

THE PATHOLOGY OF BACTERIAL INFECTION OF THE GENITALIA IN RAMS

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ABSTRACT

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Details are given of the macroscopic and histopathological changes brought about by infection of the genitalia of rams by bacteria other than *Brucella ovis*. Lesions of the vesiculae seminales and ampullae are described which, in addition to the clinically evident lesions of the testes and epididymides, could be an important reason for impaired fertility.

The name "bacterial infection of the genitalia", abbreviated to BIG, is suggested as a more appropriate designation for this condition than "ram epididymitis".

Résumé

LA PATHOLOGIE DE L'INFECTION BACTÉRIENNE DES ORGANES REPRODUCTEURS CHEZ LE BÉLIER

Des détails des lésions macroscopiques et histopathologiques apportés par l'infection des organes reproducteurs des béliers par des bactéries autres que le *Brucella ovis* sont procurés. Des lésions des vesicules seminales sont décrites qui, en plus des lésions cliniquement évidentes des testes et des epididymes pourraient être une importante raison de l'affaiblissement de la fertilité.

Le nom de "Infection bactérienne des organes reproducteurs", en abréviation BIG, est suggéré comme désignation mieux appropriée pour cette condition que: "Epididymite du bélier".

INTRODUCTION

There was a time when *Brucella ovis* was regarded as the main, if not the only, cause of ram epididymitis. Of the many publications that have appeared on this subject, those by Kennedy, Frazier & McGowan (1956) and by Biberstein, McGowan, Olander & Kennedy (1964) give the most detailed account of the pathological changes brought about by the organism. But the significance of the infection has diminished markedly since it was proved that vaccination against *Br. ovis* is effective. Van Drimmelen (1960) showed that a live vaccine prepared from the strain Elberg Rev 1 *Br. melitensis* rendered sheep sufficiently immune to withstand challenge under practical conditions. McGowan (1979) showed that ram epididymitis caused by *Br. ovis* can be effectively controlled.

Although ram epididymitis caused by *Br. ovis* had been brought under control, the continued incidence of the clinical condition was reported from many sheep-breeding districts (Van Tonder, 1977; Watt, 1978; Delong, Waldhalm & Hall, 1979). A large variety of organisms have been isolated from the genital tracts of rams affected by epididymitis not caused by *Br. ovis*. Delong *et al.* (1979), for instance, recovered *Actinobacillus actinomycetemcomitans*, *Staphylococcus* spp., *Corynebacterium pseudotuberculosis* and *Pseudomonas maltophilia* from such rams. Watt (1978) examined the scrotal contents of 2 281 Merino rams and found that 40% showed 1 or more gross lesions. *Corynebacterium* spp. were the most frequent isolates from these lesions, *Actinobacillus seminis* was isolated once and *Br. ovis* not at all. Ekdahl, Money & Martin (1968) isolated a number of different organisms from the testes and epididymides of rams in New Zealand. The following bacteria other than *Br. ovis* are listed in the publications of these workers: Gram-negative pleomorph, *Actinobacillus*-like organisms, *C. pyogenes*, *C. ovis*, *Streptococcus* spp., *Staphylococcus* spp., *Pasteurella haemolytica*, *Pasteurella multocida*, *Pasteurella pseudotuberculosis*, *Bacteroides*, *Br. abortus* and *Br. abortus* (S19).

Jansen (1980), investigating the distribution of infection in the genitalia of South African rams, found, among a variety of organisms, *Pasteurella haemolytica*, *Staphylococcus* spp., *Corynebacterium pseudotuberculosis*, *C. paurometabolicum*, *Micrococcus luteus*, *M.*

varians, *Streptobacillus* sp., *Flavobacterium*, *Streptococcus bovis*, various *Enterobacteriae* and *Pseudomonas* sp.

A detailed study of the pathological changes effected by these organisms similar to that undertaken with respect to *Br. ovis* infection by Kennedy *et al.* (1978) and Biberstein *et al.* (1964), seemed essential to assess the significance of these organisms as causes of infertility. In planning to do this I had to take note of what had already been done in this respect. Jamieson & Soltys (1947) described the gross pathological changes and pathological histology of the testes and epididymides of rams brought about by *Pasteurella pseudotuberculosis*. Dodd & Hartley (1955) described the changes seen in epididymal abscessation caused by a gram-negative bipolar staining pleomorphic bacillus. Claxton & Everett (1966) referred to the macroscopic and histopathological changes in the testes and epididymides of a ram infected with *Histophilus ovis*. Watt (1978) reported on the gross lesions and histopathological changes in the testes of rams infected with *C. ovis*, *C. pyogenes* and *A. seminis*.

The present study was aimed at studying the lesions of the testis and epididymis in greater detail and investigating the changes brought about by infection in the sections of the genital tract other than the testis and epididymis.

MATERIALS AND METHODS

Experimental animals

One hundred rams were studied. All the lesions studied were from natural cases. Rams of the Merino and Dorper breeds, culled on account of palpable lesions of the external genitalia, the presence of neutrophils and bacteria in their semen, and/or poor breeding performance, were made available from studs distributed over the whole of the Republic of South Africa. Only rams from flocks that had been regularly vaccinated against *Br. ovis* infection were selected for study and they were furthermore tested for the absence of *Br. ovis* infection by a cultural examination of their semen.

The rams were slaughtered in an abattoir and their entire genito-urinary tract, except the kidneys, was removed and transferred to a laboratory some 100 m away for further examination.

Examination of tissues

A detailed bacteriological examination was carried out on the different organs composing the genital tract, and the bacterial species isolated from the different organs were recorded in tabular form (Jansen, 1980). Specimens for histopathological examination were taken from all the organs and fixed in 10% formalin buffered at pH 7.0. Sections were prepared from the organs yielding positive bacteriological cultures and from those obviously affected in spite of negative culture results.

The Haematoxylin-eosin staining method was applied routinely. Semen smears were prepared and stained in the manner described by Van Tonder (1977).

RESULTS

Clinical changes

Rams affected by acute orchitis and epididymitis showed a painful, oedematous swelling of the scrotum, usually more pronounced unilaterally. On palpation the scrotal contents were firm, immovable and covered by a greatly thickened layer comprising the tunicae and the skin.

In some of these cases the scrotum had ruptured and formed a fistula which discharged thick, purulent material.

In the chronic stage the rams showed hard lumps up to the size of a golf ball localized in the cauda or caput epididymidis or even in the cord immediately above the testes. In these instances the testis on the affected side was usually markedly reduced in size.

Microscopic changes

When at autopsy the acutely affected scrotum was incised, the tunica vaginalis and subcutis were found to be markedly oedematous and congested. The tunica albuginea appeared thickened and congested, and was sometimes covered with fibrinous deposits.

The testis was usually normal in size and the epididymis was sometimes enlarged and firm.

The chronic cases at autopsy showed adhesions and gross fibrosis of the parietal and visceral layers of the tunica vaginalis. When the testis had ruptured, its substance showed fistulous cavities with thickened walls and one or more openings to the outside. Usually, however, the testis itself, though rarely directly affected by the suppurative process, was smaller than usual and had a thickened tunica albuginea. The cut surface showed strands of fibrous tissue and sometimes areas of calcification.

When examined post-mortem, the lumps or enlargements that could be detected on clinical examination in the cauda or caput epididymidis or above the testis were of 2 different types. Some were thick-walled, fluctuating masses which liberated a smooth, greyish purulent material on incision. Others, which frequently affected the cauda epididymidis, cut with increased resistance and on the cut surface this resistance could be seen to be caused by a more or less uniform increase in dense, white connective tissue and thickening of the overlying tunica albuginea.

In some of the rams either one or both vesiculae seminales were enlarged and had an exaggerated lobulated appearance. In many rams the vesiculae seminales and ampullae appeared normal in spite of the isolation of bacteria from them.

Microscopic changes

The incidence of microscopic changes in the different parts of the genital tracts of the 100 rams examined is given in Table 1.

TABLE 1 The incidence of histopathological lesions in the genitalia of rams

Organ	Number affected
Sheath.....	79
Vesicula seminalis.....	60
Ampulla	63
Epididymis.....	42
Testis.....	28

No bacteria were discernible in the sections.

Not all organs from which bacteria were isolated showed histopathological changes. On the other hand bacteria were always isolated from either the accessory glands or epididymides or testes or from all of them in rams showing pathological changes in one or more of the component parts of their genitalia.

It was impossible to associate a particular histopathological lesion with a single bacterial species, because in many instances the same type of lesion was caused by different bacteria. Also, more than one type of organism was often isolated from an affected organ (Jansen, 1980).

Sheath

The mucosa of the prepuce showed moderate activation of lymphoid follicles with an accumulation of lymphoblasts in the germinal centres. There was also a marked infiltration of lymphocytes and plasma cells into the subepithelial tissue, the lymphocytes being more concentrated in the perivascular areas.

Small focal areas of degeneration and necrosis of the epithelial cells were evident.

Ampulla

A variety of pathological changes were seen in the epithelium and wall of the ampulla of the vas deferens.

In the normal epithelium there were focal areas of necrosis, characterized by cytolysis and pyknosis (Fig. 1). Close to these areas accumulations of desquamated cells with pyknotic nuclei were present in the lumen.

There were accumulations of neutrophils in the lumen of the sac-like dilatations formed by the folds of the epithelium. Neutrophils were also present among the epithelial cells and in some areas pockets of neutrophils formed in the epithelium (Fig. 1). They were also distributed among the spermatozoa in the lumen of the ampulla.

In the specimens derived from some rams there were large accumulations of plasma cells in the lamina propria of the septa (Fig. 1). Lymphocytes were present among the plasma cells. Foci of plasmablasts were evident and the series of changes to plasma cells could be followed from the centre to the periphery of these foci.

Vesicula seminalis

Neutrophils were distributed among the cells of the lamina propria of the septa and in the epithelium itself.

Accumulations of these cells were also present in the lumen of the saccular dilatations formed by the epithelium.

Some specimens showed accumulations of plasma cells and some lymphocytes in the lamina propria of the septa (Fig. 2). Others showed the infiltration of predominantly lymphocytes into the lamina propria of

the septa of epithelium as well as accumulations of lymphocytes in the lumen of the saccular dilatations formed by the epithelium.

The epithelium in some sections showed foci of hyperplasia with loss of its normal structure (Fig. 3). In others, it also showed areas of karyolysis and pyknosis of epithelial cells (Fig. 4).

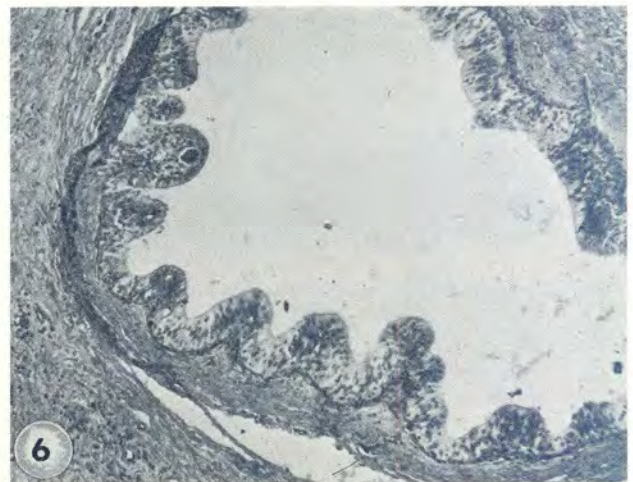
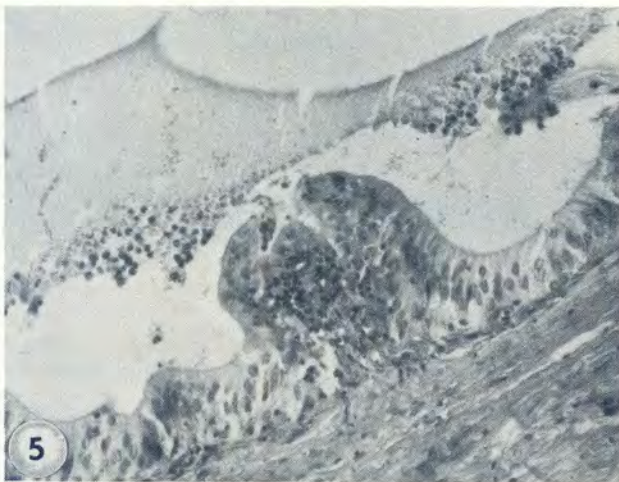
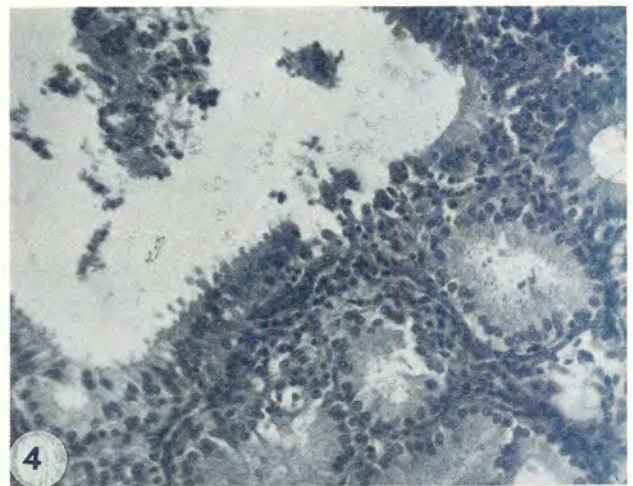
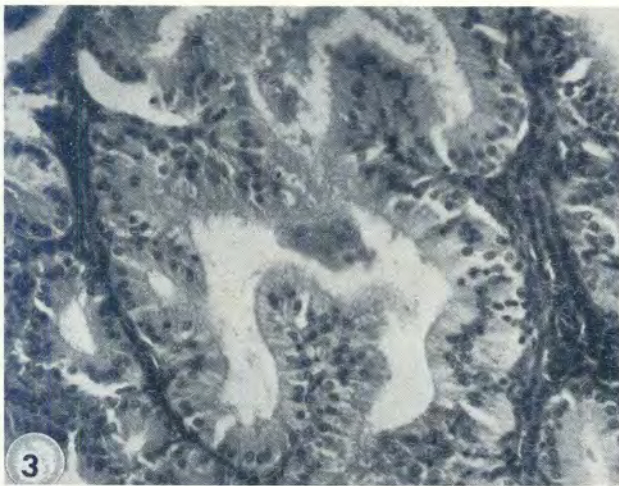
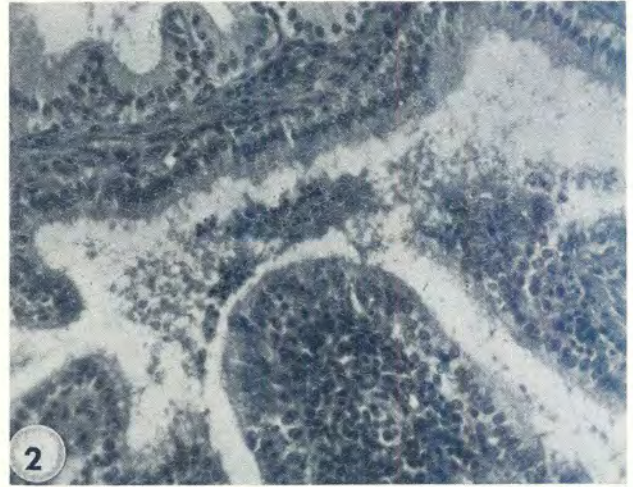
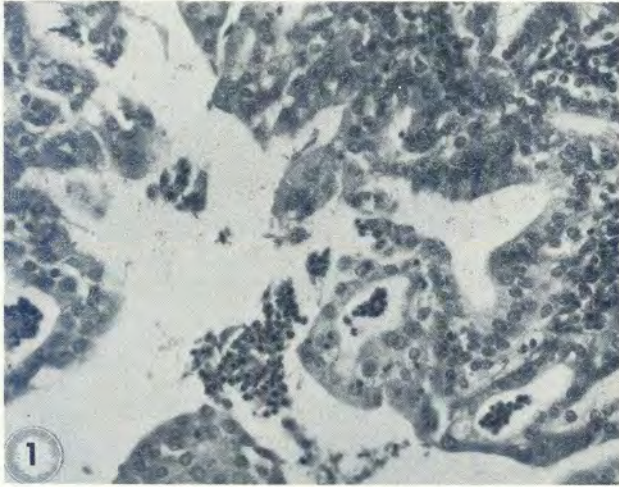


FIG. 1 Foci of necrosis in epithelium of ampulla. PMN in lumen. Pockets of neutrophils in epithelium. Plasma cells in lamina propria; $\times 500$
 FIG. 2 Vesicula seminalis: Plasma cells in lamina propria; $\times 500$
 FIG. 3 Vesicula seminalis. Hyperplasia of epithelium; $\times 500$
 FIG. 4 Vesicula seminalis. Necrosis of epithelium; $\times 500$
 FIG. 5 Epididymis. Accumulation of inflammatory cells in epithelium and in lumen of tubule. Rupture of epithelium; $\times 500$
 FIG. 6 Epididymis. Papillary hyperplasia of epithelium. Pocket of PMN near tip of papilla; $\times 500$

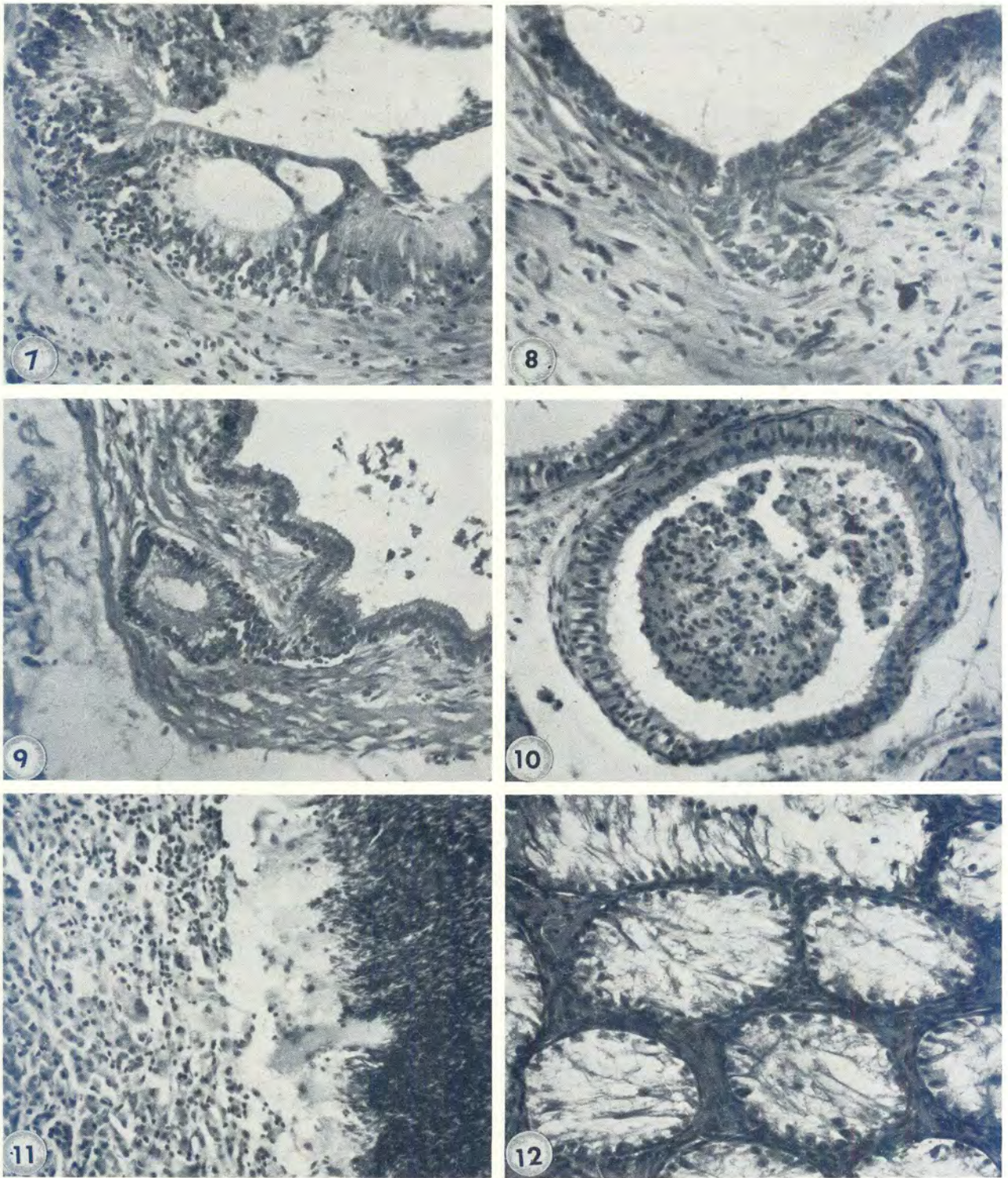


FIG. 7 Epididymis. Hyperplasia with cyst formation in epithelium of tubule; $\times 500$
 FIG. 8 Epididymis. Evagination of epithelium of tubule; $\times 500$
 FIG. 9 Epididymis. Evaginated epithelium surrounded by fibrous tissue; $\times 500$
 FIG. 10 Epididymis. Desquamation of epithelial cells; $\times 500$
 FIG. 11 Epididymis. Spermatic granuloma showing from L to R; macrophages and lymphocytes, foreign body giant cells, sperm heads; $\times 500$
 FIG. 12 Testis. Only Sertoli cells remaining in tubules, $\times 500$

Epididymis

The epididymis showed a wide variety of pathological changes. The mildest of these consisted of cuffing with lymphocytes of the vessels adjacent to the basal layer of tubular epithelium. Sometimes large collections of lymphocytes migrated to a position among the epithelial cells.

In some specimens polymorphonuclear neutrophils accumulated in the interstitial tissue and also infiltrated the epithelium.

At some sites degenerated lymphocytes and neutrophils were present in the lumen of the tubules (Fig. 5).

In many cases plasma cells accumulated in the interstitial tissue.

The following epithelial changes were evident:

- (i) Papillary hyperplasia (Fig. 6);
- (ii) hyperplasia with cyst formation (Fig. 7);
- (iii) evagination of epithelial cells (Fig. 8), some of the pockets of evaginated cells being surrounded by fibrocytes and collagenous material (Fig. 9).
- (iv) necrosis of the epithelium with cytolysis, pyknosis and karyolysis of its cells;
- (v) large scale desquamation of epithelial cells. These cells accumulated in the tubular lumen and showed degeneration (Fig. 10).

Some specimens showed an increase of fibrous tissue in the intertubular areas.

Typical spermatic granulomata as described by Jubb & Kennedy (1970) were frequently present. In some sections the successive layers of foreign body giant cells, macrophages and lymphocytes distributed in fibrous connective tissue could be clearly seen adjacent to masses of sperm heads (Fig. 11).

In the sections from acute cases of epididymitis the interstitial tissue was flooded with large and small lymphocytes and neutrophils. The same cells together with amorphous material distended the tubules and caused compression and degeneration of the epithelium. Many of the inflammatory cells showed degenerative changes. The blood vessels were dilated. Fibroblastic activity was evident around the lesions.

Testis

Masses of lymphocytes and neutrophils were present among the spermatocytes and spermatogonia in lesions of acute orchitis. The inflammatory cells also infiltrated the interstitial tissue. The spermatocytes, spermatogonia, Sertoli cells and many of the inflammatory cells in the tubules and interstitium showed pyknosis, karyorrhexis and karyolysis. The blood vessels were distended, and along the edge of the lesions there were obvious fibroblastic activity and infiltration of plasma cells.

In several specimens the spermatogonia and spermatids had virtually disappeared, leaving only the nuclei of the Sertoli cells and their thread-like cytoplasm (Fig. 12). Lymphocytes accumulated around the vessels of the interstitium and collections of plasma cells were evident. There was also an increase in fibrous tissue in the interstitium.

Some chronic lesions showed an increase in fibrous tissue in the intertubular areas with infiltration of plasma cells. In addition, all cells had disappeared from the seminiferous tubules and their lumen was filled with a partly calcified amorphous material.

Semen

The findings were in agreement with those of Van Tonder (1977). In rams with normal epididymides and infection of the vesiculae seminales or ampullae many bacteria and neutrophils were distributed among the spermatozoa. On the other hand, rams with lesions occluding the tubuli epididymidis as well as infection of the vesiculae seminales or ampullae showed no spermatozoa but only neutrophils and bacteria in their semen smears.

DISCUSSION

This investigation has shown that profound pathological changes are present in the genital tracts of rams from which a variety of bacteria was isolated. When these changes are mild and do not affect the free flow of semen, they might not noticeably impair the fertility of the ram concerned. But in most of the cases

examined the lesions were sufficiently severe to reduce the breeding potential of the ram, especially when the testes or epididymides were affected.

The study confirms the notion that a clinical examination only is insufficient for the assessment of the breeding ability of rams. It also indicates that the term "ram epididymitis" gives an inadequate description of the condition. While it is true that in some rams only one or both epididymides were affected clinically, in others these organs were completely normal and free of bacteria, but their ampullae and vesiculae seminales showed marked histopathological changes. Such animals no doubt account for the finding in practice of rams showing contamination of their semen with neutrophils, lymphocytes and bacteria, but no outward clinical lesions. I, therefore, suggest that the designation "bacterial infection of the genitalia" (BIG) of rams would be more appropriate.

The lesions found in the testes and epididymides closely correspond to those described by Kennedy *et al.* (1956) for infections by *Br. ovis*. These lesions, therefore, seem to be characteristic of bacterial infections in general of the genitalia.

A striking feature of the chronic lesions seen in all the organs was the accumulation of plasma cells in the tissue. They were usually accompanied by an abundance of lymphocytes in their immediate vicinity, creating the impression of an active immune response to the infection. Even the bacteria that might be regarded as mere commensals in the sheath elicited this supposed immune response.

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REFERENCES

- BIBERSTEIN, E. L., MCGOWAN, B., OLANDER, H. & KENNEDY, P. C., 1964. Epididymitis in rams. Studies on pathogenesis. *Cornell Veterinarian*, 54, 27-41.
- CLAXTON, P. D. & EVERETT, R. E., 1966. Recovery of an organism resembling *Histophilus ovis* from a ram. *Australian Veterinary Journal*, 42, 457-458.
- DELONG, W. J., WALDHALM, D. G. & HALL, F., 1979. Bacterial isolates associated with epididymitis in rams in Idaho and Eastern Oregon. *American Journal of Veterinary Research*, 40, 101-102.
- DODD, D. C. & HARTLEY, W. J., 1955. A specific suppurative epididymitis of rams. *New Zealand Veterinary Journal*, 3, 105-110.
- EKDAHL, M. O., MONEY, D. F. L. & MARTIN, C. A., 1968. Some aspects of epididymitis of rams in New Zealand. *New Zealand Veterinary Journal*, 16, 81-82.
- JANSEN, B. C., 1980. The aetiology of ram epididymitis. *Onderstepoort Journal of Veterinary Research*, 47, 101-107.
- JAMIESON, S. & SOLTYS, M. A., 1947. Infectious epididymo-orchitis of rams associated with *Pasteurella pseudotuberculosis*. *Veterinary Record*, 59, 351-353.
- JUBB, K. V. F. & Kennedy, P. C., 1970. Pathology of domestic animals. 2nd Ed. New York and London: Academic Press.
- KENNEDY, P. C., FRAZIER, L. M. & MCGOWAN, B., 1956. Epididymitis in rams: Pathology and bacteriology. *Cornell Veterinarian*, 46, 303-319.
- MCGOWAN, B., 1979. Epididymitis in rams: Effects of vaccination and culling on the clinical incidence of the disease. *Cornell Veterinarian*, 69, 67-72.
- VAN DRIMMELEN, G. C., 1960. Control of brucellosis in sheep and goats by means of vaccination. *Journal of the South African Veterinary Medical Association*, 31, 129-138.
- VAN TONDER, E. M., 1977. *Actinobacillus seminis* infection in sheep in South Africa. D.V.Sc. Thesis, University of Pretoria.
- WATT, D. A., 1978. Testicular pathology of Merino rams. *Australian Veterinary Journal*, 54, 473-478.