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CONTENTS/INHOUD

Addresses **Voordragte**

Address to the South African Veterinary Association Congress – A.R.W. Porter 3
Current trends in veterinary education – N.C. Owen 7

Articles **Artikels**

Wider aspects of veterinary public health – M.M. Greathead 11
A retrospective study of 120 natural cases of canine ehrlichiosis – J. van Heerden 17
The use of doxycycline to control heartwater in sheep – A. Immelman and Gillian Dreyer 23
The treatment of *Urginea sanguinea* Schinz poisoning in sheep with activated charcoal and potassium chloride – J.P.J. Joubert and R. Anitra Schultz 25
The safety of injectable rafoxanide in cattle – J. Schröder 29
Genetic markers in South African Thoroughbred stallions – D.R. Osterhoff, Lynette le Grange and Michèle Robinson 33
Biochemical polymorphisms in the South African springbok (*Antidorcas marsupialis*) – D.R. Osterhoff, Sanita M. Schoeman and A. Conroy 37
Surgical approach to the rostral cranial fossa by radical transfrontal craniotomy in the dog – P.D. de Wet, I.I. Ali and D.N. Peters 40

Continued education **Voortgesette opleiding**

Reaction patterns in myocardium in response to injury – S.J. Newsholme 52

Case reports **Gevalverslae**

Unilateral orchitis in a bull caused by *Brucella abortus* biotype 1 – C.J. V. Trichard, S. Herr, S.S. Bastianello and D. Roux 60
Gross horn malformation in an African buffalo (*Syncerus caffer*) – V. de Vos 63
A case of dysuria as a result of a communication between the urinary bladder and corpus uteri in a Cairn Terrier – Selma J.E.M. van Schouwenburg and G.J. Louw 65

Short communication **Kort berig**

Absence of urea toxicity in young pigs – C. Button, J.P.J. Joubert and B.P. Maartens 67

To the Editor **Aan die Redaksie**

Paneth cells in the pig – a controversial issue 69

Association news **Verenigingsnuus**

Committee report: The performance of cosmetic surgery by veterinarians 72

Book reviews **Boekresensies**

Domestic Animal Behaviour – J.V. Craig 6
Genetics of Livestock Improvement – J.F. Lasley 9
Horses and Horse Sense: The Practical Science of Horse Husbandry – J. Blakely 16
Principles of Animal Virology – W.K. Joklik 24

Contents continued on page 1 *Inhoud vervolg op bladsy 1*

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Book reviews **Boekresensies**
Veterinary Aspects of Feline Behaviour – B. BEAVER 59
National Decision-making for Primary Health Care 62
Klinische Propädeuk der Inneren Krankheiten und Hautkrankheiten der Haustiere – W. JAKSCH UND E. GLAWISCHNIG 62
Bovine haematology – O.C. STRAUB 68
Essentials of Canine and Feline Electrocardiography – L.P. TILLEY 71

Abstracts **Uittreksels**
Levels of glucose, serum albumin and somatic cells before and during early stages of acute clinical mastitis artificially induced in cows by means of human strains of group-B streptococci administered intracisternally .. 22
Studies on schistosomiasis 22
Attenuation of turkey meningo-encephalitis virus 22
Identification of the serotype-specific and group-specific antigens of bluetongue virus 51
Rhipicephalus zambeziensis sp. nov., a new tick from eastern and southern Africa, together with a redescription of *R. appendiculatus* 64
An investigation into the toxic principle in eggs of the tick *Amblyomma hebraeum* 64

CORRIGENDUM

The author of the abstract "Sarcocystosis in domestic animals" which appeared in Vol. 52 No. 4 p. 350 of this Journal is M.B. Markus and not M.B. Marcus.

Index to Advertisers **Advertensie-Opgaaf**

Biolyte .....
Trivitrin en Tribrissen .....
Forray 65 .....
Frazon .....
Triatix LS .....
Clamoxyl .....
Curatik .....
Rintal .....
Bolfo .....
Milvet ..... Inside front cover
Coopers 10
Coopers 14
Beecham 31
Coopers 32
Beecham 70
Agricura 74
Bayer ..... Inside back cover
Bayer ..... Outside back cover

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ADDRESS **VOORDRAG**

ADDRESS TO THE SOUTH AFRICAN VETERINARY ASSOCIATION CONGRESS*

A.R.W. PORTER**

It is with some trepidation, Mr. President, that I venture to address an audience of veterinary surgeons and their supporters when I am a mere barrister – particularly since I am not on particularly familiar territory in so far as the problems, the fears and the aspirations of the profession in this country are concerned. Happily, however, I have travelled sufficiently widely to appreciate that I should therefore proceed with similar caution to the member of staff of one of our large London medical hospitals whose telephone rang in his duty office at one o'clock in the middle of one winter's night. "Hello, there" he said. "Ah," said the voice on the other end of the 'phone. "This is Dr. Smith. I am in some difficulty and I wonder if you can help. All the pharmacies are closed, and I have an urgent need of some beta-naphtha-oxy-ethanol with thial barbitone. Can you help me?" There was, at the other end, what is normally described as a pregnant pause and then the first voice spoke again. "When I told you hello" it said, "I told you all I know."

Maybe it would be safer if I contented myself with saying 'hello' and smiling sweetly at you all. The Japanese do, after all, say that the best way of saving face is to keep the bottom half shut – but that would seem a pity after coming all this way, and anyway who ever heard a barrister who could resist the opportunity of getting on his feet and talking?

In the course of our correspondence, Mr. President, you indicated to me that you were moving towards a new Veterinary Surgeons Act, and that current issues included animal welfare, or cruelty to animals and ethical issues such as cosmetic surgery – and I smiled because we too are trying to persuade our Government that a new Veterinary Surgeons Act is required, we too are heavily involved in animal welfare issues such as those relating to intensive farming, and the ethical issues concerning such procedures as the docking of dogs' tails seem for ever to be on the agenda. I saw, therefore, that I would at once be on the same wave-length as your audience if I were to speak to you of matters with which I was most familiar at home. Veterinary problems appear to be no respecter of international boundaries – or indeed of the passing years. When I take down from the shelves of our Royal College library a copy of the Veterinary Journal for 100 years ago, I very quickly discover that, in 1881, the agendas of the Council meetings bear a striking resemblance to those of today. For example, just as we have been struggling over the past 18 months to raise the money for an extension to the Royal College building, so I read in the correspondence columns of the Journal for a century ago how hard it was to raise just £5 000 from which "all the profession required could have been procured".

If I go even further back, those of you who consider that the use of acupucture in veterinary medicine is a new and as yet un-tested and unproven procedure, might be interested to read in the Veterinarian of 1828 an article on the uses of "acupuncturism in veterinary medicine"

But, returning to the volume of the Veterinary Journal for 1881, that edition was very concerned with the proposed Veterinary Surgeons Act, which was indeed passed in that year. Of course, the practice of veterinary medicine in the United Kingdom goes back much further than that. The farriers and the leeches had anticipated the creation of a separate body of veterinary surgeons and it was not until 1791 that the first veterinary school was established in England – the Veterinary College in London, followed some years later by William Dick's Veterinary College in Edinburgh.

In the early 1840's, those who had qualified at these Colleges decided to petition the Queen for the grant of a Royal Charter, with a view to seeking a declaration that veterinary surgery was a profession and only those holding recognised qualifications would be entitled, thereafter, to describe themselves as veterinary surgeons. This initiative meant a considerable sinking of differences between those who had qualified at the 2 Colleges – for there was great rivalry between them. The story goes that, in petitioning Queen Victoria, they searched for satisfactorily obsequious words. The first proposal of the draftsman, that they should begin by saying "Conscious as we are of our manifold deficiencies" was quite unacceptable to the proud alumni of the 2 Schools – but it is alleged that they were subsequently prepared to accept the amended version – "Conscious as we are of each other's manifold deficiencies."

However, joking apart, the sort of clarion call to which they all rallied was of the kind which Thomas Mayer sounded. He was one of the leaders of the profession who saw the urgency of unity and said in 1836 –

"I respectfully call upon veterinary surgeons everywhere, if they have the slightest love for their profession, if they value their own station in society, or have any regard for their brethren, to unite in forwarding by every means in their power, the real advancement of the veterinary art; and, in all her several departments, to raise her in the estimation of society by their conduct and respectability; and also to support, by their exertions, those who seek to carry out the real wishes of the profession. The labours of one or a few of us, highly talented though they be, and animated by a degree of zeal worthy of the cause in which they have embarked, will be of no avail unless they are supported by the well-thinking portion of the profession." These words still speak to us across the ages.

*Address delivered at the Biennial Congress of SAVA, Cape Town, September 1981.

**Secretary, Royal College of Veterinary Surgeons, 32 Belgrave Square, London, England, SW1X8QP.

Then in 1881, as I have said, the profession in the United Kingdom sought its first Veterinary Surgeons Act. At first there was some public and Parliamentary reluctance to agree to such an Act. After all, the profession had been practised for a century without such an Act and the Queen herself had given the Royal College its Charter. It was one of those difficult situations in which the profession does not control its own destiny. As a wise man once said – "There are 3 stages in the acceptance of a new idea. When it is announced, people say that it is not true" (i.e., that a new Act is required) "and a little later when its truth has been borne in on them, they say it is not important, and after they see that its importance has become sufficiently obvious, they say it is now new." However, eventually, Parliament was persuaded and the Act passed, providing statutory protection not only for the title of veterinary surgeon but, in the process, a measure of protection for members of the public and their animals against unqualified practitioners – and that has been the purpose of all subsequent Veterinary Surgeons Acts.

This being the centenary of the first Act, the Royal College held a reception in St. James's Palace, London, in February to mark the occasion. Magnificent State Rooms, all that was good in the way of food and wine and many distinguished persons present although, as I said to the Queen Mother, "I am no name-dropper" and therefore I will not list them.

What changes have we seen in the 100 years since the Act of 1881 was passed – not only in the profession in the United Kingdom but in the veterinary profession generally? Well, we have seen a growing awareness that statutes governing the practice of veterinary medicine have to be supported by Acts of other kinds – outlawing cruelty to animals, ensuring their anaesthetisation during operative procedures, safeguarding their welfare and protecting them when they are used for experimental purposes. In the early stages, much was learned from the practice of surgery and medicine upon human patients. Now the boot is often on the other foot, and veterinary research workers have things of value to say to their medical colleagues. While on the subject of anaesthetisation, you may be interested in the little exchange which is alleged to have taken place between the surgeon and the university professor when the latter, to his intense irritation was called in to hospital, in the middle of term, for an operation. After various vain attempts to talk his way out of it, or at least delay the operation till the vacation, the professor finally gave in – but with one last forlorn hope that the whole business might take less time than he feared, enquired of the surgeon "How long after the administration of the anaesthetic before I can hope to be talking and thinking rationally?" To which the reply was simply "That's asking a hell of a lot of the anaesthetic."

One of the major changes within the veterinary profession in Europe in recent years has followed upon the creation of the European Community – now consisting of 10 Member States – and the Community law which provides that citizens of any of those countries who hold a recognised veterinary qualification may practise as veterinary surgeons in any part of the Community. This law came into force only 9 months ago, and the movement of veterinary surgeons from one country to another has so far been very small. What, however, is much more important, is that the profession, years previously, anticipated this freedom of movement and the need therefore to work towards common standards of professional training, a common ethical approach to various problems and a common policy upon such issues as the use of lay assistants. This co-operation, fostered of necessity, has been immensely rewarding and we would not now be without it.

This does not mean that every member of the profession in the EEC is a pale imitation of his neighbour. Everyone maintains his or her national characteristics and as some wag observed, if you were to invite a representative of each of the countries to write a paper on some aspect of the elephant, you would very soon discover from which country the authors came.

The Frenchman would assuredly write about "The love life of the elephant", the Dutchman about "Tulips as an elephant feeding stuff", the Irishman about "Pink elephants I have seen", the Englishman about "Elephants and the Royal Wedding" and the German's title would be "The complete anatomy, physiology and psychology of the elephant in 17 volumes – with appendices". You may laugh, but in South Africa the paper would no doubt be entitled "Elephants as second row forwards".

Of course, as we enjoy this freedom of movement within the Community there has been no departure from the existing rule that veterinarians from South Africa, Australia, Canada and New Zealand can, generally speaking, come and practise in the UK. So each summer we see plenty of bronzed young men and women, with their veterinary parchments in their hands, on the doorstep of the Royal College first thing in the morning waiting to register. And until they speak you can't always tell which country they are from. It is fun guessing and there are some who say "You can always tell a South African, but you can't tell him much." (I only dare to say that because I have heard it said so often about the Poms in other parts of the world. When I went to New Zealand a few years ago their favourite riddle which they would inflict upon visitors from the UK was this – "Tell me how is it that India has leprosy and New Zealand has Poms?" – and the answer was obvious – "because India had first choice.")

All this movement of veterinarians between one country and another can do nothing but good, especially if they have the opportunity to do this when they are young, enthusiastic – and willing to learn. Sure, there is an exchange of knowledge and information on techniques, on research methods, on surgical procedures and so on. Even more important, however, is the opportunity to see how other nations live and to find out that the citizen of one country has very similar hopes and fears to this opposite number in another. Quite frankly, the possibility of Germans and Italians on the one side and the French, British and their allies on the other getting into the sort of armed conflict which engulfed the whole world twice in this century is now non-existent and, if that has been the real benefit of the EEC, one will put up gladly with some of the less sensible parts of the deal like the Common Agricultural Policy.

One is glad to know that when young veterinarians move around the world and are involuntarily their country's ambassadors, they are the cream of their generation. Highly intelligent and academically first class (or they would never have got into veterinary school in the first place), there is no evidence of which I am aware that they are, as a result, less practical, less motivated or poorer contributors to the sporting or the social scene. I

have known veterinary students in recent years who have played top class professional soccer, rowed in the Oxford/Cambridge boat race, ridden in equestrian events at national level and so on, and still have coped with a demanding course which, if we are honest, would have stretched their fathers and completely floored their grandfathers. So that is change, and that is progress.

In passing, may I say that one of the new generation of veterinary surgeons who made me feel particularly humble was a young man from Ethiopia who had left the country to study veterinary medicine in Thessalonika. He had to learn Greek to enable himself to study at all, but he passed his degree examination in that difficult language with flying colours. Then, being unable to return home, because he was out of sympathy with the present regime, he came to England and sat and passed our MRCVS examinations, in English, again at the first attempt.

All over the world, too, more women are being admitted to the profession and accepted as equals. It is a long cry from the year 1770 when the British Parliament, horrified by the increasing use of cosmetics by women, introduced a Bill which read – "All women of whatever age, profession or degree, whether virgins, maids or widows that shall . . . impose upon, seduce or betray into matrimony any of His Majesty's subjects by use of scents, paints, cosmetic washes, artificial teeth, false hair, Spanish wool, iron stays, hoops, high-heeled shoes or bolstered hips, shall incur the penalty of the law in force against witchcraft and like misdemeanours and the marriage, upon conviction, shall stand null and void."

Now we have women veterinary surgeons more than holding their own in every aspect of veterinary life – and at the Royal College in 1976 we had our first women President, Dr. Olga Uvarov. We receive many letters every week from little girls wishing to become veterinarians. I particularly liked the one which began –

"I am already extremely worried about my career, despite being only 13. My mother was unfortunately not the most successful pupil at school. So she has great plans for me to make up for her wasted education. I am fortunate enough to have inherited my father's brains and my mother's body. Please could you tell me what qualifications I will need to become a vet." (Since she is now 18, I plan to ring her up some time to see how she is doing.)

Of course, there have been advances too – particularly in recent years – in veterinary facilities, in equipment and diagnostic aids, in knowledge of intricate techniques. Veterinary surgeons have begun to specialise in a way comparable to the training undertaken by the members of the medical profession – no longer claiming to be de facto specialists in equine medicine, or small animal orthopaedics merely because they do nothing else. That, as someone observed, is not necessarily anything remotely like specialisation. It may just be a case of repeating the same mistakes year after year, so long as the client does not find out. It may indeed be the case that the man does not have a life time of experience but merely one year's experience, repeated over and over and over again.

One accepts that further education after one has qualified, and is doing a job, and has a family and a mortgage and commitments, is not easy. After all, I heard one University professor say of under-graduate education that it consisted of the inculcation of the incomprehensible into the indolent by the incompetent, and even if that is not true of veterinary students, it is a hard task to study when you are past 30 and doing a full-time job as well – but thank heaven there are those who are dedicated enough to do just that. So the professional bodies have to react to make such further education possible, to encourage it, to recognise it and reward it. So far in the Royal College we have 7 specialty boards, and we hope to establish more in due course.

This will be one of the means whereby the profession can maintain the respect of the animal owning public, of other professions and of itself. Let us be quite frank. We *all* care about our image and there is nothing wrong in that if this causes us to try harder all the time. The only wrong thing would be if we were to become obsessed by the show we put on in the shop window and become uncaring about the service provided in the workshop.

It is said of the great musician Berlioz when he was rehearsing an orchestra in one of his compositions that he rapped his baton on the stand, stopped the music and said with unutterable scorn – "You are playing like a lot of veterinarians." That's all right – there is no real insult to the profession there; they are not expected to be expert musicians. What they are expected to be is competent, caring and compassionate in the exercise of their own profession.

In the UK one of our members has been making a great reputation for himself as an author whose books have been filmed and presented with great success on television. His name is James Herriot – that's his pen name, not his real one. He writes mostly about the period immediately prior to the last war and that is a nostalgic era for many people. In the television series in particular, he comes across as someone who cares deeply not only about his patients, but also about the people who figure in his daily life, showing an innate courtesy at all times – and taking time and having the concern to communicate to people that he does feel for their problems. That is what made him, and still makes him a successful veterinary surgeon – in reality and not just in some romantic portrayal of him. For when we talk of his success as a veterinary surgeon, we don't mean his monetary success. That had to wait for his books. Success to him means the respect of the community in which he lives, and the peace that goes with the knowledge of a job well done. It is something which specialist training, and modern equipment and superb facilities will not be able to guarantee without the maintenance of the highest personal standards. So in the midst of a century of change, there are some things which it is important should remain unchanged.

In all this, what is the role of the professional bodies – my Royal College – your Association? There are some who say that our job is to remain in the background and that we will play our part the best if, like a good referee at a rugby match, we are hardly noticed. That we should not be like the little boy in his bath who discovered his tummy button and began to twiddle it. To his astonishment, he discovered that, as he carried on twiddling, it came out further, like a screw – and, finally, it came out all together. Deciding that he felt none the worse without it, he chucked it out of the window. He stood up, to get out of the bath, and his bottom fell off.

Now the moral of that story is said to be – don't fiddle with things unless you are absolutely sure how they will turn out. That's fine in theory – but it is also a

recipe for conservative medicine of the worst sort – 'the mixture as before' cough bottle – and for inaction and the stultification of thought and ambition.

The story which I prefer to the tale of the screw in the bath is the story of the little man who went into the opticians in Belfast and said that he wanted his eyes tested. "Ah yes", said the optician. "Have your eyes previously been checked?" "No", he replied: "They've always been blue." "Put your left hand over your right eye", sighed the optician. Left hand went over left eye. "No", said the optician. "Left hand – *right* eye." This time it was right hand over right eye. The optician sighed again and went to the back of the shop. He took a cardboard box, cut two eye holes in it and – the better to control proceedings himself – placed it over the wee fellow's head. Whereupon he heard crying from inside the box. "What are you crying for?" he enquired. "Oh", sobed the wee man, "I wanted a horn-rimmed pair like me brother had."

The Lord preserve us from always wanting to have what our brothers – and our forefathers have had before us. We must avoid being like the man in the train with his back to the engine, who never sees anything until he has gone past it. God grant us the ambition to - *want* to improve, to do better, to make progress. Not for purely selfish reasons but because as Thomas Mayer whose words, spoken in 1836, I have already quoted, went on to say – in his call for action by the profession –

> "If, in after-times, some one more capable than myself of handling this subject should take it up, my earnest desire is, that they may be able to say of the present race of veterinary surgeons, that their labours made the dark periods of our profession more visible and added lustre to the light."

That, Mr. President, would seem to be a worthy ambition for us all.

BOOK REVIEW — BOEKRESENSIE

DOMESTIC ANIMAL BEHAVIOUR

JAMES V. CRAIG

Prentice-Hall, Inc., Englewood Cliffs, New Jersey 07632. 1981 pp xvii 346 Figs 68 Tabs 76 Published price R26,55
(ISBN 0-13-218339-0)

The study of animal behaviour, termed Ethology, is a "new" discovered field of animal production and care. The demand for more scientific understanding, has been created by our human population and has produced profound changes in the environment. This becomes distinct in the international literature scene in which an increasing amount of scientific information is being released. A most recent example is: "Domestic Animal Behaviour" of James Craig who skillfully handles an enormous amount of recent and previously available information.

The author describes in a pleasant way and with the excellent use of the English language, the causes and practical implications of animal behaviour.

Instead of monotonously dealing with pure science from one species to the other, this book is organized by topics rather than by comprehensive discussions.

An interesting entrée on natural selection, domestication and senses of communication is followed by an elaboration on the basically founded genetics of behaviour. The controversial question of instinct and behaviour is dealt with.

The importance of the knowledge of socialization and association on adoption and fostering is plainly explained through quotations of numerous trials and observations. The importance of critical periods in farm and pet animals in relation to handling is stressed. In the final chapters the sexual behaviour of the male and female animal is discussed.

A book of this nature would not be complete if psychological stress were not brought into perspective in relation to welfare, handling, restraint and handling facilities of which plans and photographs are displayed.

Animal behaviour is approached from both theoretical and practical viewpoints which should be useful for students of agriculture and veterinary science, for breeders and producers, for those with companion animals and for any one with a lively curiosity about animal behaviour.

E.C. Genis

ADDRESS **VOORDRAG**

CURRENT TRENDS IN VETERINARY EDUCATION*

N.C. OWEN**

It was my privilege to visit a fairly large number of veterinary faculties in several Western countries during the latter part of 1980. This, together with previous experience in veterinary and dental education, including a brief visit to the new medical schools at Nottingham and Southampton in the United Kingdom, have I believe given me a somewhat unique insight into veterinary educational trends *vis-a-vis* those which are current in our sister professions.

While there are obviously distinct differences between the educational objectives of the various health science professions, the developmental trends are remarkably similar. This suggests that the educational problems faced by these professions are perhaps more common than is generally acknowledged.

During my recent trip I was fortunate to visit several veterinary faculties in the United States of America and Great Britain, a few in Brazil and West Germany and one in each of Canada and the Netherlands. My overall impression is one of considerable variation in physical facilities, equipment and curricula between the various countries and occasionally within the same country. Thus, while the clinical facilities in North America are of a particularly high and uniform standard, I was struck by the paucity of attention given to animal production subjects in their veterinary programmes. The requirements in this regard must be met prior to entry to the veterinary college and tend to vary considerably. In fact, it would seem that some candidates may gain entry to certain colleges with virtually no exposure to animal production tuition.

The British veterinary programmes tend to be more comprehensive in this regard, but generally have the disadvantage of fragmented physical teaching facilities. Several of the faculties in the United Kingdom have to make use of outlying field stations for their large animal instructional purposes. I do not wish to imply that the use of satellite facilities is necessarily a disadvantage in veterinary education, in fact the contrary is probably true, especially when it comes to exposure to clinical material not always available at the veterinary hospital. However, where outlying facilities affect large numbers of staff and students, the dichotomy presents organisational and communicational problems.

We in South Africa are indeed fortunate to have the opportunity to develop facilities as comprehensive entities.

*Address delivered at the Biennial Congress of SAVA, Cape Town, September 1981.

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FOOD ANIMAL VETERINARY MEDICINE

In recent times increased emphasis has been placed on so-called "food animal veterinary medicine" in the USA. This I believe, has, at least partially, been stimulated by Federal and State Government funding programmes in that country. It is, however, difficult to assess whether this trend reflects an actual demand for increased food animal veterinary services in response to supply and demand forces[1].

EQUINE VETERINARY MEDICINE

An interesting observation is the resurgence of equine veterinary medicine as evidenced by the large number of horses being treated in veterinary hospitals generally. This trend can apparently be related to the increasing suburbanisation and affluence of the society[1]. It is also in keeping with the predictions made by the Committee on Veterinary Medical Research and Education in 1972 that the horse population would increase 4-fold between 1960 and 1980 in the USA[2]. A similar re-emergence of the horse as an important educational determinant has occurred in South Africa insofar as the demands for veterinary services by the affluent section of the community are concerned.

CURRICULAR REFORM

Perhaps the most striking educational trend encountered by me, was that of veterinary curricular reform which was especially evident in the USA. Educational reform in many spheres has characterised the 1970's and has been ascribed in part to the campus unrest of the turbulent "publish or perish" era of the 1960's[5]. It is also noteworthy that in West Germany the traditional rigidly organised faculty structures have been relaxed to accommodate reform. After much experimentation, it would seem that most veterinary colleges in the USA now offer what is called a "core-elective" curriculum consisting of compulsory subjects common to all students ("core") with a variable number of optional subjects ("electives") to cater for individual student preferences. My impression, however, is that in practise the electives have been limited to an extent where they are insignificant from an undergraduate specialisation point of view.

Integrated teaching

It is generally accepted that educational reform in the health sciences has resulted from the ever expanding tendencies of individual subjects to accommodate new "essential" material in their courses. It is this "subject

imperialism" which has placed the orthodox disciplinary curriculum under considerable strain and has given rise to a number of curricular innovations. The most significant innovation has been the introduction of integrated curricula consisting of courses based on the organ systems rather than on classical disciplines. This approach necessitates interdepartment teaching where each organ system would constitute a single course covering the anatomical, physiological, pathological, diagnostic, therapeutic (including surgical) and preventive aspects of each disease entity. The design and presentation of these courses cut across classical departmental divisions and are usually supervised by interdepartmental committees.

As it is my belief that this trend in veterinary education may be traced back to a similar trend in medical education and may therefore be of interest, allow me to briefly review these educational developments in the latter profession.

Integrated teaching by organ system appears to have originated in medical education at Western Reserve University in the USA during the 1950's. At about the same time the General Medical Council adopted a more permissive attitude regarding central control of medical education in the United Kingdom. This in turn resulted in a spate of medical curricular experiments and the adoption of various integrated forms of course design in that country[4].

A similar evolutionary pattern has emerged in medical education in South Africa since the adoption of less monolithic curricular regulations by the South African Medical and Dental Council in 1976[3]. In keeping with the intentions of these more flexible regulations, medical curricula are now less stereotype and several faculties in this country offer more integrated forms of teaching.

In the USA where education is less subject to central control, several veterinary colleges have experimented with integrated teaching programmes. The degree of success of these programmes would appear to be highly variable and in some instances a tendency to revert back towards a more conventional (orthodox) approach was evident.

The potential advantages of integrated undergraduate teaching are obvious, but numerous practical difficulties arise with the implimentation thereof. These difficulties are mainly of an organisational nature and it can be said that the systems-integrated approach has a high tendency toward entropy, i.e. it requires a constant input of energy to retain the highly organised state necessary for continued success. Indeed, the success of this approach is vitally dependent on the continued enthusiasm of the participants. Too often the programme tends to collapse as the originators move on to other appointments and are replaced by less committed staff members.

Co-ordinated Teaching

Another attempt at reform which is current in veterinary and medical education, is that of co-ordinated teaching. This approach should be clearly distinguished from the "integration by organ system" approach mentioned earlier.

In co-ordinated teaching the conventional departments remain responsible for their respective courses, but by interdepartmental co-operation an attempt is made to deal with related topics in the various courses at a given time. This approach is less demanding from an organisational point of view and therefore probably more stable in the longer term.

It is my belief that veterinary educational reform in this country could initially be sought along these lines. At this point, allow me to mention in all fairness to veterinary education in South Africa, that both integrated and co-ordinated teaching have received some attention and shall no doubt continue to do so. In fact, subjects such as "medicine", "pathology" and "promotive veterinary health" represent attempts at integrated teaching across departmental barriers. The dangers inherent in adopting a hybridisation of both the "orthodox" and the "integrative" approaches are unnecessary repetition and conflicting factual interpretation between clinicians and non-clinicians and concomitant overloading of the syllabi.

PREVENTIVE VETERINARY MEDICINE

Preventive veterinary medicine continues to receive increasing emphasis in veterinary education with the establishment of separate departments to accommodate this discipline in certain faculties. This trend represents an attempt to emphasise the maintenance of animal health as opposed to currative medicine and may be related to the concern of funding and educational authorities for the decline in rural veterinary services in most countries.

Preventive veterinary medicine as a separate entity aims to bring together all those aspects of disease prevention previously scattered amongst a host of other courses. As such this subject may lend itself admirably to a multi-disciplinary presentation.

SEEING PRACTICE ("PRECEPTORSHIP")

The concept that undergraduate students be required to gain limited clinical experience under the supervision of private practitioners has long been in existence both in South Africa and in Britain ("seeing practice").

Similar requirements have recently been introduced into the curricula at several veterinary colleges in the USA ("preceptorship"). An interesting innovation is the appointment of practitioners as part-time staff of the college to involve them more directly in the educational process. In one instance preceptorship has, however, been discontinued because of possible legal implications.

POSTGRADUATE EDUCATION

It is my impression that more attention is being given to the introduction of veterinary specialisation degree courses in both the USA and the United Kingdom. Although a variety of diploma and degree courses exist in these countress, Williams (1975) stated in reference to the situation in the USA that "the rapid growth of veterinary knowledge has created a demand for postgraduate education facilities in addition to the MSc and PhD degree courses which are mostly research orientated"[5].

In this regard, I believe that our M.Med.Vet. programme of the University of Pretoria which was started in the 1960's, is in the forefront of these developments.

CONTINUING EDUCATION

The realisation that it is no longer possible to qualify a veterinarian for life at the undergraduate level has established a definite role for veterinary faculties in the continuing education of graduates. The increasing involvement of a number of faculties in this sphere was evident, however, until such time as the funding authorities accept continuing education as a function of the university, the contribution made by faculties must remain *ad hoc* and sporadic.

In conclusion, it may be said that veterinary education is at present characterised by a lack of conformity as regards both curricular design and faculty organisation. It is perhaps in this diversity that our future strength is to be found.

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BOOK REVIEW — BOEKRESENSIE

GENETICS OF LIVESTOCK IMPROVEMENT

JOHN F. LASLEY

3rd Edn. Prentice-Hall Inc., Englewood Cliffs, New Jersey 07632 ISBN 0-13-351106-5; pp xvi & 492 Figs 82, Publ. Price R29,25

Established principles of animal breeding do not change over a period of years. Information about these principles, and methods of presenting and using them do change, however, with the publication of more research results and with the experience of the teacher, breeder, and author. In the 3rd edition the author presented the principles of animal breeding as simple and practical as possible for the student and the breeder of farm animals.

In the first chapters of the book the foundation for animal breeding is laid by presenting some of the fundamental concepts of animal genetics: chromosomes, abnormalities of chromosomes, the genes, their function and role in animal genetics, gene mutations and lethal genes, the phenotypic expression of genes and the concept of gene frequencies. Chapters dealing with the variations in the economic traits in farm animals, principles of selection, inbreeding, linebreeding, outcrossing and crossbreeding are included to present some of the basic principles involved in the improvement of quantitative traits.

A new chapter on "Genetic Resistance to Disease and Parasites" has been added and should be of greatest interest to Veterinarians. The last chapters of the book show how the fundamental principles of qualitative and quantitative genetics may be applied to the improvement of pigs, beef cattle, sheep, dairy cattle and horses.

At the end of each chapter questions and problems are brought forward to reinforce the reader's knowledge and in some cases the answers are provided.

An appendix has been added which includes more technical information on statistical procedures, the calculation of selection indices for those desiring more advanced information.

A glossary of genetic terms has also been included at the back of the book which is certainly very useful.

Genetics is rather difficult for students of Veterinary Science and Agriculture to understand, but this book helps to overcome the antagonism towards the subject and opens new avenues to really appreciate this comparable new branch of science.

D.R. Osterhoff

WIDER ASPECTS OF VETERINARY PUBLIC HEALTH

M.M. GREATHEAD*

ABSTRACT: Greathead M.M. **Wider aspects of veterinary public health.** *Journal of the South African Veterinary Association* (1982) **53** No. 1, 11-16 (En) City Health Department, P.O. Box 1477, 2000 Johannesburg, Republic of South Africa.

The field of public health work covered by the Veterinary Branch of the Johannesburg City Health Department is described. This comprises meat hygine, milk hygiene and the investigation of zoonoses. Meat hygiene embraces hygienic control of wholesale meat premises and inspection of meat imported into the municipal area. Results of the bacterial monitoring of premises and the mass of meat inspected and condemned are given. Inspection of dairy herds, diseases monitored by laboratory tests and other aspects of milk hygiene are discussed. Zoonosis control includes the investigation of unusual animal behaviour and mortality. In carrying out this work the Branch co-operates closely with the Laboratory and Technical Services and Environmental Health Branches of the City Health Department.

Key words: Veterinary public health,.meat hygiene, milk hygiene, zoonoses.

INTRODUCTION

Veterinary Public Health has been defined as a component of public health activity devoted to the application of professional veterinary skills, knowledge and resources to the protection and improvement of human health. Veterinary public health activities involve a diverse range of functions. These reflect the broad community of interests between veterinary and human medicine and indicate opportunities for profitable interaction[8].

In South Africa veterinary public health in the municipal field has been associated with the veterinarian's role in abattoir meat hygiene and the control of milk supplies, but in Johannesburg, which now has no municipal abattoir, the Veterinary Branch of the City Health Department works in the following spheres:

(a) Meat hygiene from the abattoir gate, through wholesalers to deliveries to retail butchers. This includes the inspection of meat and offal imported into the municipal area from other towns and countries.
(b) Milk hygiene from the cow to the processing depots.
(c) Co-operation with the Environmental Health Branch of the City Health Department on dairy farms and at retail meat and milk outlets when necessary.
(d) Investigation of zoonoses and other aspects of general animal health in the city.
(e) Participation in amendment and revision of relevant legislation.

VETERINARY BRANCH FUNCTIONS

Meat Hygiene

A number of different activities fall under this heading:

(a) *Abattoir environs*

Although the City Council has no responsibilities in the Johannesburg Abattoir, the operation of this complex has an impact on the surrounding municipal area with periodic complaints from adjacent properties and townships regarding smells and flies.

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(b) *Approval of building plans*

The City Deep township, in which the abattoir is situated, was planned for the development of industries allied to the meat trade. The larger stands adjacent to the abattoir have been taken up by meat wholesalers and new premises have been erected by 3 large undertakings. The Veterinary Branch plays an integral role in the Council's approval of plans for such new premises as well as plans for the modification of older establishments in other areas.

(c) *Licensing of premises*

When new wholesale premises are taken into occupation or when new wholesalers start operating they must comply with health requirements for licensing. Existing premises may be inadequate or deteriorate necessitating a review of the current licence. In recent years there has been a tendency for retailers to extend their activities into wholesaling which reqires increased refrigeration capacity, loading and offloading facilities, vehicle washing bays and a new licence.

(d) *Hygiene control of wholesale premises*

The Veterinary Branch controls the hygiene at wholesale meat premises in the city and at the offal pool associated with the abattoir. The wholesalers situated in old buildings which in some cases are structurally inadequate for modern hygienic practices, require constant supervision. The new premises which have recently been built have been designed to accommodate modern methods and have up-to-date refrigeration facilities.

These premises are controlled by regular inspections which may reveal defects in equipment, working methods and management. In old buildings good housekeeping and progressive management is essential to maintain a high level of hygiene. Particularly in new premises the management is encouraged to appoint a hygiene officer to train staff, supervise operational procedures and control the cleaning of the plant at the end of the day.

The overnight cleaning of equipment is monitored by bacteriological swabbing before work starts in the morning. It reveals the effectiveness of cleaning which has been shown to be related to the frequency of inspection and swabbing. The procedure used was described by Meara et al.[3] and their evaluation of laboratory results as given in Table 1 is applied.

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If all items swabbed at one time have counts of <100 the overall rating of the premises is 100 %.

Results recorded over an 18 month period are presented in Table 2.

More than 88 % of premises swabbed were rated as satisfactory. Effective application of efficient sanitizers is essential in the attainment of low bacterial counts.

Another swabbing procedure carried out during work is used to assess the bacterial load and identify indicator or pathogenic organisms with which the equipment may have been contaminated. Together with the Standard Plate Count for total bacteria present the following organisms are looked for on the swabs by the cultural methods used by Meara et al.[3]

coliforms
Escherichia coli
Salmonella spp.
Staphylococcus aureus
Clostridium spp.

In addition *Bacillus cereus* is cultured on egg agar at 37 °C for 48 hours and typical colonies are confirmed with Gram's stain.

Results recorded over a 12 month period are given in Table 3.

TABLE 1: EVALUATION OF LABORATORY RESULTS

Bacterial Count/25 cm²	Item Rating	Overall Rating	Remarks
< 100	6	76 – 100%	Very Good (V G)
101 – 500	5	61 – 75%	Good (G)
501 – 1 250	4	46 – 60%	Satisfactory (S)
1 251 – 2 500	3	31 – 45%	Fair (F)
2 501 – 5 000	2	16 – 30%	Poor (P)
5 001 – 10 000	1	1 – 15%	Excessive (E)
> 10 000	0	0	Unacceptable (U)

TABLE 2: EFFICACY OF CLEANING AT WHOLESALE PREMISES

No of Premises Swabbed	Overall Rating (Symbol)*						
	VG	G	S	F	P	E	U
90	47 (52,2%)	24 (26,7%)	9 (10,0%)	5 (5,55%)	5 (5,55%)	–	–

*For meaning of symbols, VG etc, see Table 1

TABLE 3: TWELVE MONTH BACTERIAL CULTURES FROM EQUIPMENT DURING WORK

Month	No. of Swabs	Standard* Plate Count	*B. cereus*	Coliforms	*E. coli*	*Salmonella*	*S. aureus*	Clostridia
July	132	54 (40,9%)	8 (6,1%)	66 (50,0%)	57 (43,2%)	0 (–)	8 (6,1%)	0 (–)
August	90	14 (15,5%)	11 (12,2%)	19 (21,1%)	11 (12,2%)	**2 (2,2%)	5 (5,5%)	4 (4,4%)
September	90	49 (54,4%)	3 (3,3%)	35 (38,9%)	17 (18,9%)	0 (–)	7 (7,8%)	1 (1,1%)
October	140	78 (55,7%)	13 (9,3%)	56 (40,0%)	53 (37,8%)	†2 (1,4%)	18 (12,8%)	0 (–)
November	120	65 (54,2%)	6 (5,0%)	72 (60,0%)	46 (38,3%)	0 (–)	47 (39,2%)	0 (–)
December	–	–	–	–	–	–	–	–
January	120	79 (65,8%)	0 (–)	68 (56,7%)	54 (45,0%)	0 (–)	20 (16,7%)	0 (–)
February	119	104 (87,4%)	0 (–)	82 (68,9%)	75 (63,0%)	0 (–)	57 (47,9%)	0 (–)
March	120	104 (83,3%)	0 (–)	77 (64,2%)	77 (64,2%)	0 (–)	34 (28,3%)	0 (–)
April	120	54 (45,0%)	1 (0,8%)	43 (35,8%)	14 (11,7%)	0 (–)	36 (30,0%)	0 (–)
May	60	26 (43,3%)	2 (3,3%)	30 (50,0%)	30 (50,0%)	0 (–)	13 (21,7%)	0 (–)
June	60	22 (36,7%)	5 (8,3%)	3 (5,0%)	3 (5,0%)	0 (–)	5 (8,3%)	0 (–)
Total	1 171	645 (55,1%)	49 (4,2%)	551 (47,0%)	437 (37,3%)	4 (0,3%)	250 (21,3%)	5 (0,4%)

*Counts $>10^4$ organisms
**S. arizoniae* and an untyped *Salmonella* species
†S. subgenus II

Few *Clostridium* and *Salmonella* spp. were isolated: the latter were found at 3 different premises.

Standard plate counts of >10 000 organisms and the number of isolations of coliforms, *E. coli* and *S. aureus* show a definite seasonal variation with a peak in the summer months. This confirms the desirability of having a controlled temperature in meat handling and cutting areas of the building.

(e) *Imported meat*

Although the Johannesburg Abattoir is one of the largest in the country it has no facilities for slaughtering pigs and it is unable to supply all the other meat needs of the city. All pork and large quantities of other meat and offal are imported daily. The types and quantities of meat brought into Johannesburg from other South African abattoirs (excluding those in the Meat Board's Witwatersrand and Pretoria Controlled Marketing Area), neighbouring countries and from overseas during 1980 are given in Table 4[5].

TABLE 4: TYPE AND MASS OF IMPORTED MEAT (1980)

Type	Mass (Tons)
Beef	9 490
Mutton	884
Pork	4 904
Veal	15
Fat	278
Offal	1 062
Total	16 633

The City Council's Meat By-laws require such imported meat to be presented for inspection before distribution and sale. The Meat Board authorises introduction of meat from other areas and consignments are accompanied by identifying documents. The inspection is designed to assess the hygiene of transportation and the state of the meat on arrival, not the efficiency of the primary meat inspection though this is noted and if

routine cuts have not been made or if pathological lesions are found, appropriate action is taken.

The following aspects are checked:

(a) Appearance of the meat.
(b) Evidence of spoilage or decomposition.
(c) Refrigeration (bone temperatures in chilled meat and the state of frozen material).
(d) Evidence of soiling (during slaughter, loading or transportation).
(e) Efficiency of loading (chilled carcases must be suspended; no overloading).
(f) Cleanliness of the vehicle.
(g) Compliance with other aspects of the Meat By-laws and Hygiene Act Regulations (vehicle construction, maintenance and identification; efficient primary inspection).

Johannesburg has been criticised in the past for insisting on re-inspection of meat brought into the municipal area, but the volume of material condemned as unfit for human consumption, given in Table 5, demonstrates the need for this control.

TABLE 5: TONNAGE OF MEAT AND OFFAL IMPORTED AND CONDEMNED

Year	Imported Meat and Offal	Condemned Meat and Offal
1979	12 182	17,72 (0,14%)
1980	16 633	36,30 (0,22%)

Reasons for condemnation were:

(a) Decomposition due to loading inadequately chilled material, overloading of the vehicle, breakdown of refrigeration en route or transportation delays.
(b) Soiling.
(c) Pathological lesions not removed at primary inspection (abscesses, arthritic joints, etc.)

It is important that the transporting vehicle – rail or road – be opened by the veterinarian or health inspector so that at the time of arrival the physical appearance of the meat can be seen, the temperature measured, the smell of the newly opened truck noted, the presence of drip on the floor and loading, spacing or possible overloading checked before the meat is removed. With high transport costs operators are tempted to use money saving methods of overloading and stacking which do result in deterioration and loss of the product.

Steps taken to correct deficiencies are:

(a) Condemnation of defective material.
(b) Prosecution of offenders.
(c) Education of exporters, importers and transporters.

Milk Hygiene

Control of farm milk hygiene is carried out by inspection of dairy herds, premises and equipment linked with observation of milking methods and backed by laboratory tests on milk samples drawn at the point of production.

Similarly, control of processing and distribution is done by inspection with laboratory support.

(a) *Laboratory control of milk*

In the City Council's laboratory complex routine tests are carried out and special investigations are undertaken.

In the veterinary laboratory the following examinations are done:

(i) Tuberculosis – biological test on herd and individual quarter milk samples
– microscopic examination of quarter samples
(ii) Brucellosis – milk ring test (MRT) screening of herd samples and serum agglutination tests on biologically tested MRT positive samples
(iii) Mastitis – somatic cell counts on herd milk samples
– Identification of pathogens and antibiograms on selected quarter samples from cows in problem herds
(iv) Thermo-resistant inhibitory substances – disc assay tests on 'Thermocult' plates[2]

In the microbiological laboratory the following tests are done:

(i) Raw milk – Standard agar plate count
– Modified Eijkman test
– Butterfat and freezing point determinations
(ii) Pasteurised milk – phosphatase test
– methylene blue reduction test
– standard agar plate count
– coliform test

In the chemical laboratory raw herd milk, pasteurised milk, fodder and feed concentrates have been surveyed for pesticide residues. The intermittent presence of dieldrin levels higher than those specified in the Regulations under the Foodstuffs, Cosmetics and Disinfectants Act[6] in raw and pasteurised milk requires further investigation, considering that the use of dieldrin has been restricted for some years. Final prohibition of its use with effect from 1 January 1982 has recently been promulgated[7]. The problem cannot be easily resolved when fodder and concentrates containing unknown quantities of pesticides are freely fed to dairy cows.

The use of a computer greatly facilitates the keeping of laboratory records and each producer's history can easily be extracted for review.

(b) *Inspection of dairy herds and disease control*

There are more than 700 producers in possession of Johannesburg milk control permits. Their herds are situated throughout the Southern Transvaal and Northern Orange Free State and are subject to inspection by veterinarians. Their premises are visited by dairy inspectors. Particular attention is paid by the veterinarians to milk-borne zoonoses and udder disease, while advice is given on other aspects of herd health.

(i) Occasional outbreaks of anthrax occur on dairy farms.
(ii) Although the Dairy Control Board requires registered producers to have their herds tested for tuberculosis, cases of udder tuberculosis causing contamination of milk supplies are still traced by biological testing of herd milk samples. A number of these have been in cows with negative skin tests, thus underlining the continued importance of the milk biological test.

(iii) The incidence of brucellosis in dairy herds as demonstrated by positive guinea-pig serum agglutination tests following biological testing of the milk was as high as 20,3 % of samples tested in 1972[4], but declined to 10,7 % in 1980. The latter figure represented 166 actual dairy herds. With the knowledge of the Division of Veterinary Services these farmers have been given notice to remove all infected animals from their herds by the end of 1982.

(iv) Mastitis as revealed by high somatic cell counts in herd milk supplies remains a serious problem in herds producing fresh milk. In 1980 14,7 % of samples tested gave counts in excess of 1 000 000 cells per ml on the Coulter Counter[5]. This is higher than than the true percentage of high cell count herds because this type of herd was tested more frequently than herds with lower cell counts. Attention is paid to predisposing causes of mastitis, in particular the milking hygiene and correct functioning of the milking machines. Use of a vacuum recorder enables the pulsation and efficiency of a machine to be rapidly checked and the farmer advised to have a defective system serviced. All producers are supplied with the cell counts of the samples taken from their herd milk supplies. An owner of a herd with repeated high cell counts is advised to take steps to reduce these and if he does not, his control permit is suspended.

Examination of milk samples from cows with sub-clinical mastitis for identification of the causative organisms and their resistance to antibiotics is done.

(v) Antibiotic contamination of milk supplies continues to be a problem in spite of the introduction of dye-marked intramammary infusions in 1976. During 1980 0,8 % of 2 517 samples of herd milk was found to contain thermo-resistant inhibitory substances. Of 999 samples of bulked tanker milk 1,9 % showed the presence of penicillin, as did 4,0 % of the 273 samples of pasteurised milk tested[5]. A contaminated farm milk supply is suspended until it is known to be free from antibiotics but a batch of affected pasteurised milk cannot be recalled once it has been distributed.

Co-operation with the Environmental Health Branch

The Veterinary Branch maintains contact with the inspectors in the Environmental Health Branch in respect of milking and milk handling hygiene on dairy farms and also with regard to problems which may arise along the distribution chain at food factories and in retail meat and milk outlets. The latter Branch is responsible for hygiene control of these premises and veterinary assistance or advice is available when required.

Investigation of Zoonoses

On the agricultural plots and farming areas within the Johannesburg municipal area there are numerous large domestic animals while throughout the municipality a large number of dogs, cats and birds are kept as pets. In addition there are many free-living animals such as cats, pigeons, wild small carnivores and rodents. Consequently the possibility of zoonoses transmission is very real. Cases of unusual animal behaviour, unexplained deaths and the appearance of wild animals such as mongooses and meercats in residential areas are often reported to the City Health Department.

(a) *Rabies*

The wide publicity given to the Natal rabies outbreak produced much concern in Johannesburg in 1980 with many cases of dog and cat bites, and aggressive and abnormal behaviour being reported. Such cases were investigated with the assistance of the State Veterinarian and private practitioners. Exposed persons considered to be at risk were referred to the District Surgeon for medical attention. The public was advised to have their pets immunised against rabies and consideration was given to making immunisation compulsory in the municipal area. The municipal veterinarians and numerous other Council employees considered potentially at risk were immunised against rabies. One case of rabies was confirmed in a heifer in the Council's beef herd. The source of infection was never confirmed but was assumed to be from the meercat population on the farm. The State Veterinarian instituted control measures immunising the surrounding dog population and gassing meercat colonies. On a number of occasions the public reported the presence of mongooses on their properties with some being killed by dogs. No rabies was diagnosed in any of the brains submitted for examination.

(b) *Toxocariasis*

This condition and other dog helminths are receiving greater attention from public health authorities and the Veterinary Branch is instituting an investigation into the incidence of ascarid and other zoonotic internal parasites of dogs in the municipal area.

(c) *Pigeons*

Pigeons are sometimes the cause for annoyance and complaint from the public and a potential source of ornithosis. Abnormal behaviour and several deaths were reported amongst pigeons in a central city park. Investigation into a possible public health hazard was conducted and examination of live and dead birds with tests on their crop contents established that they had been poisoned with parathion in grain. This chemical was also found in the water circulating in the fountain displays.

Legislation

The most important legislation under which the Veterinary Branch works is:

The Animal Slaughter, Meat and Animal Products Hygiene Act No. 87 of 1967 and Regulations
The Health Act No. 63 of 1977
The Foodstuffs, Cosmetics and Disinfectants Act No. 54 of 1972 and Regulations
The Transvaal Local Government Ordinance No. 17 of 1939
The Transvaal Licences Ordinance No. 19 of 1974
Johannesburg City Council Public Health By-laws

Public Health By-laws

Draft new or amended Regulations under the parliamentary Acts are periodically published in the Government Gazette for general comment but amendment and revision of municipal by-laws originates within the local authority.

The municipal veterinarian is concerned with the application of the Meat, Milk, Food-handling and Keeping of Animals By-laws. New developments and changing circumstances in these and other areas of public health occur continuously, requiring changes to the legislation. The veterinarian must be in a position to initiate or assist with this task.

In 1979 Johannesburg had new Meat By-laws promulgated[1].

A revised and comprehensive set of by-laws governing the keeping of animals and the control of pet shops is before the City's legal advisers. It is also obvious that the Standard Milk By-laws of the Transvaal require review.

CONCLUSION

In South Africa veterinarians are employed full-time in some large municipalities only and where there is considered to be sufficient work in smaller ones, on a part-time or consultancy basis. There is no doubt that the veterinarian has a place to fill in the public health team other than as an abattoir meat hygiene officer, but in spite of this the number of veterinarians in full-time municipal employment has declined in the past decade.

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BOOK REVIEW — BOEKRESENSIE

HORSES AND HORSE SENSE
THE PRACTICAL SCIENCE OF HORSE HUSBANDRY

JAMES BLAKELY

Reston Publishing Co., Inc. Reston, Virginia. 1981. (ISBN 0-8359-2887-1) Price R25,20

Many books and literature have been published on specific subjects and fasets of Horse Science. This publication succeeds in providing a down-to-earth study on this subject. From a complete theory of evolution to the most up-to-date practical information on breeds, history, husbandry and 2 very interesting chapters on training and psychology of the horse, it puts facts over to the horse enthusiast as well as to students in this field.

It lacks unnecessary technical terms and is provided with numerous suitable illustrations which makes it easy to read and to understand.

It gives concise information of over 50 breeds of horses and classes. It includes a section on reproduction and genetics with many practical applications – even for the veterinary student. The basic principles of nutrition and disease are well dealt with, without creating situations where too little knowledge could create problems if applied as stated in the text. It therefore does not endanger the reader, to think that he/she will be a specialist in nutrition or a qualified veterinarian after reading through these chapters.

The sections on reproduction, selection and judging are very well put. The chapters on Tack, Horseshoes and Horseshoeing are very practical and logically arranged. This is also one of the few books today that gives an excellent discussion on the psychology of the horse – a topic which is extremely important in breaking, training and handling of horses.

This book provides teaching aids, it provides basic knowledge of husbandry for the enthusiast as well as practitioner and it will provide suficient reference material and illustrations to the student in this subject.

L.J. Kritzinger

A RETROSPECTIVE STUDY ON 120 NATURAL CASES OF CANINE EHRLICHIOSIS

J. VAN HEERDEN*

ABSTRACT: Van Heerden J. **A retrospective study on 120 natural cases of canine ehrlichiosis.** *Journal of the South African Veterinary Association* (1982) **53** No. 1 17-22 (En) Department of Medicine, Faculty of Veterinary Science, University of Pretoria, Box 12580, 0110 Onderstepoort, Republic of South Africa.

In a retrospective study on 120 natural cases of canine ehrlichiosis, it was found that cases were presented in all months of the year. The disease was diagnosed in 26 different breeds amongst which the German Shepherd was the best represented. Of the different breeds of dog, German Shepherds showed the highest incidence of chronic cases as well as the highest mortality rate. In a randomly selected sample of 50 dogs, the clinical signs of canine ehrlichiosis were found to be mainly non-specific. The terms acute, subacute and chronic are proposed to describe the different stages of disease. The total white cell count was found to be of prognostic value. Treatment with either doxycycline or oxytetracycline was found to be effective in most of the cases. Blood transfusion was the most important method of supportive therapy.

INTRODUCTION

Canine ehrlichiosis has been described in many different breeds of dog[1 2 11 17] as well as in many different parts of the world[3]. The relatively great susceptibility of the German Shepherd Dog has been suggested by the studies of Spence, Giam & Theis[13]; Huxsoll et al.[4] and Klopfer & Nobel[6].

The different stages of the disease as well as the clinical signs thereof have been described by Walker et al.[18] and Buhles, Huxsoll & Ristic[1]. Troy et al.[15] also reported on the relative incidence of the various clinical signs in a series of natural cases.

This report is a retrospective study of natural cases of canine ehrlichiosis.

MATERIALS AND METHODS

Data was collected from 120 clinical cases of ehrlichiosis presented for diagnosis and treatment over a 3-year period to the Department of Medicine, Faculty of Veterinary Science, University of Pretoria. The majority of cases (108) originated from the Pretoria District which includes the city and adjacent rural areas. Most cases came from the northern suburbs and adjacent areas. Only 12 cases came from elsewhere in Southern Africa namely:

(a) a Bull-Mastiff and a Pomeranian from Rustenburg, Transvaal
(b) a Beagle from Jan Kempdorp, Northern Cape
(c) a Samoyed from Johannesburg, Transvaal
(d) 8 German Shepherd Dogs from military operational areas which include areas in South West Africa/ Namibia.

On admittance the date, breed, age, sex and body mass were recorded. All cases were also subjected to a full physical examination, examination of a peripheral capillary blood smear and a haematological investigation of venous blood; in some cases serum was collected for the determination of serum proteins and an electrophoretic study.

The diagnosis of ehrlichiosis in all cases (except in dogs with severe pancytopaenia) was confirmed by the demonstration of morulae of *E. canis* in peripheral blood smears stained with Diff-Quik (Harleco).

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The diagnosis of ehrlichiosis in dogs with severe pancytopaenia was based on

(1) clinical signs of disease
(2) the presence of a leucopaenia, an anaemia and a thrombocytopaenia
(3) the presence of a hypergammaglobulinaemia
(4) post-mortem findings (of which a variable plasma-cell infiltration into many organs and tissues was the most outstanding histopathological finding).

Clinical cases of ehrlichiosis were classified as either acute/subacute or chronic. A case was classified as chronic based on the presence of

(a) clinical signs of chronic disease such as chronic progressive loss in body mass, chronic intermittent pyrexia, epistaxis, etc.
(b) a white cell count of $6 \times 10^9/\ell$ or less, and
(c) the presence of a hypergammaglobulinaemia.

All other cases were classified as acute/subacute.

The incidence of the various clinical signs amongst such randomly-selected cases was expressed as a percentage thereof. These cases were divided into 5 groups according to their respective total white cell counts. The percentage mortality in each group was established. They were also subdivided into 4 groups according to their respective red cell counts and again the percentage mortality in each group was established.

Cases were treated as follows:

Specific treatment with one of the following drugs was given:

(i) Oxytetracycline (Terramycin capsules, Pfizer) orally at a dosage rate of 100 mg/kg once daily or 50 mg/kg twice daily for not less than 10 days.
(ii) Oxytetracycline (Liquamycin – Pfizer) intravenously at a dosage rate of 5-10 mg/kg once daily for not less than 6 days. If the drug was given for less than 6 days, it was followed by the oral administration of oxytetracycline as indicated above until the dog was treated for not less than 10 days in total.
(iii) Doxycycline (Doxyvet, Milvet) either orally or intravenously at a dosage rate of 5-10 mg/kg for not less than 10 days.

Supportive treatment which was given in some instances, consisted of one or more of the following:

(i) A blood transfusion. A transfusion with fresh

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blood was given whenever an anaemic dog was presented in a bleeding state. (Fresh blood being blood collected immediately prior to transfusion.) Anaemic, non-bleeding dogs were given stored blood. Depending on the physical state of the patient, a blood transfusion was usually given when the haematocrit dropped to below 0,20. A dosage of 40 ml blood/kg was usually aimed for.

(ii) Intravenous fluids (Sodium Chloride 0,45 % m/v and Dextrose 2,5 % m/v) injection or polyionic Plasmalyte B, Sabax, Aerotan, Johannesburg).

(iii) Essential phospholipids at a dosage rate of one tablet (Essentiale, Nattermann) twice daily.

Concurrent infection with *B. canis* was treated with one or more of the following:

(i) Diminazine (Berenil, Hoechst) at a dosage rate of 3,5 mg/kg intramuscularly.

(ii) Trypan blue (Trypan Blue, Centaur Labs) at an approximate dosage rate of 10 mg/kg intravenously.

(iii) Imidocarb dipropionate (Forray-65, Coopers) at a dosage rate of 6 mg/kg subcutaneously.

(iv) Phenamidine isethionate 5 % (Phenamidine, Maybaker) at a dosage rate of 0,4 ml/kg subcutaneously.

In severe chronic cases one or more of the following drugs were used:

(i) Prednisolone (Medrol 4 mg tabs, Upjohn) at an oral dosage rate varying from 0,5-2 mg/kg daily up to 28 days.

(ii) Nandrolone (Laurabolin 50 mg/ml, Intervet) at an intramuscular daily dosage rate of 150 mg up to 19 days.

(iii) Levamisole (Tramisol 2,5 % m/v I.C.I.) at an oral daily dosage rate of varying from 3,3-10,0 mg/kg for up to 70 days.

RESULTS

Average monthly presentation of cases

The monthly distribution of 120 cases of canine ehrlichiosis over a 3-year period is given in Fig. 1.

Clinical cases occurred in all months of the year. The highest number of cases were seen during the months April (16), May (16) and June (17). The majority of cases were presented during the first 6 months of the year.

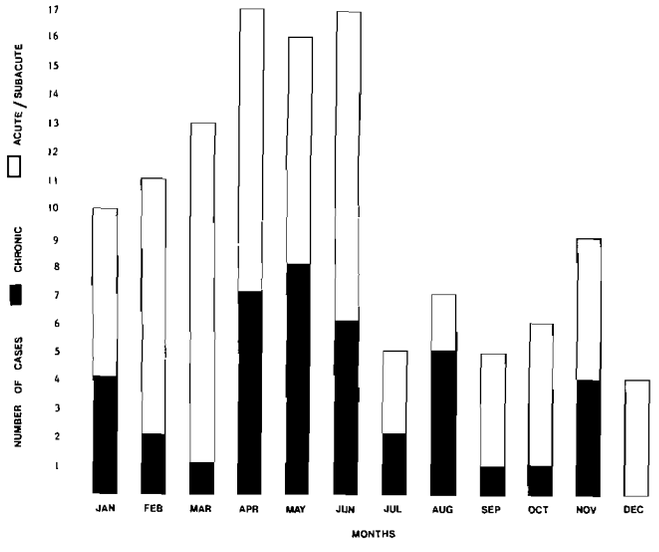

Fig 1: Monthly distribution of clinical cases of canine ehrlichiosis

Breed frequency

The 120 clinical cases of ehrlichiosis included dogs of 27 different breeds and crossbreds:

Breed	*Number*	*Percentage of the total number of cases*
German Shepherd Dogs (and German Shepherd Dog crosses)	28 (4)	23,3
Doberman Pinchers	17	14,1
Crossbreds	14	11,6
Labradors (and Labrador-crosses)	9 (4)	7,5
Bull Terriers	6	5
Fox Terrier Crosses	6	5
Boerboel	4	3,3
Collie	3	2,5
Pomeranian	3	2,5
Rottweiler	3	2,5
St Bernard	3	2,5
Pyrenian Mountain Dog	3	2,5
Greyhound	3	2,5
Other: (Boxer, Bull Mastiff, Bassett, Beagle, Dalmation, Irish Setter, Great Dane, Keeshond, Maltese Poodle, Old English Sheepdog, Schipperke, Samoyed, Scottish Terrier, Rhodesian Ridgeback, Wirehaired Terrier)	18	15,0

The 3 best represented groups/breeds were:

(a) German Shepherd Dogs and German Shepherd Dog crosses

(b) Doberman Pinchers

(c) Crossbreds

Chronic cases within the different breeds

Chronic cases of ehrlichiosis occurred in 16 different breeds;

Breed	*Number*
German Shepherd Dog	14
Labrador	5
Crossbreds	4
Bull Terrier	3
Collie	2
Bull Mastiff	2
Pomeranian	2
Boerboel	1
Wirehair Terrier	1
Dobermann	1
Greyhound	1
Fox Terrier	1
St Bernard	1
Rhodesian Ridgeback	1
Keeshond	1
Pyrenian Mountain Dog	1

Forty-one or 34,2 % of the 120 cases were classified as chronic cases. Fourteen or 58,3 % of the German Shepherd Dog cases were classified as chronic cases.

Incidence of mortality

Thirty or 25 % of the 120 cases died. Mortality occurred in 13 different breeds:

Breed	*Number*
German Shepherd Dog	10
Bull Terrier	4
Dobermann	4
Labrador	2
Crossbreds	2
Boerboel	1
Fox Terrier	1
Maltese Poodle	1
Pyrenian Mountain Dog	1
Greyhound	1
Pomeranian	1
Wirehair Terrier	1
Great Dane	1

More than 41 % of the German Shepherd Dogs did not survive the disease.

Incidence of clinical signs

The incidence of specific clinical signs amongst the randomly-selected cases was as follows:

Anorexia	84 %
General depression	84 %
Anaemia	76 %
Fever	70 %
Peripheral lymph node enlargement	64 %
Loss in body mass	59 %
Splenomegaly	36 %
Epistaxis	22 %
Mucopurulent ocular and/or nasal discharges/conjunctivitis	8 %
Icterus	4 %
Vomition	4 %
Limb-oedema	2 %
Bronchial râles	2 %

"*Anorexia*" – a varying degree of anorexia was reported by owners and observed amongst hospitalized cases. In most cases only a reduced appetite or an intermittent anorexia was observed. Total anorexia was only recorded from dogs in the acute (febrile stage) or terminal chronic phase of the disease.

"*General depression*" – a varying degree of general depression was observed and dogs often showed only intermittent listlessness.

"*Anaemia*" – the clinical diagnosis of anaemia corresponded well with the laboratory diagnosis. According to the latter, 84 % of the cases were anaemic and on physical examination 75 % of the cases were found to be anaemic.

"*Loss in body mass*" – this varied from a very slight drop in condition to extreme emaciation in advanced chronic cases.

"*Other haemorrhages*" – these included haemorrhages in the skin and into the chambers of the eye.

Incidence of blood parasites other than *Ehrlichia canis*

A concurrent infection with *Babesia canis* was found in 20 of the 50 randomly-selected dogs. Gametocytes of *Hepatozoon canis* were also found in peripheral blood smears in 8 of the cases.

The relationship between leukopaenia and mortality

The total white cell counts of 50 cases are given in Table 1.

Table 1: CLASSIFICATION OF *EHRLICHIA* CASES ACCORDING TO THEIR TOTAL WHITE CELL COUNTS

Group	White cell count × $10^9/\ell$	Number of cases	% Mortality in group
A	0-1	4	75
B	1,1-2	5	100
C	2,1-6	11	9,1
D	6,1-12	21	9,5
E	12+	9	22,2

The percentage mortality for the different groups A, B, C, D & E was 75, 100, 9,1, 9,5 and 22,5 % respectively. All cases, except one, with a total white cell count of 2 × $10^9/\ell$ or lower, died.

Relationship between red cell count and mortality

The results of classification of 50 clinical cases of ehrlichiosis according to their red cell counts are given in Table 2.

Table 2: CLASSIFICATION OF CLINICAL CASES OF EHRLICHIOSIS ACCORDING TO THEIR RED CELL COUNTS

Group	Red cell count × $10(^2/\ell$	Number of cases	% Mortality within group
A	0-2	8	62,5
B	2,1-3	19	31,5
C	3,1-4	10	20
D	4+	13	0

There was a tendency towards greater mortality in more anaemic dogs.

Treatment

Thirty or 25 % of the 120 cases did not survive the disease despite treatment (Fig 1). Most cases that died were either patients with severe pancytopaenia or cases complicated by concurrent infections with *B. canis* and/or *H. canis*.

Treatment of cases with severe pancytopaenia often was an expensive and prolonged commitment. The use of corticosteroids and/or anabolic steroids in a few cases did not seem to alter the outcome of severely pancytopaenic cases.

Two severely pancytopaenic cases were treated with laevamisole (antibiotic treatment was given at the same time) over an extended period of time (Table 3). One of them, Case A, was treated for 70 days at a daily oral dosage rate of 3,3 mg/kg and is one of 2 cases that recovered from severe pancytopaenia. The other patient, Case B, initially showed a good response to treatment, but subsequently deteriorated and died. There is, however, some uncertainty as to whether the owner continued with the treatment at home.

DISCUSSION

Average monthly distribution of cases

The highest number of clinical cases were presented during late autumn and early winter. With the exception of the month December, the lowest number of cases were presented during July, August, September and October. Cases of canine ehrlichiosis were, however, presented in all months of the year.

Table 3: TOTAL WHITE AND RED CELL COUNTS OF 2 DOGS TREATED WITH LEVAMISOLE

CASE A

Day	2	5	7	9	12	15†	19	22	28	34	52	65	72	86	124
Red cell count × $10^{12}/\ell$	4,77	3,58	3,81	3,58	2,6	2,7	1,79	1,93	1,85	1,94	2,33	3,92	4,06	5,42	7,14
White cell count × $10^{9}/\ell$	1,7	2,6	1,4	2	1,7	1,5	0,99	0,80	1,05	1,6	3,7	3,3	3,2	4,5	6,7

†levamisole therapy initiated at a daily dosage rate of 3,3 mg/kg

CASE B

Day	1*	4	11**	47***	66
Red cell count × $10^{12}/\ell$	1,54	2,92	2,95	4,53	4,36
White cell count × $10^{9}/\ell$	1,3	2,3	5,5	1,1	3,3

*levamisole therapy initiated at a daily dosage rate of 9,6 mg/kg
**levamisole therapy reduced to a daily dosage rate of 3,3 mg/kg
***levamisole therapy increased to a daily dosage rate of 9,6 mg/kg

If the relatively long (subclinical) subacute phase of the disease is kept in mind, then the observed monthly distribution of presented cases bears some resemblance to the monthly fluctuation in population size of *Rhipicephalus sanguineus* as found by Horak. (Horak IG, Department of Parasitology, Faculty of Veterinary Science, University of Pretoria, personal communication 1980). Horak found population numbers of *R. sanguineus* to be at its lowest during June, July and August with an increase in population size from September onwards. The population size started to decrease during April/May. More extensive studies on both population dynamics of *R. sanguineus* as well as on the incidence of canine ehrlichiosis in a dog population within the same area might well reveal a closer relationship.

It should also be borne in mind that the relatively long and variable duration of the subacute and chronic phases of the disease would make it very difficult to correlate presented clinical cases with seasonal fluctuation in tick numbers. Owners are often inclined to present dogs only when they show clear signs of disease – at which time the dog might well already be in the chronic stage of disease. It is impossible to determine the exact duration of the disease in a dog, even with the aid of haematological and serological parameters.

The small number of cases presented during December is probably misleading due to the fact that referrals to the Medicine Department during this period are usually very low. The drop in clinical cases during winter might well be ascribed to lower number of ticks.

Breed-frequency

Although the presented figures do not represent the relative susceptibility of the different breeds in the dog population of the study area, they do seem to indicate an over-representation of the German Shepherd Dog. The breed frequency in the dog population within the study area has, however, not been determined. In a survey conducted by Osterhoff[9] amongst 600 dog owners in the Pretoria-Johannesburg area and elsewhere in the Republic of South Africa, German Shepherd Dogs represented 5,3% of the dog population. Twenty-eight percent of the dogs in Osterhoff's study were crossbreds and might well explain the relatively higher number of crossbreds suffering from ehrlichiosis in this study.

Chronic cases/mortality within the different breeds

The fact that the percentage of chronic cases and the percentage mortality for German Shepherd Dogs was considerably higher than the same figures for all of the remaining dogs combined, might well be an indication of the relatively greater susceptibility of the German Shepherd Dog for *E. canis*. It might well be an indication of a decreased ability by this breed to develop a cell-mediated immunity against *E. canis*.

Incidence of clinical signs

The clinical signs of canine ehrlichiosis are mainly non-specific signs. This is indicated by the high incidence of clinical signs such as anorexia, general depression and anaemia. The recorded clinical signs in general agree with clinical features as described by Walker et al[18] and Buhles et al[1].

In an analysis of clinical signs in 30 cases of canine ehrlichiosis, Troy et al.[15] recorded anaemia, loss in body mass, anorexia, general depression and a fever reaction as the most common clinical signs of ehrlichiosis.

The higher incidence of a fever reaction in this study is probably misleading. Factors such as transport of the dog to the clinic, a period of waiting in the waiting room and physical examinations at the outpatient clinic might have contributed to the observed fever-reaction. This is supported by the fact that patients often had normal rectal temperatures within 24 h of admittance and before treatment was initiated.

The relatively low incidence of epistaxis in the present investigation illustrates the fact that epistaxis should not be regarded as a classical clinical sign of canine ehrlichiosis.

Walker et al.[18] described the course of clinical disease in canine ehrlichiosis as a fever phase followed by a subclinical phase which might proceed to a terminal phase. Buhles et al.[1] described the subclinical phase as a mild chronic pancytopaenic phase and the terminal phase was described as a severe chronic pancytopaenic phase.

The terms acute, subacute and chronic should, however, be used to describe the different stages of canine ehrlichiosis. The term "subclinical" should not be used

to describe the stage of disease following on the acute stage of disease, because "subclinical" implies the complete absence of clinical signs of disease. In my opinion this is not the case. Although dogs might only display subtle signs of disease, they are nevertheless present. These signs include one or more of the following: (intermittent) slight depression; varying degree of anorexia, often intermittent; varying degree of loss in body mass; mild enlargement of peripheral lymph nodes; mild anaemia. Describing the subacute phase as a mild chronic pancytopaenic phase is also incorrect in that dogs in this stage often do not suffer from a pancytopaenia. It should be stressed that it is often very difficult to distinguish between the different stages of canine ehrlichiosis.

Blood-parasites other than *E. canis*

The relatively common occurrence of a combined infection of *B. canis* and *E. canis* in the dog necessitates meticulous examination of peripheral blood smears; especially in so-called cases of biliary relapse.

Leukopaenia

A very high mortality rate occurred amongst dogs with a total white cell count of $2 \times 10^9/\ell$ and lower.

Although pancytopaenia occurs in both the acute[1,4] and chronic stages[18] of the disease, the leukopaenia in the acute stage of the disease is usually less severe and of a relatively transient nature. Observations on the total white cell count of experimentally infected dogs in the acute stage of canine ehrlichiosis has shown it to be a less constant feature than anaemia and thrombocytopaenia. The drop in total white cell count during acute stages was also less severe than in the chronic stages of the disease (author's unpublished data). Seamer & Snape[12] also reported the changes in white blood cell counts to be the least striking of the haematological changes in the acute stage of the disease. Reardon & Pierce[10] also reported a slight decrease in total leukocyte counts 14 days after infection.

In view of the high mortality rate found amongst severely leukopaenic dogs, the total white cell count thus seems to be of prognostic value in canine ehrlichiosis. A prognostic parameter is of great importance especially in chronic cases where treatment is often prolonged, expensive and unrewarding.

In the present investigation the mortality rate, based on red cell counts, was the highest amongst dogs with a red cell count of lower than $2 \times 10^{12}/\ell$. There was, however, not such a dramatic difference in mortality (as was the case in the white cell count groups) between the different groups of anaemic dogs.

Treatment

The high oral dosage rates of oxytetracycline have been used according to the recommendations of Immelman[5] who has shown that such a dose rate would result in adequate blood levels of oxytetracycline for 24 h.

One of the most important supportive treatments in patients with canine ehrlichiosis is the administration of blood. In bleeding (thrombocytopaenic) anaemic patients, a fresh blood transfusion should be given, whilst in non-bleeding anaemic patients one need not necessarily administer fresh blood. (Fresh blood here refers to blood that was collected immediately prior to transfusion.)

The decision to treat severely pancytopaenic dogs with levamisole was based on

(1) Possible immunosuppression of the cell-mediated immune response in severe canine ehrlichiosis[8];
(2) the observation that both monocyte-derived macrophages and antibodies are necessary to destroy or suppress the growth of *E. canis* in vitro[7];
(3) the immuno-stimulant properties of levamisole[14].

Levamisole restores polymorphonuclear, macrophage or T cell functions such as random migration, phagocytosis and cell mediated cytotoxicity. Its effects are especially marked on hypofunctional cells. The effects of levamisole on leukocytes are brought about by its ability to increase intracellular levels of cyclic guanosine monophosphate and by reversing the inhibitory effects of cycle adenosine monophosphate (c AMP) elevating agents. On the contrary, levamisole also induces T cell differentiation, an apparently c AMP mediated phenomenon. A similar dual mechanism has also been sugested for thymic hormone. Like levamisole, thymic hormone specifically effect T cells and not B cells[14].

It is interesting to note that thymic atrophy has been a remarkable post mortem finding in experimentally induced canine ehrlichiosis in dogs (JW Nesbitt 1981 Department of Pathology, Faculty of Veterinary Science, personal communication) used in experimental studies reported elsewhere[16].

No definite recommendations can be made as to the treatment of severe chronic cases with canine ehrlichiosis. Since completion of this study 2 more severely leukopaenic German Shepherds have been cured with levamisole-therapy and it is suggested that levamisole should be used as follows:

(i) at a daily oral dosage route of 3-10 mg/kg and
(ii) for not less than 60 days.

When using the drug for any length of time, the possibility of aggravating the leukopaenic state should, however, be kept in mind. A reversible granulocytopaenia has been described in man[14]. The use of immunostimulants such as levamisole should be investigated further.

In conclusion, it can be stated that dogs were presented with clinical signs of canine ehrlichiosis in all months of the year. A variety of breeds were presented but the German Shepherd breed was especially well represented. A high percentage of German Shepherd Dogs developed the chronic phase of the disease and died. The clinical signs of the disease were found to be mainly non-specific. The terms acute, subacute and chronic are proposed to describe the different stages of the disease. A total white cell count determination was found to be of prognostic value.

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ABSTRACT: Giesecke, W.H. & Van den Heever, L.W., 1981. **Levels of glucose, serum albumin and somatic cells before and during early stages of acute clinical mastitis artificially induced in cows by means of human strains of group-B streptococci (GBS) administered intracisternally.** *Onderstepoort Journal of Veterinary Research,* **48,** 69-75 (1981).

The investigation was performed on 3 cows, sampled repeatedly before and during the initial 48 h of artificially induced, acute, clinical mastitis. The results of the investigation both augment and support those of earlier work on the levels and significant correlations of glucose, serum albumin and somatic cells in normal and abnormal secretions monitored before and after the usual milking of healthy lactating cows had been suspended.

During acute mastitis, udder secretions from artificially infected quarters showed highly significant escalations of somatic cell counts which coincided with equally significant increases on a high and intermediate level of serum albumin values in both the infected and non-infected quarters. Corresponding glucose values fluctuated from 0,07-0,22 and 0,18-0, 32 mM in the former and latter quarters respectively.

The selective and elevated transfer of serum albumin in otherwise unaffected quarters of acutely mastitic udders suggests rather specific collateral vascular and epithelial changes of unknown nature and magnitude.

The data indicate that marked fluctuations of glucose may occur within and between quarters of individual and different cows respectively. Such variations could significantly affect phagocytosis and killing of bacteria challenging the intramammary leucocytic udder barrier before and particularly during manifestation of mastitis. Hence, udder health, although dependent on specific natural defence mechanisms such as the leucocytes and related systems in milk, may depend even more significantly on the supplies of glucose to and within the bovine mammary gland.

ABSTRACT: Van Rensburg, L.J. & Van Wyk, J.A., 1981. **Studies on schistosomiasis. 10. Development of *Schistosoma mattheei* in sheep infested with equal numbers of male and female cercariae.** *Onderstepoort Journal of Veterinary Research,* **48,** 77-86 (1981).

The development of the female *Schistosoma mattheei* was significantly higher than that of the male ($P < 0,0001$) in 12 sheep when each was exposed to equal numbers of male and female cercariae. Many more male than female worms usually develop after infestation with pools of cercariae of mixed sexes, a phenomenon which in the light of the present results seems to be due to a preponderance of male cercariae and not to the more efficient development of male than female cercariae.

The female worms recovered fell into 2 distinct population groups as regards length and pigmentation. Some overlap in the measurements of the breadths and in the numbers of ova in the uteri of the worms, however, makes the demarcation of the different populations less distinct in these respects. The female worms from 3 single-sex infestations contained either no ova (72 days after infestation) or fewer (after 134-137 days of development) than the small females from the 12 sheep.

The number of large females (602) recovered from the mesentery was approximately the same as that of the males (605) from this site. Similarly, although varying numbers of small female worms were recovered from the liver of every sheep, only 2 males and 2 large females were recovered, and these were from the same liver.

Because of the similarity between the numbers of male and large female worms, it is clear that, for *S. mattheei*, physical contact with male worms is essential for development to maturity of female worms, the mere presence of males in the host not being sufficient for this development to take place.

ABSTRACT: Barnard, B.J.H. & Geyer, H.J., 1981. **Attenuation of turkey meningo-encephalitis virus in BHK21 cells.** *Onderstepoort Journal of Veterinary Research,* **48,** 105-108 (1981).

Turkey meningo-encephalitis virus was adapted to BHK21 cell culture. Cytopathic effects were characterized by rounding and detachment of cells within 48 hours. Attenuation was achieved by 41 successive passages in BHK21 cell cultures. Turkeys and Japanese quail (*Coturnix coturnix japonica*), kept under laboratory conditions and inoculated with the attenuated virus, did not develop symptoms of turkey meningo-encephalitis but reacted by the production of haemagglutination inhibition antibody. They resisted intracerebral challenge with pathogenic strains of turkey meningo-encephalitis virus.

THE USE OF DOXYCYCLINE TO CONTROL HEARTWATER IN SHEEP

A. IMMELMAN and GILLIAN DREYER*

ABSTRACT: Immelman A.; Dreyer G. **The use of doxycycline to control heartwater in sheep.** *Journal of the South African Veterinary Association* (1982) 53 No. 1, 23-24 (En) Department of Physiology, Pharmacology and Toxicology, Faculty of Veterinary Science, University of Pretoria, P.O. Box 12580, 0110 Onderstepoort, Republic of South Africa.

A ready-to-inject solution, containing 100 mg/ml doxycycline was used. Artificially infected sheep were divided into 2 groups: a control group consisting of 10 animals, and a treated group consisting of 12 animals. Doxycycline was administered intramuscularly to the animals in the treated group at a dosage rate of 2 mg/kg body mass. All the treated animals recovered. One control animal recovered spontaneously, the others all died.

Key words: Doxycycline, heartwater, sheep.

INTRODUCTION

The disease, heartwater caused by *Cowdria ruminantium* is of great economic importance in the Republic of South Africa. It is restricted to certain regions of the country. When cattle, sheep and goats are introduced from heartwater-free areas into these regions, a high rate of mortality may occur. Even in animals that are raised in heartwater areas and attain a natural immunity, individual cases do occur. If treatment is not instituted early in the course of the disease the prognosis is very poor.

In 1939 Neitz, as quoted by Weiss, reported successful therapy of the disease with the drug Uleron[6]. Other sulphonamides, e.g. sulphanilamide, sulphapyridine and sulphadiazine were used later with success to combat the disease. It was stated that the effective blood concentrations required were very low and the exposure time needed was much shorter than that required for bacteria. The parenteral route was the route recommended[6].

With the development of tetracycline antibiotics it was established that they were more effective than the sulphonamides to control *C. ruminantium* infection. Chlortetracycline was initially used at a dosage rate of 5 mg per lb (11 mg/kg) for 4 days. This treatment resulted in a prompt recovery and with the development of a good immunity against the organism. It was subsequently established that 2,5 mg per lb (5,5 mg/kg) body mass as a single treatment administered in the early stages of the disease was effective. A dose of 1,25 mg per lb (2,75 mg/kg) has a marked beneficial effect but was not always reliable[6].

When oxytetracycline became available it was established that the intravenous administration of 2,5 mg per lb (5,5 mg/kg) body mass was a very effective treatment for heartwater[2]. The intramuscular injection with procaine was effective, although there was a delay of 24-48 h before the body temperature returned to normal.

The standard dosage rate of oxytetracycline used today to control heartwater is 10 mg/kg body mass. This treatment is usually repeated on the following day if the body temperature is still above normal. Should the fever persist for more than 48 h it is suggested that an intravenous dose of sulphadimidine should be included with the third dose of oxytetracycline[4].

Doxycycline is a tetracycline of more recent vintage and is a synthetic derivative of methacycline. It is more lipid soluble than oxytetracycline. In humans it was established that doxycycline has a longer serum half-life than oxytetracycline, with the advantage of smaller therapeutic doses and longer intervals between treatments[1].

In veterinary medicine the efficacy of doxycycline in treatment of anaplasmosis in cattle and canine ehrlichiosis has been established[3 5], and it was the purpose of this experiment to determine its efficacy in the treatment of heartwater.

MATERIALS AND METHODS

Twenty-two crossbred Dorper ewes varying in age from 24 to 36 months were used in this experiment. The animals were raised in the North-Western Cape where heartwater does not occur and were therefore regarded as being susceptible.

The average mass of the ewes was 34,6 kg (22,7-44 kg). After arrival at the Faculty of Veterinary Science where the experiment was carried out, they were treated with albendazole (Valbazan, Smith, Kline, French), a broad-spectrum anthelmintic, and were housed indoors in individual pens on concrete floors. They were fed lucerne hay and water was freely available.

The heartwater-infected blood, issued by the Veterinary Research Institute at Onderstepoort as a vaccine was used to infect the animals. Five millilitres of the blood was administered to each sheep by a slow intravenous injection (Day 0). From the 9th day onwards, rectal temperatures were recorded in the mornings and late afternoons. A temperature above 40 °C was regarded as indicative of a positive reaction. At the commencement of the experiment the sheep were arbitrarily divided into 2 groups: the 12 animals in one group to be treated with doxycycline and the 10 in the other group to be kept as infected controls. Treatment was administered only after the temperature remained above 40 °C for 3 consecutive recordings, i.e. 24 h after the initial rise.

Doxycycline was administered as doxycycline hyclate in a ready-to-inject solution containing 100 mg/ml active material (Doxyvet, Samvet Laboratories). A single administration of 2 mg/kg body mass was injected into the *Musculus gluteus*. After treatment rectal temperatures were taken once daily till the end of the trial.

The trial was concluded when the control animals had either all died or recovered spontaneously.

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RESULTS AND DISCUSSION

All the animals in the treated group reacted; the first administration of doxycycline took place on the 13th day after infection, and the last after 15,5 days. The average rectal temperature of these animals at the time of treatment was 40,5 °C and the average time taken for the temperature to return to below 40 °C, was 3,2 days. All the treated animals recovered.

In the control animals temperature reactions were recorded from Day 11 to Day 15. Symptoms progressed and the first death occurred on the 16th day after administering the infected blood and the last on Day 23. The 9 animals that died survived for an average period of 6,1 days (3-12 d) after the first rise in temperature. One sheep in this group developed a fever of 40,4 °C on Day 15. On Day 18 the fever had risen to 41 °C but from Day 20 there was a decline in the fever and she recovered completely.

All the carcasses were submitted for autopsy and in each case heartwater was confirmed as the cause of death.

The efficacy of doxycycline as established during this trial corresponds to the findings of Haig using low doses of oxytetracycline[2]. Van der Merwe reporting on her experience in a large number of cattle vaccinated against heartwater indicated that a large percentage of animals will not show a reduction of body temperature after the first administration of oxytetracycline at a dose of 10 mg/kg body mass. She suggested that repeated treatment with oxytetracycline[4] should be used as a routine.

As the ewes in this trial were under unstressful conditions, the influence of heat, movement and other stress factors on the course of the disease with or without treatment could not be studied.

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The authors wish to express their appreciation to Samvet Laboratories for financial assistance and supply of the drug.

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BOOK REVIEW — BOEKRESENSIE

PRINCIPLES OF ANIMAL VIROLOGY

W.K. JOKLIK

Appleton-Century-Crofts, New York 1980 pp X 373 Figs 152 (1 colour plate) Tabs 30 Publ. Price not stated.

The title of this book is somewhat misleading in that it only deals with medical virology.

The book is divided into 2 sections. Section 1 (written by Dr Joklik) is devoted to basic virology. It constitutes about 60 % of the book and deals among others with the nature, morphology, replication, genetics and classification of viruses. It is extremely well written with very few printing errors or inaccuracies (e.g. the capsomere number for Iridoviruses – p 19). The undisputed value of the book for both the under- and post-graduate veterinary student undoubtedly lies in this section. It may even be useful for the practising veterinarian who needs to update his knowledge on newer virological concepts such as antiviral chemotherapy and the role of viruses in cancer.

Section 2 is written by 5 different authors and deals with clinical virology. This first chapter on diagnostic virology is disappointingly brief (5 pages) and barely touches on highly important issues such as the collection and handling of specimens, the isolation and identification of viruses and the examination of specimens by light – and electron microscopy. The remaining 14 chapters describe the human disease conditions caused by the major families of viruses. This section would, for obvious reasons, be of marginal interest only to veterinarians, veterinary students and non-medical virologists.

B.J. Erasmus

THE TREATMENT OF *URGINEA SANGUINEA* SCHINZ POISONING IN SHEEP WITH ACTIVATED CHARCOAL AND POTASSIUM CHLORIDE

J.P.J. JOUBERT and R. ANITRA SCHULTZ*

ABSTRACT: Joubert, J.P.J. & Schultz, R. Anitra. **The treatment of *Urginea sanguinea* Schinz poisoning in sheep with activated charcoal and potassium chloride.** *Journal of the South African Veterinary Association* (1982) **53** No. 1 25-28 (En) Department of Toxicology, Veterinary Research Institute, 0110 Onderstepoort, Republic of South Africa.

An 80 % survival rate was achieved when sheep which received lethal doses of *Urginea sanguinea* were treated with activated charcoal at 5 g/kg and potassium chloride at 1 g/kg body mass *per os*. A brief description of *U. sanguinea* poisoning and of the mechanism of cardiac glycoside action is given. Clinical examinations, ECG recordings and serum potassium levels were done to monitor developing signs of poisoning.

INTRODUCTION

Urginea sanguinea Schinz (= *Urginea burkei*), also known as Transvaal slangkop, contains a bufadienolide cardiac glycoside, transvaalin, as its toxic principle[10]. The bulbous plant grows in the Transvaal, the western Orange Free State, Griqualand West and South West Africa[16], and together with the large number of other cardiac glycoside containing plants, can be regarded as one of the 6 most important groups of poisonous plants hazardous to cattle, sheep and goats in the Republic of South Africa[11].

Heavy losses may occur amongst stock grazing slangkop infested pastures. In general, the clinical signs are those typical of cardiac glycoside poisoning: an increased respiratory rate with forced expiration, tachycardia leading to arrhythmias, ruminal atony, a severe watery diarrhoea which may become blood stained, tympany and weakness of the hindquarters. Depending on the quantity ingested, death may occur within a day or two, or after a few days. Clinical signs of poisoning may sometimes be seen only 2 days after ingestion of the plant. Most cases occur during spring or in a drought when the succulent green flowerhead appears above the ground while no other vegetation is available. Stock reared on slangkop veld rarely graze the plant unless they are forced to do so. The repeated ingestion of small amounts of the plant does not produce toleration, but has a cumulative effect[16 17].

Although a large number of remedies, varying from cardiac stimulants to the universal antidote are being used, treatment of poisoning in ruminants has never been evaluated scientifically (T W Naudé, 1981 Veterinary Research Institute, Onderstepoort, personal communication).

Cardiac glycosides, when in the bloodstream, eventually bind to extracellular binding sites on cell membranes, especially of conducting tissues such as nerve fibres and cardiac-, skeletal- and smooth muscles. Cardiac glycosides thus inhibit a specific cell membrane sodium potassium adenosine triphosphatase which facilitates the active transport of Na ions out of, and K ions into the cell. This active transport process ensures a stable membrane electropotential necessary for the conduction of impulses. The positive inotropic effect (at therapeutic levels) and toxicity (at poisonous levels) of cardiac glycosides are due to the resultant increase of Na^+ and Ca^{++} ions and depletion of K^+ ions intracellularly[1 13 14].

In human medicine, cardiac glycoside poisoning is of great importance, since cardiac glycosides such as digoxin are often used to treat cardiac failure and, because of a narrow therapeutic index, overdosage often occurs[5]. The numerous evaluations of therapy of cardiac glycoside poisoning in humans[3 4 5 15], may beneficially be applied to veterinary science.

Potassium chloride is an old therapeuticum which is still in use. The improvement brought about with potassium chloride in cardiac glycoside poisoning has been noted, amongst others, by Loewi[9], De Graff & Lyon[4], Chung[3], and Follath[5]. The increased concentration of potassium ions in the extracellular fluid may alleviate the condition by competition with cardiac glycoside molecules for binding sites on cell membranes and by increasing the amount of K^+ ions available for active transport by the few enzyme systems still active.

Another well-known therapeuticum which is used with good results in cardiac glycoside poisoning, even in suicide cases, is activated charcoal. Recently, the activated charcoal particles are coated with dextran and used as a filter in a process of haemoperfusion. Cardiac glycoside molecules are thus removed from the patient's blood as it absorbs onto the activated charcoal[7 15]. This technique is also described for experimental use in rabbits[6]. Per os dosing of activated charcoal to rats injected parenterally with digoxin significantly increased the excretion of digoxin in the faeces[2] as a result of the interruption of the enterohepatic recirculation of digoxin.

This paper decribes the per os dosing of sheep with a combination of activated charcoal and potassium chloride as a treatment for experimentally induced slangkop poisoning.

MATERIALS AND METHODS

Urginea sanguinea bulbs were collected on the farm "Tesame", 40 km north of Pretoria and identified at the Botanical Research Institute. The bulbs were milled, dried and stored at 4 °C.

Twenty-three Merino and Dorper sheep of both sexes, aged from less than 15 months to full-mouthed, and with live masses between 31 and 70 kg, were used.

Sheep dosed with charcoal and KCl

Initially, one sheep was dosed per stomach tube with activated charcoal suspended in water at 20 g/kg body

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TABLE 1: TREATMENT OF *URGINEA SANGUINEA* POISONING IN SHEEP WITH 5 g/kg ACTIVATED CHARCOAL PLUS 1 g/kg POTASSIUM CLORIDE PER OS.

Treated group									Control group						
Sheep			*U. sanguinea* dose g/kg	Interval before treatment (h)	ECG changes and clinical signs before treatment ▼	Clinical signs after treatment	ECG changes	Fate	Sheep			*U. sanguinea* dose g/kg	Clinical signs	ECG changes	Fate
No.	Sex	Age							No.	Sex	Age				
1	♂	6t	2	5	n/a	n/a	n/a	Survived	11	♂	4t	2	Respiration abdominal, decreased rumination leading to ruminal atony, anorexia, diarrhoea, remain recumbent on Day 9	Day 2: QRS-wave widens Day 7-9: coupled rhythm, ST depression and ectopic foci	Euthanazed in *extremis* Day 9
2	♂	fm	2	17,5	Anorexia, slight arrhythmia	Faeces soft for 1 day, eat and ruminate	n/a	Survived	12	♂	fm	2	Decreased rumination, diarrhoea from Day 2, poor appetite	Day 1-12: n/a Day 13: ST depression	Survived
3	♀	4t	2	24	Faeces soft, heart rate 220, QT depression	Faeces soft for 1 day, eat and ruminate	n/a	Survived	13	♀	6t	2	Decreased rumination, poor appetite serum K^+ 6,5 mmol/ℓ on Day 2	Day 6-8: sinus arrhythmia	Survived
4	♂	2t	2,25	24 + 48*	Diarrhoea, forced abdominal respiration, rumen atony, depressed	Groan with expiration severe diarrhoea Day 2 eat on Day 3 Serum K^+ 7,98 mmol/ℓ	Day 1-5: n/a Day 6: QT depression	Survived	14	♂	2t	2,25	Forced respiration, listless Serum K^+ 6,19 mmol/ℓ	Lead II: T amplitude increased to 1,8 mV	Died in 44 h
5	♂	2t	2,5	24	Very depressed, lying down Lead II: QRS amplitude increased to 4,3 mV	Remains depressed died 4 h later		Died	15	♂	2t	2,5	Respiration forced, ruminal atony, severe diarrhoea	Lead II: QRS amplitude increased to 3,1 mV	Died in 48 h
6	♂	2t	2,5	24	Respiration deep, irregular, rumen atony, diarrhoea	Rumination restored in 24 h	n/a	Survived	16	♂	2t	2,5	Respiration shallow, rumen atony, faeces soft	Extra sistoly, Lead II: Increased amplitude of QRS and T	Died in 48 h
7	♀	fm	2,5	19	Forced abdominal respiration, foam at mouth Lead I: Increased amplitude of P&T Lead II: Increased amplitude of QRS and T	No improvement died within 24 h		Died	17	♀	mt	2,5	Forced respiration, rumen atony, diarrhoea, foam at mouth	Lead II: QRS amplitude increased to 3,7 mV and T to 1,6 mV	Died in 48 h
8	♀	fm	2,5	19	Forced respiration severe diarrhoea Lead I: Increased amplitude of P	Rumination restored in 24 h, serum K^+ 6,7 mmol/ℓ	Day 3: slight sinus arrhythmia	Survived	18	♀	mt	2,5	Forced respiration, anorexia	Lead I: Increased amplitude of P	Died in 72 h
9	♀	fm	2,5	17	Forced respiration rumen atony, anorexia	Rumination restored in 72 h	n/a	Survived	19	♀	fm	2,5	Forced respiration, diarrhoea foam at mouth, rumen atony	Sinus arrhythmia, ST depression. Lead II: Increased amplitude of QRS	Died in 48 h
10	♀	fm	2,5	17	Slight tachycardia rumen atony, anorexia Lead I: Increased amplitude of P	Rumination restored within 24 h	n/a	Survived	20	♀	fm	2,5	Forced abdominal respiration rumen atony, diarrhoea	n/a	Died in 72 h

▼ = treatment
* = treatment repeated after 24 h
Day 0 = dosed with *U. sanguinea*
mt = milk tooth
t = tooth
fm = full-mouthed
n/a = nothing abnormal

mass once only. Another was dosed with a watery solution of KCl at 2 g/kg twice daily for 2 days, followed by a 5 day interval and 2 more days of dosing. A third sheep was dosed with KCl at 1 g/kg twice daily for 2 days followed by a 5 day interval and 4 more days of dosing.

Sheep dosed with *U. sanguinea*

The remaining 20 sheep were divided in 2 equal groups according to sex and breed. The control and treatment groups were housed in individual crates in a stable and fed the usual sheep ration of the Veterinary Research Institute. They were used in the trial in groups of 1 or 2 or more of each group as was practically feasible.

Electrocardiographic (ECG) recordings[12]

Clinical examination and blood chemical pathology were done daily. Firstly blood specimens were analysed comprehensively, while subsequent specimens were used for the determination of serum K, Na, Mg and Ca levels only. The sheep were deprived of food and water for 24 h before mass measurements were recorded and dosing per stomach tube with slangkop material. The initial dose of slangkop material of 2 g/kg was increased to 2,25 g/kg and again to 2,5 g/kg. Food and water were supplied again after the sheep had been dosed with the slangkop material.

These sheep were kept under observation to detect the onset of clinical signs of poisoning. At this stage the treatment group received per os as one dose, 5 g/kg activated charcoal and 1 g/kg KCl. The same dose was repeated after 24 h in one sheep only. Sheep which died were autopsied.

RESULTS

The initial comprehensive blood chemical pathology, ECG recordings and clinical examinations indicated that the sheep were healthy.

Sheep dosed with charcoal and KCl

The sheep dosed with activated charcoal at 20 g/kg passed soft black faeces, but never stopped eating and ruminating.

After being dosed with KCl at 2 g/kg for 2 days, the second sheep went off its feed and appeared listless. The treatment was repeated after 5 days and the sheep died within 12 h after the last dose. The third sheep, dosed with KCl at 1 g/kg, never developed any signs of illness.

The haemodynamic, respiratory, clinical changes and fate of the 20 treated and control sheep are given in Table 1. The ECG changes observed conformed with those expected in cardiac glycoside poisoning.

Sheep dosed with 2 g/kg *U. sanguinea*

Three treated and 2 control sheep survived. The other control was euthanazed 9 days after being dosed. Apart from slight QT suppression and tachycardia in one sheep nothing unusual was noted in ECG recordings of treated sheep. In the controls more severe changes such as arrhythmias and firing of ectopic foci occurred after a few days.

Sheep dosed with 2,25 g/kg *U. sanguinea*

The control sheep developed respiratory distress, was listless and on ECG showed an enlarged T-wave before it died. The treated sheep was the only one which received a second dose of activated charcoal and potassium chloride 24 h after the first and it started to eat and ruminate on the following day.

Sheep dosed with 2,5 g/kg *U. sanguinea*

There were 4 survivors out of 6 treated sheep and no survivors among 6 controls. Clinical signs included respiratory distress marked by forced abdominal expiration, ruminal atony, diarrhoea, anorexia and tachycardia. ECG changes occured in both groups before treatment. Abnormal enlargement of the QRS-wave (6 sheep), P-wave (4 sheep) and T-wave (3 sheep)[12] as well as sinus arrhythmia (2 sheep) were observed. Two of the sheep had normal ECG recordings prior to treatment (treatment group) ECG changes of the other 2 survivors of this group became normal soon after treatment. The control sheep either died before another ECG recording was made (3) or developed more severe ECG changes before they died (3).

Serum levels of cations revealed increases above normal of potassium of only 2 treated sheep which both survived and in 2 controls, of which 1 survived.

DISCUSSION

To evaluate properly the treatment of experimental slangkop poisoning, the lethal dose of slangkop material used must be well established. From unpublished data (Joubert[8]) it was ascertained that only 4 out of 36 Merino ewes survived the initial dose of 2 g/kg of the same material in another experiment. As the material was well dried, finely milled and stored at 4 °C, it was assumed that similar results would be obtained. When 2 controls out of 3 survived the dose, however, the dose was increased.

The time lapse between slangkop dosing and treatment was the next important factor. An attempt to allow some signs of poisoning to develop before treatment resulted in some sheep being treated at too early a stage and in 2 others the treatment was too late.

These 2 sheep were the only ones with abnormally enlarged QRS- and T-waves before treatment. It seems that treatment is ineffective when the heart is affected to that extent.

After the first 3 sheep were dosed, the doses of activated charcoal and potassium chloride were estimated to be as large as possible without being toxic. This was justified by the results obtained with these huge doses.

The fact that increased serum potassium levels were seen in a few sheep only, precludes any deductions regarding predictions or evaluations. However, according to Bismuth et al.[1], there is a significant correlation between serum potassium levels of human suicide cases taken 3-18 h after ingestion of a cardiac glycoside and imminent mortality.

The survival of 8 out of 10 treated sheep compares well with only 2 survivors out of 10 controls. This indicates that activated charcoal and potassium chloride can be used beneficially in the treatment of slangkop poisoning in sheep.

The questions arising from this work are whether potassium chloride and activated charcoal are both necessary for effective treatment and how efficacious would this treatment be in cattle and goats against cardiac glycoside plant poisonings other than slangkop.

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THE SAFETY OF INJECTABLE RAFOXANIDE IN CATTLE*

J. SCHRÖDER**

ABSTRACT: Schröder J. **The safety of injectable rafoxanide in cattle.** *Journal of the South African Veterinary Association* (1982) 53 No. 1 29-31 (En) MSD Research Centre, Private Bag 3, 1685 Halfway House, Republic of South Africa.

Thirty-two weaned steer calves were injected subcutaneously with rafoxanide solution to determine the lethal dosage. The recommended therapeutic dosage is 3 mg/kg. Eight of 12 calves treated at 45 to 60 mg/kg displayed signs of toxicity 24 hours to 8 days after treatment. These included recumbency, polypnoea, muscle tremors and clonic spasms, opisthotonus, paddling movements of the feet, and blindness with mydriasis and death. An easily detected histopathological lesion was status spongiosus of the central nervous system. This study demonstrated that injectable rafoxanide has a wide safety margin when used at the recommended therapeutic dosage.

INTRODUCTION

The usual recommended dosage of rafoxanide in a preformed suspension (Ranide, MSD) for cattle is 7,5 mg/kg by the oral route (as compared to 3 mg/kg by subcutaneous injection). At that dosage the compound is used to treat cattle infested with *Fasciola gigantica, Fasciola hepatica* and *Haemonchus placei*. It appears that the 7,5 % solution of rafoxanide (Ranide Injectable, MSD) is 2,5 times more bioavailable than the oral suspension when injected subcutaneously[4]. At 3 mg/kg injectable rafoxanide is effective against *Bunostomum phlebotomum* and *Oesophagostomum radiatum*, in addition to the previous 3 parasites.

In order to satisfy requirements for registration under the Fertilizers, Farm Feeds, Agricultural Remedies and Stock Remedies Act (Act 36/1947), the following experiment was performed to determine the lethal dose of injectable rafoxanide in cattle.

MATERIALS AND METHODS

Experimental animals

Thirty-two weaned Friesian bull calves, 3-15 months of age, were kept in concrete floored pens. They were fed a mixture of lucerne and *Eragrostis curvula* hay and maize silage and had free access to borehole water in steel troughs.

Treatment and observation

Each calf was identified with a numbered eartag and individually weighed prior to treatment. The calves were treated in 4 groups with doses of rafoxanide ranging from 15-21, 24-30, 33-42 and 45-60 mg/kg. Two calves were treated at each dosage. The trial extended over 2½ months. The calves were injected subcutaneously on the neck with half the dose immediately cranial to the left shoulder. The other half of the dose was injected caudal to the shoulder. There were no untreated controls.

The first 3 groups were examined at least once every day for 10-20 d after treatment for signs of toxicity. The pupillary reflex of each calf was tested 2-4 d after treatment. The fourth group of calves was examined every 2 h from 22-28 h after treatment, and thereafter once or twice daily until 10 d after treatment.

Calf 32 was killed by exsanguination for necropsy because it displayed most of the spectrum of clinical signs.

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Necropsy

All calves (8) that died were necropsied. Tissue specimens were taken for histopathology from most parenchymatous organs and fixed in 10 % formalin. From the central nervous system, specimens were taken of the optic chiasma, hippocampus, colliculus oralis (corpora quadrigemina), and cerebellum[5].

Tissue specimens of 5 calves were sectioned and stained with haematoxylin and eosin. Additional liver blocks were subsequently frozen, sectioned and stained with Oil Red O by a private medical pathology laboratory (Dr W.J. Pepler and partners, Pretoria).

RESULTS

Clinical signs

No signs of toxicity were seen in any of the 20 calves treated at 15-42 mg/kg, i.e. 5-14 times the recommended therapeutic dosage. One each of the 2 calves treated at 36 and 42 mg/kg became moribund because of pneumonia, and were killed for humane reasons. Eight of the 12 calves treated at 45-60 mg/kg started displaying signs of intoxication 24 h to 8 d after treatment. These signs consisted of recumbency (8/8), rapid shallow respiration (6/8), muscle tremors and clonic spasms (6/8), foaming at the mouth (4/8), and in 2 cases opisthotonus, paddling movements of the feet, nystagmus, prolapse of the nictitating membrane, and blindness with mydriasis.

The clonic muscle spasm occurred spontaneous at 30-60 second intervals, but could be elicited by external stimuli such as touch and sound, and apparently affected all skeletal musculature.

With the exception of calf 32 that was killed, the other 7 calves that showed clinical signs eventually died. The data from these animals are summarized in Table 1. Calves treated at 60 mg/kg died within 25-66 h after treatment, and calves treated at 45 mg/kg within 2-8 d.

Necropsy findings

Seven of the 8 calves that had shown clinical signs of acute toxicity had grossly visible subdural oedema at necropsy. Another prominent lesion was a mild to moderate hepatomegaly (roundness of the liver's edges, bulging of cut surfaces). In 4 calves there were subepi- and endocardial petechiae or ecchymoses, and 3 calves had a mild pulmonary oedema.

Histological examination revealed a mild to moderate status spongiosus in all the sections of nervous tissue (see Fig. 1). A marked feature in many sections was the wide Virchow-Robin spaces, indicating pericapillary

0038-2809/82/01/0029-0031

oedema. In the hippocampus the status spongiosus was confined to the fimbria, alveus and lacunar layer.

Acute cellular swelling was seen in the hepatocytes from all the livers that were examined. In some livers the periacinar hepatocytes had large intracytoplasmic vacuoles that stained negative for lipids, and resembled hyaline droplets. In 3 of the livers the spaces of Disse were conspicuously widened, which is compatible with perisinusoidal oedema (see Fig. 2).

No other histological lesions were seen which could be attributed to the treatment.

TABLE 1: DOSAGE USED, PERIOD FROM TREATMENT TO DEATH AND THE CLINICAL SIGNS IN CALVES INJECTED SUBCUTANEOUSLY WITH RAFOXANIDE SOLUTION

Animal No.	Dosage (mg/kg)	Period from treatment to death	Clinical signs*				
			Recumbancy	Polypnoea	Clonic muscle spasms	Frothing at the mouth	Blindness
14 calves	15-33	No signs seen					
Y 8	36	–	–	–	–	–	–
18	36	Euthanasia (7d)	+	–	–	–	–
30	39	–	–	–	–	–	–
39	39	–	–	–	–	–	–
45	42	–	–	–	–	–	–
96	42	Euthanasia (7d)	+	–	–	–	–
P 8	45	8d	+	–	+	–	–
107	45	48h	+	+	+	+	–
114	48	–	–	–	–	–	–
130	48	–	–	–	–	–	–
35	51	6d	+	+	–	+	+
124	51	42h	+	+	+	+	–
26	54	10d	+	–	–	–	–
32	54	Euthanasia (48h)	+	+	+	–	+
37	57	–	–	–	–	–	–
113	57	–	–	–	–	–	–
33	60	ca. 60h	+	+	+	–	–
117	60	26h	+	+	+	+	–

+ = Present, – = Absent
*Clinical signs of the 8/12 animals that died.

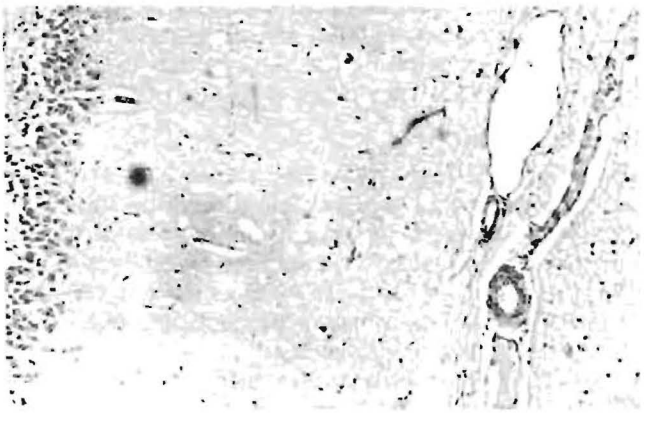

Fig. 1: Hippocampus from calf P8 showing status spongiosus and pericapillary oedema. Haematoxylin and Eosin stain. X100

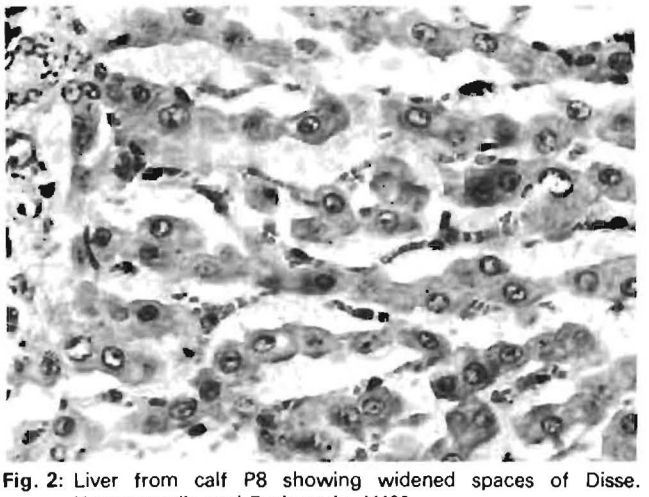

Fig. 2: Liver from calf P8 showing widened spaces of Disse. Haematoxylin and Eosin stain. X400

DISCUSSION

A variety of differential diagnoses exist for the range of clinical signs and gross lesions observed in this experiment. Those animals that were in lateral recumbency and paddled with their feet while displaying opisthotonus and nystagmus were very similar to "typical" cases of heartwater. The blindness and pronounced mydriasis, which are the most obvious signs of rafoxanide toxicity in sheep[3], are similar to descriptions of sheep suffering from progressive retinal degeneration ("bright blindness", thought to be caused by the plant *Pteridium aquilinum*[6]), or *Helichrysum argyrosphaerum* toxicity[3].

The best known differential diagnosis of the status spongiosus of the central nervous system in this country is *H. argyrosphaerum* toxicity in sheep[3]. This lesion may, however, also be caused by a variety of other toxic substances[5].

In view of the list of possible differential diagnoses, it is advisable that any diagnosis of rafoxanide toxicity be confirmed by chemical assay for plasma rafoxanide concentrations[5]. Such assays were not performed on any of the animals in this experiment.

The overall impression one gets from the above descriptions of gross and microscopic lesions, is one of oedema, probably as a result of increased vascular permeability. Rafoxanide toxicity is associated with papilloedema and vacuolation of nervous tissue in dogs[1], sheep[3] and cattle. This oedema can possibly be explained by the fact that rafoxanide has been shown to inhibit adenosine triphosphatase (ATPase) production, and that a membrane-bound ATPase plays an important role in the active transport of ions and water across cell membranes[3].

With regard to the therapeutic index, the results of this experiment are in agreement with those published for sheep[2], where it was concluded that the toxic dose of rafoxanide by the oral route lies in the region of 15-20 times the therapeutic dosage, which is 7,5 mg/kg.

From the data in this trial it can be concluded that injectable rafoxanide solution possesses a wide safety margin in cattle. Cases of intoxication present clinical signs and lesions that are easily recognisable, but which should be confirmed through plasma rafoxanide assay[5].

ACKNOWLEDGEMENTS

The author is grateful to Prof. R.C. Tustin, Faculty of Veterinary Science, University of Pretoria for assistace in interpreting the microscopic liver lesion.

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GENETIC MARKERS IN SOUTH AFRICAN THOROUGHBRED STALLIONS

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ABSTRACT: Osterhoff, D.R.; Le Grange, L.; Robinson, M. **Genetic markers in South African Thoroughbred stallions.** *Journal of the South African Veterinary Association* (1982) 53 No. 1 33-36 (En) Department of Zootechnology, Faculty of Veterinary Science, University of Pretoria, P.O. Box 12580, 0110 Onderstepoort, Republic of South Africa.

Genetically controlled markers are ideal for the identification of individual animals, and throughout the world laboratories have been established whose chief function is to provide a blood-typing service for animals including horses.

In order to achieve the aim of improved recording of foals almost all South African sires at stud were tested and their blood type identification completed. The genetic markers included in this survey were 14 blood group factors, transferrin, plasma esterase, haemoglobin, carbonic anhydrase, 6-phosphogluconate dehydrogenase, phosphoglucomutase and phosphohexose isomerase. Gene frequency calculations were performed and comparisons made with similar surveys in Thoroughbreds overseas.

The results indicate that the strict selection for speed in Thoroughbred racing horses has resulted in a high degree of genetic uniformity between South African and overseas racing Thoroughbreds.

INTRODUCTION

Concurrent with advances in the field of horse breeding such as assessment of racing and reproductive performance and systematic progeny testing there has arisen the need for a reliable method of identifying individual animals. Errors in parentage records can occur extremely readily. There also may be deliberate falsification of records by unscrupulous dealers. It is therefore highly desirable that exact records of animals are kept. In this connection, blood typing cannot prove parentage, but it is a reliable method of excluding parentage. The determination of genetic markers of stallions is, however, not only of importance in parentage exclusions, but also in solving the serious problem of neonatal iso-erythrolysis (a new name for the well known haemolytic disease of the newborn).

Studies on blood groups and genetic blood protein polymorphisms in horses, commenced in our department in 1967 and a routine blood-typing test for the Jockey Club commenced at the beginning of 1978. This paper provides a summary of the results of the tests on Thoroughbred stallions that been performed to date.

MATERIALS AND METHODS

Blood amples from 400 Thoroughbred stallions at stud were supplied for blood typing by the Jockey Club. Veterinarians in the different regions of the country collected the blood and certified in each case that the correct horse was bled.

The blood samples were collected in isotonic citrate solution. Isotonic saline (0,91 % NaCl) was used in washing and suspending red cells and as diluent for serum. Saline agglutination tests were performed in parallel with lytic tests using absorbed rabbit complement undiluted.

The saline agglutination tests were performed at room temperature (22-28 °C) in acrylic plastic plates (Perspex) using 2 drops of serum to one drop of a 2,5 % suspension of red cells. The lytic tests were performed in the same manner except that one drop of complement was added to the 2 drops of serum just prior to the addition of red cells. Test results were recorded at ½, 1 ½ and 2 ½ hours after setting up the tests. Further details are given elsewhere[4 6].

The method of starch gel electrophoresis was used for the determination of blood protein and enzymes. Starch gels have high resolving power and separation of protein molecules on these is based on electrophoretic mobility of the proteins as well as on its molecular size. A difference in electrophoretic mobility can be the result of an amino acid substitution leading to a difference in the net charge of the molecule or to a change in the configuration of the molecule.

The procedure is for each genetic marker somewhat different[2 4]. Basically, 300 ml of the appropriate gel buffer was heated to 100 °C and added rapidly to a weighed amount of starch (Connaught, Toronto) suspended in 100 ml of cold gel buffer.

The resulting viscous mass was shaken vigorously, degassed with a vacuum pump and poured into the gel trays. Gel trays with inner dimensions of 20,4 × 13,6 × 0,5 cm were used and the above mixture was enough to fill 2 of these trays. Glass plates were placed over the hot mixtures and the gels left at room temperature to set and cool. After cooling the gels were cut across their width, the distance depending on the separation required. One drop of the serum or haemolysate specimens was absorbed onto pieces of Whatman 3M filter paper, cut to measure 0,7 × 0,4 cm, blotted and inserted into the gel. The gels were connected to the electrolyte buffer with paper wicks which consisted of 6 layers of strips of Whatman No 1 filter paper.

To prevent evaporation during the electrophoretic run the gels were covered with sheets of polythene. Wet cheese cloth were placed on the polythene and the electrophoresis conducted in a specially built cupboard with fans (to increase evaporation) or in a cold room at 4 °C.

After the completion of each run, the running time being dependent on the system under investigation, the gel was removed from the tray and placed on a cutting board. It was then sliced horizontally into two with a very thin fuse wire and stained. A 1 % amido black solution, dissolved in a mixture consisting of methanol : water : acetic acid (5 : 5 : 1 v/v) was used as a general protein stain. After approximately 15 seconds the excess dye was poured off and the gel destained with methanol : water : acetic (5 : 5 : 1 v/v). Three to four changes of the destaining solution were usually adequate.

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RESULT AND DISCUSSION

1) Blood groups

The results of blood-typing of the Thoroughbred stallions are compared with those of other breeds including American Shetlands and South African Endurance horses (Table 1). The latter comprized a variety of different breeds and crossbreeds, e.g. Arab, Nooitgedacht, Boerperd, etc.

Table 1: COMPARISON OF GENE FREQUENCIES OF BLOOD FACTORS IN S.A. THOROUGHBREDS, ENDURANCE RACE HORSES AND U.S.A. SHETLAND PONIES

Blood factors*	S.A. Thorough-breds (N = 400)	Endurance horses (Varying numbers)	U.S.A.** Shetland Ponies (N = 391)
Aa	0,98	0,93	0,53
Ab	0,84	0,27	0,58
Ac	0,18	0,02	0,18
Ca	0,88	0,57	0,88
Pb	0,25	0,05	0,09
Qa	0,79	0,33	0,52
Ua	0,39	—	0,53
Da	0,02	0,66	0,25
Db	0,33	0,32	—
Dc	0,73	0,88	—
Dd	0,61	0,64	—
De	0,29	0,50	0,23
Df	0,05	0,39	—
Ka	0,01	—	0,33

*Blood factors Aa, Ab, Ac, Ca, Pb, Qa, Ua are determined by lytic tests, the others by agglutination.
**Data of Suzuki[6].

In Table 2 the comparison with Thoroughbreds of other countries are depicted. Only 6 blood factors could be compared, but these present an excellent concordance in the frequency of blood factors.

Table 2: COMPARISON OF THE FREQUENCIES OF CERTAIN BLOOD FACTORS IN FOUR POPULATIONS OF THOROUGHBREDS

Blood factors	S.A. (N = 400)	U.S.A.* (N = 276)	France* (N = 162)	Poland* (N = 100)
Aa	0,98	0,93	0,79	1,00
Ac	0,18	0,01	0,00	0,00
Ca	0,88	0,93	0,94	0,93
Da	0,02	0,00	0,00	0,00
De	0,29	0,28	—	0,17
Ka	0,01	0,12	—	0,22

*Suzuki[6]

2) Transferrin

The gene frequencies of transferrins in South African Thoroughbreds and endurance horses are presented in Table 3.

Table 3: FREQUENCY OF TRANSFERRIN ALLELES IN HORSES

	No	Tf^D	Tf^F	Tf^H	Tf^M	Tf^O	Tf^R
Thoroughbreds	400	0,283	0,523	0,036	0,000	0,095	0,063
Endurance race horses	88	0,256	0,489	0,102	0,000	0,097	0,057

Frequencies of transferrin alleles in Thoroughbreds of different countries are compared in Table 4 and a good agreement of frequencies exists. It seems that strict selection for performance shaped the gene frequencies in these countries in a similar way. Fifteen different transferrin phenotypes are shown in Fig. 1.

Table 4: FREQUENCY OF TRANSFERRIN ALLELES IN THOROUGHBREDS OF DIFFERENT COUNTRIES

Country	No	Tf^D	Tf^F	Tf^H	Tf^O	Tf^R
Belgium[1]	333	0,314	0,488	0,066	0,090	0,042
Netherlands[1]	35	0,300	0,442	0,043	0,129	0,086
U.S.A.[1]	150	0,267	0,563	0,027	0,090	0,053
Hungary[5]	208	0,313	0,471	0,019	0,101	0,096
France[2]	182	0,32	0,47	0,07	0,09	0,05
Sweden[2]	65	0,231	0,639	—	0,076	0,054
South Africa (1973)	337	0,261	0,551	0,042	0,074	0,072
South Africa (1979)	400	0,283	0,523	0,036	0,095	0,063

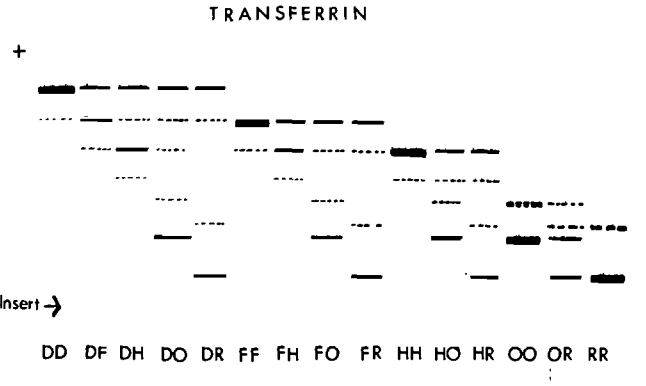

Fig. 1: Transferrin phenotypes in S.A. Thoroughbreds.

3) Albumin

There is a concordance in the frequency of albumin alleles in different Thoroughbreds though they differ considerably from endurance horses (Table 5).

Table 5: FREQUENCY OF ALBUMIN ALLELES IN HORSES

Country	No	$A1^A$	$A1^B$
Th. Netherlands[1]	40	0,29	0,71
Th. U.S.A.[1]	145	0,21	0,79
Th. France[3]	182	0,27	0,73
Th. South Africa	400	0,24	0,76
End. South Africa	90	0,48	0,52

Th. = Thoroughbreds End = Endurance race horses

Three different phenotypes can be identified in Fig. 2.

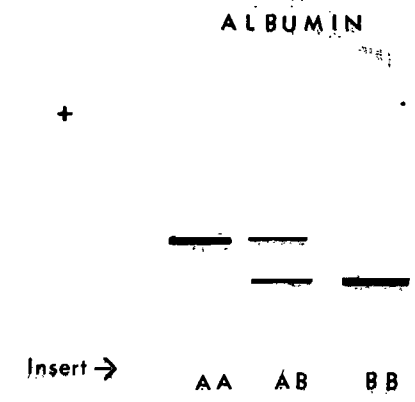

Fig. 2: Albumin phenotypes in S.A. Thoroughbreds.

4) Plasma esterase

The frequency of plasma esterase alleles (Table 6) show relatively small differences.

Table 6: FREQUENCY OF PLASMA ESTERASE ALLELES

Country	No	Es^I	Es^S	Es^F	Es^G
Th. Netherlands[1]	40	0,92	0,04	0,04	0,00
Th. South Africa	400	0,88	0,06	0,06	0,00
End. South Africa	82	0,90	0,00	0,05	0,05

Th. = Thoroughbreds End = Endurance race horses

Six different plasma esterase phenotypes are depicted in Fig. 3.

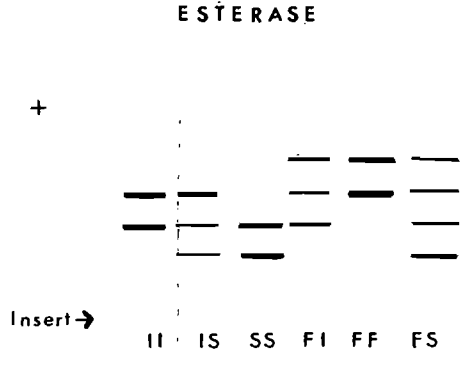

Fig. 3: Esterase phenotypes in S.A. Thoroughbreds.

5) Haemoglobin

There was no haemoglobin polymorphism in Thoroughbreds (Table 7), which is in agreement with previous findings[1,4].

Table 7: FREQUENCY OF HAEMOGLOBIN ALLELES

Country	No	Hb^A	Hb^a
Th. Netherlands[1]	40	1,0	0,0
Th. South Africa	400	1,0	0,0
End. South Africa	86	0,95	0,05

Th. = Thoroughbreds End = Endurance race horses

6) Carbonic anhydrase

The frequency of carbonic anhydrase alleles are also very similar (Table 8).

Table 8: FREQUENCY OF CARBONIC ANHYDRASE ALLELES

Country	No	Ca^F	CA^I	CA^L CA^O CA^S
Th. Netherlands[1]	40	0,08	0,92	0,0
Th. South Africa	400	0,04	0,96	0,0
End. South Africa	87	0,04	0,92	0,04

Th. = Thoroughbreds End. = Endurance race horses

Clear differences in the carbonic anhydrase phenotypes can be seen in Fig. 4.

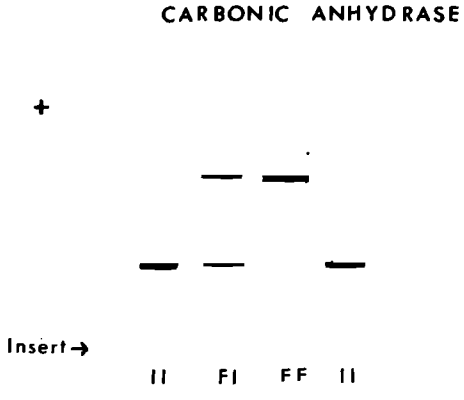

Fig. 5: Carbonic anhydrase phenotypes in S.A. Thoroughbreds.

7) 6-Phosphogluconate dehydrogenase

The 6-phosphogluconate dehydrogenase plays an important rôle in the hexose monophosphate shunt pathway. The oxidative decarboxylation of 6-phosphogluconate dehydrogenase (6-PG) to ribulose 5-phosphate (R 5P) is catalyzed by 6-phosphogluconate dehydrogenase (6-PGD), generating a second molecule of nicotinamide adenine dinucleotide phosphate (NADP). This enzyme, has been found to vary both in electrophoretic pattern and catalytic activity. There were only minor differences between South African and French Thoroughbreds and Endurance race horses in the 6-PGD systems (Table 9).

Table 9: FREQUENCY OF 6-PGD ALLELES

Country	No	$6\text{-}PGD^F$	$6\text{-}PGD^S$
Th. France[3]	182	0,59	0,41
Th. South Africa	398	0,61	0,39
End. South Africa	90	0,67	0,33

Th. = Thoroughbreds End. = Endurance race horses

Three 6-PGD phenotypes can clearly be seen in Fig. 5.

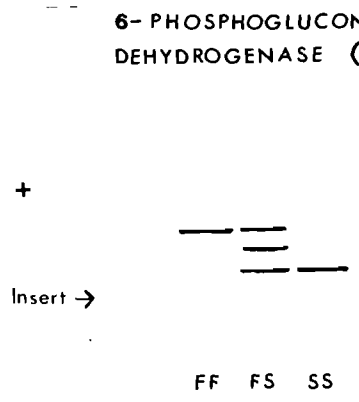

Fig. 5: 6-Phosphogluconate dehydrogenase phenotypes in S.A. Thoroughbreds.

8) Phosphoglucomutase

Phosphoglucomutase is a phosphotransferase which catalyses the conversion of glucose-1-phosphate to glucose-6-phosphate in the presence of catalytic amounts of glucose 1,6-diphosphate. The frequency of the F alleles seems to be slightly higher in endurance horses than in the Thoroughbreds (Table 10). No homozygous F-animals were found.

Table 10: FREQUENCY OF PGM ALLELES

Country	No	PGM^F	PGM^S
Th. France[3]	182	0,0	1,0
Th. South Africa	399	0,007	0,993
End. South Africa	87	0,063	0,937

Th. = Thoroughbreds End. = Endurance race horses

In Fig. 6 three phenotypes of phosphoglucomutase are depicted.

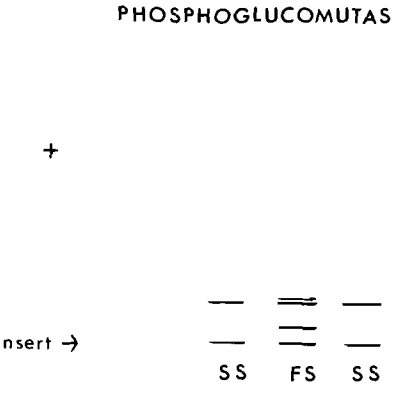

Fig. 6: Phosphoglucomutase phenotypes in S.A. Thoroughbreds.

9) Phosphohexose isomerase

Phosphohexose isomerase (PHI) also known as phosphoglucose isomerase or glucose phosphate isomerase catalyses the reversible conversion of glucose-1-phosphate to fructose-6-phosphate. There is in Thoroughbreds no polymorphism in the PHI system (Table 11).

Table 11: FREQUENCY OF PGI ALLELES

Country	No	PGI^I	PGI^F
Th. France[3]	182	1,00	0,00
Th. South Africa	399	1,00	0,00
End. South Africa	90	0,99	0,01

Th. = Thoroughbreds End. = Endurance race horses

In Table 12, a comparison is made between different breds and types of horses, including mules, with regard to those alleles which according to a previous study[4], were most frequent in Thoroughbred winners: Tf^F, Al^B, Est^I and $6\text{-}PGD^S$. For most of these alleles there is an increased frequency from mules to "common" horses, riding horses, endurance horses through to Thoroughbreds.

Table 12: GENE FREQUENCY CHANGES DUE TO SELECTION IN HORSES

Breed/ Type	Mules	Common Horses	Riding Horses	Endurance Horses	Thoroughbred		
					General	Winners	Stallions
No. of Animals	110	288	154	90	443	206	400
Marker							
Tf^F	0,26	0,48	0,48	0,49	0,55	0,55	0,52
Al^B	0,27	0,52	0,63	0,52	0,72	0,66	0,76
EST^I	0,75	0,82	0,84	0,90	0,83	0,95	0,88
$6\text{-}PGD^S$	0,10	0,16	0,13	0,33	0,41	0,39	0,39

The significance of these findings will be investigated in future work including other enzymes and possibly also leucocyte antigens. There is no doubt that from the practical point of view enough variation is present to provide a service to horse breeders with regard to parentage determination.

Future research should concentrate on those systems which are directly involved with the physiological and biochemical processes, and could provide new parameters indicative of the performance ability of the equine.

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BIOCHEMICAL POLYMORPHISMS IN THE SOUTH AFRICAN SPRINGBOK (*ANTIDORCAS MARSUPIALIS*)

D.R. OSTERHOFF*, SANITA M. SCHOEMAN** and A. CONROY***

ABSTRACT: Osterhoff, D.R.; Schoeman, S.M.; Conroy, A. **Biochemical polymorphisms in the South African Springbok (*Antidorcas marsupialis*).** *Journal of the South African Veterinary Association* (1982) **53** No. 1 37-39 (En) Department of Zootechnology, Faculty of Veterinary Science, University of Pretoria, 0110 Onderstepoort, Republic of South Africa.

An attempt was made to use polymorphic markers in the genetic taxonomy of springbok populations. Three independent free ranging springbok populations could be identified on the basis of albumin and transferrin gene frequencies. Even if most of the enzyme markers appeared to be monomorphic it can be stated that in any comparative study of this kind, cryptic genetic markers are of greater use than other environmentally sensitive taxonomic criteria. From the given data and others mentioned it can be stated that little support exists for the continued application of the subspecific status in the species *Antidorcas marsupialis*.

It is shown that the use of environmentally sensitive parameters in the springbok are both misleading and highly unsatisfactory. Biochemical polymorphism can be utilized better than other parameters but here also there are several limitations. Basically, only the albumin and transferrin types can be used when blood samples are available and only IDH and 6-PGD polymorphisms are of value when liver samples can be utilized for the genetic differentiation.

INTRODUCTION

Generally, polymorphism is the simultaneous occurrence of 2 or more varieties in the same population at the same time in such proportions that the rarest cannot be maintained by mutation alone. Biochemical polymorphism has been established in zebra[1], white rhino[5] and in gerbil[3]. It has not been found in the black rhino[5] and in blesbok[6].

In this study an attempt has been made to identify blood substances displaying polymorphic characteristics in the South African springbok in order to be able to differentiate between populations. Although the present taxanomy allows for 3 subspecies (*Antidorcas marsupialis marsupialis, A.m. hofmeyri* and *A.m. angolensis)* their identification and distribution is vague.

MATERIAL AND METHODS

As *A.m. marsupialis* occurs most widely and is thus the most freely available of the subspecies in Southern Africa it was chosen for studying genetic markers in the blood of springbok. In a search for the best genetic markers to differentiate between subspecies, liver samples were collected from all 3 subspecies.

The blood samples were obtained on 3 privately owned game farms in the Karoo, Cape Province. The springbok populations (A, B and C) on each farm had been isolated from other springbok in their respective areas for over 30 years. The animals were shot at night and immediately thereafter blood was collected into tubes containing EDTA. The following morning, the samples were dispatched by train to the laboratory. The number of samples collected were: Population A: 244 samples; Population B: 75 samples and Population C: 19 samples.

In all, 91 liver samples were obtained from a variety of localities, some of them from South West Africa, Angola and Botswana, but also from the farm mentioned above.

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Biochemical markers were studied by starch gel electrophoresis in serum, erythrocyte haemolysates and liver homogenates. In blood, the systems included albumin, transferrin, haemoglobin, 6-phosphogluconate dehydrogenase (6-PGD), phosphoglucomutase (PGM), phosphoglucose isomerase (PGI), catalase and diaphorase. In liver homogenates, the isoenzymes of isocitrate dehydrogenase (IDH), 6-PGD and sorbiton dehydrogenase (SDH) were analysed. Details of the methods on electrophoresis of blood and liver homogenates are reported elsewhere[2 3 10].

RESULTS AND DISCUSSION

Considerable polymorphic variation was found in the following systems which proved to be most useful in the general characterization of springbok populations.

Albumin

Five phenotypes were distinguishable from banding patterns and mobility; originally only the allels Al^A and Al^B were seen with the A-band migrating further than the B-band. Only in later investigations a C-band was found which migrated faster than the A-band (Fig. 1). Electrophoresis was repeated on all samples but in only 11 samples was a C-band established.

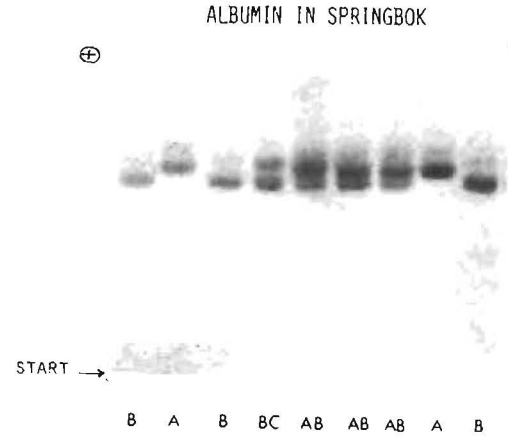

Fig. 1: Albumin types in springbok.

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Table 1 depicts the distribution of albumin types in three populations of springbok.

Table 1: DISTRIBUTION OF ALBUMIN TYPES IN SPRINGBOK

Population	No investigated	Phenotypes					Gene frequency		
		AA	BB	AB	AC	BC	Al^A	Al^B	Al^C
A	224	29	108	101	1	5	0,33	0,66	0,01
B	75		70			5	0,00	0,97	0,03
C	19		19				0,00	1,00	0,00

In Table 2 the χ^2 test for genetic equilibrium is presented.

Table 2: THE χ^2 TEST FOR EQUILIBRIUM IN THE ALBUMIN SYSTEM IN ONE LARGE POPULATION OF SPRINGBOK

Albumin	Obs	Exp	d	d^2/exp
AA	29	26,57	−2,43	0,22
AB	101	106,29	+5,29	0,26
AC	1	1,61	+0,61	0,23
BB	108	106,29	−1,71	0,23
BC	5	3,22	−1,78	0,98
CC	0	0,02	−0,02	0,02
χ^2 3 df				1,94 not significant

Although the homozygous C-phenotype was not found, the expected frequencies were in close agreement with the observed values in the largest springbok population (A), indicating that we are dealing with a balanced population. The populations B and C were actually too small to speculate about their genetic equilibrium. Nevertheless, it seems that a high degree of inbreeding has taken place, since the Al^A-allele is not at all present in populations B and C and the polymorphism in the albumins has thus been drastically diminished.

Transferrin

The transferrin results are depicted in Table 3. Altogether 11 phenotypes were found which could be classified as being dependent on 6 different alleles – Tf^A, Tf^C, Tf^E, Tf^F and Tf^H. The bands (fig. 2) resulting from the allelic functions were named from the fastest moving band A to the slowest band H. Intermediate bands B and G have not yet been found; likewise not all possible phenotypes of the alleles given have been seen yet.

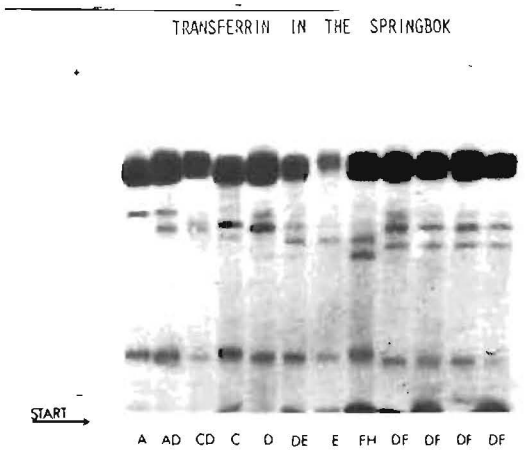

Fig. 2: Transferrin types in springbok.

Table 3: DISTRIBUTION OF TRANSFERRIN TYPES IN SPRINGBOK

Population	No investigated	Phenotypes									
		AA	CC	DD	EE	AC	AD	CD	DE	DF	EH	FH
A	246		6	176	3		11	44	2		1	
B	75			67				6		2		
C	19	3	2	9		1	1		1		1	1

The differences in the gene frequencies between the 3 populations can clearly be seen in Table 4.

Table 4: TRANSFERRIN GENE FREQUENCIES IN SPRINGBOK

Population	Gene frequency					
	Tf^A	Tf^C	Tf^D	Tf^E	Tf^F	Tf^H
A	0,022	0,114	0,844	0,018	0,000	0,002
B	0,000	0,040	0,947	0,000	0,013	0,000
C	0,211	0,132	0,526	0,053	0,027	0,053

Haemoglobin

Only one type of haemoglobin was found. It is of great interest to note that in several antelope species like impala, haemoglobins can be very useful as gene markers and in other species only a monomorphic haemoglobin situation exists.

Red cell enzymes

Most of the red cell enzymes were monomorphic providing very little use as gene markers in population studies. Table 5 lists the red cell enzymes investigated.

Table 5: RED CELL ENZYMES AS GENETIC MARKERS IN SPRINGBOK

Enzyme	Remarks
6 – PGD	1 band: no variation
PGM	2 bands: no variation
PGI	2 bands: no variation
Catalase	1 band: no variation
Diaphorase	2 bands: no variation

Isocitrate dehydrogenase

The IDH patterns found in the liver homogenates indicate that this enzyme is probably a dimer which is made up of subunits coding for the same structural gene.[10] The enzyme from the homozygotes AA and A′A′ appears as a single band on the zymogram (fig. 3). In a dimeric model 3 bands are expected in the heterozygous form, one representing the normal and the variant gene respectively and the intermediate band consisting of a normal and variant subunit (Fig. 3).

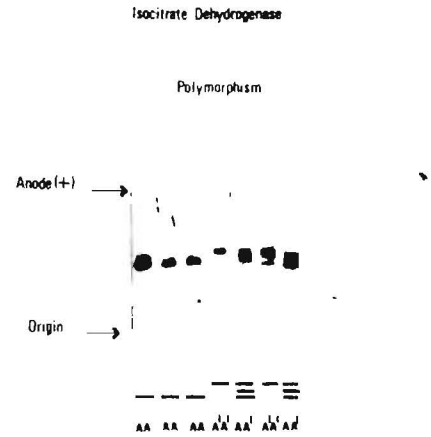

Fig. 3: Isocitrate dehydrogenase in springbok.

6-Phosphogluconate dehydrogenase

Similar patterns to those obtained for the dimer IDH were observed. The single migrating bands AA and A′A′ represent the homozygous forms consisting of identical subunits, while the intermediate patern AA′ is formed by the heterozygote (Fig. 4).

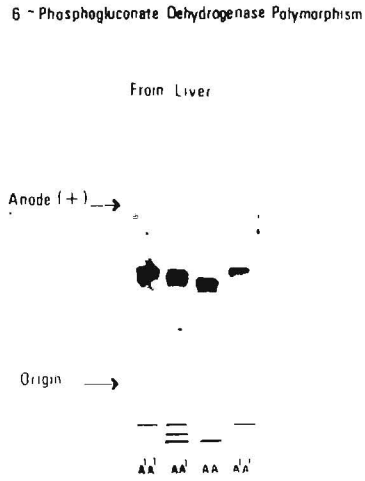

Fig. 4: 6-Phosphogluconate dehydrogenase in springbok.

In Table 6 the test for genetic equilibrium of IDH and 6-PGD alleles is depicted in the material which could be used for gene frequency analysis. Both genetic systems can be used for population gene frequency studies, since the actual frequency shows a very useful distribution: $IDH^A - 0,55$; $IDH^{A'} - 0,45$ and $6\text{-}PGD^A - 0,66$; $6\text{-}PGD^B - 0,34$.

Table 6: TEST FOR GENETIC EQUILIBRIUM OF IDH AND 6-PGD-ALLELES

System	Phenotypes						χ^2
	AA		A'A		A'A'		
	Obs	Exp	Obs	Exp	Obs	Exp	
IDH	34	28,02	31	44,94	25	18,02	8,77*
6 PGD	55	40,22	11	40,56	25	10,22	48,32*

*$P < 0,01$

An excess of homozygotes is illustrated which can be explained by the fact that most of the liver samples came from isolated populations. Here, a very high degree of homozygosity could be expected as reflected in the very low number of heterozygotes in Table 6.

Sorbitol dehydrogenase

This system was also studied in liver homogenates, but proved to be monomorphic. This system also did not provide useful genetic markers for taxonomic studies.

It would seem that the differences between the 3 populations under study are as great as the differences between the so-called subspecies. Attempts to substantiate the subspecies status of the springbok was hampered by the difficulty to obtain a greater number of samples of animals in other habitats.

Also in earlier attempts[1,9] no conclusive evidence could be provided for the subspecies differentiation in *Antidorcas marsupialis*. The size of the springbok was studied[9] and it was established that the protein content of the preferred plants eaten was responsible for differences in growth and final body size. Rumen samples were analysed to study the preferred plants.

Also a dermatological study[1] including hair analysis by stereo-microscopy could not prove subspecies differences. Not even dermatological differences between the common, black and the white springbok except for the hair colour itself could be found.

From the presented data and those mentioned from other authors it can be stated that little support exists for the continued application of the subspecific status in the species *Antidorcas marsupialis*.

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SURGICAL APPROACH TO THE ROSTRAL CRANIAL FOSSA BY RADICAL TRANSFRONTAL CRANIOTOMY IN THE DOG*

P.D. DE WET†, I.I. ALI†† and D.N. PETERS†††

ABSTRACT: de Wet P.D., Ali I.I., Peters D.M. **Surgical approach to the rostral cranial fossa by radical transfrontal craniotomy in the dog.** *Journal of the South African Veterinary Association* (1982) 53 No. 1 40-51 (En) Department of Veterinary Anatomy, College of Veterinary Medicine, Ohio State University, Columbus, Ohio 43210, USA.

To overcome the problems of restricted visibility and surgical mobility, as well as those posed by anatomical features of the canine subfrontal sinus region and associated dura mater, aggravated by hazards of infection from potentially infected frontal sinuses, of uncontrollable haemorrhage and post-operative brain compression, a radical approach was devised, combining invasion of the frontal sinus with a lateral rostrotentorial craniotomy. This offers adequate surgical manoeuvring space, excellent visualisation of the rostral cranial fossa and its contents, proper asepsis, minimal haemorrhage and no untoward after-effects. The technique lends itself to diverse neurosurgical applications in the rostral cranial fossa of the dog.

Particulars of instrumentation, and pre-operative, operative and post-operative procedures, done on 3 Beagles with excellent results, are described.

INTRODUCTION

With the advance of veterinary neurology, the development of reliable techniques for intracranial neuro surgery becomes more imperative. The most common indications for such neuro surgery include intracranial tumours, compressive skull fractures, infection, subdural haematomas[17 18] and behavioural disturbances[6 23], yet there is a paucity in veterinary literature concerning techniques for adequate exposure of the olfactory bulb and frontal lobe regions. Any exposure of this area involves invasion of the frontal sinus.

The results of the "blind" transfrontal burr hole approach to perform prefrontal lobotomies to alleviate or alter canine and feline over-aggressiveness, destructive behaviour, fear-biting and self-mutilation from any cause[6], thus far have been unpredictable[23]. It may be noted in passing that similar behavioural disturbances were reported in dogs with laceration and scarring of the basilar region of the frontal lobes, often accompanied by profuse haemorrhage in the rostral cranial fossa owing to accidental fractures of the floor of this fossa[13]. Other workers modified the technique by using a limited dorsal transfrontal sinus craniectomy but the results obtained were equally unreliable[1], such as: persistent abnormal behavioural traits, motor deficits caused by accidental surgical invasion of basal nuclei and internal capsules, seizures from contracting scar tissue, and death from uncontrolled brain haemorrhage. Consequently it was concluded that surgery should be reserved only as a last resort[1 23].

In man, treatment of anterior communicating aneurysms in the rostral cranial fossa has led to accidental occlusion of the recurent artery of Heubner resulting in hemiparesis with brachial dominance, aphasia, and paralysis of the face, palate and tongue[4]. In addition, lesions in the anterior hypothalamic region irrigated by the anterior cerebral circulation (A-I segment[19]) have caused alterations of emotional behaviour, personality disorders and intellectual deficits, which may be ascribed to occlusion of the arterial supply to this area[16].

Others have elaborated in detail upon the difficulties, dangers and risks of a surgical approach to the frontal lobe cortex via the frontal sinus and upon means to minimise these. In their patients the transfontal sinus craniotomy (or craniectomy) was limited to the development of a bone flap 20 mm wide which extended from the floor of the sinus to 10-20 mm caudally into the frontal bone; this resulted only in a limited exposure of the dorsolateral surface of the frontal lobe[18]. These limited exposures of the frontal lobes (prefrontal cortices) are hardly conducive to precise surgical or microsurgical intervention involving the entire extent of the frontal lobes and rostral cranial fossa.

Not only are these limited transfrontal craniectomies or craniotomies inadequate for exploration of the rostral cranial fossa, from lateral as well as midline approaches, but they also severely restrict visibility and surgical or microsurgical mobility between, and along, the ventrolateral surfaces of the frontal lobes. Additionally, accidental haemorrhage from the frontocortical vessels and the rostral cerebral, rostral communicating and recurrent artery complex, located between the lobes and on the floor of the fossa, was difficult to control owing to the inadequate visualisation of these areas. Moreover, owing to inadequate exposure, blind bilateral ablations (lobectomy), or leucotomies of the frontal lobes (prefrontal cortices) to alter aggressive behaviour[1 23] may contribute to disappointing results.

The complex structural and functional interrelationship which exists between the frontal lobe cortices and thalamo-hypothalamic nuclear complex, which serves as the seat for the highest control of all shades of emotional and motivational behaviour, personality and intellect[21], warrants a more sophisticated surgical, or, preferably, microsurgical approach, as well as effective control of haemorrhage in the rostral cranial fossa to prevent a wide spectrum of variations in the results and/or serious complications.

Neurosurgical techniques, especially those involving the frontal sinus, which is a potent source of infection, require the utmost attention to asepsis and haemoatasis[9 17 18]. Such techniques and patient care have been described[9 8 15 24].

This project was undertaken as a research endeavour to examine the complex structure and function of the

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basilar region of the frontal lobes in the dog, and to evaluate the feasibility at combining, in a radical approach, the invasion of the frontal sinus with a lateral rostrotentorial craniotomy. We have endeavoured to assess the feasibility of the latter surgical application through evaluation of several techniques special to this type of procedure. These include closure of the frontal sinus, use of a fascial graft as a means of providing a watertight dural closure, and the use of a "floating" bone flap as a means of combating post-surgical complications owing to cerebral oedema.

At all times a good cosmetic appearance was an important aim. Pre- and post-operative radiographs and photographs were used to evaluate this.

We believe that the adaptability of this technique provides for a multitude of possible surgical applications, including removal of tumours and haematomas involving both the frontal lobes of the cerebral hemisheres and rostral half of the cerebral arterial circle[11] (Circle of Willis), pericallosal vessels, and midline and basilar regions of the frontal lobes, with possible research applications to narrow the range of so-called idiopathic neuropathies.

This paper is presented in 2 sections.

Section I deals briefly with the applied anatomy, consideration of which supplied the data required before surgical intervention could be undertaken. Section II concerns the surgical procedure.

SECTION I: APPLIED ANATOMY

MATERIALS AND METHODS

Anatomical Preparations

Seven fresh canine cadavers were prepared for dissection of which four were injected with red-coloured latex via both carotid arteries. The anatomy of the rostral cranial fossa and its contents, especially the arterial supply and venous drainage in this region, was reviewed by dissection and observed with a surgical microscope.

In all instances the surgical approach to this region was simulated on these cadavers to establish basic techniques, to train assistant surgical personnel, and to design special surgical instruments necessary for such an approach and for microsurgical intervention in this region.

RESULTS

Surgical mobility is severely restricted, particularly in the subfrontal region of the neurocranium, by the close proximity of the reflected temporalis muscle mass, and by the orbital cone further rostrally, in spite of the additional space obtained by severing the zygomatic arch[15]. Also, in the subfrontal sinus region the somewhat acute rostro-ventral slanting of the floor (inner table – lamina interna – of the frontal bone) of the frontal sinus in some breeds, the rather snug fitting of the frontal lobes in the rostral cranial fossa, the delicate nature of the dura mater in this region, and tight adherence of the dura to the inner table of the neurocranium in older dogs, contribute further to surgical difficulties in this region. In addition, the sulcus for the dorsal sagittal venous sinus, located in a median groove of the inner table beneath the frontal sinuses, increases in depth with age. Furthermore, in older dogs, the increased prominence of the transversely located inner table and the bony ridges (cerebral juga[5]) which oppose the presylvian, cruciate and ansate sulci of the frontal lobe cortex present a transverse barrier, causing difficulty in separation of the dura in a caudorostral direction along the paramedian frontal region. The sunken position of the dorsal sagittal sinus and cerebral juga contributes further to surgical obstacles to develop a complete, large, intact, frontal bone flap without damage to the dura and the brain in this region.

SECTION II: SURGICAL PROCEDURE

MATERIALS AND METHODS

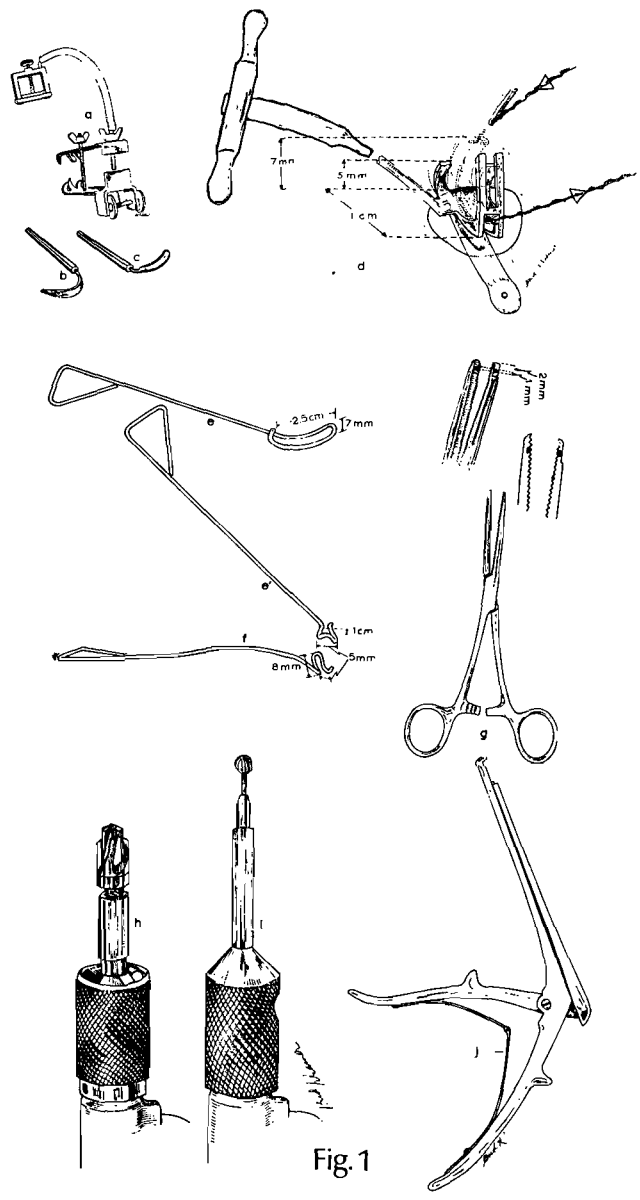


Fig. 1: *a. de Wet self-stabilising retractor[9]; *b. de Wet Zygomatic Gigli wire hook[9] *c. de Wet subtemporal dural elevator[9]; *d. Gigli saw sliding guide; *e,e'. subfrontal sinus dural elevators; *f. subfrontal dural elevator; *g. drill guide and dural protector (modification of the human Adson drill guide and dural protector); h. Smith automatic perforating drill; i. burr adapter and spherical burr; j. Kerrison laminectomy rongeurs.

*The experimentally designed instruments were constructed from miscellaneous hardware, haemostats and laboratory dissection probes.

Experimental Animals

Three Beagle dogs (mesocephalic heads) ranging in mass from 11,36 to 15,9 kg, and aged 11 months, 5 years, and 12 years, were used.

Instrumentation

In addition to routinely used neurosurgical equipment, some essential micro- and ophthalmic surgical instruments were acquired. Special neurosurgical instruments were designed or acquired and were specifically adapted to the unique and surgically difficult topographical anatomy of the frontal region of the neurocranium of the dog.

The following experimentally designed* or acquired neurosurgical instruments (Fig. 1) were used to overcome most of the above-mentioned difficulties;

*Self-stabilising retractor[25]
*Zygomatic Gigli wire hook[25]
*Subtemporal dural elevator[25]
*Gigli saw sliding guide
*Subfrontal sinus dural elevators
*Subfrontal dural elevator
*Drill guide and dural protector (a modification of the human Adson drill guide and dural protector)
Codman and Shurtleff variable speed motor drive[a]
Smith automatic perforating drill[b]
Burr adaptor[c] and various sized and shapes of burrs[d].
Kerrison laminectomy rongeurs.

Presurgical Treatment

Animals were prepared for surgery using the following schedule. Preoperative radiography of the head was found necessary to study the extent of the frontal sinuses. The dogs received 40 u Azium[e], 30 000 u procaine penicillin intramuscularly and 50 mg/kg chloramphenicol[f] every 8 h orally 24 h prior to surgery.

Anaesthesia and Treatment during Surgery

The dogs received injections of 0,005 mg/kg atropine and 0,05 mg/kg acetyl promazine before being anaesthetized. General anaesthesia was induced by the intravenous administration of 2 mg/kg sodium thiamylal[g] and maintained with methoxyflurane[h] and nitrous oxide on a standard inhalation anaesthetic machine. The bladder was catheterised to facilitate emptying following mannitol[i] therapy. The vital signs were continuously monitored electrocardiographically, by telethermometer and by oesophageal stethoscope. Automatically controlled heating pads were placed beneath the patient to control body temperature when needed. At the time of the skin incision 500 ml of a 25 % mannitol solution[i] (60 drops/min) was administered intravenously. Lactated Ringer's solution was then administered for the duration of the surgery (± 5-7 h). Antibiotic therapy was continued during surgery with chloramphenicol[f] intravenously and procaine penicillin intramuscularly.

Surgical Procedure

The animal was placed in sternal recumbency and its head secured in an adjustable head holder to provide an unrestricted arterial supply and venous drainage, to minimise the possibility of air embolism, and also for easy positioning of the surgical microscope and for microsurgical intervention (Fig. 2). The presurgical preparation of the head and manipulation of the jaw and ears has been described[9 25] previously.

The skin incision was carried rostrodorsally from the lateral palpebral commissure of the eye, and curved about 2 cm behind the dorsal palpebral margin to terminate on the midline between the medial palpebral commissures of the eyes. The incisions did not extend through the subcutaneous tissue, to prevent severing the frontal and palpebral nerve branches[11] of the rostral auricular plexus which innervate the muscles of the superior and inferior eyelids. Caudally, the incision followed the midline of the head (external sagittal crest) as far as the external occipital protuberance. The incision was then carried ventrolaterally along the dorsal nuchal line (nuchal crest) to the suprameatal spine just dorsal to the base of the ear (Fig. 2). In reflecting the skin flap, care was taken not to invade the superficial fascia and muscles over the vertex of the head or to disturb the closely related rostral auricular nerve plexus, then exposed in its entirety.

The skin flap was reflected latero-ventrally, the de Wet self-stabilising retractor[25] positioned on the right zygomatic arch (Fig. 3) and the left zygomatic arch transected rostrally, as had been reported previously by us[25] (Fig. 4). A superficial fascia and combined frontalis and interscutularis muscle flap was outlined and developed, sparing the rostral auricular plexus on the lateral side (Fig. 5). The muscle flap was reflected over the midline to the right side in a rostrolateral direction, and weighted down and wrapped in surgical sponges[j] soaked in physiological saline. This exposed the roof of the frontal sinus on the left side. The auricular and the rest of the interscutularis and frontalis muscles were then incised 3 mm from the external sagittal crest, reflected ventrolaterally and wrapped in similar fashion as the previously mentioned muscle flap. This exposed the superficial temporal artery, vein and zygomatic process of the temporal bone (Figs. 6, 6a). The artery was ligated between double ligations (Fig. 6) and the process cut in the same manner as reported[9 25] (Fig. 6a). The exposed temporalis muscle was cut and separated from the neurocranium with a periosteal elevator and reflected and treated as previously described[15 25]. Further rostral and ventral separation was undertaken, if necessary, on a horizontal line just dorsal to the optic canal, orbital fissure, rostral alar foramen, rostral border of the zygomatic process of the temporal bone and suprameatal spine further caudally. The muscle was also separated from the external crest, orbital ligament and periorbita as far ventromedially as the dorsal border of the pterygopalatine fossa by undermining the frontalis muscle. In doing so, the frontal and palpebral nerve branches[11], which are closely associated with the frontalis muscle in this region, were spared. The reflected temporalis muscle was forcefully retracted in a ventral direction and stabilised with several towel clamps and elastic bands hooked onto the base of the head holder.

The head was rotated 45° away from the surgeon. This further enhanced separation of the temporalis muscle, thus giving a better exposure of the orbital cone (periorbita) and subfrontotemporal region, and facilitating surgical mobility further ventrally along the neurocranium.

The orbital cone was then gently retracted laterally in the orbital fossa with a curved brain retractor[k] or finger rake retractor to expose the external ethmoidal artery, vein and ethmoidal nerve, ensheathed by periosteoperiorbita[5], as they emerge from the medial surface of the cone to enter the ethmoidal foramina in the medial wall

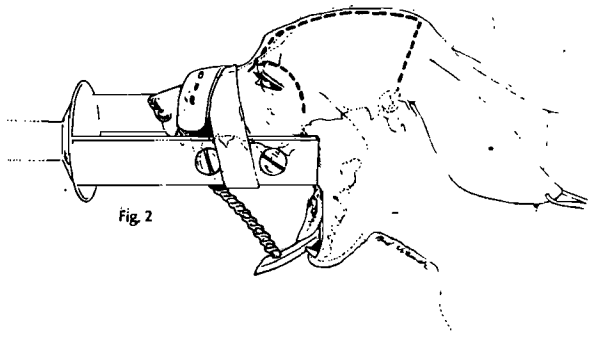


Fig. 2

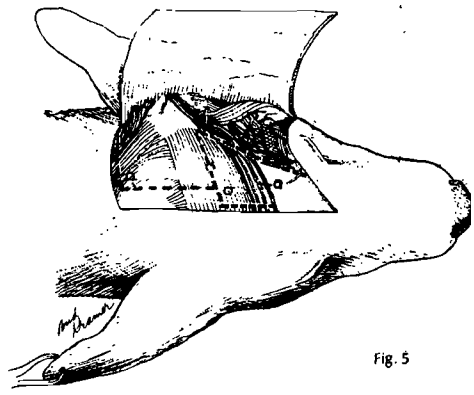


Fig. 5

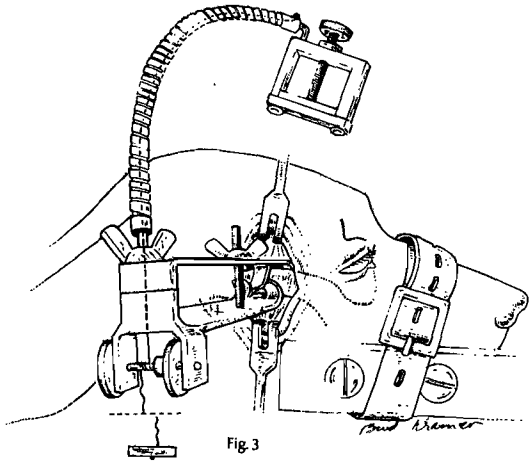


Fig. 3

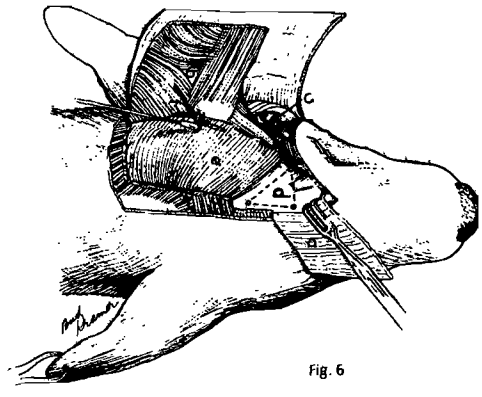


Fig. 6

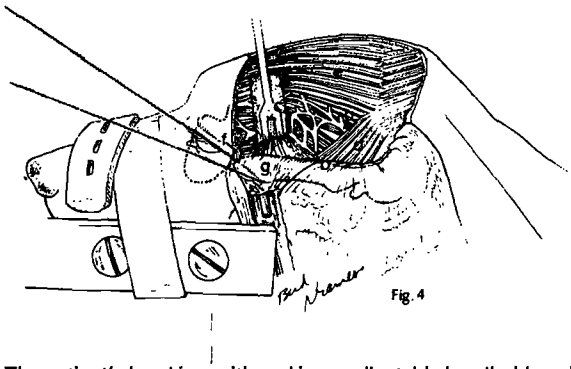


Fig. 4

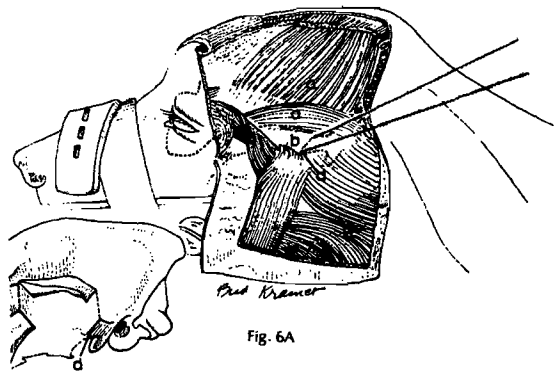


Fig. 6A

Fig. 2: The patient's head is positioned in an adjustable headholder with the mandible widely retracted, the ears secured caudally. The skin incision is outlined by a broken line.

Fig. 3: The de Wet self-stabilising retractor is placed on the contralateral zygomatic arch, just caudal to the orbital ligament. The flexible arm remains out of the surgical field until required.

Fig. 4: Gigli saw wire in position for osteotomy of the zygomatic bone, just caudal to the orbital ligament. Superficial muscles and nerves.

a. rostral auricular branch (rostral auricular n.)
b. zygomatic branch (palpebral n.)
c. rostral auricular nerve plexus
d. sphincter colli prof. and zygomatic mm.
e. frontalis m.
f. orbicularis oculi m.
g. zygomatic bone

Fig. 5: The incision to develop a combined frontalis (a) and interscutularis (b) muscle flap is outlined by broken line (c). The incision for the auricular and other superficial muscles is indicated by broken line (d).

Fig. 6: The combined frontalis and interscutularis muscle flap (a) and the rest of the superficial muscles (b), with the closely associated rostral auricular nerve plexus (c), are reflected exposing the roof of the frontal sinus (d), temporalis muscle (e), superficial temporal artery (f) (associated vein not shown), and the zygomatic process of the temporal bone. The latter is only visible from a lateral view. (See Fig. 6a).

Note also the outline of the triangular bone flap to be developed in the roof of the sinus as indicated by a broken line and also the placement sites of the three burr holes.

Fig. 6(a): Gigli saw wire in position for osteotomy of the zygomatic process of the temporal bone just rostral to its tubercle.

a. temporalis muscle
b. zygomatic process of temporal bone
c. frontal process of zygomatic bone
d. tubercle of zygomatic process of temporal bone

Inset: the zygomatic arch severed at 45° outward in the shape of a wedge.

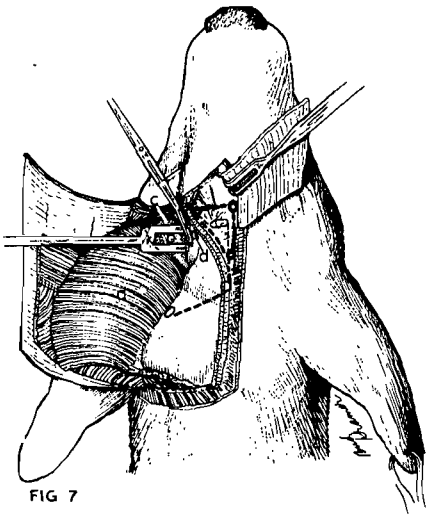


FIG 7

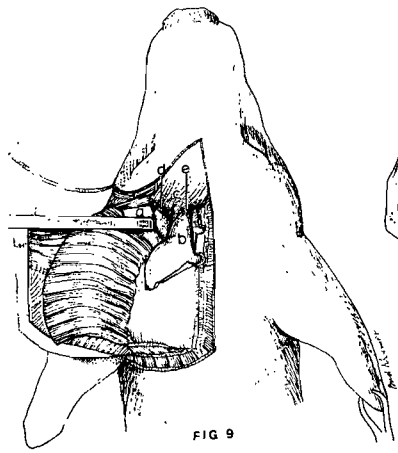


FIG 9

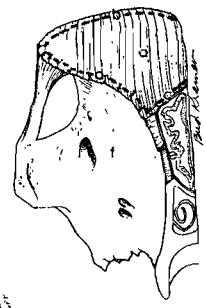


FIG 9a

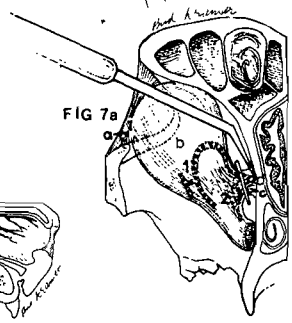


FIG 7a

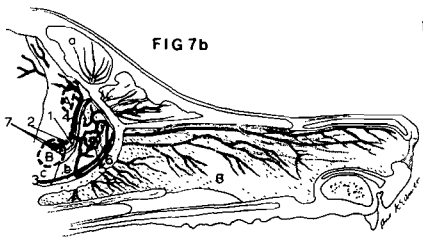


FIG 7b

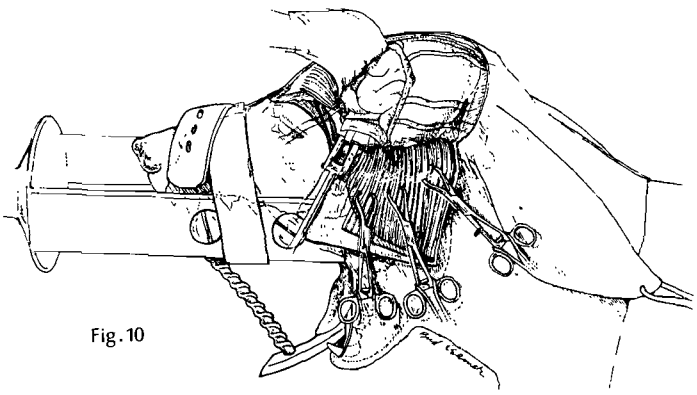


Fig. 10

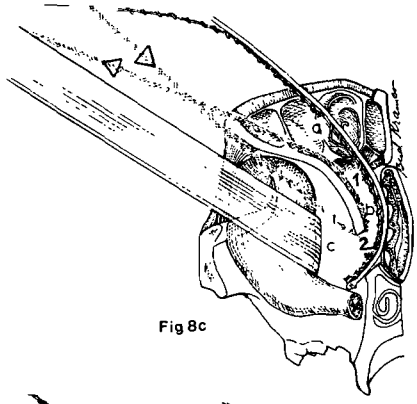


Fig 8c

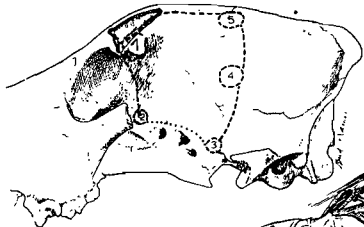


FIG 8a

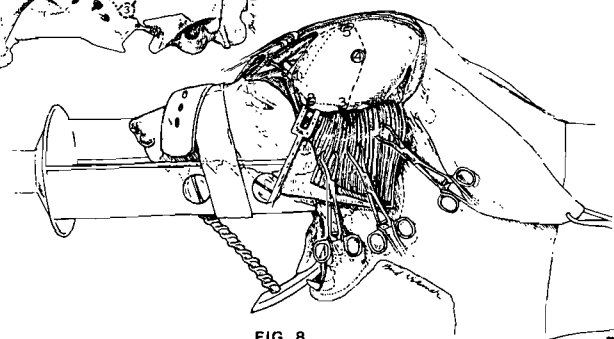


FIG 8

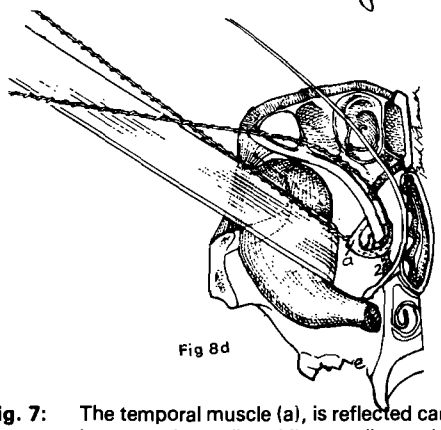


Fig 8d

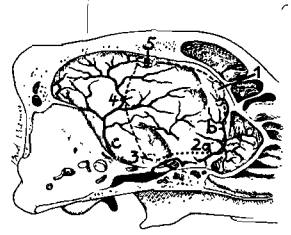


FIG 8b

Fig. 7: The temporal muscle (a), is reflected caudo-ventrally from the orbital cone (periorbita) (b) and the orbital ligament (c). The exposed cone is retracted laterally, while a small vascular clip is applied with a Scoville-Lewis aneurysm forceps to the external ethmoidal artery, vein and ethmoidal nerve, which are all ensheathed by the periorbita (d), adjacent to the orbital cone.

Note also the outlines of the pentagonal and triangular bone flaps, as indicated by broken lines, and placement sites of burr holes, only visible from a dorsal view.

Fig. 7(a): Transverse section of the cranium (half) through the rostral cranial and orbital fossae, frontal sinus at the level of the ethmoidal foramina, indicating the positions of the clip and the cauteriser tip:

a. orbital ligament
b. orbital cone, ensheathed by the periorbita
c. periosteo-periorbital ensheathment of external ethmoidal a., v. and ethmoidal n.
d. Ethmoidal foramina
   1. external ethmoidal a. (external ethmoidal vein not shown)
   2. ethmoidal nerve

Fig. 7(b): Parasagittal section of the nasal cavity, frontal sinus (a), ethmoid (b) and rostral cranial (c) fossae, to show the distribution of the ethmoidal nerve (1), external (2) and internal (3) ethmoidal arteries. Also shown is the rostral meningeal artery (4), ethmoidal rete of the cribriform plate (5) (number "5" is located just below and to the left of "6"), anastomosis of the internal and external ethmoidal arteries (6), ethmoidal foramina (7) and nasal septum mucosa (8).

The outlines of the intrasinal (A) and subfrontal (B) burr holes are superimposed upon the dural lining of the rostral cranial fossa, as indicated by broken lines.

Fig. 8: The triangular bone flap of the roof of the frontal sinus removed, and five burr holes of the pentagonal bone flap developed, craniotomy (pentagonal bone flap) with Gigli saw is illustrated by the broken line. Manual fracture of the base of the pentagonal bone flap is illustrated by the ventrally located dotted line.

Fig. 8(a), (b): The placement sites of the five burr holes, and the outline of the pentagonal bone flap are indicated by broken and dotted lines, superimposed upon the external and internal surfaces of the neurocranium (lined by dura mater).

a. external ethmoidal a.
b. rostral meningeal a.
c. middle meningeal a.

Fig. 8(c): Transverse section of the cranium (half) through the frontal sinus (a), rostral cranial (b) and orbital (c) fossae at the level of the ethmoidal foramina, indicating the position of the Gigli saw guide with the Gigli saw wire tied to its bulbous end, inserted through the exposed frontal sinus cavity, intrasinal burr hole (1), rostral cranial fossa, subfrontal burr hole (2), into the orbital fossa. Note also the proposed route and direction of to and fro traction of the wire, indicated in broken lines and by △△.

Fig. 8(d): Showing the position of the mouth piece of the Gigli saw sliding guide (a) firmly hooked onto the dorsal rim of the subfrontal burr hole (No. 2), with the Gigli saw wire sliding through it, while cutting the lateral walls of the frontal sinus and rostral cranial fossa from a dorsal to a ventral direction.

Fig. 9: Dorsal view of the calvarium with bone flaps removed, and the orbital cone (a) retracted laterally, to show the exposed dural covered frontal lobe (b). The combined frontalis and interscutularis muscle flap (c) is folded ventrally and sutured to the cut edges of the floor (d) and lateral wall (e) of the intact rostral part of the frontal sinus.

Fig. 9(a): Transverse section of the cranium (half) through the frontal sinus, orbital and rostral cranial fossae at the level of the ethmoidal foramina, illustrating the combined frontalis and interscutularis muscle flap (a) tightly folded ventrally over the cut edge of the outer table (b) and sutured to the cut edges of the inner table (c) and the lateral wall (d) of the intact rostral part of the frontal bone, thus effectively sealing off the contaminated intact rostral part of the frontal sinus and naso-frontal aperture from the cranial cavity (e) and orbital fossa (f).

Fig. 10: Surfaces of the frontal, rostral parts of temporal and parietal lobes of the cerebral hemisphere exposed allowing adequate access to rostral cranial fossa structures.

of the orbital fossa. The artery, vein and nerve adjacent to the cone (Fig. 7) were clipped[1] (vascular clip[m]) or ligated, as well as cauterised and severed in the foramina (Fig. 7a). The latter procedure prevented reflux bleeding into the orbital fossa.

These foramina served also as an external cranial landmark for the internal cranial location of the lateral border of the cribriform plate, which is located 3-5 mm rostral to these foramina. It was necessary to sacrifice the above-mentioned vessels and nerve because the subfrontal burrhole (Fig. 7b; B. Fig 8; No. 2) was to be developed over these foramina. In addition, the rostral meningeal artery, which supplies the floor (inner table) of the frontal sinus and the underlying dura, is a branch of the external ethmoidal artery; thus, preocclusion of the external ethmoidal artery resulted in a nearly bloodless surgical field with the development of the intrasinal burrhole (Fig. 7b; A. Fig. 8; No. 1).

The extent of the frontal sinus cavity was outlined with the aid of pre-surgical radiographs, (Figs. 7, 8) and a triangular bone flap developed in the roof of the frontal sinus by drilling three small burrholes in the apices of the roof, and completing the cut with a Gigli saw[n], bevelling the cut edges outwards (Fig. 7). The mucosa was stripped from the flap and stored in warm physiological saline solution with an equal part of providone iodine complex[o]. The exposed frontal sinus cavity was then flushed with the same solution, and the mucosal lining removed by curettage. Any bony trabeculae and the caudal protrusion of the ethmoid bone in the cavity were removed with a duckbill rongeurs and the remaining mucosa pushed into the nasofrontal aperture and sealed off with bone wax[p]. The sinus cavity was again flushed with the above-mentioned solution. A small (5 mm diameter) burrhole (Fig. 8, 8a, b; No. 1) was drilled far rostrally in the floor (inner table) of the sinus, medial to the supraorbital process, in the ventrolateral extension of the sinus floor. A small spherical burr[d] and burr adaptor[c] were used at a slow speed, the burr being slanted at a 30° angle towards the bone surface. Special care had to be taken in developing this hole owing to the delicate nature of the bone of the inner table and the underlying dura.

A subfrontal sinus dural elevator was introduced through the hole to separate the dura surrounding the hole, before enlarging the hole with a Kerrison laminectomy rongeurs[q] in a medial direction as far as the septum of the frontal sinus. The transverse elongation of this burrhole facilitated the separation of the dorsal sagittal venous sinus from the midline groove in the inner table underlying the frontal sinuses and the introduction of the Bailey Gigli saw guide[r] from the burrhole (fig. 8, 8a, b; No. 5) caudal to the sinus to the intrasinal burrhole (no. 1). The guide was kept as close to the midline as possible to avoid the paramedian cerebral juga[5] (transverse barrier) of the inner table under the floor of the frontal sinus.

The sinus cavity was plugged with a surgical sponge soaked in equal parts of providone iodine complex and physiological saline solution. All the instruments and gloves which had been used during the invasion of the potentially contaminated frontal sinus were replaced, the surgical site redraped, and the surgical sponges covering the temporalis muscle and orbital cone substituted[18].

The other 4 burrholes were then developed. A small (5 mm) burrhole (Fig. 8, 8a, 8b; No. 2) was placed subfrontally over the ethmoidal foramina. A similar burrhole (Fig. 8, 8a, 8b; No. 3) was positioned ventrorostrally to the border of the zygomatic process of the temporal bone, and 2 large (10 mm diameter) burrholes were developed by placing one (No. 4) 10 mm rostral to

the parietal prominence, and the other one (No. 5) 15 mm caudal to the caudal limit of the frontal sinus cavity and 7 mm paramedian to the external sagittal crest.

In all instances the Codman & Shurtleff variable speed motor drive was used. The 2 larger burrholes (No. 4 and No. 5) were drilled with a Smith automatic perforating drill[b], while the smaller burrholes (Nos. 1, 2, and 3) were made with a small spherical burr[d].

Owing to restricted surgical mobility in the subfrontotemporal region, the spherical burr had to be applied at a 30° angle to the surface of the bone at a very slow speed, when developing burrholes Nos. 2 and 3. These burrholes were slightly enlarged with a Kerrison laminectomy rongeurs to permit free passage for the Bailey Gigli saw guide. It was found best to separate the dura in the vicinity of these holes with a specially designed de Wet subtemporal[25] and subfrontal dural elevators prior to the use of the rongeurs, to prevent cutting the dura and damaging the brain, and to facilitate the introduction of the Bailey Gigli saw guide.

Particularly in older dogs with thick diploic bone in this region, drilling of burrhole No. 5 resulted in a deep pitlike burrhole. It was then necessary to groove the caudal border of this burrhole with a duckbill rongeurs. This lessened the angle with which the dural elevator and Bailey Gigli saw guide entered this burrhole. Care had to be taken in separating the dura between burrholes No. 5 and No. 1, owing to the forward slanting and prominent transverse cerebral juga[11] of the inner table of the frontal bone in this region underneath the sinus. It was found, furthermore, that the close adherence of the dura in older dogs, and the well-developed frontal diploic veins[11] entering the dorsal sagittal venous sinus in this region in younger dogs, respectively caused tearing of the dura and/or unavoidable rupture of these veins of the region. (In the later case they were sealed off by packing them with oxidized cellulose pledgets[s] after the craniotomy was completed.) It was impossible to pass a Bailey Gigli saw guide completely through from burrhole Nos. 1 to 2 or Nos. 4 to 3 owing to restricted surgical mobility in the region of burrholes Nos. 2 and 3. This surgical obstacle was overcome by tying the end loop of the Gigli saw wire to the dorsal surface of a bulbous (holed) end of the guide and passing them together through hole No. 1 to 2 and No. 4 to 3, stabilising the guide with its bulbous end protruding 13 mm through the ventral holes (Nos. 2 and 3) and pulling the wire through with a curved Kelly haemostat (Fig. 8c).

Another obstacle was the acute angle with which the wire leaves the ventral holes, which may cause locking and breaking of the wire in these holes during to and fro traction. This problem was resolved by developing a Gigli saw sliding guide, which was hooked onto the dorsal rim of the ventral holes through which the wire could now slide freely while cutting the bone from the dorsal holes (Fig. 8d).

In this way a large pentagonal transfontal lateral rostrotentorial bone flap was made by connecting (sawing) these burrholes dorsally, rostrally and caudally with a Gigli saw wire introduced by a Bailey Gigli saw guide. The cut edges were bevelled outwards. The ventral break was completed by grooving the outer table of the bone with a small burr[d] between burrholes Nos. 2 and 3 along the proposed facture line. The bone flap was removed by applying pressure over the fracture line and prying it open dorsally with a bone elevator or chisel[7]. The bone flap was kept moist in warm physiological saline.

The combined frontalis and interscutularis muscle flap was replaced, pulled tightly in a ventral direction over the cut edges of the outer and inner tables and lateral wall (frontal bone) of the exposed rostral part of the sinus cavity, cut to size to close the exposed frontal sinus cavity, so, in effect, sealing off the potentially contaminated frontal sinus from the sterile cranial cavity. The flap was sutured to the dura or to the cut edge of the lateral wall of the sinus, or preferably to the floor (inner table) and lateral wall of the sinus, with 6-0 chronic catgut (Figs. 9, 9a). The latter procedure was accomplished by drilling small holes 3 mm from the cut edge of the bone and 4 mm apart. A modification of the human Adson drill guide and dural protector was utilised for precise drilling of these holes.

A dural flap was developed in a similar manner as has been previously described by us[25].

At this point the whole frontal lobe, as far rostrally as the olfactory bulb, and a small part of the temporal and parietal lobes of the cerebral hemisphere were exposed in the craniotomy site for further exploration and surgical intervention (Fig. 10).

With the dog's head still rotated 45° away from the surgeon, the floor of the rostral cranial fossa could now be explored from a lateral approach. One end of a brain retractor[k] was inserted loosely in the gate of the de Wet selfstabilising retractor[25] and the other curved end, covered by a strip of cottonoid[t] soaked in warm physiological saline, was inserted gently underneath the frontal lobe, just rostral to the presylvian sulcus, as far medially as the curved rostroventral part of the falx cerebri on the midline of the floor of the rostral cranial fossa. The flexible arm of the de Wet self-stabilising retractor was placed in position gently to elevate the frontal lobe and then fix it in the desired position to allow access to the previously hidden structures on the left side of the floor of the rostral cranial fossa. The surgical microscope (draped[u]), was placed in position. If necessary, the gate clamp of the de Wet stabilising retractor was loosened to adjust the brain retractor while viewing the field through the microscope (Fig. 11).

The microsurgical anatomy of this region was then well visualised (Fig. 11) and ready for microsurgical intervention, such as blunt microdissection of arachnoid bands, trabeculae, arachnoid ensheathment of the internal carotid, middle cerebral, rostral cerebral and rostral communicating arterial complex, which to some extent mask the vessels and external features of the brain in this region. Microsurgical manipulation is surprisingly well tolerated by these vessels. On completion of microvisualisation (and thus of any micro-surgical task that might be required) the gate clamp was gently loosened and slowly lowered; the brain retractor was then withdrawn.

The dog's head was then either rotated 45° towards the surgeon or returned to a normal position. In the latter position the surgical microscope was placed in front of the patient and directed at a 45° angle towards the horizontal plane of the head, to continue bilateral exploration of the median and paramedian region of the floor of the rostral cranial fossa through a midline approach between the frontal lobes. The left and right frontal lobes were gently separated from the falx cerebri, just rostral to the cruciate sulci, with a finger, and the curved end of a brain retractor, covered by a

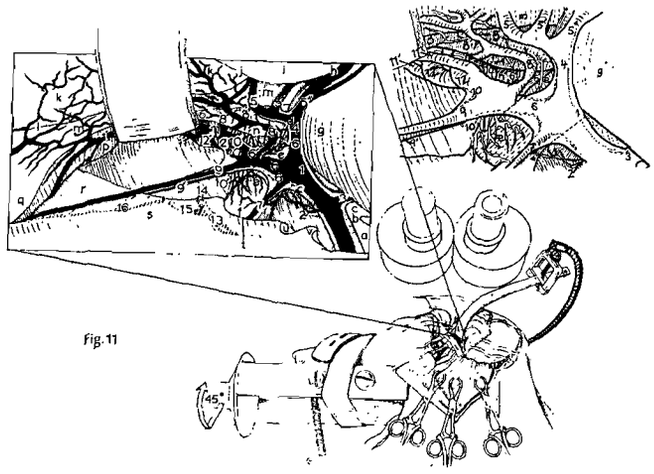


Fig. 11

Fig. 11: A lateral approach, the head is rotated 45° away from the surgeon. A brain retractor is gently placed under the frontal lobe which is slowly elevated and stabilised in the desired position with the aid of the de Wet self-stabilising retractor. The surgical microscope is moved into position and used for further adjustments of the retractor, and for viewing the microsurgical field.

Inset: Microscopic view of the microsurgical anatomy on the left side in the caudal region of the rostral cranial fossa.

a. hypophysis
b. infundibulum
c. tuber cinereum
d. lt. optic tract
e. optic chiasm
f. lt. optic n.
g. piriform lobe
h. pseudosylvian fissure
i. ventral banks of rostral parts of sylvian, ectosylvian and suprasylvian gyri
j. presylvian sulcus
k. ventral bank of prorear gyrus
l. rostral part of lateral rhinal sulcus
m. lateral olfactory gyrus
n. rostral perforated substance
o. lateral olfactory tract
p. olfactory tubercle
q. olfactory bulb
r. falx cerebri
s. floor of rostral cranial fossa (left side)
t. lt. optic canal
u. tuber sella
v. lt. rostral clinoid process

1. lt. internal carotid a.
2. basal perforating branches of rostral intercarotid a.[5]
3. lt. caudal communicating a.
4. lt. middle cerebral a.
5, 5', 5". rostral, middle and caudal branches of lt. middle cerebral a.
6. lt. rostral cerebral a. (lt. A-1 segment[19], pars precommunicalis sinistra[13])
6'. basal perforating branches of lt. rostral cerebral a. (lt. A-1 segment[19])
7. lt. internal opthalmic a.
8. lt. recurrent a. of Heubner[19] (a. centralis longa sinistra [10])
8'. basal perforating branches of lt. recurrent a. of Heubner[19] (a. centralis longa sinistra[10])
9, 9'. lt. and rt. internal ethmoidal aa.
10. rostral communicating a.
10'. basal perforating branches of rostral communicating a.[19]
11, 11'. lt. and rt. frontobasal cortical aa.[14] (aa. frontobasalis medialis sinistra et dextra[10])
12, 12'. lt. and rt. rostral cerebral aa. (lt. and rit. A-2 segments[19]; pars postcommunicalis sinistra et dextra[10])
13. left dorsal petrosal venous sinus
14. (13) anastomosis with rt. dorsal petrosal venous sinus
15. (13) anastomosis with dorsal sagittal venous sinus
16. dorsal sagittal venous sinus

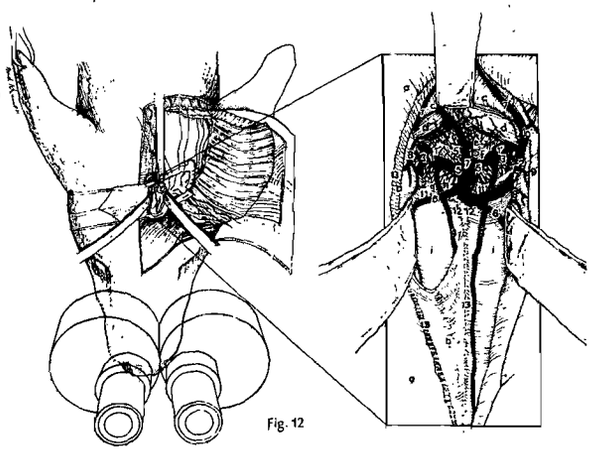


Fig. 12

Fig. 12: A midline approach, the head is returned to the normal position. Brain retractors are gently inserted between the frontal lobes, which are then gently retracted laterally. The curved bulbous end of a Gigli saw guide is hooked around the exposed genu of the corpus callosum, which is slightly retracted caudally. All of them are stabilised in desired positions with de Wet self-stabilising retractors. The surgical microscope is positioned in front of the head, and used for further adjustments of the retractors and guide, and for viewing the microsurgical field.

Inset: Microscopic view of the median and paramedian microsurgical anatomy in the caudal region of the rostral cranial fossa.

a. dural flap (reflected to right side)
b. falx cerebri
c. genu of corpus callosum
d. paraterminal gyri
e. optic tracts
f. optic chiasm
g. optic nn.
h. optic canals
i. floor of rostral cranial fossa

1, 1'. rt. and lt. rostral cerebral aa.
1". basal perforating branches of rt. and lt. rostral cerebral aa. (rt. and lt. A-1 segments[19]; pars pre-communicalis dextra et sinistra[10])
2, 2'. r.t and lt. internal opthalmic aa.
3, 3'. rt. and lt. recurrent aa of Heubner[10] (aa. centralis longa dextra et sinistra[10])
4, 4'. rt. and lt. internal ethmoidal aa.
5. rostral communicating a.
5'. basal perforating branches of rostral communicating a.[10]
6, 6'. rt. and lt. frontobasal cortical aa.[14] (aa. frontobasalis medialis dextra et sinistra[10])
7, 7'. rt. and lt. rostral cerebral aa. (lt. and rt. A-2 segments[10]; pars post communicalis dextra et sinistra[10];
7". basal perforating branches of rt. rostral cerebral a. (rt. A-2 segment[19]; pars postcommunicalis dextra[10])
8, 8'. rt. and lt. ventromedial frontocortical br.[14] (aa. frontopolaris dextra et sinistra[3])
9'. lt. intermedio-medial frontocortical br.[14] (a. frontopolaris sinistra[3])
10'. lt. dorsomedial frontocortical br.[14] (a. callosomarginalis sinistra[3])
11, 11'. rt. and lt. dorsal petrosal venous sinuses
12, 12'. (11, 11') anastomosis with dorsal sagittal venous sinus
13. dorsal sagittal venous sinus

cottonoid strip, was slowly inserted, sliding it down along the surface of the falx cerebri until it reached the floor of the rostral cranial fossa. It was then pressed against the falx to the right, the other end of the retractor inserted into the gate of the de Wet self-stabilising retractor[25], and the retractor slightly retracted to the right before closing the gate clamp and fixing the stabiliser. The left frontal lobe was then gently retracted with the finger and another curved brain retractor gently inserted in a similar manner down to the floor of the fossa, snugly against the first one. It was then stabilised in the same manner as the previous one, after slight left lateral retraction. These manoeuvres exposed the genu of the corpus callosum. The genu was then covered with a cottonoid surgical patty before it was gently and slightly retracted caudally with the curved bulbous end of a Bailey Gigli saw guide, which was then also stabilised.

With the aid of the surgical miocroscope correctly positioned, the retractors and guide were adjusted in such a manner as to allow bilateral access to the previously hidden structures on the midline of the floor of the fosa (Fig. 12).

In this approach care had to be exercised to prevent excessive lateral traction of the lobes to prevent dislodging the olfactory bulbs from the ethmoid fossae, so causing profuse haemorrhage from the ethmoidal rete of the cribri-form plate and/or tearing of the genu of the corpus callosum.

The microsurgical anatomy of this region, from the optic chiasm to the crista galli, was clearly visible and there was sufficient microsurgical mobility between the frontal lobes, which might have been impossible from the lateral appoach. The 2 rostral cerebral arteries (A-2 segments[19]) were separated by blunt microdissection of the arachnoid sheath surrounding them, thus providing a better view of the suprachiasmatic area, optic chiasm, optic nerves, and rostral cerebral-rostral communicating artery complex (Fig. 12).

Accidental haemorrhage in these areas could be controlled by suction, low frequency bipolar electrocautery, small vascular clips[v] or by packing with oxidised cellulose pledgets, gel foam sponges[w] or crushed muscle, depending upon the functional importance of the vessel.

After completion of the inspection (and thus of any desired microsurgical task) the area was flushed with warm physiological saline. Care was taken to see that all haemorrhage in this area was brought under control before the guide and retractors were withdrawn in the same manner as previously mentioned.

The dual flap was closed in a watertight fashion with 6-0 catgut, especially where it underlies the craniotomy defect in the floor of the frontal sinus, to prevent possible meningitis[18]. Closure of the dural defect was impossible in one of our dogs owing to some brain swelling which might have occurred as a result of excessive manipulation of the frontal lobes and/or owing to the fact that the left jugular vein and left carotid artery had been occluded experimentally prior to surgery. In this case the defect in the dura was repaired by an autograft of temporalis muscle fascia[18]. Prior to replacing the large pentagonal bone flap, the small triangular bone flap was secured to the large pentagonal bone flap by fine orthopaedic wire, of which the twisted ends were turned face downward into the cavity of the sinus to minimise foreign body irritation and penetration of the overlying skin.

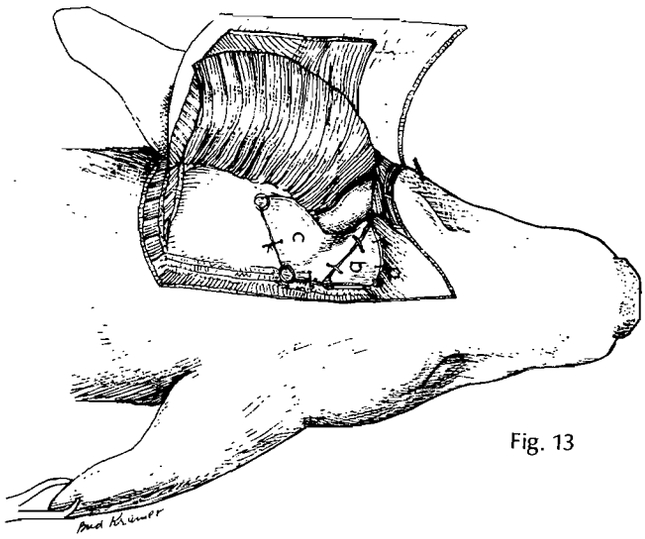


Fig. 13

Fig. 13: Dorsal view of the calvarium showing the combined frontalis and interscutularis muscle flap (a) folded ventrally and the triangular (b) and pentagonal bone (c) flaps wired in place.

The pentagonal bone flap was wired to the dorsal edge of the craniotomy defect between burrholes No. 4 and No. 5, and between No. 5 and No. 1. In this manner the bone flaps were secured to the skull in a hinge-like fashion, resting rostrally on the ventrally folded combined frontalis and interscutularis muscle flap with perfect alignment along the rest of the craniotomy defect, but with still some room for outward movement ventrally, cushioned by the replaced temporalis muscle mass (Fig. 13). This condition was found to be exceptionally favourable for the release of immediate post-surgical intracranial pressure and to avoid post-surgical compression of the cerebral cortex by any dural replacement graft underneath the bone flap[18]. The bone flaps stabilised themselves with good appositional alignment, as was borne out by successive radiographs, normal symmetry of the head, and good cosmetic appearance.

A penrose or preferably a silastic drain[x] was placed beneath the temporal muscle sutured in place. It was found advisable to secure the zygomatic arch tightly with fine orthopaedic wire to prevent misalignment of the jaws and malocclusion of the teeth. The ear muscles and other superficial muscles of the head and the superficial fascia then were successively sutured into place. Finally another penrose, or, preferebly, a silastic drain, was placed in a subcutaneous position before suturing the skin into place. The latter drain was very effective in preventing the formation of a post-surgical subcutaneous seroma[25]. These drains were removed 48-72 h later.

Posturgical Treatment

The patient was transferred to a padded foam rubber cage in the intensive care unit. A slow intravenous drip of lactated Ringer's solution was continued and the bladder periodically expressed through the catheter. Maintenance of a patent airway was ensured and oxygen administered via a face mask on a continued basis for the duration of the recovery period, to prevent cerebral anoxia and brain swelling.

Antibiotic therapy was continued for one week post-operatively using chloramphenicol intravenously and procaine penicillin intramuscularly. The Azium dosage

was gradually reduced over the first 48-72 h unless there were signs of cerebral oedema.

It was found advisable to sedate the patient immediately post-operatively in the presence of strong vital signs, to maintain comfort. Our dogs were sedated with 5 mg of Valium[y] every 2 h and 30 mg of Talwin-V[z] every 4 h. Strong narcotic analgesics were avoided to prevent further depression in a patient that is already depressed.

Vital and other physical signs possibly indicative of post-surgical brain swelling, oedema and/or intracranial haemorrhage were monitored continuously. Suggestive signs included dilation of the pupils, bradycardia, depressed respiration and decreased response to stimuli. Cerebral oedema was effectively controlled by hyperventilation, mannitol intravenously or continued Azium therapy.

The patient could be removed from the intensive care unit from 24 to 48 h post-operatively, depending on the state of progress.

Cerebrospinal fluid culture and analyses were done and radiographs of the head taken one week post-operatively. The dogs were fed soft food and observed 3 times daily for neurological and behavioural status.

Histopathology

After 1 to 2 months post-operatively the dogs were perfused with 10 % buffered formalin by intracardiac cathertisation under deep anaesthesia. Their brains were removed and tissue blocks of dura, temporalis muscle fascia autograft and floor and roof of the frontal sinus were collected. For light microscopy, fixed tissues were dehydrated, embedded in paraffin, and stained with haematoxylin and phloxine-safranin. The frontal lobes of the brains were coronally sectioned at 6 μm and stained with haematoxylin and eosin, and with luxol fast blue/cresylecht violet.

RESULTS

Physical and Neurological Examination

All dogs recovered from anaesthesia with no neurological and/or behavioural disfunction, except for immediate post-operative depression and discomfort. Within a week the younger and middle-aged dogs returned to their normal attentive and responsive behaviour, with the older dog slightly lagging behind in its progress to normality. No signs of cerebrospinal rhinorrhoea, sinusitis, meningitis and encephalitis were observed in these cases. This was confirmed later by autopsy and histopathological findings. Nevertheless, one dog developed a superficial infection of a purulent nature along the suture line of the skin between the eyes, which was probably due to an infected cage. The infection was brought under control within a week with 50 mg Gentocin[zz] therapy.

At 2½ days and 3 weeks after surgery the symmetry and cosmetic effect of the surgery on the heads was compared with the normal preoperative ones, with remarkably little difference. This was also borne out by radiographs of the excellent alignment of the bone flaps and zygomatic arch of the cranium. Restoration of normal ear, eye, eyelid, jaw and facial expression movements had been accomplished. No difference in olfaction and vision were detected.

Clinical Pathology

Cerebrospinal fluid was slightly xanthochromic in one case; the remaining CSF analyses were normal. No evidence of bacterial contamination was found on the cultures. A swab culture of the skin infection yielded a growth of *Enterococcus* as well as *Escherichia coli*, which were both susceptible to antimicrobial agents, such as Gentamicin[zz] and Tribrissen.

Pathology

Healing of all incisions proceeded by first intention without complications, except in the one case with the skin infection, where eventual healing was accompanied by slight granulation and some scarring. The osseous union of the wired bone flaps, and in 2 cases where the severed zygomatic arch were secured by wire, progressed in a normal manner, except in the area where the bone flaps were separated by the infolded muscle flap from the intact rostral part of the frontal bone. In one case, where the arch was not fixed, permanent misalignment of the arch, and some exostosis and delayed ossification of the cutting defects were radiographically observed and later confirmed at autopsy. After 1 to 2 months no osseous union was grossly observed, radiographically or histologically, between the cut edges of the bone flaps and opposing cut edge of the intact rostral part of the frontal bone (separated by the muscle flap). Some reaction of new bone formation was histologically evident along the opposing cut edges (inner and outer tables of the frontal bone, and lateral wall of the sinus). Grossly and histologically a fibrotic reaction from both sides of the infolded muscle flap and the periosteal lining of the sinus cavity was responsible for a collagenous invasion of the sinus cavity, which effectively and completely sealed off the potentially contaminated rostral part of the sinus from the neurocranial cavity (Fig. 14A, B).

The left internal ethmoidal artery was slightly hypertrophied. The dural incision healed by first intention and the fibrous union between the temporal muscle fascia autograft and adjacent dura was complete (Fig. 15A, B).

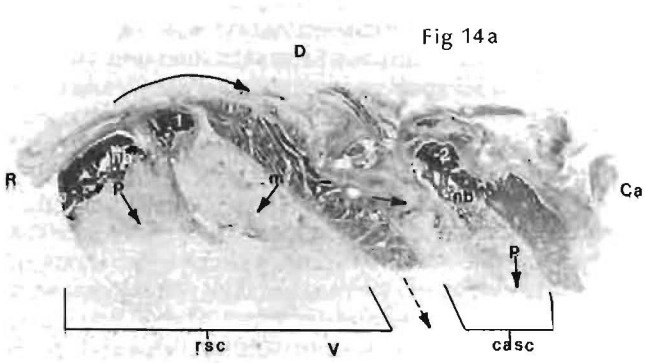
No lesions were detectable by gross examination of the brains, and histopathological changes were absent in the frontal lobes.

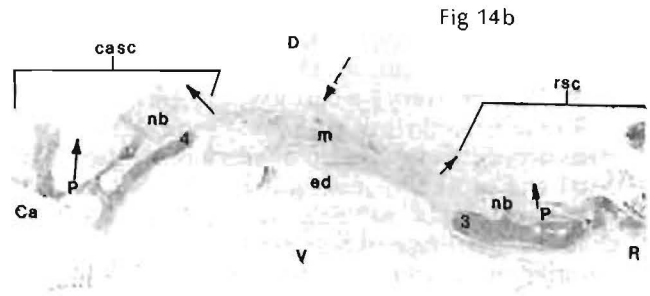
DISCUSSION AND CONCLUSION

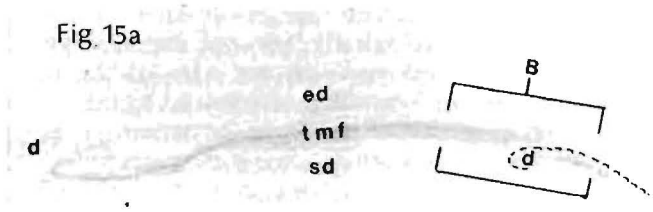
The surgical approach to the rostral cranial fossa through extensive transfrontal craniotomy and an effective closure of the potentially contaminated frontal sinus with a muscle flap prior to durotomy, combined the advantages of an aseptic field, minimal haemorrhage, better control of haemorrhages, with much wider exposure and detailed visualisation of anatomical structures in the rostral cranial fossa. Such an approach also lends itself to a greater degree of surgical and microsurgical mobility with no post-operative behavioural, personality or intellectual disturbances or neurological deficits.

The sacrificed external ethmoidal artery, vein and ethmoidal nerve had no detrimental effects upon blood supply and sensation in the nasal cavity, owing to post-surgical development of collateral circulation and regeneration respectively.

In spite of the tight closure of the frontal sinus by the muscle flap, the cerebral cortex needs further separation from the sinus cavity by an intact water-tight dura to prevent possible meningitis[22]. Where this is not possible,







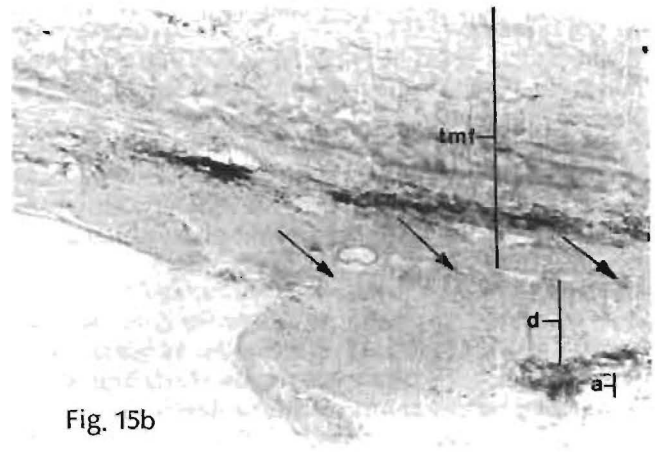


Fig. 14A, B: 1. Cut edge of outer table of intact rostral part of frontal bone, 2. opposing cut edge of triangular bone flap (roof of frontal sinus). m. Combined frontalis and interscutularis muscle flap (ventrally folded) separating rostral (rsc) and caudal (casc) parts of the frontal sinus, p. periosteum. Fibrotic reactions along rostral and caudal surfaces of the muscle flap (m→), and along the periosteal lining (p→) of the sinus, which resulted in a collagenous invasion of the frontal sinus (fibrous granulation). nb. Reaction sites of new bone formation. D = dorsal; V = ventral; R = rostral; Ca = caudal. Haematoxylin Phloxine Safranin stain; X 5.6 (30 days post-op).

Fig. 14B: 3. Cut edge of inner table of intact rostral part of frontal bone, 4. opposing cut edge of pentagonal bone flap (floor of frontal sinus). m. Combined frontalis and interscutularis muscle flap, sealing off the frontal sinus from the cranial cavity. Haematoxylin Phloxine Safranin stain; X 5.3 (30 days post-op).

Fig. 15A: The dural defect filled by a temporalis muscle fascial autograft. Note complete fibrous fusion between the dura mater and the fascial autograft at their overlapping junctions. ed. Epidural region, sd. subdural region, d. dura mater, tmf. temporalis muscle fascia autograft. Haematoxylin Phloxine Safranin stain; X 5.3 (30 days post-op).

Fig. 15B: Enlargement of a overlapping dural-fascial junction. tmf. Temporalis muscle fascia autograft, d. dura mater, a. arachnoid membrane. Note the complete fibrous union (arrows) between the outer surface of the dura mater and inner surface of the fascial autograft. Haematoxylin Phloxine Safranin stain; X 50 (30 days post-op).

other tissues can be used to fill the defect in a watertight fashion; e.g., in man a free periosteal flap[22], free fascial or periostreal grafts[12], lyophilised dural grafts[12], or a stamp of muscle have been used[20]. We are in agreement with others[18] that temporalis muscle fascia is a good source for such a graft in the dog.

The bone flap should be stabilised with special care over any graft, in such a way as to avoid compression of the cerebral cortex[18]. In our patients, with or without a graft, these bone flaps were wired in place in a hinge-like fashion, resting rostrally upon the ventrally folded muscle flap. This "floating" arrangement prevented compression of the brain, and allowed some release from possible post-surgical oedema and swelling of the brain without later sacrificing the normal symmetry and cosmetic appearance of the head.

The technique is designed for application by veterinary surgeons in a well-equipped surgical practice, or by investigators, with minimal investment in special neurosurgical instruments, except for the possible addition of a surgical microscope or dissecting microscope (table model) with a shadowless surgical lamp. Ophthalmic surgical instruments may be used instead of microsurgical instruments.

This technique should first be attempted on a cadaver so that the surgeon may familiarise himself with the use of a surgical microscope and the *in situ* macro- and microsurgical anatomy. In afflicted animals abnormal anatomical states are to be expected, such as distortion or masking of vessels in the rostral cranial fossa by frontal lobe, rostrocellar, other rostral cranial fossa tumours (e.g., meningiomas) and haematomas. Debulking of the tumours before their removal may be advisable to prevent unnecessary surgical invasion of normal tissue, and to appreciate better the normal variation of arrangement of vessels in this region in the dog. Such variations have been reported in man[19].

With special attention to pre-, intra-, and post-surgical treatment, and by abiding by a strict surgical protocol, this technique can be a feasible surgical alter-

native for management of lesions of the frontal lobes and in the rostral cranial fossa of the dog.

ACKNOWLEDGMENTS

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FOOTNOTES

a. Model NO1023
b. Model N-61
c. Model N-1023 j
d. Models 0-6539b; 0-6539e,f; 0-6531a,c,d,e; 0-6534d Codman & Shurtleff, Inc., Paceller Park, Randolph, Mass. 02368
e. Azium-Dexamethozone. Schering Corp., Kenilworth, N.J. 07033
f. Chloramphenicol-chloromycetin. Med. Tech. P.O. Box 338 Elwood, K.S. 66024.
g. Sodium thiamylal – Surital, Parke, Davis & Co., Detroit, MI.
h. Methoxyflurane – Metofane, Pitman-Moore Division Dow Chemical Co., Indianapolis, IN.
i. 25 Mannitol Injection – Cutter Laboratories, Inc., Berkley, CA 94710.
j. Gardlok, Neurological sponge strips. Edward Weck & Company, Inc. Research Triangle Park, N.C. 27706.
k. Models J-270-271 "M.Y." Pattern Brain Retractors. Ruggles, Corp., 38 Billings Rd., Boston, MA 02171.
l. Scoville-Lewis clip applying forceps, 165 MM, J-432-04. Ruggles Corp., 38 Billings Rd., Boston, MA 02171.
m. Mackenzie 100 ea. aneurysm silver clips. V. Mueller, Chicago, IL 60648.
n. Gigli saw blades, R-126, 127. Ruggles, Corp., 38 Billings Rd., Boston, MA 02171.
o. Betadine® solution. Purdue Frederick Corp., 38 Billings Rd., Boston, MA 02171.
p. Horsley's Bone Wax. A.S.R. Medical Industries, 100 Park Ave., N.Y. 10017.
q. Codman Kerrison laminectomy rongeurs, E.A. 530-1370 (3 MM up bite, 6" shaft) Codman & Shurtleff, Inc. Paceller Park, Randolph, MA 02363.
r. Bailey Gigli saw guide, R-110, Rugles Corp., 38 Billings Rd., Boston, MA 02171.
s. Oxidized cellulose pledgets – oxycel.® Parke, Davis Co., Detroit, MI 48232.
t. Codman surgical patty. Codman & Shurtleff, Inc., Randolph, MA 02368.
u. Xomed Sterile Disposable Operating Microscope Drape, xo-2921. Xomed, 8641 Baypine Rd., Jacksonville, FL. 32216.
v. Scoville-Lewis intracranial clip (4 mm × 0,75 mm) J-432-10, Ruggles Corp., 38 Billings Rd., Boston, MA 02171.
w. Gel foam-absorbable gelatin sponge. The Upjohn Co., Kalamazoo, MI 49001.
x. Silastic medical grade tubing, (0,62" ID × 0,095" OD). Dow Corning Corp. Medical Products, Midland, MI 48640.
y. Valium-Diazepam. Roche Laboratories, Nutley, N.J. 7110.
z. Talwin-V-Pentazocine lactate. Winthrop Laboratories. New York, N.Y. 10016.
zz. Gentocin-Gentamycin sulphate. Schering Corp. Kenilorth, N.J. 07033.

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ABSTRACT: Huismans, H. & Erasmus, B.J., 1981. **Identification of the serotype-specific and group-specific antigens of bluetongue virus.** *Onderstepoort Journal of Veterinary Research,* **48**, 51-58 (1981).

The bluetongue virus (BTV) core particle contains 2 major polypeptides, P3 and P7, and is surrounded by an outer capsid layer that is composed of the 2 major polypeptides, P2 and P5. Analysis of the immune precipitates from soluble ^{14}C-labelled BTV polypeptides and hyper-immune rabbit and guinea-pig sera indicated that polypeptide P2 precipitates only with homologous BTV sera. This would indicate that P2 is the main determinant of serotype specificity. It was also found that in sheep infected with BTV the P2-precipitating antibodies in the serum correlate with the neutralizing antibody titres, whereas the appearance and subsequent decline of P7-precipitating antibodies correspond well with those of the complement fixing antibodies. This suggests that BTV group specificity, as measured by a complement fixation test, is determined by the core protein P7. This result was supported by the observation that mouse ascitic fluid, which contains a high titre of BTV-specific complement fixing antibodies and a very low titre of neutralizing antibodies, contains almost exclusively antibodies that precipitate P7.

CONTINUED EDUCATION VOORTGESETTE OPLEIDING

REACTION PATTERNS IN MYOCARDIUM IN RESPONSE TO INJURY

S.J. NEWSHOLME*

ABSTRACT: Newsholme S.J. **Reaction patterns in myocardium in response to injury.** *Journal of the South African Veterinary Association* (1982) **53** No. 1, 52-59 (En) Vet. Research Institute, 0110 Onderstepoort, Republic of South Africa.

Some reactions and their components at a microscopical level in myocardium are described using evidence obtained from natural and experimental cardiac conditions. Necrosis of different types, hypertrophy, mineralization, and other changes in myocytes, changes in myocarditis, and patterns involving conductive tissue are considered.

A short summary of some collected values concerning the chronology of myocardial reactions is presented.

The pathogenesis of myocardial reactions is complex and is influenced by diverse factors. Although certain patterns are recognizable, firm aetiological interpretation from the pattern alone is rarely justifiable.

INTRODUCTION

Cardiac pathology, although it has much to contribute in the assessment and understanding of cardiac disorders, is beset with special problems which probably reflect the complexity of cardiac structure and function. One problem lies in the assessment of the functional significance of lesions. In some cases, lesions which appear severe and extensive may be clinically silent, whereas in others, for example where conductive tissue is involved, relatively mild lesions have been associated with severe cardiac dysfunction, arrhythmias and death. Lesions which are very early, and may be difficult to detect at all, may also be responsible for severe cardiac dysfunction and death. This is well illustrated by cases of recent myocardial infarction.

Another problem arises from the chronic nature of many heart lesions. Often all that remains is scar tissue, representing only the monument to previous changes and providing no clue to the aetiology or pathogenesis of a previously active process[52]. This difficulty underlies study of the cardiomyopathies and has stimulated quest for animal models which might elucidate pathogenesis.

Some reaction patterns at a microscopical level, involving the various components of myocardium, are considered here and, where possible, their sequence, specificity, and relation to injury are described. Hopefully, this will lay grounds for a clearer interpretation of myocardial lesions and their practical significance in relation to aetiology and pathogenesis.

CARDIAC MYOCYTES

Injury

The cardiac myocyte is particularly susceptible to injury by numerous conditions and agents. This susceptibility has been attributed to the high degree of differentiation and complex physiological and biochemical activities of this cell[52]. Morphological features of damage, produced by various injurious influences in both experimental and natural conditions, have been the subject of many electron microscopic (EM) studies, reviewed elsewhere[20]. These include electrolyte changes, administration of drugs, toxins, hormones, infectious agents and ischaemic and hypoxic influences. It was concluded that different agents cause a variety of organellar changes so that myocyte necrosis may evolve along a number of different pathways. In general, however, even at an ultrastructural level, the range of possible reactions is non-specific and limited. Myocyte reactions may be considered under a general concept in the pathogenesis of cell necrosis, as has been illustrated with reference to hepatocytes[23] i.e. that any cell may react ultimately to injury only by assuming a new steady state, manifest as degeneration, recovery, or hypertrophy. Alternatively, the cell may fail to adapt, manifest as necrosis.

As for any tissue, the precise point at which damage ends and reaction begins cannot be defined accurately. What may be more important to determine is the stage of irreversible change after which subsequent reaction becomes inevitable. Experimental evidence suggests that, in ischaemic injury, the stage of irreversibility is determined by structural and functional changes in mitochondria. For example, the stage of irreversible injury to canine myocardium following experimental coronary arterial occlusion[22] was marked by several morphological alterations in mitochondria, associated with breakdown of Krebs cycle metabolism. Beyond this stage, necrosis ensued, whether or not circulation was restored.

Necrosis

Although, as has been outlined, necrosis of the cardiac myocyte represents a final, non-adaptative sequel, common to various types of injury, it has been proposed[4 5] that there are 3 distinct morphological types of myocyte necrosis, and that certain features of these 3 types suggest cellular death in different functional states. This is important in considering myocardial reaction patterns because each type has been claimed to reflect a distinct pathogenesis and also influences the subsequent tissue reactions.

I. *Coagulation necrosis*[4 5 32]

This type is characterized by cellular swelling, early loss of striations and hyaline, or sometimes granular, sarcoplasm. The nucleus becomes hypochromatic and disappears at an early stage. Corresponding EM changes have been followed in experimentally induced myocardial

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infarcts in rats[24], and have been summarized as early glycogen depletion, relaxation of myofibrils, swelling of mitochondria and sarcoplasmic reticulum, and early condensation and margination of nuclear chromatin followed by nuclear dissolution.

Coagulation necrosis has been recognized in human myocardial infarcts as well as in a number of experimental ischaemic and anoxic situations. The morphological features have been reported to suggest death of the myocyte in a state of relaxation, or atony.

II. *Coagulative myocytolysis* (myofibrillar degeneration with contraction bands)[4 5 43]

This type is characterized by the early loss of both transverse and longitudinal striations and the appearance of thick, irregular, transverse, eosinophilic bands, referred to as contraction bands. These bands suggest coagulation of hypercontracted sarcomeres[43]. They alternate with paler staining, granular zones. Nuclear changes occur at a later stage. By EM the bands appear as aggregations of hypercontracted sarcomeres. The granules between the bands are translocated mitochondria which sometimes become mineralized. These features are reported to suggest death in a hypercontracted, tetanic state.

Coagulative myocytolysis has been described in various circumstances including myocardial infarction, following cardiovascular surgery, in association with subarachnoid haemorrhages[17], and in cases of phaeochromocytoma[43].

Experimentally, similar lesions have been associated with hypokalaemia, temporary ischaemia, catecholamine and corticosteroid administration, haemorrhagic shock, cobalt intoxication and following stimulation of various centres of the brain[43]. Under experimental conditions this type of necrosis can be prevented or alleviated by prior administration of adrenergic blockers, such as propanalol. The above evidence has led to a unifying theory in the pathogenesis of coagulative myocytolysis i.e. that it results from stimulation of excessive release of endogenous catecholamines, either from myocardial nerve endings to explain localized lesions or into the circulation to explain more diffuse and widespread lesions[43].

The association of coagulative myocytolysis with human cases of subarachnoid haemorrhage, and also with experimental electrical stimulation of various brain areas in cats[17], led to the deduction that the cardiac lesions resulted from activation of sympathetic "centres" in the brain, with disturbance of autonomic outflow, leading to excessive catecholamine release. It has been suggested[43] that this mechanism might be important in the apparent association between stress or anxiety states in man and raised incidence of heart attacks.

III. *Colliquative myocytolysis* (myofibrillar lysis)[4 5]

This type is characterized by loss of striations and a homogenous, weakly eosinophilic cytoplasm. Nuclear changes occur at a late stage. By EM there is evidence of intracellular oedema with disintegration of fibrils and other organelles, leading to vacuolation and dissolution of the sarcoplasm[24]. The pathogenesis is not known. However, cells showing similar changes have been described in infarction[4 5], in rheumatic heart disease[51] cobalt intoxication[20], and in myopathic hamsters[37].

Other lesions, possibly related to necrosis

Zonal lesions

These lesions have been decribed in dogs following experimental haemorrhagic shock[34]. Affected myocytes were distributed in subendocardial areas of ventricles and septum, and lesions were most prominent in the right papillary muscles. Light microscopy showed very prominent intercalated disks. EM revealed hypercontraction and disorganisation of myofibrils, with collapsed Z lines, near the intercalated disks. The cytoplasm towards the cell centre was paler and contained translocated mitochondria. The sarcolemma was scalloped at each end of the cell, adjacent to the intercalated disks. These changes were interpreted to represent a local, disruptive hypercontraction involving the ends of each myocyte.

Features suggest similarity to coagulative myocytolysis and it is not certain whether the zonal lesions represent the same process in a more localized form or at an earlier, possibly reversible, stage. Evidence suggests that the cause of both may be closely related. Zonal lesions in dogs with haemorrhagic shock are related to the intensity of catecholamine response and can be augmented by norepinephrine treatment and diminished or prevented by beta blockade, cardiac denervation and adrenalectomy[18].

Shredding lesions

These lesions were described as an apparently new and distinctive type, observed in the right ventricular myocardium of rats following experimental pulmonary embolism[11]. The lesions could be visualized initially by EM only, but later, fully developed lesions could also be seen by light microscopy (LM). By EM the whole myocyte appears stretched with myofibrils distorted, having a shredded appearance with irregular gaps containing random, apparently normal mitochondria. Intercalated discs are distorted and Z lines and I and A bands are lost. In well developed lesions, the gaps appear as irregular, clear zones by LM.

The pathogenesis of the shredding lesions is not understood. As circulation appeared normal and mitochondria were not swollen, an ischaemic pathogenesis was considered unlikely. Mechanical stretching is a possibility, arising from abnormal myocardial contraction. This would be consistent with the apparent pulling apart of the contractile elements. It is unclear whether this lesion is reversible and whether or not it occurs under other conditions.

Waviness

Waviness of myocardial fibres has been described as a constant feature of human myocardial infarction and may be a useful early indicator of infarction[8]. Areas of regular waviness of single fibres ("first order" waviness), fibre bundles ("second order" waviness) and undulations of wavy fibre bundles ("third order" waviness) have been described. Bundles of wavy myocytes were thinner than normal with broader transverse striations, giving the appearance of having been previously stretched. Shortly after infarction, the ischaemic area bulges outwards. It was suggested[8] that the ischaemic area loses contractility and is irreversibly stretched by a compensatory contraction of the surrounding myocardium. Elasticity of the stromal framework then retracts

the fibres which are unable to fit the framework, and so assume waviness.

It is uncertain whether waviness progresses to necrosis although wavy fibres undergoing coagulation necrosis have been described[8]. Waviness has also been reported in congestive cardiomyopathy in cats[48], and occasionally in human congestive cardiomyopathy[49]. Its true significance is in doubt since it has been claimed to occur in normal human and rat myocardium[14].

Myocyte necrosis – discussion and conclusions

Distinction of the morphological types of necrosis and related lesions is not always easy. The stage at which the material is examined may cause difficulties. Early stages of both coagulation necrosis and coagulative myocytolysis show only acidophylic changes[4 5]. Contraction bands and perinuclear vacuolation, it has been claimed, can result from fixation and manipulation of tissue too soon after death[26].

Myocyte necrosis may also be confused with autolysis. It has been shown[21] that morphological changes of myocytes in autolysis and ischaemia have a very close resemblance.

From a practical diagnostic standpoint, the distinction of different types of necrosis is not very useful. Although clues to the pathogenesis of each type have been provided, the field is still controversial and the finding of a particular type has little aetiological significance. This is not surprising in view of the complexity of cardiac processes. Myocardial infarction again provides a good example. The myocardial infarct is fundamentally an ischaemic lesion, yet coagulation necrosis, coagulative myocytolysis and colliquative myocytolysis have all been seen associated with the same infarct[4 5].

Other lesions may be connected intimately with necrosis and develop secondarily to abnormal contractile activity associated with the primary area of necrosis. This is, possibly, the case for shredding lesions and for waviness.

The types of necrosis are important, however, because they influence the nature and sequence of subsequent rections[4 5].

Mineralization

The deposition of calcium salts within degenerating or necrotic myocytes, to which the classical term, dystrophic calcification, might be applied, has been studied by EM in rats undergoing experimental Mg^{2+} depletion[20]. Ca^{2+} salts were deposited within mitochondria, which subsequently aggregated to form large, mineralized masses.

A clear association between mineralization and a particular type of necrosis is not established. However, descriptions of mineralization following experimental catecholamine and cortisone administration, in haemorrhagic shock and various forms of cardiac ischaemia[20] suggest an association with coagulative myocytolysis.

If the proposed mechanism in the pathogenesis of coagulative myocytolysis is correct[43], i.e. that it is a form of hypercontraction resulting from failure of Ca^{2+} efflux, this could explain a higher concentration of Ca^{2+} salts in sarcoplasm and mitochondria and might serve to explain the apparent association between coagulative myocytolysis and mineralization.

However, other investigators consider that calcification only occurs following irreversible damage to the myocyte and is attributable, at least in part, to increased Ca^{2+} influx through a damaged sarcolemma which has lost its selective permeability[46]. It is also of interest that in a study of Vitamin E/ selenium deprived pigs[50], calcification was associated with necrotic cardiac myocytes having features of coagulative myocytolysis. Calcification was assumed to occur by uncontrolled Ca^{2+} influx across membranes damaged from lack of Vitamin E/selenium control of cellular peroxidation.

In experimental ischaemia it was found that mitochondrial calcification was accelerated in irreversibly damaged cells only after blood flow was re-established[7], suggesting that mitochondrial calcification in the damaged cell takes place by transfer of blood calcium.

To summarize, this type of calcification involves mitochondria and may be associated with coagulative myocytolysis. However, the mechanisms involved and the specificity of association with any particular type of necrosis are still unclear. Thus, intracellular calcification associated with necrosis cannot be regarded as a reaction of specific diagnostic value.

Hypertrophy

Cardiac hypertrophy, which is recognizable grossly as an increase in cardiac size and/or increased myocardial thickening, is a salient feature of certain myopathies in both animals and in man. These increases are accountable histologically by hypertrophy of myocytes. Myocyte hypertrophy occurs under conditions of increased load and might be considered as a basic cellular reaction in response to a type of injury, possibly ischaemic, assuming a new steady state in accordance with the principles considered above. However, problems arise in the histological interpretation of cardiac hypertrophy, associated with difficulties in differentiating normal growth, physiological and pathological hypertrophy.

Hypertrophy and cardiac growth

Myocyte mitosis decreases rapidly prior to birth, and very little mitotic activity can be measured in these cells later than a few weeks after birth. Furthermore, there is no firm evidence that cardiac myocytes are capable of division in response to any stimuli in the adult heart. If the Anitschow cell originates from the myocyte, which is a possibility still in dispute, proliferation of this cell would represent an exception. However, it is generally accepted that, from a few weeks after birth, all myocardial growth must be achieved by hypertrophy and that hyperplasia makes no contribution to the increasing myocardial size[56].

There is good evidence that both growth and hypertrophy of myocardium are determined by haemodynamic load[56], and that myocardial growth might be expressed as a form of compensatory hypertrophy in response to increased haemodynamic load. In this sense, growth and hypertrophy are one and the same. This also corresponds with the microscopical features which are apparently indistinguishable. Thus the only factor which separates growth and physiological hypertrophy of myocardium from pathologically induced hypertrophy as a tissue reaction is not the reaction itself but the nature of the stimulus.

Changes associated with myocyte hypertrophy

As might be expected, appreciable increase in myocyte size has been measured in growth and hypertrophy, as has been demonstrated in canine myocardium[25 55].

Early changes in hypertrophy have been followed, morphometrically, using rats as model in which haemodynamic load was increased by surgical aortic banding[3].

It was found that by 8 days the mean myocyte volume had increased by 86%. This increase was accountable by an increase in number, but not size, of organelles. The number of myofibrils was greatly increased, with the formation of many new sarcomeres in each myocyte.

Morphological changes in myocyte nuclei in growth and hypertrophy have also been studied in human autopsy material[1]. These changes included increased nuclear volume and angularity, and fine, netlike heterochromatin in childhood growth and hypertrophied adult hearts. "Ledge" nuclei were also described, being forms with longitudinal ridges, which probably represented nuclear indentation by thickened myofibrils. Changes in growth and hypertrophy showed no essential differences, although increases in nuclear length were reported to be significantly greater in adult hypertrophy.

Polyploidy has been demonstrated in myocardium of man and of some other species[39]. Development of polyploidy has been shown to be related directly to age and to the size of the heart. It is also accelerated in primates in induced hypertrophy[40]. This was assumed to indicate incomplete mitosis of myocytes in order to provide enough DNA to support the increased protein synthesis necessary for growth and hypertrophy. Species differences, however, have been reported. Polyploid myocytes have been seen frequently in man, primates and in the pig but occur rarely in cat and dog[40]. In the pig, files of nuclei have been observed within myocytes, associated with growth[16], suggesting that the myocyte DNA content is increased in this species by amitosis. This mechanism has been referred to as "nuclear hyperplasia"[16] and is thought to represent a cellular adaptation to increasing demand for protein synthesis in a situation where true hyperplasia is not possible.

Thus, histologically, growth and hypertrophy have certain recognizable features. Nuclear changes are probably more useful in recognition than are changes in cell size. However, apart from a possible difference in nuclear length, mentioned above, growth cannot be distinguished qualitatively from hypertrophy. Therefore any assessment of hypertrophy must take into account comparative gross data from other animals of the same species, including sex, age, level of activity and also the gross dimensions and mass of the heart from which the myocardial sample was taken.

Physiological and pathological hypertrophy

As considered above, hypertrophy may be regarded as physiological and essentially indistinguishable from growth except in the nature of the stimulus. Furthermore, the process has been shown to be reversible if the stimulus e.g. aortic banding, is removed[12].

An important question arises concerning the nature of hypertrophy that cannot be reversed and its relation to the decompensating heart. Pathological hypertrophy has been recently defined and distinguished from physiological hypertrophy by physiological and biochemical parameters[53] as having decreased contractile function, decreased myosin ATP-ase activity and decreased cyclic AMP. In physiological hypertrophy the levels of these parameters are normal or increased. The factors and mechanisms determining the evolution of either type are complex but depend on the nature of the stimulus and the species, age and health of animal.

Pathological hypertrophy is encountered in situations where the stimulus is prolonged and intense. It has been attributed to the disproportionate biosynthesis of organelles and proteins[53]. Hypoxia, resulting from the disproportionate growth of myocytes and capillary supply, has been proposed as another possible mechanism[42].

Morphological changes, if any, which might be correlated with this definition of pathological versus physiological hypertrophy have not yet been investigated. However, 3 distinct phases in hypertrophy have been proposed[36] as:

1. Increased energy and protein production.
2. A stable state.
3. Gradual exhaustion of protein synthesis associated with myofibrillar damage and myocyte atrophy.

This third stage may correspond to pathological hypertrophy. It has been demonstrated[33] that myocyte degeneration occurs commonly in hypertrophic human hearts and it was proposed that these degenerated cells are responsible for the poor cardiac performance associated with the 3rd stage of ventricular hypertrophy.

The implication, therefore, is that myocytes degenerate as a final stage in extreme hypertrophy where no further adaptation to increased haemodynamic load is possible, leading to necrosis and associated fibrosis. This might be viewed as marking an extreme stage in a cellular adaptation to injury.

Pattern in asymmetrical hypertrophic cardiomyopathy

The hypertrophic myocardium of the interventricular septum in hearts with asymmetrical hypertrophic cardiomyopathy shows a unique pattern microscopically, which is apparently distinct from other types of hypertrophy. The pattern has been described in man[15 38], cats[30 48] and dogs[31].

The essential histological feature in all these species is a very irregular arrangement of myocytes in the septum, which run in all directions. The myocytes are shortened and show features of extreme hypertrophy, being up to 100 μm in diameter with bizarre shaped nuclei, often with a peri-nuclear halo. There is a great increase in glycogen, particularly in the peri-nuclear halo, which has been regarded as a feature of diagnostic value. The hypertrophic myocytes are separated by increased fibrous tissue.

An abnormal, haphazard organisation of the myofibrils has also been described from EM studies of human material[15]. Scanning electron microscopy (SEM) has also been recommended for visualizing the pattern[49]. In tissue from patients with non-obstructive hypertrophic cardiomyopathy, SEM showed the abnormal disposition of the myocytes very strikingly.

The origin of the pattern is uncertain. The abnormal disposition of myocytes in the septum may be the primary derangement, probably genetically determined, and hypertrophy, necrosis and fibrosis result because the disposition of the fibres makes efficient contraction impossible. Alternatively, the abnormal disposition of myocytes may represent a reaction to the presence of fibrous tissue laid down, for reasons unknown, as the initial change. A similar pattern has been seen related to myocardial scars of ischaemic origin with no evidence of hypertrophic cardiomyopathy. It has been suggested[2] that the fibre disarray in these instances is secondary to the scar, possibly as a result of a mechanical "splinting"

effect on the remaining myocardial fibres. Another explanation offered for the pathogenesis of the fibre disarray in hypertrophic cardiomyopathy is that it arises through isometric cardiac contraction. This suggestion is supported by the finding of a similar pattern in infant hearts with pulmonary or aortic value atresia[9]. Isometric myocardial contraction occurs in both these conditions.

INFLAMMATORY CHANGES

Classification of myocarditis according to pathogenesis is limited by ignorance of the pathogenetic processes in myocardial necrosis and myocarditis. However, some morphological patterns are recognizable which have been associated with a particular pathogenesis. For example, certain patterns occur consequent to necrosis and have been shown to be dictated by the type of necrosis[44]. Patterns with an alternative pathogenesis have also been investigated.

Inflammatory changes consequent to necrosis

Sequential studies of myocardial infarcts[4 5] have shown that coagulation necrosis is followed rapidly by intense neutrophil infiltration and oedema with tissue disruption and, at a later stage, by macrophage infiltration and phagocytosis of debris.

By contrast, coagulative myocytolysis and colliquative myocytolysis are followed by minimal neutrophil reaction[4 5 43] and there is very little tissue disruption. There follows only macrophage activity with phagocytosis of necrotic cellular debris.

Granulomatous myocarditis

A distinct pattern of myocarditis is characterized by myocyte damage and necrosis associated with predominantly round cell infiltrates, comprising mainly lymphocytes and/or histiocytic types and, rarely, giant cells[28]. This pattern has been referred to as granulomatous myocarditis.

The pattern appears to be associated with a variety of conditions, both natural and experimental, including acute viral and bacterial infections, chronic viral and parasitic infections, in rheumatic heart disease and also in heat stroke, following burns and in some types of autoimmune disease[35].

The occurrence of this common pattern in response to a large variety of noxious stimuli suggested the possibility that it represents an immune-mediated reaction in myocardium, initiated by myocyte damage. It was proposed that the initial step would be extension of damage to other myocytes by release of lysosomal enzymes and that this would be followed by production of antigenically altered material.

This concept of immune mediated myocarditis is strengthened by a number of different lines of evidence, reviewed elsewhere[27 28 29].

The granulomatous myocarditis associated with *Trypanosoma cruzi* in Chaga's disease has provided an interesting model for investigating these possible autoimmune mechanisms. One advantage for study is that the parasite is relatively large and readily seen. In both human cases and in experimental animals, many lesions in both acute and chronic stages are unrelated to the presence of the parasites[47]. By EM, a very intimate association has been described between lymphocytes and the sarcolemma of myocytes undergoing necrosis in these lesions. This was interpreted as evidence of a direct, specific lymphocytotoxicity to the myocytes[10]. The presence of an antigenic determinant common to both myocytes and *T. cruzi* has been demonstrated and it has been suggested that the basis for injury of myocytes in this disease is recognition of cross-reactive antigen of myocytes by lymphocytes sensitized to *T. cruzi*. Such a mechanism might also underlie the pathogenesis of rheumatic heart disease. Cross-reactivity has been demonstrated for certain antibodies between both heart muscle and streptococcal antigens in rheumatic heart disease[54].

Fibrosis

Myocardial fibrosis may be associated with necrosis and/or acute inflammatory exudation or it may occur independantly.

There appear to be several factors involved in stimulating the onset, rate and degree of myocardial fibrosis. These include the degree of stromal damage, effective blood supply and the influence of other processes such as hypertrophy.

1) *Stromal Damage*

Fibroplasia is influenced by the amount and type of myocardial necrosis. Following myocardial infarction[5 6] it has been shown that areas in which coagulation necrosis occurred there is intense fibroplasia with the formation of dense fibrous scars.

In the marginal areas of the infarct, where coagulative myocytolysis took place the original stroma remains with little evidence of fibroplasia. These areas may collapse, resulting in thin, linear scars. The variation in the amount of fibroplasia related to different types of necrosis reflects the degree of the original stromal damage since, in coagulation necrosis, there is severe stromal disruption associated with exudation, whereas, in the other types, there is minor exudation with good stromal preservation.

2) *Vascular Supply*

The effect of vascular supply on fibrogenesis in an injured area of myocardium has been studied in rats after surgical constriction of the coronary artery[44]. The rate and degree of fibrosis of the initial necrotic lesions produced is profoundly influenced by the subsequent removal of the coronary artery constriction. This resulted in more rapid fibroplasia with, ultimately, smaller scars than in rats where the coronary artery constriction was not removed. Vascular influences such as this are relevant to the important subject of the healing of human myocardial infarcts, although the beneficial effects of natural and artificial myocardial revascularization are yet to be explored fully.

3) *Hypertrophy*

Association of myocardial fibrosis with pathological hypertrophy and in hypertrophic myopathy has already been considered. Another possible way in which hypertrophy may influence fibrogenesis has been illustrated by experimental work on a myopathic strain of Syrian hamsters[6]. In this strain of hamsters there is a consistent, spontaneous development of areas of focal cardiac myocytolysis which eventually heal as small focal, fibrous scars. There is compensatory hypertrophy but congestive heart failure progresses to death. When myopathic hamsters were rendered parabiotic with

healthy ones, it was shown by various parameters that parabiosis, while it did not affect the progress of myocardial fibrosis, did have some protective effect on the myopathic heart, so that hypertrophic changes and congestive failure did not develope. The cardiac pathology of the parabiosed hamsters was subsequently studied. Scars were very much more numerous and extensive in the parabiosed hamster hearts than in the non-parabiosed ones. The explanation offered is that hypertrophic changes in the compensating heart conceal or prevent a large proportion of scar formation, the full degree of which is apparent only in the heart protected from hypertrophy through parabiosis.

Thus, in assessing fibrosis, the possible influence of various factors in the development of fibrosis, independant of the original stimulus, must be considered before making any inferences on the nature and severity of the original lesions.

Although, as has been stated, fibrosis represents only an end-stage common to numerous possible previous events, certain histologically distinct patterns of fibrosis have been distinguished which may have some diagnostic significance. Four distinct types have been described in human autopsy material[2]:

1. *Microscopical scars.* These are interpreted to represent areas of replacement fibrosis, indicating previous myocyte necrosis.
2. "*Interfibrosis*". This term was applied to a diffuse interstitial fibrosis with fine collagen, separating and encircling individual myocytes. The cause is unclear.
3. *Perivascular fibrosis.* This pattern represents an increased fibroplasia related to myocardial vascular adventitia and suggests a previous perivascular inflammatory process. In man this pattern would suggest sarcoidosis or rheumatic heart disease.
4. *Flexiform fibrosis.* This corresponds to the pattern described above in relation to asymmetrical hypertrophy.

Thus, recognition of certain patterns of fibrosis is possible. Although the associations of the different patterns are not very specific, recognition of the prevalence of one or more types may assist in histopathological diagnosis of certain myocardial disorders[2].

Anitschow cells and Ashoff Bodies

Ashoff bodies represent a myocardial reaction apparently unique to rheumatic heart disease. They are focal lesions characterized by lymphocytic cell infiltrates and large, sometimes multinucleated, cells with cytoplasmic processes[51]. Anitschow cells are intimately associated with these lesions.

The pathogenesis of this apparently specific reaction, and, particularly, the origin of the Anitschow cell, are still disputed.

Anitschow cells are not specific to Ashoff bodies and have been described in myocardial granulation tissue and following a variety of natural and experimetal forms of myocardial necrosis. However, in these lesions they occur singly and no structures resembling Ashoff bodies are present.

A mesenchymal origin has been proposed. This is based on the failure to find any features of myocytes such as myofilaments or nucleoli in EM studies. Furthermore, histochemical studies and topographical data suggest a macrophage function. In a sequential EM study of rat hearts undergoing granulation following penetration by silk sutures[41], morphological evidence was presented in favour of an origin from either pericytes or vascular endothelium.

An alternative theory supports a myocyte origin. Histological and EM evidence[35] from lesions of rheumatic fever have been presented to suggest that Anitschow cells arise from nuclei of myocytes which have previously undergone lysis. It is claimed that the nuclei, and, presumably, some cytoplasm, from these lysed cells migrate out of the original intact sarcolemma and subsequently divide, with redifferentiation into myocytes.

The available morphological evidence does not appear to be conclusive for either theory. The point, however, is important to an understanding of the pathogenesis of Ashoff bodies. If the Anitschow cell arises from mesenchyme, the reaction can be considered a unique type of granuloma. However, if it arises from the myocyte, the lesion would represent attempted myocardial regeneration.

REACTION PATTERNS IN CONDUCTIVE TISSUE

Conductive tissue has been considered less susceptible to ischaemic damage than is ordinary myocardium[19 45]. An explanation proposed is that conductive tissue is less dependant on aerobic metabolism so that it is relatively resistant to damage due to hypoxia or ischaemia[19].

Evidence of necrosis involving the conductive tissue has been found recently. Critical histological studies of human hearts with early myocardial infarction and with arrhythmias[45] have shown that, where there was widespread coagulative myocytolysis, evidence could also be found of coagulative myocytolysis in conductive tissue in 8 of the 15 hearts studied. The possible connection was considered between such conductive tissue lesions and arrhythmias, and also the proportion of sudden, unexplained deaths that occurs early in myocardial infarction.

Contrary to the concept of relative resistance of conductive tissue to ischaemic damage, certain patterns suggest that, in some situations, it is relatively sensitive to injury. Idiopathic bundle branch fibrosis (IBBF), a cardiopathy associated with some cases of chronic, complete heart block in old people, provides such an example. In IBBF there is gradual destruction of conductive tissue and associated fibrosis. The initial changes have been described as vacuolation and hyaline clumping of the special fibres, leading to degeneration and collapse of the sarcolemma, followed by mononuclear cell infiltration and replacement by fibrous tissue[13]. Thus, the conductive tissue in this condition displays a full sequence of tissue reactions to injury. The related vessels appear normal and the cause is unknown.

Chronology of changes

Useful assessment of a reaction pattern in a tissue at any stage is dependent on knowledge of the time scale required for the changes seen to occur. Little chronological data is available in cardiac pathology and it is concerned mostly with early changes associated with infarction. Some sequential observations have also been made, however.

The intervals quoted below can be regarded only as rough yardsticks, and their considerable variation under different conditions is, in itself, a striking and noteworthy feature. The picture is complicated further by the ongoing, active nature of some myocardial reaction pat-

effect on the remaining myocardial fibres. Another explanation offered for the pathogenesis of the fibre disarray in hypertrophic cardiomyopathy is that it arises through isometric cardiac contraction. This suggestion is supported by the finding of a similar pattern in infant hearts with pulmonary or aortic value atresia[9]. Isometric myocardial contraction occurs in both these conditions.

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Ashoff bodies represent a myocardial reaction apparently unique to rheumatic heart disease. They are focal lesions characterized by lymphocytic cell infiltrates and large, sometimes multinucleated, cells with cytoplasmic processes[51]. Anitschow cells are intimately associated with these lesions.

The pathogenesis of this apparently specific reaction, and, particularly, the origin of the Anitschow cell, are still disputed.

Anitschow cells are not specific to Ashoff bodies and have been described in myocardial granulation tissue and following a variety of natural and experimetal forms of myocardial necrosis. However, in these lesions they occur singly and no structures resembling Ashoff bodies are present.

A mesenchymal origin has been proposed. This is based on the failure to find any features of myocytes such as myofilaments or nucleoli in EM studies. Furthermore, histochemical studies and topographical data suggest a macrophage function. In a sequential EM study of rat hearts undergoing granulation following penetration by silk sutures[41], morphological evidence was presented in favour of an origin from either pericytes or vascular endothelium.

An alternative theory supports a myocyte origin. Histological and EM evidence[35] from lesions of rheumatic fever have been presented to suggest that Anitschow cells arise from nuclei of myocytes which have previously undergone lysis. It is claimed that the nuclei, and, presumably, some cytoplasm, from these lysed cells migrate out of the original intact sarcolemma and subsequently divide, with redifferentiation into myocytes.

The available morphological evidence does not appear to be conclusive for either theory. The point, however, is important to an understanding of the pathogenesis of Ashoff bodies. If the Anitschow cell arises from mesenchyme, the reaction can be considered a unique type of granuloma. However, if it arises from the myocyte, the lesion would represent attempted myocardial regeneration.

REACTION PATTERNS IN CONDUCTIVE TISSUE

Conductive tissue has been considered less susceptible to ischaemic damage than is ordinary myocardium[19 45]. An explanation proposed is that conductive tissue is less dependant on aerobic metabolism so that it is relatively resistant to damage due to hypoxia or ischaemia[19].

Evidence of necrosis involving the conductive tissue has been found recently. Critical histological studies of human hearts with early myocardial infarction and with arrhythmias[45] have shown that, where there was widespread coagulative myocytolysis, evidence could also be found of coagulative myocytolysis in conductive tissue in 8 of the 15 hearts studied. The possible connection was considered between such conductive tissue lesions and arrhythmias, and also the proportion of sudden, unexplained deaths that occurs early in myocardial infarction.

Contrary to the concept of relative resistance of conductive tissue to ischaemic damage, certain patterns suggest that, in some situations, it is relatively sensitive to injury. Idiopathic bundle branch fibrosis (IBBF), a cardiopathy associated with some cases of chronic, complete heart block in old people, provides such an example. In IBBF there is gradual destruction of conductive tissue and associated fibrosis. The initial changes have been described as vacuolation and hyaline clumping of the special fibres, leading to degeneration and collapse of the sarcolemma, followed by mononuclear cell infiltration and replacement by fibrous tissue[13]. Thus, the conductive tissue in this condition displays a full sequence of tissue reactions to injury. The related vessels appear normal and the cause is unknown.

Chronology of changes

Useful assessment of a reaction pattern in a tissue at any stage is dependent on knowledge of the time scale required for the changes seen to occur. Little chronological data is available in cardiac pathology and it is concerned mostly with early changes associated with infarction. Some sequential observations have also been made, however.

The intervals quoted below can be regarded only as rough yardsticks, and their considerable variation under different conditions is, in itself, a striking and noteworthy feature. The picture is complicated further by the ongoing, active nature of some myocardial reaction pat-

terns, in which acute, subacute and chronic processes may overlap. In any process in which necrosis has been occurring for longer than a few days, acute, subacute and chronic changes will all be taking place simultaneously, making interpretation very difficult. The species must also be defined. Intervals recorded for rats, for example, may be significantly shorter than equivalent intervals for man.

Irreversible damage through ischaemia has been studied by ligating the left coronary artery of dogs. The time elapsed for irreversible damage to occur was approximately 20 minutes[22].

Following infarction, the development of wavy fibres has been claimed to take only a few systoles, corresponding to a few seconds[8]. Loss of dehydrogenase activity was detectable by the nitroblue tetrazolium test 1½-6 hours after induced infarction in rats and 1-5 hours in man[14]. Histological evidence of necrosis in infarction was not recognizable until 12-24 hours[32]. In experimental rats, it was detectable at 24 hours, with a peak at 48 hours[44].

The zonal lesion was detectable in dogs 15-45 minutes after the onset of haemorrhagic shock[34].

Necrosis has been recognized in rat hearts as early as 1 hour after experimental pulmonary embolism[11]. Coagulative myocytolysis, as recognized by eosinophilic banding and nuclear pyknosis can develope within 24 hours of human cardiac surgery[43]. In canine hearts it developed 30-60 minutes after restoration of blood supply following experimental ischaemia[22].

Aortic banding studies in rats[3] have shown an increase in myocyte size detectable within 20 hours. However, what was considered as a true hypertrophy, characterized by re-establishment of normal cytoplasmic organellar proportions, was only complete at 8 days.

Neutrophil infiltration occurs from 12-24 hours after infarction in man[32] and phagocytosis of debris by histiocytes occurs as early as 72 hours after infarction in the rat[44]. In this last study a granulomatous reaction was apparent after 7 days and healing, with a linear scar, was apparent within 16 weeks. Other figures, however, suggest a much shorter time scale for necrosis and sequelae.

Macrophage activity has been seen as early as 24 hours following both human cardiac surgery[43], and experimental embolism in rats[11]. In the rats replacement of the right ventricular myocardium by connective tissue was reported within 3 days of embolism[11]. This short interval was attributed to preservation of the vascular framework.

CONCLUSION

It is evident that certain reaction patterns are recognizable histologically in myocardium. Equally evident, however, is the complexity of factors influencing these patterns so that a strict aetiological interpretation is rarely justifiable.

Certain features do give a clue to pathogenesis and interpretation can be made in the knowledge of all the possible factors, exogenous and endogenous, influencing the pathogenesis.

The foregoing illustrates some practical difficulties and potential pitfalls in microscopical interpretation of myocardial pathology. Increased clarity awaits further research, particularly into aspects of pathogenesis.

The veterinary importance of this field is underlined by the incidence and variety of plants and infectious agents, particularly in southern Africa, capable of causing myocardial injury in domestic animals.

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BOOK REVIEWS

BOEKRESENSIES

VETERINARY ASPECTS OF FELINE BEHAVIOUR

BONNIE BEAVER

1st Edn. C.V. Mosby Company, St. Louis, 1980 pp. viii and 217, illustrations 279, Price not stated
(ISBN 0-8016-0542-3)

A comprehensive study of feline behaviour which first deals with the sensory and neural origins of behaviour, while the subsequent chapters describe social, sexual, feeding, excretory, locomotive and grooming behaviour patterns. In discussing these aspects the authoress commences the description with the behaviour patterns of the foetus and neonate and continues through to adulthood. Particular attention is given to the neurological development of each behaviour patern. There is a comprehensive and up-to-date bibliography at the end of each chapter which greatly enhances the value of the book.

I must admit that I read this book with mixed feelings. Some of the chapters such as that on locomotion are outstanding, but the interpretation of some of the behaviour patterns are unacceptable to me personally. On page 55 it is stated "A cat may purr in almost any situation, including just before death, which probably reflects a state of euphoria similar to that experienced by terminally ill humans".

This book will be of value to ethologists but is of limited value to the practising veterinarian as the behaviour patterns described and case presentations are based on observations of cat colonies rather than that of the domestic pet in a home situation.

Maureen K. Baker

CASE REPORT GEVALVERSLAG

UNILATERAL ORCHITIS IN A BULL CAUSED BY *BRUCELLA ABORTUS* BIOTYPE 1

C.J.V. TRICHARD, S. HERR, S.S. BASTIANELLO and D. ROUX*

ABSTRACT: Trichard C.J.V.; Herr S.; Bastianello S.S.; Roux D. **Unilateral orchitis in a bull caused by *Brucella abortus* biotype 1.** *Journal of the South African Veterinary Association* (1982) **53** No. 1 60-62 (En) Veterinary Research Institute, 0110 Onderstepoort, Republic of South Africa.

A case of *Brucella* orchitis in a Brahman bull is described. Positive serological titres were detected 1 month before clinical symptoms became obvious, whereas the seminal plasma only contained detectable levels of antibody after the onset of orchitis. It is concluded that serological assays on the seminal plasma alone are inadequate to detect sub-clinically infected animals, and consequently particularly bulls that are intended for use in AI centres should be subjected to a full battery of serological tests as well, before they are certified to be free of brucellosis. The pathology seen in this case was an acute, necrotic orchitis characterized by the presence of numerous granulomas containing the *Brucella* organisms centrally.

CLINICAL PROGRESS OF THE INFECTION

A hemiorchidectomy was done by a private practitioner on a 4-year-old Brahman bull in September 1980 and testicular tissue was submitted to the Veterinary Research Institute, Onderstepoort. Post-operative complications set in and were only resolved by January 1981, at which stage the fate of the remaining testicle became of particular concern. On 24 February 1981 an acute swelling of the other testicle was noticed. The bull was slaughtered on 3 March 1981.

SEMEN EVALUATIONS

Semen was collected on 6 January 1981 by electro-ejaculation. A 5 ml sample was obtained which was thin and milky, its pH was 6,5 and its motility 1. It contained 25 % live sperm but no leukocytes. Subsequent to the development of the acute orchitis on 24 February 1981, semen was again collected. The evaluation of this sample, described as thick yellow pus, revealed leukocytes exclusively.

BACTERIOLOGICAL ISOLATIONS

Brucella abortus biotype 1 was isolated from the testicular tissue submitted after the hemiorchidectomy. Isolation attempts of bacteria from the semen collected on 6 January 1981 were unsuccessful. *Brucella abortus* biotype 1 was isolated from the semen collected on 24 February 1981 and from the testis, epididymis, seminal vescicles, ampullae, iliac and lumbar lymph nodes after slaughter.

SEROLOGICAL RESULTS

Serum taken on 6 January 1981 proved positive for brucellosis by the rose bengal test (RBT), with titres of 186 and 86 International Units/ml (IU/ml) in the serum agglutination test (SAT) and complement fixation test (CFT) respectively. Serological tests on the seminal plasma, however, were negative on this date. After the episode of acute orchitis on 24 February 1981 serum was once again positive in the RBT, with titres of 134 and 120 IU/ml in the SAT and CFT respectively. On this day the seminal plasma was also positive in the RBT, with titres of 40 and 98 IU/ml in the SAT and CFT.

PATHOLOGY

Gross pathology

Testes

White to yellow strands 1-2 mm wide traversed the testis on the cut surface. Between these strands the tissue was swollen and oedematous. Numerous pinpoint, yellow foci (considered to be either granulomas or foci of caseous necrosis) were disseminated throughout the testicular tissue.

Epididymis

This tissue revealed extensive fibrosis around the periphery and in the interstitium. In some places the fibrous tissue formed thick, white bands up to 7,5 mm in diameter.

Histopathology

Sections of the testis and epididymis were stained for histo-pathological examination with haematoxylin and eosin (HE). In addition, the testicular sections were stained with Giemsa's Gram's[2] or Stamp's[1] (modified Ziehl-Neelsen's) stains.

Testes

Almost the entire testis had undergone necrosis and there was complete loss of normal architecture. Extensive, well-delineated areas of acute, fibrinopurulent inflammation bordered by a narrow rim of suppurative tissue were present throughout the testis (Fig. 1). Surrounding these acutely inflamed areas were wide tracts of a more chronic, cellular exudate. This latter exudate was composed predominantly of plasma cells, but macrophages, fibroblasts and isolated neutrophils were also present, as well as varying amounts of collagen fibres (Fig. 2). A few remaining, isolated, atrophic, seminiferous tubules were still present within these tracts of inflammatory tissue. The tubules showed no indication of any spermatogenic activity and were lined only by a layer of Sertoli cells (Fig. 2).

*Veterinary Research Institute, 0110 Onderstepoort.

0038-2809/82/01/0060-0062

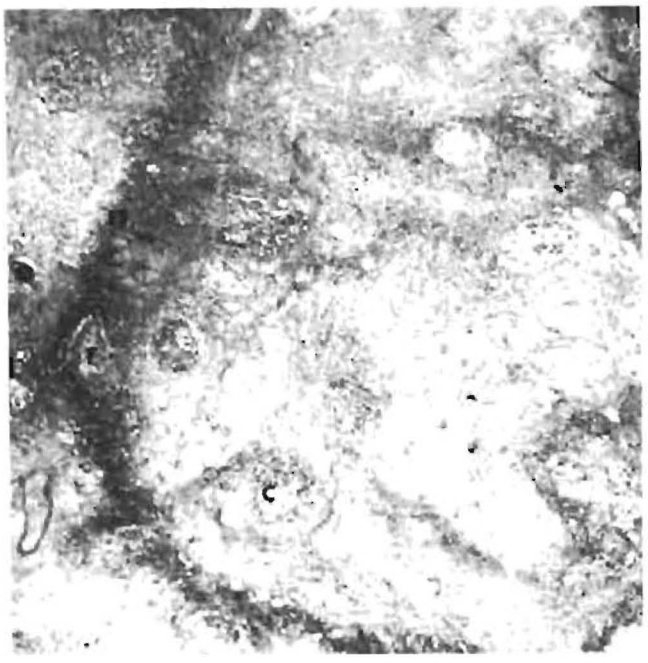

Fig. 1: A focus of fibrinopurulent inflammation bordered by a zone of neutrophils (n). Note granulomas within the centre (c) and in the rim (r) of the focus. X30

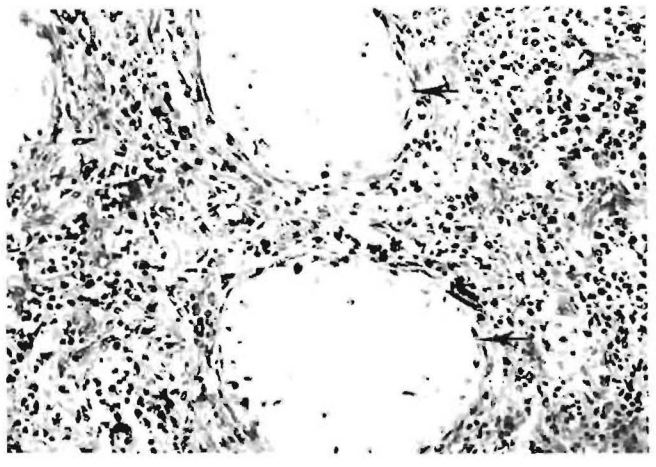

Fig. 2: Atrophic seminiferous tubules lined only by Sertoli cells (arrows) within a zone of cellular inflammatory exudate. X180

Several granulomas, scattered within the acutely inflamed areas as well as within the suppurative tissue rim bordering these areas were present (Fig. 1). They were composed of 3 zones: an inner zone of macrophages having abundant eosinophilic cytoplasm and containing numerous basophilic organisms, a middle zone consisting mostly of neutrophils, but also some lymphocytes and a peripheral zone of lymphocytes, fibroblasts and collagen fibres (Fig. 3).

In HE stained sections, numerous indistinct organisms were visible in the cytoplasm of the macrophages in the centre of these granulomas. The organisms stained negatively with Gram's stain[2] but were acid-fast with Stamp's stain. In sections stained with Stamp's stain numerous bright-red organisms, round to oval in shape, were seen tightly packed within the macrophages. The bacteria were also visible in sections stained with Giemsa, but did not stain as brightly as the organisms in sections stained with Stamp (Fig. 4).

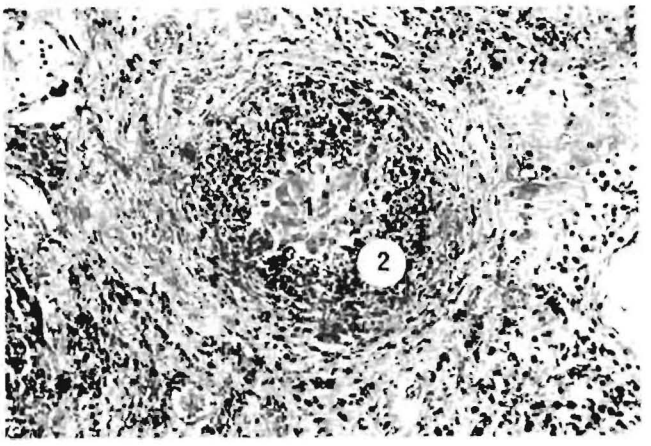

Fig. 3: A typical granuloma: 1. inner zone of macrophages containing bacteria; 2. middle zone composed of neutrophils and lymphocytes; 3. peripheral zone of fibroplasia. X75

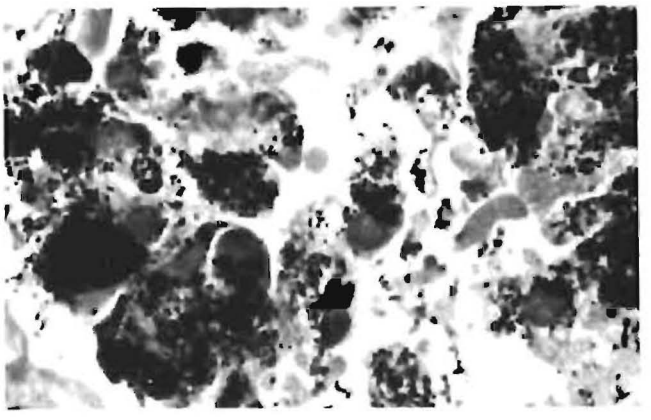

Fig. 4: Round to oval bacteria packed in the cytoplasm of macrophage (Stamp's stain). X1200

Epididymis

There was extensive fibrosis around and between the epididymal tubules. Focal cellular aggregates, composed of lymphocytes and plasma cells dispersed throughout the fibrous tissue were present (Fig. 5). The epithelium lining the tubules showed vacuolar degeneration and in some tubules neutrophils were seen migrating through and towards the lumens of the tubules. No spermatids were present within any of the tubule lumens but some tubules contained necrotic, cellular debris (Fig. 5).

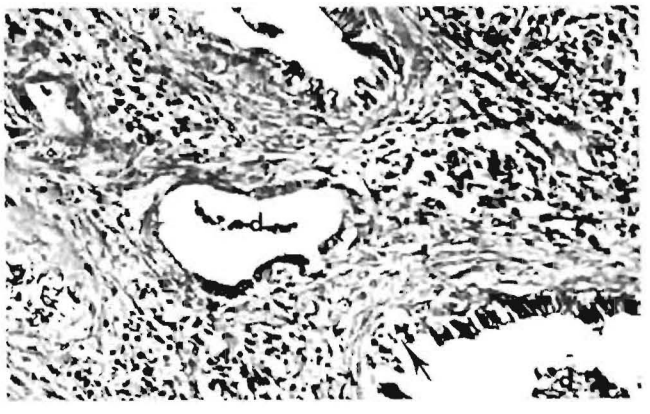

Fig. 5: Fibrosis and foci of round cells in interstitium of epididymis. Note vacuolar degeneration of tubular epithelium (arrow) and cellular debris (d) within tubular lumens.

DISCUSSION

The fact that the bull showed positive reactions to brucellosis in the serum at least a month before antibodies could be detected in seminal plasma from the second testicle and accessory glands, is a significant finding. As a blood/testes barrier is generally accepted as a physiological entity, the absence of antibody from the testis, epididymis and ampulla, until local production is stimulated by active infection in these organs, is acceptable or, alternatively, the breakdown of this barrier by the infection is an equally tenable explanation. However, with the infection of the first testicle and the subsequent isolation of the organism from the seminal vescicles it is only logical to assume that these organs were infected from the start. Even if a blood/seminal vescicle barrier is postulated, it becomes difficult to explain the absence of antibody in seminal plasma in the presumed presence of active infection in these organs, unless their capacity for local antibody production is either absent or minimal or the infection had not yet caused barrier breakdown. Whatever the explanation of this phenomenon, the practical implication is that serological tests on seminal plasma alone are insufficient to eliminate the possibility of a bull carrying the infection, and a battery of tests is indicated. This becomes additionally important where the bull is intended for use in artificial insemination as transmission of *Brucella* infection by this means is certainly not uncommon.

There are only few reports describing the pathology of brucellosis of the genital tract of bulls. Most of them describe either a unilateral or bilateral orchitis together with an epididymitis and seminal vesiculitis[3 5 7 8]. The lesions present in bulls and the African buffalo have been described by various authors to be by nature either necrotic or granulomatous[4-6]. The granulomas were characterized by a central zone of necrosis often accompanied by mineralization and surrounded by a zone of lymphocytes, histiocytes and giant cells of the Langhans type[4-6]. In the bull described in this report, the granulomas encountered in the testis were similar, but they showed neither central mineralization nor giant cells.

In the majority of cases described in the literature, no *Brucella* organisms were found in histopathological sections of various parts of the bull's genital tract. However, McCaughey & Purcell[5], when examining sections (stained with a modified Ziehl-Neelsen's stain) of the epididymis of a bull with brucellosis, noted small, oval, red organisms singly or in clumps in the centre of granulomas. In the bull described in this report, similar acid-fast organisms (Stamp's stain) were seen as tightly packed, round to oval bacteria within macrophages in the centre of the testicular granulomas. The bacteria were also visible with Gram's and Giemsa's stains but were less distinct with these stains than the Stamp's stain.

ACKNOWLEDGEMENTS

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BOOKS RECEIVED BUT NOT REVIEWED

NATIONAL DECISION-MAKING FOR PRIMARY HEALTH CARE

A study by the UNICEF/WHO Joint Committee on Health Policy. World Health Organisation, Geneva, 1981 pp 69, Publ. price not stated ISBN 92 4 156009 X

KLINISCHE PROPÄDEUK DER INNEREN KRANKHEITEN UND HAUTKRANKHEITEN DER HAUSTIERE

W. JAKSCH und E. GLAWISCHNIG

Verlag Paul Parey, Berlin and Hamburg, 1981 pp 273, Price DM48 ISBN 3-489-62716-4

CASE REPORT **GEVALVERSLAG**

GROSS HORN MALFORMATION IN AN AFRICAN BUFFALO (*SYNCERUS CAFFER*)

V. DE VOS*

ABSTRACT: De Vos V. **Gross horn malformation in an African buffalo (*Syncerus caffer*).** *Journal of the South African Veterinary Association (1982)* **53** No. 1, 63-64 (En) National Parks Board, Private Bag X402, 1350 Skukuza, Republic of South Africa.

A case of grossly malformed horns in a free-living African buffalo bull is described and photographically presented. The effect of the malformation on feeding behaviour is briefly discussed.

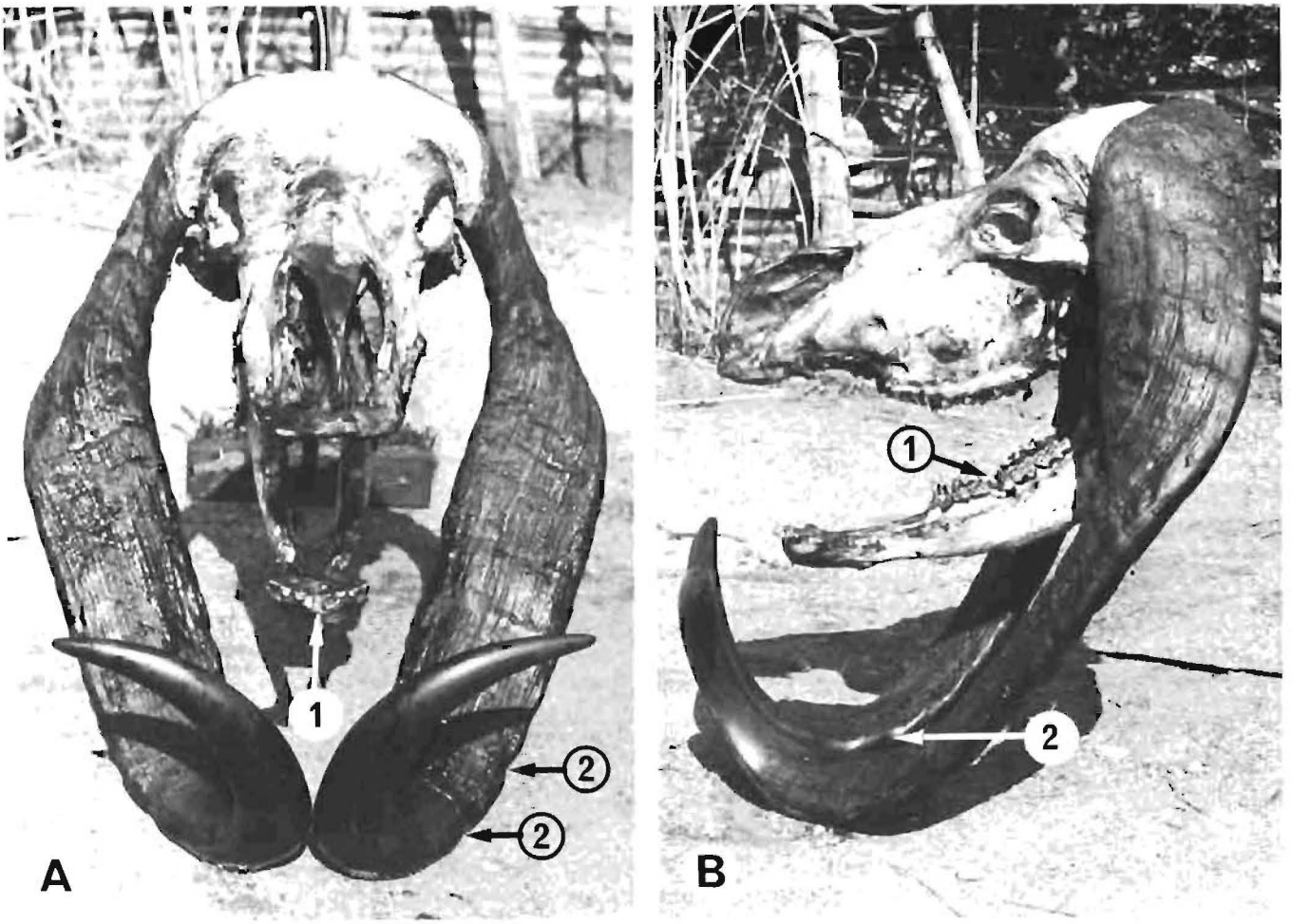

Fig. 1: Front (A) and side view (B) of an African buffalo bull with malformed horns. 1. Excessive wear of central incisors and first molars. 2. Smoothly worn grooves. Note the absence of a horn boss.

HISTORY

During June 1980 a free-living African buffalo (*Syncerus caffer*) with grotesquely malformed horns was noticed near Punda Milia Rest Camp in the Kruger National Park, South Africa. Being obviously such a rarity and due to the fact that buffaloes were being culled in that area during the time, it was decided to kill the animal in order to collect and preserve the "trophy".

*National Parks Board, Private Bag X402, 1350 Skukuza.

CLINICAL FINDINGS

Figures 1(a) and 1(b) adequately depict the macroscopic findings of the case.

DISCUSSION

Judging from a normally developed African buffalo (Fig. 2) the horns of the case depicted by Fig. 1(a) and (b) must be considered grossly malformed. Instead of having grown sideways as in a normal case (Fig. 2) the

0038-2809/82/01/0063-0064

horns swept downwards, forwards and towards the midline in a very symmetrical fashion. The horns were also relatively long, thin and flat and horn boss development was strikingly absent. As described by Sinclair[2] for normal African buffaloes, the horn boss is formed by a gradual thickening of the base of the horn where the bone core leaves the head. The thickening grows inward as the animal becomes older until the 2 bosses meet at the midline of the head. It seems therefore that instead of laying down horn for boss development, growth was chanelled into a horn lengthening process causing the malformation as depicted. When compared with the ordinary African buffaloes the length that was attained must be considered quite extraordinary. Best & Best[1] describe the Rowland Ward record length of front curve for African buffalo horns as 134,6 cm. For the equivalent measurement in this case, 136,8 cm and 133,2 cm were recorded for the left and right sides respectively. The left horn reading must therefore be considered a world record for the species. The average length of the 2 horns is 135 cm, also exceeding the present record. However, being a freak, this reading will probably not be recognized.

Fig. 2: An African buffalo with normal horn development as a comparison to Fig. 1. Note the sideways and upwards sweep of the horns and the presence of a heavy shield of horn, the boss, which covers the top of the head above the eyes.

No reason could be found for this malformation. The symmetry of the case excludes the possibility of local trauma earlier in life. It must therefore be ascribed to some systemic disorder. Yet, no other external morphological abnormalities could be found. It was a male animal with normally developed sexual organs in the prime of life and in fair condition. The internal organs were, however, unavailable for examination.

Judging from Fig. 1 it is quite apparent that the horns must have hampered normal eating routine considerably. This is further born out by excessive wear of certain teeth. The central incisors and first molars were worn down to such an extent that a gap or break in continuity formed (Fig. 1). Shallow but smoothly worn grooves were also noticed on both sides of the lowest turn of both horns (Fig. 1).

From this evidence the feeding routine of the animal was reconstructed. He was presumably dependent on long grass and browse, grasping the vegetable matter either in front with the aid of his incisors or on the side with his premolars and then using his horns in a sideways and upwards sweeping motion to sever the material. This explains the disproportionate wear of teeth and groove formation along the horns.

Survival and fair physical condition indicate that the animal had adapted and maintained itself quite well under natural free-ranging conditions, in spite of this obviously restrictive malformation.

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ABSTRACT: Walker, Jane B., Norval, R.A.I. & Corwin, M.D., 1981. ***Rhipicephalus zambeziensis* sp. nov., a new tick from eastern and southern Africa, together with a redescription of *Rhipicephalus appendiculatus* Neumann, 1901 (Acarina, Ixodidae).** *Onderstepoort Journal of Veterinary Research*, **47**, 87-104 (1981).

All stages of *Rhipicephalus zambeziensis* sp. nov. are described from laboratory-reared specimens of a strain originating from cattle near West Nicholson, Gwanda District, Zimbabwe. The redescription of all stages of *Rhipicephalus appendiculatus* is based primarily on laboratory-reared specimens of a strain originating from a mountain reedbuck (*Redunca fulvorufula*), Loskop Dam Nature Reserve, Transvaal, Republic of South Africa. The differences between these 2 species are discussed briefly.

Details of the life cycle of *R. zambeziensis* under laboratory conditions are given, and attempts to cross-breed this species with *R. appendiculatus* are described and discussed.

ABSTRACT: Neitz, A.W.H., Prozesky, L., Bezuidenhout, J.D., Putterill, J.F. and Potgieter, D.J.J. 1981. **An investigation into the toxic principle in eggs of the tick *Amblyomma hebraeum*.** *Onderstepoort Journal of Veterinary Research*, **48**, 109-117 (1981).

A purification procedure involving iso-electric focussing by means of which a toxic principle may be obtained in a pure form from crude egg extracts of *Amblyomma hebraeum* is described. The molecular mass of the toxin is approximately 10 000 according to sedimentation equilibrium sedimentation, Sodium dodecyl sulphate (SDS) gradient gel electrophoresis and calculations from the amino acid composition. Non-competitive proteinase inhibitory activity was found to be associated with the toxin. Histopathological lesions, observed in guinea-pigs inoculated with crude egg extracts or the purified toxin, included the following: focal areas of necrosis in the liver, with mineralization and oedema of the mucosa of the urinary bladder, and vacuolation of the lining epithelium. The genesis of the lesions in the various organs appears to be vascular.

CASE REPORT **GEVALVERSLAG**

A CASE OF DYSURIA AS A RESULT OF A COMMUNICATION BETWEEN THE URINARY BLADDER AND CORPUS UTERI IN A CAIRN TERRIER

SELMA J.E.M. VAN SCHOUWENBURG* and G.J. LOUW**

ABSTRACT: Van Schouwenburg S.J.E.M.; Louw G.J. **A case of dysuria as a result of a communication between the urinary bladder and corpus uteri in a Cairn Terrier.** *Journal of the South African Veterinary Association* (1982) 53 No. 1, 65-66 (En) 223 Bronkhorst St., New Muckleneuk, 0181 Pretoria, Republic of South Africa.

A 6½-month-old Cairn Terrier, considered to be a bilateral cryptorchid male, was presented with dysuria and urinary incontinence. This was found to be due to a congenital communication between the urinary bladder and corpus uteri and resulted in distention of the uterus with urine which could not be voided. An ovariohysterectomy was performed. The dog was found to be a genetic female with what resembled external male genitalia, i.e. a female pseudohermaphrodite.

Key words: Pseudohermaphrodite, dysuria, Cairn Terrier.

INTRODUCTION

This report deals with a "male" Cairn Terrier which was a regular patient at the time since its owners brought it in frequently for its routine vaccinations. It had been obtained from a reputable breeder and was pedigreed. When it was 6 months old, the owners expressed concern at the fact that neither of the testicles had descended. The dog had also not yet started lifting its leg when urinating, but still assumed the female squatting posture. On clinical examination, no testes could be palpated extra-abdominally and there was no visible scrotum. The prepuce was underdeveloped, but at this stage no attempt was made to examine the penis. It was assumed that the genitalia would develop further as the dog matured. Apart from this, it was in excellent health and extremely playful – to the extent of being hyperactive.

HISTORY, CLINICAL FINDINGS AND TREATMENT

When it was 6½ months old, the Cairn Terrier was presented showing signs of acute abdominal pain. It frequently squatted down in the urinating posture, seemed to be in severe pain then, and passed only a few drops of urine. It also seemed to be incontinent as urine dribbled continously from the prepuce. It had lost its playfulness and preferred hiding away in the house.

On examination, the lumbar and abdominal area was extremely tense and painful. The rectal temperature was 38,5 °C. A tentative diagnosis of cystitis was made with a possibility of trauma to the lumbar region as an alternative. An antispasmodic (Avafortan, Noristan) was injected intramuscularly and the dog was put on an oral course of antibiotics (Penbritin V, Beecham). The owners were asked to report back.

The next day there was a slight improvement and it was decided to complete the course of antibiotics.

A fortnight later the dog was brought back for reassessment. The condition had not improved. It was decided that an examination under general anaesthesia should be done in order to explore other differential diagnoses such as urolithiasis and stenosis or neoplasia of the urinary tract. A sterile sample of urine for bacterial culture could also be taken if necessary.

A short acting intravenous barbiturate (Intraval Sodium V, Maybaker) was administered. When attempting to pass a catheter into the bladder it was found that the dog had no macroscopically visible penis or os penis. The small genital opening was approximately 20 mm caudal to the preputial opening. A catheter of only very small diameter could be passed. After the bladder had been emptied by manual expression, 20 ml of a contrast medium (Urografin 76 %, Schering) was injected into the bladder via the catheter. Radiographs were taken. These showed that the bladder communicated with a vesicular structure which was, at the time, thought to be a distended ureter (Fig. 1).

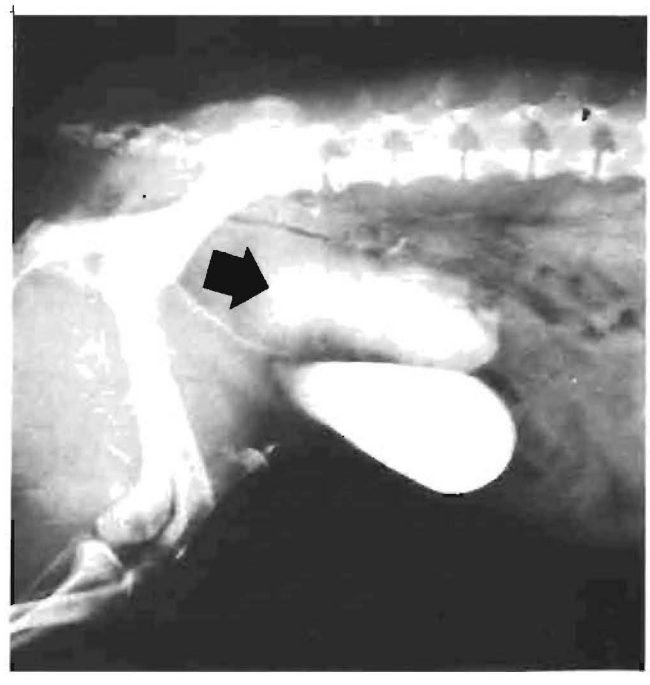

Fig. 1: Radiograph showing contrast medium in the urinary bladder as well as in a communicating vesicular structure (arrow).

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At this stage it was suspected that the dog was an intersex. Blood smears were examined and the presence of drumsticks attached to the nuclei of some neutrophiles showed that the dog was, in fact, a genetic female[1].

A laparotomy was advised. However, the dog's condition improved markedly after the examination, probably because of the emptying of the bladder and the stretching of the urethra. When abdominal spasms and dysuria reappeared after about 10 days, a laparotomy was performed.

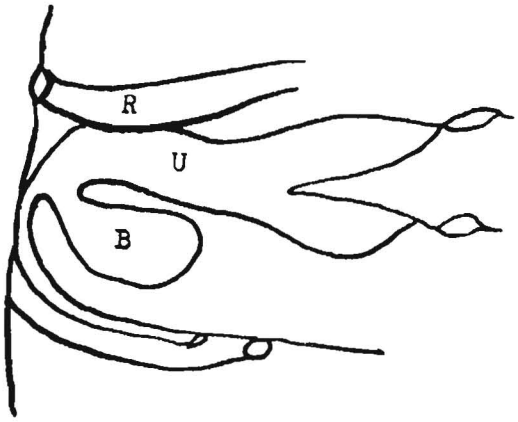

Fig. 2: Diagram showing anatomical relationship of the uterus (U), urinary bladder (B) and rectum (R).

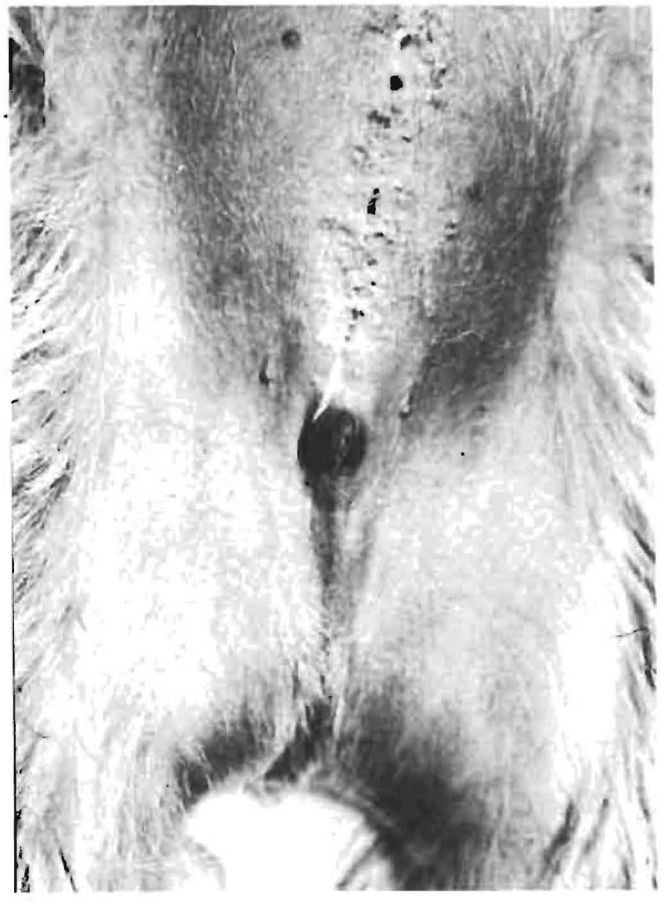

Fig. 3: Photograph of the external genitalia of the dog taken 14 days after ovariohysterectomy was performed, indicating the position of the laparotomy through which the ovariohysterectomy was performed. Note the hypoplasia of the penis and prepuce, and the absence of descended testes within the hypoplastic scrotum.

The dog had a normal uterus and ovaries, but the caudal part of the corpus uteri communicated freely with the neck of the bladder. The result of this was that both body and horns of the uterus were grossly distended with urine. Dorsally, the caudal part of the corpus uteri was adhered to the rectum but there was no communication between the two (Fig. 2).

The communication between the uterus and bladder was closed by means of a purse-string suture and an ovariohysterectomy was performed. No intra-abdominal testes were found (Fig. 3).

Histologically the uterus showed that the normal simple columnar epithelium had been compressed into a squamous epithelium by the pressure of the urine contained within the organ. Both ovaries contained developing follicles and appeared normal on histological examination.

After surgery the dog made an uneventful recovery. Within weeks it regained its normal playful nature.

DISCUSSION

This dog should be regarded as a gonadal female that developed certain of the male external genitalia, i.e. a prepuce. It closely resembles the case of a Cocker Spaniel described in 1965 by McFeely & Biggers, the main point of difference being that they found no communication between the vagina and male urethra[2]. This condition is thought to be caused by some masculinising influence during the prenatal development[2 3].

Attempts to trace litter-mates of the Cairn Terrier failed because the breeders refused to co-operate, presumably to protect their good reputation.

The abdominal pain experienced by the dog was caused by the distention of the uterus with urine. Clinical signs possibly appeared at 6½ months of age as the dog would have reached puberty and the continuing development of the uterus would have been under the influence of increasing oestrogen production.

ACKNOWLEDGMENTS

We wish to thank members of the Faculty of Veterinary Science, University of Pretoria, namely Mrs. S.E. van der Hoven for preparing the photographs and Professors W.H. Gerneke and C. Roos for their advice.

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SHORT COMMUNICATION **KORT BERIG**

ABSENCE OF UREA TOXICITY IN YOUNG PIGS

C. BUTTON*, J.P.J. JOUBERT† and B.P. MAARTENS†

ABSTRACT: Button C.; Joubert J.P.J.: Maartens B.P. **Absence of urea toxicity in young pigs.** *Journal of the South African Veterinary Association* (1982) **53** No. 1, 67-68 (En) Department of Physiology, Pharmacology and Toxicology, Faculty of Veterinary Science, University of Pretoria, P.O. Box 12580, 0110 Onderstepoort, Republic of South Africa.

Urea was non-toxic to a 10 week-old pig in an acute dose as high as 16 g /kg body mass. Ten % m/m urea in pig food over a period of 5 days was also without apparent deleterious effect.

Key words: Urea, toxicity, pigs.

INTRODUCTION

Urea is toxic to ruminants when ingested by mouth[3 5]. This is because urea is hydrolysed to carbon dioxide and ammonia by ruminal urease-producing micro-organisms[3]. Ammonia is lipid soluble and is absorbed into the systemic circulation. If the concentration of blood ammonia exceeds the ability of the liver to detoxify it, hyperammonaemia and the typical clinical syndrome of urea intoxication results[3 5]. As little as 0,5 g urea per kg body mass can kill an unadapted ruminant[3 5].

Horses are susceptible to urea poisoning despite being monogastric animals[2]. When approximately 3,5 g urea per kg body mass was given to 8 ponies, 7 of the 8 died in less then 12 hours. Hydrolysis of urea presumably occurred in the large intestine and significant urease activity was demonstrated in equine caecal fluid. Blood ammonia concentrations rose approximately 16-fold just before death occurred in these ponies[2].

Man and other monogastric mammals are supposedly not affected by oral urea ingestion by virtue of lack of gastrointestinal urease producing micro-organisms. In such animals urea is absorbed, circulates and is eliminated by glomerular filtration[3 5]. Intravenous urea solutions are used for osmotic diuresis in man at doses as high as 1,5 g/kg body mass[4].

In 1979 a pig growth meal became contaminated with urea. The meal as fed to piglets had an estimated 10,5 g urea/kg meal. Whilst on this meal piglets between the ages of 2 and 12 weeks developed diarrhoea and more than 460 died. A claim for losses was lodged against the feed company.

In a limited review of the literature all authorities stated that urea is non-toxic to monogastric animals providing water is freely available but no reference to a specific trial in young pigs could be found. To clear up this problem we administered various doses of urea to 3 young pigs.

MATERIALS AND METHODS

Three litter mate male cross-Landrace piglets, designated A, B and C, weighing 14, 16 and 13 kg respectively were used. They were 10 weeks old when testing started. Testing was over a period of 15 days.

The piglets were housed in individual stalls, bedded with straw and had water freely available.

Testing commenced with acute oral doses of 1, 2 and 4 g laboratory grade urea/kg body mass given to Piglets A, B and C respectively. Urea was dissolved in water and administered by dosing gun.

All 3 pigs were fed a dry pig meal containing 2,5 g % m/m urea per day for 5 days. Each piglet consumed the 1 kg food offered making daily intake approximately 25 g each. Following this, 5 % m/m urea was mixed with the pigs' food for a further 5 days. Again 1 kg of meal was offered and completed daily making daily intake approximately 50 g.

Pig B was then fed 7,5 % m/m and Pig C 10 % m/m urea in their meal for a further 5 days. At these levels acceptability of the meal seemed to decrease and the piglets took longer than usual to finish the 1 kg meal offered. Daily intake was thus 75 g and 100 g for Pigs B and C respectively.

Piglet A was given further single doses of 8 and finally 16 g urea in solution per kg body mass by mouth with one urea free day between trials. The protocols of the above trials are summarised in the table below.

TABLE 1: PROTOCOL FOR ADMINISTRATION OF UREA TO PIGLETS

Piglet	A	B	C
Day 1	1g/kg b m in solu + 2,5% m/m in meal	2g/kg b m in solu + 2,5% m/m in meal	4g/kg b m in solu + 2,5% m/m in meal
2–5	2,5% m/m in meal	2,5% m/m in meal	2,5% m/m in meal
6–10	5,0% m/m in meal	5,0% m/m in meal	5,0% m/m in meal
11	–		
12	8g/kg b m in solu	7,5% m/m in meal	10% m/m in meal
13	–		
14	16g/kg bm in solu		
15	–		

Doses are of grams of urea per kilogram body mass (b m) in solution (solu) or percentage urea mass/mass (m/m) in pigmeal.

RESULTS

The 3 piglets remained perfectly healthy throughout the trials. No symptoms suggestive of urea intoxication were seen. Diarrhoea did not occur in any of the piglets. Appetite was good throughout although Piglets B and C seemed to take somewhat longer to finish meal contain-

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ing higher levels of urea. Water intake was not measured but Piglet A was seen drinking frequently after his 8 and 16 g/kg doses. All piglets gained approximately 2 kg in body mass during the 15 day course of the trial.

DISCUSSION

The piglets in this trial showed no signs of intoxication at urea dosage rates far in excess of those alleged to have caused poisoning in the field. These findings support the contention of various writers[3 5] that urea is non-toxic to most monogastric animals. One should caution, however, that these results might not apply to pigs of all ages and on different diets. It is conceivable that urease producing organisms could occur in some numbers in the large intestine of pigs fed a diet high in roughage, and that such pigs might, like horses, be susceptible to urea given in high doses orally. In addition urea, salt and other osmotically active substances can, when occurring at high concentrations in the diets of pigs and in association with water deprivation, result in malacia of the cerebral cortex[1]. Such poisonings are the result of disturbed water and electrolyte balance and are not a manifestation of urea poisoning per se[1].

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BOOK REVIEW **BOEKRESENSIE**

BOVINE HAEMATOLOGY

O.C. STRAUB

Paul Parey, Berlin & Hamburg. 1981 pp iv 66 Figs 14 Tabs 35 Publ. Price not stated.

1 *Presentation* This little book is in fact not a book, but a combination of 4 articles which appeared in Zentralblatt für Veterinärmedizin, Series B, Vol 25, 1978, pp 14-28, 245-267 and 484-498. Therein lie its weakness and its strength. Its strength is in its brevity and accuracy which are a hallmark of the above journal. Its weakness is its lack of coherence as a direct result of binding 4 articles together. This may well be an energy and time-efficient manner of producing a book, but it is felt that with a little effort in terms of editing and rearranging a much more acceptable volume could have been produced.

2 *Content* The title, though theoretically accurate, belies the contents. To the average reader the title suggests that here, at last, is the definitive exposition on interpretative bovine haematology that veterinary science has been waiting for. However, on closer inspection it is seen to be merely a chronicle of the frustrations encountered by the Common Market of Europe in establishing a "European Community Leukosis Key", to which is appended (section IV) a series of tables comparing haematological data from 16 European cattle breeds represented by some 3 600 animals.

3 *Conclusion* The above is by no means intended to detract from the value of the publication in its own context. It is high time that someone should dispel the myth of infallibility of electronic cell counters and quantitate the inter-laboratory errors between respected central laboratories. For the research scientist and technologist using cell counters this book should be made compulsory reading.

However, the average veterinary practitioner, attempting to interpret haematological data from his patients, will find very little in this book that he can usefully apply.

F. Reyers

LETTER TO THE EDITOR **AAN DIE REDAKSIE**

PANETH CELLS IN THE PIG – A CONTROVERSIAL ISSUE

Paneth cells, normally occurring as a small group or as isolated cells in the deepest part of the duodenal crypts of Lieberkühn, have traditionally been described as absent in the intestinal crypts of the domestic pig *Sus scrofa*[1 2 3 7 9]. This widely-held belief has recently been challenged by Louwers & De Vos[5].

In a recent systematic survey of the histology of the porcine intestine, Paneth cells could be demonstrated with the aid of two different histochemical stains, namely, Mallory's phosphotungstic acid haematoxylin (MPAH)[6 8] and the phloxine-tartrazine (PT) method according to Lendrum[4].

These cells are mainly grouped along the sides in the lower half of the crypts of Lieberkühn (Fig. 1). The Paneth cell granules (p) were stained by MPAH; X 192. In the region of the basal lamina, the Paneth cells were lightly basophilic, with proximally situated nuclei (n) and secretory granules (gr) placed distally (Fig. 2). The Golgi apparatus (g) was seen to be situated just distal to the nucleus, where it appeared as a negative image because of lack of staining ability. These granules were stained with PT; X1200.

Full details of this work will appear in a report now in preparation.

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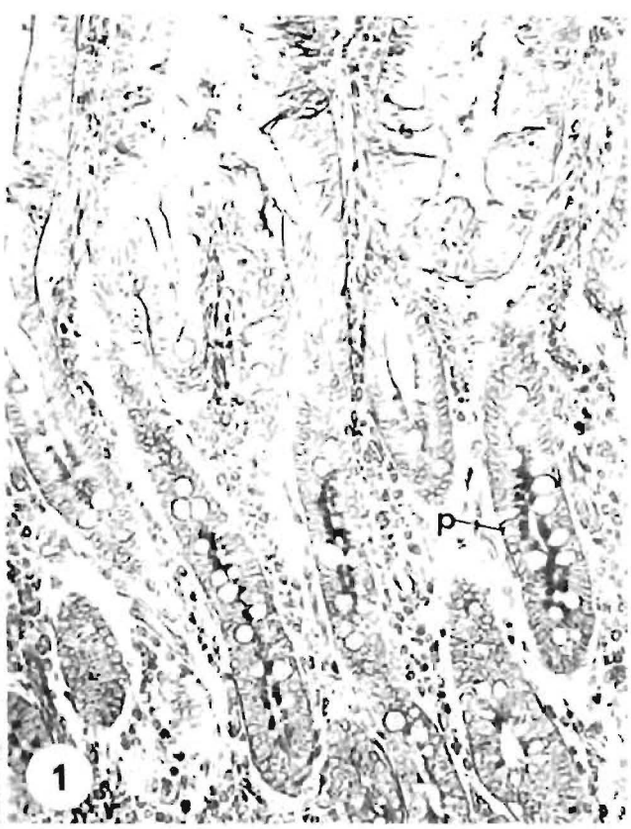

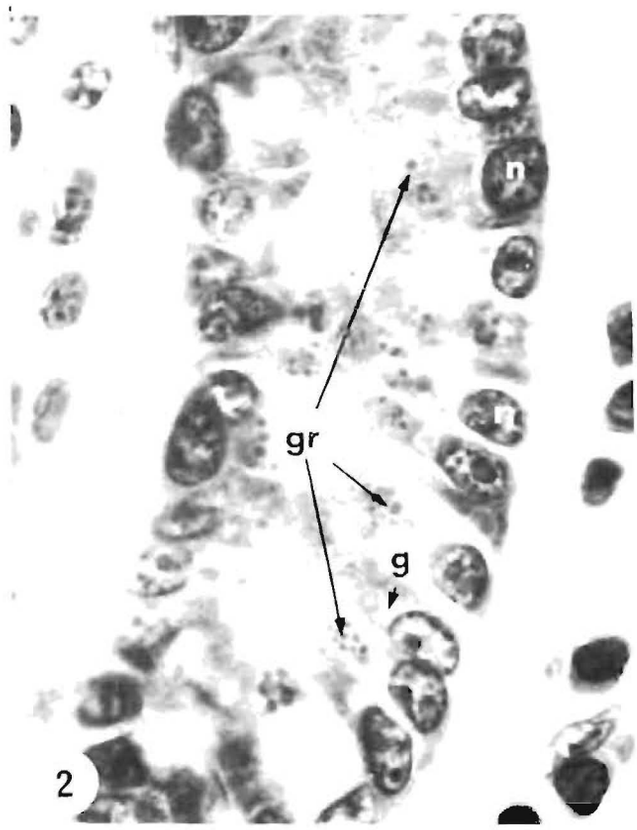

BOOK REVIEW **BOEKRESENSIE**

ESSENTIALS OF CANINE AND FELINE ELECTROCARDIOGRAPHY

LAWRENCE P. TILLEY

1st Edn. The C.V. Mosby Company, 11830 Westline Industrial Drive, St. Louis, Missouri 63141, USA. 1979 pp 337, illustrations 1188. Price not stated ISBN 0-8016-4963-3

This book provides a comprehensive review of most aspects of canine and feline electrocardiography. Its primary purpose is outlined by the author as an attempt in presenting a "practical approach to the interpretation of canine and feline electrocardiograms".

Starting with an introductory chapter on electrophysiological principles, it proceeds with an explanation of the principles of electrocardiographic recording. A step by step, well illustrated account then follows on the diagnostic approach to the electrocardiogram.

The sections on electrocardiographic interpretation in the dog and cat include information, given in a concise form, on the electrocardiographic features, differential diagnosis and therapeutics of specific entities. All conditions are highlighted with superb reproductions of electrocardiograms. The brevity of the text is sufficiently compensated for by the extensive use of references.

For the more advanced veterinary cardiologist the chapters on "uncommon complex arrhythmias" and the electrophysiologic basis of cardiac arrhythmias should make excellent and stimulating reading.

The book is concluded with a stimulating chapter on "self-assessment tracings" and a useful extensive list of references.

This well-illustrated text on canine and feline electrocardiography is highly recommended for both students and practitioners, especially for those with a keen interest in electrocardiography.

J. van Heerden

THE PERFORMANCE OF COSMETIC SURGERY BY VETERINARIANS*

INTRODUCTION

In November 1980 the Council of the South African Veterinary Association (SAVA) were informed that the Veterinary Board, the statutory body appointed to set standards of professional conduct and deal with disciplinary matters, had suspended its 2 year ban on the performance of ear cropping by veterinarians.

This reversal of the Board's earlier ruling placed the SAVA in an invidious position. It was interpreted by the public as an endorsement of the propriety of this and other cosmetic surgery procedures which had been performed by the profession in the past. It also questioned the judgement of the Association's ethical committees whose recommendations had led to the implementation of restrictions on the performance of cosmetic surgery by the Veterinary Board in 1978.

The latter aspect was of particular concern to Council because the recommendations of its ethical committees had been made after a 4 year period of deliberations on the matter.

These deliberations began in 1974 when an SAVA Committee was appointed to investigate allegations of cruelty lodged by members of the profession in respect of questionable training methods and cosmetic surgical procedures which were associated with the preparation of American Saddler horses for competitive showing. The principal complaint was directed towards the practice of "nicking" and "setting" of horses' tails in a permanently elevated position for aesthetic reasons. It was concluded that the practice caused prolonged pain and discomfort for the animal and that it was an unnecessary mutilation and should therefore be considered to be an unprofessional and unethical procedure.

A memorandum reflecting the views of the Committee was submitted to SAVA Council and at the 1976 Annual General Meeting of the Association it was resolved that "tail nicking" be abolished as a professional procedure. Deliberations on the matter had, however, focused attention on other forms of cosmetic surgery which were being routinely performed on other animal species. It was therefore decided by members that before the Veterinary Board was approached with a request that tail nicking on horses be ruled as an unethical operation, a general study of the ethical implications of cosmetic surgical procedures in all animal species be carried out.

During 1977 opinions and comments on the subject were invited from individual members of the Association and from its branches and special groups. A detailed memorandum was compiled on the subject and submitted to Council in July of that year. The memorandum recommended that the following procedures should be declared to be unethical:

(a) Ear cropping in dogs.
(b) Tail docking and tail nicking in horses.
(c) Surgical procedures which obscured any inherited abnormality without there being a clinical justification for therapeutic surgical treatment to alleviate any disability arising from such an abnormality.

The main reason for these recommendations was that in general no justification could be found for inflicting unnecessary pain and discomfort by performing unnecessary surgery. On the basis of this memorandum it was resolved at the 1977 Annual General Meeting that these procedures be declared unethical and that the Veterinary Board be formally approached to support this resolution. The Board complied with this request and in December 1978 formal notice of the Board's ruling was served upon the profession. This action, however, elicited an angry response and objections were immediately lodged by veterinarians and by animal breeders, notably those of Boxers and Doberman Pinchers in which the ear cropping operations had been customarily performed. Dissatisfaction with the ruling was brought to the notice of both the SAVA and the Veterinary Board.

Veterinarians who opposed the cosmetic surgery ban were unhappy because they felt that a statutory limitation had been imposed on their freedom to undertake any surgical procedures they wished to carry out. It was argued that this freedom was a fundamental professional right which should not be subjected to any limitations. The debate rapidly escalated into a provocative issue and the ear cropping question was therefore once again tabled for discussion at the Association's Annual General Meeting in 1979. In view of the emotive nature of the issue and largely subjective arguments put forward in favour of lifting the ban, it was decided at that meeting not to put the matter to a vote but to investigate the pros and cons of the issue for a further year.

At the Association's next Annual General Meeting in September 1980 it was resolved that the Veterinary Board be requested to review its ruling on the banning of cosmetic surgical procedures. This request was motivated amongst other things by the realisation that the ban had resulted in the procedures being performed by laymen in order to circumvent the restriction which had been placed upon the profession.

The Veterinary Board responded to the SAVA request for a review of its earlier ruling by lifting its ban on the performance of cosmetic surgery in November 1980.

TERMS OF REFERENCE OF THE SPECIAL COMMITTEE

To review all issues associated with the performance of cosmetic surgery on animals by veterinarians and submit recommendations for consideration by SAVA Council.

*A report of the Special Committee appointed by the South African Veterinary Association on 8 November 1980, under the Chairmanship of Dr J.C. Austin, to review all issues associated with the performance of cosmetic surgery by veterinarians in South Africa.

DEFINITION OF COSMETIC SURGERY

Before proceeding with this task the Committee felt that it was necessary to formulate a clear definition of the term cosmetic surgery as it is applied to surgical procedures which are performed on animals. The following definition was agreed upon:

Animal Cosmetic Surgery

Surgery which is performed on animals with the prime objective of altering the natural appearance of an animal for aesthetic reasons.

Other points pertinent to this definition were that:

(a) The procedure did not improve the health status or bodily functions in the animal subject upon which it was performed.
(b) The procedure was not carried out as an aid towards facilitating animal management or performed in the interest of the animal's well-being when maintained under normal conditions of husbandry.

In the absence of any relevant justifications for the performance of cosmetic surgery, it was by definition therefore considered to be an *unnecessary procedure*.

Examples of animal cosmetic surgery which were consistent with the terms of the definition were:

Tail nicking and setting in horses
Ear cropping in dogs
Tail docking in dogs.

IDENTIFICATION OF THE MAJOR ISSUES CONCERNED WITH COSMETIC SURGERY IN ANIMALS

In pursuing its brief, the Committee attempted an objective assessment of the most common cosmetic surgical procedures in terms of their legality, rationality and humaneness and in terms of public opinion. The Committee felt that these criteria represented the major issues which needed to be resolved by the profession in this matter. Cosmetic surgical procedures which were specifically examined in terms of these criteria included tail nicking in horses and ear cropping and tail docking in dogs.

LEGALITY OF PERFORMING COSMETIC SURGERY IN ANIMALS

The existence of laws to protect the welfare of animals at both a national and international level serve to emphasize the fact that there is wide acknowledgement that animals have legal rights.

In general, the purpose of legislation concerned with animals exist to:

(a) Prevent unnecessary suffering
(b) Control disease
(c) Provide for the control of animals.

In terms of the definition of cosmetic surgery, it must be acknowledged that cosmetic surgery is an unnecessary procedure and that it results in a period of pain and discomfort for the animal subject upon which it is performed. It would seem logical therefore that any legislation concerned with the prevention of unnecessary suffering in animals would be concerned with limiting cosmetic surgical procedures.

This premise is borne out by the existence of specific legislation to prevent certain types of cosmetic surgical procedures in member states of the European Economic Community[3]. Prohibited acts in terms of this legislation which are related to cosmetic surgical procedures include:

(a) United Kingdom (Protection of Animals Act of 1911)

Cropping of dogs' ears and the docking of horses' tails or the causation of any unnecessary suffering by doing or omitting to do any act, are defined as instances of cruelty.

(b) Denmark (Protection of Animals Act of 1950)

Docking of tails in the horse, cow and dog, and the cropping of ears of dogs over 1 week of age may be carried out only by a veterinary surgeon using anaesthesia and only for medical reasons.

(c) Netherlands (Animal Protection Act of 1944)

It is a punishable offence for any person deliberately or unnecessarily to dock horses' tails and crop dogs' ears.

(d) Sweden (Animal Protection Act of 1944)

Horses' tails may only be docked by a veterinarian to cure disease, injury or vices. Ear cropping in dogs is classified as an offence in terms of this Act.

(e) Federal Republic of Germany (Animal Protection Act of 1972, Part 4)

Anaesthesia is required for the docking of the tails of dogs over the age of 7 days. Dogs' ears may not be cropped after the third month of life. Unless for veterinary reasons, it is illegal to completely or partially amputate parts of the body of any vertebrate animal.

(g) Republic of South Africa

No specific referral is made to cosmetic surgical procedures in the statutes of the Republic of South Africa in the Animal Protection Act No. 71 of 1962. However, considerable protection is extended to animals in the broadly defined list of offences in respect of animals in Section 2 of the Act. Paragraph (r) of this section has a direct bearing on the performance of cosmetic surgery in terms of the definition adopted by the Committee for reviewing the issue.

This paragraph states that "any person who by wantonly, unreasonably, or negligently doing or omitting to do any act or causing or procuring the commission or omission of any act causes any unnecessary suffering to any animal shall subject to the provisions of this Act and any other law be guilty of an offence."

The interpretation of the word *unnecessary* is important in this context because whatsoever may be convenient, desirable or profitable to mankind may not automatically be deemed to be a necessity.

The Committee felt that the performance of cosmetic surgery could perhaps be considered to be an offence in terms of the Animal Protection Act No. 71 of 1962 and that a legal opinion on the interpretation of legislation in relation to cosmetic surgical procedure should be obtained to clarify this point.

RATIONALITY OF COSMETIC SURGERY AS A PROFESSIONAL PRACTICE

In view of the definition of cosmetic surgery adopted by the Committee and the fact that the veterinary profes-

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sion was the only scientific discipline whose primary objective was the welfare of animals, the Committee was of the opinion that the performance of cosmetic surgery such as tail nicking in horses and ear cropping and tail docking in dogs could not be considered to be anything other than a totally irrational practice.

HUMANENESS OF COSMETIC SURGERY

Humane behaviour as applied to animals is concerned with reducing the sum total of pain, fear and discomfort which is inflicted on animals by man.[5] The practice of humane behaviour is therefore concerned with eliminating or reducing the severity of these conditions and the numbers of animals affected by them as a result of any particular practice.

The principles of humane practice which are adhered to in promoting humane procedures in wild animals, domesticated animals and laboratory animals are threefold[4]:

> Elimination of all *unnecessary* procedures which cause fear, discomfort or pain.
> *Reduction* of pain and suffering by reducing unavoidably painful procedures to a minimum.
> Refinement of the techniques which are used in those procedures which have to be carried out by the application of technological and scientific advances.

In reviewing the practice of cosmetic surgery against the background of these concepts the Committee felt there was a sound justification for the elimination of all cosmetic surgical procedures on the basis of humaneness. The Committee also felt that there was little scope for refining cosmetic surgical procedures to enable the pain and discomfort which they caused to be significantly reduced. In reviewing the pain factor in these procedures the problem was almost entirely caused with the post-operative period and not the surgical procedure as such. In spite of good anaesthetic and surgical techniques in tail nicking in horses and ear cropping in dogs, the results of the procedure were clearly unavoidably unpleasant for the animal subjects upon which they were performed.

In principle, no differences could be found between the more severe types of cosmetic surgical procedures and tail docking of puppies under one week old without anaesthesia or analgesia. The latter procedure differed in that it was acutely painful when performed. The pain was, however, of short duration and the post-operative period did not appear to be noticeably unpleasant for the tail-docked animals.

The Committee felt that no veterinarian who was well-informed could honestly claim that pain and discomfort were not a feature of the post-operative phase of tail nicking in horses and ear cropping in dogs. It concluded that the ethical position of the profession would be weakened if it exhibited ignorance of these facts or if it attempted to conceal them.

In view of the divided opinions on humane issues within the profession the Committee felt it necessary to examine this aspect in more detail because the public looked to the profession for leadership and guidance in animal welfare matters.

The views of Dwight Ingle, Chairman of the Departmet of Physiology of the University of Chicago on the scientific and ethical responsibilities associated with the care and use of animals were recorded as pertinent to this matter[2].

Quoting Ingle, "it needed to be appreciated that humans were not naturally endowed with an instinct to be kind to animals. Although based in part on the conditioning of inborn emotional responses, the evolution of humaneness was associated principally with the psychological and social development of individuals. Although humaneness was acknowledged as a part of our culture, it did not always suffice to protect the interests of animals when they are used to promote human welfare." There was, he said, nothing in the process of becoming an animal technician or scientist that would ensure humaneness unless their education included attention to this problem.

The Committee felt therefore that the evolution of humaneness within the veterinary profession and society had to be based on an appropriate programme of education. To their knowledge no such programme had ever been part of any formal course of instruction at the Faculty of Veterinary Science, University of Pretoria. This omission would seem to cast doubt upon the profession's ability to evaluate animal welfare issues on a humane basis.

It was also noted that the cropping of dogs' ears was extensively employed as a teaching operation for undergraduate students in the Department of Surgery at the Faculty of Veterinary Science. This practice must have had a profound influence on the establishment of attitudes in the profession about the propriety of performing an unnecessary operation which was followed by post-operative pain and discomfort. The Committee concluded that on humane grounds there could be no justification or support for the continued performance of cosmetic surgical procedures on animals.

PUBLIC OPINION

In considering public opinion it was clear to the Committee at the outset that, with the exception of tail docking in dogs, cosmetic surgical procedures were only sought after by a very small sector of the public.

The major body representing the interests of dog breeding and showing enthusiasts – the Kennel Union of South Africa – does not permit dogs which have had their ears cropped to participate in any shows which are held under their auspices. This ruling has not been detrimental to the breed sections within the Kennel Union which cater for breeds such as Boxers, Doberman Pinchers, Great Danes. The commercial interests of ear cropping enthusiasts of these breeds therefore need not necessarily be jeopardised if the practice were discontinued.

Tail nicking is only sought after by the American Saddler enthusiasts who represent a minority group amongst equine breeders and riding, jumping and showing enthusiasts in South Africa.

Tail docking of dogs is almost universally supported and practiced by the dog breeding sector. Docked tails are stipulated in the breed standards of many dog breeds. The overwhelming weight of public opinion in favour of tail docking provided an example of the profound impact which social habits have in motivating the behaviour of society. These habits are so profoundly entrenched that it is unlikely that they will ever change. Tail docking will probably only disappear with the ex-

tinction of breeds in which it has become a man-made feature.

The Committee felt that public opinion was overwhelmingly unsupportive of ear cropping in dogs and tail nicking in horses, and that abolishment of these procedures on the basis of legal, rational and humane considerations would be accepted and supported by the public in general.

WELFARE OF ANIMALS KEPT UNDER INTENSIVE AND OTHER LIVESTOCK HUSBANDRY SYSTEMS IN RELATION TO COSMETIC SURGERY

Economic pressures combined with an increase in scientific knowledge have made it possible to intensify animal production. More animals are being kept in large units on less land than in the past. For this to be possible, a wide range of surgical procedures are carried out to aid in the management of animals and the control of disease. These include ear-tagging, horn debudding, dehorning, castration, debeaking, nose-ringing, oöphorectomisation, etc.

It was appreciated by the Committee that these operations inflict pain and discomfort on the animal subjects on which they are performed which may equal or exceed that which may occur with cosmetic surgical procedures. If such procedures are carried out on a routine basis without being absolutely necessary, they must be considered to be a contravention of the Animal Protection Act, to be irrational and to be inhumane.

The Committe recognised the existence of inhumanity and malpractices in this field. This could not, however, provide any basis for justifying or excusing the pain and discomfort which are associated with cosmetic surgical procedures whether they be of a minor or severe nature.

ANIMAL EXPERIMENTATION IN RELATION TO COSMETIC SURGERY

Animal experiments are carried out with the purpose of advancing biological knowledge or obtaining knowledge which will be useful for prolonging life or alleviating suffering in both man and animals, or to enable students to acquire biological knowledge which will be useful for saving or prolonging life or alleviating suffering.

The justification for using laboratory animals in the pursuit of scientific and medical and veterinary objectives is overwhelming when it is viewed against a background of the vast benefits to both animals and man which have arisen directly from past animal research and the reasonable expectation that such research will be of equal or greater benefit to all forms of life in the future[1].

A prime concern in animal experimentation is the protection of the animals from unnecessary discomfort and pain. Researchers have a moral and legal obligation:

(a) to take effective precautions to prevent or reduce to a minimum any pain or distress or fear in the animals used in experimental studies;
(b) to painlessly kill any animal which is suffering discomfort or pain if the condition is likely to endure as soon as the experiment is completed;
(c) not to subject any animal to any procedure which will cause severe pain which is likely to endure.

Whilst it cannot be claimed that laboratory animals never experience pain or discomfort or that the quality of animal care and treatment is optimal in all laboratories and institutions, the Committee felt that such shortcoming could not be taken as any justification or excuse for the performance of cosmetic surgery.

MANAGEMENTAL SURGICAL PROCEDURES IN COMPANION ANIMALS IN RELATION TO COSMETIC SURGERY

Surgical procedures which are performed as an aid to the management of companion animals include procedures such as sterilisation, devocalisation, declawing of cats, tattooing of ears and removal of dew claws as an aid to grooming were considered to be necessary for the proper management of dogs and cats under certain conditions of husbandry and accommodation.

These procedures were performed to prevent indiscriminate breeding, to reduce expenses, to control disease, to curb destructive and/or disturbing behaviour and to reduce the risk of injury. The various indications for practising the procedures were considered by the Committee as ample justification for these operations. In most instances the procedures had a direct beneficial effect on the welfare of the animal subjects on which they were performed.

The Committee could not find any justification for the performance of cosmetic surgery on the basis of managemental surgical procedures which are performed in companion animals.

CONCLUSIONS

After due consideration of all factors associated with the performance of cosmetic surgery, the Special Committee was of the opinion that cosmetic surgical procedures were inhumane, irrational, and with the exception of tail docking in dogs, were largely unsupported by public opinion. It was also felt that they could constitute an offence in terms of the Animal Protection Act, No. 71 of 1962.

The findings of the Committee also concurred with those of the SAVA Ethical Committee in 1975 which had led to the banning of ear cropping in dogs and tail nicking in horses by the Veterinary Board in 1978.

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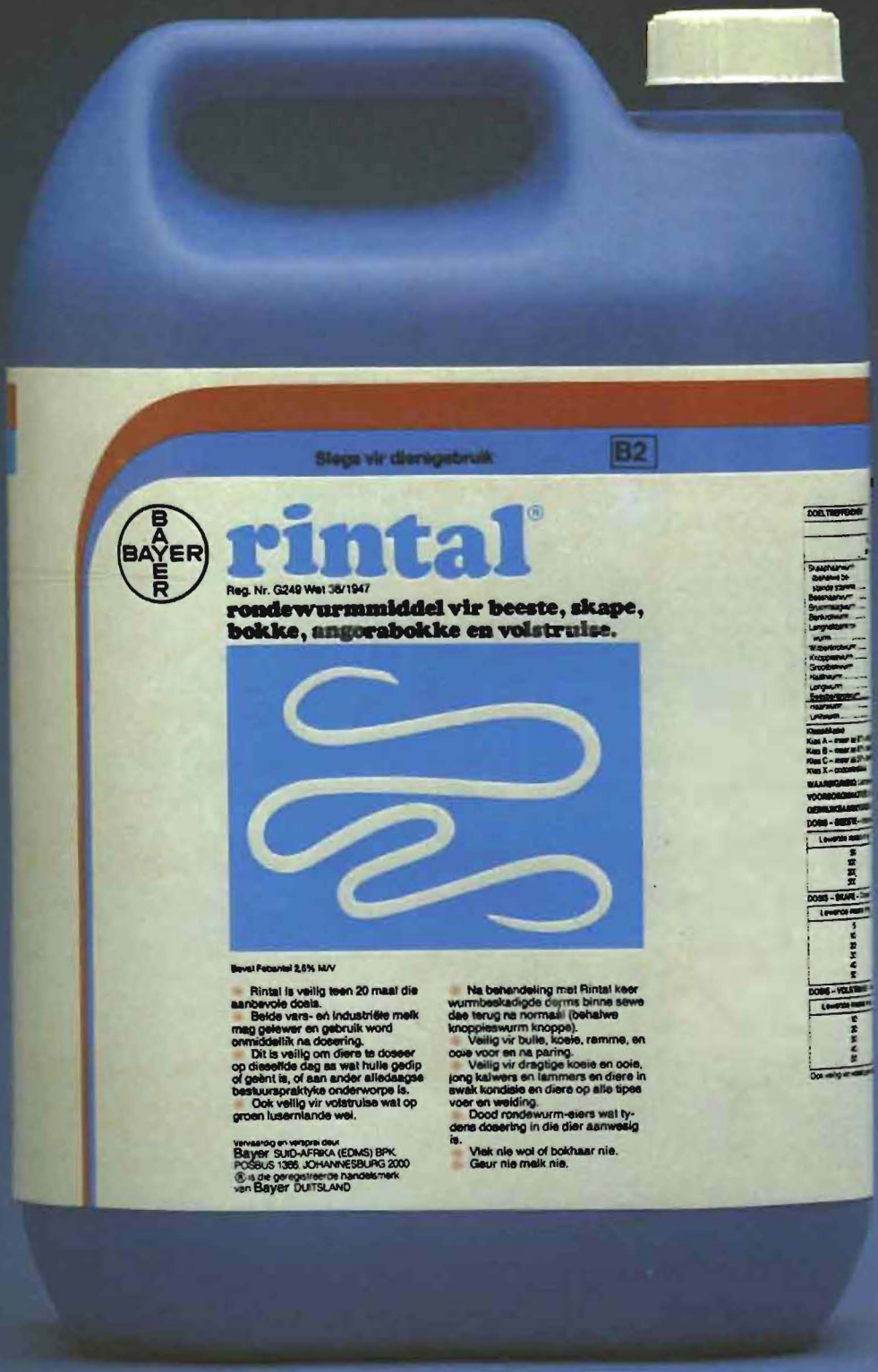

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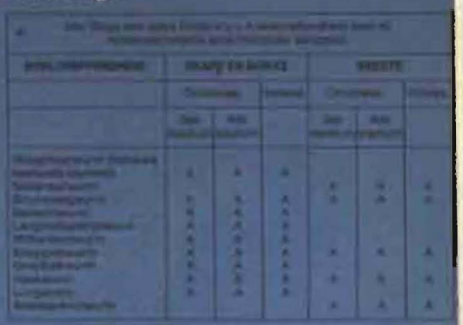

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