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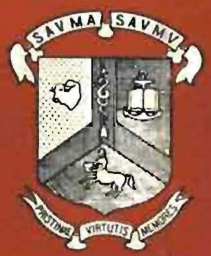

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VOLUME 39 NUMBER 2
JAARGANG 39 NOMMER 2

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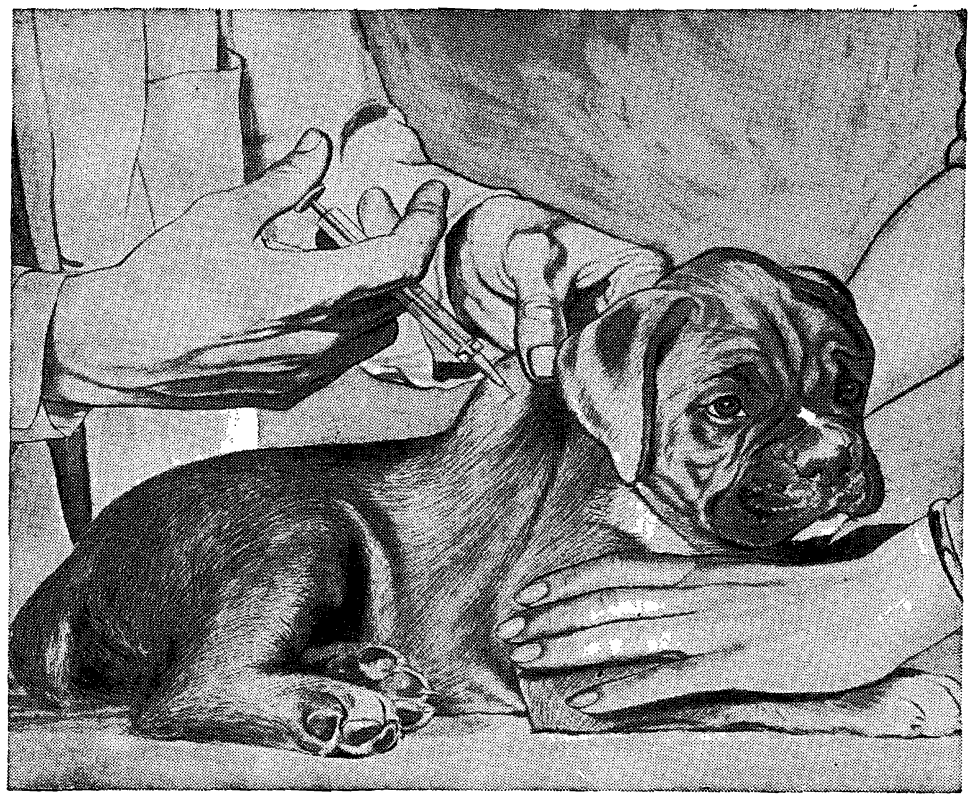

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VERENIGING

VOLUME 39 No. 2

JAARGANG 39 Nr. 2

JUNE 1968 JUNIE

CONTENTS / INHOUD

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Jl S. Afr. vet. med. Ass. 39 (2): 3-8 1968

COPPER METABOLISM IN THE MERINO SHEEP IN SOUTH AFRICA

I. The determination of Copper in body tissues and fluids and of the various Copper fractions in blood.

J. M. M. BROWN*

SUMMARY

Methods, based on complex formation between copper ions, acetaldehyde and oxalyldihydrazide, are presented for the determination of copper in body fluids and tissues. These methods include the determination of total plasma copper, red-cell copper, loosely bound plasma albumin copper and globulin bound copper (by difference).

INTRODUCTION

Peterson and Bollier[1] introduced bis-cyclohexanone oxalyl-dihydrazone as a reagent for determining copper in body tissues and fluids. Various studies on the condensation products of aldehydes and ketones with oxalylhydrazide and the blue colours which the hydrazones gave with copper ions had been conducted previously[1]. The introduction of this procedure was soon followed by the publication of numerous methods for the determination of copper in a variety of media using other reagents of this nature, e.g. oxalyldihydrazide and acetaldehyde[2] or 1,5-diphenylcarbohydrazide[3]. We made use of the Peterson and Bollier[1] procedure for a while but found it to be very sensitive to pH variations. The colour produced with copper ions at the concentrations encountered in blood also gave very low readings on the various photo-electirc colorimeters then in use in our laboratory. The pink colour developed by combination of copper ions with oxalyldihydrazide and acetaldehyde was far more satisfactory and gave results of better reproducibility. Methods using these reagents, however, all suffered from the same serious disadvantage, notably the extremely volatile nature of acetaldehyde. This property made accurate work with this reagent most difficult during the hot summer months, particularly under the semi-desert conditions of the Karoo. It was decided therefore to develop the methods outlined below in which this objection is removed without decreasing the sensitivity of the colour reaction.

PRINCIPLE OF THE METHODS

Copper forms a deep pink coloured complex with bis-acetaldehyde - oxalyldihydrazone at pH 8.4-9.1. The hydrazone is itself formed directly in the reaction medium by the interaction of acetaldehyde-ammonia in ammoniacal solution. Acetaldehyde-ammonia (1-amino-ethanol, α-amino-ethyl alcohol or "aldehyde ammonia") is a crystaline solid which is conveniently stored at low temperature without appreciable decomposition and is easily handled at room temperatures. For use in the methods described below, fresh solutions of the compound are made daily. Such solutions give all the reactions typical of acetaldehyde. The primary product of the reaction of a primary amine or ammonia with an aldehyde can be considered as the addition compound R.CH(OH) .NHR, which may lose the elements of water to give an azomethine or may undergo further condensation. When dry ammonia is passed through an ethereal solution of acetaldehyde, the well-known acetaldehyde-ammonia is formed. Although analytical figures correspond to the formula $CH_3.CH(OH).NH_2$, various investigations have shown that the product is in fact more complex. Its molecular weight in water corresponds with the formula, $3(CH_3.CH(OH).NH_2)$.[4] No general agreement on its structure has yet been reached. It appears that the formation of this type of compound from aromatic aldehydes like benzaldehyde and anisaldehyde is preceded by the production of an unstable additive compound of the general formula $(R.CHO)_2NH_3$[4]. Whether this occurs with the aliphatic derivatives or whether such compounds form in solution is not known. Hydrolysis of the compound in aqueous solution might occur thus:

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$$CH_3.CH(OH).NH_2 + H_2O = CH_3.CHO + NH_4OH$$

The overall reaction of acetaldehyde and oxalyldihydrazide with copper ions is, however, more complex than mentioned here. Formation of the pinkish purple complex takes place best in the presence of ammonium ions[1]. It was found that colour development is similar when acetaldehyde-ammonia is used instead of acetaldehyde in the presence of ammonia.

PREPARATION OF THE COLOUR REAGENTS

These are easily and rapidly prepared as follows:- (1) Oxalyldihydrazide: Two molecules of hydrazine react with one of diethyloxalate to form one molecule of oxalyldihydrazide and two of ethanol, i.e. 69 g of hydrazine and 146 g of diethyl oxalate are required to produce 118 g of the hydrazide. Add approximately 130.0 g of 50% hydrazine hydrate slowly to 20 ml absolute ethanol. In a separate container dissolve 14.6 g diethyl oxalate in 20 ml absolute ethanol. Mix the two solutions. The hydrazide crystallizes out almost at once. Place the reaction flask in the refrigerator overnight to allow for complete precipitation. Separate the crystals by filtration with suction and wash them twice with ice cold ethanol and twice with ice cold water. Recrystallize the product twice by solution in the minimum of boiling water and rapid cooling in the refrigerator. Dry in a desiccator over sulphuric acid or phosphorus pentoxide.

(ii) Acetaldehyde-ammonia: This is conveniently prepared by passing dry ammonia gas through an ice-cold solution of the required amount of acetaldehyde in anhydrous ether as described by Cohen.[5] The reaction vessel is kept cooled by surrounding it with an ice-salt mixture. When the reaction is completed the crystals of aldehyde-ammonia are separated by filtration with suction (preferably in the dark), then washed with repeated small volumes of ice cold ether, and finally dried *in vacuo*, over calcium chloride and paraffin chips in the dark. The finished product which is initially white turns light yellow in time. It should be stored in an amber bottle in the refrigerator, in which case it will keep very well for at least eighteen months without appreciable deterioration. The reagent is still active even when yellowish-brown in colour.

OUTLINE OF THE METHODS

(a) General reagent solutions: The following general reagents are required for the determination of the various copper fractions in blood and for the photometric part of the procedures for estimation of copper in urine, bile and body tissues: (i) 2N HCl; (ii) 60% w/v aqueous trichloracetic solution; (iii) concentrated ammonia (±28% NH_3); (iv) "dilute ammonia" (conc. NH_4OH 1:water 2); (v) oxalyldihydrazide solution: 0.25 g of the hydrazide dissolved in 15 ml water with the aid of heat. The solution is a saturated one and much oxalylhydrazide separates out on cooling. The supernatant solution is kept over the crystals and used when required. When it is exhausted, the same crystals are redissolved in another 15 ml water and after cooling the supernatant is kept and used in the same way. Larger batches of the reagent may be made using the proportions given above. The crystals may be used repeatedly for making these solutions in the manner described above, until a saturated solution is no longer obtained; (vi) acetaldehyde-ammonia solution: 0.5 g acetaldehyde ammonia dissolved in 15 ml water. This reagent is generally made up freshly each day, although the solution keeps quite well for at least a week, if it is stored at 4° C.; (vii) stock copper standard solution, containing 100 mcg Cu/ml; * (viii) "working" copper standard solution, containing 100 mcg Cu/100 ml (i.e. 1 mcg/ml); (ix) saturated aqueous solution of sodium pyrophosphate (5.4 g $Na_4P_2O_7$ $1OH_2O$ dissolved in 100 ml water); (x) saturated sodium citrate solution (64 g $Na_3C_6H_5O_7.2H_2O$ dissolved in 100 ml water); (xi) 0.9% w/v NaCl; (xii) acetate buffer for loosely bound plasma copper, pH 8.6: Place 0.7708 g ammonium acetate crystals in a 200 ml volumetric flask. Dissolve in about 100 ml distilled water, then add 1 ml of concentrated ammonia and 0.5 ml glacial acetic acid. Make to volume, mix and determine the pH. Adjust to 8.6, if necessary.

(b) Total plasma copper: 2 ml plasma, 2 ml water for the reagent blank and 2 ml "working" standard solution, are each placed in separate clean centrifuge tubes. Add to each 2 ml 2N HCl. Let stand for 10 minutes after mixing, then add 0.4 ml 60% trichloracetic acid. Stir the contents of the tube containing plasma with a thin glass rod until the protein coagulum has been reduced to a fine slurry. Allow to stand for 10 minutes in the refrigerator and then centrifuge down the protein precipitate. Transfer 2 ml of the

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clear supernatant centrifugate and 2 ml of the mixture from the "blank" and "standard" tubes to three clean test tubes. Add to each 0.65 ml of "dilute" ammonia. Adjust the pH to 8.6, if necessary, with more ammonia or glacial acetic acid. Adjust the volume of each to 3.5 ml. Mix, then add to each tube 0.25 ml oxalyldihydrazide solution and 0.25 ml acetaldehyde-ammonia solution. Mix and let stand for 1 hour; read the optical density at 544 mμ.

$$\text{Total plasma copper (mcg\%)} = \frac{\text{OD Test}}{\text{OD Standard}} \times 100$$

(c) *Loosely bound (albumin) copper in plasma:* The standard used here is the "working" standard diluted one in ten to contain 10 mcg/100 ml copper. A reagent blank and standard are necessary for each batch of samples and for each individual sample. A tube labelled "test" and one labelled "control" is required. Place in the various tubes concerned 1 ml water, 1 ml standard, and 1 ml plasma (this, in both "test" and "control" tubes). Add to each tube 2.5 ml acetate buffer. The pH of each should be 8.6. Add to the control tube 0.25 ml water and to the rest of the tubes 0.25 ml oxalyldihydrazide solution. Add 0.25 ml acetaldehyde-ammonia solution to all the tubes. Mix and let stand one hour. Read optical densities at 544 mμ.

$$\text{Loosely bound plasma copper} = \frac{\text{OD Test} - \text{OD Control}}{\text{OD Standard}} \times 10 = \text{mcg\%}.$$

(d) *Total red cell copper:* Centrifuge the blood sample to obtain the erythrocytes and wash these twice with 0.9% NaCl. After centrifuging following the second saline wash, the packed red cells are suspended in an equal volume of saline. Mix thoroughly and withdraw some of the suspension to fill two Wintrobe haematocrit tubes. The packed cell volume of this suspension must be known. Let this value be C in the final calculation. Set up three centrifuge tubes labelled "Blank", "Standard" and "Test" and in each of these place 2 ml water, "working" standard and cell suspension respectively. Add to each 2 ml 2N HCl. Mix and allow to stand for 10 minutes, then add to each 0.4 ml of 60% trichloracetic acid. The contents of the tube containing the cell suspension are stirred with a thin glass rod until a smooth slurry is obtained. Allow to stand for 10 minutes in the refrigerator then centrifuge off the protein precipitate.

Into three clean tubes labelled as above place 2 ml of the blank and standard mixtures and 2 ml of the clear centrifugate. To each tube add 0.4 ml saturated sodium pyrophosphate solution, 0.4 ml saturated sodium citrate solution and 0.65 ml of "dilute" ammonia. Adjust the pH, if necessary, to 8.6 and the volume of each solution to 4.25 ml (with water). To each tube add 0.25 ml oxalyl-dihydrazide and 0.25 ml acetaldehyde-ammonia solutions. Mix and let stand for 1 hour at room temperature. Read optical densities at 544 mμ.

$$\text{Mcg Cu/100 ml red cells} = \frac{\text{OD Test}}{\text{OD Standard}} \times \frac{10{,}000}{\text{C}}$$

(e) *Bile and urine copper:* Ten ml of bile or urine, 10 ml of working standard and 10 ml of water (for the blank) are placed into separate 100 ml Kjeldahl flasks together with 0.75 ml concentrated sulphuric acid, 5 ml concentrated nitric acid and some copper free glass beads, porcelain chips or carborundum chips. Digest rapidly to charring. Cool and add to each flask 0.5 ml 60% perchloric acid a further 2.5 ml concentrated nitric acid. Digest until the solution is light yellow. Cool and add 5 ml water and 0.5 ml 100 vols % hydrogen peroxide. Digest down to a volume of about 1.5-2.0 ml. If the digest is not colourless, add 2.5 ml water and 0.25 ml hydrogen peroxide. Digest again. Cool, add 5 ml water only. Digest till white fumes are evolved and a residue of 0.5-1.0 ml of digest remains. The final addition of water and peroxide indicated above can be repeated until the digest is colourless, but the final strong heating with water only must be done to remove all traces of peroxide. Allow the digests to cool. To each add about 3 ml water, then transfer the contents to a 10 ml measuring cylinder. Rinse the flask with two 1 ml portions of water and add this to the main digest. Rinse the flask with 2 ml concentrated ammonia solution which is also carefully added to the main digest. Cool, mix, check the pH and adjust to 8.6 with "dilute" ammonia or glacial acetic acid. Adjust the volume to 10 ml with distilled water.

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A 5 ml aliquot of this mixture is placed in a test tube for copper determination. The remaining 5 ml can be used for the determination of iron (see later) if so desired. Add to the 5 ml aliquot 0.5 ml oxalyldihydrazide and 0.5 ml of acetaldehyde-ammonia solutions. Mix and let stand for 1 hour at room temperature. Read optical densities at 544 $m\mu$.

$$\text{Urine or bile copper (mcg\%)} = \frac{\text{OD Test}}{\text{OD Standard}} \times 100$$

(f) *Tissue copper:* The standard used in this method is the "stock" standard containing 100 mcg Cu/ml. Place in separate Kjeldahl flasks 1 g of tissue, 1 ml of standard and 1 ml water. To each add 5 ml conc. H_2SO_4 and 5 ml nitric acid. Digest to charring. Proceed with the digestion from this step onwards as outlined under (e) above. The final step includes strong heating to the evolution of white fumes after adding 5 ml of water and reduction of the volume of the digest to about 5 ml. The digests are transferred quantitively to a 50 ml volumetric flask, the Kjeldahl flasks are thoroughly washed with small portions of distilled water and the washings are added to the digest, the volume of which is finally made up to the mark with distilled water. Mix, take an aliquot of 5 ml for copper determination and reserve the remainder for the estimation of iron if desired. To the 5 ml aliquots from the "Blank", "Standard" and "Test" digests add 1 ml saturated sodium pyrophosphate, 1 ml saturated sodium citrate and 2 ml "concentrated" ammonia solutions. Adjust the pH of each to 8.6 Make all the tubes to the same volume with distilled water. Mix and add 0.5 ml oxalyldihydrazide and 0.5 ml acetaldehyde-ammonia solutions. Mix, let stand for an hour and read optical densities at 544 $m\mu$.

$$\text{Mg Cu/100 g tissue} = \frac{\text{OD Test}}{\text{OD Standard}} \times 10$$

EXPERIMENTAL

(a) *General remarks on the methods:* The choice of the quantities of plasma, erythrocytes, body fluids and tissue used in these methods, the use of 2N HCl and 60% trichloracetic acid for liberation and extraction of bound copper and precipitation of plasma proteins and the use of sodium pyrophosphate and sodium citrate, in the amounts specified here, to complex interfering ions, are all based on principles embodied in earlier methods for the determination of copper in animal tissues and body fluids.[1,6,7]. The methods of digestion of body fluids and tissues are likewise based on earlier work [8-10]. The modifications introduced mainly concern the colour reaction with copper and the following sections are therefore devoted to experimental data concerning this aspect of the method only.

(b) *Spectral absorption of the copper complex:* The light absorption of the coloured complex, as prepared by using 2 ml of working standard and subjecting it to the procedure outlined under "total plasma copper", was studied over the spectral range 200-600 $m\mu$. The curve obtained by plotting optical density against wave length is a smooth bell-shaped curve with maximum absorption at 544 $m\mu$. The compound absorbs very strongly over the range 535-555 $m\mu$. If a photo-electric colorimeter is used for the estimation of copper by the methods outlined here, the light filter of choice would thus be an Ilford 625 or equivalent.

(c) *The influence of pH on complex formation and colour development:* A mixture of "working" standard, 2N HCl and 60% trichloracetic acid was made up using the proportions described earlier under (b). Two ml aliquots of this were taken and dilute ammonia (NH_4OH 1.: water 3) and water were added in the amounts shown in Table 1. After measurement of the pH on the contents of each tube, 0.25 ml of oxalyldihydrazide and acetaldehyde-ammonia solutions were added, the contents of the tubes mixed and allowed to stand for one hour before reading optical densities against a reagent blank. The optical densities found are given in Table 1. It can be seen from this table that the colour is maximal over pH range 8.4-9.1. Below pH 7.4 complex formation is negligible and at pH values higher than 9.1 the intensity of the colour is also decreased. These experiments were repeated over the pH range of 8.4-9.1. Maximum colour development was found to occur at pH 8.6. The "dilute" ammonia solution described under (a) earlier was found to be the most satisfactory reagent for adjusting the pH.

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TABLE 1: INFLUENCE OF pH ON COMPLEX FORMATION

Tube No.	ml NH_4OH	ml. Water	pH	OD
1	0.05	1.45	1.1	0
2	0.10	1.40	1.2	0
3	0.15	1.35	1.2	0.001
4	0.20	1.30	1.2	0.002
5	0.25	1.25	1.2	0.001
6	0.30	1.20	1.3	0.001
7	0.35	1.15	1.35	0.001
8	0.40	1.10	1.4	0.001
9	0.45	1.05	1.6	0.002
10	0.50	1.00	1.9	0.006
11	0.55	0.95	1.9	0.006
12	0.60	0.90	7.46	0.058
13	0.65	0.85	8.46	0.071
14	0.70	0.80	8.75	0.072
15	0.75	0.75	8.95	0.072
16	0.80	0.70	9.0	0.071
17	0.85	0.65	9.1	0.068
18	0.90	0.60	9.15	0.069
19	0.95	0.55	9.25	0.055
20	1.00	0.50	9.35	0.055

(d) *Time taken for maximum colour development:* Two ml aliquots of the working standard, 2N HCl: 60% trichloracetic acid solution, made up according to the proportions given under (b) were treated with 0.25 ml of oxalyldihydrazide and acetaldehyde-ammonia solutions. After mixing, optical densities were determined at ten minute intervals for the first hour and thereafter hourly for the next six hours. Colour develops within fifteen minutes and is maximal one hour after addition of the colour reagents. Very slight increases in optical density may be found over the next five hours, but thereafter the colour remains stable for at least three days if the tubes are corked and kept in the dark. Typical data are presented in Table 2.

TABLE 2: THE INFLUENCE OF TIME ON COLOUR DEVELOPMENT

Tube No.	15 mins.	30 mins.	45 mins.	60 mins.	120 mins.	24 hours
1	.064	.057	.062	.062	.064	.065
1A	.064	.057	.062	.062	.065	.064
2	.065	.063	.064	.066	.065	.065
2A	.066	.064	.066	.066	.072	.070
3	.065	.060	.064	.064	.069	.069
3A	.064	.060	.064	.064	.068	.067

For practical purposes we read optical densities of each batch of determinations at any time following one hour after addition of the colour reagents.

(e) *Reproducibility of results:* Table 2 also shows values obtained from six different samples prepared simultaneously by three different operators. If the pH is carefully adjusted and the reagents are added using highly accurate micropipettes, the reproducibility is excellent as shown by these figures which are representative of our work.

(f) *Obedience to Beer's Law:* The method gives linear results over the concentration range of 40-600 mcg% or 1-12 mcg in the test medium to which the colour reagents are added. Dilutions of the stock standard were made to cover this range, and each dilution was treated as described under (b) above. Optical densities were plotted against concentration and the resulting plot was a straight line throughout this range.

(g) *Tests for interfering substances:* The specificity of the reaction between oxalyldihydrazide, acetaldehyde and copper ions under conditions similar to those described here has been discussed by previous authors[2,9]. The following ions were tested for interference at the concentrations indicated in parenthesis: Ca (10 mg%); Mg (2 mg%); Fe^{III} (200 mcg%); Fe^{II} (200 mcg%); Na (160 meq/l); K (6 meq/l); Cl (100 meq/l); HCO_3^- (27 meq/l) and PO_4^- (5 meq/l). No interference was noted in any of the experiments when these ions were added to "working" standard copper solutions at the concentrations indicated. Urea and glucose have been tested at concentrations of 60% in each instance and have no effect on colour development.

(h) *Comparisons between the methods described here and other methods which we have used:* Table 3 shows the results obtained in the determination of total plasma copper on a number of samples of sheep plasma by our method and those of Cartwright, Jones and Wintrobe[6] (which specifies sodium diethyldithiocarbamate as colour reagent) and Peterson and Bollier[4] (which is a bis-cyclohexanone oxalyldihydrazone method). It is apparent from this table that there is good agreement between the results obtained with our method and that of Cartwright et al[6]. The Peterson and Bollier[1] procedure is inclined to give very variable results since colour development is weak if the pH of

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the reaction medium is not controlled very rigidly.

TABLE 3: COMPARISON OF RESULTS OBTAINED BY THE PROPOSED METHOD WITH THOSE OBTAINED BY EARLIER PROCEDURES

Sample No.	Cartwright et al mcg%	Peterson & Bollier mcg%	Our method mcg%
9584	155	100	130
9596	144	80	130
9606	133	100	130
9613	133	67	124
1546	101	100	92
7888	111	100	117
9585	112	100	96
9586	73	67	71
9585B	100	95	92

The results obtained by our method for copper in tissues also compare exteremely well with those obtained by the older diethyldithiocarbamate procedures [10]. We have not tried to adapt the Peterson-Bollier procedure to tissue work.

(i) *The effect of environmental temperature on colour development:* The method for total plasma copper outlined here has been used extensively by us during our field work described elsewhere. A very large number of determinations have been performed under bitterly cold winter and extremely hot summer conditions of the Karoo. Records have been kept of the optical density readings of the working standards used with every batch of samples analysed.

Although very low environmental temparatures retard *the rate* of colour development and high environmental temperatures markedly accelerate it, optical density readings obtained one hour after addition of the colour reagents were virtually identical under both sets of conditions and identical to results obtained under the more temperate summer and winter conditions of Pretoria.

ACKNOWLEDGEMENTS

The Chief, Veterinary Research Onderstepoort, is thanked for permission to publish this paper.

I am exteremely grateful to Adriana Wagner and Anna Brink and Mr. R. Gray for help during the development of the methods outlined here.

REFERENCES

1. Peterson R. E. & Bollier M. E. 1955 *Anal. Chem.* 27:1195
2. Stark G. R. & Dawson C. R. 1958 *Anal. Chem.* 30:191
3. Stoner R. E. & Dasler W. 1960 *Anal. Chem.* 32:1207
4. Hickinbottom W. J. 1959 *Reactions of organic compounds.* 3rd ed. London, N.Y. & Toronto. Longmans, Green & Co. Ltd.
5. Cohen J. B. 1937 *Practical organic chemistry.* London. MacMillan & Co.
6. Cartwright G. E., Jones P. J. & Wintrobe M.M. 1945 *J. biol. Chem.* 160:593
7. Gubler C. J., Lahey M. E., Aschenbrucker H., Cartwright G. E. & Wintrobe M.M. 1952 *J. biol. Chem.* 196:209
8. Cartwright G. E., Gubler C. J. & Wintrobe M.M. 1954 *J. clin. Invest.* 33:685
9. Markowitz H., Shields G. S., Klassen W. H., Cartwright G. E. & Wintrobe M.M. 1961 *J. biol. Chem.* 33:1594
10. Van Niekerk P. le R. 1937 *Onderstepoort J. vet. Sci. & Anim. Ind.* 9:623
11. Brown J. M. M. 1967 *Biochemical studies on geeldikkop and enzootic icterus D.V.Sc.* Thesis University of Pretoria.

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Jl S. Afr. vet. med. Ass. 39 (2): 11-12 1968

COPPER METABOLISM IN THE MERINO SHEEP IN SOUTH AFRICA

2. Normal levels of the blood copper fractions.

J. M. M. Brown, Anna Brink and Adriana M. Wagner*

SUMMARY

The normal ranges for plasma albumin- and globulin bound copper, total plasma copper, ceruloplasmin and red cell copper as determined by a previously described method, are presented in this paper.

INTRODUCTION

The previous paper in this series contained descriptions of the methods used for the determination of the various copper fractions present in blood and other body fluids. In this paper the normal ranges for the various copper fractions in the blood of Merino sheep as established by using these methods are presented.

MATERIALS AND METHODS

Heparinized blood specimens were taken taken from 96 adult Merino sheep maintained in the pool of available animals at Onderstepoort.

No attempt was made to separate the sexes in this group, but it consisted predominantly of wethers. The sheep were on a diet of lucerne and teff hay fed *ad libitum* and received also the concentrate mixture described elsewhere [1].

Blood copper fractions were determined as described in the first paper of this series [2]. Ceruloplasim was determined by the method of Houchin [4].

The statistical method of analysis of the data obtained is based on the construction of cumulative relative frequency curves and is described elsewhere [1, 3].

RESULTS

The normal ranges found for the various blood copper fractions after processing the raw data are presented in Table 1.

TABLE 1: RANGES OF BLOOD COPPER FRACTIONS

(Ceruloplasmin values are mg/100 ml. All other values are mcg/100 ml.)

	Globulin-bound Copper	Albumin-bound Copper	Total Plasma Copper	Red Cell Copper	Cerulo-plasmin
98% Range	90-220	0-20	90-223	2.5-190	2.0-13.0
80% Range	92-163	0-6.7	97-160	17.5-118	4.5-10.1

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Histograms indicating the distribution of the various data and the cumulative relative frequency curves from which Table 1 was compiled are presented in Figures 1, 2 and 3.

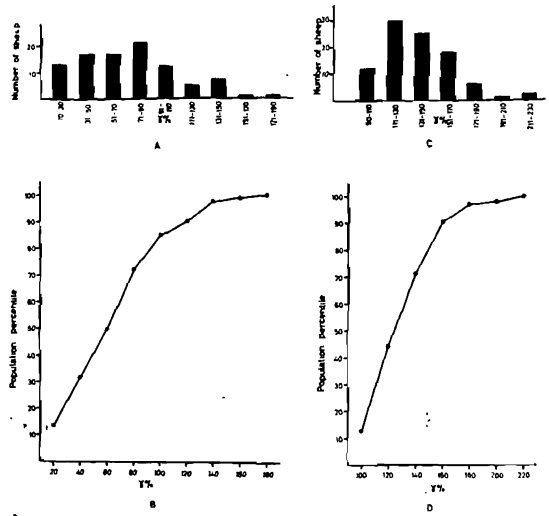


Fig. 1: A and B = Red blood cell copper; C and D = Total plasma copper.

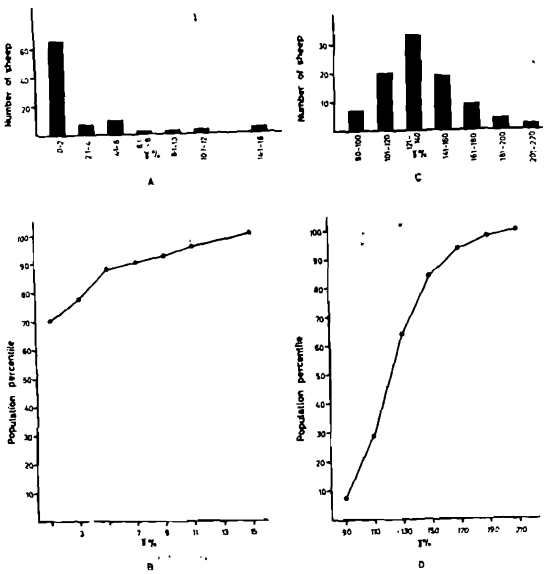
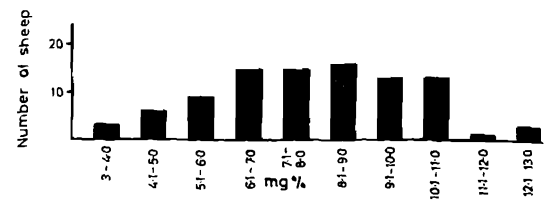


Fig. 2: A and B = Albumin-bound copper; C and D = Globulin-bound copper.



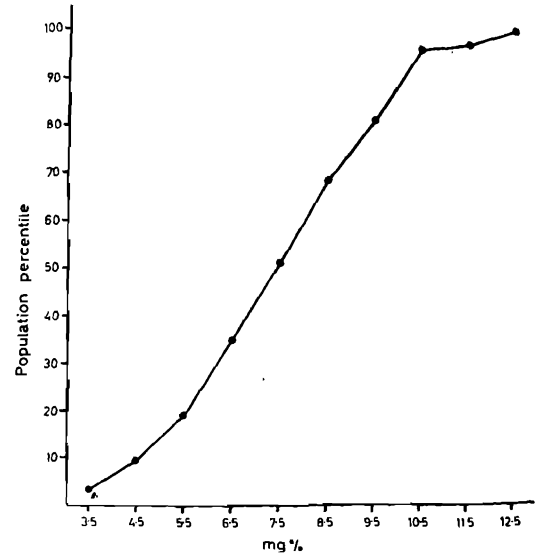


Fig. 3: Ceruloplasmin.

ACKNOWLEDGEMENT

The Chief, Veterinary Research Institute, Onderstepoort, is thanked for permission to publish this paper.

Messrs. R. Gray, R. J. Briel and P. J. de Wet are thanked for technical assistance.

REFERENCES

1. Wagner A. M. 1964 *Onderstepoort J. vet. Res.* 31:77
2. Brown J. M. M. 1968 Copper metabolism in the Merino sheep in South Africa. 1: The determination of copper in body tissues and fluids and of the various copper fractions in blood. *Jl S. Afr. vet. med. Ass.* This Journal.
3. Sion J. J. G. 1966 *Onderstepoort J. vet. Res.* 33:353.
4. Houchin O. B. 1958 *Clin. Chem.* 4:519

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Jl S. Afr. vet. med. Ass. 39 (2): 13-19 1968

THE LOW TOXICITY, ABSORPTION, TURNOVER AND EXCRETION OF COPPER IN THE MERINO SHEEP

L. P. NEETHLING*, J. M. M. BROWN** AND P. J. DE WET**

The absorption, storage and excretion of copper has been investigated in Merino sheep following intra-abomasal, intraruminal and intravenous administration of Cu-64. The excretion of the element via bile or urine is strictly limited. There is a most effective mechanism operating in the sheep which limits the intestinal absorption of the element when given in physiological or reasonable pharmacological doses. The absorption is increased to a certain threshold value in animals depleted of copper. In such animals administration of single abnormally large doses of copper does not force more copper through the intestinal mucosa. The ovine sheep liver clears administered copper rapidly from the blood stream and retains it avidly. Excess copper presumably previously loosely bound to albumin is rapidly eliminated by the kidneys.

INTRODUCTION

Copper metabolism has been extensively studied in small laboratory animals, dogs and man. Studies in sheep have largely centred around acute and chronic intoxications and the relationship of the copper status of the animal to certain diseases, e.g. toxaemic jaundice and enzootic icterus. The conditions under which chronic copper poisoning in sheep can be produced are still the subject of considerable controversy. The development of chronic intoxication is dependent upon many factors, not the least of which is the form in which the copper is ingested, the dosage levels (as a rule sustained and very high) and the amounts of molybdenum and inorganic sulphate present in the diet. The literature on copper metabolism and intoxication in man and animals is voluminous; useful reviews have been written [1-11].

Our interest in this field lies in the role of copper in the pathogenesis of geeldikkop and enzootic icterus in sheep in South Africa. Earlier work on the latter syndrome demonstrated the presence of abnormally high copper levels in the livers of fatal acute or chronic cases of the disease [12]. It was thus thought that the disease might be due to an increased copper absorption from the gut [12]. More recent work has shown that these increased liver copper levels are probably secondary to the simultaneous disturbances in urinary and biliary excretion of numerous waste products and the concomitant severe haemolysis [12-17]. The same disturbances of copper metabolism occur in the related syndrome geeldikkop [12, 13].

Since the excretion of copper by the sheep is strictly limited [12, 14] one of the main arguments in our hypothesis regarding the aetiology of these syndromes was that the assimilation of dietary copper must necessarily also be strictly limited. This paper contains the results of our studies in this regard.

MATERIALS AND METHODS

Adult Merino wethers were used throughout this work and, with the exception of those rendered copper deficient, were maintained on a diet of green lucerne hay, crushed maize and water given *ad libitum*. Copper depletion was induced in three sheep used for experiment 4 by feeding them for four and a half months on a diet containing 250 ppm (dry matter) of molybdenum (as ammonium molybdate).

Common bile duct cannulation of the animals concerned was performed as described elsewhere [18] and urine and faeces were collected by means of standard equipment in use in our laboratories for this purpose [18].

Isotopic Cu-64 was produced for us by the Atomic Energy Board of South Africa in

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their SAFARI I reactor. This Cu-64 was presented to us as cupric chloride in physiological saline solution (specific activity; 1-5 mc/mg Cu). Cuprous (Cu-64) chloride with similar specific activity, was also obtained from the same source.

Counting of samples was performed with a Philips Automatic Well-type Scintillation Detector (Type P W 4003) equipped with a $1\frac{3}{4}$ x 2" NaI/TL crystal.

Heparin was used throughout as anticoagulant for all blood samples.

RESULTS

Experiment 1. Four Merino wethers provided with biliary cannulae to the exterior were used. Each animal was given an intra-abomasal injection, following laparotomy under *local anaesthesia*, of 1 *mg copper* (as $CuCl_2$ equivalent to 5 mC Cu-64) in physiological saline. Blood specimens were collected from these animals every 20 minutes for the first four hours after injection and thereafter at 22 hours after injection.

Bile, urine and faeces were collected as voided during the 30 hours after injection and the respective samples concerned were pooled for counting. The relative amounts of copper recovered from the faeces, urine and bile samples from the four sheep or found to be present in their blood during the experimental period are presented in Table 1 as mean percentages of the total radio-activity of the injected material.

The experiment lasted for 30 hours after injection of the Cu-64. Because of the short half-life (12.8 hrs.) of this isotope, the experimental period covered probably little more than two complete turnovers of rumen contents. Although we were able to recover in the faeces of these animals only about 35% of the radio-activity we administered to them, our figures for activity in blood, bile and urine indicated negligable uptake of the isotope.

Experiment 2. Two Merino wethers provided with biliary cannulae to the exterior were used for this work. Each animal was given 1 mg copper (as $CuCl_2$ equivalent to 5 mC Cu-64) in physiological saline by intra-ruminal injection, directly through the abdominal wall. Blood, bile, urine and faeces samples were collected and prepared for counting as described in the previous experiment. The results are presented in Table 1 in the same manner as for Experiment 1. The duration of this experiment was again 30 hours. The mean relative quantity of radio-active material recovered from the faeces of the two animals (62%) is almost double that of the previous experiment. The handling of the abomasum in the first four animals coupled with the trauma of abdominal surgery could have markedly reduced gastro-intestinal motility and hence contributed to the lower recovery figures. The relative quantities recovered from blood, bile and urine are almost identical to those found in the first experiment.

TABLE 1: MEAN PERCENTAGE RECOVERY OF RADIO-ACTIVE MATERIAL ADMINISTERED TO ANIMALS USED IN EXPERIMENTS 1 AND 2, IN FAECES, BLOOD, BILE AND URINE

Medium Examined	Experiment 1 % of total activity recovered	Experiment 2 % of total activity recovered
Faeces	35.0	62.0
Blood	0.006	0.003
Bile	0.008	0.005
Urine	0.1	0.1
Excretion Ratios	**Experiment 1**	**Experiment 2**
Urine : Bile	12 : 1	20 : 1
Bile : Blood	4 : 3	5 : 3

The two sets of data indicate an almost negligible uptake of the label by the six animals concerned over the thirty-hour experimental period of at least two complete rumen turnovers. Assuming a normal whole blood copper level of 1.0 mcg/ml and an average total blood copper of 2000 mcg per sheep [10-12], the amount of copper label recovered from the blood represents no more than 0.002% of the total blood copper. The results obtained by the two routes of administration were virtually identical.

The data presented in Table 1 indicate that contrary to earlier findings [14-17], the urinary excretion of copper might be quantitatively more important than the biliary route when single doses of copper in excess of the normal daily intake are given.

Experiment 3. Following complete disappearance of radio-activity from the faeces, blood, bile and urine of the animals used in Experi-

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ment 2, two of these sheep were each given 1 mg of copper (as $CuCl_2$) equivalent to 5 m C Cu 64 by intravenous injection. One sheep was slaughtered three hours and the other 40 hours after injection. Immediately after slaughter various tissues were removed for monitoring for radioactivity. The results are presented in Table 2. The figures shown here represent the percentage of total radio-activity found in the tissues of each organ mentioned. All radioactivity was completely cleared from the circulating blood of each animal within 15 minutes of injection. The results shown in Table 2 indicate that it was almost entirely removed from the circulating blood by the liver and retained by this organ for at least 40 hours. The kidneys take up an appreciable amount of the isotope soon after its administration, but appear to rid themselves of the load fairly rapidly. This finding may be associated with an apparently greater activity of the urinary excretory mechanism, as compared to the biliary system. It is remarkable that appreciably more of the isotope is located and retained in the left lobe of the liver than in the right.

TABLE 2: PERCENTAGE DISTRIBUTION OF Cu-64 RADIO-ACTIVITY IN TWO SHEEP FOLLOWING INTRAVENOUS ADMINISTRATION

Tissue	Sheep No. 1 3 hours after injection (%)	Sheep No. 2 40 hours after injection (%)
Left lobe of liver	50.5	57.5
Right lobe of liver	24.3	31.6
Kidney	16.2	4.0
Lungs	2.4	2.3
Spleen	1.2	0.6
Head	1.5	1.1
Heart	0.8	1.1
Rest of carcass	3.0	1.7

Experiment 4. This experiment involved two adult Merino wethers maintained on the rations of lucerne and maize mentioned earlier and three sheep depleted of copper by administration of large amounts of molybdenum. The former two animals, designated sheep A and B, could be considered as being on a nutrionally adequate copper intake. The latter three sheep were designated C, D and E. Each sheep was given 1 mg of Cu (as $CuCl_2$, equivalent to 1 mC Cu-64) in physiological saline. Sheep A and C received this by intravenous injection and sheep B, D and E by intra-ruminal injection directly through the abdominal wall. Blood samples were collected from all five animals 2, 4, 6, 12, 22, 24 and 26 hours after injection. The results obtained from these animals are presented in Figure 1, in

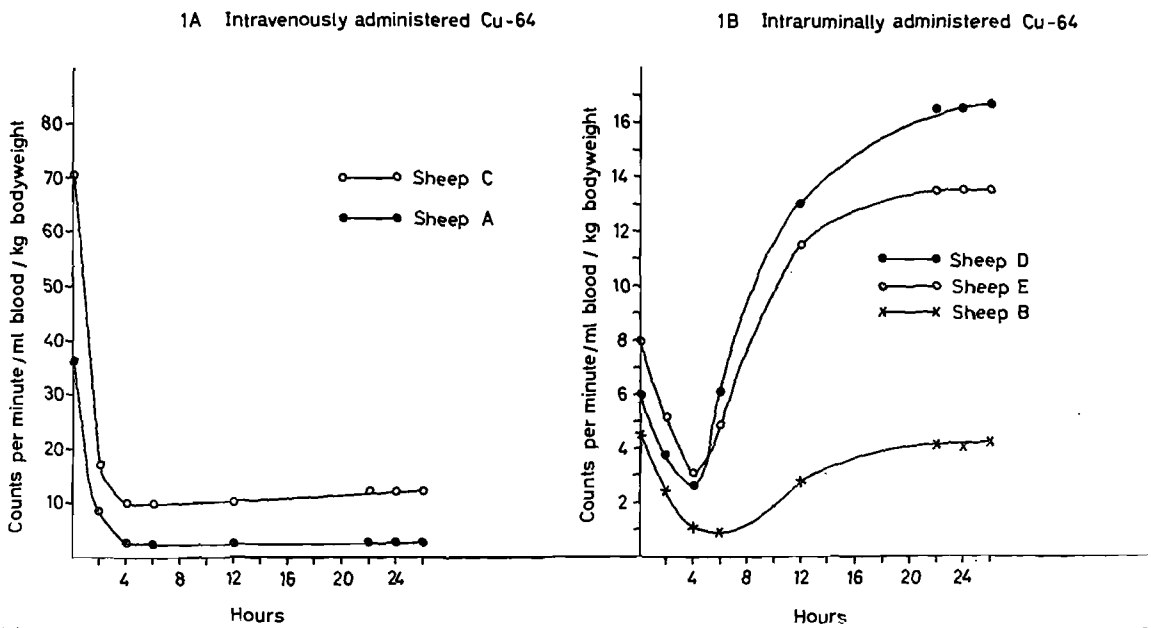

Fig. 1 : (A) Radioactivity in blood following intravenous administration of Cu-64, expressed as cpm / ml blood / Kg bodyweight plotted against time; (B) Uptake of copper from the gut in copper depleted- and normal sheep as reflected by blood levels of radioactivity.

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which blood radioactivity expressed as cpm/ml blood/kg bodyweight is plotted against time after injection. It is apparent from figure 1 A that although the rate of clearance of the copper from the blood of sheep A and C is apparently initially the same, the copper-depleted animal C retains more of the activity in its blood, from 4 hours after injection onwards, than does its "normal" counterpart.

Figure 1 B indicates quite clearly that there is a marked difference in the rate of copper uptake from the gut of copper-depleted and normal sheep before steady-state concentrations are reached in the blood of all these animals at approximately 20-22 hours after injection. The copper depleted animals apparently absorbed copper more rapidly than their normal couterpart and rapidly attained and maintained appreciably higher blood levels of the label than the latter animal. All three curves in this figure exhibit a marked inflexion during the first six hours after administration. This could be due to differences in ruminal and later intestinal absorption or, assuming that all the copper label was absorbed from the gut during the four hours following dosing, the inflexions might represent trapping of copper by the liver as demonstrated in the previous experiment, with subsequent release, possibly as caeruloplasmin into the blood stream. We are inclined to accept the latter hypothesis on the basis of our findings.

Urine and faeces were collected from four of the five animals during the twenty-six hour period following administration of the copper isotope. The results are presented in Table 3 as percentages of the total activity recovered from urine and faeces. These data indicate that after intraruminal injection the copper-depleted animals excreted about two-thirds less of the copper label in their faeces than their normal counterpart. No difference was apparent between the three animals as far as urinary excretion was concerned. The figures for copper-depleted Sheep C, which received the label by intravenous injection, are interesting from two aspects. They illustrate once more the negligible amounts of copper which leave the body following intravenous administration of small doses of the element and further they indicate, that in this particular animal, intestinal excretion either via the bile or the mucosa of the intestinal tract, was greater than the urinary excretion.

TABLE 3: URINARY AND FAECAL EXCRETION OF Cu-64 AFTER ADMINISTRATION TO COPPER-DEPLETED AND NORMAL ANIMALS

Sheep No.	Faeces % of total activity recovered	Urine % of total activity recovered
B	27.2	0.005
D	8.1	0.005
E	8.8	0.009
C	0.2	0.02

Experiment 5. Chronic copper intoxication is generally produced experimentally by prolonged administration of large and quite unphysiological doses of the element [10, 11]. Since our data so far indicated a negligible uptake of the element from the gut in animals in an adequate copper status and a somewhat increased uptake in those which were copper depleted, we decided to investigate the uptake of Cu-64 when given together with large doses of stable carrier $CuCl_2$. For this purpose we used sheep B (normal copper status) and sheep D and E (copper-depleted) from the previous experiment, once the residual activity left in their bodies from this experiment had disappeared. Each animal received 1 mg copper (= 1 mC Cu 64) together with the following amounts of $CuCl_2$ in physiological saline, by one single intra-ruminal injection: sheep B, 10 mg Cu; sheep D 100 mg Cu; Sheep E 1000 mg Cu. The results were identical in every instance to those obtained in the previous experiment. In other words, the administration of up to 1000 mg of copper in a single dose does not increase the absorption of the element in any individual sheep even when such sheep are apparently copper-depleted to a certain degree. There appears to be a distinct physiological control over the entry of copper from the gut into the body of the sheep.

The form in which copper is present in the diet is known to influence its absorption [3-5, 7]. We administered the soluble chloride. Since copper ions will enter into combination with basic phosphate, bicarbonate and hydroxyl ions in the rumen and intestinal tract, we decided to investigate the form in which the element appeared in the faeces. Aliquots of well mixed faeces from

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the above animals were extracted as follows: (i) one extraction with cold 1N HCl; (ii) two extractions with hot 1 N HCl, the extracts being combined; (iii) one extraction with cold 1 N KOH and (iv) one extraction with cold 0.05 M phosphate buffer, pH 7.4. The cold acid extracted only 20% of the activity in the sample, while the cold base and buffer extract only 10% and 5% respectively of the activity in the aliquots concerned. Hot 1 N HCl on the other hand removed 60% of the activity after brief shaking. The remaining 40% could not be removed with this mild treatment. From these few brief experiments we conclude that soluble inorganic copper is converted in the intestinal tract mainly into basic inorganic salts of low solubility.

Experiment 6. In this experiment we investigated whether the administration of cuprous Cu-64 as Cu_2Cl_2 produced results different to those obtained with the cupric salt. Two sheep were given 1 mC Cu-64 each (as Cu_2Cl_2) by intravenous and intraruminal injection. The experiment was carried out as described under Experiment 2 and the results obtained were identical to those found in this experiment. It is apparent that the cuprous ion is rapidly oxidized to the higher valency state in the rumen or liver of the sheep and then handled as cupric copper.

DISCUSSION

The absorption and transport of copper has been extensively studied in dogs, small laboratory animals and man [3, 19-25]. In spite of this, little is known of the mechanisms of absorption of copper and a precise definition of the sites of absorption is lacking [19]. The form in which copper is present in the diet is known to affect its absorption, e.g. neutral or anionic complexes such as are found in herbage are more available to copper deficient animals than copper in salts like cupric sulphate [5].

After oral administration to dogs and small laboratory animals copper levels in the blood rise rapidly over a period of 2-5 hours [19]. The copper responsible for the rise is present as loosely bound plasma albumin copper and protein bound erythrocyte copper. It appears to be transported by the albumins immediately after entry into the systemic circulation irrespective of whether it was given orally, injected as such or added *in vitro* to plasma. Albumin-bound copper is transferred to cuproproteins in the liver and then reappears in the blood after a short but variable time in association with the plasma α_2-globulins and particularly caeruloplasmin [3, 19-21]. Caeruloplasmin has been thought to regulate absorption of copper from the gut via a feed back mechanism [20]. The validity of this hypnothesis, however, has been questioned [26]. In contrast to iron it seems that in the animals studied copper homeostasis is accomplished by an adjustment of the rate of excretion to that of absorption [3].

The liver is the chief organ of copper storage in domestic animals and man [1, 3, 5, 6, 28-30]. The sheep is known to have a pattern of copper metabolism different from other species. The concentration of copper in the liver is normally higher than in most species. It has the ability to complex quite large amounts of copper in relatively stable form and to lose that excess at a very slow rate of elimination [28, 29]. Because of the ruminant portal system copper absorbed from the small intestine tends to be located first in the right part of the liver, while in copper deficiency lowest levels of stored copper are encountered first in the left lobe [19].

The main route of excretion of copper from the body of man, dogs and small laboratory animals has been shown to be via the bile. Urine, sweat and intestinal mucosal constitute secondary channels [3, 5, 19]. The rate of urinary copper excretion can be correlated with the plasma concentration of non-caeruloplasmin copper. When isotopic copper is administered to humans the greatest rate of urinary excretion is noted in the first two hours following its administration [5-22]. The close relationship between the non-globulin copper concentration and urinary copper excretion suggests that copper loosely bound to albumin is the main source of copper in urine [5, 32]. A considerable diurnal variation in urinary copper excretion has been demonstrated in normal adults [35]. Bush and coworkers [32] have demonstrated that intravenous administration of Cu-64 to healthy human subjects is followed by a greater rate of urinary elimination compared to biliary excretion than when the element is given orally.

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The data obtained by us in the current series of experiments demonstrate a number of important factors with regard to copper metabolism in the sheep. In the first instance, excretion of the element, once it has been assimilated by the sheep's body, is limited. The data presented here are conflicting. The primary route of excretion of the element was established previously by conventional chemical means as being via the bile [14]. Experiments 1 and 2 demonstrate that single small doses of copper, given intra-abomasally or intraruminally, are followed by an appreciably greater urinary than biliary excretion over the 30 hour experimental period. On the other hand, intravenous injection of the element into sheep not supplied with biliary cannulae was followed by a greater faecal than urinary excretion (sheep C in Experiment 4: see Table 3). Intravenous injection of copper loads the liver rapidly with this element and presumably forces the overflow of unbound copper to follow a biliary rather than a urinary excretory route. Whatever the mode of administration is, the fact remains that once copper enters the systemic circulation of the sheep its excretion is limited. The data presented in Table 3 demonstrate that the meagre urinary excretion remains essentially unchanged in copper depleted animals even when uptake from the intestinal tract is increased.

It is apparent from the data presented here that the intestinal tract of the sheep behaves cuprophobically towards reasonable intake levels of the element. This is analagous to the regulatory mechanism known to exist in many species for the intestinal uptake of iron. Copper absorption by the sheep seems to be regulated very efficiently. The mechanism permits the entry of more copper up to a certain and undetermined threshold in copper-depleted sheep, but even in such animals it appears to be capable of resisting the entry through the intestinal mucosa of completely unphysiological and ridiculously high doses of the element. Although it is probable that single doses higher than those used by us in experiment 5 will break this protective mechanism down completely, it seems that in practice chronic copper intoxication in the sheep can only occur under highly abnormal circumstances.

Our results support those of McCosker [10, 11] and others cited by him who demonstrated that very prolonged dosage of unphysiological amounts of copper resulted in chronic intoxication in sheep. This is known to occur under natural conditions: (a) when there is a straight forward long-continued high gross intake of highly cupriferous vegetation. (i.e. plants containing 50-60 ppm of copper or more); (b) when molybdenum levels are very low in pastures of normal copper content and (c) when liver storage of copper is abnormally raised due to failure of hepatic excretion following various forms of impairment of liver function [4, 12].

It seems likely therefore that under the conditions which generally pertain in South African pastures (including also certain parts of the Karoo where many plants may contain up to 28 ppm of the element or more) chronic copper intoxication will not occur if the molybdenum status of the animals concerned is physiologically adequate.

The results demonstrate again the well-known fact that the liver of the sheep retains administered copper avidly. It clears the element rapidly from the blood, and if our interpretation of Figure 1 is correct, releases it slowly into the plasma again firmly bound to protein.

At present some of this work is being repeated with Cu-67, which has a longer half-life and thus allows the study of the absorption of the element over a period covering the passage of all the labelled copper through the entire gastro-intestinal tract.

These studies indicate that radioactive copper dosed orally or intraruminally may be useful in determining the rate of passage of ingesta down the ovine digestive tract, because of the meagre absorption of the element.

ACKNOWLEDGEMENTS

The Chief, Veterinary Research Institute is thanked for permission to publish this paper.

REFERENCES

1. Eden A. 1943 *J. comp. Path Therap.* 53:90
2. Bull L. B., Dick A. T., Keast J. C. & Edgar G. 1956 *Austr. J. agric. Res.* 7:281
3. Gubler C. J. 1956 *J. Am. med. Ass.* 161:530

Reproduced by Sabinet Gateway under licence granted by the Publisher (dated 2011)

4. Russel F. C. & Duncan D. L. 1956 *Minerals in pasture: Deficiencies and excesses in relation to animal health.* 2nd Ed. Commonwealth Bureau of Animal Nutrition Tech. Comm. No. 15. Commonwealth Agricultural Bureaux, Farnham Royal, Slough, Bucks.
5. Adelstein S. J. & Vallee B. L. 1962 "Copper" in *Mineral Metabolism* Vol 2 B Ed. C. L. Comar F. Bronner. New York & London. Academic Press
6. Todd J. R. Gracey J. F. & Thompson R. H. 1962 *Br. vet. J.* 118:482
7. Underwood E. J. 1962 *Trace elements in human and animal nutrition* 2nd Ed. New York & London, Academic Press
8. Schütte K. H. 1964 *The biology of the trace elements* London, Crosby Lockwood & Son, Ltd.
9. Gallagher C. H. 1964 *Nutritional factors and enzymological disturbances in animals* London, Crosby Lockwood & Son, Ltd.
10. McCosker P. J. 1968 *Res. vet. Sci.* 9:91
11. McCosker P. J. 1968 *Res. vet. Sci.* 9:103
12. Brown J. M. M. 1968 *Biochemical Studies on Geeldikkop and Enzootic icterus* DVSc Thesis. University of Pretoria
13. Brown J. M. M. 1963 *Ann. N.Y. Acad. Sci* 104:504
14. Brown J. M. M. 1968 *Jl S. Afr. vet. med. Ass.* 38:365
15. Brown J. M. M. 1967 *Jl S. Afr. vet. med. Ass.* 38:311
16. Brown J. M. M. 1968 *Jl S. Afr. vet. med. Ass.* 38:383
17. Brown J. M. M. 1968 "The chemical pathology of ovine icteric states. 3. Icterogenin induced cholestasis. *Jl S. Afr. vet. med. Ass.* In Press
18. Brown J. M. M. 1959 *Jl S. Afr. vet. med. Ass.* 30:395
19. Van Koetsveld E. E. 1954 *Tijdschr. Diergeneesk.* 79:495
20. Scheinberg I. H. & Morrell A. G. 1957 *J. clin. Invest.* 36:1193
21. Bearn A. G. & Kunkel H. G. 1954 *J. clin. Invest.* 33:400
22. Bearn A. G. & Kunkel H. G. 1955 *J. Lab. clin. Med.* 45:623
23. Earl C. J., Moulton M. J. & Selverstone B 1954 *Am. J. Med.* 17:205
24. Walshe J. M. 1957 *Br. med. Bull.* 13:132
25. Cumings J. N., Goodwin H. J. & Earle C. J. 1955 *J. clin. Path.* 8:69
26. Sternlieb I., Morell G., Tucker W. D., Green M. W. & Scheinberg I. H. 1961 *J. clin. Invest.* 40:1834
28. Beck A. B. 1956 *Aust. J. Zool.* 4:1
29. Beck A. B. 1963 *Aust. J. Agric. Res.* 14:129
30. McDougall E. I. 1947 *J. agric. Sci.*, Camb. 37:329.
31. Gubler C. J., Brown H., Markowitz., Cartwright G. E. & Wintrobe M. M. 1957 *J. clin. Invest* 36:1208
32. Bush J. A., Mahoney J. P., Markowitz H., Gubler C. J., Cartwright G. E. & Wintrobe M. M. 1955 *J. clin. Invest.* 34:1766
33. Van Ravensteyn A. H. 1944 *Acta Med. Scand.* 118:163
34. Mahoney J. P., Bush J. A., Gubler C. J., Moretz W. H., Cartwright G. E. & Wintrobe M. M. 1955 *J. Lab. clin. med.* 46:702
35. Butler E. J. & Newman E. E. 1956 *J. clin.Patch.* 9 : 157

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Jl S. Afr. vet. med. Ass. 39 (2): 21-24 1968

ADVANCES IN GEELDIKKOP (*Tribulosis ovis*) RESEARCH 10.

The Symptomatology and Gross Pathology of Enzootic Icterus.

H. P. A. DE BOOM* AND J. M. M. BROWN**

SUMMARY

The symptomatology and gross pathology of enzootic icterus are described. Two forms of the disease are generally encountered *viz.* an acute haemolytic episode and the more common chronic wasting form characterised by lowgrade intravascular haemolysis and kidney pathology. The conditions unnder which either form occurs are mentioned together with their main symptoms and autopsy findings. Some thoughts are expressed regarding the progress of liver and kidney lesions in enzootic icterus and the causes of death in this disease.

INTRODUCTION

Enzootic icterus is an acute or chronic haemolytic syndrome of sheep very closely related to geeldikkop [1]. Both constitute two outwardly different extreme manifestations of a single disease entity, possibly sub-clinical chronic selenium intoxication [1]. The acute episodes of both conditions can be precipitated by various non-specific forms of stress but it is thought that the most important factor in this regard is a mild myotropic virus infection [1-3]. The acute episodes of enzootic icterus, which are often fatal within a short time, are characterised by intense icterus, severe haemolytic anaemia, severe renal pathology, severe gastro-intestinal stasis and many biochemical disturbances of note which will be described in subsequent papers. Chronic forms of the disease are common, often dominating any particular large scale outbreak and are characterised by marked anaemia and renal lesions.

The epizootiology of the disease and historical notes on research into the condition have been described in a previous communication [4].

GENERAL DISCUSSION

Two main forms of enzootic icterus may be recognised, depending largely on the nature of the precipitating agent or factors. Acute haemolytic crises represent one form and are generally seen in isolated instances throughout the year in flocks in the areas where the disease is enzootic or amongst animals which have been removed from these areas [1, 4]. The acute episodes are precipitated by such non-specific forms of stress as transport over long distance by rail or road, changes in diet, intercurrent infections, handling, dosing with anthelmintics and inoculation with live vaccines [1, 4]. Cases have been seen to appear unpredictably amongst individual animals at varying intervals after having been removed from enzootic areas, in the absence of any definable stress conditions [4].

Chronic forms of the disease can be detected on careful examination of the animals in flocks in the areas where the disease is enzootic They are generally found on a large scale during the severe epizootics of the disease which occur periodically in the affected areas and seem to follow widespread dramatic changes in the natural grazing [4]. It is possible that some outbreaks of this nature are precipitated by the same infectious agent which is operative in geeldikkop outbreaks [1]. Chronic forms of the disease are also frequently found in flocks where an outbreak of geeldikkop is rampant [1, 5].

THE ACUTE HAEMOLYTIC CRISES

Cases presenting acute attacks of the disease are incorrectly regarded as representing the typical form of the disease. This is largely because the acute episode is drama-

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tic and easily recognised and was the form most commonly seen by the earlier workers *at Onderstepoort*[1, 4, 6]. The acute form appears as a rule sporadically amongst individuals in flocks in the affected areas throughout the year; sporadically amongst animals moved out of these areas and to a fair extent in any flock during extensive outbreaks of the disease in affected areas. It is common amongst animals moved long distances in motor and rail transport and is typically an acute explosive haemolytic crisis.

Affected animals are severely icteric and anaemic, and often have a fever of 104-106° and haemoglobinuria. Malaise, apathy, anorexia and rapid loss of condition are prominent symptoms and are accompanied by hyperpnoea, a strong bounding pulse in the early stages and frequently cardiac arrhythmia. There is generally evidence of photosensitization in the form of a *mild* swelling of the ears, eyelids and lips, rhinitis, blepharitis, conjuctivitis and keratitis in the cases which occur during natural epizootics of the syndrome. These symptoms are not generally observed in cases precipitated by travel over long distances or in sporadic cases which occur amongst stabled animals. Whatever the precipitating factor, the condition is almost invariably accompanied by an extremely severe gastro-intestinal stasis, the like of which is seen only in geeldikkop. As a rule the animals die within one to five days of the onset of symptoms, although we have been fortunate in being able to study exacerbations of the condition in odd animals which have survived from one to three known prior haemolytic episodes[1].

The macroscopic pathology and histopathology of the acute attacks of enzootic icterus were described by de Kock in 1928[6]. His description included the following points of note: severe icterus and anaemia; methaemoglobincythaemia in some cases (this has been deduced from his description of the blood in these instances); enlargement and pigmentation of the liver with greyish-yellow zones around the central veins in most cases; marked enlargement and dark reddish purple pigmentation of the kidneys; tumour splenis, often marked in many cases, with the Malphigian bodies and trabeculae of this organ often appearing indistinct; the lymph nodes are generally swollen, oedematous and pigmented; hyperaemia and oedema of the lungs is a general finding and severe impaction of the caecum and colon is seen frequently; haemoglobinuria is a fairly common symptom, while mild hydrothorax and hydropericardium are seen occasionally. In addition to these findings we have observed ulcerations of the abomasum in some animals and marked atrophy of the adrenals and lymphoid tissue in others. Enlargement and pigmentation of the kidneys and the extremely severe gastro-intestinal stasis were the autopsy changes which were most striking. The cortices of the affected kidneys are intensely brownish-black in colour and the kidneys may be twice their normal size. Icterus is generally most intense in these animals.

The histopathology of the condition has been described in De Kock's original paper[6] and has been reviewed by Brown[1]. Pienaar and Van der Merwe[7] have made a special study of the cytomegaly and karyomegaly which are histological features in the liver of these cases and have reported the occurrence of intranuclear inclusions and globules in the megalocytes.

THE CHRONIC FORMS OF THE DISEASE

The commonest form of the disease is a chronic wasting syndrome which is encountered most generally during extensive outbreaks. It is this form which, strictly speaking, should be regarded as the typical disease. The onset of symptoms is insidious and early cases are not easily recognised. Some affected animals appear to look about for food with the rest of the flock but do not eat or drink, (even though many have been seen to stand with their muzzles immersed in water), while others merely nibble at their food and wander off listlessly. We have relied upon the Griqua shepherds on the farms concerned to pick out these cases from their flocks for us which they are able to do with unerring facility. These animals in the early stages of enzootic icterus generally have a fever of 103-105°F, their visible mucous membranes are frequently injected but sometimes anaemic and in a few instances the typical chocolate brown discolouration of a methaemoglobincythaemia has been observed. Gastro-intestinal stasis is invariably present[5].

The subsequent course of the disease may occupy three to five weeks before recovery or death occurs. It is typically a steady decline in condition, affected animals rapidly becoming severely anaemic and cachectic. They are apathetic, disinclined to eat and fall readily if chased. There is invariably marked gastro-intestinal stasis, a fever of

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103-105°, hyperpnoea, a bounding or a weak thready pulse, rapid heartbeat, mild icterus and methaemoglobincythaemia or cyanosis. The wool of these animals is easily pulled out and they have a mangy appearance. Rhinitis, conjunctivitis, blebharitis and keratitis occur frequently and are accompanied by some necrosis of the skin on the ears, eyelids and nostrils. This chronic form of the disease is subject to frequent exacerbations and remissions, the former being provoked by subjecting the animals to any stressful condition.

The chronic forms are seen mainly in adult animals and particularly in aged ones. We have seen cases in suckling lambs and hoggets although they occur rather infrequently and then only during severe outbreaks of the disease.

The autopsy findings in chronic cases have not been described before. The following are briefly the general changes observed: severe anaemia; cachexia; mild or negligible icterus, mild hydrothorax, hydropericardium and ascites; severe atrophy of the forestomachs and small intestines; impaction of the colon (which may be extremely severe); tumor hepatis and pigmentation of the liver; distention of the gallbladder with concentrated bile; mild to marked enlargement and pigmentation of the kidneys; atrophy of the spleen or, in many cases, marked tumor *lienis;* atrophy of the cortex of the lymph nodes, with scattered deeply pigmented nodules being present in the cortices of many such nodes; degeneration of the myocardium in some instance, pigmentation and oedema of the lungs (in at least 50% of cases seen); degeneration of the adrenal cortex; hyperplasia of the red bone marrow and brownish pigmentation of the skeletal system. In long standing cases the pigment is seen more in the cartilages than in osseous tissue. Ulceration of the pylorus was observed in a few instances. We have seen many cases in which there is a apparently an hypertrophic cirrhosis present in the liver associated with localized areas of atrophic cirrhosis particularly around the portal tracts and have described extreme cases in which the entire area around the portal fissure of the liver has been replaced by fibrous tissue giving this organ a dumbbell appearance [8].

The histopathology of these cases is similar to that already described, except that the lesions are milder. Portal cirrhosis, pigmentation and macrophage invasion are seen mainly in the livers of affected animals, mild nephrotic lesions are present and mild atrophic changes coupled with an accumulation of pigment bearing macrophages are generally seen in the lymph nodes. The histopathology of the cases of this nature, is described in detail elsewhere [1].

CONCLUDING REMARKS

The same tetrad of symptoms as seen in geeldikkop, *viz.* intravascular haemolysis, impairment of hepatic excretion, renal insufficiency and adrenal dysfunction are also found in enzootic icterus [1] The disturbances of liver and kidney function commence by being mild in enzootic icterus, but in acute cases the severe haemolytic episode changes the nature of the condition to that of a potentially lethal disease. Widespread liver cell death may follow the anoxia in severe cases and this in turn has asits sequel the invasion of macrophages, which is so prominent in enzootic icterus, and eventual replacement fibrosis. Succeeding non-fatal exacerbations all add their quota of fibrous tissue to the injured liver and eventually the extreme cirrhotic forms described earlier [8] are the result. The renal insufficiency changes very rapidly from the same type of metabolic derangement seen in geeldikkop to a malignant type of nephrosis in the severe enzootic icterus cases [1] Whereas the development of a biliary nephrosis is the general sequel to the renal disturbances in geeldikkop, the nephrosis in enzootic icterus is related directly to the renal ischaemia resulting from the anaemia and the rapid accumulation of free haemoglobin in the tubules of the kidneys [1].

Death in enzootic icterus is largely due to a combination of anoxia as a result of anaemia, the severe kidney lesions and adrenal insufficiency [1].

ACKNOWLEDGEMENTS

The Chief, Veterinary Research Institute, Onderstepoort is thanked for permission to publish this paper. We wish to thank particularly the farmers of Murraysburg, Rietbron, Beaufort West, Fraserburg, Hondefontein, Sutherland, Loxton, Victoria West, Britstown and Vosburg who have placed most of the cases studied by us at our disposal. Drs. C. W. Belonje, J. A. Badenhorst, D. E. Truter, D. J. Thornton, L. Steel, F. de St. J. van der Riet, M. van Tonder, C. Wilkens and K. M. van Heerden, have all at various times lent valuable assistance to us in the field.

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REFERENCES

1. Brown J. M. M. 1967 *Biochemical studies on geeldikkop and Enzootic Icterus.* D.V.Sc. Thesis: University of Pretoria
2. Brown J. M. M. 1964 *Jl S. Afr. vet. med Ass.* 35:507
3. Brown J. M. M. 1966 *Jl S. Afr. vet. med. Ass.* 37:454
4. Brown J. M. M. & de Boom H. P. A. 1966 *Jl S. Afr. vet. med Ass.* 37:321
5. Brown J. M. M., le Roux J. M. W. & Tustin R. C. 1960 *Jl S. Afr. vet. med. Ass.* 31:179
6. de Kock G. 1928 *13th/14th Rep. Dir. vet. Educ. Res.* p. 648
7. Pienaar J. G. & van der Merwe J. L. de B. 1966 *Jl S. Afr. vet. med. Ass.* 37:53
8. Brown J. M. M. & de Wet P. J. 1962 *Onderstepoort J. vet. Res.* 29:111

BOOK REVIEWS

ANIMAL HEALTH AND HOUSING

DAVID SAINSBURY

Bailliére, Tindall and Cassell, London, 1967, pp IX, 323; Tabs. 25; Figs. 123;
Publ. price: 50/-

This book has reproductions of black and white photographs showing the exterior, interior and fittings of animal buildings. Plans and diagrams illustrating the layout and basic principles of ventilation and other features of buildings are given. Graphs showing the effects of environmental factors on animals are included. At the end of each chapter there is a list of references to articles for those who wish for further or more detailed information. The book is printed on good quality paper with a distinct, well spaced print which is easy to read.

There is a wealth of essential information on building construction and the features necessary for the efficient management, comfort and promotion of health in animals. Special attention has been given to dairy hygiene and the buildings necessary for dairy cows and calves. Pigs and poultry including turkeys have been well covered. There are short chapters on beef cattle and sheep. The horse, goat, dog and cat and buildings for them have not been included.

Measures for the control of disease and transport of animals in the British Isles are outlined. A list of the acts and regulations covering these, are given.

The following suggestions can be made: In the chapter on disinfection, sodium hydroxide is discussed under sodium carbonate; it is surely important enough to have a heading of its own. A chapter on equipment essential for the handling of animals such as weighing scales, crushes and foot baths would also be a useful addition.

This first edition, which deals in a very practical way with the problems encountered in the intensive management and housing of animals will be found very useful by all concerned with animal production and disease. It can be recommended without reservation. It is written for conditions in the northern hemisphere. In countries like the Republic of South Africa building plans will have to be transposed so as to have buildings facing north or northeast instead of south or southwest and special attention will have to be given for counteracting the problem of heat in the summer months.

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Jl S. Afr. vet. med. Ass. 39 (2): 25-31 1968

ADVANCES IN GEELDIKKOP *(Tribulosis ovis)* RESEARCH

11. The Haematology of Enzootic Icterus

J. M. M. BROWN* AND H. P. A. DE BOOM**

SUMMARY

The haematology of cases representing different stages or forms of enzootic icterus is described. The development of erythrocyte and leukocyte dyscrasias during and after haemolytic episodes is discussed. The anaemia seen during various stages of the disease has been characterised.

INTRODUCTION

These studies, which have been fully documented earlier and elsewhere,[1] formed part of an extensive investigation into the chemical pathology and disturbed biochemistry of geeldikkop and enzootic icterus. The results of the studies on the haematology of enzootic icterus are summarised in the present paper.

MATERIALS AND METHODS

The place of origin and the most prominent symptoms of the cases used in these studies on the haematology of enzootic icterus are listed in Appendix 1. The majority of the sheep emanated from farms in the Fraserburg, Sutherland and Aberdeen districts during severe outbreaks of enzootic icterus in the late summer of 1957/1958 and 1961/1962. Animals from Rietbron were obtained during investigations into geeldikkop outbreaks in this area and those from Calvinia were examined on a farm in the Clarens district of the Orange Free State during a massive movement of small stock from drought-stricken areas in 1964.

The sheep concerned in this report were adult Merino ewes and wethers and have been classified as follows:-

(a) Early clinical cases of 1-5 days standing showing the first detectable signs of a haemolytic episode, viz. anaemia, icterus and hyperpnoea. They are subdivided for the purposes of discussion into

  (i) group 1 — mild cases and

  (ii) group 2 — severe cases (i.e., when first examined);

(b) Chronic cases.

Details of their symptomatology were discussed in the previous paper in this series[2]. These animals have been subdivided into (i) "post-haemolytic" cases of 7-14 days standing which were known to have just survived an acute haemolytic episode; (ii) mild chronic cases of 7 - 14 days standing, in which low-grade intravascular haemolysis was constantly present in the absence of acute manifestations; and

(iii) severe chronic cases of 7-14 days standing where animals were severely anaemic, more or less icteric and uraemic[1] but were not passing through a recognisable acute haemolytic crisis, i.e. the rate of haemolysis was somewhat more accelerated than in the previous group.

The animals 2206 - 2208, Bekker 1 - 3, NB2 - NB5, 5114, 5116 - 5121, 5126, 5128 and 5130 - 5132 were obtained during outbreaks of enzootic icterus on their farms of origin and brought to Onderstepoort for study. All the cases except animals 5114 - 4132 were kept for no longer than 24 hours after arrival before being slaughtered for histopathological studies.

Sheep 5114 - 5132 were transported for three days by road and rail from Fraserburg

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and Sutherland to Onderstepoort with the specific aim of precipitating acute attacks of the disease. On arrival they were fed teff hay and crushed maize *ad libitum*. This represented a complete change of diet sufficient to induce a severe gastro-intestinal disturbance. The symptoms listed in Appendix 1 are those observed when the animals arrived at Onderstepoort. Three weeks before movement to Onderstepoort sheep 5116, 5117, 5118, 5120 and 5127 had been classified as chronic cases on the grounds of clinical and chemical pathological examinations. Before leaving the respective farms they were considered recovered and clinically normal. The combination of subsequent stress factors, *i.e.* prolonged transport and change of diet, provoked the exacerbations in these cases to be described in this paper.

The remaining animals listed in Appendix 1 were maintained in sheltered pens at Fraserburg [1] where they were fed a diet of mixed Karoo bushes (*Pentzia spp, Aridaria spp. Chenopodium album, Atriplex semibaccata, Eriocephalus ericoides* and *Salsola* spp) *ad libitum*. In addition each received 200 g of crushed maize daily and had free access to lucerne hay and water. Standard techniques were employed for the haematological determinations.

RESULTS

The haematological data obtained from cases representing all stages of the disease are presented in Appendix 2. Normal ranges for the Merino sheep in South Africa, [1] are: packed cell volume, 33 - 56%; red cell count, 8 - 14 x 10^6/Cu mm; haemoglobin, 9 - 14.5%; white cell count, 4.9 - 9.15 x 10^3/Cu mm; MCHC, 24.1 - 38.6%; MCV, 21.3 - 41.7 Cuμ, MCH, 6.5 - 16.1 $\mu\mu$g and differential leukocyte count, N = 30 - 35%, L = 50 - 55%, M = 4%, E = 8%, B = O - 1%.

It is evident from data presented in appendix 2 that in the mild early cases, apart from sheep 2206 - 2208, the only constant changes were a definite neutrophilia and lymphocytopaenia. Very mild anaemia was found in some cases, *e.g.* 12224 and FB-2. The bloodsmears of these animals revealed nothing of note. Sheep 2206, 2207 and 2208 were anaemic and blood smear examination revealed severe anisocytosis, polychromasia and the presence of numerous Jolly bodies and normoblasts. There was no evidence of thrombocytopaenia. The anaemia in these cases was either normocytic normochromic (sheep 2207) or macrocytic hyperchromic (sheep 2206 and 2208). All three animals showed a marked leukocytosis generally due to a neutrophilia. Lymphotopaenia was evident in two of these cases (2207 and 2208).

Anaemia, generally macrocytic hyperchromic, of varying severity was present in the severe early cases (Group 2) and it was often accompanied by neutrophilia and severe lymphocytopaenia.

The posthaemolytic stage of enzootic icterus appears to be dominated by normocytic normochromic anaemia of varying severity, as is evident from the data obtained from sheep F-9, F-10 and F-11. In animal 12226 the anaemia appeared to be hypochromic microcytic and in sheep 12227 of a severe hyperchromic macrocytic type. Marked leukopaenia was evident in sheep F-10, F-11 and 12226, while animals F-9 and 12227 showed neutrophilia and lymphocytopaenia. The anaemic changes described for sheep 2206 - 2208 were seen in the bloodsmears of some of these animals as well.

Normocytic normochromic anaemia of varying severity was generally present in the mild (group 2) chronic cases. In some animals leukopaenia or mild neutrophilia and lymphocytopaenia were also evident. The severely affected group 3 of chronic cases showed mainly macrocytic normochromic anaemia. The development of the haemolytic syndrome in enzootic icterus is illustrated by the data presented in Tables 1 and 2. These contain the results of daily serial studies on the haematology of some of the cases brought to Onderstepoort from Fraserburg and Sutherland, and illustrate some of the effects of severe stress on apparently asymptomatic cases of the basic disease entity.

The animals shown in Table 1 had no known history of a previous haemolytic episode and were clinically normal ot the time they left their places of origin.

It is evident that the haemolytic episode developed rapidly after their arrival at Onderstepoort and in sheep 5121, 5128 and 5132 terminated in death 6 days later. Animals 5114, 5119, 5130 and 5131 all died during the course of the following week. The interval between the initial application of the combined stress factors and the appearance of

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TABLE 1: SERIAL STUDIES ON THE HAEMATOLOGY OF MILD CASES OF ENZOOTIC ICTERUS (ON DAYS AFTER ARRIVAL AT ONDERSTEPOORT)

Sheep No.	Determination	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	
5114	RCV	45	36	25	21	19	16	MCHC = 26.87
	RCC	9.22	4.73	4.50	4.36	3.98	3.59	MCV = 44.56
	Hb	11.30	8.80	4.45	4.80	4.62	4.30	MCH = 11.97
	WCC	6,320	7,850	15,600	15,750	16,250	33,600	
5119	RCV	34	28	25	21	17	16	MCHC = 36.87
	RCC	7.61	6.68	6.53	3.71	3.00	3.61	MVC = 44.32
	Hb	7.90	7.12	7.12	5.95	5.20	5.90	MCH = 16.34
	WCC	2,370	4,300	5,200	11,700	7,750	15,650	
5121	RCV	28	20	12	12	12	Died	MCHC = 34.91
	Hb	7.88	5.29	2.42	1.86	2.01		MCV = 59.7
	RCC	9.10	10.50	3.75	4.28	4.19	Met Hb at death	MCH = 20.84
	WCC	10,250	10,850	11,750	9,800	10,500		
5128	RCV	29	30	27	22	20	Died	MCHC = 35.00
	RCC	10.61	12.61	7.90	5.93	5.01		MCV = 39.92
	Hb	9.10	9.10	8.10	7.62	7.00		MCH = 13.97
	WCC	4,200	12,850	8,950	15,800	16,820		
5130	RCV	30	27	21	20	19	21	———
	RCC	8.20	6.35	4.56	4.25	4.62	4.32	———
	Hb	8.20	6.90	—	—	—	—	———
	WCC	4,200	6,250	7,950	7,450	6,850	2,500	
5131	RCV	37	30	25	22	18	16	———
	RCC	9.57	8.92	6.20	5.90	—	4.02	———
	Hb	9.10	—	6.20	5.50	5.00	4.17	———
	WCC	1,120	—	4,500	—	2,400	10,100	
5132	RCV	38	36	16	12	11	Died	MCHC = 34.09
	RCC	10.28	10.47	3.19	2.13	1.90		MCV = 57.89
	Hb	10.50	6.17	4.80	3.92	3.75		MCH = 19.73
	WCC	4,560	3,520	15,100	16,750	12,250		

N.B. RCV = red cell volume (%); RCC = red cell count (10^6/Cu mm); WCC = white cell count (10^3/Cu mm); Hb = haemoglobin (g%); MCHC = Mean Corpuscular haemoglobin content; MCV = mean cell volume; MCH = mean cell haemoglobin.

the haemolytic syndrome was in all cases 6-7 days. The absolute haematological indices given in Table 1 have been calculated from the data obtained on the last day of study (i.e. day 5 or 6 as the case may be). It can be seen from these that the anaemia which developed in these animals was generally macrocytic hyperchromic. In the case of animal 5119 it was macrocytic normochromic and in the case of 5128, normocytic normochromic.

In five out of the seven cases marked leukocytosis developed concurrently with the anaemia.

Methaemoglobincythaemia was clinically obvious in animal 5121 at the time of death.

The four animals shown in Table 2 were all classed as chronic cases of the disease when examined at their places of origin about a month before being sent to Onderstepoort. Just prior to this event they appeared to have recovered from the earlier attack. As can be seen from the data in Table 2 the ef-

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TABLE 2: SERIAL STUDIES ON THE HAEMATOLOGY OF CASES SHOWING A KNOWN EXACERBATION OF THE HAEMOLYTIC SYNDROME IN ENZOOTIC ICTERUS (ON DAYS AFTER ARRIVAL AT ONDERSTEPOORT)

Sheep No.	Determination	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
5116	RCV	38	—	25	—	16	—	—
	RCC	—	6.38	—	3.86	—	2.57	—
	Hb	10.50	—	9.42	—	7.12	—	—
	WCC	4,200	6,820	—	30,100	—	20,600	—
5117	RCV	36	29	19	17	16	12	—
	RCC	9.12	9.23	6.49	4.97	5.24	5.00	—
	Hb	10.90	7.62	5.75	3.75	3.00	—	—
	WCC	9,450	15,800	5,550	14,000	16,350	15,000	—
5118	RCV	30	33	28	20	21	15	11
	RCC	8.22	7.97	7.79	5.60	4.95	3.87	2.80
	Hb	8.50	6.17	6.65	6.40	6.90	7.30	6.20
	WCC	4,150	5,500	3,700	2,500	7,700	11,950	4,350
5120	RCV	38	40	33	29	25	24	12
	RCC	13.32	9.01	10.40	8.90	6.63	6.30	5.80
	Hb	10.50	11.75	8.50	7.35	8.50	7.62	5.75
	WCC	4,025	8,150	7,000	10,150	8.200	12,500	8,600

N.B.: The abbreviations in this table are as given at the foot of Table 1.

fects of the combined stress (duration 6-7 days) provoked a severe exacerbation of the subclinical haemolytic syndrome. Animal 5116 died during the following week, animals 5118 and 5120 two weeks later, and sheep 5117 apparently recovered, but died about two months later from a further attack. Haematological indices were calculated from the data of day 5 in the case of 5117 and from the data of day 7 in the case of animals 5118 and 5120. The anaemia in all three instances was found to be normocytic (hyperchromic in the case of 5118 and 5120 and hypochromic in the case of 5117). The development of anaemia in these animals was once more accompanied by leukocytosis, neutrophilia and lymphocytopaenia.

No deviation from normal was seen in the erythrocyte sedimentation rates of any of the animals studies (Appendix 2).

DISCUSSION

Normocytic anaemias may result from sudden loss of blood, destruction of blood, lack of blood formation or dilution of blood with fluid. Under all these circumstances, other things being equal, the remaining red corpuscles are as normal in size or haemoglobin content as they were before blood loss, destruction or dilution had occurred. The total plasma protein figures of the animals examined are given in Appendix 2. These figures were obtained from the same blood used for the haematological studies indicated in each case and were generally within normal limits. It can be assumed, therefore, that haemodilution due to water retention does not contribute towards the low values for the haemoglobin, haematocrit and red cell counts.

Blood regeneration of varying degrees occurs in acute posthaemorrhagic anaemia, in haemolytic and sometimes even in aplastic anaemias. When the stimulus to new red cell formation is great and the capacity for haemopoiesis is good, a large number of immature cells may pass into the circulation and the anaemia may appear to be macrocytic and hyper- or hypochromic. Macrocytosis may be marked in acute haemolytic anaemia and it may be prolonged in the chronic forms[3]

Enzootic icterus is of rapid onset following stress, as shown, and the anaemia is typically *hypocythaemic normocytic normochromic* or *hypocythaemic macrocytic hyperchromic (or normochromic)* in the early stages of the

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disease. The development of anaemia is accompanied generally by leukocytosis, which is due to an absolute neutrophilia and lymphocytopaenia in most cases.

The "posthaemolytic" blood picture in enzootic icterus is that of normocytic normochromic or macrocytic hyperchromic anaemia as before, which may be accompanied by a severe leukopaenia or relative neutrophilia and absolute lymphocytopaenia. These changes were first observed by De Kock in 1928[4]. The same changes are seen in mild and severe chronic cases of the disease.

Leukopaenia and lymphocytopaenia are frequent findings in stress conditions in sheep[1]. The existence of adrenal insufficiency in enzootic icterus has been demonstrated[1]. Absolute neutrophilia is generally associated with acute or localised infections. It is, however, a very prominent finding in acute haemolytic episodes and intoxications associated with metabolic disorders such as uraemia[3] Uraemia is known to be a pronounced symptom in enzootic icterus[1]. The neutrophilia in this disease is accompanied by a very marked "shift to the left", as is found in geeldikkop[1];[5].

ACKNOWLEDGEMENTS

The Chief, Veterinary Research Institute, Onderstepoort, is thanked for permission to publish this paper. We wish to thank particularly the many farmers in the areas where the disease is enzootic who have unhesitatingly placed at our disposal the cases used in this work.

REFERENCES

1. Brown J. M. M. 1967 Biochemical Studies on Geeldikkop and Enzootic Icterus. Thesis University of Pretoria.
2. de Boom H. P. A. & Brown J. M. M. 1968 *Jl S. Afr. vet. med. Ass.* This Journal.
3. Wintrobe M. M. 1947 *Clinical Haematology.* Philadelphia. Lea and Febiger.
4. de Kock G. 1928 *13th/14th Rep. Dir. vet. Educ. Res.* p. 648
5. Brown J. M. M., le Roux J. M. M. & Tustin R. C. 1960 *Jl S. Afr. vet. med. Ass.* 31:179

Appendix 1: Details of sheep used in haematological studies of enzootics icterus

A. Early clinical cases (1 - 5 days standing)
(1) Group 1: Mild cases when first examined:-

Sheep No.	District of origin	Relevant clinical data
2206	Sutherland	Listlesness, anaemia
2207	Sutherland	Listlesness, anaemia
2208	Sutherland	Fever, listlessness, anaemia
12223	Fraserburg	Very mild icterus, anaemia
12224	Fraserburg	Listlessness, anorexia
12225	Fraserburg	Anorexia
12228	Fraserburg	Nothing specific
12229	Fraserburg	Anorexia
Bekker 3	Rietbron	Icterus, anorexia
FB-2	Fraserburg	Very weak, anaemia, hyperpnoea, apathy
5114	Sutherland	Weakness, listlessness, anorexia
5119	Sutherland	Weakness, listlessness, anorexia
5121	Fraserburg	Weakness, listlessness, anorexia
5128	Fraserburg	Ewe with suckling lamb, weakness, listlessness
5130	Sutherland	Nothing unusual when first examined
5131	Sutherland	Nothing unusual when first examined
5132	Sutherland	Nothing unusual when first examined
5116	Fraserburg	Exacerbation. Severely anaemic, hyperpnoea
5117	Fraserburg	Nothing unusual when first seen. Exacerbation later
5118	Fraserburg	Nothing unusual when first seen. Exacerbation later
5120	Fraserburg	Nothing unusual when first seen. Exacerbation later

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(2) Group 2: Severe early cases:

Sheep No.	District of origin	Relevant clinical data
Bekker 1	Rietbron	Severe icterus, dyspnoea, lesions of photosensitization
Bekker 2	Rietbron	Severe icterus, dyspnoea, lesions of photosensitization
Beker 4	Rietbron	Severe icterus, hyperpnoea, lesions of photosensitization
FB-12	Sutherland	In extremis, Severe icterus, anaemia, hyperpnoea
F6	Aberdeen	Temp. 105°, Cachexia, Rhinitis, keratitis
F8	Aberdeen	Severe icterus, anaemia, hyperpnoea rhinitis, keratitis, cachexia
		Marked icterus, anorexia, listlessness, cachexia

B. **Chronic cases (7-14 days standing)**

(1) Group: "Posthaemolytic" cases:

Sheep No.	District of origin	Relevant clinical data
F9	Aberdeen	Anaemia, hyperpnoea, cachexia, keratitis, rhinitis, anorexia
F10	Aberdeen	Anaemia, hyperpnoea, mild icterus, cachexia, keratitis, rhinitis, anorexia
F11	Aberdeen	Cachexia, anorexia, gastro-intestinal stasis, hyperpnoea
12226	Fraserburg	Cachexia, anaemia, hyperpnoea, anorexia
12227	Fraserburg	Severe anaemia, hyperpnoea, rhinitis, keratitis, mild icterus, anorexia

(2) Group 2: Mild chronic cases:

NB-2	Calvinia	Weakness, cachexia, anorexia, rhinitis
NB-3	Calvinia	In Extremis, severe anaemia, hyperpnoea, rhinitis, keratitis
NB-4	Calvinia	Weakness, very thin, anorexia
NB-5	Calvinia	Weakness, very thin. Anorexia, gastro-intestinal stasis
F-7	Aberdeen	Anaemia, hyperpnoea, cachexia, anorexia, rhinitis
F-12	Aberdeen	Icterus, anaemia, cachexia, gastro-Intestinal stasis
FB-9	Fraserburg	Anaemia, hyperpnoea, rhinitis, Temp. 105°, icterus
FB-11	Fraserburg	Severe anaemia, hyperpnoea, cachexia
5115	Sutherland	Anaemia, anorexia, gastro-intestinal stasis
5123	Fraserburg	Anaemia, anorexia, gastro-intestinal stasis

(3) Group 3: Severe chronic cases:

FB-3	Fraserburg	In extremis, severe anaemia, hyperpnoea, cachexia, gastro-intestinal stasis, dehydration
FB-7	Hondefontein	Severe anaemia, hyperpnoea, cachexia, in extremis, Temp. 104°
FB-8	Hondefontein	Anaemia, hyperpnoea, listless, anorexia, gastro-intestinal stasis

APPENDIX 2: STUDIES ON THE HAEMATOLOGY OF ENZOOTIC ICTERUS

(Note: RCV = Red cell volume (%); Hb = haemoglobin (g%); RCC = red cell count (10^6/Cu mm);
TPP = total plasma proteins (g%); ESR = erythrocyte sedimentation rate (22/hr);
MCHC = Mean corpuscular haemoglobin concentration (%); MCV = mean corpuscular volume (C/μ);
MCH = Mean corpuscular haemoglobin ($\gamma\gamma$); WCC = White cell count (10^3/Cu mm);
N = neutrophiles (%); L = lymphocytes (%); M = monocytes (%); E = eosinophiles (%);
B = basophiles (%)).

A. Early clinical cases: Group 1—Mild cases when first examined

Sheep No.	RCV	RCC	Hb	TPP	ESR	MCHC	MCV	MCH	WCC	Differential White Cell Count
2206	13.0	1.57	3.4	—	0	26.15	82.80	21.65	25,200	N41, L54, M5, E0, B0
2207	14.5	3.49	5.05	—	0	34.82	41.54	14.46	18,300	N57, L39, M4, E0, B0
2208	14.5	1.80	3.4	—	0	23.44	80.55	18.88	14,100	N52, L42, M6, E0, B0
12223	36.5	8.76	11.1	7.0	0	30.41	41.66	12.67	6,850	N62, L34, M7, E0, B0
12224	33.8	7.05	12.06	7.38	1	35.68	47.94	17.10	8,100	N62, L36, M2, E0, B0
12225	35.3	8.05	11.34	7.63	0	32.12	43.85	14.08	6,000	N51, L35, M7, E7, B0
12228	35.8	11.66	10.38	—	0	28.99	30.70	8.90	6,600	N65, L29, M6, E0, B0
12229	29.0	9.07	9.17	—	0	31.62	31.97	10.11	8,900	N75, L22, M3, E0, B0
Bekker 3	27	—	8.68	7.47	0	32.14	—	—	—	———
FB-2	—	4.66	7.00	—	0	—	—	15.02	12,600	———

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B. *Early clinical cases: Group 2—Severe early cases*

Sheep No.	RCV	RCC	Hb	TPP	ESR	MCHC	MCV	MCH	WCC	Differential White Cell Count
Bekker 1	28	—	10.14	9.62	0	36.21	—	—	—	———
Bekker 2	24	—	7.80	7.42	0	32.50	—	—	—	———
Bekker 4	32	—	10.62	7.56	0	33.18	—	—	—	———
FB-12	—	—	8.00	6.60	1	—	—	—	—	———
F6	35	6.28	6.03	6.75	0	17.22	55.73	10.56	8,500	N72, L13, M9, E6, B0
F8	35	8.59	9.41	7.00	0	26.88	40.74	26.88	7.050	N85, L10, M4, E1, B0

C. *Chronic cases: Group 1—"Posthaemolytic" cases*

Sheep No.	RCV	RCC	Hb	TPP	ESR	MCHC	MCV	MCH	WCC	Differential White Cell Count
F-9	28.0	7.02	7.96	6.32	0	28.42	39.88	11.33	9,200	N69, L18, M10, E3, B0
F-10	32.0	7.49	9.65	7.00	0	30.15	42.72	12.88	2,250	N38, L59, M3, E0, B0
F-11	28.0	8.65	12.55	7.72	0	44.82	32.36	14.50	3,850	N52, L46, M2, E0, B0
12226	11.0	4.90	3.38	—	1	30.72	22.44	6.89	2,900	N55, L43, M2, E0, B0
12227	12.7	1.85	3.86	—	4	30.39	72.57	22.05	5,150	N55, L40, M5, E0, B0

D. *Chronic cases: Group 2—Mild chronic cases*

Sheep No.	RCV	RCC	Hb	TPP	ESR	MCHC	MCV	MCH	WCC	Differential White Cell Count
NB-2	25.0	7.94	7.48	7.00	2	29.92	31.48	9.42	7,100	———
NB-3	19.5	4.03	5.55	7.56	0	28.46	48.38	13.77	8,850	———
NB-4	25.0	8.62	6.76	7.90	0	27.04	29.00	7.84	7,700	———
NB-5	31.0	8.55	8.20	7.72	0	26.45	36.25	9.59	8,650	———
5115	27.0	—	7.35	—	0	27.22	—	—	—	———
5123	28.0	11.37	7.90	—	0	28.21	24.62	6.94	3,800	———
F-7	14.0	5.10	4.34	7.00	0	31.00	27.45	8.50	6,800	———
F-12	37.9	8.33	8.44	6.50	0	22.26	45.49	10.13	7,050	N54, L42, M4, E0, B0
FB-9	—	6.76	9.50	—	0	—	—	14.05	3,800	———
FB-11	—	3.24	6.00	5.20	1	—	—	18.51	5,725	———

E. *Chronic cases: Group 3—Severe chronic cases*

Sheep No.	RCV	RCC	Hb	TPP	ESR	MCHC	MCV	MCH	WCC	Differential White Cell Count
FB-3	25.0	3.79	6.00	—	1	24.00	65.96	15.83	8,300	———
FB-7	—	4.03	6.50	—	—	—	—	16.12	6,250	———
FB-8	—	5.25	8.40	—	—	—	—	16.00	7,450	———

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Jl S. Afr. vet. med. Ass. 39 (2): 33-35 1968

THE WEIGHTS AND MEASUREMENTS OF CERTAIN ENDOCRINE GLANDS OF PREGNANT DORMER AND MERINO EWES

P. C. Belonje and C. H. van Niekerk*

SUMMARY

Dormer and Merino ewes were subjected to various forms of malnutrition during the first sixteen days of pregnancy. This had no significant effect on the weights of the superior parathyroids, hypophyses, thyroids and adrenals. The weights and sizes of these glands were pooled for each breed and means, as well as correlations to bodyweight and to one another, are reported.

INTRODUCTION

During the course of an experiment designed primarily to study the effect of malnutrition and embryonal survival, various endocrine glands were removed and measured. This paper deals with the results obtained from these measurements.

MATERIALS AND METHODS

Dormer and Merino ewes ranging in age from fifteen months to adult were randomly divided into eight groups of equal size, and after mating were subjected to the following regime presented in Table 1 for the first 16 days of pregnancy, at which time they were slaughtered and the necessary specimens removed.

TABLE 1: THE FEEDING REGIMES OF THE GROUPS DURING THE FIRST 16 DAYS OF PREGNANCY

Group	Type of basic ration	Modifications
1	Maintenance	Fed throughout as control
2	,,	Starved on days 8 to 10
3	,,	,, ,, ,, 7 to 11
4	,,	,, ,, ,, 10 to 12
5	,,	,, ,, ,, 9 to 13
6	Submaintenance	Fed throughout
7	Maintenance	Submaintenance on days 7 to 13
8	Submaintenance	Starved on days 9 to 13

In all cases the glands were dissected out, cleared of superfluous tissue and weighed as soon as possible to prevent desiccation.

The measurements of the adrenals, after removal, included total length and midpiece breadth and depth.

The superior parathyroids were consistently found in the most anterior dorsal portion of the cervical thymus and lay immediately ventral to the carotid artery, lateral to the trachea, medial to the jugular vein and posterior to the mandibular salivary gland [1]. The size of the cervical thymus varied considerably.

The hypophysis was reached after transecting the cranium in a frontal plane just posterior to the eyes and removing the brain.

RESULTS

Because of technical difficulties, the four endocrine glands of all the 16 day pregnant ewes could not be removed. Therefore, in order to assess whether there was any significant difference in the gland weights between the breeds and groups, these figures were analysed by a multifactor classification with unequal numbers of observations [2]. Analyses revealed that in all four cases there was neither a significant difference between the groups nor a breed-treatment interaction, but there was a highly significant diference ($p < 0.01$) between the two breeds of animals. For further analyses therefore it was decided to treat each breed separately.

Within each group it was found that some animals had gained while others had lost weight. Standard variance analyses revealed significant difference between the gland weights of these groups within each breed.

As the treatments had had no affect on the gland weights these were pooled for each breed and the mean weight and standard error, correlation of gland weight to body-

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weight, and, in the case of the thyroids and adrenals, the mean sizes, were calculated. A Chi-square random sampling test confirmed that the left adrenal was highly significantly ($p < 0.005$) heavier than the right in both breeds, so the mean weight and standard error of each were calculated.

The results of the analyses for the two breeds are to be found in Table 2 for Dormer and Table 3 for Merinos.

TABLE 2: WEIGHTS, SIZE AND CORRELATIONS OF DORMER EWES
(MEAN BODYWEIGHT 69.89 ±1.38 kg)

Gland	Mean Gland weight and standard error		Correlation of gland to bodyweight		Mean gland measurements* and standard error	
	No. of Animals	Weight	No. of Animals	Significance	No. of Animals	Size in cm
1. Superior Parathyroids	36	47.27 ± 1.83mg	36	Nil	—	—
2. Hypophysis	36	1.16 ± 0.04g	36	Nil	—	—
3. Thyroids	37	3.78 ± 0.22g	37	Nil	18	Right 3.91 ± 0.14 × 1.27 ± 0.06 Left 4.48 ± 0.19 × 1.27 ± 0.07 Isthmus < 0.1—1.5 cm wide
4. Adrenals	37 35	Total 3.90 ± 9.15g (Right 1.86 ± 0.07g (Left 2.04 ± 0.09g	37	Nil	19	Right 2.2 ± 0.06 × 1.3 ± 0.05 × 0.8 ± 0.03 Left 2.6 ± 0.08 × 1.3 ± 0.05 × 0.8 ± 0.04

1. }
2. } Length (SE) × breadth (SE)
3. }
4. Length (SE) × breadth (SE) × depth (SE)

TABLE 3: WEIGHTS, SIZE AND CORRELATIONS OF MERINO EWES
(MEAN BODYWEIGHT 44.53 ±1.13 kg)

Gland	Mean gland weight and standard error		Correlation of gland to bodyweight		Mean gland measurements* and standard error	
	No. of Animals	Weight	No. of Animals	Significance	No. of Animals	Size in cm
1. Superior Parathyroids	38	38.74 ± 1.87mg	38	Nil	—	—
2. Hypophysis	40	0.88 ± 0.04g	40	**	—	—
3. Thyroids	42	2.74 ± 0.13g	42	**	22	Right 3.62 ± 0.20 × 1.23 ± 0.05 Left 3.81 ± 0.16 × 1.24 ± 0.06 Isthmus < 0.1—1.9 cm wide
4. Adrenals	41. 40	Total 3.38 ± 0.14g (Right 1.62 ± 0.07g (Left 1.77 ± 0.08g	41	**	19	Right 2.1 ± 0.05 × 1.2 ± 0.03 × 0.8 ± 0.04 Left 2.4 ± 0.08 × 1.1 ± 0.03 × 0.8 ± 0.03

* See Table 2.
** = Highly significant ($p < 0.01$) positive correlation.

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Further analyses included correlations between the weights of each of the four glands. The results are presented in Table 4.

DISCUSSION

As the various forms of treatment had had no effect, it is asumed that the results obtained represent a reasonable indication of the sizes and weights of these endocrines glands and may be used to supplement the figures currently appearing in two anatomy textbooks [3,4], summaries of which appear in Table 5.

Of interest is the finding that in the Dormer there is no correlation between endocrine weight and bodyweight, whereas in the Merino this is highly significant in the case of the hypophysis, thyroids and adrenals. Although there is no correlation between parathyroid weight and bodyweight, Belonje[1] has indicated that the ratio between the two could possibly be useful in assessing the calcium status of the sheep.

The correlations between the glands are merely reported for the sake of completeness although the significance is not apparent.

ACKNOWLEDGEMENTS

Thanks are due to Mr. D. van Schalkwyk and Mrs. J. Bezuidenhout of the Department of Statistics for their help in the selection of the methods used and to Messrs. G. L. Smit, J. v.d. Westhuyzen and P. Engelbrecht for their assistance.

TABLE 4: THE CORRELATION BETWEEN THE GLAND WEIGHTS OF DORMER AND MERINO EWES

	34 Dormers			34 Merinos		
	Adren.	Thyr.	Hypoph.	Adren.	Thyr.	Hypoph.
Sup. Parathy.	N.S.	*Neg.	N.S.	*Pos.	N.S.	N.S.
Hypophysis	N.S.	*Pos.	—	N.S.	N.S.	—
Thyroids	N.S.	—	—	N.S.	—	—

N.S. = Not significant: * = Significant $p < 0.05$.
Neg. = Negative correlation: Pos. = Positive correlation.

TABLE 5: WEIGHTS AND SIZES OF SHEEP ENDOCRINE GLANDS ACCORDING TO TWO TEXTBOOKS

Gland	May[3]	Sisson[4]
1. Superior Parathyriods	Not mentioned	Not mentioned
2. Hypophysis	2 × 1.2 × 1.6 cm 1.75 g	Not mentioned
3. Thyroids	4—5 × 1—1.5 cm Isthmus 0.25—0.5 × 2 cm	5—6 × 1.5 cm Isthmus 0.6—0.8 cm
4. Adrenals	Right: 2 × 1 cm Left: Longer	Right: ± 3 × 1.5 cm Left: Longer and flatter

REFERENCES

1. Belonje P. C. 1967 *Onderstepoort J. vet. Res.* 34:647
2. Federer W. T. & Zelen M. 1966. *Biometrics* 22:525
3. May N. D. S. 1964 *The anatomy of the sheep* 2nd ed. Melbourne. Wilke and Co. Ltd.
4. Sisson S. 1955 *The anatomy of the domestic animals* — 4th ed. Philadelphia & London. W. E. Saunders Co.

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Jl S. Afr. vet. med. Ass. 39 (2): 37-39 1968

METHYL-THIOURACIL AS FEED ADDITIVE IN A FEEDING TRIAL WITH OXEN

J. F. W. GROSSKOPF AND T. MARKRAM*

SUMMARY

Ten of 20 three year old oxen finished for market were each given an additional supplement of 1.25 g of methyl-thiouracil twice a day for 30 days. The mean increase in live weight of the treated group was 43.6 lb. higher and the dressed carcasses were 18 lb. heavier than the untreated oxen. The carcasses of the treated group were graded one half of a grade higher and had a better fat covering. The mean moisture content of the muscle was 77.4 per cent in the treated and 77.9 per cent in the control group. At open auction the carcasses of the treated group realised mean price of R6.05 more per carcass than those of the control group. From this and other work done in South Africa it is concluded that methyl-thiouracil can be profitably used in the fattening of slaughter catle over three years.

INTRODUCTION

The use of methyl-thiouracil as an additive to cattle fattening rations is at present a controversial matter in the Republic of South Africa. The trials conducted in this country, especially with older animals, proved to be successful and increased the profit margin by approximately R3 to R5 per ox [7, 9, 13, 14]. In younger animals carcass weights were usually not increased by the drug but there was a reasonable saving on feed [7, 14] to obtain the same dressed weight. In repeated feeding trials with a great number of aged beef cows, the addition of methyl-thiouracil caused dramatically improved liveweights gains. Carcasses weighed from 15 to 22 per cent more, less feed was consumed and grading was improved by one to two grades above that of the control cows [16]. Similar results were obtained with old sheep [16].

In the U.S.A., where mainly young steers or lambs are fattened for the market, results with thiouracil and thiourea were inconsistent and did not warrant its general use in feedlots [1, 2, 3, 4]. From Europe on the other hand, favourable results were reported on the use of methyl-thiouracil. Schole [12] found better liveweight gains in oxen and in dairy cows treated with the drug. Feed conversion in the treated groups were also approximately 50 per cent better. In another experiment in Germany [5] the methyl-thiouracil fed group of bulls weighed an average of 18.4 kg more on the hook than the control group. A Dutch experiment showed a difference of 12 kg in favour of the carcasses of treated cows.

In those experiments where moisture determinations on muscle had been done, it was found that the beef fed on methyl-thiouracil contained approximately one per cent more water than that of the control animals [5, 10]. Frens [8] concluded that the additional carcass weights were apparently not due to an increased moisture content of the muscles but that carcass weights were not in relation to the phenomenal live weight gains. Van der Merwe's [16] results are in accord with this.

Residual methyl-thiouracil in the tissues of treated animals seems to be negligible within 24 hours after discontinuation of the drug [6]. Rib-eye muscle, liver, kidney, fat and digestive organs of animals fed 0.5 g of the drug per 100 lb bodyweight per day contained negligible amounts of the drug [15]. In South African experiments the residual levels of the drug in the meat were found to be zero or less than 1 ppm [14]. These traces should not present any hazard to human health [14]. In Germany meat, liver and kidneys from treated animals were found to be completely free from methyl-thiouracil when the animals were slaughtered 40 to 46 hours after discontinuation of the drug [12].

The future fertility of female cattle is apparently not affected by thiouracil. Ten heifers previously treated with thiouracil (2

* Dept. of Animal Science, Faculty of Agriculture, University of Pretoria.

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lb thiouracil per 1000 lb feed) over a period of 201 days all became pregnant. An average of 1.3 services were required for conception. (The amount of methyl-thiouracil given in the present study represented only 0.35 lb per 1000 lb of feed).

MATERIALS AND METHODS

Twenty Afrikander/Hereford/South Devon crossbred oxen varying in age from 30 to 33 months were used for the experiment. All originated from the same farm. Because of severe drought conditions during their yearling stage the animals were not well grown. They were used in a silage feeding experiment which lasted four months and from which they were discharged just prior to this trial. They were divided into two similar groups of ten each on the basis of bodyweight, previous performance and previous experimental treatment. The oxen were kept in groups of five. Separate troughs for concentrates and roughages were provided for each animal.

Each animal in one group of ten received 1.25 g of methyl-thiouracil mixed with their maize meal twice a day. The daily ration fed per head to all the steers consisted of the following:

Yellow maize meal	6 lb.
Fish meal	1 lb.
Chopped teff hay	3 lb. (for first 9 days only)
Chopped lucerne hay	3 lb. (as from 10th day on)
Maize silage	*ad lib*
Bonemeal/Salt lick	*ad lib*

Roughage was given once a day and the concentrates twice a day in two equal quantities. The animals were weighed weekly after being starved for approximately 20 hours. The feeding trial lasted 30 days. On the 31st day the oxen were transported to an abattoir and slaughtered the following morning.

Immediately after slaughter, samples of *M. triceps brachii* were transferred to sealed containers. The samples were weighed and dried to constant weight at 80° C in order to determine their moisture content. Carcasses were graded to the nearest third of a grade and given a fat score by three competent judges. Warm carcass weights were recorded. All carcasses were auctioned on the open market.

RESULTS

Apart from the silage, the animals ate their feed well. It appeared that 6 lb. of maize meal was almost the maximum that they would consume per day. During the first 11 days of the trial the consumption of silage declined steadily. From the 12th day, however, the intake improved when silage from another silo was offered. Because of the enormous fluctuations in the moisture content of the silage, its consumption is also presented on a dry matter basis.

The results are given in the Table.

TABLE: DATA ON METHYL-THIOURACIL ADMINISTRATION TO STEERS ON FINISHING RATIONS

	Methyl-thiouracil group	Control group
Mean initial liveweight, lb.	676.6	674.0
Mean final liveweight, lb.	793.0	749.4
Mean gain, lb.	116.4	75.4
Mean daily gain, lb.	3.88	2.51
Silage consumed per ox in 30 days, lb. (wet)	501	478
Silage consumed per ox (as dry matter), lb.	171	163
Mean feed consumed, lb.*	471	463
Feed conversion ratio	4.05 : 1	6.14 : 1
Warm carcass weight, lb.	423.0	405.0
Dressing percentage	53.4%	54.0%
Carcass grading	3, Prime A	2, Prime A
	1, Prime B+	—
	2, I+	—
	1, I	3, I
	1, I−	3, I−
	1, II−	1, II
	—	1, II
Mean carcass fat score (max. 10)	4.0	3.1
Mean moisture content of Meat	77.42%	77.96%
Mean carcass price realised		
Republic's weighted mean price for 1966/67	R88.26	R82.21
	R78.90	R74.28

CONCLUSIONS

The daily addition of 2.5 g of methyl-thiouracil to the ration of oxen markedly improved live weight gains. The carcasses of the treated group also weighed more, but, as stated by Frens[8] and by Fourie and Louw[7], the increased carcass weights were not quite as great as the liveweight advantage of the treated group. In this trial the treated oxen weighed on the average 43.6 lb. more and dressed 18.0 lb. more than the control oxen. If gain in carcass weight is taken as 60 per cent of liveweight gain then the 18 lb. higher

*Silage calculated on dry matter basis

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dressing weight represents only 30 lb. or 73 per cent of the higher liveweight gain.

Whereas in other known experiments [5, 10], the meat of the methyl-thiouracil treated animals contained approximately 1 per cent more moisture, the beef of the treated group in this experiment had an insignificatly lower level than the controls.

The carcasses of the treated group graded better than those of the control animals and also had a better fat covering. Mathematically estimated, the difference between the two groups was half a grade. In other similar experiments where feed intake was measured, the methyl-thiouracil treated groups consumed less feed and this often represented the greatest profit [5, 7, 13, 14]. In this trial the treated animals consumed 8 lb more silage, on a dry matter basis, over the month, a figure that may be disregarded.

As can be seen from Table 1, the carcasses of the methyl-thiouracil treated oxen realised an average of R6.05 more per carcass than those of the control animals. On the other hand, if valued at the weighted average prices for the various grades on the controlled markets in the Republic during 1966/67, the treated carcasses would be worth R4.62 more per carcass than the untreated ones. It therefore seems that the buyers were prepared to pay a little more than the normal differences between grades for the carcasses of the treated animals in this experiment. The price of the methyl-thiouracil used was R2.55 per animal which, if subtracted, still leaves a handsome profit.

This small contribution adds to the list of publications reporting on the successful use of methyl-thiouracil as a feed additive to cattle. All publications on methyl-thiouracil feeding to cattle over the age of three years that were consulted, reported significantly higher carcass weights and apparent economic advantages in the treated groups. With younger experimental animals, however, inconsistent results were recorded. It can be concluded, therefore that the addition of methyl- thiouracil to suitable fattening rations for cattle over the age of three years can be recommended.

ACKNOWLEDGEMENTS

The financial assistance by the South African Livestock and Meat Industries Control Board, which made this investigation possible, is gratefully acknowledged.

REFERENCES

1. Andrews F. N., Beeson W. M., Barrick E. R. & Harper C 1947 *J. Anim. Sc.* 6 : 3.
2. Barrick E. R., Beeson W. M., Andrews F. N. & Harper C 1949 *J. Anim. Sc.* 8 : 243.
3. Beeson W. M., Andrews F. N. & Brown P. T. 1947 *J. Anim. Sc.* 6 : 16.
4. Blaxter K. L., Reineke E. P., Crampton E. W., & Petersen W. E. 1949 *J. Anim. Sc.* 8 : 307.
5. Drepper K., Zucker H. 1963 *Der Tierzüchter*15 : 389.
6. Ely R. E., Olson K. J. & Reineke E. P. 1948 *J. Anim. Sc.* 7 : 208.
7. Fourie P. C., Louw G. N. 1965 *Proc. S.Afr. Soc. Anim. Prod.* 4 : 111.
8. Frens A. M. 1949 *Landbouwk. Tijdschr.* 12 : 916.
9. Greef W. L. 1967 *Proc. S. Afr. Soc. Anim. Prod.* 6 (In press).
10. Grosskopf J. F. W. 1967 Unpublished data.
11. Kline E. A., Ensminger M. E., Cunha T. J., Heinemann W. W. & Ham W. E. 1949 *J. Anim. Sc.* 8 : 411.
12. Schole 1956 *Tierärztl. Umsch.* 11 : 148.
13. Serfontein W. J. 1967 Personal communication.
14. Terblanche H. J. J. 1967 *Jl S. Afr. vet med. Ass.* 38 : 3.
15. Turner C. W. 1957 *J. Anim. Sc.* 16 : 1064.
16. Van der Merwe P.K. 1967 Personal communication.

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Jl S. Afr. vet. med. Ass. 39 (2): 41-45 1968

AMMELIDE AND AMMELINE AS NON-PROTEIN NITROGEN SUPPLEMENTS FOR SHEEP

H. I. MACKENZIE* AND I. B. J. VAN RENSBURG**

SUMMARY

Toxicity and feeding trials were carried out to assess the value of ammelide and ammeline as sources of NPN for sheep.

Neither ammelide nor ammeline was acutely toxic and ammelide was not cumulatively toxic over a period of 6 weeks.

Of the sheep fed ammeline or mixtures of ammeline and ammelide, half died of crystalluria within 58 days.

It is concluded that neither of these compounds was utilized as a source of nitrogen and that they are not suitable as NPN supplements for sheep.

INTRODUCTION

Two of the cyclic derivatives of urea namely cyanuric acid and melamine differ in their effects when fed as sources of non-protein nitrogen (NPN) to sheep. Cyanuric acid (32% N) is neither acutely nor cumulatively toxic and is effectively used as a supplementary source of nitrogen [1, 2, 3]. In contrast, melamine (66% N) can be utilized to a limited extent as a source of NPN but it is toxic, particularly when fed to sheep which do not have access to unlimited drinking water [4, 5].

Two other cyclic urea derivatives namely ammelide (43% N) and ammeline (53% N) were tested to determine their effects on sheep.

I. TOXICITY TEST

Methods

Eight three-year-old German Merino ewes which had been on a diet of low quality hay *ad lib* and a daily supplement of 100g maize meal and 15g biuret for three weeks were each weighed and drenched once with a suspension of ammelide or ammeline in water. After drenching they were kept under continuous observation for a period of one hour. During the following three days they were observed intermittently and were then returned to the flock. The details of this experiment are given in Table 1.

TABLE 1: DOSAGES AS A SINGLE DRENCH

Sheep No.	Ammelide (g)	Ammeline (g)	Water (ml)	gNPN/kg body-weight
73	24	—	100	0.696
54	48	—	200	1.304
58	72	—	300	1.823
74	88	—	370*	2.768
6	—	20	100	0.647
100	—	40	200	1.036
48	—	60	300	1.887
32	—	74	370*	1.874

* These sheep refused to ingest the whole amount.

Results

The drenches had no obvious adverse effect on the sheep.

II. FEEDING TRIAL

Methods

Fifty, three-year-old German Merino maiden ewes were dosed with an anthelmintic (Thibenzole*) and a vitamin A concentrate before conditioning to pen feeding for 22 days. During the first 14 days a ration of good quality hay (10.7% crude protein) and a lick consisting of equal parts of dicalcium phosphate and salt were fed *ad lib*. For the following eight days poor quality hay was fed.

After the conditioning period the sheep were weighed and divided into five equal groups of similar weight. Treatments were allocated at random to the groups. The basal ration was poor quality veld hay (5.6% CP) and the abovementioned lick *ad lib*. The sheep were fed once daily as groups, with supplements consisting of a mixture of maize meal and a NPN source for a period of 42

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TABLE 2: COMPOSITION OF NPN SOURCES

NPN Source	Total N%	Ammelide %	Ammeline %	Melamine %	Na_2O %	K_2O %	Moisture %
Ammelide	43.2	91.4	3.1	—	0.33	—	0.8
Ammeline	52.6	3.1	89.6	2.2	0.16	0.99	1.1
Mixture I	47.7	43.3	45.1	2.4	0.58	0.58	1.1
Mixture II	46.2	62.7	28.4	3.5	0.40	0.24	1.1

days. The NPN source consisted either of ammelide, ammeline or of mixtures of both these substances in different proportions (designated Mixture 1 and Mixture 2'; the details and composition of which are given in Table 2. Neither the ammeline nor the ammelide used was in pure form.

The composition of the supplements fed to each group is shown in Table 3.

TABLE 3: COMPOSITION AND AMOUNTS OF SUPPLEMENTARY MIXTURES

Group	Source of NPN	Amount of NPN (g per group per day)	Amount of maize meal (g per group per day)
1	Nil	Nil	1000
2	Ammelide	138.9	1000
3	Ammeline	114.1	1000
4	Mixture I	125.8	1000
5	Mixture II	129.9	1000

The NPN sources suplied 60 g of N. per day.

During the test the sheep were weighed at fortnightly intervals after being deprived of feed and water for 16 hours. Group hay intakes were measured daily and lick consumption was calculated weekly.

Four sick sheep were slaughtered and autopsied and organ specimens were taken for histological examination. Before slaughter two of the sheep were bled on several occasions as indicated in Table 5 and the blood samples analysed for their urea and creatinine contents by routine procedures.

Results

After four weeks the sheep receiving supplements containing ammeline began showing symptoms similar to those that were observed in sheep on diets containing melamine[4]. The experiment was continued for a further two weeks and then all the sheep were put on a recovery ration of good quality hay plus a daily supplement of 100g maize meal per sheep.

The result of the first four weeks only are presented since the effects of ammeline showed a marked increase in severity during the fifth week of feeding and the first sheep died on the 39th day of the trial.

During the first 28 days there was little difference between the performance of the five groups. During the fifth week the hay intake of Groups 3, 4 and 5 dropped to an average of 0.32 kg per sheep per day, while the corresponding figure for Groups 1 and 2 was 0.41 kg per sheep per day. On the other hand, the supplementary mixture were completely consumed by the sheep in the affected groups even when they were showing relatively advanced symptoms.

A total of 15 animals in Groups 3, 4 and 5 died between the 39th and the 58th day. No deaths occurred during the four months after the experiment was terminated.

TABLE 4: LIVEWEIGHT AND FEED INTAKE OF SHEEP OVER 28 DAYS

	Group 1 (Control)	Group 2 (Ammelide)	Group 3 (Ammeline)	Group 4 Mixture I	Group 5 Mixture II
Av. initial wt (kg)	38.0	37.4	38.5	38.2	37.7
Av. final wt (kg)	32.3	31.4	30.0	32.3	31.0
Av. wt loss (kg)	–5.7	–6.0	–8.5	–5.9	–6.7
Av. hay consumption (kg per head per day)	0.51	0.56	0.48	0.54	0.48
Av. lick consumption (g per head per day)	5.95	9.64	7.65	7.37	5.67
No. of animals	10	10	10	10	10

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Symptoms

All the sheep in the affected groups showed inappetence, emaciation, excessive urination and a serous discharge from the nostrils. A large number of them also developed a watery green diarrhoea.

Post-mortem investigations

Four sheep showing typical symptoms were examined at the Veterinary Research Institute, Onderstepoort.

The two weakest sheep were killed for detailed postmortem examination. The lesions observed in the two carcasses were indentical and were as follows:

The adipose tissue was in an early stage of serous atrophy, while ruminal atony, acute catarrhal to haemorrhagic enteritis and diarrhoea were also present. The kidneys were markedly enlarged and were estimated to be one and a half times the normal size. Their colour was a light fawn, the surface was irregular and had a mottled appearance. On incision the edges everted and the cut surface appeared very moist. Both the cortex and medulla showed numerous thin radiating yellowish-white, dull streaks which varied in length from 1 to 10 mm (Fig. 1). It was considered that these streaks in the kidneys were caused by crystal formation which somewhat resembled calcium deposits. The urine was strongly positive for sugar. Hydropericardium was present and the pH of the contents of the rumens varied from 7.4 to 7.6.

The other two less severely affected sheep were kept under observation for several days during which blood samples were collected and analysed (See Table 5).

TABLE 5: CHEMICAL-PATHOLOGICAL FINDINGS IN BLOOD SAMPLES

	Day	Creatinine mg %	BUN mg %
Sheep I	1	7.6	92
	5	6.7	156.4
Sheep II	1	3.8	64.4
	5	3.0	83.8
	7	3.25	104.8
	20	2.2	75.44
Normal		1.8–2.7 (2.1)	10–20

The figures given as normal are those generally accepted as such at the Veterinary Research Institute.

The result of these analyses indicated that the kidneys were severely damaged and that due to uraemia the prognosis of these two cases was not favourable.

One sheep (No. I) became recumbent after a week and was slaughtered. Necropsy revealed identical lesions with those observed in the previous two cases.

The other sheep (No. II) lingered for several weeks and during this period it consumed small quantities of feed, but its condition neither improved nor deteriorated to any extent. After six weeks it was electrocuted for post-mortem examination. In this animal only the kidneys showed macroscopic lesions of significance, namely slight enlargement, and although not so severely affected, their appearance resembled those of the previous cases. Neither diarrhoea nor enteritis were present.

Histopathological examination

The major organs of the body were collected for histopathological examination from each of these sheep and, with the exception of the kidneys, did not reveal any significant changes which would indicate primary involvement of the organ concerned. The kidneys, however, were affected by a severe nephrosis in addition to a few rather rare scattered foci of sub-acute lymphocytic interstitial nephritis. The nephrosis was manifested by hydropic degeneration of the tubular epithelium and the presence of casts in many lumens of the tubules. These casts were brownish with a dark blue tint in parts and appeared to be the precursor to the crystals. Tubular dilatation was marked in both the cortex and medulla and the presence of numerous crystals in the lumens of the tubules was very obvious in all parts of the kidney (Fig 1:2). The crystals could easily be seen under the lower magnification of an ordinary light microscope and were luminous under polarised light, being very easy to demonstrate by this method. They were all of a very characteristic structure namely roundish in shape with a laminated appearance and radiating streaks from the centre (Fig. 1:3). The colour in haematoxylin-eosin stained sections was a light to pinkish yellow. In the sections of the pancreas no distinct islets of Langerhans could be detected by the routine examination method.

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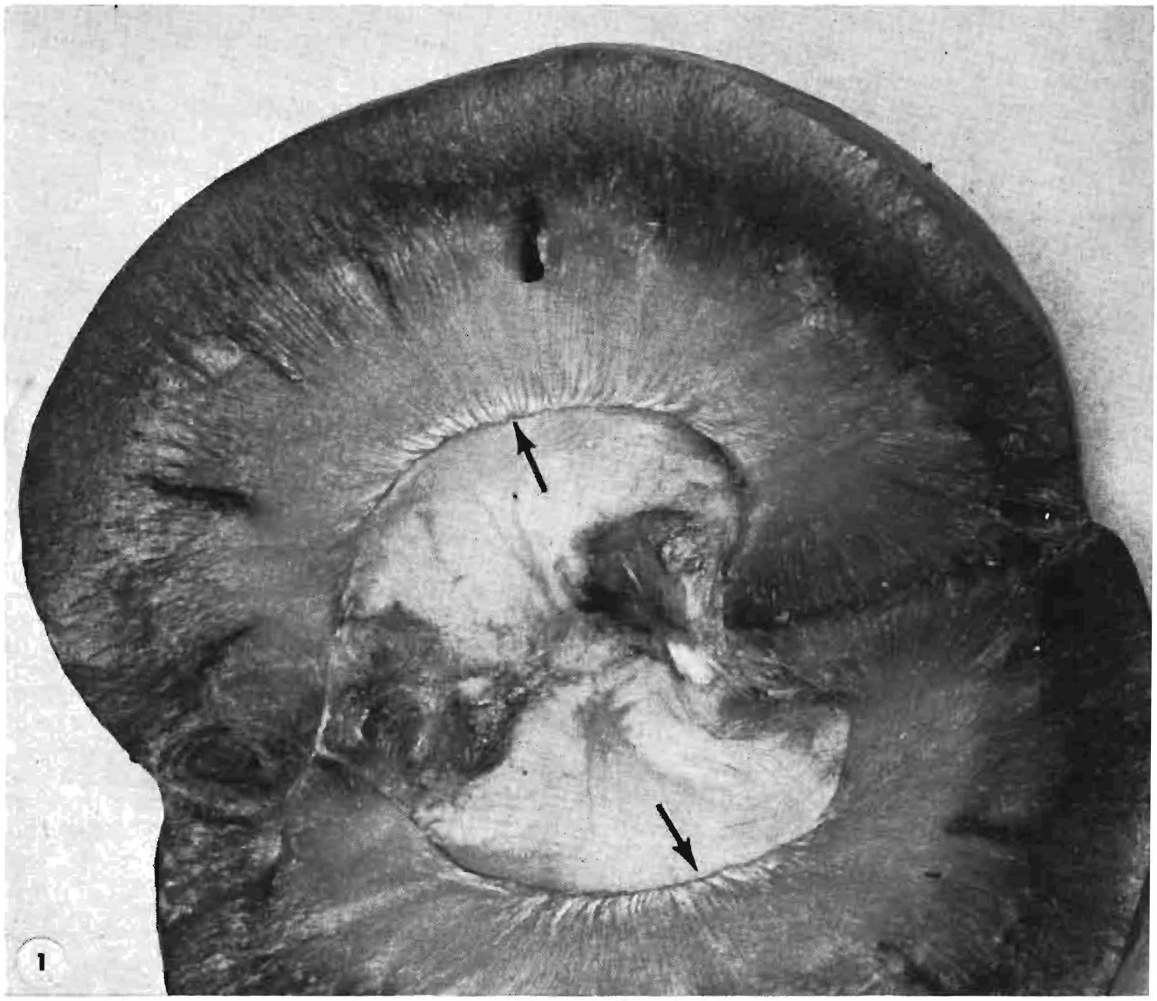

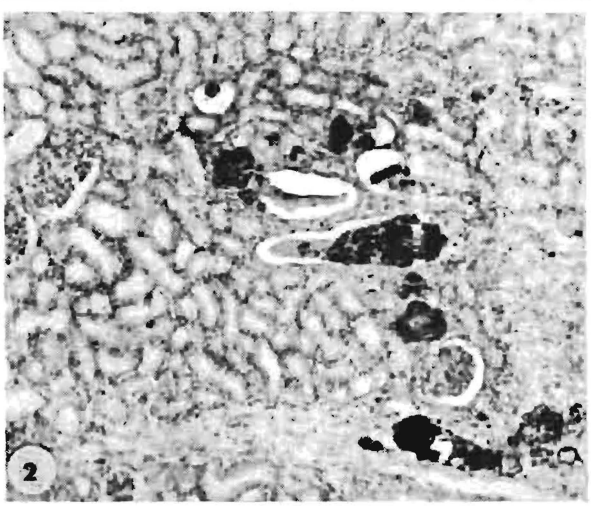

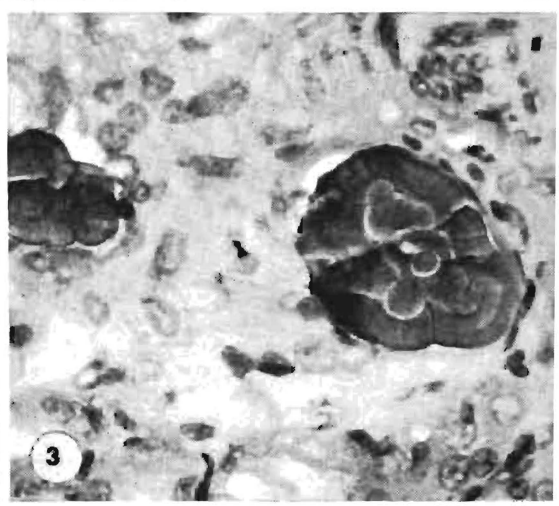

Fig. 1: (1) Macroscopic examination of kidneys.; (2) tubular crystallaria., (3) crystals examined under lov magnification in polarised light.

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DISCUSSION AND CONCLUSIONS

Ammelide was not acutely toxic and did not cause toxicity when fed to sheep for a period of 6 weeks. There was no evidence that this compound was used as a source of nitrogen by the sheep. The weight losses and feed intakes were similar to those of the control group which received no NPN. Ammeline, although not acutely toxic, resulted in the development of a crystalluria syndrome, similar to that observed when melamine was fed[4, 5]. Half of the sheep which were fed ammeline or mixtures of ammeline and ammelide died within 58 days.

It is of interest to note that in Group 5 the average daily intake of 3.7 g ammeline in the mixture was sufficient to cause the death of five of the 10 sheep in the group.

The results indicate that neither ammelide nor ammeline are suitable sources of NPN for sheep.

ACKNOWLEDGEMENTS

The authors wish to thank the Chief, Veterinary Research Institute for permission to publish this article.

Appreciation is expressed to the Section of Photography and the Technical Staff in the Departments of Pathology and Toxicology for their assistance.

REFERENCES

1. Altona R. E. & Mackenzie H. I. 1964 *Jl S. Afr. vet med. Ass.* 35 : 203.
2. Clark R., Barratt E. L. & Kellerman J. H. 1965 *Jl S. Afr. vet med Ass.* 36 : 79.
3. Mackenzie H. I. 1965 *Jl S. Afr. vet. med. Ass.* 36 : 369.
4. Mackenzie H. I. 1966 *Jl S. Afr. vet. med. Ass.* 37 : 153.
5. Clark R. 1966 *Jl S. Afr. vet. med. Ass.* 37 : 349.

BOOK REVIEW

PROTEIN UTILIZATION BY POULTRY

Edited by

R. A. MORTON AND E. C. AMOROSO

Oliver & Boyd, Edinburgh & London, 1967. Pages 215. Price 90s.

This book contains the proceedings and discussions of a twelve-paper second symposium held by the British Egg Marketing Board.

The contributing authors include C. Calet (Jouy-en-Josas, France), J. D. Summers (Guelph, Canada), G. F. Combs (Maryland), U.S.A.), D. C. Snetsinger (Minnesota, U.S.A.) and D. Lewis (Nottingham, Britain). The excellence of these and other authors and contributors has resulted in a publication of particular brilliance and unique value to specialist nutritionists associated not only with poultry but with proteins as a whole.

The book is presented in four overlapping parts commencing with methods used in the evaluation of proteins, their amino-acids and availability. This is followed by papers dealing with the requirements of poultry, tests for quality and the evaluation of cereal proteins. The third part deals with the allowances for chick, hen and turkey requirements and the fourth part deals with the relationships of amino-acids with other nutrients as well as factors affecting protein requirements.

Each paper is of exceptional quality deserving careful assimilation and the discussions at the end of each of the four parts warrant special commendation.

The book is presented on good paper, is attractively bound and the clear condensed graphs and tables facilitate the assimilation of the material presented. One can consider it a privilege to have a book of such a high calibre on one's bookshelf.

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Jl S. Afr. vet. med. Ass. 39 (2): 47-55 1968

PARASITISM ON PASTURES *

I. G. HORAK AND A. J. SNIJDERS**

SUMMARY

The life cycles, ecology of the free-living stages and epizootiology of helminths and coccidia are reviewed. The pathogenesis and pathology are discussed and methods of control based on pasture conditions suggested. The findings in several South African trials are presented and discussed.

INTRODUCTION

Parasitism in domestic animals on permanent improved pastures has not been studied in detail in South Africa. Surveys of the seasonal fluctuations of helminth populations have been confined mainly to sleep under veld grazing conditions [1-4]. At the Outeniqua Experimental Station at George in the Cape Province, however, a seasonal incidence survey, helminth control programme and production trial have been conducted in sheep grazing on permanent, improved pastures [5, 6].

Relevant data acquired at Outeniqua will be discussed, but a considerable portion of this paper will be devoted to overseas results and laboratory findings in South Africa. The discussion on coccidiosis will be confined to results obtained under conditions of intensification in South Africa. No pasture findings are available.

LIFE CYCLES

The life cycles of the common nematodes, trematodes and cestodes have been described by Soulsby [7] and those of the various coccidial species by Pellérdy [8]. Under conditions of intensification the free-living stages of these parasites play an important rôle in the epizootiology of disease and greater attention will be given to them.

ECOLOGY OF THE FREE-LIVING STAGES

(i) *Nematodes*

Eggs are passed out with the faeces of the host animal. The eggs require moisture, oxygen and heat for development and hatching and considerable numbers of eggs fail to hatch. After hatching, the larvae undergo two moults before reaching the third or infective stage; large numbers of larvae die before reaching this stage. The infective larvae are negatively geotropic, positively phototropic to mild light, but strong light repels them, active in warm conditions; moisture is essential for movement. Too much moisture, however, retards vertical migration [9].

Kauzal [10] found that 16 per cent of larvae migrated on to the surrounding grass blades, but Silangwa and Todd [11] demonstrated that only two to three per cent of larvae migrated up grass blades and of these only 59 per cent reached the first inch above the soil. It would thus seem fortuitous that animals should become heavily infested with nematodes if one considers the enormous losses occurring before the larvae migrate on to the grass blades. If, however, the almost incredible egg output of some worms is taken into account, this loss would seem necessary to prevent the extermination of the host animals.

Using *Haemonchus contortus* as an example, a single sheep infested with 2,000 adult worms can excrete 10-million eggs daily. If only 10 per cent of these reach the infective stage and only 10 per cent of the infective larvae migrate on to the grazing, a theoretical total of 200 larvae per square yard of grazing is available daily if ten infested sheep graze a paddock one acre in extent. In South Africa 10,000 to 20,000 *H. contortus* larvae are capable of killing a laboratory housed sheep.

The work of Crofton [12] and Gibson [13] indicates that the micro-climate on a pasture is more important than the macro-climate. The "mat" of the pasture is most important in supplying the correct micro-climate — it prevents desiccation and drastic alterations in temperature. After ploughing and reseeding an old pasture there was no or very little infestation left [12] and the absence of "mat"

* Paper presented to the 62nd Annual Congress of the SAVMA Durban, October 1967.
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in a new pasture would also reduce larval survival.

Larvae of the genera *Ostertagia* and *Trichostrongylus* are better able to migrate on to grass under wet conditions than those of *Haemonchus* which, however, are more active at higher temperatures than the previous two genera [9]. The free-living stages of the genus *Nematodirus* differ from the other common nematodes in that development to the infective stage occurs within the egg. The development within the egg and the hatching of the infective larvae of the various species of this genus are governed by temperature, light and moisture, delay in hatching being common in some species. It has been suggested by Thomas and Stevens [14] in Britain that *Nematodirus spathiger* develops and hatches normally without delay, that some delay occurs with *N. filicollis* and hatching is erratic in late winter and spring, while the hatching of *N. battus* is delayed until spring.

(ii) *Trematodes*

The climate in South Africa is generally not suitable for the survival on pasture of the intermediate snail hosts of the common trematodes. *Lymnaea truncatula*, the intermediate host of *Fasciola hepatica* is semi-aquatic and thus is able to survive in shallow permanent patches of water, hence *F. hepatica* is the most common trematode encountered in sheep and cattle. Animals on pasture adjoining permanent natural water sources may be exposed to severe trematode infestation.

The metacercariae of liver fluke and conical fluke are capable of surviving for a month or longer under favourable conditions and thus may be a potential danger, even though snails have been eradicated. The cercariae of *Schistosoma mattheei* die rapidly, but animals may be infested by being turned out to a pasture soon after it has been irrigated from an infested water source.

(iii) *Cestodes*

Pastures form an ideal habitat for the oribatid mites which are the intermediate hosts of the common cestodes of herbivores. Rayski [15] found few mites on newly seeded pastures, but as the amount of humus in the soil increased with ageing of the pastures, the numbers of mites increased. The majority of mites are found on grass during darkness, but during the day their presence on the herbage is dependant upon the amount of moisture present. Seasonal fluctuations in the number of mites are small and *Moniezia* cysticercoids occur in mites throughout the year, thus infestation of the definitive host can occur at any time [15].

(iv) *Coccidia*

The non-parasitic stage exists in two forms as the unsporulated and the sporulated oocyst. Sporulation does not occur until the cocysts reach an environment of suitable temperature, humidity and aeration. They are easily killed by excessive heat or desication, unsporulated oocysts being more susceptible than sporulated oocysts [16]. Direct sunlight is also lethal to the oocyst.

Although work on *Eimeria* spp of sheep and of pigs has indicated that the unsporulated oocyst can survive for long periods [17, 18] other observations seem to indicate that unless sporulation occurs soon after excretion by the host, the oocysts lose their viability [19, 20]. Under ideal conditions of shade and humidity, the oocyst can survive for many months in nature, but the vast majority of oocysts would not survive for this time. The enormous numbers of oocysts excreted by infested animals compensate for the losses that occur.. Oocyst counts in excess of 1-million oocysts per gram of faeces are not uncommon in sheep and goats, giving a total daily output of 500 million oocysts by a single animal.

EPIZOOTIOLOGY

The epizootiology of helminthiasis and coccidiosis under pasture conditions is generally similar in that every animal in the flock or herd is infested at some time or other and contamination of the pasture is continuous.

Soulsby [7] states, "three types of epidemiological situations can be defined in gastrointestinal parasitism of sheep. These are, first, the susceptibility of an already infected community is increased; secondly, susceptible individuals are introduced into an already infected community; and thirdly, the infecting mass of the organisms is increased in an already infected community".

The susceptibility of an already infested community can be increased by conditions of stress, of which insufficient feed is the most important. On pasture insufficient feed should not play a rôle, but the milk yield of individual ewes may be insufficient for their lambs and these lambs will be more susceptible to infestation [21]. Pre- and post-parturient stress in ewes may also play a rôle in making these animals more susceptible to infestation. Severe infestation in the ewe will reduce

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milk yield [22] which in turn will increase the susceptibility of her lamb.

The introduction of susceptible individuals into an already infested community occurs whenever animals are born. In addition, the mothers of these young animals frequently exhibit a post-parturient rise in faecal worm egg counts, thus increasing the infestation available on the pasture.

An increase in the mass of infesting organism is the most frequently occurring epidemiological situation. It occurs when too many animals are kept on too few acres or when animals are kept in a certain restricted area for too long. It also occurs under favourable climatic conditions.

Crofton [23] has shown that flock behaviour results in non-random distribution of faeces over the pasture, thus resulting in areas of high pasture contamination. He observed that the flock avoided grazing these heavily contaminated areas for five to eight days and the regrazing of these areas would thus coincide with the maximum numbers of infective larvae being available under suitable conditions.

PATHOGENESIS AND PATHOLOGY

Soulsby [7] and Muller [24] have published reviews of the effects of parasites on their hosts and this aspects will be summarised briefly.

In the grazing animal parasitic infestations rarely consist of single species and the contribution of individual species to the disease complex cannot be ascertained with certainty. It is thus necessary to rely on laboratory findings where pure infestations have been studied to obtain a picture of the disease caused by the various parasites.

(i) *Haemonchosis*

The fourth stage and adult worms are avid blood suckers [25, 26]. In addition, a severe hypo-albuminaemia develops [27] and submandibular oedema is a fairly constant symptom.

(ii) *Ostertagiasis*

This disease has been studied extensively in cattle and sheep [28-32]. The changes of importance when applied to pastured animals are the anorexia, decrease in apparent nitrogen absorption and low plasma inorganic phosphate, particularly if the pasture is already low in phosphates.

(iii) *Nematodiriasis*

This is a disease primarily of young animals, particularly lambs. Very large numbers of larvae are necessary to produce symptoms which are diarrhoea, dehydration and retarded weight gain [33].

(iv) *Trichostrongyliasis*

Worms of the genus *Trichostrongylus* are the most ubiquitous nematode parasites of sheep throughout the world and many workers have studied the disease produced by them [34-37]. There is apparently an interference with nitrogen, phosphate and selenium absorption and the animals go into a negative nitrogen and phosphate balance, have low plasma inorganic phosphate levels and develop lesions of white muscle disease.

(v) *Oesophagostomiasis*

The disease has been studied in sheep, cattle and pigs [38-40]. The most important changes are anorexia, diarrhoea, loss in bodyweight, hypo-albuminaemia and anaemia. On pasture, the failure to gain weight is of great importance. This decreased weight gain may persist for many months after initial infestation.

(vi) *Fascioliasis*

This disease is characterised by anorexia, hypo-albuminaemia, a decrease in serum magnesium and calcium, anaemia [41], oedemas and icterus.

The hypo-magnesaemia and hypocalcaemia could theoretically play a rôle in the occurrence of grass tetany in lactating ewes, especially where the pasture is already low in these elements.

(vii) *Cestode infestation*

Usually only young animals are affected, and unless infestations are particularly severe when weight gain might be retarded, no apparent effects can be determined [42].

(viii) *Coccidiosis*

The endogenous stages of the life cycle in the epithelium of the small and large intestine and particularly the gametocytes and gametes are responsible for the lesions and symptoms of acute coccidiosis [8, 43]. The parasites invading and leaving the intestinal cells presumably cause discomfort which leads to anorexia and decreased bodyweight. The severe intestinal damage causes diarrhoea

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and haemorrhage, the haemorrhage being responsible for the anaemia which develops. (ix) *Parasitism* of the pregnant ewe might adversely affect the birthweight of the lamb [44]. Lambs suckling from infested ewes may not get sufficient milk as Gordon [22] has shown that a ewe infested with *H. contortus* had a marked decrease in milk yield. It has also been suggested that an imporant predisposing factor to outbreaks of parasitism in lambs is a deficient milk supply [21]. In addition, such lambs will have to supplement their diet by grazing, thus acquiring still heavier infestation.

The adverse effects of parasitism on bodyweights and fleeceweights have been demonstrated by numerous authors [6, 44-46]. Baker and Douglas [47] found greater worm burdens in lambs on irrigated pasture, fed sodium molybdate at a level equivalent to 45.5 mg elemental molybdenum daily than in lambs not fed this substance. Bremer [48] found that *Bunostomum phlebotomum* infestation produced a marked drop in plasma copper in dairy calves. Davis, Herlich and Bowman [49, 50] have shown that combined infestations of coccidia and *Cooperia punctata* or *Trichostrongylus colubriformis* in calves are more severe than either a pure coccidial or pure nematode infestation.

CONTROL

(i) *Management*

The control of infestation by management practices strives to present the grazing animal with uncontaminated pasture, but as this would require unlimited pastures, rotational grazing has to be practiced.

Levine [51] in the United States found that with a two-day grazing period and 48 days of rest, lambs picked up heavy infestations from pastures. In Australia a rotational system of one week's grazing and three week's pasture rest did not increase production or provide better parasite control than continuous grazing on native pasture [52]. Gibson [13] found that the number of larvae per kilogram of herbage was slightly less in a set stocked paddock when compared to the number of larvae in a similar paddock divided into six camps rotationally grazed at weekly intervals. *T. colubriformis* larvae persisted on herbage in plots at a high level for 20 weeks. Donald [53] found virtually no decline in the number of larvae per square yard of pasture eight to nine weeks after the removal of infested sheep and Rose [54] successfully infested lambs with *H. contortus* larvae which had been on herbage for 8½ months.
Coccidia on pasture can survive for at least as long as infective larvae [8, 43].

Thus it is apparent that if rotational grazing alone was to be a successful method of combating infestation the periods between grazing would be too long to make economical use of the pasture.

(ii) *Life cycle*

At least two nematodes can be controlled by management utilising a knowledge of their life cycles.

In Britain lambs become infested with *N. battus* soon after birth when grazing with their dams on spring pasture. They excrete eggs for a few weeks and then become immune; these eggs do not hatch until the following spring when the next season's lambs become infested. By not allowing lambs to graze a pasture grazed by the previous season's lambs, infestations with *N. battus* can be virtually eliminated as the infestation can only be propagated by young lambs[55], previously infested sheep being completely immune.

In the United States *Stephanurus dentatus*, the kidney worm of pigs, can be controlled by virtue of its long prepatent period of aproximately six months. In endemic areas gilts are bred and raise one litter before being slaughtered. This means that had they themselves become infested as piglets, the worms would not yet be patent by the time they were slaughtered, thus infestation is eradicated [56].

(iii) *Chemo-therapy*

Because pastures provide the ideal environment for the survival of nematodes, the value of strategic drenching cannot be overstressed. Strategic drenching is based not only on the seasonal incidence of the parasites but on the prevailing animal husbandry practices.

Strategic drenching based on seasonal incidence has been suggested by numerous workers in South Africa [1-4] and this discussion will be confined to that based on management practices.

The ideal to be aimed at is keeping ewes worm-free from flushing until they wean their lambs, and the lambs worm-free from birth until marketing as fat lambs or six months of age. This can be achieved by drenching the ewes just prior to flushing [57], at

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which time they are placed on a fresh pasture, again eight to six weeks before lambing, to ensure maximum foetal growth, immediately prior to or subsequent to lambing when again they must be placed on a rested or new pasture, and thereafter at monthly intervals until their lambs are weaned. The lambs must be treated at monthly intervals until six months of age. These relatively short interval drenches apply particularly to animals on irrigated pastures — those on pastures dependent upon rainfail can be drenched at six-weekly intervals and even longer during periods of drought.

In Britain Gibson and Everett [58] treated ewes at mating in autumn. Conditions must have been unfavourable for re-infestation in the *interim* since these ewes had nematode egg counts of only 16 eggs per gram when they were turned out with their lambs. This low level of infestation in the ewes prevented infestation of the lambs which had egg counts of only 31 eggs per gram when they were marketed without any further treatment at 3½ to four months of age weighing 65 to 79 lb.

When shifting sheep from one pasture to the next they must be drenched, providing the grazing intervals on each pasture are not so short as to make drenching uneconomical. If sheep are drenched and allowed to remain on the same pasture, particularly if it is heavily infested, no long-term advantage from drenching may be detected.

A useful method of keeping young stock worm-free, particularly fat lambs, is by incorporating an anthelmintic in a lick thus ensuring a daily low level intake [6, 44, 59].

In coccidiosis, therapeutic treatment of the acute disease will prevent mortalities but may supress oocyst output for a short while only. As in nematode infestations it is the adult animal which provides the source of infestation for the young susceptible animal and low level treatment of the pregnant and in-milk ewe and her lamb can be of material benefit in those areas where coccidiosis is a problem.

(iv) *Immunisation*

With the exception of lungworm vaccination in cattle and sheep, no successful practical helminth vaccines for ruminants have been produced [60]. Young sheep until the age of six to nine months cannot be uniformly immunised against helminths. In fact, severe infestation during this period of their lives may prevent them from being able to develop immunity to infestation when they are adults [61].

The immunity which develops in coccidiosis is specific and apparently animals fail to become immune to some species [8], even though they may be immune to one species of *Eimeria*, another species may become dominant and produce disease.

Immunity to helminths and perhaps coccidia can be broken down by conditions of stress and here one thinks particularly of parturition, the post-parturient rise in nematode eggs being well-known in ewes. The authors have also observed acute coccidiosis with high faecal oocyst counts and mortality in ewes shortly after parturition while non-pregnant ewes from the same flock had negative or very low faecal oocyst counts.

Generally, however, older animals have acquired a degree of resistance due to previous infestations with helminths and coccidia and it is possible to make use of this resistance in grazing programmes by allowing the older animals to follow the young susceptible animals on pastures.

SOUTH AFRICAN FINDINGS

(i) *Helminths*

(a) On irrigated pasture in the Grabouw district of the Cape Province, 50 lambs were drenched with anthelmintics at monthly intervals from birth in August, to slaughter, while a further 25 were maintained as untreated controls. The nematode egg counts in the treated group were virtually negative throughout the trial while the average counts in the controls never exceeded 290 eggs per gram of faeces. At slaughter the average total worm burden in five of the controls was 4,756 worms consisting mainly of *Nematodirus* and *Ostertagia* spp., and yet this low burden was responsible for an average of 0.1 lb less wool at six months of age and an average bodyweight 3.7 lb lighter than that of the treated group at 6½ months of age.

(b) Cattle on irrigated pasture adjoining a river in the Johannesburg district were found to be infested with *paramphistomum* spp., *F. hepatica* and *Schistosoma* spp. Other cattle born and reared on the same farm were infested with *O. ostertagi* a parasite normally found only in certain regions of Natal and the Cape Province ,thus illustrating how con-

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ditions on a pasture can differ from those of the surroundings.

(c) An interesting finding was made recently in a random survey of 20 pigs of unknown origin at the Cape Town abattoir. Twelve of these pigs were infested with *Ascarops* spp. — one of them severely. The worms seemed to be associated with ulcerations, haemorrhages and scar tissue formation in the stomachs of the more heavily infested pigs.

(d) At a country club in the Pretoria district horses had been put out in the same paddock for many years and had become heavily infested with nematodes. At the time a single therapeutic dose of an anthelmintic was administered in March, the average faecal egg count was 833 eggs per gram of faeces. Faecal worm egg counts were virtually negative for the following two months and it was five months after treatment before egg counts reached an average of 280 eggs per gram of faeces. From these results it is obvious that the drier, colder winter climate interfered with the development of infective larvae in the paddock and thus reinfestation was gradual. A dosing programme for horses could thus be based on treatments in autumn, spring and mid-summer.

(ii) *Coccidia*

The following results were all obtained under conditions of intensification but not on pastures.

(a) In the Johannesburg district young sheep kept in a battery for fattening purposes suffered from severe acute coccidiosis. Average oocyst counts before treatment were approximately 40,000 per gram of faeces with individual counts as high as 172,000. Specific treatment rapidly reduced the oocyst count to zero and the treated sheep showed an average weight gain advantage of 1.2 lb over a group of untreated controls during a 36-day period.

(b) Young Angora goats were purchased in the Willowmore district of the Cape Province and transported to the Hennops River district in Transvaal. Despite their being housed on concrete, which was thoroughly swept daily, they exhibited rising faecal oocyst counts for the following two months. Specific treatment was instituted, which, although it usually only depressed oocyst counts for a limited period of time, always resulted in increased bodyweight gains in treated goats when compared with untreated controls. The unreliability of oocyst counts as a method of estimating severity of infestation was well illustrated in these trials. Two goats died with oocyst counts below 12,000 oocysts per gram of faeces, while other goats were apparently quite healthy with no signs of diarrhoea yet oocyst counts were in excess of 0.5 million per gram of faeces.

(c) In the Heidelberg district of the Transvaal young calves exhibited symptoms of acute coccidiosis, profuse, bloody diarrhoea, listlessness, emaciation and death. Average oocyst counts never exceeded 10,300 oocysts per gram of faeces, with the highest individual count being 51,600 oocysts per gram of faeces. Specific therapy rapidly reduced faecal oocyst counts, while a gradual decrease in oocyst counts was noted in a group of untreated calves 17 days after the commencement of treatment in the treated group. Daily low level administration of a coccidiostat would seem to be a practical means of control in calves, as severe infestation is usually confined to very young calves and then only for relatively short periods.

OUTENIQUA TRIALS

Work done at the Outeniqua Experimental Farm indicates that daily provision of a mineral lick containing one to two per cent thiabendazole after a therapeutic dose of the same drug gave almost complete nematode control[6]. The experiments were conducted with six month old German Merino and Dormer sheep, Merino sheep and Merino lambs. The criteria of efficacy were based on production measured by weight at birth and weaning final weight and wool production.

Experiment 1

Snijders *et al*[6] used six month old German Merino and Dormer ewes and wether weaners, grazing together during the day from 14 November, 1961 to 21 November, 1962, and compared daily administration of phenothiazine or thiabendazole in a lick with thiabendazole treatment at intervals of approximately six weeks. The estimated daily intake of thiabendazole decreased from 5 mg/Kg liveweight to approximately 2.2 mg/kg and that of phenothiazine from 30 mg/kg to 14 mg/kg liveweight.

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The production results are summarized in Table 1.

TABLE 1: EXPERIMENT 1. BODYWEIGHTS AND WOOLWEIGHTS OF SIX MONTH OLD SHEEP

Group	Average liveweight in lb			Woolweight in lb
	14/11/61	21/11/62	Gain	
Thiabendazole 1.25% in lick	61.9	143.3	81.4	8.0
Thiabendazole six-weekly	62.2	138.2	76.0	7.7
Phenothiazine 10% in lick	62.3	128.0	65.7	7.0

The stocking density during this and successive trials was up to six sheep per acre.

Subsequently, Stapelberg, Snijders and Muller[44] conducted other trials using Merinos of various ages.

Experiment 2

Four groups each consisting of 15 six-tooth Merino wethers ran together during the day from 15 October 1963 to 4 November 1964. Two of the groups were dosed six-weekly, one with thiabendazole and the other with phenothiazine and the other two groups received the same anthelmintics in lick form.

The production results are summarised in Table 2.

Compared to Experiment 1 it is obvious that older sheep show less response in terms of liveweight, but the effect on woolweight is apparent.

Experiment 3

This experiment ran from May to February 1966. Two groups of ewes, each consisting of 40 Merinos and five German Merinos, were used to produce lambs for the actual trial.

One group of ewes received thiabendazole medicated lick prior to lambing and their lambs were dosed with medicated lick from four to 42 days of age and were thereafter allowed access to the medicated lick. The ewes and lambs of the other group received no treatment.

The production results are summarised in Table 3.

TABLE 3: EXPERIMENT 3. BIRTHWEIGHTS AND BODYWEIGHTS OF LAMBS

Treatment	Birthweight in lb	Average liveweight in lb	
		23/8/1965 (weaning)	15/2/1966
Thiabendazole 2% in lick	8.8 (32)	58.7 (31)	98.0 (21)*
Controls	7.8 (35)	49.3 (32)	70.1 (21)

*Excludes animals slaughtered for worm counts.
Figures in brackets indicate number of sheep.

The lambs in the treated group had markedly higher birthweaning- and final weights than the lambs in the untreated group.

Experiment 4

The 80 Merino ewes from Experiment 3 were used and the experiment ran from April 1966 to March 1967.

The ewes had access to thiabendazole medicated lick from the time the rams were introduced until the first lamb was born,

TABLE 2: EXPERIMENT 2. BODYWEIGHTS AND WOOLWEIGHTS OF ADULT SHEEP

Group	Treatment	Average liveweight in lb			Woolweight in lb	
		15 Oct. 1963	3 Nov. 1964	Gain	15 Oct. 1963	3 Nov. 1964
1	Phenothiazine therapeutically 6-weekly	96.2	103.9	7.7	14.2	14.6
2	Phenothiazine 10% in lick	95.3	101.6	6.3	14.6	14.6
3	Thiabendazole therapeutically 6-weekly	94.8	102.6	7.8	14.0	15.3
4	Thiabendazole 1.25% in lick	95.4	105.7	10.3	14.2	15.9

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when the ewes and their lambs were separated into treated and untreated control groups.

The treated lambs were divided into two groups, the one dosed daily with 5gm of two per cent thiabendazole medicated lick and the other dosed daily with thiabendazole at 8mg/kg liveweight until six to eight weeks of age. Medicated lick was available during this period and thereafter. Half of the control lambs were dosed daily with 5mg of basic lick while the other half received 2ml of distilled water daily until they were six to eight weeks of age.

The production results are summarised in Table 4.

As in Experiment 3 the bodyweight differences became more apparent after weaning, when the weaners had to increase their consumption of herbage and thus exposure to parasites increased.

The preceeding results clearly demonstrate the adverse effect that some nematodes have on the productive abilities of sheep under intensive conditions.

Two objections may be mooted against the use of low level treatment with an anthelmintic which eliminates infestation:

(a) the animal will not develop an immunity to infestation when low level treatment ceases;

(b) the parasites will develop a drug tolerance or even resistance.

Considering the first objection it should be mentioned that low level treatment should primarily be used for fat lamb production, where no immunity and a quick return are required, or for lamb rearing during the period when the lambs are immunologically immature, i.e. up to six months of age. Theoretically these lambs should then be able to develop a better immunity than those exposed to severe infestation during this period.

The second objection can be countered by the fact that low level treatment with thiabendazole has been used at Outeniqua for six years without any indication of a tolerance or resistance developing.

TABLE 4: EXPERIMENT 4. BIRTHWEIGHTS AND BODYWEIGHTS OF LAMBS

Group	Treatment	Birthweight in lb	Average liveweight in lb	
			30/8/1966 (weaning)	22/11/1966
T1	5 gms lick 2% Thiabendazole dosed	7.5 (20)	55.6 (19)	79.6 (15)
T2	Thiabendazole 8 mg/Kg	7.5 (20)	59.6 (16)	81.1 (14)
Group Average		7.5	57.4	80.3
C1	5 gms basic lick dosed	7.5 (19)	54.1 (18)	70.9 (14)
C2	2 ml distilled water	7.5 (20)	51.0 (19)	64.6 (17)
Group Average		7.5	52.5	67.4

Figures in brackets indicate number of sheep.

REFERENCES

1. Barrow B. D. 1964 *Onderstepoort J. vet. Res.* 31 : 151
2. Rossiter L. W. 1964 *Onderstepoort J. vet. Res.* 31 : 143
3. Viljoen J. H. 1964 *Onderstepoort J. vet. Res.* 31 : 133
4. Reinecke R. K. *Jl S. Afr. vet. med. Ass.* 35 : 603
5. Muller G. L. 1967 *Onderstepoort J. vet. Res.* In press.
6. Snijders A. J., Stapelberg, J. H. & Muller G. L. 1964 *Jl S. Afr. vet. med. Ass.* 35 : 83
7. Soulsby E. J. L. 1965 *Textbook of Veterinary Clinical Parasitology. Volume 1. Helminths.* Oxford. Blackwell Scientific Publications.
8. Pellérdy L. P. 1965 *Coccidia and Coccidiosis.* Budapest Akadémiai Kiadó.
9. Rogers W. P. 1950 *Parasitology.* 32 : 208
10. Kauzal G. P. 1941 *Aust. vet. J.* 17 : 181

Reproduced by Sabinet Gateway under licence granted by the Publisher (dated 2011)

11. Silangwa S. M. & Todd A. C 1964 *J. Parasit.* 50 : 278
12. Crofton H. D. 1952 *Parasitology.* 42 : 77
13. Gibson T. E. 1966 *Biology of Parasites.* New York. Academic Press Inc.
14. Thomas R. J. & Stevens A. J. 1960 *Parasitology* 50 : 31.
15. Rayski C. 1949 *Proc XIVth Int. vet Congr. London.* 2 : 51
16. McCullough S. F. 1952 *Cit.* Davies S. F. M., Joyner L.P. & Kendall S. B. 1963
17. Christensen J. F. 1939 *J. agric Res.* 59 : 527
18. Avery J. L. 1942 *J. Parasit* 28 (Suppl.) : 27
19. De Vos A. 1967 Personal communication.
20. Horak I. G. 1967 Unpublished data.
21. Whitlock J. H. 1951 *Cornell Vet.* 41 : 299
22. Gordon H. McL. 1950 *Aust. vet J.* 26 : 14, 46, 93
23. Crofton H. D. 1958 *Parasitology* 48 : 251.
24. Muller G. L. 1964 *JlS. Afr. vet med. Ass.* 35 : 535
25. Boughton I. B. & Hardy W. T. 1935 *cit.* Soulsby . J. L. 1965.
26. Andrews J. S. 1942 *J. agric Res.* 65 : 1
27. Malan J. R. 1963 Personal communication.
28. Ross J. G. & Todd J. R. 1965 *Brit. vet. J.* 121 : 55
29. Jennings F. W., Armour J., Lawson R. D. & Roberts R. 1966 *Am. J. vet. Res.* 27 : 1249
30. Ritchie J. D. S., Anderson N., Armour J., Jarrett W. F. H., Jennings F. W. & Urquhart G. M. 1966 *Am. J. vet. Res.* 27 : 659
31. Horak I. G. & Clark R. 1964 *Onderstepoort J. vet. Res.* 31 : 163
32. Horak I. G., Clark R. & Botha J. C. 1965 *Onderstepoort J. vet. Res.* 32 : 147
33. Kates K. C. & Turner J. H. 1953 *Proc Helm. Soc. Wash.* 20 : 117
34. Franklin M. C., Gordon H. McL & MacGregor Catherine H. 1946 *J. Coun. Sci. Ind. Res. (Aust.)* 19 : 46
35. Fitzsimmons W. M. 1966 *Res. vet. Sci.* 7 : 101
36. Gallagher C. H. 1963 *Aust. J. agric Res.* 14 : 349
37. Horak I. G., Clark R. & Grey R. S. 1967 *Onderstepoort J. vet. Res.* In Press.
38. Horak I. G. & Clark R. 1966 *Onderstepoort J. vet. Res.* 33 : 139
39. Bremner K. C. 1961 *Aust. J. agric. Res. 12 :* 498
40. Taffs L. F. 1967 Personal communication.
41. Sinclair K. B. 1962 *Brit. vet. J.* 118 : 37
42. Kates K. C. & Goldberg A. 1951 *Proc. Helm. Soc. Wash.* 18 : 87
43. Davis S. F. M., Joyner L. P. & Kendall S. B. 1963 *Coccidiosis.* Edinburgh. Olivier and Boyd Ltd.
44. Stapelberg J. H., Snijders A. J. & Muller G. L. 1967 *In press.*
45. Thomas R. J. & Bainbridge M. H. 1967 *Vet. Rec.* 80
46. Brunsdon R. V. 1963 *N. Z. vet. J.* 11 : 144
47. Baker N. F. & Douglas, J. R. 1962 *Vet. Med.* 57 : 412
48. Bremer K. C. 1965 *Aust vet. J.* 41 : 372
49. Davis L. R., Herlich H. & Bowman G. W. 1959 *Am. J. vet. Res.* 20 : 281
50. Davis L. R., Herlich, H. & Bowmann, G. W. 1960 *Am. J. vet. Res.* 21 : 188
51. Levine N. D. 1959 *cit.* Donald, A. D. 1967
52. Roe R., Southcott W. H. & Turner Helen N. 1959 *Aust. J. agric Res.* 10 : 530
53. Donald A. D. 1967 *Aust. vet. J.* 43 : 122
54. Rose J. H. 1963 *Parasitology* 53 : 469
55. Gibson T. E. 1965 *Vet. Rec.* 77 : 1,034
56. Douvres F. W. 1967 Personal communication.
57. Anema S. G. 1967 Unpublished data.
58. Gibson T. E. & Everett G. 1967 *Brit. vet. J.* 123 : 247
59. Ross D. B. 1965 Vet. Rec. 77 : 672
60. Jarrett W. F. H., Jennings F. W., McIntyre W. I. M., Mulligan W. & Urquhart, G. M. 1957 *Vet. Rec.* 16 : 1329
61. Urquhart G. M. 1967 Personal communication.

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Jl S. Afr. vet. med. Ass. 39 (2): 57-59 1968

THE EFFICACY OF NEGUVON* AND RAMETIN* AGAINST *NEOASCARIS VITULORUM (GOEZE 1782)*

S. STAMPA**

SUMMARY

Neguvon was found to be highly effective against *N. vitulorum* at 25—50 mg/kg. Rametin was partially effective at 15 mg/kg and fully effective at 25 mg/kg. Adaptations of the "Egg Counting" technique and the "Critical Test" technique for test work with *Neoascaris vitulorum* are described.

INTRODUCTION

The cattle ascarid worm, *Neoascaris vitulorum* (Goeze, 1782) has received comparatively little attention by research workers in recent years. This may be due to its restricted distribution, limiting its importance to the cattle industry as a whole, although it appears to do a great deal of harm where present. Only young calves are found to carry the parasite. They seem to become free of the worm when 4—6 months old through some form of defence mechanism and remain resistant for the rest of their lives. The larvae migrating through the lungs are held responsible for a transient cough, stunted growth and occasional pneumonia. Adults feed on the intestinal contents but also gnaw on the mucosa and cause further harm through excreted waste products. Farmers maintain that the growth of calves is considerably retarded by this parasite, but no reports of detailed investigations are available.

In the Republic of South Africa,*N. vitulorum* has been found in a narrow strip along the southeast coast of the Cape Province and in the eastern parts of the Soutpansberg in the northern Transvaal. This survey cannot be considered as complete and infestation may occur elsewhere. In Angola, most of the highveld around Nova Lisboa is infested.

EXPERIMENTAL PROCEDURE AND TECHNIQUE

Naturally infested dairy and beef calves from 1 — 4 months old were available for investigation. During the trial they were kept on a similar pasture as they had grazed before the test. The "Critical Test" technique [1] lends itself to efficacy trials of drugs used against this parasite, as such a large worm could hardly be expected to become completely digested after being killed by a drug. As calves found to carry this parasite could not be purchased for slaughter it was necessary to develop techniques not requiring sacrifice.

Neoascaris eggs are readily recognised on coprological examination and egg counts tend to be high. More than 10,000 e.p.g. were noticed in the faeces of some calves. Whether the absence of ascarid eggs indicates absence of worms was investigated in the following way: the droppings of 47 calves were examined three times, on successive days after dosing or at slightly longer intervals. In 39 of these cases all three counts were negative, in seven instances all three counts were positive, and in one instance two counts were positive and one was negative. Six calves with three negative counts were dosed with 250 mg/kg piperazine plus 25 mg/kg Neguvon. They were bagged for 24 hours and no *Neoascaris* specimen was excreted by any animal. The animal with two positive and one negative egg counts was dosed and bagged in the same way. This animal excreted one adult *N. vitulorum*. Absence of *N. vitulorum* eggs in three coprological examinations therefore is regarded as indicating the absence of adult worms.

Using only animals with *Neoascaris* eggs in the droppings the following techniques were employed:

1. *Egg counts only:*

Treatment is regarded as 'fully effective' when no ova are found at all three coprological examinations after treatment. When post-treatment examinations reveal ova, the treatment is known to be not fully effective. Whether it is partially effective or wholly

* Neguvon and Rametin are registered trade names of Farbenfabriken Bayer AG.

** Veterinary Research Station of Farbenfabriken Bayer, P.O. Box 247, Grahamstown.

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ineffective cannot be stated, since eggs were not counted, nor is anything known about the correlation of *Neoascaris* eggs per gram of faeces and the number of worms present. For practical purposes, such cases are regarded as ineffective and classified as such in the Table.

2. *Excretion of worms and egg counts:*

The treatment is regarded is 'fully effective' when worms were excreted after dosing and subsequent coprological examinations for ova were all negative. The treatment is regarded as 'partially effective' when worms were found in the bags and the subsequent coprological examinations were positive. The treatment is regarded as 'not effective' when no worms were excreted.

Neguvon, also known as Dylox, Trichlorphon or Bayer L 13/59 is chemically 0,0 — dimethyl — 2,2,2 — trichloro —1— hydroxyethyl phosphonate. A 10% suspension was prepared for the trials from a wettable powder containing 50% active ingredient.

Rametin, also known as Naphthalophos, or Maretin is chemically 0,0-diethyl-0-naphthalox-imido-phosphate. A 10% suspension was prepared for the trials from a 60% wettable powder. Both drugs were administered orally by means of a 20 ml syringe. Dosages were calculated according to individual liveweights as established with a girdle measuring tape.

RESULTS

The results are listed in the Table.

Treated calves voided from one to 108 specimens of *N. vitulorum* per animal. Within these limits the efficacy of Rametin was not influenced by the number of worms present.

DISCUSSION

Rametin was not effective at 25 mg/kg against *Ascaris suis* in a preliminary trial.

Both Neguvon and Rametin can be regarded as highly effective against *N. vitulorum* at comparably low dosing rates. At the same time both control various other gastrointestinal parasites of cattle. Neguvon is highly effective (80-100%) against *Haemonchus placei* at a dosing rate of 22 mg/kg. It is also highly effective against *Desophagostomum radiatum* at 45 mg/kg. It is effective (60-80%) against *Cooperia punctata* and *C. pectinata* at 55 mg/kg [2]. Higher rates cover a wider spectrum but are not practical under South African conditions [2, 3]. Rametin is highly effective against *H. placei* and *Trichuris spp.* at a dosing rate of 10 mg/kg. It is also moderately effective (50-70%) against *Ostertagia, Trichostrongylus axei, T. rugatus, C. punctata, C. pectinata and C. oncophora* at 15-25 mg/kg [3]. It is higly effective against *Ostertagia* spp.*Cooperia* spp and *Nematodirus* spp. at 50 mg/kg. and effective (60-80%) against *Strongyloides* spp. [4, 5]. Against roundworms in young calves they have thus advantages over piperazine salts which were perviously recommended for the control of *N.*

TABLE: RESULTS OF EFFICACY TRIALS AGAINST **N. VITULORUM** INDICATING THE NUMBER OF ANIMALS IN WHICH THE TREATMENT WAS FULLY-, PARTIALLY- OR NOT EFFECTIVE

No. of animals	Drug	Dose mg/kg	Technique	Efficacy: Full	Partial	Not effective
2	Neguvon	25	egg-count only	2	—	—
9	Neguvon	50	egg-count only	8	—	1
1	Neguvon	50	excretion and egg-count	1	—	—
Total: 12	Neguvon	25-50		11	—	1
1	Rametin	15	egg-count only	—	—	1
2	Rametin	15	excretion and egg-count	2	—	—
Total: 3	Rametin	15		2	—	1
6	Rametin	25	egg-count only	6	—	—
5	Rametin	25	excretion and egg-count	5	—	—
Total: 11	Rametin	25		11	—	—

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vitulorum and which have only an additional effect against *O. radiatum* [6], possibly a degree of efficacy against *Ostertagia* spp and *N. helvetianus*, but are not effective against *H. placei* and *T. axei*, both important roundworms of calves [7].

ACKNOWLEDGMENTS

The author acknowledges the assistance rendered by his Technical Assistant Mr. W. Köpke. He also wishes to express his gratitude towards Dr. R. K. Reinecke for his valuable suggestions concerning the presentation of the manuscript.

REFERENCES

1. Reinecke, R. K. 1963 *Jl. S. Afr. vet. med. Ass.*34 : 233
2. Riek R. F. & Keith R. K. 1958 *Aust. vet. J.*34 : 93
3. Stampa S. In press.
4. Federmann M. 1964 *Dt. tierärztl. Wschr.* 71 : 62
5. Stöber M. & Ende H. 1964 *Berl. Münch. tierärztl. Wschr.* 77 : 100
6. Daykin P. W. 1969 *Veterinary Applied Pharmacology & Therapeutics.* P.131 London. Bailliere, Tindall & Cox.
7. Lapage G. 1962 *Mönnig's Veterinary Helminthology & Entomology,* pp.239,252,256, London. Bailliere, Tindall & Cox.

BOOK REVIEW

VIRUSES OF VERTEBRATES

C. ANDREWES AND H. G. PEREIRA, 2ND ED.

Bailliére, Tindall and Cassell, London, 1967. pp viii & 432, 6 tables, no figures.
Publ. price 70s.

In the revised second edition of this book the authors have adhered to the same brief and concise method of presentation of available information, which has no doubt been the key-note of success in the first edition.

In the light of more recent published and unpublished information, the subject matter has been re-arranged systematically to serve as a basis for the classification of the viruses of vertebrates. In parts I and II of the book, the RNA and DNA viruses, respectively, have been divided into different groups according to accepted criteria. Part III deals with the unclassified, or hitherto unclassifiable, viruses of the various species and includes a chapter on the virus diseases of fish and amphibia. The *Chlamydozoaceae,* whch are not regarded as viruses, have now been excluded from this edition.

In such a bold attempt at classification of viruses the lack of essential and correct information on the one hand and the rapid rate at which new information becomes available on the other are unavoidably responsible for certain errors in grouping.

For instance, the inclusion of horsesickness and bluetongue viruses in the Reovirus group, in this edition, is not warranted at this stage and probably incorrect. However, obvious errors are remarkably few and this seemingly presumptious approach by the authors may serve the useful purpose of stimulating further research and bringing forth the knowledge essential for the final classification of all viruses of vertebrates. Since a wealth of unpublished information may be available at the time, it is respectfully suggested that personal contact with medical and veterinary virologists throughout the world may be helpful in preparing the next edition of this book.

The factual knowledge concerning each virus is presented in such a manner that this book retains its exclusive value as a ready reference and a guide to further study and systematic identification of new viruses. This book is a *sine qua non* to the virologist and the student in this specialized field. Its value may probably be enhanced by the inclusion of illustrations of the morphology and development of individual or representative groups of viruses.

K. E. W.

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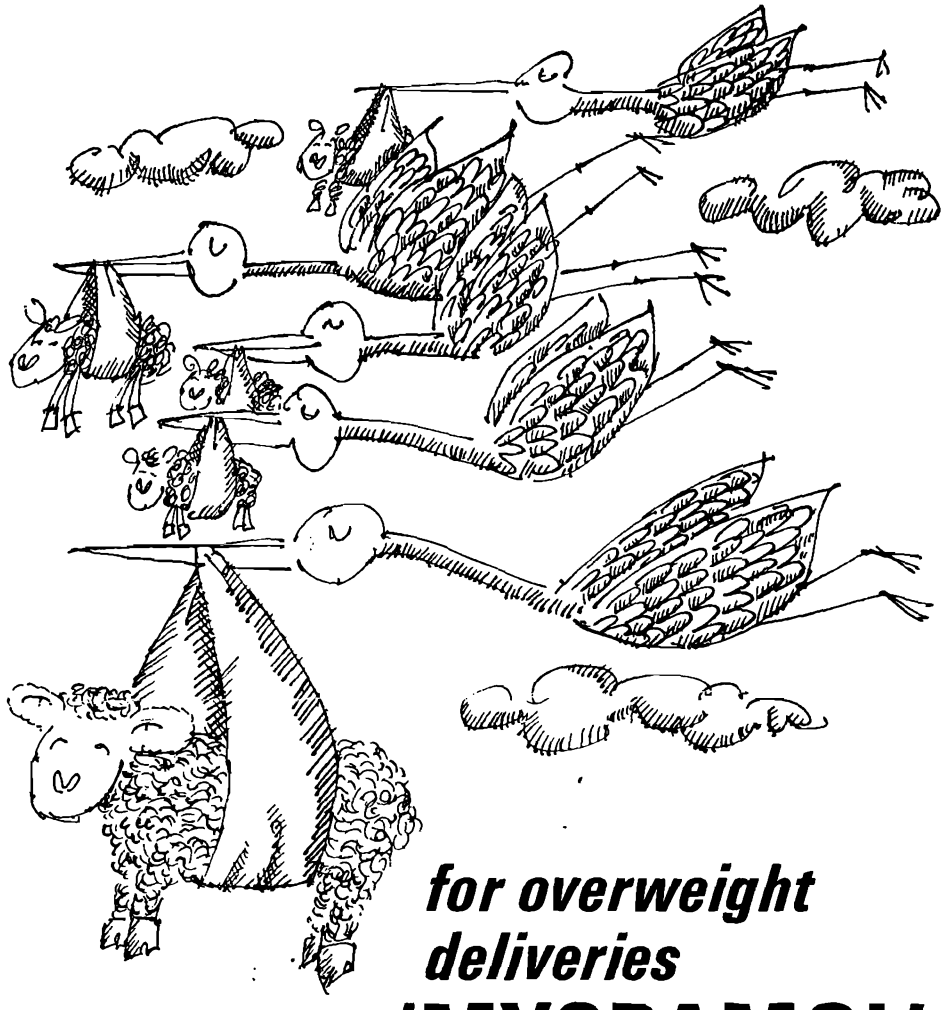

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Jl S. Afr. vet. med. Ass. 39 (2): 61-67 1968

SUPONA* (CHLORFENVINPHOS) FOR CATTLE TICK CONTROL

2. Plunge Dipping Trials

G. E. THOMPSON** AND J. A. F. BAKER**

SUMMARY

Plunge dipping trials were carried out with Supona formulated as a 30% wt/vol water miscible concentrate to establish its suitability for wide scale cattle dipping. Dipwashes prepared from the formulation proved to be chemically stable under foul wash conditions. The dipwashes remained biologically effective for upwards of two years. Excellent biological control was achieved over blue tick *(Boophilus decoloratus)*, red-legged tick *(Rhipicephalus evertsi)*, brown ear tick *(Rhipicephalus appendiculatus)* and bont tick *(Amblyomma hebraeum)* infestations.

In common with all commercially available water insoluble ixodicides, Supona was subject to a measure of preferential removal (stripping) of the active ingredient from the dipwashes. This never reached unacceptable proportions and the diswashes tended to become stabilised after a period of weekly dippings.

Supona as formulated for these trials is suitable for wide scale use against ticks. The initial target concentration in plunge dipping tanks should be 0.05% Supona.

INTRODUCTION

Drummond [1] and Shaw and Baker [2] demonstrated the ixodicidal efficiency of Supona in *in vitro* trials with different species of ticks and field handspraying trials have been reported by Baker and Thompson [3]. Further investigation of the possible use of Supona for general field control of ticks was necessary.

The widest potential use of Supona is in areas of Africa where single- and multi-host tick species occur, especially where ticks have developed resistance to the chlorinated hydrocarbon group of ixodicides [4, 5, 6, 7]. The common method of applying ixodicidal dipwashes to cattle in these areas is by plunge dipping.

Plunge dipping makes certain demands which any chemical must satisfy before it can be considered suitable. The formulated material must be chemically stable under foul wash conditions; it must be physically stable, in that the active ingredient must not undergo too high a degree of preferential removal (stripping) from the dipwash; it must not undergo progressive loss of biological efficiency, as described by Whitnall *et al* [8]; it must be safe when used over extended periods when applied to working oxen and milch cows and when used in conjunction with dosing programmes employing organophosphorus anthelmintics.

The purpose of this paper is to describe a series of trials designed to test the suitability of Supona for use in cattle plunge dipping tanks.

EXPERIMENTAL METHODS

The Supona used in these trials was formulated as a 30% wt/vol water miscible concentrate*. For a short period a 20% wt/vol water miscible concentrate was used in one tank (Wiltonside Fig. 3), but was abandoned as being unsuitable because of preferential removal.

Five tanks situated in the eastern Cape coastal belt, were cleaned, calibrated and charged with Supona. Two, (Tayside and Begha Mouth), had target concentrations of 0.025% Supona; at the other three, (Wiltonside, Burnside and Gusha Mouth), the target was 0.05% Supona. All five tanks were uncovered and subject to flooding by heavy rain, as are the majority of tanks in Southern Africa. When flooding occurred, the super-

* Supona is a SHELL trade mark.

** Cooper & Nephews S. Af. (Pty.) Ltd. East London, South Africa.

* formulated by Research Department, African Explosi ves & Chemical Industries Ltd., Modderfontein, Transvaal.

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ficial excess fluid was skimmed off the surface of the dipwash with as little disturbance of the deeper wash as possible. The quantity of fluid removed was measured and discarded. Thereafter the remaining flood water was treated as a normal replenishment.

Dippings were carried out at seven-day intervals if the weather was suitable. At each dipping, dipwash samples were taken after fifty head had been dipped and again while the last animal was swimming through the tank. Replenishments were made at 150% of the filling rate, i.e., at 0.0375% and 0.075% respectively. At Tayside, the replenishment rate was raised from 150% to 200% at the 19th dipping. The Wiltonside tank was maintained as a long term trial. At the 59th dipping, a new replenishment rate of 0.091% Supona was introduced. This was reduced to 0.083% at the 89th dipping.

At each of two 0.05% Supona tanks, (Wiltonside and Gusha Muoth), and one 0.025% Supona tank, (Begha Mouth), two groups of four oxen were selected. One group was dipped each week immediately before the second dipwash sample was taken; the other group was not dipped. Tick counts were made on both groups each week before treatment according to the method of Baker and Thompson [3]. After 5—7 treatments, the treated and untreated groups were interchanged as a further test of the efficiency of the dipwash.

During the 57th dipping at Wiltonside, three heavily tick infested animals were subjected to tick counts and then passaged through the tank while the second dipwash sample was taken. Care was taken to ensure that all parts of the animals were well wetted, and dipwash was taken from the dip tank whilst the animals were being dipped and poured into the ears. Twenty-four hours later a further tick count was made to test the biological efficiency of the fouled dipwash.

At Burnside and Wiltonside, routine whole blood cholinesterase determinations were made on regularly dipped animals and on untreated animals running in the same herd. At Wiltonside, these tests were made on adult oxen, whereas the Burnside test animals included dry cows, cows in calf, cows in milk, heifers and young calves (all being pedigree Guernsey cattle.) On this farm a routine dosing programme embodying the anthelmintic, Haloxon* was carried out together with the weekly dippings. Cholinesterase determinations were made according to Jolly and Ratcliffe [9].

RESULTS

Stability trials

The graph of Wiltonside (Fig. 1) and the histograms for the four other tanks (Fig. 2), show that Supona, as formulated for these trials is preferentially removed from the dipwash. A replenishment rate of 150% of the filling rate is insufficient to maintain the target concentration in all instances. A 186% replenishment rate is too high. The regular differences between samples one and two at each dipping are shown in the histograms (Fig. 2), and indicate that the loss of active ingredient is due mainly to preferential removal and not to chemical degradation.

Biological trials

Fig. 3, 4 and 5 illustrate the biological efficiency of Supona in plunge dip tanks against single- *(B. decoloratus)* and multi-host ticks *(R. evertsi, R. appendiculatus, A. hebraeum)*.

The table shows conclusively that Supona does not lose its biological efficiency under foul wash conditions.

Safety trials

Results of routine whole blood cholinesterase determinations at Burnside and Wiltonside are shown in Fig. 7. No unacceptable degree of blood cholinesterase depression was observed on either farm. No clinical signs of toxicity were seen on any of the farms used in the trials.

DISCUSSION

Supona, as formulated for these trials, is subject to preferential removal from dipwashes. The degree of preferential removal is not severe and falls well within the range of water insoluble ixodicides previously marketed for general use. As expected, the degree of preferential removal was highest during the first few dippings and tended to decrease as the dipwash matured. At all diptanks, with the exception of Burnside, con-

* Haloxon: (O, O di (2-chlorethyl) O-(3-chloro-4-methyl coumarin-7yl phosphate). Registered Trade Mark - Cooper, McDougall & Robertson, Ltd., Berkhamsted, U.K.

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centrations of Supona in the dipwash tended to rise towards the target concentrations with replenishments. When the replenishment rate was increased from 150% to 186% of initial filling rate, at Wiltonside the concentrations of Supona in the dipwash rose to 0.060%.

Data supplied in Figures 1 and 2 show that in average size tanks with relatively small numbers of cattle being dipped, such as Gusha Mouth and Begha Mouth, which were not flooded, each replenishment gave a marked rise in active ingredient and brought the ixodicide content back to the initial target concentration. At Wiltonside and Burnside, on the other hand, most of the big replenishments resulted from flooding and there was not the same sharp rise in the dipwash concentrations. The lowest recorded level at Wiltonside was at the 39th dipping and after the tank had been flooded three times in a period of four weeks. Burnside went without replenishment from the 14th to the 23rd dipping. During this period 1350 animals were dipped and the ixodicide level dropped only six points from 0.044% to 0.033%. Tayside data are not comparable as 800 head per week were dipped in this 3000 gallon tank, which is of inadequate size for this number. In this trial it was necessary to increase the replenishment rate to restore the desired concentration of Supona.

No detectable chemical degradation of Supona took place in any of the dipwashes used in the trials. With due allowance for unavoidable variations in sampling and analysis, the consistent drop in ixodicidal content between the first and second samples at each dipping indicates that chemical loss is due to preferentical removal. The satisfactory behaviour of all the dipwashes shows that this formulation of Supona is suitable for use in the preparation of dipwashes.

The biological results achieved were most satisfactory. This is particularly noticeable with bont ticks and blue ticks, which, because of their preferential attachment sites, are vulnerable to efficient ixodicides applied by plunge dipping. Regular blue tick scrapings at Wiltonside (Fig. 3) and regular total bont tick counts at Begha Mouth, (Fig. 5), Wiltonside (Fig. 3) and Gusha Mouth (Fig. 4.) show that excellent control of these ticks was achieved even against such severe challenges as experienced at Gusha Mouth (Fig. 4) and Begha Mouth (Fig. 5). No engorged specimens of these ticks were found and, after the second treatment, only larval stages of the blue tick were seen. When the treated and control groups were interchanged the pattern was quickly reversed. These results do not differ from the findings of Baker and Thompson [3].

The immature stages of the red-legged tick and to a lesser extent, the adult stage of the brown ear tick, have attachment sites which show up the limitations of the plunge dip for controlling these ticks. At Gusha Mouth and Begha Mouth, the nymphal stages of the red legged tick were seen at most inspections, a factor not encountered when Supona washes are applied by handspraying[3].

The brown ear tick was not easily controlled. The trials were conducted during the short period of severe brown ear tick activity. Early in the experiment the tick challenge was low and as it rose the ears of the treated group remained relatively clean, and contained only odd engorged specimens. The ears of the untreated animals became very dirty with damaged skin surfaces which bled easily. When the two groups were interchanged there was no immediate improvement to these damaged ears and no marked improvement until the tick challenge started to wane. These results are contrary to those previously reported [3], when ixodicide application was handsprayings. They emphasise the necessity for the handdressing of ears infested with brown ear tick in addition to general application by means of plunge dipping. At Wiltonside, (Table), where care was taken to ensure thorough wetting of all parts of the animals, excellent effect on all stages of the tick species was achieved. Thus it is the method of application rather than the insecticide which is at fault.

The continued efficiency of Supona in cattle dipping tanks is well demonstrated by the spectacular effect on heavy tick infestations of the three animals dipped at the end of the 57th weekly dipping at Wiltonside. This excellent result was achieved despite a dipwash analysis of 0.043% Supona. Reference to Figures 3, 4 and 5 shows that tick control remained good when the ixodicidal level in the dipwashes dropped as low as

* Toxaphene, registered trade mark of Hercules Incorporated, Wilmington, U.S.A.

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0.017% (Begha Mouth). At this concentration, however, residual protection would be too short to control severe challenges from the quick feeding rhipicephalid ticks[3]. A target concentration of 0.05% Supona allows for preferential removal of insecticide whilst ensuring excellent tick control.

No signs of toxicity were observed at any time time in any class of cattle used in these trials. Regular whole blood cholinesterase determinations confirmed that there is no increased toxicity hazard when Supona dipwashes are allowed to become foul. In all cases maximum cholinesterase depression occurred in the treated animals during the winter months when all animals were under stress.

The increasingly widespread incidence of strains of blue, brown ear and red-legged ticks resistant to the chlorinated hydrocarbon group of insecticides enchances the value of Supona, particularly as it is the first organo-phosphorus compound which can match the former efficiency of Toxaphene* against the multi-host ticks at safe and economical concentrations.

REFERENCES

1. Drummond R. O. 1962 *J. econ. Ent.* 54:1050
2. Shaw R. D. & Baker J. A. F. 1966 *Vet. Rec.* 78:864
3. Baker J. A. F. & Thompson G. E. 1966 *Jl. S. Afr. vet. med. Ass.* 37:367
4. Whitnall A. B. M., Thorburn J. A., McHardy W. M., Whitehead G. B. & Meerholz F. 1952 *Bull. ent. Res.* 43:51
5. Whitehead G. B. 1959 *"Insecticide resistance in the Blue tick,* (Boophilus decoloratus) Koch, in South Africa". Ph. D. Thesis Rhodes University.
6. Whitehead G. B. & Baker J. A. F. 1961 *Bull ent. Res.* 51:755
7. Baker J. A. F. & Shaw R. D. 1965 *Jl. S. Afr. vet. med. Ass.* 36:321
8. Whitnall A. B. M., Bradford B., McHardy W., Whitehead G. B. & Meerholz F. 1948 *S. Afr. J. Sci.* 45 : 113
9. Jolly D. W. & Ratcliffe B. D. 1958 *Vet. Rec.* 70:289

TABLE: CHECK ON CONTINUED BIOLOGICAL EFFICIENCY OF SUPONA DIPWASH: WILTONSIDE; 57th DIPPING; MULTIHOST TICK COUNTS AVE. OF 3 ANIMALS

	BONT TICKS			RED LEGGED TICKS				BROWN TICKS			
	Nymphs	Adults	E.F.	Larvae	Nymphs	Adults	E.F.	Larvae	Nymphs	Adults	E.F.
Ante dipping	+ + F	198	F	+ + F	+ + F	51	F	+ F	+ F	63	F
Post dipping	0	1	f	0	0	0		0	0	0	

Dipwash sample showed 0.043% Supona.
E.F.—Female Engorgement. f—unengorged females only. F—Fully engorged females present.
+—1-25 specimens approximately.
+ +—26-60 specimens approximately.
+ + +—More than 60 ticks.

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Fig.1

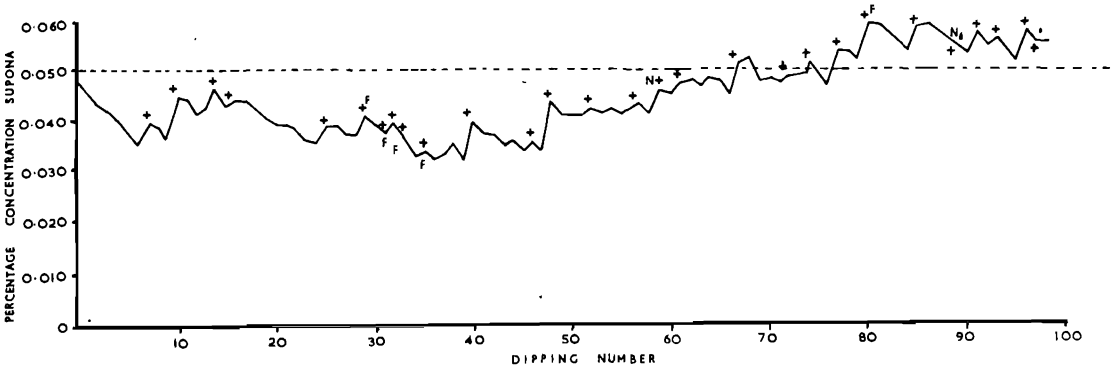
WILTONSIDE LONG TERM DIPPING

TARGET CONCENTRATION 0·05% SUPONA

AVERAGE OF TWO SAMPLES FROM EACH DIPPING

TANK CAPACITY: 3,750 GALLONS

AVERAGE OF 250 HEAD OF CATTLE PER DIPPING



\+ REPLENISHMENTS MADE PRIOR TO THESE SAMPLES BEING TAKEN

F TANK FLOODED

N REPLENISHMENT RATE INCREASED TO 186 % OF FILLING RATE

N_1 " " DECREASED " 166 % " " "

TANK FLOODED PRIOR TO DIPPINGS	
DIPPING NUMBER	GALLONS REMOVED BEFORE REPLENISHMENT CALCULATED
29	300
31	170
33	190
35	210
80	700

Fig.2

DIPWASH SAMPLES

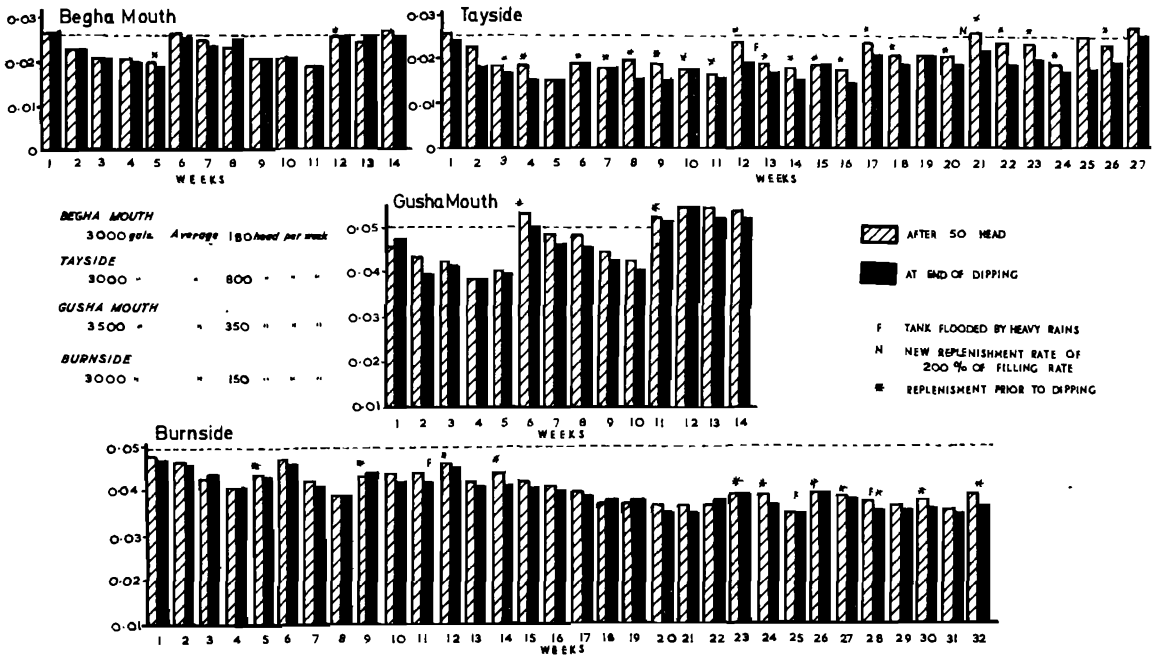


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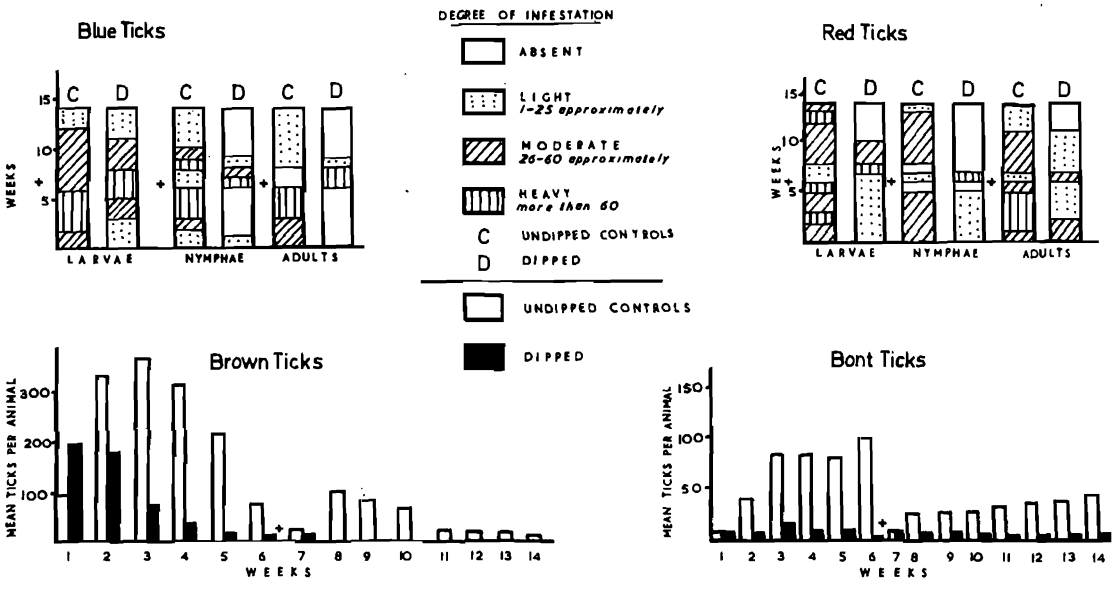
Fig. 3

WILTONSIDE

WEEKLY ASSESSMENTS, COUNTS AND TREATMENTS



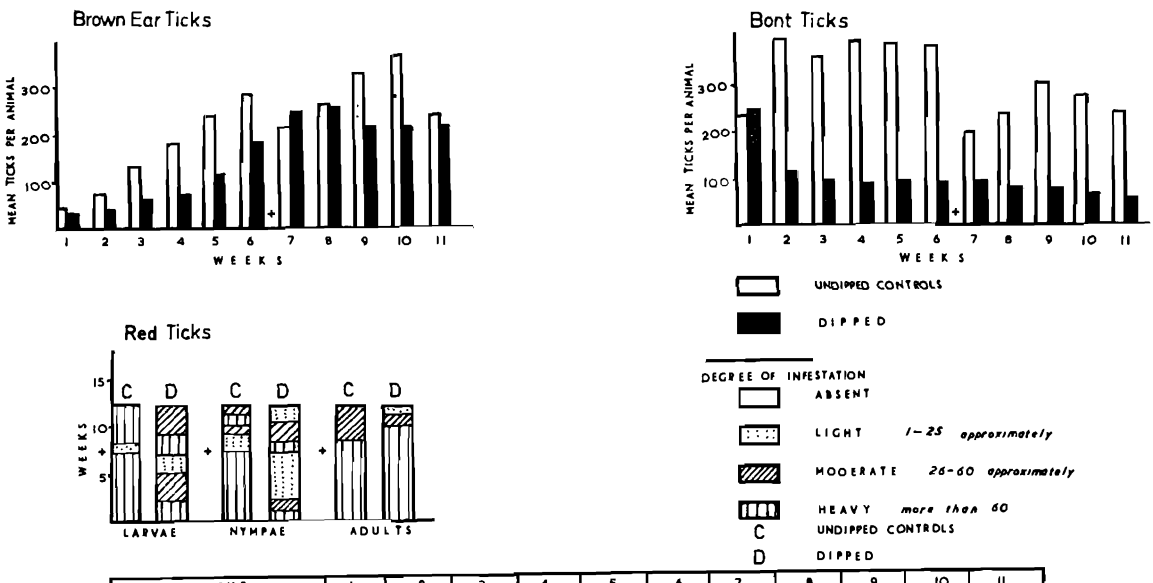
TREATMENT	1	2	3	4	5	6	7	8	9	10	11	12	13
% SUPONA IN DIPWASH	0·046	0·041	0·037	0·040	0·039	0·036	0·038	0·036	0·035	0·033	0·031	0·030	0·029

+ CONTROL AND TREATED GROUPS INTERCHANGED

Fig. 4

GUSHA MOUTH

WEEKLY ASSESSMENTS, COUNTS AND TREATMENTS



TREATMENT	1	2	3	4	5	6	7	8	9	10	11
% SUPONA IN DIPWASH	0·039	0·041	0·038	0·039	0·049	0·046	0·045	0·042	0·040	0·051	0·054

+ CONTROL AND TREATED GROUPS INTERCHANGED

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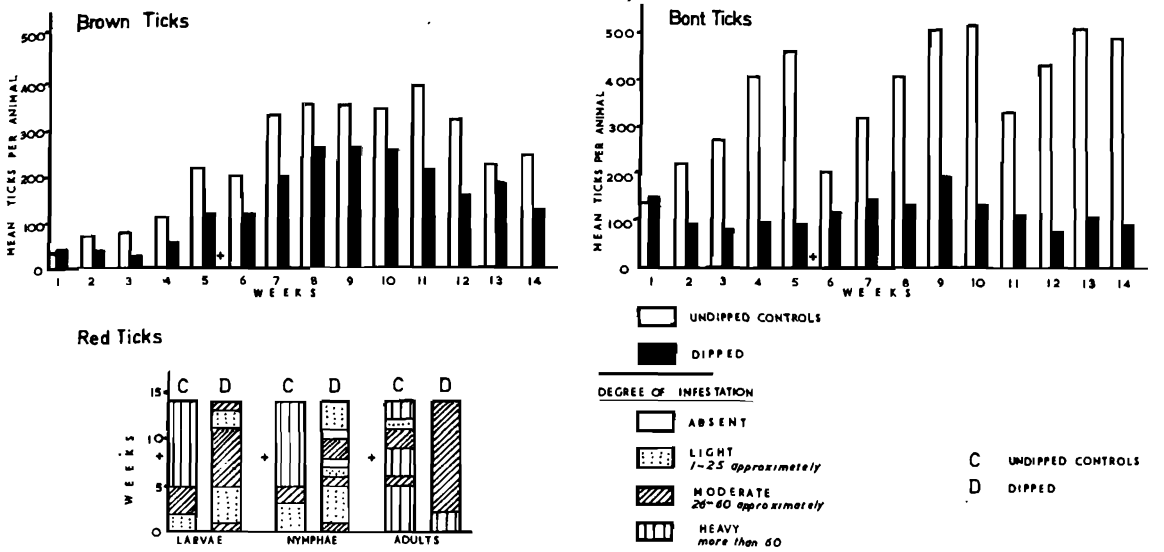
Fig. 5

BEGHA MOUTH

WEEKLY ASSESSMENTS, COUNTS AND TREATMENTS

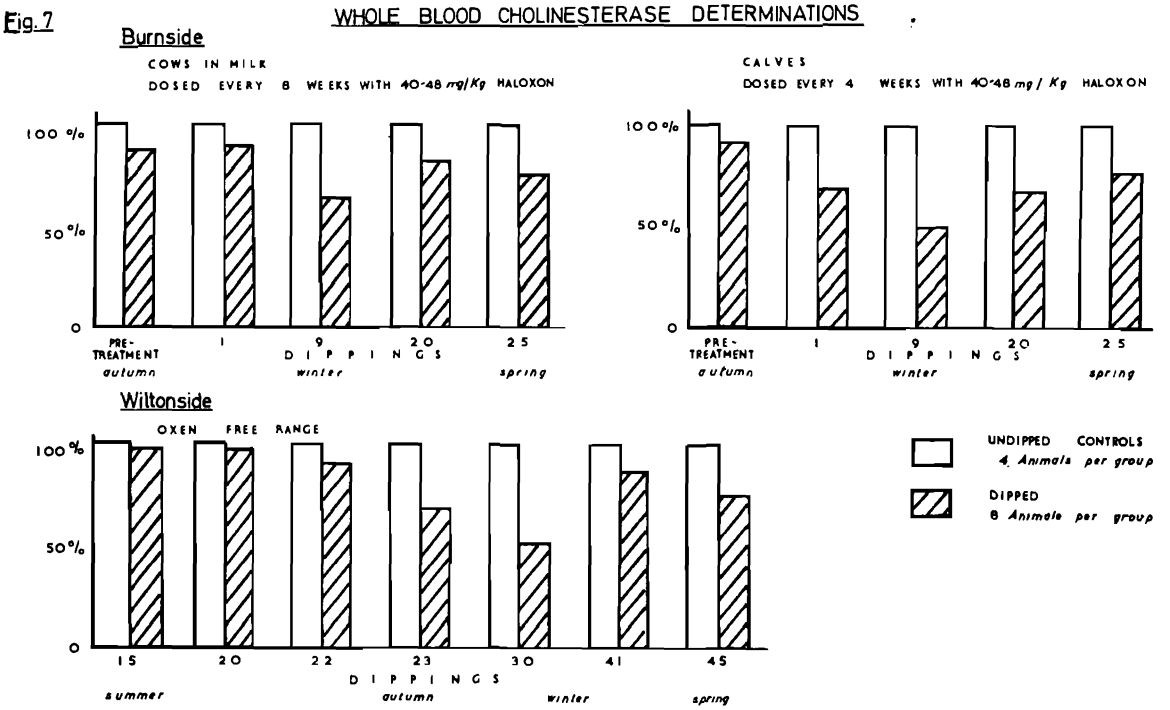


TREATMENT	1	2	3	4	5	6	7	8	9	10	11	12	13
% SUPONA IN DIPWASH	0·026	0·022	0·020	0·019	0·018	0·024	0·022	0·024	0·020	0·020	0·017	0·025	0·023

+ CONTROL AND TREATED GROUPS INTERCHANGED

Fig. 7

WHOLE BLOOD CHOLINESTERASE DETERMINATIONS



CHOLINESTERASE LEVELS DEPICTED ARE THE AVERAGES OF GROUPS, SEVEN DAYS AFTER THE TREATMENTS SHOWN

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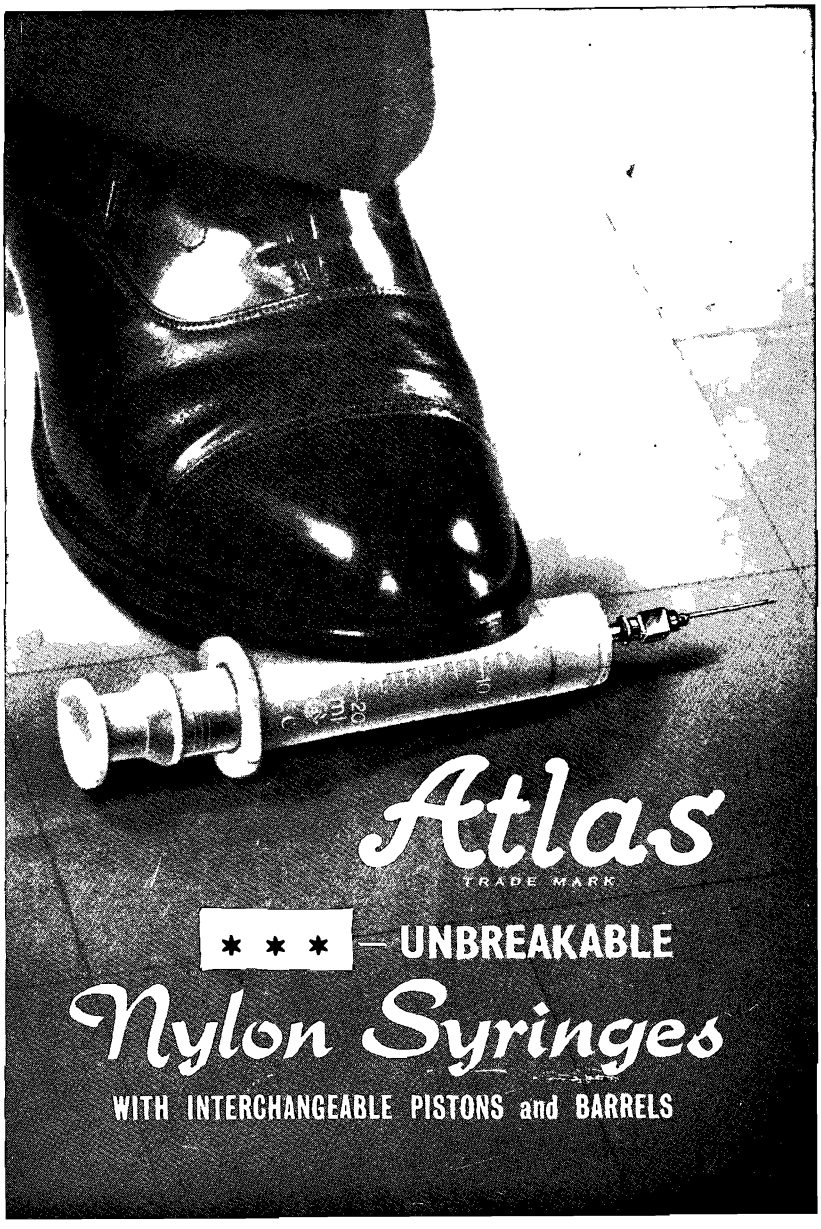

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ORGANOPHOSPHATE POISONING IN FARM STOCK IN SOUTHERN AFRICA*

H. J. J. TERBLANCHE**

SUMMARY

An account is given of organophosphate poisoning in livestock by dealing with absorpion, excretion, storage, effect on body tissues, symptomatology, pathology, diagnosis and treatment. Various factors considered to be contributory to poisoning are also briefly discussed. These include factors such as nutrition, metabolic diseases, diseased organs, viral and bacterial diseases, poisonous plants, urea, copper sulphate, malpractices and stress.

INTRODUCTION

Phosphoric acid esters (commonly known as organophosphates) are widely used in the veterinary field and there is every reason to believe that they have come to stay. They are applied externally as dipping materials, dusting powders, blowfly remedies, washes and shampoos, and internally as anthelmintics. Many organophosphates are used as plant pesticides. Both man and beast are frequently poisoned by these compounds via inhalation, skin absorption or oral intake.

ABSORPTION

These compounds vary as regards their dermal, oral and inhalation toxicity. The rate of absorption depends largely on the solubility in fats and water. Some are highly toxic percutaneously but much less toxic orally, e.g. trichlorphon, which is well tolerated orally at 75 to 100 mg/kg by cattle, causes toxic symptoms at 10 mg/kg when applied dermally in an oily base as a pour on. Organophosphates are all absorbed from the gastro-intestinal tract where they are hydrolysed. They are usually demonstrable in the blood about half-an-hour after oral intake. The pH of the digestive tract influences the way in which the radicals are split, and this again determines the rate and course of poisoning. In a more acid medium, for instance, the phenols are split off whereas in a more alkaline *milieu* the alcohol radical separates. The blood stream transports the compound to the body tissues, where it is further hydrolysed by esterases. These esterases are particularly active in the plasma, liver and kidneys.

EXCRETION

Organophosphates and their decomposition products are mainly excreted by the kidneys. Maximum levels in the urine are usually reached within 6-12 hours after absorption. Some are excreted in small quantities in the milk, e.g. trichlorphon, whereas others such as parathion are not. Small quantities are also excreted in the faeces. The latter phenomenon is well demonstrated by the way flies are killed when they settle on the faeces of sheep, cattle, pigs and poultry, which have been treated with trichlorphon or coumaphos. Storage takes place mainly in the kidneys, liver, skin, fat and, to a small degree in nervous tissues.

EFFECT ON BODY TISSUES

In the normal animal acetylcholine is released at the synapses of cholinergic nerves to carry the stimulus across. The acetylcholine has to be destroyed (hydrolysed) immediately post-synaptically into choline and acetic acid. This hydrolysis is accomplished by cholinesterases. Organophosphates inhibit these cholinesterases and consequently acetylcholine accumulates post-synaptically. The different organophosphates vary in their ability to inhibit cholinesterases. Not only breeds, but individuals within a breed, also vary considerably in their susceptibility to cholinesterase inhibition. Angora goats, for

* Paper presented at the 62nd Annual Congress of the S.A.V.M.A. Durban, October, 1967.

** P.O. Box 1366, Johannesburg.

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instance, were found to be more susceptible to poisoning than sheep. Furthermore, it is well known that very young animals are more easily poisoned than mature animals.

POISONING

General Symptomatology:

The symptoms are those of acetylcholine accumulation, *viz.* respiratory disorders, such as spasms, increased bronchial secretion and diaphragmatic contractions, bradycardia, salivation, diarrhoea, colic and flow of digestive juices, lacrimation, muscular tremors, cramps, disturbed equilibrium and paralysis.

Sheep and Goats:

Restlessness, profuse salivation, laboured breathing, stretching, frequent micturition, diarhoea, weakness and paralysis.

Cattle:

Distress, listlessness, colic, recumbent position with legs outstretched, muscular tremors and inco-ordination, frequent micturition, diarrhoea and paralysis.

Pigs:

Salivation, muscular tremors and inco-ordination, jerking of extremities, twitching of ears and tail, paresis and paralysis.

Horses:

Restlessness, inco-ordinated movement, sweating and general colic symptoms.

PATHOLOGY

Organophosphate poisoning causes functional disturbances of organs. The morphological changes which take place are either slight or atypical or secondary and consequently no definite diagnosis is possible on macroscopic or microscopic examination of organs.

Macroscopic changes

These are usually one or more of the following: hyperaemia and oedema of lungs, congested liver, catarrh and inflammation of the anterior portion of the gastro-intestinal tract, petechiae in mesentery, congestion of kidneys. Hyperaemia and oedema of the lungs and congestion of the kidneys are the most constant lesions. In Angora goats, haemorrhagic duodenitis is common, and in a large percentage of these cases the duodenum is ruptured, resulting in acute peritonitis.

Microscopic changes

These include usually mild degrees of degeneration of the myocardium, adrenal medulla and liver. Hyperaemia and oedema of the lungs and both interstitial and glomerulonephritis are common. In some cases, abomasitis, gastritis and duodenitis are present.

DIAGNOSIS

Since no pathognomonic lesions are produced by organophosphate poisoning, diagnosis can only be arrived at by considering the case history, symptoms, pathological changes, by elimination of bacterial and viral infections as well as chemical analysis of skin, digestive tract contents, milk, urine, blood and tissues.

TREATMENT

Poisoned animals should be treated swiftly and specifically as soon as possible after onset of symptoms. In addition to specific treatment, symptomatic nursing is desirable. Atropine is still the antidote of choice and P.A.M. (pyridine-2-aldoxime-N-methyl-iodide) is an excellent drug for supplementary treatment. The dosage of the latter is recommended at a level of about 10 mg/kg bodyweight, *viz.*, the standard 0.5 gram pack is sufficient for about 100 lb bodyweight. Where animals have been poisoned percutaneously it is essential to wash them thoroughly with soap and water.

Sheep and Goats:

Inject atropine at the rate of 16 mg per adult animal (0.35 mg/kg). Half this dose should be given intravenously and the other half subcutaneously. Atropine dissolved in peanut oil is an ideal antidote for repeat treatments, which should follow four hours after the intitial dose. When necessary, follow with P.A.M. intravenously, at a dosage level of 10 mg/kg.

Cattle, Pigs and Horses:

Inject atropine at the rate of 2 mg/kg body weight: half the dose intravenously and the other half subcutaneously. In addition, inject P.A.M. intravenously at the rate of about 10 mg/kg. Repeat the treatment, if necessary, after 4 - 6 hours.

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FACTORS CONTRIBUTING TO ORGANOPHOSPHATE POISONING

Nutritional factors:
Nutritional state and mineral balance are difficult to assess accurately and do not constitute a fixed pattern. A high percentage of lactating dairy cows in various bodily conditions receiving concentrates were found to react adversely to dosing with some organophosphates. Lactating veld cows, on the other hand, tolerated these organophosphates much better. It is not clear whether the concentrated feed, or the continuous drain on the mineral balance via milking or both, account for this higher susceptibility.

Metabolic diseases:
The ketonaemic complex in cattle and "domsiekte" in sheep have been directly incriminated in organophosphate poisoning. Animals with a high intake of grain, where acidosis was apparent, have shown a high susceptibility to some organophosphates. In the latter case ulceration of the ruminal wall is possibly a major factor.

Organ disorders:
Factors which fall into this category are pathological changes and particularly conditions such as abscessation and adhesions of the lungs and cirrhotic and abscessed livers, both commonly found in sheep and cattle. Liver damage caused by fascioliasis must be included here. A faulty liver, which cannot play its normal rôle in the detoxification process of the body, is commonly seen in animals which have succumbed to organophosphate application.

Viral and bacterial diseases:
Febrile conditions such as blue tongue, bacterial icterus and ephemeral fever, have been found to be underlying factors which rendered affected animals highly susceptible to organophosphate poisoning. Several cases of enterotoxaemia in sheep have followed organophosphate dosing.

Poisonous plants:
Cases of organophosphate poisoning have occurred where plants containing prussic acid were grazed, *e.g. Cynodon* (kweek) and sorghums, such as kaffircorn and babala, and pods from certain trees. Toxicities were also encountered where *Amaranthus* spp. (hanekam) and radishes were eaten in large quantities prior to dosing. These plants contain nitrates, which themselves are often toxic. Some animals on senecio veld have also displayed sensitivity to organophosphate poisoning. In these cases liver pathology predisposed to these toxicities.

Urea:
Urea in itself is often responsible for mortality in sheep and cattle. Where animals have consumed this material, the dosing of some organophosphate has been found to cause side-reactions.

Copper sulphate:
This material is used as an oesophageal groove stimulant prior to administration of some anthelmintics. At times this material is not only incorrectly used but unfortunately rather recklessly. Many farmers even use the crystals (powder) for predosing and not the recommended 10% solution. In several instances the crystals were used in quantities of 2-6 g per sheep, and the solution in volumes of 20 to 45 ml per sheep. This represents an overdosing factor of tenfold and higher. Recommended copper sulphate solution dosage is equivalent to approximately 20 mg/kg. Specific trials in sheep with copper sulphate and organophosphates produced the following results:-
Sheep tolerated copper sulphate in both solution and crystal form to a level of 130 mg/kg. Above this level, serious side reactions and mortalities occurred. When copper sulphate at various lower levels was tested in combination with organophosphates like Trichlorphon and Coumaphos at recommended dosage levels, side reactions occurred at copper sulphate levels of 60 mg/kg and mortalities at 100 mg/kg. When these organophosphates were dosed at double dosage levels combined with copper sulphate at recommended level, no mortalities occurred. High dosage levels of copper sulphate, therefore, are dangerous in combination with some organophosphates.

Simultaneous dipping and dosing:
This practice can only increase the possibility of poisoning. Not only should the simultaneous use of organophosphates internally and externally be avoided, but also other combinations *e.g.* chlorinated hydrocarbon dips and organophosphate and arsenical anthelmintics. Such combinations are used often and are responsible for unnecessary losses.

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Maldosing:

Both overdosing and dosing the wrong materials have resulted in intoxication. Overdosing results mostly from wrong dilution rates and faulty dosing equipment. Cases have been investigated where materials such as Dieldrin † (insecticide) and Lujet (R) (Blowfly remedy) and BHC powder have inadvertently been dosed. Furthermore, dosing into the respiratory tract is also encountered.

Stress:

Organophosphates and other stock remedies are often blamed for losses when administered to animals under stress. Stress factors which have been encountered in this connection are: dehorning (cattle); docking and castration (lambs); high ambient temperature (pigs); obesity and emaciation; fatigue, particularly after having being driven or transported over long distances; and parturition.

REFERENCE

Debackere M. 1963 *Vlaams diergeneesk. Tijdschr.* 32:361

† Registered Trade Mark of Shell Chemicals.
(R) Registered Trade Mark of BAYER GERMANY.

LETTER TO THE EDITOR

Sir,

The following clinical information may interest your readers:

1. *Unusual symptoms of contagious pustular dermatitis:* Recently I observed some unusual cases of distribution of the lesions of CPD amongst young lambs and I would be interested in knowing if other practitioners in sheep areas have had similar experiences. In one outbreak severe typical lesions were found on the teats of ewes, with milder lesions on the lips. Some of the lambs of these ewes showed mild lip lesions and severely affected areas on their backs. The latter lesions were acute, multiple, superficial and disseminated with a purulent exudate. Since the exudate caused matting of the wool in the areas concerned the animals appeared to have Lumpy Wool. Material taken from these lesions proved to be CPD positive following biological and histo-pathological examination. In a flock of 50 lambs twenty showed body lesions, 12 showed lip lesions and four died. Complete recovery occurred amongst the surviving lambs in two to three weeks but the animals concerned remained poor doers.

2. *Treatment of rectal prolapse with 2% phenol oinment:* (i) A three to four year old mare with a rectal prolapse was treated by cleaning the prolapsed part with "Cetavlon" and covering it with 2% phenol ointment before correction. The patient was given 500 mg of chloropromazine intramuscularly and one ounce of magnesium sulphate daily for a week. On the day following initial treatment the prolapse recurred twice and was corrected each time after application of phenol ointment. It recurred again two days later and after treatment in the same manner did not recur again. (ii) A rectal prolapse of about 3-4 inches and of four days standing was encountered in a one-year old Dorper ewe. Phenol ointment was applied once before reduction, three times the following day and once on the third day after correction. The animal was kept on green food for six days and then fed concentrates and dry hay. Prolapse occurred again after the change in diet. After reduction again and treatment as above the animal was kept on green food. Prolapse did not recur again during a two week period of observation following the final operation.

D. J. Thornton, Graaff Reinet.

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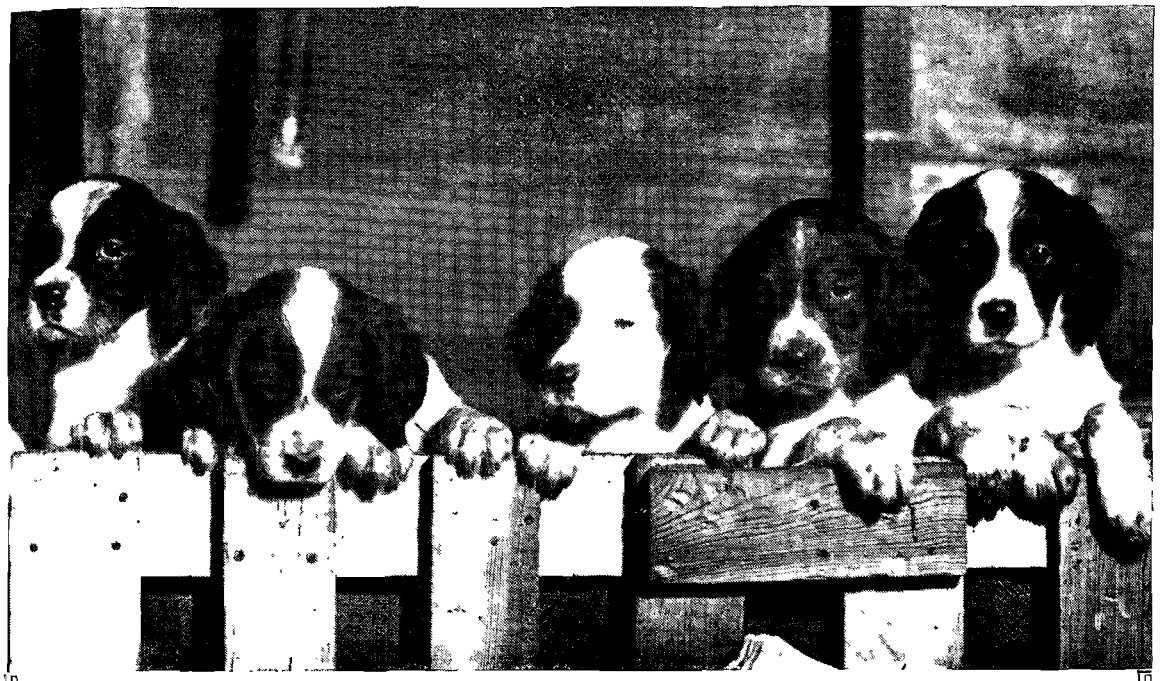

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DIE DIAGNOSE VAN VIBRIOSE BY BEESTE

B. J. H. BARNARD*

SUMMARY

The serological and bacteriological diagnosis of *Vibrio fetus* infection in cattle is discussed. Attention is paid to the more important aspects of specimen collection and dispatch to the laboratory. Methods for isolation, culture and typing are briefly summarised.

INLEIDING

Weens die toenemende belangrikheid van vibriose by beeste en die probleme wat ontstaan ten opsigte van diagnose, gesien van die kant van die privaatpraktisyn of staatsveearts wat monsters moet neem, sowel as van laboratoriumwerkers in verskeie diagnostiese sentra, word hiermee praktiese aanwysings in die lig van die jongste bestaande kennis gegee.

SEROLOGIESE DIAGNOSE

Die gebruik van serum vir 'n diagnose van vibriose is van geen praktiese waarde nie, omdat besmetting baie selde gepaard gaan met 'n waarneembare serumtiter. 'n Positiewe titer is gewoonlik ook van korte duur. Derhalwe word die slymagglutinasietoets gebruik vir die diagnose van die siekte by beeste.

Die vaginale slymagglutinasietoets is 'n kuddetoets. Dit is dus nutteloos om slegs individuele diere te toets. Die aantal monsters wat per kudde getoets word speel 'n belangrike rol in die diagnose van die siekte. Van alle monsters wat op Onderstepoort gedurende die periode 1964-67 getoets is, was 21 persent positief of verdag. Waar vyf of minder monsters per kudde getoets was, is slegs 15 persent van die kuddes positief of verdag positief gevind. Waar daar egter 10 of meer monsters per kudde ondersoek is, kon 'n positiewe of verdag-positiewe diagnose van 53 persent van die kuddes toegesê word. Bevindings van die Animal Husbandry Research Centre, Australië, is soortgelyk: met gemiddeld agt monsters per kudde, was 21 persent daarvan positief terwyl 41 persent van die getoetste kuddes besmet was. Hulle beveel 8-10 monsters per kudde aan. Die optimum getal monsters kan dus op 10-15 per kudde gestel word.

Agglutinerende teenliggaampies is in die vaginale slym waarneembaar vanaf vier tot tien weke na besmetting. Enkele diere kan vir jare daarna nog positief bly, maar in die meeste gevalle is die toets na 'n paar maande weer negatief. Dit is gevolglik van groot diagnostise waarde om slegs monsters te neem van verse, of van koeie wat voorheen nie besmet was nie, 1-6 maande nadat hulle deur 'n verdagte besmette bul gedek was. Dragtige diere kan ook besmet wees en monsters van sulke diere kan ook gebruik word vir die slymagglutinasietoets.

Dit is verder van belang dat slegs die standaardtampon, deur Onderstepoort uitgereik, gebruik moet word. Tuisgemaakte tampons verskil in grootte, sodat daar nie 'n konstante hoeveelheid slym opgeneem word nie, met gevolglik verwarrende resultate. Tampons met bloed, mis of urine besoedel gee ook dikwels misleidende resultate. Die tampon moet in dieselfde houer teruggeplaas word en sonder enige byvoeging na die naaste laboratorium gestuur word.

DIE VAGINALE SLYMMAGGLUTINASIETOETS

By ontvangs bevat die tampon ongeveer 0.85 gram vaginale slym; 8.5 ml van 'n 0.85% soutoplossing word by die tampon in die houer gevoeg wat dan vir minstens 12 uur by 4°C gelaat word. Tampons kan vir etlike dae in die soutoplossing by 4°C gelaat word voordat die toets gedoen word.

Stel nou vir *elk* van die drie antigene,

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verkry van drie verskillende stamme van *V. fetus* waarteen getoets word, 'n reeks glasbuisies van 5 mm x 80 mm as volg op: vier buisies vir 'n positiewe kontrole met 'n positiewe serum vir die besondere antigeen; vier buisies vir 'n negatiewe kontrole met 'n standaard negatiewe serum, en vier buisies vir elke monster wat getoets moet word; merk die buisies.

Plaas nou 0.5 ml formolsoutoplossing * in elke buisie. In die eerste buisie van elke groep van vier, word nou onderskeidelik 0.5 ml positiewe serum, 0.5 ml negatiewe serum en 0.5 ml van die tamponekstrak geplaas, sodat daar nou in elke eerste buisie 1 ml vloeistof is.

Die vloeistof in die eerste buisie van die positiewe serumgroep word nou gemeng en 0.5 ml oorgedra na die volgende buisie. Hou so aan tot by die vierde buisie en gooi die laaste 0.5 ml weg. Herhaal die verdunningsmetode vir die negatiewe- en vir die tamponekstrakgroep. Twee verdere soortgelyke reekse soos die pas beskrewe een word opgestel. Laastens word 0.5 ml van die onderskeie drie antigene in elke buisie van die betrokke reeks gevoeg.

Die toetsbuisies word nou vir 12-18 uur in 'n broeikas by 37°C geplaas en dan gelees teen 'n swart agtergrond met 'n helder lig van agter.

Vertolking van die resultate: Die volgende verdunnings word deur bogenoemde tegniek verkry: buis een 1:40, buis twee 1:80, ens. 'n Verheldering van ongeveer 100% in die boonste gedeelte, met 'n duidelike wit oorgangslyn na die geagglutineerde gedeelte, word as 'n vier plus reaksie beskou. Waar die verheldering ongeveer 75% is, met 'n 'n duidelike oorgangslyn, word die reaksie as drie plus beskou. By 'n twee en een plus reaksie is die verheldering ongeveer 50% en 'n onduidelike of geen oorgangslyn sigbaar nie.

'n Positiewe reaksie vir 'n bepaalde antigeen word verteenwoordig deur 'n drie- of 'n vierplusreaksie in die tweede of verdere buisie, d.w.s. 'n titer van 1:80 of hoër. Om 'n positiewe diagnose van 'n monster te maak moet met minstens twee antigene 'n positiewe reaksie by 'n titer van 1:80 of hoër verkry word.

'n Verdagte diagnose word gemaak waar agglutinasie voorkom hetsy by 'n verdunning van 1:40 of net by een antigeen.

* 1 ml formalien per liter 0.5% NaCl-oplossing.

BAKTERIOLOGIESE DIAGNOSE

Die bakteriologiese diagnose van *Vibrio fetus* lewer die betroubarste resultate op die individuele dier. Veral by hulle is dit van groot waarde.

Met die vordering wat die afgelope paar jaar gemaak is met antibiotiese-verrykte, selektiewe media en ‚Millipore'-filtrasietegniek, lewer die isolasie van *V. fetus* nie meer die groot probleem van vroeër op nie. Ten einde te verseker dat 'n betroubare diagnose gemaak word, moet die monsters die laboratorium so spoedig moontlik bereik nadat hulle geneem is.

(a) Monstering by Bulle:

V. fetus-organismes word in die skede en op die *glans penis* aangetref. 'n Direkte of indirekte metode van diagnose kan gevolg word.

(i) Direkte diagnose:

'n Direkte diagnose word gemaak deur die direkte isolasie van *V. fetus* uit die semen of uit 'n skedewassing. Dertig ml voedingsop met pH 7.0 word as skedewasvloeistof gebruik. Die lewensvatbaarheid van *V. fetus* in 0.85% soutoplossing neem vinnig af, sodat dit nie geskik is as wasvloeistof nie. As meer as 30 ml gebruik word, is die konsentrasie van organismes dikwels te laag vir 'n betroubare diagnose.

'n Wasvloeistofhouer word gemaak deur 'n gomlastiek buis 0.5 cm x 1 meter aan 'n bottel, gevul met 30 ml voedingsop, te heg. Die punt van die buis word toegeknyp en die bottel dan gesteriliseer. Net voor gebruik word 'n steriele plastiese pipet by die toegeknypte punt ingedruk.

Knip die hare om die skede opening af en was met skoon water. Droog die area goed af. Stoot nou die pipet 10 cm in die skede op. Hou die skede-opening met een hand toe. Lig die bottel nou op, laat die wasvloeistof in die skede vloei en masseer dit met die ander hand uitwendig vir een minuut. Laat nou die bottel sak sodat die vloeistof daarin terugvloei. Vul nou 'n 20 ml steriele buis met die skedewassing en verseël dit lugvry.

(ii) Indirekte diagnose:

'n Ejakulaat, tesame met die besinksel van 'n skedewassing wat vir 30 minute teen 3,000 o.p.m. uitgeswaai is, word in die baarmoeder van bronstige negatiewe verse ge-

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spuit. Monsters word dan van die verse geneem vir diagnose.

(b) Monstering by verse en koeie:

V. fetus kan binne drie dae na 'n natuurlike besmetting in die vagina gevind word en kan vir maande daar teenwoordig bly, selfs in dragtige diere. Kort na besmetting sprei die infeksie van die vagina na die baarmoeder, vanwaar dit weer binne 40-60 dae begin verdwyn.

By die indirekte toets van bulle word vier biopsies, met weeklikse tussenposes, deur middel van 'n Folmer-Nielson-apparaat van die baarmoederslymvlies van gedekte verse geneem.

Vaginale slymmonsters kan met plastiese pipette opgesuig word. As die pipet ingestoot word, moet die vulva deur 'n handlanger oopgetrek word om te voorkom dat ander, uitwendig teenwoordige bakterieë die slym besoedel.

Waar aborsies voorkom, kan fetale maaginhoud in steriele plastiese pipette opgesuig word. Pipette word in alle gevalle met 'n warm yster lugvry verseël. Smere van maaginhoud en kotiledons kan ook gemaak word.

(c) Die versending van monsters:

V. fetus is mikro-aërofilies en dus baie gevoelig vir lug. Waar monsters binne ses uur die laboratorium kan bereik, moet dit net lugvry verseël word voor versending. Waar dit langer gaan neem om 'n laboratorium te bereik, moet dit lugvry verseël en in droë ys verpak word. Indien die versendingstyd langer as 24 uur is, sal slegs 'n klein aantal monsters positiewe resultate lewer. Versending per pos is dus uiters ondoeltreffend.

(d) Kweking van die organismes:

Weens sy mikro-aërofiliese aard, moet *V. fetus* onder 'n verlaagde O_2 spanning by 37°C gekweek word. Petrivlakkulture kan in 'n wye-bekfles gepak word. Deur 'n kers daarin te laat uitbrand nadat die deksel toegemaak is, kan die verlangde lugsamestelling verkry word. Waar apparaat beskikbaar is, kan 80% van die lug in die bottel met N_2 of CO_2 vervang word. Halfvloeibare media kan net so in die broeikas geplaas word.

(e) Media: Of Brucella-agar[D] óf Thiol medium[D] + 15 gram agar/liter met en sonder antibiotika word vir isolasie gebruik.

Halfvloeibare media wat gebruik kan word, is Thiolmedium[D] of Brucella-sop[D] met byvoeging van 1.5 gram agar per liter. Dit word in ongeveer halfvol sopbuise gehou. *V. fetus* kan tot twee maande in sulke buise by kamertemperatuur lewend gehou word.

Antibiotika moet bygevoeg word teen 45°-50° C, net voordat petrivlakke gegooi word. Maak die volgende oplossings: 1.) Bacitracin[S] 30 mg/ml; 2) Novobiocin[P] 5 mg/ml; en 3) Mycostatin[L] 50,000 E/ml. By elke liter media word 1.25 ml van elke oplossing gevoeg en dit word geskud, waarna die petrivlakke gegooi word. Die antibiotiese oplossings kan tot drie maande by 4° C gehou word.

(f) Enting: Ongeveer 0.1 ml vaginale slym, biopsiemateriaal, semen of fetale maaginhoud word op petrivlakke met en sonder antibiotika geënt en uitgestryk. Skedewassings word op petrivlakke met en sonder antibiotika en in halfvloeibare media geënt. Om van die grofste onsuiwerhede in die skedewassings ontslae te raak, word dit vir vyf minute teen 1,500 o.p.m. uitgeswaai. Die helder vloeistof word nou vir 25 minute teen 3,000 o.p.m. uitgeswaai en die boonste vloeistof afgesuig. Met die laaste drie ml vloeistof in die buis word die besinksel losgespoel en in 'n spuit opgetrek. Om van die vinniggroeiende *Proteus* en *Pseudomonas* spp., wat dikwels teenwoordig is, ontslae te raak, word 'n 0.65 μ ,Millipore-filter** in 'n filterhouer aan die spuit geheg. Laat 3-5 druppels van die laaste helfte van die filtraat op die medium drup.

Waar min organismes teenwoordig is, mag dié tegniek 'n negatiewe resultaat gee. Die voordeel is dat 'n redelike suiwer kultuur van *V. fetus* meesal verkry kan word

(g) Ondersoek: Kulture word na drie en ses dae ondersoek. Half-vloeibare kulture moet tot veertien dae gehou word voordat 'n negatiewe diagnose gemaak kan word. 'n Positiewe groei in 'n halfvloeibare medium ver-

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D: Difco Laboratories, Detroit 1, Michigan, V.S.A.
S: Squibb. Lab., Isando, Transvaal, S.A.
P: Propan Pharmaceutical, Wadeville, Germiston, Transvaal.
L: Lilly Laboratories, Isando, Transvaal.
** Millipore Filter Corporation, Bedford, Mass. V.S.A.

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toon as 'n sambreelvormige dofheid ongeveer 1 mm onder die oppervlakte. Kolonies op vaste media is deurskynend, opgehewe en glinsterend, tot 1 mm in deursnee maar dikwels kleiner. Soms word net 'n egalige dofheid op die kweek bodem gesien.

Van die verdagte groei word smere gemaak en volgens metode van Gram gekleur. *V. fetus* vertoon as gram-negatiewe, komavormige organismes met enkele "S"- en nou en dan langer spiraal vorms by primêre isolasie.

(h) Differensiasie van *Vibrio* spp: Morfologies kan die drie belangrikste tipes, nl. *V. fetus venerealis, V. fetus intestinalis* en *V. bubulus* nie onderskei word nie en biochemiese toetse word vir die doel gebruik. Die basiese medium wat hiervoor gebruik word is Bactothiol [D]-medium in sopbuise.

(i) Katalase-toets: By 'n drie-dae oue kultuur van 5 ml word 5 ml van 'n 1% H_2O_2-oplossing gevoeg. Die watteprop word vervang met 'n rubberprop, waar deur 'n haarbuisie gaan. Die buis word onderstebo gehou en die verplasing van medium deur gas word gemeet. Die verplasing van 5 mm of meer, binne 20 minute, word as positief beskou.

(ii) H_2S-toets 0,02% sistien word by die basiese medium gevoeg. Nadat die buise geënt is, word 'n loodasetaat-geïpregneerde filtreerpapierstrokie in die bek van die buis geplaas en die watteprop opgesit. Swartverkleuring van die papier binne drie dae by 37° C word as positief beskou.

(iii) Glisien-toets: Groei na bebroeiing teen 37°C na drie dae, in 'n 1% glisienbyvoeging by die medium, word as positief beskou.

(iv) Sout-toets: Slegs *V. bubulus* sal groei in basiese medium waarby 3.5% NaCl gevoeg is.

Onderskeid tussen die drie bogenoemde *Vibrio* spp. word gemaak volgens die onderstaande tabel:

	V. fetus venerealis	*V. fetus intestinalis*	*V. bubulus*
Katalase	+	+	—
3.5% NaCl	—	—	—
H_2S	—	±	+
1% glisien	—	+	+

OPSOMMING

1. As kuddetoets is die vaginale slymagglutinasietoets betroubaar, maar heeltemal ongeskik om individuele diere te toets. Minstens tien monsters per kudde is nodig om 'n betroubare resultaat te verkry.

2. 'n Bakteriologiese diagnose kan goeie resultate lewer van individuele diere mits aan die volgende vereistes voldoen word: (a) Vermy kontaminasie van die monsters; (b) Gebruik voedingsop as skedewasvloeistof; (c) Besorg monsters binne ses uur by die laboratorium; indien dit langer sal neem, gebruik droë ys as bewaarmiddel en verseël altyd die monsters lugvry; (d) Bepaal die sp. of variëteit wat geïsoleer is, sodat duur behandelingsmetodes nie voorgeskryf sal word vir die saprofitiese *V. bubulus* nie; (e) Indien al bogenoemde vereistes nie nagekom word nie, is 'n negatiewe resultaat van geringe waarde; (f) Diagnose by streekslaboratoriums kan in dié verband 'n belangrike rol speel deurdat die tydsfaktor uitgeskakel kan word.

DANKBETUIGING

Die Hoof, Navorsingsinstituut vir Veeartsenykunde, word bedank vir sy toestemming tot publikasie en dr. H. J. W. Botes vir sy advies by die opstelling hiervan.

VERWYSINGS

1. Vandeplassche M., Florent A., Bouters R., Hysman A., Brone E. & Dekeyser P. 1963 *The Pathogenesis, Epidermiology and Treatment of Vibrio fetus infection in cattle.* Brussel. Instituut tot Aanmoediging van het Wetenschappelijk Onderzoek in Nijverheid & Landbouw.
2. Laing J. A. 1960 *Vibrio fetus infection of Cattle.* Rome. FAO Agricultural Studies.
3. Dunn H. O., Burda K., Wagner W. C. & Gilman H. L. 1965 *Cornell Vet.* 55:220
4. Hoerlein A. B. & Kramer T. 1963 *J. Am. vet. med. Ass.* 14:866
5. Shepler V. M., Plumer G. J. & Faber J. E. 1963 *Am. J. vet. Res.* 24:749
6. Winkenwerder W. & Bisping W. 1964 *Zentbl. Vet. Med.* Reihe B:603
7. Clark B. L. 1965 *Victorian Vet. Proc.* 1965-66:32

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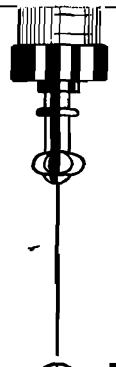

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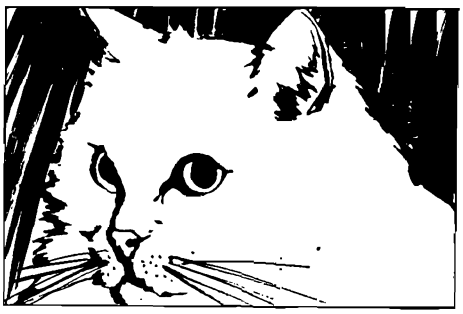

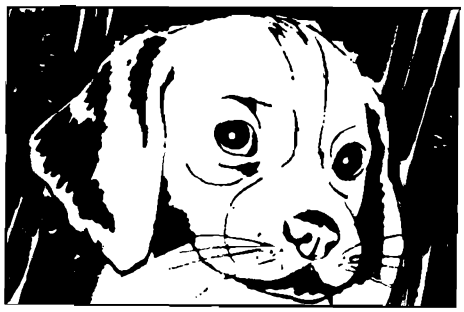

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ISOLATION OF *ACTINOBACILLUS SEMINIS* IN SOUTH AFRICA

R. W. WORTHINGTON* AND P. P. BOSMAN**

SUMMARY

A case of epididymitis in a Dorper ram is described. *A. seminis* was isolated from the ram's semen. The organism proved to be identical in serological, cultural and biochemical characteristics to a strain of *A. seminis* obtained from Australia. The ram excreted the organism in the semen for nine months and *A. seminis* was cultured from the accessory glands after slaughter. Epididymitis was produced in six out of eight rams experimentally infected with the South African isolate and the Australian strain, three of these six rams also developed an orchitis. The lesions were similar in those infected with the South African and Australian strains. Seven of the eight infected rams excreted the organism in their semen and all developed titres against *A. seminis* antigen.

INTRODUCTION

Since the first isolation in South Africa of *Brucella ovis* from rams with epididymitis [1] this organism has generally been regarded as the main cause of infectious epididymitis. Vaccination with the attenuated *Brucella melitensis* strain Rev. 1 introduced by van Drimmelen in 1960 [2] generally proved to be effective as a means of controlling the infection. In isolated cases, however, epididymitis continued to occur in flocks in which rams had been vaccinated. Epididymitis was also occasionally seen in rams which did not show titres against *Br. ovis* and from whose semen it was not possible to isolate this organism. It was therefore felt that other organisms might be involved in these cases.

In 1955 Dodd & Hartley[3] working in New Zealand isolated an organism from cases of epididymitis, which was provisionally described as a gram negative pleomorph. Baynes & Simmons (1960) [4] later isolated an organism which they named *Actinobacillus seminis* from three cases of epididymitis in two flocks in Queensland and New South Wales. They were able to cause lesions by inoculating cultures of the organism or infected semen into the testis or the epididymis of rams and developed a complement fixation test for detecting antibodies. In 1966 Simmons, Baynes & Ludford [5] described their observations on the epidemiology of the condition in an infected flock of Border Leicester sheep. The condition has also been found in America [6]. It is of interest to note that Baynes & Simmons (1960) state that "it is unlikely that the disease would be readily distinguished from *Brucella ovis* infection by clinical examination", and Livingston & Hardy (1964) [6] state: "It is possible that *A. seminis* infections have been confused with infections caused by *Br. ovis*."

The purpose of this report is to describe the isolation of *A. seminis* from a ram with epididymitis in South Africa.

CASE HISTORY

A batch of Dorper rams was acquired for experimental work from a farm in the Boshoff district. On arrival all rams were bled and semen was collected from them. Serum from one ram (No. 22824) was found to react negatively to the C. F. test for *Br. ovis* but examination of semen revealed large numbers of neutrophils.

The volume and concentration of the semen was in the normal range, and smears stained with nigrosin-eosin [7] showed 60% of sperm to be alive.

Clinical examination at this stage revealed no lesion. A fortnight later the ram developed clinical epididymitis with unilateral enlargement and hardening of the cauda epididymidis. Palpation elicited no pain response.

Within three days of the development of the epididymal lesion, a unilateral orchidectomy was performed and the diseased testis removed *in toto*. A typical spermatic granuloma, as described by Jubb and Kennedy, [8]

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with a diameter of 2 cm was found on incision of the cauda epididymidis. There was also obvious macroscopic fibrosis of this epididymal tail.

Semen collected at an interval of two and again at three weeks after the operation showed no improvement compared to preoperative semen. No palpable lesion developed in the remaining epididymis until five months later, when hardening of the cauda epididymidis could be felt. The quality of semen at this stage was still unchanged. The fact that semen could still be obtained, indicated that the epididymis was still patent, in spite of the lesion.

Bacteriological examination of the semen resulted in the isolation of an organism resembling *A. seminis*.

MATERIALS AND METHODS

Isolation of the organism:

Semen was streaked on tryptose blood agar and serum agar plates which were incubated at 37°C in air and in a 15 percent CO_2 atmosphere. Typical small dewdrop-like colonies were selected and subcultured on tryptose blood agar plates.

Semen samples were examined periodically until the ram was slaughtered nine months after the first isolation was made. At autopsy cultures were made from the following organs: lung, liver, spleen, kidney, iliac and inguinal lymph nodes, testicle, epididymis, vesiculi seminales, bulbourethral glands and ampullae.

Cultural and Biochemical tests:

A strain of *A. seminis* K3844-C was obtained from Mr. G. C. Simmons, of the Animal Research Institute, Yeerongpilly, Queensland, Australia, and his strain and the South African isolate 22824 were tested in parallel as follows:-

Tenfold serial dilutions of a suspension of organisms were plated on tryptose blood agar plates and incubated under 15 percent CO_2. The plates were observed daily and the colony forms studied on plates having well spaced single colonies. Smears stained according to the Gram, Hansen and modified acid fast techniques were examined. The growth in air and in a 15 percent CO_2 atmosphere on serum agar, tryptose blood agar, agar, serum broth and broth was observed. The strains were tested for ability to ferment the following carbohydrates: arabinose, rhamnose, glucose (anaerobically and aerobically), fructose, sucrose, maltose, lactose, trehalose, starch, glycerol, mannitol (aerobically and anaerobically), dulcitol, sorbitol, inulin and dextrin; and for ability to reduce nitrates to nitrite, split urea and sodium citrate, produce indole, liquefy inspissated horse and bovine serum, hydrolyse arginine and to produce catalase.

Serological investigation:

(a) Rabbit antisera

The growth from one well-grown tryptose blood agar slope after 48 hours' incubation was harvested in 10 ml of sterile normal saline and to this was added 9 ml of Bayol F* and 1 ml of Arlacel A**. The suspension was shaken until a stable emulsion was produced. Three ml of the emulsion was injected into rabbits each week for three weeks. After the last injection they were bled weekly until a high titre serum was obtained (usually after 1-2 weeks) and then a large amount of blood was collected by heart puncture. The serum was freeze-dried in 1 ml amounts and stored at 4° C. Antisera were produced in this manner against the strains 22824 and K3844-C as well as the following three *Br. ovis* strains: 6010 (Onderstepoort isolate), 292 (received from Dr. Buddle, Wallaceville Research Station, New Zealand), Q 28 E (received from Mr. Simmons, Animal Research Institute, Yeerongpilly, Queensland, Australia).

(b) *Antigens*

Strain 22824 was grown on tryptose blood agar medium in Roux flasks. After 72 hours' incubation the flasks were individually checked for sterility, the organisms washed off the slopes with sterile normal saline and the suspension killed by heating to 60° C for one hour. The cells were washed twice in 0.5% phenol saline suspended in a small amount of phenol saline and placed in a boiling water bath for 10 minutes. Merthiolate was added to give a final concentration of 1/10,000 and the suspension allowed to stand at room temperature for three weeks. It was then centrifuged to deposit the cells, the clear supernatant was stored at 4° C and used as antigen. In the case of strain K3844-C and 6010 the organisms were grown in a fluid medium[9] in an atmosphere of 15 percent CO_2.

* Esso Petroleum Co.

** Mannide mono-oleate; Atlas Power Co., Wilmington. Delaware.

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The Roux flasks were shaken on a shaking machine during incubation. A soluble antigen was produced from the harvested cells in exactly the same manner as described for strain 22824.

The method of performing complement fixation tests was that used routinely at Onderstepoort for testing sera for anti-*Br. ovis* antibodies. Details may be obtained from the authors. Each of the five rabbit anti-sera were tested against the three antigens. The three antigens were also used for testing the sera of ram 22824 and the experimentally infected rams.

Transmission experiments: A group of 8 adult Dorper rams were used. All rams were bled and their sera tested for antibodies against 6010 *(Br. ovis)*, 22824 and K3844-C antigens: Semen smears stained with Gram and modified acid fast staining methods were examined and semen cultures made from semen collected from each ram. There was no evidence of infection with either *Br. ovis* or *A. seminis* in the eight rams. Cultures of strain 22824 and K3844-C grown for 24 hours on tryptose blood agar were harvested in normal saline. The suspensions were adjusted to a density of a Brown's tube No. 4. One ml suspension of strain 22824 was injected into the testis of each of two rams and two received 1 ml into the tail of the epididymis. The other group was similarly challenged with K3844-C and the two groups housed in separate pens. The rams were bled at weekly intervals for six weeks after the exposure and their sera tested against 6010 *(Br. ovis)*, 22824 and K3844-C antigens. Semen specimens were also collected at weekly intervals for six weeks and examined culturally. Strains isolated were identified by the same cultural and biochemical tests used to identify the original strain.

RESULTS

The South African isolate proved to be culturally and morphologically identical to the Australian strain. Neither strain would grow to any appreciable extent on medium not containing serum and both strains grew better in an atmosphere containing CO_2 than in air, although growth did occur in air. Growth was distinctly better on tryptose blood agar than on blood or serum agar without tryptose. The colony forms of the two strains were identical and similar to those described by Baynes & Simmons[4]. The organisms were non-motile, non-sporulating gram negative pleomorphs with individual organisms varying from coccoid forms to rods of up to 4 μ in length. They stained negatively with Hansen's and the modified acid fast straining techniques. No sugars were fermented even after prolonged incubation (4 weeks). Neither strain was able to produce indol, split urea or sodium citrate or lyse inspissated bovine and equine serum. They were able to reduce nitrate to nitrite, hydrolyse arginine, and were strongly catalase-positive. The only difference between the two strains was that the South African strain produced a clot but no acid in litmus milk whereas the Australian strain produced neither clot nor acid.

The results of the complement fixation tests done on the rabbit antisera are given in Table 1.

TABLE 1: COMPLEMENT FIXATION TITRES OF RABBIT ANTISERA (RECIPROCALS OF THE END TITRE)

	Antigen		
	22824 (A. seminis)	K3844-C (A. seminis)	6010 (Br. ovis)
22824 (A. seminis)	64	64	—
K3844-C (A. seminis)	128	128	—
292 (Br. ovis)	—	—	512
Q28E (Br. ovis)	—	—	64
6010 (Br. ovis)	—	—	256

Of the rams injected with strain K3844-C three developed lesions within three days. The ram 22997, which had received an intratesticular injection, did not develop any lesions and remained clinically normal for the duration of the experiment. Its counterpart developed an acute orchitis and epididymitis which subsided within 14 days and thereafter remained clinically normal. The two rams which had received an intra-epididymal injection both developed acute epididymitis within three days and by the seventh day 22998 presented a hyperacute orchitis; the testicle was about twice the normal size and the whole scrotum was hot, oedematous and painful. The swelling gradually subsided and the testicle became hard, although it remained slightly enlarged; the epididymis remained very enlarged and hard. The other ram (22985) developed an acute epididymitis

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which gradually became hard and less painful, although remaining very large and appearing like a typical field case of chronic epididymitis. One of the rams (22984) which had received an intratesticular injection of strain 22824, failed to develop any lesions and remained clinically normal throughout the trial. The other three all developed acute epididymitis within three days. Intratesticular injection into ram 22996 also produced an acute orchitis, the testicle being slightly swollen, hot and painful and both head and tail of the epididymis remained hard and swollen. Within three weeks the testicle had atrophied and was hard and about half the size of the other, normal, testicle. Both head and tail of the epididymis remained hard and swollen. The other two rams which had receved an injection into the epididymis developed acute inflammation of the tail of the epididymis within three days, thereafter the swollen epididymis became progressively harder and the picture was similar to that seen in a typical field case of chronic epididymitis.

All the rams subjected to immunity challenge developed CF titres against *A. seminis* antigen but failed to develop titre against *Br. ovis* antigen. The results are given in in Table 2. For the sake of brevity only the titres before infection and 1, 3 and 6 weeks after injection are given.

Organisms indentical to *A. seminis* in cultural and biochemical characteristics were isolated from the semen of seven of the eight rams. The results of the cultural examinations are given in Table 3.

TABLE 2: COMPLEMENT FIXATION TITRES OF SHEEP INFECTED WITH **A. SEMINIS** (RECIPROCALS OF TITRES)

Ram No.	Infecting strain	Before infection			PERIOD AFTER INFECTION								
					1 week			3 weeks			6 weeks		
		6010 (Br. ovis)	22824 (A. seminis)	K3844-C (A. seminis)	6010 (Br. ovis)	22824 (A. seminis)	K3844-C (A. seminis)	6010 (Br. ovis)	22824 (A. seminis)	K3844-C (A. seminis)	6010 (Br. ovis)	22824 (A. seminis)	K3844-C (A. seminis)
22985	K3844-C	—	—	—	—	8	16	—	16	16	—	8	16
22998	,,	—	—	—	—	4	4	—	64	64	—	64	64
22994	,,	—	—	—	—	64	32	—	8	8	—	8	8
22997	,,	—	—	—	—	8	8	AC	AC	AC	—	16	8
22999	22824	—	—	—	—	32	16	2	64	64	—	64	64
22989	,,	—	—	—	—	64	32	—	64	64	—	64	64
22996	,,	—	—	—	—	8	16	—	64	64	—	64	64
22984	,,	—	—	—	—	16	32	—	32	32	4	64	32

AC = anticomplementary

TABLE 3: ISOLATION OF **A. SEMINIS** FROM THE SEMEN OF 8 EXPERIMENTALLY INFECTED RAMS

Ram No.	Infection strain	Cultural examination of semen						
		Before exposure	Period after infection:					
			1 week	2 weeks	3 weeks	4 weeks	5 weeks	6 weeks
22985	K3844-C	—	−	−	−	−	+	+
22998	,,	—	−	+	+	+	+	+
22994	,,	—	+	+	+	+	+	+
22997	,,	—	−	−	−	−	−	−
22999	22824	—	−	+	+	−	+	+
22989	,,	—	+	+	+	+	+	+
22996	,,	—	−	+	−	+	+	−
22984	,,	—	−	+	−	+	+	−

−: negative. +: positive.

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The post mortem examination of ram 22824 revealed an enlarged fibrotic epididymis and what appeared to be enlarged *vesiculi seminales*. No macroscopic abnormalities were seen in any of the other organs. A more detailed description of the pathology will be published later. *A. seminis* was isolated in pure culture from the glandular part of the deferent ducts and the *vesiculi seminales*; a few colonies were also isolated from the bulbourethral glands. No organisms were isolated from the testicle, epididymis or any of the other organs.

DISCUSSION

A three year investigation of *A. seminis* infection in a flock of border Leicester sheep in Queensland by Simmons et al [5] revealed 31 infected rams. They considered the condition to be a primary genital infection of rams and described clinical and subclinical cases. *A. seminis* could be isolated regularly from clinical cases for periods varying from three months to four and a half years. The high incidence of the disease in unmated rams suggested a method of spread other than by venereal transmission. In the case described in this article and in some of the cases described by Simmons et al [5], the organism was found in the accessory glands and was excreted in the semen for long periods.

In the Australian investigation complement fixation titres were also demonstrated in 23 ewes and two newborn lambs but no clinical or bacteriological evidence of infection could be found and the serological response was of a short duration.

It is unlikely that *A. seminis* infection was recently introduced into South Africa. The case described in this communication occurred in a Dorper ram, a breed of sheep developed in this country. The condition probably previously went unnoticed because of the high incidence of *Br. ovis* infection before the introduction of REv. 1 vaccination. Epididymitis lesions caused by *A. seminis* and *Br. ovis* are very similar and the two conditions can only be distinguished by laboratory examination.

A seminis is serologically unlike *Br.. ovis* and usually morphologically dissimilar, but in some cultures, where predominantly short cocco-bacillary forms occur, these could be mistaken for *Br. ovis* in Gram stained smears. The colony forms may also quite closely resemble *Br. ovis*. If, however, a careful examination is made, there should be no difficulty in distinguishing the two organisms.

At the present time nothing is known of the incidence or economic importance of this condition in South Africa. Only isolated cases have been described in other countries and despite the fact that extensive testing was undertaken in Queensland flocks, only a few positive samples were revealed; the disease was a problem in one flock only [5]. In South Africa a similar organism has also recently been isolated in the Middelburg area [10] and the infection could perhaps be sufficiently widespread to constitute a problem. More information is needed about the incidence of the organism in the semen of rams with excessive numbers of neutrophils in their semen, in rams showing other abnormalities of testes or accessory glands and apparently normal rams.

ACKNOWLEDGEMENT

The Chief, Veterinary Research Institute, Onderstepoort and the Chief, Division of Veterinary Services are thanked for their permission to publish this article.

REFERENCES

1. Van Rensburg S. W. J., Van Heerden K. M., Le Roux D. J. & Snijders A. J. 1958 *Jl. S.Afr. vet. med. Ass.* 29:223
2. Van Drimmelen G. C. 1960 *Jl S. Afr. vet. med. Ass.* 31:129
3. Dodd D. C. & Hartley W. J. 1955 *N.Z. vet. J.* 3:105
4. Baynes I. D. & Simmons G. C. 1960 *Aust. vet J.* 36:454
5. Simmons G. C., Baynes I. D. & Ludford G. C. 1966 *Aust. vet. J.* 42:183
6. Livingston C. W. & Hardy W. T. 1964 *Am. J. vet. Res.* 25:660
7. Blom E. 1950 *Nord. vetmed.* 2:58
8. Jubb K.V.F. & Kennedy P. C. 1963 *Pathology of domestic animals* Vol. I p. 370 New York Academic Press.
9. Lawrence W. E. & Jones R. F. 1961 *Aust. vet J.* 37: 406.
10. Van Tonder E. M. & Bolton T. F. W. 1968 This Journal.

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Jl S. Afr. vet. med. Ass. 39 (2): 87-90 1968

EPIDIDYMITIS IN RAMS CAUSED BY *ACTINOBACILLUS SEMINIS*

E. M. VAN TONDER AND T. F. W. BOLTON*

SUMMARY.

An organism resembling *Actinobacillus seminis* was isolated from the semen of 21 rams on seven properties. *A. seminis* was originally described by Baynes and Simmons [1]. Intra-epididymal and intratesticular transmission of Simmons' organism and one of our isolates to eight susceptible rams and clinical, bacteriological and serological tests confirmed beyond doubt that this organism was indentical to *A. seminis.*

INTRODUCTION

Epididymitis in rams caused by a pleomorphic gram negative, non acid-fast bacillus was first described by aynes and Simmons [1] in Australia. In view of its general characteristics and isolation from semen, the name *Actinobacillus seminis* was provisionally proposed for this organism. Simmons, Baynes and Ludford [2] reported a high incidence of infection by the same organism in a flock of Border Leicester sheep. Infection caused by *A. seminis* has also been reported by Livingston and Hardy [3] in a single case in a Rambouilliet ram in the United States of America.

The high incidence of epididymitis amongst stud and flock rams in the Middelburg, Cape area on farms where the application of Rev 1 vaccine is regularly practised, led to the suspicion of a cause other than *Brucella ovis.* During the past year investigations were carried out on a number of properties on rams showing clinical signs of epididymitis. Bacteriological examination of semen samples from 21 rams on seven properties yielded an organism similar to that described by Baynes and Simmons [1] as regards its cultural and biochemical properties.

This paper describes the transmission experiments with cultures of the organism isolated from one of these rams. Comparative studies with Simmons' organism are also described.

MATERIALS AND METHODS

Experimental animals

Ten two-tooth Merino rams were kept together under isolation for a period of 14 days prior to the onset and during the entire experiment. They were subjected to a clinical examination, bacteriological semen examination and serum-complement fixation test for *A. seminis* on two occasions during the pre-inoculation period. A complement fixation test for *Brucella ovis* was carried out once during the pre-inoculation period and again 12 days after inoculation.

Infective material.

Subcultures were made on 5% horse blood agar slants of the organism obtained from an infected ram (No. 87) as well as of Simmons' organism. These cultures were examined after four days for possible contamination and suspended in normal saline. The suspensions of each organism were pooled and standardised against Brown's opacity tube No. 4.

Transmission to rams.

Rams 70 and 23 received injections into the tail of the left epididymis and rams 2 and 3 into the left testicle of 1 ml of suspension of the organism isolated from ram 87. Likewise 1 ml of suspensions of Simmons' organism were injected into the epididymis of rams 7 and 24 and into the testes of rams 47 and 53. Rams 57 and 88 were left as controls to check contact transmission.

Specific examinations

Clinical examination of the external genitalia and rectal temperatures were recorded daily for 14 days after injection. Microscopic and cultural examination of semen, and serum complement fixation tests were carried out at given intervals. The results obtained are recorded here.

Serological tests for *A. seminis*:-

* Veterinary Diagnostic Centre, Middelburg, Cape.

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TABLE 1: SCROTUM EXAMINATIONS ON SPECIFIC DAYS DURING POST-INOCULATION PERIOD

Ram No.	Inoculum	Inoculation site	DAYS AFTER INOCULATION							
			0	1	3	5	7	9	11	14
70	87	L.E.	N.A.D.	L.E.*	L.E.**	L.E.*** L.T.*	L.E.**** L.T.**	L.E.**** L.T.**	L.E.**** L.T.**	L.E.**** L.T.*
23	87	L.E.	N.A.D.	L.E.*	L.E.*** L.T.*	L.E.*** L.T.**	L.E.**** L.T.**	L.E.**** L.T.**	L.E.**** L.T.**	L.E.*** L.T.*
2	87	L.T.	N.A.D.	N.A.D.	N.A.D.	N.A.D.	N.A.D.	N.A.D.	N.A.D.	N.A.D.
3	87	L.T.	N.A.D.	N.A.D.	L.T.*	L.T.***	L.E.* L.T.***	L.E.** L.T.***	L.E.** L.T.***	L.E.** L.T.***
7	Simmons	L.E.	N.A.D.	L.E.*	L.E.**	L.E.*** L.T.*	L.E.**** L.T.**	L.E.**** L.T.**	L.E.**** L.T.**	L.E.**** L.T.**
24	Simmons	L.E.	N.A.D.	L.E.*	L.E.**	L.E.**	L.E.*** L.T.*	L.E.*** L.T.*	L.E.*** L.T.*	L.E.*** L.T.*
47	Simmons	L.T.	N.A.D.	N.A.D.	L.T.*	L.T.**	L.T.***	L.E.* L.T.***	L.E.* L.T.***	L.E.* L.T.*
53	Simmons	L.T.	N.A.D.	N.A.D.	L.T.*	L.T.*	L.T.**	L.T.***	L.T.***	L.T.**
57	Control	—	N.A.D.	N.A.D.	N.A.D.	N.A.D.	N.A.D.	N.A.D.	N.A.D.	N.A.D.
88	Control	—	N.A.D.	N.A.D.	N.A.D.	N.A.D.	N.A.D.	N.A.D.	N.A.D.	N.A.D.

L.E. = Left Epididymis
L.T. = Left Testes
* = Degrees of enlargement
N.A.D. = No abnormality detected

Specific antigen and positive serum was obtained from the Veterinary Research Institute, Onderstepoort. Serum from a six month old ewe lamb reared in absolute isolation was used as the negative control. A serum dilution of 1/20 with 50% or greater complement fixation was considered as positive.

RESULTS

The experimental animals reacted negatively to all examinations and tests carried out during the pre-inoculation period. The results of scrotum examinations on specific days following inoculation are given in Table 1.

Enlargement occurred sooner in the rams infected intra-epididymally than in those infected intra-testicularly. All rams, except No. 2, which never reached clinically, as well as the two control rams, reacted by the third day. The lesions attained their maximum size by the seventh to the ninth day. All rams infected intra-epididymally also had testicular enlargement on the fifth to the seventh day, whereas two out of three rams infected intra-testicularly developed enlargement of the epididymis from the seventh to the ninth day. The enlarged testes and epididymi tended to be soft and tender at the initial stages but became more firm and insentitive by the 14th day. Pain was more evident in those rams infected intra-testicularly.

TABLE 2: RESULTS OF EXAMINATIONS OF SEMEN SMEARS STAINED WITH THE MODIFIED ZIEHL NIELSEN TECHNIQUE

Ram No.	Inoculum	Inoculation Site	DAYS AFTER INOCULATION			
			4	7	11	14
70	87	L.E.	N B	N B	N —	N B
23	87	L.E.	N B	N —	N B	N —
2	87	L.T.	—	—	—	—
3	87	L.T.	N —	N B	N —	N B
7	Simmons	L.E.	N —	N —	N —	N —
24	Simmons	L.E.	N —	N B	N B	N B
47	Simmons	L.T.	—	—	—	—
53	Simmons	L.T.	N —	N —	N —	N —
57	Control	—	—	—	—	—
88	Control	—	—	—	—	—

N = Neutrophils
B = Non-acid fast bacilli
L.E. = Left Epididymus
L.T. = Left Testis

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Temperature recordings were variable and the fact that reasonably high reactions were obtained on the third to the sixth and again about the ninth day in all sheep, suggests that environmental temperatures and conditions were probably responsible.

The results of the semen smear examination are summarised in Table 2. In six of the infected rams neutrophils were detected in the semen but non-acid fast bacilli in only four of them at one stage or another.

Bacteriological examinations

The bacteriological results are given in Table 3. Organisms resembling Simmons' organism culturally and biochemically were isolated from the semen of all artificially infected rams except ram no. 2 on one or more occasions.

TABLE 3: RESULTS OF BACTERIOLOGICAL SEMEN EXAMINATIONS

Ram No.	Days after inoculation			
	4	7	11	14
70	+	–	N.T.	+
23	+	+	+	–
2	–	–	–	–
3	–	–	+	+
7	–	–*	+	–
24	–	+	+	+
47	–	+	–	N.T.
53	–	+	N.T.	N.T.
57	–	–	N.T.	–
88	–	–	N.T.	–

+ = **Actinobacillus seminis** isolated
– = **Actinobacillus seminis** not isolated
* = Organism died
N.T. = Not tested

Serological examinations.

All sera from the rams used in this experiment reacted negatively to the complement fixation test for *Brucella ovis* on both occasions.

Table 4 summarises the results of the complement fixation test for *A. seminis* on serum obtained from these rams on specific days after inoculation.

TABLE 4: RESULTS OF THE COMPLEMENT FIXATION TEST FOR **A. SEMINIS**

Serum	Inoculum	Days after inoculation			
		4	7	11	14
Positive control	—	160	160	160	160
Negative control	—	0	0	0	0
Ram No. 70	87	0	40	80	80
,, 23	87	0	40	40	40
,, 2	87	0	10	0	10
,, 3	87	0	10	40	40
,, 7	Simmons	10	10	20	20
,, 24	Simmons	0	20	40	80
,, 47	Simmons	0	40	80	80
,, 53	Simmons	0	40	80	80
,, 57	Control	0	0	0	0
,, 88	Control	0	0	0	0

DISCUSSION

Clinical, bacteriological and serological results indicated that all the rams except ram no. 2 receiving suspensions of either organism, became infected. This ram had received the organism isolated from ram 87 intratesticularly.

The results of the scrotum examinations were in accordance with those described by Baynes and Simmons[1] in as far as scrotal enlargement was apparent in all the infected rams, except ram No. 2, from the seventh day and in most rams from the fifth day onwards. The rams used in this experiment, however, were infected either intra-epididymally or intra-testicularly and not by a combination of the two routes. Rams infected by the former route reacted after 24 hours and those by the latter on the third day following inoculation, regardless of inoculum used. Scrotal enlargement was not clearly visible at this stage.

These early reactions were probably due to the high infective doses used in this experiment. The earlier epididymal responses may suggest a higher susceptibility of this structure to these organisms. This could also explain why the spread from epididymis to testis occurred sooner than spread from testis to epididymis which took place in three out of four cases anyway. The possibility of semen accumulation due to epididymal obstruction should also be considered. This

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is further supported by the fact that pain is more evident in the cases of intra-testicular inoculation. The more acute course of infection produced by the organism isolated from ram 87 seems to indicate that this organism may be slightly more virulent than Simmons' organism.

Smear examinations indicated that the appearance of neutrophils in the semen of all clinically affected rams, except ram 47, was a constant feature. The non-acid fast bacilli, however, could not be demonstrated regularly.

On bacteriological culture *A. seminis* was recovered from the semen of all infected rams except ram No. 2 on one or more occasions.

Serological tests were positive in seven out of eight infected rams on the 11th and again on the 14th day of the post-infective period. Five rams reacted positively on the seventh day. Ram No. 2 developed a titre of 1/10 on the seventh and again on the 14th day, despite the fact that all other examinitions were negative. This ram is therefore still regarded as a suspicious reactor.

Three out of four rams (Nos. 70, 23 and 3) artficially infected with our organism reacted positively to all tests and one (No. 2) suspiciously to the serological test only, while all rams (Nos. 7, 24, 47 and 53) inoculated with Simmons' organism reacted positively to all tests. It has therefore been established that the organism isolated from ram 87 is identical to the organism described by Baynes and Simmons[1]. Worthington and Bosman have recorded similar results in their recent comparative study[4].

ACKNOWLEDGEMENTS

We have pleasure in thanking the Chief, Veterinary Field Services, for his permission to publish this paper, the Chief, Veterinary Research Institute, Onderstepoort for supplying the required reagents and the Chief, Agricultural Research Institute, Middelburg, C.P. for the experimental animals made available to us.

REFERENCES

1. Baynes I. D. & Simmons G. C. 1960 *Aust. vet J.* 36: 454
3. Livingston C. W. & Hardy W. T. 1964 *Am.J. vet. Res.* 25: 660
2. Simmons G. C., Baynes, I. D. & Ludford C. G. 1966 *Aust. vet. J.* 42: 183
4. Worthington R. W. & Bosman P. P. 1968 This Journal.

BOOK REVIEW

VETERINARY BACTERIOLOGY AND VIROLOGY

I. A. MERCHANT AND R. A. PACKER, 7TH ED.

The Iowa State University Press, Ames, Iowa, 1967. 752 pages.

This book, written by two expert microbiologists with the assistance of several contributing authors, contains a vast amount of information on the general biology of bacteria, infection, resistance, immunity, the classification of pathogenic micro-organisms and of animal viruses and general aspects of viruses and the diseases caused by them. It is to be understood that, as all these subjects are covered in 752 pages, not much detailed data is given, but the information is presented concisely, systematically and attractively. Veterinary students will find it a useful textbook in their microbiology course and also for easy reference when they encounter an infectious disease during their clinical studies. Practitioners could derive material assistance from the descriptions of the various micro-organisms and viruses and the diseases they cause—especially since the descriptions are given in such a compact form.

The seventh edition is a decided improvement on the sixth which was remarkably popular. It seems a pity that some of the newest microbiological techniques, e.g. the use of fluorescent antibodies, are only mentioned in passing.

B. C. J.

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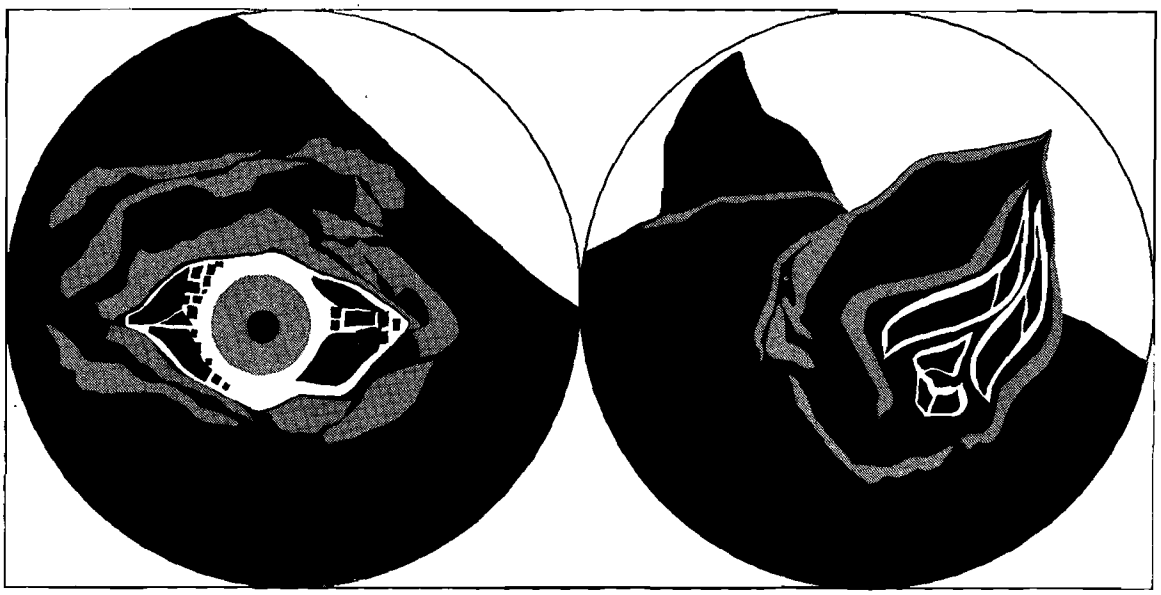

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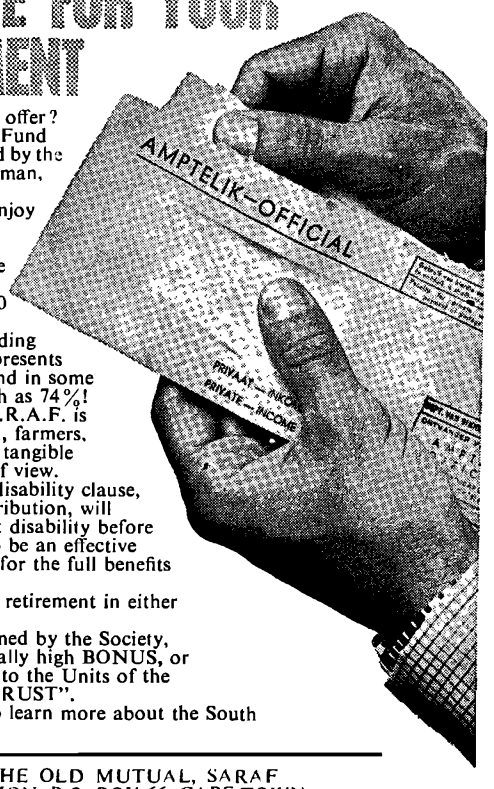

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SURGERY OF BOVINE IMPOTENTIA COEUNDI

V Surgical Conditions of the Preputial Skin and Prolapsed Lining (Excluding Stenosis Proximal to the Orifice)

C. F. B. HOFMEYR*

SUMMARY

This paper concerns surgical conditions of the preputial skin and prolapsed parietal layer, excluding stenosis posterior to the orifice in a series of 27 bulls. Three cases did not fit completely into this classification.

In the other 24 cases the excessive length of the preputial skin and prolapse of the parietal layer were the main predisposing factors in four Herefords, two Afrikaners, three Santa Gertrudis and 13 Brahmans. One Jersey and one Friesland were the exceptions.

Two main operative techniques were developed; one for amputation of a long preputial skin or prolapsed parietal layer and the other one for rectifying orifice stenoses over 1.5 cm thick. Operating on a pendulous sheath caused excessive and prolonged oedema, particularly in the Brahman. Some cases had to undergo more than one operation.

Treatment failed in all three atypical cases. In the remaining 24 there were only two failures, both Brahmans, where secondary stenosis supervened after premature discharge on the insistence of the owners.

INTRODUCTION, DEFINITION AND INCIDENCE

This paper is one of a series dealing with surgical *impotentia coeundi* in 176 bulls [1-4].

The observations recorded are based on a group of 27 bulls suffering from conditions varying from stenosis of the preputial orifice to inflammation, infection and induration of the prolapsed parietal layer of the prepuce. Of these thirteen were Brahmans, five Herefords, three Santa Gertrudis, three Afrikaners, two Frieslands and one Jersey. Their ages varied from 13 months to eight years.

It is noteworthy that *Bos indicus* (Brahman and Afrikaner) and a breed developed from this species (Santa Gertrudis) supplied 19 of these cases. The Afrikaner breed is very popular in South Africa, but the Brahman is as yet represented by relatively small numbers while the Santa Gertrudis is uncommon.

AETIOLOGY AND PATHOGENESIS

Like any exposed part, the prepuce, and particularly its orifice, is subject to trauma. In this series, one Friesland bull showed laceration of the prepuce extending from the orifice to near the fornix (Fig. 1). In the other Friesland there was active infection of the *ostium praeputiale*, and in the Jersey there was narrowing of the preputial orifice due to the presence of a fibrous ring, which obstructed the tumescent penis near the base of the glands upon emergence. Both conditions are presumed to have been caused by tick bites.

Congenital stricture of the orifice has been reported [5]. One Afrikaner bull in this series suffered from hypoplasia of the parietal preputial layer.

Apart from these aspects, cases cited in the literature as well as those studied by me, consist mainly of prolapse of the parietal layer of the prepuce, the pathogenesis of which has been regarded by some as being primarily an ulceration of the sheath resulting in prolapse when infection enters [6, 7]. Mostly, however. conformation of the prepuce is blamed, either overtly, or by implication in emphasis upon breed disposition.

Amongst 452 German Holstein bulls, Aehnelt, *et al* [8] found 29 per cent with short sheaths, 64 per cent with sheaths of medium length and seven per cent with long, relaxed sheaths. Of the latter, three bulls suffered from habitual prolapse, one of them develop-

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ing a purulent posthitis. Donaldson and Aubrey [6] recorded 20 cases of preputial prolapse of which 19 had "phimosis" and one "paraphimosis". Of these 19 were Santa Gertrudis; two crosses of this breed; four, Zebu crosses and one as a Hereford. Their ages varied from 18 months to nine years. A pendulant sheath with inflammation is common in beef breeds, particularly Brahmans and Brahman crosses [9-13].

When the sheath hangs close to the ground, it is exposed to further trauma [9], allowing entry of pyogenic organisms, particularly *Corynebacterium pyogenes.*

Scar tissue forms after injury and may cause stenosis [10, 23 17] or deviation of the penis [10]. Initial irritation often leads to increasing prolapse [10]. As a result of narrowing of the sheath, urinary obstruction and rupture of the bladder may complicate the issue [15]. Unless inflammation is present, prolapse does not interfere with service [16, 17].

In general the aetiology in the present group is not connected with copulation injury, but to trauma associated with environmental features such as the presence of thorn bush usually acting in conjunction with predisposing anatomical features.

The only Jersey bull included in this group suffered from stenosis of the preputial orifice. As this breed has a minimum of preputial skin without any tendency towards prolapse of the parietal layer, its virtual absence from this group is according to expectations.

The same applies to the Friesland although they have a relatively better developed preputial skin.

All the other cases can be considered as having had a conformational predisposition.

The Hereford is represented by five bulls. In South Africa it is a well-known fact that this breed has a greater tendency than other breeds of *Bos taurus* (Fig 2) to have preputial skin of excessive length with a consequently greater propensity of the preputial parietal layer to prolapse. In two cases, stenosis of the prolapsed prepuce was actually the main disabling lesion. The Aberdeen Angus, which is not represented in this group, is known locally for prolapse of the prepuce, but the preputial skin is almost invariable of normal length. Furthermore, a length of parietal layer of the prepuce may be visible when an Aberdeen Angus bull stands but it tends to retract while he is in motion. As this breed is not common in South Africa, it cannot be stated with certainty whether the absence of recorded cases among it is incidental or not.

The Afrikaner breed has been bred to thrive under hot climatic conditions, where tick life is very active. To promote heat loss, the dewlap and the preputial skin are pendulous. In certain individuals the latter feature is exaggerated, thus exposing the sheath to trauma by low, hard and spiny vegetation. Ticks, particularly those with long mouthparts, favour the prepuce for attachment. Conformation, therefore, predisposes to pathology of the preputial skin. On the credit side is the fact that the skin of the Afrikaner is tough and resists infection more effectively than that of the imported breeds. Another positive feature is the low incidence of prolapse of the prepuce so that the tender parts are well protected. It is noteworthy, and may be significant in spite of the small number. that the parietal layer of preputial skin was undamaged in both the Afrikaner bulls with stenosis due to trauma. In one case the integumental layer of the prepuce was definitely a great deal longer than is regarded normal for the breed.

The Brahman breed furnishes close to half of the total number of cases in this group. As the total number of Brahmans in South Africa is insignificant relative to the numbers of the other breeds represented in this group, this high incidence indicates a major weakness of the breed under South African conditions. Although bred to thrive under similar environmental conditions as the Afrikaner, the pendulous nature of the preputial skin has been carried to extremes, so that the preputial opening often has a remarkably small ground clearance. This difference between the two breeds may well lie in the fact that the imported representatives of the Brahman breed were obtained from those prototypes that had been evolved in a relatively protected physical environment.

The pendulous nature of the sheath is exaggerated by a general tendency of the parietal preputial layer to prolapse (Fig. 4) not only while the bull is standing, but also during locomotion. As the penis slants obliquely downwards from the scrotum to the preputial orifice, there is a considerable effect of gravity on the retraction apparatus. The prolapse is thus due to a relatively inadequate tone of the *Mm. retractores penis.* Prolapse is further favoured by excess skin in the parietal layer, apparently in relation to the excess of integumentary layer. This was so

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excessive in one case that it in itself was a cause of obstruction.

In the Santa Gertrudis, (developed from cross-breeding Brahman and Shorthorn) excessive length of the preputial skin is also a common feature unfortunately (Fig. 5). Small numbers of this breed are found in South Africa at present. Apparently, it is not as susceptible to prolapse as is the Brahman. Breeders particularly of the last two mentioned breeds should heed the implications of these very undesirable anatomical features described which have profound implications on the future of particularly the last two mentioned breeds.

DIAGNOSIS

In view of what has been stated, the diagnosis is self evident. Stenosis at the preputial orifice is determined by the usual examination. A prepuce skin of excessive length or a prolapse of the parietal layer, is evident from a distance, particularly if there is well marked infection and induration (Figs. 4, 5). The prolapse may hang so low as to be almost in contact with the ground (Fig. 4) and the bull may actually tread on it.

TREATMENT

A retaining purse string suture at the preputial orifice or the application of antibiotic or sulphonamide ointment under pudendal block have been advised [10, 13]. Hoffman [18] considers treatment unrewarding where stenosis is present, as operations on two bulls failed.

For the relief of stenosis, generally, surgery is favoured. For this purpose, various forms of anaesthesia have been employed, i.e. epidural analgesia [19] or general anaesthesia [5, 6, 7, 11] with or without tranquilizer or local infiltration analgesia [5, 7, 11].

The simplest form of open surgery advocated is making a slit from the orifice posteriorly [7, 9, 10, 19], or removing a triangle of skin with the base against the orifice, then slitting the parietal layer [5, 9, 15, 20]. In both instances the parietal layer is sutured to the integumentary layer. To shorten the prolapsed parietal layer, more radical surgery was found necessary in certain cases [6]. Two circumferential incisions were made—one at the edge of the prolapsus and the other at the preputial orifice. The two incisions were joined by a longitudinal incision, the intervening parietal layer was removed and the edges of skin and prepuce united by sutures. Where infection supervened and caused stenosis, a triangle of skin was removed as described above. Postoperative treatment consisted of application of Propamidine* cream and Varidase** injections (the latter in two bulls). The area of operation easily became infected. Out of 11 bulls so treated, eight returned to service but four broke down subsequently. Of the three primary failures, one had severe adhesions, one could not serve because too much of the parietal layer had been excised and one died of toxaemia [6].

Another method consisted of placing mattress or interrupted sutures through skin and prepuce at the end of the sheath. The tissues were divided a short distance distal to the suture line. In addition a longitudinal ventral incision was made or a triangular area of skin was removed and sutured [7].

In a similar technique the external layer of prolapsed parietal layer was incised and stripped down slightly. The internal layer was then cut and the two sutured together progressively as the incision was extended further round the circumference [9, 11, 16].

A minor variation of this technique consisted of making two elliptical incisions on either side and then applying closely spaced thin catgut sutures [13]. A warning has been issued that amputation of too much of the prolapsus may terminate the bull's breeding life [9].

In order to counter the tremendous postoperative swelling that usually occurs in Brahman bulls, and to avoid recurring stricture, Megale [11] devised an apparatus for the prevention of postoperative stricture. This consisted of a tube and clamp applied to the prepuce to allow amputation without haemorrhage. The visible blood vessels were ligated and parietal and integumentary layers joined by sutures. The tube remained *in situ*. The end passed through a cotton pouch which was laced round the preputial skin like a shoe, with the purpose of controlling local oedema. It was stated that good results had been obtained but no subsequent report has been seen.

For the relief of a thin ring stenosis of the *ostium praeputiale*, I have found the technique described by others effective. As this technique is ineffective where the stenosis is thicker than about 1.5 cm, a new operation

*May-Baker.
**Cyanamid.

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has been developed. For amputation of the prolapsed and indurated prepuce a modification of the method reported by others had to be introduced.

The operative techniques employed by me are as follows:

Technique 1 (Fig. 9) is employed when there is stenosis of the *ostium praeputiale* in the absence of a thick fibrous ring. Local infiltration anaesthesia with or without injection of a tranquillizer is usually sufficient. A wedge of integumentary layer, intervening fascia and parietal layer, with its base at the preputial orifice and the apex on the midline posteriorly as far back as deemed necessary, usually about five cm or slightly more, is removed. The integumentary and parietal layers along the cut edges of the wedge are approximated by interrupted silk sutures. Application of technique 1 leaves unyielding fibrous tissue, causing some impedance to erection even though it might result in a wide opening.

Technique 2 (Figs. 6, 7, 10, 11) is employed when the preputial opening is stenosed by a thick fibrous ring, usually 1.5 cm or thicker. Even in an extreme case, where the stenosis is 18 cm long technique 2 can be employed.

A wide circular incision is made some distance away from the stenosing mass of fibrous tissue, close to the hair line. Tissue forceps are attached to the edge of the stenosed opening and tension is maintained while the incision is deepened very gradually to allow careful attention to bleeding points or recognition of large blood vessels in time to divide them between ligatures. Because of the excellent blood supply and the formation of new capillaries near the granuloma, bleeding may be copious and haemostasis has to be ensured at every step. When the subcutaneous tissues have been incised, the incision is deepened by dissecting the fascia against the scar tissue so as not to include any granulations. If the stenosis is longer than about three cm, it can be felt like a cord. The stenosed part of the orifice and parietal layer are thus gradually mobilised and drawn away from the circular incision in the skin. In this manner the more deeply (posteriorly) situated unaffected part of the parietal layer is brought into view. If the distance dissected is more than about four cm, the divided tissues can create excessive potential space after operation, if not given attention. This exigency is met by putting in a series of single No. 0 chromic catbut sutures between the normal aveolar tissues on either side of the granulomatous mass.

The normal part of the parietal layer is incised just beyond the stenosis. If no retaining catgut sutures have been inserted, the cut edge is grasped by tissue forceps to prevent retraction into the depths of the wound and secured to the integumentary layer by interrupted catgut or silk sutures. As the skin is firm, accurate apposition to the parietal layer can be achieved with simple interrupted sutures.

Basically, silk sutures are preferable because of less capillary action than catgut, but subsequent removal is difficult, particularly in fractious bulls. Hence, where little swelling is anticipated, catgut is acceptable, in spite of the mentioned disadvantage. Where extensive resection has to be done, silk has to be used.

In small bulls, the resulting opening admits three fingers with ease, but in big bulls four fingers can be inserted.

Technique 3 is applicable when the preputial skin is of excessive length and indurated by infection and succeeding fibrosis or when the parietal preputial layer has prolapsed and been subjected to repeated trauma with its sequelae. The technique is closely related to that for amputation of the stenosing lining inside the preputial cavity [3].

When the preputial skin is excessively long, the circumferential incision is made at such a site as to reduce the skin to normal

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Fig. 1. The preputial cavity exposed by lateral laceration and healed in that state.
Fig. 2. Hereford — lengthened preputial skin and indurated orifice.
Fig. 3. Hypoplasia of the prepuce. At the orifice a circular incision was made and the prepuce drawn out. The bulge of the prepuce represents a diverticulum containing the curled glans penis.
Fig. 4. Braham — Prolapse of prepuce with severe infection and induration.
Fig. 5. Santa Gertrudis — lenghthened preputial skin and indurated orifice.
Fig. 6. See fig. 10 and 11.
The stenosed part of the prepuce has been amputated, revealing the galea glandis. The next step is suturing the edge of the prepuce to the skin.
Fig. 7. Amputation of prolapsed prepuce — the operation completed.
Fig. 8. Brahman showing extensive post-operative oedema of the preputial area.

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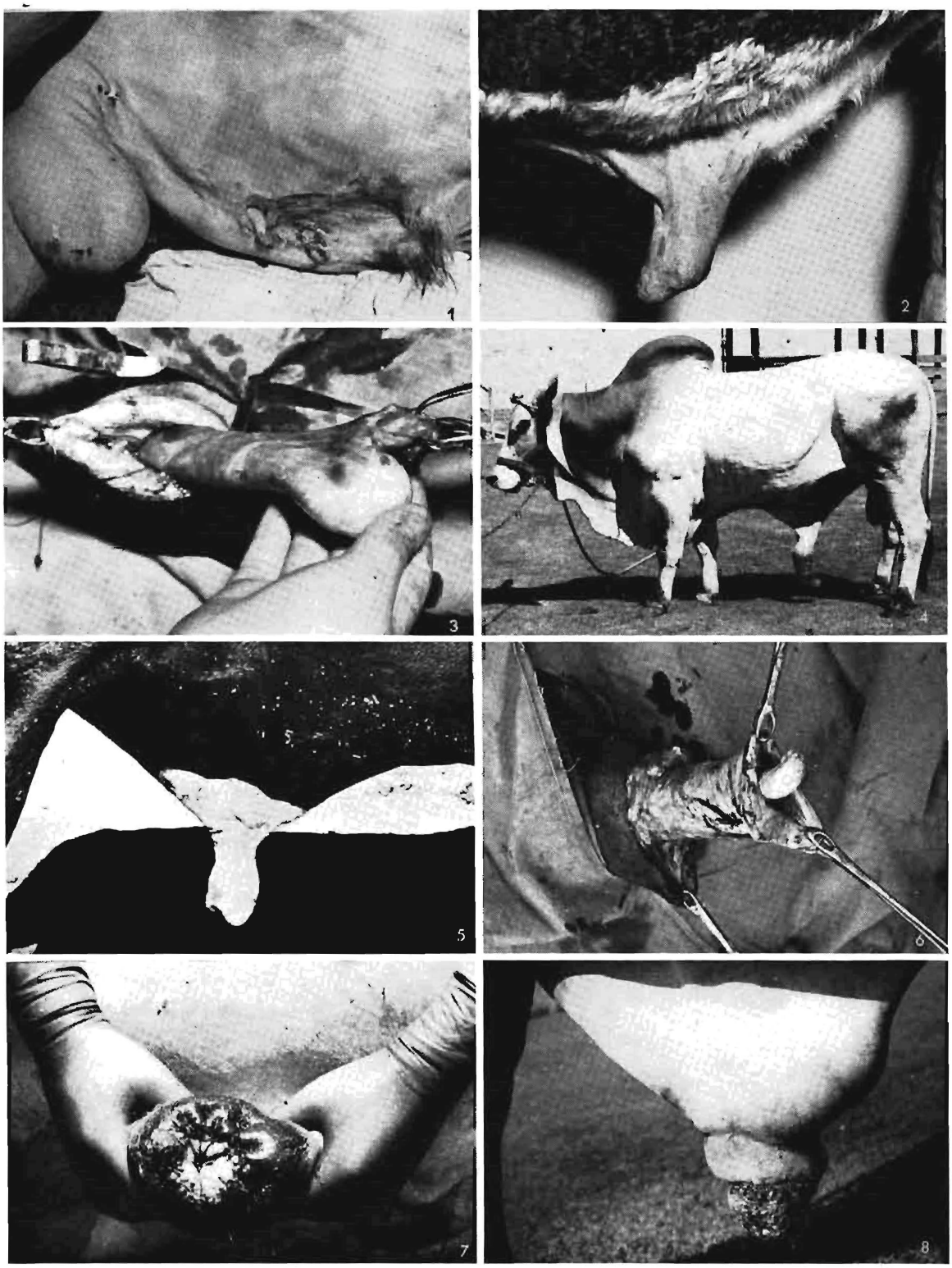

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length, unless, of course, the extent of the lesion forces one to amputate at a higher level, when the remaining preputial skin would be shorter than normal.

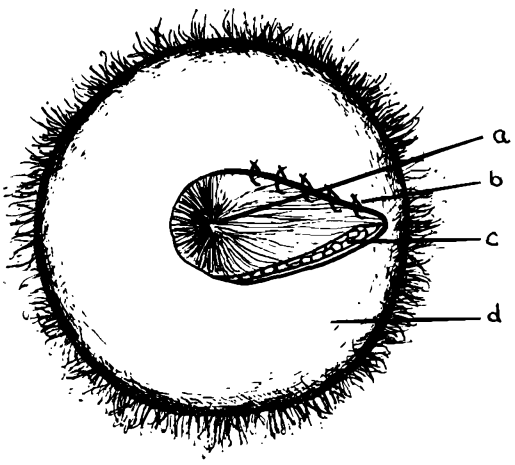
In the case of prolapse of the parietal layer, the circumferential incision is made proximal to the hair line if possible, so that the eventual suture line would not be at the lowest point of the reduced prepuce but a short distance inside the natural preputial opening. The reasons will be set out later.

The circumferential incision is mostly made at right angles to the long axis of the parts to be amputated and not obliquely or in zig-zag fashion[3]. After the initial incision the general technique is similar to technique 4 described in the preceding paper[3]. In suturing, however, the approximation of tissues at the end of the operation involves integumentary and parietal layer when preputial skin has been amputated and between proximal and distal parts of the parietal layer if the prolapsus has been removed. In both cases interrupted sutures are used, of silk if the suture line forms the most dependent point with the patient in standing position, or of No. 3 chromic catgut if the suture line is at a level within the sheath, i.e. proximal to the preputial orifice. Interrupted sutures are applied here to a greater extent than in internal sutures. If preputial skin has been shortened, the remaining skin is firm enough to allow accurate approximation of the parietal layer. If the prolapsed parietal layer has to be removed, the more distal part thereof is found to be less flaccid than the more proximal part, which also permits correct suturing.

After completion of the operation the opening permits the access of three or four fingers depending on the size of the bull.

EVALUATION

There were five failures out of 27, giving a recovery rate of 82 per cent. The five failures included one bull with a nearly completely opened preputial cavity, which healed in that position despite operation according to technique 1; one of extensive haemorrhage and infection around the penis, causing secondary asymmetrical preputial prolapse; one of congenital hypoplasia of the preputial mucous membrane upon which an operation according to technique 3 was done, and two cases of secondary stenosis after premature discharge upon owner's demand.



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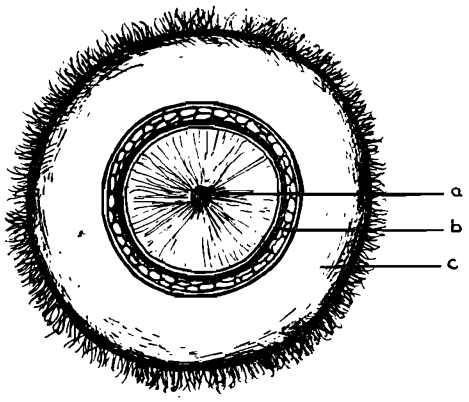

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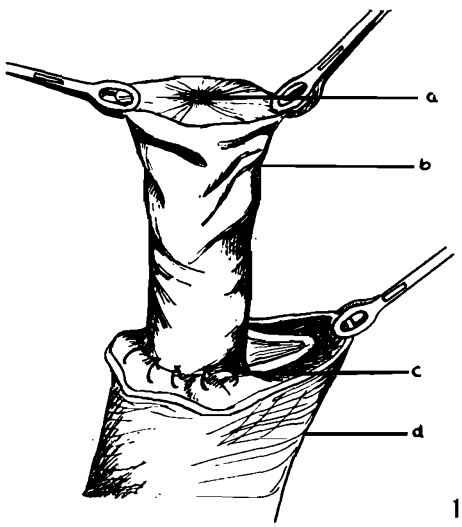

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Certain aspects of the operative techniques have to be discussed.

Technique 1, involving either a linear or wedge-shaped incision of the preputial orifice, is not original and is simply executed. The indication is stenosis at the orifice provided the fibrous ring is narrower than 1.5 cm and not too hard. (If the ring is hard and fibrous, erection will not be complete, even if the opening is sufficiently large). The technique was used only on two bulls after previous operations had been followed by secondary stenosis. Despite success, there was a tendency, during urination, for urine to leak out at the posterior commissure of the previous wound and then to run down to the edge of the circular part of the opening. The repeated moisture at the posterior commissure caused slight eczema. This operation would not have succeeded if applied initially, as the stenosing rings were too wide. Previous operations reduced them to become amenable for operation according to technique 1.

Technique 2 was developed for those cases of stenosis of the preputial orifice where technique 1 was not indicated because of thickness of the stenosing ring or where the ring was so thick as to form a stenosed canal. Operation according to technique 2 succeeded in avoiding exposure of the stenosed area which would have been caused had the preputial skin been incised from the lateral aspect, a procedure likely to lead to adhesions between skin and parietal preputial layer or even penis.

Technique 2 was performed on mild cases. It was the only operation used on five bulls. Of these, it failed in two bulls, in one of which there was some secondary stenosis. This could not be rectified as the owner had insisted on removing the bull prematurely while the opening was ample but the tissues were still actively contracting. The other failure had hypoplasia of the preputial lining and did not represent a fair test of the technique. One bull underwent the operation twice. Several veterinarians had given a hopeless prognosis before admission of this bull. The operation immediately relieved the stenosis of 18 cm. The tremendous postoperative swelling of this Brahman bull (a phenomenon to be discussed fully later) led, as expected, to another ring stenosis which was only 1.5 cm in thickness and almost thin enough for technique 1. Two cases recovered with technique 2 as the only operation. A Jersey bull improved after this operation but a thin stenosing ring formed which was rectified by technique 1.

A Brahman required an amputation (technique 3) which was followed by tremendous and prolonged oedema (Fig. 8). The resultant stenosis was permanently cured by following technique 2.

Another case in a Brahman had a similar course, except that there was a slight secondary stenosis after operating according to technique 2. This was put right by applying technique 1.

Finally, then, against the cases where technique 2 contributed to recovery, only two failures were recorded: the one case that was discharged prematurely and the other that suffered from congenital hypoplasia.

Technique 3 is indicated in cases with a lengthened preputial skin or with prolapse of the parietal layer. It was the method employed in 16 cases. In one case only was there failure, as stenosis occurred after operation, but by that time the bull had been discharged on insistence by the owner. A second operation almost certainly would have led to recovery. Two out of the 16 cases had to be operated on again and were then normal, the first after technique 2 had been used and the second after two operations, one according to technique 2 and one according to technique 1.

With an excessively long preputial skin a circular incision is made at, or close to, the site which would reduce the preputial skin to normal length. This means that the skin

Fig. 9. V shaped enlargement of stenosed preputial orifice.
(a) Ostium.
(b) Sutures uniting parietal layer of prepuce and preputial skin.
(c) Wound still to be sutured.
(d) Preputial skin.

Fig. 10 Operation for relieving deep stenosis of ostium.
(a) Stenosed ostium.
(b) Circular incision around ostium.
(c) Preputial skin seen end-on.

Fig. 11. See fig. 10 — Second stage of operation.
(a) Stenosed ostium.
(b) Stenosed prepuce dissected out, surrounded by granulation tissue.
(c) Anchoring sutures between subcutis and tissues around the prepuce.
(d) Preputial skin.

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carrying the long preputial hair, is lost. As this suture line lies at the lowest point, urine tends to keep it moist for long periods. This causes local irritation and possibly infection, a state of affairs not materially relieved by growth of ordinary hair. This emphasizes the physiological importance of long hair at the preputial orifice to lead off the urine.

When the prolapsed mucous membrane is amputated, it is found that the incision need not be so close to the hairline as to cause the suture line to lie at the lowest point. It is best to amputate some distance away, so that the suture line would be drawn up into the preputial cavity after operation. It is more protected there from outside contamination, and the skin providing growth of long hair is not disturbed.

In two cases the zig-zag incision was employed[3]. Both were completely successful without a suggestion of narrowing the normal lumen. Experience with the zig-zag incision in the previous and present groups is thus very encouraging.

The most recent operations according to technique 3 were done with the specific purpose of amputating the prolapsed prepuce as far from the natural preputial orifice as possible, bearing in mind the purpose of the operation. These cases showed much less postoperative oedema than those where the amputation was done at the hairline.

Any operation on the prepuce is likely to give rise to considerable oedema, as the site is the most dependent part of the abdomen. The extent of the oedema and the time it persists in a particular type of operation, is directly related to the length of the preputial skin. As could be expected, the Brahman is by far the worst, in that the oedema can last well over a month.

Postoperative inflammatory oedema naturally occurs after all operations and this is related to the type and severity of operation and not to conformation of the prepuce and thus to the breed. The tremendous swelling encountered, particularly in the Brahman, is mainly *stasis* oedema. In amputation operations such large blood vessels have to be ligated as to grossly interfere with local circulation. Venous and lymphatic return is hampered by minimal *vis a tergo*. The persistent oedema lowers local resistance against infection which can supervene easily as the prepuce is particularly exposed to contamination. Further, the oedema results in excessive

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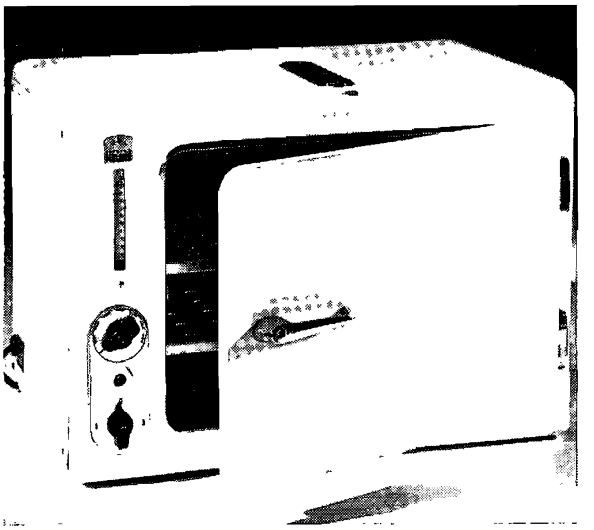

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fibroplasia and has a prospensity towards stenosis.

Various forms of treatment have been used by different surgeons in order to control the oedema. These include corticosteroids, the use of which is open to severe criticism. As the oedema is mostly due to *stasis* and not of inflammatory nature, the corticosteroids will have little effect. Even though these hormones reduce fibroplasia, their administration has to be continued for such a long time as to make the venture very expensive—this is apart from the well known undesirable effects of prolonged corticosteroid administration. The report of Megale [11], who employed special apparatus to control oedema, is interesting but a follow-up report would have permitted better final assessment of his methods.

By and large it can be concluded that the methods described have given good results: Nevertheless, it is essential to have the bull under observation for a long time; lack of co-operation from the owner will greatly reduce the success obtained. In those cases suffering from oedema without fibrosis and with slight preputial prolapse, non-operative treatment may be effective.

REFERENCES

1. Hofmeyr C. F. B. 1967 *Jl S. Afr. vet. med. Ass.* 38:275.
2. Hofmeyr C. F. B. 1967 *Jl S. Afr. vet. med. Ass.* In press.
3. Hofmeyr C. F. B. 1968 *Jl S. Afr. vet. med. Ass.* In press.
4. Hofmeyr C. F. B. 1966 *Surgery of Bovine Impotentia Coeundi.* Thesis. University of Pretoria.
5. Smythe R. H. 1959 *Clinical Veterinary Surgery.* London. Crosby, Lockwood & Son, Ltd.
6. Donaldson J. E. & Aubrey, J. N. 1960 *Austral. vet J.* 36: 380.
7. Frank E. R. 1955 *Veterinary Surgery.* Minneapolis, Minn. Burgess Publishing Co.
8. Aehnelt A. E., Liess, J. & Dittmann, J. 1958 *Dtsch. tierärztl. Wschr.* 65: 385.
9. Milne F. J. 1954 *J. Amer. vet. med. Ass.* 124: 6.
10. Gibbons W. I. 1956 *N. Amer. Vet.* 37: 650.
11. Lenert A. A. 1956 *J. Amer. vet. med. ass.* 129: 506.
12. Megale F. 1943 *Arquivos das Escol Superior de Veterinaria de Universidade Ruvae do Estado de Minas Gerais.* 1: 67.
13. Roberts S. J. 1956 *Veterinary Obstetrics and Genital Diseases.* Michigan. Edwards Brothers Inc.
14. Götze R. 1934 *Dtsch. tierärztl. Wschr.* 42: 589.
15. Silbersiepe E. & Berge, M. 1958 *Lehrbuch der speziellen Chirurgie.* Stuttgart. Ferdinand Enke Verlag.
16. Wheat J. D. 1951 *J. Amer. vet. med. Ass.* 118: 295.
17. Zemjanis R. 1962 *Diagnostic and Therapeutic Techniques in Animal Reproduction.* Baltimore. The Williams and Wilkens Co.
18. Hoffmann W. 1939 *Schweiz. Archiv. Tierheilk.* 79: 418.
19. Berge E. & Westhues, M. 1958 *Tierärztliche Operationslehre.* Hamburg. Verlag Paul Parey.
20. Laing J. A. 1955 *Fertility and Infertility in the Domestic Animal.* London. Bailliere, Tindall & Cox.

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DISTOKIE BY 'N MERRIE: 'N KLINIESE GEVAL

JOHANN VAN STADEN*

SUMMARY

A mare was relieved of a normal, live foal about thirty-four hours after rupture of the allantochorion and thus some considerable time after the onset of partus. This occurred in spite of the fact that examination *per vaginam* elicited no suckling reflex and no response to pinching of the nostrils. The dystokia was due to an easily corrected lateral flexion of head and neck of an otherwise normally positioned foetus.

OPSOMMING

'n Merrie is verlos van 'n normale, lewendige hingsvul, ongeveer vier-en-dertig uur nadat die allanto-chorion gebars het en dus moontlik ietwat langer na aanvang van partus, dit ten spyte van die feit dat by ondersoek *per vaginam* geen suigrefleks en geen reaksie op knyp van die neus ontlok kon word nie. Die distokie was te wyte aan maklik regstelbare fleksie van nek en kop van die andersins normaalliggende fetus.

INLEIDING

Distokie by perde is relatief seldsaam. Nog seldsamer en dus noemenswaardig is die verlossing van 'n lewendige en gesonde vul tenminste vier-en-dertig uur en moontlik veel langer na aanvang van partus.

GEVALVERSLAG

Pasiënt: 'n Ligte tipe plaasperdemerrie, ses jaar oud in redelik goeie voedingstoestand. *Anamnese:* Die merrie het eenmaal tevore op normale wyse geboorte gegee aan 'n gesonde vul. Sy was drie jaar gelede haltermak en het sindsdien uitsluitlik op en van die veld gelewe, met slegs periodieke hantering in 'n drukgang vir behandeling teen bosluise. Vroeg die oggend is deur die eienaar gesien dat sy besig was om te vul. Sy het opgestaan en by die volgende sametrekking het 'n groot hoeveelheid vloeistof afgekom.

Ondersoek: Ongeveer drie uur namiddag die volgende dag is die merrie deur my gesien. Sy het op haar sy gelê met pote gestrek. Geen deel van die vul of vrugvleise was te bespeur nie. Slegs een enkele buiksametrekking is gesien. Sy was sterk genoeg om op te staan en is in 'n drukgang gejaag, waarna met enige moeite 'n toom aangesit is. Dit was die enigste vorm van bedwang, aangesien berustings- en kalmeermiddels nie beskikbaar was nie.

Die algemene habitus was goed en geen tekens van toksemie was bespeurbaar nie.

Ondersoek *per vaginam* is uitgevoer deur regs van die merrie te staan en met die linkerhand deur die dwarspale van die drukgang te werk. Die skede was bedek met 'n dun lagie taai slym, die cervix wyd oop en die fetus nog in die baarmoeder. Slegs die pote was tasbaar deur 'n vlies, wat later geblyk het die amnion te wees.

As gevolg van protesreaksie van die merrie kon die een poot slegs by 'n tweede poging buite die vulva gebring word, die amnion deur middel van 'n skêr oopgesny word en albei pote bevry word. Op hierdie stadium het 'n tweede abdominale sametrekking die uitvloei van 'n groot hoeveelheid geelbruin, troebel amnionvloeistof tot gevolg gehad, maar geen verdere uitstoting van die fetus nie.

Verdere ondersoek het getoon dat die vul in anterior longitudinale voorstelling en dorso-sakrale ligging was, met fleksie van kop en nek na links. Die suigrefleks was afwesig en geen reaksie op knyp van die neus is verkry nie.

Regstelling: Regstelling het met min moeite plaasgevind deur die bokaak vas te vat met die linkerhand, palm oor die neus, vingers in die bek en koudaalwaartse trekking. Verdere trekking op die pote het 'n terugtrekreaksie van die doodgewaande vul en hewige gesteier van die merrie verwek, sodat sy weer met veel moeite in die drukgang teruggekry moes word.

* Posubs 81, Grootfontein, S.W.A.

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Die pote van die fetus is met vultoue gefikseer en die bokaak met die linkerhand vasgevat. Met die hulp van 'n enkele handlanger aan die toue is die vul binne twee minute uitgetrek.

Nabehandeling: Die vul, 'n hings, het sy kop gelig, asemgehaal en sy pote beweeg onmiddellik nadat slym uit bek en neusgate verwyder is. Die naelstring, wat gebreek het toe hy grond getref het, is met jodium ontsmet.

Die vrugvliese was stewig vas en is nie verwyder nie. Twee steekpille (Agricura Laboratoria) is in die baarmoeder geplaas. Die besoek het in sy geheel twee uur geduur.

Die merrie en haar vul is in 'n kampie geplaas en met die volgende waarneming vyf uur later deur die eienaar, was die vrugvliese reeds uitgestoot. Die vul het normaal gelyk en rondgeloop.

Een inspuiting van 30 ml Reverin is op my voorskrif deur die eienaar gedoen, maar daarna was die merrie te sku vir verdere behandeling. Na twee maande was merrie en vul nog in uitstekende toestand.

BESPREKING EN GEVOLGTREKKINGS

Volgens die ondersoek en verloop van die verlossing kan byna met sekerheid aanvaar word dat die fleksie van kop en nek van die vul vir die distokie verantwoordelik was. Met baarmoeder- en buiksametrekkings het die merrie wel daarin geslaag om die allanto-chorion te laat bars, maar nie die amnion nie.

Die feit dat slegs twee buiksametrekkings gedurende meer as 'n uur en geen baarmoedersametrekkings waargeneem is nie, regverdig die stelling dat die uterus as gevolg van volgehoue isometriese sametrekkings uitgeput geraak het, wat dan moontlik opheffing van kontak tussen uterus en allanto-chorion vertraag het. Dit het op sy beurt fetale lewe onderskraag en dus ook die plasenta-bloedsomloop. Nadat laasgenoemde onderbreek en die fetus verwyder was, het die vrugvliese spontaan en spoedig losgelaat.

Gewoonlik word aanvaar dat verlengde partus by die merrie tot spoedige dood van die vrug lei. In hierdie geval is 'n lewendige vul gebaar, ongeveer vier-en-dertig uur nadat die allanto-chorion gebreek het. Binnebaarmoederlike ondersoek van die vul kon geen reflekse van die vul ontlok nie. Hierdie geval toon dus dat daar nie te gou aanvaar moet word dat 'n vul dood is nie en dat 'n verwagte onhanteerbare pasiënt tog selfs sonder verdowingsmiddels gehanteer kan word.

ERKENNING

Die toestemming van die Direkteur van Landbou, Administrasie van Suidwes-Afrika tot publikasie van hierdie verslag word met dank erken, asook die advies van prof. S. J. van Heerden en dr. R. Coubrough by die opstel hiervan.

BOOK REVIEW

THE VETERINARY ANNUAL. EIGHTH ISSUE 1966/1967

Edited by

W. A. Pool, mrcvs.

John Wright & Sons Ltd., Bristol. Pages 354, Figs. 22. Publ. Price 63s.

The Eighth Issue of The Veterinary Annual represents a review of the latest developments over a wide field of activity in Veterinary Science. Thirty-six specialists have contributed on different subjects. Like the previous issues, this book is particularly suited to the needs of the busy practitioner who has very little time for extensive reading. The reader is assisted by a complete index. An introduction to interesting new topics such as veterinary ethology or the study of animal behaviour and a description of recent pharmaceutical preparations and appliances, add to the value of the book.

B. C. J.

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Jl S. Afr. vet. med. Ass. 39 (2): 105-108 1968

THE HANDLING, HOUSING AND NUTRITION OF CAPTIVE WILD MEERKATS

I. F. ZUMPT*

SUMMARY

Various methods of handling, housing and nutrition of wild meerkats are discussed. It was found that these animals can be kept successfully in the laboratory.

INTRODUCTION

Since the first known outbreak of rabies in South Africa was recorded in 1892 [1,2], research into the epizootiology of the disease has been sporadic in nature but the contributions to the epizootiology of rabies made by du Toit [2,3] and Snyman [1,4,5] have never been surpassed in this regard and our present knowledge is mainly based on the findings of these authors. It was felt that the numerous problems of investigation into the habits of rabies carriers necessitated another approach. Observations and experiments were carried out with the object of eliminating adverse factors which prohibited the successful use of meerkats in the study of rabies in the laboratory. The following species were studied:

Cynictis penicillata, Suricata suricatta and *Xerus inauris* [6,7]. *C. penicillata* (G. Cuvier) Order: *Carnivora* Family: *Viverridae* Sub-family: *Herpestinae* Common names: English: Cape Yellow Mongoose, Bushytailed, Yellow or Red Meerkat Afrikaans: Geel-, Rooi-, Witkwas- or Witpuntstertmeerkat *S. suricatta* Erxl. Order: *Carnivora* Family: *Viverridae* Sub-family: *Herpestinae* Common names: English: True, Common, Cape, Suricate or Slender-tailed meekat Afrikaans: Stokstert, Graatjie or Gewone Meerkat.

X. inauris Zimmermann Order: *Rodentia* Family: *Sciuridae* Sub-family: *Sciurinae* Common names: English: Ground Squirrel, Bush- or Fantailed Meerkat. Afrikaans: Waaierstertmeerkat.

No subspecies were taken into consideration [8].

HANDLING

Careful handling of these small mammals is essential in experimental work. Painful, deep bite-wounds can be inflicted with the danger of contracting rabies ever present. It is also necessary to prevent injury to the animals, which could affect experimental results. In order to capture the animals various methods of trapping were used, *e.g.,* several variations of the boxtrap [9, 10, 11, 12, 13], two box-type traps designed by the author, fine-wire or nylon snares [9], "funnel-no return" rat traps, and two types of jackal-traps. All these had several disadvantages. In the majority of cases, especially when there was an abundant food supply, it took days before a single animal was trapped. The animal had to become accustomed to the trap and by this time the bait was dispersed by insects or had lost its attraction. In many cases traps were set off by birds and had to be re-set. Many of these traps are difficult to conceal and attracted the attention of humans, who made the efforts to catch meerkats difficult. The above-mentioned methods of trapping were discarded as they were all inefficient and caused many injuries.

The present method of capturing meerkats was developed by Mr. H. de Bruyn** and is superior in all respects. In the routine extermination of meerkats all burrows treated with carbon monoxide are closed up with sand, and only some small isolated active colonies are not treated. Two holes are transformed into a short cul-de-sac on the periphery of a selected colony. The colony is then dug-up systematically by following each tunnel from its exit hole. As the digging is progressing the holes are probed with a stick from time to time. The presence of a meerkat is easily determined by the type of resistance felt or the presence of some hairs on the end of the stick. When the space be-

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comes limited, it is easy to remove the meerkat with a doubly gloved hand. Should one escape, it will enter a prepared hole after a short search, and it can be retrieved as described.

If burrows are selected where several meerkats have been seen to enter, success is virtually assured and between three and eight meerkats can be caught per colony. A strong butterfly-net is also useful for catching escaping meerkats.

RESTRAINT

Once the meerkats have been caged, the actual handling begins. There are several useful methods described in the literature [7, 9, 10, 11, 14, 15]:

1) Snare on short stick: This is a safe method for the handler but causes strangulation and lacerations to the animal. It is also very tedious as the meerkat is very agile and quickly learns to avoid the snare.

2) Hand-method: Meerkats have to be caught behind their heads, but even gloves do not always protect against bite-wounds. Experienced handlers prefer the hand-method as this causes the least injuries to the animal [16].

3) "Squash-box": Cages are fitted with a solid wooden board, which is fixed to a two foot long handle which, when pushed, moves the board across the cage and so traps the animal between cage wall and board. It is then easy to remove the meerkat by seizing it behind the neck.

4) Indirect: The meerkat to be caught is transferred into a narrow bag or cage by tilting the cage so that the meerkat falls into the former where it cannot move due to limited space. This method is effective, but skill is required to remove the animal from the bag.

Melcior and Iwen [11] describe methods of restraint in arctic ground squirrels by means of a girth-hitch, a stretching board and a cone-bag, all of which have disadvantages; the danger of being bitten is great. Baumgartner [10] constructed a handling box for fox squirrels with a false bottom similar in nature to the described squash-box. Emlen [14] used a reinforced cloth-cone for catching wild rats. It might be useful, although a meerkat cannot be persuaded to enter it. A zipper-tube described by Shadle and Skarupinski [15] is similar to that of Emlen.

HOUSING

Every wild animal removed from its natural habitat and kept in captivity changes its behaviour patterns. As the three species of meerkats all live in burrows, their habits are very difficult to study in detail. In cages or cement runs, the change in environment influences the reproduction cycle in particular. This is proved by the fact that few meerkats were born in captivity in the past, the National Zoological Gardens of South Africa [17] recording the birth of only six litters of *C. penicillata* during 52 years.

The majority of zoological gardens use stone houses with cement runs to house meerkats. In some instances, sand or soil boxes are added. Meerkats for experimental purposes are kept in metal cages in which the behaviour studies are difficult.

Standards for the management of the ferret, which has similar habits to *C. penicillata*, were established by Edwards [18], Bleby [19] and Murray [20]. Their instructions were used as a general guide.

Good care and management is indispensable to experimental success. Cleanliness, static conditions of temperature and humidity, and the absence of draughts are essential. With the ultimate aim of studying the ecology of rabies, several different methods of keeping meerkats in captivity were studied, and only those which were found useful will be discussed. Meerkats are shy in cages, but can be handled efficiently. In artificial burrows their behaviour is normal, but they can only be caught with great difficulty. The choice of cage always depends on the type of experiment:-

(1) Simple Cage: A standard wire cage useful for most experiments measures 20 x 13 x 12 inches. Foot lesions may be prevented by the introduction of a wooden floor which must be cleaned thoroughly every week. Fixation of the water- and foodtroughs is essential, and these are kept in place by wire clips. In the case of squirrels, the addition of nesting material in the form of a piece of hessian is advocated. The biggest disadvantage of such a cage is the danger of draught. The use of a bag to cover the cage at night time is sufficient to prevent losses from pneumonia.

(2) Compound Cage: This wood and wire cage measures 2 x 1½ x 1⅓ feet and comprises an exercising, a feeding and a

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sleeping compartment. This is a suitable breeding cage. Wooden surfaces should be lined with asbestos to facilitate the frequent cleaning. Small peepholes and four inches popholes with sliding hatches are useful. The dark sleeping quarters are covered with a layer of sawdust and during the gestation period nesting material must be added. This cage is warm and draught-free. It eliminates most disturbances, but is difficult to clean and is expensive.

(3) Enclosed Burrow: This is a small burrow covered with fine chicken wire and is surrounded by iron sheets, four inches above, and four feet below, ground level. On two sides a box-trap is built into the wire cover. Feeding is done in these traps, and if required a meerkat can then be caught. But the animals cannot be observed continuously, which is a great disadvantage during the breeding season.

NUTRITION

Successful maintenance of captive wild animals depends largely on correct nutrition. In the case of meerkats the difficulty consisted in obtaining a balanced diet for each species. It is out of the question to feed the different animals on those foods obtainable in their natural habitats, as this would necessitate the breeding of various insects on a large scale and the collection of roots, bulbs, plants and seeds in huge quantities. Experiments were carried out to find alternative food:-

(a) *C. penicillata* is carnivorous. In nature its feeding habits depend on the availability of foods and are thus influenced by the season. In spring and summer various types of insects eg, termites and crickets are eaten, whereas insects with hard limbs or shield are only eaten when the supply of termites decreases. During autumn and winter virtually anything available is eaten, eg. locusts, beetles, other insects, eggs, carrion and even birds and chickens will be attacked. Red meerkats caught in late spring or summer adapt to artificial feeding only with great difficulty, whereas those caught in late winter do so very quickly. Meerkats have been kept successfully for one year and longer on two ounces of cold, raw meat per day. Those caught in summer must be adapted over a few days by feeding warm meat or that from freshly killed animals. If this is not done, a large percentage of meerkats will refuse to eat and will die. Animals caught in the winter months are starved for one day and take the cold meat presented without difficulty the following day. In practice each meerkat receives two ounces of semi-lean meat in the morning. In the evening the feeding-tray should be removed and cleaned. The development of a rough coat after some weeks in captivity, is countered by the addition of a few drops of multivitamin syrup. The same effect may be obtained by putting the cages into the sun every second day. Raw eggs, fried and boiled meat, various insects, intestines and other offal are uneconomical and not always available. In order to decrease the feeding costs and raise the standard of hygiene, various types of balanced ration pellets and cubes have been tested, but without success. Dog, rat and mouse cubes are refused in dry and in moistened form. All attempts at gradual adaptation fail. Food mixtures containing maize-, carcass- and fish-meal were used in several experiments and it was found that the following mixture proved most successful:- maizemeal 50%, carcass-meal 20%, fish-meal 30%.

(b) *X. inauris* is herbivorous. In nature it lives on groundnuts, maize, tubers, bulbs, roots, soft stems of plants[21], seeds (sunflower, kaffircorn) and other vegetable matter. The method of giving groundnuts, whole maize and fresh lucern *ad. lib.* is very successful and is used in zoological gardens. Experiments carried out showed that a mixture of maizemeal 85%, carcass-meal 10% and sunflower-meal 5% gave the best results. If only fed maize-meal animals progressively loose weight and may develop dermatitis. Both species (a and b) responded favourably to the described mixtures as soon as the period of adaptation had been overcome. These food stuffs proved to be not only easily available and economic but also hygienic and all test animals gained weight and were clinically healthy. Several females gave birth to normal viable young.

(c) *S. suricatta* is omnivorous in nature and lives on insects as well as on maize,

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seeds, roots and other vegetable matter. When caught at an early age, they become very tame and feeding is no problem. They can be fed best on mince-meat and brown bread and will eat almost anything such as groundnuts, maize, sunflower and kaffircorn seeds, carrots, insects and eggs[1]. All aged specimens refused all food and became progressively weaker until they died after a few days.

CONCLUSION

The experiments and observations have shown that it is possible to maintain *C. penicillata* and *X. inauris* successfully on various mixtures of maize-, fish-, and carcass-meal. Young *S. suricatta* responded well to a brown bread and mince-meat mixture. Feeding is relatively easy once adaptation to a certain food mixture has taken place.

ACKNOWLEDGEMENT

I wish to express my thanks to the Chief, Veterinary Field Services, for the permission to publish this paper and Mr. H. W. de Bruyn for his untiring field-work.

REFERENCES

1. Snyman P. S. 1940 *Onderstepoort J. vet Sci. Anim. Ind. 15 : 9.*
2. Du Toit P. J. 1929 *Rabies in South Africa.* Union of S.A. Dep. Agrig.
3. Du Toit P. J. 1936 *Quart. Bull. Health Org.* V:162.
4. Snyman P. S. 1937 *Jl. S. Afr. vet. med. Ass.* 9:126.
5. Snyman P. S. & Thomas A. D. 1939 *Acta Conventus Tertii de Tropicis Atque Malarias Morbis.* 1:616.
6. Ellerman J. R., Morrison-Scott T. C. S. & Hayman R. W. *South African mammals, a re-classification.* Brit. Mus. (Nat. Hist.) London.
7. Roberts A. 1951 *The Mammals of South Africa.* Johannesburg. Central News Agency.
8. Lundholm B. G. 1955 *Ann. Transv. Mus.* 22:305.
9. Horn E. E. & Fitch H. S. 1946 *J. Mammal.* 27:220.
10. Baumgartner L. L. 1940 *J. Wildl. Mgmt.* 4:444.
11. Melchoir H. R. & Iwen F. A. 1965 *J. Wildl. Mgmt.* 29:671.
12. 1935 *Rabies* Dep. Publ. Health & Dep. Agric. Bull. No. 50, Un. S. Africa.
13. Rongstad O. 1965 *J. Mammal.* 48:76.
14. Emlen J. T. 1944 *J. Wildl. Mgmt.* 8:264.
15. Shadle A. R. & Skarupinski I. 1935 *Science* 82:335.
16. Ewer R. F. 1966 *Afr. Wild Life* 20:53. ?
17. Brand D. J. 1963 *Proc. Zool. Soc. Lond.* 140:651.
18. Edwards J. T. G. 1957 *Handbook on the Care and Management of Laboratory Animals.* 2nd ed. p. 459 London. U.F.A.W.
19. Bleby J. 1966 *Vet. Rec.* 79:869.
20. Murray G. N. 1941 *Onderstepoort J. vet. Sci.* 17:297.
21. Bigalke R. 1921 *JL. Dept. Agric.* South Africa. 431.

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Obituary

GEORGE FREDERIK VAN DER MERWE

It was a great shock to the veterinary profession when Dr. "Okkie" van der Merwe suddenly passed away on the 16th of January, 1968, after a short illness. He is survived by his widow, son and daughter, to whom we extend our sincerest condolences.

George Frederik van der Merwe was born on the 14th of April, 1909. His outstanding capabilities were first recognised while he was at the Agricultural School at Gamman near Windhoek, when he won a bursary awarded by the administration of South West Africa, to study veterinary science. He gained the B.V.Sc. degree at the University of Pretoria in 1935.

On the 6th of January 1936 he assumed duty as a Government Veterinary Officer in Windhoek. He married Aletta Jacoba Magdalena Pyper on the 1st February, 1937.

While stationed at Gobabis he assisted in a campaign against Bovine Pleuropneumonia in the Kaokoveld. Later he was responsible for the inspection of meat for export from Walvis Bay.

Within his own area he had to implement the Cattle Improvement Ordinance and soon had a reputation for his good judgement leading to his appointment as a junior judge by the Shorthorn Breeders Association.

In 1947 Dr. van der Merwe was transferred to Windhoek to assist the Director of Agriculture while still a State Veterinarian. He was officially appointed to act for the Director in his absence. Promotion to Senior State Veterinarian followed.

Dr. van der Merwe's health was never robust and eventually he was compelled to ask for a transfer to the then Union of South Africa. He arrived at Grahamstown in 1956 and there took a particular interest in heartwater, making some valuable contributions to our knowledge of immunisation of small stock.

In 1959 he was made Sub-Director of Veterinary Services with headquarters at Vryburg. After some 19 months he was transferred to the Veterinary Field Services head office in Pretoria where his title was subsequently changed to Assistant Chief. His section was responsible for the co-ordination of a variety of functions, *inter alia*, animal health schemes, diagnostic services, projects, pig and poultry diseases, surveys, artificial insemination and sterility diseases. On a number of occasions he acted for the Deputy Chief and in 1967 was promoted to that rank with the task of co-ordinating animal health schemes and diagnostic services.

During his long service Dr. van der Merwe became known for his neatness, meticulous attention to all facets of a problem and unruffled conduct in difficult situations, characteristics which made him most valuable to the Service and well-liked in public life. He enjoyed his bowls in his latter years and did very well in club and national championships.

HERBERT HENRY CURSON

On January 15th, 1968, there passed away a respected colleague and teacher of many of the older Onderstepoort graduates, Herbert Henry Curson.

Born on 22nd September, 1892, at Umzinkulu, as eldest son of Henry Charles Curson and only one of the family to return to South Africa, he was educated at Michaelhouse,

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Balgowan and graduated from the Royal Veterinary College, London, in 1914, having gained fourteen College medals.

His record of service may be briefly summarised thus: Jan. 1915—March 1917: Field Officer in the Division Veterinary Services at Western Cape; until October 1918 served on the Inter-Colonial Veterinary Commission on rinderpest in German East Africa; until February 1920 research officer at Onderstepoort (vaccine production); until March 1921 in charge of Grahamstown Laboratory, where he made observations on toxic plants; until March 1923 in charge of research station on trypanosomiasis he established in Zululand; until September 1936 research officer at Onderstepoort; until September 1952 Deputy Director of Native Agriculture. In addition he was Lecturer in Hygiene and Animal Management in the recently established Faculty of Veterinary Science for two years and from April 1926 until September 1936 occupied the Chair of Anatomy. During World War II he served in the army as Captain (August 1940—November 1943). On retirement he immediately commenced work in the Military Archives at Defence Headquarters, to which he was attached for almost ten years.

In 1925 he spent a year overseas, receiving the degree Dr. Med. Vet. from the Tierärztliche Hochschule, Hanover, and the Fellowship of the Royal Veterinary College.

He has written 175 papers on subjects ranging from teratology and anatomy, reproductive physiology, botany, entomology, pathology and infectious diseases to history of the veterinary profession and military history. Two monographs, namely "Colours and Honours in South Africa, 1783—1948" and "Regimental Devices in South Africa, 1783—1954" stand to his credit, as well as a joint work on "The South African Field Artillery in German East Africa and Palestine, 1915—1919."

As Honorary Secretary to the South African Veterinary Medical Association he assisted in drafting the original Veterinary Act and was a member of the Veterinary Board. He officiated as Editor of this Journal and represented his colleagues on the Public Servants' Association. On 13 September 1956 he was unanimously elected Honorary Life Vice-President of our Association. Long before the present era he took particular pains to develop Afrikaans as a scientific language.

To the three sons of this indefatigable worker and gentleman in the finest traditional sense, their wives and his grand-children, the members of this Association extend their sincere sympathy and share in their gratitude for such a life.

CONRAD MENTZ THESEN MELDAL-JOHNSON

The tragic death in February of Dr. C. M. T. Meldal-Johnson in an accident at Estcourt has saddened and shocked his many friends and colleagues.

Born in the Eastern Cape some 48 years ago, Mentz or "Johnnie," as he was affectionately called by so many, was educated at Graham College, Grahamstown, Rhodes University and Onderstepoort, graduating in 1944.

He was thereafter commissioned in the S.A. Veterinary Corps and saw active service on the animal transport ships between South Africa and Karachi. Following demobilisation this valuable experience of sea transportation led him into the employ of UNRRA, where he accompanied several shiploads of animals to Europe.

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In 1949 he married Mavis Bryant and there are two children. A long period of useful service as State Veterinarian, Kingwilliamstown, where he was highly esteemed by the farming community, ended in 1961 when the family emigrated to New Zealand. They stayed there until September 1967. During this period he was largely concerned with the tuberculosis eradication scheme and also made several trips transporting stock to South America. On returning to the Republic he rejoined the Field Service and was stationed at Escourt for only five month prior to his untimely death.

Johnnie was an exceptional character, high spirited, cheerful and bubbling over with fun and a generous friendship for all. His energy and infectious enthusiasm for his work was legendary. We can only salute his bright and joyous memory and also extend our heartfelt sympathy to his wife, his daughter Yolande and son Eric.

RICHARD PAINE

We regret to announce the death on the 29th of September last yearof Richard Paine F.R.C.V.S. Born in England in 1879 he qualified at the Royal College, London in 1901. In 1902 he joined the service of the Cape Colony and was stationed at East London and later Beaufort West. He was closely associated with the campaign against glanders and wrote his thesis for the fellowship on this subject. He was at Elsenburg from 1905 to 1910 and acted as Principal for some time. Treasured possessions from this period were two letters of appreciation from the Prime Minister of the Colony, John X. Merriman.

He was then associated with the campaign against East Coast fever in the Transkei and moved to the Grahamstown laboratory in 1922 where he worked with Dr. Martinaglia on tuberculosis in kudu and baboons. In 1934 he took over the control of Allerton Laboratory, Pietermaritzburg, where he remained till his retirement in 1939. He then practised in Pietermaritzburg till 1947.

We extend our sincere sympathy to his family and friends. He is survived by his wife, daughter and two sons.

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BOOK REVIEW

A GUIDE TO COMPARATIVE VETERINARY ANATOMY

WILLIAM M. STOKOE

Bailliére, Tindall and Cassel Ltd., London, 1967. X & 162 pages; off-set litho, loose-leaf, plastic ring binding; 8½"x6". Publ. price 21s.

In consonance with the modern mini-trend —mini-cars and mini-skirts—here we have a mini-anatomy. (Yet bear in mind that the Anatomia Porci was written a thousand years ago and was twentyfive-fold shorter). This is a statement of fact, neither of disparagement nor of applause, that depends entirely on the reader's attitude. As the horse still serves as type animal—and there is nothing wrong with that except that it may cause the neophyte in many schools some initial discomforture—and as the older terminology and some out-dated concepts are still employed (e.g. the fore-stomachs as dilatations of the abdominal oesophagus, the classification of joints, the difference between deciduate and non-deciduate placentation, the direction of flow in the *Vena perinealis*, the status of Metacarpus V in the bovine) it is clear that this mini-skirt has been cut from old-fashioned tweed.

The book is written in the style of abbreviated lecture notes and the material on mammalian anatomy presented system by system. A general description is followed by brief, comparative notes. Usually a single characteristic is given whereby the bones of the various species may be identified; in the case of organs, one or two, occasionally three points of difference are mentioned. Whether the most typical features, from the practical point of view, have been selected in each and every case is a moot point.

Often the general description is based on the anatomy of the horse, without always saying so (e.g. ligaments of the fetlock).

Of the skull, only the main regions, the orbital and pterygo-palatine foramina and the paranasal sinuses of the horse and frontal and maxillary sinuses of the ox and mandible and hyoid bone are dealt with.

Only the muscles of the thoracic and pelvic limb of the horse are treated, whereas the arterial and venous systems of the dog only are considered. Of the latter system only the caudal vena cava and main tributaries, the portal vein and main tributaries, and the azygos vein come in for mention. The lymphatic system of only the ox is described and is the best chapter in the book in the reviewer's opinion. The ear has been omitted; under "Eye," the cornea is not mentioned, not even as one of the refractive media of the eye. The word "parasympathetic" never appears. The goat has been left out entirely and the cat only comes in for mention by way of dental formulae, stellate veins of the kidneys and as having a zonary placenta. The anatomy of the fowl is accorded the last five pages in the book.

There are some misconceptions: the issue between simple and compound stomachs on the one hand and uni- and multiocular on the other is confused; the common bile duct is regarded as union of pancreatic and bile ducts; the definition of "Selenodont" is not a happy one. A number of errors has also crept in: the fourth carpal bone of ruminants is indicated as a fusion of fourth and fifth carpals; the deep pectoral muscle is not an adductor of the shoulder joint, nor is the middle gluteal muscle primarily and abductor of the hip joint, nor are the obturator internus and gemelli muscle extensors of this joint; the biceps femoris and gemelli muscles do not arise from the ilium but from the ischium; the foetal lung can hardly be said to be pale grey; the palatopharyngeus muscle is not a muscle of the soft palate (why try to distinguish, in a text of this brevity, between extrinsic and intrinsic muscles of the soft palate —if one wants to do it, surely the M. palatinus is the only one that can be regarded in a sense as intrinsic); there are not only two buttresses, one dorsal and one ventral, arising from the common renal papilla in the sheep; by omission the impression is created that the sow has no suburethral diverticulum; the ileoinguinal and genitofemoral nerves do not supply the testes.

Typographical errors—the bane of authors' and publishers' lives—are particularly irritating in a text of this type, as each word is critical. The following have been spotted:

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bifed (p. 5); root (of pelvis) (p. 12); Gamellus (p. 41—twice, and p. 154; should one not rather stick to the comparative anatomical concept and speak of mm. gemelli?); rakes instead of takes (p. 131); **cilated** epithelium (p. 135); glands of humeral secretion! (p. 142). The words INTERNAL CAROTID and OCCIPITAL have been omitted (p. 52).

Then there are a number of statements which, although not overtly wrong, are so worded that they may give rise to misconception in the students' mind. Usually it is a problem inherent in the brevity of style. Some examples may be quoted. The olfactory nerve is said to pass through the ethmoidal foramen, the opening of the ureter to be valved, the hock joint to consist of four synovial sacs, the mare's placenta, *by implication* is diffuse and complete; similarly the iliohypogastric and ilioinguinal nerves belong to the last three of the lumbar group, only in the dog has the greater omentum to be removed to see the abdominal organs (including the liver!), and the horse has a tonsillar sinus. One may raise one's eyebrows at the description of the ovulation fossa, of the trigone of the bladder and the anterior and posterior limits of the equine frontal sinus.

This book focuses attention sharply on the greatest dilemma in the teaching of veterinary anatomy: extent of coverage. In this respect one can argue till the cows come home whether the author has really succeeded in "cutting out all the deadwood" (why cling to the traditional anatomical liturgy of so many sides, surfaces, borders, unless they have specific names?) and whether he has not thrown out the baby with the bath-water. It is doubtful whether this book, with its lack of explanatory matter, will "lay a foundation" as the author claims; only a good teacher, with adequate material and facilities at hand, and aided by more extensive text-books, can do this. As a skeleton for further extension it should be very useful indeed. It is a pity that a blank page for notes does not face every page of text, instead of being inserted at the end of each chapter only.

As "a student's laboratory companion—be he Meat Inspector, Veterinarian or Animal Nursing Auxiliary", and, one may add, "Animal Husbandry Student", as "a means of consolidating knowledge" and as "a review source" it should serve its purpose well.

If it is true that a book such as this is needed to correct the present-day "tendency for veterinary anatomists to skirt the elemen's of their subject in an effort to keep up with the clinical Jonesses," then the teaching of veterinary anatomy (presumably in Britain) is in a sad, sad state.

The type is small, but generously spaced; unfortunately the inking is uneven. The review copy had two back covers.

H. P. A. de B.

YOUR HORSE — A VETERINARY BOOK

ANON. VETERINARY CORRESPONDENT TO SPORTING LIFE

Published by "The Sporting Life," 1968. Longacre, London, WC 2.

pp 149. Publ. price 21/-.

This volume originated as a series of articles by a veterinarian writing in a British magazine, "The Sporting Life." As such, its aim is to promote understanding of the horse in the modern age, and is intended for those connected with horses in a non-veterinary capacity, e.g. owners, breeders, trainers and studmen.

Emphasis is placed chiefly on the thoroughbred, but most of its contents may be applied to all other breeds of horses. The author systematically describes all the facets of present-day stud procedures, and stresses the importance of a close liaison between stud manager and veterinarian.

The first nine chapters are devoted to conditions of the limbs, are suitably illustrated and cover conformation, joints, tendons and fractures. Breeding, infertility, infectious diseases and parasites are adequately dealt with. Treatments are not described in great details as the value of veterinary assistance is emphasised.

The book will be an aid to veterinary students and to veterinarians as it contains a wealth of practical suggestions not readily gleaned from textbooks. Although intended primarily for the layman, it is a compact and interesting fund of information.

J. M. O'G.

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FLUID BALANCE IN CANINE SURGERY

L. W. HALL

Bailliére, Tindall and Cassell, London, 1967. pp. viii & 117, Figs. 22,

Publ. Price 25s.

Although fluid balance in the normal and in the sick dog has been the subject of numerous articles over many years, comprehensive information in this field could usually only be obtained after spending very many hours in a good library. The need for a succinct work, embodying the essentials of fluid therapy in the dog thus became ever more insistent. The appearance of this small volume is very welcome. The author wisely and lucidly first brings the reader up-to-date regarding acid-base equilibrium, distribution of water, water balance and the relevant physiological features of the electrolytes. He then passes on to disturbances due to body fluid, sodium and potassium depletion and various forms of acidosis and alkalosis. Clinical diagnosis and laboratory tests are then described, followed by the principles of treatment. The next chapter on practical procedures provides valuable information as to ways and means of implementing treatment. The frequency and the importance of shock justify the chapter devoted to it. Towards the end of the book space is given to special problems and selected cases. The last pages are occupied by a pot pourri of useful information like formulae and, finally, recommended laboratory techniques.

The book is well written and the information strictly relevant. As such it is recommended to senior undergraduates and to clinicians.

C.F.B.H.

TEXTBOOK OF MEAT INSPECTION

HORACE THORNTON, 5TH ED.

Bailliére, Tindall and Cassell, London, 1968. pp vii & 596, Figs. 238,

Colour plates 16, numerous Tabs., Publ. Price 75s (R7.50)

The first edition of this well known book was published nineteen years ago, and this latest edition needs no introduction to the many who have come to rely on previous editions over so many years. The appearance of a new edition is proof of the continued demand for this work, and is undoubtedly the result of the authors' maintained interest in the field of meat hygiene, extended as it has over a lifetime of work in so many countries of the world. It is perhaps this broad approach, not confined to the conditions and legislation of one country only, which is responsible for the widely accepted value of this book.

The author and co-author have added considerably to both the text and the illustrations, and have brought the book up to date by revising existing and adding new sections on abattoirs and meat hygiene, sanitation in the abattoir, the role of the abattoir in the control of diseases, sources of contamination of meat, the assessment of meat quality and the effects of preslaughter handling on meat. The co-authors' chapter on chemical residues in meat deals with this increasingly important aspect of meat hygiene in an authoritative way, thereby including in the sphere of interest and concern of the meat hygienist some of the man-made problems of this modern technological world.

This standard work now includes chapters on abattoirs and methods of slaughter, post mortem inspection, comparative anatomy, uses of organs as food, pathology of food animals, bacterial and parasitic infections, the preservation and bacteriology of meat, the processing of by-products and the inspection of rabbits, hares and poultry. The book more then justifies its existence on the desk or in the lecture room of all concerned with meat hygiene in its broadest sense of the word. It now represents a most comprehensive text for reference and study, and can be strongly recommended.

This latest edition is again a credit to the publishers; the paper, printing, binding and reproductions (black and white, line drawings and colour plates) leave nothing to be desired. I have only two criticisms. The

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absence of any references is bound to be felt by those readers who wish to delve more deeply into certain aspects; it is however fully appreciated that inclusion of references would add materially to the cost and size of a publication which already extends over more than 600 pages. A more serious criticism relates to what might be called a case of anachronism: a new edition dealing with a variety of recent developments in the field, but wherein one still encounters rather antiquated and certainly no longer internationally recognised names for some micro-organisms such as *Staphylococcus pyogenes aureus, Fusiformis necrophorus* and *Salmonella suipestifer*. Synonyms are sometimes given in brackets, and these are usually even older names. This is not in keeping with the rest of the book. It will undoubtedly confuse the younger generation and tend to perpetuate these now extinct names in the minds of the older readers. It is hoped that this matter will be corrected in a following edition.

L. W. v. d. H.

ATLAS OF RADIOGRAPHIC ANATOMY OF DOG AND HORSE

H. SCHEBITZ AND H. WILKENS

Verlag Paul Parey, 1968. Berlin and Hamburg. Pp. 198, 94 Radiographs, 94 line drawings (7 colour), 72 positioning drawings. Publ. price DM. 148 (approx. R27).

To date, veterinary textbooks on radiology have dealt mainly with radiographic technique and pathology. In spite of being familiar with the contents of these books, even veterinarians experienced in radiological diagnosis on occasion have difficulty in deciding whether certain features on a radiograph are normal or abnormal. When these problems of interpretation arise, the availability of a normal radiograph for purposes of comparison is invaluable.

The authors, a clinician and an anatomist, have collaborated to provide students and practising veterinarians with a comprehensive atlas illustrating the normal radiographic anatomy of the dog as well as the extremities and head and neck of the horse. Not only is the reader provided with an authoritative outline of the anatomy necessary for proper interpretation of radiographs, but he is also thoroughly instructed in the relevant positioning aspects of technique required to attain radiographic images identical in outline to those reproduced in the book. These instructions must be followed meticulously if full benefit is to be derived from this book. Even a slight deviation in positioning alters the radiographic anatomical record.

Ninety-four large radiographs, unsurpassed in their detail, definition and contrast, are reproduced free from any inscriptions, i.e. as they will appear on the illuminator. Each radiograph is accompanied by a comprehensively annotated line drawing of identical dimensions, explaining the anatomical features of the relevant radiograph. The line drawings of the carpal and tarsal joints are done in four colours to facilitate recognition of the component bony structures. Special contrast methods, applicable to the thorax and abdomen, viz. bronchography, pyelography, pneumoperitoneum and barium enemas, to name a few, are also described and illustrated.

A list of the accessory equipment and exposure factors used accompanies every radiograph. In order to attain the high quality radiographs reproduced in their book, the authors made extensive use of high definition screens and a Bucky-diaphragm. Exposures were mostly made from a distance of 120 cm. with reasonably high kilovoltages and milliampere-seconds. The following figures were compiled by the reviewer to serve as a general indication of the kV's and mAS used by the authors:

	Dog		Horse	
	Average	Range	Average	Range
mAS	29	7—58	53	10— 98
kV	66	45—75	70	50—115

Veterinarians in private practice will in all probability find that their X-ray units are incapable of delivering the same kV and mA outputs, with the result that they will have to compromise. Despite this fact, the book can nevertheless be recommended to private practitioners who will find it invaluable as a reference work with which to compare their own radiographs.

The text is in German and English. A full list of contents and an extensive bibliography is provided.

This is an excellent book with outstanding illustrations and definite practical value. Both the authors and the publishers can be justly proud of this publication. The companion volume in preparation, covering all the other domestic animals, is eagerly awaited.

C. J. R.

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QUIZ / VASVRA

DO YOU KNOW THIS ONE? KEN U HIERDIE EEN?

Kalf-fetus, geaborteer of voltyds gebore. Matige hidrokefalus; min of meer spits toelopende bokaak wat sy- of ventraalwaarts verbuig kan wees; gesplete verhemelte; baie kort onderkaak; snytande verdruk en verminder in getal; meesal prominente oë en langgerekte oogspleet; kort, asimmetriese oorskulpe; ledemate distaal van humerus/femur afwesig of slegs as afgeknotte radius-ulna/tibia teenwoordig; soms rudimentêre kloutjies.

Calf foetus, aborted or carried to-term. Moderate hydrocephalus; more or less pointed upper jaw which may be deviated sideways or ventrally; cleft palate; very short lower jaw; incisors compressed out of position and decreased in number; usually prominent eyes and elongated palpebral fissure; short, asymmetrical ears; limbs absent distal to humerus/femur or represented by stumps of radius-ulna/tibia; sometimes rudimentary claws.

Acroteriasis congenita (Lethal factor A5) also known as "amputated" or "otter calves." Inherited as a simple autosomal Mendelian recessive. Thus far recorded in Fries cattle.

Please report such cases to Prof. de Boom, if possible forward specimen, and give full ancestry.

Acroteriasis congenita (Letale faktor A5) ook bekend as „amputated" of otterkalwers oorgeërf as 'n enkelvoudige outosomale Mendeliaanse resessief. Tot dusver in Friesbeeste aangeteken.

Rapporteer asseblief sulke gevalle aan Prof. de Boom, stuur monsters in indien moontlik, en dui volle voorgeslag aan.

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