrowing backward, producing a series of concentric excavations, each deeper than the preceding and removing the soil with the head, or by the simple removal of the soil until the sides of the pit become somewhat stable. Locomotion is invariably backward, never forward. "Any invertebrate, be it insect, arachnid, or crustacean, that happens to fall into the trap is accept-

able as food." Letisimulation (death-feigning) often follows rough handling or other similar treatment and, though variable in duration, mutilation may frequently fail to produce response. No well-defined relation appears to exist between the duration of the feint and temperature, fasting, or strength of the stimulus.

Reaction of Epeira.—Barrows ('15, Biol. Bull., 19:316-326) finds that the large orb weaving spider, *Epeira sclopetaria*, orients itself, when going from the center of the web to capture entangled flies, in response to a vibratory stimulus. The stimulus can be submitted experimentally and produces the normal response, the latter being analyzable into (1) the orientation, (2) the forward movement, and (3) the attack on the vibrating object. Mutilation experiments yielded evidence indicating the probability that the organs used in detecting the vibratory stimulus are sense hairs on the tarsi.

Regeneration.—Schmit-Jensen ('15, Smithsonian Report for 1914, pp. 523-536) made a study of homœotic regeneration of the antennæ in a phasmid, *Carausius morosus*. A "spontaneous case of substitutional homœosis" was noted among a lot of individuals (nymphs) which had suffered from cannibalism. An antenna, bitten off near the base, developed, after molting, a termination consisting of a distinct tarsus-like segment with a large empodium and two small claws. Subsequent molts witnessed the appearance of paired protuberances corresponding exactly to the paired plantulæ on the underside of normal tarsal regions. Amputation experiments on fifty specimens, involving removal of either or both antennæ at the level of the joint between the first and second, or the second and third antennal segments, resulted in the production of twenty cases of regeneration. These regenerations varied to some extent but all developed the tarsus-like terminations. Amputations made on young nymphs showed that regenerations developed more and more with each succeeding molt and the resemblance between the regenerated portion and the true tarsal region became greater. Under certain conditions of the experiments a tibia-like segment was also produced at the base of the tarsal region.

Orientation of Ephemera.—Krecker ('15, Biol. Bull., 29:381-388) finds that the positive reaction of May-flies to air currents

is evidently due to strains exerted on the muscles of those appendages which serve the function of attachment, rather than to sensations resulting from contact of moving air with the body. The normal resting position on vertical supports which is usually negative with respect to the earth's surface, is not, apparently, a pure reaction to gravity but is entirely or in part accounted for by the unstable character of the insect's attachment when in any other position. May-flies react negatively to bright sunlight but positively to certain artificial lights. An optimum zone surrounds colorless, sixteen candle-power, incandescent lights, beginning about six inches from each light and extending to about thirty inches, in which the positive reaction seems to be satisfied. Individuals alighting in this zone tend, if large numbers are present, to arrange themselves in rows corresponding to the radii. The area within six inches of the light is designated as the "excitement zone" since individuals entering this zone become excited and perform confused movements. The positive reaction is much stronger for slightly yellowish light than for red or blue. Red and blue lights produce the above-described alignment but a well-defined excitement zone is lacking.

Population of "Blanket-Algæ."—Platt ('15, Am. Nat., 49:752-762), in a study of the life of floating masses of filamentous algæ ("blanket-algæ") in fresh-water pools near Ithaca, N. Y., finds that insects are prominent components of such a community. The larvæ, nymphs, and adults of five orders of insects are regular inhabitants of the alga-masses and constitute the largest individuals of the population. Nymphs of Ephemera (*Callibaetes*, *Batis*, et al), nymphs of Odonata (*Enallagma*, *Ischnura*, et al), larvæ of Diptera (*Chironomus*, *Ceratopogon*, *Odontomyia*), larvæ of Coleoptera (*Hydrophilus*, et al), and adult beetles (*Helophorus*, *Crenophilus*, et al) were found to constitute the principal insect population, the Diptera representing the greater part. *Chironomus* was one of the two animal forms which appeared most regularly. Members of this genus were present at all seasons. The larvæ of May-flies and midges are the principal plant consumers and since the

latter are so prolific they are of great importance as food for other animals. Important data concerning the plant and other animal members of this population are given.

Lepidopterous Larvæ.—Fracker ('15, Illinois Biological Monographs, 2:1-169), in an extensive paper on the classification of lepidopterous larvæ, reports, among others, the following general results: The primary setæ, which constitute important taxonomic characters, have an arrangement in all segments of the body of the larva which has been derived from the same ancestral type, the latter including twelve primary setæ which are arbitrarily designated by Greek letters. Modification of this type has come about in three different and independent ways, namely, tendency of prothorax to retain the maximum number of setæ, partial reduction on the meso- and metathorax, and reduction of abdominal chætotaxy without much change of positions. Other general conclusions of a more special nature are also given. Extensive keys to families, genera and species are presented, accompanied by detailed discussion. This paper covers the whole order Lepidoptera and is the most comprehensive and thoroughgoing treatment of the subject which has yet appeared.

Poisons of Plant-Lice.—Dewitz ('15, Ann. Ent. Soc. Am., 8:343-346) reports that the diluted extract of certain plant-lice obtained by triturating these animals in physiological salt solution or a mixture of glycerine and physiological salt solution, when submitted to ox blood, hæmolyzes the red blood-corpuscles even at ordinary temperatures. "1 gr. of plant-louse matter will completely dissolve the red blood-corpuscles of 25 ccm. of undiluted blood or 40 gr. will dissolve a litre of blood." The substance causing this hæmolysis is given the name *aphidolysin*. Nothing is known concerning the particular part of the body of the insect which contains this poison.

Stimuli and Egg Hatching.—Severin, Severin, and Hartung ('15, Psyche, 22:132-137) have carried on experiments to determine the stimuli which cause the eggs of *Chætogædia monticola*, a leaf-ovipositing tachinid (Diptera), to hatch. These eggs are deposited on various grasses and weeds and, at the time of oviposition, contain fully developed larvæ. Since the larva is a parasite in certain

other insects which consume the vegetation on which these eggs occur, what accounts for the sudden hatching in the digestive tract of the host which permits the parasite to begin penetrating the wall before being expelled with the excreta? Eggs, placed in the alkaline liquids exuded from the mouths of certain host caterpillars, began hatching within less than one minute, almost all of them hatching within three hours. Similar results were obtained using alkaline liquids from insects which are not the normal hosts. Hatching occurred, in most cases, in the mid-intestine. Emergence was partially prevented, sometimes completely so, in various acid media. Experiments seemed to show that increased turgidity causes the larvæ to emerge when the eggs were immersed in water or diluted alkaline solutions for thirty-six hours or longer. Evidence was secured which points to the probability that the digestive juices of the host, reaching the larva through the micropyle, stimulate it to perform the body movements and contractions by which the escape from the chorion is affected.

Polyembryonic Development.—Patterson ('15, Biol. Bull., 29:333-372) finds that in *Copidosoma gelechiæ*, a hymenopterous parasite of the *Solidago* gall-moth (*Gnorimoschema gallæsolidagonis*), polyembryonic development occurs, the egg giving rise, on the average, to about 191 individuals. The cleavage stages were not secured, the earliest development studied being that in which the division of the egg into embryonic primordia had begun. The young polygerms have an outer nucleated membrane and a central region containing the true embryonic nuclei. The latter, by segregation into groups surrounded by a dense layer of granular protoplasm, develop into the primordia of the multiple embryos. The polygerm elongates, breaks up into several spherical primary masses, each of which contains several primitive embryos. Primary masses give rise to secondary masses and the embryos separate from each other, each developing a covering composed in part of granular protoplasm and in part of protoplasm from the nucleated membrane. The intervening space becomes filled with an inter-embryonal substance, the subsequent dissociation of which liberates the larvæ, thus introducing them into the body-cavity of the host where they ultimately destroy the unchitinized parts of the caterpillar.

The emergence of males and females from the same caterpillar is interpreted as the result of the deposition of two or more eggs in the same host. However, the possibility "that such broods may also arise from a single fertilized egg by a process of disjunction of the sex chromosomes during the early cleavage stages" is suggested.

Gynandromorph Bees.—Morgan ('16, Am. Nat., 50:37-45), in a paper on the Eugster gynandromorph bees, reviews the evidence pertaining to the two chief theories as to the origin of these anomalous insects. Recent studies by Boveri and Mehling on the original Eugster gynandromorphs show that the male parts of these bees are *maternal* while the female parts are *paternal*, thus supporting the original hypothesis of Boveri that these anomalous insects are produced by a delayed fertilization or by some irregularity in the penetration of the sperm into the egg, resulting in the fusion of the sperm nucleus with one of the two nuclei produced by the division of the egg, with the ultimate production of an individual which possesses the characters of the male on one side and those of the female on the other. Morgan's theory of gynandromorph origin is based on the possible entrance of two or more spermatozoa into the egg with a subsequent union of one with the egg nucleus and the independent development of an outlying spermatozoan, the former producing female structures and the latter producing male structures. According to the theory of Boveri, the male parts resulting from the single egg nucleus should be *maternal* while by Morgan's theory the male parts derived from the single sperm nucleus should be *paternal*. Attention is called to the discovery by Newell that the drone bees inherit the characters of the mother. It thus appears that Boveri's theory holds, at least for the case of the honey-bee. Boveri's cytological argument in support of his theory is reviewed but it is found by Morgan to be of uncertain support. Mention is made of a third possible origin for gynandromorphs, namely, "dislocation during ontogeny of the two sex chromosomes." Gynandromorphs would be expected to arise in insects when conditions would prevail in which certain nuclei come to contain two sex chromosomes and others only one. Attention is also called to Goldschmidt's explanation of the gynan-

dromorphs which he secures in crosses of *Lymantria dispar* and *L. japonica*, an explanation which "involves the relative potencies of the sex factors in the different races."

Marine Insects.—Arndt ('15, Proc. Indiana Acad. Sci., 1914, pp. 323-336) presents a paper on the habits of the insects and spiders of the "between tides zone" at Cold Spring Harbor, Long Island. Particular attention is given to *Megamelus marginatus*, *Grammonata trivittata*, *Clubonia* sp., *Bembidium constructum*, *Heterocerus undatus*, *Bdelidæ* sp. (?), and *Lycosa communis*. The first three are characteristic of the *Spartina* grass area, while the last four are common in the outer *Juncus* area. Almost all insects inhabiting the zone between high and low tide present peculiar protective features, some of which are as follows: (1) Certain unique instincts serve to prevent them from being washed away by the tide, as for example, the tenacious clinging to the blades of grass by those insects which inhabit the *Spartina* zone, the habit of crawling under the gravel, and the wandering about for food only during sunny days at low tide. (2) The instinct of certain forms to swim to the *Fucus* thallus during the disturbance of high tide, an instinct which is interpreted as one resulting in protection from aquatic enemies. (3) Marked resistivity of tide zone species to drowning. (4) Structural modifications which facilitate locomotion, enabling the possessors to inhabit an environment in which safety may be dependent upon retreat. Attention is called to the necessity of tide zone insects being air breathers since the terrestrial conditions prevail for one-half of the time. During periods of high tide or submergence they are inactive. "The most striking phenomena is the strictly zonal distribution of the insects of the between tides zone."

Salts Required by Insects.—Loeb ('15, Journ. Biol. Chem., 23:431-434) has raised five generations of the banana fly, each of normal motility, on the following nutritive mixture: grape sugar, 0.5 gm.; cane sugar, 0.5 gm.; ammonium tartrate, 0.1 gm.; citric acid, 0.05 gm.; K_2HPO_4 , 0.005 gm.; $MgSO_4$, 0.005 gm.; H_2O , 3 cc. No $NaCl$ or $CaCl_2$, other than that which may appear as impurities in the chemicals used, was necessary. K and PO_4 appeared to be indispensable and SO_4 and Mg must be present. These ex-

periments show that as highly organized an animal as this fly can be reared on a culture medium as simple as that required for certain micro-organisms. Attention is called to the general assumption that the evolution of the higher animals could have occurred only after the appearance of green plants since the latter serve directly or indirectly as food for the former. Although this is true in general of the present fauna there is the possibility that "an evolution of animals as highly specialized as insects might have taken place independently of the existence of green plants." Attention is also called to the results of certain other experiments which indicate that the nitrite and nitrate bacteria are capable of forming carbohydrates from carbon dioxide, or possibly other atmospheric carbon compounds, independently of light, and other micro-organisms might have the same power. Such micro-organisms might furnish the carbohydrates necessary for development of other micro-organisms requiring sugars for their growth. It thus seems obvious that the evolution of animals as complex as the banana fly might have been possible without the existence of chlorophyll.

Inheritance of Pink Coloration.—Hancock ('16, Ent. News, 27:70-82) has made a study of inheritance of the coloration in the unusual pink form of the katy-did, *Amblycorypha oblongifolia*. He has found that this pink katy-did crosses freely with the normal green form. When a pink female was crossed with a normal green male, some of the hybrid F_1 progeny emerged from the egg after two years while the rest of the eggs hatched after a period of three years. Nine of the hybrids had pink coloration like the mother, and four were green like the male parent. The sexes were about evenly divided in both the pink and the green forms. These hybrid F_1 types have been inbred but the F_2 generation has not yet emerged. The pink and green color which appeared after the first molt was not materially affected by subsequent ecdyses. The pink and the green colors are hereditary and are regarded as germinal in origin. The theory that the color is the result of absorption of coloring matter accompanying the food is rejected.

PAUL S. WELCH.

Kansas State Agricultural College.

PROCEEDINGS

of the American Microscopical Society

MINUTES OF THE COLUMBUS MEETING

The thirty-fifth annual meeting of the American Microscopical Society was held in connection with the A. A. A. S. at Columbus, Ohio, Dec. 29, 1915.

In the absence of President Kofoid, Professor Henry B. Ward acted as chairman. The meeting was called to order at 5 P. M.

The report of the Custodian was read and referred to an auditing committee, consisting of Dr. H. J. Van Cleave and Professor Frank Smith, both of Urbana, Ill.

On motion of Past-president Bleile the Secretary was instructed to send the greetings of the Society to Mr. Pflaum and to express to him our regret that he was unable to be present.

The Treasurer's report was read and accepted, being referred to the auditing committee named above.

The term of Treasurer Hankinson expired at this meeting, and the Society acquiesced with regret in his decision not to accept the office for another term. Unanimously the members present voiced their appreciation of his faithful and effective service of the Society during six years of its history, including the difficult period of reorganization.

The following officers were duly nominated and elected: President, Professor Michael F. Guyer, University of Wisconsin; First Vice-President, Professor T. L. Hankinson, Eastern Illinois Normal School; Second Vice-President, Professor L. E. Griffin, University of Pittsburg; Treasurer, Dr. Harley T. Van Cleave, University of Illinois. Dr. George R. LaRue, University of Michigan, Professor H. S. Brode, Whitman College, and Professor W. M. Rankin, Princeton University, were elected members of the Executive Committee.

The Secretary was selected as a member of the Council of the American Association for the Advancement of Science and was authorized to name as the second representative for 1916 some member actually in attendance at that meeting.

On the recommendation of the Executive Committee it was voted to allow the Secretary \$50.00, or such part thereof as may be necessary, in defraying the expense of attending the annual meetings of the Society.

The Chairman and Secretary were empowered to approve the minutes and prepare them for publication.

The Society adjourned.

T. W. GALLOWAY, Secretary.

SPENCER-TOLLES FUND

Custodian's Report for the year 1915

Reported at Philadelphia Meeting.....		\$4184.65
Dividends received	\$ 254.67	
Life membership No. 6, Seth Bunker Capp.....	50.00	304.67
		<hr/>
Total		\$4489.32
		<hr/>
Net increase during the year.....		\$ 304.67

GRAND TOTAL

All contributions to date.....	\$ 800.27	
All sales of proceedings.....	758.38	
All life memberships	300.00	
All interest and dividends	2820.67	\$4679.32
		<hr/>

LESS

All grants to date.....	\$ 150.00	
All dues on life memberships.....	40.00	190.00
		<hr/>
Balance invested.....		\$4489.32

Life Members: (Robert Brown, dec'd.); J. Stanford Brown; Seth Bunker Capp; Henry B. Duncanson; A. H. Elliott; John Hately.

Contributors of \$50 and over: John Aspinwall; Iron City Microscopical Society; Magnus Pflaum; Troy Scientific Society.

MAGNUS PFLAUM, Custodian.

Columbus, Ohio, Dec. 29, 1915.

We, the undersigned committee hereby certify that we have carefully examined the foregoing account of Magnus Pflaum, Custodian, and found the same correct.

H. J. VAN CLEAVE,

FRANK SMITH,

Auditing Committee.

ANNUAL REPORT OF THE TREASURER OF THE AMERICAN
MICROSCOPICAL SOCIETY

December 24, 1914, to December 24, 1915

RECEIPTS

Balance on hand from 1914.....	\$ 243.61
Dues of old members.....	442.87
Dues of new members.....	70.00
Initiation fees	102.00
Subscriptions for volume 33.....	100.00
Subscriptions for volume 34.....	34.40
Subscriptions for volume 35.....	26.00
Subscriptions for other volumes.....	103.00
Sales of Transactions.....	3.30
From Y. H. Tsou, as part expense of publishing his paper.....	75.00
From H. E. Metcalf, as part expense of publishing his paper.....	25.00
Life membership of S. B. Capp.....	50.00
Advertisers in volume 33.....	131.25
Advertisers in volume 34.....	18.50
Advertisers in volume 35.....	7.50
Advertisers in other volumes.....	90.00
Total	<u>\$1,522.43</u>

EXPENDITURES

Printing Transactions, volume 33, no. 4.....	\$ 198.29
Printing Transactions, volume 34, no. 1, 2, 3.....	483.65
Plates for Transactions, volume 33, no. 4.....	6.25
Plates for Transactions, volume 34, no. 1, 2, 3.....	37.70
Plates for volume 34 no. 4.....	24.22
Postage and express for Secretary	41.61
Postage and express for Treasurer	18.50
Office expenses, stationery, stenography, etc.....	126.40
Office expenses of the Treasurer.....	9.10
Advertising literature	39.88
Spencer-Tolles fund from S. B. Capp's dues.....	50.00
Sundry	10.18
Balance on hand	476.65
Total credits	<u>\$1,522.43</u>

T. L. HANKINSON, Treasurer.

Signed by Auditing Committee,

We hereby certify that we have examined this account, have checked items against vouchers on file, and have verified the totals. We find the report as given above to be correct.

H. J. VAN CLEAVE,

FRANK SMITH,

Auditing Committee.

TRANSACTIONS
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1916

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<i>Custodian:</i> MAGNUS PFLAUM.....	Meadville, Pa.

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GEORGE R. LARUE.....	Ann Arbor, Mich.
H. S. BRODE.....	Walla Walla, Wash.

EX-OFFICIO MEMBERS OF THE EXECUTIVE COMMITTEE

Past Presidents Still Retaining Membership in Society

R. H. WARD, M.D., F.R.M.S., of Troy, N. Y., at Indianapolis, Ind., 1878, and at Buffalo, N. Y., 1879	
ALBERT MCCALLA, Ph.D., of Chicago, Ill.	at Chicago, Ill., 1883
GEO. E. FELL, M.D., F.R.M.S., of Buffalo, N. Y.,	at Detroit, Mich., 1890
SIMON HENRY GAGE, B.S., of Ithaca, N. Y.,	at Ithaca, N. Y., 1895 and 1906
A. CLIFFORD MERCER, M.D., F.R.M.S., of Syracuse, N. Y.,	at Pittsburg, Pa., 1896
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E. A. BIRGE, LL.D., of Madison, Wis.	at Winona Lake, Ind., 1903
HENRY B. WARD, A.M., Ph.D., of Urbana, Ill.,	at Sandusky, Ohio, 1905
HERBERT OSBORN, M.S., of Columbus, Ohio,	at Minneapolis, Minn., 1910
A. E. HERTZLER, M.D., of Kansas City, Mo.,	at Washington, D. C., 1911
F. D. HEALD, Ph.D., of Philadelphia, Pa.,	at Cleveland, Ohio, 1912
CHARLES BROOKOVER, Ph. D., of Little Rock, Ark.,	at Philadelphia, Pa., 1914
CHARLES A. KOFOID, Ph.D., of Berkeley, Calif.,	at Columbus, Ohio, 1915

The Society does not hold itself responsible for the opinions expressed by members in its published *Transactions* unless endorsed by special vote.

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No 2

GRANTS FROM THE SPENCER-TOLLES FUND

By T. W. GALLOWAY, *Secretary*

The Spencer-Tolles fund is an endowment to encourage research. It has been accumulated by the American Microscopical Society thru a period of thirty-two years by means of Life Memberships, by gifts of individuals and societies, by sale of Transactions, and by accumulated interest. It is intended as a memorial to C. A. and H. R. Spencer and, R. B. Tolles, pioneer workers in America in optical instruments. It has been the desire to have this memorial a permanently active one,—continually stimulating scientific work and lending to scientific progress in the field so long served by the American Microscopical Society.

The funds are favorably invested and will soon amount to \$5000. Until that sum is reached it is the purpose of the committee to award less than the annual income each year. In the course of two or three years it is the hope that the annual grants may total \$250 or \$300.

Members of the American Microscopical Society are invited to aid the Society in reaching the ends for which the fund was established. In aiding the Committee find the very best avenues for the awarding of the grants, members will benefit themselves and the membership at large, since the results of the researches will be published in the *Transactions*. It is the confident expectation that the Spencer-Tolles Fund will not only advance research, but greatly strengthen the American Microscopical Society during the years to come.

The terms regulating the making of grants from the income of the fund, quoted from the announcement of the Committee, are given herewith.

Regulations Governing Grants from the Spencer-Tolles Fund.

1. The Committee will receive formal application for grants from the Spencer-Tolles Fund at present only from members of the American Microscopical Society.

2. For the present, under ordinary circumstances not more than \$100 will be voted in any one year to research purposes. No money can be granted for any other purpose from the income of this Fund.

3. Applications for grants shall be filed with the chairman of the committee.

4. On completion of the work each recipient of a grant shall give a report of the use to which the grant allowed has been put, preferably in the form of a paper ready for publication and embodying the results of the work in connection with which the grant was used.

5. Every grant is made upon the express condition that all results obtained by its aid shall be offered to the American Microscopical Society for publication in advance of their announcement elsewhere. Wherever published, the publications including the results of this work shall contain the distinct statement that the work contained in the paper was done with the aid of a grant from the Spencer-Tolles Fund of the American Microscopical Society.

6. The expenditure of the money shall be entirely in the control of the person receiving the grant and he shall not be asked to secure or furnish any vouchers covering the expenditure in detail. On completion of the work he shall file with his report a statement that such a sum, mentioning the amount, has been expended and the results of the work are contained in the accompanying report. Any unexpended balance retained by the custodian in making the final payment, or if paid out of the grant not covered by this statement, shall be returned to him and shall be again placed in the Fund.

(Signed) HENRY B. WARD, Chairman,
S. H. GAGE,
MAGNUS PFLAUM,
H. R. HOWLAND,
A. M. BLEILE.

The Committee or the Secretary will welcome correspondence from members regarding research work which for its preparation or publication needs the assistance of this fund. It is the purpose to interpret the conditions with the utmost generosity in the interest of scientific discovery. It is not the desire to hamper the investigator by limiting the use of grants to very precise purposes or control narrowly their expenditure. The fullest possible freedom for the exercise of individual judgment will be allowed the grantee.

SNOW-FIELD AND GLACIER OLIGOCHÆTA FROM MT. RAINIER, WASHINGTON*

By PAUL S. WELCH

The material which forms the basis of this paper was collected on the snow-fields and glaciers of Mt. Rainier, Washington, during February, March, April, and June, 1915. Specimens first came to the writer through the courtesy of Professor Frank Smith, of the University of Illinois, to whom they had been sent by the United States Bureau of Biological Survey. The collections were made by Mr. J. B. Flett of the Longmire Ranger Station who later supplied the writer with six collections from the same region. The writer wishes to express his indebtedness to Mr. Flett for his continued interest and his many courtesies in furnishing carefully preserved material and in supplying data on the habits and appearance of the living worms.

Genus Mesenchytræus

Both of the species discussed in this paper belong to the genus *Mesenchytræus* (*Enchytræidæ*), which, at the present time, includes almost sixty species and varieties, a few of which are of uncertain standing. Of this assemblage, twenty species and two varieties are recorded from North America. They are as follows, the type locality for each being given: *beringensis* Eisen (Bering Island, Bering Strait, Alaska), *beumeri* Mchlsn. (?) (Hamburg, Germany), *eastwoodi* Eisen (Hoods Peak, Sonoma Co., Calif.), *fontinalis* Eisen (Pine Ridge, Fresno Co., Calif.), *fontinalis* var. *gracilis* Eisen (Fresno Co., Calif.), *franciscanus* Eisen (San Francisco, Calif.), *fuscus* Eisen (Pit River, Calif.; No. Calif.), *fuscus* var. *inermis* Eisen (West Fork of Feather River and Goose Lake, Modoc Co., Calif.), *grandis* Eisen [Alaska (Sitka? or Juneau?)], *harrimani* Eisen (Kadiak, Orca, Metlakatla, Sitka, Yakutat, Unalaska, Alaska; Lowe Inlet, British Columbia), *kincaidi* Eisen (Ice-House Lake, St. Paul Island, Bering Sea, Alaska), *maculatus* Eisen (Popof

*Contribution from the Entomological Laboratory, Kansas State Agricultural College, No. 18.

Island, Alaska), *nanus* Eisen (Popof Island, Alaska), *niveus* Moore (Mt. St. Elias, Alaska), *obscurus* Eisen (St. Paul Island, Pribilof Group; Popof Island, Alaska), *orca* Eisen (Orca, Alaska), *pedatus* Eisen (Goose Lake, Alturas, Modoc Co., Calif.), *penicillus* Eisen (Port Clarence, Alaska), *setchelli* Eisen (Unalaska Island, Alaska), *solifugus* Emery (Muir Glacier; La Perouse Glacier, Alaska), *unalaskæ* Eisen (Unalaska, Alaska), and *vegæ* Eisen (Port Clarence, Alaska).

All but one of the above-mentioned species were originally described from North America and are not yet known to occur elsewhere. *Mes. beumeri* Mchlsn., a European species, has been doubtfully reported by Moore ('99, p. 141) from the vicinity of Philadelphia, Pa. He also reported an undescribed species of this genus from the same locality. The last American contribution to the knowledge of *Mesenchytræus* is that of Eisen ('05) in which most of the above-listed species are described. However, since that time new representatives of the genus have been reported from other parts of the world.

Mesenchytræus gelidus n. sp.*

(Plates XIV-XVI; Figs. 1-19)

Definition.—Length of alcoholic specimens, 21-32 mm., average about 24.7 mm. Diameter, 1.25 mm. Somites, 66-77. Color, dark reddish brown to almost black. Prostomium blunt, rounded, smooth. Head pore at tip of prostomium. Setæ sigmoid; all of same size and approximately uniform in length; in anterior part of body, 4-5 in lateral bundles, and 7-9 in ventral bundles; in posterior part of body, 3-4 in lateral bundles and 4-6 in ventral bundles. Clitellum on $\frac{3}{4}$ XI-XIII, continuous around body. Two septal glands on IV/V and V/VI. Brain with width two to two and one-third times the length; anterior margin deeply emarginate, posterior margin almost straight, lateral margins diverging cephalad. Dorsal blood-vessel arises in XIV; cardiac body present. Nephridia with small, slender anteseptal part and large, irregular, compressed postseptal part; efferent duct arises from ventral surface

*A typographical error which the writer was unable to correct appears in the abstract of this paper (Welch, '16, p. 143). The name of this species appears therein as *gelicus* instead of *gelidus*.

of latter about mid-way of its length. Spermiducal funnel large, somewhat cylindrical, bent; length three to four times the diameter; collar present, variable, usually reflected or flaring, set off from body of funnel by distinct, broad constriction. Sperm duct 6-7 times length of funnel. Penial bulb large, somewhat globular; atrium globular but smaller than body of bulb; about twelve, elongate, finger-like, multicellular glands opening into ental extremity of atrium about entrance of sperm duct; two sets of glands within penial bulb. Sperm sacs extend caudad to XXXI-XXXV. Ovisac single, tubular, bifurcates in XVI; extends to XXXI-XXXV; contains sperm sacs. One pair of unusually developed spermathecae; ectal opening laterad at IV/V, two inconspicuous groups of unicellular glands, surrounded externally by definite, light yellowish area; duct short, cylindrical, symmetrical, straight; two well-developed diverticula at ental end of duct, oppositely placed, elongate, slightly expanded at extremities; ampulla very long, usually terminating in IX-XI; devoid of connection with digestive tract; irregular, more or less unsymmetrical, constricted in regions corresponding to position of septa, caudal end usually dilated.

This description is based on twenty-seven sexually mature specimens. Many others of uncertain sexual maturity were examined in the study of external characters. The type and most of the paratypes are in the collection of the writer. Paratypes have also been deposited in the collection of the United States National Museum and in the collection of Professor Frank Smith.

The habitat of this species will be described in some detail in another part of the paper.

Affinities.—The determination of the affinities of this enchytræid is a matter of considerable difficulty, especially when the foreign species are considered. Many of the latter, recorded a number of years ago, are not described in sufficient detail to make possible a profitable attempt to discover relationships. Furthermore, the assemblage of species assigned to *Mesenchytræus* includes a number of poorly described ones, such as *Mes. armatus* Lev., *Mes. mencli* Vejd., and *Mes. montanus* Bret., the affinities of which cannot be judged either because of the failure to describe

a number of the essential details of structure or because of the use of sexually immature specimens. The validity of several foreign species seems to be questionable but, until they and other poorly described forms receive more intensive work, little can be done in the accurate determination of the synonymy. After a careful scrutiny of the literature dealing with the foreign species, the writer has not found any of them, as described, to closely approach *Mes. gelidus*.

Mes. gelidus belongs to the group of species having two diverticula on each spermatheca. This single point of agreement does not necessarily indicate close relationship and it is very possible that the convenient grouping of species on the basis of the number of diverticula is somewhat artificial. However, *Mes. gelidus* belongs to the group in which the spermathecae are prolonged caudad through several somites and lack connection with the lumen of the digestive tract. It appears that at least five American species, namely, *Mes. harrimani* Eisen, *Mes. setchelli* Eisen, *Mes. franciscanus* Eisen, *Mes. obscurus* Eisen, and *Mes. maculatus* Eisen, are to be regarded as close relatives. They have been described in considerable detail and fairly satisfactory comparisons are possible.

Mes. harrimani differs from *Mes. gelidus* in having a length of more than twice the average of the latter; a larger number of somites; a smaller number of setae in both lateral and ventral sets; brain square, not markedly wider than long; a longer and more slender spermiducal funnel; a much shorter sperm duct; and less compact nephridia, each possessing a distinct bladder-like chamber near the ectal opening of the efferent duct. Slight differences occur in the position of the clitellum and in the distribution of pigment. The penial bulb and associated structures are similar in some respects but a satisfactory comparison is prevented owing to a discrepancy in Eisen's description ('05, pp. 24-25) in which the following statement is made: "Atrium medium size, with about sixteen large gland-fascicles opening at the entrance of the atrium into the bulb." In the "Synopsis of species of Mesenchytræus" in the same paper (pp. 18-20) the following statement is made: "Penial glands, about 12 long atrial glands". Eisen's text figure No. 6 shows nine atrial

glands and another figure (Plate II, Fig. 4) shows fourteen. Both figures are, however, diagrammatic. No discussion of such variation occurs in the description and the writer is at a loss to know what interpretation to put upon the matter. Disregarding the atrial glands, the structure of the penial bulb, particularly the internal glands, differs from that of *gelidus*.

Mes. setchelli differs from *Mes. gelidus* in possessing a rounded brain with a decidedly convex posterior margin; in having the origin of the dorsal blood-vessel in XVIII; in having sperm sacs which reach only to XVIII; and in possessing only five atrial glands in connection with the penial bulb. Smaller differences exist in the character of the spermiducal funnel, nephridia, and in the details of structure of the penial bulb.

Mes. franciscanus differs from *Mes. gelidus* in the distinctly smaller number of setæ per bundle in both sets; in the origin of the dorsal blood-vessel in XVI; in the much longer and more slender spermathecal diverticula; and in the spermiducal apparatus which has a single, large, well-defined accessory gland in connection with the penial bulb, a sperm duct not longer than one and one-half times the length of the funnel, and distinctly sessile, globular, atrial glands. Minor differences exist in the position of the clitellum and in the finer structure of the penial bulb.

Mes. obscurus is distinguished from *Mes. gelidus* by the position of the clitellum on XII and XIII; the very long, slender, spermathecal diverticula; the very slender spermiducal funnel; the loosely constructed, three-lobed nephridia; and the possession of 16-20 atrial glands in connection with the penial bulb.

Mes. maculatus seems to be a close relative of *Mes. gelidus* but differs from it in the number of setæ per bundle in the lateral set; in the deltoid shape of the brain; in the spermathecae in which the slender diverticula are not located at the junction of the duct with the ampulla and are much shorter than the former; and in the possession of three sets of internal penial bulb glands, one set consisting of multicellular glands.

External Characters

The body of *Mes. gelidus* is elongate, sub-cylindrical, smooth, and of about uniform diameter, except in the regions cephalad of

the clitellum and near the posterior end where there is a gradual tapering towards the extremities. In alcoholic specimens, there is a slight but distinct dorso-ventral flattening accompanied by a faint, shallow, mid-ventral, longitudinal depression which is present throughout the greater part of the body. However, since the writer has not had the opportunity of studying living material, no statement can be made concerning the constancy of these characters and there is the possibility that they are unnatural results incident to preservation and that the true form is a cylindrical one as is the case in many of the *Enchytræidæ*. The length, in preserved material, varies from 21 to 32 mm., the average of twenty-five sexually mature specimens being 24.7 mm. The maximum diameter, in the region of the clitellum, is about 1.25 mm. The segmentation is distinct in all parts of the body. The intersegmental grooves are narrow, shallow, and regular in outline, except in the vicinity of the extremities where they are broader, deeper, and more conspicuous, particularly on the cephalic end. The deepening of the intersegmental grooves in the regions of the extremities produces an antero-posterior convexity of the surface of intervening somites which elsewhere is uniformly plane. On the surface of each somite, midway between the margins, is a faint elevation or ridge which encircles the body, including the four setæ bundles. A number of the specimens show, in the middle region, a series of ruptures in the intersegmental grooves, thus producing whitish rings which consist of interruptions, more or less regular, of the cuticula and hypodermis, exposing the underlying muscle layers of the body-wall. Further discussion of these ruptures occurs in another part of the paper.

The number of somites varies from 66 to 77, the average of twenty-five specimens being 69. A few mature specimens were studied in which the number was as low as 50 but these were disregarded since there was some evidence of loss of somites and subsequent regeneration in the posterior region. The color of most mature specimens is, in general, deep reddish-brown to almost jet-black. The distribution of color is not uniform. The ventral surface is often of a slightly lighter hue and the anterior and posterior regions are invariably lighter in color. In occasional specimens,

the terminal somites are light yellow. Sometimes the black color predominates over almost the entire body. A few specimens were found in which a distinct, rather abrupt transition from also black to yellow occurred in the vicinity of LX but they are referred to above as displaying some evidence of regeneration, a possible explanation for the difference in color. The maximum intensity of the black color occurs dorsad and laterad, in the region behind the clitellum. An examination of the surface under magnification shows the universal presence over the body, save on the clitellum, of innumerable, minute, irregularly distributed, light yellow spots, which give the surface a flecked appearance. These microscopic areas are slit-like, the long axes predominantly extending in the direction of the circumference.

Certain, special, superficial areas show distinct color differences. The ectal opening of each spermatheca at IV/V is surrounded by a broadly fusiform area which occupies about one-half the width of the two adjacent somites and is rendered rather conspicuous by the contrast with the surrounding surface. The crescent-shaped mouth and the transverse slit-like openings of the penial bulbs and oviducts are surrounded by narrow but distinct, light yellow areas. On the ventral surface, beginning with VII, a pair of small, circular, widely separated, yellow spots occur on either side of the mid-ventral line and in close proximity to the cephalic margin of each somite. These are the ectal openings of the nephridia.

The clitellum occurs on $\frac{3}{4}$ XI-XIII. Some slight variation in extent was noted but on completely mature specimens the above-mentioned limits hold. It is moderately thick, increasing, to a limited extent, the diameter of the body in that region. The distinct, uniform, yellow color renders it a conspicuous external character. It completely surrounds the body, no diminuation of thickness occurring on the ventral side. The surface is smooth and lacks the flecked appearance described for other regions of the body.

The head pore is distinct externally and located on the apex of the prostomium. The ectal opening has the form of a trans-

verse slit and is surrounded by a narrow, yellowish area. Dorsal pores are absent.

The setæ are distinctly sigmoid and arranged in fan-shaped bundles which are disposed in four longitudinal rows, two ventral and two lateral. The setæ of a bundle are all of approximately equal development. In the anterior part of the body, there are 4-5 setæ in the lateral bundles and 7-9 setæ in the ventral bundles. In the posterior part, there are 3-4 setæ in the lateral bundles and 4-6 in the ventral bundles. The setæ on XI are not specialized.

Internal Characters

Lymphocytes.—In the alcoholic specimens examined, the lymphocytes (Pl. XIV, Fig. 4) are scanty in the anterior region of the body but posterior to the clitellum they occur in some abundance. They vary in shape to some extent but, in general, are oval or elliptical. All lymphocytes are so heavily loaded with dark, non-staining pigment-granules that the cytoplasm is almost entirely obscured and frequently the nuclei are almost completely hidden.

Chloragoc Cells.—Aside from the anterior five or six somites, the digestive tract is covered with chloragoc cells (Pl. XIV, Fig. 5) for the greater part of its length. They are closely set together, usually elongated, and expanded at the free ends. In most of the specimens examined, these cells were heavily loaded with numerous, minute pigment-granules. In many instances, the amount of pigment present was almost sufficient to render the ectal portion of the cell unstainable.

Brain.—The brain (Pl. XV, Fig. 10) lies almost entirely in I, although in some of the specimens it extends slightly into II. It is easily dissected out *in toto*, thus facilitating the study of the organ as a whole. In all of the preparations, the shape and proportions are quite constant. The width is approximately from two to two and one-third times the length, an average measurement being: length, 0.132 mm.; width, 0.301 mm.; maximum thickness, about 0.011 mm. The anterior margin is deeply emarginate and slightly angular in character while the posterior margin is very slightly concave. From the rounded latero-caudal angles, the lateral margins diverge cephalad. Two pairs of supporting strands extend to the

body-wall, one arising from the anterior part near the emargination and the other from the posterior margin.

Nephridia.—The nephridia (Pl. XV, Figs. 14, 18) begin on VI/VII. Their shape varies somewhat in different regions of the body and in different specimens but, in general, the anteseptal part is composed of a nephrostome borne on an elongated, narrow pedicel. The postseptal part is an enlarged, irregular, compressed mass, the posterior end of which is reflected cephalad and gives rise to the efferent duct. In the large majority of the specimens examined, the nephridia are distinctly compressed and lie flat against the ental surface of the body-wall. Structurally, the body of the nephridium is of the usual mesenchytræid type. No evidence of a reservoir at the ectal end of the efferent duct was observed.

Spermiducal Funnel.—This organ (Pl. XV, Figs. 15-17) lies in the usual position in XI. The dimensions show limited variation but the length is commonly from three to four times the maximum diameter. A well-developed, variable collar is present. In all of the specimens examined, the funnels are bent in varying degrees, and the opening is directed caudad. The sperm duct is approximately 6-7 times longer than the funnel and extends caudad, usually to XIII/XIV. It is then reflected cephalad, extending into XII to unite with the penial bulb.

Sperm Sacs and Ovisac.—A large part of the cœlom posterior to the clitellum is occupied by the extensive sperm sacs (Pl. XIV, Figs. 1-2) and the single ovisac. They lie ventrad and laterad of the alimentary canal and are the most conspicuous of the internal organs. They are formed by very long caudal extensions of certain septa in the clitellar region. In the specimens examined, the delicate nature of these septa and the crowded condition of the internal organs in the clitellar region make the exact determination of the origin of the sperm sacs and the ovisac difficult and it is only by careful reconstructions of serial sections that the beginnings of these storage sacs can be followed. Septum XII/XIII continues caudad as a single, tubular outgrowth to XVI where it divides into similar halves, a right and a left, which lie on either side of the median line. The posterior termination, in the specimens exam-

ined, varies from XXXI to XXXV inclusive. Definite constrictions occur at intersegmental regions while between the latter distinct swellings are present. In sexually mature specimens, developing masses of ova occur in great quantities throughout the entire length of the sac.

Septum XI/XII continues caudad as two tubular outgrowths, one corresponding to each spermiducal funnel, which enter the ovisac and are completely contained within the latter and its branches. In XVI, where the ovisac divides, each sperm sac enters the corresponding branch of the ovisac and is coterminal with it, the variation in the position of the end being XXXI-XXXV. The sperm sacs also show constrictions and swellings which correspond to the segmentation. They fill approximately one-half of the lumen of the ovisac and, in sexually mature specimens, are crowded full of developing spermatozoa. The opening of each spermiducal funnel is directed towards, and is in close proximity to, the anterior opening of its corresponding sperm sac, in some specimens being partly contained within it. The sperm duct apparently lies entirely outside of these sacs.

A somewhat similar extensive provision for the storage of the developing reproductive cells has been reported previously in a few other species. Eisen ('05, pp. 25, 47, 49) found that in *Mes. harrimani* the sperm sacs extend "back some thirty somites" but made no mention of an ovisac; that in *Mes. fuscus* the sperm sacs are very large, "extending as far back as somite XXVII or further"; and that in *Mes. fuscus* var. *inermis* the sperm sacs extend to XXII and the ovisac to XXVIII. Other species are described in which sperm sacs and ovisacs are recorded as "extending far back" or by some other similar, indefinite statement. Many descriptions contain no mention of sperm sacs or ovisacs. Until such described species have been re-examined, it will not be possible to know whether or not these extensive sacs are peculiar to the American fauna.

Penial Bulb.—In general structure, the penial bulbs (Pl. XVI, Fig. 19) conform to the mesenchytræid type of Eisen. They present a noteworthy complexity of structure and agree in a number of respects with the same organs in certain Pacific Coast species.

They are attached to the ventral, ental surfaces of the body-wall in XII, one on either side of the median line. Each organ occupies a large part of the cœlom, having a total length, exclusive of the atrial glands, of about two-thirds the diameter of the body in that region. The maximum diameter slightly exceeds one-fourth of the transverse body-dimension. In the specimens studied, it was easy to dissect out these organs and to study them *in toto* as well as in serial sections.

Each organ is composed of two distinctly differentiated regions: (1) the globular atrium, and (2) the slightly elliptical body of the bulb. Superficially, the sperm duct unites with the ental apex of the atrium and is uniform in diameter from that point to the spermiducal funnel. About twelve, well-developed, finger-like, atrial glands are attached to the apex of the atrium near the union with the sperm duct. All are similar in shape and dimensions and extend freely into the cœlom. The attachments of these glands are not distributed uniformly about the apex of the atrium but are aggregated largely on one side.

The internal structure of the penial bulb is complicated and presents some interesting detail. In the completely retracted condition, the extension of the sperm duct forms at least one-half of the length of the bulb as a whole. The remainder of the organ is that part surrounding the penial bulb invagination. The continuation of the lumen of the sperm duct widens within the atrium to form a definite, spacious chamber, corresponding in contour to the external surface of the globular atrium. This chamber, at its ectal side, leads into an elongated lumen which expands slightly, forming another chamber, just entad of the penial pore. This expansion seems to correspond to the "penial chamber" described by Eisen ('05, p. 8). The penial bulb invagination is deep, bounded on the mesal side by a rather smooth wall, but on the opposite side two strong folds are present. It is lined throughout by a continuation of the external cuticula. The lining epithelium of the atrium differs from the corresponding tissue in the sperm duct in that the cells are elongated, reduced in transverse dimension, and lack cilia. The muscle-layer is in close proximity to the bases of these cells and while it varies somewhat in thickness and is interrupted at

intervals, it can be traced to its origin from the circular muscle-layer of the body-wall. The epithelium, at the penial pore, graduates into the hypodermis which forms the greater part of the lining of the penial bulb invagination. Structurally, it differs from the peripheral hypodermis only in the reduced length of the cells and the absence of the heavy, ectal zone of pigment, except at the entrance of the invagination and on the extremity of the first fold. The lateral portion of the bulb contains a large number of loosely associated muscle-strands which extend, in general, from the periphery towards the interior. The retractor muscle is formed largely by the union of strands from the circular muscle-layer of the body-wall with two other bands, each extending into one of the large folds which project into the invagination. The mesal side of the bulb, particularly near its base, also contains a loose meshwork of muscle-strands. In the ental half of the organ, a circular muscle-layer is present, exterior to and in contact with the longitudinal muscle-layer. The former is particularly well developed in the lateral half of the bulb. It seems probable that this circular layer is a continuation of the longitudinal muscle-layer of the body-wall. A very delicate peritoneum separates the muscle-layers from the coelom.

Sections show that the atrial glands are composed of two very definite regions, a peripheral region and a central one. The peripheral region is constructed of large, somewhat cuboidal, distinctly nucleated, gland cells arranged in a single layer. The central region is composed of the extensions of the peripheral gland cells which evidently function as ducts. The gland cells take artificial stains intensively but the central region seems to have little or no affinity for them. These extensions of the gland cells composing the central region penetrate the wall of the atrium; extend beneath the muscle-layers, separating the latter from the epidermal lining of the atrial and "penial" chambers; and open into the penial bulb invagination on the surface adjacent to the penial pore.

Two sets of glands occur within the penial bulb. Many large unicellular glands of varying size and shape intermingle with the strands of the circular muscle-layer in the ental half of the organ. It has not been possible to follow out the extensions of these cells

and determine with what part of the lumen they are related. Another set of glands is present in the lateral side of the ectal half of the organ and are related to the penial bulb invagination. Small unicellular, fusiform, gland cells occur in the heavy folds of the wall, in close proximity to the hypodermis. Each gland has a very fine extension which is related to the hypodermis but the exact nature of this relation has not been determined. It seems probable that these prolongations extend to the surface of the cuticula.

Spermathecæ.—Among the prominent internal organs of the body is a pair of spermathecæ (Pl. XV, Figs. 7-8, 12-13) which first appears in V. These organs are greatly elongated, often extending caudad as far as XI and occupying the large part of the cœlom in that region. Each organ is composed of three distinctly differentiated parts, namely, duct, diverticula, and ampulla. The duct is straight, elongate, cylindrical, and slightly greater in diameter near its middle. The ectal opening is laterad in position in IV/V, slit-like, and surrounded on the external surface of the body by the distinctly light colored area already described. Two groups of unicellular glands are associated with the ectal opening, one on the cephalic and the other on the caudal side. The component glands of each group are elongate, club-shaped, and distinctly nucleated. These groups of glands are apparent only in sections and because of their small size may be overlooked in a casual examination. At the junction of the duct with the ampulla are two, smooth, club-shaped, oppositely situated diverticula, each of which is slightly longer than the spermathecal duct and is reflected caudad, parallel with the long axis of the ampulla. The bulk of the spermatheca is composed of the greatly elongated ampulla. It is characterized by a series of swellings and constrictions, the latter corresponding to the intersegmental grooves, and the whole ampulla often has a moniliform appearance. There is no connection of any kind with the digestive tract but the caudal, free extremity of the organ is an expanded sac, variable in form, and usually larger than any of the other distended portions of the ampulla.

The spermatheca shows some interesting variations. The duct and diverticula are comparatively constant in shape and size but the ampulla is variable. Commonly, the ampullæ in the same speci-

men are of equal development and approximately of the same shape but a number of specimens were examined in which one ampulla (Pl. XV, Figs. 12-13) was greatly elongated, extending into IX, while the corresponding one on the opposite side of the animal was reduced, extending through only one or two somites, and considerably different in shape. The usual position of the ampulla is parallel to and latero-ventral of the digestive tract but specimens were examined in which the ampullæ were somewhat contorted and partially wrapped around the digestive tract. It is possible that such a condition is responsible for the reduced size of one ampulla since in certain specimens the ampulla on one side had apparently exceeded the other in growth, had filled the available space on its side of the cœlom, and had become crowded to the opposite side while the ampulla of the opposite spermatheca had grown caudad until it came into contact with the other one and had evidently ceased development, resulting in its reduction in length. An examination of these reduced ampullæ *in situ* sometimes showed the free extremities in contact with the opposite ampullæ and doubled upon themselves, suggesting a crowding of parts in development.

The excellent state of preservation of the specimens made possible a study of the finer structure of the spermatheca. The external cuticula is reflected into the ectal opening and forms a complete lining for the lumen of the spermathecal duct, disappearing at the bases of the diverticula. The hypodermis merges into the lining epithelium of the duct which composes by far the greater part of the thickness of the wall. The cells are elongated, glandular in appearance, closely set together, and distinctly nucleated at their bases. They are longer in the middle region of the duct and are responsible for that position of the maximum diameter of the organ. At the ental end of the duct, these cells gradate rather abruptly into the lining epithelium of the diverticula. The circular muscle-layer of the body-wall forms the longitudinal muscle-layer of the duct for its entire length, diminishing in thickness at the bases of the diverticula. It has not been possible to determine whether the longitudinal muscle-layer of the body-wall is related to the spermatheca. Unicellular glands occur over the outer surface of the duct throughout its entire length.

The walls of the diverticula are composed of a lining of epithelium, constructed of cuboidal, closely set, glandular, distinctly nucleated cells, bounded on their ectal ends by a very thin muscle-layer which, in turn, is covered by a continuation of the above-mentioned peritoneum.

Structurally, the ampulla is composed of two regions, a short ectal portion adjoining the duct, and a very long ental part extending caudad. The lining epithelium of the ectal region shows numerous transverse folds, thus increasing its surface to a considerable extent. This epithelium usually has irregularly shaped, small, non-staining pigment-granules in the free ends of the cells. The muscle-layer is well developed in this part of the diverticula. Unicellular glands occur exterior to the muscle-layer and are bounded at their outer ends by the peritoneum. In the remainder of the ampulla, the wall is greatly reduced in thickness so that the layers are difficult to distinguish. A thin lining epithelium and a delicate peritoneum are present and there are some hints of the presence of a muscle-layer, the exact character of which could not be determined. Small, scattering, irregular thickenings occur on the inner surface of this region.

The contents of the spermathecae present some interesting features. In all of the sexually mature specimens examined, the spermathecae contain spermatozoa which have a definite distribution in the organ. No spermatozoa were found in the duct but the diverticula almost invariably contained them, each mass having a definite and constant relation to the epithelial lining. The heads are all in contact with the lining of the diverticulum and the tails extend out into the lumen. The meaning of this arrangement is not clear. Spermatozoa are almost constantly absent from the ectal portions of the ampulla but, in the long, expanded part, surprising quantities, crowded into large masses, are present in the lumen and are not related to the wall in any way.

The striking structural feature of the spermathecae is the unusual size. It appears from a study of the literature on *Mesenchytraeus* that greatly enlarged and elongated spermathecae, such as the type just described, have been found only in American species from the Pacific Coast. Eisen ('05) described such organs for the

first time in the following species: *Mes. harrimani*, *Mes. setchelli*, *Mes. franciscanus*, *Mes. obscurus*, *Mes. maculatus*, *Mes. vegæ*, and *Mes. orca*. As descriptions stand at present, it appears that in this large genus only a small group of species possesses this particular type of spermatheca. Eisen ('05, p. 15) makes the following statement in this connection: "There is some little reason to suspect that this enlargement of the spermathecæ in this genus may have been overlooked in some species, and that some spermathecæ which have been described as short and as immediately connecting with the intestine, in reality are greatly prolonged posteriorly. The part adjoining the diverticles is always narrow and closely approaches the intestine. This peculiarity causes it to tear readily and I am satisfied that such torn spermathecæ have been considered as entire." Whether Eisen's speculations are true remains to be proved but since some of the descriptions of foreign species are very meager, unsatisfactory, and evidently made without careful dissection of specimens or the study of serial sections, it is possible that this type of spermatheca is not quite as unique as it now seems. However, since the spermathecæ have long been used as a taxonomic character and therefore called for special attention, it would appear that the number of cases in which such exceptionally large organs would be overlooked must, at best, be quite small. An inspection of the above list of species possessing such spermathecæ shows that these forms are all Alaskan in distribution, save *Mes. franciscanus*. It might be suspected that greatly enlarged and prolonged spermathecæ are characteristic of species inhabiting cold regions but *Mes. falciformis* Eisen, *Mes. fenestratus* (Eisen), *Mes. primævus* Eisen, and others are found in the arctic regions of the Old World and yet do not possess this peculiarity. Likewise, *Mes. solifugus* Emery and *Mes. niveus* Moore, found in frigid conditions in Alaska, have spermathecæ of the ordinary enchytræid type.

Pigmentation.—Examination of sections of the various regions of the body shows that the color described in foregoing pages has its basis in dark, brownish, non-staining pigment-granules which occur in marked abundance in the body-wall and certain internal organs. The hypodermis bears the principal load, the pigment being distributed through this whole layer. It occurs chiefly in the

outer ends of the hypodermal cells but may be scattered all through them. Pigment-granules are present in varying amounts in the following internal organs: (1) lymphocytes; (2) chloragoc cells; (3) epithelial lining of the ectal, folded end of the spermathecal ampulla; (4) ectal portion of the hypodermal lining of the penial invagination; (5) setigerous glands; (6) lining of the buccal cavity and the pharynx; and (7) ectal ends of the efferent nephridial ducts. Of the above-mentioned internal structures, the lymphocytes and the chloragoc cells contain the largest amounts of pigment. Granules do not seem to occur in connection with the nervous systems as is the case in *Mes. solifugus*.

BIOLOGICAL NOTES

The only recorded observation of "snow worms" on Mt. Rainier which the writer has been able to find is given by Moore ('99, p. 142): "In a letter Prof. Russell adds the interesting information that he has observed similar worms on the snows of Mt. Rainier, Wash., thus indicating for them a wide distribution."

All of the information concerning the living form has been supplied by Mr. J. B. Flett, who, as stated before, furnished the writer with the material on which this paper is based. Sexually mature specimens were collected from February 23 to April 5. Whether this represents the seasonal period of sexual maturity is not known. Between the above-mentioned dates, these worms occurred abundantly on the snow-fields of Mt. Rainier, at an elevation of from 2700 to 5600 feet. They also occurred on the snow on the mountain slope in a dense forest of fir and hemlock. These worms have not thus far been found on ice nor on the glaciers though they occur on the snow below the ice front and outside of the lateral moraines of Nisqually Glacier. The snow on which they were found is not permanent through the entire season but melts with the coming of summer and it therefore appears that a part of their life history must be spent on or in the ground. During midwinter when the temperature is very low, they are inactive and do not appear on the surface of the snow. Appearance at the surface accompanies the rising temperature in the spring and their activity becomes noticeable when the snow is beginning

to melt. When placed on hard packed snow during their active period, they are able to bore down through it at will. Under conditions of softening snow, they exhibit a rather efficient locomotion. When taken in the hand, they perform lively squirming movements for a time but soon relax and become quiet. Blue jays and several other species of birds prey upon these worms, picking them off the surface of the snow.

There is the possibility that these enchytræids undergo a color change during the first few days of their appearance. The first specimens, which were collected January 7, are very light yellowish in color and show no evidence, externally or internally, of pigment. Since these specimens are sexually immature, specific identification is not possible but they seem to be the species under discussion. As already stated, the sexually mature form is very dark in color and bears a conspicuous amount of pigment. Mr. Flett is of the opinion that, since the light colored specimens only appear very early in the season and since dark specimens of similar size appear later in the same localities and in approximately the same numbers, this difference in color is probably not a species difference but rather a life history difference in the development of the same species. None of the light colored specimens examined by the writer have been sexually mature but all of the dark colored individuals have either been completely mature or very close to complete sexual maturity. The evidence thus far is circumstantial only and does not justify any definite conclusions. It may be mentioned in this connection that Moore ('99, p. 135), quoting Mr. Bryant, reports what seems to be a similar color change in *Mes. solifugus*.

Nothing definite is known concerning the food of these snow-worms. Mr. Flett reports that the snow over which these enchytræids crawl has a red color, due to a minute, unicellular plant, which, in his opinion, serves as food for the worms. The writer made some examinations of the material contained in the alimentary canal of these worms, and found what seems to be microscopic algæ composed of very minute, globose cells, containing greenish and reddish colors, occurring singly or in clusters, and having the appearance of *Pleurococcus*. This material occurs

in considerable quantity in the digestive tract and offers evidence leading to the conclusion that the minute snow algæ constitute at least a part of the food of these worms.

A number of the collections made by Mr. Flett contained representatives of the associated animal life. Insects belonging to seven orders, Collembola, Hemiptera, Plecoptera, Coleoptera, Diptera, Lepidoptera, and Hymenoptera (several species in five of the orders), one species of Gastropoda, and three species of Araneida were found in the same habitat with the snow-worms. The Collembola (*Isotoma* sp.) occur in enormous numbers, especially on the snow below the glacier, making it black in appearance. In this respect the situation resembles that described by Moore ('99, p. 136) for *Mes. solifugus* which had great quantities of a collembolan, thought to be *Achorutes nivicola*, associated with it. All of these associated animals are black or very dark, except one species of spider and one species of Hemiptera, both of which are largely of a dark red color.

Mesenchytræus solifugus rainierensis n. var.

(Plate XVII, Figs. 20-26)

A collection from the upper snow-fields of Mt. Rainier, made on June 17, 1915, by Mr. Flett, contained seventy-five specimens of any enchytræid, blackish in color but smaller than *Mes. gelidus*. A thorough study of this material, which is in good histological condition, showed that it must be regarded as *Mes. solifugus* Emery. However, several structural characters fail to correspond to the descriptions of Emery ('98a, '98b, '98c, '00a, '00b), Moore ('99), and Eisen ('05). Previously known material of *Mes. solifugus* could not be secured for study but a very careful comparison of the characters of the material from Mt. Rainier with the published descriptions indicates that the deviations are apparently not sufficient to justify the separation into another species. Nevertheless, the variations are so constant that the writer feels convinced that it must be considered as a new variety.

Mes. solifugus was first described by Emery under the name *Melanenchytræus solifugus*, from specimens collected by Filippi on Malaspina Glacier, at the base of Mt. St. Elias, Alaska. The fol-

lowing year, Moore ('99) published a more extended account of the same species from material collected by Mr. H. C. Bryant, also on Malaspina Glacier. His paper contained not only the results of a careful anatomical study and a quoted account from Mr. Bryant of the habitat and the activities of these annelids, but also an interesting discussion of their relation to the peculiar habitat, with special reference to temperature and light, and an account of the possible function of the dense pigmentation. In the same material, Moore found another form which he described under the name *Mes. niveus*. He also rightly placed *Melanenchytræus* as a synonym of *Mesenchytræus*. Eisen ('05) published a short account of *Mes. solifugus*, specimens of which were collected by Professors T. Kincaid and W. E. Ritter during the Harriman Alaska Expedition on Muir Glacier and on La Perouse Glacier. It thus appears that up to the present account *Mes. solifugus* has been known only from glaciers in a very limited region of Alaska.

Definition.—Length, 12-18 mm., average about 15 mm. Diameter, about 0.52 mm. Somites, 51-60, average 55. Color, very dark brown to black. Prostomium rounded, smooth, blunt. Head pore at tip of prostomium. Setæ sigmoid, abruptly bent at distal ends; uniform in size and shape; in anterior fourth of body, 2 setæ, rarely 3, per bundle in lateral rows; in remainder of body, 1 seta, rarely 2, in lateral rows; in anterior third of body, 2-4 setæ, usually 3, per bundle in ventral rows; 2 setæ per bundle in ventral rows in remainder of body. Clitellum absent. Brain but very slightly longer than wide; anterior margin deeply emarginate, posterior margin shallowly concave, lateral margins approximately parallel. Origin of dorsal blood-vessel in XIII-XIV; small cardiac body present. Nephridia with small, slender, anteseptal part and large, irregular, lobate postseptal part; origin of efferent duct on ventral surface of latter very near posterior end. Spermiducal funnel moderate, cylindrical, tapering slightly towards origin of sperm duct; length about three times the maximum diameter; collar absent. Sperm duct very long, extending caudad within ovisac to XX; masses of coils in XIII-XIV; diameter uniform throughout. Ovisac extending from XII/XIII to XX, bifurcating in XIII or XIV. One pair of sperm sacs extending from XI/XII to about

XV; within ovisac. Penial bulb large, subglobular; well-developed, fusiform atrium with five atrial glands; numerous groups of multicellular glands within bulb. Spermathecae confined to V; duct short, cylindrical, numerous unicellular glands externally; ampulla but little greater in diameter than duct, elongate, tapering slightly anteriorly and uniting in posterior part of V with lateral aspect of digestive tract; two opposite, elongated, cylindrical diverticula reflected caudad.

The description is based on twenty-seven sexually mature specimens. Forty-eight other specimens of uncertain sexual maturity were examined in the study of external characters. The type and most of the paratypes are in the collection of the writer. Paratypes have been deposited in the collection of the United States National Museum and in the collection of Professor Frank Smith.

The habitat of this species will be described in another part of the paper.

External Characters

The body is elongate, cylindrical, smooth, and uniform in diameter except at the extreme anterior and posterior ends where there is a slight but gradual tapering. The length of the alcoholic specimens varies from 12 to 18 mm., the average length of twenty-seven sexually mature specimens being approximately 15 mm. The diameter varies from 0.47 to 0.63 mm., the average being about 0.52 mm. Except in the extreme anterior region, the external segmentation is indistinct, the surface being smooth and the intersegmental grooves difficult to detect. As in *Mes. gelidus*, external examination shows the presence of ruptures in some of the intersegmental grooves, thus producing whitish rings between somites. Longitudinal sections show that these ruptures are breaks in the cuticula and hypodermis and the underlying muscle-layers are thus exposed, producing the whitish band. Such ruptures are present in almost every specimen sent to the writer and occur uniformly in the middle region of the body, the anterior and posterior ends only being invariably free from them. No information is available at present concerning the origin of these interruptions. Moore ('99, p. 126) reports a similar feature in some of the specimens

of *Mes. solifugus* which he examined. He also states that Mr. Bryant, the collector who secured the material on Malaspina Glacier, Mt. St. Elias, and furnished data on the living animal, informed him that these white or yellow bands "were present when the worms were collected". On a later page (p. 135), Moore quotes directly from an account by Mr. Bryant, a part of which is as follows: "Some of the specimens I obtained had also distinct whitish bands around their bodies." From this account it might appear that these light bands occur in the living worms. Whether it is true of *Mes. gelidus* and *Mes. solifugus* var. *rainierensis* remains to be discovered. It seems unlikely, from an examination of the available material, that these interruptions are normal. Longitudinal sections show ragged edges of tissue about these interruptions, a fact which seems to constitute evidence against such a view. The appearance suggests the killing and preserving operations as the cause, although none of the specimens showed signs of extreme contortion or severe treatment.

The external surface of each somite is smooth and free from ridges, secondary constrictions, or striations of any sort. The number of somites varies from 51 to 60, the average being about 55. Except at the extreme anterior and posterior ends, the somites are of approximately uniform width. The color of the entire body is very deep brown to black, except for the above-mentioned whitish bands between somites. To the unaided eye, the specimens are black but under magnification, using transmitted or reflected light, they are a deep, rich brown. The distribution of the color is almost uniform throughout the body. Specimens, cleared and mounted *in toto*, show on the surface, except on XI-XIII, innumerable, minute, polygonal areas, usually hexagonal, each of which has a long and short dimension, at right angles to each other, the former extending in the direction of the circumference of the body. Each area contains in its center a light, oval spot.

Certain special, external areas are distinctly lighter in color than the surrounding surface. The tip of the prostomium and the lateral parts of the groove O/I are yellow and offer contrast with the adjacent parts. The ectal opening of the spermatheca in IV/V is surrounded by an ovate, yellowish area which, however, is not as

distinct as in *Mes. gelidus*. The posterior end of the last somite and the immediate vicinity of the penial bulb invaginations are also much lighter than surrounding parts.

These specimens lack the conspicuous, elliptical, swollen areas about the ectal openings of the spermathecae which Moore ('99, p. 125; Figs. 1, 2) found in the Malaspina Glacier specimens. Such areas are represented only by the lighter color in the immediate vicinity of the openings described above. Emery and Eisen make no mention of these areas.

A distinctly differentiated clitellum seems to be absent. The region of XI-XIII differs from the adjacent somites only in the obscurity of the intersegmental grooves and the slightly increased body diameter in that part containing the penial bulbs. Transverse and longitudinal sections of sexually mature specimens show no increase in the thickness of the hypodermis. The apparent absence of a clitellum causes some difficulty in distinguishing sexually mature individuals in superficial examination but in the material on which this paper is based it has been found that all specimens showing distinctly protruding penial bulbs are sexually mature. The uniform absence of a differentiated clitellum might cast some doubt, at first thought, upon the sexual maturity of all of the specimens but serial sections and dissections have demonstrated the presence of spermatozoa in the spermathecae, developing ova and spermatozoa in the storage sacs, and well developed ovaries and testes, thus furnishing proof of sexual maturity. Moore ('99, p. 125) reported that "In none of the specimens examined (about twenty in number) is the clitellum very distinctly developed, but on the contrary is thin and scarcely extends beyond the limits of the twelfth somite." Eisen ('05, p. 59) found it "probably confined to XII."

The head pore is distinct and located very near the tip of the prostomium. The external opening is slit-like in appearance, transverse in position, and surrounded by a very narrow, yellowish area.

The setae are distinctly sigmoid and arranged in fan-shaped bundles, two lateral and two ventral. Those of a bundle are of approximately equal development. Anterior to the penial bulb invagination, the lateral bundles contain 2 setae, very rarely 3;

posterior to that region, they contain 1 seta, very rarely 2. Anterior to the vicinity of XX, the ventral bundles contain 2-4 setæ, usually 3; posterior to that region, they contain 2 setæ. The setæ are somewhat distinctive in the very abrupt bend at the distal, exposed end (Pl. XVII, Fig. 24). The proximal end is broadly curved and deeply imbedded in the body-wall. A comparison with the descriptions of Moore and Eisen shows that the number of setæ per bundle which they report is greater than has been found in the Mt. Rainier material. In the latter, the writer has found but a single bundle which contained five setæ. It is not possible to determine whether the abrupt bend at the distal end of each seta described above is a characteristic only of *rainierensis* since Moore ('99, p. 126) states that in his Alaskan specimens "The setæ have the form usual in the genus, being feebly sigmoid and arranged in fan-shaped bundles, but are mostly imperfect, owing to the points being worn or broken off." He also reports that "Enlarged setæ are found in the ventral bundles of XI; these are about one-third longer and much thicker than the others." Such enlarged setæ have not been found in the specimens from Mt. Rainier. Emery ('00b, p. 225, Fig. 10) states that "The *chaetae* are slightly sigmoid, more markedly bent at their apical end (Fig. 10). They are about a third longer in the posterior half of the body than in the anterior segments, as it appears by comparing Figs. 12 and 13. Each bundle consists of four nearly equal chaetae. The ventral bundle is absent in the 12th (clitellar) segment, which receives the opening of the sperm-duct." From this description it might appear that the setæ resemble closely those of *rainierensis* but the figure which accompanies Emery's description portrays setæ quite different from those found in the Mt. Rainier specimens. Instead of being very slender, approximately uniform in diameter, distinctly sigmoid, broadly curved at the proximal end, and abruptly curved at the distal end, they are short and very stout, having, in the posterior bundles, a diameter of one-sixth the length. The diameter increases from the distal end to the opposite extremity which is almost straight. The distal end is very acute.

Internal Characters

In many respects, the internal anatomy corresponds to that described, by previous observers, in the Alaskan form and for that reason only new data and the features which present differences will be discussed.

Brain.—Dissections and frontal sections show the brain (Pl. XVII, Fig. 22) to correspond, in a general way, to the accounts of the same organ in Alaskan specimens. It resembles more closely the figure and description given by Moore, differing in being a little longer than broad and in having the posterior margin distinctly but shallowly concave. However, these are minor differences. It also resembles the figure given by Emery ('00b, Fig. 2) except that the concavity of the anterior margin is much deeper. A typical measurement is as follows: maximum length, 0.137 mm.; maximum width, 0.129 mm.; maximum thickness, 0.043 mm. A supporting strand extends latero-caudad from each latero-caudal angle to the body-wall.

Dorsal Blood-vessel.—According to the previous descriptions of *solifugus*, the origin of the dorsal blood-vessel occurs in XII. In the Mt. Rainier specimens, this vessel becomes distinct from the perivisceral blood-sinus in the posterior part of XIII or the anterior part of XIV. In none of the specimens examined did it arise in XII. An inconspicuous cardiac body is present.

Septal Glands.—In position, the septal glands agree with the description by Moore ('99, p. 127) but cannot strictly be called "large". Eisen ('05, p. 59) found the "Septal glands small" but the position is not indicated. Of the two indefinite terms, the latter is more nearly correct for the specimens studied by the writer.

Emery ('98a, pp. 110-111) includes the following statement in his original description of *solifugus*: "Nei segmenti 4-8 la cavita viscerale è in gran parte occupata da ghiandole unicellulari, i cui lunghi e sottili condotti sboccano all' esterno in vicinanza dei gruppi ventrali di setole." Michaelsen's description of *Mes. solifugus* ('00, p. 87) contains the following: "Liebhöhle des 4.-8. Segm. von grossen einzelligen Drüsen erfüllt (Kopulationsdrüsen?), die in der Nähe der ventralen Borstenbündel ausmünden." This statement, evidently taken from Emery, contains the tentative suggestion

by Michaelsen as to the character of the glands. Emery ('00b, pp. 226-227) in his later and more complete paper, describes these glands as follows: "In the segments 4-8, the most part of the body-cavity is filled by *unicellular glands* (Fig. 11 gl); their very thin excretory prolongations form numerous threads directed towards the ventral side, which can be easily followed on the sections to the sides of the ganglion chain. Their thinness and flexuous course make it difficult to follow them to their end on the surface of the skin. I believe that they converge towards the bundles of chaetae of the ventral series. As Mr. Michaëlsen writes me, these glands may be regarded as morphological equivalents to those gland-cells which in other Enchytraeids are related to the chaetae of the genital segments. In *Melanenchytraeus*, I don't think that these glands have any relation to the function of reproduction, because I find them no less developed in immature specimens." No mention of such special unicellular glands is found in the descriptions of Moore and Eisen and the writer has found no evidence of them in his material. Since the position of these unknown glands coincides with the position of the septal glands, as described by Moore and as found by the writer in *Mt. Rainier* specimens, it might be suspected that the two have been confused. Emery ('00b, Fig. 11) figures these glands as they appear in a longitudinal section of the body and although the component cells are not associated in close, compact masses, they are aggregated into rather loosely associated groups and columns with their prolongations approaching each other and extending in the same directions. The appearance of the whole very strongly suggests loosely constructed septal glands and since Emery worked with a very few sexually mature specimens which were not in the best state of preservation, the writer is inclined to suspect these masses of "unicellular glands" of being the usual series of septal glands.

Nephridia.—The nephridia (Pl. XVII, Fig. 25) agree closely with previous descriptions. A well-developed, ciliated nephrostome, borne on a slender base, comprises the anteseptal part. The post-septal region is enlarged, somewhat lobate, and slightly compressed. The structure of the interior is of the usual mesenchytræid type. The efferent duct arises from the ventral surface of the postseptal

part near the caudal end. In the specimens examined, there is considerable variation in the size and shape of the nephridia in different regions of the body but the general structure is fairly constant. The first pair is located on VI/VII. Moore ('99, p. 127) found nephridia "in every somite posterior to VII." No nephridia have been found on XI/XII and XII/XIII in the Mt. Rainier specimens.

Lymphocytes.—Lymphocytes are very scanty, in some specimens almost absent. A few scattering cells occur in the anterior region and occasional ones more caudad. They are small, elliptical, nucleated, and contain pigment-granules.

Spermiducal Funnel.—The spermiducal funnels (Pl. XVII, Fig. 23) depart somewhat from Moore's descriptions of Alaskan specimens of *solifugus*. Instead of being constricted at the middle and bent upon themselves so that the free end is directed caudad, they show only a slight diminution in diameter and a slight bend at the middle so that the free end is directed cephalo-dorsad. A much greater disagreement is found when comparison is made with Emery's figure ('00b, Fig. 16) of the funnel which is represented as a very short, funnel-shaped organ which is broader than long. However, Emery states that his figure was drawn from a reconstruction. The length is about three times the maximum diameter, a typical set of measurements being as follows: length, 0.344 mm.; diameter, 0.113 mm. The collar is entirely absent.

Sperm Duct.—The sperm duct is very long and of approximately uniform diameter. Because of its coiled and contorted condition, the exact determination of its length has not been possible. It extends from the end of the spermiducal funnel into the ovisac; forms contorted masses in XIII and XIV; continues caudad within the ovisac to XX; thence is reflected cephalad to XII to unite with the atrium. Throughout the entire course it is characterized by numerous coils and curves. Its extent is represented by the combined length of sixteen somites plus an allowance of the length of several somites to compensate for the contortions. Except for a very small part at the end of the spermiducal funnel and at the union with the penial bulb, the entire duct is contained in the ovisac, and does not, according to the writer's observations, enter either of the sperm sacs. Moore ('99, p. 129) found that, in his material,

the sperm duct on the right side lies within the ovisac and extends to the region of XVIII, but the one on the left side lies free in the body-cavity, extending also to the region of XVIII. Eisen's description contains no account of these ducts.

Sperm Sacs.—The sperm sacs are a pair of caudal evaginations of XI/XII, one on each side of the median line, which lie latero-ventrad of the digestive tract and extend to XV or XVI. In the specimens examined, they are rather small, but both are of about equal development. Both lie within the ovisac and both contain masses of developing spermatozoa. The structure of the walls of these sacs is identical with that of the septa.

Moore ('99, p. 128) found a different condition in Alaskan material. The sac on the right side is almost rudimentary, extending only into XII, while the one on the left side is well developed, extends caudad to XX, and is apparently not contained within the ovisac. Other descriptions do not include data on the structure and relations of these sacs. Allowing for a liberal variability, such differences seem rather large for intra-species variation.

Ovisac.—The ovisac is formed by the caudal outgrowth of XII/XIII. It arises as a single, well-developed sac and extends caudad, ventrad to the digestive tract, to the posterior part of XIII or to XIV where it bifurcates, forming two branches, one on either side of the median line, which extend to the vicinity of XX. There is some variation in the length since specimens were examined in which the caudal terminations were in XXII, while in others they were in XIX. As stated above, the ovisac contains the sperm ducts and the sperm sacs. In addition, it contains masses of developing ova. Here, again, is an interesting divergence from the condition described by Moore ('99, pp. 129-130) who found, in the Alaskan material, that the ovisac is single throughout its entire length and contains only the right sperm duct and the masses of developing ova. The length, however, is very similar in both cases.

Penial Bulb.—In the general plan of structure, the penial bulb (Pl. XVII, Figs. 20, 26) agrees with the description by Eisen ('05, p. 60) for Alaskan specimens. However, a number of differences exist in some of the finer structural detail. The whole

organ is of the mesenchytræid type as described by Eisen ('05, p. 7) and consists of three sets of structures, viz., the penial bulb proper, the atrium and its associated parts, and the accessory glands. The whole organ is conspicuous in size so that both bulbs occupy the greater part of the cœlom in that region.

The penial bulb proper is situated on a deep invagination which, in transverse section of the body, is slit-like but in longitudinal section appears as a narrow channel leading, at its inner extremity, into an expansion which takes the form of a chamber, narrow in dorso-ventral dimension but much wider in transverse dimension. This inner chamber caps the ental part of the invagination like the top to a mushroom. Structurally, the wall of the invagination is essentially a continuation of the body-wall. The ectal end of the sperm duct connects with the inner, expanded chamber. The body of the bulb is composed of (1) a large number (over twenty-five) of multicellular glands, (2) scattering unicellular glands, (3) a large number of muscle-strands, and (4) the ectal end of the sperm duct. The multicellular glands are pear-shaped, the expanded ends being entad of the invagination. Each is composed of a number of gland cells aggregated in the expanded ends and a narrow, tapering extension, composed of the elongated ends of the gland cells, which connects with the penial invagination. None of these glands opens into the sperm duct. The unicellular glands are sessile, very inconspicuous, and are scattered singly about the invagination into which they apparently open. They are the only glands which occur laterad of the invagination. In the interior of the bulb, muscle-fibers extend in different directions. They lie between the various glands and many of them are attached to the wall of the invagination. They vary in size from very fine threads to rather strong strands. The ectal end of the sperm duct, which is contained within the body of the bulb, has the usual structure except that it is surrounded by a very strong, longitudinal muscle-coat which is apparently derived from the longitudinal muscle-layer of the body-wall.

Within the penial bulb but near its ental surface, the sperm duct begins to expand into the atrium so that a small portion of the ectal part of the latter is included within the envelope of the

bulb. The atrium is a stout, fusiform organ, about 0.3 mm. long and 0.13 mm. in maximum diameter. Its histological structure is very similar to that indicated in Eisen's figure ('05, Pl. VIII, Fig. 1), except that the epithelium on the surface does not seem to be as distinct and thick. At the ental end of the atrium is a set of about five large, club-shaped, multicellular atrial glands, arranged in a whorl. These glands extend into the cœlom and are conspicuous organs in sections of that part of the body.

A number of large, multicellular, pear-shaped, accessory glands are present. They lie just outside of the envelope of the bulb and open into the invagination, commonly at its origin. In general structure, they resemble the multicellular glands within the penial bulb. The surrounding envelope is very delicate and it is difficult in some of the specimens to determine just how many of the glands at the edge of the bulb are to be classed as accessory.

The structure of the penial bulb and its associated parts in *solifugus* has been briefly described by Moore ('99, p. 129) and Eisen ('05, pp. 59, 61) as it occurred in the Alaskan material which they examined, and a comparison with the Mt. Rainier specimens is of interest. Moore states that "Before entering the atrium in somite XII the recurrent limbs of the sperm ducts expand into narrow fusiform sacs (.), having glandular, epithelial and muscular walls, which receive the ductules of a group of unicellular spermiducal glands. This structure probably serves to form and eject the spermatophores. A narrow curved duct, which is also provided with some unicellular glands, perforates the mesial wall of the atrium and opens into its lumen. Unlike the remainder of the male efferent apparatus, the atrium (.), is, in part, of ectodermal origin, as is indicated by the pigmented lining epithelium. It is a spheroidal thick-walled partly eversible sac, with an internal cavity having a mushroomlike shape in the retracted organ. Its walls are composed of a cuticle-covered, rather deep, pigmented and perhaps glandular epithelium, surrounded by a thick muscular layer in which the fibres are partly longitudinal, but largely radial, especially about the place of entrance of the sperm duct. A number of groups of unicellular glands are attached to the organ, and probably empty into its lumen."

At first sight, it might appear that a radical difference exists between the structure of the Alaskan and Mt. Rainier specimens. In the first place, a difference in the use of terminology is evident. Moore applies the term *atrium* to the structure which the writer and others call the *penial bulb*, and speaks of the expansion of the sperm duct just entad of the bulb as "narrow fusiform sacs". The writer follows Michaelsen ('00, p. 9) and Eisen ('05, p. 4) in using the term *atrium* to designate the enlargement of the sperm duct which is situated just entad of its union with the penial bulb. Apparently, Moore regards the atrial glands and the multicellular glands within the penial bulb as "groups of unicellular glands" rather than multicellular glands as described by the writer. If this interpretation of Moore's description is in error, then the structure of the penial apparatus in the Alaskan specimens is very different from that described in the present paper. No distinction is made in the above-quoted description between the penial glands within the bulb and the accessory glands.

Eisen ('05, pp. 59, 61) describes the penial apparatus in *solifugus* as follows: "A large atrium in which opens about eight atrial glands of large size. Many large accessory glands open along the base outside of the penial bulb. About fifteen penial glands inside the penial bulb. . . . The accessory glands, which are characteristic, open along the base of the penis outside of the bulb. They are long and of trefoil shape, with enormous long narrow ducts." It will be noted that Eisen found a greater number of atrial glands in his material. Furthermore, the accessory glands are evidently much longer and the number of penial glands in the bulb greater than in *rainierensis*.

Emery ('00b, pp. 227-228, Fig. 16) describes the penial apparatus as follows: "The last tract [Sperm duct] forms a spherical bulb (*a*), but before reaching it the tube presents a fusiform swelling (*c*), whose wall is very thick and made of long cells, directed radially on the transverse section, the lumen being not widened. Bundles of prostatic (spermiducal) glands (*b*) are related to the bulb; another little group of glands (*e*) lies around the tube, above its fusiform thickening." Some difficulty is experienced in interpreting this description, especially when the figure is con-

sulted. The spherical body of the bulb and the atrium agree with the other descriptions but the "prostatic glands" are represented as large, rather numerous, lobular organs which lie out in the body cavity, opening into the penial bulb much as do the corresponding organs in *Mes. gelidus*. The other group of glands referred to is of uncertain identity, judging from either the description or the figure. Emery's figure was made from the reconstruction of a series of sections and is possibly not fully dependable, although it seems improbable that a mistake could have been made in the matter of so large a group of glands as those which he calls prostatic. Assuming that his observations have been fairly correct, the penial apparatus differs from those described by Eisen and Moore as well as from *rainierensis*.

Spermatheca.—The spermatheca, in all of the specimens examined, consists of (1) a short, stout, cylindrical duct, covered externally, in part, by numerous attenuated, unicellular glands, (2) two almost oppositely placed, elongated, cylindrical diverticula which are directed caudad, and (3) an elongated, slightly curved ampulla which decreases slightly in diameter towards the ental end and joins the digestive tract independently on its lateral aspect. The character of the area surrounding the external opening of the duct has already been discussed.

Certain variations are apparent when these organs are compared with the descriptions of *solifugus* from other localities. Moore ('99, pp. 130-131) found three spermathecal diverticula. He also found that the ampullæ of the two spermathecæ unite to form a short duct before joining the digestive tract on its dorsal side. No mention is made of unicellular glands on the duct. Eisen's description ('05, p. 60; Fig. 32b) agrees with that of Moore in almost every respect. However, his figure shows, on one of the spermathecæ, one diminutive and three equally developed diverticula, indicating a possible variation, although no mention is made of it in his description. Emery's original description of *solifugus* ('98a, pp. 110-111) contains the following statement: "I ricettacoli del seme non comunicano con l'intestino; sono in continuità l'uno coll', altro ed hanno ciascuno, alla base della loro ampolla, due o tre diverticoli." His more complete paper ('00b) corroborates this

statement. It appears, from the above, that specimens with as few as two spermathecal diverticula were found. The reported absence of the communication of the ampulla with the digestive tract may be an error as has been suggested by Moore. Emery ('00b, p. 230) re-examined his material and found no connection but states that the spermathecae lie in close contact with the intestine and points out the possibility that his single sectioned specimen might be not fully mature or abnormal.

Pigmentation.—All of the specimens from Mt. Rainier are deeply pigmented. Microscopic examination of sections of all parts of the body shows that the hypodermis bears a heavy load of minute, dark brown, non-staining pigment-granules, especially in the outer ends of the cells. This pigmentation extends to the internal organs. The chloragog cells, which first appear in IV and are present throughout the remainder of the body, are heavily loaded with pigment. It also occurs in the glands surrounding the bases of the setæ, in the lymphocytes, in the lining of the buccal cavity and the pharynx, in the spermathecal duct, in the lining of the penial invagination, in the nerve cord, and in the brain. It will be noted that this distribution of pigment corresponds closely with that described by Moore ('99, p. 127) for specimens of *solifugus* from Alaska. Eisen ('05, pp. 59, 60) states that the pigment is distributed in the body-wall and also in most of the internal organs "even in the ganglia and the brain" but does not specify further detail.

Summary of Comparisons.—The variety, *rainierensis*, differs from the Alaskan specimens of *solifugus* in the following respects: (1) the smaller number of setæ per bundle, (2) the absence of enlarged setæ on XI, (3) the origin of the dorsal blood-vessel in XIII-XIV, (4) the concavity of the posterior margin of the brain, (5) the constant presence of only two diverticula on the spermatheca and the independent opening of each spermatheca into the lateral wall of the digestive tract, (6) the shorter accessory glands, the smaller number of atrial glands, the larger number of multicellular glands within the penial bulb, and the straighter spermiducal funnel, and (7) the complete enclosure of the sperm sacs and sperm ducts by the ovisac. All of these differences seem

to be constant and are sufficient to raise the question as to whether they are too wide to be considered within the range of intra-species variation. However, they are differences of narrow margin and when compared with the sum total of the points of agreement with *solifugus* as described, the writer is convinced that the Mt. Rainier material is not a distinct species but can be considered only as a new variety.

BIOLOGICAL NOTES

As stated before, all of the information concerning the living specimens has been furnished by Mr. Flett. He found *Mes. solifugus* var. *rainierensis* abundant on the higher snow-fields and glaciers of Mt. Rainier in early summer. The collection which has furnished the material for the preceding description was made on June 17, 1915, at an altitude of 7,500 ft. Nothing is known concerning the winter or early spring conditions since at those seasons collections at such heights are not possible. These worms occur on snow-fields which seldom thaw during the summer and they evidently pass the entire existence, generation after generation, in the snow and ice. Mr. Flett states that on one occasion he has seen what he thought was this worm at an elevation of only 6,000 feet where the snow melts and grass and flowers grow in profusion during three or four months of the year. However, this was an unusually low altitude for these worms since they occur regularly and more abundantly much higher up the mountain on the permanent snow-fields and on the snow and ice of the glaciers. The writer has not had the opportunity of studying specimens from an altitude as low as 6,000 feet, but if the worms seen at that level are *solifugus* var. *rainierensis*, as Mr. Flett thinks they are, it appears that those individuals which chance to be developed at the unusually low levels must pass a part of the life history in midsummer on or in the ground.

On the glaciers, these worms coil up so as to appear as small spherical black dots on the snow or solid ice and it requires a considerable exposure to sunshine to warm them up to the active stage. According to the observations of Bryant (Moore, '99, p. 134), the specimens of *solifugus* on Malaspina Glacier, Mt. St.

Elias, "remain on the surface during the night; but when the sun appears in the morning they again burrow into the snow". This does not agree with the observations of Mr. Flett for the variety *rainierensis* since he has noticed no tendency on the part of the worms to avoid sunlight, the warmer the day the more active they become, this activity being manifested on the surface. The period of greatest activity is usually from the middle of the afternoon to about five or six o'clock.

No data on the associated life are available except the statement by Mr. Flett that snow algæ ("*Sphærella nivalis*") occur in great abundance and that spiders and snow-fleas (*Collembola*) are present.

The question of the food of a worm living in such a habitat is of considerable interest. Obviously, in the case of those individuals living on the permanent snow-fields and the ice of the glaciers, the number of substances serving as food are extremely limited. Emery ('00b, p. 226) found the contents of the intestine of Alaskan specimens, especially in the posterior part, to consist of "very fine crystalline mineral detritus, which seems to be the ordinary food of this worm". An examination of the intestinal contents in the Mt. Rainier specimens yielded practically no definite data. A certain amount of what seems to be fine, angular, mineral particles is present. However, the conspicuous content was a more bulky material, which the writer was unable to identify. In some respects, it has the appearance of partly disintegrated vegetable matter. This material was found in every specimen examined and evidently represents a part of the usual food.

GENERAL CONSIDERATIONS ON SNOW-FIELD AND GLACIER WORMS

Temperature Relations.—A striking thing about the environment of these enchytræids is the very low temperature of the medium in which they live. They exist and carry on their life processes under freezing temperatures and in a medium of snow and ice. Species, such as *Mes. gelidus*, which regularly occur at an altitude below the limit of permanent snow-fields evidently pass

a part of the life history on or in the ground, but aside from the midsummer months they are covered with snow and live in the latter or else in the earth beneath it. In either case, they are passing the greater part of the year in cold conditions, appearing in the melting snow in early spring. On the other hand, *solifugus*, which regularly inhabits the permanent snow-fields and the ice of the glaciers, spends its entire existence, generation after generation, under these conditions. Apparently, it must deposit its eggs in the snow, on the ice, or in the small pools of ice water which sometimes occur on the surface of the lower parts of the glaciers, and the young worms must be able to withstand the rigid conditions and successfully solve their problems of maintenance.

Pigmentation.—Nothing is known concerning the rôle of the large amount of pigment. Reference has already been made to the speculative discussion of this problem by Moore ('99, pp. 135-142). It may be related to the maintenance problems of heat or light, or both, but until some careful observations and experiments are made no definite conclusions can be drawn. If such pigmentation were confined exclusively to glacier forms the circumstantial evidence might be stronger in favor of a theory that it is an adaptation to some of the factors peculiar to that environment, perhaps light and temperature, but the fact that species other than glacier forms are known (*Mes. harrimani* Eisen, *Mes. obscurus* Eisen, *Mes. maculatus* Eisen, and others) which possess similar pigmentation throws some doubt on such an assumption. If it be true, as is suspected in some cases, that the younger stages of these glacier worms are not pigmented, a critical study of the life history may be necessary to the accurate solution of this problem.

Other Snow-field and Glacier Worms.—It is possible that future investigations of the northern glaciers and the arctic snow-fields will reveal still other enchytræids which occupy these frigid habitats and extend the knowledge of the distribution of the species already known. Certain arctic explorers make mention of the presence of "worms" on the snow and ice and while no hint is given of their identity, it is quite possible that they are enchytræids. Mr. Flett states that he found a snow-worm in the Olympic Mountains which occurred in enormous numbers, making the snow black,

and which resembles *Mes. solifugus* var. *rainierensis* in size and general appearance, but since material has not been secured its identity is unknown. Among the collections from Mt Rainier, there is at least one other enchytræid which appears to be distinct from those described in this paper but has not been carefully studied.

Thus far, all of the known glacier worms belong to the genus *Mesenchytræus*. While our knowledge concerning the geographical distribution of *Enchytræidæ* is incomplete, it appears fairly certain that the family is one of temperate and frigid distribution. Ude ('01, p. 23) found, among other things, that available data showed that "Die Gattungen *Mesenchytræus* und vielleicht *Henlea* (vergl. *H. ventriculosa* (Udek.) lassen in ihrer Verbreitung Cirkumpolarität vermuten." Certain other genera (*Bryodrilus*, *Lumbricillus*, and others) have representatives in the arctic zone but none of them seem to have been reported as living continuously in snow and ice. Whether this habit is confined to *Mesenchytræus* is a problem for future investigation.

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EXPLANATION OF PLATES

ABBREVIATIONS

ac. gl.,	accessory gland.
atr.,	atrium.
atr. gl.,	atrial glands.
cut.,	cuticula.
ec. op.,	ectal opening.
hyp.,	hypodermis.
in. pen. gl.,	intra-penial glands.
lum. amp.,	lumen of ampulla.
lum. div.,	lumen of diverticulum.
ov's.,	ovisac.
pen. b. i.,	penial bulb invagination.
pen. ch.,	penial chamber.
pen. gl.,	penial glands.
pen. po.,	penial pore.
r. m.,	retractor muscle.
sp. d.,	sperm duct.
sp'r.,	spermatheca.
sp'r. d.,	spermathecal duct.
sp. s.,	sperm sac.
I-XXXV.,	somites.

PLATE XIV

Mesenchytræus gelidus

- Fig. 1. Diagram of XI-XXXV, showing position and extent of ovisac, sperm sacs, spermiducal funnels, and sperm ducts. Broken lines indicate omission of somites.
- Fig. 2. Diagram of I-X, showing extent of the exceptionally large spermatheca. The broken lines across spermatheca in posterior part of VIII indicate most anterior observed termination of these organs.
- Fig. 3. Longitudinal section through ectal region of spermatheca.
- Fig. 4. Lymphocytes, indicating abundance of pigment-granules in cytoplasm.
- Fig. 5. Chloragog cells. Pigment-granules distributed through cells.
- Fig. 6. Longitudinal section through ectal opening of spermatheca, showing unicellular glands which occur in connection with end of spermathecal duct.

PLATE XV

Mesenchytræus gelidus—cont.

- Fig. 7. Spermatheca.
Fig. 8. Spermatheca.
Fig. 9. Setæ. Bundle from ventral row.
Fig. 10. Brain.
Fig. 11. Penial bulb and associated structures.
Figs. 12-13. Spermathecæ from one specimen, drawn to same scale, showing a rather exceptional form of variation in development.
Fig. 14. Nephridium.
Figs. 15-17. Diagrams of different forms of spermiducal funnel.
Fig. 18. Nephridium.

PLATE XVI

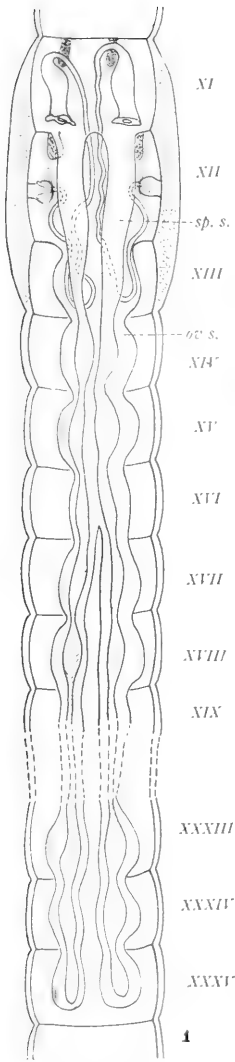
Mesenchytræus gelidus—cont.

- Fig. 19. Longitudinal section through penial bulb.

PLATE XVII

Mesenchytræus solifugus var. *rainierensis*

- Fig. 20. Penial bulb as it appears in transverse section of body.
Fig. 21. Spermatheca.
Fig. 22. Brain.
Fig. 23. Spermiducal funnel.
Fig. 24. Setæ.
Fig. 25. Nephridium.
Fig. 26. Penial bulb and associated structures.



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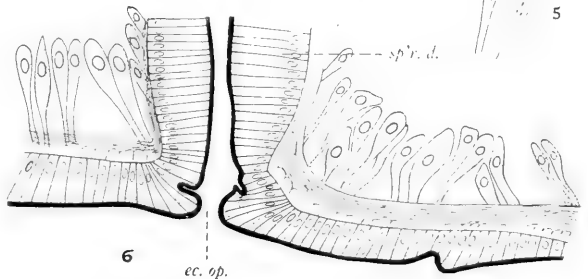
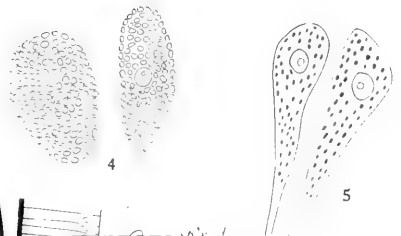
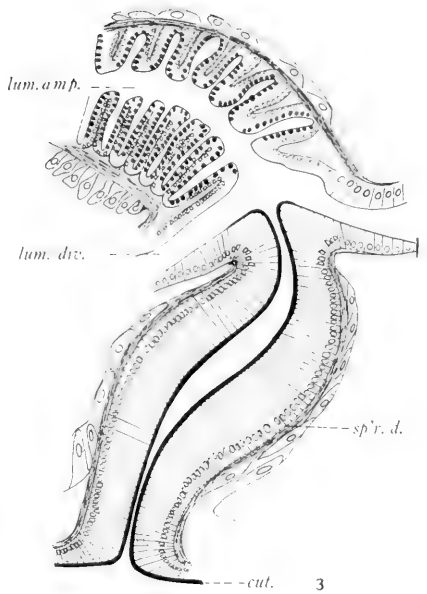
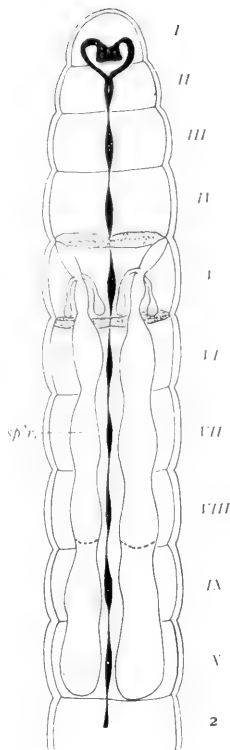
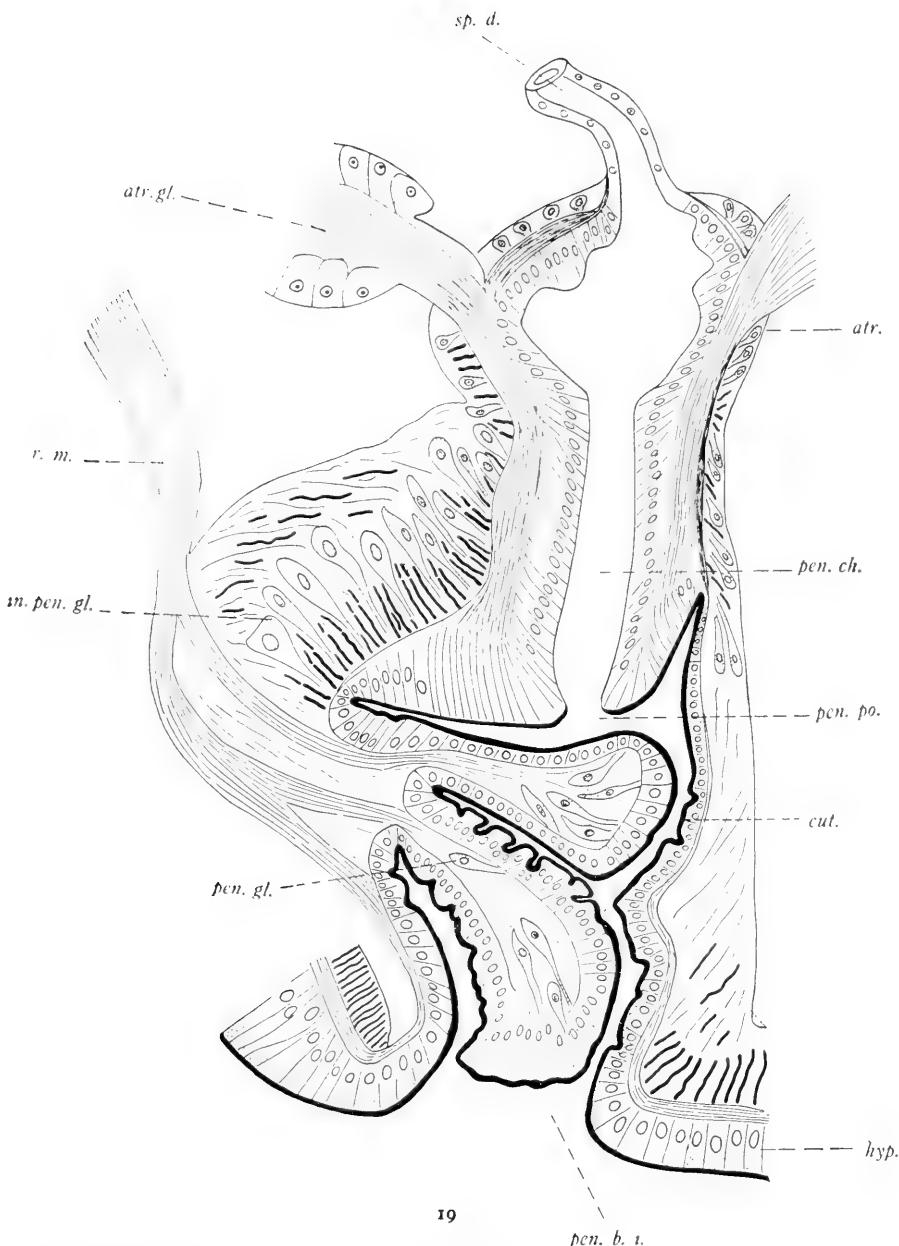


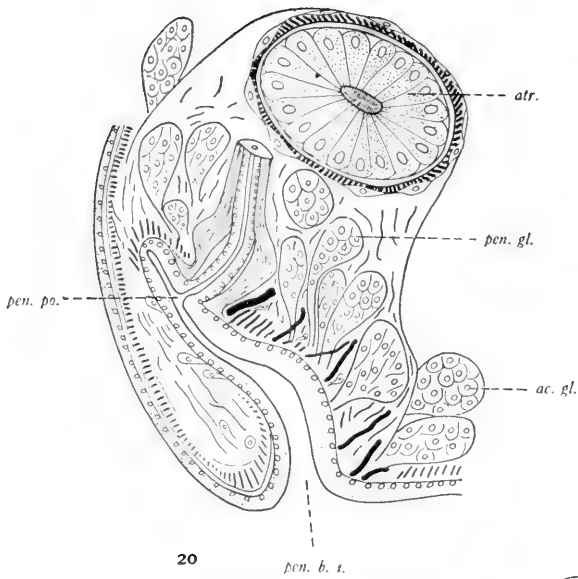
PLATE XIV



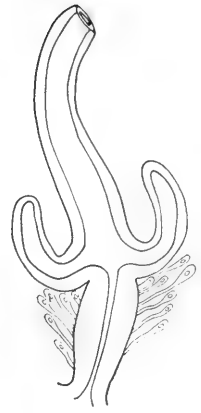
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PLATE XV

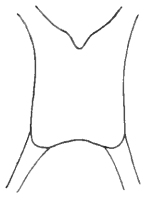




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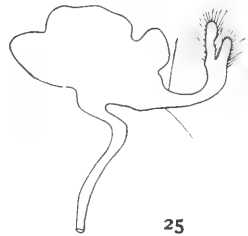
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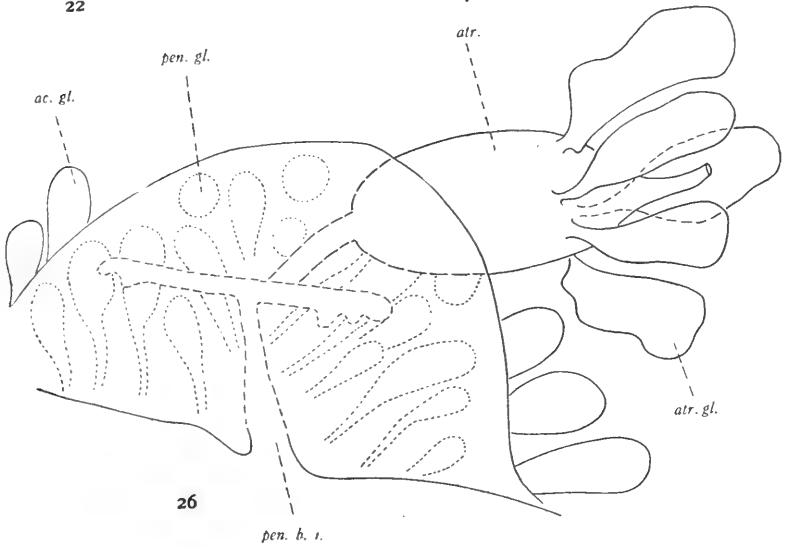
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ON THE SO-CALLED INTESTINAL GLANDS IN NECTURUS MACULATUS

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Introduction.

Imbedded in the submucosa and connected to the mucosa in the intestines of *Necturus* are groups of cells whose function has not been determined. The finer structure and function of these groups of cells constitute the object of the present work.

Groups of cells of similar character have been described in *Proteus*, the European Salamander, Newt, Triton and *Amblystoma*, but have never been mentioned in connection with other animals. It is of interest to note that forms as closely allied as *Rana* do not possess these groups of cells, nor, according to a recent paper by A. M. Reese, does the Alligator. Since the forms mentioned above represent two of the three suborders of *Urodeles* it is probable that they are characteristic of *Urodeles* only.

The function of these groups of cells may be similar in the different animals which possess them, but two views have been held as to their function. One view is that they are glands discharging into the intestine. This other view is that they serve as proliferation centers where the epithelial cells which constitute the musoca are produced.

Various names, more or less suggestive of function, have been applied to these structures, such for example as "gland," "Sprossen" (sprout), "Bud," "Zapfen" (spigot) and "Ersatzzellen" (compensation cells). It is clear that it is not advisable to use for these structures, a name which suggests a function until their function has been ascertained. So, for the sake of convenience, I shall use the term "protuberance" inasmuch as it does not suggest a function.

In the present work I shall confine myself to these protuberances in *Necturus*.

Review of Literature.

Hoffman (1878) was probably the first writer to mention these protuberances in *Necturus*. I have not been able to see Hoffman's work but according to B. F. Kingsbury, Hoffman described these

protuberances in *Necturus* as glands and "speaks of their almost circular opening upon the surface epithelium of the intestine."

Oppel in 1889 saw protuberances in the intestinal submucosa in *Proteus anguineus* and pronounced them glands (Drüsen). He noted that mitotic cells were abundant in the structures and in the intestinal mucosa in the immediate vicinity of the structures.

Protuberances in the intestinal mucosa of *Triton* were described in 1892 by Bizzozero. He referred to them as groups of compensation cells (Ersatzzellen) and sprouts (Sprossen). He said that cells proliferate not only at the base of the mucosa but press the wall of the mucosa out in places to form sprouts. For saying that these sprouts are epithelial, he gave the following reasons, which are summarized from the translation; (1) their general character and constitution are those of epithelial tissue, (2) cells can be seen in all stages of transformation from the cubical cells of the sprout to the columnar cells of the mucosa. He remarked that mucous granules can be seen between the epithelial elements and that a great many mitoses are present in the sprouts.

In *Salamander* protuberances have been described by Nicholas (1894) who called them proliferation buds (bourgeons germinatifs). Nicholas said that they represent cell proliferation centers, and that they could not be glands inasmuch as they possess no lumen. He said that dividing cells were to be seen only in the buds (bourgeons) or in the surface epithelium near the neck of a bud.

Kingsbury in 1894 working on the enteron of *Necturus* regarded these protuberances as glands. He wrote "my study of them in *Necturus* would, however, lead me to regard them as glands. The arrangement of the cells as if surrounding a lumen, and indeed a lumen itself, which Nicholas declared did not exist in these structures, could be seen upon almost all of my sections in some of the glands. I was unable, however, quite satisfactorily to demonstrate the existence of a neck, although it seemed in numerous glands to be quite well indicated."

The work of Bizzozero was resumed and confirmed by Sacerdotti in 1896. The latter also working on *Triton* sought to ascertain the relation between the period of cell division and the period of mucous secreting activity. He determined that karyo-

kinetic forms were assumed by cells which had already begun to secrete mucous, that these karyokinetic cells are found in the deeper parts of the epithelial layer but more especially in the epithelial spigots (Zapfen) pressing into the connective tissue. He added that occasionally and exceptionally karyokinetic cells were seen near the surface of the epithelium. Sacerdotti explained their presence in that region by saying that the cells in the animal concerned were possessed of very great growth capacity and thus certain mitotic cells had been forced out of the epithelial spigots and had reached the surface while still in mitosis.

Bates in 1904 described protuberances in the American *Amblystoma* and remarked that their structure would suggest glandular activity yet said he was not able to demonstrate the existence of a lumen.

Technique.

Inasmuch as the main object of my work was to determine the finer structure of these protuberances and with a view to distinguish whether they were glands or not, I endeavored to obtain a technique which would give to glandular secretions a differential coloration. For this purpose some of Dr. R. R. Bensley's special methods for glands were used.

Mallory's triple stain (Guyer, *Animal Microcology*, p. 172) was used extensively. It proved to be especially rapid and practical for general work. It gave a conspicuous differential coloration to connective tissue and epithelial tissue.

Weigert's Hematoxylin (Guyer, p. 178) proved to be very effective for preparing slides for the purpose of studying mitosis and details of nuclear structure.

Some of the animals were killed very soon after being brought into the laboratory, others were kept in an aquarium for a few months. Most of the fixing was done in Gilson's fluid but some fixing was done in Bensley's Acetic Osmic Bichromate fluid.

Description.

These protuberances in *Necturus* are located for the most part at the peripheral or outer parts of the folds. Occasionally one is found on the side of a fold a short distance from the peripheral bend. In one animal, however, out of over twelve which were

studied, in which the intestinal mucosa was more abundant and more folded, I saw a few of these protuberances occupying positions at the very central part of the sections at the central apices of some of the folds.

As for the distribution of these protuberances throughout the length of the intestine, I found that they are more abundant anteriorly than they are posteriorly. To ascertain this, I made a count of the number of protuberances that appeared in cross sections in the respective regions. The count involved twenty slides from each region from about five different animals. The results of the count are here summarized.

Region	Maximum No.	Minimum No.	Average No.
Duodenal region	109	43	70.3
Middle intestine	104	45	66.6
Rectum	75	37	52.6

Thus it is seen that the number of protuberances decreases with distance from the pylorus.

The size of these protuberances varies in diameter from about 50 micra to about 127 micra. In length they vary from about 127 micra to about 293 micra. These measurements made with a stage micrometer and an ocular micrometer, are the extreme measurements of a great many taken.

The shape of these protuberances is naturally variable but it is usually possible to distinguish three parts. (1) the body, that part of the structure which lies wholly outside the mucosa, (2) the mucosal portion, or the portion that lies wholly within the mucosa and (3) the neck which connects the body and the mucosal portion. I shall describe each part separately and in detail.

The body is commonly roughly spherical in shape but is also frequently columnar or conical with the point more or less blunt. The body is also frequently branched. These structures are invariably connected to the mucosa. In mounted sections one sees many bodies apparently not connected to the mucosa but by tracing such bodies through serial sections a connection with the mucosa could always be demonstrated.

Those protuberances that are situated quite at the peripheral ends of the folds almost always extend into the submucosa in a direction perpendicular to the mucosa. If however the protuberances are situated on the sides of the folds, as related above, the body part bends over so as to lie close to the mucosa. In other words, the body parts of the protuberances always extend in a direction radiating from the center of the intestine.

The cells in the body portions are polygonal in shape. The nuclei are more or less spherical and occupy nearly all of the cell. I studied carefully the narrow zone of protoplasm which surrounds the nucleus in cross sections to see if I could discover any granules the presence of which would suggest a secretory activity, but none of my preparations showed any.

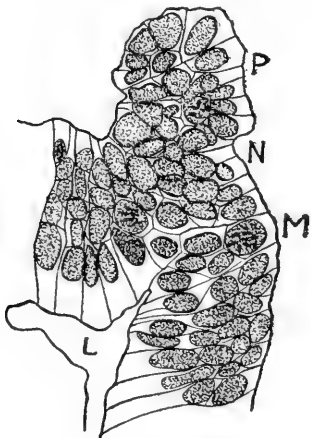
I paid particular attention to the arrangement of cells within the body, and was unable to demonstrate a universal definite order of arrangement. This is contrary to the findings of Kingsbury, who stated that the cells were arranged as if surrounding a lumen, and added that a lumen itself could be seen in some of his sections. I was unable to discover a lumen nor did I find an arrangement of cells that would suggest the presence of a lumen.

Therefore, I have concluded that these protuberances cannot be glands. I consider the presence or absence of a lumen a sufficient criterion on which to base such a conclusion, for if these structures were glands the existence of a duct through which the secretions might pass and a corresponding arrangement of cells would be necessary.

The mucosal portion is that portion which extends into the epithelial mucosa. It commonly spreads out within the mucosa so that cross sections appear fan shaped. There is a gradual transition in the shape of the cells of the body of the protuberance and the columnar cells of the mucosa. The transition takes place in the mucosal portion of the protuberance. The cells in the mucosal portion of the protuberance become more and more elongate and are arranged in crescentic layers in optical section, as if suspended hammock-like from the outer boundary of the mucosa. The farther the cells are from the center of the protuberance, the more columnar they are. The nuclei become more and more oval.

The neck of the protuberance is the portion at the peripheral boundary of the mucosa, which connects the body of the protuberance to the mucosal portion. It is almost always somewhat constricted.

Cells in various stages of mitosis are frequent in all parts of the protuberance. To determine the ratio of mitoses present to the total number of cells in the protuberances, I made a count involving several thousands of cells and calculated that the ratio of mitotic cells to the total number of cells is one to forty-one. There are an average number of 39 cells in a section of a protuberance.



Camera lucida drawing of a portion of a cross section of the mucosa of the mid intestine of *Necturus maculatus* showing protuberance P, lumen L, neck N, mitotic cells M. The cell walls were not drawn by means of a camera lucida. Spencer Ob. 10X, Oc. 4.

Thus there is an average of about one mitotic cell in a protuberance. In some protuberances I saw three cells in mitosis while, on the other hand, there were many sections of protuberances that did not show a single mitosis.

It is significant that not one case of mitosis was seen in the mucosa at any considerable distance from a protuberance.

Upon these facts I have concluded that cells which are to compose the intestinal mucosa in *Necturus*, are formed in these protuberances. It would seem that the cells are forced through the neck of the protuberance, while perhaps sometimes still in mitotic condition. In the mucosal portion of the protuberance, the cells appear to be forced in a lateral direction along the mucosa. Since mitotic cells are not found at points along the mucosa excepting near the protuberances, it may be concluded that the intestinal mucosal cells do not divide after they have become functional cells.

According to this view, these protuberances in *Necturus* are cell proliferation centers for the mucosa as was concluded for some other Urodeles by Bizzozero and Nicholas.

Mitchell, South Dakota.

FILICOLLIS BOTULUS N. SP., WITH NOTES ON THE CHARACTERISTICS OF THE GENUS*

By H. J. VAN CLEAVE

Lühe¹ founded the genus *Filicollis* (1911:30) upon the characteristics of the species formerly known as *Echinorhynchus anatis* Schrank, of which he considered *E. filicollis*, Rud., *E. polymorphus* Bremser, and *E. laevis* vonLinst. as synonyms. In this species, *F. anatis* (Schr.), the female has a peculiarly modified proboscis at the end of a slender neck. Only a portion of the surface of the large inflated proboscis carries hooks. This characteristic was incorporated into Lühe's definition of the genus. Regarding the development of the proboscis in *Echinorhynchus filicollis* Rud., which as indicated above Lühe accepted as a synonym for *Folicollis anatis*, de Marval² (1905:267) has given a complete account of the changes in form accompanying advance in age of the female. His figures 103, 104, and 106 indicate in the development of the female a gradual change from an ovoid proboscis slightly larger than the neck in very young forms to the inflated, spherical form characteristic of the fully mature female. These three figures have been copied by the writer in the present article as figures 1, 2, and 3. In order to evaluate this point of structure fairly it should be borne in mind that these changes in the form of the proboscis occur after the individual has found lodging in the final host. To the writer it seems within the bounds of reason that such a characteristic as the shape of the proboscis appearing near the end of the development of the individual carries with it but slight phylogenetic significance. Consequently it could signify nothing more than a specific character and no longer should be considered as diagnostic for the genus.

The writer has found individuals belonging to a new species which agree in all essential details with the description of the genus *Filicollis* except in the lack of this inflated proboscis of the female. The creation of a new genus for such a minor variation would necessitate the separation of forms which are evidently closely related.

*Contributions from the Zoological Laboratory of the University of Illinois, No. 63.
¹Lühe, M. 1911. Die Süßwasserfauna Deutschlands, Heft 16, Acanthocephalen. Jena; 116 pp.
²Marval, L. de 1905. Monographie des acanthocéphales d'oiseaux. Rev. suisse de Zool., 13:195-387.

To avoid this confusion the description of the genus should be modified so that the species *F. botulus*, described later in this article, may be included within its limits. In view of the fact that this adds but one more species to the genus the writer does not feel justified in offering a complete emendation of Lühe's definition but offers as a suggestion that until more is known of members of this genus the original definition (Lühe 1911:30) should be qualified by adding the statement: "The proboscis of the female *may* be dilated, or in some species the proboscis of the female may assume the same form as that described for the males of the genus."

Filicollis botulus n. sp.

Description. Body both sexes cylindrical, large, thick, sausage shape; about 20 mm. long; about 4 mm. in diameter. Neck long, naked, retractable; 0.76 mm. long; 0.38 mm. diameter at posterior end. Proboscis sheath 2 mm. long. Body of male spined short distance back from neck, spines about 0.012 mm. long. Mature female with no distinct spines in this region, cuticula of anterior part of body in minute elevations. Proboscis ovoid, 0.65 mm. long, 0.57 mm. in diameter, armed with sixteen longitudinal rows of seven or eight hooks each. Hooks practically uniform in size, basal hooks 0.060 to 0.062 mm. long, apical hooks slender, basal hooks without distinct roots. Embryos elliptical, with three concentric membranes; 0.071 to 0.083 mm. long, 0.030 mm. broad. Type host *Somateria dresseri*, in intestine. Type locality Maine, U. S. A. Cotypes in collection of Bureau of Animal Industry, Washington, D. C., catalog number 2080; and in collection of the writer at Urbana, Illinois.

The material upon which this specific description is based was collected by Mr. Albert Hassall in Maine during the month of April, 1892. There are fifty specimens in the collection examined by the writer. In addition to this material the collections of the Bureau of Animal Industry contain several hundred specimens of this species from the type host and from *S. mollissima* [*S. m. borealis*?]

Acanthocephala of this general form from American ducks of various species frequently have been assigned by earlier workers to the species '*Echinorhynchus polymorphus*.' The confusion of

the species *E. polymorphus* Bremser, *E. anatis* Schrank, and *E. filicollis* Rudolphi of the early workers has been very general. After various investigators had attempted to solve the problem of the relationships of these species Lühe (1911; 27 and 30) has finally made a careful analysis of the characteristics of these species through which he was led to establish two independent genera, *Filicollis* and *Polymorphus*. The addition of another species to the genus *Filicollis* furnishes support to the contention for the validity of this genus, which is very clearly a natural group in the classification of the *Acanthocephala*.

EXPLANATION OF FIGURES

All original figures were drawn with the aid of a camera lucida. A projected scale indicating the magnification accompanies each drawing.

- Fig. 1. *Filicollis anatis*, fully mature female redrawn from deMarval 1905, fig. 103.
- Fig. 2. *F. anatis*, young form. Redrawn from deMarval 1905; fig. 104.
- Fig. 3. *F. anatis*, very young form. Redrawn from deMarval 1905; fig. 106.
- Fig. 4. *F. botulus* n. sp., male. Neck retracted. Body spines not shown. Arrow indicates posterior limit of spined region.
- Fig. 5. Cuticular body spines from anterior region of body of male shown in fig. 4.
- Fig. 6. *F. botulus* n. sp., female. Tip of proboscis slightly inturned.
- Fig. 7. *F. botulus*. Proboscis and part of neck of female.
- Fig. 8. Profile, ventral surface, of proboscis shown in fig. 7.
- Fig. 9. *F. botulus*. Embryos from body cavity of female.

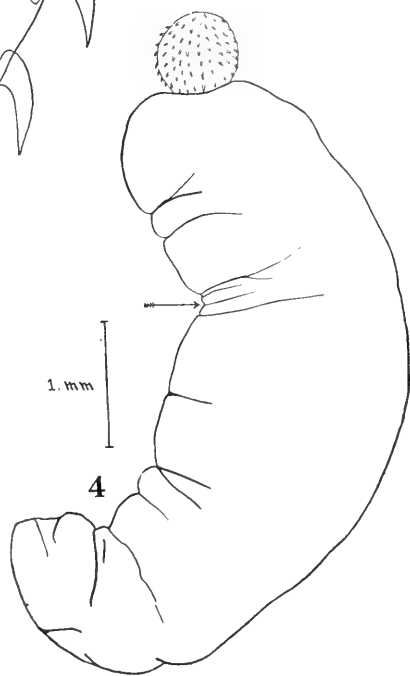
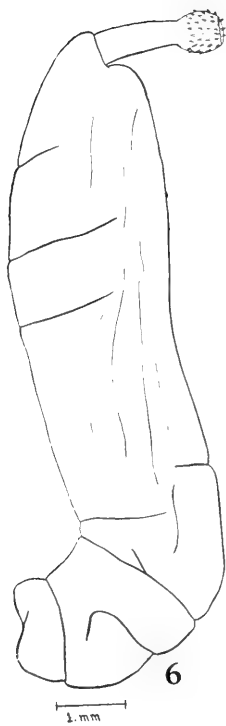
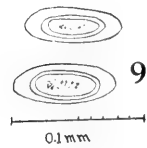
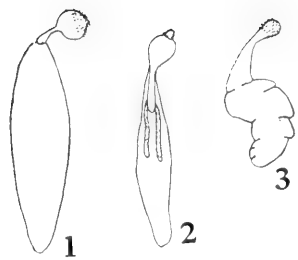
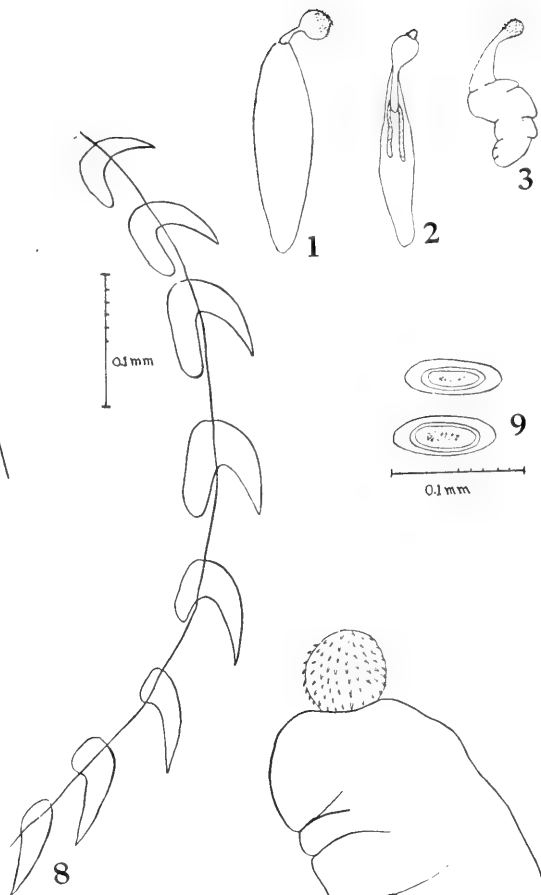
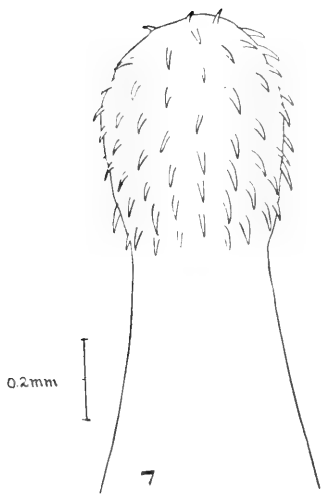


PLATE XVIII



DEPARTMENT OF NOTES. REVIEWS. ETC.

It is the purpose, in this department, to present from time to time brief original notes, both of methods of work and of results, by members of the Society. All members are invited to submit such items. In the absence of these there will be given a few brief abstracts of recent work of more general interest to students and teachers. There will be no attempt to make these abstracts exhaustive. They will illustrate progress without attempting to define it, and will thus give to the teacher current illustrations, and to the isolated suggestions of suitable fields of investigation.—[Editor.]

NOTES ON HANDLING PROTOZOA IN PURE LINE WORK

During the past year the writer has been engaged in experiments on the inheritance of extra contractile vacuoles in a new race of *Paramecium* and has worked out some methods of technique that have so facilitated his work that he is led to publish them in hope that they may be of benefit to others.

Maintaining pure cultures.—The greatest care is necessary to prevent pure line cultures from becoming mixed with others. Even with labelled pipettes accidents may occur. The scheme shown in

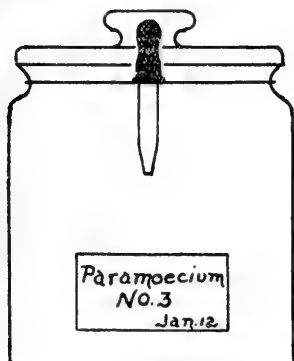


FIG. 1

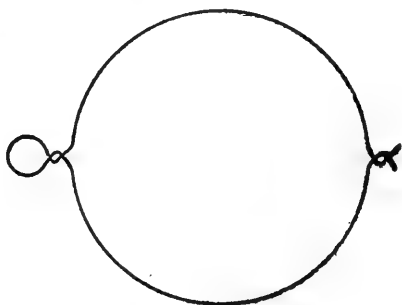


FIG. 2

the cut was recently devised and has proved most convenient. A piece of soft brass wire is shaped about some round object of a diameter slightly larger than a pipette and is held by several twists. Then the long ends of the wire are bent around the culture jar and again fastened by twisting the ends. In the jars used by the writer (humidors bought at any five and ten cent store) there is a convenient groove near the top into which the wire fits nicely. When finished the small circle protrudes from the jar and into

this ring the pipette is dropped giving the appearance of Fig. 1. With this method pipettes are always at hand and there is no danger of mixing the lines by transferring animals (clinging to the walls of a pipette) from one culture to another.

Preparation of watch glasses.—Syracuse watch glasses have been used for single individuals throughout the writer's work and considerable difficulty was experienced at first in locating animals which were close to the edge of the container. They frequently found their way there as the fluid had a tendency to spread evenly over the surface of the watch glass. Several methods were tried to correct this tendency of the culture medium to spread over the bottom but the best one was hit upon accidentally. There was a trace of paraffin in a pan in which the glasses were being sterilized one day and this coated the glasses imperceptibly but sufficiently to give the liquid no hold on the glass. In vessels treated in this way the surface tension of the medium tends to draw it into a spherical mass. Should the liquid roll to the edge of the glass where the animals would be hidden from view, it is easily rolled out again by tilting the glass and none of the animals in the drop are left behind. When animals are being kept in very small drops of water the writer has placed as many as twenty individual drops of liquid containing protozoa in a single watch glass and they have not run together. The surface tension of the liquid draws it up, when on a paraffined surface, until it gives a very fair picture of a drop of mercury. Furthermore, being contracted to the smallest area possible there is less evaporation than when the same amount of fluid is spread out and the chances of losing a valuable specimen through drying are very much less. The writer's practice is to use a piece of paraffin about the size of a pea to a quart of water. This will be sufficient for a surprising number of watch glasses. When the sterilized glasses are removed they are wiped while hot and polished. No paraffin is visible although a faint trace of it can be felt.

Making Pipettes.—In this work the most useful pipette has been found to be one that has a short but very fine tip. The usual methods of drawing them tend to make a pipette with a rather long tip as the glass tube has been heated for some distance

equally along its length. To cut down the area heated the tube to be drawn is placed transversely across a fish tail flame, which heats equally an area certainly not more than a quarter of an inch at found their way there as the fluid had a tendency to spread evenly the most, and the tube is pulled with considerable force when the glass is just commencing to melt. Several trials will show the best time to start pulling. This method gives pipettes with very fine tips not more than from three-quarters to one and one-half inches long.

*Zoological Laboratory,
University of Pennsylvania.*

ROBERT T. HANCE.

NOTES ON EMBEDDING IN PARAFFIN

When embedding very small objects, such as insect larvæ or small flowers or anthers, in paraffin it is most convenient to orient them one behind the other. This method allows a single block to be made of from three or four to a dozen pieces of tissue and these may be cut in one ribbon. This obviously eliminates a great deal of the labor involved in making a block of each separate object, cementing it to the holder, trimming it and adjusting the microtome each time. In the ribbon it is easy to see where one piece of tissue ends and the other begins as there is usually several blank sections of paraffin between them. It is relatively simple to arrange the tissue in line under a carbon bulb with warm needles but a difficulty is met with when an attempt is made to place the paraffin mold in water for cooling. The material is shaken from position and must be reoriented. This had been overcome in the following way. A watch glass is used as a mold for embedding small objects and a petri dish is convenient for larger tissue. When the tissue is ready to be embedded the dish is heated to the melting point of the paraffin under the electric bulb. It is then placed in a crystallization dish with two slides beneath it to prevent it from touching the bottom of the container. Paraffin is then poured into the small dish and the objects oriented as desired the heat of the electric bulb keeping the paraffin melted. Then the light is turned off and cold water is poured into the crystallization dish. Since the dish containing the paraffin is raised from the bottom the water flows under it and soon solidifies the paraffin in the lower part of

the dish which consequently holds the objects fast. As soon as a surface film is formed enough water can be added to cover the embedding mold to complete the hardening of the paraffin.

In petri dishes or watch glasses the bottom is practically flat and true and the tissue is allowed to sink to the bottom. When the tissue is cut out as a block the part that rested against the bottom makes one of the two parallel sides and requires little or no trimming.

When a number of pieces of tissue or a number of series (as described above) are embedded in one disk of paraffin it is dangerous to attempt to separate them with a knife as one can never be sure of the direction the crack in the paraffin will take. I have found that a hand scroll saw or coping saw (which may be purchased for ten to twenty-five cents) does admirably for cutting a block of tissue from the main disk. A hot wire is used by some but is not nearly so convenient nor so accurate as the saw. The use of the saw permits many more pieces to be placed in the same space as no care need be taken to have well defined pathways for the paraffin to split along as is necessary when a knife is used for separating the pieces.

ROBERT T. HANCE.

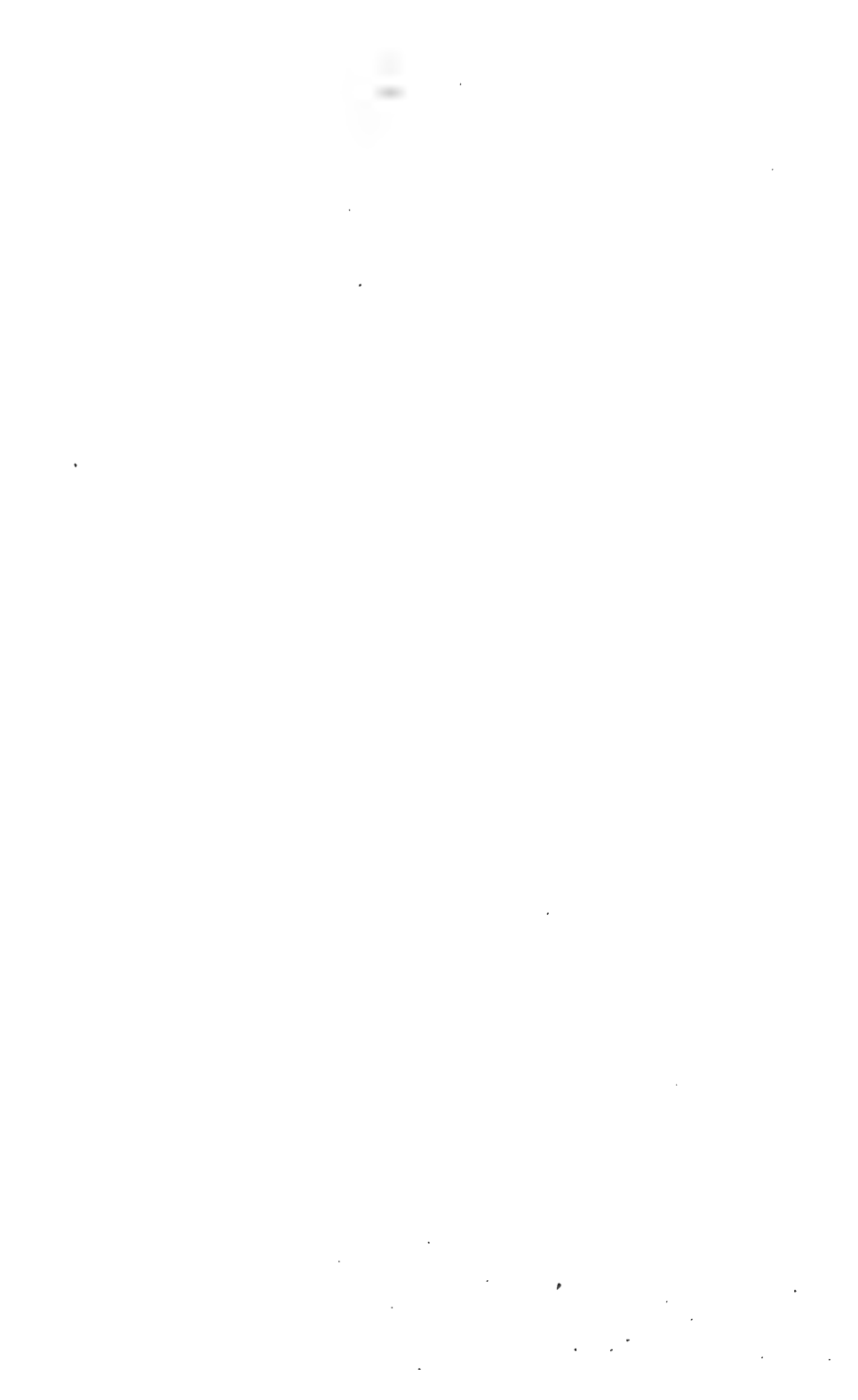
A NEW SPECIES OF OPERCULARIA

Opercularia wallgreni Grier n. sp.

Plate XIX. Figs. 1 and 2

Bodies ovate or attenuate fusiform, about 3 times as long as broad, tapering mostly toward the pedicle extremity. Ciliary disc never elevated above the margin of the peristome a greater distance than $\frac{1}{2}$ the length of the animal, apparently with but one circlet of cilia. Membranous collar moderately large, but obliquely set. Endoplast band-like, curved, parenchyma beneath granulated. Pedicle tree-like, slender, branching profusely and dichotomously, attaining a considerable proportionate altitude, delicately striate in a longitudinal direction. Transverse articulations wanting or present only where branching occurs.

Height entire polypidum 1.4 mm., length extended zooid .10 mm., width .022 mm., width of pedicle .005 mm.



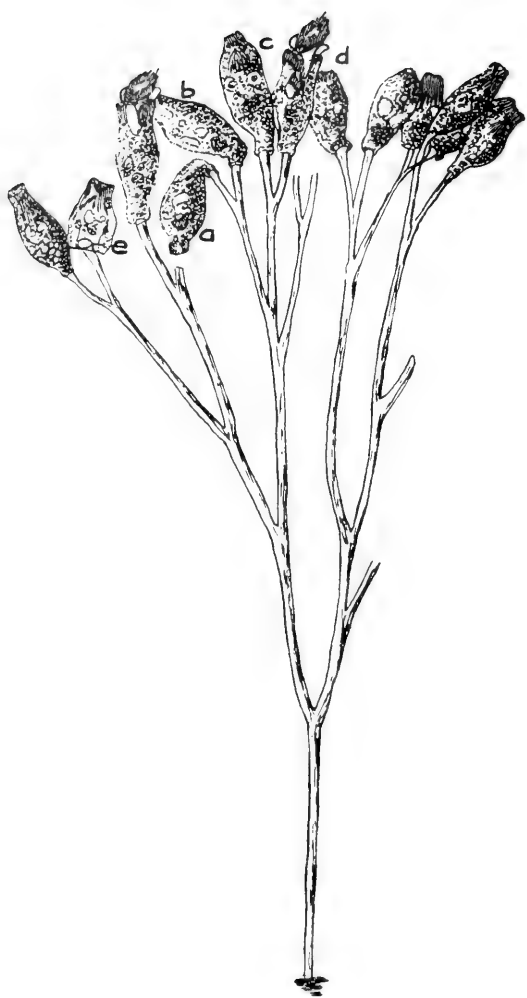


Fig 1

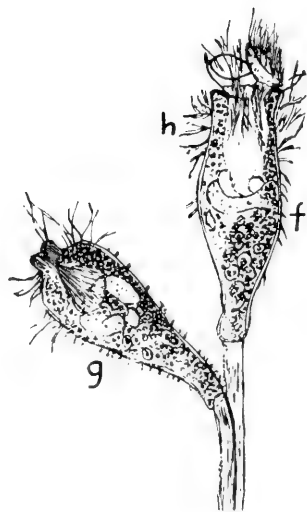


Fig. 2

Habitat.—Fresh water, apparently only upon aquatic plants. The colonies may include from 2 to about 200 zooids, which assume a nodding or pendant position after contracting. The species described was found in an aquarium in the writer's laboratory at the Central High School, first growing upon *Sagittaria platyphylla* Smith, but readily attaching itself to *Elodea*, *Myriophyllum*, and other aquatic plants. Its food consists for the most part of unicellular Algæ although Protozoa were sometimes observed in process of digestion. The species is very prolific, and while it does not grow in a hay infusion, quickly covers the walls of aquaria, growing thickest on the sides nearest the light. It is apparently of great longevity. This form I have respectfully dedicated to Dr. A. B. Wallgren, Professor of Zoology, University of Pittsburgh.

EXPLANATION OF FIGURES

PL. XIX.

Fig. 1. Entire colony x about 90.

a, b, c, d. Successive positions assumed by zooids during acquisition of food.

e. Curious introverted position noted.

Fig. 2. Two zooids x about 400.

f. Expanded, taking in food.

g. Contracted.

h. Attached bacterial growth.

Central High School,
St. Louis, Mo.

N. M. GRIER.

A METHOD OF MAKING TOTO MOUNTS OF UNICELLULAR FORMS

The matter of making toto mounts of unicellular forms often presents considerable difficulty. The cells or cœnobia settle so slowly that there is danger of losing them in the changes of liquid, and this slowness in settling makes the use of the more precise stains difficult. A method which has been successfully used for small forms like *Scenedesmus* is described in Chamberlain's "Methods in Plant Histology," University of Chicago Press, 1915. It consists of drying the cells down on the slide and then carrying them through all subsequent processes on the slide as in the case of paraffin sections. This method seems to cause some distortion

even in the smaller forms and a large form like *Closterium* is ruined. The following method, discovered in the botanical laboratories at the University of Nebraska, has been found to combine the good fixation and preservation of the bulk method with the precision of staining and the ease of handling secured by drying the cells to the slide.

The material is killed and fixed in whatever solution the investigator has found most satisfactory for the particular group of algae or Protozoa with which he is working. It is washed in bulk in the usual manner and carried through a graded series of alcohols until a strength of about sixty per cent. is reached. It is allowed to settle completely in this grade. A very thin layer of albumen fixative is smeared upon the thoroughly cleaned slides. A drop of the material is then drawn up with a pipette and placed upon the slide. The sixty per cent. alcohol in which it is lying coagulates the albumen and causes a surprisingly large number of cells to be firmly fixed to the slide. They may now be dipped directly into sixty per cent. alcohol and successively into higher grades. It is possible to use such stains as Flemming's triple and iron-alum hæmatoxilin rapidly and with precision. Before using a stain like Flemming's triple it is usually well to harden the cells thoroughly in ninety-five per cent alcohol, and then proceed as usual.

Univ. of Nebraska.

ROBERT A. NESBIT.

METHOD TO CLEAN USED MICROSCOPIC SLIDES

Especially where a course is given in microscopic technic there are usually a large number of worthless slides prepared. To throw them away seems an extravagance and yet to clean them in waste-xylol is practically a waste of time.

The method I am about to suggest may be well known, yet I think it will bear repeating. There had been a large number of old slides collecting from year to year in our department, worthless and merely occupying space, yet no one cared to assume the responsibility of throwing them away. Recently Professor Reese head of the department, suggested we try gold dust in an attempt to clean them.

A liberal amount of gold dust and a number of the slides, some of them dated 1902, were placed in water, and thoroughly boiled. As soon as the cover slips came off of their own accord, the slides and slips were placed in a pan of water. These were wiped dry while others were being boiled. The ease with which they can be cleaned and dried and the small amount of time required compared to the waste-xylol method, makes it a very profitable undertaking.

On taking them from the gold dust solution they were first placed in waste alcohol, but it was found by placing them in water they could be cleaned and dried much easier.

Zoological Laboratory,
West Virginia University.

J. THERON ILLICK.

ENTOMOLOGICAL NOTES

Chromosomes of Notonecta.—Browne ('16, Journ. Morph., 27:119-162) has made a comparative study of the chromosomes of five North American species of *Notonecta* (*undulata*, *irrorata*, *insulata*, *shooterii*, and *indica*) and one species of the same genus from Europe (*glauca*). Among other things, it was found that an XY pair of chromosomes is present in each of the above-mentioned species, the components of which divide separately in the first division and go to the opposite poles in the second. The X and Y chromosomes vary in size in the cells of the different species as well as in the cells of different individuals of the same species. They are almost equal in most of the cells of certain individuals of *shooterii*, while in *indica* they are distinctly unequal. *Undulata*, *indica*, and *shooterii* have 14 chromosomes in the first division, 13 in the second, and 26 in the diploid groups. *Irrorata* and *glauca* have 13 chromosomes in the first division, 12 in the second, and 24 in the diploid groups. *Insulata* has 14 or 13 chromosomes in the first division and 12 in the second. Large double chromosomes occur in *insulata*, *glauca*, and *indica*. No definite correlation of the somatic characters of the different species with the difference in chromosomes number and arrangement was discovered, although it was found that the 14-chromosome species are the smaller and the 13-chromosome ones are the larger. It thus appears that while,

in general, a definite number and arrangement of the chromosomes of each species exist, the status of each species in relation to the others cannot at present be determined on this basis. The chromosomes and somatic characters seem to indicate that *indica* has been derived from *undulata*.

Mitochondria.—Lewis and Robertson ('16, Biol. Bull., 30:99-124) studied, by the tissue culture method, the mitochondria and certain other structures in the male cells of the grasshopper *Chorthippus curtipennis*. By using a culture medium which very closely approached Locke's solution, it was found that, in addition to the fact that the minute structures of the living cells could be examined from day to day, these structures could be experimented upon as readily as those of the chick embryo. Any stage in the development of the germ cell was obtained by this method and was studied in the stained and unstained conditions, the staining being done with Janus green and neutral red. By these methods, mitochondria and neutral red granules were demonstrated in the primary spermatogonium. The former are present in the primary spermatogonium as small, delicate granules and increase in quantity during the growth stage, becoming definitely arranged along the spindle during the spermatocyte division. In the spermatid, they form the nebenkern, later developing "into two equal homogeneous threads in the tail of the spermatozoön."

Gynandromorphism.—Cockayne ('15, Journ. Genetics, 5:75-131), in a paper entitled "Gynandromorphism and Kindred Problems", presents a comprehensive discussion of gynandromorphism among animals, the greater part of the data being drawn from insects. Descriptions of a number of new examples are presented. The data included in the paper are not readily summarized and cannot be included here. A somewhat elaborate and suggestive classification of "hermaphrodites" is given. The theoretical explanations of gynandromorphism are taken up in some detail and considered critically. A number of text figures illustrate the condition of the internal reproductive organs in some of these anomalous insects. Four plates devoted exclusively to Lepidoptera illustrate certain forms of gynandromorphs. A bibliography of sixty-eight titles accompanies the paper.

Color Changes in Dynastes.—Andrews ('16, Journ. Exp. Zool., 20:435-456) reports results of studies on color changes in adults of the rhinoceros beetle, *Dynastes tityrus*. It was discovered that when live specimens of either sex which were light yellow with dark spots were confined for a time in receptacles with wet decayed wood, they all became very dark reddish with the spots scarcely visible. When removed from these conditions, they rapidly returned to the usual light coloration. Since such changes in an animal having no changeable pigment cells or blood vessels so distributed as to make color change possible seemed to be unrecorded, experiments were undertaken to determine the nature of this phenomenon. Variations in moisture were found to underlie the conditions which produced the color changes. These color differences can be explained without the assumption of internal nervous activity since, in general, the reactions of a dead, dried specimen were the same as those of the living beetle in all respects concerning the change of color from light to dark and the reverse. Experiments with light, heat, and moisture yielded results which made it evident that both living and dead beetles behaved alike in changing color under conditions which were interpreted as chiefly involving differences in the amount of water presented to the surfaces of the elytra and the thorax. Results of experiments pointed to the conclusion that any liquid which can enter the shell may cause it to change from light to dark color. Microscopical examination of the elytron showed that it is composed of an outermost layer of such a nature that it readily absorbs and gives off moisture. The absorption of liquids permits the color of the underlying part of the exoskeleton to appear as dark red, whereas when air replaces the moisture in this layer, it prevents the underlying color from showing through. The dark spots which do not change appear to be due to the presence of some material which so fills the pores or interstices that it acts as if it makes the area permanently wet or to some degree soaked with liquid, thus rendering visible the underlying color. The relation of these color changes to the life activities of the insects is not known.

Light Reactions of Vanessa antiopa.—Dolley ('16, Journ. Exp. Zool., 20:356-420) finds that *Vanessa antiopa* is invariably

positively phototactic and in direct sunlight comes to rest with the head away from the source of light. Specimens with one eye blackened and placed face foremost in a horizontal beam usually turn toward the functional eye, occasionally continuing to turn in this fashion, thus performing circus movements, but usually reversing the movement at the edge of the beam and moving toward the source of light. In non-directive light, the insects perform circus movements only, each turning toward the functional eye. Apparently, the stronger the light the larger are the circles described by the insect. Non-directive light of very low intensity does not deflect either way specimens with one functional eye but a further reduction of this light leads to the performance of circus movements toward the blinded eye. The behavior becomes modified by repeated trials, the modification being manifested in three ways: (1) decrease in number of circus movements; (2) decrease in angle of deflection; and (3) increase in promptness of orientation at edge of beam. With one eye blackened, this insect, when moving toward a source of light, can re-orient when the direction of the rays is changed and always in such a way as to turn toward the source of light. In darkness, blinded specimens move in circles toward the blinded eyes, showing that the covering acts as a stimulus. In light, blinded specimens circle in the opposite direction, showing that light received by the functional eyes is the stimulus. Evidence indicates that the reaction may depend upon the localization of photic changes within the eyes and that orientation is not wholly dependent upon the relative intensity of light on the functional and blinded eyes.

Origin of Wings.—Crampton ('16, Journ. N. Y. Ent. Soc., 24:1-39) presents a thoroughgoing summary and critical examination of the various theories relating to the origin of wings in insects. Special attention is given to a comparison of the evidences advanced in support of the *tracheal gill theory* and the *paranotal theory* of the origin of insect wings. The latter is favored and some original data are offered in its support. The wings of all insects are regarded as homologous and of common origin, thus being subject to the same principles regardless of the kind of metamorphosis. Tracheal gills and wings have been shown by em-

bryological studies to belong to different developmental series and are not homologous, facts which offer very strong counter-evidence against the derivation of wings from tracheal gills. Since the paranota (integumental outgrowths on the sides of the tergum) are homodynamous with the wings, the latter "were doubtless derived from them, since they occur in the most diverse forms". The only reliable evidence available at present is that of embryology, such evidence indicating that the wings are of tergal origin. It is also concluded that the paranota from which the wings are thought to have originally developed were entirely or in part expansions of the tergum. A bibliography of two hundred twenty-two titles accompanies the paper.

Wing Venation of Hymenoptera.—Rohwer and Gahan ('16, Proc. Ent. Soc. Wash., 18:20-72) have published a paper on the "Horismology of the Hymenopterous Wing" in which a modified form of the old Cresson system of wing venation terminology is proposed. The paper is valuable because of the extensive synonymy and comparisons of the systems used in the past, thus making much easier the translation of one system into another and the determination of equivalent names. The new system which is proposed is based on the opinion of the writers that "it is better for taxonomic work to designate a given area by a given name and call it that regardless of its possible homologies or analogies." The new system is constructed only for insects of the order Hymenoptera and is apparently a revival of the old custom of using different systems of terminology for venation in the different orders and of using purely arbitrary systems rather than those based on homology. The writers point out what they consider to be objections to the Comstock-Needham system but there is doubt that these objections are well taken. Unfortunately, such a system as the one proposed in this paper, in spite of its possible commendable points, will have the effect, if adopted at all extensively, of impeding the development of a common system, based upon homologies, for all of the orders of insects, a system much to be desired.

Parasites.—Timberlake ('16, Can. Ent., 48:89-91) finds that certain insects (beetles) may be parasitized by *Dinocampus americanus*, a common braconid parasite, without producing the death

of the host. It was observed that certain individuals of *Hippodamia convergens*, *Coccinella 9-notata*, and *Olla abdominalis* might be parasitized, the larva attain full growth and escape, and the recovery of the host be apparently complete. Experiments in which individuals of *Hippodamia convergens* and *Olla abdominalis* were exposed to the parasite showed that this particular parasite does not injure the vital organs of the host, although its fatty lymph tissues are often left in such a depleted condition that the beetle soon dies. The exit aperture of the parasites is itself sometimes fatal in effect. These experiments showed that successive parasitism may occur, resulting in the emergence of more than one generation of the parasite from the same host.

Insects and Fire Blight.—Stewart and Leonard ('16, *Phytopathology*, 6:152-158) report the results of experimental studies on the rôle of insects in the dissemination of fire blight bacteria. Experiments with certain Diptera (*Pollenia rudis* and *Sapromyza bispina*) seem to indicate they are not active agents in increasing the number of twig blight infections, since their method of feeding is such that it is doubtful if the blight bacteria which they may carry can gain entrance to the tissues. Possibly the bacteria carried by the flies may gain entrance through the punctures of other insects and produce a few infections. Flies are thought to be most important in those cases where they carry the causal organism to blossoms and occasionally to wounds, such as those produced by hail stones. The authors believe that all of the sucking bugs in nurseries are important in this connection. The numbers, methods of feeding, and seasonal distribution make the various species of different interest and importance in the transmission of blight bacteria. It is suspected that those which feed upon the tender tips of the twigs are of more importance than those which feed upon other parts of the foliage. *Lygus invitus*, *Heterocordylus malinus* and *Lygidea mendax* are of importance in spreading fruit blight in orchard trees.

Embryology of Honey-bee.—Nelson ('15, Princeton Univ. Press) has recently published a book on "The Embryology of the Honey Bee", which is the most comprehensive and thoroughgoing treatise that has yet appeared on this subject. The author has been

concerned not only with the details of the development of the honey-bee but he has employed the comparative method of treatment, drawing extensively upon the works of other investigators on the embryology of insects in general, thus making the book valuable in connection with any work in insect embryology. The large mass of data cannot be summarized here but certain special features deserve mention. The duration and rate of development were specially considered and it was found that the total time normally required for embryonic development is 76 hours. This period is divided into stages I-XV. Cleavage requires 14-16 hours; formation of blastoderm, 14-16 hours; formation of mesoderm, rudiments of mesenteron and embryonic envelope, 12-14 hours; remainder of development, including differentiation of tissues and organs, 32-34 hours. Discussion of the formation of the mesenteron is accompanied by an extensive comparison of the various opposing views and interpretations of other investigators on this subject. The different writers are classed according to the following theories of mesenteron origin: (1) derivation from yolk cells; (2) derivation from the lower layer (mesoderm, entomesoderm, primary entoderm); (3) derivation from proliferations of the blind inner ends of the stomodæal and proctodæal invaginations; (4) derivation, independent of the mesoderm, from two proliferating areas of the blastoderm, one at each end of the germ band, corresponding to the future location of the stomodæum and proctodæum, respectively; and (5) derivation from cells migrating inward from thickenings or islands of the blastoderm. Nelson holds that "the relation of the mesenteron rudiments in the honey bee may be interpreted in two ways, and the one chosen will probably depend largely on the theoretical bias of the interpreter. First, the mesenteron rudiments may be referred to the mesoderm Second, the mesenteron rudiments may be considered, as purely blastodermal in origin." A final decision between these two interpretations is not attempted. Segmentation is considered rather fully and is found to be as follows: head, six segments; thorax, three segments; and abdomen, twelve segments. Appendages were observed on the antennal, the three gnathal, and the three thoracic segments. There is evidence that in the honey-bee the tritocerebral (intercalary) segment should be

considered as "exaggerated ganglionic swellings" which probably do not represent appendages. Nelson has found nothing which could be "safely construed as abdominal appendages". Degenerating cells of unknown significance occur in the rudiments of the brain, particularly in the region between the second and third lobes of the protocerebrum. The tracheal system is formed from eleven pairs of invaginations of the lateral ectoderm, one pair, which seems to be observed for the first time, occurring on the second maxillary segment. This discovery is significant since "Now that a pair of tracheal sacs is known to exist in the second maxillary segment, the homology of the second pair of tentorial invaginations with the stigmata of the second maxillary segment is completely excluded, and the homology of similar invaginations with those of the trachea is decidedly problematical". The tentorium develops from two pairs of ectodermal invaginations, one in front of the bases of the mandibles and the other behind the bases of the first maxillæ. The œnocytes develop from the migration of cells from eight pairs of localized areas of the lateral ectoderm which occur on the first eight abdominal segments at the same level as the tracheal invaginations. The mesoderm differentiates laterally into a somatic and a splanchnic layer, the mid-ventral region remaining single layered. Separate cœlomic sacs are wanting. The single layered area develops into the rounded blood cells; the somatic layer forms the trunk muscles, the pericardial fat cells, and the dorsal diaphragm; and the splanchnic layer gives rise to the muscle layer of the mid-intestine and the two main divisions of the fat body. A mid-dorsal union of two rows of cardioblasts, derived from the two mesodermal layers, forms the heart. Two masses of mesodermal cells, one forming the anterior and one the posterior end of the mesoderm, produce the muscle layer of the stomodæum and the proctodæum respectively. One hundred fifty-seven titles are included in the bibliography at the end of the book.

Kansas State Agricultural College. PAUL S. WELCH.

NOTES ON OLIGOCHÆTA

Galvanic Response of Earthworm.—Moore and Kellogg ('16, Biol. Bull., 30:131-134) have tested the galvanic reaction of

Lumbricus terrestris to constant current. Orientation began as soon as the current was submitted and the body of the animal took the form of a U with the concave side toward the kathode. Writhing movements accompanied this orientation and as a rule the animal ultimately crawled to the kathode. Pieces of the body of the worm, 3-4 cm. long, subjected to the constant current, responded in the same way, except that progressive movements were absent. Reversal of the current produced a reversal of the response. The reactions are interpreted as due to the tension of the longitudinal muscles on the kathode side of the worm, resulting in a stronger contraction than that which occurs in the anodal region.

South Indian Oligochæta.—Stephenson ('15, Memoirs of the Indian Museum, 6:35-108) reports results of studies on *Oligochæta* of Ceylon and Southern India. The eversible pharynx of certain enchytræids was studied and a sensory function is suggested for this organ. The constant presence of setæ within the cœlom of *Fridericia carmicheali*, surrounded by masses of lymphocytes, is discussed and the tentative explanation is that they are formed, not as are the setæ of the body-wall, but as excretory products which take the form of rods or spicules, persisting in the body-cavity or disintegrating and being eliminated through the usual channels. The so-called iridescent sperm funnels of some earthworms have been found to be due, not to iridescence of the funnel itself, but to the spermatozoa which are so disposed that they form innumerable extremely fine threads lying parallel and which function as a "diffraction grating". New species of the genera *Enchytraeus*, *Fridericia*, *Drawida*, *Pontodrilus*, *Megascolides*, *Comarodrilus*, *Perionyx*, *Megascolex*, and *Erythræodrilus* are described.

Regeneration.—Hyman ('16, Journ. Exp. Zool., 20:99-163) has studied regeneration in a number of species of *Oligochæta* belonging to the families *Acolosomatidæ*, *Naididæ*, *Lumbriculidæ*, and *Tubificidæ*. By the use of Child's cyanide method, a gradient in the rate of metabolism of two forms, primary and secondary, was demonstrated. In the former, the rate of metabolism decreases posteriorly from the head and was found only in *Aeolosoma* and the zooids of *Naididæ*, while the latter is superimposed upon the primary gradient but runs in the reverse direction. The secondary

gradient involves the caudal third of the body in *Dero limosa*, the caudal half or more in *Lumbriculus inconstans*, and all but the first 5-15 somites in the *Tubificidæ*. Only the typical number of head somites are regenerated following amputation of varying numbers of anterior somites. The head regenerates a tail only when accompanied by a certain number of trunk somites and the tail regenerates a head only when it is of a certain minimum size whereby the gradient is eliminated. Under these conditions, regardless of length, a piece of *Dero limosa* regenerates a normal worm. If of appropriate length, any part of the body of *Lumbriculus inconstans* regenerates a normal worm, normal posterior regeneration occurring at any level. Formation of the head in *Tubifex* ceases at about the level of the fifteenth somite, while in *Limnodrilus* it ceases at the level of the seventh somite. The gradient of an axial series of pieces is not the same as that of the whole worm since temporary stimulation results from the cutting. Rate of metabolism is an important factor in anterior regeneration in short pieces since head formation will be inhibited in proportion to the metabolic rate of the old piece. If this rate be low, no inhibition of the new tissue occurs and a normal head is produced. Normal heads are always formed on long pieces since the dynamic factors are not important and the primary gradient determines the increased independence of cells at an anterior level over those of a more posterior level.

Kansas State Agricultural College.

PAUL S. WELCH.

NOTES ON THE COLLECTION AND REARING OF VOLVOX

Since the rearing of *Volvox* for laboratory use seems to be more or less unusual the writer has deemed it worth while to present here the results of some work done by a class of students and himself along this line.

Colonies of *Volvox aureus* Ehrenberg were found last fall in some abundance in small sphagnum pools on the west shore of a small glacial lake west of Ann Arbor known as the First Sister Lake. Collections were made on October 27, 1915. Small water-filled depressions in the sphagnum 10 to 30 inches in diameter were

sought out and into these depressions pint jars were thrust in such a way as to secure some bits of sphagnum and decaying vegetable matter together with water. The contents of the jars were sedimented and the liquid just above the sediment examined for colonies. When found the water at the top was poured off since the organisms were settling to the bottom and the remaining water and sediment from several jars poured together. Examination of many small pools on the shore of this lake showed that *Volvox* was not present in many of them. Some pools yielded *Pleodorina*. No colonies were found in the water of the lake itself.

Smith (1907) who has collected *Volvox* in the vicinity of Ann Arbor has found it in "small glacial pools containing *Riccia* and duckweed" but apparently had not found it among sphagnum. He (Smith, 1905) found it best in permanent pools but apparently never in great abundance. He collected it by "dipping it up together with a little of the water" and also "by sweeping a bolting cloth net over water plants, or better, using a 'Birge net'". Smith's method of dipping up the water together with plant material has been found by me to be the best method for collecting the organism here in Michigan. While in Nebraska the writer found *V. perglobator* Powers in such great abundance and in such clean open water that it was best collected with a small dipnet covered with bolting cloth or India linen. In this way large quantities of water could be passed through the net in a short time.

The collections made on October 27, 1915, were brought to the laboratory in the pint jars filled almost to the top with the water in which the organisms grew, together with about a half inch of the decaying vegetable material from the same source. Arrived at the laboratory these jars, still covered, were placed on the outside window ledge on the north side of the laboratory where no direct sunlight could strike them. They were kept here until November 29 when ice began to form on the surface of the cultures. At this time all jars except three belonging to the writer were brought inside the building. The writer's cultures remained outside for some time longer. While outside the window the number of colonies had increased greatly but when brought inside the numbers steadily diminished and soon disappeared. One student, Miss Rose Mayer,

placed her cultures in an unheated room, not in direct sunlight, where they continued to thrive for some time. An abstract from her report on her cultures is here given:

1915.

- Oct. 27. Volvox collected. (Other data as given above.)
- Oct. 28. Jars containing Volvox were placed outside of the window on the north side of the building.
- Nov. 3. Volvox was multiplying.
- Nov. 15. Still increasing in number.
- Nov. 29. Cold outdoors, some ice on surface of water. As many colonies as could be gathered were preserved. The remainder were placed in an unheated room but not in direct sunlight, i. e., the windows (south) were covered with gauze to diffuse the light.
- Dec. 13. A considerable increase had taken place. Another lot was removed and fixed. Remainder was returned to the cold room.
- Dec. 18. Colonies were quite numerous, but were beginning to fall to the bottom of the jar.
- Jan. 25. No colonies were observed. Since the last examination the temperature of the room had increased considerably.

When the cultures belonging to seven other students were brought inside they were so placed that they received some morning light. These cultures soon ran out and the writer has no further record of them. The writer's cultures (three pint jars) remained on the window ledge on the north side of the building until about December 5, but were protected at night with a cloth thrown over them. When brought inside they were placed on the window sill near the radiator. They received only north light. The colonies gradually disappeared but no record was kept of their last appearance. At various times the cultures were examined ocularly or with a hand lens but no colonies were seen until February 19 when a few colonies were seen in one jar only. They were noted again on February 26, and on March 7 the record states that they were in fair abundance. On March 16 the estimated number was 200 to 400 colonies and on March 27 several thousand were seen. At the time of writing, April 27, the water is green with them and they lie close together on the surface of the decaying vegetation.

During the fall and winter no colonies in sexual stages were seen. On March 25 a single zygote was found in the debris from the bottom of the culture. April 25 several colonies with ripe zygotes were found but they were not numerous.

Of the other two cultures one was destroyed by accident about the middle of March but no colonies had appeared in it at that time. The other culture was observed from time to time and in it 3 colonies were found March 20, about 20 on March 25, and about 50 on March 27. April 20 there were several thousand colonies present. These cultures are being kept in the hopes that further information may be gained in regard to the culture of this organism.

In conclusion it should be stated that since these cultures were not kept under controlled conditions it is possible that this success could not be repeated. However, certain points in the culture of this organism may be emphasized, viz., the water for the cultures should be from the same source as the organisms. Never use tap-water for making up the culture or for making good evaporation. Keep the cultures covered to prevent evaporation and consequent change in density of the medium and to exclude the dust and bacteria. The presence of organic material seems to be beneficial. Direct sunlight is unnecessary and is to be avoided because it causes too great variations in temperature in closed vessels. North light is good; in fact many algæ thrive in it as evidenced by the good growth of algæ in these cultures. Low temperature, above freezing, in early winter seemed to favor development. Old cultures should not be destroyed unless they have become hopelessly foul but they should be kept and the organisms given a chance to reappear.

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GEORGE R. LA RUE.

A NEW EMBEDDING STAGE

A new electrically heated embedding stage prepared according to designs prepared by laboratory men in this university has been recently put on the market by Eberbach & Co. of Ann Arbor. The essential parts of this embedding stage (see cut, Pl. XXI, Fig. 2) are a transite base $17\frac{3}{4}$ inches long by $4\frac{1}{2}$ inches wide mounted on three levelling screws, a copper stage made in two parts, 4 by 13 inches and 4 by 4 inches respectively, and under one end of the longer copper stage an electric heating unit. The heating unit may be wound for any voltage and to yield any desired temperature. Those in use in the Zoölogical Laboratory are designed for 110 v. alternating or direct current and the current requirement is 0.5 ampere. This yields a temperature of about 74° C. No regulator or rheostat or other provision for controlling or varying the temperature is provided but since the coil is situated under one end of the stage lower temperatures may be secured by moving the object away from coil. A scale to indicate the gradations of temperature could be attached if desired. In practice the coil is attached to a convenient electric receptacle near the paraffine bath and that part of the stage over the coil is heated sufficiently to melt paraffine in a few minutes. The embedding tray may now be warmed over the hot stage, filled with melted paraffine and moved to a point on the stage where the paraffine is kept just melted. Objects to be embedded are now transferred to the embedding tray, oriented, and the label inserted at the end of the tray with the legend towards the margin of the tray. Now the tray is gently moved to the unheated end of the stage where the paraffine is permitted to congeal on the bottom sufficiently to hold the objects in place. Then the tray is trans-

ferred to a dish of cold water or alcohol standing at the end of the embedding stage and into which it is immersed as soon as the paraffine is cooled sufficiently to prevent the breaking of the surface film by the water.

The use of this embedding stage makes unnecessary the use of the top of the paraffine bath for this purpose. Its use helps greatly in securing good embedding because it permits the paraffine to be melted clear to the bottom of the embedding tray and thus the orientation is made easy. The plan of allowing the object to lie on a layer of congealed paraffine is not only unnecessary but is faulty in that the paraffine is too soft to permit accurate orientation of the object and also because a cleavage plane is formed at which the paraffine frequently breaks during the sectioning. The stage is convenient to use, does away with the necessity of using gas, and largely obviates the danger of overheating the tissue which danger is always present when a gas flame is used for heating the ordinary stage. This stage because of its low construction is very stable, unlike the very insecure stage used with the gas flame, and with the levelling screws it may be levelled. In several months' use by a class no objectionable features have appeared and its good points are only the better appreciated.

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GEORGE R. LA RUE.

MAKING GLASS PLATES FOR COVERING MUSEUM JARS

At this time when it is impossible to secure from abroad the glass plates for covering museum jars it is worth while to know that after a little practise passable plates may be made in any laboratory equipped with power grinding and buffing machinery. Double strength glass plates may be purchased cut to size or they may be cut in the laboratory. Their edges may be rounded and a narrow ground surface at the margin may be secured by grinding on a carborundum wheel designated 120J-G5 which can be purchased from the Carborundum Co., Niagara Falls. The size of the wheel will depend somewhat on the power and speed of the grinder. In this laboratory a $4\frac{1}{2}$ by $\frac{1}{2}$ inch wheel belted to a $\frac{1}{2}$ h. p. motor

of about 1800 R. P. M. is used. While plates made in this way are not as good as the imported article they are usable and cheap, and by this means museum jars whose covers have been broken may be put into use.

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GEORGE R. LA RUE.

THE POSSIBLE NATURE OF THE "BOOK LUNGS" OF SPIDERS

The abdomen of spiders is now unsegmented, and yet it is probable that spiders have descended from ancestors whose bodies were segmented throughout.

The breathing apparatus in spiders is varied, some forms showing some development of tracheal tubes. On the forward end of the abdomen are found two sacs, each of which encloses a folded membrane which exposes the blood to the air. These are the book lungs.

In the section of such a lung from an *Aglena* (Plate XX, Fig. 1) the membranous character of the organ will be seen. Red blood cells may be seen between the double membranes. The outer surface of the membrane is covered with short spines, which prevent the moist membranes from adhering.

It is possible that this arrangement is derived from an ancestral form which had external gills at this point, somewhat similar to the tracheal gill membranes of insect nymphs.

A figure of a section of the young wing membranes of an Ephemera nymph is shown (Pl. XX, Fig. 2) for comparison. The similarity of structure is striking.

E. W. ROBERTS.

NOTE ON THE NATURE OF THE CYTO-PLASTID

The cyto-plasm of a cell contains unit plastids which themselves bear a great resemblance to a complete cell with its nucleus and cyto-plasm.

Using the Tussock Moth egg for an illustration we get a suggestion of this condition. The egg is filled with nutritive material supplied by numerous nurse cells from their own cyto-some system.

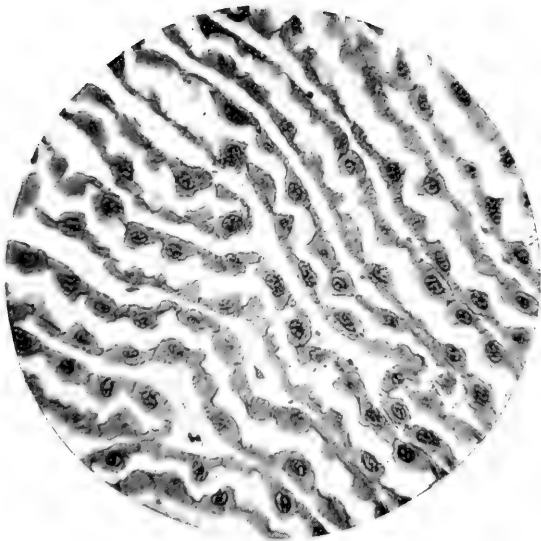


FIG. 1. Book-lung of Spider.

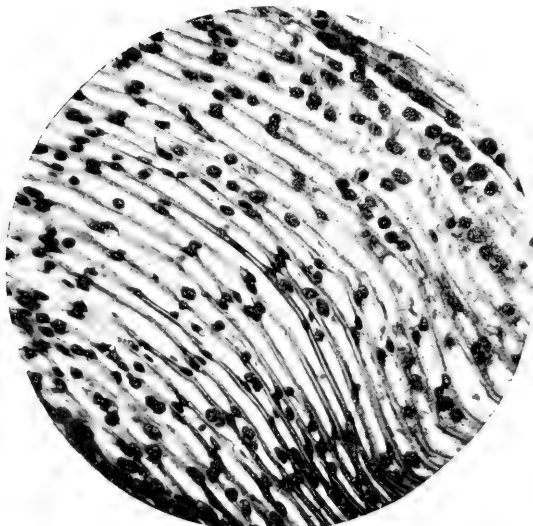


FIG. 2. Section of growing wing of Ephemerid Nymph.

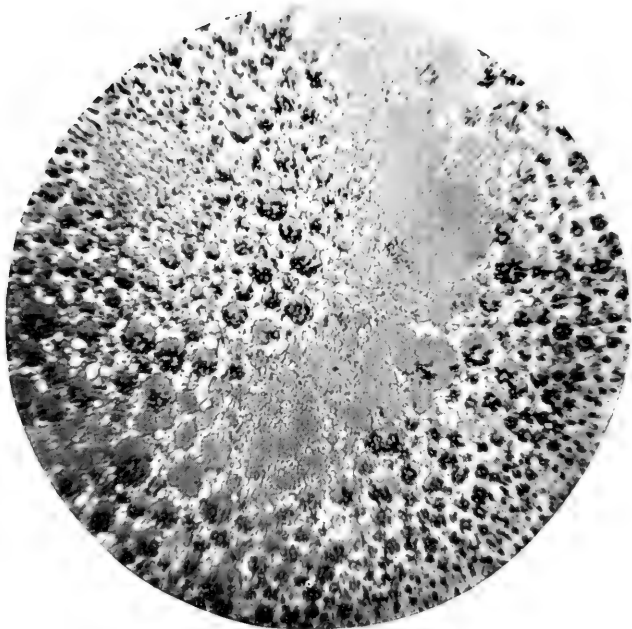


FIG. 1. Cyto-plastid in Tussock Moth Eggs.

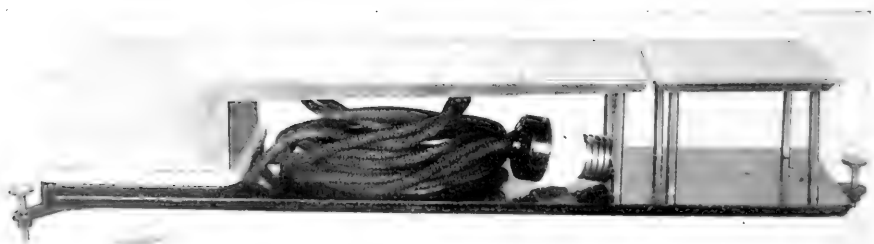


FIG. 2. Electrically heated embedding stage, Eberbach & Co.

Into this mass of raw food material, are extruded from the walls of the oviduct certain granules which stain as nucleo-somes. These appear singly at first as seen at the edges of the photograph (Plate XXI, Fig. 1). These chromatic bodies seem to multiply by binary divisions and thus produce a progeny of various numbers of staining bodies.

These bodies sometimes group themselves into paired threads, and in some cases separate into distinct groups resembling the anaphase of nuclear mitosis. The cyto-plastids thus seem to act as a miniature cell, containing grouped staining bodies, a surrounding body of plasm, and a definite membrane.

These efforts of the staining bodies to divide and to group themselves gives one the impression that the process is a modified or incomplete sequence of what we see in mitotic divisions of the chromosomes, from which they may be derived.

In the lower animals such as Rhizopods and others, the nucleus buds out into the cytoplasm similar bodies, which undergo analogous binary divisions and finally form a large progeny of staining bodies. Later these group and form new nuclei which surround themselves with a plasmic body, about which a new cell wall is formed. In the Tussock egg these processes seem to stop short of this final result, and produce objects whose fate is to be used as food.

E. W. ROBERTS.

SENESCENCE AND REJUVENESCENCE

The fact that an organism at its individual beginning is "young" and gradually loses some of the powers of youth with the passage of time, is so commonplace that its real significance escapes us. The further fact that one of these older organisms can, in spite of its age, produce offspring which apparently do not have the age of the parent, but are as young as the parent originally was at the beginning, brings us acutely upon the problems at the basis of this book. In what does growing old consist? How does age differ from youth? When reproduction takes place, is youth absolutely restored? If so, do the old materials become young again, or is there some material within the aging body which does not itself grow old?

Many investigators believe that youth is the eternal possession of the germ-plasm, and from this the cells of the body form,—these latter alone becoming aged. There is in this view only one process,—senescence. Rejuvenescence is apparent only.

Professor Child, thru the study of the lower forms particularly, in which differentiation is not so pronounced and non-sexual reproduction and regeneration are more frequent, comes to the conclusion that rejuvenescence is not confined to the debatable case of the germ cells, but is found as characteristic a feature of life as is senescence. He finds it also in agamic reproduction, regeneration of lost parts, in the restoration of starved tissues to activity; and the like. The author believes that senescence is not continuous therefore; that it is interrupted now and again by processes essentially restorative of youth; that there is no eternally youthful stuff, but that rejuvenescence and dedifferentiation are just as really a part of the life-cycle as is growing old.

As a criterion of youth one may take as his standard the normal changes which are observed in the embryo formed by the union of gametes,—e. g. rapid growth, cell division, differentiation and other vital processes. Senescence would imply on the other hand a decrease in these dynamic processes. Professor Child undertakes to erect a more exact standard based upon the principle that youth and age may be measured by the rate of metabolism in the organism. As a test of the rate of metabolism, and hence of the youthful or senile tendencies of tissues, the author holds that their susceptibility to certain chemical substances is satisfactory. In solutions strong enough to kill in a short time, the more actively metabolic (younger) objects are killed first; whereas in weaker solution to which acclimation is possible, those more metabolic live longer.

The captions of the five principal divisions or parts will sufficiently indicate the order and scope of the work, which is in large part based upon the author's original researches here given to the world for the first time:—I. The Problem of Organic Constitution; II. An Experimental Study of Physiological Senescence and Rejuvenescence in the Lower Animals; III. Individuation and Reproduction in Relation to the Age Cycle; IV. Gametic Reproduc-

tion in Relation to the Age Cycle; V. Theoretical and Critical. There are seventeen chapters in all.

The book is a worthy illustration of a monographic treatment of a subject instead of in numerous detached papers. The biological workers will be grateful.

Senescence and Rejuvenescence, by Charles Manning Child. Illustrated, 480 pages. University of Chicago Press.

A TEXT BOOK OF HISTOLOGY

Messers Jordan and Ferguson, in this book, have tried to give to students and teachers a treatment of the relatively stable matter of histology which will overcome, thru interest, the difficulties which the average student has in approaching and mastering the subject. There is no doubt that histology may be, and often is, so presented as to be deadening and full of drudgery. Indeed in the opinion of the reviewer this is an indictment that will stand against very much of the work done in College and University laboratories in America today, not alone in histology but in all aspects of morphological work. The writers justly conceive that such interest may be stimulated and held by relating the facts of structure to their practical ends in terms of function; and to their meaning in terms of the generalizations which give vitality and zest to investigation. The student of any science is entitled to the pleasure that comes naturally from following his discoveries on thru into conclusions that relate his facts into a system. Whenever it has been necessary, in order to accomplish this, the authors have not hesitated to introduce the facts of development and of function, and the theoretical explanation which will enable the student to appreciate the facts. This is illustrated, for example, by the discussion of the neuron theory under nervous tissues, and the theories of the inversion of the vertebrate retina in the discussion of the eye. In the opinion of the reviewer they have had good success in accomplishing their announced purpose.

The order of discussion will be made clear by giving the chapter headings. An introductory chapter deals with protoplasm and the cell. Then follow chapters on Epithelial Tissues; Connective Tissues; Muscular Tissue; Nervous Tissues; End Organs; Blood Vessels; Blood; Lymphatic System; Mucous Membranes and

Glands; Skin; Respiratory System; Digestive System; Urinary System; Reproductive System; Ductless Glands; Nervous System; Eye; Ear. A chapter of 40 pages on Histological Technic concludes the book.

The illustrations are well chosen, excellently executed and liberal. The text is well written and clear. Mechanically the book is a beautiful one.

A Text Book of Histology, by H. E. Jordan and J. S. Ferguson. 800 pages, 594 illustrations. D. Appleton & Co., New York.

MEDICAL AND VETERINARY ENTOMOLOGY

It has long been recognized that the richest ore is often found where two veins intersect. Under the title *Medical and Veterinary Entomology*, Professor Herms treats the territory common to *Economic Entomology* and *Parasitology*. This treatment, in the hand of one who has himself made noteworthy contributions to both fields, in both a scientific and a practical way, assures a book helpful to those immediately interested in the subjects and illuminating to the general zoologist.

The author expresses his purpose as being to systematize the subject and thus assist in securing for *Medical Entomology* a place among the applied biological sciences, rather than to supply a comprehensive treatise. He indicates that he hopes to be of special service to the physician, the veterinarian, the health officer and sanitarian, as well as to the teacher and student.

Biologically the field includes the morphology and ecology of insects (and of the ticks and mites which are embraced in the treatment); their relations to the host when they are the direct causes of the diseases; the biology of the bacteria and protozoa by which the diseases are caused when the insects are merely the carriers of the germs; the physiology and pathology of the animals attacked. While in itself a specialization, it thus rapidly broadens out into the general interests of biologists, theoretical and practical, when its connotations are realized.

The content and mode of treatment are apparent from an examination of the chapter headings:—Introduction; Parasites and parasitism; Insect anatomy and classification; Insect mouth parts;

How insects carry and cause disease; Cockroaches, beetles, thrips; Lice; Bedbugs; Mosquitoes as disease bearers, and their control; Buffalo gnats and horseflies; Common house fly; House fly control; Bloodsucking Muscids; Myiasis; Fleas; Ticks; Mites. An interesting final chapter is given to the discussion of venomous insects and arachnids,—as bees, wasps, spiders, scorpions, etc. The nature of the venom, the manner of its introduction, and its effects are treated.

The book is well illustrated with half-tones, has numerous keys for the identification of the principal genera, and gives the best accepted treatments for control of the insects and of the resultant diseases.

Medical and Veterinary Entomology, by William B. Herms. 394 pages, illustrated. The Macmillan Co., New York, 1915. Price, \$4.00.

CLASSIFICATION OF LEPIDOPTEROUS LARVÆ

Number one of Volume two of the Illinois Biological Monographs bears the above title, and is the thesis of the author, S. B. Fracker, offered in the Graduate School of the University of Illinois toward the degree of Doctor of Philosophy in Entomology.

The paper is divided into two parts; the first being devoted to the question of the homology of the setæ of the larvæ of Lepidoptera, and the second to the systematic outline of the families and genera.

The position taken by the author, that a final classification of the insects based upon both the larval and adult characters necessarily eliminates errors that belong to a classification based solely on either, must commend itself to the general student. It is surely true also that anything which will make identification of insects more possible in the larval stages will save much trouble and time required to rear them to maturity for more certain identification.

The proffered classification is based largely upon the setæ and the armature developed in connection with them, the head parts, the size and shape of the spiracles, the prolegs and the hooks they bear, and other structures somewhat less certain.

The portion of the paper that will prove most suggestive to the general biologist is the discussion of the setæ, the method of homologizing them, and the evidence for their sufficient identity for comparative purposes.

The first step is to get a standard or type segment so far as setal arrangement is concerned. This is done by taking the various thoracic and abdominal segments in the more generalized members of the two sub-orders, and plotting from these the setal arrangement by superimposing them so as to get a composite. This composite gave about 15 setæ with approximately the distribution found on the pro-thorax of the most generalized and primitive types. With this composite all the different segments of every larva were compared. In a similar way a type of abdominal segment is worked out and used as a standard.

The author concludes that the first-stage larva, before entering upon the various moults, best represents the ancestral type of Lepidoptera and that the setal arrangement of the first instar is essentially the same as in the ancestors, and thus serves as a connecting link between the more generalized type and the modern, specialized older stages.

The various tests of homology of setæ used by the author are:— (1) similar grouping of setæ in mature caterpillars generally; (2) similar position of setæ on certain segments of modern mature caterpillars; (3) similar arrangement of setæ on all the segments of generalized groups of caterpillars; (4) similar arrangements on all the segments of newly hatched larvæ; and (5) evidences of migration from these similar positions.

The author believes that the setal arrangement of every segment of the body of the larvæ of Lepidoptera has been derived from the same ancestral type; that 12 such primary setæ can be homologized; that these primary setæ are present in the first instar; that they may be modified by loss chiefly (abdominal segments) or by loss and change of position. Sub-primary setæ appear in later instars and may become associated with the primary setæ in ways more or less confusing. They may develop in tufts of various kinds.

The systematic part is based upon maturer larvæ, but the characters apply for the most part to the earlier also. Elaborate keys of the order and of the families, leading to the genera, are supplied.

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NOTICE TO MEMBERS:

Business Sessions of the *American Microscopical Society* will be held at New York City in connection with the American Association for the Advancement of Science. See Program of A. A. A. S. for time and place.

T. W. GALLOWAY, Secretary.

Beloit, Wisconsin.

TRANSACTIONS
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APRIL, 1916

No 2

THE INNERVATION OF THE AMPULLÆ OF LORENZINI
IN ACANTHIAS VULGARIS

By HERBERT EDMOND METCALF

Teaching Fellow in Animal Biology, University of Minnesota

INTRODUCTION

In a former paper (1914) the author gave a general description of the ampullæ of Lorenzini in *Acanthias vulgaris*, presenting evidence as to the sensory nature of these organs. Since that time further work has been done on their innervation and is embodied in this paper.

Peabody (1897) published a short article on the innervation of these structures as shown by methylin blue preparations, and the present author's findings as published in the 1914 paper closely agreed with his in the main, altho there was some doubt expressed at that time as to the nodules described being the actual termination of the nerve fiber. Later findings indicate that a complete impregnation was probably not present either in Peabody's preparations, or in the author's in 1914. Later methods have demonstrated the final terminations of the nerves supplying the ampullæ.

METHODS

New material impregnated by the methylin blue method was sectioned, and several preparations were made by the Bielschowsky double silver and gold process. The results therefore, were checked

up between those two methods, as well as by maceration and isolation of cells.

It was considered probable that if the nerves which supply the ampullæ terminate on particular cells in the ampullary pockets, isolation by means of maceration and teasing would be likely to show isolated sensory cells with their nerve terminations attached. Material was macerated in 3% caustic potash for 24 hours, teased immediately in glycerine and mounted in glycerine jelly; or carried through the alcohols, teased in balsam and mounted. In this way thousands of isolated sensory cells were obtained as well as isolated supporting cells. This proved to be a very valuable method of studying the general structure of the epithelium as well as for the sensory cells with their nerve terminations.

THE GROSS INNERVATION OF AN AMPULLA

There are eight definite groups of ampullæ in *Acanthias*; two on the dorsal surface, four on the ventral, and two on the lateral surfaces of the snout and head. As might be expected these groups are bilaterally symmetrical. The number in each group, however, varies somewhat and each ampulla has not a fixed position on each side. The position of four of these groups may be seen in Figure 2 (dorsal Fig. 2-A, ventral Fig. 2-C.) The two ventral groups have not yet been divided into four by the cartilage bars of the rostrum.

There are two kinds of ampullæ in *Acanthias*. Those with a single duct extending from the alveolar portion to the surface, and those with a split duct. As was explained in the previous paper these two types are essentially alike, only in the ampullæ which have a double duct there are no primary ducts, the division between the two secondary tubules being carried completely to the surface.

The model in Figure 1 (Pl. XXII) was made from sections 5 microns thick by the wax plate method and shows approximately the size and shape of the ampullæ having a double duct. In this case an ampulla having a short duct leading to the surface was selected, in order that the entire course might be modelled. This particular ampulla had alveoli and does not follow an equal division

into secondary and tertiary ducts. There are several alveoli which come off the main or secondary ducts. (Fig. 1-D.)

The external opening is nearly round and divided by the partition between the two secondary ducts. Immediately below the skin the duct widens out to about twice the size of the opening, and proceeds downwards to the alveolar portion. Here there is a sharp constriction to about seven eighths its size and then the alveoli are budded off at different levels. This fact is important for it can be readily seen that the number of alveoli cannot be counted from a single section, as they will not all be included. The average number of alveoli in the ampullæ of *Acanthias* is twenty-two. The model shows the general appearance and proportion faithfully with the exception that there is some slight shrinkage so that the alveoli in the fresh total mount are somewhat more rounded and not so long in proportion to their width. This is probably due to the coagulation of the mucus in the lumen of the ampulla due to the fixing fluid.

The ampullæ in the two groups on the dorsal surface of the snout are innervated by the ophthalmicus superficialis; those of the ventral groups by the buccalis; and those of the spiracular region by the mandibularis externus branches of the seventh or facial nerve. These branches also innervate the lateral line system in those regions. The ophthalmicus superficialis may be seen under the dorsal group in Figure 2-B, and the buccalis directly over the ventral group in Figure 2-D.

From the main nerve trunk as it passes along close to the alveolar portions of the ampullæ, there is given off a single twig to each ampulla containing from 5 to 15 medullated fibers. The number of medullated fibers does not correspond at all to the number of alveoli, altho in cases where there are a large number of alveoli there is a large number of fibers. Roughly speaking there are about twice as many alveoli as there are fibers supplying the ampulla. These twigs run to the ampulla, and as the alveoli are arranged in a circle, there is a central space into which the nerve twig runs. (Figs. 5 and 6.) This twig does not give off branches until it reaches a level just below where the duct begins and where the ampullary pockets are budded off. At this point the nerves bend laterally (Fig. 5-b) with respect to the ampulla,

and radiate out between the alveoli close to where the alveoli join the duct. (Fig. 6.)

This radiation takes place practically at the same plane for all of the nerve fibers, and all turn at right angles to their first course in 15 to 20 microns.

While doing this, the nerve fibers, which have up to this time been medullated, lose their sheaths, and when they appear on the external surface of the ampulla, between the alveoli, they are entirely devoid of covering. (Fig. 3.) Thus it may be seen that the course of the neurofibrils over the ampullary pockets themselves is in the majority of cases, from the bases of the alveoli toward their distal ends, speaking in respect to the orientation as shown in the model. (Fig. 1.)

As the neurofibril reaches the external surface of the ampulla, it divides into a great number of neurofibrillæ which anastomose over the entire surface of the pockets making a network over that portion of the ampulla which is sensory in character. (Fig. 4.) Whenever two fibrillæ branch a spot or nodule is formed. Nodules are also often seen along the course of the fibrils, as well as where a fibril apparently ends. (These nodules are labelled *b* in Figures 9 to 14.) These were taken by Peabody (97) and by the author (1914) to be the endings of the neurofibrils upon the bases of the sensory cells. This, however, I have now seen is not the case.

NERVE TERMINATIONS

The sensory cells in the epithelium are well seen both in sections and in isolated specimens, and are the large pear-shaped cells with broad bases and a short projection which extends out to the lumen of the ampulla. Each of these cells is surrounded by four or five supporting cells which fit over the top as shown in Figure 9. In macerated preparation the pear-shaped cell comes away from the supporting cells and leaves the small hole where its projection reached the lumen. (Figs. 7-a and 8-a.) The supporting cells in macerated preparations give a table-like appearance as shown in Figure 8, and they occasionally stick together as in Figure 7. Figure 9 is a drawing of a sensory cell complete with its four supporting cells and its nerve termination. All of these cells are more

or less rounded due to the fact that they swell somewhat in the maceration process. This gives a more or less rounded appearance to their bases which appear flat in sections. Figure 10 is a drawing of an isolated cell which was cleared and drawn in optical section so as to show the small projection from its upper surface to the lumen.

The neurofibril, which has been traced to the small nodule on the base of the cell, does not stop there but has a small process extending up between the sensory cell and the interstitial cell. (Labelled *c* Figs. 9-14.) This process does not readily impregnate with methylin blue but can be seen occasionally by very careful focussing with the oil immersion lens in cases where it does occur. It has been seen a sufficient number of times to justify the statement that in all probability all of the pear-shaped cells have such a termination. This process which was entirely overlooked in the previous paper, is the final termination of the nerve. This process arises from the nodule on the base of the cell and is very slender. It extends to the level of the lower border of the nucleus and there flares out with a fork and encloses an area which in my preparations appeared lighter than the rest of the nerve. It must be understood that this oval area is NOT on the inside of the cell, but closely applied to its external surface. This form of ending is much like the terminations of the sensory fibers in the ear of *Mustelus* as described by Morrill (97), and may be taken as evidence of the homology of these ampullæ with those of the ear.

In isolated sensory cells impregnated by the methylin blue method the cell may sometimes be seen with this process connected with the basal nodule as well as with part of the torn neurofibril. (Figs. 11 and 14.) Morrill (97) in his figures shows many cells which have no hairs, and yet were sensory in character. These cells in the ampullary pockets look somewhat like hair cells with the exception that the process is rather thick and does not project out into the lumen. In every case, however, it connected with the lumen. This is not as readily seen in sections as it is in isolated specimens, because in sections these sensory cells are so large that it is only occasionally that a section cuts the process exactly to show it extending to the lumen. In thick sections, by focussing, it may

be recognized. It may thus be seen that the real termination of the nerve supplying the ampullæ of Lorenzini in *Acanthias* is a slender process extending from the basal nodule to a point about half way up on the cell, and there flaring out into an oval plate in close contact with the sensory cell.

SUMMARY

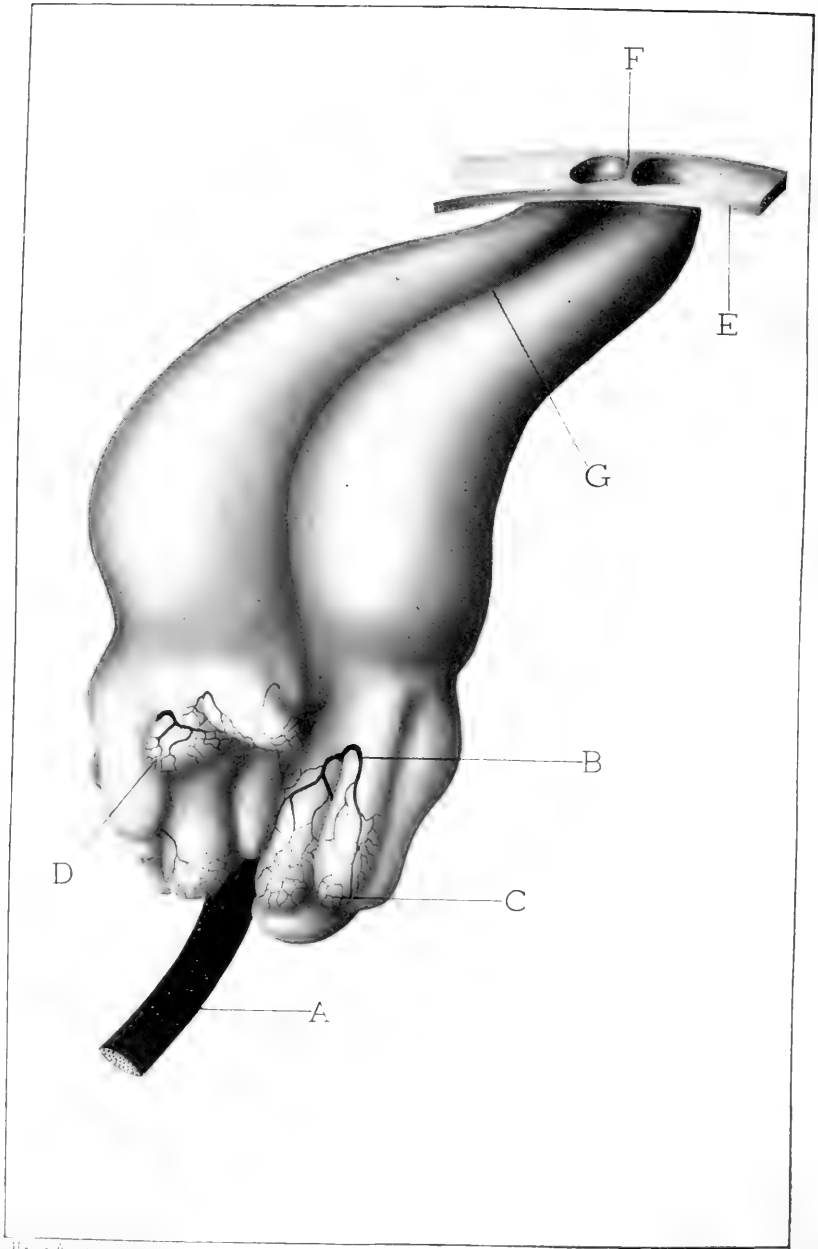
1. Each ampulla is innervated by 5 to 15 medullated fibers from the seventh or facial nerve.
2. These fibers run into the central space of the ampulla, turn sharply and radiate laterally to reach the external surface of the ampulla.
3. From there they radiate downwards forming a network with nodules on the bases of the sensory cells.
4. The termination in *Acanthias vulgaris* is a light oval plate closely applied to the sensory cell at about its middle and connected with the basal nodule by a slender strand.

I wish to thank Dr. J. B. Johnston for his kind help during the course of this investigation.

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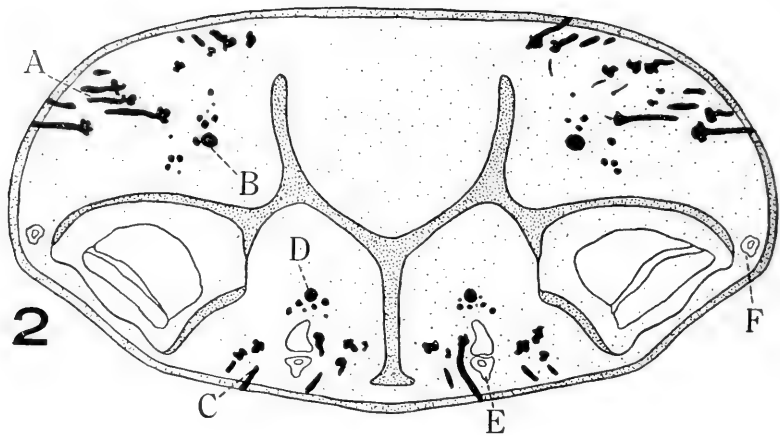
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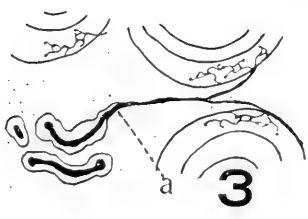


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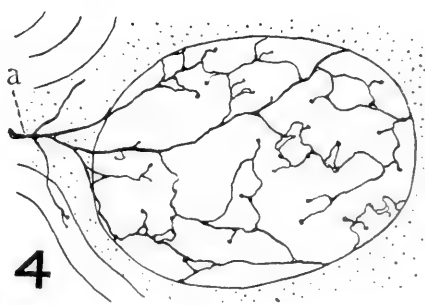




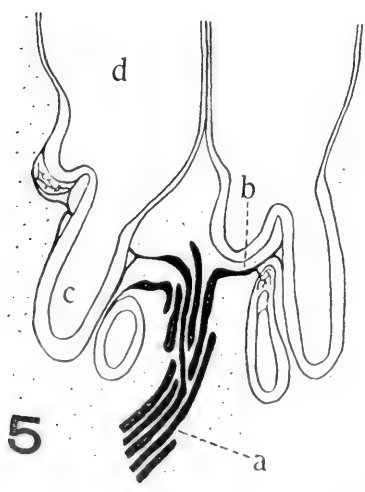
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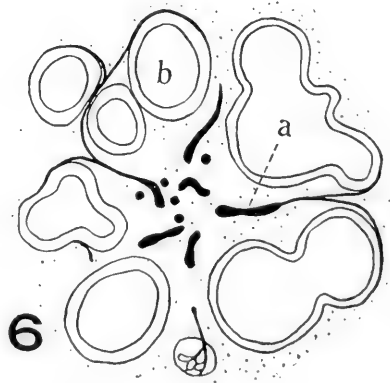
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DESCRIPTION OF PLATES

PLATE XXII

- Fig. 1. Drawing of a wax model of an ampulla having a double duct. A—nerve bundle entering central space. B—ramification of unmedullated fiber over alveolus. C—anastomosis of neurofibrils over distal end of alveolus. D—alveolus coming directly off secondary duct. E—skin of fish. F—partition between the two secondary ducts and openings.

PLATE XXIII

- Fig. 2. A transverse section of an *Acanthias* pup just forward of the nasal capsules. A—Dorsal group of ampullæ. B—Ophthalmicus superficialis. C—Ventral group of ampullæ. D—Buccalis nerve. E—Lateral line system. F—Lateral line system. X 15
- Fig. 3. Drawing showing the loss of the medullary sheath as the nerve bends laterally. Bielschowski method. a—point where loss of sheath occurs. X 180
- Fig. 4. Showing the anastomosis of the neurofibrils over the ampullary pocket. Methylin blue impregnation. a—point where nerve leaves sheath. X 250
- Fig. 5. Diagram to show nerve entrance and lateral radiation. Longitudinal section, Bielschowski method. a—nerve twig. b—nerve radiating laterally. c—ampullary pocket. d—duct. X 180
- Fig. 6. Transverse section showing nerves radiating laterally. Bielschowski method. a—nerve radiating laterally. b—ampullary pocket. X 180.

PLATE XXIV

- Fig. 7. Group of supporting cells from which three sensory cells have been removed by maceration. a—opening by which the sensory cell connected with the lumen.
- Fig. 8. Group of supporting cells from which one sensory cell has been removed by maceration. a—opening by which the sensory cell connected with the lumen.
- Fig. 9. A group of supporting cells enclosing a sensory cell with its nerve termination. a—projection of the sensory cell to the lumen. b—basal nodule. c—nerve termination.
- Fig. 10. Optical section of an isolated sensory cell with three supporting cells, and nerve termination. a—process extending to lumen. b—basal nodule. c—nerve termination.
- Fig. 11. A sensory cell isolated by maceration to show process and nerve termination. a—process extending to lumen. b—basal nodules. c—nerve termination.
- Fig. 12. Section directly through the process extending to the lumen showing nerve termination on side of cell, and two supporting cells. Methylin blue impregnation. a—process. b—basal nodule. c—nerve termination.
- Fig. 13. Section directly through the process extending to the lumen showing nerve termination on middle of cell, and two supporting cells. Lettering as in Figure 12.
- Fig. 14. A sensory cell isolated by maceration to show process and nerve termination. Lettering as in Fig. 11.

All figures in Plate XXIV were drawn with the oil immersion lens and have a magnification in the plate of 2000 diameters.

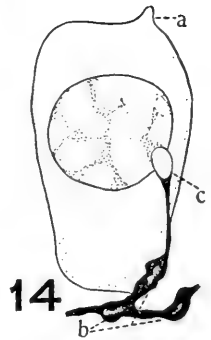
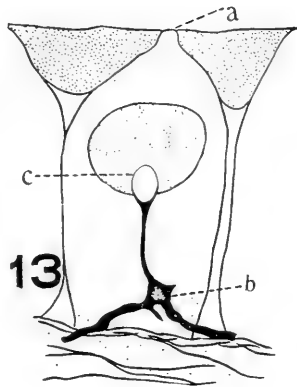
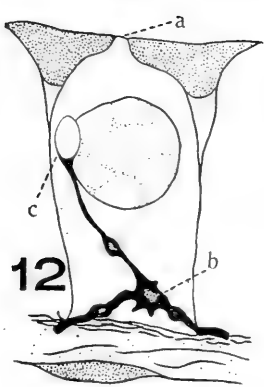
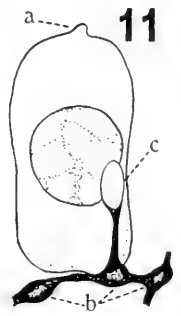
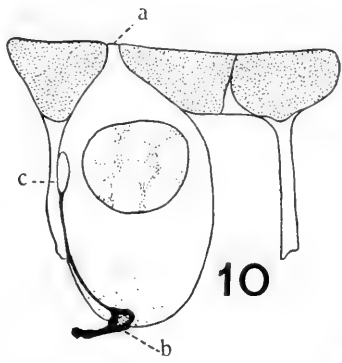
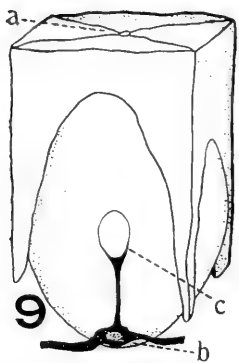
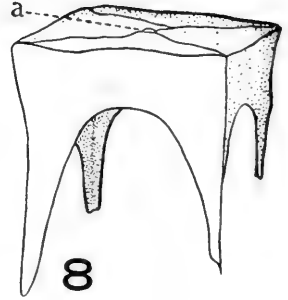
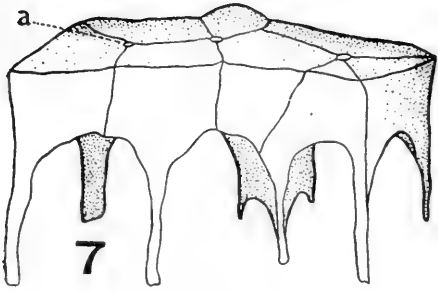


PLATE XXIV



A NEW MONOSTOME TREMATODE PARASITIC IN THE MUSKRAT WITH A KEY TO THE PARASITES OF THE AMERICAN MUSKRAT*

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INTRODUCTION

In studying the parasites of muskrats shot on Lake Chisago, Minnesota, last summer (August, 1915), in addition to several species previously reported for muskrats Barker (:15, p. 184) more than a hundred specimens of a small monostome trematode heretofore unrecorded for the muskrat were found in one male animal.

MORPHOLOGY

To the unaided eye the trematodes appear as very small, thick ovals or globules, milk white in color. The length varies from 0.709 to 0.899 mm., the breadth from 0.501 to 0.657 mm. Under the hand lens or binocular the anterior third or fourth of the body is seen to taper gradually. The posterior third of the body also tapers slightly, but the end is markedly truncate. A rather wide but shallow indentation is fairly constant on each side in the region of the middle third of the body, and the body is widest just anterior to this constriction. A slight, wide but shallow, indentation also occurs in the center of the posterior end of the body, which marks the outlet of the excretory vesicle. The dorsal surface of the body is strongly convex in both diameters; the ventral surface is concave or cupped.

The lateral and posterior margins of the body turn ventrad and mesad forming a ventral horse-shoe-shaped shelf one-fifth to one-

*Contributions from the Zoological Laboratory of the University of Nebraska, No. 115.

fourth the width of the body. The anterior fifth of the body is often strongly flexed ventrad, this flexed condition together with the ventral shelf forms a characteristic elongated ventral cavity or cup-like groove which undoubtedly functions as an effective hold-fast in lieu of an acetabulum, which is absent in this form. The ventral groove is wide and shallow anteriorly and narrow and deep posteriorly. In some specimens the anterior end of the body is straightened out obliterating the cavity at that end. The posterior end likewise may be straightened, thereby breaking the continuity of the shelf and altering the shape of the ventral depression. Ventral papillæ or ridges are entirely absent and the body is devoid of spines or spinelets.

Digestive tract: Only one sucker, oral in position is present. It is slightly sub-terminal, quite muscular, circular or oval in shape and measures 0.05 to 0.06 mm. long by 0.052 to 0.065 mm. wide. The esophagus is straight and about twice as long as the diameter of the oral sucker. A pharynx is absent. The intestinal caeca lie in the median third of the body, and are slightly undulating but without pockets. Their course is nearly straight to the posterior half of the body where they make a decided wide outward bend, but in the posterior fifth of the body they again turn medianward and approach each other but remain separated by a median space equal to about four times their diameter. Both caeca end blindly in the posterior eighth of the body.

Genitals, female: The ovary lies in the extreme posterior end of the body, just to the left of the median line and dorsal to the left testis. Its bulk lies between the posterior ends of the testes, but a small portion overlaps one testis. The ovary is elongated and somewhat convoluted or twisted and the convolutions do not all lie in the same plane which gives a lobed or segmented appearance to the organ in any one plane of focus. The oviduct arises from the internal lobe of the ovary and almost immediately penetrates the shell gland and emerging as the uterus proceeds with several coils anteriorly and to the left around the base of the cirrus pouch. It then passes transversely across the entire body of the worm and in increasingly larger ascending transverse coils extends anteriorly to the level of the bifurcation of the digestive tract, then descend-

ing in wide transverse folds or coils to the level of the cirrus pouch again it passes posteriorly in a sharp right angle turn around the base of the pouch and merges with the vagina. The vagina is narrower than the uterus and lies just posterior to and parallel with the cirrus pouch. The lumen of the terminal third of the vagina is widened and its wall wrinkled affording considerable expansion for the reception of the unusually large thick cirrus. The vagina terminates externally in a large opening the vulva, or female genital pore, slightly posterior and to the left of the opening of the cirrus pouch. The lumen of the terminal portion of the vagina is lined with thick cuticula and surrounded by two muscular sheets, one of longitudinal and one of circular muscle fibers. This muscular wall is in turn surrounded by a large oval mass of gland cells. In many specimens the lumen of the vagina and the contiguous transverse descending arm of the uterus are filled with sperm cells. The vitellarium consists of two compact convoluted tubular masses extra-cæcal in position, one lying on each side of the body in the middle third. From the posterior portion of each gland a transverse vitelline duct passes posteriorly and mesad and enters a small but definite oval vitelline reservoir median in position and lying between the fourth and last posterior fifths of the body just anterior to the shell gland. Neither a Laurer's canal nor a receptaculum seminis were found.

The shell gland lies in the median plane of the posterior fifth of the body between the testes. It is triangular in shape, fairly large, and well defined, tho not prominent. The oviduct passes thru the center of its mass, the cells of which are arranged in a characteristically radiating manner around the lumen of the oviduct.

Eggs: The eggs are very numerous, of a light straw color, oval in shape and about twice as long as wide. The length varies from 0.02 to 0.024 mm., and the breadth from 0.010 to 0.013 mm. A well defined operculum or lid is present at the narrower end of the egg. At each pole the shell substance is drawn out into a very long solid tapering filament. Each polar filament is often five times as long as the egg proper. The filaments of the eggs *in utero* intertwine forming tenacious strings of eggs which can withstand considerable tension. The manner in which these long polar filaments

are formed in the moulding of the egg shell offers an interesting problem.

Genitals; male: The two large testes are lateral in position, extracæcal, one lying on each side, in the posterior third or fourth of the body. They are solid oval or longated bodies longer than wide and are frequently two to five lobed. Their entire mass does not lie in the same vertical plane of the body. The left testis is generally larger than the right one. From each testis a short transverse vas efferens runs medianward, the two uniting to form a wide tubular convoluted seminal vesicle having its base in the median plane at the level of the anterior margin of the shell gland. From this point it passes forward turning to the right around the base of the cirrus pouch which it enters from the dorsal surface. The cirrus pouch is comparatively very large, its length being one-third to one-half the width of the body and lies transversely across the median portion of the body at the anterior level of its posterior half. It is pear-shaped with its base to the right. The base contains an enlargement of the seminal vesicle which is surrounded by the cells of the prostate gland. The middle portion and neck contain the large thick cirrus. The lumen of the cirrus is markedly eccentric, lying nearer the under or ventral surface, and is lined with a thick cuticula. Internal to this lining is a single sheet of longitudinal fibers which aid in the retraction of the cirrus. The bulk of the cirrus is composed of parenchyma tissue and its musculature is poorly developed. No cirrus spinelets are present. The extruded cirrus is always markedly flexed or curved.

Excretory system: A slight depression occurs in the middle of the posterior end of the body and in the center of this depression is a small excretory pore. A very short canal lined with cuticula and surrounded by a row of radiating gland cells connects the pore with a small stellate shaped excretory vesicle or reservoir. From the reservoir four to eight fine radiating canals lead off in all directions and planes. The excretory tubules immediately branch and rebranch, soon becoming so attenuated as to make it impossible to follow their course.

SYSTEMATIC DISCUSSION

Kossack (:11, p. 553) in a thoro revision of the monostome trematodes, groups the five heretofore recognized families in two families, the *Cyclocælidæ* and the *Notocotylidæ*. He clearly points out that the *Notocotylidæ* differ essentially from the other families of monostomes with respect to the following striking characters: the absence of an acetabulum; the presence of ventral glands (drüsenpakete); the absence of a pharynx; the position of the extracæcal testes and the intercaecal ovary in the same plane in the extreme posterior end of the body; the elongated vagina and cirrus sac, which incloses a part of the much convoluted seminal vesicle; the strongly developed vitelline glands lying lateral to the intestinal cæca in the posterior half of the body; the absence of a receptaculum seminis; the uterine coils posterior to the cirrus pouch, and the presence of a polar filament at each end of the egg.

Lühe, (:09, p. 33) created a new genus *Paramonostomum* to contain *Monostomum alveatum* Mehl. *Paramonotomum alveatum* Lühe, conforms to the diagnostic characters of the *Notocotylidæ* with the exception of the presence of ventral glands, which both Lühe and Kossack report they were unable to find either in toto preparations or in sections.

We seriously question the validity of creating a new genus for this species on a single important differentiating character, namely the absence of ventral glands. The number of rows of these ventral glands varies from two in *Notocotylus diserialis* Ssinitzin to three in *Notocotylus triserialis* Diesing and five in *Notocotyle quinqueserialis* Barker and Laughlin and their complete absence in a species might naturally be expected. Their absence then, it seems to us is of specific rather than generic import.

Linton (:10, p. 69) has described a new species representing a new genus of monostomes which he found in three species of tropical fishes. As described by Linton the characters of this new species tally with the characters given for the *Notocotylidæ* with the following exceptions: ventral glands are absent; "the genital aperture is on the left margin approximately at the anterior third of the body;" "the cirrus pouch is relatively large," and a seminal vesicle is present lying "anterior and dorsal to the ovary."

The monostome which we have described has a number of the diagnostic characters of the *Notocotylidæ* such as, the absence of a pharynx; the caudal and parallel position of the extracæcal testes and the ovary; the strongly developed and laterally placed vitelline glands and the eggs with long polar filaments. On the other hand it differs essentially from all of the described species of *Notocotylidæ* in the absence of ventral glands, with the exception of *Paramonostomum alveatum*; the posterior position of the cirrus pouch, vagina and genital pores; the shape and large size of the cirrus and pouch; the position of the uterine coils anterior to the cirrus pouch, and the compact convoluted vitelline glands. Notwithstanding these differences it obviously more closely resembles the *Notocotylidæ* than the *Cyclocaelidæ*. The question immediately arises as to what characters and what combination or complex of characters must be present in a species to rightly place it in the family *Notocotylidæ*. As Kossack aptly remarks "Man ist bei der Schaffung des Trematoden—systems induktiv vorgegangen." It is often difficult to determine what morphological characters are of real phylogenetic import and therefore afford a reliable basis for a natural classification and what characters are due possibly to environment and therefore may be expected to vary and can not be relied on in determining the true systematic position of a given species. It seems to us that such a complex of characters as the absence of a pharynx; the posterior position of the testes and ovary in the same transverse plane with the testes extracæcal and the ovary situated between them and the presence of polar filaments on the eggs may be considered of real phylogenetic significance and for this reason we do not hesitate to place this new species in the family *Notocotylidæ*. Kossack (:11, p. 554), divides this family into the two sub-families *Notocotylina* and *Ogmogasterina* on the difference in the character or arrangement of the ventral glands. In the *Notocotylina* they are in rows (Reihen Drüsenpakete), in the *Ogmogasterina* the glands are arranged along long ribs ("Längsrippen, auf denen Drüsenpakete ausmünden"). We agree with Kossack that such a division is well founded and further propose the creation of a third sub-family to contain such species as those described by Linton and the present paper and others that may be found later which have the general character complex of the *Notocotylidæ* but differ from the

two recognized sub-families with respect to particular characters such as the position of the genital pore; the character of the cirrus; the nature and extent of the vitelline glands and the uterine coils and the absence of ventral glands. For this new sub-family we propose the name *Nudacotylinæ* (devoid of cups) and designate the following characters as diagnostic.

Sub-family Nudacotylinæ

Small *Notocotylidæ*, with thick bodies and without ventral glands. Ventral surface may be strongly cupped. Genital pore lateral (to right or left of median line) decidedly posterior to the intestinal bifurcation. Cirrus pouch large, thick, pear-shape, enclosing a small portion of a convoluted winding seminal vesicle. Vitelline glands strongly developed, compact convoluted masses, extracæcal and lateral in the posterior half of the body.

Uterus strongly developed, transverse folds extend laterally over the intestinal cæca and lie anterior or posterior to the cirrus pouch. Eggs with long polar filaments.

Type genus: *Nudacotyle*, mihi; other genus, *Barisomum* Linton. As the type species of the genus *Nudocotyle* we designate the form described in the present paper which we have named *Nudocotyle novicia*.

KEY TO THE PARASITES OF THE AMERICAN MUSKRAT

- Body flat, unsegmented III
 Body flat, segmented II
 Body cylindrical, unsegmented I
- I. Body differentiated into long slender region and shorter thicker region 2
1. Body not differentiated into two regions. Body stiff, opaque; mouth surrounded by prominent lips..... b
- a. Body thread-like; mouth not surrounded by prominent lips.
- (1) Body hair-like (capillary): male with small bursa and single long spicule; eggs with polar plugs.....
*Capillaria ransomia* (Barker, 1915: 197)
- (2) Body thread-like; male with well developed bursa and two short spicules; eggs without plugs.....
*Trichostrongylus fiberius* (Barker, 1915: 196)
- b. Body stiff, opaque; mouth surrounded by three prominent lips
*Ascaris* sp.¹
2. Body stiff, opaque, divided into long slender cephalic region and shorter, thicker body region.....
*Trichuris opaca* (Barker, 1915: 195)
- Species inquirendæ.*
- Filaria* sp. (in collection of Bureau of Animal Industry, Washington, D. C.)
- II. Found in cysts; in liver..... 2
 Not in cysts; in intestine..... 1
1. (1) Body thin, flabby; genital pores unilateral; single row of hammer-shaped hooks on rostellum; three large testes present
*Hymenolepis evaginata* (Barker, 1915: 194)
- (2) Body thick, stiff; genital pores alternate; double row of long and short hooks on rostellum; testes numerous.....
*Anomotania telescopica* (Barker 1915: 194)
2. Bladder-like cysts in liver.....
*Cysticercus fasciolaris* (Stiles and Hassall, 1894; Linton, 1915: 46)
- III. Two suckers, oral and acetabular, present..... 2
 One sucker, oral or caudal, present..... 1
1. Oral sucker not present..... B
 Oral sucker present..... A
- A. Ventral papillæ absent..... b
 Ventral papillæ present..... a
- a. (1) Three longitudinal rows of ventral papillæ present.....
*Catantropis filamentis* (Barker, 1915: 190)
- (2) Five longitudinal rows of ventral papillæ present..*Notocotyle quinqueserialis* (Barker and Laughlin, 1911: 261)

¹Unpublished research by Bessie Noyes.

- b. (1) Body thin, spatulate.....
*Monostomum affine*² (Leidy, 1858: 110-112)
 (2) Body thick, ventral aspect cupped.....
*Nudacotyle novicia* (the present paper)
- B. Caudal sucker present
 (1) Testes branched.*Cladorchis* (*Stichorchis*) *subtriquetrum*=
*Amphistomum subtriquetrum*³ (Leidy, 1888:126-127)
 (2) Testes slightly lobed.....
*Wardius zibethicus* (Barker, 1915: 192)
2. Body divided into cephalic and caudal regions..... B
 Body not divided A
- A. Oral sucker surrounded by collar, bearing spines..... b
 a. Oral sucker without collar.....
*Plagiorchis proximus* (Barker, 1915: 192)
 b. Eggs numerous bb
 aa. Eggs few (30 to 100).....
*Echinoparyphium contiguum* (Barker, 1915: 187)
 bb. Body 3 to 8 times longer than wide..... bbb
 aaa. Body 10 to 15 times longer than wide.....
*Echinostomum coalitum* (Barker, 1915: 185)
 bbb. Uterine coils definitely transverse..... bbbb
 aaaa. Uterine coils compact.....
*Echinostomum callawayensis* (Barker, 1915: 188)
 bbbb. (1) Anterior testis immediately behind the shell gland...
*Echinostomum armigerum* (Barker, 1915: 189)
 (2) Anterior testis separated from shell gland.....
*Echinostomum echinatum* (Leidy, 1888: 126-127)
- B. Body divided into cephalic and caudal regions.....
*Hemistomum craterum* (Barker, 1915: 191)

²We question the correctness of the identification of this species and surmise that it is a species of *Notocotyle*.

³The identification of this species is questionable and we surmise that it is *Wardius zibethicus*. We wish to express our thanks to Doctor Joseph Leidy, Jr., for his efforts to secure for us the original material of Professor Leidy and regret that it has been misplaced or destroyed and is not available for comparative study.

SUMMARY

1. A new species of monostome trematode, *Nudacotyle novicia*, from the intestine of the American muskrat is described with four figures.
2. The suggestion is made that *Paramonostomum alveatum* Lühe rightly belongs in the genus *Notocotyle*.
3. A new sub-family, *Nudacotylinæ* is created under the *Notocotylidæ* with *Nudacotyle novicia* as the type genus and species.
4. A key to the parasites of the American muskrat is given.

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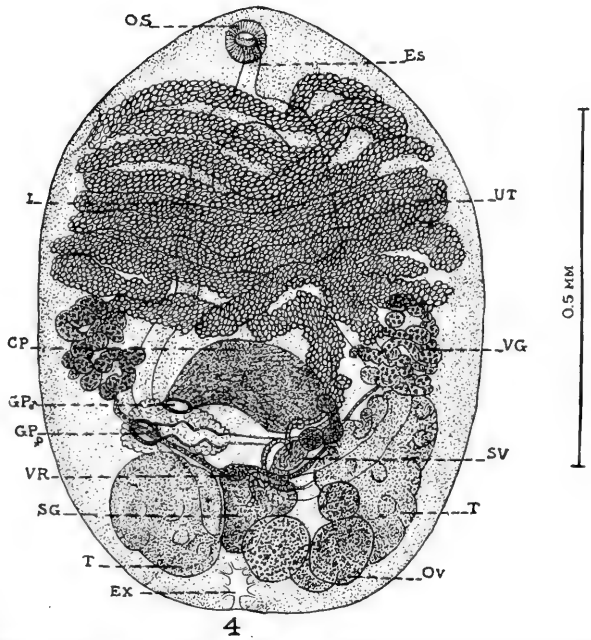
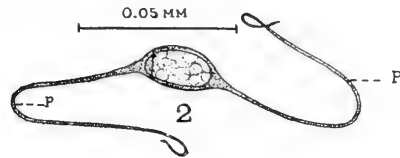
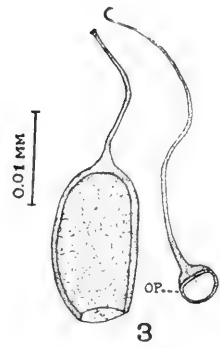
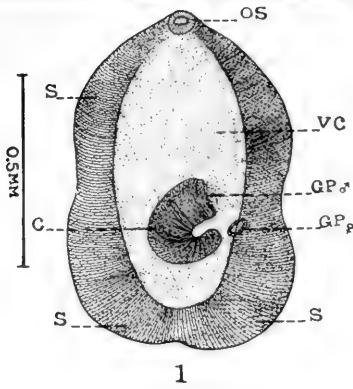
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EXPLANATION OF PLATE XXV

Fig. 1.—*Nudacotyle novicia* Barker, free hand drawing of unstained and uncompressed specimen, ventral aspect, under binocular. OS, oral sucker; C, cirrus, extruded; GP ♂, genital pore, male; GP ♀, genital pore, female; S, ventral shelf; VC, ventral cup.

Figs. 2 and 3.—Eggs of *Nudacotyle novicia*. P, polar filament; Op, operculum.

Fig. 4.—*Nudacotyle novicia* Barker, camera lucida drawing of stained and slightly compressed specimen, dorsal aspect. CP, cirrus pouch; ES, esophagus; Ex, excretory reservoir; GP ♂, genital pore, male; GP ♀, genital pore, female; I, intestine; Ov, ovary; OS, oral sucker; SG, shell gland; SV, seminal vesicle; T, testis; Ut, uterus; VG, vitelline gland; VD, vitelline ducts; VR, vitelline reservoir.



F.D.B. del.

DEPARTMENT OF NOTES. REVIEWS, ETC.

It is the purpose, in this department, to present from time to time brief original notes, both of methods of work and of results, by members of the Society. All members are invited to submit such items. In the absence of these there will be given a few brief abstracts of recent work of more general interest to students and teachers. There will be no attempt to make these abstracts exhaustive. They will illustrate progress, without attempting to define it, and will thus give to the teacher current illustrations, and to the isolated suggestions of suitable fields of investigation.—[Editor.]

SOME METHODS OF PREPARING INSECTS FOR DEMONSTRATION PURPOSES

In teaching entomology it is of considerable importance to provide the students with as much material as possible to aid them in identifying their specimens, to give them an adequate idea of the great number of insects available for study, and to impress upon them the value of insects for the demonstration of biological principles. It is therefore the custom to display upon the walls of the laboratory or on tables collections of insects which will serve these purposes. In the following paragraphs I wish to describe, with the aid of photographs, three methods that have been used in the entomological laboratory at the University of Michigan, which may be of interest to other teachers of the subject.

1. A method of displaying insects for purposes of identification. Photograph 1 shows a laboratory table, two feet wide and five feet long. On it is a removable frame which will carry six Comstock boxes inclined in such a way that their contents can be studied with ease. These boxes can be removed and others put in their place without difficulty. It is thus possible to display a large number of insects in a small space with the use of a laboratory table from which the frame that holds the insect cases can be removed and which therefore becomes available for other purposes. The framework above the boxes may be used for posting laboratory directions, notices, drawings, etc.

2. A method of displaying the life histories of insects. As is well known, an excellent method of displaying stages in the life histories of insects is to place the various stages in Riker mounts. These may be given to the students for study but may also be grouped in such a way as to make beautiful and instructive wall ex-

hibitions. Photograph 2 shows a number of such groups, each group consisting of three or four mounts and arranged according to the economic importance of the insects represented, e. g., one group contains insects of the household (clothes moth, cheese skipper and carpet beetle) and another includes three shade tree pests (the horned tail, tussock moth, and leopard moth.) Students and visitors show considerable interest in these exhibits and, perhaps unconsciously, derive a great deal of information from them.

3. A method of displaying insect galls. Biologically the gall insects are among the most interesting of the whole class. The insect galls may be prepared for exhibition in the following manner (see Photo 3.) Racks three feet long, eight inches high, and two and one half inches deep are made, with the top piece hinged at both ends. Strips on the edges at the top and bottom prevent the bottles from falling out. Galls on stems are dried and placed in large mouthed vials about two inches in diameter, and a label is placed at the bottom. Galls on leaves are preserved in 10% formalin and placed in bottles about two and one half inches in diameter. The bottles or vials fit loosely enough in the racks so that they can be turned around and all sides of the galls can thus be examined but their removal is prevented by the strip near the top and bottom. If, however, it becomes necessary to take out a bottle, the hinge at one end of the top can be disjoined and the desired specimen removed. A background of white cardboard helps to bring out the characteristics of the galls. Such a rack as that described may, of course, be used for other material both zoological and botanical.

*Zoological Laboratory,
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R. W. HEGNER.

THE SEDGWICK-RAFTER OCULAR MICROMETER AND ITS USES

In 1889 Prof. W. T. Sedgwick and Mr. George W. Rafter developed the so-called Sedgwick-Rafter method for enumerating microscopic organisms to be found in water,—a procedure which has since come into general use among biologists, chemists and engineers investigating or in charge of water supplies, and has been incorporated as a part of the Standard Methods of Water Analysis adopted

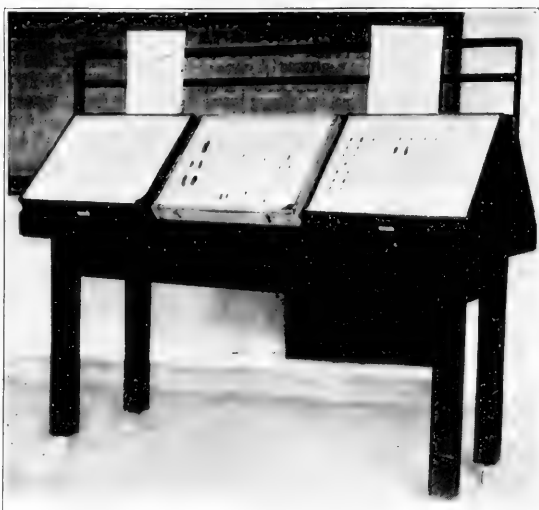


PLATE XXVI FIG. 1



PLATE XXVI. FIG. 2

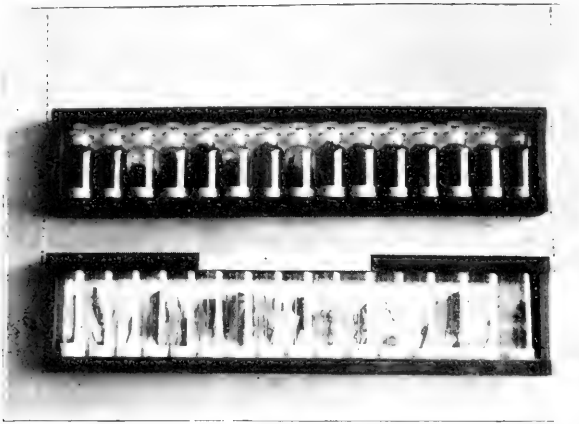


PLATE XXVII. FIG. 1

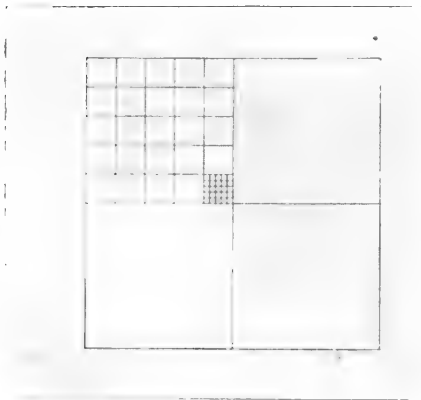


PLATE XXVII. FIG. 2

by the American Public Health Association. This process, the details of which need not be mentioned here, involves the concentration of organisms by means of a sand filter and the microscopic examination of cubic millimeter volumes of the concentrate. The ocular micrometer which was devised for the purpose of ruling off a square millimeter surface on the "ringed" slide has been found useful not only in the enumeration and identification of organisms, but also in the broad field of general microscopic work and perhaps deserves a word of commendation to microscopists.

The micrometer is sub-divided as shown in the accompanying diagram and when used with an ocular of the No. 4-x Spencer type (2 inch) and a 16 mm. objective, can be so standardized by the aid of a stage micrometer that the large square will outline a square millimeter surface. The smallest square will then be 20 micra on a side and the micrometer will form a convenient measure for the larger microscopic objects. By only a slight change in the tube length, moreover, a standardization with the 4 mm. objective can be made which will increase the magnification five-fold, thereby reducing the side of the large square to 200 micra and that of the smallest square to four, so that, with the high power, the dimensions of all but the very smallest microscopical organisms can be readily obtained.

But ready adaptability to either high or low powers of the microscope is not the only commendable feature of this type of micrometer. There is a marked advantage in the way it is ruled into squares of various sizes. It enables one to measure the length and the breadth of an object at the same time and yet leaves the greater part of the field of vision relatively free from lines interfering with clear definition of objects. It is thus adapted to the use of the beginner as well as to that of the seasoned microscopist. The student beginning microscopic work in zoölogy, botany or histology, for example, is studying anatomy with no basis or standard of size-comparison, other than that offered by different objects in the same field. The standardization and use of the micrometer gives him an appreciation of the magnifying power of the microscope by demonstrating the apparent size of a familiar and fundamental unit, the square millimeter, and teaches him the approximate relative

size of the microscopical linear unit or micron. The student finds the device convenient to handle, and easy to use and soon acquires a mental scale of unit areas by which he can estimate the size of organisms or other structures in micra much as he judges of macroscopic structures in centimeters or inches.

Ocular micrometers may be prepared either by engraving or photography and are not difficult to procure. Engraved micrometers are somewhat superior and are sold by Bausch and Lomb under the name of "Whipple's Eye-Piece Micrometer" at \$3.50 each. Those made by photography are much less expensive and have been found entirely satisfactory. In either case, the actual size of the large square ruled on the glass should be 7 mm. on a side. In photography the lines are produced on lantern-slide glass by photographing a specially prepared sketch. The glass is then cut and mounted on a clean cover slip with gum damar. The only difficulty in using this type arises from the fact that the lantern-slide glass is rather thick and cannot be perfectly cleared without removing the fine lines. The fault is not serious, however, and may be readily overlooked if expense must be considered, for these micrometers can be made for about 75c each.* This is relatively very inexpensive, for the common linear ocular micrometer, which is much less adapted to work with living forms and hardly more valuable for exact measurements, is not obtainable for less than \$1.25.

The advantages of this type of micrometer are its ready adaptability to general use with either high or low powers of the microscope; the definition of measured squares or unit surfaces, which are easier to use and to fix in mind than are linear units; a comparatively unobstructed "field"; and (in the photographic product) low cost.

*Unmounted ocular micrometers may be secured at this price from Mr. B. S. Turpin, 30 Trinity Place, Boston, Mass.

C. E. TURNER,

*Instructor and Research Associate Department
of Biology and Public Health. M. I. T.*

FORMATION OF SPORANGIA IN STEMONITIS

Hilton (Jour. Q. M. C., Apr. 1916) contributes an interesting note on the method of forming sporangia in this common mycetozoon. He had the good fortune to find considerable masses of plasmodium just in condition to observe the whole process. At noon the plasmodium formed somewhat rounded, solid, cushion-shaped masses. This surface differentiated in about a quarter of an hour into frothy, bubble-like hemispheres which divided and covered the entire surface regularly. By 4:00 P. M. each mass of hemispheres had contracted in width and increased in height; and the basal part constricted into flutings corresponding to the surface hemispheres. These flutings gradually contracted to pillars, the creamy protoplasm withdrawing more and more to the upper third. In another half hour black stalks were visible as the cores of the pillars. By 8:30 o'clock all the protoplasm had risen clear of the substratum and the still cohering heads of the sporangia appeared resting on a forest of black stalks. By 10:00 P. M. the sporangia had virtually assumed their permanent shape and were beginning to darken.

A DROUTH-ENDURING ZYGNEMA

Fritsch (Ann. Bot. 1916, pp. 135-149) reports upon a *Zygnema* especially adapted to terrestrial conditions. The longitudinal walls become much thickened, showing two or three successive layers. The outer layer of the wall is mucilaginous. This doubtless is an adaptation which prevents too rapid drying and aids resorption of water on return of moist conditions. It was found on Hindhead Common (England.)

There are two chloroplasts in the mature cell. Division takes place by an infolding or growth from the inner layer of the cell wall. The growth of this gradually constricts the protoplast, but the division may not be completed for considerable time. In this way the two daughter protoplasts have at first a connection thru the center of this plate.

With the oncoming of drouth the fat globules of the cell pass to the outer surface, and form there a dense layer just beneath the cell wall. When the plant begins to absorb water the fat drops

come to be distributed again. When a drouth begins, the protoplast develops a new layer of the cell membrane. A cell divides not more than twice between two drouths. Agamic reproduction may take place early in the year, by the cell dividing unequally into a smaller pigment cell and a large, simple reproductive spore (akinete.) The pigment body disintegrates, the wall weakens and this becomes a breaking point for the dividing up of the filament.

BACTERIA AID IN FORMATION OF EUROTIIUM

Sartory and Roger (C. R. Soc. Biol. Paris; 79: pp. 174-5) found, in a variety of *Aspergillus B* grown on damp straw, that they could secure promptly and abundantly and constantly the formation of perithecia, provided the culture was innoculated with microorganisms of the *B. mesentericus* group. Otherwise, he found, with pure cultures of the *Aspergillus* he could not get the *Eurotium* even with the aid of the various media hitherto suggested by students as valuable in this connection.

BACTERIAL INFECTION IN FRESH EGGS

Hadley and Caldwell (Bul. 164: R. I. State Col. Ag. Exp. Sta.) have discovered 8.7% of fresh eggs show bacterial infection of the yolk. The whites were sterile in all cases examined. The fertilization of the egg made no difference in the percentage. Forty different bacterial forms were found. There were no streptococci, and none of the groups causing hæmorrhagic septicæmia, enteritis, typhoid-dysentery, or diphtheria.

The study was instituted to throw light on the mortality of embryos in incubation, and the degree to which the mortality of chicks in brooders may be influenced by egg infection from mothers harboring the germs of diseases.

SOME REMARKABLE FEEDING ACTIONS OF AMEBÆ

Mast and Root (J. Exp. Zool. July, 1916) report studies of the capture of rotifers, paramecia, and other ciliates, by *Ameba*. They capture rotifers by flowing around the foot while attached. The protoplasm gradually flows upward along the stalk. The rotifer contracts in the effort to relieve the pressure; but when it extends

the ameba begins its flow again. In the meantime the rotifer is gradually weakened by the digestion of the foot. It may require days to engulf the rotifer.

In the capture of paramecia the amebæ take a sort of mushroom shape. The free margin is made irregular by numerous short pseudopodia. This furnishes a space beneath the umbrella and recesses at the margin in which paramecia tend to come to rest. Pseudopodia enclose the paramecia from either side. In some instances pseudopodia reached only about half the length of the paramecium and by turning toward the body of the prey compressed it so as to cut the animal in two,—engulfing only the inner half. This whole process required only about ten seconds. This was not an isolated incident, but was observed many times.

The writers, by computation based on the amount of pressure required to cut paramecia with a thin glass thread, have reached the conclusion that this amputation of paramecia by *Ameba* could not be explained by surface tension in the protoplasm of *Ameba*.

CASE OF BROODING IN HOLOTHURIANS

Ohshima (Ann. Zool. Japon., June, 1916) reports a new case of internal brooding in holothurians,—*Pseudocucumis africanus*. As many as 25 and 27 young were found in the body cavity of the mother. Three such brood-carrying individuals were found. The author was unable to discover either how fertilization occurs or the young escape.

EFFECTS OF ACTIVITY ON NERVE CELLS

Kocher (J. Comp. Neur., June, 1916) after fifteen separate experiments, completely controlled, on six different species of animals, using cells from various regions of the nervous system, reaches the conclusion that there is no deviation from the normal, either qualitative or quantitative, that can be revealed by the most exacting cytological tests in nerve cells even in the most advanced fatigue. He ascribes the varying results of former investigators to:—(1) difficulty in separating the effects of normal activity from unavoidable shock or injury to the nervous system in killing the animal; (2) post mortem changes taking place before complete fixation; (3)

varying chemical action of fixing agents; (4) solvent action of materials used in fixation and imbedding; (5) varying effects of chemical reaction between basic or acid stains and the different cell structures; and (6) effect of subjecting tissues to the temperatures necessary in manipulation. The author used an elaborate series of checks in control of the studies.

TRYPANSOME INFECTION IN MAMMALS

Lanfranchi (Atti. R. Ac. Linc. 1916, pp. 369-73) believes that *Trypanosoma brucei*, *T. gambiense* and *T. vodiense* can pass from the blood of a pregnant mother into the milk; and that by the first two, at least, the fœtus may also be infected in the same way.

IMPROVING TECHNIC FOR SHOWING DETAILS OF DIVIDING CELLS

Allen (Anat. Rec., July, 1916), as the result of a series of critical experiments to perfect the technic for demonstrating the details of mitosis in the central nervous system and in the testis of the albino rat, without producing distortion of adjacent tissues, offers the following suggestions:—

“For cytological work, the slightest gain at any point in the technic is worth working for.

“Very gradual changes of fluids, agitation of fluids during changing, and slow infiltration appear essential in order to get the best results from any fixative.

“The addition of a low percentage of urea to fixing fluids results in sharpening the chromosomes and preserving the structure of the achromatic nuclear material. It may help the penetration of the fluids.

“Picro-formol-acetic mixtures are more effective when used at about 38° C. Cold is detrimental.

“Flemming’s fluid is more effective if used at 0° C. or a few degrees lower.

“Flemming’s fluid is of no value as a brain fixative at any temperature. At times (if urea is added) it isolates metaphase and anaphase chromosomes in spermatocytes somewhat better than any

other fixative tried (except B-15*), but is hard on the rest of the tissues and shrinks heavily.

"Anilin oil is an excellent substitute for the higher alcohols.
"Xylol shrinks tissues more than the vegetable oils."

VISUAL EFFICIENCY IN THE USE OF OPTICAL INSTRUMENTS

Purkis (J. R. M. S., June 1916) makes some good practical working suggestions for those who use the microscope for prolonged observations, where necessity of accurate observation and minimum fatigue are necessary.

1. The fact that only one plane is in sharp focus at a time and that other planes show dimly tempts the novice to strain the eye in the effort to make it see things which are not clearly in focus, instead of adjusting the distance.

2. This suggests also that care should first be taken to discover the limitations of the instrument which cannot be corrected by manipulation and to accept these, refusing to try to make the eye compensate for these limitations, by intensely close observation.

3. The eye should look at the field almost casually. What it cannot see by looking quietly at the object, it cannot see by an intense and strained gaze.

4. Recognizing that there is a certain amount not only of shock to the eye in sudden changes from dark to light luminosities and conversely, but also strain of the eye in the effort to see well before the re-adjustment is completed, it is important to avoid so far as possible sudden changes of luminosity. In this connection it is well (a) to see that the illumination of the room is not in too great contrast with that of the instrument; (b) to modify luminosity to compensate when passing from one objective to another; (c) to avoid sharp contrasts of luminosity within the field itself; and (d) when resting the eye preparatory to observation or during observation to do so in approximately the same illumination to be used in the field of the instrument.

*B-15 is a picro-formol chrome-acetic mixture in urea:

Picric acid	75cc
Formol	25cc
Acetic acid	5cc
Chromic acid (crystal).....	1.5 grams
Urea	2.0 grams

NORTH AMERICAN DIATOMACEÆ

A new guide for the American student of Diatoms will be of interest to many members of the American Microscopical Society. Charles S. Boyer has issued a monograph of most painstaking character on the Diatomaceæ of Philadelphia and vicinity. The book opens with an introduction descriptive of the physical condition of the Philadelphia region,—which means a territory with a radius of about 100 miles from Philadelphia. A short chapter is devoted to the structure, reproduction, evolution, and rôle in nature of the diatoms.

The body of the book is given to keys, descriptions, and figures of nearly 600 species and varieties. There are 700 drawings by the author illustrating these species. All these drawings are to uniform scale,—about 800 diameters. In his classification the author adopts Schuett's synthesis of the various bases of classification of Pfitzer and Petit, Smith, Mereschkowsky, as well as the older methods. The synonymy is for the most part omitted.

The appendix contains suggestions as to the collection and preparation of diatoms for mounting and study. Many of these will prove most helpful for the inexperienced student of the group. The headings:—Collection of fresh water material; Collection of marine material; Blue clay deposit; Cleaning material; Preparation of strewn mounts; Preparation of selected mounts; Instruments required,—will sufficiently indicate the scope of these hints. Mechanically the book is very attractive. It is bound in art vellum cloth, the pages are 9x12, and the text is printed on heavy unglazed paper.

The Diatoms of Philadelphia and Vicinity, by Charles S. Boyer, A.M., F.R.M.S., 143 pages, 700 illustrations. J. B. Lippincott Company, Philadelphia, Pa., 1916. Price \$5.00.

NECROLOGY

- Burrill, Thomas J, Ph.D. '78, President A.M.S.,
1886 and 1904..... Urbana, Ill.
- Lewis, Ira W. '87..... Dixon, Ill.

IRA W. LEWIS

Mr. Lewis, who was for nearly twenty years a member of the American Microscopical Society, died May 26, 1915, at the age of 73. He was a man of most substantial character and attainments. For 45 years he was connected with the office of the Clerk of the Circuit Court of Lee County, Ill., as Deputy or as Circuit Clerk. The Bar Association of the County testified in a public Memorial Service to his unvarying efficiency and high mindedness.

He was a devoted churchman and gave much of his energy to work in the moral and religious education of youth. He was a self cultured man, and his memorialists testify with great uniformity that he was one of the most remarkable and versatile men of his community.

In the meantime he gained local note as a student of books and of nature. He was very devoted to his work with the microscope and collected an extensive library of microscopical journals and scientific books. It would be greatly to the credit of American culture if we had more such amateur students with the microscope.

His wife survives him.



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HERBERT OSBORN, M.S., of Columbus, Ohio,	at Minneapolis, Minn., 1910
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F. D. HEALD, Ph.D., of Philadelphia, Pa.,	at Cleveland, Ohio, 1912
CHARLES BROOKOVER, Ph. D., of Little Rock, Ark.,	at Philadelphia, Pa., 1914
CHARLES A. KOFOID, Ph.D., of Berkeley, Calif.,	at Columbus, Ohio, 1915

The Society does not hold itself responsible for the opinions expressed by members in its published *Transactions* unless endorsed by special vote.

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NOTICE TO MEMBERS

Business Sessions of the *American Microscopical Society* will be held at New York City, as follows:

Dec. 28 (Thursday), 8:15 A. M., Executive Committee will meet at breakfast at *Hotel Martinique*.

Dec. 28, 4:30 P. M., General business meeting of the Society, immediately following the adjournment of the American Society of Zoologists, in the Hall used by them.

The Headquarters of the *American Microscopical Society* will be the Hotel Martinique, Broadway, 32-33d Sts. It is convenient—near the Pennsylvania Station—and reasonable in rates.

T. W. GALLOWAY, *Secretary*,
Beloit, Wisconsin.

(This Number was issued on December 16, 1916.)

TRANSACTIONS
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OCTOBER, 1916

No. 4

REPORT OF THE SECRETARY AND EDITOR

T. W. GALLOWAY

Beloit College, Beloit, Wis.

Three years ago the Secretary reported to the Society the outstanding facts relative to the state of the organization during the first term of his service. The present number marks the close of the second term of three years. In the absence of large and well attended meetings of the members, where discussions both of scientific and business details would place in the minutes much that would enlighten the members as to the work of the Society, it appears that a triennial report by the Secretary may well supplement the annual reports of the Treasurer and the Custodian.

It seems necessary, for the sake of efficiency in such a Society, that a large amount of the leadership and decision of policies fall upon the Secretary. The organization pays for this freedom of action on his part, however, in a certain loss of responsibility and mutuality on the part of the membership. The Secretary feels very strongly the lack of positive and constructive,—or for that matter any kind of,—criticism.

Attention was recalled in the last report to the transitional period through which the Society has passed in the last quarter of a century, and to the decision four years ago to try to find a function for the Society which would be at once worthy of its best traditions and place it in a position in which it may continue, in spite of the multiplication of special societies, to serve American science and scientists. It is too much to hope that this has been done as well as it might have been done. Yet evidences are not wanting that the policy adopted then has been on the whole well chosen.

There is, however, one group of our most faithful and effective members whom we have not been able to serve as the Secretary would like. This is the group of students who are working with the microscope independently, usually out of contact with colleges and laboratories, browsing more or less in the numerous interesting fields opened to them by the microscope. In many cases they are working in an amateur way, more or less intensively upon one or more of the groups of microscopic forms. In some instances these students are serving, by private correspondence, as a clearing-house for information for amateurs. The Secretary has the impression from correspondence that this group is again becoming larger than it has been for 25 or 30 years. It finds our research articles too technical and would greatly prize a much enlarged discussion of elementary and advanced, up-to-date methods of manipulation, such as distinguished the "Journal of Applied Microscopy" some 15 years ago. A department devoted to these interests would probably find acceptance with a considerable number of people. Two English journals,—the "English Mechanic and World of Science" and the "Journal of Micrology and Natural History Mirror" serve this end in some measure. The Secretary-editor has tried, in an experimental way, to relate the Society to some of the local amateur microscopical clubs with the hope that at least a small department of this type of material might be established in the *Transactions* to encourage again in America high grade amateur work on the part of intelligent people who do not expect to use the microscope for professional ends. Thus far we have been unable to do anything significant in this direction. All his own spare time is so taken up in carrying out the policy adopted as our main objective that he could not, even if his ability ran in this direction, do the work himself. It has not been possible to find any one at once able and willing to do this in the large and scientific way which would be necessary to make it worth while. Suggestions from the members will be welcome.

In other respects the work of the Society seems to have progressed reasonably. The usual volume of four numbers and about 300 pages has been published each year. It is still true that we do not receive contributions enough in the varied fields of interest

to make it possible for us to select articles in such a way as to furnish a balanced biological ration. For some years the zoological contributions have dominated. Thanks to our botanical friends, chief among whom was Professor Bessey, a much larger botanical element has appeared in the *Transactions*. For various reasons it has been impossible to issue the quarterly promptly in the month of its date.

The Secretary has been particularly disappointed in respect to one class of contributions which he hoped to feature in the *Transactions*. It is a strong conviction of his that nothing we can do would serve American science so distinctively and give the *Transactions* so welcome a place in every biological library as a series of digests covering part by part the special fields that go to make up biology. If this territory were marked off into 20 or 25 subdivisions, and an expert were to present once in five or six years a thorough-going summary of each of these, we could by giving one such digest in each issue cover the field in this time. Such a digest would not undertake to be exhaustive as might be needed by the men in the field; but would be interpretative of the most important results, the main conclusions and tendencies and prospects, together with such references to the important papers as would serve the need of the student outside the particular field rather than of those within it. A few such have appeared and these in every way have confirmed the editor in his conviction of the tremendous service such an enterprise would render to the rank and file of American students and teachers of biology. The attempt has thus far failed because the men who are most able to command both the territory and the audience are unable to command the time. The Secretary is coming to be of the opinion that the income of our Spencer-Tolles Fund apportioned as an honorarium for digests of this kind would render a more distinctive and a more vital service than it can do as our present rules demand.

Several suggestions have come urging that emphasis be placed upon the technic and methods of biology and microscopy. Some progress in this direction is being made and more is planned. We have not yet secured a steady stream of such communications; but

there has been an encouraging response. The Secretary urges strongly that every member who sees this notice "highly resolve" to supply this department with a description, illustrated if desirable, of every new unpublished device for investigation, illustration, and teaching which his own experience approves; and furthermore secure similar returns from all his colleagues, whether members of this Society or not. There is no present point at which so vital a service can be rendered the *Transactions* and the membership by the individual member. We shall be glad to become a clearing house for the best biological methods developed in America.

We are still growing in numbers and in financial strength. When the present Secretary took charge in 1909 there were on the roll 226 names of members and 33 subscribing libraries and individuals. A considerable number of these quickly dropped out. In all, 127 of this list have ceased to be members, leaving 99 members at present whose connection with the Society goes back of 1911. In the six years there have been added 300 members and 67 subscribers. Of these members 57 or about 20% have discontinued. Of the subscribers only three or four have been lost. There is now an enrollment of 343 members and 97 subscribers, or a total of 440 supporters. One of the chief deductions from our membership history is that the list of subscribing libraries should, if possible, be run up to 150 or 200. This would furnish a group of supporters not nearly so fluctuating as individual memberships. A library once possessed of a fairly complete file of our volumes is increasingly likely to continue its connection. Because of this the Executive Committee at the Cleveland meeting authorized the Secretary to adopt with libraries a liberal policy in the distribution of partial sets made up of those back volumes of which we have excessive numbers. We have been able to supply partial sets, sometimes running as high as 18 or 20 volumes, on condition that the library would begin a subscription with Vol. 29, at which point we began to publish in parts. The Secretary will welcome correspondence from members connected with institutions with growing and permanent libraries, which may not be

able to buy a complete set, relative to the conditions of this distribution of the back volumes.

The total resources of the Treasurer in 1911 were \$782. The annual resources for the succeeding years were \$1234, \$1433, \$1379, and in 1915, \$1522. The society is very nearly to a point of self-support, which will not demand a high-pressure campaign for members, but merely demand effort enough to replace those who are lost. It ought to be possible within the next year or two to push the members and subscribers to the 500 mark.

The Spencer-Tolles Fund under the wise and enthusiastic management of the custodian, Mr. Pflaum, has climbed steadily during the period, from \$3352 in January, 1912. The custodian will report at the next meeting close to \$5000. This will mean an annual income of \$300,—which may be devoted to the advancement of science. The Secretary urges those members of the Society who are engaged in research, in any field where such a grant would signally advance the investigation, to make inquiry of the Chairman of the Committee on Grants,—Professor H. B. Ward, University of Ill., Urbana, Ill.



A COMPARATIVE STUDY OF EPIGYNY IN CERTAIN MONOCOTYLEDONS AND DICOTYLEDONS*

By MARGARET HANNAH

It is not the purpose of this study to give the complete details of the organography of the different flowers, which have been chosen for this comparison but the steps in the development of the floral organs have been given in sufficient detail to show the development of the inferior ovary. From these a comparison of epigyny in the two great divisions of the flowering plants has been made. Very little work has been done on this special phase of the subject, although different writers have their own explanation for the manner in which epigyny is produced.

Martin (6) in 1892 wrote, "The real origin and behavior of the floral organs in their younger stages of development as correlated with the inferior ovary has attracted but little attention, and therefore, no definite statement can be made as to the true relationship existing between the floral organs in their embryonic condition". In the same paper, in speaking of the tubular mass of tissue, he wrote, "In which there is a complete fusion of the parts until liberated", and a theory was proposed "that all the floral organs are coalesced in their initial state in the annular wall, and each appears as the upper parts are liberated".

Gray (5) wrote, "Where the adnation is complete to the top of the ovary and none beyond it".

Goebel (3) in 1887 defined an epigynous flower in this manner, "The walls of the ovary are formed from the torus itself, which is hollowed out into the shape of a cup, or even a long tube, while the carpels which form the entire wall of the free superior ovary, spring like the perianth and androecium from the margin of the hollow torus and only close its cavity above, being there prolonged into the style and bearing the stigma. The inferior ovary is formed by the terminal growth being retarded and by the outer rim growing upward. The placentae may be regarded as the margins of the carpels running down the inner surface of the ovary wall".

*Contribution from the Department of Botany, The University of Nebraska.

In his *Organography of Plants*, published in 1901, Goebel (4) said in contradiction to what he had said in his book, published in 1887, "On account of deficient historical investigation, the view was formerly advanced, that the ovary of the epigynous flower is formed from the cup-like flower axis, and the carpellary leaves only produce the style and stigma. Comparative morphology has rightly contradicted this interpretation, which is still found in many books. As the history of the development shows, the carpels share in the formation of the ovarian cavity, and the ovules have no other origin than in the superior ovary.

"In all inferior ovaries, the vegetative point becomes at an early period more or less concavely hollowed out, and the leaf structures of the flower arise, partly from the margins, and partly from the inner surface of the depression. Whether one describes the marginal part of the flower axis as a 'congenital concrescence' of the different leaf whorls of the flower is an arbitrary matter, because the flower axis ends its active existence with the bringing forth of the leaf structures of the flower. The earlier the flower axis assumes the cup-like form, the more will we in general ascribe its character to the flower axis. The later this form is assumed the more will its features approach the more primitive condition as we find it in hypogynous flowers".

Wylie (8) in 1904 worked out the morphology of *Elodea canadensis*. In this study, he showed how epigynous flowers have developed in this primitive type of monocotyledon.

Coulter (1) in 1883 investigated the dandelion flower, giving as his purpose an investigation of the development of the inferior ovary. He said, "The inference is that all four of the floral organs are blended together in the primitive ring, which rises from the original obconical mass, that they are essentially hypogynous and that their separate appearance is a freeing of their upper extremities. It was attempted in vain to detect in the primitive ring or later in the wall of the ovary any evidence of the blending of two or more distinct parts. No such indication could be found, and the inference that all four floral parts are represented in the wall of the inferior ovary rests, not so much upon the structure of the wall, as upon the order of the succession in the appearance of the floral parts".

He believes the theory, that the primitive ring belongs to the receptacle, is not tenable for two reasons: (1) "the late appearance of the calyx"; (2) the fact that the corolla lobes appear with the ring and not after it, indicating that the ring belongs to the floral organs. "The inferior ovary is produced by an arrest of the development of the floral axis the rising in a peripheral ring of the floral organs and a gradual arching over of the carpelary leaves".

Merrell (7) in his paper on *Silphium* said, "The outline of the receptacle soon became angular by the upward growth of the marginal ring, which is the beginning of the corolla tube".

Six species of Monocotyledons and nine species of Dicotyledons, representing twelve different families, have been included in this comparative study.

The different species are named below under their respective groups:

1. Monocotyledons: *Sisyrinchium angustifolium*; *Gladiolus gandavensis*; *Iris germanica*; *Freesia refracta*, *Var alba*; *Musa sapientum*; and *Canna indica*.
2. Dicotyledons: *Malus ioensis*, *Ribes aureum*, *Fuchsia speciosa*, *Citrullus vulgaris*, *Sanicula canadensis*, *Galium aparine*, *Sambucus canadensis*, *Valeriana officinalis* and *Helianthus annuus*.

MONOCOTYLEDONS. Plates XXVIII-XXXI

Sisyrinchium angustifolium. Plate XXVIII

The flowers are produced in clusters, developing in centripetal order. The buds begin as protuberances of meristematic tissue, which appear slightly triangular in cross section, due to the arrangement of the flowers in the cluster. Soon three lobes, the beginning of the stamens, grow up from the rim of the receptacle, which has become flattened (Plate XXVIII, Fig. 2). The lobes of the perianth soon appear at the sides of the mass (Fig. 3). At about the same time, the tissue just below the origin of the perianth and the stamens begins to elongate, forming a tubular ring about a shallow cavity in the center (Figs. 4, 5 and 7). The three lobes, which are to form the carpels, soon grow out from the inner

rim of the tubular cavity (Figs. 5 and 6). A mass of tissue soon roofs over the central cavity, and a continuation of the three lobes upwards forms the style branches (Fig. 12). From three sides of the cavity, tissue begins to grow towards the center (Fig. 8). The fusion of these three outgrowths divides the ovary into three parts (Figs. 10 and 11). The tissue at the point of origin of the sepals and the petals grows *en masse*, so that the sepals and petals are joined together for a short distance, forming a tubular ring.

Gladiolus gandavensis. Plates XXVIII and XXIX

The earliest stages in the development of the flower of the *Gladiolus* were not found, but stages showing the growth of the tissue forming the tubular rim and the central cavity are shown in Plate XXVIII, Fig. 13. The perianth and stamen lobes have appeared. Further development is similar to *Sisyrinchium*. The figures show the origin of the parts and the final development of the inferior ovary. The order of the succession of the floral leaves is sepals, petals, stamens and pistils.

Iris germanica. Plate XXIX

The development of the flower of the *Iris* is almost the same as that of *Sisyrinchium angustifolium*, differing in the order of the succession in the development of the floral leaves. In the *Iris*, the order is perianth, stamens, and carpels (Figs. 2, 3 and 4). The petals and stamens appear almost at the same time, so nearly so, that it is difficult to determine which lobes really appear first. In *Sisyrinchium*, the stamen lobes grow out first (Plate XXVIII, Fig. 2) and the lobes of the perianth grow out at the sides, as described under *Sisyrinchium* and shown in (Plate XXVIII, Fig. 3.)

Freesia refracta. Plates XXIX and XXX

The flowers are produced on two sides of an elongated axis, each flower subtended by a bract. The flower begins as a protuberance from the side of the flower axis. This undifferentiated mass of cells soon flattens and broadens, and distinct lobes appear, the sepals (Fig. 7). Inside the whorl of sepals, the stamens ap-

pear (Fig. 8). At this time, the tissue below the lobes begins to elongate, forming a tubular ring with a shallow cavity in the center (Fig. 9). The lobes of the carpels appear (Fig. 1 and 2, Plate XXX). There is further elongation of the tissue just below the point of origin of the floral organs. The three carpel lobes grow out into the central cavity, and, growing upward and toward the center, cover the cavity. An upward extension of the three masses forms the three style branches, which appear as three lobes only at the top (Fig. 3).

Musa sapientum. Plate XXX

The method of origin and early development of *Musa sapientum* is like the *Iris*, *Freesia* and *Gladiolus*. The later development shows a zonal development of the tissue near the bases of the sepals and petals, so that the sepals and two of the petals are joined together nearly to their tips. The other petal remains free.

Canna indica. Plates XXX and XXXI

The flower begins as an outgrowth which broadens; the sepals grow out from the top of this undifferentiated mass (Fig. 11, Plate XXX), and just inside of these lobes, other masses grow out, and very soon separate into two parts (Plate XXXI, Fig. 1). The inner whorl forms the four staminoidia and the one fertile stamen, and the other whorl the petals. The tissue at the base of these lobes now elongates in the peripheral portion leaving a hollow central cup (Figs. 2 and 3). Further development resembles *Sisyrinchium*, differing in the form of the style. The tissue at the upper portion of the central cavity grows up into a somewhat flattened mass forming the style (Figs. 4 and 5).

Wylie (8) found the development of the primitive, Monocotyledon, *Elodea canadensis*, quite like that given above for the higher Monocotyledons. Buds begin as protuberances, apex of receptacle flattens and broadens, a mass of tissue grows up leaving a tri-radiate slit down the center. The order in *Elodea* is sepals, three sterile stamens, three stigmatic lobes, and finally the petals.

In the Monocotyledons described above the tri-radiate slit did not form until after a circular cavity formed and three masses of tissue grew out into this cavity.

DICOTYLEDONS. Plates XXXI-XXXV

Malus ioensis. Plates XXXI and XXXV

Material was not collected early enough to show the first stages in the development of the flower of the *Malus*, but early stages to show the development of the inferior ovary were found. The floral organs, sepals, petals and stamens are shown in Plate XXXI, Fig 6. At the time that the lobes of the floral organs appear, there is an upward growth of the outer rim of the receptacle, leaving a shallow, broad, central cavity (Fig. 7). The carpel lobes grow out from the bottom of this shallow cavity (Fig. 8). The elongation of the rim of this cup produces the wall of the inferior ovary (Fig. 9). Very soon after the appearance of the lobes of the carpels, a cross section of the ovary shows five masses of tissue growing in toward the center (Fig. 10).

Ribes aureum. Plate XXXII

The order of the development of the floral organs of *Ribes* is similar to that of *Malus*. The elongation of the cylindrical mass of tissues leaves a narrow slit (Fig. 3), from the sides of which, two protuberances grow out, forming two opposite lateral placentae (Figs. 4, 5 and 7). Two lobes of the style form with only one cavity in the ovary. The cross section shows two bundles, one for each carpel (Fig. 7 d).

Fuchsia speciosa. Plates XXXII and XXXIII

The flowers are solitary in the axils of opposite leaves. The flower buds begin as hemispherical masses of undifferentiated tissue (Plate XXXII, Fig. 8a). The receptacle flattens, and the four lobes of the sepals appear on the peripheral portion. This outer portion elongates, forming a shallow cup-shaped cavity in the center (Figs. 9 and 10). The petals and stamens grow out from the inner surface of this cup, near the bottom (Fig. 11).

The zonal development continues. The cup-like cavity elongates. Just below the stamens, the four lobes of the carpels appear (Fig. 12). An upward prolongation of these lobes forms the style branches and roofs over the central cavity (Fig. 13). The four cavities of the ovary are formed by four masses of tissue growing toward the center and fusing. (Plate XXXIII, Figs. 1, 2 and 3).

Citrullus vulgaris. Plate XXXIII

The origin and development of the floral organs of the flowers of *Citrullus* are very similar to those of *Fuchsia*, except for the number of parts. There are the five sepal lobes (Fig. 6), petals (Fig. 7) and stamens (Fig. 8). As in the other Dicotyledons, there is the hollow cavity in the center (Fig. 9). The sides of the cup elongate to form the inferior ovary (Fig. 11).

Sanicula canadensis. Plates XXXIII and XXXIV

What has been said about the *Fuchsia* is also true, in part, for *Sanicula*. From the flattened receptacle the five lobes of the sepals grow out (Fig. 13). The petals appear just inside but alternate with the sepals (Fig. 13). At the point of origin of these parts there is the zonal elongation, so that the sepals and petals are adnate a short distance above the ovary. After the cavity of the ovary is formed, a two-lobed mass of tissue grows up from the bottom of the cavity (Fig. 15). The base of this tissue elongates carrying the lobes upward, until they meet the tissue, which roofs over the cavity of the ovary Plate XXXIV (Figs. 1 and 2).

Galium aparine. Plate XXXIV

The petals are the first of the floral organs to appear in *Galium* (Fig. 4). The sepals are late in appearing, and then do not develop very far (Fig. 5). Just below the stamens the stigmatic lobes appear and between these a convex mass grows out from the bottom of the cavity. This mass soon becomes two-lobed (Fig. 6), enlarges (Fig. 7), and develops two sporangia with their integuments (Fig. 8). With the appearance of the stigmatic lobes and the sporangia, there is an elongation of the

tissue at the base of the sporangia. This upward growth and a downward growth from the roof tissue divides the central cavity into two separate locules (Figs. 9 and 10).

Sambucus canadensis. Plate XXXIV

The origin of the floral organs is the same as the others described. The sepals grow out from the edge of the flattened mass of tissue (Fig. 11). The petal lobes appear (Fig. 12) while the receptacle is still flat (Fig. 13). The sepals elongate and enclose the other parts. Figure 14 shows the appearance of the stamens. The central cavity flattens out, so that it becomes broad and shallow (Fig. 15). The carpel lobes appear as outgrowths from the bottom of the cavity (Fig. 16). This upward growth coalesces with four lobes growing in from the inner surface of the cavity. Tissue grows down from above and meets the upward growing tissue (Figs. 1 and 2, Plate XXXV).

Valeriana officinalis. Plate XXXV

The five lobes of the petals appear at the edge of the flattened receptacle (Fig. 4). The lobes on one side of the flower grow more rapidly than the others, forming a slightly zygomorphic flower. A single stamen grows out (Figs. 6 and 7). At the same time, there is an upward growth of the tissue at the base of the petals and the stamens (Fig. 7). The sepals grow out from the outer surface of the tubular ring, and the carpel lobes form near the bottom part of the inner surface of this ring. There are then two rapid zonal elongations, one of the tissues growing up to form the ovarian cavity, and the other the tissue at the base of the sepals and stamen. The petals form a tube and the stamen is joined to the petal some distance above its point of origin.

Helianthus annuus. Plate XXXV

The details of the development of the flowers of *Helianthus* are essentially like those for *Valeriana*. The order of the succession of the parts is petals, stamens and carpels (Figs. 10-16). The sepals appear at about the same time as the carpels. There

are the two zonal elongations; one forms the tubular corolla, and the other the wall of the inferior ovary.

SUMMARY

In conclusion, some of the points observed in the study of the development of the inferior ovary in the above species of Monocotyledons and Dicotyledons are summed up in the following comparisons of the two groups:

1. The flower buds of all originate in the same manner by a protuberance of undifferentiated, meristematic tissue growing out from the flower axis. This mass flattens and broadens, and several whorls of lobes appear at the upper outer edge.

2. The order of the succession in the development of the floral parts is not the same for all the genera of the Monocotyledons, nor for all the Dicotyledons. The order, sepals, petals, stamens and carpels is found in all the Monocotyledons except *Sisyrinchium*. The same order is found in all the Dicotyledons, except *Helianthus*, *Galium* and *Valeriana*. In these the order is petals, stamens, sepals and carpels. In *Sisyrinchium*, so far as could be determined, the stamens appeared first.

3. In none of the cross sections could there be found any evidences that several floral parts had begun as separate parts and then joined together.

4. In both groups, there is evidence of adnation of parts, but this union of parts is formed by a zonal elongation of the tissue at the point of origin.

5. An examination of one similar stage of development, through the whole series of plants studied (the stage chosen was the one showing the appearance of carpels, Plate XXXVI), shows one difference between the Monocotyledons and Dicotyledons. In the Dicotyledons the lobes of the carpels grow out from the bottom of a very shallow cup, while in the Monocotyledons the carpel lobes push out from the sides of a slightly elongated cup.

6. After the appearance of the carpel lobes the annular rings in all elongate, forming the inferior ovary. In some cases, the tissues above the ovary also elongate, so that the stamens appear to branch out from the petals, and in others the petals and sepals are joined.

The differences of opinion as to the origin and development of epigyny seem to be based on the question, whether inferior ovaries are formed by a coalescence of all parts until they are liberated above, or whether the receptacle grows up in the form of a cup with the lobes of the sepals and petals coming from the rim. After making a careful study of the different plants described above, it seems that epigyny in both the Monocotyledons and the Dicotyledons develops in practically the same manner. This appears to be by a zonal elongation of the tissues just below the point of origin of the floral leaves. This elongation may begin at the appearance of the first lobes or it may not begin until the lobes of all the parts have appeared.

The origin and development is similar, with the one exception given under 5 above, whatever theory as to the nature of the wall of the ovary, is accepted, whether it has the character of the floral axis or the character of the floral leaves.

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EXPLANATION OF PLATES

The lettering for all the figures is the same:

se = sepals	pl = placenta
pe = petals	s = stamens
p = perianth	st = style and o = ovules

PLATE XXVIII

Figs. 1-12 *Sisyrinchium angustifolium* (X 68.7).

Fig. 1. The young stem tip on the left, a, young flower bud; b, bract.

Fig. 2. Longisection of a young flower bud, showing the beginning of the lobes of the stamens.

Figs. 3, 4, 5, 6 and 9. Longisections thru the center of young flower buds, showing the origin and growth of the sepals, petals, carpels and ovary cavity.

Figs. 7, 8, 10, 11. X—sections of ovary, showing central cavity in Fig. 7; the development of placenta and ovules in Figs. 8, 10, and 11; Fig. 7 about same stage as Fig. 4; Fig. 10 same as Fig. 9 and Fig. 11 as Fig. 12.

Fig. 12. Longisection of an older bud, showing the inferior ovary.

Figs. 13-15. *Gladiolus gandavensis* (X 68.7).

Fig. 13. Longisection of flower bud, s, beginning of stamen; p, perianth; b, bract.

Fig. 14. Older stage of same.

Fig. 15. Shows the beginning of carpels, c.

PLATE XXIX

Fig. 1. *Gladiolus gandavensis* (X 23.2) se, sepals; pe, petals; st, style.

Figs. 2-6. *Iris germanica*. Figs. 2, 3, 4 (X 68.7). Figs. 5 and 6 (X 38.7). All are longisections of young flowers, cut near center of bud.

Fig. 2. Young flower bud; p, perianth; b, bract.

Fig. 3. s, beginning of stamens.

Fig. 4. Further development; c, carpels.

Figs. 5 and 6. Later stages of same.

Figs. 7-10. *Freesia refracta* (X 68.7). Longisections of young flower buds. s, stamens; perianth; se, sepals. Figures numbered in order of the appearance of different parts.

PLATE XXX

Figs. 1-3. *Freesia refracta*. Figs. 1 and 2 (X 68.7).

Figs. 1 and 2. Longisections of the same flower. Fig. 1 cut thru the center and Fig. 2 at one side of the center.

Fig. 3. (X 38.7). Longisection of an older flower.

Figs. 4-9. *Musa sapientum* (X 68.7).

Fig. 4. Young flower bud.

Fig. 5. Longisection of an older flower bud.

Figs. 6-9. Longisections, showing origin and growth of parts as lettered.

Figs. 10 and 11. *Canna indica* (X 68.7).

Fig. 10. Longisections of three flower buds, a; b, bracts.

Fig. 11. L-section of one older flower. s+pe, stamens and petals; se, sepals.

PLATE XXXI

Figs. 1-5. *Canna indica*. Figs. 1-3 (X 68.7); Fig. 4 (X 19.3).

Figs. 1, 2, 3 and 4. L-sections, numbered in order of development of parts and lettered as above.

Fig. 5. Cross section of pistil (X 19.3). a, cut near top of style; b, near center; c, below b.

Figs. 6-10. *Malus ioensis* (X 31.2).

Figs. 6, 7, 8, 9. Longisections, showing the origin and development of the parts of the flower.

Fig. 10. Cross section of a young ovary. pl, placenta.

PLATE XXXII

Fig. 1. *Malus ioensis* (X 18.7). Longisection of a flower, the inferior ovary well developed.

Figs. 2-7. *Ribes aureum*. Figs. 2-5 (X 68.7); Figs. 6 and 7 (X 31.2).

Figs. 2, 3 and 6. Longisection of young flower numbered in order of development.

Fig. 4. Cross section of the ovary of Fig. 3.

Fig. 5. X-section of an older stage.

Fig. 7. X-section of pistil, stage same as Fig. 6.

a, cut at dotted line "a"; c, on dotted line

"c"; and b, cut between "a" and "b"; d, cut thru ovary, shows young ovules.

Figs. 8-13. *Fuchsia speciosa*. Figs. 8-10 (X 68.7). Figs. 11-13 (X 31.2).

Fig. 8. Longisection of young stem tip.

a, a, young flower buds on either side of stem tip.

Figs. 9 and 10. Development of sepals.

Figs. 11, 12 and 13. Appearance of sepals, petals, stamens, and carpels. Dotted lines in Fig. 12 show where petals will appear as slide is moved along.

PLATE XXXIII

Figs. 1-4. *Fuchsia speciosa*.

Fig. 1. X-section of very young ovary (X 68.7). Same stage as Plate V, Fig. 10.

Fig. 2. X-section (X 31.2) of older ovary.

Fig. 3. X-section of ovary of Fig. 4. (X 15.5).

Fig. 4. L-section of an older flower. (X 15.5).

Figs. 5-11. *Citrullus vulgaris*. Figs. 5-10 (X 68.7); Fig. 11 (X 31.2).

Fig. 5. Beginning of flower bud.

Fig. 6. Sepal lobes appearing.

Figs. 7, 8, 9, 10 and 11. L-sections of young flowers, showing the beginnings and development of sepals, petals, stamens and pistils.

Figs. 12-15. *Sanicula canadensis* (X 68.7).

Fig. 12. Beginning of flower bud, a;

Fig. 13. Three flower buds in different stages of development.

Fig. 14. Carpels have appeared.

Fig. 15. Two ovules growing up into central cavity.

PLATE XXXIV

Figs. 1 and 2. *Sanicula canadensis* (X 31.2). Longisections of flower, with inferior ovary and ovules.Figs. 3-10. *Galium aparine*. Figs. 3-8 (X 68.7). Figs. 9 and 10 (X 31.2).

Fig. 3. Longisection of young flower. Receptacle flattened.

Fig. 4. L-section; sepals and petals appear.

Fig. 5. Same. Carpels and placenta forming.

Fig. 6. Placenta two-lobed.

Figs. 7 and 8. Further development of parts, with two ovules.

Fig. 9. X-section of ovary cut at "m" Fig. 8.

Fig. 10. Same cut below Fig. 9.

Figs. 11-17. *Sambucus canadensis*. (X 68.7).

Fig. 11. L-section of fl. bud; lobes of sepals.

Figs. 12 and 13. Beginning and growth of petals.

Fig. 14. Stamens appearing.

Fig. 15. Growth of parts shown in Fig. 14.

Fig. 16. L-section. Carpel lobes just beginning.

Fig. 17. Placenta growing out from central cavity.

PLATE XXXV

Figs. 1 and 2. *Sambucus canadensis* (X 31.2).

Fig. 1. Longisection of fl; beginning of ovules.

Fig. 2. Further development of parts already begun.

Figs. 3-9. *Valeriana officinalis* (X 68.7).

Fig. 3. Longisection of two fl. buds.

Fig. 4. Two fl. buds showing different stages of development.

Fig. 5. L-sections; petals present.

Figs. 6 and 7. L-sections; carpels appearing and developing.

Fig. 8. Elongation of two regions, forming inferior ovary and corolla tube.

Fig. 9. Inferior ovary and ovules.

Figs. 10-16. *Helianthus annuus*. (X 68.7).

Fig. 10. Flower buds appearing on flattened receptacle.

Fig. 11. One flower. Lobes of petals appearing.

Figs. 12 and 13. Beginning of stamens and carpels.

Figs. 14 and 15. Further development of floral organs.

Fig. 16. L-section of inferior ovary, stamens adnate to petals.

PLATE XXXVI

Figs. 1-14. (X 68.7). L-sections of similar stages in the development of the floral leaves of the Monocotyledons and the Dicotyledons.

Figs. 1-6. Monocotyledons.

1. *Sisyrinchium angustifolium*.

2. *Gladiolus gandavensis*.

3. *Freesia refracta*.

4. *Iris germanica*.

5. *Canna indica*.

6. *Musa sapientum*.

Figs. 7-15. Dicotyledons.

7. *Malus ioensis*.

8. *Ribes aureum*.

9. *Fuchsia speciosa*.

10. *Citrullus vulgare*.

11. *Sanicula canadensis*.

12. *Galium aparine*.

13. *Sambucus canadensis*.

14. *Valeriana officinalis*.

15. *Helianthus annuus*.

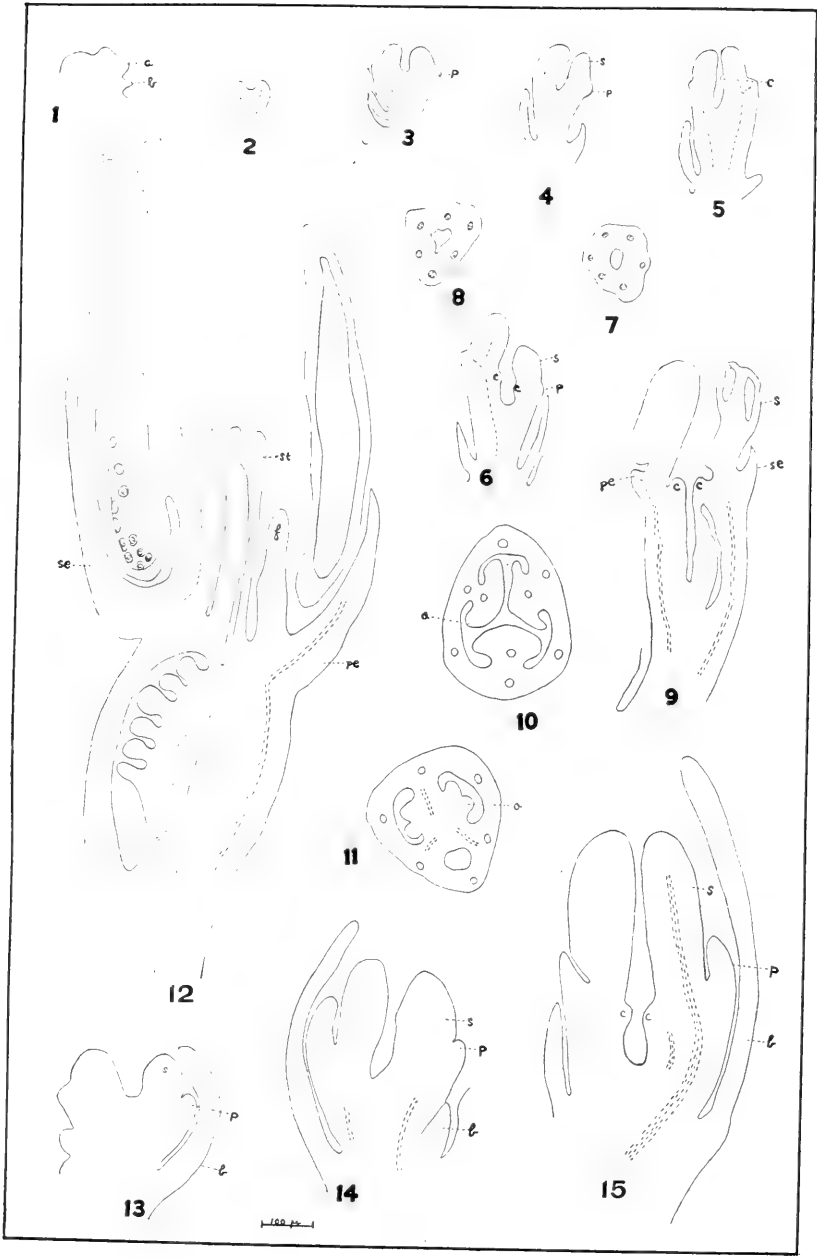


PLATE XXVIII

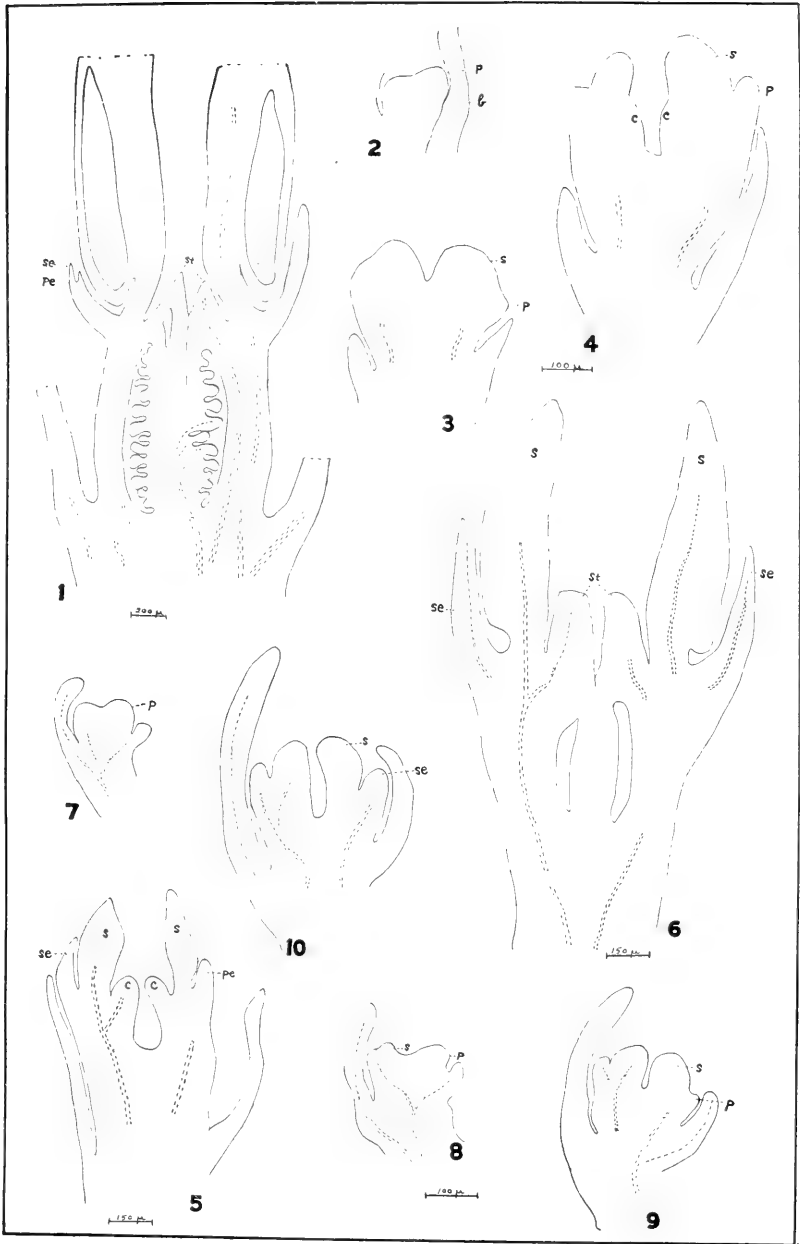


PLATE XXIX

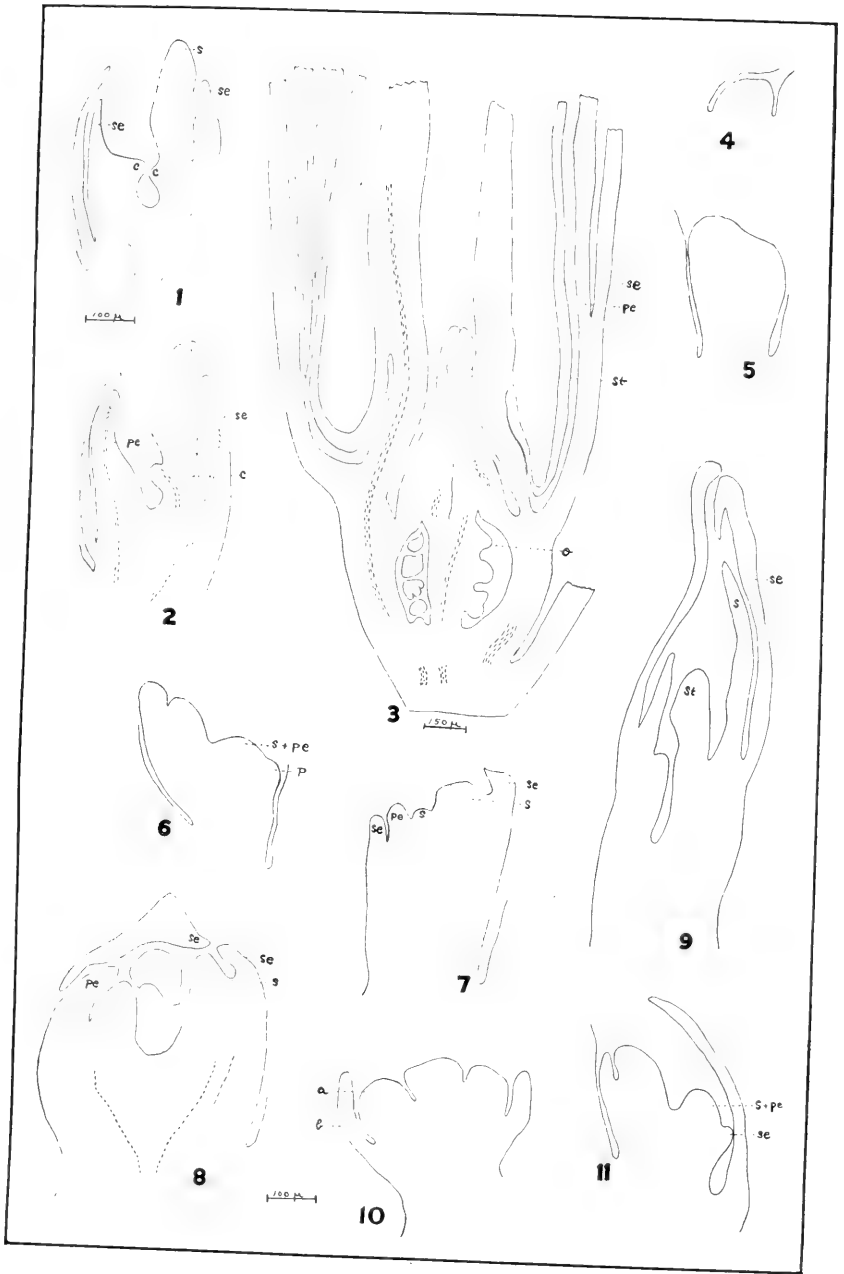


PLATE XXX

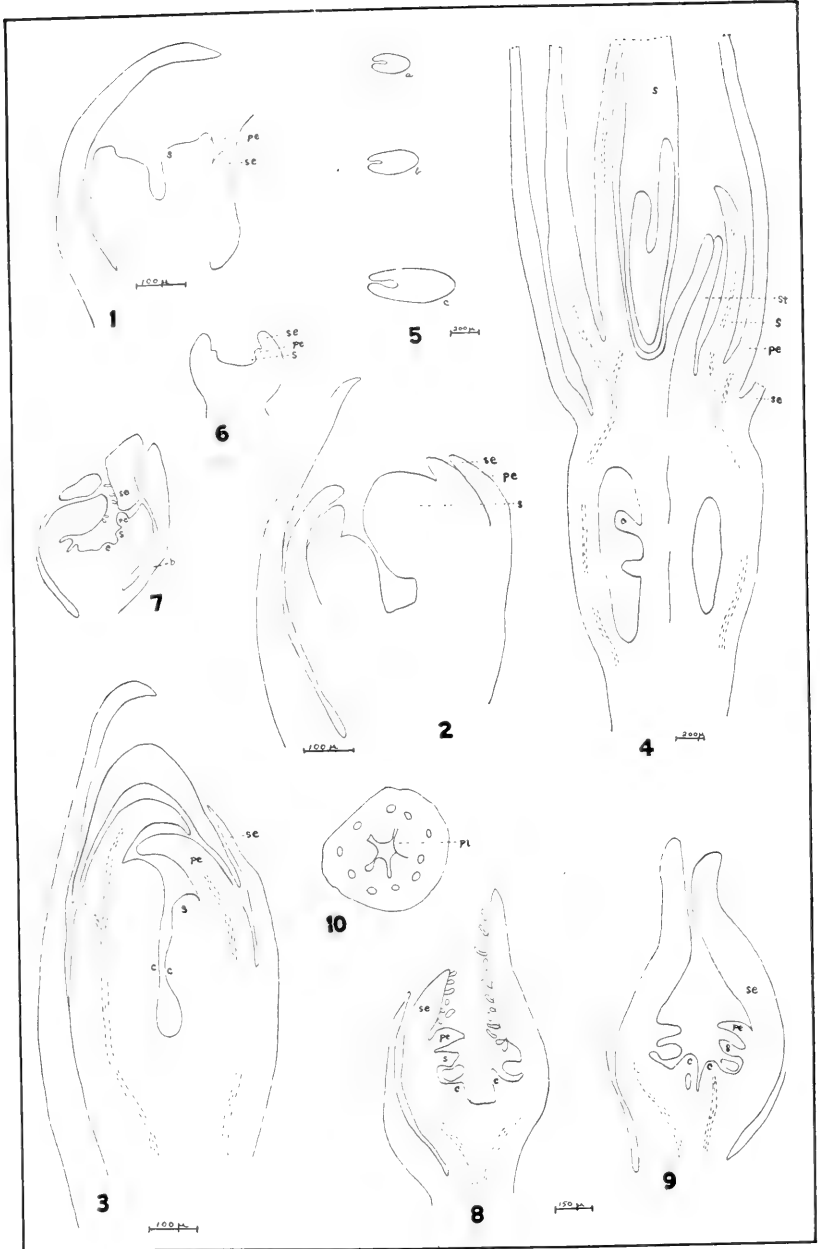


PLATE XXXI .

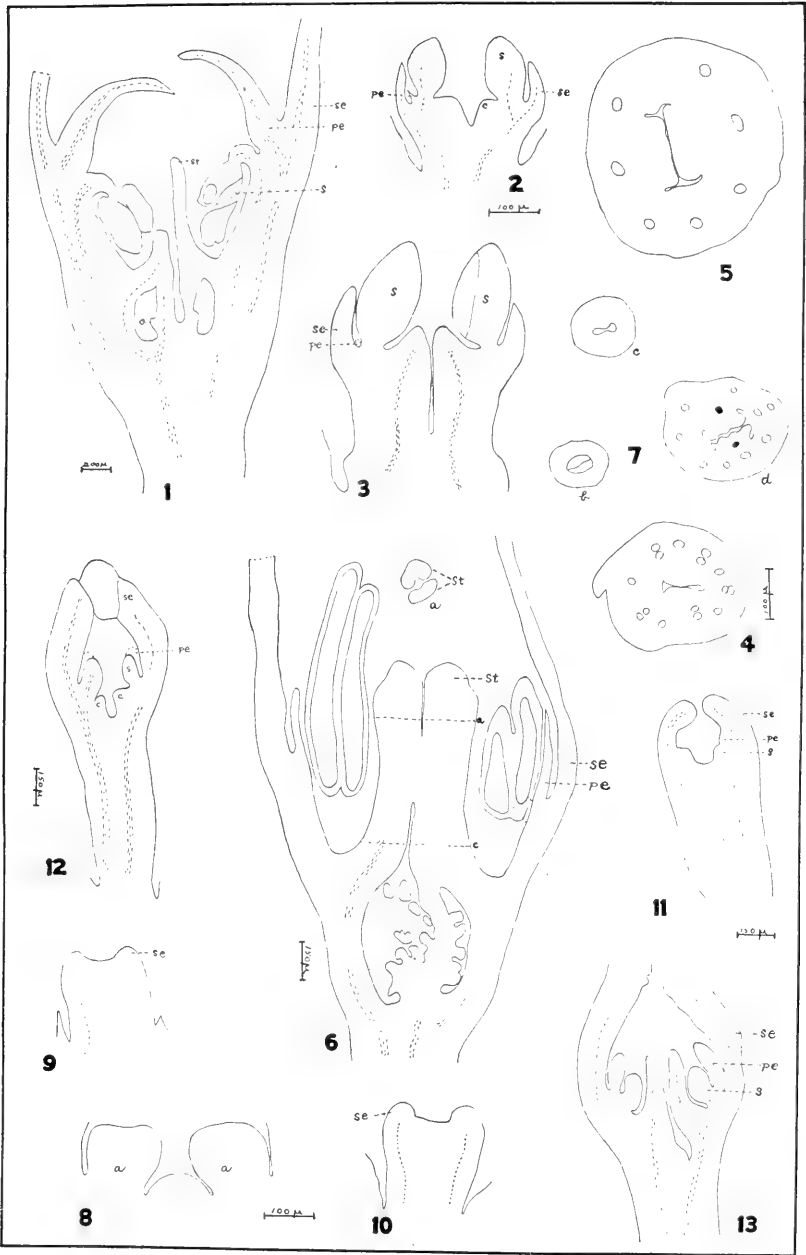


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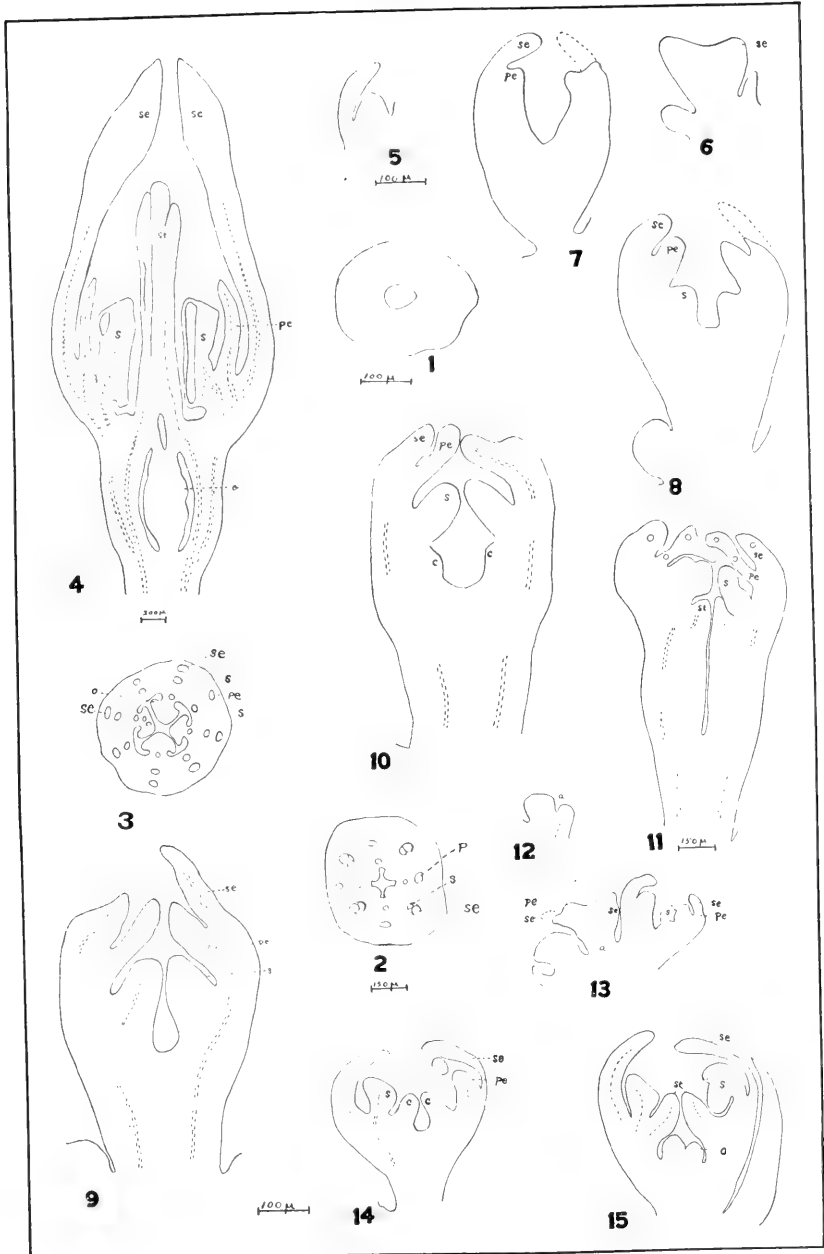


PLATE XXXIII

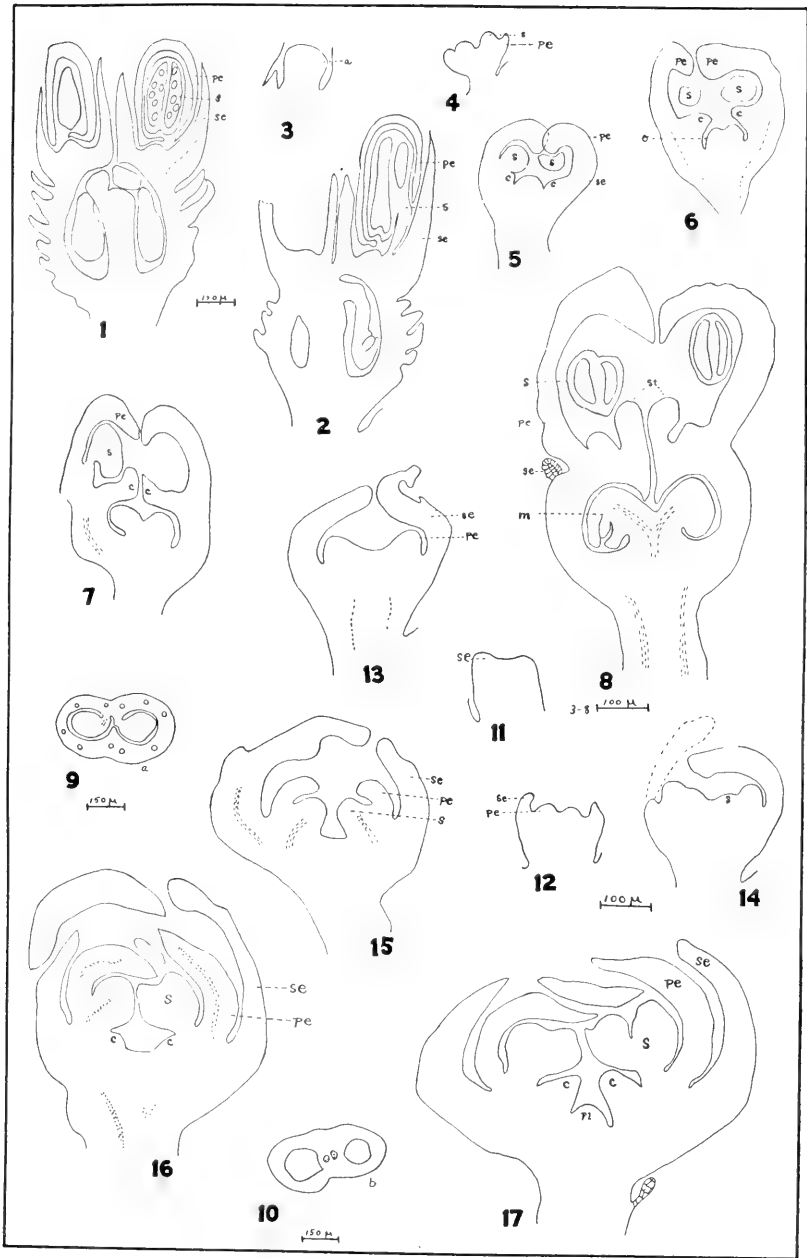


PLATE XXXIV.

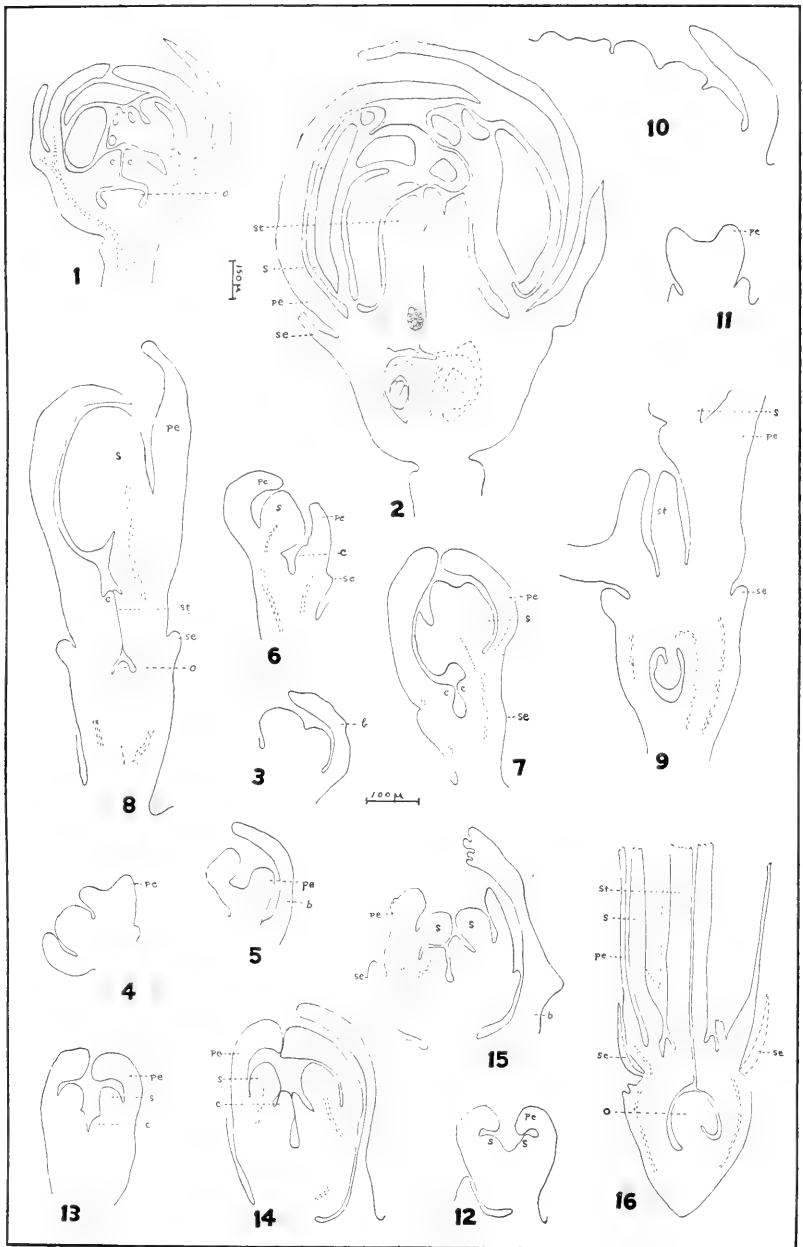


PLATE XXXV

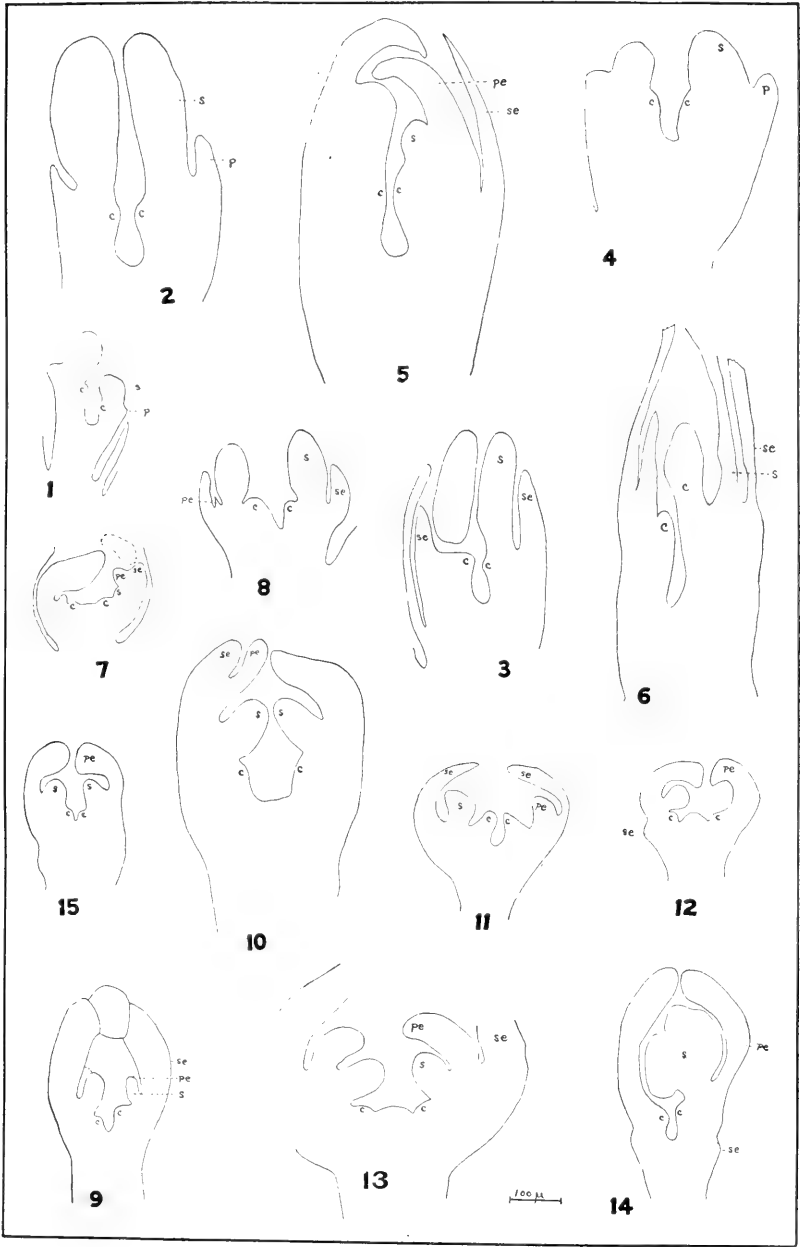


PLATE XXXVI

ACANTHOCEPHALA OF THE GENERA CENTRORHYNCHUS AND MEDIORHYNCHUS (NEW GENUS)
FROM NORTH AMERICAN BIRDS*

H. J. VAN CLEAVE

INTRODUCTION

The writer has made a thorough study of the avian Acanthocephala in the collections of the U. S. Bureau of Animal Industry. In this study a number of specimens comprising four undescribed species have been discovered in which the proboscis receptacle finds its insertion near the middle of the proboscis wall. This method of insertion is characteristic of but a single known genus of Acanthocephala. The morphology of the four newly discovered species possessing a proboscis receptacle of this type varies too broadly in points of fundamental structure to permit of including all four species within the Genus *Centrorhynchus*. But one species of the four, and that represented by a single individual in the collection under consideration, agrees with the characters of the genus *Centrorhynchus* as given by Lühe (1911:41). For the other three species the writer has found it necessary to create a new genus, the characters of which are enumerated in another part of this paper.

METHODS

The specimens for study have been stained in toto with Ehrlich's acid hematoxylin, dehydrated, cleared in synthetic oil of wintergreen, and mounted in damar. All the drawings have been made with a camera lucida.

GENUS *CENTRORHYNCHUS* Lühe 1911

The genus *Centrorhynchus* comprises a well defined group of parasitic worms, belonging to the Class Acanthocephala, which reach maturity in the alimentary canal of birds. One of the most striking characteristics of this genus is the insertion of the proboscis

*Contributions from the Zoological Laboratory of the University of Illinois, No. 76.

receptacle in the middle of the proboscis wall. This peculiar departure from the ordinary arrangement of the organs and parts of the body, while not the sole means of distinguishing the members of this genus, has led to considerable confusion among various workers. The attempt on the part of some investigators to homologize the basal region of the proboscis with the neck characteristic of some of the other genera of Acanthocephala has been most distinctly refuted by Lühe (1912:274). He has shown that if the insertion of the proboscis receptacle marks the boundary between neck and proboscis the genus *Gigantorhynchus* must be considered as having no true proboscis for, according to Lühe's observations, the proboscis receptacle in the genus *Gigantorhynchus* finds its insertion at the tip of the organ of fixation, which, according to the proposed distinction, would of necessity be considered an armed neck. The folly of this argument is self evident. There is no reason for doubting that the organ of attachment in the genus *Gigantorhynchus* is a true proboscis. Similarly the spined regions both anterior to and posterior to the insertion of the proboscis receptacle constitute the proboscis of the *Centrorhynchi*.

Little has been done toward establishing the synonymy within the genus *Centrorhynchus*. Lühe (1911:41) has listed the forms from central Europe which might be attributed to it but has added the statement that at least some of the names included in his list are certainly synonyms. Kostylew (1914:186) has given a list of four species which he considered as valid for this genus but has not presented the data used in reaching his conclusion.

Of the species attributed to the genus *Centrorhynchus* but a single one has been reported from North America. Leidy (1888:22) has recorded the occurrence of individuals by him determined as "*Echinorhynchus caudatus* Zeder" from the swallow tail kite, *Elanoides forficatus* (*Elanoides furcatus*)* and two specimens of the same species from the owl *Scotiapterx nebulosa* (*Strix nebulosa*). The specific identity of these specimens with the European species must be sharply questioned, for, as earlier works of the writer have shown, the Acanthocephala of North America, and

*Here, and elsewhere in the text, scientific names of birds quoted from another writer or taken from records accompanying the collections follow in parenthesis after the name given for the species in the A. O. U. Check List.

especially those from fresh-water and terrestrial hosts, constitute a list of forms in the main peculiar to the American continent. A fuller account of the evidences of this development is given at the end of this paper.

On the basis of the present study it becomes impossible to determine whether the Acanthocephala described by Leidy belong to the Centrorhynchi or the genus Mediorhynchus. The only valid record of a species of Centrorhynchus from North America is in a single collection of one individual in the Collections of the Bureau of Animal Industry. A comparison of this specimen with descriptions of other species of the genus has revealed differences which make it necessary to consider this a new species. The specific diagnosis follows.

CENTRORHYNCHUS SPINOSUS NOV. SPEC.

(Figs. 1-3)

Specific diagnosis. With the characters of the genus. Proboscis closely set with numerous hooks. Species description based on a single female which becomes type. Body 20 mm. long; diameter anterior part of body slightly larger (6.6 mm.) than posterior part (0.5 mm.); posterior extremity slightly pointed, conical. Proboscis 0.65 mm. long; with hooks of two distinct types, those anterior to insertion of proboscis receptacle with recurved roots; strongest hooks (0.038 mm. long) appearing near the middle of the proboscis just behind the cylindrical apical portion. Hooks in thirty-two longitudinal rows of about twenty-four hooks each. Hooks posterior to insertion of proboscis receptacle thornlike, about 0.05 mm. long, often with recurved tips. Proboscis constricted at insertion of proboscis receptacle, diameter at constriction 0.25 mm.; posterior portion (0.35 mm. in diameter) not sharply set off from body proper; slightly swollen anterior to insertion of receptacle; tip smaller, cylindrical, 0.17 mm. in diameter.

Type host *Herodias egretta* (*Ardetta egretta*), in intestine. Collected by Hassall, Sept. 1894. Type deposited in the U. S. Bureau of Animal Industry Helminthological Collections. Catalog number Hassall Collection 6307.

In general body form this species closely resembles the figures given by Lühe (1911 Fig. 54) for *C. aluconis*. Unfortunately the

male of *C. spinosus* is unknown so a comparison of the male organs with those of *C. aluconis* must be deferred until other materials are available. One of the most strikingly characteristic points about this species is the abundance of the spines (see Figs. 1 and 2). The entire proboscis, even under a low power of the microscope, fairly bristles with projecting spines. While the hooks of the anterior part of the proboscis are stronger than those posterior to the insertion of the proboscis receptacle (see Fig. 3) the difference in size is not as conspicuous as in the species of *Mediorhynchus* described in this paper.

MEDIORHYNCHUS NEW GENUS

The remaining three species differ from the characterization of the genus *Centrorhynchus* in the following particulars: (1) The males possess eight rounded or pear shaped cement glands instead of the three long tubular cement glands described for *Centrorhynchus*. (2) The wall of the proboscis receptacle is composed of a single muscular layer instead of two layers as specified by Lühe for *Centrorhynchus* and shown in figure 2 of *C. spinosus*. (3) The invertors of the proboscis pass through the sides of the proboscis receptacle considerable distance from its base and continue backward through the body cavity as the retractors of the proboscis receptacle, while in *Centrorhynchus* the invertors pass through the wall of the receptacle at its rounded posterior extremity (see Fig. 10).

Generic diagnosis. Acanthocephala of medium size reaching sexual maturity in the alimentary canal of birds. Proboscis receptacle inserted near the middle of the proboscis wall. Receptacle a single walled muscular sac with invertors of proboscis passing through its wall some distance anterior to the posterior tip of the receptacle. Central nervous system near the center of the proboscis receptacle between the invertor muscles. Cement glands of male a compact mass of rounded or pear shaped glands, usually eight in number. Proboscis hooks of two distinct types; those anterior to the insertion of the proboscis receptacle in surface view with flask shaped roots, bases of roots broad; those on the posterior portion of the proboscis without reflexed roots. Embryos with three concentric membranes.

Descriptions of three species belonging to the genus *Mediorhynchus* follow:

MEDIORHYNCHUS PAPILLOSUS NOV. SPEC.

(Figs. 4-10)

Specific diagnosis. With the characters of the genus. Proboscis armed with inconspicuous hooks, each hook embedded in a papilla (see Figs. 5 and 8.). Body of both sexes cylindrical, almost uniform diameter throughout. Type male 9.3 mm. long; diameter 0.75 mm., tapering to 0.57 mm. at base of proboscis, and tapering slightly at posterior extremity. Cement glands (see Fig. 7) eight, rounded to pyriform, in compact mass slightly posterior of posterior testis. Testes two, elongated, elliptical, contiguous; in chief axis of body. Proboscis 0.65 mm. long; largest diameter 0.30 mm., near the middle. Lemnisci 3 mm. long.

Type female 18 mm. long, diameter 0.75 mm., tapering to 0.57 mm. at base of proboscis. Embryos 0.038 to 0.047 mm. long by 0.018 to 0.024 mm. across, with three concentric membranes (see Fig. 9.).

Proboscis anterior to insertion of proboscis receptacle with eighteen longitudinal rows of six or seven hooks each, longest hooks 0.027 mm. long, each hook with a root process (0.040 mm. long) usually longer than the recurved spine (see Fig. 6.). Surface view of roots pyriform (see Fig. 8.). Hooks posterior to insertion of proboscis receptacle without reflexed roots; thornlike, with tips bent posteriorly almost at right angles to axis of the spine; four to six hooks in a longitudinal row.

Host *Myiochanes virens* (*Contopus virens*). Collected May 30, 1892, by Albert Hassall. Type male and type female deposited in the U. S. Bureau of Animal Industry Helminthological Collection. Catalog number 6320 Hassall Collection.

Besides the types of *M. papillosus* the collections of the U. S. Bureau of Animal Industry contain one specimen of this species, a male from the intestine of *Porzana carolina* in the Hassall collection, catalog number 6303. The locality from which this individual was taken is not given in the records accompanying the specimens.

MEDIORHYNCHUS GRANDIS NOV. SPEC.

(Figs. 11-14)

Specific diagnosis. With the characters of the genus. Specific description based upon the study of one male and three fully mature females, constituting two collections from different hosts. Body of females 27 to 35 mm. long, practically cylindrical; diameter anterior region 1.2 mm.; maximum diameter considerable portion in middle of body 0.9 to 1.4 mm.; diameter near posterior extremity 0.7 to 1.1 mm. Anterior and posterior extremities of body flexed ventrally. Anterior part of body just behind proboscis expanded and sharply set off from neck. Proboscis of all specimens partially inverted; largest female anterior to insertion of proboscis receptacle approximately 0.6 mm. long if extended (all but 0.3 mm. inverted in type); proboscis posterior to insertion of receptacle 0.6 mm. long. Hooks in anterior region of proboscis with massive roots; twelve longitudinal rows of approximately four hooks each (two in each row show on surface of inverted proboscis). Basal portion of proboscis with numerous small spines, not in perfect rows but about thirty longitudinal rows with three to six spines each. Hooks on anterior proboscis 0.05 mm. long (measurement taken as longest straight line from the tip of the spine to the region where the hook and root join). Length of root 0.075 to 0.086 mm. (from base of root to top of angle between root and hook). Embryos with three concentric membranes, about twice as long as broad; 0.043 by 0.021 mm.

Male 8.2 mm. long. Maximum diameter 1 mm; diameter posterior tip 0.18 mm.; anterior to tip 0.5 mm. Diameter anterior end of the body proper 0.61 mm. Lemnisci about 2 mm. long. Testes oval, slightly separated, 1.2 mm. long and 0.35 mm. wide. Eight cement glands, each rounded, usually pear shaped.

Type host *Quiscalus quiscula*, in intestine. Cotypes deposited in U. S. Bureau of Animal Industry Helminthological Collection. Catalog number Hassall Collection 6319.

One fully mature female determined by the writer as belonging to this species has been found in the intestine of *Sturnella magna*. Collected by C. S. Brimley, Nov. 30, 1902, in North Caro-

lina. In the U. S. Bureau of Animal Industry Parasitological Collection. Catalog number 6772.

MEDIORHYNCHUS ROBUSTUS NOV. SPEC.

(Figs. 15 and 16)

Specific diagnosis. With the characters of the genus. Body both sexes robust, medium length; largest diameter near the middle, tapering slightly toward either end; dorsal surface slightly convex. Proboscis small, setting on obliquely truncated anterior end of body, pointing slightly ventrad. Lacunar system of subcuticula highly developed. Lemnisci about one-fourth to one-third the length of body. Specific descriptions based on one male and one female which become types.

Type female 16 mm. long. Maximum diameter body proper 2.4 mm., diameter posterior extremity 0.8 mm., anterior extremity 0.9 mm. Proboscis short, globular, 0.2 mm. in diameter; tip partially inverted and base partially retracted within body; exposed portion anterior to insertion of proboscis receptacle armed with twenty-four longitudinal rows of hooks. Hooks very small and inconspicuous with small pyriform roots 0.032 mm. long. Each hook in an elevation of the proboscis wall but not in a distinct papilla. Basal region of proboscis entirely retracted within body, hooks not observable.

Type male 7 mm. long, maximum diameter 1.25 mm.; diameter posterior extremity 0.52 mm.; anterior extremity the same. Proboscis short, globular, 0.156 mm. in diameter, partially inverted. Hooks small, inconspicuous, about 0.025 mm. long; only about 0.005 mm. extending beyond the elevations on proboscis.

Embryos 0.038 mm. long by 0.016 mm. wide.

Type host *Icteria virens* in intestine. Collected by A. Hassall at Washington, D. C., in June 1893. Type male and female deposited in the Helminthological Collection of the U. S. Bureau of Animal Industry, catalog number 2316.

FAMILY CENTRORHYNCHIDÆ

When Hamann (1892:195-197) first pointed out the presence within the Acanthocephala of distinct groups and showed the necessity of recognizing a number of genera to displace the old all

inclusive genus *Echinorhynchus* he founded three families; the Echinorhynchidæ, the Gigantorhynchidæ, and the Neorhynchidæ. The latter by recent change in name of the type genus becomes Neoechinorhynchidæ. Within each of these families he recognized a single genus. Since his work on this subject a long list of Acanthocephalan genera has been created but usually with the creation of a new genus the founder has neglected to point out the affinities of the genus. As a result today there are few references to groupings of genera into larger groups or families. As a start toward organizing this part of the system the writer proposes that since the genera *Centrorhynchus* and *Mediorhynchus* have the same type of proboscis receptacle and both reach sexual maturity in the alimentary canal of birds, they should be united to form a family which should take the name *Centrorhynchidæ* from the name of the oldest genus.

Family Centrorhynchidæ. Diagnosis. With the characters of the Class Acanthocephala. Living as mature adults in the alimentary canal of birds. Proboscis receptacle inserted near the middle of the proboscis wall.

THE AMERICAN ACANTHOCEPHALA AS A DISTINCTIVE FAUNA

The development of a typical American fauna among the Acanthocephala, independent from and in time of relatively remote separation from the European fauna, is evidenced in the extent to which the American representatives of a genus differ from the European representatives upon which by far the greater amount of work has been done. In many cases a generic diagnosis based entirely upon European representatives of a genus fails in some points of detail to permit of the inclusion of American species subsequently discovered. In the genus *Neoechinorhynchus* the two species *N. gracilientis* (Van C.) and *N. longirostris* (Van C.) show structures and hook arrangement at considerable variance from the conditions typical of described European forms. On the other hand the three species of the genus, *N. emydis* (Leidy), *N. cylindricus* (Van C.) and *N. tenellus* (Van C.), closest in their affinities to the European species agree among themselves in possessing eight syncytial cells in the cement gland. Bieler (1913:235) has called attention to the fact that the two generally recognized Euro-

pean species *N. rutili* (Müll.), the common fresh-water species of Europe, has twelve nuclei in the cement gland, while *N. agilis* (Rud.), the marine representative of the genus, possesses eight nuclei in the cement gland of the male. These with other differences in structure of European and American Neoechinorhynchi would tend to indicate that the fresh-water representatives of the genus have either had (a) an independant origin on the two continents or (b) each developing from marine species of the genus at one time common to the two continents they have been separated a sufficient length of time to allow distinctive characteristics to develop in the two groups. Of these possibilities the latter seems the more plausible.

In the genus *Filicollis* the writer (1916:132) found it necessary to suggest a modification of the original characterization of the genus in order that *F. botulus* Van C. be admitted to a position near to *F. anatis* (Schrank) which general body structure and hosts of the two parasites demanded. Similarly in the genus *Arhythmorhynchus* the writer (1916a:169) has described two North American species which, though agreeing with the European species in fundamental generic characteristics, differ from them in some points of detail of structure.

Finally, in the Centrorhynchidæ the discovery of a new genus from North America adds still further evidence of the trend in North America toward the development of a distinctive Acanthocephalan fauna.

SUMMARY

A study of avian Acanthocephala in the Collections of the U. S. Bureau of Animal Industry has revealed four new species, one belonging to the genus *Centrorhynchus*, and three with characters such as to prevent inclusion in any known genus. For the latter a new genus, *Mediorhynchus*, has been created.

The two genera *Centrorhynchus* and *Mediorhynchus* agree in the method of insertion of proboscis receptacle, and in the fact that both occur as parasites in the alimentary canal of birds. They differ in the size, shape, and number of the cement glands of the

male; in the structure of the wall of the proboscis receptacle; and in the relations of the invertors of the proboscis to the proboscis receptacle.

Upon the basis of agreement the writer has suggested the establishing of a new family Centrorhynchidæ to include these two genera.

Evidence has been assembled to show the tendency toward the development of an Acanthocephalan fauna peculiar to terrestrial and fresh-water hosts of the North American Continent.

A key to the species of Centrorhynchidæ from North American birds is given.

KEY TO THE SPECIES OF THE FAMILY CENTRORHYNCHIDÆ FROM NORTH AMERICAN BIRDS

Acanthocephala parasitic in birds, with the proboscis receptacle inserted near the middle of the proboscis wall.—Family Centrorhynchidæ.

1. (2) Proboscis receptacle a two layered muscular sac, cylindrical, with the invertors of the proboscis passing through the posterior rounded tip and continuing backward through the body cavity as the retractors of the proboscis receptacle.—Genus Centrorhynchus. A single species, *C. spinosus*, reported from North America.
2. (1) Proboscis receptacle a single layered muscular sac with the retractors of the proboscis receptacle passing from its sides some distance anterior to the posterior tip. Receptacle not cylindrical in form. Genus *Mediorhynchus*3
3. (6) Anterior and posterior regions of proboscis with the same number of longitudinal rows of hooks.....4
4. (5) Twenty-four longitudinal rows of hooks on proboscis. Maximum diameter of body : length of body :: 1 : 5 (or 6).
.....*M. robustus*
5. (4) Eighteen longitudinal rows of hooks on proboscis. Maximum diameter of body : length of body :: 1 : 9....*M. papillosus*
6. (3) Twelve longitudinal rows of hooks on anterior region of proboscis; thirty on posterior region.....*M. grandis*

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EXPLANATION OF PLATES

Abbreviations used:

- | | |
|---|--|
| <i>b.</i> brain | <i>l.</i> lemnisci |
| <i>c. g.</i> cement gland | <i>p. r.</i> proboscis receptacle |
| <i>e.</i> egg mass | <i>r. p.</i> retractor of proboscis receptacle |
| <i>ins.</i> point of insertion of proboscis | <i>t. a.</i> anterior testis |
| receptacle on proboscis wall | <i>t. p.</i> posterior testis |
| <i>i. p.</i> inverter of proboscis | |

PLATE XXXVII

Centrorhynchus spinosus nov. spec.

- Fig. 1. Type female showing body form and general arrangement of parts.
 Fig. 2. Proboscis and anterior region of body, showing also insertion of proboscis receptacle and location of the retractors of the receptacle with reference to the wall.
 Fig. 3. Profile of the same proboscis showing a single longitudinal row of hooks.

PLATE XXXVIII

Mediorhynchus papillosus nov. gen. et nov. spec.

- Fig. 4. Entire male from intestine of *Prozana carolina* showing arrangement of organs.
 Fig. 5. Proboscis and anterior region of body, surface view, of type male from *Myiochanes virens*.
 Fig. 6. Profile of proboscis shown in figure 5, showing a single longitudinal row of hooks.
 Fig. 7. Posterior end of body of male in optical section showing especially the shape, number, and arrangement of the cement glands characteristic for the genus *Mediorhynchus*.
 Fig. 8. Surface view of roots and papillæ from proboscis.
 Fig. 9. Embryos from body of mature female.

PLATE XXXIX

- Fig. 10. *M. papillosus*. Optical section of proboscis and anterior region of body showing attachment and structure of the proboscis receptacle and course taken by invertors of the proboscis through the wall of the receptacle.
 Figs. 11, 12, 13, and 14 of *Mediorhynchus grandis* nov. spec.
 Fig. 11. Anterior region of body of female.
 Fig. 12. Proboscis of same specimen. In surface view the delicate spines on the posterior region of the proboscis are discernable only as small circular markings.
 Fig. 13. Profile of proboscis showing single longitudinal row of hooks.
 Fig. 14. Embryos from body cavity of female.
 Figs. 15 and 16. *Mediorhynchus robustus* nov. spec.
 Fig. 15. Male in optical section showing arrangement of organs.
 Fig. 16. Embryos from body cavity of female.

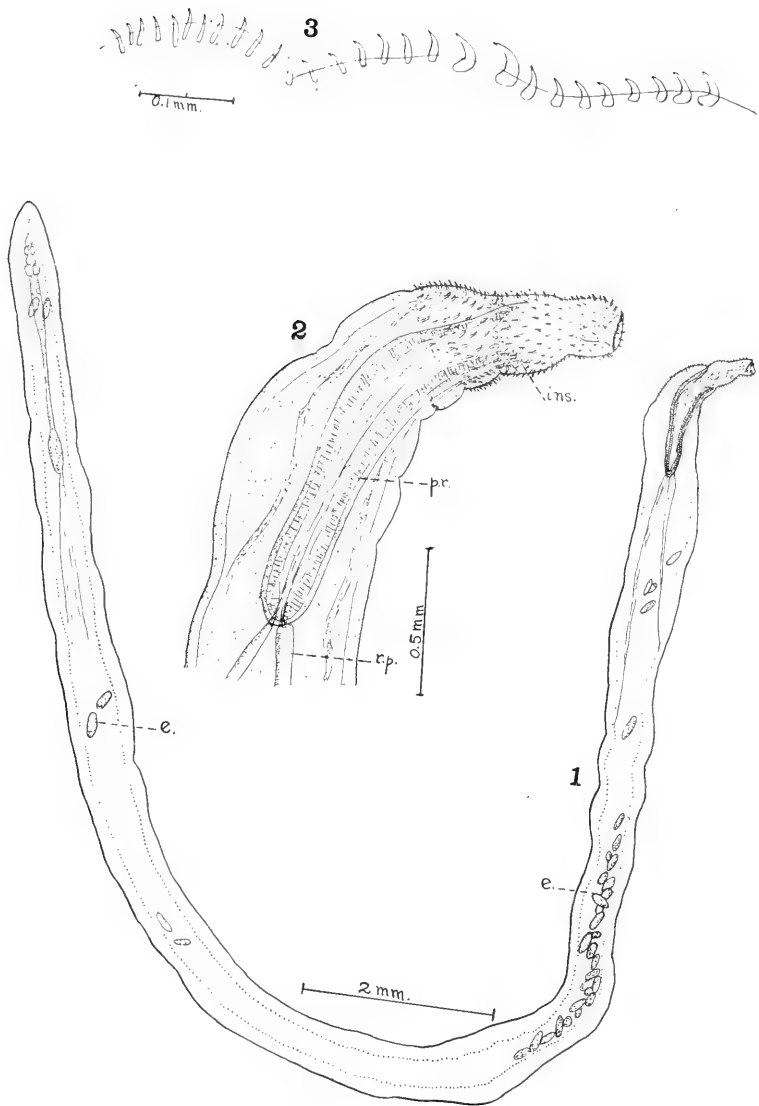


PLATE XXXVII

H. J. VAN CLEAVE

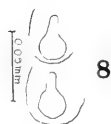
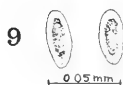
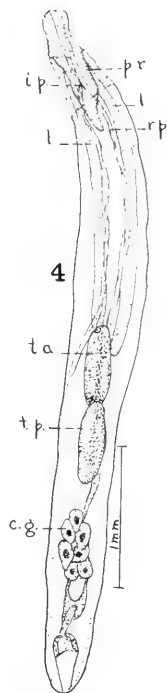
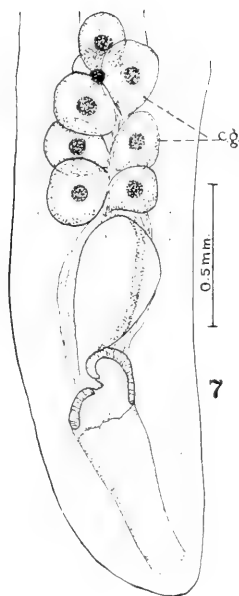
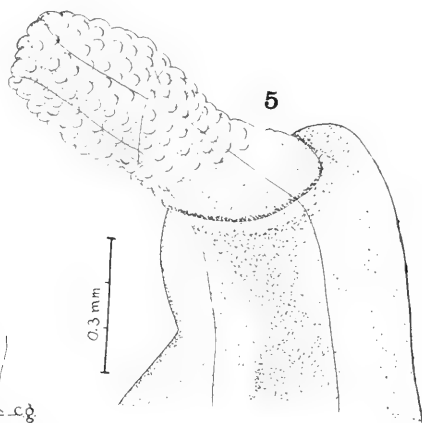


PLATE XXXVIII

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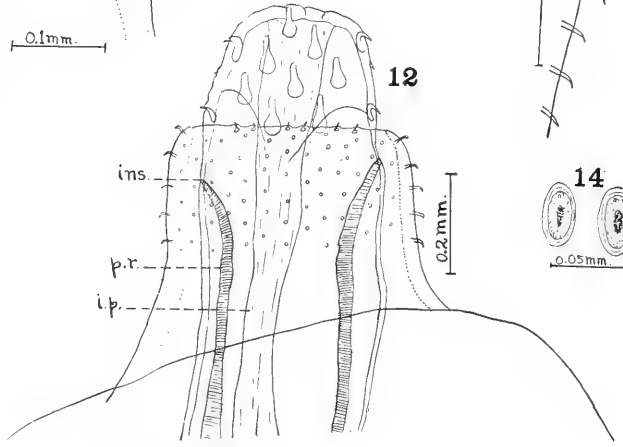
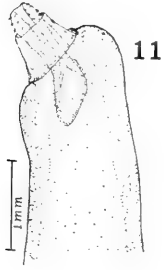
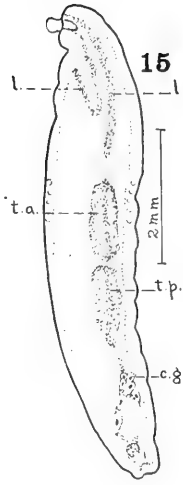
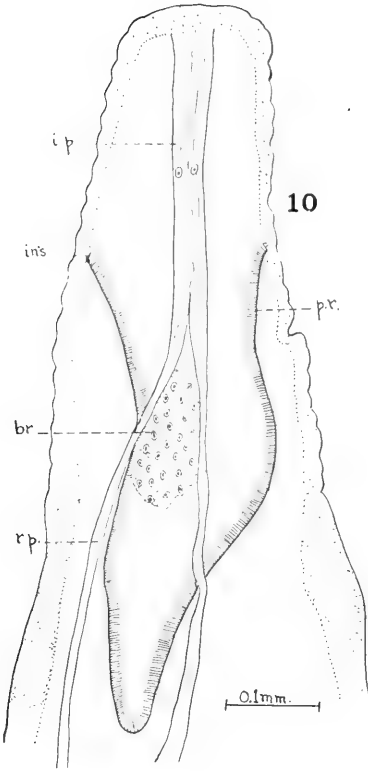


PLATE XXXIX

H. J. VAN CLEAVE

THE GENUS ASPIDISCA EHRENBERG

HAROLD H. PLOUGH

The following systematic account of the genus *Aspidisca* was undertaken with the idea that at some future time the writer would use one of the species for a series of culture experiments. In order to do this intelligently an extensive study of the literature of the genus was suggested by Prof. G. N. Calkins. This study had disclosed one or two rather obvious mistakes in the recent literature, and showed the need of a new systematic account of the genus. The confusion in a relatively simple and homogeneous group like the *Aspidiscas* indicates that considerable work still remains to be done in systematizing the whole group of Hypotrichs.

ASPIDISCA EHRENBERG 1830

O. F. Muller 1773, 1780, 1786, 1788. Bory 1824. Ehrenberg 1830, 1831, 1833, 1838. Dujardin 1841. Perty 1852. Claparede at Lachmann 1857. Stein 1859. Fresenius 1865. Diesing 1865-6. Quennerstedt 1867-8. Fromental 1874. Mereskovsky 1878. Kent 1881. Rees 1884. Fabre-Domergue 1885. Perejaslawzewa 1886. Gourret et Roesser 1886, 1888. Calkins 1902. Hamburger und Buddenbrock 1911.

The genus *Aspidisca* was originally separated off from O. F. Muller's *Trichoda* by Ehrenberg 1830 and placed in a separate family *Aspidiscina*. The family was given the following rather fanciful characterization: "animaux polygastriques à carapace, ayant un canal intestinal distinct à double orifice, dont seulement celui de l'anus est terminal." Under Bory de St. Vincent's name *Cocculina* Dujardin 1841 and afterward Perty described several species which are probably *Aspidiscas*, though the fact can be established only in the case of Dujardin's *Cocculina costata*. The principle character which distinguishes the genus *Cocculina* from his other *Ploesconien* was the absence of a mouth. This does not stand for as Claparede et Lachmann 1857 first showed the mouth is at the base of a peristomial field of cilia on the left side and between the upper and lower plates of the cuirass. These investigators dis-

tinguished *Aspidisca* from the other genera in their family Oxytrichina by the absence of frontal cirri. Stein 1859 again made of the genus a separate family under Ehrenberg's name and considered that it represented a connecting link between his family Chlamydodonta (*Chilodon*, *Ergvilia* etc.) and the Euplotina. Kent 1881 recognized that the main point of difference between this genus and the remainder of his family Euplotidae was "in the more rearward location of the peristome field and consequent non-projection of the adoral fringe of cilia beyond the lateral border," and included the genus in that family. This classification is generally accepted. The location of the peristome field as above described and the absence of marginal cirri are sufficient to distinguish the genus from the other genera of the family.

Description of the Genus *Aspidisca* Ehr. 1830.

Synonymy: *Trichoda* p p—Muller 1773, 1780, 1786, 1788.

Tribulina? *Ratulus*? p p—Bory 1824.

Oxytricha? p p *Euplotes* p p *Loxodes plicatus* ??—Ehrenberg 1838.

Coccudina p p—Dujardin 1841, and Perty 1852.

Onychaspis—Diesing 1856.

Monostylus ??—Perejaslawzewa.

Animalcules ovate, small, rigid, encuirassed, with a convex dorsal and a plain ventral surface; left side nearly straight, right sharply convex; the right border having a thickened margin; peristome limited to the left ventral side where it forms a small depression which may or may not just reach—but in no case extends beyond—the anterior border; associated with it is a simple fringe of adoral cilia which do not extend beyond the border; the ventral surface on the left side extends to a greater or less degree toward the right under the peristome, so that the latter is located in a pocket between the upper and lower surface; several large claw-like styles toward the anterior end and in the center of the ventral region; a variable number of posterior or anal styles arranged in a single row just inside the posterior ventral margin; anal aperture placed far back, debouching a little in advance of the anal styles. Cosmopolitan, fresh and salt water.

The number and position of the ventral and anal cirri has thus far been considered the important diagnostic character of the species of *Aspidisca*. Yet *A. polystyla* Stein was described as having from ten to twelve anal cirri, and a variety described by Calkins 1902 from Woods Hole has thirteen anal and eight rather than seven ventral cirri—which number had not previously been

exceeded in the genus. Calkin's variety of *A. hexeris* Quenn. has also eight ventral cirri, while *A. Andrewii* Meresch.=*A. hexeris* has the seven very differently arranged. Fresenius 1865 described *A. leptaspis* with five anal styles, but Rees 1884 showed that one of these often frays out into three, making seven in all. Fabre-Domergue 1885 described this seven styled form as a new species *A. crenata*. It appears then that the number and position of the cirri is to a large degree variable, and that they must be discarded as of importance in differentiating species. A re-examination of the genus discloses that the species can be readily distinguished by the character of the stiff cuirass. This may be smooth or serrated dorsally, and the left border may be plain or incised so as to produce a varying number of posteriorly directed spurs. The form of the stiff cuirass is apparently extremely constant within the genus, and a few minutes observation of it will generally suffice to identify the species. In view of this fact it appears that *A. lynceus* and *A. polystyla* are really very similar, and further work may disclose that they are one and the same species. The latter of these species was placed in a separate subgenus—*Onychaspis*—by Stein and a separate genus by Diesing on the basis of its larger number of styles, yet one might easily be derived from the other by the continuation of the fraying out process which has been noted above in *A. leptaspis*. From the simple smooth bordered forms like *A. lynceus* it is possible that the spurred and serrated forms have been derived. On the basis of the form of the cuirass a simple key for the quick identification of the species of *Aspidisca* may be constructed.

KEY FOR THE IDENTIFICATION OF THE SPECIES OF ASPIDISCA

- A. Right and left border smooth.....a
- B. Left border incised to form a single backwardly directed spur in the posterior third.....A. *hexeris*
- C. Left border with two spurs, one in the anterior and one in the posterior third.....c
- D. Left border with three spurs.....A. *sedigita*
- a. Dorsal surface with recurved thorn-like appendage...*A. *turrita*
 Dorsal surface without thorn.....a'
- c. Dorsal surface and posterior border serrated.....A. *leptaspis*
 Dorsal surface smooth.....A. *lynceaster*
 a'. Ventral plate projecting beyond left border of carapace*A. *costata*
 Ventral plate not projecting.....a"
 a". Peristome reaching anterior border, anal cirri 5.....A. *lynceus*
 Peristomial cilia not reaching anterior border, anal cirri more than 5.....A. *polystyla*
- A. Right and left border smooth.

A. *lynceus* (Muller).

Trichoda lynceus Muller 1773, 1780, 1786, 1788.

Ratulus lynceus ? Bory 1824.

Aspidisca lynceus Ehrenberg 1838.

Coccludina crassa ? Dujardin 1841.

Aspidisca lynceus Claparede et Lachmann 1867 t6.

Body ovate, widest and somewhat truncate posteriorly; the marginal border of the carapace entirely even, the left one straight, the right one somewhat convex; the dorsal surface smooth, or marked longitudinally with three feeble furrows; the inferior surface bearing 7 ventral styles in two rows—anterior with 4, posterior with 3- 5 anal styles. L—1/540". Salt water.

Distribution: Baltic (Ostsee) at Hapsal (Eichwald), Warberg and Wisby (Quenn.), Wismar (Ehr. and Stein), Ostershelde (Rees), Mittelmeer Cette (Duj.), Gulf of Naples (Geta-Fentz) Gulf of Mexico (Smith), Siberia and Egypt (Schewiakoff).

*Indicates the only forms so far reported.

This species is the type of the genus as correctly designated by Claparede et Lachmann, and later by Fromental.

A. polystyla Stein.

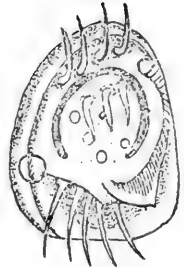
A. polystyla Stein 1859 (subgenus *Onychaspis*).

Onychaspis Diesing 1865.

A. polystyla var. *maxima* Gourret et Roeser 1886.

A. plana Perejas. 1886.

A. polystyla Calkins 1902.



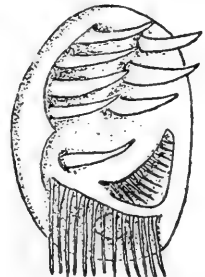
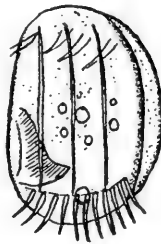
A. lynceus ventralside
Stein-Taf. III, Fig. 4

Body oval, the margin entire, the left border nearly straight; the dorsal surface slightly convex, traversed by three longitudinal furrows; ventral styles 7 to 8, forming two anterior, oblique, parallel rows of 3 or 4, and 3, one separate ventral style stationed by itself to the right and rear of the other six; anal styles variable 10-13. L—1/510". Sea water.

Distributions:—Warberg (Quenn), Bay of Concarneau (Fabre-Domergue) Harbor of Marseille and Bastia Corsica (Gour. et Roes.), Harbor of Trieste (Stein), Gulf of Naples (Geta-Entz), Black Sea (Perejas.), Woods Hole, U. S. A. (Calkins).



A. polystyla—ventral and dorsal side.
Stein-Taf. III—Figs. 18 and 19.



A. polystyla—ventral side
Calkins—Fig. 57.

This species is evidently close to the preceding in all except the number and distribution of the cirri. In these it is very variable. Calkin's variety from Woods Hole differs from Stein's original form in the possession of 8 ventral and 13 anal styles—the former banked and very massive. *A. plana* as described by Perejaslawzewa is identical in all essentials with this species. A form described by Gourret et Roeser 1886 as *A. polystyla* var. *maxima* has 7 anal styles. The drawing (Pl. XXXIV, Fig. 1), which is evidently from the dorsal rather than the ventral side as stated, is so

inaccurate that it is of little value. Except for the small number of anal cirri, the description is fairly close to that of Stein.

A. turrita (Ehr.)

Euplotes turritus Ehr. 1838.

Aspidisca turrita Clap. et Lach. 1857.

Body suborbicular, widest and somewhat truncate posteriorly, its marginal border even, the left side nearly straight, the opposite one rounded; a thorn-like recurved spine developed from the center of the dorsal surface; ventral and anal styles as in *A. lynceus*. L—1/450". Fresh and salt water.

Distributions: Baltic (Ostsee) at Hapsal (Eichwald), Wismar (Ehr.) Gulf of Naples (Geta-Entz), Gulf of Mexico (Smith), common in pond water about New York City.

A. costata (Duj.)

Trichoda cicada? Muller 1786, 1788.

Oxytricha cicada? Ehrenberg 1838.

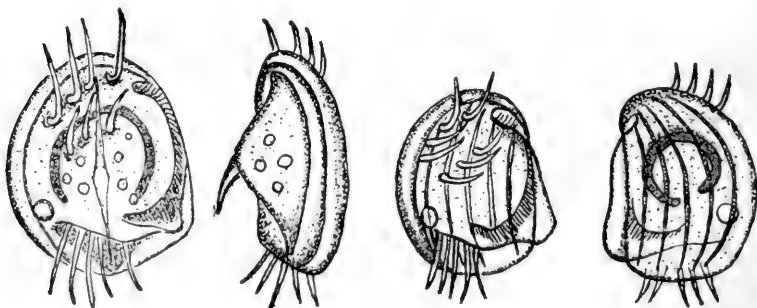
Coccudina constata Duj. 1841.

Aspidisca cicada Clap. et Lach. 1857.

Aspidisca costata Stein 1859.

Body nearly oval, or left side nearly straight, right convex; the ventral portion developed outwards and backwards beneath the peristome in the form of a triangular plate toward the posterior extremity of the left side; dorsal surface grooved by six longitudinal furrows; ventral styles 7, in two oblique rows—anterior 4, posterior 3; anal styles 5. L—1/690". Brackish or pond water.

Distribution:—North Sea Water Aquarium (Fresenius), Finland Brackwasserbuchten at Ransösund (Levander), Gulf of Genoa (Gruber), Gulf of Mexico (Smith), Buenos Aires (de la Rue), common in pond water in vicinity of New York City.



A. turrita—ventral and side view.
Stein-Taf. III—Figs. 11 and 14.

A. costata—ventral and dorsal view.
Stein-Taf. III—Figs. 15 and 16.

This is the only species recognizable from Dujardin's drawings. It is there drawn with seven anal cirri, and the ventral plate is not shown. In spite of these inaccuracies, however, we must agree with Stein that the form of the body and the grooved dorsal surface are unmistakable.

B. Left border armed with a single backwardly directed spur in its posterior third.

A. hexeris Quenn.

A. hexeris Quennerstedt 1867 v. 4.

A. Andrewii Mereshkowsky 1878.

A. hexeris Calkins 1902.

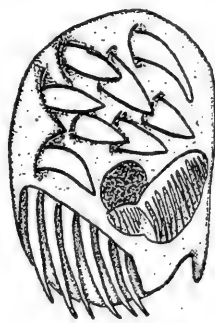
Body elliptical, about one and one-half times as long as broad, equally rounded at each extremity; the left border armed close behind its center with a single backwardly directed spurlike projection; dorsal surface usually grooved by three faint longitudinal furrows; ventral styles short and thick, 7 to 8 in number, generally in two rows of 3 and 3, with one placed by itself posteriorly, 6 anal styles. L—1/500". Salt water.

Distribution: Wisby, Gotland (Quenn), White Sea (Meresch), Woods Hole, U. S. A. (Calkins).

A. hexeris as described by Calkins from Woods Hole agrees closely with Quennerstedt's original description except for the possession of 8 ventral styles. As already indicated this character is not believed to be of importance in differentiating species. *A.*



A. hexeris—dorsal side.
Quenn. II—Fig. 19.



A. hexeris—ventral side.
Calkins—Fig. 50.

Andrewii Meresch. is in all respects identical with this species. Hamburger und von Buddenbrock 1911 consider this species to be identical with *A. leptaspis* Fresenius. The resemblance between the two is merely superficial, however, for the anterior spur and

the marked serration of the cuirass, both of which are extremely constant in *A. leptaspis*, are never found in *A. hexeris* either in the European or American varieties.

C. Left border armed with two spurs, one in the anterior and one in the posterior third.

A. lyncaster (Muller).

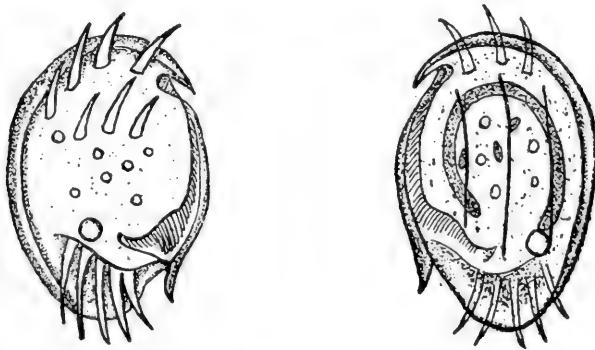
Kerona lyncaster? Muller 1786.

Trichoda lyncaster Muller 1788.

A. lyncaster Stein 1859.

Body ovate, rounded on the right side and at the two ends, the anterior border of the left side incised so as to produce a posteriorly directed spur-like projection, a similar aculeate spur, curved slightly outwards, projecting from the ventral surface of the carapace on the same side within a short distance of the posterior margin; ventral styles short and thick, 7 in number, placed in two oblique rows of 4 and 3; anal styles 5. L—1/450'—1/360". Salt water.

Distribution: Finland See at Löfo (Levander), Baltic Sea at Travemünde and Stralsund (Stein), Kiel Harbor (Möbius), Gulf of Genoa (Gruber), Gulf of Naples (Geta-Entz), Harbor of Bastia Corsica (Gour. et Roes.)



A. lyncaster—ventral and dorsal view.
Stein-Taf. III—Figs. 1 and 2.

A. leptaspis (Fresenius).

A. leptaspis Fresenius 1865.

A. crenata Fabre-Domergue 1885.

Body oval, blunt at either end, the left side somewhat less curved than the right; the right border smooth, the left incised in the anterior third so as to produce a feeble spurlike projection; in its posterior third a similar but more prominent and sickle shaped spur from both the upper and lower plates of the cuirass; outer posterior border of upper plate evenly notched or ser-

rated; lower plate similarly serrate anteriorly and posteriorly; upper surface may show three feeble longitudinal striations; ventral styles 7—two rows of 3 each, with one placed posteriorly by itself; anal styles 5 to 7. L—.065—.07 mm. Salt water.

Distribution: North Sea Water Aquarium (Fresenius), Osterschelde, Belgian coast (Rees), Bay of Concarneau (Fabre-Domergue).

This peculiar form is of especial interest because of the observation of van Rees 1884 with regard to the fraying out of one of the anal cirri so as to form three, making a total of seven instead of five as the species was originally described by Fresenius. The seven styled form was described by Fabre-Domergue 1885 as a new species *A. crenata*. The fact that such a condition occurs proves that the actual number of cirri is of little value in recognizing species, and suggests that a similar condition may exist in other Hypotrachs which have not been observed for an extended period. The anterior tuft of cilia figured by van Rees is also of interest. If the observation is correct it appears that the adoral cilia may sometimes extend a considerable distance towards the anterior end. On the other hand it seems more likely that the tuft represents an additional ventral cirrus—a condition observed by Calkins in at least two other species of *Aspidisca*.



A. leptaspis—ventral view.
van Rees—Fig. 11.



Ventral view
Fabre-Domergue—Fig. VII.

D. Left border with three spurlike projections.

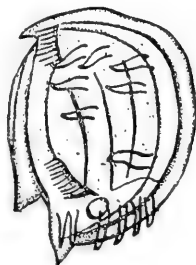
A. sedigita Quenn.

A. sedigita Quennerstedt 1868 v6.

Body broadly ovate, suborbicular, the right border smooth and rounded, the left one incised so as to produce an anterior, middle, and posterior spurlike projection—the middle spur extending from the ventral surface as does the posterior one in *A. lyncaster*; ventral styles 7, short and thick, centrally

disposed; six stout, scarcely prominent anal styles. L— $1/325''$. Salt water.
Distribution: Wisby, Gotland (Quenn).

This species is made identical with *A. lyncaster* by Hamburger und von Buddenbrock 1911. According to Quennerstedt's original description, however, *A. sedigita* possesses a third spur at the posterior end of the body giving it a characteristic pointed appearance. This together with its smaller size makes it necessary that the species be retained.



A. sedigita—Dorsal side.
Quenn-Taf. II—Fig. 2.

DOUBTFUL SPECIES

A. denticulata Ehr. 1838 is insufficiently described and the figure is not recognizable as an *Aspidisca*. The name suggests that it may be identical with *A. leptaspis* Fres. but the description of the denticulations as on the left side precludes this.

A. radiata Fromentel and *A. pulvinata* Fromentel 1874 probably cannot be considered good species because of insufficient description. The former has ten ventral cirri and the adoral fringe of cilia at the anterior end, which probably put it outside the limits of the genus. The latter has no apparent differences from *A. costata* (Duj.).

A. bipartita Gourret et Roeser 1886 very clearly has none of the characters of the genus, such as a group of ventral styles and an adoral fringe of cilia on the left side of the ventral surface. The description and pictures look very much like a minute crustacean belonging to the group Cladocera.

Columbia University, New York City.
October 19th, 1916.

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NEMATODE TECHNIQUE*

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Difficulties in making satisfactory preparations for examination have prevented many from taking up problems with nematodes. The author, in his study on some of the parasitic species, undertook to solve a few of the general problems in the technique and also some of the more particular processes for demonstrating certain organs and systems. This work has by no means exhausted the problem, but perhaps the methods given here will serve as a guide for future work along this line. These methods, while applicable to free-living nematodes, have been worked out in particular for the parasitic forms.

Collecting parasitic nematodes differs little from collecting other parasitic worms. However, there are some things that have to be kept carefully in mind. In the first place many forms are encountered with such well developed mouth parts that it is difficult to free them from their attachments without injury. The best method is to take hold of the host tissue very close to the mouth of the parasite with a pair of fine-pointed forceps and with gentle pressure and slight traction, to pinch the animal off; in this procedure the forceps take up so little tissue that when the worm is freed they close with nothing between their points. When the worms are not so firmly attached, by gently stroking them with a fine camel's-hair brush in a direction away from the tissue, they will often come loose; such a brush will be found in general to be the most convenient instrument for handling parasitic nematodes.

In looking for alimentary parasites it is a safe plan to split the gut with a needle, for then the danger of cutting a worm is nil; however, in the examination of the larger animals this is not always practical and here one must very cautiously cut the wall with blunt scissors. In most cases gripping the opened gut

*Contributions from the Zoological Laboratory of the University of Illinois under the Direction of Henry B. Ward, No. 77.

tightly between the two prongs of the forceps and drawing it thru them will free, in good condition, all of the parasites that adhere. If an examination is to be made of the lungs, liver, kidneys or muscles of an animal it will be found of advantage to tease them out with needles. In case nematodes are found encysted, they should be carefully dissected out, before preservation, under a good lens or binocular.

Most nematodes are rather sensitive to changes in the osmotic condition of the medium in which they are placed. Tap water has been found to be far better for temporary keeping than physiological salt solution, and while some of them live for a while in distilled water, this medium should be generally avoided. For most of the nematodes found in fresh-water fishes a 0.3% salt solution will be found to be about right. As a matter of fact the best medium to keep them in while making the necessary observations on the living material is that which collects in the dish in which the examination is being made, and into which the isolated organ has been placed for examination. In spite of the fact that Looss found the Sclerostomidae of horses and donkeys remain in good condition after being kept for hours in physiological salt solution, it will be found that there is an advantage in killing and fixing the worms as soon as possible after they have been removed from the host. Under no condition should they be allowed to dry at any stage in the process.

Owing to the nature of the cuticula nematodes are very hard to preserve. Most cold killing fluids penetrate so slowly that nematodes live for hours in fluids which will kill other parasitic worms in a few seconds; the hot fluids coagulate the proteins of their bodies before they get in, thus making penetration harder, but they have the advantage over the cold reagents in that the specimens are killed in a straight position. While this last statement is in general true, it does not always hold, for with most of the Trichinellidae the anterior region of the body will coil up like a spring even tho the fluid be fairly hot. This may be prevented in a great measure, if not entirely, by placing the anterior end of the worm in the angle formed by pressing the points of a pair of forceps together and working the specimens to and fro in the hot fluid as

soon as placed into it. In applying this method the fluid should be first heated and then the worms transferred into it. In most species the posterior region of the males curl up, and no method has yet been devised to prevent this. No successful method of anesthetizing the worms before fixation has been found by the author, and this procedure should be avoided. If worms are cut into pieces before fixation the organs crowd out at the cut end and the whole animal presents a very abnormal picture when sectioned, due to the pulling and stretching of the tissues. However, for certain purposes this is not an objection.

Without doubt the most successful killing fluid ever recommended for general work in this group is the one devised and used by Looss. Because of the simplicity of its use and general good results, it has been followed by nearly all workers since he (1901) published the method. The procedure is this: worms, carefully freed from debris, are placed in a mixture of 20% glycerol in 70% alcohol, if the worms are small, and in 10% glycerol and 70% alcohol, if they are large, the mixture previously being heated to 80° C. They may be preserved in this mixture for future use or put into an incubator or on a paraffin bath with the cover off of the dish in which they are contained. The alcohol and water are allowed to evaporate slowly from the glycerol, care being used to keep the worms from coming into pure glycerol too quickly, else they will collapse.

Twenty-four hours is slow enough for some small worms, but twenty-four days is too fast for some of the large ascarids. The time may be regulated by the temperature or by covering the dish partially. In glycerol they are very transparent and may be studied or kept in this fluid or transferred directly into glycerine-jelly and mounted on a slide. Of course they are not stained by this process, but many characters come out beautifully in material prepared in this way. Looss transfers material directly into 96% alcohol for sectioning, after making a few incisions in the cuticula with a very fine sharp knife. They are then brought into absolute alcohol, oil of cedar and finally paraffin. He found that oil of cedar was the best medium for this purpose; on the other hand the author has had no success with it, whatsoever, but since the product as

sold commercially varies so greatly, no importance can be attached to his negative results. In connection with the very excellent work of Looss it is interesting, from the standpoint of what is to follow, to quote one of his statements: "Canada-balsam was almost entirely excluded because it made unstained objects too transparent, and stained nematodes were less favorable for mounting than unstained ones". On the same subject Braun and Lühe express themselves in no uncertain terms: "Man wirt von vornherein auf das Färben ganzer Tiere verzichten müssen und kann auch wegen der kaum zu vermeidenden Schrumpfung der Cuticula beim Aufhellen mit Kreosot, Terpentin, etc. den Einschluss in Canada-balsam oder anderen Harzen nicht anwenden".

As for general nematode technique, there seems to be in literature few references or results published other than this work of Looss, altho there are indications that other killing fluids were used to some extent by the authors prior to 1901. Since this time a method for handling nematodes was proposed by Langeron which can be used to advantage, since it is very rapid and reliable. Nematodes are killed in diluted formol (5:100) and then transferred, after several hours, into a lacto-phenol mixture, made as follows and used first in half strength:

Glycerol	2 parts
Phenol	1 part
Lactic acid	1 part
Distilled water	1 part

After a few hours the nematodes are placed in this fluid of full strength and either preserved in it, or mounted on a slide in a drop of the mixture, enclosed in a gold-size ring, covered with a cover glass, and sealed.

Looss and Braun and Lühe claim that it is not practical to mount nematodes in balsam after staining, yet when material is handled in the correct manner, it becomes the very best medium and further allows of staining to suit the needs of the student. It is obvious that animals so sensitive to changes in osmotic pressure will require very cautious treatment while bringing them into paraffin or balsam, and it was Cobb who first proposed a plan by which they could be brought into these reagents, and for this purpose he

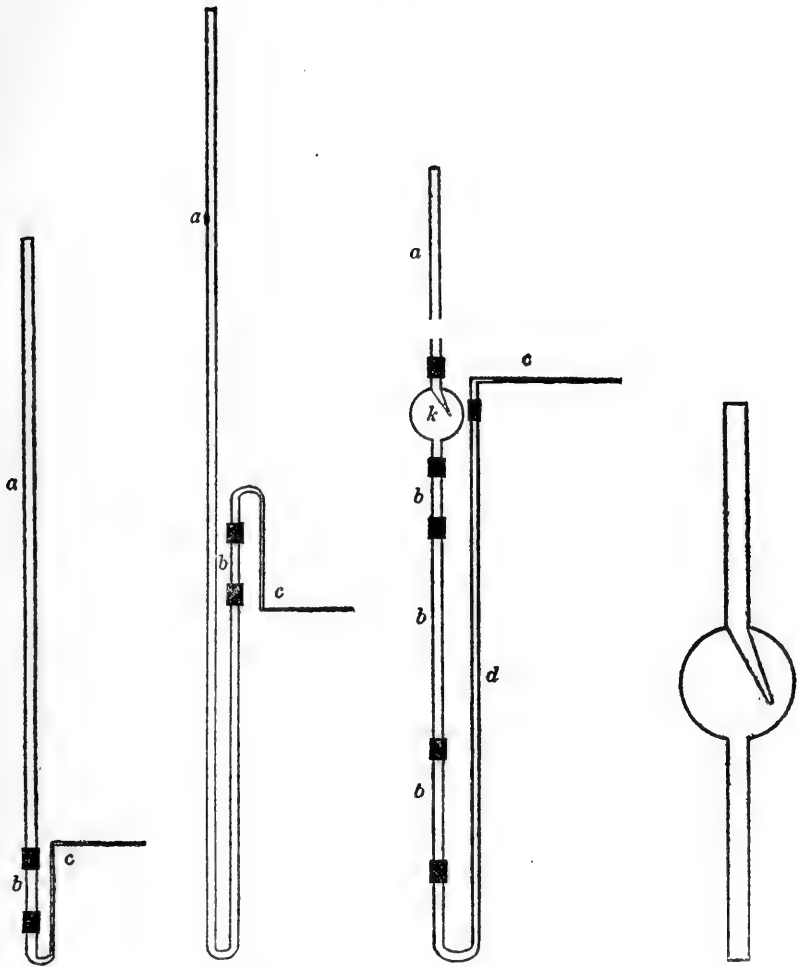


FIG. 1.

FIG. 2.

FIG. 3.

FIG. 4.

- FIG. 1. Differentiator used for dehydration. *a*, reservoir; *b*, object box, plugged at each end with cotton; *c*, filter, also with cotton plug. (After Cobb.)
- FIG. 2. Differentiator used for transferring objects from absolute alcohol into a clearing fluid. Letters as in Fig. 1. (After Cobb.)
- FIG. 3. New type of differentiator for dehydrating. *a*, reservoir; *b*, object holders; *c*, filter and regulation devise; *d*, safety tube; *k*, mixing chamber. The reservoir should be two metres long, and is shown sectioned in the figure. In filling, avoid bubbles.
- FIG. 4. The mixing chamber of the new differentiator. Made of glass tubing, in which a bulb is blown and a pointed piece of tubing is fused into one end.

devised the differentiator. This ingenious devise (Fig. 1) consists of a glass tube 5 mm. or more in diameter, which is used for the reservoir. To this is attached, by rubber tubing, the object box, a short piece of tubing, plugged at each end with a cotton plug and holding the specimens. The object box is also attached to the filter, a third piece of glass tubing, which is drawn out into a fine capillary tip and bent as shown in the figure. The filter is filled with whatever fluid the worms are in, for instance corrosive sublimate solution, and is attached to the object box, after inserting a cotton plug in the proximal end. The box is filled and the object placed into it, the distal plug inserted and the whole connected to the reservoir, which is filled in the following manner: mix equal parts of sublimate solution and 35% alcohol (solution 2), equal parts of solution 2 and sublimate solution (solution 1), equal parts of solution 2 and 33% alcohol (solution 3). The reservoir is filled one-fourth full of each solution in order, 1, 2, 3, and then on the top 33% alcohol is added to fill the reservoir. A wire is passed down the reservoir to cause a little mixture, and withdrawn. Minute drops flow from the filter and the rate of flow can be regulated by tilting the differentiator, so that the worms are brought into 33% alcohol in from two to five hours. As the reservoir is emptied it is filled with solutions of the next higher grade of alcohol made up in the manner described. Specimens may be stained here, destained, and finally brought into absolute alcohol. Because the clearing fluids are heavier than alcohol it is necessary to use a reverse form at the tip of the differentiator. (Fig. 2). Here the pressure is equalized by bending the reservoir as figured and by the same general plan the worms are brought into the clearing fluid (Cobb used oil of cloves if the worms were uncut and chloroform if they were cut) and finally into balsam.

For some time this method was used with a fair amount of success but certain disadvantages attended it. The most obvious one is that in using the instrument so much time is required in the making up of the many solutions and the constant refilling of the reservoir. Then, too, the change is not gradual enough for the best work, especially with some very delicate worms. Further, clearing fluids attack the rubber connections. After many

trials the following method and instrument for handling the worms were devised: worms are killed in 50% alcohol heated to 60-75° C. and transferred at once to any one of the following mixtures: (A) Carnoy's (33 parts chloroform, 33 parts glacial acetic acid, 33 parts alcohol, the mixture is then saturated with corrosive sublimate), (B) a mixture made of equal parts of alcohol, water and acetic acid, saturated with corrosive sublimate, or best of all (C) using the latter mixture with the addition of enough osmic acid to make the solution contain from 0.05% to 0.1% osmic acid. These killing fluids give very good results, but for the very fine preservation of histological detail, the last one given is by far the best. In these fluids the material is left for from one to ten hours, depending upon the size of the worms. It is possible to get good results by killing in hot water and then transferring into a saturated aqueous solution of corrosive sublimate, with or without acetic acid, or in combination with osmic acid. If the nematodes are killed in osmic mixtures they should be bleached by adding a little hydrogen peroxide to the water into which they are brought after fixation in water solutions, or to 50% alcohol if they are killed in alcoholic solutions. After this treatment they are brought very gradually into 70% alcohol and the corrosive sublimate removed with iodine solution. One can usually succeed in doing this within a period of ten or twelve hours and six or eight changes, so that the nematodes are perfectly round and not distorted, but to be safe, the new differentiator (Fig. 3) to be described later should be used. After the iodine is removed they are graduated into 80% alcohol and preserved in this strength, being careful not to let it become weaker. If they are to be used for totos at once they should be stained in Ehrlich's acid hematoxylin (diluted 1:25-50) or in Delafield's hematoxylin of the same dilution, in which event it will be a saving of time to remove the corrosive sublimate when they are in water or 35% alcohol with iodine dissolved in water to which a little potassium iodide is added. For sectioning it is useless to try to stain in toto, yet material may well be stained in one of the hematoxylin in order to make them easy to see and handle, or Mayer's pararcarmin may be used for this purpose when they are in 70% alcohol. In either case they are stained for twenty-four hours

and then destained to the proper intensity in 5% hydrochloric acid in water or 35% alcohol and the hematoxylin material "blued" by transferring into a 5% solution of ammonia water, in either water or 35% alcohol. They are then ready for the differentiator. The new form of this instrument (Fig. 3) consists of the following parts: the reservoir, the mixing chamber, the object holders, the safety tube, and the filter and flow regulation device. The reservoir is a shell tube with an inside diameter of 5 mm. and two meters long, so that it will hold about 45 cc. of alcohol. The mixing chamber (Fig. 4) is made in the following manner: A piece of good thick-walled glass tubing, with an inside diameter of 4 mm. is pulled out at one end and sealed. Ten centimeters from the sealed end the tube is heated in a good flame and a bulb blown that will hold between six and eight cubic centimeters, then the sealed end is cut off as near the bulb as possible. The end that has been drawn out is bent in the flame near the tip (which has been broken off) at a slight angle and inserted into the bulb and the two held so that the tip points to one side and a little below the middle of the bulb; in this position they are fused together, and both ends of the tube are cut and rounded off about 5 cm. from either end of the bulb. If for any reason this type of mixing chamber cannot be made, the substitute shown in Figure 5 can be used. It is made with a piece of large glass tubing and rubber stoppers. The object holders are pieces of shell tubing with an inside diameter of from 5 to 7.5 mm. and varying in length from 5 cm. to that needed to hold the worms without bending. These are plugged with cotton at either end to keep the worms in place. The safety tube is 2 or 3 mm. inside diameter and bent in the shape of a U with one arm 5 mm. long and the other long enough to reach to the middle of the mixing chamber when in use. The filter is a tube 3 mm. in diameter, bent in the form of a right angle and pulled out into a fine capillary at one end; a cotton plug in it acts as a filter to prevent stoppage by particles in the alcohols. Rubber tubing is used for connections. To manipulate the instrument requires some practice and the order is essential. The filter is filled and connected to the safety tube as shown in Figure 3. A pinch-cock on a piece of rubber tubing at the end of the safety tube will keep it full while the rest of the

apparatus is being filled with the grade of alcohol or water in which the specimens are. The object holders are next filled and the specimens put into them and plugged with cotton. As many holders can be filled as necessary and connected together in a long line, each being properly labeled. These are then connected to the safety tube and a pinch-cock applied to the rubber-tube-capped free end. The mixing chamber is capped with a short rubber tube, then filled by drawing in the liquid of the same strength in which the specimens are, whereupon it is closed with a pinch-cock. This is then connected to the terminal object holder, the other end of the mixing chamber being connected to the reservoir. The reservoir is then filled by putting in four grades, one on top of the other, in order,



FIG. 5.

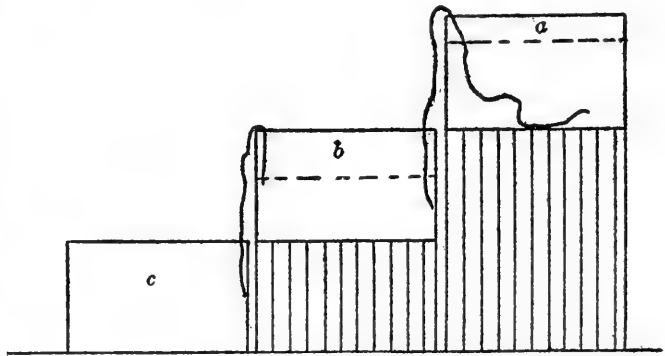


FIG. 6.

FIG. 5. Mixing chamber made of a piece of glass tubing with rubber stoppers and inlet and outlet tubes.

FIG. 6. String siphon system for handling small embryos and eggs and transferring nematodes from absolute alcohol into a clearing fluid. *a*, for the clearing fluid; *b*, for the objects; *c*, waste. *a* and *b* are supported on wooden blocks. The whole is placed under a bell-jar with sulphuric acid in a beaker to absorb the moisture.

such as 35%, 50%, 70%, 85% alcohol, the lowest being always the next step above the fluid in which the worms are contained. The capillary tip is regulated to yield 5 to 10 small drops per minute, and as the reservoir is emptied, it is filled with 95% alcohol until 20 cc. have been added. It is then filled with absolute alcohol until 50 cc. have been added, whereupon the apparatus is allowed to empty itself

as far as the safety tube will permit, as this will never allow it to run dry. The mixing chamber will attend to the mixing of the alcohols as the lighter one is forced under the heavier one and mixes very efficiently. After the absolute alcohol is all in the reservoir a calcium chloride tube is attached to the top of the reservoir, and the specimens allowed to remain in the alcohol for several hours. The object holders are now taken out ready for the clearing fluid.

The best method for getting the material into the clearing fluid is by means of a string siphon. Three Stendor dishes are arranged in stair-step fashion (Fig. 6). The objects are placed in the middle dish, either in the tubes or free, and this dish is connected with the receiving end of a siphon, made by using any suitable piece of string, regulating the flow by the size of the string. From this dish another string is led to the waste dish below. If the nematodes are to be used for sectioning one puts in the upper dish a mixture of absolute alcohol and xylol, half and half, and after this has run out xylol full strength, regulating the flow so that the specimens come into pure xylol in about 36 hours. If the specimens are intended for toto mounts synthetic oil of wintergreen (methyl salicylate) is used in the same way as xylol. The xylol-cleared worms are gradually brought into paraffin either after being cut into pieces or still whole, by allowing small pieces to dissolve in the xylol; they are finally saturated in xylol-paraffin at 35° C., and embedded after infiltration for one-half to one and a half hours in paraffin melting at 56° to 58° C. By coating, with soft paraffin, the block of hard paraffin in which the nematodes are embedded, it will be found that good sections in series can be made, provided the knife is kept sharp. For this convenient method, the author is indebted to Mr. H. G. May. Some little success has been met with infiltration in vacuo and this method, if adapted somewhat, will undoubtedly yield results worth while. The worms cleared in oil of wintergreen should be put in parchment boxes or boxes made of heavy linen bond paper and placed in Stendor dishes with damar dissolved in oil of wintergreen, and the mounting medium allowed to slowly dialyse and penetrate. It is best to pierce the cuticula in one or two places before the worms are placed into the cups, and this can be done with a sharp needle so that none of the internal organs are injured.

After twenty-four hours treatment in this manner they may be mounted under a cover glass on a slide in damar in oil of winter-green. For staining sections good results are obtained with Delafield's hematoxylin, Ehrlich's acid hematoxylin, Unna's orcein method, Mallory's connective tissue stain and thionin in saturated solution in 1% phenol. Borax carmine is of no value and strange as it may seem, iron hematoxylin yields also poor results.

It will often be found of advantage to dissect out parts of nematodes and for this purpose glass rods drawn out into fine points are useful. For dissecting out the mouth parts, maceration in concentrated potassium hydroxide, and mounting in glycerine-jelly will give good results.

Nematode embryos and eggs are killed in strong Flemming's or the fluid of Ripart and Petit (camphor water, not saturated, 75 gms., distilled water, 75 gms., glacial acetic acid, 1 gm., copper acetate, 0.3 gm., copper chloride, 0.3 gm.) to which a few drops of osmic acid have been added, and are either carried thru the staining and dehydrating process in a differentiator or in a string siphon system. In using the latter it is best to cover it with a bell-jar and enclose also a beaker of sulphuric acid to absorb the moisture.

Goldschmidt found that the nerve technique of Bethe, Apáthy, Wolff-Bielschowsky and Cajal could be applied to nematodes as well as to other forms. He offered a modification of the Cajal method that gave good results. Material is fixed in ammonical alcohol for 24 hours, 6 days in 10% silver nitrate in an incubator, 24 hours in hydrochinon or pyrogallol and formol; it is then embedded and sectioned. The sections are treated for twenty minutes in 0.1% goldchloride and reduced in a sodium fixing bath for half an hour. His method of preparing toto mounts of the large forms to show the nervous system was to split the cuticula and remove the esophagus. The stretched out "shell" was then stained for 6 to 8 hours in Nissl's alkaline methylene blue at 60° C., dehydrated and cleared in oil of cloves, in which they were kept, the excess stain being taken out by the action of the oil of cloves in two or three days.

The methods herein described have given good results for the author and it is hoped that others will be able to reproduce and

improve upon them. Several failures should not discourage any one, for at best time is required to obtain really good results in this very difficult field.

The author wishes to express his sincere thanks to Professor Henry B. Ward for his careful suggestions and criticism of the manuscript of this article. Valuable assistance in many ways has been received thru the close personal association with my friend Mr. Henry G. May.

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DEPARTMENT OF NOTES, REVIEWS, ETC.

It is the purpose, in this department, to present from time to time brief original notes, both of methods of work and of results, by members of the Society. All members are invited to submit such items. In the absence of these there will be given a few brief abstracts of recent work of more general interest to students and teachers. There will be no attempt to make these abstracts exhaustive. They will illustrate progress, without attempting to define it, and will thus give to the teacher current illustrations, and to the isolated suggestions of suitable fields of investigation.—[Editor.]

ENTOMOLOGICAL NOTES

Collembola.—Folsom ('16, Proc. U. S. Nat. Mus., 50:477-525) presents a paper treating of all of the known species of North American *Poduridæ* (*Collembola*), with the exception of the subfamily *Onychiurinaæ*. Keys to all of the taxonomic groups and full descriptions of the species are features of the paper. A bibliography of one hundred seventeen titles, and eighteen plates containing two hundred fifty figures, add to the value of the work. It is a paper indispensable to persons interested in the *Collembola*.

Pure Lines in Aphids.—Ewing ('16, Biol. Bull., 31:53-112), in a paper entitled "Eighty-seven Generations in a Parthenogenetic Pure Line of *Aphis avenæ* Fab.," finds no summation effect by selection, using six different fluctuating variations, and is of the opinion that summation through continued selection is not to be expected since fluctuations in such a pure line "are in general not dependent upon germinal variations." Selection from extreme variants was without effect on somatic characters of succeeding generations. Long continued selection from the extreme variant in each succeeding generation produced no more change in the mode of the variable than did the selection from individuals differing but slightly from the mean of the line and for only a few generations. Nor was the mean of the line shifted when selections from extreme variants for forty-four successive generations were made, using a character (body-length) "which is known to be inherited in the higher animals that reproduce sexually." Variations in body-length were found to be largely due to variations in temperature and food. The rarely appearing discontinuous variations were apparently not inherited. Size, fecundity, and color were not affected by long continued parthenogenetic reproduction. Pædogensis occurred occasionally

among the nymphs of both winged and wingless forms and the offspring which reached maturity were normal. It is thought that pædogogenesis in this aphid is due to "the arrested development of the body in general, while the reproductive organs become completely functional."

Insect Flagellates and Disease.—Fantham and Porter ('16, Journ. Parasitology, 2:149-166) have studied "the significance of certain natural flagellates of insects in the evolution of disease in vertebrates." Flagellates, including members of the genera *Herpetomonas* and *Crithidia*, secured from the alimentary tracts of several species of insects, were introduced into representatives of the *Pisces*, *Amphibia*, *Reptilia*, *Aves*, and *Mammalia*, by inoculation or by feeding. The latter was accomplished by feeding the host with the infected insects, or with the intestines of the insects, or with food contaminated with the feces of insects containing the resistant stages of the *Flagellata* concerned. It was found by these methods that *herpetomoniasis* can be induced in various warm and cold-blooded vertebrates, the nature of the infection and the protozoan parasites found in the vertebrate hosts resembling those of human and canine leishmaniasis. *Herpetomonas jaculum*, *H. stratiomyiæ*, *H. pediculi*, *H. ctenocephali*, *H. culicis*, and *Crithidia gerridis* were found pathogenic to warm-blooded vertebrates, the first and last mentioned having been also successfully introduced into cold-blooded hosts, such as fishes, frogs, toads, lizards, and grass-snakes. In acute cases of the induced disease, the flagellate form of the parasite was most numerous at the death of the host, while in chronic cases the non-flagellate forms predominated. These writers hold that a *Leishmania* is morphologically a *Herpetomonas*, that leishmaniasis are invertebrate-borne *herpetomoniasis*, "and that these maladies have been evolved from flagellates of invertebrates (especially insects), which have been able to adapt themselves to life in vertebrates."

Phylloxera Galls.—Rosen ('16, Am. Journ. Botany, 3:337-360) finds that the leaf-gall produced by *Phylloxera vastatrix*, which occurs on *Vitis vulpina*, starts to develop on the embryonic bud leaves and soon produces a depression due to the upward growth of tissue at the sides. That portion of the leaf surrounding the proboscis and beneath the insect shows no proliferation.

Gall development depends upon leaf development, the gall becoming mature when the leaf reaches maximum size. No support for the theory that the insect causes gall formation by the injection of some chemical appears in this study. The initial stimulus for gall development is believed to be the continuous sucking action of the insect at a single fixed point.

Color Inheritance in Phasmidæ.—MacBride and Jackson ('15, Proc. Royal Soc. London, (B) 89:109-118) have studied the inheritance of color in a phasmid, *Carausius morosus*, from southern India, which exhibits marked color varieties. Of the several thousand insects reared, only six males and one gynandromorph appeared, the results therefore being concerned with parthenogenetic inheritance. All the insects are alike at hatching, having a definite color pattern of green and brown pigments. In subsequent growth, the green pigment overpowers the brown so that the insect appears pure green in the majority of cases, although, in about three per cent., the reverse occurred. The color of the mother does not influence the proportion of predominantly green or predominantly brown offspring. Pure green forms were secured from larvæ reared in complete darkness. Conditions of reduced light did not increase the proportion of brown forms. Males are very rarely produced from the eggs of unfertilized females.

Cuticula vs. Parasites.—Thompson ('15, Proc. Cambridge Philosoph. Soc., 18:51-55) presents argument and data in support of "the cuticula of insects as a means of defence against parasites." Some of the positive points stressed are: (1) Thickness and resistance of the cuticula sometimes effectively prevent the entrance of dipterous parasites. (2) In the process of molting, the larvæ of insects often succeed in freeing themselves from the unhatched eggs of dipterous insects. (3) Certain parasites, which have made entrance into the body of the host but have not withdrawn completely into the body-cavity, are thrown off in molting. (4) The probability that the cuticula is some protection even against the well-developed ovipositor of hymenopterous parasites. It is contended that the marked parasitism to which insects are subject is not necessarily a proof of the inefficiency of the cuticula and a possible explanation can be found in the fact that a great many of the parasites

are themselves arthropods and that the arthropod structure and life history render the members of the group especially able to support parasitic invasions. "It seems highly probable that the cuticular armour, and the function of ecdysis correlated with it, in reality arrests a very considerable part of the violent attack which many members of the Arthropoda are obliged to sustain."

Polyhedral Bodies.—Glaser and Chapman ('16, Biol. Bull., 30:367-390) have studied the nature of the curious crystal-like structures called *polyhedral bodies* or *polyhedra* which are constantly associated with certain diseases of insects, known in America by the vernacular name *wilt*. The larval stages of thirteen species of *Lepidoptera*, belonging to eight different families, have been found to be susceptible to the polyhedral diseases, and at certain times these diseases kill off from 30 to 70 per cent. of some of the most noxious pests (gipsy-moth, tent-caterpillars, and army-worms). These structurally complicated polyhedra, which arise in the nuclei of certain tissue cells, are specific for a certain type of disease. They are "nucleoprotein crystal-like degeneration-products and not organisms."

Photosensitivity of Blowfly Larvæ.—Patten ('16, Journ. Exp. Zool., 20:585-598), in studying the changes of photosensitivity with age in the larvæ of *Calliphora erythrocephala*, tested the specimens daily, from hatching to pupation, by subjecting a larva, crawling under the influence of a horizontal beam of light, to an instantaneous change of 90° in the direction of the beam and measured the resulting change in the direction of locomotion. The curve of photosensitivity, constructed on individual averages, showed constant negative reaction and that increased amplitude occurred during the first days of larval life, the maximum of 81° being attained on the fourth day. Steady decrease followed until the seventh day and thence to pupation the amplitude remained almost constant. Decrease in sensitivity was coincident with the initiation of the migration period.

Chemotropic Response of House-fly.—Richardson ('16, Science, 43:613-616) experimented with a number of organic and inorganic compounds which occur as products of fermentation in barnyard manures in an effort to discover whether the distinct oviposition

preference of the house-fly for horse manure is due to the odor of some volatile chemical substance which was liberated in the manure during the early stages of decomposition. Ammonia was shown to be a strong alluring agent and was particularly attractive to the females. In experiments with acidulated manure, oviposition response was approximately in inverse ratio to the distance from the source of the ammonia. Butyric acid, and, to some extent, valerianic acid, augmented the oviposition response when added to moist ammoniated cotton. Ammonium carbonate and moist cotton lacking these acids produced no response. Since these acids are found in barnyard manure, the evidence points to them as the attracting agents for the fly.

Aquatic Lepidoptera.—Welch ('16, *Annals Ent. Soc. Am.*, 9:159-190) reports on the biology of certain aquatic *Lepidoptera* (*Nymphula maculalis* and *N. icciusalis*). Eggs of *N. maculalis* are invariably deposited about the egg holes of a chrysomelid beetle (*Donacia*) in the floating leaves of the yellow water-lily. Laboratory experiments showed that in the absence of *Donacia* egg holes, egg masses of *N. maculalis* may be deposited, after some delay, about the leaf margin or artificial punctures and incisions. The orientation of the eggs in a mass is definite and constant. Tracheal gills are absent in the first instar but appear in the second. The total number of gill filaments per larva increases from forty in the second instar to over four hundred in the full-grown larva. Construction of cases from excised pieces of food plant leaves is a constant larval activity, these cases functioning as a protection and a support in water. Larval dissemination is accomplished by crawling, by voluntary propulsion in detached cases, by effects of winds, waves, and currents on detached cases, and indirectly by the work of certain other aquatic insects which separate the leaves of the food plant from the petiole. The larvæ and pupæ usually pass the entire existence under water. The adult is aerial and nocturnal. Eggs of *N. icciusalis* are laid on the margins of leaves of *Potamogeton natans* and independently of the activities of other aquatic animals. Tracheal gills are absent in all instars. Case-making, similar to that of *N. maculalis*, is a normal activity of the larva.

Syrphidæ.—Metcalf ('16, *Maine Agr. Exp. Sta., Bull.* 253),

in a study of the *Syrphidæ* of Maine, gives particular attention to the larvæ of these flies, treating of the structure and habits in considerable detail. Five different structural types of larvæ occur, the species of each having approximately the same habits: (1) the aphidophagous type, which is composed mainly of predaceous species; (2) the boring type, which includes the species that feed in the bulbs of living plants; (3) the short-tailed, filth-inhabiting type, which includes a number of species that feed on exposed decaying animal and vegetable matter; (4) the long-tailed, filth-inhabiting type, which is characterized by an elongate, posterior, flexible, telescoping, respiratory process at least half as long as the body; includes a number of forms which are scavengers in habit; and (5) the microdon type, includes those anomalous forms, sometimes mistaken for *Mollusca* and *Coccidæ*, which live in the nests of ants. Descriptions of the life stages and the life histories of the species of Maine are given and the beneficial and injurious habits of the larvæ discussed. Keys to the known larvæ and pupæ of *Syrphidæ* are included. Nine plates include a large number of figures of structural detail and life history stages.

Nematode Parasites.—Merrill and Ford ('16, Journ. Agr. Research, 6:115-127) found two new species of *Nematoda* parasitic in insects. *Diplogaster labiata* parasitizes the adults of *Saperda tridentata* (*Colcoptera*), infesting the digestive tract in such large numbers that they rupture the wall, escape into the body-cavity of the host, and cause its death. These nematodes were reared in water cultures to which macerated beetles were added as food, thus affording opportunity to work out the life history. Another nematode, *Diplogaster arivora*, was found infesting the heads of termites (*Leucotermes lucifugus*) in numbers as high as 75 per host. They were found in the soil about infested termite colonies; also in the dead bodies of termites and other decaying matter. This parasite was successfully introduced into the termites. In cases of heavy infestation, the mortality of the host was high. *D. arivora* was also reared in water cultures and the life history determined.

Cestodes in Musca domestica.—Gutberlet ('16, Journ. Am. Vet. Med. Assn., pp. 218-237) has carried on experiments which show that the cysticercoid stage of *Choanotania infundibuliformis*,

a cestode which infests chickens, occurs in the common house-fly (*Musca domestica*). Flies which fed on the eggs of this tapeworm developed the cysticercoïd stage and chickens fed on flies developed the adult worm, the identity of the two stages being determined by morphological comparison. Circumstantial evidence points to the probability that certain other insects which commonly occur about poultry yards and which are readily eaten by fowls are the intermediate hosts of other species of cestodes.

Spermatogenesis in Dragon-flies.—Smith ('16, Biol. Bull., 31:269-303) describes spermatogenesis in the dragon-fly, *Sympetrum semicinctum*, and for comparison has examined another dragon-fly, *Libellula basalis*. The maturing sex cells in the testes of the nymphs occur in globular cysts arranged, one or two layers deep, around a central duct which extends zig-zag through each organ. The cysts seem to have no definite arrangement in the tubule according to age but all of the developing stages of the spermatozoa may be found in a single transverse section. In both species, there are 25 spermatogonial chromosomes which are so closely crowded together that they are difficult to study. Apparently, the leptotene threads unite side by side to form a spireme which breaks up into segments that seem to open out along the original axis of synapsis to form rings. These rings condense into crosses and then into quadripartite bodies or prophase chromosomes. Twelve bivalent autosomes and one sex-chromosome occur in the primary spermatocyte. In the second spermatocyte division, the sex-chromosome passes to one pole undivided, thus giving rise to two kinds of spermatids and subsequently to two kinds of spermatozoa. In *Libellula basalis*, the sex-chromosome passes undivided to one pole in the primary spermatocyte division, thus forming two kinds of secondary spermatocytes, while in the secondary division, it divides equally. Two kinds of spermatozoa are also produced but by a slightly different process.

Insect and Mite Galls.—Wells ('16, Ohio Journ. Sci., 16:249-290) has made a survey of insect and mite galls on the hackberry (*Celtis occidentalis*), giving particular attention to the histology of the galls and the gall bearing parts. The seventeen species of zoöecidia found were distributed as follows: *Acarinæ* 1, *Lepidop-*

tetra 1, *Hemiptera* 5, *Diptera* 10. All produce abnormal cell and tissue formations. The acarinous and lepidopterous galls are *kataplasmas* (cells and tissues differing but slightly from the normal), while the hemipterous and dipterous galls are *prosoplasmas* (cells and tissues differing fundamentally from the normal ones). The latter show definite specificity. Eight plates containing many figures of these galls and their morphological characters accompany the paper.

Viability of Mosquitoes.—Chidester and Patterson ('16, Ent. News, 37:272-274), in an experimental study of the influence of various concentrations of sea water on the viability of the salt marsh mosquitoes, *Aedes sollicitans* and *Aedes cantator*, find that under laboratory conditions the viability of the larvæ in salt water depends upon the salinity of the water from which they are taken and varies with the species. Larvæ of *A. cantator* died quickly in distilled water and in the higher percentages of salinity. All larvæ in water of 22 per cent. or above died within two days. Field records indicate that *A. sollicitans* lives and thrives in marsh water of a higher salinity than that which appears to be suitable for *A. cantator*. Evidence seems to indicate that the distribution and date of appearance of the two dominant species are, in part, dependent upon the salinity of the marsh water at various distances from the sea. It is suspected that investigation will show a certain amount of dissolved salt more favorable for the development of the eggs of one species than another.

Gregarines of Insects and Myriapods.—Watson ('16, Illinois Biol. Monographs, 2:1-258) reports the results of a study of the gregarines found as parasites in various *Orthoptera*, *Coleoptera*, and *Myriapoda*. Although primarily a work on gregarines, it contains much of interest to entomologists, since considerable attention was given to the relations of these parasites to their arthropod hosts. Twenty-two new species are described and additional data are given for many others. The paper includes a synopsis of the eugregarine records of the *Myriapoda*, *Colcoptera*, and *Orthoptera* of the world; also a list of the cephaline gregarines of the world and their hosts, followed by a second list arranged according to the hosts. Records of two hundred forty-three gregarines distributed among

two hundred seventy-six hosts are given, these numbers including a few incomplete identifications. A valuable bibliography and fifteen plates containing three hundred thirty-eight figures are included in the paper.

Pupæ of Lepidoptera.—Mosher ('16, Bull. Ill. State Lab. Nat. Hist., 12:17-159) has presented an extensive paper on the "Classification of the Lepidoptera based on Characters of the Pupa." The external morphology of lepidopterous pupæ is worked out in detail. The paper is rich in analytical tables to the superfamilies, families, subfamilies, and genera. Full descriptions and discussions of the various groups are given. Attention was given to the phylogeny of the order, using the following characters as the basis: the number of movable segments; the freedom of the appendages; the number of sutures in the head; the relative length of the body segments; the presence or absence of visible labial and maxillary palpi; the presence of exposed portions of the prothoracic femora in specialized pupæ; and the method of dehiscence. The nature of the paper makes impossible a summary here, but it is the most comprehensive and connected study of lepidopterous pupæ which has appeared and forms an important basis for work on these quiescent stages.

Classification of Pupæ.—Mosher ('16, Annals Ent. Soc. Am., 9:136-158) reports on the classification of the pupæ of the saturniid moths (*Saturniidae*). The general characters of the pupæ of this family are described and a key to nine genera is given. Keys to certain species are also included. Detailed generic and specific descriptions are given for the pupæ of the following: *Copaxa lavendera*, *Telea polyphemus*, *Trophæa luna*, *Agapema galbina*, *Callosamia promethia*, *Callosamia angulifera*, *Eupackardia calleta*, *Rothschildia orizaba*, *Rothschildia jorulla*, *Samia californica*, *Samia cecropia*, *Samia columbia*, *Samia gloveri*, and *Philosamia walkeri*.

Breeding Habits of Orthoptera.—Turner ('16, Annals Ent. Soc. Am., 9:117-135) has made a survey of the breeding habits in the *Orthoptera* and finds that preliminary copulatory movements are, within narrow limits, constant for each group of this order but vary from very simple ones in *Mantidæ*, *Phasmidæ*, and *Acrididæ* to complex ones in *Blattidæ*, *Gryllidæ*, and *Locustidæ*. All males show sex discrimination but the females are aggressive and show sex

discrimination only in some groups while in others they are entirely passive. Habits of copulation are typical for each family, e. g., in *Mantidæ*, *Phasmidæ*, and *Acrididæ* there is superposition of the body of the male; in *Blattidæ* and *Gryllidæ* superposition of the female occurs; and in *Locustidæ* end to end copulation is characteristic. Generalized reproductive behavior occurs in those families having the largest number of subfamilies. A significant parallelism between a classification based on reproductive behavior and one based upon palæontological evidence occurs, suggesting "that the different types of reproductive behavior have been fairly constant since their origin."

Brain of Termites.—Thompson ('16, Journ. Comp. Neurology, 26:553-603) has made a study of the brain of *Leucotermes flavipes* (termite) in the different castes, with reference to its finer structure, making, in addition, a comparison with corresponding organs in the castes of true ants. This comparison is interesting since both termites and ants have a complex social organization but differ in degree of specialization and intelligence. The study included the nymphs of the first and second form, the soldier, the worker, and the true adult. No sex differentiation occurs between the brains of the different castes or stages and but very little caste differentiation appears, although the optic apparatus shows a correlation between the degree of development of the compound eyes and the size of the optic lobes. The structure of the brain in termites resembles very closely that of ants, except that the mushroom bodies are much simpler and more primitive. The ocelli, present in the nymphs and adults of the sexual forms but absent in the worker or soldier, are simple, primitive, without lens or pigment, and lack the ocellar lobes of the ocellar nerves which occur in ants. The problematical frontal gland, found in all castes and situated on the postero-dorsal surface of the brain between the mushroom bodies, is composed of epithelial cells continuous with the hypodermis and innervated from the brain. It seems to be functional only in the true adult and soldiers. "The suggestion is made that the frontal gland may have arisen phylogenetically from the ancestral medial ocellus which is now lacking in the

termites, and that the 'fontanel' nerve may be a vestige of the former median ocellar nerve."

Reflex "Bleeding."—McIndoo ('16, *Annals Ent. Soc. Am.*, 9:201-222) finds that the "reflex bleeding" (the ejection of drops of liquid from the femoro-tibial articulations in certain coccinellid and meloid beetles) is a true reflex in *Epilachna borealis*, one of the *Coccinellidae*, but that the liquid, instead of being blood, is a secretion from hypodermal glands and passes to the exterior through innumerable tubes opening near and in the articular membrane. Hypodermal glands are distributed widely over the integument of this species, groups of them occurring on the tarsi and around the femoro-tibial articulations, two at the proximal end of the tibia and two at the distal end of the femur. All four contain about 100 pores. The articular membrane contains about four hundred pores of still another kind. Fluid is emitted from these groups of pores in response to irritation. The discharge of the secretion is accomplished by muscular contraction in the femur whereby the blood is forced into a specially devised chamber containing the glands. The glands associated with the femoro-tibial articulation lack the reservoirs which characterize those glands distributed widely over the body. The secretion is bitter and disagreeable in odor. Its function is thought to be that of protection and it is suggested that possibly it aids in sex recognition and in distinguishing between different individuals.

Effect of Röntgen Rays.—Runner ('16, *Journ. Agr. Research*, 6:383-388) has experimented with the effect of Röntgen rays on the tobacco, or cigarette, beetle (*Lasioderma serricornis*), using a new form of Röntgen tube designed by Coolidge. Heavy dosages are demanded in the treatment of cigars or tobacco infested with this insect. Heavier exposures must be used for eggs near the hatching point than for those recently laid. Dosage equivalent to 150 milliamperes minutes exposure with spark gap of 5.5 inches gave satisfactory results with eggs in tobacco placed 7.5 inches from the focal spot of the tube. Under this exposure, eggs in an advanced stage of development hatched but all observed specimens failed to reach the adult stage. Adults submitted to an exposure of 600 milliamperes minutes, with spark gap of 5.5 inches, "giving

an approximate voltage of 65,000," and distance from focal spot of tube being 7.5, apparently lived the usual length of time but the large number of eggs deposited after exposure were infertile. Larvæ, receiving the same treatment, showed decreased activity and development, remaining in a dormant condition for a considerable period, and all died before reaching the pupal stage.

Parasitized Larvæ of Army-worm.—Tower ('16, Journ. Agr. Research, 6:455-458), in a comparative study of the amount of food eaten by parasitized and nonparasitized larvæ of *Cirphis unipuncta*, has found that when attacked by an internal parasite (*Apanteles militaris*), the parasitized larvæ of the army-worm ate approximately half as much food as unparasitized larvæ during corresponding periods, indicating that the parasitism becomes directly beneficial in the generation attacked. Four newly molted fifth-stage specimens when parasitized ate respectively 16.21, 12.16, 11.97, and 14.50 square inches of corn foliage during the last two stages previous to the emergence of the parasites while the average of twenty nonparasitized larvæ during the same stages was 33.6 square inches. Five partially developed fourth-stage larvæ when parasitized ate respectively 20.63, 17.36, 21.24, 17.64, and 17.99 square inches, while twenty nonparasitized larvæ ate, on the average, 34.77 square inches during the same stages.

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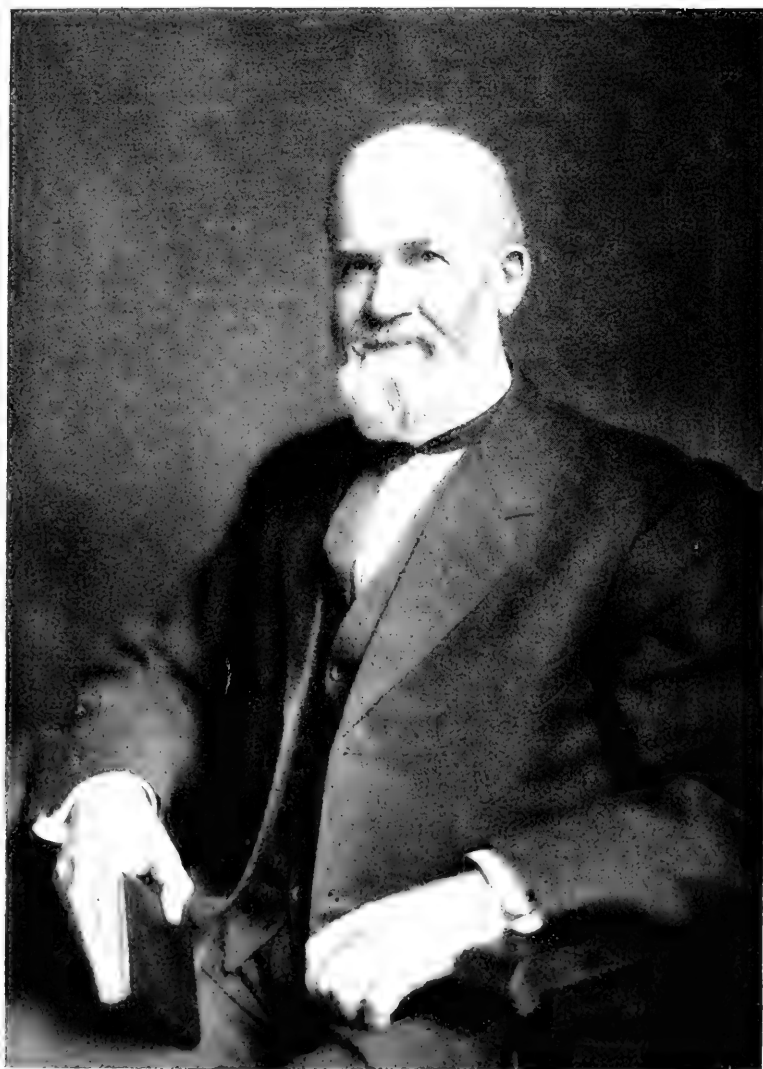


PLATE XI.

THOMAS JONATHAN BURRILL

THOMAS JONATHAN BURRILL

April 25, 1839

April 14, 1916

In the death of Professor Thomas Jonathan Burrill, the American Microscopical Society has lost one of its charter members whose activity in behalf of the Society may best be judged from his having twice served as its President and at another time as Secretary. A history of forty-four years of active service in one institution—the University of Illinois—where he watched the growth of teaching and investigation in natural science from the time when he was sole instructor in ‘natural history and botany’ to the time of his introduction of the laboratory method of instruction in his classes in that institution, and finally to the division of his work among four fully equipped independent departments,—all of them the direct outgrowth of his early enthusiasm for science; such is a history that is rarely recorded for any individual.

The first to discover a bacterial cause of disease in plants he opened a broad new field for investigation in bacteriology. His discovery of the cause of pear blight in 1880 assured for him a place as a leader in investigation but circumstances prevented his following this line of work. As one of his closest friends has expressed it “he loved people better than things, education better than science, and others better than himself; and he turned aside into what seemed to him the path of his duty, with no shadow of hesitation or appearance of regret, leaving it to others to note and remember his steady loyalty to his ideal.” This turning aside was in response to the constant and growing demands of educational

and administrative duties which prevented his devoting himself to extended investigations.

Professor Burrill retired from active service to the University in 1912. This same year he was made Professor Emeritus of Botany. His withdrawal from administrative duties gave him time even at his advanced age to begin again upon an active research program. Until within the week of his death he was busily engaged in attempting to induce nitrogen forming bacteria to grow on non-leguminous plants. His interest in modern problems of bacteriology was most fittingly recognized in his election to the Presidency of the American Society of Bacteriologists in December of the year preceding his death.

The development of biological equipment during a single life time is most vividly shown in the collection of microscopes and apparatus used by Professor Burrill and now on exhibition at the University of Illinois. One of his most intimate colleagues has promised an account dealing with his work and his apparatus for an early number of the Transactions.

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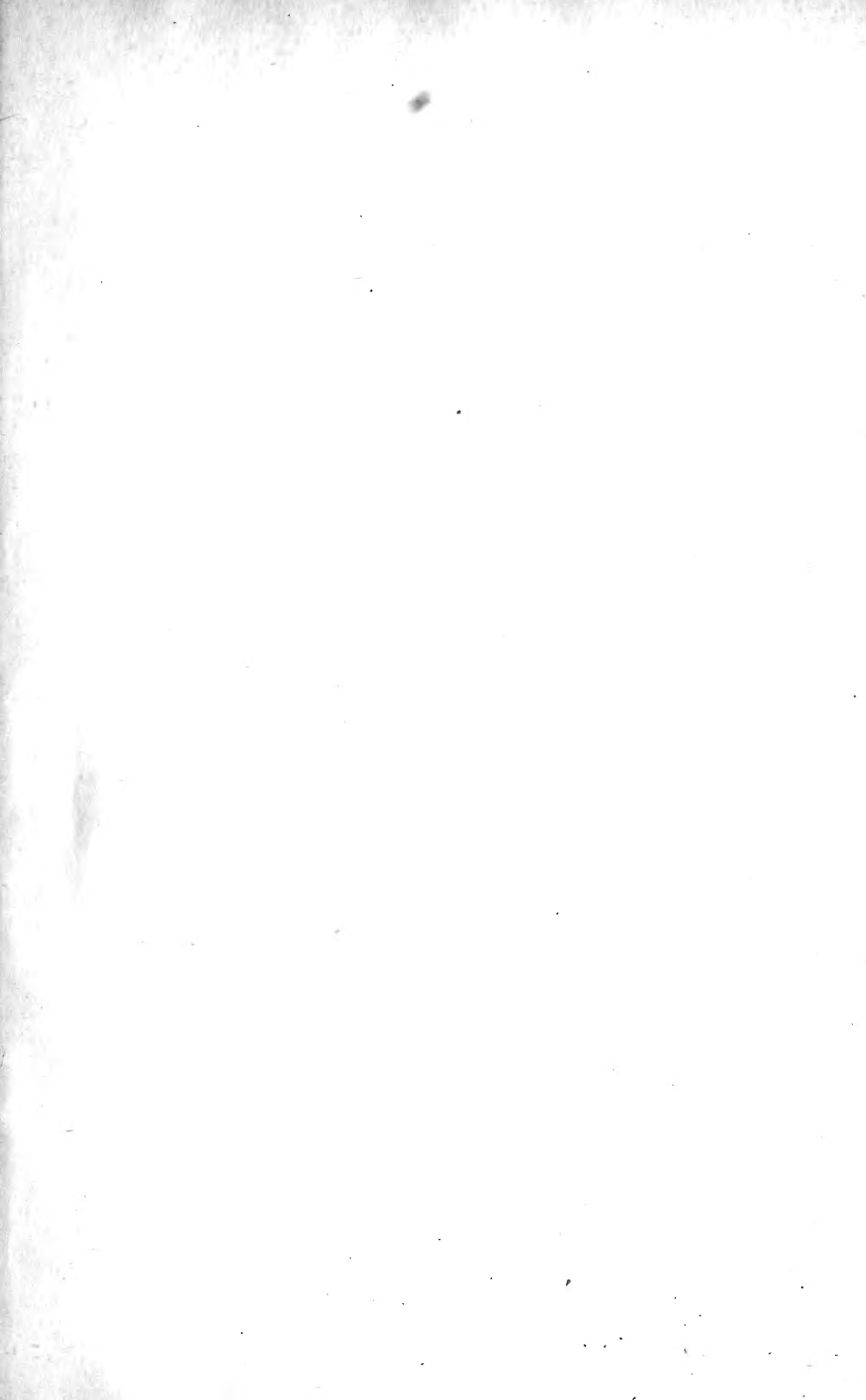
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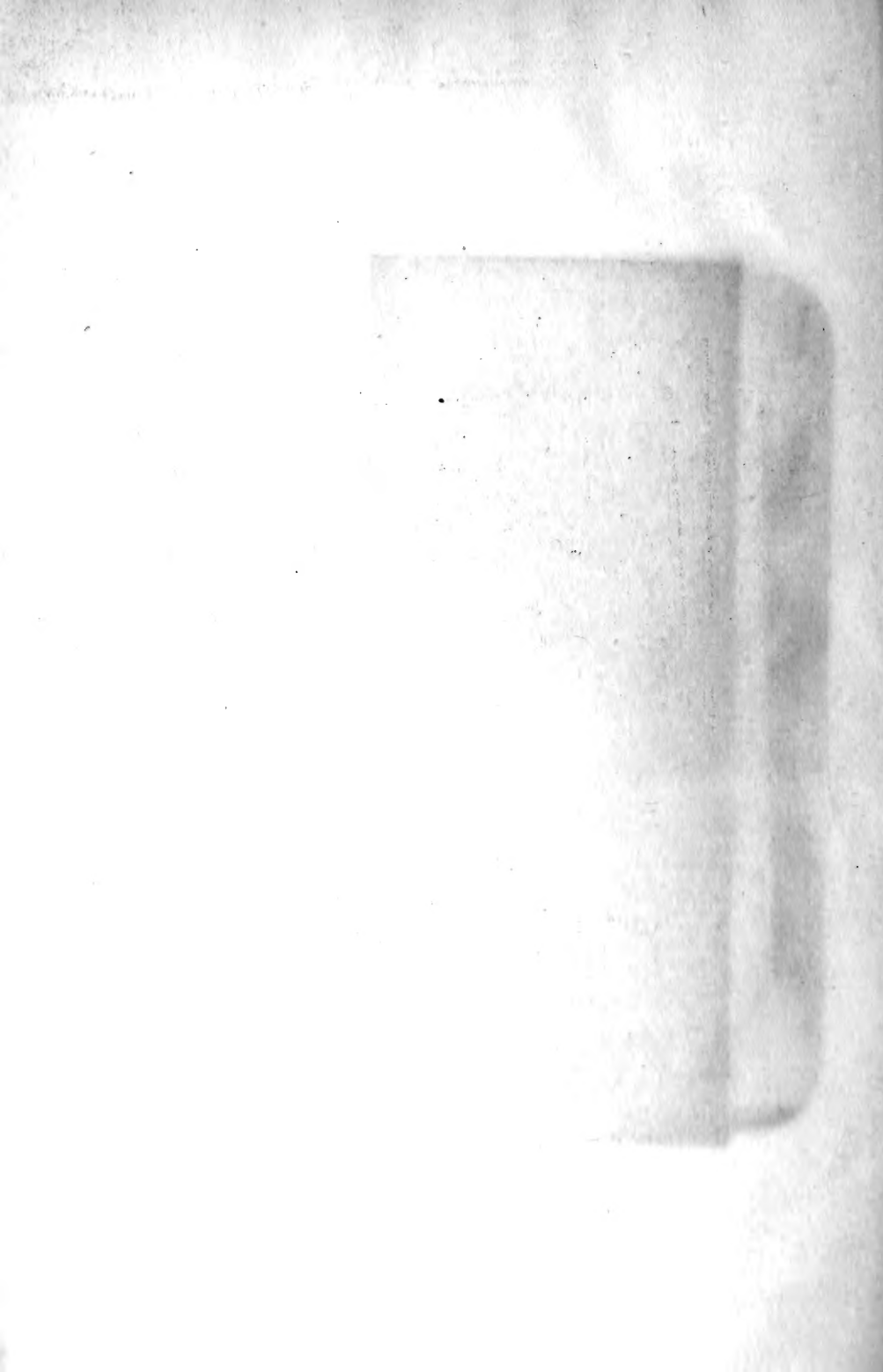
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