## WELCH

## Studies on the Enchytraeidae

## of North America

## Zoology Ph. D.

1913

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### STUDIES ON THE ENCHYTRAEIDAE OF NORTH AMERICA

BY

PAUL SMITH WELCH A. B. James Millikin University, 1910. A. M. University of Illinois, 1911.

#### THESIS

Submitted in Partial Fulfillment of the Requirements for the

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Studies on the Enchytracidae of north America

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I. INTRODUCTION.

In spite of the fact that the forms belonging to the family Enchytraeidae are common in many parts of North America, it is a group of which little is known. Less than a dozen references constitute the literature on the North American species. The writer has been carrying on investigations in this field in Illinois for the past three years and the following paper represents some of the results of this study and is offered as a thesis in partial fulfillment of the requirements for the degree of Doctor of Philosophy.

The writer wishes to express his indebtedness to Professor Frank Smith under whose direction this work has been done. Acknowledgements are also due the following persons:- the Director of the Sewage Testing Station at Chicago for permission to work in the laboratories of that institution: Dr. Arthur Lederer, Chief Chemist of the Sewage Testing Station, and his associates for the many courtesies extended to the writer during his work at that place: and Professor S. A. Forbes for material from the collections of the Illinois State Laboratory of Natural History.

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#### II. FAMILY ENCHYTRAEIDAE.

This family as known at present includes a large number of species distributed among sixteen genera. These worms are wide spread in their distribution, being common in various parts of the American continents. generally distributed in Europe and reported from Siberia, New Zealand and North Africa. They are found in various kinds of situations. Many are terrestrial, some are aquatic and others are reported as being common to either kind of situation. Although the majority of the aquatic forms are from fresh water several species are found along the shore in marine situations. They are quite similar to the earthworms in some respects; viz., (1) the simple setae; (2) the wide separation of the spermathecae from the spermiducal pores; (3) the presence of paired or unpaired glands comparable to the calciferous glands of the earthworms; (4) the thick tody wall. In other respects they resemble the lower oligochaetes; viz., (1) the presence of numerous lymphocytes in the coelomic rluid; (2) the limitation of the sperm ducts to two consecutive somites, one of which contains the internal opening and the other the external opening; (3) the reduction of the oviduct to a mere pore.

Definition of the family Enchytraeidae.

Michaelsen('00, p. 66) defines the family as follows:-

"Borsten stiftförmig oder einfach hackenförmig, ohne deutlichen Nodulus, gerade oder schwach S-förmig gebogen, einfach-spitzig, meist zu mehrenen (3-12) in facherförmigen Bundeln, selten zu 2, einzeln oder ganz fehlend. Kopfporus vorhanden. Nephridialporen vor den ventralen Borstenbändeln. Gurtel am 12. Segm. und über mehr oder weniger grosse

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teile der benachbarten Segm. Mannliche Poren 1 Paar, am 12 Segm., vor den ventralen Borstenböndeln; weibliche Poren 1 Paar, am 18. Segm., vor den ventralen Borstenböndeln; Sumentaschenporen meist 1 Paar, auf Intsegmtf. 4/5, selten 2 Paar auf Intsegmtf. 6/4 und 4/5. Darm mit dorsalen Schlundkopf, durch den mehrere Paare Septaldrösen, vor dem Bessep. 4/5 und einigen folgenden gelegen, ausmönden. Blutgefässesystem einfach; Röckengefäss nur im Vorderkörper, mit dem Bauchgefäss durch wenige, meist 3, Transversalgefässpaare verbunden. Weganephridisch; Nephridien mit massigen Postseptale. Hoden an Dissep. 10/11; Samentrichter mit dicken, drösiger Wandung und engem Lumen, walzen- oder tonnenförmig, selten schief trichterförmig, vor Dissep. 11/12. Ovarien an Diesep. 11/12; Eitrichter rudimentär, an Dissep. 12/18; Eier gross, dotterreich, einzeln oder su mehmeren in Cocons abgelegt. In einzelnen Fallen sästliche Geschlechtorgane mit Ausnahme der Samentaschen um 3 oder 4 Segm. nach vorn verschoben".

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III. SYSTEMATIC AND MORPHOLOGICAL STUDIES.

1. Preliminary discussion.

The Enchytraeidae is fast coming to be one of the larger families of the Oligochaeta. It now includes sixteen genera, as follows:-

| Mesenchytraeus Eisen. | Henlea Mchlan.             |
|-----------------------|----------------------------|
| Enchytraeus Henle.    | Fridericia Mchlan.         |
| Nichaelsena Ude.      | Distichopus Leidy.         |
| Lumbricillus Orst.    | Achaeta Vejd.              |
| Marionina Mohlan.     | Propappus Mchlan.          |
| Buchholzia Mchlsn.    | Euenchytraeus Bretscher.   |
| Stercutus Mchlen.     | Hepatogaster Cejka.        |
| Bryodrilus Ude.       | Hydrenchytraeus Bretscher. |

The genus Chirodrilus Verrill has been a subject of some dispute. Both Vejdovsky and Vaillant placed it among the Tubéfucidae while Beddard('95,p.314) and Michaelsen('00,p.88) placed it among the Enchytraeidae. Later Michaelsen('05,p.50) included it with the Tubificidse. As there seems to be good grounds for putting it with the Tubificidae, it has been omitted from the above list.

The richness of Enchytraeid life in North America is indicated by the fact that in spite of the small amount of investigation which has been made in this group, nine of the above listed genera are known to have representatives on this continent and it seems safe to predict that future investigation will reveal still other genera as yet unknown. A promise of what future investigation may reveal is shown in the work of Eisen on the Pacific Coast where random collecting gave material from

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which he described forty eight new species distributed among eight genera and in his introduction he states that he had at the time of writing come fifty or more additional new species from the same region, the publication of which was at the time prevented by certain existing circumstances.

Of the above listed genera only Distichopus is, so far as our knowledge goes, limited to North America. It was described by Leidy in 1882 from Eastern United States. The genera represented in North America are as follows:-

| Mesenchytraeus | Bryodrilus  |
|----------------|-------------|
| Enchytraeus    | Henlea      |
| Michaelsena    | Fridericia  |
| Lumbricillus   | Distichopus |
| Marionina      |             |

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|--|--|--|
| 2. KEY TO THE GENERA OF ENCHYTRAEIDAE OF NORTH AMERICA.              |  |  |
| A. Setae not disposed in bundles; occur singly when present; usually |  |  |
| absent on many of the somites MICHAELSENA.                           |  |  |
| AA. Setae disposed in bundles.                                       |  |  |
| B. Setae disposed in two bundles on each somite DISTICHOPUS.         |  |  |
|  |  |  |
| BB. Setae disposed in four bundles on each somite.                   |  |  |
| C. Dorsal pores present FRIDERICIA.                                  |  |  |
| CC. Dorsal pores absent.   |  |  |
| D. Oesophagus merges suddenly into intestine HENLEA.                 |  |  |
| DD. Oesophagus merges gradually into intestine.                      |  |  |
| E. Setae straight and of equal length ENCHYTRAEUS.                   |  |  |
| EE. Setae signoid.   |  |  |
| F. Testes pleurilobed LUMBRICILLUS.                                  |  |  |
| FF. Testes undivided.  |  |  |
| G. Origin of dorsal blood vessel intraclitellar;                     |  |  |
| blind diverticula in connection with alimentary                      |  |  |
| canal somewhere in VI-VIII BRYODRILUS.                               |  |  |
| GG. Origin of dorsal blood vessel postclitellar;                     |  |  |
| no diverticula in connection with anterior                           |  |  |
| part of alimentary canal.  |  |  |
| H. Nephridia with a wide, closely wound canal                        |  |  |
| and slight intermediate substance                                    |  |  |
| MESENCHYTRAEUS.  |  |  |
| HH. Nephridia with narrow, loosely coiled canal                      |  |  |
| and well developed intermediate substance                            |  |  |
| MARIONINA.   |  |  |
|  |  |  |
|  |  |  |



#### 8. Genus HENLEA Mohlen.

The genus Henles was established by Michaelsen in 1889 and although it includes a somewhat heterogeneous assemblage of species there does not seem to be at present sufficient grounds for breaking up the genus into several generic types. It is distinguished fr.cm the other genera by the following characters: - (1) The sudden change in the diameter of the digestive tract where the oesophagus passes into the intestine; and (2) the anteolitellar origin of the dorsal blood vessel. As a rule there are diverticula at the beginning of the intestine. Henles has Bryodrilus for its nearest relative although Buchholzia also stands close. Michalsen ('03, p. 51) in discussing the phylogeny of the Enchytraeidae places Henlea at the base of the system as the most primitive genus on the ground that the forms belonging to this group show the greatest diversity in the character of the setae. Later the same writer ('05, p. 24) described a new genus, Propappus, which is in a number of respects more primitive than Henles and must be regarded as the oldest of the known Enchytracid genera. One of the striking characters of this new genus is the presence of forked setse.

Forty two species and two varieties are included in this genue at the present time. Of this number seven are doubtful either as regards their being valid species or as regards the correctness of their position in this genus. Of these the following seem worthy of notice:- H. lefroyi, described by Beddard('05,p.62) from India and placed provisionally in this genus, is described as having the origin of the dorsal blood vessel intraclitellar, the intestinal diverticula lacking, and the oesophagus as passing gradually into the intestine. Beddard finds it possible to

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eliminate mafely, by other characters, all of the genera except Bryodrilus and Henles. He places the species in Henles because the characteristic intestinal diverticuls are sometimes absent, a reason which is open to question. H. scharffi, described by Southern ('10, p. 18) from the White Nountains and also placed provisionally in this genus, is described as having no intestinal diverticuls and the cesophagus passes gradually into the intestine. The anteolitellar origin of the dorsal vessel seems to be the only good ground for placing the species in this genus.

Taking the genus as a whole there is a remarkable variation in the different organs. H. puteana Vejd. is unique in having two pairs of spermathecae. The species of the genus can be grouped in a number of ways depending upon the character of the setae, the presence of absence of intestinal diverticula, the presence or absence of peptonephridia, the place of origin of the dorsal vessel, and the presence or absence of ampullae on the spermathecae.

Of the assemblage of species included under Henlea, four species and two varieties have been described from North America. They are as follows:-

|    | Nare                             | Type_Locality                       |
|----|----------------------------------|-------------------------------------|
| H. | californica Eisen                | Santa Rosa, Sonoma Co., Calif.      |
| Ħ. | Californica var. monticola Eisen | West Fork, Feather River, Calif.    |
| H. | californica var. helenae Eisen   | St. Helena, Napa Co., Calif.        |
| H. | guatemalae Eisen                 | Guatemala City, Central America.    |
| H. | ehrhorni Eisen                   | Mountain View, San Mateo Co. Calif. |
| H. | scharffi Southern                | White Mountains, N.H.               |

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Leidy(1850, Jour. Ac. Soi. Phil. ser. 2, v. 81, p.48) described a epecies under the name of Enchytraeus socialis from eastern Pennsylvania. The description is so inadequate that it is very uncertain as to just what this species is. Michaelsen('00,p.69) places it as a synonym of Henles ventriculoss Udek. and there appears to be evidence in favor of this view. It thus appears that Henles ventriculoss Udek. may be added tentatively to the list of North American species of this genus.

#### HENLEA Mohlsn.

Michaelsen('00, p. 67) defines the genus as follows:-

"Borstan gerade oder schwach S-förmig gebogen. Kopfporus klein, zwischen Kopflappen und I Segn.; Röckenporen fehlen. Lymphkörper von einerlei Gestalt gross, meist discusförmig, selten ehliptisch, dunkel granuliert. Der Oesophagus geht in 7., 8. oder 9. Segm. plotzlich in der weiten Mitteldarm über. Ursprung des Röckengefässes anteolitellial in 8. oder 9. Segm.; Blut farblos; Herzkörper fehlt. Nephridien mit kleinem, einfachem Anteseptale. Hoden massig. Samentaschen einfach, ohne Divertikel, mit dem Oesophagus kommunizierend."

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KEY TO THE SPECIES OF THE GENUS HENLEA KNOWN TO OCCUR IN NORTH AMERICA.

A. Spermathecae with diverticula - - - - - H. shrhorni Eisen. AA. Spermathecae without diverticula.

B. No intestinal diverticula present - - H. scharffi Southern.
BE. Intestinal diverticula present.

C. Two intestinal diverticula present.

- D. Spermatheca with ampulla having diameter little or no greater than duct; anteseptal region of nephridia small.
  - E. Dorsal vessel arises in VIII; peptonephridia connected with digestive tract in IV.
    - F. Two accessory glands at the ectal opening of spermathecal duct; spermathecae slightly bent.
      - G. Brain wider than long, concave anteriorly and posteriorly; spermathecae with lumen approximately straight - -H. californica Eisen.
      - GG. Brain almost square with posterior margin only concave; spermathecae with an expansion in lumen for storage of spermatozoa which is connected with lumen of intestine by a long narrow contorted canal - - -- - H. californica var.helenae Eisen.
    - FF. Four or more accessory glands at ectal opening of spermathecal duct; spermathecae sharply tent - - H. califormica var.monticola Eisen.

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- EE. Dorsal vessel arises in IX; peptonephridia connected with digestive tract in V - - - -- - H. urbanensis n. sp.
- DD. Spermatheca with well developed oval ampulla; anteseptal part of nephridia approximately as long as the postseptal part - - - H. guatemalae Eisen.

CC. One intestinal diverticulum which completely surrounds the digestive tract in VIII - - - H. moderate n. sp.

#### Henles moderata n. sp.

Plate I, figs. 1-12.

#### Definition .-

Length 18-19 mm., average about 16 mm. Diameter C.48 mm. Somites 46-58. Color whitish yellow. Prostomium somewhat tapering. Head pore at O/I. Doreal pores absent. Setae of unequal length, inner ones slightly shorter; slightly bent; varying in size, inner ones finer; 8-6 per bundle in anterior part of body, 4-2 in posterior part. Clitellum on 1/2XI - XIII. Lymphocytes elliptical. Brain about one third longer than wide; anterior margin concave; posterior margin deeply emarginate; lateral margins converge rapidly cephalad. Peptonephridia present and well developed, connected with alimentary canal in V; dorsal and ventral strands lie in close contact with alimentary canal; ventral strand gives rise to a number of tubules in VI and VII which project into the coelom. Four "taste organs" in buccal cavity each provided with a muscular strand which extends to body wall. Ossophagus passes abruptly into

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intestine in VIII. Intestinal diverticulum present in VIII; entirely surrounde digestive tract; composed of numerous tubules which ultimately unite into about twelve main tubules by means of which connection with digestive tract is effected. Dorsal vessel arises in IX. Each nephridium with small anteseptal part; postseptal part about one and one half times larger; efferent duot arises from anterior part of latter. Spermiducal funnel small; length about twice diameter. Spermathecae with owal expanded ampulla near ectal end; duot diminisging in diameter towards ental end; rosette of four glands at ectal opening of duot; duots unite dorsad of digestive tract to form a short oval tube through which they communicate with lumen of digestive tract in posterior part of V.

Described from 11 sexually mature specimens. Cotypes in the collection of the writer. Cotypes also in the collection of Professor Frank Smith.

The specimens which are the basis of this description were found near Urbana, Illinois, in rich soil and under decaying leaves in undisturbed forest land, in late March, 1911. All of the specimens were sexually mature showing spermatozoa in the spermathecae, well developed egg masses in the body cavity, and developing spermatozoa in XI.

#### Affinities.

It is somewhat difficult to determine the systematic relation of this species owing to the fact that some of the species which are included in the genus are so incompletely described. Species which, so far as described seem to be closely related, might, if more thoroughly worked

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out, reveal characters which would separate them widely. However, until these meager descriptions are supplemented by further study one must be content to place any new species as nearly as possible to what seems to be its natural position in the genue. If the minimum number of distinct differences be considered this species seems to approach H. gemmata Eisen, H. ochraces Eisen, and H. dorsalie Bretscher, all of which are imperfectly described and as a consequence the assumption of this relationship must be tentative. H. gemmata Eisen differe from H. moderate in the character of the setse, the spermiducal funnel, the nephridia, the brain and the spermathecae. H. ochraces Eisen differs in the character of the brain, nephridia and spermathecae. H. dorsalis Bretscher shows differences in the length and in the character of the brain.

#### DETAILED\_MORPHOLOGY.

#### EXTERNAL CHARACTERS.

The body is slender and has an average length of about 16 mm., the extremes being 18 and 19. In transverse section the body is circular. The diameter is greatest in the region of the olitellum where it averages about 0.47 mm. Posterior to the clitellum the diameter diminishes only to a very slight degree. In the living specimens the body is opaque and yellowish-white in appearance. The prostomium(Fig.6) shows a slight but gradual tapering. The intersegmental grooves are quite distinct in the first S-5 somites but elsewhere are obscure. The number of somites is variable, the average being about 54, the extremes 46 and 58. The head pore is small and located on O/I. The clitellum occupies 1/2 XI-XIII and is usually well developed. In the anterior region there are 8-6 setae

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per bundle and 4-2 in the posterior region. In the last four or five somites there are usually 2 setse per bundle. The arBangement of the setse in the bundle(Fig. 3) resembles that of Fridericia. The component setse of a bundle are of different lengths, the outer ones being longer and heavier, the inner ones shorter and finer. It should be noted that in this species there is not so great a difference in the size of the setse of a bundle as is usually found in Fridericia. In each bundle the proximal ends of the setse are in rather close proximity to each other and are arranged in transverse linear sequence. Outside of the body wall they spread out fan-wise. Close examination shows that, as in Fridericia, the setse are not arranged in pairs but represent a series of different sizes. The proximal ends(Fig.9) are distinctly bent.

#### INTERNAL CHARACTERS.

Lymphocytes.

The lymphocytes(Fig. 5) are large and abundant: Their distribution in the coelom is not uniform but certain regions are well supplied while other regions are almost destitute. They begin to appear near IV. The intervening space between the septal glands and the reproductive organs is almost completely filled with them except in the region of the intestinal diverticulum where they are greatly reduced in numbers. Few if any are present in the somites which contain the reproductive organs but beyond them the lymphocytes are always present although not in such numbers as in the anterior part of the body. They are disc-like or broadly elliptical. The granular cytoplasm contains a conspicuous nucleus. Measurements average as follows:- length - 9.085; width 0.045.

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Brain.

The brain(Fig. 1) occupies a median dorsal position in II, projecting into III for a distance of approximately 1/5 the length of the brain. It also projects into I for a very short distance. The brain is somewhat heart shaped. The posterior margin is deeply emraginate and the anterior margin is decidedly concave. The lateral margins converge rapidly cephalad and approach each other closest at a point just caudad to the origin of the commissural trunks. Measurements shows that the posterior part of the brain is approximately 2.7 times wider than the anterior part. Measurements of three brains which had been dissected out show the dimensions of that organ to be practically uniform, the ratio of the the greatest width to the greatest length being 6 : 9. The actual measurements are 0.108 mm. for the greatest width, and 0.162 mm. for the greatest length. In transverse section the organ is elliptical in outline. The brain is anchored to the body wall by two pairs of supporting strands which arise from the latero-posterior and posterior parts of the organ. The anterior region gives rise to the usual nerve trunks which extend cephalad, diverging only slightly up to the point where they begin to extend around the oesophagus. Near this point these trunks divide, giving rise to a pair of trunks which extend into the prostomium. The main pair of trunks extend around the digestive tract forming the circum-ocsophageal commissures, uniting again on the ventral side to form the sub-oesophageal ganglion which lies partly in I and partly in II.

Peptonephridia.

Two peptonephridia are present, one arising from the doreal and the other from the ventral side of the digestive tract in the anterior part

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of V. Both adhere closely to the alimentary cindal as they extend caudad and for a considerable part of their extent lie between the epithelial layer and the muschfar coat of the oesophagus. Each is composed of two strands which give off at intervals a number of tubules that project freely into the coelom. In VI both the dorsal and ventral peptonephridia become greatly thickened and enlarged. Near this enlargement the ventral peptonephdidias give rise to a number of tubules which escape from the wall of the digestive tract and extend around it on either side. The termini of these tubules lie around the dorsal vessel or in its immediate vicinity. They come into close proximity to the dorsal strands but so far as observed do not unite with them. After giving off these tubules the strands become reduced to their former size and continue thus as far as VII where the ventral peptonephridian becomes thickened again and gives rise to tubules which project freely into the body cavity. These tubules resemble those in VI in the general structure but differ in being shorter and fewer in number. They extend dorsad on each side of the digestive tract but do not reach the dorsal side. Both the dorsal and ventral peptonephridia end immediately anterior to the intestinal diverticulum in the posterior part of VII.

#### Taste Organs (Geschmackslappchen)

This species is somewhat unique in having four of these organs (Fig.8) instead of the usual number two. They extend from the floor of the buccal carity, are products of the lining epithelium and are structally like it. These tongue-like organs are sometimes directed caudad, sometimes cephalad, depending upon the state of retraction of the pharynx. The surface is covered by the cutusula which lines that part of

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digestive tract. Four muscular strands, two on either side of the median line, are attached to the wall of the buccal cavity at the bases of these organs and extend obliquely ventro-caudad to the body wall. Vejdovsky('84,p.99) describes ganglion cells in the bases of the "Gdschmackslappohen" which he studied but they have not been seen in the preparations of this species.

#### Intestinal divertioulus.

At the junction of the intestine with the oesophagus in VIII there arises a structure which is reflected cephalad over the latter, investing it closely for the greater part of the length of the somite. In cleared mounts it appears as a brownish, almost opaque mass filling the greater part of the coelom at the above mentioned position. It is a single organ with two shallow longitudinal depressions, one dorsal, and the other ventral, the former being the more distinct. Sections(Figs. 10 and 11) show the organ to be made up of a series of branching, rather thick walled tubules, about twelve in number, which extend radially and cephalad. The region of the digestive tract(Fig. 10) from which these tubules arise is ciliated and and the basal parts of the lumina of the tubules (tdo) are also cilisted. These tubules give off branches as they extend cephalad until the whole mass of the anterior part of the organ is composed of finer tubules(Fig. 11, td) which lie in very close proximity to each other. The walls of these tubules are distinctly nucleated and appear to be composed of glandular tissue. The whole diverticulum is invested in a layer of peritoneum beneath which is a very much reduced muscle layer, a continuation of the muscle layer of the digestive tract. The perivisceral blood sinus(Fig. 10, bsp) appears at the point of origin

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of the tubules from the digestive tract. This sinus is continued cephalad and the reduced spaces(Fig. 11, bs) which appear between the tubules of the diverticulum are continuations of this sinus. In all of the specimens studied the structure of this organ is uniform in all respects.

The presence of the mid dorsal and mid ventral longitudinal grooves suggests the possibility that this organ may have developed from two lateral outgrowths from the digestive tract which grew together and fused at the points of contact. However an examination of the point of origin of the diverticulum shows no evidence that it arose as two separate parts.

#### Dorsal blood vessel.

The dorsal blood vessel arises from the perivisceral blood sinus in IX. In some of the specimens it shows a conspicuous expansion in IX immediately after it originates from the sinus but this is not a constant feature since some specimens do not show it at all while others show only a moderate expansion.

#### Nephridia.

The first nephridia appear on V/VI. There is some variation is size and shape in the various specimens and in the different regions of the body although this variation is within rather narrow limits. The anteseptal part(Fig. 2) is reduced in size, the postseptal part being about two and one half times larger. The efferent duct is longer than the postseptal part and arises near the septum, opening to the exterior just cephalad to the ventral setae.

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Spermiducal funnel.

The spermiducal funnel(Fig.4) is small in comparison to the size of the body. It is situated in the posterior part of XI with its base in close proximity to the lower part of XI/XII with its long axis almost parallel to the long axis of the body. The whole organ lies close to the ventral body wall. It varies in shape within narrow limits but in general it resembles an elongated cask. It also varies somewhat in its dimensions but on the average the length is about twice the diameter. The anterior end has a well differentiated collar which is set off from the body of the organ by a constriction. This collar varies in the degree of the reflection of the margin which is sometimes reflected to the extent of about 180° and in other preparations only about 45°. The anterior opening is in close proximity with the extremity of the testis.

The sperm duct extends through XI/XII very near its union with the body wall. It is long, much coiled, and confined to XII.

#### Penial bulb.

This organ(Fig. 12) conforms to the Lumbricillid type of penial bulb as defined by Eisen('04, p. 8) and does not differ markedly in structure from that of the other American species of this genus. It is small and is not nearly so conspicuous in transverse sections as is usually the case in other Enchytraeid species. It is covered by a definite musculature (m), a continuation of the circular muscle layer of the body wall, which does not at any point penetrate into the Dody of the bulb. The bulb is composed of two kinds of cells, namely, those surrounding and opening into the sperm duct extension\* (ibc), and those which fill

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the peripheral parts of the bulb (glp) some of which appear to open to the surface below the penial pore (pp). The former are elongated, nucleated, and stain very lightly. They are arranged radially around the sperm duct extension (sde). The peripheral cells are irregularly spindle -shaped and tend to take the stains heavily. The sperm duct (sd) penetrates the bulb near the ectal mide and joins the sperm duct extension (sde) well within the body of the bulb. When the penial bulb is in the retracted state the sperm duct extension curves strongly towards the penial invagination (pb) and the penial pore (pp) is located well towards the base of the latter.

#### Ovaries.

These organs occur as usual in XII attached to the ventral part of XI/XII. They are massive, filling a considerable part of the coelom in XII. The terminal part of each which bears the developing mass is usually pushed up into the body cavity until it lies dorsad to the digestive tract.

#### Spermathecae.

A single pair of these organs lie in V. The ectal opening of each ' is laterad in the intersegmental groove IV/V and is surrounded by a number of glands(Fig. 7) which forms a sort of rosette. There is no differentiation of duct and ampulla. Within the ectal region which is somewhat swollen the lumen attains its maximum diameter. This swollen

\* In this paper the name "sperm duct extension" will be used to designate that differentiated part of the spermduct which lies within the penial bulb, extending from the terminus of the sperm duct proper near the periphery of the bulb to the penial pore.

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region involves about one half of the entire spermathece. The diameter decreases entad, the swollen region merging gradually into the ductlike portion which extends obliquely across the coelom to a point dorsad of the digestive tract where it bends caudad. It meets and unites with the duct of the spermathece of the opposite side in the posterior part of V to form the large common duct(Fig.7) which soon enters the digestive tract.

#### Henles urbanensis n. sp.

Plate VI, figs. 56-58.

#### Definition.

Length 25 mm. Diameter 0.57 mm. Color whitish yellow. Prostomium blunt and rounded. Head pore at O/I. Dorsal pores absent. Setae of unequal length, the inner ones slightly shorter and finer: slightly bent; 6-8 in ventral bundles in anterior part of body, usually 8; 4-6 in lateral bundles: about 7 in ventral bundles in middle region; 4-6 in lateral bundles. Clitellum on 1/2 XI - XIII. Lymphocytes elliptical, large. Brain about as wide as long: anterior margin concave, posterior margin emarginate; lateral margins diverge cauded. Peptonephridis present and well developed, connected with digestive tract in V; dorsal and ventral strands lie in close contact with digestive tract; both dorsal and ventral strands show conspicuous thickenings at origin which extend freely into coelom; dorsal strand gives rise to tubules in VI, the ventral gives rise to similar tubules in VI and VII. Two "taste organs" in buccal cavity. Ossophagus passes rather abruptly into intestine in VIII. Two lateral sac-like diverticula in VIII; cavity of each

-21-

communicates with lumen of digestive tract by one lateral opening. Dorsal vessel arises in IX. Nephridia with small anteseptal and large well developed postseptal part; duot arises from ventral surface of latter near septum. Spermiducal funnel moderate, length about two and one third times greater than diameter. Spermatheca not strongly developed; no distinct ampulla; diameter greatest at region of ectal opening where it is surrounded by a rosette of glands; communication with lumen of digestive tract on dorsal side effected by a short channel formed by the fusion of the two spermathecas at their ental ends; ectal opening near IV/V.

Described from one sexually mature specimen. Type in the collection of the writer.

The specimen which is the basis of this description was found in woods near Urbana, Illinois, in the rich soil of undisturbed forest land, in late March, 1911. The specimen is sexually mature since it shows the presence of spermatozoa in the spermathecae, well developed egg masses in the body cavity and developing spermatozoa in XI.

#### DETAILED MORPHOLOGY.

#### EXTERNAL CHARACTERS.

The length of the type sepcimen is 25 mm. The body is long, slender, and in transverse section is circular in outline. The greatest diameter is in the region of the clitellum where it measures 0.576 mm. The body is opaque and of a whitish yellow color in the living specimen. Unfortunately the data regarding the number of somites has been lost and the type series of transverse sections includes only fifteen somites, hence

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that point must be left until additional material is secured. The prostom -ium is rather blunt and rounded. The intersegmental grooves are rather distinct. The head pore is at O/I. The clitellum is well developed and occupies 1/2 XI - XIII. Near its middle the cells are about eight times longer than their width. There are no specially differentiated areas.

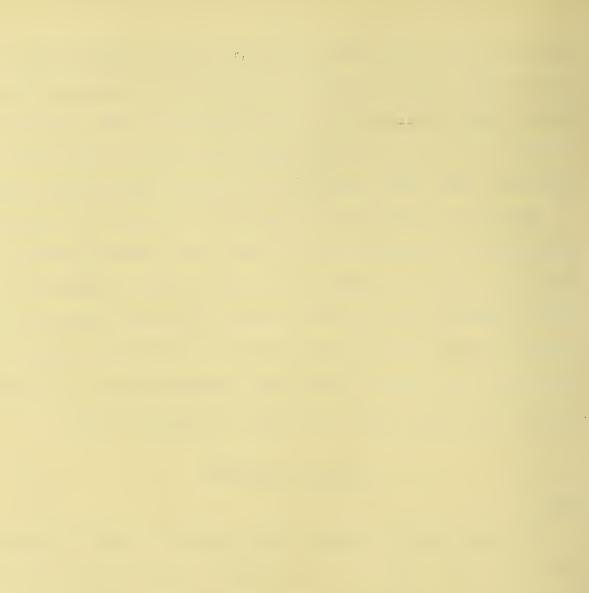
The shape and arrangement of the setae correspond very closely to the condition in Henles moderata but there is a distinct difference in the number of setae per bundle. In the first thirteen somites the ventral bundles contain 6-8 setae, usually the latter number; the lateral bundles contain 5-6 setae, never more than 6. The middle region of the body usually has 7 setae in the ventral bundles and 4-6 in the lateral bundles. The proximal ends of the setae are distinctly bent.

#### INTERNAL CHARACTERS.

#### Brain.

The brain occupies a median doreal position in I and II, the greater part of the organ beind in II. The length is about the same as the greatest width, the measurements beind as follows:- length 0.146; greatest width 0.142 pp. The posterior margin is distinctly emarginate and the anterior margin is guite concave. The smallest width, 0.125 mm., is in the region of the origin of the commissural trunks. From this point the lateral margins diverge caudad until they reach the region of greatest width which is about midway of the length of the organ. Thence the lateral margins round off gradually into the posterior margin. In transverse section the brain is elliptical in outline. The anterior region gives rise to the usual commissural trunks which extend around the alimentary canal, one on either side and meet to form the sub-cesophageal

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ganglion in II. One pair of supporting strands extends from the sides of the brain to the body wall: another pair extends from the two Terminal lobes to the body wall.

#### Peptonephridia.

These organs ars two rather complicated structures arising from the dorsal and ventral surfaces of the alimentary canal in the anterior part of V. They recemble those of Henles moderats in the general plan of structure but present certain marksd differences. The dorsal and ventral psptonephridia are quite dissimilar. The dorsal one, at its origin, gives rise to two divisions, one sotal and the other ental. The ectal portion is a large, thick walled, tubular structure which extends fresly into the coelon. Immediately beyond its origin this ectal portion assumes a position parallel to the alimentary canal and extends caudad for about the length of one somite. The ental portion has a very intimate relation to the wall of the digestive tract since it lies under ths outer coat of the latter. Immediately anterior to the third pair of septal glands it gives rise to two irregular, branching outgrowths, one on the right and one on the left side, which escape from the enclosing sheath of the digestive tract and extend freely into the coelom showing a tendency to lie in close proximity to the dorsal vessel. The main part of the dorsal peptonephridium continues caudad, maintaining its intimate relation to the digestive tract, and ends just anterior to the origin of the intestinal diverticula. The ventral psptonsphridium is also composed of two parts, one ectal and the other ental. The ectal division resembles the corresponding dorsal ectal one in structure and mass but differs in shape and distribution. Immediately beyond its origin it

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enlarges and extends into the body cavity. It is composed of two parts, one of which extends cephalad for a distance of about one half a somile forming about your fifths of the bulk of the structure, whereas the other extends caudad for a short distance. The former is unbranched. The ental part resembles the corresponding dorsal one in structure and in its relation to the digestive tract. Just anterior to the third pair of septal glands it gives off right and left branches which project freely into the body cavity, extending dorsal around the alimentary canal. These branches are in close proximity to the corresponding ones of the ventral peptonephridium but do not unite with them. The main part of the ventral peptonephridium continues caudad, maintaining the same intimate relation to the digestive tract, until a point just anterior to the intestinal diverticula is reached where it again gives rise to right and left branches which extend into the coelom. These oranches are similar to the corresponding anterior ones except that they are not so extensive. The main part of the gland which is longer than the dorsal peptonephridium, ends just anterior to the origin of the intestinal diverticulus. All of the various parts of these organs have essentially the same structure. They are rather thick walled, tubular, and conspicuously nucleated. The peculiarly thickened postions at the origin of the glandsvary in staining capacity from the other parts and to some extent recall the condition of the peptonephridia in Henlea leptoders Vejd. as figured by Vejdovsky('79, Taf. X, fig. 2).

Taste bodies.

A pair of these organs arise from the floor of the buccal cavity one on either side of the median line and extend out into the lumen.

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They are about 0.057 mm. long. The basal part of each is somewhat constricted forming a sort of pedicel. The remainder of the organ is rather epindle-shaped, thick at the middle and tapering off to a point at the extremity. The body of each of these organs is composed of elongated, nucleated cells which resemble the other epithelial cells of the lining of that part of the digestive tract in structure and staining reaction. The extremities are characterized by the disappearance of all traces of cell walls and nuclei, thus presenting a somewhat homogeneous appearance. The entire outer surface is covered by the cuticula which lines the buccal cavity. Each organ is provided with a muscle which extends ventrocaudad from its base to the body wall.

#### Intestinal diverticula.

The intestinal diverticula comprise two lateral sac-like evaginaticks which arise from the intestine in the posterior part of VIII. They extend cephalad from the point of origin and fill the greater part of the coelom in that region. Their dimensions increase from the point of origin towards the anterior part of VIII. They are somewhat flattened laterally, the greatest diameter being in a dorsi-ventral direction. The structure of these organs(Fig. 57) is very interesting. Each diverticulum contains a large central cavity (cd), a continuation of the lumen of the intestine, which communicates with it by a single, dorsilateral, slit-like opening. One of the characteristic features of the structure of this organ is the intricate folding of the inner lining. The dorsal portion of the side (ens) adjacent to the alimentary canal shows but little if any folding. However, the entire opposite wall is

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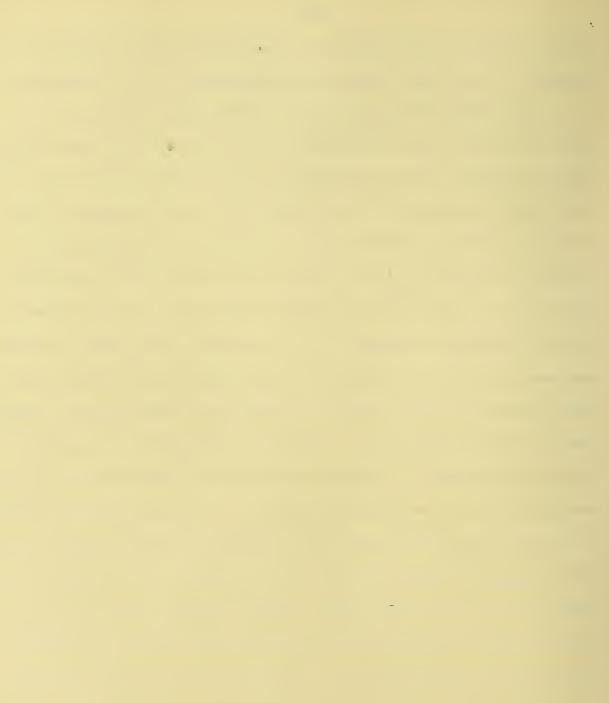
conspicuously folded. This folding which involves most of the thickness of the wall is found all along the ectal and ventral sides. Examination with high power shows a series of blood sinuses (be) which are intimately related to the walls of the diverticula. In the anterior part of each organ they are inconspicuous being confined to small spaces in the walls and folds. Toward the intestinal connection they become more apparent and occupy the more or less continuous space between the parts of the walls as well as the numerous spaces in the folds as indicated in figure 57. These spaces in the folds appear slit-like in transverse sections. The sinuses increase in size and diminish in numbers caudad until finally there appear a few large sinuses which result from the union of the smaller ones. Ultimately there appears one very large sinus which is the main channel of connection with the perivisceral sinus at the junction of the diverticulum with the intestine. The ectal surface of the diverticulum is to some extent covered with chloragog cells(chc). The ental surface(ens) shows no such cells but is covered with a layer of peritoneum of the usual type. The wall of the diverticulum is composed of (1) an external peritoneum, either modified into chloragog cells or of the simple type, (2) a middle region occupied by a blood sinus, and (3) an inner rather thick, greatly folded, non-ciliated epithelium. Just cephalad to the junction with the intestine the epithelium begins to take on the appearance of the lining epithelium of the intestine and has a similar staining reaction. This communication is ciliated for a short distance. The staining reaction of the bulk of the diverticulum is quite different from that of the epithelial lining of the intestine from which it originates. At the point of union of the diverticulum

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with the digestive tract there is a marked and abrupt increase in the diameter of the latter which marks the beginning of the intestine.

A comparison of the structure of these organs with that of H. moderata reveals a wide difference in the two species. In the latter a totally different plan of structure is found. Owing to the meager treatment in the literature of these structures in other species of Henles it is not possible to make extensive comparisons. Michaelsen('86) figures the structure of the intestinal diverticula in H. leptodera Vejd. and in H. ventriculosa D'Udekem. Both conform to the general plan of structure as presented in H. urbanensis. They have one rather spacious central cavity bounded by walls which show infoldings and which contain the blood sinuses in somewhat the same relation. However they are quite different from those of H. urbanensis as indicated by the following facts. H. ventriculosa has four diverticula which alone distinguishes it from the other species. H. leptodera has two diverticut la but they differ from those of H. urbanensis since the folds in the inner lining are larger and much fewer in number and are present on the ental as well as the ectal wall. Also the branches of the blood sinus are fewer in number. Michaelsen('89) describes and figures the structure of the intestinal diverticulum in H. nasuta. It also presents the same general type of structure as H. urbanensis but the folds of the lining membrane are much wider and more irregular than in the latter. The folds also show branching, a condition which does not appear in H. urbanensis. Furthermore according to Michaelsen's figure there is no difference in the structure of the ectal and ental walls. Eisen('04, p. 100) makes the following brief statement concerning these organs in H. californics:-

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" Intestinal pouches in VII are similar to those figured by Wichaelsen from H. nasuta. The villi are fully as intricately folded". The same writer( 1.0.p.100) makes the following meager statement concerning H. guatemalae:- "Intestinal pouches in VII; epithelium with comparatively few folds". Again Eisen( 1.0.p.105) makes the following statement concerning H. ehrhorni:-"Intestine.- The tubular part is furnished in VIII with a pair of diverticles which not only fill the greater part of VIII but also project into VII. The inner lobes of the diverticles are much coarser than in H. californica, the villi being less numerous and more of the nature of the diverticles in Benhamia. At the posterior end of the diverticles there is a large valve opening into the saoculated intestine."

It appears from any information which can be gleaned from the literature that the structure of the intestinal diverticula in H. urbanensis is different from anything yet described.

## Dorsal vessel.

The dorsal vessel arises from the perivisceral blood sinus in IX. At its origin, and extending somewhat into VIII, is a very large heartlike expansion which in the specimens studied is filled with a substance probably the remains of the blood. In the posterior part of VIII this swelling decreases rapidly and the dorsal vessel proper appears, lying near the mid dorsal line of the alimentary canal. Immediately anterior to the third pair of septal glands there is another expansion of the vessel, although not nearly so pronounced as the one at the origin of the vessel. This also soon becomes reduced to the usual diameter. The greater part of the external surface of the vessel is covered by

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chloragod cells.

The marked change in the diameter of the vessel in VIII is accompanied by marked changes in the walls of the vessel. In the anterior part of VIII the vessel is thick walled, the external surface is covered with chloragog cells, the inner surface has a number of cells projecting our radially into the lumen, and is of the usual diameter. Near the middle of eighth somite there occurs a sudden change in which the diameter increases greatly, the wall decreases in thickness to a mere membrange, the chloragog cells are lost, and the only cells which can be identified in connection with the walls are the few cells which lie flattened against or are contained within the extremely thin wall.

## Nephridia.

The anteseptal part is small; the postseptal part is large, broad, and somewhat pointed at the posterior extremity. The efferent duct arises from the postseptal part near the septum and usually is about as long as the former. The internal lumen is tortuous throughout its entire length. The right nephridium on XIII/XIV is missing in one of the preparations. This is probably due to the fact that that side of the body is filled with developing egg masses thus producing, a crowded condition which brought about the elimination of the nephridium.

# Spermiducal funnel.

The spermiducal funnel lies in the ventral part of XI with its base in close proximity to XI/XII. It  $l_A^{\prime}$ es close to the body wall and parallel to the long axis of the body. It is about two and one third times longer than the diameter, the measurements being - length **9.192** mm.,

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diameter 0.085 mm. The sporm duct is long, much contorted, and confined to XII.

# Penial bulb.

This organ(Fig. 58) is of the Lumbricillid type of structure. It lies in the usual position in the ventral part of XII on a deep invagination of the body wall. It is large in comparison to the size of the body and is conspicuous in sections. The body of the bulb is composed of three kinds of cells. The first kind (ibc) forms a series in which the cells are arranged in a radial fashion around the sperm duct extension for its entire length. They are uniform in character and have but a slight staining reaction. The nuclei lie at the bases of the cells and are so regular in their distribution that they appear as a distinct row in sections. The cells of the second kind(glp') occupy the dorsal peripheral part of the bulb. They are in general fusiform and arranged in such a way that the oval nuclei appear somewhat scattered. The cells are so placed that their long axes point towards the sperm duct extension. They stain intensely and have the general appearance of gland cells. The third series of cells(glp) occupies the ventral ectal part of the bulb. lying between the inner bulb cells(ibc) and the ventral periphery. They are much larger than those of the other two kinds and their boundaries are less strongly marked. Their contents as indicated by the staining reaction are quite distinct from the other cells being less dense and taking the stain sparingly. The point of entrance of the sperm duct(sd) occurs at the ectal side of the bulb not far from the innermost extent of the penial invagination. It penetrates the bulb and meets the sparm duct extension(sde) at a point about half way between

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the dorsal periphery and the penial pore(pp). The sperm duct extension curves laterad and meets the penial invagination(pb) in the upper half of its length. The cuticula which lines the invagination is also continued into the sperm duct extension as a lining. The bulb is covered externally by a musculature (m) which is a continuation of the circular muscle layer of the body wall. From the inner extremity of the penial invagination a muscular strand(mr) extends diagonally dorsad and unites with the muscle layer of the body wall a short distance away.

It will be noticed that the transition from the clitellar cells to the bulb cells is very abrupt and that this transitional region occurs very close to the penial invagination both on the ventral and on the lateral sides.

# Ovaries.

The ovaries are massive, extending dorsad around the digestive tract. Egg masses are present in the coelom and the type specimen seems to be at the height of sexual maturity.

# Spermathecae

A pair of these organs lie in V. The external opening of each is laterad and in intersegmental groove IV/V where it is surrounded by a rosette of glands(Fig. 56). The diameter of each organ is greatest near the eotal opening and from thence entad the diameter is reduced and becomes nearly uniform for the greater part of its entire length. There is no well developed ampulla and spermatozog are present along the whole length of the lumen. The spermatheca extends obliquely across the coelom to the digestive truct where it bends caudad. In the anterior

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part of V each spermatheca meets with the spermatheca from the opposite side to form a single common lumen(Fig. 57) through which both communicate with the digestive tract. This communication is not middorsad as is usually the case but is latero-dorsad in position.

#### 4. Genus LUMBRICILLUS Oersted.

The name Lumbricillus was first used by Oersted in 1844 but was not given a permanent place in the no@Aclature until 1900. Previous to that time the old name Pachydrilus of Claparede was in common use but Michaelsen finding that the name Lumbricillus antedated Pachydrilus replaced the latter by Lumbricillus which is now commonly accepted by most workers in Oligochaete.

The genus is defined by Michaelsen('00, p. 78) as follows:-

"Kopfporus klein, zeischen Kopflappen und 1. Segn. Borsten S-förmig gebogen. Rückenporen fehlen. Blut gelb bis rot. Das Rückengefäss entspringt postclitelliel and besitzt keinen Herzkörper. Peptonephridien fehlen. Hoden aus einer Anzahl birnförmiger Teilstäcke bestehend. Samenleit er lang. Samentaschen ohne Divertikel."

Eisen extends the above description by adding points concerning the testes, nephridia, and penial bulb. His definition of the genus is as follows:-

"Setae sigmoid, arranged in fan shaped fascicles. Head pore small, situated between the prostomium and the peristomium. Brain generally deeply emarginated posteriorly. Ventral sex<sup>ual</sup> glands around the ventral ganglion menerally present. Blood red or yellow. Dorsal vessel rises

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posterior to the clitellum. No cardiac gland. No peptonephridia. Testes multi-lobed, each lobe capped by a small sperm sac. Sperm ducts comparatively narrow. Penial bulb without inner muscular strands, containing numerous glands of various kinds, some of which may open into the basal part of the sperm duct. No atrium and no glands outside of the penial bulb. Nephridia with entire postseptal and with an anteseptal which consists merely of the nephrostome."

It will be noted that the chief characters of distinction are the absence of dorsal pores, peptonephridia, and cardiac gland, and the presence of sigmoid setue and pluri-lobed testes.

That the presence of the ventral glnds (Kopulationsdrüsen of Michaelsen and Ude, " opulatory Glands" and "Outgrowths of the Nerve Cords" of Beddard, "Ventral Glands" of Eisen) is not uiagnostic of the senus is made apparent by the fact that Southern('09,p.149 and 158) found them in Marionina semifusca Clap. and Enchytraeus lobatus Southern. Stephenson('11,p.52,58,62) found them in Enchytraeus nodosus Stephenson and evidences of them in E. sabulosus Southern and in Fridericis tulbosa Rosa. The presence of these glands is a feature of the genus but evidently not a distinctive character.

It has been recently shown that there is a close relationship between Lumbricillus and Enchytraeus. Stephenson('11) has found intermediate species which serve to bridge over the interval between the two genera. He calls attention to the usual characters serving to distinguish them, namely -

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| Lumbricullus                  | Enchytraeus              |
|-------------------------------|--------------------------|
| (1) Sigmoid setae             | Straight setae           |
| (2) Penial bulb present       | Penial bulb absent       |
| (3) Copulatory glands present | Copulatory glands absent |
| (4) Multilobed testes         | Undivided testes         |

He describes a new species from the Clyde, Lumbricillus viridis, which is typically Lumbricillid in all respects with the exception that the anterior part of the body has setze of the form typical of the genus Enchytraeus while in the posterior part the setae show only a very faint double curve. He also describes a new species from the same general locality, Enchytraeus nodosus, which is safely placed in that genus but which shows the following Lumbricillid affinities :- the presence of ventral glands, a penial bulb, and in certain cases the setse show an indication of a double curve. Again, Enchytraeus dubius, a new species described from the same general locality has setae typical of the genus Enchytraeus but has ventral glands, a penial bulb("although it is bifid internally"), the testes are loted and resemble the lobed condition in Lumbricillus. It also has red blood. It may also be added that Enchytraeus albidus, a very typical species of that genus, has an imperfect penial bulb surrounded, however, by other and smaller aggregations of gland cells. All of this data is interesting in view of the fact that heretofore these two genera have been regarded as standing some distance apart and have been placed in different subfamilies.  $\mathbb{T}^{n}$  is point will be discussed at length in another connection.

Michaelsen('00) placed seventeen species in this genus, one of which he regards as somewhat doubtful. Later('03) he added L.henkingi

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Ude to the list making a total of eighteen species. Of this assemblage of species none had been reported from North America. Recent investigations have increased the number so that at present thirty one species, three varieties, one doubtful species, and one doubtful variety are referred to this genus. Of this number six species and three varieties have been described from North America. They are as follows:-

Type Locality. Name. San Mateo Co., Calif. L. santaeclarae Eisen Metlakatla, Alaska. L. merriari Eisen Metlakatla, Alaska. L. merriari var.elongatus Eisen Metlakatla, Alaska, and L. annulatus Eisen Orca, Prince William Sound. Farragut Bay, Alaska. L. ritteri Eisen Santa Clara River, Calif. L. franciscanus Eisen St. Paul Island, Pribilof L. franciscanus var. borealis Eisen Group, Alaska. Unalaska. L. franciscanus var. unalaska Eisen Casco Bay, Me. to Vineyard L. agilis Moore Sound, Mass.

KEY TO THE SPECIES OF THE GENUS LUMBRICILLUS KNOWN TO OCCUR IN NORTH AMERICA.

A. Spermatheca with crown of glands limited to the ectal opening.

B. Spermathecal duct distinctly set off from the ampulla; ventral glands in XIV-XV; brain two and one half times longer than wide, posterior margin deeply emarginate - -- - L. santaeclarae Eisen.

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Lumbricillue rutilus n. sp. Plate II, figs. 13-22.

Definition.

Length 15-19 mm. Diameter 0.44-0.68 mm. Somites 41-49. Color red with slight tinge of yellow. Prostomium Bather short and rounded. Head pore on B/I. Dorsal pores absent. Setae sigmoid; all of same size and approximately of same length; in anterior 2/8 to 3/4 of body 0-7 in lateral bundles and 5-10(usually 5-7) in ventral bundles; 4-2 in posterior part of body. Clitellum on XII-XIII; interrupted on mid ventral surface. Brain about one and one half times longer than wide: anterior margin concave, posterior margin distinctly emarginate, lateral margins diverge slightly cauded. Peptonephridia lacking. Dorsal vessel varies slightly in position of its origin, arising in XIII-XIV. Nephridia with large postseptal region and very small anteseptal part which consists only of nephrostome; efferent duct arises from ventral surface of posterior part of postseptal part. Spermiducal funnel cylindrical, 4-5 times longer than diameter, strongly bent at middle; collar present, slightly reflected, slightly wavy in outline, and set off from body of funnel by a slight constriction. Testes multilobed, with about nine lobes on each side of body; each lobe capped by a small sperm sac. Spermathecae in V; without diverticula and each consisting of a well developed ampulla and a short duct; ampulla consisting of an expanded, barrel-shaped, thick walled middle region and a much narrower ental region which is reflected cephalad before uniting with digestive tract; duct not sharply set off from ampulla, is much shorter -than ampulla and surrounded by a well developed gland which shows a

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number of lobes arranged in the form of a rosette; ectal opening laterad and near IV/V, internal opening on lateral wall of digestive tract in posterior part of V. Ventral glands in XIII and AIV, differ slightly in shape in the two somites, surround ventral ganglis closely on ventral, lateral, and part of dorsal surfaces leaving only median dorsal line free.

Described from 32 sexually mature specimens. Many other specimens were examined in determining the external characters.

Cotypes in the collection of the writer. Several cotypes also in the collection of Professor Frank Smith.

The specimens which are the basis of this description were collected June 22, 1911 by A.A.Girault. They were found in sprinkling filter no. 5 of the Chicago Sewage Testing Station. They occurred in great abundance in sludge which covered the lime stone rocks which compose the filter bed. A complete description of the habitat of these worms will be given in another part of the paper.

# Affinities.

This form is easily separated from the other known American species and the differences are so distinct and so numerous that it can scarcely be said to have any close relatives among the American forms. When compared with the foreign species of this genus it appears to resemble L. litoreus Hesse, L. subterraneous Vejd., L. lineatus Mäll., L. verrucosus Clap., and L. tenuis Ude. However there are a number of distinct differences in each case and furthermore bhe descriptions of

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the above named species are very brief and make no reference to some characters which are now considered to be of use in separating species. When these descriptions are made more complete it is reasonable to expect that other points of difference not yet known will be found.

# DETAILED MORPHOLOGY.

#### EXTERNAL CHARACTERS.

The body is smooth, slender, and cylindrical and tapers gradually towards the two extremities. The length in alcoholic specimens varied from 9-14 mm., the more common length being 10-12 mm. However studies made on the living specimens showed that these figures are not an accurate estimate of the length of mature specimens. This form has certain limits of elongation and contraction and a series of measurements taken on the living forms showed that the lengths indicated in the alcoholic specimens were too low, probably due to the effect of the killing and fixing fluids. In the living specimensthe length varies from 15-19 mm. These measurements were made on specimens which were not in an extremely elongated or contracted condition. The diameter is greatest in the region of the clitellum where, in alcoholic specimens, it is 0.446-0.684 mm. The anterior 5-9 intersegmental grooves are very distinct but the remaining ones are rather obscure. The number of somites is variable but the limits of this variation are rather definitely fixed. Counts in thirty five specimens showed this variation to be 40-49. The moderately developed clitellum is on XII-XIII and occurs only on the dorsal and lateral surfaces of the worm. In alcoholic specimens the body is opaque and light brown in color except on the clitellum where it is much lighter. In the living animal the color is reddish

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with a slight tinge of yellow. The prostomium is blunt, smooth and rounded. The head pores is small and located at O/I.

The setae(Fig. 18) are distinctly sigmoid and are arranged in fanshaped bundles. These bundles are disposed in four longitudinal rows, two ventral and two lateral. In the ventral rows the number of setae per bundle varies from 4-8, occasionally 9 and in rare instances 10. In the lateral rows the number varies from 4-7. In the last few comites the number in both sets of rows varies from 4-2.

#### INTERNAL CHARACTERS.

# Brain.

The brain(Fig. 20) lies in I and II. The length is about one and one half times the width. The posterior margin is distinctly emarginate while the anterior margin is concave. The lateral margins vary to some extent, being in some cases nearly parallel, in others slightly divergent caudad. Two pairs of supporting strands extend from the posterior end of the brain to the body wall while from the anterior end a rather strong muscle strand extends from the mid ventral region to the wall of the prostomium.

## Blood vascular system.

Since the blood in this species is colored and the vessels are rather large and remain distended after death it is possible to follow out the course of the chief vessels. Furthermore owing to the fact that the integument of the body is semi-transparent and the blood vessels stand out prominently it was possible to study the vascular system in the living form and thus verify the observations made on the alcoholic

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material. The system(Fig. 18) consists of two principal longitudinal vessels, one dorsal and one ventral, and transverse vessels which connect them in the anterior region. The dorsal vessel arises from the perivisceral sinus in WIII-XIV. An examination of twenty specimens showed that the origin of this vessel is not constant in position and that it varies within the limits of XIII-XIV. In the majority of cases the origin is at XIII/XIV. At the intersegmental regions this vessel shows constrictions which are slight in its anterior part but distinct in its posterior half. Distinct swellings are present in XI, XII and XIII. From its origin the dorsal vessel extends cephalad, parallel and dorsad to the digestive tract and throughout its course it maintains a rather close relation to it. In I this vessel divides into two symmetrical branches, one of which passes around to the right and the other around the left side of the digestive tract. These branches extend to the ventral side of the buccal cavity and each comes to lie parallel to it thus forming the right and the left ventral trunks. They extend caudad into IV where they approach each other and unite forming a single vessel which extends to the posterior region of the body. In III there arises from each of the yentrals vessels a branch which extends dorsad to connect with the dorsal vessel at a corresponding position, thus forming the first pair of transverse vessels. The second pair of transverse vessels occurs in IV anterior to the point of union of the right and left ventral trunks. The third pair arises from the ventral vessel very near IV/V and immediately posterior to the point of union of the right and the left ventral vessels and it unites with the dorsal vesselat a corresponding point of union of the second pair of transverse vessels.

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The fourth pair connects the dorsal and the ventral trunks in V.

# Nephridia.

The nephridia(Fig. 15) are of the usual Lumbricillid type in which the postseptalpart is represented only by the nephrostome. The efferent duct arises from the ventral surface of the posterior end of the postseptal part and opens to the exterior in front of the ventral setae.

# Testes.

The testes are located in the usual position in XI and are multilobed. Each lote is club shaped and its attachment to the ventral surface of X/XI is very slender. In this species there are approximately 8-10 of these lobes on each side of the digestive tract, all somewhat similar in sixe and shape and radiating fan-wise from the point of common attachment, the anterior and posterior ones extending out into the adjacent somites. Each lobe is capped by its own individual sperm eac. The presence of these sperm sacs on the testes of Lumbricillus was first reported by Eisen in 1900 and he also found that this condition is not confined to Lumbricillus since an exceptional case of the same occours in Ocnerodrilus occidentalis, the only species of that genus which shows this peculiarity. The testes have been studied by other workers in a number of species of Lumbricillus and while they have been found to differ from each other to a limited extent, it has not been practical to use these differences as specific characters.

# Spermiducal funnel.

This organ(Figs. 14 and 16a) lies in the usual position in XI. The

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length is about 4-5 times the diameter. In all of the specimens examined the funnels are cylindrical and are strongly bent near the middle, usually reflected upon themselves. The collar at the anterior end of the funnel is usually somewhat wavy in outline and varies in shape being sometimes slightly relfected and sometimes flaring. The duct extends through XI/XX and after presenting a few contortions extends directly to the penial hulb.

Southern('09, p. 149) claims that the sperm funnel in the genus Lumbricillus is "very contractile" and "varies greatly" in its relative proportions according to the amount of tension on it. He holds that "specific determinations which rely on these two characters(the funnel and the ventral glands) must be regarded with suspicion, especially when preserved material is used". No evidence of such variation has been found in L. rutilus, at least not enough to warrant a statement as strong as the above. It is quite doubtful if the spermiducal funnel in L. rutilus is "very contractile" since the structure is such as to preclude conspicuous contractility. The muscular tissue is reduced to almost a minimum and since the bulk of the organ is composed of long closely set cells and the lumen is very fine it does not appear to be reasonable to expect great contractility in the funnel of this species.

# Penial bulb.

This organ(Fig. 22) is rather simple in its structure and conforms to the Lumbricillid type. It is rather large as compared with the diameter of the body, is approximately globular in shape, and is covered with a musculature (m) which is a reflection of the musculature of the body wall. Both the circular and the longitudinal layers take part in

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this muscular covering. The circular muscles lie in contact with the gland cells of the bulb and are very much reduced in thickness, so much so that it is difficult to demonstrate them with high powers. However close examination shows that there is a very thin layer present and in transverse sections it is easy to distinguish the strands of muscle fibers passing from the circular layer of the body wall to the periphery of the bulb at two different regions, namely, between the entrance of the sperm duct and the body wall, and the periphery of the bulb and and the ventral body wall. The longitudinal layer is well developed and in transverse sections it shows the same structure as that of the body wall the only difference being a reduction in thickness. The interior of the bulb is filled with cells of but one kind. These cells differ somewhat in shape in the different parts of the bulb but in general they are elongate, somewhat fusiform, and most of them are arranged radially around that sperm duct extension (sde). The nuclei are located for the most part in the peripheral ends of the cells. The peripheral part of each cell stains deeply while the opposite part which is approximated to the sperm duct extension stains lightly. At the ventral side of the bulb the cells merge gradually into the hypodermis (h) of the body wall. The sperm duct (sd) meets the dorsal part of the bulb near the lateral body wall. It unites with the bulb in such a way that the terminus of the duct appears to be imbedded in the periphery of the bulb. The union of the sperm duct with the sperm duct extension (sde) is not accompanied by any kind of differentiation and it is a little difficult to determine the exact junction of the two. The sperm duct extension (sde) is quite spacious and is differentiated into three distinct parts, an inner, a middle, and an outer region. The inner region constitutes about one

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half the length of the sporm duct extension and extends from the junction of the latter with the sperm duct to the middle region. It is approximately uniform in diameter and strongly sigmoid in shape. The middle region is a distinct bulbous swelling in the lumen of the sperm duct extension in which the diareters are about equal. The outer region is that part which lies between the middle region and the penial invagination (pb). It is funnel shaped, the greater diameter being at the outer end. This last named region might be regarded as a part of the penial invagination but the examination of a number of specimens shows that it is constant in shape in the retracted bulbs and since a slight angularity, present at the junction of the invagination and this region, seems to mark the boundary between the two it will be treated as a part of the sperm duct extension. Since the union of the sperm duct with the bulb is slightly posterior the sperm duct extension follows a cephaloventro- lateral course in reaching the exterior. The cuticula (ou) of the body wall is reflected inwards into the sperm duct extension and lines it for its entire length.

#### Spermatheca.

A pair of these organs(Fig. 16) is present in the usual position in V and as in all of the previously described species they have no diverticula. There is no well defined line of separation between the duot and the ampulla. The ectal opening has a well developed crown gland which shows a series of emarginations on the ental margin giving it the appearance of having about six lobes. When the spermatheca, is dissected out and mounted in toto it is observed that the gland is large enough to completely hide the spermathecal duct since the ental

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periphery of the gland extends to the ampulla. The duct gradually merges into the ampulla, a fusiform structure, constituting the greater part of the mass of the spermathece. The ental portion of the ampulla gradually becomes reduced in diameter forming a short dust-like terminus which has approximately the same diameter and length as the spermatheceal duct. The ental part of the ampulla bends cephalad upon itself and unites with the digestive tract. This union is lateral in position and is exactly opposite the corresponding opening of the other spermathecea. An examination of sexually immature specimens shows that the spermathece lacks the connection with the digestive tract, a fact which meens to point to the conclusion that the ental part of the ampulla is the last to be acquired by the developing spermathece. The ectal opening of each spermathecea is surrounded by a thickened region of the hypodermis.

The abundance of "terial and its perfect histological condition has made it possible to study in some detail the histology of the spermathees. A number of interesting structural features are present and seem worthy of extended discussion. The meager treatment of the structure of spermatheeae in species of Lumbricillus makes comparisons almost impossible and gives no hint as to whether the condition in this species is unique or common to the group.

The wall of the ampulla(Fig. 28) is composed of three layers, the enveloping peritoneum(p), the muscle layer(mcl), and the lining epithelium (el). The peritoneum is of the usual type being merely a thin membrane with scattered nuclei. The muscle layer is well developed and consists of a single layer in which the component fibers extend around the organ in a transverse direction thus forming a circular layer. It is about equally developed in all parts of the ampulla and in the

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region of the junction with the intestine passes over into the muscle layers of the digestive tract. However it is very difficult to determine the exact structure of this transitional region and the writer has not been able to demonstrate with absolute certainty to which of the muscle layers of the digestive tract it becomes allied. It seems to pass into the circular layer as one would expect, and further evidence in support of this conclusion will be given later when it will te shown that this muscle layer connects with the circular layer of the body wall. The epithelial layer of the ampulla constitutes the greater part of the mass of the organ. It is composed of tall columnar cells set closely together and distinctly nucleated at the bases. In all parts of the organ this layer is thick and especially so in the enlarged part where it forms a very thick heavy wall and the length of the component cells is many times longer than their diameters. This layer is one cell thick throughout. In longitudinal sections of the organ this fact is clear enough in the ental region where the ampulla connects with the digestive tract but in the swollen region where the layer has its maximum thickness it appears at first sight to be composed of more than, layer since several rows of nuclei appear in the same section. These cells: are closely set and sections of the usual thickness include parts of from two to three superimposed layers thus giving the appearance referred to above.

The part of the spermathece extending from the ampulla to the external opening is really the duct but is not strikingly set off from the ampulla. It has a rather peculiar structure. The epithelial layer (el) which lines the ampulla is interrupted completely and the wall of

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the duct is composed of two kinds of elements only, the muscle layer (ms), and the gland cells(eog) of the large gland which surrounds that region. This gland is composed exclusively of very long cells which extend from the periphery to the lumen of the duct. The nuclei are situated in the peripheral ends. In a longitudinal section of the spermatheces these cells have the appearance of having their inner ends out off by a band of muscle tissue(ms) thus separating the glandular region into two parts, one lying between the lumen and the muscle band and the other part lying on the other side of the muscle band. This appearance is further emphasized by the fact that the tissue outside of the muscle band stains readily while that between the muscle band and they lumen does not take the stain at all. In reality these apparent regions are not distinct but the gland cells are continuous from the periphery of the gland to the lumen and are not interrupted by the muscle band as will be shown in the following paragraph.

The muscle elements in the epermathecal duct exhibit an interesting peculiarity in arrangement and derivation. At the ectal end of the ampulls the well developed circular muscle layer instead of continuing over the duct unchanged becomes converted into a series of muscle bands(ms) which extend at right angles to the direction taken by the fibers of the same layer on the ampulla. These bands do not form a continuous layer but occur at intervals thus forming a cylinder of muscle bands each of which is separated from the neighboring ones by spaces through which extend the prolongations of the gland cells to meet the lumen. These muscle bands continue through to the external hypodermis and become allied to the circular muscle layer of the body

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wall. Each band appears to be composed of two Similar parts closely approximated and in all cases this double condition of the bands seems to be constant.

The peritoneum (p) continues over the gland as an envelope. The external cuticula (cu) is reflected into the lumen of the spermatheca but only for a short distance.

## Ventral glands.

The peculiar and problematic organs which have been given several names( "Kopulationdräsen" of Michaelsen and Ude, "Copulatory Glands" and "Outgrowths of the Pentral nerve cord" of Beddard, "Ventral Glands" of Eisen) are present in specimens of this speciee. They are moderately developed (Fig. 17) and are closely and uniformly associated with the ventral nerve cord in XIII and XIV. They almost completely surround the ventral ganglia leaving only a small free space on the dorsal median line of the nerve trunk. In transverse section(Fig. 19 and 21) they appear to be made up of a mass of distinctly nucleated spindle-shaped gland cells which lie parallel to each other and extend ventrad. They do not have the lateral wing-like developments which are present in some species but are compact and bulbous in appearance. Each penetrates the body wall in the mid ventral region immediately under the nerve cord, thus opening to the exterior. In the majority of the specimens examined these glands are not bilaterally symmetrical but one side is more strikingly developed than the other giving the whole the appearance of being turned over to one side. In all of the specimens examined the increased growth occurred on the same side for both glands of the same specimen but in some individuals it was found on the left

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side while in others it is on the right side. The two glands differ but little in size but there is a difference in the shape. The ental ends of the one in XIII extend above the level of the ventral ganglion forming a mid dorsal fossa While the ental ends of the gland in XIV do not extend above the level of the ventral ganglion and a fossa is not formed.

It appears that in the past undue stress may have been placed on the importance of the ventral glands as a specific character in the genus Lumbricillus and also in Marionina. Southern('09,p.149) has found great variation in them. "In some cases individuals have shown well developed glands while in others from the same locality they were either small, absent, or in different segments". Ditlevsen('04,p.432) also throws doubt on the value of this character in the separation of species.

No evidence of variation has been observed in the ventral glands of Lumbricillus rutilus. In every specimen of the long series examined they occurred uniformly in XIII and XIV and in all cases showed practically the same degree of development. The only variable feature is the one referred to above, namely, the occurrence of the increased growth on one side of the glands on the right side in some specimens and on the left eide in others.

# 5. Genus FRIDERICIA Mchlsn.

Fridericia is a well defined genus separated from the other genera by the following important characters:- (1) the presence of dorsal pores; and (2) the development and arrangement of the setae. They are

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produced in each bundle two at a time, the outer pair consitituting the oldest and largest ones. The next pair lies immediately within the oldest and these in turn include still younger ones in the same manner. The group thus formed is fan-shaped with a pair of outer large setae and gradating pairs between. Sometimes an old seta may fall out thus destroying the symmetry and frequently but one or two fully formed setae may be present in a bundle.

Michaelsen('00, p. 94) defines the genus as follows:-

Borsten in 4 Bändeln, gerade, zu 2 im Bändel und dann gleich lang oder zu mehreren und dann die inneren des Bändels mehr oder weniger regelmässig paarweise ind stufenweise kleiner als die äussern. Räckenporen mit Verschlusszellen meist vom 7., selten vom 6 Se.m. an vorhanden: Kopfporus meist klein, dorsal zwischen Ropflappen und 1. Segm. Lymphkörper von zweierlie Gestalt. Peptonephridien stets vorhanden. Der Oescphague gehts allmählich in den Mitteldarm über. Das Räckengefäss entsprings meist postclitellial: Elut farblos. Nephridien meist mit grossem Anteseptale, in dem der Flimmer kanal echon Windung beschreibt. Samenleiter lang. Samentaschen meist mit dem Darm kommunizierend, enfach oder mit Divertikeln".

Fridericia is the largest genus of the family Enchytraeidae. In 1900 Michaelsen allowed twenty one species and since that date the number has increased considerably so that at present there are about ninety species referred to this genus. It appears that at least three of the species included in this number are doubtful. Of this large assemblage of species sixteen have been described from North America although Michaelsen claims that one of them, F. parva Moore, is a

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synonym of F. bulbosa Rosa. he North American species are:-

| Name                           | Type Locality                        |
|--------------------------------|--------------------------------------|
| F. alba Moore                  | Philadelphia, Pa.                    |
| F. agricola Moore              | Philadelphia, Pa.                    |
| F. longa Noore                 | Philadelphia, Pa.                    |
| F. parva Moore(bulbosa Rosa ?) | Philadelphia, Pa.                    |
| F. agilis Smith                | Havana, Ill.                         |
| F. firma Welch                 | Urbana, Ill.                         |
| F. tenera Welch                | Urbana, Ill.                         |
| F. harrimani Eisen             | Mountain View, Calif.                |
| F. fuchsi Eisen                | Santa Cruz Mts., near Boulder Creek. |
| F. johnsoni Eisen              | Santa Barbara, Calif.                |
| F. santaerosae Eisen           | Santa Rosa, Sonoma Co. Calif.        |
| F. santaebarbarae Eisen        | Santa Barbara, Calif.                |
| F. maogregori Eisen            | Napa Co. Calif.                      |
| F. californica Eisen           | San Francisco, Calif.                |
| F. sonorae Eisen               | Sonora, Mexico.                      |
| F. popofiana Eisen             | Popof Island, Alaska.                |

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KEY TO THE SPECIES OF THE GENUS FRIDERICIA KNOWN TO OCCUR IN NORTH AMERICA.

A. Spermathecae without diverticula.

B. Posterior margin of brain convex.

C. Spermathecae do not connect with the digestive tract - - - F. sonorae Eisen.

CC. Spermathecae connect with the digestive tract.

D. Peptonephridia simple and unbranched; brain about twice as long as wide - - - - - F. sime n. sp.

DD. Peptonephridia with four to six branches projecting from a common base; brain almost circular - F. fuchsi Eisen.

BB. Posterior margin of brain truncate or concave.

C. Peptonephridia simple and unbranched - - F. parva Moore. (bulbosa Rosa)

CC. Peptonephridia with distinct branches.

D. Length over 10 mm.; somites over 50; dorsal pores begin on VI; dorsal vessel arises in XXII - - -

DD. Length under 10 mm.; somites not more than 50; dorsal pores begin on VII.

E. Dorsal vessel arises in XIV; peptonephridia short, each with at least two branches; brain deltiod, posterior margin almost straight, anterior margin conical - - - - F. harrimani Eisen.

EE. Dorsal vessel arises in XIII; peptonephridia thick and compact, with the free end frayed; brain with posterior margin slightly concave, anterior margin convex - - - F. johnsoni Eisen.

AA. Spermathecae with diverticula.

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B. Spermathecae each with two diverticula.

C. Brain circular - - - - - - - - - - - F. santaerosae<sup>1</sup>Eisen.

1. Spermathecae as a rule with only two large diverticula as the key indicates but in one specimen Eisen found that the large diverticulum on one side was replaced by three smaller ones.

|  |                      |                |                |     |                   |               |           |                |           |                |                |              | - 0           | 204      | -              |               |               |               |              |                 |                |          |          |                |                |                |          |               |             |                |                   |              |                     |          |
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|  |                      |                | D.             | ŝ   | Sp                | er<br>le      | n         | at             | h         | ecat           | a]             | l<br>th      | di            | V (      | er             | ti<br>al      | ou<br>C       | ila<br>ope    | e<br>en      | su<br>in        | b-g            | 0        | y1       | ir<br>tł       | nd<br>ne       | ri             | olu      | al<br>ot      |             | n              | 0                 |              |                     |          |
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|  | DD.                  | Se<br>du       | tae            | ea  | pr<br>ri          | 8             | do<br>es  |                | in<br>fr  | ar             | at<br>n        | ly<br>ar     | , i<br>it     | fo<br>Br | ur<br>io       | r             | e             | n d           | or           | of              | Dit            | er<br>he | ł        | ou<br>oo       | nd<br>st       | 11             | e;       | t             | al          | gi<br>I        | hr<br>pa          | ic<br>r1     | 11 e                | 91       |
|  |                      | E.             | Aı             | nt  | er                | i             | or        | 1              | ra        | r              | 21             | n            | 01            |          | br             | aj            | n             | C             | or           | 108             | av             | e.       |          |                |                |                |          |               |             |                |                   |              |                     |          |
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EE. Anterior margin of the brain convex.

- F. Branches of the peptonephridia arise from a common base; length 8 mm. F. macgregori Eisen.
- FF. Franches of the peptonephridia aries dendritically and not from a common base. length over 8 mm.
  - G. Dorsal vessel arises in XX -F. oconeensis n. sp. GG. Dorsal vessel arises in XVI - -- - F. californica Eisen.

Fridericia douglasensis n. sp. Plate III, figs. 24-33.

Definition.

Length 11-20 mm. Diameter 0.48-0.54 mm. Somites 43-55. Color white. Prostomium short, blunt, slightly angular at tip. Head pore on O/I. Dorsal pores begin on VII. Setae 2-6 per bundle in ventral rows and 3-5 in lateral rows in anterior part of body: 3-6 in the middle region, and 3-2 in posterior region. Clitellum on 1/3 XI = 1/2 XIII. Lymphocytes elliptical. Erain about one and two thirds times longer than wide: anterior margin conceve, posterior margin truncate, lateral margins diverge caudad. Peptonephridis large, tubular, slightly tuberculate, unbranched. Dorsal vessel arises in XX. Nephridis with anteseptal and postseptal parts about same size: efferent duct arises from middle of latter. Spermiducal funnel well developed; length about one and one half times diameter; collar distinctly set off from body of funnel by constriction; margin of collar not reflected. Sperm duct shorter than in most species and with few contortions. Spermatheces with • -

duct, ampulla and diverticula; ampulla funnel-shaped, with 7-11 globular, unequal, irregularly disposed diverticula; no glands at ectal opening of duct; each ampulla with independent connection with digestive tract.

Described from 16 sexually mature specimens. Many more were examined in estimating the external characters. Cotypes in the collection of the writer. Cotypes also in the collection of Professor Frank Smith.

The specimens on which this description is based were collected in a deciduous forest near the north shore of Douglas Lake, Michigan, July 14, 1911. They were found in considerable abundance under chips, pieces of bark and other debris at the base of a large fallen tree where there was considerable moisture present. The majority of the specimens were sexually mature.

#### Affinities.

This species belongs to the group having more than two diverticula on the spermathece. It appears to have no close relatives among the foreign species. It is also distinct from the other American species and apparently approaches none of them unless it be F. longs Moore and unfortunately the description of that species is inadequate thus making the establishment of relationships uncertain.

# DETAILED\_MORPHOLOGY.

## EXTERNAL CHARACTERS.

The body is slender and has a length of 11-20 mm. In transverse

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section the outline of the body is circular and is greatest in the region of the olitellum where the diameter is 0.45-0.54 mm. The living specimens are opaque and whitish in color. The intersegmental grooves are obscure for almost the entire length of the body; only the first two or three are at all distinct. In nearly all of the specimens examined the first intersegmental groove is deeply marked. The number of somites is variable, the extremes in a count of nine specimens being 43 and 55. The prostomium is short and somewhat angular at the extremity. The degree of this angularity is somewhat variable although all of the specimens show it to some extent. The clitellum is moderately developed and is situated on 1/S XI-1/2 XIII.

The number of setse pair bundle varies in the different regions of the body and to a limited extent in the different rows. In general there are 2-6 per bundle in the ventral rows in the anterior region(more often 5 or 6), and 8-5 in the later rows: 2-6 per bundle in middle region in both sets of rows; and 3-2 in the posterior region. Aside from a decided bend at the proximal end(Fig. 29) each sets is straight.

### INTERNAL CHARACTERS.

### Brain.

This organ(Fig. 80) lies in I and II, chiefly in the latter. The anterior margin is concave while the posterior margin is distinctly truncated. The lateral margins diverge caudad for about two thirds of their length. Figure 20 shows the characteristic shape of this organ. In transverse sections it is broadly elliptical in outline. The anterior part gives rise to two nerve trunks which form the circumcesophageal

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commissures. The entire organ is surrounded by a well developed neurilemma which is somewhat thickened between the roots of the commissural trunks. Two sets of supporting strands attach the posterior region to the body wall.

#### Peptonephridia.

There is a pair of these organs ventrad to the digestive tract, one on the right and one on the left. Each opens separately into the oesophagus in a latero-ventral position in IV. There is some variation as to the exact place of opening, some specimens showing it mear the middle of IV while in others it is in the posterior part. However in all cases the opening is in IV. These organs(Fig. 32) are long and unbranched. They resemble the peptonephridia in Fridericia oconeensis in general appearance but are not quite so, long, are larger in transverse section, and the tuberculate appearance is not so pronounced. they are tubular , with but a single lumen which is large in comparison to the size of the organ. Izrediately beyond the union with the digestive tract each peptonephridium turns abruptly caudad and extends parallel to it. The length varies somewhat in the various specimens. In some cases they extend into VI but in otners only into V. Sometimes the terminal parts extend around the alimentary canal and end dorsad to it. One of the specimens examined showed a peculiar condition in which one of the organs, about midway of its length, gave rise to a single long well developed branch which extended cephalad and terminated anterior to the opening of the peptonephridium. The structure of this branch is identical with that of the main trunk. This is probably an accidental condition since it ocours only in one specimen and only on one side.

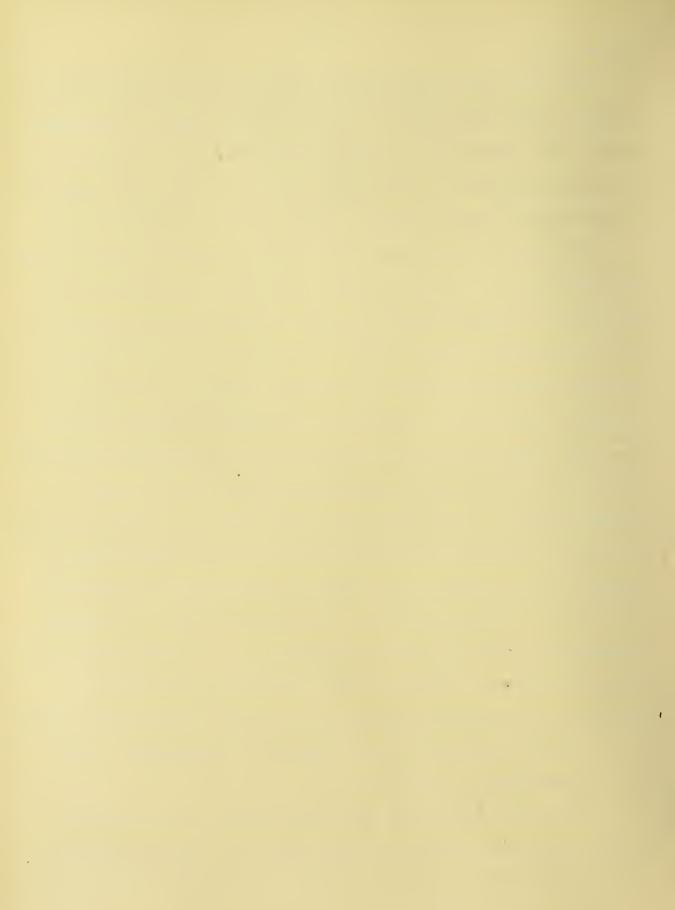
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Chylus cells.

The chylus cell region occupies XV-XVIII in all of the specimens examined except one in which a few chylus calls appear in the posterior part of XIV. Cells of two kinds are present(Fig. 32), the chylus cells (ch), and the epithelial cells(epc). The chylus cells form a continuous layer. They are flask-shaped, the broader ends being ectal. The base of each is more or less truncate and the sides converge gradually entad. The apical half of the intracellular canal(icc) is straight but the basal half is somewhat spiral in shape and appears sigmoid in section. This intracellular canal is lined by a gelatively thick specialized layer of cytoplasm(cyl) which is everywhere uniform in thickness and structure. The ganal is alliated for the greater part of its length. The blood sinus (bs) somes into contact with little more than the basal end of each chylus cell. The epithelial cells(epo) are long and trumpet-shaped, the broad end being ental. Their ectal ends are inserted between the spices of the chylus cells. In longitudinal sections usually but one epithelial cell appears between each chylus cell and its neighbor. The ental surface of the spithelial cells is ciliated. Interstitial cells are absent. There appears to be a distinct difference in the structure of the chylus cells and the epithelial cells as indicated by the staining reaction. In the former the cytoplasm appears to be somewhat granular while the cytoplasm of the epithelial cells is more homogeneous; and more dense.

The general character of these chylus cells is confirmatory of Eisen's conclusion, that the location, shape, and size of such cells offer specific differences. The characters of the chylus cells in this

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species are distinct from those of other species so far as they are known.

### Vascular system.

The blood vessels in the specimens of this species remain better distended than in the others examined making it possible to follow them and to study the chief vessels. The system(Fig. 24) consists of the usual dorsal longitudinal vessel(bvd), the ventral vessel (bvv). and transverse vessels which connect them forming loops around the anterior part of the digestive tract. The dorsal vessel ariges from the proivisceral blood sinus in XX. It shows distinct swellings in each of the somites posterior to the clitellum. From its origin it extends cephalad, parallel, and dorsad to the digestive tract with which it maintains a more or less close relation throughout its course. In the anterior it lies between the brain and the buccal cavity and is much reduced in diameter so that it requires high magnification to follow it. At a point immediately under the anterior part of the brain it divides into two trunks, one passing to the right and the other to the left of the digestive tract, both extending to the ventral side of the buccal cavity where they lie parallel to it. They extend caudad to the anterior part of IV where they approach each other and units near IV/V to form the single ventral trunk. Near the middle of III a transverse vessel extends dorsad from each ventral vessel and unites with the dorsal vessel near the posterior part of III. In the anterior part of IV similar transverse cessels connect the ventral vessels with the dorsal trunk. No transverse vessels occur in II.

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Nephridia.

The anteseptal part(Fig. 27) is of about the same size as the postseptal although there is a slight variation, the latter being sometimes a little larger than the former. The efferent duot arises from the mid ventral surface of the postseptal part about midway of its length and opens externally just anterior to the ventral setae bundles of the same somite. The lumen is very tortuous throughout its entire course.

## Spermiducal funnel.

This organ(Fig. 28) lies in the usual position in XI with its base close to the ventral surface of X/XI. The length is about one and one half times greater than the diameter. It is slightly bent and in the majority of the specimens examined it was bent in such a way than the anterior end was directed dorsad or laterad. The collar is distinctly set off from the body of the funnel by a constriction. Its diameter is about one half that of the funnel. The free margin is not refeloted but projects funnel-wise towards the developing sperm mass.

The sporm duct is confined to XII and seems to be much shorter than is usual in other species. Instead of the mass of convolutions in XII there is at most only three or four coils and from these the duct extends directly to the penial bulb.

# Penial bulb.

In structure the penial bulb(Fig. 21) conforms to the Lumbricillid type as defined by Elsen. It is well developed and is a conspicuous organ in transverse sections of the body through XII. It is completely

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invested by a strong ausculature which ultimately connects with the muscle layer of the body wall. When the bulb is completely retracted the penial invagination(pb) is large, having a depth equal to the length of the bulb. The sperm duct(sd) meets the bulb on the dorsal-ectal surface and penetrates it for a short distance. Within the bulb it is replaced by the sperm duct extension (sde) which, curving boldly laterad, extends to the penial invagination, opening into the latter about mid way of its length. The external outicula lines the penial invagination and also extends into the sperm duct extension as a lining. The bulb is composed of two groups of cells. One group occupies the dorsal part of the organ and is composed of cells which occur near the periphery but send long prolongations to the sperm duct extension. The second group of cells occupies the ventral part of the bulb and opens only onto the surface of the penial invagination. In nearly all of the preparations studied in this connection it was not difficult to determine the line of separation between these two groups of cells owing to the fact that they tend to separate and produce a distinct split in the bulb at that place.

All of the preparations examined showed a few nuclei scattered among the extensions of the doreal cells at a position about half way between the periphery and the sperm duct extension. This recalls the condition in F. firms, agilis, agricols and others in which there is another group of cells between the sperm duct extension and the peripheral doreal cells which is conspicuous in sections because of the arrangement of numerous similar nuclei in a definite row. Critical @Asmination of this bulb has failed to reveal a third definite group of cells in that region. High magnification shows that the scattering nuclei represent

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only occasional cells interpolated between the extensions of the other cells. It is possible that the structure of this bulb represents a transition between the type represented by F. firms and that represented by E. tenera.

#### Spermathecae.

A rather surprising variation exists in these organs in the different specimens. In order to be sure of sexual maturity each spermatheca was examined carefully for spermatozoa, their presence being taken as evidence that the specimen was sexually mature. The number of diverticula (Fig. 25) varies from about 7-11. They are of unequal size on the same spermatheca and their number may vary in the members of the same pair. They are more or less spherical in shape and the cavity of each has a wide communication with the lumen of the ampulla. In some cases the diverticula are quite distinct. Sometimes they are obscured so that at first sight the spermathecae appear to be devoid of them, but closer observations shows that they are uniformly present. The ampulla is pearshaped and connects with, the digestive tract. There is considerable variation in the position of this connection. In some specimens it is very near the mid dorsal line and the ental ends of the two ampullae lie so closely together that only very close examination of sections reveals the fact that they enter separately. In other specimens the two ampullae unite with the digestive tract laterally, one exactly opposite the other. Furthermore there is some variation in the way in which they approach the digestive tract. Both ampullae may approach symmetrically, or the ampulla on one side may curve dorsad and unite with the digestive tract near the mid dorsal line while the other extends

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directly to the mid lateral surface and unites with that point. The spermathecal duct is straight and devoid of glands.

Fridericia oconeensis n. sp. Plate IV, figs. 34-41.

Definition.

Length 13-17 mm. Diameter 0.34-0.43 mm. Somites 44-60. Color whitish. Prostomium blunt and rounded. Eead pore at O/I. Dorsal pores begin on VII. Setae of unequal length, inner ones finer and shorter: almost straight except for a distinct bend at the proximal end; 4-6 per bundle for almost the entire length of the body, sometimes as many 8 per bundle; 4-1 per bundle in the last few somites. Clitellum on XII-XIII. Lymphocytes numerous, elliptical to circular in outline. Brain about twice as long as wide; posterior margin distinctly convex; anterior margin slightly convex; lateral margins diverge caudad. Peptonephridia long, tuberculate, with but few branches arising irregularly along the main part of each organ. Dorsal vessel arises in XX. Nephridia with anteseptal and postseptal parts of about same size; efferent duct arises from anterior part of latter near septum. Spermiducal funnel with length about twice that of diameter, cylindrical; bent new posterior end, with well marked funnel-shaped collar; duct long, much contorted and confined to XII. Spermathecae each with pear-shaped ampulla which connects with digestive tract and which bears a circle of seven diverticula; duct about twice as long as ampulla; ectal end with a few small unicellular glands.

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Described from six sexually mature specimens. Cotypes in the collection of the writer. Cotypes also in the collection of Professor Frank Smith.

The specimens which are the basis of this description were collected near Oconee, Illinois, November 5, 1910. They occurred rather abundantly under the decaying bark of fallen timber. Of the total number of specimens taken only about one half were sexually mature.

### Affinities.

The character of the spermathecae places this species in the group having more than two diverticula on the spermatheca. A careful comparison of the characters of this species with the corresponding ones of the other species in this group shows that it is a distinct form. Difficulty is encountered in such a comparison with some of the foreign species since the descriptions are so brief that the relationships cannot, with any degree of accuracy, be determined. However no foreign species appear to be closely related to this form. As regards the American species of this group there is only one species which might be regarded as a near relative, namely Fridericia tenera Welch, and disregarding minor differences, the characters of the brain and the peptonephridia are sufficient to distinguish the two species.

## DETAILED MORPHOLOGY.

### EXTERNAL CHARACTERS.

The body is slender and has a length of 13-17 mm. In transverse section

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it is oircular and has in the region of the olitellum a maximum diameter of 0.34-0.43 mm. In the living specimens the body is opaque and whitish in appearance. The prostomium is blunt, rounded and rather short. The intersegmental grooves, excluding the first three or four, are very obscure. The number of somites is variable, the extremes being 44 and 60. The head pore is small and located at 9/I. The clitellum is well developed and occupies XX-XIII.

The setae bundles usually contain 4-6 setae. Sometimes the number is as high as E but this happens rarday. In the last four or five somites the number varies from 4 to 1. The setae are simple and straight except for the distinct bend at the proximal end. They are of different lengths in a bundle, the outer ones being longer and stouter, the inner ones shorter and more slender.

The head pore is on O/I. In one of the preparations the lymphooytes which are abundant in the anterior part of the body were passing out through this opening at the time of fixation. This seems to corroborate the statement of Coénot('97,p.90) that the coelomic fluids are often exuded through the "dorsal pores" when the animal comes in contact with some irritating substance and it is possible that in this instance the chloretone was the irritating substance.

The cuticula is thick, firm, and approximately uniform in thickness throughout the length of the body. In the first few somites the hypodermis is about two to two and one half times thicker than the cuticula but throughout the greater part of the body the latter has about the same thickness as the former and is sometimes even slightly thicker.

The presence of the thick cuticula in these specimens evidently

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supports that Vejdovsky makes('70, p. 11), namely, that those species living in comparatively dry localities are characterized by a thick outicula. The specimens of this species were found under the decaying bark of fallen timber and while the decaying wood in which they lived was somewhat damp, yet the percentage of moisture was low.

## INTERNAL CHARACTERS.

## Brain.

This organ(Fig. 34) occupies a median dorsal position in I and II, ohiefly in the latter. Both anterior and posterior margins are convex. the lateral margins converge cephalad. The averages of a number of measurements show that it is about twice as long as wide. These two dimensions differ somewhat in the different specimens but the ratio is nearly uniform. One set of measurements which is fairly representative is as follows:-length 0.153 mm., width 0.085 mm. A distinct neurilemma encloses the organ and is slightly thickened on the posterior margin. Two pairs of supporting strands attach the organ to the body wall, one arising near the mid lateral part and the other from a latero-posterior position. In transverse section the brain is almost circular. The anterior part gives rise to the usual nerve trunks and at a short distance anterior to their origin each divides into two parts one of which extends into the prostomium while the other extends arcund the digestive tract as one of the circumcesophageal commissures.

Peptonephridia.

Two of these organs(Fig.38) lie ventrad to the digestive tract, one on either side of the median line. Each opens independently into the

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into the cescophagus in a latero-ventral position in the posterior part of IV. There is some variation in length. In some specimene they extend to VII and in others only to VI. However the general structure is the same. Each organ immediately subsequent to its opening extends caudad parallel to the digestive tract. Each gradually diminishes in diameter towards the terminus. Each is  $\operatorname{roug}_{A}^{H}$  y tuberculate in appearance and gives off short single tubular branches at irregular intervals. The branching is much more marked at the extreme posterior end although even there it is sparse. Anteriorly the branches are given off only at wide intervals, are short, and show no secondary branching.

### Chylus cells.

Chylus cells occur in 1/2 XIV - XVIII. As in certain other species of Fridericia previously described the epithelial layer of the intestinal wall in this region(Figs. 35 and 39) is composed of two distinct kinds of cells, the epithelial cells(epc) which line the lumen of the intestine and the chylus cells(ch) which lie deeper in the intestinal wall and meet the lumen only at points between the former. The epithelial cells are wedge-shaped, the larger ends being entad. In sections the smaller ends appear inserted between the apices of the chylus cells either singly or in groups of two, usually the latter. Each epithelial cells contains a large elliptical nucleus(nu) in the ental end and the surface bordering onto the intestinal lumen has numerous long cilia. The ectal end is in contact with the perivisceral blood sinus(bs). Each chylus cell is flask-shaped; the broader end being ectad, and is about three times as long as broad. The base is broad and somewhat truncate, the ppex rather pointed and exposing little surface to the lumen of the

-69-



intestine, and the sides of the cell converge entad. The intracellular canal(ico) extends the full length of the cell. Its apical half is straight or nearly so but the basal part often shows two distinct spiral turns. The lining of this canal appears to be little more than a mere bounding membrane. The apical half of the intracellular canal only is ciliated. The relation to the perivisceral blood sinus is intimate. As shown in figures 35 and 39 the surface of the base and from one half to two thirds the length of the sides are in contact with the sinus. A distinct spherical nucleus lies in the base of each cell, usually in one of the broad angles made by the curves of the intracellular canal. Interstitial cells are absent.

The characters of the chylus cells are uniform and help to confirm Eisen's conclusion that they have taxonomic value. A comparison with other species in which the chylus cells have been worked out shows that none approach closely to the condition described above.

### Nephridia.

The nephridia are rather large and conspicuous. The anteseptal part (Fig.36) is approximately as large as the postseptal part. The first pair appears on VI/VII. The efferent duct arises from the postseptal part a short distance from the septum and opens exteriorly just anterior to the ventral setae bundles. The lumen is very tortuous throughout its entire length.

Spermiducal funnel.

The spermiducal funnel (Fig. 40) is cylindrical. The posterior end diminishes uniformly to the diameter of the sperm duct which arises from

-70-

it. The anterior end is characterized by a well marked, protruding, funnel-shaped collar, distinctly set off by a transverse constriction. The length of the funnel is about twice the diameter. Measurements show the length to be 0.135 - 0.15 mm., the diameter 0.074 - 0.085 mm. It lies in the usual position with the anterior end pointing dorsocephalad, due to the distinct bend in the body of the funnel. The lumen is eccentric as shown in transverse section, being nearer the ectal surface of the organ.

### Penial bulb.

The structure of the penial bulb (Fig. 41) conforms to the Lumbricillid type as defined by Eisen. It is composed of cells of one kind only. Each cell has two parts, the main body which lies near the periphery of the bulb and contains a large oval nucleus, and the prolongation which extends to the sperm duct extension. The body of each cell has a strong affinity for stains while the prolongation stains only very slightly. The sperm duct(sd) unites with the bulb on the dorso-ectal surface and there meets the sperm duct extension(sde). The whole organ is covered by a well developed musculature(m). In some respects the structure of this organ recalls that of Fridericia tenera Welch; the general structure being the same. However, when the bulbs of the two species are carefully compared distinct differences are noted in the shape of the organ, the mode of union of the sperm duct with the bulb, the musculature, and the oharacter of the component cells.

## Spermathecae.

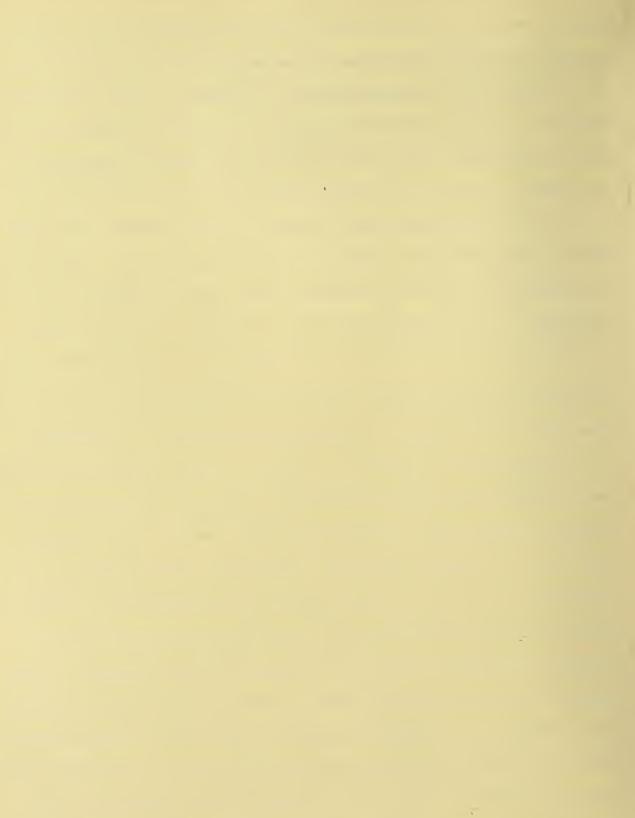
Each spermatheca has three well differentiated parts, the duct, the

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ampulla, and the diverticula. The duct opens laterad in IV/V. Two or three inconspicuous unicellular glands occur at this opening but they are so small that high magnification is required in order to distinguish them. The duct is approximately uniform in diameter throughout its length and is usually more than twice as long as the ampulla. Measurements of this duct in a number of specimens shows the length to be about 13-15 microns. The pear-shaped ampulla unites with the digestive tract well down on the side of the latter in the posterior part of V, thus making the openings of the two spermathecae almost directly opposite each other. The ampulla is 6-7 microns long, has a conspicuous lumen, and bears seven globular diverticula which are arranged in a single whorl. The diverticula differ somewhat in size but the general plan of each is the same. The lumen of each is continuous with the lumen of the ampulla.

An examination was made of a number of sexually immature specimens and it appears that the spermathecae are the last of the reproductive organs to attain their complete development. Some of these specimens with all of the other reproductive organs well developed showed small spermathecae with the ducts and ampullae well developed but lacking the diverticula, the only indication of their future position being the rounded collar-like shoulder on the ectal end of each ampulla. A later stage in the development was noted in which the above mentioned collarlike shoulder of the ampulla has begun to show slight divisions and the boundaries of the diverticula are becoming distinct. Intermediate stages showing transition between the two above described stages were not available but it is quite probable that the course of the development of these organs in this species is as follows:- (1) the development of the

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duct, (2) the development of the ampulla, and (3) the development of the diverticula.

Fridericia sima n. sp. Plate V. figs. 42-50.

pefinition.

Length 15-19 mm. Diameter 0.45-0.57 mm. Somites 52-58. Color whitish with slight tinge of yellow. Prostomium prominent and pointed. Head pore at O/I. Dorsal pores begin on VII. Setae of the typical Fridericia type; 4-6(rarely 7 or 8) setae per bundle in the anterior region; 4 (rarely 5) per bundle in middle region, and 3-2 setae per bundle in posterior region. Clitellum on XII - 2/3 XIII. Brain about twice as long as wide; posterior margin moderately convex, anterior margin very convex and slightly pointed at the extremity; lateral margins almost parallel. Peptonephridia simple, unbranched and confined to V. Dorsal vessel arises in XX. Nephridia with well developed anteseptal and postseptal, the latter being longer and slightly larger than the former; the efferent duct arises from the ventral side of the postseptal part about mid way of its length; small distinct reservoir present at the ectal opening of efferent duct. Spermiducal funnel cylindrical, about twice as long as the greatest diameter, with protruding collar. Spermathecae with duct and ampulla distinctly differentiated; ampulla simple, thin walled, pear-shaped, and devoid of diverticula; junction with digestive tract on dorsal surface of latter but distinct from corresponding junction of opposite spermatheca; duct about twice as long as ampulla, and with no glands at ectal opening.

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Described from 10 sexually mature specimens. Cotypes in the collection of the writer. Cotypes also in the collection of Professor Frank Smith.

The specimens of this species were collected in Cotton Woods, near Urbana, Illinois, April 1, 1911. They were found in undisturbed forest land, under the decaying leaves and to a slight depth in the rich humus, which contained a moderate amount of moisture. The majority of the specimens collected were sexually mature. The entire collection was made at one very small locality.

### Affinities.

The structure of the spermathecae puts this form in that group of species having no diverticula on the ampulla of the spermatheca. Of this group of species there are, at present, only six represented in North America, the majority being found in Europe. This worm evidently has no close relatives among the foreign forms since the differences existing between them are numerous and well marked. Of the six American species only two, F. alba and F. parva Moore(bulbosa Rosa), appear to be at all closely allied and even these show several distinct differences and no difficulty is experienced in separating them.

# DETAILED MORPHOLOGY.

## EXTERNAL CHARACTERS.

The body is slender, cylindrical and tapers from XII and XIII gradually towards the two extremities. The length varies from 15 to 19 mm. The diameter is greatest in the region of the clitellum and varies from 0.45 to 0.57 mm. The intersegmental grooves are obsolete in all

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parts of the body except in the extreme anterior region where the first and sometimes the second and third intersegmental grooves are well marked. The somites vary from 50 to 58. The olitellum is on XII-2/3XIII and is well developed. The prostomium(Fig. 43) is prominent and pointed. It varies slightly in shape but the typical condition is that indicated in the figure which shows the well developed, pointed prostomium overhanging the mouth and the slightly upturned tip. One or two of the specimens showed a condition in which the prostomium was shorter and more rounded but it is possible that this is due to a contracted condition of the animal. The color of the living worm is whitish with a slight tinge of yellow. The dorsal pores begin on VII. The setae are of the typical Fridericia type. They are well developed and the greater part of their length projects beyond the body wall. In the anterior region there are 4-6(rarely 7 or 8) setae per bundle; 4 setae(rarely 5) per bundle in the middle region, and 3-2 setae per bundle in the posterior region.

# INTERNAL CHARACTERS.

### Brain.

The brain(Fig.48) lies in the dorsal part of I and II, ohiefly in the latter. It is about twice as long as wide. The posterior margin is broadly convex while the anterior margin is strongly convex. The lateral margins are approximately straight and almost parallel, diverging only slightly caudad. In transverse section the brain is elliptical in outline, the longer diameter being approximately twice the length of the short one. Two pairs of supporting strands arise from the posterior part of the brain and pass obliquely across the coelom to unite with the body

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wall.

Peptonephridia.

The peptonephridia(Fig.42) are simple and unbranched. They arise from the digestive tract in the anterior part of V and extend caudad approximately parallel to it, ending at the posterior limit of V. The openings into the digestive tract are latero-ventrad. Both peptonephridia are practically of the same length and the free ends are usually closely approximated. In shape these organs are cylindrical, sometimes slightly flattened. The maximum diameter is just caudad of the point of union with the digestive tract and from there the organs taper gradually to their free extremities. Slight irregularities on the surface produce a tuberculate apearance, which however is not strikingly distinct. Sections show these organs to be tubular and conspicuous nuclei appear in the walls.

## Chylus cells.

These cells(Fig. 45) form a distinct region extending from the caudal third of XIV to the anterior part of XVII. In longitudinal sections this region is very easily located by the abrupt increase in the thickness of the intestinal walls. These cells are rather closely packed together forming an almost continuous layer. The chylus cells(ch) are flask-shaped, somewhat truncated at the ectal end, and the sides converge slightly entad. The intracellular canal(icc) is distinct. Its apical course is straight but the basal portion is bent almost at a right angle and is sometimes slightly sinuous. The lining of the canal appears to be a very thin cytoplasmic layer but in many of the preparations it

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is rather difficult to distinguish. Cilia are present for almost the entire length of the canal. The periviscenal blood sinus is quite spacious in this region and transverse sections of the worm show the sinus(bs) in contact with the greater part of the surface of the cells. The epithelial cells(epc) are more or less block-shaped, distinctly nucleated, heavily ciliated on the ental surface, and form a layer of which the continuity is broken only by the apical parts of the chylus cells. No interstitial cells are present.

#### Nephridia.

The nephridia appear first on VI/VII. The anteseptal part(Figs. 44 and 46) is well developed, ovate, and provided with a distinct ciliated nephrostome. The postseptal part is longer and slightly larger than the anteseptal part. There appears to be two divisions in the former, an anterior smaller one and a posterior larger one. The smaller region is a kind of neck by which the posterior larger one is joined to the anteseptal part. The origin of the efferent duct presents an interesting feature: It becomes distinct from the ventral surface of the postseptal part at about the middle of its length. A critical examination of the nephridia in the various parts of the body and in the various specimens reveals two rather distinct conditions which seem to throw light upon the true origin of the duct. A number of nephridia were studied in which it was evident that the posterior end of the nephridium was reflected ventrad upon itself (Fig. 44) for some distance and then diverged as the duct proper, thus pointing to the fact that the duct really arises from the posterior end of the postseptal part. It should be noted that this reflected portion comes into very close contact with the

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ventral side of the nephridium, so close that it is attached there, the line of union appearing in sections only as a single line of fusion. Quite a number of nephridia were found (Fig. 46) in which the condition of the caudal end of the postseptal part is not shown and the duct apparently has its origin from the ventral surface of the postseptal part about midway of its length. This variation may be interpreted by regarding the first condition as the transitional stage and the second as the resulting stage.

Another interesting condition connected with the efferent duct is the presence at its ectal extremity of a small but distinct reservoir. Just before the lumen of the duct reaches the exterior it expands into a more or less flask-shaped reservoir which in turn opens to the exterior through a small pore. The lumen of the nephridium is much longer and more contorted in the postseptal than in the anteseptal part.

### Spermiducal funnel.

The spermiducal funnel(Fig. 49) is cylindrical; usually slightly bent, and approximately twice as long as the greatest diameter. There is a distinct collar set off from the body of the funnel by a constriction. This collar is not reflected but is somewhat funnel-shaped.

### Penial bulb.

The penial bulb (Fig. 50) is of the Lumbricillid type. It is rather simple in its structure and moderate in size. In transverse section it appears as an elliptical structure with the long diameter approximately parallel to the penial invagination (pt). The greater part of the ental surface of the organ is covered by a well developed muscle layer (m) which

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is a continuation of the circular muscle layer of the body wall. The body of the bulb is composed of cells of one kind(glp) which have their enlarged nucleated parts near the periphery and each of which sends a long process in an eotal direction. These prolongations lie parallel to each other and the majority of them end radially around the sperm duct extension(sde). Others end on the ectal surface ventrad to the penial pore. The sperm duct(sd) unites with the bulb on the ectal surface. In the retracted state the sperm duct extension shows a very decisive bend of about 120° before it opens into the invagination. None of the specimens examined showed the bulb in the everted condition. Many of the preparations showed a break or cleft in the body of the bulb which conformed in direction to the long diameter and in a single section appeared to separate the nucleated ends of the cells from the greater part of the cell prolongations

### Spermatheoae.

Each spermatheca(Fig. 47) consists of two clearly differentiated parts, the ampulla and the duct. The ampulla is simple, pear-shaped, and has no diverticula. Its inner end is united with the latero-dorsal part of the digestive tract in the posterior part of V. The cavity of the ampulla conforms in general to the shape of the exterior. The walls are rather thin except at the ectal end where they are conspicuously thicker. The duct extends, with very few curves, to its external opening in the anterior part of V. There are no glands at the ectal opening. The duct is about twice as long as the ampulla. The external hypodermis surrounding the external opening of the duct is distinctly thickened.

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## 6. Genus ENCHYTRAEUS Henle.

The genus Enchytraeus was established by Henle in 1837 and although the limits of the group have altered from time to time it appears to have a permanent place in the nomenclature. Michaelsen('00,p.88) defines the genus as follows:-

"Borsten in 4 Bündlen, 2 ventralen und 2 lateralen, gerade, die eines Bündels gleich lang. Kopfporus klein, dorsal zwischen Kopflappen und 1. Segm.; Rückenporen fehlen, Ursprung des Rückengefässes postclitellial; Blut meist farblos; Herzkörper fehlt. Lymphkörper von einerlei Gestalt. Peptonephridien vorhanden oder fehlend; der Oesophagus geht allmählich in den Mittledarm über. Ausführungsgang der Nephridien am hinten Pol des Postseptal entspringend, Meist sehr kurz. Samenleiter lang. Samentaschen mit dem Darm kommunizierend, ohne Divertikel".

In the light of recent work this definition is somewhat faulty. The statement that the spermathecae communicate with the digestive tract does not always hold since in E. modestus Eisen there is no such connection. Also the statement that the spermathecae have no diverticula can no longer be made since species have been described(E. alaskae Eisen, E. saxicola Eisen, E. citrinus Eisen) in which there are diverticula on the ampulla.

Eisen('04,p.61) modified the definition of the genus as follows:-"Setae of equal length and straight. Head pore between the prostomium and somite I, always small. No dorsal pores anterior to clitellum. Intestine and cesophagus gradually merging into each other. Dorsal vessel rises posterior to clitellum from a vascular sinus of the

-80-

intestine. One pair of sperm-sacs, surrounded by peritoneal membrane, project from the testes forward. No single penial bulb, but one or more isolated glandular papillae situated in the vicinity of the spermiducal pores, generally and principally ventral to the pores. Numerous transverse muscles connect the ventral and lateral parietes surrounding the spermiducal pores. Peptonephridia glands present or absent. One kind of lymphocytes. Intestine generally with chylus cells".

It will be noted that in the above definition statements concerning the sperm-sacs and the structure of the penial bulb have been added. It seems doubtful if either of these two points can be considered as constant features of the genus. In E. gillettensis n. sp. and E. indicus Stephenson sperm sacs are absent and in certain species recently described, E. nodosus Steph., E. dubius Steph., and others, no mention is made of them. Furthermore E. gillettensis n. sp. has a single compact penial bulb, not broken up into isolated glandular papillae. E. nodosus Steph. is similar in this respect. It thus appears that the penial bulb of this genus as described by Eisen is not a constant feature.

About thirty two species have been assigned to this genus and of this number eight have been recorded from North America and its adjacent islands. Seven of the eight species were originally described from this continent. The list is as follows:-

| Name                    | Type Locality                        |
|-------------------------|--------------------------------------|
| E. modestus Eisen       | Oraca, Prince William Sound, Alaska. |
| E. metlakatlensis Eisen | Metlakatla, Alaska.                  |
| E. kincaidi Eisen       | Popof Island, Alaska.                |

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-82-E. alaskae Eisen Garforth Island, Muir Inlet, Glacier Bay, Alaska. Muir Inlet, Glacier Bay, Alaska. Muir Inlet, Glacier Lowe Inlet, British Columbia. Bermuda Islands. E. albidus Henle Germany(American locality = Mass.). KEY TO THE SPECIES OF ENCHYTRAEUS KNOWN TO OCCUR IN NORTH AMERICA.

A. Diverticula present on ampulla of spermatheca.

- B. One diverticulum on ampulla of spermatheca.
  - C. Spermiducal funnel with length not exceeding four times the diameter.
    - D. Peptonephridia present; brain with convex posterior margin; length 20-25 mm. - - - - +E. kincaidi Eisen.
    - DD. Peptonephridia absent; posterior margin of brain with slight median concavity; length about 10 mm. - -E. marinus Moore.
  - CC. Spermiducal funnel very long and narrow, length exceeding four times the diameter; posterior margin of brain with deep emargination; extensive sperm sacs present; length 15-20 mm.

BB. Two diverticula on ampulla of spermatheca.

- C. Posterior margin of brain concave; diverticula of spermatheca equal in size - E. alaskae Eisen.
- CC. Posterior margin of brain convex; diverticula of spermatheca unequal is size - E. citrinus Eisen.

AA. No diverticula on the ampulla of the spermatheca.

B. Spermatheca not connected with digestive tract; straight and more or less covered throughout its length with small glandular cells; no distinct and enlarged ampulla - E. modestus Eisen.

BB. Spermatheca connected with the digestive tract.

C. Spermatheca short and stout, the duct being of about same length as ampulla; length not exceeding 10 mm.; somites over 50.



- D. Duct of spermatheca not set off distinctly from ampulla; connection with digestive tract situated at one side of ampulla and not at apex; large compact orown of glands at ectal opening  $- - \div - - - - - E$ . metlakatlensis Eisen.
- DD. Duct of spermatheca distinctly set off from ampulla; connection with digestive tract apical; ectal half of duct covered with closely set pear-shaped gland cells - + - + - - - - - E. albidus Henle.
- CC. Spermatheca long and slender, the duct being about 4 times as long as ampulla; glands cover the entire length of duct; length 2.5 - 4.5 mm; somites 25-27 - E. gillettensis n. sp.

Enchytraeus gillettensis n. sp.

Plate VI, figs. 51-55.

#### Definition.

Length 2.5 - 4.5 mm. Diameter 0.144-0.188 mm. Somites 25-27. Color whitish. Prostomium blunt and rounded. No dorsal pores. Lymphocytes few in number, ovoid, nucleated. Setae straight, those of a bundle of equal size; number per bundle 2-3, rarely 4 or 5. Clitellum on XII-XIII. Brain about twice as long as broad; anterior margin concave; posterior margin almost straight; lateral margins converging cephalad. Peptonephridia present; arise from dorsal surface of pharynx in III as two separate structures; slightly branched; usually terminating in a large mass. Dorsal vessel arises in XIV. Anteseptal part of nephridia small and inconspicuous, comprising little more than the nephrostome; postseptal part large; short efferent duct arises from posterior end of postseptal part. Length of spermiducal funnel 3-4 times the greatest diameter: collar distinct; distinct bend in middle of funnel. Duct of spermatheca about four times the length of ampulla and completely covered with small glands; ampulla spherical, thin walled, destitute of diverticula, and connected with digestive tract.

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Described from 15 specimens all of which are sexually mature. Cotypes in the collection of the writer. Cotypes also in the collection of Professor Frank Smith.

The specimens which have formed the basis for this description were collected by Dr. George R. LaRue at Gillette Grove, Iowa, in August, 1910. They occurred in damp soil under the drip of a building. The soil was black and contained some humus.

# Affinities.

Nothing definite can be accomplianed in attempting to establish the relationshpis of this species owing to the incomplete and ambiguous descriptions of some of the European species. All that can be done at present is merely to call attention to the apparent similarity which exists between this form and some of the other species as judged by those characters which are described in sufficient detail to be used in diagnosis, keeping in mind meanwhile the possibility that future investigation may show that they are not at all closely related.

This species approaches E. argenteus Mohlen. closely in some respects but shows differences in the character of the nephridia, the spermiducal funnel, and the spermathecae. In some respects it resembles E. sabulosus Southern but distinct differences exist in size, in number of somites, brain, and one or two other organs. It resembles E. indicus Stephenson but differences exist in the peptonephridia and spermathecae. Of the American species it more nearly approaches E. albidus Henle but here again distinct differences occur in length, number of somites, and spermathecae.

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# DETAILED MORPHOLOGY.

## EXTERNAL CHARACTERS.

Twenty seven alcoholic specimens have a length of 2.5 - 4.5 mm. They had been carefully killed and fixed and since microscopical examination of the specimens in toto showed no evidence of conspicuous contraction it is safe to consider these measurements as fairly accurate. The number of somites is rather constant varying within the limits of 25 and 27. The greatest diameter which is in the region of the clitellum is 0.144 - 0.188 mm. The body is smooth, cylindrical, and tapers very gradually audad from the clitellum. The clitellum is only moderately developed and occurs on XII and XIII. It is interrupted on the mid ventral surface. The intersegmental grooves are more distinct in the anterior region; posteriorly they tend to become obsolete. The prostomium is blunt and rounded. The setae have been sufficiently described in the definition.

### INTERNAL CHARACTERS.

#### Lymphocytes.

The lymphocytes are scattered throughout the greater part of the coelom, but occur in only small numbers. They are nucleated, ovoid, and have a decided affinity for stains.

## Chloragog cells.

The elongated chloragog cells are club-shaped and very highly developed. They first appear in V and from that point caudad they almost completely cover the digestive tract, filling the greater part of the

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coelom. The cytoplasmic contents are distinctly reticular and sometimes appear to be alveolar in structure.

### Brain.

The brain(Fig. 52) lies chiefly in I and II. The length varies slightly in the different specimens but the ratio of the length to the greatest diameter appears to be almost uniformly 11 : 6. The posterior margin is approximately straight although in some specimens there is a slight convexity. The lateral margins converge anteriorly so that the smallest width occurs just posterior to the origin of the commissural trunks. The anterior margin is concave and slightly V-shaped. A neurilemma surrounds the brain and appears to be thickest about the posterior margin. Two pairs of strands connect the brain with the body wall, one pair arising from the lateral margins near the point of greatest diameter, the other arising from the region of the junctions of the lateral with the posterior margin. In transverse section the brain is ovoid in shape. Close examination of cleared specimens showed the presence of an elliptical area located in the region of greatest width. Eisen('04, p. 62) calls attention to this area as follows "- " The brain in Enchytraeus is characterized by the circular mass of fibers in the posterior part of the fiber belt in the brain. As this structure has not been studied in detail its nature is not understood". Dr. Eisen evidently regarded this structure as more or less characteristic of the genus although he makes no mention of it in his descriptions of the brains of E. alaskae Eisen, or E. citrinus Eisen, and his figures of these species do not show any indication of its presence. The writer has nothing to contribute to our knowledge of this structure but there is

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no doubt of its presence in E. gillettensis.

#### Peptonephridia.

These organs are a conspicuous component of the anterior region of the body. Both are similar and arise, one on either side, from the dorsal surface of the pharynx immediately posterior to its dorsal epithelial thickening in III. They extend caudad for about the length of one somite. Each shows irregular branches and makes irregular contortions in the coelom on either side of the digestive tract. These organs terminate in a peculiar manner. In some specimens both peptonephdidia merge into a large mass dorsad of the digestive tract; in other specimens only one organ ends in this mass and in one preparation this mass is entirely absent.

# Nephridia.

These organs(Fig.51) are large and appear as conspicuous masses on the floor of the coelom. The anteseptal part is small and inconspicuous, being little more than a nephrostome. The postseptal part is large, ovoid, and comprises the bulk of the organ. It appears to differ slightly in shape in the various regions of the body, the anterior ones being somewhat shorter and thicker while those posterior to the clitellum are a little more elongated. The short efferent duct arises from the posterior surface of the postseptal part, bends abruptly ventrad, sometimes also slightly cephalad; and opens to the exterior just ventrad of its posterior end.

## Spermiducal funnel.

The spermiducal funnel (Fig. 54) is rather small, cylindrical, and is

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3-4 times as long as broad. The collar is well developed and is set off by a distinct constriction. The funnel usually shows a marked bend in the middle, the convex aspect being dorsæd. The maximum diameter is just back of the collar. From this point it diminishes gradually so that the funnel merges into the sperm duct without any abrupt decrease of diameter.

## Penial bulb.

The penial bulb(Fig.55) is compact, globular, and enclosed in a simple muscular investment. It is composed of cells of a single kind and all appear to empty onto the body surface. Large spherical nuclei lie in the ental ends of the cells so that in a transverse section there is is a continuous row of them around the periphery of the bulb. A few nuclei appear in the vicinity of the sperm duct. This duct(sd)penetrates the bulb on its ectal side and, after making a bold curve within the bulb, opens into the shallow invagination(pb). The bulb is covered by a thin sheet of peritoneum beneath which lies a thin layer of muscle tissue(m). This musculature does not penetrate the bulb nor is the bulb divided into separate parts as seems to be the case in some species of this genus.

# Spermathecae.

Two moderately developed spermathecae are present in V. Each(Fig.53) is distinctly differentiated into two regions, the duct, and the ampulla. The duct is about four times longer than than the ampulla and is covered throughout its entire length with small glands which give it a tuberculate appearance. The lumen is very fine and the walls of the duct are thick.

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There seems to be a few additional unicellular glands at the ectal opening but they are small and not easily seen. The ectal opening is somewhat latero-ventral in position and occurs near IV/V. From this point the duct extends, without contortions, in a dorso-meso-caudal direction. Before reaching the digestive tract it merges into the ampulla. The ampulta is spherical, thin walled, and unites with the lateral surface of the digestive tract by means of a short duct-like extension: All of the specimens studied showed masses of sperm in the globular ampulta.

7. THE PENIAL BULB AS A CHARACTER IN CLASSIFICATION.

Previous to 1904 the structure of the penial bulb had not been oritically examined and no attempt had been made to discoved in it characters of taxonomic importance. It had been seen and very briefly described by some of the earlier workers(Vejdovsky, Michaelsen, et al) but the finer details of structure were neglected. As a consequence soarcely any of the earlier publications on Enchytraeidae give information either in text or figures which can be used in estimating the taxonomic value of this organ. Eisen('04) made critical studies of it in about fifty species distributed among eight genera and gave descriptions and figures of the structure of the organ in each case. Unfortunately these species were not evenly distributed among the eight genera since in two of them only one species of each was examined and only two species in another. The most thorough going examination was

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made in the genus Mesenchytraeus in which twenty species were studied. The results of this extended study are given by Eisen('04,p.6) as follows:-

"The present arrangement of the various genera is partly tentative. Until now the structure of the penial bulb has not been critically examined, except in a few species besides those described in this paper, and it is in reality only a supposition that the structure of the penial bulb is uniform in the respective species of a genus. I think, however, this assumption will prove to be correct. The species within each of the genera which have been examined have proved to correspond in all particulars to such an extent that it may be safely assumed that the other species will also agree". \*\*\*\*\*\*\*\*\*\* The copulatory cushion or penial bulb is of considerable importance in the classification of Enchytraeidae, and I have as far as it has been possible investigated its structure in all of the species described in this paper. " \*\*\*\*\*\*\* \*\*\* "It seems almost certain that a great uniformity of structure exists in the different species of the same genus; or in the same genera of the various subfamilies. The structure of the penial bulb or corresponding organs can therefore be said to be highly characteristic of both species, genera and subfamilies".

According to Eisen there are three distinct kinds of bulbs which he defines as follows:-

I. The Mesenchytraeid Bulb.

"The Mesenchytraeid Bulk is a single muscular structure, containing circular as well as fan-shaped muscular bands connecting the body wall with the periphery of the bulb. Between the muscular bands are generally

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found numerous penial glands which open on the surface of the bulb around the penial pore. The sperm-duct penetrates the bulb, opening on the center of its outer surface".

II. The Enchytraeid Bulb.

"The Enchytraeid bulb is multiple, consisting of several separate cushions grouped around the penial pore. In these cushions we find several sets or fascicles of glands, each fascicle opening by itself on the surface of the body. There are no muscular bands connecting the base of the cushions with its periphery. The sperm-duct never penetrates the bulb or cushions but opens close to and independently of them. Exterior to the cushions there are numerous muscles connecting the body wall immediately surrounding the pore with other parts of the same somite."

# III. The Lumbricillid Bulb.

"The Lumbricillid Bulb is always single and covered with a strong muscular layer, which however never penetrates down between the cells of the bulb. There are generally two or three distinct sets of gland cells in the bulb. Some of these open in the lower part of the spermduct, or rather in a narrow groove in the elongation of the sperm-duct. Others open on the free surface of the bulb, either irregularly or in narrow circular fields, bunched into fascicles. The sperm-duct penetrates one side of the bulb. In Bryodrilus the gland which opens into the extension of the sperm-duct is covered with a thin cushion of muscular strands, forming a bulb within a bulb".

Eisen distributed the eight genera examined as follows:-I. Mesenchytraeid Bulb. Mesenchytraeus.

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II. Enchytraeid Bulb.

Enchytraeus.

Michaelsena.

III. Lumbricillid Bulb.

Lumbricillus.

Marionina.

Bryodrilus.

Henlea.

Fridericia.

Eisen was convinced that the structure of the penial bulb is of "great taxonomic importance" and he used it as the chief character in distinguishing subfamilies, added it to the definitions of the genera, and gave it a prominent place in his descriptions of new species.

The family Enchytraeidae now contains sixteen genera and nearly three hundred species. Since Eisen's investigation was based on about fifty species distributed among eight genera it is evident that his work must be extended and his conclusions tested on other species and genera before the structure of the penial bulb can be considered as a safe diagnostic character. Considerable work has been done on foreign Enchytraeidae since 1904 but it has been in the form of numerous small papers, no comprehensive works having appeared. As a consequence the systematic value of the penial bulb has been but little discussed. Stephenson('11,p.b4) is inclined to doubt the importance of the use of the penial bulb"as a basis for the distinction of subfamilies or even perhaps of genera". Most other foreign workers have been noncommittal on this subject.

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There is no doubt that the discovery of taxonomic characters in the structure of a rather conspicuous internal organ such as the penial bulb is a step in advance and the desirability of adding to the somewhat limited list of specific and generic characters in Enchytraeidae is obvicus to one who has worked in the group. The writer has given special attention to the structure of this organ in all of his work and oritical studies have been made on all of the species and genera available not only with the view of determining the minute structure of this interesting organ but with the view of testing the validity of Eisen's conclusions as to its systematic importance, and if possible, of adding something new to the data already accumulated.

Some interesting results have been obtained. In some cases they lend support to Eisen's conclusions, in others it is apparent that certain limitations and alterations must be made in Eisen's system, and in still others certain generic differences given by Eisen do not hold. Owing to the importance of this subject the results of the present study will be discussed in some detail.

The studies of the writer on the penial bulb have been made on fourteen species distributed among five genera as follows:-

- Henlea urbanensis n. sp. Henlea moderata n. sp.
- 2. Marionina forbesae Smith and Welch.
- Lumbricillus rutilus n. sp.
   Lumbricillus insularis Ude.
- Enchytraeus gillettensis n. sp. Enchytraeus albidus Henle.

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5. Fridericia agilis Smith. tenera Welch. firma Welch. agricola Moore. sıma n. sp. cooneensis n. sp. douglasensis n. sp.

For convenience each genus will be discussed separately.

## Henlea.

H. moderata and H. urbanensis have penual bulbs of the Lumbricillid type as defined by Eisen and exhibit no peculiarity of structure which calls for alteration in his conclusions.

### Marionina.

Eisen('04,p.90) makes the following statement concerning the character of the penial bulb in this genus:- "Penial bulb without internal muscular strands." \*\*\*\*\*\*\*There are two sets of glandular cells opening into the bulb. One set opens into the lower part of the sperm duct, while the other opens into the base around the pore". According to his investigations both M. elaskae and M. americana conform to this description.

Studies on M. forbesae have shown that the bulb is of the Lumbrioillid type in being a compact single structure invested in a musculature which does not penetrate into the interior but it does not have two sets of glands as described by Eisen. Instead the cells are all of one kind and all apparently open onto the surface of the bulb. None could be found which emptied into the sperm duct extension. Michaelsen('05,pl.1) figures the penial bulb of M. falclandica and

although it cannot be determined whether any of the cells open into the sperm duct, all of the cells in the bulb appear to be of the same kind. The bulb, however, is of the compact Lumbricillid type.

Benham('05, p. 294, pl. XIV, fig. 9) describes and figures the penial bulb of M. antipodum which is a very small structure composed of similar cells through which the sperm duct penetrates. Opening into the dorsal surface is a conspicuous accessory gland which lies entirely outside the bulb and is much larger than the latter. Michaelsen('05) described the penial bulb in M. werthi as a small structure entirely concealed in the body wall and possessing an accessory(prostate) gland which extends into the body cavity.

It appears that the original limits of constancy of structure in the penial bulb as laid down by Eisen do not hold. This organ in M. antipodum and in M. werthi does not fall under the Lumbricillid type since it has an accessory gland, the absence of which is emphasized as a character of this type. Therefore it will be necessary to modify the definition of the subfamily Lumbricillinae by making provision for the presence of accessory glands in Marionina. Furthermore it can no longer be said that the bulb in Marionina has uniformly two sets of gland cells within it.

#### Lumbricillus.

Studies on L. rutilus and L. insularis show that in both species the structure of the penial bulb conforms to the general Lumbricillid type as defined by Eisen and call for no special comment in this connection. It also appears that the descriptions of this organ which have appeared in the literature since 1904 agree with the definition

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of the Lumbricillid type. Thus far no deviation from this type is known to occur in this genus.

#### Enchytraeus.

The characteristics of the Enchytraeid type of penial bulb as described by Eisen have already been discussed. In defining the subfamily Enchytraeinae the following statement('04,p.61) is given concerning the penial bulb:- "In this family the penial bulb structures are not confined within a single bulb as in Lumbricillinae, but are broken up in two or more masses of papillae, often of unequal size. In a cross section of the body these papillae may be seen to extend from the median line to the other side of the spermiducal pore, and even in the long diameter of the body the glands have a more or less considerable extension. In some species these glands are situated close to each other; in others again they are separated by the common tissue of the body wall".

According to Eisen two genera, Enchytraeus and Michaelsena; are characterized by this type of penial bulb. In defining the genus Enchytraeus the following statement(l.c.p.61) is made:-" No single penial bulb, but one or more isolated glandular papillae situated in the vicinity of the spermiducal pores, generally and principally ventral to the pores. Numerous transverse muscles connect the ventral and lateral parietes surrounding the spermiducal pores".

The studies made by the writer on this genus show that Eisen's diagnosis connot be used safely in distinguishing it. E. gillettensis conforms in no respect to the type of penial bulb which Eisen claims to be uniform for the genus but instead has a single compact glandular bulb in which the muscular investment does not penetrate into the

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• , interior. No penial structures are present outside of the bulb and the sperm duct penetrates the bulb. In fact, it is a typical Lumbricillid bulb. Stephenson('11,p.50,pl.XLVIII,fig.10) describes and figures the penial structure in E. nodosus and here also the Lumbricillid type of bulb is present. It appears that at least two species are radically different in the structure of the penial bulb from the Enchytracid type.

The writer has also made studies on the penial structure in sections of E. albidus, made from specimens collected at Woods Hole, Mass., by Professor Frank Smith. Michaelsen('86,p.39,pl.II,fig.3) described and figured the structure of the penial bulb in E. mobil which has been shown to be a synonym of E. albidus. The specimens from Mass. have identically the structure described by Michaelsen. However the penial structure does not entirely conform to the Enchytraeid type. It is broken up into a number of glandular parts or fascicles but departs from the Enchytraeid type in having a large central division which is simple, compact, globular, invested in a musculature and penetrated by the sperm duct, all of which are characteristics of the Lumbricillid bulb.

Stephenson('11,p.56) described a new species, E. dubius, in which the penial bulb is described as follows:-"The penial gland is not large; its peculiarity is that it is bifid internally; thus in a series of longitudinal sections it is first met with as a single mass, while, nearer the middle line, it is completely double. It is attached by two thick strands, composed of cells with large oval nuclei, to the dorsolateral body wall". The same writer('12,p.240) described a new species, E. indicus, in which the penial bulb is a typical Lumbricillid bulb. Southern('09,p.158) described a new species, E. lobatus, in which the character of the bulb is given as follows:- "Duct(sperm) ends in a penial

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bulb, half as large as the funnel".

The results may be stated as follows:-

I. It is evident that the limitations laid down by Eisen concerning the variation of the penial bulb in Enchytraeus cannot hold. Future studies may show that the Enchytraeid form of penial structure is the common one but it cannot be used as a diagnostic character.

2. It appears that there are transitional stages in the structure of the penial bulk in the various species of this genus ranging from the Lumbricillid to the Enchytraeid type. The bulb in E. gillettensis and E nodosus is distinctly Lumbricillid; in E. dubius there is a partial division of a Lumbricillid bulb; in E. albidus one of the several fascicles is Lumbricillid in type; and finally in a number of species the typical Enchytraeid type prevails. This may be regarded as additional evidence in support of the contention of Stephenson('11) that Enchytraeus and Lumbricillus are intimately related and are not widely separated as was formerly supposed.

3. Until the structure of the penial bulb is thoroughly worked out in many of the known species of this genus, any attempy to make necessary modifications in Eisen's classifications must of necessity be only tentative. At present the stability of the subfamily Enchytraeinae is in question and future work may necessitate its elimination. There is a bit of evidence which may prove to be of service in the final adjustment of the matter, namely, the absence of penial bulb cells which open directly into the sperm duct. This seems to be the condition in all of the species of Enchytraeus which the writer has studied as well as those worked up by other investigators. However it seems doubtful if this one

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feature can be used as a diagnostic character of the genus since a similar condition appears to exist in certain species of at least one other genus.

# Friderioia.

Eisen('04,p.108) oharacterizes the penial bulb of Fridericia as follows:- "The penial bulb of Fridericia is quite characteristic and seems to be of similar structure in all of the species investigated by the author. There is only one kind of cells filling the bulb. The cells all open in the extension of the sperm duct and along the surface of the bulb; the duct convects with the bulb at the base of the latter and cannot strictly be said to enter the bulb".

It is necessary to note that Dr. Eisen refers to those cells which open into the sperm duot extension and those which open onto the surface of the bulb as one kind of cells in his general statement and in the description of several species he refers to them as different kinds of cells stating that the bulb in several species"has two kinds of cells". This inconsistency is due to the fact that in the bulbs studied by him the cells are all similar but in some cases the cells of a bulb open onto two different surfaces and these were unfortunately referred to as "two kinds of cells". However it is evident from his descriptions and figures that all of the species which he studied had bulbs in which the cells were all of one kind.

The work done by the writer shows that although the bulb is uniformly Lumbricillid in all respects it is necessary to make a slight alteration in the characterization of the bulb in Fridericia. It was found that the seven species examined by him can be divided into two groups, one

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composed of F. tenera, F. sima, and F. cooneensis in which the bulb is made up of cells of one kind only some of which open onto the extension of the sperm-duct and some may open onto the external surface; the other group composed of F. agilis, F. firma, and agricola in which the bulbs possess two distinct types of cell, one of which occupies the peripheral parts of the bulb and may open either into the sperm duct extension or onto the external surface; the other cocupies the interior of the bulb and the cells are arranged radially about the sperm duct extension, opening directly into the latter. These two types of cell are quite distinct showing uniform, marked differences in position, shape, size, size of nuclei, and staining reactions.

It appears that the structure of the penial bulb in F. douglasensis represents a transitional stage between the two groups mentioned above since the inner cells which open into the sperm duct extension in F. agilis, F. firma, and F. ggricola are represented only by a few scattering cells disposed at irregular intervals between the extensions of the peripheral cells.

The above results show that the original statement regarding the character of the bulb must be revised so that provision is made for the occurrence of two kinds of cells in the penial bulb of certain spocies of the genus. Otherwise Eisen's conclusions are supported.

At present little can be positively stated concerning the importance of the penial bulb in separating the species of any given genus owing to the incompleteness of the data for the whole group. The writer has found that in each species studied the structure of the bulb is constant

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and that in no case is it exactly duplicated in the bulk of another species. In certain genera, as for example Mesenchytraeus and Enchytraeus where the penial structure is usually very complicated and the variation rather wide it is not difficult to find disinguishing characters for species in the structure of the penial apparatus. The main difficulty appears to occur in the Lumbricillid bulb, particularly in Fridericia where the structure of the organ is at its simplest and where the variation among the species is so small that although the structure is uniform for each species, it is difficult to get a distinct diagnostic character.

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IV. OBSERVATIONS AND EXPERIMENTS ON LUMBRICILLUS RUTILUS N. SP.

1. Introduction.

The writer's attention was first called to this species in April, 1911, when alcoholic material of the same was turned over to him by Professor S.A.Forbes to whom it had ben sent by the Director of the Thirty-ninth Street Sewage Testing Station, Chicago, Illinois. This material was accompanied by the information that this worm occurred in great abundnace in the sprinkling filter beds. The specimens were in such poor histological condition that the attempt to determine the species was abandoned. Later, June 22, 1911; Mr. A.A.Girault made a collection of similar material at Chicago, made a brief record of the conditions of the habitat, and had a large number of these specimens properly killed and fixed. This material formed the basis for the morphological and systematic work on this species which appears elsewhere in the paper. During October, 1912, the writer spent three weeks at the Chicago Sewage Testing Station making certain investigations on this species and the major part of the data on the living worms which follow were accumulated during that period.

#### 2. Habitat.

Since this is the first published record of an American species of Lumbricillus which ocours in connection with sewage and since the great abundance indicated a particularly favorable environment, the habitat was carefully studied and will be described at some length. It may be noted here that there are records of about six European species which

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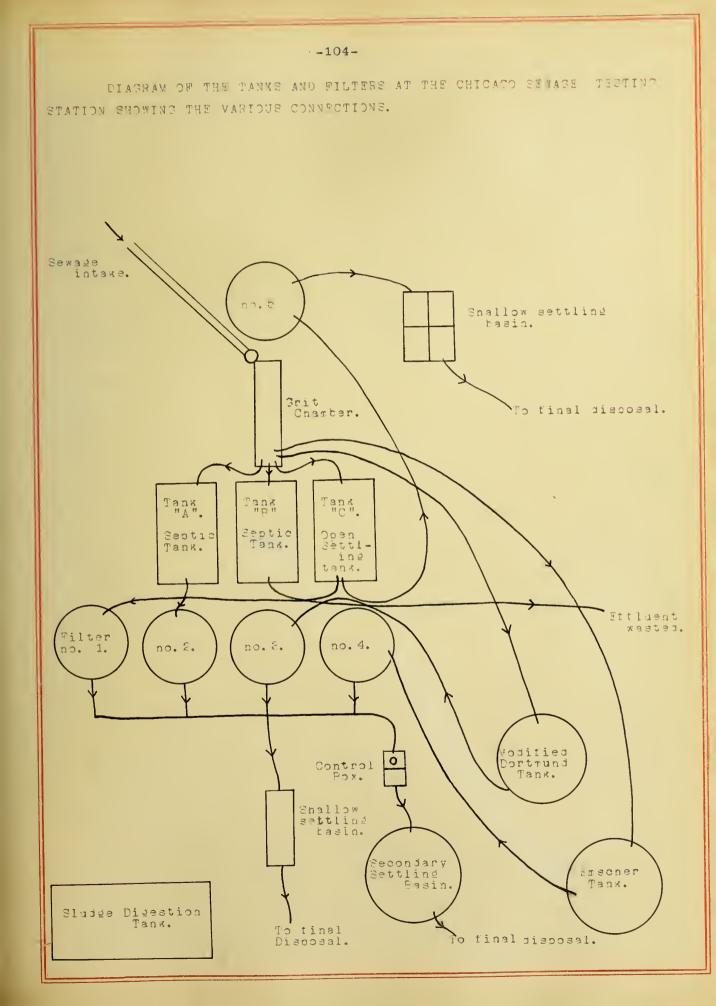
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ocour normally in connection with city water supplies, in sewere, near dung heaps, and various places containing decaying organic matter. Much of the detail which follows is essential to subsequent discussions and explanations of results.

The Thirty-ninth Street Sewage Testing Station is equipped with a Grit Chamber,  $20 \times 4 \times 3$  feet, with a capacity of 1,000 gallons at flow depth; three tanks, each 40 feet long, 9-7 feet deep and 7 1/2 feet wide, total capacity each 19,000 gallons, flow 17,000 galbons; Tank "A" is an open Septic tank with a nominal eight hour period, Tank "B" is a closed Septic tank, Tank "C" is an open settling tank. An Inhoff or Emscher Tank 7 1/2 feet in diameter and 17 feet deep and a modified Dortmund tank 7 1/2 feet in diameter and 9 feet deep are also in operation. Five sprinkling filters in cylindrical wooden tanks are connected up with the settling and septic tanks. Four are open and one is closed. Various settling basins and a Sludge Digestion Tank 29  $\times$  7  $\times$  6 feet with a total capacity of 8,900 gallons are also in operation. The following diagram will indicate the relation of these various tanks and filters to each other:-

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The one important thing to note in connection with the work which is to follow is the nature of the effluents of the various Settling and Septic Tanks. In brief, the aim of modern sewage disposal involves two distinct steps:- (1) the disposal of the small amount of suspended matters as soon as possible and (2) the oxidation of the fresh sewage on biologic filters in case additional treatment is demanded.

The first step is accomplished by passing the sewage through the Grit Chamber in which the heavier mineral particles are deposited. At the Testing Station; for experimental purposes, the sewage then passes in part to the Septic Tanks and in part to the Settling Tank. In the former anaerobic decomposition of the sewage goes on, eventually resulting in a series of chemical changes which from the point of view of purity often render the effluent a great deal worse than the raw sewage entering it. In the Settling Tanks the sludge is removed at frequent intervals in the early stages of decomposition with the result that the effluent is improved by this process. The principles of the Septic Tank and the Settling Tank are combined in the Emscher or Inhoff Tank which is a device permitting the escape of a"fresh" effluent, while the suspended matter settles, dropping into a separate sludge digestion chamber. The modified Dortmund Tank( a kind of Settling Tank) or Biolitic Tank is of such construction that sulphate reduction; due to bacterial activity is typical and at times as much as 40 p.p.m. of hydrogen sulphide have been observed in its effluent.

The second step consists in biologic treatment or oxidation of the sewage. This is accomplished in the sprinkling filters which receive the various effluents from the Septic and Settling tanks. Since these filters are intimately connected with this work a brief description will

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be given. Sprinkling filters are not built primarily as a device for removing suspended matter, but as a means of oxidizing and mineralizing the organic matter delivered to it. Each sprinkling filter consists essentially of a bed of orushed lime stone, 4 1/2 to 10 feet deep, supplied with a central top-surface intermittent spray which constitutes the influent. The size of the stone, the depth of the bed, and the period of the spray is different in each filter. For example in filter no. 4 the flow period is about 60 seconds with an intermission of about 30 seconds. The daily flow is approximately 10,000 gallons.

Sprinkling filters function to considerable extent in the catching of suspended matter and as a consequence sludge accumulates on the stones. The chemical nature of this sludge depends upon the character of the influent.

According to the observations of Dr. Arthur Lederer and others connected with the Testing Station Lumbricillus rutilus has a seasonal distribution. The worms apparently disappear completely at the approach of winter(November or early December). In Varch or April they begin to make their appearance in all of the sprinkling filters and their effluents. In a short time they become extremely abundant in the filters and large numbers of them are carried out into the effluents. The period of maximum abundance is apparently rather short but the worms are abundant throughout most of the summer and begin to diminish in numbers as the autumn wanes. An interesting fact in connection with the period of maximum abundance will be discussed in another connection. No careful observations have as yet been made regarding the relative abundance in the various filters. Girault(June 22, 1911) found them most abundant in

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filter no: 5 which is covered. However the Sanitary Engineer states that there is usually an abund(nace in all of the filters, except no.3, at that time of year.

During the writer is residence at the Station (Oct. 5-25) a careful examination of all of the Tanks, Settling Basins, and Filters was made with the view of determining the distribution of the species at that time of year. It was found that the worms were confined to certain of the Filters and their effluents. The greatest abundance occurred in Filter no.4, a few were found in Filters no. 1 and 5 and some were found in the general filter effluent.

Some observations were made which throw light upon the reasons for this distribution. Absence of these worms in the Septic Tanks is, no doubt, due chiefly to the absence of dissolved oxygen and the presence of inimical gaseous decomposition products. Absence from the Settling Tank is due probably to the very low dissolved oxygen content. Absence in Sprinkling Filter no.2 can be accounted for by the fact that the influent of this tank comes directly from the Septic Tank and the same unfavorable factors are introduced: Constant absence in Sprinkling Filter no. 3 finds an easy explanation in the fact that its influent comes directly from the modified Dortmund Tank where considerable subphate reduction occurs and as a consequence the effluent of the latter (the influent of the former) is laden with hydrogen sulphide which is harmful to animal life, not only because it draws upon the available oxygen but it is in itself poisonous in its effects. The appearance of the worms in Filters 1,4, and 5 is presumably due to the fact that the influents of the above named filters are effluents of the settling tank and are "fresh". The great abundance in Filter no. 4 as compared with the scarcity in Filters

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no. 1 and 5, in October, seemed to depend upon the fact that Filter no. 4 contained an unusual amount of sludge for the time of year while the quantity of sediment on the stones in Filters no. 1 and 5 was decidedly smaller. The reason for the appearance of the worms in the filter effluents is a purely mechanical one since they are carried there by the descending currents of sewage.

The vertical distribution of the worms in the filters during the summer is not known but an examination of Filter no. 4 in Ootober showed them to be largely confined to the upper two feet of the filter bed. Below that limit they occurred only rarely. This vertical distribution coincided with the vertical distribution of the "load" of the filter since the greater part of the sludge deposited at this time of year is confined to this zone and below it the stones are conspicuously cleaner. It seems very probable that this is the factor which determines the distribution of the worms at this time since they show a decided affinity for the sludge.

These worms do not appear normally on the upper surface of the filter bed but may be found by removing the uppermost rock and examining the ones which lie below. The physical conditions of this environment are as follows:- the light is pr\_actically excluded beyond the first six to eight inches. Moisture is at a maximum since large quantities of sewage are constantly flowing down through the interstices. The temperature is cool and in general fairly uniform for a given season. As indicated by the effluent, the temperature has a variation of about 10° C in winter and is always above 0°C. The average temperature in September, 1911 was 68°F. and in October 63°F. An abundance of the finer

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settling suspended matter finds lodgment on the surfaces of the rock and in the interstices.

Adults are distributed to the various parts of the plant by the streams of sewage. They have a specific gravity slightly greater than water and therefore sink slowly when placed in quiet water of some depth but a stream of moderate velocity will carry them when they are once loosened from their hold on the supporting rocks. The writer has observed instances where several worms had penetrated a mass of the sludge which because of decomposition changes had acquired a low specific gravity and when loosened from the point of accumulation it floated away easily carrying the additional weight of the worms.

The associated animal life.

These Lumbricillid worms were not the only animal forms present in the filters. In fact, when examined in October, it was found that there were other forms which were more generally distributed and more abundant. No attempt was made to list the microscopic life of the filters and attention was confined to the macroscopic forms of which the following were more or less abundant in October, 1912:-

- 1. Prorhynchus sp ?.
- 2. Small Nematodes(not identified).
- 3. Pristina sp.
- 4. Nais communis Piguet(?).
- 5. Helodrilus subrubicundus Eisen.
- 6. Psychoda albimaculata Welch.
- 7. Chironomidae.
- 8. Collembola (Isotoma sp.)
- 9. Water Mites(not identified).

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3. Color of the living form.

To the unaided eye this worm presents a general reddish appearance. The region posterior to the olitellum appears uniformly reddish except that the ventral blood trunk shows as a deeper red line. The region anterior to the posterior margin of the clitellum is distinctly lighter in color and the region of X-XII is sexually mature specimens shows as a distinct white spot with no trace of red. The surface layer of the lime stone rocks composing the sprinkling filter beds acquires after a time a reddish brown color due partly to a formation of certain ferrous compounds and with this sort of a back-ground the worms are often a little difficult to distinguish but the white spot on X-XII is conspiouous and reveals their presence. When large numbers are placed in water they often tend to accumulate into a compact mass and in such an aggregation the red color seems to be intensified as the whole mass gives one the impression of a deeper red than occurs in a single isolated worm.

Examination under magnification shows that the body is somewhat transparent, with a light yellowish tinge and that the red color is really due to to the red blood in the vascular system. The translucency of the body wall permits the vascular system to show through boldly enough to effect the external appearance. The principal vessels and their connecting branches can be easily traced and the pulsations of the dorsal vessel watched with ease. The white spot in the region of X-XII owes its existence to the presence of developing reproductive elements and constitutes an external means of judging the sexual maturity of the specimens.

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4. Locomotion.

The sole mode of locomotion is an active crawling against a supporting surface. A number of experiments were performed with the view of determining the efficiency of this mode of locomotion. Specimens were tested with the following substances as supporting surfaces:-

- 1. Filter rock.
- 2. Sludge.
- 3. Wood(planed surface).
- 4. Ground glass.
- 5. Smooth sheet iron.
- 6. Smooth glass.

Each of the above surfaces were tested in two ways:-(1) dry, and (2) moist. The results showed that the efficiency of the crawling was very much higher on all of the surfaces when they were moist and that dry surfaces were an important barrier against dispersal. As regards the moist surfaces of the above named substances, the tests showed that the efficiency varied approximately in the order indicated in the list, the maximum occurring on the filter rock and the minimum on the smooth glass. The worms crawl with considerable ease over the surfaces of the rocks in the filter beds and owing to the irregular shapes of these rocks the interstices form a continuous series of chambers and passages abundantly supplied with sewage and settled solids, through which the worms pass easily. They also have a surprising ability for working their way through the masses of sludge which accumulate on the surface of the stone. On a moist ground glass surface the worms make moderately efficient progress but on moist polished glass the progression is slow. The difference in the rate of locomotion on the various supporting surfaces is due to the fact that the setae, which are important agents in locomotion, easily

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or with difficulty find temporary hold on the irregularities of the surface.

The crawling consists of an extension of the anterior region and a drawing of the posterior region after it. Although this procedure is the normal and usual one it is, under some circumstances, reversed and the result is a temporary backward movement.

There is no evidence whatever of an ability to progress by swimming. A series of tests were made by placing worms in test tubes full of sewage. In all cases they sank to the bottom and were never able to leave it. If placed in a dish of water whose depth was less than the length of the worms they could often get to the surface by crawling up the side. If the water in the vessel was deeper than the length of the worms it was impossible for them to get away from the bottom. When placed in water they often exhibit random wriggling movements but such motions were ineffectual so far as locomotion was concerned.

5. Relation to external factors.

a. Relation to light.

The specimens of this species has a decided negative response to light. They occur normally below the surface of the filter bed where practically all light is excluded. No worms were found on the well lighted surface of the filter bed. In the Spring and early Summer when the maximum abundance accurs the worms often make their appearance in consider -able quantity in the effluents of the sprinkling filters and regularly accumulate on the shaded side of the secondary settling basins. Filter bed rock was frequently brought into the laboratory in large battery jars and placed on a table before a large window. The worms regularly

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-/ migrated away from the light side towards the dark side and then if undisturbed migrated into the central region of the mass of rook. Sudden exposure to light calls forth a response in the form of active orawling movements which cease when the organisms find themselves in a place where the light is distinctly less in intensity. Exposure to direct sunlight produces an immediate, active negative response.

## b. Resistance to desiccation.

It was noticed that when masses of filter rock which contained worms were brought into the laboratory and exposed to the air sc that evaporation could take place the gradual drying of the surfaces at successive depths caused a migraticn of the worms deeper and deeper into the mass where moisture was still present. A number of tests were made to determine how long these worms could live when removed from water or moist sludge and transferred tc a dry place. The specimens were cleaned and tested singly. There was some variation but in general the time was limited to 3-5 minutes when the experiments were carried on under the conditions which existed at the Testing Laboratory. Beyond this time the worms failed to revive when returned to moist conditions. A mass of worms composed of several individuals had a much higher resistance than a single individual.

## o. Thigmotactio response.

These Lumbrioillid worms exhibit a considerable degree of positive thigmotaxis. This is shown by the frequency with which numbers of them were seen under natural conditions to progress in aggregations side by side. They are also often found grouped together in masses on the surface

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of the filter rocks. When a large number of specimens were transferred to a watch glass containing tap water they showed a distinct tendency to assemble into a mass around any solid particle or small mass of sludge which may have been transferred with them. This often proved to be of advantage in the putrescibility tests since the transfers from the different containers could be easily made by pricking up the whole mass of one hundred worms at one time. The tendency to accumulate into masses seems to be accentuated when the worms are placed under slightly unusual conditions. In case these masses are left undisturbed for a time the worms often ultimately begin to disperse in a rather characteristic way in that they start to move away on the supporting surface in several separate aggregations, the individuals of each lying compactly side by side and all moving in the same direction.

Spesimens were frequently placed in temporary storage in Syracuse watch glasses each containing a small quantity of water and covered by another watch glass. After these glasses had stood for a time the worms almost invariably accumulated around the upper rim of the lower watch glass at the point where the upper glass touched the lower. They apparently prefer a position where they secure the maximum contact with the glass. Specimens kept in Petri dishes commonly accumulated between the perpendicular sides of the upper and lower dish. Specimens kept in dishes containing wet filter paper commonly sought positions between the filter paper and the sides and bottom of the container.

A normal habit of the worms involves a crawling about over the surface of the rook to which they cling rather tenaciously, often making it difficult to pick them off with a needle and a stream of water must have at least a moderate velocity in order to wash them off. They

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normally bore into the sludge and are also often found in the pores of the rock from which they are extracted with considerable difficulty.

d. Behavior with reference to dry and moist surfaces.

A series of experiments was performed in order to determine the mode of behavior with reference to dry and moist surfaces. These experiments were carried on in the laboratory in diffuse sunlight and

in a temperature of about 75°F.

Experiment 1.

A drop culture slide with concave center and ground glass top surface was used and the concavity filled with tap water. By means of a large needle a straight continuous trail of water was drawn from the concavity to very near the end of the slide. Worms were placed at the

end of the water trail. Results: -

- Random movements in the immediate vicinity with the anterior half of the body. These movements showed the following uniform characteristics:
  - a. Refusal to pass out onto the dry surface.
  - b. Refusal to pass off the end of the slide.
  - c. An apparent recognition of the edge of the water followed by an immediate response resulting in the withdrawal of the anterior region.
  - d. Exploring movements at the end of the water trail restricted to a small area.
  - 2. The worms ultimately found the water trail and almost invariably followed it rapidly and uninterruptedly to the central cell.

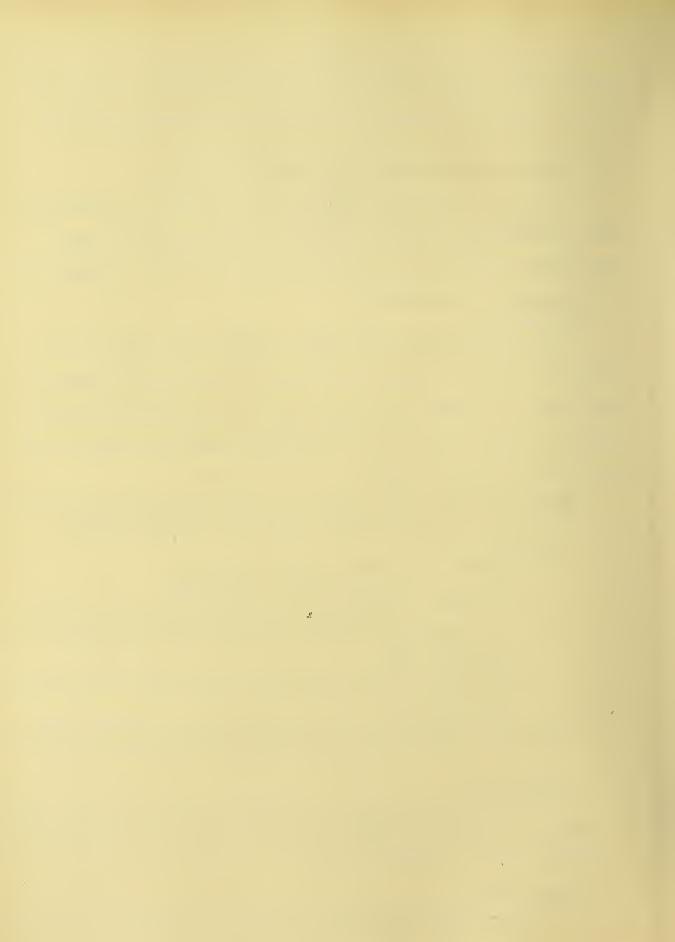
Experiment 2.

In this experiment the water trail was made very tortuous and and the procedure and conditions were the same as in Experiment 1. Results same as in Experiment 1.

Experiment 3.

A ground glass slide which did not have a central cell was used.

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A long straight water trail was drawn on the surface and worms were placed at one end. Results: - Aside from occasional stops accompanied by searching movements the worms followed the water trail to the opposite end; retraced the course, and often repeated this performance for considerable length of time.

## e. Relation to temperature.

The abundance of material made it possible to carry on a long series of experiments involving large numbers of individuals with the view of determining the temperature limits of life and the effect of different temperatures on the general activities of the organisms. In all of the experiments vigorous worms just removed from the natural habitat were used. All of the tests were made using tap water as the medium. Beginning at 2°C tests were made for every degree(in some cases every 1/2°) up to 45°C. This was also done by starting at 45° and reducing the temperature by steps of 4° down to 2°C. Unfortunately facilities were not at hand for making tests with temperatures lower than 2°C and consequently the minimum temperature was not determined. Results indicated that the maximum temperature is very near 36°C. Specimens sometimes survived 36°C. but an additional 1/2° proved fatal in a short time. All temperatures above 38.5°C caused immediate death. The specimens submitted to the higher temperatures were observed under the microscope so that the effect of the temperatures could be judged more accurately. All specimens survived 2°C., the lowest obtainable temperature at that time, and were apparently uninjured. It is interesting to note that these worms seem to be adapted to withstand successfully rather low temperature.

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A series of tests was carried on by putting a considerable number of worms into flasks containing water to a depth of about 3 cm. and keeping them in the ice box where the temperature was a trifle less than 5°C. The worms lived indefinitely under these conditions. One flask remained in the refüßgerator for ten days and at the end of that time all of the worms were alive, active and apparently in as good condition as when they were first put in. As to the effect on the activities of the worms, little if any difference could be detected from 10° to 25°C. However below 10°C, the activities decreased so that at 2°C only a very moderate activity was manifest. It was noted that this decrease was not uniform but from 5° to 2° the decrease was more marked than from 10° to 5°. Above 25° the activity became increased as the temperature was raised.

From this data it would appear that so far as temperature is concerned ed the worms could exist in the filters the year around since the latter are in constant operation and they never freeze. Evidently some factor other than temperature is effective in the reduction of the numbers of the worms in late Summer and Autumn.

#### f. Relation to oxygen.

1). The normal supply of oxygen.

As has been stated before a sprinkling filter is a device designed primarily to effect the oxidation of the sewage which is delivered to it and as a consequence the organisms which live in the filters are well supplied with oxygen. This element comes to them in two ways, (1) from the air by direct contact, and (2) from the sewage which contains dissolved oxygen.

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In all of the filters except no. 3 the stone composing the filter bed is of a size which produces a series of continuous and rather spacious interstices. These may be reduced to some extent by the settling material from the sewage but never become completely filled. These interstices constitute a series of air spaces and thus form a source of oxygen supply for the organisms inhabiting then. That these Lumbricillid worms take oxygen directly from the air is shown by the fact that specimens have been kept for several days in vessels containing moist filter paper which served to prevent the undue drying of the worms. This was also demonstrated when at one time an accident to the pumping machinery eliminated the possibility of oxygen from sewage for over three days the only liquid present in the upper zone being that held by the eludge.

The sewage which constitutes the influent of the filters, coming as it does from the Septic and Settling Tanks has a very low dissolved oxygen content, often showing a total absence of dissolved oxygen partioularly during the hot season. However this influent passes through a nozzle which preaks the stream up into a spray. This spray is thrown out into the air and in falling is distributed over the whole of the upper surface of the filter bed. The result of spraying the liquid into the air and passing it through the air spaces between the rocks is that the liquid becomments oxygenated to considerable degree. The spraying also brings about the loss of most of the carbon dioxide. Consequently regardless of its source the sewage which flows over the worms has a considerable dissolved oxygen content. The effectiveness of the sprinkling filter as an oxygenating device is shown by the fact that filter no. 4 on certain days in September and October, 1912, when the influent(at entrance to nozzle) showed an absence of dissolved oxygen the effluent contained

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from 11.2 to 14.7 p.p.m. That this oxygen can be utilized by the worms is shown by the fact when placed in tap water they lived for days and even weeks under conditions which prevented their getting the oxygen from any other source.

2). Resistance to decreased oxygen.

It would seem that organisms as active as these worms would require a considerable amount of free oxygen and that the sprinkling filter environment is such as to afford a generous supply. A number of extended experiments were made with the view of determining the effect of decreased oxygen. The methods employed in these experiments are as follows:-A series of samples of tap water whose dissolved oxygen content ranged from 1.2 p.p.m. to 9.2 p.p.m. was used as a medium. The highest dissolved oxygen sample was obtained by agitating the water in air. Tests were also run using sprinkling filter effluent in which the dissolved oxygen varied from 10 to 13 p.p.m. Samples with a lower dissolved oxygen content were made up by mixing boiled tap water with different quantities of tap water; in still other cases they were made by boiling a considerable quantity of tap water and allowing it to stand in an open vessel and taking samples at successive intervals as the water gradually absorbed oxygen from the air. Sampling bottles of 128 cc. corrected capacity were used and care was taken that each was stoppered in such a way that no air was included in the form of bubbles at the region of the stopper. Two samples were taken of each of the different grades of water, one being used for the worms and the other for the determination of the dissplved oxygen content. Ten vigorous worms, fresh from the filter, were quickly transferred to one of the bottles and the time noted. The

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worms were thoroughtly cleaned before they were put into the bottles. This procedure was followed for each of the samples taken. The bottles were all kept under conditions which simulated those existing in the sprinkling filter and were observed frequently.

It might appear that there is a source of error in this work due to the fact that no account was taken of the carbon dioxide and the nitrogen. When water is boiled it loses most of the dissolved oxygen, nitrogen and carbon dioxide and when oxygen was added to the samples by exposure to the atmosphere or by the addition of ordinary tap water certain small quantities of nitrogen and carbon dioxide were also added. However the writer believes that this source of error did not interfere with the essential results of the experiments. The carbon dioxide in the sprinkling filter is a very variable quantity since much of the gas present in the influent must be lost when it is sprayed out into the air and consequently the water coming to the worms probably contains only small quantities of the gas, a condition comparable to that in the sampling bottles of the experiments. Only a very limited amount of dissolved nitrogen is present in the sprinkling filter influent. When tap water which contains a certain amount of N is boiled the major part of the N is lost. In those samples made up by mixing the boiled with the tap water some N was added with the latter while in the other samales made by exposure the N is taken up from the air very slowly so that in all cases the samples closely approximated the conditions in the sprinkling filters. Of the three gases involved the oxygen was the only one which was varied widely in the experiments.

Other workers have found that it is very difficult, in fact, almost impossible to vary absolutely only a single gas in this kind of

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experimentation by methods known at present and a small source of error must of necessity be introduced. Since the aim of these experiments is to determine the general effect of reduced dissolved oxygen content, it is thought that this source of error does not materially effect the general result.

The dissolved oxygen content of each sample was determined by using Winkler's Method as outlined in "Standard Methods of Water Analysis 1912, published by the American Public Health Association. Briefly stated, a solution of manganous sulphate was added to the sample and followed by an alkaline solution of potassium iodide. The precipitate of manganous hydrate was allowed to settle. Sulphuric acid was added and the free iodine in solution was titrated with a standardized solution of sodium thiosulphate. The equivalent of free iodine was calculated to oxygen and the results expressed in parts per million.

The following are the results of one of the experiments which will serve as a representative of other similar experiments made in the same connection:-

|             |                        | Exportano.         |                        |                       |                 |                        |   |
|-------------|------------------------|--------------------|------------------------|-----------------------|-----------------|------------------------|---|
|             | Corrected<br>of bottle | capacity<br>in cc. | $Na_2S_2O_3$<br>in cc. | Oxygen<br>in p. p. m. | Number<br>worms | Time lived<br>in hours | _ |
| ÷÷-÷-<br>1. | 128                    |                    | 0.8                    | 1.25                  | 10              | 21                     |   |
| 2.          | 128                    |                    | 1.1                    | 1.7                   | 10              | 27                     |   |
| з.          | 128                    |                    | 1.7                    | 2.6                   | 10              | 42                     |   |
| 4.          | 128                    |                    | 1.9                    | 2.9                   | 10              | 47                     |   |
| 5.          | 128                    |                    | 3.5                    | 5.4                   | 10              | 65                     |   |
| 6.          | 128                    |                    | 5.7                    | 8.9                   | 10              | 75                     |   |
| 7.          | 128                    |                    | 5.99                   | 9.2                   | 10              | 142                    |   |
|             |                        |                    |                        |                       |                 |                        |   |

Experiment III.

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The data of the other experiments carried on in this connection varied to some extent from the above but the general result is the same, namely, that these worms require oxygen, that they must have it in considerable quantities, and that a continued low dissolved oxygen content has a deleterious effect. In interpreting these results it is necessary to keep in mind the fact that in each of the samples the dissolved oxygen content does not remain constant but the water becomes gradually poorer in oxygen since it is being used up in the respiratory activity of the worms and it is very probable that the excretory products of the worms aid in reducing the dissolved oxygen to a small extent. Although the data seem to show that the worms are sensitive to reduced oxygen and that a lowering of the dissolved oxygen content is inimical, it does not necessarily follow that death was due to complete exhaustion of oxygen since it was shown that death occurred sometimes when considerable oxygen was still unconsured, as for esample, in number 6 of the above table the water which at first contained 8.9 p.p.m. was tested for dissolved oxygen at the end of the experiment and was found to still contain 4.2 p.p.m. of Koygen, a quantity which was sufficient, as an initial quantity, in ong of the other experiments to allow worms to live for 98 hours. It was not possible in these experiments to remove the excretory products from the water and thereby eliminate the possibility of the accumulating waste influencing the vital activities of the worms but in an experiment made in another connection a similar quantity of worms was kept in 45 cc. of water in an open flask to which the air had free access and at the end of ten days when the experiment was ended by an accident all were alive and apparently as active as at the beginning. Since the quantity of water was so much smaller and the length of time

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so much greater than in the above experiment it shows fairly conclusively that accumulating wastes do not constitute a factor which brings about that death of the worms. Furthermore the normal habitat is one in which organic wastes are at a maximum.

g. Relation to sewage.

1). To crude sewage.

In another connection the fact was brought out that no worms were nore found in the Septic and Settling Tanks which receive the orude sewage and the cause was attributed to the lack of dissolved oxygen. Tests were made in which vigorous worms were placed in bottles of crude sewage and corked so that no air was included or could gain access. In all cases they lived but a short time(10-12 hours). Another test was made by placing worms in a flask which was about half filled with crude sewage, thus leaving a large air space atove the surface. One lot lived under these conditions for 72 hours which is to be accounted for by the fact that the sewage absorbed oxygen from the air space above. However since the sewage itself contained putrescible matter which also drew heavily upon the oxygen supply, the continued low oxygen content had a fatal effect.

2). Behavior in the presence of sludge.

The following experiments were performed in this connection:-Experiment 1.

A ground glass slide was used and a water trail was drawn lengthwise of the slide. A mass of moist sludge from the filter stones was placed at one end of the water trail and worms were placed at the other

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end. Results: - the worms followed the trail to the sludge mass, crawled around it and through it and became quiet. Did not leave it.

Experiment 2.

Supported a circular cover glass on bits of filter paper as feet, ran water under it and drew a water trail leading to it. Ten worms were placed at the end of the trail. Results: - the worms followed the trail to the cover glass but would not pass under it.

Experiment 3.

The procedure was the same as in experiment 2 except that the cover glass was covered with black paper so that the space under it was darker than that outside. Results:- same as in experiment number 2.

Experiment 4.

The procedure was the same as in experiment 2 except that a mass of sludge from the filter was placed under the cover glass. Results: - in every case the worms passed under the cover glass in a short time and became quiet after surrounding and penetrating the mass.

It is not possible to explain, from the data, the behavior of the worms with reference to the cover glass in experiments 2 and 3. In neither case did the worms pass under and in spite of the fact that these forms are negatively phototecic, the area under the darkened cover glass was avoided. There was evidently some unknown factor present which was sufficiently active to overcome the negative phototactic tendency of the worms and prevent their migration into more favorable light conditions. However the experiments show a distinct regognition by the worms of the presence of sludge and a positive reaction. Furthermore it is evident that this positive reaction is sufficiently strong to overcome the opposing influence which prevented the worms from migrating under the cover glass in experiments 2 and 3. The experiments

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showed also that the positive reaction to the sludge was not due to the negative phototaxis since the worms did not pass under the cover glass in experiment 8.

## 3] Relation to the sludge in the filters.

That these worms have ammechanical effect on the settling suspended matter which accumulates in the filters it not difficult to (b) serve. These organisms are active and are constantly burrowing through the masses of sludge and since they occur in such large numbers they must play a prominent part in loosening up the sludge and working it over, thus facilitating the oxidation of the unstable organic matter.

It has been observed at the Testing Station that during the winter the sprinkling filters become clogged to considerable extent, that is, they build their "load", by accumulating a large quantity of sludge. This sludge is held in the filter until the temperature begins to rise in early Spring and then the filter begins to "unload". The effluent becomes laden with large quantities of heavy earthy matter and the sludge in the filter becomes rapidly reduced. The significant thing is the fact that this "unloading period" is coincident with the maximum abundance of the Lumbricillid worms and after the greater part of the unloading has occurred the abundance of the worms decreases somewhat. Whether or not the worms are responsible for the unloading remains to be proven but the circumstantial evidence points to these forms as at least one of the factors which play a part in the unloading. Cognisance must be taken of the fact that other organisms are also abundant at this period and it is possible that the unloading is the result of the combined mechanical action of a number of associated organisms.

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4). Relation to Putrescibility.

Before going into the discussion of the experiments which were made in this connection it will be necessary to make clean certain important terms which are in constant use in sewage investigations.

In order to explain what is meant by the terr Putrescibility and to indicate its precise application in sewage disposal work it is necessary to explain in considerable detail certain chemical and physical conditions which exist in ordinary sewage. Phelps('09, p. 75) gives a very clear account of the application of this term in the following rather lengthy "Putrescibility as applied to organic matter in general, quotation: implies the ability to undergo offensice putrefactive decomposition" \*\* \*\*\*" Such decomposition is always anaerobic and is accompanied by the evolution of offensive odors. These two phenomena have, therefore, formed the basis of most putrescibility tests. Some criteria of putrefaction which have been employed are = (1) Development of offensive odors; (2) formation of black sediment; (3) reduction in the amount of dissolved oxygen; (4) loss of all dissolved oxygen; (5) loss of all available oxygen including that of nitrites and nitrates; and (6) increase in the oxygen-consumed figure. Some of the tests are based on partial reduction of the available oxygen and subsequent anaerobic fermentation in the effluent; others depend on the complete reduction of the available oxygen and the subsequent anaerobic fermentation. The tests most commonly employed belong to the latter group, depending on the production of odor or of hydrogen sulphide, blackening of the liquid, or reduction of organic dyes. The test which depends on an increase in the oxygen-consumed figure during incubation is also in this class,

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because anaerchic fermentation alone renders organic matter more readily oxidizable."

"These two types of test illustrate two distinct points of view which should be clearly differentiated. An effluent may be regarded as being composed of a given mass of organic matter diss clved or suspended in a definite amount of water. The water contains also a definite amount of available oxygen in the form of free dissolved oxygen, nitrites, nitrates, and possibly other compounds. All of the organic matter is oxidizable to some extent, and to that extent it serves as bacterial food. The greater the [maount of organic matter and the greater its oxidizability, the greater is the absorption of oxygen from the medium. Consequently a reduction of available oxygen in the effluent during inoubation is a measure both of the amount of organic matter present and of its capability of oxidation. As a small amount of readily oxidizable matter has the same effect on the result as a larger amount of more stable matter, a test of this kind indicates whether or not the organic matter consumes oxygen; but it does not show whether or not the supply of available oxygen is sufficient to prevent the establishment of anaerobic conditions. This important question of the balance between the oxygen demanded by the organic matter and the oxygen available in the liquid is taken into consideration by tests of the second kind; namely, those dependent on the establishment of anaerobic conditions. Such tests do not involve the estimation of the amount and the kind of organic matter; indeed, organic matter which does not absorb any oxygen from the liquid under the conditions of an incubation test must be very highly oxidized; and, furthermore, most organic matter derived from sewage is putrescible in itself - that is, if it is stored by itself in the absence

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of oxygen, it undergoes putrefactive changes. The question at issue is not however, whether the organic matter itself will putrefy, but whether the effluent as a whole will become so reduced in oxygen that putrefaotion will be possible. In other words, it is: simply a question of a balance between the available oxygen of the effluent and the oxygen which the organic matter will require during the incubation period. It would seem that the problem might readily be solved by determining this balance, but; unfortunately, it is not a simple matter, because the action involved is bacterial. Many attempts have been made to determine the oxygen balance analytically, but such tests answer only with very good and very bad effluents, for which an inspection of the sample would serve just as well. When there is doubt about the character of the effluent - the condition for which such information is of most value - all such analytical procedures have peretofore failed. It is evidently impossible to imitate with any degree of precision the bacterial activities that are involved. There remains, then, but one satisfactory expedient: To let the action proceed by itself and note the result. But here also there are difficulties, because bacterial reactions of this sort are necessarily slow in reaching equilibrium, and the time required by a nicely balanced effluent is greater than can be allowed in routine work. Some arbitrary perior of time, therefore, is usually adopted, and it is in respect to this factor that the confusion arises. If stability is to be considered as a definite qualitative characteristic of an effluent that characteristic should be determined by a test sufficiently prolonged to insure equilibrium, but such procedure is not feasible for obvious practical reasons, and it is not desirable, because it is not enough simply to know that the available oxygen is sufficient or insufficient to

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to satisfy the demands of the bacteria that are working on the organic matter. If the available oxygen is sufficient, there is gerfect stability -a definite condition; if it is insufficient, there is still stability in the quantitative sense - a relative stability determined by the relation of the available oxygen to the total amount of xoygen required by the organic matter for perfect stability. In practice the latter condition is the one usually encountered".

Owing to the varied meanings which are attached to the word putrescibifity, Phelps(l.c. p.77) has recommended the word Stability to designate" that desirable quality which is the usual object of sewage purification - the transformation of organic matter to such a form that it is incapable of undergoing offensive decomposition". He argues that the term stability implies a positive characteristic which is acquired

during the purification process, while the term putrescibility refers to a negative characteristic. Stability describes that condition in which the available oxygen exceeds the required oxygen. However the term putrescibility has been retained in this paper since it is still largely in use in the literature which deals with sewage investigation.

From the economic point of view questions bearing directly upon the putrescibility or stability of sewage are of the greatest importance. The ultimate aim of all sewage disposal operations is to render the putrescible matter as stable as possible and any factors which facilitate or hinder this process are of considerable practical importance. Since these Lumbricallid worms occur in such abundance in connection with devices which are operated in order to increase stability, the relation of these worms to this process becomes pertinent. From the point of view

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of sewage disposal it is desirable to determine whether or not these forms have any relation to putrescibility and if so whether it is of a beneficial or detrimental nature.

The purpose of these experiments was to discover if possible just what effect these organisms have on putrescildility. Tests were made on the following kinds of sewage: - (1) Raw sewage, (2) Septic Tank effluent, (3) Settling Tank effluent, and (4) Sprinkling filter effluent. The suspended matter is sewage is of two kinds, namely, the settling suspended matter, and the non settling suspended matter. The former is of such a nature that it can be removed by filtration through ordinary filter paper, or will be deposited when sewage is stored. The latter does not settle and is not removed by ordinary filtration but must be removed by chemical precipitation, by biologic treatment, or by the use of certain special filtering devices. This non settling suspended matter is colloidal in nature and is known as pseudo-colloidal matter in sewage. Each of the above mentioned effluents was tested in three ways, (1) by using the raw material, (2) by using the material after passage through ordinary filter paper which removed the settling suspended matter, and (3) by using the material from which the pseudocolloidal matter had been removed. This pseudocolloidal matter was removed by filtering the liquid through a Gooch crucible which was connected with a filter pump. Since investigation has shown (Lederer, 1912, p.5)" that the finaly divided slowly settling matter and the pseudocolloidal matter not capable of settling make up the greater part of the putrescibility" the tests were made in such a way as to allow the study of the effect of the worms on the liquid when one or both of the above mentioned substances were

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present.

The sampling bottles of the Testing Station were used as containers for the tests. These bottles have a capacity of 128 cc. All of the glass ware such as pipettes, sampling bottles and etc. was sterilized before using. The sampling bottles were filled with the various grades of sewage and then worms were transferred to each. Vigorous worms, fresh from the filters were used in every case. They were carefully cleaned by transferring them through a series of vessels containing pure water before they were transferred to the test bottle in order that no additional matter might be carried with them. Then they were counted out into lots of 100 and after the removal of all excess water each lot was weighed on a fine analytical balance. Those lots which weighed approximate -ly the same were selected for the tests. Each lot was placed in a separate boille which was corked in such a way that no bubbles were enclosed. For each individual test a check experiment was carried on similar in all respects except that no worms were used. Thus a single series involved twenty four tests.

Determinations of putrescibility involve the use of delicate indicators which aid in the accurate detection of the beginning of anaerobic conditions. In these experiments Spitta and Weldert's Methylene Blue Putrescibility Test was used. This test depends upon the formation of a colorless leucobase as the oxygen in the sample becomes exhausted. The technique is simple. One cc. of 0.1 percent aqueous solution of Methylene Blue is added to the sample which is then kept in an incubator at either 20°C or at 37°C, and observed frequently. The blue color of the sample remains practically unchanged until the available oxygen contained in it has been consumed and putrefactive conditions have been established. At

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this time the dys is reduced and the color disappears. The time for the decolorization(reduction time) therefore indicates quite closely the time at which the available oxygen is consumed. Phelps('09,p.77) added further value to the Methylene Blue Test by putting it on a quantitative working basis so that the putrescibility of a given sample can be expressed in terms or relative stability. This makes it possible to indicate the proportion of oxygen present as compared with the total amount of oxygen required to oxidize a given sample.

This test lends itself well to this kind of experimentation since it not only makes it an easy matter to determine the reduction time but the presence of the Methylene Blue in the sample has: little or no deleterious effect on the worms. At the same time that the worms were transferred to the sample 1 co. of a 0.1% aqueous solution of Methylene Blue was added to each bottle and the time carefully noted. These test bottles together with the ohecks were placed in a constant bemperature incubator at 20°C. Careful watch was kept on these samples and the reduction time of each noted.

The worms in the sampling bottles were frequently observed in order to determine whether or not any of them died while under these conditions since it is evident that the death of any of them would constitute a source of error by increasing quantitatively the amount of putrescible matter in the sample. Fortunately the mortality was very low, so low that the writer feels confident that it did no vitiate the results.

The following table indicates the results of one of the series:-

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|---|----------------------------|-----------------------|-------------------------------------|---|--|--|--|--|--|
| Origin.   | Putrescitility<br>in hours | Relative<br>Stability | Loss<br>in<br>Relative<br>Statility | Percentage<br>relation of<br>actual lose to<br>possible loss. |  |  |  |  |  |
| Crude sewage. (blank)   |                            | 11                    | 1                                   | 9.09  |  |  |  |  |  |
| Crude sewage(100 worms<br>Crude sewage. Filtered<br>+ pseudocolloids<br>(tlank) |                            | 28                    |                                     |   |  |  |  |  |  |
| (100 worms)   | 24                         | 21                    | 0                                   | 19.2  |  |  |  |  |  |
| Crude sewage. Filtere<br>- pseudocolloids*<br>(blank)                           |                            | 47                    |                                     |   |  |  |  |  |  |
| •••• (100 worms)  | 29                         | 24                    | 28                                  | 48.9  |  |  |  |  |  |
| Septic tank effluent.<br>(tank A)(clank   | 8                          | 8                     |                                     |   |  |  |  |  |  |
| ·· (100 worts)  | ٤                          | 2                     | 1                                   | 38.8  |  |  |  |  |  |
| Septic tank effluent.<br>Filtered + pseudo-<br>colloids.(tlank)                 | 17                         | 15                    |                                     |   |  |  |  |  |  |
| ··· (100 worss)   | 14                         | 12                    |                                     | 20.   |  |  |  |  |  |
| Septic tank effluent.<br>Filtered - pseudo-<br>colloids(tlank)                  | 50                         | 27                    |                                     |   |  |  |  |  |  |
| · (100 worns)   | 29                         | 24                    | 31                                  | 95, 1   |  |  |  |  |  |
| Settling tank efflue<br>Crude. (blank)  | nt. 14                     | 12                    |                                     |   |  |  |  |  |  |
| ·· (100 morns)  | 12                         | 11                    | 1                                   | 8.2   |  |  |  |  |  |
|   |                            |                       |                                     |   |  |  |  |  |  |

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In relation of Selative actual loss Statility Statility to possible Lelative. Putrescitility Origin in hours \_\_\_\_ Settling tank effluent. Filtered+pseudocolloids. (tlank) 20 25 • • (100 worms) 2 10.7 25 31 \_\_\_\_\_ Settling tank effluent. Filtered-pseudocolloids. 37 õõ (tlank) .. .. . . 22 15 32.4 27 (100 worns) \_\_\_\_\_ Sprinkling tilter effluent. Tank 2. (tlank) 99 20 JAVS 69.6 89 (100 woris) 20 . . 27 hrs. Sprinkling filter effluent. Filtered+pseudocolloids. (tlank) 58 20 days. 65.6 35 34 . . . (100 worms) 42 hrs. Sprinkling filter effluent. Filtered-pseudocolloids. (Elank) 20 days. 65.6 05 (100 worms) 34 42 hrs. Incubation temperature 20° C. Relative statility numbers calculated according to Phelps. \* For the sake of brevity the signs + and - have been used to indicate the presence or absence of the substance whose name follows the sign.

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An examination of all of the results of the experiments made in this connection of which the above series is a part shows that the most conspicucus result is the marked <u>increase</u> of putrescibility in the samples containing the worms as compared with the corresponding samples which contained no worms. This is a constant feature of these experiments as in no case was there an opposite result. The presence of the worms increases the putrescibility under all conditions as regards the presence or absence of the various kinds of suspended matters in sewage. The reduction time increases with the removal of suspended matter and the difference between the reduction time of the test and that of the check experiments tends to become greater as the suspended matters are removed. Increased putrescibility means loss in stability and loss in relative stability apparently increases with the removal of the suspended matters.

The explanation of the manner in which this increase in putrescibility is effected by the worms has not been determined. The exhaustion of the oxygen may be accomplished in two ways, (1) by the respiratory activity of the worms, or (2) by means of the organic matter contributed by the worms in the form of excreta. It is very probable that both are factors which work towards the same end. From a practical stand point it does not matter how the oxygen is used up since the important thing is the fact that it is <u>being used up</u>. As has been stated before the sprinkling filter is a device for oxygenating the sewage which is delivered to it thus rendering it more stable. The presence of anything in the filter. The evidence seems to be conclusive that the presence of these worms in the rilter <u>increases</u> the putrescibility of the sewage and this is accomplished by using up a part

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of the available oxygen and since these worms occur in great numbers in the filter for the greater part of the year there is good reason for thinking that so far as their relation to the available oxygen is concerned the effect of the presence of these worms in the filter is a <u>detrimental</u> one. If, on the other hand, it be true that at all times of the year there is a distinct vertical distribution of these worms in the filter in which the larger number are confined to the upper two or three feet the detrimental effects of their presence may be overcome to some extent since the interstices of the filter throughout its depth are air spaces and it is possible that the loss of the oxygen in the upper zone due to the activities of the worms may be made up to some extent by the passage through the air filled spaces. Nevertheless the fact remains that the worms increase the putrescibility and in this connection are apparently not desirable organisms.

It is possible that when the problem of the relation of these worms to sewage has been completely worked out it will be found that advantageous relations may more than offset the harmful ones so that the sum total of their activities may show them to be desirable forms. However until more investigation is carried on this point must remain unsettled.

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#### V. SUMMARY.

1. The following new species of Enchytraeidae distributed among four genera have been added to the list of North American forms:-

| Name                     | Type Locality         |
|--------------------------|-----------------------|
| Henlea moderata          | Urbana, Ill.          |
| Henlea urbanensis        | Urbana, Ill.          |
| Lumbricillus rutilus     | Chicago, Ill.         |
| Fridericia douglasensis  | Douglas Lake, Mich.   |
| Fridericia oconeensis    | Oconee, Ill.          |
| Fridericia sima          | Urbana, Ill.          |
| Enchytraeus gillettensis | Gillette Grove, Iowa. |

2. Chylus cells were found only in Fridericia. The characters of these cells are distinct for each species and show evidences of taxonomic value.

3. Studies on the penial bulb have shown that in all of the available material its structure is uniform in all of the specimens of a given species and that characters of taxonoric importance are to be found in this organ. However Eisen's classification of the subfamilies and genera based on the characters of this organ is faulty. In Marionina provision must be made in the definition for the presence of an occasional accessory gland in connection with the bulb. The stability of the subfamily Enchytaeinae is very uncertain since it contains only one genus, namely, Enchytraeus, which is now known to contain several species which have penial bulbs of both the Lumbricillid and Enchytraeid types as well as

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transitional forms connecting the two. Alterations must be made in Eisen's oharacterization of the bulb in the genus Fridericia to provide for wider variation in the number of sets of Component cells in this organ.

4. Stephenson has recently described forms which have transitional characters between Lumbricillus and Enchytraeus. Additional evidence of the close relation of these two genera is now offered since it is shown that the penial bulb of the latter shows distinct transitions from the so called Enchytraeid type to the Lumbricillid type. These two genera were formerly regarded as standing far apart.

5. The following statements refer to a single species, Lumbricillus rutilus n. sp.

- a. L. rutilus occurs in abundance in the sprinkling filters of the Chicago Sewage Testing Station during the warm months of the year. Its distribution in the various tanks and filters depends chiefly upon the dissolved oxygen content, the H<sub>2</sub>S content, and the "freshness" of the influent.
- b. This worm is associated with a number of other animal forms of which the larvae and pupae of Psychoda albimaculata, Prorhynchus sp., Pristina; sp., Nais sp., Helodrilus subrubicundus, Chironomidae, nematodes, Water Wites, and Collembola(Isotoma sp.) are the most common.
- c. The sole mode of progression is by crawling. It is much more efficient on moist surfaces than on dry ones, the latter being an important barrier against dispersion. Progression is more efficient on rough than smooth surfaces. There is no evidence whatever of

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ability to swim.

- d. These worms are sensitive to light and show a decided negative response to it.
- e. Exposure to dry conditions results fatally within a short time, usually less than five minutes.
- f. These worms are positively thigmotactic. They show a distinct tendency to accumulate in masses and to orient themselves in such a way that a maximum of contact with the sludge and the filter bed rock is secured.
- g. The maximum temperature of the organisms is very near 36°C. From 25° to 10° no difference in the activities of the worms was noted. From 10° to 2° the activity was reduced. They can live in a temperature of 5°C for days and even weeks.
- h. These worms require an abundant supply of oxygen. Continued low dissolved oxygen content in the medium has a deleterious effect and great abundance of these forms in the sprinkling filters is due in part to the high dissolved O content of the sewage which comes in contact with them. They cannot thrive in crude sewage because of its low dissolved oxygen content.
- i. The worms show a distinct recognition of the presence of sludge and react positively to it.
- j. The worms have a mechanical effect on the accumulating sludge in the sprinkling filters by loosening it up and working it over thus facilitating the oxidation of the unstable organic matter. Also

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oiroumstantial evidence points to these forms as at least one of the factors which bring about the "unleading" of the filters in the Spring.

k. It has been shown that these worms increase the putrescibility of the sewage in which they occur by using a part of the available oxygen. This is of great economic importance. They interfere with the efficiency of the sprinkling filter and aid in rendering the sewage unstable, facilitating anaerobic decomposition. In this particular respect they are undesirable organisms in sewage disposal plants.

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VII.APPREVIATIONS USED IN THE PLATES.

a. - - - Amoulla. bs. - - Elood sinus. bso. - - Perivisceral blood sinus. bvd. - - Dorsal tlood vessel. bvv. - - Ventral blood vessel. cc. - - Clitellar cells. cd. - - Cavity of diverticulum. ch. - - Chylus cell. chc. - - Chloragog cell. cil. - - Cilia. cu. - - - Cuticula. cyl. - - Lining layer of cytoplasm. dtl. - - Luren of digestive tract. ecg. - - Glands at ectal opening of spermatheca. eco. - - Ectal opening. ecs. - - Ectal surface. 91. - - - Epithelial layer. ens. - - Ental surface. epc. - - Epithelial cell. glo. - - Perioheral gland cells. h. - - - Hypodernis. ibc. - - inner bulb cells. icc. - - Intracellular canal.

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T. - - - Musculature.
Col. - - Circular suscle layer.
Tr. - - Retractor suscle.
TS. - - Muscle strand.
n. - - - nucleus.
p. - - - Peritoneum.
pb. - - Penial bult invagination.
pp. - - Penial pore.
sd. - - Spers duct.
sde. - - Extension of the spers duct.
spd. - - Spersathecal duct.
td. - - Tubules of diverticulum.
tdo. - - Origin of the tubules of the diverticulum.

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VIII. EXPLANATION OF PLATES.

Plate I.

Henlea moderata n. sp.

- Fig. 1. Dorsal view of the brain.
- Fig. 2. Lateral view of a nephridium.
- Fig. 3. Bundle of setae.
- Fig. 4. Spermiducal funnel.
- Fig. 5. Lymphocyte.
- Fig. 6. Lateral view of the anterior region showing the shape of the prostomium.
- Fig. 7. Spermatheoa.
- Fig. 8. Transverse section through the floor of the buccal cavity showing the four "taste organs".
- Fig. 9. Seta.
- Fig. 10. Transverse section of the digestive tract in VIII showing the origin of the tubules of the intestinal diverticulum.
- Fig. 11. Transverse section of the digestive tract in VIII showing the structure of the intestinal diverticulum.
- Fig. 12. The penial bulb as it appears in a transverse section of the worm in that region.

#### Plate II.

#### Lumbricillus rutilus n. sp.

- Fig. 13. Diagram of the anterior region showing details of the circulatory system.
- Fig. 14. Spermiduoal funnel.
- Fig. 15. Nephridium.
- Fig. 16. Spermatheca.

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|---|
| Longitudinal section through the spermiducal funnel.                    |
| Outline sketch of the ventral glands in XIII and XIV.                   |
| Bundle of setae showing typical shape and arrangement.                  |
| Transverse section through the ventral gland in XIII.                   |
| Dorsal view of the brain.   |
| Transverse section through the ventral gland in XIV.                    |
| The penial bulb as it appears in a transverse section through the worm. |
|   |

Fig. 23. Longitudinal section through the spermatheca showing the details of structure.

#### Plate III.

Fridericia douglamensis n. sp.

- Fig. 24. Diagram of the circulatory system in the anterior region of the worm.
- Fig. 25. Spermatheca.
- Fig. 26. Transverse section of the clitellar cells.
- Fig. 27. Nephridium.
- Fig. 28. Spermiducal funnel.
- Fig. 29. Seta showing the ourved proximal end.
- Fig. 30. Doreal view of the brain.
- Fig. 31. Penial bulb as it appears in a transverse section of the worm.
- Fig. 32. Peptonephridium.
- Fig. 33. Chylus cells as they appear in longitudinal section through the wall of the intestine.

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#### Plate IV.

#### Fridericia oconeensis n. sp.

- Fig. 34. Dorsal view of the brain.
- Fig. 35. Chylus cell as it appears in longitudinal section through the wall of the intestine.
- Fig. 36. Nephridium.
- Fig. 37. Spermatheoa.
- Fig. 38. Peptonephridium.
- Fig. 39. Transverse section through the intestine in the region of the chylus cell zone.
- Fig. 40. Spermiduoal funnel.
- Fig. 41. Penial bulb as it appears in a transverse section of the body of the worm.

#### Plate V.

Fridericia sima n. sp.

- Fig. 42. Peptonephridium.
- Fig. 43. View of the anterior region of the worm showing the shape of the prostomium.
- Fig. 44. Nophridium.
- Fig. 45. Chylus cell as it appears in a longitudinal section of the wall of the intestine.
- Fig. 46. Nephridium.
- Fig. 47. Spermatheoa.
- Fig. 48. Dorsal view of the brain.
- Fig. 49. Spermiducal funnel.
- Fig. 50. Penial bulb as it appears in a transverse section of the worm.

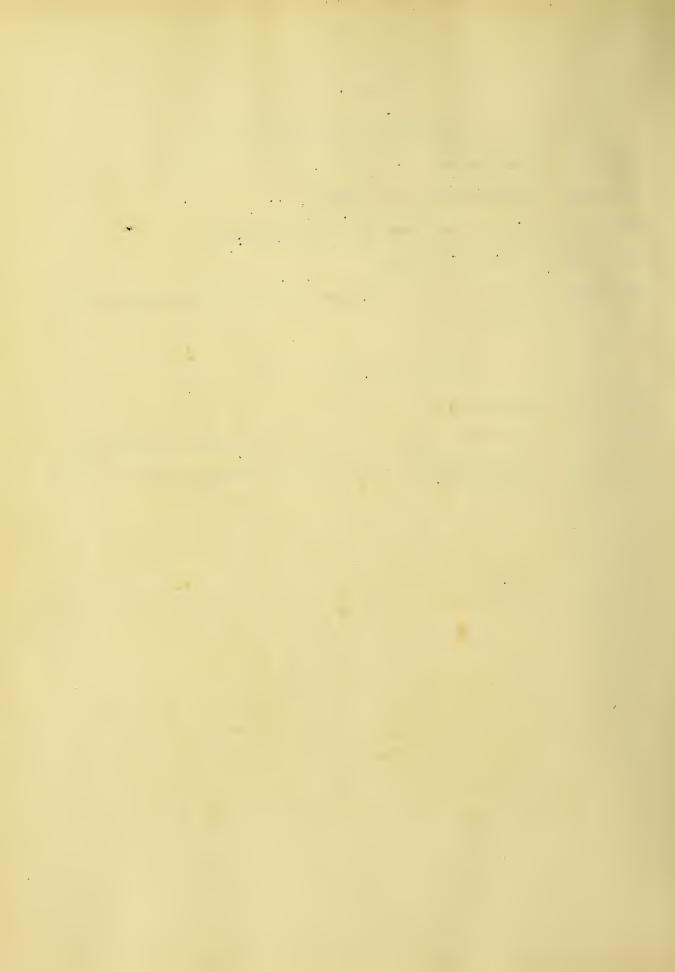
# Plate VI.

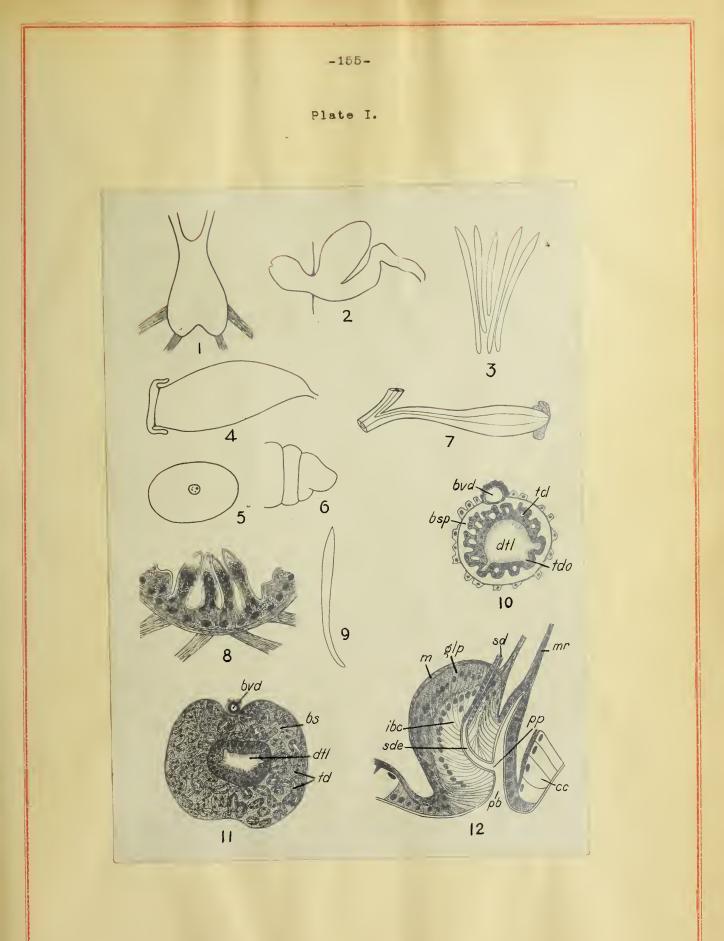
# Enchytraeus gillettensis n. ep.

- Fig. 51. Nephridium.
- Fig. 52. Dorsal view of the brain.
- Fig. 53. Longitudinal section of a spermatheca.
- Fig. 54. Spermiducal funnel.
- -Fig. 55. Penial bulb as it appears in a transverse section of the worm.

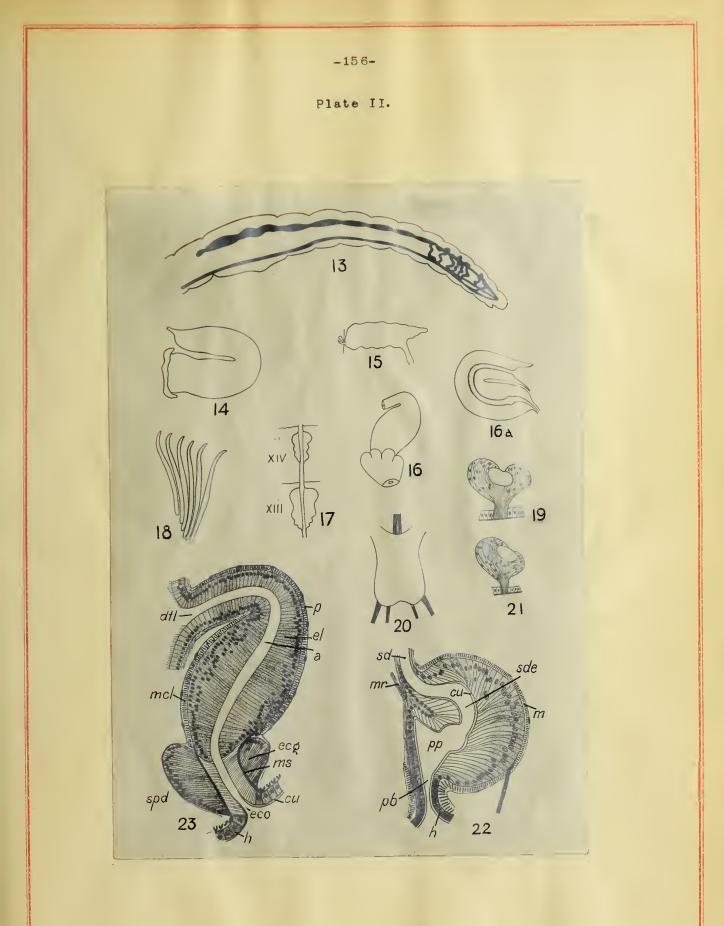
Henlea urbanensis n. sp.

- Fig. 56. Spermatheca.
- Fig. 57. Transverse section through an intestinal diverticulum.
- Fig. 58. Penial bulb as it appears in a transverse section of the worm.

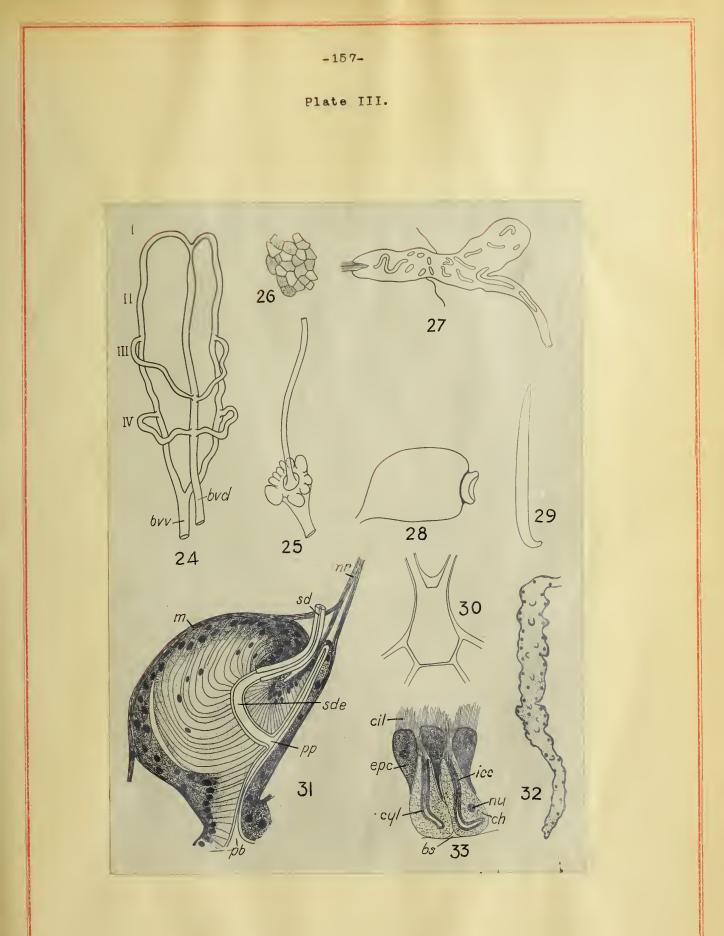




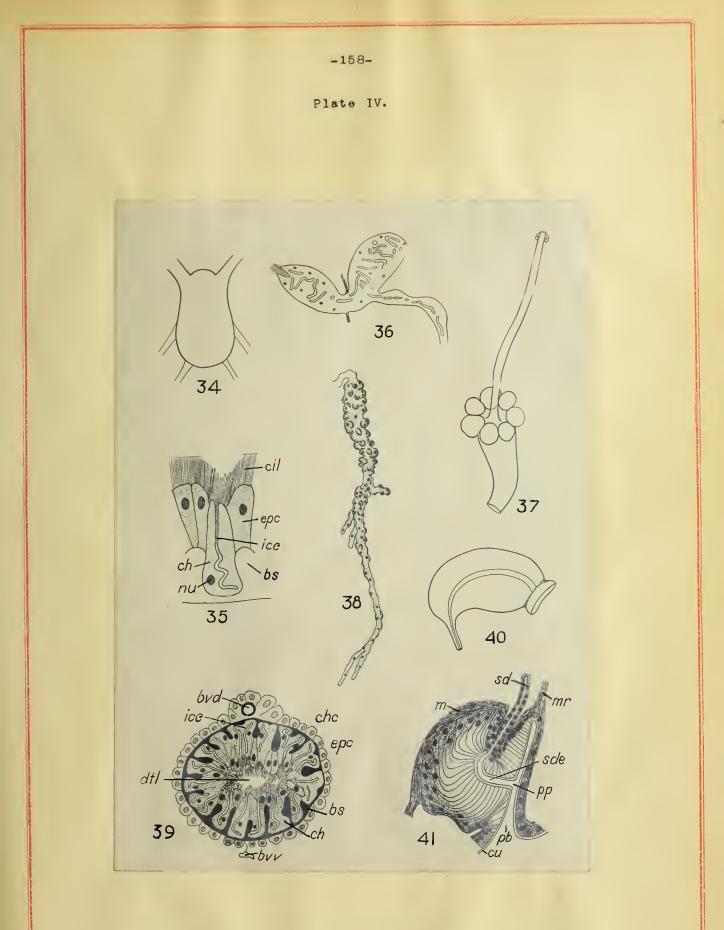




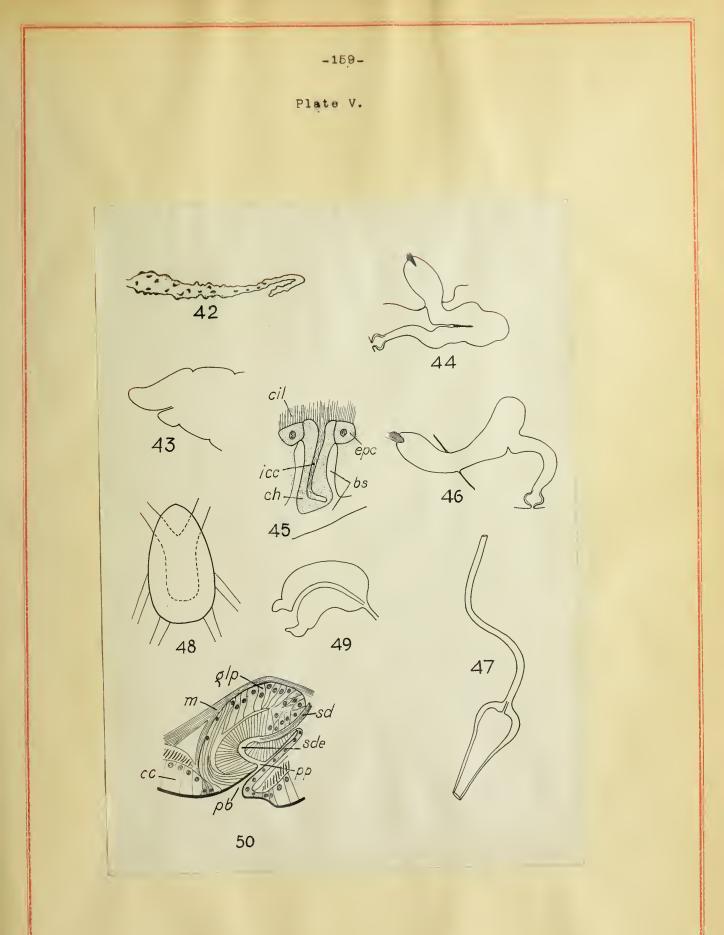




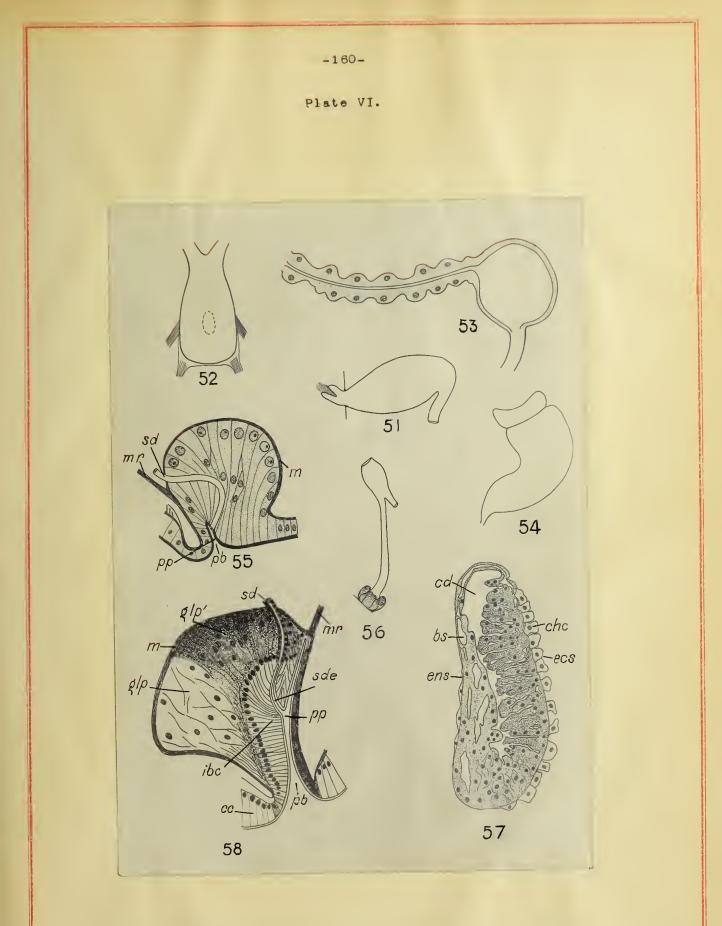












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Paul S. Welch .

# Vita.

| 1852, January 22th. | Forn in Montgomery County, Illinois.  |
|---------------------|---|
| 1989-1999           | Attended Public Schools in Auduton Township,<br>Montgowery County, Illinois.        |
| 1839-1901 .         | Attended Public Schools at Oconee, Illinois.  |
| 1901 <b>-1</b> 904  | Teacher in Putlic Schools, Shelty County,<br>Illinois.                              |
| 1904-1903           | Academy of James Millikin University, Decatur,<br>Illinois.                         |
| 1906-1910           | College, James Villikin University.   |
| 1910                | Honor Student in Class of 1910.   |
| 1306-1907           | Student Assistant in Piology, Villikin Univ.  |
| 1907-1909           | Student Assistant in Physics, Villixin Univ.  |
| 1909-1910           | Assistant in Fiology, Villikin Univ.  |
| 1903 and 1909       | Assistant in Eiclopy in Summer Sessions,<br>Villikin Univ.                          |
| 1906 <b>-191</b> 0  | Assistant in the Farnes Depidoptera Collection,<br>Decatur, Illinois.               |
| 1910-1911           | Grappate Scholarsnip in Zoology in University<br>of Illinois.                       |
| 1911-1918           | Fellow in Zoology, University of Illinois.  |
| 1911 and 1912       | Assistant in Entorology at the Eiological<br>Station of the University of Michigan. |
| 1918                | Appointed Instructor in Entorology at the   |
|                     | Fiological Station of the University of<br>Vichigan.                                |
| 1911                | Elected member of Illinois Chapter of Signa Xi.                                     |



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#### legrees.

- 1910. A.F. Janes Willikin University.
- 1911. A.M. University of Illinois.

### Publications.

- 1911. "Studies on a Phosphorescent Persudan Annelia, Odontosyllis enopla Verrill." Joint author with Professor T.W. Salloway. "rans. Am. Vier. Soc. vol. XXX, no. 1, pp. 18-89, 5 pls. 30 tigs.
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- 1912. "Otservations on the Life History of a new species of Psychoda". Annals of the Ent. Soc. Ar. vol. V, no. 4, ob. 411-419, 2 pls., 11 figs.
- 1218. " A rethod for the Preparation of Earthwords and smaller Oligochaeta for study". School Science and Mathematica vol. 18, pp. 15-19.





