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DESEMBER 1986

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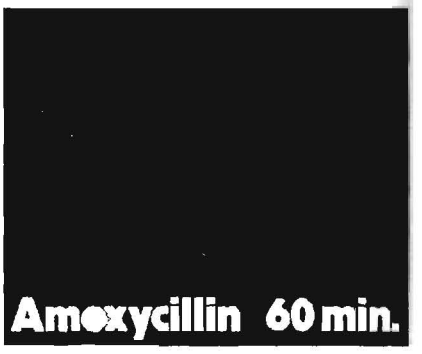

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SIR ARNOLD THEILER MEMORIAL LECTURE
SIR ARNOLD THEILER-GEDENKLESING

THEILER GEDENKLESING*

B.C. JANSEN**

Geagte Meneer die Dekaan, ek beskou dit as 'n besondere voorreg om deur lewering van die Theiler gedenklesing hulde te bring aan iemand wie se naam 'n legende geword het in veeartsenykunde in Suid-Afrika. Hy het as jong Switserse veearts die uitdaging wat veesiekte in Suidelike Afrika gebied het, aanvaar. Met sy inherente belangstelling in die natuur en biologie het hy gou te staan gekom in die middel van die stryd teen runderpes, perdesiekte en bloutong wat toentertyd in raaisels gehul was en onberekenbare ekonomiese verliese veroorsaak het. Weens sy bekwaamheid is hy gou ook betrek by die bestryding van menslike siektes soos pokkies.

Biologie was in die dae van Sir Arnold Theiler 'n beskrywende wetenskap soos duidelik blyk uit die talle nuwe parasiete wat toentertyd beskrywe is. Hy het geleef in die dae voor spesialisasie in veeartsenykunde toe die veld vir navorsing nog totaal braak gelê het. Geen wonder dus dat hy aan sy veelsydigheid kon uiting gee deur te beweeg op die gebied van virologie, protosoölogie, bakteriologie, helmintologie en patologie.

Theiler het egter ook geleef in 'n tydperk van vinnige uitbreiding van kennis en sal weens sy bydraes altyd gereken word as behorende aan die garde van manne soos Pasteur, Koch, Lister, Laveran en Bruce wat 'n revolusie teweeggebring het in die opvattings oor die besmetlike aard van siektes.

Onderstepoort sal altyd bly bestaan as nagedagtenis aan Theiler as die vader van veeartsenykundige navorsing en opleiding in Suid-Afrika.

*Voordrag gelewer by geleentheid van die 3de Fakulteitsdag van die Fakulteit Veeartsenykunde, Universiteit van Pretoria op 1 Oktober 1986.
Address delivered at the 3rd Faculty Day of the Faculty of Veterinary Science, University of Pretoria held on 1 October 1986.

**Dept. of Medicine, Faculty of Veterinary Science, University of Pretoria, P/Bag X04, 0110 Onderstepoort, Republic of South Africa.

But already during the last years of Theiler's life a revolution in biological research was started, the extent and significance of which could not have occurred to Theiler or any of his contemporaries. This revolution was fueled by a continuous series of advances and has been progressively gaining momentum to this very day. It has reached the stage where scientists can understand many disease processes at the molecular level.

The revolution started with the discovery of transformation in pneumococci by Griffith in 1928. He converted strains of non-virulent pneumococcus bacteria into capsulated virulent strains by exposing them to virulent cells that had been killed by high temperature. In the early 1930's the protein nature of all enzymes became universally accepted and in the early 1950's Frederick Sanger worked out the exact sequence of amino acids of the enzyme insulin.

Slowly the one-gene-one-protein concept emerged with important support from studies on sickle-cell anaemia by the chemist Linus Pauling. But everybody realised that the one-gene-one-protein idea, important as it was, could provide no clue to the molecular mechanisms involved in the cell so long as the nature of the gene itself remained a total mystery.

Through a series of advances, DNA which appeared to locate exclusively in the nucleus, and RNA, which was found in the nucleus and cytoplasm, were discovered. But the problem remained to decide whether the gene was made of DNA, proteins or RNA. The first hint that the essential genetic material could be transmitted from one organism to the next came from the abovementioned observation by Griffith. But Avery, McCarty and Macleod finally identified the active transforming fraction as DNA.

Stepwise progress was made in various laboratories using sophisticated techniques such as X-ray crystallography to determine the spatial arrangement of the atoms within compounds. This culminated in 1953 when James Watson and Francis Crick showed that the

DNA molecule is a double helix in which two polynucleotide chains running in opposite directions are held together by hydrogen bonds between pairs of centrally located bases.

The basic features of the double helix were simple and told how DNA stores genetic information which resides in the linear sequence of the 4 bases. They also suggested a chemical mechanism for the self-replication of DNA. From this moment on, the way in which geneticists investigated the gene entered a completely new phase and fundamental discoveries followed in rapid succession.

The transcription of RNA upon DNA templates and the role of the various types of RNA in protein synthesis were elucidated. It was shown that the genetic code is largely, if not entirely universal in all living beings and plants. The replication of even the smallest of viruses is a very complicated affair, achieved only with the aid of highly evolved genetic regulatory systems designed to see that the right molecules are synthesized at just the right time in the life cycle of the virus.

Biochemists could now combine their expertise with that of geneticists and concentrate on proteins that function as enzymes, catalyzing the several thousand biochemical reactions that in the aggregate constitute the metabolism of living cells.

The identification of reverse transcriptase in 1970 and the discovery of DNA restriction enzymes in 1968 and their subsequent application constitute the cornerstone of the all-important recombinant DNA technology of the present day. DNA can be cut apart, modified and reassembled, it can be amplified to many copies. With DNA one can generate RNA and then protein molecules of predetermined size and constitution. Vaccines now being designed with the use of recombinant DNA technology will have a great impact on animal disease control in the future. During the 1970's it was demonstrated that proteins isolated from the surfaces of viruses and some bacteria could induce the production of neutralizing antibody and protect animals against challenge with homologous agents e.g. short segments cleaved from the surface proteins of the foot-and-mouth disease virus served as effective immunogens. It is pleasing to know that Onderstepoort has made considerable advances in this field by identifying the polypeptide fraction responsible for the serotype specificity of the bluetongue virus. The gene encoding for this fraction has also been identified. These sophisticated laboratory procedures are also applied to the study of various other pathogens e.g. the retrovirus of 'jaagsiekte' and *Babesia* parasites.

The production of proteins by molecular cloning has developed since 1973. Cloning, more than any other single factor, has changed the face of biology. It consists of splicing a segment of DNA representing a gene encoding for the desired protein into a bacterial plasmid or viral DNA and subsequently transferring it to a single-celled host for replication of the guest gene and its expression as protein. Cloned protein vaccines have several advantages over whole-agent vaccines. They are non-infectious and stable to temperature variation.

Thus biology in 1986 is dramatically different from its antecedents only 10 years ago. New investigative techniques have made commonplace many experiments that were previously far beyond the reach of even the cleverest experimental biologist. The new molecular biology has done much more than expand the repertoire of laboratory techniques. It has with remarkable rapidity, established a biotechnology industry. Molecular biology has changed the ways people think about living things because they have come to understand the fundamental aspects of life processes. Investigators nowadays think about biological systems in terms of their molecular components and they have come to manipulate molecules. Biologists have become biochemists. They now possess the highly sophisticated electron microscopic, biochemical and genetic engineering procedures to let them tackle the cell's almost overwhelming complexity. Even children at school are familiar with the double helix of DNA as the symbol of the biological revolution that began earlier during this century.

Sir Arnold Theiler, due to the period during which he lived, could not contribute to the revolution in biological research. But his ideal of service, consciousness of endeavour, his pride in a task, his confidence of success in the face of difficulties will always serve as a stimulus to the scientists at Onderstepoort who cannot avoid applying the most advanced modern techniques.

Sir Arnold Theiler het in 1920 die veeartsenykundige fakulteit te Onderstepoort gestig. Hy het navorsing ten volle geïntegreer met opleiding en as leermeester van sy hoogste prestasies bereik. Die klem van veeartsenykundige opleiding het geval op tropiese siektes en parasitologie met die gevolg dat die graduandi goed toegerus was om die heersende probleme die hoof te bied en betrokke te raak by betekenisvolle navorsing. Maar met die jare het veeartsenykunde in verskillende rigtings ontwikkel en het die behoeftes van opleiding dienooreenkomstig verander. Van veeartse word nou verwag om te voorsien in die behoeftes van troeteldiere, 'n hoogs gespesialiseerde melkbeesbedryf, voedselhigiëne, die pluimveebedryf, die farmaseutiese bedryf en nog verskeie ander rigtings. Deur die breë basiese opleiding in die voorgraadse kursus en aangevul deur gespesialiseerde nagraadse opleiding word graduandi gelewer wat meeste van die vertakkings van die veeartsenykunde goed kan behartig. Maar of ons veeartsenykundiges voorsien met 'n genoegsame basis in biochemie om met vertroue sekere navorsingsrigtings te betree is hoogs twyfelagtig. Dit stem tot kommer want as ons nie nou navorsing doen op die mees gevorderde wyse nie, sal ons gou nie meer oplossings hê vir ons mees brandende probleme nie. Die min veeartse wat navorsing as 'n permanente roeping kies, beskou ek as 'n probleem wat dringende aandag vereis van beide opleidingsinrigtings en die persone in beheer van navorsing.

ADDRESS **VOORDRAG**

THE VETERINARIAN'S ROLE IN FOOD PRODUCTION**

F.J.H. LE RICHE*

The news headlines of the eighties describe the worst world-wide economic crisis in half a century. In many countries incomes are falling. Record budget deficits plague national and local governments on every continent. The external debts of several countries in the third-world and Eastern Europe verge on the unmanageable. Corporate bankruptcies in major industrial countries are more numerous than at any time since the great depression. Unemployment races upwards in both industrial and developing countries. More countries are threatened with famine than in any other time in the modern era.

The unprecedented doubling of world food supplies over the last generation was achieved in part by adopting agricultural practices that lead to excessive soil erosion. Erosion that is draining the land of it's productivity. After a point agriculture can then no longer be sustained and the land is abandoned.

Although technology has greatly expanded the earth's human carrying capacity, most obviously, with advances in agriculture the human ingenuity embodied in advanced technology can raise the natural limits on human economic activity, it cannot entirely remove them.

Amongst the most serious problems facing our world are surely those of a political nature, the threats of nuclear wars and the lack of understanding between the peoples and the populations of the various continents. The rapid growth of the world's population is surely one of the most significant problems we have to tackle. This leads to a depletion of resources, deforestation, overfishing, over-grazing and all the problems which we as people interested in the production of food, have to contend with. In all spheres of human endeavour much is being done to reduce the effects of this bleak picture. We, however, have only at this conference to deal with aspects relating to securing food supplies and the part that various scientifically trained people play in this picture.

Measured just in terms of output, the past generation has been one of unprecedented progress in world agriculture. In 1950 the world's farmers produced 320 million tons of grain. In 1984 they produced nearly 1,5 billion tons – an increase of nearly 1,2 billion tons was all the more remarkable because it occurred where there was little new crop land to bring under the plough.

On closer examination this 34-year-span breaks into two distinct eras – before and after the 1973 oil price increase. Modern agriculture thrives on cheap energy and the age of cheap energy came to an end in 1973. For twenty three years the world food-output expanded at over 3% per year whereas since 1973 this figure has dropped to 2% per annum and the world's farmers are struggling to keep pace with the increase in population.

The global increase in world food-output also obscures wide variations in individual geographic regions. In North America production has steadily outstripped demand, generating ever larger export surplusses. In the Soviet Union output has fallen behind demand over the past decade making the country the largest grain importer in history. In Africa which has a population of 550 million and which has to feed 14 million additional people each year, food production per person has fallen steadily since 1970.

Despite trippling of grain imports since then, hunger has become chronic and an enduring part of the African landscape. When world food supplies are discussed or measured, the only or principle criteria is the production of world *grain*. The actual part played by animal fats and proteins is insignificant and disregarded in terms of a measuring yard-stick.

In 1950 the world population was 2,5 billion people and during that year 320 million tons of grain was produced. In 1984 the world population was 4,66 billion people and 1,5 billion tons of grain was produced. The actual grain production per person in kilograms was 248 in 1950 and this has increased to 310 in 1984.

The principle reason for this increase has been the correct and much larger usage of fertilizer in crop lands. During the period under discussion the use of chemical fertilizers between 1950 and 1984 climbed from 15 million tons to 114 million tons. Nearly and 8-fold increase within a generation.

On a world-wide basis these figures are interesting because the increases took place primarily in North America and in Europe. Today, the countries with significant exportable surplusses of grain can be counted on the fingers of one hand namely the United States, Canada, Australia, Argentina and France. Of these the United States accounts for over half and with Canada covers close to 70% of the total. The rest of the world's dependence on these supplies, varies widely.

A FOA team of agronomists assessing the food situation in Africa in the late 1984's, identify 22 countries where crisis seemed eminent. Amongst these were Angola, Botswana, Central African Republic, Chad, Ethiopia, Ghana, Lesotho, Mozambique, Senegal, Somali, Swaziland, Tanzania, Zambia and Zimbabwe.

Now in 1986 certain of these countries like Zimbabwe have at least been able to produce enough grain for their own demand.

The position in Africa, however, remains alarming and we in southern Africa have also suffered because of the droughts which we have had over the last 4 years. South Africa itself has had to import grain which was something we never expected and never thought could happen.

In spite of the difficult years which we've lived through, it is interesting to note that the growth in

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**Opening address presented at the biennial scientific congress of the SAVA, Sandton Holiday Inn, Johannesburg, August, 1986.

agricultural exports from South Africa to the rest of Africa has grown from 1976 to 1984 by more than 25%. The population of the Republic of South Africa and the National States over the last two decades has increased by 21%, whereas agricultural production has increased by 34%. The Republic of South Africa which is only 3,75% of the total area of Africa, produces more than 33% of the total agricultural products of this continent.

On analysing the total agricultural production in South Africa, it is interesting to note that in spite of this country being a relatively large wool and mohair producer, that only 33% of the total agricultural production is derived from animal sources. The significance therefore of animals is considerably less than that of products produced in crop production. The scientifically trained people engaged in animal husbandry include, the geneticists, the chemists, the nutritionists and the wide variety of qualified people who handle the various aspects of animal production on products obtained.

The veterinarian plays his part and I believe that this part is far too specific and limited in the total picture of food production in our country.

The veterinarian, most certainly, plays a very important part in the area of small animals and more particularly in the fields of pets in the cities. Here I have no views other than the highest praise to your profession.

An analysis of the South African scene until the turn of the century indicates that the population will increase to about 48 million people. More people percentage wise will live in the urban areas and therefore also the food habits and food requirements will change to a large degree. Grain and other crop and horticultural products will make up approximately 75% of the daily diet, while animal products will constitute only 25% of the total. This figure even may be on the high side.

Some years ago Dr Von Marle projected that the animal product usage in the year 2000 would be more or less as follows:

(1) Milk: 5 400 million litres will have to be produced from 1,2 million cows at a rate of 4 500 kg of milk per lactation of 15 litres per day. This compares with the figure of approximately 9 litres per day average at present.
(2) Beef: 810 thousand tons will be required and this could only come from 3,7 million carcasses each weighing 220 kg. In order to achieve this our beef herd will have to increase to approximately 14 million animals.
(3) Lamb and mutton: 280 thousand tons will be required from 40 million sheep and 15 million carcasses of approximately 18,5 kg each.
(4) Pork: We will require 180 thousand tons from 198 sows producing 902 kg pork per sow per annum.
(5) Poultry: 870 thousand tons of which 90 thousand tons will come from old laying hens and 780 thousand tons from broilers. Our present production is in the vicinity of 350 thousand tons per annum.
(6) Eggs: We will require 5 600 million eggs produced by 20 million hens at 280 eggs per hen per annum. Our present production of eggs is just over 3 000 million eggs per annum.

In order not to destroy our land and the feed resources, a great deal of *intensification in animal production will* have to take place with immediate effect. This challenge becomes far greater in South Africa because of the tremendous learning curve still to be experienced by our black farmers in the National States. Similar sentiments were recently experienced by Dr Frans van der Merwe, the Chief Director of Animal Production of the Department of Agriculture. Greater production per animal and greater intensification is the only answer for us in South Africa. He indicated that significant results have over the last 15 years been obtained in programmes of performance testing of various animal groups. These are only indications of what is possible. In these tests which were limited and controlled by animal scientists the following improvements were recorded:

(1) Milk production over 15 years improved from 920 kg per day to 4 500 kg per annum per cow per lactation. The intercalving period improved by 10 days. This is still not acceptable but at least improvements have been recorded. At present the average milk production per cow in our national herd is only 2 100 kg against the 4 500 which were obtained in performance tested herds. This clearly indicates that over the next 14 or 15 years we have to double the production of our national herd.
(2) From 1971 to 1982 the average weaning weight, that is at 205 days, of beef calves improved from 175 kg to 195 kg. Our herd average today would be less than 150 kg per calf.
(3) From 1974 to 1982 the feed conversion in performance tested pigs improved by 12,6%, while the fat thickness decreased by 25%.
(4) Lamb carcasses, and the experiments were done in Dorpers, improved by 9,2 kg over the same period.
(5) The average weight of broilers improved by 200 gm to 2 kg over a period of 7 years, while the feeding period decreased by 7 days to just under 7 weeks.
(6) The average egg production over 17 years improved from 90 eggs to 270 eggs per hen per annum.

Mr President, the above figures clearly indicate that in order to feed the people of South Africa, a major input is required by the managers and the scientists engaged in animal production. In South Africa the animal scientists are making a major contribution to this effort but they too feel that their training should be extended so as to cover a wider field of knowledge in general.

Where does the veterinarian fit into the total picture and should changes in his contribution be made or not.

The contribution made by the Department of Veterinary Services is invaluable. They are primarily engaged in research, testing of medicaments and the developing and production of vaccines, etc. The overall work done, and part played by Onderstepoort in South Africa, has in certain instances ensured the survival of certain animal groups. There can be no doubt at all about the positive and enormous part Onderstepoort and the Department of Veterinary Services are playing in the overall picture and production of animal proteins, fats and related products.

I see a problem, or an area for discussion, when we get to the practising veterinarian.

He plays a significant part in the cities and in the field of pets overall.

In the rest of his area of activity, I believe he is too specialised in veterinary medicine and limited in his training and approach over too great a span of animals to be a real constributer to food production and to the improvement of our herds in South Africa.

In addition to this limitation I venture to tread upon very dangerous ground by indicating that he often presents the picture of being an aloof person who finds

terrific satisfaction in his professional status.

The practising veterinarian, with his training and knowledge should play a far greater part to improve our food production to the norms I mentioned previously. There should be a far greater co-operation between him and the Animal Scientist. His knowledge of nutrition, genetics, chemistry, breeding practices and management, could be conveyed to the farmer to great advantage of all.

In South Africa we have to improve the production capacity of our animals in all fields and the practising veterianarian has a new part to play. He must make a greater effort to be part of the *agricultural scene* and not only find satisfaction in his professional status.

A further thought is that the veterinarian should be able to major in e.g.: small animals and pets; large animals; poultry; horses, etc., etc.

Die praktiserende veearts kan 'n baie groter en meer beduidende rol speel in die verbetering van ons diereproduksiegroepe en volgens my beskeie mening, kan hy die rol vervul van super inligting- en praktiese hulpbron vir die boer. Ons sal hom beloon en saam ons land ontwikkel.

Meneer die President, dit was vir my 'n voorreg wat aan my toegestaan was om hierdie kongres te open. My beste wense aan u vir hierdie kongres en ook vir die toekoms vir die veearts in Suid-Afrika. Ek glo dat 'n kongres van hierdie aard veel sal bydra tot 'n nuwe siening van die veearts se plek in die produksieketting in ons land.

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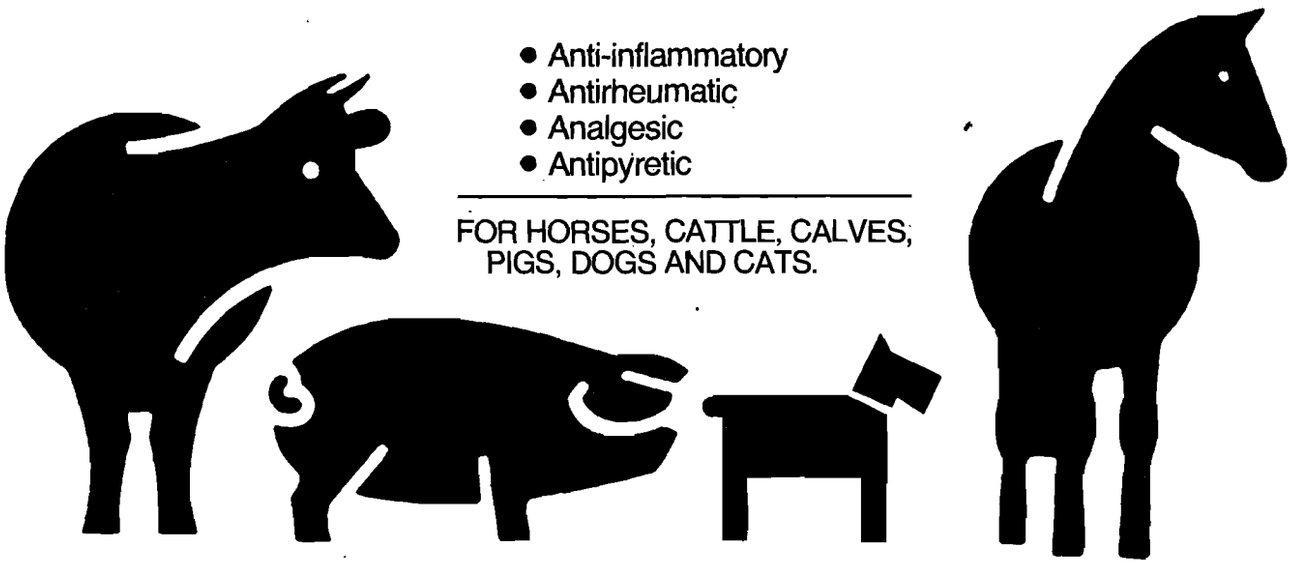

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ARTICLE ARTIKEL

THE PREVALENCE OF TEAT CANAL INFECTIONS IN LACTATING DAIRY COWS AS DETERMINED FROM FOREMILK AND TEAT CANAL SWAB SAMPLES

J.H. DU PREEZ*

ABSTRACT: Du Preez J.H. **The prevalence of teat canal infections in lactating dairy cows as determined from foremilk samples and teat canal swab samples.** *Journal of the South African Veterinary Association* (1986) **57** No. 4, 193-198 (En) Department of Veterinary Public Health, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, 0110 Onderstepoort, Republic of South Africa.

The diagnostic reliability of techniques for determining the prevalence of teat canal infections varies: bacteriological results obtained by examination of teat canal swabs were 20% higher on average than those of foremilk samples (FMS) examined and classified according to the criteria of the International Dairy Federation (IDF). Since they are based exclusively on the bacteriocytological results of the FMS, the IDF criteria for the classification of the various forms of subclinical udder conditions do not permit an accurate classification of the health status of the udder.

Key words: Foremilk sampling, teat canal infections, teat canal swab sampling

INTRODUCTION

Long before the development of knowledge relevant to the aetiology of mastitis, detection of the disease was based upon examination of the milk for gross deviations from the normal and upon the physical examination of the udder for swelling, scar tissue and atrophy[30]. In 1916 Moak[25] developed a procedure for the examination of foremilk for evidence of mastitis. The California Mastitis and Whiteside Tests, both used for the detection of subclinical mastitis, are based on the presence of somatic cells in milk[31]. The conductivity of milk is another method used for the detection of mastitis[11 23]. Dodd[5] pointed out the need for further research into teat canal infections or colonization (TCI). Classification of subclinical bovine mastitis according to the International Dairy Federation's (IDF) criteria[21 22] does not provide for a condition such as TCI.

From literature on clinical and subclinical mastitis[7 24 34] it is apparent that the disease complex is in all respects multifactorial. From various points of view it is equally clear that the diagnosis of subclinical mastitis depends on definition and diagnostic criteria which must therefore be pathologically, aetiologically and pathogenetically correct[15].

Modern bacteriological and cytological techniques are much more sensitive for the diagnosis of bovine mastitis than those available at the turn of the century. The sensitivity of these methods has apparently engendered great confidence in their use in the diagnosis of mastitis. During the early seventies this culminated in a definition of clinical and subclinical mastitis as well as the standardization of diagnostic criteria by the IDF[21 22].

The IDF definition of mastitis is based on recommendations made by Kästli[21] and Tolle[32]. The definitions given by Kästli[21] are as follows:

(1) Normal udders are those which show no outward sign of a pathological condition and produce milk which is free from pathogenic organisms and has a normal cell count.
(2) Latent infections are present when the milk shows the presence of pathogenic organisms but nevertheless has a normal cell count.
(3) Subclinical mastitis shows no macroscopic evidence of inflammation but examination of the milk reveals udder infection, an increased cell count and also changes in the chemical properties of the milk.
(4) Clinical mastitis: acute mastitis is present when there are obvious symptoms of inflammation of the udder such as heat, pain and swelling. The milk is macroscopically abnormal and the animal may have an elevated body temperature. Subacute mastitis is present when there are persistent clots especially in the foremilk.
(5) Non-specific or aseptic mastitis is present when there is no recognisable infection and the symptoms may be subclinical of clinical.

All these definitions are associated with the cell counts of the milk[21]. Therefore the following cell counts have been proposed and established. "With due consideration to Table 1, a threshold value of more than 500 000 cells per ml is suggested as indicating that the cell count is abnormal and that a diagnosis of mastitis has been established. This threshold value is acceptable for diagnostic classification on condition that the milk is sampled: from the first fractions of milking; from cows in normal lactation; aseptically at milking times".[21]

Tolle[32] explains and elaborates further on the definition of mastitis. The term is derived from the Greek word *mastos*, meaning breast and in this context udder, and the suffix *itis* meaning inflammation of [31]: "Mastitis is an inflammatory change of the mammary gland which, along with physical, chemical and microbiological changes, is characterised by an increase of somatic cells, especially leucocytes, in the milk, and by pathological changes in the mammary tissue".

From the above it is apparent that the diagnosis of mastitis in lactating cows is currently based on clinical examination of the udder tissue and its secretion, the somatic cell content of milk, and bacteriological data. The general emphasis on the cytological and bacteriological parameters has promoted the diagnostic importance and use as criteria of potentially pathogenic bacteria and elevated somatic cell counts (threshold 5×10^5 cells per ml) in aseptically collected samples of foremilk[15].

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However, neither the historical development[15] nor the more recent standardization of the diagnoses of subclinical udder health conditions[21 22] guarantees that the present diagnosis of such conditions is correct. In fact, it is evident that both the definition and diagnostic criteria recommended by the IDF[21 32] are rather controversial[15 18]. Tolle[33] suggests the need for a revision of the definition of mastitis. Doubts should be resolved on the present concepts of latent udder infection[26 27 35] and non-specific or aseptic mastitis[16 27].

A major reason for the controversy is the initially complete[21 32] and more recently[22] almost equally inadequate recognition by the IDF of the rather important udder health problem of teat canal infections. According to the definitions and criteria proposed by Kästli[21] and Tolle[32] no such condition as TCI exists, nor can it be diagnosed. However, it has long been known that TCI in fact not only exists, but can be diagnosed and is of particular significance in regard to the development of mastitis.

The diagnosis of TCI is made by means of examination of teat canal swab foremilk samples[4] and teat canal swab, foremilk and teat-wall puncture milk samples[13]. Other researchers[1 17–20 35 36] bacteriologically examined milk samples collected aseptically both via the teat canal and through the wall of the udder or teat cistern of the same quarters in order to prove the separate existence of TCI. From such investigations it is clear that the isolation of bacteria from milk may frequently indicate TCI and not intramammary infection[15 18 26] as suggested in the IDF[21 32].

Major difficulties are still being encountered in the diagnosis, therapy and prevention of subclinical mastitis in particular[8 14]. Giesecke & Van den Heever[15] consider that the combination of bovine serum albumin levels in milk and the IDF criteria may permit accurate assessment of udder health and TCI.

The aim of this investigation was, firstly, to classify TCI according to the existing IDF definitions and criteria no such condition exists nor can it be diagnosed. Secondly, to compare the prevalence of TCI as diagnosed according to the results of examination of teat canal swab (TCSS) and foremilk samples (FMS) from the same quarter.

MATERIALS AND METHODS

The investigation was performed on 365 Friesian dairy cows (1 430 quarters) on a zero grazing system of a herd milked by hand three times a day on the Transvaal Highveld. The standard of management, animal husbandry and hygiene in the herd was relatively poor.

Collection of FMS and TCSS

Sampling routine: After thorough udder washing with clean running water, drying with a disposable paper towel, disinfecting the teat with 70% alcohol and cotton wool and discarding the initial three jets of foremilk, a quarter milk sample was aseptically collected into a sterile 5 ml 'Monoplast' (Labotec, Johannesburg) tube, following the standard procedure[22].

The teat canal was thereafter swabbed with a special sterile miniature teat canal swab made by wrapping good quality highly absorbent cotton wool for a distance of 3 – 4 mm around the tip of 3,5 cm long, slim wooden toothpicks. Twelve swabs were placed in a 20 ml screw cap vaccine type bottle and sterilised in an autoclave. The swabs were carefully removed from the storage container and the cotton wool tip was inserted into the teat canal for a distance of 3 – 6 mm, whereafter it was placed in a sterile 5 ml 'Monoplast' tube to facilitate transport. Residual milk in the teat canal after foremilk sampling ensured comfortable teat canal swab sampling. FMS and TCSS were obtained immediately before routine milking. All samples were transported on ice and examination proceeded within 3 – 6 hours of sampling.

Before implementation of this sampling regime the effectivity of the disinfection process was established by swab sampling and culturing in a test sample size of 50 teat tips subsequent to disinfection (99% were bacteriologically negative).

Somatic cell count

A defatted Prestcott-Breed smear[2] of each foremilk sample was stained according to the Broadhurst-Paley method[30]. The somatic cell count per ml milk was determined microscopically after counting the cells in 10 microscopic fields[2].

Isolation and identification of bacteria

Facultative anaerobic and micro-aerophilic bacteria were isolated by streaking a loopful (± 0,01 ml) of foremilk sample onto each of two blood tryptose agar plates. Both plates were incubated at 37° C for 48 h, one of them under micro-aerophilic conditions (± 8% carbon dioxide). The swabs were similarly streaked and cultured. In addition, a loopful (± 0,01 ml) of each FMS and the aseptically removed tip of each swab were separately enriched by incubation in 5 ml serum broth prior to streaking onto solid media as above. This served as a back-up in those cases where no growth materialised on blood tryptose agar plates. The identification of bacteria was done according to the methods and criteria described by Cowan & Steel[3].

Anaerobic bacteria: For several practical reasons samples were not examined for anaerobic bacteria. Du Preez[9] has shown that with a few exceptions of so-called aseptic mastitis (ASM), anaerobic bacteria are always isolated together with facultative anaerobic or micro-aerophilic bacteria.

For the purpose of this report it was assumed that the presence of bacteria in TCSS indicates TCI.

Statistical evaluation

The log-linear model was used for statistical analyses[10].

Health status of quarters

The health status of quarters, determined on the basis of clinical examination and data obtained by examination of FMS, was classified according to the IDF criteria but the health status of quarters, diagnosed by means of the TCSS, was classified according to the IDF criteria based on the cytological results of the FMS and bacteriological results of the TCSS.

RESULTS

From the results it was apparent that the udder health states which were diagnosed, differed according to the type of sample investigated. Such differences become still more obvious from a comparison of the percentage values of udder quarters classified according to their health status (Table 2).

Table 1. Normal quarters (N), teat canal infection/colonization (TCI), aseptic mastitis (ASM) and subclinical mastitis (SCM) diagnosed according to the results of the foremilk sample (FMS) and teat canal swab sample (TCSS), respectively (n = 1 430 quarters)

Status of the quarter	FMS	TCSS	Difference between TCSS and FMS	FMS-isolates				TCSS-isolates			
				Sa	Sc	O	Sa∝Sc	Sa	Sc	O	Sa∝Sc
N	849	528									
TCI	241	562	321	201	30	4	6	430	74	5	53
ASM	172	118	54								
SCM	168	222	54	91	63	4	10	148	53	6	15
Total	1 430	1 430	750	292	93	8	16	578	127	11	68

Sa = *Staphylococcus* spp.
Sc = *Streptococcus* spp.
O = Other mastitis pathogenic bacteria, soil bacteria, saprophytes and fungi

Table 2. The prevalence of normal quarters (N), teat canal infection/colonization (TCI), aseptic mastitis (ASM) and subclinical mastitis (SCM) diagnosed according to the results of the foremilk sample (FMS) and teat canal swab sample (TCSS)

Status of quarter	FMS %*	TCSS %*	Percentage between TCSS and FMS	FMS-isolates %				TCSS-isolates %			
				Sa	Sc	O	Sa∝Sc	Sa	Sc	O	Sa∝ Sc
N	59,4	36,9	22,4								
TCI	16,8	39,3	22,4	14,1	2,1	0,3	0,4	30,1	5,2	0,3	3,7
ASM	12,0	8,3	3,8								
SCM	11,8	15,5	3,8	6,4	4,4	0,3	0,7	10,3	3,7	0,4	1,0
Total	100,0	100,0		20,5	6,5	0,6	1,1	40,4	8,9	0,7	4,7

* = expressed in terms of the amount of quarters examined (n = 1 430)
The prevalence of clinical mastitis was 5%
Sa = *Staphylococcus* spp.
Sc = *Streptococcus* spp.
O = Other mastitis pathogenic bacteria, soil bacteria, saprophytes and fungi

Table 3. Existing International Dairy Federation's (IDF) and newly proposed classification of health status of quarters for subclinical bovine udder infection/inflammation as determined by the somatic cell count of the foremilk sample (FMS) and the bacteriology of the teat canal swab sample (TCSS).

Criteria			Diagnoses	
1 Somatic cell count x 10^3 per ml of milk	2 Mastitis pathogens in foremilk sample (FMS)	3 Mastitis pathogens in teat canal swab sample (TCSS)	IDF diagnosis from 1 and 2	Proposed diagnosis from criteria 1 and 3 in comparison with criterion 2
<500	Absent	Absent	Normal quarter	Normal quarters
<500	Present	Present	Latent infection	Teat canal infection and/or colonization (TCI)*
>500	Absent	Absent	Aseptic mastitis	Aseptic mastitis
>500	Present	Present	Subclinical mastitis	Subclinical mastitis**

*Although TCSS's data was responsible for the proposed classification, FMS diagnosed as latent infection according to the IDF's criteria must also be diagnosed and classified as TCI. Bacteriological positive TCSS diagnosed TCI exclusively and does not reflect the bacteriological status of the udder parenchyma
**SCM and TCI may occur simultaneously in the same quarter

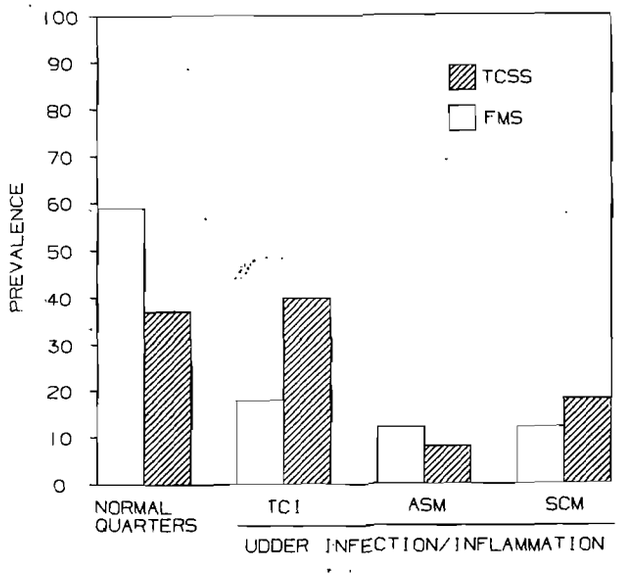
TCI: The prevalence of TCI diagnosed by means of the TCSS was 22,4% higher than when diagnosed by means of the foremilk sample. The 22,4% increase in TCI diagnosed by means of the TCSS corresponded with the 22,4% decrease in normal quarters diagnosed by means of the same method.

Most TCI and SCM were caused mainly by pure cultures of coagulase-negative staphylococci and *Staphylococcus aureus,* followed by a combination of the above-mentioned bacterial species, other pathogenic bacteria, soil bacteria, saprophytes and fungi.

Clinical mastitis: The prevalence of clinical mastitis was 5%.

TCI and SCM: In all cases where the FMS indicated a SCM the TCSS were bacteriologically positive.

The prevalence of TCI as determined by the examina-



tion of TCSS from all four quarters of 25 first and second lactation cows was 86% compared with 88% in 25 third to sixth lactation cows in the same herd, i.e. under identical conditions of relatively poor management, hygiene and animal husbandry.

The prevalence of TCI, diagnosed according to the TCSS, in 25 high-yielding cows (>15 ℓ/day), was 80% compared with 78% in 25 low-yielding cows (<10 ℓ/day) in the same herd.

The results of this investigation are illustrated in Fig. 1.

DISCUSSION

Tolle[33] has pointed out that the IDF definition of mastitis and infection was very useful but that in the light of modern knowledge the IDF's mastitis group should revise the definition with due regard to the dynamics of the udder. This study represents an extensive in-depth investigation into the prevalence of TCI.

The classification of the IDF criteria[21,32] for subclinical bovine mastitis does not provide for a condition such as TCI. Because all the corresponding TCSS of quarters diagnosed and classified according to the bacterio-cytological results of the IDF criteria as latent udder/quarter infection* were bacteriologically positive and because it is assumed that the presence of bacteria in TCSS indicates TCI, TCI must therefore be classified according to the IDF criteria in the same category as latent udder infection although the somatic cell count of a small percentage TCI exceeds 500 000/ml. According to the findings of this study, TCI and latent udder infection are two different entities but the existence of latent udder infection is questionable, as Newbould[26] and Verhoef & Smit[35] have already pointed out. The following classification of the health status of quarters for subclinical udder inflammation/infection are proposed (Table 3).

Some of those quarters classified as ASM according to the IDF criteria were proved to be TCI with a high somatic cell count. Although these TCI have a somatic cell count of more than 500 000/ml they should be classified in the same category as latent udder infection because only 3,8% quarters were involved. The present IDF criteria for the classification of the various forms of SCM fall short as a procedure for making accurate diagnoses since such a diagnosis can only be based on the bacterio-cytological results of the FMS.

Many quarters in this study, diagnosed according to the FMS and classified as normal according to the IDF criteria, were proved to be true cases of TCI by means of teat canal swab sampling. An explanation for this phenomenon could be that the bacteria adhere so strongly to the keratin and epithelial cells of the teat canal wall that with foremilk sampling, no contamination of the milk takes place from the teat canal.

In teat canal swab sampling the bacteria in the teat canal are removed, together with the keratin and epithelial cells, by a canal swab for purposes of culturing. The increased mechanical friction caused by teat canal swab sampling permits greater diagnostic accuracy than the flushing action during foremilk sampling.

With the use of the TCSS instead of the FMS for diagnosing udder infections, fewer quarters are indicated as being normal, in other words, teat canal swab sampling is a more sensitive and accurate diagnostic method for determining the health status of quarters.

With the use of the FMS to establish the health status of the quarter, false negative results (normal quarters) were obtained in more than 20 % of the cases, that is to say when compared with the results of the TCSS. The milk obtained from the quarter by FMS is therefore not necessarily a true reflection of the bacteriological status of the quarter.

Organisms multiplying in infected lesions or colonized teat canals are ideally situated for transmission to the parenchyma of the udder[28]. In other words, in routine diagnostic work based on cytobacteriological examination of FMS, some cases of TCI are consequently not diagnosed. This explains the 3,8% higher prevalence of ASM based on the results of examination of FMS compared with TCSS.

Toxins possibly liberated by the colonized bacteria in the teat canal are probably responsible for the increased somatic cell count in the so-called ASM cases. In this study it has been proved that the status of many quarter is incorrectly diagnosed as ASM on the grounds of the FMS – which implies that no bacteria are present – instead of TCI, where bacteria are in fact present. This TCI may possibly give rise to or develop into udder infections/mastitis, with all the harmful consequences involved. By relying solely on the FMS to establish the health status of a quarter, negative results may be obtained in certain cases in respect of the bacteriological infection of the quarter. By means of TCSS more cases of SCM are diagnosed than with a corresponding decline in the number of cases of ASM.

According to the FMS (IDF criteria) a quarter suffering concurrently from TCI and SCM cannot be identified as such. Only the SCM can be diagnosed because, in terms of the IDF criteria, the presence of elevated numbers of somatic cells and mastitis bacteria in the milk automatically results in the conditions being classified as SCM. This study shows that SCM may occur concurrently with TCI and be identified as such provided suitable samples are examined.

It is considered important from a diagnostic, clinical and therapeutic point of view to be able to distinguish

*Because foremilk is sampled from the bovine udder or quarter it is more explanatory to talk to latent udder/quarter infection and not of latent infection.

between SCM and TCI occurring simultaneously in the same quarter, since udder infection caused by *S. aureus* and *S. epidermidis* is maintained by TCI, according to Forbes & Herbert[13]. If the TCI can be eliminated, the incidence of udder infection could be reduced accordingly because for microbial invasion to lead to udder infection, micro-organisms have to migrate into the teat canal[12 29]. Dodd[6] has pointed out that the main reason for mastitis control is economic efficiency and that the main purpose of control programmes is to reduce microbial infections. Diagnosis of subclinical cases of mastitis based on IDF criteria does not produce a result which distinguishes between a TCI and a latent udder infection.

Where management, hygiene and standard of animal husbandry are poor, the prevalence of TCI does not increase with increasing age, number of lactations, increasing production and milkability. Since the udders were exposed to numerous bacteria, many environmental, managemental and hygiene factors predisposing to mastitis were active in this herd. The prevalence of TCI therefore appears not to be the only determining factor in the incidence of clinical mastitis and SCM although their prevalence is normally higher in older, higher yielding and more easily milkable cows. There is a correlation between the high prevalence of TCI and SCM and clinical mastitis. This statement is borne out by the fact that in this herd the prevalence of SCM, diagnosed according to the FMS and TCSS, was 11,75% respectively, and that of clinical mastitis 5%, whereas the prevalence of TCI, diagnosed according to the FMS and TCSS, was 16,8% and 39,3%, respectively.

CONCLUSIONS

Various mastitis pathogenic bacteria, soil bacteria, saprophytes and fungi colonize the teat canal. Only with TCSS can TCI be diagnosed accurately. In the routine examination of FMS many cases of TCI (up to 20% or more) are not recognised. TCI must be classified according to the IDF criteria in the same category as latent udder infection. The existence of latent udder infection is questionable. TCi and latent udder infections are two different entities. Many cases of SCM and ASM, classified according to the IDF criteria, are actually TCI. By using only the bacterio-cytological results of FMS, the IDF criteria for the classification of the various forms of SCM do not facilitate accurate classification of the health status of the udder. The prevalence of TCI, diagnosed according to the TCSS, is almost the same in high and low-yielding cows.

Statistical conclusions

Tables 1 & 2: In establishing the prevalence of TCI, ASM and normal quarters in this dairy herd, the results of the FMS and TCSS differ significantly (significance level (P) of less than 0,01%). In respect of SCM the difference is fairly significant ($P < 5\%$).

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REFERENCES

1. Black R F, Marshall R T, Bourland C T 1972 Locus of mammary gland infections of *Corynebacterium bovis*. Journal of Dairy Science 55: 413 – 416
2. Carter G R 1973 Diagnostic Procedures in Veterinary Microbiology, 2nd edn. Charles C Thomas, Springfield, Illinois
3. Cowan S T, Steel K J 1974 Manual for the Identification of Medical Bacteria. Cambridge University Press
4. Cullen G A, Herbert C N 1967 Some ecological observations on micro-organisms inhabiting bovine skin, teat canals and milk. British Veterinary Journal 123: 14 – 25
5. Dodd F H 1975 Review of the Seminar. In: Dodd F H, Griffin T K, Kingwill R G (ed) Proceedings of the International Dairy Federation Seminar on Mastitis Control, Bulletin Document 85: 501 – 506
6. Dodd F H 1980 Mastitis control. In: Bramley A J, Dodd F H, Griffin T K (ed) Mastitis control and herd management. Technical Bulletin 4, National Institute for Research in Dairying, Reading, England: 11 – 23
7. Dodd F H, Griffin T K The role of antibiotics at drying-off in the control of mastitis. In: Dodd F H, Griffin T K, Kingwill R G (ed) Proceedings of the International Dairy Federation Seminar on Mastitis Control, Bulletin Document 85: 282 – 302
8. Dodd F H, Jackson E R 1971 The control of bovine mastitis. National Institute for Research in Dairying, Reading, England
9. Du Preez J H 1981 Die prevalensie, aard en betekenis van anaërobiese bakterieë in die melkkoei-uier. MMedVet (Hyg) Verhandeling, Universiteit van Pretoria
10. Everitt B S 1977 The analysis of Contingency Tables. Chapman & Hall, London
11. Fernando R S, Rindsig R B, Sphar S L 1981 Effect of length of milking interval and fat content on milk conductivity and its use for detecting mastitis. Journal of Dairy Science 64: 678 – 682
12. Forbes D 1969 The pathogenesis of bovine mastitis. Veterinary Bulletin 39: 529 – 541
13. Forbes D, Herbert C N 1968 Studies in the pathogenesis of staphylococcal mastitis. The Veterinary Record 83: 69 – 73
14. Giesecke W H 1985 The diagnosis and control of bovine mastitis as a means of improving the quality of milk. South African Journal of Dairy Technology 17: 3 – 21
15. Giesecke W H, Van den Heever L W 1974 The diagnosis of bovine mastitis with particular reference to subclinical mastitis: A critical review of relevant literature. Onderstepoort Journal of Veterinary Research 41: 169 – 212
16. Giesecke W H, Van den Heever L W 1975 Milk hygiene considered in terms of recent observations on definition and diagnosis of bovine mastitis. South African Journal of Dairy Technology 7: 51 – 61
17. Giesecke W H, Van den Heever L W, Hope D C, Van Staden J J 1968 Laboratory mastitis diagnosis: The microbiological content of parallel teat and gland cistern milk samples from quarters of known status. Journal of the South African Veterinary Medical Association 39: 33 – 44
18. Giesecke W H, Viljoen M H 1974 The diagnosis of subclinical mastitis in lactating cows: A comparison of cytological methods and a monovalent radial immunodiffusion test. The Onderstepoort Journal of Veterinary Research 41: 51 – 74
19. Heidrich H J, Grossklaus D, Mülling M 1964 Vergleichende bakteriologische Untersuchungen an durch Handmelken und durch Venülenpunktion der Drüsenzisterne gewonnenen Milchen. I. Untersuchungen an Venülenmilchen. Berliner under Münchener Tierärtzliche Wochenschrift 77: 83 – 84
20. Heidrich H J, Mülling M, Birkner H W 1964 Vergleichende bakteriologische Untersuchungen an durch Handmelken und durch Venülenpunktion der Drüsenzisterne gewonnenen Milchen. II. Vergleichende Untersuchungen Handmelken Venülenmilchen. Berliner under Münchener Tierärtzliche Wochenschrift 77: 85 – 87
21. Kästli P 1967 Definition of mastitis. Annual Bulletin of the International Dairy Federation, Part III: 1 – 5
22. Laboratory methods for use in mastitis work 1981 Document 132. International Dairy Federation Bulletin. Square Vergote 41, 1040 Brussels, Belgium
23. Linzell J L, Peaker M, Rowell J G 1974 Electrical conductivity of foremilk for detecting subclinical mastitis in cows. Journal of Agricultural Science 83: 309
24. Mayer E 1980 Reports and summaries. XI International Congress on Diseases of Cattle. Association for Buiatrics, Tel Aviv, Israel
25. Moak H 1916 Control and eradication of infectious mastitis in dairy herds. Cornell Veterinarian 6: 36 – 40
26. Newbould F H S 1974 Microbial diseases of the mammary gland. In: Larson B L, Smith V R (ed) Lactation: A Comprehensive Treatise, Academic Press: 269 – 316
27 Pearson J K L 1975 What we are attempting to control – mastitis

or udder infection. *In:* Dodd F H, Griffen T K, Kingwill R G (ed) Proceedings of the International Dairy Federation Seminar on *Mastitis Control*, Bulletin Document 85: 59 – 61
28. Philpot W N 1979 Control of mastitis by hygiene and therapy. Journal of Dairy Science *62: 168 – 176*
29. Prasad L B M, Newbould F H S 1968 Initial response of the bovine mammary gland to invasion by *Staphylococcus aureus*. Canadian Veterinary Journal 9: 170 – 177
30. Schalm O W 1964 A *syllabus* on the bovine mammary glands in health and disease. Department of Clinical Pathology, School of Veterinary Medicine, University of California, Davis
31. Schalm O W, Carrol E J, Jain N C 1971 Bovine Mastitis. Lea & Febiger, Philadelphia
32. Tolle A 1971 A monograph on bovine mastitis. International Dairy Federation Bulletin. Square Vergote 41, 1040 Brussels, Belgium, Part I: 1 – 23
33. Tolle A 1975 Final conclusions. *In:* Dodd F H, Griffen T K, Kingwill R G (ed) Proceedings of the International Dairy Federation Seminar on Mastitis Control, Bulletin Document 85: 507 – 510
34. Van den Heever L W, Giesecke W H (ed) 1979 Proceedings – 1st SA Symposium on Mastitis Control in Dairy Herds. South African Vetrinary Association, Pretoria, Republic of South Africa
35. Verhoeff J, Smit J A H 1981 Bovine serum albumin and cell counts in the diagnosis of subclinical udder infections. The Veterinary Quarterly 3: 38 – 45
36. Wiesner H U 1969 Zur Aussagekraft von Staphylokokken befunden in Viertelgemelksproben. Vth Symposium of the World Association of Veterinary Food Hygienists, Opatija 397 – 402

ABSTRACT **SAMEVATTING**

LEPTOSPIRA INTERROGANS SEROVAR *HARDJO* ASSOCIATED WITH BOVINE ABORTION IN SOUTH AFRICA

Leptospira interrogans serovar *hardjo* was isolated from urine from dairy cattle in the Onderstepoort area. This was the first successful isolation of this serovar as sole agent causing an abortion storm in the Republic of South Africa. Abortions occurred as early as at 4 months' gestation. (Te Brugge, Lesley A. & Dreyer, Tersia, 1985. *Leptospira interrogans* serovar *hardjo* associated with bovine abortion in South Africa. *Onderstepoort Journal of Veterinary Research*, 52, 51-52 (1985).)

ABSTRACT **SAMEVATTING**

FLUCTUATIONS IN THE GLUCOSE LEVEL OF COW'S MILK FROM NORMAL AND SUBCLINICALLY DISEASED UDDERS

Individual quarter samples from some 19 cows on average were investigated monthly over 12 months for determining the udder health status of cows and the glucose concentrations of foremilk and strippings.

Foremilk showed a mean 0,1311 mM concentration of glucose which remained fairly stable during the period of investigation and lactation. A fluctuating mean value of 0,2037 mM was determined in strippings in which glucose levels were consistently and appreciably higher than those of foremilk.

Foremilk from completely normal quarters and others affected by non-specific cellular reaction, relevant or irrelevant teat canal infection and aseptic or septic subclinical mastitis, showed mean glucose concentrations of 0,1410; 0,1392; 0,1337; 0,1417; 0,1262 and 0,1248 mM, respectively. Strippings from the same quarters showed corresponding values of 0,2056; 0,2861; 0,2100; 0,1733; 0,1661 and 0,1617 mM glucose.

(Giesecke, W.H., Durand, Anette M. & Petzer, Inge-Marié, 1984 Fluctuations in the glucose level of cow's milk from normal and subclinically diseased udders. *Onderstepoort Journal of Veterinary Research*, 51, 15-19 (1984).)

ARTICLE ARTIKEL

A COMPARISON OF THE TICK BURDENS OF WILD ANIMALS IN A NATURE RESERVE AND ON AN ADJACENT FARM WHERE TICK CONTROL IS PRACTISED

I.G. HORAK* and M.M. KNIGHT*

ABSTRACT: Horak, I.G.; Knight, M.M. **A comparison of the tick burdens of wild animals in a nature reserve and on an adjacent farm where tick control is practised.** *Journal of the South African Veterinary Association* (1986) 57 No. 4, 199-203 (En) Tick Research Unit, Department of Zoology and Entomology, Rhodes University, 6140 Grahamstown, Republic of South Africa.

Acaricidal treatment of cattle, sheep and goats on a farm in Valley Bushveld in the Grahamstown district resulted in a reduction in the numbers of certain tick species on kudu (*Tragelaphus strepsiceros*), scrub hares (*Lepus saxatilis*) and crowned guinea fowl (*Numida meleagris*) on the same farm. The tick species most severely affected was *Amblyomma hebraeum*, while *Haemaphysalis silacea, Hyalomma marginatum rufipes* and *Rhipicephalus appendiculatus* were affected to a lesser extent. *Rhipicephalus glabroscutatum* and *Rhipicephalus oculatus* appeared to be unaffected.

Key words: Tick burdens, wild animals, domestic stock

INTRODUCTION

Large herbivores such as kudu (*Tragelaphus strepsiceros*), eland (*Taurotragus oryx*) and buffalo (*Syncerus caffer*) are generally better hosts of adult ixodid ticks than are smaller antelope species[5 8 14]. Both the large and small herbivores may be efficient hosts of immature ticks[5 8]. This implies that if only small antelope species are kept in small or medium-sized nature reserves, massive tick infestations are unlikely to occur because of the absence of large herbivores which could maintain large populations of adult ticks.

It has been suggested that, where wild animals run with domestic stock, effective control of the ticks on the domestic animals will also reduce tick burdens on the wild animals[8]. Consequently the wild animals on such a farm will no longer serve as a possible source of tick infestation for the domestic stock.

An opportunity to test this hypothesis arose during a survey conducted to ascertain the seasonal abundance of ixodid ticks on cattle, sheep, goats, kudu, scrub hares (*Lepus saxatilis*) and crowned guinea fowl (*Numida meleagris*) on the farm "Bucklands" and on kudu, scrub hares and crowned guinea fowl on the adjoining Andries Vosloo Kudu Reserve in the Grahamstown district. A comparison of the tick burdens of the wild animals on the two properties indicated that acaricidal treatment of the domestic stock contributed to a marked reduction in the numbers of certain tick species on the wild animals on the farm.

MATERIALS AND METHODS

The farm "Bucklands" is 5 480 ha in extent and it shares a common 11 km boundary with the Andries Vosloo Kudu Reserve which is 6 497 ha in extent. The vegetation on the farm and reserve is classified as Valley Bushveld[1]. There are approximately 185 cattle, 300 Dorper sheep, 4 000 Angora goats and 300 kudu on the farm. The reserve contains approximately 54 hartebeest, 450 kudu, 140 eland and 100 buffalo. The numbers of scrub hares and guinea fowl on both properties are unknown.

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At monthly intervals, from February 1985 to January 1986, 2 beef-type calves, 2 Dorper sheep, 2 Angora goats and 2 scrub hares and, from May 1985, 2 crowned guinea fowl were slaughtered on the farm and processed for tick recovery by methods previously described[7 9]. Only 1 calf and not 2 was slaughtered during December 1985 and during January 1986. This was because the other 2 calves had died from unknown causes prior to slaughter at each occasion and only their carcasses had been found. At 3-monthly intervals 1 male kudu was shot for tick recovery on the farm, but during June 1985 2 kudu were shot. At monthly intervals 1 male kudu, 2 scrub hares and 2 crowned guinea fowl were shot in the Andries Vosloo Kudu Reserve and processed for tick recovery. It was, however, not always possible to obtain scrub hares or guinea fowl on either of the properties. The numbers of animals slaughtered each month are summarized in Table 1.

Although other cattle on the farm were treated at 2 to 4-weekly intervals with the acaricide fenvalerate (Sumitik Cattle Dip, Shell SA (Pty) Ltd), 4 yearling cattle were not treated, except when very heavily infested with ticks, and ran in camps separate from the other cattle. At monthly intervals 2 calves were sprayed with the acaricide fenvalerate or treated with the acaricide flumethrin (Drastic Deadline, Bayer SA (Pty) Ltd) and then kept under tick-free conditions for 7 days before being placed on the farm in the same camp as the 4 untreated cattle, where they ran for a month before being slaughtered and processed for tick recovery. This was done in order to determine the tick burdens of the survey calves under more natural conditions and to see whether these increased during the 2-year-period the survey is intended to run.

Sheep and goats on the farm were treated at monthly intervals with fenvalerate. The 2 sheep and 2 goats to be killed for survey purposes were not treated with acaricide for approximately 8-12 weeks before slaughter. The tick burdens of the sheep and goats were determined in order to see which ticks were still being acquired by the small stock on the farm.

RESULTS

The total numbers of ticks recovered from the cattle,

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Table 1. The total numbers of animals slaughtered each month from February 1985 to January 1986 on the farm "Bucklands" and in the Andries Vosloo Kudu Reserve

Month	Numbers of animals slaughtered each month								
	"Bucklands"						Andries Vosloo Kudu Reserve		
	Cattle	Dorper sheep	Angora goats	Kudu	Scrub hare	Guinea fowl	Kudu	Scrub hare	Guinea fowl
February, 1985	2	2	2		2		1*	2	
March	2	2	2	1	2		1	2	1*
April	2	2	2		2		1*	1	1*
May	2	2	2		2	2	1*	2	2
June	2	2	2	2	2	2	1	2	2
July	2	2	2		2	2*	1*	1	
August	2	2	2		2	2	1*	2	2
September	2	2	2	1	2	2	1	2	2
October	2	2	2		2	2	1*	2	2
November	2	2	2		2		1*	2	2*
December	1	2	2	1	1*		1		
January 1986	1	2	2		2	2	1*	2	2
Total numbers available for comparison	22	24	24	5	22	12	4	20	12

*The tick burdens of these animals were not used for comparative purposes because no animals of the same species had been shot on the other property during the same month

Table 2. The total numbers of ixodid ticks recovered from beef cattle, Dorper sheep and Angora goats on the farm "Bucklands"

Ixodid tick species	Total number of ticks recovered from								
	22 cattle			24 sheep			24 goats		
	Larvae	Nymphs	Adults	Larvae	Nymphs	Adults	Larvae	Nymphs	Adults
Amblyomma hebraeum	1258	166	477	30	40	0	64	30	0
Amblyomma marmoreum	876	13	0	42	4	0	158	12	0
Boophilus decoloratus	171	197	155	0	0	0	22	0	2
Haemaphysalis silacea	1328	960	432	618	250	24	576	125	23
Hyalomma marginatum rufipes	0	0	28	0	0	0	0	0	0
Rhipicephalus appendiculatus	836	322	320	270	56	0	703	55	26
Rhipicephalus evertsi evertsi	516	478	48	24	6	2	138	2	1
Rhipicephalus glabroscutatum	3047	775	742	1852	136	92	736	661	232
Rhipicephalus simus	0	0	17	0	0	0	0	0	0
Rhipicephalus (near *R.oculatus*)	0	0	17	0	0	2	0	0	2
Totals	8032	2911	2236	2836	492	120	2397	885	286

sheep and goats slaughtered during the course of the year are summarized in Table 2.

A total of 10 ixodid tick species were recovered from the cattle and 7 and 8 species from the sheep and goats respectively. *Rhipicephalus glabroscutatum* was the most abundant tick on each of the 3 hosts.

The cattle harboured considerably more ticks in each stage of development than did the sheep or goats. The difference was particularly noticeable for adult ticks. This is understandable as the cattle ran in a camp with untreated cattle while the sheep and goats ran with animals which were regularly treated.

The total numbers of ticks recovered from only those kudu, scrub hares and crowned guinea fowl that were shot during the same months on the farm and in the nature reserve are summarized in Tables 3-5.

The kudu were infested with 11 ixodid tick species. Those on the farm harboured considerably fewer *Amblyomma hebraeum* and slightly fewer *Haemaphysalis silacea* and *Rhipicephalus appendiculatus* than did the animals on the reserve.

The scrub hares were infested with 14 tick species. On the farm the hares carried markedly fewer *A. hebraeum* and slightly fewer *Hyalomma marginatum rufipes, R. appendiculatus* and *Rhipicephalus evertsi evertsi* than the animals in the reserve. Large numbers of *Rhipicephalus oculatus* were recovered from the scrub hares on both properties, but none were found on the domestic animals, kudu or guinea fowl. The large numbers of *R. oculatus* larvae recovered from the hares on the farm were particularly due to one animal which harboured 2 156 larvae.

Table 3. The total numbers of ixodid ticks recovered from kudu on the farm "Bucklands" and on the adjoining Andries Vosloo Kudu Reserve

Ixodid tick species	Total number of ticks recovered from					
	5 kudu on "Bucklands"			4 kudu on the Kudu Reserve		
	Larvae	Nymphs	Adults	Larvae	Nymphs	Adults
Amblyomma hebraeum	87	8	2	3756	1058	459
Amblyomma marmoreum	22	0	0	21	0	0
Boophilus decoloratus	4	0	16	0	0	2
Haemaphysalis silacea	441	224	259	2362	299	363
Hyalomma marginatum rufipes	0	0	2	0	0	0
Hyalomma truncatum	0	0	0	0	0	2
Ixodes pilosus	0	0	0	0	0	2
Rhipicephalus appendiculatus	20	40	50	408	45	104
Rhipicephalus evertsi evertsi	6	16	2	36	22	5
Rhipicephalus glabroscutatum	8632	5019	446	3583	1543	640
Rhipicephalus (near *R. oculatus*)	0	0	8	0	0	44
Totals	9182	5307	785	10166	2967	1621

Table 4. The total numbers of ixodid ticks recovered from scrub hares on the farm "Bucklands" and on the Andries Vosloo Kudu Reserve

Ixodid tick species	Total number of ticks recovered from					
	22 scrub hares on "Bucklands"			20 scrub hares on the Kudu Reserve		
	Larvae	Nymphs	Adults	Larvae	Nymphs	Adults
Amblyomma hebraeum	3	1	0	730	323	0
Amblyomma marmoreum	90	1	0	32	4	0
Boophilus sp.	7	0	0	0	0	0
Haemaphysalis leachi	0	1	0	0	0	0
Haemaphysalis silacea	17	10	2	16	20	0
Hyalomma marginatum rufipes	48	12	0	146	145	0
Hyalomma truncatum	1	2	0	0	0	0
Ixodes sp.	1	0	0	1	0	0
Rhipicephalus appendiculatus	44	62	0	112	233	0
Rhipicephalus evertsi evertsi	42	40	0	122	159	0
Rhipicephalus glabroscutatum	1127	353	0	740	260	0
Rhipicephalus oculatus	4605	770	134	604	641	109
Rhipicephalus (near *R.oculatus*)	0	0	1	0	0	0
Rhipicephalus sp.	5	3	0	22	2	0
Totals	5990	1255	137	2525	1787	109

Table 5. The total number of ixodid ticks recovered from crowned guinea fowl on the farm "Bucklands" and on the Andries Vosloo Kudu Reserve

Ixodid tick species	Total number of ticks recovered from					
	12 guinea fowl on "Bucklands"			12 guinea fowl on the Kudu Reserve		
	Larvae	Nymphs	Adults	Larvae	Nymphs	Adults
Amblyomma hebraeum	428	3	0	5295	275	0
Amblyomma marmoreum	6	7	0	70	5	0
Haemaphysalis silacea	529	61	0	648	169	0
Hyalomma marginatum rufipes	1	0	0	16	2	0
Ixodes sp.	1	0	0	0	0	0
Rhipicephalus appendiculatus	0	0	0	5	0	1
Rhipicephalus evertsi evertsi	0	0	0	1	0	0
Rhipicephalus glabroscutatum	1	0	0	10	2	0
Rhipicephalus sp.	0	0	0	1	0	0
Totals	966	71	0	6046	453	1

The guinea fowl were infested with 9 tick species. Those on the Kudu Reserve harboured considerably more *A. hebraeum* than did the birds on the farm.

DISCUSSION

In terms of numbers the major tick species on the farm "Bucklands" and in the Kudu Reserve are *A. hebraeum, H. silacea, R. appendiculatus* and *R. glabroscutatum* as well as *R. oculatus;* the latter tick only infesting the scrub hares. These findings support those of earlier surveys on the same properties[11 19].

The immature stages of *A. hebraeum* will feed on a large variety of hosts[9 15 20], while the adults prefer cattle and the very large antelope and wild bovid species[6 7 10 18]. MacIvor & Horak[12] have recovered fairly large numbers of adult *A. hebraeum* from Angora and Boer goats, but contrary to the findings in the Kudu Reserve in the present survey, kudu generally do not carry large numbers of adults of this tick[11]. The regular acaricidal treatment of the domestic stock on the farm thus affected particularly *A. hebraeum* and resulted in a marked reduction in numbers. This in turn is reflected in the small burdens of *A. hebraeum* recovered from the Dorper sheep and Angora goats and from the wild animals slaughtered on the farm. The cattle examined in the survey had fairly large burdens of adult ticks but it must be remembered that they ran in a separate camp with 4 cattle that were hardly ever treated.

It could be argued that the differences in the *A. hebraeum* burdens of the wild animals on the 2 properties were due to factors other than acaricidal treatment of the domestic stock on the farm. The Kudu Reserve harboured a total of 240 eland and buffalo, and as these animals are excellent hosts of adult *A. hebraeum*[7 18] the numbers of this tick could have increased considerably on the reserve. This would account for the large burdens on the kudu, scrub hare and guinea fowl shot on the reserve. The farm, however, was stocked with 185 cattle. These animals are also excellent hosts of adult *A. hebraeum*[6] and the numbers of this tick would also no doubt have increased on the farm but for the regular applications of acaricide.

It is possible that the large number of Angora goats on the farm, unmatched by numbers of similarly-sized antelope in the Kudu Reserve, altered the habitat so that it was no longer suitable for *A. hebraeum*, or they could have diluted the tick numbers to such an extent that there were fewer to infest the wild animals. To discount these possibilities it must be remembered that Angora goats are also fairly good hosts of all stages of development of *A. hebraeum*[12], and instead of diluting the population, would probably have assisted in increasing it if it had not been for the acaricidal treatment applied to the goats. If the large number of goats had actually altered the habitat to make it unsuitable for *A. hebraeum* one would have expected the other tick species to be similarly affected.

In a way the domestic stock on the farm acted like "vacuum cleaners" collecting the ticks and bringing them to the dipping tank or spray race to be killed. Because Angora goats[12] and cattle[6] are generally better hosts of adult *A. hebraeum* than kudu[11] the majority of adult ticks on the farm were killed and consequently the population on the wild animals was reduced.

Adult *Amblyomma marmoreum* are found almost exclusively on tortoises and other reptiles[4 17 20], while the immature stages will infest tortoises and several other hosts[9 17]. As could therefore be expected the numbers of immature *A. marmoreum* found on all host species at both localities in the present study indicate that the adults were unaffected by the acaricidal treatment of the domestic stock.

Adult *H. silacea* prefer herbivores such as bushbuck, kudu and eland[7 11 16] while the immatures may be found on a variety of animals[16 20]. Those adults and immatures present on the kudu would be unaffected by acaricidal treatment applied to the domestic stock. Consequently infestation would continue and this is reflected by the small reduction in numbers on the wild animals running on the farm.

The adults of the *Hyalomma* spp. prefer cattle and the larger wild herbivores (Table 2)[3 6 7 10]. The immature stages prefer scrub hares, certain rodents and some ground-frequenting birds (Tables 4, 5)[2 3 10 15]. The acaricidal treatment of the cattle could thus be expected to reduce the number of immature *Hyalomma* spp. on the farm, as was found in this survey.

Both kudu and cattle are good hosts of immature and adult *R. appendiculatus*[6 7 10 11]. Acaricidal treatment of the cattle would consequently only control a portion of the population, while the remainder on the kudu continue to reinfest the pastures. The moderate reduction in *R. appendiculatus* numbers on the wild animals on the farm is a reflection of this fact.

Amongst the animals we have examined, kudu and goats appear to be preferred hosts of all stages of development of *R. glabroscutatum*[7 12]. Acaricidal treatment of the domestic livestock would consequently only partially reduce the total population. In addition the vast majority of both immature and adult ticks of this species are found on the lower legs and around the feet of domestic and wild ruminants[7 13]. The constant contact of the feet and lower legs with soil, grass and dew is likely to have an adverse effect on acaricidal efficacy against these ticks. It is thus not surprising that no reduction in numbers is evident, and kudu and scrub hares on the farm actually harboured greater burdens of this tick than those in the reserve.

R. oculatus is a tick which, in all its stages of development, seems to prefer hares as hosts (Table 4)[20]. Hence acaricidal treatment of domestic stock will have no effect on the numbers of this tick.

In conclusion it can be stated that the greater the preference of adult ticks of a particular species for domestic stock such as cattle, sheep and goats, the greater the possibility of controlling that species by adequate acaricidal treatment of the domestic stock irrespective of the number of wild animals present on the same farm. If, however, several really large wild bovids such as eland or buffalo are present on the farm, tick control will be considerably more difficult as these animals are good hosts of the adults of a large number of tick species[7 10 14 20], and these ticks will consequently be less affected by acaricidal treatment of the domestic stock.

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REFERENCES

1. Acocks J P H 1975 Veld types of South Africa with accompanying veld type map. Memoirs of the Botanical Survey of South Africa No 40, 128 pp
2. Clifford C M, Flux J E C, Hoogstraal H 1976 Seasonal and regional abundance of ticks (Ixodidae) on hares (Leporidae) in Kenya. Journal of Medical Entomology 13: 40-47
3. Hoogstraal H 1956 African Ixodoidea. I. Ticks of the Sudan (with special reference to Equatoria Province and with preliminary reviews of the genera *Boophilus, Margaropus,* and *Hyalomma*). Department of the Navy Bureau of Medicine and Surgery, Washington DC 1101 pp.
4. Hoogstraal H, Aeschlimann A 1982 Tick – host specificity. Bulletin de la Société Entomologique Suisse 55: 5-32
5. Horak I G 1980 The control of parasites in antelope in small game reserves. Journal of the South Arican Veterinary Association 51: 17-19
6. Horak I G 1982 Parasites of domestic and wild animals in South Africa. XV. The seasonal prevalence of ectoparasites on impala and cattle in the northern Transvaal. Onderstepoort Journal of Veterinary Research 49: 85-93
7. Horak I G, Potgieter F T, Walker Jane B, De Vos V, Boomker J 1983 The ixodid tick burdens of various large ruminant species in South African nature reserves. Onderstepoort Journal of Veterinary Research 50: 221-228
8. Horak I G 1984 Cattle, wild animals and parasites – epizootic or delicately balanced ecosystem? Proceedings 13th World Congress on Diseases of Cattle, Durban, September 17-21 1984: 390-393
9. Horak I G, Williams E J 1986 Parasites of domestic and wild animals in South Africa. XVIII. The crowned guinea fowl *Numida meleagris,* an important host of immature ixodid ticks. Onderstepoort Journal of Veterinary Research 53: 119-122
10. Howell C J, Walker Jane B, Nevill E M 1978 Ticks, mites and insects infesting domestic animals in South Africa. Part 1. Descriptions and biology. Department of Agricultural Technical Services, Republic of South Africa, Science bulletin No 393, v + 69 pp
11. Knight M M, Rechav Y 1978 Ticks associated with kudu in the eastern Cape Province: Preliminary report. Journal of the South African Veterinary Association 49: 343-344
12. MacIvor K M, Horak I G 1984 The internal and external parasites of Angora and Boer goats in Valley Bushveld near Uitenhage. Angora Goat and Mohair Journal 26: 7-14
13. MacIvor K M 1985 The distribution and hosts of *Rhipicephalus glabroscutatum.* Onderstepoort Journal of Veterinary Research 52: 43-46
14. MacLeod J 1970 Tick infestation patterns in the southern province of Zambia. Bulletin of Entomological Research 60: 253-274
15. Norval R A I 1974 Studies on the biology and ecology of *Amblyomma hebraeum* Koch, 1844 and other tick species (Ixodidae) of the eastern Cape. PhD thesis: Rhodes University
16. Norval R A I 1975 Studies on the ecology of *Haemaphysalis silacea* Robinson 1912 (Acarina: Ixodidae). Journal of Parasitology 61: 730-736
17. Norval R A I 1975 Studies on the ecology of *Amblyomma marmoreum* Koch 1844 (Acarina: Ixodidae). Journal of Parasitology 61: 737-742
18. Norval R A I 1983 The ticks of Zimbabwe. VII. The genus *Amblyomma.* Zimbabwe Veterinary Journal 14: 3-18
19. Rechav Y 1982 Dynamics of tick populations (Acari: Ixodidae) in the eastern Cape Province of South Africa. Journal of Medical Entomology 19: 679-700
20. Theiler Gertrud 1962 The Ixodoidea parasites of vertebrates in Africa south of the Sahara (Ethiopian Region). Report to the Director of Veterinary Services, Onderstepoort 260 pp.

ABSTRACT

SAMEVATTING

SEASONAL ABUNDANCE AND DISTRIBUTION OF *PARAFILARIA BOVICOLA* OVIPOSITIONAL BLOOD SPOTS ON CATTLE IN SOUTH AFRICA

More than 23 000 cattle of both sexes and different ages were examined for blood spots caused by egg-laying females of *P. bovicola.* Although these studies extended over four years and involved 5 farms in different parts of the Transvaal Bushveld, the overall results were the same.

Ovipositional bleeding was strongly seasonal with blood spots first appearing in winter (June), reaching a peak in spring (September-November) and thereafter declining rapidly as summer progressed. In a single year at Zoutpan Research Station up to 92,1 % of the 1st year heifers had already bled by November and this proportion increased only slightly to 95,1 % by the end of the bleeding season (May). The number of blood spots per animal showed a similar seasonal abundance except for a second peak of abundance in June for 1st year heifers and oxen.

The prevalence of blood spots in cattle of different ages and sex varied markedly. At Mara Research Station half as many oxen bled in their 2nd year as in their 1st year, while at Zoutpan 19,2% fewer heifers bled in their 2nd year than in their 1st. Bulls bled the most, then 1st year oxen, 1st and 2nd year heifers and 2nd year oxen, with breeding cows bleeding the least. A high female hormone level appears to be associated with the development of immunity.

The shortest period from birth to 1st blood spot (the apparent prepatent period) was 191 days, while 81,8 % of oxen bled for the 1st time within 279 days after birth. Blood spots were equally distributed on the left and right sides, with 92,1 % on the dorsolateral regions and 59,9 % on the shoulder and rib regions. The blood spot distribution more or less matched the carcass lesion distribution. This suggests that ovipositing females are largely responsible for carcass lesions in these areas. (Neville, E.M., 1984. The seasonal abundance and distribution of *Parafilaria bovicola* ovipositional blood spots on cattle in South Africa. *Onderstepoort Journal of Veterinary Research,* 51, 107-114 (1984).)

ARTICLE ARTIKEL

DIE SKADELIKE GEVOLG VAŃ INTRA-UTERINE TOEDIENING VAN JODIUM BY MERRIES

ENETTE VAN DYK* EN A. LUCIA LANGE**

ABSTRACT: Van Dyk Enette; Lange A Lucia. **The detrimental effect of the use of iodine as an intra-uterine instillation in mares.** *Journal of the South African Veterinary Association* (1986) **57** No. 4, 205-210 (Afrik) Department of Genesiology, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, 0110 Onderstepoort, Republic of South Africa.

Fifty ml of a 0.2% iodine solution were instilled into the uteri of six mares selected for culling. Biopsy specimens were taken for microscopic examination before the commencement of the experiment and at various times over the course of a year after the single instillation. Severe oedema and haemorrhage was present in the lamina propria of all the post-instillation biopsy specimens. The epithelium showed vacuolisation and necrosis, as well as focal areas of epithelial loss. In some cases hyperplasia or metaplasia was seen. Cell infiltration was characterized by the presence of neutrophils, eosinophils and mild lymphocyte infiltration around the blood vessels. Leukostasis was present around the blood vessels in some specimens. Blood vessel changes were characterized in the early stages by oedema of the *tunica media* of the medium-sized arteries and later by arteriosclerosis with the complete obliteration of the lumen of the particular vessels in some cases. The endometrial glands showed hyperplasia and mitotic figures were more often seen in these glands than normal. The most pronounced lesion in the later stage of the experiment was fibrosis of the lamina propria with the resultant collapse of the *stratum compactum* and contraction of the *stratum spongiosum*. The severe fibrosis of the endometrium is the most probable reason for infertility in mares treated with iodine.

Keywords: Iodine, uterine instillation, mares, endometrial fibrosis.

INLEIDING

Die gebruik van jodium as 'n chemiese kuretasie in die baarmoeder van koeie is 'n welbekende praktyk waardeur regenerasie van die endometrium gestimuleer word en suksesvolle teling in bepaalde gevalle daarna moontlik is[1,2,8,11]. Die irritasie van die endometrium wat deur die jodium veroorsaak word stimuleer die produksie en vrystelling van die luteolitiese faktor in die baarmoeder wat dan regressie van die corpus luteum veroorsaak en die aanvang van estrus tot gevolg het[7].

Die effek wat jodium as intra-uterine installasie in die merrie het, is deur Mather & Hurtgen[5] getoets. Hierdie merries het egter nie estrus getoon nie. Histologiese ondersoeke van die endometrium het ernstige interstisiële inflammasie aangetoon wat tot by die miometrium gestrek het. Fokale areas van nekrose, bloeding en interstisiële fibrose is tot 4 weke na behandeling waargeneem.

In 'n eksperiment om die gebruik van vesel-optiese tegnieke te evalueer is merries eksperimenteel met verskillende middels behandel. Hier het die jodium installasie ernstige inflammasie, fibrien neerlegging en ulserasie van die endometrium veroorsaak[6].

Die installasie van jodium intra-uterien in merries word egter wel in praktyk gedoen, moontlik met die aanname dat dieselfde reaksie soos wat in die bees verkry word, ook hier verkry sal word.

Sommige merries met erge bakteriese besmetting van die baarmoeder word suksesvol behandel met antibiotika volgens die antibiogram aangedui, soos blyk uit die onvermoë om enige bakterië te kweek en die suksesvolle teling van die merries na behandeling[13]. Ander merries onder soortgelyke omstandighede kan egter nie beset raak nie. In hierdie merries se geskiedenis kon altyd 'n intra-uterine behandeling, met jodium, op 'n vroëer stadium, òf vir ontsmettingsdoeleindes òf vir die regulering van die estrussiklus, opgespoor word[13].

Vrugbaarheid in die merrie word gedefinieer as die vermoë om beset te raak, die konseptus te behou en aan 'n lewende vul geboorte te gee[3]. Onvrugbaarheid is dus 'n onvermoë om aan enige van die vereistes te voldoen en om die moontlike rede vir die onvrugbaarheid by merries wat met jodium behandel is te probeer vasstel, is opeenvolgende mikroskopiese ondersoeke van die baarmoeder gedoen na 'n enkele intrauterine behandeling met jodium.

MATERIAAL EN METODIEK

Ses uitskot merries van verskillende ras en ouderdom, waarvan die teelgeskiedenis bekend is, is gebruik. Die geslagstelsel is in alle gevalle deeglik ondersoek deur beide rektale palpasie en vaginale inspeksie voor die aanvang van die eksperiment. Monsters vir histologiese ondersoek van die baarmoeder is geneem met 'n Hauptner instrument volgens die metode beskryf deur Rickets[9].

Weefselmonsters vir histologie van die baarmoeders is vooraf geneem en weer na 3, 6, 12, 27 en 120 uur; 1, 2, 3 en 4 weke; 6 maande en 1 jaar na 'n enkele installasie van jodium.

Die weefselmonsters is fikseer in Bouin se oplossing en na prosessering is snitte gekleur met hematoksilien en eosien (HE) en Mason se trichroom kleurtegniek[12] vir ligmikroskopiese ondersoek.

Geen kwantifisering van histologiese veranderinge is gedoen nie. Die veranderde histologiese beeld is met die kontrole vergelyk en daarvolgens geïnterpreteer. Daar is geen skaal gebruik om die selinfiltrasie te beskryf nie. Die veranderinge is met die kontrole vergelyk.

Povidoon jodium (Betadine, Keatings) is as 'n enkel intra-uterine installasie gebruik. Dit is verdun na 'n 0,2% jodium oplossing waarvan 15 ml per merrie gebruik is.

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RESULTATE

Volgens die beskrywing van Rossdale & Ricketts[10] van die normale anatomie en histologie van die vroulike geslagstelsel van die merrie, het die kliniese beeld, in vyf van die merries tydens vooraf ondersoek geen afwykings getoon nie en was die histologiese beeld in al vyf merries normaal. In Merrie 3 was 'n pneumovagina teenwoordig en die uterus was deegagtig met palpasie. Die eierstokke was aktief en geen ander abnormaliteite het voorgekom nie. Histologiese ondersoek het 'n ligte periglandulêre fibrose getoon.

In die snitte wat voor die toediening van jodium van die baarmoeder biopsies gemaak was, het die voorkoms van die baarmoederepiteel gewissel na gelang van die estrussiklus van die betrokke merrie. Ligte bloeding was in al die snitte teenwoordig. Haemosiderien pigment is in drie merries waargeneem en ligte periglandulêre fibrose was in Merrie 3 teenwoordig.

Edeem en bloeding in die *stratum compactum* en *stratum spongiosum* was deurgaans in al die biopsies teenwoordig na installasie (Fig. 1.) Gedurende die eerste paar ure na installasie van die jodium was die bloeding redelik uitgesproke net onder die epiteel. In latere biopsies was bloeding multifokaal in beide bogenoemde lae; soms was dit om die bloedvaatjies. Edeem was deurgaans teenwoordig in beide lae maar veral in die *stratum spongiosum*. Selfs biopsies wat 1 jaar na die enkele installasie van jodium geneem was, het duidelike edeem van hierdie laag getoon (Fig. 2.)

Drie uur na die toediening van jodium in utero was vakuolisasie van die proksimale gedeeltes van die oppervlakkige epiteelselle uitgesproke in 5 van die 6 gevalle. Epiteelselle wat kubies was voor die toediening het 3 uur daarna silindries vertoon met opvallende vakuolisasie. Diè wat vooraf silindries was se opvallendste veranderinge was vakuolisasie van die proksimale gedeeltes asook kariopiknose. Daar was in sommige gevalle bloeding tussen die epiteelselle. Nekrose van die oppervlakkige epiteel was waargeneem in 4 van die 6 merries.

Eosinofielinfiltrasie was in een merrie so vroeg as 3 uur na die toediening van jodium waargeneem. Die eosinofielinfiltrasie was in die lamina propria.

Ses uur na installasie van jodium in utero was gevind dat behalwe bloeding en edeem, wat weereens opvallend was, die epiteel by Merrie 2 en 6, wat silindries was voor die aanvang van die eksperiment nou fokale areas van epiteelverlies gewys het (Fig. 3.) Die oorblywende epiteelselle het ook meer kubies vertoon. Op hierdie stadium is neutrofielinfiltrasie vir die eerste keer in beide merries waargeneem en was eosinofiel infiltrasie ook teenwoordig.

Twaalf uur na installasie van jodium het neutrofiel infiltrasie in 4 uit die 6 merries voorgekom. Eosinofiel infiltrasie het in 3 merries voorgekom (Fig. 4) Nekrose van fokale dele van die epiteel het in 5 gevalle voorgekom en edeem van die *tunica media* in 3 gevalle.

Op hierdie stadium het leukostase in veral die venas van die *stratum spongiosum* voorgekom. Soos reeds

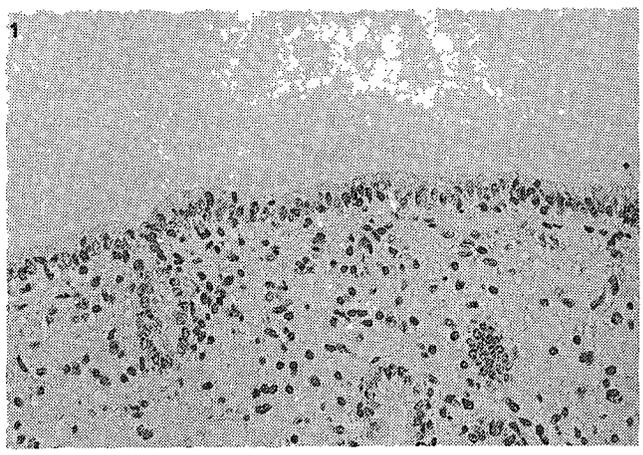

Fig. 1: Bloeding en edeem. Merrie 1; 3 uur na installasie van jodium. HE X1000

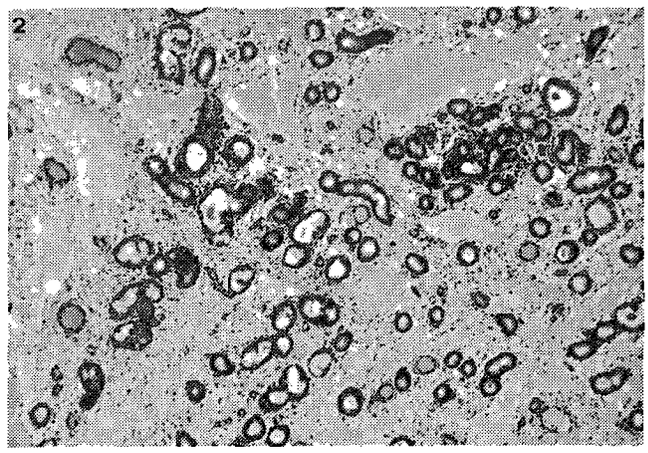

Fig. 2: Edeem en fibrose teenwoordig 1 jaar na die installasie van jodium in Merrie 4. HE X250

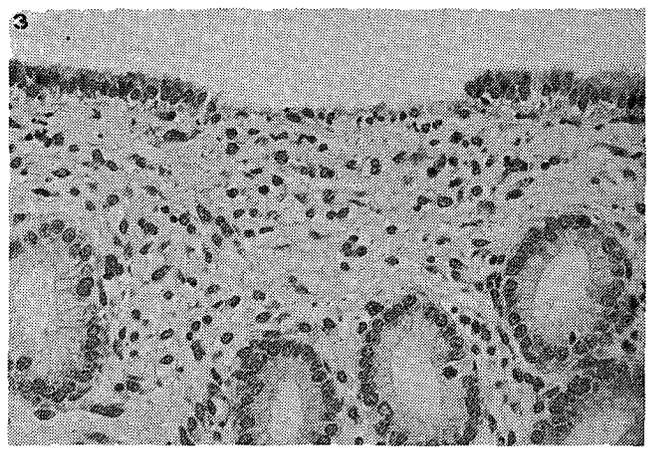

Fig. 3: Erosie met neutrofiele in submukosa. Mitotiese figure kan in die kliérepiteel gesien word. Merrie 1; 27 uur na installasie van jodium. HE X1000

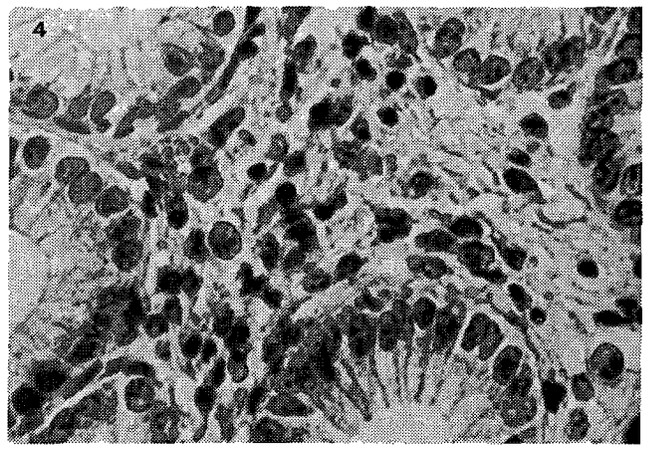

Fig. 4: Eosinofiele in Merrie 1; 12 uur na die installasie van jodium. HE X2500

genoem was bloeding en edeem ook konstante bevindinge gedurende dié periode.

Die biopsies wat 27 uur na die installasie van jodium geneem was, het neutrofiel infiltrasie in al ses merries getoon asook nekrose van die epiteel, eosinofiel infiltrasie, bloeding en edeem, en edeem van die *tunica media* van die middelslag arteries. Vakuolisasie van die oppervlakkige epiteel het in 3 gevalle voorgekom en hiperplasie van kliere in 1 merrie.

Vyf dae later was dieselfde veranderinge steeds teenwoordig asook die vermeerdering in die aantal epiteelselle in beide die mukosa, waar die selle kubies en meerlagig vertoon het in sekere areas, en in die baarmoederkliere. Duidelike mitotiese figure is in die kliere waargeneem (Fig. 3.) asook 'n vermeerdering in die aantal kliere in die *stratum spongiosum*. (Fig. 5 & 6).

Die veranderinge 7 en 14 dae na installering van jodium was dieselfde soos hierbo beskryf vir 5 dae.

Drie weke na die installering van jodium kon bykomend in 3 merries waargeneem word dat die edeem in veral die *stratum compactum* tekens van organisasie begin toon het; daar was naamlik 'n vermeerdering in die onvolwasse fibroblaste in dié gebied. Hiperplasie van kliere is in 4 merries waargeneem.

'n Opvallende letsel wat gesien is 28 dae na die aanvang van die eksperiment, was fibrose van die bloedvatwande wat voorheen edeemagtig vertoon het. In alle gevalle was dit die middelslag en groter arteries wat geaffekteer was en in sommige snitte was die fibrose so erg dat dit gelyk het asof die lumen afgesluit was as gevolg van bindweefsel neerlegging. Die ander veranderinge was nog soos hierbo beskryf.

Ses maande na die aanvang van die eksperiment kon bykomstig tot die uitgesproke bloeding en edeem in die *stratum compactum* en *stratum spongiosum* fibrien thrombi in verskeie klein bloedvaatjies waargeneem word. Laasgenoemde is in 4 van die 6 merries gesien. Terselfdertyd is gevind dat kollageen neergelê is en dat fibroblaste meer volwasse geraak het met uiteindelike periglandulêre fibrose in 3 merries. Hiperplasie van die kliere is in 5 merries gesien terwyl die oppervlakkige epiteel in 4 gevalle metaplasties vertoon het (Fig. 7).

Een jaar na die installasie van die jodium in utero is waargeneem dat die epiteel silindries vertoon het in al die merries. Sklerose van die bloedvatwande was steeds teenwoordig in al die merries. In alle gevalle was dit die middelslag en groter arteries wat geaffekteer was en in sommige snitte was die fibrose so erg dat dit gelyk het of die lumen afgesluit was as gevolg van die bindweefsel neerlegging (Fig. 8).

Hiperplasie van kliere is in 5 merries waargeneem terwyl trombi ook opgemerk is by 3 van die 6 merries in verskeie klein bloedvaatjies (Fig. 9.)

Die mees uitgesproke letsels in die biopsies wat na 1 jaar geneem is en deurgaans by al die merries gevind is was erge fibrose van die lamina propria. Die fibrose was uitgesproke met gevolglike kollaps van die *stratum compactum* wat gelei het tot 'n indruk dat die hele baarmoederwand saamgetrek het (Fig. 10 & 11.)

Die histologiese veranderinge word ook opsommen-

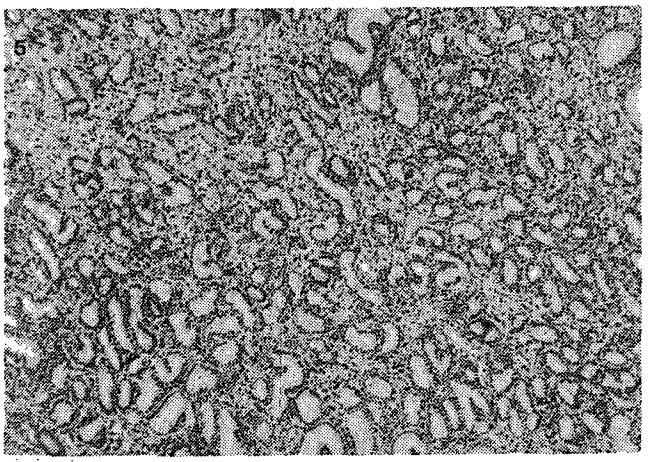

Fig. 5: Hiperplasie en fibrose in Merrie 5; 1 jaar na die installasie van jodium. HE X250

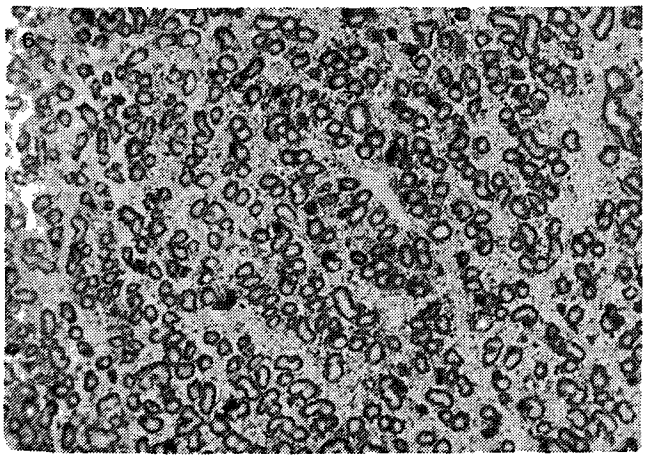

Fig. 6: Hiperplasie, bloeding en edeem in Merrie 5; 6 maande na die installasie van jodium HE X500.

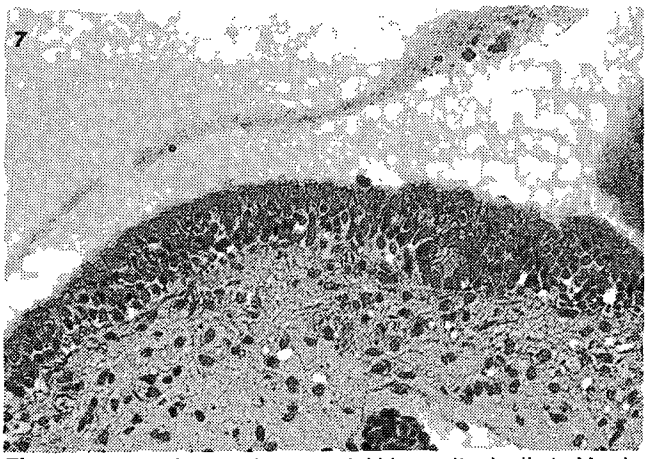

Fig. 7: Metaplasie van die oppervlakkige epiteelselle in Merrie 5; 6 maande na die installasie van jodium. HE X1000.

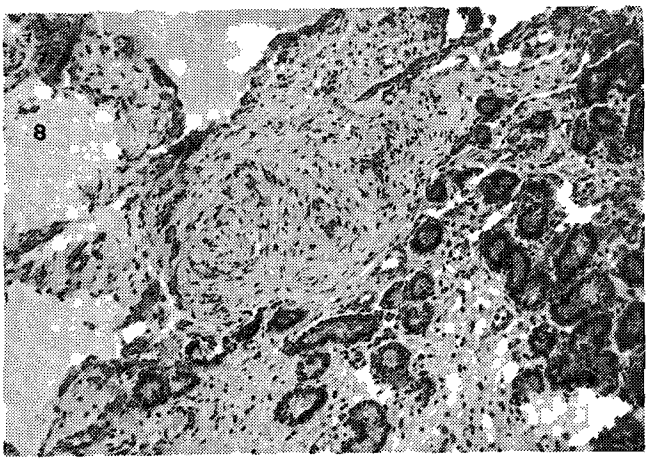

Fig. 8: Arteriosklerose. Merrie 5; 6 maande na die installasie van jodium. HE X500

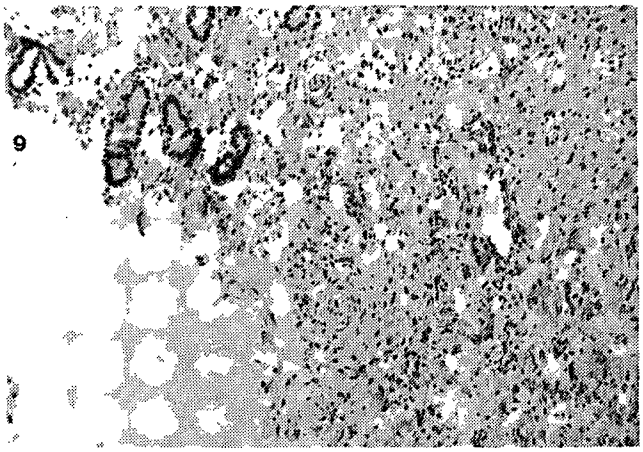

Fig. 9: Trombi en arteritis teenwoordig in Merrie 1; 6 maande na die installasie van jodium. HE X500.

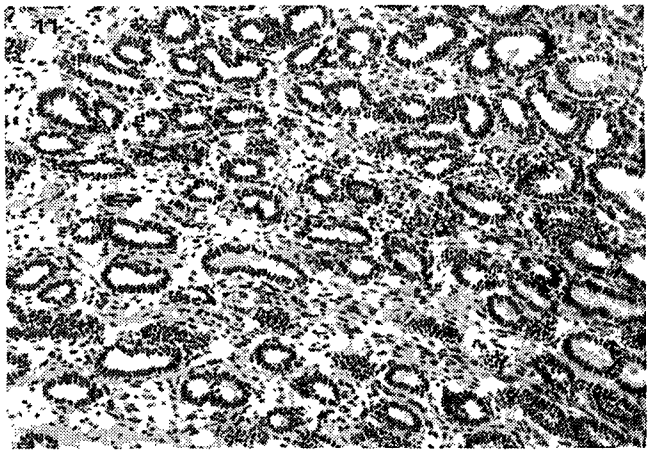

Fig. 11: Fibrose teenwoordig in Merrie 1; 1 jaar na die installasie van jodium. HE X500

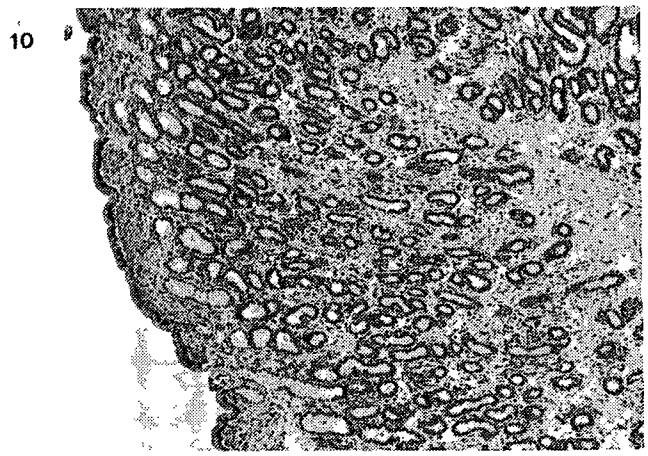

Fig. 10: Fibrose teenwoordig 1 jaar na die installasie van jodium in Merrie 1. HE X250

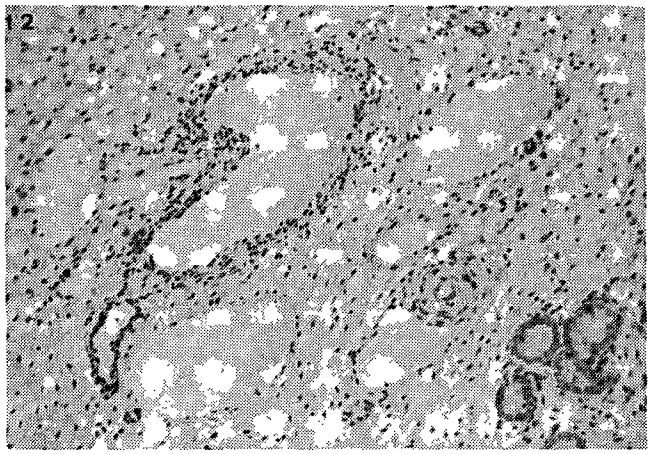

Fig. 12: Perivaskulêre infiltrasie van 'n vena en edeem van die *tunica media* van die arterie. Merrie 1; 7 dae na die installasie van jodium. HE X500

derwys in Tabel 1 weergegee.

Op geen tydstip gedurende die eksperiment was daar 'n erge selinfiltrasie teenwoordig in enige van die biopsies nie. Daar was neutrofiele binne 6 uur na die installering van jodium en variërende hoeveelhede van hierdie seltipe was nog teenwoordig in biopsies geneem 28 dae na die aanvang van die eksperiment. Eosinofiele was opgemerk by al die merries. In sommige merries was eosinofiele binne 3 uur na die begin van die eksperiment gesien en binne 12-27 uur het al die biopsies eosinofiel infiltrasie getoon. Eosinofiele was nog teenwoordig 8 dae na die jodium installering by Merrie 3. Leukostase en perivaskulêre flensing met beide neutrofiele en limfosiete was teenwoordig in sommige snitte maar dit was nie konstant by al die merries nie (Fig. 12).

Die baarmoederkliere was ook deurgaans geaffekteer. Binne 5 dae na die toediening van jodium kon hiperplasie van die klierepiteel waargeneem word. In hierdie stadium sowel as in opeenvolgende biopsies kon mitotiese figure in die epiteelselle en 'n vermeerdering in die aantal kliere waargeneem word.

Opsommenderwyse kan die belangrikste veranderinge as volg saamgevat word:

1. Edeem en bloeding was deurgaans in al die biopsies teenwoordig.
2. Die epiteel het eerstens vakuolisasie en nekrose gewys, daar was fokale areas van epiteel verlies, herstel het plaasgevind en die hele siklus verandering het weer herhaaldelik voorgekom. In sommige gevalle het die selle hiperplasie of metaplasie gewys.
3. Selinfiltrasie was gekenmerk deur die teenwoordigheid van neutrofiele, eosinofiele en geringe limfosiet infiltrasie gewoonlik om bloedvaatjies. Soms was daar leukostase ook in bloedvate teenwoordig.
4. Bloedvatveranderinge was in die vroeë stadia gekenmerk deur edeem van die *tunica media* van middelslag arteries wat later verwerk is tot arteriosklerose soms met algehele afsluiting van die lumen van die betrokke vate.
5. Die endometriumkliere het hiperplasie getoon en mitotiese figure is meer dikwels in dié kliere opgelet as wat normaalweg die geval sou wees.
6. Die mees uitgesproke letsel in die latere stadia van die eksperiment was fibrose van die lamina propria met gevolglike kollaps van die *stratum compactum* en sametrekking van die *stratum spongiosum*.

BESPREKING

Alhoewel 'n baarmoeder biopsie slegs 0,2% van die oppervlakte weergee is dit tog verteenwoordigend van wat verwag kan word in die res van die baarmoeder[4].

Gedurende die proses van die neem van biopsies vind geringe beskadiging van die weefsel plaas met gevolglike bloeding. Geen kwantifisering is gedoen nie maar die bloeding wat deurgaans in alle biopsies gevind is, was

Tabel 1: Histologiese endometriale veranderinge na jodium installasie

Merrie no.	Vooraf	3 uur	6 uur	12 uur	27 uur	5 dae	7 dae	14 dae	21 dae	28 dae	6 maande	1 jaar
1	Ek Hs B	B E Ve N Eos Hs Es	N/O	B E N Neu	B E Ve Neu Eos A	B E N Neu Eos A H Hs	B E N Neu Eos A H	B E N Neu Eos A H	B E Ve Neu A H	B E Hs Eos Neu H As	B E Es T H As	B E F Es H As
2	Es B	B E Ve N	B E Ek Neu Neu	E N (erg) Eos	B E Ve N A	B E N Neu Neu Eos H	B E N Neu Eos H A	B E N H H H	B E Neu Neu F (vroeg)	B E N F As	B E Ek N H As	B E F Fs As
3	Es B F (matig)	B E	N/O	B E Ve Neu Eos A	B E N Neu Eos A	B E N Neu Eos H	B E N Neu Eos H A	B E N Neu Eos H A	B E	B E Ve Eos A M	B E M H F (erg)	B E F T As Es H
4	Ek B	B E N E_s	N/O	B E N Neu	B E N Neu Eos A	B E N Neu H A	B E Ve	B E Neu A	B E F (vroeg)	B E Ve N As	B E M H T As	B E T As F Es H As
5	Ek Hs B	B E Ve N F_s	N/O	B E N Eos Hs A	B E N Ve Neu Eos	B E N Neu Eos H A	B E N Ve Neu Eos A	B E Ve Neu Eos A H	B E Neu A	B E N Ve Neu As	B E H M As T	B E F Es T H As
6	Es Hs B	B E Ve N	B E Ek N Neu Eos	B E N Eos A	B E N Neu A H	B E N Ve Neu Eos	B E N H A Neu	B E Neu H	B E H F (vroeg)	B E N Neu H F As	B E HF As Es T	B E F Es H As

A = edeem van *tunica media*
As = arteriosklerose
B = bloeding
E = edeem
E_k = epiteel kubies
Eos = eosinofiele
E_s = epiteel silindries
F = fibrose
H = hiperplasie van kliere
H_s = hemosiderien
M = metaplasie
N = nekrose van epiteel
Neu = neutrofiele
N/O = nie ondersoek nie
T = fibrien trombi
Ve = Vakuolisasie epiteel

meer uitgesproke as by die biopsies wat geneem is voor die aanvang van die eksperiment. Ons is dus van mening dat die bloeding wat gesien is in die biopsies nie net was as gevolg van die tegniek nie, maar deel uitmaak van die letsels wat met jodium installering geassosieer word.

Edeem van die lamina propria was in meeste van die biopsies teenwoordig. Uitgesproke edeem van die *stratum compactum* en *stratum spongiosum* is gevind tot ongeveer 21-28 dae na die aanvang van die eksperiment; hierna was daar gaandeweg organisasie van die losserige struktuur tot meer neerlegging van kollageen met uiteindelike fibrose en sametrekking van die baarmoederwand. Veral die periglandulêre fibrose was baie opvallend in sommige gevalle. Alhoewel die uiteinde van die erge edeem fibrose was (6 maande en 'n jaar), was daar nog steeds 'n mate van edeem teenwoordig in laasgenoemde biopsies. Die vermoede bestaan dat die jodium wat slegs eenmalig geïnstalleer is, óf 'n depot-effek het en dus 'n baie langdurige irriterende invloed op die baarmoeder wand uitoefen óf dat dit onmoontlik is vir die liggaam om die jodium af te breek met dieselfde gevolg as hierbo genoem. Dit moet egter nog bepaal word waar die jodium gestoor word asook die konsentrasies daarvan.

Dat daar 'n irriterende effek op die baarmoederwand uitgeoefen word blyk ook duidelik uit die veranderinge wat in die epiteel waargeneem is. Die vakuolisasie wat voorkom in die luminale epiteelselle gedurende die geslagsiklus is gewoonlik beperk tot die basale derde van die epiteelselle[3]. In hierdie eksperimentele gevalle was daar konstant vakuolisasie in die proksimale gedeelte van die epiteelselle. Daar word beweer dat hierdie vakuolisasie patologies is indien dit saamgaan met transepiteliale migrasie van neutrofiele[3]. In die biopsies wat ons ondersoek het, het ons gevind dat neutrofiele deur die epiteel migreer het en dat daar elders ook neutrofiele teenwoordig was. Ander veranderinge wat in die epiteel waargeneem was, was nekrose van die epiteelselle met afstoting en erosievorming. Hierdie veranderinge was meer dikwels in die vroeë stadia van die eksperiment gesien, maar selfs in biopsies wat na 6 maande en 'n jaar geneem is, was daar kariopiknose in die epiteelselle.

Nog 'n verandering wat waargeneem was, was dat in sekere dele van biopsies geneem na 6 maande daar 'n neiging vir die epiteel was om meerlagig en plat te word óf dat daar hiperplasie van die meer kubiese epiteel was. Dit is moontlik dat die epiteelveranderinge wat hierbo beskryf is net die normale sikliese variasie by merries weerspieël maar indien die ander veranderinge van bloeding en edeem bygereken word, is dit heelwaarskynlik dat hierdie letsels saamhang met 'n irriterende effek deur die jodium.

Endometriale hiperplasie word gesien in gevalle van

lankstaande endometritis. Rossdale & Ricketts[10] meen dat dit die gevolg van herhaaldelike estrusstimmulasie kan wees. Verder word diffuse fibrose dikwels in sulke gevalle gevind.

Selinfiltrasies wat in die biopsies waargeneem was, was eosinofiele, neutrofiele en limfosiete. Die rede vir die eosinofiel infiltrasie is onbekend, alhoewel Kennedy[3] bevind het dat eosinofiele dikwels teenwoordig is in merries wat aan endometritis lei tydens estrus.

Die middelslag arteries het binne 12 uur reageer met edeem van die *tunica media*. Na 28 dae het hierdie letsel na arteriosklerose van die bloedvate verander. In sommige gevalle was die letsel so erg dat daar afsluiting van die lumen plaasgevind het. Hierdie letsel was by al die biopsies gesien wat na 6 maande en 'n jaar geneem was. Daar kan bespiegel word dat die jodium moontlik 'n direkte effek op bloedvatwande het en dat die uiteinde hiervan arteriosklerose is, maar hierdie bewering verdien verdere ondersoek. 'n Ander verandering wat gesien was in die bloedvate was klein fibrienstolsels in die kleiner arteries. Hierdie trombi was slegs waargeneem in biopsies wat na 6 maande en 'n jaar geneem was. Die letsel het uitgebreid voorgekom in alle klein bloedvaatjies. Die rede vir die verskynsel is onduidelik, die moontlikheid bestaan egter dat die stadiger bloedvloei in die arteriosklerotiese vate kan aanleiding gee tot die vorming van die trombi in kleiner vaatjies.

Die belangrikste letsel wat in die eksperiment waargeneem was, was die uiteindelike fibrose van die lamina propria. Dit is 'n welbekende feit dat verhoogde kollageenneerlegging op chroniese ontsteking of irritasie volg. Chroniese degeneratiewe endometritis en herhaaldelike akute endometritis lei dikwels tot bindweefselproliferasie[10]. Fibrose vind eerstens rondom die kliere plaas en later betrek dit die hele *stratum spongiosum*[10]. Bykomend tot die fibrose was daar ook epiteel- en klierhiperplasie en metaplasie.

Die uiteindelike gevolgtrekking is dus dat jodium installering in die baarmoeder die orgaan sodanig beskadig dat dit sal lei tot onvermoë om beset te raak as gevolg van die erge fibrose asook die epiteelveranderinge in die baarmoederwand. Die merries is nie getoets of hulle dragtig sou word nie aangesien Rossdale & Ricketts[10] fibrose beskou as 'n hoofoorsaak van subfertiliteit en dit die mees algemene histopatologiese bevindinge in biopsies van onvrugbare merries is.

VERWYSINGS

1. Ekman L, Holmberg O, Settergren I, Thoreli C B 1965 Resorption of iodine in Lugol's solution and in an iodophor from the uterus of cows. Nordisk Veterinaer-Medicin 17, 391-396
2. Ginter O J, Meckley P E 1972. Effect of intra-uterine infusion on length of diestrus in cows and mares. Veterinary Medicine/Small Animal Clinician 67: 751-754
3. Kennedy R M 1978 Cyclic and pathologic changes of the mare endometrium as detected by biopsy with a note on early embryonic death. Journal of the American Veterinary Medical Association 172: 241-262
4. Kennedy R M 1975 Prognostic value of endometrial biopsy of the mare. Journal of Reproduction and Fertility Supplement 23: 347-348
5. Mather E C, Hurtgen J P 1976 The effect of intrauterine manipulation and treatment on the equine estrus cycle. VIII th International Congress on Animal Reproduction and Artificial Insemination, Cracow: 601-603
6. Mather E C, Refsal K R, Gustafsson B K, Seguin B E, Whitmore H L 1979 The use of fibre-optic techniques in clinical diagnosis and visual assessment of experimental intrauterine therapy in mares. Journal of Reproduction and Fertility (Supplement) 27: 293-297
7. Morrow D A 1980 Current Therapy in Theriogenology W B Saunders Company Philadelphia London Toronto
8. Nakahara T, Domeki I, Yamauchi M 1971. Synchronization of estrus cycle in cows by intra-uterine injection with iodine solution. National Institute of Animal Health Quarterly 11: 219-220
9. Ricketts S W 1975 The technique and clinical application of endometrial biopsy in the mare. Equine Veterinary Journal 7: 102-108
10. Rossdale P D, Ricketts S W 1980 Equine Stud Farm Medicine. Balliére Tindall, London
11. Seguin B E, Morrow D A, Oxender W D 1974 Intra-uterine therapy in the cow. Journal of the American Veterinary Medical Association 164: 609-612
12. Thompson S W, Hunt R D 1966 Selected Histochemical and Histopathological Methods. Charles C Thomas, Springfield.
13. Van Dyk E, Immelman A, Van Heerden J S 1982 Die gebruik van amikasien in die behandeling van *Pseudomonas aeruginosa* veroorsaakte endometritis in die merrie. Tydskrif van die Suid-Afrikaanse Veterinêre Vereniging 53: 124-126

ABSTRACT

SAMEVATTING

MYCOPLASMAS RECOVERED FROM BOVINE GENITALIA, ABORTED FOETUSES AND PLACENTAS IN THE REPUBLIC OF SOUTH AFRICA

A total of 917 *Mycoplasma* isolations were made from 4 092 specimens from 2 874 cattle in private herds and at AI stations. The percentages of isolations from the different sources were: cervico-vaginal mucus 14,6 %, semen 43 %, preputial wash 25 %, foetuses 3,3 % and placentas 15 %. *Mycoplasma bovigenitalium*, the most common isolate, was recovered from 39 % of males, 47 % of females, 25 % of foetuses and 11 % of placentas. A wide spectrum of mycoplasmas was present, and varying combinations were common. The possible pathogenic significance of the isolates is discussed. (Trichard, C.J.V. & Jacobsz, Elsie P., 1985. Mycoplasmas recovered from bovine genitalia, aborted foetuses and placentas in the Republic of South Africa. *Onderstepoort Journal of Veterinary Research*, 52, 105-110 (1985).)

ARTICLE ARTIKEL

EFFECT OF IVERMECTIN ON THE REPRODUCTIVE POTENTIAL OF BREEDING RAMS

J. SCHRÖDER*, G.E. SWAN**, RUTH A. BARRICK*** and J.D. PULLIAM***

ABSTRACT: Schröder J; Swan G E; Barrick R A; Pulliam J D. **Effect of ivermectin on the reproductive potential of breeding rams.** *Journal of the South African Veterinary Association* (1986) 57 No. 4, 211-213 (En) MSD Research Centre, Private Bag X3, 1685 Halfway House, Republic of South Africa.

Ivermectin was administered orally 6 times at 21d intervals at 400 µg/kg (double the recommended dosage) to 10 Merino rams. Ten other rams served as untreated controls. Semen samples were collected serially before and after each treatment, and evaluated for volume, density, colour, motility, pH, percentage live sperm and sperm morphology. One testis was removed from each ram 9d after the last treatment, and histological examinations were performed on testicular sections. The semen of the 2 groups was similar before the first and after the last treatments. The seminal pH of treated rams was lower during the periods following the first 5 treatments. The volume of ejaculates of treated rams was generally higher during the periods following the first 5 treatments, but not consistently so. No histological differences were observed in the testicular tissue of treated and control animals 9 days after the sixth treatment. It was concluded that repeated treatment with ivermectin at the recommended dosage of 200 µg/kg will not impair the reproductive potential of rams.

Key words: Ivermectin, reproduction, safety, semen quality, sheep testis histology.

INTRODUCTION

Ivermectin (Ivomec Liquid, MSD) is an antiparasitic agent which is effective against gastrointestinal nematodes, nasal bot (*Oestrus ovis*), and some arthropod ectoparasites in sheep[2,3,5]. The recommended therapeutic dosage is 200 µg/kg per os, and sheep tolerate dosages of up to 4 mg/kg[2]. This paper describes a trial in which the effect of repeated treatments with ivermectin on the reproductive potential of rams, was evaluated.

MATERIALS AND METHODS

Animals:

Twenty purebred Merino rams were used. Eighteen rams came from one source, of which 17 were from the same crop (c. 18 months old), while the eighteenth one was from a previous crop (c. 5 months older), and 2 mature rams came from another source. All the rams were clinically sound at the start of the trial and each was identified with a numbered ear tag.

Allocation:

The rams from each source were paired by scrotal size (length and circumference) measured 5d before the first treatment. Within each pair, one animal was randomly allocated to the treated group and the other to the untreated control group.

Treatment:

Ten rams (i.e. one from each pair) were treated orally with ivermectin 6 times at 21d intervals at 400 µg/kg (twice the therapeutic dosage). The other 10 served as untreated controls. An aqueous micelle formulation (Ivomec Liquid, MSD) was used. For accurate calculation of the dosage, the mass of each ram was determined prior to each treatment. Day 0 was the day when the first treatment was given.

Semen collection:

A semen sample was collected from each ram 14, 11 and 5d before, and 1 and 3d after each treatment (except that only one sample was collected after the second treatment (Tables 1 & 2). The first 4 collections were made using a commercially available electroejaculator (Animal Breeding Services, Brooklyn, Pretoria). All subsequent electrostimulations were with a home-made ejaculator[10]. The ejaculates were collected in graduated test tubes.

Semen evaluation:

Each ejaculate was appraised visually to determine its volume, colour, and density[9]. A semen drop was examined microscopically to evaluate sperm motility, and pH was determined to the nearest 0,5 by means of pH indicating paper. An eosin and nigrosin-stained, heat-fixed semen smear was examined microscopically for estimation of the percentage live sperm, and to assess sperm morphology.

Testicular histology:

One testis was removed from each ram 9d after the last treatment. A cross-section from the centre of each testis was fixed in 10% formalin. The fixed specimens were processed routinely for light microscopy. Haematoxylin and eosin-stained tissue sections were examined with a light microscope.

Statistical analysis:

Several statistical analyses were carried out. A preliminary analysis using diagnostic techniques such as normal probability plots and examination of trends, revealed that parametric methods would be appropriate for the analysis of data on semen volume, density, col-

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our, motility, pH and percentage live sperm. A repeated measures analysis of data from collections before and after the first treatment (Days -14 to 16) was done to test whether the first treatment had an effect on semen quality.

To test if any short-term patterns following any treatment could be detected, polynomials were fitted to the 5 observations following each of the first 5 treatments. Since these fits indicated that any difference between treatments would be sufficiently detected using only average values of data collected after each treatment, a repeated measures analysis was done on the averages following each of the first 5 treatments.

Finally, the average post-final treatment (Days 106 and 108) minus the average pre-initial treatment (Days -14, -11, and -5) values for each animal were analysed, to test whether there was any difference between the status before treatment and the status at the end of the trial. Live mass at the times of treatment was analysed using analysis of covariance for a repeated measures design, with mass on Day 0 used as a covariate.

RESULTS

Density, colour, motility, and percentage live sperm:

No significant ($p>0,10$) differences were found between the ivermectin-treated and untreated control groups in the analysis of the above characteristics. The status for both groups at the end of the trial, was similar to that at the beginning.

Volume:

The average volume of the ejaculates of the ivermectin group (0,88 ml) was greater than that of the control group (0,75 ml) following the first treatment (Table 1). This difference was statistically significant ($p<0,05$). A significant ($p<0,05$) treatment x repeated treatment interaction was found; the average ejaculate volume of the ivermectin group was generally greater than that of the control group after each of the first 5 treatments, except after the third treatment when the reverse occurred.

The difference between the pre-initial (Days -14, -11, and -5) and post-final treatment (Days 106 and 108) values was close to zero for the 2 groups.

Semen pH;

The average pH of semen from the treated group was lower than that from the control for all collection periods (Table 2). The treatment x repeated treatment interaction was not significant ($p>0,10$). Although the difference was statistically significant ($p<0,05$) for ejaculates collected after the first and fourth treatments, the overall treatment effect in the repeated measures analysis was not statistically significant ($0,05<p<0,10$).

Sperm morphology:

There was a comparable low incidence of abnormalities (loose heads, bent tails) in untreated and treated rams. The incidence of cytoplasmic droplets was also similar in sperm from rams in each group. These data were not statistically analysed.

Live mass:

The rams in the ivermectin group were initially heavier, and continued to be so until the last treatment. They gained slightly more mass than the untreated controls during the trial. When the masses were adjusted for the initial diferences by analysis of covariance, their live mass on Day 105 (day of the sixth treatment) was greater than that of the controls, but this difference was not significant ($p>0,10$).

Histology:

No histological differences were seen in the testicular tissue of treated and control groups. The seminiferous tubules contained spermatogonia, primary spermatocytes, secondary spermatocytes, spermatids, spermatozoa, and Sertoli cells. No difference was found in spermatogenesis between the treated and control groups.

Interstitial cells and other cells were histologically similar in the 2 groups. No signs of testicular degeneration such as hypoplastic tubules, spermatic giant cells, multinucleated cells, vacuole formation, loss of germinal cells, interstitial cell hyperplasia, basement membrane thickening, or increased peritubular connective tissue which exceeded the normal background incidence of these phenomena in untreated control animals, were found. Within sections, the seminiferous tubules were in various stages of the spermatic cycle with minor variations in intensity within and between animals. The histological data were also not statistically analyzed.

DISCUSSION

Collection of semen from rams by electrostimulation normally yields ejaculates of variable volume (0,5 – 2,5 ml)[9]. In the period following the first treatment, differences occurred in the ejaculate volume of the 2 treatment groups. These differences were statistically significant ($p<0,05$), but generally so small (0,02 – 0,15 ml for period averages) that they were not considered to be clinically important (R I Coubrough 1983 Faculty of Veterinary Science, Onderstepoort, personal communication).

It is difficult to assess the importance of the differences in semen pH between the 2 groups. These differences attained statistical significance ($p<0,05$) only for ejaculates collected after the first and fourth treatments. The pH of ram semen tends to decrease on standing following collection[7], but it is unlikely that the semen of treated rams consistently stood for longer than that of the controls prior to pH measurement.

Few authors appear to attach much significance to the reliability of pH as a criterion of semen quality[4,7,8], particularly following electrostimulation[1,6]. If anything, ram semen with a pH value in the lower section of the range is considered to be highly fertile[8] (all other factors being equal), and rams with increased semen pH tend to be less fertile[1]. Our difficulty in assessing the importance of the observed differences in pH could most likely have been obviated by using a measuring method more refined than pH-indicating paper.

It was concluded that repeated treatment with ivermectin at the recommended dosage of 200 µg/kg will have no harmful effect on the reproductive potential of rams.

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REFERENCES

1. Ball L 1976 Electroejaculation. In: Klemm W R (ed.) Applied Electronics for Veterinary Medicine and Animal Physiology. Charles C Thomas, Illinois.
2. Campbell W C, Benz G W 1984 Ivermectin: A review of efficacy and safety. Journal of Veterinary Pharmacology and Therapeutics 7: 1-16
3. Egerton J R, Birnbaum J, Blair L S, Chabala J C, Conroy J, Fisher M N, Mrozik H, Ostlind D A, Wilkins C A, Campbell W C 1980 22,23-Dihydroavermectin B1, a new broadspectrum antiparasitic agent. British Veterinary Journal 136: 88-97
4. Foote R H 1978 Factors influencing the quantity and quality of semen harvested from bulls, rams, boars and stallions. Journal of Animal Science 47 (Suppl 2): 1-11
5. Hotson I K 1982 The avermectins. A new family of antiparasitic agents. Journal of the South African Veterinary Association 53: 87-90
6. Hulet C V 1977 Prediction of fertility in rams: factors affecting fertility, and collection, testing, and evaluation of semen. Veterinary Medicine/Small Animal Clinician 72: 1363-1367
7. Salisbury G W, Lodge J R 1962 Metabolism of spermatozoa. Advances in Enzymology 24: 35-104
8. Symington R B 1961 Studies on the adaptibility of three breeds of sheep to a tropical environment modified by altitude. V. The annual fluctuation in breeding ability of rams maintained on the Rhodesian highveld. Journal of Agricultural Science 56: 165-171
9. Van Tonder E M 1977 Examination of rams for genital soundness. Journal of the South African Veterinary Association 48: 267-272
10. Van Tonder E M, Bolton T F W, Robertson A O, Greef L 1973 Modification and use of the generator type of electro-ejaculator for rams. Journal of the South African Veterinary Association 44: 437-439

Table 1. Mean ejaculate volumes for each treatment group for each period of the trial

Trial period*	Volume (ml)	
	Untreated	Ivermectin 400µg/kg
Before treatment (Days -14, -11, -5)	0,74	0,69
After 1st treatment (Days 1, 3, 7, 11, 16)	0,75[a]	0,88[b]
After 2nd treatment (Days 22, 28, 32, 37)	0,67	0,82
After 3rd treatment (Days 43, 45, 49, 53, 58)	0,75	0,72
After 4th treatment (Days 64, 66, 70, 74, 79)	0,75	0,82
After 5th treatment (Days 85, 87, 91, 95, 100)	0,74	0,76
After 6th treatment (Days 106, 108)	0,74	0,69

*Day 0 is the day of the first treatment
[a,b]Means for the same period having no superscript in common, are statistically different ($p<0,05$)

Table 2. Semen pH for each treatment group for each period of the trial

Trial period*	pH	
	Untreated	Ivermectin 400 µg/kg
Before treatment (Days -14, -11, -5)	6,09	6,07
After 1st treatment (Days 1, 3, 7, 11, 16)	6,24[a]	5,72[b]
After 2nd treatment (Days 22, 28, 32, 37)	6,15	5,71
After 3rd treatment (Days 43, 45, 49, 53, 58)	5,90	5,53
After 4th treatment (Days 64, 66, 70, 74, 79)	5,85[a]	5,38[b]
After 5th treatment (Days 85, 87, 91, 95, 100)	5,88	5,65
After 6th treatment (Days 106, 108)	6,18	6,12

*Day 0 is the day of the first treatment
[a,b]Means for the same period having no superscript in common, are statistically different ($p<0,05$)

ABSTRACT **SAMEVATTING**

RESPONSE OF SHEEP AND CATTLE TO COMBINED POLYVALENT *PASTEURELLA HAEMOLYTICA* VACCINES

The antibody response to various combined polyvalent *Pasteurella haemolytica* vaccines was studied in sheep and cattle.

In sheep, certain oil adjuvant vaccines gave rise to a better antibody response to *P. haemolytica* than an $A1(OH)_3$-adsorbed vaccine. This finding, however, was not consistent for all serotypes, and with respect to *P. multocida*, oil adjuvants had no advantage. Furthermore, it was found that the removal of all the culture supernatent fluid during the production process had no deleterious effect on the antigenicity of the product.

In cattle, good responses were obtained with both alum-precipitated and $A1(OH)_3$-adsorbed vaccine where all culture supernatent fluid was not removed during the production process. No advantage was gained with oil emmulsion vaccines.

The degree of immunity afforded to mice and the antibody response to different serotypes of *P. haemolytica* varied considerably. Further detailed studies with respect to specific serotypes of *P. haemolytica* are therefore required. (Cameron, C.M. & Bester, Faith, J., 1986. Response of sheep and cattle to combined polyvalent *Pasteurella haemolytica* vaccines. *Onderstepoort Journal of Veterinary Research*, 53, 1-7 (1986).)

BOOK REVIEW **BOEKRESENSIE**

TEXTBOOK OF VETERINARY DIAGNOSTIC RADIOLOGY

DONALD E. THRALL

1st Edn., W B Saunders Co., Philadelphia, PA 19105, 1986, pp vii and 563, illustrations 705, Price £46
(ISBN 0-7216-1199-0)

As mentioned in the preface the purpose of this book was to provide a comprehensive book on veterinary radiology which can be used as a text by veterinary students and as a reference work by both equine- and companion animal practioners.

It has a multiple-author format to which no less than 31 distinguished radiologists, mostly from the United States of America, have contributed chapters.

Where possible the roentgen sign method of radiographic interpretation was utilized in the text. The basis of this method, as explained in the first chapter, is the recognition of basic radiographic abnormalities involving changes in size, shape, number, location, margination and radiopacity which are then correlated with the signalment, history, and clinical and laboratory data in order to formulate a differential diagnosis.

A discussion of the basic principles of radiography and radiographic technique falls outside the scope of this book. Excellent books on this subject already exist so this omission is no shortcoming. Where necessary the reader is informed about newer techniques and positionings which may not yet have found their way into the standard texts on radiographic technique.

The book is divided into 9 sections and 47 chapters of which 4 sections (31 chapters) deal with conditions affecting companion animals and 3 sections (12 chapters) with conditions affecting equines.

The 2 chapters in the introductory section expound the basic principles of image formation and radiographic interpretation in general as well as basic guidelines for the classification of bone lesions into aggressive, semi-aggressive and non-aggressive categories. As orthopaedic conditions form the bulk of radiographic examinations in general practice these guidelines are extremely helpful diagnostic and prognostic tools.

The next 6 sections deal, in tandem fashion, with the axial skeleton, appendicular skeleton, and neck and thorax of companion animals and equines. With the exception of the salivary glands every conceivable radiologically significant anatomical part or organ is discussed in the 3 sections (179 pages) concerning companion animals. In the 12 chapters (139 pages) included in the 3 corresponding sections concerning equines discussion was confined to the skull, the nasal passages, sinuses and guttural pouches, the spine (mainly cervical), the stifle and tarsus, the carpus, the metacarpus/metatarsus, the fetlock, the phalanges, the navicular bone, the larynx, pharynx and trachea, the pleural space, and the lungs.

Section 8 contains 14 chapters (165 pages) on abdominal disorders affecting companion animals only. In addition to discussion of the abdominal organs usually contained in radiology textbooks this section also contains chapters on abdominal masses, the adrenal glands, abdominal lymphnodes, and the ovaries and testes.

The final section (25 pages) has chapters on avian radiography and alternate imaging techniques. The latter techniques (X-ray computed tomography, magnetic resonance imaging, scintigraphy and diagnostic ultrasound) will probably not be of much direct interest to private practitioners.

The book is profusely illustrated with good quality reduced photographs of the original radiographs but it is not intended to be a comprehensive atlas of pathological conditions that could affect animals. Where necessary line drawings were used to illustrate the text or help to explain the photographs. Some sonograms were also included in various chapters but mainly in the one dealing with the uterus and pregnancy.

A further good point of the book is the fact that the normal radiological anatomy of the region or organ under discussion is explained and illustrated at the beginning of each chapter.

A list of references concludes each chapter. These references are very up to date and include many from 1984 and even some from 1985. A comprehensive index is also provided at the end of the book.

The book succeeds admirably in providing a comprehensive review of modern veterinary radiology. As far as could be ascertained most of the known pathological conditions are illustrated and/or discussed or at least mentioned. The reader must however not expect to find answers to all his radiological problems or in depth discussions of all the conditions that can be diagnosed radiologically. There are for instance no explicit guidelines for the grading of hip dysplasia. Although a differential diagnostic list of the causes of wobbler syndrome in dogs is provided the reader is not told how to distinguish between the numerous causes. This shortcoming is probably inherent in any book which tries to deal comprehensively with a vast subject in one single volume.

Some conditions like *Spirocerca lupi* granuloma of the oesophagus are mentioned briefly in the text but no further differential diagnostic details are provided. This particular condition was not listed in the index. The scant attention it receives is probably due to the fact that it is rarely if ever seen in the United States.

The roentgen sign method is strictly followed in some chapters but in others the authors have preferred to use the more usual classification based on aetiological agents e.g. infection, neoplasia, etc. In some chapters the two approaches are combined. Both have advantages and merits.

Although metabolic bone conditions are referred to and illustrated in several chapters, it might be a good idea for future editions to contain a chapter specifically dealing with them in a more coherent and comprehensive way and including some up to date information about their aetiologies.

This book is reasonably priced for what it contains and can be recommended wholeheartedly.

C.J. Roos

ARTICLE ARTIKEL

BRUCELLOSIS SEROLOGY: REDUCED DOSE S19 VACCINATION OF YEARLING HEIFERS VERSUS THE USE OF THE STANDARD DOSE AT 5-7 MONTHS OF AGE IN A CLEAN HERD

S. HERR*, P.P. BOSMAN**, W.J. EHRET***, LESLEY A. TE BRUGGE*, CATHERINE C. WILLIAMSON* and P.M. PIETERSON*

ABSTRACT: Herr, S.; Bosman P.P.; Ehret W.J.; Te Brugge L.A.; Williamson C.C.; Pieterson P.M. **Brucellosis serology: reduced dose S19 vaccination of yearling heifers versus the use of the standard dose at 5-7 months of age in a clean herd.** *Journal of the South African Veterinary Association* (1986) 57 No. 4, 215-219 (En) Veterinary Research Institute, 0110 Onderstepoort, Republic of South Africa.

In the 427 heifers vaccinated at 12 to 14 months of age with the reduced dose (3 x 10^8 to 1 x 10^9 viable organisms per dose) 3 had antibody titres in the complement fixation test of 196, 688 and 748 IU ml^{-1}, respectively, at 5 months post-inoculation. At 23 to 25 months of age 8/128 (6,3%) of these heifers had rose bengal test (RBT) reactions, at least 6/128 (4,7%) a serum agglutination test (SAT) antibody titre in excess of 30 IU ml^{-1} and 1/128 had a suspicious reaction in the complement fixation test (CFT) of 30 to 49 IU ml^{-1}.

In the 116 heifers inoculated with the standard dose (4 to 12 x 10^{10} viable organisms) at 5 to 7 months of age and tested at 22 to 24 months of age, a reactor rate of 37/116 (31,9%) in the RBT was seen. At least 34/116 (29,3%) had antibody titres in excess of 30 IU ml^{-1} in the SAT. Only 1 animal had a suspicious reaction in the CFT of 18 to 24 IU ml^{-1}.

No abortions could be attributed to the vaccine strain and no other isolates of the vaccine strain were made.

The possible role of vaccine residues in syringes as a cause of persistent reactions is discussed. The use of disposable syringes is recommended. On serological grounds there appears to be little advantage in using the reduced dose vaccine in yearling heifers as opposed to the standard dose vaccination of 5 to 7 month old heifers.

Key words: Bovine brucellosis, serology, vaccination.

INTRODUCTION

The work done by Plommet[20] proved the effective immunogenicity and lowered antigenicity of reduced doses of *Brucella abortus* strain 19 (S19) vaccine in cattle. Subsequent experimental and field work has been done establishing these 2 criteria to a lesser or greater degree. Woodard & Jasman[21], working with heifers of over 275 to 350 kg and using a dose of $5,9 \times 10^7$ viable organisms, found no card test or complement fixation test (CFT) positive reactions after 5 months post-inoculation. At 20 weeks after the inoculation of 13-month-old heifers other workers[7] found no card test, no serum agglutination test (SAT) reactions above 100 International Units per millilitre (IU ml^{-1}) and no CFT reactions of 53 IU ml^{-1} or higher. Corner & Alton[5], using a dose of 3×10^8 organisms, found 1,8% of the animals serologically positive after 3 months. Barton & Lomme[3] listed residual vaccinal titres as one of the disadvantages of the adult reduced dose vaccination. A high percentage of rose bengal test (RBT) reactions, persisting for up to 23 months in infected herds, was recorded after using a dose of 1,9 to $3,8 \times 10^9$ viable organisms[14]. In Australian work[1], where a 3×10^8 dose was used in 15-month-old heifers, some RBT reactions persisted for 7 months and 2/14 animals recorded CFT titres of 86 and 688 IU ml^{-1}, respectively, at that time. In the USA[9], work done on brucellosis-free properties, showed that both RBT and CFT titres persisted for more than 6 months and that breed and/or location played significant roles in the numbers of animals reacting to the tests.

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Doses that varied from 1×10^7 to 3×10^8 provided adequate immunity when used in heifers of 12 to 15 months of age[17,21].

An advantage of using the reduced dose vaccine in heifers at 12 to 14 months of age as opposed to the standard dose at 5 to 7 months of age would lie in a reduction in residual vaccinal titres. This is especially important when and if the heifers are offered for sale.

In order to compare the residual vaccinal titres resulting from these procedures 2 groups of heifers on brucellosis-free properties were selected and subjected, for the first time, to either the standard dose vaccination at 5 to 7 months of age or the reduced dose ($3 \times 10^8 - 1 \times 10^9$ viable organisms) at 12 to 14 months of age, respectively.

The serological results were compared with groups of unvaccinated bulls and groups of older cows which had been inoculated as heifers, all from these same properties.

MATERIALS AND METHODS

Animals and Specimens

Location and herd history

This beef herd was composed of the Bovelder breed (a mixture of mainly the Simmenthaler, British beef breeds, Charolais and various *Bos indicus* breeds) located on the two Johannesburg Municipal sewage farms, south and north of the city. The herd had been monitored over a number of years for the presence of brucellosis by examination of a large percentage of any abortions occurring on the properties. *Brucella* had never been isolated from the foetuses submitted and no unexplained abortion storms had occurred.

Group A

Four hundred and twenty seven heifers between the ages

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of 12 and 14 months were inoculated subcutaneously (s/c) for the first time with S19 containing the reduced dose of 3×10^8 to 1×10^9 viable organisms. Serum was taken on the day of inoculation, at 5 and 11 months post-inoculation for serological testing. Two heifers had died so that only 425 heifers were tested at 5 months post-inoculation. Three heifers with high CFT titres were slaughtered after the 5 month test and 294 were sold on being certified pregnant shortly thereafter. Only 128 heifers remained for the test carried out at 11 months post-inoculation.

The heifers were bred 3 weeks post-inoculation and the non-pregnant ones 7 months later. The few abortions that occurred after the first breeding season were submitted for diagnostic examination together with serum from the dam on the day of abortion and 3 weeks later. In this group there was no possibility of S19 vaccine residues in syringes used for other purposes acting as an antigenic booster.

Group B

One hundred and sixteen heifers were inoculated s/c, once only, with the standard dose S19 (4 to 12×10^{10} viable organisms) between the ages of 5 to 7 months. Their sera were tested serologically for brucellosis 17 months post-inoculation when they were 22 to 24 months of age and were in the same stage of pregnancy as the later bred group A heifers. Some booster residues of S19 vaccine may possibly have been present in syringes used for other purposes in this group.

Group C

Eighty four bulls which were kept together with the group B heifers until weaning, at 7 months of age, and which were subjected to all other inoculations together with this group were themselves not inoculated with S19. Serum was submitted for brucellosis serology at the same time as the group B heifers. Until weaning (at 7 months) this group was exposed to the same risk of S19 vaccine residues as the group B heifers, but not thereafter.

Group D

One hundred and ninety cows of 3 to 11 years of age had been inoculated s/c, once only, with the standard dose S19 as heifers when between 5 and 7 months of age. Serum from these animals was submitted for brucellosis testing at the same time as the group B heifers and group C bulls. These cows were exposed as heifers to S19 vaccine residues in syringes to the same extent as the group B heifers. Later they were exposed on an annual basis at the same risk rate as the group C bulls and group B heifers, but for a number of years as opposed to 1 year in the case of the bulls and the heifers.

Examination of abattoir material from 3 heifers in group A

When 3 of the group A heifers showed CFT serological titres of 196, 688 and 784 IU ml^{-1} respectively at 5 months post-inoculation, they were slaughtered and the isolation of *Brucella* attempted. Separate specimens were taken from left and right retropharyngeal, lumbar, medial iliac and supramammary lymph nodes respectively, from each quarter of the udder, from the uterine wall (none was pregnant) and from the spleen. On the day of slaughter a bacteriological needle was used to pick up a globule of material from a freshly cut surface of each specimen as a modification of the method of directly plating out from a freshly cut surface[2]. This globule was streaked out on 5 different agar media as previously described[12 13]. The specimens were stored at $-12°C$ for 2 weeks. They were then thawed and the plating-out was repeated. All plates were incubated in air as well as in an atmosphere containing 10% CO_2 at 37°C. Cultures were examined after 48 and 96h before being discarded as negative.

Serological methods and interpretation of results

The RBT, the SAT and the CFT were applied as previously described[10] and modified[11 15 16]. In previous work in adult vaccinated cattle a CFT antibody titre of 392 IU ml^{-1} or higher at 2 to 5 months post-inoculation was regarded as indicative of infection[14]. These criteria were arbitrarily applied to the group A heifers. After 6 months post-inoculation a CFT antibody titre of 30 to 49 IU ml^{-1} was regarded as suspicious and a titre of 60 IU ml^{-1} or higher as positive in the group A heifers[14]. In all the other groups an 18 to 24 IU ml^{-1} end-point in the CFT would be regarded as a suspicious result and a titre of 30 IU ml^{-1} or higher as positive[15]. The highest titre recordable was 784 IU ml^{-1} in the CFT and 1352 IU ml^{-1} in the SAT. All references to titres of 784 IU ml^{-1} in the CFT identify titres of this magnitude or higher. In the SAT, due to the dilutions used, titres in the lower 2 dilutions represented 19 to 21 IU ml^{-1} and 54 to 98 IU ml^{-1}, respectively (Table 2). When titres of 30 IU ml^{-1} or greater in SAT are reported on, those animals with a titre of 54 IU ml^{-1} or greater can be recorded as the least number involved but some of those with a titre of 21 IU ml^{-1} may have exceeded the 30 IU ml^{-1} antibody concentration.

RESULTS

Isolation of *Brucella* and serological tests on aborters

No isolation of *Brucella* was achieved either from the aborted foetuses or from the abattoir specimens taken from the 3 serologically positive heifers in group A at 5 months post-inoculation. None of the aborters in group A was serologically positive at the time of or after the abortions.

Persistence and magnitude of serological reactions

RBT

Of the 8 animals in group A which showed a RBT reaction prior to inoculation 2 (25%) were positive on the subsequent tests. There were 53/425 (12,5%) RBT reactors in group A at 5 months post-inoculation (Table 1). At the age of 23 to 25 months, 11 months post-inoculation, 8/128 (6,3%) of the group A animals had RBT positive reactions compared with 37/116 (31,9%) of the group B (22 to 24 months old and 17 months post-inoculation), 0% in the unvaccinated bulls and 8/190 (4,2%) in the group D cows (Table 1).

SAT

The group A heifers showed SAT antibody titres of 19 to 21 IU ml^{-1} in 5/427 (1,2%), and a titre of 54 IU ml^{-1} in 3/427 (0,7%) of cases prior to inoculation (Table 2). By 5 months post-inoculation 1/5 (20%) of the animals with titres of 19 to 21 IU ml^{-1} still showed a titre of 21

Table 1: The persistence of serological reactions in the Rose Bengal test

	Group A	Group B	Group C	Group D
Vaccine dose used	Reduced[a]	Standard[b]	Unvaccinated	Standard
Age at vaccination (months)	12 – 14	5 – 7	Unvaccinated	5 – 7
Number and percentage positive before vaccination	8/427 (1,9%)[c]	—[d]	—	—
Number and percentage positive 5 months post-vaccination	53/425 (12,5%)	—	—	—
Age at final test (months)	23 – 25	22 – 24	22 – 24	>36
Number and percentage positive at final test	8/128 (6,3%)	37/116 (31,9%)	0/84	8/190 (4,2%7)

[a] Reduced dose = $3 \times 10^8 - 1 \times 10^9$ viable organisms per dose
[b] Standard dose = $4 - 12 \times 10^{10}$ viable organisms per dose
[c] Of these 8 animals only 2 were positive at the subsequent tests
[d] — = Not done

Table 2: The persistence and magnitude of serological reactions in the Serum Agglutination Test (SAT)

	Group A			Group B	Group C	Group D
Time elapsed post-vaccination (months)	0	5	11	17	Not vaccinated – same age as group B	30
Number and percentage of animals with SAT titres of: 19-21 IU ml^{-1}	5/427 (1,2%)	87/425 (20,5%)	15/128 (11,7%)	23/116 (19,8%)	3/84 (3,6%)	26/190 (13,7%)
54-89 IU ml^{-1}*	3/427 (0,7%)	25/425 (5,9%)	6/128 (4,7%)	34/116 (29,3%)	0/84	2/190 (1,1%)
215-338 IU ml^{-1}	0/427	2/425 (0,5%)	0/128	0/116	0/84	0/190
860-1352 IU ml^{-1}	0/427	0/425	0/128	0/116	0/84	0/190

*Some importing countries require a SAT titre of less than 30 IU ml^{-1} which would make at least this number and percentage ineligible.

Table 3. The persistence and magnitude of serological reactions in the Complement Fixation Test (CFT)

	Group A			Group B	Group C	Group D
Time elapsed post-vaccination (months)	0	5	11	17	Not vaccinated – same age as group B	≥30
Number and percentage of animals with CFT titres of: 18-24 IU ml^{-1}	0/427	3/425 (0,7%)	2/128 (1,6%)	1/116 (0,9%)*	0/84	0/190
30-49 IU ml^{-1}	0/427	4/425 (0,9%)	1/128 (0,8%)*	0/116	0/84	0/190
60-98 IU ml^{-1}	0/427	2/425 (0,5%)	0/128	0/116	0/84	0/190
120-196 IU ml^{-1}	0/427	1/425 (0,2%)	0/128	0/116	0/84	0/190
240-392 IU ml^{-1}	0/427	0/425	0/128	0/116	0/84	0/190
480-784 IU ml^{-1}	0/427	2/425 (0,5%)**	0/128	0/116	0/84	0/190

* = Suspicious results
** = Positive results

IU ml^{-1} while the other 4 had reverted to negative status. Of the 3 animals with original titres of 54 IU ml^{-1}, 2 had reverted to negative status by 5 months post-inoculation and 1 still recorded as titre of 21 IU ml^{-1}.

At 5 months post-inoculation the group A heifers had SAT antibody titres of 19 to 21 IU ml^{-1} in 87/425 (20,5%), of 54 to 89 IU ml^{-1} in 25/425 (5,9%) and of 215 to 338 IU ml^{-1} in 2/425 (0,5%) of cases (Table 2). The 2 animals with titres of 215 and 338 IU ml^{-1} were among the 3 animals slaughtered to determine the presence of *Brucella* organisms. Thus they and other animal slaughtered were not included in the final serological tests carried out at 11 months post-inoculation.

When the final serological tests were done no animals in any of the groups had SAT antibody titres of 215 IU ml^{-1} or higher (Table 2). Of those animals with titres of 19 to 21 IU ml^{-1} there were 15/128 (11,8%) in group A, 23/116 (19,8%) in group B, 3/84 (3,6%) in group C and 26/190 (13,7%) in group D. Antibody titres of 54 to 89 IU ml^{-1} were seen in 6/128 (4,7%) in group A, 34/116 (29,3%) in group B, 0% in group C and 2/190 (1,1%) in group D (Table 2).

CFT

The group A heifers showed negative results in the CFT before inoculation with the reduced dose of S19 at 12 to 14 months of age (Table 3). At 5 months post-inoculation 3 of these animals showed antibody titres of 196, 688 and 784 IU ml^{-1} respectively. These 3 animals were slaughtered and attempts were made to isolate *Brucella* from them. They were therefore not included in the final serological tests of 11 months post-inoculation.

In the group A heifers 2/128 (1,6%) showed antibody titres of 18 to 24 IU ml^{-1} in the CFT at 11 months post-inoculation (Table 3). This compares with 1/116 (0,9%) in group B, 0% in the uninoculated bulls in group C and 0% in group D (old cows inoculated as heifers) with similar antibody concentrations (Table 3). Group A heifers registered titres of 30 to 49 IU ml^{-1} in 1/128 (0,8%) of cases while no titres of this magnitude were seen in any of the other groups (Table 3). No antibody titres greater than 49 IU ml^{-1} were recorded in any of the groups at the end of the experiment (Table 3).

DISCUSSION

The finding that none of the abortions could be attributed to S19 and that none of the aborters in group A was serologically positive for brucellosis is in agreement with reports of other workers that S19 causes less than 1% abortions[17]. The failure to isolate *Brucella* from the 3 animals with high CFT titres 5 months after inoculation, is in keeping with findings in Britian[17], where excretors of S19 following vaccination are a rarity. In the USA[18,19], on the other hand, S19 was frequently isolated from serological reactors following adult inoculation.

The occurrence of the 3/425 cases with CFT titres of 196, 688 and 784 IU/ml, respectively, 5 months after inoculation with the dose of 3×10^8 to 1×10^9 viable organisms, was comparable with results from Australia[1], where 2/14 animals had CFT titres of 86 and 688 IU ml^{-1}, respectively, 7 months after using a dose of 3×10^8 viable organisms. In contradistinction, several workers[7,21] found no such problems at this dosage level. Recent work has demonstrated that the persistence of titres may vary on different properties and in different breeds of cattle[9]. These varying results will undoubtedly cause problems if this reduced dose vaccine is used at 12 to 14 months of age and the animals are sold within a period of less than 7 months post-inoculation. Whether these problem reactors would return to negative status by 12 months after inoculation requires further research. The indications that reactions will vary between herds, locations and breeds make the task of undertaking such research more complicated and probably not worthwhile.

The use of a reduced dose S19 (1,9 to $3,8 \times 10^9$ viable organisms) presented problems with persistant RBT reactions in infected herds[14]. As these reactors to the RBT have all to be subjected to the technically more exacting CFT, a high percentage of reactors could easily overload laboratory facilities[14]. The percentages of RBT positive reactors recorded at the end of the trial differed little between the old cows vaccinated as heifers (Table 1, Group D – 4,2%) and the reduced dose yearling vaccinates (Group A – 6,3%) and were in both cases of manageable proportions. The 31,9% reactor rate in the group B heifers, vaccinated with the standard dose S19 at 5 to 7 months of age, gives cause for concern. In this group the possibility that this high reactor rate could be due to the booster effect of S19 residues in syringes, used for other inoculations, could not be excluded. The fact that the uninoculated bulls (Table 1 – Group C), which were running with the Group B heifers for 1 to 2 months after the S19 inoculations and were subjected to the same other inoculations with the same syringes, failed to show any RBT reactions did not exclude this possibility. The initial dose of 4 to 12×10^{10} *Brucella* organisms may be necessary before the booster effect of residues in syringes is capable of activating an anamnestic response. Recent work[8] has shown that 5 to 7 rinses of a S19 syringe may still leave 1×10^5 organisms *in situ*. The antigenicity of this residue is unaffected by death of the organisms[4,6]. It may therefore have the effect of increasing the antigenicity of S19 subsequently used in that syringe or acting as a booster when applied together with other vaccines. We would strongly recommend that S19 vaccination be done with disposable syringes which should be burnt after use. In the absence of field strains of *Brucella*, as was the case in this trial, these persistent reactions may be due to idiosyncrasies of breed[9], or cross reactions with organisms such as *Yersinia enterocolitica* type 9[2]. This type has not been recorded in animals in South Africa to date. If either of the latter 2 conditions is the cause of the persistent titres no easy solution presents itself as in the case of vaccine residues.

The persistence of SAT antibody titres is of little consequence except in the case of animals for export. Some importing countries stipulate that animals must record antibody titres of less than 30 IU ml^{-1} in the SAT. If export of animals is envisaged it becomes clear that at least 6/128 (4,7%) of Group A, 34/116 (29,3%) of Group B and 2/190 (1,1%) of Group D animals would not qualify at the time of the final test (Table 2).

There was no significant difference at the final test between the suspicious reactions of 1/128 (0,8%) in Group A, 1/116 (0,9%) in Group B and 0% in Group D in the CFT (Table 3). It would appear that whatever the cause of the high percentage of residual titres in the RBT and SAT in the Group B heifers, it was not opera-

tional in the CFT. The serological results that would have been seen at the end of the trial in the 3 animals from group A slaughtered at 5 months after inoculation will remain unknown. At worst they could have contributed a 3/128 (2,34%) component of positive reactors to the CFT in this group. As far as the titres in the CFT are concerned, it must be concluded that there appears to be no advantage to the use of a reduced dose vaccine at 12 to 14 months of age as opposed to the standard dose at 5 to 7 months of age.

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REFERENCES

1. Alton G G, Corner L A, Plackett P 1983 Vaccination of cattle against brucellosis using either a reduced dose of strain 19 or one or two doses of 45/20 vaccine. Australian Veterinary Journal 60: 175-177
2. Alton G G, Jones L M, Pietz D E 1975 In: Laboratory Techniques in Brucellosis 2nd ed. WHO Monograph Series No. 55, Geneva
3. Barton C E, Lomme J R 1980 Reduced-dose whole herd vaccination against brucellosis: a review of recent experience. Journal of the American Veterinary Medical Association 177: 1218-1220
4. Beck C C, Ellis D J, Fichtner G J, Laiho E R, Whitehead G L 1964 Brucella tube agglutination titres resulting from the use of syringes containing viable strain 19 vaccine. Journal of the American Veterinary Medical Association 144: 620-625
5. Corner L A, Alton G G 1981 Persistence of *Brucella abortus* strain 19 infection in adult cattle vaccinated with reduced doses. Research in Veterinary Science 31: 342-344
6. Cullen G A, Corbel M J 1970 Observations on some possible causes of variation in the titre of *Brucella* antibodies in cattle. Veterinary Record 87: 101-106
7. Deyoe B L, Dorsey T A, Meredith K B, Garrett L 1979 Effect of reduced dosages of *Brucella abortus* strain 19 in cattle vaccinated as yearlings. Proceedings 83rd Annual Meeting of the United States Animal Health Association: 92-104
8. Durand M P, Leplatre J 1984 Etude sur les serologies bovines observe es apres inoculations de faibles quantites de B19. Note a propos d'une contamination accidentelle apres une vaccination anti-aphteuse. Bulletin de l'Academie Veterinaire de France 57: 55-60
9. Hendricks J B, Lomme J, Odenweller R 1983 Reduced dose strain 19 vaccination of adult cattle in brucellosis negative herds. Proceedings of the 87th Annual Meeting of the United States Animal Health Association: 116-128
10. Herr S, Bishop G, Bolton T F W, Van der Merwe D 1979 Onderstepoort brucellosis serology laboratory manual. Department of Agriculture and Fisheries, Veterinary Research Institute, 0110 Onderstepoort.
11. Herr S, Huchzermeyer Hildegard F A K, Te Brugge Lesley A, Williamson Catherine C, Roos J A, Schiele C J 1985 The use of a single complement fixation test technique in bovine brucellosis, Johne's disease, dourine, equine piroplasmosis and Q Fever serology. Onderstepoort Journal of Veterinary Research, 52 : 279-282.
12. Herr S, Roux D 1980 The isolation of *Brucella abortus* biotype 1 from serologically positive reactors following on S19 adult inoculation of dairy cows. Journal of the South African Veterinary Association 51: 255-257
13. Herr S, Roux D 1981 The efficacy of bacteriological procedures for the isolation of *Brucella abortus* from abattoir material. Onderstepoort Journal of Veterinary Research 48: 7-12
14. Herr S, Te Brugge Lesley A 1984 Experience with reduced dose *Brucella abortus* strain 19 vaccination of adult cattle in infected herds in South Africa. Proceedings of the XIIIth World Congress on Diseases of Cattle 1 : 359-365
15. Herr S, Te Brugge Lesley A 1985 Profiles of serological reactions following adult cow inoculation with standard dose *Brucella abortus* strain 19 vaccine. Journal of the South African Veterinary Association 56: 93-96
16. Herr S, Te Brugge Lesley A, Guiney M C M 1982 The value of the microtitre serum agglutination test as a second screening test in bovine brucellosis. Onderstepoort Journal of Veterinary Research, 49, 23-28.
17. Morgan W J B, Nicoletti P 1977 Immunization discussion. In: Crawford, Hidalgo (ed.). Bovine Brucellosis: An International Symposium. Texas A & M University Press, College Station and London
18. Nicoletti P 1979 The effects of adult vaccination with strain 19 on the incidence of brucellosis in dairy herds in Florida and Puerto Rico. Proceedings of the 83rd Annual General Meeting of the United States Animal Health Association: 75-80
19. Nicoletti P 1981 Prevalence and persistance of *Brucella abortus* strain 19 infections and prevalence of other biotypes in vaccinated adult dairy cattle. Journal of the American Veterinary Medical Association 178: 143-145
20. Plommet M 1977 Experimental brucellosis in cows in France. In: Crawford, Hidalgo (ed.) Bovine Brucellosis: An International Symposium. Texas A & M University Press, College Station and London
21. Woodard L F, Jasman R L 1983 Comparative efficacy of an experimental S45/20 bacterin and a reduced dose of strain 19 vaccine against bovine brucellosis. American Journal of Veterinary Research 44: 907-910

ABSTRACT

SAMEVATTING

IMMUNITY AGAINST GENITAL INFECTION BY *HISTOPHILUS OVIS* IN RAMS

Rams have been immunized against an infection of their genitalia by *Histophilus ovis*. An alum-precipitated antigen and an antigen plus Freund's complete adjuvant proved equally effective.

An injection of live *H. ovis* into the epididymal tissue proved to be a better method of challenging immunity than an injection into the vas deferens.

It was shown that cell-mediated immunity, as evidenced by tests for lymphocyte transformation, the presence of a macrophage migration inhibition factor and a delayed hypersensitivity skin reaction did not play a role in the resistance, nor did specific IgG antibodies have any protective influence. It was shown that neutrophils play a cardinal role in the immunity against *H. ovis* infection in so far as they phagocytize and destroy the organisms and are attracted to them by chemotaxis in immune animals. (Jansen, B.C. & Hayes, Marianna, 1984. Immunity against genital infection by *Histophilus ovis* in rams. *Onderstepoort Journal of Veterinary Research*, 51, 203—207 (1984).)

BOOK REVIEW **BOEKRESENSIE**

FARM ANIMAL WELFARE: CATTLE, PIGS AND POULTRY

DAVID SAINSBURY

1ST Edn. Collins Professional and Technical Books, London. 1986 pp 175, illustrations 47, photographs 9, tables 20 Price not given. (ISBN 0-00-383157-4)

In Britian the subject of animal welfare is currently an emotional issue between politicians, "direct-action" activists, and those involved in intensive animal production.

David Sainsbury is a veterinarian and lecturer at the University of Cambridge. Most of this book consists of an account of housing and management under intensive farming systems, and the way in which they relate to animal welfare. He shows how animal behaviour and husbandry affect efficient livestock production, and includes much valuable information about the design of facilities and the practical management of these three classes of livestock.

It might be tempting to assume that these principles have limited application in Southern Africa; and perhaps in many situations we could manage with less sophisticated facilities. Nevertheless, the degree of intensification in the pig and poultry industries, and a similar trend in dairy farming, mean that this book will be a valuabe source of practical information.

It is generally clearly written and well presented.

E.F. Donkin

BOOK REVIEW **BOEKRESENSIE**

ANIMAL HEALTH IN AUSTRALIA VOL. 8 HELMINTH PARASITES OF SHEEP AND CATTLE

V.G. COLE

Australian Government Publishing Service, Canberra, 1978, pp. × + 225, Many illustrations and tables. Price not stated (ISBN 0-644-03286-3)

The book is a review of parasitological information of Australia and is written in an easy to understand language. It is divided into 4 parts, each with several chapters and a list of references following each chapter.

Part 1 deals with general parasitology and provides lists of parasites of cattle and sheep, describes parasitic life cycles, the regional distribution of parasites in Australia and epidemiological studies with cattle and sheep.

Parts 2 and 3 form the bulk of the book. Part 2 deals with the factors that cause parasitic diseases, the symptoms of helminth "infections", the effects of parasitic diseases on animal production and the diagnosis of parasites in the living and dead animal. Useful tables, listing the parasites and the lesions they produce are provided here.

Part 3 deals with the treatment and control of parasites, as is currently practised in Australia. This includes treatment with anthelmintics, control by grazing management and remarks regarding the anthelmintics themselves.

Part 4 deals with various aspects of larval tapeworms, as well as some parasites that are seldom or occasionally encountered. This part is followed by 3 appendices, which summarize the symptoms produced, the diagnosis, control and prevention of the parasites of cattle and sheep, and which list the anthelmintic efficiency.

The book gives a good account of what has been and is being done to control parasites in Australia. Because of the similarities of the climates and the fact that the major pathogenic worms are distributed world-wide, many of the recommendations are applicable to South African conditions as well. Some anthelmintics listed in Appendix 3 are not registered for use in South Africa, and caution is advised when dealing with the epidemiology of some of the worms.

To conclude with the words of the author, "this book is a summary of parasitological information considered to be of specific interest to the field veterinarian involved with sheep and cattle".

J. Boomker

ARTICLE ARTIKEL

TOLAZOLINE AS AN ANTAGONIST IN FREE-LIVING LIONS IMMOBILISED WITH A KETAMINE-XYLAZINE COMBINATION

T.C. VAN WYK* and H.H. BERRY*

ABSTRACT: T.C. van Wyk.; Berry H.H. 1986 **Tolazoline as an antagonist in free-living lions immobilised with a ketamine-xylazine combination.** *Journal of the South African Veterinary Association* (1986) 57 No. 4, 221-224 (En) Directorate of Nature Conservation and Recreational Resorts, Private Bag 13306, 9000 Windhoek, South West Africa/Namibia.

A combination of ketamine at 8,0 mg/kg and xylazine at 3,2 mg/kg was found effective in immobilising lions *(Panthera leo)* for 4 hours. Ataxia and immobilisation were rapidly induced, with stable respiratory rate, heart rate and body temperature recorded. Tolazoline effectively antagonised xylazine via intravenous or intramuscular injection, resulting in a return to mobility within approximately 20 and 60 minutes, respectively. Tolazoline also elevated the respiratory rate. No mortalities occurred during 97 immobilisations of 76 lions.

Key words: Tolazoline antagonist, ketamine-xylazine combination, lion, wildlife.

INTRODUCTION

The major advantage of using ketamine hydrochloride (Ketalar, Parke-Davis) as opposed to phencyclidine hydrochloride (Sernylan, Parke-Davis) for the immobilisation of lions *Panthera leo* L. is the significant time difference in the post-anaesthetic recovery stage[4]. Using dosage rates of 7,8 – 15,1 mg/kg ketamine ($\bar{x}$ = 11,5 mg/kg) Smuts et al.[4] recorded the time to full recovery as 3 – 6 h ($\bar{x}$ = 4h12), whereas phencyclidine at a dosage rate of 1,4 – 2,4 mg/kg ($\bar{x}$ = 1,7 mg/kg) resulted in a time of 8 – 15 h ($\bar{x}$ = 12h) before the lions were completely recovered. Phencyclidine has further disadvantages, namely that the long period required for recovery makes the animal susceptible to hyperthermia[1], the occurrence of violent, epileptiform convulsions[1,2] and the restrictions placed on its prescription and handling due to its Schedule 8 rating by the South African Medical, Dental and Drugs Control Amendment Act, 1974. Ketamine's main disadvantage as an anaesthetic for free-ranging lions has been the relatively large volume required, necessitating projectile darts of 10 ml capacity[2] or applying a divided dosage in two separately fired darts[5]. Consequently, as part of a study on aspects of lion ecology at Etosha National Park, we also investigated the possibility of utilising the apparent synergistic effect of ketamine in combination with xylazine hydrochloride (Rompun, Bayer) to reduce the dosage rate of ketamine. Wiesner[6] recommended a concentration of 100 mg/ml ketamine in combination with 125 mg/ml xylazine. For immobilising captive, adult lion Wiesner[6] administered this combination at a dosage rate of 3 ml mixture plus 1 ml of ketamine. Furthermore, because the effects of xylazine include pronounced muscle relaxation with concomitant respiratory depression and a marked lack of co-ordination upon recovery, especially with high dosage rates, the use of tolazoline hydrochloride (Weimer Pharmaceuticals, Rastatt) as an antagonist to xylazine was also investigated during the present study. The antagonistic effect of tolazoline was reported by Zingoni et al.[7] who administered it to xylazine-sedated domestic sheep.

Tolazoline (2-benzyl-2-imidazoline hydrochloride) acts as an alpha-adrenoreceptor blocking agent which also has a direct dilator action on the peripheral blood vessels, especially the arterioles and capillaries[3]. By intramuscular injection tolazoline produces maximum effects after 30 – 60 minutes and is rapidly excreted largely unchanged via the urine. Minor side effects in humans include pilo-erection and flushing, while major side effects may include severe tachycardia, cardiac arrhythmia, vomiting and diarrhoea. Large doses may result in orthostatic hypotension. Tolazoline is light-sensitive and unstable at high ambient temperatures and should be stored under dark, cool conditions.

MATERIALS AND METHODS

Ketamine in powder form was made up to a standard concentration of 250 mg/ml by using isotonic saline and agitating and heating the solution to 45°C. Since this solute formed crystals when stored below 25°C, reheating and agitation were necessary shortly before preparing the combination with xylazine. Xylazine in powder form was obtained in bottled units of 500 mg. An experimental supply of tolazoline solution at a concentration of 20 mg/ml in sterile water (pH 3-4) was donated by Weimer Pharmaceuticals, Rastatt, West Germany.

Five ml ketamine at 250 mg/ml were injected into a bottle containing 500 mg of xylazine powder and the mixture shaken briefly. This resulted in a concentration of 250 mg/ml ketamine and 100 mg/ml xylazine. Crystallisation of this mixture was less frequent than with the ketamine in solution alone. When crystals formed, only moderate heating (35°C) and agitation were necessary to dissolve them. Due to the light-sensitivity of ketamine, the mixture was always stored in a dark, cool place.

Ketamine-xylazine was administered in 3–5 ml reusable aluminium dart syringes with barbed NC 2 needles and propelled by a 32-gauge rifle fitted with a ,22 adaptor (Palmer Chemical and Equipment Co., Georgia, U.S.A.). "Stun-load" 0,22 blank cartridges

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(Abattoir Engineering Supplies, Ravenmoor, R.S.A.) were used to fire the darts at distances of 20 – 30 m. For distances less than 20 m a Palmer CO_2 gas Cap-chur Rifle was preferable, because of the reduced impact of the dart on the lion. Based on an estimated body mass of 150 kg for an adult lioness and 200 kg for an adult lion, darts of 3 and 4 ml, respectively, were used, resulting in a calculated initial dosage rate of 5 mg/kg ketamine and 2 mg/kg xylazine. This was intended to render the lion sufficiently immobile so that a booster injection of 1,5 ml for females and 2 ml for males could be given by syringe. Thereby the total dosage rate was theoretically set at 7,5 mg/kg ketamine and 3 mg/kg xylazine. Each immobilised lion was weighed to the nearest kilogram to calculate the actual dosage rate. At a later stage this procedure was modified by firing a complete dose in 4 ml darts for adult females and 5 ml darts for adult males. Boosting by hand syringe was only done when it was evident that the animal was insufficiently immobilised. Lions were considered immobile when they became sternally or laterally recumbent after darting and could no longer raise their head. The body mass of large cubs (12 – 24 months) and sub-adults (2 – 4 years) was first estimated and the volume of the drug reduced accordingly. The embedded dart was removed as soon as the lion was tractable to check if complete injection of the drug had occurred. To facilitate rapid absorption of the drug combination, the dart was fired into a body area with a rich blood supply such as the shoulder muscle or the buttocks. Lions were approached to within darting range by vehicle during daytime. If they were not in a suitable position or were found lying in dense vegetation, the gutted carcase of a springbok (*Antidorcas marsupialis*) was dragged as bait around their immediate vicinity. Alternatively, to attract lions, a larger carcase such as gemsbok (*Oryx gazella*), kudu (*Tragelaphus strepsiceros*), or zebra (*Equus burchelli*) was gutted and dragged in the vicinity of a pride's favoured waterhole and then chained to a tree.

Captured lions were routinely given an injection of 15 – 20 ml of long-acting penicillin (Compropen, Glaxo-Allenbury). Dart wounds or any naturally occurring skin injuries were treated with an antibiotic aerosol (Liquamycin, Pfizer). Vitamins and amino-acids concentrate (Stress-Vitamin, Weimer Pharmaceuticals) were injected at a dosage rate of 15 – 20 ml per animal. Possible eye damage through dryness or foreign particles was prevented by liberal applications of antibiotic ophthalmic ointment (Terramycin, Pfizer). To protect the eyes from sunlight a cloth blindfold was applied immediately after the lion was immobile. To prevent hyperthermia, immobilised lions were shaded and when their rectal temperature increased to 40°C they were cooled with water spray, using a pressurised cylinder to obtain finely-dispersed droplets. Critical body functions, namely respiration, heart beat and temperature were monitored every 30 minutes until the lion showed signs of recovery. Recovery from immobilisation was considered to have started when a lion was able to move into a sternal position and raise its head. Under field conditions the emergence from ketamine anaesthesia was gauged by reflexes in the eyes, tongue and jaw muscles.

Because a dosage rate of tolazoline was unknown for lion, increasing doses from 0,5 – 5,0 mg/kg were given by intramuscular and then intravenous route to ascertain the effect. The preferred intravenous route was via the jugular vein.

Statistical evaluations were done, using Student's t-test for 2 means and applying a null hypothesis of no difference between mean recovery times of different treatments.

RESULTS

The effects of a combination of ketamine and xylazine on free-living lions were summarised in Table 1. The effects of tolazoline on lions immobilised with ketamine-xylazine combination are presented in Table 2.

Lions receiving ketamine-xylazine in a single dose, followed by tolazoline, remained immobile for a significantly shorter period than lions not receiving tolazoline ($t=7,61$; $P<0,001$). Similarly, consecutive doses of ketamine-xylazine, followed by tolazoline, resulted in significantly reduced immobilisation periods compared to when tolazoline was not administered ($t = 5,99$; $P<0,001$). When tolazoline was administered intravenously following a single dose of ketamine-

Table 1. Effects of ketamine and xylazine in combination on free-living lions in Etosha during 1981–84

Parameter measured	Drug combination			
	Ketamine-xylazine in single dose (n = 20)		Ketamine-xylazine in two or more consecutive doses (n = 34)	
	$\bar{x}$	±S D	$\bar{x}$	± S D
Body mass (kg)	127	21	161	33
Total ketamine (mg)	1 006	143	1 318	263
Total xylazine (mg)	403	57	519	92
Dosage ketamine (mg/kg)	8,0	1,2	8,3	1,1
Dosage xylazine (mg/kg)	3,2	0,5	3,3	0,4
Time to ataxia (min)	3'06"	47"	6'02"	2'08"
Time to immobilisation (min)	4'33"	1'26"	9'08"	5'47"
Clinical after 60 min				
Respiratory rate/min	16	3	15	3
Heart rate/min	56	11	54	17
Rectal temperature (C°)	38,4	0,9	38,9	1,2
Total immobilisation time (hrs/min)	4h33	1h13	4h23	1h15

Table 2. Effects of tolazoline on ketamine-xylazine immobilised lions in Etosha during 1981–84

Parameter measured	Drug combinations			
	Ketamine-xylazine in single dose followed by tolazoline (n = 18)		Ketamine-xylazine in two or more consecutive doses, followed by tolazoline (n = 24)	
	$\bar{x}$	± S D	$\bar{x}$	± S D
Body mass (kg)	136	16	161	51
Total ketamine (mg)	1 038	83	1 305	319
Total xylazine (mg)	415	33	552	128
Dosage ketamine (mg/kg)	7,7	0,9	8,5	2,2
Dosage xylazine (mg/kg)	3,1	0,4	3,4	0,9
Time to ataxia (min)	3'14"	36	3'39"	1'42"
Time to immobility (min)	5'39"	1'32"	7'20"	3'57"
Clinical after 60 min				
Respiratory rate/min	16	4	15	3
Heart rate/min	54	11	53	7
Rectal temperature (°C)	38,5	1,3	38,6	1,0
Tolazoline given				
Intramuscular (mg)	379	190	428	182
Intravenous (mg)	430	209	639	284
Dosage (mg/kg)	4,0	0,6	3,7	0,9
Respiratory rate after 15 min	18	6	17	3
Respiratory rate after 30 min	21	11	23	12
Respiratory rate after 45 min	41	24	29	28
Respiratory rate after 60 min	67	53	69	44
Time to mobility (min)				
via intramuscular route	74	40	50	23
via intravenous route	20	15	22	16
Total immobilisation time (hrs/min)	2h27	45 min	2h19	38 min

xylazine, lions recovered their mobility in significantly less time than when tolazoline was given intramuscularly ($t = 3,27$; $P < 0,02$). Likewise, intravenous administration of tolazoline following consecutive doses of ketamine-xylazine, reduced the immobilisation period compared to when tolazoline was given intramuscularly ($t = 3,01$; $P < 0,02$).

The ketamine-xylazine combination retained its potency after a storage period of 4 months.

DISCUSSION

The results show that a dosage level of approximately 8,0 mg/kg ketamine in combination with 3,2 mg/kg xylazine provides an effective immobilisation of both sexes of lion for up to 4 hours. It is preferable to administer this combination in a single dose, which is usually possible with all lionesses, but sometimes, in the case of heavy males weighing over 200 kg, a booster injection of additional drugs may be necessary. Ataxia, which was characterised by an hypnotic stare and uncoordinated movement, was noticeable within 3–6 minutes and complete immobilisation took place within 4 – 9 minutes.

Clinical measurements reflected an even respiratory and heart rate, whilst body temperature was not unduly elevated. Prevention of hyperthermia in lions immobilised with ketamine-xylazine is a critical consideration when ambient temperatures are high, if it is considered that panting rates of 120 – 150 per minute were recorded in resting lions subjected to Etosha's summer daytime shade temperatures of 40°C. Tables 1 & 2 show a respiration rate of 15 – 16 per minute after 60 minutes of immobilisation, which represents a decrease of nearly 90% over panting rate.

When tolazoline was administered, a significant reduction in immobilisation time was achieved in all cases. It allowed a return to mobility, albeit uncoordinated, within 50–74 minutes following intramuscular injection and 20 – 22 minutes when infused intravenously. A dosage rate of 4,0 mg/kg was found to produce the best reaction. Increasing the dosage rate to 5,0 mg/kg resulted in muscular seizures in 2 cases. These may have been caused by the total removal of the sedative affects of xylazine, leaving the lion susceptible to typical ketamine excitation. Because lions which were returned to mobility by tolazoline at 4,0 mg/kg showed ataxia to varying degrees, it appears that some effects of xylazine and ketamine were still present. With our present experience this seems preferable to exposing the recovering lion to convulsions and resultant stress and hyperthermia. Tolazoline, in our opinion, should be administered at a sub-maximal dosage rate. When tolazoline is administered it is important to allow the anaesthetic effect of ketamine to wear off which is normally about 2 hours after administration.

Without tolazoline, immobilised lions required about 4,5 hours to become sternal and still displayed ataxia after 5 – 6 hours. In contrast, tolazoline-treated lions were able to walk more steadily 2,5 hours after being darted and joined up with their pride members soon afterwards. An additional advantage of tolazoline is that the lion can defend itself within 30 – 60 minutes of antidoting and need not be guarded for periods of up to 6 hours as was previously the case. Moreover, a tolazoline-aided return to mobility was accompanied by an elevated respiration rate which facilitated cooling. Table 2 shows mean respiration rates of 67 and 69 per minute, 60 minutes after administration of tolazoline, whereas lions which did not receive tolazoline had a

mean respiration rate of 17 per minute (n = 34) after the same time had elapsed (4 hours following immobilisation).

Although no problems were experienced during or after the intravenous infusion of tolazoline, we draw attention to the major side effects listed under the introduction of this paper, especially the possibility of sudden drop in blood pressure which may accompany the intravenous administration of a vasodilator. The intravenous injection of tolazoline was found to be particularly important when rapid recovery was required, for example, to return a lioness to small cubs, or to prevent hyperthermia on hot days. However, if such urgency was not present the intramuscular route was preferred, which reduced the likelihood of severe side effects. Furthermore, 4 out of 10 lions receiving intravenous infusion of tolazoline began reviving whilst it was being administered. During this investigation 97 immobilisations on 76 lions were successfully performed with no mortality or permanent disablement to any of them. No ill-effects of the immobilisations were detectable after 24 hours and no increase in shyness or aggressiveness were noted in any of the animals and even in individuals which were immobilised on 4 separate occasions. We can therefore report with confidence on the efficacy of a combination of ketamine-xylazine for immobilising free-living lions.

ACKNOWLEDGEMENTS

We thank all staff members who assisted in this work. In particular we wish to express our appreciation to Senior Nature Conservators E Cronjé, I Behrens and Nature Conservators P Stander and M Paxton for the help they gave during this investigation. We are also indebted to Dr J Orford, Mrs R Orford and Mrs C Berry for their support during immobilisations. Miss P Rankin assisted with the statistical analysis. Dr W Weimer is thanked for supplying experimental samples of tolazoline and for his personal interest in this study.

REFERENCES

1. Bertram B C R 1976 Lion immobilisation using phencyclidine (Sernylan). East African Wildlife Journal 14: 233-235
2. Ebedes H 1973 The drug immobilisation of wild carnivorous animals. In: Young E (ed.) The Capture and Care of Wild Animals 1st edn Human and Rousseau, Cape Town: 62-68
3. Martindale W 1982 Tolazoline hydrochloride. In: Reynolds J E F (ed.) The Extra Pharmacopoeia 28th edn The Pharmaceutical Press, London: 1633-1634
4. Smuts G L, Bryden B R, De Vos V, Young E 1973 Some practical advantages of CI-581 (ketamine) for the field immobilisation of large wild felines, with comparative notes on baboons and impala. The Lammergeyer 18: 1-14
5. Smuts G L 1983 Lion 1st edn Macmillan South Africa, Johannesburg.
6. Wiesner H 1977 Tranquilization by the "blowgun rifle" method. (Translation) Kleintierpraxis 22: 327-330
7. Zingoni M R, Garcia-Villar R, Toutain P L 1982 La Tolazoline comme Antagoniste de la Sèdation par la Xylazine chez le Mouton. Revue de Mèdecine Vètèrinaire 133: 335-339

ABSTRACT **SAMEVATTING**

SIMULIUM CONTROL IN A SMALL POLLUTED RIVER IN SOUTH AFRICA

The effects on *Simulium adersi* and *S. hargreavesi* larvae of 2 *Bacillus thuringiensis* var. *israelensis* products, the liquid formulation "Teknar" (Sandoz) and a powder formulation produced by the Ben Gurion University, Israel, were compared in the laboratory and in the Pienaars River. This river was heavily polluted with effluent from a nearby sewage works and contained 77 mg/ℓ chloride.

In the laboratory *S. adersi* and *S. hargreavesi* larvae showed 26; 48; 95 and 100 % mortality 6 hours after a 10-minute application of 0,8; 1,6; 3,2 and 16 ppm "Teknar" in rain water. The powder formulation applied at 0,2; 1,2; 2,0 and 30 ppm resulted in a 7; 17; 35 and 100 % mortality. In polluted river-water the mortality was 85 % with 16 ppm "Teknar" and 80 % with 30 ppm *B. thuringiensis* powder.

In the field trials "Teknar" at 1,6 ppm and *B. thuringiensis* powder at 3 ppm did not cause any larval mortality at flow rates of 3 060ℓ/min and 2 040ℓ/min, respectively. However, 24 hours after application of the powder formulation, numbers of *S. hargreavesi* decreased significantly (P = 0,05) 20 m below the application point. A further 24 hours later, after "Teknar" had been applied, the numbers of *S. adersi* decreased and those of Chirnomidae increased significantly. There was a significant increase in *S. hargreavesi* 200 m downstream after treatment with "Teknar".

(Car, M., 1984. Laboratory and field trials with two *Bacillus thuringiensis* var. *israelensis* products for *Simulium* (Diptera: Nematocera) control in a small polluted river in South Africa. *Onderstepoort Journal of Veterinary Research*, 51, 141-144 (1984).)

ARTICLE ARTIKEL

EXAMINATION OF LOCHIA AS AN AID TO THE EARLY DIAGNOSIS OF BOVINE BRUCELLOSIS

J.A. ERASMUS*

ABSTRACT: Erasmus J.A. **Examination of lochia as an aid to the early diagnosis of bovine brucellosis.** *Journal of the South African Veterinary Association* (1986) 57 No. 4, 225-226 (En) Directorate of Veterinary Services, Veterinary Laboratory, P.O. Box 625, 9500 Kroonstad, Republic of South Africa.

The standard technique for diagnosing bovine brucellosis is the serological examination of blood samples. As affected females may excrete large numbers of the *Brucella* organism in the lochia, the bacteriological and smear examination of such material appeared to be a suitable alternative for early diagnosis.

Of the lochia samples collected from 210 cows and heifers within 12–24 h after parturition or abortion, 10,9% were bacteriologically positive. Only 70% of these could be diagnosed correctly as positive by microscopic examination. This technique also resulted in 3,3% false negative and 1,4% false positive diagnoses. Results of the serological examinations of blood, collected simultaneously with the lochia samples, correlated fairly well with those obtained microscopically. Culture of lochia samples on a suitable medium appears to be the method of choice, when dealing with the early diagnosis of bovine brucellosis.

Key words: Bovine brucellosis, examination of lochia, early diagnosis.

INTRODUCTION

Brucellosis is usually introduced into a non-infected, susceptible herd by the purchase of an infected female[2,10]. When such an animal aborts or calves normally, enormous numbers of the causative organism may be shed via the foetal tissues and fluids to contaminate the environment[9,10,14].

Direct and indirect techniques for identifying an infected female are available. Of the indirect techniques, the Rose Bengal plate test (RBPT) and the complement fixation test (CFT) are the most common methods on which a diagnosis is based. The phenomenon of an infected animal aborting or calving and developing a positive CF titre 1 to 2 weeks later, is well documented[9]. Herr et al.[12], however, are of the opinion that the incidence of this feature of brucellosis might be low, indicating that serology might be an adequate route for early diagnosis.

The lochia from a brucellosis infected case may be infected with enormous numbers of the *Brucella* organism. This material might thus conveniently be used for demonstrating the presence of infection by culture[3] and smear examination[5]. When slides of lochia are stained, using the modified Ziehl-Neelsen staining technique[7], *Brucella* organisms can be detected microscopically as partially acid fast extra- and intracellular coccobacilli[5]. Unfortunately *Brucella* is not the only partially acid fast staining organism which may occur in the lochia of cattle. This property is also shared by *Chlamydia* spp. and by *Coxiella burnetti*[6]. When employing smear examination as the method of diagnosis, a number of false positive diagnoses could thus be expected. Such false positive diagnoses should, however, be eliminated by the bacteriological examination of lochia samples.

In this report the advantages of examining lochia samples from cattle by smear examination and culture, are compared with routine serological methods.

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MATERIALS AND METHODS

Lochia samples of 210 cows and heifers were collected in sterile plastic pipettes which are normally used for the artificial insemination of cattle. These samples were collected within 12 – 24 hours after calving or abortion. Duplicate smears of an aliquot of the lochia were stained by the modified Ziehl-Neelsen technique[7]. If such smears revealed partially acid-fast staining intra- and extracellular coccobacilli, measuring about 0,5 – 0,6 by 0,6 × 1,5 μm, a tentative diagnosis of brucellosis was made[4,15]. As these dimensions are within the same range as organisms of *C. burnetti*[4,13], a definite diagnosis of brucellosis could not be made by a smear examination only.

A second aliquot of a lochia sample was plated onto Farrell's medium, containing the prescribed range of antibiotics[7]. All plates were incubated at 37°C in an atmosphere of 10% CO_2. Plates were examined daily from the 4th up to the 8th day for the presence of *Brucella* colonies. Suspect colonies were identified by employing standard bacteriological techniques[1].

Blood samples of the same animals were taken simultaneously in sterile empty vacuum tubes void of any anticoagulant. Serum from these samples was subjected to the RBPT[11] and the CFT[11]. Any sample having ≥ 30/IU CF antibodies/ml was designated positive for brucellosis.

For the purpose of this study, the disease status of an individual animal was defined according to the presence of *Brucella* organisms in the lochia as revealed by bacteriological examination. False negative and false positive diagnoses on smear and serological methods, were based on the definition of Crawford & Hidalgo[8] (Table 1).

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Table 1. Definition of disease status based on bacteriological, microscopical and serological examinations

Test employed	Result	Disease status (According to culture)	
		Infected	Non-infected
Smear, RBPT	Positive	True positive	False positive
or CFT	Negative	False negative	True negative

RESULTS

All relevant results are summarized in Table 2

Table 2. The results of serological and microscopical examinations compared to the culture of lochia

Test performed	Result	Disease status No. of cases	
		Positive	Negative
Smear examination	Positive	16(7,6%)	3(1,4%)
	Negative	7(3,3%)	184
Rose Bengal plate test	Positive	15(7,1%)	110(4,8%)
	Negative	9(4,3%)	176
Complement fixation	Positive	14(6,7%)	2(1,0%)
	Negative	10(4,8%)	184
Number of cases positive on culture:		23(10,9%)	
Number of cases correctly diagnosed as positive on microscopic examination:		16/23(61%)	
Number of cases correctly diagnosed as positive on the CFT:		14/23(61%)	

According to the above definitions, 23(10,9%) of the samples examined bacteriologically were positive for *Brucella abortus* (Table 2). On smear examination, the number of positive cases diminished to 7,6% indicating that smear examinations of lochia resulted in about 70% correct positive diagnosis. This technique also resulted in 1,4% false positive and 3,3% of false negative diagnosis (Tables 1 & 2). The close correlation in the number of true positive as well as false negative diagnoses, when applying smear examination and the RBPT, is obvious (Table 2).

The CFT resulted in 6,7% positive diagnoses. This figure does not significantly differ from that found in smear examination. As in the latter case, the CFT also results in false positive and false negative diagnoses.

DISCUSSION

Accepting the general finding that bovine brucellosis is mainly spread when an infected animal calves or aborts, it stands to reason that calving in isolation would be an important step to eradication of the disease in highly susceptible herds (herds where calfhood vaccination with strain 19 vaccine were not meticulously applied). In order to relieve the pressure which may exist on an isolation system, the early diagnosis of the disease becomes an important factor. Due to the time interval which may elapse between parturition and the production of a detectable level of circulating antibodies[9], serological techniques apparently are not the most efficient method of identifying positive cases (Table 2). In fact, only 61% of infected cases could correctly be diagnosed as positively by employing the CFT on blood, collected within 24h after calving or abortion. Smear examination of the lochia and the RBPT could not improve this figure significantly. On the other hand, the number of positive cases, when the lochia samples were subjected to a bacteriological examination, increased to 10,9%.

Of the 4 techniques investigated, the bacteriological examination of lochia samples, appears to be the most sensitive technique in identifying positive brucellosis cases soon after calving or abortion.

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REFERENCES

1. Alton G G, Jones L M, Pietz D E 1975 Laboratory Techniques in Brucellosis. World Health Organisation Monograph series no 55
2. Blood D C, Henderson J A, Radostits O M 1979 Veterinary Medicine 5th edn Bailliere Tindall, London
3. Brinley-Morgan W J 1977 The diagnosis of *Brucella abortus* infection in Britain In: Crawford R.P., Hidalgo R.J. (ed.) Bovine Brucellosis, An International Symposium. Texas A & M University Press. College Station and London.
4. Brinley-Morgan W J, McCullough N B 1975 Genus *Brucella* Meyer and Shaw 1920, 173 In: Buchanan R.E., Gibbons N.E. (ed.) Bergey's Manual of Determinative Bacteriology 5th edn. The Williams and Wilkins Co., Baltimore
5. Buxton A, Fraser G 1977 Animal microbiology Vol 1 Immunology, Bacteriology, Mycology, Diseases of Fish and Laboratory Methods. Blackwell Scientific Publications, London
6. Buxton A, Fraser G 1977 Animal microbiology Vol 2 Rickettsias and Viruses. Blackwell Scientific Publications, London
7. Corbell M J, Gill K P W, Thomas E L 1978 Methods for the identification of Brucella. Ministery of Agriculture, Fisheries and Food. Research Section, Diseases of Breeding Department, Central Veterinary Laboratory, New Ham, Weybridge, Surrey KT 15 3 NB
8. Crawford R P, Hidalgo R J 1977 Diagnosis discussion In: Crawford R.P., Hidalgo R.J. (ed.) Bovine Brucellosis. An International Symposium. Texas A & M University Press, College Station and London
9. Cunningham B 1977 A difficult disease called brucellosis. In: Crawford R.P., Hidalgo R.J. (ed.) Bovine Brucellosis. An International Symposium. Texas A & M University Press, College Station and London
10. Henning M.W. 1956 Animal Diseases in South Africa. Central News Agency, South Africa.
11. Herr S, Bishop G, Bolton T F W, Van der Merwe D 1979 Onderstepoort Brucellosis Serology Laboratory Manual Veterinary Research Institute, 0110 Onderstepoort.
12. Herr S, Roux D, Pieterson P M 1982 The reproducibility of results in bovine brucellosis serology and their correlation with the isolation of *Brucella abortus*. Onderstepoort Journal of Veterinary Research, 49: 79-83
13. Moulder J W 1975 The rickettsias In: Buchanan R.E., Gibbons N.E. (ed.) Bergeys' Manual of Determinative Bacteriology. The Williams and Wilkins Co., Baltimore
14. Plommet M 1977 Experimental brucellosis in cows in France In: Crawford R.P., Hidalgo R.J. (ed.) Bovine brucellosis. An International Symposium. Texas A & M University Press, College Station and London
15. Singleton P, Sainsbury D 1978 Dictionary of Microbiology John Wiley and Sons, Chichester

RESEARCH NOTE NAVORSINGSNOTA

SCANNING ELECTRON MICROSCOPY OF *BUNOSTOMUM PHLEBOTOMUM* (RAILLIET, 1900)

F.S. MALAN*, MARYNA DE KOCK** and H.J. ELS***

ABSTRACT: Malan F.S.; De Kock M.; Els H.J. **Scanning electron microscopy of *Bunostomum phlebotomum* (Railliet, 1900).**- *Journal of the South African Veterinary Association* (1986) 57 No. 4, 227-230 (En) Hoechst Research Station, P.O. Box 124, 1320 Malelane, Republic of South Africa.

Live specimens of *Bunostomum phlebotomum* were collected from the small intestine of a calf and processed for scanning electron microscopy using standard methods. This paper describes the surface morphology of adult worms. All these hookworms showed a dorsally bent anterior end with its buccal capsule, which opens antero-dorsally with a pair of chitinous cutting plates on the ventral margin. Prominent cervical papillae and a well developed male bursa with a dorsal and 2 lateral lobes, were observed. The female tail is slender and pointed with the anal opening close to the tip. Further studies are necessary to demonstrate morphological differences from other hookworms and to obtain more detail of the male bursa and female tail.

Key words: Hookworm, *Bunostomum phlebotomum,* scanning electron microscopy, external characteristics.

INTRODUCTION

The utilization of scanning electron microscopy (SEM) in helminthology provided new information on the morphology of several helminths. A literature survey, however, revealed only a few SEM studies on hookworms[2-4], and none on the cattle hookworm, *Bunostomum phlebotomum* (Railliet, 1900).

In this paper the ultramicroscopy of this nematode is described and illustrated, with particular reference to the anterior and posterior ends of both sexes.

MATERIALS AND METHODS

About 25 adult *Bunostomum phlebotomum* were collected from the small intestine of a calf and immediately placed into physiological saline (0,85% NaCl solution). After 10 min they were thoroughly washed in sodium cacodylate buffer for 10 min at room temperature (27°C).

Specimens were fixed in 2,5% glutaraldehyde containing 0,1 M sodium cacodylate and 0,1 M sucrose at pH 7,2[1]. Thereafter the specimens were washed twice for 15 min in 0,1 M sodium cacodylate buffer with 0,1 M sucrose at room temperature. Specimens were post-fixed with 1% $0s0_4$ in 0,15 M sodium cacodylate buffer for 1 – 2 hours at room temperature. In some cases sonification followed a rinsing in 0,15 M sodium cacodylate buffer. The specimens were infiltrated with 1% tannic acid containing 0,05 M sodium cacodylate buffer at pH 7,2 at room temperature for 1 hour. This was followed by 3 rinses in 0,85% saline for 5 min each and then a 2nd incubation in 0,5% uranyl acetate in distilled water containing 0,45 g/ℓ sucrose at room temperature for 1 hour. A final rinse was carried out in saline for 5 min.

All the specimens were dehydrated in 50%, 70%, 90%, 100% (twice) acetone for 15 min in each concentration. They were then critical point dried with CO_2 in a critical point drying apparatus. The specimens were mounted on stubs and coated with a thin gold layer in a sputter coater. Specimens were examined and photographed in a Jeol JSM-35C scanning electron microscope operating at 12kV.

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RESULTS

Anterior end

An antero-lateral view of the helminth showed the marked dorsal curvature of the anterior end, typical of hookworms (Fig. 1 & 2). A fairly large buccal capsule displayed distinct ridges resembling fused lobes. On the surface of the buccal capsule, 3 symmetrically arranged lighter (less electron-dense) areas alternating with darker (more electron-dense) areas could be observed (Fig. 3 & 4).

The buccal cavity opened antero-dorsally. A pair of chitinous cutting plates were situated on the anterior rim of the buccal capsule (Fig. 3 & 4). A V-shaped depression was evident in the outer wall of the buccal capsule above the cutting plates (Fig. 3 & 4).

By removing the wall of the buccal capsule, the 2 cutting plates were found to be separate; they were semilunar in shape and appeared to be sharp at the inner edges. Some deformation could be seen beneath the collar of the buccal capsule (Fig. 3).

A closer view of the cervical area illustrates the strutted pattern of the cuticle and a protruding cervical papilla. The papilla had a conical shape with a few ridges encircling its base (Fig. 5).

Copulatory bursa

The male bursa was formed by a small dorsal and 2 large lateral lobes. These lobes were not observed in a completely expanded position. The long filiform spicule projected from the genital cone, often in a spiraling fashion (Fig. 6&7).

Female tail

The tail was slender and pointed and the slit-like anal

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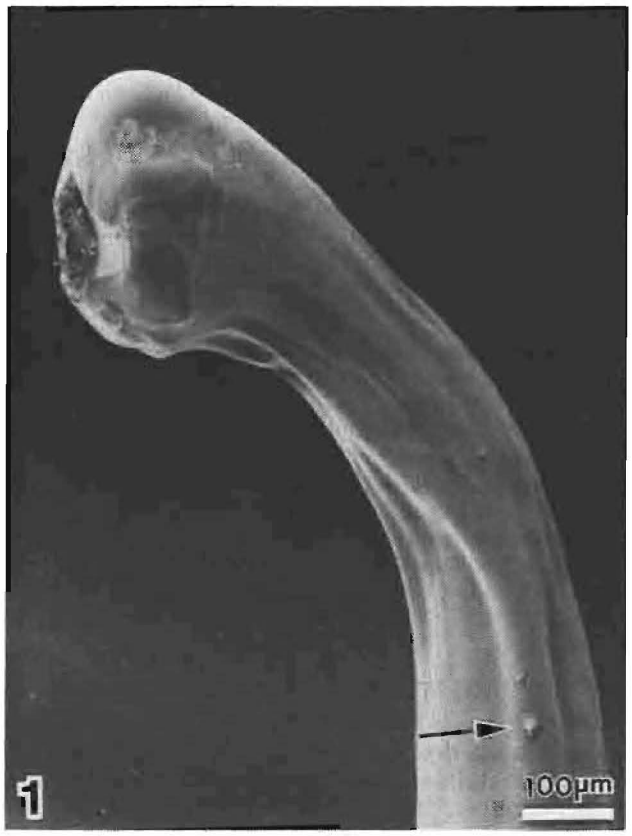

Fig. 1: Lateral view of *B. phlebotomum*. Anterior end bent dorsally giving rise to the name hookworm. Note position of cervical papilla (arrow).

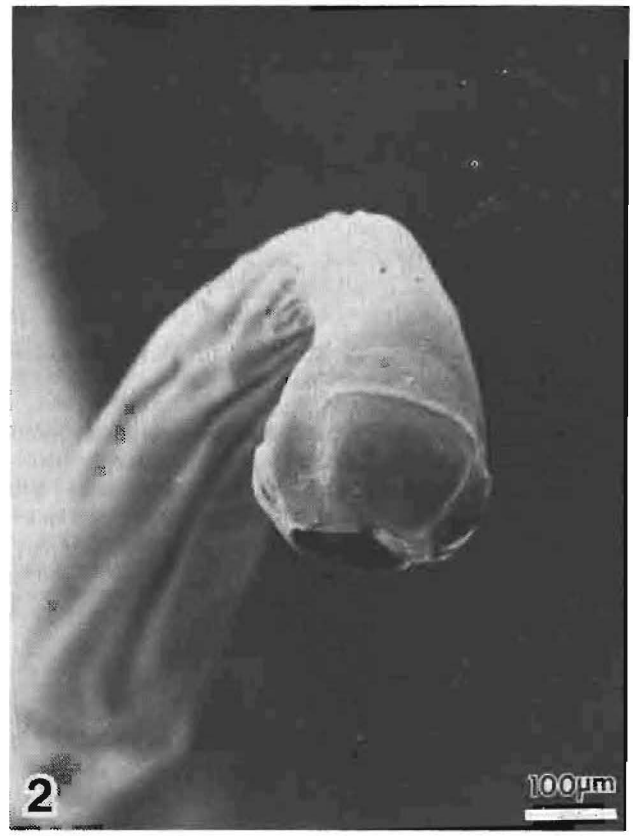

Fig. 2: *B. phlebotomum*. Anterior end, ventral view. Ridges on the external surface of the buccal cavity giving the appearance of fused lobes.

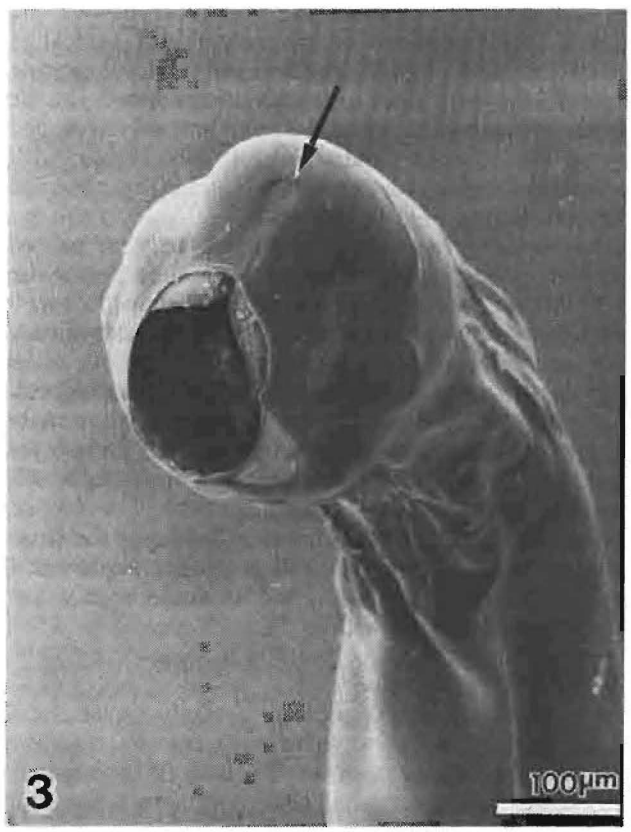

Fig. 3: *B. phlebotomum, en face* view – buccal cavity opens antero-dorsally with a pair of cutting plates ventrally. A V-shaped depression is seen on the exterior of the buccal cavity where it covers the cutting plates (arrow).

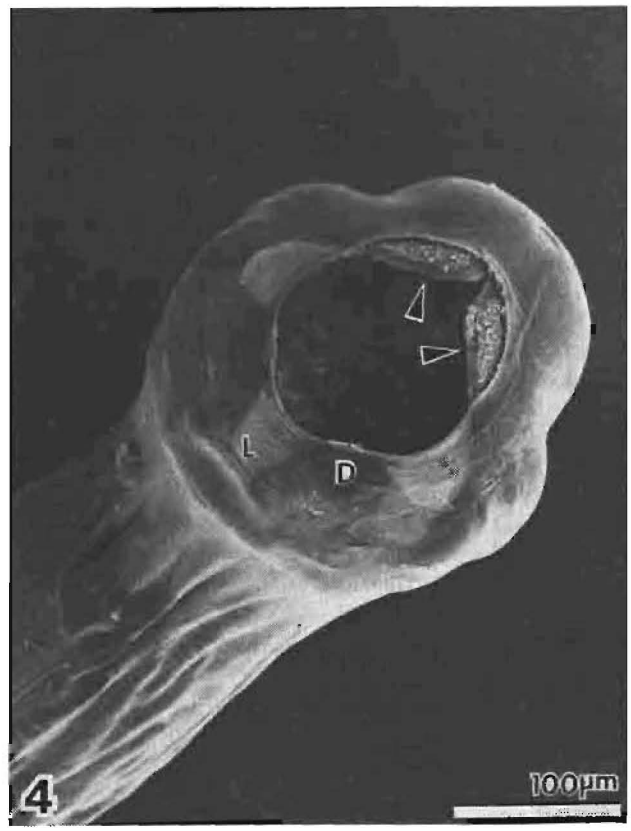

Fig. 4: *En face* view of *B. phlebotomum*. Note appearance of ventral semilunar cutting plates (arrow heads). Alternating lighter (L) and darker (D) areas can be seen on outer wall of buccal capsule.

Fig. 5: *B. phlebotomum*. A straight conical cervical papilla and straited pattern of the cuticle are shown.

Fig. 6: Copulatory bursa and spicule of *B. phlebotomum*.

Fig. 7: Copulatory bursa and spiraling spicule of *B. phlebotomum*.

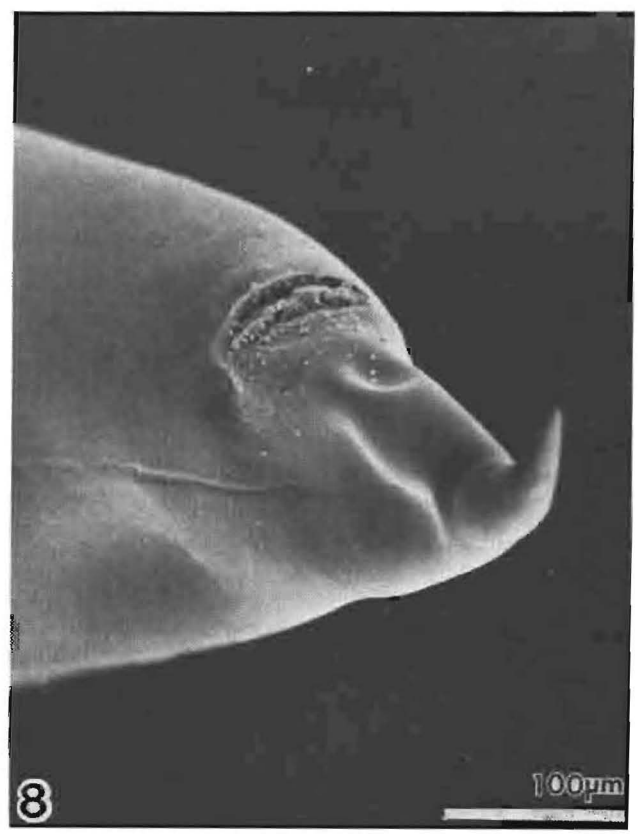

Fig. 8: Female tail and anus of *B. phlebotomum*.

opening relatively close to the tip (Fig. 8). A small amount of collapse could be observed.

DISCUSSION

The study of hookworms by means of SEM has been reported by only a few authors[2-4]. The present paper provides new information on the morphology of the adult hookworm *B. phlebotomum*. Morphological features observed include the typical dorsal curvature of the anterior end and the presence of a large buccal cavity with 2 ventral cutting plates. The cuticular ridges, giving the impression of fused lobes (Fig. 2), on the external surface of the buccal cavity and the V-shaped depression (Fig. 3) most likely form part of a flexible underlying structure. This idea is supported by the presence of the 3 lighter areas (Fig. 4, L), which reflect a stretched buccal wall when underlying structures are withdrawn resulting in an open buccal cavity.

The presence of conical cervical papillae and a regularly striated cuticle are additional features which may be important distinguishing characteristics of this particular hookworm. The semi-expanded position of the dorsal and lateral lobes of the male bursa may not represent the natural condition. A complete withdrawal of these lobes is necessary to describe the genital cone and spicule, and inner surface of the lobes. Due to the few male bursas and female tails processed in this study, further investigation is needed to determine their taxonomic importance. These SEM observations on the morphology of *B. phlebotomum*, obtained from a calf, supplement in general the available information reported for other hookworms[2-4].

ACKNOWLEDGEMENTS

The authors wish to express their appreciation to Dr J P van Niekerk, Department of Electron Microscopy, Medunsa for his advice in preparing the paper. The technical assistance of Mr G Shabangu of the Hoechst research station is gratefully acknowledged.

REFERENCES

1. Els H J, Malan F S, Scialdo-Krecek R C 1983 Scanning electron microscopy of *Strongylus* spp. in zebra. Onderstepoort Journal of Veterinary Research 50: 257-265
2. Setasuban P 1974 Scanning electron microscopy of hookworms: Morphological differences between infective stages of *Ancylostoma caninum* (Ercolani, 1859) and *Ancylostoma tubaeforme* (Zeder, 1800). Southeast Asian Journal of Tropical Medicine and Public Health 5: 519-523
3. Yoshida Y, Matsuo K, Kondo K, Arizono N, Ogino K 1974 Scanning electron microscopy of hookworms 1. Adults and infective-stage larvae of *Necator americanus* (Stiles, 1902). Southeast Asian Journal of Tropical Medicine and Public Health 5: 510-514
4. Yoshida Y, Matsuo K, Kondo K, Arizono N, Ogino K 1974 Scanning electron microscopy of hookworms 2. Adults and infective-stage larvae of *Ancylostoma duodenale* (Dubini, 1843). Southeast Asian Journal of Tropical Medicine and Public Health 5: 515-518

ABSTRACT **SAMEVATTING**

SERODIAGNOSIS OF BOVINE BESNOITIOSIS

Sera from non-infected cattle and cattle infected with *Anaplasma, Babesia, Theileria* and *Sarcocystis* were tested for antibodies to *Besnoitia* in ELISA and immunofluorescence tests (IFT) with *Besnoitia besnoiti* of blue wildebeest origin as antigen. Only 2 out of 86 sera gave false possitive reactions in ELISA and none in the IFT, indicating a high specificity for the tests.

Three-hundred-and-three bovine sera from 3 farms in an area endemic for besnoitiosis were similarly tested and the results were correlated with clinical findings based on visual inspection for typical symptoms and the presence of cysts in the scleral conjunctiva. Most of the positive tests were observed in cattle older than 1 year. Of the cases with scleral cysts, 68,7 % were positive in the ELISA and 81,74 % in the IFT. However, 45,74 % (ELISA) and 49,47 % (IFT) of the clinically negative cattle were clinically positive, indicating a high incidence of clinically inapparent infection. These results indicate a relatively low sensitivity for these serological tests.

An unexpected finding was that the ELISA remained negative for at least 60 days after experimental infection of the cattle, the maximum period for which tests were done, whereas the IFT became positive.

No antibodies against *B. besnoiti* could be found in human sera.

Besnoitia jellisoni antigen gave positive results with *B. besnoiti* antibodies in ELISA, but not in the IFT. (Janitschke, K., de Vos, A.J. & Bigalke, R.D., 1984. Serodiagnosis of bovine besnoitiosis by ELISA and immunofluorescence tests, *Onderstepoort Journal of Veterinary Research*, 51, 239-243 (1984).)

SHORT COMMUNICATION KORT MEDEDELING

RABIES IN SOUTH AFRICA: EPIDEMIOLOGICAL TRENDS FOR THE PERIOD 1980–1984

B. GUMMOW* and G.V. TURNER**

ABSTRACT: Gummow B.; Turner G.V. **Rabies in South Africa: Epidemiological trends for the period 1980 – 1984.** *Journal of the South African Veterinary Association* (1986) 57 No. 4, 231-237 (En) Department of Infectious Diseases, Faculty of Veterinary Science, University of Pretoria, Private Bag x04, 0110 Onderstepoort, Republic of South Africa.

Efficient surveillance and evaluation of data collected is a useful adjunct to a rabies prevention/control programme. Epidemiological analyses were carried out on all rabies cases reported for the period 1980 – 1984, with emphasis on the frequency and distribution in domestic and wild animals. A significant increase in reported rabies cases since 1982 was noted, with a greater prevalence in domestic animals contributing to the total increase. Rabies in domestic animals was of major importance in Natal, Gazankulu, KwaZulu, Lebowa, Qwa Qwa and Kangwane. Of all the domestic animal rabies cases reported, canines and bovines accounted for 54,5% and 33% of the cases, respectively. The genus *Cynictis* accounted for 69% of all the wild animal rabies cases reported. The greatest number of rabies cases were reported in the winter months. A definite seasonal trend was demonstrated with domestic animal rabies reaching a peak during August.

Key words: Rabies, frequency, distribution, regions, seasonal, domestic animals, wild animals.

INTRODUCTION

Rabies is a notifiable disease in the Republic of South Africa (RSA). The disease is caused by a virus belonging to the family Rhabdoviridae[1]. All mammals, including humans, appear to be susceptible. Rabies was first recorded in South Africa in 1892[3]. The disease is now known to be endemic in certain parts of southern Africa and is always a potential threat to animal and human health. The dynamic and complex nature of rabies, especially in wildlife populations, appears to have contributed to various changes in the epidemiological pattern of the disease in the region. The significance of such changes in relation to the prevention and control of rabies can only be determined by an efficient surveillance programme and the continual evaluation of all data reported.

The aim of this study was to analyse recent rabies data to determine any epidemiological trends relative to the three major epidemiological variables, namely, populations at risk, temporal and spatial distributions of disease.

MATERIALS AND METHODS

Data concerning reported rabies cases were obtained from the records of the Veterinary Research Institute, Department of Agricultural and Water Supply, and the Epidemiology Section, Department of Health and Welfare. Epidemiological analyses were carried out on all rabies cases reported for the period 1980 – 1984. For this study a rabies case was one where brain specimens from affected animals were diagnosed as positive for rabies by one of the following routine methods:

(a) Indirect fluorescent antibody technique
(b) Biological test in 7-day old mice
(c) Histopathological identification of Negri bodies

The animal populations included in this study were divided into domestic and wild animals. These were further subdivided into genera according to the record system adopted by the authorities from which the data was obtained. Domestic animals were recorded as follows: *Canis, Bos, Felis, Ovis, Equus, Capris* and *Sus*. The wild animal rabies cases included the following genera: *Cynictis, Genetta, Suricata, Herpestes, Otocyon* and *Canis*. The number of rabies cases reported and their distribution within these various groups were determined.

Data relating to the temporal distribution of rabies cases within the various populations considered was used to determine the prevalence of reported cases on a monthly and annual basis for the period 1980 – 1984. Analyses pertaining to the spatial distribution of the disease was confined to determining the occurrence of reported rabies cases in the following regions: Cape Province, Natal, Orange Free State (OFS), Transvaal, Gazankulu, KwaZulu, Lebowa, Qwa Qwa and Kangwane. In addition, mortality rates per 100 000 animals were calculated for each region. Mortality rates could only be determined for domestic animals. In calculating the mortality rates the only total animal numbers available for the numerical denominators were those obtained from the 1984 Livestock Census, Department of Agriculture and Water Supply.

Correlations were made between the epidemiological variables studied in order to ascertain whether any significant epidemiological trends existed, with special reference to the frequency and distribution of reported rabies cases.

RESULTS AND DISCUSSION

The total number of rabies cases in animals reported for the period 1980 – 1984 is presented in Fig. 1. There was an increase in the number of cases reported in 1984 in comparison to the preceding three years. This demonstrates that rabies is still an important potential health hazard to animals and humans. These figures only represent the reported cases. From a practical standpoint it can be assumed that the actual number of rabies cases occurring in South Africa is in excess of those

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0038-2809/86/04/0231-0237

reported. Correspondingly there was an increase in rabies cases reported in domestic animals (Fig. 2). This suggests that the total increase of rabies cases can be attributed to the increase in rabies in domestic animals. Such a trend is considered to be of major public health significance.

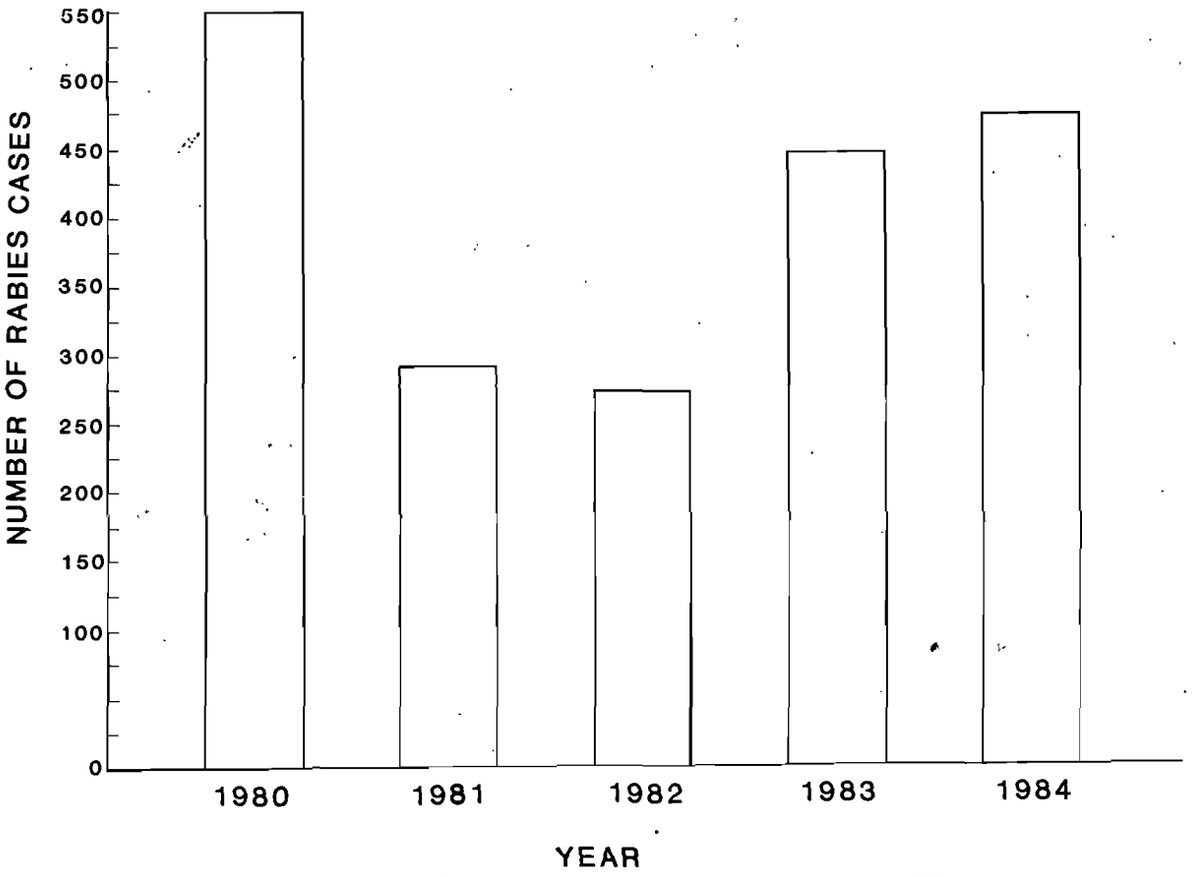

Fig. 1: Total number of rabies cases in animals reported for the period 1980-1984.

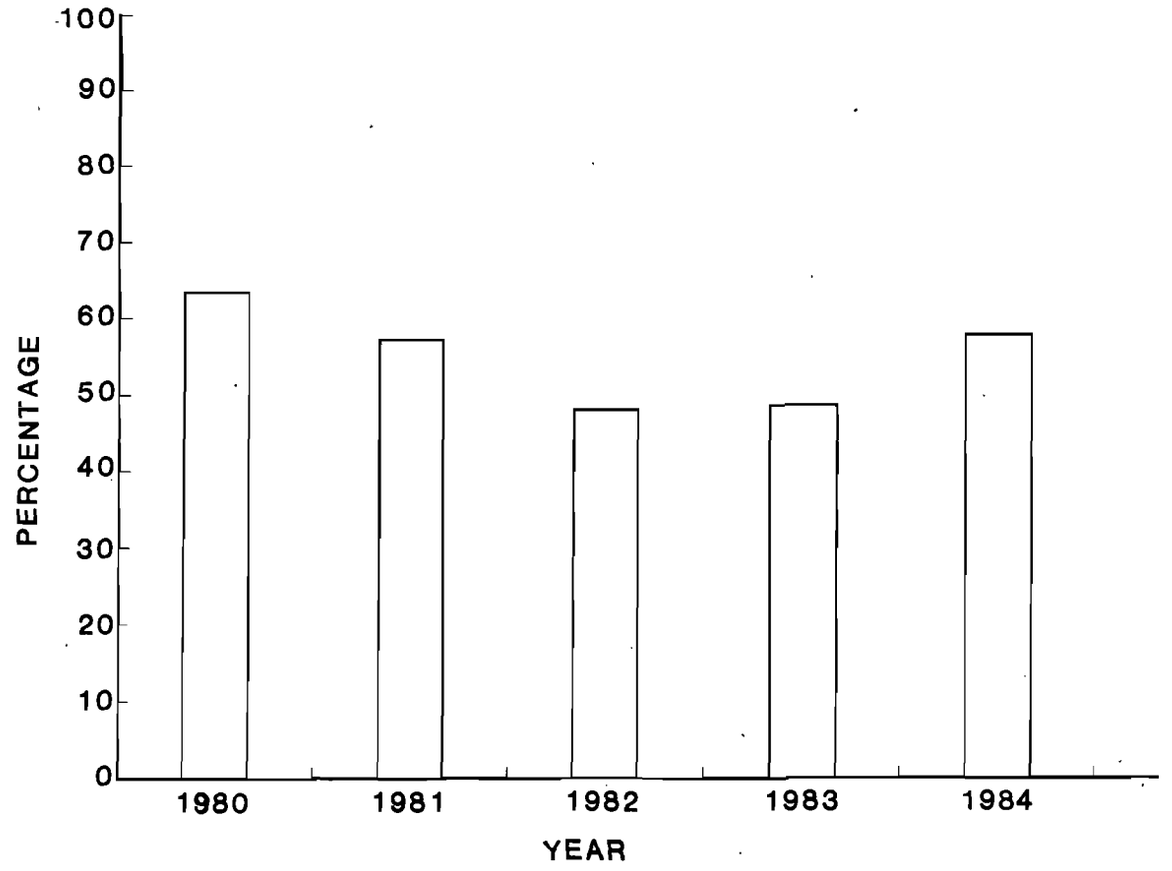

Fig. 2: Number of rabies cases in domestic animals expressed as a percentage of the total number of animal rabies cases reported for the period 1980-1984.

The importance of domestic animals in the epidemiology of rabies was compounded further when the frequency of cases was analysed according to the various regions included in the study (Fig. 3). In Gazankulu, KwaZulu, Lebowa, Qwa Qwa and Kangwane the reported rabies cases were almost exclusively confined to domestic animals. In Natal the majority of cases reported were domestic animals. For the period 1981 through 1984 there was an increase in the number of domestic animal rabies cases reported in the OFS, whereas in the Transvaal there was a noticeable decrease. With regard to reported rabies cases, Natal had the highest mortality rate per 100 000 domestic animals (Fig. 4). An increase in mortality rates was noted in the OFS and KwaZulu.

These three regions also had the highest mortality rates due to rabies in humans during 1984 (Gummow B. unpublished data). In the Transvaal there was a definite decrease in the rabies mortality rate in domestic animals for the period 1980 – 1984. There were no reported cases of rabies in humans during this period (Gummow B. unpublished data).

Of all the domestic animal rabies cases reported canines and bovines accounted for 54,5% and 33% of the cases, respectively. The distribution of rabies cases in the different genera of domestic animals is presented in Fig. 5. The dramatic increase in canine cases reported for 1984 is of epidemiological significance when correlated with the findings shown in Fig. 1 & 2.

Epidemiological data on wildlife rabies up to 1980 was analysed by Meredith[2]. In this study the results of analysing the distribution of reported rabies cases in the different genera of wild animals for the period 1980 – 1984 is presented in Fig. 6. The family Viverridae was the most important group affected by rabies, accounting for 77% of all wild animal cases reported. In this study the Viverridae, which are the mongooses, included the following genera: *Cynictis, Genetta, Suricata* and *Herpestes. Cynictis,* the yellow mongoose, accounted for 69% of the wild animal cases reported. The number of Viverridae rabies cases in relation to the different wild animals included in this study was in agreement with Meredith's findings[2] prior to 1980.

The family Canidae was the other important wild animal group. This included the genera *Otocyon* (bat-eared fox) and *Canis* (jackals). The Canidae accounted for 17% of the wild animal rabies cases reported. The genus *Otocyon* appears to be playing a greater role in the epidemiology of wildlife rabies. The reason for this is difficult to explain. Of the wild animal rabies cases reported *Otocyon,* and *Canis* accounted for 13% and 4% of the cases, respectively. Other wild animal types played a lesser role and only accounted for 6% of the cases reported.

When comparing the frequency and distribution of rabies in domestic and wild animal populations, it was found that the genera *Cynictis, Bos* and domestic *Canis* accounted for 80% of all rabies cases reported (Fig. 7). Meredith[2] suggested that species predominance in a given area was linked to habitat and that species with a

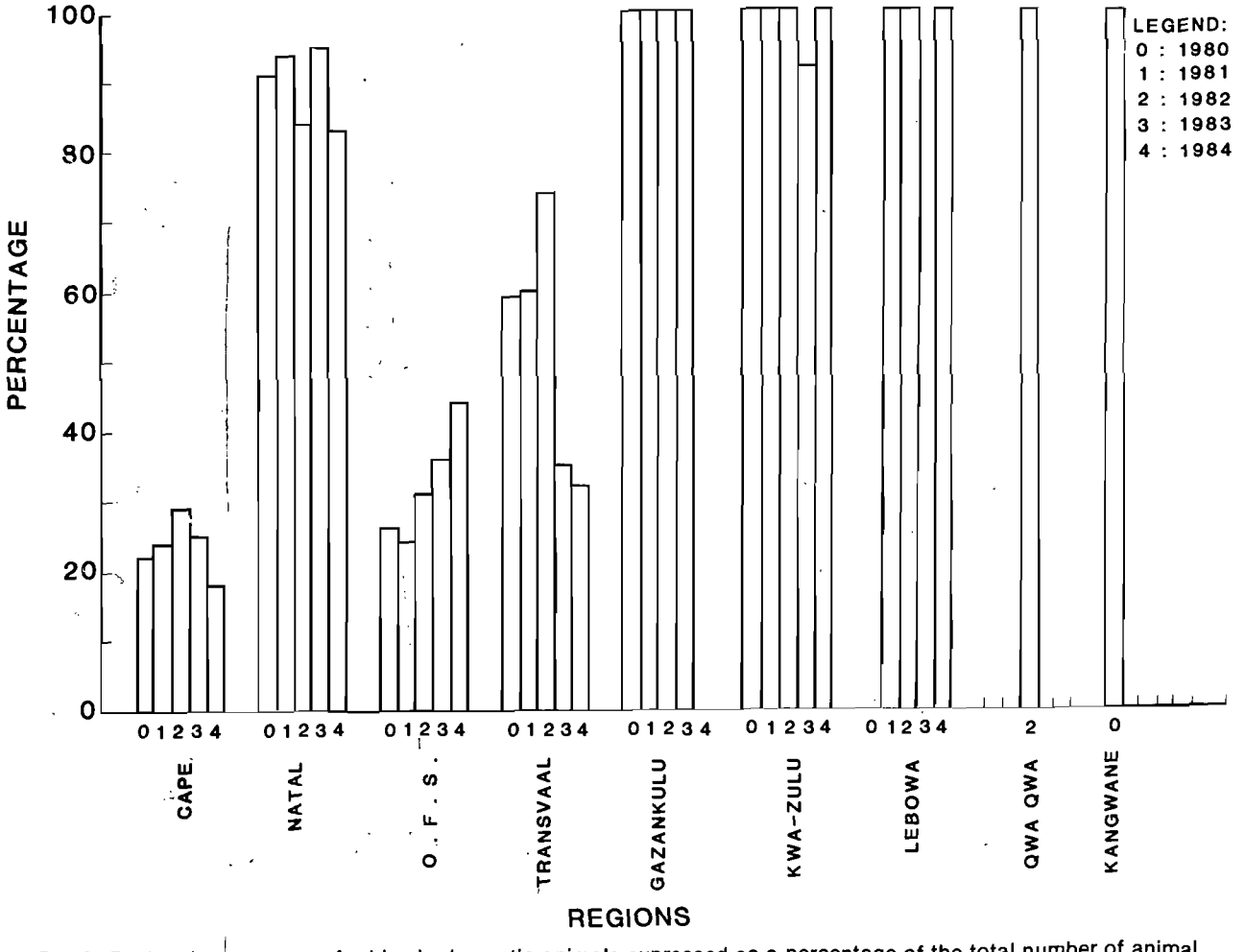

Fig. 3: Regional occurrence of rabies in domestic animals expressed as a percentage of the total number of animal rabies cases reported for each region (1980-1984).

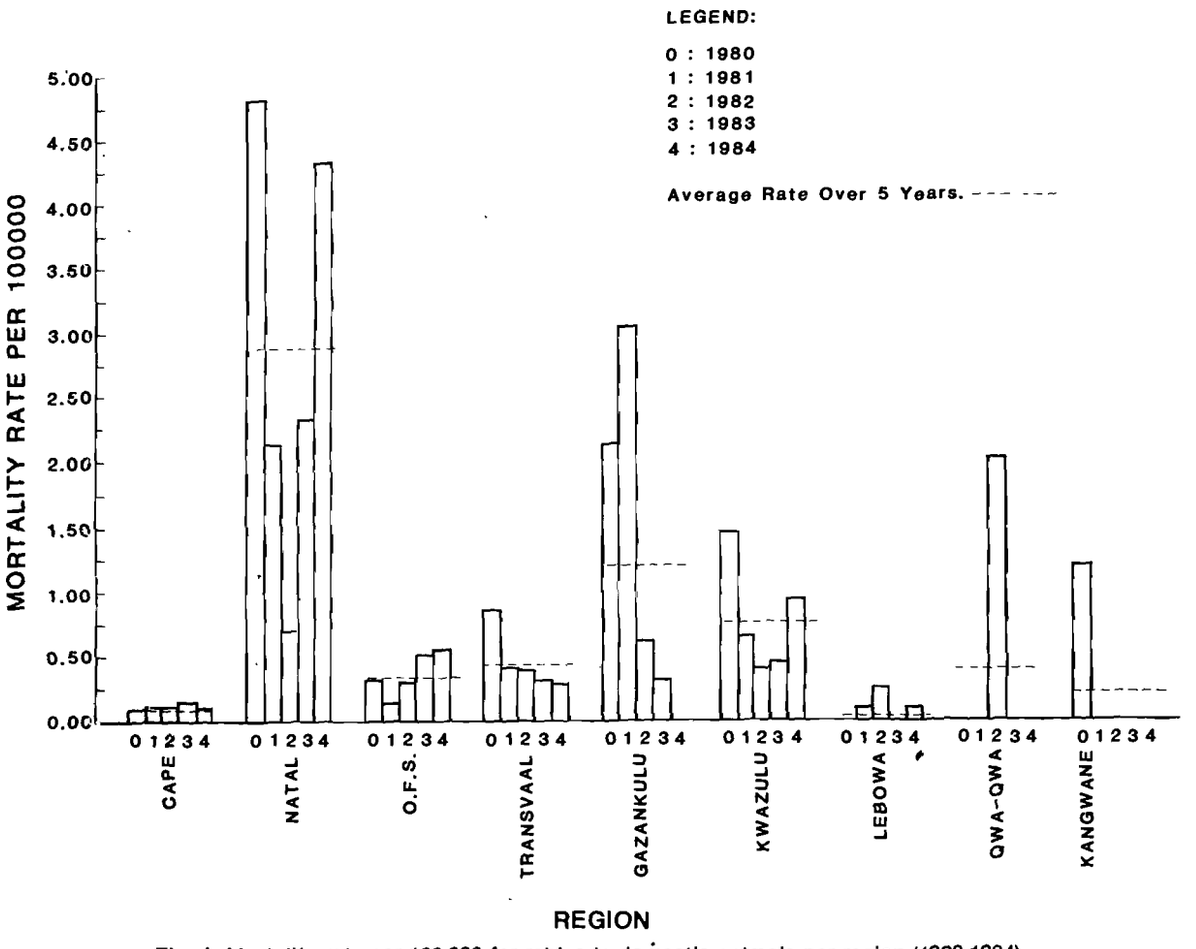

Fig. 4: Mortality rate per 100 000 for rabies in domestic animals per region (1980-1984).

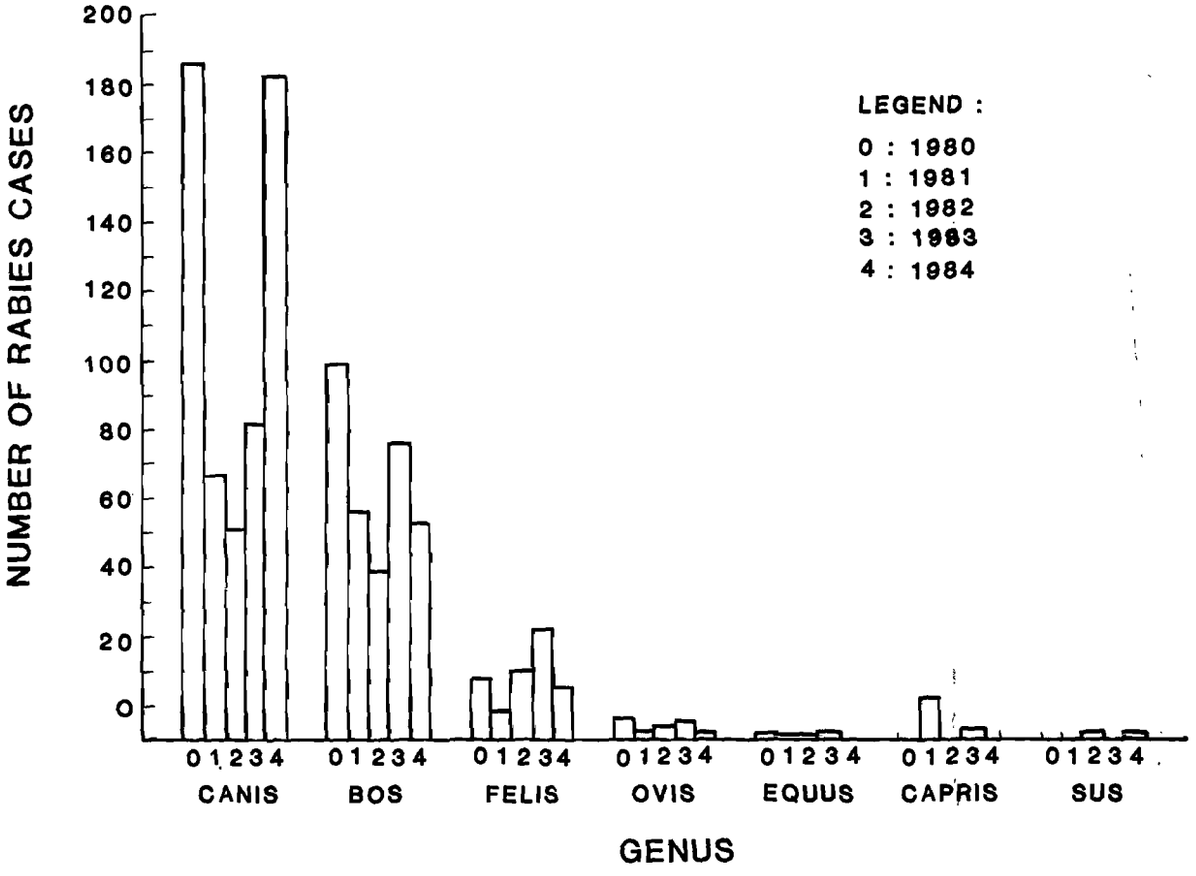

Fig. 5: Distribution of rabies cases in different domestic animals for the period 1980-1984.

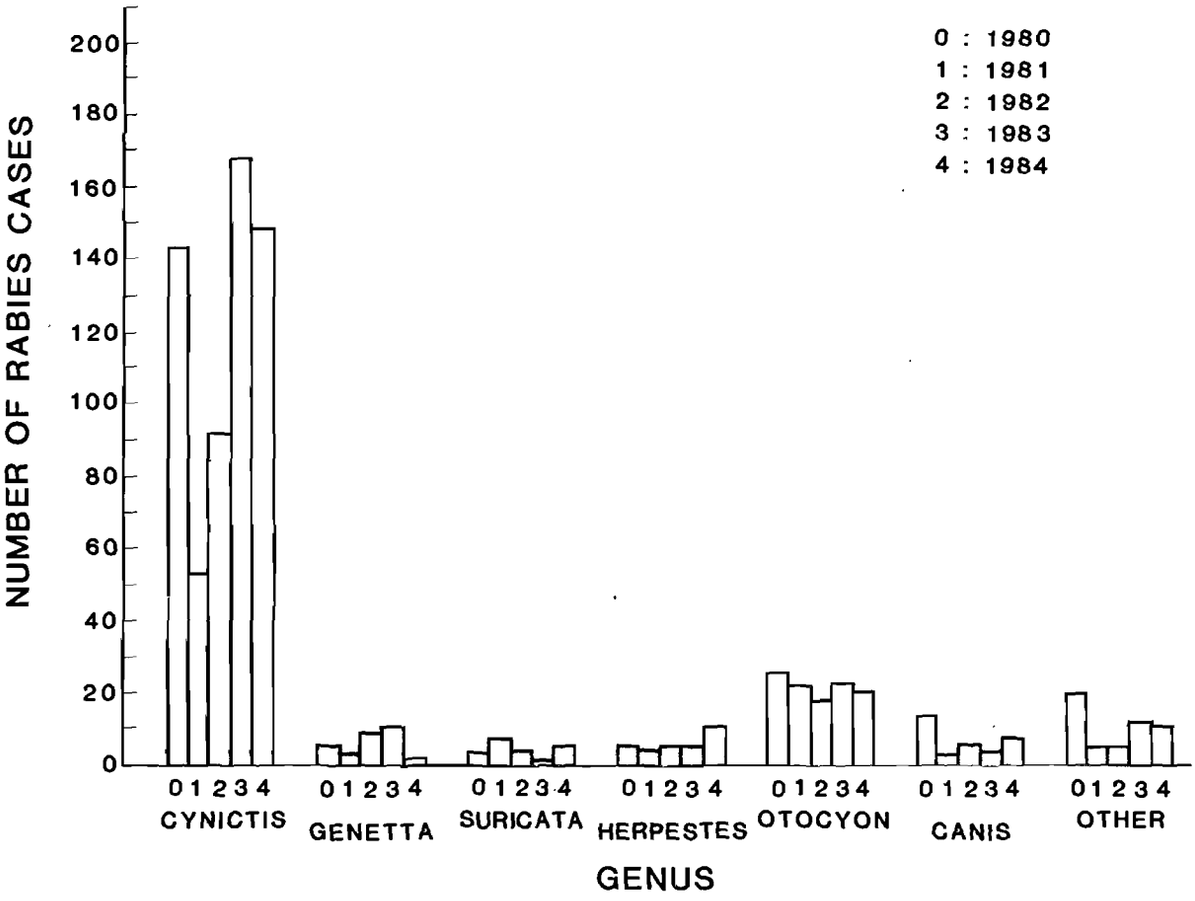

Fig. 6: Distribution of rabies cases in different wild animals for the period 1980-1984.

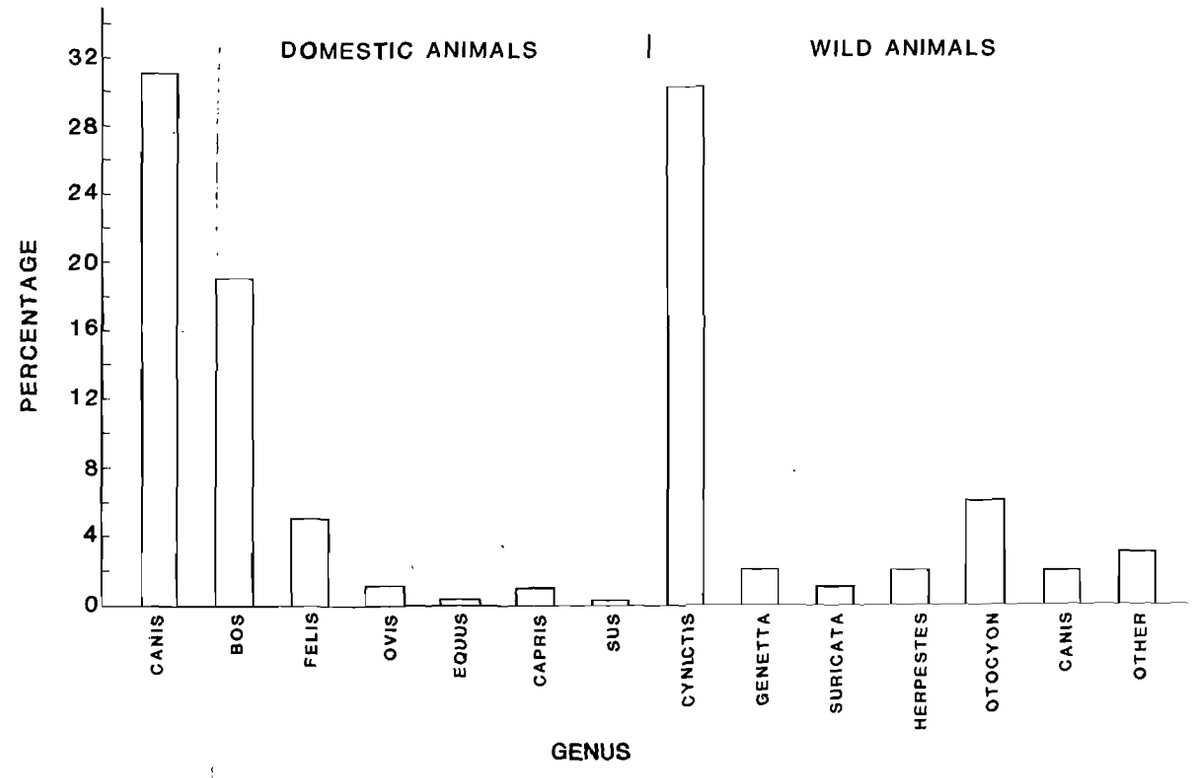

Fig. 7: Number of rabies cases in different animal genera expressed as the mean percentage of the total number of animal rabies cases reported for the period 1980-1984.

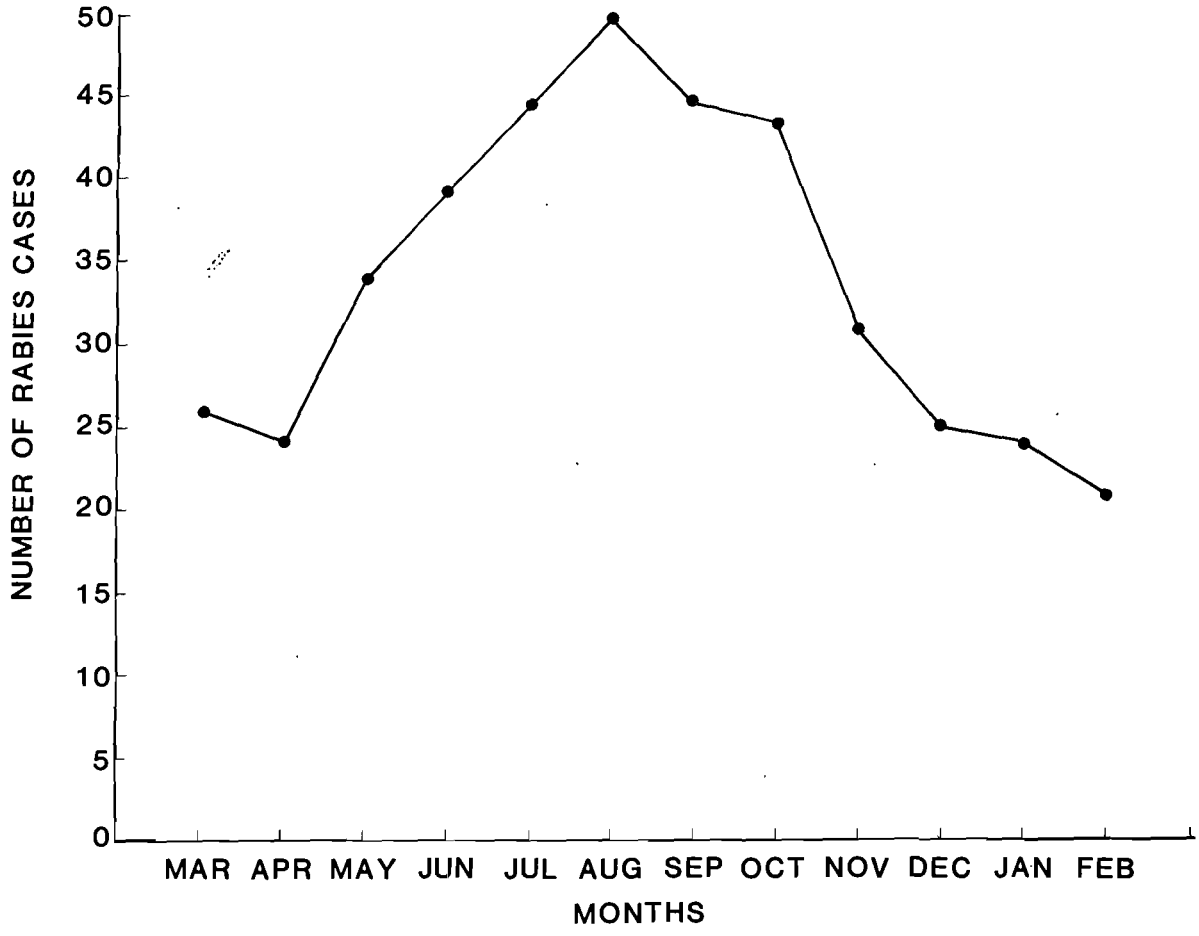

Fig. 8: Average monthly occurrence of rabies in animals reported for the period 1980-1984.

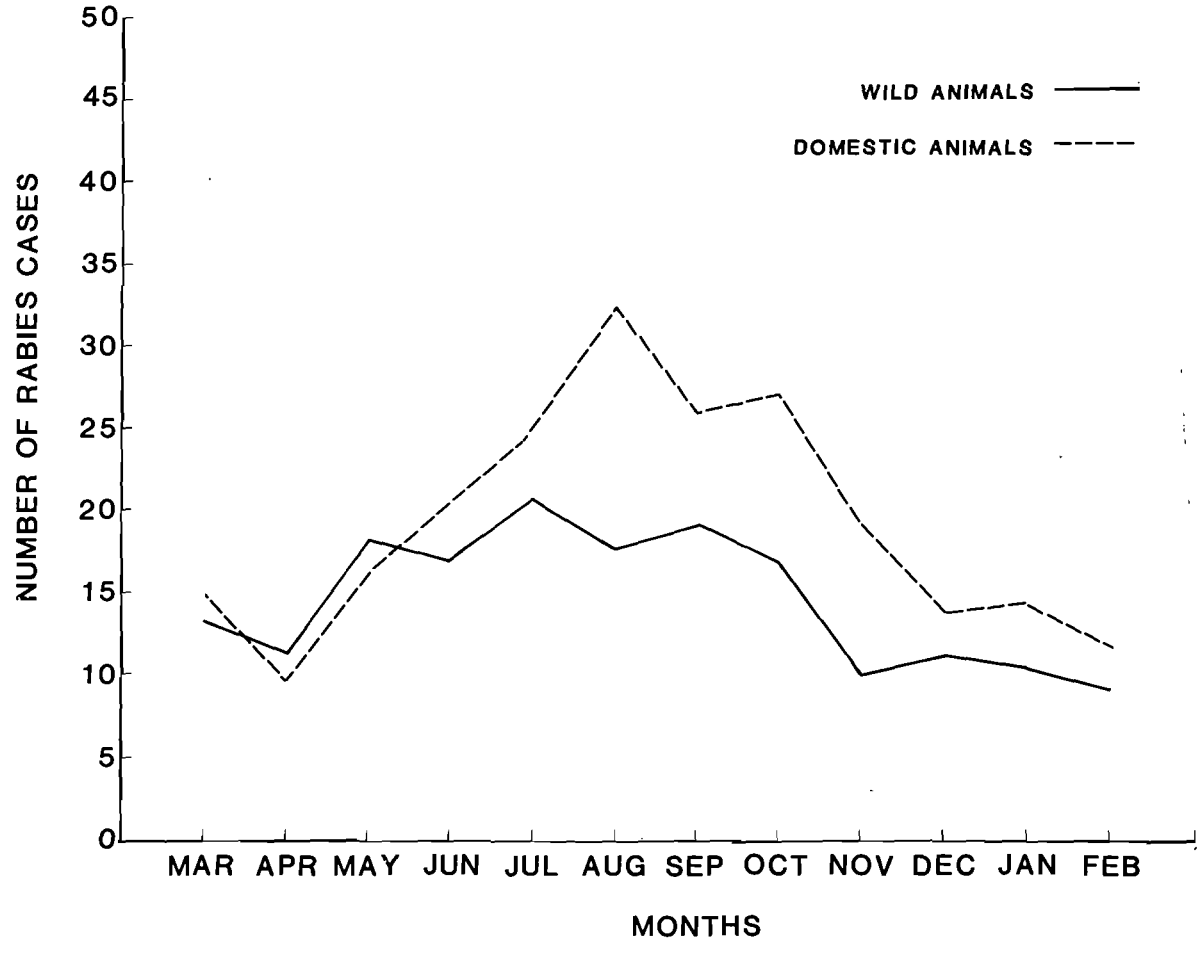

Fig. 9: Average monthly occurrence of rabies in domestic and wild animals reported for the period 1980-1984.

high degree of habitat specificity were important as a source of rabies only over a relatively small part of their distribution range. Conversely those with less habitat specificity were more widely distributed and therefore played a more significant role as a source of rabies over a wider area[2]. The genus *Bos* has a relatively high habitat specificity and therefore is unlikely to play a significant role in the epidemiology of rabies. Thus of the three genera mentioned above, *Cynictis* and *Canis* are possibly the most likely animals to contribute to the perpetuation of rabies in South Africa.

This study also demonstrated that rabies exhibits a definite seasonal trend (Fig. 8 & 9). There was a consistent increase in the number of rabies cases reported during the winter months with a peak in August. The increase in wild animal rabies cases occurred from May to October (Fig. 9), whereas the increase in domestic animal cases only began in July with a pronounced peak in August.

It is therefore suggested that the initial rise in wild animal rabies is due to increased animal population densities as a result of the colder climatic conditions and the shortage of food and water. In addition, behavioural patterns of *Cynictis* may also contribute to this seasonal increase[4]. In the winter months the young mongooses are evicted from their parents' territory. The young mongooses are then forced to scatter over a wider area because brother-sister mating does not occur[4]. The opportunity arises for the wide-spread distribution of rabies with a concommittant increase of the disease in the wild animal population. The probability of domestic animals coming in contact with rabid animals is thus enhanced, which possibly explains the increase in domestic animal rabies cases at a slightly later stage.

A greater chance therefore exists for humans to come in contact with rabid animals, especially domestic animals, during the winter months. It has been shown that the majority of reported human contact cases with rabid animals, occurs in the winter months (Gummow B. unpublished data)

CONCLUSIONS

Rabies is still an important health hazard to animals and humans. Domestic animals are playing an increasingly important role as a potential source of rabies to susceptible populations. This is especially of significance in Natal, Gazankulu, KwaZulu, Lebowa, Qwa Qwa and Kangwane and during the winter months.

This study demonstrated some subtle but important epidemiological changes in the frequency and distribution of rabies in South Africa during the period 1980 – 1984. The epidemiological trends regarding the populations at risk and the seasonal and regional occurrence of rabies should be of interest to those involved in the prevention and control of rabies in South Africa.

ACKNOWLEDGEMENTS

The authors wish to express their appreciation to Dr H Küstner and the members of the Epidemiology Section, Department of Health and Welfare for their assistance, advice and use of their computer facilities.

REFERENCES

1. Howell P G 1982 Rabies and rabies-like viruses. South African Journal of Science 78: 409-411
2. Meredith C D 1982 Wildlife rabies. Past and present in South Africa. South African Journal of Science 78: 411-415
3. Snyman P S 1940 The study and control of vectors of rabies in South Africa. Onderstepoort Journal of Veterinary Science 15: 9-140
4. Zumpt I 1982 The yellow mongoose as a rabies vector on the central plateau of South Africa. South African Journal of Science 78: 417-418

BOOK REVIEW **BOEKRESENSIE**

CLINICAL LABORATORY ANIMAL MEDICINE

DONALD D. HOLMES

1st Edn. The Iowa State University Press, Ames, Iowa, 50010. 1984 pp v and 138. Price R33.00 (ISBN 0-8138-0328-4).

This book has been written to provide a convenient reference manual on the biology, husbandry, diagnosis and treatment of disease in laboratory mammals and reptiles. It comprises chapters on mice, rats, Mongolian gerbils, guinea pigs, rabbits, ferrets, non-human primates and aquatic and terrestial reptiles. These are supplemented with appendices on normal biological data on reproductive, physiological, haematological and biochemical features of the species covered. The book is supported with source references for much of the information which it contains.

It serves as an excellent introductory text for persons who are entering laboratory animal science as technicians or clinicians or for veterinarians who have to treat small mammals, rodents and reptiles in their practices.

The book has been reproduced from a typewritten manuscript and presented in a ring binder format on good quality paper.

While it is not intended to be a comprehensive source of information on the species it covers, it is a valuable source of information in a condensed form for biomedical science students, laboratory animal technologists and veterinarians who are involved with laboratory animal care.

J.C. Austin.

BOOK REVIEW **BOEKRESENSIE**

CANINE ANATOMY

DONALD R. ADAMS

1st Edn. The Iowa State University Press, Ames, Iowa 1986 pp viii + 513, illustrations 527 Price £29.50 (ISBN 0-8138-0281-4)

The book is intended as a guide for the systematic dissection of the dog for first year veterinary students. It deals with the superficial morphology, musculoskeletal, respiratory, digestive, urogenital, cardiovascular, lymphoreticular and peripheral nervous systems. The brain is not included. No dissection instructions are provided. The annotations for the line drawings are part of the text and do not appear as separate legends. Not all the illustrations are technically of a very high standard. However, it is refreshing to see new approaches to known structures in the hundreds of original drawings. Helpful explanations of anatomical terminology are given, making the text very accessable to undergraduate students. A number of useful differences between the dog and cat are mentioned where appropriate.

The book is not intended to be a textbook. Neither is it a true dissection guide as no dissection instructions are included. In veterinary schools such as ours, where anatomy is taught comparatively, this expensive study guide cannot be recommended without reservation.

Malie S. Smuts

CASE REPORT **GEVALVERSLAG**

THE USE OF METHYL METHACRYLATE BONE CEMENT AS AN INTERNAL SPLINT IN THE TREATMENT OF FRACTURES OF THE CANINE AXIS

D. G. STEYN*

ABSTRACT: Steyn D G. **The use of methyl methacrylate bone cement as an internal splint in the treatment of fractures of the canine axis.** *Journal of the South African Veterinary Association* (1986) 57 No. 4, 239-241 (En) Department of Surgery, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, 0110 Onderstepoort, Republic of South Africa.

Three dogs with fractures of the body of the axis showing moderate to severe displacement were presented for treatment. The fractures were exposed, screws were fixed into each fragment and the heads of the screws were embedded in bone cement to serve as an internal splint. The surgical approach and technique are described.

Key words: Methyl methacrylate, axis fractures, bone cement, dogs.

INTRODUCTION

Fractures of the cervical vertebrae are uncommon compared to fractures of other bones, particularly the bones of the extremities. In a survey of 126 cases of blunt spinal trauma, only 5,5% involved fractures of the cervical vertebrae[3].

Stone et al.[8] reviewed cases of cervical vertebral fractures of which 21 (78%) had fractures of the axis. Denny[2] reported 9 fractures of the cervical vertebrae of which 4 involved the axis. Cervical fractures, therefore, frequently involve the axis with the most common site being the dens and/or the vertebral body.

The majority of the reported cases were treated surgically. Denny[2] used plaster casts to immobilize the necks of patients presenting displaced and unstable fractures. Wire and plates in combination with laminectomy, hemilaminectomy and removal of the dens were used for internal fixation. Rouse[7] described a method in which 4 pins were inserted into the vertebral body, 2 cranial and 2 caudal to the fracture. The ends of the pins were left protruding about 1 cm and incorporated in bone cement to serve as an internal splint.

This report describes fracture of the body of the axis in 3 dogs treated by internal fixation with screws and bone cement.

SURGICAL TECHNIQUE

The patient is positioned in dorsal recumbency with the neck extended and surgically prepared. The approach to the vertebra is similar to a technique previously described[1]. A midline incision extends from immediately cranial to the larynx to a point midway between the larynx and the manubrium. The right sternocephalic muscle is retracted to the right, and all the structures ventral to the longus colli muscles are retracted to the left. The longus colli muscles will now be exposed. Haemorrhage in the musculature may be evident in the region of the fracture. The longus colli muscle is then severed at its implantation on the ventral vertebral process and reflected laterally with the aid of a periosteal elevator. With the ventral surfaces of the atlas and axis now exposed, the fracture may be reduced and stabilized.

On completion of these procedures, the anatomical structures are pushed into position. A few interrupted sutures of absorbable material may be used to bring the reflected longus colli muscles into position. The subcutaneus tissue and the skin are sutured in a routine manner.

Postoperative care consisted of cage rest, antibiotics and analgesics for 3 days. Intravenous fluids were administered prior to and during the surgical procedure.

CASE REPORTS

The 3 cases reported here, were referred for treatment to the Surgery Department of the Faculty of Veterinary Science, University of Pretoria. Clinical, neurologic and radiologic examinations were performed to establish the diagnosis.

Case 1

An 8½-month-old male Bull Terrier-cross was presented 2 days after an automobile accident. Cervical pain, ataxia of the hind legs and proprioceptive deficits of all four limbs were present. The animal resisted manipulation of the neck. Radiographs revealed a fracture of the body of the axis with dorsal displacement of the caudal fragment. The proximal fragment, however, was not displaced and the dens was in its normal position (Fig. 1).

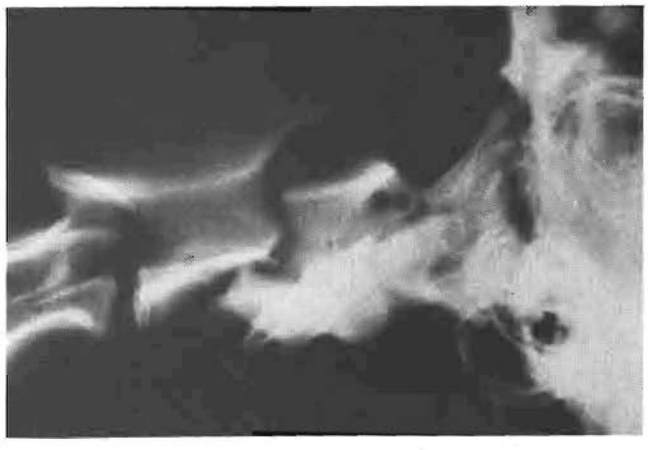

Fig. 1: Fracture of the axis. The caudal fragment is displaced in a cranial direction with overriding of the fragments.

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The fracture was exposed in the manner described, and an attempt was made to reduce it. Although it was not possible to achieve perfect reduction, the position of the fragments was improved considerably. A hole was drilled from the ventral surface of the cranial fragment extending into the dens. The hole was tapped, and a 3,5 mm cortical screw was inserted with the head protruding about 8 mm. Another screw was placed in the caudal fragment. In order to maintain reduction, orthopaedic wire was looped around the head of the screw to exert traction on the vertebra.

While the partial reduction was maintained with the end of the wire loop, the area between and surrounding the two screw heads was filled with methyl methacrylate bone cement. During the setting time, the area was irrigated with cold saline to dissipate the heat of polymerization. When the cement had set, the wire was cut off. The remaining piece can be seen in the postoperative radiograph (Fig. 2).

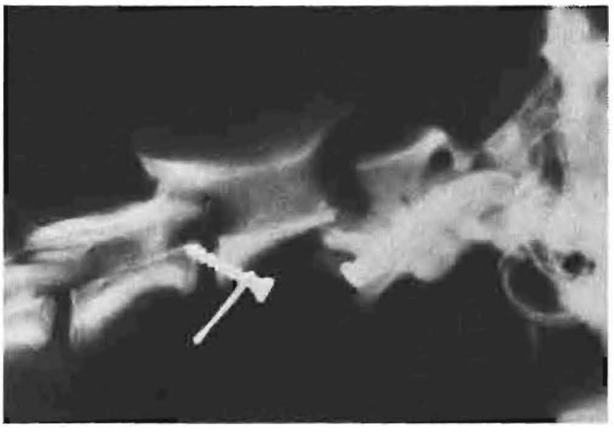

Fig. 2: The fracture incompletely but sufficiently reduced. The piece of wire around the head of the screw and embedded in the bone cement is clearly visible.

Case 2

A 9-month-old female German Shepherd with acute cervical pain and discomfort of unknown aetiology, was referred by a private practitioner. Radiographs of the neck were taken, and a transverse fracture of the axis was diagnosed (Fig. 3).

A screw was put into the cranial fragment and anchored in the base of the dens. Another screw was placed in the caudal fragment as described in Case 1. The 2 screw heads were again incorporated in the bone cement to maintain reduction (Fig. 4).

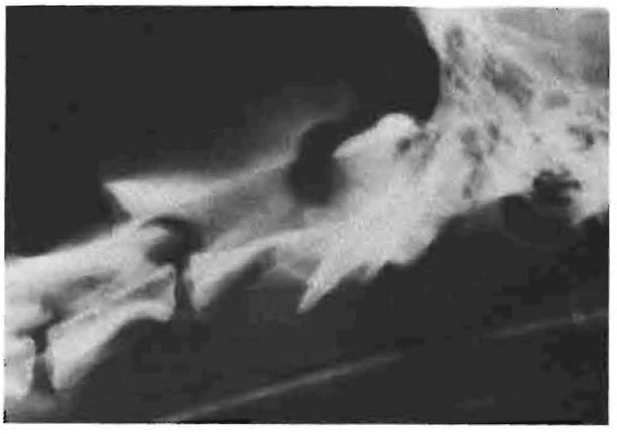

Fig. 3: Fracture of the axis. Overriding is evident at the ventral surface of the axis.

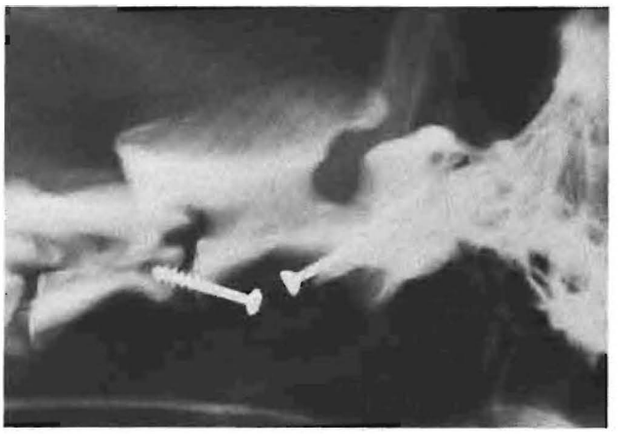

Fig. 4: Good reduction of the fracture has been achieved with the two screw heads incorporated in bone cement.

Case 3

A 2-year-old male Corgi, hit by a car, sustained a fracture of the cranial part of the body of the axis (Fig. 5). Severe cervical pain, front limb paresis and loss of proprioception were present. The animal resisted manipulation of the neck and cried out in pain when it was forced to move.

Fig 5: Fracture of the axis with dorsal displacement of the caudal fragment. Note the degree of displacement at the ventral surface of the axis.

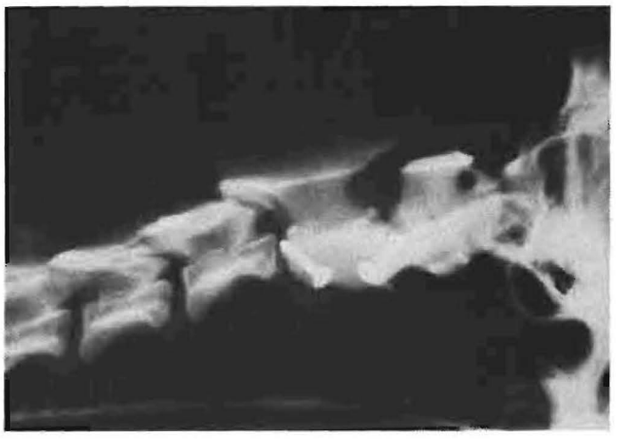

Fig. 6: Anatomical reduction has been achieved. The mass of bone cement is radiopaque because of the presence of barium mixed with the bone cement.

The ventral surface of the axis was exposed, the fracture reduced and screws placed as described. Barium sulphate was mixed in with the bone cement before it was packed into the area incorporating both screw heads. The bone cement is clearly visible on the postoperative radiograph (Fig. 6).

RESULTS

Internal fixation had a dramatic postoperative effect on the patients. In all 3 cases, the animals moved freely with very little obvious discomfort, although manipulation of the neck was still resisted to some degree. The general behavior of the animals improved markedly following surgery.

DISCUSSION

Besides the use of methyl methacrylate for the implantation of prostheses, many adjunctive uses of this substance have been described. These include the treatment of comminuted trochanteric femoral fractures[4], acetabular fractures[6], multiple fractures[9] and cervical fractures[7]. In addition, experimental spinal instabilities following dorsal laminectomies were successfully stabilized by placing Steinmann pins and embedding them in bone cement[10]. Although temperatures of 104°C to 107°C develop within the bone cement and 75°C to 85°C at the bone interface, necrosis extending only approximately 0.5 mm into the bone, occurs when the bone cement sets in contact with bone[9]. The heat of polymerization can, however, be dispersed effectively by irrigating the area with a cold saline solution[10].

Rouse[7] described a technique in which 2 Steinmann pins, wire and plates have also been used in the repair of fractures of the axis[2].

A ventral approach is necessary to expose the body of the axis and to achieve proper anatomical positioning of the fragments for accurate screw placement. Rouse[7] also stressed the danger of haemorrhage from the vertebral sinuses which can occur when the fragments are manipulated. Respiratory arrest may be the end result if extensive haemorrhage occurs in the neural canal.

The screws must not protrude into the vertebral canal and should, therefore, be placed with great care. Two screws on either side of the fracture line would give better anchorage and would conform to basic orthopaedic principles. This would have been technically difficult as a result of the small size of the cranial fragment and the lack of sufficient bone substance in that area. Postoperative loosening of the screws is possible after a few weeks. In the clinical cases reported here, this did not happen before clinical healing was completed.

Complete reduction is not always possible in spite of all efforts as can be seen in Case 1. The cervical spinal canal is large relative to the spinal cord. Compression of the cord does not occur if displacement of fractures is not excessive. Perfect anatomical reduction, therefore, is not essential provided that the pressure on the spinal cord has been relieved and reduction is maintained to prevent movement and further damage to the cord. The screws and the bone cement serve as an internal splint which maintains the reduction and provides good internal stability. This method will not replace other methods, but it can be successfully used in cases of fracture of the vertebral body, particularly fractures of the axis.

REFERENCES

1. Cechner P E 1980 Ventral cervical disc fenestration in the dog: A modified technique. Journal of the American Animal Hospital Association 26: 647-650
2. Denny H R 1983 Fractures of the cervical vertebrae in the dog. Veterinary Record 23; 236-240.
3. Feeney D A, Olivier J E 1980 Blunt spinal trauma in the dog and cat: Insight into radiographic lesions. Journal of the American Animal Hospital Association 16: 885-890
4. Lau H K, Lee P C, Tang S C, Lin J T K, Chow S P 1983 Treatment of communicated trochanteric femoral fractures with Dimon Hugston displacement fixation and acrylic cement – a preliminary report of sixteen cases. Injury 15: 129-135
5. Park J B, Rouse G P, Kenner G H, Gendrean C L 1978 Spinal fixation using acrylic bone cement: mechanical property measurements. Biomaterials, Medical Devices and Artificial Organs 6: 77-84
6. Renegar W R, Griffiths R C 1977 The use of methyl methacrylate bone cement in the repair of acetabular fractures. Journal of the American Animal Hospital Association 13: 582-588
7. Rouse G P 1979 Cervical spine stabilization with methylmethacrylate Veterinary Surgery 8: 1-4
8. Stone E A, Betts C W, Chambers J N 1979 Cervical fractures in the dog: A literature and case review. Journal of the American Animal Hospital Association 15: 463-471
9. Sumner-Smith G, Walters E H 1976 The adjunctive use of the methyl methacrylate bone cement in the stabilization of multiple fractures. Journal of the American Animal Hospital Association 12: 778-781
10. Swain S F 1975 Evaluation of four techniques of cervical spinal fixation in the dog. Journal of the American Animal Hospital Association 166: 1080-1086.

ABSTRACT **SAMEVATTING**

LYSOSOMAL STORAGE DISEASE IN ABYSSINIAN CATS

Blood lipid analysis was performed on the serum of 2 normal kittens and 1 adult cat and on serum from 3 affected kittens.

Thin layer chromatography was done on tissue extracts of various organs from clinically affected kittens and unaffected unrelated kittens of a similar age, and on serum from carrier cats, affected kittens, related unaffected kittens unrelated kittens. Spleen and lymph node cell cultures were prepared from 1 affected kitten and the growth medium cell cultures were analysed for lipids. A lecithin-like phospholipid was identified in the serum of an affected kitten, a carrier cat and a related unaffected kitten. This substance was produced by the liver of affected kittens and also by macrophage-like cells in spleen cell cultures prepared from the spleen of a kitten with signs of the disease. (Lange, A. Lucia, Brown, J.M.M. & Maree, Charlotte C., 1983 Biochemical studies on a suspected lysosomal storage disease in Abyssinian cats. *Onderstepoort Journal of Veterinary Research*, 50, 149-155 (1983).)

BOOK REVIEW **BOEKRESENSIE**

PRECIS DE PATHOLOGIE DES POISSONS

P. DE KINKELIN, CH. MICHEL AND P. CHITTINO

INRA, Service des Publications Route de Saint-Cyr, F-78000 Versailles, France. pp. 348, 224 figures, 47 tables, 14 colour plates. INRA-OIE Edition, 1985 – 360FF including postage.

The development of aquaculture and leisure fishing has brought to light the increasing effect of fish diseases on output; these activities often favour the occurrence, or at least the dissemination, of certain diseases.

Fish health therefore takes on an increased economic importance, making the control of disease necessary.

This book, written in French, was realized on the recommendation of the OIE Commission for Fish Diseases, and co-edited with the Institut national de la Recherche Agronomique (INRA) in France, constituting a veterinary approach to ichtyopathology. From a miminum of theoretical data on the fish, its environmental and causes of disease, the authors show how these elements lead to diagnosis and justify certain methods of preventive treatment.

The book comprises five sections:

1. General remarks on fish diseases
2. Aetiological pathology
3. Chief pathologic characteristics of different types of breeding
4. Diagnosis in the field and in the laboratory
5. Prevention and treatment of diseases

Due to its extensive bibliography, numerous illustrations, glossary and index of key words, the book brings to the reach of Veterinary Services, veterinary practioners and laboratory technicians an entire range of theoretical and practical information which, up to now, has been lacking in veterinary literature.

BOOK REVIEW **BOEKRESENSIE**

EQUINE DISEASES
A TEXTBOOK FOR STUDENTS AND PRACTITIONERS

HANS-JURGEN WINTZER

1st Edn. 1982. Translated and revised by David A Weaver 1985. Verlag Paul Parey 1986 pp XVI and 439 with 364 illustrations.

This book covers all the systems of the horse starting with respiratory diseases, diseases reproductive diseases of the limb, metabolic diseases and toxicology to mention just a few.

To include such a variety of conditions into one volume necessitates very brief and sometimes abrupt definitions only. In depth discussions therefore is not a characteristic of this book. To students and colleagues with limited equine practises this book can be of help. Experienced equine practitioners will not benefit materially.

S.S. van den Berg

REVIEW OORSIG

THE IMMUNE SYSTEM OF THE NEONATAL AND WEANER PIGLET: A REVIEW

PAMELA HUNTER*

ABSTRACT: Hunter P. **The immune system of the neonatal and weaner piglet:** *Journal of the South African Veterinary Association* (1986) 57 No. 4, 243-245 (En) Veterinary Research Institute, 0110 Onderstepoort, Republic of South Africa.

The article reviews aspects of the immune system of the piglet from birth to weaning which may have practical application for the veterinarian. Subjects discussed are colostral immunity, the ability of the piglet to mount an immune response, the effect of weaning on the immune system, factors involved in the cause of post-weaning scours and its control.

Key words: Pig, immune system.

INTRODUCTION

Newborn piglets, being immunocompetent 50-70 days after conception[47], have the potential to mount an immune response as effectively as older animals[47]. Their vulnerability arises from the unprimed state of the immune system, having been sheltered in an antigen-free environment during gestation. The neonate is therefore dependent on maternal colostrum as a source of protective antibodies, while it mounts an immune response to the organisms in its environment.

THE ROLE OF COLOSTRUM

Porcine colostrum contains roughly 80% IgG, 15% IgA and 5% IgM[47]. All three[40] classes of immunoglobulin are absorbed through the intestinal mucosa. The survival of the immunoglobin fraction is promoted by a trypsin inhibitor present in colostrum, as well as the low acidity of the neonate's intestine[27]. Colostral uptake is maximal within the first few hours after birth[38] but can occur up to 36-48 hours[41]. Lecce et al.[29] prolonged the absorptive ability of the intestine for 5 days by maintaining newborn piglets on parental nutrients. They later showed that closure could be elicited by non-immunoglobulin-containing food[28]. Feeding newborn piglets with artificial colostrum can therefore prevent absorption of antibodies if colostrum is fed later.

The efficacy of colostrum in preventing neonatal infections is dependent on piglets receiving sufficient colostrum as soon as possible after birth. Factors such as large litters where piglets are underweight and weak and parturition is prolonged, will result in low colostral intake[11].

Temperature can influence suckling behaviour. Piglets experimentally exposed to a temperature of 10°C for short periods, failed to ingest sufficient colostrum[6]. Maternally derived antibodies supress[15] active antibody production in the piglet, until passively derived antibodies begin to decline[23]. Maternal IgM levels decline to a minimum at 2 weeks, while IgG levels are low at 4 weeks[17]. Active synthesis of IgM occurs at 2 weeks, of IgA at 3 weeks and IgG at 5 weeks[23]. Colostrum-deprived piglets show earlier development of the different antibody classes[23]. Similarly those fed on bovine colostrum showed synthesis of IgA and IgG during the first week while actively synthesized IgM is detectable almost imediately[23].

In recent years attention has focused on the cellular components of colostrum. Schlesinger & Covelli[42] reported that human infants born to mothers with positive tuberculin skin tests were sensitised. Evidence for colostral transfer of T-lymphocytes was advanced by other authors[14 34] at roughly the same time. They postulated lymphocyte transfer via colostrum through the gut of the neonate, resulting in a transfer of skin sensitivity and graft rejection abilities.

Seelig & Billingham[43] later succeeded in detecting the uptake of lymphocytes by the intestine of the neonatal rat. Sheldrake & Husband[44] were recently able to repeat this work with syngeneic lymphocytes (genetically identical) and were also able to show transfer of allogeneic lymphocytes (genetically dissimilar but of the same species) in neonatal lambs.

In the latter case the lymphocytes were shown to be transported to the lacteal lymph ducts and to establish themselves in the mesenteric lymph nodes. No work has yet been done to confirm this mechanism in pigs.

However, work on oral immunisation of sows with K88 antigen[24 25] revealed that there is a homing of sensitised lymphocytes (both B and T) to the udder of the sow. The role of these T-lymphocytes is not clear, but it is possible that they may be involved in transfer of cell-mediated immunity to the neonate.

THE IMMUNE STATUS OF THE PIGLET AT BIRTH

At birth the piglet has a high cortisol level, as a result of the hormonal trigger mechanisms at the inception of parturition[30]. These high levels drop rapidly to normal adult levels within a few days[30]. Dvorak[12] describes high cortisol levels persisting up to 60 days postpartum and claims that the neonate is therefore in an immunosuppressed state. High post-partum cortisol levels are said to cause depressed chemotactic and phagocytic responses in the neonatal calf[39], but this has not been shown in piglets. McCauley & Hartmann[30] argue that the leukocyte dynamics reflect a minimal suppression of the immune function of the newborn piglet as the neonatal piglet has a neutrophilia[30] which persists longer than 48 hours. Neutrophilia of newborns is also seen in other species[1]. A colostral factor[26] has been identified which promotes a neutrophilia in suckling calves. This factor has not yet been demonstrated in sows' milk. Neutrophils are powerful effector cells in a cytotoxic

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response dependent on specific antibodies (antibody-dependent cell-mediated cytotoxicity). The neutrophilia seen shortly after birth, together with the specific antibody present in colostrum may be important defence mechanisms[48] for the unprimed neonate.

The proportion of circulating B-lymphocytes of the newborn piglet is similar to that in adults but the total number is lower until 10 days after birth[30]. Nevertheless, piglets are able to mount antibody response to antigens such as TGE virus as efficiently as older animals[16], if there is no colostral interference.

The gastrointestinal immune system is fully responsive to antigen at one week of age[4 47]. Oral vaccination at 3 days with TGE virus brings about the production of specific antibody 5-14 days later[47]. Secretory IgA is produced and reaches adult levels earlier than other immunoglobulin classes[48].

THE EFFECT OF WEANING ON IMMUNITY

Weaning before the age of 5 weeks has an *in vivo* and *in vitro* suppressive effect on cell-mediated immunity[8], as assayed by inhibition of the phytohaemagglutinin (PHA) skin test and mitogen-induced blastogenesis of lymphocytes. Weaning at 5 weeks does not have the same suppressive effect on cell-mediated immune response[8].

Blecha[9] recently investigated the putative role of stress at weaning and its effect on the immune system, by assaying cell-mediated immunity and antibody production in 2 groups of weaners; one group was weaned with their littermates while the other group was placed with non-littermates. The latter group showed elevation of plasma cortisol levels but there was no difference between the lymphocyte blastogenesis, PHA skin test and antibody production between the two groups. The authors concluded that the acute, short-lived rise in cortisol levels seen at weaning does not impair immunological function. This view has been stated by other authors[10], who feel that the concept of stress as a function of cortisol levels is an oversimplification and that other endocrine systems should be examined more closely to elucidate the role of stress in precipitating swine disease. The immunosuppressive effects of early weaning should be borne in mind when formulating a vaccination program for weaners.

POST-WEANING SCOURS: THE ROLE OF NUTRITION AND TEMPERATURE

The cause of post-weaning scours, was until recently attributed primarly to enterotoxigenic *Escherichia coli* strains (ETEC). It was thought that piglets became susceptible due to stress and declining levels of maternal antibody. This hypothesis, while widely accepted, was not satisfactory, as the dosing of weaners with large numbers of ETEC did not consistently induce scours[20]. Kenworthy & Allen[21] showed that the post-weaning scours syndrome occurred in the absence of ETEC.

On histological studies typical intestinal changes were seen in piglets which had died from post-weaning scours. These were crypt cell mitosis, crypt hyperplasia and villous atrophy[21 22]. Similar intestinal pathology was noted by Stokes[46] who fed pigs ovalbumin and elicited a delayed-type hypersensitivity response. The pathology noted in the post-weaning scours cases as well as the experimental cases of Stokes et al., were noted to be similar to the picture described in humans suffering from food-sensitive enteropathies[35]. Miller et al.[31 32] tested the hypothesis that the post-weaning scours syndrome was due to hypersensitivity to dietary antigen: piglets were fed diets containing bovine casein at weaning. One group was fed casein in its natural from while a second group was fed hydrolysed casein (non-allergenic). The results supported the hypothesis that post-weaning scours was initiated by allergy to proteins in the weaning diet, as the group which received hydrolysed casein showed no cases of diarrhoea, while those fed untreated casein all developed diarrhoea.

A number of authors[3 18 36] presented evidence that tolerance to orally ingested antigen is preceded by a transient phase of hypersensitivity. Miller et al.[33] showed that piglets fed creep feed had a higher incidence of scouring than piglets weaned abruptly. They concluded that creep feeding promoted sensitisation while abruptly weaned piglets are unprimed and therefore experience a limited hypersensitivity response. The phenomenon of large doses of orally presented antigen inducing tolerance while small doses induce sensitisation has been described for various other species[39] as well as pigs[5]. English[13] has reported that if total creep intake exceeded 600 g before weaning, scouring was considerably reduced. He suggested that this intake was sufficient to induce tolerance and avoid the severe hypersensitivity reaction at weaning. Implementation of either abrupt weaning or use of non-allergenic feeds such as hydrolysed casein could therefore be considered as practical methods of controlling post-weaning scours.

Armstrong & Cline[2] noted that cold weather conditions exacerbated post-weaning scours (which at that time was attributed to ETEC infection). In the light of the dietary antigen hypersensitivity hypothesis, this can be explained by the findings of Kelly et al.[19] who found that cold ambient temperatures enhance delayed-type hypersensitivity responses. More specifically low temperatures bring about an elevation of cortisol levels which inhibit T-lymphocyte supressor cells or stimulate T-lymphocyte helper cells[7]. This also results in the phenomenon of increased antibody synthesis in cold conditions[7]. However, the role of low temperatures on the immune system appears to be complex, as cold is known to predispose piglets to TGE virus[45].

REFERENCES

1. Archer R K, Jeffcott L B 1977 Comparative Clinical Haematology Oxford Blackwell Scientific Publications: 178, 220, 315, 384
2. Armstrong W D, Cline T R 1977 Effects of various nutrient levels and environmental temperature on the incidence of colibacillary dairrhoea in pigs. Journal of Animal Science 45: 1042-1050
3. Asherson G L, Zembala M, Peira M A C C Mayhew B, Thomas W R 1977 Production of immunity and unresponsiveness in the mouse by feeding contact sensitising agents and the role of suppressor cells in the Peyer's patches, mesenteric lymph nodes and other lymphoid tissues. Cellular Immunology 33: 145-155.
4. Banks K I 1982 Host defence in the newborn animal. Journal of American Veterinary Medical Association 181: 1053-1056
5. Bennell M A, Husband A J 1981 Study of immunisation regimes for the stimulation of local immunity of the pig intestine. Research in Veterinary Science 30: 353-361
6. Blecha F, Kelley K W 1981 Cold stress reduces the acquisition of colostral immunoglobulins in piglets. Journal of Animal Science 52: 594-600.
7. Blecha F, Kelly K W 1981 Effect of cold and weaning on antibody mediated immune responses of pigs. Journal of Animal Science 53: 439-447
8. Blecha F, Pollmann D S, Nichols D A 1983 Weaning pigs at an

early age decreases cellular immunity. Journal of Animal Science 56: 396-400
9. Blecha F, Pollmann D S, Nichols D A 1985 Immunologic reactions of pigs regrouped at or near weaning. Americal Journal of Veterinary Research 46: 1934-1937
10. Dantzer R, Mormède P 1983 Stress in farm animals: a need for re-evaluation. Journal of Animal Science 57: 6-18
11. Dechamps P, Nicks B 1984 Influence de parametres non-infectieux et plus particulierement du confort thermique sur la viabilité et la vitalité de porcelets en maternite. Annales de Medecine Vétérinaires 128: 261-283
12. Dvorak M 1972 Adrenocortical function in foetal, neonatal and young pigs. Journal of Endocrinology 54: 374
13. English P R 1981 Proceedings of Pig Veterinary Society 7: 29-37
14. Head J R, Beer A E, Billingham R E 1977 Significance of the cellular component of maternal immune endowment in milk. Transplantation Proceedings 9: 1456.
15. Hoerlein A B 1957 The influence of colostrum on antibody response in baby pigs. Journal of Immunology 78: 112.
16. Jensen M T, Stone S S, Kemeny L J 1982 Serum antibody responses of neonatal and young adult pigs to TGE corona virus. Veterinary Immunology and Immunopathology 3: 529-533
17. Jönsson A 1973 Transfer of immunoglobulin from mother to offspring in the pig. Acta Veterinaria Scandinavica Supplement 43: 1-64
18. Kagnoff M E 1978 Effects of antigen feeding on intestinal and systemic immune response 1. Priming of precursor cytotoxic T-cells by antigen feeding Journal of Immunology 120: 395-399
19. Kelly K W, Greenfield R E, Everman J F, Parish S M 198 Delayed-type hypersensitivity, contact sensitivity, PHA responses to heat and cold stressed calves. American Journal of Veterinary Research 43: 775-779
20. Kenworthy R Allen W D 1966 The significance of *Escherichia coli* in the young pig. Journal of Comparative Pathology 76: 31-44
21. Kenworthy R, Stubbs J M, Syme G 1967 Ultrastructure of small intestinal epithelium in weaned and unweaned pigs with post-weaning diarrhoea. Journal of Pathology and Bacteriology 93: 493-498
22. Kenworthy R 1976 Observations on the effects of weaning in the young pig: Clinical and histopathological studies of intestinal function and morphology. Research in Veterinary Science 21: 69-75
23. Klobasa F, Werhahn E, Butler J E 1981 Regulation of humoral immunity in the piglet by immunoglobulins of maternal origin. Research in Veterinary Science 31: 195-206
24. Kortbeek-Jacobs N, Van der Donk H 1981 mRNA from porcine colostral and milk cells coding for specific antibodies in response to orally administered *Escherichia coli* strains. Veterinary Immunology and Immunopathology 2: 441-454
25. Kortbeek-Jacobs J M C, Van Kooten P J S, Van der Donk J A, Van Dijk J E, Rutten V P (1984) The effect of oral immunisation on the population of lymphocytes migrating to the mammary gland of the sow. Veterinary Microbiology 9: 287-299
26. La Motte G B, Eberhardt R J 1976 Blood leucocytes, neutrophil phagocytosis and plasma corticosteriods in colostrum-fed and colostrum derived calves. American Journal of Veterinary Research 37: 1189-1193
27. Laskowski M, Kassell B, Hagerty G 1957 A crystalline trypsin inhibitor from swine colostrum. Biochimica et Biophysica Acta 24: 300
28. Lecce J G, Matrone G, Morgan D O 1961 Porcine neonatal nutrition, absorption of unaltered porcine proteins and polyvinyl pyrrolidone from the gut of piglets. Journal of Nutrition 73: 158-166
29. Lecce J G, Morgan D O 1962 Effect of dietary regimen on cessation of intestinal absorption of large molecules (closure) in the neonatal pig and lamb. Journal of Nutrition 78: 263-268
30. McCauley I, Hartmann P E 1984 Changes in piglet leukocytes, B-lymphocytes and plasma cortisol from birth to 3 weeks after weaning. Research in Veterinary Science. 37: 234-241
31. Miller B G, Newby T J, Stokes C R, Hampson D, Bourne F J 1983 The role of dietary antigen in the aetiology of post-weaning scours. Annales de Recherche Veterinaire 14: 487-492
32. Miller G B, Newby T J, Stokes C R, Bourne F J 1984 Influence of diet on post-weaning malabsorption and diarrhoea in the pig. Research in Veterinary Science 36: 187-193
33. Miller B G, Newby T J, Stokes C R, Bourne F J 1984 Creep feeding and post-weaning diarrhoea in piglets. Veterinary Record 14: 296-297
34. Mohr J A 1973 The possible induction or acquisition of cellular hypersensitivity associated with the ingestion of colostrum Journal of Pediatrics 82: 1062
35. Mowat A Mc I, Ferguson A 1981 Hypersentitivity in the small intestinal mucosa V Induction of cell-mediated immunity to dietary antigen. Clinical and Experimental Immunology 43: 574
36. Newby T J, Stokes C R, Bourne F J 1980 Effects of feeding bacterial lipopolysaccharide and dextran sulphate on the development of oral tolerance to contact sensitising agents. Immunology 41: 617-621
37. Osburn B I, Stabenfeldt G H, Ardans A A, Trees C, Sawyer M 1974 Perinatal immunity in calves. Journal of American Veterinary Medical Association 164: 295-297
38. Payne L C, Marsh C L 1962 Gamma-globulin absorption in the baby pig: The non-selective absorption of heterologous globulin and factors influencing absorption time. Journal of Nutrition 76: 151-158
39. Pierce N F, Koster F T 1980 Priming and suppression of the intestinal immune response to cholera toxoid/toxin by parenteral toxoid in rats. Journal of Immunology 124: 307-311
40. Porter P 1969 Transfer of immunoglobulins IgA, IgG and IgM to lacteal secretions in the parturient sow and their absorption by the neonatal piglet. Biochemica et Biophysica Acta 181: 381
41. Porter P 1970 Serological changes in immunoglobulins IgG, IgA and IgM and *E. coli* antibody in young pigs. Immunology 18: 565-573
42. Schlesinger J J, Covelli H D 1977 Evidence for transmission of lymphocyte responses to tuberculin by breast feeding. Lancet: 529
43. Seelig L L, Billingham R E 1981 Transplantation 32: 308-314
44. Sheldrake R F, Husband A J 1985 Intestinal uptake of intact maternal lymphocytes by neonatal rats and lambs. Research in Veterinary Science 39: 10-15.
45. Shimizu M, Shimizu Y, Kodama Y 1978 Effects of ambient temperature on the induction of TGE in feeder pigs. Infection and Immunity 21: 747
46. Stokes C R, Newby T J, Bourne F J 1981 Altered immune function associated with dietary factors. Current Topics in Veterinary Medicine and Animal Science 12: 224-239
47. Tizard I 1982 An Introduction to Veterinary Immunology 2nd ed. W B Saunders Company
48. Zarkower A, Eskew M L, Scheuchenzuber W J, Ferguson F G Confer F. 1982 Antibody-dependent cell-mediated cytotoxicity in pigs. American Journal of Veterinary Research 43: 1590-1593

BOOK REVIEW **BOEKRESENSIE**

PATHOLOGY OF DOMESTIC ANIMALS

K.V.F. JUBB, P.C. KENNEDY AND N. PALMER

3rd Edn. Academic Press Inc., Orlando, Florida, 32887 U.S.A. 1987. Vol. 1 pp.XIX and 574; Vol. 2 pp.XVII and 582; Vol.3 pp.XVII and 527. Numerous figures.
ISBN 0-12-391601-1; 0-12-391602-X; 0-12-391603-8 Price ± R520.00

This handbook dealing with veterinary pathology and which is now published in 3 volumes, is considered to be the standard text in Veterinary Pathology.

The text is arranged according to systems and subdivided into 3 volumes on this basis. Each system is dealt with first on a general basis and then followed by a discussion on the specific diseases traditionally dealt with under each system. Numerous figures (all in black and white), mostly of a good quality supplement the text. Each volume is supplied with an index which in the first 2 volumes only refer to the information contained within that specific volume; that in the 3rd volume contains a comprehensive index covering all 3 volumes. Ample references are provided at the end of each chapter in a systematic way sub-divided into the various topics dealt with in each chapter.

Since each chapter has been authored by 1 or more different authors, the standard of presentation and content vary inevitably and it will be obvious in one or two sections. A disturbing deficiency in general is the lack of electron-microscopical detail both in respect of text and illustrative material – a sad omission if one considers the wealth of available information and the extent to which it forms part of pathology in general.

Probably the greatest criticism that may be directed against the textbook is the lack of detail in respect of specific diseases and species differences; a fact acknowledged by the authors in the introduction. These comments are probably more appropriate when referring to the diseases and intoxications of Africa; many important disease have only been dealt with sketchily and a number have been omitted altogether.

Despite these comments, this remains an excellent treatise on what is known about veterinary pathology and it is recommended for use by practicing and academic pathologists and should be obtainable as a reference work in all libraries specialising in veterinary literature. The price in South Africa, precludes its acquisition by practitioners and students.

N.P.J. Kriek

DIE KLINIESE FARMAKOLOGIE VAN GLISEROL GUAIAKOLAAT ETER IN DIE PERD – 'N OORSIG

P. STADLER*

ABSTRACT: Stadler P. **The clinical pharmacology of glyceryl guaiacolate ether in the horse – A review.** *Journal of the South African Veterinary Association* (1986) 57 No. 4, 247-249 (Afrik) Department of Medicine, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, 0110 Onderstepoort, Republic of South Africa.

The physical and chemical properties, administration, biotransformation, pharmacological effects, clinical applications, side-effects, toxicity and contraindications of glyceryl guaiacolate ether in the horse are reviewed.

Key words: Glyceryl guaiacolate ether, horses, clinical pharmacology.

INLEIDING

Vanweë die grootte en anatomiese bou van die perd is daar soms praktiese probleme en gevare verbonde aan die hantering en die uitvoer van prosedures op hierdie spesie. In 'n poging om hierdie probleme te voorkom of te verminder word daar van verskeie middels gebruik gemaak. Een so 'n middel wat met groot sukses gebruik kan word is gliserol guaiakolaat eter (GGE) wat in hierdie artikel bespreek sal word.

FISIESE EN CHEMIESE EIENSKAPPE

Gliserol guaiakolaat eter (GGE) is die generiese naam vir 3-(0-methoksiephenoksie)-1,2 propaandiol[4 18]. Dit is 'n wit poeier met 'n bitter smaak[1 13]. Dit is swak oplosbaar[7 13] en verder is dit onstabiel in oplossing, sodat dit deels van 'n 5% oplossing uitkristalliseer by kamertemperatuur[3 5]. Die kristalle kan maklik weer opgelos word deur dit te verhit en te roer[3 13].

Beskikking in die liggaam

GGE kan oraal toegedien word, maar vir 'n waarneembare effek is 'n hoë dosis nodig[1 3]. Die intraveneuse roete word dus verkies[1 13] en dit word gewoonlik as 'n 5-10% oplossing in 5% dekstrose of steriele water toegedien[1 8]. As 'n 5% oplossing ekstravaskulêr beland, veroorsaak dit gewoonlik nie 'n erge weefselreaksie nie[13], maar 'n 15% oplossing is wel irriterend[22].

By 'n plasmakonsentrasie van minder as 150$\mu g/m\ell$ in ponies kon geen sigbare effekte waargeneem word nie[4]. Die afname in plasmakonsentrasie volg in beide geslagte eerste orde kinetika[4]. Die plasma halfleeftyd is egter 84,4 minute in die hings teenoor 59,6 minute in die merrie[4]. Die tyd wat dit effektief is, is ook langer in die hings as die merrie teen dieselfde dosis[4]. Al die effekte keer egter terug na normaal by dieselfde plasmakonsentrasie wat bewys dat dit nie 'n verskil in reseptorsensitiwiteit is nie[4]. Die beste moontlike verklaring is 'n verskil in biotransformasie na aktiewe produkte[4].

Die lewer is die hooforgaan van biotransformasie[4]. GGE ondergaan naamlik 0-dealkilasie om katekol te vorm wat dan gekonjugeer word na meer polêre substanse soos die glukuronied of etersulfate[4]. Daar mag ook moontlik nog ander metaboliete wees[4]. Die demonstrasie van vry katekol in die uriene dui op 'n verbreking van die eterverbinding tussen die guaiakol nukleus en die gliseriel eenheid[4].

Uitskeiding vind hoofsaaklik in die uriene plaas[9].

FARMAKOLOGIESE EFFEKTE

Sentrale senuweestelsel

a) Spierverslapping

GGE is 'n sentraalwerkende spierverslapper[15 16] wat polisinaptiese reflekse meer effektief blokkeer as monosinaptiese reflekse[13]. Dit blokkeer naamlik die oordraging van senuwee-impulse by die internunsiële neurone van die rugmurg, breinstam en subkortikale areas[3 13]. Die aanvang en duurte van verslapping hang af van die spoed van toediening[19]. Die eerste teken van verslapping is dat die perd se kop begin hang, gevolg deur oorbreking by die kootgewrigte[7]. Dit is nadat c. 40mg/kg GGE intraveneus toegedien is[18]. Vinnige toediening gee 'n vinnige, gladde neergaan, maar as die toediening te lank duur, vind steiering plaas voordat die perd gaan lê, hoewel opgewondenheid of agteroorval nie voorkom nie[19]. 'n Gemiddelde dosis van 11 mg/kg intraveneus, dit wil sê, 2,2 $m\ell$/kg van 'n 5% oplossing GGE, is nodig om 'n perd te laat lê[6]. Maksimale verslapping kom eers 'n paar minute na die toediening voor[15 18].

Hoewel die skeletale spiere verslap, hou die diafragma aan met werk en anders as met die perifereelwerkende spierverslappers is daar dus nie 'n gevaar van respiratoriese paralise met terapeutiese dosisse nie[17 21]. In ponies is volledige spierverslapping, met uitsondering van die diafragma, gesien by plasma konsentrasies van meer as 238 $\mu g/m\ell$[4]. Wanneer die perd lê, bly die palpebrale, pupillêre en korneale reflekse teenwoordig en meeste gevalle reageer op pynprikkels deur middel van dilatasie van die pupille en beweging van die ore en ooglede[1]. Hoewel die slukrefleks gewoonlik nog teenwoordig is[18], is die verslapping van die faringeale en laringeale spiere voldoende om die plasing van 'n endotrageale buis te vergemaklik[9 17]. Die tonus van die kremastor spier word ook nie geaffekteer nie[17].

Die tydsduur van spierverslapping na 'n enkele dosis is ongeveer 15-30 minute wat beter is as die korter werking van die perifereel-werkende spierverslappers[4]. Wanneer 'n perd weer opstaan, vind dit stil plaas sonder veel steiering en inkoördinasie[1 19].

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b) Kalmering

'n Mate van lomerigheid en kalmering word met GGE verkry[11 13]. Dit is blykbaar as gevolg van die middel se effek op die retikulêre formasie van die breinstam[7 11]. Die toediening van 50 mg/kg GGE intraveneus het 'n kalmerende effek vir ongeveer 5-10 minute waartydens die perd min reageer op eksterne stimuli[1 7].

c) Pynstilling

Daar blyk 'n mate van pynstillende effek te wees[11 13] as gevolg van die middel se effek op die subkortikale areas van die brein[11]. Die perd sal egter nog altyd op pyn reageer na toediening van kliniese dosisse[7 19] deur dilatasie van die pupille en beweging van die ore en ooglede[1]. Vir chirurgiese prosedures moet lokale of algemene verdowing dus saam toegedien word.

Kardiovaskulêre stelsel

GGE het 'n minimale effek op die kardiovaskulêre stelsel teen terapeutiese dosisse[1 10]. Die hartspoed bly onveranderd[10 19] of styg effens[10 20]. 'n Effense daling in bloeddruk word waargeneem, geassosieer met 'n daling in periferele weerstand[10 17]. Daarvoor word gekompenseer deur die verhoogde hartspoed[20]. Geen aritmië word veroorsaak nie[17].

Respiratoriese stelsel

Net soos op die kardiovaskulêre stelsel is die effek op die respiratoriese stelsel minimaal[10 11]. Anders as met die perifireel-werkende spierverslappers gaan die diafragma ongehinderd aan met werk[17 21] en alhoewel daar verslapping van die faringeale en laringeale spiere is[9 21], is daar geen respiratoriese paralise met terapeutiese dosisse nie[17 21]. Die respiratoriese tempo mag effens styg[7 19] of onveranderd bly[18 19], terwyl die getyvolume mag daal[1 17]. Die minuutvolume bly egter onveranderd[17].

By toksiese dosisse mag respiratoriese stilstand soms die hartstilstand voorafgaan[7].

KLINIESE GEBRUIKE

(a) Hulpmiddel by algemene verdowing – As gevolg van die middel se weglaatbare effekte op die kardiovaskulêre en respiratoriese stelsels, blyk dit die middel van keuse te wees in die hoë risiko pasiënt voordat die pasiënt onder inasemingsnarkose geplaas word[11 18]. Induksie is glad en sonder paniek of neerval van die perd[4 11]. Beide die verslapping van die faringeale en laringeale spiere[17] en die feit dat die perd beheer is[10], bevorder intubasie[10 14].

Die middel se spierverslappende effek is baie voordelig tydens chirurgie[11 14], veral in gevalle waar inasemingsnarkose nie moontlik is nie of 'n narkotiseur nie beskikbaar is nie, aangesien daar nie met respirasie ingemeng word nie[9]. Na toediening, alleen of in kombinasie met algemene verdowingsmiddels, is die herstel ook glad met baie min inkoördinasie[1 11].

GGE potensieer die effek van algemene verdowingsmiddels sodat minder van die verdowingsmiddels dus gebruik kan word[15 18]. Dit kan suksesvol in kombinasie met verskeie algemene verdowingsmiddels gebruik word, soos halotaan of enfluraan[6 12], tiamilal[5 12], tiopentoon[2 3] en ketamien[16] sowel as met chloraalhidraat[9].

(b) Chemiese beheer – Dit is 'n bruikbare middel as 'n perd byvoorbeeld op 'n kanteltafel of operasietafel geplaas moet word of om hom lêend te vervoer[7].

(c) Neertrek van perde[9 18] – Die dosis het varieer van 73 tot 139 mg/kg intraveneus om dieselfde perd herhaaldelik neer te trek, maar geen toleransie is opgemerk nie[7]. Ongewenste steiering kan voorkom word deur die perd te ondersteun of teen 'n muur te druk totdat hy reg is om te gaan lê[7].

(d) Ortopedie – Toediening teen 'n dosis van 50 mg/kg intraveneus nadat algemene narkose gestaak is, lei tot 'n gladde herstel na ortopediese operasies[1].

(e) Kalmering[1 7] – 'n Dosis van 50 mg/kg intraveneus gee kalmering vir 5-10 minute wat voldoende is vir die oplaai van perde en die uitvoer van kort diagnostiese en terapeutiese prosedures[7].

(f) Obstetrie – Regstelling van wanpresentering word bevorder deurdat abdominale sametrekking en spartel deur die merrie beheer word[6 7]. 'n Dosis van 50 mg/kg intraveneus is gebruik[6].

(g) GGE is al suksesvol gebruik in die behandeling van tetanus[17].

NADELE EN NEWE-EFFEKTE

(a) Een van GGE se grootste nadele is dat 'n groot volume toegedien moet word, aangesien 'n te hoë konsentrasie hemolise sal veroorsaak[9 16]. Gevolglik word die werking van die middel vertraag.

(b) As gevolg van sy swak oplosbaarheid en gevolglike uitkrisstallisering moet 'n vars oplossing elke keer voor gebruik, opgemaak word[19].

(c) As GGE alleen gebruik word, mag 'n ongewenste periode van steiering voorkom[1 9]. Dit kan egter voorkom word deur die perd ferm te ondersteun of teen 'n muur te druk totdat hy verslap genoeg is om stil te kan gaan lê[9].

(d) 'n Effense daling in bloeddruk word veroorsaak[10 17].

(e) Hemolise – Gekonsentreerde oplossings mag intravaskulêre hemolise veroorsaak[13 22]. Dit is verwant aan die konsentrasie eerder as die totale dosis[19 22]. Die drumpelwaarde sonder byvoegings is tussen 16 en 20%[19]. Die oorsaak is onbekend, maar 'n direkte toksiese effek op die rooibloedsel word vermoed, aangesien die byvoeging van byvoorbeeld glukose, dekstrose of levulose blykbaar die rooibloedselmembraan stabiliseer teen die effek[19 22].

(f) Tromboflebitis van die jugulare vena het al voorgekom nadat oplossings van 10% en meer gebruik is[5 18]. Die presiese oorsaak is onbekend, maar die konsentrasie van die oplossing, vinnige toediening en onverskillige plasing van die naald mag bydraende faktore wees[19].

(g) Hoë konsentrasies soos 15% is irriterend indien dit ekstravaskulêr beland[22].

TOKSISITEIT

GGE is 'n veilige middel[1 7]. Die toksiese dosis is 2-3[17] tot 4 [7] keer die terapeutiese dosis. Kliniese tekens wat met oordosering gesien word is tetanie, konvulsies, hipotensie, diep koma, apnee en vrekte[4]. Hierdie tekens kan gedeeltelik toegeskryf word aan die katekol wat tydens GGE se biotransformasie gevorm word[4]. Erge ekstensorspasmas het voorgekom na toediening van gemiddeld 182 mg/kg intraveneus en dit kan dien as 'n waarskuwing van oordosering[7 21]. Dit verdwyn as die toediening dan volgehou word[6 7]. Vrekte as gevolg van hartstilstand

het na ongeveer 450 tot 462 mg/kg intraveneus ingetree[7].

Indien die konsentrasie van die oplossing nie so hoog is dat hemolise voorkom nie, word lewer- en nierfunksie nie benadeel[9,17].

Herhaalde toedienings het ook geen nadelige effekte opgelewer nie.

KONTRA-INDIKASIES

Geen kontra-indikasies vir GGE se gebruik is bekend nie[17]. Toediening aan dragtige merries was ook sonder nadele, hoewel die middel deur die plasenta gaan en fetale beweging onderdruk[11].

BESPREKING

As gevolg van GGE se onstabiliteit in oplossing moet 'n vars oplossing elke keer gebruik word. Dit word gewoonlik as 'n 5-10% oplossing intraveneus toegedien, aangesien 'n te hoë konsentrasie hemolise veroorsaak. Dit het die toediening van 'n groot volume tot gevolg wat lastig is.

Dit is egter 'n goeie spierverslapper aangesien dit nie met respirasie inmeng nie en veral tydens chirurgie is die middel dus van groot waarde. Daar is ook net minimale effekte op die kardiovaskulêre stelsel. Die wye veiligheidsgrens en weinige newe-effekte maak dit 'n baie bruikbare middel. Die belangrikste newe-effek naamlik hemolise kan voorkom word deur die konsentrasie te verlaag of glukose of dekstrose by te voeg.

Daar is dus beslis 'n baie goeie plek vir hierdie middel in die behandeling en hantering van perde.

VERWYSINGS

1. Bishop W J, 1978 Glyceryl guaiacolate in equine anaesthesia. New Zealand Veterinary Journal 26: 284
2. Brouwer C J 1985 Use of guaiacol glycerine ether in clinical anaesthesia in the horse. Equine Veterinary Journal 17: 133-136
3. Coffman M T, Pedersoli W M 1971 Glyceryl guaiacolate as an adjunct to equine anesthesia. Journal of the American Veterinary Medical Association 158: 1548-1553
4. Davis L E, Wolff W A 1970 Pharmacokinetics and metabolism of glyceryl guaiacolate in ponies. American Journal of Veterinary Research 31: 469-473
5. Ellis R G, Lowe J E, Schwark W S, Taylor J L 1977 Intravenously administered xylazine and ketamine HCl for anesthesia in horses. Journal of Equine Medicine and Surgery 1: 259-265
6. Funk K A 1970 Glyceryl guaiacolate: a centrally acting muscle relaxant. Equine Veterinary Journal 2: 173-177
7. Funk K A 1973 Glyceryl guaiacolate: some effects and indications in horses. Equine Veterinary Journal 5: 15-19
8. Grandy J L, McDonell W N 1980 Evaluation of concentrated solutions of guaifenesin for equine anesthesia. Journal of the American Veterinary Medical Association 176: 619-622
9. Hall L W 1971 Wright's Veterinary Anaesthesia and Analgesia 7th edn Bailliere Tindall, London
10. Health R B, Gabel A A 1970 Evaluation of thiamylal sodium, succinycholine and glyceryl guaiacolate prior to inhalation anesthesia in horses. Journal of the American Veterinary Medical Association 157: 1486-1494
11. Jackson L L, Lundvall R L 1970 Observations on the use of glyceryl guaiacolate in the horse. Journal of the American Veterinary Medical Association 157: 1093-1095
12. Jackson L L, Lundvall R L 1972 Effect of glyceryl guaiacolate – thiamylal sodium solution on respiratory function and various hematologic factors of the horse. Journal of the American Veterinary Medical Association 161: 164-168
13. Jones L M, Booth N H, McDonald L E 1977 Veterinary Pharmacology and Therapeutics 4th edn AMES, The Iowa State University Press
14. Kalhoro A B, Rex M A E 1984 Observations on the use of glyceryl guaiacolate as an adjunct to general anaesthesia in horses. Australian Veterinary Journal 61: 49-53
15. Mostert J W 1963 A centrally acting relaxant in anaesthesia. Suid-Afrikaanse Tydskrif vir Geneeskunde 1281-1284
16. Muir W W, Skarda R T, Sheehan W 1978 Evaluation of xylazine, guaiafenesin and ketamine hydrochloride for restraint in horses. American Journal of Veterinary Research 39: 1274-1278
17. Roberts W D 1968 The role of glyceryl guaiacolate in a balanced equine anesthetic. Veterinary Medicine/Small Animal Clinician 63: 157-162
18. Schatzmann U 1974 The Induction of general anaesthesia in the horse with glyceryl guaiacolate. Comparison when used alone and with sodium thiamylal (Surital). Equine Veterinary Journal 6: 164-168
19. Schatzmann U, Tschudi P, Held J P, Muhlebach B 1978 An investigation of the action and haemolytic effect of glyceryl guaiacolate in the horse. Equine Veterinary Journal 10: 224-228
20. Schatzmann U, Koehli M, Dudan F, Jones R S 1984 Some cardiopulmonary effects of glyceryl guaiacolate in combination with preanesthetics in the standing horse. Equine Practice 6: 17-21
21. Tavernor W D 1970 The influence of glyceryl guaiacolate ether on cardiovascular and respiratory functions in the horse. Research in Veterinary Science 11: 91-93
22. Westhues M, Fritsch R 1965 Animal Anaesthesia Vol 2 Oliver and Boyd Ltd, London

ABSTRACT

SAMEVATTING

BUFFALO IN THE NORTHERN NATAL GAME PARKS SHOW NO SEROLOGICAL EVIDENCE OF INFECTION WITH FOOT-AND-MOUTH DISEASE VIRUS

A total of 594 sera collected from buffalo (*Syncerus caffer*) in the Hluhluwe/Umfolozi Game Reserve complex, Ndumu Game Reserve and the eastern shores of Lake St Lucia were examined for antibody to SAT 1, 2 and 3 types of foot-and-mouth disease (FMD) virus in neutralization tests. No neutralization of SAT 2 or 3 viruses was exhibited by any of the sera tested at final dilutions $>10^{0,9}$. A small proportion (2,9 %) of sera neutralized SAT 1 virus at dilutions up to $10^{1,7}$, but these were considered to be due to non-specific reactions. This, together with the absence of clinical FMD in both cattle and game in this region over at least a 45-year period and the failure to isolate FMD virus from pharyngeal scrapings of buffalo sampled in the area, leads to the conclusion that FMD does not occur in these buffalo populations. (Esterhuysen, J.J., Thomson, G.R., Flammand, J.R.B. & Bengis, R.G., 1985. Buffalo in the northern Natal game parks show no serological evidence of infection with foot-and-mouth disease virus. *Onderstepoort Journal of Veterinary Research*, 52, 63—66 (1985).)

TO THE EDITOR — AAN DIE REDAKSIE

EFFECT OF A GONADOTROPIN-RELEASING HORMONE ANALOGUE INJECTED IN BULLDOG BITCHES AT THE TIME OF MATING

A variety of uncontrolled methods, based largely on speculation, have been used recently by bulldog breeders in attempts to obtain larger litters. The drugs most often used are gonadotropin-releasing hormone (GnRH) analogues, even though none are registered for use in dogs. Conflicting verbal reports on the efficacy of treatments led to analysis of the accurately kept records obtained from a breeder.

Four fertile bulldog males were used on 43 bitches. The dogs and bitches were housed separately in concrete runs and fed a commercial dry dog food *ad lib*. Bitches were mated at least twice during their oestrus periods, at two day intervals. Over a period of one year, twenty two bitches were randomly treated once with 0,004 mg buserelin (1 ml' Receptal, Hoechst) injected subcutaneously at the time of the first mating. The remaining twenty one bitches were not treated.

In the group of bitches treated, seven bitches which had conceived on every previous mated cycle did not produce pups. Ten bitches of unknown breeding history did also not produce pups. Five bitches conceived and produced litters. Thus, 22,73 % of the treated bitches produced litters, with a mean litter size of 8,6 (6-13). The average number of pups born per cycle was 1,95. Amongst the untreated bitches, 38,1 % produced litters, with a mean litter size of 4,88 (2-11). The average number of pups born per cycle was 1,86.

Several workers have investigated the effects of GnRH analogues on the cycle of the bitch. McRae et al.[2] found that the long term treatment of bitches with naferelin, a potent GnRH agonist, exerted anti-reproductive effects. There was a suppression of oestrus, a delay in puberty, and bitches treated in anoestrus displayed an induced but sterile oestrus. However, it appeared not to interfere with an already entrained ovulation. In another report, single injections of a GnRH analogue were found to be ineffective in inducing oestrus in anoestrus bitches[3]. Chakraborty et al.[1] were able to cause ovulation with a GnRH analogue in bitches which had been induced with PMSG or FSH. The effect on a natural oestrus was not investigated. It may thus be noted that GnRH analogues produce varied effects on the cycle of the bitch, and appear to depend on the stage of the cycle and the period of administration.

The data derived from the analysis of the information supplied by the bulldog breeder indicated a large variation in response pattern in the treated bitches. A large number of previously fertile bitches failed to produce litters, and unsuccesful matings were seen in a further 10 bitches, resulting in a very low overall number of pups being born. From the clinical viewpoint therefore, this use of the GnRH analogue did not produce the desired effect.

While this randomly derived data concurs with the results obtained from planned trails, it highlights the possible hazards which may result from the uncontrolled use of GnRH analogues by breeders or veterinarians. Until more conclusive data are available therefore, the use of these analogues for this purpose should be discouraged.

REFERENCES

1. Chakraborty P K, Wildt D E, Seager S W J 1982 Induction of oestrus and ovulation in the cat and dog. Veterinary Clinics of North America: Small Animal Practice 12: 85-92
2. McRae G I, Roberts B B, Worden A C, Bajka A, Vickery B H 1985 Long term reversible suppression of oestrus in bitches with nafarelin acetate, a potent LHRH agonist. Journal of Reproduction and Fertility 74: 389-397
3. Oettel M 1979 Möglichkeiten zur hormonellen Beeinflussung bei der Hündin. Monatshefte für Vet-Med 942-945

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DIFFERENTIATION OF *BOOPHILUS DECOLORATUS* AND *BOOPHILUS MICROPLUS*

ONDERSKEID TUSSEN *BOOPHILUS DECOLORATUS* EN *BOOPHILUS MICROPLUS*

Fig. 1: *B. decoloratus:* Female: Capitulum, ventral.
Fig. 1: *B. decoloratus:* Wyfie: Kapitulum, ventraal.

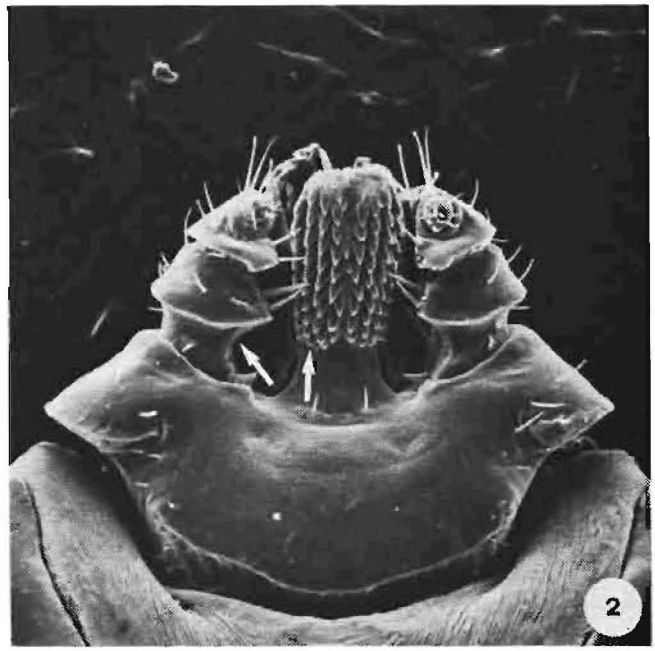

Fig. 2: *B. microplus:* Female: Capitulum, ventral.
Fig. 2: *B. microplus:* Wyfie: Kapitulum, ventraal.

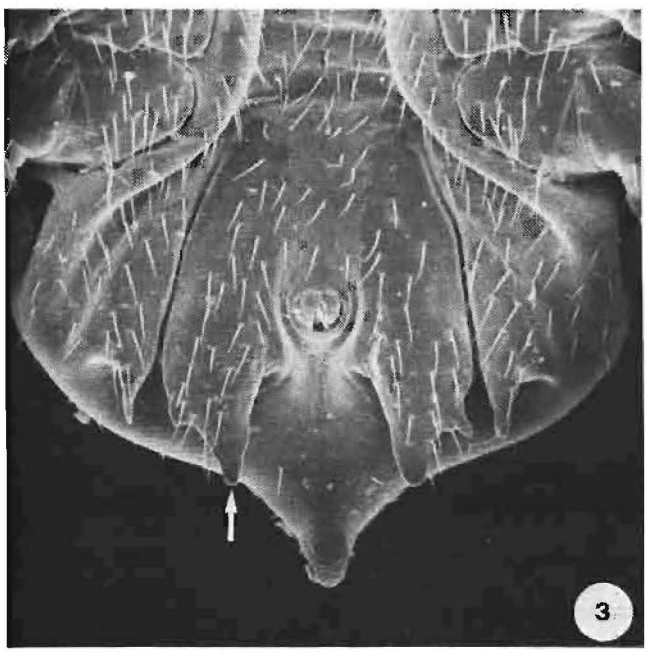

Fig. 3: *B. decoloratus:* Male: Anal plates, ventral.
Fig. 3: *B. decoloratus:* Mannetjie: Anaalplate, ventraal.

Fig. 4: *B. microplus:* Male: Anal plates, ventral.
Fig. 4: *B. microplus:* Mannetjie: Anaalplate, ventraal.

Redwater (babesiosis), which is one of the most economically important cattle diseases in South Africa and causes large losses annually, is transmitted by the one-host blue ticks.

Boophilus decoloratus is the commoner of the 2 species occurring here and transmits only one of the organisms that causes redwater, *Babesia bigemina*. *Boophilus microplus* has a more restricted distribution:

Rooiwater (babesiose), wat een van die mees ekonomies belangrike veesiektes in Suid-Afrika is en jaarliks grootskaalse verliese veroorsaak, word deur die eengasherige bloubosluise oorgedra.

Boophilus decoloratus kom plaaslik meer algemeen voor en dra slegs een van die organismes wat Rooiwater veroorsaak, nl. *Babesia bigemina* oor. *Boophilus microplus* het 'n meer beperkte verspreiding en kom

it occurs in some of the coastal areas of the Cape Province, the Transkei and Natal and in parts of northern Natal and the Transvaal. It can transmit both *Babesia bovis* and *B. bigemina*. It is therefore important to be able to identify these 2 *Boophilus* species accurately. The main differences between them are illustrated here by scanning electron micrographs.

B. decoloratus has a 3/3 dentition (i.e. 3 rows of teeth on each side on its hypostome) and its palpal segment I has a ventral bristle-bearing protuberance (Fig. 1). *B. microplus* has a 4/4 dentition and its palpal segment I is concave ventrally (Fig. 2).

It is sometimes difficult to examine the mouthparts of the males microscopically and the shape of the adanal plates can also be used as a taxonomic feature. In *B. decoloratus* males the adanal plates reach beyond the posterior body margin and they have a relatively long internal spur (Fig. 3). In *B. microplus* the adanal plates do not reach beyond the posterior body margin and they have a short internal spur and an even shorter external spur (Fig. 4).

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voor in sommige kusstreke van die Kaapprovinsie, die Transkei en Natal asook in dele van noord Natal en in die Transvaal. *B. microplus* kan beide *Babesia bovis* en *B. bigemina* oordra. Dit is dus belangrik om die twee naverwante *Boophilus* spesies noukeurig te kan onderskei. Die vernaamste verskille tussen hulle word hierin d.m.v. skandeer elektronmikroskopie geïllustreer.

B. decoloratus het 'n 3/3 dentisie (3 rye tande aan elke kant van die hipostoom) en die eerste segment van die palpe dra ventraal 'n haaragtige stekel op 'n uitstulping (Fig. 1).

B. microplus het 'n 4/4 dentisie en die eerste segment van die palpe is ventraal konkaaf (Fig. 2).

Dit is soms moeilik om die mannetjies se monddele mikroskopies te ondersoek en hier kan die vorm van die adanale plate as taksonomiese kenmerk gebruik word. By *B. decoloratus* besit die adanale plate 'n relatiewe lang interne stekel, wat verby die postero-liggaamswand uitsteek, en 'n korter eksterne stekel (Fig. 3). By *B. microplus* besit die adanale plate 'n kort interne stekel, wat nie verby die postero-liggaamswand uitsteek nie, en selfs 'n korter eksterne stekel (Fig. 4).

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ABSTRACT

SAMEVATTING

FURUNCULOSIS IN RAINBOW TROUT

Ulcerative skin lesions were encountered in rainbow trout raised in sea water by a commercial concern in the Western Cape, South Africa. Grossly, the lesions resembled furunculosis but, histopathologically, they differed from typical furunculosis in that bacterial colonies were rarely found in the organs, and also the kidneys and spleens were minimally involved. The causative organism was identified as an achromogenic *Aeromonas salmonicida* that shared characteristics with all 3 subspecies, *salmonicida, masoucida* and *achromogenes*. This is the first report of an outbreak of this disease in South Africa. (Boomker, J., Henton, M.M., Naudé, T.W. & Huchzermeyer, F.W., 1984. Furunculosis in rainbow trout (*Salmo gairdneri*) raised in sea water. *Onderstepoort Journal of Veterinary Research*, 51, 91-94 (1984).)

GOUE MEDALJE VAN DIE SAVV VIR 1986
GOLD MEDAL OF THE SAVA FOR 1986

RICHARD KARL REINECKE

Richard Karl Reinecke was born in Johannesburg on 7 May 1924. He matriculated in 1936 at the King Edward School, Johannesburg. He received the BVSc degree at Onderstepoort in 1947 and then spent nearly 8 years in private practice in Cape Town and in Krugersdorp. During 1954 he joined the staff of the section of Helminthology, Veterinary Research Institute, Onderstepoort as a State Veterinarian. He later became Senior State Veterinarian and also Senior Lecturer in Helminthology at the Faculty of Veterinary Science of the University of Pretoria. When this Faculty became autonomous in 1973, he was appointed Professor and Head of the Department of Parasitology.

In 1959 he was awarded the DVSc degree by the University of Pretoria for research on the epidemiology of nematode infestations of cattle in the north-western Cape Province. This was followed by an M.Med.Vet (Parasitology) awarded in 1978 by the same university for research on the effect of abomasal nematodes on subsequent infestations with *Haemonchus contortus*. In 1980 he received the DSc degree in Zoology from the University of Potchefstroom for his research on the development of a larval anthelmintic test in ruminants.

Shortly after Richard Reinecke joined the staff of the Veterinary Research Institute in 1954 he was sent to "Armoedsvlakte" in the north-western Cape to investigate severe helminth parasitism in young cattle. Here he established that the kraal, which was never cleaned, was the most important source of infestation. On dairy-ranches in the district, the calves, which never left the calf-pens, became heavily infested when suckling the teats and udders of cows that had become contaminated with nematode larvae when they lie down in the kraals. The calves also became infested when they licked each other and themselves and in so doing, ingested larvae clinging to skin and hair.

This research so fired his enthusiasm that he encouraged a number of state veterinarians employed by the Division of Veterinary Services to conduct surveys on the epidemiology of nematode parasites of ruminants in the southern, eastern and Karoo regions of the Cape Province and in Natal. The results of these surveys are still used as the basis for helminth control programmes in these regions. In recent years he has himself commenced research on the seasonal abundance of helminths of domestic ruminants in the western Cape Province, a region sadly neglected up till now.

During the early 1960's, a number of new anthelmintics came onto the market and Reinecke became involved in testing the efficacy of these remedies. What interested him more, however, was improving the methods by which these anthelmintics were tested. He soon realised that most of this work would have to be done by veterinarians in the employ of the pharmaceutical companies, and these veterinarians, like the state veterinarians conducting the seasonal incidence surveys, needed training in the taxonomy and collection of helminths. To achieve this, he conducted a number of short courses attended by numerous veterinarians, some from as far afield as Zimbabwe, and in this way made a magnificent personal contribution towards continuing education. To assist with this training, he compiled a laboratory manual which to this day is a remarkably informative document.

He continued his personal research into the improvement of anthelmintic efficacy testing and with Prof Groeneveld developed the non-parametric method of testing the efficacy of an anthelmintic. This method precludes the subjective evaluation of efficacy and predicts the efficacy of an anthelmintic on a flock basis. His research in this field led to his recognition as an expert in this field by the World Association for the Advancement of Veterinary Parasitology and his appointment by this body, to an international panel for standardising anthelmintic tests.

He also served as technical advisor to the Registrar for Stock Remedies registered in terms of Act 36 of 1947. Because of his insistence on the generation of local efficacy data for anthelmintic registration purposes, he was to a large degree instrumental in the establishment in South Africa of a number of research farms by various pharmaceutical companies.

During research into the methodology of anthelmintic testing he noticed that there appeared to be competition for survival between the various nematode species infesting the abomasum of sheep. He exploited this

tendency and developed a vaccine using the relatively non-pathogenic *Trichostrongylus axei* to protect sheep against the highly pathogenic *H. contortus*. Unfortunately for a number of practical reasons, this vaccine is not a commercially viable proposition.

During all this time he was actively engaged in teaching under-graduate veterinary students and directing the research of post-graduate students. Realising the unsuitability of the available text books on veterinary helminthology for South African conditions, he proceeded to write the textbook "Veterinary Helminthology" which appeared during 1983.

He is comprehensively funded by the Foundation for Research Development of the CSIR in respect of research on the B-scale. He is also the only individual at the Faculty of Veterinary Science, University of Pretoria to be funded at this high rating. Furthermore, 3 out of the 4 C-rated research workers at this faculty, are from his department. He is either sole author, senior author or co-author of 72 scientific publications. The publications of his post-graduate students, of whom there have been 8 doctoral and 4 masters candidates, number about twice this number. He has thus had a remarkable influence on veterinary parasitology in this country.

As a man's man and a most likeable rebel in our profesion, he has never been afraid to differ openly from people if he was convinced of his point whether it was on scientific grounds or just common horse sense. This included not only his juniors and peers, but also his superiors – especially the directors and deans under whom he served. His own words are: "I went through life insulting people". However, and most important, he always did this in an extremely good spirit.

He retired at the age of 62 in June this year and is now *professor emeritus* and has received the singular distinction of being re-appointed as a full-time research worker by the University of Pretoria for 3 years in order to continue his work.

He has 4 children and 5 grandchildren by a previous marriage and recently married Mrs Norah Donelly.

In view of his considerable and sustained contribution to veterinary research and his investment in the future of the profession through his undergraduate teaching programme and post-graduate guidance, Richard Karl Reinecke is a most worthy recipient of the Gold Medal of the SAVA for 1986.

AWARDS **TOEKENNINGS**

SILWER MEDALJE VAN DIE SAVV VIR 1986
SILVER MEDAL OF THE SAVA FOR 1986

JOHN CHRISTOPHER AUSTIN

The third Silver Medalist of the Association in acknowledgement of outstanding long-term service to and advancement of the veterinary profession or calling is, very fittingly, John Austin.

He was born in Johannesburg on 11 September 1940 and attended King Edward VII School, Johannesburg, where he matriculated in 1957. He obtained the degree BVSc at the University of Pretoria in 1963.

He was in private practice in Johannesburg from 1964 – 1971 and since 1972 has been Consultant Veterinarian to the University of the Witwatersrand.

He was involved in the design, briefing and tender specifications of inter alia the Biomedical Resources Centre, the New Medical School Laboratory Animal Facility and the New Biological Sciences Building Central Campus Animal Facility. From 1976 to date he has been consultant in this field at 7 other laboratory animal facilities including the Universities of Durban-Westville and Potchefstroom and the SA Institute for Medical Research's new Serum and Vaccine Laboratories.

He has been Director of Central Animal Service at the University of the Witwatersrand since 1982; he is honorary professor in the Department of Veterinary Public Health at the University of Pretoria and Honorary Lecturer at both the Department of Experimental Odontology and the Department of Physiology at the University of the Witwatersrand.

Dr Austin has excelled in his contributions in the two interrelated fields of Laboratory Animal Science and Animal Welfare.

It is generally accepted that all animals are kept for the benefit of man. Veterinarians are basically trained to protect and promote the health and welfare of animals and, by so doing, contribute directly to improving the well-being of the human population. Modern veterinary science is finding the delivery of veterinary services expanding dramatically in many non-private-practice areas e.g. laboratory animal medicine, wildlife medicine, research, regulatory and consumer protection.

Both the State and the public in general have shown a substantial increase in interest and concern for the welfare especially with regard to intensive food animal operations, the use of experimental animals in research, preservation of endangered wildlife species, and the role of companion animals in society. In its broadest sense animal welfare is regarded by many as one of the most important sociological issues of this decade.

The role of the veterinary profession in these fields is of paramount importance and Dr Austin has been very successful in creating and stimulating awareness of this in both veterinary practice and education. He did this in his typical clear, logical, fearless way, regardless of the possible ridicule of colleagues and has changed the profession's thinking in many regards.

Over the past ten years, he has made significant contributions to the veterinary profession in relation to non-private-practice services. Dr Austin has spearheaded the SAVA's activities and policies regarding animal welfare and matters relating to laboratory animals. As a member of the SAVA, Dr Austin has served the profession unselfishly, admirably and with dedication in the following capacity:

* Chairman and Co-ordinator of the SAVA Liaison with the Federation of SPCA's Committee.
* He was responsible for drafting of "Blue Book" with regard to the interactions between the SPCA and the veterinary profession.
* He drafted the SAVA policy documents regarding rodeos, cosmetic surgery and the use of experimental animals.
* He organised and co-ordinated the formulation of a booklet and posters concerning guidelines for the use of laboratory animals in schools.
* He served in an advisory capacity on animal housing and drafted the guideline booklet for kennels.

Dr Austin has made a significant contribution of furthering the ideals and image of the profession. His efforts and dedication have been especially noteworthy in the area of the biomedical sciences where the humane use of experimental animals has been utilised to further the aims of human medicine and dental care and thereby, the well-being of man. Dr Austin's vast experience and, obviously, unselfish professional service in the area of laboratory animal science and biomedical

research is well substantiated by his impressive list of 71 scientific publications, the 120 papers he read at scientific congresses and the numerous committees he has served on. He is also a founder member of the South African Association for Laboratory Animal Science and he has played a significant role in developing the Diploma Course in Laboratory Animal Technology at the Technikon RSA.

Dr Austin has earned respect and appreciation from colleagues in the veterinary and other health professions and has set a professional standard of which the veterinary profession can justifiably be proud. There is no doubt that Dr Austin's efforts have directly and indirectly contributed to the rapidly expanding career opportunities being offered veterinarians in the field of laboratory animal medicine.

John married a fellow veterinarian, Dr Georgina Crewe, and they have two children.

By way of personal sacrifice, unselfish service, professionalism, example, persistence, diplomacy, forethought, integrity, deep sincerity and compassion, Dr Austin has established himself as a leader in his field in the RSA. By so doing, he has served the veterinary profession with unequalled distinction in the important areas of animal welfare and laboratory animal medicine and he is unreservedly awarded the Silver Medal of this Association.

JACK BOSWELL TOEKENNING VIR 1986
JACK BOSWELL AWARD FOR 1986

RODRICK ATTWOOD WILSON

This award for 1986 in acknowledgement of unstinting dedicated service to the Veterinary Profession through the South African Veterinary Association is awarded to Dr Ricky Wilson.

He was born on 15 June 1939 and matriculated at Rondebosch Boys' High School, Cape Town, in 1956. He obtained the degree BSc(Med) at Stellenbosch in 1959 before qualifying as a veterinarian at Onderstepoort in 1963.

After two year's of locums he became State Veterinarian first at Baberton and later at Dundee but then moved to Caledon in 1965 where he has been in private practice ever since.

He has always had the Association at heart, having been Chairman of the Western Cape Rural Practitioners' Group from 1978 – 82, Vice Chairman of the Cape West Branch in 1980 – 82 and Chairman of this branch from 1982 – 85. In addition he was National Chairman of the Rural Practitioners' Group during the period 1980 – 83 and a very capable Chairman of the Federal Council's Advisory Committee on Ethical Matters from 1982 – 85.

He is an active member of the SA Society of Animal Production and has been intimately involved with Organised Agriculture. Here he is a member of several agricultural unions and study groups and a committee member of the West Cape Branch of the SA Society for Agricultural Extension. He has been the organiser of numerous veterinary orientated farmers' days where the emphasis has been on herd/flock health.

Ricky is a firm believer that the rural veterinary practitioner is, and should be, the most important cog and leader in the livestock production industry and that armed with a veterinarians training we are the persons that should co-ordinate all the facets affecting animal production. This belief he enthuses to people he comes in contact with.

It is also evident that he is a believer that involvement starts at a grass-root level and that challenges or problems should be well researched, and then discussed with as many colleagues as possible, often obtaining the farming community's viewpoint about the particular issue and that only then does one present the proposal, suggestion or innovation to the Branch, Group or Association.

He has been nicknamed "the farmyard philosopher" by many of his colleagues as he believes that any new veterinary practice or innovation must be put before a representative group of farmers for their comments.

Together with Dr Bill Sykes he dedicated a tremendous amount of thought, time, effort and personal involvement towards the SA Veterinary Foundation's efforts to investigate the plight of rural practice and from which the concept of "Herd/Flock Health Schemes" was born.

Under the Presidency of Prof P Belonje he served on the Association's Sub-Committee that had numerous meetings with the South African Society for Animal Production.

Together with his partner, Ian Herbst, they researched and initiated the concept of the veterinarian's involvement in the sheep production industry and they are considered the leaders in the various aspects of flock health, management and production. This knowledge they have open haertedly shared with the profession and numerous rural practitioners throughout the Republic have been motivated to become involved within the sheep industry at the same level. They have also organized numerous practical seminars and lectures for their colleagues on the same subject.

Ricky's grass-root involvement as a production orientated veterinarian has made him a very popular speaker at farmers' meetings and he is very often invited to address meetings many hundreds of kilometres away from their practice and on all occasions his belief in a veterinarian's involvement within the livestock industry is enthused to his audience.

It is clear that he devotes an enormous amount of time and involvement with Organized Agriculture and farmers at their level of interest by delivering lectures at their meetings and serving on their committees.

He believes that in the past veterinarians have kept

themselves too aloof from the farming community and their way of life. Veterinarians have not always recognized the frustrations and problems farmers experience at farm level and hence the farmers (our clients!) are often hesitant to adopt new veterinary procedures or innovations.

He is always most approachable regarding advice and the dissemination of knowledge to his collegues. This he does in an openhearted way for those in all spheres of veterinary science, especially those facets related to rural practice.

He married Julie Bartels and they have two children.

This full-blooded rural practitioner richly deserves this Association's in-house award for his unstinting service to the profession.

NAVORSINGSTOEKENNING VAN DIE SAVV VIR 1986
RESEARCH AWARD OF THE SAVA FOR 1986

I.G. HORAK

The sixth recipient of this award is Prof Ivan Gerhard Horak, Professor and Director of the Tick Research Unit of the University of Rhodes, Grahamstown.

He obtained his veterinary degree at Onderstepoort in 1957 and was manager at Mkuze Estates up to 1960. He then turned to helminthological research at the Veterinary Research Institute at Onderstepoort where he worked for 7 years and also obtained his D.V.Sc. in 1966. He then became Director of Research and Development of the firm Merck Sharpe and Dohme for the next 7 years. He returned to Onderstepoort in 1974 but this time as Senior Lecturer and later Associate Professor in Parasitology at the Veterinary Faculty where his main interest was Entomology. He obtained a second doctor's degree, PhD (Zoology) through the University of Natal in 1980. In 1982 he took up his present position at Rhodes University where he has built up a most productive and active unit extensively funded by various organisations but *inter alia* with comprehensive funding by the Foundation for Research and Development of the CSIR.

Prof Horak has some 75 publications to his credit and the Awards Committee in scanning veterinary publications of members of the SAVA came across a remarkable series of publications on "The Parasites of Domestic and Wild Animals in South Africa No's I – XVII" which appeared in the Onderstepoort Journal of Veterinary Research from 1977 – 1984 and for which this award is now presented to him.

These publications deal with extensive surveys of the parasites, both internal and external, of domestic animals – cattle, sheep, goats, pigs, dogs – and of wild animals – blesbok, impala, wildebees, zebra, springbok, vaalribbok, bontebok and warthog.

These surveys, involving 1 280 animals, have made a tremendous contribution to our knowledge of the seasonal prevalance, host preferences and geographical distribution of the parasites under different farming practices, thereby providing basic information on the epidemiology of the parasites. In addition, many new species and new host records have been reported. The original surveys were carried out at different localities in the Transvaal but since then, work has commenced and is continuing in the western and eastern Cape, Natal, Orange Free State, as well as S.W.A./Namibia, and the numbers of animals now involved make the 1 200 mentioned previously, pale into insignificance. As examples may be mentioned that papers in press at present deal with 343 sheep, angora goats and calves, while another in the Mountain Zebra National Park involves 287 animals such as larks, chats, guinea fowl, francolin and rodents.

In 1977 he published a classical report on the epidemiology of *Oestrus ovis* in sheep. He examined 542 heads of sheep and found that 73,4% were infested. The highest burdens occurred in May and June and the lowest in September. Mature larvae that pupated from the end of March to mid-August did not develop into viable flies. The length of the pupal stage decreased from 50 days in August to 25 days in mid-summer (December – January) and then again lengthened to 50 days in May. From this data he concluded that sheep were repeatedly infested during the spring, summer and autumn, and that the flies survived as 1st instar larvae in the sheep from June to August, when climatic conditions are unfavourable. They leave the sheep to pupate and emerge as flies in October, thus beginning the cycle again.

It has been known for some years that nematodes of cattle and sheep undergo hypobiosis, i.e. retarded or arrested development, but he showed that this also occurred with the nematodes of African antelope. In cold countries such as Canada, the stimulus for arrested development is the low temperatures that prevail there, while here in the Republic the main stimulus is drought. He pointed out the similarities between the arrested development in *Haemonchus contortus* and diapause of *Oestrus ovis* in sheep, and concluded "that seasonal ar-

rested development in nematodes is similar to diapause in insects. It is triggered by stimuli of which temperature and to a lesser extent photophase are probably the most important, acting on the free-living stages and resulting in inhibited development in a subsequent stage of the life cycle. Non-specific arrested development occasioned by host- or parasite-related factors is similar to quiescence in insects, in that the immediate environment exercises a restraining influence''.

At a time when game and domestic stock are increasingly being run together on farms, his work also highlighted the very real danger of transmission of internal and external parasites to each other.

This award which is aimed at stimulating research of the highest quality is perhaps a little belatedly but certainly most deservedly, given to a most worthy and honoured member of our Association for this outstanding series of articles.

AWARD TOEKENNING

CLINICAL AWARD OF THE SAVA FOR 1986
KLINIESE TOEKENNING VAN DIE SAVV VIR 1986

CHRISTOPHER HARRY BADEN MARLOW

The sixth recipient of this prestigious award in acknowledgement of clinical excellence and the first rural and equine practitioner to get it, is Dr Chris Marlow of Cradock.

He was one of the four to acquire the BVSc degree with distinction at Onderstepoort and he did so in 1956; also receiving the Clinical Medal that year. In 1985 he was awarded the degree MSc.Agric (Stellenbosch) *cum laude* for his work entitled "The oestrus cycle, mating practices, conception rates and foetal losses in Thoroughbreds in the Eastern Cape Province."

In 1957 he started a private practice with a radius of up to 400 km in Cradock which at this stage was regarded as the heart of the SA Stud Breeders of Thoroughbreds.

Because of the vast distances he had to travel he was one of the first to introduce preventive medicine schemes with emphasis on reproduction, production, nutrition and management of all species, including chinchillas, in his area. Artificial insemination was introduced on a large scale to Angora, Merino and cattle breeders.

In 1956 a preventative medicine approach was started in earnest in equines, but with emphasis on the individual and not a group approach. Chris has the best and most comprehensive patient record system on Thoroughbreds in this country. He has provided an exceptional service to his clients and has always been an inspiration to his colleagues. He is regarded as a world authority on the interpretation of uterine biopsies and is presently acting as a referral practice for many private practitioners in this respect.

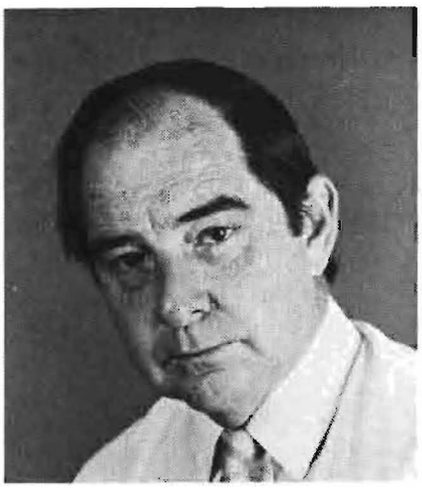
He plays an active role in veterinary education by receiving students into his practice since 1963.

He is also active in the Association being a member of different groups and branches of the SAVA – in particular the Equine Practitioners Group where he is Chairman of the Stud Health Committee. He also belongs to the Thoroughbred Breeders' Association and the South African Endurance Riders' Association.

He has given 9 presentations at Scientific meetings and has 4 publications to his credit.

He has fulfilled all the criteria of this award and through his contribution towards clinical excellence, study, research and implementation of new ideas is indeed an example for colleagues.

Clearly the status of the profession has been advanced in the eyes of the public through his attempts.



E.M. NEVILL

Dr Errol Matson Neville obtained his B.Sc. (Agric), Entomology at the University of Natal in 1959, and joined the Department of Agriculture in 1960 as Assitant Professional Officer.

Stationed at the Entomology Department, Glen College of Agriculture, he was engaged mainly in research on the control of the elegant grasshopper.

In 1962 he joined the Entomology Section at the VRI, Onderstepoort, and at present is a Chief Veterinary Research Officer. During 1968 he was awarded a M.Sc. (Agric.) Entomology degree by the University of Pretoria on "Studies of *Parafilaria bovicola* in South Africa".

Although *Culicoides* and *Parafilaria* research have been his major projects over the past two decades, substantiated by some 21 publications on various aspects of these two ectoparasites, his interest in entomology is much wider as is shown by at least ten other publications on a variety of ectoparasites of importance in the veterinary and agricultural fields.

As a senior and experienced entomologist he is involved in diagnostic and identification functions of the section and is also a Technical Adviser to the Registrar for Act 36 of 1947.

Dr Nevill is responsible for all research on insects in the section which includes liaison with the Section of Virology at the VRI on arbovirus work and also the Directorate of Veterinary Services on the collection of arthropod vectors.

He has lectured to B.V.Sc. III students on veterinary entomology and has been external examiner for B.Sc. Entomology (Hons) and B.V.Sc. (Hons) and M.-Med.Vet. students at the University of Pretoria.

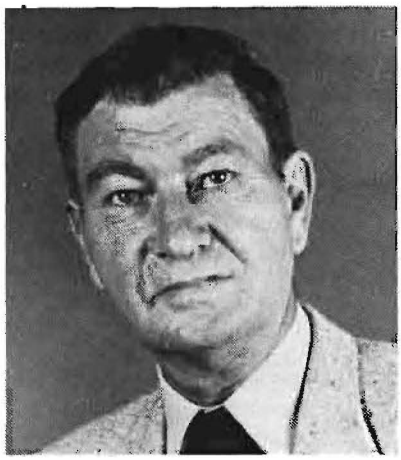
Dr Nevill is regarded as a specialist in his field and enjoys international recognition. He is a member of the WHO/FAO working team on bluetongue, and has been consulted as referee for a research proposal on vectors of the bluetongue virus involving the USA-Israel Binational Agricultural Research and Development Fund. His advice on *Parafilara* research has been requested on two occasions from Sweden over the past few years.

Overseas congresses and research visits include a paper delivered at the World Ceratopogonidae Group, Australia in 1972 and research visits to Australia, the USA and UK on *Culicoides* – arbovirus studies.

Errol Nevill is a highly respected member of the staff of the VRI and has made considerable contributions of great value to our profession as a veterinary entomologist.

He is regarded as a world authority in his chosen field of research and Honorary Membership of the SAVA will contribute appreciably to the reputation of our profession.

HONORARY MEMBER — ERELID



L.A.P. ANDERSON

After matriculating at the Sarel Cilliers Hoërskool, Hobhouse, Dr L.A.P. Anderson or "Lap" as he is popularly known, attended the University of the Orange Free State where he graduated with a BSc. degree in 1950. His first appointment after graduation was as an assistant to Dr M.G.A. Henrici, who, at that time, was investigating the aetiology of geeldikkop. This appointment was to make such a profound impression on him that he devoted his entire professional career thereafter to research on the chemistry of South African poisonous plants. From 1957 – 1971 he conducted his research at the CSRI, to which he was temporarily seconded, as well as at the Research Institute for Soil and Irrigation. He also studied further during this period. A gifted student, he obtained an MSc (*cum laude*) and a DSc in organic chemistry at his *alma mater*. In 1972 Lap was transferred to the Veterinary Research Institute, Onderstepoort where he was promoted to Assistant Director in 1978.

Publishing mostly in chemical journals as he does, most veterinarians can be forgiven for not knowing that this unassuming man, probably is the most distinguished organic chemist working in the field of plant poisons of veterinary importance in South Africa today. He has made major contributions to our knowledge of the bufadienolides of 'plakkies', 'slangkop', and *Melianthus comosus*. Recently he was able to show that the toxic principle(s) of *Thesium linneatum* were also cardiotoxic glycosides. The importance of cardiac glycoside-containing plants to the livestock industry of South Africa need not be elaborated upon. Before Lap and his co-workers at the National Chemical Research Laboratory of the CSIR and Onderstepoort VRI began their chemical investigation of the Crassulucene, only 1 bufadienolide had been isolated from 'plakkies', viz. cotyledoside from *Tylecodon wallichii*. Now at least 13 have been identified – 4 from *Cotyledon orbiculata*, 6 from *T. grandiflora* and 3 from *Kalanchoe lanceolata*. In his latest work he combined chemistry and veterinary science in 3 publications in the *Onderstepoort Journal of Veterinary Research* which have helped to define 'krimpsiekte' more clearly and which have thrown new light on the factors that govern the commulativity of bufadienolides. Including the 9 bufadienolides from *Melianthus comosus*, 4 from *Urginea physodes* and 1 from *Thesium linealium*, he has been instrumental in the identification of no less than 27 bufadienolides, a truely unique achievement.

While pursueing his interest in photosensitivity awakened by his research with Dr Henrici, he isolated a triterpenoid acid from *Lantana camara* that was 6 times more toxic than the then already known icterogenin. Not content with this he systematically changed the functional groups of the triterpene molecule to determine which groups were responsible for toxicity, thereby greatly improving our understanding of the action of these compounds. The chemistry of vermeerbos *(Geigeria* spp.) also received his attention. Some 30 years previously Rimington and his co-workers had isolated the active principle of vermeerbos, vermeeric acid, a dibasic acid that passed readily into the dilactone vermeerin. Many workers had tried unsuccessfully to re-isolate vermeerin before Lap managed to do so. Not only did he isolate vermeerin, but he also determined its structure.

Cardiac glycoside poisoning, photosensitivity and vermeersiekte are rated as 3 of the most important stock poisonings in South Africa, but Lap also found time to study some of the less important poisonings. The toxic principles of 2 neuro-toxic plants, *Cynanchum* sp. and *Sarcostemma viminale* have been isolated and publications on the structures of these compounds are now being prepared. He has also purified the hepatotoxin of *Lasiospermum bipinnatum*, a poisonous plant that causes significant stock losses in winter, especially in the vicinity of Graaff Reinett.

The importance of chemistry to the veterinary profession is well illustrated by the investigation he carried out on the toxicity of *Albizia tanganyicensis* last year. *A. tanganyicensis* or 'paper bark' is a medium-sized tree that grows on the slopes of granite koppies in the Northern Transvaal. Usually it is harmless, because the toxic pods are borne well out of the reach of stock. Outbreaks of albiziosis thereafter can occur only in about August – September when the pods are blown by high winds from the trees. Lap extracted a methoxy derivative of pyridoxin and its acetate from the pods of *A. tanganyicensis* and, noting the similarity between this

toxin and vitamin B_6, he and Mr G.E. Erasmus correctly surmised that it acted as an antagonist to the vitamin. Trials with guinea-pigs confirmed that vit B_6 was a remarkably effective antidote against *Albizia* poisoning.

Knowledge of the molecular structure of a toxin is essential for understanding the mode of action of poisons, as well as their detoxification, metabolism and excretion from the body. Without this knowledge, quantitative methods for the assay of toxins in feedstuffs cannot be devised, and rational methods of treatment often cannot be developed.

Honorary Membership of the South African Veterinary Association is awarded in recognition of his 35 years of distinguished service to the profession during which time he has been instrumental in the isolation and characterisation of more toxins from South African plants than any worker before him.

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