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IV.—The Larval stage of *Hypoderma bovis*

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transversely striated, the striæ running into reticulations which are stronger in the centre. Pro- and mesopleuræ closely punctured except at the apices; the metapleuræ rugosely punctured except at the base above; the basal furrow distinct, deep, and marked all over with short stout keels. Mesosternum closely punctured, except in the centre; the furrow is wide and deep and becomes wider and deeper towards the apex. Near the base the mesopleuræ are raised; the apex of the raised part curved above, and it is marked at irregular intervals with some stout keels. Legs black, covered with white hair, the calcaria rufous. Wings fusco-violaceous; the stigma and nervures black; the first transverse cubital nervure is oblique, faint above, completely obliterated below; the first recurrent nervure is received shortly beyond the middle, the second in the basal third. Abdomen shining, the basal segments slightly, the apical strongly punctured; the segments strongly constricted at the base, most strongly on the lower side; the constriction on the second segment finely striated.

As the fact is not alluded to by Col. Bingham, it may be pointed out that the males in this genus have hairy eyes. This makes the fourth Indian species.

Khasia Hills.

IV. — *The Larval Stage of Hypoderma bovis.* By P. KOOREVAAR, Veterinary Surgeon to the Amsterdam Public Abattoir *.

AT a meeting of the Nederlandsche Dierkundige Vereeniging held in October 1895 Dr. C. Ph. Sluiter exhibited certain *Æstrus*-larvæ which had been found by myself in the spinal canal of a young bullock.

Since then I have met with *Æstrus*-larvæ in the vertebral canal in the case of a large number of cattle from nine to eighteen months old and in certain full-grown animals up to the age of six years.

The larvæ lie freely in the fatty tissue between the dura mater spinalis and the periosteum, especially in the hindermost portion as far as the cauda equina.

In the fresh condition the larvæ are of a transparent white colour, with a light green interior. They are segmented and oblong in shape; the segmentations are more distinctly

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visible in specimens preserved in spirit than in the fresh larvæ.

The dimensions of these spinal larvæ vary considerably ; in a collection that I formed between October and February there is one specimen measuring 5 millim. in length by $\frac{1}{2}$ millim. in breadth ; the remainder are from 6 to 14 millim. long and from 1 to $2\frac{1}{2}$ millim. broad.

In the warm fat, before it has become solid, the larvæ lie extended ; if they are set free they assume a curved shape and contract, becoming in consequence considerably shorter and thicker ; in this contracted condition the annulations are also distinctly visible. As regards other details these grubs have the characteristics of *Cæstrid* larvæ.

In the year 1884 M. Hinrichsen, a veterinary surgeon, in dissecting a tuberculous bullock, found the first specimen of these larvæ in the spinal canal ; in 1888 he published in the 'Archiv für wissenschaftliche und praktische Thierheilkunde,' Bd. xiv., an account of the examination of thirty-nine cattle.

In the case of fourteen of these (for the most part the younger animals) he met with from one to twenty larvæ in a portion of the vertebral column. So long ago as 1863 Prof. Brauer described and figured these larvæ in his 'Monographie der *Cæstriden*,' but the fact that they occur in numbers in the spinal canal was nevertheless new.

Further statements as to the finding of these larvæ in the neural canal in cattle are also given by Hinrichsen, Horne, and Ruser in the 'Zeitschrift für Fleisch und Milchhygiene' for 1895.

Hinrichsen considered these *Cæstrus*-larvæ to be the first stage of *Hypoderma bovis*, which was till then unknown ; and Prof. Brauer agreed with this opinion.

Owing to the frequent occurrence of this larva in the spinal canal Horne was led to consider the latter as the normal hidden resting-place of *Hypoderma bovis*.

In the middle of January I met with the first specimens of *Hypoderma*-larvæ beneath the skin ; on a closer examination ten more *Cæstrus*-larvæ were found in the epidural fat of the vertebral canal.

The occurrence of *Hypoderma*-larvæ beneath the skin, in what are known as warbles, and at the same time of *Cæstrus*-larvæ in the spinal canal in the same animal, was repeatedly observed in the months of January, February, and March in the slaughtered cattle at the abattoir.

It is remarkable that in size and form the largest spinal larvæ do not differ from the youngest *Hypoderma*-larvæ in the subcutis ; the subcutaneous larvæ are somewhat less

transparent; the older subcutaneous larvæ (third stage) are dull white and marked with darker transverse bands; later on they become noticeably thicker and pyriform; the colour changes to more of a greyish yellow, and subsequently to dark brown; these larvæ have become encapsulated, have perforated the integument, and produce the well-known warbles in the skin.

In the months referred to one frequently finds that the spinal larvæ have crawled out of the fat and are lying in the spaces between the vertebræ; occasionally they are found with one extremity in the intervertebral spaces.

On the 28th of last February [1896] I also made an interesting discovery; in a yearling beast with a large number of *Hypoderma*-larvæ beneath the skin—the subcutis on the back and loins was violently inflamed and suppurating—three spinal larvæ were discovered in addition. On examining the very œdematous œsophagus, I found thirteen *Æstrus*-larvæ in the connective tissue between the mucosa and the muscularis; in size, shape, and colour they were similar to the larvæ lying in the spinal canal.

In the same way on two occasions after this I also met with *Æstrus*-larvæ in the wall of the œsophagus. *Æstrus*-larvæ have likewise been found in the œsophageal wall by Curtice; it afterwards appeared that these were the larvæ of *Hypoderma lineatum*, which occurs in the United States.

The interesting statement is also made by Horne that in the case of a young bullock he found the whole carcass so full of *Hypoderma*-larvæ that it had to be withdrawn from consumption.

Are these spinal and œsophageal larvæ really those of *Hypoderma bovis* in the first stage?

The great agreement between the youngest subcutaneous and the largest spinal larvæ in the same animal, the appearance of larvæ under the skin, coupled with the disappearance of the larvæ from the spinal canal, is a strong argument in favour of the view that this is the case. It is remarkable that no single investigator has met with the larvæ on the way from the vertebral canal to the subcutis. Horne asserts that he has more than once noticed in the flesh dirty green larva-tracks, which led from the spinal canal through between the muscles to beneath the skin. In one instance, where both subcutaneous and spinal larvæ were present, a larva was met with between the spinous processes of the vertebral column; hitherto I have not seen definite larva-passages. It is true that when *Æstrus*-larvæ are present the epidural fat has a dirty yellow colour; sometimes there is found

in the fat a green granular mass; above it is flabby and cedematous.

With a view to making sure whether these spinal larvæ were the larvæ of *Hypoderma*, I inserted them under the skin of another animal, in order that they might there become adult and in order from them, if possible, to breed *Hypoderma bovis*.

I selected as subject of experiment a small dog, which since the beginning of January had been supplied with food infected with *Echinococcus veterinorum*. On February 3rd I introduced, under aseptic precautions, through an opening 2 centimetres wide, beneath the skin in the left lumbar region eleven spinal *Æstrus*-larvæ taken from a calf. The wound was closed and healed quickly, the state of the dog remained normal, and nothing was seen of the eleven larvæ inserted.

Eight days later in the same manner fifteen larvæ were introduced under the skin of the right side; scarcely an hour later one of my colleagues and I reopened the wound, and saw to our astonishment that all the larvæ with the exception of one had disappeared. Two days afterwards I observed a larva lying under the skin on the costal wall, 8 centimetres away from the wound; its rounded oblong form was distinctly visible through the thin skin of the dog; one could feel it roll under the finger. On the third day it had crawled forward as far as the spinous processes, and on the fourth it had disappeared.

The dog remained normal; fourteen days after the first insertion there was still nothing to be seen under the skin.

What had become of the twenty-six larvæ in the dog?

To settle the point I decided to open the animal.

The autopsy was interesting.

On removing the skin five still living larvæ were found in the subcutis—one on the left costal wall, one in front of the shoulder, one on the right thigh, one on the skull, and the fifth larva on the point of the jaw. In the subcutis and in the underlying muscles no traces of their wanderings were observable; but certain cedematous spots were found in the subcutis and the muscles.

On opening the abdomen some bloody matter exuded. Six larvæ were found between the folds of the intestine, and therefore free in the peritoneal cavity.

I found, further, five larvæ in the fat of the spleen, kidneys, omentum, inguinal canal, and the retro-peritoneal tissue; the removal of the kidneys disclosed three more larvæ upon the psoas muscles.

In the pleural cavities no larvæ were found lying free; still five larvæ were met with, three of which were in the wall of the œsophagus and two in the peritracheal tissue.

The discovery in the spinal canal was very remarkable; on exposing the cord it was found that two larvæ were ensconced in the fat between the dura mater spinalis and the periosteum: the fat around the place was somewhat softer and injected with blood.

All the twenty-six larvæ introduced were found, and for the most part alive.

They had wandered far in a relatively short time—eight to fourteen days after they had been introduced under the skin.

In spite of careful examination no traces or tracks of the course followed by the larvæ could be found.

Certain small œdematous patches were indeed present in the subcutis and between the muscles, and some bloody humour was found in the abdominal cavity.

On February 17th I introduced in the same manner twenty spinal *Æstrus*-larvæ under the skin of an eighteen-months-old he-goat. So far as I am aware, the larvæ of *Hypoderma bovis* are not found in the goat.

On February 29th I found in the he-goat five subcutaneous swellings, each with a central opening, from which there exuded a dirty white fluid. I propose to allow these larvæ to mature.

I fed two young dogs simultaneously with spinal *Æstrus*-larvæ, introduced into the pharynx by means of a spatula. The dogs were killed after two and four days respectively. On autopsy I have been unable to find a single larva outside the alimentary tract or remains of the larvæ in the stomach or intestine.

I introduced ten larvæ directly into the œsophagus of a rabbit by means of a rubber tube. For three days I examined the fæces; but neither in them nor in the rabbit, which I afterwards killed, could I find any trace of the injected larvæ.

As it appeared from the autopsy of the dog, the larvæ introduced under the skin wandered about into various places agreeing with those in which they are found in cattle. This discovery, in connexion with the negative result produced by the introduction of the larvæ *per os*, causes me to incline to the opinion that the young larvæ of *Hypoderma bovis* at first pass beneath the skin, and from thence betake themselves to the spinal canal and other places, to return later into the subcutis and there undergo further development under the well-known conditions.

I hope later on to publish a communication as to the discovery of the further development of the spinal larvæ beneath the skin of the he-goat.

March, 1896.