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OTTAWA

HEALTH OF ANIMALS BRANCH

SPECIAL REPORT

ON

SARCOSPORIDIAE

AND THEIR ASSOCIATION WITH

“LOCO” DISEASE AND DOURINE

BY

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Assistant Pathologist

In charge Experimental Station, Lethbridge, Alberta.

Published by order of the Hon. SYDNEY A. FISHER, Minister of Agriculture, Ottawa, Ont.

October, 1908

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OTTAWA, October 16, 1908.

To the Honourable

The Minister of Agriculture.

SIR.—I beg to transmit herewith a bulletin, being a study of 'Sarcosporidiae' and their association with 'Loco' disease and Dourine. The investigation on this subject has been conducted by E. A. Watson, V.S., assistant pathologist in charge of the experimental station at Lethbridge, Alberta. I recommend that this be printed for distribution.

I have the honour to be, sir,

Your obedient servant,

J. G. RUTHERFORD,

Veterinary Director General.

SARCOSPORIDIOSIS.

A contribution on, with especial reference to its associations with 'Loco' disease and Dourine, and the possibility of mistaking the spores of Sarcocystis for certain so-called developmental forms of trypanosomata.

Although Sarcosporidiae are of common occurrence in the musculature of domesticated animals, very little is known concerning the life-history of the parasites, and nothing but supposition as to the means by which their hosts become infected. Further, a marked uncertainty or contradiction exists as to the power these parasites possess in producing a serious and recognizable disease.

It would appear that in the majority of cases of infection, in otherwise healthy animals, the invasion is a very limited and harmless one, involving to a more or less slight extent the muscles of the oesophagus and tongue, but that in certain conditions, generally of obscure origin, and briefly designated under the term 'cachectic', the parasites may overrun the entire muscular system and become a grave menace to the life of their hosts.

A brief resumé of the more important observations on the subject by various authors is here given:—

NOMENCLATURE.

- (a) Muscular Psorospermiosis.
- (b) The sarcocysts, tubes, sacs or utricles of Miescher.
- (c) Rainey's corpuscles (the spores of the cysts).
- (d) Sarcosporidiosis.

It is stated in Neumann's Parasites, that about 40 per cent of pigs may be infected. Moulé, quoted by the same authority, found the parasites in 98 per cent of cachectic sheep, usually numerous in proportion as the cachexia is more accentuated. In 100 sheep in good condition he met with them in 44, and then always in small numbers. Of 100 oxen condemned for being in extremely bad condition he found 37 infected with Sarcosporidiae. In cattle in good condition he found them only in three instances.

Schnitz noted their presence in the muscles of the forehand of a horse, destroyed on account of paralysis of the anterior limbs. Some similar examples are also recorded by Neumann. The following extracts are quoted from Minchin's² account of the Sarcosporidiae:—

'In acute cases all the skeletal muscles may be infected, even those of the head . . . the parasite grows until it distends the fibre to five or even ten times its normal breadth absorbing the contractile substance as it does so. . . . The cysts are observed to degenerate in some cases, their adventitious walls becoming calcified, in other cases the cysts burst and spread their contents in the surrounding tissues, destroying the muscles and producing tumors and abscesses.

'The symptoms of Sarcosporidiosis in the pig are paralysis of the hinder extremities, a skin eruption, and general symptoms of sickness, such as thirst, increased body-temperature, and dim, streaming eyes.

'The disease is sometimes the cause of fatal epizootics among domestic animals, especially sheep. In the mouse also Sarcocystis muris is a very deadly parasite. Laveran and Mesnil have isolated the toxine of the Sarcosporidian parasite of the sheep, and named it Sarcocystine, a substance found to be extremely toxic for the experimental rabbit.'

From some feeding experiments by Smith and others, quoted by Minchin it seems extremely probable that infection naturally occurs through the digestive tract. Sarcosporidiae have rarely been found in man; a few cases of fatal infection are mentioned by Neumann, and others by Bulloch.³

Ostertag⁴ does not consider the eating of flesh of infested animals dangerous to man. Neumann again thinks that infested meat should be prohibited for human use 'because of its bad appearance and diminished nutritive value.' According to the same author 'muscular Psorospermiosis is not betrayed during life by any appreciable signs and the parasites can be detected only at autopsy.'

The following cases have come under the writer's personal observation:—

(a) In Cattle Suspected of Loco-poisoning.

CASE I.—(Animal No. 27) heifer, 3 years old; said to have developed symptoms of loco-poisoning as a yearling.—(Dr. Hilton.) Received at quarantine station October 27, 1907; stunted, undersized appearance, and in poor flesh; coat staring and rough, on passing hand over the body numerous small scurfy nodules could be felt, knots of hair matted with moist, brain-like scales. There were frequent prolonged extensions of the head and neck, accompanied by a more or less constant trembling and agitation of the muscles of these parts, especially of the jaws and most marked during attempts at feeding, the power of the prehension of food being practically lost, and mastication very difficult and incomplete. Death occurred November 14, 1907.

Principal features of Autopsy.—Gelatinous infiltration, light yellow to an orange colour, of the connective tissues, lungs and kidneys. The inner covering of the brain, the pia, very dark, almost black in places, and of a metallic lustre. The heart enlarged, appearing soft and flabby, yet proving tough on section. The endocardium sprinkled with minute whitish granules, very numerous.

Microscopical.—Heart and its endocardium very extensively infested with Sarcosporidiae. Unfortunately, by the time these preparations had been examined and the parasites recognized, the carcass had been disposed of, and none of the skeletal muscles preserved. There was an immense deposition of greenish brown pigment granules in the covering of the brain and the cells of the cortex.

CASE II.—Heifer (animal No. 88). The symptoms and condition being so similar to Case I., they need not be further described, beyond adding that there was a thin watery discharge from the nostrils, dimmed eyes, and occasional lachrymation. After being under observation for three months, in which the disease made steady progress, until the animal became quite helpless, she was destroyed.

Principal lesions at autopsy.—Fibrinous inflammation of epicardium. Minute whitish specks showing through endocardium. The heart as a whole too voluminous.

Lungs.—Chronic, suppurative inflammation. Small sacs or cysts, about the size of a bean or small nut, and rather resembling lymph nodes, could be squeezed from the cut surface of the parenchyma. The appearance of the skeletal musculature slightly granular, cloudy and light brownish-red colour. The brain in this case did not reveal any abnormalities visible to the eye. The molar teeth were very irregular and badly developed, and the bones of jaws and face thin and brittle, though rather enlarged.

Microscopical.—The following muscles, organs or tissues were examined for Sarcosporidiae. (1) Psoas, (2) diaphragm, (3) tongue, (4) larynx, (5) upper and lower lips, (6) the gums, (7) the jaws, masseter and buccal, (8) the muscles of the poll, splenius and complexus, (9) of the eye, (10) of the limbs, the adductor magnus, flexor pedis perforatus, flexor metacarpi externus, (11) the heart, (12) the liver, (13) spleen, (14) kidneys, (15) lungs. In every specimen of every muscle examined,

the parasites were exceedingly numerous; Plate I., figs. 1 and 2, representing the degree of invasion. In the lungs the semi-purulent cysts consisted of leucocytes and disintegrating cellular tissues; a few Sarcosporozoa in process of decay were seen. It is extremely probable, but not proved, that these inflammatory areas in the lungs resulted from the presence and disintegration of Sarcocysts. In the spleen, liver and kidneys, a very few degenerating spores were seen and much granular pigment matter.

The myocardium contained very numerous parasites. While, as already stated, all of the skeletal muscles were extensively invaded, those of the head, poll and extremities were, if anything, the more heavily infested.

CASE III.—(Animal No. 83) Steer, 3 years old. Similar condition to preceding cases, but the disease not so far advanced. During the past three months, in which the animal had been hand-fed and well cared for, the symptoms have increased in severity. Depression is deepening, muscular action stiff and slow. Jaws swollen. The animal was cast and a small fragment of muscular tissue excised from the tongue, Flexor Metacarpi Externus, the masseter, and one the cervical muscles. Sarcosporozoa were found in preparations of each, not as numerous as in cases I. and II., but still fairly plentiful. The present condition of the animal leaves little chance for recovery.

CASES IV., V. and VI.—(Animals 84, 85 and 86) Steer and two heifers, two, three and two years old respectively. The condition is approximately the same in each, namely, slow, sluggish muscular action, depression, and a dejected, unthrifty appearance. These animals were cast and operated on as in Case III. The muscles examined, and the results as follows:—

Case IV.—Masseter—Sarcosporidiae numerous.

Upper cervical—Sarcosporidiae fairly numerous.

Tongue—Sarcosporidiae fairly numerous.

Case V.—Flexor metacarpi externus—Sarcosporidiae not found.

Middle cervical—Sarcosporidiae fairly numerous.

Tongue—Sarcosporidiae very numerous.

Case VI.—Lower cervical—Sarcosporidiae numerous.

Tongue—Sarcosporidiae numerous.

Thus in all muscles examined, with the single exception of a very small fragment of the flexor metacarpi, Sarcosporidiae were present.

(b) In the Equines Suspected of Loco-poisoning.

CASE VII.—(Animal No. 91.) Gelding, rising 3 years old showing symptoms of loco-poisoning as a yearling.—(Dr. Hilton.)

The condition, briefly, on admittance to the quarantine station, October 27, 1907, depression, restlessness; slowly wandering about without aim or object. Slow, high, hesitating gait, as if walking over obstacles. When undisturbed the head is carried very low, down to the level of the knees, the neck often twisted. When suddenly startled, there is a brief period of excitement and the animal may stagger and fall. At present date, after a period of five months, the condition has become greatly aggravated, and it does not look possible for the animal to live much longer. Flexion and extension of the limbs are extreme, and brought about very slowly, and it is quite impossible for the animal to run, trot or move out of a slow walk. The bones of the face appear much swollen, and the expression truly melancholic and pitiable. Small sections of the tongue and masseter muscle were removed and Sarcocysts were found in micro-sections of each. The parasites were few in number, and small, though containing spores.

CASE VIII.—(Animal No. 93.) Gelding, rising 3 years old, also showing symptoms of loco-poisoning as a yearling. On October 27, 1907, the condition was very fair, symptoms only slightly noticeable; but during the past five months the disease has greatly advanced until the present condition is quite as bad as the preceding case. A fragment of the flexor metacarpi externus muscle only has been examined. Sarcocysts were present, few in number, and in a young stage of growth. Previous to the finding of Sarcosporidiae in any of the foregoing cases, five loco-diseased equines had succumbed. The parasites not being suspected none of the skeletal muscles were microscopically examined. A portion of the myocardium was preserved in each case, in which no parasites were detected. A section of the tongue of one of these animals had been preserved, and in this Sarcosporidian cysts were found.

(c) In Dourine-affected Equines.

CASE IX.—(Filly, 9 months old.) Experimentally infected with *Trypanosoma Equiperdum*. In the fluid of a cutaneous swelling that followed the inoculations free spores of Sarcocysts together with trypanosomata were found, each species of parasite being present in about equal numbers.

CASE X.—(Filly, 2 years old.) Inoculated with the blood of a Dourined stallion. Seven months later, in the fluid of a cutaneous swelling, free Sarcospores were present, mostly crescentic forms, and indistinguishable from the crescents described by certain authors (see Plate II.) as developmental forms of Trypanosomata. Trypanosomes were not seen in this animal at this time.

CASE XI.—(Mare, 3 years old), in final stages of naturally acquired Dourine. Sarcospores were found free in the fluid of a swelling on two occasions, the swellings resulting from some serum-injection experiments.

(d) In a Cachetic Filly, the Cause of the Cachexia Not Known.

CASE XII.—(Filly, 2 years old), one of six supposedly healthy fillies shipped to quarantine station for experimental work. It was noted on arrival, that this animal (No. 68) had an unthrifty appearance, stiffened gait, rough hide and was in poor flesh. The mucous membranes were pale. She was several times carefully examined for signs of Dourine, but none were detected. The cachexia became more evident during the autumn and winter, with increasing stiffness of the muscle and gait, especially of the hinder extremities. The animal was found dead in the pasture on March 4th last, the body frozen.

The muscles of the oesophagus, tongue, and extensors of the fore-arm were infested with Sarcosporidiae. They were not seen in the myocardium or the muscle of the eye; none other were examined.

In cases I. to VIII. of Sarcosporidiosis in 'locoed' animals certain symptoms and conditions are described. These are more or less characteristic of 'loco-disease.' This disease itself is now under investigation at this station and will be the subject of a separate and later report, and is only mentioned here in those respects in which it may bear a possible relation to Sarcosporidiosis, or where the writer is unable to differentiate between the symptoms of the one and the other. Certainly, a great variety of conditions and symptoms occur in the course of the malady or maladies attributed to loco-poisoning. There appear various forms of mental derangement, from mania in the first stages to a dense stupor or comatose condition in the latter; disorders of the muscular system from increased reflexes to spasms, atrophy and paralysis; osseous enlargement or degeneration, especially of the jaws and parts of the skull; impaired vision; depravity, emaciation, &c. The history in most cases shows that the disease is very chronic, and often of a fluctuating or intermittent character. The muscles of the head and neck, especially of the jaws, the lips, and the tongue exhibit tremors, spasmodic contractions or prolonged after-contractions. The

jaws sometimes remain rigid, the head and neck extended. In equines cramps are not infrequent, a limb is slowly extended as far as possible, remaining in that position for a short time, then slowly and extremely flexed, held high above the ground for a minute or more, the animal moving off at a limp. The contractility of the muscles is delayed, slow, and hesitating, not arrested at the proper moment, resulting in over-flexion and extension. This might be explained by absorption of, pressure on, or breaks in the continuity of the protoplasm of the muscle fibres by the Sarcocysts, and consequently, a greatly reduced conductivity. It is equally true, that the origin of this muscular disturbance may be seated in the nerve cells of the brain, as a result of loco-poisoning. If the latter hypothesis is correct, it is a strange coincidence that in the few cases examined the muscles most affected were those in which the parasites were most numerous. It is difficult to trace any relationship between the disease of the osseous structures of the jaws and face and the disease of the muscles; and yet, as mentioned before, Laveran and Mesnil isolated an extremely active poison from the *Sarcocystis-tenella* of the sheep (unfortunately, the original publication by these savants is not a hand) and it is permissible to assume that a similar toxin is elaborated by the parasites infesting the ox and horse. Sarcosporidiae appear more numerous in 'locoed' oxen than in horses, in the latter intestinal parasites are exceedingly plentiful, and very formidable-looking verminous aneurisms have been found at each autopsy. The condition favourable to these parasitic invasions may very likely originate from ingestion of the loco-weed, and possibly to some other dietetic conditions peculiar to the ranges where loco-disease prevails. It might be mentioned in passing that the writer has on several occasions examined specimens of meat, purchased from a local butcher, for Sarcosporidiae. In all, three ox-hearts, an ox-tongue, and several joints of beef have been examined, each specimen being from a different animal. Of these only one was found infected with Sarcosporidiae, a heart, the parasites being very numerous. This heart looked decidedly unhealthy. There were a few fibrinous growths on the epicardium, considerable gelatinous oedema, and the little fat remaining, very soft and yellow.

Concerning the Parasite, *Sarcocystis*.

Class—Sporozoa; sub-class—Neosporidiae; Order—Sarcosporidiae; Genus—*Sarcocystis*, Ray Lankester. (After Minchin.) Infesting domestic animals there are probably several distinct species of *Sarcocystis*, but these have not been clearly differentiated.

In those infections of the horse and ox, already described, and even in the most heavily infested cases, very young forms of parasites could never be found; only the trophic phase was met with, accompanied by spore-production, as in all of the sub-class of Neosporidiae. In cattle the youngest trophozoites observed were lodged in the muscle fibre, and, though microscopically invisible, had attained a considerable size and formed several hundred spores. The larger parasites, distending the muscle fibres, are just visible to the naked eye as minute whitish rods or specks. In the equines, Cases VII. and VIII., the parasites averaged a much smaller size than in the bovines, the smaller trophozoites containing only the spore mother-cells or pansporoblasts, preliminary to sporulation. In these young forms the radially striated enveloping membrane is well marked (Plate I., fig. 3); as the parasite matures the membrane becomes thinner and the striations disappear, until, in the largest forms as seen in cattle, the parasite appears to be held only by the sarcolemma of the muscle fibre. Evidently the parasites are able to multiply within the body of their host until every part of the muscular system is invaded; this endogenous mode of infection is hypothetically brought about by rupture of the cysts and dissemination of the spores, and yet, if this is actually the case it is strange that even in such severely infected cases as I. and II., no intermediate forms between the relatively minute spore and large young trophozoite could be found.

The spores themselves, the clamydospores, vary considerably in shape, size, and

in the arrangement of their chromatin and polar capsule. In form they appear in all gradations from a perfect sphere to an oval, bean-shaped or crescent form, though each type is symmetrical and doubtless represents different stages in the development of the spore-cell. The spherical forms would appear to be the younger and the crescentic the older, as in each trophozoite containing many spores, some are found disintegrating and these are almost invariably of a crescentic variety.

The chromatin granules of the nucleus of the spherical spore are relatively small and scattered, and the polar capsule is granular and only partially developed; in the next stages, the chromatin collects into small bodies, and one end of the cell becomes pointed, and at this end is always found the polar capsule. This capsule stains with difficulty with most dyes, and not at all with the ordinary basic stains; even with some of the Romanowsky preparations only a faint pink ectoplast at one end of the spore, and where the protoplasm has receded, denotes the position of the capsule (see Plate II., fig. 3, No. 6). The Romanowsky stains, however, are preferable to any other, and, properly prepared, differentiate very clearly and beautifully the structures of the cell. A single granule, staining black, and, more rarely a double granule, may occasionally be seen, situated about half-way between the nucleus and the pointed extremity; this is doubtless the centrosome. (Plate II., fig. 7—the end spore, fig. 3, Nos. 1 and 6). In the older spores the nuclear chromatin collects into round or irregular bodies, eight to sixteen in number. The frequent finding of a large spherical spore and a crescent lying intimately, and in some cases apparently fused, together, is suggestive that the former is the female cell and the latter the male gamete (Plate II., fig. 3, No. 5). A large spherical form, probably resulting from the fusion of these two elements, may be seen dividing directly into two, three or four. (Plate II., figs. 2 and 3).

The possibility of mistaking the spores of *Sarcocystis* for developmental forms of *Trypanosomata*.

From time to time mention is made by various workers, of large 'crescentic bodies' or 'vermicule-like forms' occurring in the blood of animals affected with trypanosomiasis, and are generally described as developmental stages in the life-history of a species of trypanosomata.

Holmes, in an article entitled 'Trypanosomiasis among cattle in India,' states, 'In the blood taken from two infected bulls I found peculiar developing forms bearing a striking resemblance to the crescents of human tertian fever. These are large crescent-shaped bodies staining blue and containing numerous chromatin granules.' Some of these forms of trypanosomata (?) are illustrated by author.

Lingard⁶ in 'Species of trypanosomata observed in Bovines in India' mentions the above observations of Holmes, and further describes similar forms met with by him (Lingard), also illustrating them in coloured plates (Plate II., fig. 20, and Plate III., fig. 7, *Journal Tropical & Science*, Vol. II., No. 7, 1907), stating 'These crescent forms, above described, are frequently co-existent with the piroplasma bigeminum, or the smaller forms in the blood of Indian cattle. Possibly the presence of the crescents in Queensland bovines may point to the fact that these animals in some instances are also the host of a large form of trypanosoma, which up to the present has not been demonstrated in their blood.' Hunt, quoted by Minchin (page 269) and also by Lingard, 'found crescents in the blood of cattle, and observed their change into a spheriodical shape, but while comparing these bodies to the crescents of malarial parasites, he at the same time regards them as a form of sporulating body, producing spores endogenously.' Martin in criticizing the observations of Holmes states, 'Raymond also afterwards found them (i.e., the crescents) in the cattle of Bengal, and he is inclined to the opinion that they have nothing to do either with trypanosomiasis or piroplasmiasis, but that they are really *Sarcosporidia*.' The writer ventures to express a similar opinion. At first, in finding them in the cutaneous swellings of Dourine-affected equines, side by side with mature and unmistakable trypanosomes,

PLATE I.

Fig. 1.

Fig. 2.

Gd. S. H. E.A.W.

Fig. 3.

PLATE II.

Fig. 1.

Fig. 2.

Fig. 3.

Fig. 4.

Fig. 5.

and after a careful comparison with the illustrations and descriptions furnished by Holmes and Lingard already referred to, and to which they appeared identical, he was inclined to consider them as developmental forms of trypanosomata. But subsequently, when positively no difference could be detected between these forms and certain of the spores of *Sarcocystis* isolated from cases I. to VIII. and case XII., he (the writer) became convinced that they belong to the latter genus of a parasite, and excludes any relationship between these crescents and trypanosomata. Certainly, such forms of the spores of *Sarcocystis* as are here depicted, Plate II., fig. 5, Nos. 2 and 6 and some others, may very easily be mistaken for trypanosomes in which the flagella and undulating membrane have been cast off, for 'stumpy,' swollen forms. It is of great importance, therefore, in diagnosing such a serious infection as Trypanosomiasis from blood specimens to remember the possibility of the presence of *Sarcosporidia* and their likeness to questionable forms of trypanosomata.

Conclusions.

1. The parasite *Sarcocystis* under certain conditions becomes a very important factor in disease, invading the entire musculature of their hosts, with serious or fatal consequences.

2. *Sarcosporidiosis* may be closely associated with, and is probably a very frequent sequel to, the disease of horses and cattle known as 'Loco-disease.' It may complicate the diagnosis of this disease, and also of Dourine, and probably of some others, and retard or prevent recovery from these and similar cachectic conditions.

3. The crescentic spores of *Sarcocystis* bear a striking resemblance to 'Crescentic-bodies' that have been described as developmental forms of trypanosomata, and it would be unsafe, or quite erroneous to diagnose an infection by the latter from the presence alone of those crescentic bodies.

4. The *Sarcosporidia* are deserving of more detail study and investigation than has hitherto been accorded them, both from a zoological and a pathological standpoint.

EXPLANATION OF PLATES.

PLATE I.

Fig. 1.—Cross-section of bundle of muscle fibres infected with *Sarcocysts* (from splenius muscle of heifer, Case II., hamatoxylin and picric acid. Leitz objective No. 3 Ocular No. 3.

Fig. 2.—Longitudinal section of the same.

Fig. 3.—Young parasites showing radially striated envelope, and pansporoblasts from flexor metacarpi externus muscle of a horse, Case III. Objective No. 6, Ocular No. 3.

PLATE II.

Showing the spores of *Sarcocystis*, $\frac{1}{2}$ oil immersion, ocular No. 3.

Fig. 1.—Variations in size, shape nuclear arrangement, and formation of polar capsule.

Fig. 2.—Nos. 1-4, and fig. 3, Nos. 1-3. Dividing forms. Note the centrosomes in fig. 3, No. 1.

Fig. 3. No. 5.—Gametes or Zygote formation (?).

Fig. 3. No. 6.—An example of spores more lightly stained, and showing only the position of the polar capsule.

Fig. 4. Nos. 1-6.—Constriction of the protoplasm, and extended bodies (†), No. 4, suggesting that the pointed extremity of the crescent spore had penetrated one of these bodies.

Fig. 1-4.—Are taken from Sarcosporidiae of the ox.

Fig. 5.—Represents types of spores found free in the fluid of cutaneous swellings in dourine-affected equines.

All figures in Plate II. are stained by the writer's modification of the Romanowsky method, prepared as follows:—A polychrome blue is made by one per cent solution of medicinal methylene blue in a one-half per cent aqueous solution of sodium bicarbonate, heating and evaporating to dryness over a water bath. The residue is powdered and two solutions then prepared—

A.—35 ccm. B. 4 ccm. in 16 ccm. of methyl alcohol.

B. 0.5 per cent eosin in methyl alcohol and mixed in the following proportions:—

A.—35 ccm. B. 4 ccm. in 16 ccm. of methyl alcohol.

This solution is slightly alkaline (very useful for some purposes) for the study of *Hæmatozoa* and *Sarcosporidiae*. A drop or two of an exceedingly dilute solution of acetic acid in alcohol should be added, care being taken not to over neutralize, and the stain applied as in Leishmann's and other alcoholic Romanowsky stains.

The proportions of A and B may be varied slightly increasing the amount of Eosin according to the reaction desired, and the stain may also be further diluted with methyl alcohol to advantage.

References.

1. Numann, L. G. *Parasites and parasitic diseases of domesticated animals*. Second edition, Macqueen p. 555-561—1905.
2. Minchin, E. A.—*Sporozoa*.—Treatise on zoology. Edited by Ray Lankester. Part I—1903.
3. Bulloch, W.—*Psorospermiosis*. *System of Medicine*. Edited by Allbutt and Rolleston, Vol. II., Part II, p. 828—1907.
4. Ostertag, R.—*Handbook of Meat Inspection*, translated by E. V. Wilcox. Second edition—1905.
5. Holmes, J. D. E.—*Trypanosomiasis among cattle in India*. *Jour. Comp. Path. and Therap.* Vol. XVII., p. 323—1904.
6. Lingard, A.—*Trypanosomata observed in Bovines in India*. *Jour. Tropical Vet. Science*, Vol. II., No. 1, p. 43—1907.
7. Martin, E. E.—*Trypanosomiasis in cattle of India*. *Jour. Comp. Path. and Therap.* Vol. XVIII., p. 145—1905.