JOURNAL
OF THE
SOUTH AFRICAN
VETERINARY
ASSOCIATION



TYDSKRIF
VAN DIE
SUID-AFRIKAANSE
VETERINÊRE
VERENIGING

VOLUME 43 No. 4

DECEMBER/DESEMBER

JAARGANG 43 Nr. 4

1972

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#### CHANGE OF EDITORIAL POLICY

Will intending contributors please note that henceforth all references must:

- (a) be listed in alphabetical order according to the surname of the first author. (The superscript numeral system is retained, but the references must no longer be numbered in order of appearance in the text, but according to their alphabetical order);
- (b) give the full title of the article to which reference is made.

#### WYSIGING VAN REDAKSIONELE BELEID

Sal alle voornemende bydraers asseblief kennis neem dat voortaan alle verwysings-

- (a) in alfabetiese volgorde moet verskyn, volgens die eerste outeur se van. (Die boskrifnumereringstelsel word behou, maar verwysings word nie meer genommer volgens orde van aanhaling in die teks nie, maar volgens alfabetiese volgorde);
- (b) die volle titel van die artikel, waarna verwys, moet dra.

#### **CORRIGENDUM**

The legends to the two figures on page 264 of the Journal (No. 3 — September issue) should be transferred seriatim to the two histograms on page 269. In their place the following captions should appear:—

- Fig. 1. Colon showing leucocytic infiltration of lamina propria and submucosa. 75 ×
- Fig. 2. Higher magnification of colon showing primarily eosinophil infiltrate and crypts of Lieberkühn distended with mucus. 150 ×.

## JOURNAL OF THE SOUTH AFRICAN VETERINARY ASSOCIATION TYDSKRIF VAN DIE SUID-AFRIKAANSE VETERINÊRE VERENIGING

THE JOURNAL OF THE S.A.V.A. is owned and published by the South African Veterinary Association, of which it is the official organ. It appears quarterly and is devoted to matters of veterinary importance generally.

The statements made and opinions expressed by contributors to this Journal are their responsibility only; such statements are not necessarily endorsed by the Editorial Committee, neither do their opinions reflect those of the Committee.

SUBSCRIPTION — A free copy of each issue is sent to all Members of the Association in good standing. The subscription rate for non-members is R10.00 per annum, post free surface mail.

BACK NUMBERS are obtainable at R3.00 per number.

CONTRIBUTIONS — The Editor will consider contributions of veterinary interest. Double spaced, carefully revised, typewritten manuscripts should be submitted in triplicate (original plus first two copies). Layout and references should be in the style of this number. The number of figures and tables may be limited at the Editor's discretion unless the author contributes to the cost of reproduction. Complete titles of references must be given.

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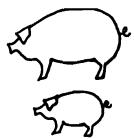
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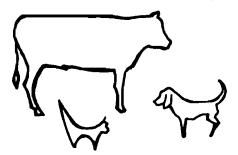
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#### REDAKSIONEEL

#### EDITORIAL

# DIE VEEARTS EN BEHEER OOR ARTSENYMIDDELS

#### Besorgdheid oor dwelmmiddelverslawing. kwaadwillige gebruik en agtelosige hantering van giftige stowwe, artsenymiddelresidue in voedsel van dierlike oorsprong, bakteriële weerstandopbouing teen middels en artsenyverwekte ensiemversterking of -remming is die wêreld oor aan die toeneem. Verskeie wette en wetsontwerpe op beheer artsenymiddels het onlangs by die Parlement van die Republiek gedien of staan ingedien te word. Uit hoofde van sy professionele status geniet die veearts die voorreg om die hele reeks geneesmiddels, wat hy in die beoefening van diergeneeskunde nodig mag hê, aan te skaf, te besit, saam te stel, te resepteer, toe te dien of voor te skrywe. Hierdie stowwe kan giftig, skadelik, potenskadelik of afhanklikheidswekkend wees, en groot verantwoordelikhede is dus verbonde aan besit van hierdie voorreg.

Die Professie het in hierdie opsig altyd op verantwoordelike wyse opgetree, maar die noodsaak om hierdie reputasie te handhaaf kan nie oorbeklemtoon word nie. Onlangse en voorgestelde wetgewing is meer ingewikkeld en vereis gedurige herinnering van onsself en van anderes aan ons professionele verbintenis.

Dit is trouens een van die algemene verpligtings van 'n veearts om hom ten volle op hoogte te stel (en te hou) van wette wat hom en sy werk raak. Op hierdie tydstip kan alle lede van die professie voordeel trek om juis dit te doen aangaande die beheer oor artsenymiddels. Die Wet op Misbruik van Afhanklikheidsvormende Stowwe Nr. 41 van 1971 maak voorsiening vir besonder swaar strawwe vir enigeen, insluitende professionele persone, wat onder hierdie Wet aan misdryf skuldig bevind word. Dit dien onthou te word dat dit bowendien die uitdruklike verpligting van die veearts is om met die owerhede saam te werk en om die Regering en sy beamptes, sover hy by magte is, te help om die wette ten uitvoer te bring.

# THE VETERINARIAN AND THE CONTROL OF MEDICINE AND DRUGS

Throughout the world man is becoming increasingly concerned over matters such as drug addiction, malicious use and careless handling of toxic substances, drug residues in food of animal origin, bacterial drug resistance and drug induced enzyme potentiation or inhibition. Various Acts and Bills dealing with medicine and drug control have recently come before the Republic's parliament or are due for early consideration. By virtue of his professional status the veterinarian enjoys the privilege of acquiring, possessing, compounding, dispensing, administering or prescribing the full range of medicinal substances which he may require in practising veterinary medicine. These substances may be poisonous, harmful, potentially harmful or dependency producing, and great responsibilities therefore accompany possession of this privilege.

The profession has always acted responsibly in this regard, but the need to maintain this reputation cannot be overemphasized. Recent and proposed legislation is more complex and requires that we constantly remind ourselves (and others!) of our professional obligations. It is, in fact, one of the General Obligations of a Veterinarian to 'make (and keep) himself fully conversant with the laws affecting himself and his work'. At this point in time all members of the profession could benefit from doing just that as far as medicine and drug control is concerned. The Abuse of Dependency Producing Drugs Act No. 41 of 1971 makes provision for extremely heavy penalties for anyone, including professional persons, found guilty of an offence under this Act. It is also worth recalling that it is also the specified obligation of a veterinarian to co-operate with the authorities and to assist, as far as lies within his power, the Government and its officials in carrying out its laws.

Wet 36 van 1947 is in 1970 gewysig om voorsiening te maak vir die registrasie as veemiddels van stowwe wat ,geskik en effektief genoeg is vir die doel van gebruik' met dien verstande dat registrasie ,nie teen openbare belang indruis nie'. Sodanige geregistreerde middels is vryelik sonder voorskrif bekombaar en dit moet onthou word dat stowwe wat nie dusdanig geregistreer is nie, deur die owerhede as ongeskik vir onbeperkte gebruik sonder geskikte toesig geag word. Die aanbeveel van ongeregistreerde stowwe of formulasies, of die voorstel vir hul aanskaffing en gebruik van vee-eienaars, behalwe waar dit geresepteer of voorgeskrywe word ooreenkomstig die Wet op Geneeshere, Tandartse en Aptekers Nr. 13 van 1928, kan dus as onprofessionele gedrag beskou word.

Namate beheer verskerp word, is daar dikwels sterker druk op veeartse om voorskrifte blindelings uit te reik. Dit kan nie sterk genoeg beklemtoon word nie dat die Wet op Geneeshere, Tandartse en Aptekers gevolmagtigde veeartse toelaat om middels te besit, te resepteer en voor te skrywe in die loop van wettige uitoefening van sy beroep ,vir 'n dier onder sy sorg en behandeling'. Genoemde Wet word nou hersien en voorgestelde wysigings maak voorsiening dat 'n ,veearts' (as sodanig by die Veeartsraad geregistreer) middels en medisynes Groepe B tot G geklassifiseer) onder sekere voorwaardes vir veeartsenykundige doeleindes mag aanskaf, besit, saamstel, resepteer, toedien en voorskrywe, m.a.w. die eienaar en die dier, waarvoor die middel geresepteer of voorgeskrywe word, moet gespesifiseer word. 'n Verdere vereiste is dat ,'n veearts sulke geskeduleerde stowwe mag verkoop of verskaf slegs vir die behandeling van 'n dier onder sy sorg in die loop van sy praktyk'. Dit beteken nie dat 'n veearts noodwendig elke dier moet sien en ondersoek alvorens medisyne te resepteer of voor te skryf nie. Indien 'n bekende kliënt 'n veearts sou nader omtrent 'n probleem en die veearts kan homself tevrede stel insake die diagnose en die beste behandeling omrede vorige kennis van toestande en siektes op die plaas, asook omtrent die bekwaamhede van die vee-eienaar, sou hy sy oordeelvermoë gebruik en nog verantwoordelik optree en die diere sou nog onder sy sorg en behandeling val. Dit is die voorneme dat hierdie wysigings aan die Wet op Geneeshere, Tandartse en Aptekers in die gewysigde Wet op Beheer oor Medisyne Nr. 101 van

Act 36 of 1947 was amended in 1970 to provide for registration as livestock remedies of substances that are 'suitable and sufficiently effective for the purpose intended' provided that registration 'is not contrary to the public interest'. Such registered remedies are freely available without prescription and it should be borne in mind that substances not so registered are considered by the authorities to be unsuitable for unsupervised and unlimited use. Recommending or suggesting to stock owners the acquisition and use of unregistered substances or formulations (except where dispensed or prescribed in terms of the Medical, Dental and Pharmacy Act No. 13 of 1928) may therefore be considered as unprofessional conduct.

As control is tightened there is often increased pressure on veterinarians to issue prescriptions. It cannot be overemphasized that the Medical, Dental and for Pharmacy Act provides 'authorized veterinarians' to possess, dispense prescribe drugs in the course of the lawful exercise of his calling 'for an animal under his care and treatment'. The said Act is now under review and proposed amendments provide for a 'veterinarian' (i.e. registered as such with the Veterinary Board) to acquire. possess, compound, dispense, administer and prescribe drugs and medicines (classified in Groups B to G) for veterinary purposes under certain conditions, i.e. the owner and the animal for which the drug was dispensed or prescribed must be specified. Another requirement is that 'a veterinarian may sell or supply such scheduled substances only for the treatment of an animal under his care in the course of his practice'. By this is not meant that a veterinarian must of necessity see and examine each and every animal before dispensing or prescribing drugs. If a known client were to approach his veterinarian about a problem and the veterinarian could satisfy himself as to the diagnosis and the best treatment because of previous knowledge of conditions and diseases on the farm as well as the capabilities of the livestock owner, he would be using his discretion and still be acting responsibly, and the animals would still be under his care and treatment. It has been proposed that these amendments to the Medical, Dental and Pharmacy Act be incorporated into the amended Drugs Con1965 ingelyf word. U Vereniging ondersteun hierdie wysigings en het die professie tot instemming daartoe verbind.

Indien ons ons rol in die rasionalisering van gebruik van middels in diergeneeskunde en veeteelt gaan vervul, en indien ons aan die beperking van sekere stowwe tot professionele gebruik of gebruik slegs onder veeartsenykundige toesig glo as 'n manier om wangebruik, omgewingsbesoedeling en blootstelling van die mens aan skadelike stowwe te minimiseer, kan ons nie anders as om die groter veiligheid in bovermelde opsigte te verwelkom nie. As ons ons voorregte as professie hoog skat, moet ons ook strenge neerlegging by die wet verseker en nie toelaat dat ons onsself in enige mate tot eksploitasie blootlê wat skadelik sal wees teenoor diegene teen wie ons 'n verpligting het, t.w. die vee-eienaar en -bedryf, die algemene publiek, en, natuurlik, onsself.

Voorgestelde Wysigings van die Wet op Beheer van Artsenymiddels 1965 (Nr. 101 van 1965) om Beheer oor "Geskeduleerde Middels" te verleen.

\*=Op voorskrif.

Skedule A: (Vir gebruik, ens., slegs op permit van die Sekretaris van Gesondheid.) Amfetamiene, cannabis en derivate.

\*Skedule B: Asetorfien, kokaïen, kodeïen, dihidromorfien, metadoon, morfien, opium, petidien, ens.

\*Skedule C: Barbiturate, ens.

\*Skedule D: Barbituursuur, ens.

\*Skedule E: Antibiotika (sinteties en natuurlik), hormone, sulfonamiede, fenielbutasoon, ens.

\*Skedule F: Alkaloïede en glikosiede, digitalis, atropien, apomorfien, emetien, arseen, antihistamiene, chloroform, chloraalhidraat, lignokaïene, strignien, blousuur, ens.

\*Skedule G: Trichlooretileen, fenasoon, loodasetaat en pleisters, eter, asetanilied, ens. trol Act No. 101 of 1965. Your Association fully supports these amendments and has committed the profession to compliance.

If we are to play our part in the rationalisation of the use of drugs in veterinary medicine and animal husbandry, and if we believe in the limitation of certain substances to professional use or under veterinary supervision as a means to minimize abuse, environmental pollution and the exposure of man to harmful substances, then we cannot but welcome greater security in this area. If we value our privileges as a profession, we must also ensure that we comply strictly with the legal requirements and do not allow ourselves to be exploited in any way which will be harmful to those to whom we have an obligation, i.e. the livestock owner and industry, the public at large and, of course, ourselves.

Proposed Amendment of Drugs Control Act 1965 (No. 101 of 1965) to Provide for Control of 'Scheduled Drugs'.

\*=on prescription.

Schedule A: (For use, etc., only under permit from Secretary for Health).

Amphetamines, cannabis and derivatives.

\*Schedule B: Acetorphine, cocaine, codeine, dihydromorphine, methadone, morphine, opium, pethidine, etc.

\*Schedule C: Barbiturates, etc.

\*Schedule D: Barbituric acids, etc.

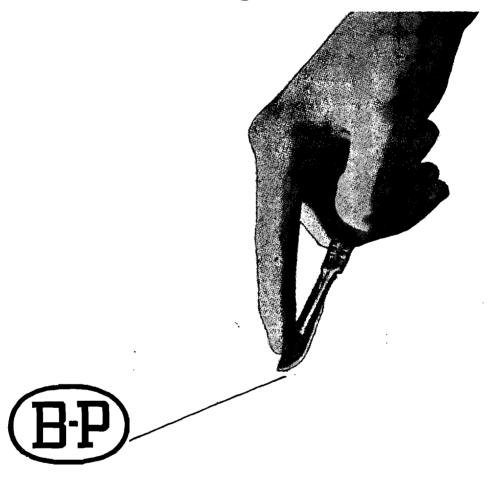
\*Schedule E: Antibiotics (synthetic and natural), hormones, sulphonamides, phenylbutazone, etc.

\*Schedule F: Alkaloids and glycosides, digitalis, atropine, apomorphine, emetine, arsenic, antihistamines, chloroform, chloral hydrate, lignocaines, strychnine, hydrocyanic acid, etc.

\*Schedule F: Trichlorethylene, phenazone, lead acetate and plasters, ether, acetanilide, etc.

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**REVIEW** 

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IEW OORSIG

# A REVIEW OF SOME ASPECTS CONCERNING MATING AND REPRODUCTION IN SHEEP\*

E. M. VAN TONDER\*\*

#### SUMMARY

Various factors concerned with successful mating and reproduction are reviewed with reference to the available literature.

The different mating practices employed in small stock farming are discussed with evaluation of their particular advantages and disadvantages.

Practical aspects on mating management conducive to successful mating are given.

#### INTRODUCTION

The domestication of our farm animals and their exploitation have inevitably lead to practices aimed at increasing production and reproduction. The latter has attained a significance far beyond the level required for the survival of a particular breed. Successful mating is an absolute and basic prerequisite for maximal reproduction. As far as our small stock is concerned it is limited to the natural breeding season and is subject, furthermore, to specific environmental, climatic and other conditions. Specific systems of mating practice have been evolved. These include selection of suitable mating and lambing seasons, inducement of maximal sexual activity and fertility, handling of ram and ewe flock and control of the actual mating process, aspects which are often ignored when considering the causes of lowered reproduction.

A general outline of the various natural mating practices employed in the Karoo and adjacent mixed and grassveld areas will be given with reference to the available literature and will include personal observations and experience. Although emphasis will be placed on the Merino breed, the same considerations are applicable to small stock farming in general.

REVIEW OF REPRODUCTIVE PHENOMENA

Puberty in the ewe

Puberty is determined by the commencement of full sexual activity. In the female animal it is characterized by production of ova and manifestation of oestrus. In animals with unrestricted sexual activity throughout the year, puberty would be determined by the age at which oestrus is exhibited for the first time.

In animals with defined annual breeding seasons, signs of puberty will not be observed before the breeding season is reached, although they might be physiologically competent beforehand. Age and mass could possibly be used as indicators of such competence: Hafez² found no difference in live masses at the time of their first oestrus between the Suffolk lambs born early or late in the lambing season, provided that the difference in age did not exceed the limits of the breeding season. Nevertheless, the first noticeable heat period remains the only practical means of determining the age of puberty in ewes.

Others factors apart, the variation between the dates of birth in relation to the subsequent breeding season was probably the main reason for the great difference in age of puberty observed in sheep: Watson & Gamble 3 found that Merino ewe lambs born in spring, summer or winter reached puberty in the first breeding season at mean ages of 6, 12 and 9 months respectively.

In South Africa, earlier workers be observed the first heat in Merino ewes at the age of 9 to 10 months. Roux be determined that only 40 per cent of Merino ewes reach puberty at the age of 19 months under Transvaal conditions, while in the Karoo 50 per cent

\*\*Regional Veterinary Investigation Centre, Middelburg, Cape.

<sup>\*</sup>Paper presented at the Biennial Scientific Congress, South African Veterinary Association, East London, September, 1971.

do so between the age of 12 to 17,5 months. Hugo 6 established that 83 per cent of Merino ewe lambs born in autumn reached puberty in the first post-natal season at the mean age of 11.5 months, as against 17 per cent of the spring lambs, which did so at the mean age of 9 months. In the second season following birth, the remainder of the autumn lambs and 75 per cent of the spring lambs came on heat. Thus 92 per cent of the springborn lambs attained puberty in the second season at a mean age of 16 months: the majority was too young at the first season. There is a considerable variation between individual sheep: the youngest age of puberty observed by Hugo 6 was 7 months (200 days). Puberty in Rams

Puberty in rams is related to age and more closely to body mass <sup>7</sup>. It usually occurs when 40—60 per cent of the mature body mass has been attained. In Merino ram lambs, sperm were first observed at a mean age of 191 days and live mass of 63 lbs. These figures are comparable with those reported by Dun. Complete descent of the testes and penis development were found to occur at the mean ages of 5 and 6 months respectively. These figures were more or less the same for ram lambs on high and medium feeding levels while those in the low level group were considerably tardier. Sexual Maturity

Sexual maturity represents that age, at which the animal can be bred successfully for the first time without permanent harmful effects on growth and reproduction; it obviously occurs for a variable length of time after puberty. It is usually advisable to postpone breeding until well after puberty, although it should not be delayed too long 10. The consequences of breeding animals too early are clearly demonstrated by Spencer et al". Although the ewes mated as lambs produced more lambs over the whole period, their average lambing percentage up to the fourth year was distinctly lower than those mated as yearlings, despite excellent nutrition.

The age of sexual maturity is greatly influenced by the breed of sheep as well as the nutritional level during growth and development. While studying the breeding histories of prolific Merino ewes from various studs, Roux 5 determined that 90 per cent of the ewes were mated at the age of 18—24 months for the first time and therefore re-

commended that maiden ewes should be mated as soon as they are 18 months old.

In more recent studies 6, it has been found that, in an autumn mating practice, springborn Merino ewes can be mated at the age of 16 months.

Although rams can be used at a relatively young age, especially under excellent feeding conditions, this is not recommended. Two-tooth (14—18 month-old) Merino rams are sometimes used, particularly for progeny testing but the most usual and recommended age is from 18 to 20 months.

#### The Breeding Season

It is well known that in most animals sexual activity is restricted to more or less defined breeding seasons. This is apparently aimed at delivering the young at a time most favourable for development and growth.

The control of the breeding season by an internal rhythm under the influence of light is abundantly substantiated in the relevant literature. In contrast to most animals, in which increased daylight irradiation acts as the most important stimulus for sexual activity <sup>12</sup>, sheep respond more actively to a decrease in light <sup>13</sup>. A regular rhythm of alternate shorter light and longer dark periods seems to stimulate the occurrence of oestrus rather than an actual decrease in the light intensity <sup>14</sup>.

Working with British breeds of sheep, Yeates 15, supported by Hafez 2, determined that the breeding season ranged from September to March, with the peak corresponding to the shortest day. Subsequently these authors 15-18 proved beyond doubt that, in some British breeds at least, sexual activity was markedly influenced by a change in duration of hours of daylight. Yeats 19 and Watson 20, working on Merinos, found similar responses to changes in duration of daylight hours. The occurrence of oestrus in a certain percentage of ewes during a period of increasing length of day caused Watson & Radford 21 to doubt control by decreasing light and to conclude that their effective light-dark regime must have been different from that in the previous studies 14-18. Radford <sup>22, 23</sup> furthermore, considered alternative suggesions for photoperiodic control of sexual activity, including a possibility of a suppressing mechanism during anoestrus rather than

a stimulation of activity during the sexual season. He concluded that factors other than light may play as equally important a rôle in controlling sexual activity. Neverthess, the studies referred to as well as additional and subsequent reports on studies on British breeds <sup>24</sup>. <sup>25</sup>, Merinos <sup>25-29</sup> and various breeds on a comparative basis <sup>50-32</sup> afford overwhelming evidence of the seasonal nature of sexual activity in sheep and its photoperiodic control.

Although Quinlan & Maré <sup>33</sup> were convinced that the Merino in South Africa shows sexual activity throughout the year, sufficient evidence to the contrary was provided by Kupfer <sup>34</sup> and Roux <sup>5</sup>. Hugo <sup>6</sup> made a close study of the reproductive pattern of various breeds and cross-bred sheep; he confirmed beyond doubt the seasonal incidence of sexual activity of sheep in this country. He established a sexual year ranging from October to September on the calendar year, with a peak during May: maximal increase in sexual activity corresponded to the sharpest decline in length of day. A graphic representation of the breeding season in three different breeds is given in the figure.

of the sexual year (possible number of 16,8 day cycles per sheep during the sexual year) does not reach 40 per cent.

Although no significant seasonal influence on the duration of oestrus could be demonstrated, there was a tendency to increased duration correlated with increased sexual activity of the flock <sup>6</sup>.

Apart from a seasonal influence on ovarian activity as such, there is also conclusive evidence of a seasonal tendency in ovulation rate. The occurrence of twin and multiple ovulations as opposed to single ovulations, closely follows the pattern of the breeding season <sup>20</sup>, <sup>35,40</sup>.

In rams subjected to artificially decreased light, Moule 1 demonstrated strongly increased libido. Personal observations strongly support this work. While reasonable difficulty is experienced in selecting Merino rams interested in artificially stimulated but fully receptive ewes during spring (September—October), no problems are encountered with the same batch of rams with ewes with synchronized oestrus during the breeding season.

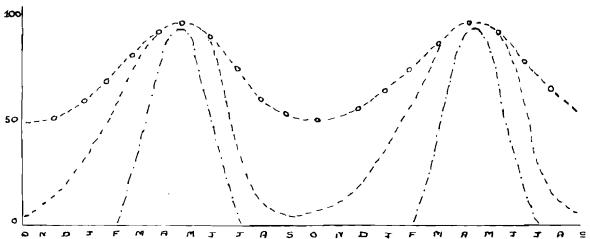


Fig.: Graphic representation of the sexual season of various breeds. (Adapted from Hugo, 1955). O---O Blackhead Persian. ---Merino. ----British mutton breeds.

It should be borne in mind that the breeding season of a particular flock is not representative of the duration of the breeding season of its individual members but merely the collective result of relatively short individual seasons distributed over a longer period. The mean duration of the breeding season (number of oestrous cycles) of individual sheep in relation to the length

Concerning seasonal changes in semen production, Moule <sup>52</sup> suggested that these probably occur in all species breeding seasonally. Decreases in motility, concentration and total numbers of sperm and increases in the proportion of dead and/or morphologically abnormal sperm outside the breeding season were also reported by various workers <sup>17, 26, 43,46</sup>.

#### Breed Differences

Domestication, by way of selection, improved shelter, nutrition and disease control, causes an adjustment of breeding habits: a major effect is extension of the sexual season <sup>47</sup>. Nevertheless, the limited sexual season of primitive breeds is regarded as being dominant over the longer season of improved breeds <sup>13</sup>.

Distinct differences in the duration of the breeding season exist between various breeds of sheep 16, 21, 25, 30, 31, 38. In South Africa Roux 5 and Kupfer 34 classified the different breeds in order of decreasing duration of the breeding season as follows: Blackhead Persian, Ronderib Afrikander and Karakul, Merino and Woolled Persian and lastly the British breeds. Hugo 6 classified the different breeds of sheep into 'short', 'intermediary' and 'long season' breeds (see Figure). He also estimated the mean period of sexual rest in the Merino as 220 days, as opposed to 117 days in the Blackhead Persian. Variations in the duration of the oestrous cycle in the same breed at different periods of the breeding season 6, 40 as well as individual differences within breeds 48, nevertheless make differences between breeds difficult to estimate accurately.

Breed differences in ovulation rate, as evinced by the occurrence of multiple births, are well known and a table of comparative figures has been compiled by Thomson & Aitken <sup>49</sup> and adapted by Hafez <sup>7</sup>.

Only one reference regarding breed differences in semen production could be found in the available literature. Starke <sup>45</sup> pointed out that the Ronderib Afrikander rams used in his experiments produced semen of superior quality to Merino rams, probably due to their younger age.

Repeated personal observations, with which Marlow <sup>50</sup> agrees, indicate that there are distinct differences between various breeds of rams in libido and semen quality, with pronounced manifestation during the summer months. Amongst indigenous breeds, the earlier maturing and long breeding season types are usually superior in semen qualities, libido and mating dexterity, followed by cross-breds, woolled-mutton breeds and Merino.

The mass and age at which puberty occurs relative to adult mass and age are not the same for all breeds of sheep. Roux 5

observed ewes of the faster growing breeds to reach puberty at an early age, whilst the slowly growing breeds did so closer to adulthood. The Blackhead Persian, purebred Ronderib and Welsh Mountain and Ronderib crosses reached puberty at the early age of 5—6 months as against 12 to 17,5 months in Merino ewes. The relative ages at which sexual maturity is attained will obviously follow similar trends in the various breeds. Ewes of the fast growing breeds are usually bred at approximately 10—14 months old.

Puberty in rams also occurs earlier in fast growing breeds than in slower growing breeds, and earlier in cross-bred rams than in their purebred parents 7, 51-53. In practice, mutton and cross-bred rams are normally bred from 12 to 16 and Merino rams at 18 to 20 months of age.

#### FACTORS INFLUENCING REPRODUCTION

As the results to be discussed exert more or less similar influences and effects on both sexes, they will be dealt with in connection with ewes only.

#### Heredity

Although reproduction, like all the production traits, has an hereditary basis, it is of a very low order <sup>54.59</sup>. Selection based on heritability in order to raise the reproduction rate in any particular flock can be aimed at increasing the proportion of ewes lambing or by increasing the number of lambs per ewe lambing.

In the first instance it is based on culling ewes which fail to bear a lamb, either at the first or second occasion. This method, however, was proved to be of little or no value in increasing reproduction rate <sup>56</sup>, <sup>57</sup>, <sup>60,62</sup>. Selection for the incidence of multiple births, although it also has a low heritability value <sup>54</sup>, <sup>55</sup>, <sup>57</sup>, <sup>62</sup> proved to be a more practical and rewarding means <sup>38</sup>, <sup>56</sup>, <sup>57</sup>, <sup>60,62</sup>, not only owing to an increase in the number of lambs but also to an actual decrease in the percentage of dry ewes.

#### Nutrition

Low lambing percentages owing to protein deficiency during the breeding season were reported by Hart & Miller 69 while Hafez 7 and Hugo 6 found reduced oestrous activity in ewes on low levels of feeding during the breeding season. This was reflect-

ed both in the number of ewes exhibiting oestrus as well as the number of oestrous cycles exhibited by these ewes <sup>6</sup>.

As the duration of the oestrous period was significantly reduced when the dietary level decreased from high to medium plane, Hugo <sup>6</sup> concluded that duration of the oestrous period is indeed very sensitive to changes in nutritional levels. These findings are substantiated by results obtained by various workers <sup>40, 64, 69</sup> who also determined greater oestrous activity, earlier onset and increased duration of the breeding season in ewes on higher levels of nutrition. Evidence that increased nutritional levels cause an increased duration of the breeding season was also put forward by Roux <sup>5</sup> and by McKenzie & Terrill <sup>26</sup>.

Although it was stated by Hafez 17 that variations in nutritional level may influence fertility more by converting oestrus to silent heat than by modifying ovulation rate, Wallace 70 demonstrated significant differences in ovulation rate between groups on high, medium and low planes of nutrition. Wodzicka-Thomaszewska & Dobbie n also obtained a 10 per cent difference in twinning rate between two-tooth ewes on a high as against those on a low plane of nutrition. The provision of protein rich, lush feeding to sheep prior to and during the mating season not only increased ovarian activity but also the incidence of twin lambing 72-75. Differences in lambing percentage or percentage of dry ewes between animals on maintenance and higher as against low levels of nutrition have also been reported by Hart  $^{38}$  (up to 15%), Wallace  $^{70}$  (5—10% and 13-22% lambs to ewes mated), Wodzicka-Thomaszewska & Dobbie<sup>71</sup> (5—28%), Inkster<sup>76</sup> (12-15%), Clarke 77 (3-10%) and Coetsee, Vermeulen & Dyason 78 (15%).

The value of improved nutrition in increasing sexual activity and stimulating the onset of the breeding season is manifested in the practice of flushing. This subject has been studied by Hunter 40, 64 and by Smith 65,68 who found that increased levels of nutrition during the preceding winter and spring, apart from a direct influence, also have a delayed effect on oestrous activity persisting for a period of 12 months.

Coetsee, Vermeulen & Dyason <sup>76</sup> reported an increase in lambing percentage of 15 per cent when ewes were flush-fed for a period of three weeks before mating commenced but failed to show beneficial results when they were fed for longer periods or only during the mating period.

The influence of nutrition on puberty and sexual maturity will be evident. Thus, while Quinlan, Maré & Roux & reported puberty at the age of 9 to 10 months in lambs born in autumn and early winter and maintained in good condition, Roux 5 found that, under Karoo veld condtions, 50 per cent of ewes reached puberty at the age of 12 to 14 months, whereas all failed to do so at 18 months with poor nutrition.

The fact that Hugo was only able to demonstrate significantly delayed puberty in low level nutritional animals as compared to the high and medium level groups, led him to state that a decrease in nutritional level beyond a certain critical stage was required before the hereditary tendency towards early puberty will be depressed. In this respect Inkster for proved that well-reared hoggets (weighing on an average of 15 lb more) have 12—15 per cent higher lambing percentages than their poorly reared mates.

With regard to overfeeding, Quinlan & Maré <sup>33</sup> and Quinlan <sup>79</sup> reported the detrimental effect on sexual activity by fat deposition in and around the ovaries. This is supported by Marshall & Hammond <sup>10</sup> and by Barton <sup>80</sup> who regard overfatness as the most important cause of infertility in two-tooth ewes. These observations are contradicted by Wallace <sup>70</sup>, who in consecutive years obtained the highest conception figures at first service as well as lambing percentage in ewes fed on a high level diet from weaning till mating and which he described as excessively or over-fat.

#### Aae

It is generally accepted that fertility in the ewe increases with age up to an optimum age; sufficient evidence to this effect is also available. Kelly <sup>13</sup> estimated that fertility increases until the 8th year and then declines but in a subsequent report <sup>81</sup> stated the age of maximum fertility to be 3,5 years while Terrill & Stoehr <sup>82</sup> suggested an age of 5 years. McKenzie & Terrill <sup>26</sup> reported that the breeding season in sheep is longest in their prime, shorter in older animals and shortest in lambs. This is supported by Hafez <sup>2</sup>, although he did not indicate that

it becomes shorter once a certain age was reached.

Hugo 6 indicated that the number of ewes exhibiting oestrus reached a peak at the early age of 19 months, while the average number of oestrous cycles of the flock gradually increased. The optimal age at which maximal sexual activity (number of cycles per ewe as against the possible maximum) was reached, was calculated to be at 5 years, while the maximum length of the breeding season (mean number of oestrous cycles) was attained at 4,5 years.

Based on the lambing percentages of a flock, the results obtained by Spencer et al 11 and Hart 38 also indicate that the age of maximum fertility of a flock centres around an age of 5 years and that increased ovulation rate with advancing age was at least partly responsible for the increased lambing percentage.

Further evidence of increased fertility with age is supplied by Watson & Gamble <sup>3</sup> who in the corresponding next season obtained conception rates of 63, 95 and 50 per cent respectively in young lambs born in spring, summer and winter.

The influence of age on puberty concerns primarily the date of birth in relation to the onset of the first post-natal breeding season. This has already been discussed.

#### Climate

The fact that animals will breed throughout the year in parts where more even climatic conditions prevail, seems to indicate that apart from nutrition, climatic conditions are also involved. It was determined by Johnson 83 that such parts are also more densely populated with sheep.

Marshall <sup>34</sup> indicated that Scottish blackfaced sheep exhibit only two oestrous cycles on the Scottish highlands as compared to low-lying areas, where the breeding season is substantially longer. Exposure of sheep to low temperatures depresses their sexual activity <sup>84</sup>. <sup>85</sup>.

The seasonal incidence of the breeding season in Merino ewes in the Orange Free State as compared to their uninterrupted sexual activity in central Europe was ascribed to a difference in climatic and nutritional conditions <sup>34</sup>.

Although a depressive action of hot summer condtions on sexual activity was reported by Asdell <sup>1</sup>, subsequent evidence by various workers <sup>19, 27, 86, 87</sup> seems to indicate that high ambient temperatures interfere with fertilization and cause embryonic loss rather than depressing sexual activity. In this respect, Moule <sup>87</sup> also demonstrated a negative correlation between high ambient temperatures and low lamb marking percentages.

#### Wool Production

The influence of increased wool production on reproduction was clearly demonstrated by Hugo 6, who found that higher wool production decreased duration of the breeding season and oestrous period and retarded the onset of puberty. Bosman 88 showed that well developed highly productive Merino ewes yielded lower lambing percentages than their less developed, lower productive counterparts.

In various experiments, Iinkster 76 obtained differences in lambing percentages ranging from 16 to 17 per cent in Merino ewes shorn prior to mating as compared to unshorn ewes, as well as a delayed lambing period in the unshorn ewes. In subsequent experiments he ascribed the difference in lambing percentages to lower lamb mortality in the shorn group. Similar results have been reported by Dun 89 and Drinan & Dun 90 who concluded that the difference in lambing percentage and twinning rate in favour of less developed ewes was more pronounced in two-tooth than in mature ewes. The advantage in twinning rate in less developed ewes dwindled when twinning rates were high.

#### Parturition and Lactation

Roux 5, in support of Anderson 91, found that Merino ewes do not exhibit oestrus while suckling their lambs, although ovulation may take place. He estimated average intervals of 154 and 201 days between lambing and recurrence of oestrus in ewes that lambed in September and in July-August respectively. According to Quinlan Maré 33, ovulation takes place 10 to 15 days post partum without signs of oestrus but oestrus will occur within 17 days if the lamb is removed. Granger 92 stated that a number of ewes conceived within 29 to 61 days when joined by rams after lambing down.

In more recent experiments <sup>93</sup> on ewes under optimal conditions post partum oestrus was observed at an average of 68,7 days in ewes that lambed during July-August. In 90 per cent of these Merino ewes oestrus occurred before the lambs were weaned at 90 days.

#### Mating Behaviour

The stimulating effect of the presence of rams on ewes prior to and at the onset of the breeding season has been widely studied.

Coleman <sup>94</sup>, <sup>95</sup> reported earlier onset of the breeding and lambing season when vasectomized rams were first put to the ewes, while Radford & Watson <sup>96</sup> advanced the onset of the breeding season by one week when teasers joined the ewes before December.

Riches & Watson <sup>97</sup> obtained no beneficial effect by allowing rams to run with the ewes continuously instead of putting them to the ewes for short intervals only, while Watson <sup>98</sup>, subsequently supported by Edgar & Bilkey <sup>99,101</sup>, concluded that the presence of vasectomized rams two weeks prior to mating the ewes with fertile rams had no advantage over a system of putting fertile rams to the ewes from the start.

Ewes which had some previous association with rams, exhibited oestrus within 17 days on being joined by intact rams as compared with other ewes which did so between the 18th and 26th day <sup>102</sup>. Direct contact was not essential as smell and sound had the necessary stimulating effect <sup>102</sup>. Intensive treatment or mustering improved the breeding performance of two-tooth ewes only <sup>103</sup>.

In many instances the ewe on heat seeks male company, follows him around, butting others when more ewes are on heat 104. The variation in number of matings per ewe is determined by a variety of factors, such as duration of oestrus, anatomical fit of ram and ewe, tendency of ewe to stay with the ram, her standing attitude and stimulus value. During an average heat period, a ewe was teased 18 times, mounted 25 times and mated 6 times. The number of matings averaged 3,9 for the first half and 2,4 for the second half of the oestrous period. As the number of ewes on heat increased, the number of teases, mounts and matings decreased as well as the interval from the first to the last mount.

Studies on mating behaviour of rams 105-107 revealed pronounced differences between individual rams. The most important factors affecting mating behaviour were the number of ewes on heat and the individual differences in rams. The number of times a ewe was mated after an initial drop remained constant and the number of mounts per mating was unaffected by the number of ewes on heat.

Marincowitz & Pretorius 108 classified rams into eight groups in order of sexual dominance and mating dexterity. They observed that the number of ewes served increased and the number of attempts and time taken to copulate decreased with an improving order of dominance. Under veld conditions the superior types of rams produced higher weekly lambing percentages.

#### MATING SEASON

The basic consideration is the selection of a mating practice most suitable to a specific set of circumstances.

In sheep, autumn and spring mating will result in spring and autumn lambing respectively, consequently the seasonal aspect of mating and of lambing must be compared and contrasted.

#### 1. Autumn versus Spring Mating

Mating during autumn, when the maximum number of ewes in the flock exhibit sexual activity, is to be recommended if maximal conception is to be obtained. Conception was proved to be higher during the period of maximal increase of sexual activity immediately prior to the peak of the breeding season, than at any other time <sup>20</sup>.

Over a period of four years Hugo 6 observed only 40 per cent of ewes coming into heat in the spring mating period of four months, beginning on 15 October, as against 80 per cent for a similar autumn period beginning 15 April. Comparable figures for a period of six years were quoted from Bosman 88, namely 87,4 to 88,4 per cent for autumn mating as against 46,8 per cent, which in consecutive years increased to 84,6 per cent for spring mating, in the stud at the Grootfontein Agricultural College. Comparing the lambing percentages of flock sheep on the same farm over a number of years. Marincowitz 109 found that 83,9 per cent of ewes mated from March to May every year lambed with a lambing percentage of

90,2, while 67,0 per cent of ewes mated from September to November lambed, with a lambing percentage of 70,3 per cent.

Too long a mating and hence lambing period is not desirable from a management and economical point of view. In practice, a period of 6 to 8 weeks is usually preferred. By extending the mating period from 8 to 10 weeks during autumn no corresponding increase in conception was encountered. On the other hand, a gradual increase from 34,2 to 40 per cent was noted when the spring mating period was extended from 8 to 16 weeks <sup>6</sup>. Limiting spring mating to a practical period of 8 weeks thus leads to very poor conception rates.

From Hugo's 6 observations it can be calculated that, over a period of four years, maximal conception in the autumn-mated ewes occurred within a reasonably constant average of eight days after introduction of the rams, whereas in spring mated ewes this period decreased from 100 days during the first year to 30 days during the fourth year. These figures indicate an advancement of the mean mating date from 24 February for the first year to 15 December for the fourth year. Furthermore, they also clearly indicate that spring mating, especially in young sheep, does not occur. The average mating dates rather indicate a summer mating period advancing from late to early summer with increasing age.

The age effect is also clearly demonstrated by the fact that in the spring mated ewes the average conception rates for 42 and 56 days mating periods varied from 0 to 33,3 per cent during the first year, and from 0 to 55,6 per cent during the fourth year 6. The over-all poor results obtained by spring mating are depressed even further by the presence of young ewes. A tendency to better performance wih increasing age is mainly due to an increased duration and advanced onset of the breeding season. From a practical point of view, the percentage of young ewes in any particular flock will play an important rôle in the breeding performance of such a flock during spring mating but not during autumn mating. Furthermore, a large percentage of young ewes at the age of approximately 18 to 19 months will be missed in a limited spring mating period, and they will only be mated again a year later at the age of 2,5 years.

Towards the peak of the breeding season, the oestrus period tends to become longer and the incidence of twin or multiple ovulation tends to increase <sup>20</sup>, <sup>35</sup>, <sup>36</sup>, consequently the chances of ewes being served and conceiving will be greatly increased during autumn mating as against spring mating. Twinning rates of 40 per cent in autumn and six per cent in spring mated ewes have been established <sup>110</sup>.

Genetically speaking, the order of duration of the breeding season of the different breeds forms an epistatic series when crossbreeding 1, 6: the 'shorter season' dominate over the 'longer season' ones. British breeds, except Dorset Horns and their crosses, therefore, will have short seasons, while crosses with the Merino will have intermediate seasons. As the shorter seasons become more concentrated around the peak in April-May, it will be evident that spring and even summer mating in the shortseasoned breeds will be more unsuccessful than in the Merino or other breeds having intermediate seasons. In some of these crosses and in certain areas a mid- and late summer mating system can be attempted however, particularly with older ewes.

The cooler weather conditions prevailing during autumn would be more conducive to successful mating. In rams, high ambient temperatures and overheating adversely affect the semen picture, while in ewes it interferes with fertilization and early embryonic growth.

Finally, in the main sheep areas, autumn mating usually occurs under good natural grazing conditions, with the result that flushing is obtained by natural means and need not be effected by supplementary feeding.

#### 2. Autumn versus Spring Lambing

Apart from the substantially higher lambing percentage that can be expected by mating in autumn, there are distinct advantages and disadvantages in both lambing practices.

The practical implications of Hugo's findings  $^6$  are that autumn-mated ewes lambing in spring will have a period of  $\pm 117$  days in which to recover before being mated again—allowing for a weaning age of 90 days— while ewes lambing in autumn have a mean period of only 49 days. This short recovery period coincides with the most un-

favourable grazing conditions in the main sheep areas, thus compounding the disadvantage of autumn lambing.

Since the autumn mating is carried out at the peak of the breeding season and will be concentrated in a short period, the spring lambing season will also be of short duration as opposed to the more protracted autumn lambing, even if the spring mating period is limited to the same extent as the autumn mating period. From a management as well as an economical point of view, spring lambing is therefore preferable.

On the other hand it has been proved 111 that the cool weather prevailing in autumn and early winter favours growth and development of lambs. Furthermore, parasitic infestations and lambhood diseases have a more restricted incidence during the autumn season. The peak of the rainy season in the Karoo and adjacent sheep farming areas occurs during autumn with resultant good grazing and increased milk production. Autumn thus is superior to the spring season from a lamb-rearing point of view. This favourable position is reversed at the time of weaning, as well as the two-tooth or shedding age which will then have to take place under very poor veld conditions, whereas in a spring lambing practice both these events will usually occur under the most favourable climatic conditions. These disadvantages of autumn lambing apply particularly to the Karoo, the advantages of spring lambing to the true summer rainfall areas.

Considering that true spring mating results in low levels of conception unless extended to the summer season, and that the onset of the breeding season can be advanced to a limited extent only, a practice must be adhered to which exploits the natural season, in other words an autumn mating practice based on the natural breeding season must be adopted and means should be found to eliminate its disadvantages. These centre around lambing and rearing of lambs: most of them can be eliminated by the provision of additional feeding, shade and disease control. Despite the expense involved, the increased lambing percentage (up to 30 per cent and more) would be amply rewarding. Extending the spring mating season into the summer months would increase the lambing percentage to a little over 40 per cent at best. It would present problems of its own and neither exclude the disadvantages of

spring mating, nor fully benefit from the advantages of an autumn mating season.

In areas where autumn mating practice cannot be employed, an early and mid-summer or late autumn mating practice should be considered.

A combination of the two practices can also be carried out to a reasonable extent. This is either done by having specific groups mated in autumn and spring respectively or by mating the dry ewes from the autumn season during the spring season. Despite problems involved with management, it has the advantage of reducing the average cost of rams per lamb, as either less rams have to be kept or the same rams are used in succession.

#### MATING SYSTEMS

Four natural mating methods are used, viz. flock (mass or group mating), individual mating, hand mating and continuous mating. In flock mating a number of rams are placed in a camp with a number of ewes for a specific period, usually eight weeks, whereas in continuous mating the rams are kept with the ewes throughout the year, or are removed for a short period only. Flock mating is also known as group mating, which term is often used to denote individual mating. Since ewes are usually mated in groups, it is felt that the terminology should be based on the number of rams associated with a group of ewes.

In individual mating each ram is allowed to run separately with a specific number of ewes for a certain period, while in hand mating ewes on heat are placed individually with any particular ram and allowed to be served once or twice. These ewes are identified beforehand by the use of teaser rams.

#### 1. FLOCK MATING

#### (a) Mating Season

It has already been recommended that the natural mating season should be employed wherever possible. As the peak of the breeding season is reached during May and it has also been proved that better conception figures are obtained by mating during the period of increasing sexual activity, it would appear as if a mating season centred around the middle of April would be most beneficial. Because a longer mating season is not desirable and offers no real advantage, a maximum period of 8 weeks and not less than 6 weeks is indiciated.

#### (b) Management of Ewes

Management of the ewe flock in connection with mating actually starts a few weeks before mating, when the necessary feeding, dipping, shearing and vaccinations are carried out.

Adequate nutrition of the ewe must already start at birth of the young ewe lamb. The effects of underfeeding and starvation during lambhood and before puberty cannot be corrected by flush feeding a few weeks before mating. Breeding ewes should be protected from severe nutritional stress at any time during their lifetime. Although it has been reported that overfatness causes sterility, especially in two-tooth ewes, the natural conditions in this country would probably not allow such a problem to occur.

Flush feeding during the period of three weeks before mating should be introduced and maintained during the breeding season. The type of flush feeding is apparently not important and flushing can be effected by putting ewes on green pastures, or in rested camps, or by offering suitable supplementation.

The effect of wool production on reproduction is a matter of selection and breeding, and can only be compensated for by increasing the nutritional level. Mating of long-woolled ewes should not be encouraged, as shearing prior to mating has a definite beneficial influence on lambing percentages, especially of young ewes.

#### (c) Management of use of Rams

Rams should be examined for genital soundness as close to the mating season as possible and those with doubtful fertility, libido and mating dexterity replaced by rams possessing the desired qualities. All inoculations should be done at least six weeks prior to the rams being put with the ewes, or otherwise, postponed until they are taken out at the end of the mating season.

To avoid the stress and insulation of a long fleece and the effects of overheating on semen production, rams should be shorn shortly before the mating season, leaving only sufficient time for shearing wounds to heal. Special attention must be given to the scrotal cover, which must be clipped short. Shearing becomes more important in developed rams, as the cervical skin-folds carrying long wool will interfere with the mating act.

As the ram will have to search for and serve a large number of ewes repeatedly, they require optimal stores of semen and energy. Feeding time will be restricted; hence it is deemed absolutely necessary that supplementary concentrates or a complete ration should be given for at least three, but preferably for 6—8 weeks, before mating commences.

A practical and rewarding procedure that can be strongly recommended, is to divide rams into two groups and to put alternative groups with the ewes for a period of 14 days, except for the last fortnight, when all the rams are put to the ewes. During the period when they are not with the ewes the rams should be fed well. Another method used with excellent results is to put the rams with the ewes at night while they are rested and fed during the day. This entails more work, however.

Although the average ram does not become sexually exhausted very easily, it is my personal opinion that under natural veld conidtions in the Karoo this happens more often than is believed. It should also be remembered that when one ram is affected, his companions are additionally burdened The number of rams commonly used varies from two to three per hundred ewes. Apart from the work reported by Edgar 101, no scientific evidence for any particular figure could be obtained. In experiments carried out by him on easy country, an average of 160 ewes per ram were settled in 18 days. Only one ram served less than 100 ewes, while the figures for the other rams ranged from 100 to over 250 ewes. No signifactnt differences could be demonstrated in the docking percentages between ewes whether one, two or three rams per 100 ewes were used.

Whether these results are applicable in this country, with its topography, climatic conditions and veld cover, is questionable. Since the mating of one ram per 100 ewes becomes a risky undertaking, it is felt that the use of two to three rams per 100 ewes should be continued until more information is obtained. A decrease in the number of rams to 1 per 100 ewes would naturally reduce the cost of rams per lamb drastically. It would be a strong economical consideration, on condition that the reproduction rate remains unaffected.

There is considerable variation in opinion as regards the value of introducing teaser rams for 14 days prior to the mating season. Everybody seems in agreement that ewes are stimulated only when in the transitional period from the non-breeding to the breeding season and that vasectomized rams are less effective than normal rams, or even completely ineffective. The practice of introducing two teasers per 100 ewes will only be of value in spring mating and when young ewes are involved. Until this aspect is fully investigated in this country, it is suggested that this practice should be continued since it involves very little additional cost and effort.

#### (d) Flock Composition

The available evidence indicates that it is sound practice to separate maiden ewes entirely, especially at their first mating. The fact remains that their proportional presence in an existing flock will have a definite influence on the reproductive performance of that flock. Although this is particularly true for a spring mating practice owing to the restricted breeding season of young ewes, it is also true in autumn mating. In this system, inexperienced youngsters with short heat periods are forced to compete with older ewes, which are more inclined to seek the ram, follow him around and stay on heat much longer. Evidently young ewes on heat will easily go undetected.

When maiden ewes are kept as a separate flock, more attention can be given to them. It is also recommended that they should be mated to mature and experienced rams, while the young replacement rams are put to the older ewes.

#### (e) Camps and Camp Sizes

Rested camps should be chosen for mating because of their flushing value. It should always be endeavoured to select the more open type of camps with a more even surface and favourable topography.

Sheep are often inclined to form groups, grazing and drinking separately from other groups. More often than not the distribution of rams is uneven. Since multiplicity of watering points and large-sized camps encourage group formation, smaller camps with single watering points should be utilized for mating.

A more practical solution yielding excellent results is mustering of the sheep once or twice a week for a few hours or even overnight. This is especially rewarding in the case of maiden ewes.

#### (f) Flock Size

No scientifically founded information can be given on this aspect. Generally, smaller flocks of 200—300 ewes are preferred in practice. Smaller flocks are easier to handle and less inclined to form groups. The presence of fewer rams could possibly lead to less fighting and competition. In large flocks, group formation with uneven distribution of rams will be encouraged. Some ewes, especially the younger ones, will be missed, while the potential of some rams will be wasted.

In large flocks, where more ewes will come on heat simultaneously, mustering is essential in order to increase the opportunity for individual ewes to make contact with the ram.

#### 2. INDIVIDUAL MATING

This system is mainly used by stud farmers for progeny testing of their rams. Rams to be tested are individually assigned to a specific number of ewes in order to identify the progeny.

The number of ewes in a mating group will also be determined by factors like size of camps, topography, and the age, libido and mating dexterity of a ram. Normally, a group of 60 ewes is allowed per ram but in smaller camps or when rams are only put to the ewes at night-time, up to a 100 ewes can be served by one ram. When very young rams are used, the number of ewes should be restricted to 25 to 40 ewes.

On large stud farms insufficiency of number of camps might prove a stumbling block. Small paddocks and putting the rams to the ewes overnight could be used to overcome this problem but it would entail a great deal of work, sorting the animals and driving them to the paddocks from their camps and vice versa twice a day. With this system more ewes can be allowed per ram, thus reducing the cost of rams per lamb. Proper records of individual rams can be kept, while their breeding potential and mating ability can also be assessed.

#### 3. HAND MATING

This is the most effective natural mating system: accelerated breeding progress can be achieved, with a minimal cost of rams per lamb. It is used almost exclusively on stud farms, where specific rams are mated to specific ewes and where young rams are progeny-tested. As each mating is directly supervised, the ewes are retested after 15 to 17 days and genitally sound rams are usually used, high lambing percentages are more or less guaranteed. Any possible factor detracting from high lambing percentages can be identified within the course of one oestrous cycle and can be corrected timeously.

#### 4. CONTINUOUS MATING

This method is still practised to a certain extent today, especially in the Karakul industry. Rams are usually kept with the ewes throughout the year or are removed for a short period only. This system offers no means of checking on the lambing percentage and factors influencing it cannot be identified. It has no application to profitable farming.

#### MATING MANAGEMENT

As it concerns the consideration of all factors opposed to, and the introduction and application of all possible measures in favour of maximal reproduction, mating management constitutes the most important single aspect determining the success of reproduction.

It therefore includes all those factors previously discussed, as well as their practical application in a predetermined and organized reproductive pattern.

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#### IMMUNITY TO CORYNEBACTERIUM PSEUDOTUBERCULOSIS\*

C. M. Cameron\*\*

#### SUMMARY

Experimental results pertaining various aspects of the immunogenicity of C. pseudotuberculosis are reviewed.

It is concluded that immunity to C. pseudotuberculosis is primarily antibacterial and humoral in nature, because whole bacteria will induce immunity and mice can be protected easily with hyperimmune antiserum.

The protection-inducing antigen is an integral part of the cell wall and cannot be extracted without destroying its antigenicity. The protoplasm also contains protective antigen in small quantities as well as a toxin which is similar to the exotoxin.

Sheep which were immunized with a formalinized vaccine containing aluminium phosphate adjuvant showed an increased resistance to infection but they were not solidly immune.

#### INTRODUCTION

Infection by Corynebacterium pseudotuberculosis, also known as C. ovis or the organism of Nocard and Preisz and causing classical caseous lymphadenitis in sheep, is becoming increasingly prevalent and assuming economic importance. This communication summarizes the author's efforts at producing a vaccine of practical value.

#### IMMUNOGENICITY AND TOXICITY

Initial experiments in guinea-pigs showed that only bacteria grown under favourable conditions were able to produce any measure of immunity. A medium was ultimately formulated which supported the spontaneous development of a dense pellicle and by growing the bacteria in large, flat flasks a very high yield could be obtained 1, 2.

The protoplasm of the bacteria was toxic, and, even when killed with formalin, the bacterial cells could still produce sterile abscesses<sup>3</sup>. No relationship could be established between toxicity and immunogenicity 4.

#### MICE AS EXPERIMENTAL ANIMALS

As the use of large numbers of sheep was uneconomical for preliminary experiments and because it was difficult to measure the extent of subcutaneous abscesses in guinea-pigs, the method used by Jolly 5 in mice was adapted with very good results. The challenge dose, however, was extremely critical: doses of between 4×104 and 2×105 usually gave rise to subacute or chronic infections in controls 6.

#### IMMUNITY IN MICE

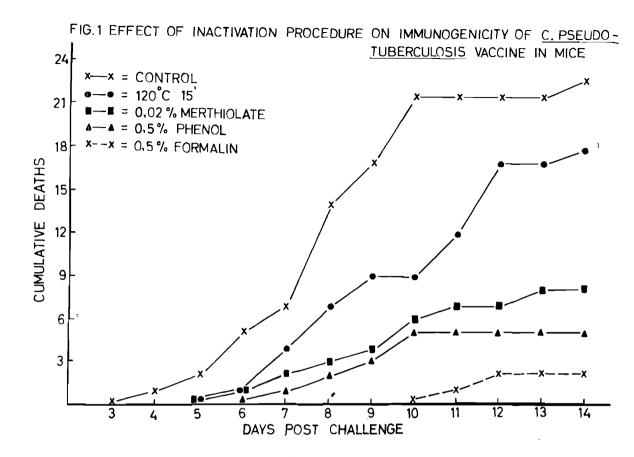
The degree of immunity, assessed by counting the total number of live bacteria in the spleens of immunized and control mice one week after challenge, gave somewhat erratic results, hence the cumulative death rate was adopted as standard measure.

A solid immunity could be obtained in mice by using washed whole bacteria as vaccine: it would thus appear that antibodies to exotoxin are of little consequence in protection 6.

Formalinized bacteria were considerably more immunogenic than those killed by other means (Fig. 1).

Hyperimmune rabbit antiserum C. pseudotuberculosis was shown to protect mice passively (Fig. 3). This antiserum was totally devoid of anti-exotoxic activity, thus again showing that antitoxin is not primarily involved in protection. Furthermore, the pro-

<sup>\*</sup>Based on a paper presented at the Biennial Scientific Congress of the South African Veterinary Association. East London, September 1971. The table and figures as well as part of the text have been reproduced from the Onderstepoort J. vet. Res. with permission of the editor.
\*\*Section of Bacteriology, Vete\*inary Institute, P.O. Onderstepoort, Rep. of South Africa.



tective effect of the serum could be markedly reduced by absorption with whole bacteria, thus proving that the immunity is primarily antibacterial in nature and mediated by serum antibodies 7.

Immunity could not be established in recipient mice by the infusion of peritoneal macrophages from immune donors. This finding would argue against the participation of a primary cellular immune mechanism being of any consequence in protecting mice against *C. pseudotuberculosis* infection.

By comparing the bacterial population changes in the peritoneal cavity of passively protected and control mice, certain interesting features were observed. It was shown that there was a similar and parallel decrease in the number of bacteria in both groups during the first phase of infection. Only after two hours was any difference detectable between the two groups, when the number of bacteria in the control group rapidly increased until death of the recipients

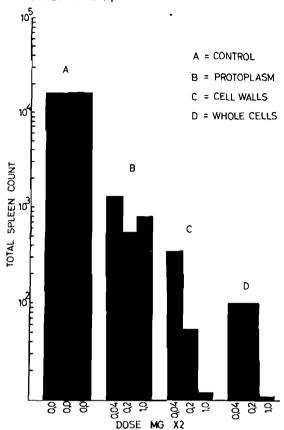
resulted. The same pattern was observed when comparing actively immunized mice to controls 7.

These findings, as well as other experiments with opsonized bacteria, indicate that the protective effect of the serum is not due to an initial opsonization resulting in enhanced phagocytosis and killing, but that the antibodies somehow effectively restrict the secondary multiplication of the bacteria.

#### NATURE OF ANTIGEN

To locate and characterize the bacterial antigen or component which was directly responsible for stimulating a protective immune response, bacteria were mechanically disrupted and the cell walls and protoplasm separated. A comparison of the immunizing properties of these components is shown in figure 2 and it is clear that, although the protoplasm has some activity, the cell wall is considerably more active, but neither is as efficient as whole bacteria <sup>6, 8</sup>.

FIG.2 COMPARISON OF IMMUNIZING PROPERTIES OF WHOLE BACTERIA, CELL WALLS AND PROTOPLASM.



Although the amount of antigen in protoplasm was small, it had the advantage of being soluble. It could be separated into two toxic components, designated A and B, by gel chromatography. By toxin neutralization tests it was possible to show that toxin B was related to the exotoxin and was not immunogenic, whereas toxin A was associated with material that had some immunizing activity. Indirect evidence was also obtained that the protoplasm also contains antigens which reside in the cell wall and that it is indeed this material which may be responsible for the immunizing activity of the protoplasm. Unfortunately, the material containing toxin A had a marked tendency to aggregate, which precluded further fractionation and an exact elucidation of its toxic and immunizing properties in relation to cell wall components 9.

Extraction of the cell wall with ether: ethanol, trichloracetic acid and hot formamide did not yield immunogenic material, nor did these procedures reduce the immunogenicity of the residual cell walls. The immunity-inducing antigen is, therefore, firmly attached to the cell wall 6. 9.

Neither detergents nor mucolytic enzymes in conjunction with chemical procedures succeeded in lysing the cell wall. Successive treatments with acid, alkali and *Streptomyces albus* enzymes, however, resulted in almost total disintegration of the cell walls but unfortunately the soluble material was devoid of all immunogenic activity. This was shown to be due to the hydrolytic effect of the alkali. Further attempts to prepare soluble antigen by using crystalline trypsin also failed <sup>9</sup>.

Soluble material was extracted from whole bacteria with hydrochloric acid under various conditions but again no immunogenic activity could be found. The same applied to acetic acid extracts and only a minute quantity of active material could be obtained by citrate extraction.

It was concluded that the antigen which induces protection is an integral part of the cell wall matrix and that chemical or enzymatic processes which alter the chemical configuration of the cell wall simultaneously result in the destruction of the antigen or antigens which are capable of evoking a protective immunity 8.

#### ARTIFICIAL INFECTION OF SHEEP

Preliminary attempts to establish lymph node abscesses by percutaneous injection of living bacteria invariably resulted in failure. Abscesses were seldom seen in the nodes but extensive subcutaneous lesions developed. These lesions were so variable in size that they were not considered to be a satisfactory criterion for assaying the immunity.

Results of experiments with different intravenous doses of dispersed bacteria derived from shake cultures were very disappointing. Doses in the range of 10 to 10 usually resulted in acute death due to pulmonary oedema, while sheep which received doses between 2×10 and 10 seldom developed any lesions. When a dose of 5×10 was used, abscesses developed in the lungs and kidneys but some sheep still died acutely (wihin 3 days) while others recovered completely.

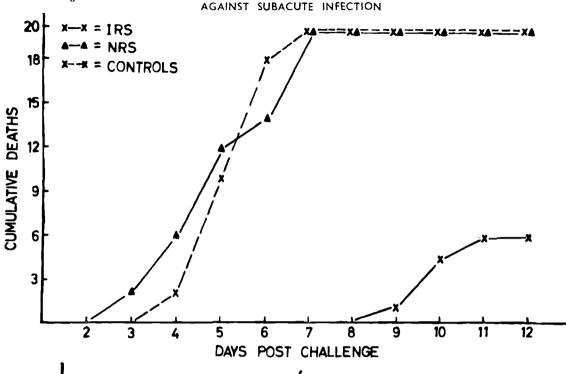


Fig. 3: PROTECTIVE EFFECT OF HIPERIMMUNE WHOLE CELL RABBIT ANTISERUM

Fig. 4: ANTIBODY RESPONSE OF SHEEP TO VACCINE GIVEN AT DIFFERENT INTERVALS

NOT THE PROPERTY OF SHEEP TO VACCINE GIVEN AT DIFFERENT INTERVALS

WEEKS AFTER LAST INJECTION

--- = PRIMARY AND SECONDARY INJECTIONS GIVEN AT 2 WEEKS INTERVAL
--- = PRIMARY AND SECONDARY INJECTIONS GIVEN AT 4 WEEKS INTERVAL
--- = PRIMARY AND SECONDARY INJECTIONS GIVEN AT 6 WEEKS INTERVAL

Far better results were obtained when fresh bacteria derived from pellicles in static cultures were injected intravenously. The small floccules or clumps of bacteria appeared to lodge in the lung and kidney capillaries and subsequently gave rise to discrete abscesses. The abscesses were usually well dispersed throughout the lung tissue but there was a tendency for more frequent occurrence in the right apical lobe. Abscess formation was considered to be a true chronic infection. In some instances sheep died 7 to 14 days after infection with diffuse purulent pneumonia or early abscess formation. Such cases were regarded as subacute infections and the superficially situated abscesses could be counted quite readily. This system proved to be a most satisfactory method for infecting sheep artificially and evaluating immunity.

Counts of the total live bacteria in weighed lung samples and density measurements for measuring and expressing the degree of infection <sup>11</sup> yielded erratic results.

#### IMMUNE RESPONSE IN SHEEP

Because the protection-inducing antigen is part of the cell wall and because serum antibodies are responsible for mediating immunity, an agglutination test should be a reliable index of the immune status of an animal which has received vaccine. Consequently, this test was adopted to follow the immune response of sheep after administration of vaccine.

C. pseudotuberculosis organisms have a marked tendency to auto-agglutinate; this presents particular problems when conducting agglutination tests. A method, however, was devised whereby these problems could largely be overcome <sup>11</sup>.

Using this method, the immune response of groups of sheep to various vaccines was compared. Whole culture vaccine containing a total of 0,25 ml packed bacteria was used in all the experiments.

Freshly prepared vaccine was shown to be highly toxic for guinea-pigs and sheep owing to incomplete toxoiding of the exotoxin. Guinea-pigs died three to four days after receiving the vaccine, while sheep developed a severe haemolytic anaemia followed by intense icterus, liver degeneration and death. If toxoiding were allowed to continue for at least a week, the vaccine was quite safe.

Increasing the dose from 0,25 ml packed cells to 1,0 ml packed cells did not improve the immune response. Similarly, the administration of three initial injections, instead of the conventional two, also had no beneficial effect.

One of the most disappointing findings during these studies was the failure to find an adjuvant which would significantly enhance the immune response. Ten different adjuvants were tested. Freund's complete and Freund's incomplete adjuvant were slightly better than other oil adjuvants, while chemical adjuvants were equally poor. Nevertheless, aluminium phosphate gel was routinely added to the vaccine because it at least served to keep the bacteria in homogenous suspension.

Another discouraging feature of the immune response was that the agglutination titres returned to pre-immunization levels within four months. Despite these rather disappointing results, attempts were made to establish the actual resistance of sheep to infection at the peak of the immune response.

The only factor which did improve the immune response to some extent was an extension of the interval between administration of the primary and secondary doses of vaccine to six weeks.

The results of an experiment in which sheep were given two injections of 5,0 ml vaccine (0,5 per cent packed cells) with an interval of six weeks, are shown on table 1. The outstanding feature of this experiment was that 9 out of 14 of the control sheep died before termination of the experiment, while only one of the immunized sheep died. This was probably due to a somewhat high challenge dose but proved that immunized sheep are clearly immune to subacute infection. When the surviving control sheep were slaughtered one month after infection, they were found to have just as many pulmonary abscesses as the immunized sheep.

In a second experiment two groups of sheep were immunized; one received two doses of 2,5 ml aluminium phosphate adjuvate vaccine (1,2 per cent packed cells) and the other group vaccine containing Wellcome oil adjuvant. The average agglutination titres at challenge were 1:35 and 1:59 respectively for the two groups. The results of this experiment are presented graphically in figure 5. As in the previous experiment, three of the six control sheep died before

Table 1: ABSCESS DEVELOPMENT IN THE LUNGS OF SHEEP FOUR WEEKS AFTER CHALLENGE WITH 10° LIVE BACTERIA INTRAVENOUSLY

	Agglutinaiton titre at time of challenge	Interval until death	Abscesses in lungs four weeks after challenge
Immunized* sheep	1:320 1:40 1:40 1:40 1:40 1:40 1:40 1:80 1:20 1:80 1:80 1:80 1:160	7 days	215 106 273 476 185 56 123 56 193 249 40 144
Mean	1 : 90		172
Unimmuniz- ed sheep	1 : 20 1 : 20 1 : 10 1 : 10 1 : 20 1 : 20 1 : 40 1 : 80 1 : 20 1 : 20 1 : 20 1 : 40	11 days 4 days 7 days 7 days 6 days 4 days 4 days 4 days 4 days 4 days	173 108 198 111 ——————————————————————————————
Mean	1 : 28		168

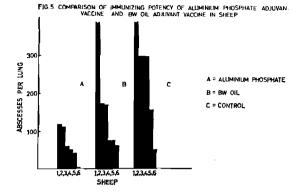
<sup>\*</sup>Immunized sheep received two 5 ml injections of vaccine containing 0,5 per cent packed cells.

termination of the experiment; 300 abscesses were arbitrarily allocated to each of these three animals.

It is clear that the sheep which received aluminium phosphate adjuvant vaccine had appreciably fewer abscesses than the control group, while the group which received the BW oil adjuvant vaccine exhibited no detectable degree of resistance.

A curious feature was that sheep which had received aluminium phosphate vaccine developed a mean agglutination titre of only 1:43 four weeks after challenge, while the group which had received BW adjuvant vaccine had a mean value of 1:183.

A certain degree of immunity to a chronic infection was thus established but, with the exception of a single sheep, it was



not absolute. Although immunized sheep developed fewer abscesses than control sheep, they were not solidly protected.

The immunized sheep probably had destroyed the majority of bacteria, or had effectively restricted their multiplication for a brief period, but a sufficient number of organisms had survived and eventually they had given rise to abscesses.

There are a number of possible explanations for the above observation.

- (a) Despite the fact that hyperimmune rabbit serum protected mice passively, exclusive humoral immunity may nevertheless not suffice in sheep and the use of live vaccines which could also induce a cellular immunity might prove to be of value, although generally subcutaneous administration of a vaccine prepared from attenuated live bacteria does not produce a better immune response than that prepared from virulent killed bacteria 10.
- (b) Conventional methods of immunization may be inadequate to afford a solid immunity. Manipulation of the antigen in order to render it more immunogenic, or the application of alternative methods of immunization may overcome the problem.
- (c) Hitherto only Merino sheep have been used and this breed may be exceptionally unresponsive to *C. pseudotuberculosis* antigen. Furthermore, sheep in general may be inherently less responsive to this antigen than other animals. In view of the immune response in different species and breeds, this aspect would not appear to play a significant rôle.

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- (d) Immunized sheep may be quite capable of destroying single bacteria but may not be able to cope with clusters of organisms which might be more difficult to phagocytose and destroy.
- (e) The challenge doses used in this investigation were probably vastly in excess of any infection which could conceivably be encountered under natural circumstances.

#### CONCLUSION

The vaccine may indeed prove to be effective when used under field conditions. This can only be proved by very extensive trials. In the light of this and the briefness of the

antibody response, it is suggested that vaccine be administered shortly before expected exposure to infection, e.g. shearing, so that the animals are maximally protected at this particular time.

Studies are being undertaken to see if chemical alteration of killed bacteria or stimulation of the animal's immunological system will enhance the immune response and improve the level and duration of immunity <sup>10</sup>.

#### ACKNOWLEDGEMENT

I thank Mrs. L. v. Reenen and the section of photography for preparing and reproducing the figures.

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DRUGS MIDDELS

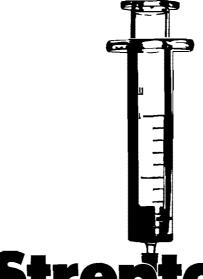
#### NARCAN

The availability of NARCAN (Naloxone Hydrochloride), a new narcotic antagonist without narcotic like properties of its own, is announced.

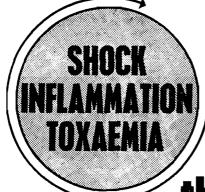
The following summary is extracted from an article which appeared in the Journal of the American Veterinary Medical Association with regard to NARCAN.

"The synthetic drug, Naloxone Hydrochloride, was found to be a safe and effective narcotic antagonist. It is more potent than nalorphine and levallorphan and does not depress respiration as commonly occurs with other narcotic antagonists. One part Naloxone Hydrochloride prevents the respiratory depression caused by 15 to 20 parts oxymorphone. Reversal of all effects of oxymorphone, including analgesic, was evident when a ratio of 0,4 mg antagonist to 1,5 mg narcotic was given. Morphine and meperidine were counteracted by Naloxone Hydrochloride to a lesser degree than was oxymorphone."

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#### THE MORPHOLOGY OF THE BOVINE TEAT CANAL: . A PRELIMINARY REPORT

W. H. Giesecke\*, W. H. Gerneke\*\* and I. B. J. van Rensburg\*\*\*

#### SUMMARY

Preliminary investigation on the morphology of the bovine teat canal indicates that the folds in the teat canal are arranged not longitudinally, but in spiral fashion. This arrangement suggests that a spiralling recoil of elastic fibres virtually wrings out and dries the canal after extraction of milk, at the same time closing it effectively.

The fine structure and function of the teat canal has been investigated repeatedly 1-15. The teat canal is described as that part which connects the external teat orifice with the teat cistern. The length of the teat canal varies from 8 to 14 mm and averages 10 mm. Usually the teat canal follows a straight and vertical course, and has a distal, central, and proximal width of about 0.4 mm, 0.5 mm and 0.8 mm respectively. The teat canal tends to lengthen and dilate considerably as the lactational age increases 16-19.

The stratified squamous epithelium of the skin of the teat is inflected into the teat canal, maintaining its stratified nature but differing from the outside epithelium in having a better developed stratum granulosum and therefore giving rise to very soft keratin. It is thrown up into a series of longitudinal dermal and epidermal folds, collectively termed 'junctional folds', and gives way to the double-layered cuboidal epithelium of the teat cistern at Fürstenberg's rosette.

The current concept of the course of the junctional folds implies that these folds proceed in a straight line from Fürstenberg's rosette towards the external orifice.

Observations on the direction of inclination of the dermal papillae as seen in cross and longitudinal sections of the teat canal have suggested that the junctional folds

are arranged in a spiral fashion. From examination of histological preparations of material obtained from two lactating cows suckled by their calves, and from one dry. but sexually mature, heifer it appears that steep junctional spirals the distal in portion of teat cistern (Fig. 1) and proceed through the teat canal (Fig. 2). These spirals make an almost horizontal, full turn at, or just distal to, Fürstenberg's rosette. The horizontal portion of each epithelial spiral is followed by a more vertically inclined part which makes a complete turn around the lumen in the middle portion of the teat canal. Distally from this second turn the junctional spirals change again gradually from the more vertically inclined course into a more horizontally arranged tight coil, terminating at the external orifice of the teat canal in horizontal folds derived from the papillary body of the skin of the teat (Figs. 3, 4). The junctional spiral of a heifer appears to be shorter than that of a lactating cow. The arrangement is shown schematically in figure 5.

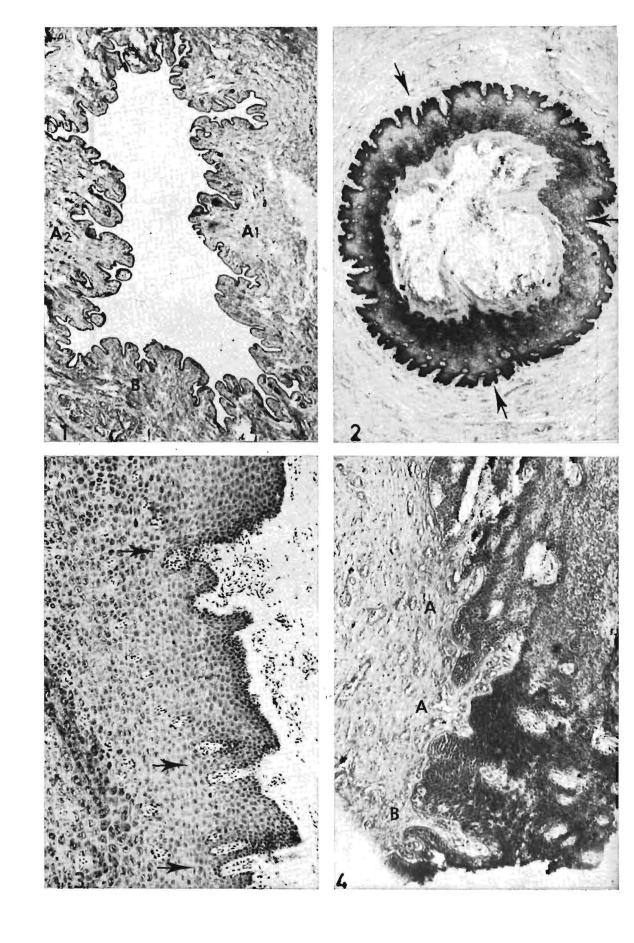
The exact arrangement of elastic fibres and of smooth musculature around the teat canal remains to be determined. In view of the direction of the spiral folds, it is possible that the teat canal is opened by contraction of smooth muscle and closed by a twisting motion produced by recoil of the elastic fibres. Therefore it is presumed that, after removal of milk, the normal teat canal is virtually wrung out and dried by a spiralling contraction proceeding from Fürstenberg's rosette towards the external orifice. At the same time the canal would be closed off effectively.

This preliminary report is to be followed by more explicit data when further investigations into the exact nature of these junctional folds are completed.

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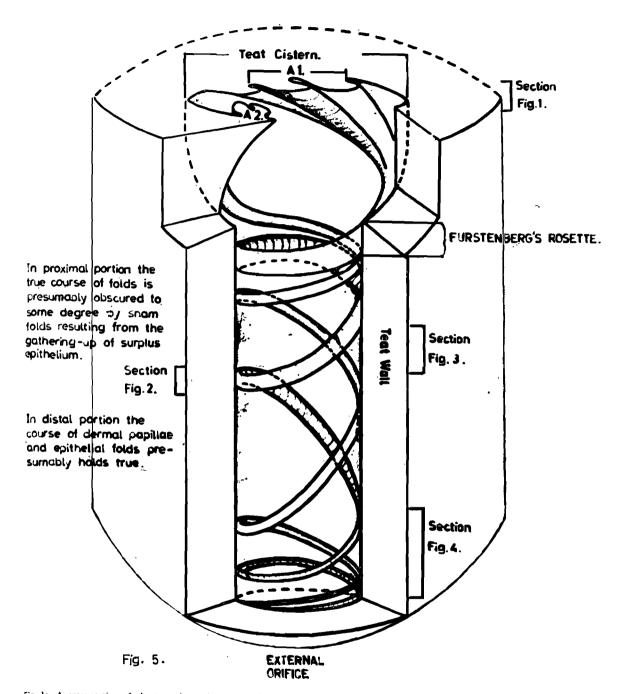


Fig. 1: A cross-section of the teat cistern just proximal to Fürstenberg's rosette. The direction of the epithelial folds at A suggests spiralling in at least one direction, whereas the folds at B possibly suggest couter-spiralling. HE, 30 ×.

Fig. 2: A cross-section of the proximal portion of a dilated teat canal. The direction of epithelial folds and dermal papillae (arrows) suggests spiralling in at least one direction; counter-spiralling is possible. HE, 30 ×.

Fig. 3: A longitudinal section of the proximal portion of a dilated teat canal. The direction of dermal papillae (arrows) — including the partially cut ones — suggests spiralling of the teat canal epithelium. HE, 70 ×.

Fig. 4: A longitudinal section of the external surface of a dilated teat canal. The direction of proximal dermal papillae at A suggests a suggests a horizontal course. Gomori's silver technique. 70 ×.

Fig. 5: A proposed scheme of epithelial spirals in a teat canal reconstructed from a series of micrographs. Only two epithelial spirals in a teat canal reconstructed from a series of micrographs. Only two epithelial spirals in a teat canal reconstructed from a series of micrographs.

Fig. 5: A proposed scheme of epithelial spirals in a teat canal reconstructed from a series of micrographs. Only two epithelial spirals have been shown.

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#### INFORMATION

#### EGG SHELL QUALITY

INLIGTING

The question is always asked about increasing the level of Vitamin D in order to increase calcium absorption. Research at Washington State on levels of Vitamin D ranging from 600-39 000 units per kilogramme of feed showed no improvements above normally prescribed levels. It seems that Vitamin D activity reaches a plateau. The only times in recent years when extra Vitamin D seemed to help were in exceptional cases where the initial Vitamin D of a ration was of poor quality due either to manufacturing or marketing problems.

Dr. Milton Scott of Cornell found that the substitution of large oyster shell flakes for some of the ground limestone in the ration increased blood levels of calcium and also resulted in thicker shells. But Dr. E. P. Singsen of the University of Connecticut encountered difficulties in reproducing these results. Dr. Singsen recently conducted a series of experiments where he varied the source and quantity of calcium and levels of sodium bicarbonate in the feed but decreased the common salt level. The results of these trials directed his thinking to high temperature effects during the summer.

Birds pant to control temperature. As they do so, they lose an increased amount of carbon dioxide from the lungs. This condition results in an increased acidity of the

blood and decreased level of bicarbonate. The lack of the bicarbonate ion in the bird's blood system then affects shell thickness. If salt is decreased, there are less chloride ions to be involved in the acidity. Adding the sodium bicarbonate provides both those elements at levels the birds can use. Dr. Singsen feels this may be another technique of value during the hot summer months.

Dr. Singsen concluded that egg production is not affected adversely except where salt is removed and the sodium not replaced. In his trials egg weight and shell thickness were most favourably affected by the final treatment which was full level of ground limestone, limestone chips free choice, lowered salt and increased sodium bicarbonate. His suggestions for feeding hens during hot weather would be to use a ration with all the normally recommended ground limestone, salt level of four pounds per ton, five pounds of sodium bicarbonate added per ton and offer oyster shell or limestone chips on free choice.

Combings, New Hampshire Cooperative Extension Service (June 1972); University of New Hampshire, Durham, N.H. 03824.

(Taken from: AGRICULTURAL REPORT of the Agricultural Counsellor, Embassy of South Africa Washington, D.C. as published by the Department of Agricultural Technical Services, Pretoria, 1972).

## NUTRITION AND OVARIAN ACTIVITY OF MARES EARLY IN THE BREEDING SEASON

C. H. van Niekerk\* and J. S. van Heerden\*\*

#### SUMMARY

The length of the oestrous cycles, the size and activity of the ovaries, and the time and incidence of ovulation early in the breeding season of mares kept on veld grazing (Group A) were compared with those of mares which received a supplementary diet of concentrates and lucerne hay (Group B).

The average mass of Group A remained more or less constant against a marked increase in body mass in Group B.

All the mares in Group B came into oestrus and ovulated within a period of 43 days. In Group A only 25% of the mares ovulated, while 25% remained in anoestrum. The remaining 50 per cent came on heat but did not ovulate during the 53-day experimental period.

A marked difference in ovarian activity between the two treatment groups was noticeable.

#### INTRODUCTION

Foals are born in every month of the year; this implies that the mare is polyoestrous and capable of reproducing all year round. The difficulty in getting all mares to settle during winter and early spring?, however, supports the classic statement of Heape 3 that the mare is a polyoestrous animal with a tendency to the monooestrous state.

The mechanisms which control the reproductive function of the mare have long been the object of speculation. Livestock breeders have credited the appearance of green grass, sunshine and warm weather, whereas research workers have suggested daylight length as initiators of sexual activity 1, 4, 5.

Kupfer<sup>6</sup>, who worked mainly on donkeys and a few horse mares kept under ordinary

veld conditions, concluded that the appearance of oestrum in the South African donkey and horse is seasonal. Quinlan, van Rensburg & Steyn<sup>5</sup>, on the other hand, observed that the oestrous cycles of the majority of mares, stabled at night and fed on concentrates and hay, continued throughout the year. They also speculated on the possibility of nutritional factors probably being responsible both for the irregularities noted in the oestrous cycle and partly for the low fertility.

Much research has been done on flushing of ewes, which are also seasonal breeders, before mating 7. On the mare, however, except for the work of Bengtsson & Knudsen 8 on trotting mares in training, no references could be found on the effect of flushing on the ovarian activity early in the breeding season.

As irregularity of the oestrous periods, such as anoestrum and long oestrous periods with active, non-cycling ovaries early in the breeding season, is one of the main problems in horse breeding <sup>2</sup>. <sup>4</sup>. <sup>9</sup>. <sup>10</sup>, this experiment was designed to study the effect of supplementary feeding on the ovarian activity of mares early in the breeding season.

#### MATERIALS AND METHODS

The observations which form the basis of this study were made at Onderstepoort on 15 maiden mares about 4 years old and of the light farm type.

These mares were kept as a group on veld grazing for at least one year prior to this experiment. On the 28th July, 1964, (Day 0) the 15 mares, all in good veld condition, were divided according to mass, incidence of oestrus, and ovarian activity, into two groups, A and B.

All the mares were kept on veld grazing, but Group B received a supplementary diet of 11,4 kg (25 lbs) lucerne hay and 2,7 kg (6 lbs) of concentrate daily for a period

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of 53 dayst. All the mares were teased daily by an active stallion. Ovarian changes were assessed by daily rectal palpation of the ovaries. The estimated size of the ovaries and follicles, as well as the day of ovulation, was recorded. Mares near ovulation were served by fertile stallions and a rectal diagnosis of pregnancy 11 was made ±20 days after ovulation. All the mares, diagnosed as pregnant, either foaled, or were slaughtered

this experiment began on the 28th July (Day 0). The average mass of the mares in Group A on veld grazing remained more or less constant throughout the experiment. In Group B, however, there was a marked increase in average body mass from 370 kg to 423 kg over the 53 days.

Mares 37 and 9, in Group A, lost 32 and 20 kg respectively; they were the only two mares in both groups that never came on

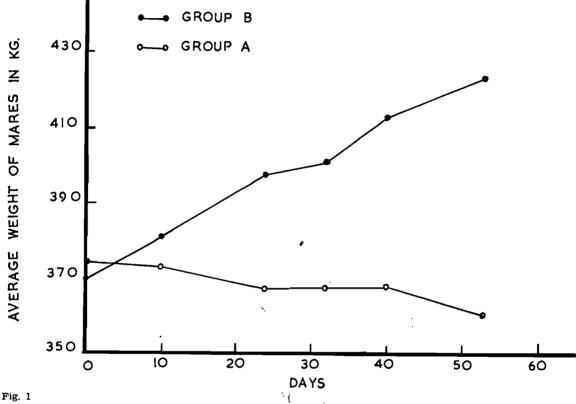


Fig. 1

before their normal foaling date, the foeti being found in the mares.

All mares in both groups were weighed at regular intervals.

#### RESULTS AND DISCUSSION

#### Mass changes

The average masses of the mares in Groups A and B during the 53-day period of treatment are presented in Fig. 1.

There was only 4 kg difference in average body mass between groups A and B when

heat. On the other hand, Mares 84 and 58 in Group A gained in mass during the first few weeks of the experiment; they were the only two mares in Group A that ovulated (Table 1).

#### Oestrus and ovulation

The number of days each mare was on heat during the 53-day period of the experiment, the length of the period between Day 0 and ovulation, and the incidence of ovulation and conception, are given in Table 1.

Veld grass: Protein 3,2 per cent Lucerne hay: Protein 15 per cent Concentrates: Protein 11,9 per cent Fibre 33,6 per cent 32 per cent Fibre Fibre 6,3 per cent

<sup>†</sup>Analysis of feedstuffs:

Table 1: THE NUMBER OF DAYS EACH MARE WAS ON HEAT, THE LENGTH OF THE PERIOD BETWEEN DAY 0 AND THE DAY OF OVULATION, AND THE INCIDENCE OF OVULATION AND CONCEPTION OF THE MARES

Group A (on veld grazing only)

Total days on Mare No. heat	Length in days from Day 0 to ovulation or Day 53	Ovula- tion	Concep- tion
84* 42 58 24 37 0 42* 51 44* 24 55 15 61* 23 9 0	49 38 53 53 53 53 53	Yes Yes No No No No No	Yes Yes No No No No No
Total: 8 179	171	2	2
Average 30 (8 mares)	44 (2 mares)	25%	25%

Group B (veld + supplementary diet)

11* 38* 39 43* 45 66 74*	10 24 28 15 36 12 33	8 24 43 17 41 16 32	Yes Yes Yes Yes Yes Yes	Yes Yes Yes Yes Yes Not served Yes
 otal: 7	158	- 171	7	6
verage:	23	24	100%	100% of 6 mares

<sup>\*</sup>Mares on heat at the beginning of the experiment.

On Day 0 five of the eight mares in Group A and four of the mares in Group B were on heat. During the following 53 days all the mares in Group B but only 75% in Group A came on heat. The mares in both groups showed the fairly long heat periods common to this time of the year 4.5.9.

The heat periods were shorter on the average in Group B, which received supplementary feeding. The big difference between the two groups lies in the fact that 100% of the oestrous periods in Group B ended in ovulation. Therefore, all the mares in this group ovulated within the 53-day period. In Group A only 33 per cent of the mares that came on heat, i.e. only 25% of all the mares in this group, ovulated. Osborne 12 found in her slaughter experiments an ovulation rate of 18,5% in August and 21,5% in September. Conclusive evidence is given that the fertility of the ovum is not affected by the treament, as the conception rate of the mares that ovulated was the same in both groups.

#### Ovarian changes

The sizes of the ovaries and follicles of mares in both groups, when the experiment began on 28 July (Day 0), are given in Table 2. The ovaries of all the mares in both groups on Day 0 were in about the same state of activity. The ovaries of all the mares were active, with a knobbly surface owing to the presence of small follicles less than 1 cm in diameter. The largest follicles present measured only 2 cm in diameter. There was little difference between the ovarian activity of mares on heat and those not showing heat. This indicates that, although the ovaries were all active, the mares were probably not cycling at this

Table 2: THE SIZE OF THE OVARIES AND FOLLICLES OF THE MARES ON THE 28th JULY (DAY 0)

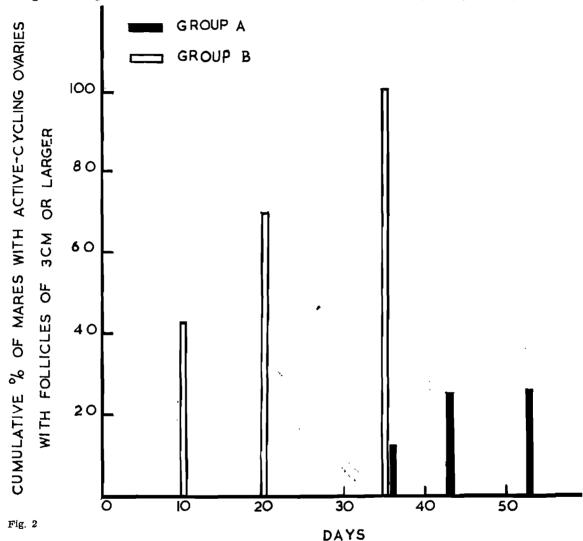
Group A				Group B	
Mare No.	Size of ovary cm	Size of follicle	Mare No.	Size of ovary cm	Size of follicle
84*	5 ×3 ×3	2	11*	5 ×3 ×2	2
58*	$3.5 \times 2.5 \times 2$	Knobbly	38*	$3.5 \times 2.5 \times 2$	Knobbly
37	$3,5 \times 2,5 \times 2$	Knobbly	39	$3 \times 2 \times 1.5$	Knobbly
42*	$4 \times 3 \times 2.5$	2 ,	43*	$3 \times 2.5 \times 2$	Knobblý
44*	4 ×3 ×2,5	2	45	$3.5 \times 2.5 \times 2$	1,5
55	3 ×2 ×2	Knobbly	66	$3 \times 2.5 \times 2$	1,5
61*	$2.5 \times 2 \times 2$	Knobbly	77*	$3.5 \times 2 \times 1.5$	Knobbly
9	$3 \times 2.5 \times 2$	Knobblý			•

<sup>\*</sup>Mares on heat

stage, a phenomenon described by van Nie-kerk<sup>1</sup>.

The changes in ovarian activity and the incidence of ovulation in Groups A and B are given in Figs. 2 and 3.

The ovaries of Group B showed active cyclic changes at a much earlier stage than Group A. Thus, within the first 10 days, 43% of the mares in Group B had follicles of 3 cm and larger. By 20 days this had



There is marked difference in ovarian activity measured by ovarian size and follicular growth between the two treatment groups. The ovaries of all the mares in Group B became active and cycling and ovulation occurred, while only 25% of the ovaries of mares in Group B, Mares 84 and 58, became active and cycling, but the rest of the ovaries of the mares in this group remained active but non-cycling, with follicles less than 2 cm in size.

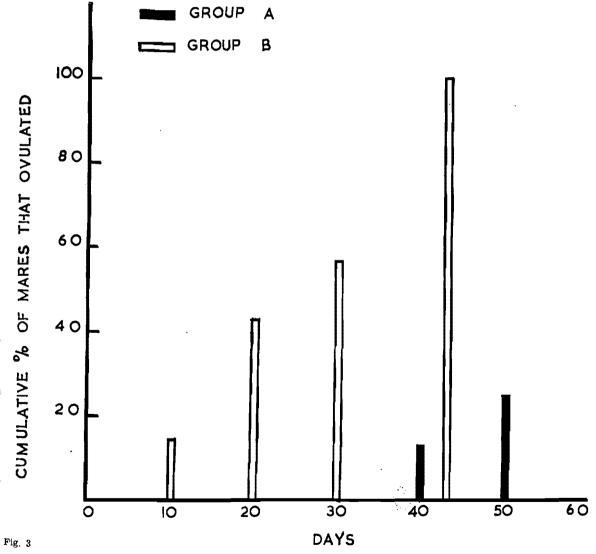
risen to 70% and by 35 days to 100%, all of which ovulated. On the other hand, no mares in Group A had follicles of 3 cm and larger by 20 days, and only one mare (12%) by 35 days, which ended in the first ovulation in this group. At the end of the experimental period of 53 days, only 25% of the ovaries of mares in this group had cyclic changes terminating in ovulation.

In Group B, the first mare ovulated eight days after receiving supplementary

feeding and by 43 days all the mares in this group had ovulated (Fig. 3). In Group A, however, only the first mare had ovulated at this stage, and the second and last one just before the end of this experiment (Fig. 3).

#### CONCLUSION

From the results obtained it is obvious that a diet high in protein and carbohydrates resulted in an increase in body mass, and this had a beneficial effect on the fertility of mares early in the breeding season. In the group on veld grazing, follicular growth and ovulation were suppressed. It seems that the release of FSH and LH from the anterior hypophysis, which is responsible for follicular growth and ovulation, is stimulated not only by an increase in daylight length, but also by the diet of mares. Supplementary feeding high in protein and carbohydrates can, therefore, be prescribed as a treatment for mares in anoestrum as well as for mares with abnormal anovulatory oestrous periods early in the breeding season.



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#### INFORMATION

#### INLIGTING

#### RABBITS

Consumer resistance to rabbit meat is gradually disappearing in the United States. Last year U.S. Department of Agriculture estimated that 18 466 000 lbs of rabbit meat were consumed in the U.S.A. With the increasing demand for variety in foods the rabbit industry can be expected to grow considerably within the next few years. At present the broiler industry enjoys the distinction of producing the cheapest meat in this country. This is due largely to research in this field which has resulted in the remarkably high feed efficiency of chickens. Even without the benefit of this well organized and concentrated research programme, however, rabbit meat has a fair chance of competing for the market. Feed efficiency is already relatively high, being in the vicinity of 1 kg of rabbit per 3,5 kg intake of feed and while its present supermarket price is more than twice that of broilers it has one major advantage over them—bone comprises no more than 8% of the total carcass weight.

These facts emerged during a recent interview with Mr. Peats, owner and manager of Hill Crest in Hill City, Kansas, which is one of only two rabbit abattoir and meat packing plants which participate in the voluntary U.S.D.A. rabbit plant inspection scheme. This scheme involves the presence of a U.S.D.A. appointed inspector at all stages, from slaughter to packing, costing the company \$1800 per month. One assumes that the advantage to the company comes by way of advertising.

The plant is at present handling between 5000 to 7000 rabbits a week. It operates on a dis-assembly line basis with 17 men and women working a 30 to 35 day week. They slaughter, skin, viscerate and cut into portions before packing for deep-freeze and of that time 1½ hours a day are spent in keeping the premises spotlessly clean. There is no fuss or distressing noise and the whole operation runs smoothly at a rate of just over four minutes per rabbit.

The financial aspects of this seemingly efficient operation are not what one might expect. Mr. Peat is confident after five months of operations that, when he has 10 000 rabbits going through the line each week, his present small monthly loss will turn to profit. The profit will grow when he adds pelts and blood serum (veterinary grade) and other byproducts like thrombo-plastin and carcass meal.

There are 40 000 farms in the U.S. where rabbits are raised and Mr. Peat's is one of these, with 500 producing does on his farm (600 are considered full time employment for one man), which can bring in a profit of \$10 per head/pa.

Neither primary producer nor processor seems to be headed for a quick profit in this venture. However, the consumer gains, as rabbit is very good to eat and very nutritious too.

#### Personal communication.

(Taken from: AGRICULTURAL REPORT of the Agricultural Counsellor, Embassy of South Africa, Washington, D.C. as published by the Department of Agricultural Technical Services, Pretoria, 1972).

#### HAEMOGLOBINS IN WILD ANIMALS

D. R. OSTERHOFF\*, I. A. PETRIE\*\* AND E. YOUNG\*\*\*

#### SUMMARY

Haemoglobins of lion, impala, black rhinoceros, zebra, buffalo, elephant and wildebeest were investigated and their migration pattern in starch gel electrophoresis established.

#### INTRODUCTION

The haemoglobins of vertebrates display a similarity of composition and structure which bears witness to their possible common evolutionary origin 1. The haemoglobin molecule is of tetrameric structure, composed of two pairs of identical polypeptide chains. The presence of more than one normal haemoglobin is common in domestic animals 2-5. In each, the variant can be traced to one or other of the polypeptide chains; in every case where the analysis has been carried out. it has been found that the variant chain differs from its normal counterpart by the substitution of only one or two amino acid residues 6. Many studies have been performed on domestic animals 7, and questions of linkage and natural selection have also been discussed in other species. A series of abnormal haemoglobins has been found in man, but in animals none of the known variants is apparently associated with a pathological condition.

#### MATERIAL AND METHOD

In a survey performed on stored blood samples of wild animals in the Kruger National Park, the following specimens were included: 19 lions, 101 impalas, one black rhinoceros, 33 zebras, 27 buffaloes, 109 elephants and 12 head of wildebeest. The samples were very often haemolysed before treatment, hence the faint bands in samples A, B, E, F and G (Fig. 1), which are actually serum proteins.

The original technique of starch gel electrophoresis but with a few modifications was applied.

#### RESULTS AND DISCUSSION

The migration rates of the haemoglobins of different species are depicted in figure 1, where sheep haemoglobin type BB (H) and cattle haemoglobin type AC (1) are included for comparison.



# RATE OF MIGRATION OF DIFFERENT HAEMOGLOBINS

A—Lion, B—Impala type AB, C—Black Rhino, D—Zebra, E—Buffalo, F—Elephant, G—Wildebeest, H—Sheep type BB and I—Cattle type AC.

The results obtained can be summarized as follows:

Lion (A): All 19 samples investigated showed only one band, and no variation was found. Impala (B): It was established that there are three haemoglobin alleles responsible for the different phenotypes found in the following frequencies: 8 AA, 49 BB, 2 CC, 29 AB, 11 BC and 2 AC, giving the following gene frequencies Hb<sup>A</sup>=0,233; Hb<sup>B</sup>= 0,683 and Hb<sup>C</sup>=0,084. In figure 1 the haemoglobin sample B represents the haemoglobin AB. The faint bands behind and in front of the haemoglobins originate from the serum proteins.

Black Rhinoceros (C): Only one sample was included in this comparison which, in figure 1, appears as one thick band (sample C). From earlier investigations it was established that the black rhinoceros possesses two types of haemoglobin, which present themselves as two bands migrating very closely together. The white rhinoceros, however, possesses only one type of haemoglobin, identified by a single band.

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Zebra (D): Two types of zebra haemoglobin were identified, one type showing a single line and the other type possessing two lines. Of the 33 samples investigated 32 belonged to the double band type, the one (D) in figure 1 being haemoglobin of the animal with the single band. The frequency of the different types in zebra haemoglobins is in agreement with an earlier investigation 4. Buffalo (E): One type of haemoglobin was found in the 27 samples; it was represented by a double band. The bands were very close together and the front band always appeared to be darker than the slower migrating one. In figure 1, sample E, the two bands can be clearly seen. Again this is in agreement with an earlier investigation on 178 samples of buffalo where, in all cases, the typical double band pattern was obtained 11.

Elephant (F): Haemoglobin typing was performed on 109 blood samples, but all samples exhibited only one migration line,

and no variation was found <sup>12</sup>. From figure 1, sample F, it is obvious that the elephant haemoglobins migrate faster than all the other haemoglobins investigated.

Wildebeest (G): Only one type was found in the 12 samples investigated, expressed by a single band in the electrophoretogram.

The chemical nature of the haemoglobins of wild animals is not known, but it is obvious from figure 1 that distinct differences in migration rates exist. The elephant haemoglobins migrated the fastest, followed by that of black rhinoceros, with lion, impala and zebra haemoglobins migration almost the same rate; buffalo and wildebeest haemoglobins exhibit the slowest migration rate.

This study has to be regarded as a preliminary one, and should be followed up with chemical studies of the nature of the haemoglobins; this could lead to interesting phylogenetic conclusions.

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# GEELDIKKOP: EXPERIMENTAL INDUCTION BY FEEDING THE PLANT TRIBULUS TERRESTRIS L. (ZYGOPHYLLACEAE)\*

E. M. van Tondert, P. A. Bassontt and I. B. J. van Rensburgtt

#### SUMMARY

For the first time since the work of Theiler (1918) and Quin (1928, 1929), geeldikkop has been successfully produced in various feeding experiments where Tribulus terrestris was used in order to assess the significance of this perennial plant as a possible aetiological agent of the disease. The results obtained afford convincing evidence that the ingestion of T. terrestris during certain seasons can cause geeldikkop. The latent period varied from 3 to 34 days with an average of 13 days. Although the precise mechanism is still unknown, it is evident that a toxic substance, be it an unstable aberrant metabolite or a mycotoxin within the plant or in adhering fungal spores on its surface, is responsible for a syndrome of photosensitization and icterus. The lesions observed correspond to those of natural cases and the most significant ones to those reported by Theiler. Of these lesions, the most characteristic one is the presence of a crystalloid cholesterol-like material (and the clefts left behind when it has been dissolved during processing) in hepatocytes, Kupffer cells and within bile ducts, frequently occluding them.

A brief history of the incidence of the disease on the farm Grootfontein in the district of Middelburg, Cape, is given and details of the outbreaks on this property during 1971 are discussed.

#### INTRODUCTION

Although geeldikkop (Tribulosis ovis) has been studied intensively since some time before the turn of the century, the primary cause has remained an enigma.

Interesting theories have been advanced, embracing a wide variety of possible aetiological factors; on subsequent investigation no conclusive evidence helpful in elucidating the true nature and underlying cause of this particular disease entity has been produced.

Contrary to the fact that in relatively recent work 1 Tribulus terrestris was relegated to a minor rôle as stress factor, by virtue of its saponin content, in the aetiology of the disease, latest evidence and observations seem to indicate that this particular plant is more directly associated with geeldikkop. The majority of small stockowners in this country, like their predecessors, has remained adamant that ingestion of the plant is associated with occurrence of the disease; it is invariably the only consistent feature in outbreaks of geeldikkop. Moreover, the only successful results in reproducing this disease under experimental conditions have thus far been obtained by feeding of Tribulus to the animals concerned 2-4. It must be admitted that these successful attempts were few and far between and are outnumbered by a mass of unsuccessful work in this respect 2-9. Nevertheless, it would appear from the relevant literature that there are certain specific conditions which must be fulfilled in order to obtain the desired results. In this respect Theiler<sup>2</sup>, notwithstanding previous negative results 10, 11, decided to repeat feeding experiments with sheep exposed to similar atmospheric conditions as those prevailing in natural outbreaks. Feeding experiments carried out at Onderstepoort, using either green or dried plants sent up from the Cape Province and Orange Free State, or green plants collected locally, consistently failed to produce the disease. On the other hand, in a series of experiments on the farm Bergrivier, near Luckoff, in the district of Fauresmith, Orange Free State, Theiler 2 was able to produce geeldikkop in 60% of sheep fed in a paddock on Tribulus plants only. Although these results could not be repeated, he was convinced that the

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<sup>\*</sup>Paper given at Biennial Scientific Congress, South African Veterinary Association, East London, September, 1971. †Regional Veterinary Investigation Centre, Middelburg, Cape.

flowering stage of this plant was the exclusive cause of geeldikkop. Quin3, in two experiments carried out approximately 10 years later, was able to produce geeldikkop in sheep grazing on wilted Tribulus, but failed to do so in a third experiment where the green, luxuriant plant was grazed. In subsequent experiments, he was able to cause geeldikkop in lambs grazing different growth stages, viz., young pre-flowering, flowering and late fruiting stages, but failed repeat experiment successfully. the Drenching experiments with the late fruiting stage of Tribulus, carried out concurrently and also subsequently 5, failed to cause the disease, but did cause deaths, presumably owing to nitrate poisoning. Finally, it would appear from the literature that nearly all subsequent work in assessing the possible rôle of Tribulus in the aetiology of geeldikkop 6-9 was carried out by drenching plant material or extracts prepared from it. All these attempts failed to produce the disease.

As geeldikkop has not been reproduced since the time of Theiler 2 and Quin 3.4, the present study was undertaken in order to re-investigate the possible rôle of *Tribulus* in the aetiology of this disease.

#### RELEVANT HISTORY

An opportunity to study geeldikkop arose when a comparatively severe outbreak of the disease occurred on the experimental farm of the Grootfontein Agricultural College, Middelburg, Cape Province. It is therefore pertinent to present a brief review of the incidence of geeldikkop on this property, which falls within the enzootic area and has an average annual rainfall of 360,5 mm.

#### Previous Outbreaks

Reliable reports reveal that only four previous outbreaks were experienced on the farm since 1938. The first outbreak occurred in 1953, followed by outbreaks in 1959, 1963 and 1967. According to the available records, it would appear as if these incidents could hardly be referred to as 'outbreaks', as that of 1959, the severest of the three, involved only nine lambs and six ewes. The total number of sheep involved in the preceding outbreaks includes one lamb in 1953, a Merino ewe in 1963 and two Merino lambs in 1967. Thus there were only 18 reported cases of geeldikkop on this farm during the last 33

years, involving a total of six different camps on the property.

#### Recent Outbreaks (Fig.)

Two outbreaks were encountered during late summer and early autumn of 1971. In the first of these, a flock of 90 mature Merino ewes was moved to a specific camp (Camp 1) during the third week of January. When these animals were collected five days later. four of them were suffering from geeldikkop. The flock was immediately transferred to a mountain camp and no further cases developed. As result of a poor grazing, this flock was again moved to a lower lying camp after approximately ten days. When a further case of geeldikkop developed after the sheep had been in this particular camp for almost 14 days, they were finally put into a different mountain camp, where they remained without further cases appearing.

The second outbreak, which was of a more severe nature, occurred almost a month later. In this case, a flock of two-tooth Merino ewes was placed in a camp (Camp 2). adjoining Camp 1 and bordering its western side, on the afternoon of 10/3/71. During the afternoon of 15/3/71 (i.e. within five days) a few sheep appeared to be ill, but geeldikkop was only reported and confirmed early the following morning. These sheep had been moved to a nearby camp the previous afternoon but were then returned to Camp 2 in order to allow certain investigations to be made. Altogether 22 acute cases of geeldikkop were removed from the flock on 16/3/71. One sheep could not be found and was assumed to have succumbed to this disease. A further two acute cases occurred on the following day and were similarly removed from the flock. As the animals in this particular camp (Camp 2) were included in Experiment 5, their fate will be discussed in the appropriate section of this paper.

# GENERAL OBSERVATIONS, EXPERIMENTAL ANIMALS AND METHODS

#### General observations

These observations were made in connection with the incidence and appearance of *Tribulus* and the grazing habits of sheep in Experiments 3, 4, 5 and 6, while observations on *Tribulus* in the different experimental camps were made daily on a comparative basis with regard to density, stage and lush-

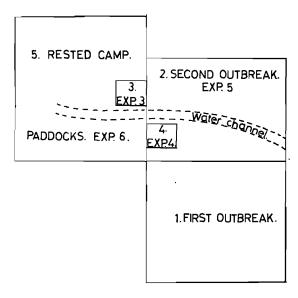
ness of growth. Individual sheep in each group were closely observed with regard to plant preference, where they concentrated in the large camp, and to what extent *Tribulus* was grazed in all the camps.

#### Rainfall

As no rain gauges were available in the experimental camps, the figures from two camps 'Boesmankop' and 'Perdekamp' situated 2—3 km away and in an easterly and a westerly direction respectively, were taken. It was assumed that the average of these figures would be more or less representative of the rainfall in the experimental camps.

#### Experimental Camps (Fig.)

In addition to Camp 2, two small temporary camps, approximately 10,9 ha in size, were fenced off; one situated in an adjoining rested camp (Camp 5) and the other in Camp 2. For easy reference, the numbers 3 and 4 respectively, which also coincide with the appropriate experiments, were allocated to them. These camps were placed intentionally so as to contain *Tribulus* almost exclusively.



A movable paddock, enclosing an area of approximately 25 square metres, was constructed from gates of different sizes in order to adapt its shape to particular patches of *Tribulus* and to exclude as many other plants as possible. All other plants, except *Tribulus*, were removed before introduction of the experimental animals.

#### Plant Survey

In order to determine the *Tribulus* population in relation to the total plant population in Camps 3 and 4, plant surveys were carried out. The Descending Point Method <sup>12</sup> was used and 500 readings per camp were made.

#### Experimental Animals

The flock of 129 two-tooth Merino ewes in Camp 2 involved in the second outbreak of geeldikkop was included in one experiment, as well as a group of six Merino ewes of varying ages, previously fed on lucerne hay exclusively for up to four years, and four four-month old Merino ram lambs.

All other animals used were bred on the farm and consisted of recently shorn mature and two-tooth Merino ewes and six to eight month-old Merino wethers. These animals were selected at random from flocks kept in camps with no previous history of geeldikkop. The remainder of these flocks either stayed in the same camp from where the experimental animals were selected or were changed to other 'non-toxic' camps, i.e. camps with no previous history of the disease.

#### Examination of Experimental Animals

All sheep introduced into the various experiments from elsewhere on the farm were examined individually and daily throughout the duration of each experiment, except for a few occasions when such work was interrupted or prevented by rain.

Animals in the original flock were gathered in an existing set of pens in the camp once or twice daily and closely observed for any signs of illness. Only suspicious cases were examined individually. These examinations consisted of a close visual inspection of the head and hoofs for characteristic signs of photosensitization and of the mucous membranes for icterus.

Autopsies were made on natural and experimental cases, many of which were sacrificed at various stages of the disease. Specimens were collected in 10% buffered formalin for histopathological studies. These specimens were processed and stained in a routine manner.

#### EXPERIMENTAL DESIGN

Experiment 1: Attempted transmission with blood

Notwithstanding negative results obtained by previous workers 2. 10. 11, this experi-

ment was carried out in order to exclude the possibility of a transmissible causative Four young Merino ram lambs. agent. approximately four months of age, were suitably marked and kept on lucerne hay in a paddock at the Veterinary Investigation Centre, Grootfontein together with a group of lambs and ewes of mixed age which acted as controls. These lambs were divided into two groups and each pair received 10 ml heparinized blood intravenously from two different donors on 16/3/71. The two donors represented acute incipient cases of geeldikkop not noticeably sick earlier that specific morning but with grossly swollen faces three hours later.

#### Experiment 2: Feeding Tribulus

Twenty young (±8 months old) Merino wethers were divided at random into two groups and placed in adjoining paddocks at the Veterinary Investigation Centre without protection against prevailing climatic conditions. They were transferred to their respective paddocks during the morning of 16/3/71, when this experiment commenced.

The experimental group was fed exclusively on *Tribulus* collected from Camp 2 as well as from the rested camp (Camp 5), while the control group was given lucerne hay only. Both groups were fed and watered from troughs.

From 16 to 18/3/71 the experimental group received Tribulus collected on the sixteenth March and from 19 to 21/3/71, Tribulus collected on the nineteenth. Between collections this material was stored at room temperature in jute bags at the Veterinary Investigation Centre. From 22/3/71 onwards fresh material was collected and fed daily. As it became extremely difficult to gather sufficient amounts of Tribulus from Camp 2, daily collections from 23/3/71 onwards were made from the rested camp with its more abundant growth. The other members of the flock from which these animals were selected, acted as controls; they were kept in the original or an adjoining camp for the duration of the experiment.

# Experiment 3: Grazing Tribulus mainly (Camp 3)

Five recently shorn two-tooth Merino ewes and five young Merino wethers were placed in Camp 3, erected in the rested camp, during the afternoon of 17/3/71 and allowed to graze unrestrictedly. Water was supplied

in a trough and the supply replenished daily. The rest of the animals in the respective flocks from which these animals were selected, was either kept in the original or other camps as controls for this experiment.

# Experiment 4: Grazing Tribulus mainly (Camp 4)

Five recently shorn two-tooth Merino ewes and five young Merino wethers were placed in Camp 4, erected in Camp 2, during the afternoon of 17/3/71 and allowed to graze there continuously for the duration of the experiment. Water was supplied in a trough and the supply replenished daily.

As these animals originated from the same flocks as those in Experiment 3, the remaining sheep in those flocks also acted as controls to this experiment.

# Experiment 5: Introduction into a 'toxic' camp

The original flock of 129 sheep involved in the second outbreak was kept in Camp 2 for the duration of these experiments.

A total of 26 experimental sheep from elsewhere on the farm was introduced into, this flock during the morning of 17/3/71. These sheep were distinctly marked and subjected to daily individual examination, whereas the other members of the flock were only inspected closely.

This group of sheep consisted of ten recently shorn two-tooth Merino ewes originating from the same flock as those used in experiments 3 and 4, ten recently shorn mature Merino ewes from a distant 'non-toxic' camp and six Merino ewes of mixed ages previously referred to.

`The original flocks served as controls for the experimental sheep.

#### Experiment 6: Grazing Tribulus exclusively

Two young Merino wethers were put into a movable paddock at noon on 29/3/71. This paddock was placed in Camp 2 for one day only and then moved to the rested camp until this experiment was terminated. Water was supplied in a plastic container and the paddock moved once or twice daily in order to provide adequate feeding material. The paddock was carefully placed on each occasion to ensure the enclosure of as few other plants as possible. These other plants were meticulously removed so that only *Tribulus* was left to be eaten.

Two Merino wethers from the same flock were again introduced into the movable paddock during the morning of 6/4/71 when this particular experiment was repeated.

On each occasion a similar number of Merino wethers selected as controls was placed into an open paddock at the Veterinary Investigation Centre and given lucerne hay only. These control sheep, due to practical problems encountered, could unfortunately not be kept on natural veld devoid of *Tribulus*,

#### RESULTS

#### General Observations

Although it was claimed that Camp 1, where the first outbreak had occurred, was entirely free of *Tribulus*, huge patches of this plant were found on the north-western side when the camp was properly surveyed. There was also sufficient evidence that sheep had concentrated in this part.

Camp 2 is traversed by a narrow water channel running almost from its northern to southern corner (Fig.). Almost the entire area east of this channel and a smaller south-western area were covered mainly with *Tribulus*. This cover extended to the adjoining rested camp for a distance of several hundred metres. The remaining areas of both camps were mainly covered by *Eriocephalus* spp. ('kapokbos').

The Tribulus-covered areas were densely populated by these plants, which ranged from pre-flowering to early seeding stages. It was noticed that the more advanced growth stages occurred in small furrows and waterways and, judged by their appearance, had grown more luxuriantly than those on higher ground between the water-ways (fingers of erosion), although they were all in a dry and wilted stage. All Tribulus were wilted and many even dry, their leaves and shoots were curled up and were of a dirty brown or purplish brown colour with a slight greenish hue. The majority of these plants were of the younger stages and carried only three to five shoots, at most approximately 15 to 20 cm in length. There was a striking difference in density and height of the Tribulus cover in Camp 2, when compared with the cover in the rested camp. In the former Tribulus was less abundant and grazed short,

whereas those in the rested camp were abundant, dense and undisturbed.

Towards the end of March the established Tribulus at first assumed a yellowishgreen and later on a darker green colour. The more advanced growth stages started flowering again, followed by the younger stages. The flowers, however, appeared premature and smaller than usual, probably owing to interrupted growth. Some patches, especially in hollows, furrows and on softer soil surface did not grow again and kept their brown or even dark-brown colour. It was assumed that these plants had been watered better, had grown quicker and had therefore completed their growth cycles.

Close observation revealed that sheep were extremely fond of *Tribulus* in all stages of growth but preferred the drier and more wilted stages to the green and flowering ones.

At the onset of these experiments, the sheep in Camp 2 were found on the *Tribulus* patches every day and at practically all times of the day. A few days after the first rains during the experiment, i.e. 23 to 24/3/71, these sheep changed their grazing habits and were feeding mainly on 'kapokbos' (*Eriocephalus* spp.) until 18/4/71, when they returned to the *Tribulus* area. They were then eating both types of vegetation as they were found grazing on the *Tribulus* patches in the forenoon and on 'kapokbos' during the afternoon.

The gradual change-over from one plant to the other, even before the rains, was probably due to overgrazing of *Tribulus*.

Observations on individual sheep in all the grazing experiments revealed that they were eating *Tribulus* almost exclusively. During long periods of close observation, sheep were seen grazing these plants only, passing by many other attractive and palatable plants, on their way from one *Tribulus* to the next. While observing the animals kept in the movable paddock, it was noticed that they would first eat at the dry patches until these were almost denuded before going on to the surrounding green and flowering stages.

#### Rainfall

The rainfall figures of two nearby camps for the latter half of 1970 and the first part

Table 1: RAINFALL EXPRESSED IN MILLIMETRES: 1.7.70—13.5.71

Date	'Boes	manskop'	'Per	dekamp'	A	erage/
	Rainfall	Monthly Total	Rainfall	Monthly Total	Average	Mean Monthly Total
16/ 7/70	12,2	12,2	10,7	10,7	11,51	11,51
28/ 8/70	116,5	116,5	105,1	105,1	110,8	110,8
12/10/70 26/10/70	6,2 6,5	12,7	5,4 9,1	14,5	5,8 7,8	13,6
16/11/70	5,5	5,5	6,2	6,2	5,9	5,9
7/12/70 21/12/70 23/12/70 28/12/70	49.3 8.8 1.5 1.7	61,3	25,1 8,0 1,6 2,1	36,8	37,2 8,4 1,6 1,9	49,1
8/ 1/71 11/ 1/71 25/ 1/71	8,3 11,2 10,0	29,5	8,2 6.7 4,1	19,0	8,3 9,0 7,1	24,4
5/ 2/71 8/ 2/71 11/ 2/71 15/ 2/71	13,1 31,6 4,6 5,0	54,3	19,5 21,2 3,4 4,8	48,9	16,3 26,4 4,0 4,9	51,6
19/ 3/71 24/ 3/71	12,4 35,5	47,9	12,7 30,2	42,9	12,6 32,9	45,5
2/ 4/71 5/ 4/71 8/ 4/71	27,4 30,5 7,4	65,3	15,5 31,9 6,0	53,4	21,5 31,2 6,7	59,4
3/ 5/71 11/ 5/71	2,5 21,5	24,0	1,5 18,5	20,0	2,0 20,0	22,0
Total		429,2		357,5		

of 1971 until termination of all the experiments are given in table 1.

Examination of these figures reveals that approximately six weeks elapsed from the last showers on 8/2/71 till the first rains in March. A similar dry period of four weeks existed between 7/12/70 and 8/1/71. Although a heavy downpour occurred during August 1970, as well as light showers in October and November with comparable dry spells in between, exceptionally cold weather was experienced almost continuously up to the middle of December. Furthermore, cloudy to overcast and cool weather persisted from the first week of April onwards.

Plant Survey

The results of a plant survey conducted in experimental Camps 3 and 4 on 25/3/71 are listed in table 2.

From these results it can be seen that the total plant count was slightly greater, while the *Tribulus* cover was distinctly higher in Camp 4 than in Camp 3. According to these figures, *Tribulus* constituted approximately 76% of the total canopy cover and 73% of the total canopy spread cover in Camp 4, while the corresponding figures for Camp 3 amounted to 82% and 81% respectively.

Table 2: PLANT SURVEY IN CAMPS 3 AND 4 ACCORDING TO THE DESCENDING POINT METHOD<sup>12</sup>
(500 READINGS PER CAMP)

PLANTS		CAMP 4			CAMP 3		
	Base	Canopy cover	Canopy spread cover	Base	Canopy cover	Canopy spread cover	
Ephemeral Weeds:							
Amaranthus thunbergii		1	1				
Atriplex semibacoata		5	6 8				
Chenopodium spp.		3	8		ł		
Salvia claudestina				1	j <b>5</b>	11	
Tribulus terrestris	8	62	107	8	77	139	
Total	8	69	122	9	82	150	
Grasses:			, ,				
Aristida namaquensis	1	ĺ	1		1		
Cynodon incompletus		4	6		_		
Total		4	7				
Palatable Bushes:		-	2		1	2	
Eriocephalus glaber	]				'		
Pentzia incana		1	1				
Total		1	3		1	2	
Unpalatable Bushes:			_     .				
Chrysocoma tenuifolia			2		4	1	
Lycium spp.		1	3		6	8	
Pentzia globosa	2	5	8		, ,	-	
Pteronia glauca					ı	9	
Zygophyllum incrustatum			<u> </u>		1	i	
Total	2	6	14		11	19	
Grand Total	10	80	146	9	94	171	
% Cover	2,0	16,0	29,0	1,8	18,8	34,2	

#### Symptoms observed

Complete descriptions of the symptomatology of geeldikkop are to be found in the publications of Theiler<sup>2</sup>, Steyn<sup>13</sup> and Brown<sup>1, 14,17</sup>. Although additions ware made by each subsequent worker, these descriptions have much in common and feature the different stages in the development of the typical disease, viz., erythema and oedema of the head and its structures, icterus, necrosis and sloughing of the affected skin. Other symptoms such as fever, rhinitis, conjunctivitis, keratitis, dyspnoea and coronitis were

usually present in a variable percentage of cases <sup>13</sup>, <sup>14</sup>. Although the symptoms of photosensitization and icterus could vary in severity, they were usually present in typical cases <sup>13</sup>.

Close observations on natural and experimental cases in our investigations, however, revealed the occurrence of different clinical manifestations. For the purpose of these investigations, the following classes or types based on the onset and development of the most typical symptoms were distinguished.

Class 1: Reddening and crust formation of the muzzle, nose and upper lip, without overt swelling of the head and ears. Appearance of red coronary band after 3 to 5 days and eventual slipper formation. Absence of clinical icterus.

Class 2: Acute swelling of ears and face, which usually subsides within twenty-four hours without discernable icterus, although it could be followed by other symptoms as well as icterus. Recovery usually without sloughing of the necrotic epidermal layer.

Class 3: Reddening of the muzzle, nose and upper lip, appearance of red coronary band and of icterus but no swelling of the face and ears. Recovery with sloughing of necrotic epidermal layer of the skin on the ears, around the eyes and on the bridge of the nose. Eventual slipper formation.

Class 4: Typical cases. Similar to Class 3 but also accompanied by severe swelling of the face and ears. Complete sloughing of the necrotic epidermal skin on the face and ears on recovery and slipper formation.

Class 1 seemed to be a mild and transient form, Class 2 moderate, sudden in onset and usually, but not invariably, transient, while Classes 3 and 4 might be similar and determined only by exposure to direct and intense sunlight. Icterus either preceded or followed the syndrome of photosensitization. This sequence was sometimes influenced by the weather (sunlight) and the time of the day during which the crisis was reached. The hoof lesions appeared either simultaneously with the face lesions or followed shortly afterwards. Other exposed areas, such as the lower legs and vulva, could also be affected. The eyelids were sometimes completely immobilized and glued together by exudate. Mortalities occurred mainly in Classes 3 and 4.

#### Experiment 1

None of these lambs showed any symptoms of geeldikkop during the entire period of observations, although all four developed contagious pustular dermatitis within six to nine days after injection of heparinized blood.

#### Experiment 2

The first positive case developed 13 days and the last one 33 days after commencement of this experiment. Altogether five of the ten experimental sheep were clinically affected, while none of the ten controls showed any symptoms. Details of these cases are shown in table 3.

Table 3: POSITIVE CASES: EXPERIMENT 2

Sheep No.	Clinical classification (Class)	Date of Onset	Latent period (days)
A70.7	3	29/3/1971	13
A70.84	2/3*	1/4/1971	1,6
A70.24	2/3	5/4/1971	20
A70.181	2/3	16/4/1971	31
A70.40	2	19/4/1971	34
	1	1	I

\*2/3: Intermediate between Class 2 and 3

In four out of the five sheep, with no distinct or visible symptoms, subclinical geel-dikkop was subsequently confirmed on histopathological examination (see *Pathology*), while the two slaughtered controls were negative. This gave a total incidence of 90%.

#### Experiment 3

Details of positive cases are given in table 4.

Table 4: POSITIVE CASES: EXPERIMENT 3

Sheep No.	Clinical classification (Class)	Date of Onset	Latent period (days)
A69.487	3	20/3/1971	3
A69.311	3	21/3/1971	4
A70.6	2	22/3/1971	5
A70.212	2 / 3*	25/3/1971 relapse on 14/4/71	8
A70.120	2/3	25/3/1971 relapse on 3/5/71	8
A69.535	3	27/3/1971	10
A69.669	2/3	26/3/1971 relapse on 24/4/71	9

\*2 / 3: Intermediate between Class 2 and 3

Altogether seven out of the ten sheep in this experiment developed typical symptoms of geeldikkop within a period of three to ten days after introduction into Camp 3. Furthermore, three of these positive cases which were left in the camp recovered and subsequently developed relapses from 20 to 39 days later. Although the three remaining sheep showed a slight yellowish tint of the conjunctivae at some time or other during the course of the experiment, they were regarded as clinically negative. Only one of these animals was autopsied; distinct liver lesions characteristic of geeldikkop were found (see Pathology). It was also confirmed as a subclinical case on histopathological examination. This gave an incidence of at least 80%.

#### Experiment 4

Details regarding the positive cases are given in table 5.

Table 5: POSITIVE CASES: EXPERIMENT 4

Sheep No.	Clinical classification (Class)	Date of Outbreak	Latent period (days)
A69.527	2	25/3/1971	8
A70.185	3	29/3/1971	12
A69.533	3	5/4/1971	19
A70.146	2	9/4/1971	23

Four out of ten sheep in this experiment developed distinct clinical geeldikkop.

Two other animals which had a slight yellow discoloration of the conjunctivae could possibly have had mild attacks but were nevertheless classed as clinically negative. Only one of them, Sheep A59.541, was autopsied and extensive liver lesions were found. It also proved to be positive on histopathological examination (see *Pathology*). Another animal from this group, which was completely negative on clinical examination, was also found to be suspicious on histopathological examination.

#### Experiment 5

From the onset and during the course of this experiment 41 sheep (excluding the missing one) were removed from the camp with clinical geeldikkop. Except for the 23rd and 24th of March when the flock was not

gathered on account of rain, cases were encountered every day until the 26th. Further cases occurred on the 29th and 30th but none developed during the rest of the experimental period.

During close inspection and examination of sheep in this flock on 18/3/71, a number of cases with red and crusted noses and red banded hooves were marked and examined daily. Only one of them developed clinical geeldikkop, while the others recovered completely except for peeling of the nose and slipper formation of the hooves.

None of the animals introduced into the flock became affected.

#### Experiment 6

One sheep had a light yellow tint of the conjunctivae on the third day and this became distinct and intense on the fourth day. Although this animal was severely sick, swelling of the face and ears was never obvious, probably owing to the low intensity of sunlight as result of the overcast weather prevailing at the time. The second animal was acutely affected on the seventh day, the whole of its face and its ears being swollen to a severe degree.

The second pair of animals was not distinctly affected except for reddening and slight swelling of the nose in one of them (Sheep A70.537). On post-mortem examination some two months later this sheep had a distinct red band and the other one a distinct ridge on all four hooves, as well as substantial liver damage. Both animals were positive on subsequent histopathological examination (see Pathology).

#### Pathology

A comprehensive comparative study of the pathology of geeldikkop and related conditions of photosensitization and hepatosis has been undertaken and is still in progress. Hence only a short summary of the most significant findings to date will be given.

Several contributions have been made in this field 1, 2, 17; although most of the described lesions were noticed, our observations are more in accordance with those of Theiler 2.

A critical analysis at this stage, however, is not indicated: it suffices to mention the

most prominent features. As the lesions in natural and experimental cases were indistinguishable microscopically except for degree of severity, no distinction will be made between the two groups.

Liver: Varying degrees of bile duct proliferation accompanied by fibroplasia, mild to moderate degeneration of the hepa-(swelling and vacuolar changes with scattered necrosis of individual cells), Kupffer cell mobilization and proliferation, pigmentation (bile, lipofuscin and iron) and the formation of polarizing crystalloid, cholesterol-like material in the hepatocytes, Kupffer cell mobilization and proliferation, very regular and characteristic microscopic features in the pathogenesis of the disease. Most of the cholesterol-like crystalloid material is dissolved in haematoxylin and eosin stained sections and is represented only by clefts of typical shape. On the other hand, a small amount of polarizing material, unlike cholesterol, frequently remains undissolved in the portal areas. Hence it is considered not to be cholesterol. Some bile ducts are often occluded by this material and this is sometimes associated with necrosis of the duct epithelium. It is evident that these crystalloid bile plugs or 'thrombi' are responsible for more pronounced patchy lesions which are macroscopically characteristic of the more advanced and more severe cases. This results in irregular, patchy fibrosis in which small circular lesions eventually may lead to pit formation and the more linear lesions to folds. The lobulation is usually distinct, especially in more advanced cases. This is due to the portal lesion mentioned previously. It is also evident that the colour varies considerably, depending on the degree of pigmentation (vide supra) and progression of the lesions. It ranges as a rule from yellowish-brown to ochrish-brown, often with irregular lighter patches of varying sizes.

Very mild to mild megalocytosis is sometimes noticeable and mononuclears, neutrophils or eosinophils may be present, especially in the portal areas. Vascular lesions are absent or exceptional and very mild. Fatty changes are infrequent (±14%) and usually mild. In subacute and chronic cases large purplish-brown or greyish-brown pigment-laden macrophages are present and usually more numerous around the proliferative portal lesions.

Kidneys: The colour varies from light brown to greenish-brown, sometimes medium

brown in chronic cases. Many olive-green and dull grey spots are frequently present and the surface may be uneven owing to cystic changes. The kidneys may also be swollen.

Pigmentation and nephrosis, characterized by cystic dilatation of a number of tubuli, are present on microscopic examination. Proteinaceous, pigment and crystalloid casts are found during various stages of the disease. Tubular epithelial hypertrophy and hyperplasia are evident in many cases and mild fibroplasia is seen in some chronic cases. The presence of crystalloid material is an inconstant feature and is mainly seen in some of the more severe subacute and chronic cases. As in the liver, the incidence of crystalloid material is considerably more apparent after staining of frozen sections with oil red-O. There is no evidence of haemoglobinuria.

Myocardium: Sometimes small foci of degeneration and very mild mononuclear infiltration are noticeable. This is appreciable macroscopically in exceptional cases only.

Skin and coronet: Typical necrotizing lesions of photodynamic dermatitis are present in which usually some vessels in either cutis or subcutis are affected by eccentric hyaline changes. Medium-sized and fairly large vessels are also frequently affected. Oedema may be very pronounced in the acute stages and mild haemorrhages are present. The dermal and coronary papillae are particularly prone to damage. Slipper formation in the hoof is very frequently noticed in cases older than one to two weeks.

Urinary bladder: Except for localized foci of dedema in a few cases, no specific lesions are noticeable.

#### DISCUSSION AND CONCLUSION

The only remark that can be made on the first outbreak is that, contrary to initial reports, considerable areas of *Tribulus* were found in Camp 1. A similar experience was also reported by Theiler<sup>2</sup>.

With regard to the second outbreak, the abundance of this plant was very evident. Furthermore, these plants were mostly in the pre-flowering stages and considerably wilted. The fact that the sheep were constantly found grazing on the *Tribulus*-infested areas during the first part of the experiment

and only sought alternative grazing when these areas were almost denuded, seems to be convincing evidence of their preference for this plant. This is in accordance with Theiler's <sup>2</sup>, Quinn's <sup>3</sup> and Brown's <sup>9</sup> observations. The cut and overgrazed appearance, so strikingly different from the more abundant and longer growth in the rested camp, also supports this view.

Close observations of the grazing habits proved beyond doubt that sheep are exceedingly fond of the *Tribulus*, and indeed preferred the dry, wilted, early stage to the luxuriant, green, advanced stage. These findings are supported by the observations of Brown<sup>9</sup>, who incriminated the presence of the thorny seeds as a repelling factor in the preference by sheep for this plant. The observations also afforded evidence that whatever the active, causative agent of geel-dikkop, *Tribulus* should be the plant to be studied first.

The wilted appearance of Tribulus during the period of observation was evidently caused by the low rainfall figures for that particular area and period, by the prevailing heat, and possibly other climatic conditions. The fact that Tribulus did not make an earlier appearance, despite ample late winter and spring rains, can be accounted for by the almost continuous cold weather experienced till mid-December of 1970. A good downpour early in December followed by a hot and relatively dry period, despite the occurrence of scattered showers, appeared to have been favourable for growth and subsequent withering of Tribulus prior to the first outbreak of geeldikkop. Likewise the second outbreak of the disease and the appearance of Tribulus were also preceded by a similar set of conditions. The ample rains occurring in early February followed by a relatively dry and hot spell, lasting approximately six weeks, could be held responsible for the initial growth and subsequent wilting of the plants.

The particular association between the 'fate or well-being' of *Tribulus* and the incidence of geeldikkop as indicated above, and specifically stated by Theiler<sup>2</sup>, Quin<sup>3,4</sup>, Brown<sup>9</sup> and a multitude of stock owners, suggests an interdependent relationship rather than mere coincidence.

The different clinical manifestations recorded during the current studies are by no

means new, as variations of symptoms, as well as the occurrence of subclinical cases, have already been described previously 1, 2, 14. Whereas these workers have described mainly the sequence of events in a typical case of geeldikkop, our observations seem to indicate that different forms of the clinical disease does occur. These various forms were encountered in both natural and experimental cases. It should be emphasized, however, that the four basic symptoms in our classification, viz., reddening and crust formation of the nose and muzzle, swelling of the face and ears, reddening of the coronary band and eventual slipper formation (all three representing the syndrome of photosensitization) and icterus, are all present in a typical case, while only some of them are found in the other types. This variability evidently reflects the degree of affliction, which is influenced by the amount of toxic substance ingested and frequency of ingestion, individual susceptibility, intensity of daylight irradiation 14, the levels of circulating photodynamic agents 8, 15 and bile pigments 15, 16. It has been noticed frequently that there need not necessarily be a direct correlation between the severity of photosensitization and the severity of hepatic lesions 1. Furthermore, reddening and crust formation of the muzzle, nose and upper lip, without any swelling of the face and ears, especially in mild cases, were not clearly mentioned by previous workers 1, 2, 9, 13, 14, while coronitis and evential slipper formation, described in only 36% of cases 14, were constant features in our observations.

The results of Experiment 1 are in accordance with those obtained by previous workers 2. 10, 11, inasmuch as inoculation of blood from acutely affected cases failed to cause or transmit the disease, and hence afford some evidence against a possible infectious agent.

The development of clinical geeldikkop in five out of ten sheep fed exclusively on *Tribulus*, as against the absence of symptoms in all the control animals fed on lucerne hay, undoubtedly incriminates this plant as an aetiological agent. These results are in agreement with those obtained by Theiler<sup>2</sup>, except that in his experiments the first cases occurred after nine days, while all cases studied by us occurred within 13 to 34 days. The more protracted onset and course of the disease in the present experiments may

be attributed to the fact that freshly collected material was not fed from the start. This offers some explanation for the unsuccessful attempts <sup>2, 3, 4</sup> at producing geeldikkop by feeding or drenching material transported from elsewhere or dried beforehand. The cloudy and overcast weather also could have influenced the production of a toxic substance and the appearance of the syndrome of photosensitization.

Although the camps involved in Experiments 3 and 4 contained vegetation other than *Tribulus*, a plant survey clearly indicated the overwhelming incidence of this plant in both camps. The paucity of other edible plants present, and the marked preference shown by sheep for *Tribulus* at that stage, further stress the significance of this plant and clearly support the finding of Theiler<sup>2</sup> and Quin<sup>3</sup>.<sup>4</sup>, who incriminated *Tribulus* as the aetiological agent in geeldikkop.

Explanation for the higher incidence of the disease in Camp 3 seems obvious, if considered that it was situated in a rested camp with more abundant and dense growth of *Tribulus*.

Another important observation from Experiment 3 was the fact that three sheep contracted the disease twice within 20 to 39 days. This supports the observations of previous workers<sup>2, 9</sup>, and presents evidence against a possible infectious cause.

Failure of the experimental animals in Experiment 5 to develop geeldikkop was not unexpected and, in view of the specific circumstances, was fairly obvious. Considering the small size of the remaining areas covered by *Tribulus* and the rapid overgrazing, followed by a subsequent change in feeding habits of the original flock, the total intake of *Tribulus* could have been inadequate. The small number of experimental animals also would have been dominated in their choice of food by the gregarious feeding habits of the original flock.

The response within 3 to 7 days of the first two animals kept in the movable paddock (Experiment 6) affords overwhelming evidence of the importance of *Tribulus* in causing geeldikkop. The lesions in both animals were also identical to those of uncomplicated natural cases of the disease. As all other plants were removed and only this plant offered, the objections that could be

raised against the other grazing experiments are completely eliminated. The reason why the second pair of sheep developed only very mild or subclinical geeldikkop evidently lies in the complete climatic change at that time, which could have affected both the physiology of the plant and the surrounding or associated microflora. The appearance of a more luxurious stage of *Tribulus* also supports the view that the wilted, pre-fruiting stage is usually involved in causing geel-dikkop<sup>3</sup>. <sup>4</sup>.

Although only distinct clinical cases were taken into account, it should be mentioned that, with few exceptions, the clinically negative or doubtful cases in the specific grazing or feeding experiments had distinct macroscopic and microscopic liver lesions. These lesions were indistinguishable from those observed in clinical cases. It suffices to state that a higher incidence of clinical cases would have occurred if suitable weather conditions would have prevailed.

Considering the pathology in general at this stage where the study is still in progress, it can be stated that certain characteristic features of geeldikkop apparently distinguish it from other known disease entities such as facial eczema 18 and enzootic icterus 1, 19. In the small number of natural and experimental cases of facial eczema examined 20, the polarizing, cholesterol-like material of geeldikkop was absent. Also in facial eczema additional characteristic lesions are known to occur in the vessels of the liver and in the mucosa of the gall bladder and urinary bladder 18. The crystalloid material is also absent in enzootic icterus, whereas a haemolytic crisis accompanied by severe haemoglobinuria is a constant feature in the latter 1, 19. The so-called 'dikoor' syndrome 21 which occurs on artificial grazing and old lands, differs from geeldikkop only in degree of severity. In all, it is a milder syndrome but with all the typical histopathological changes including the crystalloid material so characteristic of geeldikkop.

In view of the information obtained, it can be concluded that *Tribulus* contains a toxic agent during certain specific climatic conditions which produces geeldikkop. Initial good rains after a period of drought, followed by a spell of hot, sunny weather seem to be prerequisites for severe outbreaks. Wilted *Tribulus* proved to be at the most toxic stage

and hence it is suggested that the toxic agent is produced by the plant under these circumstances. This may be coincidental, however, as such climatic conditions will also affect possibly associated microflora 20, 22, such as fungi, be they parasitic 2 or saprophytic 20, 22. In the latter instance *Tribulus*, being a surface creeper with markedly hairy leaves and stems, could also efficiently act as a trap for fungal spores 22. It can therefore be postulated that either an unstable aberrant metabolite of the plant itself, or a mycotoxin contained in spores adhering to the hairy plant, or one produced by a

parasitic or saprophytic fungus is responsible for the disease.

#### ACKNOWLEDGEMENTS

The generous co-operation of the Director, Karoo Region and staff in providing experimental sheep and necessary facilities, and the whole-hearted support of the Director, Veterinary Research Institute, Onderstepoort are greatly appreciated. The assistance of the technical staff of the Section of Pathology at this Institute and at the Veterinary Investigation Centre, Grootfontein has been invaluable in these studies.

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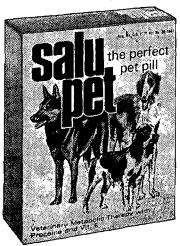
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# THE TOXICITY OF THE POLLEN BEETLE ASTYLUS ATROMACULATUS BLANCH.

T. S. KELLERMAN, T. F. ADELAAR AND J. A. MINNE\*

#### SUMMARY

Homogenized pollen beetle, Astylus atromaculatus Blanch., suspected of causing an outbreak of diarrhoea amongst cattle in Marico, were dosed per stomach tube to sheep and guinea-pigs. Some of the experimental animals developed enteritis and some of the affected ones died. The unidentified toxic principle does not appear to be cantharidin.

#### INTRODUCTION

Few insects are known to be harmful when ingested by domestic animals. Cocoons of the mopani tree moth, Gonometa rufobrunnea Aur. and Molopo moth, G. postica (Walker) can cause ruminal impaction. Saliva of locusts and possibly also the saliva of army worms, Laphygma exempta (Walker) can poison pastures and pupae of the moth Nudaurelia cytheria Fabr. may be toxic to pigs 3.

Steyn' reported the case of a child who died of heart failure after eating a grasshopper, *Phymateus leprosus* Fabr. He produced a similar condition by administering the minced grasshopper to rabbits, and concluded that the grasshopper was either toxic per se or that it became toxic after feeding on a cardiac glycoside containing plant such as *Nerium oleander* (Afrikaans: Ceylonroos) or *Asclepias fructicosa* (milkweed). Subsequent workers showed that the cardenolides present in body tissues of the North African grasshopper, *Poekilocerus bufonius* Klug. were similar to those of the milkweeds on which it fed.

The only other poisonous insects in South Africa (apart from bees, wasps, etc.) are beetles (order Coleoptera). According to Koch 6, larvae of the following beetles (Chrysomelidae) are used by bushmen to poison arrows: Polyclada flexuosa Baly., Diamphidia nigro-ornata Stal. and D. vittatipennis Baly.

The Chrysomelid larvae are parasitized by larvae of the beetles Lebestina peringueyi Liebke., L. subcruciata Fairm. and L. holubi Peringuey, of the family Carabideae. These ectoparasites are equally, if not more, toxic than their hosts. The as yet unidentified arrow poison is thought to immobilize game by acting on the central nervous system.

Blister beetles of the family Meloidae contain a severe blistering agent, cantharidin 7. Animals sometimes contract cantharidin poisoning when they accidentally ingest blister beetles, e.g., cattle grazing in the cooler parts of day when the beetles are sluggish and dogs that snap at flying insects. The family is well represented in South Africa and includes *Mylabris oculata* Thunb, the well-known C.M.R. beetle (named after the Cape Mounted Rifles).

In this study it will be shown that Astylus atromaculatus Blanch. of the family Dasytidae, popularly known as the pollen beetle or spotted maize beetle (Afrikaans: bont mieliekewer), is toxic to sheep and guineapigs. Its characteristics are also given.

#### IDENTIFICATION OF BEETLE

A. atromaculatus was introduced into South Africa from South America, and has become widely distributed in the country. It is a small beetle, approximately a centimetre long, with black markings on yellow elytra. Superficially, pollen beetles bear some resemblance to the C.M.R. beetle (Figs 1 and 2) but can easily be distinguished in the following way 8:—

- (a) Pollen beetles are smaller than most C.M.R. beetles and have different markings.
- (b) The elytra of pollen beetles cover the entire abdomen. In the case of the C.M.R. beetle the posterior tip of the abdomen is visible when viewed from the dorsal aspect.
- (c) Pollen beetles occur together in great numbers, whereas C.M.R. beetles are solitary.

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Fig. 1. Pollen beetles clustering on a flower.

#### LIFE CYCLE

The eggs are laid under dry leaves, where, within about two weeks, the larvae hatch.

Larvae feed principally on decomposing organic matter and hibernate in the soil. In summer, after a pupal period of approximately two weeks, the adults emerge. Only one generation is completed in a year 8.

The adults are strong fliers, feed mainly on pollen, and are most abundant in the period January to March, when they are frequently seen clustered in masses on a variety of flowers, grasses and crops such as maize and sunflowers.

The beetles are sluggish early in the morning and evening when it is cool and, if disturbed, make little attempt to fly away. Towards noon when it is hot they become more active but large numbers may still be seen on flower heads.

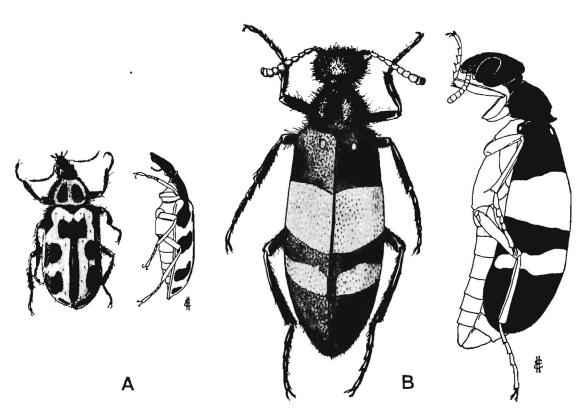


Fig. 2. Scale drawing of a pollen beetle (A) and C.M.R. beetle (B) (approximately life size × 5).

#### HISTORY

Our attention was first drawn to the possible toxicity of *A. atromaculatus* in January, 1970 when it was reported from the Marico district that 20 cows in a herd of 24 had developed diarrhoea while grazing on a land of sunflower regrowth. Four of the affected cows died.

Mr. W. C. Koker, of the Division of Veterinary Services, Mafeking described the clinical signs as follows: severe mucoid diarrhoea, slight bloating, reluctance to rise when lying down, and congestion of the mucous membranes. Some cows groaned periodically, as if in pain, and those lactating dried off.

The cattle had grazed in the land without mishap for 6 weeks. During the outbreak, which occurred a few days after a heavy shower, many pollen beetles were seen clustering on the sunflower heads.

Old inhabitants of the district claimed that similar outbreaks had occurred before when the beetles were present.

TOXICITY TRIALS ON GUINEA-PIGS
ORAL ADMINISTRATION OF BLENDED BEETLES

#### Method

Live pollen beetles from the Marico district were blended in distilled water and dosed per stomach tube to mature female guinea-pigs of live mass c. 750 g. The following dosage rates were used:—

MASS POLLEN BEETLE DOSED PER GUINEA-PIG (g)
Trial A 5.0\* 2.5\* 1.25 0.5 0.2
Trial B 0.25 0.187 0.125\* 0.0625
\*Two guinea-pigs were used in these groups. Other groups each contained one.

#### Results

All the guinea-pigs in Trial A died within 24 hours. Those in Trial B showed no apparent ill effects.

Clinical Signs: The affected animals developed anorexia, became listless and died. No diarrhoea was observed.

Necropsy Findings: The most marked feature was a severe, acute, haemorrhagic enteritis affecting the small intestine. The stomach and large intestine appeared normal but in some the caecum was affected.

The lungs were frequently congested and more rarely oedematous. In most cases the livers were swollen and degenerated.

No macroscopic lesions were observed in the urogenital tract.

Histopathological Findings: Examination of the most affected organs confirmed that damage was greatest to the intestine and liver. A haemorrhagic enteritis was present and there was fatty degeneration with a few disseminated foci of early necrosis in the liver. Little pathological change was observed in the urogenital tract, apart from some cloudy swelling of the kidneys.

#### DERMAL TESTS

Subcutaneous injection of an aqueous suspension of 2,5 g ground-up *Astylus* beetle into two guinea-pigs produced no conspicious lesions.

Topical application of the suspension to shaved areas of skin over the abdomens of two guinea-pigs did not produce inflammation.

TOXICITY TRIALS ON SHEEP

TRIAL 1

Method

An aqueous suspension of pollen beetles collected in the Pretoria district was administered per stomach tube to three Merino ewes weighing between 28 kg and 33 kg. The following dosage rates were used:—

Ewe 1 received 1,2 g pollen beetle/kg body mass daily from Day 1 to Day 3. Treatment was suspended from Day 4 to Day 7 because the rumen was static. On Day 8, when the ruminal motility had returned to normal, dosing was resumed and continued until Day 23.

Ewe 2 received 3,0 g pollen beetle/kg body mass on Day 1, Day 3 and Day 4. Treatment was suspended because the rumen was static and, as ruminal motility did not return to normal, no further treatment was given. The sheep died on Day 9.

Ewe 3 received a single dose of 5,0 g pollen beetle/kg body mass. On Day 3 the rumen became static and remained so until the sheep died on Day 6.

The sheep were examined daily. Periodically standard chemical pathological determinations were done on the blood. These included blood urea nitrogen, creatinine, serum glutamic oxaloacetic transaminase, serum glutamic pyruvic transaminase, bilirubin, total plasma protein, glucose, serum calcium, sodium and potassium. Sheep that died were necropsied and specimens for histopathological examination were taken from various organs, fixed in 10% formalin, cut in a routine manner and stained with haematoxylin and eosin.

#### Results

Clinical signs: The interval between dosing and development of signs was 2 to 4 days depending on individual susceptibility and dosage rate. The first signs to be noticed were listlessness, anorexia, ruminal stasis, and a mild, almost inapparent, bloat. These were followed in the case of Ewes 2 and 3, by a severe, foul smelling, light green, mucoid diarrhoea. Ewe 1, which did not have diarrhoea, made a rapid recovery.

Chemical pathological findings: No significant changes from the normal were observed.

Necropsy and histopathological findings:

The most serious lesions were found in the intestinal tract. An acute catarrhal to pseudomembranous enteritis of the small intestine and, in Ewe 3, of the caecum was present. Microscopic examination revealed haemorrhage into the mucosa and a fairly severe neutrophil infiltration in the case of Ewe 3, while in Ewe 2 only hyperaemia, oedema and little cell infiltration were present. The forestomachs and colon were not macroscopically affected.

General venous congestion, slight enlargement of the liver (which was found histopathologically to be due to cloudy swelling and early hydropic degeneration), congestion of the meninges, and congestion of the lungs with mild oedema were the other main features of necropsy.

The urogenital tract appeared macroscopically to be normal but on histopathological examination cloudy swelling of the kidneys and, in Ewe 3, scattered pyknosis of the bladder epithelium were observed.

#### TRIAL 2

#### Method

A single administration of an aqueous Astylus suspension that had been stored at 4°C for 5 weeks was dosed per stomach tube to four sheep at 2 g, 3 g, 4 g and 5 g Astylus beetle/kg body mass, respectively. Blood samples were withdrawn periodically from the sheep; the same chemical determinations were done as in the case of Trial 1.

#### Results

At the lowest level of administration there was no response but at the other levels ruminal stasis set in at about 5 days after treatment. The rumens remained static for 8 to 11 days. No significant chemical patho-

logical changes were observed, there was no diarrhoea, and all the sheep recovered.

#### CHEMICAL TESTS: IDENTIFICATION OF THE TOXIC PRINCIPLE

#### Method

Dry, ground Astylus beetles were extracted with acidified chloroform (hydrochloric acid: chloroform, 2:100 v/v) by shaking at room temperature for 2 hours. Some of the beetle material was also refluxed in acidified chloroform for 2 hours.

The extracts were dried separately in a rotary evaporator and taken up in acetonitrile. Oils and fats were removed by washing three times with n-hexane.

The cleansed residues and a cantharidin standard were spotted onto Mercks H.F. 254/366 T.L.C. silicagel plates. The chromatograms were developed with a benzene: dioxane (80:20 v/v) mixture, chromogenized in iodine vapour and viewed under longwave ultra-violet light to intensify the cantharidin spots.

Cantharidin for the standard was extracted from C.M.R. beetles using the same technique.

#### Results

Cantharidin appeared as a single spot at  $R_f$  0,7 visible at concentrations above c. 5  $\mu$ g. No cantharidin could be detected in the Astylus beetle extract.

#### DISCUSSION

Although probably uncommon, poisoning with pollen beetles should be considered as a possible cause of diarrhoea in ruminants during the latter part of summer. As the beetles are attracted to pollen, special care should be taken, particularly when stock graze on harvested lands on which volunteer crops in the flowering stage are growing. The most dangerous times for grazing plants infested with A. atromaculatus are in the cooler parts of the day when the beetles are more sedentary and thus most easily ingested. These danger periods coincide with the peak grazing times.

The nature of the toxic principle is not known but chemical tests indicate that it is not cantharidin. The lowest single dose of the material from Marico that resulted in the death of a guinea-pig was approximately 0,27 g/kg body mass. In the case of sheep dosed with material from Pretoria the lowest single lethal dose was 5 g/kg body mass. This could either have been due to a species difference, the ruminant being less susceptible than a monogastric animal, or it might have been due to a difference in toxicity between insects from different localities. Storing the homogenized insect material at 4°C for 5 weeks appeared to reduce its toxicity.

The toxicity for bovines was not determined but it is highly probable that A. atromaculatus was responsible for the mortality and diarrhoea in cows at Marico.

#### ACKNOWLEDGEMENTS

We are grateful to:

1. Dr. I. Zumpt and Mr. W. C. Koker, Divi-

- sion of Veterinary Services, Mafeking, for bringing the case to our notice and supplying insects for the trials.
- 2. The following staff members of the Veterinary Research Institute, Onderstepoort: Dr. R. C. Tustin for undertaking the histopathological work; Mr. B. P. Maartens for his devoted technical assistance; Mr. A. M. de Bruyn and staff for photography; Dr. G. D. Sutton for supplying many of the beetles from the Pretoria district and Dr. T. W. Naudé for his interest and advice.
- Mr. E. Holm of the Plant Protection Research Institute for the pen drawing and assistance in taxonomic matters.

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#### HAEMOPHILUS CORYZA: THERAPY WITH SELECTED DRUGS

S. B. Buys\*

#### SUMMARY

The therapeutic effect of selected antimicrobial agents was compared in artificially produced cases of haemophilus coryza (Haemophilus gallinarum infection) in mature fowls. Sulphachlorpyrazine, a combination of sulphachlorpyrazine and dihydrostreptomycin, and sulphadimidine prevented the spread of the disease during the period of medication. A combination of chlortetracycline and sulphadimethylpyrimidine; was found to be less effective. None of the compounds used could sterilize all infected birds of infection.

#### INTRODUCTION

A number of drugs has been used in the treatment of haemophilus coryza with varying success. Sulphathiazole 4, sulphadimethoxine10, and chlortetracycline plus sulphadimethoxine 7 were chemotherapeutically active against H. gallinarum infections in chickens. Streptomycin<sup>2</sup> has also been found effective in the treatment of haemophilus coryza. A few strains of H. gallinarum resistant to streptomycin and dihydrostreptomycin have been found by Matsui et al 9. Erythromycin thiocyanate (poultry formula) afforded clinical relief to a significant number of H. gallinarum affected birds 11. Spectinomycin and combinations spectinomycin-erythromycin could not contain the infection, but there was a marked decrease in the number of clinically affected birds 5.

The object of this investigation was to evaluate the therapeutic and prophylactic effects of chlortetracycline-sulphadimethylpyrimidine combination and sulphachlorpyrazine in haemophilus coryza. For comparative purposes, sulphadimidine and dihydrostreptomycin sulphate, both currently used in this country to treat haemophilus coryza, were included.

#### MATERIAL AND METHODS

Mature White Leghorns obtained from the Onderstepoort flock, which has been closed and apparently free from haemophilus coryza for 20 years, were used in these studies.

The isolate of H. gallinarum (No. 6) used to infect the birds was isolated from a local field case. The organism was isolated on blood tryptose agar plates with Staphylococcus epidermidis as a feeder  $^3$ , then inoculated into a chicken meat infusion broth  $^6$  enriched with 5% chicken serum and finally propagated in the yolk sac of seven day embryonated eggs before being harvested and stored at -20°C.

A 24 hours broth culture, inoculated with stored yolk material, was used to infect birds. In Trials 1 to 8, 0,2 ml of the culture was injected into both infra-orbital sinuses. In Trials 9—12 only the right sinus was infected. The infra-orbital route was selected, as this ensures the development of clinical symptoms within 24 hours.

The experimental design is set out in table 1. Medication was applied to both experimentally infected and uninfected birds exposed to contact infection in Trials 1, 2, 3; 5, 6, 7; and 9, 10, 11. In this way both therapeutic and prophylactic effect could be assessed. Each trial group consisted of 18 birds, 12 infected and 6 non-infected. The birds were housed in three-tier battery laying cages-one bird per cage-with a total of six cages on each tier (see sketch 1). One water trough supplied the three individual cages of every row. In each tier birds nos. 1, 3, 4 and 6 were experimentally infected. Birds nos. 2 and 5 were left uninfected but exposed to infection from an experimentally infected bird on each side.

The birds of trial groups 4, 8 and 12 served as unmedicated controls. Those of Trial 4 were housed in the same manner as outlined above, and the same design for experimental and contact infection was followed. Trials 8 and 12 consisted of 16 birds per group, 8 infected and 8 non-infected.

<sup>\*</sup>Poultry Section, Veterinary Research Institute, Onderstepoort.

<sup>†</sup>Trade name: Esb<sub>3</sub>, Ciba Ltd., Basle.

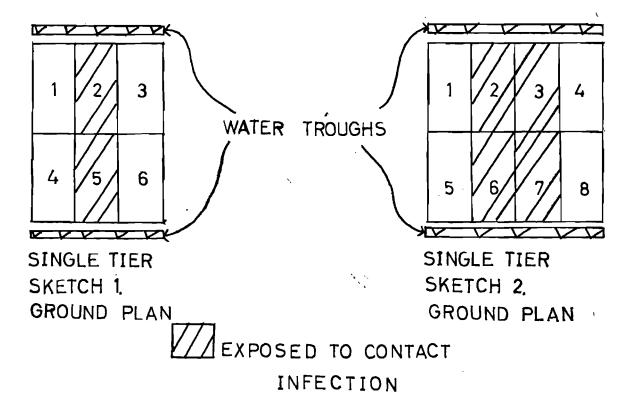
<sup>††</sup>Trade name: Aureo-Sulmet, American Cyanamid Company.

Table 1: EXPERIMENTAL DESIGN

Trial No.	Total no. of birds	Housing	Experimentall infected Cage No./	•	Non-Infect Contacts Cage No., Total no. of i	,	Drugs Used	Days of treatment starting on Day 1
1	18	3 Tiers	1, 3, 4, 6/	12*	2, 5/	6*	Chlortetracycline- sulphadimethyl-pyrimidine	1—5
2	18	3 Tiers	1, 3, 4, 6/	12	2, 5/	6	Sulphachlorpyrazine	15
3	18	3 Tiers	1, 3, 4, 6/	12	2.5/	6	Sulphadimidine	15
4	18	3 Tiers	1, 3, 4, 6/	12	2, 5/	6	No medication	No treatment
5	` 18   '	3 Tiers	1, 3, 4, 6/	12	2, 5/	6	Sulphachlorpyrazine	1, 2, 5, 6, 7
6	18	3 Tiers	1, 3, 4, 6/	12	2, 5/	6	Sulphadimidine	1, 2, 5, 6, 7
7	18	3 Tiers	1, 3, 4, 6/	12	2.5/	6	Diyhdrostreptomycin	1
8	16	2 Tiers	1, 4, 5, 8/	8† !	2, 3, 6, 7/	8†	No medication	No treatment
9	18	3 Tiers	1, 3, 4, 6/	12	2, 5/	6	Sulphachlorpyrazine	1, 2, 5, 6, 7
10	18	3 Tiers	1, 3, 4, 6/	12	2, 5/	6	Sulphadimidine	1, 2, 5, 6, 7
11	18	3 Tiers	1, 3, 4, 6/	12	2, 5/	6	Sulphachlorpyrazine- dihydrostreptomycin	1, 2, 5, 6, 7
12	16	2 Tiers	1, 4, 5, 8/	8	2, 3, 6, 7/	8	No medication	No treatment

<sup>\*</sup>See Sketch 1 Ground Plan.

<sup>†</sup>See Sketch 2 Ground Plan.



These birds were housed in two-tier battery laying cages with a total of 8 cages on each tier (see sketch 2). Each water trough supplied four individual cages. In each tier, birds nos. 1, 4, 5 and 8 were experimentally infected. Birds nos. 2, 3, 6 and 7 were left uninfected as contact birds.

The drugs included in this study were:

(1) Chlortetracycline (CTC)-sulphadimethylpyrimidine (S) (Concentration 100 g CTC+100 g S/kg) at a level of 250 ppm CTC+250 ppm S in the drinking water.

- (2) Sulphachlorpyrazine (30 per cent w/w powder) at a level of 300 ppm in the drinking water.
- (3) Sulphadimidine (33<sup>1</sup>/<sub>3</sub> per cent w/v solution) at a level of 2 220 ppm in the drinking water.
- (4) Dihydrostreptomycin sulphate (250 mg/ml) at a dosage rate of 250 mg per bird administered intramuscularly.

The day of experimental infection of the birds was designated as Day 0 in all 12 trials.

In Trials 1—4, continuous water treatment was carried out for a period of five days, beginning on Day 1. In Trials 5 to 12 water-treatment was furnished on Days 1, 2, 5, 6 and 7. Dihydrostreptomycin was administered on Day 1 only.

Individual daily records were kept of each bird, except for Days 9 and 10 in Trials 1 to 8

For the purpose of statistical analysis and plotting the graphs, each bird was given a 'score' according to the following criteria. Facial swelling (normal to severely affected: 0 to 3+), abnormal respiratory sounds (when present, one+) and general habitus and food consumption (normal 0 to maximum affected 4+). A dead bird was given the maximum score of 8+.

Only experimentally infected birds were used for evaluation of the therapeutic effect. In Trials 1 to 4 all four groups had 12 expe-

rimentally infected birds. Since the unmedicated groups in Trials 8 and 12 had only eight experimentally infected birds, these were compared in respect of 'score', as explained above, with eight out of 12 birds taken at random from the medicated groups. Those in Trial 8 were compared with 5, 6 and 7, and those in Trial 12 with 9, 10 and 11. To plot the graphs, the total 'score' of each group of birds was plotted against time. To evaluate the results statistically, Friedman's two-way analysis of variance by ranks, followed up by Nemenyi's test for differences between different treatments, was used 8

A post-mortem examination was done on all birds that either died during the trials, or were killed at the end of each trial. H. gallinarum isolations were made from the infra-orbital sinuses 3 in Trials 1 to 4 and 9 to 12

#### RESULTS

The rate of recovery of each group of birds in Trials 1 to 12 are recorded in figures 1, 2 and 3. The number of birds from which H. gallinarum could be isolated at postmortem examination in Trials 1 to 4 and 9 to 12 are tabulated in tables 2 and 3. In table 4 the percentages of birds in Trials 1 to 4 and 9 to 12 showing airsac lesions at post-mortem examinations are recorded.

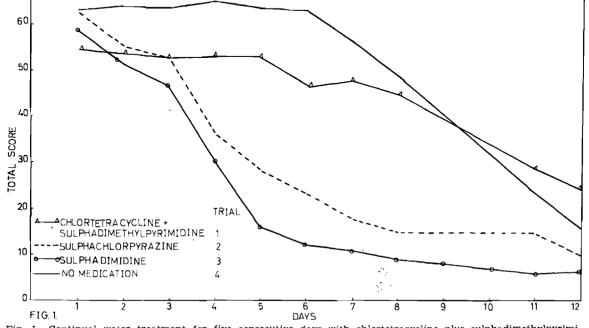
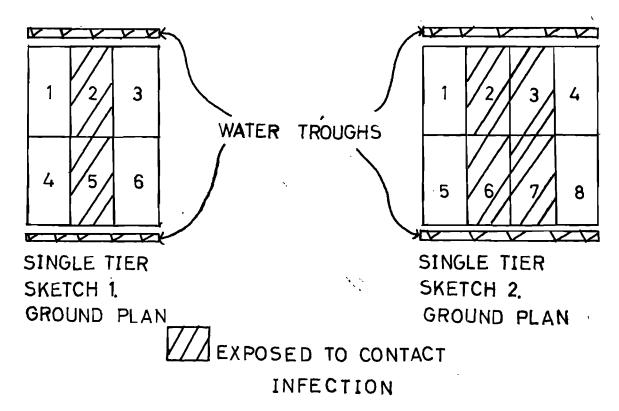


Fig. 1. Continual water treatment for five consecutive days with chlortetracycline plus sulphadimethylpyrimidine, sulphachlorpyrazine and sulphadimidine.

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4	18	3 Tiers	1, 3, 4, 6/ 12	2. 5/	6	No medication	No treatment
5	18	3 Tiers	1, 3, 4, 6/ 12	2, 5/	6	Sulphachlorpyrazine	1, 2, 5, 6, 7
6	18	3 Tiers	1, 3, 4, 6/ 12	2, 5/	6	Sulphadimidine	1, 2, 5, 6, 7
6 7	18	3 Tiers	1, 3, 4, 6/ 12	2, 5/	6	Diyhdrostreptomycin	1
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10	18	3 Tiers	1, 3, 4, 6/ 12	2, 5/	6	Sulphadimidine	1, 2, 5, 6, 7
11	18	3 Tiers	1, 3, 4, 6/ 12	2, 5/	6	Sulphachlorpyrazine- dihydrostreptomycin	1, 2, 5, 6, 7
12	16	2 Tiers	1, 4, 5, 8/8	2, 3, 6, 7/	8	No medication	No treatment

\*See Sketch I Ground Plan. †See Sketch 2 Ground Plan.



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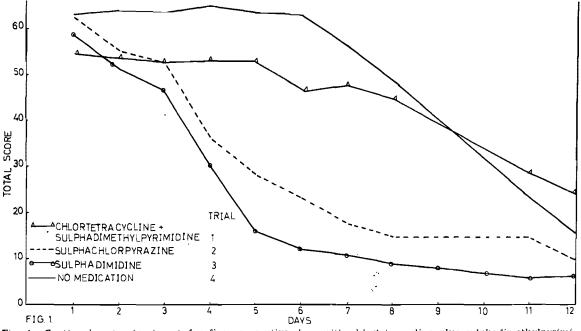
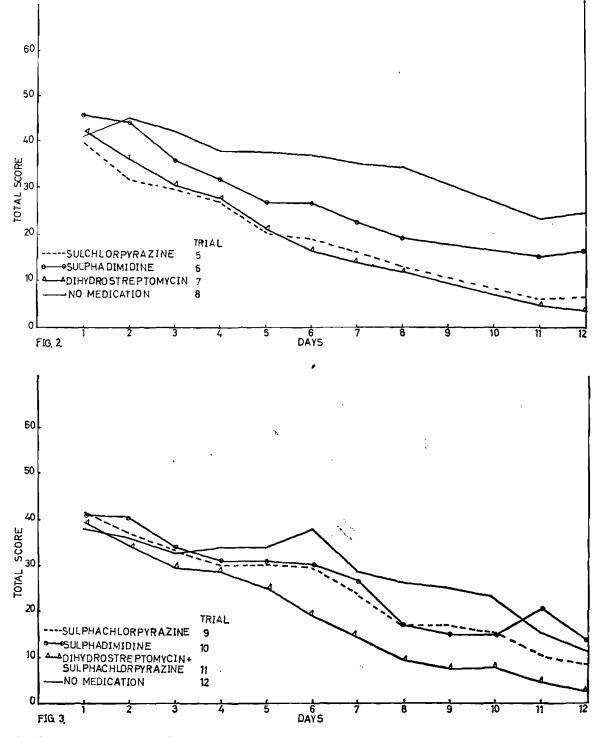


Fig. 1. Continual water treatment for five consecutive days with chlortetracycline plus sulphadimethylpyrimidine, sulphachlorpyrazine and sulphadimidine.



Figs. 2 and 3: Water treatment on Days 1, 2, 5, 6, and 7 with sulphachlorpyrazine and sulphadimidine. Dihydrostreptomycin was admi nistered on Day 1 only.

Table 2: STERILIZING EFFECT OF DRUGS. TRIALS 1-4

Trials		Infected (12 per		Non-in contact (6 per	birds
	DRUGS USED	H. galli isolations		H. gallinarum isolations at P.M.*	
_		Attempted	Pos.	Attempted	Pos.
1	Chlortetracycline and sulphadimethylpyrimidine.	12	8	6	3
2	Sulphachlorpyrazine 300 ppm	12	3	6	3
3	Sulphadimidine 2 220 ppm	12	4	6	4
4	No medication	12	10	6	3

<sup>\*</sup>P.M. = Post-mortem examination 12 days after infection.

Water treatment for five consecutive days from Day 1 with chlortetracycline and sulphadimethylpyrimidine, sulphachlorpyrazine and sulphadimidine.

Table 3: STERILIZING EFFECTS OF DRUGS. TRIALS 9-12

Trials		H. gallinarum isolations at P.M.*		Non-infected contact birds		
	DRUGS USED			H. gallinarum		
		Attempted	Pos.	Attempted	Pos.	
	·	12 per group		6 per group		
9	Sulphachlorpyrazine 300 ppm	12	1	6	3	
10	Sulphadimidine 2 200 ppm	12	6	6	6	
11	Dihydrostreptomycin sulphate (250 mg/bird) plus sulphachlorpyrazine 300 ppm	12	2	6	2	
12	No medication	8 per g	group	8 per group		
12	NO medication	8	2	8	2	

<sup>\*</sup>P.M.=Post-mortem examination 12 days after infection.

Treatment on Day 1 and 2 and on Days 5, 6 and 7 with water containing sulphachlorpyrazine and sulphadimidine. Dihydrostreptomycinsulphate administered on Day 1 only.

Table 4: INCIDENCE OF AEROCYSTITIS (TRIALS 1-4 AND 9-12)

,	Infected birds	Non-infected contact birds
,	Percentage with aerocystitis	Percentage with aerocystitis
No medication	33%	6%
Chlortetracycline and sulphadimethyl-pyrimidine	17%	17%
Sulphachlorpyrazine	4%	6%
Sulphadimidine	29%	25%
Dihyrdrostreptomycin plus sulphachlorpyrazine	0%	0%

The incubation period of haemophilus coryza is given by Yoder <sup>13</sup> as 1 to 3 days in susceptible birds exposed by contact to infected cases. With sulphachlorpyrazine, sulphadimidine and dihydrostreptomycin sulphate and sulphachlorpyrazine combined, coryza symptoms appeared 5 to 6 days after cessation of treatment in contact birds. With a combination of chlortetracycline plus sulphadimethylpyrimidine and dihydrostreptomycin sulphate, coryza symptoms appeared in contact birds 1 to 2 days after cessation of treatment.

Under the experimental conditions of these trials, the minimum period for clinical signs to develop after exposure in unmedicated contact groups was 3 days. Thus, it would appear that the first group of drugs could limit the spread of the disease during time of medication, but the second group could not.

None of the drugs tested could completely sterilize all experimentally infected birds of infection. (See Tables 2 and 3).

Appearance of haemophilus coryza symptoms after cessation of treatment has been found by several authors <sup>4, 10, 11</sup>. H. gallinarum could also be re-isolated from these birds <sup>10</sup>.

The data for each day (except Days 9 and 10, Trials 1 to 8) were statistically examined. A significant difference at the 0,05 level was found between:

- 1. Trial 4 (unmedicated) and Trial 2 (sulphachlorpyrazine) on Days 4, 5, 6, 7, and 8.
- Trial 4 (unmedicated) and Trial 3 (sulphadimidine) on days 4, 5, 6, 7, and 8.
- 3. Trial 8 (unmedicated) and Trial 5 (sulphachlorpyrazine) on days 3 and 7.
- Trial 8 (unmedicated) and Trial 7 (dihydrostreptomycin sulphate) on Days 6, 7, and 8.
- Trial 12 (unmedicated) and Trial 11 (dihydrostreptomycin sulphate plus sulphachlorpyrazine) on Day 6.

No significant differences could be found between:

- Trial 4 (unmedicated) and Trial 1 (chlortetracycline plus sulphadimethylpyrimidine).
- Sulphachlorpyrazine, sulphadimidine, dihydrostreptomycin, and dihydrostreptomycin plus sulphachlorpyrazine.
- 3. Any of the drugs on Days 11 and 12 of any of the trials.

The faster recovery of sulphachlorpyrazine and sulphadimidine treated birds in Trials 2 and 3 compared with Trials 5, 6, 9 and 10 may be due to the continuous treatment for five days in Trials 2 and 3, compared to Trials 5, 6, 9 and 10, in which the birds were treated on Days 1, 2, 5, 6, and 7, and this may warrant further experiments to determine optimum treatment schedules.

Sulphachlorpyrazine, according to Rüttimann and Heim <sup>12</sup> can be given to laying hens without causing any significant drop in egg production, fertilization rate or hatchability, whereas sulphadimidine, according to Bankowski <sup>1</sup>, can cause a decrease in egg production in laying flocks.

#### CONCLUSION

If recovery rates, percentage of birds showing aerocystitis, and *H. gallinarum* isolations are considered, it would appear that sulphachlorpyrazine, sulphadimidine and dihydrostreptomycin sulphate can be used to good effect to counteract the more severe symptoms of haemophilus coryza.

These experiments again emphasize the relative importance of chemotherapy in the control of *H. gallinarum* infection. The drugs used did not sterilize the birds from infection, but economic losses could be limited. Recovered carrier birds usually remain on a farm; they infect new introductions, and a new cycle starts. It is evident, therefore, that successful control should rely on stringent zoosanitary measures; the need for an effective vaccine is emphasized.

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#### **BOOK REVIEW**

BOEKRESENSIE

#### JONES'S ANIMAL NURSING

R. S. PINNIGER, EDITOR

Pergamon Press; Oxford, etc., 2nd Edition, 1972; pp xvi+471, Figs 135, Tabs 8. Price R13.25

This beautifully produced second edition has been edited for the British Small Animal Veterinary Association and contains contributions from twenty authors, two of whom are Registered Animal Nursing Auxiliaries. The experience of the six years which have elapsed since the first edition has been well utilized to update and expand the information presented. The book not only covers the requirements of the two-year syllabus for the Animal Nursing Auxiliary examinations of the Royal Veterinary College but often provides considerable additional information for those interested in further study. It may be mentioned here that the contents also cover all aspects of the syllabus for the twoyear Diploma in Animal Nursing proposed by the Faculty of Veterinary Science, Onderstepoort.

The ten chapters comprise anatomy and physiology, animal management, hygiene and feeding (including sections on cage birds,

small animals and genetics), first aid, diagnostic aids and laboratory tests, theory and practice of nursing (including sections on ethics, medical nursing, common diseases and dispensing), surgical nursing (including sections on anaesthesia and theatre practice), obstetrics and pediatric nursing, radiography, legal aspects and finally, a chapter on the veterinary profession. Most chapters are concluded with a bibliography for further reading. There are twenty-four black and white photographs among the figures. A useful table of biological data on some smaller mammals is provided. A comprehensive index is included.

While this book deals exclusively with small animals, this is not regarded as a defect. Information is presented in a lucid and comprehensive manner and the book is a pleasure to read.

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#### RIFT VALLEY FEVER

#### 1. VECTOR STUDIES IN THE FIELD\*

#### B. M. McIntosh\*\*

#### SUMMARY

During field investigations into possible vectors of Rift Valley fever in Southern Africa at a time when epizootics were occurring, or had recently occurred, this virus was isolated six times from Culex theileri, twice from Aedes lineatopennis and once each from Anopheles coustani, Aedes dentatus and Eretmapodites quinquevittatus. It was concluded that Culex theileri plays an important part as vector of this virus during epizootics in cattle and sheep in the highveld region of South Africa where it is prevalent. Aedes lineatopennis was numerically predominant in Rhodesia, where it might have been the main vector. Multiple isolations of Wesselsbron virus and two isolations of Middelburg virus were also made from this species.

#### INTRODUCTION

After an apparent absence of 13 years, Rift Valley fever (RVF) re-appeared in domestic animals in Southern Africa in 1969. In the summer of 1968/69 the disease appeared in a widespread epizootic in Rhodesia as well as in limited outbreaks in the coastal lowlands of Natal, South Africa and Moçambique.

In Rhodesia the disease occurred from January to May, during which a laboratory diagnosis was made on 127 farms while on a further 89 farms its presence was suspected on clinical grounds. As far as is known, these outbreaks involved cattle only and infection was most intense in the highlands around Salisbury but its presence was also established in the eastern lowlands at Chiredzi, Fort Victoria and Chipinga. An isolated outbreak also occurred in the extreme south-western region, near Figtree†.

In Moçambique the disease was diagnosed in March among cattle at Chibuto on the coastal plain about 200 km north of Lourenço Marques. Shortly afterwards outbreaks were diagnosed at João Belo at the mouth of the Limpopo River and near Vila Luisa, a coastal town about 30 km north of Lourenço Marques, where outbreaks were extensive. The last outbreak was diagnosed in July††.

In the autumn of 1969 there were outbreaks of abortion accompanied by low mortality, suspicious of RVF, among cattle near Empangeni and Hluhluwe in northern Natal†††. From an aborted foetus, submitted in July from one of the affected farms near Empangeni, RVF virus was isolated by Dr. P. G. Howell of the Onderstepoort Veterinary Research Laboratories.

Although extensive immunization had been carried out in Rhodesia in 1969, this had apparently not been done in South Africa and so it did seem likely that further outbreaks would occur here in 1970. The ovine and bovine populations in South Africa would probably still be largely susceptible, having experienced neither immunization nor natural infection on a large scale. In any event, no outbreaks occurred in Rhodesia in 1970, while in South Africa outbreaks were reported from the districts of Standerton, Frankfort, Kroonstad, Koppies and Odendaalsrus. During the following year there were several suspected outbreaks in the Orange Free State and one suspected case in Natal but none was confirmed according to the Annual Reports, 1967—71, of the Department of Agricultural Technical Services, Pretoria, Nevertheless, despite the probable absence or low incidence of the disease in 1971 in South Africa, RVF virus was isolated from mosquitoes collected at Port Shepstone in February of that year as will be described later.

Opportunities for vector studies in the field during epizootics of RVF in Southern

<sup>\*</sup>The studies and observations on which this paper are based were financed jointly by the South African Institute for Medical Research and the Poliomyelitis Research Foundation.

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Dr. H. M. Hodkin, Assistant Director of Veterinary Services, Rhodesia; in press

<sup>††</sup>Dr. F. G. Valadão; personal communication.

<sup>†††</sup>Dr. L. J. Kritzinger; in press.

Table: NUMBER OF INSECTS, INSECT POOLS, AND ISOLATIONS OF RVF VIRUS FROM VARIOUS LOCALITIES: 1969—1971

Locality/Date	Species	Total Tested	No. Pools	RVF Isolations
Rhodesia May 1969	Aedes lineatopennis Anopheles coustani Culex theileri Aedes dentatus 21 mosq. spp. Culicoides spp.	3,842 284 103 23 477 929	63 8 7 5 62 4	2 1 1 1
Kroonstad district March 1970	Culex theileri Anopheles squamosus Aedes caballus A. dentatus Anopheles coustani 3 mosq. spp. Culicoides spp.	1,398 267 18 3 3 72 522	47 12 5 3 2 13	4
Johannesburg February 1970	Culex theileri C. pipiens C. univittatus Aedes caballus 6 mosq. spp. Culicoides spp. Simulium spp.	144 4.833 744 1 48 339 476	3 15 16 1 6 2	1
Port Shepstone February 1971	Eretmapodites quinquevittatus Aedes demeilloni A. strelitziae Anopheles coustani 20 mosq. spp.	423 838 242 14 542	15 20 10 1 40	1

Africa have been limited in the past and only on two occasions have worthwhile observations been possible. In 1953 in the Luckhoff district, Orange Free State, Gear et al. obtained multiple isolations of virus from Culex theileri and Aedes caballus during an outbreak in sheep and also infected laboratory mice by the bite of naturally infected A. caballus. In 1956 RVF virus was again isolated from C. theileri during an outbreak of RVF in sheep in the Wesselsbron district, Orange Free State, according to unpublished data of the Arbovirus Research Unit.

The recent outbreaks provided further opportunities for vector studies in the field and with this object in mind visits were made to Rhodesia in 1969 and to the Kroonstad district, Orange Free State, in 1970. In addition to these studies, the circumstances surrounding the isolation of RVF virus from mosquitoes collected near Johannesburg in 1970 and at Port Shepstone, Natal, in 1971 are described.

#### METHODS

A variety of mosquito collecting methods was used. These included human and sheep

bait, light traps and portable suction traps for collecting off vegetation. Shortly after collection insects were killed and identified. Mosquitoes were pooled according to species and Culicoides and Simulium species according to genus. The insect pools were stored on solid carbon dioxide. Pools were processed in the laboratory and the suspensions inoculated intracerebrally into infant mice, two days old, for attempted virus isolation.

It is not the intention to describe in this paper the methods of virus isolation and identification. It is considered sufficient to mention that on the basis of haemagglutination-inhibition, complement-fixation and neutralization tests the strains of RVF virus isolated between 1969 and 1971 were immunologically indistinguishable from those isolated during the 1950's.

#### RESULTS

Rhodesia, 1969

From 20th to 28th May insect collections were made on seven farms within a radius of about 80 kilometres of Salisbury. On each farm the diagnosis had been confirmed by

laboratory methods by the Veterinary Laboratory, Salisbury. At the time collections were made, no further reports were being received of abortions but on one farm an aborted foetus was seen in the veld. Climatic conditions were dry, nights were cold and insect numbers on most farms were low and it seemed fairly obvious that, because of the onset of winter, the disease was subsiding at the time collections were made.

As shown in the table, Aedes lineatopennis was by far the most prevalent species. At Pearson, 15 km north-east of Salisbury, large numbers of this species were collected off human and sheep baits in low-lying marshy pasture shortly after sunset and the same species was also collected in fair numbers on most farms. Two strains of RVF virus were isolated from A. lineatopennis collected at Pearson. On the same farm this species also yielded 17 strains of Wesselsbron virus as well as two of Middelburg virus. Among mosquitoes, Anopheles coustani ranked second in prevalence and on a farm at Nyabira, 25 km north of Salisbury, this species yielded one strain of RVF virus. A strain of this virus was also obtained from each of C. theileri at Nyabira and Aedes dentatus at Pearson, although neither species was prevalent.

#### Kroonstad district, 1970

Following on information received from Dr. K. E. Weiss, Director, Onderstepoort Veterinary Research Laboratories, that RVF virus had been isolated from a bovine near Steynsrust, insect collections were made on four farms in the area from 18th to 24th March, including the farm 'Berlyn' on which the isolation had been made.

At the time collections were made, conditions were extremely dry. Only 251 mm of rain had fallen since the previous November, as against an average of 442 mm for the same period in previous years. The effect of this was evident in the paucity of Aedes species collected, a genus well-known for its dependance upon heavy rains. On the other hand, C. theileri and Anopheles squamosus, breeding in relatively permanent water, were still prevalent.

On 'Berlin' C. theileri was present in large numbers, particularly in an irrigated

lucerne field located near where the infected dairy herd was pastured for the night. Four strains of RVF virus were isolated from *C. theileri* collected on this farm, the only isolations made.

#### Johannesburg, 1970

On a sewage farm on the outskirts of Johannesburg, this Unit has maintained a field study on arthropod-borne viruses where insect collections have been made on a routine basis for several years. The table shows collections during February, 1970, during which month a strain of RVF virus was isolated from C. theileri. These collections, not intentionally concerned with RVF, were designed to collect a bird-feeding species, Culex univittatus, and the small number of C. theileri collected is probably not truly representative of the large numbers of this species generally present on this farm. There is a large herd of cattle on the sewage farm and, unknown to us at the time, a calf had died with symptoms suggestive of RVF, but this diagnosis could not be confirmed by isolation of the virus from the organs of the calft.

#### Port Shepstone, 1971

During January, 1971, a mild febrile illness had occurred among human beings near Port Shepstone on the coast of Natal. Mosquitoes were prevalent at the time and a mosquito-borne virus was suspected as the cause. To investigate this possibility, mosquitoes were collected in the area from 3rd to 10th February and tested for virus. As shown in the table, one strain of RVF v rus was isolated from Eretmapodites quinquevittatus. This was the most prevalent mosquito species in coastal bush at Cslo Beach, where the illness had occurred. The species fed readily on man but serological tests on sera from several convalescent patients showed that they had not been infected by RVF virus and there were also no reports of outbreaks in domestic stock at the time. There is, therefore, no apparent reason to associate the infection in E. quinquevittatus with human or domestic animal infection.

#### DISCUSSION

Until mosquito species can be excluded as vectors by experimental proof of their inability to transmit virus, all species found

<sup>†</sup>Dr. W. J. Ehret, Johannesburg Municipality; personal communication.

infected in nature clearly come under suspicion as possible vectors. On the other hand, cattle and sheep circulate RVF virus at high concentrations in the blood, thus it is to be anticipated that during epizootics a variety of species feeding on these animals will be infected without all of them necessarily being important vectors. In addition to infection, supportive evidence in the form of feeding habits, prevalence, geographical distribution and vector capability should also be considered. In attempting to assess the vector status of species it should be borne in mind that epizootics are the result of an abnormal situation in the ecology of the virus, usually an indication that the virus has fortuitously gained access to an incidental vector which feeds readily on cattle and sheep but which is not normally concerned in virus maintenance. Furthermore, it is more than likely that different species would participate as vector in such diverse habitats as exist on the South African highveld, the Rhodesian highlands, and the subtropical coastal lowlands of Mocambique and Natal. Varying climatic conditions in the same region may also influence the species involved at any particular time. For instance, during dry spells, a Culex species is more likely to assume the dominant rôle, while Aedes species might become dominant after heavy rains have resulted in high populations of these species.

The isolation of RVF virus during the present investigations from C. theileri in Rhodesia, as well as in the Kroonstad and Johannesburg districts is in accord with observations made during the outbreaks in the Luckhoff district in 1953 and in the Wesselsbron district in 1956, when C. theileri was also found to be infected. Further evidence implicating this species are the facts that it feeds readily on cattle and sheep and has been shown experimentally to be an efficient vector2. Hence on the South African highveld where this species is both widespread and numerically predominant, the indications are strong that it plays an important rôle as vector of RVF virus during epizootics. Furthermore, since this species is relatively drought-resistant and adults have been collected during winter on the highveld3, it also seems that this species could have been the means by which RVF virus had survived the winter on the highveld over a limited number of years after past epizootics. The real numerical status of C. theileri in Rhodesia is not known. During the time collections were made, it was rare and, because of this, it is possible that in Rhodesia it may not be as important as in South Africa.

The field observations in Rhodesia suggested that A. lineatopennis may have been the dominant vector. It was the most prevalent species, was widespread and was observed to feed readily on sheep, from which it may be inferred that it would also feed on cattle; it also yielded two strains of virus, Although A. lineatopennis is relatively shortlived in the adult stage, good rains had fallen during the summer of 1968/69 in Rhodesia and large numbers of this species were still present as late as May, so it is quite possible that, on the highlands at least, this species was sufficiently prevalent throughout the summer to have made it possible for it to have been the main vector. It is unfortunate that the present lack of information on the vector capability of this species precluded a more definite conclusion on the actual rôle of this species in Rhodesia.

As mentioned earlier, A. caballus was implicated as a vector during the outbreak in Luckhoff district in 1953. During the recent outbreaks this species was not collected in Rhodesia and, probably because of the prevailing drought, it was rare in the Kroonstad district. From this it seems unlikely that it could have been a significant vector on either occasion. A further complication with regard to this species is that it has recently become apparent that what is at present called A. 'caballus' probably consists of two species. Both forms have been collected at Hopetown, near Luckhoff, so there is at present doubt as to which species was actually implicated during the Luckhoff studiès.

Since no observations were made on the coastal lowlands of Moçambique and Natal during the recent outbreaks, comments on possible vectors there must remain largely speculative. Because of its known scarcity in the region, C. theileri can be excluded from any major rôle. Mr. J. de Sousa, Brigada para a Erradicacão do Paludismo, Moçambique, has informed me that common species collected in infected areas in Moçambique included Aedes durbanensis, Aedes circumluteolus, Culex thalassius, Culex poicilipes and Anopheles coustani. At one locality, A. circumluteolus predominated and this species is known to be common in the coastal

zone of northern Natal where RVF virus was isolated twice from it in 1955. Despite failure to demonstrate transmission with this species in recent tests<sup>2</sup>, it seems possible that A. circumluteolus may have been a vector where it occurred in very large numbers.

While the isolation of RVF virus at Port Shepstone in 1971 may merely be the outcome of extensive virus dispersal as a result of the widespread outbreaks in the previous two years, the implication of an Eretmapodites species is noteworthy because another member of this genus not present in South Africa was incriminated as a vector of RVF in Uganda in 1944. There are suggestions in the isolation at Port Shepstone that a forest maintenance transmission cycle may exist in the coastal region of Natal, although transmission experiments with E. quinquevittatus, described in the next paper, indicated that if this were so, this species seems an unlikely candidate for the rôle of maintenance vector.

The multiple isolations of Wesselsbron virus from A. lineatopennis at Pearson, near Salisbury, raises the question of what part,

if any, this virus had in the outbreaks of abortion in cattle attributed at the time entirely to RVF. It is not possible to answer this question satisfactorily, as the effect of this virus on pregnant cattle is apparently not yet known<sup>6</sup>. Our own limited observations on inoculated calves showed that at this age, at least, mortality does occur in cattle and that high levels of viraemia occur even in mild cases, from which it may be deduced that a bovine-mosquito-bovine transmission cycle almost certainly occurs. The high infection rate in A. lineatopennis is strongly suggestive that this species was transmitting the virus between cattle, at least on the farm where the isolations were made.

#### ACKNOWLEDGEMENT

I am very much indebted to the Director and Staff, Department of Veterinary Services, Salisbury, Rhodesia, for providing laboratory space and assistance in many other ways during my visit to Rhodesia.

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#### TRALS WITH RAFOXANIDE\* 5.

EFFICACY STUDIES AGAINST FASCIOLA HEPATICA, FASCIOLA GIGANTICA, PARAMPHISTOMUM MICROBOTHRIUM AND VARIOUS NEMATODES IN SHEEP

I. G. HORAK, A. J. SNIJDERS AND J. P. LOUW\*\*

#### SUMMARY

Anthelmintic efficacy studies involving 50 sheep artificially infested with *F. hepatica* and 100 sheep similarly infested with *F. gigantica* are described.

At a dosage rate of 5 mg/kg live mass, rafoxanide exhibited anthelmintic activity greater than 95 per cent against 75 day-old *F. gigantica* and 4 to 11 day-old (fourth stage) *H. contortus*.

At a dosage rate of 7,5 mg/kg live mass, an efficacy greater than 90 per cent was obtained against 50, 75 and 99 day-old *F. gigantica*, all stages of development of *H. contortus* and adult *G. pachyscelis* and greater than 85 per cent against 42 day-old *F. hepatica* and adult *C. ovina*.

At 15 mg/kg live mass, rafoxanide was 63,6 per cent effective against 25 day-old *F. gigantica* and 92,8 per cent effective against immature *P. microbothrium*.

#### INTRODUCTION

The recently discovered compound rafoxanide, 3,5 - diiodo - 3' - chloro - 4' - (p - chlorophenoxy) - salicylanilide, has exhibited activity against a wide range of parasites. Its anthelmintic acivity against Fasciola hepatica in sheep 1.5, Fasciola gigantica in cattle 6.7, Haemonchus contortus in sheep 8, Haemonchus placei in cattle 7 and larvacidal activity against Oestrus ovis in sheep 9, 10 have already been described.

Recent titration studies at this laboratory have indicated the dosage levels at which rafoxanide is effective against F. hepatica, F. gigantica, Paramphistomum microbothrium, H. contortus and Gaigeria pachyscelis 11. The present paper describes anthelmintic efficacy trials conducted in sheep against artificial infestations of the above-mentioned species and against Chabertia ovina.

#### GENERAL MATERIALS AND METHODS

Two experiments were carried out. Sheep were infested with the metacercariae of *F. hepatica* or of *F. gigantica*, and some with the metacercariae of *P. microbothrium*. Some sheep were also infested orally with the infective larvae of *H. contortus* and *C. ovina* or percutaneously with the larvae of *G. pachyscelis*.

For treatment, rafoxanide as a 2,5 per cent preformed suspension was administered intraruminally by means of a trocar and cannula.

At autopsy the larger bile ducts and gall bladders were opened and all visible flukes removed. The livers were cut into slices 2 to 5 mm thick and the slices incubated at 42°C in a normal saline solution. The flukes which had emigrated from the sliced liver were collected and stored for counting.

The liver flukes recovered from the indicator controls were counted microscopically and those from the other sheep macroscopically.

The *P. microbothrium* were counted in toto after recovery by methods already described <sup>12</sup>.

The nematodes were recovered either by processing the gastrointestinal tract in a waterbath  $^{13}$ .  $^{14}$  or by washing the ingesta over sieves with  $150 \mu m$  apertures.

#### EXPERIMENT 1

#### Materials and Methods

Fifty Merino ewe lambs, six to eight months old, were each infested with 150 metacercariae of *F. hepatica* and 210 larvae of *G. pachyscelis*, in addition 19 of them were infested daily for three days with 590 larvae of *H. contortus*.

The lambs were treated in groups, each consisting of 11 sheep. Rafoxanide was ad-

<sup>\*</sup>RANIDE: Reg. Trade Mark of MSD (PTY) LTD., Merck Sharp & Dohme International, Division of Merck & Co. Inc., Rahway, N.J., U.S.A.

<sup>\*\*</sup>MSD Research Centre, Hennops River, P.O. Box 7748, Johannesburg.

ministered at a dosage rate of 7,5 mg/kg live mass so that anthelmintic activity could be determined against:—

- (i) 28, 35, or 42-day old F. hepatica,
- (ii) 1 to 3 day-old (third stage) H. contortus, and
- (iii) 10, 17, or 24 day-old G. pachyscelis infestations.

An indicator control was slaughtered at the time of each treatment and 14 sheep were kept as untreated controls. All the sheep were slaughtered 50 to 65 days after the original infestation.

#### Results

The ranked burdens of F. hepatica are summarized in table 1.

The mean efficacy of rafoxanide dosed at 7,5 mg/kg live mass was 46,0, 58,7 and 88,9 per cent against 28, 35 and 42 day-old *F. hepatica* respectively.

Five treated sheep died within seven days of treatment and their worm burdens were excluded when calculating anthelmintic efficacy.

Two untreated sheep died during the course of the experiment and these harboured nine and 93 flukes. The worm burdens of these sheep are not included in the tables.

Table 1: EXPERIMENT 1: THE EFFICACY OF RAFOXANIDE AGAINST F. HEPATICA

Indicator		Sheep tre	ated at 7,5 m	ig/kg on:
Controls	Controls	Day 28	Day 35	Day 42
Day 28	19	6	.8	0
64	22 37	6 9 9	15 16	1 2
Day 35	37	1Ś	17	2 2
30	45	26	17	4
D 43	61	31*	19	10*
Day 42 14	73 76	33 49	23	13 14
	82	50	29 44	16
	83	68	57*	19*
	91	74	74	53*
	124			
Mean	63	34	26	7
% Eff.		46.0	58.7	88.9

<sup>\*</sup>Died within 7 days of treatment and excluded from

The worm burdens of the sheep treated when the *H. contortus* they harboured were one to three days old and of their untreated controls are summarized in table 2.

The mean burden in the untreated controls was 1 087 and that in the treated sheep 74 worms, giving a mean efficacy of 93,2 per cent.

Table 2: EXPERIMENTS I AND 2: THE EFFICACY OF RAFOXANIDE AGAINST H. CONTORTUS

I to 3 days old at treatment		4 to	ll days old at treatme	ent	11 to 22 days old at treatment		
Controls	Treated at 7,5 mg/kg	Controls	Treated at 5,0 mg/kg	Treated at 7,5 mg/kg	Controls	Treated at 7,5 mg/kg	
582 939 1 149 1 164 1 232 1 258 1 285	0 1 2 4 5 7 12 112 159 222 294	480 582 970 1 214 1 279 1 394 1 661	0 3 4 8 17 18 23 34 38 74* 177*	0 0 0 0 0 0 2 3 109* 209* 372*	498 784 885 1 043 1 048	0 0 1 2 2 2 7 8 10 12 23 486*	
1 087	74	1 083	16	1	<b>852</b>	6	
% Eff.	93,2	_	98,5	99,9		99,3	

<sup>\*</sup>Died within 6 days of treatment and excluded from the mean.

The *G. pachyscelis* burdens are ranked in table 3.

Table 3: EXPERIMENT 1: THE EFFICACY OF RAFOXANIDE AFAINST G. PACHYSCELIS

Numbers of G. pachysceis recovered from:											
Indicator	Controls	Sheep tre	ated at 7,5 m	ng/kg on:							
Controls		Day 10	Day 17	Day 24							
Day 10	0 3 3 6	0*	0*	0* 0*							
U	3	0	3	0*							
Day 17	6	1	4	Ī							
4th stage	17	2 3 5 7 8	10	2							
Day 24	19 22	5	17 21	2 3 4 4							
4th stage	30	7	22								
40	34		27	10							
	41 43 86	11	56 64	25 29							
Mean	25	5	22	10							
% Eff.		80,0		60,0							

<sup>\*</sup>Died within 7 days of treatment and excluded from mean.

The worm burdens in the controls varied considerably; consequently, not much reliance can be attached to the efficacy data. Against 10, 17, and 24 day-old *G. pachyscelis* rafoxa-

nide was 80,0 per cent effective, ineffective and 60,0 per cent effective respectively.

#### EXPERIMENT 2

#### Materials and Methods

One hundred Dorper lambs approximately eight months old were each infested with 90 metacercariae of *F. gigantica*. The lambs were then randomly allocated to various groups; the animals in these groups were infested with other helminths and treated at times such that anthelmintic activity could be determined against:—

- (i) 25, 50, 75, or 99 day-old F. gigantica,
- (ii) 4, 13, and 18 day-old (immature) P. microbothrium,
- (iii) 4 to 11 (fourth stage), or 11 to 22 dayold (adult) H. contortus,
- (iv) 15 (fourth stage), 30 (fifth stage), or 75 day-old (adult) G. pachyscelis, and
- (v) 1 to 8 (third stage), 9 to 24 (fourth stage), or 22 to 54 day-old (adult) C. ovina infestations.

Rafoxanide was administered to the sheep intraruminally with the exception of those sheep treated when the liver flukes were 99 days old, where treatment was per os. Dosage levels of 5,0 7,5 10,0 and 15,0 mg/kg live mass were employed. An indicator control was slaughtered at the time of treatment

Table 4: EXPERIMENT 2: THE EFFICACY OF RAFOXANIDE AGAINST FASCIOLA GIGANTICA

		Number	of F. gigantica	recovered from:							
1-41			Sheep treated on:								
Indicator Controls	Controls	Day 25 at 10,0 mg/kg	Day 25 at 15,0 mg/kg	Day 50 at 7,5 mg/kg	Day 75 at 5,0 mg/kg	Day 75 at 7,5 mg/kg	Day 99 at 7,5 mg/kg				
Day 25	11	9	0	0	0	0	0				
48	13	13	2	! o	0	0	0				
	15	15	2	0	0	' 0	. 0				
Day 50	20	16	3	1	0	0	0				
40	21	17	4	1	0	. 0	0				
	23	18	7	1	0	0	0				
Day 73	24	· 20	9	1	0	0	0				
41	24	21	9	2	. 0	0	0				
	25	26	9	i 3	0	0*	0				
	26		. 14	4	18*	19*	0				
	30	33	. 29	5	20*	25*	0				
	35	· C					4*				
Mean	22	20	8	2	0,0	0,0	0,0				
% Eff.		9,1	63,6	90,9	100,0	100,0	100,0				

<sup>\*</sup>Died within 6 days of treatment and excluded from the mean.

on Days 25 and 50; a sheep which had died on Day 73 was used as an indicator control for the sheep treated on Day 75.

The sheep were slaughtered 86 to 108 days after the original infestation with *F. gigantica*.

#### Results

The ranked burdens of *F. gigantica* are summarized in table 4.

At 5,0 mg/kg live mass, rafoxanide was 100 per cent effective against 75 day-old worms; at 7,5 mg/kg it was 90,9 100 and 100 per cent effective against 50, 75 and 99 day-old flukes respectively. At 10 mg/kg it had no effect and at 15 mg/kg it was 63,6 per cent effective against 25 day-old *F. gigantica*.

Eighteen untreated sheep died as a result of infestation, and four to 50 F. gigantica were recovered from these sheep with a mean of 25 worms. The worm burdens of these sheep are not included in the tables.

Six of the 34 sheep treated on either Day 75 or Day 99 died within six days of treatment and five of these sheep still harboured living flukes. The remainder of the sheep in these groups were slaughtered nine to 18 days after treatment and harboured no living flukes. Consequently, the flukes recovered from the sheep that had died were excluded when calculating the mean residual burdens.

The group of sheep which were infested with *F. gigantica* and treated with rafoxanide at 10 mg/kg live mass when the flukes were 25 days old, were slaughtered 86 and 87 days after initial infestation. The anthelmintic had little effect on the total worm burden. Considerable stunting of the flukes in the treated animals was observed; this in turn probably prevented death from infestation, as only one of these 11 treated sheep died before the slaughter date, whereas 15 untreated sheep with similar burdens had died by Day 87.

The numbers of *P. microbothrium* recovered from control and treated sheep are summarized in table 5.

A dosage of 15 mg/kg live mass resulted in a mean efficacy of 92,8 per cent against *P. microbothrium*, which were 4, 13 and 18 days old at the time of treatment.

Table 5: EXPERIMENT 2: THE EFFICACY OF RAFOXANIDE AGAINST P. MICROBOTHRIUM

	Nos. of paramphistomes reco	vered
	Treated 15 mg/kg at age 4, 13 and 18 days	Control
	0 0	266 376 399
	i	451
	3 7	477 532
	2 3 7 8 9	
	37 268	
Mean	30	417
Efficacy %	92,8	

In table 2 the ranked *H. contortus* burdens are summarized with those of Experiment 1.

The six sheep that died within six days of treatment all had residual worm burdens considerably greater than those of the remaining sheep in the group; these burdens were excluded for the purpose of determining efficacy.

At a dosage level of 5,0 mg/kg live mass rafoxanide was 98,5 per cent effective against 4 to 11 day-old *H. contortus*, and at 7,5 mg/kg live mass it was 99,9 and 99,3 per cent effective against 4 to 11 and 11 to 22 day-old worms respectively.

The numbers of *G. pachyscelis* recovered from the treated and control sheep are summarized in table 6.

Four of the six sheep which died within six days of treatment had the highest worm burdens in their respective groups and all six were excluded for the purpose of determining the mean worm burdens.

Rafoxanide had no or little effect on worms 15 or 30 days old. At 7,5 mg/kg live mass rafoxanide was 98,4 per cent effective against adult (75 day-old) *G. pachyscelis*; in fact ten of the eleven treated sheep were entirely cleared of hookworm infestation.

Table 6: EXPERIMENT 2: THE EFFICACY OF RAFOXANIDE AGAINST G. PACHYSCELIS

		Numbers	of G. pachyscelis reco	overed from		
	Treated		Tre	ated		Treated
Controls	Day 15 at 7,5 mg/kg	* Controls	Day 30 at 5,0 mg/kg	Day 30 at 7,5 mg/kg	Controls	Day 75 at 7,5 mg/kg
1 20 25 25 28 28 29 38 39 46 61	21 25 26 33 34 35 36 37 38 50 51	38 40 49 54 66 68 71	19 22 22 29 30* 34 53 62 80 86 93*	18* 19 33 38 40 47 50 50 54 76*	48 53 65 71 79	0 0 0 0 0 0 0 0 0 0 8 59*
31	35	55	45	41	63	1
% Eff.	0,0		18,2	25,5		98,4

<sup>\*</sup>Died within 6 days of treatment and excluded from the mean.

The ranked *C. ovina* burdens are presented in table 7.

Efficacy against one to eight, and nine to 24 day-old worms was poor, but increased to an efficacy of 88,9 per cent against 22 to 54 day-old worms.

#### DISCUSSION

At the commercially recommended dosage rate of 7,5 mg/kg live mass, rafoxanide exhibited efficacy greater than 85 per cent against 42 day-old F. hepatica, against 50, 75 and 99 day-old F. gigantica, against all stages of development of H. contortus, and against

Table 7: EXPERIMENT 2: THE EFFICACY OF RAFOXANIDE AGAINST C. OVINA

		Numb	ers of C. ovina recover	red from		
1 to 8 days	s at treatment	9	to 24 days at treatme	22 to 54 days at treatmen		
Control	Treated at 7,5 mg/kg	Control	Treated at 5,0 mg/kg	Treated at 7,5 mg/kg	Control	Treated at 7.5 mg/kg
51 78 265 279 291 325 359 363 363 391 431	86 129 159 178 199 202 204 220 236 263 292	158 186 213 230 273 295 296	9* 74 103 110* 160 217 227 290 313 314 316	20 53 60 65* 126 130 187* 194 263 270 333*	148 163 232 306 412	0 0 1 2 5 6 25 28 67 89 89
291	197	236	224	140	252	28
% Eff.	32,3	<u>-</u>	_	40,7	_	88,9

<sup>\*</sup>Died within 6 days of treatment and excluded from the mean.

adult G. pachyscelis and adult C. ovina. When this dosage rate was doubled, similar efficacy was obtained against immature P. microbothrium. To the above helminths can be added adult Haemonchus placei<sup>7</sup> and the larval stages of Oestrus ovis<sup>9, 10</sup> to complete the known spectrum of activity.

With the exception of *P. microbothrium*, the feeding habits of which are unknown, the other helminths are generally accepted as being haematophagous, particularly in the mature or adult stage, whilst *O. ovis* is considered to live off the secretions of the nasal passages and sinuses. As rafoxanide, being bound to plasma proteins, reaches and maintains high levels in the blood <sup>15</sup>, this possibly explains the susceptibility of these parasites to the compound at relatively low dosage levels.

When, however, the dosage is increased to 15 mg/kg, those stages of these parasites which are not truly haematophagous, are also affected. Examples are very young F. hepatica and F. gigantica, as well as immature P. microbothrium, which may, as is the case with Cotylophoron spp., be tissue feeders <sup>16</sup>. In this case blood containing the compound may be ingested accidently, and the higher concentration present be sufficient to be lethal.

The results of the present experiments indicate that rafoxanide may be slow acting, as can be seen from the number of living helminths of various species still present in treated animals six to seven days after treatment. It is, thus, important when conducting anthelmintic tests with this compound to allow a period of at least 10, and preferably 14, days between treatment and slaughter.

Infestations of *F. gigantica* proved to be highly pathogenic to sheep, the most common

cause of death being rupture of an hepatic vessel or the liver capsule with consequent fatal haemorrhage. The reason for the death of sheep soon after treatment may be two-fold: firstly, irreversible liver damage had already taken place before treatment, or, secondly, peritonitis, resultant upon intraruminal injection in comparatively young sheep, coupled with already severe liver damage, proved too great an assault on the body. For this reason it was decided in Experiment 2 not to treat the last group of 12 sheep intraruminally: only one death occurred after treatment which was adminstered per os.

The small numbers of F. gigantica recoverer from some of the untreated sheep that died as a result of infestation are not necessarily a true reflection of the total worm burden. Because of the severe damage to the liver, decomposition of this organ appears to be particularly rapid; the flukes are similarly affected and are not easily recovered subsequently and counted. In these sheep, flukes appeared to leave the liver for the peritoneal cavity prior to death; this, too, may account for the small numbers recovered.

The results obtained in Experiment 2 indicate that, although rafoxanide dosed at 10 mg/kg live mass had no effect on *F. gigantica* burdens 25 days old at treatment, it caused considerable stunting of the fluke. It is possible, therefore, that rafoxanide may prevent deaths from acute fascioliasis by stunting the growth of the liver flukes and hence reducing their pathogenicity.

#### ACKNOWLEDGEMENTS

Mesdames S. M. Raymond and I. Pienaar are thanked for their able technical assistance.

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#### INLIGTING INFORMATION

#### ONVRUGBAARHEID BY VERSE WEENS TEMPERATUUR-WISSELING

Indien verse vanaf koue na warm klimate, net voor die teelseisoen verskuiwe word, kan bevrugting vertraag word, wat 'n uitgawe vir die boer in terme van voerkoste verteenwoordig.

Staking van die bronssiklus en ander opmerklike fisiologiese veranderinge in beeste mag deur skielike blootstelling aan hoë temperature veroorsaak word. Met tyd en met voorsiening van die regte rantsoen, egter, kan beeste hulle by die hoër temperature aanpas.

Veekundiges van die VSDL se Landbounavorsingsdiens, dr. James Bond en dr. Robert E. McDowell het onlangs 'n studie van die gevolge van spanning a.g.v. hitte, voltooi. Beeste wat gedurende die somer of winter buite aangehou is, is aan 90°F temperature in kamers met lugreëling blootgestel. Kontrolebeeste is in konvensionele skure gehou.

Die verse is op omhulde, hoëkrag, laevesel rantsoen en water geplaas in proewe wat van 56 tot 200 dae geduur het. Die hoeveelheid wat hulle gevreet het is nie beperk nie. Weekliks is die velhaardiepte en gewig gemeet en daagliks is daar nagegaan om vas te stel of bevrugting plaasgevind het aldan nie.

Resultate het gewys dat die bronssiklus verwant is aan die mate van spanning—die verskil tussen Winter- en hoë Somertemperature.

Om sulke verwantskap te bewys is ses verse wat vir Somer gewend gemaak is, in die kamers gedurende Augustus (Somer) die volgende jaar, geplaas. Hulle is aan temperature van 100°F blootgestel. Aanvanklik het hul liggaamstemperature gestyg, toe effens gedaal, en daarna konstant gebly. Gedurende die sesweektydperk is die siklus van 5 uit die 6 verse belemmer.

Alhoewel die siklus van verse wat vir Winter gewend gemaak is vir verskillende tydperke gestaak is, is hulle bevrug en het hulle normale kalwers afgegooi toe die gereëlde siklus weer begin is. Ander veranderings wat in verse wat aan hitte-spanning gely het, waargeneem is, was o.a. 'n gemiddelde temperatuurverhoging van 1.8°F, en 'n verhoogde waterinname-13-kwart meer as in die geval van verse wat gewoonweg gehuisves is. Die mees besondere fisiese reaksie op hittespanning wat waargeneem is, was verharing en verminderde velhaardiepte in verse wat vir Winter gewend gemaak is. Velhaardiepte was skynbaar naverwant aan aanpassing deur verse aan aanhoudende hittespanning. Die wetenskaplikes het gevind dat indien verse baie water en hoëkrag, lae-vesel rantsoene ontvang het hulle makliker by die hoër temperature aangepas het. Boere hoort dus soiets te onthou indien hul beeste vanaf gebiede met lae temperature na dié met hoë temperature vervoer, aangesien kragrantsoene gedurende vertering die liggaamstemperatuur verhoog. Volgens dr. Bond is voorafgaande klimatiese kondisionering belangrik in die uitwerking van hitte op verse. Hy raai boere aan om die aankoop van beeste indien moontlik binne hul eie klimaatstreek te doen, veral gedurende die teelseisoen.

(Agricultural Research, U.S.D.A., Augustus 1972). (Oorgeneem uit: LANDBOURAPPORT van die Landbouraad van die Suid-Afrikaanse Ambassade, Washington, D.C., soos uitgereik deur die Departement van Landboutegniese Dienste, Pretoria, 1972).

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#### BAILLIÈRE \* TINDALL

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#### PROTECTIVE ACTION OF A LIQUID SOAP AGAINST **BILHARZIA**

L. J. van Rensburg\*

#### SUMMARY

It was found that the application of a soap containing hexachlorophene protected 31 of 32 Praomys (Mastomys) natalensis against subsequent infestation with Schistosoma mattheei. The protective effect of this preparation persisted when the tails of these animals were washed in running tap water (100 ml per minute) for periods of up to 40 minutes and then immediately exposed to S. mattheei cercariae.

#### INTRODUCTION

Kemp et al. 1 and Hunter et al. 2 showed that ointments containing hexachlorophene protected laboratory animals against infestation with schistosomes. Subsequently Armstrong & Fripp<sup>3</sup> applied a soap containing hexachlorophene (Schistopel\*\*) to the tails of multimammate rats, *Proamys* (Mastomys) natalensis, and found that the animals were resistant to infestation for 48 hours. In these trials, however, the tails of the rats were not rinsed for prolonged periods prior to exposure to schistosome cercariae. This paper describes an experiment which was conducted to determine the protective effect of Schistopel after rinsing the treated skin surface in running water for periods up to 40 minutes and then exposing it to Schistosoma mattheei cercariae immediately after rinsing.

#### MATERIALS AND METHODS

Infective material

A strain of S. mattheei obtained from Zululand and maintained in the laboratory in sheep and Bulinus (Physopsis) globosus and B. (P.) africanus 5, 6 was used.

Application of the soap and subsequent

rinsina

After anaesthetization with pentobarbital sodium the test soap was rubbed into the tails of 32 P. (M.) natalensis. The animals were then divided into 4 groups of 8 animals each. Immediately thereafter their tails were suspended in test tubes, into each of which tap water flowed through a rubber tube at the rate of  $\pm 100$  ml per minute. This rinsing continued for 3, 8, 20 and 40 minutes for groups A, B, C, and D respectively.

#### Infestation

The treatment and rinsing of the 4 experimental groups was staggered so that they could be exposed to infestation immediately after rinsing and simultaneously with the 8 untreated, unwashed rats in the control group. The animals were individually exposed to S. mattheei, as described by Purnell 7. Twenty five aliquots of 10 ml were removed after thorough mixing 6 from each of two cercarial suspensions, S1 and S2, each aliquot containing either 397 cercariae (S1) or 344 (S2). These were used to infest three, four, or five animals from each experimental group as well as the controls as indicated in the table. The remaining aliquots (10) were used to estimate the number of cercariae in the 2 suspensions. The rats were exposed to cercariae for 30 minutes and thereafter the number of cercariae remaining in each test tube was counted. The animals were placed into separate cages and the worms were recovered by perfusion 9 54 days after infestation. The fluid collected from the vena porta was examined with a binocular stereomicroscope.

#### RESULTS

The results are summarized in the table.

No worms were recovered from Group A (3 minutes' rinsing), Group C (20 minutes' rinsing) and Group D (40 minutes' rinsing). Six worms were recovered from one animal in Group B (8 minutes' rinsing), but the other animals in this group were negative. Parasites were recovered from all the untreated (control) animals: the number varied from 32 to 74 with a mean of 46,4 per rat. The mean percentage of cercariae remaining in the test tubes after infestation was 67,9%; 51,3%; 52,5%; 56,8% and 42,1% in groups A, B, C, D and the control group respectively.

\*\*South African Commercial House (Pty) Ltd.

<sup>\*</sup>Section of Helminthology, Veterinary Research Institute, Onderstepoort.

Table: EFFECT OF RINSING ON THE REPELLANT ACTION OF HEXACHLOROPHENE (SCHISTOPEL)
APPLICATION AGAINST EXPERIMENTAL S. MATTHEEI INFESTATION

	Animal	Duration of	Cerca	ariae	Warms
Group	No.	rinsing in water (minutes)	Number per dose	% that failed to penetrate	recovered
A	1	3	397 (SI)**	58,94	0
	2 3	3	397 (SI)	57,18	0
*	3	3	344 (S2)	72,97	
*	4 5 6	3 3	397 (S1)	57,43	
	5	3	344 (S2)	92,44	0
ļ	7	3 3	344 (S2)	65,99	0
į	8	3	344 (S2) 344 (S2)	66,57 71,80	0 0
MEAN		3		67,9	0
В	1	8	397 (S1)	45,84	0
	ż	8	397 (SI)	39,30	.0
	2 3	8	344 (S2)	40,41	6
	4	8 !	397 (SI)	42,32	ŏ
	4 5 <b>6</b>	8	344 (S2)	57,56	ō
	6	8	344 (S2)	60,17	0
*	7	8	344 (S2)	53,78	_
	8	8	344 (S2)	70,64	0
MEAN		8		51,3	0,86
c	1	20	397 (SI)	48,87	0
	2	20	39 <b>7</b> (S1)	51,64	0
	3 4	20	397 (51)	42,82	0
	4	20	397 (SI)	47,86	0
	5 <b>6</b>	20	344 (S2)	57,27	Q
_	<u>6</u>	20	344 (S2)	45,06	0
Ť	7 8	20 20	344 (S2) 344 (S2)	59,01 67,15	<u> </u>
MEAN		20		52,5	0
		40	207 / (1)		
·	l o	40	397 (S1) 397 (S1)	48,36 F9.19	Ŏ
	2 3	40	397 (SI)	58,19 64,48	Ŏ
	4	40	397 (SI)	55,16	0
*	4 5	40	344 (52)	43,90	
*	6	40	344 (S2) ·	61,63	_
*	7 8	40	344 (S2)	70,35	
*	8	40	344 (S2)	52,33	_
MEAN		40		56,8	0
 CONTROLS	1		397 (\$1)	36,27	52
*		-	397 (S1)	15.11	
	2 3		397 (SI)	32.75	
	4		344 (S2)	51,16	35
1	5	<del>-</del> i	344 (S2)	51,16 40,70	74 35 39
*	6	- i	344 (S2)	66,57	_
*	4 5 6 7 8	: =	344 (S2) 344 (S2)	48,55 45,35	32
— MEAN				42,1	46,4

<sup>\*</sup>These animals died under anaesthesia after infestation. \*\*S1 represents Suspension 1 and S2 Suspension 2.

#### DISCUSSION

Although the tails of the treated rats were washed in running water for periods up to 40 minutes, all but one of them were resistant to infestation with *S. mattheei*. This is in agreement with the findings of Armstrong & Fripp<sup>3</sup> who showed that mice of which the tails had not been rinsed for prolonged periods, were completely resistant to infestation with *S. mansoni* for 48 hours after treatment with this soap.

In all cases, including the rats that did not become infested, fewer cercariae remained in the test tubes after infestation than were present in the infective dose. It is not known whether these cercariae penetrated or merely adhered to the skin when the tails were removed from the suspensions. A relatively large percentage of cercarciae also failed to penetrate the untreated (control) group. This finding agrees with that of Van Wyk<sup>8</sup> who also found that large numbers of cercarciae fail to penetrate the skin of multimammate rats.

These results suggest that this product may be effective for short periods in protecting human beings against infestation with bilharzia and its action in higher primates should be investigated.

#### ACKNOWLEDGEMENTS

I am grateful to Dr. J. A. van Wyk and Dr. Anna Verster for support and advice during the investigation and with the manuscript and to Miss H. Pakendorff for technical assistance.

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#### CASE REPORT

#### **GEVALVERSLAG**

#### URETHROPLASTY IN A CASE OF TRAUMATIC RUPTURE OF THE ABDOMINAL URETHRA IN A CAT

P. ALCANTARA AND M. E. COSTA\*

#### SUMMARY

The successful performance of urethroplasty in a case of traumatic rupture of the abdominal portion of the urethra in a 6 months-old female Siamese is reported.

#### HISTORY

A female Siamese cat, which had been run over by a car on the previous day, was referred to our Department with a diagnosis of fracture of the right femur.

#### CLINICAL DATA

The clinical examination confirmed the fracture of the femur and disclosed a moderate paleness of the mucosae, slow vasomotor reactions, moderate dehydration, bradycardia, hypotension and a temperature of 37,8°C. The PCV was 30%. Abdominal pain and tension of the abdominal wall were present. In view of the signs of shock and of an acute post-traumatic abdomen, it was decided to undertake an exploratory laparotomy and the patient was immediately prepared for surgery.

Before and during the operation 100 ml of 5% dextrose and 100 ml of "Haemaccel" (Behringwerke) were administered by intravenous drip, and 600 000 IU of procaine penicillin plus 0,05 g of streptomycin sulphate were given by intramuscular injection.

#### OPERATIVE TECHNIQUE

After pre-medication with atropine suphate (0,25 ml of a 1 mg/kg solution), anaesthesia was induced by inhalation of a mixture of 4% "Fluothane" (I.C.I.) in oxygen through a face mask. When plane III of anaesthesia was attained, an endotracheal tube was inserted and anaesthesia maintained with the same anaesthetic by means of a semi-closed circuit, the concentration of the anaesthetic agent being controlled to maintain the patient in as light an anaesthetic plane as possible.

The abdominal cavity was opened by means of a midline incision extending from a point approximately 3 cm cranial to the umbilicus to a point approximately 2 cm cranial to the pubis.

The examination of the abdominal viscera disclosed a complete rupture of the urethra at a point about 1 cm caudal to the neck of the bladder (cervix vesicae). The bladder was moderately distended with urine which had not leaked into the peritoneal cavity in any appreciable amount, apparently because of the retraction of the severed mucosa and longitudinal smooth muscle fibres of the urethral stump and contraction of the circular smooth muscle fibres. The urethral lumen at the site of rupture was covered by a blood clot. The ligaments of the bladder were intact but the bladder had profuse subserous haemorrhages, probably of traumatic origin (Fig. 1). The distal portion of the urethra was not accessible even after the abdominal incision was extended to the pubis.

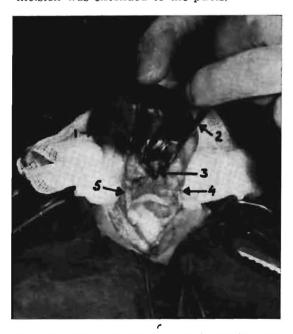


Fig. 1. The bladder, protected on its dorsal aspect by a gauze sponge, has been pulled from the abdominal cavity. Between the tips of the dissection forceps lies the neck of the bladder, and, slightly towards the bottom of the figure, the severed proximal end of the urethra can be seen, its lumen covered by a blood clot. Bladder (1); dissection forceps (2); proximal urethra, severed and covered by a blood clot (3); left (4) and right (5) lateral ligaments of the bladder.

<sup>\*</sup>Departamento de Cirurgia, Faculdade de Veterinária, Universidade de Lourenço Marques, Moçambique.

A sterile rubber catheter was introduced aseptically through the external urethral orifice by an assistant. By these means the distal portion of the abdominal urethra was pushed into the pelvic cavity and the tip of the catheter, showing through the lacerated orifice, was manually guided into the proximal portion of the urethra and the bladder. The bladder could thus be emptied.

The urethra was sutured with the catheter in situ by means of three simple interrupted sutures of no. 3/0 non-capillary braided silk, the knots being tied over the serosa (Figs. 2 and 3).



Fig. 2. Three simple interrupted sutures have been passed, connecting the ends of the severed urethra. A cathter, previously passed through the distal portion of the urethra and into the bladder, can be seen. Bladder (1): neck of the bladder (2): catheter (3): distal portion of the urethra (4).

After the abdominal cavity had been flushed with warm sterile isotonic saline to which penicillin and streptomycin had been added, the abdominal incision was closed in the usual manner.

The catheter was left in place and secured to the vulva with a suture.

During the operation two cardiac arrests occurred. Immediate cardiac massage through the diaphragm and hyperventilation of the lungs with pure oxygen resuscitated the patient in both cases.

#### POST-OPTRATIVE THERAPY

A daily therapy consisting of 200 000 IU of procaine penicillin, 0,025 g of streptomycin sulphate, 100 ml of 5% dextrose in isotonic

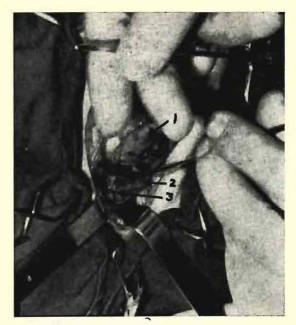


Fig. 3. The sutures have been tied, pulling the severed ends of the urethra together. Bladder (1); flat end of a probe inserted urethra (2); approximated ends of severed urethra (3).

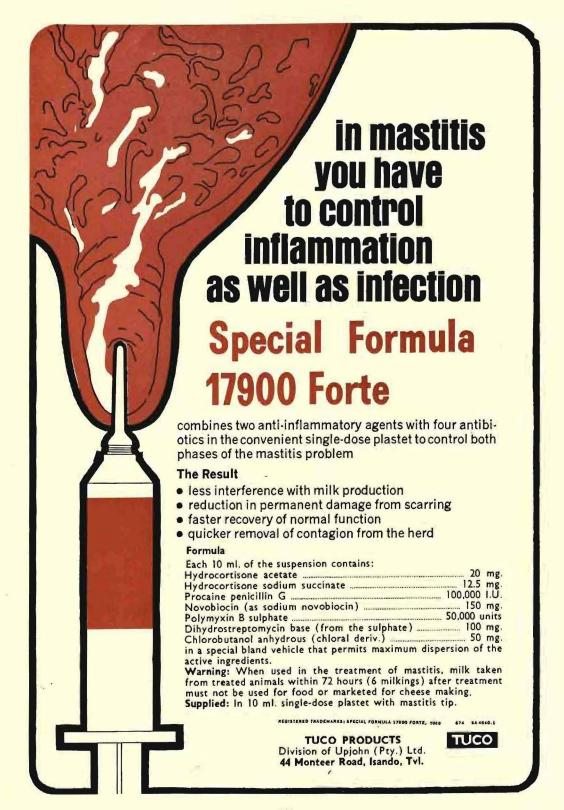
saline and a polyvitamin compound was established. The patient recovered steadily and on the fourth day was considered to be in a condition to withstand a second operation.

Under general anaesthesia with Fluothane (I.C.I.), a Steinman pin was used to stabilize the fracture of the femur, after the fracture site had been exposed by a lateral approach. This operation was easily performed and no incidents occurred.

The general condition of the patient worsened a little during the three days after the second operation, but improved afterwards.

A cystitis developed and was diagnosed seven days postoperatively. Treatment with nitrofurantoin ("Furadantina", Laboratório Normal) and nalidixic acid ("Wintomilon", Winthrop) proved ineffective. The infection, caused by a Streptococcus sp. and a Corynebacterium sp., was finally controlled with ampicillin ("Penampla", Laboratório Sanitas).

On the fourteenth day of the postoperative period the catheter was removed by the animal. As it was considered that healing of the urethra should have occurred by then, no attempt was made to replace it. The healing of the fracture was delayed, but on the 53rd post-operative day the Steinman pin was removed. Five months after the operation the animal is in perfect condition.



#### In Memoriam



#### MARKUS ZSCHOKKE 1893—1972

'n Baanbreker op die gebied van veeartsenykunde en 'n alombekende persoon in Suidwes, is in Swakopmund oorlede waar hy in 'n hospitaal verpleeg was.

Dr. Markus Zschokke, gebore in 1983 te Basel, Switserland, het in 1919 na Suid-Afrika gekom nadat hy sy universitêre opleiding voltooi het te Zürich, om hom as bakterioloog te Onderstepoort te vestig.

In 1929 word hy na Suidwes-Afrika verplaas as staatsveearts in welke hoedanigheid hy die gebied vir 38 jaar lank in verskeie distrikte gedien het en 'n bekende persoonlikheid oor die hele gebied geword het.

In 1942 is hy vir die laaste keer verplaas na Grootfontein waar hy in 1965 afgetree het op die hoë ouderdom van 72 jaar. Terwyl hy in Grootfontein gestasioneer was, het hy ook spesiaal opdrag gehad om toesig en kontrole te hou oor die mees noordelike bantoegebiede, nl. Okavango, Owambo en Kaokoveld. Hierdie taak volvoer hy vir 18 jaar lank.

As kultuurhistories-bewuste persoon het hy o.a. 'n leidende aandeel gehad om Fort Namatoni by die oostelike ingang van die huidige Etosha-wildreservaat as Nasionale Monument verklaar te kry.

Die oorledene is onder blyke van groot belangstelling op Nuwejaarsdag begrawe op sy klein plasie "Hisuma-Hill" naby Grootfontein. Hy laat sy eggenote en twee getroude kinders na ('n seun wat in Port Elizabeth woon en 'n dogter te Walvisbaai), aan wie ons ons opregte meegevoel betuig.

(Oorgeneem, met sekere byvoegings, uit ,Die Suid-wester').

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#### In Memoriam

#### WALTER FREI

Prof. Dr. Med. Vet., Dr. h. c. Walter Frei, after a brief illness, died in Zollikon, Zürich, on September 29, 1972, in his ninetieth year, according to information kindly sent by one of his pupils, Dr. Hans E. Suter, (who, we regret to learn, himself passed away on November 12) to the Dean of the Faculty of Veterinary Science, Onderstepoort. With his decease, the last living link with the pioneering days of veterinary research in South Africa—the 'Daspoort era'—has fallen away.

Professor Frei remained active in research until the end. His attachment to Onderste-

poort always remained strong and, were it not for serious indisposition of his wife, he would have visited South Africa in 1963. A token of his esteem is to be found in the form of his book "Allgemeine Pathophysiologie der Infektionskrankheiten" published in 1966 and donated by him to the Faculty Library, housed in the Students' Residence, as part of the University's Merensky Library.

We pay homage to a great figure in the history of veterinary science in South Africa and extend our deepest sympathy to his wife, Lina, son Ernst and daughter Gerda.

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Health and Meat Inspection; Section B of lectures and practical work in the Zoonoses, Food Hygiene and Meat Inspection and Section C of lectures and practical Work in Pathology, Mycology, Microbiology and Immunology, Poultry Diseases, Parasitology and Clinical Chemistry, with particular emphasis on laboratory diagnostic techniques.

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Written and Oral and Practical examinations are held in June with a resit in September/ October.

Candidates desirous of taking the course must have a Veterinary qualification registrable with the Royal College of Veterinary Surgeons or such other veterinary qualification as may be recognised for the purpose by the University Court.

Further particulars and forms of application may be obtained from the Dean, Royal (Dick) School of Veterinary Studies, Summerhall, Edinburg, EH9 1QH., to whom application should be sent not later than 30th June, 1973.

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#### PURPOSES OF WORLD VETERINARY CONGRESSES

President, World Veterinary Association
[Editorial Note: This article was written in preparation for the 1975
World Veterinary Congress.]

The purposes for which the World Veterinary Association was created are set out in general terms in Article 1 of its Constitution. From that statement one can infer that the purposes of World Veterinary Congresses are:

 To promote a sense of unity in the profession by bringing together veterinarians of all nationalities and all specializations

Although today veterinarians in differnt countries are brought closer together by modern communications, some sections of the profession are drifting apart from another point of view, namely, specialization, and this could lead to fragmentation of the profession and consequent erosion of its standing. Thus, there is as much need today as ever to foster the concept of one veterinary profession embracing all specializations as well as all nationalities. National and regional congresses play an important rôle in advancing the profession in some parts of the world, but there is need for world congresses to symbolize the concept of a unified profession extending throughout the world. Similarly, specialist associations perform a valuable function in advancing their subject, but they should also be outward-looking, and World Veterinary Congresses provide them with an opportunity to communicate with other sections of the profession. Chauvinism, whether in the form national isolationism or specialist separatism, is a short-sighted policy that is inappropriate for a scientifically based profession.\*

To increase the prestige and influence of the veterinary profession and make its value to the community more widely appreciated

Professional ethics do not allow individual veterinarians to advertise the services they have to offer. Governments and the public are not aware of the full scope and potentialities of the profession; this is especially so now when the profession is expanding into many new fields. Publicity associated with World Veterinary Congresses and reporting of the scientific programme in the world press provides a unique and dignified way to make widely known the services which the profession has to offer to the community today. Congresses also provide an opportunity for the veterinary profession to communicate with government officials and leading members of sister professions by inviting them to take part in the proceedings.

 To advance all branches of veterinary science by the pooling and exchange of information and ideas

Before the 18th World Veterinary Congress in Paris in 1967 the scientific programme consisted mainly of papers reporting research. It became increasingly obvious that most of these papers were so specialized and highly technical that they were of interest to very few congressists. It was felt by many that such papers are more appropriate for meetings of specialists in particular disciplines and that the special and unique function of professional congresses is to provide an opportunity for colleagues with similar interests to exchange views on topics of current importance, to gain an understanding of each other's problems and to become personally acquainted. Therefore, at the Congresses in Paris and Mexico a new way of arranging the scientific program-

The Editor of this Journal in this respect announces with concern and regret receipt of a letter from the Central Agricultural Library, East Berlin, containing a scurrilous attack on the South African Government and issuing an ultimatum to our Association to oppose the Government or cease exchange. Presuming that the letter had been written under extreme political pressure and not wishing to embarrass further our colleagues using the aforesaid Library, your Editor had no option but to cease exchange.

me was adopted. Now all Members and Associate Members are invited to suggest topics which they wish to hear discussed. After all the proposals have been received and a selection made, the Members and Associate Members are asked to nominate speakers for the chosen topics. To select the topics and speakers, a Scienti-Programme Advisory Committee (S.P.A.C.) has been set up, consisting of the Bureau, 3 Members of the Permanent Committee and one representative of each of the Associate Members. The task of the S.P.A.C. is not only to find the best speakers available, but to choose speakers bearing in mind the need for a fair representation of all Member countries; this could not be achieved if Associate Members acted independently in selecting speakers, especially as many of the Associate Members are not represented in some of the Member countries. The selected speakers are asked to present a concise review of current knowledge on some aspect of the topic, to include their own views but not to present a detailed report of their own research. Four speakers are chosen for each topic. Short communications reporting briefly the results of recent research or experience relevant to the particular topic may be offered after the main presentation. Then follows a discussion in which any congressist may take part. This programme offers wide opportunities for communication across language barriers by individual veterinarians, many of whom would not otherwise experience this type of intellectual stimulation.

The scientific programme is not the only opportunity for congressists to discuss matters of current scientific and professional interest. The social pro-

gramme provides opportunities for colleagues from all parts of the world to meet under congenial circumstances and hold informal discussion. Also, it is valuable for colleagues to get to know each other on a personal basis.

Finally, it should be mentioned that the commercial exhibits enable congressists to see an international display of the drugs, biologicals and instruments, which form an essential part of veterinary activities, and to meet representatives of the manufacturers.

#### 4. To improve veterinary education

Associated with the slightly different rôles which the profession is called on to play in different countries, and with the new fields into which the profession is moving, there are important changes taking place in veterinary education and these vary in different countries. The World Veterinary Congresses provide a unique opportunity for veterinary educationalists from all countries to meet and exchange informaton, experience and views. Here again it is essential for the profession to maintain some degree of uniformity as well as to adapt to a variety of needs.

In conclusion, World Veterinary Congresses are essential for attaining the aims of the World Veterinary Association, and they provide a unique and valuable service to the profession as a whole. All major professions require, and have, some such international forum. World Veterinary Congresses constitute an impressive expression of universal collaboration by veterinarians, and their functions differ somewhat from national and specialist congresses. Their value should be appreciated by all whose vision extends beyond their own parochial sphere.

#### **TYDSKRIFNUUS**

JOURNAL NEWS

#### ÉCONOMIE ET MÉDECINE ANIMALES

Hierdie tydskrif, uitgegee deur die "Expansion Scientifique", Rue St. Benoît 15, Paris (6°), Frankryk, verbind suiwere wetenskap en praktyk in die veeartsenykunde oor 'n wye gebied. Die referate oor omgewingsbesoedeling en toksiese residue gelewer tydens die sittings insake menslike en dierlike patologie, gehou te Parys op 4 en 5 Oktober 1972, sal ook in dié tydskrif opgeneem word. Abonnement beloop 42F per jaar, betaalbaar per bankwissel aan die uitgewers.

#### EFFECT OF REV 1 VACCINATION

Sir.

We wish to refer to our article "The incidence of *Brucella* ovis infection in South African rams: a serological survey", which appeared in the March issue of this Journal (Jl S. Afr. Vet Ass. 43 (1):83).

It would appear from the figures produced that Rev 1 vaccination had little value in reducing the incidence of *B. ovis* infection (5,0 per cent in the non-vaccinated as against 3,6 per cent in the vaccinated animals) which is contrary to our experience in the field. We also expressed our doubts as to the reliability of the histories on vaccination procedures obtained in such an extensive survey.

In order to evaluate the position more accurately, the data obtained from stud farms in the Middelburg and Graaff Reinet districts during the above survey were subjected to a careful analysis. These districts were chosen as we are well acquainted with the majority of stud farmers and the operational procedures practised on their properties. All the farms involved in our previous survey were listed and divided—according to our knowledge and experience—into three categories, viz. (A) where Rev 1 vaccination is

ed farms are summarized in the appended table.

From these figures it can be seen that 7,1 percent of animals from group A farms had titres of 1/8 and higher (titres regarded as postive) against B. abortus and 0,3 per cent had the same titres against B. ovis. Comparable figures were 14,5 and 18,1 per cent for rams from group B farms and 0,0 and 3,4 per cent for animals from group C farms. If titres of 1/4 and higher (suspicious and positive titres) against B. abortus and B. ovis respectively, are considered, the percentages of animals involved for each group of farms are: group A farms 15,0 and 1,3; group B farms 21,5 and 25,9; and group C farms 0,5 and 6.3.

It is obvious that the incidence of positive *B. abortus* titres was high (7,1 and 14,5 per cent for A and B groups of farms respectively) on farms where Rev 1 vaccination is practised, while no positive titres occurred on farms (group C) where vaccination is not done. It seems justified, therefore, to assume that the *B. abortus* titres were induced by vaccination. The 0,5 suspicious reactors on group C farms could possibly be attributed to vaccinated introductions or to non-specific

Table: COMPARISON OF CF TITRES AGAINST B. ABORTUS AND B. OVIS OF SERA FROM RAMS ACCORDING TO REV 1 VACCINATION PRACTICE

Conserve No		No. of	B. abortus titres					B. ovis titres						
Category	Noof farms				1/16	1 8	1	1/2	Neg	1/16	1	14	1	Neg
A. Vaccinations: Meticulous	22	719	31	20	57	155	456	1	1	7	91	619		
B. Vaccinations: Unreliable	9	304	34	10	16	62	182	35	20	24	67	158		
C. Vaccinations: Non-existent	16	414	0	0	2	26	386	13	1	12	62	326		

meticulously carried out, (B) where vaccination procedures are unreliable, and (C) where vaccination is not practised. The second group (group B) includes those farms where vaccinations are incorrectly rather than irregularly done. All farms which could not be placed with certainty, were omitted. The results of the CF test on sera from the select-

reactions. The higher incidence of positive (14,5 as against 7,1 per cent) or positive and suspicious (21,5 as against 15,0 per cent) B. abortus titres on farms where vaccination is not properly carried out when compared to farms where it is meticulously done, seems to be convincing evidence of the faulty procedures followed on these properties. Recent

work (in press) has shown that following Rev 1 vaccination CF titres rise sharply but fall to negative levels within 6 months, and that Rev 1 vaccination induces higher titres to B. abortus antigen than to B. ovis antigen. As yearling and older rams were involved in our survey, the higher incidence of B. abortus titres in group B indicates that on these farms sheep were vaccinated at an older age than on the farms in group A. The official recommedation is that rams should be vaccinated at weaning time, at a time when they have still had minimal contact with naturally infected rams. This is an extremely important point in the control of the disease by vaccination under practical farming conditions.

The incidence of positive B. ovis titres is highest (18,1 per cent) in rams from farms where unreliable vaccination is practised, followed by farms where no vaccination is carried out (3,4 per sent), and lowest (0,3 per cent) on farms where vaccination is properly done. The incidence of positive or positive and suspicious B. ovis titres on properties in group A (0,3 and 1,3 per cent) was therefore drastically reduced, compared with farms in group C (3,4 and 6,4 per cent). If it is considered that the farms in group A probably had severe problems due to B. ovis infection and therefore resorted to Rev 1 vaccination, while the farmers in group C probably were still unaware of its occurrence

on their properties, the beneficial results of Rev 1 vaccination are most striking.

The severe incidence of B. ovis infection on farms in Group B is, of course, representative of the situation on properties where the problem is rife, and also affords the reason why preventative vaccination is employed. It must be admitted, however, that some of the positive B. ovis titres may have been caused by recent Rev 1 vaccination.

The situation on these farms is in our opinion more closely comparable to that which prevailed on properties in group A, previous to their introduction of Rev 1 vaccination. The difference in incidence of B. ovis titres between these two groups of farms is even more dramatic and conclusive evidence of the excellent results that can be obtained with Rev 1 vaccination, on condition that it is carried out meticulously. Proper use of the vaccine includes correct handling and storage and strict compliance with the instructions issued with the vaccine, particularly with regard to the age of vaccination.

E. M. van Tonder\*
R. W. Worthington\*\*
Maria S. G. Mülders\*\*

#### BOOK REVIEW

BOEKRESENSIE

#### DIE GEFÄSSVERSORGUNG DER WIRBELSÄULE UND DES RÜCKENMARKES VOM HAUSSCHWEIN

(Sus scrofa F. domestica L., 1758)

#### HABILITATIONSSCHRIFT

(THE VASCULAR SUPPLY OF THE VERTEBRAL COLUMN AND SPINAL CORD OF THE DOMESTIC PIG)

HORST WISSDORF

Paul Parey; Berlin & Hamburg, 1970; 104 pages; 68 figures; paper back. Price DM 30.-.

This work was inspired by a research project at the Tierārztliche Hochschule, Hannover, on the subject of rheumatoid diseases in animals. The changes noted in the vertebral column and spinal cord of the pig seemed to justify a thorough investigation of the blood supply of these structures.

Part I (66 pages) covers the macroscopically demonstrable arteries and veins of the vertebral column. This includes a rather superficial description of the distribution of the nutrient foramina of the vertebrae. A bibliography of more than 150 references is given.

Regional Veterinary Investigation Centre Middelburg, Cape Province.

<sup>\*\*</sup>Veterinary Research Institute, Onderstepoort.

Every relevant artery and vein is desscribed from its origin to its extraosseus ramifications at the vertebrae and intervertebral discs. The intraosseus circulation is barely touched. Forty-seven photographs and good drawings illustrate this section.

Part II (24 pages) deals with the arteries and veins of the spinal cord demonstrable by macroscopic dissection and by light microscopy. It is illustrated with 21 photographs and drawings and one table. The bibliography includes 50 references. In conclusion a short summary of parts I and II is given in English, French and Italian.

The author must be congratulated on presenting a scholarly account of this in-

creasingly vital and long-neglected field. It is a valuable book for the specialist—whether medical or veterinarian. The author has taken great pains acknowledging every known worker in this field, covering comparative, veterinary and human anatomy and embryology, so much so that these references often seem irrelevant and detract from the flow of the text.

Considering the magnitude of the subject, the readability of this slender volume would have been enhanced considerably if more space were allowed for both text and figures.

M. M. S. S.

#### **BOOK REVIEW**

**BOEKRESENSIE** 

#### VETERINÄRHYGIENE

E. ROOTS, H. HAUPT, H. HARTWIGK AND D. STRAUCH

Paul Parey; Berlin and Hamburg, 2nd Edition, 1972; pp xii & 345, Figs. 86 & Tabs. 30.

Price DM 68.

The classification of veterinary hygiene, preventive medicine, managemental hygiene and animal hygiene proves to be difficult, but the old title "Veterinarhygiene" was kept for the second edition of Roots and Haupt's well-known book which appeared in 1955.

The different parts of the book cover almost all subjects dealt with in the Animal Management Course of the curriculum as given in the Faculty of Veterinary Science of the University of Pretoria at Onderstepoort. Here it could be suggested to change the name of the mentioned course to Veterinary Hygiene.

In the first part, the health problems through harmful feeds are dealt with very well: a complete list is included of the effects of feed deficiencies and excesses of nutrients. In the second part, on the provision of water, limiting values of detrimental elements in drinking water are given and a section on disinfection of drinking water is included. In the next sections, complete information is provided on buildings and building materials, stable systems and management and climate. Even if some of the information is not applicable under South African conditions, the trend to greater intensification certainly will continue and the sections on stables for pigs, horses, dairy cattle and poultry provide valuable information.

The section on management of artificial

pastures is of little value to the South African reader but the sections on handling facilities, animal behaviour and vices are of great importance. The parts on general hygiene and stud management and disease prevention, including disinfection, are excellent and should be studied by every student in Veterinary Science and Animal Husbandry (if he is able to read German).

The sections on the removal of animal waste products and cadavers provide some new information, aspects which become more and more important in the practice of the future Veterinarian and Animal Scientist, since almost everybody nowadays is concerned with prevention of pollution.

The section on transport of animals is valuable and the aspects of animal protection in intensive management systems are extremely well presented. For the local reader the section on animal hygiene in the tropics is useless, but the parts on the germfree and gnotobiotic animal and the protection from radiation provide the latest information on these modern aspects.

This book is one of the best of its kind and should become a standard reference book for students in Veterinary Science, Animal Husbandry, for the Animal Breeder and the numerous persons dealing in many different ways with animal products.

D. R. O.

#### SUDDEN DEATHS FROM CLOSTRIDIUM SEPTICUM INFECTION

#### IN THE GOBABIS DISTRICT

Sir.

From time to time sudden deaths have been encountered in mature cattle, sheep and goats in the district of Gobabis, South West Africa. The peracute course of the disease and the frequent appearance of a sanguineous discharge from the nostrils and anus resembled the signs of anthrax. Nevertheless, the animals had been vaccinated against this disease.

In bloodsmears, bacilli in strings were found. At certain stages of sporulation the organisms bore a vague resemblance to B. anthracis. Generalized and advanced decomposition, despite a brief interval between death and autopsy, was a striking feature. The characteristic smell of clostridial decomposition was obvious. By comparison, the liver seemed to have decomposed more rapidly than the other organs. In most cases the spleen was enlarged, once more suggesting anthrax.

Goats were affected with a circumscribed pneumonia and a gelatious infiltration of the area around the pericardium, a severe pericarditis and myocarditis. Tumor lienis was moderate.

As this area is densely bushed, it was frequently impossible to get to the carcass early enough, but in one outbreak, during which 34 head of cattle had died, specimens could be collected for bacteriological examination within one hour of death. From these, Dr. J. Bergmann of the Windhoek Regional Veterinary Laboratory was able to grow Clostridium septicum in pure culture.

During the past winter only seven deaths from suspected Clostridium septicum infec-

tion were encountered in goats. A nanny goat that had aborted the previous evening was found dead next morning. The autopsy was performed at 10 a.m. It had been extremely cold that night and it was possible to isolate *Clostridium septicum*, diplococci and haemolytic *E.* coli.

On account of the extremely rapid decomposition, specimens for bacterial isolation should be collected within one hour after death. Even so, Clostridium septicum is so ubiquitous and such a rapidly growing organism, that it may easily mask other organisms. Smears from fresh cases should really be examined by applying fluorescent antibody techniques. Under the circumstances this could not be done, but the evidence was considered sufficiently suggestive to warrant the use of a trial batch of Clostridium septicum vaccine (±2000 doses) obtained from the Veterinary Research Institute, Onderstepoort. The results were dramatic and were considered as final evidence that Clostridium septicum was responsible. despite experience that this infection tends to be self-limiting under field conditions.

A polyvalent clostridial vaccine, incorporating Clostridium septicum, Clostridium chauvei and possibly Clostridium oedematiens would be most useful.

T. C. Wessels

State Veterinarian, Gobabis, S.W.A.

chauvei and Clostridium septicum will shortly be issued by the Veterinary Research Institute, Onderstepoort.—Ed.)

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#### **PERSONALIA**

#### Dr. W. BÜTTIKER

An authority on the 'eye-frequenting moths', which feed around the eyes of cattle, sheep, game and even humans, and which are the suspected vectors of ophthalmia in mammalian animals, recently visited South Africa.

He is Dr. W. Büttiker from Ciba-Geigy, Switzerland—leading manufacturers of specialized chemicals—who had come to investigate certain aspects of citrus entomology in South Africa. Dr. Büttiker previously lived in South Africa for four years—from 1953 to 1956—doing research in Nelspruit on tropical and sub-tropical crops, and in Johannesburg on plant protection and veterinary and medical entomology.

Apart from being a menace to many species of domestic animals in Africa and Asia, the 'eye-frequenting moths', which are related to the commonly known fruit-sucking moth in this country, are also a grave threat to sheep in the Karoo, says Dr. Büttiker. Their diet includes the lachrymation and pus of the eyes of animals. As a result of intensive studies a group of skin-piercing, blood-sucking moths have been discovered in Asia by H. Bänziger, a former student of Dr.

Büttiker's.

Following on research previously done by Dr. R. du Toit and Dr. S. Stampa at Onderstepoort, Dr. Büttiker carried out extensive investigations and 'already has considerable proof that these moths are indeed the vectors of ophthalmia'. In an attempt to prove the last steps of this transmission and to isolate the causative agent of the eye disease, Dr. Büttiker will be going on another entomological expedition to the Ivory Coast in October this year.

He anticipates visting South Africa again in 1974 in order to conduct an entomological expedition in various parts of the country. His wife, Sonya, will be accompanying him as technical assistant, as she has done on all previous expeditions.

Every second year since 1963 Dr. Büttiker has gone on entomological expeditions to Asia and Africa in connection with parasitology, including the 'eye-frequenting moth'; and as a member of the World Health Organisation's Malaria Advisory Team he has done research on the eradication of malaria in Afghanistan, Ceylon, Burma and Cambodia.

INLIGTING INFORMATION

#### SOUTBOSSTUDIES MAG NUWE WEIVELDE TOT GEVOLG HÊ

'n Spesiale studie, gemik op die ontwikkeling van nuwe weivelde in die dorre en halfdorre gebiede van die wêreld, word tans deur die Fakulteit Biologie van die Texas Universiteit, onder leiding van dr. J. R. Goodin, onderneem.

Vir die doel van hierdie soutbosfisiologiestudie, word die genus Atriplex (soutbos) gebruik, en reeds is verskeie betekenisvolle eienskappe waargeneem. Deur middel van aftastende elektronemikrografie van poreuse blaartjies op Atriplex-blare, is 'n menigte blasies waargeneem, wat of besig was om te ontwikkel, óf reeds gebars het. Die selle ontspruit uit die opperhuid van 'n stam-sel, baie van die kenmerke waarvan gelyksoortig is aan dié van 'n gewone kliersel. Beide die stam-sel en die blasie bevat kloroplaste, en sitoplasmastroming in die blasie is deur fase-mikroskopie waargeneem.

Die mening is dat 'n sterk "soutpomp"-aksie die akkumulasie van beide katione en anione, van 'n betreklik geringe bron, veroorsaak en dat sodra 'n sekere versadigingspunt bereik word, die selle bars en soutkristal-selwandafval oor die blaaroppervlakte versprei word. Daar is meer sout in die poreuse haartjies as in die blaarsap te vinde.

As gevolg van die soutlaag, behoort die meganisme evapotranspirasie vanaf die blaaroppervlakte te verminder, wat sy droogteweerstandmeganismes natuurlik sou verhoog.
Die proses is aaneenlopend en, namate die soutdekking op die blare toeneem, word die blare minder smaaklik vir diere, tot op die stadium wanneer dit reën en die sout afgewas word en in die grond beland.
Daar neem dit weer 'n kringloop en word dit deur die plant, tot op die blaaroppervlakte, gevoer.

Proewe is reeds uitgevoer met verskeie Atriplex-soorte, wat as weidinggewasse onder gewone agronomiese omstandighede gekweek is. Aangesien die opbrengste hiervan dikwels so hoog was as dié van lusern, kan die soutbos, volgens dr. Goodin, in dorre gebiede aangepas word waar besproeiing buite die kwessie is a.g.v. 'n tekort aan water, of waar watergehalte so sleg is dat tradisionele voergewasse nie gekweek kan word nie.

Die woestynstruike het 'n hoë proteïeninhoud en kan in dorre gebiede suksesvol aangepas word aangesien hul wortelstelsels breed is en dus die vermoë het om oor 'n groter grondruimte te absorbeer. Hulle is ook besonder bestand teen droogtes, asook hoë temperature, sterk lig en hoë soutinhoud, en alhoewel hierdie plante nie as vervangers van normale weidinggewasse beskou kan word nie mag hulle wel van belang wees om die landbou na gebiede wat voorheen onbruikbaar was, uit te brei.

Voortspruitend uit hierdie navorsing het reeds twee referate verskyn, nl.: Oxalate Metabolism in Atriplex; en An Ultrastructural Study of the Vesiculated Hairs in Atriplex.

Die "International Center for Arid and Semi-Arid Land Studies (ICASALS)" het reeds finansieel tot hierdie navorsing bygedra. (ICASALS Newsletter, Vol. 5. nr. 2).

(Oorgeneem uit: LANDBOURAPPORT van die Landbouraad van die Suid-Afrikaanse Ambassade, Washington, D.C., soos uitgereik deur die Departement van Landboutegniese Dienste, Pretoria, 1972).

INFORMATION INLIGTING

#### NEW TECHNIQUE TO BRING COWS INTO PRODUCTION WITHOUT PREGNANCY

Ohio scientists who developed a technique using a combination of natural hormones to bring a dairy heifer or cow into production without pregnancy have established levels of hormones that have proven successful in an average of 60 percent of the cows tested.

Dr. K. L. Smith, research dairy scientist at the Ohio Agricultural Research and Development Center, Wooster, said that he and co-worker Dr. F. L. Schanbacher have refined the techniques which they first revealed in late 1971. The level and ratio of estrogen and progesterone have been established which will initiate substantial milk yields in non-pregnant, non-lactating cows and heifers.

In normal practice, dairy cows do not begin to produce milk until they calve. A major problem encountered by dairymen is that 5 to 10 per cent of all dairy cows have some sort of reproductive malfunction which prevents conception when they are mated. Ultimately, this means the slaughterhouse for the cows even though they may be potentially high milk producers.

Scientists have tested a number of methods for initiating lactation, with only

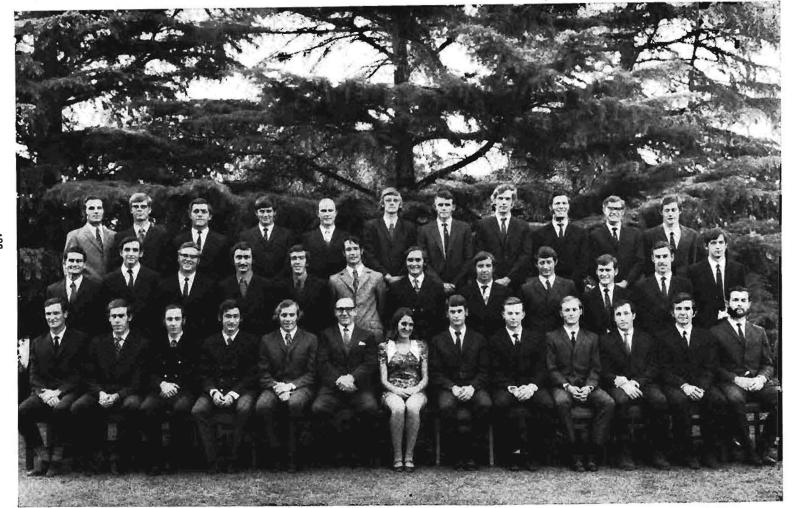
limited success. The Ohio technique appears to be the most reliable yet developed. It has the added advantage of requiring hormone administration for only 7 days as opposed to the 60 to 120-day period of time required by older hormone injection methods. The Ohio scientists have also achieved higher levels of milk production.

Studies indicate that the manner in which the hormones are administered to the animals may be critical. Ratio and level of the hormones do not require a high degree of precision, according to test results. This makes the method more practical for adaptation to commercial use.

The method has not been reviewed and approved for commercial use by the US Food and Drug Administration but it is hoped that some day commercial dairymen will be able to retain high-producing cows which have reproductive problems.

News from Ohio Agricultural Research and Development Center, Wooster, Ohio 44691; 28 July 1972.

(Taken from: AGRICULTURAL REPORT of the Agricultural Counsellor, Embassy of South Africa, Washington, D.C. as published by the Department of Agricultural Technical Services, Pretoria, 1972).



Front Row/Voorry: H. W. Aucock, C. P. Marwick, J. H. J. Coleman, P. H. Mapham, P. A. Gordon (Class Captain/Klasleier), Dr. R. K. Loveday (House Warden/Huisvader), Miss/Mej. J. E. Jordaan, J. D. Prinsloo, T. P. Spotswood, D. H. Havelaar, A. M. Navid, A. W. H. Smith, H. W. Bosch.

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Webber, J. J.
Woudstra, F. S.

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> Clinical Medal and Bag Kliniese Medalje en Tas Webber, J. J.

I.C.I. South Africa (Pharmaceuticals) Ltd.

Prys vir Chirurgie en Geslagskunde
Prize for Surgery and Genesiology

MacFarlane, M. D.

Spots

Medicine and Infectious Diseases Geneeskunde en Infeksiesiektes Spotswood, T. P.

Maybaker Clinical Prize Kliniese Prys

Francis, A. B. G.

Agricura Prize Agricuraprys Spotswood, T. P.

Pfizer Prize Pfizerprys Woudstra, F.S.

Prize awarded by the Reproduction Group of the SAVA Prys toegeken deur die Reproduksiegroep van die SAVA Webber, J. J.

> Lilly Laboratories Prizes -Lilly Laboratories-pryse

Diseases of Swine Varksiektes Aucock, H. W. Poultry Diseases Pluimveesiektes Holloway, J. M.

The Faculty of Veterinary Science expresses its sincere appreciation and thanks to the donors of the above-mentioned medals and prizes.

Die Fakulteit Veeartsenykunde betuig sy besondere dank en erkentlikheid jeens die skenkers van bovermelde medaljes en pryse.

## JOURNAL of the SOUTH AFRICAN VETERINARY ASSOCIATION

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#### MANAGEMENT COURSE FOR VETERINARY PRACTITIONERS

Good management practices are as important in a veterinary practice as in any business but there is little opportunity for the veterinarian to obtain formal instruction on the business aspects of veterinary practice.

Good management of a practice could mean the difference between success and failure or significantly improving on an existing practice.

It is vital that this gap be filled and the Veterinary Foundation in collaboration with the National Development & Management Foundation in Pretoria is planning a Management Course for veterinary practitioners.

This seminar will be the first of its kind for Veterinary Practioners and should be attended by all veterinarians in practice. It will also be a one day course for which the fee will be R15,00, morning and afternoon tea and midday lunch included.

Saturday, March 24th, 1973, has been selected as the day as convenient as any for the majority of practitioners. The Swartkop Country Club has been selected to provide a pleasant venue. The following is the preliminary programme, in which some of the practical as well as the theoretical aspects of management will be covered.

- 1. Welcoming address.
- 2. The importance of Scientific Management in running a Veterinary Practice.
- The principles of scientific decision making as applied to the Veterinary Practice.
- 4. The interpretation of financial statements.
- 5. A financial case study.
- 6. Practical applications of accounting methods and systems in Veterinary practice.
- 7. Legal aspects of the veterinary practice.
- 8. Human relationships.
- 9. Film Show "Interviewing".
- Panel discussions.

Formal applications will be sent out in due course but in the interim, cheques may be made payable to the South African Veterinary Association and forwarded to the Secretary. Early action will avoid any possible disappointment.





#### ABNORMALE HORINGGROEI IN 'N KOEDOE

Die skelet van 'n groot, uitgegroeide koedoebul is deur dr. T. W. Naudé in 'n dig beboste kol op die plaas Bezuidenhoutskraal in die distrik Cullinan naby Pretoria gevind.

Soos op die foto gesien kan word, het die regterhoring abnormale groei ondergaan, met so 'n sterk rostro-ventrale kromming dat die afgeknotte punt in die oog gesteek het. Die enigste spoor wat die onvermydelike oogbesering op die skedel agtergelaat het, was 'n retroversie van die ventrale orbita-rand, vermoedelik 'n effek van volgehoue druk op die oogbal.

Daar is nog heelwat wild op die plaas, van die roofdiere selfs die enkele hiëna. Die plaasmense het nooit die betrokke bul lewendig gewaar nie.

Fotografie: A. M. du Bruyn.

#### ABNORMAL HORN GROWTH IN A KUDU

The skeleton of a large, mature Kudu bull was found by Dr. T. W. Naudé in a thicket on the farm Bezuidenhoutskraal, Cullinan district, near Pretoria.

As can be seen on the photo, the right horn had undergone abnormal growth, with such a marked a rostroventral curvature that the truncated end projected into the eye. The only trace that the inevitable eye injury had left on the skull was a retroversion of the ventral orbital rim, presumably the effect of sustained pressure on the ocular bulb.

There is still a considerable amount of game on this farm, of the beasts of prey even the occasional hyena. The affected bull had never been seen alive.

Photography: A. M. du Bruyn.