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By G. DE KOCK, M.R.C.V.S., Dr.Med.Vet., and P. J. J. FOURIE, M.R.C.V.S., Research Officers, Onderstepoort.

Necrosis in Musculature and Myocard of Sheep—Pseudo-tuberculosis.

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Kitt (1), describes in bovines under the name of "Pseudo-tuberculose des Fleisches," peculiar nodules in the panniculus, the muscles of the abdomen, shoulder, leg, and the tail. The nodules vary in size from a millet seed to that of a hazel-nut. The nodules are of a caseous nature and microscopically they resemble the lesions of tuberculosis.

Microscopically the lesions consist of central necrotic masses in which are groups of abscess-like round cell accumulations. "(Multiple abszessartige Rundzellen-ansammlungen)." This is surrounded by a fibrous and fibroblastic capsule. Histologically these lesions are therefore unlike tuberculosis.

Kitt further states that, because animals affected with pseudotuberculosis had in many cases also typical tuberculosis of lymphatic glands and intestines, may simply be coincidental, and that tuberculosis and some other disease are occurring simultaneously in one and the same animal. He mentions the possibility, however, that the lesions described as pseudotuberculous may be true tubercles, in which the organisms have died out and have lost their staining properties, seeing that giant cells and a coagulative necrosis like those occurring in tuberculosis are occasionally present. Subcutaneous inoculation in cattle with ground up nodules always gave negative results in his experiments (observed for months).

Occasionally badly stained groups of bacilli resembling the bacillus pyogenis bovis were found in the nodules. Hungerbühler could in one case obtain cultures of this organism which was pyogenic

for small animals.

Kitt points out that the lesions are described as Blastomycosis in the literature without sufficient evidence.

Hutyra and Marek (2) state that, according to Glässer, homogeneous caseous nodules resembling muscle tuberculosis are due to

bacteria of the coli-typhoid group in cattle.

The same authors in the first volume of the above work discuss under Pseudotuberculosis, all disease processes which resemble tuberculosis anatomically, but which are caused by parasites and organisms, other than the acid-fast tubercle bacillus of Koch. They mention that Preisz suggested that only such disease processes that are caused by bacteria, other than acid-fast organisms should be so designated. Numerous organisms are mentioned as the cause of pseudotuberculosis in the various animals. The only organism associated with pseudotuberculosis of sheep is the bacillus pseudotuberculosis ovis of Preisz, otherwise known as the Preisz-nocard organism.

The authors describe the pseudotubercle in connection with the serous membranes, the subcutaneous tissues, and in the organs, as resembling the true tubercle macroscopically, but from which it can be differentiated microscopically. The pseudotubercle is of an exudative nature (neutrophiles) as against the proliferating nature (tuberculous granulation tissue) of the true tubercle. Necrosis occurs in the later stages of true tubercle formation. The pseudotubercle develops more rapidly, consists throughout of necrotic detritus surrounded by a narrow peripheral layer of round cells; and necrosis and caseation is an early feature in its development. It contains neither giant cells nor epithelioid cells.

In speaking more particularly of pseudotuberculosis of sheep, the authors decribe the well-known macroscopic lesions in lymphatic glands, lungs, liver, kidneys, spleen, etc., and further mention that "at times caseous purulent foci, which may vary up to the size of a fist, may be found accidentally in slaughtered animals in the muscles, as for instance, in the muscles of the thigh."

Joest (3) describes the lesions caused by the bac. pseudotuberculosis ovis (Preisz-nocard organism). The young lesions consist of neutrophiles, very soon necrosis occurs and a central structureless mass remains, at the periphery of which there may still be a narrow zone, in which cells are recognised. These are of two kinds, (1) neutrophiles, and (2) macrophages. The whole lesion is surrounded by a fibrous and fibroblastic capsule. The necrotic portions become calcified, and this is often seen in concentric layers.

Ostertag (4) describes in addition to the usual situations also caseous nodules of pseudotuberculosis in the musculature. He states further that the only thing that the pseudotubercle has in common with the true tubercle is caseation, and that in the former, there are never giant cells or epithelioid cells present. Ostertag maintains that there is no calcification in the pseudotubercle, but as a result of inspissation the arrangement of concentric layers is produced.

The first case (specimen 4826) that came under our observation was sent in from the Municipal Abattoirs, Johannesburg. It consists of the forearm of a sheep—see plate I. Lesions are present in the flexors and extensors of the forearm, and that portion of the triceps inserted at the olecranon process. The lesions are in the form of small foci varying in size from 2 by 1 mm. to 4 by 1 mm. They are spindle-shaped, and have the long axis in the same direction as that of the muscle fibres. They are greyish white in colour and opaque. They are irregularly distributed throughout the muscle substance, but in places they have a linear arrangement. The rest of the muscle substance is normal as regards colour and transparency.

Microscopically.—The nodules vary in size and age. In the younger nodules (plate II, fig. 1) calcified particles are present towards the centre, around these are giant cells and it seems as if these are attempting to phagocyte the calcified particles.

The nodules show as yet no other evidence of necrosis, they consist mainly of epithelioid cells which have a peculiar spherical appearance, and in many of them the nuclei are situated towards the periphery of the cells. This is nearly always true of epithelioid cells having two and more nuclei, but is also true of some of those having only one nucleus. Although the nodules consist mainly of epithelioid cells, quite a number of round cells and infrequently also

neutrophiles occur in an irregular manner amongst the epithelioid cells. Towards the periphery of the young nodule round cells are more frequent. Here also the granulation tissue is fibroblastic, but in places already well formed fibrous tissue is present forming a well-

defined capsule.

The older nodules vary somewhat in their microscopic appearance—(1) see plate II, figs. 3 and 4. These show the presence of fairly extensive central areas which are completely calcified and the homogeneous pink staining mass so typical of necrosis of the epithelioid cells in the tubercle is not present. Around the calcified masses are numerous giant cells and in places giant cells are seen to have phagocyted fairly large calcified particles. The impression that one has when examining such a nodule is that the calcification was not preceded by necrosis of the epithelioid cells. The pheripheral portions consist of epithelioid cells as described for the young nodule, and in places beautiful examples of giant cells that are indistinguishable from the Langhans giant cells. The whole is surrounded by a fibroblastic, but mainly fibrous capsule. (2) See plate II, fig. 2. These nodules show in addition the presence of a homogeneous pink staining zone around the central calcified area. This is due to necrosis of epithelioid cells. Such nodules are less frequently met with than those described under (1).

No acid-fast organisms could be demonstrated by the Ziehl-Neelson's method of staining. Neither was it possible to demonstrate the presence of other organisms by staining with methylene blue or

Giemsa.

the abattoirs were forwarded.

Nodules are also described in the myocard of the sheep but this material was unfortunately not available for microscopic examination.

The second case that was investigated was sent to this Institution by the Chief Sanitary Inspector, Uitenhage, C.P. Specimens (6124) of the heart and of striped muscle of a sheep that was slaughtered at

The animal was in good condition, but numerous nodules were described in the musculature and in the heart. The chief sanitary inspector's report of the examination of the rest of the carcase is:—lungs, spleen, pancreas, stomachs and intestine normal. The liver contained a degenerated echinococcus cyst. The following lymphatic glands were carefully examined, and all were found to be normal: submaxiallary, subparotid, bronchial, mediastinal, periportal, prescapular, renal, iliac, precrural, inguinal, and mesenteric.

Macroscopically. Striped Muscle (not identified).—Only a small piece is available for description and examination. Several nodules are present, some of which are embedded deeply in the substance of the muscle, others are superficially placed and are raised above the surface of the muscle. The nodules measure up to 7 mm. in diameter; they are circumscribed and rather sharply marked off from the surrounding muscle substance. They are surrounded by a well formed capsule. They are opaque and of a pale greyish white colour. The central portions can be easily removed and consist of a caseous and a calcareous substance.

Myocard.—Similar nodules are present. See plate IV, fig 1. Some are situated superficially and cause a bulging of the epicardium. Others are embedded in the substance of the myocardium, and a few are situated towards the endocard.

Microscopically. Myocard.—The nodules are composed of cells which are for the most part well preserved. In a few nodules central necrotic areas are present, and this appears as a homogeneous structureless mass (plate III). Around this is a zone of epithelioid cells, amongst which the remains of muscle fibres can still be recognized. Infrequently giant cells, which are not very characteristic, are present at the periphery of the caseous mass. The fibroblastic and fibrous tissues at the periphery of the nodules do not form a well-defined capsule. In some portions of the perhiphery of the nodule, there may be a great deal of this tissue, whilst in other portions very little. The whole nodule is therefore not sharply marked off from the remaining tissues. In the immediate vicinity of the nodules there is very marked atrophy of the muscle fibres and in places vessels are seen in cross-section literally choked with round cells. Amongst the epithelioid cells are also very numerous round cells.

In places the myocard shows exudative changes of a diffuse nature. The exudate is entirely cellular and consists mainly of round cells, but quite numerous neutrophiles are also present. The leucocytic infiltration is present along the interstitium and appar-

ently also in the parenchyma of the organ (plate IV, fig. 2).

Striped Muscle.—The nodules are well defined and circumscribed (plate III). Centrally there is a comparatively small area in which calcification has occurred. This is surrounded by a wide zone which is structureless and of a homogeneous nature, but nuclear debris and nuclei in varying stages of disintegration can still be recognized in this zone. This is surrounded by a zone of epithelioid cells, amongst which are also round cells. Giant cells are present at the periphery of the necrotic zone and also in the epithelioid zone, some are like foreign body giant cells, but others are indistinguishable from Langhans giant cells. In the case of the older nodules there is a well developed fibrous capsule; in the younger nodules this consists of granulation tissue.

No tubercle bacilli could be demonstrated in the myocard or the striped muscle by Ziehl-Neelson's method of staining, neither could other bacteria be demonstrated with Giemsa and methylene blue

staining.

Discussion.—The lesions are precisely like those caused by the Koch's tubercle bacillus, but these organisms could not be demonstrated in sections by the usual methods. It was unfortunate that no material was available for a bacteriological examination. But there seems to be very little doubt that the second sheep (specimen 6124)

could not possibly have had tuberculosis.

The chief sanitary inspector's examination shows clearly the normal state of the lungs and the important lymphatic glands. An animal that has numerous tubercle nodules in the musculature and the heart must have had a generalization of the disease, and it is inconceivable that such lesions in the musculature and myocard can exist without any lesion in the lungs and the lymphatic glands of the head and neck, or the bronchial and mediastinal lymphatic glands, or any lesions in the intestines and the mesenteric and other lymphatic glands.

Most authors are agreed that the lesions of the so-called "pseudotuberculosis" resemble the true tuberculosis macroscopically, but can be differentiated from the latter microscopically.

Kitt, however, described lesions in bovines that were undoubtedly

also affected with tuberculosis. In some of these lesions he found epithelioid cells and giant cells, but could not demonstrate Koch's tubercle bacilli bacteriologically or biologically. He believes that some of them may indeed be true tubercles, in which the organisms have died out, but would, on the other hand, also appear to commit himself in regarding some of these lesions as of the nature of pseudotuberculosis, caused by the bacillus pyogenes bovis. When dealing with bovines one is not on such safe ground as far as tuberculosis is In sheep, and especially South African sheep, tuberculosis can be definitely excluded, especially as no lesions were found in the lungs and the lymphatic glands.

It is therefore quite clear that we are dealing here with lesions which are not in all cases absolutely like tuberculosis macroscopically, but which are precisely like those of tuberculosis microscopically.

Dunkel believes that the bacillus pyogenes bovis, the bacillus pyogenes suis and the bacillus pseudotuberculosis ovis are varieties of one and the same species, because they are reciprocally agglutinated by their respective immune sera, because mice after treatment with the one bacillus become immune to the other, and further because the bacillus pyogenes after being passed through the rabbit, assumes the appearance of the bacillus pseudotuberculosis ovis when inoculated into sheep.

No material was available for cultural or biological examination, but it would appear that these lesions in the sheep are analogous to those described by Kitt in bovines as being due to the bacillus pyogenes bovis in which histologically the structure of the true tubercle was also present.

It is therefore possible that these lesions in the sheep were caused by the bacillus pseudotuberculosis ovis and if further material becomes available an attempt will be made to prove this culturally and biologically.

Conclusions.—Lesions are described in the myocardium and musculature of South African sheep, which are not in all cases absolutely like those of tuberculosis macroscopically, but most of which are peculiarly like those of tuberculosis microscopically. reasons given in the text we believe that these lesions are not those of ordinary tuberculosis. Unfortunately no material was available to investigate the cause bacteriologically or biologically.

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