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XV.—*On some Organisms found in the Excrement of the Domestic Goat and the Goose.*

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(Read 8th November, 1882.)

PLATE VII.

WHEN studying lately the appearances of the hay-bacillus, both in the fresh and putrid infusion of hay, it occurred to me that it would be worth while to examine the excrement of a herbivorous animal, and a grass-feeding bird. Accordingly, I procured the fresh excreta of the goat, and of the goose, taking from the latter only the part but little, if at all, contaminated with urates. The examinations were begun in the month of August last, and as they proved rather interesting, and may open up a study that may furnish results for experiment, I venture briefly to offer a few remarks upon the organisms found. Photomicrographs were made of some of these, by a Seibert's $\frac{1}{6}$ water-immersion objective, without collar adjustment, kindly lent to me by Mr. Curties for this purpose. I may remark that the objective answered well with the

EXPLANATION OF PLATE VII.

(Lithographed from some of the Photomicrographs exhibited.)

- Fig. 1.—A few of the free and growing spores found in the mixture of the goose excrement.
„ 2.—Part of the layer immediately beneath the upper layer, containing micrococci, in the mixture of the goose excrement.
„ 3.—Part of the felted mass of short rods, and part of some long free filamentary chains. These appeared later in the same mixture.
„ 4.—The earliest notice of *Spirillum* in the goat excrement.
„ 5.—Shows the marked increase in the number of *Spirilla*, and the diminution in the number of the rods.
„ 6.—*Spirillum* from goat excrement. [The original photomicrograph was taken with the addition of a Zeiss amplifier, and magnified to 651 diameters; the others, each, to 365 diameters.]

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camera extended to a certain length, and also when used with a Zeiss amplifier at the same extension; but at distances much beyond, the want of the collar adjustment became rather apparent; hence the enlargement of the objects was chiefly kept within very moderate dimensions, yet sufficient for illustration. I may add, that I believe they are within easy reach of the engraver's power, which apparently was not the case with the photographs of the minute organisms found in rainwater-ice and hail, described in a previous paper.*

The examination was proceeded with in the following manner: a portion of fresh excrement of the goat was broken up with a clean glass rod in some freshly distilled water, and immediately covered with a glass plate. Examined by the Microscope at the time, numerous bacteria of various sizes, and a very few rods of, apparently, hay-bacillus were present, besides large quantities of partially digested vegetable matter suspended in a somewhat glutinous material, most likely mucus from the intestinal canal. The mixture was set aside in the room without artificial heat, the ordinary temperature ranging between 60° and 70° F., and fully exposed to daylight. On the third day when examined there was a thin scum extending over a large part of the surface. This scum contained numerous bacteria, some small rods, and here and there in the thinner parts of the pellicle some bright oval bodies with sharp outlines, whilst free from the scum were many larger and longer rods, both straight, and with a well-marked wide curvature, not angular; these rods were in active movement; there were also a few spores with outgrowths, these had the spore ends nearly globular, and the outgrowth or extension very pale and slightly granular, with a *gentle curve*. These spores had a slight forward and backward movement, also a peculiar swaying motion from side to side, the spore end forming the fulcrum. I believe these must be regarded as the spores of the *Spirillum*, which appeared later. A portion of the upper part of the fluid was removed, and freshly distilled and reboiled water added both to it, and to replace the quantity removed from the original portion. Both vessels, being covered, were placed in a dark box, and kept at a pretty constant temperature of 90° F. for twelve hours, when a fresh examination was made. Both vessels now teemed with infusoria and bacteria, the fluids had become more or less ropy, especially the original one. The organisms in the diluted portion consisted chiefly of active rods of very variable lengths, many having the wide curvature. No spores were visible. The original portion which had been diluted was divided into three layers, the surface one being of a dark greenish-brown colour, the second very much paler, whilst the lowest consisted of the debris of the food.

* *Ante*, p. 449.

The rod organisms in the upper layer resembled those in the diluted portion, but were far more numerous, and in very active movement, the bend in the curved ones being used apparently as an axis for locomotion. The bacteria had very little motion, the fluid being most likely too slimy; no spores were visible at this stage.

Hay was steeped for twelve hours in cold water and the liquid sterilized by boiling; when cold, a portion was added to the original but diluted mixture and gently stirred, whilst another portion of the hay infusion was infected from the former. All three vessels were now left exposed to daylight and the ordinary temperature of the room for a couple of days. Re-examined, the long rods had almost disappeared in the hay infusion that had been infected, and chiefly very short rods could be found; bacteria and infusoria were also present.

In the mixture simply diluted with distilled water, the rods were now fewer and less active; a pellicle on the surface was crowded with motionless bacteria; the infusoria still abounded; there was no offensive smell. The original fluid, or mixture, now diluted with the hay infusion, was examined. It was densely crowded with straight and curved rods of very variable lengths, and a few spirilla were visible, some having only one-and-a-half turns, others two to six; these were in active movement. In Fig. 4 (Pl. VII.) is represented the first notice of *Spirillum* in the mixture. In different parts of the slide some single large micrococci and also smaller double ones were noticed. The infusoria still abounded, and the mixture had now a faint sickly odour. Attention was confined to this mixture. After another twenty-four hours the rods and spirilla appeared nearly equally abundant, some of the latter having as many as thirty-three angular turns. The curvatures, both in width and depth, differed considerably. These organisms continued about matched for four days, when the *Spirillum* got the upper hand, the number of rods lessening; this is fairly well shown in Fig. 5. The survival of the fittest was evidently taking place, but at the same time also appeared another organism contending for the mastery, viz. a very delicate mycelium spreading in every direction through the fluid, which quickly rendered all further observation useless. The fluid was, however, kept for five weeks, and at the end of that period the rods and spirilla had well nigh disappeared, and nothing could be found by which to determine to what object the mycelium belonged. The mycelium was in very long twisted threads not larger than the rods, and at first I fancied they might be the rods in their filament stage, but close examination soon showed this not to be the case, as the threads had short outgrowths at very variable distances.

In the excellent contribution upon the life-history of *Spirillum*

given by P. Geddes and J. Cossar Ewart, M.D., in the Proceedings of the Royal Society, No. 188, 1878, it is shown that at one term of existence, the screw-shaped rods become less twisted, and finally straight, passing into the ordinary rod form as in Fig. 7, given by them. They also suggest that the term *Vibrio* should not be considered as generic. When examining the above-named fluids I have repeatedly found many of the long spirilla motionless, with one half having lost all twist except a large gentle curve, but that end presented a very delicate pale, very finely granular condition, differing entirely from the other part, the end being scarcely visible even when stained, and I have regarded this as a progressive dissolution of the organism. In the case I have mentioned we might have expected the spirilla to have reverted to rods, which was not the case, so far as I could determine, and from the *Spirillum* found in the goat excrement, appearing after and so largely replacing the rods, I think it offers a fair plea that *Bacillus* and *Spirillum* are to be considered distinct, though the latter, when broken up, may greatly resemble bacillus rods. The straight rods I should regard as *Bacillus subtilis*, and the curved ones as merely an accidental variation in their form, though many with a single curvature had very much the appearance of *Vibrio rugula*.

In the same paper it is stated the parent or spore-bearing hyphæ are locomotive, "and the spores quiescent." The authors say, "The life-history of *Spirillum*, so far as we at present know, may be thus summarized. The well-known motile corkscrew may alternate between the active and resting states, and ultimately lengthen out into a small filament, which loses its definite twist, and may freely bend or straighten. This thread grows into a much larger and longer motionless filament in which spores appear. These rapidly divide and acquire a bright brown colour, the filament re-assuming the motile condition, and sooner or later breaking up."

The spiral organisms were rigid, with a spiral movement. In size they appear rather smaller than the figures given of *Spirillum volutans*, and larger than *Spirillum tenue*, approaching nearer to the *Vibrio serpens* of Cohn. If the term vibrio were put aside, would it not be as well to substitute for the curved forms of *Bacillus*, *Bacillus curvatus*, or *Bacillus subtilis* var. *curvatus*, and thus help to get rid of the objectionable term? Having some doubt as to the exact species of *Spirillum*, I have not given more than the generic name.

I found the organisms varied so considerably according as they were left dry upon the cover-glass after or before staining, or mounted in distilled water, or in a semi-saturated solution of acetate of potash, or in dammar medium, that I have not given the measurement. The one method of mounting would not agree with the

others, as shown in some other photomicrographs, taken with the same objective, at the same distance from object to screen; but I have given the measurement of the $\frac{1}{1000}$ of an inch at the same distance.

The examination of the goose excrement, the part not covered with urates, was made by breaking up a portion in freshly distilled water with a clean glass rod, then covering it with a plate of glass, and setting it in the daylight at the ordinary temperature of the room, at the same time as the former experiment. Examined on the slide, chiefly vegetable debris of grass, coarse and fine granular organic and mineral matters, with here and there a bright point, like an ordinary bacillus spore, amongst various bacteria, and a very few short rods were noticed.

After twenty-four hours, a thin scum appeared in several places on the surface of the fluid, which had now settled into three layers, the heavy solid constituents having sunk to the bottom. The top one was of a dense brown colour, the middle much clearer and less coloured. After another twenty-four hours the top liquid was examined; being diluted with a droplet of water on the slide, it was seen to contain numerous very bright oval forms, many with outgrowths of varying lengths, evidently germinating spores, apparently of the hay-bacillus. These had motion forwards or backwards; but not the singular swaying movement from side to side from a fixed point. There were also a few short rods, micrococci, and bacteria present. These were photographed, Fig. 1. On the following day the short rods had notably increased in number, but they did not appear to grow in length; fewer spores in growth were visible. The pellicle on the surface had increased, the part exposed to the air consisting, so far as I could make out, of bacteria mingled with micrococci, whilst immediately beneath, the micrococci formed a layer in a delicate transparent medium. This layer is seen pretty distinctly in Fig. 2. In various parts of the pellicle on the slide, small masses of minute bodies, highly refractive and set in a gloea, larger than the spores of the hay-bacillus, were seen. I believe they belong to a *Bacillus* of larger dimensions, as I have many times noticed similar bodies in connection with a short chain of stout short rods, in other preparations. Continued examinations for many days revealed nothing further; the rods had not grown, and the entire fluid was becoming of a greenish colour throughout, but at last upon several slides the bacilli were seen in chains of some length and in nearly all attached to a felted mass of small rods, and rods lying free, but close to the mass, as depicted in Fig. 3. In the filamentary chain, the joints appeared to be passing into the spore condition in a few. The little mass of free rods were motionless and of rather paler appearance than the ordinary rods of hay-bacillus; the fluid was crowded with infusoria,

there were numerous bacteria, but not active to any extent. The fluid had become ropy, the dark colour had lessened, the odour had become disagreeable. An attempt was made to cultivate the organisms in fresh sterilized hay infusion, but it was unsuccessful.

The original fluid was now stirred up, and allowed to re-settle, still it yielded nothing of change that I could discover upon the examination of many slides. It was kept for more than a month when the fluid had a sour smell and acid reaction. To the latter I think we may attribute the want of growth of the rods generally; evidently the pabulum was not favourable; at this stage a few octahedra of oxalate of lime were seen. The results offered a great contrast to those of the excreta of the goat.

Possibly, by fractional cultivations in proper media, we might be able to arrive at a more perfect study of the different organisms, and test their physiological peculiarities or their pathological reactions, if any. We may, I think, however take for granted that the spores have resisted the entire digestive process in both cases, but whether they, or the spirilla, would prove detrimental to guinea pigs or mice, I must leave to the care of those armed with the necessary powers, in this country, for such studies.
