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JOURNAL OF THE SOUTH AFRICAN VETERINARY ASSOCIATION

TYDSKRIF VAN DIE SUID-AFRIKAANSE VETERINÊRE VERENIGING

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BERIA

EDITORIAL REDAKSIONEEL

## CONTINUING EDUCATION — A PERSONAL VIEW OF INFORMAL EDUCATION AFTER GRADUATION

The very concept of continuing education has profound implications. It is an admission, on the one hand, that undergraduate study is incomplete and therefore the attainment of a degree is not a final goal, and, on the other hand, that unaided, personal experience after graduation is also insufficient.

If one accepts the necessity of post graduate education then it is obvious that this does not mean only learning from one's day-to-day experiences and mistakes. Although this experiental learning is lauded by the uninformed, it is a very tedious and inefficient way of learning. Therefore it is imperative to gain further knowledge more effectively. This is done by communicating with those who have had the opportunity to gain the necessary expertise by study and experience.

This communication can occur in various ways, i.e. congresses, discussions, refresher courses and self-study.

#### Congresses

This is probably the worst medium for information transferral. The usual multidisciplinary congress covers a large variety of subjects within each of which something highly esoteric is delivered to a largely uncomprehending or uncaring audience.

But, and this is important, congresses serve an admirable purpose of being a meeting place of colleagues. Here they meet new colleagues, reaffirm old friendships, discuss general matters, relate experiences and, most importantly, increase communication and understanding amongst one another. It would appear more logical to scrap the word "Congress" and rather use the word "Symposium" (Gr. Sumposiazo = to drink together).

### **Discussions**

Here I include informal discussions, second opinions and the general seeking of advice. This is an area where much can still be achieved.

Single-man practices are often so overloaded with work or so remote in distance that informal discussions are a very real problem, but the problem should be attacked with vigour. In large multi-partner practices there is often reasonable internal communication, but, without an input from outside, ideas can easily become stultified and entrenched. It is a pity that in this country each large practice has its own hospital. Communication between colleagues would be far better if hospitals were centralised. One could then have small yet efficient libraries, journal clubs and discussion groups.

Seen from the other side, I have gained the impression that "knowledgeable people" who are asked advice are often rather cagey and appear reluctant to disseminate knowledge. They imagine this may be to their own detri-

ment. This is false reasoning because he that knows that he knows not everything, will always consult again — he that does not know, thinks he knows everything and therefore never consults. I would also appeal to laboratories to make their reports precise, descriptive and educational. By doing this they will not only be serving their colleagues but they will move away from being prescriptive and divorced from reality.

#### Refresher courses (Workshops, Wet Labs)

Refresher courses should be, and usually are, designed around a specific subject with in-depth theory and intensive discussion combined with hands-on practical experience. This is where one can really get to grips with new concepts and techniques one wishes to use in one's practice. The immediate implementation of the new knowledge is essential as a positive reinforcement of the learning process. It is almost useless to go through an exercise at a workshop and then only attempt to apply the expertise much later.

#### Self-study

This is the most important procedure as it encompasses all the previous considerations. But, you must know what you wish to know and, once, having established that, you go ahead to find the information with determination, open eyes and an open mind. Deciding what is important and hence what you wish to know is one of the most difficult tasks facing anyone. Nevertheless, one must identify the knowledge that will cover the most important aspects of your job, that which is less important, and you will have to decide which knowledge is outside of your expertise. Once this has been done, one goes about learning more about the important fields, then narrow down the unimportant even more and refer the rest to someone else. If referral is difficult or impossible it is imperative that you inform the client that you are not really competent in the procedure, but by using general principles you will attempt to help as best you can. It is absolutely no good to give the impression that you know and are competent in all branches of Veterinary Science — this will lead to personal frustration and be a source of bitterness to your client.

Having decided what you wish to do well, it is easy to select the appropriate Symposia, Advisers and Refresher Courses. In addition, you will be able to select the correct textbooks, and replace them at regular intervals. It will also not be difficult to establish which particular journals to subscribe to.

This expertise in specialised fields is nothing new, but should be stimulated and recognised. It should be developed in such a way that in a particular area one should find, and know of, and consult with, and refer cases to these specialists, thereby giving an admirable

service to the local community. In addition, with this interest specialisation one becomes more inquisitive and more inclined to approach research workers and others to investigate real and important problems in your area. This has a spin-off value as it means that research workers will be attracted to real problems and not drift off into the minutiae of own-interest research. I allude here to real research on unimportant esoteric diseases do not confuse this with basic research. Basic research is fundamental, but if real problems are identified, applied research workers will draw the basic scientists into these projects. This is sound, as often the latter find it difficult to motivate their work to others and also in certain fields they lack a sense of belonging. Once having interested others in your problems you will find yourself to be an active participant in problem-solving rather than an inactive, reluctant specimen gatherer and forwarder. These active, knowledgeable people should publish their views, be positive in their criticism of the views of others, and especially debate objectively when they are criticised.

Finally, it is imperative that one keeps track of the knowledge you are accummulating, as well as the source of knowledge. As a professional person one is particularly vulnerable to criticism and you must be able to counter criticism with hard facts. Should a complaint be laid against you, you must be able to defend the allegations with documented proof of your competence in the procedure in question. In other words you must know what you are competent at and must be able to show how you came by that competence. A continually updated curriculum vitae and the keeping of excellent records are therefore essential.

I do not wish to dwell too long on undergraduate experience, but it must be patently obvious from the foregoing that the habit of self-study should be emphasised in the curriculum; no one who has been spoonfed as an undergraduate can ever really know how to undertake self-studies. Also, one must be taught how to study selectively. One must realise at the end of the

course what you know and what you have yet to learn in the branch of veterinary service you are going to enter. There is agreement, I am sure, that while the undergraduate should be taught principles, basic to the whole of veterinary service, it is futile to spend large amounts of valuable time on specific branches of that service which are only going to attract a very small number of graduates.

Together with training in self-study, methods of communication should be taught and practiced. A qualified veterinarian should have no difficulty at all in reaching out for a phone or a note pad to ask for, or give, advice. Even more important perhaps, we should feel far more comfortable in writing to the Journal or Vet News giving of our knowledge, or constructively criticising the opinions of others.

Finally, the undergraduate must never be given the idea that the attainment of the degree is an end in itself. Rather it must be understood that the degree indicates a certain level of competence, enabling the holder thereof to register as a Veterinarian. Once having got his foot on this, the bottom rung of the ladder, he must know that the learning process has to continue, and look forward to doing so with enthusiasm and pride in his chosen profession.

"For the protection of wisdom is like the protection of money; and the advantage of knowledge is that wisdom preserves the life of him who has it" (Ecclesiastes 7:12).

#### **ACKNOWLEDGEMENTS**

I would like to acknowledge all those who have added knowledge and richness to my life and experience. This article, for instance, is certainly not entirely of my own making, but it is my responsibility. Many have influenced me; they, and I, know who they are. And I thank them all.

P.C. Belonje, Department of Physiology, Medical School, University of Cape Town, 7925 Observatory.

## THE USE OF ACUPUNCTURE IN CANINE EPILEPSY

A nine-year-old, male maltese poodle with a body mass of 5,5 kg, was presented with a history of mild generalised seizures. The seizures had started 18 months previously and occurred approximately once per month. Each episode was preceded by prodromal signs and the seizures lasted for approximately 2 - 4 min, after which the dog fully recovered. The patient had received anticonvulsant therapy for 7 months prior to presentation with no improvement in the seizures. The anticonvulsant therapy consisted of phenobarbitone only (Lethal, Lennon LTD), then phenobarbitone and diphenylhydantoin (Epanutin, Parke-Davis) in combination. There were no abnormal findings on physical examination. The diagnostic work-up included haematology chemical pathology, electrophoresis, blood glucose, urine analysis, faecal analysis, radioimmunoassays (cortisol and insulin levels) electrocardiography, cerebrospinal fluid analysis and a computerised axial tomography scan (CAT scan). Analysis of the results of these tests revealed no abnormalities.

No seizures were observed while the patient was in hospital. The patient was started on a course of phenobarbitone syrup (Phenobarbitone vitalet, Adcock Labs LTD) at a dose rate of lmg kg<sup>-1</sup> every 12 h. This dosage was gradually increased and blood phenobarbitone levels were measured at various intervals (Table 1). Chemical pathology (including liver enzymes) blood glucose and haematology were monitored at various intervals as well. From day 75, liver enzyme (ALP, AST, LDH and ALT) concentrations started to rise. At no time during this treatment regimen did the seizure interval, or the severity of the seizures improve.

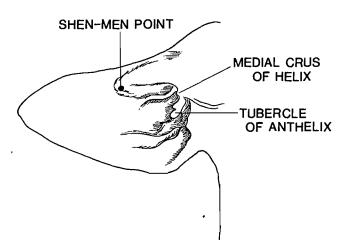
From day 120 the owner was requested to give diazepam (Valium, Roche) at a dosage rate of 0,5mg/kg bid at the first indication of prodromal signs. This treatment regimen exacerbated the seizures.

Blood phenobarbitone concentrations approached top effective serum concentrations, (i.e. before toxic side effects are noticed) and it was decided to wean the dog off all medication. Acupuncture was performed 150 d after first presentation.

Table 1: Dosages of phenobarbitone and serum concentrations

Day dose	Phenobarbitone mg kg <sup>-1</sup> , bìd	Serum concentration μg ml <sup>-1</sup>
1*	1	-
30	1,5	5
45	1,5	8,5
75	3 .	18
120	6	30
135	6	40

1\* — Day treatment was started.



# RIGHT EAR OF THE DOG - CONCAVE SURFACE

Traditional acupuncture for canine convulsive diseases by veterinarians involve the placing of needles in up to ten acupuncture points. The points are situated on the gallbladder, bladder, spleen, liver, conception vessel and governing vessel meridians as described in man and related (transposed) to the dog. In this case a single auricular needle (2 mm) (Auricular needles, Brentwood Marketing) was placed bilaterally aseptically in the Shen-men point (Fig 1). The needle that has a penetrating depth of 1mm is placed over the Shen-men point and advanced until the circular part of the needle is flush with the auricular skin. The needle is held in place by means of elastoplast. The needle can be left in for an indefinite time ranging from twenty min to up to a month or longer. The area should, however, be regularly inspected for redness and should this occur the needle should be removed.

The dog has been free of seizures since treatment was initiated 200 days ago. The needles were left in for 10 days. During this period the dog twice showed clinical signs of restlessness that might have led to a seizure and auricular needles were inserted.

J van Niekerk, Department of Anaesthesiology and Radiology and G N Eckersley, Department of Companion Animal Medicine and Surgery, Faculty of Veterinary Science, Medical University of Southern Africa, 0204 Medunsa

#### HUIDSTEELTJIES IN 'N BOKSER HONDJIE

'n Drie-dae-oue Bokser reuntjie is saam met die res van 'n werpsel van vyf aangebied vir die amputasie van sy stert. Hy het 'n aantal velgroeisels, vanaf 2x1 cm tot 1x0,5 cm, rostraal van die oor op albei kante van sy kop gehad (Fig 1). Die swart masker van hierdie hondjie het verder koudaalwaarts oor sy kop gestrek as wat gewoonlik by die Bokserras aangetref word (Fig 2). Dit was in hierdie areas waar die steeltjies van vel vas was. Die groeisels is onder lokale verdowing chirurgies verwyder.

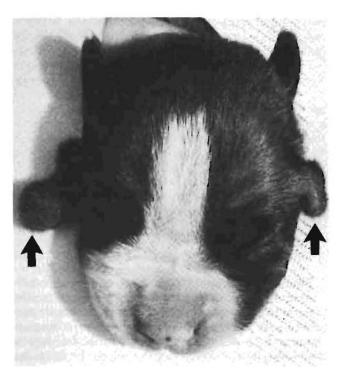


Fig. 1: Die kop van die hondjie met die twee huidsteeltjies (pyltjies).

Mikroskopiese ondersoek van hierdie groeisels het getoon dat daar hiperplasie van die epidermis en dermis, asook hiperplasie van die aangrensende weefsel was. Daar was talle vetkliere teenwoordig, maar geen sweetkliere nie. Daar was meer haarfollikels as wat gewoonlik in die vel voorkom. Die dermis het uit baie bindweefsel, spesifiek meer kollageen en fibroblaste, bestaan. Die mikroskopiese voorkoms van die vel het gelyk soos die van 'n kroniese ontsteking. Omdat hierdie hondjie met hierdie steeltjies gebore was, is hierdie oorsaak onwaarskynlik. Die oorsaak van die hiperplasie in hierdie twee spesifieke areas van die vel is onbekend.

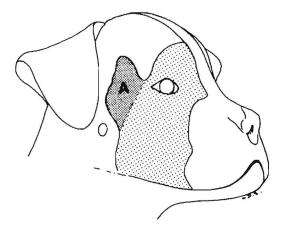


Fig. 2: 'n Lyntekening van die kop van die hondjie op 6 weke ouderdom, wat die grootte van die masker aandui. Area A is die bykomstige area wat in die teks beskryf word.

G J Louw, Departement Anatomie, Fakulteit Veeartsenykunde, Universiteit van Pretoria, Privaatsak X04, 0110 Onderstepoort en Selma J E M van Schouwenburg, Privaat praktisyn, Pretoria

#### STRUCTURE AND FUNCTION OF THE SKIN

G J LOUW, Department of Anatomy, Faculty of Veterinary Science University of Pretoria Private Bag X04 0110 Onderstepoort

Dr O M Briggs gave a comprehensive account of the functions of the skin in his article (JSAVA 58: 229-232). I wish to add some information. The skin has not only a protective function in the mechanical sense, but is also a means of communication between the animal and its environment, and is therefore an important organ of sensation. It has numerous structures which participate in the regulation of the entire organism, such as Meissner corpuscles, end-bulbs of Krause, Ruffini's nerve endings, free nerve endings, nerves, blood vessels, lymph vessels. The numerous functions of the skin are performed by its cells, and their performance depends on the milieu surrounding them. The complexities of blood flow in the skin and the presence of arteriovenous

anastomoses are well documented. The environment plays an important role in alterations in the blood flow to and within the skin. Cutaneous fissures connected to blood vessels and smooth muscle fibres play a role in the swelling of the papillary body. The nerve supply to these muscles is important, and the surrounding environment will, again, affect the transmission of impulses along adrenergic, myelinated and nonmyelinated nerve fibres. Since the organism uses its skin as a means of communicating with the environment, acupuncture as a means of treatment or anaesthesia is successful in animals.

G J Louw, Department of Anatomy, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, 0110 Onderstepoort

#### **DOURINE IN SOUTH AFRICA**

In your September 1986 issue, Williamson and Herr reported on the results of dourine tests done on horse sera received from various parts of southern, Africa (JSAVA 57: 163-165). No positive tests were reported on Natal sera and very few on horses tested in the central Karoo and Western Cape areas.

A large number of tests on Natal horses, as well as horses in the Karoo and Western Cape area, were done by our Regional Veterinary Laboratories at Allerton (Pietermaritzburg), Middelburg (Cape) and Stellenbosch. From April 1981 to March, 1985, 12069 dourine tests were done at these laboratories, using the same methods and interpretations as those used by Williamson and Herr. Our results will supplement the information presented in their article and give a more complete picture of the incidence of dourine in the main horse-breeding areas.

Sera tested positive originated from the magisterial districts of Allerton, Middelburg and Stellenbosch (Table 1).

This represents an overall incidence of 0,65% of the sera tested, but shows a distinct difference between regions, viz: 1,08% in Natal, 0,85% in the central Karoo area and 0,03% in Western Cape area. However, a decrease in the number of serological positives has been noted in the Natal region over this period, viz:

1981/2 18/788 positive (2,3%) 1982/3 11/841 positive (1,2%) 1983/4 7/920 positive (0,7%) 1984/5 2/910 positive (0,2%)

The sera derived from three main sources:

- 1. horses showing clinical signs of the disease
- 2. horses bled for certification for export to neighbouring states
- 3. routine annual testing of horses prior to the breeding season, most of which were Thoroughbreds.

Virtually all stud Thoroughbred mares are tested annually, so that the true incidence of the disease in this breed is known with reasonable confidence. However, the repeat testing of breeding Thoroughbreds (which are invariably negative) each year can therefore lead to an erroneous estimation of the true incidence of the disease. The other breeds are not tested in a routine way, nor are more than a small fraction of the breeding population tested in one year. Therefore, the relatively high incidence of positive tests amongst a relatively

Table 1: Results of dourine tests on horses

District	Positive	Total
Allerton Regional Veterinary Laborato	pry	
Alfred	2	11
Dannhauser	6	68
Dundee	1	50
Estcourt	3	39
Glencoe	1	13
Ixopo	1	48
Mount Currie	6	84
Paulpietersburg	2	24
Pietermaritzburg	1	145
Underberg	8	224
Utrecht	7	19
Horses from 29 other districts	_	2 784
Total	38	3 509
Middelburg Regional Veterinary Labo	ratory	
Albany	2	29
Bloemfontein	5	161
Boshof	, 2	66
Colesberg	1	833
Cradock	1	18
De Aar	23	1 213
Kuruman	2	83
Maclear	1	5
Port Elizabeth	1	289
Vryburg	2	88
Horses from 79 other districts		1 934
Total:	40	4 719
Stellenbosch Regional Veterinary Lab	poratory	
Malmesbury	1	856
Horses from 5 other districts	_'	2 985
	_	
Total:	1	3 841

small sample of the total population of nonthoroughbreds leads one to believe that the incidence of Dourine in these horses may be considerably higher than the average figure above.

However, the true incidence of the disease in the total horse population is unknown, as tests were not conducted in the form of a survey and this letter merely serves to indicate the various findings at our Regional Veterinary Laboratories.

Information submitted by: G C Bishop, G F Bath, P J Posthumus: Allerton RVL; E M van Tonder: Middelburg RVL; P M S Masters: Stellenbosch RVL

A Faul, Director of Veterinary Services, Private Bag X318, 0001 Pretoria

## **BOOK REVIEW**

**BOEKRESENSIE** 

#### A COMPENDIUM OF SMALL ANIMAL SURGERY

R.L. LEIGHTON & K. JONES

1st Edn. The Iowa State University Press, Ames, Iowa 50010, USA. 1983 pp VIII and 274, numerous illustrations, Price £23,95 (ISBN 0-8138-0366-7).

The author, prof. Robert Leighton, former Head of Department of Surgery at Davis, California, is still very active in the field of continuing education despite being retired, and a hand-out prepared for a series of continuing education lectures given in Japan has formed the basis of this book.

As the title indicates, this book contains a concise and comprehensive account of surgical procedures in small animals. Each procedure is described under the following headings: indications, instrumentation, anaesthesia, technique and remarks. The emphasis is greatly on technique; little attention is given to pathophysiology, surgical anatomy and aftercare. The scope of the procedures covered is wide and includes most common and advanced surgical techniques; however the omission of common procedures like ovario-hysterectomy, castration, bone plating, skin flaps and most laryngeal surgery is notable while certain rare and even obscure procedures such as adrenalectomy and palatine mucocoele are included.

One technique, usually well accepted and of proven value, is described for each procedure. There are, however, some original techniques (e.g. quadriceps contracture) and unfortunately also some techniques that should be considered obsolete (e.g. the recommended technique for linea alba closure). The author himself sums his choice of procedures up as 'some old, some new, some unusual' in the preface. The same comment also applies to the choice of materials, such as suture materials and orthopaedic implants. On many occasions, newer and better materials could easily replace the materials advocated.

This book contains numerous relatively simple but very clear line drawings made by the co-author. The text is easily readible. The binding is of the multiple ring type, which is not very pleasing for a bibliophile, but which is in accordance with the primary use of this book, namely as a hand-out for refresher courses. The total absence of references should be seen in the same light.

In summary, this book should be evaluated within the context for which it is intended. The lack of pathophysiological background is worrying. It is also not a state of the art surgical text, which in the present times would necessitate a multi-author approach. It is rather a compilation of surgical techniques favoured by a single albeit very experienced author.

With this in mind the book can not be recommended for veterinary students, but could be of value as a reference for small animal practitioners who are to embark on an unfamiliar surgical procedure and who want a quick refresher before doing so.

Frank J.M. Verstraete

ARTICLE ARTIKEL

## PROPHYLACTIC EFFICACY OF SUSTAINED-RELEASE IVERMECTIN AGAINST INDUCED NEMATODE INFESTATIONS IN CATTLE

M.D. SOLL\*, I.H. CARMICHAEL\* AND R.G. HARVEY\*

ABSTRACT: Soll M.D.; Carmichael I.H.; Harvey R.G. 1987 Prophylactic efficacy of sustained-release ivermectin against induced nematode infestations in cattle. *Journal of the South African Veterinary Association* (1987) 59 No. 1, 9-11 (En). M.S.D. Research Centre, Private Bag 3, 1685 Halfway House, Republic of South Africa.

The efficacy of sustained-release ivermectin was evaluated against challenge infestations of gastro-intestinal nematodes in a laboratory study involving 12 treated and 12 untreated control cattle. A weighted, orally administered osmotically activated device designed to lodge in the rumeno-reticulum and to deliver ivermectin at a dosage rate of approximately 8 mg/day for a 120-day period, was administered to treated cattle. Animals were challenged with infective larvae of Bunostomum phlebotomum and Oesophagostomum radiatum approximately 4 weeks, and with Haemonchus placei, Ostertagia ostertagi, Trichostrongylus axei and Cooperia pectinata approximately 4 and 6 weeks after bolus administration.

No parasites were recovered from ivermectin-treated animals, representing 100% efficacy (p < 0.01) against challenge infestations with larvae of all of the above species. Efficacy against an incidental infestation of *Trichuris* spp was 92% (p < 0.01).

Key words: Ivermectin; sustained-release; cattle; challenge nematode infestations

#### INTRODUCTION

Under conditions in which grazing cattle are continuously challenged with parasitic nematodes single dose anthelmintic treatment is generally curative, rather than prophylactic.

Techniques for sustained release of anthelmintics have been developed to increase the prophylactic value of some remedies. The main aims of this dosage form are to reduce pasture contamination with nematode eggs at strategic times and hence to lower future infection rates and to offset the labour costs associated with repeated treatments.

Ivermectin is highly effective against important cattle nematodes at extremely low dose levels<sup>456</sup>. It is ideally suited to sustained release administration because the mass of active ingredient is not a limitation on formulation. Administered as an erodable intraruminal device it was found to be < 99% effective against challenge infestations of Cooperia oncophora and Ostertagia ostertagi<sup>11</sup>. An osmotically activated pump which delivered ivermectin intraruminally at a controlled zero order rate<sup>10</sup> was also < 99% effective against mature induced infestations of O. ostertagi, C. oncophora and Dictyocaulus viviparus<sup>3</sup> and challenge infections with C. oncophora, Cooperia pectinata, Nematodirus helvetianus, Oesophagostomum radiatum and D. viviparus<sup>3</sup>

This paper describes a trial conducted in South Africa to evaluate the efficacy of ivermectin administered by a specially weighted, osmotically activated sustained-release bolus (Alza Corporation, United States of America) against challenge infestations of a number of common nematode species in cattle.

#### MATERIALS AND METHODS

## Animals and Management

Twenty-four Friesland-cross calves weighing 150-210 kg were included in the trial. Treatment groups were hous-

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ed separately in concrete-floored pens which limited, as far as possible, accidental infection with nematode larvae. A ration of milled lucerne hay was provided, and fresh borehole water was supplied in concrete troughs.

#### Infestations

All trial animals were challenged with approximately 2000 Bunostomum phlebotomum, 1000 O. radiatum, 3000 H. placei, 4800 O. ostertagi, 4800 Trichostrongylus axei and 8000 C. pectinata 27-28 d after administration of the bolus; except for B. phlebotomum and O radiatum, the infestations were repeated 41-42 d after treatment.

The number of larvae per infective dose was determined by initially counting the number of larvae in 10 aliquots (5 aliquots each counted by 2 different observers) from a well-mixed suspension of larvae of each species. From these results the volume of suspension required to deliver the infective dose was calculated.

Infestations were orally induced (with the exception of Bunostomum) by pipetting the required quantity of suspension of larvae onto a moist filter paper disc in a Buchner funnel while negative pressure was applied. The filter paper disc was then rolled up and inserted into a gelatine capsule. Using a rubber hose extension to a standard dosing gun filled with water, the capsule was expelled to the back of the pharynx; the animal was observed briefly to ensure the capsule was swallowed. Bunostomum infestations were induced by pipetting the required volume of suspension onto the skin on the withers.

### Allocation and treatment

Animals were ranked by mass within groups of the same sex. Replicates of two animals each were formed starting with the heaviest pair. Those within a replicate were randomly allocated to treatment groups using a table of random numbers. Those in Group 1 formed an unmedicated infested control and Group 2 received one sustained-release bolus designed to release ivermectin at 8mg day<sup>-1</sup> for 120 d.

Boluses were administered orally on Day 0 using a specially designed balling gun which precluded damage to the bolus. Animals were observed for a few minutes immediately after administration to determine if the bolus was regurgitated.

All calves were tested for the presence of metal objects in the rumeno-reticulum using a metal detector prior to treatment. Bolus retention was confirmed using a metal detector at regular intervals throughout the trial period.

#### Slaughter, worm recovery and counts

Animals were slaughtered by replicate 70-72 d after treatment. Worms were recovered from the abomasum and small and large intestines using the methods described by Reinecke<sup>12</sup>.

Total macroscopic counts were conducted on the abomasal and intestinal ingesta. Parasites were removed as they were counted. Two aliquots, each representing 1/10 of the volume of the abomasal ingesta, abomasal digests and small intestinal ingesta were counted with the aid of a stereoscopic microscope and the numbers of parasites recorded.

#### Statistical Methods

Total counts for each animal were estimated by multiplying the number of parasites counted in each location by the aliquot factor for that location and summing over locations. The data were transformed to the natural logarithm (of (count + 1)) for analysis and calculation of geometric means.

Because no worms were found in any medicated animal, the treated group was compared to the control group using a t-test for means with unequal variances.

#### **RESULTS**

The numbers of parasites recovered are given in Table 1.

Table 1: Efficacy of sustained release ivermectin against challenge infestations of cattle nematodes

	_	Group Me	ean <sup>1</sup>	
	Control	Ivermectin <sup>2</sup>	Efficacy	prob
Bunostomum phlebotomum,				
adult	308	0	100%	< ,01
Cooperia pectinata, adult	655	0	100%	<,01
Cooperia pectinata, L	0,5	0	100%	_
Haemonchus placei, adult	616	0	100%	<,01
Oesophagostomum radiatum,				
adult	232	0	100%	< ,01
Ostertagia ostertagi, adult	1 681	0	100%	< ,01
Ostertagia ostertagi, L	0,6	0	100%	.0855
Trichostrongylus axei, adult	100	0	100%	<,01
Trichostrongylus axei, L	3,1	0	100%	< ,01
Trichuris spp, adult	7,0	0,5	92,2%	<,01

<sup>&</sup>lt;sup>1</sup>Retransformed mean, based on transformation to 1n (count + 1)

No parasites from the induced infestations were recovered from any ivermectin-treated animal, representing 100% efficacy (p<0,01) against infestations of B. phlebotomum, C. pectinata, H. placei, O. radiatum, O. ostertagi and T. axei. Incidental Trichuris spp. burdens were 92% lower in treated animals relative to untreated controls. This difference was statistically significant (p<0,01).

Based on the non-parametric method of anthelmintic assessment of Reinecke<sup>12</sup>, and 'A' claim was obtained (>80% effective in >80% of the treated animals) against challenge infestations of the parasites tested.

Although medicated cattle gained 7kg more than the controls, this difference was not statistically significant (p>0,10).

#### DISCUSSION

This study demonstrates that sustained treatment with ivermectin is highly effective in preventing establishment of challenge infestations of the economically important gastrointestinal nematodes of cattle. The high efficacy is comparable to that of ivermectin administered subcutaneously or orally as a single treatment<sup>456</sup>.

Efficacy against the 4th larval stages of C. pectinata, H. placei, O ostertagi and T. axei was 100%; however, due to variability of these larvae in the controls, differences were not statistically significant for C. pectinata and O. ostertagi. Egerton et al. infer that larvae predestined to become inhibited in the  $L_4$  stage are either inherently more susceptible to ivermectin than those undergoing normal, uninhibited development or that these larvae are metabolically more active prior to time of inhibition and as a result encounter a greater cumulative ivermectin exposure. These authors found that, even at very low doses, sustained-release ivermectin was more effective against inhibited  $L_4$  stages of intestinal nematodes (predominantly N. helvetianus) than against the adults.

The efficacy of the ivermectin sustained-release system in totally eliminating ingested parasites over a period of months has yet to be tested under conditions of natural challenge. This system has valuable potential for nematode parasite control particularly when applied strategically at critical times of the year.

Donald<sup>7</sup> considers that controlled release systems may increase the risk of selecting for resistance compared with intermittent conventional dosing. Several factors may reduce this potential danger. One is to use the device to control parasites over a limited period within epidemiologically based control schemes. Another important factor is the release profile of the device. Because it functions on a diffusion basis, the release rate of the morantel bolus declines throughout its life. This suggests that at some stage drug concentrations may be discriminating between susceptible and heterozygous resistant parasites7. By contrast, another delivery device, the Laby capsule, has a constant release rate and a sharp cut-off when exhausted2. Osmotically activated pumps such as those used to deliver ivermectin in the present studies can be designed to have a similar release profile to the Laby system, with a rapid 'start up' and rapid 'shut down'<sup>10</sup>. Such systems would be expected not to result in prolonged exposure of parasites to sublethal levels of ivermectin. Anthelmintics adminis tered in this way can be expected to have a low risk of selection for resistance<sup>1 13</sup>.

#### **ACKNOWLEDGEMENTS**

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<sup>&</sup>lt;sup>2</sup>Controlled release bolus to deliver 8 mg ivermectin per day

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**BOOK REVIEW BOEKRESENSIE** 

## DIAGNOSTIC RADIOLOGY OF THE DOG AND CAT

#### K.J. KEALY

2nd Edn. W.B. Saunders. Philadelphia, PA 19105. 1987 528 pages, 210 illustrations. Price not supplied (ISBN 07216-1853-7).

More comprehensive than the 1st Edition, and updated, this textbook will provide the practitioner and student with a valuable aid in diagnostic radiology of the dog and cat.

After a brief introduction of "The Radiograph", the author discusses specific disease entities under the following main headings: The abdomen, thorax, bones and joints, skull and vertebral column as well as a "Miscellaneous" chapter in which soft tissue pathology is discussed.

Each section is preceded by a discussion on normal anatomy and specific radiographic technique pertaining to the organ being discussed. Radiographic changes associated with pathological conditions are given and illustrated by means of radiographs and line drawings. A concise description of the clinical signs associated with the pathology is usually given as well as other conditions that may be confused radiologically.

This book is not concerned to any great extent with radiography but rather with the radiographic images associated with specific diseases in the dog and cat.

This book is detailed but concise, easily read, clearly understandable and would be an asset in any small animal practice.

R.D. Gottschalk

ARTICLE ARTIKEL

## EFFECTS OF CHEMICAL IMMOBILISATION ON THE BLOOD COMPOSITION OF IMPALA (AEPYCEROS MELAMPUS (LICHTENSTEIN)

C.S. CHENEY® and J. HATTINGH\*

ABSTRACT: Cheney C.S.; Hattingh J. Effects of chemical immobilisation on the blood composition of impala (Aepyceros melampus (Lichtenstein). Journal of the South African Veterinary Association (1987) 59 No. 1, 13-18 (En). Vaalbos National Park, Sydney-on-Vaal, 8376 Longlands, Republic of South Africa.

The effects of chemical immobilisation on impala (Aepyceros melampus) were investigated. The results indicate that etorphine HCl or Carfentanil should not be used in isolation but that xylazine should be included in the immobilisation "cocktail". The highest dose possible, commensurate with safety, should be used in order to minimise time and distance to recumbency. Body temperature should be monitored and if cooling measures are not effective the animal should be aroused. Either diprenorphine HCl alone or in combination with yohimbine and 4-aminopyridine should be used as antidote/s.

Key words: Stress, chemical immobilisation, capture, Aepyceros melampus

#### INTRODUCTION

Stress-related mortalities arising from capture and/or restraint procedures in wild animals can take peracute, acute or chronic forms<sup>4</sup> 6 12. Deaths have been documented in many species; waterbuck<sup>5</sup>, blue wildebeest, buffalo, impala, zebra, white rhinoceros, sable and elephant<sup>17</sup>, baboon<sup>21</sup>, black rhino, blesbok, eland and nyala678, giraffe3, wolves22, white tailed deer1422, mountain goat<sup>11</sup>, elk<sup>13</sup> and moose<sup>20</sup>, to mention but a few. Whereas deaths are more likely to occur in animals that are chased, physically restrained or mechanically captured, mortalities also occur in chemically immobilised wild animals. Recent work by Hattingh and his associates9 suggests that stress during chemical capture plays a greater role than has been understood. The purpose of the present study was to investigate physiological responses to chemical immobilisation in impala (Aepyceros melampus) because, although chemical agents are used extensively in game capture operations, very little is known about the response of wild animals to these procedures and drugs. In the present context, stress is regarded as the phenomenon of a stressor causing a physiological response.

#### MATERIALS AND METHODS

Thirty-six adult impala (30 male, 6 female) were immobilised in the Malelane section of the Kruger National Park between August 1985 and March 1986. Drug regimens and dosages (from the literature but modified by experience) are indicated in Table 1. Higher dosages were necessary during the summer as the mean body mass per individual increased on average by 22,7% or 10,9 kg  $(47,9 \pm 8,5$  to  $58,8 \pm 8,9$  kg). Winter dosages were found to be marginal or too low for summer when animals were in better condition. Three environmental conditions were recorded during each capture operation ambient temperature, cloud cover (expressed as frac-

tions of eights) and wind (visual Beaufort scale) — to determine their possible contribution towards the response.

Animals were immobilised and blood samples drawn at regular intervals for periods up to 90 min, after which they were weighed and the antidote/s administered (Table 1). A sample of animals was manually restrained for 5 min after arousal to ascertain the effects on the physiological response.

Heart rate (HR) was measured by auscultation and respiratory rate (RR) by observation. Rectal temperature (Tb) was recorded by inserting a rectal thermometer to a depth of 10cm. Heparinised jugular venous blood was centrifuged and the plasma stored frozen. These samples were assayed within 2 days for cortisol (Cortisol Coat-A-Count solid phase radioimmunoassay kit, Diagnostic Products Corporation) and Free T<sub>3</sub> (Free T<sub>3</sub> Solid phase radioimmunoassay kit, Diagnostic Products Corporation).

Plasma was also analysed for lactate (enzymatic UV-method and fluoride/EDTA reagent as anticoagulant, Boehringer Mannheim) and glucose concentrations (Ames model 5581 Glucometer). Full blood was analysed for haematocrit (Hct, microhaematocrit technique) and haemoglobin concentration (Hb, Compur M1000 Haemoglobin-meter, Compur Electronics). Plasma containing 4 mmol  $\ell^{-1}$  reduced glutathione and 5 mmol  $\ell^{-1}$  EDTA was analysed for total catecholamines (T.C.) using high performance liquid chromatography.

For controls, 10 impala were brain shot with a .222 calibre rifle and samples taken as soon as possible (20-40s). With the exception of HR and RR, all variables monitored in the experimental animals were also recorded in the control samples. Normal HR was taken as 74 (sheep and goat<sup>2</sup>) and RR as 20 ( $\pm$  8) from observing free-ranging impala at rest.

Results were compared statistically using the Students t-test and relationships determined by stepwise variable regression analysis using the method of least squares.

### **RESULTS**

No consistent statistically significant differences existed between data from males and females nor between those

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Table 1: Drug regimens and dosages

	Dose	e (mg)		Antidotes (I V)		
Drug/s	Winter	Summer	Diprenorphine HCloo (mg)	Yohimbinexxx (mg kg <sup>-1</sup> )	4-Aminopyridine <sup>oooo</sup> (mg kg <sup>-1</sup> )	N
Etorphine HCI+	0,8	1,2	1,6	-		6
Carfentanil <sup>o</sup>	0,7	1,0	1,4	~		6
Carfentanil plus xylazine <sup>x</sup>	0,6 15,0	1,0 15,0	1,2 1,2	0,1	0,1	6
Etorphine HCI and xylazine	0,7 15,0	1,0 15,0	1,4 1,4	0,1	0,1	6 6

<sup>+</sup> Etorphine HCL (M99, Reckitt and Colman)

Table 2: Immobilisation data (means ± s.d.)

			Recumbency			Antidote	
Drug/s	N	Time to ataxia (min)	Time to (min)	Distance to (m)	N ,	Drug/s	Up (s)
Etorphine HCI	6	4,1 (0,8)	12,9 ( 4,9)	242 ( 92)	6	M	37 ( 15)
Carfentanil	6	3,1 ( 1,7)	8,6 (10,0)	195 (232)	6	M	84 ( 58)
Etorphine HCI and Xylazine HCI	12	2,8 ( 0,9)	6,4 ( 2,6)	155 (114)	6	M.Y.A. M	88 ( 32) 86 ( 12)
Carfentanil and Xylazine HCl	12	2,8 ( 1,3)	5,2 ( 2,5)	96 (104)	6	M.Y.A. M	170 ( 59) 204 (111)

M ≈ diprenorphine HCl

M.Y.A. = diprenorphine HCl, yohimbine and 4-aminopyridine administered together

obtained from animals immobilised in the winter (August to October) or summer (November to March). Results from all animals were thus pooled where appropriate.

## Immobilisation data

Table 2 summarises the relevant data. Animals immobilised with etorphine HCl and xylazine and Carfentanil and xylazine took significantly less time (P < 0.05) to recumbency than animals immobilised with etorphine HCl only. In general, animals immobilised with Carfentanil and xylazine took longer to get up after administration of the antidotes than animals with other regimens.

Table 3 summarises statistically significant correlations between time and distance to recumbency and certain blood variables. In addition, a subjective evaluation of the level of excitement and cortisol and lactate concentrations is shown. In general, the greater the level of excitement after darting and the greater the time and distance to recumbency, the greater the physiological response.

Physiological response to different drug regimens: These results are shown in Fig. 1. Significant differences (\*) indicate that the mean results (over 90 min) for animals immobilised with solutions containing xylazine were different from the mean results of animals only receiving etorphine HCl or Carfentanil (P at least <0.05). The mean results after manual restraint were obtained from 2-8 animals with etorphine HCl/xylazine, 3-7 animals with Carfentanil/xylazine and 1-3 animals with etorphine HCl only.

Three out of 6 impala immobilised with etorphine HCl and 5 out of 6 immobilised with Carfentanil had rectal temperatures in excess of 40°C, the highest being 41°C and 41,9°C respectively. Mean RR and HR were significantly higher in etorphine HCl and Carfentanil immobilised animals than for the other two regimens (P < 005). Body temperature, RR and HR remained elevated throughout the period of immobilisation. Cortisol levels continued to increase during immobilisation in the etorphine HCl and Carfentanil immobilised animals showing significantly higher mean levels than etorphine HCl/xylazine and Carfentanil/xylazine samples (P < 0.05). Mean haematocrit and haemoglobin values were significantly higher with etorphine HCl and Carfentanil than for the other drug regimens (P < 0.05) during the period of immobilisation. Mean lactate levels in animals with etorphine HCl were significantly higher than those for etorphine HCl/xylazine immobilised animals (P<0.05) and Carfentanil/xylazine samples

Carfentanil — (Janssen Pharmaceuticals)

Xylazine HCl — (Rompun, Bayer)

Diprenorphine HCI (M5050, Reckitt and Colman)

xxx Yohimbine — (Centaur Labs)
4-Aminopyridine — (Centaur Labs)

(P < 0.025) but were lower (P < 0.01) than those for animals immobilised with Carfentanil alone.

#### Effects of environmental conditions on physiological responses:

Significant relationships were found to exist between ambient temperature, cloud cover and wind and certain physiological responses. These are shown in Table 4.

#### DISCUSSION

The control values used in the present study were obtained from undisturbed animals killed instantaneously. It is known that this procedure may produce physiological responses. Until remote controlled blood sampling procedures have been applied to impala<sup>10</sup>, these control values are accepted as being a reasonable reflection of what the values are in these animals at rest.

Significant relationships existed between the level of excitement immediately after dart impact and the maximum recorded cortisol and mean lactate levels. Animals that responded with little excitement on darting had the lowest maximum cortisol levels whereas those that responded by running off at speed and having extended flight distances had the highest levels. Animals darted with barbed needles in which the dart remained embedded showed the greatest fright and flight response — the higher cortisol levels in these animals as opposed to animals immobilised with drop-out darts would in-

dicate that the use of barbed darts will elicit a greater response. Lone animals showed greater excitement and flight distances than animals that were darted in a herd. The greater flight distances were accompanied by higher cortisol and catecholamine levels and higher haematocrits and RR and HR although not all relationships were statistically significant.

Significant relationships were also found between time to recumbency and initial haematocrit, mean total catecholamines concentrations, initial cortisol concentrations and mean HR. All relationships were linear, indicating that an increased time to recumbency will result in elevated concentrations. Further, linear relationships were found between distance to recumbency and mean and maximum RR, maximum HR and mean cortisol concentrations. The physiological response is therefore enhanced by both time and distance to recumbency. It is preferable to use the highest dose possible, commensurate with safety, to "drop" the immobilised animal quickly, as low or marginal doses usually result in extended times and distances to recumbency, and, as has been shown, result in greater physiological responses.

Five of the 6 impala darted with etorphine HCl alone would not go down and had to be caught by hand after which they were easily forced down into a recumbent position. An excitable phase was evident and muscle fasciculations were observed in some animals. They were very sensitive to tactile and auditory stimuli. They were not completely tractable during immobilisation

Table 3: Correlations between time and distance to recumbency and level of excitement at darting and certain physiological responses.

/ariables Linear regression		r	p<	N	
* Level of excitement at darting:					
vs max. cortisol concentration vs mean lactate concentration	Y = 19,48X + 64.29 Y = 1.60X + 6.63	0.5084 0.3881	0.001 0.021	36 34	
	1 - 1.00% 1 0.00				
Time to recumbency:					
vs initial Hct	Y = 0.43X + 7.19	0.6619	0.005	33	
vs mean T.C. concentration	Y = 0.42X + 3.99	0.4200	0.029	27	
vs initial cortisol concentration	Y = 1.9X + 18.02	0.3535	0.050	35	,
vs mean heart rate	Y = 1.81X + 69.38	0.4333	800.0	36	
Distance to recumbency:					
vs max, heart rate	Y = 0.08X + 76.53	0.4623	0.004	36	
vs max, respiratory rate	Y = 0.11X + 18.84	0.3824	0.021	36	
vs mean respiratory rate	Y = 0.10X + 12.75	0.4195	0.011	36	
vs mean cortisol concentration	Y = 0.10X + 39.57	0.4958	0.002	36	

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Table 4: Significant correlations between environmental conditions and physiological response.

Variable	Regression equation	r	P<	N	
Ambient temperature:					
vs maximum rectal temperature	$Y = 0.49X \ 0.18$	0.7369	0.001	36	
vs mean rectal temperature	Y = 0.49X 3.07	0.7409	0.001	36	
Cloud cover:			<del>-</del>		
vs mean heart rate	Y = -4.44X + 94.2	~0.4002	0.015	36	
vs mean cortisol	Y = -6.48X + 75.52	-0.4404	0.007	36	
vs mean lactate	$Y = e^{(1.98 - 0.13 x)}$	~0.4740	0.004	34	
Wind strength:					
vs mean ACTH level	Y = -74.82X + 335.50	~ 0.4238	0.016	32	

<sup>\*</sup>A subjective evaluation based on the following:

1 — no excitement — little or no fright response on dart impact.

2 — slight excitement — moved of a few paces and resumed feedi

3 — moderate excitement — ran off a distance and then stopped. 2 - slight excitement - moved of a few paces and resumed feeding.

<sup>4 -</sup> marked excitement - ran off a great distance.

and on occasion struggled and made attempts to stand. Breathing was laboured in some animals, and three impala had transient arrythmias. Animals darted with etorphine HCl alone showed the greater distances and times to recumbency/capture. The antagonist diprenorphine HCL, however, worked very well on animals immobilised with etorphine HCl — the time up after antidote administration being on average 37 s.

16

Five of the 6 impala darted with Carfentanil went down, only one having to be caught by hand. Although immobilised animals were also sensitive to sound and touch and also showed increased muscle tone, this was less pronounced than with etorphine HCl and they were generally more sedate. In the absence of noise and tactile stimuli they lay quietly. Elevated HR, RR and rectal temperature were again a feature. Mean cortisol, total catecholamines, glucose, lactate, haematocrit and haemoglobin levels were higher compared to animals immobilised with a combination of etorphine HCl/xylazine or Carfentanil/xylazine, although this was not always significant. The latter animals generally showed a good response. Relaxation was more than adequate. Distances and times to recumbency were shorter as opposed to animals darted with etorphine HCl or Carfentanil alone. Two out of the 12 animals immobilised with the etorphine HCl/xylazine combination had elevated rectal temperatures (above 40°C) and one of the animals immobilised with Carfentanil/xylazine. The general impression with etorphine HCl/xylazine animals was that although they remained fairly aware of their surroundings they remained relaxed and were not unduly disturbed by local stimuli. With carfentanil/xylazine, impala appeared unaware of surroundings. Tongue protrusion was noted in some animals and increased salivation was a common feature — one known to accompany xylazine administration.

The use of diprenorphine HCL, 4-aminopyridine and yohimbine in combination as an antidote (the latter two for xylazine) is advocated as this appears to work effectively in reversing xylazine sedation. An important feature when using xylazine in combination with either etorphine HCl or Carfentanil is that physiological responses tend to either return to normal quickly after initial deviation from normal or to remain very stable.

From the above it is concluded that etorphine HCl or Carfentanil when used alone to immobilise impala cause greater physiological responses than when xylazine is included in the combination. The use of Carfentanil/xylazine or etorphine HCl/xylazine is therefore advocated, the latter combination being the preferred one because of possible residual effects of Carfentanil (V. de Vos 1987 National Parks Board, Skukuza, personal communication).

Ambient temperature appears to exert an important effect on the physiological responses. Death due to hyperthermia in captured wild animals has been well documented<sup>5</sup> <sup>17</sup>. The stress response is increased with an increase in body temperature and this is very closely linked to ambient temperature, the effect of shade and cooling by wind. These results underline the importance of confining capture operations to cooler hours of the day, attempting to restrict times and distances to recumbency to the minimum, including a muscle relaxant/sedative in the drug 'cocktail', avoiding chasing and keeping periods of immobilisation as short as possible. Body temperature should be monitored regularly and the animal wetted and/or fanned if temperature remains

elevated. If cooling measures do not appear to be beneficial, the animal should be aroused.

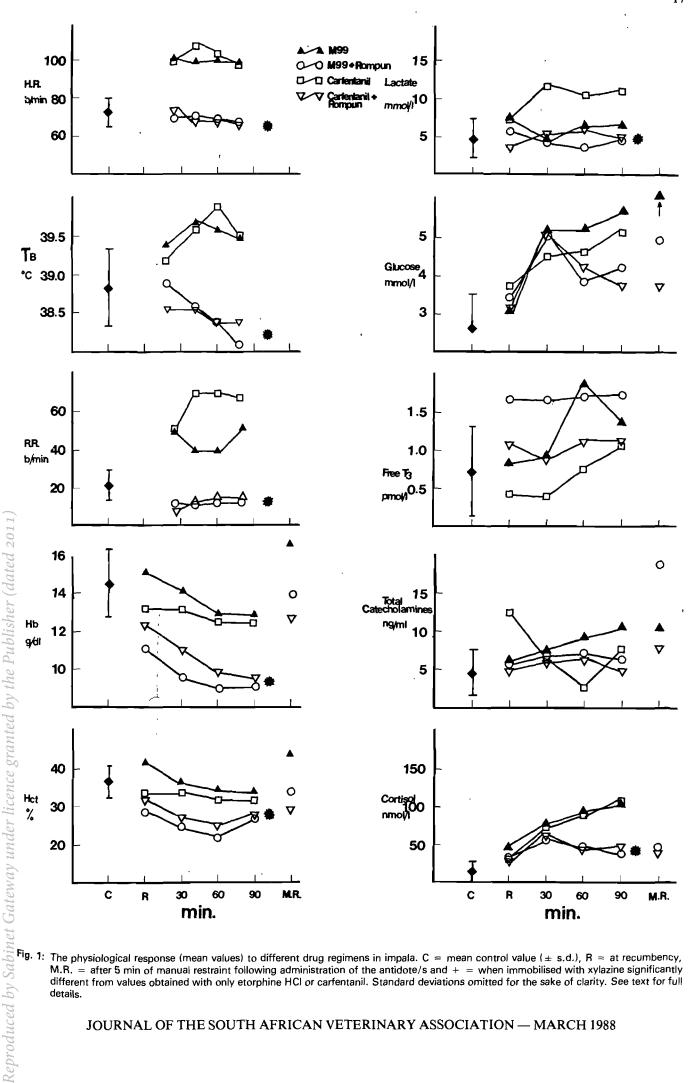
Results obtained from animals manually restrained after arousal show that this procedure constitutes a "severe form of stress". This is in accordance with the findings of Olivier<sup>16</sup>, Presidente et al.<sup>18</sup>, Wesson et al<sup>22</sup> and Drevemo and Karstad¹ who concluded that blood stress indicators were highest in manually restrained animals, intermediate in shot animals and lowest in chemically immobilised deer. Work by Murray et al.<sup>15</sup> on impala showed that confinement in a crush led to 30% mortalities and restraint with tranquilisation to 22% mortalities — most of the deaths were, according to the authors, caused by "fatal stress" during individual restraint. The above points to the fact that wild antelope should, if at all possible, not be manually restrained or handled when not sedated.

#### **ACKNOWLEDGEMENTS**

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The physiological response (mean values) to different drug regimens in impala. C = mean control value ( $\pm$  s.d.), R = at recumbency, M.R. = after 5 min of manual restraint following administration of the antidote/s and + = when immobilised with xylazine significantly different from values obtained with only etorphine HCl or carfentanil. Standard deviations omitted for the sake of clarity. See text for full details.

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ARTICLE ARTIKEL

## THE RELATIONSHIP IN DOGS BETWEEN PRIMARY RENAL DISEASE AND ANTIBODIES TO ENCEPHALITOZOON CUNICULI

C.G. STEWART\*, F. REYERS\*\* and HELENA SNYMAN\*\*\*

ABSTRACT: Stewart C.G.; Reyers F.; Snyman H. The relationship in dogs between primary renal disease and antibodies to Encephalitozoon cuniculi. Journal of the South African Veterinary Association (1986) 59 No. 1, 19-21 (En). Department of Infectuous Diseases and Public Health, Faculty of Veterinary Science, Medical University of Southern Africa, 0204 Medunsa, Republic of South Africa.

Fifty-two serum samples from dogs with primary renal failure were tested for antibodies to Encephalitozoon cuniculi. Twelve were positive as compared to two positive samples from a control group of 42 dogs. There was a statistically significant difference between these two groups which suggests an association between infection with E. cuniculi and the development of chronic renal disease.

Key words: Encephalitozoon cuniculi, renal disease, dogs

#### INTRODUCTION

Encephalitozoon cuniculi is a protozoon parasite that affects a large variety of mammals including rabbits, rodents<sup>7</sup>, blue foxes<sup>17</sup> and cats<sup>27</sup>. Encephalitozoonosis in dogs has been reported from England<sup>20</sup>, Europe<sup>11</sup>, U.S.A.<sup>8</sup> <sup>22</sup>, East Africa<sup>21</sup>, Zimbabwe<sup>13</sup> and the Republic of South Africa<sup>14524</sup>. Since the first reported case in the Republic of South Africa in 1966, encephalitozoonosis has been diagnosed fairly frequently, based mainly on microscopical examination of tissue specimens collected at necropsy. The disease occurs mainly in young dogs between 4-12 weeks of age and has also been recognised as an important cause of losses in breeding kennels<sup>24</sup>. The main clinical signs are a loss in mass combined with an encephalitis-nephritis syndrome<sup>20</sup>.

A number of other infectious agents, including Ehrlichia canis, and Leptospira spp. have been associated with nephritis<sup>12</sup>. The kidney appears to be a target organ in infections with E. cuniculi and chronic interstial nephritis regularly occurs in young dogs following experimental infection<sup>3 5</sup> and in natural cases of encephalitozoonosis<sup>13 20 21</sup>. Although the renal lesion may becomes irreversible in many cases<sup>3 21</sup> leading to uraemia and death 12 19 20 it is difficult to demonstrate the organism at this later stage of the disease<sup>625</sup>.

Azotaemia is defined as an abnormal concentration of urea, creatinine or other non-protein nitrogenous substances in blood and may be due to prerenal, renal or post renal causes<sup>18</sup>. It may be caused by increased production of urea by the liver or creatinine by the muscles or by decreased rate of loss (primarily by the kidneys). Renal failure may be caused by any disorder that damages the functional capacity of 70-75% or more of the nephrons and may be caused by a variety of different causal agents. Renal disease may therefore precede renal failure and likewise renal failure may

precede azotaemia. Blood urea (BU) levels above 30 mmol/ℓ usually indicate the presence of primary renal disease10 whereas prerenal azotaemia seldom gives values above 30 mmol/l. In our experience a value in excess of 20 mmol/ℓ is strongly suggestive of primary renal disease.

In many cases of renal failure it is not possible to establish an aetiological diagnosis. This study was carried out to determine if there was a relationship between E. cuniculi infection and primary renal failure in older dogs.

#### MATERIALS AND METHODS

Azotaemic Dogs: The recorded results of canine BU determinations conducted over a four year period in the Department of Medicine, Faculty of Veterinary Science, Onderstepoort, were examined. The case histories were obtained for all dogs with a BU level above 20' mmol/l. These records were examined and where an aetiological diagnosis was made, the case was excluded. The stored serum samples from the 52 remaining dogs with azotaemia, were then collected. Necropsies had been carried out on 29 of these dogs.

Control Dogs: As a control group, serum samples were obtained from dogs whose BU levels had not been determined or was below 8 mmol/ $\ell$ . It is possible that some of these specimens may have had BU values greater than 20 mmol/ $\ell$ ; however this is unlikely as none of them had a clinical history of disease which would cause a high BU. The samples of all 42 cases had been obtained during the same period as those of the azotaemic group.

Indirect Fluorescent Antibody (IFA) Test: All serum samples were tested for the presence of antibodies against E. cuniculi by means of the IFA test previously described23.

Immunoglubulin (Ig) Determinations: Radial immunodiffusion kits (Miles Laboratories) were used for the quantitative determination of canine IgG and IgM. The test was carried out according to the manufacturer's instructions, except that each serum sample

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was measured accurately with a Hamilton syringe and then added to the relevant well.

**Statistics.** A X<sup>2</sup>-squared test was used to test the significance of the relationship between azotaemia and a serological response to *E. cuniculi*.

#### **RESULTS**

There were 12 dogs from the azotaemic group and 2 dogs from the control group which had antibodies to E. cuniculi. There was a significant difference between these two groups (P < 0.05).

Five of the 29 azotaemic dogs necropsied had antibodies to *E. cuniculi*. Four of the 5 showed evidence of interstitial nephritis which was confirmed on histopathological examination in 3 cases. The fifth dog showed pyelonephritis at necropsy.

Examination of the records of the 2 dogs in the control group which had antibodies to *E. cuniculi*, showed that one had died of old dog encephalitis and the other of distemper with focal renal lymphocytic infiltration seen on histopathological examination. The serum antibody titres of these two animals were 1/80 and 1/60, respectively.

The clinical pathology findings and immunoglobulin levels from the serologically positive animals in the azotaemic group are shown in Table 1. The specific gravity (SG) of the urine varied from 1,010 to 1,022, the serum creatinine levels were high; and the protein levels in the urine were slightly to moderately elevated in most cases, but high in one animal having a high protein content in the urine. The IgG levels in the serum varied from 12 000 to 26 000 mg  $\ell^{-1}$  and the IgM level from 820 to 4 200 mg  $\ell^{-1}$ .

#### DISCUSSION

Previous results have shown that a titre > 1/20 usually represents infection with E. cuniculi  $^{23}$ . As 23% of the azotaemic group had titres of 1/20 and over, these results suggest that E. cuniculi may play a significant role in the aetiology of chronic renal failure. Determination of the underlying cause(s) of azotaemia is of great clinical importance since this information will significantly influence therapy and prognosis. In many cases, however, it is not possible to give an aetiological diagnosis. In this study cases were specifically chosen

where it had not been possible to reach an aetiological diagnosis. The diagnosis in these cases was limited to the pathophysiological level<sup>19</sup>.

The selection of dogs in the azotaemic group with a BU level 20 mmol  $\ell^{-1}$  would in our experience eliminate dogs with prerenal azotaemia. The detection of concentrated urine with SG > 1,030 indicates a population of functioning nephrons adequate to prevent signs caused by primary renal failure<sup>16</sup>. In the azotaemic group 42 dogs had urine SG values below 1,030 and 2 dogs had a value of 1,032. The SG values of the remaining 8 dogs were not available. The two dogs with urine SG's values of 1,032 had BU levels of 32 and 98 mmol  $\ell^{-1}$  respectively. These low urine SG values combined with azotaemia confirmed the presence of primary renal disease<sup>10</sup> in the majority of dogs in the azotaemic group and in all the dogs positive to *E. cuniculii*.

Four of 5 dogs on which necropsies were performed showed evidence of chronic interstitial nephritis (CIN), the aetiology of which has not been totally clarified. Most evidence suggests that CIN follows a non-fatal episode of *Leptospira canicola* infection<sup>14</sup>. However, acute leptospirosis is not commonly diagnosed at the Outpatients Clinic at Onderstepoort the source of our specimens (personal observations). The disease does occur from time to time, but appears to be more prevalent in the coastal areas<sup>2</sup>.

The exact mechanism by which *E. cuniculi* causes interstitial nephritis has not been studied. Both young and adult dogs experimentally infected with *E. cuniculi* failed to develop clinical encephalitozoonosis<sup>3</sup>. However, on histopathological examination interstitial nephritis with plasma cell infiltration occurred in all cases. This lesion could be seen up to 350 days after experimental infection<sup>25</sup>. Demonstration of the parasite in these lesions was difficult particularly in older dogs<sup>5</sup> <sup>25</sup>. Whether some of these cases would eventually have developed renal failure is not known, but the serological results obtained in this study suggest that it may have been possible.

In foxes it has been shown that in utero infection is necessary for clinical signs of encephalitozoonosis to develop. Infections which occur post-natally result in mild subclinical disease. In young fox cubs interstitial nephritis can be demonstrated<sup>15</sup>. Unlike dogs, infection of adult foxes results in endometritis whereas interstitial nephritis does not seem to develop<sup>16</sup>.

Young puppies with encephalitozoonosis pass large

Table 1. Clinical pathology findings in 12 azotaemic dogs serologically positive to Encephalitozoon cuniculi

<i>E. cuniculi</i> titre	Age (years)	Blood Urea mmol ( <sup>-1</sup>	Creatinine $\mu$ mol $\ell^{-1}$	Urine S G	Urine protein**	Blood $lgG$ $mg \ell^{-1}$	Blood IgM mg $\ell^{-1}$	Necrospy performed
160	7	85	312	1,014	+	20 000	2 500	Yes
80	3	79	783	1,014	++	16 000	4 200	Yes
80	Adult	90	1378	1,013	+	19 000	2 100	Yes
40	4	61	ND*	1,017	+	14 000	2 200	Yes
20	6	52	705	1,011	+	21 500	3 100	Yes
80	Adult	37	ND	1,019	+	24 000	820	_
40	7	26	ND	1,010	+	24 000	1 000	_
40	16	34	444	1,022	+++	18 000	2 200	
40	7	29	ND	1,011	++	20 000	4 200	o
40	7	53	617	1,011	+	24 000	820	_
40	6	52	571	1,010	++	26 000	2 800	_
20	Adult	163	1070	1,019	+	12 000	3 400	-

<sup>\*</sup>ND = Not done

<sup>\*\* =</sup> Scale of - to + + + (Ames multistix) Ames Corp, Iowa.

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numbers of *E. cuniculi* spores in the urine<sup>5</sup> and where close contact occurs between dogs, as in kennels, many animals become infected. Stewart et al<sup>23</sup> showed a 75% prevalence of antibodies to *E. cuniculi* in breeding dogs following an outbreak of the disease in a breeding kennel. None of the adult animals developed encephalitozonosis. In this instance horizontal infection appeared to be widespread. How many of these animals develop sub-clinical interstitial nephritis was not known, but, experimental evidence suggests that the incidence might have been high<sup>3</sup>. It is conceivable that at least some of these animals may have developed chronic renal disease later on in life.

Tizard<sup>26</sup> gives the serum IgG levels of dogs as 10 000-20 000 mg  $\ell^{-1}$  and 700-2 700 mg  $\ell^{-1}$  for IgM. In the azotaemic group positive to the IFA test for antibodies to *E. cuniculi*, 5 dogs had above normal IgG levels and 4 dogs had above normal IgM levels (Table 1). These increases were only slightly above normal suggesting that hypergammaglobulinaemia is not an important finding in adult chronically infected dogs.

#### **ACKNOWLEDGEMENTS**

Prof R K Reinecke, Department of Parasitology, University of Pretoria, is thanked for the facilities to carry out this work.

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BOOK REVIEW BOEKRESENSIE

## THE RUMINANT ANIMAL DIGESTIVE PHYSIOLOGY AND NUTRITION

D.C. CHURCH (Ed)

Prentice Hall, Englewood Cliffs, New Jersey 07632. 1988 pp IX and 564, 273 illustrations and 124 tables, Price not given (ISBN 0-8359-6782-4).

This book was previously published in two volumes under the name Digestive Physiology and Nutrition of Ruminants. It is a major multi-author book with contributions from 30 leading authorities and researchers in the field of ruminant physiology and nutrition. In its new format it can be expected to become a standard text in its field for some time to come.

The text is subdivided in three parts. Part I deals with the basic digestive physiology of ruminants including a chapter on comparative anatomy of concentrate selectors, grass and roughage eaters and an intermediate type of ruminant. The growth and development of the ruminant digestive system is also comprehensively covered. The physiological background to motility of the gastro-intestinal tract is handled in detail and the rumen ecosystem is very adequately covered.

Part II deals with the consumption, metabolism and requirements of ruminants under a variety of conditions and also has a chapter on the nutrient needs of ruminants versus monogastric species.

Part III deals with nutritional problems related to the gastro-intestinal tract like bloat, acidosis, nitrate and urea toxicity and acute pulmonary oedema and emphysema. It also has chapters on metabolic problems related to nutrition like milk fever, hypomagnesemia and bovine ketosis which are covered in a fairly detailed way while the fat cow syndrome and urinary calculi are also discussed but in less detail. The effect of stress on nutrient needs is also explored.

The last chapter in this section is on therapeutic nutrition and superficially explores the nutrient imbalances in stressed and diseased animals and feeding regimens for patients under treatment. This is one chapter in the book that will benefit by the inclusion of more hard facts. Admittedly this is more a reflection of the dearth of information or this subject in the literature than on the ability of the author.

In general it can be said that most of the chapters are fairly comprehensive and up to date, the book is well indexed and the references are current. The authors did not use SI units which may be a minor irritation for South African readers.

Veterinary students, academic clinicians, research workers and to a lesser extent, veterinary practitioners will find much of the information applicable and this book should surely grace the shelves of all libraries specialising in veterinary, agricultural and zoological literature as a standard reference work.

G.H. Rautenbach

ARTICLE ARTIKEL

## A REVIEW OF *LEGIONELLA PNEUMOPHILA* IN HORSES AND SOME SOUTH AFRICAN SEROLOGICAL RESULTS

C.A. WILKINS\* and N. BERGH\*

ABSTRACT: Wilkins C.A.; Bergh N. A review of Legionella pneumophila in horses and some South African serological results. Journal of the South African Veterinary Association (1988) 59 No. 1, 23-26 (En). Department of Infectious Diseases, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, 0110 Onderstepoort, Republic of South Africa.

An examination of the sera of 329 horses for *L. pneumophila* antibodies revealed a much lower exposure rate than that reported in the United States of America. Further serological investigations of persons closely associated with a sero-positive horse indicated that the horse could not be considered to be a source of infection but that both humans and animals were probably exposed to a common source of infection. The results showed that 192/329 (58.4%) of the sera tested negative, 114/329 (34.7%) had end-point titres of 1/2, 22/329 (6.7%) end-points of 1/16 and one an end point of 1/256 (0.3%). Serological testing of the people closely associated with horses showed that out of 22 people, 3 had a positive end-point titre of 1:64 and only one person showed an end-point titre of 1:256.

Key words: L. pneumophila, equines, serological, South Africa

#### INTRODUCTION

L. pneumophila is a ubiquitous organism and has been isolated from a wide variety of sources, especially from water and soil. It is therefore reasonable to expect that animals would be exposed to the organisms to a greater extent than the human population. As a result of this a number of serological surveys have been undertaken to determine the incidence of antibodies against L. pneumophila in a wide variety of species. Results obtained in the USA<sup>5</sup> indicated that 31,4% of equine, 5,1% bovine, 2,9% porcine, 1,9% ovine, 1,9% canine and 0,5% caprine sera were positive. In the same investigation the occurrence of human positive sera was 0,4%. As a result of the high incidence of positive reactions in equine sera, it was decided to investigate the situation in horses in South Africa.

Subsequent work in the United States has shown that the organism does not cause clinical disease in, nor is shed by horses. The horse does not play any role in the clinical maintenance of the disease. Infection of humans from horses is unlikely, more probable is infection of both human and horse from a common source.

The world has now had a decade of Legionnaires' disease and since the first outbreak of Legionnaires' disease in Philadelphia in 1976, it has become one of the most intensively studied diseases and organisms<sup>18</sup>. There are presently no fewer than 23 recognised Legionella species with 10 serogroups of L. pneumophila, including serogroup I which was the organism associated with the original outbreak of Legionnaires' disease<sup>17</sup>. It was soon established that the major mode of transmission was airborne. The first ideas were that the organism was associated with disturbed soil and water. Subsequently the organisms were isolated from water in a large variety of conditions, stretching from the hot springs of Yellowstone Park, to the rain forests of Puerto Rico, numerous lakes, estuarine waters, irrigation sprinklers in Israel, industrial cooling fluids, power sta-

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tion cooling towers and circulating water, not only in large buildings such as hotels, but also in domestic supplies. Sludge systems in effluent handling and stationary water tanks also provide suitable conditions for the multiplication of Legionella. The creation of aerosols in water seems to be the most likely mode of transmission of the organisms<sup>10</sup> 12 23. The organism is stable in 5 micron water particles which is the correct size to reach the alveoli so that water aerosols are the most probable method of transmission. As the organism is widely distributed in nature in water, mud and soil and transmitted by aerosol, there is every reason to expect that animals under these conditions would be exposed to infections with L. pneumophila. Several serological surveys were carried out to test this hypothesis. Snowman et al22 investigated the role of indoor and outdoor occupations in the sero-epidemiology of L. pneumophila infections in humans. They found no significant differences between these occupations.

The disease is known to occur worldwide. The bulk of reports of outbreaks of the disease have come from the Northern Hemisphere in so-called developed countries, mainly the United States, Europe and the USSR. There are some reports of the disease in New Zealand, Australia and South Africa, although from the literature it would appear that the incidence of the disease in these countries is very low. There are few reports of the situation in tropical areas or from the developing countries. Whether this is simply a fact of more sophisticated diagnostic capabilities in the developed countries or that the disease does not occur in the tropical areas, is at this stage undetermined. The details of 16 cases in South Africa have been published.

L. pneumophila characteristically gives negative reactions for most biochemical tests with the exceptions of catalase, oxidase and betalactamase<sup>20</sup>. A definitive identification is therefore made, using the direct FA test, gas liquid chromatography or DNA hybridization<sup>25</sup>. Plasmids have been found in Legionella and Legionella-like organisms, however, at this stage no function for these plasmids has been described<sup>15</sup>. A broad range of extracellular enzyme activity has been described<sup>24</sup>. Both

endo- and exotoxins have also been found. The exotoxins have a variety of cytotoxic effects and haemolytic exotoxins have also been described<sup>1 11 14 29</sup>.

In development of an animal model for the study of L. pneumophila infections, Fitz George et al<sup>9</sup> found that mice were resistant to induction of disease by aerosols, by strains of serogroups I and III of L. pneumophila. Both of these strains were, however, pathogenic for guinea-pigs by aerosol infection over a wide range of doses. Intra-nasal instillation of 5 x 109 viable organisms did not induce disease, but when a small particle aerosol (less than 5 microns in diameter) was used, 104 viable organisms produced fatal widespread bronchopneumonia within 3 days. Milder illness with a less extensive bronchopneumonia, was produced in Rhesus monkeys and marmosets by one of these strains i.e. 74/81 serogroup I. Intraperitoneal injection of a laboratory adapted strain of L. pneumophila caused fatal disease in 8 out of 8 guinea-pigs within 24 hours.

The most significant finding in this series was that while 10<sup>4</sup> L. pneumophila organisms caused a fatal infection in guinea-pigs when given as a fine particle aerosol, 5 x 10<sup>9</sup> organisms of the same preparation, given intranasally, did not.

The most obvious difference between these methods is the size of the infected particle and the eventual deposition in the respiratory tract. The aerosols generated with a Collison spray in a Henderson type apparatus<sup>13</sup>, contain a majority of particles of 5 microns in diameter, which would penetrate to and be deposited on the surface of the alveoli and terminal bronchioles. Intranasal instillation on the other hand, results in the massive flooding of the upper respiratory tract only and produces relatively few particles small enough to be deposited in the terminal respiratory bronchioles or alveoli. The pneumonic lesions produced by aerosol infection in guinea-pigs, Rhesus monkeys and marmosets were very similar to those seen in Legionaires' disease in humans<sup>9</sup>.

A number of techniques have been developed for the diagnoses of Legionaires' disease. The fluorescing antibody technique has proven to be one of the most useful. The indirect fluorescent antibody (IFA) test as developed for detection of antibodies in human sera, is presently the most useful test for serological surveys<sup>26 27 28</sup>.

### MATERIALS AND METHODS

In this series IFA Legionella screening kits with 8 wells per slide with L. pneumophila groups I to IV as the antigen were used. The conjugate used in these tests was a fluoroscein isothiocyanate conjugated fraction goat anti-horse IgG (heavy and light chains specific) (Lot 19635)). In a block titration the optimal results were obtained with a dilution of 1:250 for the conjugate. This dilution was then used throughout the tests. A control positive serum obtained from Centre for Disease Control in Atlanta, Georgia was used as the control serum. This titrated out to a titre of 1:512. All sera on the screening tests were tested at a dilution of 1:2. Any serum showing positive reaction on these screens was then titrated out to its end point. Human sera were screened at a dilution of 1:64.

Sera, collected from apparently normal horses (n = 329) without signs of respiratory disease, were tested in this way. Sera from 22 human contacts with a

sero-positive horse which reacted at a titre of 1:256 were also tested.

#### RESULTS

Of the sera tested, 137 (41,6%) showed a reaction on the primary screening test. Twenty-three of these samples were positive at a titre of 1:16 and higher, thus giving a percentage reaction of 7%. Only one of these animals was positive at a titre of 1:256 which would be considered an indication of probable recent contact with the organisms (Table 1).

Table 1: The number of horses showing a positive reaction to

L.pneumophila

	End-point IFA titre					
	Negative -	1/2	1/16	1/256		
Total sera Percentage	192 58,4	114 34,7	22 6,7	1 0,3		

Clinically this horse had at no stage shown any clinical signs of a respiratory infection. None of the 22 human contacts showed any signs of current respiratory infections either. The results of serological analyses of human sera are reflected in Table 2.

Table 2: Serological reactions of people in close contact with a seropositive horse

	End-point IFA titre				
	Negative	1/64	1/256		
Total sera	18	3	1		
Percentage	81,8	13,6	4,6		

#### DISCUSSION

Collins et al.<sup>5</sup> examined a series of 2 586 animal sera using the micro-agglutination technique as described by Farshey et al.8. The initial serum dilution used in the Farshey study was 1:2 and the tests were carried out against four serogroups of L. pneumophila. The microagglutination titres greater than 1:64 were considered positive which is consistent with the criteria set for the micro-agglutination technique. The high percentage of sero-positive horses suggested that horses are commonly infected with L. pneumophila or related organisms and not unexpectedly, the older animals showed a far higher incidence of positive reactions. The indirect fluorescent antibody test was used to examine the sera of 109 horses3. The results were compared with the titres obtained by the micro-agglutination technique. A high correlation (r = 0.89) was found between the titres measured by the two tests. A further serological survey was carried out using five L. pneumophila serogroups and 4 other Legionella species: 29% of the horses and 24% of the sheep tested were positive to the Legionella species other than L. pneumophila at a titre of 1:256. At a titre of 1:128, 72% of the pigs, 56% of the sheep and 50% of the horses reacted to at least one of the Legionella species antigens. These tests were carried out using the IFA technique.

Although no prior history of people in close contact with a sero-positive horse was available, the fact that 82% of them remained negative to serological tests, would appear to indicate that the chances of transmission from a non-clinical state in horses are small. In the work of Collins et al.7 and Cho et al.4 they were unable to re-isolate the organism from horses after they had been infected both intravenously and by aerosol and, although these horses showed a sero-conversion they did not excrete the organisms again at any time. It is therefore unlikely that any of the people working in close contact with horses would have become infected from the horse, but would more likely have come into contact with the organisms from the same sources as the horse and it may be that in a large number of cases, the humans in contact with these organisms in this manner, may also show a similar reaction. The horses in these trials carried out at Colorado State University, USA, showed only a transient febrile response with a transient decrease in circulating lymphocytes 2 days after inoculation. Although these horses did not develop any clinical disease, all showed a sero-conversion which was evident as early as 4 days after inoculation. The serological response was confirmed by indirect immunofluorescence and was shown to consist predominantly of IgM. The agglutinating antibodies persisted for at least 4 months after infection. Histologically the lungs contained evidence of a low-grade inflammatory response, characterised by focal proliferation of the alveoli lining cells with few neutrophils and eosinophils, while the lymph nodes showed evidence of a reactive hyperplasia and these workers were unable to re-isolate the organisms from any of the horses which they had infected. It would, therefore, appear that the pathogenicity of L. pneumophila at least for serogroups I - III is very low in horses and that horses are most unlikely to act as carriers of the disease or as maintenance hosts in nature. As the research into the genus Legionella continues, it is nevertheless possible that organisms may be found which are more pathogenic to animals. However, at this stage, even serological surveys carried out in England in investigations into outbreaks of severe respiratory disease, failed to incriminate L. pneumophila21 in cattle and pigs.

The possibility of cross-reactions between the L. pneumophila and other organisms commonly infecting the horse was investigated. It was found that of 51 possible antigens, routinely demonstrated in L. pneumophila, up to 3 of these antigens cross-reacted to those from other equine pathogens. The number and type of the cross-reactions however were not different from those in 18 other bacteria tested. These reactions were quantified by cross-line immuno-electrophoresis and the organisms tested against were Streptococcus equi, Rhodococcus equi, Salmonella abortus equi, Actinobacillus equuli and Pseudomonas pseudomallei. From these tests it can be assumed that naturally occurring antibody titres to L. pneumophila in horses, are not due to cross-reactions with other bacteria.

As a result of all this work, it can be assumed at this stage that, although specific antibody titres to L. pneumophila can be demonstrated in horses, the pathogenicity of the organisms is extremely low and of little clinical significance. Further the organisms are not excreted by horses after exposure and that horses are unlikely to act as carriers of the disease.

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CASE REPORT **GEVALVERSLAG** 

## AN OUTBREAK OF LYMPHOSARCOMA IN MERINO SHEEP IN THE SOUTH WESTERN CAPE

JENNIFER R. GREEN\*, I.A. HERBST\*\* and D.J. SCHNEIDER\*

ABSTRACT: Green, Jennifer R.; Herbst I.A.; Schneider D.J. An outbreak of lymphosarcoma in Merino sheep in the South Western Cape. Journal of the South African Veterinary Association (1988) 59 No. 1, 27-29 (En). Regional Veterinary Laboratory, Private Bag X5020, 7600 Stellenbosch, Republic of South Africa.

Ten per cent of a flock of 481 Merino sheep died from suspected lymphosarcoma in the Caledon district, during a period of one vear. In 9 sheep the diagnosis was confirmed by histopathological examination of the tumours. All the sheep and the cattle on the farm were tested for enzootic bovine leukosis using a Bovine Leukemia Glycoprotein Immunodiffusion kit. At the first test 20,5% of the sheep had antibodies to enzootic bovine leukosis. A control programme resulted in a drop in infection rate (3%) within 17 months.

Key words: sheep, lymphosarcoma

#### INTRODUCTION

Sheep are highly susceptible to bovine leukemia virus (BLV) infection3, but only isolated cases of lymphosarcoma have been diagnosed in sheep and cattle in the South Western Cape in the past. Virus recovered from tumours in sheep is biologically, morphologically and antigenically indistinguishable from BLV<sup>3</sup>.

BLV may be transmitted via colostrum, milk and through contact with infected animals3. It is also transmitted by biting insects1 and by blood on vaccination needles and on surgical equipment<sup>2</sup>.

The agar gel immunodiffusion test is widely used to detect antibodies to BLV and to identify infected animals. As in cattle, BLV in sheep persists despite the continuous presence of antibodies<sup>3</sup>.

## HISTORY OF OUTBREAK

During August 1984, lymphosarcoma was diagnosed in 8 sheep on a farm in the Caledon district following histopathological examination of organ samples. According to the farmer 50 sheep had died during a period of about one year. All of them had shown the presence of large growths in various organs or in the subcutaneous tissue.

The farmer owns 2 farms, but at the time of the outbreak of lymphosarcoma in the sheep, ran a flock of 481 Merino sheep and 62 cattle on only one (the larger) of them. As far as could be ascertained the disease had never occurred in overt form in any of the cattle.

#### CLINICAL AND POST MORTEM FINDINGS

One live affected sheep was brought to the Stellenbosch Veterinary Laboratory for diagnostic purposes. On clinical examination several large, firm subcutaneous wellings were found over the left shoulder and thorax Fig. 1). The sheep was euthanased by intravenous injecion of an overdose of pentobarbitone sodium.

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Macroscopic Pathology: At necropsy large grey, firm tumours were found in the liver, kidney, lung, heart, intestinal wall and subcutaneous tissue of the shoulder and thorax. The peripheral and mesenteric lymph nodes and the spleen were enlarged.

Microscopic Pathology: With the light microscope the neoplasms appeared as sheets of uniform round cells with round to oval nuclei and the occasional reniform nucleus (Fig. 2 & 3). The mitotic index was high and there was focal necrosis in the larger tumours.

#### SEROLOGICAL FINDINGS

Serum of this sheep (see above) was tested for BLV antibodies using a Bovine Leukemia Glycoprotein Immunodiffusion kit in which Leukassay B reagents are used. Antibodies to BLV were found.

#### CONTROL

All the sheep and cattle on the farm were identified using ear tags and the farmer was advised to institute and maintain strict hygiene measures during all vaccination, inoculation, bleeding, tail docking and shearing procedures. It was advocated that a sterile hypodermic needle be used for each animal during bleeding, inoculation and vaccination procedures and that shearing and surgical equipment be sterilised by dipping in chlorhexidine and sodium hypochlorite after use on each animal.

BLV infected animals were identified by periodical serological testing of sheep and cattle on the farm by use of the immunodiffusion test (vide supra). A group of 160 sheep, purchased by the farmer, were all bled. They all tested negative before being brought to the farm, a week before the second flock test was performed. According to the results of the serological tests, the animals were divided into 2 groups: the first group that consisted of positive sheep with their lambs and positive cattle was taken to the second farm where their numbers were gradually reduced by sale for slaughter for human consumption. The second group that consisted of animals that were serologically negative for BLV, remained on the larger farm and was tested a second and a third time, 5 months and one year and 5 months after the first test respectively.



Fig. 1: Subcutaneous tumours on the shoulde and on the thorax

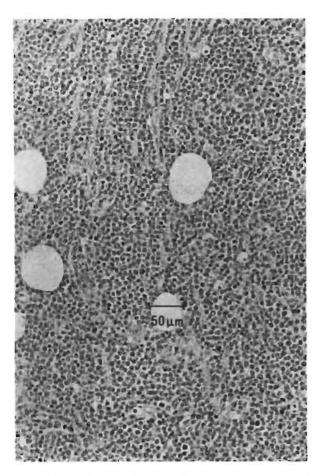


Fig. 2: Sheets of neoplastic cells with adipose cells between them. HE  $\!x\,200$ 

The numbers, categories and types of animals tested as well as the results of the test are presented in Table 1.

The second text revealed that one of the remaining 366 original sheep was positive for BLV. This was an adult ewe. During the interval between the first and second tests, 3 ewes had died and 12 old ewes had been culled. All 60 cattle were negative and at this time they were all removed to another camp, which placed them out of any contact with the sheep.

The third test included 844 sheep. This comprises the 160 purchased sheep as well as the remaining

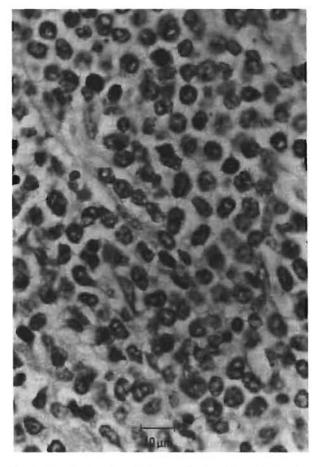


Fig. 3: Neoplastic cells at high magnification. The majority of nuclei are round. Some are oval to reniform in shape. HE x 1000

Table 1: Results of the first serological test for BLV in the Merino sheep performed on 13.9.85

Age of sheep	Number tested	Number positive	Percentage positive
Lambs	190	0	0
2 tooth - 6 tooth	74	12	16
6 tooth and older	217	88	40,55
TOTAL	481	100	20,7



Fig. 1: Subcutaneous tumours on the shoulder and on the thorax

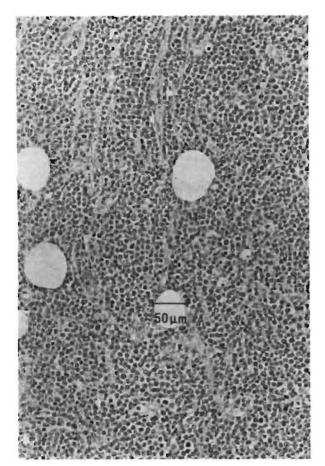


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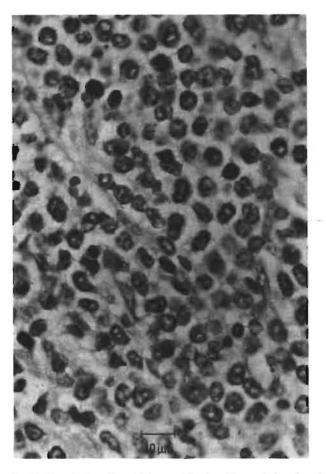


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TOTAL	481	100	20,7

serologically negative animals from the original flock and the progeny of both these groups. A group of 37 cattle that had recently been purchased and that had grazed in an adjacent camp for 2 weeks were also tested.

One cow and 29 sheep had antibodies to BLV. Two of these sheep were 2 year-old ewes from the 160 sheep that had been purchased. The others were adult ewes.

#### DISCUSSION

The results of the second test were encouraging. The results of the third test were, however, disappointing. On investigation it was found that the farmer had relaxed his flock and herd hygiene. The newly purchased sheep and cattle had been in contact with the original flock before the latter were tested. All the sheep had been vaccinated against pulpy kidney and blue-tongue disease and both the sheep and cattle had been subcutaneously injected with a worm remedy. Instead of using a sterile hypodermic needle for each sheep, 5 sheep had been vaccinated with each needle. It is speculated that there were a few sheep that carried the virus, but that had not yet developed antibodies4. The virus was most likely passed on to other sheep during vaccination. This emphasizes the fact that eradication of the BLV virus from a flock or herd will take perseverance, dedication, attendance to detail and a thorough understanding of all aspects of the disease on the part of the owner. To date (2 years after the original outbreak) no new cases of overt disease have occurred in the sheep or cattle. It is, however, realised that this time interval is much too short to make any unequivocal statement concerning this.

BLV is a highly infectious agent that can result in severe economic loss. Strict management together with regular testing is needed to control the disease. It is interesting to note that there were an unusual number of clinical cases in this outbreak.

#### **ACKNOWLEDGEMENTS**

The authors thank Mrs C van Reenen, Mr A Terblanche and Mrs V Krömker of the Regional Veterinary Laboratory, Stellenbosch, for their technical assistance and Mr P van der Merwe of the Fruit Technology Research Institute, Stellenbosch for the photographs of the sections. The farmer is also thanked for his co-operation.

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**BOOK REVIEW BOEKRESENSIE** 

#### FUNDAMENTAL TECHNIQUES IN VETERINARY SURGERY

C.D. KNECHT, A.R. ALLEN, D.J. WILLIAMS, J.H. JOHNSON

3rd Edn. Bailliere Tindall, W.B. Saunders Company 1987. 771 Figures, 7 Tables, 1 Chart, Price £23,50 (ISBN 0-7216-1397-7).

The publication of a third edition twelve years after the first edition confirms the acceptance of the book by students and practitioners.

The contents of the book are presented in eleven chapters. Under Surgical Instrumentation, the authors describe the instruments that form part of a basic set, draping of both small and large animal patients, as well as methods routinely used.

The section on Orthopaedic instruments is, however, of doubtful significance because the instrumentation discussed is not indicative of what a basic orthopaedic set should include. The chapter is concluded with a useful table showing the contents of stan-

The second chapter is devoted to suture materials and knotting of sutures. The description of materials could have been more detailed while materials like kangaroo tendon should only be mentioned within historical context. The different knotting techniques are excellently illustrated. Suture patterns are well defined, explained and schematised.

Routine procedures — from preparation of packs to cleaning of the theatre — are discussed in detail. Sterilization, preparation of the operative field, scrubbing as well as the donning of gowns and gloves are simply and clearly described.

The excellent illustrations in the chapters on dressings, casts and splints of the different body regions of small animals and horses allows for easy understanding and should be of practical significance to private practitioners.

Chapter 10 is mainly devoted to an oversimplified description of selected surgical procedures for small animals. The book is concluded with a discussion on the basics of cardio-pulmonary resuscitation.

This profusely illustrated and well presented "little red book", as nicknamed by the authors, can be recommended as a good textbook on general surgery as well as a helpful guide for the new practitioner. It offers pleasant reading at the not so pleasant price of £23,50.

E.J. Durante

CASE REPORT GEVALVERSLAG

#### SERVIKALE INTERVERTEBRALE DISKUS PROLAPS IN 'N PERD

P. STADLER\*, S.S. VAN DEN BERG\*\* en R.C. TUSTIN\*\*\*

ABSTRACT: Stadler P.; Van den Berg S.S.; Tustin R.C. Cervical intervertebral disc prolapse in a horse. Journal of the South African Veterinary Association (1987) 59 No. 1, 31-32 (Afrik). Department of Medicine, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, 0110 Onderstepoort, Republic of South Africa.

A Hansen type I cervical intervertebral disc prolapse was diagnosed in a 16-year-old American Saddler showing clinical signs of paresis and ataxia. An ante-mortem diagnosis was made by means of plain radiographs and a myelogram. The horse was euthanased and the diagnosis confirmed on a post-mortem examination.

Key words: intervertebral disc prolapse, cervical, horse

#### INLEIDING

Intervertebrale diskus uitbulting of prolaps is 'n relatief seldsame toestand in die perd<sup>8</sup>. Tydens 'n literatuurstudie kon slegs drie gevalle van diskus uitbulting<sup>1</sup> <sup>6</sup> en een geval van ruptuur van die anulus fibrosus met bloeding in die neurale kanaal<sup>5</sup> opgespoor word. Van die drie gevalle van diskus uitbulting was twee voordoods gediagnoseer<sup>1</sup> <sup>6</sup>. Sover bekend is hierdie die eerste bevestigde geval van servikale intervertebrale diskus prolaps in 'n perd in Suid-Afrika.

#### **GESKIEDENIS**

'n Gewese Suid-Afrikaanse enkeltuigkampioen, die sestienjarige hings Cameo's Brigadoon, is deur 'n privaatveearts aangebied vir ondersoek en behandeling. Agt dae tevore, 'n week na die hings se laaste deelname aan 'n skou, is hy lêend in sy stal gevind en was hy nie in staat om op te staan nie. Gedurende die daaropvolgende paar dae het sy toestand sodanig verbeter dat hy weer kon loop, maar hy het ataksie getoon.

#### KLINIESE ONDERSOEK

Met opname was sowel die perd se kondisie as habitus goed. 'n Volledige kliniese ondersoek is uitgevoer en afgesien van die neurologiese afwykings was die enigste ander abnormaliteit 'n beskadiging van die vel oor die regter skouer en beide tuber coxae.

'n Neurologiese ondersoek het getoon dat die kraniale senuwees se funksie normaal was. Die perd het tetraparese getoon, maar met kliniese tekens meer opvallend aan die linkerkant. Hy het die linker voor- en agterhoewe gesleep met beweging en het selfs teen 'n muur aangeleun om sodoende die minimum gewig aan die kant te dra. Hy was onwillig om te beweeg, maar indien hy forseer is om te beweeg, was sy gang ataksies. Tydens beweging kon gesien word dat al vier ledemate wel aangetas was. Verder was hy ook geneig om te struikel.

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Wanneer hy forseer was om in 'n baie klein sirkel te draai, het hy sy hoewe so lank as moontlik op die grond probeer hou. Met beweging het die buitenste been oormatig uitgeswaai en het die perd probleme ondervind om sy hoewe weer in die korrekte gewigdraende posisie neer te plaas. Hy het met moeite agteruit beweeg.

Geen pyn kon verwek word met betasting en manipulasie van die kop en nek nie.

Ondersoek van die bloed, uriene en mis het geen abnormaliteite opgelewer nie.

Vier dae na opname is die perd onder algemene verdowing geplaas. Induksie is toegepas met die gebruik van gliserol guaiakolaat eter (G.G.E., Centaur) en tiopentoon (Intraval, Maybaker) waarna die verdowing met halotaan (Fluothane, ICI) instandgehou is.

Met die perd in 'n laterale ligging is oorsigfoto's eers geneem wat 'n vernouing van die  $C_{2\cdot 3}$  intervertebrale spasie aangetoon het. Serebrospinale vog is deur die atlanto-occipitale opening versamel voordat 40,5g metrizamied (Amipaque, Winthrop) opgelos in 60 ml verdunningsvloeistof subarachnoidaal ingespuit is en 'n miëlogram gedoen is. Analise van die serebrospinale vog het geen abnormaliteite opgelewer nie.

Die miëlogram het 'n ekstradurale drukking op die rugmurg bo die vernoude  $C_{2-3}$  intervertebrale spasie aangetoon. Die ventrale kontrasmediumkolom was daar vernou en die rugmurg dorsaal verplaas (Fig. 1).

Die bogenoemde radiologiese bevindinge het 'n diagnose van intervertebrale diskus prolaps bevestig. Die perd het herstel van die verdowing en weer rondbeweeg. Drie dae later het hy egter gaan lê en kon nie weer opstaan nie. Weens die swak prognose is genadedood toegepas.

## NADOODSE ONDERSOEK

Die enigste abnormaliteite van belang wat gevind is, was geassosieer met die intervertebrale diskusse tussen servikale werwels 2-3 en 3-4 en hul onderskeie artikulasieprosesse. Die nukleus pulposus van die betrokke diskusse was geelgroen verkleur en het 'n bros, kaasagtige konsistensie gehad. Van hierdie materiaal het die anulus fibrosus in 'n dorsale rigting penetreer en 'n klein hoeveelheid was op die ventrale vloer van die vertebrale kanaal sigbaar. Die kraakbeen van die kraniale en kaudale dele van die werwelliggame by die betrokke

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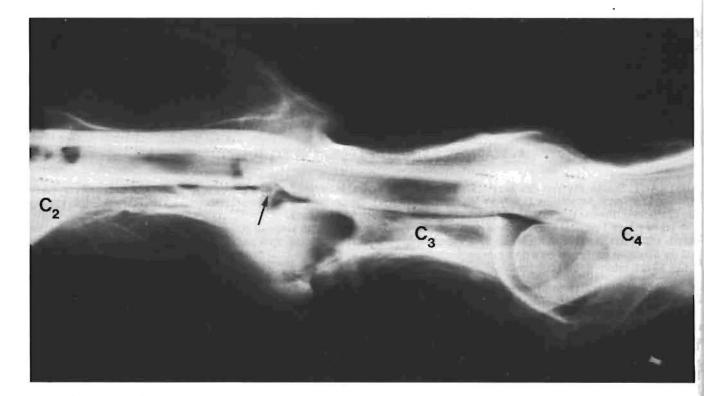


Fig. 1: Miëlogram van die servikale vertebrae. 'n Massa (pyl) dorsaal van die vernoude C 2-3 intervertebrale spasie vernou die ventrale kontraskolom en verplaas die rugmurg dorsaal

intervertebrale spasies asook van die betrokke artikulasieprosesse het erosies getoon. Verder was daar ook nuutgevormde been en kraakbeen teenwoordig op die rande van die artikulasieprosesse.

#### **BESPREKING**

In honde is dit meer dikwels die intervertebrale diskusse van die torakolumbale werwels wat degenerasie en uitbulting ondergaan<sup>2</sup>. In die perd daarenteen is daar nog geen gevalle aangeteken waar uitbulting van die torakolumbale diskusse voorgekom het nie. Die redes hiervoor is moontlik dat hierdie diskusse in die perd dun is en uit fibrokraakbeen bestaan; die sterkte van die longitudinale vertebrale ligamente en die relatiewe onbuigbaarheid van die perd se torakolumbale werwelkolom in vergelyking met die van die mens en hond<sup>4</sup>.

Diskus degenerasie in die perd is al gevind in asimptomatiese ouer perde<sup>7</sup> sowel as in perde met kliniese tekens van ataksie<sup>10</sup>, maar in laasgenoemde geval is daar nie gesê of uitbulting ook teenwoordig was nie. Diskus degenerasie is as oorsaak van trombo-embolisme van die spinale vate met gevolglike iskemiese nekrose van die rugmurg beskryf9. Uitbulting sonder tekens van degenerasie van die betrokke diskus, soos wat dikwels in chondrodistrofiese honde gevind word3, is ook al in die perd beskryf1 en daar is gespekuleer dat hulle moontlik traumaties van oorsprong kon wees. In die geval onder bespreking was daar egter degenerasie sowel as prolaps van intervertebrale diskusse wat aanleiding tot die kliniese tekens gegee het. Daar was in hierdie geval geen aanduiding van 'n traumatiese insident kort voor die aanvang van kliniese tekens nie.

Hoewel intervertebrale diskus degenerasie en uitbulting of prolaps in die perd dus 'n relatief seldsame toestand is, waarskynlik as gevolg van die meer fibreuse aard van die nukleus pulposus<sup>6</sup>, behoort dit sterk oorweeg te word as 'n moontlike oorsaak van ataksie in perde. Met hierdie kennis tot ons beskikking is dit moontlik dat meer gevalle van die aard gediagnoseer en aangemeld sal word.

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CASE REPORT GEVALVERSLAG

### CHLAMYDIOSIS IN A SPRINGBOK (ANTIDORCAS MARSUPIALIS)

J.J. VAN DER LUGT\* and J.C. KRIEK\*\*

ABSTRACT: Van der Lugt J.J.; Kriek J.C. Chlamydiosis in a springbok (Antidorcas marsupialis). Journal of the South African Veterinary Association (1988) 59 No. 1, 33-37 (En). Section of Pathology, Veterinary Research Institute, P.O. Box 12502, 0110 Onderstepoort, Republic of South Africa.

Chlamydiosis was diagnosed in a 3-month-old springbok (Antidorcas marsupialis) on a farm where 46 out of 65 springbok died over a period of 3 months. Nervous signs, which included circling, opisthotonus, loss of balance, recumbency and intermittent paddling movements of the legs were seen in lambs and adult animals. Gross lesions consisted of a fibrinous peri- and epicarditis and interstitial nephritis, while the microscopical lesions were characterised by multifocal encephalomyelitis and meningitis, interstitial pneumonia, and vasculitis in the brain, spinal cord and myocardium. Colonies of suspected chlamydial organisms were seen in a few mononuclear cells and tubular epithelium in the kidneys. Ultrastructurally the colonies were composed of 3 morphological types of particles, consistent with the different stages in the life cycle of the chlamydiae.

Key words: Chlamydia psittaci, systemic infection, springbok

#### INTRODUCTION

The chlamydiae are obligatory intracellular parasites<sup>16</sup>. In view of their unique growth cycle, these organisms have been placed in their own order, Chlamydialis, with one genus, *Chlamydia*<sup>9</sup> <sup>13</sup>. Currently two biochemically, biologically and antigenically distinguishable species are recognised, namely, *Chlamydia trachomatis* and *C. psittaci*<sup>8</sup>. Except for a few isolates from mice <sup>8</sup>, all *C. trachomatis* isolates have come from humans. Chlamydiosis caused by *C. psittaci*, however, is not as species-specific and it has been isolated from many avian and mammalian species<sup>16</sup>, man (human psittacosis)<sup>13</sup> and amphibians<sup>6</sup>.

Serological evidence suggests that chlamydial infections exist in several species of wildlife<sup>7</sup> <sup>14</sup>, but naturally occurring chlamydiosis has only been described in opossums (Didelphis paraguayensis)<sup>12</sup>, northern fur seals (Callorhinus ursinus)<sup>3</sup>, muskrats (Ondatra zebethicus)<sup>15</sup>, snowshoe hares (Lepus americanus)<sup>15</sup> and koala (Phascolarctos cinereus)<sup>2</sup>.

In this paper clinical and pathological findings in a 3-month-old springbok (Antidorcas marsupialis), which was destroyed while suffering from chlamydiosis, are reported.

#### HISTORY AND CLINICAL SIGNS

In August 1985 a farmer near Kimberley bought a group of 40 adult springbok, composed of 30 ewes and 10 rams, and placed them in a natural veld camp together with 70 adult sheep ewes and 3 rams. The sheep were born and raised on the farm and had not been vaccinated during the previous 3 years. Wildebeest, blesbok and rhebok were kept in separate camps on the farm, but no direct contact was possible with the springbok and sheep.

Seventeen of the 25 springbok lambs born from the end of November to the middle of December died between 2-3 weeks and 3½ months of age. No stillbirths,

abortions or complications during birth were noticed by the farmer. During the period December to March, 29 adult springbok (20 ewes and 9 rams) also died. No mortalities occurred in the adult sheep during this period.

According to the farmer a range of clinical signs were noticed in approximately 30 young and adult springbok, which included unsteadiness, circling, loss of balance and opisthotonus. The ataxic animals fell frequently and made galloping leg movements while recumbent. Weakness of the hindquarters, which invariably progressed to total paralysis, was noticed in some animals. Most springbok died within 3-4 days of the onset of the clinical signs, although in a few adult springbok, the course of the disease was protracted and extended up to 3 weeks. In addition to the nervous signs, these animals showed severe loss of weight, while a few salivated.

Soon after the first mortalities occurred in the springbok, the sheep were moved to an adjacent camp. During February 1986, thirty of the 70 sheep ewes aborted at  $c.\,3\%-4\%$  months of gestation. No complications were reported in the ewes and no attempt was made to identify the cause of the abortions.

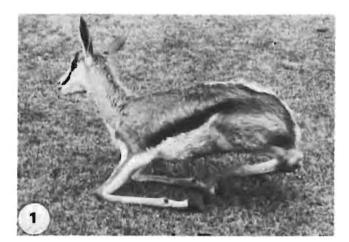
A 3-month-old springbok ram lamb which had been sick for 4-5 days was referred for necropsy to the Section of Pathology, Veterinary Research Institute, Onderstepoort. The animal was still in a good condition and made attempts to graze, but at times it became restless, walked with an unsteady gait, and had a bewildered facial expression, often standing with its feet wide apart. At times the animal stumbled, fell and frequently was unable to rise (Fig. 1), showing intermittent paddling movements of the legs (Fig. 2).

#### MATERIALS AND METHODS

A necropsy was performed after the springbok was killed by an intravenous overdose of barbiturates. Specimens of heart, lung, spleen, liver, kidneys, intestines, skeletal muscle, mesenteric and peripheral lymph nodes, brain, spinal cord, optic nerves and the eyes fixed in 10% buffered formalin, were routinely processed and stained with haematoxylin and eosin (HE) for histopathological examination. Selected sec-

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Figs. 1 & 2: The springbok frequently fell (Fig. 1) and showed paddling leg movements (Fig. 2)

tions of the kidneys were stained with Giemsa, the periodic acid-Schiff (PAS) reaction and Gram's stain.

Small blocks of formalin-fixed kidney were post-fixed in 2,5% gluteraldehyde, routinely processed and stained with uranyl acetate and lead citrate for transmission electron microscopy.

Brain smears were prepared from the hippocampus, stained with Giemsa and examined for the presence of *Cowdria ruminantium* colonies.

Specimens of epicardium and brain, and swabs of epicardial surfaces were submitted to the Section of Bacteriology, VRI, for routine aerobic bacterial isolations. Specimens of liver, brain and fat were tested for pesticides containing chlorinated hydrocarbon and organophosphorus compounds.

#### RESULTS

#### Macroscopical pathology

Lesions were limited to the kidneys and the heart. The kidneys were pale and slightly enlarged, and bulged on cut surface. Numerous pinhead-sized yellowish-white foci were disseminated throughout the cortex and medulla of both kidneys. Some of the foci were surrounded by a narrow dark-red zone.

Fibrinous adhesions between the epi- and pericardium were most prominent at the base of the heart, giving the epicardium a greyish-white and irregularly thickened appearance in some areas.

#### Microscopical and ultrastructural pathology

Kidneys: Numerous inflammatory foci were randomly distributed in the interstitium throughout the cortex and medulla of both kidneys (Fig. 3). The foci were of variable size and consisted predominantly of large mononuclear cells, lymphocytes and macrophages, some plasma cells and a few neutrophils which frequently obliterated the tubules (Fig. 4). Neutrophils were the main cell type in a few foci. Mitotic figures often occurred in mononuclear cells and some inflammatory cells were necrotic. Tubules entrapped by the cellular infiltrate were occasionally dilated and contained cellular casts, or, in some instances, mineralized deposits. Several of the inflammatory foci, especially at the cortico-medullary junction and in the outer medulla, were bordered by interstitial haemorrhages (Fig. 3). A mild perivascular infiltration of lymphocytes and plasma cells was evident at the cortico-medullary junction.

The epithelium of the renal pelvis was hyperplastic and accompanied by a mild but widespread subepithelial infiltration of mononuclear cells which sometimes formed discrete nodules.

In a few of the cellular foci basophilic, granular, intracytoplasmic inclusions were discernible in large mononuclear cells and tubular epithelium (Fig. 4). The inclusions consisted of numerous small, round to oval or pleomorph bodies, which stained dark-blue to purple-blue with Giemsa, and negative with Gram's stain and the PAS reaction. These bodies were densely packed in mononuclear cells, and dispersed in epithelial cells (Fig. 4).

Ultrastructurally the inclusions contained three distinct types of particles (Fig. 9): small spherical particles, 250-375 nm in diameter, with a single eccentric aggregation of electron-dense material, or nucleoid, partially surrounded by dense cytoplasm; round to oval intermediate particles, 350-550 nm in size, with a nucleoid situated in the centre of granular cytoplasm; and oval to slightly irregular large particles, 600-1200 nm in diameter with granulo-fibrillar cytoplasm. Some of the small particles were surrounded by a distinct cell wall and a few large particles were undergoing binary fission. As a rule the inclusions were partly enveloped by a membrane and densely packed with particles of all three types. In some tubular epithelial cells, the particles were distributed throughout the cytoplasm and the majority were of the small type.

Central nervous system, eyes and optic nerves: A few foci of gliosis, accumulations of mononuclear cells and vacuolization of nervous tissue were evident in the grey matter of the midbrain and the white matter of the cerebellum (Fig. 5). Some neurons were necrotic and mineralized, and many axons and glial cells were prominently swollen. There was also evidence of demyelination. Pigment, varying from granular darkbrown to homogenous yellow-brown, was seen in some macrophages.

Vasculitis was noticed in most inflammatory foci and was also randomly distributed throughout the cerebrum, cerebellum and meninges (Fig. 6). The vasculitis was characterized by swelling and necrosis of the endothelial cells; fibrinoid changes, necrosis and vacuolation of the tunica media; and infiltration of the wall and perivascular spaces by variable numbers of lymphocytes and macrophages, some plasma cells and

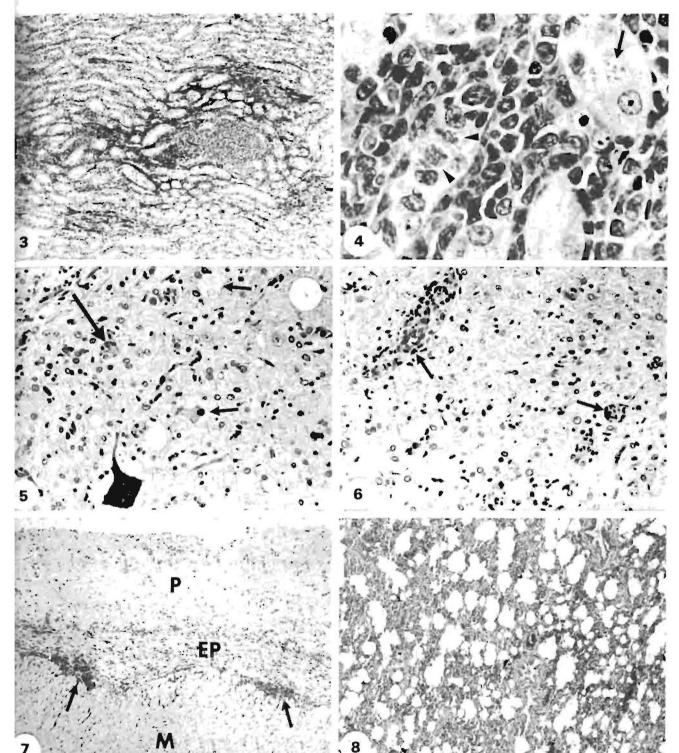


Fig. 3: Focal interstitial nephritis in the medulla surrounded by haemorrhages. HE X 100

- Fig. 4: Interstitial mononuclear cell infiltrate in renal cortex. Compact chlamydial inclusions in mononuclear cells (arrowheads) and organisms dispersed in a tubular epithelial cell (arrow). HE X 1000
- Fig. 5: Vacuolation of neuropil, gliosis, foci of mononuclear cells (large arrow) and necrosis of neurons (small arrows) in the grey matter of the midbrain. HE X 400
- Fig. 6: Vasculitis with perivascular infiltration constituted mostly by lymphocytes (arrows) in grey matter of midbrain. HE X 500
- Fig. 7: Fibrinous pericarditis (P) and thickening of epicardium (EP) by fibrosis. Mild but widespread infiltration of inflammatory cells in the epicardium and adjacent myocardium (M), forming discrete foci (arrows). HE X 200
- Fig. 8: Diffuse acute interstitial pneumonia characterized by an exudation of fibrin and inflammatory cells. HE X 100

neutrophils. In certain areas the leptomeninges were oedematous and infiltrated by mononuclear cells.

A few foci of mild gliosis, vacuolation and mononuclear cell infiltrates occurred in the grey matter of the spinal cord. Vasculitis and perivascular cuffing by lymphocytes and occasionally multiple small haemorrhages were also present.

Focal perivascular and adventitial accumulations of a small number of lymphocytes and plasma cells occurred in the optic nerves.

No colonies of *C. ruminantium* were present in the brain smears.

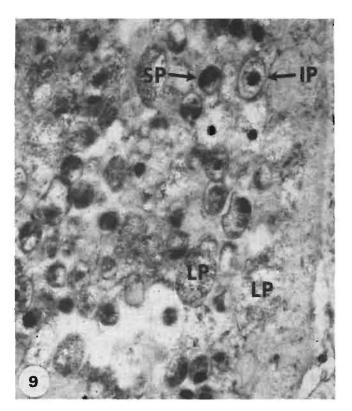


Fig. 9: Electron micrograph of part of an intracytoplasmic inclusion in a mononuclear cell in the renal cortex. Three types of particles are discernable: small dense particles (SP), inter mediate particles (IP) and large granular particles (LP) X 25 000

Heart: There was a mild to moderate fibrinous peri- and epicarditis (Fig. 7). The epicardium of the ventricles was focally thickened by fibrosis, oedema and a mild but widespread exudate which consisted of macrophages, lymphocytes, plasma cells and some neutrophils. The inflammatory cells infiltrated perivascular spaces and the adjacent myocardium, and occasionally formed discrete foci (Fig. 7). Vasculitis and perivascular lymphocytic cuffing similar to those described in the brain, were occasionally seen in the myocardium.

Lung: The alveolar walls were moderately distended by fibrih, macrophages, neutrophils and lymphocytes (Fig. 8). In addition, there was also a mild focal fibrinous pleuritis.

Other organs: Lesions included lymphoid and reticuloendothelial hyperplasia, sometimes accompanied by necrosis of lymphocytes, in the spleen, lymph nodes and Peyer's patches, and a mild focal fibrinous peritonitis.

### Bacteriology and toxicology

No pathogenic bacteria were cultured from the organs and swabs submitted for isolation, and no chlorinated hydrocarbon and organophosphorus compounds could be demonstrated in the tissues.

### DISCUSSION

The staining and morphologic characteristics of the intracytoplasmic inclusions in the kidney conform to those described for chlamydiae<sup>8</sup> <sup>16</sup>. Ultrastructurally the small particles were comparable with the infectious elementary bodies, and the large particles represented

the vegetative initial or reticulate bodies in the life cycle of chlamydiae<sup>1</sup>.

In domestic animals *Chlamydia psittaci* often elicits lesions in one or more organs<sup>10</sup>. Inflammatory changes in various organs in the springbok indicated that the animal suffered from a systemic infection. The lesions in the springbok were very similar to those reported in sporadic bovine encephalomyelitis (SBE) and polyarthritis in lambs, which are both systemic chlamydial infections<sup>5 16</sup>. In addition to the involvement of the central nervous system, a fibrinous peritonitis, pleuritis and/or pericarditis as well as foci of granulomatous inflammation in the kidney were observed in SBE<sup>4 16</sup>. In lambs with chlamydial polyarthritis, concomitant lesions included diffuse interstitial pneumonia, encephalomyelitis and fibrinous pleuritis and peritonitis<sup>10 11</sup>.

Although chlamydial infection was confirmed in only one animal, the clinical signs and course of the disease in the other springbok were very similar and can possibly also be attributed to chlamydiosis. The source of the infection in this outbreak was not determined, but the sheep grazing together with the springbok in the same camp could have played a role. It is perhaps significant that c. 30 sheep ewes aborted two months after the first mortalities occurred in the springbok. Unfortunately the cause of these abortions was not determined, but may have been an indication of chlamydial infection in the sheep. Intestinal chlamydial infections are common in sheep and usually subclinical<sup>16</sup>. Apparently healthy sheep with an intestinal infection can excrete the agent intermittently over several years, thus providing a rich source of organisms for contamination of the environment17.

### **ACKNOWLEDGEMENTS**

The authors wish to thank the staff of the Photography and Electron Microscopy Sections for the preparation of the photomicrographs.

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### BOOK REVIEW BOEKRESENSIE

### DISEASES OF THE THORAX — RADIOGRAPHIC DIAGNOSIS

### PETER F. SUTER, JUAN A. GOMEZ

1st Edn. The Iowa State University Press, Ames, Iowa, 500100. 1981 pp 77, figures 9 and tables 32, Price £9,95 (ISBN 0-8138-0370-5)

This concise soft cover book based on the lecture notes of Dr Suter, is a welcome addition to the complex subject of thoracic radiographic interpretation of dogs and cats. The authors have managed to explain the various conditions in an easy to follow, well structured manner as well as by utilising extensive tables of the various differential diagnosis of radiographic findings. The book contains no radiographic reproductions or index but a comprehensive table of contents overcomes the latter problem.

Technical factors and basic principles of radiographic interpretation are covered in the first few pages followed by sections on the thoracic wall and diaphragm, pleural, mediastinal, tracheal and oesophageal disorders. Pulmonary and cardiac diseases are covered more intensively including diagnosis by means of selective and non-selective angiocardiography. A section is included on the pathophysiological dimensions in the interpretation of thoracic radiographs. A comprehensive reference list is included.

This book is strongly recommended for students and private practitioners as well as those who teach clinical cardiology and other thoracic conditions.

R.M. Kirberger

# BOVINE BRUCELLOSIS IN THE HIGHVELD REGION: INCIDENCE IN DAIRY HERDS

J.A. ERASMUS\* and J. FLOOR\*\*

ABSTRACT: Erasmus J.A.; Floor J. Bovine brucellosis in the highveld region: Incidence in dairy herds. Journal of the South African Veterinary Association (1987) 58 No. 1, 39-40 (En). Directorate of Veterinary Services, Veterinary Laboratory, P.O. Box 625, 9500 Kroonstad, Republic of South Africa.

Bulk tank milk samples which were collected twice with an interval of 2 months from 2103 herds were tested for brucellosis by employing the brucella ring test. Farmers involved were all supplying industrial milk to the National Co-operative Dairies. A questionnaire was circulated to these farmers in which they were asked to indicate how heifer calf vaccination with strain 19 vaccine was practised on their properties.

Of the herds tested, 18,1% could be regarded as infected. This figure varied from 8,9% in the Potchefstroom area to 30,9% in the northern districts of the Hoopstad area. An important cause of this rate of infection should be sought in improper calfhood vaccination. At least 9% of the respondents did not practice heifer calf vaccination whilst another 8,1% only commenced with vaccination during 1985 or later. About 12,4% of farmers commenced heifer calf-vaccination prior to 1970.

Key words: Bovine brucellosis, brucella ring test, heifer calf vaccination

### INTRODUCTION

The national incidence of brucellosis has decreased from 10,5% during 1975/6 to about 1,7% during 1985/6<sup>1</sup>. These figures were calculated from routine brucellosis tests performed at the various veterinary laboratories during a specific year. Blood samples examined originated from both dairy and beef herds. These samples were presumably collected several times per annum from infected herds and at least once from brucellosis free herds.

During 1985/6 the incidence of reactors in beef and dairy herds in the Highveld region amounted to about 2,5% (Director, Veterinary Services, Private Bag X138, 0001 Pretoria, personal communication) indicating the significance of bovine brucellosis in this region. In order to estimate the incidence of brucellosis in dairy herds in different parts of the Highveld region, a survey was conducted in which the brucella ring test (BRT) was performed on bulk milk samples, collected from 2103 industrial milk producers. A questionnaire, in which particular attention was given to heifer calf vaccination with strain 19 (S 19) vaccine, was simultaneously distributed to these farmers.

### **MATERIALS AND METHODS**

Two bulk tank samples were collected from each participant supplying industrial milk to the local depots of National Co-operative Dairies (NCD) in the region. These samples were collected after mechanical stirring of a bulk tank for at least 5 min and were transported to the nearest laboratory on ice. A BRT was performed on each sample and the results were interpreted according to the method of Herr et al.<sup>2</sup>.

In order to present the incidence of brucellosis in the relevant herds on an area basis, the highveld region was subdivided according to the operational areas of the 5

state veterinarians. As a non-significant number of producers from the Bethlehem area (Fig. 1) supply industrial milk to NCD, this area was omitted from both surveys.



Fig. 1: Highveld region subdivided into 5 areas.

### **RESULTS**

The relevant data are summarised in Tables 1 and 2.

Of the 2103 samples tested, 18,1% resulted in positive reactions on both occasions, while 11,7% resulted in one positive reaction (Table 1). About 31% of herds from the Northern districts of the Hoopstad area (Fig. 1; Table 1) tested positive on both occasions. This figure is obviously more than the 12,5% and 8,9% positive reactions found in the Lichtenburg and Potchefstroom areas respectively.

A total of 230 farmers responded to the questionnaire (Table 2). Due to the poor response, the data could not

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National Co-operative Dairies, Heilbron

Table 1: Incidence of bovine brucellosis in industrial milk producing

Area	Number one sample positive	Number of Farms	
Hoopstad ** . (southern districts)	81 (21,9%)	97 (26,3%)	. 369
Hoopstad * (northern districts)	 20 ( 9,8%)	63 (30,9%)	207
Lichtenburg	105 (10,4%)	126 (12,5%)	1005
Kroonstad	26 ( 9,1%)	74 (26,0%)	285
Potchefstroom	15 ( 6,3%)	21 ( 8,9%)	237
Total	247 (11,7%)	381 (18,1%)	2103

Hoopstad area; north of Vaal river.

Table 2: Heifer calf vaccination with strain 19 vaccine.

Action on farms	Number o	of herds%
Heifer calves not vaccinated at		
all	21	9,1
Heifer calves vaccinated once a	t	
age of 3 to 10 months	198	86,1
Heifer calves vaccinated at age of 3 to 10 months and again at		
18 to 24 months	11	4,8
Number of farms	230	-
Heifer calves vaccinated since:		
1955-59	8	3,8
1960-64 -	2	1,0
1965-69	16	7,6
1970-74	31	14,8
1975-79 •	53	25,4
1980-84	82	39,2
1985 to date	17	8,1
Number of farms	209	_

be broken down for a comparison in the different areas. It is, however, interesting to note that 9,1% of the respondents have not been practising heifer calf vaccination. Only 12,4% of farmers commenced with vaccination prior to 1970, whilst 8,2% commenced with vaccination during 1985 or later.

### DISCUSSION

Since only a small quantity of antigen is required to test a relatively large milk sample, the BRT is considered to be convenient, cheap and a very sensitive procedure for detecting bovine brucellosis<sup>4</sup>. There are a number of reasons for obtaining false positive and false negative BRT reactions in a bulk tank sample. False positive BRT reactions can be expected when fresh milk, colostrum, or milk from cows in the drying-off period is tested or in cases of mastitis or after recent brucellosis vaccination. False negative BRT reactions, on the other hand, could be expected in milk heated to 45°C for 5 min, milk excessively agitated or in infected cows when Brucella infection is absent in the udder<sup>2</sup>. For the purposes of this paper, only those herds where both samples resulted in a positive BRT reaction, were regarded as positive. Where one positive BRT reaction was obtained, the herd was designated suspicious for brucellosis. Based on these assumptions, about 18,1% of industrial milk producing dairy herds in the region, could be regarded as being infected with this disease.

It is generally agreed that heifer calf vaccination with S19 vaccine alone, cannot eradicate brucellosis from a herd. The effect of vaccination however, leads to the reduction of infected material excreted and thus to reduction of spread of infection to other animals within such a herd<sup>3</sup>. The positive effect of heifer calf vaccination is clearly demonstrated by Faul & Bosman<sup>1</sup>, who observed that the lowest incidence of brucellosis was experienced in that province of the Republic of South Africa where strict control over heifer calf vaccination has been maintained for the past 18 years. In contrast, the majority of the respondents have practised heifer calf vaccination for 10 years or less. Another 9,1% of these farmers (Table 2) have not been practising heifer calf vaccination at all.

If these figures could be extrapolated to the results of the BRT survey, it could be said that a vast number of adult females in the dairy herds of the region are, even at this point in time, fully susceptible to bovine brucellosis.

Although heifer calf vaccination with S19 vaccine is a legal obligation since 1968, one of the reasons for the high incidence of brucellosis in the different areas can be assumed to be the fact that meticulous heifer calf vaccination has not been practiced in the region.

### **ACKNOWLEDGEMENTS**

The authors wish to thank the field staff of National Co-operative Dairies for collecting and the laboratory staff for testing the samples.

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<sup>\*\*</sup> Hoopstad area; south of Vaal river.

# OESTRUS SYNCHRONIZATION IN DAIRY HEIFERS USING A PROGESTERONE RELEASING INTRAVAGINAL DEVICE

### D.C. LOURENS\*

ABSTRACT: Lourens D.C. Oestrus synchronization in dairy heifers using a progesterone releasing intravaginal device. Journal of the South African Veterinary Association (1988) 59 No. 1, 41-43 (Eng). Department of Theriogenology, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, 0110 Onderstepoort, Republic of South Africa.

This paper describes a pilot trial carried out, to investigate the use of progesterone releasing intravaginal device(PRID) in controlled breeding in heifers under South African conditions. PRID's containing progesterone and oestradiol benzoate were employed for oestrus synchronization and reproductive management in ten 18-month-old, well-managed, heifers in a commercial Transvaal highveld dairy herd. PRID's were inserted into the vagina of each animal and removed after 12 days. Oestrus observation was done continuously and on the first observed oestrus the heifers were artificially inseminated once. Oestrus detection continued normally for returns to oestrus. Pregnancy was confirmed by rectal palpation eight weeks after breeding if no return to oestrus had occurred. The incidence of clinical oestrus within 3 days after removal of the PRID's (defined as the oestrus response) was 90%. The conception or pregnancy rate following the first artificial insemination was 77.8%.

Key words: Oestrus synchronization, dairy heifers

### INTRODUCTION

There are two basic approaches available for the successful synchronization of oestrus and ovulation in cattle. The first makes use of natural progesterone or synthetic progestogens, alone or in combination with oestrogens, whereas the other approach involves the use of prostaglandin  $F_{2\alpha}$  or synthetic analogues which are luteolytic<sup>3 4 5 10 11</sup>.

Extensive field trials have also been conducted with combined treatments involving progesterone intravaginally or progestogen implants for variable periods followed by prostaglandins and or pregnant mare serum gonadotrophin, before, at, or after withdrawal of the progestogen<sup>14678</sup>.

Progesterone containing drugs may be administered in various ways. They may be administered by daily injections (progesterone in oil) or orally (synthetic progestogens). The numerous parenteral injections, the unreliability of the oral route of administration, and the subsequent poor results have led to the development of subcutaneous implants and intravaginal devices<sup>3 6 7</sup>. A subcutaneous implant containing the potent synthetic progestogen, Norgestomet, is currently widely used<sup>3 6 10</sup>.

The use of intravaginal sponges (similar to those used in sheep) have resulted in better conception rates when compared to the parenteral or oral routes of administration. A disadvantage, however was a high rate of loss of the sponges<sup>67</sup>. This has in turn led to the development of a solid phase intravaginal system for the delivery of progesterone<sup>29</sup>.

A progesterone releasing intravaginal device (PRID, Abbott Laboratories) is a stainless steel spiral coated with inert silicone rubber containing progesterone. This provides a large surface area which remains in contact with the vaginal mucosa leading to a continuous supply and absorption of exogenous natural progesterone to

 Department of Theriogenology, Faculty of Veterinary Science, University of Pretoria, P. Bag X04, 0110, Onderstepoort, Republic of South Africa the recipient animal<sup>269</sup>. Oestradiol benzoate in a gelatin capsule is attached to the coil. The inclusion of oestradiol has been shown to result in higher conception rates in synchronized oestrus<sup>29</sup>.

Progesterone releasing intravaginal devices (PRID) have been extensively investigated and used in some countries. To date PRID's have not been commercially available in the Republic of South Africa. This paper describes a pilot trial which was carried out to investigate the oestrus response and the first service conception rate of heifers following the use of PRID's in a small group of well-managed heifers.

### MATERIALS AND METHODS

The experiment was carried out in a commercial dairy farm on the Transvaal highveld during March, 1986. The general management and nutrition on the farm was of a high standard.

Ten 18-month old well-grown Holstein heifers were selected for the trial. To assess the reproductive status, the reproductive tracts of all heifers were palpated per rectum immediately before treatment commenced. The heifers had been fed on good quality maize silage, which was supplemented with a commercial high protein-mineral summerlick and yellow maize meal. This ration was fed from 2 weeks prior to the commencement of the trial.

The PRID's (Abbott Laboratories) contain 2,25g of natural progesterone and 10 mg oestradiol benzoate. A speculum and rod were used to insert the PRID into the anterior vagina according to instructions of the manufacturer. When the speculum is removed, the coil is expelled into the vagina leaving a previously attached nylon string protruding from the vulva which is used for removal. The PRID's were left in situ for 12 days<sup>9</sup>.

The heifers were observed continuously for signs of oestrus following PRID removal. At the first observed oestrus after PRID removal, heifers were artificially inseminated once by a proficient inseminator. Oestrus observations were continued as part of the normal

Table 1: Oestrus response and first insemination conception rate

Date PRID in : 10.3.86 Date out : 22.3.86

Heifer	Oestrus/	Pregnancy e	xamination	PRID rer	noval to	Inseminations per
No.	Insemination dates	Date	Result	First oestrus	First insemi- nation	Conception
84116	24/3 .	26.5.86	+	2 days	2 days	1
84110	2 <u>4</u> /3"	26.5.86	+	2 days	2 days	1
84107	23/3	26.5.86	+	1 day	1 day	1
84109	18/4 (not insemi- nated), 6/5, 27/5.	23.7.86	+	27 days	45 days	2
84103	24/3, 5/6, 14/7, 30/7, 22/8	14.11.86	+	2 days	2 days	5
84124	23/3, 24/5	23.7.86	+	1 day	1 day	2
8488	24/3	26.5.86	+	2 days	2 days	1
84111	24/3	26.5.86	+	2 days	2 days	1
84117	25/3	26.5.86	+	2 days	3 days	1
84106	23/3	26.5.86	+	1 day	1 day	1

Oestrus response : 9/10 - 90% First insemination conception rate : 7/9 - 77,8%

routine on the farm and heifers repeating during subsequent oestrus periods were inseminated again. After a non return period of 8 weeks heifers were examined rectally for pregnancy which was reconfirmed at 90 days.

### **RESULTS**

After the insertion of PRID's all heifers showed a small amount of blood on the speculum as well as slight discomfort for a day after the insertion. The retention rate was 100%.

On removal of the PRID's a thick brownish mucoid discharge was observed in all heifers which had completely disappeared by the time of insemination.

All relevant data on occurrence of oestrus, inseminations, interval from PRID removal to first oestrus and insemination, pregnancy diagnosis and number of inseminations per conception is shown in Table 1.

The signs of oestrus were excellent in all the heifers showing synchronized oestrus.

### **DISCUSSION**

The insertion of the PRID was a fairly simple procedure which resulted in minimal trauma and discomfort.

The thick brownish mucoid vaginal discharge observed on removal of the device was probably due to the accumulation of vaginal secretions and secondary bacterial growth. The condition usually rapidly resolves following on removal of the PRID's and has no adverse effect on fertility<sup>9</sup>. This fact was clearly demonstrated in that at the time of oestrus each heifer exhibited a clear bullstring. An excellent oestrus response was obtained (Table 1).

The degree of synchronization observed in this small trial was comparable with that reported in the literature<sup>4</sup>. The stage at which the PRID is inserted, is an important determinant of the overall response<sup>12</sup>. The inclusion of oestradiol benzoate has also been shown to improve the results<sup>12</sup>. Good results have also been reported when the PRID was used in combination with prostaglandins<sup>1248</sup> and pregnant mare serum gonadotrophins<sup>7</sup>.

The first insemination conception rate (Table 1) on observed oestrus correlates very well with reported results of 72-82%<sup>8</sup>.

Extensive reported experimental investigations have, however, been based on fixed time single or double inseminations. This relieves the farmer of the need to observe oestrus and has the additional benefit of giving non-oestrus but synchronously ovulating animals the opportunity of conceiving. Conception rates following fixed time inseminations, have however yielded variable results with synchronization programmes.

The cost factor with PRID is an important factor to consider. There are indications that PRID may be used more than once, and this should result in a significant reduction in the cost of this technology<sup>5</sup>.

In conclusion, the excellent synchrony of oestrus and high fertility after PRID treatment obtained in this small trial, emphasizes the fact that this regimen would be precise enough to provide acceptable pregnancy rates when insemination is carried out on observed oestrus under conditions of optimal management and nutrition. Different nutritional and environmental factors may cause the synchrony of oestrus to vary considerably among groups of animals synchronized by any scheme even under controlled experimental conditions<sup>3 6 8 9 10</sup>. Users of oestrus control programmes must always be aware of this variability and that it may effect their results.

### **ACKNOWLEDGEMENTS**

The author wishes to thank Mr W van Zyl, the dairy farm manager of Beeskor for providing the heifers and for his skillful assistance with oestrus observation and artificial inseminations as well as Maybaker S.A. (Pty) Ltd, P O Box 1130, Port Elizabeth for providing the PRID's. Sister M E Louw is thanked for technical assistance.

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BOOK REVIEW BOEKRESENSIE

### **MYCOTOXICOLOGY**

### W.F.O. MARASAS and PAUL E. NELSON

The Pennsylvania State University Press, University Park, Pennsylvania, 16802 USA and London, 1986 pp 102, illustrations 76, Price \$34,50 (ISBN 0-271-00442-8).

This book is more than just an introduction to some naturally occurring mycotoxicoses as claimed by the authors; it is an up-to-date, lucid, concise account of several important veterinary mycotoxicoses. Having one of South Africa's most noted mycotoxicologists as a senior author has ensured that local aspects of these mycotoxicoses have not been ignored. Eleven mycotoxicoses are dealt with, of which seven — aflatoxicosis, stachybotryotoxicosis, vulvo-vaginitis (hyperoestrogenism), equine leukoencephalomalacia, facial eczema and lupinosis — are known to occur in South Africa. Of the remaining conditions described, one (mouldy sweet patato toxicosis) is suspected of occurring in this country. Another three (ergotism induced by *Claviceps purpurea*, ochratoxicosis and possibly 'feed refusal') are similar to syndromes occassionally seen here. The book has wide application in southern Africa and, since the authors had mainly North American readers in mind, they can be forgiven for omitting diplodiosis and *Aspergillus clavatus* poisoning.

The information on each mycotoxicosis is presented in a standardised format and includes the mycology, phytopathology, chemistry, clinical signs, epidemiology, pathology and control. The colour plates and black and white illustrations are excellent. As a unique marketing feature the 48 colour slides used in the book can be purchased for an additional outlay of \$100.

I highly recommend this book to veterinary students, veterinary practitioners, colleagues in the feed-processing industries, extension specialists and anyone else who has an interest in mycotoxicology.

T.S. Kellerman

### DERMATOSPARAXIS IN WHITE DORPER SHEEP

A. VAN HALDEREN\*, JENNIFER R. GREEN

ABSTRACT: Van Halderen A.; Green Jennifer R. Dermatosparaxis in White Dorper sheep. Journal of the South African Veterinary Association (1988) 59 No. 1, 45 (En). Regional Veterinary Laboratory, Private Bag X5020, 7600 Stellenbosch, Republic of South Africa.

A diagnosis of dermatosparaxis in a lamb with clinical signs of extreme skin fragility was based on the gross changes as well as on light and scanning electron microscopical observations. Similar cases had occurred on the same farm during the previous year.

Key words: Sheep, White dorper, dermatosparaxis

A White Dorper breeder in the Porterville district of the Western Cape Province reported an unusual condition in his lambs in August 1985. Nine out of 900 lambs of 3 weeks to 2 months of age showed signs of extreme skin fragility with tearing off of large strips of skin. The lambs had to be euthanased for humane reasons. This condition was first noticed during the 1984 lambing season when fewer cases occurred. The farmer bought in 4 White Dorper rams from another breeder in 1983 and since then used these rams for breeding.

Apart from the obvious skin lesions there were no other macroscopical changes<sup>3</sup>. Skin sections of the affected lamb and a normal control lamb of the same age were prepared for histopathological examination and stained with haematoxylin and eosin as well as with the Warthin-Starry method. Sections of skin were also prepared for scanning electron microscopy. The skin from the affected lamb showed markedly thinner, more irregular and less dense collagen bundles in the dermis<sup>2,3</sup> than the control lamb. This was especially evident in the sections stained with the Warthin-Starry method and those viewed under the scanning electron microscope (Fig 1 & 2).

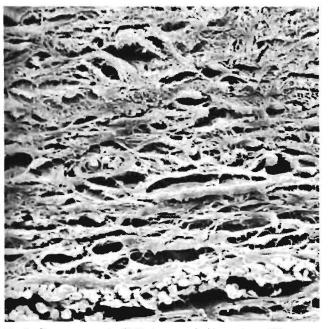


Fig. 1: Dermatosparaxis: SEM photograph skin section x 500.

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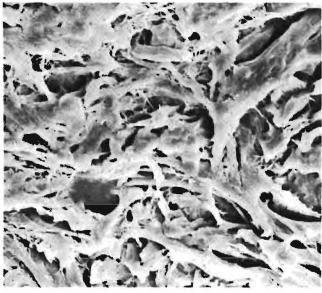


Fig. 2: Normal lamb: SEM photograph skin section x 500

A diagnosis of dermatosparaxis was made based on the clinical signs, histopathological and scanning electron microscopical observations.<sup>123</sup>. This is a simple, autosomal, recessive hereditary condition that has been described in cattle, sheep and dogs<sup>12</sup>. Skin fragility is caused by impaired collagen synthesis arising from an enzyme deficiency for the conversion of procollagen to collagen<sup>12</sup>. As far as is known it is the first report of dermatosparaxis in White Dorper. Hypermobility of the joints as described by McOrist et al.<sup>3</sup> has not been observed in affected sheep. Further cases have subsequently been reported from two other White Dorper breeders.

### ACKNOWLEDGEMENTS

We would like to thank Dr H J de Wet for referring the case, Mr P van der Merwe of the Research Institute for Fruit Technology for the scanning electron microscope photographs and Mrs R de Kock for typing the manuscript. The Director of Veterinary Services is thanked for permission to publish this note.

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# AN UNDESCRIBED RHIPICEPHALUS SPECIES ASSOCIATED WITH FIELD PARALYSIS OF ANGORA GOATS

L.J. FOURIE\*, I.G. HORAK\*\* and L. MARAIS\*\*\*

ABSTRACT: Fourie L.J.; Horak I.G.; Marais L. An undescribed Rhipicephalus species associated with field paralysis of Angora goats. Journal of the South African Veterinary Association (1988) 59 No. 1, 47 -49 (En). Department of Zoology and Entomology, University of the Orange Free State, 9300 Bloemfontein, Republic of South Africa.

Paralysis of Angora goat kids caused by adult ticks, which most probably belong to an undescribed species of the *Rhipicephalus* pravus group, is described. Confirmed cases of paralysis occurred in the south-western Orange Free State between the second half of September and the first half of November as well as during the first half of February. The mean female tick burdens ( $\bar{x} = 21,4$ ) of paralysed Angora kids were significantly higher than those of healthy kids ( $\bar{x} = 4,4$ ). The predilection attachment site of the adult ticks was the ears of the goats. The elephant shrew *Elephantulus myurus* is a preferred host of the immature stages whereas the scrub hare *Lepus saxatilis* serves as an important host for the adult ticks.

Key words: Rhipicephalus sp., Angora goats, paralysis

### INTRODUCTION

Eight different forms of tick toxicosis are known, of which tick paralysis is the most important<sup>1</sup>. Fifty-three of the approximately 800 described tick species have been implicated as a cause of tick paralysis. The pathophysiology of this syndrome has been studied in detail for paralysis produced by Argas walkerae, Dermacentor andersoni, Ixodes holocyclus and Rhipicephalus evertsi evertsi<sup>1 3 4</sup>. Clinical signs, which are generally very similar, are seen 4-7 days after tick attachment<sup>2 3 4</sup>. Initial signs are lower motor neuron paralysis with varying degrees of incoordination, ataxia and muscular weakness, progressing to total paralysis with the animal in lateral recumbency. Death is usually the result of respiratory paralysis or sometimes aspiration pneumonia<sup>8</sup>.

In South Africa the engorging females of the ixodid ticks R. evertsi evertsi and Ixodes rubicundus may both cause paralysis of small stock<sup>2</sup> <sup>6</sup>. During an intensive study of the epidemiology of Karoo paralysis caused by I. rubicundus in Fauresmith district, a tick apparently belonging to the Rhipicephalus pravus group was observed to cause paralysis and mortalities among Angora goat kids. This paper records the tick burdens of animals paralysed by this tick, the predilection attachment sites and seasonal occurrence of the ticks and also comments on the natural hosts of this tick.

### MATERIALS AND METHODS

The majority of observations were made on the farm Preezfontein (29° 50'S; 25° 23'E) which is situated near the town of Fauresmith in the south-western Orange Free State. Fourteen of 51 Angora kids became paralysed on this property during the second half of September 1986. The ears of the remaining kids (except for 5 'tracers') were treated with a tick dressing (Milborrow

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Tick Dressing (chlorfenvinphos 3% mm)) at approximately 2 week intervals. An attempt was made to observe the 5 untreated kids on a weekly basis from October — December 1986 in order to collect the *Rhipicephalus* sp. responsible for causing paralysis as well as any other ticks present. In each case the sex of the ticks and attachment sites were noted. Kids which developed paralysis were weighed, the ticks removed and the recovery time recorded. Ticks were also counted on the treated animals.

The seasonal abundance of this tick species was determined in a flock of 60 adult sheep which utilized the same habitat as the Angora goats. From January to December 1986, 20 randomly selected sheep from this flock were thoroughly examined once a week for the presence of ticks. The natural hosts of the ticks were determined from a large variety of small and large mammals and birds which were shot or trapped on a monthly basis. These animals were processed for tick recovery by methods described by Horak and Fourie<sup>5</sup>.

Cases of paralysis of small stock in other districts were also investigated.

### **RESULTS**

Although all the ticks were removed from the 14 Angora goat kids which intially became paralysed, 10 subsequently died giving a mortality rate of 71%. These kids were less than one month of age.

All 5 'tracer' Angora kids became paralysed in the period between the second half of September and the first half of November. These animals harboured mean burdens ( $\pm$  S.E.) of 21,4  $\pm$  5,0 female ticks of an undescribed *Rhipicephalus* species similar to *Rhipicephalus pravus* compared with 4,4  $\pm$  0,9 female ticks of the same species carried by 23 healthy but treated kids from the original flock. The masses of the paralysed Angora kids varied between 2,6 and 5,0 kg.

The first of the 5 experimental Angora kids which showed signs of paresis was allowed to become completely paralysed after which all the ticks were removed. This kid died approximately 5 hours after becoming completely paralysed. A total of 25 engorging Rhipicephalus sp. females was removed. Ticks were

removed from the other 4 experimental kids when they showed clinical signs of leg weakness, in order to prevent further mortalities. Between 10 and 38 engorging *Rhipicephalus* sp. females were collected from these kids which recovered completely 1 to 2 days after tick removal. Apart from the unknown *Rhipicephalus* sp. tick, two *Hyalomma marginatum rufipes* ticks were found on one of the Angora kids which showed signs of paresis. Five of the healthy kids also harboured *Hyalomma* spp.

The sex ratio of the ticks on the Angora kids did not differ significantly (p > 0.05) from parity (1.00%) 0.99%). Most ticks attached in and on the ears (n = 353). Ticks were also recovered from the anogenital region (n = 23) and the neck (n = 20) with the smallest number (n = 10) found around the buds of the developing horns.

In comparison with the high burdens of the undescribed Rhipicephalus sp. found on Angora kids (up to 38m and 45m per individual), the burdens of this tick species on adult sheep were consistently low. The maximum number of Rhipicephalus sp. collected from an individual sheep was 14. Although adults of this *Rhipicephalus* sp. occurred on the sheep throughout the course of the year, the highest numbers were recorded during spring and summer. Tick counts done on a large number of small mammals and birds (Table 1) indicated that the elephant shrew Elephantulus myurus and the scrub hare Lepus saxatilis serve as important natural hosts of the immature and adult ticks of this species respectively. The immature stages were identified by comparing them to laboratory-reared specimens.

Table 1: List of birds and small mammals examined for the presence of the *Rhipicephalus pravus*-like tick

Species	Birds Common name	Sample size
Oena capensis	Namaqua Dove	3
Myrmecocichla formicivora	Anteating Chat	4
Galerida magnirostris	Thickbilled Lark	17
Stenostira scita	Fairy Flycatcher	1
Mirafra apiata	Clapper Lark	4
Mirafra passerina	Monotonous Lark	1
Chersomanes albofasciata	Spikeheeled Lark	34
Calandrella cinerea	Redcapped Lark	3
Motacilla capensis	Cape Wagtail	2
Anthus novaeseelandiae	Richard's Pipit	3
<i>Anthus</i> sp.		3 2 3 7 2 2
Anthus similis	Longbilled Pipit	2
Anthus leucophrys	Plainbacked Pipit	2
Pycnonotus nigricans	Redeyed Bulbul	1
Oenanthe pileata	Capped Wheatear	1
Cercomela sinuata	Sicklewinged Chat	1
Cursorius temminckii	Temminck's Courser	1
Amadina erythrocephala	Redheaded Finch	1
Serinus sulphuratus	Bully Canary	1
Prinia flavicans	Blackchested Prinia	1
Cisticola fulvicapilla	Neddicky	1
	Mammals	
Elephantulus myurus	Rock Elephant-shrew	52
Lepus capensis	Cape Hare	33
Lepus saxatilis	Scrub Hare	47
Pronolagus rupestris	Smith's Red Rock Rabbit	25
Pedetes capensis	Spring Hare	3
Rhabdomys pumilio	Striped Mouse	3
Aethomys namaquensis	Namaqua Mouse	238
Saccostomus campestris	Pouched Mouse	13
Malacothrix typica	Large-eared Mouse	1

Confirmed cases of paralysis caused by the *Rhipicephalus* sp. tick were also recorded in the Jagersfontein and Philippolis districts. In one of these cases, 52 of 140 five month old Angora kids became paralysed during the first 2 weeks of February 1987 and 35 subsequently died. In addition to the undescribed *Rhipicephalus* sp. a few *Hyalomma* spp. were also present on some of these Angora kids.

### **DISCUSSION**

No laboratory studies have as yet been conducted to confirm that this *Rhipicephalus pravus*-like tick produces paralysis. However, the findings reported above indicate that there is a relationship between burdens of this tick and cases of paralysis in Angora goat kids in the field. This is supported by the fact that no ticks of other species known to cause paralysis in small stock were recovered from the affected kids and that the occurrence of paralysis in late September to early November and during February, does not correspond to the times during which *I. rubicundus* generally causes paralysis (March-August).

Some of the affected goats recovered when the ticks were removed. Those that did not recover were possibly already too severely affected for recovery to take place.

The exact taxonomic status of this paralysis inducing tick is still unclear. During a survey conducted in 1949, ticks resembling Rhipicephalus appendiculatus were collected at Fauresmith and the surrounding district. Subsequent collections, however, indicated that these ticks were actually Rhipicephalus pravus specimens?. Walker (1987, Veterinary Research Institute, Onderstepoort, personal communication) examined some of the adult ticks collected from the paralysed Angora kids and is of the opinion that they may represent an undescribed species in the Rhipicephalus pravus group. The taxomonic status of this tick is currently being investigated.

The higher tick burdens on paralysed animals compared to non-paralysed animals was not unexpected. Gothe and Bezuidenhout<sup>2</sup> demonstrated that the intensity of tick paralysis caused by R. evertsi evertsi was directly related to the infestation rate. It is not, however, the number of ticks per se that influences the degree of paralysis, but rather the number of ticks at a certain stage of engorgement. Only female R. evertsi evertsi in the mass range of 15-21 mg are reported to be toxic<sup>2</sup>. Comparisons between the pathogenicity of the R. pravus sp. tick and that of other paralysis ticks can, however, only be made after detailed in vivo studies. Data on the geographic distribution of this tick will assist with the collection of various strains and the determination of possible intraspecific differences in their paralysis-inducing capacity.

Because of the paucity of information no recommendations regarding control measures can be made at this stage. The fact that the predilection site of this tick is the ears of the host may complicate chemical control.

### **ACKNOWLEDGEMENTS**

We would like to thank Mr J van Niekerk of the farm "Preezfontein" for the use of the facilities on his farm.

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RESEARCH NOTE NAVORSINGSNOTA

### THE GENETIC RELATIONSHIP BETWEEN THE DÖHNE- AND THE WALRICH-MERINO

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ABSTRACT: Osterhoff D.R.; Groenewald J.C.; Nürnberger H.; Van Vuuren C.H. The genetic relationship between the Döhneand the Walrich-Merino. Journal of the South African Veterinary Association (1988) 59 No. 1, 51-52 (En). Department of Zootechnology, Faculty of Veterinary Science, University of Pretoria, 0110 Onderstepoort, Republic of South Africa.

The Döhne- and the Walrich Sheep breeders' societies amalgamated in 1982 to form an enlarged Döhne Merino Breed Society. Both breeds are dual-purpose, polled breeds with high fertility as well as good wool and mutton characteristics. Apparently they have a similar genetic background, both being developed out of local Merino ewes and dual-purpose Merino-type sheep from Europe. Studies of biochemical polymorphisms in the blood of 224 Döhne and 204 Walrich Merino established a close relationship between these two breeds.

Key words: Genetic markers, biochemical polymorphism, Döhne Merino breed, Walrich Merino breed

### INTRODUCTION

Döhne and Walrich sheep breeders, after many years of negotiations, amalgamated in November 1982. Both breeds are dual-purpose, polled breeds with a high fertility, good mutton characteristics and a strong constitution. Both carry high-quality Merino wool falling in the 20 to 22 micron range. Their breed standards are identical and they are phenotypically indistinguishable.

The Döhne Merino is a synthetic breed that was developed by the Department of Agriculture at the Agricultural Research Institute at Döhne. The late Mr J J J (Koot) Kotzé, former head of the Döhne Experimental Station, began breeding trials in 1939 using Merino ewes and various mutton-type rams. The crosses derived from imported German Merino rams proved to be so well adapted and successful that a developing programme was started. The high production potential of the Döhne and its efficiency and adaptability in tough commercial environments resulted in the phenomenal growth of the society, and the spread of the breed to all parts of South Africa.

The founder of the Walrich breed was the late Mr W A (Jock) Higgs, a Merino stud breeder from the farm Richmond, in the Zastron district. His aims were realised by mating his Merinos in 1932 to French Précoce Merino rams which led to the development of a synthetic breed. When a breed society was formed, it was decided to give the breed a distinctive name — Walrich — derived from Jock Higgs's first name, Walter, and the name of his stud, Richmond.

The Döhne Merino and Walrich breeds are thus similar because they both have the local Merino as female parent, and closely related European breeds as male parents. In the present study genetical markers were used to investigate the relationship between these two breeds.

### MATERIALS AND METHODS

The techniques for the determination of genetic markers in sheep blood have been described extensively by Tucker<sup>4</sup>.

 Department of Zootechnology, Faculty of Veterinary Science, University of Pretoria, 0110 Onderstepoort, Republic of South Africa Blood samples (n = 428) of the two breeds were collected from sheep on four different farms: the Döhne Merino samples from the Döhne nucleus stud (Dr T Laas) and from the Potchefstroom Agricultural College, and the Walrich samples from the farms of Mr D Steyn, Breyten and Mr D Hatting, Heilbron. More than 100 samples in each case from each farm could be collected.

Data of other South African Breeds, the South African Merino, the Letelle and the Namaqua used in a previous study on genetic polymorphism in South African Merino sheep<sup>2</sup>, as well as data from Spanish Merino<sup>3</sup> were included in these breed comparisons.

### **RESULTS**

Gene frequencies of haemoglobin, transferrin and "X"-proteins are given in Table 1. In Table 2 gene frequencies of carbonic anhydrase, aryl esterase and nucleoside phosphorylase are given.

### **DISCUSSION**

Table 1 clearly indicates that the haemoglobin gene frèquencies between the Döhne and Walrich Merino are closer to each other than to the other breeds included in the table. The same can be said of the transferrin gene frequencies.

Albumins are almost 100 percent of the slow moving type; they are thus not of great help in the identification of breed differences, and are therefore not presented.

When sheep red cell lysates are subjected to electrophoresis in starch gels at pH 7.6, another zone which exhibits polymorphism can be demonstrated. This protein has not yet been identified and is given the preliminary designation of 'X'. It comprises three bands which are either all present or all absent. The two phenotypes are X-positive and X-negative and are inherited as if controlled by a pair of autosomal alleles. Again, in Table 1 it can be seen that the Döhne and the Walrich merino are the closest related, because the gene frequency in this polymorphism is very similar.

In almost all breeds only the slow migrating carbonic anhydrase band has been discovered<sup>4</sup> and both the Döhne and the Walrich Merino are falling in line with these early discoveries.

Table 1: Comparison of gene frequencies in certain protein types

		Haemo	oglobin			Transferrin			"X"-	protein
	N	ньА	ньВ	TfA	TfB	$Tf^C$	TfD	TfE	×	x
 Döhne	224	.125	.875	.292	.257	.438	.013	,000	.557	.443
Walrich	204	.164	.836	.316	.186	.375	.123	.000	.664	336
Spanish Merino	442	.327	.673	.165	.164	.126	.485	.060	.414	.586
S A Merino	162	.309	.697	.246	.042	.352	.328	.032	.191	.809
Letelle	64	.336	.664	.025	.123	.467	.377	.008	.099	.901
Namagua	<sup>2</sup> 85	.065	.935	.214	.007	.013	.766	.000	.000	1.000

Table 2: Comparison of gene frequencies in certain enzyme types

	•	Carbonic anhydrase		Aryl e	sterase	Nucleoside phosphorylas	
	N	CaS	CAF	EsA +	EsA —	NPH	NPL
Döhne	224	1.000	.000	.131	.869	.529	.471
Walrich	204	1.000	.000	.259	.741	.539	.461
Spanish Merino	442 .	.952	.048	.179	.821	-	_
S A Merino	162	.988	.012	.016	.984	.926	.074
_etelle	64	.992	.008	.375	.625	.883	.117
lamaqua	85	1.000	.000	.018	.982	.576	.424
	. [						

Some serum samples show a brown "smudge-like" zone of staining which developes rapidly within 15 min when alpha-naphthyl acetate is used as substrate. This zone was classified as being aryl esterase dividing sheep into either Es A positive or Es A negative sheep. Some differences between the Döhne and Walrich Merino breeds are indicated but these two breeds are also in this system closer to each other than the other breeds included in the comparison.

Nucleoside phosphorylase (NP) (EC 2.4.2.1) catalyses the phosphorolyteic cleavage of certain purine nucleosides. The enzyme is widely distributed in animal tissues and inherited variants have been reported in human. Tucker described for the first time the NP activity of sheep red cells and established the inherited deficiency of NP in the red cells of certain sheep<sup>5</sup>. Again the gene frequencies of the Döhne and Walrich Merinos are those which indicate the closest relationship. No explanation can be given for the observation that the Namaqua sheep seem to be nearer to these two-breeds.

Summarising all these results it appears that the Döhne and Walrich breeds are closer to each other than

to the other breeds included in the study. It can therefore be said that the amalgamation of the two breeds can be regarded as a distinct advantage for both breeds, providing a greater genetic pool which can only lead to better selection and improvement.

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# THE SUPPRESSION OF ELECTRO-EJACULATION IN THE CHACMA BABOON (PAPIO URSINUS) BY AZAPERONE

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ABSTRACT: Meltzer D.G.A.; Van Vuuren M.; Bornman M.S. The suppression of electro-ejaculation in the Chacma baboon (Papio ursinus) by azaperone. Journal of the South African Veterinary Association (1988) 59 No. 1, 53 (En). Roodeplaat Research Laboratories, P.O. Box 13873, 0129 Sinoville, Republic of South Africa.

Six baboons (*Papio ursinus*) males were immobilised on five occasions at intervals of two weeks using a combination of ketamine hydrochloride and xylazine. The animals were subsequently successfully electro-ejaculated. No semen was obtained from animals when azaperone was used instead of xylazine, despite the use of a successful standardised method of electrical stimulation.

Key words: electro-ejaculation, Papio ursinus, azaperone

### INTRODUCTION

The use of ketamine and xylazine for the immobilization of non-human primates has been described by Booth<sup>2</sup>. This drug combination is used routinely for the immobilisation of Chacma baboons kept at the Roode-plaat Research Laboratories. However, xylazine was found to have a prolonged sedative effect and azaperone was therefore used in its place. Azaperone, a butyrophenone derivative, produces its neuroleptic effect by a central adrenergic blockade<sup>2</sup>. It is used extensively in clinical practice for the sedation and restraint of pigs<sup>2</sup> and also in combination with etorphine or carfentanil for the immobilisation and capture of freeranging wild herbivores<sup>45</sup>.

### MATERIAL AND METHODS

Six chacma baboons were immobilised with ketamine (Ketalar, Parke Davis) at a dosage rate of 15 mg kg<sup>-1</sup> together with 5 mg total dose of xylazine (Rompun, Bayer). Semen was collected using a standard electrical stimulation procedure as described by Bornman et al<sup>3</sup> every 2 weeks, for a period of 10 weeks. Fresh preparations of the semen collected were evaluated with minor modifications<sup>3</sup> by the standard procedure for man<sup>6</sup>. Two weeks later, azaperone (Stresnil, Janssen Pharmaceutica) was used together with ketamine in the same animals at a dosage rate of 20 mg per animal. The standard electrical stimulation procedure was used for semen collection.

### **RESULTS**

The mean semen density obtained from the 6 baboon males during the period of the trial ranged from 69 - 136 x 106 ml<sup>-1</sup> when ketamine and xylazine were used. When azaperone was used instead of xylazine in the im-

mobilizing drug mixture, no ejaculates were obtained and four of the animals urinated in response to the electrical stimulation.

### DISCUSSION

In man, emission and ejaculation, together with bladder neck closure, have been shown to be controlled, primarily, by the sympathetic nervous system acting through alpha-adrenergic receptors<sup>1</sup>. Butyrophenone derivatives produce their effects through an adrenergic receptor blockade which appears to interfere with the usual ejaculatory response to an electrical stimulus. It is therefore advisable to avoid the use of this group of neuroleptic drugs when immobilising animals to be electro-ejaculated.

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## PRACTICAL SMALL ANIMAL DERMATOLOGY II: DIAGNOSTIC APPROACH

### O.M. BRIGGS\*

ABSTRACT: Briggs, O.M. Practical small animal dermatology II: Diagnostic approach. Journal of the South African Veterinary Association (1987) 59 No. 1, 55-58 (En). St. Francis Veterinary Hospital, 157 Main Road, Bergvliet, 7800 Cape Town, Republic of South Africa.

The approach to the evaluation of a small animal patient with a dermatological condition is discussed with emphasis on systematic methods. The importance of obtaining an adequate history is stressed and the correct performance of screening tests is emphasised. Evidence of the body's reaction to disease or injurious agents is described since it must be recognised. Pruritus and alopecia are two common presenting clinical signs and systematic approaches to eliciting the aetiology of these are proposed.

### INTRODUCTION

When presented with a dermatological disease which cannot be immediately diagnosed, one should not resort to guesswork. Recognition of the way in which the skin reacts to the disease combined with a step-by-step diagnostic approach including systematic rule-outs is more rewarding to the pet, owner and clinician. There are a number of textbooks on veterinary dermatology to consult once a diagnosis has been established. The pet owner can be advised and a therapeutic approach initiated. However, an incorrect diagnosis sets off a chain-reaction of mishaps which can be frustrating and costly.

### **IDENTIFYING THE PROBLEM**

A skin complaint has often been observed by the pet owner for a considerable period prior to being presented to the clinician. The step-by-step approach (Table 1) must therefore include a detailed history (Table 2) with emphasis on the owner's observations. Where did the condition start? Where did it spread to next? Did the lesions appear prior to the itch or did the patient cause the pathology by scratching? The physical examination should begin with a complete clinical examination of the

Table 1: A systematic diagnostic approach

- Obtain a thorough history
- 2. Do a physical examination
- 3. Draw up a list of differentials
- 4. Carry out screening tests
- 5. Narrow the differentials6. Carry out definitive tests
- 7. Arrive at a diagnosis
  - Table 2: Taking a history
- Animal details (breed, age, sex, origin)
   History of other illnesses
- 3. Diet (type and quantity)
- 4. Environment (bedding, in-contacts)
- 5. Owner's observations
- 6. Evidence of transmission
- 7. Case history or referral details
- \* St Francis Veterinary Hospital, 167 Main Road, Bergvliet, 7800 Cape Town, Republic of South Africa

patient before concentrating on the skin. Direct examination of the skin must include all areas of the body.

### **SCRAPINGS**

The screening tests include microscopic examination of scrapings, direct smears and epilated hairs (Table 3).

Table 3: Diagnostic tests

- A. Direc
  - 1. Strong light and magnifier
  - 2. Woods lamp
- B. Microscopic
  - Scrapings
  - 2. Direct smears and fine needle aspirates
  - 3. Epilated hairs
- C. Growth on culture media
  - 1. Bacteria
  - 2. Fungi
- D. Biopsy
  - 1. Histopathology
  - 2. Immunofluorescence
- E. Hormone assays
  - Thyroid hormones
  - 2. Cortisol
- Sex hormonesBlood screens
  - Haematology
  - 2. Chemical profiles
- G. Allergy tests
  - 1. Hypoallergenic diet
  - 2. Patch testing
  - 3. Isolation and provocative exposure
  - 4. Intradermal allergy testing

Scrapings should be taken with a blade held vertically and the cutting edge scraped over the skin. Squeezing a skin fold while scraping for *Demodex* expels these parasites from the hair follicle. Collected epidermal debris is placed on a slide to which a drop of oil has been added. Examine first without a coverslip as parasites are often found because of their movement. The coverslip depresses the sample and spreads it. The use of 10% potassium hydroxide is considered essential by some clinicians. A few drops are added to the collected sample on the slide. This is then warmed (often by placing the slide on the microscope light apparatus for 2 minutes) until the debris has dissolved to reveal parasites or ectothrix spores.

### DIRECT SMEARS

A slide is pressed firmly onto the lesion. It is fixed and stained with a screening stain such as Diff-Quik (RapiDiff, Clinical Sciences Diagnostics). The "roof" of a pustule or vesicle can be lifted off (with a corner of the slide or a fine needle) prior to making the impression. The slide is examined microscopically for microbes and cell types (cytology). Other stains which can be used include Giemsa which stains the granules in mast cells blue.

### **NEEDLE ASPIRATES**

A sterile needle attached to a syringe is inserted into a nodule, tumour, or cyst and cells, fluid, or solid material withdrawn. The aspirate may be very slight but when expressed onto a slide and stained invariably reveals useful information. Do not immediately discard the aspirate. It may be necessary to send it to a laboratory for culture or further analysis.

### **WOODS LAMP**

Wood's lamp examination has limited application as it must be done in a darkened room (not always available) and only 40-60% of ringworm caused by *Microsporum canis* fluoresces. Lesions caused by *Trichophyton mentagrophytes* and *Microsporum gypseum* do not fluoresce. A more reliable test is to culture hair and scales on dermatophyte test medium (DTM) (Fungassay, Pitman-Moore). Although there is an incubation period of 3-5 days, it has the advantages of accuracy and ease of interpretation.

### **EPILATED HAIRS**

Hairs are removed from normal and diseased looking coat or skin with a small mosquito forceps and placed on a drop of mineral oil on a slide. A coverslip is applied. The preparation is examined for the presence of fungal spores, fractured hairs, mites attached to the hair root, the stage of the hair growth cycle (club root—telogen or resting phase; large, pigmented root—anagen or active phase). Telogen/anagen ratios are useful in determining abnormal coat conditions such as telogen defluxion of the postpartum female. While plucking the hairs, it is useful to note the ease with which this can be accomplished. Easily epilated hair indicates an endocrine abnormality. If it is difficult to epilate, especially in a partially alopecic area, suspect a severe pruritus and self-epilation.

The clinician must select the appropriate diagnostic tests and use them in an optimum sequence. For instance young animals presenting with focal alopecic areas must be scraped for mites and cultured (DTM) for ringworm on the first visit. It is essential to rule out these two diseases in neonates and juveniles as early as possible.

### **CULTURE ON DTM**

The accuracy of DTM is such that it is worth the delay while the sample is incubating. The culture must be dated, named, and examined daily. The time period for a colour change is important. Any changes in colour after 14 days are usually due to contamination by saprophytes. If there is any doubt, a small "flag" of cellotape attached to a forceps should be pressed onto the thallus and then placed on a drop of lactophenol cotton blue stain on a slide. Microscopic examination

reveals cigar-shaped macroauleurospores with ring worm.

### **BACTERIAL CULTURE**

Empirical selection of antibiotics is adequate in the initial therapy of pyoderma. The author reserves bacterial culture and sensitivity testing for non-responsive pyodermas, recurrent pyoderma, deep pyoderma and life- (or limb-) threatening pyoderma. Interpretation of sensitivity testing is necessary. A drug with which one is familiar, which is not too expensive and with a narrow spectrum of activity against the pathogen should be selected. In all cases, select a drug effective against coagulase-positive staphylococci.

### **BIOPSY**

Close clipping and disinfection of the biopsy site should be avoided as this affects the superficial layers of the specimen. Four and six mm diameter punches usually provide adequate tissue for the dermatohistopathologist. A local anaesthetic (0,5 ml) is injected subcutaneously. The punch is rotated until the dermis is penetrated and the specimen is removed and blotted dry. A single suture is usually sufficient to close the wound. A scalpel should be used to biopsy nasal, footpad, and interdigital areas. The resultant eliptical incision heals without puckering. Excisional biopsies must be performed if total removal of the lesion is required such as a small, suspected malignant nodule. Pustules and vesicles should be biopsied in toto. The histopathologist may also require some normal skin tissue at the edge of the lesion to assist in making a diagnosis. Incisional · biopsies, where only a piece of the lesion is removed, are necessary for cosmetic reasons or when the lesion is toc large to be totally excised.

Specimens for histopathology are fixed and transported in 10% neutral phosphate buffered formalin. Formalin is obtained as 40% formaldehyde, i.e. "concentrated" formalin. This must be diluted 1 in 9 to give 10% formalin and to every litre of this, 4g acid phosphate monohydrate and 6,5g anhydrous disodium phosphate should be added as buffer. Specimens for immunofluorescence (autoimmune diseases such as pemphigus) must be preserved in Michel's formula. Contact the histopathologist in this regard.

### LESION RECOGNITION

Lesions are described according to their distribution (localised, generalised, bilaterally symmetrical, asymmetrical, focal, multifocal), arrangement (discrete, confluent), depth (elevated, depressed), consistency (soft, fluctuant), quality (dry, moist, greasy), colour and size.

Primary and secondary lesions are described in Tables 4 and 5 respectively. Primary lesions develop spontaneously as a direct reflection of the disease. Secondary lesions either evolve from the primary lesions or are artifacts induced by the patient (self-trauma) or external factors (medications).

### **PRURITUS**

Pruritus is defined as an unpleasant sensation that provokes the desire to scratch. Physiological pruritus is moderate and transient and is the normal animal's reaction to stimuli such as humidity, dust, sand and occasional ectoparasites. Pathological pruritus is persistent and often leads to skin changes. Pruritus is likely to be a

Table 4: Primary lesions

Lesion	Description
macule	circumscribed area of pigmentary alteration which is neither elevated nor depressed
patch	as for macule but greater than 1 cm in diameter small, solid elevation of the skin
<sub>nodule</sub>	larger (greater than 1 cm in diameter) solid eleva- tion
plaque	solid elevation greater than 1 cm in diameter that is flat-topped
tumour	solid elevation that is neoplastic and involves or in- filtrates the dermis as well
cyst	soft, fluctuating epithelial-lined cavity containing fluid or a combination of viscid fluid and solids
pustule	small, circumscribed elevation containing purulent exudate
vesicle	small, sharply circumscribed elevation containing fluid
bulla	vesicle greater than 1 cm in diameter
wheal	sharply circumscribed, flat-topped elevation consisting of oedema

Table 5: Secondary lesions

Lesion	Description
scale	accumulation of loose fragments of the horny layer
crust	dried exudate on the surface of a lesion
scar	fibrous tissue that has replaced damaged tissue
excoriation	superficial excavations of epidermis caused by self-trauma
lichenification	thickening and hardening of the skin characterised by an exaggeration of the superficial skin markings
hyperpigmenta- tion	excessive pigmentation
hypopigmenta- tion	decreased pigmentation
fissures	linear cleavages of the skin
hyperkeratosis	increased thickness of the horny layer

presenting complaint in the majority of dermatologic cases and, if pathological, requires investigation. Elucidation of its multifactorial aetiology poses problems for the busy clinician. However the final outcome is more likely to be satisfactory if a detailed systematic approach is followed initially rather than administering purely symptomatic therapy. The history should include whether the pruritis or the lesions appeared first, whether the condition is seasonal or perennial, and whether any agent appears to exacerbate it. A plan such as that given in Table 6 can be followed. The conditions in each stage must be ruled out before proceeding to the next stage. In stage one, the condition will be recognised at clinical examination. Stage 2 would probably require more detailed examination such as screening tests. Allergy tests (Table 3) are often needed in pruritic cases. Intradermal allergy testing in suspected atopic patients requires expertise in performance and experience in interpreting the results. Allergens selected for that particular area are injected intradermally and the reaction measured and compared to negative and positive controls. This is best referred to a veterinarian who does it frequently.

Table 6: Stages in the diagnosis of the aetiology of pruritus

Stage 1	Stage 2	Stage 3	Stage 4
large parasites	microscopoic parasites	allergic diseases	idiopathic pruritus
trauma	bacterial disease	auto-immune disease	•
chemical irritant	sdiseases resembling infections		
incorrect diet anal sacculitis acute moist dermatitis acral lick granuloma	neoplasia		

Table 7: Alopecia: contributing factors

Endogenous Primary
hormonal
female endocrine imbalance (ovarian imbalance) hypothyroidism
hyperadrenocorticism
hyposomatotropism
male feminizing syndrome
telogen defluxion .
genetic
alopecia universalis
colour mutant alopecia

alopecia universalis colour mutant alopecia black hair follicular dermatitis

Secondary
inflammatory
allergic
secondary to disease
autoimmunity
pemphigus (foliaceous, vulgaris, erythematosis, vegetans)
bullous pemphigoid
systemic lupus erythematosis

Exogenous Nutritional

zinc responsive dermatosis

Biological parasites (Demodex, Sarcoptes, fleas) micro-organisms (bacteria, fungi, viruses)

Physical self-trauma, burns

Chemical caustic burns

latrogenic

glucocorticoids antimitotic drugs (cyclophosphamide, methotrexate)

parasitic, atopy, food, contact

Non-scarring cutaneous disease Alopecia areata Acanthosis nigricans Seborrhoea

Isolation from suspected allergens for 14 days followed by provocative exposure is less exacting and may even be done by the owner in pets suspected of having drug hypersensitivities or contact allergies. Dietary hypersensitivity testing involves feeding a selected diet for at least 21 days. If the pruritus ceases, a single new dietary component is added each week until the pruritus

returns. In this manner, careful management and record keeping can elicit the dietary allergen. Patch testing has fallen into disuse in the clinical field as the patient does not usually tolerate the patches and bandages.

### **ALOPECIA**

Alopecia is defined as partial or complete absence of hair in an area where hair is normally present. The aetiology (Table 7) can be either internal (endogenous) or external (exogenous). Although alopecia is readily noticed by owners, it is often the result of internal factors which are not readily apparent. Negative screening tests then result in further diagnostic tests (Table 3) being necessary.

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### **BOEKRESENSIES**

### THE MARE AND THE FOAL

Proceedings of the ninth Bain-Fallon Memorial Lectures,

August 6th - 9th, 1987

Australian Equine Veterinary Association, 1987 pp v and 190, numerous illustrations and tables. Price \$A50,00 or £22,00 (ISBN 0 959 3977 5 2). Available from Miss A.M. Best, P.O. Box 371, Artarmon, N.S.W. 2064, Australia.

This book represents a collection of papers on equine reproduction presented by three well-known authorities as part of the annual Bain-Fallon Memorial Lectures. These lectures are dedicated to the memory of Murray Bain and Peter Fallon for their contributions to stud practice in Australia.

The book is presented by the Australian Equine Veterinary Association as the Proceedings of the ninth meeting and is divided into three sections, each consisting of a number of short "chapters":

- 1. Hormonal mechanisms during oestrus and pregnancy.
- 2. Clinical examination of the reproductive tract in the mare; infertility and obstetrics.
- 3. The neonate.

In the first seven "chapters", WR 'Twink' Allen of the TBA Equine Fertility Unit deals with overviews of the hormonal mechanisms during the oestrus cycle including aspects of environmental stimulation and abnormalities of the cycle; control of the cycle with artificial lighting and hormonal therapy; reproductive efficiency in the United Kingdom; early pregnancy failure; pregnancy and pregnancy loss; progesterone assays; and parturition including induction of parturition. These "chapters" are of necessity concise but offer a wealth of information which is further augmented by the bibliographies at the end of each presentation. Both practitioner and postgraduate student will find reading this section fascinating.

The second section has been compiled by AC 'Woody' Asbury, University of Florida. It consists of eight presentations dealing with the clinical examination of the genital tract of the mare; endometrial biopsies; non-infectious infertility; infectious infertility and its immunological basis; the management of endometritis; abortion; obstetrics; and puerperal complications. There are a few shortcomings in this section but it nevertheless provides valuable information to the practitioner and student of equine reproduction.

In the last section, Anne M. Koterba of the University of Florida gives a clear concise review of the neonate and its problems. The section starts with the identification and management of the high risk foal and leads the reader through "chapters" on neonatal asphyxia, respiratory diseases, fluid therapy, nutritional support, neonatal infection, neonatal immunity, prematurity or immaturity, maladjustment syndrome, intestinal disorders and urogenital disorders. There is again a wealth of information available in the subsections of this section from which the practitioner and student will certainly benefit.

The book is highly recommended to equine practitioners, post-graduate students and lecturers. It is well presented, concise and clear and should be readily acceptable due to its handy format and size. I find the absence of an index unfortunate in an otherwise excellent publication.

H.M. Terblanche