HOSTED BY THE
WHO COLLABORATING CENTRE FOR RESEARCH AND MANAGEMENT OF ZOONOTIC DISEASE CONTROL, VETERINARY RESEARCH LABORATORY, HARARE

PROCEEDINGS OF THE THIRD INTERNATIONAL CONFERENCE OF THE SOUTHERN AND EASTERN AFRICAN RABIES GROUP

HARARE, ZIMBABWE

7-9 MARCH 1995

EDITED BY

JOHN BINGHAM

GEORGE C. BISHOP

AND

ARTHUR A. KING

CONTENTS

EPIDEMIOLOGY AND DIAGNOSIS

FOREWORD

The Southern and Eastern **African Rabies Group (SEARG)** was founded at a gathering of rabies scientists, diagnosticians and policy makers in Lusaka in June 1992. The following year, in South Africa, two meetings were held under the auspices of SEARG, in Pietermaritzburg and Onderstepoort.

At the meeting in Pietermaritzburg, Mozambique was elected to host the next meeting of SEARG, to be held in late 1994. However, due to logistical difficulties, this venue was changed and Zimbabwe was requested to be host. This meeting took place in Harare on 7-9 March 1995. These proceedings contain the presentations given at that meeting.

This meeting would not have been possible without the financial assistance of the OIE Rabies Reference Centre at Onderstepoort; the World Health Organization, Geneva; the Department of Foreign Affairs of the South African Government; Pasteur Merieux, France; Rhone Merieux, France; the Office International des Epizooties, France; RDP Livestock Services, The Netherlands; Virbac Laboratories, France; Mallinckrodt Veterinary, United Kingdom; Central Veterinary Laboratory, Weybridge, United Kingdom; the Cattle Producers Association, Zimbabwe and Zimvet, Zimbabwe. We gratefully thank all these sponsors and we look forward to their support at future meetings.

The meeting was hosted by the WHO Collaborating Centre for Research and Management of Zoonotic Disease Control, based at the Veterinary Research Laboratory in Harare. The support of the Director of Veterinary Services, Dr Stuart Hargreaves and the Assistant Director of Veterinary Services/Diagnostics and Research, Dr Unesu Ushewokunze-Obatolu is gratefully acknowledged.

The meeting was well attended by the SEARG member countries. The Veterinary Department of every invited country was represented, as were representatives from Health ministries of nearly every country. All the member countries gave reports, which although making depressing reading due to the increasingly overwhelming task of controlling rabies, were very forthright. Four authorities on rabies from Europe and North America accepted our invitation to present papers. In addition, several rabies scientists working in the region attended and presented papers on their work. We would like to thank all the presenters for their valuable contributions.

The organisers would also like to thank those who chaired the sessions and the various rapporteurs who recorded the discussions.

During the business meeting, held on 8 March 1995, it was proposed that the next meeting be held in Nairobi, Kenya in early 1997. An incoming committee to organise this meeting was elected as follows: Dr Philip Kitala (Chairman), Dr John Bingham (Vice-Chairman), Dr Brian Perry (Secretary) and Dr George Bishop (Treasurer).

John Bingham, George Bishop and Arthur King (editors)

PROGRAMME

7 MARCH 1995 FIRST DAY

0730 - 0810 Registration

0810 **Chair**: Director of Veterinary Service **Dr S.K. Hargreaves**

Welcoming address: Honourable Minister of Health and Child Welfare

Theme: National Reports Chair: Dr G. Bishop

0830 - 1000 National Reports:

Uganda	Dr J. Illango
Kenya	Dr Karugah
Tanzania	Dr Sembiko
Malawi	Dr C. Mwiyeriwa
Zambia	Dr H. Munang'andu

1000 - 1030 TEA BREAK

1030 - 1230 National Reports (continued)

Mozambique	Dr Rodrigues
Madagascar	Dr R. Tsiresy
Namibia	Dr O.J.B. Hübschle
Botswana	Dr K. Sehularo
Zimbabwe	Dr J. Bingham
Swaziland	Dr R. Dlamini
Lesotho	Dr L. Khomari
South Africa	Dr G. Brückner

1230 - 1330 LUNCH

Theme: Human rabies **Chair: Dr F.-X Meslin**

1330 - 1400 Review and Introduction	Dr R. Swanepoel
1400 - 1430 Rabies video	Dr P. Kloeck
1430 - 1500 Human rabies in Zimbabwe	Dr A. van Geldermalsen

1500 - 1530 TEA BREAK

1530 - 1600 Ante-mortem diagnosis in humans	Dr M. Fekadu
1600 - 1630 Pre- and post-exposure treatment for rabies: the latest developments	Dr J. Lang
1630 - Discussion on human rabies	

Theme: Role of International Organisations

Chair: Dr A.A. King

1630 - 1640	Role of WHO	Dr F.-X. Meslin
1640 - 1650	Role of OIE	Dr G.R. Thomson
1650 - 1700	Role of a Regional Collaborative Centre	Dr U. Ushewokunze-Obatolu

8 MARCH 1995 SECOND DAY

Theme: Canine rabies. **Chair: Dr A. King**

0800 - 0830	The epidemiology and significance of canine rabies worldwide	Dr A. I. Wandeler
0830 - 0900	Dog ecology studies in Zimbabwe	Dr J. Butler
0900 - 0930	Dog ecology studies in Kenya	Dr J. McDermott
0930 - 1000	Rabies in dogs in South Africa	Dr G. Bishop

1000 - 1030 TEA BREAK

Chair: Dr M. Obwolo

1030 - 1050	Rabies in dogs in Tanzania	Dr S. Cleaveland
1050 - 1110	Vaccination coverage required to prevent outbreaks of dog rabies	Dr P. Coleman
1110 - 1130	Multi-sectoral collaboration in rabies control in Uganda	Dr C.S. Rutebarika
1130 - 1200	A two day massive rabies campaign: a sustainable intervention?	Dr K. de Balogh
1200 - 1230	Rabies control in dogs in Zimbabwe	Dr W. Madzima

1230 - 1330 LUNCH

Chair: Dr A.I. Wandeler

1330 - 1400	Progress in oral vaccination of wildlife in Europe	Dr A.A. King
1400 - 1430	Oral vaccination as a means of rabies control in dogs	Dr F.-X. Meslin
1430 - 1500	Oral delivery of rabies vaccines to dogs	Dr C. Schumacher

1500 - 1530 TEA BREAK

Chair: Dr M. Fekadu

1530 - 1600 General discussion on rabies in dogs.

1600 - Business meeting. Chair: Dr G. Bishop

The role of SEARG in promoting rabies control, election of officers, discussion of diagnostic manual etc.

1900 Dinner for conference participants and invited guests (sponsored by Rhone Merieux).

9 MARCH 1995 THIRD DAY

Theme: Epidemiology and Diagnosis. Chair: Dr R. Swanepoel

0800 - 0830 Surveillance and diagnosis: some thoughts	Dr A. I. Wandeler
0830 - 0900 Epidemiology of antigenic and genetic variants of African rabies viruses	Dr G.R. Thomson
0900 - 0930 Diagnostic techniques	Dr A.A. King
0930 - 1000 Antibody measurement	Dr J. Barrat

1000 - 1030 TEA BREAK

Theme: Rabies Surveillance. Chair: Dr B. D. Perry

1030 - 1100 Data gathering and analysis for improved decision support to rabies control	Dr B. D. Perry
1100 - 1120 The European approach	Dr J. Barrat
1120 - 1140 The South African approach	Dr G. Bishop
1140 - 1200 The Zimbabwean approach	Dr J. Bingham
1200 - 1230 Discussion	

1230- **Closing address:**
Director of Veterinary Services **Dr S.K. Hargreaves**

1230 - 1330 LUNCH

1400 - 1630 Practidal sessions for country representatives.

(Venue: Veterinary Research Laboratory, Borrowdale Road, Harare.)

Practical session A: Data collection and reporting. Leader George Bishop

Practical session B: Diagnostic methods. Leader: Jacques Barrat.

OPENING SPEECH

The opening speech was given by the Honourable Minister of Health and Child Welfare, **Dr Timothy Stamps:**

"It gives me pleasure to welcome to Zimbabwe the international rabies experts who have given their time to address this meeting and also to welcome the representatives of fourteen eastern and southern African countries who will share with us their experiences of working with this disease.

Rabies is a disease of animals and infection of humans is, almost always, a "spill-over" event from an infected dog. Over ninety per cent of human deaths from the disease occur in the tropics, where some three-quarters of the world's population lives. Throughout the near global distribution of the disease various mammalian species other than the dog may act as principal hosts and vectors. For example, within recent decades the disease has become widely reported in jackals, bat-eared foxes and mongooses in many southern African countries.

The early history of rabies in Zimbabwe is somewhat anecdotal, but indigenous inhabitants of Matabeleland and Manicaland recalled that the disease had been prevalent prior to European settlement in the country. The first irrefutable record was of an outbreak in dogs which began in 1902 after introduction from the north-west where rabies was rife in western Zambia. The outbreak was brought under control by measures which included the large-scale destruction of "unowned" dogs and the muzzling and tying-up of other dogs. Sporadic cases continued to occur until 1913, but apart from an episode of dog rabies at Victoria Falls in 1938, the country remained free until 1950.

The present outbreak of rabies in Zimbabwe began in 1950 when infected dogs were brought into the country by people moving freely across the border with eastern Botswana. A concomitant episode occurred some 250km further east near Beitbridge, the source being a rabies outbreak in the Messina area of Transvaal, adjacent to the border with Zimbabwe. During the next two years these outbreaks spread with considerable, momentum, but regressed following the application of efficient control measures. In 1952, however, the first serious outbreaks in wildlife, especially jackals, were confirmed in Chipinge and Marondera, although unconfirmed cases had previously been reported.

Since 1950, over 23 000 specimens have been submitted for rabies examination to the Veterinary Research Laboratory in Harare and more than 9 000 have been confirmed positive. Regrettably, 164 human rabies cases have also been confirmed, although this figure does not take into account an additional number of people who have died from unconfirmed clinical rabies.

Today, although dogs are still the most prolific vectors of rabies in Zimbabwe, jackals are a serious threat, not only to people and dogs, but also to livestock. Our jackal population appears to be becoming more highly urbanized with contact between jackals and dogs more frequently reported. It is encumbent upon us all to seek every means available to control this growing threat to our people, pets and livestock.

I congratulate the now well established Southern and Eastern Mrican Rabies Group on the programme which they have devised for this meeting. As Minister for Health and Child Welfare I am particularly pleased to see that, for the first time, a complete session has been given over to rabies in humans. A closer association between veterinary and medical scientists is essential if we are to better understand the disease and to reduce its impact on our lives. I note that the programme also contains papers which are at the very forefront of rabies research - dealing with the oral vaccination of dogs and wildlife. Oral vaccination is a technique which holds out hope for real progress in rabies control in Zimbabwe.

I also congratulate the Veterinary Research Laboratory, who are hosting this meeting, on their recent appointment as a World Health Organization Collaborating Centre for Research and Management of Zoonotic Disease Control. The appointment comes as recognition of the excellent work of the Laboratory and will provide a focus for continued research, training and co-operation within the region.

It is my hope that you have a successful and rewarding meeting and that you return home safely, full of enthusiasm for the continued fight against rabies".

CLOSING ADDRESS

The closing address was given by the Zimbabwe Director of Veterinary Services, Dr **Stuart Hargreaves:**

"1n general, Mr Chairman, we can say that the meeting has been a success - maybe even a great success. There were 13 different countries represented, all those from the Southern and Eastern African Rabies Group were present, with others from as far afield as Canada, the USA, France and the UK. There were, I am informed, more medical doctors than at previous meetings. This is to be encouraged as it was emphasised that co-operation and liaison with our medical colleagues needs to be greatly improved. It was therefore encouraging and fitting that the meeting was opened by our own Minister of Health and Child Welfare.

The meeting began with country reports, and here I must compliment speakers on their frankness and honesty. Speakers were not shy to point out their country's deficiencies and weaknesses. It was good that we could all talk freely, and there was good interaction with everyone.

However, it was tragic that many member countries reported a gross shortage of vaccine due to lack of funds. It is in my opinion unacceptable that priority is given to the importation of many luxury goods over the need for rabies vaccine. This is totally unacceptable. Public pressure is needed to change this attitude.

In just about all countries there was a decline in the number of dogs vaccinated and in all cases the numbers vaccinated were well below the accepted 70 percent cover required for adequate control. Much greater effort is needed in this regard, we need to get down to basics, all of us need to increase the rabies vaccination coverage. Generally, rabies is on the increase within the region, which is of great concern.

Many countries mentioned the lack of basic data and under-reporting was common. There is a general need to do more basic groundwork to get an accurate assessment of all the dogs in the country, and other basic information. This is beginning to be done in some countries as demonstrated by the studies of the ecology of dogs and wildlife vectors of the disease.

I must congratulate Lesotho for achieving and maintaining freedom from rabies, although, as with other countries, vaccinations are declining.

The general outcome of the country reports emphasised the need for education of the people and public awareness, the need to get to the schools and educate children about rabies and the need for co-operation from the people in control. Many countries emphasised that rabies had entered their country from a neighbour and it was pleasing to hear that in fact the disease entered southeastern Africa from the north! While talking to the Minister of Health last night I was informed that rabies infection in Africa was introduced from Europe!

Generally rabies in Africa was a dog/human problem and socio-political influences affected the prevalence of the disease.

Human rabies: This was a very interesting session and much debate centred on vaccination strategy and affordability of vaccine. Dr Wandeler said that each positive rabies case indicated a failure, a failure not only in the medical profession but in the system. The same applies to dog rabies, every case indicates a failure in the system. There is a need to improve control programmes, there is so much to be done. There is a very definite need to investigate vaccination strategies that will reduce costs yet remain effective, for example, by reducing the number of visits to hospital. Drug companies could assist in this regard as well as more research, for example, intradennal vaccinations and other strategies to give cheap and life-long immunity.

The rabies video, I think, said it all, and it re-emphasised the need to control this most terrible disease. We will need to have copies and to show them to politicians to increase funds for rabies control, research, vaccines, surveillance, treatments, education and public awareness. As a group, we must influence people with control over funding, such as governments, international agencies and donors. There is a real need for greater public awareness and education. It is of concern that some people thought that rabies was caused by changes in the weather, or they sought the services of a "dog bite specialist" after being bitten rather than a medical doctor. This shows how much work there is to be done.

There were several papers on dog ecology and it was good to see that much progress had been made in this area following recommendations from previous meetings. This is a step in the right direction. The ecology studies have given us a far better understanding of the disease and there is a need to work out more optimal immunisation programmes, both in dogs and humans, and it must be affordable. With the knowledge that there is a high turnover of dogs in the early years and that in one study 88 percent of the dog population under one year of age were not vaccinated, there is a need for vaccine that can be used effectively in young puppies and that will have long, if not life-long immunity. Maybe it is too much to ask.

It was emphasised that under-reporting was rife and that although rabies was increasing, control programmes were covering a smaller proportion of the dog population. A paper from Zambia did demonstrate, however, that with effort, goodwill and co-operation a lot could be achieved. However, the sustainability of such campaigns was questioned.

It was clearly evident that oral vaccines do work and are very successfully used in Europe. But there was still much work to be done. Before use it was advised that oral vaccines should be tested in each particular country, region and area. Great detail and background information was required before use regarding target and non-target species. Oral vaccination means a lot of hard work from ourselves and manufacturers and it is recommended that oral vaccination be further evaluated according to WHO protocols, especially in countries where parenteral vaccination has failed due to uncontrolled dogs and wildlife vectors.

During question time, public education again was raised and George Bishop stated that rabies had no rules. The complexities of the disease were clearly pointed out and, as the Malawi delegate said, we must not give up.

There is a great need for this group to spread information vertically and, can I say laterally, to give public and political awareness within countries and to source donor/governmental support for more funds.

In today's session it was unfortunate I missed the first presentation, but there was, I am informed, a large amount of very useful and practical information.

The scientists have done a great deal of work which indicates there are two genetic groups of rabies viruses in Africa, the mongoose and canid types. In addition, there are several types of rabies-related viruses. Our understanding of the epidemiology of the disease is improving, which is encouraging.

The diagnosis of rabies was reviewed and the collection of brain samples by the straw technique may be a way to reduce transport costs. It is clear that there is great variation in diagnostic procedures between various laboratories and there is need for standardisation. Training workshops were recommended.

The importance of both active and passive surveillance in making rational strategies was stressed, concerning for example type of vaccination system, cost and recovery. There is a need for basic data and various economic evaluations. Educationalists, economists, sociologists all need to be involved. The group should investigate the standardisation of reporting and handling of information and use of computerisation.

We had an interesting presentation of the European approach to rabies. We were able to compare what occurs in our part of the world with activities in Europe, perhaps we can learn from their strategies. The South African approach indicated a simple system of information gathering. We need to get back to basics and build from there, with better analysis of basic data. The group could look at standardisation of forms and their design. The Zimbabwean system has been tried and tested over the years, but there is room for collaboration which will lead to improvements.

In closing I must say thanks, firstly to the sponsors who provided the funding. Without funding there would be no meeting. Thanks also to those involved in the organisation of the meeting, especially George Bishop and John Bingham; the invited guests, Drs Alex Wandeler, Arthur King, Makonnen Fekadu and Jacques Barrat; our laboratory which hosted the meeting, and will be the venue for the practical sessions this afternoon. Lastly thanks to all the speakers who presented papers, Rhone Merieux for the dinner, and you the participants, for without your interest there would be no meeting.

I declare this third meeting of the Southern and Eastern African Rabies Group closed and wish you a safe journey back to your homes".

ROLE OF THE "OFFICE INTERNATIONAL DES EPIZOOTIES" (OIE) IN RABIES DIAGNOSIS AND CONTROL IN SOUTHERN AND EASTERN AFRICA

G.R. Thomson, Director of the OIE Regional Collaborating Centre for Africa, Onderstepoort, South Africa

In 1994 the Onderstepoort Veterinary Institute (OVI) of South Africa was designated as an OIE Regional Collaborating Centre for Africa. This appointment is for three years in the first instance but is renewable thereafter. No direct funding is received from OIE to cover the expenses of the Centre, although a modest amount is provided by the South African Department of Agriculture for this purpose.

Objectives of the Centre

To improve surveillance of animal diseases in southern and eastern Africa and to contribute to the implementation of disease control programmes.

Activities

1. Training: Management of information systems; registration and testing of drugs; other functions as required.

2. Diagnosis: Expanding and improving diagnosis especially in respect to diseases for which the OVI already acts as an 0IE reference laboratory (*viz.* rabies, blue tongue, African Horse sickness, African swine fever, Rift Valley fever, lumpy skin disease). supply of biological reagents required for diagnosis.

3. Support for research.

So far as rabies specifically is concerned, the OVI has recently revamped the Rabies Unit to comply with international standards for working with this agent. In particular, the aim of the Rabies Unit is to provide an efficient routine as well as a standardised diagnostic service for animal rabies in South Africa in particular, as well as other southern and eastern African countries.

Activities and techniques which are currently in operation are:

- Direct FA testing of brain specimens from animals suspected as rabid;
- Histological and immuno-histochemical examination of the brains of animals suffering from neurological disorders.
- Serological testing for rabies and rabies virus proteins:
    - Blocking ELISA (antibodies to rabies N protein);
    - Rapid fluorescent focus inhibition test (virus neutralisation);

- Screening of rabies viruses with a truncated panel of MAbs to differentiate "canid" and "viverrid" viruses as well as rabies-related viruses;
- Genome sequencing to enable the accurate determination of relationships between rabies viruses;
- Commercial production of a direct fluorescence conjugate for rabies;
- Preservation and cataloguing of rabies virus isolates from different species and localities in southern Africa,
- Maintenance of a computerised rabies database for South Africa in association with the Directorate of Animal Health;
- Participation in a number of research projects.

WHO COLLABORATING CENTRE FOR RESEARCH AND MANAGEMENT OF ZOONOTIC DISEASE CONTROL

U. Ushewokunze-Obatolu, Assistant Director of Veterinary Services (Diagnostics and Research), Harare, Zimbabwe

The Centre's terms of reference

1 Programme management: To collaborate with the WHO in regional and national projects on zoonoses control and related food-borne disease. The WHO is to provide the Centre with comprehensive programme management assistance, including formulation, planning, implementation and evaluation. Also collaboration with international centres and other WHO collaborating centres in zoonoses, food-borne disease and veterinary public health.

2 Research: To contribute to projects of operational research in national programmes of surveillance and control and to co-operate in basic (strategic) and applied research.

3 Capacity building: To collaborate in strengthening programmes for education and training of professional and technical personnel for the African region.

4 Co-ordination: To strengthen co-operation between national veterinary and public health services towards improving surveillance, prevention and control of major zoonotic diseases.

5 Regional collaboration: With regional member States at their request.

6 Operational support: To provide biological reference material and laboratory services on request and assist member States in developing surveillance systems and related laboratory services. (Rabies is singled out as the most important for most of these purposes.)

Centre's activities so far

Once communication was effected in mid- 1994, the Centre embarked on the following:

1 . Studying priorities in the area of zoonoses, food borne diseases and veterinary public health through
- internal consultations;
- consultations with Ministry of Health;
- inter-institutional workshop;
- identification of areas for immediate attention.

2. Findings:
- facts required in diagnostic methods by various institutions;

- information management needed improvements to reflect sectoral interests and to make available information on zoonoses;
- except in the case of rabies and anthrax, inter-sectoral co-ordination was poor.

3. Priority diseases: Rabies, cysticercosis, TB, plague, *Campylobacter*, *Listeria, Salmonella, Brucella,* anthrax, trypanosomiasis, *Chlamydia,* Rift Valley fever and Wesselbron disease.

4. Activities:
   - Raise awareness among public;
   - Seek support for research programmes on zoonoses;
   - Identify areas for improvement in diagnostics e.g. trypanosomiasis, *Salmonella, Brucella melitensis;*
   - Strengthen information management and exchange;
   - Seek training opportunities for staff.

5. Outputs so far:
   - Confirmation of interests among individuals and institutions;
   - Identification of problems and opportunities to be exploited;
   - Renewal of interest in zoonoses;
   - National networking stimulated towards co-ordinated action plans.
   - Supporting regional activities (e.g. SEARG)

Centre's action plan

Short term:
- Information management - creating and managing a central database and network;
- Identification of infrastructural needs for selected priorities;
- Strengthening sectoral networking;
- Operational support.

Intermediate and long term:
- Collaborative research into *Salmonella enteritidis,*.
- On the job training for professional and technical staff,
- Identification and prioritisation of new areas for research and control;
- Strengthening of programme management;
- Regional co-ordination.

Services/opportunities presently offered

Training in immunodiagnostics generally including rabies FAT, ELISA for Rift Valley fever, *B. abortus* testing surveillance and informatics

2. Provision of test control reagents

- positive and negative brain material for rabies;
- positive and negative serum samples for rabies;
- Quality control on rabies conjugate
- Information bank management
- Collaboration in research

  Anthrax molecular epidemiology;

  TB molecular test,

  S. enterititis as public health problem.

Concerns

- Resource base to cope with widening mandate;
- Position of member states in region on this;
- Sometimes a clash of priorities between sectors;
- Administrative constraints.

RABIES IN UGANDA

James Illango

Animal Health Research Centre, Entebbe, Uganda

Introduction

From the time rabies was recognised as a major public health problem in Uganda, which culminated in the enaction of the Rabies Act of 1935, it has remained a disease of considerable significance. In order to counteract the disease, the Department of Veterinary Services designed a strategy based on notification, laboratory diagnosis and application of appropriate, internationally accepted preventative measures. In 1991, it further developed a multi-sectoral policy to broaden its approach to the problem (see "Multi-sectoral collaboration in rabies surveillance and control in Uganda" page 120). Despite these measures the incidence of rabies has increased, as shown in Table 1.

Table 1. Total reported rabies cases in Uganda from 1992 to 1994.

Species	1992	1993	1994	Totals
Human	15	23	15	53
Dog	243	228	376	847
Cat			1	1
Cattle	1	2		3
Goat	2		7	9
Fox		2		2
Jackal	3	6		9
Totals	264	261	399	924

Rabies in 1992

In the year 1992 there was a total of 264 cases in the country, with high numbers of cases recorded in the months of February, July and October. The incidence was low in November and December. The dog was the major animal species accounting for 243 cases (92 percent). The jackal was the only wildlife species recorded with 3 cases. There were 15 human victims.

As shown in Figure 1, for 1992, there was a wide distribution of the disease in all regions of the country. In total 23 districts out of 40 were affected. Most cases were recorded in the east in Kapchorwa District with 67 cases, Rukungiri District with 44 cases, Masaka with 24 cases and Iganga with 13.

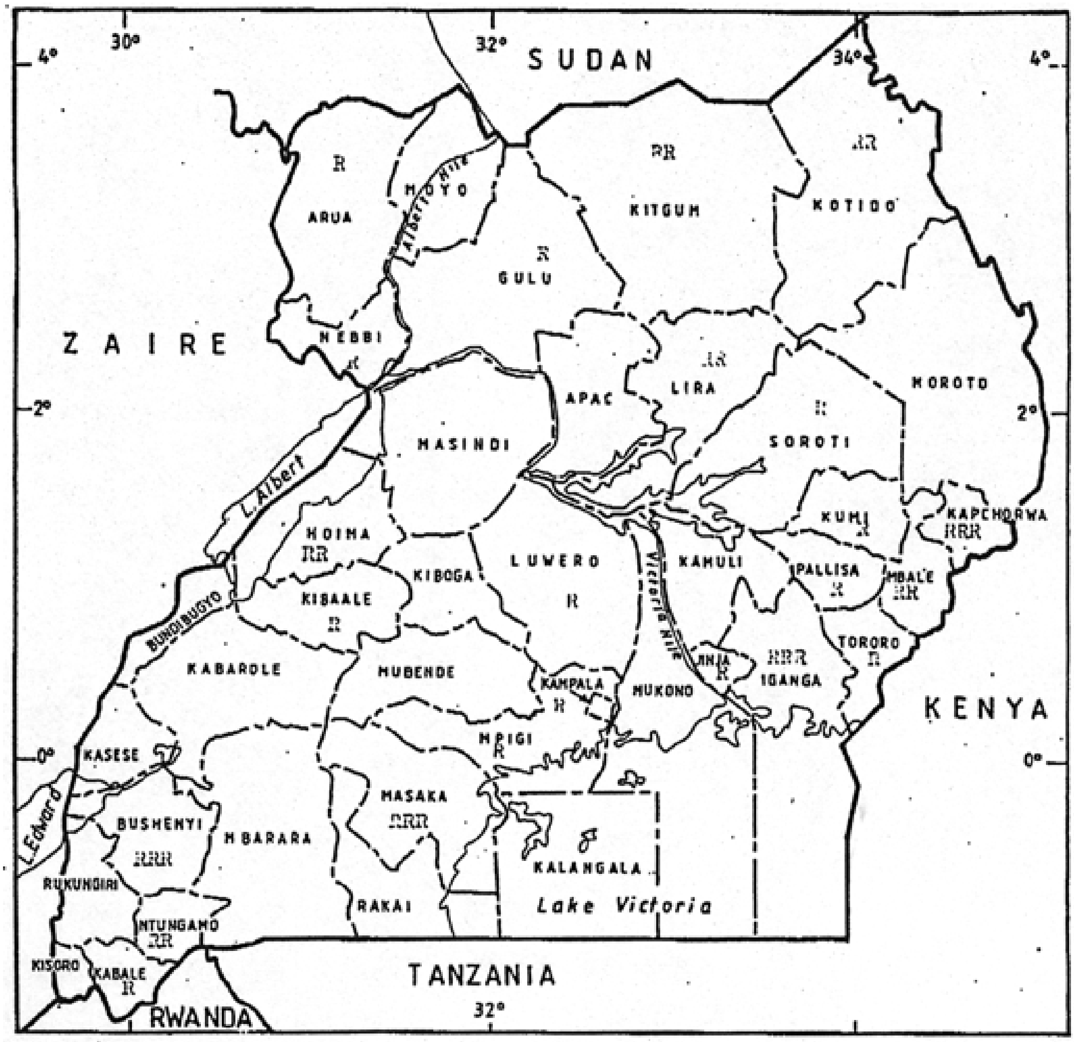


Figure 1. Rabies in Uganda, 1992. Key: R - less than 5 cases; RR;- 5-10 cases; RRR - over 10 cases.

There were specific factors as well as general factors which accounted for the rabies pattern in the country. In the northern part of the country, there was a combination of an influx of refugees and their pets from the fighting in southern Sudan where no preventive measures are taken and incidents of rebel activities which disrupted the provision of necessary services, such as vaccination. In the eastern part, the area had just recovered from massive raiding and rustling by Karamojong warriors and some incidents of rebel activities which also affected Government services, caused massive displacement of people, and social life in general. In one district, Kapchorwa, wild carnivores (jackals and foxes) were incriminated for such occurrence of the disease. A game reserve has now been demarcated in part of the area. The area is mountainous, with a lot of transport problems.

Among the general factors affecting the whole country were: stray dogs which were inaccessible for vaccination and were on the increase, poor vaccination coverage and public awareness was still low.

Rabies in 1993

During 1993 a total of 261 cases was recorded (Table 1 and Figure 2). High numbers of cases were reported in February, July, August September, October and December. The dog was the major animal species, accounting for 228 cases (87.3 percent). Foxes and jackals accounted for 2 and 6 cases respectively, all from Kapchorwa District. There were 23 human cases.

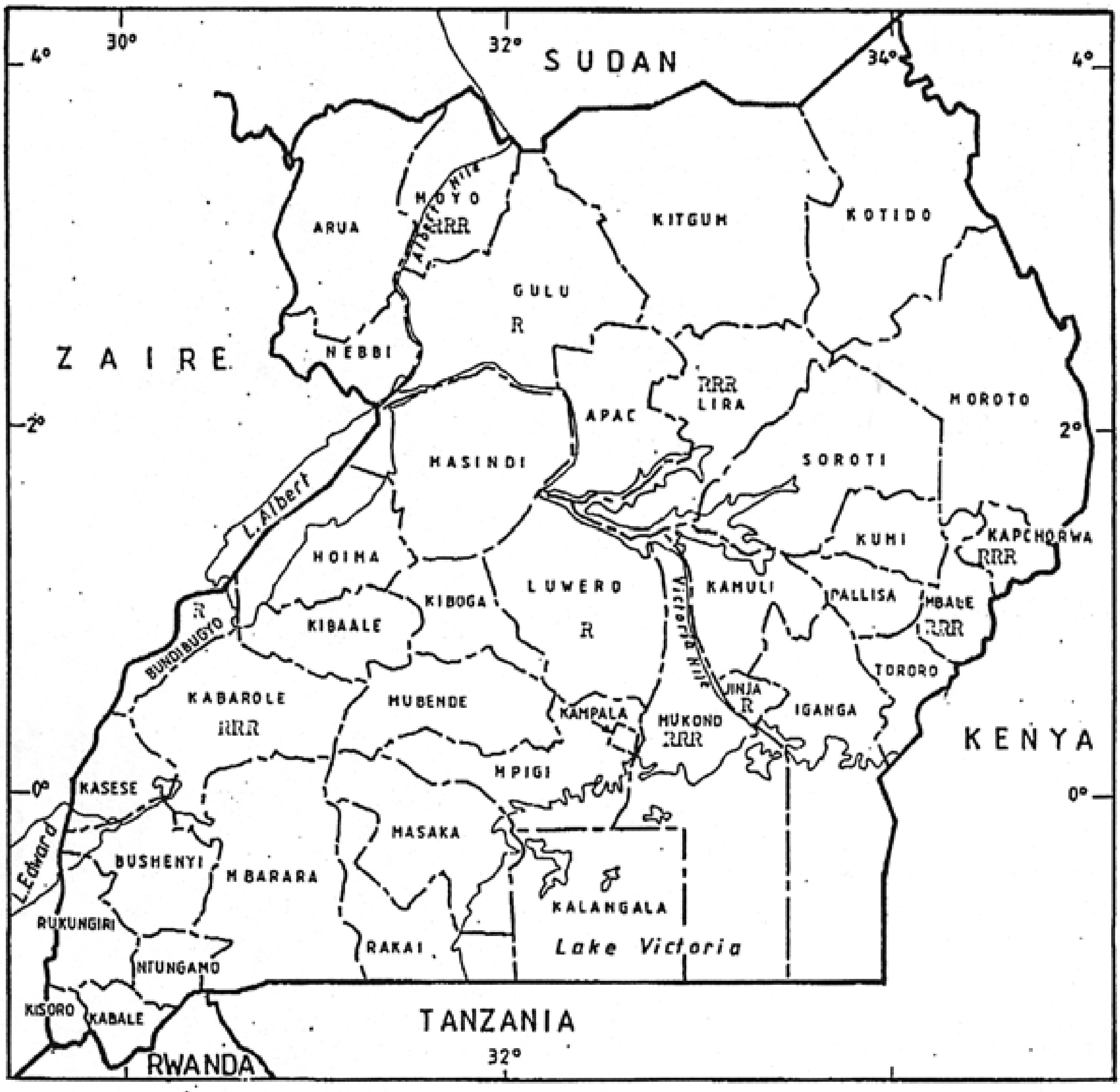


Figure 2. Rabies in Uganda, 1993. Key: R - less than 5 cases; RR - 5-10 cases; RRR -over 10 cases.

During 1993 the disease was patchy and affected only 10 districts out of 40. Most cases were recorded in the following areas: Lira (northern region) 20 cases, Kapchorwa (eastern) 73, Mukono (Central) 86 cases and Kabarole (western) with 16 cases. Other regions had sporadic cases.

Factors influencing the rabies pattern included massive vaccination campaigns done in the previous year in 23 districts. A total of 56 657 dogs and 95 cats were vaccinated. In districts where the incidence was high, either vaccination coverage was very low or no vaccination at all was done. As in 1992 stray dogs, refugees and rebel activity contributed to poor control.

Rabies in 1994

There was again an increase in the incidence of the disease with 399 cases recorded. High numbers of cases were recorded in January, February, March, June, July, August and September. The dog was the major animal species accounting for 376 cases (95 percent). Fifteen human cases were recorded.

There was wider distribution of the disease than in the previous year with 22 districts affected. The major areas were: Masaka (119 cases), Jinja (44 cases) and Kitgum (24 cases).

The major factors affecting the rabies pattern this year were the continued effect of refugees and rebel activities, affecting north-westem Uganda and parts of northern Uganda, which disrupted services. The vaccination coverage in some areas (for example Masaka) was poor. In the north-eastern areas of Kotido and Moroto there was considerable lack of public awareness which made it difficult for any meaningful preventive measures to be put in place. The areas are inhabited by nomadic pastoralist whose livelihood depends on cattle raiding. Finally, stray dogs played a significant role.

Table 2. Total rabies cases from 1992 to 1994 by month.

Year	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Total
1992	26	42	25	20	20	22	35	19	15	30	5	5	264
1993	2	25	13	10	18	23	30	40	28	24	6	42	261
1994	19	37	40	3	17	91	67	59	37	15	13	1	399
Total	47	104	78	33	55	136	132	118	80	69	24	48	924

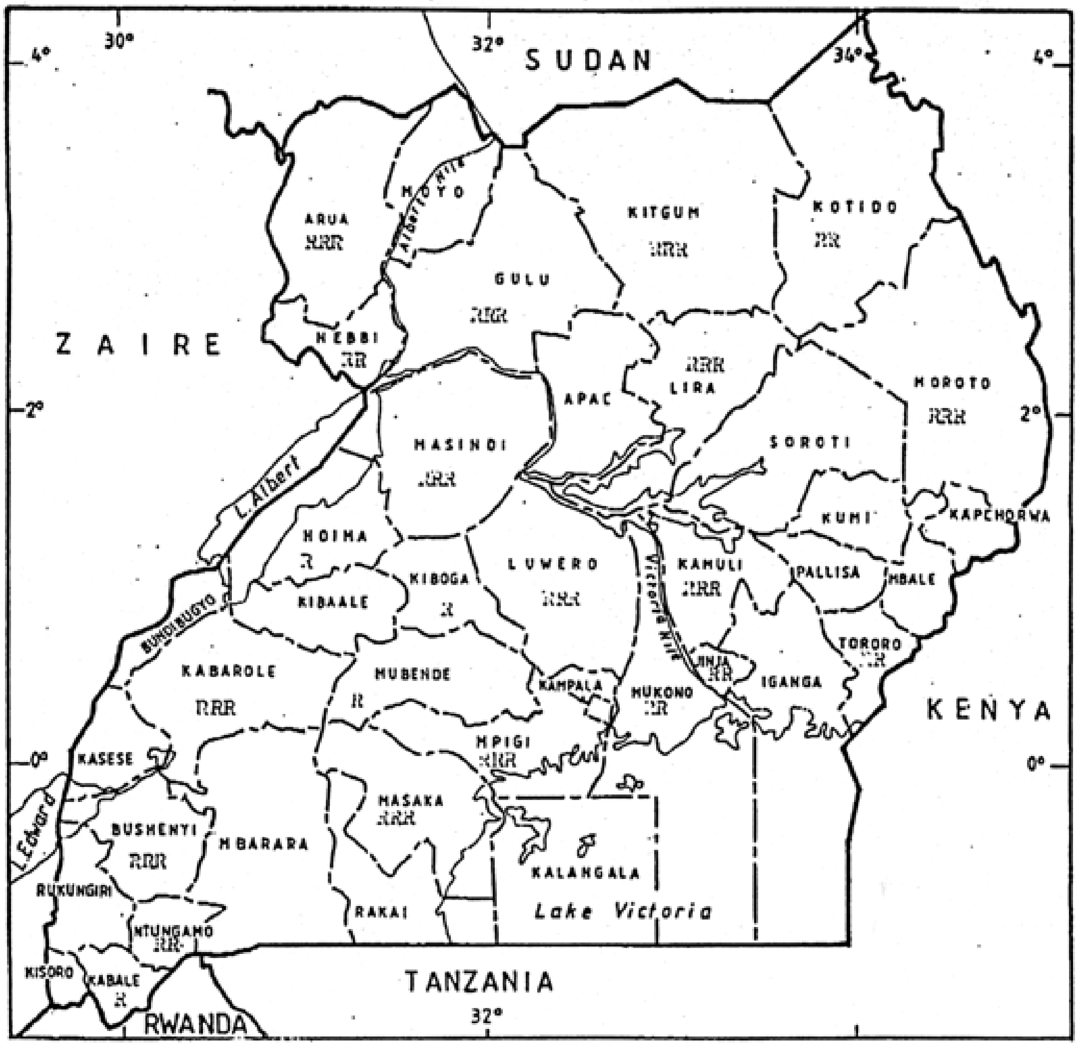


Figure 3. Rabies in Uganda, 1994. Key: R - less than 5 cases; RR - 5 - 10 cases; RRR -over 10 cases

Diagnosis

Rabies diagnosis is done at the Central Laboratory. Entebbe. Methods currently in use are the fluorescence antibody test (FAT), with the mouse inoculation test (NUT) as a back-up. In addition, laboratory diagnosis is also done at the Faculty of Veterinary Medicine, Makerere University, by histology. Major problems affecting laboratory diagnosis are:

1) Inadequate equipment. In the Central laboratory, histopathology cannot be done as there is no functional microtome nor tissue processor (old and broken down). Hence, samples for histology are sent to the University.

2) Operational funds are inadequate for the purchase of the necessary laboratory consumables. The mouse colony is currently supported by a German GTZ project.

3) Lack of transport for carrying out the necessary investigations, collection of samples and distribution of preservatives to the districts.

There are also field problems which indirectly affect laboratory diagnosis.

1) Lack of cold chain in some districts.

2) Inadequate sampling equipment. Only twenty districts received kits for rabies work.

3) Lack of protective clothing

4) Preservatives are not adequate to cover all districts, although they are available in the laboratory.

5) Transport constraints.

Control

Vaccination of dogs and cats is a major control method and it is compulsory. The main problem here is that vaccine is imported and so the supply is not regular, and sometimes not sufficient.

Table 3 shows the vaccination data from 1992 to 1994. Unfortunately, most districts do not carry out dog and cat censuses. Vaccinations are done in localities of out-breaks only, therefore an adequate percentage coverage is not achieved. In addition stray dogs are on the increase. Few cats are brought for vaccination.

Table 3. Numbers of animal vaccinations administered from 1992 to 1994.

Year	Dog	Cat	Total
1992	56567	95	56752
1993	24824	51	24875
1994	81965	341	82306
Total	163446	487	163933

Other control measures are:

1) Import restriction which require an import licence and a health certificate for any imported pet;

2) Tie-up orders and destruction of stray dogs. These are not very successful;

3) Public education to create awareness. This is addressed through a multisectoral approach.

The critical issues which need to be addressed in rabies control are:

1) Control of rabies in stray dogs which are on the increase, for example by the use of oral vaccination;

2) Public education;

3) Support to diagnostic services.

Acknowledgements

Dr W. Kaboyo, Veterinary Public Health Unit, Ministry of Health and Dr C. Rutebarika, Department of Veterinary Services, Ministry of Agriculture, Animal Industry and Fisheries, Entebbe are thanked for their contributions to this report.

RABIES IN KENYA

A.K. Karugah

Department of Veterinary Services, Kabete, Nairobi, Kenya

Since 1912, when the first case of rabies was diagnosed in a dog in Kenya, the disease has existed in the country with varying incidence. It was not until 1982, however, that the number of cases diagnosed annually rose to 200 and above. The disease, which is a problem both in the rural and urban areas, is now more widespread and prevalent than at any time in the country's history.

The rabies situation

Although all mammalian species can be affected by rabies, the species of most importance in Kenya is the dog. The dog is also the major vector of rabies in man. Approximately 55 - 65 percent of animal rabies cases diagnosed in our investigation laboratories occur in dogs. The other animal species probably play only a limited role in the maintenance of the disease in most parts of Kenya. This statistic is significant because of the close links between dog and man.

The number of positive animal rabies cases reported annually from 1991 to 1994 are shown in Table 1. Although the number of positive cases decreased from 205 in 1992 to 72 in 1994, there is no evidence that the incidence of the disease has decreased in the country. In fact, the official figures given represent only the tip of the iceberg. The reasons for this apparent decrease have to do with submission of specimens for diagnosis.

The figures for human rabies shown in Table 1 represent a significant under-estimate of the true statistics because a number of cases die in their homes and these are not reported.

Since 1987, only slightly more than 9 percent of the national requirement for post-exposure treatment was allocated, due to limited availability of funding. Control of rabies in dogs is one effective way of reducing cost of post-exposure treatment in man.

Surveillance

In Kenya, all cases of suspected rabies are dealt with by a qualified veterinarian wherever practicable and all outbreaks are reported immediately to the Provincial Director of Veterinary Services and the Director of Veterinary Services. Specimens are submitted to Kabete or Mariakani Veterinary Investigation Laboratories (VIL), these two being the only laboratories that carry out rabies diagnosis both in animals and humans in the country.

The VILs and District Veterinary Offices (DVO) are the regional collection points of all rabies specimens since they have facilities to transport the specimens to the diagnostic laboratories. However, distances between these collection points are often quite long, thus discouraging farmers or the public from submitting specimens or reporting cases. This is the major drawback in our rabies surveillance and explains, in part, the reason for the low number of samples submitted for diagnoses.

The Department of Veterinary Services (DVS) has a small computer facility that inputs rabies diagnosis data, amongst other data. These data are analysed and reports compiled, at no fixed interval, and disseminated to all offices handling technical matters in the Department.

The surveillance system described above underestimates the true incidence of rabies in the country. This is due to non-reporting and non-presentation of suspected animal cases at veterinary clinical centres and non-availability of local diagnostic facilities. The general public is not adequately educated to create the necessary awareness regarding rabies. Figure 1 is a map of Kenya showing the location of various VILs and their relative distances from Kabete and Mariakani VILs.

The wish of the Department is to have all eight VILs carry out rabies diagnosis so as to improve our surveillance system. At the moment, this is not possible due to lack of funds to purchase the required equipment and to lack of adequately trained virologists.

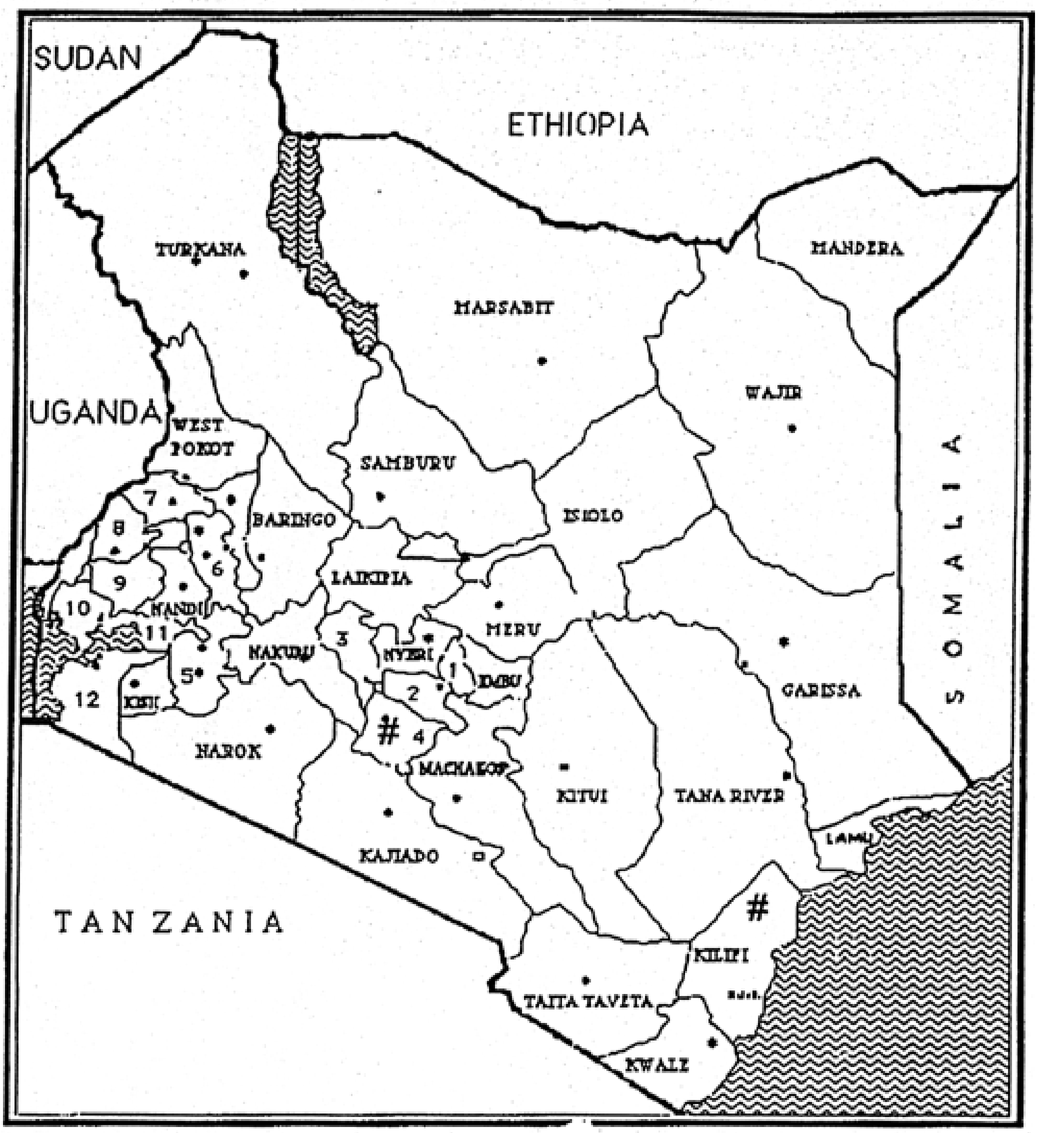
Table 1. Positive cases according to species, 1991 - 1994. (Source: Kabete and Mariakani Investigation Laboratories.)

	1991	1992	1993	1994
Human	47	31	49	UK
Dogs	112	129	113[1]	40 [1]
Cattle	32	46	50	21
Cats	17	4	7	1
Sheep	3	3	4	0
Goats	1	2	6	2
Horses	3	1	3	4
Camel	0	1	0	1
Pigs	0	1	0	3
Wildlife[2]	4	18	7	0
Total animal	172	205	190	72

UK = Unknown, no data available.

1 Mariakani VIL cases not included.

2 The wildlife species include: honey badger, mongoose, bat, jackal, squirrel, wild-dog, leopard and hyaena.



KEY

1 - KIRINYAGA	5 - KERICHO	9 - KAKAMEGA
1- MURANGA	6 - KEIVO - MARAKWET	10 - SIAYA
3-NYANDARUA	7 -TRANS NZOIA	11 - KISUMU
4 - KIAMBU	8-BUNGOMA	12 - SOUTH NYANZA

Figure1. The districts of Kenya, showing the locations of the Veterinary Investigation Laboratories (VILs). # VIL which diagnoses rabies. *other VIL.

Diagnosis

The majority of specimens arrive at the laboratory as carcasses. A few heads are submitted. Some live dogs are brought in for observation and are released to their owners if they do not develop rabies. Opening of the heads is done in our pathology laboratory and the brain specimen is subsequently sent to the virology laboratory. With human cases a brain sample or occasionally a saliva sample, is submitted.

The FAT and mice inoculation are the only methods of rabies diagnosis routinely carried out in the country at the moment. The FAT is done on all samples as a first procedure, then all cases that are suspicious on FAT and all cases with a history of human contact are put into mice. Results of FAT can be available within two hours of submission of the specimen while results of mice inoculation take up to 30 days. Depending on the urgency, the results are communicated by telephone.

Control

In Kenya, legislation exists to ensure adequate control of rabies. If fully implemented, such legislation would be perfectly effective in the control and eventual eradication of the disease. However, in most cases, we operate under significant financial constraints leading to shortages of vaccines and insufficient logistical support for rabies control policies.

Our control measures are a combination of dog vaccination, destruction by baiting of "stray" dogs and restriction of dog movement.

The main factor limiting the efficacy of dog rabies control at the moment is our inability to vaccinate an adequate proportion of the dog population during any one year. Furthermore, given the rapid turnover of the dog population, it is essential that any such vaccination programme is sustained in the long-term, and not carried out as a "one-off" exercise.

Kenya's human population in 1994 was approximately 24 million people. Using a dog:person ratio of 1:8 (Bögel and Meslin 1990) and 1:6.5 in Zimbabwe (Brooks 1990), Kenya's dog population in 1994 can be estimated at between 3 - 3.7 million. To eliminate canine rabies in the country, it is desirable that approximately 75 percent of the dog population be vaccinated, i.e. approximately 2.3 - 2.8 million dogs. As all the vaccine used in the country is imported, the kind of funding necessary to mount and sustain campaigns of this nature would be colossal and unaffordable through the government's normal expenditure allocations.

The amount (doses) of rabies vaccine purchased in recent years is as follows: 44 000 in 1991, 150 000 in 1992, nil in 1993 and 150 000 in 1994. The use of vaccine doses is as follows : 85 026 in 1991, 96 510 in 1992, 42 108 in 1993 and 42 249 up to October 1994. It is therefore evident that the country has fallen short of the amount of vaccine required to conduct meaningful vaccination campaigns.

Vaccine development

Production of LEP vaccine started at Kabete in 1967 and continued until 1989 when it was discontinued due to equipment breakdown. Commercial vaccines made outside Kenya have been in use since then. However, with increasing use of tissue culture systems worldwide, Kenya Veterinary Vaccine Production Institute (KEVEVAPI) is developing an inactivated rabies vaccine in tissue culture. It is hoped that with the current equipment production of approximately one million doses of the vaccine will be possible by the end of this year.

Production levels can be increased substantially (up to 10 million doses per year) with the proposed acquisition of modem equipment.

Acknowledgement

I wish to register my gratitude to the Director of Veterinary Services, Kenya for enabling me to attend the meeting. While compiling this paper, I held useful discussions with officers involved in rabies work in various institutions. The officers include: Dr K. Bangat and J. Macharia (Department of Veterinary Services, Kenya), Dr J. McDermott (University of Nairobi), Dr Githaiga (Kenya Wildlife Service), Dr KinyiIi (Kenya Veterinary Vaccine Production Institute), Dr Ngichabe (Kenya Agricultural Research Institute) and Dr Sigei (Ministry of Health).

References

Bogel K and Meslin FX (1990). Economics of human and canine rabies control: Guidelines for programme orientation. *Bulletin of the World Health Organization* **63***:* 2 81-29 1.

Brooks R. (1990) Survey of the dog population of Zimbabwe and its level of rabies vaccination. *Veterinary Record* **127***:* 592-596.

RABIES IN TANZANIA

Y.S.S. Sembiko

Veterinary Public Health, Ministry of Agriculture, Dar-es-Salaam, Tanzania

Rabies has been reported in Tanzania since 1954 in a locality in the southern highland regions. Control strategies managed to confine the disease to these regions until 1969 when the disease started to spill over into other regions. The main cause of the spread was major political changes in 1967 which declared the country a socialist State and caused increased mobility of people and collapse of zoosanitary infrastructure. To date, rabies is enzootic in almost all districts with the exception of the southern regions of Mtwara and Lindi, where the disease has not been reported. No reports of the disease in these regions does not guarantee the absence of the disease. Poor zoosanitary infrastructure and lack of awareness of the public in these regions may be an alternative reason for no reports.

Substantial effort has been applied to contain the disease in enzootic areas, but funds to sustain vaccination programmes are inadequate. It is now apparent that lack of awareness, irregular supply of vaccines, inaccessibility of dogs and inadequate funds to run vaccination campaigns are responsible for low vaccination coverage.

For the past three years rabies cases in both humans and dogs have been increasing. Regions which were quiescent are now reporting high incidences.

Nationwide, cases (confirmed and unconfirmed) numbered 457 in 1992, 404 in 1993 and 877 in 1994. Human cases are shown in Table 1.

After the political instability in Rwanda and Burundi in March 1994, with resulting heavy influx of refugees with dogs along the borders, a severe rabies outbreak was triggered which claimed several lives in Kibondo, Kasulu, Ngara and Karagwe districts. Neighbouring villages are reporting a high incidence of rabid dog bites which are unattended because of the unavailability of post-exposure immunisation. Few people are able to travel to Uganda for post-exposure immunisation at US$ 60, or travel to Bukoba where they pay US$ 35 - 50 in private hospitals.

The rabies status in other districts is similar with the exception of the ever endemic region of Mbeya in the Southern Highlands, which although reporting the highest numbers of dog bites, has reported no human deaths for the past three years. This is attributed to mass sensitisation which increased awareness of post-exposure immunisation and control of rabies in dogs.

Wildlife rabies has been reported in the past in hyaenas, jackals and vervet monkeys. Studies conducted in the Serengeti ecological zone has shown no evidence of a wildlife rabies cycle.

All cases of rabies in wildlife were associated with the domestic dog. However we cannot rule out the presence of wildlife rabies in Tanzania.

Table 1. Human exposure to dog bites and deaths due to rabies. (UK- Unknown, no data available.)

Year	Bites	Immunisations	Deaths
1980	1141	712	49
1981	1914	983	51
1982	392	20	5
1983	UK	UK	UK
1984	96	UK	UK
1985	709	216	5
1986	2457	681	55
1987	3019	2467	71
1988	2479	2493	13
1989	1772	1043	13
1990	1672	1023	8
1991	2565	2565	17
1992	5819	5819	13
1993	1044	1044	8
1994	UK	UK	58

Rabies surveillance and diagnosis

Zoosanitary infrastructures for rabies surveillance and diagnosis nationwide are at a state of collapse. At present only the facilities at the Veterinary Investigation Centre in Arusha are capable of receiving samples for histopathology only. The Sokoine University of Agriculture Veterinary Clinic receives samples from Morogoro and neighbouring regions for histopathology and FAT. The Central Veterinary Laboratory has the capacity for histopathology and FAT.

The major problem in rabies diagnosis is submission of samples. The network of Veterinary Investigation Centres can receive samples but they cannot mail the samples to the diagnostic laboratories due to financial constraints.

The lack of appropriate formats for reporting, the national database for processing of the data and the feed-back mechanism are our main constraints. Also, availability of sampling kits and equipment contribute to the problems.

Rabies control

The problems involved with rabies control has prompted the Ministries of Agriculture and Health to work together on the development of a national rabies control programme which envisages control and eradication of canine rabies.

To arrest the current epizootic along the border areas with Rwanda and Burundi, the French Food Aid Counterpart fund supported the Ministry of Agriculture by donating US$ 50 000 to conduct an emergency rabies campaign in refugee invaded districts. This campaign has started and we expect to vaccinate 50 000 of the 110 000 dogs in Kigoma and Kagera Regions. We target less than 50 percent vaccination coverage in these areas because of the type of dog ownership. Most dogs in the area are neighbourhood dogs, many of them owned by children for sporting purposes. Refugee dogs are not targeted because they are not accessible.

Plans are underway to approach Animal Protection Associations at home and abroad to assist in the evacuation of these dogs.

The Tanzania National Park is soliciting funding from Wildlife Conservation Societies to implement a three year canine rabies and canine distemper vaccination campaign in the Serengeti ecological zone to protect susceptible wildlife species at the Serengeti National Park.

Table 2. Numbers of dogs and cats vaccinated and destroyed.

	Vaccination		Destruction	
Year	Dogs	Cats	Dogs	Cats
1980	21139	50	20093	21
1981	42740	240	22028	630
1982	18433	121		21
1983	33624	949	5562	42
1984	19338	12	2913	45
1985	14319	9	1488	55
1986	86427	2 129	1498	914
1987	50620	1816	13655	172
1988	97122	690	8453	228
1989	44388	460	3566	71
1990	47328	520	3350	90
1991	15601	1442	1660	133
1992	11635	236	1448	UK

New approach

The Ministry of Agriculture with the Ministry of Health plans to implement a National Rabies Programme. This envisages control and eventual eradication of canine rabies by:

1. Implementation of a pilot project where evaluation of efficacy of different vaccination strategies will be tested.
2. Establishment of an adequate zoosanitary infrastructure.
3. Establishment of an institutional framework to strengthen intersectoral co-operation at all levels.
4. Mass sensitisation for responsible dog ownership.
5. Vaccination of 75 percent of the dog population.
6. Strengthening of rabies surveillance and training.
7. Rationalisation of international and technical co-operation.

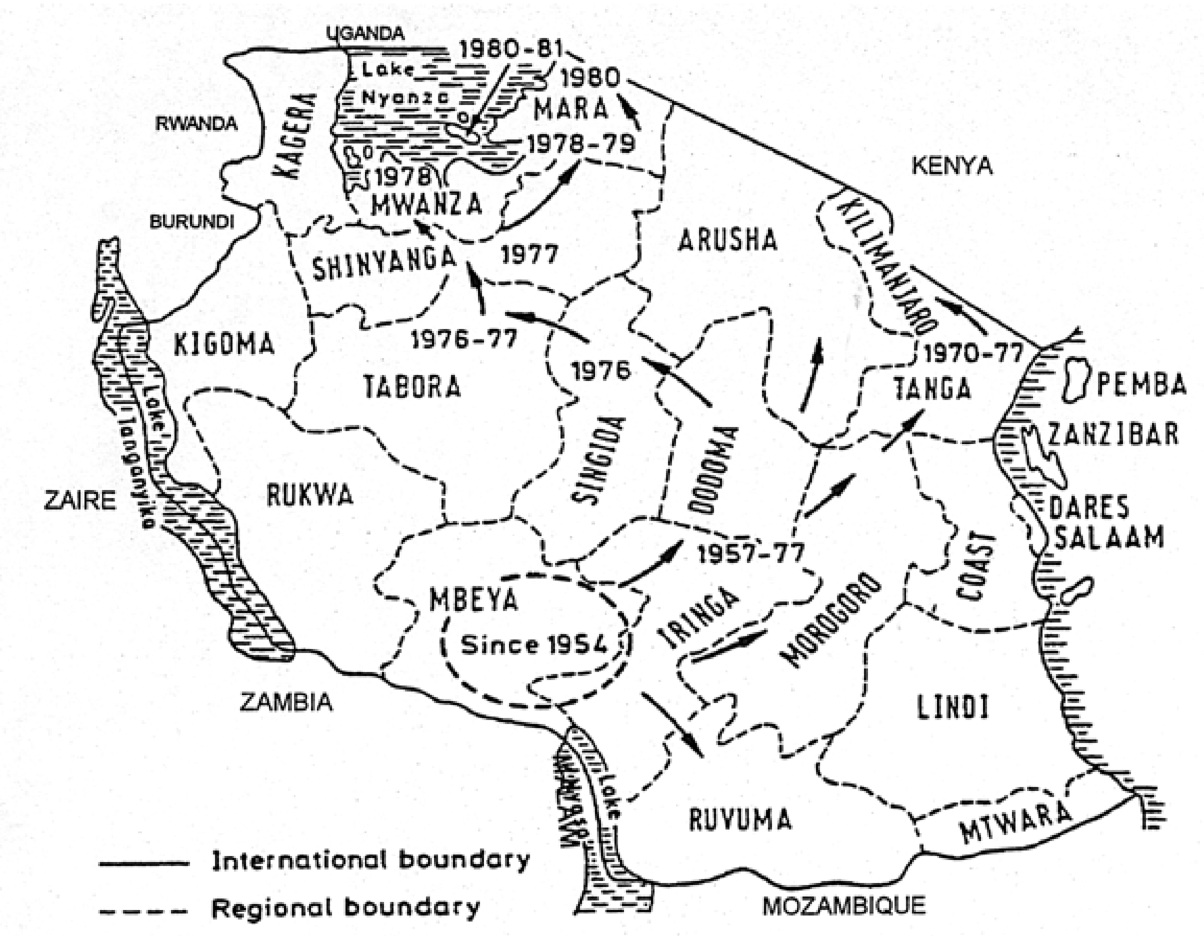


Figure 1. Map of Tanzania, showing the spread of rabies.

RABIES IN MALAWI

Chrissie N. Mwiyeriwa

Liwonde Agricultural Development, Liwonde, Malawi

The total diagnosed rabies cases from 1992 to 1994 are shown in Table 1. In 1992, 174 out of 231 submissions for rabies were confirmed positive by the Central Veterinary Laboratory, while the Southern Region (Blantyre) Laboratory confirmed 50 cases. In 1993 the CV1, alone had 198 submissions of which 154 were positive.

Of the positive cases, there is a very high percentage (80 percent) of dog cases every year. The number of cattle diagnosed positive should be a cause for concern. Considering that specimen submission is far from complete, the countrywide picture is probably much worse than the trend portrayed.

Table 1. Numbers of rabies cases in Malawi 1992 to 1994. Source: Central Veterinary Laboratory.

Species	1992	1993	1994	Total
Dog	150	149	187	486
Cat	3	2	3	8
Cattle	10	14	17	41
Goat	2	2	7	11
Sheep	1	3		4
Pig	2	1		3
Jackal	1	2	1	4
Hyaena	5	1		6
Civet		2		2
(Unclassified)	50			50
Total	224	176	215	615

Diagnostic Services

Malawi has two laboratories capable of carrying out rabies diagnosis: the Central Veterinary Laboratory (CVL) and Southern Region Laboratory in Blantyre. Both diagnose rabies using the fluorescent antibody test (FAT). Any specimen from an animal with a history of human contact is subjected to biological testing using mice.

It should be emphasised here that the level of sample submission has continued to decline. It would therefore not be an exaggeration to say that these positive cases may just be the tip of the iceberg.

Control Measures

Malawi, like many other countries, has followed the system of mass vaccination of dogs as the chief means of rabies control. Nation-wide vaccination campaigns are followed by tie-up orders and any stray dogs are destroyed. Even though this exercise never really manages to vaccinate more than 10 percent of the dogs, the destruction of stray dogs and unwanted dogs had a somewhat significant impact. Campaigns are often conducted only during emergency situations, especially around urban areas.

Rabies control implementation suffers from inappropriate administrative structures within government poor staffing, slow decision making, lack of funds and low priority compared to other diseases such as malaria and AIDS.

In an effort to strengthen the control measures, rabies control and extension resources have been prepared by the Livestock Disease Evaluation Project and are being established in a Rural Development Project (Lilongwe West RDP) in the central region. It is hoped this approach will be extended to all the RDPs in the country.

One more aspect worth mentioning here is that even though it is well appreciated that rabies is a zoonotic disease, no formal co-ordination between the two Ministries of Agriculture and Livestock Development and that of Health and Population exist.

Human rabies

For the years under review, it was not possible to get complete figures for human contacts. However, from January 1993 to June 1994, in the central region only, 5000 people (2826 men and 2174 women) were bitten by dogs. During the same period there were 25 human deaths due to rabies.

It is appreciated that treatment of humans after exposure to rabies is very expensive, almost K500 (US$33) per case. In Malawi the cost cannot be afforded by most people and the cost to government is far above sustainable levels. From January to February 1995 already 118 people have been recommended for anti-rabies treatment.

Rabies is a zoonotic disease and as such there is justification for more meaningful support, since both humans and animals are at risk. But the situation in Malawi is not encouraging, as the rabies control programme is far from being adequate. For instance, there seems to be no direct co-ordination between medical and veterinary personnel.

RABIES IN ZAMBIA

Hetron M. Munang'andu

Central Veterinary Research Institute, Lusaka, Zambia

Zambia is a landlocked country with estimated human and dog populations of 7.3 million and over 800 000 respectively at the end of 1994. Rabies has been a public health hazard since the 1900's. It was officially confirmed in 1913 when diagnostic facilities became available. Until recently rabies could only be confirmed at the Central Veterinary Research Institute (CVRI) in Lusaka. As a result only a few rabies cases were reaching the laboratory for confirmation. To counter this under reporting of rabies cases, diagnostic facilities were introduced in the Regional Diagnostic Laboratories (RDL's). With this decentralisation of rabies diagnosis, more cases were reaching the laboratories for confirmation.

In Zambia canine rabies is the most prevalent and dogs are considered to be the main vectors of the disease to other animal species. The prevalence of bovine rabies cases is also high compared to other species. Although rabies is a known problem in the wild animals of other countries in southern Africa, little has been reported from the wild animals of Zambia during the period 1992 to 1994.

Table 1. The number of rabies cases in Zambia by species.

Species	1992	1993	1994	Total
Human	24	55	13	92
Domestic animals				
Dog	30	35	36	101
Cat	3		2	5
Cattle	8	12	1	21
Goat	2		2	4
Sheep	1			1
Horse	2	1		3
Wildlife				
Fox	1		1	2
Jackal			1	1
Baboon			1	1
Total cases	71	103	57	231

Human rabies

The incidence of human rabies in Zambia is quite high, contributing about 39.7 percent of the total confirmed rabies cases during the period 1992 to 1994 (Table 1). The provincial distribution of rabies cases showed more cases in Lusaka than the rest of the provinces in the country with the highest incidence being in 1993 (Table 2).

Table 2 shows that the most affected age group consisted of people who were 15 year or older (74 cases), followed by the age group of people between 5 and 14 years old (18 cases) while those below 4 years of age only contributed 1 case of the total confirmed rabies cases for the period 1992 to 1994.

Table 2. Human rabies in Zambia, classified according to province and age classes: 4 years and below (<4), 5-14 and 15 years and over (15+).

	1992			1993			1994		
				Age					
Province	<4	5-14	15+	<4	5-14	15+	<4	5-14	15+
Central		1	1						
Copperbelt		2	7			7		3	1
Eastern		1	2		1	2			1
Luapula									2
Lusaka			3			29			
Northern		1	1			3			
Northwestern		1				2		1	1
Southern		1	3	1	3	3			2
Western			1		2	2		1	1
Total		7	18	1	6	48		5	8

Rabies in domestic animals

The total number of domestic animal rabies cases during the period 1992 to 1994 was 135, of a total of 305 submissions. There were more canine cases (101 cases) followed by bovine cases (21 cases) whilst the rest of the domestic species only contributed 13 cases (Table 1).

The provincial distribution of canine cases, with the exception of Western Province, showed more positive rabies cases in the urban areas along the line of rail extending from Southern (24 cases), Lusaka (50 cases), Central (20 cases) and Copperbelt Provinces (15 cases). The other provinces had considerably fewer cases per province (Tables 3 and 4).

Wildlife rabies

It is interesting to note that rabies has not been recorded in any of the National Parks of Zambia (Rottcher and Sawchuk 1978). This fact cannot be attributed to lack of surveillance or under reporting as qualified biologists permanently stationed in the parks under the Department of Wildlife have always been on guard to report any outbreaks of notifiable diseases like rabies.

Although rabies cases have in the past been reported from wild animals (Akafekwa 1976; Rottcher and Sawchuk 1978; Hussein 1984; Sinyangwe 1992), these have been reported from fanning areas where some wild animal species still occur. Generally there has been little reporting of rabies from wild animals during the period 1992 to 1994 (Table 1).

Table 3. Animal rabies in Zambia by province and year.

Province	1992	1993	1994	Total
Central	8	7	5	20
Copperbelt	8	4	3	15
Eastern	1	0	1	2
Luapula				
Lusaka	16	16	18	50
Northern	2	1		3
Northwestern	3	3	2	8
Southern	7	8	10	25
Western	3	9	5	17
Total	48	48	44	140

Table 4. Canine rabies in Zambia by province and year.

Province	1992	1993	1994	Total
Central	5	7	5	17
Copperbelt	6	5	1	12
Eastern	0	0	1	1
Luapula				
Lusaka	13	10	16	39
Northern	1	1		2
Northwestern	1	1	1	3
Southern	2	5	7	14
Western	2	6	5	13
Total	30	35	36	101

Diagnosis

The diagnosis of rabies is carried out at the Central Veterinary Research Institute and the School of Veterinary Medicine, both in Lusaka, and also in the five Regional Diagnostic

Laboratories, Ndola, Chipata, Mazabuka, Mongu and Isoka Regional Diagnostic Laboratories (see Figure 1).

There are three diagnostic techniques currently in use and these are the fluorescent antibody test (FAT), the mouse inoculation test (MIT) and histopathology. However, the CVRI also has the capability of diagnosing rabies using an enzyme immunoassay technique.

Figure1. The locations of the Regional Diagnostic Laboratories.

Control

In Zambia there is a statutory requirement for mass vaccination of dogs and cats annually, together with elimination of stray dogs. Other domestic animals like cattle, goats and sheep are only vaccinated in rabies-affected areas.

Upon the laboratory confirmation of rabies, legislation stipulates that a "rabies infected area" should be declared. Veterinary officials mount vigorous control programmes. Prohibition of movement, quarantine and isolation of suspected dogs may be enforced in the affected area. Usually control areas cover a radius of 20 kilometres around the nucleus of the outbreak.

Import regulations of dogs from other countries into Zambia stipulate that dogs should be quarantined for a period not less than 30 days. Such dogs should come from a rabies-free area and should have a valid vaccination certificate.

Rabies vaccine is locally produced by the Vaccine Production Unit at the CVRI. However, this is usually supplemented by imported vaccines since the local vaccine is sometimes insufficient.

**Reference
s**

Akafekwa G. (1976) Rabies in Zambia. *Bulletin de I'Office International des Epizooties* **86***:* 373-381.

Hussein N.A., Sharma RX, Ando R. and Chizyuka H.G.B. (1984) Further review of the epidemiology of rabies in Zambia (1975-1982). *Revue Scientifique et Technique de I'Office International des Epizooties* **3**: 125-135.

Rottcher D. and Sawchuk AM (1978) Wildlife rabies in Zambia. *Journal of Wildlife Diseases* **14**: 513-517.

Sinyangwe P. G. (1992) Rabies in Zambia. In *Proceedings of the International Conference on Epidemiology Control and Prevention of Rabies in Eastern and Southern Africa, Lusaka-Zambia, June 2-5 1992*. Edited by Arthur King. Fondation Marcel Merieux. p. 19-21

RABIES IN MOZAMBIQUE

M.P.R. Travassos Dias[1]

National Institute of Veterinary Research, Maputo, Mozambique

Rabies became enzootic in Mozambique in 1908 and it has remained so to the present time. The disease represents a threat to public health and causes veterinary and human health authorities continuous concern regarding appropriate measures of vigilance and disease control.

As in most countries of the region, dogs are by far the most important animal species responsible for the transmission of rabies, either between animals, or to man. So far there is no evidence that other animal species play an important role in this process. There are no reports of wild animals showing unusual behaviour or biting people. Table 1 tends to confirm this point, although it is known that such figures are an underestimate of the number of rabies cases that may occur, and most of the cases still come from urban areas.

Table 1. Animal species diagnosed with rabies.

Species	1989 to 1991	1992 to 1994	Total
Dog	26	47	73
Cat		2	2
Monkey	1		1
Goat	2	3	5
Water buffalo		1	1
Total	29	53	82

In the period from 1992 to 1994, as compared to the three previous years, more positive cases were confirmed by the laboratories. The increase (45.3 percent) is probably a consequence of the end of civil unrest in 1992, when the peace situation allowed better communication around the country.

During 1992 to 1994 animal rabies was reported in all of the provinces, but most of the cases were from Maputo (11), Manica (11), Nampula (8), Craza (6), Sofala (6) and Zambezia (5).

[1] Co-author and presenter: F. Rodrigues, National Directorate of Livestock Maputo, Mozambique.

Table 2. Confirmed rabies cases in Mozambique

	1992	1993	1994	Total
Animal rabies	18	14	21	53
Human rabies	42	9	7	58

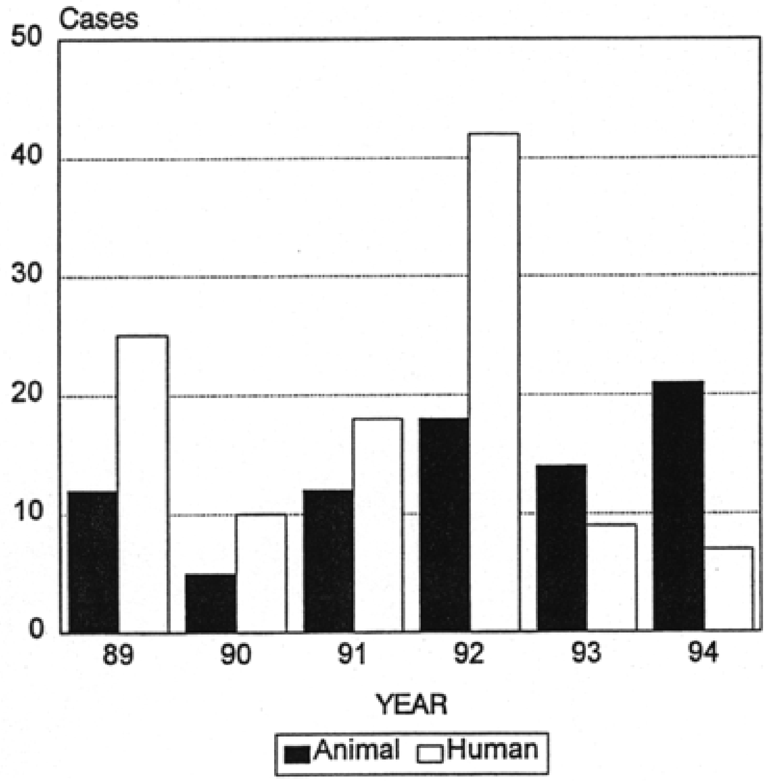


Figure 1. Confirmed rabies cases 1989 - 1994.

Monthly reports sent by the Provincial Veterinary Services to the National Directorate of Livestock mention only very few rabies cases that were not confirmed: three in Sofala province in 1992, and in Cabo Delgado province, two cases in 1992 and one case in 1993.

However, the situation is believed to be much more severe. In fact dogs in rural areas, where there are no resources either for human treatment or confinement and observation of biting dogs, are usually killed, as they are known to cause rabies. These dogs are recognised by showing signs of change of behaviour, salivation, open mouth, roaming and biting people. Nampula and Cabo Delgado provinces have reported several such instances and deserve particular attention regarding better facilities for rabies control.

Diagnosis

Although the four provincial veterinary laboratories use Seller's method, most of the specimens are still diagnosed by the Central Veterinary Laboratory, at Maputo using immunofluorescence.

Control

The capture and killing of stray dogs is now limited to Maputo and Beira, the two main towns, and is mostly done to eliminate animals. Such operations, if done on a large scale, are considered beneficial as they bring an educational effect and dog owners will tend to keep their animal enclosed, and therefore fewer people will be bitten.

Vaccination coverage of dogs is still far from reaching the desirable level, mainly due to shortage of resources such as vehicles and fuel.

Flury vaccine produced in the Central Veterinary Laboratory is still used, although there are plans to initiate the production of an inactivated vaccine of tissue culture origin. Vaccination is free of charge, except at private clinics, where in Maputo, about 3,000 animals are vaccinated each year. This is half of the annual target.

Table 3. Numbers of people bitten by animals and receiving post-exposure treatment.

	Period 1988-1990	Period 1992-1994
Number of people bitten	2433	2675
Number of people treated	446	478
Percent treated	18.3	17.9

Table 4. Species responsible for biting people, with the percentage of the total animal bites from Table 3.

Animal species	Period 1988 - 1990	Period 1992 - 1994
Dog	2371 (97.4%)	2637 (98.5%)
Cat	22(0.9%)	12(0.4%)
Monkey	25(1.0%)	23(0.86%)
Others	15(0.6%)	3(0.1%)

Human rabies

The number of cases of human rabies is similar to the number of animal cases. In 1992 there was a significant increase of human cases, as Nampula reported 21 cases. During the war, towns became overpopulated with people that came from rural areas with their animals, (chickens, goats, dogs).

Most of the reports of human cases are based on clinical symptoms and history of the disease.

Post-exposure treatment is given according to WHO recommendations. Reports of the medical authorities in Maputo town, with a population of nearly 2 million, give some figures of numbers of people who were bitten by animals and of numbers receiving post-exposure treatment (Table 3). These reports allow us to verify that more than 97 percent of people are bitten by dogs (Table 4), of which about 20 percent are stray dogs (537 of 2637 dogs during the 1992 to 1994 period). In the city of Nampula the situation is clearly critical, after reports of 21 human deaths and 426 people bitten in 1992 only.

Conclusions

Confirmed rabies cases do not reflect the real picture of the rabies situation in Mozambique, due to under-reporting caused by poor veterinary infrastructure. For the same reasons, the role that wildlife may play in the rabies cycles is not known.

As a result of the shortage of means to control the disease, the number of human victims is still high.

Availability of human rabies vaccine is of primary importance for adequate control. Availability must be ensured before starting effective information campaigns to get people involved in dog vaccinations and confinement of suspected animals.

RABIES IN MADAGASCAR

Rejo Tsiresy

Ministère du Diveloppement Rural et de la Réforme Foncière, Antananarivo, Madagascar

The current situation.

Table 1. Numbers of specimens confirmed positive in the laboratory. Source: Annual reports of the Pasteur Institute of Madagascar (IPM).

	1992	1993	1994	Total
Dog	89	183	283	555
Cat	2	26	24	52
Lemurs			4	4
Cattle	8	4	11	23
Total	99	213	322	634

Table 2. Numbers of human rabies cases and people treated against rabies. Source: Annual reports of the IPM.

	1992	1993	1994
Number of cases (human)	0	1	0
People receiving treatment	1534	1528	2177

Rabies control measures

a) Epidemiological surveillance

- All imported carnivores must have a vaccination certificate issued by an official veterinarian of the exporting country;
- People are requested to report any suspicious cases to either the veterinary services, the IPM or the nearest of the 53 anti-rabies centres;
- People bitten are treated if the wound is considered dangerous (treatment is free at IPM and anti-rabies centres);
- Biting animals are held under observation for over two weeks;
- A sample of brain tissue is sent to the IPM for confirmation.

Theoretically, the national data collection network is set up but we still need to improve the co-ordination between the Veterinary Services, the IPM, the anti-rabies centres, and private veterinarians.

The diagnostic technique, performed by the IPM is quite satisfactory, but the main limitations to surveillance are:

- Anti-rabies centres are too far away;
- Most of them lack the facilities to ensure proper storage and transport of samples;
- Suspect animals are not captured;
- Sometimes they are killed but samples for rabies confirmation are not taken.

As a result, many rabies cases are missed. Nevertheless, the annual reports give realistic pictures of the situation.

b) Actions taken

- A decree is passed every time an outbreak occurs;
- A panel of measures including an eradication campaign using poison (mainly strychnine sulphate) is implemented locally by the regional joint committee on rabies.

The joint committee on rabies includes representatives of the Ministry of Agriculture (Veterinary Services), the Ministry of Health, the Ministry of Internal Affairs, the Ministry of Defence, the Ministry of Justice, the Ministry of Population, and the Ministry of Culture and Communication.

c) Vaccination of dogs

We use a vaccine manufactured locally. It is a live attenuated Flury strain vaccine maintained by passage on embryo egg culture. Imported inactivated vaccines are also used extensively. Our regulations require us to vaccinate dogs and cats every year. The vaccination is not compulsory, therefore the number of vaccinated animals is small.

Implementation of future rabies campaigns

a) The context:

- The dog is the main if not the sole vector of the disease in Madagascar;
- Stray dogs have no wild prey, they have to return to villages to find their food.

b) The campaign:

Instead of trying to implement an epidemiological study, we intend to conduct an eradication program before the end of 1995. The main features of the campaign will be

- Free vaccination. Pet owners will be requested to vaccinate the animals they want to keep; these animals must wear collars;
- All dogs and cats without collars will be killed;
- Incentives will be proposed to have the community involved in the project.

As a pre-requisite, an expert committee will be called upon to discuss the possible consequences of a dramatic decrease of dog and cat populations on the ecosystem (for example, on the effects of rodent populations).

RABIES IN NAMIBIA

O.J.B. Hilbschle[1]

Central Veterinary Laboratory, Windhoek, Namibia.

Rabies has been enzootic, albeit sporadic, for many years in Namibia. The socio-geographic situation prevailing in Namibia governs to a large extent the nature of the biological cycle of rabies in Namibia.

In the central-northern regions, comprising mostly commercial fanning enterprises, one observes predominantly a vector to victim cycle which can be compared to the so-called sylvatic cycle as seen in Europe. Whereas in the latter case the fox (*Vulpes vulpes)* is the major vector, in Namibia we consider the black backed jackal (*Canis mesomelas)* as the major vector of rabies in this situation. The commercial farming operations rely almost exclusively on cattle, goat and occasionally on sheep production. In contrast to the southern region where commercial sheep farming prevails and hence jackal proof fencing is used, no such precautions are used in the central and northern region. Obviously cattle, sheep and goats therefore become the major victims of rabies transmitting vectors.

In contrast, the far northern densely populated regions of Namibia, which are used for intensive subsistence agricultural activities, experience what usually is described as the urban rabies cycle. In the absence of a substantial number of game, the stray dog acts as vector as well as being the victim. Figure 1 illustrates the high incidence of rabies in bovines, dogs and jackals.

Besides the total yearly incidence of rabies cases, which is relatively high if one accepts that only a small percentage of all affected animals reach the laboratory for examination, it is further interesting to note that rabies in Namibia seems to follow a certain seasonal pattern. Figure 2 shows the peak numbers of positive rabies cases in cattle during the second half of each year, which coincides with the dry season.

As early as 1987 efforts were undertaken to vaccinate the jackal via the oral administration route. In initial experiments we used the "Tübingen bait vaccine" containing the SAD B19 vaccine strain of rabies virus. Results obtained with this vaccine in caged jackals were extremely encouraging yet due to financial constraints and other factors no further investigations concerning the innocuity of the vaccine for other wildlife were done. It is, however, noteworthy that titres obtained 11 months after oral vaccine were well in excess of 1: 1000 (BFA Tübingen, Germany).

If one looks at the distribution of cases on a yearly basis during the last three years of reporting it becomes evident that the highest number of rabies cases was recorded during 1992 (Table 1). It is suggested that the high incidence during 1992 is directly related to the severe drought situation the subcontinent as a whole experienced during that year.

[1] Co-author: **Nt Uanguta,** Central Veterinary Laboratory, Windhoek, Namibia.

Table 1. Confirmed rabies cases, 1992-1994.

Species	1992	1993	1994	Total
Domestic animals				
Dog	31	18	12	61
Cat	4	1	2	7
Bovine	72	27	46	145
Caprine	4	8	7	19
Donkey	2			2
Horse	1			1
Ovine	8	2	6	16
Pig			1	1
Wild animals				
Jackal	28	14	13	55
Bat-eared fox	1	1	4	6
Honey badger	3			3
Meerkat	1	2		3
Wild cat	1	1		2
Baboon			2	2
Kudu	1			1
Total	157	73	94	324

Looking at the number of wildlife cases diagnosed from 1987 up to 1994 one observes two distinct peaks during 1988 and 1992 which coincide with relatively dry years. One tends therefore to predict that harsh environmental conditions favour the spread of rabies in a sylvatic cycle as seen in certain parts of Namibia. A further aspect worth looking at is the ratio of positive cases (laboratory confirmed) to the total number of suspected cases submitted to the laboratory. During 1992 - 1994 a total of 314 cattle suspected with rabies were submitted to the CVL. Of these 149 (47 percent) were found positive. In the other cases it was possible to establish other causes responsible for central nervous symptoms, such as lead poisoning and *Anaplasma* infections amongst others. Similarly a small percentage (18 percent) of dogs submitted for rabies examination were found positive for rabies in the central-northern regions, whereas the majority (61 percent) of specimens submitted from the far northern region were confirmed positive.

Diagnosis of rabies is performed with the FAT and in certain cases, if required, a biological test is performed in mice. The conjugate used either originated from Onderstepoort or from the Pasteur Institute.

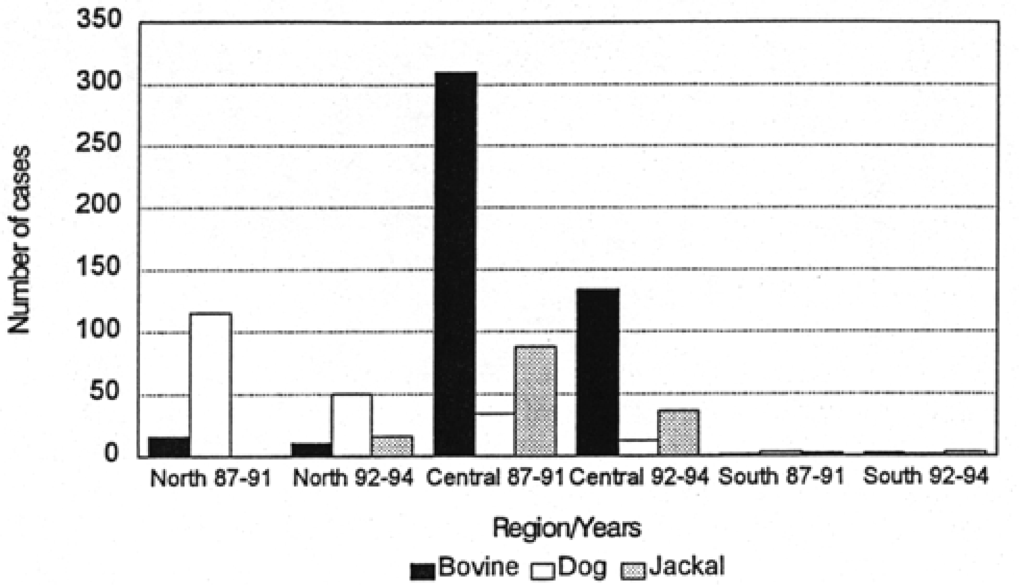


Figure 1. Distribution of confirmed cases.

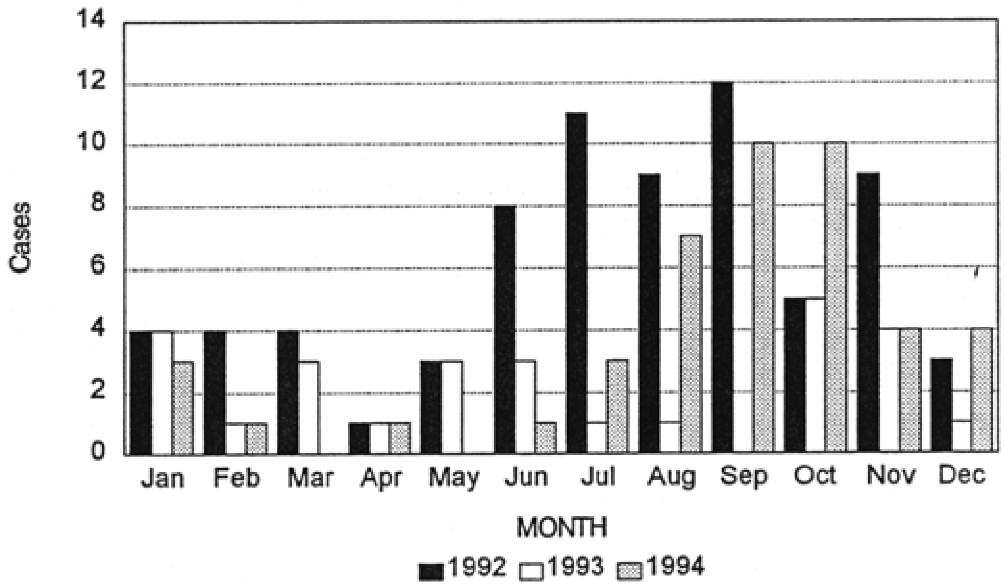


Figure 2. Bovine cases by month.

RABIES IN BOTSWANA

K Sehularo[1]

Department of Veterinary Services, Mahalapye, Botswana

The first occurrence of rabies in Botswana is not known. However, there were apparent outbreaks of the disease in the southeastern parts of the country in 1919 and 1922. According to the literature the first confirmed case of rabies was in a dog in 1938. A more serious outbreak of rabies occurred in northwestern Botswana in the early 1950's. This outbreak spread to the populated eastern and southern parts of the country. The frequency of recorded confirmed rabies cases has since increased significantly and this can be ascribed to increased public awareness brought about by public education about the disease, improved cognisance of the disease, increased submission of samples for rabies diagnosis by the concerned veterinary personnel and improved diagnostic techniques by the National Veterinary Laboratory.

Present situation of rabies in Botswana

During the reporting period 1992 to 1994 the National Veterinary Laboratory received and processed a total of 842 brain samples for rabies diagnosis of which 492 were confirmed positive by FAT and MIT (Table 1). This represents a positivity rate of 58.8 percent.

It is interesting to note that monthly distribution of rabies cases tends to increase from May and reach the highest in the months of June, July, August and September; from then there is a steady decline in confirmed cases. It is thought that this pattern is a result of jackal dispersal and breeding behaviour. Also around winter, grazing is poor, livestock travel long distances to graze and as a result there is increased interaction with wild carnivores.

Using the Tropic of Capricorn as a division between the northern and southern parts of the country, there tends to be more cases in the northern districts than in the southern districts. This might be because the northern parts of the country are more populated than the southern parts.

No human rabies case has been reported during the period 1992 to 1994.

Phylogenetic analysis of 100 rabies virus isolates from Botswana using polymerase chain reaction (PCR) based genetic sequencing has shown that there are two distinct variants: a mongoose variant and a domestic dog variant. Other species may have either variant.

[1] Co-author: D.W. Rumisha, Ministry of Health, Gaberone, Botswana

Table 1. Rabies in Botswana by species and year.

Species	1992	1993	1994	Total
Domestic animals				
Dog	28	17	23	68
Cat		1	2	3
Cattle	98	47	75	220
Goats	41	20	36	97
Sheep			2	2
Horse/donkey	2	2	6	10
Wild animals				
Jackal	19	10	33	62
Honey badger	4	2	2	8
Wild cat	7		1	8
Viverrids	.5	3	2	10
Hyaena		1	1	2
Cape fox	1			1
Mouse	1			1
Total	205	103	184	492

Rabies control

Rabies control is done in accordance with the Animal Diseases Act. It is a notifiable disease, There is free compulsory annual vaccination of dogs and cats by government personnel. Dogs and cats which are three months old or above are vaccinated and a certificate of vaccination is issued to the dog or cat owner. Also dogs and cats are marked with paint and immediately after vaccination, a tie-up order is imposed and dogs without paint are shot. Owners wishing to move their dogs and cats from one zone to another are issued with interzonal permits which should be accompanied by up-to-date rabies vaccination certificates.

Thorough investigation of suspected rabies cases and in-contact animals are carried out and the necessary samples are taken.

In cases of human contact, field officers regard anybody who has touched or consumed cooked meat of a rabid animal to be eligible for post-exposure treatment, until laboratory results are known. If the results are negative post-exposure treatment is stopped. In the case of bite contacts by unvaccinated dogs, the dog is monitored every day for 10 to 14 days. Post-exposure treatment is recommended until the termination of the monitoring period, but is continued if the dog is confirmed rabid.

In cases of animal contacts, action is only taken after laboratory results are known. In the majority of cases all in-contact animals are vaccinated against rabies.

Additional control measures are quarantine, culling of jackals and public education about the disease.

Occupational risk groups (veterinary staff who routinely handle suspected rabid animals) are, as a rule, given rabies prophylaxis.

Discussion

What is most striking about the data is that domestic ruminants (bovine = 44.7 percent and caprine = 19.7 percent) contribute high percentages of the positive rabies cases. This can be attributed to the fact that they are exposed to the three main vectors (domestic dog, jackal and mongoose). Also the population of cattle and goats is several times that of domestic carnivores, hence the total sample submissions from the domestic ruminants is higher than that of dogs, resulting in an increased number of positives. Domestic carnivores are routinely vaccinated against the disease while domestic ruminants are not.

The next highest contribution of cases come from domestic dogs (13.8 percent) and black-backed jackals *(Canis mesomelas)* (12.6 percent). The significance of the percentage of domestic dog rabies is that it reflects vaccination coverage of this species. Overall positivity rate of 13.8 percent is very high considering that domestic carnivores receive compulsory annual rabies vaccination. The high percentage of dog rabies can be ascribed to several factors. Some dog owners, especially those in the rural areas, tend not to bring their dogs for vaccination because they believe it spoils their hunting ability. Rabies vaccination campaigns are carried out at the same time as contagious abortion vaccinations of cattle and this may interfere with the administration of vaccine. Dog owners are expected to bring their dogs to the crush. In some districts veterinary personnel do not go to homesteads to vaccinate those dogs which were not brought to the crush. In urban areas some owners cannot handle their dogs, hence they fail to bring them to the designated vaccination centres.

Conclusions and recommendation

Phylogenetic analysis results have so far shown that there are two distinct rabies virus types: mongoose and domestic dog types. There is an inadequate rabies vaccination coverage of dogs and cats. For rabies incidence in the country to decline, there has to be adequate rabies vaccination coverage of the dog and cat population. Public education about the disease should be intensified. The vaccination has to be co-ordinated properly with house to house vaccination campaigns in both rural and urban areas. This should immediately be followed by destruction of stray dogs. Rabies vaccination campaigns should be carried out separately from other vaccination campaigns. Oral vaccination should be introduced to cover wildlife, in particular jackals. The question of controlling mongoose rabies should be investigated.

RABIES IN ZIMBABWE

John Bingham

Veterinary Research Laboratory, Harare, Zimbabwe

The total confirmed rabies cases from 1992-94 are shown in Table 1. Figures 1-3 show these cases displayed on the map of Zimbabwe.

Table 1. Rabies in Zimbabwe: Total laboratory-confirmed rabies cases from 1992 to 1994.

Species	1992	1993	1994	Total
Human	1	1	3	5
Domestic animals				
Dog	212	78	119	409
Cat	14	5	3	22
Cow	141	69	61	271
Sheep	1	1	3	5
Goat	16	4	13	33
Horse	1	1	1	3
Donkey	5	2	2	9
Wild animals				
Jackal	146	101	270	517
Mongoose	4		3	7
Genet	2		1	3
Civet	7	2	6	15
Aardwolf		1	1	2
Hyaena	1	1		2
Honey badger	7	3	3	13
Wildcat	3			3
Serval	1			1
Leopard			1	1
Cheetah		1		1
Eland	1			1
Kudu		1	1	2
Duiker	1			1
Sable	2			2
Total animals	565	270	488	1323

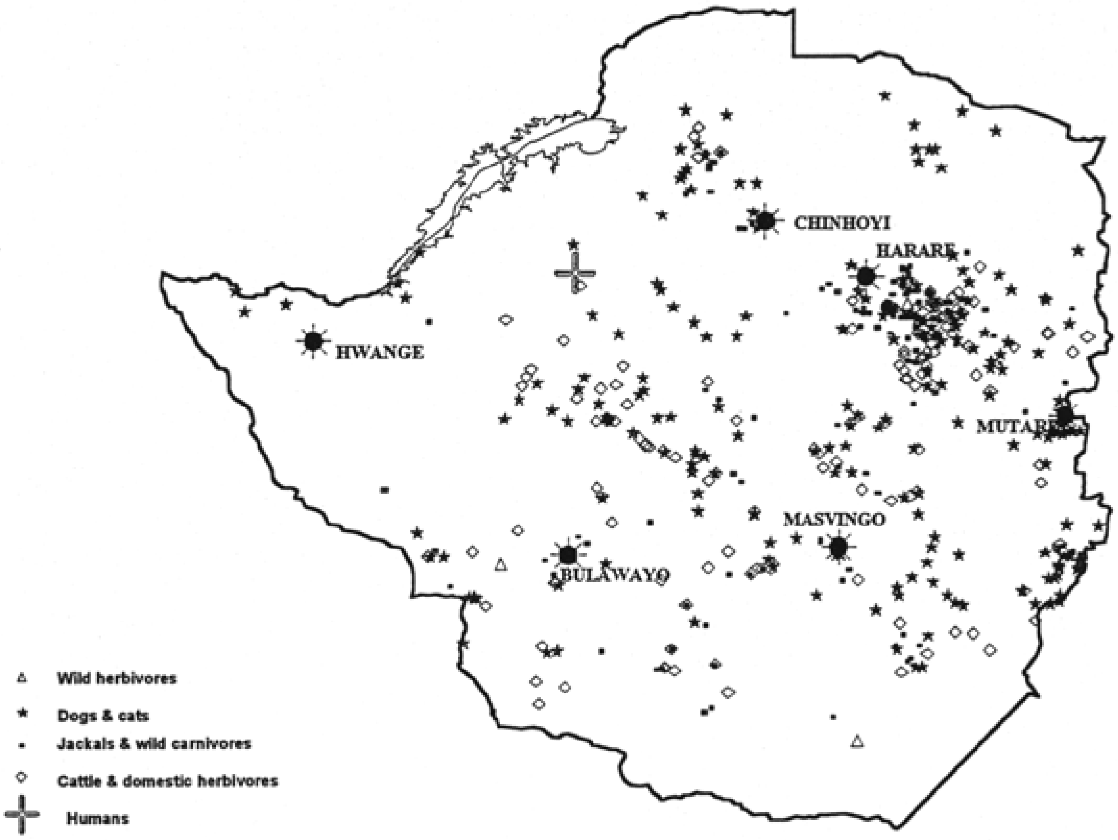


Figure1. Rabies distribution, 1992

Canine rabies

Rabies in domestic dogs declined from a peak of 219 cases in 1991 to the lowest for many years in 1993. As has been the case in most previous years, many of the dog cases originated from communal (rural subsistence farming) areas. In addition, a high proportion of dog cases was associated with jackal epidemics in 1992. The reason for the decline in dog cases during 1993 is not known but may be related to the period of drought during the 1992/93 season which probably resulted in a decline of dog numbers.

Rabies in livestock

As usual this mainly affected cattle. Cattle rabies was distributed throughout most of the country and appeared to be associated with both jackal and dog cases.

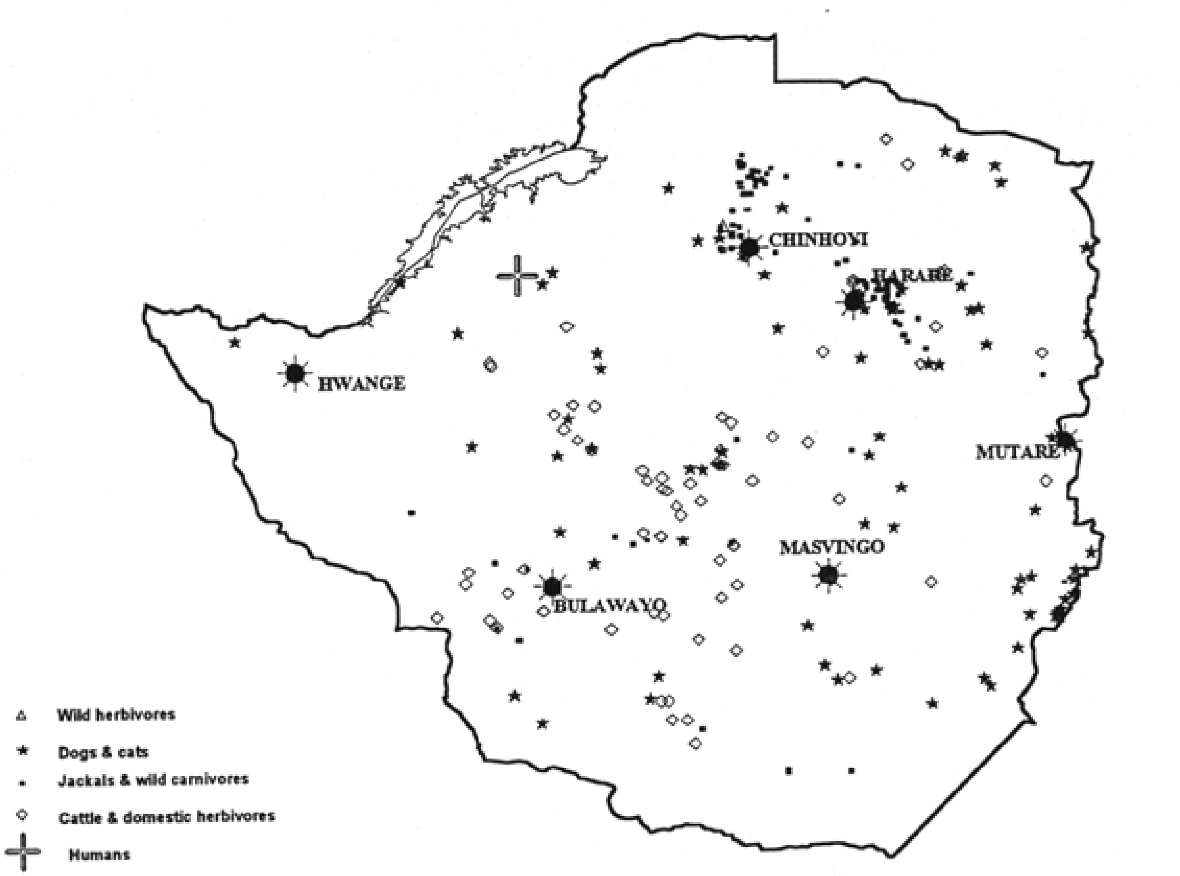


Figure 2. Rabies distribution, 1993

Wildlife rabies

The large jackal rabies epidemic which begun in the Marondera area, southeast of Harare, in 1990 and 1991 continued to spread northwards and westwards. It encroached the Harare peri-urban areas in 1993 and spread northwards.

During 1991 rabid jackals were detected in the Chinhoyi area. In 1993 and 1994 this epidemic spread northwards and eastwards and, to a small extent southwards to the area west of Harare. This epidemic became very intense during 1994.

During the period 1992-94 the southern Lowveld and Midlands regions of the country also experienced jackal rabies, although this was at a lower level and more sporadic than in the Highveld areas.

Rabies in carnivores other than jackals was reported mainly in civets *(Civettictis civetta),* honey badgers *(Mellivora capensis)* and mongooses - the white-tailed *(Ichneumia albicauda),* large grey *(Herpestes ichneumon)* and Meller's *(Rhynchogale melleri)* mongooses. These cases were generally associated with jackal epidemics.

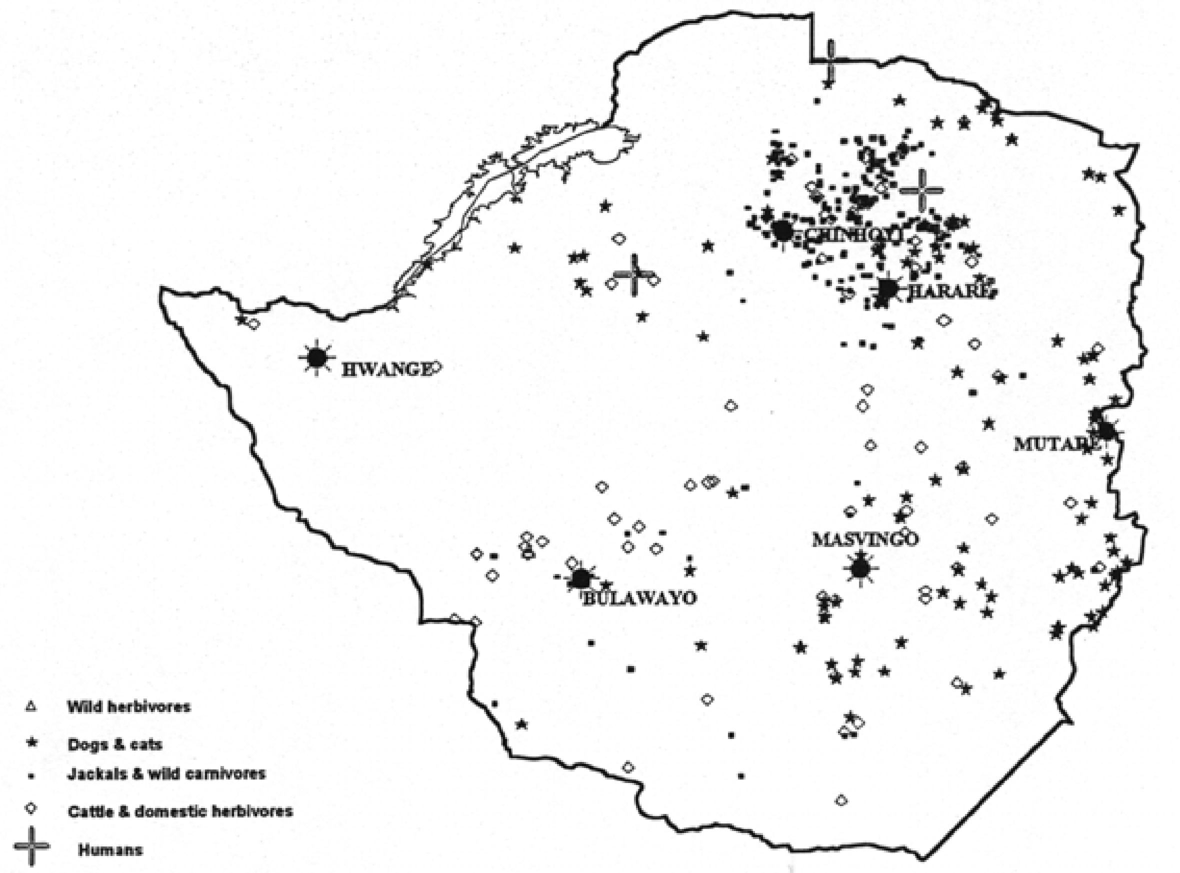


Figure 3. Rabies distribution, 1994

Rabies in Humans

Five cases of human rabies were confirmed during the period 1992 to 1994 inclusive. Their details are given in Table 2. None of these cases received post-exposure treatment.

In addition several cases were diagnosed on clinical grounds only. In these cases, dogs appear to be the vector species, except in one case which was bitten by a jackal.

Laboratory diagnosis

The main diagnostic test for rabies remains the fluorescent antibody test with mouse inoculation being the main back-up test. Histopathology is also used. During 1994 three human cases were diagnosed positive by ante-mortem. skin biopsy technique, using frozen sections stained with fluorescein-conjugated anti-rabies serum.

Table 2. Details of confirmed human rabies cases from 1992 to January 1995.

Date	Place of Origin	Age Years	Sex	Vector	Site of bite	Incubation period
Oct 1992	Gokwe	13	M	Dog	Leg	43 days
Aug 1993	Gokwe	2	M	Dog	Face	10 days
Mar 1994	Guruve	6	M	Dog	Foot	3 1 days
Mar 1994	Gokwe	6	F	Dog	Leg	25 days
May 1994	Bindura	20	M	Jackal	Arm	44 days
Jan 1995	Kamativi	3	M	Dog	NK	1.5 mths?

NK = Not known.

Control

The number of doses of vaccines given to dogs countrywide was 638 000 in 1991, 482 000 in 1992 390 000 in 1993 and 270 000 in 1994 (see page 134, "Rabies control in dogs in Zimbabwe). Most of these vaccines were given during mass vaccination campaigns in communal areas, where dog owners were requested to bring their dogs to central vaccination points.

RABIES IN SWAZILAND

Roland X. Dlamini[1]

Veterinary Department, Nlangano, Swaziland

Rabies is a deadly disease in both animals and human beings. Its control calls for an understanding of the disease concept and co-operation amongst various government Ministries including Agriculture, Health and Police.

Cases of Rabies in Animals (1992 - February 1995)

Between 1992 and 1994 an average of 22 cases per annum were confirmed in the laboratory. There has been a general increase in the number of rabies suspect specimens submitted to the laboratory (see Table 1). There also seems to be a sharp increase in the number of positive cases. In the first two months of 1995 the laboratory confirmed 10 cases. It should be noted that the laboratory only uses the fluorescent antibody test (FAT) and so false negative results are possible. Introduction of a biological test would be of benefit.

Table 1. Number of animal rabies cases for the period 1992 to February 1995. (Source: Manzini Veterinary Investigations Laboratory.)

Year	Total specimens	Number of positives
1992	29	17
1993	42	14
1994	63	33
1995 (Jan/Feb)	15	10

Human Rabies

A total of 5 suspect cases in humans were reported between 1992 and to date. This figure could be much higher as hospital records do not accurately reflect the rabies situation. For example, animal bites in hospitals are recorded as injuries, no records for those receiving post-exposure rabies vaccine are kept and there is no follow-up. Human contacts are not always followed up when a rabid animal is found. Not every dog-bite case in the community, especially minor bites in rural areas, goes to hospital.

[1] Co-author: L.D. Mathunjwa, Ministry of Health, Mbabane, Swaziland

When clinical rabies develops, some victims are taken to prophets and traditional healers. In clinics such cases can be misdiagnosed as a psychiatric illness, especially where the history is not clear. In epidemic areas the Veterinary Department has taken responsibility for alerting the medical staff, educating nurses at seminars, suggesting guidelines, tracing victims and referring them to hospital for vaccination.

Some victims do not finish their vaccination courses, and some contacts are not vaccinated at all. The Veterinary Department attempts to follow up such cases. Some nurses resent treating patients referred by veterinary officers. Rabies vaccine is only available in hospitals and the larger health centres. Some patients also resent being referred to hospitals which means travelling long distances for vaccinations of which they hardly understand the purpose.

Rabies control measures in place

The Kingdom of Swaziland conducts annual vaccination campaigns which attempt to vaccinate all dogs. Animals brought to the various veterinary stations are vaccinated all year round. In September the Veterinary Services conducts vaccination campaigns at diptanks and other community centres. Certificates are issued for vaccinated dogs. In the past, vaccinated dogs were tattooed but this was stopped because it was causing injuries to dogs and inconvenience to personnel.

The Veterinary Services conducts an annual census in August of all domestic animals and thus it is easy to compare the total dog population with the number of dogs vaccinated (Table 2). If coverage in a diptank area is poor, re-vaccination is conducted following meetings to encourage owners to bring their dogs.

Veterinary Assistants are constantly on the lookout for suspect rabies cases and report immediately to Government Veterinary Officers. In the case of an outbreak, diptank areas which fall within a radius of approximately 6 km. are declared Rabies Guard Areas and a "tie-up" order is instituted. Compulsory vaccination is conducted immediately, followed by a "shoot-out" campaign where dogs that are not tied and those that are not vaccinated are shot.

Table 2. Rabies vaccination coverage for the period 1992 to 1994. (Source: Annual reports, Ministry of Agriculture.)

Year	Dog Population	Dogs Vaccinated	Percentage Coverage
1992	74177	52260	70.5
1993	78790	57202	72.6
1994	72932	45291	62.1

Discussion

Improvements in identifying vaccinated dogs are necessary, since the present method has loopholes. Means of reviving and improving the tattoo are being considered. Rabies awareness campaigns are being extended to clinics and schools. Co-operation between the police and the Veterinary Department especially during shoot-out campaigns is very important. Rabies control measures need to be strictly adhered to.

A considerable number of rabid dogs were between 2 and 6 months of age. These dogs either were not born or were not old enough for the annual campaign in September. For this reason it may be necessary to vaccinate every three months in problem areas targeting such puppies. There is definitely a need to introduce another laboratory method to compliment the FAT, which is not infallible.

The shoot-out campaign is a very effective method for encouraging dog owners to vaccinate. It is also very popular with the veterinary staff. Dogs from areas distant from where the shoot-out is conducted are also presented for vaccination as the campaign spreads.

Close co-operation between the Veterinary Department and the Ministry of Health is highly recommended. Suspicious bites of humans by animals need to be reported promptly to the Veterinary Department for investigation. The Veterinary Department needs to refer people exposed to rabid animals immediately to hospitals, stating clearly that such people require post-exposure rabies vaccine. Hospitals need to keep adequate records so that it is possible to follow up cases. Refresher courses for the nursing staff and veterinary assistants are necessary. The Ministry of Health needs to formulate a policy or guidelines on management of suspected rabies cases.

RABIES IN LESOTHO

Lebohang Khomari

Department of Livestock Services, Maseru, Lesotho

Lesotho is divided into four geographic zones according to elevation. They are the highlands, the foothills, the rift valley and the lowlands. The elevation ranges from 1388 m to 3482 m. The average minimum temperature is -CC with snow in winter and -5'C in the lowlands. These geographic zones and the cold climate play an important role in the rabies control programme. Because of the inadequate rangeland, seasonal movement of livestock is highly important. Thus in summer livestock is grazed in the highland areas while in the winter the animals are moved to the foothills or lowlands. Herdsmen tending the livestock will frequently work with up to as many as 10 dogs. Though no studies of dog population sizes have been conducted, by using the human to dog ratios the dog population in the rural areas is thought to be high.

It is only in winter that any rabies vaccination programme could be implemented because then the dogs are in areas that are accessible by road. Secondly there is an economic and technical advantage: due to economic constraints it is not easy to keep or to maintain an effective cold chain in the mountain rural areas. But by leaving the vaccine outside overnight the vaccine stays well preserved. Even during the day the chances of the vaccine losing its potency is minimal, not least because it is the inactivated type of vaccine that is used. This is of benefit as vaccination is carried out by the extension personnel recruited from the different disciplines of agriculture, people who do not appreciate fully the principle behind a successful immunisation programme. Therefore there is an increased risk of improper handling of the vaccine which could render it ineffective. It should be noted that in the lowlands vaccine is stored as much as possible according to the manufacturers' recommendations.

The country's terrain warrants the creation of numerous vaccination points, failure to observe this fact would lead to failure of the vaccination campaign. Hence there is a need to train a large number of vaccinators.

All dogs in Lesotho are classified as family dogs. They could be restricted or semi-restricted. There is an advantage and a disadvantage to this. The advantage is that it makes vaccination easy and if all dogs are brought for vaccination a good coverage is guaranteed. In the event that they are not, it is impossible and dangerous to enforce other control measures such as dog destruction because of lack of political will.

Since 1988, when a free vaccination campaign was first launched with the help of the government of the Republic of South Africa, the incidence of rabies has been on the decline and to date it has been reduced to nil, the last laboratory confirmed case having been reported in a dog in 1991. No studies have been carried out to ascertain the reasons behind the decline in incidence of rabies but it is believed that the low dog and wildlife population densities plus enforcement of strict vaccination requirements on importation of dogs play an important role.

Subsequent to one reported human death in 1989 no other human case has been recorded to date. Ministry of Health statistics show figures of outpatients given post-exposure anti-rabies treatment as follows: 1992 - 48 patients, 1993 - 15 patients and 1994 - 10 patients. The people are treated on claims of having been bitten by dogs irrespective of whether the dog is rabid or not. Therefore these figures are no reflection of the rabies status in the country.

The vaccination coverage of dogs has been declining since 1992 (see Table 1). The reasons for this are multifactorial and include poor response of the public to the campaign, delays in obtaining the vaccine and inadequate or lack of transport for the preparation of the campaign and to convey the vaccinators to the vaccination points.

Outside of the campaign, dogs can be vaccinated all the year round at owners' request, although at their cost. As a result the total number of dogs vaccinated outside of the campaign period is very low compared to the figures attained during national rabies campaigns which are normally launched annually between June and September.

Since the incidence of rabies may suddenly increase due to the large number of susceptible dogs, our plan of action should be to maintain vigorous rabies awareness campaigns. The public has to be educated on issues of vaccine safety. This will help to remove the mistrust and lack of faith in the rabies campaign, which are attitudes held particularly by rural people.

Table 1. Dog rabies vaccination figures and estimated percent vaccination coverage for the period 1992 to 1994.

Year	Number of dogs vaccinated	Estimated percent coverage
1992	85,890	43
1993	60,885	30
1994	49.625	24

RABIES IN SOUTH AFRICA

G.K Brückner

Department of Animal Health, Pretoria South Africa.

Introduction

The control of rabies, more than most other zoonotic diseases, is influenced by a variety of factors from the external environment - especially the socio-economic environment. The nature and epidemiology of the disease cause sporadic moments of increased public awareness that quickly subsides in the presence of other more important factors affecting the socio-economic well being of people. Thus the creation of a continuous awareness of the disease for human beings demands an intensive and costly input by public authorities. Yet the results are often disappointing for those involved in the control of the disease if the outcome of their extension efforts is measured against for example, the number of dogs vaccinated.

In South Africa the control measures in those areas where sylvatic rabies are most prominent are aimed to maintain the prevalence of the disease within epidemiologically acceptable levels. In these areas it is accepted that the disease manifests itself in seasonal and cyclic patterns and that control measures to prevent the further spread of sporadic outbreaks in urban areas are at most only a public relations exercise with very little impact on the epidemiology of the disease. However, the increase in the occurrence of the disease in canine rabies areas Eke Natal, remains a major reason for concern. Very often the traditional means of control (mass vaccination campaigns or mobile vaccination clinics) are not sufficient to achieve satisfactory control. More radical methods of creating public awareness and to satisfy the needs of a wide spectrum of different communities, must then be implemented. Cognisance must be taken of basic needs of the community, for example, the means of transport available to dog owners, the distance between dwellings and vaccination points, the cultural beliefs and disbeliefs of the community and the ability of decision makers in such communities to influence the people to present dogs for vaccination.

The principles of control of rabies in South Africa.

The control measures practised in South Africa support the following principles for control recommended by the Expert Committee on Rabies of the World Health Organisation.

- Registration, licensing and taxation of dogs;
- Elimination of stray animals;
- Restraint of dogs while the control campaign is under way;
- Mass vaccination of dogs free of charge.,
- Provision of adequate facilities for diagnosis;
- Reduction in number of wildlife animals where these are a reservoir of disease,
- A continual and energetic publicity campaign.

The control of rabies in South Africa is based on three principles:

Legislation: The creation of rabies control areas, import and movement control and compulsory vaccination in certain areas. Rabies is a controlled animal disease in accordance with the Animal Diseases Act, 1984 (Act 3 5 of 1984). The act makes provision for compulsory notification of any outbreaks or suspected outbreaks of the disease, the procedure for submission of samples, the declaration of controlled rabies areas, quarantine or destruction of infected animals, compulsory vaccination, movement control and import control.

Availability of diagnostic services: The service must be reliable, easy to access by the public and must have a speedy notification of results.

Disease surveillance: The surveillance system must be reliable, the planning and execution of surveillance exercises must be based on sound epidemiological principles and the surveillance system must be supported by a reliable diagnostic service.

The primary aims of these control measures are:

To create an immune buffer between animal and man.

To immunise enough of the susceptible canine population to lower the possibility of disease occurrence below an acceptable threshold level. In urban areas it could be as high as 70 to 80 percent while in certain rural areas a vaccination level of 40 percent of the canine population can still succeed in suppressing epidemics of the disease.

The prevention of introduction of the disease from other infected areas or from infected countries by means of movement and import control.

The prevention of the spread of the disease from infected areas to non-infected areas.

The degree of control varies between endemic areas (for example, in the central plateau where rabies is primarily transmitted by viverridae) and the traditional canine rabies areas or epidemic rabies areas (in Natal and the Northern Transvaal). In the latter, stricter control measures are applied in respect of frequency of vaccination and movement control. Those areas where the dogs and jackals play the major role in the transmission of rabies, namely Natal and certain districts in the Northern Transvaal, have been declared rabies controlled areas.

Dogs in such an areas must be vaccinated with an inactivated rabies vaccine between 3 and 7 months of age, again at the age of 1 year and after that at least once every three years. In high risk areas such as Natal, vaccination of all dogs annually is practised as a routine control measure.

Vaccination of dogs and especially cats, outside the rabies control areas, breaks the chain between sylvatic rabies and human infection and therefore safeguards public health, although it does not control the spread of the disease in wildlife.

Table 1. Laboratory confirmed animal rabies cases in South Africa, given by the laboratory of confirmation. OVI = Onderstepoort Veterinary Institute, ALL = Allerton Regional Veterinary Laboratory.

		1993/94			1994/95	
SPECIES	OVI	ALL	TOTAL	OVI	ALL	TOTAL
Bovine	85	16	101	72	23	95
Canine	40	238	278	61	384	445
Feline	12	4	16	7	10	17
Equine	1	2	3	1	3	7
Caprine	4	4	8	1	6	7
Ovine	1	2	3	3		3
Porcine		2	2			
Unspec mongooses	5		5			
Cynictis	69		69	67		67
Atilax				4		4
Civettictis				1		1
Suricata	10		10	11		11
Galerella	3		3	4		4
Mungos	2		2			
Mellivora	1		1	1		1
Felis lybica				5		5
Genetta	1	1	2	2		2
Unspec *Felis*	1		1			
Canis mesomelas	26		26	22		22
Otocyon	15		15	36		36
Unspec. *Canis*	7		7			
Proteles	6		6	6		6
Xerus	4		4	1		1
Ictonyx				1		1
Procavia				1		1
Vulpes chama				1		1
Total	293	269	562	308	426	734

Factors that necessitate a re-evaluation of existing control measures in South Africa.

The sharp increase in the number of cases diagnosed in South Africa during 1994/95 can be almost entirely attributed to the higher number of rabid dogs diagnosed in KwaZulu/Natal. During 1994/95 the total number of positive cases diagnosed was 734 compared to 562 the previous year (Table 1) with dogs, the yellow mongoose (*Cynictis penicillata)* and the black-backed jackal (*Canis mesomelas*) being the three most important vectors. However, the number of dogs diagnosed positive by the Allerton Regional Veterinary Laboratory, in Kwazulu/Natal increased from 238 to 384. This figure is significant in respect of the escalation of positive cases in KwaZulu/Natal as only 63 positive cases in dogs were diagnosed by the Onderstepoort Veterinary Institute which represents the occurrence of the disease in dogs in the remainder of the country.

The canid strain of the disease was diagnosed in areas previously believed to be the "traditional" viverrid-areas. Available data indicates a southward and westward spread of the disease, while the detection of rabid dogs in and adjacent to highly densely populated areas in Gauteng and the Eastern Cape and along the Swaziland and Botswana borders, are major reasons for concern.

In the north-western areas of the country there appears to be a close correlation between outbreaks of the disease in cattle and cases in the black-backed jackal (Figure 1). In these areas the positive cases diagnosed in dogs appear to be a spill-over effect from the jackal cycle rather than primary foci of canine rabies. However, in the northern areas of the Eastern Cape (former Transkei) there appears to be a well-established canine-bovine cycle contrary to the jackal-bovine cycle in the north-western areas of the country.

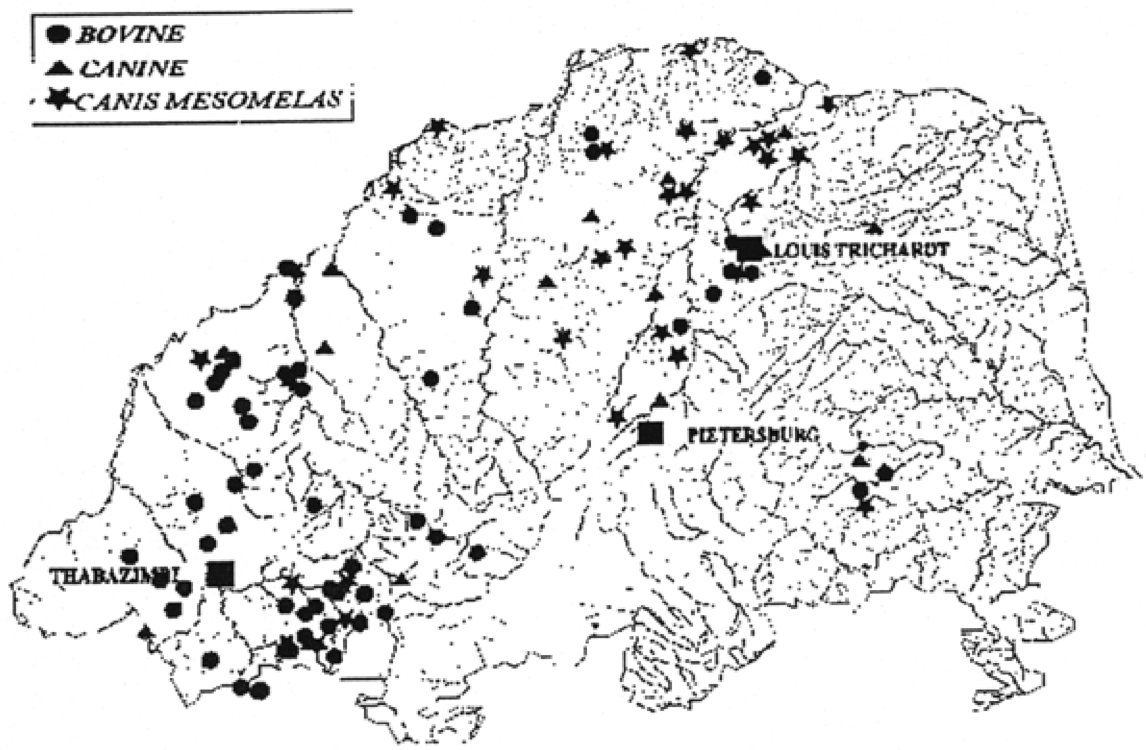


Figure 1. Rabies in the Northern Province of South Africa, January 1992 to October 1995.

The change in the epidemiology of the disease, drastic changes in socio-economic patterns and movement of people and the unprecedented increase in urban and semi-urban settlements, question the rationale of persisting with ongoing strict control measures only in the declared control areas of KwaZulu/Natal and the Northern Province. Available data indicates that similar stricter control measures are needed in other areas of the country. The southward spread of the disease has, for instance, already placed the traditional rabies-free areas of the Western Cape at risk while the need for a disease-free buffer zone or control area around the highly densely populated areas of Gauteng and the Eastern Cape, is inevitable.

Although the control of rabies in the provinces of South Africa is essentially the responsibility of the Veterinary authorities within each province, a national policy to guide the control across borders is essential. The establishment of separate legislation for animal disease control for each province should in the end be guided by norms and standards in the legislation of the National authority, to ensure uniform standards and procedures of control for a disease that respects no political boundaries.

PREVENTION AND MANAGEMENT OF HUMAN RABIES IN ZIMBABWE

Aad van Geldermalsen

Ministry of Health, Bindura, Zimbabwe

Problems in the Prevention and Management of Human Rabies

Although human rabies is endemic and occurs widely throughout Zimbabwe, its incidence is low (Foggin 1988; Bingham 1993; see also "Rabies in Zimbabwe", page 53). Under-reporting is probably not very significant. During the last epidemic some obvious cases were not confirmed in the laboratory but they were recorded and are included in the reports.

The presence of cases represents failure of the last link in the chain of preventive measures (i.e. adequate post-exposure prophylaxis).

Failure of the final preventive measure against human rabies depends partly on endemicity. The reasons for failure with low animal endemicity are lack of awareness and insufficient preparedness. Additional reasons for failure with high animal endemicity (epidemic) are: a higher chance of a missed case, "vaccine out of stock" situations and a higher chance of odd cases.

At present an outbreak of jackal rabies has reached its peak in the district (Bindura) bordering Harare to the north. The rabies 'wave' progressed over the province (Mashonaland Central) from West to East. Figure 1 shows the number of all confirmed cases in the whole of the Province. By February 1995, 20 cases of rabies were confirmed in one month for Bindura District alone, while the whole province had 35 cases. Figure 2 shows the rise to 62 exposed humans in the Bindura hospital catchment in the month of February.

By this date, three (and possibly 4) humans in the province were known to have died of rabies.

Prevention and Management of Human Rabies

The conditions that help prevent human rabies are:

- Public awareness (knowledge, understanding of seriousness, suspicion) which depends on education, special campaigns, personal experience.
- The health system: awareness and surveillance of true rabies contacts, organisation logistics (transport, communication), case of access to facilities, follow-up of patients, feedback from the veterinary diagnostic laboratory.

The system as it should function is described as follows:
In the "quiet" endemic period:

- a level of awareness should be maintained;
- regular updates of animal rabies in the country (from Veterinary Research Laboratory, Harare) should be obtained
- a stock of vaccines at the first referral level (district hospitals) should be maintained.

In the epidemic periods:

- rabies exposure should be prevented;
- dog vaccination (if necessary, tie up orders and shooting) should be maintained;
- efforts should be made to eradicate jackals;
- health education campaigns for farm workers, communal farmers, etc, should be carried out;
- vaccine stock management should be improved.

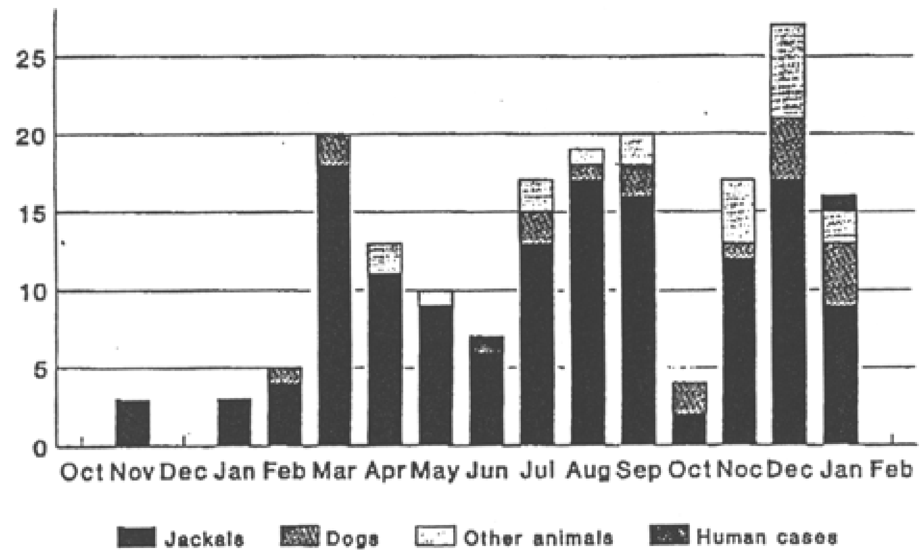


Figure 1. Rabies cases, Mashonaland Central, October 1993 to January 1995. (Source: Provincial Veterinary Office.)

For the prevention of clinical rabies:

- education and information for Health Workers (leaflets, vaccination protocols, lectures, etc) is necessary;
- vaccination registers at hospitals should be instituted;
- regular inspection of registers is essential, with correction of bad practices, reporting to the Veterinary Department and Ministry of Health Headquarters;
- study of system failures should be undertaken in order to sensitise health workers and improve awareness, referral systems etc.

The following are some of the system failures: patient delay

- the patients do not come (or are not able to come) for vaccination;

- health system's delay - including delayed transferral, the urgency is not imparted to the patient,
- combined case - for example in older bite wounds, no urge is felt by the patient or nurse for vaccination.

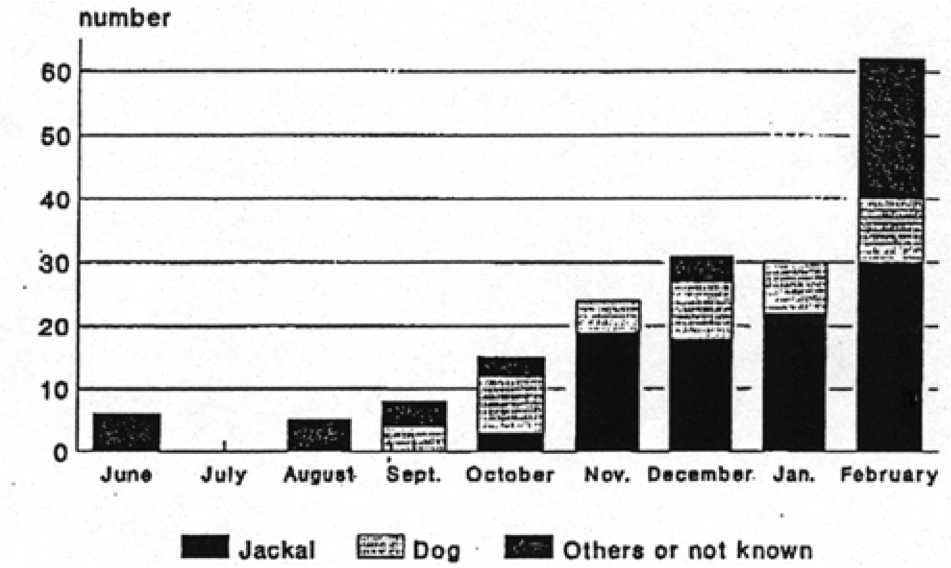


Figure 2. Post-exposure rabies immunisation courses given at Bindura Hospital, 1994 -1995. (Source: Rabies register at Bindura Hospital.)

Conclusion

Even with a high incidence of animal rabies, there need be no case of human rabies provided:

- awareness by the population and health staff is high;
- vaccine stocks are well maintained;
- communication between health and veterinary services is easy and open at all levels of operation.

References

Bingham J. (1993) Rabies in Zimbabwe. In: *Proceedings of the International Conference on Epidemiology, Control and Prevention of Rabies in Eastern and Southern Africa. Lusaka, Zambia, 2 - 5 June 1992.* Edited by King A.A. Editions Foundation Marcel Mérieux, Lyon, France.

Foggin C.M. (1988) Rabies and rabies related viruses in Zimbabwe: historical, virological and ecological aspects. PhD Thesis, University of Zimbabwe, Harare.

ANTEMORTEM DIAGNOSIS OF RABIES IN HUMANS

Makonnen Fekadu

Centers for Disease Control, Atlanta, Georgia, USA

Rabies diagnosis consists of either postmortem or antemortem procedures, or both. The first attempt of postmortem diagnosis of rabies, especially in dogs, was made by physical examination of stomach contents for ingested sticks and stones as stigmata of deranged behaviour.

Other techniques for the rabies diagnosis of infected animals evolved from Pasteur's 1881 report that the disease-producing organisms were located in the central nervous system (CNS) of infected animals and that sub-meningeal inoculation of infected CNS material into rabbits caused their death. Pathological changes indicative of neuronal involvement were made in 1892, when Babès described accumulation of embryonal cells around blood vessels and neurones in the brain tissue undergoing chromatolytic degeneration which he called "rabidic tubercules". These lesions now known as Babés' nodules are also found in other viral encephalitides; their presence is not specific for rabies diagnosis.

Antemortem diagnosis of rabies in humans can aid health care personnel in providing the best clinical treatment for the patient and protection for others who may have been exposed to the disease. Antemortem diagnosis of rabies is a difficult procedure. The psychological impact on a patients family and hospital staff should not be underestimated when rabies is considered in the differential diagnosis. To make patient management even more difficult, negative findings on all tests, while certainly reducing the possibility that rabies virus is the cause of the illness, cannot rule out rabies infection.

Methods of antemortem diagnosis are based on clinical manifestation and previous studies of rabies pathogenesis. These studies showed that, after multiplication in the CNS, rabies virus moves centrifugally along the nerves to peripheral organs. Peripheral nerves, cornea, salivary glands, skin and other tissues near the CNS may be infected early in the disease.

The detection of viral antigen in immuno-fluorescent antibody stained impressions of corneal epithelium and frozen sections of skin biopsy or the isolation of a virus from the saliva and tracheal aspirates are reliable indicators of rabies infection.

Demonstration of a significant serum antibody titre to rabies virus in the absence of passive or active immunisation or the appearance of antibody in the cerebrospinal fluid is a definitive method for diagnosis. The antibody response to infection, usually found on day 8 to 10 after onset of the first symptom, may be absent even 24 days after onset on rare occasions.

Recent techniques, such as the detection of messenger and/or genomic RNA by in situ-hybridisation and polymerase chain reaction are good candidates for early detection of rabies infection. These techniques are still in the experimental stage, and when available may need well equipped laboratories and trained personnel to be applicable for routine rabies diagnosis.

Laboratory results from rather few cases of human rabies evaluated by the Rabies Laboratory at the Centers for Disease Control showed that no single ante-mortem diagnostic test was positive in every case.

PRE- AND POST-EXPOSURE TREATMENT FOR RABIES

J. Lang

Pasteur Mérieux Serums et Vaccins, Lyon, France

The risk of rabies infection to people bitten by rabid animals varies from 3 - 70 percent depending on the site of the bite (bites to the head being considerably more likely to result in infection) and the number and depth of the bites. The presence of clothes over the site of the bite may even reduce the chances of infection to below one percent (Hattwick 1974).

Clinical diagnosis is the initial stage in the diagnostic process, and for this it is important to recognise the clinical phases of the disease. These can be extremely variable. The first stage, the incubation period, normally varies from 20 - 60 days. This is followed by the prodromal period of 2 - 10 days and then the acute neurological phase of similar duration. Finally coma sets in, which may be short or up to 14 days.

In rabies endemic areas the disease must be included in the differential diagnosis of all persons who present with signs of neurological involvement.

Table 1. Interval from bite to death in 707 cases in Thailand (Wilde *et* al. 1989).

Time interval	Percent
7-10 days	3.23
10-20 days	41.93
21-28 days	25.80
1-3 months	16.13
3-6 months	3.23
6-12 months	3.23
Over 1 year	6.45

Post-exposure immunisation

The incubation period in humans can be short. It was less than one month in 71 percent and less than 3 months in 87 percent of cases in Thailand. It is therefore vital that post-exposure treatment is not delayed. Treatment must not be delayed while observing an animal. Double or triple doses of vaccine can be given if the treatment is delayed more than 48 hours.

The WHO (1992) have recommendations for categorising rabid animal contacts according to the treatment which is necessary. These are given in Table 2. In post-exposure treatment five injections should be given on days 0, 3, 7, 14 and 30.

In Category 3 contacts, rabies immunoglobulin should also be given on day 0. This should be given at a dose rate of 20 IU/kg body weight for human immunoglobulin or 40 IU/kg body weight for equine immunoglobulin. As much as possible should be infiltrated around the wound and the remainder should be administered intramuscularly into the gluteal region. It has been shown that the local wound infiltration is extremely effective in rapid neutralisation of the virus. The use of immunoglobulins may, however, result in lower antibody titres in the short (14 days) and long term (1 year) (Suntharasamai *et al.* 1986).

Table 2. WHO guide for post-exposure treatment (WHO 1992).

Category	Type of wound	Treatment
1	Touching or feeding of animals. Licks on intact skin	No treatment
2	Nibbling of uncovered skin. Minor scratches or abrasions without bleeding. Licks on broken skin.	Start vaccination immediately. Stop treatment if animal remains healthy after 10 days or if it is negative.
3	Single or multiple transdermal bites or scratches. Contamination of mucous membranes with saliva.	Vaccine and immunoglobulins immediately Stop treatment if animal remains healthy after 10 days or if it is confirmed negative.

In fully vaccinated people who completed the vaccine course over 6 months but less than 1 year previously, 2 booster doses should be given on days 0 and 3. A complete treatment should be given if 1 year has lapsed since the full course.

Immunocompromised patients should receive two or three doses intramuscularly at day 0. The other vaccinations and the immunoglobulin should be administered as for normal patients.

There are several factors which may influence the effectiveness of post-exposure treatment:

1. Inappropriate local wound treatment;
2. The nature and severity of exposure;
3. The treatment (or lack of) with immunoglobulin particularly in cases of severe exposure, and the site of administration of the immunoglobulin;
4. Delays in vaccination;
5. The vaccine potency, which is affected by the initial potency and storage conditions;

6. Host factors such an under-responsive immune system, underlying diseases, alcoholism and concurrent treatment with immunosuppressants such as steroids and chloroquine;
7. Iatrogenic factors such as injection of vaccine in the gluteal area.

Pre-exposure immunisation

Pre-exposure immunisation should be given to people at risk of exposure, such as laboratory. staff veterinarians and animal handlers. The WHO (1992) recommend that the pre-exposure course consist of vaccinations on days 0, 7 and 28, followed by boosters at 1 year and every 3 years thereafter.

The vaccine should be administered intramuscularly in the deltoid area of the arm in adults and the anterolateral area of the thigh in infants. It should never be administered in the gluteal area.

Verorab human rabies vaccine

For the production of Verorab vaccine, the PM 1503-m rabies virus seed strain is used. It is grown on vero cells at the 137th passage level.

In post-exposure trials using Verorab, patients had mean antibody titres over 0.5 IUmI at 14 days and thereafter for over 1 year (Thongcharoen *et al.* 1986).

In a pre-exposure trial using Verorab, titres of over 0.5 IU/ml, which persisted for over 1 year, were achieved in all subjects at day 14.

The incidence of adverse reactions to the vaccine is about 1 percent and these range from local reactions at the site of injection to systemic reactions with fever, headache, fatigue and myalgia.

Challenges for research on human rabies vaccine production

There are challenges to the production of an ideal commercial rabies vaccine. These include:

1 . Reducing the production cost;
2. Producing a vaccine which requires fewer injections;
3. Producing an orally effective vaccine;
4. Broadening the protective activity to include rabies-related viruses;
5. Producing a vaccine which will give lifelong immunity.

References

Hattwick MAW. (1974) Human rabies. Public *Health Reviews* **3**:229-274.

Suntharasamai P., Chanthavanich P., Warrell M.1, Looareesuwan S., Karbwang 1, Supanaranond W., Phillips R.E., Jansawan W., Xtieref C., Pouradier-Duteil X. and Warrell D.A. (1986) Purified vero cell rabies vaccine and human diploid cell strain vaccine: comparison of neutralising antibody responses to post-exposure regimens. *Journal of Hygiene, Cambridge* 96:483~489.

Thongcharoen P. et al. (1986) *Virus Information Exchange Newsletter of South East Asia* 3:80.

WHO (1992) MMO Expert Committee on Rabies, Eighth Report. WHO Technical Report Series 824. World Health Organization, Geneva.

Wilde H., Choonikasien R, Hemachudha T. Supich C. and Chutivongse S. (1989) Failure of rabies postexposure treatment in Thailand. *Vaccine* 7:49-52.

INTRADERMAL APPLICATION OF MODERN (CELL-CULTURE AND EMBRYONATED EGG) RABIES VACCINES FOR HUMAN POST-EXPOSURE TREATMENT[1]

F-X. Meslin

Veterinary Public Health Unit, World Health Organization, Geneva

Introduction

In developing countries, where more than 99 percent of all human rabies deaths occur, nervous tissue antirabies vaccines are still the most widely used because of their relatively low cost and despite their variable potency and the risk of neurological complications. The supplies of modem and safe vaccines for many developing countries are grossly inadequate, whereas the demand for affordable and safe human post-exposure treatment (PET) is increasing in the developing world. Although the costs of modem vaccines are decreasing, the current price of a full intramuscular vaccine treatment is far beyond what an average family in Africa or Asia can afford.

Multi-site intradermal (ID) administration of small doses of cell culture rabies vaccine which have been shown to protect humans bitten by proven rabid animals and to reduce the costs of PET by 60 percent is an effective way of decreasing the cost of these much more potent, safe modern vaccines and of increasing the neutralising antibody response. In many developing countries, however, rabies vaccines are being given intradermally under inappropriate conditions and according to regimens whose efficacy is unproven.

A VMO Expert Committee in 1991 recommended intradermal application of modem rabies vaccines. This recommendation was re-assessed in January 1992. As new data had accrued since early 1993 it was time to re-evaluate the safety and efficacy of a method which should help to reduce the number of human rabies deaths. A VMO consultation was therefore held on 13-14 March 1995.

Recognised ID regimens:

An "ideal" vaccine regimen for PET should require a minimum quantity of vaccine, few visits to the clinic and rapid induction of immunity. These features need to be combined to produce an economical, efficient, safe regimen.

The Thai Red Cross (TRC) 222011 and the 804011 regimens have fulfilled these requirements. They have been used in restricted areas, mainly by experienced personnel.

[1] This paper is a summary of a paper presented at the Fifth Conference on Research Towards Rabies Prevention in the Americas, Mexico, 24-27 October 1995. It is reproduced here in response to the discussions held during the human rabies session.

From 1985 to 1994, however, approximately 70 000 TRC ID regimens were given in Thailand, with more than 29 000 in category 3 exposure.

- The Thai Red Cross 2-site intradermal method (222011), (for use with purified verocell rabies vaccine (PVRV), purified chick embryo cell vaccine (PCEC) and purified duck embryo vaccine (PDEV)) consists of injecting one ID dose at each of 2 sites on day 0, 3, 7 and a single site on day 28 and 90; the ID dose per site is one fifth of the volume after reconstitution of a single immunising intramuscular (IM) dose, i.e. if IM dose is 0.5 ml, ID dose = 0.1 ml/site; if IM dose is 1.0 ml, ID dose = 0.2 ml/Site.

- The 8-site 804011 regimen (for use with human diploid cell vaccine (HDCV) and PCEC vaccines where IM dose is 1ml) consists of injecting on day 0, 1 ml of vaccine divided between 8 ID sites (deltoids, anterior thighs, suprascapular and lower quadrant of the abdomen); on day 7, 4 x 0.1 ml ID injections (deltoids and thighs), and single 0. 1 ml ID boosters on days 28 and 90.

Proposed WHO guidelines

Knowledge of the correct methods of using modem tissue culture and embryonated egg vaccines intradermally is very poor in tropical areas with endemic dog rabies, where exposure to rabid animals is frequent vaccine is scarce, there is little money and no rabies immunoglobulin (RIG) available. For these reasons, a variety of untested, potentially dangerous ID regimens are being used in some places. The only regimens which have been demonstrated to be immunogenic today are the 222011 and the 804011 (see above).

The volume of the standard IM dose varies between different products, and some producers are in the process of changing from a 1 ml to a 0.5 ml ampoule. Attention should therefore be paid to the volume of the product after reconstitution.

It is therefore proposed to publish precise instructions on the currently recognised methods of y ED vaccination, to make optimum use of restricted resources, and on precautions to be taken to prevent vaccine contamination from multidose vials, as well as viral cross-infection.

These guidelines should be suitable for distribution to large and small clinics and rural health centres and to government organisations deciding on vaccine policies.

A draft outline of headings for the proposed Guidelines was presented during the consultation.

Future research:

There is a need for new ID vaccine regimens using products formulated in 0.5 ml vials. Immunogenicity studies in non-exposed volunteers must identify a satisfactory regimen before efficacy is tested in patients needing PET.

- The immunogenicity of a regimen beginning with 4-site ID injections (0.1 ml/site), e.g. 40202 (days 0-7-28) using the entire 0.5 ml vial on day 0 should be studied.
- The effect of smaller doses (0.05 ml/site) in 8 sites, still using one 0.5 ml vial on day 0, could be investigated. The 8-site ID PET regimen has only been tested with HDCV and PCEC in 1 ml vials.
- The value of giving a final injection on day 90 should be tested.
- Clinical trials of the efficacy of the optimum regimen emerging from prior immunogenicity studies, with and without RIG, in patients with proven exposure to rabies should be carried out. Studies without RIG could be conducted ethically only in countries where RIG is unavailable.

Conclusions

Implementing ID methods for PET would increase the use of PET globally. It was emphasised that these regimens are of comparable immunogenicity to the IM regimen and very much more effective and safer than vaccines of nervous tissue origin.

Although the original IM route of inoculation has been regarded as the optimum method of treatment, it may be considered that this is not the best way of using one dose of vaccine on day 0. The ID method has advantages especially in developing countries in the absence of RIG. There is no contraindication to the use of an ID regimen.

The decision to implement economical ID PET rests with government agencies which select policies for rabies prophylaxis in their own countries. Dissemination of information from such an authority by instruction of physicians, nurses and other health care workers is very important. Local or regional advisers, who could be contacted easily to give practical advice, would enhance the acceptance of the new methods.

Further reading

Chutivongse S., Wilde H., Supich C. *et al.* (1990) Post-exposure prophylaxis for rabies with antiserum and intradermal vaccination. *Lancet* 335: 896-8.

Cupo P., Azevedo-Marques M.M., de Menezes JR, Hering S.E. (1991) Reacoes de hipersensibilidade imediatas apos uso intravenoso de soros antivenos: valor prognostico dos testes de sensibilidade intradermicos. *Revista do Instituto de Medicina Tropical de Sao Paulo* 33: 115-122.

Khawplod P., Gluck R., Wilde H. *et al.* (1995) Immunogenicity of purified duck embryo rabies vaccine (Lyssavac-N) with use of the WHO-approved intradermal postexposure regimen. *Clinical Infectious Diseases* 20: 646-5 1.

Malasit P., Warrell DA, Chantavanich P. *et al.* (1986) Prediction, prevention, and mechanism of early (anaphylactic) antivenom reactions in victims of snake bites. *British Medical Journal 292: 17-20.*

Phanuphak P., Khawplod P., Sirivichayakul S, *et al. (1987)* Humoral and cell-mediated immune responses to various economical regimens of purified Verocell rabies vaccine. *Asian Pacific Journal of Allergy Immunology* 5: 3 3 -7.

Warrell MI, Nicholson K.G., Warrell D.A. *et al.* (1985) Economical multiple site intradermal immunisation with human diploid-cell-strain vaccine is effective for post-exposure rabies prophylaxis. *Lancet* 1: 1059-62.

Warrell MI, Suntharasamai P., Nicholson K.G. *et al,* (1984) Multi-site intradermal and multi-site subcutaneous rabies vaccination: improved economical regimens. *Lancet L* 874-6.

WHO Expert Committee on Rabies, 8th report, 1992. TRS 824, World Health Organization, Geneva.

WHO (1993) Report of informal discussions on ID application of modern rabies vaccines for PET, 22 January 1993 (WHO/RabRes./93.41).

WHO (1994) World Survey of Rabies No 28, for year 1992 (WHO/Rabies/94.210).

A SURVEY OF COMMUNAL LAND DOGS IN ZIMBABWE WITH REFERENCE TO IMPROVING RABIES VACCINATION COVERAGE

James RA. Butler

Department of Biological Sciences, University of Zimbabwe

Abstract

The majority of Zimbabwe's dog population exists in communal lands; it is here that the country's canine rabies is probably most prevalent. Results are presented of a survey of the communal land dog population. Dogs are typically unrestricted in all agro-ecological and ethnic regions, but despite this most are accessible for vaccination. Dog densities are closely linked to human densities, probably due to their dependence on human-derived food. The dog:human ratio of 1:4.7 and density of 20.92 per km^2 suggest high rates of dog-human contact and risk of rabies transmission. The largest age class was that less than one year of age, and 88.4 percent of unvaccinated dogs were in this group. Overall, 58.4 percent of dogs had been vaccinated. Methods of improving this coverage, including the feasibility of oral vaccination, are discussed.

Introduction

Domestic dogs (*Canis familiaris*) have accounted for almost half of the confirmed rabies cases in Zimbabwe since 1950 (Department of Veterinary Services (DVS) records). Brooks (1990) estimated in a national survey of the Zimbabwe dog population in 1986 that only 40 percent of dogs of three months old or above had been vaccinated for rabies, which falls far below the 70 percent minimum coverage recommended by the World Health Organization (WHO 1984) to adequately control canine rabies.

Dog rabies is probably most prevalent in Zimbabwe's communal lands (subsistence agricultural areas). Brooks (1990) estimated that 71.3 percent of Zimbabwe's dogs exist in communal lands, and 52 percent of all dog rabies cases since 1980 have occurred in these areas (DVS records). Furthermore, Foggin (1988) noted that 51.6 percent of rabid communal land dogs bit humans (compared to 14.5 percent of rabid dogs for other landuse types), and that the incidence of rabies in these areas is certainly under-reported. Since 56 percent of Zimbabwe's human population lives in communal lands (Central Statistics Office 1995), and the principal justification for the control of rabies world-wide is to prevent the death of humans (Brooks 1990, WHO/WSPA 1990, Fekadu 1993, Wandeler *et al.* 1993), it is obvious that this landuse sector should be a priority for DVS rabies control programmes.

Brooks (1990) concluded that the rabies vaccination coverage of dogs in Zimbabwe had to be improved, perhaps through the development of new vaccination techniques. The current DVS method in communal lands is the compulsory annual parenteral vaccination of dogs older than three months (Foggin 1988), which are brought for vaccination by their owners to meeting points such as dip tanks or schools. However, oral vaccination by bait is a method which has been used for wild carnivores in Europe and North America (Perry and Wandeler 1993), and the feasibility of this method for dogs in rural Mrica has not yet been fully investigated (Perry *et al.* 1988). It is possible that oral vaccination techniques could be a suitable alternative to parenteral vaccination of dogs in Zimbabwe's communal lands.

It is recognised worldwide that prior to improving or modifying existing methods of canine rabies control the ecology of the targeted dog population and its parent human population should be well understood. Consequently a study of communal land dog ecology has been initiated with the goal of improving rabies control in these areas. Six questions are addressed by the research:

1. What is the relationship between dogs and humans?
2. What are the dynamics and structure of the dog population?
3. What are the densities of dogs?
4. What are peoples' attitudes to current rabies vaccination methods and any proposed alterations, and how aware are they of rabies?
5. What is the feeding and social ecology of communal land dogs, and what are the determinants of their home ranges?
6. What is the ecological relationship between communal land dogs and wild carnivores?

This report presents the preliminary results of a questionnaire survey carried out in 1994 as the initial stage of the study. The survey aimed to answer questions (1) to (4) and, in part, question (5). Questions (5) and (6) are being addressed in 1995-96 by a detailed study of the feeding and social ecology of dogs on the boundary of Gokwe communal land and a wildlife reserve, the Sengwa Wildlife Research Area.

Methods

The questionnaire design followed the guidelines of the World Health Organisation (WHO/WSPA. 1990). Occupants of houses with dogs were asked about the number of dogs owned and their age, sex and rabies vaccination status, the history of dog deaths in the past two years, their reasons for keeping dogs, their dog-feeding and dog-restriction policies, and the presence of foreign dogs (owned or stray). Dog owners were also asked about their opinion of the current parenteral rabies vaccination method; then the oral vaccination technique was described to them, and their opinion of this alternative method was noted. Occupants of houses without dogs were also asked about the presence of other dogs (owned or stray) at their house. All households were asked about the number of people living in the premises, their waste disposal method, whether there was a toilet on the premises, and if they knew about rabies.

Sampling of households was undertaken by following roads within each communal land that were passable by vehicle. Every alternate house within a 500 m distance perpendicular to the road was sampled. Wherever possible only adults were interviewed. If the occupants of a house were absent or only children were present the adjacent household was interviewed instead. Local DVS Veterinary Extension Assistants (VEAs) and staff conducted the interviews and the author acted as recorder. Although not strictly random in structure this sampling method was random in effect as there was no prior knowledge or planning of the routes that were ultimately driven.

Five agro-ecological regions have been defined in Zimbabwe by Vincent & Thomas (1960), within which agricultural systems and human population densities vary. These areas range from Region I (high rainfall, specialised and diversified farming) to Region V (low rainfall, extensive livestock production). The following seven communal lands were selected to represent the five agro-ecological regions: Ngorima (Region I), Svosve and Kandeya (Region II), Gokwe (Region III), Tsholotsho and Dande (Region IV) and Metengwe (Region V) (Figure 1). Variations may also occur between ethnic groups; consequently the sample of communal lands was also used to represent the different ethnic areas of Shona (Ngorima, Svosve, Kandeya, Gokwe), Ndebele (Tsholotsho, Gokwe) and Venda (Mtetengwe).

Human population densities in each communal land were derived from areas (km^2) estimated on 1: 1000 000 and 1:250 000 maps and aggregated ward populations taken from the 1992 Zimbabwe National Census (Central Statistics Office 1995). Human populations for each communal land in 1994 were estimated by multiplying the 1992 population by the Census' projected annual population growth rate of 3.13 percent.

Results

A total of 705 households was interviewed (Table 1). A majority of houses owned dogs (62.0 percent), all houses had open pits for waste disposal either inside or outside the perimeter of the house, and more than half (53.3 percent) had no toilet facilities (Table 1).

The sex ratio of male to female dogs was close to parity, indicating no selection by owners for one sex or the other (Table 2). With the exception of Ngorima, which had relatively few dogs, the dogs per capita, dogs per household and dog:people ratios were similar in all communal lands. The dog density results (Table 3) were calculated from the estimated human population and area data for each communal land, using the dogs per capita data (Table 2). There was a significant correlation between human and dog densities in the communal lands surveyed (Figure 2), suggesting that dog densities are dependent on human densities.

The most common primary use of dogs by 576 houses keeping them or who intended to keep them was to "guard the house from human intruders" (60.0 percent). A further 14.9 percent of houses used dogs to "protect crops from raiding baboons and monkeys", and 14.4 percent kept them to "guard poultry from predation by civets, genets and mongooses". More minor uses were to "guard livestock against predation by jackals, hyenas and lions" (5.6 percent); to "give warning of crop raiding elephants, buffalo and wild pigs" (2.2 percent); "as a pet (1.2 percent); "for hunting" (0.9 percent); "for herding cattle" (0.4 percent) and "for breeding pups to sell (0.4 percent). The use of dogs as protection from marauding wildlife was most common in the more remote communal lands with low human population and high wildlife densities, such as Tsholotsho, Dande and Gokwe.

Table 1. General household data collected by the questionnaire survey undertaken in seven communal lands (N=Ngorima, S=Svosve, K=Kandeya, G=Gokwe, T=Tsholotsho, D=Dande, M=Mtetengwe).

COMMUNAL LANDS SURVEYED	**N**	**S**	**K**	**G**	**T**	**D**	**M**	**Total**
Households interviewed	99	110	71	110	115	100	100	705
Mean persons per household	7.34	5.74	6.61	8.46	7.09	6.91	7.91	7.17
± S.E.	0.84	0.61	0.87	0.95	0.72	0.79	0.90	0.31
Range	1-30	1-17	1-14	1-45	2-18	2-29	2-24	1-45
Households with dogs (%)	45.5	64.5	46.5	61.8	72.2	59.0	78.0	62.0
Households with open pit (%)	100	100	100	100	100	100	100	100
Households without toilet (%)	9.1	29.1	54.9	70.1	89.6	69.0	46.0	53.3

Table 2. Dog numbers, sex ratios, and dog:people ratios in the communal lands surveyed.

	COMMUNAL LANDS SURVEYED							
	N	S	K	G	T	D	M	Total
Total dogs surveyed	82	168	87	186	224	156	182	1,085
Total males	46	90	48	102	115	79	125	605
Total females	36	78	39	84	109	77	57	480
Male:Female ratio	1:0.8	1:0.9	1:0.8	1:0.8	1:0.9	1:1	1:0.5	1:0.8
Dogs per capita	0.11	0.27	0.18	0.20	0.27	0.23	0.25	0.22
Dogs per household	0.83	1.53	1.23	1.69	1.95	1.56	1.82	1.54
Dog:people ratio	1:8.9	1:3.8	1:5.4	1:5.0	1:3.5	1:4.4	1:4.3	1:4.7

Dog husbandry varied little in the communal lands surveyed (Table 4). All dog owners fed their dogs *sadza*, (maize meal porridge, the human staple), once or more times a day. Very few dogs were restricted by tying up during the day and night and households that practised this only did so occasionally. Only one dog owner had an enclosure that confined his dogs.
A vast majority of houses (78.6 percent) reported foreign dogs visiting their homes, usually at night. All of the foreign dogs seen by householders were known to have owners. The visiting dogs either scavenged leftover food or were males soliciting a resident bitch on heat. Householders and DVS staff described four cases of stray dogs since 1990, one in each of the communal lands of Ngorima, Kandeya, Tsholotsho and Mtetengwe. These dogs seemed to have been abandoned during the 1992 drought.

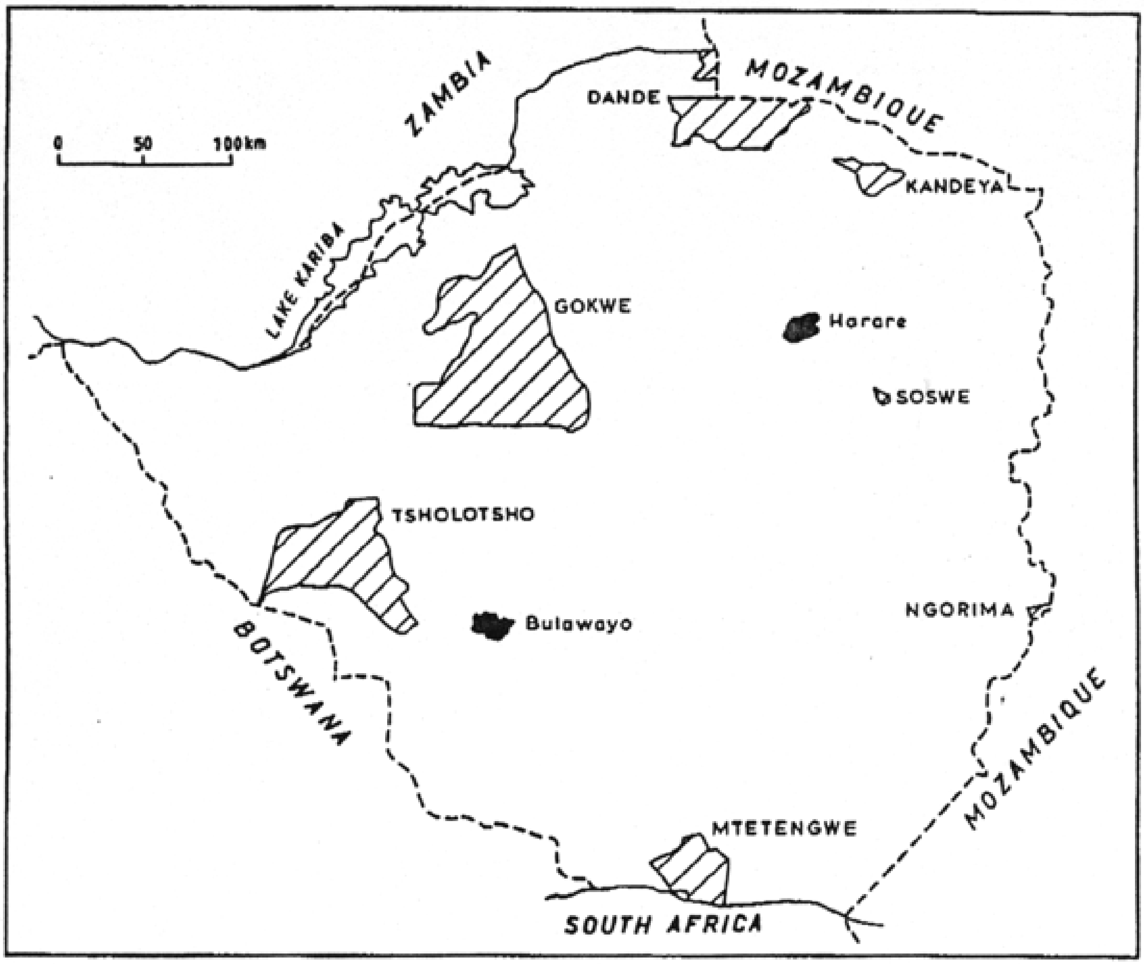


Figure 1. The locations of the seven communal lands surveyed in Zimbabwe.

Table 3. Estimated human and dog numbers and densities calculated from the dogs per capita results (see Table 2) for the communal lands surveyed.

	Communal lands surveyed							
	N	**S**	**K**	**G**	**T**	**D**	**M**	**Total**
Estimated area (km^2)	106	108	334	2344	1834	1052	480	**6258**
Estimated human popn .(1994)	16355	14064	98 866	305 531	118 938	38054	22149	**613957**
Human density (per km^2)	154.29	130.22	296.0	130.35	64.85	36.17	46.14	**98.11**
Dogs per capita	0.11	0.27	0.18	0.20	0.27	0.23	0.25	**0.22**
Estimated dog popn. (1994)	1799	3797	17796	61106	32 113	8752	5537	**130900**
Estimated dog density (per km^2)	16.97	35.16	53.28	26.07	17.51	8.32	11.54	**20.92**

Table 4. Dog husbandry characteristics among the 437 houses with dogs interviewed in the communal lands surveyed.

	COMMUNAL LANDS SURVEYED							
	N	S	K	G	T	D	M	Total
Houses feeding dog sadza (%)	100	100	100	100	100	100	100	100
Houses tying dogs up by day (%)	6.7	8.4	9.1	10.3	2.4	3.3	6.4	6.4
Houses tying dogs up by night (%)	4.4	4.2	3.0	8.8	3.6	5.1	0	4.1
Houses with dog-restricting fence %)	2.2	0	0	0	0	0	0	0.2

The mean age of the 1085 dogs surveyed was 2.01 years (:ES.E. = 0.09, range = 1 month - 16 years). The mean age of all males surveyed was 2.18 years (:ES.E. = 0. 14, range = 1 month -16 years). The mean age of all females surveyed was 1.80 years (:ES.E. = 0.12, range = 1 month - 14 years). There was no significant difference between the mean age of all males and females (p>0.01, t-test). The population age structure showed that 41.8 percent of the dog population at the time of the survey was less than one year old, as shown in Figure 3. From the ages of 1 256 dogs that had died in the past two years at households surveyed it was calculated that 71.8 percent died within the first year of life.

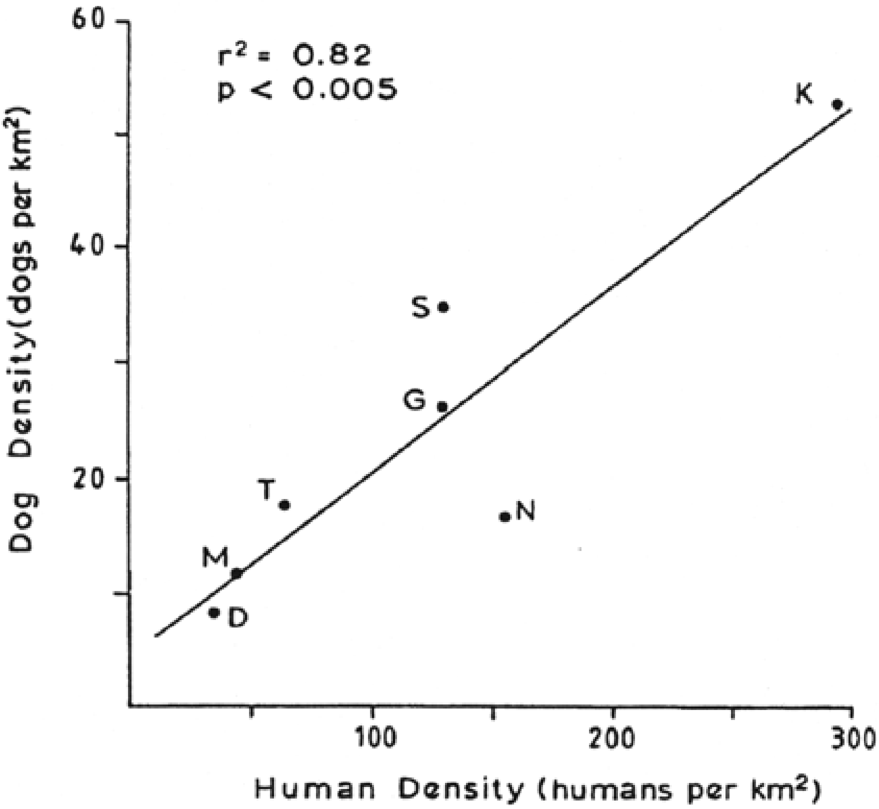


Figure 2. The correlation between human density and dog density in the communal lands surveyed (N=Ngorima, S=Svosve, K=Kandeya, G=Gokwe, T=Tsholotsho, D=Dande, M=Mtetengwe).

Of a total of 884 dogs surveyed more than three months old (and therefore eligible for rabies vaccination) 41.6 percent had never been vaccinated (Table 5), and 58.4 percent had been vaccinated at least once. There was considerable variation in vaccination coverage in the seven communal lands, with unvaccinated dogs ranging from 27.9 percent in Svosve to 56.5 percent in Dande. Interviews with local DVS staff in each communal land suggested that these data were a reflection of the DVS vaccination resources available.

The most common excuse that dog owners gave for a dog not being vaccinated was that it had been born since the last vaccination (38.2 percent); this reason was the most common in all of the communal lands (Table 5). A close second was the lack of information heard about the time and place of the last vaccination campaign (32.2 percent). Very few owners with unvaccinated dogs reasoned that their dogs could not be handled (6.3 percent) and therefore could not be taken to the vaccination point. Of all unvaccinated dogs 88.4 percent were in the three months to one year age group; beyond one year old the dog vaccination coverage per age class remained relatively high and consistent (Figure 4).

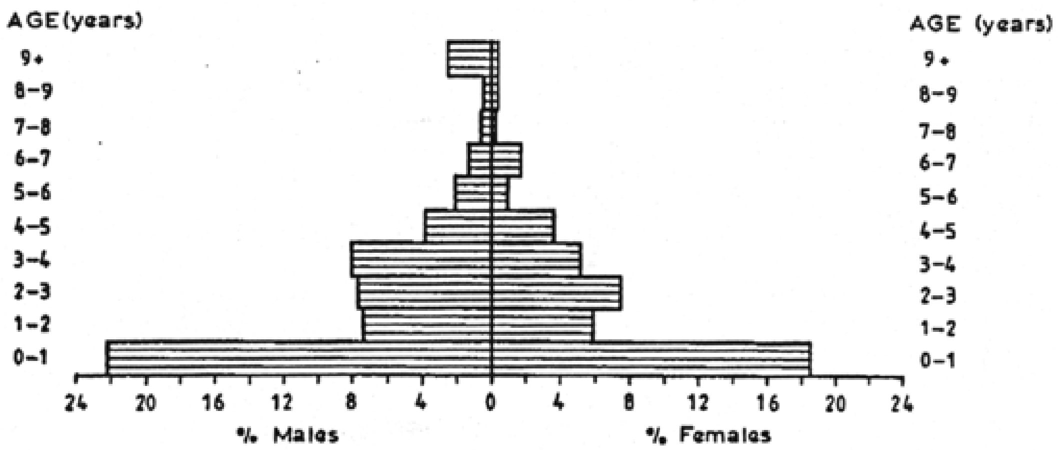


Figure 3. The population pyramid derived from the age distribution of the 1 085 dogs surveyed in the seven communal lands.

Rabies awareness among all households was high, with 70.0 percent being "quite clear" about the disease and its implications (Table 6). Awareness among the 513 houses with dogs or who had had dogs in the past two years was similarly high, and almost all approved of rabies vaccination; the two that did not had had dogs that died immediately after being vaccinated and held the Veterinary Department responsible. Response to the idea of oral rabies vaccination was also positive, with 96.5 percent of houses with dogs or who had had dogs in the past two years supporting the initiative (Table 6). The 18 houses who disagreed either did not understand the method or distrusted changing the traditional vaccination technique.

The main reason that dog-owning households gave for supporting oral vaccination was that "any method will do as long as it works" (Table 7). However, the second most popular reason was that the method would "avoid handling the dog", suggesting that dog owners would rather avoid handling their dogs if possible. The least common reason was that the method would "save walking time and avoid misinformation about vaccination dates and sites".

Table 5. The percentage of dogs more than three months old that had never been vaccinated for rabies, and the reasons given, expressed as a percefitage of unvaccinated dogs.

	COMMUNAL LANDS SURVEYED							
	N	**S**	**K**	**G**	**T**	**D**	**M**	**Total**
Dogs not vaccinated	30.0	27.9	43.7	42.7	49.3	56.5	39.1	41.6
Dog born since last vaccination	70.8	7.9	77.4	30.4	13.5	15.4	84.1	38.2
No information about last vaccination	20.8	31.6	12.9	26.8	64.9	40.0	4.8	32.2
Owner unavailable at the time	4.2	28.9	9.7	16.1	8.1	18.5	3.2	12.5
Vaccination team failed to arrive	0	0	0	25.0	4.0	7.7	6.3	7.4
Dog unhandleable	4.2	28.9	0	1.7	2.7	9.2	1.6	6.3
Dog <3 months old at last vaccination	0	2.7	0	0	6.8	9.2	0	3.4

Discussion

In terms of the World Health Organization's (WHO 1984) dog classification, Zimbabwe's communal land dogs are free-ranging and semi-dependent on humans and should therefore be termed "neighbourhood dogs". They are generally not restricted by their owners (Table 4) and are free to roam, as the evidence of most households reporting foreign dogs visiting their homes confirms. Although all dogs are intentionally fed by their owners (Table 4) the food they receive may not be nutritionally adequate, encouraging dogs to roam in search of food at other houses. Food is freely available since all houses have an open waste dump and half do not have toilets (Table 1), possibly providing dogs with a supply of human faeces. These characteristics of dog husbandry and the resulting dog behaviour were similar in all the communal lands surveyed, regardless of agricultural systems or ethnic regions.

This factor has direct consequences for the dog population structure. Since all dogs have access to other members of the population and their owners do not control breeding, reproduction is unrestricted. This results in the overall parity of the sex ratio and again this

is fairly consistent in every communal land surveyed (Table 2). The population pyramid (Figure 3) further illustrates this parity and also indicates that the majority of the population (41.8 percent) is less than one year old. The mortality rate and turnover for this age class is very high, with 71.8 percent dying per annum.
The close relationship between human density and dog density (Figure 2) and the absence of reports of true "feral" dogs ("no dependency on humans" (WHO 1984)) suggests that communal land dog numbers are governed by the availability of human-derived food and therefore by the numbers of humans. The lack of control of breeding would allow dog populations to reach levels that could be sustained by this food supply. The few reports of "stray" dogs were almost certainly owned dogs that had been lost or abandoned. These dogs should still be termed neighbourhood rather than feral dogs since they probably continue to be dependent on humans for food.

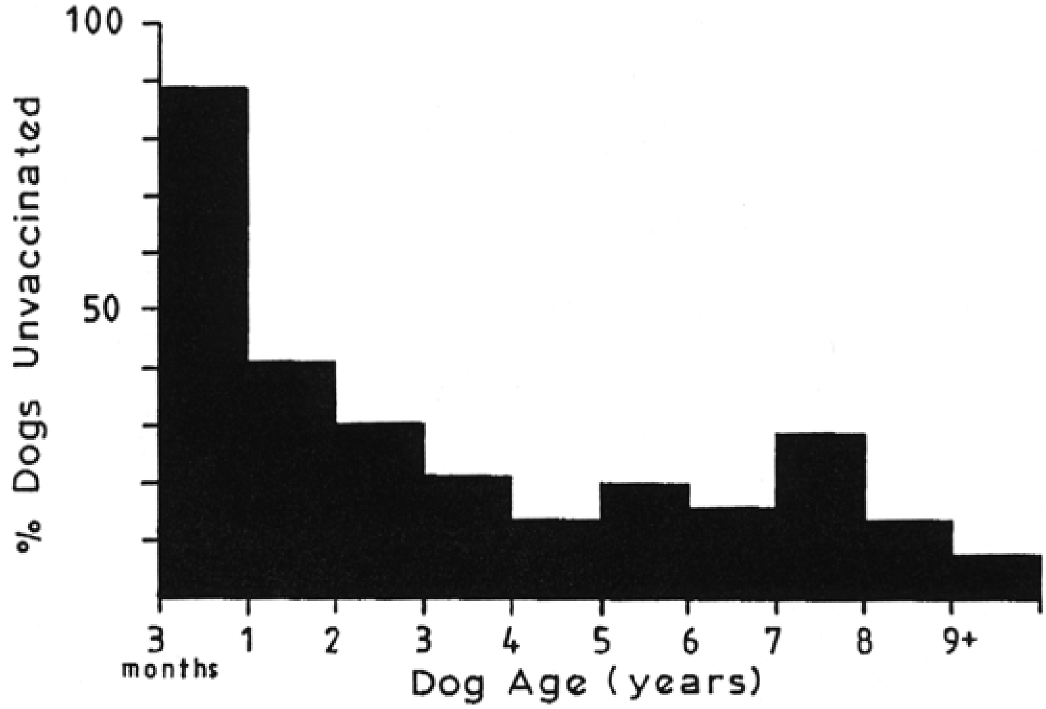


Figure 4. The age-specific distribution of dogs that had never been vaccinated for rabies.

This relationship has important implications for the risk of rabies transmission to humans. Since dog:human ratios are relatively consistent in all communal lands (Table 2) and dogs roam freely around houses which have large numbers of people resident (Table 1) the chance of dog-human contact must be high. This is probably magnified by the high overall dog density of 20.92 dogs per km^2 . The majority of houses (62 percent) owned dogs, yielding a dog:human ratio of 1:4.7. Both of these figures are greater than Brooks' (1990) estimates for the whole of Zimbabwe of 41 percent and 1:65, respectively, suggesting that dog-human contact in communal lands is probably greater than in any other landuse sector of the country.

In terms of rabies vaccination coverage the risk of rabies transmission from dogs to humans may be less in communal lands than in the whole of Zimbabwe. Brooks (1990) estimated that 40 percent of dogs three months old and above in Zimbabwe had been vaccinated in 1986, yet this survey indicates that 58.4 percent of eligible dogs had been vaccinated at least once in the communal lands. However, this figure may be an exaggeration because interviewees could have been anticipating prosecution if they admitted that their dog had in fact not been vaccinated. Regardless, this vaccination coverage is still below the 70 percent level recommended by the World Health Organization (WHO 1984) to control rabies, and therefore it must be concluded that the risk of transmission from dogs to people in communal lands is still unacceptably high.

The dogs most vulnerable to rabies infection appear to be those less than one year old, which represent 41.8 percent of the population. It is this age class that was the least vaccinated in 1994 (Figure 4), largely because many were born since the last vaccination campaign in the area or were too young to be vaccinated at the time (Table 5). To improve rabies vaccination coverage it is this age class that should be targeted. Due to the high turnover within the age class the current frequency of vaccination should be increased to at least twice per annum, and if possible more than this. Brooks (1990) also made this recommendation, having estimated that the majority (33 percent) of the Manicaland Province dog population was less than one year old. However, Foggin (1988) could not support this argument, demonstrating that only 24.1 percent of rabid dogs were less than one year old, although it is not clear whether any of his samples were communal land dogs.

The accessibility of the communal land dog population does not seem to be a problem. Dog owners almost unanimously supported the aim of the rabies vaccination campaign (Table 6) and were happy to bring their dogs to vaccination points when required. Those owners who did not attend vaccination points did so not out of apathy or objection but because they had heard no information about forthcoming vaccination campaigns (Table 5). Furthermore, very few dogs (6.3 percent) had not been vaccinated because they could not be handled and brought to the vaccination point (Table 5). Brooks (1990) suggested that public education about rabies could assist in the success of vaccination campaigns; however, this study suggests that public awareness of the disease was very high in the communal lands surveyed, and that there is little difference between awareness among dog-owning houses and all houses (Table 6).

The problem instead seems to be one of scarce Veterinary Department resources. Vaccination coverage varied considerably between communal lands (Table 5); those with high coverage such as Svosve appear to have consistently regular, efficient vaccination campaigns, and in Svosve's case this may be because of its close proximity to the Marondera Veterinary Office. Dande, on the other hand, has very low coverage and is situated far from the local veterinary office in Mount Darwin.

In light of this, oral vaccination may not be a viable alternative to the parenteral vaccination method currently in use. Oral vaccination by bait is likely to require as great an input of resources such as transport and personnel as the current method does; baits would have to be distributed, either independently of dog owners as Perry *et al.* (1988) experimented, or directly to owners. Even if the method was used, the frequency of vaccinations would still have to be increased to several times per annum, escalating. the demand for resources. Therefore one of the most important criteria to be met by oral vaccination would be cost-effectiveness relative to the parenteral vaccination method.

The suitability of oral vaccination as an alternative in terms of other criteria is mixed. Dog owners almost unanimously supported the idea, but the majority (47 percent) gave no particular reason why (Table 7). Most of the remaining respondents (36.8 percent) supported the idea largely because it avoided handling dogs, which indicates that dog-handling may be more of a problem than the results in Table 5 suggest. The free-ranging, scavenging nature of the dogs' behaviour may make them suitable for the baiting strategies. However., to design a bait distribution method more needs to be known about the typical home range movements of dogs. This issue is currently being studied in Gokwe communal land. If dogs find most of their food by scavenging around people then baits will have to be placed around houses. This would increase the risk of bait tampering by people and the chance of accidental contact with rabies vaccine (Perry and Wandeler 1993). Baiting would therefore involve intensive DVS activity around settlements, and it is possible that door-to-door parenteral vaccination would offer a more cost-effective alternative, particularly since it appears that most dogs can be **handled.**

Table 6. Rabies awareness among the 705 households interviewed and those 513 who owned dogs or who had owned dogs in the past two years ('dog-owning'), and the percentage of dog-owning houses approving of rabies vaccination and supporting the alternative oral technique.

	COMMUNAL LANDS SURVEYED							
	N	**S**	**K**	**G**	**T**	**D**	**M**	**Total**
Households 'quite clear' about rabies	97.0	81.8	67.6	76.4	86.1	46.0	30.0	70.0
Dog-owning houses 'quite clear' about rabies (%)	98.2	82.2	79.5	80.5	87.1	46.2	32.2	71.3
Dog-owning houses approving of rabies vaccination (%)	100	100	100	100	97.8	100	100	99.6
Dog-owning houses supporting oral vaccination	98.2	97.8	94.9	95.1	96.8	93.9	97.7	96.5

Table 7. The primary reasons that the 495 dog-owning households supporting oral vaccination gave for their approval of the technique, expressed as a percentage of supporting households.

	COMMUNAL LANDS SURVEYED							
	N	**S**	**K**	**G**	**T**	**D**	**M**	**Total**
Any method will do if it works (%)	3.5	26.1	86.5	51.3	80.0	11.5	67.1	47.0
Avoids handling dog (%)	42.1	71.6	10.8	26.9	10.0	62.3	27.1	36.8
Saves walking time and misinformation (%)	52.6	2.3	2.7	21.8	10.0	26.2	5.8	16.2

Acknowledgements

This survey would not have been possible without the assistance and co-operation of Provincial Veterinary Offices and the following DVS field staff, Ms Estembelle Nzvimbo (Drihori VEA, Svosve), Mr Sithole (Animal Health Inspector, Marondera), Mr Marowa (Dotito VEA, Kandeya), Mr Matavunye (Gata Rest Camp VEA, Ngorima), Mr Mapfurno (Sengwa Bridge VEA, Gokwe), Mr Zinyama (Field Orderly, Gokwe), Mr Kumalo (Beitbridge VEA, Mtetengwe), Mr Sibanda (Animal Health Inspector, Tsholotsho) and Mr Gondo (Mashumbi Pools Field Orderly, Dande). The Director of Veterinary Services is thanked for his support during this survey. Funding for this part of the dog ecology project was supplied by the British Overseas Development Administration.

References

Brooks R. (1990) Survey of the dog population of Zimbabwe and its level of rabies vaccination. *Veterinary Record* 127: 592-596.

Central Statistics Office (1995) Census 1992: Zimbabwe National Report. Central Statistics Office, Harare, Zimbabwe.

Fekadu M. (1993) Canine rabies. *Onderstepoort Journal of Veterinary Research* 60: 421-427.

Foggin C.M. (1988) Rabies and rabies related viruses in Zimbabwe: historical, virological and ecological aspects. D.Phil thesis, University of Zimbabwe.

Perry B.D. and Wandeler A. L (1993) The delivery of oral rabies vaccines to dogs: an African perspective. *Onderstepoort Journal of Veterinary Research* 60: 451-457.

Perry B.D., Brooks R-, Foggin C.M., Bleakley L, Johnston D.H. and Hill F.W.G. (1988) A baiting system suitable for the delivery of oral rabies vaccine to dog populations in Zimbabwe. *Veterinary Record* 123: 76-79.

Vincent V. and Thomas R.G. (1960) An Agricultural Survey of Southern Rhodesia: Part I, Agro-ecological Survey. Government Printer, Harare, Zimbabwe.

Wandeler A1, Matter H.C., Kappeler A. and Budde A. (1993) The ecology of canine rabies: a selective review. *Revue Scientifique el Technique de I'Office International des Epizooties* 12: 51-71.

WHO (1984) Guidelines for dog rabies control. World Health Organization, Geneva, (VPH/83.43).

WHO/WSPA (1990) Guidelines for dog population management. World Health Organization/World Society for the Protection of Animals, Geneva, (WHO/Zoon/90.165).

POPULATION DYNAMICS OF DOGS IN MACHAKOS DISTRICT, KENYA: IMPLICATIONS FOR VACCINATION STRATEGY

Philip M. Kitala[1]

Department of Public Health, Pharmacology and Toxicology, University of Nairobi, Kenya

Abstract

Results from a one year longitudinal study of dog ecology on 150 dog-owning households in Machakos District, Kenya are presented. The sample of study dogs had a high turnover rate with half of the dogs replaced over the year. Overall life expectancy was estimated at 2.8 to 2.9 years. Males had a live expectancy of 3.5 years and females 2.4 years. Only 39 percent of female dogs survived to one year of age. High mortality was balanced by high fecundity (1.3 females produced per female per year) so that the population was estimated to grow by 9 percent (± 5 percent). Two distinct density (dog/km^2) patterns were observed, one peri-urban area with a dog density of 110/km^2 and five rural areas in which dog densities average 2 approximately 10/km^2 .

Based on these results we conclude that annual rabies vaccination is insufficient and that rabies vaccines which can be given at 3 months of age are required. Further information needs for the appropriate design of vaccination programmes for rural dog populations are discussed.

Introduction

Rabies has been of particular importance in Machakos District, where the disease has been endemic since the mid 1950's, even persisting during the 1970's when rabies Was controlled in the rest of Kenya (Kariuki and Ngulo 1985; Kariuki 1988). In the period 1981 - 1990, 8027 people were officially reported to have been bitten by dogs in the district out of which 4947 received post-exposure treatment (PET) and 22 died due to rabies (Machakos District Hospital). In the same period, 505 animal rabies cases were confirmed (Machakos District Veterinary Department). These officially recorded cases provide only a rough indication of the extent of the rabies problem in the district as many cases go unreported. A recent active surveillance project covering 2.5 percent of the District (Kitala *et al.* 1994) revealed an estimated number of human bite cases of 3680 per annum and confirmed animal cases of 2160 per annum. The number of confirmed animal cases was 40 times the number indicated by passive surveillance, even with only 60 percent of the specimens from potential animal

[1] Co-author and presenter: John J. McDermott, Faculty of Veterinary Medicine, University of Nairobi, Kenya.

cases reported to community rabies workers being diagnosed. Over 90 percent of the confirmed and unconfirmed cases were in dogs.

Many authors (Beran and Frith 1988; Wandeler *et al.* 1988; WHO 1988; WHO/WSPA 1990; Perry 1993) have argued convincingly that dog ecology data are essential for planning_rabies control programmes. Dog ecological data required are: 1) population size and density (total and by risk group); 2) population structure (age and sex); 3) population characteristics (for example movement dependency and restriction) and 4) population dynamics (fecundity, mortality and population growth) (Perry 1993).

This paper is an update of a previous paper by Kitala *et al.* (1993) in which data from an initial visit of dog-owning households is extended with data from a follow-up visit to the same households one year later. In the first paper data were presented on population size, structure, and characteristics. In this paper data on population dynamics are presented, as well as imported dog population estimates. The data from both these papers are then combined to discuss their implications for planning rabies vaccination of the Machakos dog population.

Materials and methods

1. Study area and data collection

The study areas were described and displayed in Kitala *et al.* (1993). Briefly, the study areas were selected by a stratified random sample with six of ten divisions and one sublocation per division in the district selected. A list of households for each study sublocation was compiled with the assistance of the local chief and administrator. The households were re-ordered by random selection and visited in order until 25 dog-owing households had been interviewed.

Two visits were made. On the first visit, two questionnaires, one for household and one for individual dog information, were completed and serum was collected from all dogs (Kitala *et al.* 1993). During the second visit, 12 months later, follow-up data (births, deaths, dog movements and vaccinations) on each dog initially sampled and subsequent acquisitions were collected. During the period between visits, local community rabies workers collected information on animal-bites and specimens for rabies diagnosis (Kitala *et al. 1994),*

To better estimate the dog population in the study areas, a census of all households was Conducted in each study sublocation. Upper primary and secondary school children were recruited to conduct a census of dogs in each study sublocation, during the post-Easter holiday in 1993.

2. Analysis of population dynamics

Parameters for fecundity, mortality (survivorship) and population growth were estimated for the overall population and for age-specific classes (<1, 1-2, 2-3, 3-4 and >4 years). Fecundity rates were calculated as the number of female offspring per female per year using the methods of Caughley (1977). Lifetables (Chiang 1984) were used to calculate age-specific mortality rates and cumulative survival rates by the product-limit method (Kaplan and Meier 1958). Overall and sex-specific survival curves of age versus cumulative survival rate were also generated. Life expectancy (mean age at death) from birth was estimated by two methods, the method of Chiang (1984) and the inverse of the age versus ln(dog number) regression slope (Caughley 1977).

Population growth was estimated using Lotka's equation, by the method described in Caughley (1977). In brief, the sum of the products for each age-class of a function of the population growth rate (r) and the age-specific fecundity and survival rates are set to 1 and r is solved iteratively. The fecundity and survival rates are for the female segment of the population.

Results

1. Dog population

Dog census results, by sublocation, are listed in Table 1. Most sublocations had low to moderate dog densities (6 - 21/kni). One sublocation was markedly different with a density of 110 dog/kin 2 . The dog population estimated during the initial household survey (also in Table 1) had a similar pattern to the census data but systematically underestimated the dog population by approximately 10-20 percent presumably due to households not listed by chiefs but enumerated by school children.

Table 1. Dog population of study sublocations, calculated by schoolboy enumerated census and estimated from a sample of households, Machakos District Kenya, 1992 to 1993.

Sublocation	Number of males	Number of females	Census population (dogs/km^2)	Household sample estimate (dogs/km^2)
Kikambuani	486	395	110.1	83.4
Mikuyuni	176	163	10.3	9.2
Sultan Hamud	346	253	5.8	3.5
Ngoni	285	152	20.8	14.7
Mavau	894	557	16.1	13.9
Ikombe	868	542	11.4	8.6
Overall	3055	2062	13.5	10.4

2. Population dynamics

The number of dogs initially recruited into the follow-up cohort, the losses and additions over the year, and the end study population are listed by age-class in Table 2. Initially, 305 dogs of known ages (15 were unknown) were found in the 150 sampled households. Over the year 125 dogs died or disappeared and 2 were given away. Half the dogs were less than one year of age. All except one of the dogs added to the study households (Table 2) were either born there or acquired as young dogs from neighbours.

Table 2. Cohort of dogs from 150 dog-owning households in Machakos District Kenya, 1992 - 93

Age class (months)	Initial population (known ages)	Losses	Additions	End population
≤ 12	153	82	83	81
> 12 ≤ 24	54	12	1	72
> 24 ≤ 36	46	15	0	42
> 36 ≤ 48	24	10	0	31
> 48 ≤ 60	7	2	0	14
> 60	21	6	0	20
Total	305	127	84	262

Fecundity rates averaged 1.3 female offspring per female per year (Table 3). Two to three year old dogs had peak fecundity compared to younger and older dogs. This was a function of both a higher litter rate and higher number of pups born per litter (Table 3). Births were distributed throughout the year (Figure 1).

The population of dogs was young and had more males in all age classes (Figure 2). This was reflected by the uniformly lower survival rates (Figure 3) for females compared to males. Only 39 percent of females survived to one year of age. The life expectancy for dogs was short. Overall, it was estimated to be 2.9 years by the method of Chiang (1984) and 2.8 years by the method of Caughley (1977). Life expectancy was longer for males (3. 5 years) than for females (2.4 years).

Table 3. Fecundity of 123 female dogs from 150 households in Machakos District, Kenya 1992-93.

Starting age-class (months)	Number of females	Numbers of litters	Pups born per litter	Female offspring per female per year
≤ 12	64	17	3.9	0.7
>12 ≤ 24	26	16	5.3	1.8
> 24≤ 36	16	12	5.4	2.2
> 36 ≤ 48	11	4	4.8	1.1
> 48	6	4	32	1.3
Total or average	123	53	4.7	1.3

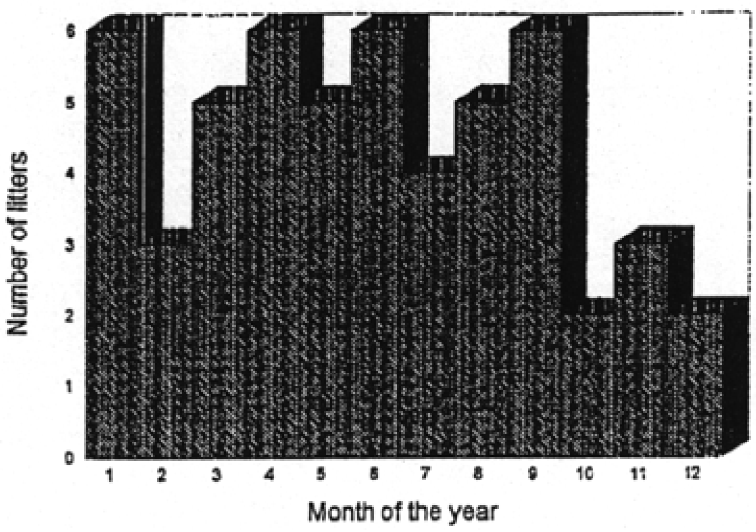


Figure 1. Distribution of births by month of the year for 53 litters born to study dogs in Machakos District, Kenya, 1992-93.

By combining the fecundity and survival data above and assuming a stable age distribution, a population growth rate (r) of 9 percent (±5 percent) was estimated (Table 4). Thus, the low survival rates were more than balanced by high fecundity. The r estimated is age-independent (Scott and Smith 1994).

Table 4. Estimates of population growth in the Machakos dog population, 1992-93.

Starting Age-class (months)	Fecundity (M_x)	Cumulative Survival of Females (L_x)	$M_xL_x\exp(-rx)$[a]
≤ 12	0.7	0.39	0.26
> 12 ≤ 24	1.8	0.27	0.40
> 24 ≤ 36	2.2	0.15	0.25
> 36 ≤ 48	1.1	0.07	0.05
> 48	1.3	0.05	0.04

Growth rate parameter, r = 0.09

[a] where x is the age-class from birth. The first age-class (≤12) is -1, the second is -2 and so on.

Discussion

Rabies occurs commonly in Machakos District and over 90 percent of confirmed rabies cases are canine (Kitala et *al.* 1994). This study had identified several important feature of the ecology of the Machakos District dog population that have implications for dog rabies control. The first is that there is a rapid turnover of the dog population. Dogs, particularly females, have a relatively low life expectancy, presumably due to difficulties in foraging for food. The average life expectancy of 2.8 years is considerably shorter than the 4.5 year life expectancy of well cared for dogs in North America and Europe (Wandeler *et al.* 1988) but higher than the 1.9 years calculated by Gascoyne (1994) for dogs in Serengeti area of Tanzania. High neonatal and post-whelping mortality is primarily responsible for the low life expectancy, with 50 percent of dogs dying in their first year of life. This low survival rate is compensated by a very high fecundity rate of 1.3 female dogs produced per female per year. Thus, over a one year period, half the dog population is replaced.

A second important feature is that despite the low survival rate of female dogs, the population continues to grow. This growth is determined by local factors, since the dog population is essentially closed, with almost no immigration and emigration. It is suspected that the growth of the dog population mainly reflects growth in the human population. Regardless, the current dog population adequately maintains rabies transmission now and will likely continue to do so in future in the absence of effective rabies control.

The third important feature is that two distinct density patterns exit. Dog:human population ratio estimates range from L5.5 to 1:16 with an overall mean of L9.5 (Kitala *et al.* 1993). These are consistent with estimates by Gascoyne (1994) in Tanzania. However, within Machakos District there are major differences in dogs/kni2, reflecting the large differences in human population densities. One of the study areas, Kikambuani sublocation in Kangundo location, had both human and dog densities 10 times higher on average than the other study areas. This area is considered a peri-urban area. In contrast, the remaining five, more rural, areas had dog densities of six to 20/km^2.

The other main feature is the pattern of dog ownership, restriction and dependency. All dogs are owned and return to their household of origin, but 69 percent of dogs have little or no restriction on their movements. Restriction is more common in the higher density sublocation where dogs are primarily used for guarding. The number of dogs per household is low and variable. On average, households have 2 dogs. Given the **frequent population turnover,** households can easily lose all their dogs.

What are the implications for rabies control in Machakos District? Let us assume that, as stated by Bogel and Meslin (1990), effective dog vaccination is the most cost-effective local approach to controlling rabies and that a vaccination coverage of 70 percent is required. First since there is such a high dog population turnover rate, annual vaccination is inadequate to maintain a 70 percent coverage. Second, since most dogs and almost all dogs-at-risk are less than one year of age, vaccination of young dogs is essential. Currently, rabies immunisation is only initiated from 3 months of age. It would be desirable to have most dogs effectively protected by that age.

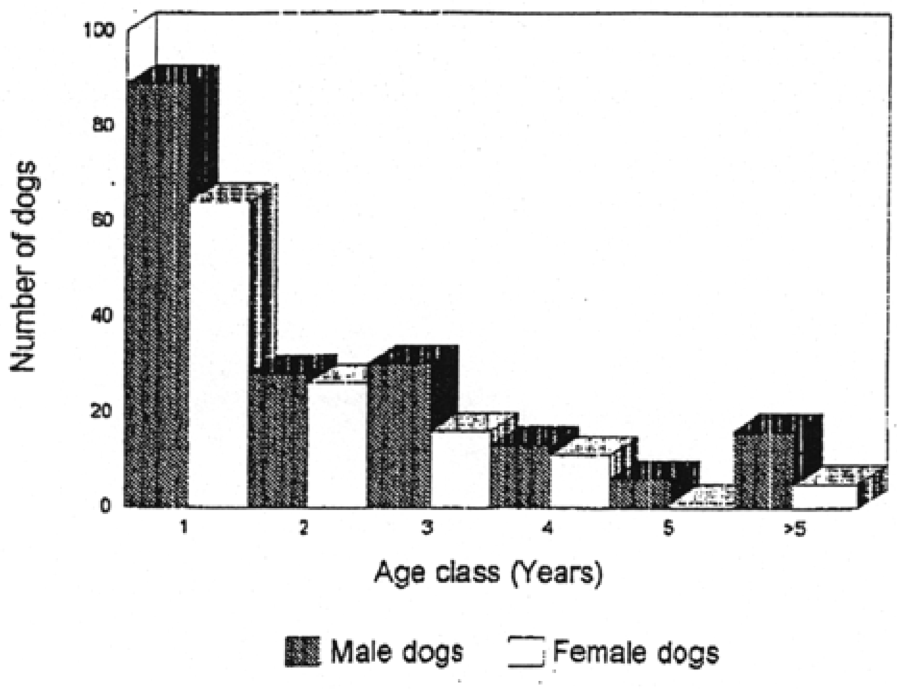


Figure 2. Age-sex structure of 305 study dogs from Machakos District, Kenya, 1992-93

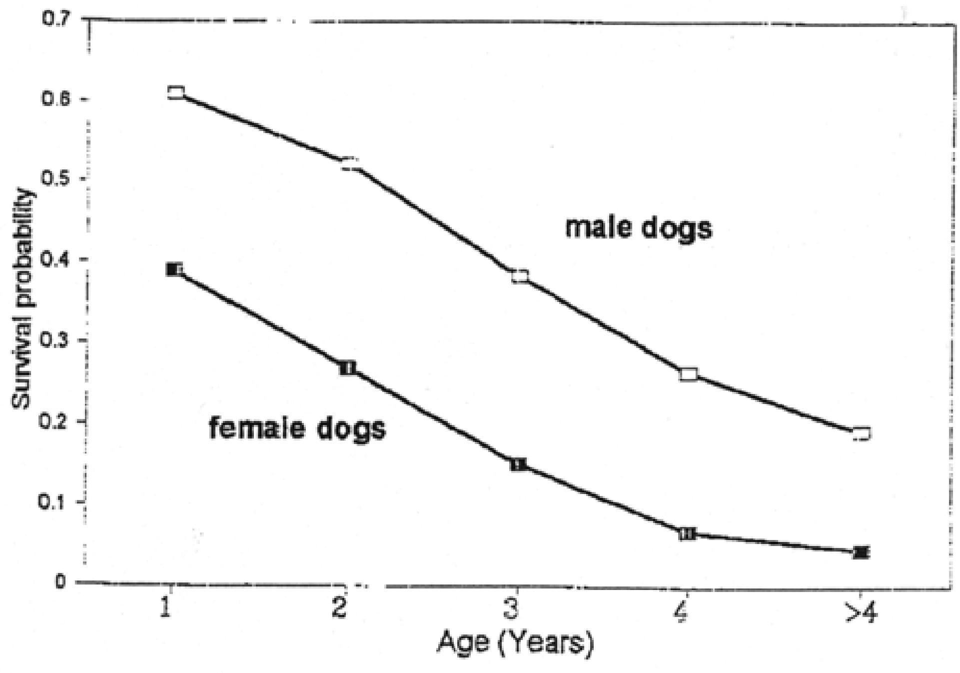


Figure 3. Cumulative survival of 305 study dogs (182 males and 123 females) in Machakos District Kenya, 1992-93.

In planning an effective vaccination programme community participation will be an essential prerequisite. Our recent active surveillance study in Machakos indicates that there is a high degree of public awareness and interest in rabies control. The type of community-based vaccination programmes will depend on dog density and socio-economic conditions. It is likely that for the higher-density peri-urban areas, the methods of Perry *et al.* (1995), who achieved 70 percent vaccination coverage in a Nairobi suburb, could be used. However, programmes for lower density rural areas need more investigation. Some of the essential questions with regards to vaccination in rural areas are: how can dogs best be contacted? what vaccination coverage is achievable? does vaccination need to be combined with some type of dog control? and what are the rural poor willing to pay for vaccination? We hope to explore these question by field trials in our rural study areas in the near future.

References

Beran G.W. and Frith M. *(1988)* Domestic animal rabies control: an overview. *Reviews of Infectious Diseases 10:S672-677.*

Bogel K. and Meslin R-X *(1990)* Economics of human and canine rabies elimination: guidelines for programme orientation. *Bulletin of the World Health Organization 68:28 1 -291.*

Caughley G. *(1977)* Analysis of vertebrate populations. Wiley, New York.

Chiang L. *(1984)* The life table and its application. Robert E. Kreiger, Malabar FL, USA.

Gascoyne S. *(1994)* Rabies in the Serengeti region of Tanzania. 0DA/NRRI) Animal Health Programme Final Report. Institute of Zoology - Regent's Park and London School of Hygiene and Tropical Medicine, London. *71 pp.*

Kaplan E.L. and Meier P. *(1958)* Nonparametric estimation from incomplete observations. *Journal of the American Statistical Association 53:457-48 1.*

Kariuki D.P. *(1988)* The epidemiology and diagnosis of rabies in Kenya. *Kenya Veterinarian 12:32-35.*

Kariuki D.P. and Ngulo W.K. *(1985)* Epidemiology of animal rabies in Kenya *(1900 - 1983).* In: *Rabies in the Tropics,* Edited by Kuwert E., Merieux C, Koprowski H. and Bogel K. Springer-Verlag, Berlin. pp. *451-464.*

Kitala P.M., McDerraott J1, Kyule M.N. and Gathuma 1M. *(1993)* Features of dog ecology relevant to rabies spread in Machakos District, Kenya. *Onderstepoort Journal of Veterinary Research 60:445-449.*

Kitala P.M., McDerinott J1, Kyule M.N. and Matere C.N. *(1994)* Investigation of rabies incidence and control by active surveillance in Machakos District Kenya. *Kenya Veterinarian 18:528-530.*

Perry B.D. (1993) Dog ecology in eastern and southern Africa: implications for rabies control. *Onderstepoort Journal of Veterinary Research* 60:429-43 6.

Perry B.D., Kyendo T.M., Mbugua SW., Price J.E. and Varma, S. (1995) Increasing rabies vaccination coverage in high human population density suburbs: a case study in Nairobi, Kenya. *Preventive Veterinary Medicine* 22:137-145.

Scott M. and Smith G. (editors) (1994) Parasitic and infectious diseases (epidemiology and ecology). Academic Press, New York.

Wandeler A1, Budde A., Capt S., Kappeler A. and Matter H. (1988) Dog ecology and dog rabies control. *Reviews of Infectious Diseases* 10: S684-688.

WHO (1988) Report of a WHO consultation on dog ecology studies related to dog rabies control, 22 - 25 February, 1988. World Health Organization, Geneva (WHO/Rab.Res/88.25).

WHO/WSPA (1990) Guidelines for dog population management. World Health Organisation, Geneva (WHO/Zoon/90.165).

CANINE RABIES IN SOUTH AFRICA

George C. Bishop

Allerton Regional Veterinary Laboratory, Cascades 3202, Pietermaritzburg, South Africa

There are many factors which have contributed to the marked increase in canine rabies in South Africa over the past two years. Dramatic political changes have led to immense social reformation and this reformation has moved the focus away from the control of infectious diseases. Rabies, particularly in the domestic dog, has flourished and control measures have been extremely difficult to implement.

There are several areas in South Africa which are associated with specific vectors (see Figure 1). This figure has been produced on overlays and when combined, they give a chilling picture of the situation in South Africa (Figure 2). Sylvatic rabies is determined by the distribution of the vector, but the domestic dog occurs where people occur - the more people, the more dogs. The area worst affected by canine rabies in South Africa is Kwazulu Natal (Figure 3) and it is on this area that my attention will be focused. Nearly 90 percent of the rabies cases in Kwazulu Natal are recorded in dogs (Figure 4).

The increase in the number of rabid dogs recorded during 1994, compared to 1993, bears testimony to the deteriorating situation (Figure 5). The worsening rabies scenario was particularly severe in Kwazulu Natal, although a proportion of these cases could be attributed to increased vigilance and improved specimen retrieval. We also know of certain areas where there have been many suspect rabid dogs observed but no specimens have been forthcoming. A large proportion of our cases can be traced back to areas close to roads - is this really the situation or does this merely reflect fewer difficulties in transport of the specimens? Our records therefore reflect where rabies cases were diagnosed and not necessarily where all the cases occurred. It has often been claimed in Kwazulu. Natal that as few as 20 percent of the cases are diagnosed. Certainly nearly half of all the cases go unrecorded because in very few instances are the dog vector and animal victim both produced at the laboratory.

In Kwazulu Natal, nearly all cases diagnosed are caused by the canid strain. The disease is clearly dog-driven and Figure 6 shows the total number of cases compared to the number of canine rabies cases for the 34 year period, 1961-1994. The interesting increase in the latter half of the year is also obvious. A cyclical pattern has been seen every year over the periods in which rabies has occurred in Natal. This phenomenon is shown for a seven year period in Figure 7.

Apart from the peaks within each year, there may be an increase in the disease every 3 to 4 years (Figure 8). Some of these tendencies can be explained on the basis of reactive control measures whereas others are much more difficult to work out.

Why is the problem so bad in Kwazulu Natal and not so serious elsewhere? Why is the disease worse in some areas of Kwazulu Natal than in others? These are questions which we ask ourselves continuously but are unable to answer. The ways in which rabies is controlled do not vary much in South Africa and it seems only a matter of time before the disease breaks out in the vast townships like Soweto (Transvaal), Mdantsane and Zwelitsha (both eastern Cape). Our vigilance in these vast townships has slipped and rabies could already be established there. The Natal situation may well be repeated elsewhere in South Africa soon.

The areas in which at least one case was recorded during 1989 are shown in Figure 9(a), and when compared with those recorded during the 3-year period, 1992 to 1994 (Figure 9(b)), the deteriorating situation becomes patently obvious. Why has this occurred? There are many factors influencing spread and these explain why the disease is worse in some areas than in others. Most of our cases occur where there are high numbers of people and therefore high numbers of dogs. The worst affected areas lie adjacent to the coast. Quarter degree plots reflecting the density of rabies cases are shown in Figure 10. The quarter degree is approximately 20 km by 20 km in Natal and this is a convenient area size to depict rabies prevalence.

Within Kwazulu Natal, there are immense differences in the topography, the concentration of people (and therefore the density of dogs) and the weather conditions. The huge Drakensberg mountain range on Natal's western border, is an effective barrier whereas most of our rivers do not impede spread of the disease. Man-made political boundaries are obviously completely ineffective. The old adage that rabies travels best by car still holds and this is where the danger lies because the urbanisation rates recorded in the Durban area are amongst the highest in the world and people continue to move into the cities, bringing with them their dogs.

In the opulent central Durban urban area, there are in excess of 3 000 people/km^2 and in the surrounding poorer peri-urban areas, informal settlements reach 6 000/km^2. In the rural areas of Impendhle, there are fewer than 3 people/km^2 . We have conducted a survey which involved nearly 250 000 respondents and from this we have calculated that the human to dog ratio is 4:1. The squatter areas of Durban have nearly 1 500 dogs/km^2 and Impendhle only 0.6 dogs/km^2 . The average dog density for Durban and the adjacent Chatsworth urban area is 465 dogs/km^2 . The marked differences in the number of recorded cases inAifferent areas during the period 1988 - 1994, when compared to the dog density, reveals the expected relationship quite clearly (Table 1).

Table 1. Dog density and rabies cases in Natal.

District	Dogs/sq.km	Cases
Durban & Chatsworth	465	184
Pinetown	175	111
Pietermaritzburg	85	67
Impendle	0.6	1
Estcourt	4	2
Mooi River	3.9	2

The feral dog is not an important factor in the epidemiology of canine rabies, as nearly all dogs have owners in Kwazulu Natal. There is, however, a high proportion of unfenced, unsupervised dogs. The level of vaccination is variable and it is certainly true that where rabies continues to ferment the levels of vaccination have not even nearly approached the required 70 percent level. Dog ecology is also receiving our attention and we are looking into dog turnover, average ages, litter sizes, the reasons for keeping dogs (pets, protection, hunting etc) and the way they are kept. These factors vary from area to area and from house to house.

Canine rabies is a multifactorial disease, but if there is one factor that could be considered the most important, it might be the socio-political one. The present outbreak occurred in 1976 and this co-incided with the fragmentation of Natal into more than double the number of districts (Figures 11). Along with this came a division of veterinary responsibility and control and a completely untenable situation resulted. At present, control measures are very difficult to apply and many of the worst affected areas are the ones where it is unsafe to enter in an organised and predetermined manner. Police protection is not always the answer as the police are often viewed with distrust and suspicion because they are perceived to be partisan. Violence, suspicion and general apathy are hardly conducive to good vaccination strategy. The poorer people, who own most of the dogs that become infected, have priorities which are far more important than rabies vaccination. Housing, education, security, food, money, health, water and electricity are often taken for granted by the affluent but these commodities are pivotal in the very fight for survival of the poor.

How can we hope to control canine rabies? Ideally we would like to eliminate strays and vaccinate at least 70 percent of the canine population. We have learned that door-to-door vaccination may be better than providing clinics in some areas. The timing of clinics is often crucial and an improved response is obtained when vaccinating out of normal work hours. We do not charge to vaccinate and free vaccine has been given to SPCM and private practitioners. Administrative bottlenecks have been eliminated, such as simplifying the "proof of vaccination" certificate.

Our immunisation figures are improving (Figure 12) and there have been a few areas where we have turned the disease around. Last year's vaccination figures are not yet available but there is reason to believe that they were much higher than the previous year. We have embarked on research which will possibly allow us to make use of bait vaccine in dogs in some of the areas which are difficult to work using our normal approach.

We most certainly are not going to give up. The control of canine rabies will improve as our country stabilises. We remain optimistic.

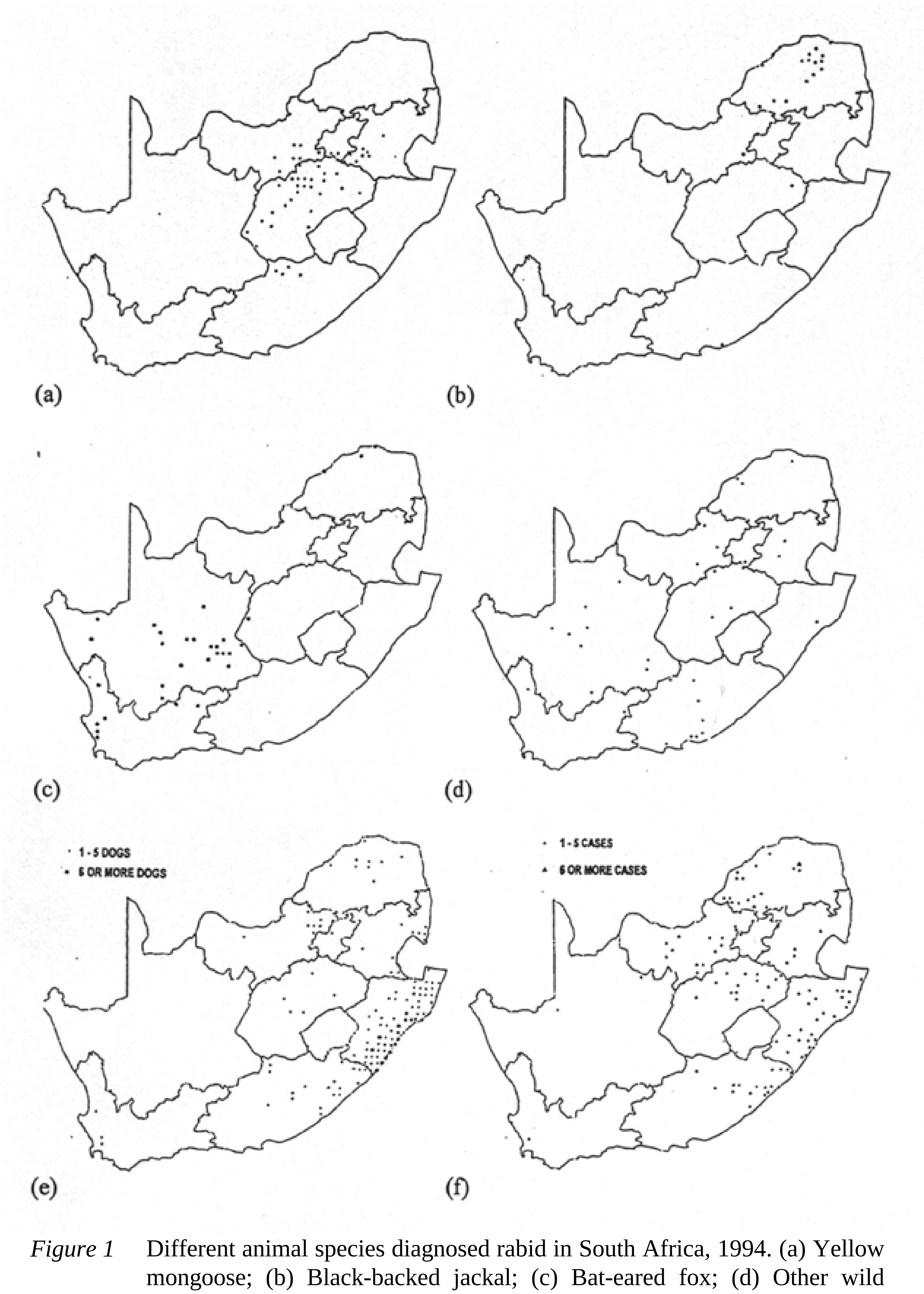


Figure 1 Different animal species diagnosed rabid in South Africa, 1994. (a) Yellow mongoose; (b) Black-backed jackal; (c) Bat-eared fox; (d) Other wild animals; (e) Dogs; (f) Other domestic animals.

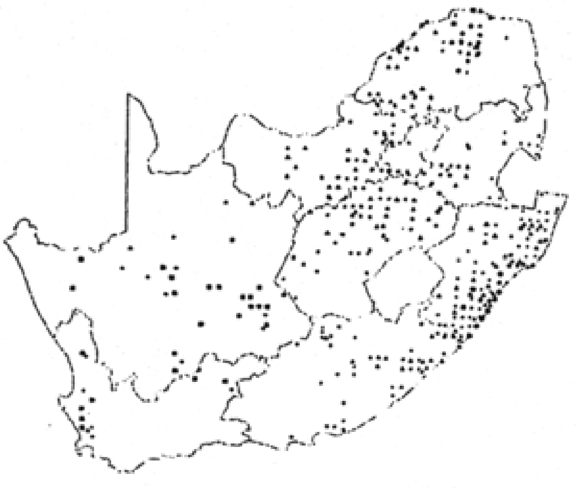


Figure 2. Rabid animals (all species) diagnosed in South Africa in 1994.

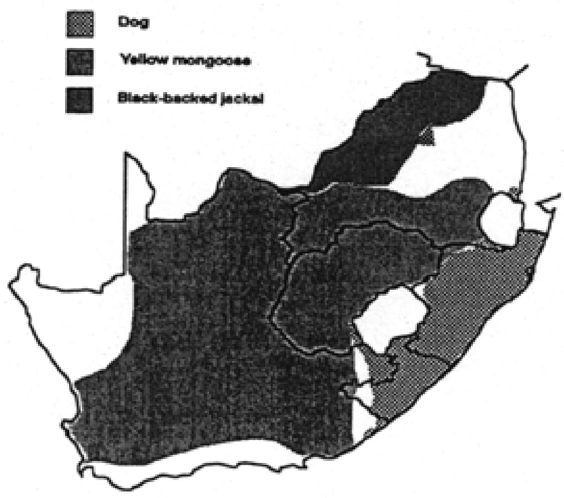


Figure 3. The main vectors of rabies in South Africa.

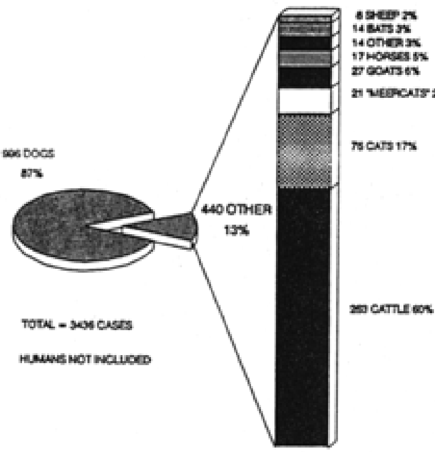


Figure 4. Natal rabies cases, 1961 - 1994.

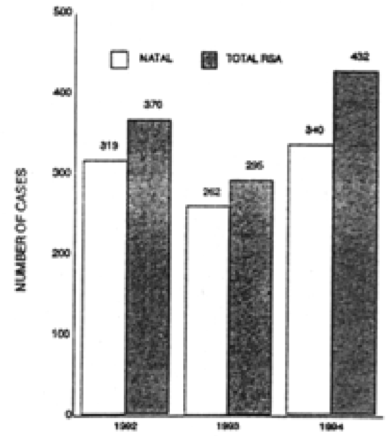


Figure 5. Canine rabies in South Africa.

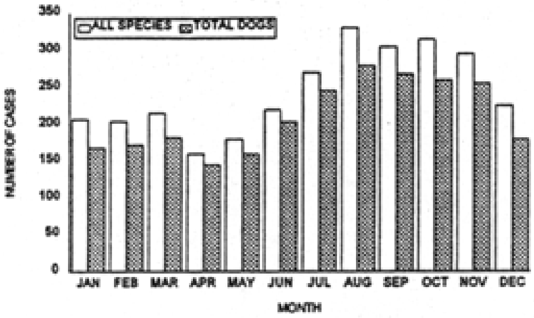


Figure 6. Rabies in Natal, 1961 - 1993.

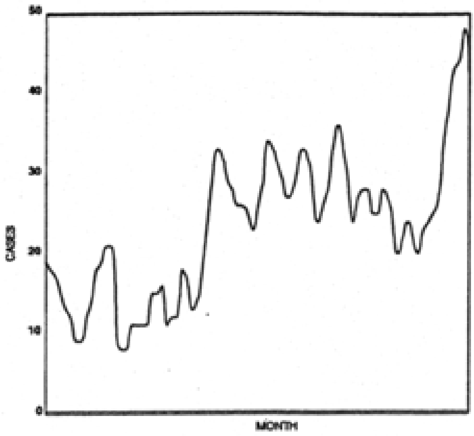


Figure 7. Natal rabies cases, all species, 3-month rolling average, 1987 - 1994.

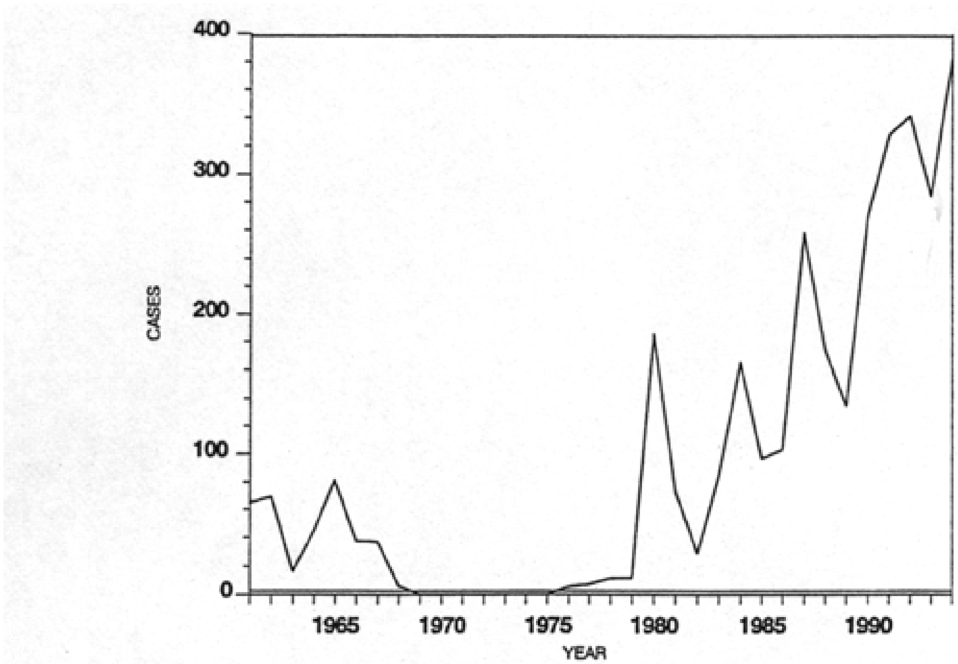


Figure 8. Natal rabies cases, domestic animals, 1961 - 1994.

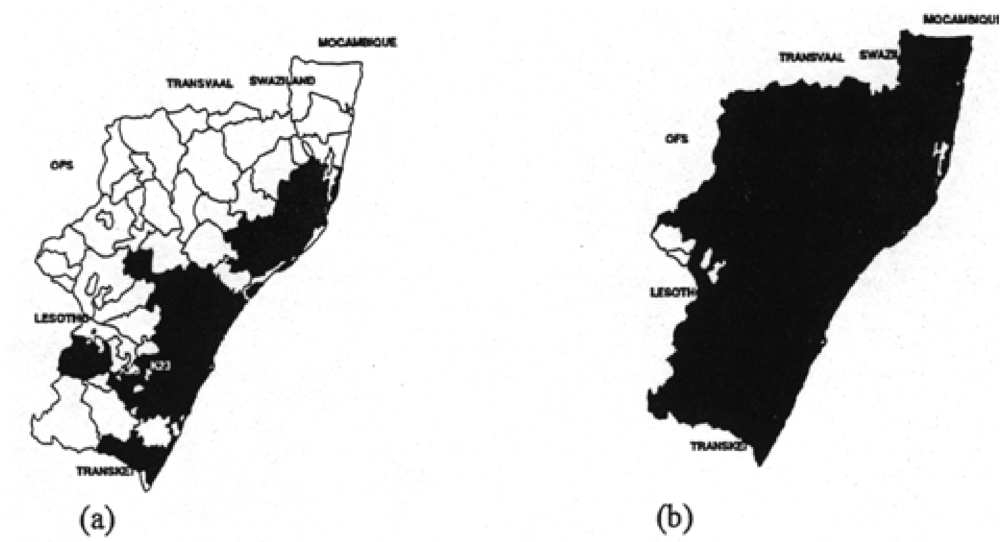


Figure 9. Natal districts infected in 1989 (a) and 1992 - 1994 (b).

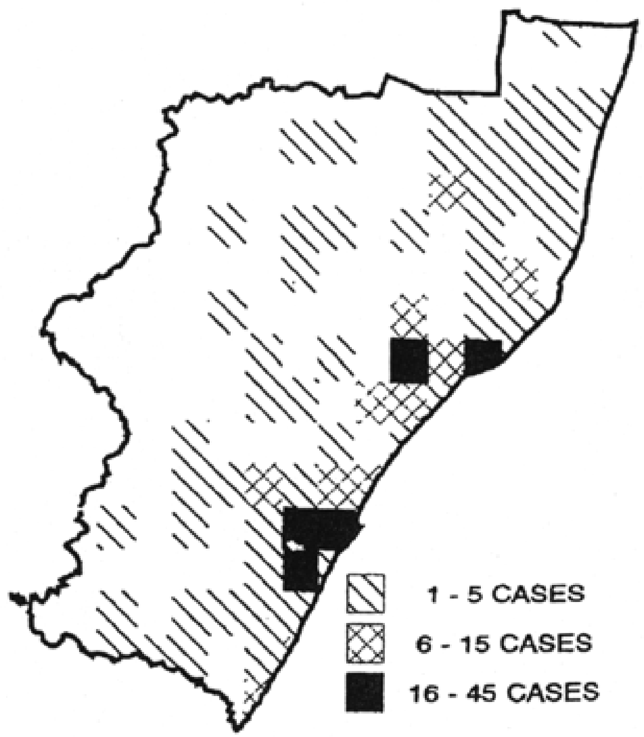


Figure 10. Rabies prevalence in Natal in 1994 - 1/4 degree data.

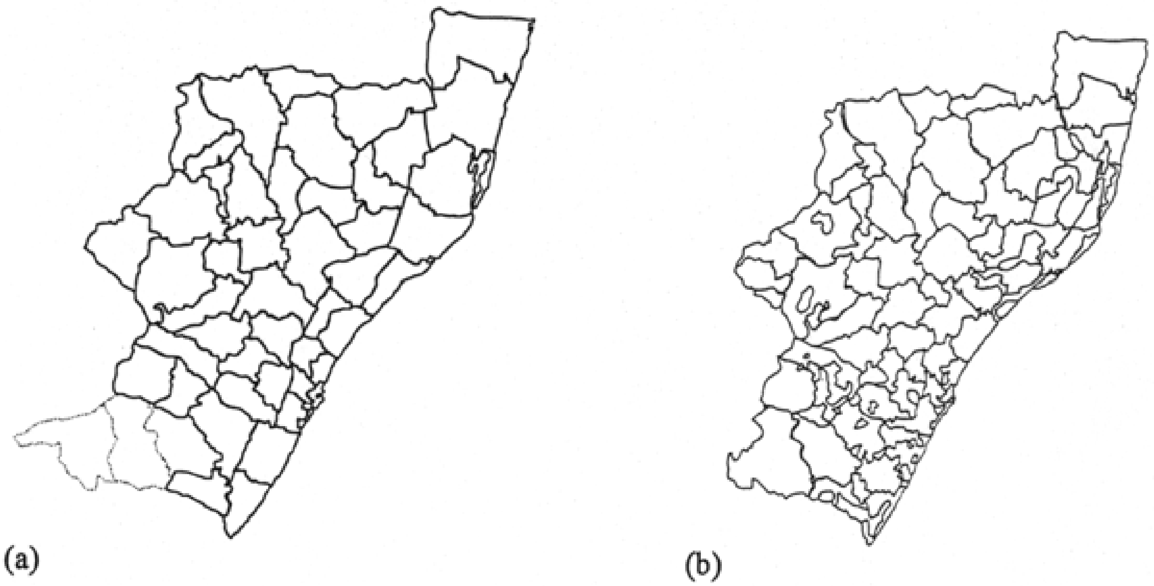


*Figure 11.*Natal districts in 1975 (a). Kwazulu Natal districts in 1994 (b)

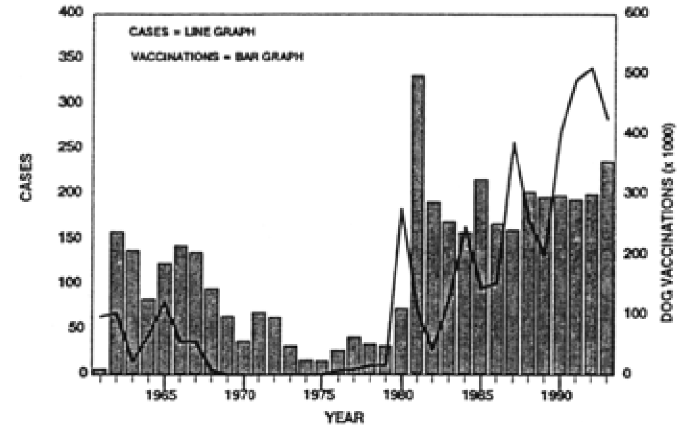


*Figure 12.*Natal rabies cases and dog vaccinations, 1961 - 1993.

RABIES IN THE SERENGETI: THE ROLE OF DOMESTIC DOGS AND WILDLIFE IN MAINTENANCE OF DISEASE[1]

Sarah Cleaveland [2,3]

Institute of Zoology, Zoological Society of London

Introduction

Although dogs account for over 90 percent of human rabies cases (WHO 1992), relatively little is known about the epidemiology of canine rabies. Much of our understanding of rabies epidemiology comes from empirical and theoretical studies, not of dogs, but of wildlife populations in Europe and North America. Here, in any given geographic area, antigenically and genetically distinct strains of rabies (virus "biotypes") are maintained by a single principal host species (Carey 1985; Blancou 1988; Smith 1989; Rupprecht *et al* 1991; Wandeler 1991; Blancou *et al.* 1991).

The increasing incidence of dog and wildlife rabies in Africa (King 1993) is causing concern, not only for public health but also for the conservation of some endangered canids (Macdonald 1993). In the Serengeti, recent rabies outbreaks have seriously affected the small population of African wild dogs *(Lycaon pictus)*(Gascoyne *et al.* 1993; Alexander *et* al 1993) and control of the disease has become a component of their conservation strategy. However, in order to control rabies effectively, we need to identify which animals are reservoirs and by what mechanisms the disease is maintained in reservoir populations.

Reservoir hosts in the Serengeti

In the first part of this study we evaluate the roles of domestic dogs and wildlife as reservoirs of rabies in the Serengeti, using five criteria: 1) reservoir host populations should show evidence of persistent infection, 2) cases should occur in the reservoir host in the absence of cases in other species, but the converse should not occur, 3) outbreaks in other species should follow cases in the reservoir host population, 4) control of rabies in the

[1]This contribution is a summary of the paper: Cleaveland and Dye (1995) Maintenance of a microparasite infecting several host species: rabies in the Serengeti. *Parasitology* (in press).

[2] Co-author: **C. Dye**, Vector Biology and Epidemiology Unit London School of Hygiene and Tropical Medicine, Keppel Street, London, United Kingdom WC1E 7HT.

[3] Formerly S.C. Gascoyne.

reservoir host should result in elimination of disease in all other species, and 5) the virus isolate that is characteristic of the reservoir species should be found in all other species.

1 . Evidence for maintenance in dog populations. Historical records show that from 1958 to 1977, rabies was apparently absent from the Serengeti (Rweyemamu *et al.* 1973; Magembe 1985). In Serengeti District (SD) (an agropastoralist area to the west of the Serengeti National Park), dog cases, or human cases derived from dogs, were reported every year for which records were available (1977-1984; 1986-1995). In contrast, rabies was reported only sporadically in pastoralist areas to the east of the park(Loliondo Game Control Area, LGCA and Ngorongoro Conservation Area, NCA).

Evidence for maintenance in wildlife populations. Since the early 1980s, rabies has only rarely been recorded in Serengeti's wildlife, despite intensive monitoring of carnivore populations. The disease has been confirmed in two wild carnivore species -African wild dogs in 1990 (Gascoyne *et al.* 1993) and bat-eared foxes *(Otocyon megalotis)* in 1987 and 1988 (Maas 1993), and in 1994 and 1995. The African wild dog population is undoubtedly too small and dispersed to maintain rabies. While recurrent cases have occurred in bat-eared foxes, the short duration of epidemics (5 and 7 weeks in 1987 and 1988 respectively)(Maas 1993) together with an absence of reported cases between 1988 and 1994 suggests that this species is also unlikely to be a reservoir host in Serengeti.

2. Independent maintenance of infection. Since rabies is maintained in domestic dog populations of SD but occurs only sporadically in wildlife, rabies in dogs can occur in the absence of cases in wildlife, but wildlife rabies does not occur in the absence of dog rabies.

3. Temporal sequence of epidemics. With so few rabies cases in Serengeti, we draw on data from Zimbabwe to investigate the temporal relationship between dog and wildlife cases. Case incidence data from 1950-1991 (Foggin 1988; Bingham 1993) together with results of a cross-correlation analysis indicate that cases in dogs preceded outbreaks in jackals with a la of one year ($r=0.523$, $p<0.05$). During this period, therefore, rabies epidemics in jackals appear to have been driven by infection in dogs and not vice versa.

4. Rabies control. In the late 1950s, dog rabies control measures (dog vaccination, movement restriction and culling) resulted in the apparent elimination of disease from the Serengeti District between 1958 and 1977 (Rweyemamu *et al.* 1973; Magembe 1985).

5. Virus isolates. Three virus isolates from Serengeti (a domestic dog, an African wild dog and a cow) were found to be antigenically and genetically indistinguishable with characteristics consistent with southern African canid-associated virus. These results support findings of other studies, which show that three African wild dog isolates from the Masai Mara National Reserve, Kenya (part of the Serengeti ecosystem) were identical both to the Serengeti African wild dog isolate and to four domestic dog isolates from the Masai Mara (Alexander *et al.* 1994).

Drawing together these data, we argue that domestic dogs, not wildlife, are the likely reservoir of rabies in the Serengeti. It is clear, however, that surveillance measures need to be improved before definite conclusions can be reached. In particular, the observation that bat-eared foxes can maintain rabies in parts of South Africa (Thomson and Meredith 1993) emphasises the need for further investigation of the role of this species in the Serengeti.

Mechanisms of disease maintenance

In the second part of the study, we explore possible mechanisms of maintenance in dog populations in Serengeti. Although rabies is among the best studied microparasite infections, explanations for disease maintenance remain problematic. Several mechanisms have been proposed to account for persistence, for example, reintroduction of disease (Voigt *et al.* 1985; Artois *et al.* 1991), inapparent reservoirs (Carey 1985), prolonged or variable incubation periods (Bacon 1985; Aubert *et al.* 1991) and carrier animals (Crandall 1991). Although many of these can be invoked for dog rabies, few studies have evaluated the quantitative significance of different possible mechanisms of disease maintenance in dog populations.

In theory, microparasite infections are more likely to be maintained at higher host densities. In this study, we provide evidence for a threshold density for rabies persistence in Serengeti, with infection appearing to be maintained in populations exceeding 5 dogs/km^2 (SD), but not in pastoralist populations (LGCA and NCA) of less than 1 dog/km^2. However, this threshold is several orders of magnitude lower than has been suggested from other studies. For example, dog rabies in Guayquil, Ecuador occurred continuously where dog densities exceeded 600 dogs/km^2, but only sporadically in lower density areas (Beran and Frith 1988). These observations led us to investigate the role of atypical rabies in possible mechanisms of disease maintenance in the Serengeti.

Rabies Serology

Results of serological analyses using a liquid-phase blocking ELISA (Esterhuysen *et al.* 1995) have shown that a proportion of healthy, unvaccinated Serengeti dogs have detectable serum levels of rabies antibody. Several lines of evidence support the view that rabies seropositivity detected by this test reflects genuine, and recent exposure to rabies virus.

a) Antibody titres in Serengeti seropositives were higher than in any individual from rabies-free islands (89 dogs from Mauritius and 99 dogs from UK).

b) Seropositive dogs were significantly more likely to be found in villages where rabies had been confirmed or reported than in villages where it had not (odds ratios with 95 percent confidence limits as follows: for villages where rabies confirmed OR= 3.96 (1.88-8.24), $p<0.001$, n=900; for villages where rabies reported OR=5.02 (2.41-10.66), $p<0.001$, n=900).

c) Individual case history data. i) Seroconversion was detected in a dog which had been bitten by a suspect rabid dog 2-4 weeks earlier and which remained healthy for at least four months; ii) Four of 12 dogs bitten by a suspect rabid dog one to two weeks earlier were seropositive. Two dogs remained healthy and seropositive for at least two years, with fluctuating antibody levels. One of the 8 bitten dogs that was seronegative died of confirmed rabies and three died of suspect rabies one to two months later.

In a follow-up study, only one of 32 seropositives subsequently died of suspect rabies and this was an animal showing neurological signs at the time of sample collection. Nine seropositives remained alive and healthy for at least two years. Case histories indicated that none of the seropositive dogs had recovered from neurological disease. Rabies virus was not isolated from saliva samples collected from six seropositive dogs. Of the atypical rabies infections that have been described (recovery, carrier states, latency, aborted rabies, reactivation of infection; Fekadu 1991), an interpretation of "aborted" rabies is the most consistent with our observations.

Mathematical models

While we cannot explain rabies seropositivity in terms of rabies pathogenesis or immunity, we can use mathematical models to explore three different interpretations of seropositivity -long incubators, carriers and immune animals. Each has been invoked as a possible mechanism for rabies maintenance (for example Bingham *et al.* 1994; Chaparro and Esterhuysen 1993 for long incubators; Andral 1964; Fekadu 1991 for carrier animals; Coyne *et al.* 1989 for immune animals). In this study, infection parameters derived from Foggin (1988) and dog demographic data from Serengeti were incorporated into a deterministic model of classic dog rabies, which generated regular cycles of disease incidence. The model was relatively insensitive to the inclusion of 5 percent of dogs as long incubators (with an incubation period of 20 weeks rather than 4 weeks), or 10 percent dogs as immune animals. In contrast inclusion of only 0. 1 percent carrier dogs had a marked impact on the dynamics, stabilising fluctuations in disease incidence and lowering the probability of extinction in troughs between epidemics. While it was beyond the scope of this study to carry out extensive surveys for carrier animals, studies in Ethiopia and Nigeria have demonstrated salivary virus excretion in 0.46 percent (5/1083) and 0.27 percent (4/1500) of healthy dogs respectively (Fekadu 1972; Aghomo *et al.* 1989).

Summary

Drawing together data from recent field studies and historical records, we present evidence to support the view that domestic dogs are the likely reservoir of rabies in the Serengeti. Rabies is maintained in dog populations of Serengeti District where densities exceed 5 dogs/km^2 but not in pastoralist dog populations of less than 1 dog/km ' suggesting not only that there is a threshold density for rabies maintenance, but also that disease control measures should be directed towards higher-density dog populations. Rabies seropositivity occurs in a proportion of healthy, unvaccinated Serengeti dogs. While an interpretation of rabies seropositivity in terms of rabies pathogenesis has not been possible, mathematical models show that maintenance of rabies in Serengeti dog populations is more likely if seropositives are infectious carriers, rather than long incubators or immune animals.

Acknowledgements

SC was supported by grants from the Agricultural and Food Research Council and ODA/NRRI) Animal Health Programme. We thank Tanzania National Parks, Ngorongoro Conservation Area Authority and Serengeti Wildlife Research Institute for permission to work in the Serengeti. We thank the following for assistance in Serengeti: Dr M. Roelke-Parker, Dr M. Kaare, Dr S. Mkumbo, Dr P. Loomu and Dr F. Gulland. Frankfurt Zoological Society and the British Development Division in Eastern Africa provided logistic support. We thank Dr G. Thomson and Mr J. Esterhuysen at the Onderstepoort Institute for Exotic Diseases for providing facilities and assistance with rabies serological analyses, Dr J. Barrat, CNEVA, Nancy, France for provision of diagnostic kits, carrying out diagnostic tests and virus isolation, and Dr J. Shuja, Mauritius for kindly collecting reference dog sera.

References

Aghomo H.O., Oduye 0.0., Tomore I. and The M. (1989) Isolation of rabies virus from clinically healthy and previously unvaccinated dogs. *Bulletin of Animal Health and Production in Africa.* 37:13 1-13 5.

Alexander K.A., Smith J. S., Macharia MJ. and King A.A. (1993) Rabies in the Masai Mara, Kenya: preliminary report. *Onderstepoort Journal of Veterinary Research* 60:411-414.

Alexander K.A., Smith J. S., Macharia M1 and Kitala P. (1994) Molecular epidemiology of rabies in Kenya. In: *Proceedings of the Ninth International Conference of Negative Strand Viruses, Portugal. p.* 180.

Andral L. (1964) Une maladie comme les autres: la rage. Dix annees d'ex~perimentation sur la rage en Ethiopie. *Annales de l'Institut Pasteur Ethiopie* 1(5):58-65.

Artois M., Aubert M., Blancou J, Barrat J, Poulle M.L. and Stahl P. (1991) Ecologie des comportements de transmission de la rage. *Annales Recherches Veterinaires* 22:163-172.

Aubert M.F.A., Blancou 1, Barrat J, Artois M. and Barrat MJ. (1991) Transmission et pathogenie chez le renard roux de deux isolats A dix ans dIntervalle du virus de la rage vulpine. *Annales Recherches Veterinaires* 22:77-93.

Bacon PJ (1985) A systems analysis of wildlife rabies epizootics. In: *Population dynamics of rabies in wildlife.* Edited by Bacon P.J. Academic Press, London. pp. 109-130.

Beran G.W. and Frith M. (1988) Domestic animal rabies control: an overview. *Reviews of Infectious Diseases* 10:S672-S677.

Bingham J. (1993) Rabies in Zimbabwe. In: *Proceedings of the International Conference on Epidemiology, Control and Prevention of Rabies in Eastern and Southern Africa, Lusaka, Zambia, 1992.* Edited by King A.A) Editions Fondation Marcel Merieux, Lyon. pp. 29-33

Bingham L, Hill F.W.G.. and Matema R. (1994) Rabies incubation in an African civet *(Civettictis civetta). Veterinary Record* 134:528.

Blancou J. (1988) Ecology and epidemiology of fox rabies. *Reviews of Infectious Diseases* **10**:S606-S614.

Blancou L, Aubert M.F.A. and Artois M. (1991) Fox Rabies. In *The Natural History of Rabies. 2ndEdiflon.* Edited by Baer G.M. CRC Press, Boca Raton. pp. 257-290.

Carey A.B. (1985) Multispecies rabies in the eastern United States. In: *Population dynamics of rabies in wildlife.* Edited by Bacon P.J. Academic Press, London. pp. 23-4 1.

Chaparro F. and Esterhuysen JJ. (1993) The role of the yellow mongoose *(Cynictis penicillata)* in the epidemiology of rabies in South Africa - preliminary results. *Ondersterpoort Journal of Veterinary Research.* 60:373-377.

Coyne MJ., Smith G., & McAllister F.E. (1989) Mathematic model for the population biology of rabies in raccoons in the mid-Atlantic states. *American Journal of Veterinary Research* 50: 2148-2154.

Crandell R.A. (1991) Arctic fox rabies. In: *The Natural History of Rabies. 2nd Edition.* Edited by Baer G.M. CRC Press, Boca Raton. pp. 291-306.

Esterhuysen LL, Prehaud C. and Thomson G.R- (1995) A liquid-phase blocking ELISA for the detection of antibodies to rabies virus. *Journal of Virological Methods* 50:31-42.

Fekadu M. (1972) Atypical rabies in Ethiopia. *Ethiopian Medical Journal* 10:79-86.

Fekadu M. (1991) Latency and aborted rabies. In: *lhe Natural History of Rabies. 2nd Effition.* Edited by Baer G.M. CRC Press, Boca Raton. pp. 191-198.

Foggin C.M. (1988) Rabies and rabies-related viruses in Zimbabwe: historical, virological and ecological aspects. PhD. thesis, University of Zimbabwe, Harare.

Gascoyne S.C., King A.A., Laurenson M.K., Borner M., Schildger B and Barrat J. (1993). Aspects of rabies infection and control in the conservation of the African wild dog *(Lycaon pictus)* in the Serengeti Region, Tanzania. *Onderstepoort Journal of Veterinary Research* 60:415-420.

King A.A. (1993) (Editor) *Proceedings of the International Conference on Epidemiology, Control and Prevention of Rabies in Eastern and Southern Africa, Lusaka- Zambia, 1992.* Editions Fondation Marcel Merieux, Lyon.

Maas B. (1993) The behavioural ecology and social organisation of the bat-eared fox in the Serengeti National Park, Tanzania. PhD. thesis, University of Cambridge, U.K.

Macdonald D.W. (1993) Rabies and wildlife: a conservation problem? *Onderstepoort Journal of Veterinary Research* 60:351-355.

Magembe S.R. (1985) Epidemiology of rabies in the United Republic of Tanzania. In: *Rabies in the Tropic*. Edited by Kuwert E., Merieux C., Koprowski H. and Bogel K. Springer-Verlag, Berlin. pp. 392-398.

Rupprecht C.E., Dietzschold B., Wunner W.H. and Koprowski H. (1991) Antigenic relationships of Lyssaviruses. In: *The Natural History of Rabies. 2nd Edition* Edited by Baer G.M. CRC Press, Boca Raton. pp. 69-100.

Rweyemamu, M.M., Loretu K., Jakob H. and Gorton E. (1973) Observations on rabies in Tanzania. *Bulletin of Epizootic Diseases in Africa.* 21:19-27.

Smith J.S. (1989) Rabies and ectopic variation: use in ecologic studies. *Advances in Virus Research* 36:215-254.

Thomson G.R. and Meredith C.D. (1993) Rabies in bat-eared foxes in South Africa. *Onderstepoort Journal of Veterinary Research* 60:315-323.

Voigt D.R., Tinline R-R- & Broekhoven L.H. (1985) A spatial simulation model for rabies control. In: *Population dynamics of rabies in wildlife.* Edited by Bacon P.J. Academic Press, London. pp. 311-349.

Wandeler A.I. (1991) Carnivore rabies: ecological and evolutionary aspects. *Hystrix* **3**:121-135.

WHO (1992) World survey of rabies, no. 25. WHO/Rabies/92.203. World Health Organization, Geneva, Switzerland.

IMMUNISATION COVERAGE REQUIRED TO PREVENT OUTBREAKS OF DOG RABIES

Paul G Coleman [1]

Department of Medical Parasitology, London School of Hygiene and Tropical Medicine

Abstract

According to WHO guidelines, the critical percentage (P_c) of a dog population which needs to be vaccinated to prevent or control epidemics of rabies is 70 percent. The empirically derived value of P_c is based on observations worldwide between vaccination coverage and the subsequent decrease in rabies incidence. In this study, by contract we estimate P. by applying epidemic theory to data available from four outbreaks of rabies in urban and rural areas of Indonesia, Malaysia, Mexico and USA. The rate of increase of cases over the early stages of epidemics is used to estimate the basic case reproduction number (k) of rabies, which falls in the range 1.62-2.33. These estimates of R. may be used to calculate Pc which lies between 39 and 57 percent. It may also be shown, by considering the errors attached to these values of Pc, that the recommended immunisation coverage of 70 percent would be sufficient to prevent a major outbreak of rabies in at least 96.5 percent of occasions.

[1]Co-author: **C. Dye**, Department of Medical Parasitology, London School of Hygiene and Tropical Medicine. (corresponding author)

MULTI-SECTORAL COLLABORATION IN RABIES SURVEILLANCE AND CONTROL IN UGANDA

C.S. Rutebarika[1]

Department of Veterinary Services, Entebbe, Uganda

Introduction

The incidence of rabies in Uganda has remained high and the disease has been widespread throughout the 39 districts during the last two decades, due to the deteriorating political and social-economic situation. Rabies is most prevalent in dogs followed by domestic livestock and wildlife (see "Rabies in Uganda" p 17). Some of the factors which have contributed to the high incidence include:

- Human displacement and consequent uncontrolled, increased dog movement;
- Lack of dog population control;
- Insufficient vaccines and transport;
- No proper dog identification and care;
- Lack of natural barriers between Uganda and her neighbours;
- Insufficient public awareness and the consequent poor presentation of dogs for vaccination;
- Inadequate funding;
- Relaxation in enforcement of the laws governing rabies control.

Dog rabies control relies mainly on the mass immunisation of dogs. This has traditionally been carried out by the Department of Veterinary Services which procures rabies vaccines, distributes them and administers them to the dogs. The same Department is also responsible for destruction of stray and unvaccinated dogs and dog movement control. This measure is enforced through the Rabies and Animal Diseases Act.

During the last two decades, conventional veterinary rabies control measures have been largely ineffective in reducing the incidence of the disease to acceptable levels. Hence the need to establish and adopt a multi-sectoral approach (MSA) involving other sectors.

In 1991, a technical committee on rabies control (TECOR) was formed in response to the increased incidence of the disease. The committee comprises members from the Department of Veterinary Services, Animal Health Research Centre and Ministry of Health Veterinary Public Health Unit (VPH). The objectives of the committee are:

- To formulate an effective preventive and control strategy for rabies;
- To formulate rabies surveillance systems for data collection, analysis and mapping, with the support of the laboratory diagnostic services;

Co-author: **W. Kaboyo,** Veterinary Public Health Unit Ministry of Health, Entebbe, Uganda.

- To foster collaboration and co-ordination of rabies control programmes in the subregion.

Implementation of the multi-sectoral approach

A multi-sectoral approach involving other sectors notably the Veterinary Department, Health, Local Administration, Information, Youth and Culture, Internat Affairs and local communities has been adopted, initially in the three disei-c-ts of Masaka, Kabarole and Tororo(Figure 1) since late 1994. In future the programme will extend to six other districts and eventually the whole country.

The multi-sectoral approach is being implemented at the district and village levels in the surveillance and control of both human and animal rabies, with co-ordination at the national level through TECOR- The initial work has been supported by the YMO through the VPH Unit.

TECOR organises seminars for various sectors to be involved in rabies surveillance and control at the district level. The seminars address improved awareness and sensitisation of the community to the dangers of rabies, community participation, establishment of a viable district surveillance system and the formation of district-based multi-sectoral committees involving the relevant sectors.

Improved awareness and sensitisation of the community to the dangers of rabies

Rabies education packages are given. Audio visual aids such as pamphlets, posters, radio and television programmes and video fillms on rabies are used to educate the public. Recently a rabies film documentary was made in Masaka district to be used in other districts and on Uganda Television for improving community awareness of rabies.

Community participation

The communities are encouraged to participate in rabies control programmes, enacting local by-laws and enforcement of rabies laws, planning, publicising and implementing dog vaccination campaigns, dog registration and stray dog control, reporting rabies cases and ensuring that dog bite victims get first aid and treatment.

Figure 1. The districts (shaded) in which the multi-sectoral approach is being implemented.

Establishment of viable district surveillance systems

The local administrators, the communities and the district medical and veterinary staff co-ordinate rabies surveillance and facilitate data collection analysis and dissemination in the district. The data are later sent to the central co-ordinating committee at the national level.

Formation of district based multi-sectoral committees involving the relevant sectors

At the district level, co-ordination of rabies surveillance and control activities is vested in the district multi-sectoral committee comprising the following sectors: Veterinary, Health, Local Government and Local Council, Community Leaders, Internal Affairs, Representatives of Women and Youth. The roles of each sector are detailed in Table 1.

Table 1. The roles of the participating sectors in MSA to rabies control.

Sector	Activily					
	Dog registration and census	Stray dog elimination	Dog vaccination	Pre- and Post-exposure treatment	Community health education	Law enforcement
Veterinary	X	X	X		X	X
Health				X	X	
Internal Affairs (Police)		X	X			X
Local Government	X		X			X
Community Leaders	X	X	X		X	X

Viability of the multi-sectoral approach and future strategy

For each district, disease data, vaccination coverage and treatment costs before and after MSA implementation will be compared. Evaluation of accuracy of dog census and registration data, stray dog elimination and control efforts will be done. The cost benefit analysis of MSA versus the traditional approach will be undertaken. The MSA will eventually be adopted if there is positive evaluation. The overall goal will be reduction of rabies incidence in both man and animals.

The initial or pilot work conducted in the three districts of Masaka, Kabarole and Tororo is still in its infancy. The work done so far is very promising. It is hoped that eventually the rabies surveillance and control for the whole country will be improved. There is also need to involve wildlife specialists in TECOR.

The TECOR and Veterinary Department have interest in carrying out tests on bait delivery systems for oral vaccination of both domestic and stray dogs in the near future.

Finally, there is need to harmonise and synchronise rabies control programmes with neighbouring countries if meaningful reduction of rabies incidence and eventual eradication are envisaged.

TWO DAY RABIES VACCINATION CAMPAIGN: A SUSTAINABLE INTERVENTION?

K.K.LM de Balogh [1, 2]

School of Veterinary Medicine, Lusaka, Zambia

Abstract

Information concerning the planning and implementation of a two day rabies vaccination campaign organised in Lusaka, Zambia, is presented. Factors which contributed to its realisation and issues regarding the sustainability of such campaigns are discussed. Sustainability, for the purpose at hand, is defined as the recurrent (yearly?) execution of a vaccination campaign with a coverage of at least 70 percent to control the disease in a predetermined area. A key to success of the Lusaka campaign might have been that motivated veterinarians managed to secure required resources by mobilising and activating different institutions and organisations such as the School of Veterinary Medicine, Department of Veterinary and Tsetse Control Services, Information Service of the Ministry of Health, charitable organisations and Non Governmental Organisations. Free rabies vaccination campaigns appear to be feasible even with tight government budgets, by making use of an intersectorial approach, using resources efficiently and by focusing control activities in problem areas. In order to make such campaigns sustainable different interacting factors need to be addressed which are mainly related to organisation and management issues.

Introduction

Dog rabies is spreading in many countries mainly due to increasing density and mobility of human and dog populations (Bögel *el al.* 1982). In Zambia, dogs account for over 70 percent of all confirmed rabies cases (Hussein *et al.* 1984). In the capital city, Lusaka, most persons bitten by dogs attend the outpatient clinic of the University Teaching Hospital. During approximately one year (4 December 1991 to 13 November 1992), 962 bites were recorded. Sixty-five percent of these patients belonged to the male sex and most bites (80 percent) had been inflicted either by unvaccinated dogs or by dogs of unknown vaccination status prior to the vaccination campaign. If available, patients bitten by such dogs received a costly rabies post-exposure vaccine free of charge. Otherwise, they were advised to purchase the vaccine on the free market from pharmacies or private clinics.

[1]Co-authors: **F. Sabbe,** School of Veterinary Medicine, Lusaka, Zambia; **P.J. de Graaf,** Faculty of Agronomy and Forestry Engineering, EMU, Maputo, Mozambique.

[2]Present address: Faculty of Veterinary Medicine, EMU, Maputo, Mozambique.

As it has been recognised that the best way to reduce the rate of human exposure is by controlling the disease in the main host (Wandeler *et al.* 1988; WHO 1984) rabies vaccination of dogs and cats is compulsory in Zambia.

According to the World Rabies Survey (WHO 1992), a total of 78 785 dogs had been immunised in 1989 against rabies in Zambia. The vaccination coverage was estimated at 25 percent. Because of financial, logistical and organisational constraints the number of rabies vaccination campaigns conducted by the Department of Veterinary and Tsetse Control Services (DVTCS) have been insufficient to control the disease as a vaccination coverage of at least 70 percent is required (Beran 1991).

A dog population study was conducted in 1992 by the School of Veterinary Medicine in conjunction with the World Health Organization (WHO) and the DVTCS. The purpose of the study was to obtain information on the owned segment of the dog population and their accessibility to rabies vaccination (de Balogh *et al.* 1993). The information obtained was used as a baseline for the organisation of a massive two-day rabies vaccination campaign in Lusaka. Costs were reduced by using an intersectorial approach for the organisation of the campaign. The experience obtained from this campaign could be used for the planning and execution of future rabies vaccination campaigns in Zambia and elsewhere.

PREPARATIONS FOR THE CAMPAIGN

The financial constraints for the organisation of the vaccination campaign in Lusaka were overcome through the mobilisation of additional resources from charitable organisations and non-governmental organisations (NGOs). Personnel were recruited from the School of Veterinary Medicine (mainly veterinary students) and from the DVTCS. No specific funds had been allocated for this campaign by the Government of the Republic of Zambia. Money was raised from personal contributions, associations and donors to purchase equipment such as needles, syringes, disinfectants and paper for rabies certificates, and to cover the expenses of the rabies awareness campaign, including printing of posters and hiring of a vehicle with loudspeakers.

The plan to carry out a rabies vaccination campaign had been presented to a Lusaka based Rotary Club. The executive board of the club agreed to participate in the campaign as part of their community service activities. They provided transport for each vaccination team and financed packed lunches and a social activity at the end of the campaign as an acknowledgement. Since a very limited budget was available and the participants joined on a voluntary basis, it was opted for an intensive two day campaign.

The rabies vaccine had been imported by the DVTCS and was to be given free of charge during the campaign.

2. Timing and selection of vaccination areas

Prior to the selection of the vaccination areas, records on dog-bites reported to the University Teaching Hospital were evaluated. Almost all cases originated from the medium to low income areas which corresponded to the more densely populated areas of the city. High income, low density areas of Lusaka were not included in this vaccination campaign, as the dogs in these areas would be less at risk of contracting rabies. This was based on the assumption that most dogs in these areas were not able to leave the fenced premises and hence would have less contact with other dogs. Moreover, the owners are in a better position to take their dogs to veterinary clinics for vaccinations.

The vaccination campaign was scheduled for the 13th and 14th of September 1992. This date was selected as it was a week-end at the beginning of the new school term and still during the dry season.

From the previously conducted dog population study in Lusaka (de Balogh *et al.* 1993), a central point vaccination had proven to be more cost-effective in comparison with a door to door campaign for the urban area. In the low income, densely populated area of Lusaka where the dog population study had been carried out a dog to human ratio of 1:44 had been calculated. This ratio was used in conjunction with the 1990 human population census data to roughly estimate the dog population for each area. A larger number of dogs per inhabitants was expected to be present in the medium class areas. In the illegal squatter settlements less than 1 dog per 44 inhabitants was expected. The number of vaccination sites per compound were selected according to the estimated dog population for each area. A total of 10 000 dogs were estimated to live in the area to be covered by the campaign. Therefore a total of 45 vaccination sites were chosen. Schools, especially primary schools, were most frequently selected as vaccination sites. Other vaccination points were located in front of market places and at a shopping centre.

3. Manpower

The DVTCS provided part of the manpower. The students of the School of Veterinary medicine were encouraged to participate in the campaign allowing them to gain first hand experience in a disease control activity. A total of 13 vaccination teams consisting of 5 persons per team were formed with a total of 70 persons participating. Besides the representative of the Veterinary Department, each team contained one final year, one 5th year and one 4th year veterinary student, and a member of the Rotary club who assisted the team by transporting them to their respective vaccination sites. During the campaign, the general task-force of each team consisted of two persons vaccinating, two persons writing the certificates and one person organising the queues and assisting in controlling the dogs and preparing them for vaccination.

Each team was to be visited several times during the day by the organisers to monitor the activities, replenish the supplies of vaccine, certificates, needles and disinfectants and distribute the packed lunches.

4. Public information

Two weeks prior to the set date a rabies awareness campaign was launched with the assistance of the information Department of the Ministry of Health. Several interviews were arranged on television and radio programmes. Radio programmes, in which the public could call in and ask questions, were very informative, especially for the organisers, as misconceptions about rabies and related issues could be clarified. The costs involved in radio announcements could be avoided by disseminating the information through interviews in established programmes in English and in several local languages.

In the target areas, radio was considered the most effective medium for information dissemination as television and newspapers only reach those sectors of the public with a higher income. Nevertheless, interviews with newspapers were arranged and sponsors were found for advertising the campaign.

Through the District Education Officer, 43 schools were contacted to serve as vaccination sites in addition to two market places and a shopping centre. The headmasters of the schools were informed about the campaign and some background information was provided for dissemination among the pupils. It was further suggested to use the weekly parade to make the announcements. Posters were printed encouraging people to have their dogs vaccinated and indicating the time and location of each vaccination site. These posters were distributed through the schools to be displayed on their premises and at different public places in the area.

A vehicle with loudspeakers from the Information Service of the Ministry of Health made announcements of the campaign in the different target areas.

THE CAMPAIGN

A short briefing session was held for the participants on the morning of the first vaccination day. Issues concerning which dogs should be vaccinated (depending on their age, health status and previous vaccinations), methods of restraint and the issuing of a vaccination certificate, were presented. The danger of transmitting parvovirosis, distemper and other infectious diseases was emphasised during the briefing session. The limited availability of equipment such as needles and syringes demanded their re-use after a careful and proper disinfection with formalin and rinsing with distilled water.

A total of 9 174 dogs were vaccinated over the week-end at 45 different vaccination sites, in the low and medium income areas of Lusaka. Some monkeys were brought to the vaccination sites for rabies inoculation. As the vaccine was not adequate for this species they were not vaccinated. No cats were presented for vaccination.

On the first day of the campaign mainly the southern belt of Lusaka and a few locations in the north-east were selected. The second vaccination day covered the northern belt of Lusaka. The vaccination teams operated from 0900 - 1200 hours and from 1400 - 1630 hours. Most vaccination points functioned either in the morning or in the afternoon with the exception of some busy locations which remained operational during the whole day. More dogs were brought during the morning than during the afternoon and the number of dogs per location varied significantly. Up to 450 dogs were vaccinated in a day by one team whereas in other less busy locations only up to 45 dogs were vaccinated in one afternoon.

An average of 9.4 person-minutes was calculated for the vaccination of one dog and the issuing of a vaccination certificate. As the vaccinated dogs had not been marked, the vaccination coverage could not be quantified. Neither could the foregoing rabies awareness campaign be quantitatively assessed in its effectiveness but the response of the public indicated that the information must have reached large numbers of the target group. Apparently, the public welcomed the opportunity to have their dogs vaccinated free of charge near their homes. The long distances to the veterinary clinics and the costs involved were given as the main reasons which had previously withheld dog owners from having their dogs vaccinated.

It was observed that most of the dogs were brought to the vaccination sites by children, mainly boys. Only in a few cases were the dogs difficult to handle and the use of a dog catcher had to be used for restraint. However, dogs that were difficult to handle were most likely not brought to the vaccination points. Generally the public was co-operative in allowing an orderly execution of the vaccinations, by keeping in a line and avoiding dog fights.

The total costs for the printing of posters, information material and vaccination certificates as well as for equipment food and drinks were calculated at 250 .000 Zambian Kwacha (exchange rate in 1992 was US$ 1.00 = ZK 200). This amount does not include the cost of the vaccine. Costs for manpower can be neglected as all participants joined on a voluntary basis.

DISCUSSION

Control of rabies in developing countries is directly related to the management of the dog population (Joshi and Bögel 1988). Previous experiences of rabies vaccination campaigns in other parts of the world have indicated that the success of such campaigns depend on the willingness of dog owners to have their dogs vaccinated, government commitment and the availability of resources, qualified personnel and infrastructure to organise such campaigns (Escobar 1988). These key issues with relation to the execution of the campaign and sustainability are briefly addressed. Sustainability, for the purpose at hand, is defined as the recurrent (yearly?) execution of a vaccination campaign with a coverage of at least 70 percent to control the disease in a predetermined area.

For the realisation of rabies control activities, there has to exist a clear commitment at the national level with the appropriate legislative support. Rabies control cannot be self-supporting and requires a certain degree of government support. Government commitment could be assessed by the quantity of funds made available and its commitment not to divert such funds for other disease control activities. Unfortunately, the budgets of developing countries are usually constrained and often insufficient funds can be made available for rabies vaccination campaigns. Hence, additional funds must be secured. The 'Lusaka experience' showed that this is not too difficult.

Small amounts of money could easily be obtained through informal channels without putting to action lengthy public relations mechanisms. NGOs, international donors and the private sector were motivated to contribute to the campaign as they recognised the importance of controlling rabies. These organisations have substantially contributed to the success of the campaigns in Lusaka. Their funds were used to purchase consurnables and information material. In fact most of the funds were acquired outside any regular budget.

The continuity of rabies control has not only failed due to deficient government funding but also due to a lack of clear designation of responsibilities. An institutional strategy is called for in which responsibilities are clearly defined and described. Rabies control is often one of the many disease control activities designated to a person within a ministry. The appointment of a Rabies Control Officer would be very desirable. Such an officer could manage the organisation of rabies vaccination campaigns and liaise with the different institutions involved, improve the forwarding of samples from suspected rabid dogs, the feedback of results to the field and the organisation of training courses and awareness programmes. Unfortunately, the creation of a new position in the government structure is a long process.

As an alternative, the organisation of local vaccination activities could be seconded to institutions outside of the DVTCS such as Veterinary faculties, Veterinary/Animal Husbandry Training Institutions and Veterinary Research Institutes. In such institutions rabies working groups could be initiated. Veterinary students, extension workers, technicians and qualified veterinarians would regularly participate in the vaccination activities. Further, these working groups could more readily initiate rabies related research such as dog ecology studies, surveys on vaccination coverage and development of information material.

Community oriented organisations are often enthusiastic to contribute to such campaigns either in kind or by providing manpower. Non-veterinary/health personnel such as the police, military, students and school teachers have also been successfully integrated in massive one day rabies campaigns in Brazil (Belotto 1988).

In any campaign the participation of the local community is a crucial factor for achieving the desired results. It is believed that the high degree of participation in Lusaka was mainly due to:

- a targeted intensive public awareness campaign preceding the actual vaccination campaign indicating all advantages of having dogs vaccinated and informing the public about rabies in general
- provision of free vaccination of dogs against rabies
- strategic selection of vaccination sites near the homes of the target group
- the issuing of vaccination certificates
- the timing of the campaign

Initiatives to organise large rabies vaccination campaigns without issuing rabies vaccination certificates are questionable, as the public is generally aware of the importance of the certificate as it proves the validity of the vaccination. Although fraud with false certificates does not seem to occur in Zambia, alternative methods to clearly identify vaccinated dogs should be considered. The use of a durable collar, issuing of a tag or the use of permanent dye to facilitate the identification of the vaccination status of the dog in case it has bitten somebody are to be assessed.

If persons and vehicles of different sectors are to be involved in the execution of a massive campaign, short intensive vaccination periods are called for. This ensures motivation and will avoid interference with their regular activities (Belotto 1988). Implementing massive campaigns on a yearly basis and on a fixed date could largely reduce the costs of the preceding information campaign once it becomes institutionalised.

In Zambia, dogs are cared for mostly by the young boys in the household and most dogs were brought by them to the vaccination points. The use of schools for the dissemination of information is often used in Zambia. In this case the information about rabies directly reached one of the main target groups, school age children. Furthermore, the main advantages of using schools as vaccination sites are:

- they are well known locations
- the main target group, young boys, are all gathered together
- facilities (desks, chairs, water) are available
- they are generally accessible and parking facilities are often available

The main disadvantages are:

- the vaccination points should be operational outside of school hours, for example, over week-ends. This, however, does not coincide with the working hours of most civil servants.
- dogs may soil the school premises.

Schools were found very suitable vaccination sites as compared to, for example, market places. Although markets are also well known to the public these are considered less suitable due to their size and by having various entrances. Furthermore., the presence of big crowds had a disturbing effect on the orderly execution of the vaccinations.

An alternative to be considered would be to organise these campaigns during the school term and to execute the vaccinations at the beginning of the holidays.

Vaccination campaigns should be conducted during the dry season, when roads are easily accessible and the risk of rains preventing people arriving at the vaccination sites is absent. After each campaign the suitability of the vaccination site should be evaluated.

It may be clear that under the prevailing unstable economic and institutional conditions in many developing countries sustainability remains difficult to foretell and continuous debates are required. Based on the Lusaka experience it seems that the following factors enhance successful implementation of a campaign and may to a certain extent contribute to sustainability:

- some motivated initiators
- recurrent involvement of NG0s, charitable organisations and the private sector to secure financial inputs
- effective public awareness campaigns
- participation of the target group
- the issuing of a 'formal document' (certificate)
- the timing and area selection
- a certain degree of government commitment to assure the regular organisation of such campaigns
- the guaranteed supply of vaccines

A key to success might have been that motivated veterinarians managed to secure required resources by mobilising and activating different institutions and organisations such as the School of Veterinary Medicine, Extension Services of the DVTCS, Information Service of the Ministry of Health, charitable organisations and NGOs.

Free rabies vaccination campaigns appear to be feasible even with tight government budgets by making use of an intersectorial approach by concentrating resources (Bögel and Meslin 1990) and focusing controlled activities in problem areas. An evaluation of the campaign is required to readjust strategies. In order to make such campaigns sustainable different interacting factors need to be addressed which are mainly related to organisation and management issues.

REFERENCES

Balogh, de K.K.I.M., Wandeler A1, Meslin R-X. (1993) A dog ecology study in an urban and semi-rural area of Zambia. *Onderstepoort Journal of Veterinary Research* 60:43 7-443.

Belotto A.J. (1988) Organisation of a mass vaccination for dog rabies in Brazil. *Review of Infectious Diseases 1* O(S4): S693 -696.

Beran G.W. (199 1) Urban rabies. In: *7he Natural History of Rabies, 2nd Edition.* Edited by Baer G.M. CRC Press, Boca Raton, pp.427-443.

Bögel K., Andral L., Beran G., Schneider L.G. and Wandeler A. (1982) Dog Rabies Elimination. *International Journal of Zoonoses* 9:97-112.

Bögel K. and Meslin RX. (1990) Economics of human and canine rabies elimination: guidelines for programme orientation. *Bulletin of the World Health Organization* 68:381-291.

Escobar Cifuentes E. (1988) Program for the Elimination of Urban Rabies in Latin America. *Review of Infectious Diseases* **10**(S4): S689-692.

Hussein NA, Sharma RX, Ando R., Chizyuka H.G.B. (1984) Further review of the epidemiology of rabies in Zambia (1975-1982), *Revue Scientifique et Technique de I'Office International des Epizooties* 3(l):125 -13 5.

Joshi D.D., B6gel K. (1988) Role of Lesser Developed Nations in Rabies Research. *Review of Infectious Diseases* **10**(S4): S600-603

Wandeler A1, Budde A., Capt S., Kappeler A., Matter H. (1988) Dog Ecology and Dog Rabies Control. *Review of Infectious Diseases* **10**(S4): S684-688.

WHO (1984) WHO Expert Committee on Rabies, 7th Report. WHO Technical Report Series 709, World Health Organization, Geneva.

WHO (1992) World Survey of Rabies 25 (for year 1989). WHO/Rabies/92.203. World Health Organization, Geneva.

RABIES CONTROL IN DOGS IN ZIMBABWE

W.N. Madzima

Department of Veterinary Services, Harare, Zimbabwe

Introduction

Rabies was first diagnosed in the country in 1902 (Sinclair 1922) and its presence continued until 1913. Between 1913 till 1950 there were no outbreaks of rabies, except for two dogs imported from Zambia which developed the disease while still in quarantine in 1938 (cited by Swanepoel and Foggin 1978). The apparent disappearance of the disease may have been partially due to a low dog population at the time.

On 6 September 1950 rabies re-emerged in the Beitbridge area and rapidly spread into Masvingo and Bulawayo areas such that by 1951 there had been 117 cases in domestic animals. The disease was present in neighbouring northern Transvaal and Botswana, from where infection is believed to have spread. Vaccination of dogs was initiated in 1950, but by 1952 the disease was also diagnosed in wild carnivores.

During the first ten years after which vaccination had been legislated, 1.1 million dogs were vaccinated and eradication of rabies seemed a realistic objective (Foggin 1988). However, a combination of two factors resulted in a vicious cycle which precluded the eradication of the disease, namely, the periodic fall in the number of dogs presented for vaccination and the resultant spill-over of infection into the jackal population. This situation was always exacerbated by cross-border transmission of the disease. As a result the average incidence continued at approximately 120 confirmed cases per year until 1976, as shown in Figure 1. Also shown in Figure 1 is the sudden increase in the number of rabies cases around 1978 that followed a sudden drop in the number of dog vaccinations. This epidemic co-incided with a period of civil disturbances that culminated in independence in 1980. Thereafter, the epidemic was effectively brought under control and although dog vaccinations have progressively improved there has not been a corresponding decrease in the number of cases. Other factors such as other major disease outbreaks have put a strain on available manpower and transport resources and periodically impacted negatively on the extent of vaccination coverage. This was the case in 1989 during a major foot-and-mouth disease outbreak and again in 1994 when the country experienced a major outbreak of Newcastle disease.

Epidemiology of Dog Rabies in Zimbabwe

The incidence of rabies in dogs appears to be influenced by a number of closely linked factors, chief among which are percentage vaccination coverage, land use, human/dog population density, jackal population and proximity to international borders.

As shown in Figure 1, a good vaccination coverage has been followed by a drop in the number of rabies cases, irrespective of the trend in the incidence of the disease at the time. In 1986 Brooks (1990) estimated that there were 1.3 million dogs in Zimbabwe of which 90 percent were in the communal (subsistence) farming areas. The highest coverage was in 1991 when just over 500 000 (3 8 percent) dogs were vaccinated in communal farming areas.

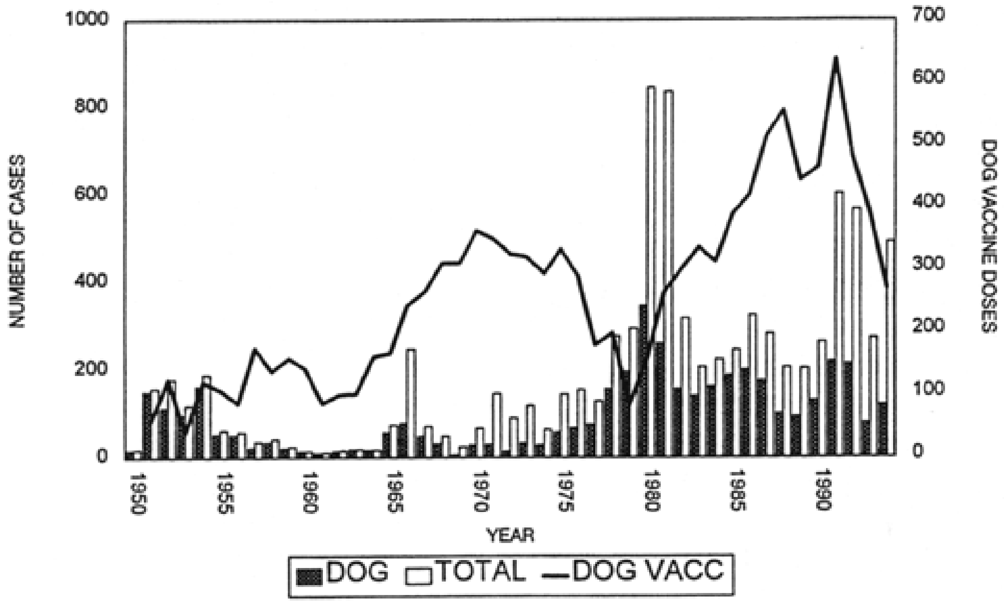


Figure 1. Levels of rabies and rabies vaccinations in Zimbabwe since 1950, showing total cases, dog cases and number of dogs vaccinated.

The incidence of rabies also varies according to land use. Approximately 75 percent of confirmed rabid dogs originate from commercial fanning areas and urban areas (Foggin 1988), perhaps more a reflection of better knowledge about the disease, hence better reporting. However, it is believed that there are more cases of rabies in communal farming areas but there is under-reporting. This is evidenced by the increasing trend of confirmed cases in spite of an increasing vaccination coverage.

Communal farming areas are generally over-populated and have poor vegetation cover which makes them less suitable as a habitat for jackals, but they have a high population of roaming unconfined dogs. Thus, dog rabies is the problem of communal farming areas. In contrast, commercial farming areas have abundant vegetation cover and account for 87 percent of cases of jackal rabies. The presence of jackals results in a self-sustaining cycle of infection between dogs and jackals. Thus, although dog vaccination coverage is considered good in commercial farming areas, it is apparently not high enough to prevent maintenance of the dog-jackal rabies cycle.

Rabies in dogs occurs throughout the country (see "Rabies in Zimbabwe" page 53), but over the years there have been more cases along the eastern border, presumably as a result of migration of dogs from Mozambique.

Since 1950 to date, approximately 47 percent (4287 out of 9091) of all confirmed rabies cases were in dogs followed by jackal rabies (24 percent) and cattle rabies (19 percent). Based on figures collected between 1987 till 1991, 58 percent of positive rabies cases in dogs involved human contact followed by cattle (42 percent) and jackals (38 percent) (Bingham 1993). Thus, from a public health point of view, dog rabies is of greatest significance. Cattle rabies is usually associated with bites by rabid jackals, but in communal farming areas dog bites also play a role.

Control of Rabies in Dogs

Control policy

The Animal Health (Rabies) Regulations, 1966, make it compulsory for an owner of a dog to have it vaccinated against rabies within one month of attaining the age of three months, followed by a second vaccination at 12-15 months of age and within every three years thereafter. Alternatively, if anyone acquires a dog whose vaccination history in not known, the dog must be vaccinated within seven days of its acquisition and again after six months and within every three years thereafter. In the Animal Health (General) Regulations, 1994, rabies is a notifiable disease and as such it must be reported to the Department of Veterinary Services. In the event of a serious rabies outbreak, the Director of Veterinary Services may declare any place as a rabies area by issue of an Animal Health (Rabies Areas) Order. The order enables veterinary staff to take additional measures to control the outbreak, such as the issuing of tie-up orders in affected areas and destruction of dogs which are not secured or closely confined.

Additionally, the Animal Health (Import) Regulations, 1989, require that, before bringing a dog into Zimbabwe, a veterinary import permit is obtained, subject to ' certain conditions as the Director of Veterinary Services may impose. In the Southern African Development Community (SADC) the conditions of importation have been standardised, thereby facilitating movement of dogs and cats between member countries.

Vaccination strategy

In urban areas dog owners may produce their dogs for vaccination on any day during working hours at Government Veterinary Offices or private veterinary surgeries. Additionally, Government Veterinary Offices have set aside every Friday afternoon as a time when owners can bring their dogs for vaccination by a team on standby for the purpose. The Department routinely mounts vaccination campaigns annually at dipping tanks throughout the communal farming areas. A similar mass vaccination campaign is carried out annually at designated points at shopping centres in urban areas. The annual vaccination campaigns are necessitated by the high turn-over in the dog population as a result of high mortality in puppies (parasites, disease) and uncontrolled breeding, especially in communal farming areas and among dogs of the urban poor.

At every vaccination, every dog receives a tattoo bearing that year's unit number, initially in the left ear, and subsequently in the right ear. If the owner requires a certificate of vaccination, a fee of Z$3,00 is charged. Thus, in general, vaccination by government is carried out free as a contribution to public health.

Rabies vaccine

The vaccine used in dogs consists of inactivated cell culture vaccine. All the vaccine used in Zimbabwe is imported. There are several proprietary rabies vaccines on the local market which must all be registered with the Drugs Control Council in terms of the Drugs and Allied Substances Control Act and its regulations. The Department of Veterinary Services procures vaccine through an annual government tender. Rabies vaccine is only imported by the Department of Veterinary Services who sell to veterinary private practitioners. This measure assists in determination of the amount of rabies vaccine used in the country.

Reporting of human contacts

All dog-bites must be reported to veterinary staff who will promptly check whether the dog in question has an up-to-date vaccination history. In the event that this is not the case, the dog is detained for observation for a period of at least seven days for signs of the disease to appear. In the meantime the person who has been bitten is referred to a hospital for treatment.

Client Education on Rabies

During mass vaccination campaigns against rabies, opportunity is taken to educate the public through the distribution of leaflets and the use of a loud-hailer mounted on a mobile unit. The mobile unit also makes use of films on rabies, which are shown at agricultural shows and at schools. Posters on rabies are also displayed in every Veterinary Office, veterinary surgery and human clinic. Additionally, veterinary staff go on radio and television, and issue articles in papers and magazines.

References

Bingham J. (1993) Rabies in Zimbabwe. In: *Proceeding of the International Conference on Epidemiology Control and Prevention of Rabies in Eastern and Southern Africa, Lusaka~ Zambia, June 2-5, 1992.* Edited by King A. Fondation Marcel Merieux, Lyon, France.

Brooks R. (1990) Survey of the dog population of Zimbabwe and its level of rabies vaccination. *Veterinary Record* **127**:592-596.

Foggin C.M. (1988) Rabies and rabies-related viruses in Zimbabwe: Historical, virological and ecological aspects. Doctor of Philosophy thesis, University of Zimbabwe.

Sinclair LM. (1922) A short history of the infectious diseases amongst domestic animals of Southern Rhodesia since the occupation. *Rhodesian Agricultural Journal 19:21-22*.

Swanepoel R- and Foggin C.M. (1978): The Occurrence, Diagnosis, Treatment and Control of Rabies in Rhodesia. *Central African Journal of Medicine 24:* 107-115.

PROGRESS IN ORAL VACCINATION OF WILDLIFE IN EUROPE

Arthur A. King

Formerly, Central Veterinary Laboratory, Weybridge, U.K

One may be forgiven for asking of what relevance oral vaccination of wildlife in Europe is to the southern African scene when, as we have heard today and in previous meetings, the region's most pressing problems are associated more with diagnosis and control of rabies in dogs than with control of rabies in wildlife. My response would be that should we succeed in controlling or eliminating canine rabies, whilst rabies exists in wildlife the possibility of re-introduction of the disease into dogs and other domesticated species remains and the cycle may begin all over again. Many of you are, of course, familiar with the principles of oral vaccination and indeed have a working knowledge of what is happening in Europe. In the limited time available for this talk I shall thus try to bring you up to date by reference to published figures and to attempt where possible, to draw comparisons with the situation in the region and to define one or two of the problems that may arise when wildlife rabies control in the region becomes a possibility.

Firstly, a few general points about rabies in animals. The majority of rabies hosts are small to medium-size carnivores that scavenge for food and/or prey on rodents, other small vertebrates and invertebrates. All have high intrinsic population growth rates, thus permitting rapid recovery from severe losses due to disease; this regenerative ability ensures survival of the species and continuation of a rabies epizootic despite high case density within it (Wandeler 1991). In Europe, the red fox is the principle vector and victim - where fox rabies is eliminated no other terrestrial animal cases are recorded. Within the southern and eastern African region, however, we have not one but several wildlife reservoirs of infection, notably jackals (three species), bat-eared foxes and mongooses (several species).

Secondly, disease surveillance in Europe is of a very high standard - almost all of the cases to which I shall refer have been confirmed as rabies by laboratory techniques, almost exclusively fluorescent antibody tests. This is not the case in Africa. Generally speaking, the more southerly countries report more cases in wildlife - whether this is a true reflection of the weight of disease, or is related to the lower levels of surveillance in more northerly countries of the region, is not clear; in the latter group, positive cases are not always confirmed by laboratory tests.

Thirdly, all of the vaccines and baits used in European oral vaccination programmes have been "tailored" to suit the target species (the fox) and although non-target species undoubtedly take a proportion of the baits, very few cases of vaccine-induced rabies have been recorded, despite the fact that a large number of field cases have been analysed for vaccine virus. This is not so in Africa. In experiments, one of the vaccines widely used in early vaccination trials in Europe (SAD_{Berne}) killed two of four wild-caught chacma baboons in which the vaccine was instilled (Bingham *et al.* 1992). The vaccine is therefore unsuitable for deployment in areas where these baboons occur. This is particularly disappointing since earlier experimental studies (though published later, (Bingham *et al.* 1995)) had shown the vaccine to be highly efficacious in the two species of jackal important in rabies transmission in Zimbabwe.

Fourthly, the vaccine of choice has to be packaged in a bait which will be preferentially taken by the target species. In Europe, a variety of shapes and sizes of baits has been used, ranging from chicken heads in early trials to fish-meal polymer baits now widely used; they are either laid by hand or, again more recently, dropped from helicopters. These baits are highly attractive to foxes. Unfortunately, some so far unpublished studies (Bingham, pers.comm.) have shown that jackals are not enamoured of baits which may smell of fish, but they do like chicken heads!

This leads us to the distribution of vaccine-laden baits. Hand-laying is hugely expensive unless "volunteers" can be found and it is time-consuming, bearing in mind that more than one bait-laying campaign will be required. Helicopter drop costs are not insignificant but in Europe the method has allowed coverage of vast geographical areas with bait. It works for foxes in Europe; in Zimbabwe, however, it may be that baits will have to be laid by hand and in groups, since jackals will often tend to travel in pairs or small groups and the dominant individuals may take many of singly laid baits (J. Bingham, pers.comm).

Of course, for a campaign to be successful a large percentage of the target species needs to be vaccinated in order to create a situation whereby an infected animal (which would eventually die of the disease) would come into contact only with immune animals and thus not be able to pass on the disease. Some indication of whether or not an animal has consumed a bait (and therefore immunised itself) is desirable and although there are several options available, the most frequently used technique is the incorporation of tetracycline in the bait. Once an animal has consumed a bait the tetracycline is laid down in teeth and bone and can be demonstrated by diamond-saw sections of either. Using fluorescence, the number of baits consumed by an animal can be counted - rather like counting the annular rings in a cross-section of the trunk of a tree. This method works very well for foxes in Europe. However, in Zimbabwe it has been shown that there are naturally occurring tetracycline-like compounds in jackal femurs, with a prevalence high enough (over 30 percent) to interfere with interpretation of baiting campaigns (Bingham *et al.* 1994).

The European fox rabies epidemic began at the Russo-Polish border after the Second World War and moved centrifugally in all directions at an annual rate of about 30-40 km. The disease entered northern West Germany from East Germany in 1950 and central West Germany in 1960; it had reached Switzerland by 1967 and France by 1968. Early control methods centred upon attempts to diminish the fox population by a variety of methods including hunting, poisoning, gassing and trapping. The overall success of such methods was highly controversial, since they were relatively non-selective and lacked public support for methods which decimated wildlife populations.

The death toll from the disease has been enormous. Taking only four of the countries affected (Germany, Switzerland, France and Belgium), with a land area of 0.98 million square kilometres, during the 1980's 112 753 laboratory confirmed rabies cases were recorded (Table 1). Nearly 83 000 of these cases were foxes. This figure, however, represents only a fraction of the true death-toll in the host species, since it has been estimated (Blancou *et al.* 1988) that only 10-20 percent of foxes which die from the disease in the wild are submitted for diagnostic tests. Putting these figures into context with other rabies epidemics, in a similar ten year period (1983 - 1992) over the 9.3 million square kilometres of the USA, where skunks, raccoons, foxes and bats are reservoir species, 57 417 cases were recorded (Krebs *et al.* 1994); over the 1.2 million square kilometres of South Africa, where dogs, jackals, bat-eared foxes and mongooses are the reservoir species, during the 1980s, only 43 15 cases were recorded (Swanepoel *et al.* 1993).

Table 1. Reported Rabies Cases in Germany, Switzerland, France and Belgium 1980 -1989

	Domestic Animals				Wildlife Animals					Total
	Dog	Cat	Other	Total	Fox	Deer	Badg	Other	Total	
Germany	1375	2959	7714	12048	56027	3892	1241	2951	64111	76159
Switzerland	48	423	631	1102	4485	231	308	284	5308	6410
France	519	986	3627	5132	19844	188	254	465	20751	25883
Belgium	51	181	1473	1705	2471	23	35	67	2596	4301
Total	1993	4549	13445	19987	82827	4334	1838	3767	92766	112753

Since 1985-86, the number of European countries, participating in control programmes has increased to 14, plus Russia, Estonia and Belarus. Over 70 million vaccine-baits, costing US$ 83 million have been distributed over an area of 4.5 million square kilometres (Stöhr 1994), an area which approximates in size to half that of the USA, or to that of South Africa, Namibia, Botswana, Zimbabwe and Mozambique together.

A number of vaccines have been used, commencing with the SAD_{Berne}, first used in chicken head baits in Switzerland in the mid-1970's, SAD B19 used in Germany and elsewhere and more recently, SAG-2 (a derivative of SAD_{Berne} in which a two-stage neutralisation of the virus using anti-G Mabs yields a vaccine which differs from the parent virus by one amino acid in position 333 but by two nucleotides from any of six possible triplets encoding for arginine at that site (Schumacher *et al.* 1993)). Lastly, a genetically engineered vaccinia-rabies glycoprotein (V-RG) recombinant vaccine, which has been shown to elicit high antibody levels when given parenterally to a wide variety of species (Rupprecht and Kieny 1988) has also been used widely.

The rabies incidence has fallen markedly in all countries where vaccine baits have been distributed, although no country so far has succeeded in eradicating the disease and remaining rabies-free for a long period. Examples of progress are shown in Table 2. Two points become immediately obvious - a dramatic fall in the incidence to 1993 in Germany, France and Belgium and a rise in incidence in 1994 in Germany, Switzerland and Belgium.

Although the rise in incidence is disappointing, one needs to plot the cases on a map to see what is actually happening. In all of the four countries, vast areas have been totally cleared of rabies. The increase of numbers is due to high incidence in a few locations - often associated with countries' borders with other rabies endemic countries. These locations will be subjected to further intense vaccination campaigns during 1995 and beyond. The prospect of a rabies-free western Europe is within sight.

Table 2. Reported Rabies Cases in Germany, Switzerland, France and Belgium 1989 - 1994

		Domestic Animals				Wildlife Animals					Total
		Dog	Cat	Other	Total	Fox	Deer	Badg	Other	Total	
Germany											
	1989	163	329	754	1246	4855	316	128	278	5577	6823
	1990	192	267	523	1082	3937	242	94	216	4489	5571
	1991	153	189	282	624	2665	130	48	132	2975	3599
	1992	59	77	138	274	1011	56	24	60	1151	1425
	1993	6	25	110	141	636	28	16	24	704	845
	1994	5	28	201	234	1044	48	28	24	1144	1378
Switzerland											
	1989	0	0	2	2	56	0	2	0	58	60
	1990	0	0	1	1	24	0	0	0	24	25
	1991	0	1	1	2	95	0	2	6	103	105
	1992	0	3	2	5	103	1	16	2	122	127
	1993	1	4	12	17	143	1	11	2	158	175
	1994	2	3	20	25	167	5	26	2	200	225
France											
	1989	53	117	554	724	3341	28	35	86	3490	4214
	1990	50	82	331	463	2406	19	37	59	2521	2984
	1991	38	83	269	390	1663	24	23	45	1755	2165
	1992	30	49	138	217	1000	16	16	35	1067	1284
	1993	4	11	26	4	198	1	6	15	220	261
	1994	2	4	14	20	73	0	1	5	79	99
Belgium											
	1989	12	33	250	295	520	6	8	13	547	842
	1990	2	11	66	79	56	0	3	6	65	144
	1991	0	1	9	10	19	0	0	0	0	29
	1992	0	0	17	17	17	0	0	0	17	34
	1993	0	1	0	1	0	0	1	0	1	2
	1994	0	1	17	18	43	0	0	0	43	61

Already benefits are beginning to accrue. Fewer dog and cat cases result in a reduction of need for human post-exposure treatments. Although the disease has not caused livestock losses of serious economic proportions, no loss to rabies is another advantage and freedom from rabies in western Europe will eventually allow free movement of animals without the need for quarantine.

In this paper I have outlined the progress in oral vaccination in Europe and made comparisons where possible with the situation in the southern and eastern African region. Is it possible that oral vaccination could be used here? The time, perhaps, is not yet, there is a more pressing need to control canine rabies. But, in a country such as the host nation for our conference, which has an excellent record of canine rabies control in the past and where the jackal appears to be the main if not the only wildlife reservoir of infection, it is appropriate that a start should be made. John Bingham and his colleagues have already carried out much of the groundwork and it is to his laboratory here in Harare that we shall be looking for advancement.

References

Bingham J., Foggin C.M., Gerber H., Hill F.W.G., Kappeler A., King A.A., Perry B.D. and Wandeler A.I. (1992) Pathogenicity of SAD rabies vaccine given orally in chacma baboons *(Papio ursinus). The Veterinary Record* 131:55-56.

Bingham J., Matema R., Kappeler A. and Hill F.W.G. (1994) Naturally occurring tetracycline-like fluorescence in sections of femur from jackals in Zimbabwe. *The Veterinary Record* 135:180-182.

Bingharn J., Kappeler A., Hill F.W.G., King A.A., Perry B.D. and Foggin C.M. (1995) Efficacy of SAD (Berne) rabies vaccine given by the oral route in two species of jackal *(Canis mesomelas* and *Canis adustus). Journal of Wildlife Diseases* 31:416-419.

Blancou J., Pastoret P.-P., Brochier B., Thomas I. and Bögel K. (1988) Vaccinating wild animals against rabies. *Revue Scientifique et Technique de l'Office International des Epizooties* 7:1005-1013.

Krebs J.W., Strine W., Smith J.S., Rupprecht C.E. and Childs J.E. (1994) Rabies Surveillance in the United States during 1993. *Journal of the American Veterinary Medical Association* 205. (15 December 1994).

Rupprecht C.E. and Kieny M.-P. (1988) Development of a vaccinia-rabies glycoprotein recombinant virus vaccine. In: *Rabies.* Edited by Campbell J..B. and CharIton K.M. Kluwer Academic Press. pp. 335-380.

Schumacher C.L., Coulon P., Lafay R, Bénéjean L, Aubert M.F.A., Barrat J., Aubert A. and Flamand A. (1993) SAG-2 oral rabies vaccine. *Onderstepoort Journal of Veterinary Research* 60:459-462.

Stöhr K. (1994) Oral immunisation of foxes in Europe: Why does it take so long? In: *Fifth annual international meeting on rabies in the Americas, Niagara Falls, Ontario, Canada, November 16-19 1994* (in press).

Swanepoel R, Barnard B.J.H., Meredith CD, Bishop C.G. Brückner G.K., Foggin C.M. and Hilbschle 0.J.B. (1993) Rabies in southern Africa. *Onderstepoort Journal of Veterinary Research* 60:325-346.

Wandeler A.I. (1991) Carnivore rabies: ecological and evolutionary aspects *Hystrix* 3:121-135.

EVOLUTION OF WHO RECOMMENDATIONS ON ORAL IMMUNISATION OF DOGS AGAINST RABIES

F.-X. Meslin

Veterinary Public Health Unit, World Health Organization, Geneva, Switzerland

Although improvements in rabies trends have been reported recently, no dog rabies-infected country has become free in the last two years and no successful dog rabies elimination programmes have been implemented during the past 10 years in spite of the availability of improved methods of surveillance and control and in spite of the increasing health significance of the disease in many countries. The reasons for not achieving an appropriate vaccination coverage of most dog rabies control programmes in developing countries are manifold but two major obstacles should be mentioned: 1) the lack of sustainable resources allocated to rabies control 2) where appropriate resources are available, the difficulty of reaching a large enough proportion of dogs for parenteral vaccination.

Although in many areas a high vaccination coverage can be achieved using the parenteral route, a significant proportion of the dogs cannot be reached by parenteral vaccination i.e. dogs aged less than two to three months, ownerless dogs, owned dogs found in or near the household which cannot be caught and uncatchable free roaming owned dogs. When these categories are put together it represents about 15 to 25 percent of the total dog population in Tunisia whereas it represents between 40 and 50 percent in a village in Turkey (Matter 1989; Matter and Fico 1992; Matter *et al.* 1993).

It was shown also that dog vaccination at permanent vaccination centres (usually local veterinary offices) was very ineffective and could not lead to a coverage much higher than 10 percent. Vaccination from mobile centres was more effective and could lead. as mentioned before, to a coverage ranging from 40 to 85 percent. Door-to-door vaccination in villages of the western part of Turkey may not lead to a much higher coverage than mobile centres. However this vaccine delivery tactic, although time consuming, may allow a very high proportion of the dogs in Tunisia, in most Latin American countries and probably in many African and Asian countries to be reached (Meslin *et al.* 1994).

The first WHO consultation dealing specifically with the concept of oral vaccination of dogs (OVD) was held in Geneva in February 1988 (WHO 1988). This consultation which analysed the causes of recent failures of dog rabies control projects, led to a clear vision of the potential contribution of the OVD concept to rabies control and elimination by stating: "OVD may offer new strategies and approaches permitting a significant increase in the dog vaccination coverage (especially of free-roaming and poorly supervised dogs) both when applied exclusively or in combination with parenteral immunisation".

Three delivery systems for OVD could be envisaged (Matter 1992):

(a) the distribution of the baits to owned dogs via their owners who would collect the bait at a central location;

(b) the placement of baits in selected spots where they can be picked-up by free-roaming dogs (so called "wildlife immunisation model") and
(c) distribution of baits to dogs encountered in the streets (so called "hand-out" model).

The first technique ideally requires a vaccine safe for humans and thermostable enough to be distributed to dog owners without any further supervision. This technique, if most dog owners can be motivated, should lead to a high coverage of the dog population (circa 90 percent if dogs less than three months old can also be immunised) which would be independent of whether the dog can or cannot be restrained by its owner and whether it is permanently confined to the household or partially free-roaming.

Deposition of the baits in selected areas would target mainly ownerless and free roaming owned dogs and its efficacy would be dependant upon the respective proportions in the dog population of these categories of dogs. It can be hypothesised that this technique would be more effective in rural areas where a higher proportion of the dogs are free (expected coverage 85 percent) than in sub-urban zones (expected coverage 40 percent). Deposition of baits over large areas especially near or within densely populated human settlements might be neither feasible nor safe as observed in Tunisia in 1994.

With regard to the "hand-out" delivery technique, according to the results acquired in a limited area in Tunisia, if the baits are handed out to free roaming dogs encountered in a given area, one should not expect to reach more than 30 percent of the total dog population living in the area. It should however be noted that even if the proportion of dogs newly vaccinated by these methods ("wildlife immunisation" and "hand out" models) is small, the increase of the immunisation coverage resulting from their application may be critical to achieving the disease elimination target.

The oral vaccination technique, whatever delivery system is selected, might become an essential complement of parenteral vaccination or the sole immunisation technique used in all places where there is a large number of unrestrainable owned dogs (e.g. Turkey) or a high percentage of feral dogs which cannot be eliminated over the long term (e.g. Yemen).

Since 1988 WHO has continuously promoted international collaboration and co-ordinated research in OVD through an informal group of specialists associating specialised WHO collaborating centres, researchers and official representatives of potential recipient countries as well as pharmaceutical companies (WHO 1989; 1991; 1992; 1993a; 1993b; 1994).

Very early on it became evident to the group that safety of the OVD concept (from candidate vaccines to bait and bait delivery systems) under the specific conditions prevailing in most dog rabies areas. was a prerequisite to promoting its use in the field. Vaccine safety for non-target species. especially humans, has remained the centre of international activities in OVD from 1991 onwards. The group very carefully looked at different probable, and also at more unlikely scenarios which could lead to human exposure to a live virus dog vaccine. Those scenarios ranged from the most obvious i.e. exposure to the vaccine contained in a bait to more elaborated situations such as exposure through licking by a freshly immunised dog, or exposure to a vaccine virus reverting to increased virulence after passaging into immunosuppressed target or non-target species.

To better assess the likelihood of these different scenarios the group requested in 1993 that all candidate vaccines be tested in immunosuppressed animal models and for safety in non-human primates. In 1994 it was fluther recommended that better quantitative tests be developed to measure input vaccine virus excretion and that levels of virus excretion with time be evaluated in young puppies as these are the most probable excreters and transmitters of vaccine virus to humans.

During the same period of time, the group also established guidelines for the determination of oral vaccine efficacy in laboratory dogs and for bait development bait preference trials and for the evaluation of bait delivery systems in the field (Matter 1993). In 1994 the group worked on elaborating specific guidelines for implementing OVD projects and it has promoted further investigation of OVD logistics and economics.

Investigating economics of OVD is essential, since it is very unlikely that all resources required for dog rabies elimination become suddenly available. The implementation of control activities will obviously remain under financial strain and require that new techniques be as cost-effective as possible. When targeting certain "high-risk" components of the dog population such as the feral and free-roaming owned dogs it may be possible to accept a cost per dog vaccinated by the oral route higher than that established for a parenteral vaccination (i.e. US$ 1 to US$ 1.3 with US$ 0.35 worth of vaccine (Bögel and Meslin 1990) as most savings accrue as from the stage of rabies elimination. However when oral and parenteral vaccination compete for the same dogs (i.e. owned and restrainable segment of the population) one should expect at least comparable costs per fully vaccinated dog. Indications on how costs of OVD would compare with parenteral vaccination have been given (Blancou 1992).

To reduce costs further and thereby open new opportunities for the initiation of large-scale vaccination programmes, inexpensive and voluntary vaccine delivery systems involving the community itself or community leaders should be promoted. In this context the results acquired recently in Tunisia on placebo bait distribution to dogs via their owners are very encouraging. This method would however necessitate modifications of regulation on the delivery and application of veterinary rabies vaccines currently enforced in many countries. It should also be kept in mind that this move might not be well received by professional associations and governments struggling to implement structural adjustment policies (Perry and Wandeler 1993).

A number of requirements regarding safety of candidate vaccines and safety, efficacy and economics of baits delivery (using placebo baits) still remain to be fulfilled. WHO co-ordinated laboratory and field research on OVD carried out over the past six to seven years has however been very fruitful and has created the proper conditions for launching limited field trials in the near future.

References

Blancou J. (1992) Oral vaccination. In *Proceedings of the third International Meeting on Advances and Research towards Rabies Control in the Americas, 5-7 October 1992, Mexico* City, ISBN 968-811-301-8, pp. 14-121.

Bö6gel K. and Meslin F.X. (1990) Economics of human and canine rabies elimination: Guidelines for programme orientation. *Bulletin of the World Health Organization* 68 (3): 281-291.

Matter H.C. (1989) Populationsbiologische untersuchungen an hundepopulationen in Tunesien. PhD thesis, University of Berne, Switzerland.

Matter H. (1992) Oral and parenteral vaccination of dogs against rabies. In *Proceedings of the third international meeting on advances and research towards rabies control in the Americas, 5-7 October 1992, Mexico city*, ISBN 968-811-301-8, pp 201-205.

Matter H.C. and Fico R. (1992) Accessibility of dogs to oral and parenteral vaccination against rabies in Tunisia and Turkey. Unpublished YMO document No. YMO/Rabies/93.206.

Matter H. (1993) Suggestions for the development of a research project for the field evaluation of several vaccine-bait delivery techniques to vaccinate dogs orally against rabies. WHO/RabRes./93.40.

Matter H.C. *et al.* (1993) Canine ecology and rabies vaccination. In *Proceedings of the Symposium on Rabies Control in Asia, Jakarta, Indonesia*, 27-30 April 1993. Fondation M. Mérieux, Lyon, pp. 75-97.

Meslin F.-X., Fishbein D.B. and Matter H.C. (1994) Rationale and prospects for rabies elimination in developing countries. In *Lyssaviruses,* Edited by Rupprecht C., Dietzschold B. and Koprowski H., Springer Verlag, pp. 1-26.

Perry B.D. and Wandeler A.I. (1993) The delivery of oral rabies vaccines to dogs: an African perspective. *Onderstepoort Journal of Veterinary Research* 60: 451-457.

WHO (1988) Report of the WHO Consultation on Oral Immunization of Dogs against Rabies, Geneva, 26-27 February 1988, WHO/RabRes./88.26.

WHO (1989) Report of the WHO Consultation on Requirements and Criteria for Field Trials on Oral Rabies Vaccination of Dogs and Wild Carnivores, Geneva, 1-2 March 1989, WHO/Rab.Res/89.32.

WHO (1991) Second WHO Consultation on Oral Immunization of Dogs Against Rabies, Geneva, 6 July 1990, WHO/Rab.Res/91.37.

WHO (1992) Third Consultation on Oral Immunization of Dogs Against Rabies, Geneva, 2 1 -22 July 1992, WHO/Rab.Res/92.38.

WHO (1993a) Fourth WHO Consultation on Oral Immunization of Dogs Against Rabies, Geneva, 14-15 June 1993, WHO/Rab.Res/93.42.

WHO (1993b) Rapport de la Consultation informelle sur la recherche en mati6re de vaccination des chiens par voie orale en Tunisie, Genève, le 15 avril 1993. Document VPW93.120.

WHO (1994) Fifth Consultation on Oral Immunization of Dogs Against Rabies, Geneva 20-22 June 1994, WHO/RabRes/94.45.

THE ORAL DELIVERY OF RABIES VACCINES TO DOGS

Carolin L. Schumacher[1]

VIRBAC, Biological Research & Development, 06517 Carros, France

Abstract

In most developing countries the dog remains the major transmission vector of rabies to man, despite the widespread use of parenteral vaccination. Parenteral vaccination does not, in general, achieve population immunity levels high enough to inhibit rabies transmission within the dog population. Following the demonstration that oral vaccination induced levels of population immunity sufficient to interrupt the epidemiological cycle of fox rabies in Europe, research has aimed at the development of this approach for dog rabies control.

Oral vaccines are likely to be easier to administer and culturally better tolerated, and therefore well adapted to overcome the logistical problems hampering rabies control in developing countries. Among several proposed rabies vaccine candidates, the attenuated rabies vaccine strain SAG-2 has been shown to induce a protective immune response to rabies in dogs when delivered by bait.

Oral live vaccines must meet high safety standards due to the risk associated with the dissemination of those replicating antigens in young and/or immunodepressed individuals. SAG-2 has confirmed its safety through the absence of clinical signs, dissemination and excretion in several carnivorous target and an important number of non-target species including non-human primates.

Present field research, carried out using a placebo bait specifically designed for the oral delivery of SAG-2 to dogs, is aiming at confirming the hypothesis according to which oral vaccination, alone or as a possible adjunct to injectable rabies vaccines, might increase vaccine coverage by immunising poorly supervised dogs which are both often inaccessible to parenteral vaccination and a high-risk group for rabies transmission to man.

Introduction

In developing countries. canine rabies control has achieved only limited success despite extensive effort and expense. One often suggested reason for this failure is that dog vaccination levels sufficient to break the dog-to-dog transmission cycles are rarely reached (B6gel et al. 1982; Perry 1993) or cannot be maintained, so that transmission is inevitably re-

[1] Co-author: **A. Aubert**, VIRBAC, Biological Research & Development 06517 Carros, France.

established (El Hicheri 1993). It is often suggested that problems such as inadequate logistics, insufficient community participation or inaccessibility of dogs are responsible for this failure and it is hypothesised that these could be overcome with oral vaccines for dogs (see "Evolution of WHO recommendations on oral immunisation of dogs against rabies" page 145). Rabies control by oral immunisation is possible and has been clearly demonstrated with respect to fox rabies in Europe (Wandeler *et al.* 1988; Brochier *et al.* 1991; Aubert *el al.* 1993). The techniques developed to control wildlife rabies in Europe and North America and their delivery does however require adaptation to be successful in the dog. The approach chosen by our laboratory was the development of the attenuated vaccine strain SAG-2. Until now, only live vaccines have been shown to confer an adequate protective, systemic immune response against rabies in dogs at concentrations conducive to an economical vaccine use (Rupprecht *et al.* 1992). These vaccines have the capacity to replicate locally in the vaccinated animal and in this way the viral antigens are presented to the host's immune system more effectively than would be the case for non-replicating agents (O'Hagan 1992). However, due to the risk of mutations during replication and the proximity of dogs to humans, vaccines intended for the oral immunisation of dogs have to meet high safety standards. Safety recommendations issued by the YMO specify that vaccines should be innocuous for humans, for the target species including very young dogs and for non-target species likely to be attracted by the bait delivery system (WHO 1995). They should not be excreted, as excretion may be indicative of local replication and consequently an increased risk of mutation, reversion to pathogenicity and transmission.

It is assumed that effective rabies control in countries with canine rabies requires the immunisation of a large proportion of the dog population (about 70 percent) in order to reduce the contact rate between infectious and susceptible dogs to a level too low to sustain rabies transmission within the population (Beran 1991). As this level of vaccine coverage is difficult to obtain and maintain, targeting rabies control measures is thought to increase the success of such measures and render them more economic and viable (Perry 1993). Studies are currently being undertaken to analyse the so-called high-risk section of the dog population (in terms of exposure to and transmission of rabies) which are composed of true feral and totally or partly unrestricted, owned dogs. These dogs, characterised by minimal supervision and high dog-to-dog contact rates (Table 1) are likely to be less accessible to parenteral vaccination and to constitute the main target for oral vaccination (Matter 1993). The field trials summarised in this document, using a freeze-dried placebo dog bait, were recently conducted in two rural villages in Tunisia in order to determine the accessibility of certain subpopulations of dogs to this method, the risk of human exposure to vaccine virus and the logistical efforts associated with bait distribution (Ben Youssef *et al.* unpublished data).

The vaccine candidate SAG-2

SAG-2 is a live rabies virus that. was selected from a pool of SAD Bern variants by monoclonal antibodies directed against the external glycoprotein of the rabies virus (Lafay *et al.* 1994). The resulting variant differs from the parental strain SAD (Bern) by two nucleotides in a triplet coding for an aminoacid (Table 2) in a region of the glycoprotein associated with the virulence of rabies viruses (Coulon *et al.* 1983). SAG-2 has lost the residual pathogenicity for mice by the oral, intramuscular and intracerebral route that characterises the parent strain SAD Bern (Lafay *et al.* 1994).

Table 1. Classification of dogs based on their dependency and restriction

	Full restriction Dogs physically separated from rest of the dog population	**Semi-restriction** Dog with access to the rest of the dog population	**No restriction** Dog with free access to the dog population at all times
Full dependency Dog given all of its essential needs intentionally by humans	**Restricted dog**	**Family Dog**	
Semi-dependency Dog is given a proportion of its essential needs intentionally by humans		**Neighbourhood Dog**	**Neighbourhood Dog**
No dependency Dog is given none of its essential needs intentionally by humans			**Feral Dog**

Inoculation of dogs, baboons and over 30 mammalian and avian animal species by the oral route produced no adverse effect and no evidence of virus excretion could be detected (E. Masson *et al.*, in submission to Vaccine; M. Fekadu *et al*, J. Bingham *et al.*, F. Chapparo *et al.*, unpublished data). Since 1992, the strain has been successfully used for fox rabies control in Europe (Masson *et al.* submitted for publication) without any report of negative impacts on the environment.

The vaccine delivery system

The vaccine delivery system developed to vaccinate dogs by the oral route with SAG-2 is composed of two main parts: a freeze-dried central core containing the vaccine and a thin layer of bait matrix attractive to dogs.

The dimensions of the internal unit are approximately 1.5 (H) x 3 (L) x 2.5 (W) cm. Contact with small amounts of aqueous solution leads to the re-hydration of the unit within seconds. This is of importance because, in order to be active, the vaccine virus needs to be reconstituted in the saliva of the dog. Since dogs swallow food generally without chewing, only small quantities of saliva are available. The reconstitution of the vaccine is promoted by the adhesive character of the central unit due to adsorption of bait particles to the mucosa. This characteristic induces a licking reflex in the dog, promoting saliva secretion and consequently bait disintegration.

The freeze-dried unit is coated by a matrix which is attractive for dogs and protects the core unit against minor shocks and humidity. It is composed of materials of animal origin, artificial taste enhancers, and a synthetic polymer responsible for impermeability and mechanical stability.

Table 2 Differences between SAD Bern and SAG-2 in codon and aminoacid sequence 333 of the rabies glycoprotein.

Virus Strain	**Nucleotide Sequence**	**Aminoacid in Position 333**
SAD Bern	AGA	Arginine
SAG2	GAA	Glutamic Acid

Efficacy of the vaccine delivery system in dogs.

Two groups of four adult laboratory beagles, from whom food was withheld 24 hours prior to the experiment, were offered a single bait containing either $10^{9.0}$ median tissue culture infectious doses ($TCID_{50}$) or $10^{8.0}TCID_{50}$ of SAG-2. Two dogs were not vaccinated and kept as challenge controls. Seven of eight vaccinated animals were interested in the bait and consumed it within a period of 2 min. 16 sec to 11 min. 55 sec. One dog of the group receiving the bait with the higher vaccine titre consumed only half of its bait. Twenty-nine days after treatment, all vaccinated and unvaccinated dogs were challenged with a lethal dose of canine street virus injected into the temporal muscle. The two control animals died of rabies as expected, 19 and 26 days later. One animal in each vaccination group also succumbed to rabies 14 and 16 days following challenge (Table 3). Both animals had consumed an entire bait. The shortened incubation period in the vaccinated dogs could indicate the occurrence of an early death phenomenon (Sikes *et al.* 1971, Blancou *et al.* 1980), but the small number of animals renders an interpretation of this observation difficult. None of the- eight vaccinated and the two control dogs presented detectable levels of seroneutralising antibodies before vaccination or before challenge on day 29. Despite the fact that neutralising antibody titres were undetectable prior to challenge, three out of four dogs which received a bait containing either $10^{9.0}$ $TCID_{50}$ or $10^{8.0}$ $TCID_{50}$ were protected against a lethal rabies virus challenge. The difference between the vaccination group and the control group is significant when considering, that the challenge virus concentration corresponds to at least 1 LD100 for dogs (100 percent of controls succumbed) (M. Aubert and C. Schumacher *et al.*, unpublished data).

Table 3 Efficacy of a single freeze-dried bait containing SAG-2 in dogs

Number of animals	**Vaccine conc. per bait**	**YNA* on day of challenge**	**% Challenge mortality****
4 dogs	10^9 $TCID_{50}$	0/4	25%
4 dogs	10^9 $TCID_{50}$	0/4	25%
2 controls	none	0/2	100%

* VNA: Virus Neutralising Antibodies
* Intramuscular challenge with $10^{3.64}$ median mouse intracerebral lethal doses ($MICLD_{50}$) of lethal canine street rabies virus 29 days following vaccination.

Modes of bait distribution

The following studies were carried out in two different test sites in an rural area in the proximity of Tunis. The data presented hereunder are estimates based on a preliminary analysis. The total dog population in either villages accounted for approximately 300 dogs, only 5-15 percent of which were considered unowned and free all the time. The majority of dogs were used for guarding and were tied up on the property.

1. Bait distribution at mobile vaccination centres

Following an information campaign, baits (placebo DBL-2) containing the serum-marker sulfadimethoxine (SDM were handed out to dog owners (one bait per dog) at a mobile vaccination centre with precise instruction for use. The next day each household of the village was visited and the bait consumption determined by questionnaire survey and by serum analysis to reveal the presence of serum marker. According to either analysis, over 90 percent of the owned dogs had consumed a bait and 5 to 15- _percent of these dogs were considered inaccessible for parenteral vaccination. Truly unowned dogs did not receive baits. The logistical effort of the method did not exceed that normally spend for parenteral vaccination campaigns. Due to good compliance with the instructions, human exposure was minimal (Table 4).

2. **Door-to-door distribution**

Following an information campaign, each household of village 2 was visited and a bait (placebo DBL-2) was given to dogs present at the time of the visit. Consumption was determined by direct observation. Sixty-five percent of all owned dogs accepted a bait. The proportion of these dogs that was inaccessible to parenteral vaccination was not determined, but can be considered low due to the fact that unapproachable dogs, freely moving on their territory, frequently left upon arrival of the vaccination team. The logistical effort of the distribution method was very important. Bait acceptance was low compared to the previous distribution mode and might be related to the presence of the vaccination team on the premises. However, for the same reason, the risk of human contamination was minimal (Table 4).

Table 4. Vaccine coverage, risk of human contamination and logistical efforts of oral vaccination compared to parenteral vaccination

Bait Distribution Mode	Theoretical Vaccine Coverage		Risk of Human Exposure to Vaccine Virus	Logistical Effort & Vaccine Cost*
	Total Dog Population	High Risk Population		
Central Point	81%	8%	low	comparable
Door to Door	59%	<8%	none observed	high (Team)
WIM**	25%	6%	high	high (Vaccine)

* Compared to parenteral vaccination campaigns
** WIM = Wildlife immunisation model

3. Wildlife immunisation model

Following an information campaign, over 1000 baits (placebo DBL-2) containing the serum marker, SDM, were distributed along roadsides and around village 2 at a density of 4.7 baits per dog and left there over night. The next day, each village household was visited and a blood sample was drawn from every accessible owned dog. Free-roaming dogs were bled after having been anaesthetised by using a dart blow pipe. Bait acceptance was assessed by SDM analysis on sera collected.

Twenty-three percent of the owned dogs, of which 7.5 percent were considered inaccessible to parenteral vaccination had consumed a bait. Forty percent of the dogs with unknown ownership status (the total dog population comprised 10-15 percent of the dogs with uncertain ownership status) also consumed a bait. The logistical effort, bait costs and the risk of human contamination were considered high in this case (Table 4).

Discussion

The live modified rabies vaccine strain SAG-2 has fulfilled most of the WHO recommendations for efficacy and safety and is due to undergo field evaluation. A suitable vaccine delivery system for carnivores, a freeze dried bait suitable for the delivery of the SAG-2 vaccine into the oral cavity of dogs is under development. The bait is well accepted by owned and unowned dogs. Consumption of a single vaccine laden bait confers protection against a severe street virus challenge to approximately 80 percent of animals vaccinated in the laboratory.

Bait distribution in two Tunisian field sites according to the central point and door-to-door distribution mode reached primarily owned dogs accessible to parenteral vaccination and to a lesser extent, owned dogs inaccessible to parenteral vaccination. Bait distribution according to the wildlife immunisation model reaches primarily unrestricted owned dogs and unowned dogs which together constitute the high risk group in terms of rabies transmission and are largely inaccessible to vaccination, However, the risk of human contamination, especially of children, and the costs of this method are unacceptable. None of the distribution modes reaches more than 10 percent of the high risk dog population, but this might nevertheless be sufficient. The solution could be provided by combining parenteral vaccination of accessible owned dogs with the distribution of baits to owners who could not bring their dog to the vaccination centre. If necessary, unowned dogs could additionally be targeted by the distribution of baits in delimited areas, where unowned and free-roaming owned dogs share resources. Used in combination with parenteral vaccination of dogs, oral vaccination is unlikely to reduce the costs of rabies control in the short run, but the increase of vaccination coverage in the vector population could, as witnessed in some European countries practising oral vaccination of foxes, lead to rabies elimination.

References

Aubert M.F.A., Masson E., Vuillaume P., Artois M. and Barrat J. (1993) Les acquis de la prophylaxie contre la rage vulpine, *Médecine et Maladies Infectieuses* 23: 537.

Beran G.W. (199 1) Urban rabies. In: *The Natural History of Rabies, 2nd Edition,* Edited by Baer G.M. CRC Press, Boca Raton. pp.427-444.

Bögel K., Andral L., Beran G., Schneider L. and Wandeler A. (1982) Dog rabies elimination: a trend analysis and program proposal prepared by a WHO working group. *International Journal of Zoonoses 9:* 97.

Blancou L, Andral B. and Andral L. (1980) A model in mice for the study of the early death phenomenon after vaccination and challenge with rabies virus. *Journal of General Virology* 50: 433.

Brochier B., Kieny M.P., Costy T., Coppens P., Bauduin B., Lecocq J.P., Languet J.B., Chappuis G., Desmettre P., Aflademanyo K., Libois R. and Pastoret P.P. (1991) Large-scale eradication of rabies using recombinant vaccinia rabies vaccine. *Nature 354*: 520.

Coulon P., Rollin P.E. and Flamand A. (1983) Molecular basis of rabies virulence. II. Identification of a site on the CVS glycoprotein associated with virulence *Journal of General Virology* 64: 693.

El Hicheri K. (1993) Resultats du programme national Tunisien de la lutte contre la rage. Rapport du Ministère de l'Agriculture.

Lafay R, Bénéjean L, Tuffereau C., Flamand A. and Coulon P. (1994) Vaccination against rabies: construction and characterization of SAG2, a double derivative of SAD Bern. *Vaccine* 12: 317.

Matter H. C. (1993) Canine ecology and rabies vaccination. In: *Proceedings of the symposium on Rabies Control in Asia, Jakarta, Indonesia, April* 27-30, 1993. Fondation Marcel Merieux, Lyon. pp.75-94.

O'Hagan D.T. (1992) The oral delivery of vaccines. *Clinical Pharmacokinetics* 22: 1.

Perry B.D. (1993) Dog ecology in eastern and southern Africa: Implications for rabies control. *Onderstepoort Journal of Veterinary Research.* 60: 429.

Rupprecht C.E., Dietzschold B., Campbell LB., Charlton K.M. and Koprowski H. (1992) Consideration of inactivated vaccines as oral immunogens of wild carnivores. *Journal of Wildlife Diseases 28: 629.*

Schneider L.G., Cox J.H. (1983) Ein Feldversuch zur oralen Immunisierung von Füchsen gegen die Tollwut in der Bundesrepublik Deutschland. *Tierärztliche Umschau 38:* 3 15.

Sikes R.K, Cleary W.F., Koprowski H., Wiktor T.J. and Kaplan M.M. (1971) Effective protection of monkeys against death from street rabies virus by post-exposure administration of tissue-culture rabies vaccine. *Bulletin of the World Health Organization* 45: 1.

Wandeler A1, Capt A, Kappeler A. and Hauser R- (1988) Oral immunization of wildlife against rabies: concept and first field experiments. *Review of Infectious Diseases 10:* S649.

WHO (1995) Report of the Sixth Consultation on Oral Immunization of Dogs against Rabies unpublished.

SURVEILLANCE AND DIAGNOSIS: SOME THOUGHTS

Alexander L Wandeler

Animal Diseases Research Institute, Nepean, Ontario, Canada

Governments have the obligation to protect people from illness and economic loss caused by transmissible diseases. Diagnosis and surveillance are integral components for the realisation of this responsibility.

Rabies Surveillance

The objective of rabies diagnosis of an individual suspect animal is to enable postexposure treatment decisions. Disease surveillance has additional objectives, such as

- to establish an understanding of the epidemiology (this will allow the formulation of strategies for public health measures, which includes postexposure treatment policies);
- to promote public awareness;
- to permit disease control planning;
- to monitor the impact of disease control operations;
- to maintain rabies-free status.

According to the OEI International Code, the recognition of rabies-free status of a country or area is dependent on the following preconditions:

1) Rabies is a notifiable disease;
2) An effective system of disease surveillance is in operation;
3) All regulatory measures for the prevention and control of rabies have been implemented including effective importation procedures;
4) No case of indigenously acquired rabies infection has been confirmed in man or any animal species during the past two years;
5) No imported case in carnivora has been confirmed outside a quarantine station for the past six months.

(Some exemptions were originally made to precondition number 4.)

Clearly, these preconditions only fit for a very limited number of countries. But they very acutely point to the question: what is an effective system of disease surveillance?

Diagnosis

Often rabies is diagnosed on the basis of clinical symptoms. Rapidly worsening symptoms in a dog such as a sudden change of disposition, apprehensive expression, change of voice, excitability, depraved appetite, drooling of saliva, attacks on objects within reach, paralysis of lower jaw and tongue, posterior paralysis, and the almost invariably fatal outcome are strong indications for the presence of a rabies virus infection. However, there is considerable variation in the clinical course within and between species. In addition, symptoms that are considered pathognomonic may never develop. It is therefore advisable to consider as suspect rabies cases all instances of rapidly progressing neurologic disorder in the absence of a binding alternative diagnosis.

It is suggested that clinically suspect cases of rabies be submitted for laboratory confirmation when they have contaminated (bitten) humans or when essential for epidemiological surveillance. Current laboratory test methods are described in detail in WHO's "Laboratory Techniques in Rabies" (WHO 1973). The method of choice is immunofluorescence for the detection of viral antigen in brain impression smears. The demonstration of inclusion bodies in neurones with histological staining methods is less reliable. A number of other methods (ELISA, PCR, etc.) cannot be recommended for general use at present. However, it is advisable that microscopic findings be confirmed by an additional test, preferably a procedure to isolate the virus in either mice or in tissue culture. Confirmatory tests are especially important when microscopic findings are negative in suspicious cases with histories of human exposure.

Material suitable for surveillance

In places where rabies is known to be endemic it might be sufficient to promote the submission of brain samples of animals that have contaminated humans in order to compile a picture of the distribution and prevalence of the disease. No doubt, this strategy holds the danger that principal hosts may go unrecognised. For example, dogs being the major source of human contamination are the predominant species submitted for diagnosis while a rarely submitted wildlife species could be responsible for maintaining the zoonosis. Dogs may be occasional victims of wildlife rabies in one location, frequent ones in a different place, and they may be the principal host species in again another area. It is obvious that a misinterpretation of the epidemiology has repercussions in the design of disease control strategies.

Specimens can be brought or shipped to the diagnostic centre as whole carcasses, heads, extracted whole brains or brain samples collected through the foramen magnum or the orbit with a straw or another suitable probe. The specimens should be transported frozen or refrigerated. It is advisable to transport small pieces of brain (brain stem, medulla oblongata, hippocampus) in 50 percent buffered glycerol if circumstances do not permit a rapid and refrigerated delivery to the laboratory. One might also consider submitting fresh material and in addition fix half of each brain in formalin for histological or immunohistological processing.

Brain specimens from animals that have acted suspiciously or that have contaminated humans are the material most desirable and suitable for surveillance. Their submission to a diagnostic laboratory should be encouraged. Dogs, wild carnivora, and bats are the species most often recognised as principal hosts. Other domestic carnivora (cats, etc.), domestic herbivores, and wild ruminants are quite often victims of the disease, but they only rarely support epizootics independent from the aforementioned species. Clearly not suitable for surveillance are rodents and birds, as well as clinically normal animals and roadkills. Not that such material should never be examined in well defined projects, but it is very likely that the disease prevalence among them is extremely low. It is also not recommended to base rabies surveillance on serology.

There is a host of reasons why serology can be misleading. Antibodies can be measured in neutralisation tests (assayed in mice or in tissue culture). The most outstanding but rarely acknowledged problem in neutralisation assays are virus inactivating substances that occur quite frequently in blood collected under suboptimal conditions. The problems with nonspecificity that must be overcome in haemagglutination inhibition tests and ELISAs are not less dreadful. Sometimes one succeeds in demonstrating that a collection of samples yields statistically significant, different results from another group of samples, but often it is dubious to make final statements on presence or absence of antibodies in individual samples. If one accepts a positive result as real, then one is left with the question of the cause for their presence. Serum antibodies are usually not of very high titre and are not accompanied by cerebrospinal fluid (CSF) antibodies if they are the result of vaccination. Antibody titres are very high in serum and in CSF if they are the corollary of survival of clinical disease, which is considered to be exceptional. We do not know what antibody levels to expect as results from different forms of abortive infections and noninfective natural exposures (for example, oral). In addition, the possibility of cross-reacting rhabdovirus antibodies has never been investigated properly.

Obligations of the diagnostic centre

The most obvious duty of the diagnostic centre is to achieve diagnosis rapidly and to report quickly to permit timely postexposure treatment and disease control decisions. A diagnostic centre is more useful if it serves a large area and not only its immediate vicinity. It is a formidable task to ensure the submission of specimens from all regions that require diagnostic services and/or need surveillance. It might not be achievable without a network of motivated and well instructed personnel in agricultural and health services in all regions concerned.

The diagnostic centre should give instructions for sample conservation, packaging, completion of submission forms, and transport. The field personnel must also be advised on the purpose of sample submissions and on what categories of specimens to collect. The diagnostic centre has to supply material for conservation and packaging if these are not readily available in the field. It also should provide blank submission forms. Submission forms should accompany every specimen. They should furnish the following information:

- species
- date (of submission)
- location (where animal was found)
- owner's name and address (for domestic animals)
- type of human exposure (none, bite, other)
- date of exposure
- names and addresses of exposed people

Supplementary information might also be collected, such as clinical symptoms, whether the animal was killed or found dead, age and sex of the animal, and observations on the source of infection.

The diagnostic centre should perform a number of additional duties aside from assisting sample submission and achieving laboratory diagnosis. A serotype/genotype identification and/or rabies virus variant characterisation should be performed on some, possibly all, specimens found positive in routine testing. Selected samples should be forwarded to laboratories qualified to perform this task if it cannot be achieved in the diagnostic centre. Reference samples need to be set aside. They should be stored as original rabid brain samples, or as virus isolates made thereof, at –70°C or better in liquid nitrogen or in lyophilised condition. The strategies of specimen selection for long term storage is to cover epizootic events spatially and temporally and to represent all virus variants that might circulate independently.

Last but not least it is the duty of the diagnostic centre to analyse the findings or to make them available for analysis, and to produce monthly, quarterly or yearly reports. None of the classical epidemiological parameters such as incidence and prevalence are very useful: the specimens submitted to the diagnostic centre do not constitute a random sample. A random sample, if one would attempt to analyse one, would have to be enormously large due to the very low prevalence of detectable rabies infections in a population (the infection is not readily detectable in its incubation stage). However, simple periodical maps and tables listing by species and area of origin, the numbers examined and the numbers found rabid will indicate trends and movements.

Reference

WHO (1973) Laboratory Techniques in Rabies. Edited by Kaplan, M.M. and Koprowski, H. World Health Organization, Geneva.

EPIDEMIOLOGY OF ANTIGENIC AND GENETIC VARIANTS OF AFRICAN RABIES VIRUSES

Gavin R Thomson

**Onderstepoort Institute for Exotic Diseases,
Onderstepoort, South Africa**

Introduction

This brief review is intended as a bridge between rabies virologists and molecular biologists on the one hand, and field veterinarians on the other who are involved in the control of rabies in sub-Saharan Africa, particularly those in southern Africa. An attempt is made to provide people with limited exposure to molecular approaches to epidemiological investigation and who are unlikely to have access to literature in that regard, with an understandable synopsis of the relationships that have so far been established between rabies viruses (*viz.* serotype or genotype 1 Lyssaviruses) in different regions of the continent as well as relationships between different carnivore species and particular rabies virus biotypes.

Rabies in Eurasia is an ancient disease: it can be traced back to the 23rd century B.C. in Mesopotamia (Blancou 1988), and has been reported throughout recorded history (Steele and Fernandez 1991). The contention that rabies evolved initially in Africa (King *et al.* 1994) based on the observation that more (4/6) Lyssavirus genotypes (Bourhy *et al.* 1993) occur in Africa than in any other continent, would make it an even older disease in this continent. If that is so, rabies has been in Africa for more than 4300 years. Recorded history in most of sub-Saharan Africa, however, only covers the last 150 - 200 years. This is fortunately not the insurmountable problem it might appear because the discovery of "molecular clocks" within the genomes of living organisms, including viruses, provides a means whereby the paucity of historical record can to some extent be compensated for. Using this approach, some historical inferences about rabies have already been made (Smith *et al.* 1992; Smith and Seidel 1993) and this capability will doubtless increase in future.

Monoclonal antibody studies

Monoclonal antibodies (Mabs) directed against either the glycoprotein (G-Mabs) or, more usually, the nucleoprotein (N-Mabs) of rabies virus, were the first sensitive means by which rabies viruses were differentiated (Wiktor and Koprowski 1978; Wiktor *et al.* 1980; Sureau *et al.* 1983) and it was soon shown that in North America, for example, particular antigenic variants of rabies virus were associated with specific wildlife Mab reaction patterns (Smith and Baer 1988). This phenomenon has been referred to as "species compartmentalisation" and conveys the tenet that there are subtle mutual adaptations between virus variants and animal species which promotes the maintenance of a particular variant in a specific animal host. Occasionally "spillover" of the species-adapted variant into a different species occurs but in the vast majority of cases no further propagation takes place, i.e. this is a dead-end situation. On the other hand, this is likely to be the means by which rabies virus becomes established in new host species and another process of virus/host adaptation is initiated.

Schneider (1982) first described the reaction patterns of African rabies viruses with both N-Mabs and G-Mabs. The patterns obtained with N-Mabs provided three "wild-type Africa" patterns but the species from which these viruses originated were not clearly stated and it is also impossible to reconcile the countries from which the African viruses originated with individual reaction patterns. The origins, both with respect to species and locality, of reaction patterns obtained with G-Mabs are likewise difficult to establish. In 1983 Sureau *et al.* published a more extensive investigation using N-Mabs and 204 virus isolates obtained world-wide, including 73 from Africa and Madagascar of which all but 12 were from dogs. The Madagascan isolates, from 9 dogs and 2 cows, produced four different reaction patterns whereas all 64 viruses originating in North and Central Africa (all dogs) provided a single but distinct reaction pattern. All 9 rabies viruses from Namibia, the only southern African country included in the survey (4 cows, 4 kudu, *Tragelaphus strepsiceros* and a honey badger, *Mellivora capensis*), similarly also produced a single but distinct reaction pattern.

The association of particular antigenic variants of rabies virus with specific wild animal species in southern Africa was first reported on by Foggin (1988). Three viruses obtained from slender mongooses (*Galerella sanguinea*) in central and southern Zimbabwe were shown to differ from those from dogs and jackals in that country. However, the epizootic of rabies in slender mongooses in Zimbabwe dissipated and has not reappeared since.

In a series of reports based on reaction patterns of 16 N-Mabs, King and colleagues (King 1992; King *et al.* 1993; 1994; Tremlett *et al.* 1994) extended Foggin's (1988) study and presented data which clearly demonstrates that rabies viruses from South Africa, Namibia and Botswana can be divided into at least two antigenic groups - those associated with domestic dogs, black-backed jackals (*Canis mesomelas*) and bat-eared foxes (*Otocyon megalotis*), i.e. members of the family Canidae, and the other group that principally infects viverrid species (a separate carnivore family including mongooses, suricates, genets and civets). These two groups were consequently referred to as "canid" and "viverrid" viruses, the former being homogenous (i.e. the reaction patterns of all members of the group obtained from South African canids were identical with the 16 Mab panel used) while the 46 isolates in the viverrid group provided 24 different reaction patterns. These reaction patterns were further divisible into three broad groups delineated on the basis of Clustan analysis (King *et al.* 1994).

Although the majority of canid and viverrid viruses identified in South Africa were isolated from canids and viverrids respectively, some were obtained from livestock and a few isolates were obtained from the "wrong" host providing evidence for "spillover" infections in both directions (King *et al.* 1994). Interestingly, most of the isolates (14/16) so far recovered from wild, feral and domestic felids in South Africa and subjected to antigenic analysis have been found to belong to the viverrid group (King *et al.* 1994; Meredith, C.D. 1995, personal communication).

An intriguing observation reported by King *et al.* (1993) was that the reaction patterns of African mongoose isolates and those from wildlife in the former USSR appeared to be related. It was speculated that this situation could be due to the wildlife isolates from eastern Europe/Asia and Africa being of ancient origin while canine viruses are derived from a variant which has subsequently been carried around the world by "man and his dog".

In a different study in which 95 rabies viruses from regions throughout the African continent (excluding Madagascar) were examined using another panel of 21 N-Mabs (Smith *et al.* 1993), five reaction patterns were obtained. Reaction pattern 1, which had previously (Smith 1989) been found in dog rabies areas of Latin America, Asia and eastern Europe, were also shown to occur widely in Africa, again exclusively in association with dogs, *viz.* in Tunisia, Algeria, Morocco, Cameroon, Gabon, Nigeria, Kenya and Tanzania. Pattern 2 was confined to southern Africa (South Africa, Namibia and Zimbabwe) and occurred in a variety of animals, *viz.* dogs, jackals (14 of 15 viruses tested) as well as domestic animals and six varied wildlife species (water mongoose *Atilax paludinosus*, two kudu, two bat-eared fox and a honey badger). The suggestion was made that this variant predominates in "areas of southern Africa where the jackal is thought to be the major vector of rabies" (Smith *et al.* 1993). Reaction patterns 3, 4 and 5 were made up of only 13 viruses which originated from South Africa and Zimbabwe in a variety of species (4 yellow mongoose *Cynictis penicillata*, grey mongoose *Galerella pulverulenta*, water mongoose, 2 wild cat *Felis lybica*, ground squirrel *Xerus inauris*, bat-eared fox, honey badger, dog and cow). The possibility that these variants are mostly associated with yellow mongooses was made on the basis that in the geographic region from which they were recovered, yellow mongooses are the most important wildlife vector.

Attempts to reconcile the findings of the various Mab studies described above is complicated by the fact that different viruses and Mab panels were used. Nevertheless, some broad conclusions are possible. In particular, it is clear that there is considerable antigenic variation among African rabies viruses with most of the variation occurring in southern Africa. Within southern Africa there is consensus that a clear antigenic difference exists between canid-associated and viverrid-associated viruses with the latter group being surprisingly heterogeneous. However, the studies of Smith *et al.* (1993) and King *et al.* (1993; 1994) disagree in interpretation of the findings. King *et al.* (1993; 1994) consider that the viruses circulating in wild canids (jackals and bat-eared foxes) were derived from canine rabies imported into southern Africa, whereas Smith *et al.* (1993) offer the possibility that the jackal rabies variant is indigenous to Africa and is only indirectly related "to the global reservoir of dog rabies". The findings of Smith *et al.* (1993) also imply that dog viruses in southern Africa differ from those which occur over almost all the rest of the southern African continent where the viruses so far examined show little if any variation. The situation in Madagascar may, however, be different if the results of Sureau *et al.* (1983) are correct.

It must be borne in mind that while considerable antigenic variation has been shown between rabies viruses using Mabs, there is at present little reason to believe that this variation is sufficient to render vaccines currently in use ineffective (King *et al.* 1994). However, this is an issue which requires further investigation, particularly in southern Africa.

Genome-based studies

To date these studies have concentrated on the nucleotide or deduced amino-acid sequences of three genomic regions of the rabies virus, *viz.* the nucleoprotein and glycoprotein genes as well as the non-coding intergenic region between the glycoprotein and polymerase (L) genes sometimes referred to, probably incorrectly, as the pseudogene (T) (Ravkov *et al.* 1995). The relative positions of these genomic regions is shown in Figure 1. There is considerable variation in the rate at which mutations occur in these different genomic regions, i.e. in the degree of conservation. This can be exploited to different ends: for example, closely related viruses can conveniently be compared by sequence analysis of highly variable regions of the genome (those with a high frequency of mutational events) such as the G-L intergenic region or the cytoplasmic region of the glycoprotein gene (Tordo *et al.* 1993). For viruses which are more distantly related it may be easier to use relatively conserved regions such as the nucleoprotein genes, especially its central region (Smith *et al.* 1992; Tordo and Kouknetzoff 1993) or the polymerase (L) gene (Poch *et al.* 1990).

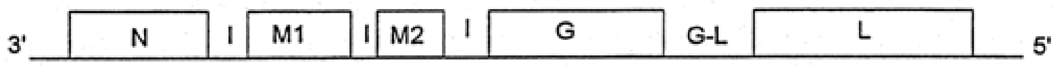


Figure 1 Arrangement of the 12 000 base, single stranded, negative-sense RNA genome of rabies virus. (N = nucleoprotein gene; M1 = polymerase-associated phosphoprotein gene; M2 = matrix protein gene; G = glycoprotein gene; GI, =long intergenic region (approximately 425 nucleotides); L = polymerase gene; I = short intergenic regions)

In a comprehensive study of canine-associated rabies viruses obtained from all the continents of the world where rabies occurs, Smith *et al.* (1992), using a 200 base region of the nucleoprotein gene, showed that the 87 viruses could be divided into six broad groups (I -VI) which shared less than 90 percent homology. Group I included viruses from the southern USA, Central and South America, Europe, the Middle East, North and West Africa and China. By contrast the other five groups occurred in relatively small geographic areas of either South-East Asia, India or West Africa. In an attempt to explain these findings, it was suggested that the six groups were derived originally from different progenitor rabies viruses enzootic in wild Canidae in Asia and Africa. The wide distribution of group I viruses was advanced as possibly being due to European colonists who spread the virus around the world with the dogs that accompanied them (Smith *et al.* 1992).

In a subsequent report Smith *et al.* (1993) described another two groups, VIII and IX (group VII contained vampire bat isolates from South America), which are indigenous to southern Africa. Group VIII contained three isolates from three diverse species (wild cat, water mongoose and ground squirrel) while group IX, which was itself heterogeneous and contained three subgroups, comprised seven viruses, six of which were from mongooses, the other being a bat-eared fox isolate. They further showed that Kenyan and Tanzanian dog viruses as well as southern African isolates associated mostly with jackals could be included in the geographically widespread group I viruses although the southern African viruses formed a distinct subgroup. This implies that jackal isolates from southern Africa are likely to have an epidemiological association with canine viruses belonging to group I and contradicts the conclusion drawn from their Mab study presented in the same paper (see above).

Using a much larger portion of the rabies virus genome (2500 residues of the so-called M-L amplicon which covers both conserved and variable regions of the genome), Tordo *et al.* (1993) found that rabies viruses from Europe and the Middle East and dog-associated viruses widely distributed in Africa (designated Africa 1) form a distinct group of related viruses. A second group of rabies viruses derived from domestic dogs in west-central Africa and Somalia (Bourhy *et al.* 1993) differed from Africa 1 viruses and were consequently referred to as Africa 2. A subdivision within Africa 1 viruses was also reported: those designated as 1a were confined to North African countries while 1b viruses were widespread in western, central, eastern and southern Africa. A map showing these distributions was produced by Bourhy *et al.* (1993). However, this study did not include viruses derived from wildlife hosts in Africa.

The genome-based studies summarised above suffered the disadvantage that there were relatively few rabies viruses available from the vastness of the African continent and it should therefore be anticipated that they lack considerable detail as far as specific regions of Africa are concerned. They nevertheless show that the epidemiological situation in southern Africa differs from that in the rest of the continent. In an effort to extend our understanding of the southern African rabies situation, Nel and his colleagues (Nel *et al.* 1993; Von Teichman *et al.* 1995; Nel *et al.* in preparation) have undertaken studies aimed at investigation of the relationships between as many rabies viruses as possible from southern Africa in relation to their geographical distributions and host animal associations. In particular, an endeavour has been made to substantiate and extend the Mab-based findings which show that viverrid-associated and canid-associated viruses in southern Africa are derived from different lineages (King *et al.* 1994).

Concentrating on the nucleotide sequences of a portion of the genome which includes the cytoplasmic region of the glycoprotein gene and the G-L intergenic region, which both have high mutation rates (Tordo *et al.* 1993), it was confirmed that canid and viverrid isolates differ clearly from each other and that while the canid viruses are essentially homogeneous, those from viverrids are heterogenous and comprise at least five subgroups (Figure 2). Furthermore, the canid viruses were more closely related to PV (Pasteur virus) than were the viverrid strains indicating an association between southern Africa canid viruses and European rabies viruses (Figure 2). Another important finding was that the five subgroups of viverrid virus were associated geographically with distinct and different localities on the central plateau of South Africa (Figure 3). This indicates independent evolution over a considerable period of time of rabies viruses in viverrids, principally yellow mongooses, which is the wildlife species most frequently infected with rabies in South Africa (Swanepoel *et al.* 1993; Swanepoel 1994) and is strongly supportive of the belief that rabies is indigenous to that region. Further support for this contention was provided by a separate investigation in which the mitochondrial DNA of yellow mongooses from different localities in southern Africa was used to infer maternal lineages (Van Vuuren, B. and Robinson, T., in preparation). This showed that while yellow mongooses are widely dispersed in southern Africa there is little evidence for gene flow between mongooses in different localities although the reason for this separation is not clear; it does not appear to be due to geographic barriers. This situation would have made it possible for rabies virus and yellow mongooses to co-evolve independently in different regions of the central plateau of South Africa.

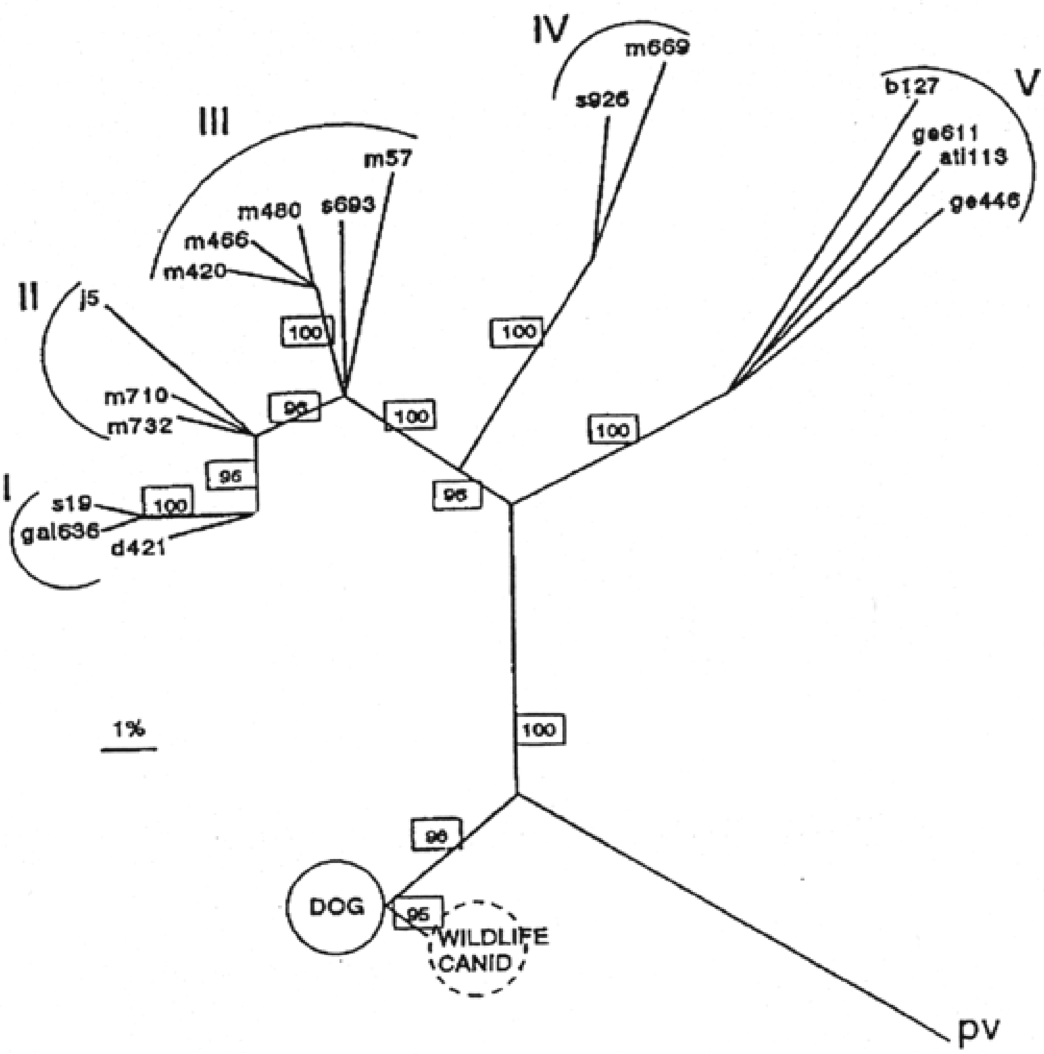


Figure 2. Dendrogram derived from gene sequences of South African rabies virus isolates. (PV = Pasteur virus.) (Figure supplied by Professor L. Nel, University of Pretoria.**)**

Conclusions

The findings of both Mab- and genome-based studies indicate that in southern Africa there are two epidemiologically separate lineages of rabies virus. The first of these comprises a diverse, indigenous group which has probably evolved, presumably over millennia, in viverrids in general and yellow mongooses in particular. No similar epidemiological situation has been described elsewhere in the world and it may therefore be unique. The other, more homogenous groups of viruses, is maintained by both wild and domestic canids and was probably only introduced into southern Africa in the late 1940's from a line which traces back to European rabies viruses, which themselves are homogenous (Tordo *et al.* 1993). Alternatively, this virus could also be indigenous to southern Africa. Canid viruses of southern Africa appear to be related, on the basis of genomic studies, to at least some of those circulating in dogs in East Africa (Smith *et al.* 1993) and those in turn are probably directly related to dog rabies viruses in the rest of the continent with the exception of some in West Africa that seem to have a different but equally ancient source.

The differentiation of southern African rabies viruses into two groups on the basis of Mab and genome sequencing studies is supported by provisional results from studies aimed at the examination of biological differences between these two virus groups (Chapparo and Esterhuysen 1993).

Figure 3. Association of viverrid virus subgroups with geographic localities. (Fig. supplied by Professor L. Nel, University of Pretoria.)

References

Blancou J. (1988) Epizootiology of rabies: Eurasia and Africa. In: *Rabies.* Edited by Campbell LB. and CharIton K.M. Kluwer, Dordrecht.

Bourhy H., Kissi B. and Tordo N. (1993) Taxonomy and evolutionary studies on Lyssavirus with special reference to Africa. *Onderstepoort Journal of Veterinary Research* 60: 277-282.

Chapparo F. and Esterhuysen LL (1993) The role of the yellow mongoose *(Cynictis penicillata)* in the epidemiology of rabies in South Africa - preliminary results. *Onderstepoort Journal of Veterinary Research* 60:373-377.

Foggin C.M. (1988) Rabies and rabies related viruses in Zimbabwe: historical, virological and ecological aspects. PhD Thesis, University of Zimbabwe, Harare.

King A.A. (1992) African overview and antigenic variation. In: *Proceedings of the International Conference on Epidemiology, Control and Prevention of Rabies in F-astern and Southern Africa. Lusaka, Zambia, 2 - 5 June* 1992. Edited by King A.A. Editions Foundation Marcel Mérieux, Lyon, France.

King A.A., Meredith C.D. and Thomson G.R. (1993) Canid and viverrid rabies in South Africa. *Onderstepoort Journal of Veterinary Research* 60: 295-299.

King A.A., Meredith C.D. and Thomson G.R. (1994) The biology of southern African Lyssavirus variants. In: *Lyssaviruses.* Edited by Rupprecht C.E., Dietzschold, B. and Koprowski, H. Springer-Verlag, Berlin, Heidelberg, New York. pp 267-295.

Nel L.H., Thomson G.R. and Von Teichman B.F. (1993) Molecular epidemiology of rabies virus in South Africa. *Onderstepoort Journal of Veterinary Research* 60: 301-3 06.

Poch 0., Bloomberg B.M., Boughueleret L. and Tordo N. (1990) Sequence comparison of the polymerase (L proteins) of unsegmented negative-strand RNA viruses: theoretical assignments of functional domains.. *Journal of General Virology* 71: 1153-1162.

Ravkov E.V., Smith 1S. and Nichol S.T. (1995) Rabies virus glycoprotein gene contains a long 3'noncoding region which lacks pseudogene properties. *Virology* 206: 718-723.

Schneider L.G. (1982) Antigenic variants of rabies virus. *Comparative Immunology and Microbiology of Infectious Diseases* 5: 10 1- 107.

Smith LS. (1989) Rabies virus epitopic variation: use in ecologic studies. In: *Advances in Virus Research.* Edited by Maramorosch K., Murphy F.A. and Shatkin A.L Academic Press, San Diego. pp 215-224.

Smith LS. and Baer G.M. (1988) Epizootiology of rabies: The Americas. In: *Rabies.* Edited by Campbell J.B. and Charlton K.H. Kluwer, Dordrecht.

Smith LS., Orciari L.A., Yager P.A., Seidel D.D. and Warner CK (1992) Epidemiology and historical relationships among 87 rabies virus isolates as determined by limited sequence analysis. *Journal of Infectious Diseases* **166***:* 296-3 07.

Smith LS. and Seidel H.D. (1993) Rabies: A new look at an old disease. In: *Progress in Medical Virology*. Edited by Melnick LL. Karger, Basel. pp 82-106.

Smith LS., Yager P.A. and Orciad L.A. (1993) Rabies in wild and domestic carnivores of Africa: epidemiological and historical associations determined by limited sequence analysis. *Onderstepoort Journal of Veterinary Research* **60***:* 307-314.

Steele J. H. and Fernandez PJ. (1991) History of rabies and global aspects. *The Natural History of Rabies, 2nd.* Edited by Baer G.M. CRC Press, Boca Raton. pp. 1-24.

Sureau P., Rollin P. and Wiktor T.J. (1983) Epidemiological analysis of antigenic variations of street rabies virus: detection by monoclonal antibodies. *American Journal of Epidemiology* **117***:* 605-609.

Swanepoel R. (1994) Rabies. In: *Infectious Diseases of Livestock.* Edited by Coetzer J.A.W., Thomson G.R- and Tustin R.C. Oxford University Press, Cape Town.

Swanepoel R., Barnard B.H.J. Meredith C.D., Bishop G.C., Brückner G.K., Foggin C.M. and Hübschle 0.J.B. (1993) Rabies in southern Africa. *Onderstepoort Journal of Veterinary Research* **60***:* 325-346.

Tordo N. and Kouknetzoff A. (1993) The rabies virus genome: an overview. *Onderstepoort Journal of Veterinary Research* **60***:* 263 -269.

Tordo N., Badrane H., Bourhy H. and Sacramento D. (1993) Molecular epidemiology of Lyssaviruses: focus on the glycoprotein and pseudogenes. *Onderstepoort Journal of Veterinary Research* **60***:* 3 15 -3 23.

Tremlett J.G., Wibberley G. and King A.A. (1994) Rabies virus typing - Preliminary survey in Botswana. *Tropical Animal Health and Production* **26***:* 15 7-160.

Von Teichman B.F., Thomson G.R., Meredith C.D. and Nel L.H. (1995) Molecular epidemiology of rabies virus in South Africa: evidence for two distinct virus groups. *Journal of General Virology* **76***:* 73-82.

Wiktor T1, Flamand A., and Koprowski H. (1980) Use of monoclonal antibodies in diagnosis of rabies virus infection and differentiation of rabies and rabies-related viruses. *Journal of Virological Methods* **1***:* 3 3 -46.

Wiktor T.L and Koprowski H. (1978) Monoclonal antibodies produced against rabies virus produced by somatic cell hybridization: detection of antigenic variant viruses. *Proceedings of the National Academy of Sciences USA* **75***:* 3938-3942.

DIAGNOSIS OF RABIES

Arthur A. King

Formerly, Central Veterinary Laboratory, Weybridge, U.K

We have heard such excellent contributions on and witnessed such a harrowing video of rabies in humans and animals that I need not dwell for too long upon the clinical disease of rabies. Suffice it for me to remind you of the salient features. In humans, the incubation period, that is, the interval between exposure and the first symptoms in the prodromal stage, may last from as short as four days to longer than ten years, although in cases with very long incubation periods the possibility of a second exposure often cannot be ruled out. In a recent analysis of the incubation periods of 1555 patients (Fishbein 1991), none was shorter than 10 days, 29.8 percent were of 10-30 days, 54.4 percent were of 31-90 days, 14.6 percent were of 91-365 days and 1.2 percent were of over 365 days duration. Factors which may influence the length of the period include the site of the bite, the quantity of virus "inoculated" and the age and immune status of the victim. Far less is known of the incubation period in naturally infected animals but it is likely to be equally variable. Bingham *et al.* (1994), citing the case of a wild-caught civet demonstrated that it can also be lengthy - almost seven months after capture in their example.

Early symptoms in rabies patients may resemble those of tetanus, typhoid and malaria, or of viral encephalitides caused by measles, mumps, herpesvirus or enteroviruses. A paralytic rabies diagnosis may be confused with poliomyelitis, acute inflammatory polyneuropathy (Guillain-Barré syndrome) or with post-vaccinal encephalitis (PVE) due to the presence of myelin in brain-tissue vaccines. Confusion in diagnosis is frequently the result of a lack of history of a biting incident - the bite may have appeared trivial or have occurred many months before and been forgotten by the patient.

Clinical signs of rabies in animals are manifest by behavioural changes and unexplained paralysis. Normally, domestic pets are friendly, are not easily disturbed, look healthy and exhibit a predictable behavioural pattern. Wild animals also exhibit characteristic behavioural patterns - normally they are afraid of man and in a free environment they usually prefer to slip away unseen and to avoid contact. Rabid animals do not behave normally. The first changes seen are somewhat benign, the animal is lethargic, appears unhealthy with some slight inco-ordination. This condition may remain stable for about 12 hours then deteriorate rapidly as quadriplegia develops; mandibular paralysis with loss of swallowing reflex may be observed (dogs do not exhibit hydrophobia) and the animal may die without other signs. Early signs in dogs last two to five days and are followed by paralytic rabies in 75 percent of cases or furious rabies in the remainder. Paralysis and death occurs in both paralytic and furious forms of the disease four to eight days after the onset of symptoms. However, virus may be present in the saliva of rabid animals for many days before clinical signs appear and it may be steadily or intermittently secreted until just before death. Cats usually develop furious rabies.

Getting samples to the Laboratory

In many countries in Africa, getting animal heads to the laboratory forms a major part of the costs of diagnosis. In ideal circumstances the entire head and neck of a suspect animal should be sent to the Laboratory, but savings can be made if, in the field, brain sampling without opening the skull can be performed. A plastic straw is passed either through the occipital foramen, heading in the direction of an eye, or, after making an entry through the posterior wall of the orbit with a trocar, screwing the straw through the brain in the direction of the occipital foramen (WHO 1988). One straw sample can be used for histological examination, in which case the straw contents are expelled into a formalin fixative; a second straw sample can be taken for FAT and/or virus isolation, in which case the brain tissue is retained in the straw in glycerol-saline during despatch to the laboratory. The best straw samples are those taken immediately after the animal has died, but rabies antigen remains detectable long after the virus ceases to be viable and has been demonstrated in brain tissue from highly decomposed animals. In humans, a needle biopsy of the brain via the foramen magnum or superior orbital fissure provides sufficient material for FAT and/or virus culture to confirm death from rabies.

Laboratory tests

1. Fluorescent antibody tests (FAT)

Brain smears of the thalamus, pons or medulla, hippocampus and cerebellum or spinal cord (or from the core sample preserved in glycerol-saline) are made on glass slides, allowed to air-dry at room temperature, inactivated for two minutes under UV light about 20 cm. above the slides and then fixed in acetone at –20°C for 1-2 hours. Upon removal from acetone, slides are again air-dried, rabies specific conjugate containing Evan's blue as a counterstain is added to the smears and, after incubation at 37°C for 30 minutes, the slides are washed in one or two changes of phosphate buffered saline (PBS), air dried and, depending upon the power of the microscope, the smears are examined either dry or mounted in PBS.

Although the FAT is the simplest and most accurate test for confirmation of rabies antigen in brain material, attention to detail is all important. If UV light for inactivation is not available, remember that the virus may still be viable after acetone fixation; never use acetone which has been stored in a bottle previously used for another chemical. Always check the pH of buffers and ensure that they are on the alkaline side of neutrality. Use the best conjugate that you can afford - the conjugate specially prepared (at Onderstepoort from anti-N protein extracted by Dr A.I. Wandeler at Animal Diseases Research Institute, Ottawa, Canada) for use in the region is highly specific, has a low background, recognises all rabies serotypes and is cost-effective.

Wherever possible, use the microscope for rabies work only. Incide nt-light microscopy is far superior to transmitted fight. Always use microscope objectives that are designed for fluorescence work. A combination of higher magnification objective and lower magnification eyepieces is better than the other way around. It is better to buy one or two top quality objectives rather than a turretful, of which x5, x 10 and x 100 will hardly ever be used. Use as a minimum a x25, but a x40w or x50w (water lenses used when a coverslip mounted on PBS is used), in conjunction x6.3 eyepieces, an incident-light microscope and the Onderstepoort conjugate, will give magnificent fluorescence.

Mercury vapour lamps do not last for ever and a record of their working life should be kept With incident light it is necessary to wait only a minute or two after switching on before reading can commence. Lamps used beyond the manufacturer's recommended time have been known to explode, causing not only a health hazard but also damaging the collector lens and mirror in the lamphouse - the repairs are expensive. The lamps may appear to be giving good light but the glass of the lamp may become blackened and its efficiency should be checked regularly by using a known positive slide which has been stained and stored frozen after sealing the coverslip, with nail varnish - such slides can be used many times provided that they are returned to the freezer immediately after use. If a result is required urgently, cut down on the staining time, never the fixation time - the longer a smear is fixed (up to overnight fixation), the better the staining will be; always aim to inform the sender of the result on the day that the specimen was received, thus building confidence between owner, field staff and laboratory.

FAT staining can be used on formalin fixed tissue. Pieces (of about $2cm^3$) from Susa-fixed cerebral cortex, basal ganglia, hippocampus (Ammon's horn), midbrain and medulla are pre-treated by emulsification, washing with PBS and then digestion with trypsin. Smears can then be made, fixed and stained in the usual way.

2. Histology

Although inflammatory lesions found in the brain during rabies diagnosis may be common to other viral infections, the specific round to oval inclusions described by Negri in 1903, usually found in the cytoplasm of undamaged nerve cells, particularly in the hippocampus, may be identified by histological and immunological techniques. In sections which have been wax-embedded, the refractory, acidophilic, RNP-containing "Negri bodies", which consist of a reticulogranular matrix containing tubular structures contiguous with maturing virus particles, are regarded as pathognomonic for rabies; their absence, however, does not exclude the disease. They may be found in almost any region of the brain, but they are seen in greater numbers in the central pyramidal layer of Ammon's horn of the hippocampus, in the lower loop and the middle layer of the ganglioneurones and in the neurones of the cerebellum, the motor area of the cerebral cortex and in the medulla. Their frequency of occurrence, size and shape may be influenced by the host species, the infecting virus strain and the clinical phase period. Although histological methods are still used in some countries, they have been largely replaced by the quicker and more accurate FAT.

3. Virus isolation in mice

Animal inoculation, using suckling or weaned mice, remains a practical and reliable method and in some countries the only laboratory method of rabies diagnosis. The clinical disease in mice is brief but the incubation periods of street strains are typically long and may vary from 7 to 28 days and occasionally longer. Diagnosis time may be shortened by using a method in which individuals of a series of inoculated mice are killed at daily intervals and their brains examined using the FAT (Webster *et al.* 1976).

4. Virus isolation in cell culture

With few exceptions rabies infected primary cells and cell lines exhibit no obvious cytopathic changes, but antigen produced during virus growth may be detected by using FAT. Baby hamster kidney (BHK-21) cells are now routinely used in many diagnostic laboratories. They are hardy, require a relatively simple medium supplemented with inexpensive bovine serum and since they do not require additional CO_2 to support growth, they can be used in a closed and therefore safer culture system. Neuroblastoma cells of human or murine origin are also widely used and in some laboratories their culture is used in the place of mouse inoculation as a method of virus isolation. Although they are said to be 200 times more sensitive than BHK-21 cells, they replicate more slowly, do require additional CO_2 and are more prone to toxicity from decomposing or less fresh specimens.

At the molecular level, rabies RNA can be detected following reverse transcription and cDNA amplification by PCR. Molecular techniques for rabies diagnosis, however, need further refinement particularly if they are to be used in countries where sample degradation is a problem because of the climatic conditions.

5. Measurement of neutralising antibody in serum

WHO recommends that all staff working with rabies virus should be vaccinated. Immunisation should preferably consist of three full intramuscular doses of tissue-culture rabies vaccine of potency at least 2.5 X per dose given on days 0, 7 and 28. Serum samples should be tested 1-3 weeks after the last dose and thereafter checked every, 6 months and a booster administered when the titre falls below 0.5 IU/ml (WHO 1992).

One widely recognised method of antibody measurement is the Rapid Fluorescence Focus Inhibition Test (RFFIT) (Smith *et al.* 1973). For many years at Weybridge. a straightforward neutralisation test in 96-well plates using six three-fold serum dilutions (1/10 - 1/2430) in duplicate (50µl/well) to which is added 50µl CVS virus at 100 median tissue culture infectious doses ($TCID_{50}$), incubated at 37°C for 90 minutes has been used. After the addition of BHK-21 cells the plates are incubated at 37°C for up to four days before being fixed in 80 percent acetone for 15 minutes. air-dried and then stained and read on an incident-light microscope, using a x25LL (long reach) objective. Wells with no virus growth indicate that the virus has been completely neutralised; wells with fluorescent cells indicate incomplete or no neutralisation. Each test includes a back-titration of the virus and an international standard antiserum. The test is accepted if the virus back-titration indicates that 30 - 300 $TCID_{50}$ of virus was added to the test and standard sera. The test serum titre is compared with that of the standard and can be expressed in international units by reference to a statistical table. Almost all sera from HDCV vaccinees titrate -within the test range, although occasionally higher titres are noted. As a rough guide, by this technique the Copenhagen standard titres at 1/810, equivalent to 10 IU/ml.

6. Ante-mortem diagnosis

"Ante-mortem tests for rabies have the advantage that the disease can be confirmed on a differential diagnosis list and if necessary, the appropriate management given to the patient" (Bingham and Mlambo 1995). It is entirely for that reason that, in the UK where rabies is no longer present, attempts have been made to isolate rabies virus from saliva samples from patients who have been bitten abroad. Such attempts have been successful in only 50 percent of the patients, but in one notable case virus was isolated from five of six daily saliva samples taken from a patient who later died of the disease (King and Turner 1993). Significantly, no virus was isolated on the four days immediately preceding death, but a rising serum antibody titre was detected over the last three days.

Rabies antigen has been reported to be demonstrated by FAT on corneal scrapings (Schneider 1969), but concerns have been expressed regarding possible damage to the cornea of non-rabid patients who recover and interpretation of results from such smears is often difficult. Antigen may be detected by FAT in 4-6 mm skin biopsies including hair follicles taken from the neck and from the bitten skin. Although it may be necessary to examine 50 or more frozen sections from each biopsy, the test is 60-100 percent sensitive (Warrell *et al.* 1988). More recently, Bingham and Mlambo (1995) were able to diagnose rabies in three patients, from skin biopsies from the nape of the neck; they modified the work described by Blenden *et al.* (1983) by cutting transverse sections, perpendicular to the epidermis and therefore cutting the hair follicle longitudinally. Significantly, perhaps, although skin biopsies from all three patients were FAT positive, corneal smears from the one patient where these were taken were negative. In regions where nervous-tissue-origin (NTO) vaccines are still used, rabies and post-vaccinal encephalitis (PVE) can present indistinguishable clinical pictures. However, neutralising antibody is not detectable in unvaccinated patients during the first week of illness and it has been proposed that a diagnosis of PVE is strongly suggested in patients at risk if antibody is detected in the serum and no rabies antigen is detected in the skin biopsy (Warrell *et al.* 1988).

References

Bingham L, Hill F.W.G. and Matema R. (1994) Rabies incubation in an African civet. *Veterinary Record* **134:528**

Bingham J. and Mlambo P. (1995) Ante-mortem diagnosis of human rabies by the skin biopsy technique: three case reports from Zimbabwe. *Central African Journal of Medicine* 41:258-260.

Blenden D.C., Bell LE, Tsao A.T. and Umoh LU. (1983) Immunofluorescence examination of the skin of rabies infected animals as a means of early detection of rabies virus antigen. *Journal of Clinical Microbiology* 18:631-6.

Fishbein D.B. (1991) Rabies in Humans. In: *The Natural History of Rabies, 2nd Edition.* Edited by Baer G.M. CRC Press, Boca Raton. pp. 519-549.

King A.A. and Turner G.S. (1993) Rabies - a review. *Journal of Comparative Pathology* **108**:1-39.

Schneider L. G. (1969) The corneal test: a new method for the intra-vitam diagnosis of rabies. *Zentralblatt für Veterinärmedizin (Reihe B)* **16**:24-3 1.

Smith LS., Yager P.A. and Baer G.M. (1973) A rapid tissue culture test for determining rabies neutralizing antibody. In: *Laboratory Techniques in Rabies 3rd Edition.* Edited by Kaplan M.M. and Koprowski H. World Health Organization, Geneva. pp. 354-357.

Warrell M.L, Looareesuwan S., Manatsathit S., White N.J., Phuapradit P., Vejjajiva A., Hoke C.H., Burke D. S. and Warrell D.A. (1988) Rapid diagnosis of rabies and post-vaccinal encephalitides. *Clinical and Experimental Immunology* 71:229-234.

Webster WA, Casey G.A. and Charlton K.M. (1976) The mouse inoculation test in rabies diagnosis: early diagnosis in mice during the incubation period. *Canadian Journal of Comparative Medicine* **40**:322-325.

WHO (1988) Simplified technique for the collection, storage and shipment of brain specimens for rabies diagnosis. World Health Organization, Geneva. (VMO/Rab.Res./88.27)

WHO (1992) WHO *Expert Committee on Rabies. Eighth Report.* World Health Organization, Geneva. (TRS 824)

RABIES SERO-ANTIBODY MEASUREMENT WITH SERONEUTRALISATION TESTS

J. Barrat[1]

Laboratoire d'Etude sur la Rage et la Pathologie des Animaux Sauvage, CNEVA, Malzeville, France

The reference method for antirabies antibody measurement is seroneutralisation. The general principle of the method is as follows: a constant amount of virus is mixed with constant volumes of serum dilutions. After an incubation step, the remaining virus is detected either by mouse inoculation (clinical signs and death of animals are observed) or by cell culture. These tests measure the "neutralising power" of the serum. This neutralisation includes a specific component, due to antibodies and a possible non-specific component.

When a large number of sera must be tested ELISA techniques may be useful because they are generally rapid and objective. Different methods may be used: direct estimation of the amount of specific antibodies, indirect estimation of the antibodies by measuring the remaining virus after an incubation step or by a competitive test using monoclonal antibodies as the competitor.

This paper is a summary of remarks and questions which arose during an epidemiological study performed on dogs.

MOUSE NEUTRALISATION TEST

This technique is well standardised (Atanasiu 1973). The critical points are described including the dilution of serum to be tested, the challenge strain used, the dose of challenge virus, the mice to be used and the calculation method.

The comparison of results between laboratories is possible with this method.

CELL CULTURE NEUTRALISATION TEST

In vitro test methods are favoured over *in vivo* methods because they do not use animals and also because *in vitro* tests are generally as sensitive as *in vivo* ones and give an answer more rapidly and at a lower cost.

[1] Co-authors: **S. Cleaveland**, Institute of Zoology, Regents Park, NW1 4RY, London, LTK; **Fl. Cliquet**, CNEVA, 54220 Malzeville, France; **J. Esterhuysen**, Veterinary Research Institute, Onderstepoort 0 110, South Africa; **M. Aubert** CNEVA, 54220 Malzeville, France.

First observation

In 1992 the WHO/OIE collaborative centre was asked by OIE to produce a reference serum of dog origin for rabies. In order to establish the titre of that serum, different freeze dried aliquots were sent to 20 laboratories for titration on cell culture. These laboratories were asked to titrate this dog reference serum against the WHO reference antirabies immunoglobulin.

Up until the present there have been 14 responses. Figure 1 shows the distribution of titres according to the number of laboratories. Two groups of titres are observed: 5 to 10 IU/ml and 12 to 19 IU/ml.

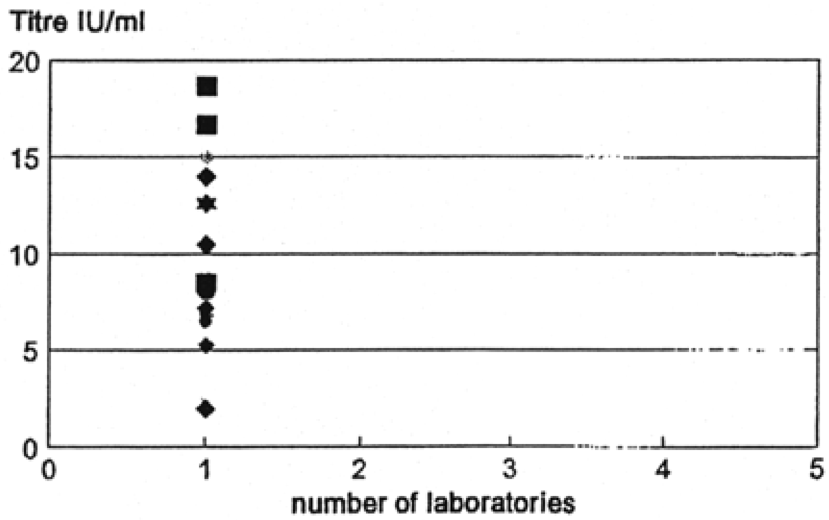


Figure 1: Range of titres obtained in 14 different laboratories for different aliquots of the same pool of immunised dogs. Results are expressed as the mean of three different tests.

The conclusion of this observation and of the analysis of the test procedures was that it was not possible to establish a standardised titre without further studies.

Blind test

The aim of this test was to control the specificity of the technique, the accuracy and the consistency of dilution factors.

Every participating laboratory received a set of 10 tubes:

- nine unknown dilutions of a pool of sera collected from vaccinated dogs in a pool of sera of naive dogs. Two of these dilutions contained no antibody. Every participating laboratory received identical panels.
- an aliquot of a pool of vaccinated cat sera.

Each panel had to be titrated three times in independent operations.

Up until the present eight out of the ten laboratories that received the panel have responded. Two incomplete answers cannot be used.

All the positive samples were found positive and all the negative ones were found negative. So the specificity of the reaction for this test was 100 percent. The intra-laboratory inter-test variation was generally lower than 15 percent. The repeatability is higher than 15 percent in only one laboratory.

The intra-laboratory consistency is illustrated in Figure 2. It is represented by the regression line between observed titres and the theoretical titres determined from the titre of the more concentrated tube and from the dilution factor. The inter-laboratory variation, that is, the range of titre for the same serum sample, illustrated in Figure 2, is close to the variation illustrated in Figure 1.

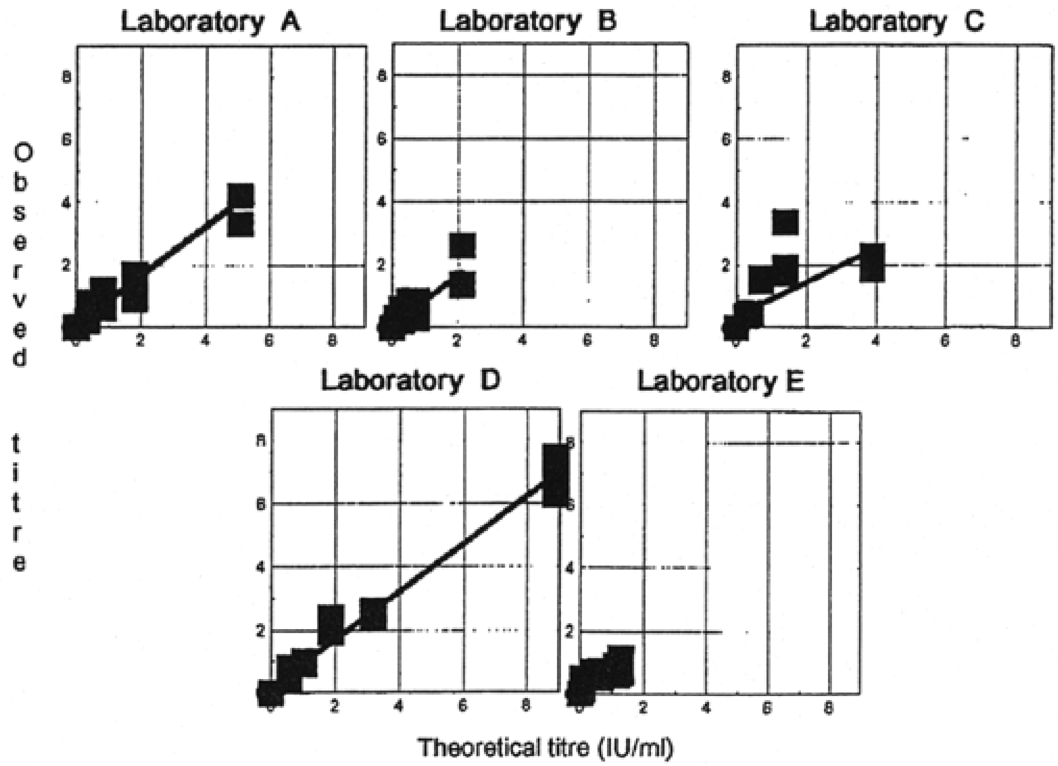


Figure 2: Comparison of results obtained by five participating laboratories, showing their intra- and inter-laboratory variations

One laboratory, different from that described above, has no correlation between titres and dilution factors.

In conclusion, the broad range observed in titres (Figure 1) is seen again during this blind test but the inter-test intra-laboratory repeatability and the intra-laboratory consistency are generally good. The difference observed in titres established in different laboratories should then come from differences in the test procedures.

Comparison of testing procedures

Most of the laboratories follow the principles of the rapid fluorescent focus inhibition test (RFFIT) as described by Smith *et al.* (1973) or Zalan *et al.* (1979). But in fact, the analysis of testing procedures shows few common points, as follows.

Cell culture support:

- six laboratories use 96 wells microtitration plates;
- two use Terasaki plates;
- no laboratory uses Lab Tek slides (which is the most frequently described technique!).

Dilution of serum:

- threefold dilutions are generally used in six laboratories, other dilution factors used include twofold (1 lab), fourfold (1 lab) and fivefold (2 labs);
- four laboratories (of 9) prepare a tenfold predilution and one uses a fourfold predilution;
- three laboratories make duplicates for each serum.

Challenge strain:

- CVS (challenge virus standard) is the challenge strain in eight laboratories;
- SAD (street Alabama Dufferin) is used in one lab;
- SAGI (SAD avirulent Gif-1) is used in one lab.

The challenge virus titre varies greatly from one laboratory to another. The comparison between laboratories is difficult because the calculation method is not standardised: median tissue culture infectious doses ($TCID_{50}$), 50 percent or 80 percent of fluorescence and fluorescent plaque forming units (PFU) are used. Two laboratories express their CVS titre in $TCID_{50}$, one uses 10 000 $TCID_{50}$ per plate and one uses 10 000 000 $TCID_{50}$ per plate. These two laboratories incubate the plates for 24 hours.

The neutralisation step is made at 35 to 37°C and for 60 to 90 minutes.

Incubation with cells:

Whatever the laboratory, a cell suspension is added to the tube or plate containing the serum and virus after the incubation step. The cell culture medium is MEM or DMEM with 5, 8 or 10 percent foetal calf serum. Two cell lines are used: BHK21 (7/9 laboratories) and BSR cells (2/9). The density of the cell suspension varies from 3×10^5 to 1.6×10^6 . The incubation time of the plate varies from 20 to 96 hours at 35 to 37°C.

Observation method:

The observation of the non-neutralised virus is made after fluorescent antibody staining of the fixed cell layer. Two reading techniques may be used:

i. Estimation of the general fluorescence of the cell layer.

Fluorescence is estimated by the proportion of fluorescent cells in the layer. This estimation may vary from one person to another. If a ratio is done when using a reference serum, the "subjective" part of the method disappears as shown in Figure 3.

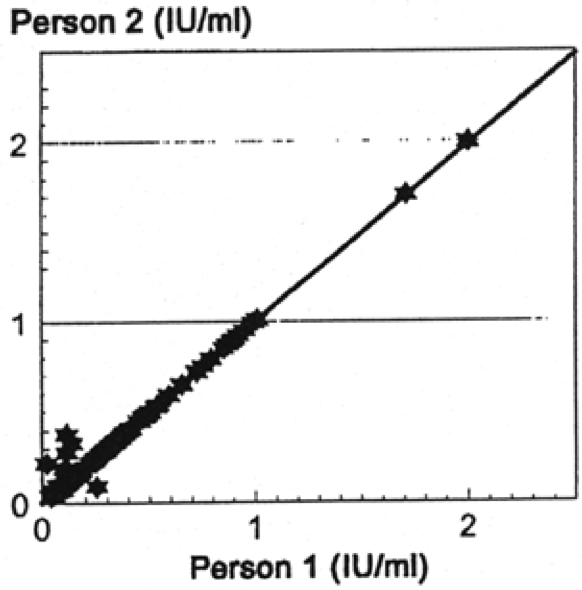


Figure 3: Results obtained independently by 2 persons when testing 70 dog serum samples in the same laboratory. The correlation factor is r = 0.99.

ii. "Numeration" methods

The method may be strictly qualitative to detect fluorescence in the complete cell layer of the well. The quantitative aspect is given by the number of replicates for every serum dilution. The calculation of the titre is then made with the Spearman-Kärber method or with a neoprobit graphic chart.

Another way is to observe 5 to 10 microscope fields in a well and to determine the proportion of positive fields (i.e. with fluorescence). In this method, the observed fields cannot be considered independent because they are all a small part of a complete layer that has grown in a single well. The observation mean must be reproducible both for magnification (objective, eyepiece and magnifying factors of the additives inserted in the optical path) and for the size of the field really observed. The diameter of the microscope visual field (in mm) is calculated as follows:

$$\frac{\text{eyepiece visual field number}}{\text{objective magnification.}}$$

Therefore, for a constant magnification rate of the optical system, according to the specifications of the eyepiece, this observed diameter may vary from 1 to 3 and the observed surface varies from 1 to 9!

iii. Titre of the challenge virus

If method (i) above is used for calculating and controlling the dose of virus, the titre is estimated from an absolute observation. The remark of point (i) shows that the dose of virus is "observer" related. Great variations are observed in challenge virus dose and in units. In order to have a rapid answer people use high doses of virus, the assumption being also that replication of virus in cells will not be disturbed by a poor quality scrum (presence of cytotoxic products or bacteria that are not eliminated by antibiotics).

Calculation of the titre of the serum:

In order for the neutralising power of the serum to be expressed in IU/ml, a reference serum is added and the titre is the result of the following ratio:

$$\frac{\text{dilution of the serum with 50\% inhibition of fluorescence}}{\text{dilution of the reference serum with a 50\% inhibition of fluorescence.}}$$

Under such conditions, the difference of estimation of fluorescence between persons (the "subjective" part of the reaction) disappears. The effective dilution of the serum is the one with a 50 percent inhibition of fluorescence. The calculation method is either a neoprobit graphic one or uses the Spearman-Kärber method.

Conclusions

Seroneutralisation detects both specific and non specific neutralising activity of a serum, which is important when estimating the immune status of animals, for instance, for cross border controls or as an alternative to quarantine measures. Great variations for *in vitro* neutralisation tests procedures are used between laboratories and therefore standardisation of techniques is important.

References

Atanasiu P. (1973) Quantitative assay and potency test of antirabies serum and immunoglobulin. In: *Laboratory Techniques in Rabies, 3rd Edition.* Edited by Kaplan, M.M. and Koprowski, H. pp. 314-318.

Smith LS., Yager P.A. and Baer G.M. (1973) A rapid tissue culture test for determining rabies neutralizing antibody. In: *Laboratory Techniques in Rabies, 3rd Edition,* Edited by Kaplan, M.M. and Koprowski, H. pp. 354-

Zalan E., Wilson C. and Pukitis D. (1979) A microtest for the quantitation of rabies virus neutralizing antibodies. *Journal of Biological Standardization* **7***:213-220.*

DATA GATHERING AND ANALYSIS FOR IMPROVED DECISION SUPPORT TO RABIES CONTROL

Brian D. Perry[1]

International Livestock Research Institute (ILRI), Nairobi, Kenya.

INTRODUCTION

In the effective control of animal diseases such as rabies, there are numerous decisions to be made by a variety of different categories of decision maker, each of which requires certain information to be available to optimise the quality of the decision taken. Information used is derived from data, and in an ideal world, it would clearly be desirable that data are collected on as many different aspects of disease occurrence as possible. In the control of rabies in the developing world, there is generally a considerable shortfall between the optimal range and quality of data, and those which are actually collected. On too many occasions we are either oversupplied with data we do not want, or undersupplied with data we need. Data collected in some countries have lost their immediate relevance, because the data collections system was set up years ago when the disease pattern was different when less was known about the epidemiology, and when the resources allocated to disease control were more plentiful for a smaller animal and human population. On equally many occasions we are undersupplied with data we do want because the diagnostic technology is inadequately disseminated, samples are not submitted for diagnostic procedures, or results are inadequately analysed or reported. Opit (1987) described Finagle's law, which although taking a rather pessimistic view of life, expresses sentiments that many Directors of Veterinary Services in the eastern and southern African region can probably identify with:

The information you have is not what you want;
The information you want is not what you need;
The information you need is not what you can get;
The information you can get costs more that you can pay.

So where are the priorities with regard to data for the most important decisions in rabies control?

As an aid to the process of evaluating data needs and the ways of collecting them, we review the decision making process for rabies control in developing countries, identifying the priority decisions to be made, the data required to make these, discussing how such data be gathered, and how these data can be analysed, synthesised and used. Issues will be dealt with in broad terms, as a discussion point for the more specific presentations from different countries to follow.

[1]Co-author: **J. J. McDermott,** Department of Public Health, Faculty of Veterinary Medicine, University of Nairobi, Kenya.

DECISIONS, DATA NEEDS AND DATA SOURCES

In considering the decisions, data needs and data sources for more effective rabies control, we will consider two major categories of decision as examples. These are:

1 What is the long-term national strategy to deal with rabies?
2. How should physicians or health clinic staff handle cases of human exposure to rabies?

What is the long-term national strategy to deal with rabies?

We have selected four components of this decision.

i. Choice of vaccine

The mainstay of rabies control programmes in much of the eastern and southern African region is the immunisation of susceptible domestic animal hosts, in particular dogs. The choice of which vaccine or vaccines to use in a given country or region should ideally depend on such factors as their efficacy, safety, lability, availability, ease of administration and cost, and the pertinent requirements for a given country should be articulated by its veterinary authorities. Although this happens in some countries, it is by no means universal, and the pharmaceutical industry, both directly and indirectly, exert influence over vaccine choice by their decision as to whether to pursue registration of their products or not. Thus the veterinary authorities may prefer a given vaccine, on the basis of the criteria listed above, but if a company does not wish to market it in that country for one reason or another, it will not be available. Information on the technical merit of vaccines submitted for registration is generally acquired through the pharmaceutical companies themselves, and unless a country requests specific registration trials, the efficacy and safety data are usually generated outside the region.

To compound these less than ideal circumstances, two other factors influence the choice of rabies vaccines used. The first involves government tenders. In many African countries, rabies vaccination is carried out primarily by the government veterinary service, which solicits tenders from interested pharmaceutical companies at regular intervals for the purchase of rabies vaccines. The tender is usually offered to one company, potentially affecting rabies control should the vaccine be ineffective for any reason, the "all eggs in one basket" syndrome. Furthermore. the successful tender is usually determined on the basis of cost, rather on technical merits.

The second factor affecting decisions on vaccine choice is when local production of rabies vaccine occurs. There has been a trend over the last 30 years to establish local animal disease vaccine producing units, to enhance national disease control capacities and to minimise the expenditure of foreign exchange on vaccine purchase. However, often the investment and operating costs for producing high quality rabies and other vaccines cannot be sustained by national markets. In some instances, these economic constraints have resulted in the production of inferior vaccines, seriously affecting the efficacy and public confidence in rabies control. We would suggest that for rabies vaccines, most countries would be better advised to maintain or obtain access to an up to date database of available rabies vaccines, including their costs and qualities. Choice of vaccine could then be determined by considering both economic and biological criteria.

ii. Target species

For much of the African region, the target species is the dog, responsible for maintaining rabies infection in most countries, and also for the vast majority of human exposures to rabies. Thus under most circumstances, the decision on target species is easy, and the crude species-specific incidence and human exposure data derived from passive disease surveillance systems serve to confirm this. However, there are some notable exceptions to this within the region where wildlife rabies has also been, or is currently, very important. It has either been demonstrated or is strongly believed that rabies is maintained in yellow mongoose (Chaparro and Esterhuysen 1993; King *et al.* 1993) and bat-eared fox (Thomson and Meredith 1993) populations in South Africa and in jackal populations in South Africa (King *et al.* 1993) and Zimbabwe (Foggin 1988; Bingham and Foggin 1993). It is assumed that rabies control in these wildlife reservoirs will require their own specific control programmes. For example, jackal rabies in Zimbabwe is considered to be of such importance that a specific research programmes to develop and deliver oral rabies vaccines for jackals is currently underway (Bingham *et al.* 1993).

Decisions on target species should be subject to periodic reviews. For example, in North America, rabies incidence has been markedly reduced in the original target species, dogs, but over time foxes, raccoons and skunks have become the main reservoir species. Now rabies control is directed both at these wildlife reservoir species and at dogs and cats which transmit rabies between these reservoirs and humans. In Africa, rabies control in the main reservoir species, dogs, may have important impacts on other target species. Despite much having been written, little is known about the outbreak of rabies in wild dogs in the Serengeti ecosystem. However, it has been assumed that the infection was derived from domestic dogs of the human habitations surrounding the parks and that dog rabies control will limit spread to wildlife species (Alexander *et al.* 1993; Gascoyne *et al.* 1993). 'This is an important consideration for many African countries in which the conservation of endangered wildlife species such as the wild dog is of critical concern

iii. Delivery of rabies vaccine: where, what age, what numbers and with what frequency?

One of the most important decisions in animal disease control is how to most effectively use the available resources to maximum effect, and for rabies control this translates into how best to deploy available vaccines. Frequently, 70 percent vaccination coverage is cited as the required threshold for controlling rabies in dog populations (WHO 1984). In this meeting, Coleman and Dye (see page 119) present a mathematical model indicating that this is a reasonable target. However, Perry (1992) showed that most countries in eastern and central Africa were not even coming close to achieve this figure. So what information can enhance the decision on deployment of vaccines to improve the situation, and how can such data be gathered?

Vaccination programmes generally are directed at entire countries, and do not vary their geographical focus from year to year. However, Perry (1993) has argued that vaccination would be more effective if targeted to certain high risk communities, placed at higher risk on the basis of geographical location or socio-economic status. High quality and up-to-date reporting of the location of rabies cases can, in some circumstances, identify the direction of movement of outbreaks, and areas of particular risk where special vaccination efforts could be undertaken. This can be achieved through passive disease surveillance schemes, and has been demonstrated to effect in Zimbabwe. A locational identity or grid reference must be provided with each suspect rabies submission, and this location entered into a geographical information system (GIS). Important components of georeferenced rabies incidence data are that georeferencing is accurate, that the incidence reporting is widely and regularly practised, and that summary distribution maps for appropriate time periods (such as weekly or monthly) are prepared and analysed.

The actual requirement of the number of dogs or other target species to vaccinate depends on the population, the age of animals to be vaccinated and the frequency of vaccination. However, the decision on the number of animals to vaccinate is generally determined solely by the government budgetary allocation. But what is the actual requirement? Dog censuses are almost non-existent in the region, being too costly and of too low a priority. The only census reported in recent years had been that of Brooks (1990) as part of a research project in Zimbabwe. For routine population estimates it will be necessary to apply other methods to estimate the size of the dog population. The most commonly used technique has been to extrapolate from the human population by a common dog:human ratio, estimated by Bögel *et al.* (1982) to be in the range of 1: 8 to 1: 11. In areas in which either the human population is not well known or variations in this ratio are expected, the population could be estimated by standard statistical sampling methods. Such methods were used by Kitala and McDermott (see page 95) to estimate dog numbers and densities in Machakos District. Regardless of the method used, it should be appreciated that for most areas of Africa, dog populations are increasing. Estimates of increase range from 4 percent in Zimbabwe (Brooks 1990) and 4.6 percent in northern Tanzania (Gascoyne 1994) to 9 percent in Machakos, Kenya (Kitala and McDermott. page 95). From these studies it is not clear whether dogs are increasing at a similar or faster rate than human populations. Further case studies within this region will help to provide better information to guide dog population estimation.

In targeting scarce vaccination resources better, decisions need to be made on what age groups to vaccinate. This depends on the age-specific incidence of rabies, the age-structure of the dog population, and the frequency of vaccination. Currently, there is little good information on the age-specific incidence of rabies in the region. The exception is in Zimbabwe, where Foggin (1988) estimated age-specific proportions as 4 percent for dogs less than 4 months, 20 percent from 4 to 12 months and the remaining 76 percent greater than one year. Kitala (personal communication 1995) estimates similar age-specific proportions in Kenya. Even though puppies and very young dogs appear to be at relatively low risk of rabies, the annual (or even six-monthly) vaccination campaigns in many countries results in young dogs being at very high risk well before the next campaign. Thus, information on the age-structure and population turnover of dog populations is of great importance. Fortunately, there have been a number of recent studies on different dog populations in the region (Brooks 1990; Butler, page 81; Gascoyne 1994, Kitala *et al.* 1993;

Kitala and McDermott, page 95) that provide useful data. In these studies, consistently high fecundity and mortality of dogs have been reported, with more than half the dogs in a population being less than one year of age. This means that by the next annual vaccination, more than half the dogs will have been replaced.

This rapid turnover of most African dog populations indicates that annual vaccination would be inadequate to maintain the desired 70 percent vaccination cover. Currently, the annual frequency of most rabies vaccination campaigns are determined more by tradition and convenience than by the epidemiology of the disease. For cost and logistic reasons, it is common to combine rabies campaigns with the immunisation of other livestock against anthrax, blackleg and other diseases. However, it seems that under most circumstances at least a six-monthly vaccination frequency is required.

Even with vaccination twice annually, the large proportion of young dogs presents a challenge to effective vaccination coverage. The minimum recommended age at vaccination for vaccines currently used ranges from 3 months for most inactivated tissue culture vaccines produced by commercial companies to 6 months for modified live vaccines produced by some national vaccine production units. This precludes a large proportion of dogs from being vaccinated. Vaccine regimes for younger dogs are urgently required (Perry 1995). Given that vaccination is uncommon in most areas in the region, the prevalence of maternal antibodies will generally be low so that the interference by maternal antibodies on the development of vaccinal immunity at the population level will likely be minimal. Some commercial vaccine producers indicate in their literature that dogs as young as 6 weeks of age could be vaccinated. Given the difficulties of mounting frequent (more than semi-annual) vaccination campaigns and the relatively young age of African dog populations, there are significant potential benefits to vaccinating younger dogs. These deserve further investigation through clinical trials to assess the efficacy and safety of different vaccines in puppies (with and without maternal antibodies). It is important that trials be conducted in the field since the responses of healthy laboratory dogs might differ from dogs kept under natural conditions.

iv. Who should pay, and how much?

The decision as to whether rabies vaccination costs should be borne by the public sector, shared with dog owners, or combinations thereof, is one constantly facing governments and veterinary departments in the region, dealing with high public expectation of public sector veterinary services but diminishing financial resources. This decision has efficacy, economic and political implications. Data required to make this decision include the actual cost of vaccines, the size and extent of the rabies problem, the purchasing power (or willingness to pay) of different sectors of society, and the opportunities or constraints within the public sector budget. Within the region, the current practice varies from country to country, from free provision of rabies vaccine by many government veterinary departments, to the levy of a nominal charge for government delivered vaccination (as in Kenya, for example), to the full cost recovery from dog owners (as carried out by private veterinary practices in most urban centres of the region). In some countries, other organisations, such as societies for the prevention of cruelty to animals, are prepared to raise funds and provide logistical support. In general, we consider that there is much scope for improved funding through all possible sectors if the economic implications of rabies and the merits of alternative intervention options are better spelled out, through straightforward techniques such as benefit cost analysis. Such studies are relatively easy to perform, using data collected in surveys, and have been carried out for other animal diseases in the region (e.g. for theileriosis of cattle; Mukhebi *et al.* 1992).

How should physicians or health clinic staff handle cases of human exposure to rabies?

Again, as with the decision on national rabies control strategy, this decision has sub-components, the most important of which are: has an exposure to rabies taken place; and if so, what are the intervention options available?

i. Has an exposure to rabies taken place?

The data needed to arrive at a decision as to whether an exposure to rabies has occurred are derived from epidemiological principles, local knowledge and, in some cases, diagnostic evidence. Factors to be considered are the species and type of animal involved in the exposure (one which requires local knowledge of the occurrence of rabies at the time), whether an exposure sufficient to transmit rabies virus occurred (one which is based on epidemiological principles), the current disposition of the animal (is it available for observation or diagnostic testing), and whether reliable diagnostic services are available (Perry 1987).

In many countries of the region, there are insufficient data to elaborate a detailed decision matrix using these factors. As far as species and type of animal are concerned, when wildlife species are involved in the exposure, it is difficult not to consider virtually all as potentially rabid. In North America, where wildlife rabies is endemic, epidemiological evidence suggests that although rodents and lagomorphs are frequently responsible for biting humans, these bites are often provoked and present minimal risks of rabies infection. However, in the recent study of Kitala *et al.* (1994), they recorded an unusually high number of confirmed rabies cases in rodents, suggesting that the extrapolation of the North American evidence to be unwise. Clearly, more and better data on species-specific incidence are required. As in North America, wild carnivores and bats appear to present a considerable risk, and the length of time they excrete rabies virus prior to the onset of clinical signs is unknown; thus any wild animal exposing a human to rabies virus should be destroyed and tested if feasible, and if rabies testing facilities are available.

With domestic dogs and cats, more is known on the excretion of rabies virus in saliva, and its association with behavioural changes, which, in combination with the feasibility in some facilities to quarantine these species, has led to less drastic, although more complex, strategies to be adopted in many countries. However, these strategies may not be completely appropriate for many circumstances in eastern and southern Africa, where recovery from rabies with virus excretion has been reported (reviewed by Fekadu 1988), the prevalence of which is unknown. Clearly, there is an urgent need for more data through well-structured epidemiological studies.

ii. If exposure to rabies has occurred, what intervention option(s) should be initiated?

Data requirements for this decision include clinical data (such as current vaccinal status, the site of exposure and time since exposure) combined with epidemiological principles (which will determine, for example, whether vaccine alone or vaccine and rabies immune globulin should be administered), inventory data (such as what vaccines and tested regimens are available) and economic data (such as the cost of the vaccine to the patient, the cost of transport to a clinic, and the "affordability" of vaccination to different sections of society).

DATA GATHERING, ANALYSIS, SYNTHESIS AND USE

The quality of information on key epidemiological features of rabies varies widely from country to country as manifested in the country reports presented at this meeting. One essential requirement for improved information is the development of standardised disease reports, based on WHO (1992) data requirements. Such a report would need to include some of the key data needs discussed above (species, age, location, contacts, etc.) as well as being concise and simple to complete.

In our opinion, passive surveillance should be continued but will need to be supplemented with selective active surveillance in representative regions to estimate the degree of under-reporting and other potential deficiencies of passive reporting systems. For example in Kenya, Kitala *et al.* (1994) noted that an active surveillance reporting system for rabies uncovered 40 times more rabies cases despite only 60 percent of potential cases identified being diagnosed in the laboratory. The problems of passive surveillance are numerous, but a fundamental concern is that the need for rapid diagnosis to guide the post-exposure human treatment of animal bites and the tracking of rabies outbreaks, for which many standard rabies diagnosis and reporting systems were designed, is not being fulfilled. If passive surveillance is to be relied upon in decision making, the needs of its clients and users must be served by the data gathering and reporting system. For individuals subjected to a potential rabies exposure, the expectation of a prompt diagnosis and report plus accurate information on the costs and availability of post-exposure treatment would be a strong incentive to report exposures. For communities and local veterinary officials, prompt reporting of rabies cases is invaluable for the planning and publicising of rabies control campaigns, especially in response to outbreaks. Such feedback is essential if a viable and sustainable surveillance system is to be developed. Given the scarcity of government resources and the logistical difficulties of delivering vaccination, community involvement is important for any rabies control programme to be effective. We believe that communities could play a valuable role in rabies surveillance if their needs were addressed and included in such a system.

For the rapid dissemination of rabies reports, some degree of computer processing will be required. The current rabies reporting system in Zimbabwe (Bingham 1993) provides an example of what can be achieved with modest resources, made more cost-effective if the system is used for multiple diseases. One software package, EMINFO, produced by the Centers for Disease Control in the United States in conjunction with the World Health Organization and currently in its 6th version, combines modules for entering, storing and manipulating data, calculating descriptive and simple analytic statistics and generating reports. It can also be linked with MAPINFO, a companion mapping software programme. Both programmes are public-domain software that are widely available at subsidised cost.

For policy makers, economic analyses such as cost-efficacy data on post-exposure treatment regimes and cost-benefit analyses on alternate rabies control strategies are required (Fishbein *et al.* 1991*;* Meslin 1994*).* Currently, such analyses are mainly limited by the lack of high quality epidemiological data on the incidence and risk of rabies using different control and treatment options. Some of these data could be generated in the region, but others, for example the efficacy of potential useful and cheaper post-exposure treatment regimes, such as the reduced dose intradermal regime of Chutivongse *et al.* (1990)*,* can be extrapolated from other areas. The limited budgets of most government health and veterinary services and the tremendous financial resources required to control other health problems in the region, means that any proposals for rabies control programmes will need to be well justified economically.

As evidenced at this meeting, improvements in data gathering, analysis, synthesis and reporting are required in many countries of the region. Common rabies case and report forms using standard protocols, would enhance both national and regional rabies control efforts and would be a valuable contribution of the Southern and Eastern Africa Rabies Group. Shared databases on available vaccines, post-exposure treatment regimes and rabies incidence and spread within the region could be developed and would serve to highlight the qualities and deficiencies of available and potentially available data on rabies in the region.

REFERENCES

Alexander K.A., Smith J.S., Macharia MJ. and King. A.A. (1993*)* Rabies in the Masai Mara, Kenya: preliminary report. *Onderstepoort Journal of Veterinary Research* **60***:411-414.*

Bingham J. (1993) Rabies in Zimbabwe. In: *Proceedings of the International Conference on Epidemiology, Control and Prevention of Rabies in Eastern and Southern Africa, 2-5 June 1992, Lusaka, Zambia.* Edited by King A.A. Edition Fondation Marcel Mérieux. Lyon. pp. 29-33*.*

Bingham J. and Foggin C.M. (1993) Jackal rabies in Zimbabwe. *Onderstepoort Journal of Veterinary Research* **60***:365-366.*

Bingham L, Perry B.D., King A.A., Schumacher C.L., Aubert M., Kappeler A., Foggin CM, Hill F.W.G. and Aubert A. (1993) Oral rabies vaccination of jackals: progress in Zimbabwe. *Onderstepoort Journal of Veterinary Research* **60***:477-478.*

Bögel K., Andral L., Beran G., Schneider L.G. and Wandeler A.I. (1982) Dog rabies elimination: a trend analysis and programme proposal prepared by a WHO working group. *International Journal of Zoonoses* **9***:97-112.*

Brooks R. (1990) Survey of the dog population of Zimbabwe and its level of rabies vaccination. *Veterinary Record* **127***:592-596.*

Chaparro F. and Esterhuysen J.J. (1993) The role of the yellow mongoose *(Cynictis penicillata)* in the epidemiology of rabies in South Africa - preliminary results. *Onderstepoort Journal of Veterinary Research* **60**:373 -377.

Chutivongse S., Wilde H., Supich C. and Baer G.M. (1990) Postexposure prophylaxis for rabies with antiserum and intradermal vaccination. *Lancet* 896-898.

Fekadu M. (1988) Pathogenesis if rabies virus infection in dogs. *Reviews of Infectious Diseases* **10**:S678-S683.

Fishbein D.B., Frontini M.G., Doggins LG, Collins E.F., Quiroz-Huerta G., Gamez Rodriguez LL, Woo Ming B., Garza Ramos J, Belotto A1, Balderas Torres LM. *et al.* (1991) Rabies control in the Republic of the Philippines: benefits and costs of elimination. *Vaccine* **9**:581-587.

Foggin C.M. (1988) Rabies and rabies related viruses in Zimbabwe: historical, virological and ecological aspects. PhD thesis. University of Zimbabwe.

Gascoyne S.C. (1994) Rabies in the Serengeti Region of Tanzania. Final Report to the 0DA/NRRD Animal Health Programme. Institute of Zoology, Regent's Park and London School of Hygiene and Tropical Medicine. 71 pp.

Gascoyne S.C., King A.A. Laurenson M.K., Borner M., Schildger B. and Barrat J. (1993) Aspects of rabies infection and control in the conservation of the African wild dog *(Lycaon pictus)* in the Serengeti region, Tanzania. *Onderstepoort Journal of Veterinary Research* **60**:415-420.

King A.A., Meredith C.D. and Thomson G.R. (1993) Canid and viverrid rabies viruses in South Africa. *Onderstepoort Journal of Veterinary Research* **60**:295-299.

Kitala P.M., McDermott J.J., Kyule M.N. and Gathuma J.M. (1993) Features of dog ecology relevant to rabies spread in Machakos District, Kenya. *Onderstepoort Journal of Veterinary Research* **60**:445-449.

Kitala P.M., McDermott J.J., Kyule M.N. and Matere C.N. (1994) Investigation of rabies incidence and control by active surveillance in Machakos District, Kenya. In: *Proceedings of the 7th International Society for Veterinary Epidemiology and Economics Symposium, 15-19 August 1994, Nairobi.* Edited by Rowlands GJ, Kyule M.N., Perry B.D. *Kenya Veterinarian* **18**:528-530.

Meslin F-.X. (1994) The economic burden of rabies in developing countries: costs and benefits from dog rabies elimination. In: *Proceedings of the 7th International Society for Veterinary Epidemiology and Economics Symposium, 15-19 August 1994, Nairobi.* Edited by Rowlands GJ, Kyule M.N., Perry, B.D. *Kenya Veterinarian* **18**:523-527.

Mukhebi AM., Perry B.D. and Kruska R.L. (1992) Estimated economics of theileriosis control in Africa. *Preventive Veterinary Medicine* **12**:73-85.

Opit L.J. (1987) *World Health Forum* **8***:* 409-417.

Perry B.D. (1987) Rabies. In: *The Veterinary Clinics of North America, Small Animal Practice.* Edited by August J.R. and Loar A. S. **17**:73-89.

Perry B.D. (1992) The epidemiology of dog rabies and its control in eastern and southern Africa. In: *Proceedings of an International Conference on the Epidemiology, Control and Prevention of Rabies in Eastern and Southern Africa, Lusaka Zambia, 2-5 June 1992.* Edited by King A. Editions Fondation Marcel Merieux, Lyon, pp 107-121.

Perry B.D. (1993) Dog ecology in southern Africa: implications for rabies control. *Onderstepoort Journal of Veterinary Research* **60**:429-43 6.

Perry B.D. (1995) Rabies control in the developing world: can further research help? *Veterinary Record* **137**:521-522.

Thomson G.R. and Meredith C.D. (1993) Rabies in bat-eared foxes in South Africa. *Onderstepoort Journal of Veterinary Research* **60**:415-420.

WHO (1992) Report of the WHO Expert Committee on Rabies. WHO Technical Report Series, No. 824. World Health Organization, Geneva.

WHO (1984) Guidelines for dog rabies control. World Health Organization, Geneva, (VPH/83.43).

RABIES SURVEILLANCE IN EUROPE

J. Barrat[1]

Laboratoire d'Etudes sur la Rage et la Pathologie des Animaux Sauvages, CNEVA Malzeville, France.

Rabies in Europe is mainly a disease of wildlife, being maintained by the red fox. Canine rabies is limited in distribution and remains sporadic, although it is important in Turkey.

Four meetings on Rabies Control in Europe have been organised in Strasbourg (1985), in Annecy (1988), in Nancy (1991) and in Piestany in Slovac Republic (October 1993). These meetings were conducted to inform veterinary services about the situation of rabies control in the European countries and to allow exchange of information.

Thirty-five heads of the veterinary services of the different countries were sent questionnaires. The different data presented in this paper are a summary of the data collected from the 22 countries that answered the questionnaire, i.e. Austria, Belgium, Croatia, Denmark, Estonia, Finland, France, Netherlands, Hungary, Italy, Latvia, Lithuania, Norway, Portugal, Czech Republic, Romania, United Kingdom, Slovak Republic, Slovenia, Sweden, Switzerland and Turkey.

The first step of rabies surveillance involves diagnosis, reporting of cases and exchange of epidemiological information. In the second step, the aim of veterinary services is to reduce the spread of the disease, the risk of domestic animal contacts and finally the risk of human contacts because most often, humans are contaminated through domestic animal contacts.

Diagnosis of rabies

Most of the 22 countries that answered have fewer than 10 diagnostic laboratories, the exceptions being Romania and Lithuania that have 49 and 42 diagnostic centres respectively. This means that most often a centralised structure exists for rabies diagnosis, which allows the regular training of technicians necessary for a reliable experimental diagnosis. In France and in Lithuania, specimens from animals that have caused human contacts are treated in special laboratories. In France, for instance, cases with human contact are diagnosed in laboratories under the Ministry of Health, while epidemiological diagnosis falls under the Ministry of Agriculture.

The fluorescent antibody test (FAT) is used in all the countries, on all the specimens. Immunoenzymatic tests for diagnosis are regularly used in France and Portugal. Nineteen countries use the mouse inoculation test while 6 use the cell culture test on a regular basis.

[1] Co-author: **M.F. Aubert** Laboratoire d'Etudes sur la Rage et la Pathologie des Animaux Sauvages, Malzeville, France.

Histology is performed in 9 countries: 2 of them use Seller's staining technique, 1 uses Mann's technique.

Sixteen out of 21 countries regularly type the strains isolated. This operation is most often made abroad using monoclonal antibodies rather than molecular biology.

Not all the techniques are used for every specimen. Table 1 summarises the average use of the different techniques according to the purpose of the diagnosis.

Table 1. The use of the different diagnostic techniques in 22 European countries according to the purpose of the diagnosis. Figures are percentage of countries.

	FAT	Histology	Mouse inoculation test	Cell culture test	Immuno-enzymatic test
Human contact	100	44	80	50	8
Domestic animal contact	100	44	67	46	8
Epidemiological Surveillance	100	16	46	36	7
Oral vaccination Survey	71	9	33	40	9

Exchange of epidemiological data

Most of the countries regularly transmit epidemiological data to international organisations:
WHO (21 countries), OIE (19 countries) and FAO (11 countries).

Seventeen countries regularly exchange information on rabies epidemiology with neighbouring countries. The exchange is most often an official one using bulletins. Six of these countries also exchange data on an informal basis, using letters and telecommunication means. This information exchange has improved since 1991.

Control measures against human rabies

All the countries that answered the questionnaire practise post- exposure treatment for humans. Dogs, cats and cattle are the principal source of human contacts. Foxes are generally the fourth or the fifth ranking source of human contact.

Treatments are either given only in specialised centers (12/20 countries) or both in specialised centres and by doctors out of these centres (5/20). In the three remaining countries, treatment is given only by doctors out of these centres. Twenty to 100 treatment centres exist per country.

The average estimated cost of a treatment is 380 US dollars. Only five countries produce the rabies vaccine they use: France, Netherlands, Romania, Russia and Switzerland. The number of treatments is related to the type of rabies that exists in the country: an average of 2 000 treatments was performed in 1992 in the European countries that have fox rabies while in Turkey (a canine rabies country) 90 000 treatments were undertaken. Both in 1991 and 1992, 75 percent of human treatments used vaccine alone, antirabies serum plus vaccination was administered in 20 percent of treatments while the remaining 5 percent corresponded to the use of antirabies serum alone.

Most often the countries use a single vaccination schedule, 6 of them use 2 schedules and Latvia uses 3. Generally the immunisation schedules correspond to WHO recommendations:

- The 2, 1, 1 scheme with 2 doses on DO, 1 on D7 and 1 on D21 is used in 4 countries.
- 14 countries use 5 injections at days 0, 3, 7, 14, 30 or 6 with the last one at D90.
- Daily injections are administered in Lithuania for 25 days and in Latvia for 7 or 28 days.

Fifteen countries use the intramuscular route of injection, 3 others the subcutaneous one. When daily injections are administered, the subcutaneous or the intraperitoneal route are used. None of the countries indicated that the intradermal administration route is used.

Vaccines used for humans are controlled using the NIH test (8/14) or the European pharmacopoeia test (7/14). Five countries use both tests for controls. The Habel test is used in only one country. These controls are performed in official laboratories of the country (13/17) and in the producer's own laboratories (8/12). All batches of vaccine are controlled.

When antirabies, serum is needed, 14 out of 22 countries use globulins of human origin. The rest use globulins of animal origin. No countries use both human and animal globulins. The serum is administered intramuscularly either by general route only or using a partial local administration also.

The only postvaccinal problems that are reported are local reactions alone (7.4 percent) or with a general reaction (2.2 percent). Fever is observed in 1. 1 percent of cases.

Control measures against domestic animal rabies

Most often vaccination of domestic animals is at least advised. Vaccination of dogs is compulsory in 13 countries, at least in the rabies contaminated part of the country.

Vaccination is always performed by veterinarians helped by technicians in two countries. The owner of an animal can never vaccinate a domestic animal against rabies. Antirabies vaccination of domestic animals is under state control in 80 percent of the countries.

The first vaccination of domestic carnivores occurs between 2 to 6 months of age (92 percent of countries, while half of all countries vaccinate at 3 months) and boosters are given yearly (85 percent). Vaccination of domestic herbivores are undertaken between 1 and 6 months of age (6 months in 50 percent of countries), and boostered yearly (90 percent).

Once vaccinated, animals must be identified to validate antirabies vaccination in 16 countries (Table 2).

Table 2. The means used to identify animals and determine their vaccinal status in European countries.

	Tattoo or electronic chip	Tag with number	Description	Verification by owner
Dog	4/16	11/16	15/16	3/16
Cat	3/16	4/16	14/16	3/16
Cattle	1/16	10/16	4/16	1/16
Horse	2/16	2/16	6/16	3/16

When a previously vaccinated domestic animal is exposed, it may be killed (in all species or livestock only). Most often, it is observed for 2 to 6 months sometimes with a booster injection of vaccine. If an unvaccinated domestic animal is exposed, 3 countries allow post-contact antirabies vaccination.

Active measures against fox rabies

1. Fox population control

Different ways to control fox populations may be used in the European countries according to the type of rabies. This information is summarised in Table 3.

The raccoon dog is considered as a possible vector of rabies in Finland, Latvia, Lithuania and Sweden where they are shot, trapped and gassed.

Table 3. The numbers of European countries which practice the different methods of fox population or rabies control.

	Shooting	Gassing	Poisoning	Trapping	Oral vaccination
Fox rabies	18/20	3/16	1/12	9/16	17/18
Dog rabies	8/11	1/9	1/9	5/9	8/9
Bat rabies	0/7	0/7	0/7	0/7	0/4

2. Oral vaccination of foxes

Most European countries that experience fox rabies either use or plan to use oral vaccination of foxes. They will carry out these campaigns until eradication of the disease has been achieved or at least maintain tests in experimental areas. Austria, Belgium, Denmark, Estonia, Finland, France, Netherlands, Hungary, Italy, Latvia, Lithuania, Czech Republic, Slovak Republic, Slovenia and Switzerland submitted data on this topic.

When such campaigns are undertaken, most often central governments finance the campaigns, sometimes together with regional structures. The European Union participates financially in the vaccination campaigns in 6 out of 15 countries.

The total area treated in Europe in 1991 was 250 000 square kilometres and in 1992 it was 400 000 (a 60 percent increase). Baiting was done on foot (9 countries) and sometimes also by vehicle (2 countries). Five countries used aerial distribution, only one used helicopter baiting. State agents distributed baits in eight countries and volunteers in nine. In four countries baiting was performed both by state agents and volunteers. The average baiting density was 16 baits/km^2. Campaigns were usually carried out in spring and in autumn, Finland and Netherlands distributed baits only in autumn while Latvia distributed only in spring, all other countries conducted two campaigns a year.

Reports of contacts of domestic animals with vaccine baits are infrequent. The control of efficacy of oral vaccination campaigns is performed both by studying the rabies cases and also by studying specially sampled foxes. These foxes are shot during day (11 countries) and also during the night (8 of the 11 countries), the average sampling pressure is 0.05 animal/km^2. Most often a single laboratory deals with the control of oral vaccination campaigns. The tetracycline used as a marker in baits is sampled either in teeth (5 countries) or in bones (9 countries, mainly the mandible); three countries test both teeth and mandible. Serological testing of foxes is carried out in 11 countries, most often using a cell culture test, sometimes by ELISA or mouse inoculation test. Eight countries look for the possibility of vaccinal rabies with monoclonal antibodies.

Average bait uptake is 60 percent for the first campaign and reaches 72 percent at the fifth. Over the same period the average seroconversion rates are 55 percent and 66 percent respectively. The average cost of oral vaccination per square kilometre may be divided as follows:

- US$ 15.5 for the vaccine itself
- US$ 9 for the bait
- US$ 3.6 for the organisation of vaccination campaigns

Since the decrease of rabies, the number of foxes killed for analysis is decreasing in spite of the fact that foxes may be killed all year round in the majority of countries. Hunters' testimony and scientific studies have shown that fox densities have increased since the decrease of rabies.

Active measures against dogs in cases of dog rabies

If canine rabies occurs, half of the countries would begin culling operations by shooting and/or trapping dogs. Vaccination would become compulsory in all the countries, oral vaccination of dogs would be performed only in Turkey which already has canine rabies. In all the countries, all the dogs would be kept on leash. A tax would be imposed in 60 percent of the countries.

Table 4. The number of countries and the doses of vaccine of the different strains used during 1991 and 1992.

	1991		1992	
	Countries	Doses	Countries	Doses
SAD Bern	0	0	Czech Republic	885000
SAD B 19	6	1990000	7	1950000
SAG1	2	1020000	2	1360000
VRG	2	1070000	2	1570000
Experimental. vaccine from Bielorussia	Latvia	14210	0	0
SAD32	Slovak Republic	11000	Slovak Republic	100000

Research into rabies control

Fifteen countries have research centres that work on rabies. These centres study the following topics:

- diagnosis (15 countries)
- vaccine control (13 countries)
- epidemiology of rabies (12 countries)
- bat rabies (10 countries)
- oral vaccination and parenteral immunisation (8 countries)
- monoclonal antibodies (7 countries)
- molecular biology (6 countries)
- quarantine efficacy (4 countries)

Among the possible research fields in rabies, heads of veterinary services were asked to indicate the ones that they considered as important for future research. The first ranked was epidemiology of rabies followed by oral vaccination, diagnosis and surveillance, vaccine control and vaccine improvement.

RABIES SURVEILLANCE IN SOUTH AFRICA

George C. Bishop

Allerton Regional Veterinary Laboratory, Cascades 3202, Pietermaritzburg, South Africa.

In most of South Africa, rabies is spread by wild life and there is little that can be done to regulate and control this form of the disease. Surveillance in these areas is based mainly on keeping records of the confirmed cases and we have data indicating the exact geographical location, the date of death and the species of all cases since the late 1920's. Pro-active surveillance is more important in Natal, however, because canine rabies is endemic in the entire province and in areas adjacent to it, information on dog ecology, the movement and number of dogs and the levels of vaccination form an integral part of the control measures which must be applied.

We have conducted a number of surveys in Natal over the past few years. The human to dog ratio is being calculated based on a survey conducted in the schools falling in 62 of the 65 districts of the province. The eldest child from each family was asked two questions, namely: "how many people live in your home?" and "how many dogs do you have?" About 78 000 questionnaires were completed in 517 schools and from this information, we should soon be able to estimate the human to dog ratios of the various racial groups in urban, peri-urban and rural communities. Our last national census was held in 1991 and using that data, along with the known geographical areas of the census districts will enable us to calculate the dog densities. This survey covered nearly 250 000 people and indications are that there may be a human to dog ratio as high as about 4,1 humans to each dog. Previous field counts put Natal's dog population at about 600 000 whereas our survey results indicate a population which may be as much as three times higher than that figure. The data obtained in this manner will be compared to figures obtained from other surveys, still to be held, based on classical sampling/ surveillance approaches.

We have also run a number of surveys at vaccination points in order to improve our control strategies. Very useful information has been obtained by asking questions such as:

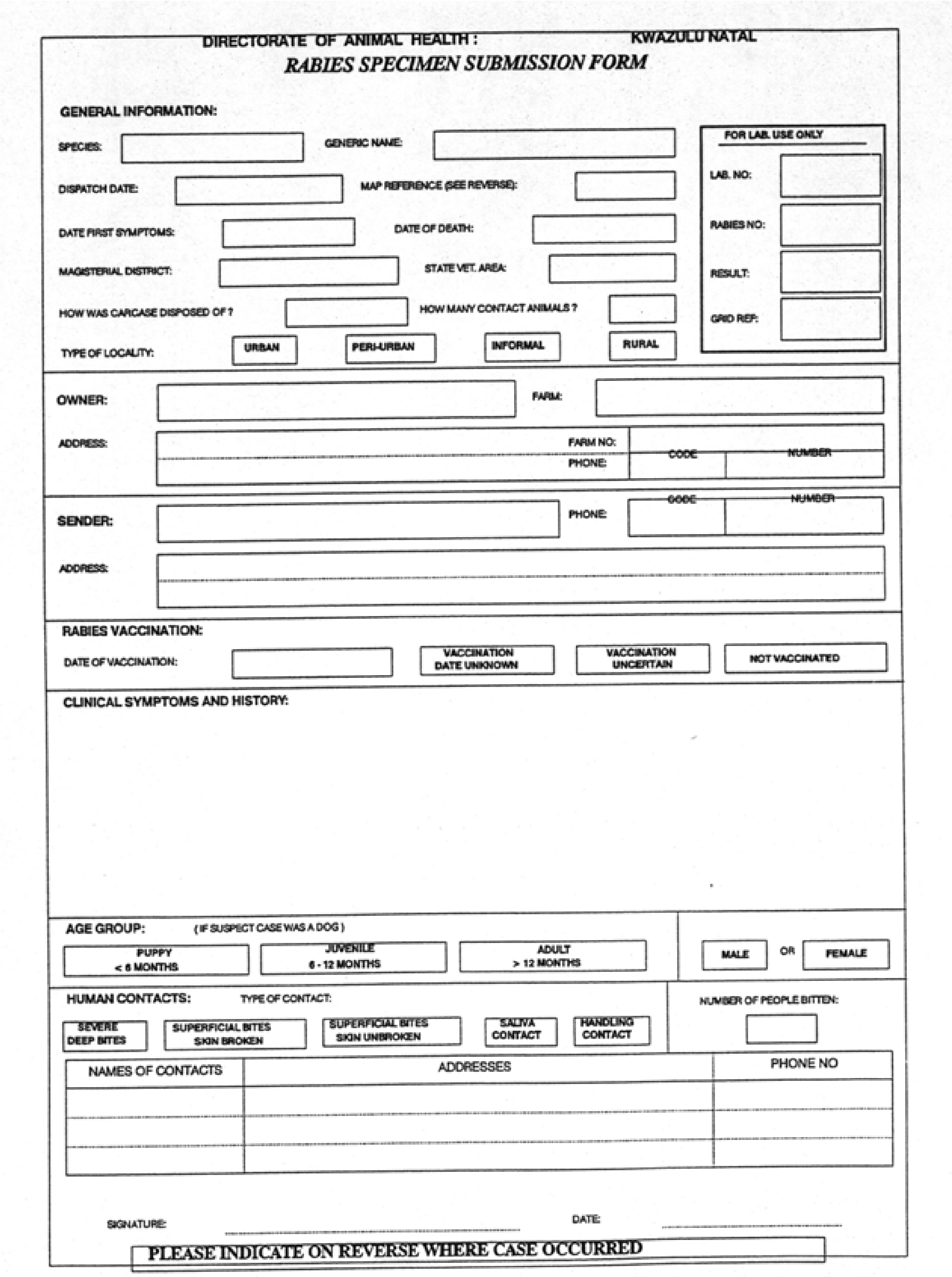
1. How far did you travel?
2. How did you travel?
3. How did you hear about this clinic?
4. Which days and what times will be most suitable for you to bring your dog?
5. What suggestions have you got which could help us to improve our service?

From the answers received to date, it appears that most people walk their dogs less than one kilometre and week-ends would be most suitable for dog vaccinations. Advertising techniques vary from one area to the next and we make use of loud-hailers, posters and the print and electronic media. Announcements made at schools are particularly effective in informing the communities. In general, winning the co-operation. of the communities so as to gain maximum voluntary response seems to be far better than making use of legislation to enforce our will. Collecting data such as the ages of dogs presented for vaccination, the number of lactating bitches and the sex ratios of dogs has taught us a lot about our dog population. We are trying to establish how often vaccination clinics must be provided in the high prevalence areas so as to be most effective. It would appear that because of the high turn-over rates in some areas, it is necessary to visit certain localities up to three or four times each year.

A well-designed rabies specimen submission form can be of tremendous value in collecting relevant data. If some careful thought is given to the design of such a form, particularly with a view to obtaining exact information in a user-friendly manner, the results can be most rewarding. Most of our specimens are submitted by relatively few people and these people have been schooled, coaxed and cajoled into filling the forms correctly. It is also important to standardise codes and fields used in data compilation if more than one laboratory is engaged in rabies diagnostics, so as to facilitate exchange of data and information. The form we use in Natal (Figure 1) has proved very successful because it has enabled us to access a lot of data that we have been able to put to good use. The information is entered on computer by a clerical assistant and the codes are then converted by the computer. Instead of asking for the exact grid reference, we request that a cross indicating the position of the case be placed on the map (Figure 2) which is on the reverse of the form. The block is recorded by our receptionist as a letter and number combination and the computer programme then converts the block code, say "D 14" to a quarter degree block reference. This approach circumvents the age-old problem of confusing degrees east with degrees south and reduces the dangers of "operator flair" in specifying the location!

By making use of this approach, we now have the date, species, quarter-degree location and recorded symptoms of every confirmed rabies case that has occurred in Natal since the disease was first diagnosed in Natal in 196 1. This total now stands at nearly 3 600.

Easily accessed, accurate and complete data is indispensable in the fight against rabies. It can also make the task of motivating an increase in budget or staff requirements a much simpler
task.


DIRECTORATE OF ANIMAL HEALTH : KWAZULU NATAL

RABIES SPECIMEN SUBMISSION FORM

GENERAL INFORMATION:

SPECIES: GENERIC NAME:

DISPATCH DATE: MAP REFERENCE (SEE REVERSE):

DATE FIRST SYMPTOMS: DATE OF DEATH:

MAGISTERIAL DISTRICT: STATE VET. AREA:

HOW WAS CARCASE DISPOSED OF ? HOW MANY CONTACT ANIMALS ?

TYPE OF LOCALITY: URBAN PERI-URBAN INFORMAL RURAL

FOR LAB. USE ONLY

LAB. NO:

RABIES NO:

RESULT:

GRID REF:

OWNER: FARM:

ADDRESS: FARM NO: PHONE: CODE NUMBER

SENDER: PHONE: CODE NUMBER

ADDRESS:

RABIES VACCINATION:

DATE OF VACCINATION: VACCINATION DATE UNKNOWN VACCINATION UNCERTAIN NOT VACCINATED

CLINICAL SYMPTOMS AND HISTORY:

AGE GROUP: (IF SUSPECT CASE WAS A DOG)

PUPPY < 6 MONTHS JUVENILE 6 - 12 MONTHS ADULT > 12 MONTHS

MALE OR FEMALE

HUMAN CONTACTS: TYPE OF CONTACT:

SEVERE DEEP BITES SUPERFICIAL BITES SKIN BROKEN SUPERFICIAL BITES SKIN UNBROKEN SALIVA CONTACT HANDLING CONTACT

NUMBER OF PEOPLE BITTEN:

NAMES OF CONTACTS	ADDRESSES	PHONE NO

SIGNATURE: DATE:

PLEASE INDICATE ON REVERSE WHERE CASE OCCURRED

Figure 1. Rabies specimen submission form.

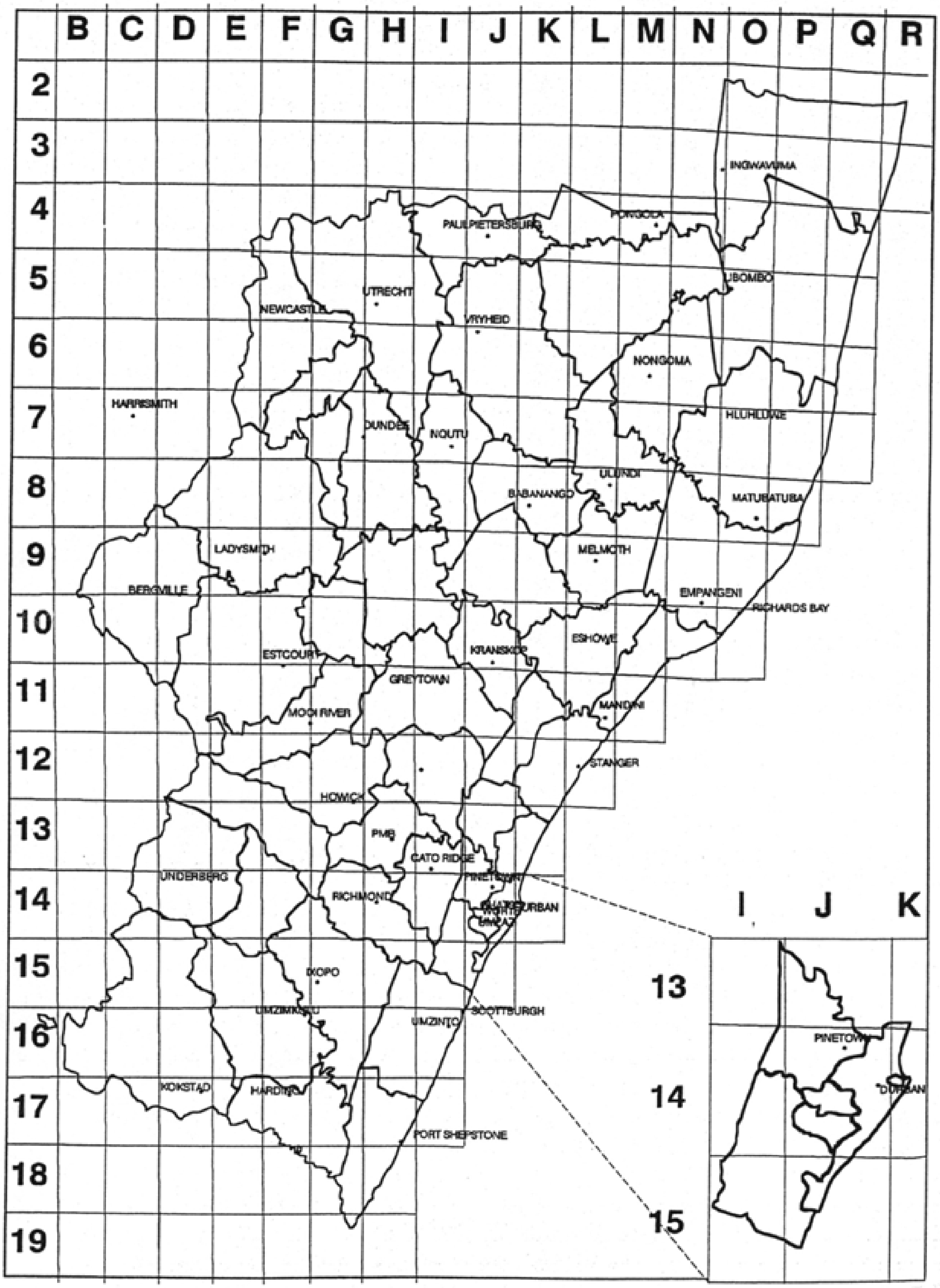


Figure 2. Rabies specimen form map.

RABIES SURVEILLANCE IN ZIMBABWE

John Bingham

Veterinary Research Laboratory, Harare, Zimbabwe

Introduction

Surveillance, in the veterinary or medical context is the monitoring of disease patterns to facilitate action. It encompasses field identification of diseases, laboratory confirmation, and dissemination of the information to the relevant authorities. It is, in turn, used in understanding disease processes, predicting disease occurrence and for making decisions on control or prevention of the disease.

Rabies has been present continuously since 1950 (see "Rabies control in dogs in Zimbabwe", page 134). Initially, diagnosis of rabies was carried out by histological tests and mouse inoculation. In 1967 the fluorescent antibody test was introduced and this soon replaced the histological test as the main diagnostic test in use for rabies (Foggin, 1988). In Zimbabwe, the case identification and reporting systems for rabies have been relatively efficient since the first introduction of the disease in 1950. This has allowed detailed studies of the patterns of rabies from the beginning of the outbreak (Foggin, 1988). During the mid-1980's the reporting system was computerised.

Structure of the Zimbabwe Department of Veterinary Services

There are three important branches of the Zimbabwe Department of Veterinary Services, not including the directorate, which are involved with rabies surveillance.

Firstly, the Field Branch, which identifies suspect rabies cases and submits the appropriate specimens to the laboratory. Three main levels of staff are involved in the disease identification procedure. Veterinarians are located at provincial or district level and are responsible for, apart from diagnostic functions, the major decision making processes at this level. Animal health inspectors (AHI's) are responsible for much of the routine livestock examination and disease monitoring, particularly in commercial farming and urban environments. Veterinary extension assistants (VEA's) are responsible within the communal areas for disease monitoring and extension work. They are based at Animal Health Management Centres (AHMC's), which are distributed strategically within each of the communal areas.

Secondly, the Diagnostics and Research Branch, under which falls the Rabies Unit, is responsible for diagnosis and reporting of confirmed cases and the management of the Rabies Surveillance database. The Rabies Unit is located at the Veterinary Research Laboratory in Harare. Its functions are overseen by a veterinarian and a technologist.

Finally, the Training Branch, which is responsible for the training of the field staff in identification of suspect rabies cases and the correct specimen sampling procedure.

Specimen sampling and submission

Specimens for rabies confirmation are submitted, as whole carcasses or as heads, by members of the public to veterinary offices. Alternatively, suspect rabid animals are collected by members of the staff of the Department. All provincial and district veterinary offices and AHN4C's are provided with post-mortem facilities and the equipment necessary for the removal and preservation of brains.

Kits for the preservation and transport of brain specimens are made up at the Rabies Unit in Harare and are transported to the provincial and district offices and to the AHMC's. The kits consist of wooden boxes painted red and the word "RABIES" printed boldly on the box. Fifty percent glycerol-saline and 10 percent formal-saline are placed separately into two half-litre glass jars (Consul, South Africa) which are placed into metal containers. The containers are placed into the boxes. To assist in preservative identification colour-coded labels, green for glycerol saline and white for formal-saline, are attached to each of the glass jars and the metal containers.

The whole brain is removed from the skull. It is bisected through the midline and one half is preserved in the 50 percent glycerol-saline solution, while the second half is preserved in the 10 percent formal saline solution. A Rabies Field Report form giving the name, address and contact numbers of the sender, details of the case and information on human contacts, is enclosed within the kit.

The kit is transported by rail or freight service to the Rabies Unit where the diagnosis is carried out. Delays in the transport of brain specimens often occur and are a major cause of poor quality specimens. Delays may happen before placing the brain into the preservative, where logistical problems occur in getting the head to a veterinary office or AHN4C which has a rabies kit or after preservation, in getting the full kit to a railway station or freight office. The former problem is usually the most serious in Zimbabwe, as this is where the longest delay will occur, and where most deterioration in specimen quality will occur. In rural areas which have poor infrastructure, veterinary personnel may carry specimens on buses, bicycles and by foot for several days before they get placed on a freight vehicle for transport to Harare.

Diagnosis

The Rabies Unit is manned every day of the week for rabies diagnostic tests. It is the only laboratory in Zimbabwe which routinely carries out such tests. Specimen kits are collected daily at railway and freight service depots.

The fluorescent antibody test (FAT) is the main test carried out for diagnosis of rabies. The mouse inoculation test (MIT) is used as a backup test on FAT-negative and inconclusive specimens. In addition, other non-routine procedures are occasionally used, including histological staining of paraffin embedded specimens, enzyme digestion of formalin-fixed specimens and skin biopsies on human ante-mortem cases.

Reporting

Results are initially reported to the sender of the specimen by telephone, usually within 24 hours of receiving the specimen. Results are reported as "positive", "negative by FAT", "uncertain, test to be repeated" or "unsuitable". If the mouse inoculation test subsequently shows the specimen to be positive, the sender is informed immediately by telephone. The "unsuitable" report is usually made with highly decomposed specimens or with formalin-fixed specimens where the FAT, following digestion, is negative.

The epidemiologically relevant details of each specimen are entered into a database (dBase 3 Plus, Ashton Tate) file. The fields of this file include specimen number, date received, sender's reference number, species, results of the different tests and the grid reference and locality of origin of the specimen. From the database a weekly report is generated which is sent to all the provincial and district veterinary offices. In addition, a quarterly report is generated for all the medical and veterinary authorities, the hospitals and the private veterinary practitioners. Both of these reports include a map of the case locations, generated by the Epidemiology Section of the Veterinary Research Laboratory.

On a quarterly basis, survey forms are sent out to all the hospitals to obtain information on the number of human rabies cases diagnosed, the number of patients receiving post-exposure prophylaxis and the numbers of vaccine and immunoglobulin doses used by the hospitals during the quarter. This information is compiled and reported in the quarterly report, giving total numbers used throughout the country. The questionnaire return rate for this survey is between 30 and 40 percent.

Reference

Foggin C.M. (1988) Rabies and rabies related viruses in Zimbabwe: historical, virological and ecological aspects. PhD Thesis, University of Zimbabwe, Harare.

NOTES ON DISCUSSION SESSIONS[1]

DISCUSSION OF NATIONAL REPORTS

Dr Karugah (Kenya).
Why are samples not reaching the laboratory?
There is not enough public awareness; people do not take an interest. There is also a lack of infrastructure to get samples to the laboratory, where the only tests available are fluorescent antibody and mouse inoculation tests.

Dr Munang'andu (Zambia).
There are more cases reported from Lusaka than other provinces. Why? Rural cases are likely to be much higher than reported.

What types of vaccine are used in Zambia? Lamb brain vaccine is used. BHK vaccine is difficult because the virus grows only to low titre. Rabisin is being imported.

Dr. **Hübschle (Namibia).**
There appears to be two cycles, urban (dog) and jackal to bovine. Is there jackal rabies in the absence of dog rabies in the Central Region? There is very little dog rabies: in the north the hit rate is 62 percent positive in dogs but only 18 percent in the Central Region, that is, in this region there is a great fear of rabies and if a case is suspected in a dog its head is cut off immediately.

Is the virus strain the same in dogs and jackals? Yes, identical.

Dr Bingham (Zimbabwe)
What skin biopsy was taken to confirm the three cases of human rabies? Full thickness biopsies, which included hair, were taken from the back of the neck.

Were serological tests and virus isolation tests on saliva carried out? No serological tests were done, saliva was tested from one of the patients: no virus was isolated.

How many days after clinical signs were the biopsies taken? All were advanced cases, one died a few hours after the samples were taken.

Dr Khomari (Lesotho)
What is the incidence in neighbouring countries? The incidence is high in Natal and our low incidence in Lesotho may also be due to our country's terrain. There may be rabies present in meerkats.

[1]Chief rapporteur: **Arthur A. King**

Is the reporting accurate? We do not seem to have any rabies.

Dr Swanepoel: Rabies control was good and cases declined even before the vaccination level fell.

GENERAL DISCUSSION OF NATIONAL REPORTS

An incentive for vaccination would be to shoot dogs, then the other owners would bring their dogs forward.

Dr Illango: After injection dogs do not seem to run as before, possibly due to a localised swelling from vaccination.

Dr Perry: As one of those who has attended the entire series of SEARG rabies meetings, I have had the opportunity to compare this session on the country reports of rabies and its control within the region with those of previous meetings and I conclude that this morning we have heard some good news, and some bad news.

First the good news! We have heard of dramatically enhanced rabies reporting in Uganda, if we compare with the report presented in Lusaka in 1992. We have heard of enhanced decentralised rabies diagnostic capacity in Zambia which, if it can be sustained, will undoubtedly improve the potential for better rabies control in the country. We have heard of the continued high quality of rabies surveillance in Zimbabwe, a model within the region. We have heard of continued effective control of rabies in Swaziland, and the absence of rabies in Lesotho, situations likely to come under increasing threat in the coming months and years. And we have heard, particularly from our colleagues in Tanzania and Lesotho, the use of the terms semi-restricted and unrestricted dogs rather than "strays", indication of greater consideration of dog ecology in rabies control programmes.

And now the bad news! It has been saddening to observe an increasing rabies incidence reported from most of the countries in the region, as well as a decline in rabies vaccine coverages. We have also seen a continued tendency to read inferences on the patterns of the disease from some countries on the basis of very limited data from a potentially biased set of submitted specimens. From many countries, we hear that rabies control strategies haven't changed at all, despite our discussion at previous meetings of the need for dog ecology studies, the need to target high risk communities, and the need to consider carefully when it is appropriate to allocate the sparse resources to dog population control in the absence of effective vaccination programmes. Lastly, it was disturbing to note that some of the presenters appeared to miss some of the more exciting things on rabies control going on in their countries. Was this an indication that some government veterinary services may be becoming increasingly isolated from events, given their severe lack of resources and the ever increasing pressure to privatise many aspects of veterinary services? While the privatisation of veterinary practice makes sound economic sense in countries where routine veterinary procedures have been carried out by the public sector, we must ensure that diseases of public health concern, such as rabies, particularly in circumstances where they affect impoverished communities, are not overlooked in this process, and remain clearly on the agenda of the public sector veterinary and medical authorities.

Dr Bishop: It is an appropriate time to ask ourselves "where are we going from here?" AIDS pulls away money and public awareness from rabies. Rabies in Africa is a people's problem; socio/political organisations cannot control rabies until we can control the movement of people. Also, within the region, we still have different levels of diagnostic capability.

DISCUSSION OF HUMAN RABIES SESSION

Dr Swanepoel's paper (no paper submitted for this proceedings) Which vaccine is the best? Human diploid cell vaccine.
Even for human cases due to European bat lyssaviruses? **Dr Meslin:** There have been very few cases of genotype 5 and 6 infections, none of which received HDCV.

Do human to human cases ever occur? They are possible, but not recorded, except through corneal transplants.

Dr van Geldermalsen's paper
You have no data on the use of human rabies immunoglobulin. Is it used? No, there are only a few ampoules in the fridge.

How many doses of vaccine are bought and used? Within the last two months, masses. Each person is given five injections.

Dr Fekadu's paper
Children on life-support systems appear to have unusually long clinical periods. Yes, they do, but even without these systems the clinical period can be very long.

Dr van Geldermalsen: You do not need a test which is not practical. Yes, but we have had six cases in the USA within the last two year, all diagnosed post-mortem - rabies was not always considered.

Dr Lang's paper
It is essential that the number of visits to the medical centre are cut down; five doses intramuscularly is totally impractical in the developing world. In Kenya there are a number of pirates selling useless reagents at inflated cost. Is there any interest in research into modifying schedules? **Dr Meslin:** At present, 1/50,000 treatments fail; it is difficult to change schedules of proven systems in current usage without putting people at risk. WHO recommends the 6th dose to be given at 90 days. If the 2.1.1. schedule of Zagreb is used, globulin should not also be used.

GENERAL DISCUSSION OF HUMAN RABIES

What if 40 people eat a rabid animal? **Dr Meslin:** One must use the intradermal route for post-exposure because of the cost. The ID route is a well recognised route and the schedule developed is in two sites on day 0, two on day 3, two on day 7, one on day 30 and one on day 90.

There appears to be a dichotomy in veterinary and medical vaccine manufacture and use; for example, adjuvants are not used in human treatment. **Dr Lang:** Adjuvants are used when the antigenicity is low, and if an adjuvant is used there may be a slower response.

What do we do with dog bite patients while waiting for the result from the laboratory? **Dr Bingham**: We recommend that the hospital assesses the situation according to the nature of contact and begins the appropriate treatment. The treatment is stopped if the brain proves negative or the animal remains healthy after ten days. **Dr Perry:** One can use a "decision tree" system, according to the nature of exposure, species etc. A framework for decision support is required. **Dr Sembiko:** Nobody can determine whether a patient should or should not be given treatment - it is too risky. **Dr Meslin:** But you must make this judgement because vaccine is so expensive and in such short supply. WHO has guidelines. Also co-operation between veterinary and medical professions must be improved - a good exercise is to bring the two groups together to design their own method of co-operation.

There is a high incidence of acute death, how many cases are missed? **Dr Fekadu:** In experiments with dog rabies, 18 percent died without showing any signs at all; one dog secreted virus for 14 days without showing signs and another survived for 125 days and on occasion secreted six logs of virus in the saliva.

Dr Kaboyo: In Uganda, two children entered hospital with advanced rabies and if the mother had not been given post-exposure treatment she too might have died. **Dr Fekadu:** I know of two cases where both mother and child died, but it was not known if one or both were bitten or who was bitten first.

DISCUSSION OF CANINE RABIES SESSION

Dr Kitala's paper

Was there a differential interaction between households' vaccine delivery and rabies exposure? **Dr McDermott:** In Kenya, dogs are a boys'thing, women have nothing to do with them and the men are working in towns, so one cannot always get a response by knocking on doors.

Is it possible to vaccinate under three months of age? What are vaccine manufacturers doing in order to give better value? **Dr Jeffries** (Rhone Merieux): It is reasonable to assume that the quality of immunity transferred (from the mother) is minimal and therefore vaccines can be used at any age after four weeks. Such vaccination may lead to a lower response and may require a second dose four weeks later. This may not be practical.

Are you doing studies to take this further? **Dr Jeffries:** No, they are not justified.

Dr Wandeler: We recommend puppy vaccination.

Dr Sembiko: Yes, but with such a high mortality of young puppies in the first three months, so much vaccine will be wasted.

Dr McDermott: We shall just have to try it but vaccination can only be done when the boys are not at school.

Dr Coleman's paper

How homogeneous is vaccine coverage? **Dr Wandeler:** In Mexico there is variation of 40-70 percent within given areas.

Dr Rutebarika's paper

What are the number of vaccine doses used and the number of dogs vaccinated? Sometimes no vaccine is used because of difficulties in reaching the animals. We are trying to change the System.

Where do the stray dogs live? Some are urban, some are rural.

Dr de Balogh's paper

Is it possible to combine vaccination with other operations and social problems? Yes it is, but this may dilute the attention to rabies. **Dr Munang'andu** (Zambia): We just go for rabies in towns, but may widen our activities in rural areas.

Has the campaign been repeated? No, the problem is who will be responsible for it?

Did you give the children post-exposure treatment? We had treatment available but it was not needed - the children brought their own dogs' along.

Can you make the programme customer driven? There was a willingness to bring dogs but no resources for vaccine etc. Radio was the best means of publicity, not many people had television sets or newspapers.

Dr Madzima's paper

Where do most of the rabies cases occur? Most dog rabies occurs in the communal farming areas, however, this is usually very poorly reported.

What proportion of dogs treated end up rabid and are there vaccine breakthroughs? We occasionally have vaccinated dogs with rabies, but the incidence is less than 1 percent of diagnosed dogs. **Dr Swanepoel:** Inactivated vaccines are now the most widely used – there used to be breakthroughs but not now. **Dr Ushewokunze-Obatolu:** There are also by-laws which assist in rabies control, for example neutering and the spaying of bitches.

Dr King's paper

Fox rabies is somewhat cyclical, how do we know that it is not just disappearing without the intervention by oral vaccination? **Dr Wandeler:** This has not been studied extensively in Europe, but it is currently being studied in Canada.

Dr Perry: How do you get rid of the last one or two cases? This is the difficulty, anyone could predict that it would be easy to eliminate the first 90 percent of cases and the rest would be a problem. One has to maintain active surveillance and re-vaccinate infected areas until no indigenous cases are reported for two years.

When will UK get rid of quarantine? I am not in a position to make such a prediction, but the UK would like to see a little more progress in the eradication programmes of our neighbours and once satisfied that animal identification, certification, vaccination and antibody testing are fool- and evasion-proof, then no doubt the relaxation of quarantine of animals which travel amongst the countries of the European Community, but not of animals which come from areas in the world where rabies other than fox-borne rabies is endemic, will be considered.

Dr Meslin's paper

Data on the economics of dog oral vaccination are being gathered, but regarding the differences of bait uptake, are they due to differences in the taste of dogs? Perhaps.

Dr Schumacher's paper

You mention delivery of vaccine to homes and then walk away, or delivery to community leaders. Are there professional and ethical questions when you give vaccines to the public? We went back to the public and took blood samples. Yes, but what did the local veterinarians say? **Dr Meslin:** In Tunisia this question was not addressed, but it might not be so much of a problem, it would not conflict with the private sector.

GENERAL DISCUSSION OF RABIES IN DOGS

Traditional healers are used for many purposes, what do you advise governments to do? **Dr Wandeler:** Its a very good question - one can only go to public education, schools etc. We had tried to ask dog bite specialists to tell patients to go to veterinarians and physicians. Can we take a legal approach? Possibly, but we should not put people out of business.

Rabies is a bigger problem where the dog population is low and this conflicts with the problem in Natal - why? **Dr McDermott:** At a density of 6-20/km^2 the population is much lower than that in Natal, the contact rate is less and there may be more restrictions in upper class areas. **Dr Bishop:** There is no real conflict, we had taken extreme cases of highest and lowest dog density and the positives made sense. The problem may be of census - unevenness of populations throughout the area but the figures are taken to represent the whole area.

Dr Swanepoel: In mongoose rabies areas there has been the odd dog case but the disease did not spread amongst dogs. **Dr Thomson:** In addition, around Cape Town there has been for many years rabies in bat-eared foxes yet there is no rabies in dogs.

Dr Bishop: It is a game without rules - rabies is found in poor peoples' areas; rich people live in better areas and have no rabies.

Dr Butler: Should we wait for dog oral vaccines to come on the.market? What evidence is there of non-target species taking the bait? **Dr Schumacher:** In our study, there were some donkeys, no cats, but there were very few in the area.

Dr McDermott: Are there endemic areas where there are always infected dogs? **Dr Wandeler:** We should be cautious of epidemic patterns, they may depend on the degree of surveillance.

Dr Fekadu: The disease of rabies falls between two chairs, the Ministries of Agriculture and of Health, how do we deal with this problem? **Dr Ushewokunze-Obatolu:** You cannot have separate budgets - if, say, the Ministry of Health asked for a budget for rabies control they might get it all.

Dr Thomson: Is rabies an important disease, compared to others? **Dr Meslin:** Yes, there may be other, bigger, problems but if you look at costs other than the existing mortality details, it costs a lot. **Dr Fekadu:** Next to AIDS, rabies is the most feared disease. **Dr de Balogh:** Yes, but rabies is preventable.

What is the use of vaccinating puppies, most of which will die soon afterwards? It is an issue of availability and costs. **Dr Wandeler:** That is a very important question - dogs less than one year of age are a very important age group. **Dr Fekadu:** Non-vaccination of dogs less than three months old was introduced when live virus vaccines wed in use. Modem inactivated vaccines are safe for puppies. **Dr Perry:** If that small window of less than three or even less than one month of age is the most important it may be the right time to vaccinate. **Dr Dlamini:** A significant number of rabid dogs are less than three months of age. People are not afraid of handling puppies whereas older dogs can be very difficult to handle.

DISCUSSION OF EPIDENHOLOGY AND DIAGNOSIS SESSION

Dr Barrat's paper

What is the specificity problem with ELISAs? We don't know why the ELISA is specific for foxes and not for dogs. Why do you say that the neutralisation test has no problem with specificity? All the positives were clearly positive and all the negatives were clearly negative, that is, 100 percent specificity. Why is such an odd test as the RFFIT always used, why not a straight, classical, neutralisation test? I don't know. **Dr Fekadu:** If you compare the RFFIT with the neutralisation test in mice. you get the same result. **Dr**

Wandeler: That is more or less correct but you get different titres. **Dr Fekadu:** With incubation of normal dog sera, a number of laboratory beagles had titres of 1/25, so we always had a cut-off.

GENERAL DISCUSSION ON EPIDEA1I0LOGY AND DIAGNOSIS

Dr Bishop: Concerning post-mortems in humans using the orbital puncture core sample technique for FAT: in two positive cases where the sample was put into mice, the glycerol saline preservative appeared to lower the titre after the sample had been left on the bench at room temperature.

Dr Perry: I have serious concerns about the conclusions being drawn from work on antigenic variation, the sample sizes are low but the conclusions huge. **Dr Thomson:** I could not agree more - the southern African analyses are being carried out on many more samples and will probably be more accurate.

Will current vaccines be effective against mongoose rabies? **Dr Thomson:** While there is evidence of antigenic variation, there is no evidence that vaccine will not protect. **Dr Swanepoel:** There was an historic case of a vaccinated person who died from mongoose rabies, but the vaccine had been given into the gluteal region.

DISCUSSION ON SURVEILLANCE

Dr Dye: It is important that more thorough analysis be done of routine data. Rabies can be controlled if all the information and tools are in place.

What is the solution to control of jackal rabies and why is it not being implemented? **Dr Bingham:** In principle, oral vaccination is effective in jackals, but more time, money and resources are needed to complete the basic research and to implement an oral vaccination system. Culling of jackals is probably not an effective solution in the long-term, due to high population turn-over.

APPENDIX I. LIST OF PARTICIPANTS.

Dr Patrick Barillot, Virbac, BP 27 - 06511, Carros Cedex, France.

Dr Jacques Barrat Laboratoire d'Etude sur la Rage et la Pathologie des Animaux Sauvages, CNEVA, BP 9, 54220 Malzeville, France.

Dr Berton, Virbac, BP 27 - 06511, Carros Cedex, France.

Dr John Bingham, Veterinary Research Laboratory, PO Box CY 551, Causeway, Harare, Zimbabwe.

Dr George Bishop, Allerton R.V.L. Private Bag X2, Cascades 3202, South Africa.

Dr Bobade, Faculty of Veterinary Science, University of Zimbabwe, PO Box W 167, Mount Pleasant, Harare, Zimbabwe.

Dr Doug Bruce, Cooper Zimbabwe, PO Box 2699, Harare, Zimbabwe.

Dr Gideon Brückner, Department of Animal Health, Private Bag X138, Pretoria 0001, South Africa.

Mr James Butler, Department of Biological Sciences, University of Zimbabwe, PO Box MP 167, Mount Pleasant, Harare, Zimbabwe.

Mrs Chasokela, Department of Nursing Services, Ministry of Health and Child Welfare, PO Box CY 1122, Causeway, Harare, Zimbabwe.

Dr Sarah Cleaveland (Gascoyne), Institute of Zoology, Zoological Society of London, Regent's Park, London NW1 4RY, United Kingdom.

Dr Paul Coleman, Medical Parasitology, London School of Hygiene and Tropical Medicine, Keppel Street, London WC1E 7HT, United Kingdom.

Dr Katinka de Balogh, University of Mozambique, c/o Royal Netherlands Embassy, PO Box 1163, Maputo, Mozambique.

Dr M.P.R. Travassos Dias. Instituto Nacional de investigacao Veterinaria, CP 1922, Maputo, Mozambique.

Dr Roland X. Dlamini, Dept. Veterinary Services, PO Box 11, Nlangano, Swaziland.

Dr Chris Dye, Medical Parasitology, London School of Hygiene and Tropical Medicine, Keppel Street, London WC1E 7HT, United Kingdom.

Mr Philip Dziva, Department of Epidemiology and Disease Control, Ministry of Health and Child Welfare, PO Box CY 1122, Causeway, Harare, Zimbabwe.

Dr Makonnen Fekadu, Department of Health and Human Services, Centers for Disease Control and Prevention, Atlanta, GA 30333, USA.

Dr Francois Flanagan, RDP Livestock Services, 10 Angel King Avenue, Chinhoyi, Zimbabwe.

Dr Peter Gamble, Veterinary Research Laboratory, PO Box CY 551, Causeway, Harare.

Dr Thierry GozIan, Virbac Laboratories, BP 27 - 06511, Carros Cedex, France.

Dr Stuart K. Hargreaves, Department of Veterinary Services, PO Box CY 66, Causeway, Harare, Zimbabwe.

Dr O.J.B. Hübschle, Central Veterinary Laboratory, Private Bag 13187, Windhoek, Namibia.

Dr James Illango, Animal Health Research Centre, PO Box 24, Entebbe, Uganda.

Dr Peter Jeffries, Rhone Merieux, 29 Avenue Tony Gamier, BP 7123, 69348 Lyon Cedex 07, France.

Dr Winyi Kaboyo, Ministry of Health, PO Box 8, Entebbe, Uganda.

Mr Kadzatsa, Department of Environmental Health, Ministry of Health and Child Welfare, PO Box CY 1122 Causeway, Harare, Zimbabwe.

Dr Githaiga Kamau, Kenya Wildlife Services, Kenya.

Dr A.K. Karugah, Department of Veterinary Services, Kabete, Kenya

Dr L. Khomari, Department of Livestock Services, Private Bag A82, Maseru, Lesotho.

Dr Arthur King, Formerly, Rabies Unit Central Veterinary Laboratory, New Haw, Addlestone, Surrey KT15 3NB, United Kingdom.

Dr Paul Kloeck, Department of Animal Health, Private Bag X2, Cascades 3202, South Africa.

W Timothy Kudinha, Veterinary Research Laboratory, PO Box CY 55 1, Causeway, Harare, Zimbabwe.

Mr Ignatius Kunda, Central Veterinary Research Institute, Central Veterinary Research Institute, PO Box 33980, Balmoral, Lusaka, Zambia.

Dr Jean Lang, Pasteur Merieux Serums et Vaccins, 58 Avenue Leclerc, BP 7046, 69348 Lyon Cedex 07, France.

Dr Rachel Madekurozya, Veterinary Research Laboratory, PO Box CY 551, Causeway, Harare, Zimbabwe.

Dr Welbourne Madzima, Department of Veterinary Services, PO Box CY 66, Causeway, Harare, Zimbabwe.

Dr Majok, Faculty of Veterinary Science, University of Zimbabwe, PO Box MP 167, Mount Pleasant, Harare, Zimbabwe.

Dr Morgan Matingo, Veterinary Research Laboratory, PO Box CY 551, Causeway, Harare.

Dr Jonathan Matongo, Dept Veterinary Services, PO Box 5, Kadoma.

Dr John McDermott Faculty of Veterinary Medicine, University of Nairobi, PO Box 29053, Kabete, Kenya.

Dr Francois Meslin, Veterinary Public Health, World Health Organization, CH- 1211, Geneva 27, Switzerland.

Mrs Lucy Mnkandla, Department of Epidemiology and Disease Control, Ministry of Health, PO Box CY 1122, Causeway, Harare, Zimbabwe.

W Mtshena, Ministry of Health and Child Welfare, PO Box 206, Gweru, Zimbabwe.

Mr Mufambanhando, Ministry of Health and Child Welfare, PO Box 323, Mutare, Zimbabwe.

Dr Hetron Munang'andu, Central Veterinary Research Institute, PO Box 33980, Balmoral, Lusaka, Zambia.

Mr Muza, Department of City Health, Ministry of Health, PO Box CY 1122, Causeway, Harare, Zimbabwe.

Dr Mwenye, Ministry of Health and Child Welfare, PO Box 10, Marondera, Zimbabwe.

Dr C. Mwiyeriwa, Liwonde Agricultural Development, Private Bag 3, Liwonde, Malawi.

Dr Chris Ncube, Veterinary Research Laboratory, PO Box CY 551, Causeway, Harare, Zimbabwe.

Mr James Nqindi, Veterinary Research Laboratory, PO Box CY 551, Causeway, Harare, Zimbabwe.

Dr Josephat Nyika, Department of Veterinary Services, PO Box 90, Bindura, Zimbabwe.

Prof. Mark Obwolo, Faculty of Veterinary Science, University of Zimbabwe, PO Box MP 167, Mount Pleasant Harare, Zimbabwe.

Dr Brian Perry, International Livestock Research Institute, PO Box 30709, Nairobi, Kenya.

Dr Brigitte Revol, 13 Roland Close, Kambanji, PO Chisipite, Harare, Zimbabwe.

Dr Fernando Rodrigues, National Directorate of Livestock, PO Box 1406, Maputo, Mozambique.

Dr D.W. Rumisha, Ministry of Health, Private Bag 00269, Gaberone, Botswana.

Dr C.S. Rutebarika, Department of Veterinary Services, PO Box 513, Entebbe, Uganda.

Dr Carolin Schumacher, Virbac, Biological Research and Development, BP 27 - 06511, Carros Cedex, France.

Dr Sehularo, Dept. Animal Health and Production, Private Bag 0032, Gaberone, Botswana.

Dr Y.S.S. Sembiko, Veterinary Public Health, Ministry of Agriculture, PO Box 9152, Dar es Salaam, Tanzania

Dr Martin Sibanda, Veterinary Research Laboratory, PO Box CY 551, Causeway, Harare, Zimbabwe.

Prof R. Swanepoel, National Institute of Virology, Private Bag X4, Sandringham 2131, South Africa.

Dr Gavin R. Thomson, Onderstepoort Institute for Exotic Diseases Private Bag X6, Onderstepoort 0110, South Africa.

Dr Rejo Tsiresy, Ministère du Développement Rural et de la Réforme Foncière, Antananarivo, Madagascar

Dr Unesu Ushewokunze-0batolu, Veterinary Research Laboratory, PO Box CY 551, Causeway, Harare, Zimbabwe

Dr Aad van Geldermalsen, Ministry of Health, PO Box 98, Bindura, Zimbabwe

Dr Alexander I. Wandeler, Animal Diseases Research Institute, 3851 Fallowfield Road, Nepean, Ontario K2H 8P9, Canada.

Dr Arthur Wellington, Rhone Poulenc, PO Box 819, Halfway House 1685, South Africa

APPENDIX II. LIST OF AUTHORS

Dr M.F. Aubert
Laboratoire d'Etude sur la Rage et la Pathologie des Animaux Sauvage
CNEVA
BP 9, 54220 Malzeville
France.

Dr Jacques Barrat
Laboratoire d'Etude sur la Rage et la Pathologie des Animaux Sauvage
CNEVA
BP 9, 54220 Malzeville
France.
Tel: +33 8329 2608 Fax: +33 8329 3313 Email: vana30@calvacom.fr

Dr John Bingham
Veterinary Research Laboratory
PO Box CY 551, Causeway, Harare
Zimbabwe.
Tel: +263 4 705885 Fax: +263 4 707952Email: ncube@healthnet.zw

Dr George Bishop
Allerton RX.L.
Private Bag X2, Cascades 3202
South Aflica.
Tel: +27 331471931 Fax: +27 331471633

Dr Gideon Brückner
Department of Animal Health
Private Bag X138, Pretoria 0001
South Africa.
Tel: +27 12 319 7679 Fax: +27 12 329 0499

Mr James Butler
Department of Biological Sciences, University of Zimbabwe
PO Box MP 167, Mount Pleasant Harare
Zimbabwe.

Dr Sarah Cleaveland (formerly Gascoyne)
Institute of Zoology
Zoological Society of London
Regent's Park, London NW1 4RY
United Kingdom.
Tel: +44 71636 8636 Fax: +44 71436 5389

Dr Fl. Cliquet
Laboratoire d'Etude sur la Rage et la Pathologie des Animaux Sauvage
BP 9, 54220 MaIzeville
France

Dr Paul Coleman
Medical Parasitology, London School of Hygiene and Tropical Medicine
Keppel Street London WC1E 7HT
United Kingdom.
Tel: +44 71636 8636 Fax: +44 71436 5389

Dr Katinka de Balogh
University of Mozambique
C/o Royal Netherlands Embassy, PO Box 1163, Maputo
Mozambique.

Dr M.P.R Travassos Dias
Instituto Nacional de Investigacao Veterinaria
CP 1922, Maputo
Mozambique.
Tel: +258 1475170/2 Fax: +258 1475161 Telex: 6-195 DEAMD

Dr Roland X. Dlamini
Dept. Veterinary Services, PO Box 11
Nlangano
Swaziland.

Dr Chris Dye
Medical Parasitology, London School of Hygiene and Tropical Medicine
Keppel Street, London WC1E 7HT
United Kingdom.
Tel: +44 71636 8636 Fax: +44 71436 5389

Dr J. Esterhuysen.
Veterinary Research Institute
FMD Laboratory
Private Bag X5
0 110 Onderstepoort
South Africa

Dr Makonnen Fekadu
Department of Health and Human Services, Centers for Disease Control and Prevention
Atlanta, GA 30333
USA.
Tel: +1404 639 105

P.J. de Graaf
Faculty of Agronomy and Forestry Engineering
EMU, Maputo
Mozambique

Dr O.J.B. HÜbschle
Central Veterinary Laboratory
Private Bag 13187, Windhoek
Namibia.
Tel: +264 61237684 Fax: +264 61221099

Dr James Illango
Animal Health Research Centre
PO Box 24, Entebbe
Uganda.
Tel: +256 42 20193 Fax: +256 42 21047

Dr Winyi Kaboyo
Ministry of Health
PO Box 8, Entebbe
Uganda.
Tel: +256 42 20201 Fax: +256 42 20440 or 20996

Dr A.K Karugah
Department of Veterinary Services
Kabete
Kenya
Tel: +254 2 631387 Fax: +254 2 631499

Dr L. Khomari
Department of Livestock Services
Private Bag A82, Maseru
Lesotho.
Fax: +266 310 146

Dr Arthur King
Formerly, Rabies Unit Central Veterinary Laboratory
New Haw, Addlestone, Surrey KT15 3NB
United Kingdom.
Tel: +44 1932 341111 Fax: +44 1932 347046
Telex: 2623 18 VETWEY G

Dr Philip Kitala
Department of Public Health, Pharmacology and Toxicology
University of Nairobi, PO Box 29053
Nairobi, Kenya

Dr Jean Lang
Pasteur Merieux Serums et Vaccins
58 Avenue Leclerc, BP 7046, 69348 Lyon Cedex 07
France.
Tel: +33 7273 7037 Fax: +33 7273 7813

Dr Welbourne Madzima
Department of Veterinary Services
PO Box CY 66, Causeway, Harare
Zimbabwe.
Tel: +263 4 791355 Fax: +263 4 707952

Dr L.D. Mathunjwa
PO Box 8
Mbabane
Swaziland

Dr John McDermott
Faculty of Veterinary Medicine, University of Nairobi
PO Box 29053, Kabete
Kenya.
Tel: + 254 2 632211 or 632401

Dr Francois Meslin
Veterinary Public Health, World Health Organization
CH- 1211, Geneva 27
Switzerland.
Tel: +4122 7912111 Fax: +4122 7910746

Dr Hetron Munang'andu
Central Veterinary Research Institute
PO Box 33980, Balmoral, Lusaka
Zambia.
Tel/Fax: +260 1236283

Dr C. Mwiyeriwa
Liwonde Agricultural Development
Private Bag 3, Liworide
Malawi.
Tel: +265 740 323 Fax: +265 743 781

Dr Brian Perry
International Livestock Research Institute
PO Box 30709, Nairobi
Kenya.
Fax: +254 2 631499email: b.perry@cgnet.com

Dr Fernando Rodrigues
National Directorate of Livestock
PO Box 1406, Maputo
Mozambique.

Dr D.W Rumisha
Ministry of Health
Private Bag 00269, Gaberone
Botswana.
Tel: +267 374 351 Fax: +267 374 354

Dr C.S. Rutebarika
Department of Veterinary Services
PO Box 513, Entebbe
Uganda.
Tel: +256 42 20321 Fax: +256 42 21047 or 20428
Telex: 61267/61494

F. Sabbe
School of Veterinary Medicine
Lusaka
Zambia

Dr Carolin Schumacher
Virbac Biological Research and Development
BP 27 - 06511, Carros Cedex
France.
Tel: +33 9208 7163 Fax: +33 9208 7199

Dr Sehularo
Department of Animal Health and Production
Private Bag 0032, Gaberone
Botswana.
Tel: +267 350 500 Fax: +267 303 744 Telex: 2543 BD

Dr Y.S.S. Sembiko
Veterinary Public Health, Ministry of Agriculture
Pamba House, PO Box 9152, Dar es Salaam
Tanzania
Fax: +255 5134534 Telex: 41246 KILIMO TZ

Dr Gavin R Thomson
Onderstepoort Institute for Exotic Diseases
Private Bag X6, Onderstepoort 0110
South Africa.
Tel: +27 12 529 9511 Fax: +27 12 529 9543

Dr Rejo Tsiresy
Ministère du Développement Rural et de la Réforme Fonci6re
Antananarivo
Madagascar
Fax: +2612 40609

Dr M. Uanguta
Central Veterinary Laboratory
Private Bag 13187, Windhoek
Namibia

Dr Unesu Ushewokunze-Obatolu
Veterinary Research Laboratory
PO Box CY 55 1, Causeway, Harare
Zimbabwe
Tel: +263 4 705885 Fax: +263 4 707952Email: ncube@healthnetzw

Dr Aad van Geldermalsen
Ministry of Health
PO Box 98, Bindura
Zimbabwe
Tel: +263 1716764 Fax: +263 1716956

Dr Alex Wandeler
Animal Diseases Research Institute
3851 Fallowfield Road, Nepean, Ontario K2H SP9
Canada.
Fax: +1613 954 0614 email: wandelea@em.agr.ca