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ANATOMICAL AND PATHOLOGICAL STUDIES
ON THE SEX ORGANS FROM
SLAUGHTERED BULLS IN HOKKAIDO

III. PATHOLOGICAL FINDINGS

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(Received for publication, January 11, 1961)

In the previous papers, the present author has reported the anatomical findings on the sex organs and the occurrence of the remnants of the Müllerian duct in the bulls. In the present paper, microscopical findings will be presented about the same materials as were used in the previous investigations. The result of the chemical analysis for urinary calculi will also be described.

MATERIALS AND METHODS

The materials used in this investigation were sex organs taken from the 15 bulls which

CHART 1. Collection Sites of the Specimens for Microscopic Examination (I)

Notes:
Aa. Testis
Ab. Head of epididymis
C. Body of epididymis
D. Tail of epididymis
E. Ductus deferens
Fa. Penis (extra-pelvic part)
Gb. Prepuce

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were previously reported in this journal). Specimens for the histological examination were taken from both sides of testis, epididymis, ductus deferens, ampulla ductus deferentis, seminal vesicle, prostate, bulbo-urethral glands, extra-pelvic and pelvic part of penis, and prepuce. The collection sites of each specimen are shown in charts 1 and 2. After having been fixed with 10 per cent formal solution, these specimens were embedded in paraffin. But, only the testes specimens were also fixed with ALLEN-BOUIN's and BOUIN's solutions immediately after slaughter. Sections were made from them and stained with hematoxylin-eosin solution for microscopical examination.

**Findings**

**Testis**  Testicular degeneration was observed in 3 cases (H-20, 27 & 32) and orchitis in 2 cases (H-18 & 33).

**Bull No. H-18, 6-year-old Holstein bull**

The semen picture and fertility rate were satisfactory, but he was slaughtered June 14, 1958 because of unfavorable factors for further demands in reproduction. Both the testes showed slight vacuolation of seminiferous epithelia and edematous thickness of basal membrane in the several seminiferous tubules. Perivascular cellular accumulation with histiocytic cells and proliferation of the connective tissue were observed in the interstitial tissue. This case was diagnosed as slight chronic interstitial orchitis.

**Bull No. H-20, clinical history was not known because no information came from the owner.**

In both testes, some seminiferous tubules had suffered from degeneration. Affected tubules showed slight vacuolation, decrease in number of seminiferous epithelial cells, arrest of spermatogenesis and thickness of basal membrane. In the interstitial tissue edema was observed.

**Bull No. H-27, 7-year-old Guernsey bull**

May 1953, this bull was imported from England. The semen picture was satisfactory but fertility rate was unfavorable (Table 1). He was affected by lameness during 1957 and slaughtered May 9, 1958 because of low fertility and lamness. The semen picture one week before slaughter is shown in table 2. In both testes, several seminiferous tubules were affected by degeneration, showing moderate atrophy, vacuolation and arrest of spermatogenesis. Edematous loosening of the interstitium were observed.
KANAGAWA, H.

TABLE 1. Fertility Rate of H-27

<table>
<thead>
<tr>
<th>YEAR</th>
<th>NUMBER OF SERVICE</th>
<th>CONCEPTION</th>
<th>CONCEPTION RATE</th>
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<tbody>
<tr>
<td>1953</td>
<td>27</td>
<td>5</td>
<td>18.5%</td>
</tr>
<tr>
<td>1954</td>
<td>24</td>
<td>5</td>
<td>20.8%</td>
</tr>
<tr>
<td>1955</td>
<td>13</td>
<td>3</td>
<td>23.1%</td>
</tr>
<tr>
<td>1956</td>
<td>20</td>
<td>6</td>
<td>30.0%</td>
</tr>
<tr>
<td>1957</td>
<td>8</td>
<td>3</td>
<td>37.5%</td>
</tr>
<tr>
<td>1958</td>
<td>4</td>
<td>2</td>
<td>50.0%</td>
</tr>
</tbody>
</table>

TABLE 2. Semen Picture of H-27

<table>
<thead>
<tr>
<th>VOLUME</th>
<th>MOTILITY</th>
<th>NO. OF SPERM</th>
<th>pH CATALASE TEST*</th>
<th>ABNORMAL SPERMATOZOA</th>
<th>LIVE-DEAD STAIN*</th>
</tr>
</thead>
<tbody>
<tr>
<td>ml</td>
<td>%</td>
<td>million/ml</td>
<td>Head</td>
<td>Body</td>
<td>Tail</td>
</tr>
<tr>
<td>4.0</td>
<td>70</td>
<td>174.5</td>
<td>6.2~6.4</td>
<td>145</td>
<td>53/100</td>
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Bull No. H-32, 5-year-old Holstein bull

The semen picture and fertility rate were satisfactory. July 20, 1959, he was slaughtered because of chronic lameness. In both testes, some seminiferous tubules showed degeneration with atrophy, vacuolation and decrease in height of seminiferous epithelium, increase in number of the Sertoli cells and only one layer of spermatogonia along basal membrane. Edema in the interstitial tissue was noted.

Bull No. H-33, 6-year-old Holstein bull

Since May 1954, he had been used for natural service. His semen picture was normal. July 1958, the owner first noted decrease in fertility rate (Table 3). After March 1959, semen examinations showed that he was suffering from oligozoospermia (Table 4). The bull was treated with 1,000 mg of testosterone propionate every other day during the period March 17

TABLE 3. Fertility Rate of H-33

<table>
<thead>
<tr>
<th>YEAR</th>
<th>NUMBER OF SERVICE</th>
<th>CONCEPTION</th>
<th>CONCEPTION RATE</th>
</tr>
</thead>
<tbody>
<tr>
<td>1955</td>
<td>45</td>
<td>28</td>
<td>62.2%</td>
</tr>
<tr>
<td>1956</td>
<td>109</td>
<td>78</td>
<td>71.6%</td>
</tr>
<tr>
<td>1957</td>
<td>207</td>
<td>112</td>
<td>54.1%</td>
</tr>
<tr>
<td>1958</td>
<td>34</td>
<td>13</td>
<td>38.2%</td>
</tr>
<tr>
<td>1959</td>
<td>13</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>
### TABLE 4. Semen Picture of H-33

<table>
<thead>
<tr>
<th>DATE</th>
<th>VOLUME</th>
<th>MOTILITY (%)</th>
<th>pH</th>
<th>NO. OF SPERM</th>
</tr>
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<tr>
<td>March 2, '59</td>
<td>3.0</td>
<td>++ 0 35 60</td>
<td>6.9</td>
<td>31~33</td>
</tr>
<tr>
<td>March 4, '59</td>
<td>6.5</td>
<td>++ 0 90</td>
<td>6.5</td>
<td>37</td>
</tr>
<tr>
<td>March 7, '59</td>
<td>6.5</td>
<td>++ 0 35</td>
<td>6.5</td>
<td>65</td>
</tr>
<tr>
<td>March 11, '59</td>
<td>6.5</td>
<td>++ 0 10 10</td>
<td>6.4</td>
<td>65</td>
</tr>
<tr>
<td>March 24, '59</td>
<td>10.0</td>
<td>++ 0 10 10</td>
<td>6.4</td>
<td>88</td>
</tr>
<tr>
<td>April 1, '59</td>
<td>7.0</td>
<td>++ 0 20 20</td>
<td>6.5</td>
<td>92</td>
</tr>
<tr>
<td>April 20, '59</td>
<td>5.0</td>
<td>++ 0 40 40</td>
<td>6.5</td>
<td>35</td>
</tr>
<tr>
<td>June 10, '59</td>
<td>7.0</td>
<td>++ 0 40 40</td>
<td>6.5</td>
<td>34</td>
</tr>
<tr>
<td>July 1, '59</td>
<td>6.5</td>
<td>++ 0 30 50</td>
<td>6.5</td>
<td>27</td>
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### CHART 3. Therapeutic Results of H-33

Notes:

- □ 100 mg of Amorisin (Testosterone propionate) every other day, 1,000 mg in total
- ■ 1,000 IU of Anteron (PMSG) every other day, 10,000 IU in total
- □ 50~60 g of Minex every day, 2,000~2,400 g in total
to April 6, 10,000 IU of PMSG every other day during the period March 25 to April 16 and 50~60 g Minex* every day from May 22 to July 25. During administration of these preparations, the number of spermatozoon increased about two to three times as compared with the finding before the therapy (Chart 3). But, when the therapy was stopped, the number of spermatozoon gradually decreased. Therefore he was slaughtered July 28, 1959.

The right testis was small in size (13.0 x 6.1 x 6.0 cm), very firm in consistency, weighing 250 g. The left testis was also small in size (12.9 x 6.5 x 6.4 cm), firm in consistency, weighing 260 g. Histologically both testes showed severe atrophy of seminiferous tubules, vacuolation of seminiferous epithelium and arrest of spermatogenesis with appearance of giant cells. In the interstitial tissue marked edema, fibrosis, histiocytic proliferation, cellular infiltration and foci with lymphocytes, plasma cells or neutrophilic leucocytes were observed. In this case, a diagnosis of severe chronic interstitial orchitis with severe atrophy was made.

Epididymis A slight degree of pathological changes was observed in the following 2 cases: cellular foci with histiocytes in a part of the serosa (H-16), and in the interstitial tissue of the head of epididymis (H-18). The remaining cases revealed no remarkable changes.

Ductus deferens No case showed remarkable pathological changes.

Ampulla ductus deferentis A slight degree of pathological changes was observed in the following 8 cases: lymphocytic cellular infiltration and histiocytic proliferation in the tunica propia and perivascular cellular foci in the muscular layer (H-15), edema of the interstitial tissue (H-21), desquamation of epithelial cells in the lumen (H-27), vacuolation in the epithelium (H-29, 30, 32 & 33), and proliferation with histiocytes in the interstitial tissue (H-31).

Seminal vesicle A slight degree of pathological changes was observed in the following 4 cases: slight infiltration of lymphocytes in the submucosa (H-21), edema in the interstitial tissue (H-29), desquamation and degeneration of the glandular epithelium (H-30), and vacuolation of glandular epithelial cells (H-31). The other cases revealed no remarkable changes.

Prostate A slight degree of pathological changes was observed in the following 3 cases: partial edematous dilation of the duct (H-16), slight cellular infiltration with lymphocytes or neutrophilic leucocytes, and histiocytic proliferation in the interstitium (H-22), and slight edema in the interstitium (H-26). The remaining cases revealed no remarkable changes.

Bulbo-urethral glands No case showed remarkable pathological changes except H-18 in which a slight degree of congestion in the interstitial tissue was observed.

Urinary tract Urinary calculi were found in 2 cases (H-20 & 32). In both of them, many small calculi were observed throughout the inside of the urethra, especially in the distal curve of the sigmoid flexure and pelvic part of the urethra. The calculi were mostly light gray or white in color, of the size of a grain of sand or of rice and of the consistency of hard stones. Chemical analysis demonstrated that these urinary calculi were composed mainly of silicic acid, as shown in the following table 5.

The urinary tract in these 2 cases was affected by chronic catarrhal inflammation; mucosal epithelium showed slight degree of congestion and hemorrhage. Cellular infiltration and foci

* Mixed mineral powder preparation including Fe, Cu, Co, Zn, Mn, Mg, Ca, NaCl and vitamin K₄ prepared by the TAKEDA Chemical Industries Ltd., Osaka, Japan
with lymphocytes or neutrophilic leucocytes, and proliferation with histiocytes were noted in the submucosa. In another case (H-33), slight proliferation of histiocyte was observed. In the remaining several cases, there were observed only slight pathologic changes such as congestion, petechiae or edema in the mucosa.

**Prepuce** In 9 cases (H-18, 21~23, 26, 27 & 31~33), chronic catarrhal posthitis was observed; lymphatic nodules of the preputial mucosa were enlarged with hemorrhage and accumulation of infiltrating leucocytes. In the corium intense cellular infiltration with leucocytes and histiocyte proliferation were noted. In one case (H-28), histiocyte proliferation in the corium was superior to that in the other cases.

**DISCUSSION**

As regards the pathology of the testis, LAGERLOF proposed that the pathological state of the testis was classified into the following four categories: hypoplasia, degeneration, orchitis and fibrosis. Hypoplasia of the testis has been thoroughly investigated in the Swedish Highland Breed by LAGERLOF and ERICKSSON. But, in Japan, no report concerning this condition of the bull has been published up to this time. In the present author's cases, three bulls (H-20, 27 & 32) demonstrated histological findings similar to those of partial hypoplasia, showing degeneration in several seminiferous tubules with a single layer of markedly vacuolized spermatogonia, moderately thickened basal membrane and almost empty cavity. But, in these cases the semen picture and the fertility rate were almost normal and also the testes were normal in size. In addition, the genetic condition of these cases could not be known. Therefore, a diagnosis not of partial testicular hypoplasia, but of partial testicular degeneration was made. The causes of these conditions are not yet entirely clear, but they may be due to some physical or traumatic factors including severe climatic condition, mechanical injury, excessive physical exercise and transportation under adverse condition.
The orchitis case with hormone therapy (H-33) showed a temporary increase in number of spermatozoa during the administration of testosterone propionate and PMSG. Tiba et al. have reported similar findings during treatment with testosterone propionate in a case of azoospermia in the bull. Heckel and McDonald reported "rebound phenomenon" of the spermatogenic activity following the administration of testosterone propionate in a case of human azoospermia. But, the present author's case did not show the "rebound phenomenon" after cessation of the treatment.

According to Gibbons, inflammations in the epididymis only are uncommon and the accessory sex glands are seldom grossly diseased in the bull. Prostatic diseases in the bull are almost unknown. In the course of the present author's investigation, no remarkable pathological changes were observed in the epididymis and other accessory sex glands, excepting slight changes such as congestion or edema observed in several cases.

Blount investigated urinary calculi in cattle, and stated that most of them were composed of silicate, calcium oxalate, xanthine, calcium or magnesium carbonate and phosphate. Newsom also reported that urinary calculi in cattle and sheep were composed of calcium, magnesium and aluminum salts of phosphoric acid. Swingle found protein and silica in urinary calculi in steers. In the present author's cases silicic acid was a main component of urinary calculi. Connell et al. have described a number of causative factors to produce urinary calculi: vitamin A deficiency, insufficient water intake, intake of highly mineralized water, nutritional imbalance especially as regards minerals, intake of certain feedstuffs, weather and climatic effects, and metabolic disturbances associated with abnormal excretion of salts. In general, however, bovine urolithiasis is considered to be nutritional in origin. In one case of the present materials (H-32) the feedstuff was largely rice straw as roughage. This case would suggest that rice straw may be concerned with the formation of urinary calculi, because of the fact that rice straw contains a high percentage of silicic acid (about 20 per cent).

As for the posthitis, Gibbons reported that inflammation of the sheath and penis is common in bulls. Milne also reported that balanoposthitis was quite commonly encountered in the bull, because the prepuce of the bull is long and narrow and the preputial orifice is small as compared with that of the horse, thereby increasing the tendency to produce inflammation of this region. In the present author's observation, chronic catarrhal posthitis was found in 9 cases. Its cause could not yet be explained clearly. According to Milne, bacterial infection from the bedding, accumulations of smegma, foreign matter such as grass awns and exposure of the penis to cold temperature, may be the principal cause of posthitis in the bull.
SUMMARY

Pathological investigations were made on the genital organs from 15 slaughtered bulls. The findings may be summarized as follows:

Testicular degeneration was observed in 3 cases and orchitis in 2 cases. The former was similar to partial hypoplasia, showing degeneration in several seminiferous tubules with a single layer of markedly vacuolized spermatogonia, moderately thickened basal membrane and almost empty cavity. The histological findings of the orchitis cases were accordant with those of chronic local interstitial orchitis, showing atrophy and vacuolation in the seminiferous tubules. In the interstitial tissue, there existed marked edema, proliferation of histiocytic cells, fibrosis and cellular infiltration or cellular foci. One of the orchitis cases had been treated with testosterone propionate and PMSG; during administration of these preparations, the number of spermatozoon increased to about 2~3 times that before the therapy.

Urinary calculi were found in 2 cases. The urinary tract in these cases was affected by chronic catarrhal inflammation. Chemical analysis of the urinary calculi showed mainly silicic acid.

In the prepuce, chronic catarrhal posthitis was observed in 9 cases. In the corium intense cellular infiltration and foci were noted.

The present author wishes to express his cordial gratitude to Professor T. ISHIKAWA, chief of this Department, for his kind guidance. Grateful acknowledgment is also made to Assistant Professor Y. FUJIMOTO for his kind instruction in the pathological investigation, to Assistant Professor H. SHIOKAWA for his helpful advice on the chemical analysis of urinary calculi, and to Assistant Professor K. KAWATA for his kind advice throughout this investigation. He is also indebted to Dr. M. YAMAKAWA, Inspector at Sapporo Abattoir, for the kind supply of the materials used.

REFERENCES


EXPLANATION OF PLATES

PLATE I.

Fig. 1. Bull No. H-18: Left testis. At central portion of picture, marked perivascular cellular infiltration can be seen in interstitial tissue. × 60.

Fig. 2. Bull No. H-20: Left testis. In two seminiferous tubules, degeneration and arrest of spermatogenesis have occurred; seminiferous tubules at lower part of picture are almost normal. × 180.

Fig. 3. Bull No. H-20: Severely degenerated seminiferous tubules of right testis. × 180.

Fig. 4. Bull No. H-27: Edematous loosening of interstitial tissue of left testis. At right portion of picture a degenerated seminiferous tubule is seen. × 180.

Fig. 5. Bull No. H-33: Marked vacuolation of seminiferous tubules, cellular infiltration and proliferation in interstitial tissue in left testis. × 180.

Fig. 6. Bull No. H-33: Marked cellular infiltration, edema and slight degree of fibrosis in interstitial tissue in right testis. × 180.

PLATE II.

Fig. 7. Bull No. H-20: A part of cervix of urinary bladder and pelvic part of urinary tract. Many small urinary calculi are observed.

Fig. 8. Bull No. H-20: Upper part of penis. Calculi are of size of a grain of sand or of rice.

Fig. 9. Bull No. H-20: Under part of penis. At upper portion of picture calculi are observed in curve of sigmoid flexure.

Fig. 10. Bull No. H-32: Pelvic part of urinary tract. Numerous small calculi can be seen (scale: cm).

PLATE III.

Fig. 11. Bull No. H-20: Inflammation of urinary tract. Cellular infiltration in submucosa is noted. × 180.


Fig. 14. Bull No. H-22: Chronic catarrhal posthitis. Preputial mucosa are thickened and cellular infiltration is noted in corium. × 60.

Fig. 15. Bull No. H-26: Chronic catarrhal posthitis. Marked cellular foci in corium. × 60.

Fig. 16. Bull No. H-28: Histiocytic proliferation and cellular infiltration in corium of prepuce. × 180.