ERRATA.

Page 12, lines 16 and 17, for one hundred read three hundred and for one thousand read six hundred.

Page 17, line 2, dele first letter in the line.

Page 168, line 12, page 177, lines 13 and 14, and page 271, line 10, for Lemna trisulca read Spirodela polyrhiza.

Page 209, line 2 of foot-note, after but insert represents.

Page 256, line 7, and page 266, line 19: snowi n. s. has been shown to be hieroglyphica, o^{\wedge} .

Page 257, insert as line 8 as follows: -ken to the office produced young in ten days. The

Page 272, line 13, for P. biguttatus read Pompilus biguttatus.

Page 278, Plate V., 16, after view insert as follows: a, mentum; b, labial rudiment; c, maxillary palpi; d, maxilla; e, labrum; f, antenna; g, eye; h, mandible.

Page 286, line 11, drop initial the one line.

Page 386, line i, for Comstocki read Comstock.

Page 399, line 17, for specimens read specimen.

Page 411, line 10, for Michaelson read Michaelsen.

Page 441, line 3 from bottom, for 66 read 68.

Page 445, line 10 from bottom, for 57 read 58.

Page 466, line 1 from bottom, for Cypria read Cypris.

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VOLUME IV.

ARTICLE I.—Bacteria Normal to Digestive Organs of Hemiptera. By S. A. Forbes.

In 1833 Léon Dufour described and figured in his "Recherches sur les Hémiptères,"* under the non-committal name of "cordons valvuleux," some curious appendages of the alimentary canal in Scutelleridæ, Pentatomidæ, and certain Coreidæ, misinterpreting their structure, however, and expressing no opinion as to their function, which, in fact, he called a mystery.† These organs had been previously distinguished by Ramdohr (1811), and they have since been several times referred to by entomologists as glands or follicles secreting a digestive fluid, presumably pancreatic. My own entomological studies did not make me particularly acquainted with structures of this class until 1888, in the autumn of which year. while studying the contagious diseases of the chinch bug (Blissus leucopterus) I made some dissections of that insect, isolating the alimentary canal and searching different parts of it for the source of a bacterial infection discovered in the fluids of chinch bugs when crushed under the microscope. This bacterial development I thus traced to certain cecal appendages of the small intestines so unlike the "cordons valvuleux" of Dufour's descriptions, that it did not for a time occur to me to connect the two; but in the course of some general dissections of

^{*} Pp. 149-151, etc., and figs. 1, 2, 6, 13, 19, 21.

^{† 1&#}x27;p. 150, 171.

Hemiptera made at my office this summer by my assistant, Mr. John Marten, and myself, it became evident that these dissimilar organs could be connected by intermediate gradations, that they had substantially the same anatomical relations and histological structure, and that all were alike, wherever they occurred, in the highly remarkable fact that they were invariably loaded with myriads of bacteria, differing in genus and species in the different insects, but always confined to these organs.* We further observed that in Coreidæ and Lygæidæ these cœcal structures might be present in one genus and absent in another of the same family, only the higher Hemiptera (Pentatomidæ, Scutelleridæ, Corimelænidæ, etc.) invariably possessing them, and the lower Hemiptera invariably wanting them. In case they were absent, their bacterial relationship was never assumed, in whole or in part, by any other organ. The occurrence of "masses of motile vibrio-like objects" in these glands in a Pentatoma was noticed by Leydig in 1857, but I have found no other mention of the matter than that on page 337 of his Lehrbuch der Histologie.

There are certain cœcal appendages of the alimentary canal of other orders of insects, which have a general resemblance to these in Hemiptera, but can be considered homologous with them only in a very loose sense of the word, since they are certainly not homogenous. These gastric pouches in grasshoppers, cockroaches, and carabid beetles do not commonly contain bacteria so far as we have been able to determine. In fact, the only other insect structures in which we have found bacteria normally present with any constancy, were the fatty bodies of various species of cockroaches. It is not absolutely certain that these objects from cockroaches are bacteria, as they have not yet been cultivated, our own recent efforts having failed, as did Blochmann's.†

^{*} No hibernating specimens have as yet been examined, and it is possible that this phenomenon will be found to disappear with the functional quiescence of these glands.

[†] Biologisches Centralblatt, Vol. VII., p. 606.

Balbiani's observations, however, (reported in *Comptes Rendus*, Vol. 103, p. 952) to the effect that bacterial forms introduced in the blood of insects are taken up by the cells of the pericardial tissue and destroyed therein, give a certain probability to the hypothesis that these seeming bacteria of cockroaches are really such. It is true that Balbiani's statements are limited to the pericardial tissue in the vicinity of the heart; but as Kowalevsky has shown* that this tissue is intermingled in many insects with the so-called fatty bodies, it is not unlikely that a more general and critical search would have shown the cells in question to have the same functions wherever found.

There can, however, be no doubt as to the nature of the objects found in the cocal appendages of the Hemiptera above mentioned. They not only present every visible characteristic of micrococci and bacilli, but by their reaction to stains, their resistance to prolonged treatment with solutions of caustic potash, and especially and conclusively by the success of our culture experiments with both fluid and solid media, they answer to all the tests applicable to the recognition of bacteria.

These cocal structures are probably shown in their simplest form in Pyrrhocoridæ (see Dufour, p. 171, and figures 17 and 21), although in the absence of specimens of this family for microscopic examination I can only repeat Dufour's surmise that the small and variable cocal pouches of the small intestine in these Hemiptera are homologues of the complicated apparatus of Anasa and Euschistus. The next simplest form of this organ which I have thus far seen, is that of the chinch bugs where it consists of five to eight large coca radiating from a common point of attachment on the intestine about .2 mm. behind the third stomach. These coca are about .12 mm. in diameter, and average 1.5 mm. in length. They are straight or slightly contorted, with smoothly rounded ends, and are nearly filled, when in normal condition, with large, pale, loosely-attached, sub-

^{*} Biologisches Centralblatt, Vol. IX., p. 44.

spherical cells, similar to those of the gastric epithelium, and like them usually binucleate, but containing more fatty granules. These cells are variable in size, and individual ones become greatly swollen, and probably break down in secretion. The lumen of the tube is an irregular linear space, not always readily distinguishable in the midst of the cells. That these tubules actually communicate with the intestine at the point of their insertion, I have repeatedly demonstrated under the microscope by carefully readjusted pressure on the cover-glass. By this means granules may be made to pass freely from any one of the cœca into the intestine, and even from the third stomach into the cœca through the slender portion of the intestine connecting them.

The microbe of these ceca (in the chinch bug commonly *Micrococcus insectorum* only) occurs primarily in the intercellular fluids of these structures, and was excessively abundant in every one of a great number of specimens, the ceca of which I examined separately. My specimens were from all parts of the State of Illinois and from Kansas, and were of various ages, from young immediately following the first moult to the adult. A thorough exploration and examination of all the other organs of these chinch bugs failed to discover any trace of this or any other bacterium, with the exception of an occasional infection of that part of the intestine into which the ceca open.

Examples of five other genera of Lygaeidæ have thus far been dissected by us, in three of which (Lygaeus turcicus, Nysius angustatus, and Geocoris uliginosus) there is no trace of these "pancreatic" organs, while in two others (Trapezonotus nebulosus, and Myodocha serripes) they are present in a stage of development quite above that of the chinch bug, but far below that characteristic of the higher Hemiptera. In Myodocha, for example, they are made up of numerous cœcal tubes arranged side by side in a single layer, in flat, leaf-like lobes, three in succession, the largest leaf anterior, and the middle one of the series the smallest, the three being bunched together,

wrapped around the stomach, and imbedded in fatty tissue in a way to require careful dissection for their display. The tube-like structures of which these leaflets are composed are thickest distally, and are attached by their narrow ends to the alimentary canal, which in the first lobe is indistinguishable from the edge of the sheet itself. This anterior sheet is irregularly palmate, the longest of the finger-like ceca measuring 1.1 mm. and the shortest (those most posterior) about .7 mm. The transverse diameter of a single cecum at its broadest end is about .07 mm. This lobe is partly folded together, the folds being held in place by branches of a large trachea, which is distributed abundantly to all parts of the structure.

The second or smallest lobe is attached to the intestine by a narrow insertion about .1 mm. behind the preceding. It is .5 mm. long by about .2 wide.

The third lobe, of medium size, is also attached by a narrow insertion to the intestine immediately beyond the preceding. It is quite regularly palmate in form, is supplied by a single much-branched trachea, and measures about .7 mm. long by .5 wide.

Crushing successively and separately all the portions of the alimentary canal upon cover-glasses, and treating by the usual methods for the demonstration of bacteria, I found all the preparations quite free from them, with the exception of those from the above-described leaf-like cœcal structures; and in these, and in every part of them, immense numbers of a minute Micrococcus occurred (not M. insectorum), situated, as in the chinch bug, chiefly between the large spherical cells of which these bodies were principally composed. Several repetitions of this experiment with other specimens gave the same result. In Trapezonotus this organ has the same structure and general appearance as in Myodocha serripes.

Among the Coreidæ I have seen it in Anasa tristis, Alydus pilosulus, and A. eurinus, but have found nothing resembling it in Corizus lateralis. In this family it has a much more considerable extent than in the foregoing,

and now takes the form of rows of short, transverse cocal tubules, standing in general at right angles to the small intestine, but with their inner ends the smaller. They are fused into a continuous layer, and make by their arrangement a broad plaited border on each side of the intestine for its whole length, from the stomach to a bladder-like expansion into which the Malpighian organs open. A large trachea runs along the intestine, and its branches are very liberally distributed, right and left, to all parts of these gland-like bodies*. The tubules are lined with a single-layered epithelium very different from that of the part of the intestine into which they open. They may be easily demonstrated, by pressure under the microscope, to open separately into the alimentary canal running along between the rows, and the same fact is evident in stained sections. In every case, again, the intercellular substance within these tubules is little more than a mass of bacteria,-micrococci or bacilli, as far as determined.

The same may be said of the Pentatomidæ and Corimelænidæ dissected,—Corimelæna, Peribalus, Mormidea, Euschistus, and Hymenarcys,—except that in these families there are always four rows of the short transverse tubules instead of two. In Capsidæ, Nabidæ, Reduviidæ, and Aradidæ—the only other families examined with this matter in mind—we have not found these structures, and Dufour notes their absence in examples of these families, and in Miris, Phymata, Cimex (Acanthia), and the lower Hemiptera generally.

In every case where they have occurred in our dissections, we have made exhaustive search for bacteria in other parts of the alimentary canal also, and in the salivary glands, the fatty bodies, etc., and in all these Hemiptera with only negative results.

^{*} The abundant tracheal supply of these organs and the minute distribution of the tracheal branches—scarcely less abundant than in the fatty bodies, and much more so than in other portions of the alimentary canal—hint at a peculiar function for this so-called pancreatic apparatus.

I have no present desire to speculate concerning the meaning of the bacterial contents of these glands, but limit myself to this preliminary account, and await the completion of the several investigations in which we are now engaged,—upon the distribution of the ecca, their variations, and their relations to the habits of the species possessing them, and upon the kinds and nature of the bacteria constantly harbored by these interesting appendages of the alimentary canal.

Concerning their relations to insect disease, I will only add that in chinch bugs perishing gradually but rapidly, we find them varying considerably in number in the cocal appendages; and that where they are most abundant, the epithelium of these structures is completely disorganized, only the basement membrane remaining, in the form of cocal tubes filled with a pure culture of *Micrococcus insectorum* and a little granular debris, the latter apparently the product of the decomposition of the epithelium.