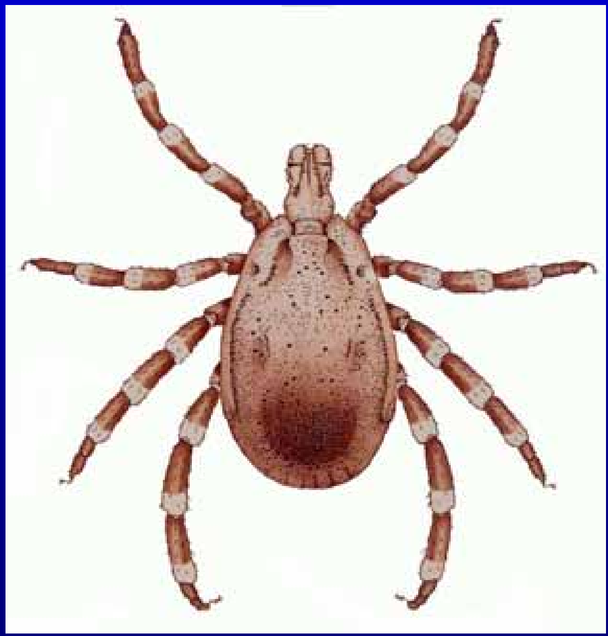
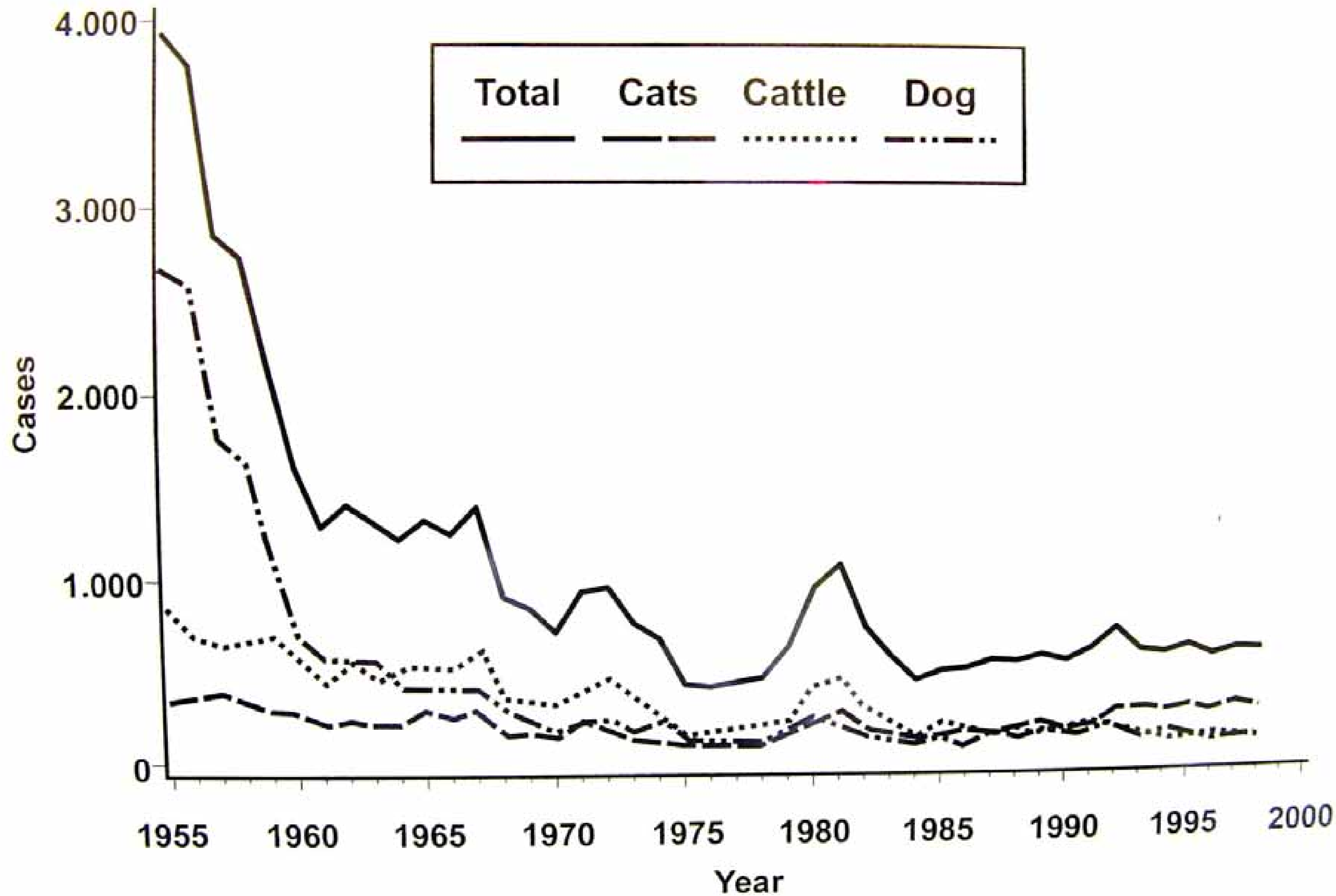
CONCEPTS OF RABIES IN AFRICA

R SWANEPOEL
SPECIAL PATHOGENS UNIT
NATIONAL INSTITUTE FOR COMMUNICABLE DISEASES
SOUTH AFRICA

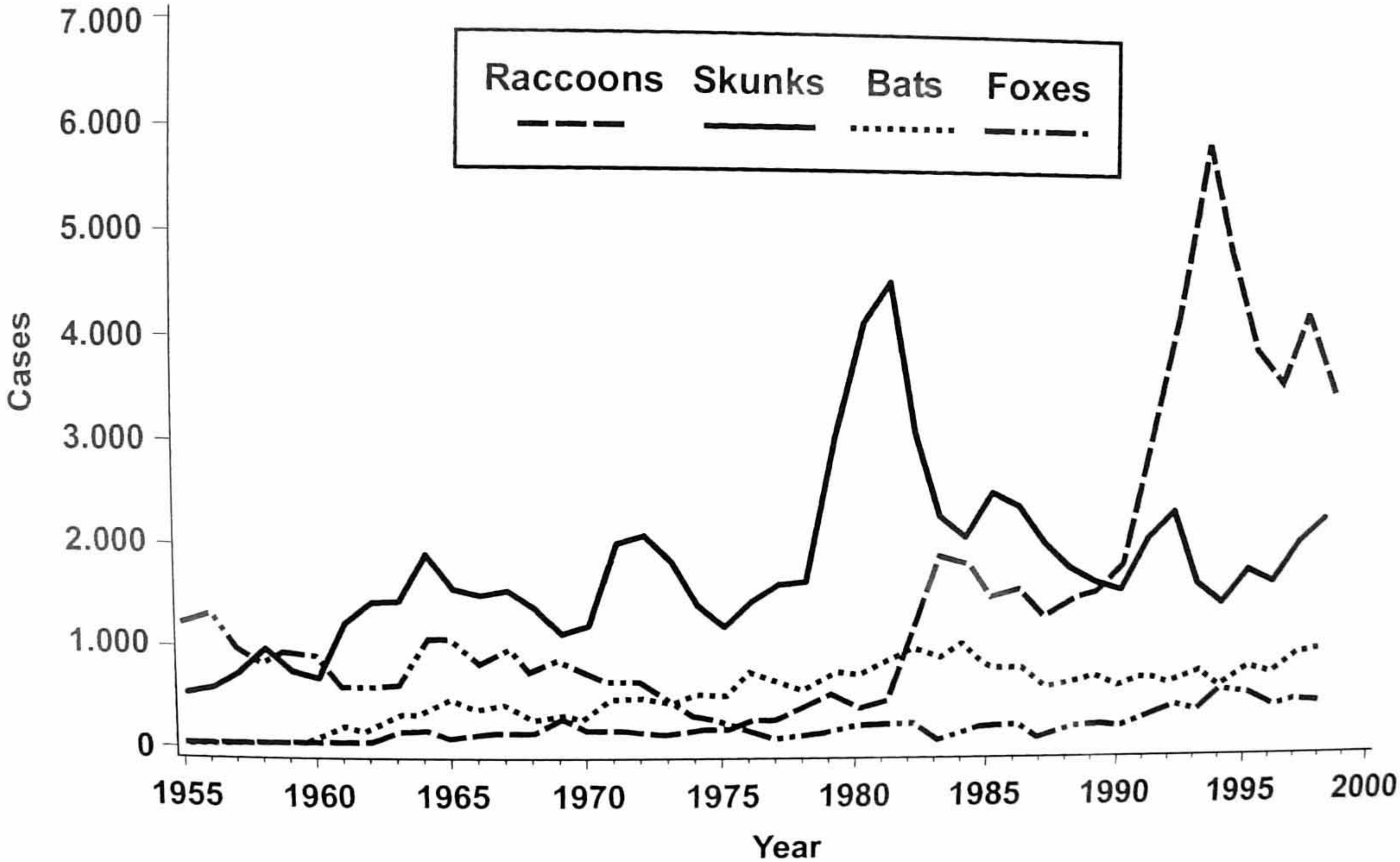


FAMILY RHABDOVIRIDAE
GENUS LYSSAVIRUS

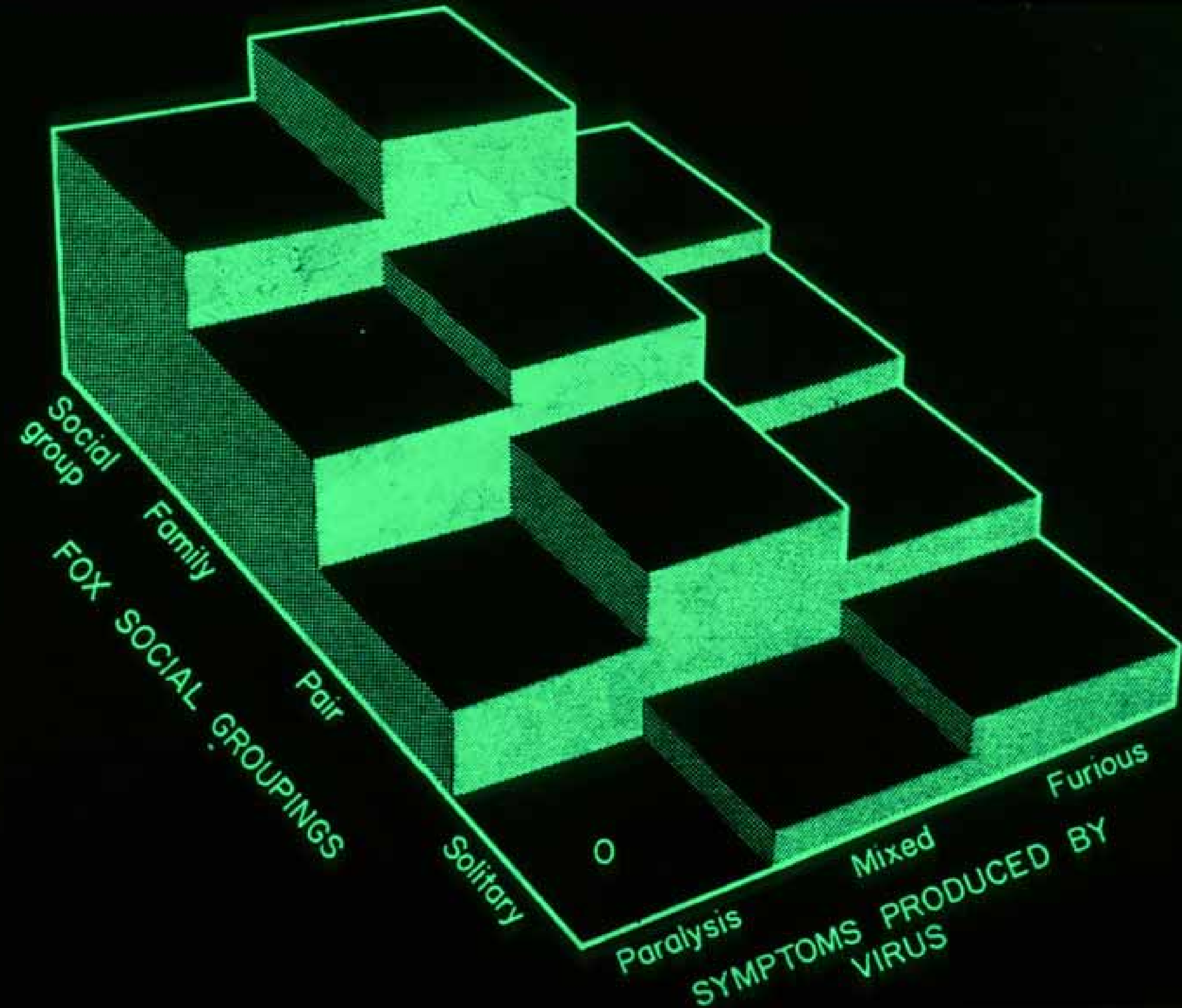
- RABIES = LYSSAVIRUS 1 (WORLDWIDE)
- LAGOS BAT = LYSSAVIRUS 2 (AFRICA)
- MOKOLA = LYSSAVIRUS 3 (AFRICA)
- DUVENHAGE = LYSSAVIRUS 4 (AFRICA)
- EUROPEAN BAT LYSSAVIRUS 1 = LYSSAVRUS 5
- EUROPEAN BAT LYSSAVIRUS 2 = LYSSAVIRUS 6
- AUSTRALIAN BAT LYSSAVIRUS = LYSSAVIRUS 7



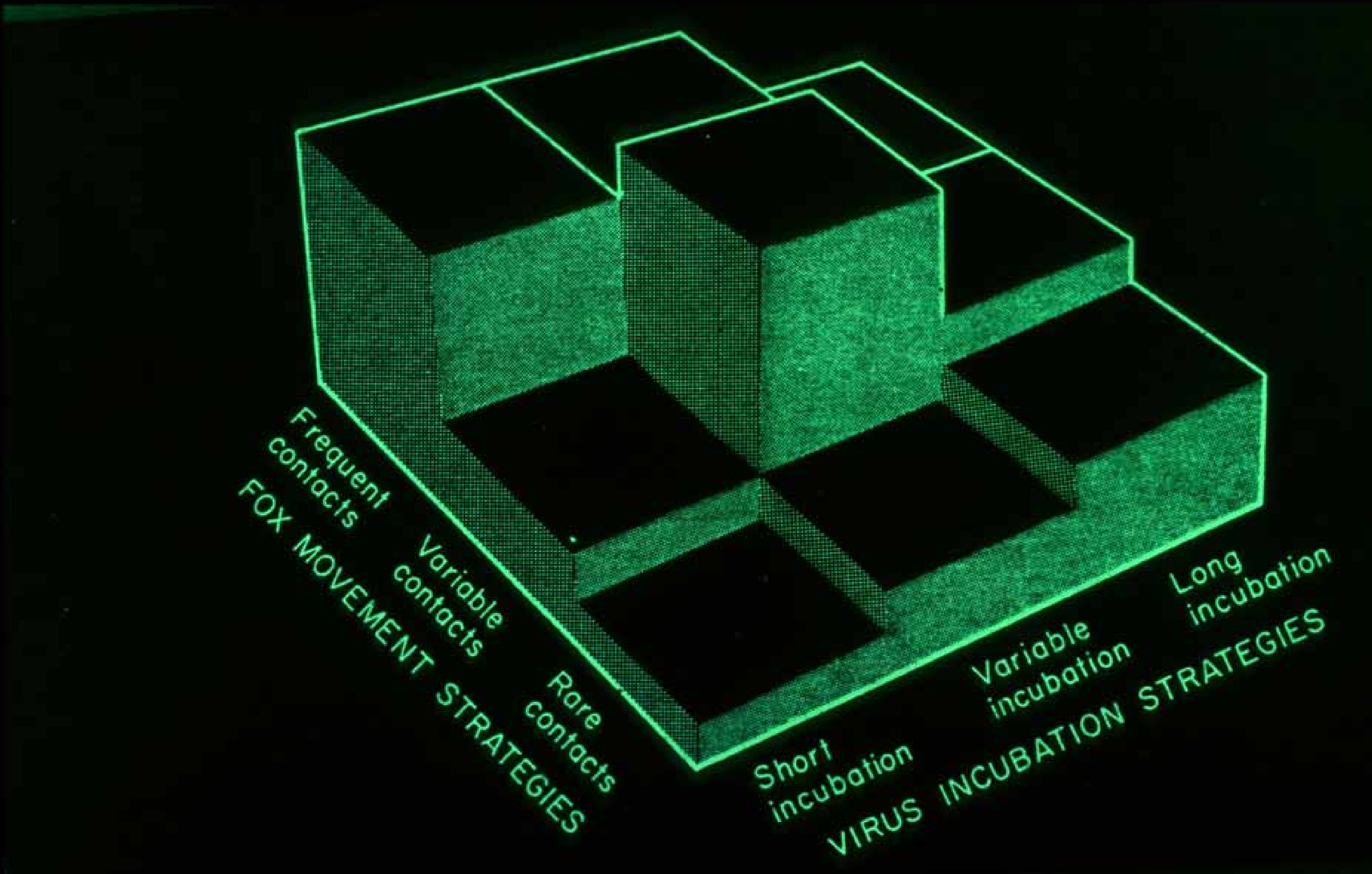
Total
Cats
Cattle
Dog
4.000
3.000
2.000
1.000
0
Cases
1955
1960
1965
1970
1975
1980
1985
1990
1995
2000
Year



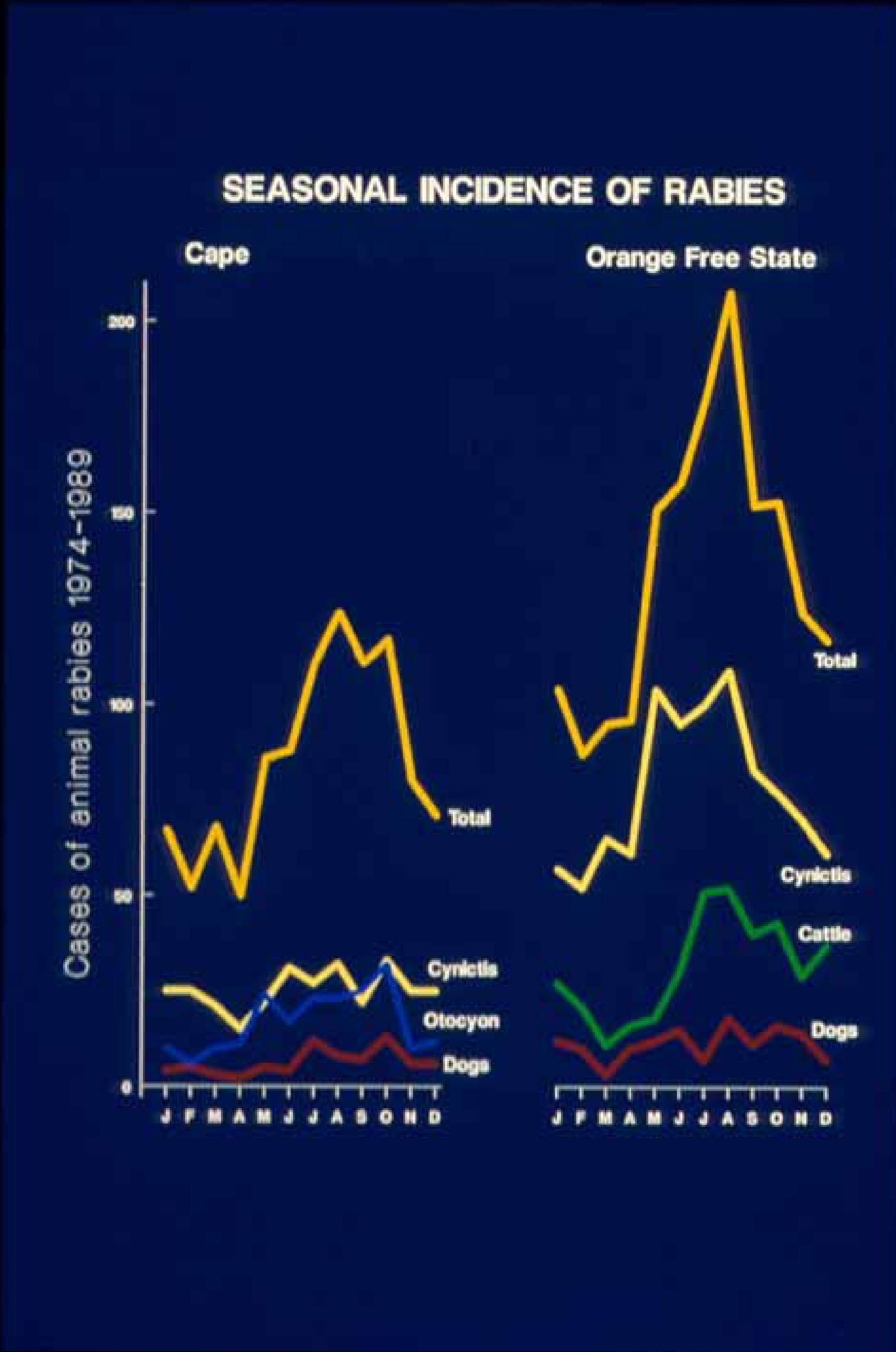
Raccoons
Skunks
Bats
Foxes
Cases
7.000
6.000
5.000
4.000
3.000
2.000
1.000
0
1955
1960
1965
1970
1975
1980
1985
1990
1995
2000
Year



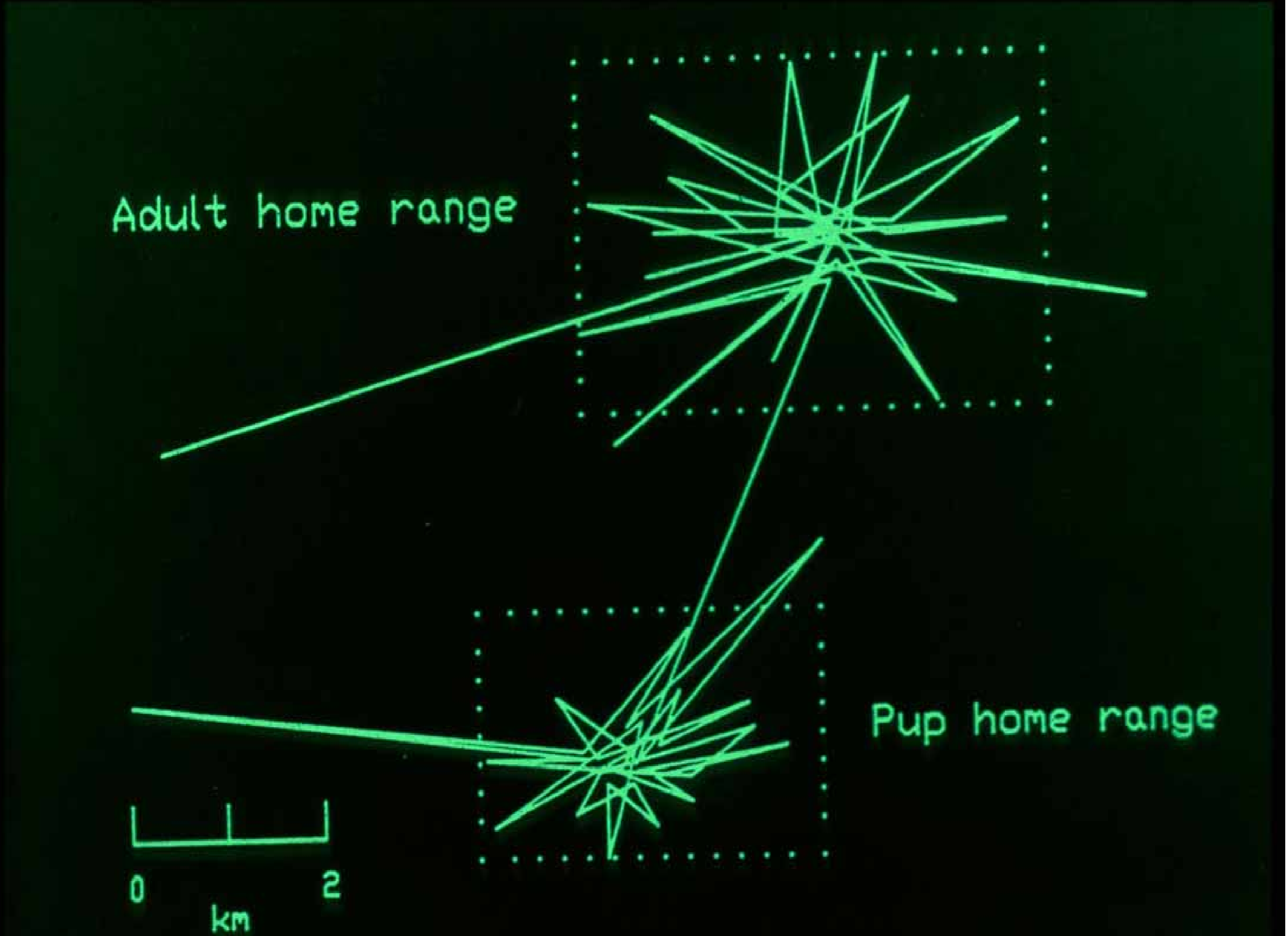
Social group
Family
Pair
Solitary
FOX SOCIAL GROUPINGS
0
Paralysis
Mixed
Furious
SYMPTOMS PRODUCED BY VIRUS


Frequent contacts
Variable contacts
Rare contacts
FOX MOVEMENT STRATEGIES
Short incubation
Variable incubation
Long incubation
VIRUS INCUBATION STRATEGIES

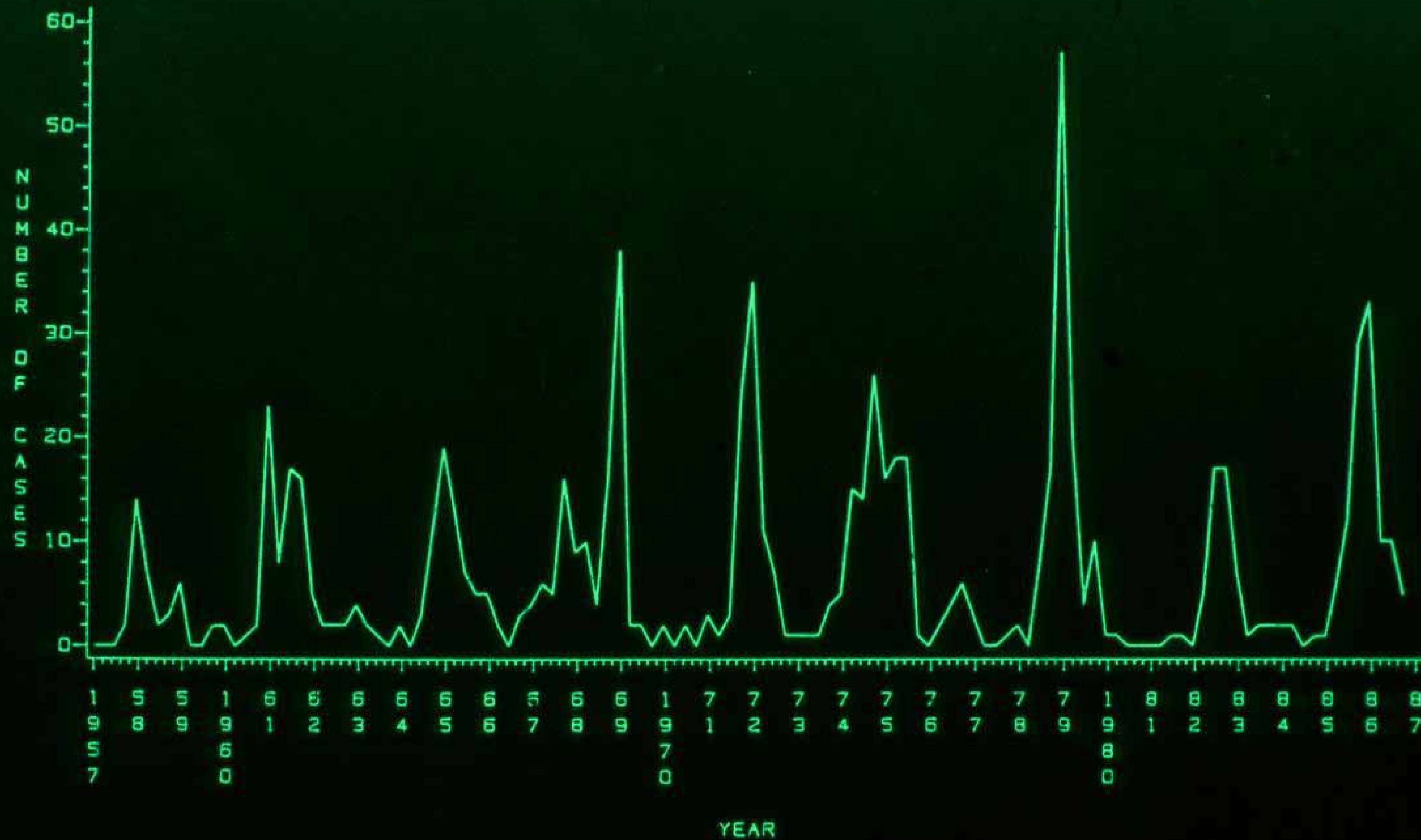

Skunk (north central states and California) gray fox (Texas)
Skunk (south central)
Raccoon
Red fox/Arctic fox
Gray fox (Arizona)



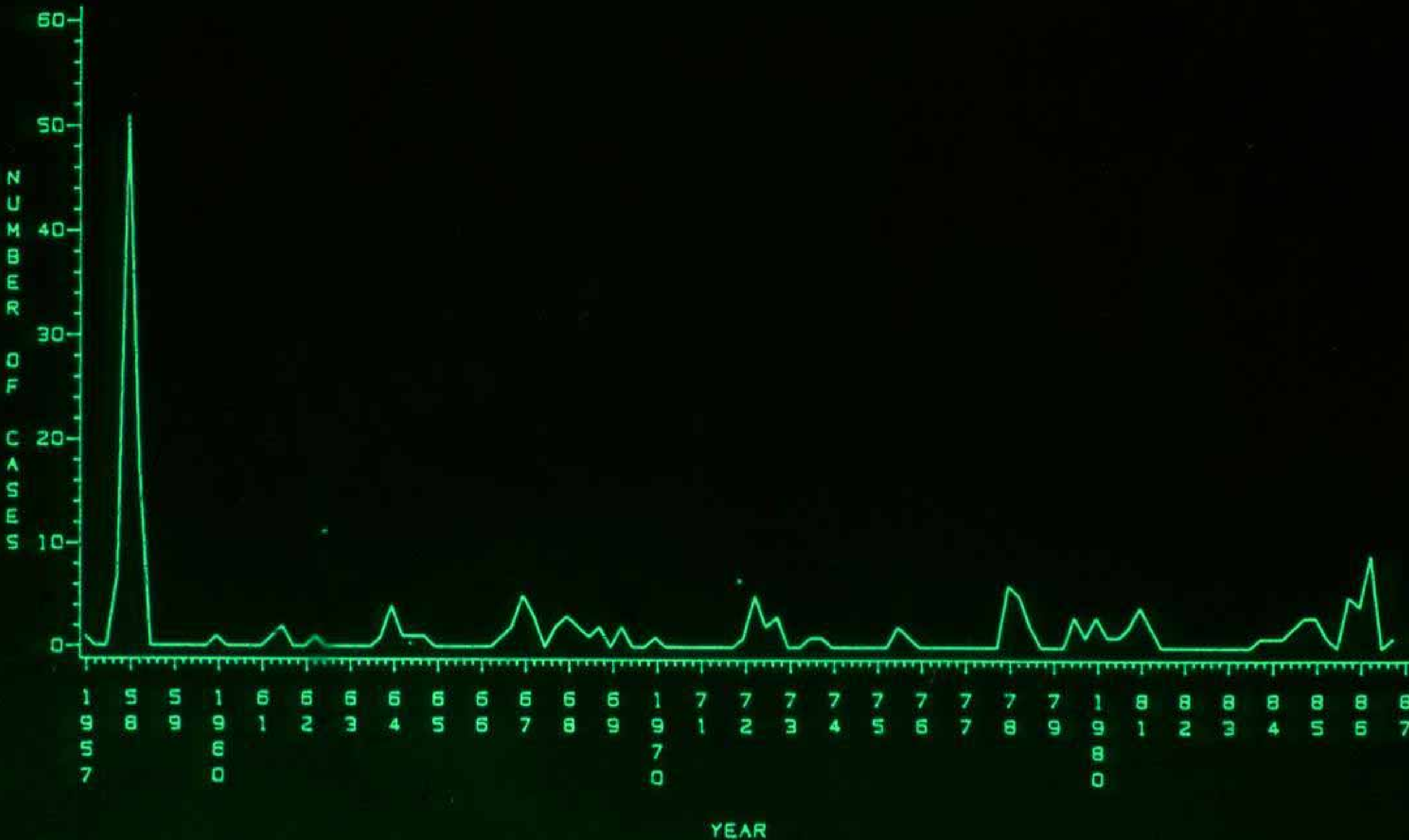
SEASONAL INCIDENCE OF RABIES
Cape
Orange Free State
Cases of animal rabies 1974–1989
200
150
100
50
0
Total
Cynictis
Otocyon
Dogs
Total
Cynictis
Cattle
Dogs
J F M A M J J A S O N D
J F M A M J J A S O N D


Adult home range
Pup home range
0
2
km

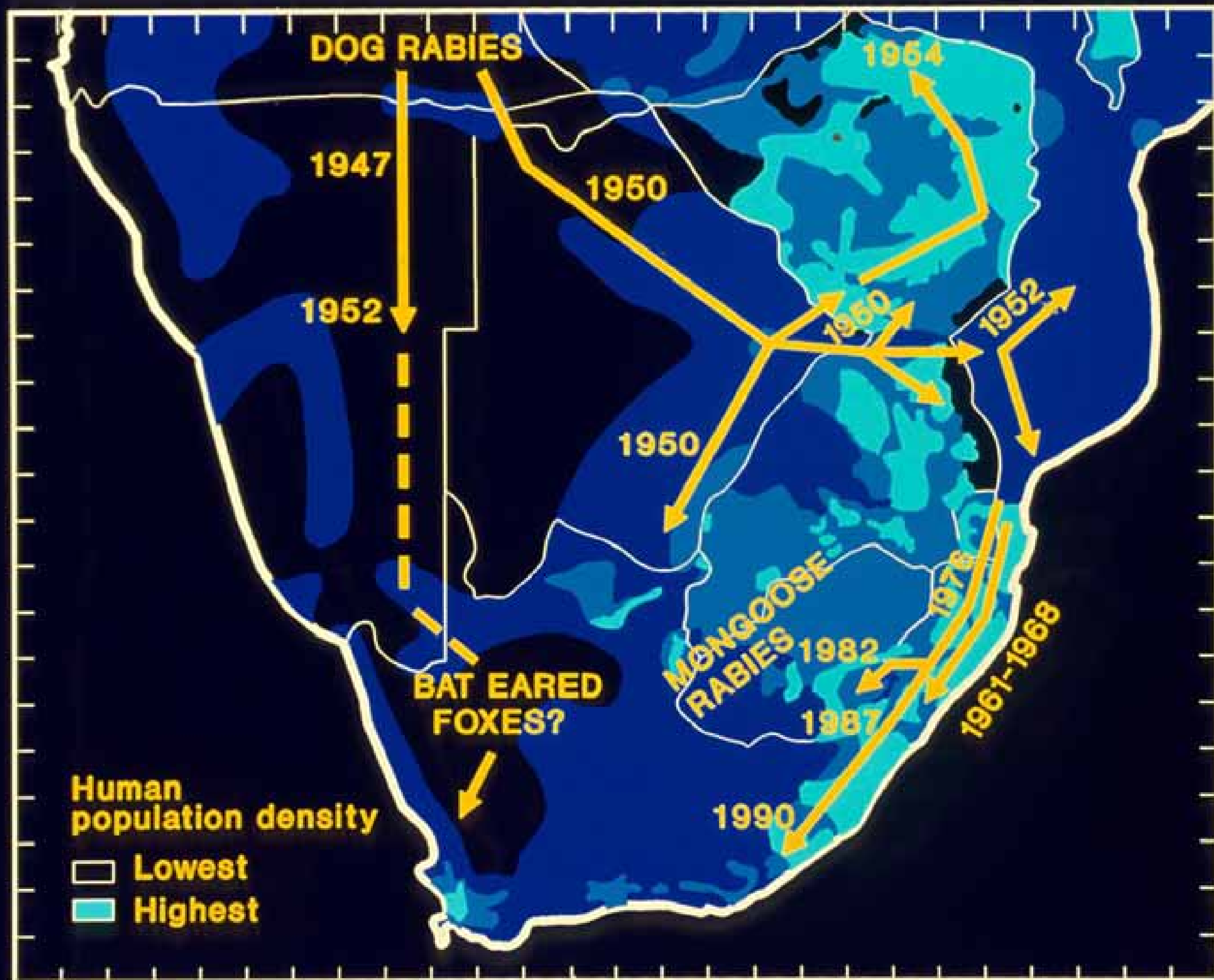

0
250
500 KM



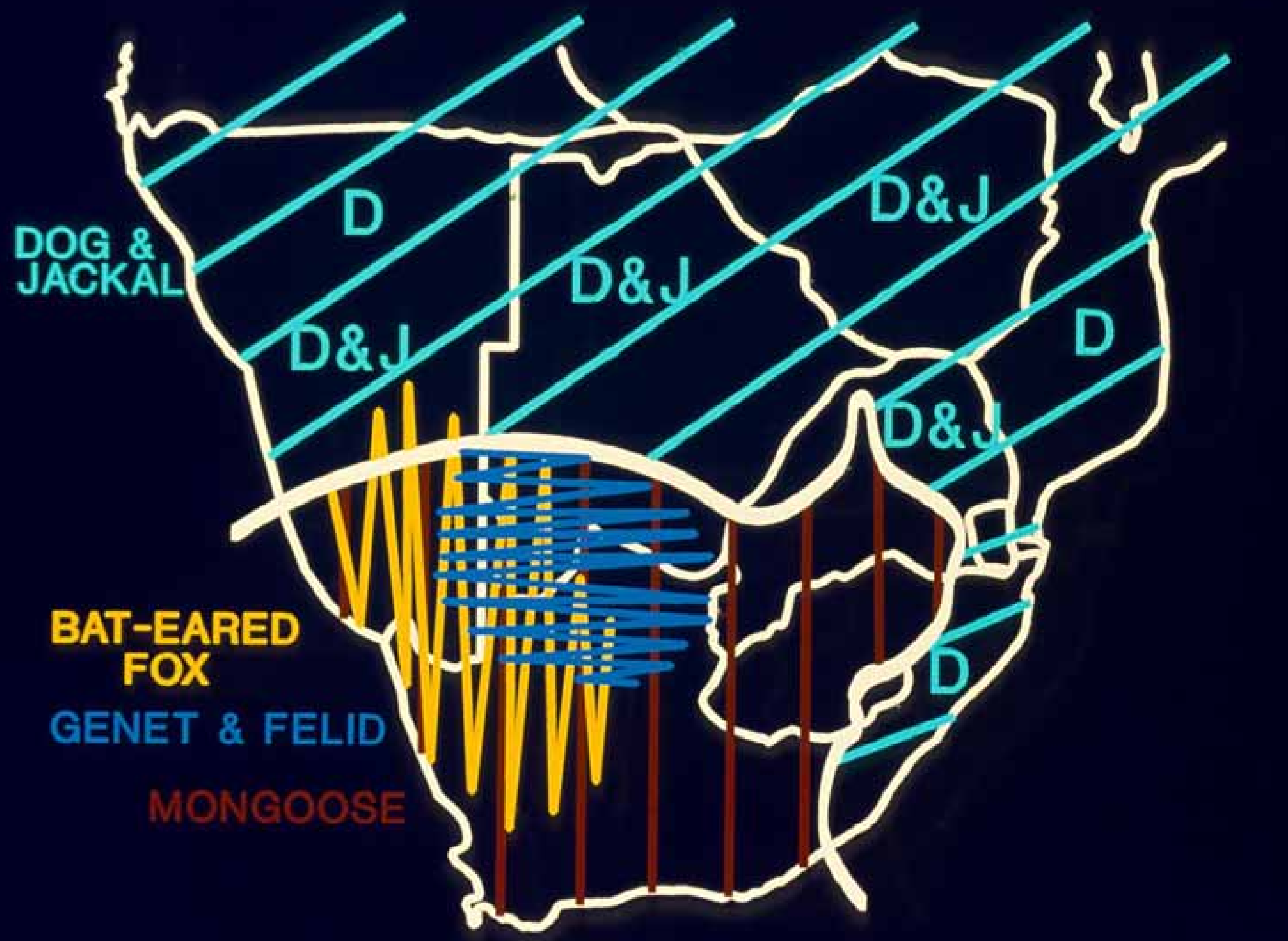
NUMBER OF CASES
0
10
20
30
40
50
60
1957
58
59
1960
61
62
63
64
65
66
67
68
69
1970
71
72
73
74
75
76
77
78
79
1980
81
82
83
84
85
86
87
YEAR



NUMBER OF CASES
60
50
40
30
20
10
0
1957
58
59
1960
61
62
63
64
65
66
67
68
69
1970
71
72
73
74
75
76
77
78
79
1980
81
82
83
84
85
86
87
YEAR


DOG RABIES
1947
1952
1950
1950
1954
1950
1952
1950
BAT EARED
FOXES?
MONGOOSE
RABIES
1976
1982
1987
1961-1968
1990
Human
population density
Lowest
Highest

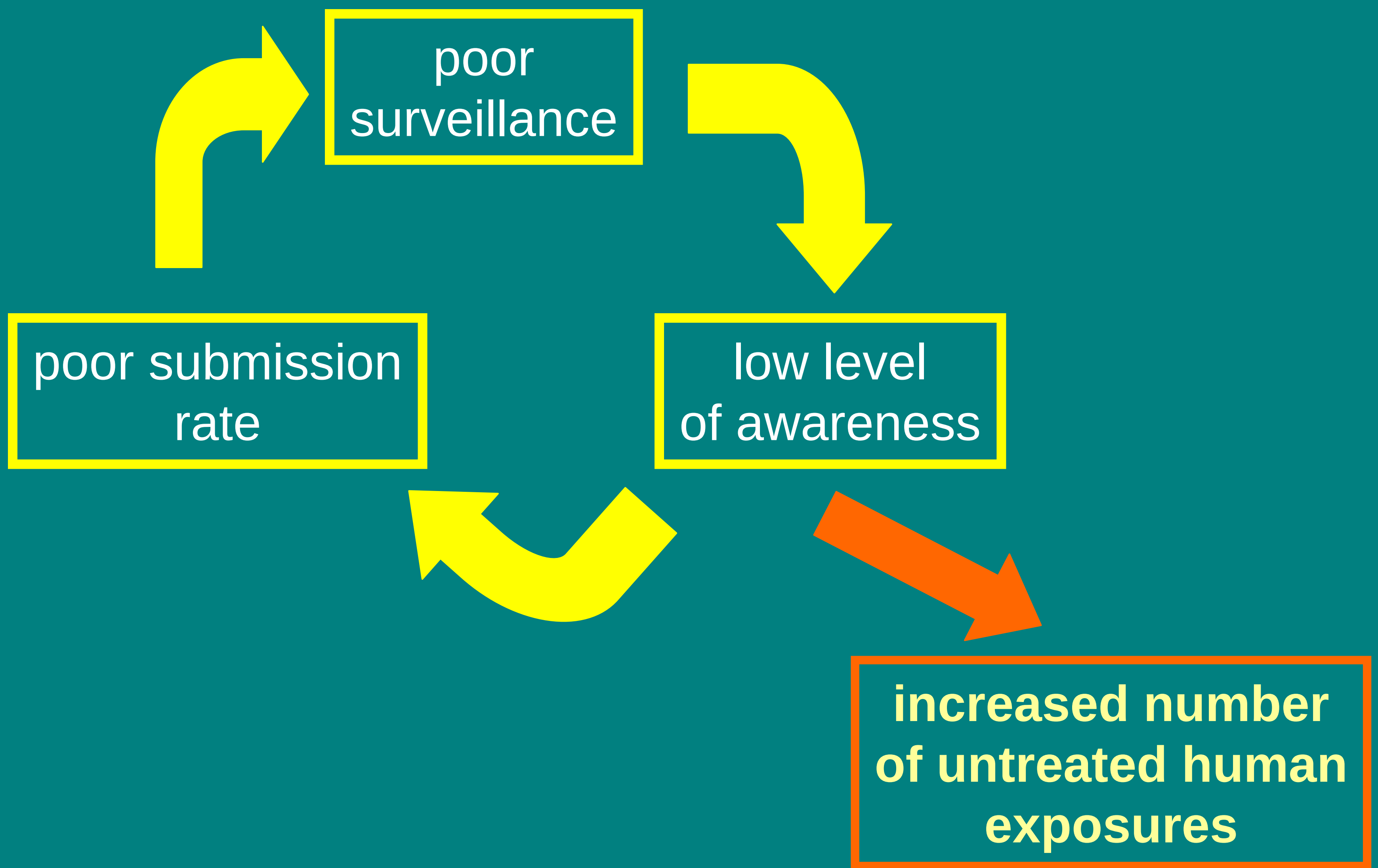
DOMINANT RABIES VECTOR AREAS OF SOUTHERN AFRICA



Rabies Surveillance

Alexander I. Wandeler
Centre of Expertise for Rabies
Canadian Food Inspection Agency

Surveillance and Human Rabies



The Purpose of Surveillance

- To understand the epidemiology:
  - to know the geographical distribution of rabies cases
  - to know the species that are involved in maintaining the epizootic
  - to know which species are important for rabies transmission to humans
- To promote rabies awareness
- To identify research needs

Surveillance

Good surveillance should

inform about the geographical distribution of rabies virus variants and their association with different host species;

allow the rapid detection of newly emerging, invading, and introduced virus variants;

permit to judge risks for humans, domestic animals and endangered species;

inform the planning and implementation of control programs

Surveillance and Epidemiology

- Adequate surveillance should permit epidemiological analysis
- Biased sampling inevitable (e.g. incidences unknown)
- Virus variant identifications necessary

Surveillance?

How good are our observations ?

How good are our interpretations ?

How good are our predictions ?

How to do surveillance

Agent (antigen) detection	Detection of immunological responses (antibodies)
Active Surveillance	Passive Surveillance

Agent (antigen) detection

- Routine rabies diagnosis

  usually by antigen detection (FAT) in brains of suspect animals

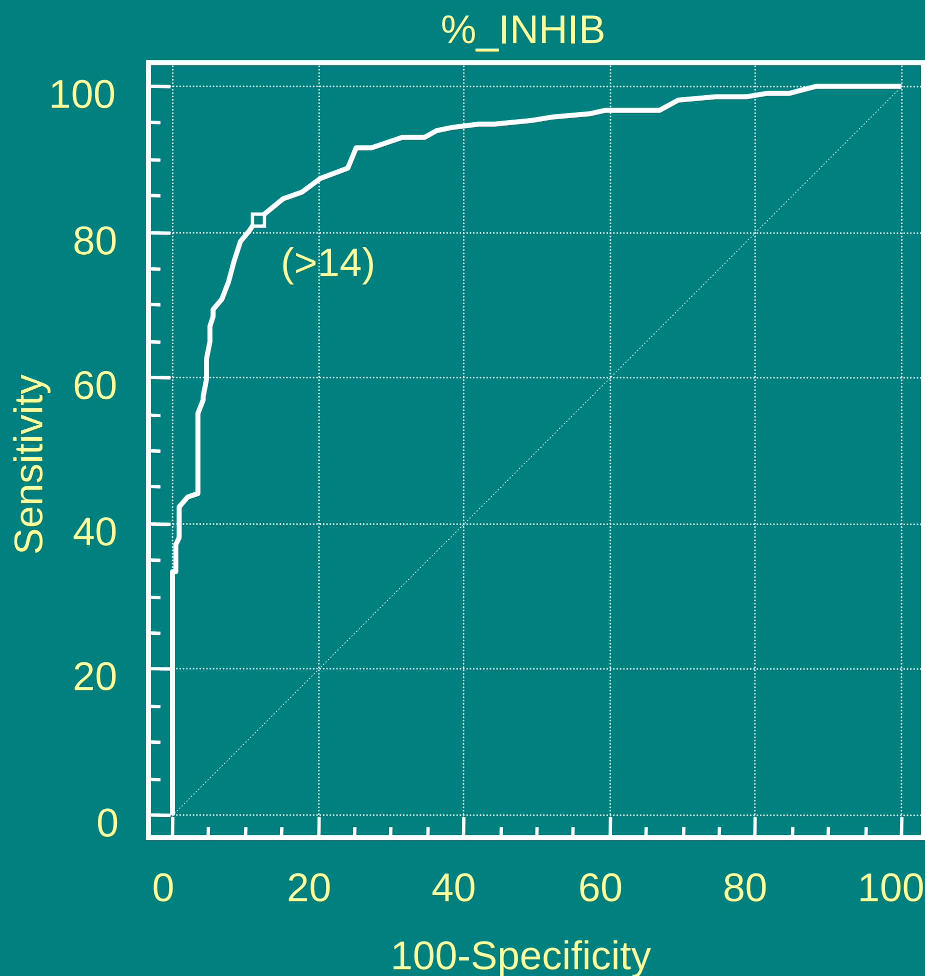
  sometimes followed by virus isolation and virus characterization
- Immediate tracking of zoonotic events

Detection of immunological responses

- Usually done by neutralizing antibody tests or ELISAs
- Historical tracking of epizootic
- Problems with test specificity and sensitivity
- Problems with interpretation
- Usually done without field controls

Raccoon Sera (exp.134)

ROC curve: c-ELISA versus NT



Active Surveillance

- Random sampling of host populations
- Difficulties:
  - coincidence of antigen presence in cns and clinical symptoms
  - short duration of clinical phase
  - low sensitivity
- Sampling method of choice for serological surveys

Passive Surveillance

Collection and analysis of suspect animals

- dependent on encouragements and restrictions
- likelihood of detection

Rabies prevalence (in %) in different samples in Switzerland

	Red fox	European badger	Stone marten
Shot by hunters	2	3	0
Trapped	9	-	0
Road kill	11	< 5	< 6
Found dead	60	44	10
Killed, abnormal behavior	**86**	**60**	**21**
n	4195	322	544

How to do surveillance

Antigen detection

recommended by WHO and OIE

Antibody detection

CAVE AT

useful in some situations, controls imperative

Active Surveillance

CAVE AT

Passive Surveillance

recommended by WHO and OIE

encouragements/incentives necessary

The OIE Reference Laboratory for Africa

23 January 2006

Heja Lodge, Windhoek, Namibia

Dr Claude Sabeta
OIE Ref. Lab. Rabies
OVI, Onderstepoort
SOUTH AFRICA

ARC • LNR

Memoranda of agreement

- Designation of OVI as an OIE Regional Collaborating Centre for Africa → reinforce the surveillance and control of animal diseses in southern Africa.
- Established in 1993, initial agreement between the OIE and the government of South Africa
- Funding expired 2002 (NDA to provide R255 000 annually)

Activities: reference laboratories

- To function as a centre of expertise and standardisation of a designated disease
- To store and distribute biological reference material for diagnosis and control of a disease
- To develop new procedures for diagnosis and control of a disease

Activities: reference laboratories (cont.)

- Coordination of scientific and technical studies in collaboration with other laboratories and/or organisations
- Publication and dissemination of any information which could be useful to member countries

Activities: additional

- To gather, process, analyse and disseminate epizootiological data
- To place expert consultants at the disposal of the OIE
- Provision of scientific and technical training for personnel from member countries
- Provision of diagnostic testing facilities to member countries
- Organisation of scientific meetings on behalf of the office

Objectives of the Centre

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

ARC-OVI OIE Reference Labs

- OIE reference laboratory for African swine fever - Dr C Phiri

- OIE reference laboratory for Rabies

  - Dr C Sabeta

- OIE reference laboratory for African horse sickness, bluetongue, lumpy skin disease and Rift Valley fever - Dr GH Gerdes

Activities, workshops and conferences

- Laboratory diagnosis of ASF to personnel at the Central Veterinary Laboratory, Entebbe, Uganda
- RVF-PCR training for groups from Uganda, Yemen and also from South Africa

Activities, workshops and conferences

- Distribution of anti-rabies polyclonal conjugate to member countries -

  (Eritrea, Uganda, Swaziland, Allerton,

  Malawi, Zimbabwe, Mali, Botswana,

  Zambia, Sudan, Swaziland, Namibia)

Workshops and conferences

- Financial support towards the Southern African Society of Veterinary epidemiology and Preventive Medicine, July 2002 (15 local and international scientists)
- ICPTV workshop – held jointly with ARC-OVI.

  Workshop on Tsetse and Trypanosiamiasis Research and Control in Southern Africa (November 2002)
- SADC-Diagnostic Subcommittee meeting, Mauritius March 2003, Quality Assurance Manager of ARC-OVI

Production, testing and distribution of diagnostic reagents

- Financial support to Mozambique for ASF diagnostic tests and virus isolations
- Providing ASF test reagents to Mozambique to perform surveillance
- Providing ASF antigen and sera to Uganda

Production, testing and distribution of diagnostic reagents

- Production of AHS antigen for molecular testing
- Lumpy skin disease sera supplied to Pirbright laboratory
- Production of Bluetongue antiserum for Onderstepoort Biological Products (OBP)
- Production of Bluetongue antigen for research purposes

Future perspectives (Rabies laboratory)

- Maintain virus archive (postgraduate student training, epidemiological studies, pathogenicity studies)
- Provide diagnostic training (animal health technicians), local and regional
- Allow for sabbatical training for other collaborators
- Provide diagnostic reagents to other regional laboratories

Control of rabies

- To improve the understanding of the epidemiology of rabies and rabies-related viruses
- Assist in the control of rabies through development of effective and socially acceptable dog immunisation strategies
- To improve diagnostic capabilities for rabies through establishment of modern, reliable and rapid diagnostic tests

Rabies reference laboratory

- Support and enhance ongoing and future scientific collaborations of the Rabies Unit, including those with Allerton Provincial Laboratory, University of Pretoria and International research institutions

- To liaise with members of the public and the veterinary and medical professions in order to give the necessary advice

==> Surveillance and control

In order to succeed,

- There is need to recruit well qualified personnel
- Continuous exposure by regular training
- Financial support (Gauteng Veterinary Government, National Department of Agriculture)

Thank you!!!

RABIES ADVISORY GROUP

(SOUTH AFRICA)

RAG HISTORY

- First meeting held in 1989 and was sponsored by pharmaceutical companies with the Onderstepoort Veterinary Institute, Institute of Virology, Veterinary Faculty and Veterinary Directorate represented
- Was formalised in 1991 as a “informal” group of scientists from the National (NDA) and Provincial Departments after a surge in rabies cases in Kwazulu-Natal

RAG HISTORY (cont)

- In 1995 formal recognition was requested for RAG to become an advisory group to the ICTA Veterinary Working Group
- Formal recognition was granted in 1996. Members include veterinarians (state and private), health professionals, Onderstepoort Veterinary Institute (OVI) and Institute of Virology
- Terms of reference drafted in 2000

MANDATE

- Review research, technology and development i.r.o. vaccines, treatment, care etc.
- Review policy on control and management of rabies
- Evaluate rabies epidemiology, nationally and internationally
- Identify research areas and motivate for funding

MANDATE (cont)

- Co-ordinate collaboration between various role players e.g. NDA, ARC/OVI, CDC
- Make recommendations to provinces i.r.o. control and management
- Access, produce, procure audio visual awareness aids
- Advisory group, not an action group
- Liaison within SEARG and internationally

ACTIVITIES OF RAG

- Meet twice a year
  - Discuss matters referred by provinces and National Department
- Identify "hot spots" and liaise with provinces with regard to control measures
- Design various forms to be used with regard to rabies control, e.g. sample submission form, vaccination certificate etc.


TOILET→

VETERINARY SERVICES

RABIES SPECIMEN SUBMISSION FORM

Please complete all sections. If there is insufficient space, please write on the back of the form.
Submit specimens on ice or in 50% glycerol saline in clearly labelled leak proof containers.
Please do NOT use Formalin.

FOR LABORATORY USE ONLY:

Date received :	Time received :	Label ref no :	Rabies no :
Report date :	Time out :	Exam. 1	Result :
RPO :	Signature :	Exam. 2	

SENDER	Name :		Sender Ref No :	
	Address :			
	Fax :		Tel :	
	Signature :		Date :	
OWNER (OR STRAY)	Name :		Tel :	
	Address :			
FARM	Farm name :		Farm No :	
	District Road :			
LOCATION OF CASE	Grid reference (KZN only) :			
	Geographic location :	East :	South :	
	Local municipality :			
	Magisterial district :		SV area :	
	District municipality :		Province :	
SPECIES	Common name :		*Certain :	*Uncertain :
	Scientific name :		*Certain :	*Uncertain :

AGE (IF DOG)	Puppy (< 6 months) :		GENDER (IF DOG)	Male :
	Juvenile (6 - 12 months) :			Female :
	Adult (> 12 mths) :			Neuter :

CLINICAL HISTORY	Date first clinical signs :	
	Date of death :	
VACCINATION HISTORY (DOGS ONLY)	Unknown :	
	Known :	
	Vaccinated, date unknown :	
	Vaccinated, date known (state) :	
HUMAN CONTACTS (NUMBER OF)	Unknown	
	Saliva or handling contact (Category 1) :	
	Superficial bites, no bleeding (Category 2) :	
	Superficial or deep bites, wounds bleeding (Category 3) :	

CONTACT DETAILS (OF HUMAN CONTACTS)

Name; street address :		Tel :
Name; street address :		Tel :
Name; street address :		Tel :
Name; street address :		Tel :
Name; street address :		Tel :

REPUBLIC OF SOUTH AFRICA
VETERINARY SERVICES

IDENTITY, RABIES VACCINATION AND MOVEMENT CERTIFICATE FOR DOGS AND CATS MOVING BETWEEN PROVINCES IN THE REPUBLIC OF SOUTH AFRICA

NB KEEP THIS CERTIFICATE AS PROOF OF VACCINATION. TAKE IT WITH YOU FOR REVACCINATIONS AND WHEN YOUR DOG OR CAT MOVES BETWEEN PROVINCES.

A1 Identification and description of animal:

Dog ☐ Cat ☐ Male ☐ Female ☐

Date of birth : ..../............./............ Colour : ....

Name of animal : .... Breed : ....

Distinguishing marks : ....

Microchip number : ....

A2 Owner's name : ....

Owner's address : ....

....

A3 This certificate serves as an official vaccination certificate for interprovincial and international movements on condition that -

1. it accompanies the animal;
2. the property of origin is free from quarantine restrictions imposed for rabies contol purposes;
3. the certificate is signed and stamped in the space below and
4. all requirements of the importing country have been met.

A4 Vaccination record

Vaccinations	Date	Name of vaccine	Batch No.	Signature and address of veterinarian / authorised official
Primary				
Revaccination				

A5 General information on rabies vaccination of dogs and cats in South Africa :

All dogs and cats 3 months and older must initially be immunised twice within a twelve month period, administered at least 30 days apart, and thereafter every three years. Vaccination must be carried out by an authorised person using a registered vaccine.

Puppies and kittens under the age of 3 months may be vaccinated, provided they are revaccinated at 3 months and then according to the above schedule.

Pregnant dogs and cats can be safely vaccinated.

For international movement, some countries require vaccination by a registered veterinarian and some stipulate annual rabies vaccinations. It is the owner's responsibility to ascertain the requirements of the importing country.

No : 13152

ACTIVITIES OF RAG (cont)

- Compile guidelines (RABIES GUIDE FOR THE MEDICAL, VETERINARY AND ALLIED PROFESSIONALS), SOP's (vaccination campaigns) related to the control of rabies
- Liaise with the Department of Health with regard to the handling of human dog bite wound patients and related problems

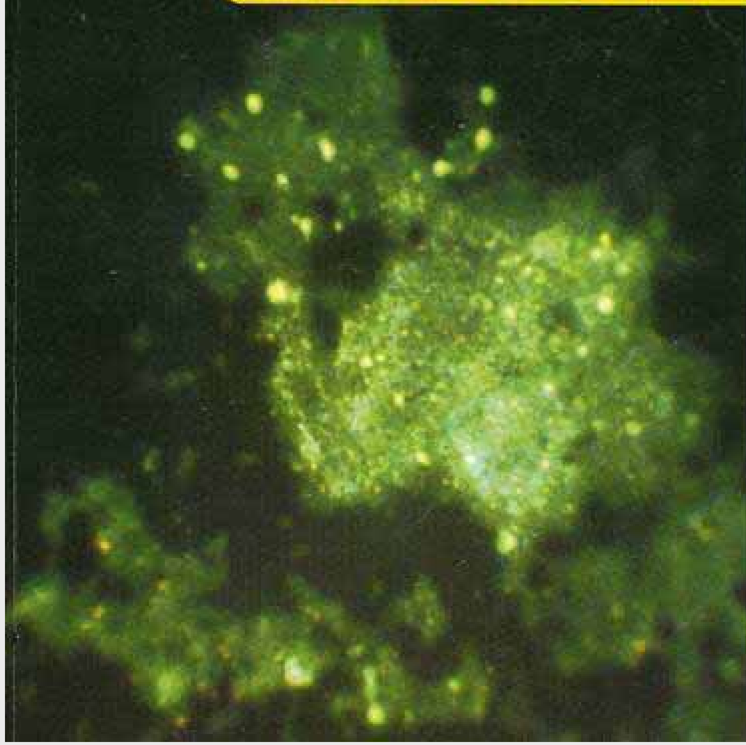
FUNCTIONS OF RAG (cont)

- Liaise with the Onderstepoort Veterinary Institute with regard to research needs
- Monitor the prevalence of rabies in the country
- Identify suitable sample containers

RABIES

GUIDE FOR THE MEDICAL, VETERINARY AND ALLIED PROFESSIONS

G.C. Bishop
D.N. Durrheim
P.E. Kloeck
J.D. Godlonto
J. Bingham
R. Speare
and the Rabie
Advisory
Group



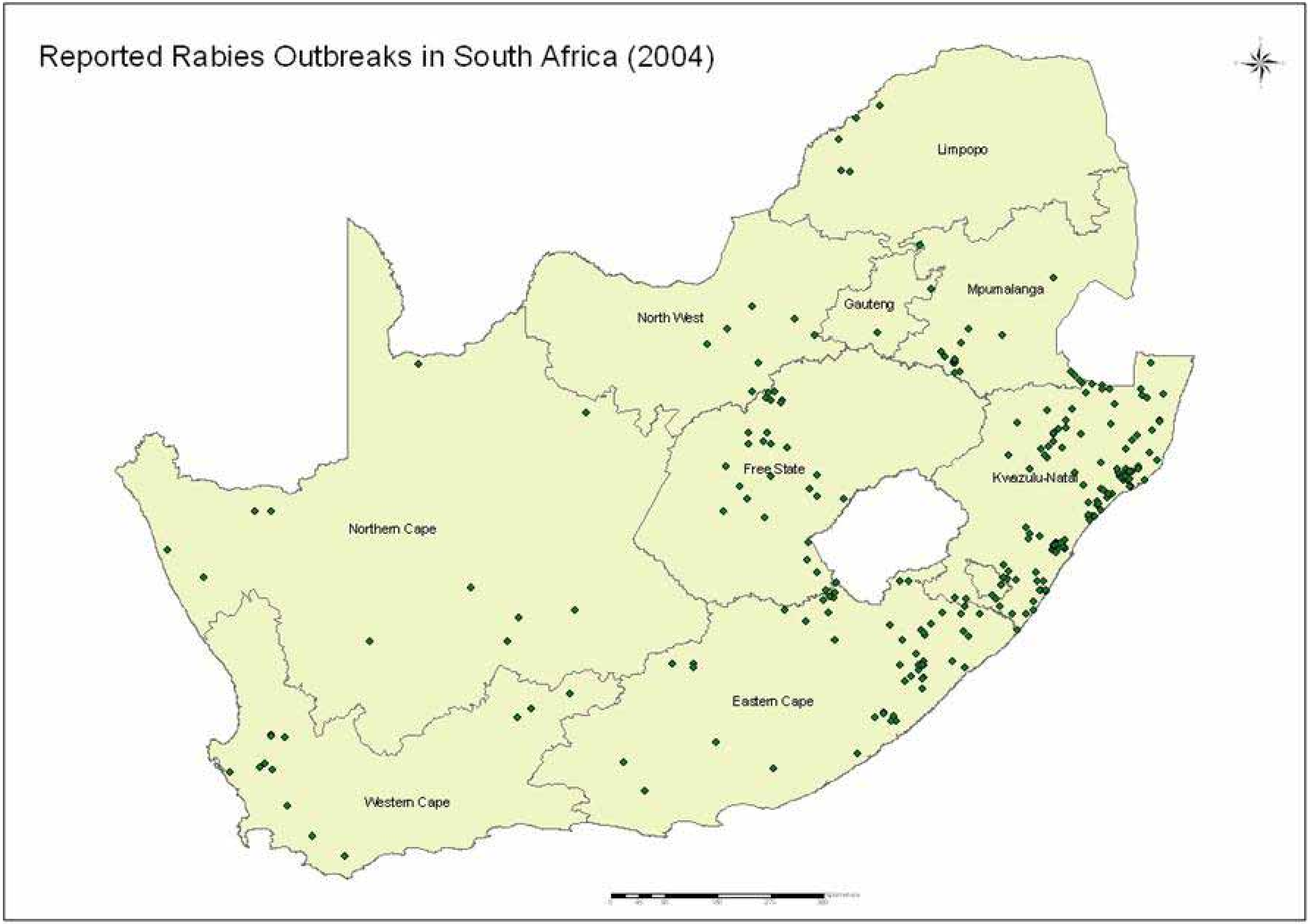
Department: Agricultu

Draft Rabies Campaign

SOP 2nd Edition

Compiled by : K. Le Roux in collaboration with KZN technical staff

CONTENTS


Reported Rabies Outbreaks in South Africa (2004)
Limpopo
Mpumalanga
Gauteng
North West
Free State
Kwazulu-Natal
Northern Cape
Eastern Cape
Western Cape

Alliance for Rabies Control

What is ARC and what does it hope to achieve?

Katie Hamilton

Alliance for Rabies Control
Easter Bush
Roslin, Midlothian
EH25 9RG, UK

email: arc@rabiescontrol.org
www.rabiescontrol.org

Concept

- What and why ARC?
- ARC – the **Alliance for Rabies Control** – charitable organisation intending to bring together national and international expertise on rabies to implement or assist in the implementation of control programmes
- Main objectives:
  - promote public health and advance the education of the public in matters regarding rabies and the risk of infection
  - relieve the suffering of animals and promote conservation and animal welfare in developing countries of Asia and Africa by alleviating the burden of rabies
- Focus on the domestic dog reservoir

Concept

- To achieve these aims, we need:
  - International commitment
  - Local commitment
  - Understanding of the epidemiological situation in target areas
  - Well defined programme objectives
  - Reliable source of human and animal vaccine
  - Consistent funding
  - Relatively small scale programmes

- Who is ARC?

Alliance for Rabies Control

Directors

- Individuals who will advise on the overall management of the charity
- Ultimate decision on grants and proposals
- Skilled managers

Technical Advisory Group (TAG)

- Experts in the rabies field
- Invited by Directors
- Permanent World Health Organization representative

Members

- Membership is free
- Any individual who has an interest in rabies
- No companies – only individuals
- Directors can decline a membership application

Current status

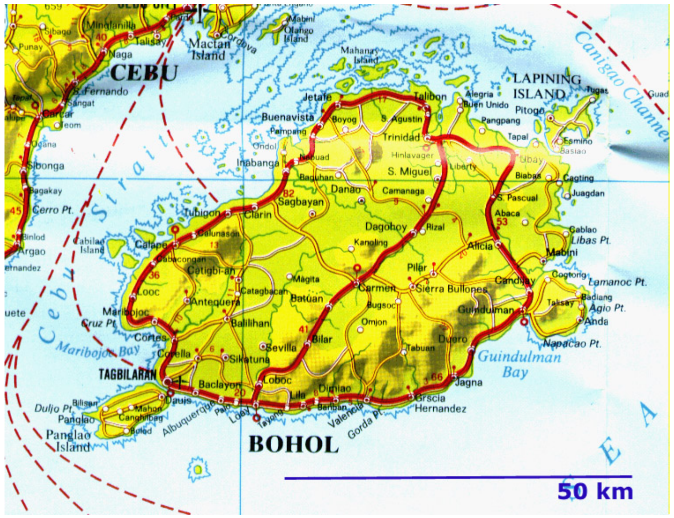
- Registration process in Scotland
- Alliance for Rabies Control (ARC) is registered as a Limited Company with Charitable status
- Somewhat long-winded administrative process. Now complete.
- Potential sources of funding will include governments, private donations, block grants (yet to be approved by Directors), etc
- Largely project-led
- Decisions as to where to apply for funding will be discussed at the first directors meeting.
- Open invitation for proposals

A model intervention

- A proposal written by Betsy Miranda to combat rabies in Bohol, Philippines is under discussion and has been agreed in principle by ARC.
- Funding required: US$ 100,000; local authorities to raise 50%.
- The programme is expected to run for 2 years.
- Year 1 - programme execution
- Year 2 - post-campaign maintenance and epidemiological follow-up.

Bohol, Philippines

- Rabies is endemic
- Human population: 1,137,268
- Estimated dog population: 98,500 (2000 figures)
- Reported 9 human cases and more than 8000 post-exposure treatments - over 50% of these cases are children below 15 years.
- Target vaccination of 69,000 dogs in 2 campaigns over 12 months.
- Pre-requisites: political will: a concept of a rabies control programme approved by local government.
- Pre-requisites: support of the local population:
- Lacking: resources: vaccine acquisition, programme funds



Relevance to SEARG

- Country proposal originating from countries, even if draft
- ARC will assist with pre- and post intervention epidemiological investigation and impact assessment – e.g.: under-reporting, human mortality, dog incidence, patient follow-up
- Assessment of programme needs - what scale does control programme need to be implemented – national or regional?
- Financial needs
- Vaccine requirements - humans and animals
- Work with government (national or sub-national) to design and implement programme - local staff, local structures
- Cost-effectiveness assessment

Where to get more

- www.rabiescontrol.org
- Regular updates of progress with projects
- Information on meetings and plans
- Consider the invitation for proposals open…
- Email us at arc@rabiescontrol.org

Making a difference: Saving lives

Rabies prevention programs

Deborah J Briggs
Kansas State University

Rabies - Facts

- Rabies has the highest case fatality rate of any known infectious disease
- The ‘real’ burden of rabies is unknown because of a lack of hard data
- Little governmental funding for rabies prevention programs
- Highest burdens of human disease are in Africa and Asia

Burden of rabies – Africa and Asia

Similarities

- Main vector – canine
- Main victims – children
- Funding – difficult to non-existent
- Status – neglected
- Vaccines – CCV and NTV
- RIG – scarce, expensive
- Canine control programs are few and erratic
- Many countries with varying degrees of wealth

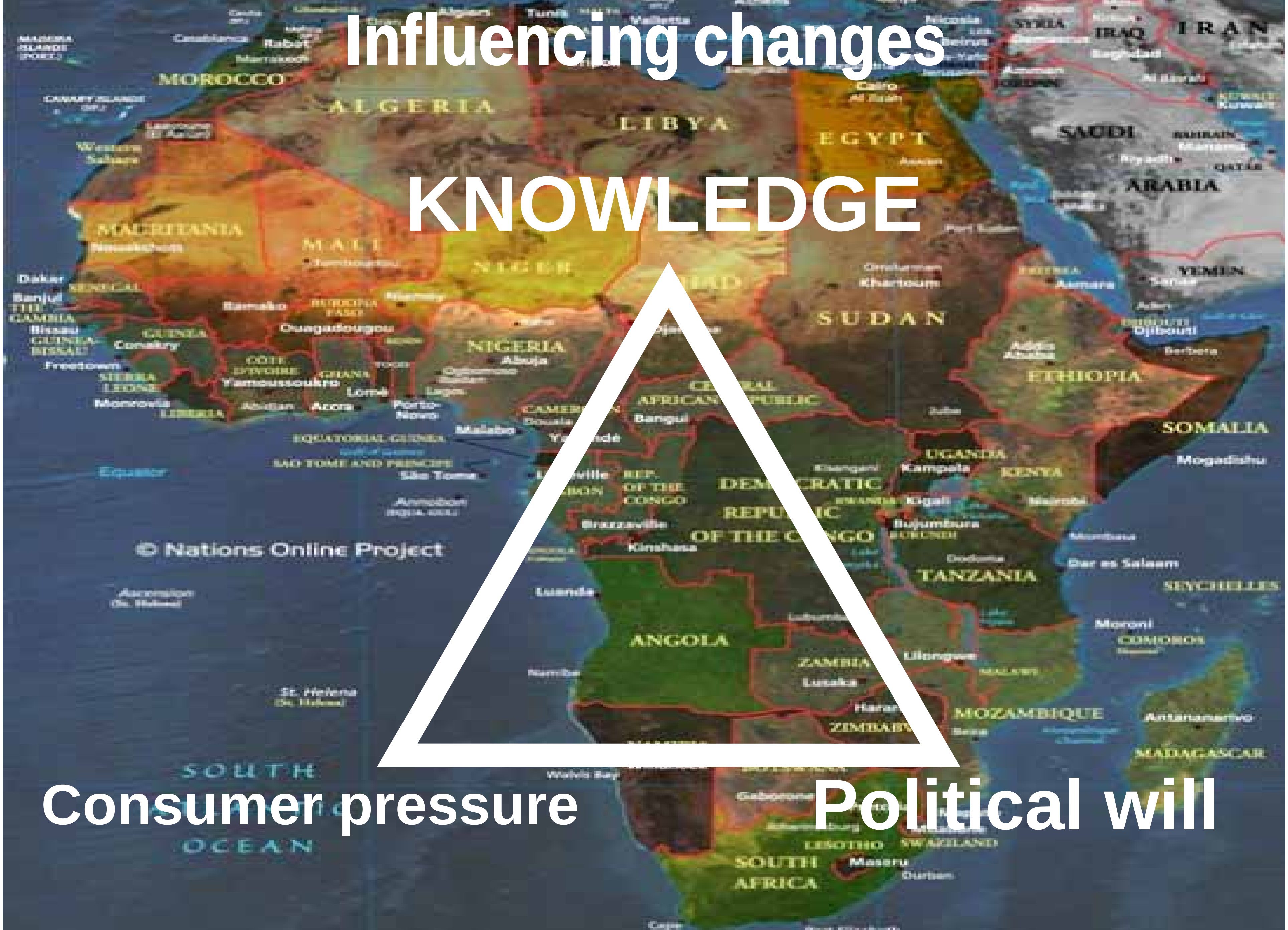
Burden of rabies – Africa and Asia

Differences

- ***Number of human deaths has significantly decreased over the past decade***

- **Asia –more local programs, reduced dose regimens, more physicians**

- **Africa – Established rabies group (SEARG), more vets, wildlife rabies captures international attention**


Influencing changes
KNOWLEDGE
Consumer pressure
Political will
© Nations Online Project

Thailand in the Eighties

Population:	**55 Mill**
Dog population:	**>10 mill**
Human rabies cases:	**appr. 300/year**
No. of postexposure treatments:	**appr. 100,000/year**

Nerve tissue rabies vaccine discontinued in 1987 in Bangkok, in the early nineties in the rest of Thailand

Deficiencies in post-exposure management due to high cost of cell culture rabies vaccines and RIG

Wilde, Chutivongse, Progress in Rabies Control, 1988

Historical view from Asia

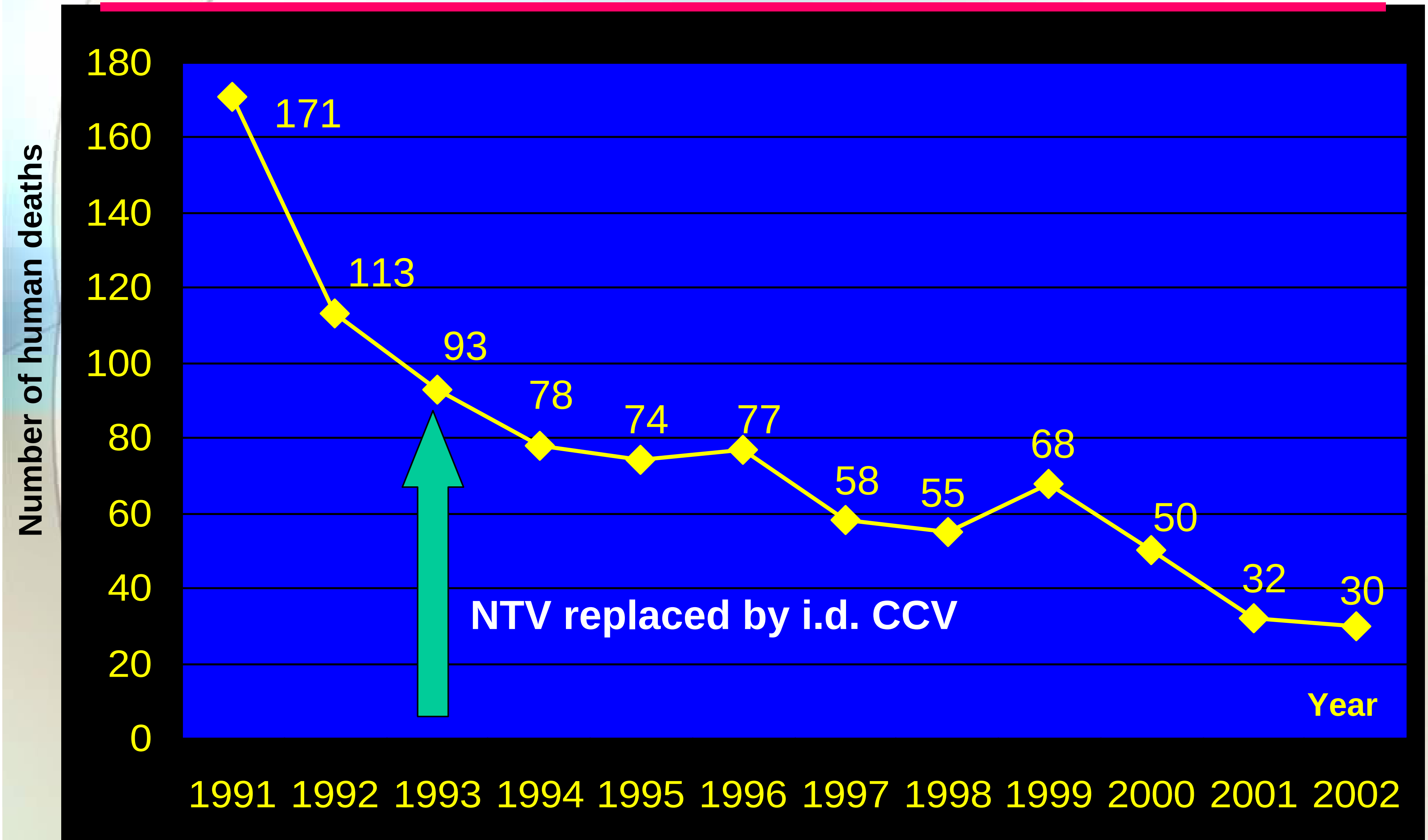
- **1984 – Thailand: Prof Supawat - QSMI**
- **"You kill your beloved Thai people everyday"…**

Rabies Control in Thailand

Year	Deaths	Number treated
1984	**228**	**74,086**
1985	**205**	**79,977**
1986	**205**	**79,977**
1987	**212**	**84,178**
1988	**219**	**79,454**
1989	**212**	**84,178**
1990	**185**	**88,312**
1991	**171**	**93,641**
1992	**113**	**116,222**
1993	**93**	**133,946**
1994	**78**	**148,112**
1995	**74**	**160,000**
2002	**30**	**>300,000**

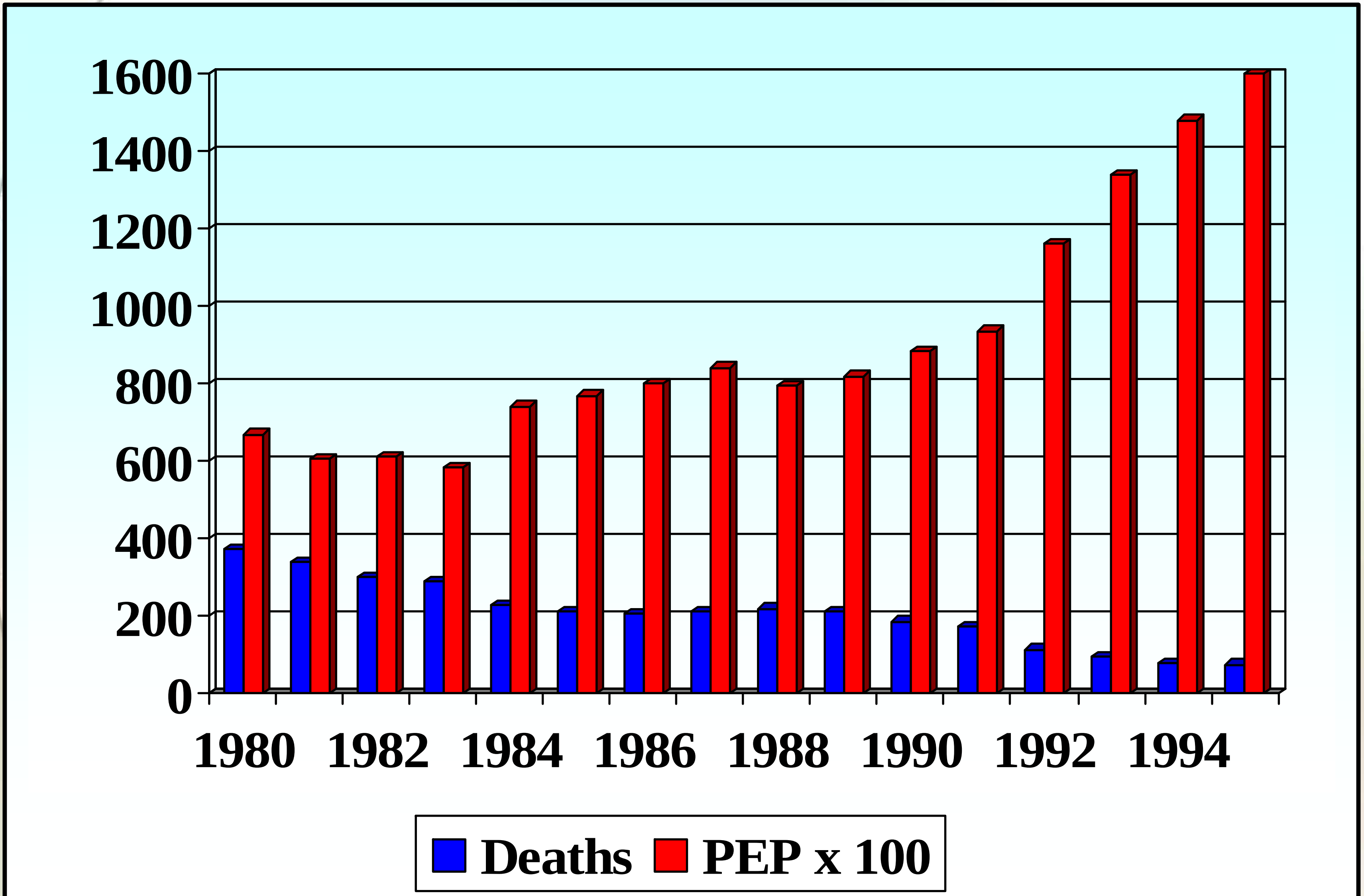
Nerve tissue vaccine replaced by the use of intradermal Cell Culture Vaccine

Human Rabies Deaths in Thailand



Source : Choomkasien P. Division of Epidemiology, Ministry of Public Health, Thailand

Rabies Control in Thailand

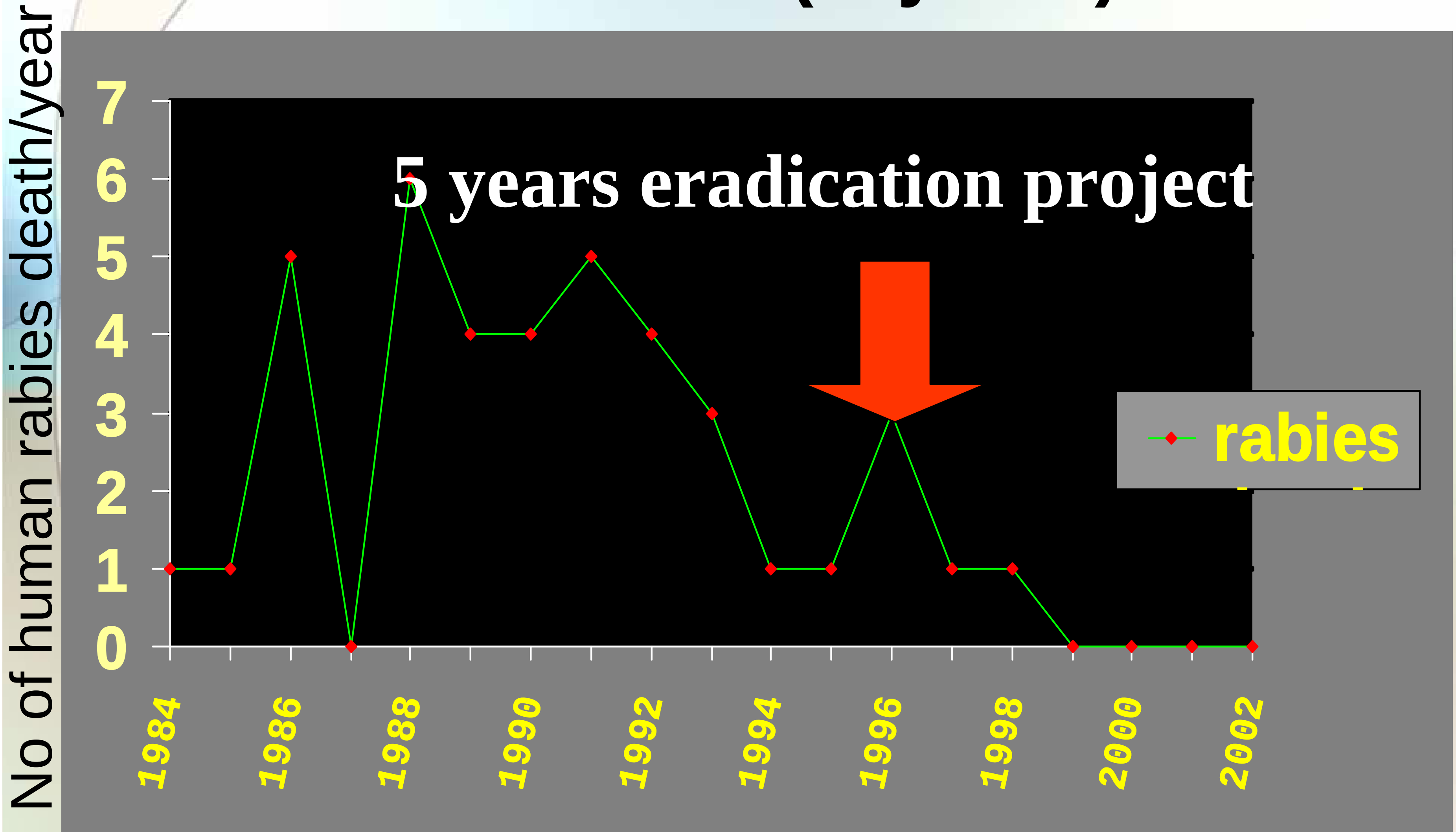


Wasi et al. Vaccine. 15:S7, 1997.

Building on past experience

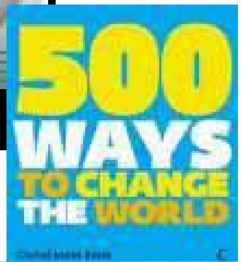
- Dr Thavatchai Kamoltham – Petchabun Province, Thailand
- Over 40 % of deaths in children under 15 years of age
- Innovative new project to reduce human rabies to 0 deaths in 5 years

Human Rabies Death in Phetchabun 1894-2002 (19years)



Source: Report 506 Epidemiological unit Provincial Health office of Phetchabun

Petchabun rabies progress



- Ministries of Health, Agriculture, Education, NGOs

Influencing other nations

- **Sri Lanka**

- **Philippines**

- **India ~~– NTV 2005~~**

Determination to change

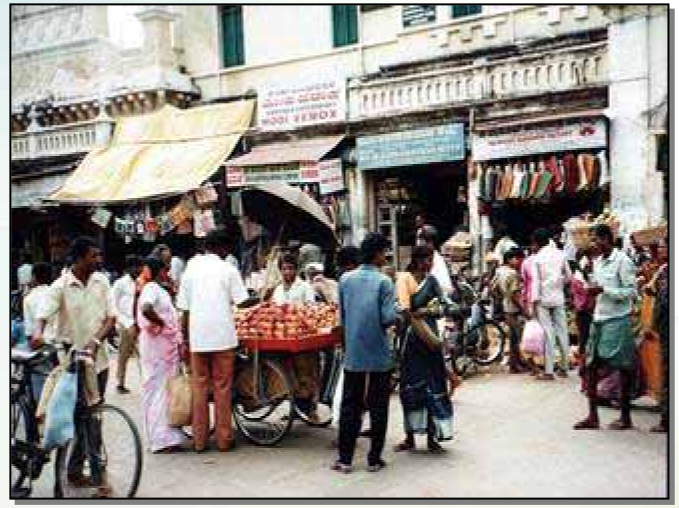
- Instituting intradermal across an entire country – Sri Lanka – stopped NTV production – Dr Omala

- Creating a cease-fire to institute teaching seminars – Tamil Tigers

Finding ways to improve diagnostic facilities – foreign governments

Funding programs

- What about places that have little or no funding?



India

- Decentralized government
- HIV
- Malaria
- TB
- Parasitic diseases
- Religious, cultural considerations

South America/PAHO success story – is it possible in Asia?

India

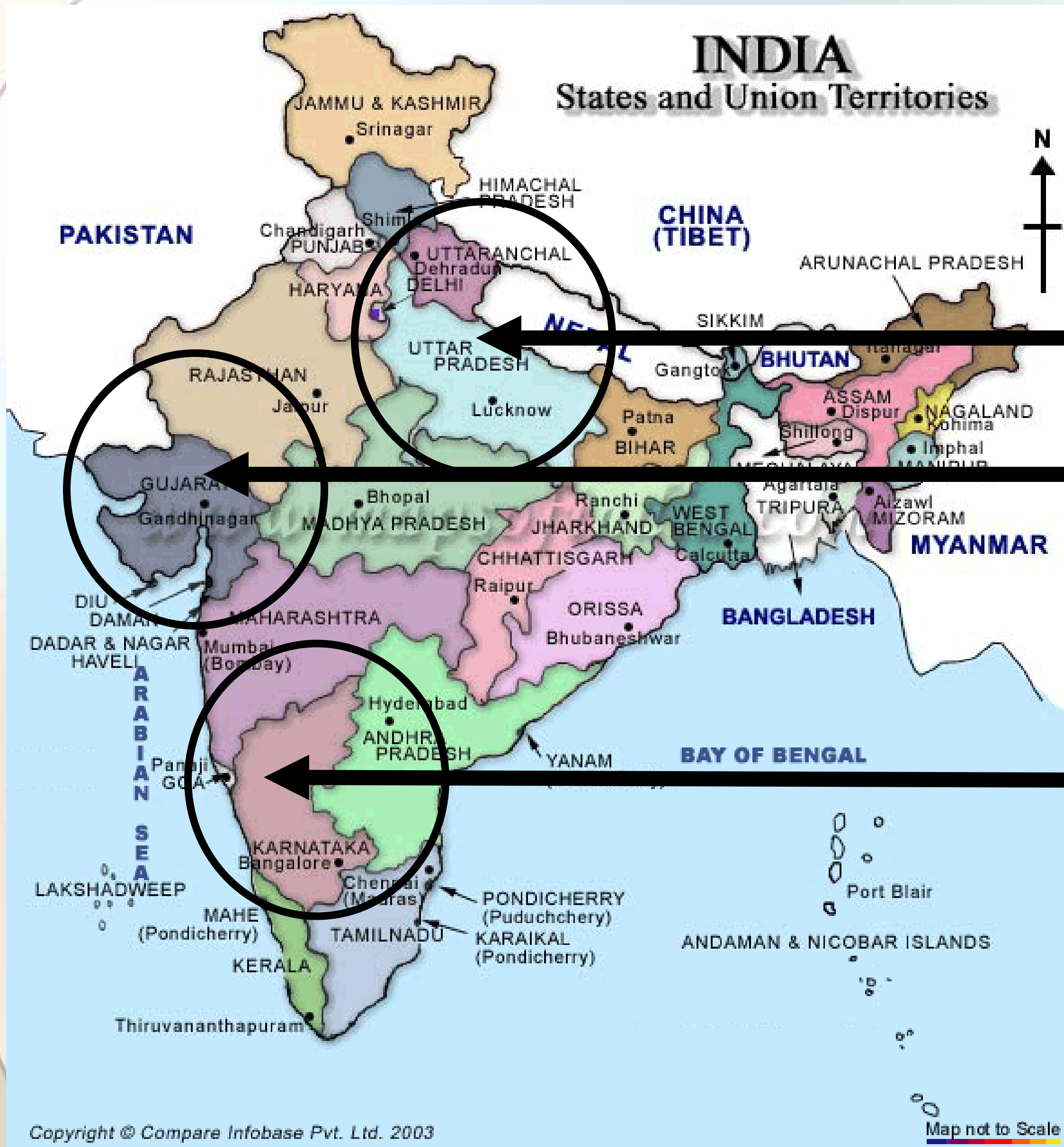
•200 to 250 new patients daily

•Long lines, crowded clinics

India – Funding PEP

- One doctor in State MoH decided to stop NTV in Uttar Pradesh
- Pushed state government to secure funding from the World Bank
- Cooperation with Health systems Development Projects of the respective states
- Included CCV in the list of essential medicines, changed the order of importance and hence could manage to get funds
- Great press and so the Chief minister of the state took personal interest to insure that there would be a continuous supply

India – Funding PEP



0.5 mio doses

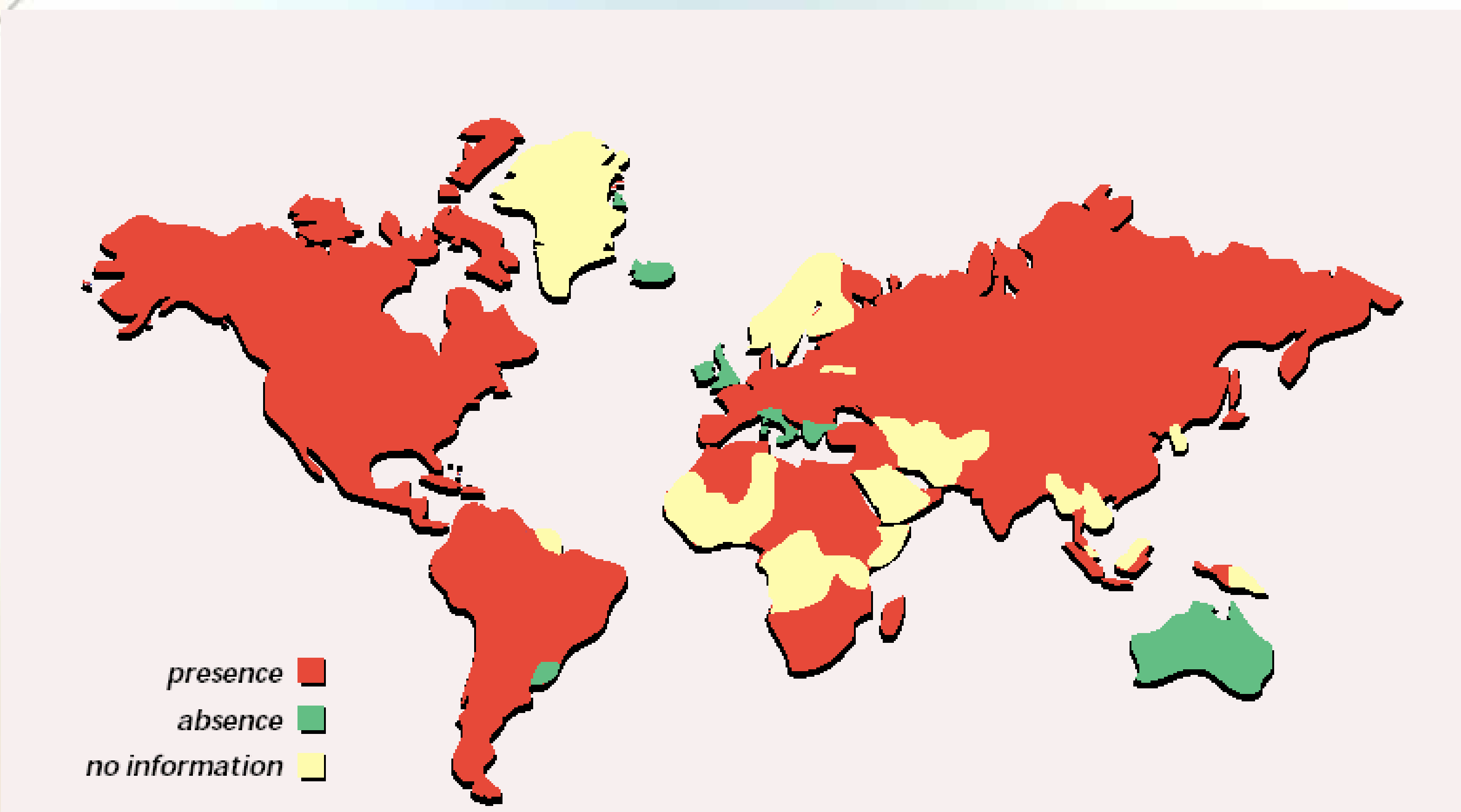
30,000 doses

0.5 mio doses

India

- In Karnataka 4 physicians created a paradigm shift in rabies education
- Are bringing rabies education down to the village level
- Developed a teaching plan, involved medical and veterinary students to work with all levels of the community

Conclusions



One person can change the face of rabies

The Importance And Value Of Dog Ecology Studies In Rabies Control.

Roland Xolani Dlamini

Introduction

- African Economic environment characterized by severely limited resources
- Intense competition for those resources
- Rabies programs must generate and justify statistics to the powers be.
- Clearly spell out amount of resources (value) required, how they will be used and the Benefits.

Introduction cont.

- Discussing benefits – include control of rabies in dogs, savings on post exposure treatments, saving human lives, preventing losses in domestic animals.
- Economists are convinced by figures rather than the passionate demonstration of the suffering of human victims before finally succumbing to death from rabies.
- Carefully crafted ecology studies can generate meaningful (useful) figures

Introduction cont.

- Constraints to effective dog rabies control are not only economic but also logistical such as inaccessibility of some dogs to vaccination.
- Precious resources employed in the traditional mass vaccination at designated centers = not effective
- Unless accompanied by aggressive initiatives to reach those inaccessible dog populations.
- Dog ecology can identify and characterize such populations.

Rabies – a population problem

- Rabies is fatal to the host, no carrier status
- Maintenance of the disease denotes indefinite transmission through members of host population.
- Successful maintenance = average transmission ratio = or > 1 (Bingham2005)
- Local population unlikely to maintain the disease
- Persistence requires many local populations (Metapopulation).
- At any one time virus must be present in at least one local population

Rabies – a population problem cont.

- 70% local population vaccination coverage = transmission + maintenance curtailed (WHO)
- However, depends on potential contact between infected and susceptible –a function of dog densities, level of restriction and dog turnover (Perry 1993)
- Therefore level of risk different for different local populations.
- Dog ecology studies – identify and characterize high risk populations
- Limited resources directed to high risk populations for greatest possible effect.

Rabies – a population problem cont.

- Movement of infected dogs from one population to another is key to maintenance of the virus.
- Rabies travels in minibuses from one local population to another (Bishop et al)
- Careful ecology studies can reveal how rabies moves between your country's local populations.
- Effective movement control thru public awareness, certification can control the disease.

Pertinent ecology Information

- Population: size, spatial distribution, age distribution, turnover, sex ratios,breeds/strains presence of ownerless dogs (numbers distribution, characterization of their habitat), fertility, fecundity.
- Management info: uses of dogs, feeding, movement restriction, accessibility (ownership + restraint), reproduction control, access to veterinary services.

Pertinent ecology information cont.

- Owners information: cultural acceptability (attitudes) towards vaccination, reproduction control, access to veterinary services.
- Human exposure information: dog bite records,circumstances of the bite, treatment received by victims, fate of bitten individuals, attitudes towards dog bites and rabies post exposure.

Methods of collecting ecology information

- No one method is adequate to collect all pertinent ecology information.
- Questionnaire Surveys have been used to collect information on owned dogs – accessible owners
- Can be supplemented with other methods such as Mark & Recapture Technique and Field Observations – especially to get information on ownerless dogs.

Conclusion

- A good knowledge of local dog ecology [dog numbers, turnover, reasons for ownership, management, attitudes of the hosts(owners)] is crucial when planning control measures and negotiating for resources.
- High risk populations are identified and formulation of appropriate strategies to effectively deliver limited resources and achieve the greatest possible positive effect for every dollar spent.

Conclusion cont

- Dog ecology studies must not be once- off or a snap shot but must be continuous.
- Integrated to the overall rabies and/or dog population control program(s)
- To document trends as they occur and adapt strategies accordingly.
- Risk level of local populations will change overtime- due to successful control interventions or major changes in the environment.
- E.g. change in disposal of hotel and other waste can create or remove habitat for ownerless dogs.

RABIES PROJECT

DOG POPULATION

SURVEY

&

PRIMARY HEALTH CARE


BEWARE!

BACKGROUND

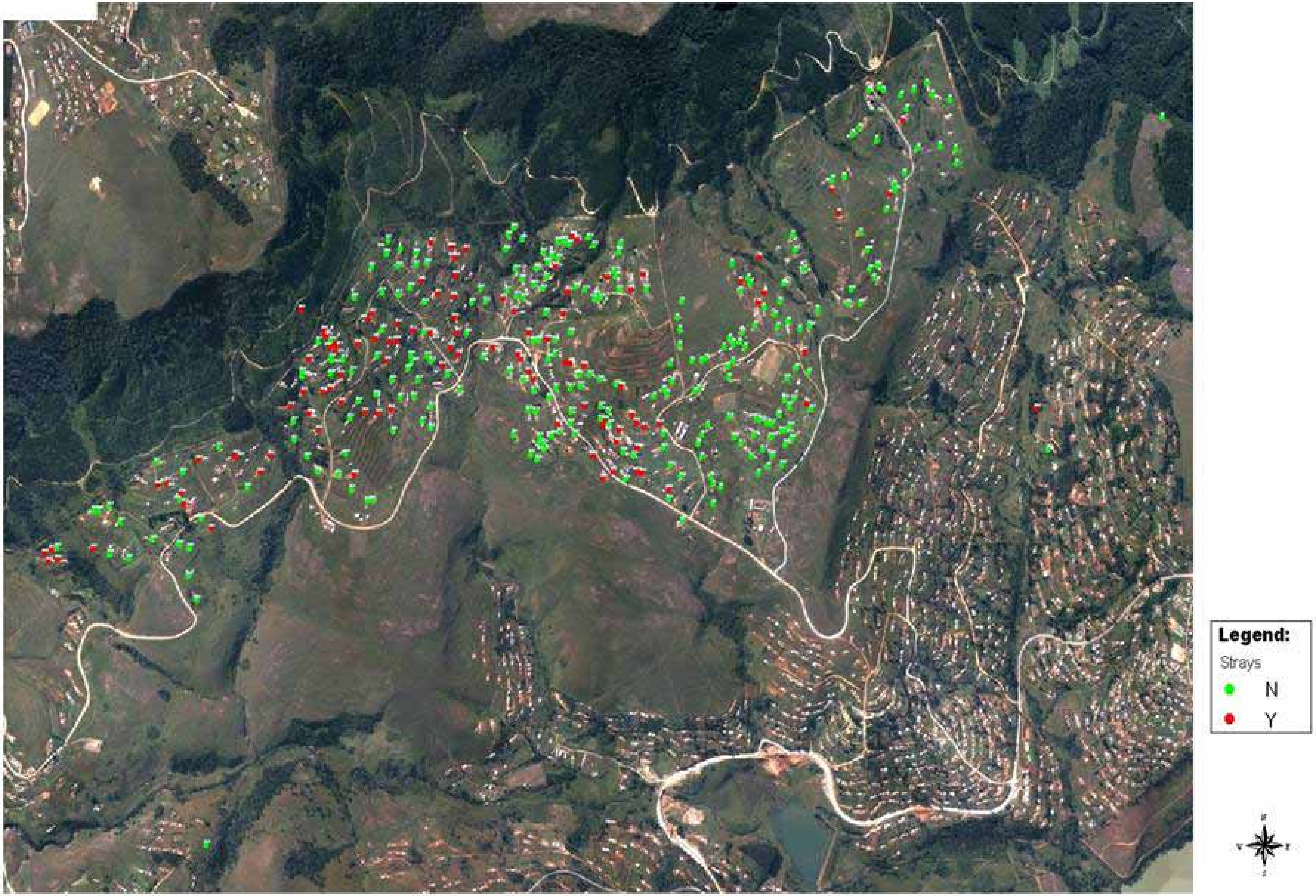
- Mass campaigns have dropped Rabies cases by 30%
- Still areas where Rabies remains in spite of vastly improved vaccination campaigns
- Question arises of WHY ?
- Once again need for accurate baseline data is demonstrated

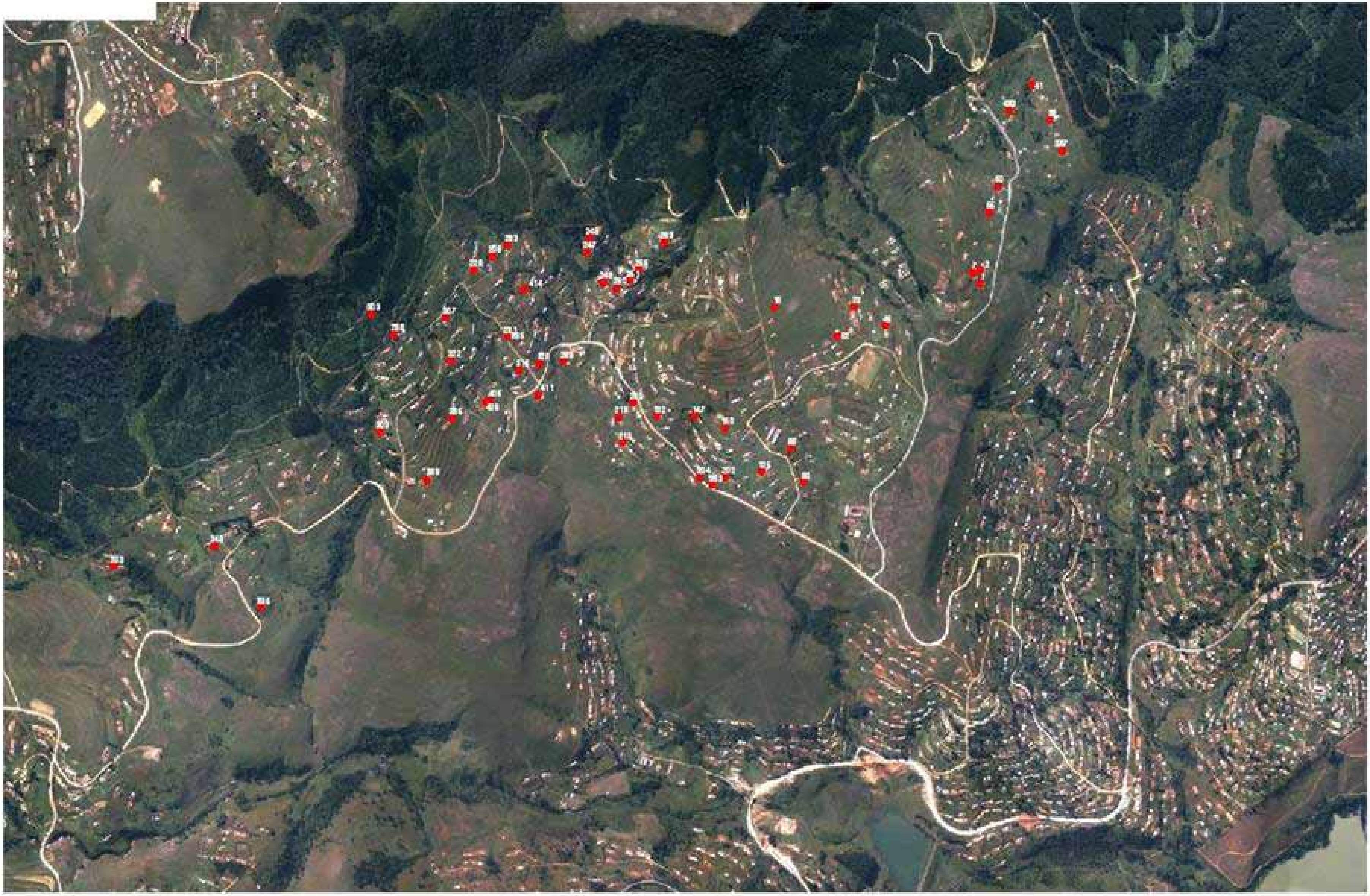
BACKGROUND(1)

- Community complaints of stray dogs
- SPCA and Departmental staff removed 25 Stray dogs (further 12 identified but not removed)
- Village is isolated and therefore ideal study area
- Full cooperation from Induna and community

BACKGROUND(3)

- Recent aerial photos available.
  - These proved to be invaluable in the project.
- Cooperative effort between Department and SPCA.
- Private practitioners involved in the primary health care.
- Involvement of other role players.
  - Traditional leaders.
  - Municipal councilors.


Legend:
Strays
N
Y



BACKGROUND(4)

- Information needed
  - Scientific dog census
  - Dog population dynamics
  - Needs assessment for primary health care
  - Importance of the "stray' dog population
  - Impacts of human population changes on dog ownership(Migration, AIDS,Unemployment, poverty)

PROCEDURES

- Aerial photos used to divide area up into sections.
- Planning meetings held with all role players.
- Technician training day held.
- Questionnaire drawn up.
- Equipment obtained.
  - Catch poles.
  - Cages.
  - Etc.

PROCEDURES(2)

- Teams of 2 technicians visited every household in village
- Questionnaires completed
- GPS coordinates taken of every household
- Unvaccinated dogs vaccinated
- Households visited marked with red hazard tape

PROCEDURES(3)

- Households with dogs for sterilization were identified by yellow hazard tape on gate.
- These households were also plotted on the aerial photos to enable technicians to collect dogs on the day of sterilization.





PRELIMINARY RESULTS

- Total of 414 sites visited (cf. 395 identified from aerial photos)
- 383 completed questionnaires used for analysis
- Non residential sites excluded (churches, stores etc.)
- A number of questionnaires excluded due to incomplete information

PRELIMINARY RESULTS

- 145 households kept dogs (37.8 %)
- 339 dogs recorded
  - Dogs per households keeping dogs 2.33
  - Dogs per household (all households) 0.89
- 2,220 people in village
  - 5.8 people per household
- People: Dog ratio 6.5:1

PRELIMINARY RESULTS

- 63 households (18.5 %) had no one home during the day
  - Reasons
    - Abandoned (12) 19.0 %
    - No Response (9) 14.3 %
    - Owners at work (42) 66.7 %
  - Number of dogs at these houses 28 (8.2 % of total)

PRELIMINARY RESULTS

- 44 dogs were removed on request of the owners
- 29% of households reported stray dogs as a problem
  - Eating eggs
  - Killing chickens
  - Chasing other livestock
  - (Very few reported a health or human danger problem)

PRIMARY HEALTH CARE

- The following week a sterilization clinic was held
  - 50 males
  - 36 female animals sterilized
- 60 of these were identified during the survey
- 26 were "walk ins" during the clinic

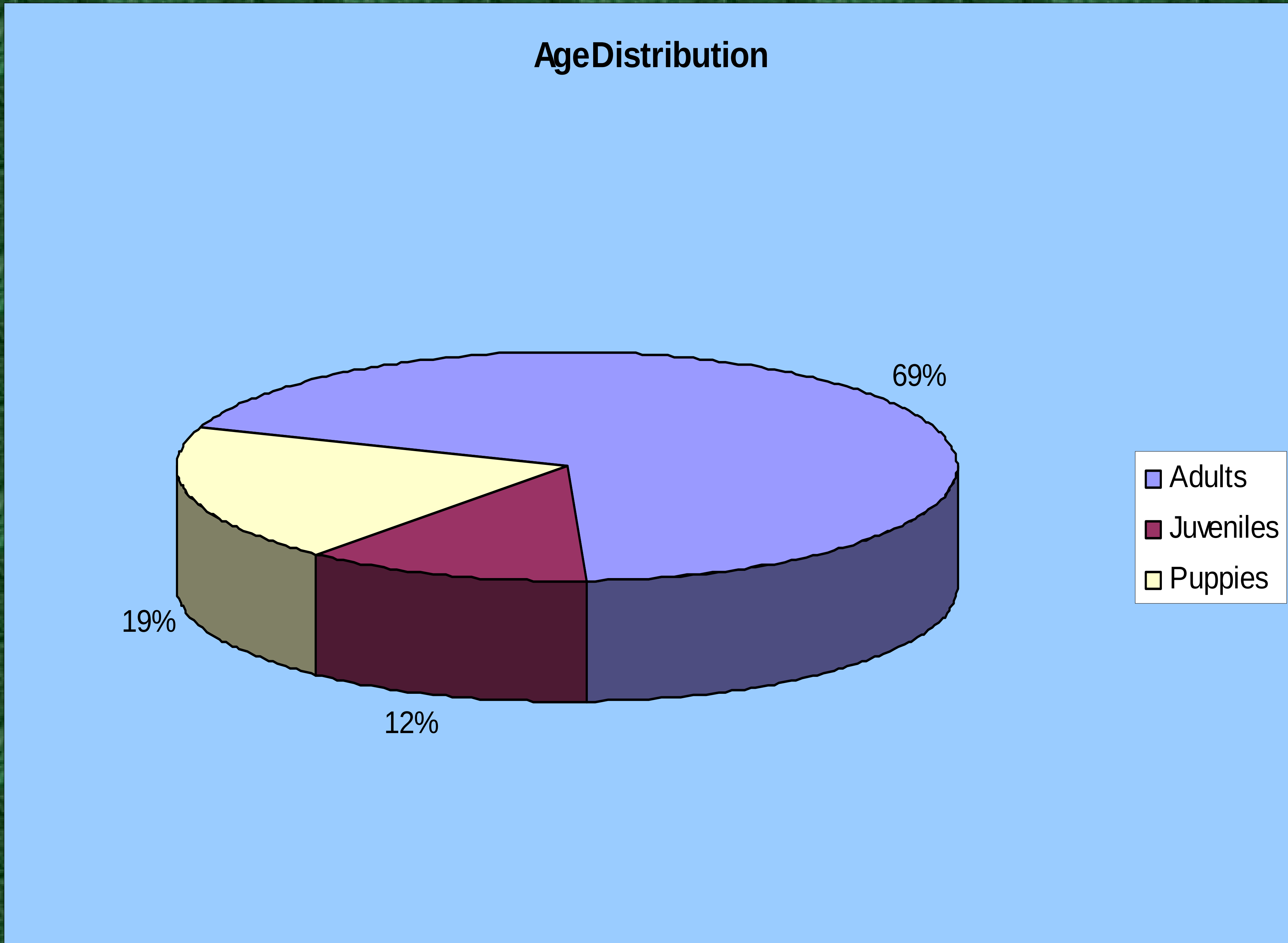
FOLLOW UP CLINIC

- On request of the community a follow up clinic was held and 23 male and 10 female dogs were sterilized
- A further 4 owned dogs were removed
- 22 puppies removed from 3 pregnant bitches
- 9 dogs for sterilization could not be caught (Importance for Rabies campaigns)

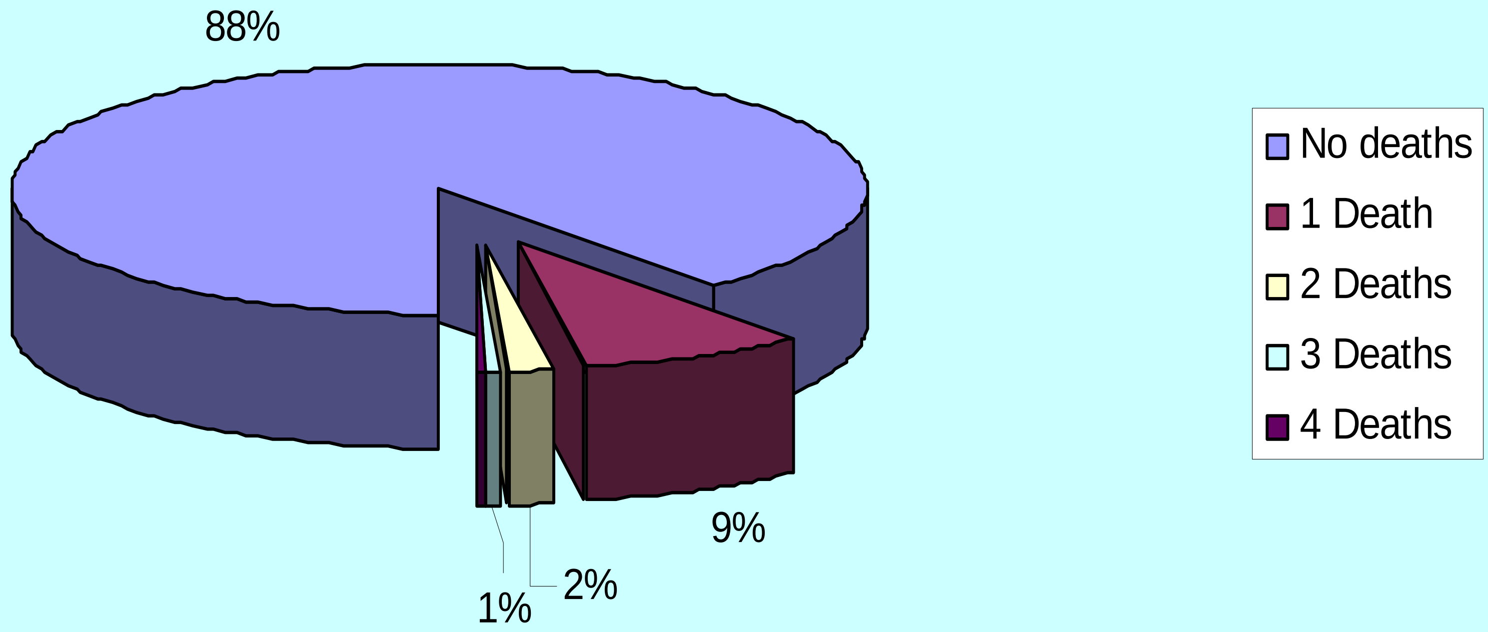
PRIMARY HEALTH CARE(2)

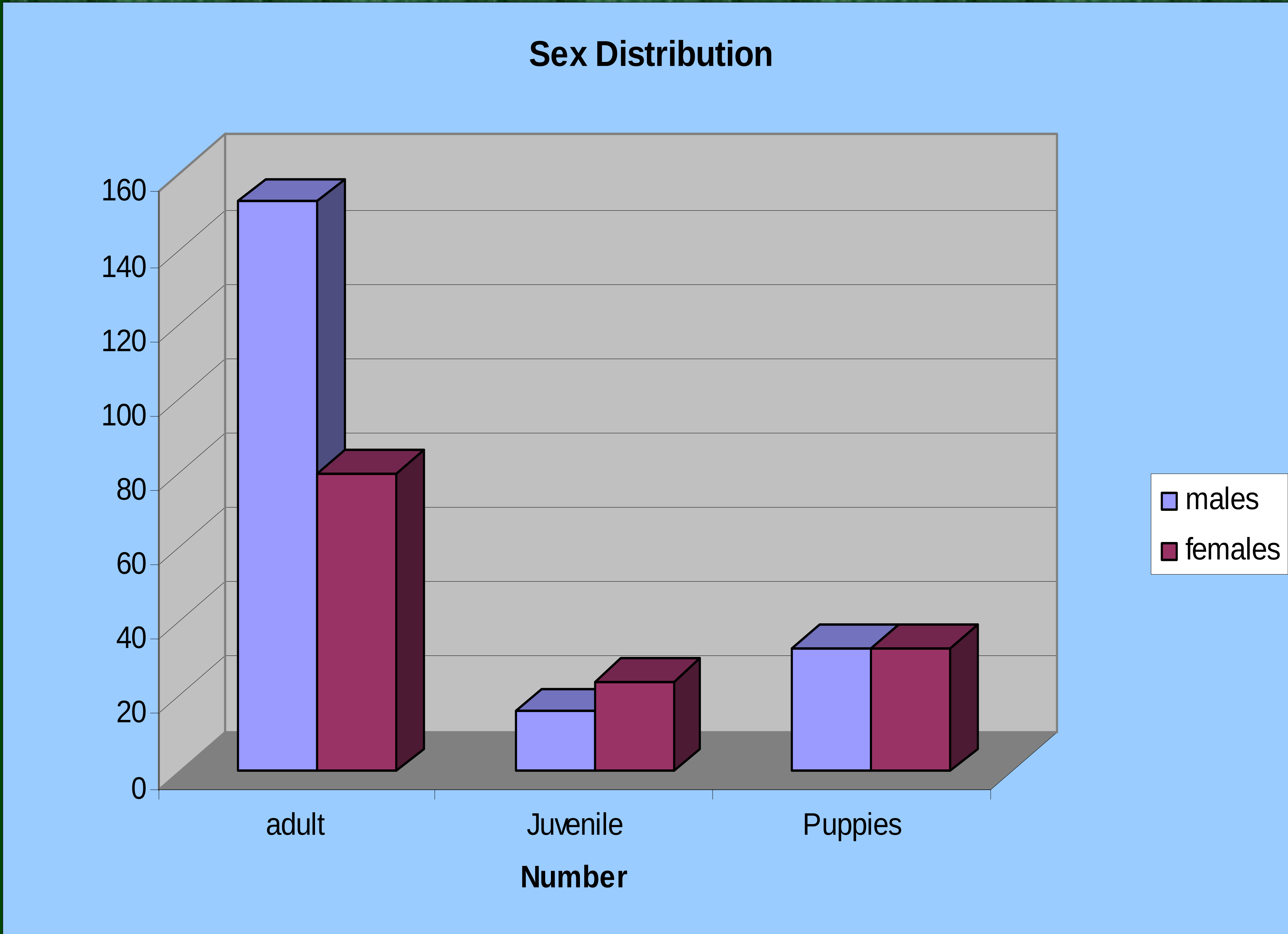
- Dogs were collected by the technicians and returned after recovery from the anaesthetic.
- This was to reduce number of dogs waiting at the clinic site.
- Sterilizations were done by State Veterinarians, the SPCA veterinarian and volunteers from private practitioners.
- Primary health care (deworming, vaccinations, mange treatments etc) also carried out.


SPCA
PIETERMARITZBURG



AgeDistribution
69%
19%
12%
Adults
Juveniles
Puppies

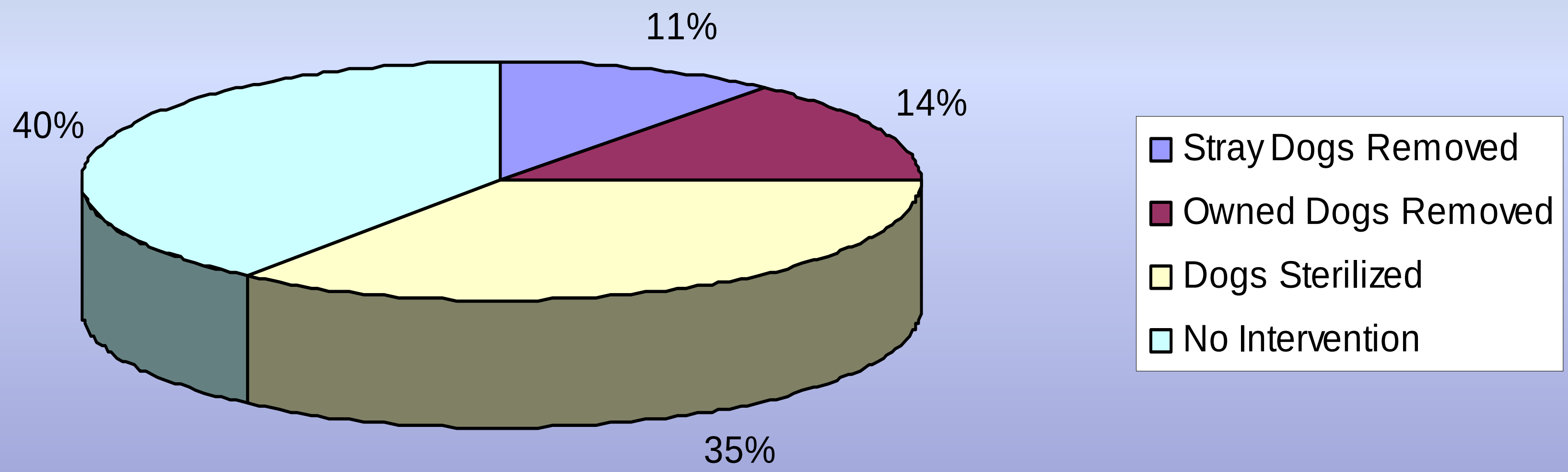
Adult Deaths Reported





Sex Distribution
160
140
120
100
80
60
40
20
0
adult
Juvenile
Puppies
Number
males
females

Interventions Nxamalala village



DISCUSSION

- Best Rabies vaccination figures for this village was 130
- This being done by a very dedicated technician

- This is only 38 % of total dog population

DISCUSSION

- At a Human:Dog ratio of 6.5:1

KWAZULU NATAL SHOULD HAVE A DOG POPULATION OF

1,300,000

DISCUSSION

DURING 2004 A TOTAL OF

497,224 DOGS WERE VACCINATED

38 %

DISCUSSION

- Is the fact that nearly 20% of households had no one home significant ?
- Is the fact that 3 % of the houses in the area were abandoned significant ?
- Is the fact that 9/33[27.2%] dogs(follow up clinic) could not be caught significant ?
- How did the removal of strays before the survey affect the response to the question re stray dogs ?
- Further analysis of the data is being carried out and this will also identify new directions to be investigated in future surveys.

DISCUSSION

- Dogs not available for “normal’ vaccination campaigns (in Nxamala village)
  - 11 % stray population
  - 8   % Owners not at home
  - 25 %   Owners not able to catch
- THEREFORE
  - No matter how good the campaign

44 %

of the dogs would not be vaccinated

DISCUSSION

- Exercise needs to be conducted in Rabies "hot spot" area
- All areas need to conduct similar surveys to further refine the data
- Technicians in general need more specialized training in survey techniques
- Recent aerial photography essential

QUESTIONS

- What is a stray dog?
  - True feral populations
  - Owned dogs joining feral packs on temporary basis
  - Owned dogs that have been abandoned
  - Dogs following bitches on heat
  - Dogs scavenging for food (not fed at home)

QUESTIONS

- **What alternative methods of vaccination can be used ?**
  - **The role of bait vaccination needs to be investigated**
- **How applicable are these results to other areas ?**
- **Census needs to be repeated in all areas to confirm ratios**
- **What other means of rabies control can be applied ?**
- **How can public awareness and cooperation be increased**

Using an Epidemiologic Model for Community-Based Rabies Control

Pamela Woods,[a b]

Laura Hungerford,[b]

David Hartley[b]

b

a

Faculty Veterinary Science

University of Zimbabwe

Mathematical Models for Communication for Rabies Control

Pamela Woods,[a b]

Laura Hungerford,[b]

David Hartley[b]

b UNIVERSITY OF MARYLAND SCHOOL OF MEDICINE 1807

a Faculty Veterinary Science

University of Zimbabwe

Rabies behaves differently in different populations

- The likely behaviour of disease epidemics described mathematically
- Rabies in Foxes in Europe, Raccoons in USA, Dogs & Jackals in Africa

- Debate about “self-sustaining” epidemics or if “outside sources” required
- Critical selecting correct control measure

Classifying disease threats from Reservoir Hosts (H_1) and Target Hosts (H_2)

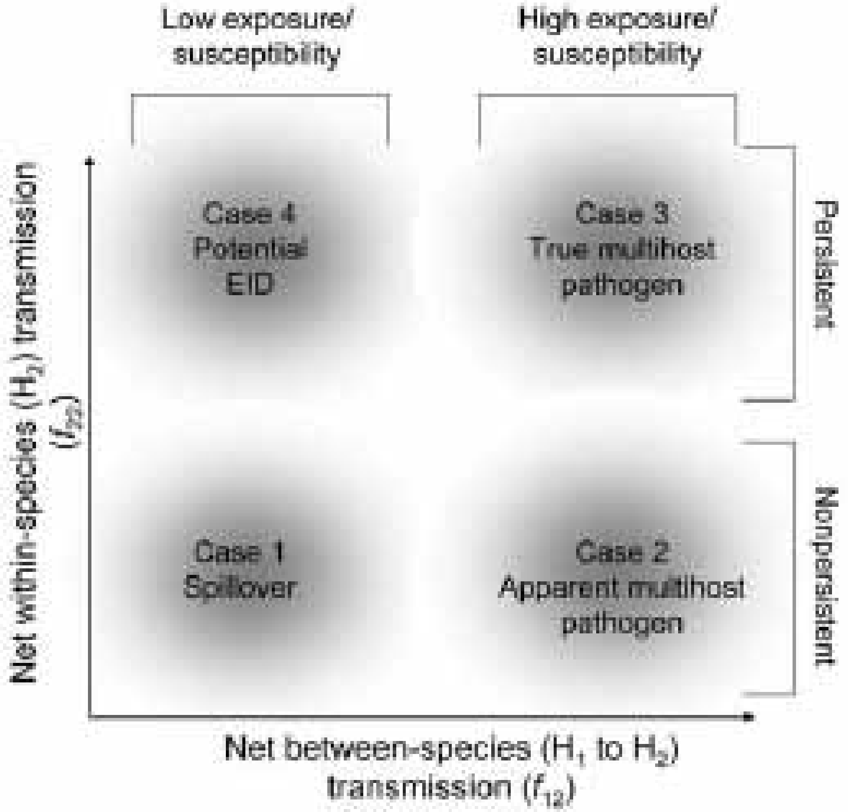


Figure 2. Community-epidemiology continuum, determined by the net between-H_1 and -H_2 transmission rate (f_{12}) and the net within-H_2 transmission rate (f_{22}). EID, emerging infectious disease.

- X axis - Transmission from Reservoir to Target
- between species

Fenton & Pedersen, 2005

Classifying disease threats from Reservoir hosts (H_1) and Target hosts (H_2)

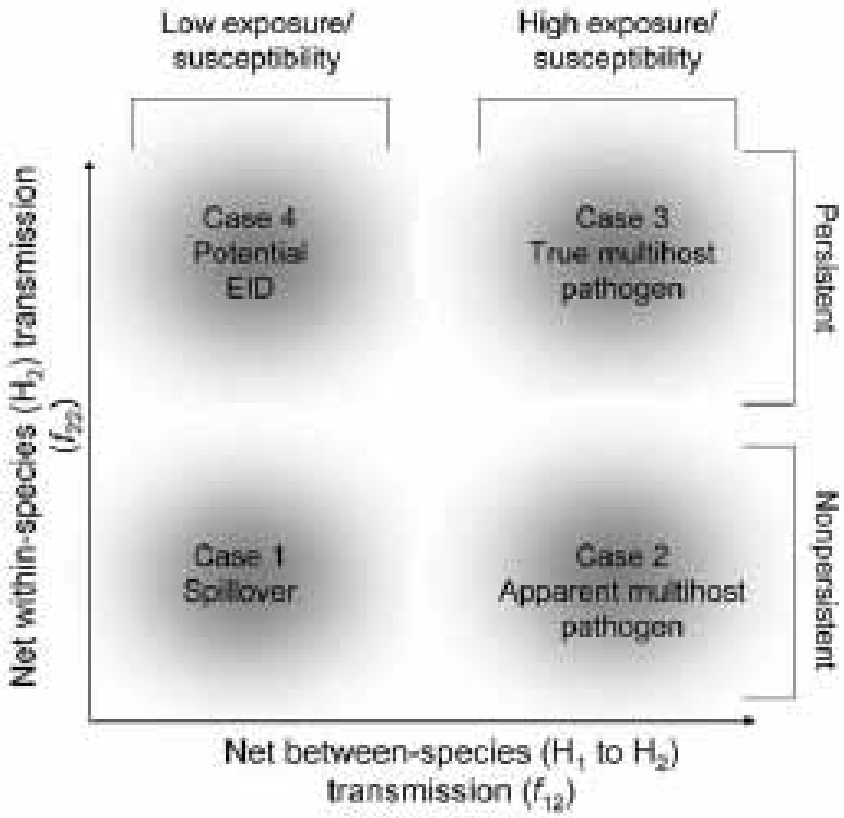


Figure 2. Community-epidemiology continuum, determined by the net between-H_1 and -H_2 transmission rate (f_{12}) and the net within-H_2 transmission rate (f_{22}). EID, emerging infectious disease.

Fenton & Pedersen, 2005

Y axis Target host to Target host (H_2 to H_2)

- within species

Classifying disease threats from Reservoir hosts (H_1) and Target hosts (H_2)

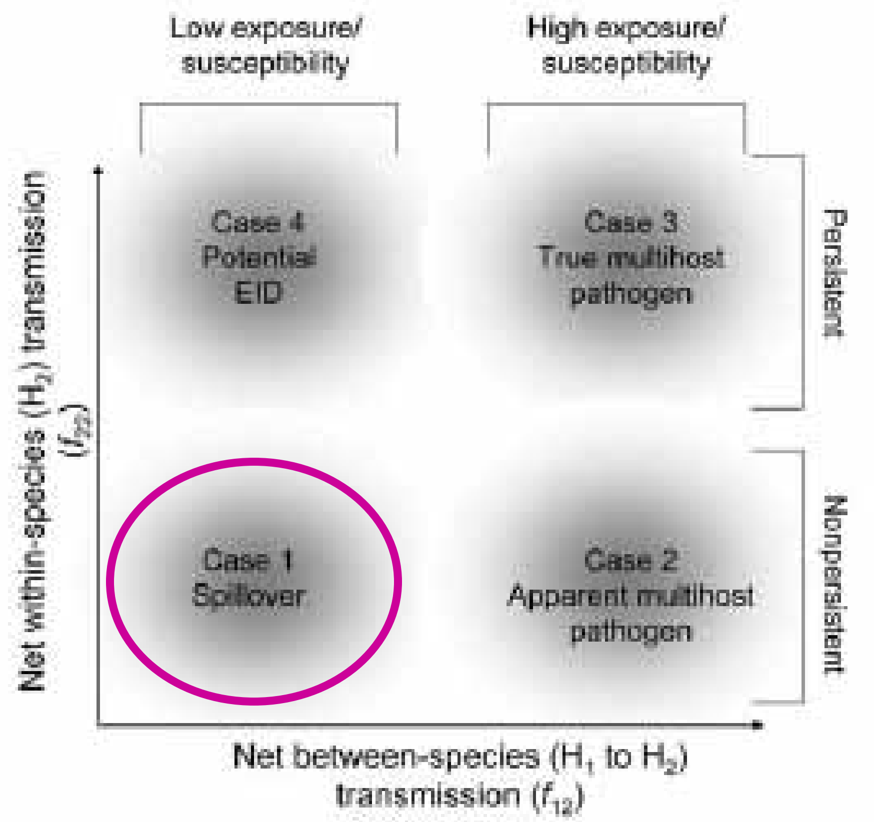


Figure 2. Community-epidemiology continuum, determined by the net between-H_1 and -H_2 transmission rate (f_{12}) and the net within-H_2 transmission rate (f_{22}). EID, emerging infectious disease.

- Raccoon rabies in cats in Eastern USA

Fenton & Pedersen, 2005

Classifying disease threats from Reservoir hosts (H_1) and Target hosts (H_2)

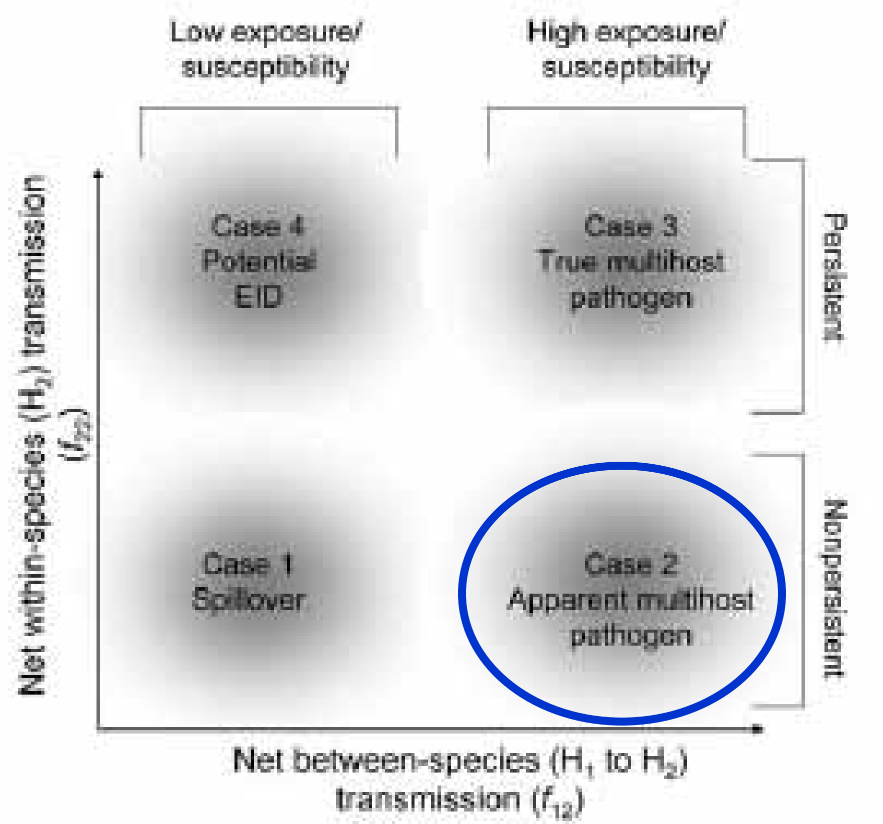


Figure 2. Community-epidemiology continuum, determined by the net between-H_1 and -H_2 transmission rate (f_{12}) and the net within-H_2 transmission rate (f_{22}). EID, emerging infectious disease.

Fenton & Pedersen, 2005

- Rabies in side-striped jackals
- not self sustaining due low density susceptible H_2 population,
- epidemics seeded from domestic dog reservoir H_1

Classifying disease threats from Reservoir hosts (H_1) and Target hosts (H_2)

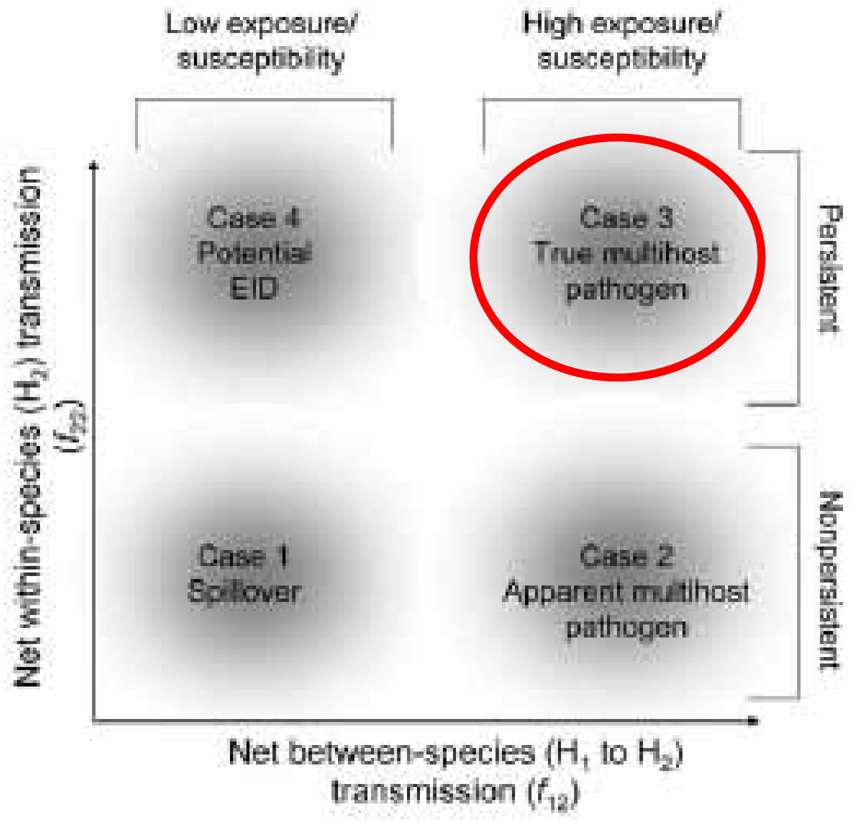
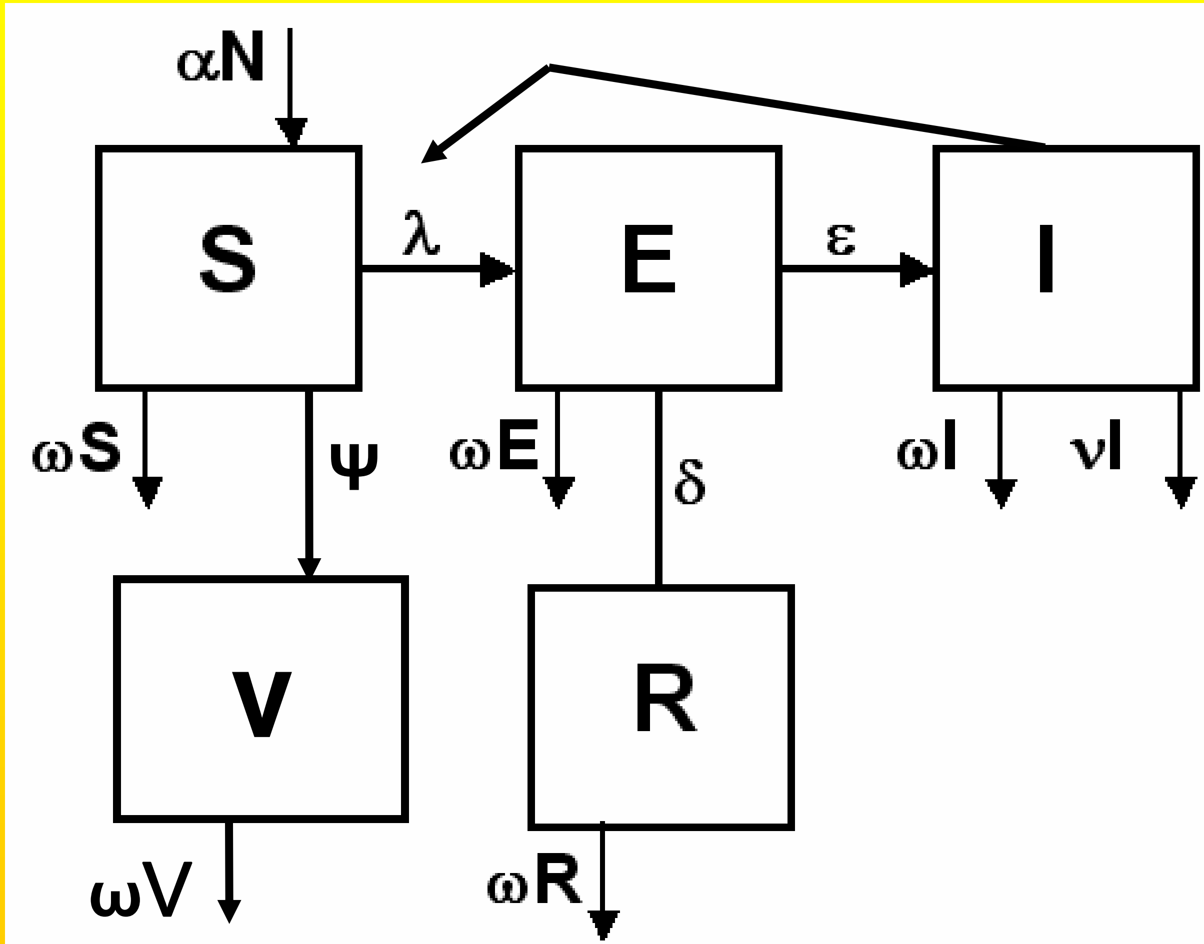


Figure 2. Community-epidemiology continuum, determined by the net between-H_1 and -H_2 transmission rate (f_{12}) and the net within-H_2 transmission rate (f_{22}). EID, emerging infectious disease.

- Self-sustaining in either host population
- Not canine rabies

Fenton & Pedersen, 2005

Rabies Dog Model



ά = birth rate

ω = general mortality

Ψ = vacc rate

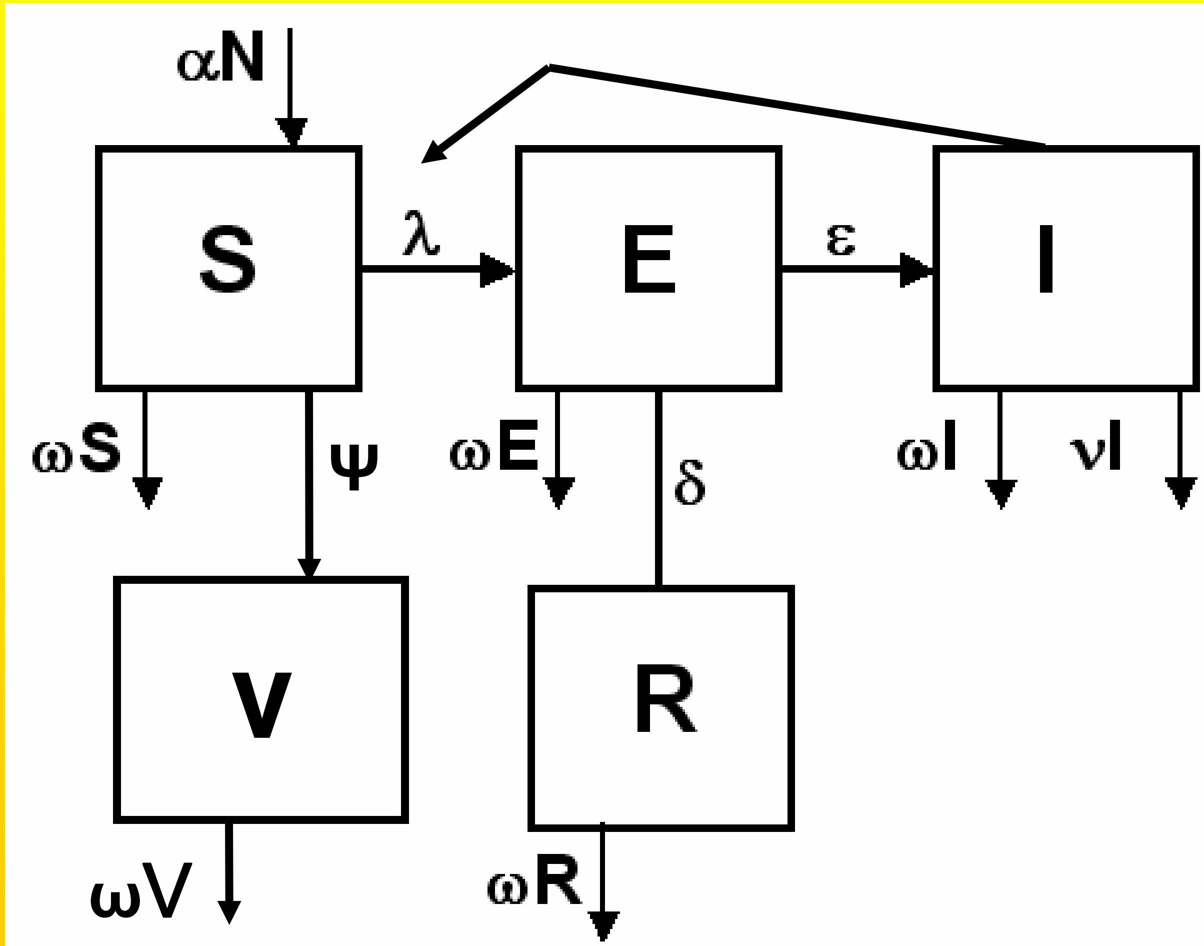
λ =force of infection

ε =1/IP

δ = recovery rate

v = rabies-specific mortality

Rabies Dog Model



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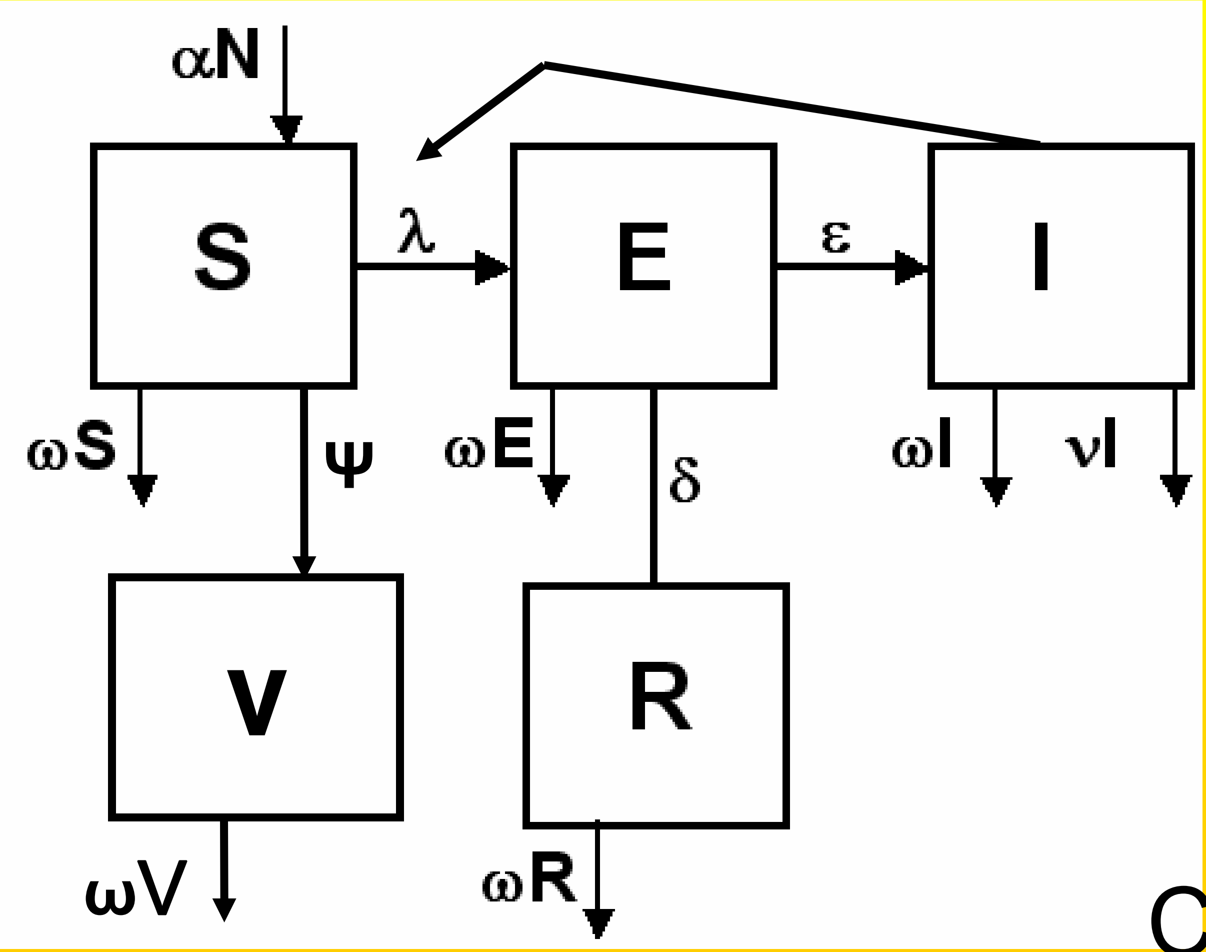
v = rabies-specific mortality

R ??

Rabies Dog Model



ά = birth rate

ω = general mortality

Ψ = vacc rate

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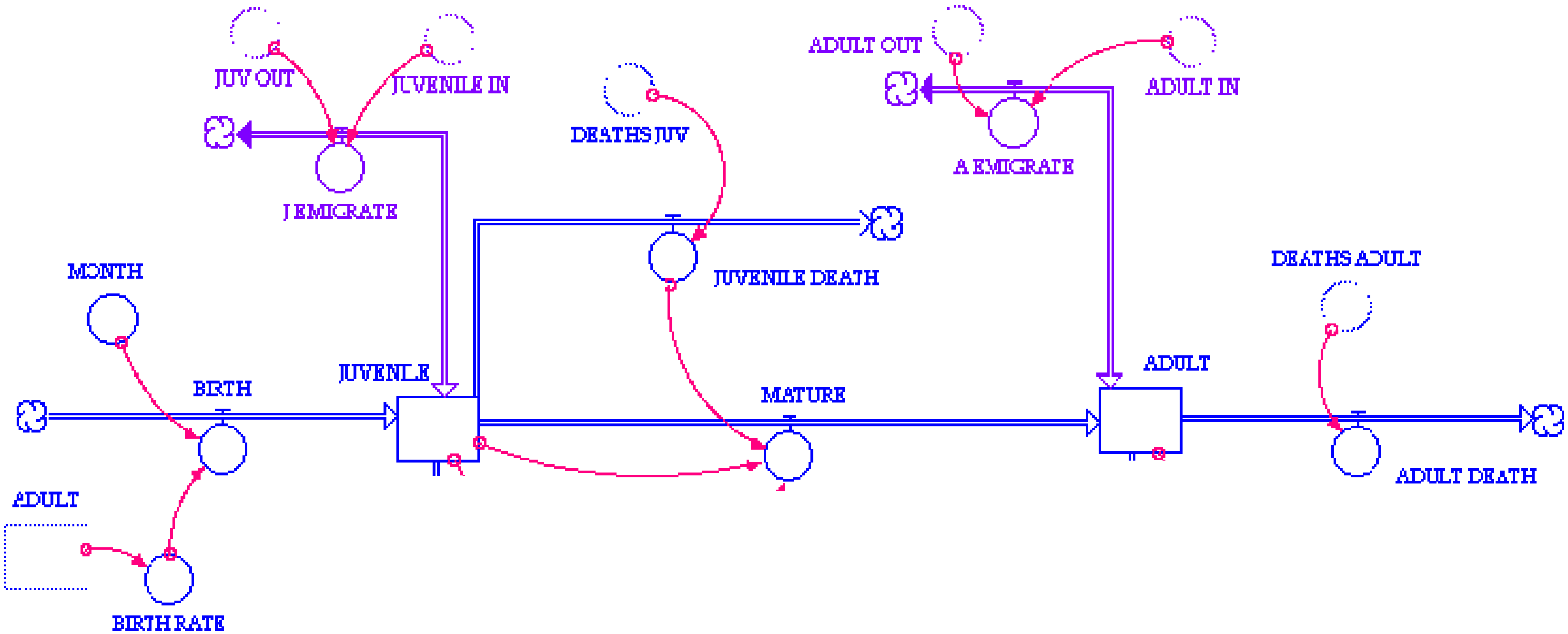
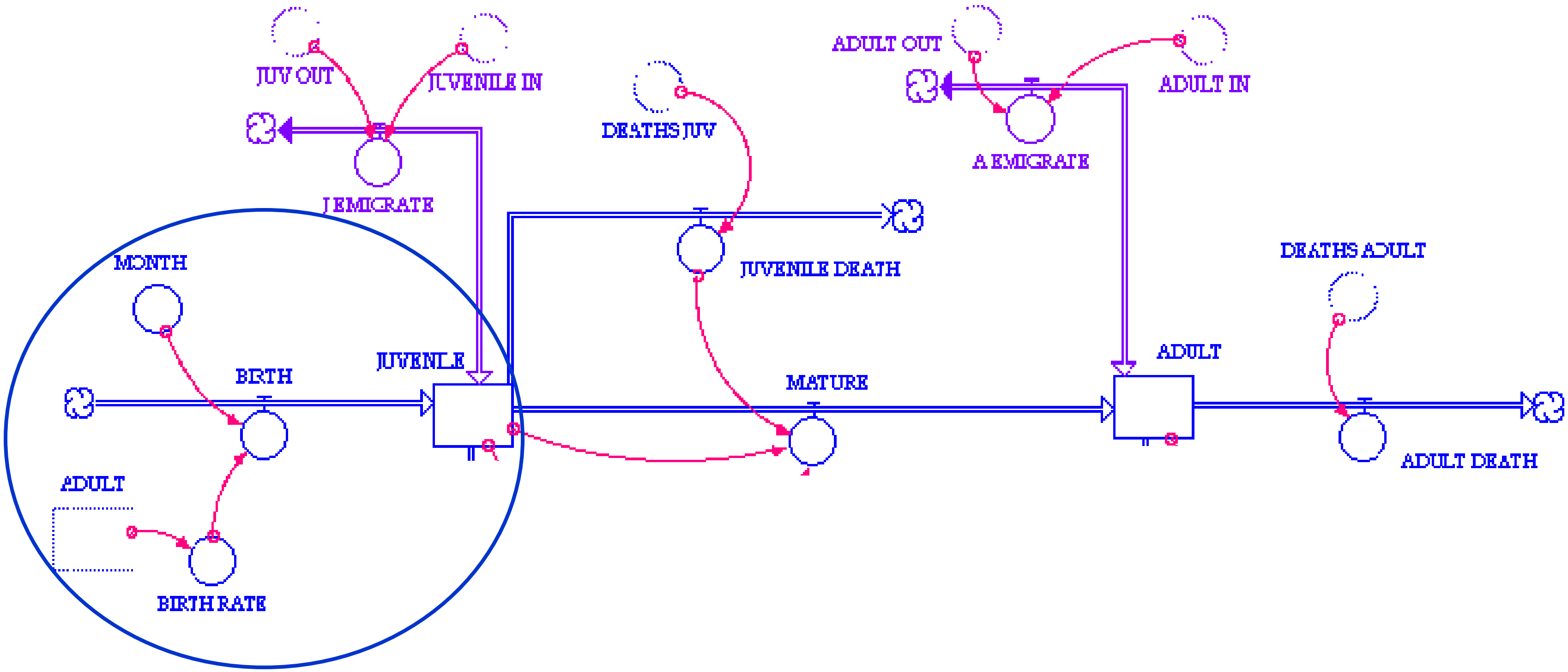
Pictorial !!
Communication

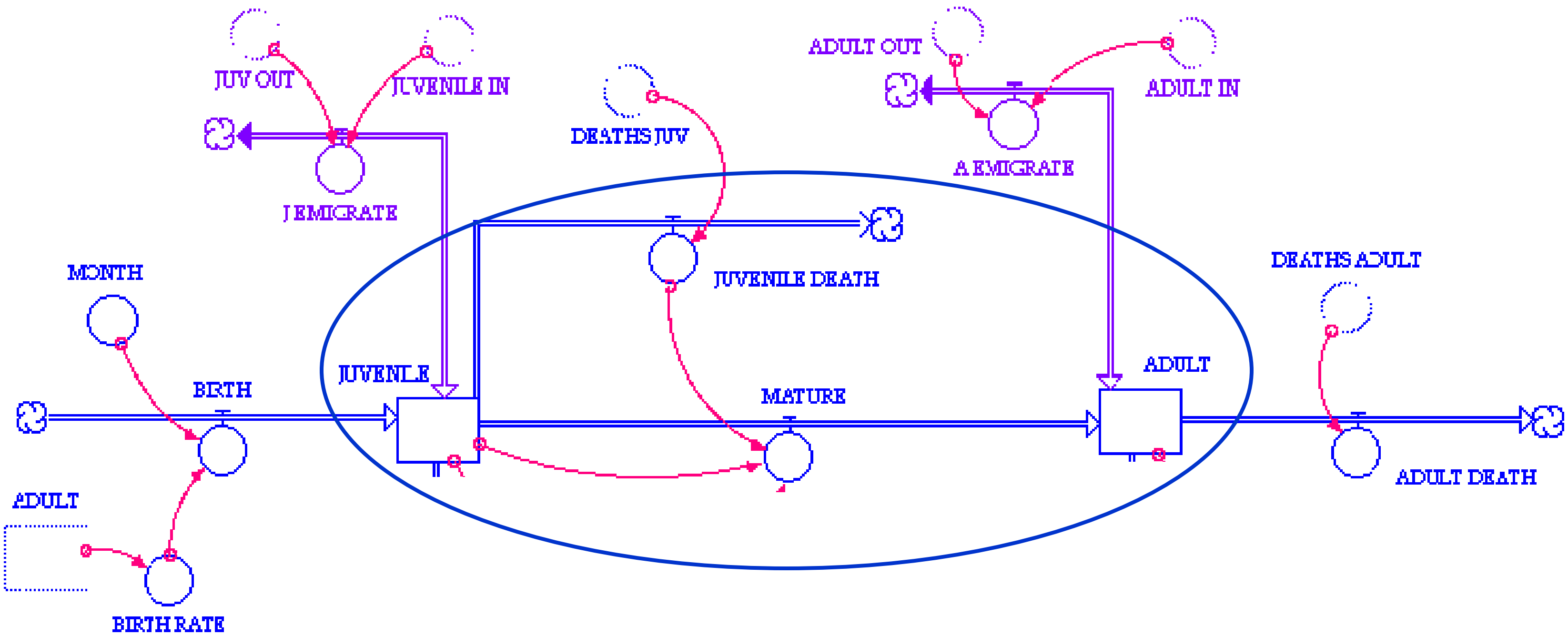
Dog rabies compartmental model



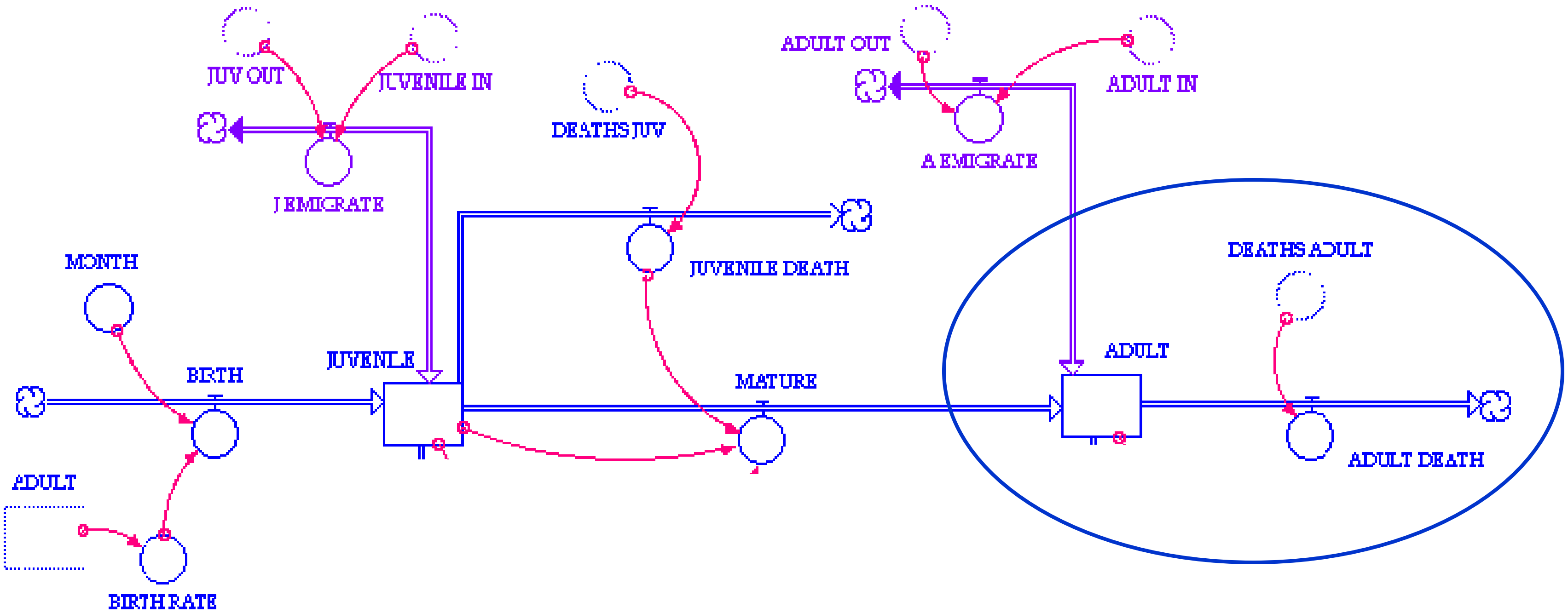
Dog rabies compartmental model



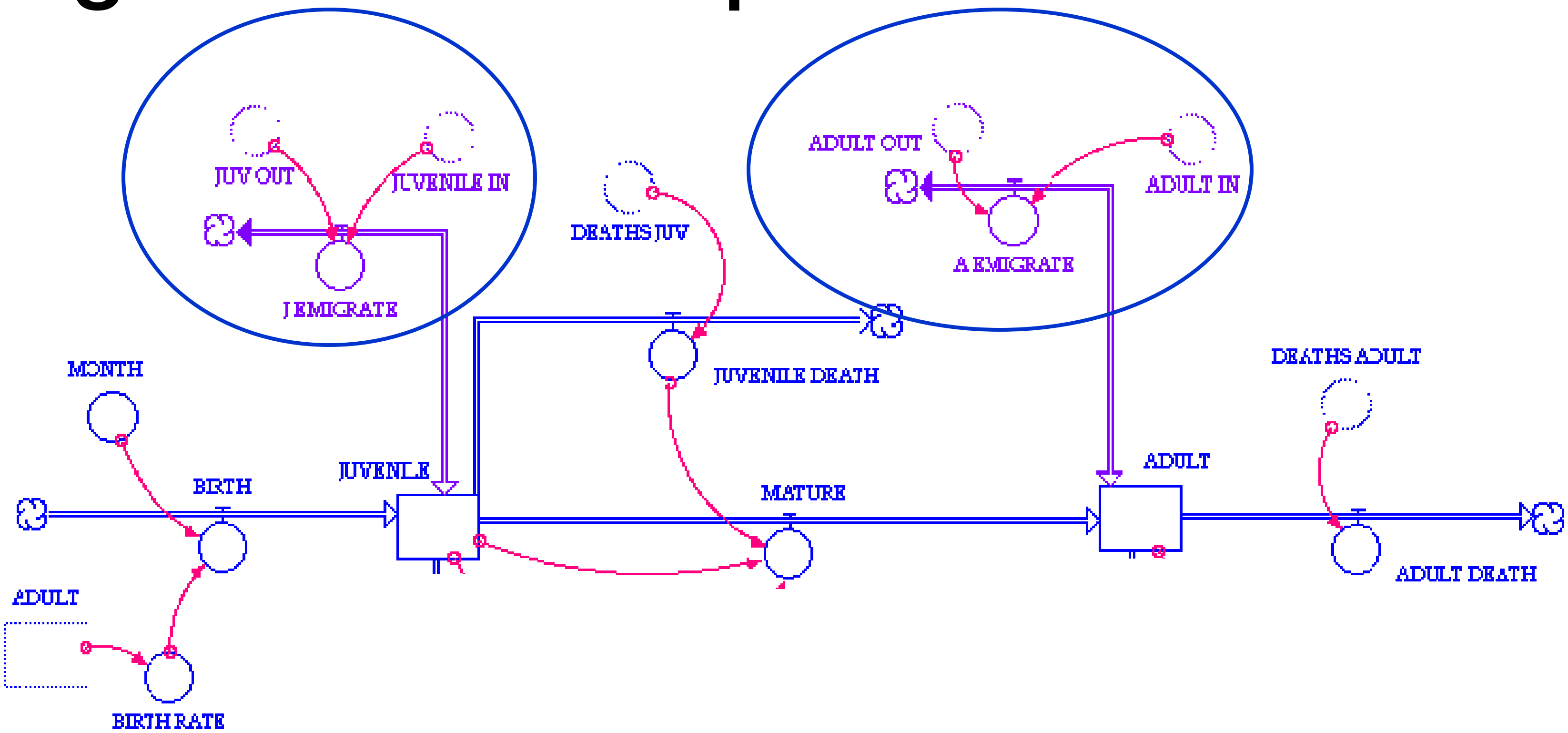
Dog rabies compartmental model



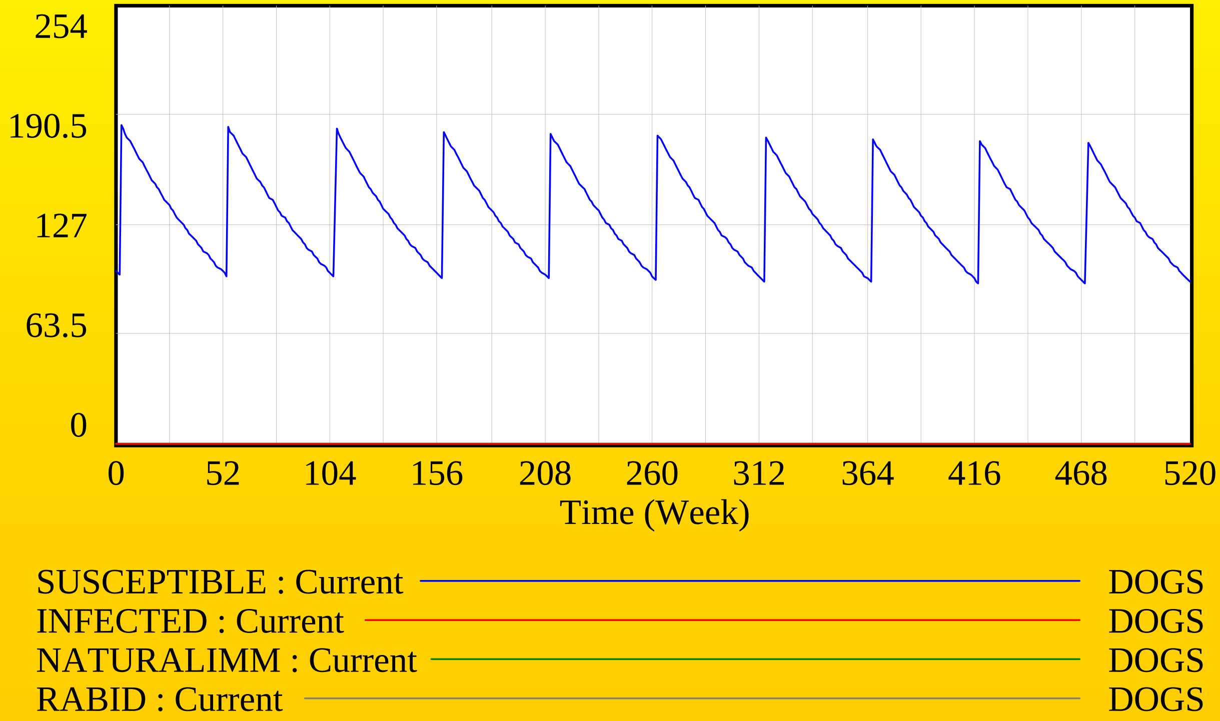
Dog rabies compartmental model



Dog rabies compartmental model



Graph showing population over 10 years

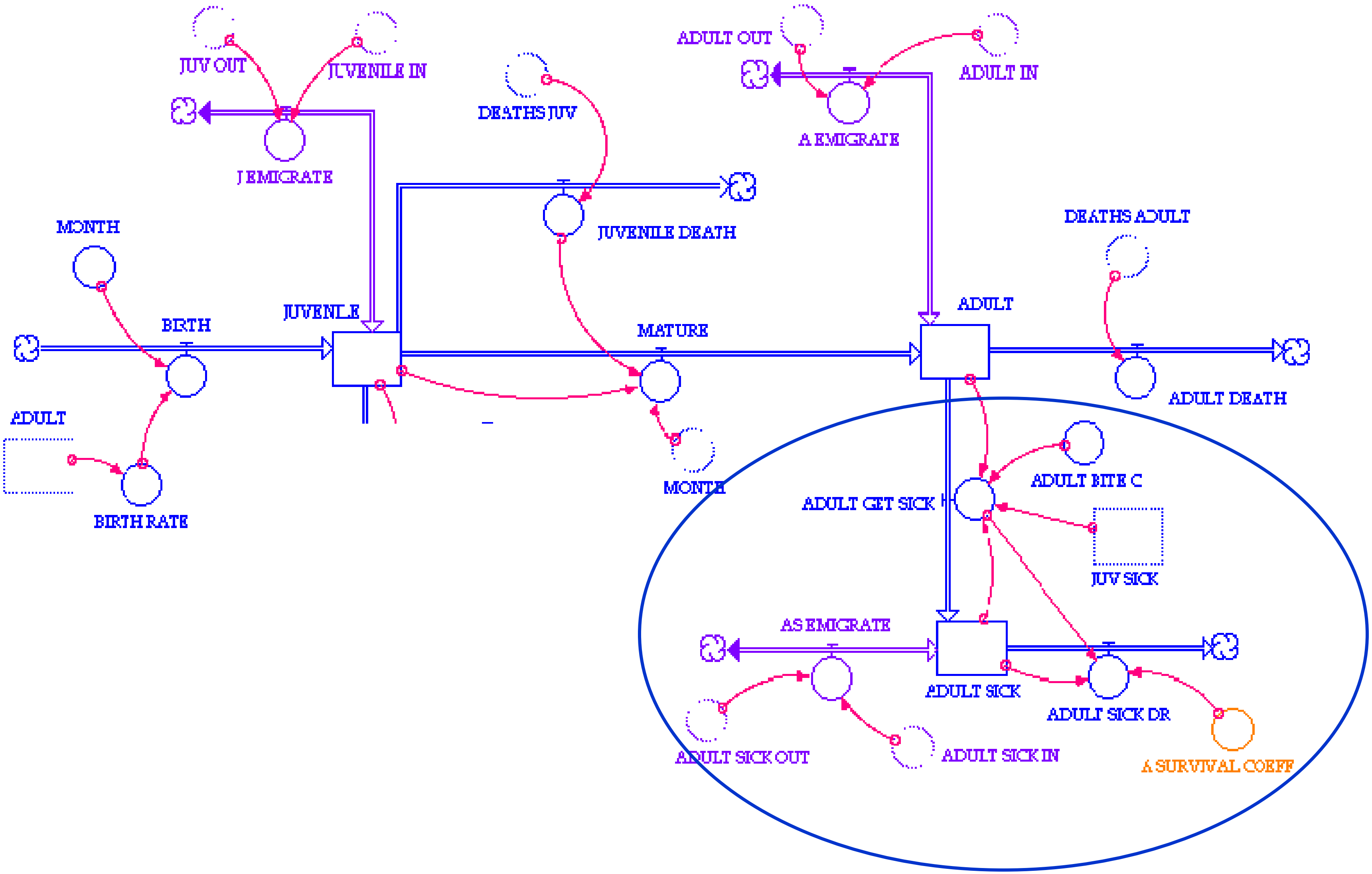


Vensim Graphs

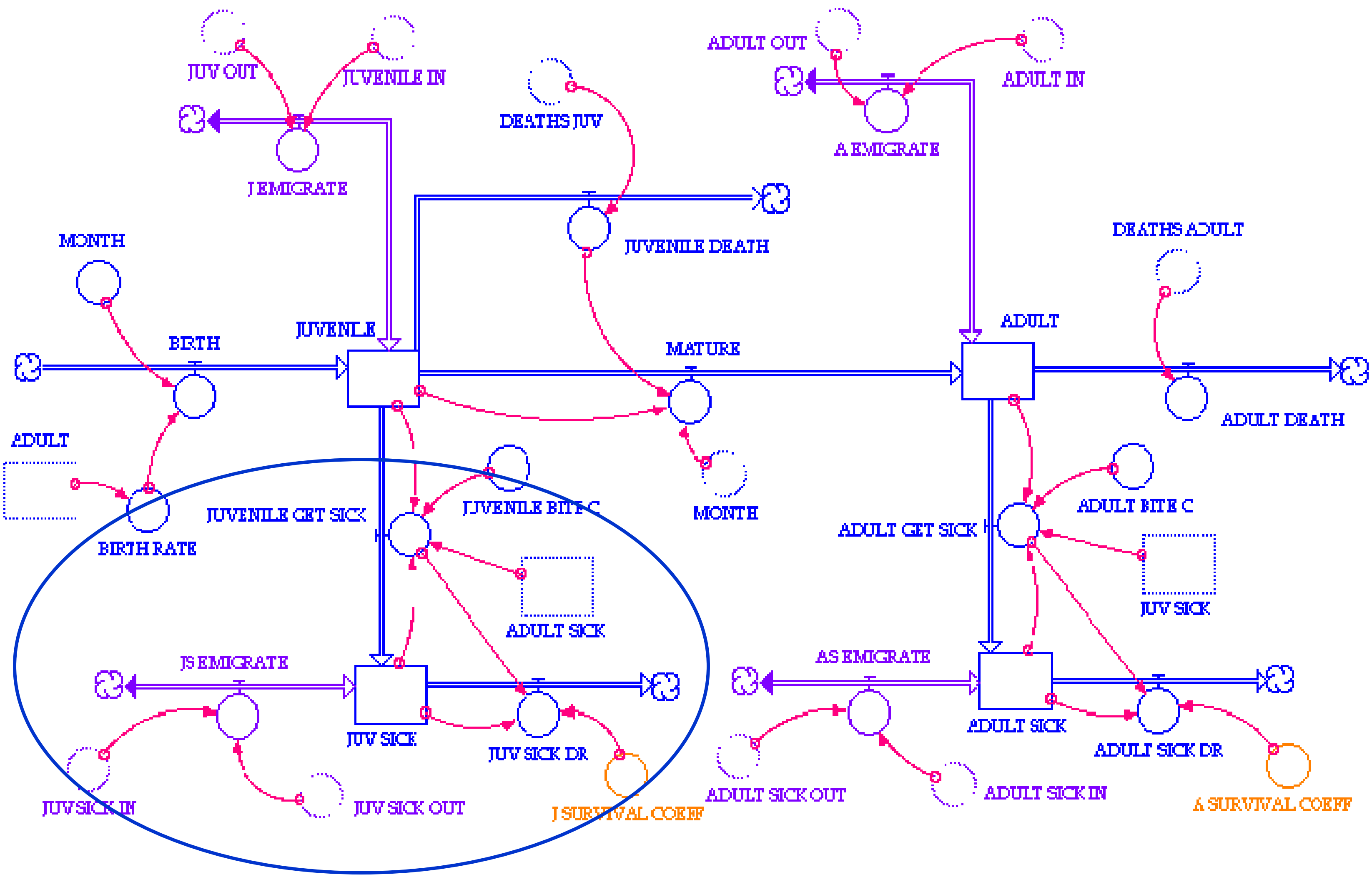
No disease

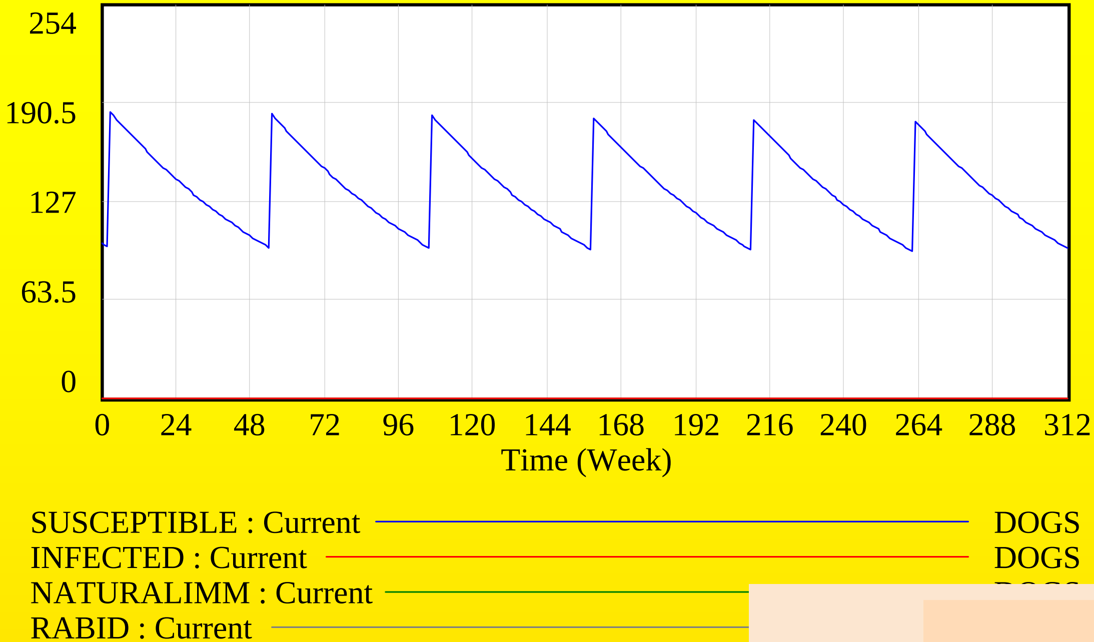
Stable Population

Dog rabies compartmental model



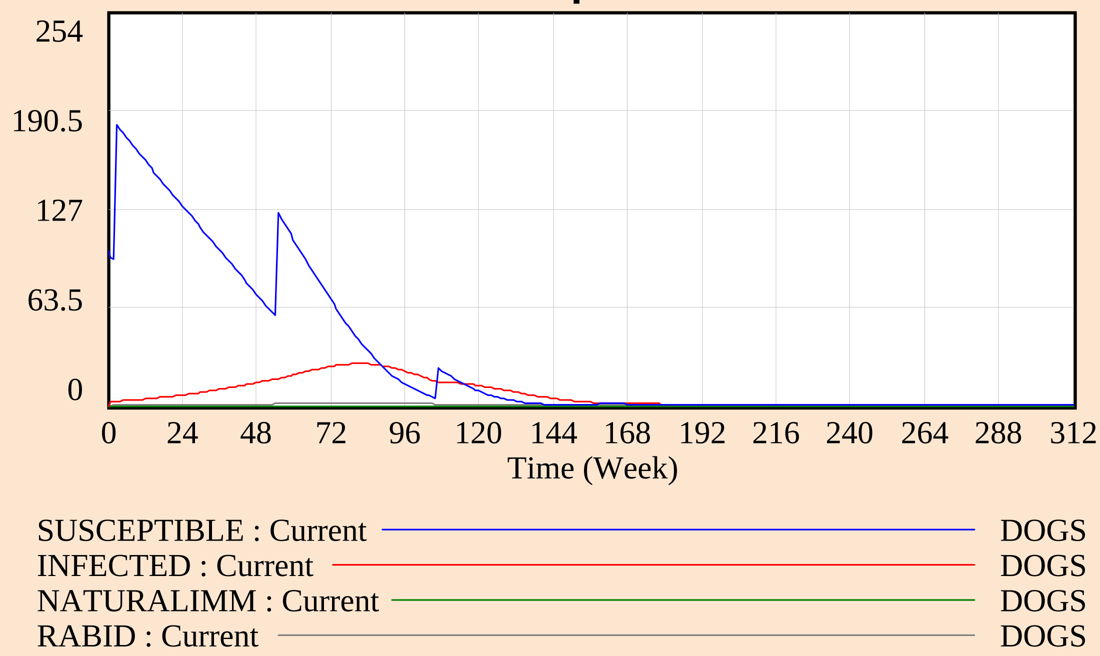
Dog rabies compartmental model



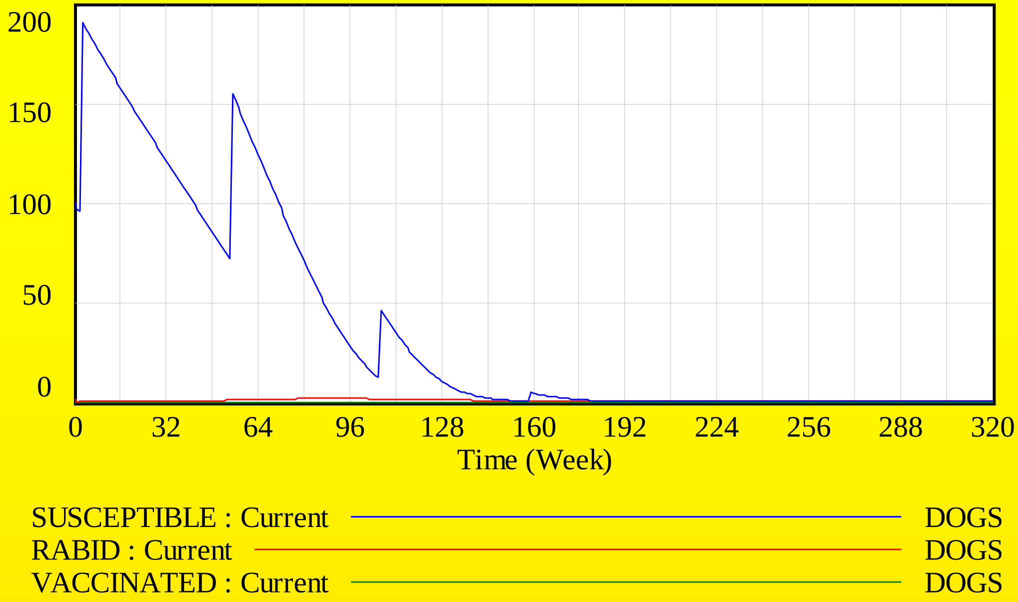


No Disease

Rabies present



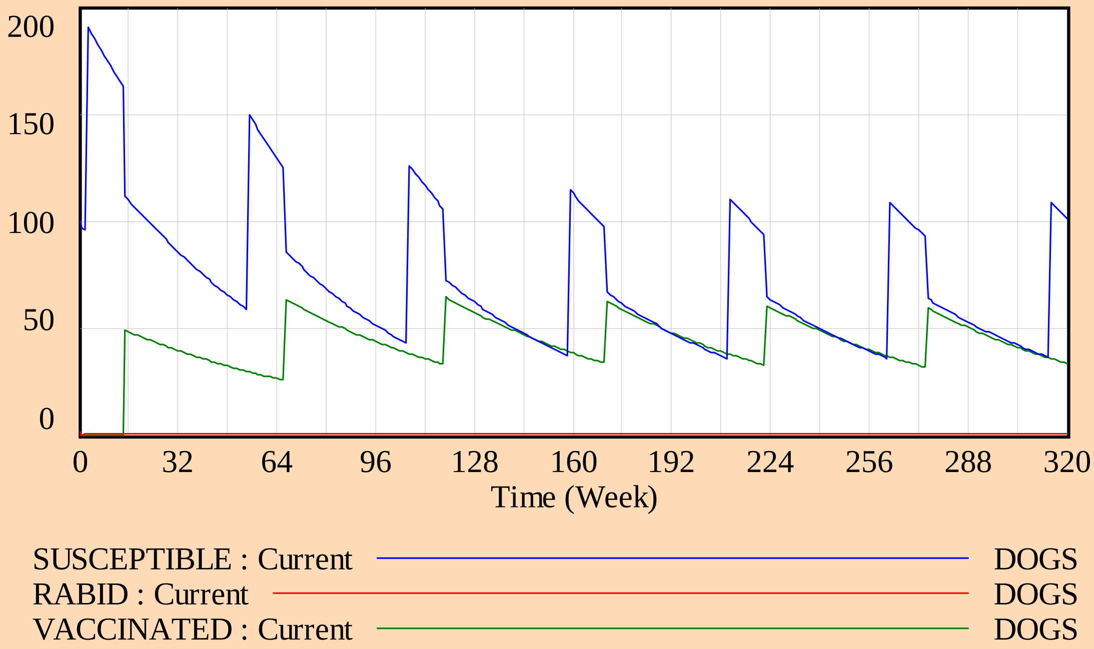
1 rabid dog
No vaccination
Other mortalities same
Birth rate not increased
Sex ratio pups same



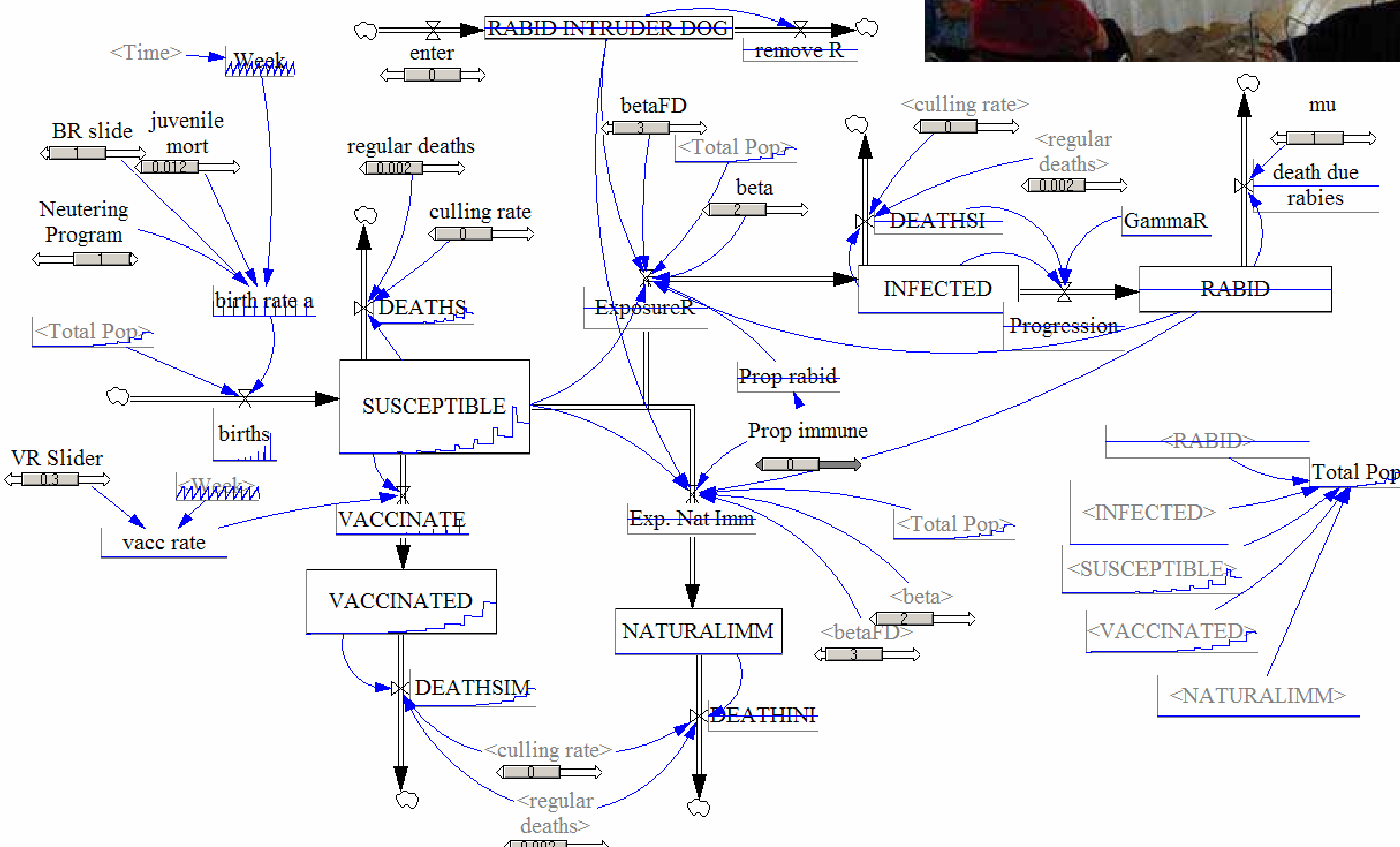
1 rabid dog
No vaccination

1 rabid dog

Vaccination rate 40%
Over limited time period
3 months post whelping
Other mortalities same



Vensim model developed by group for wild dog rabies



Advantages for Research/Policy

- Include field workers with valuable information over many years - but not published
- Inclusive, quick & transparent method investigating disease ecology & devising control schemes
- See which parameters make the most difference when changed, if sensitive then further investigations & potential as control point

Advantages of Pictorial Mathematical Models

- Can involve many stakeholders
- Different levels literacy & (dis)comfort with equations
- Pictorial interface aids participation
- Effective way for group/community to see what effects of different control methods
- Community participation essential to success

Pictorial Models are Useful !

How to proceed ?

Difference equation model in R

- Vensim Model
  - same mathematics
  - engaging pictorial interface
  - slider bars
- Developing model “at the table”
- Start simple
- Model effects specific interventions

Molecular epidemiology of rabies in KwaZulu Natal, South Africa

Peter Coetzee

University of Pretoria

Molecular epidemiology

- **Molecular epidemiology: S**cientific field of study in which molecular markers (unique patterns of nucleotide or amino acid substitutions) are used to track the diversity, and routes of dissemination of a pathogen within and between susceptible host populations.
- **Aim:** Identifying factors which influence the incidence, occurrence and spread of a disease

Molecular epidemiology - Basic principles

- **Mutations** – random errors caused during genome replication.
- Viral mutation up till a million times higher than mutations in Eukaryotes
- **Reason:** RNA-dependant RNA polymerases lacks proofreading function.
- Viral population - quasispecies.
- Genetic drift, neutral mutation - evolution of geographic variants.

Molecular epidemiology - Basic principles

- **Genetic typing techniques**

- **<u>Molecular sequence data</u>**

– Quantitative measurement of relatedness between viral variants

– Construction of phylogenetic trees which allow for the visualization of evolutionary relationships between taxa (phylogenetic characterization).

Molecular epidemiology – Methodology

- Essential to select correct region of genome for characterization
- Rates of mutation vary from gene to gene – selective pressures can either constrain or promote evolution
- Ideal target: region with maximum genetic information flanked by two regions of low variation - primer sites

Molecular epidemiology – Methodology

- RT-PCR, automated fluorescent nucleotide sequencing, multiple alignment, phylogenetic analysis.
- Distance based methods - NJ
- Sequence based methods – MP, ML
- Publications: NJ – simplest, fastest, most commonly used, genetic distances indicated on tree

Molecular epidemiology – Methodology

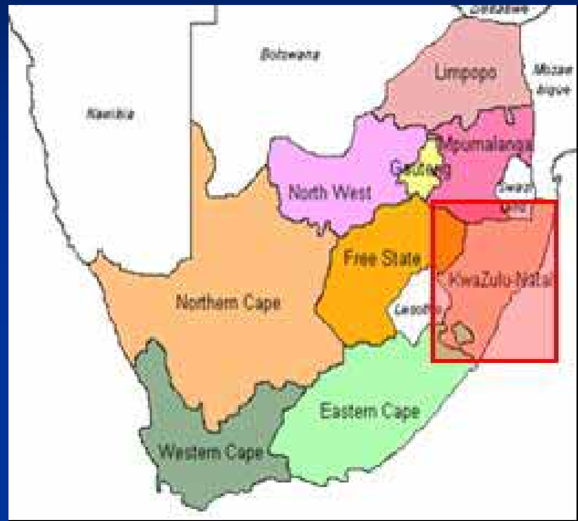
- Multiple trees with varying topology may be reconstructed from a given dataset – essential to test robustness of reconstructed tree topologies.
- Bootstrap tests:

1. Multiple random re-sampling of nucleotides from multiple sequence alignment.
2. New tree constructed from every pseudo-replicate dataset.
3. Number of times a specific grouping is reconstructed from total number of datasets – statistical support for clustering of isolates in a specific monophyletic group

- 80-100% - strong support for grouping of isolates
- Below 70% - generally considered non-significant

KwaZulu Natal

- South Africa's most populous province.
- 54 independently governed magisterial districts.
- Borders Swaziland and Mozambique in the north, Eastern Cape in the south.
- Bound inland by Lesotho, the Freestate province and southeastern Mpumalanga
- Agricultural, industrial and recreational hubs of province occur primarily along the coast – connected by N2 highway.
- Hubs typically surrounded by informal settlements



Rabies in KwaZulu Natal – Historical perspectives

- Dog rabies introduced from Angola in 1947
- Spread throughout African subcontinent in susceptible domestic and wild canid populations.
- Angola (1947) – Botswana – Northern Limpopo province, South Africa (1950)- Zimbabwe (1950) – Maputo, Mozambique (1952)
- Two epidemic initiated in northern KwaZulu Natal:

1. 1964 – 1968: brought under control through vaccination and control of dog movement .
2. 1976 – 2006: initiated by refugees fleeing political unrest in Mozambique, remains intractable

Rabies in KwaZulu Natal – Epidemiological perspectives

- Epidemic in KwaZulu Natal - majority of human and animal cases in South Africa per annum.
- Majority of human cases – children.
- Domestic dog - principal disseminator and transmission host.
- Apparently no involvement of wildlife in the epidemiology of the disease.
- Highest number of cases reported from coastal regions - Durban, Richards Bay and Port Shepstone (highest population density).
- Fewer cases reported from internal regions.

Rabies in KwaZulu Natal – epidemiological perspectives

- Reasons for the persistence of the second epidemic. .

1. Inability to control dog movement.
2. Inadequate vaccination coverage.
3. AIDS – feral dogs.

- Hotspot vaccination and euthanasia campaigns (2003) - effective in reducing number of rabies cases in target regions

Problem statement

- Little known about the molecular epidemiology of canid-borne rabies in KwaZulu Natal.
- Project aimed to contribute to the currently available knowledge base by using phylogenetic methods to characterize viral isolates from the province.

Materials and methods

- Number of cases (2003): 409 animal, 11 human
- Rabies cases not evenly distributed – localized in densely populated coastal regions.
- Representative sampling – attempted to sequence at least 50% of submitted isolates from each magisterial district from which rabies was reported from during the study year
- Pseudogene and glycoprotein cytoplasmic domain targeted – highly variable, suitable for distinguishing between closely related variants
- 123 animal and 5 human isolates included in final dataset.

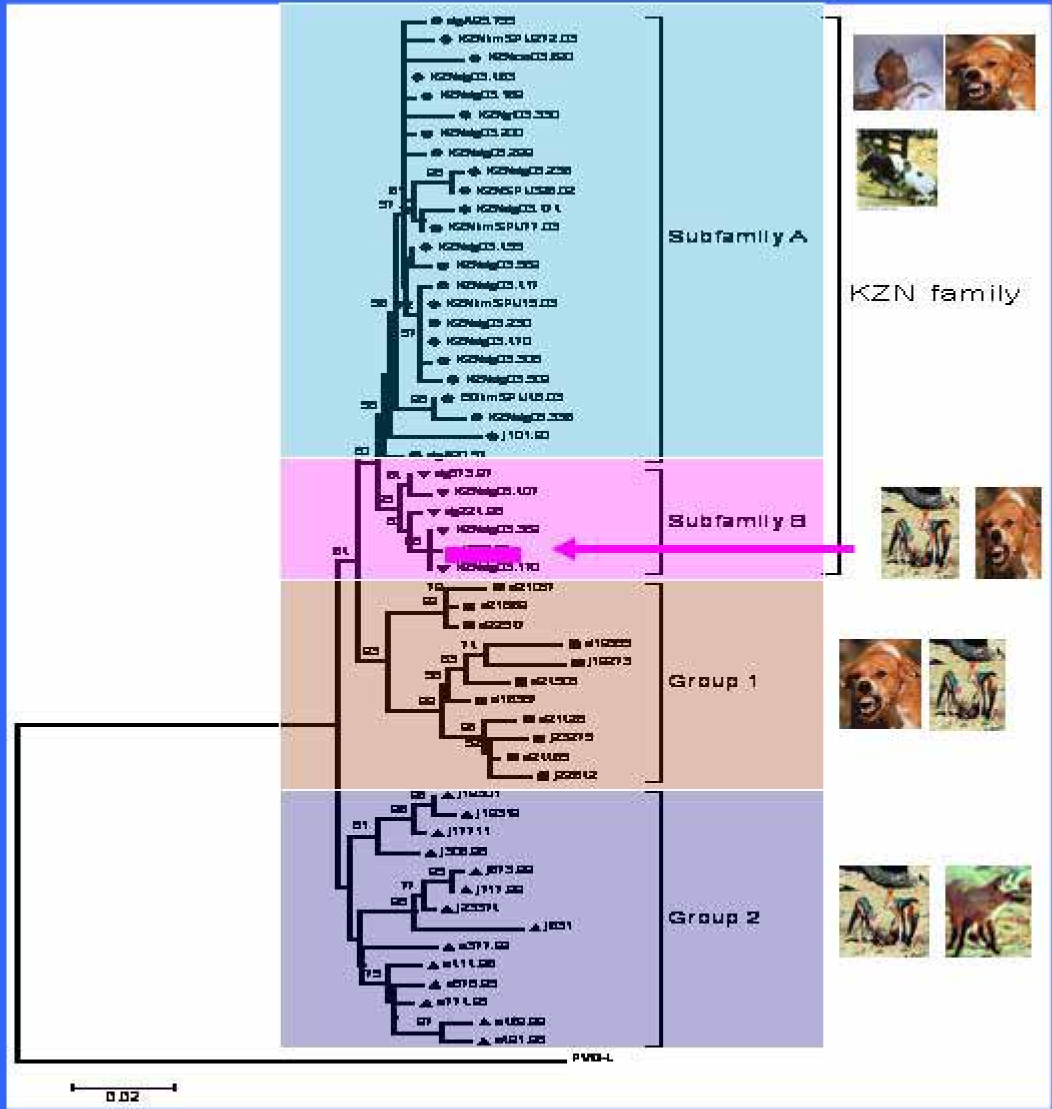
Analysis strategy

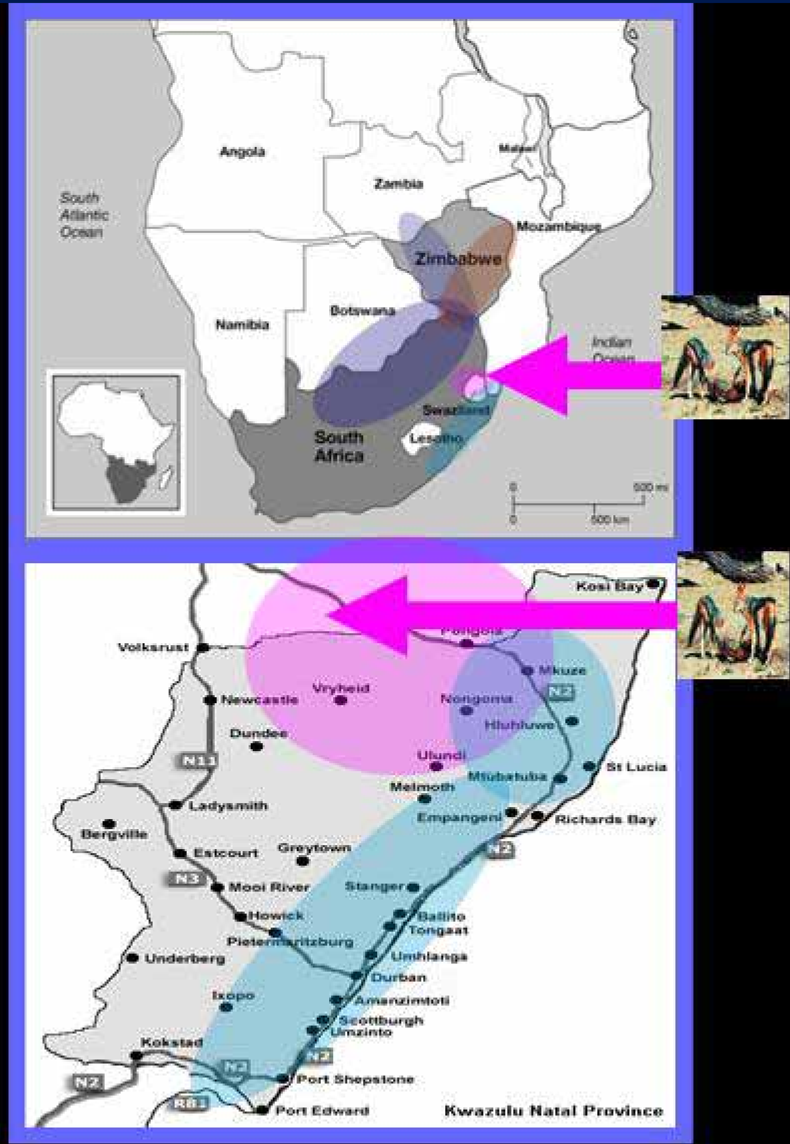
- Two phylogenetic trees constructed in order to address the different aspects of the epidemic in KwaZulu Natal.

- **First tree – KZN, Zimbabwe, South Africa**
- Regional perspective

- **Second tree** – KZN isolates only
- Clarification of the relationships which exist specifically between isolates from the different magisterial districts of the province.

Phylogenetic analysis of canine viral isolates from southern Africa – Origin of epidemic and involvement of wildlife





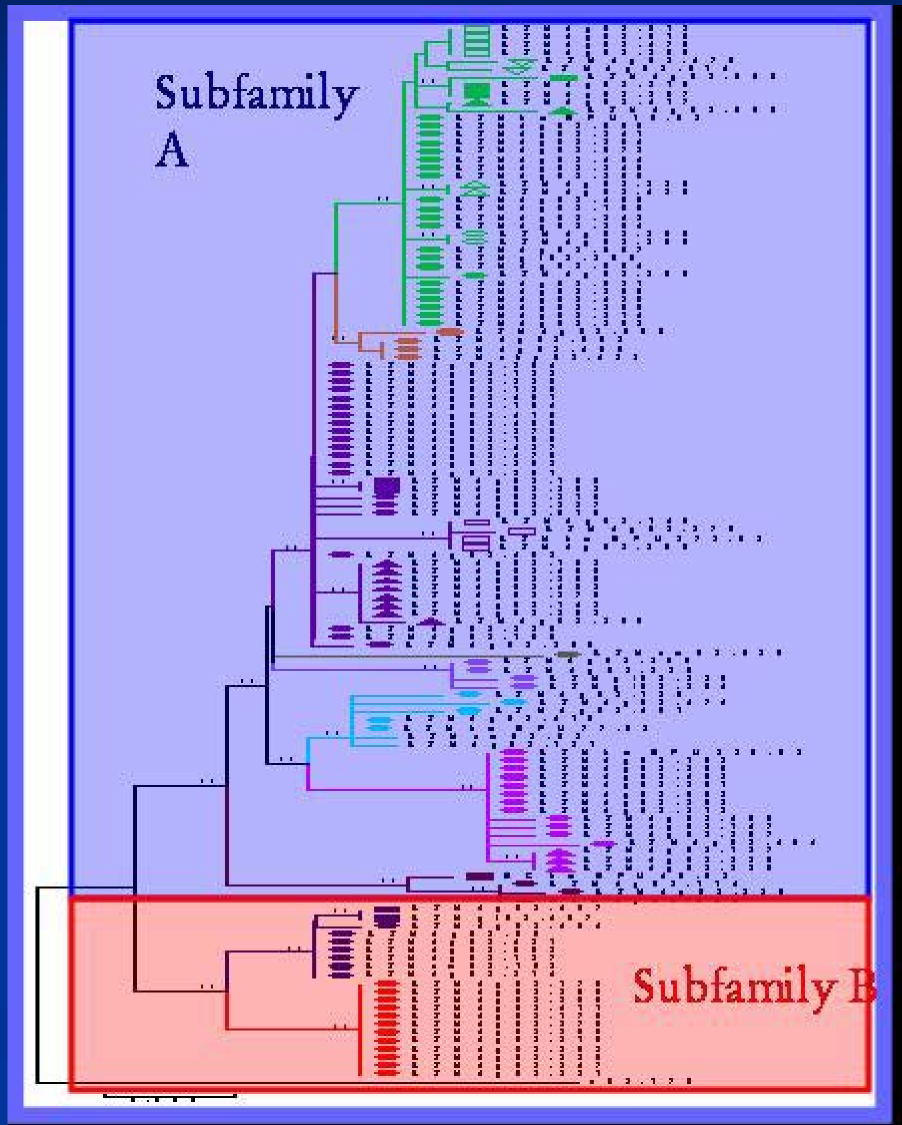
Phylogenetic analysis of viral isolates from KwaZulu Natal – identification of regional variants

- Viral isolates from KwaZulu Natal divisible into two viral groups (Subfamily A and B).

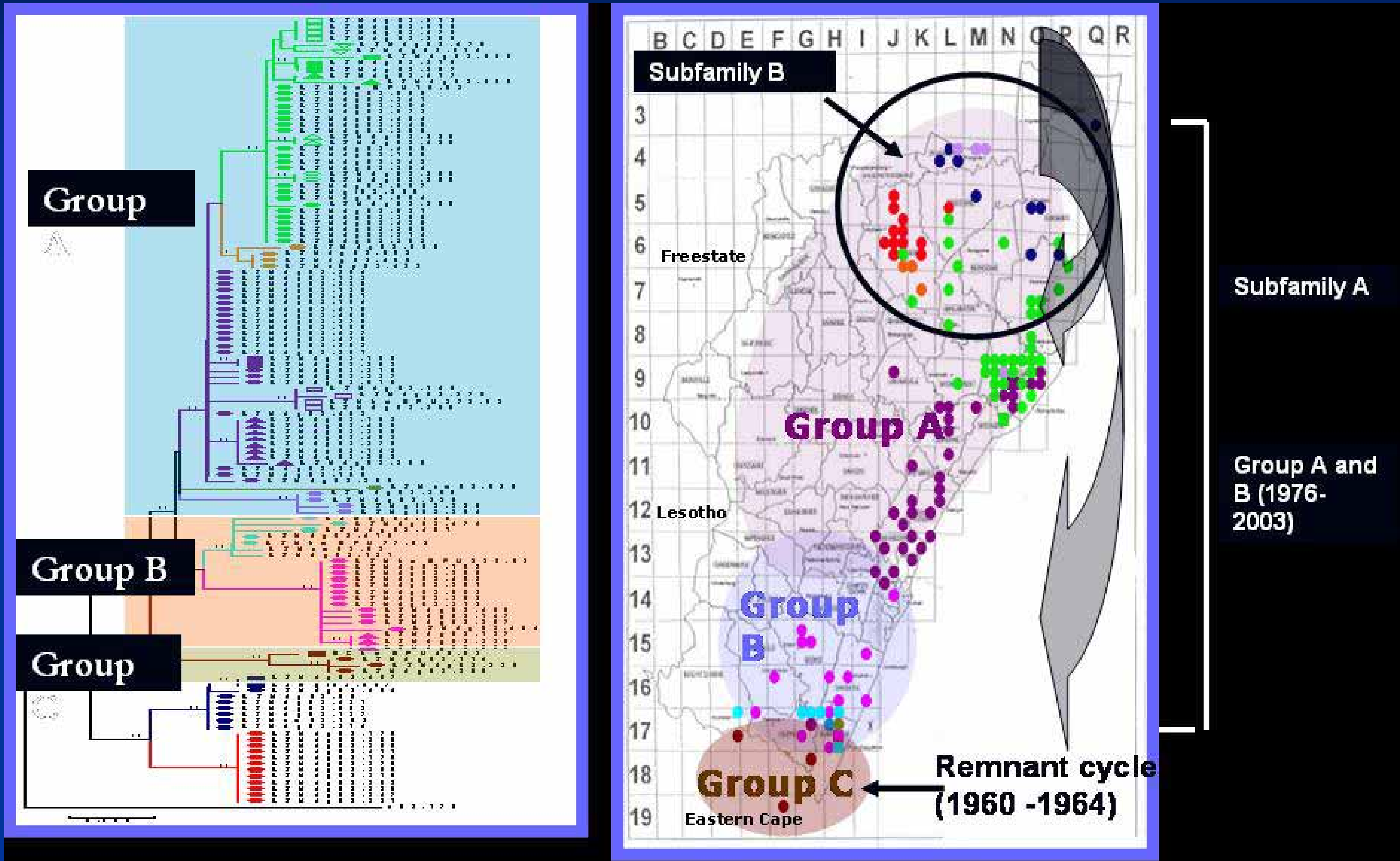
1. Subfamily A – core of current epidemic among dogs
2. Subfamily B – putative dog-jackal cycles

- Grouping of viral isolates into clusters (clades) based on geographic region of isolation.

- Subfamily A (cluster 1A-8A), Subfamily B (cluster 1B-2B)
- Phylogenetic characterization of regional topotypes –future surveillance



Phylogenetic analysis of viral isolates from KwaZulu Natal – Investigation of the origins of current epidemic



Reconstruction of human case histories from the province (2002-2003)

- **Five human cases from KwaZulu Natal analyzed**
- **Cases among children, terminal patients - complicates reconstruction of human case histories - contact routes, control measures**
- **Database of regional variants – useful for comparative studies.**
- **Human isolates without exception clustered with variants in regions these exposures presumably occurred in.**

What's new ?

- Characterization of geographic variants – sequence database for future comparisons
- Clear definition of the emergence of epidemic in KwaZulu Natal.
- Possible involvement of wildlife

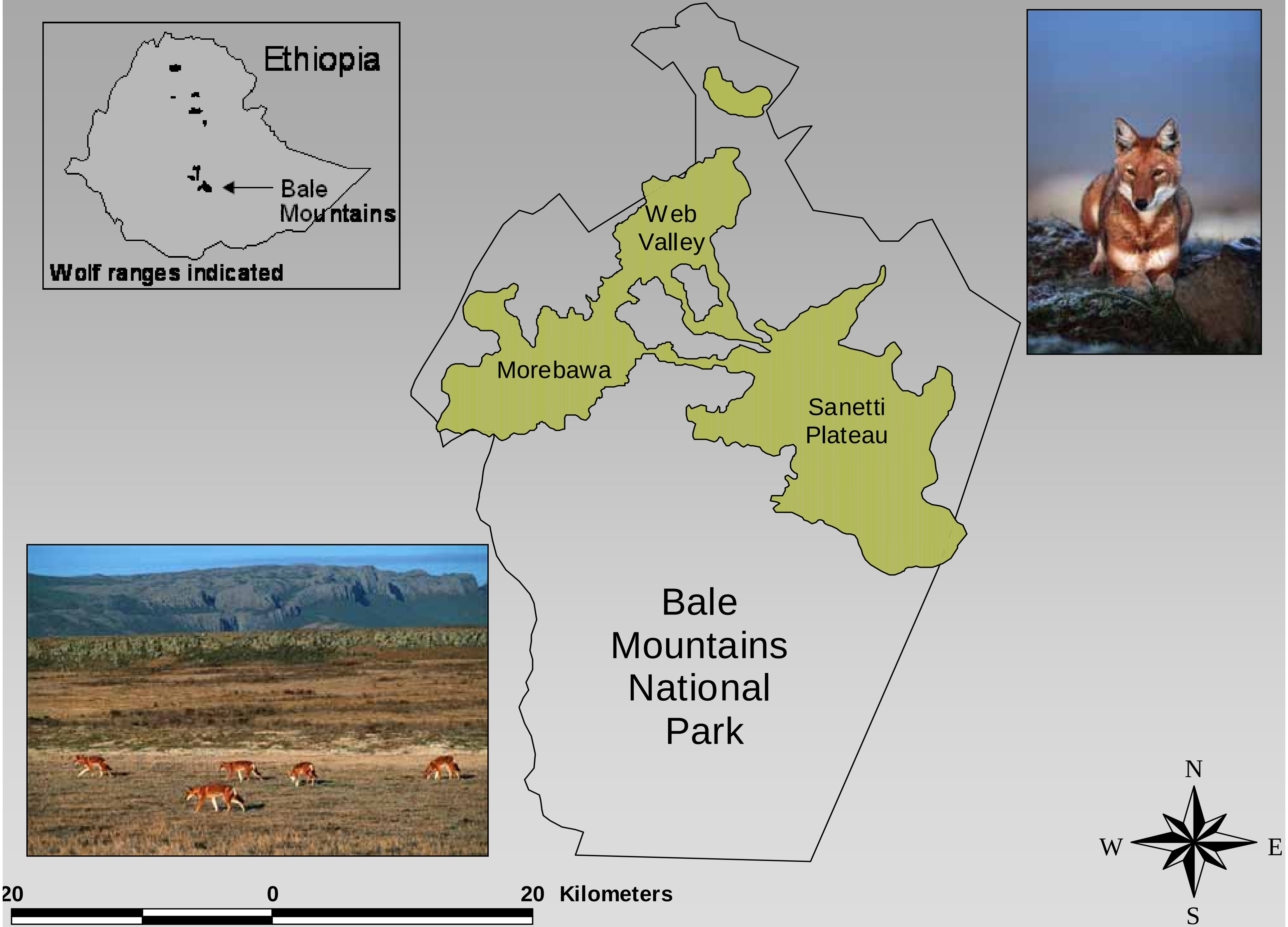
- Co-authors: Prof L. H Nel (UP), Mrs. W. Markotter (UP), Dr. J. Randles (Allerton Regional Veterinary Laboratory)
- Acknowledgement: Dr. Claude Sabetha (OP,), Dr. Felicity Burt (NICD), Nobantu, Jackie, Nicolette, Liz (UP)

Thank you !!!

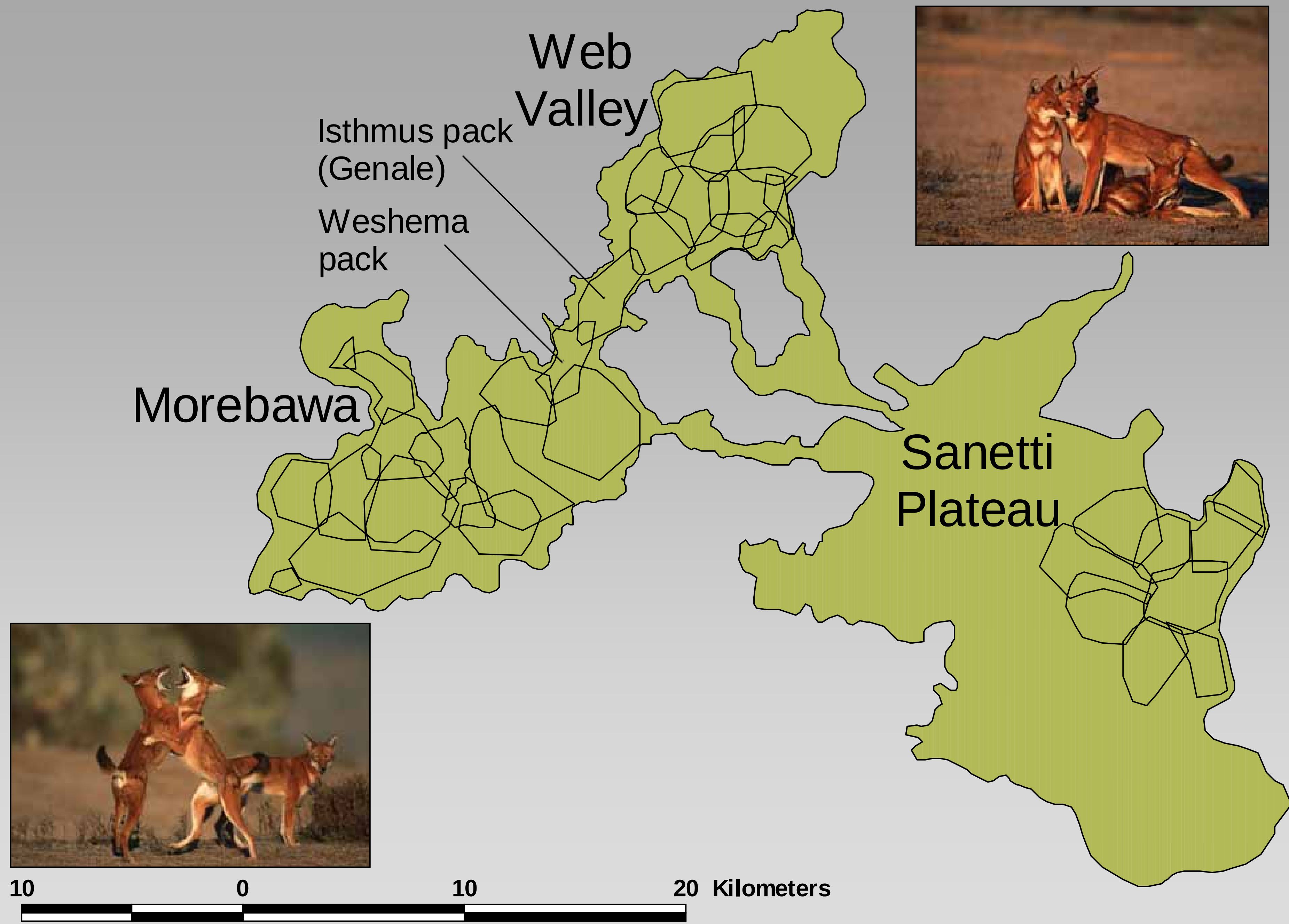
Management of a rabies epidemic in Ethiopian wolves in Bale Mountains National Park 2003/2004

Deborah Randall[2,5] Dan Haydon[3] Darryn Knobel[1,2] Anthony Fooks[4] Stuart Williams[2,5] Louise Matthews[1] Lucy Tallents[2,5] Kifle Argaw[6] Fekadu Shiferaw[6] and Karen Laurenson[1,2,7]

1. Centre for Tropical Veterinary Medicine, University of Edinburgh, Edinburgh, United Kingdom
2. Ethiopian Wolf Conservation Programme, Addis Ababa, Ethiopia
3. Division of Environmental and Evolutionary Biology, University of Glasgow
4. Veterinary Laboratories Agency, Weybridge, United Kingdom
5. Wildlife Conservation Research Unit, University of Oxford, Oxford, United Kingdom
6. Ethiopian Wildlife Conservation Organisation, Addis Ababa, Ethiopia
7. Frankfurt Zoological Society, Arusha, Tanzania

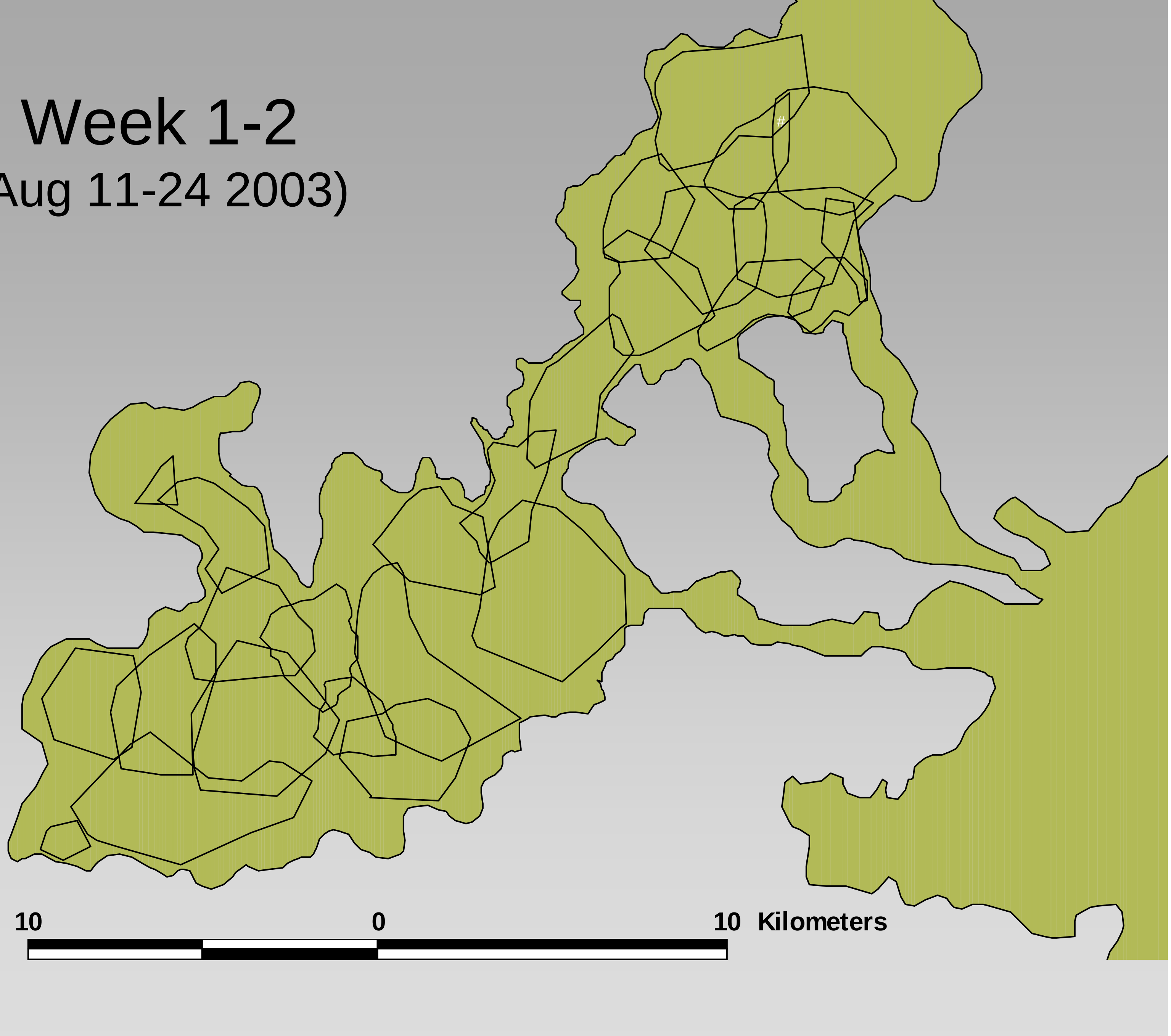


Ethiopia
Bale Mountains
Wolf ranges indicated
Web Valley
Morebawa
Sanetti Plateau
Bale Mountains National Park
N
W
E
S
20
0
20 Kilometers


Web
Valley
Isthmus pack
(Genale)
Weshema
pack
Morebawa
Sanetti
Plateau
10
0
10
20 Kilometers

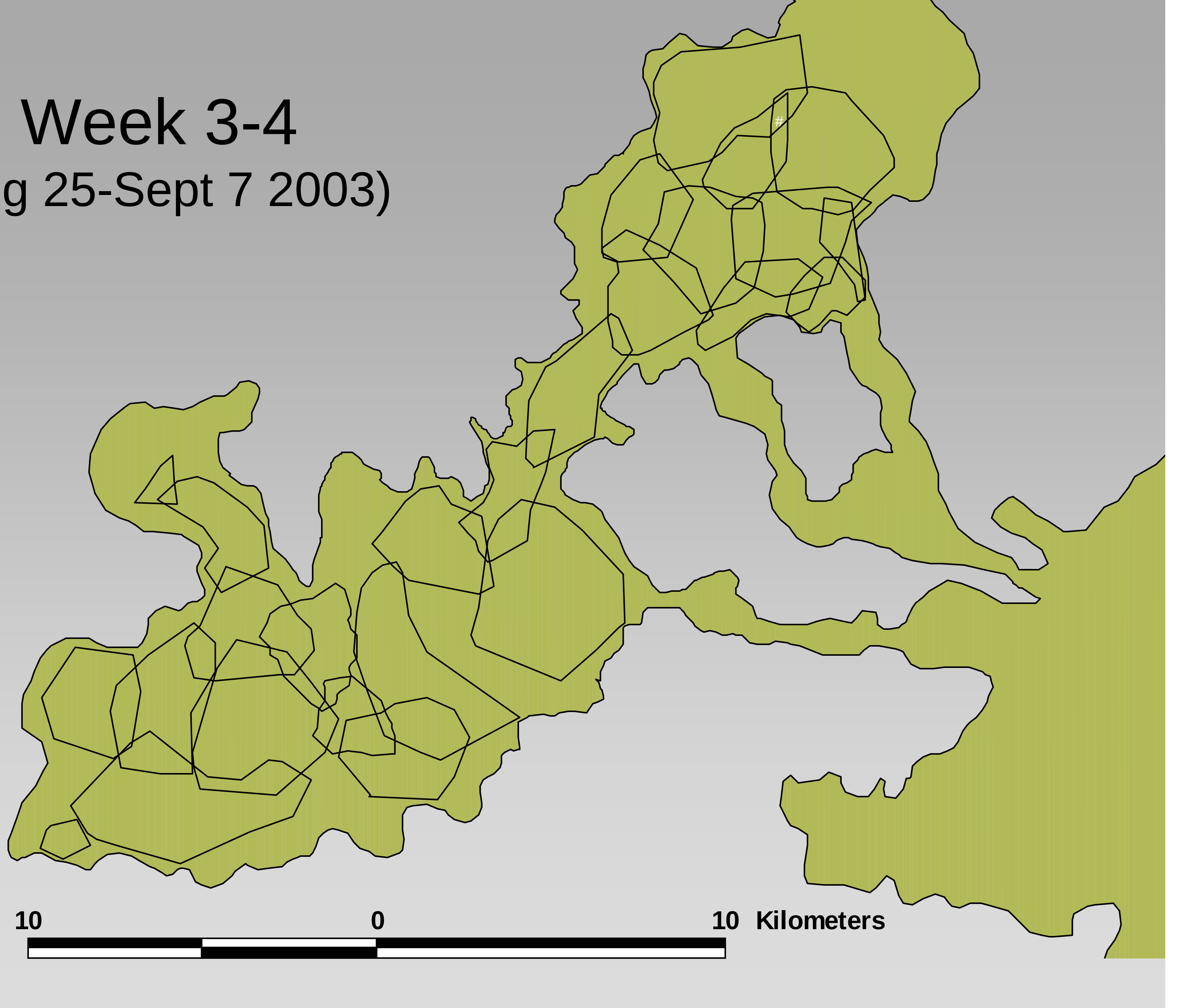
Week 1-2

(Aug 11-24 2003)



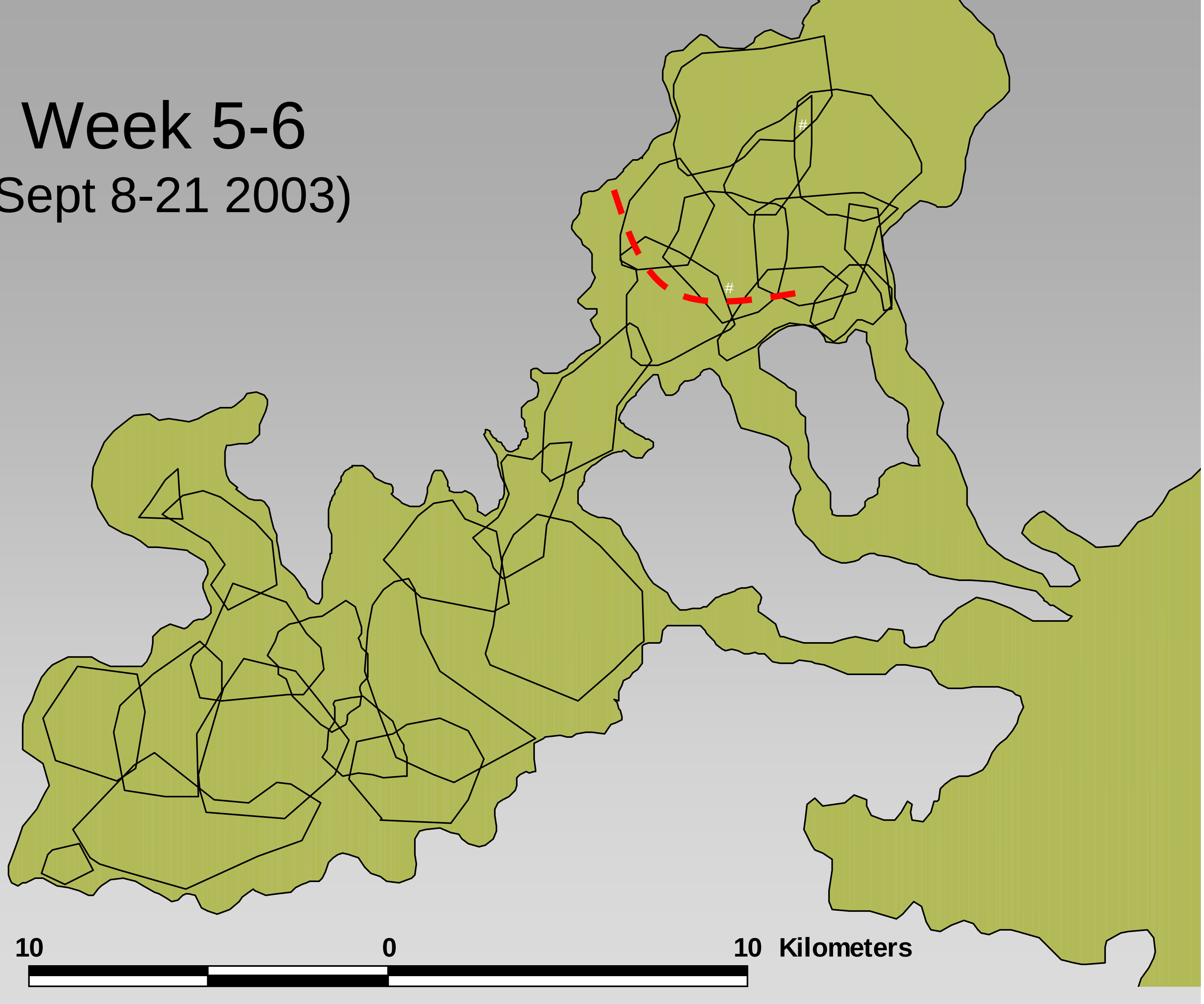
Week 3-4

(Aug 25-Sept 7 2003)



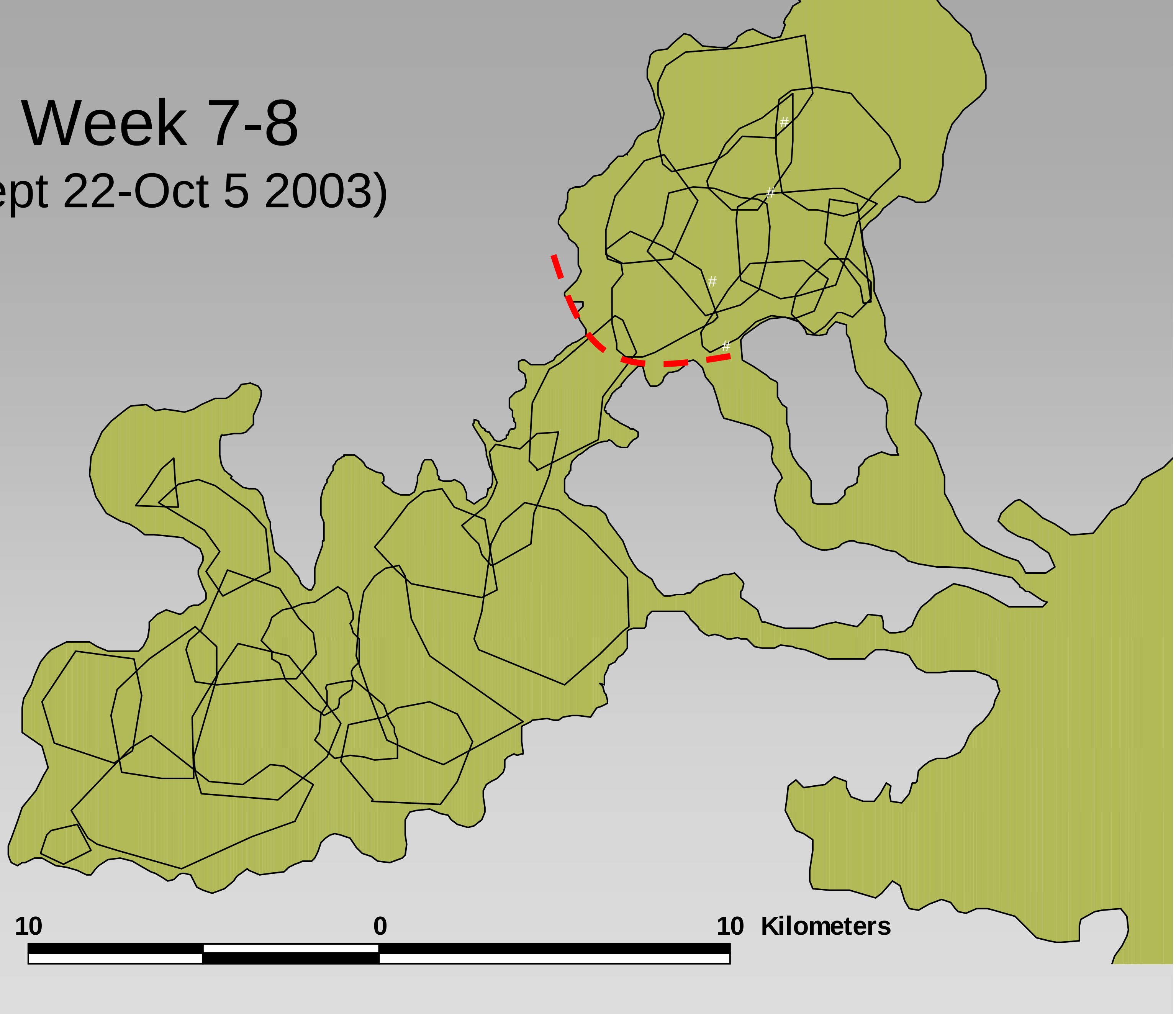
Week 5-6

(Sept 8-21 2003)



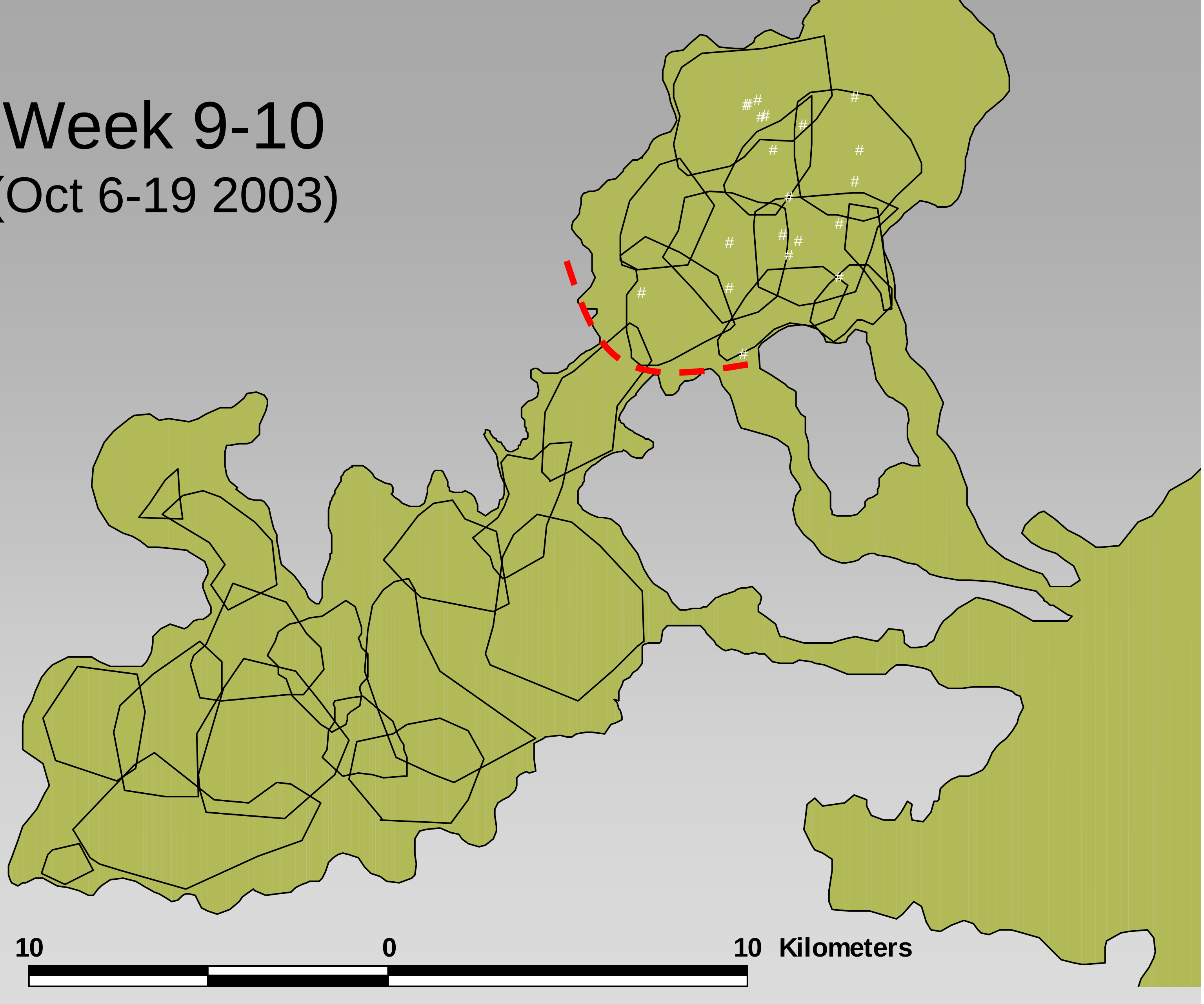
Week 7-8

(Sept 22-Oct 5 2003)



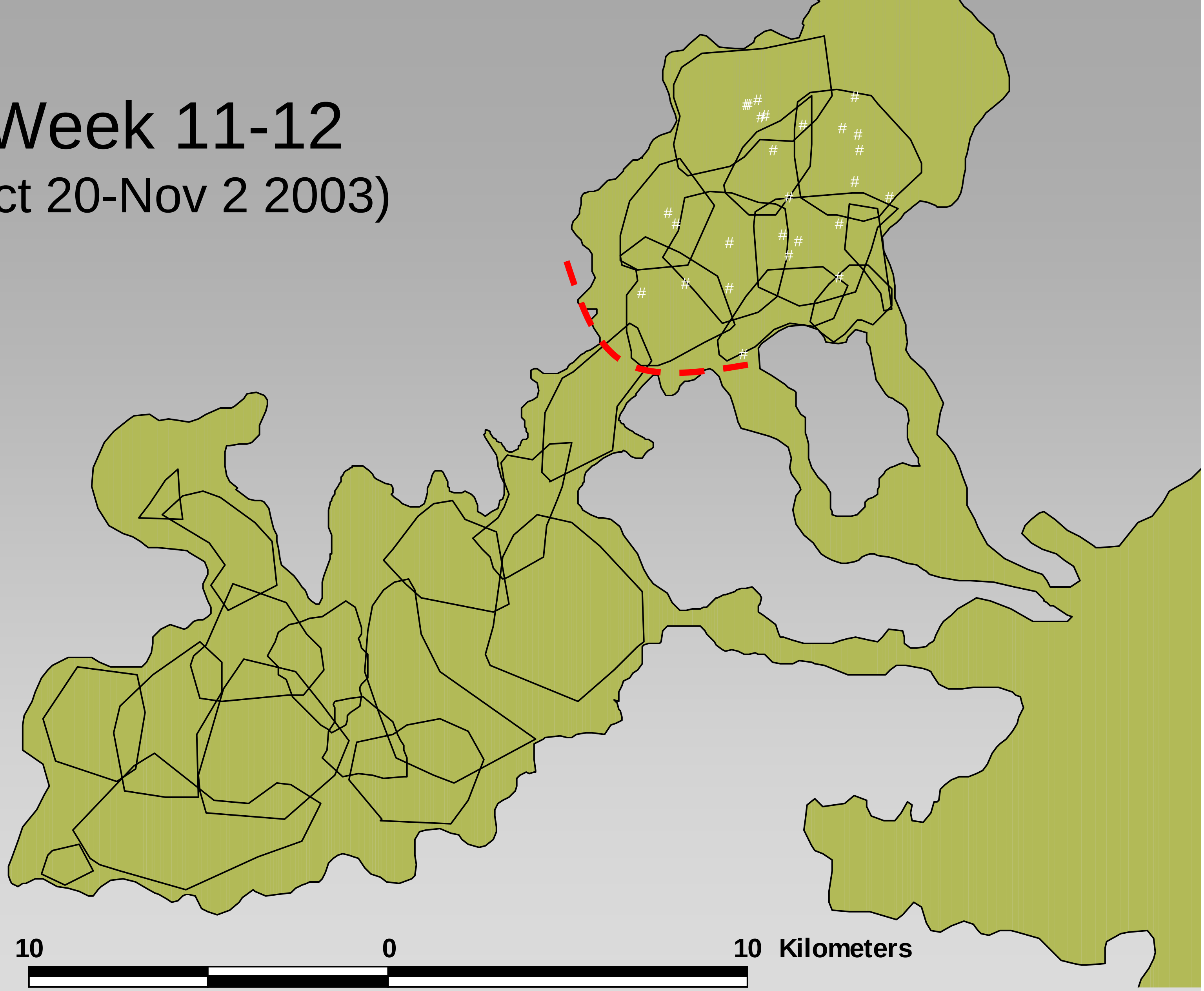
Week 9-10

(Oct 6-19 2003)



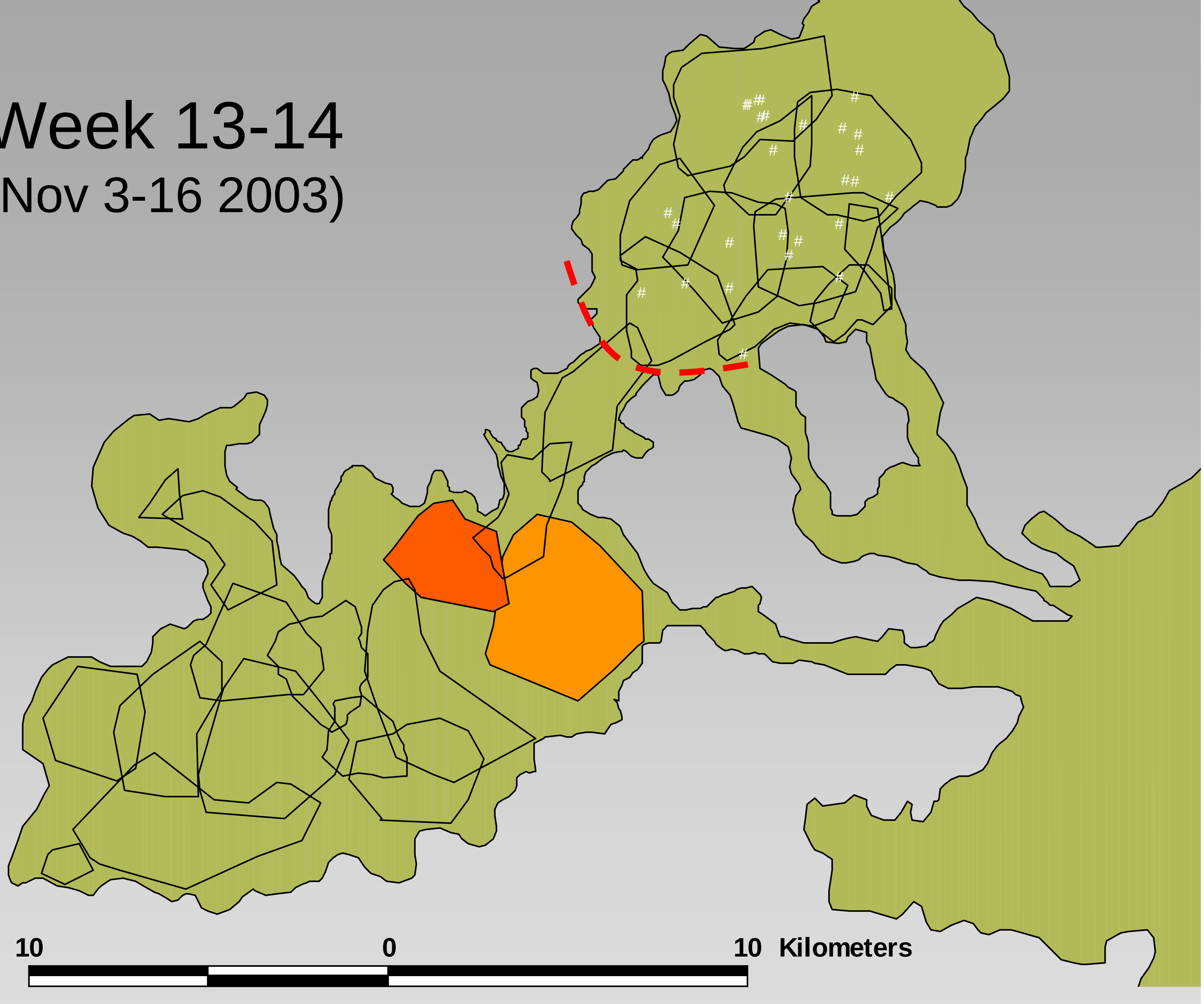
Week 11-12

(Oct 20-Nov 2 2003)



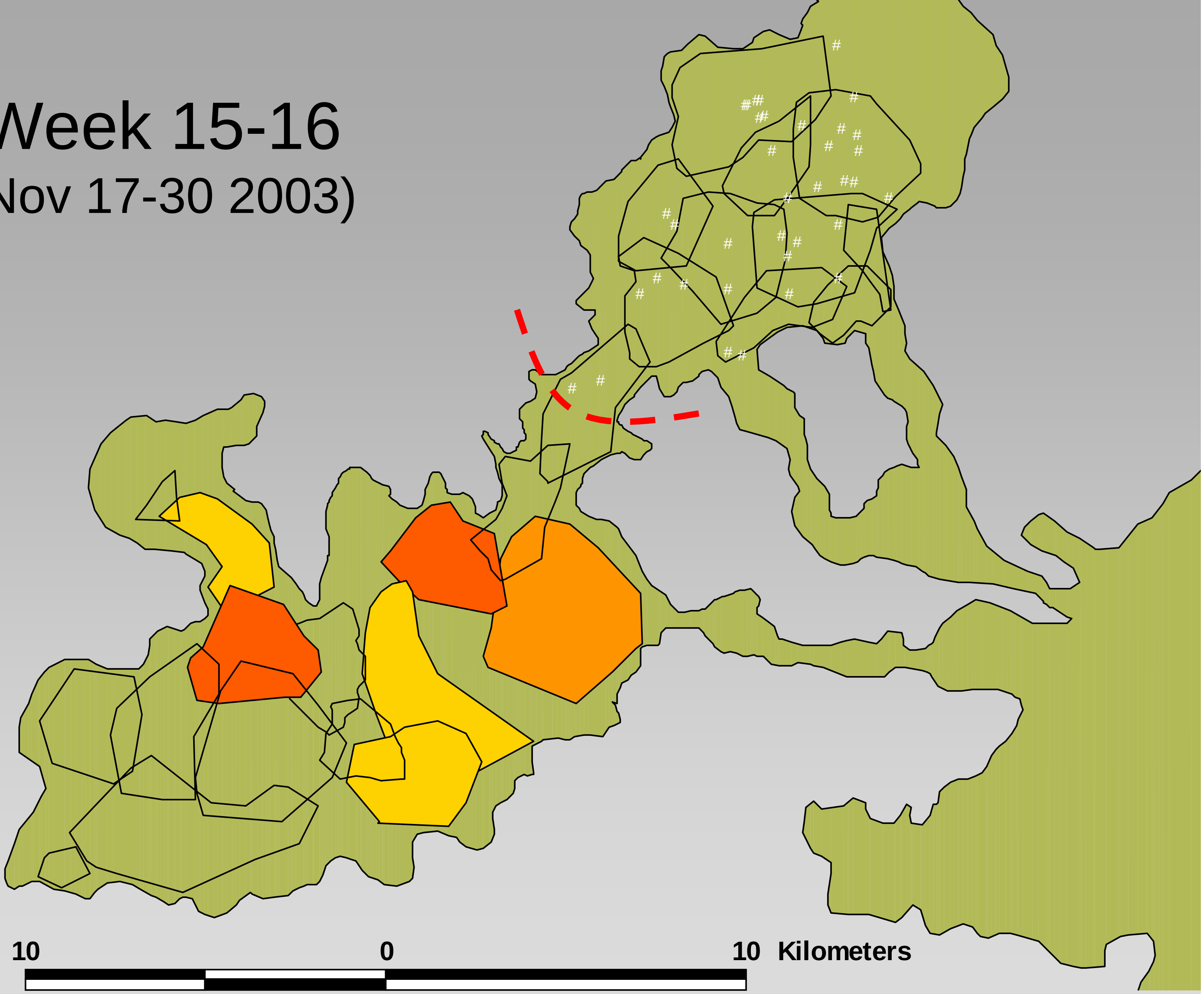
Week 13-14

(Nov 3-16 2003)



Week 15-16

(Nov 17-30 2003)

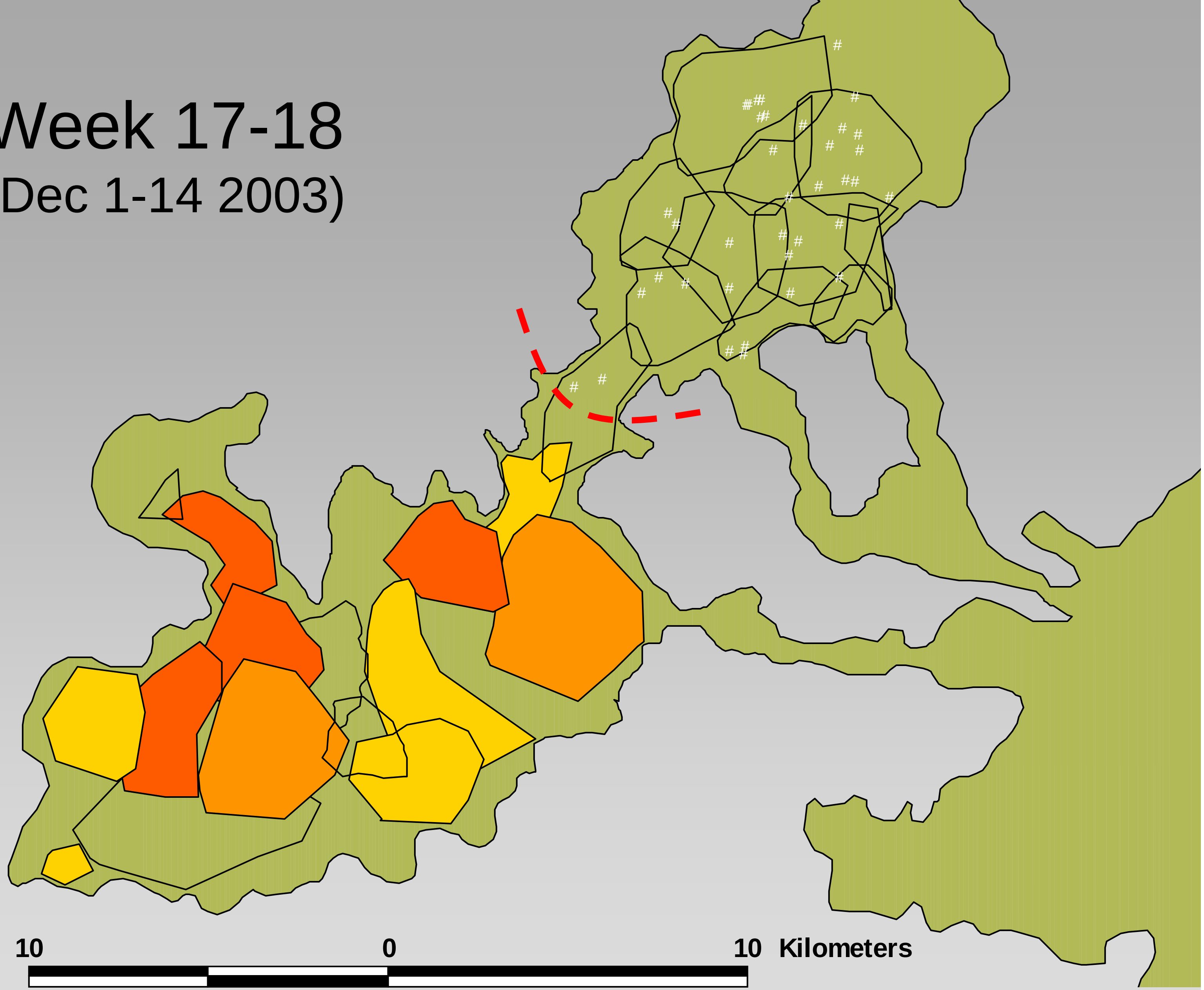


Vaccination coverage

- ≤ 20%
- 21-40%
- 41-60%
- 61-80%

Week 17-18

(Dec 1-14 2003)

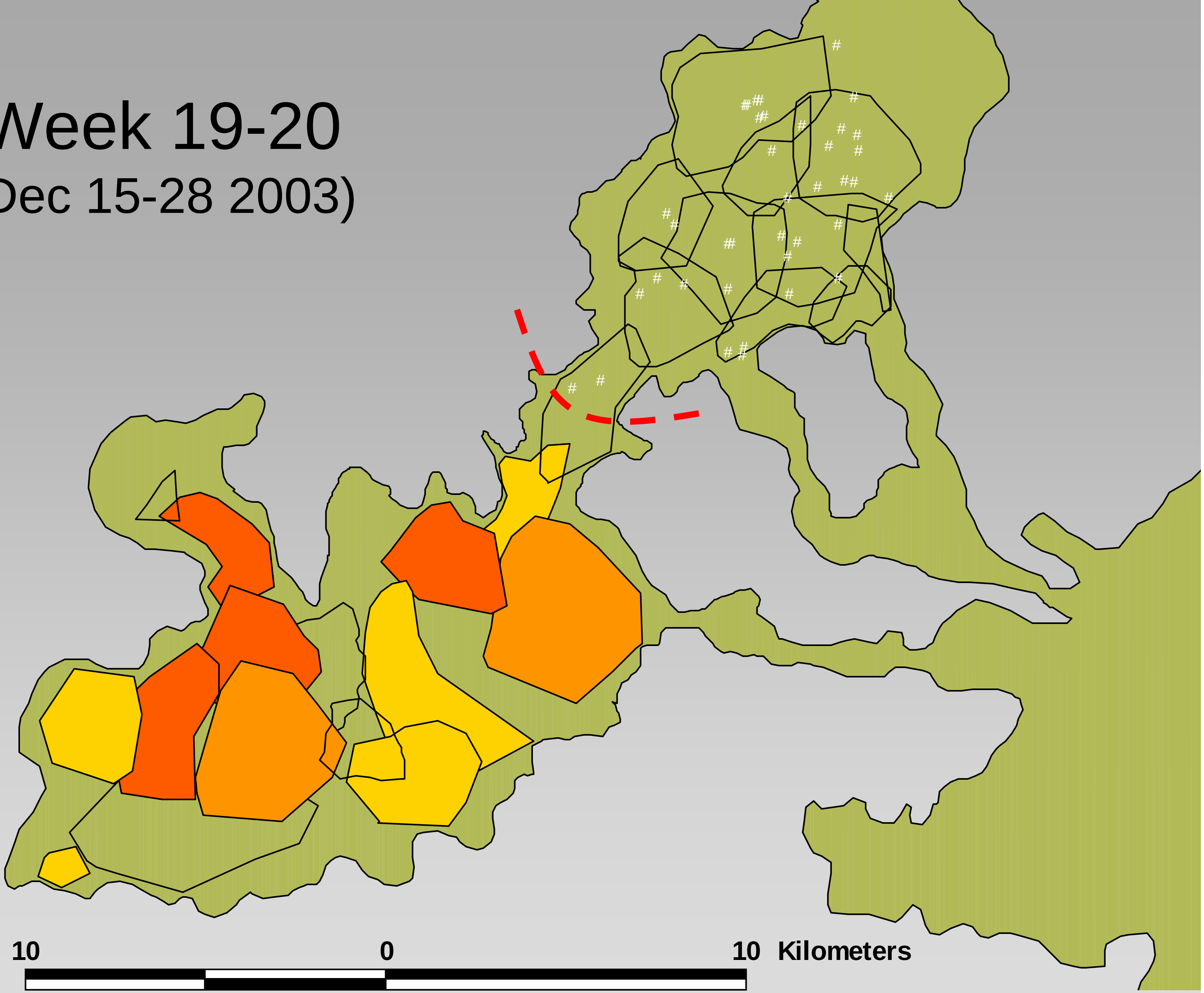


Vaccination coverage

- ≤ 20%
- 21-40%
- 41-60%
- 61-80%

Week 19-20

(Dec 15-28 2003)

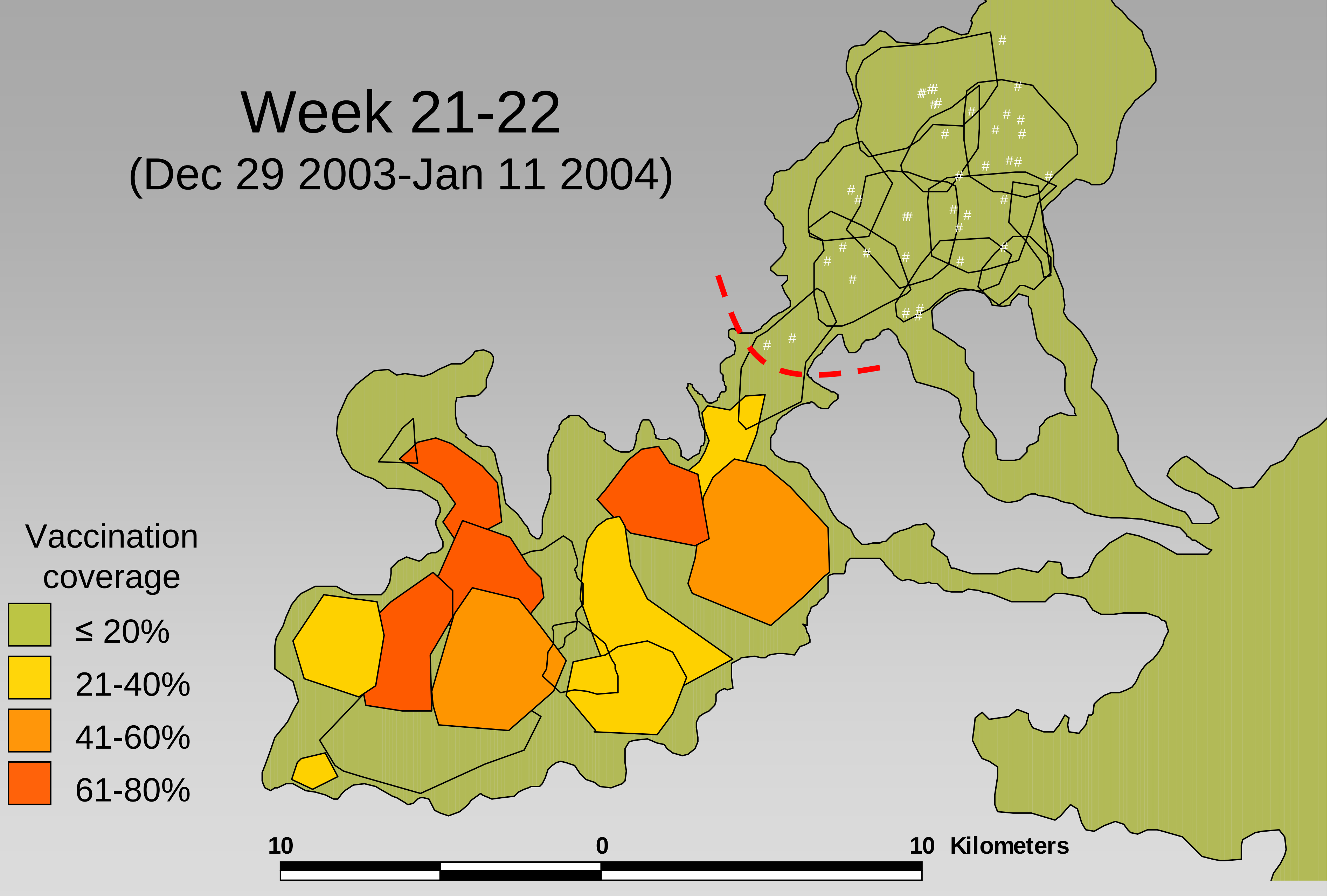


Vaccination coverage

- ≤ 20%
- 21-40%
- 41-60%
- 61-80%

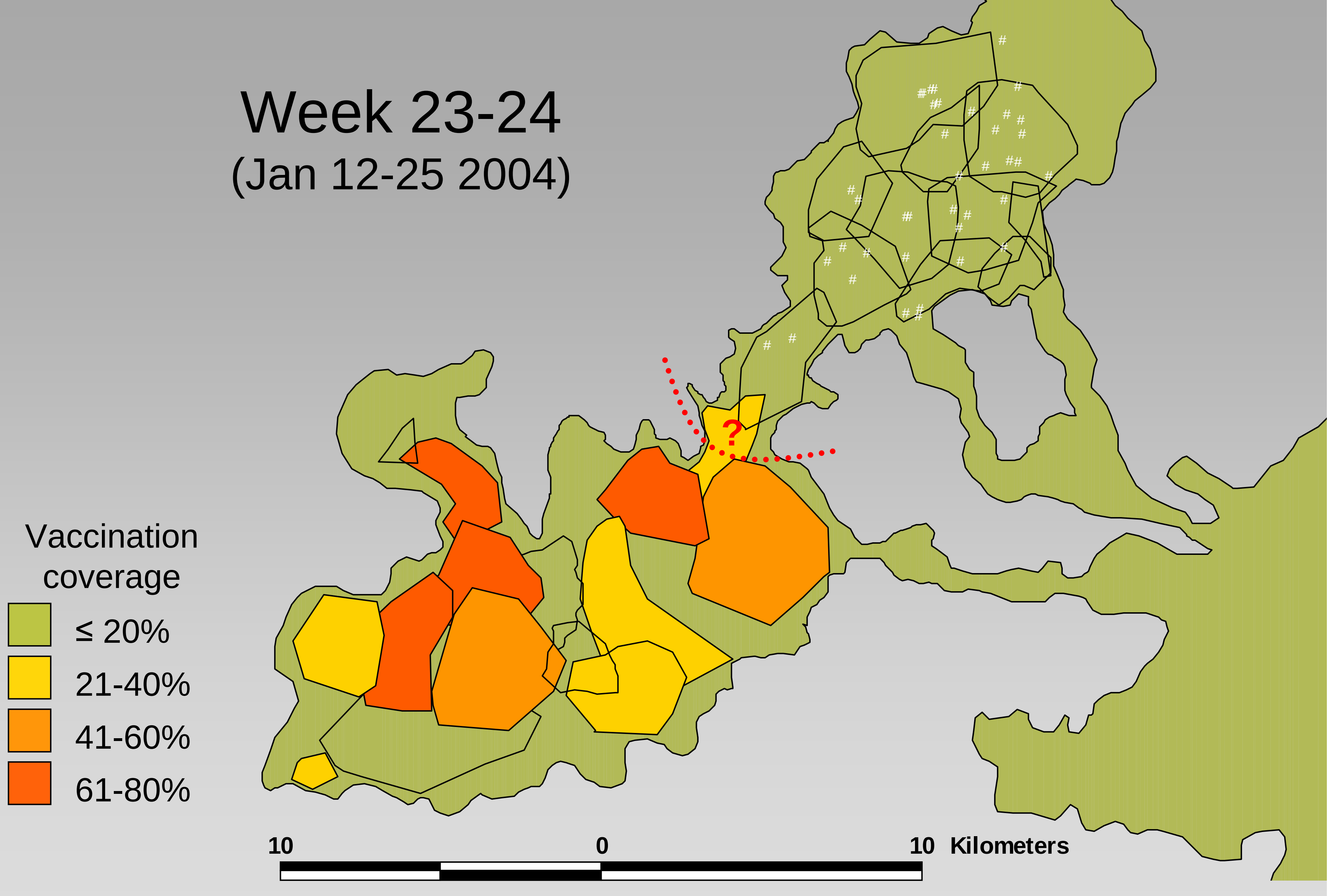
Week 21-22

(Dec 29 2003-Jan 11 2004)



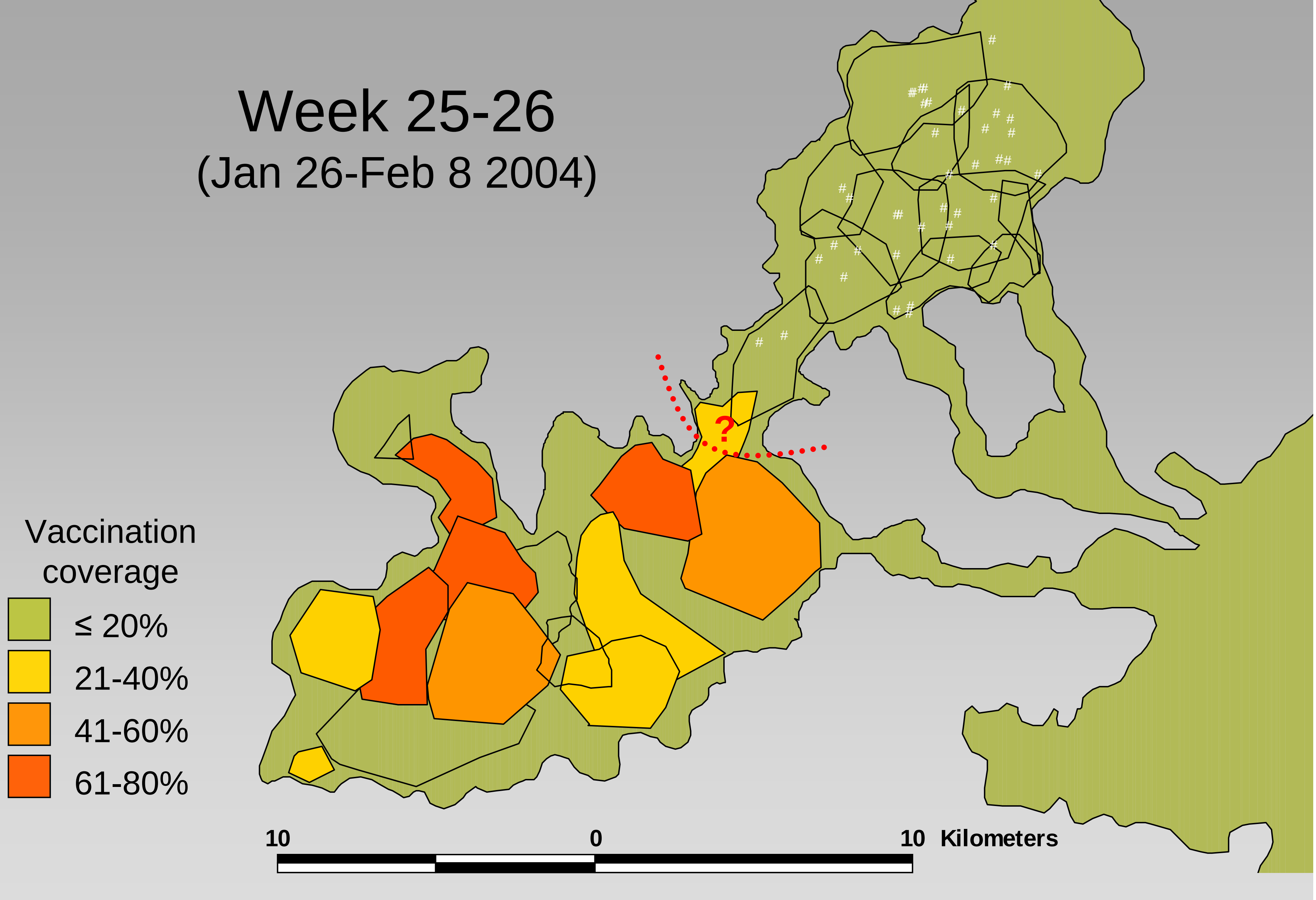
Week 23-24

(Jan 12-25 2004)



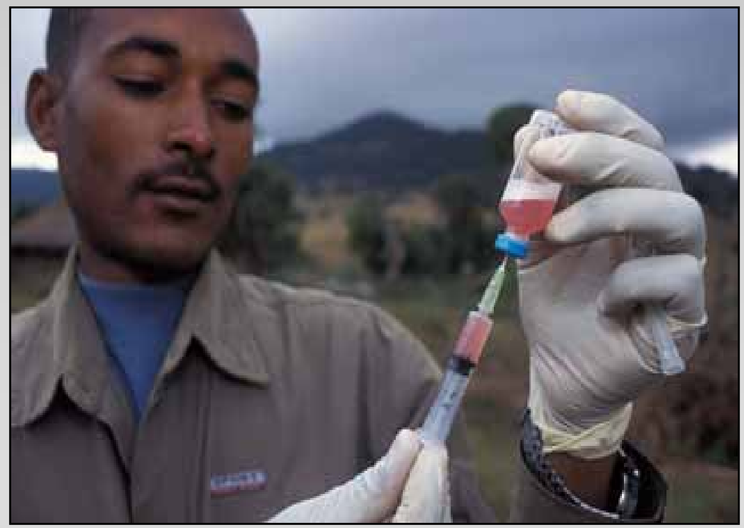
Week 25-26

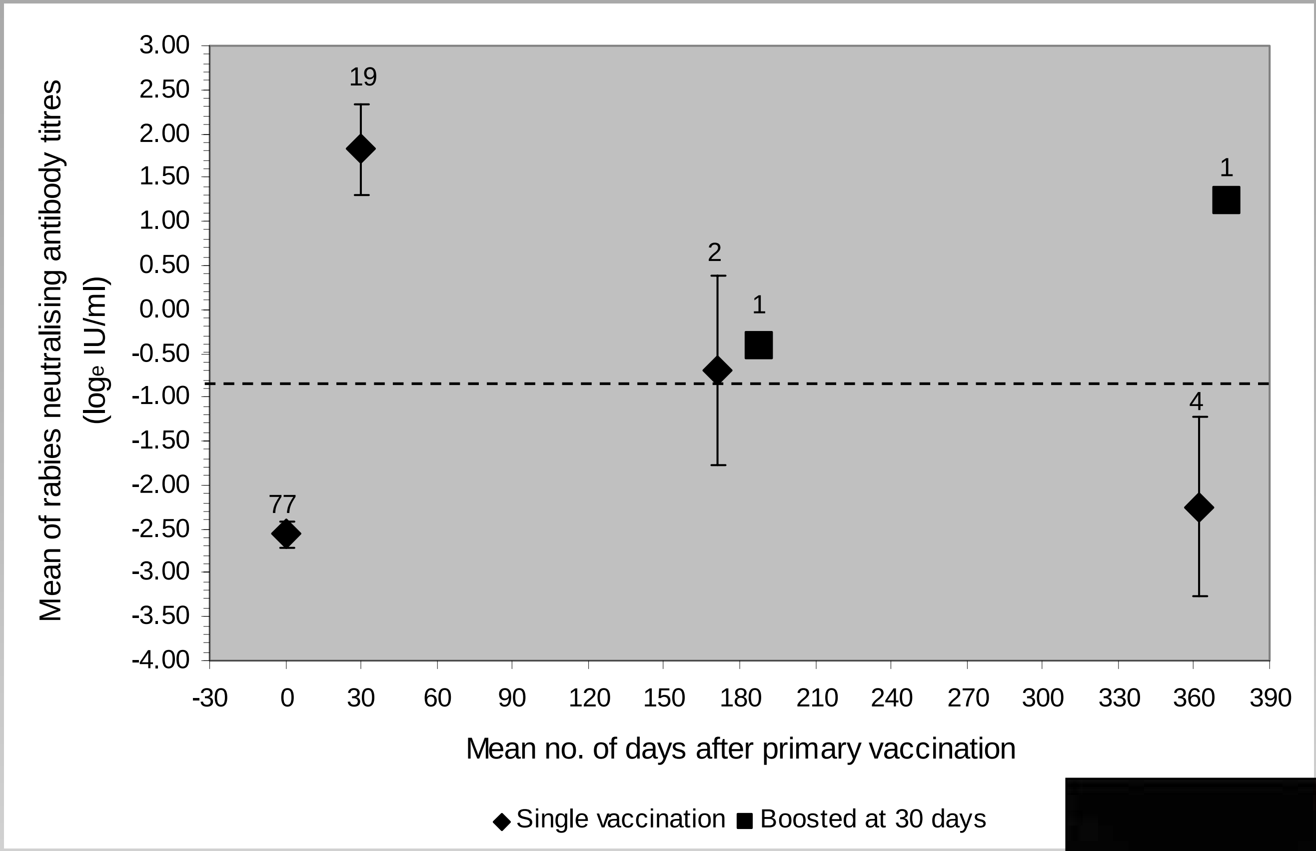
(Jan 26-Feb 8 2004)



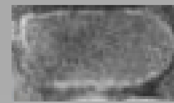
Summary of outbreak & intervention:

- 76% (72/95) known wolves died/disappeared in Web Valley & surroundings
- 84 (69+8+7) animals vaccinated in 114 capture events in Morebawa, Sanetti & Web Valley
- No rabies-related deaths within the vaccination zone

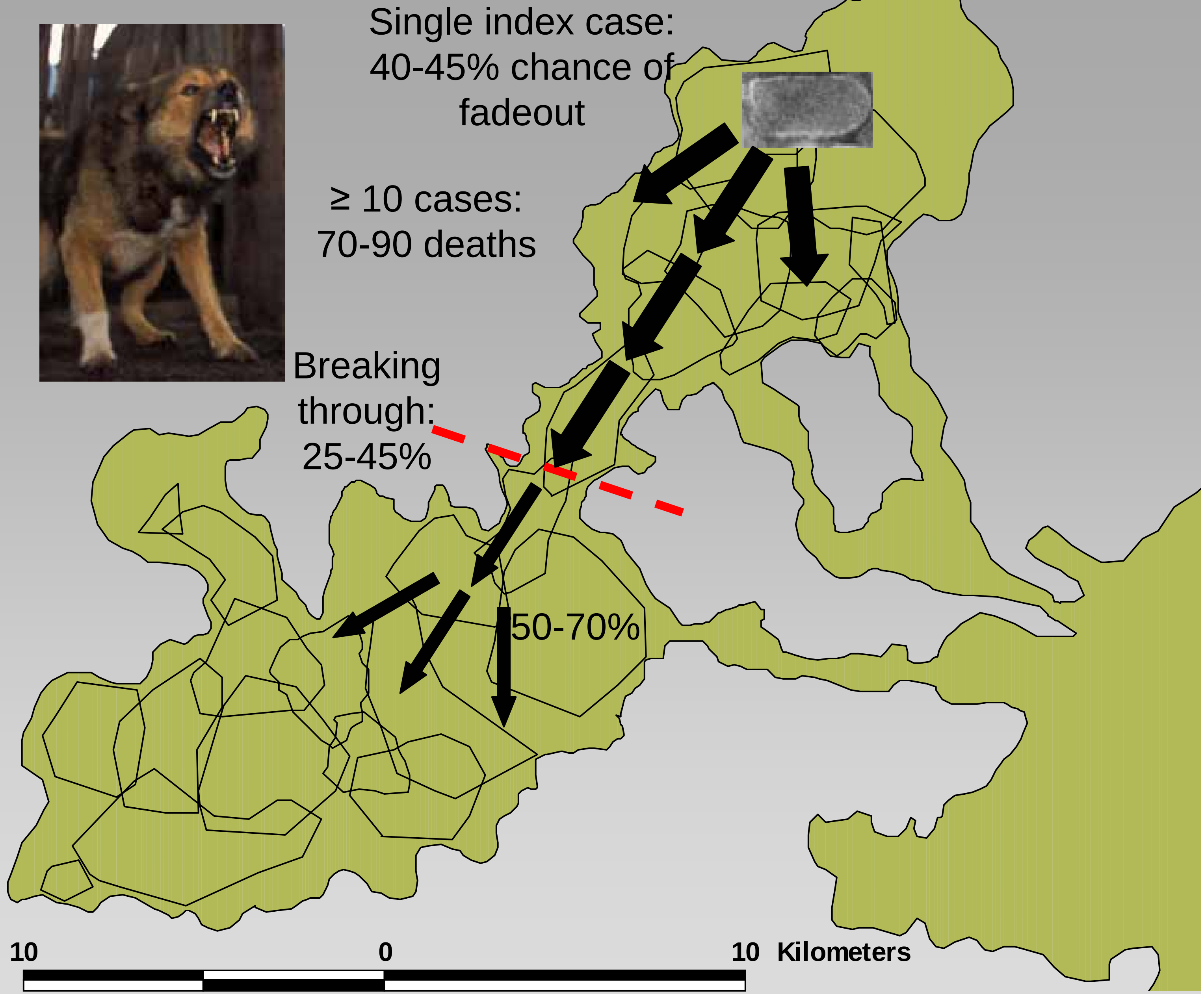




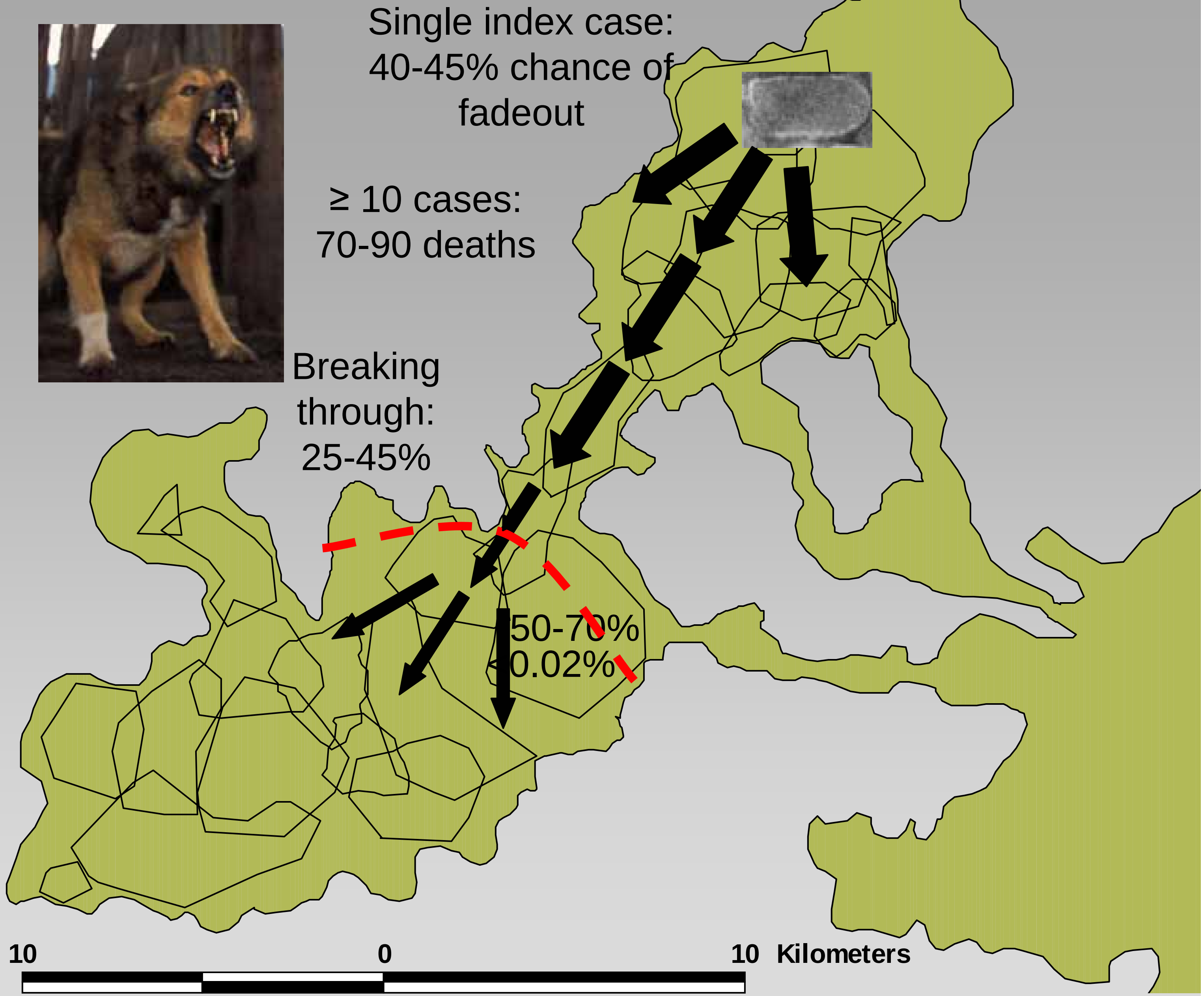
Effect	Analysis of variance of log titre at 30 days SS	Degr. of Freedom	MS	F	p
age	0.09323	1	0.09323	0.25544	0.622
sex	0.13062	1	0.13062	0.35787	0.560
batch	2.05144	1	2.05144	5.62056	0.034
dose	0.90786	1	0.90786	2.48735	0.139
Error	4.74485	13	0.36499		




10
0
10 Kilometers

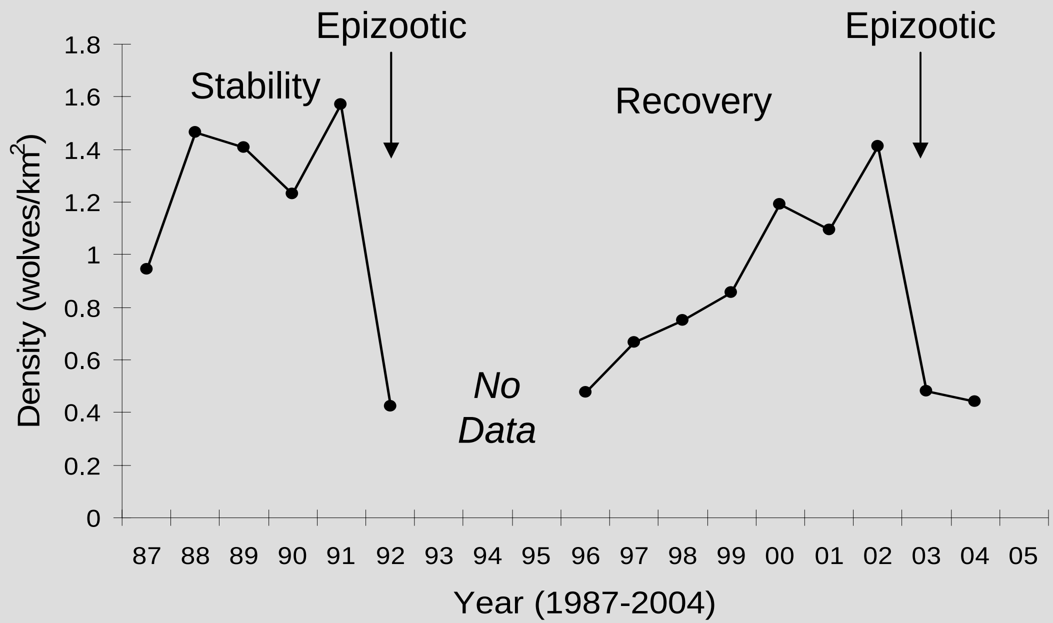


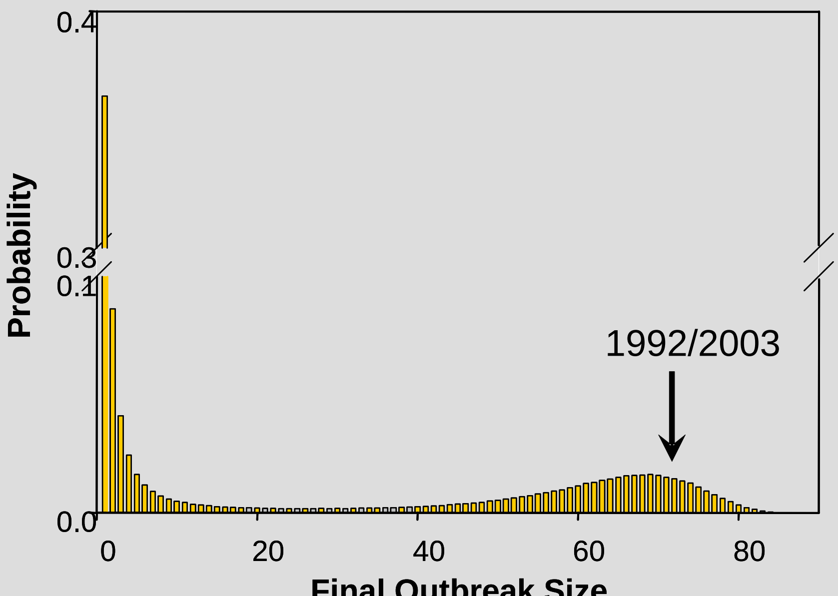
Single index case:
40-45% chance of fadeout
≥ 10 cases:
70-90 deaths
Breaking through:
25-45%
50-70%
10
0
10 Kilometers



Single index case:
40-45% chance of
fadeout
≥ 10 cases:
70-90 deaths
Breaking
through:
25-45%
50-70%
<0.02%
10
0
10 Kilometers

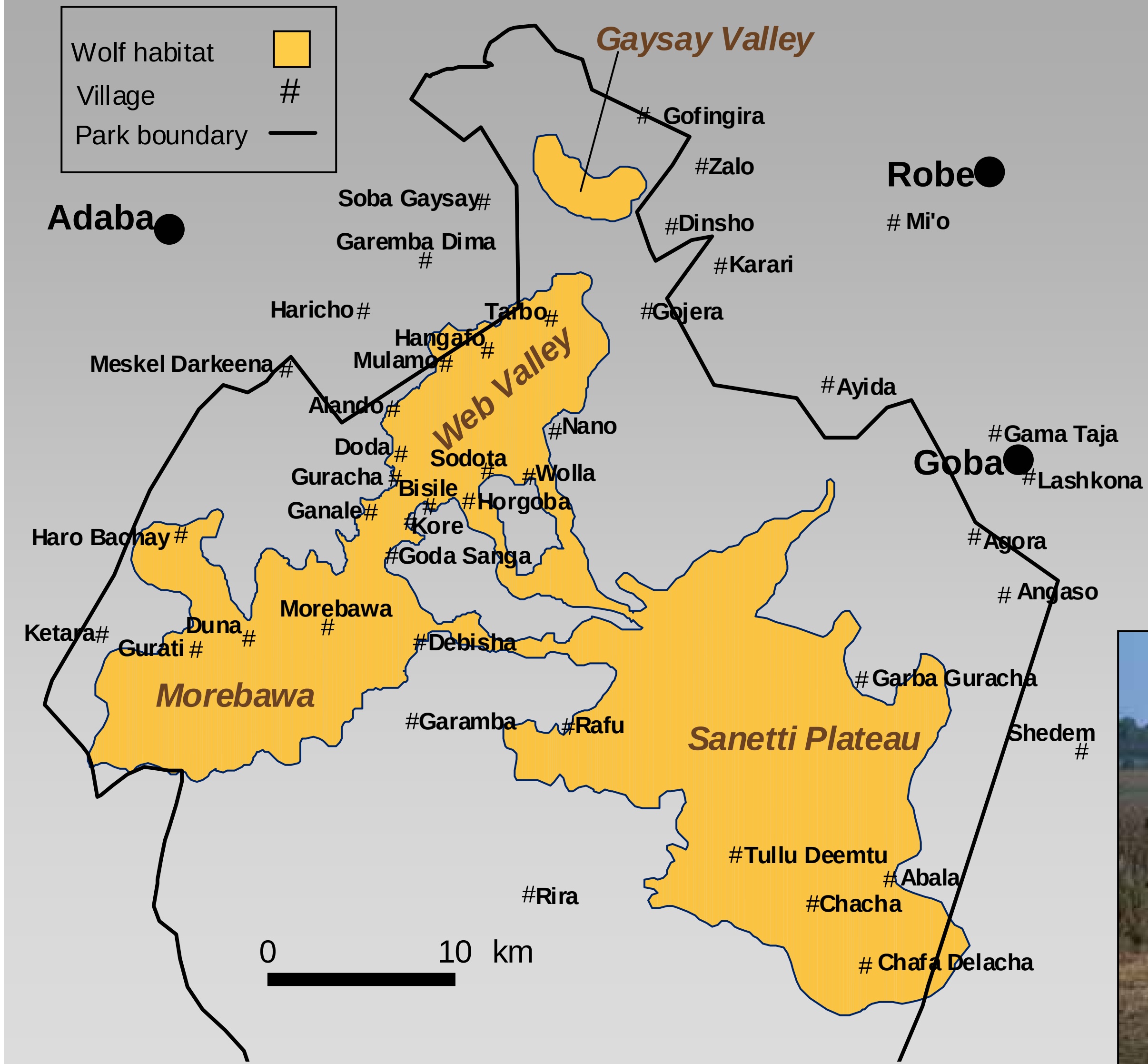
- Rise and fall of Web Valley wolves
  - 1992: 77% (41/53)
  - 2003: 76% (72/95)
  - Likely source: dogs



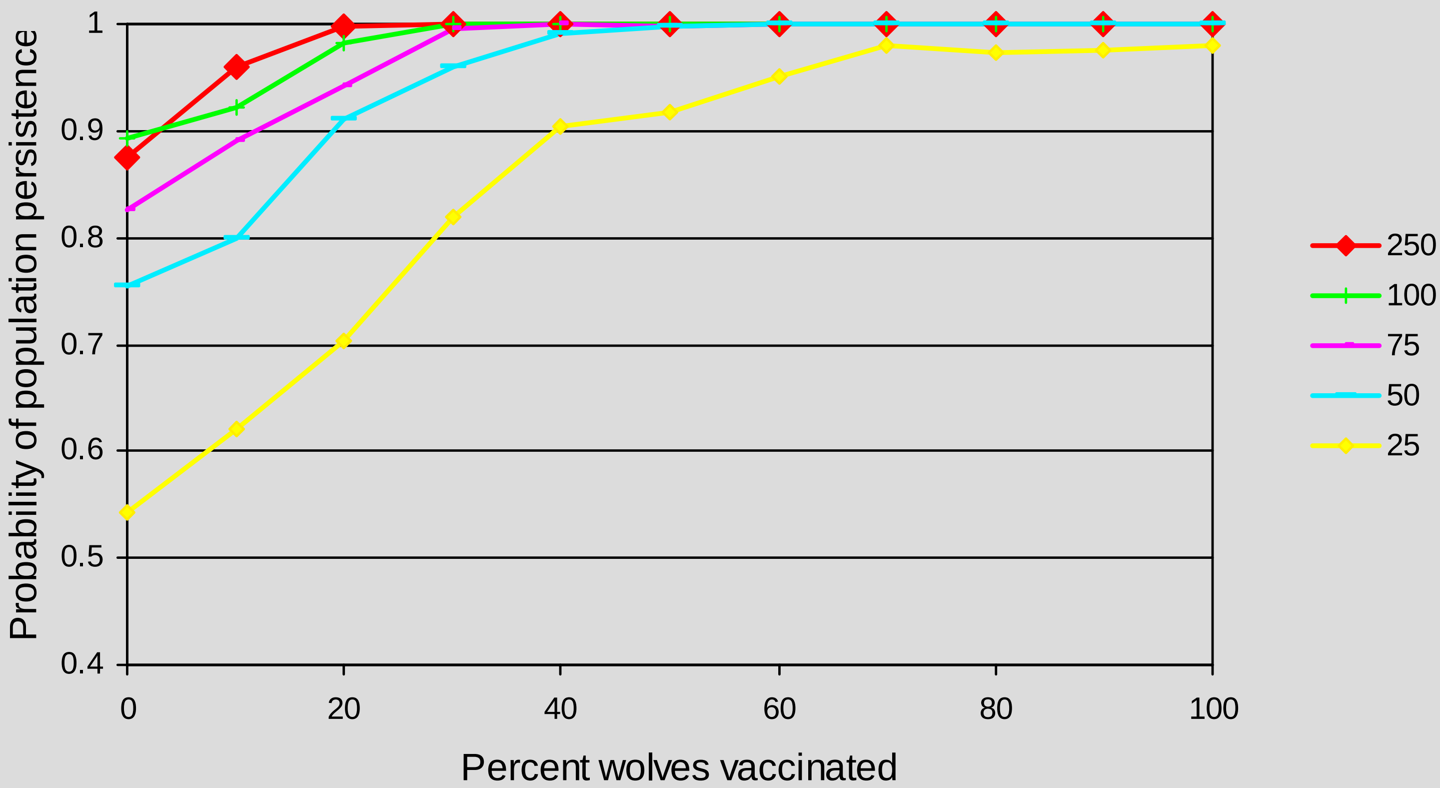


- Many small outbreaks (1-10 cases)
- > 10 case → 67-86% mortality

Bale Mountains – human settlements



- More than 42 villages in and around wolf habitat
- Dogs chase, compete for food and interbreed with wolves
- Direct dog-to-wolf contact is potential for disease transmission



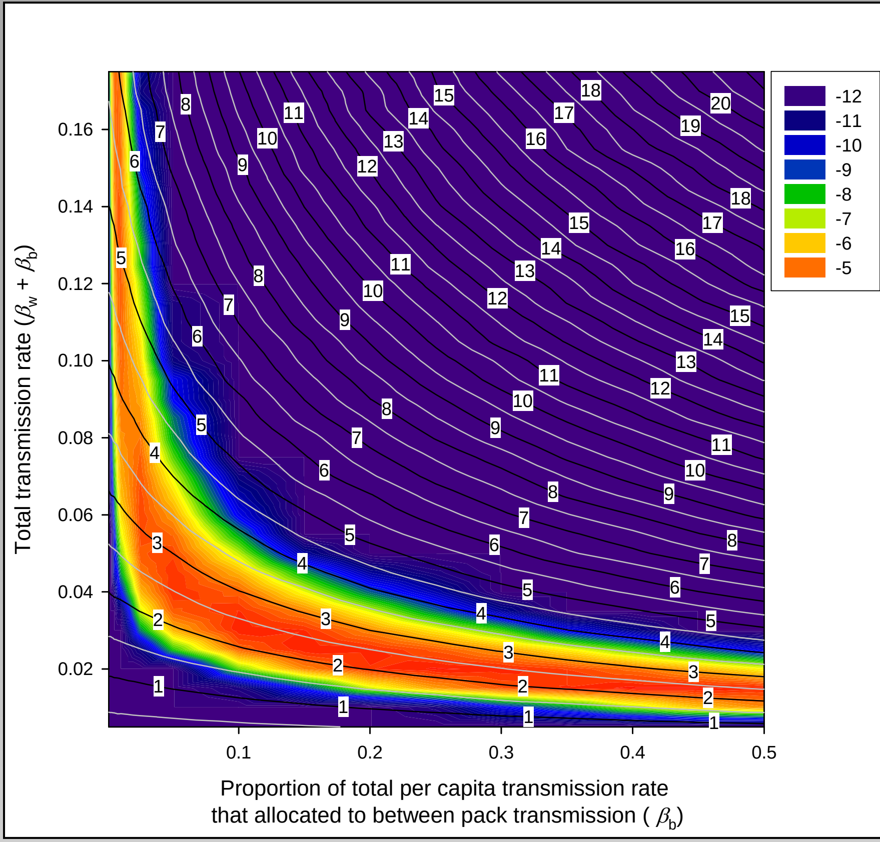
Alternate strategies:

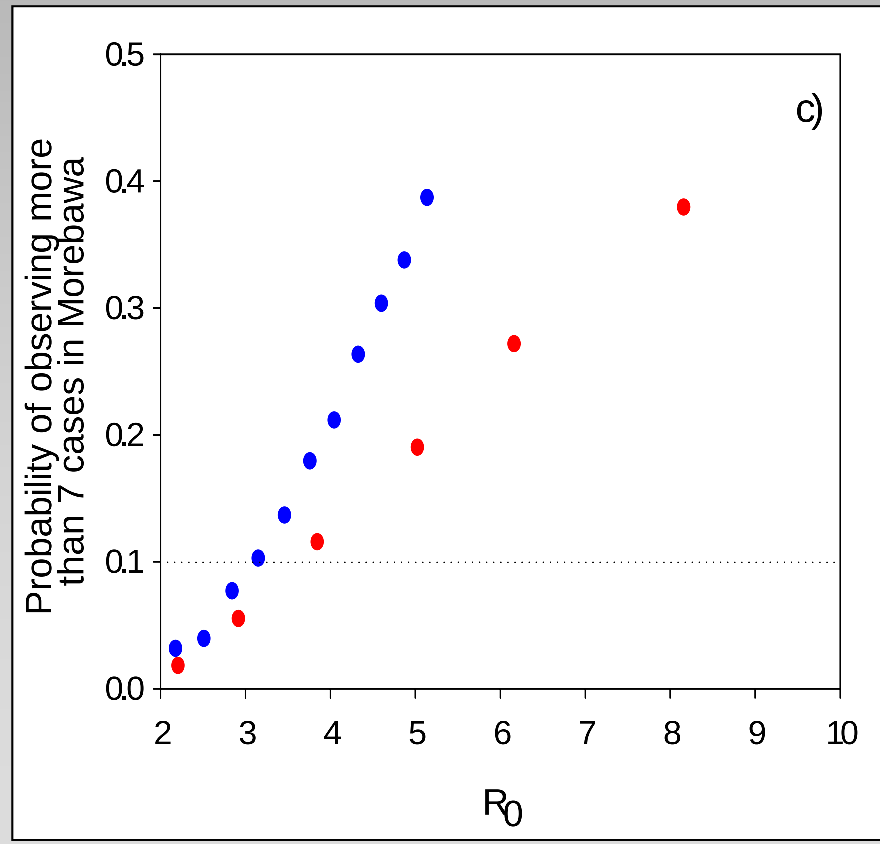
- Dog vaccination
- Oral vaccination
- Do nothing?

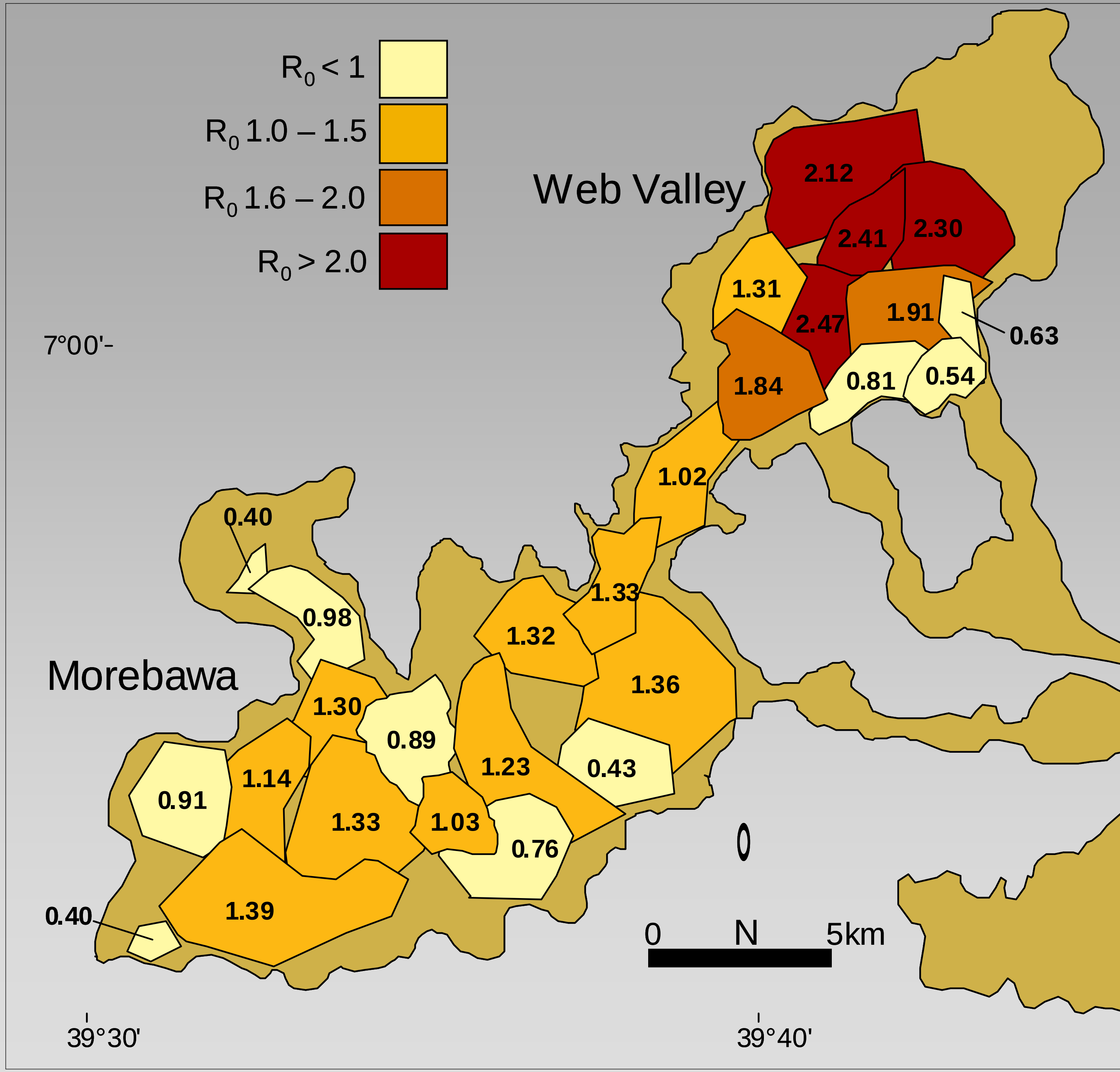
Acknowledgements

- Staff of the Ethiopian Wolf Conservation Programme
- Bale Mountains National Park, Oromia Regional Government, Ethiopia
- Ethiopian Wildlife Conservation Department, Addis Ababa, Ethiopia
- Centers for Disease Control and Prevention, USA
- Wellcome Trust; Morris Animal Foundation
- University of Edinburgh Small Project Grant

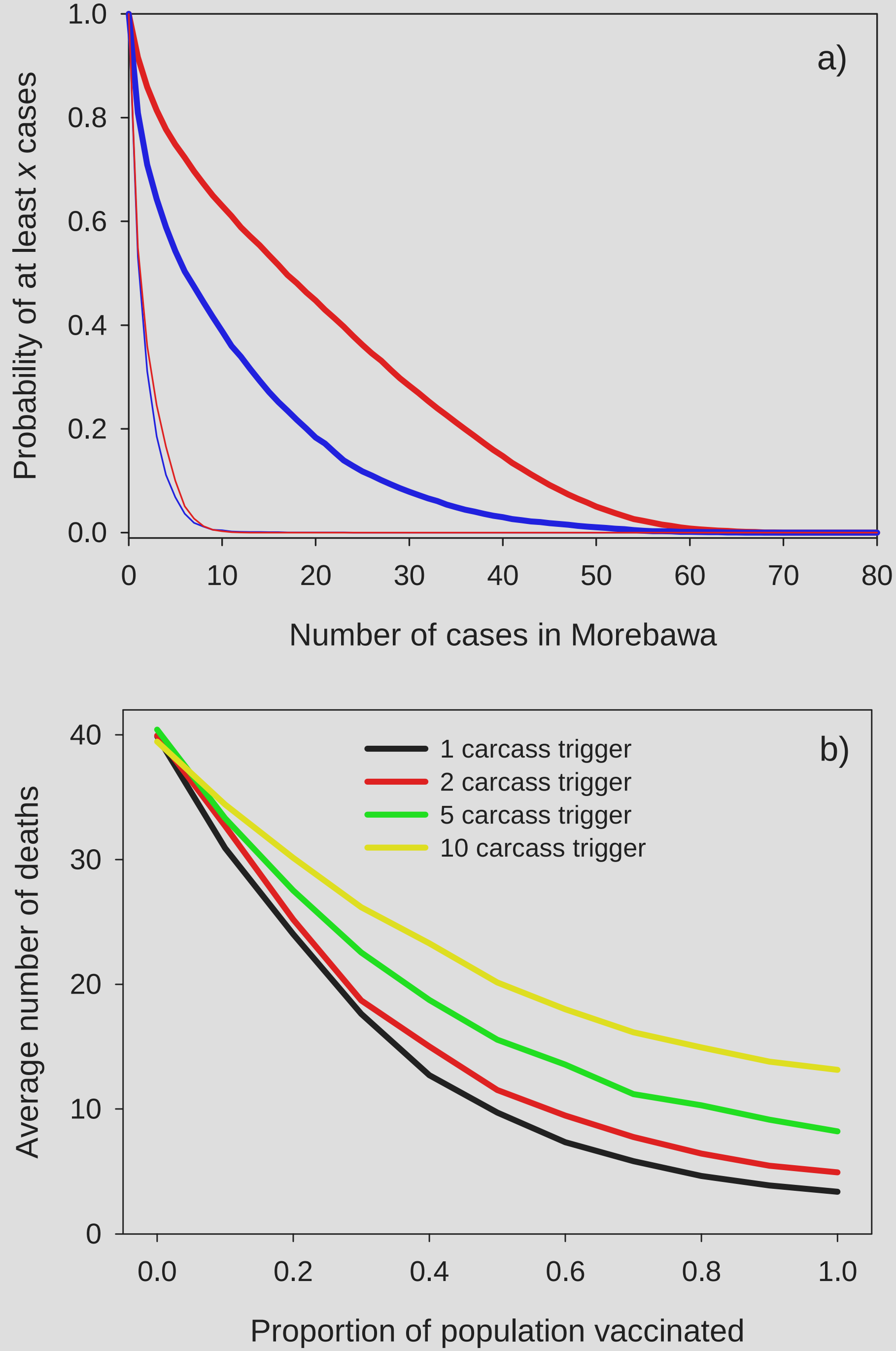
Photographs: Martin Harvey

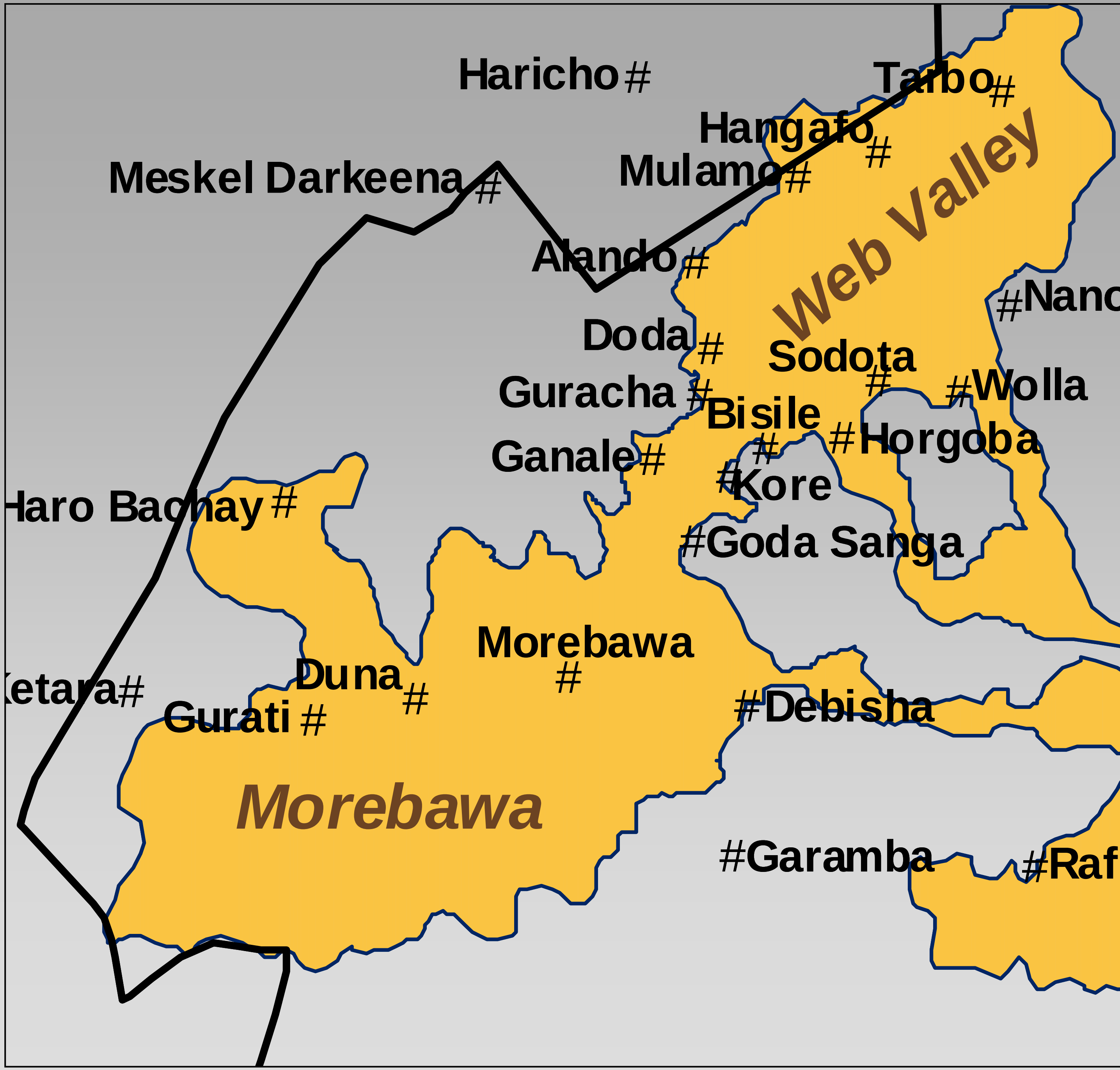

Total transmission rate ($\beta_w + \beta_b$)
0.16
0.14
0.12
0.10
0.08
0.06
0.04
0.02
0.1
0.2
0.3
0.4
0.5
Proportion of total per capita transmission rate
that allocated to between pack transmission (β_b)
-12
-11
-10
-9
-8
-7
-6
-5


c)
Probability of observing more
than 7 cases in Morebawa
0.5
0.4
0.3
0.2
0.1
0.0
2
3
4
5
6
7
8
9
10
R_0

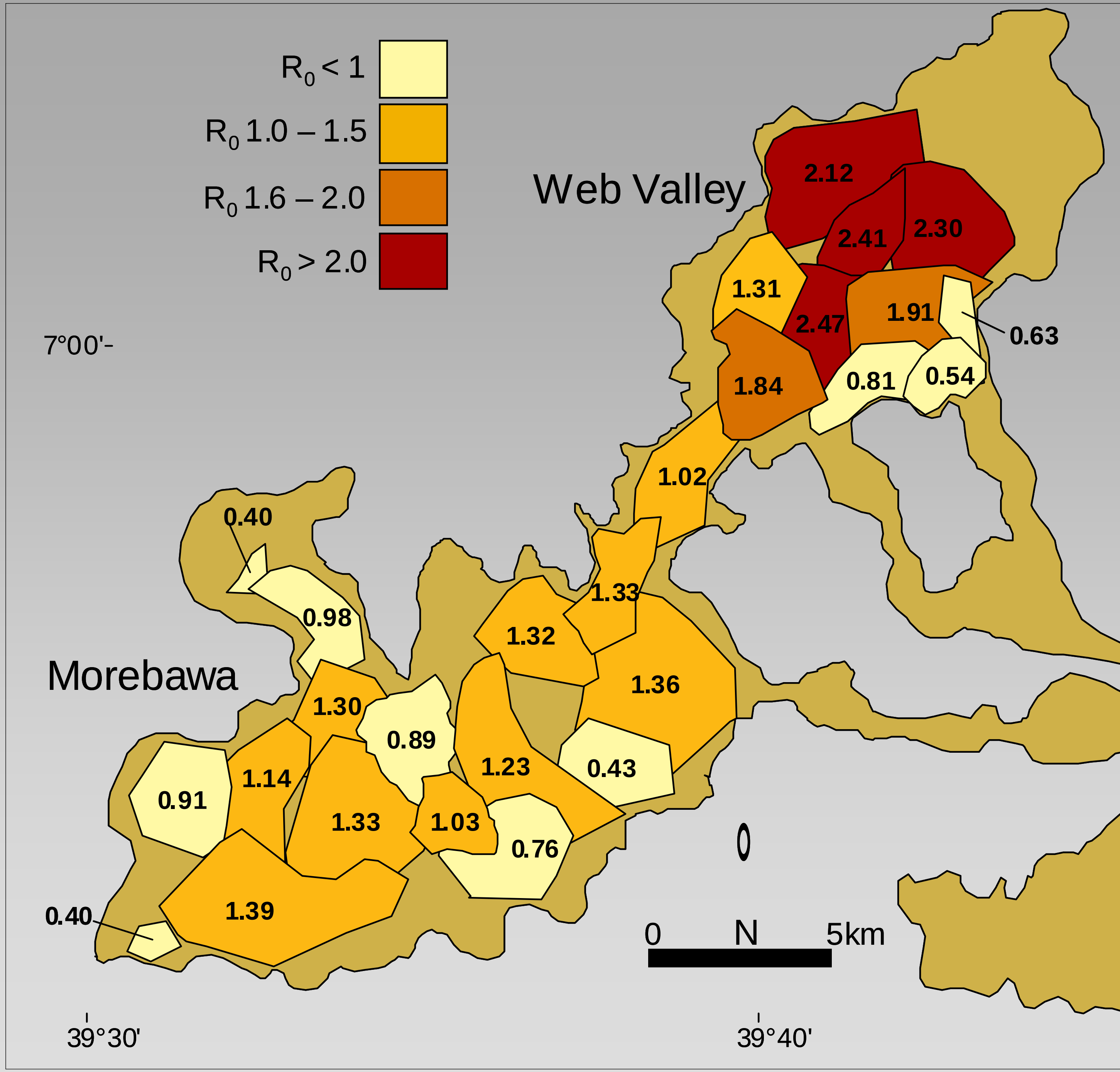


R0 < 1
R0 1.0 – 1.5
R0 1.6 – 2.0
R0 > 2.0
Web Valley
Morebawa
7°00'
2.12
2.41
2.30
1.31
2.47
1.91
0.63
1.84
0.81
0.54
1.02
0.40
0.98
1.33
1.32
1.36
1.30
0.89
1.23
0.43
1.14
0.91
1.33
1.03
0.76
1.39
0.40
0
N
5km
39°30'
39°40'


a)
Probability of at least x cases
1.0
0.8
0.6
0.4
0.2
0.0
0
10
20
30
40
50
60
70
80
Number of cases in Morebawa
b)
1 carcass trigger
2 carcass trigger
5 carcass trigger
10 carcass trigger
Average number of deaths
40
30
20
10
0
0.0
0.2
0.4
0.6
0.8
1.0
Proportion of population vaccinated



Haricho
Talbo
Hangafo
Mulamo
Meskel Darkeena
Web Valley
Alando
Nano
Doda
Sodota
Wolla
Guracha
Bisile
Horgoba
Ganale
Kore
Haro Bachay
Goda Sanga
Morebawa
Duna
etara
Gurati
Debisha
Morebawa
Garamba
Rafu



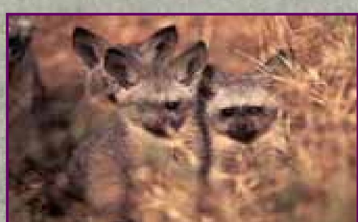
$R_0 < 1$
R_0 1.0 – 1.5
R_0 1.6 – 2.0
$R_0 > 2.0$
Web Valley
Morebawa
7°00'-
2.12
2.41
2.30
1.31
2.47
1.91
0.63
1.84
0.81
0.54
1.02
0.40
0.98
1.33
1.32
1.36
1.30
0.89
1.23
0.43
1.14
0.91
1.33
1.03
0.76
1.39
0.40
0
N
5km
39°30'
39°40'

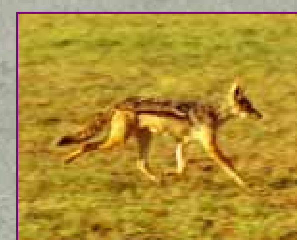
IDENTIFYING RESERVOIRS OF RABIES IN THE SERENGETI, TANZANIA

Tiziana Lembo

Sarah Cleaveland, Craig Packer, Andy Dobson

Magai Kaare, Katie Hampson, Ernest Eblate, Christine Mentzel, Meggan Craft



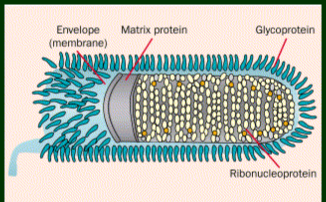


RABIES RESERVOIRS

Rabies infects all mammals, but not all mammals can act as reservoirs

For a species to act as a reservoir for rabies

- usually opportunistic host species living at high densities (e.g. foxes, raccoons, skunks, domestic dogs) with high birth rates
- adaptation between reservoir and virus that ensures maximal virus transmission

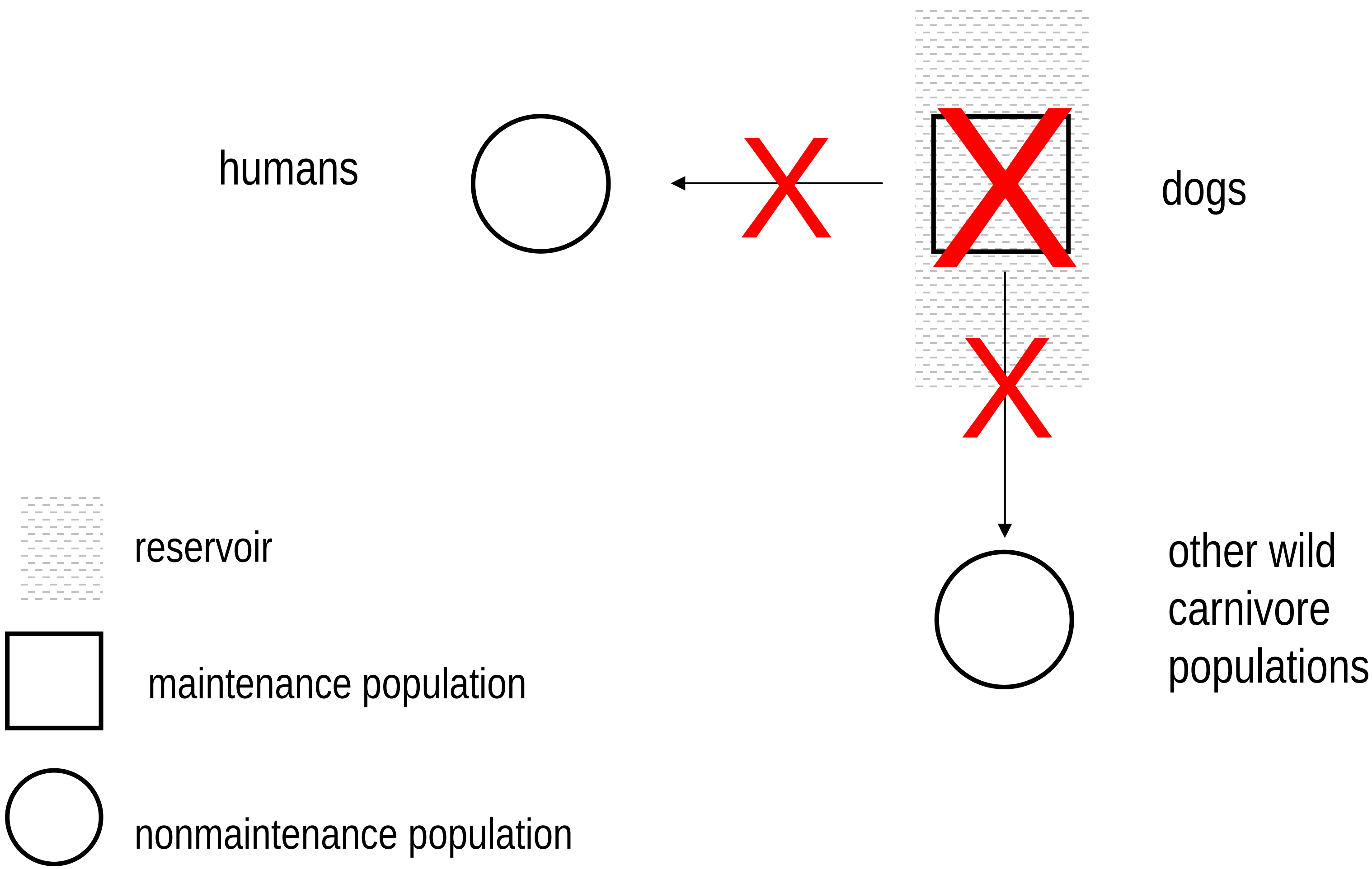


Why does identifying reservoirs matter?

- Knowledge of the reservoir species is critical for the design of effective disease control measures
- If rabies is controlled in the reservoir host population, it will disappear from all other species

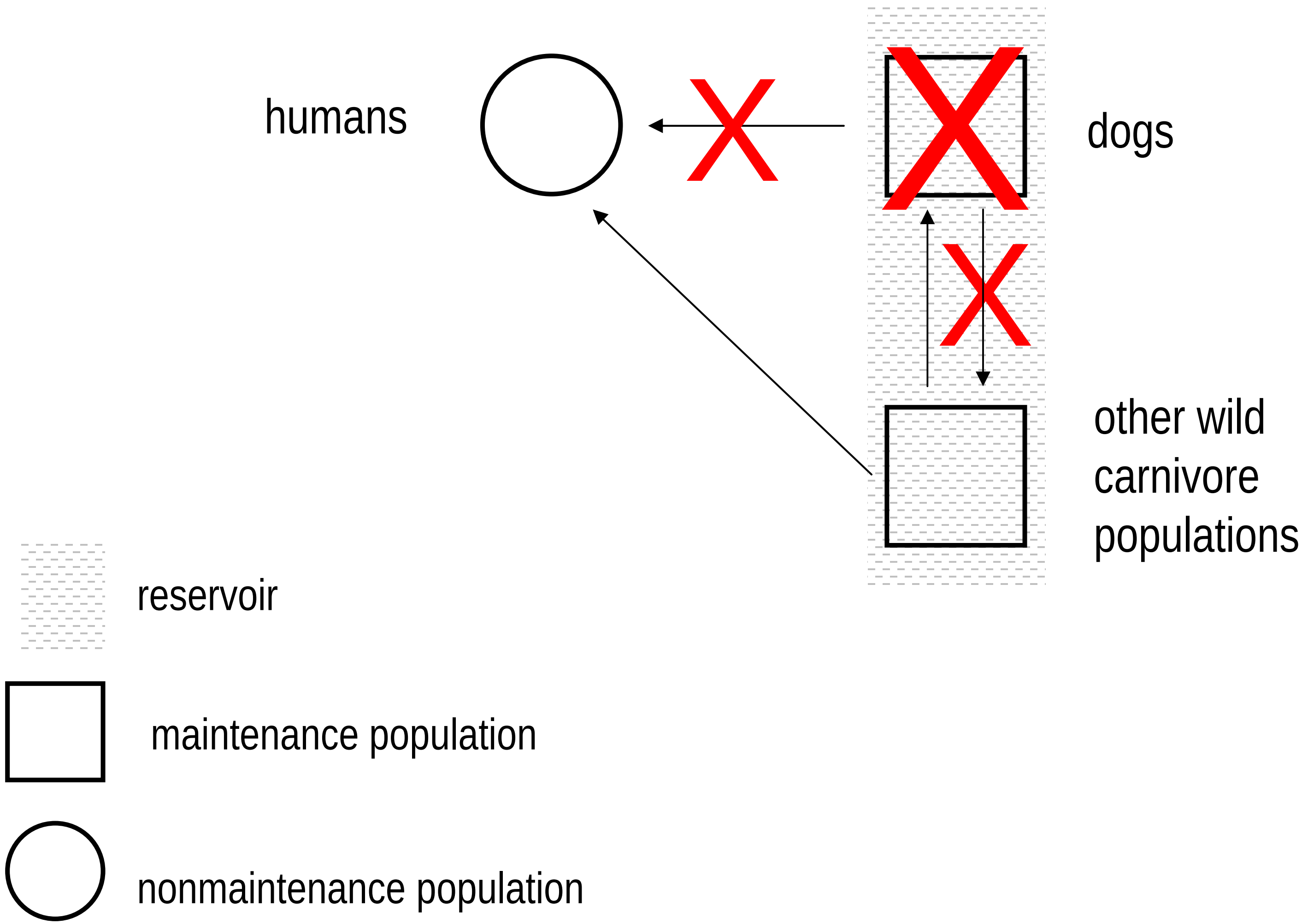
Relevance for Serengeti

Dogs as sole reservoir

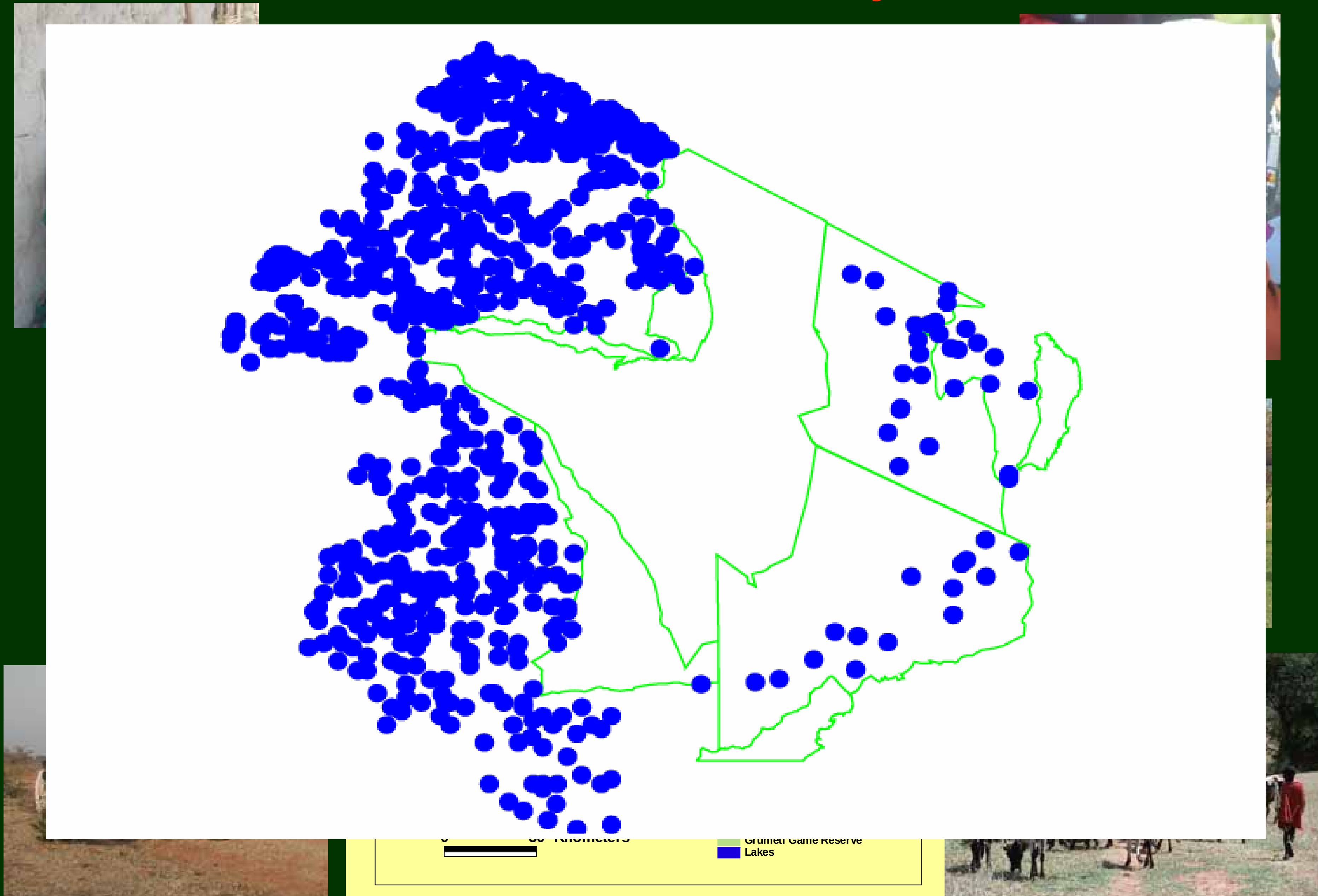


Relevance for Serengeti

Other carnivores maintaining rabies independently



Features of the ecosystem



No fences separate wildlife-protected areas and human settlements

More than 2 million people live in the ecosystem or in adjacent districts

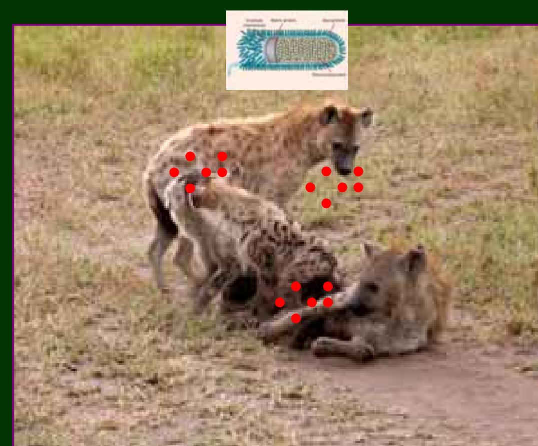
Wildlife, domestic animals and people often come into close contact

Mechanisms of rabies maintenance in the Serengeti

- Two mechanisms associated with 2 distinct variants of RABV:
  - <u>Typical</u> canid-associated isolated from domestic dog, African wild dog, bat-eared fox and white-tailed mongoose:
    - Virulent
    - Intra- and inter-specific transmission
    - Africa 1b

  - <u>Atypical</u> appearing to be maintained exclusively in spotted hyenas ("carrier state")-East et al., 2001:
    - A-virulent
    - Intra-specific transmission
    - Europe and Middle-East

BUT based on detection of viral RNA in saliva (PCR), no isolation of infectious virus



What tools can we use to help identify rabies reservoirs?

1. Evidence of persistence in the putative reservoir(s)

   Case-incidence data

2. Temporo-spatial relationship between dog and wildlife rabies cases

   Case-incidence data

   Trace-back data

   Contact patterns

3. Evidence of transmission to other species

   Is the same variant seen in different species or do different species maintain distinct variants?

   - Phylogenetic analysis

4. Intervention studies

   Control of rabies in domestic dogs through mass vaccination

1. Evidence of persistence in domestic dogs and wildlife

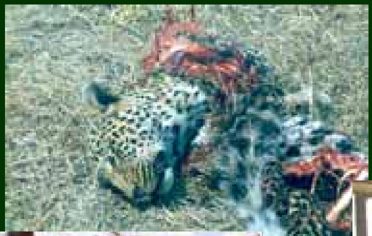
T. Lembo, K. Hampson, M. Kaare, E. Ernest, C. Mentzel, T. Mlengeya, R. Hoare and S. Cleaveland

Disease surveillance operations

- Disease surveillance networks
  - Inside SNP (park vets, scientists, rangers)
  - Outside SNP
    - Community-based active surveillance measures (LFOs)
    - Contact tracing from hospital records and livestock offices

- Post-mortem examination of domestic dog/wildlife carcasses

- Diagnosis in the field (dRIT-direct rapid immunohistochemical test)



dRIT

T.Lembo, M.Niezgoda, A.Velasco-Villa, S.Cleaveland, E.Ernest and C.E.Rupprecht

Developed and optimised at the Rabies section of the CDC as a confirmatory test for the DFA test with the potential to be used as a field test for rabies diagnosis and surveillance

Carried out on brain touch impressions

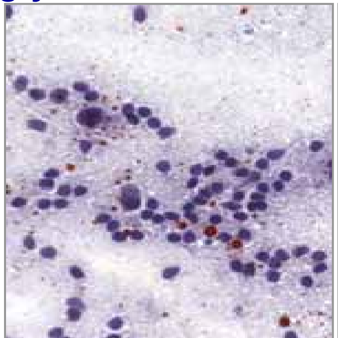
Based on antibody recognition of rabies antigen

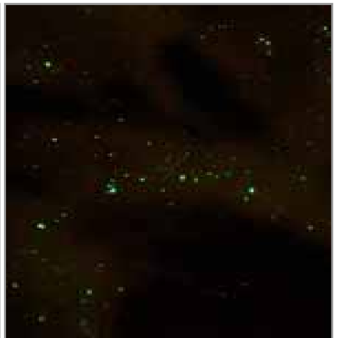
As a colorimetric test the only equipment required is an ordinary light microscope

The dRIT allows rabies diagnosis under field conditions

Preliminary evaluation in TZ:

- compared to DFA test, 100% sensitive and specific (n = 159)
- successfully performed on samples preserved in glycerol and in variable conditions of preservation

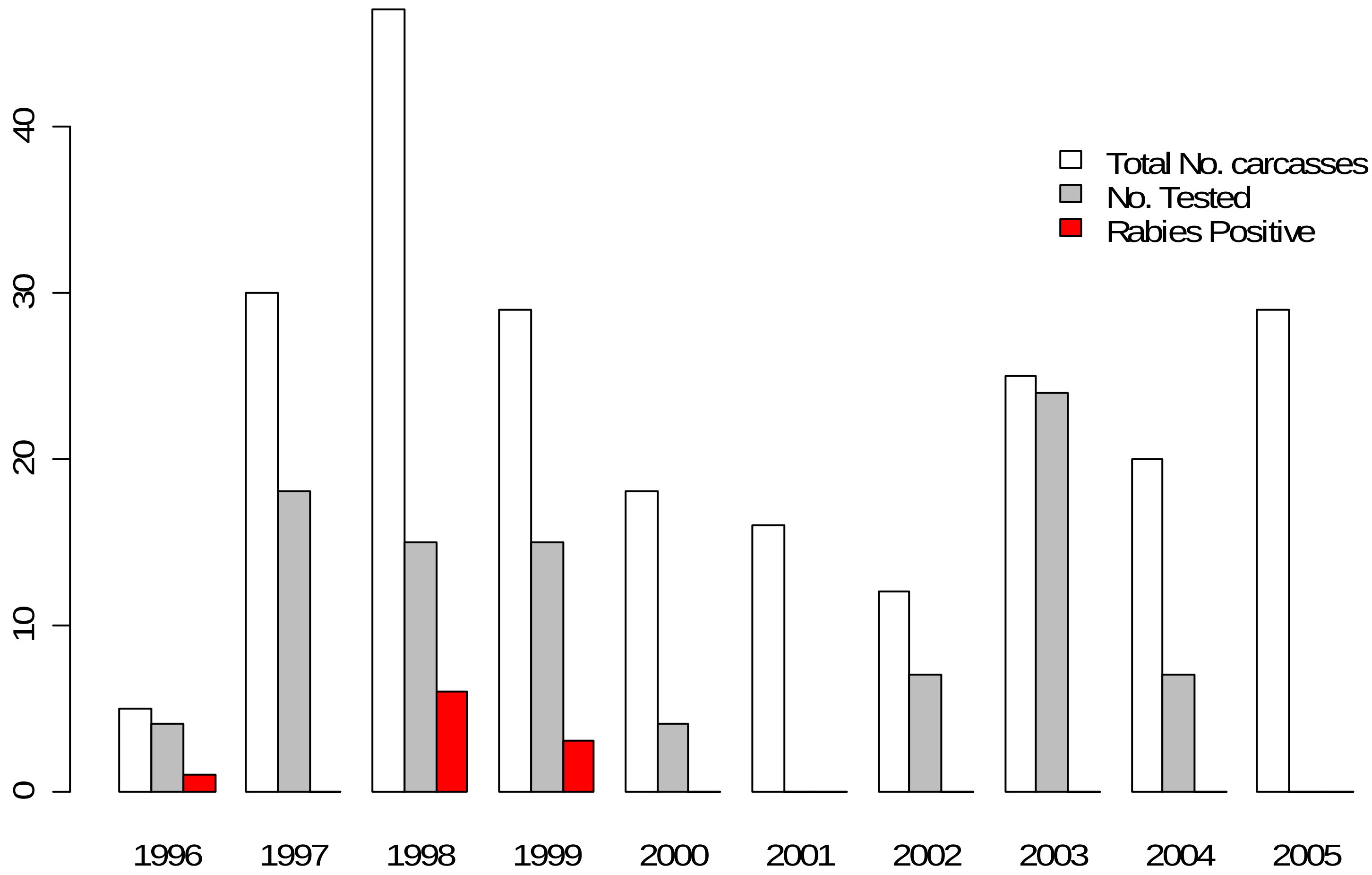




Domestic dog rabies

- Rabies does persist in domestic dogs in higher-density populations (>5 dogs/km²)
  - Dog rabies cases reported every year in districts to the west of SNP, but only sporadically in the NCA
  - Veterinary office records, hospital records (rabid dog-bite injuries), questionnaire data

WILDLIFE RABIES CONFIRMED CASES IN SNP



Laboratory confirmation AFSSA

Laboratory confirmation CDC

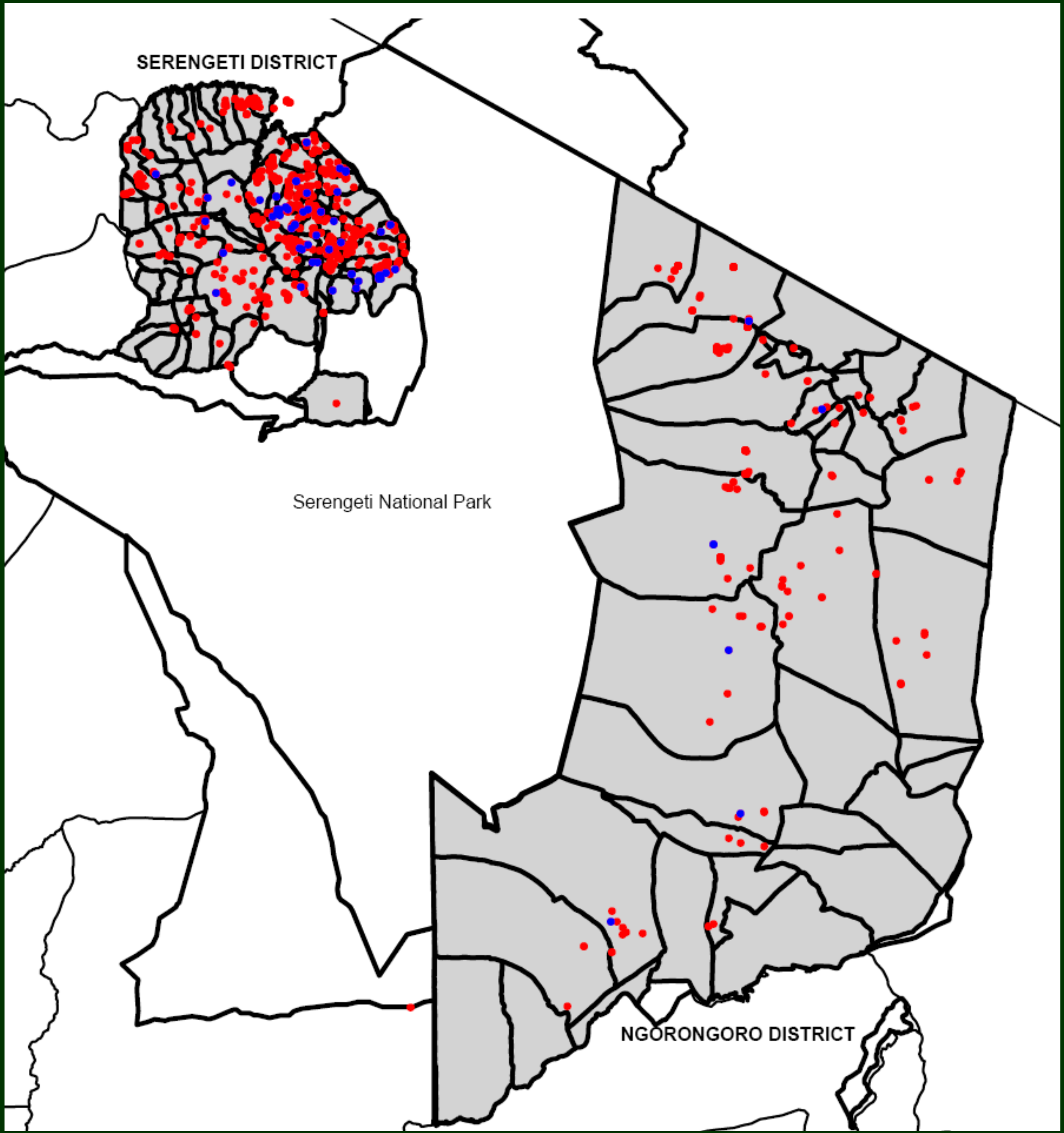
1. Conclusion

- Rabies has been detected persistently in high density domestic dog populations
- Rabies has been detected only sporadically in wildlife populations in SNP

2. Temporal and spatial patterns of dog and wildlife cases

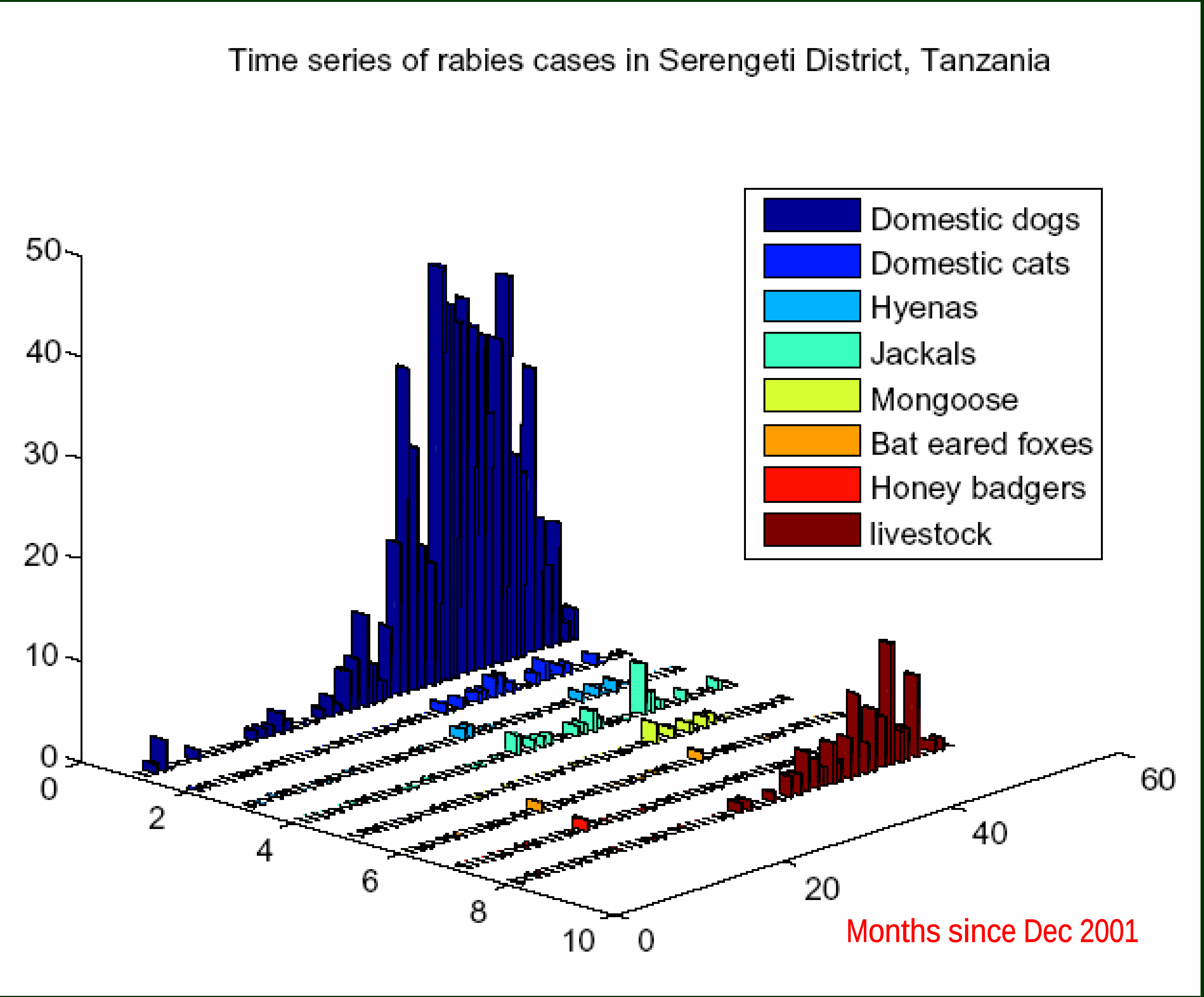
Katie Hampson

Wildlife suspect rabies cases in areas with dog rabies



Domestic dog cases

Wildlife cases

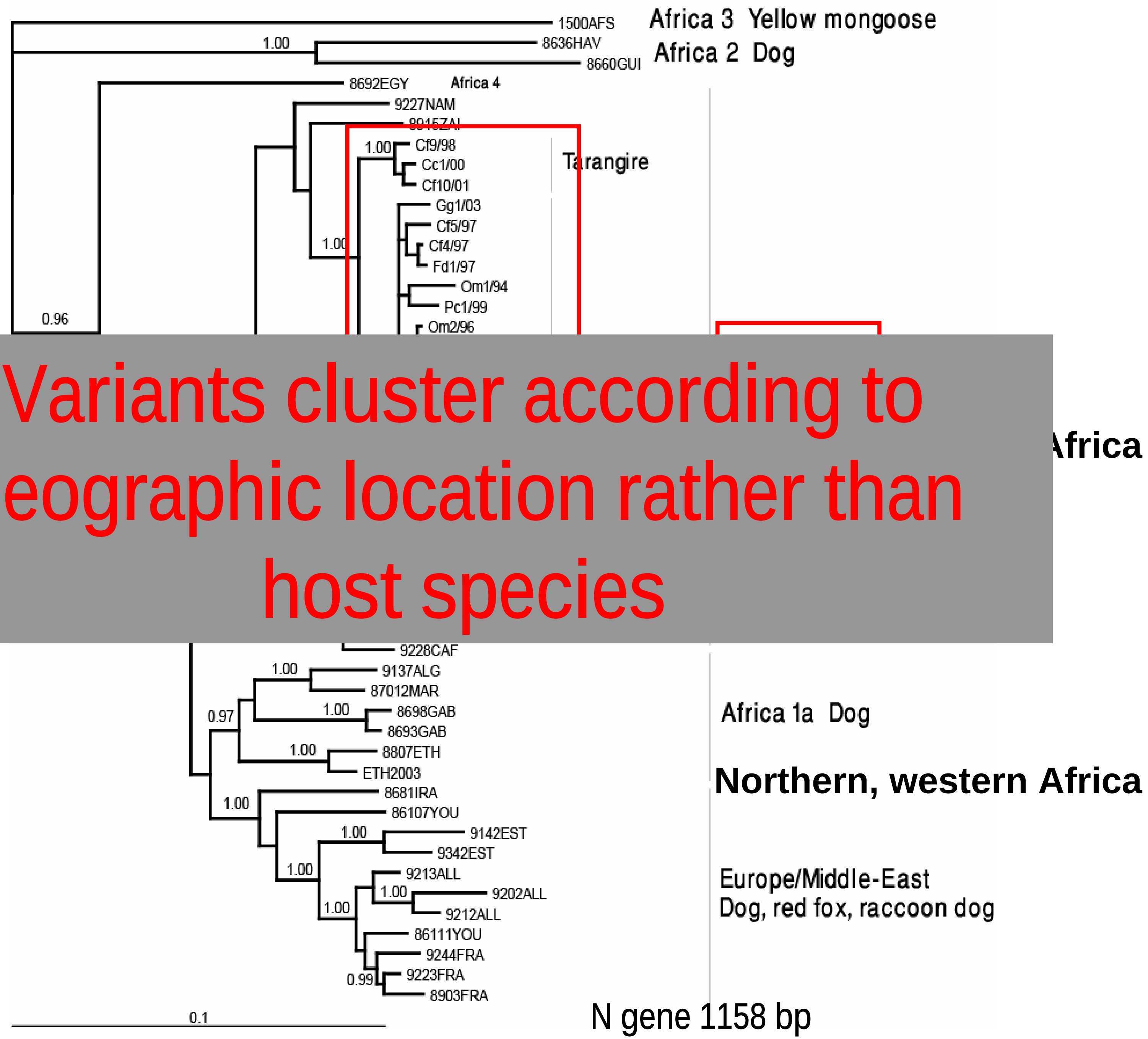

Time series of rabies cases in Serengeti District, Tanzania
Domestic dogs
Domestic cats
Hyenas
Jackals
Mongoose
Bat eared foxes
Honey badgers
livestock
0
10
20
30
40
50
0
2
4
6
8
10
0
20
40
60
Months since Dec 2001

2. Conclusion

- Wildlife rabies has mainly been detected in areas with dog rabies
- Wildlife cases have been clustered in space and time suggesting short-lived chains of transmission following spill-over from domestic dogs

3. Genetic characteristics of viruses isolated from different populations

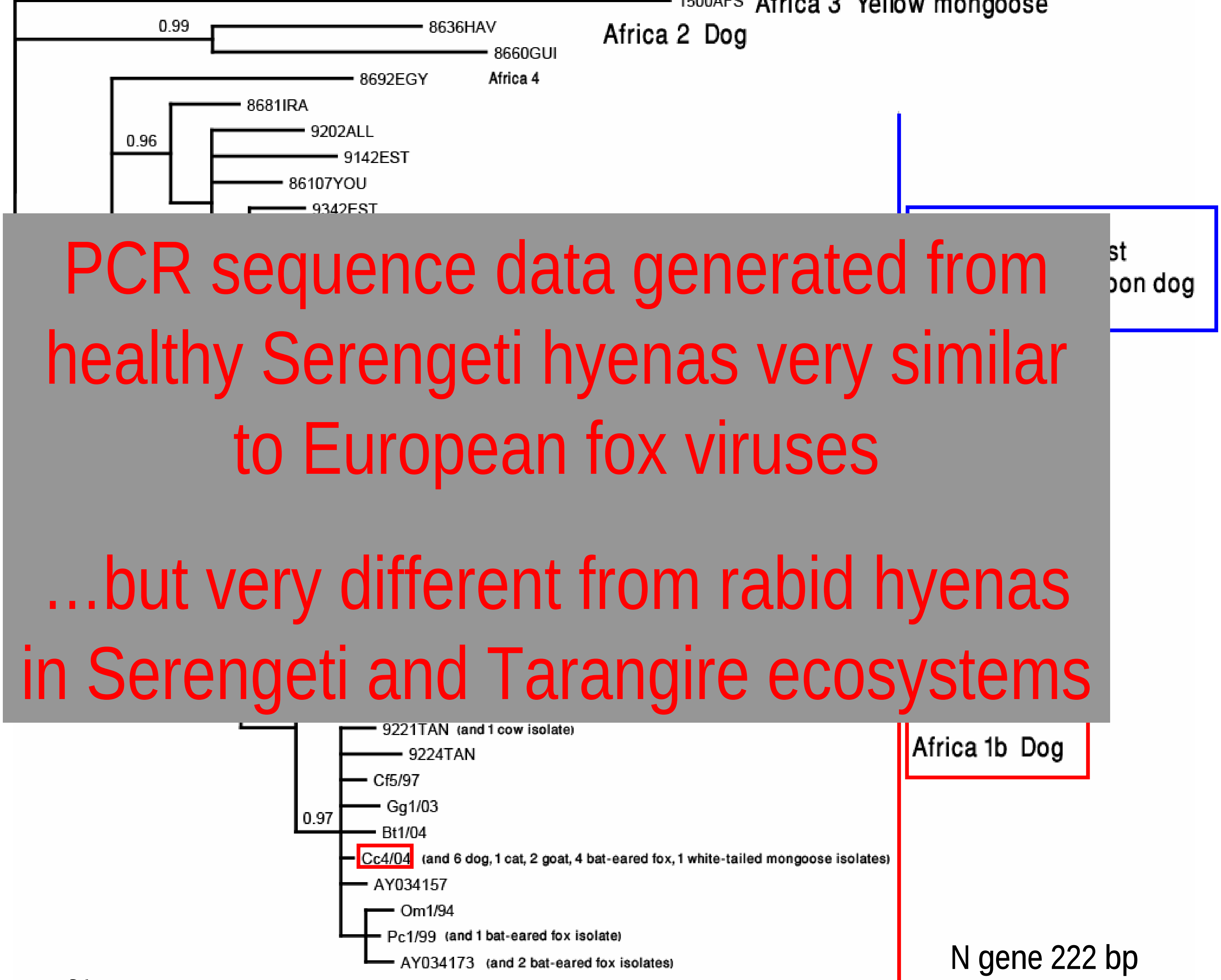
T. Lembo, A. Velasco-Villa, S. Cleaveland, P. Brandão, I.V. Kuzmin, A.R. Fooks, J. Barrat and C. E. Rupprecht



Variants cluster according to geographic location rather than host species

PCR sequence data generated from healthy Serengeti hyenas very similar to European fox viruses

…but very different from rabid hyenas in Serengeti and Tarangire ecosystems



3. Conclusions

Single virus variant (Africa 1b) circulating in multiple species

In any given area, viruses isolated from rabid hyaenas are the same as the viruses isolated from other domestic and wildlife species

Lack of PCR positivity in brains or salivary glands of healthy hyenas in our study

but power sufficient to detect a prevalence of 20% cf 13% reported by East et al. (2001) for brain PCR positives

Confirmation of the carrier state in Serengeti hyenas requires isolation of rabies virus

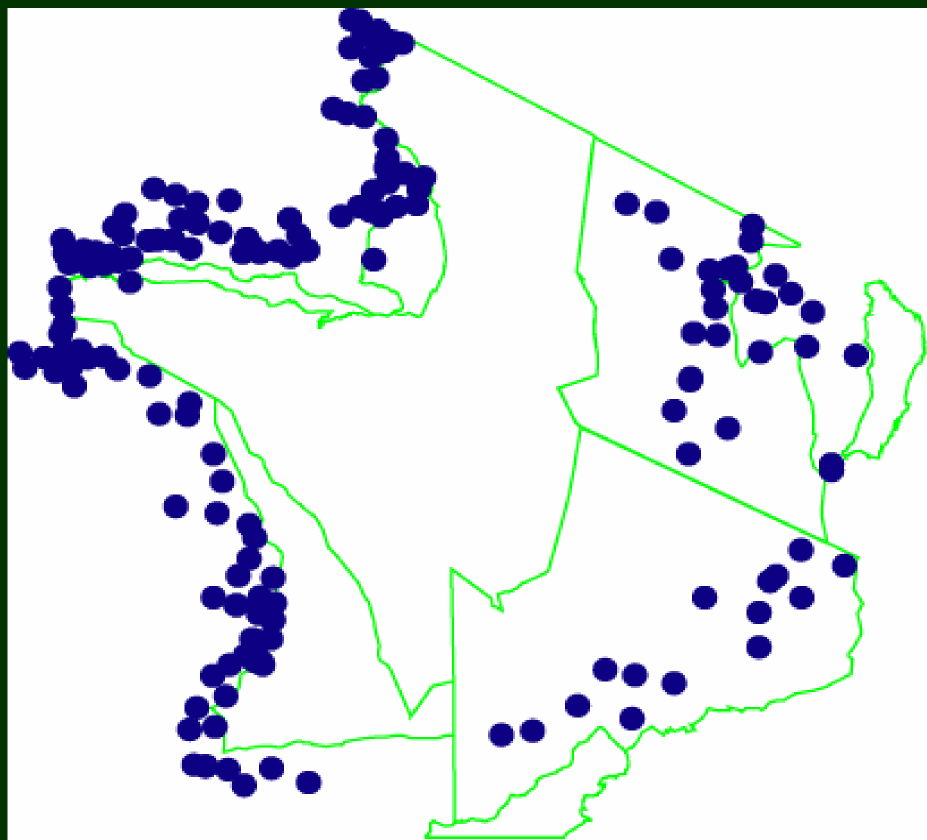
If hyena carriers do exist in the Serengeti, what is the epidemiological significance of an avirulent virus that does not infect other species?

4. Intervention studies

- If dogs only maintenance population in the reservoir, vaccination campaigns targeted at dogs should eliminate rabies in dogs and wildlife

2003 - 24,659 dogs

2004 - 26,000 dogs



2004 - 7,000 dogs

Acknowledgements

- Rabies section, CDC, USA
- Rabies section, VLA, UK
- AFSSA
- Intervet
- TAWIRI
- TANAPA
- COSTECH
- FZS
- TZ ministries
- Local communities
- District councils
- SUA (R.Kazwala)
- District Vet offices, Serengeti, Tarime, Musoma, Bunda, Magu, Bariadi, Meatu
- VIC, Mwanza & Arusha
- Serengeti National Park Staff
- Serengeti lion and cheetah projects
- NIH/NSF
- University of Edinburgh, CTVM

Temporal and Spatial Dynamics of Rabies in Northwest Tanzania

Katie Hampson

Andy Dobson, Sarah Cleaveland, Craig Packer,

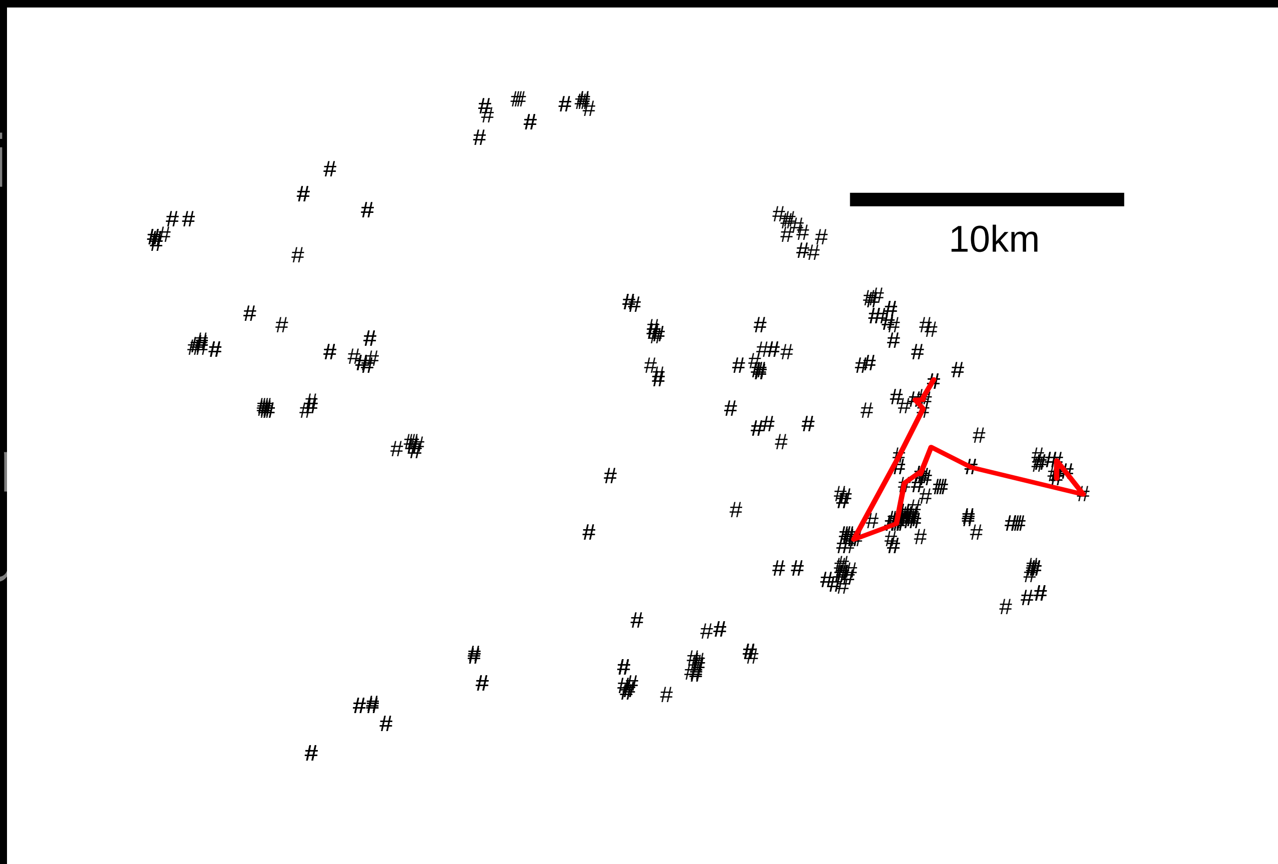
Magai Kaare, Tiziana Lembo, Ernest Eblate, Christine Mentzel, Meggan Craft

METHODOLOGY:

- Veterinary and hospital records
- Surveillance by village livestock officers
- Contact tracing

CONTACT TRACING

- Unique data on behaviour and movement of rabid animals
- Veri ... ance tool
- Esti ... rom natu



CONTACT TRACING

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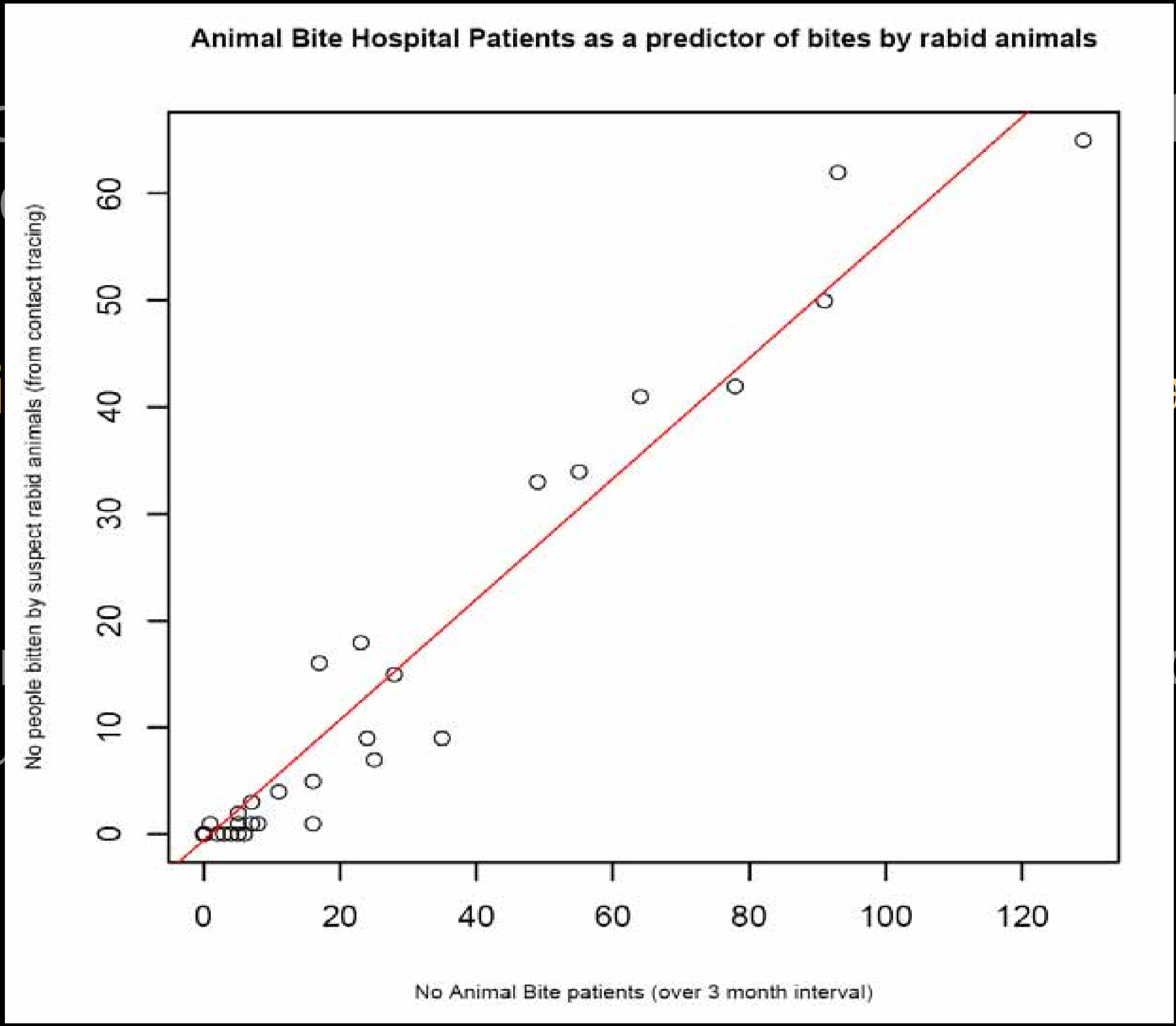
- Veri ... ance tool ...
- Esti ... om natu...



- Un
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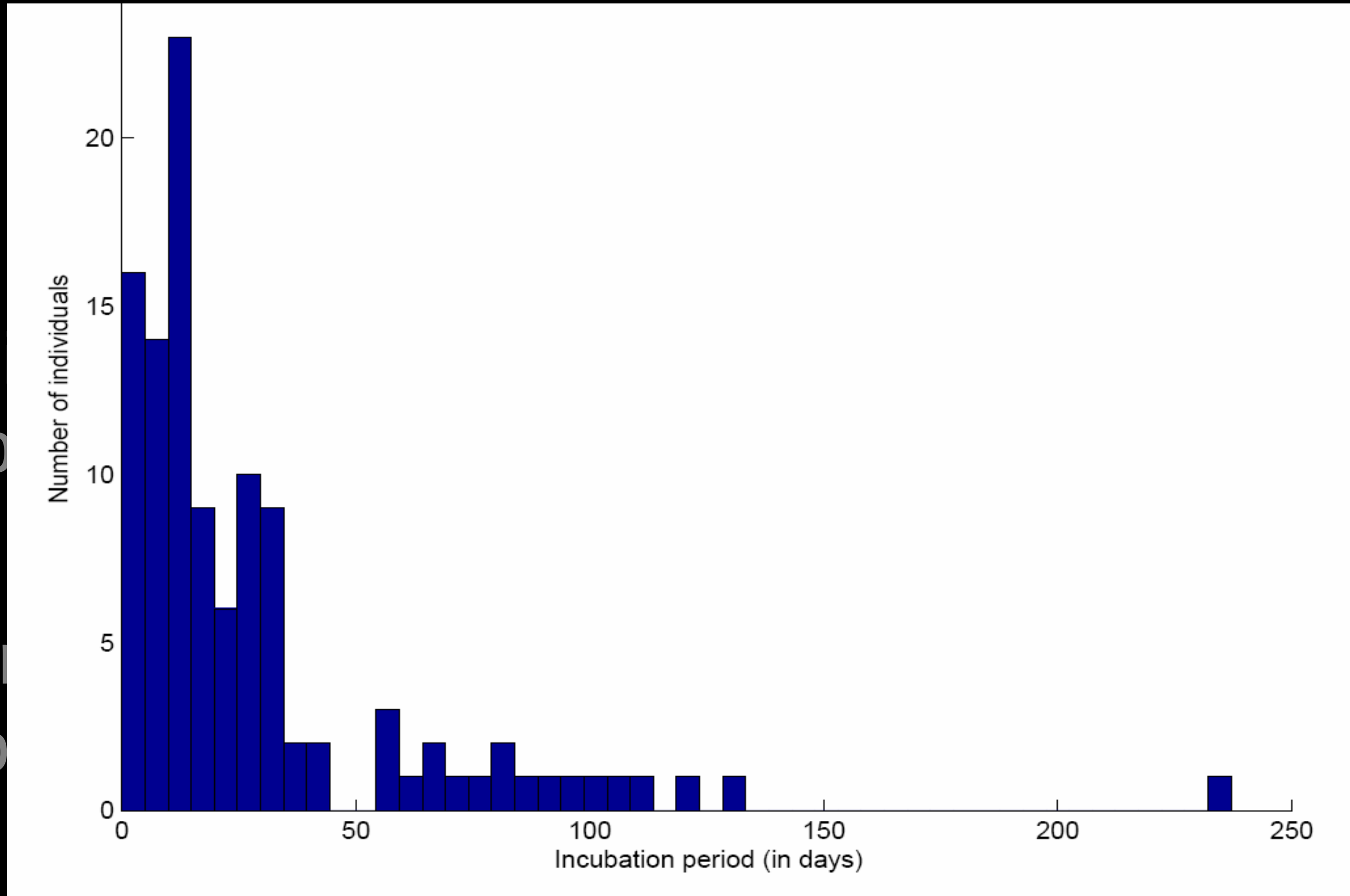


- Estimation of epidemiological parameters from natural infections

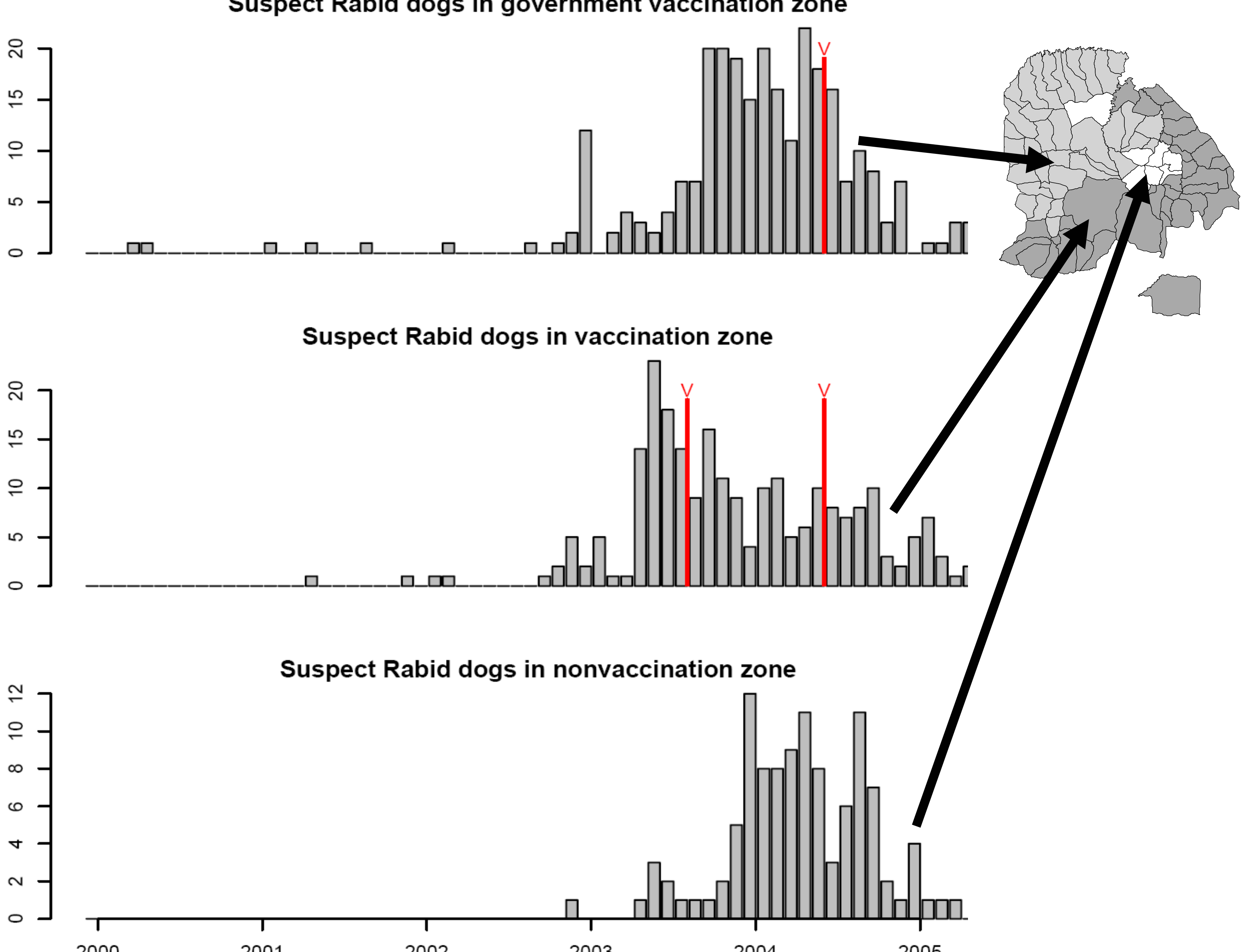
Endemic canine rabies occurs as PERIODIC epidemics

- Epidemic curve is deterministic and predictable

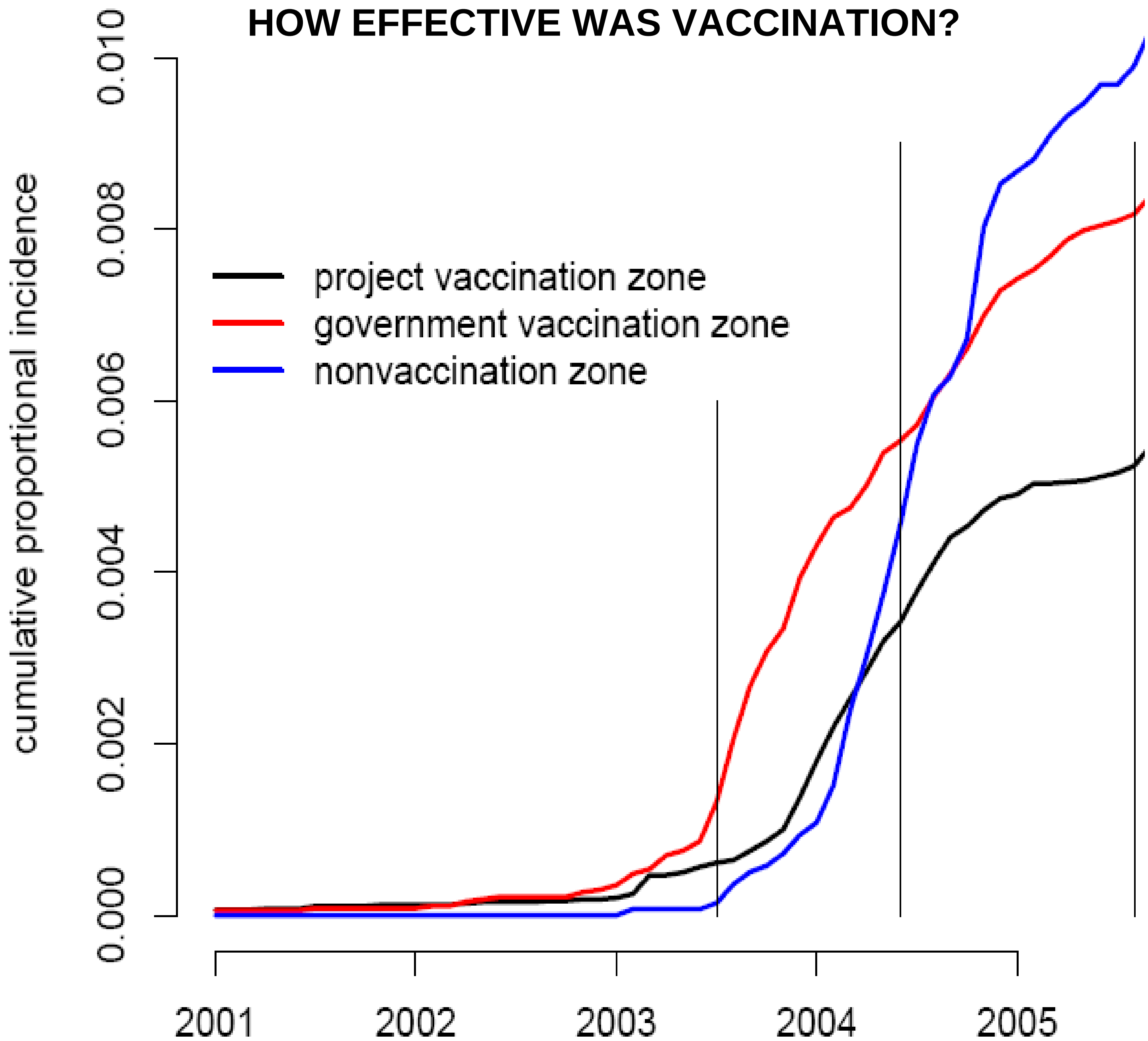
- Exact timing is less predictable (triggered by
- superspreading events?)

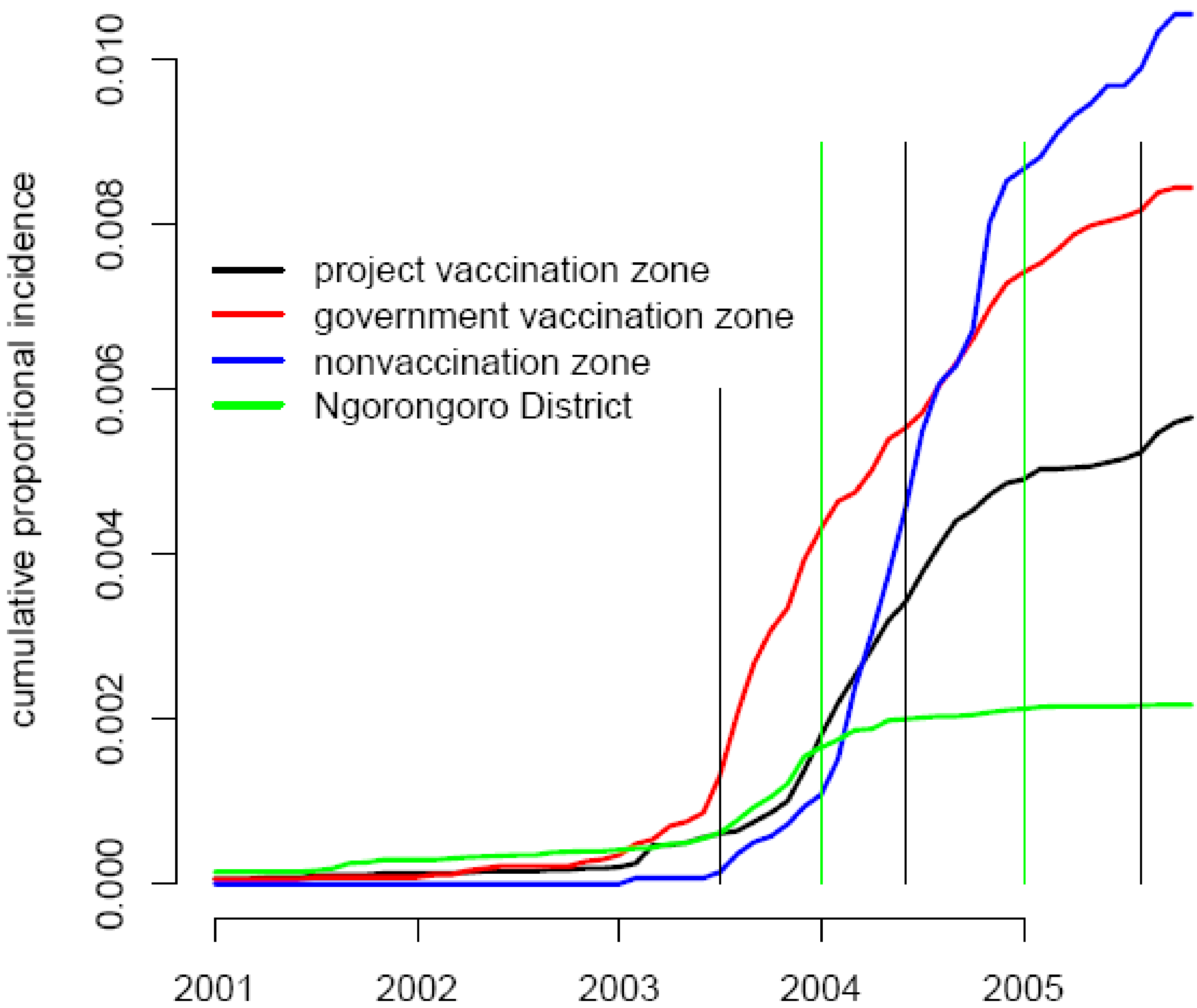
- Epidemics are shorter-lived in lower density populations

- But how have interventions affected the trajectory?



Suspect Rabid dogs in government vaccination zone
Suspect Rabid dogs in vaccination zone
Suspect Rabid dogs in nonvaccination zone
V
V
V
0
5
10
15
20
0
2
4
6
8
10
12
2000
2001
2002
2003
2004
2005


HOW EFFECTIVE WAS VACCINATION?
cumulative proportional incidence
0.000
0.002
0.004
0.006
0.008
0.010
project vaccination zone
government vaccination zone
nonvaccination zone
2001
2002
2003
2004
2005



cumulative proportional incidence
0.000
0.002
0.004
0.006
0.008
0.010
project vaccination zone
government vaccination zone
nonvaccination zone
Ngorongoro District
2001
2002
2003
2004
2005

CONTROL IMPLICATIONS

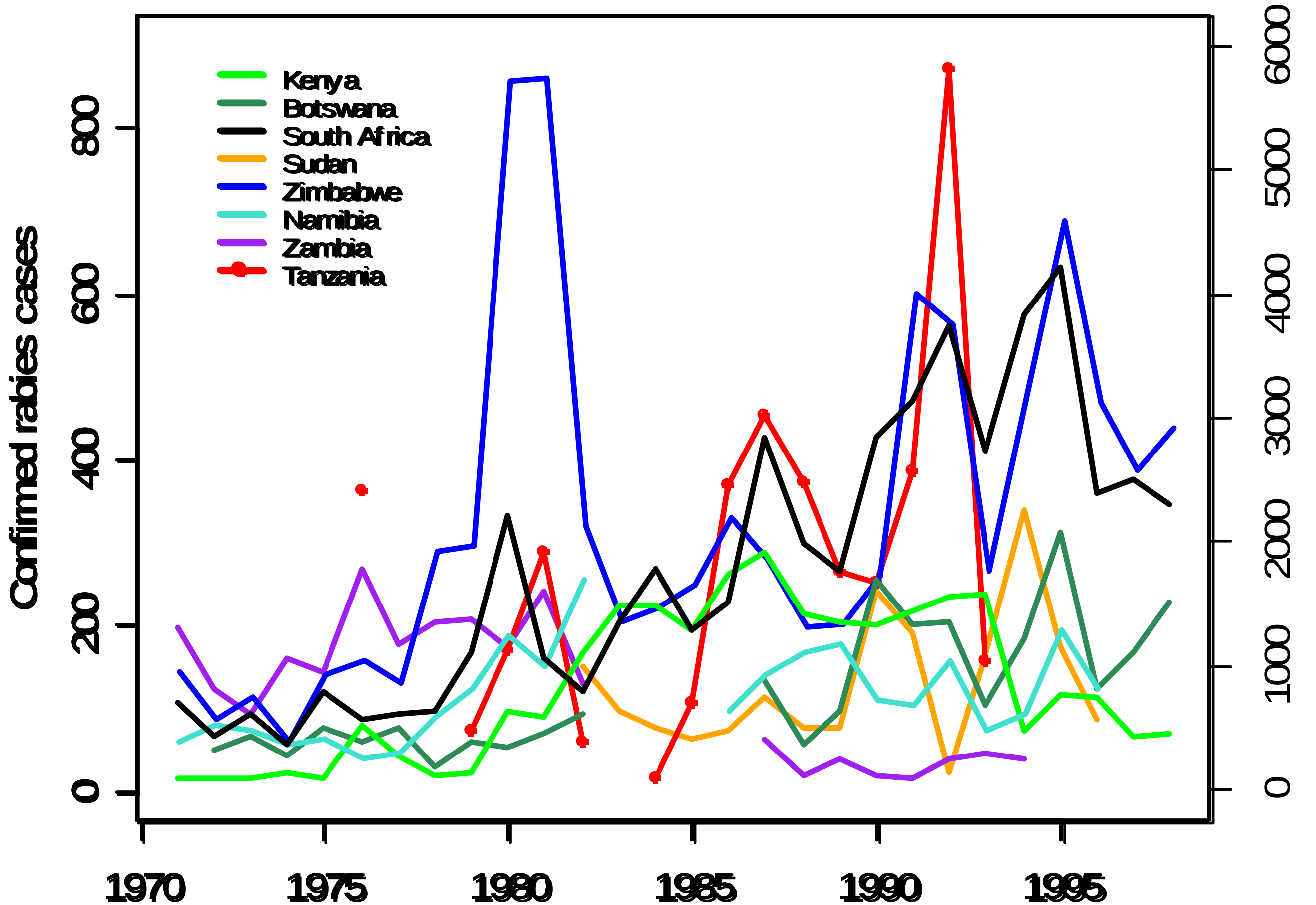
Effectiveness of vaccination dependent upon TIMING and FORCE of infection

Vaccination can reduce incidence but SUSTAINED high coverage is necessary to reduce overall burden of disease

Are epidemics periodic elsewhere in Southern and Eastern Africa?

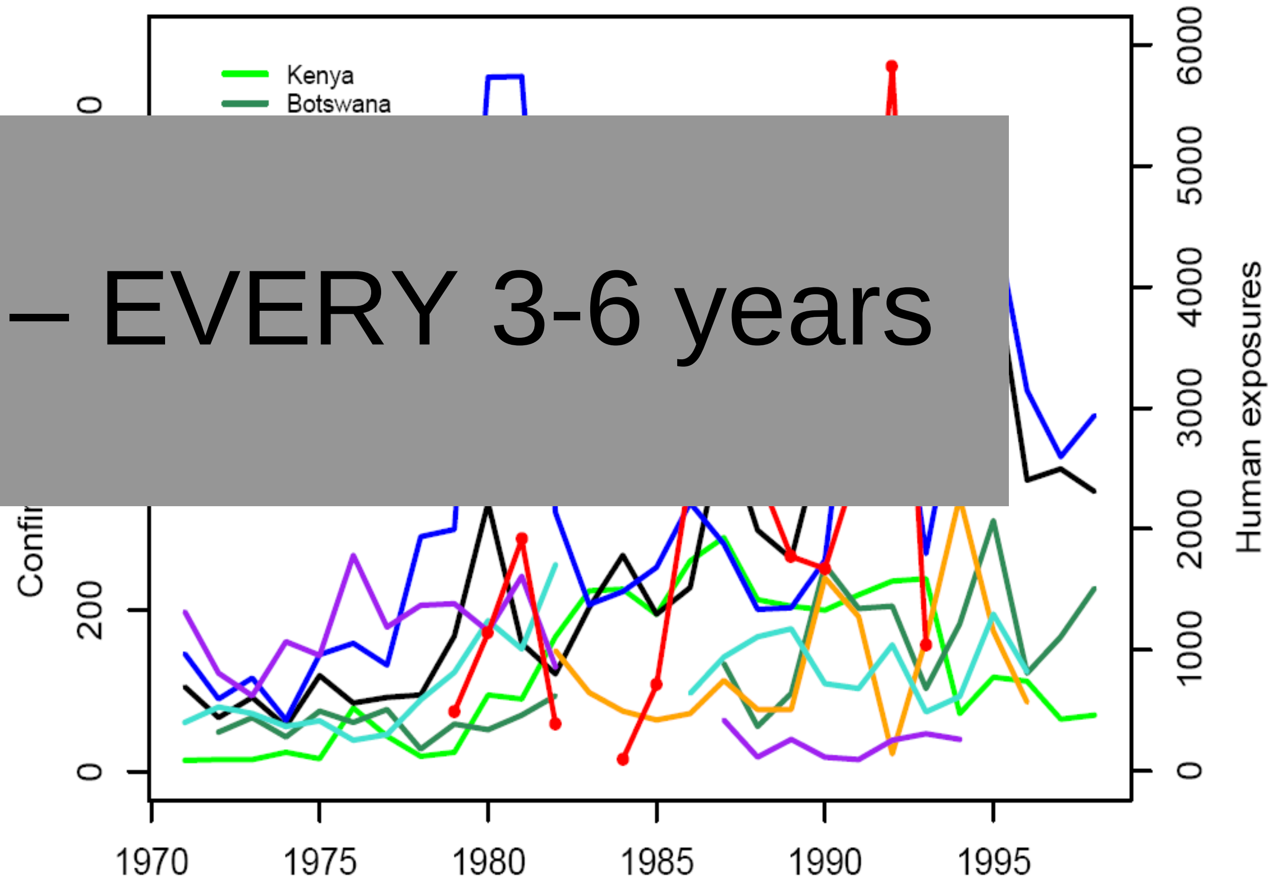
With Jonathan Dushoff, John Bingham, Gideon Brückner

Are epidemics periodic elsewhere in Southern and Eastern Africa?



With Jonathan Dushoff, John Bingham, Gideon Brückner

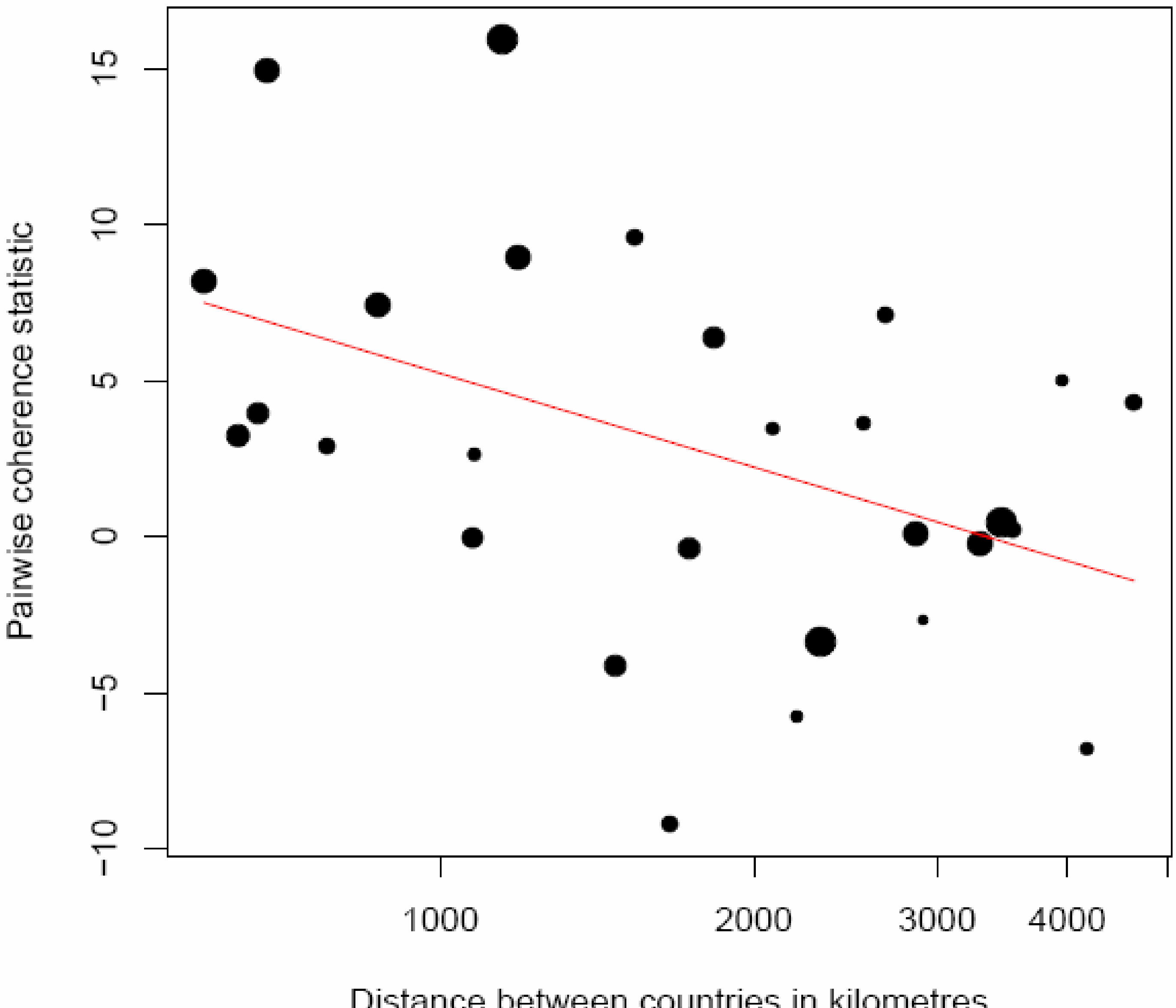
Are epidemics periodic elsewhere in Southern and Eastern Africa?



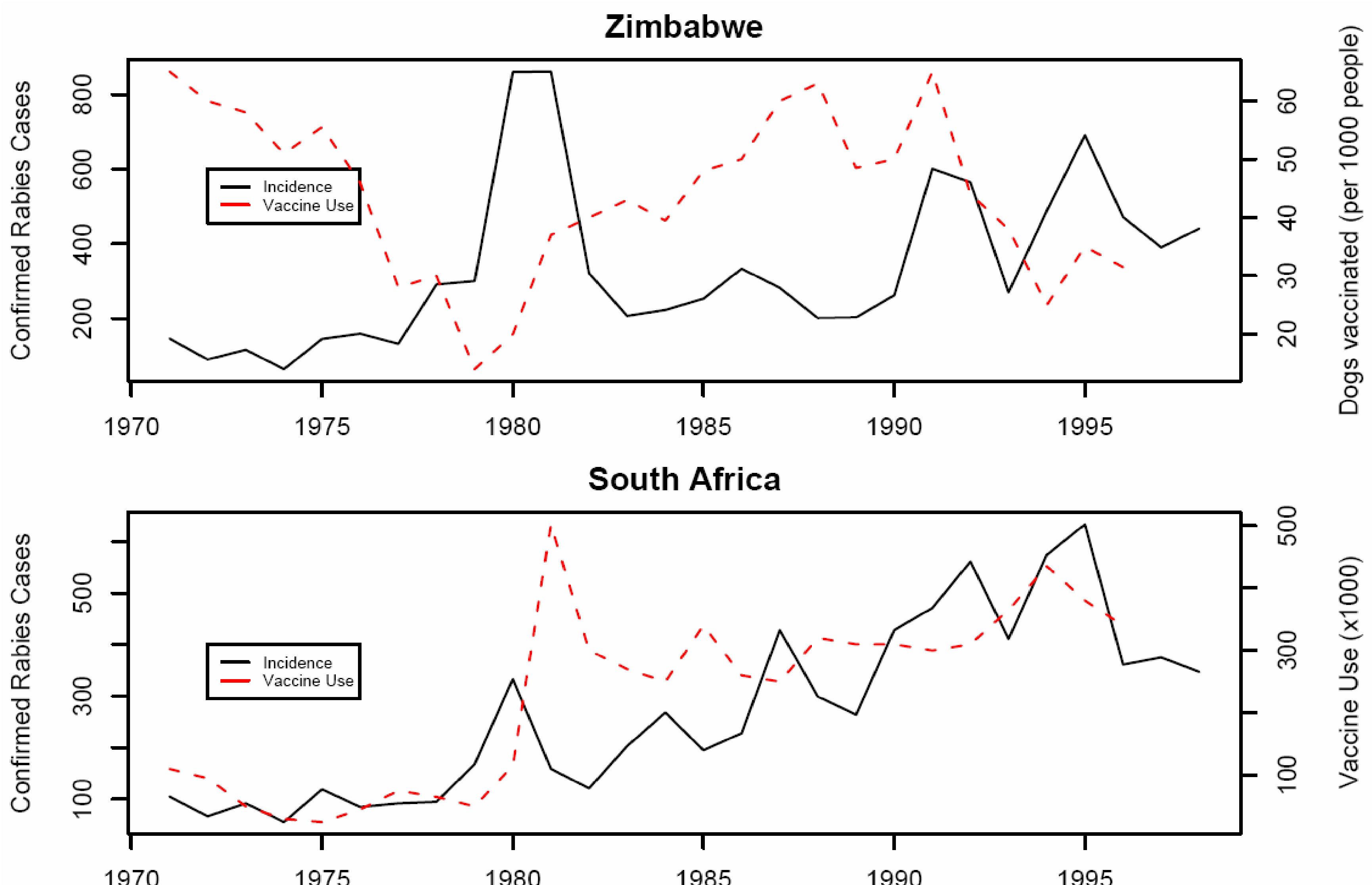
YES – EVERY 3-6 years

With Jonathan Dushoff, John Bingham, Gideon Brückner

- Epidemics are largely synchronised
- Degree of synchrony declines with distance



Interaction between control measures and disease dynamics



CONCLUSIONS:

- Periodic outbreaks may be driven by interactions between disease dynamics and social responses
- Synchrony could result from small amounts of dispersal between local populations
- Implications for predicting and preventing outbreaks
- Preliminary analyses suggest striking large-scale patterns with important practical consequences, but potential for much more investigation

THANKS!

THANKS!

Dynamics and control in NW Tanzania:

Mathias Magoto, Emmanuel Sindoya and the CDP team
Local communities, district councils, livestock offices and hospital staff, Ministry of Water and Livestock Development, Ministry of Regional administration and Local Governments, Ministry of Health
FZS, TAWIRI, TANAPA
Jonathon Dushoff, Simon Levin, Burt Singer, Dan Haydon, Daniel Bennett, Julie Pulliam and the disease cabal

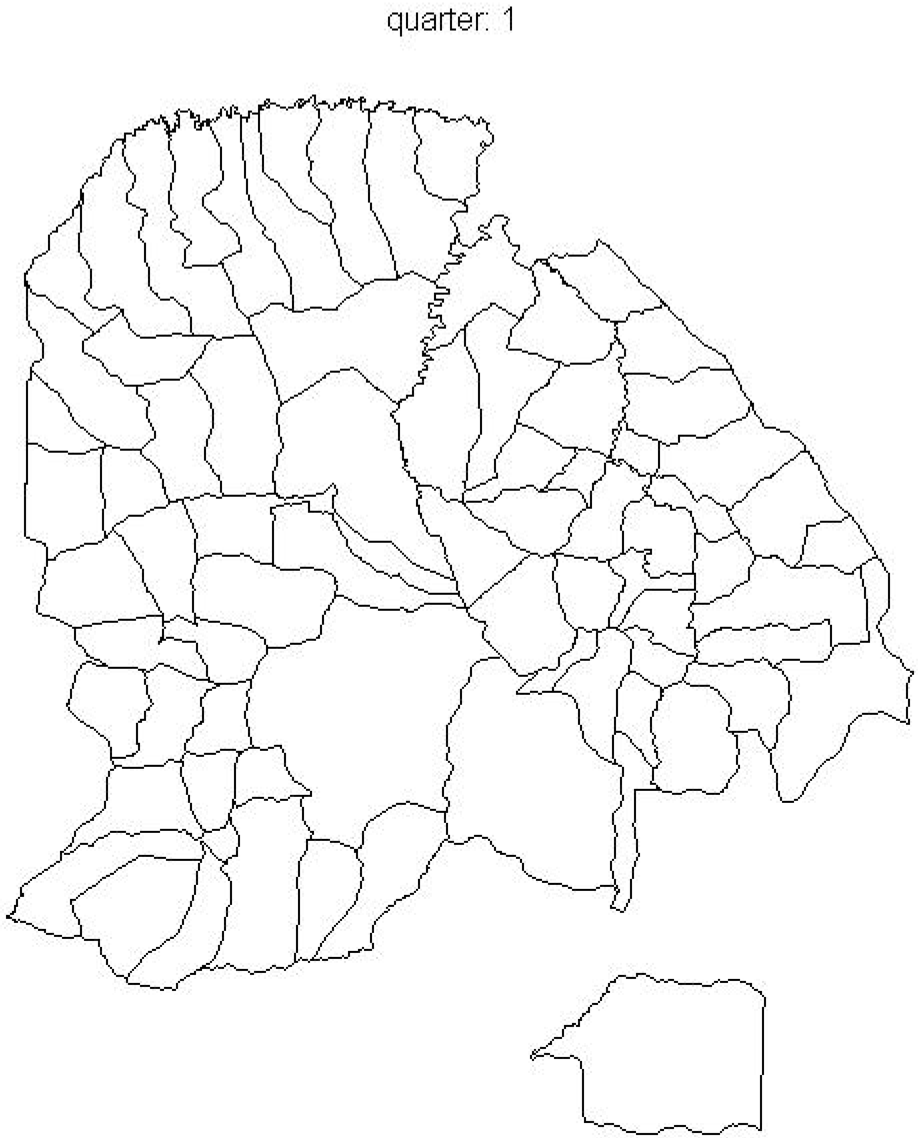
Large scale analyses:

Jonathan Dushoff, John Bingham, Gideon Brückner

FUNDING:

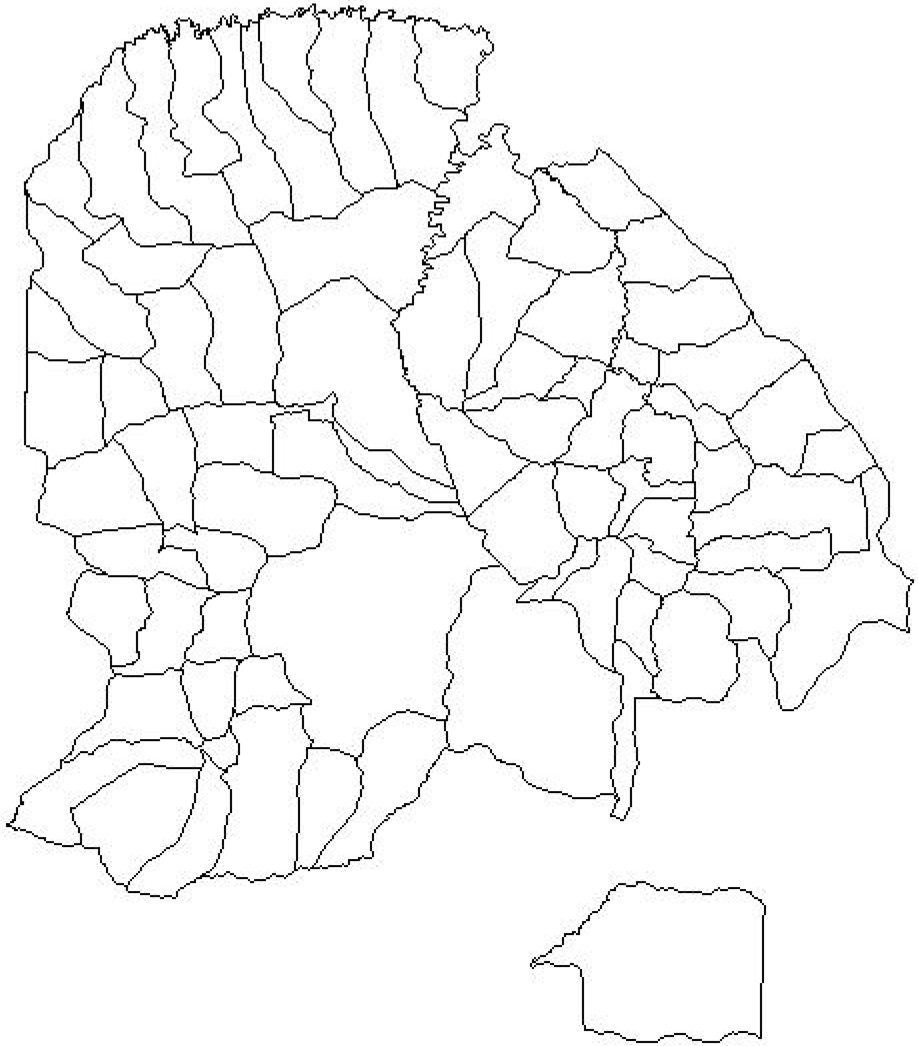
NSF, NIH, The Pew Foundation, The Teresa Heinz Foundation, The Centre for Health and Wellbeing and EEB, Princeton University, Glasgow University.

2001

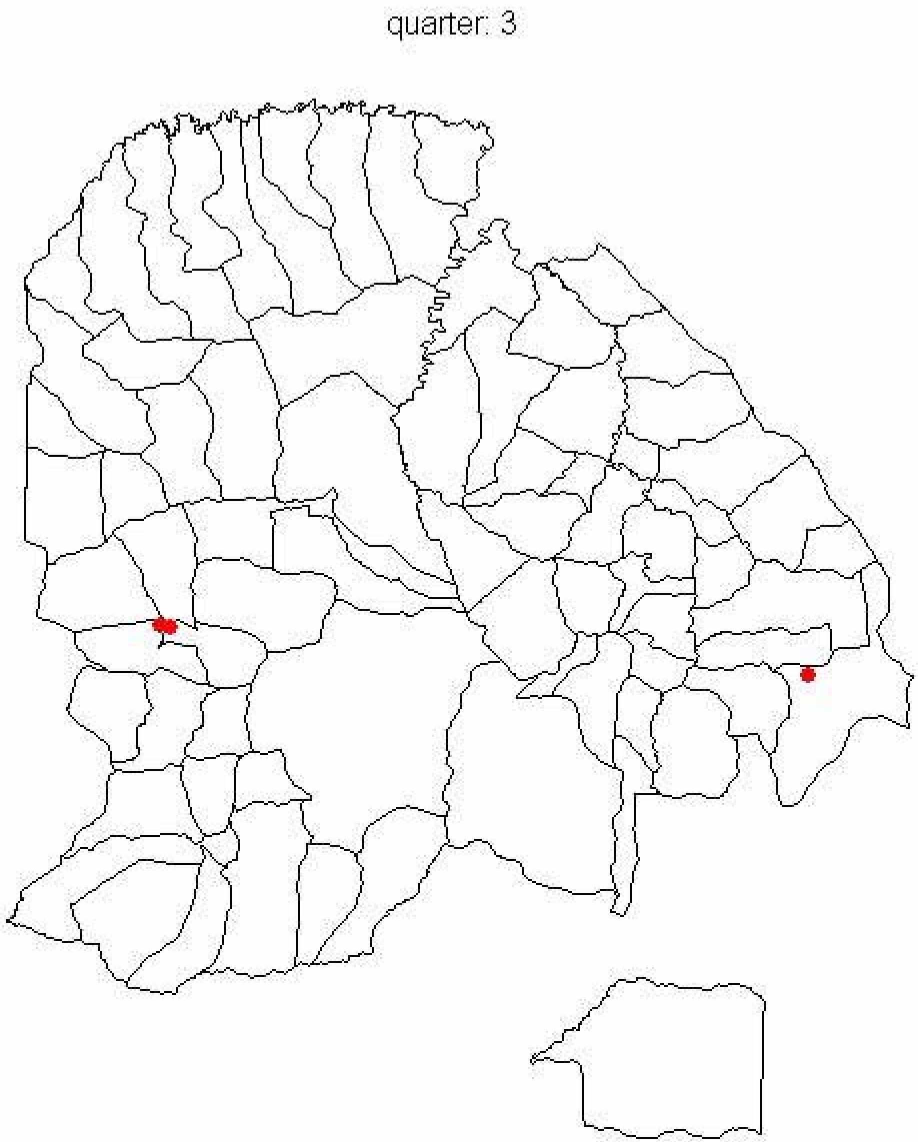


2001

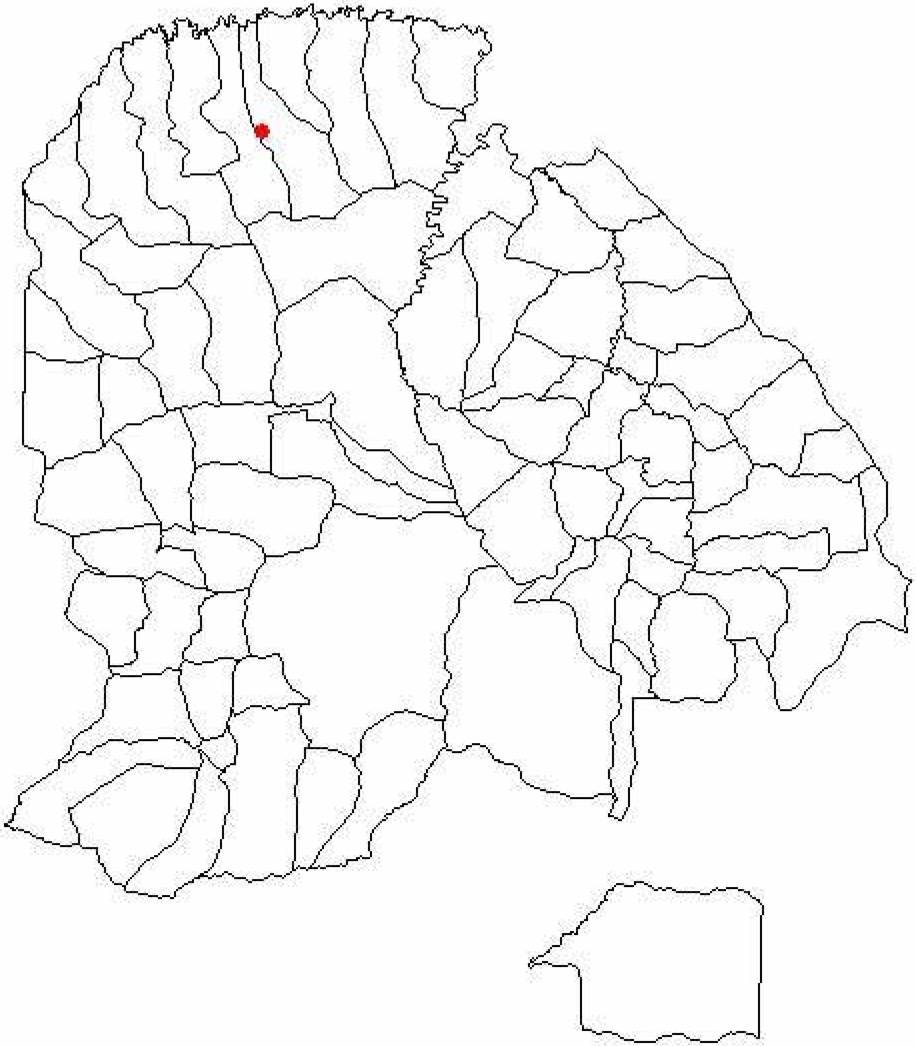
quarter: 2



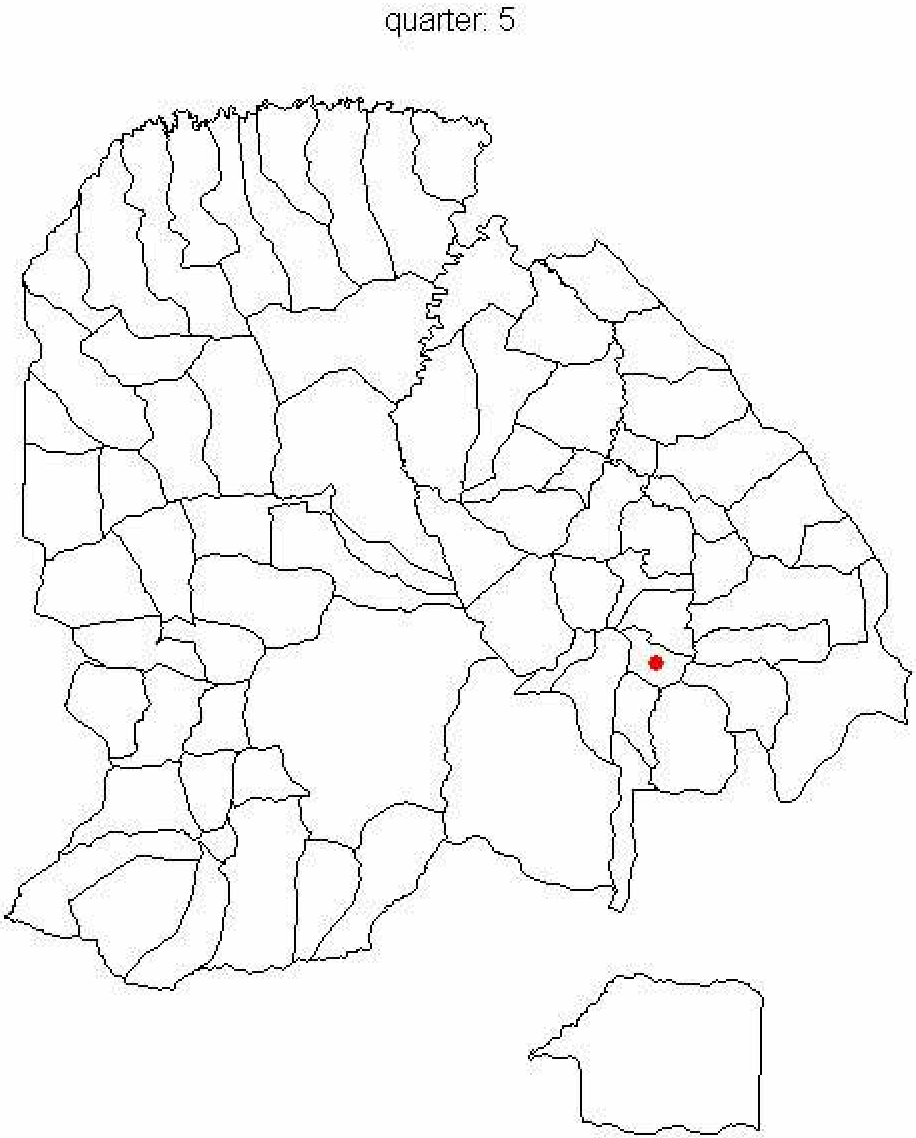
2001



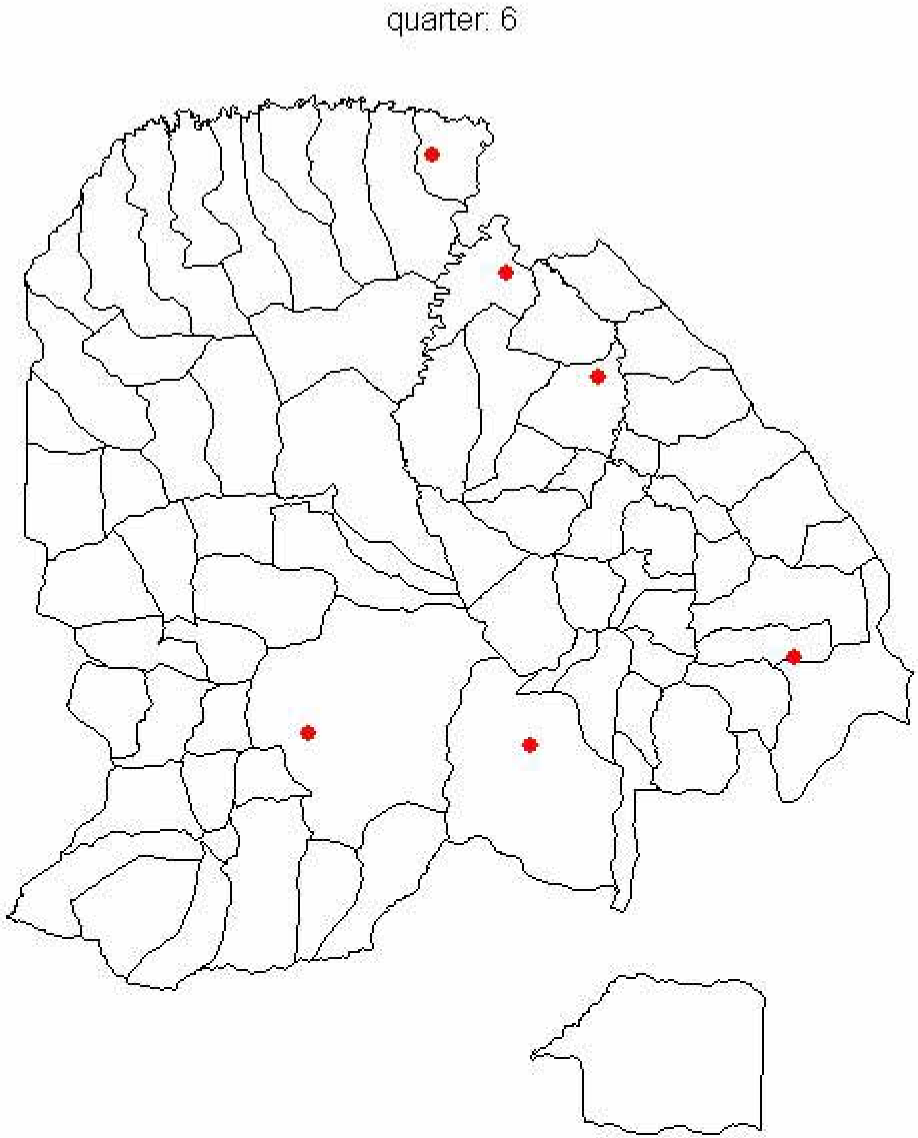
2001



2002

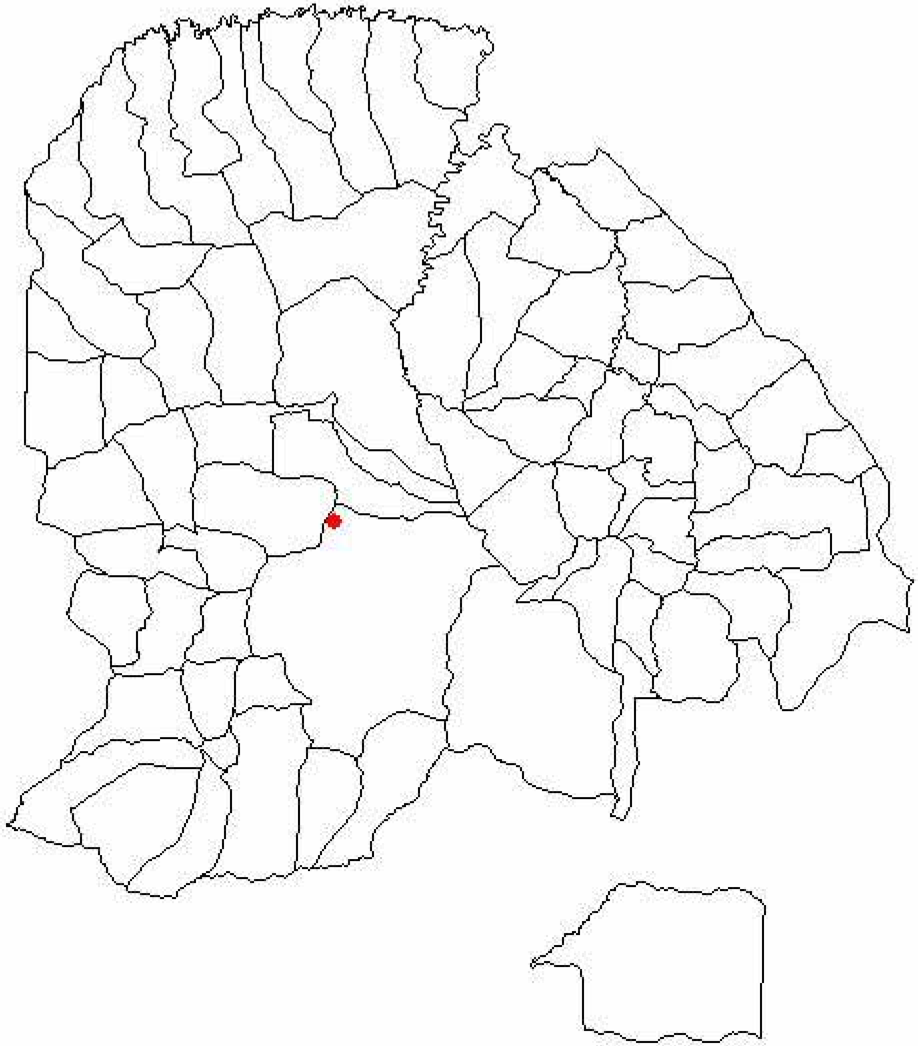


2002



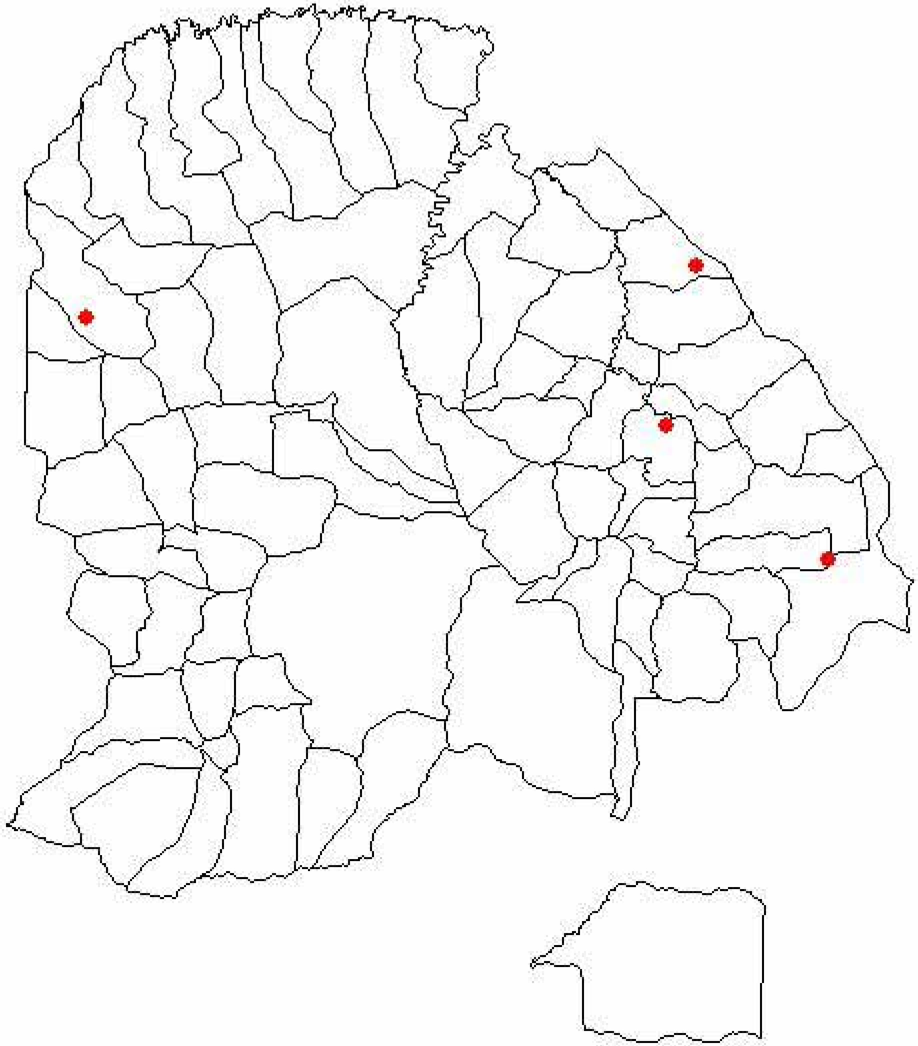
2002

quarter: 7

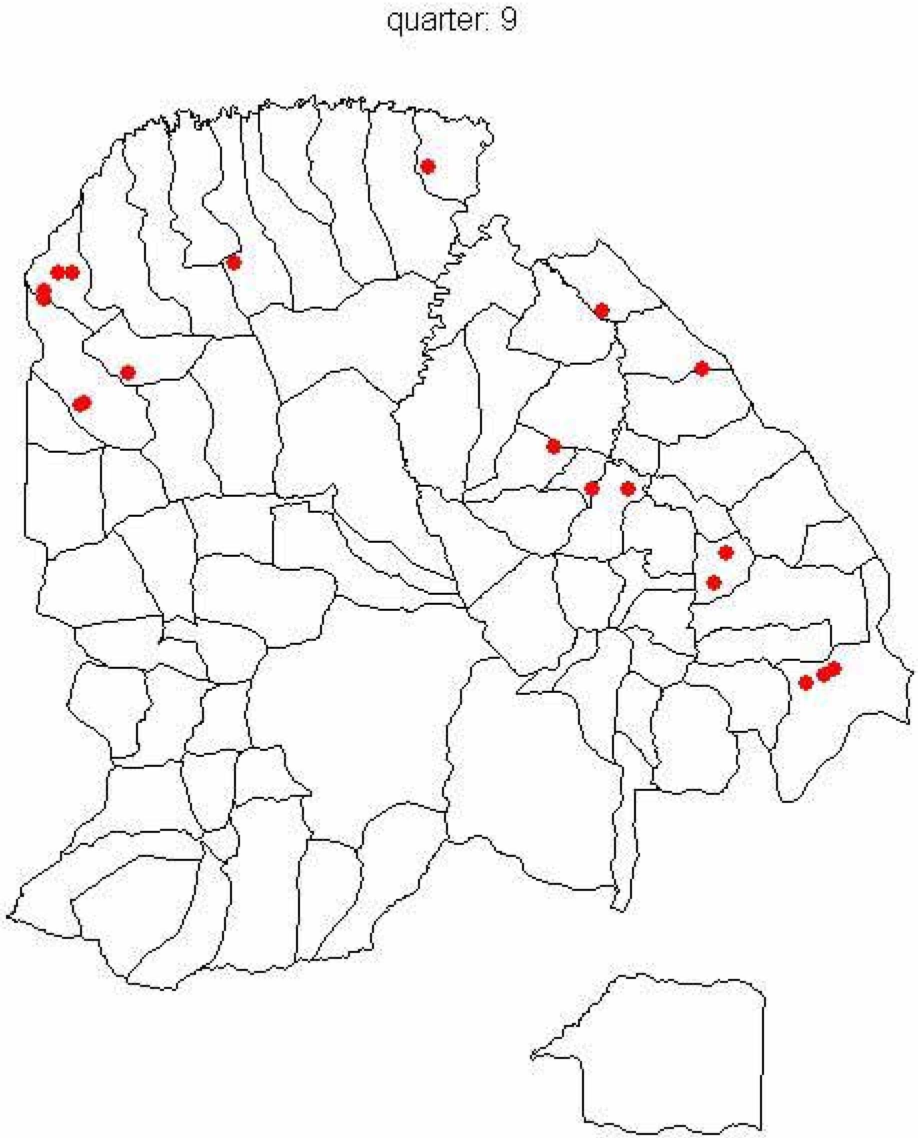


2002

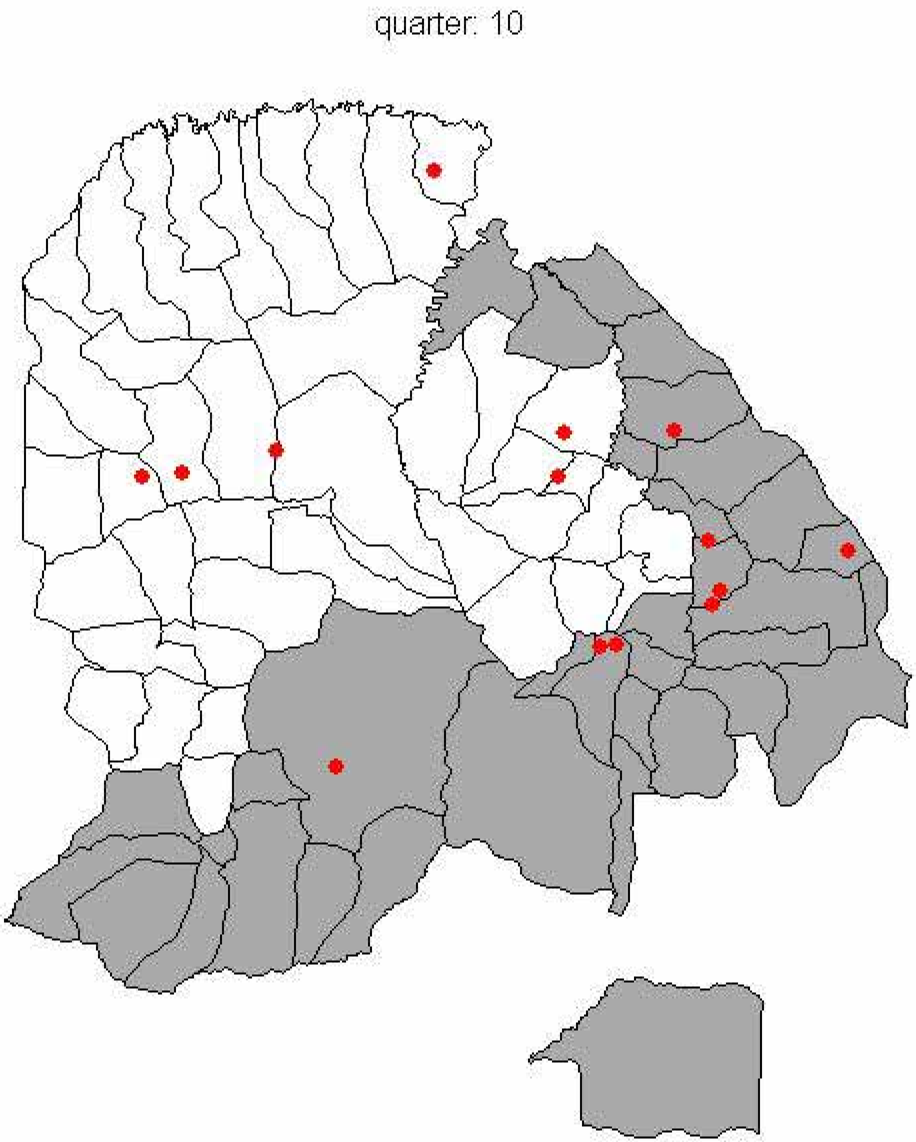
quarter: 8



2003

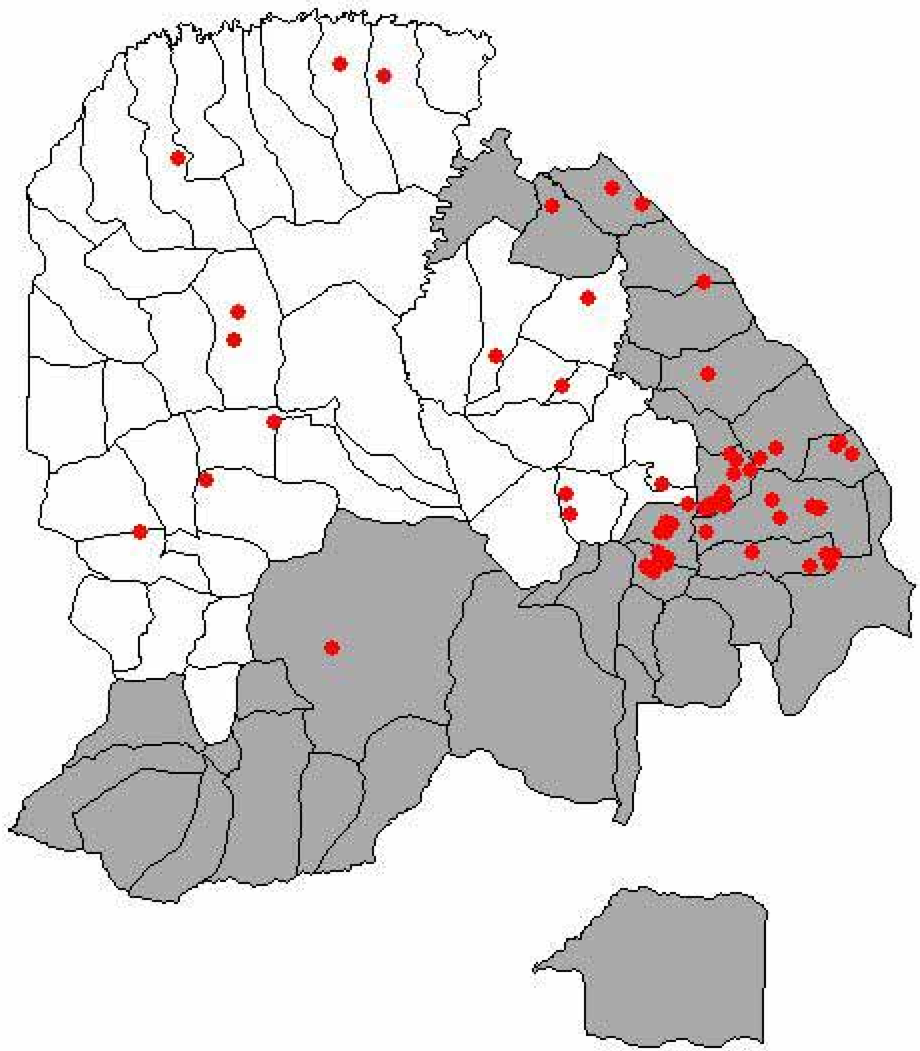


2003

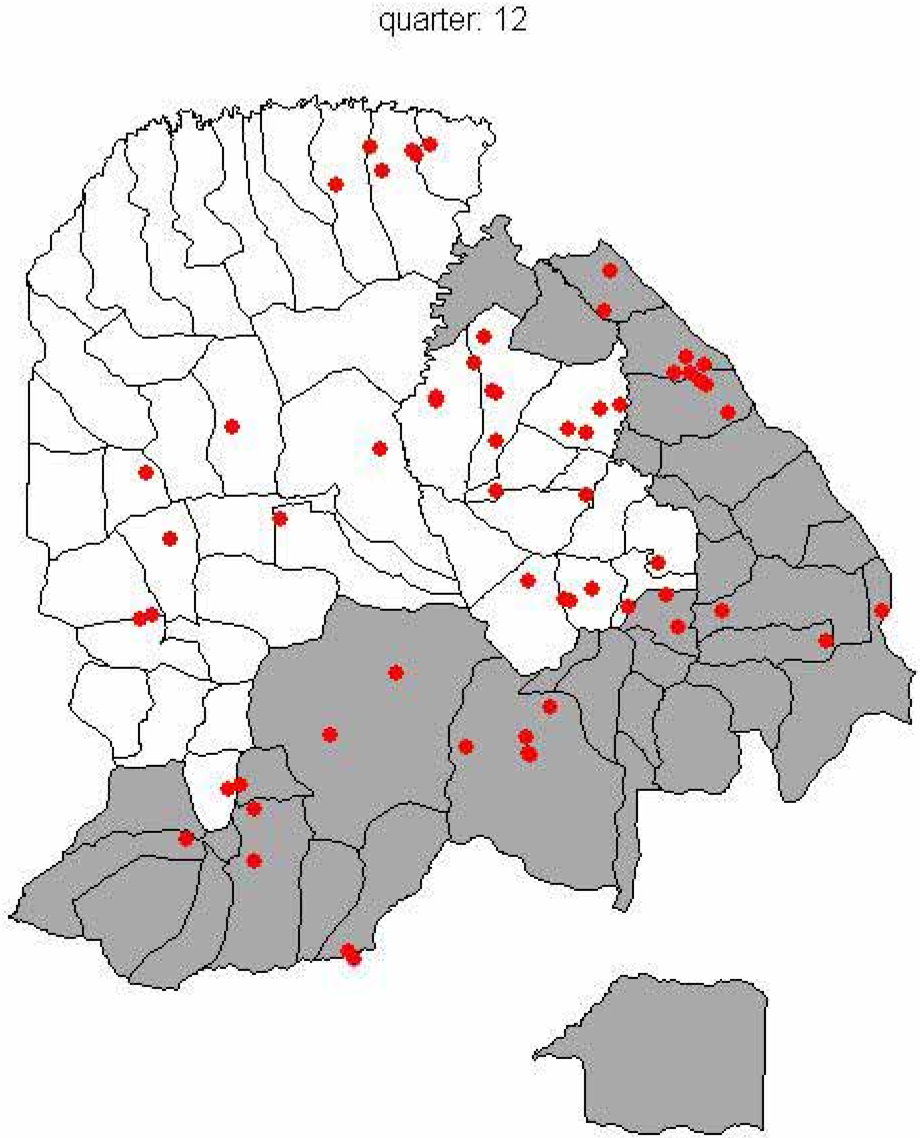


2003

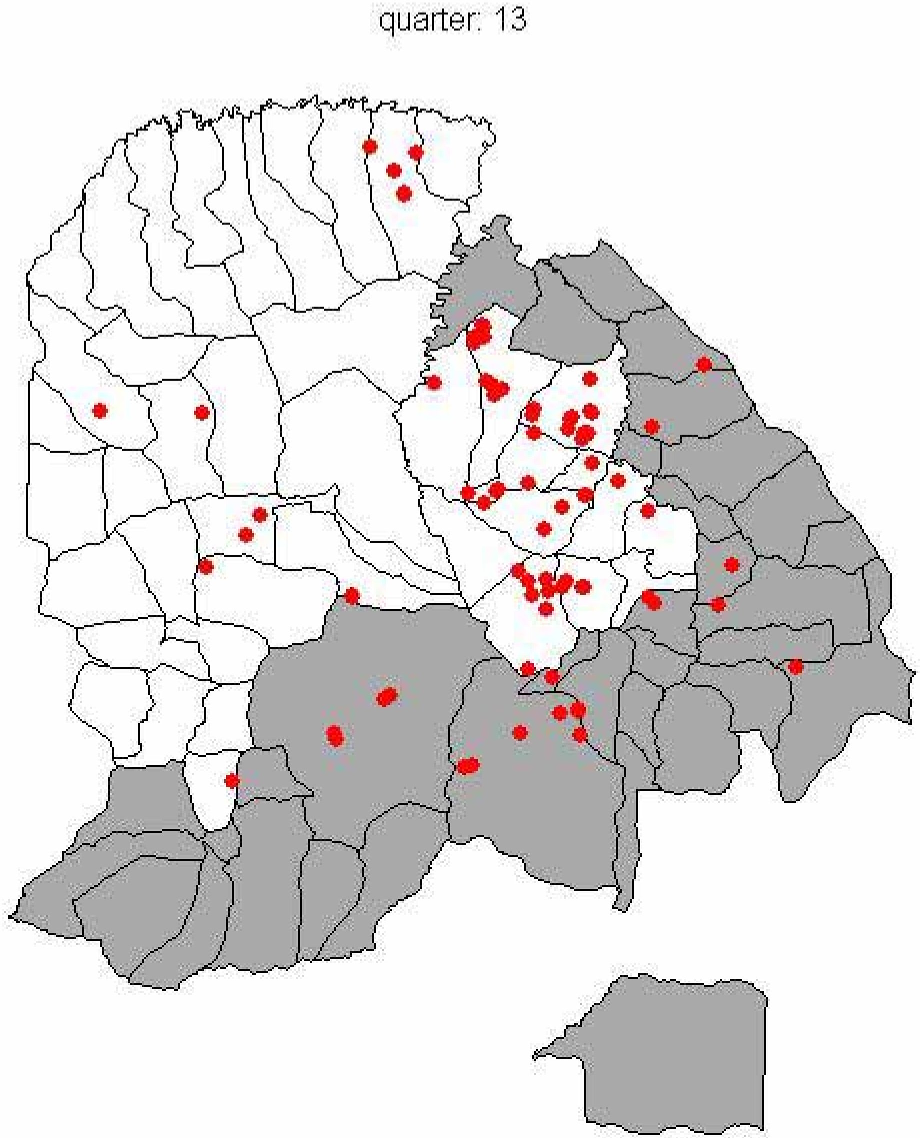
quarter: 11



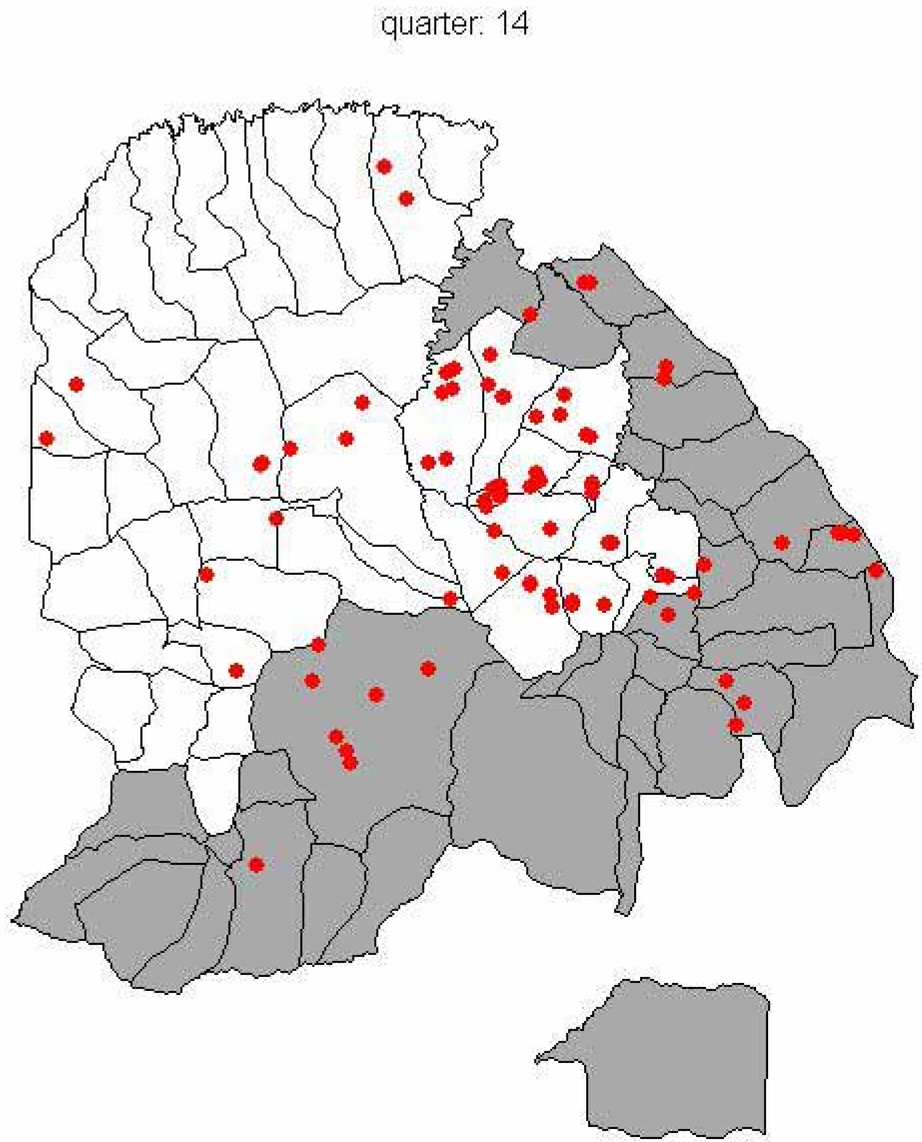
2003



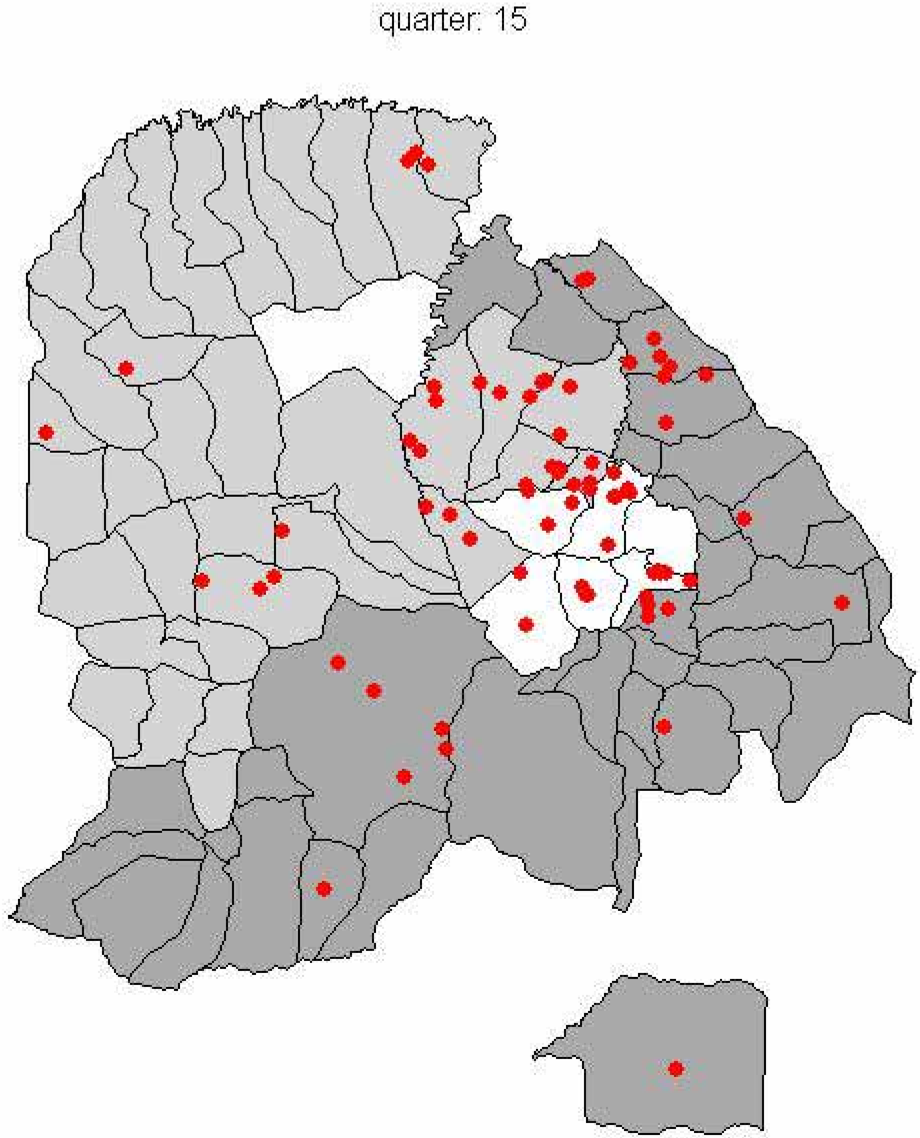
2004



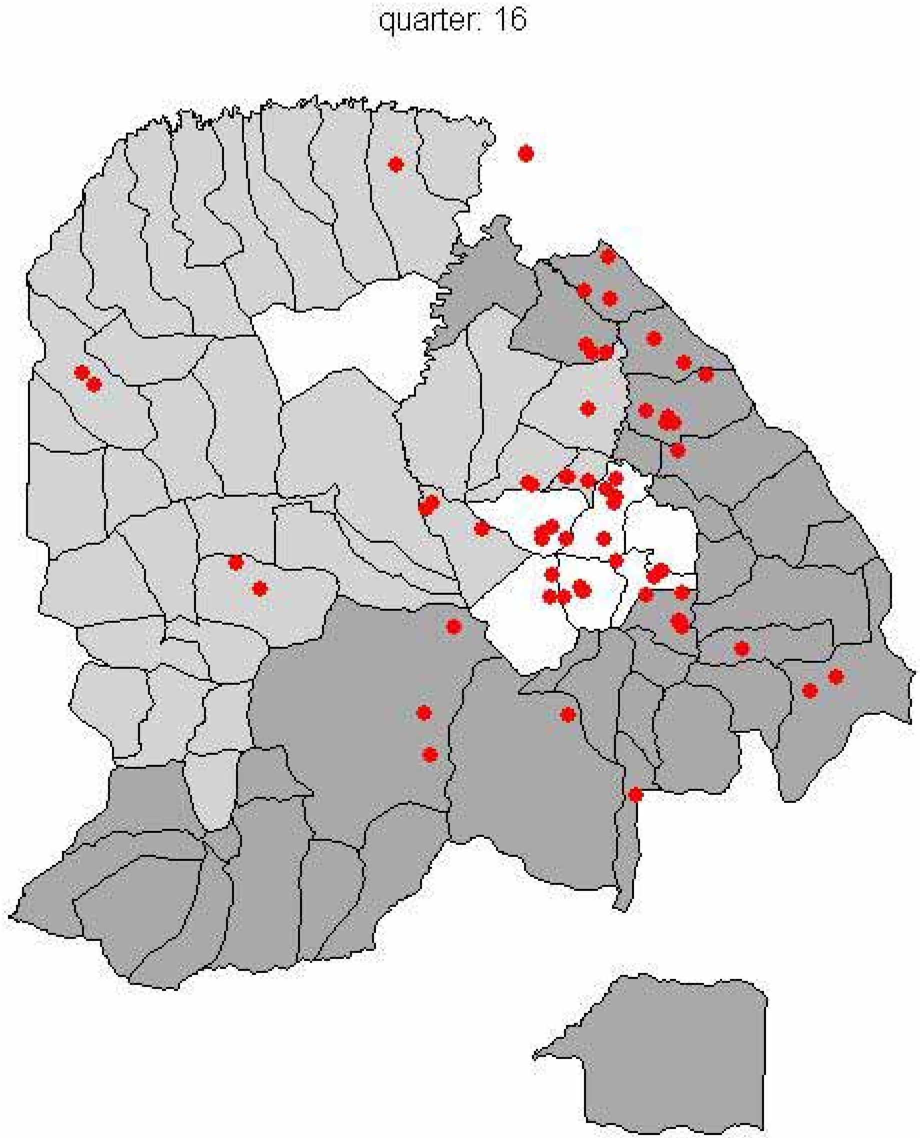
2004



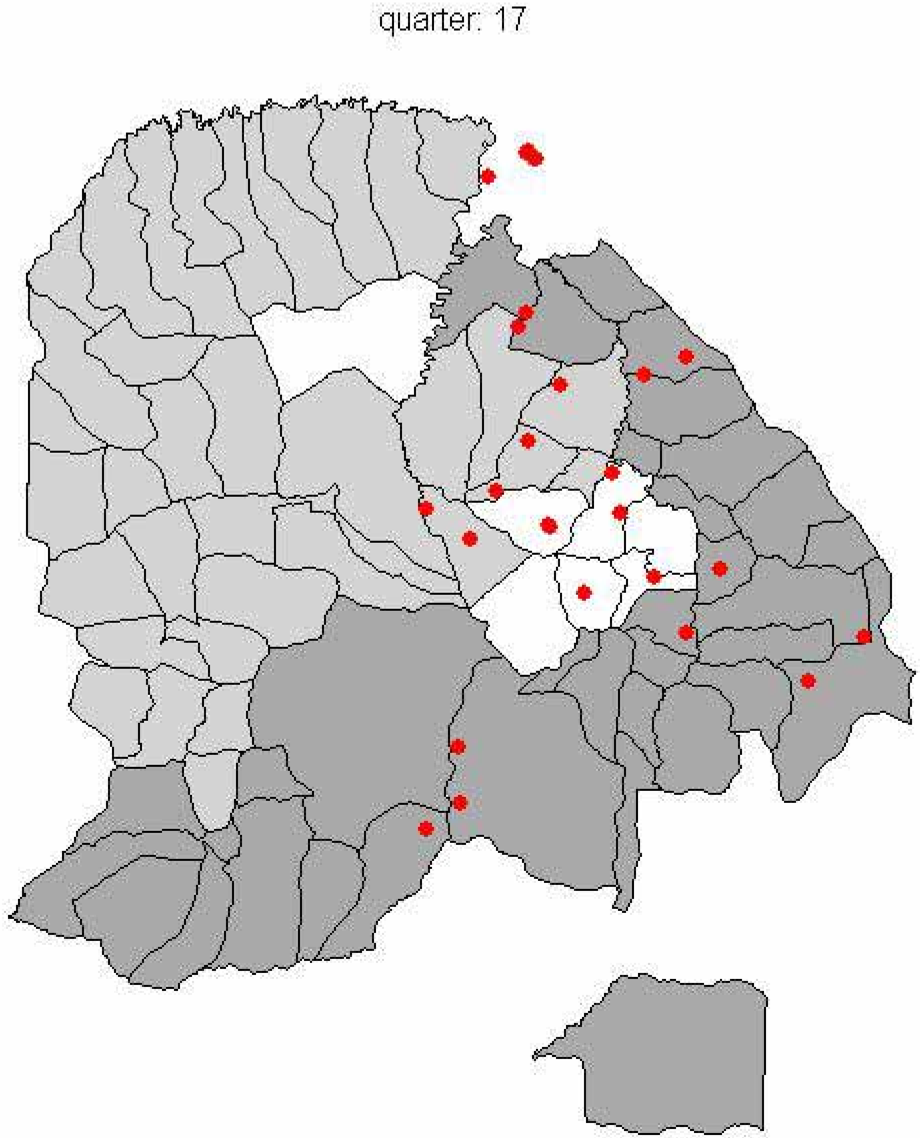
2004



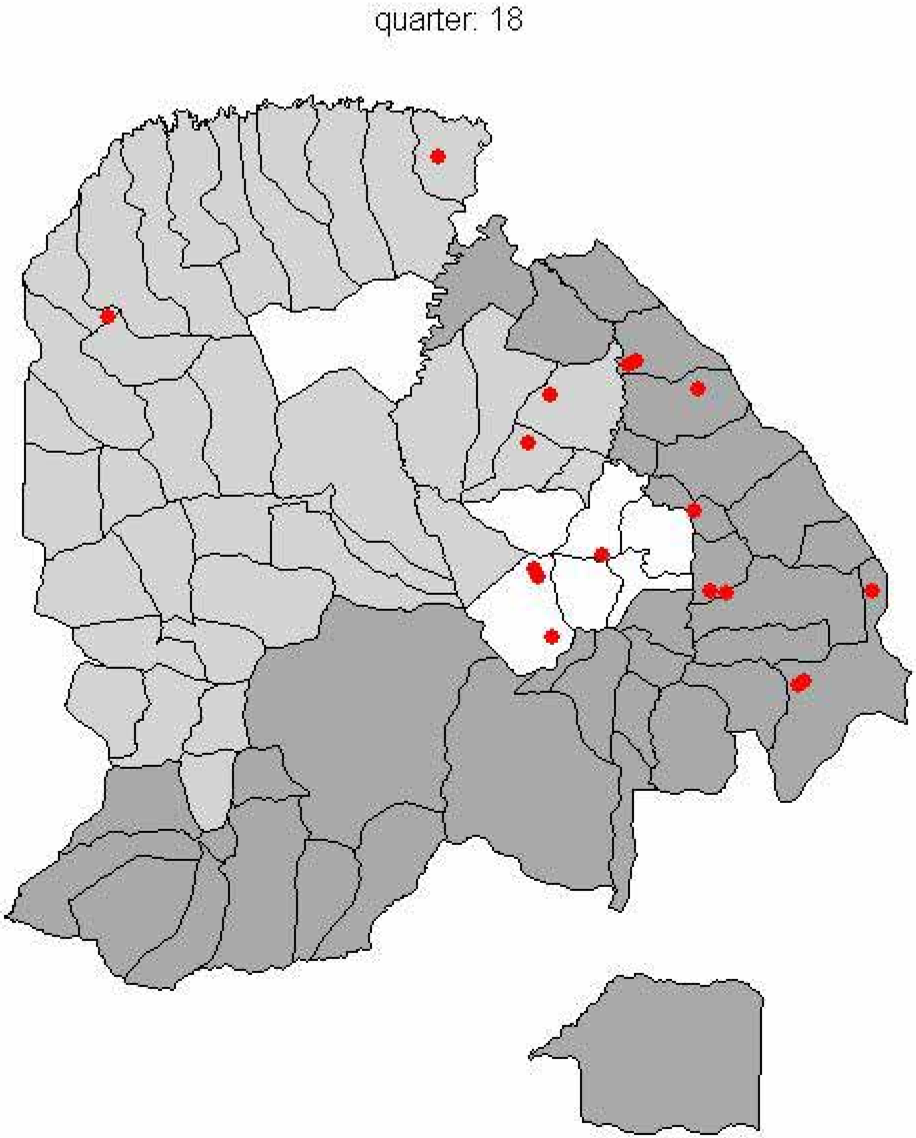
2004



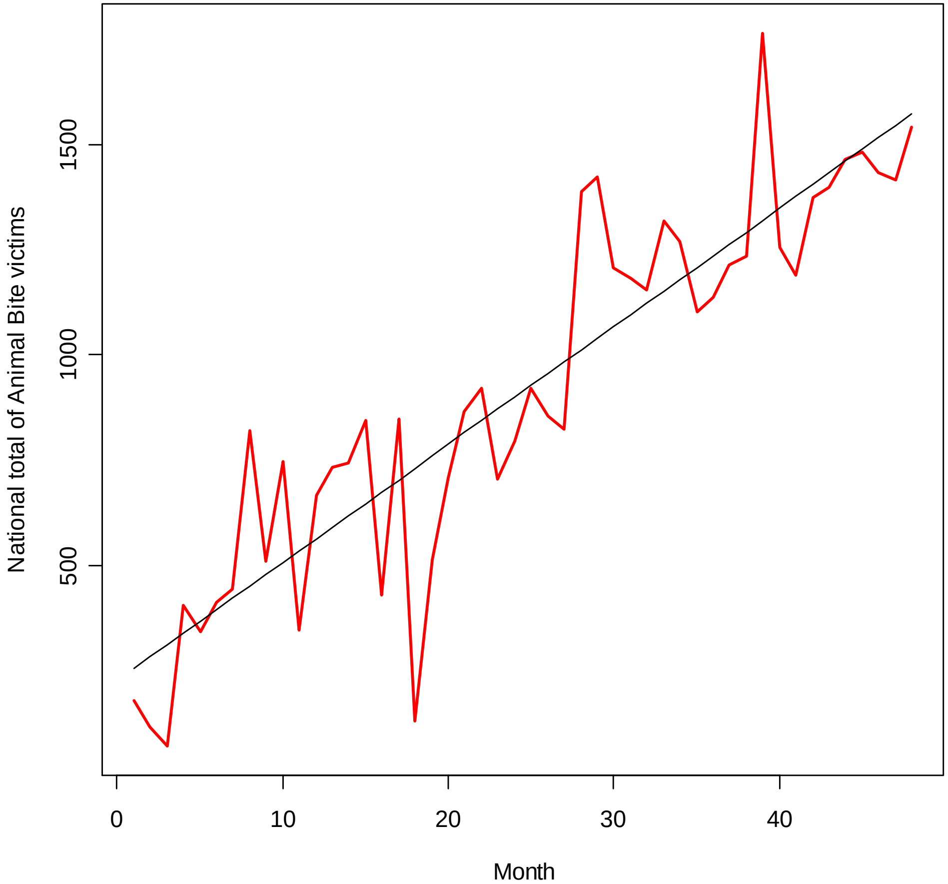
2005



2005

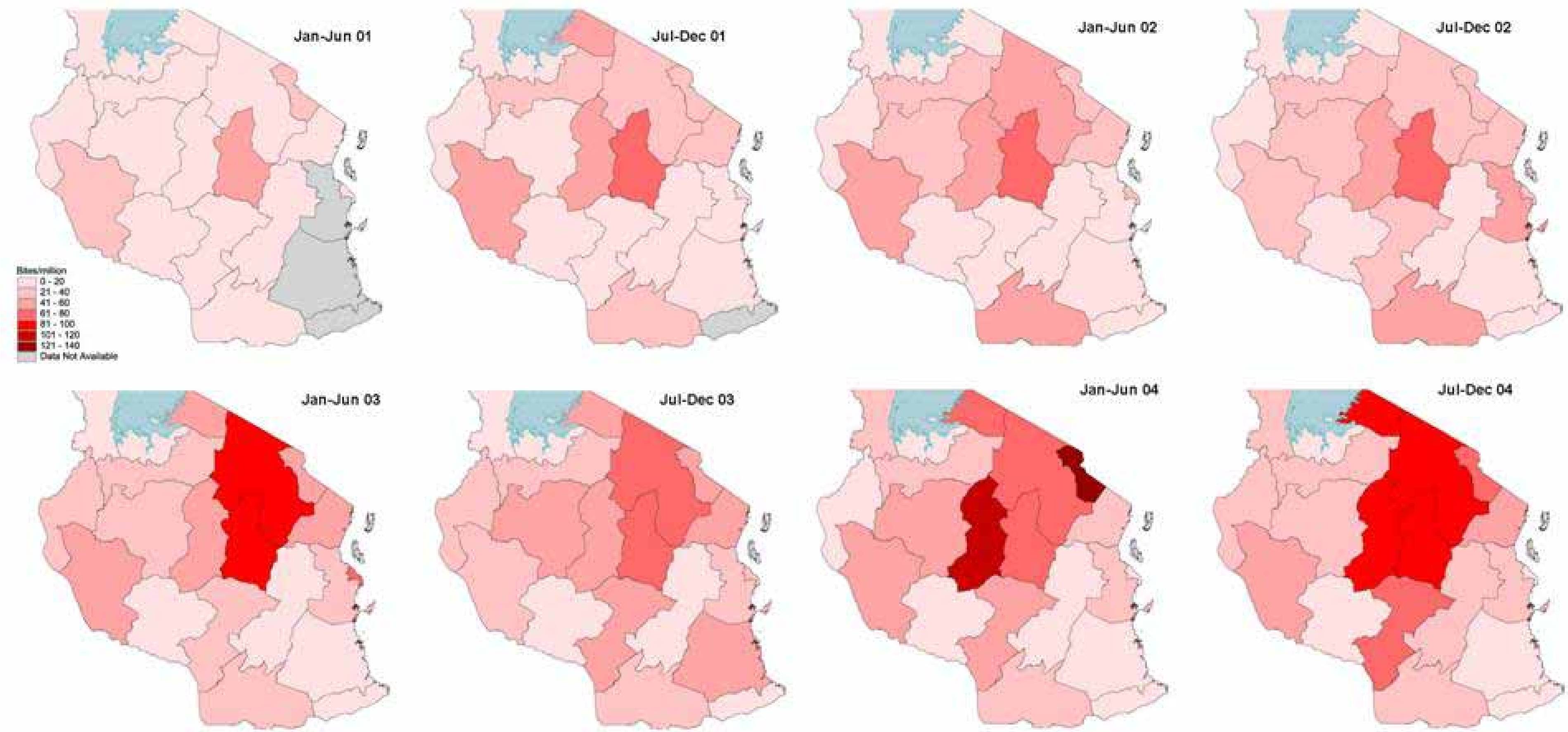


Nationwide incidence: 2001-2004

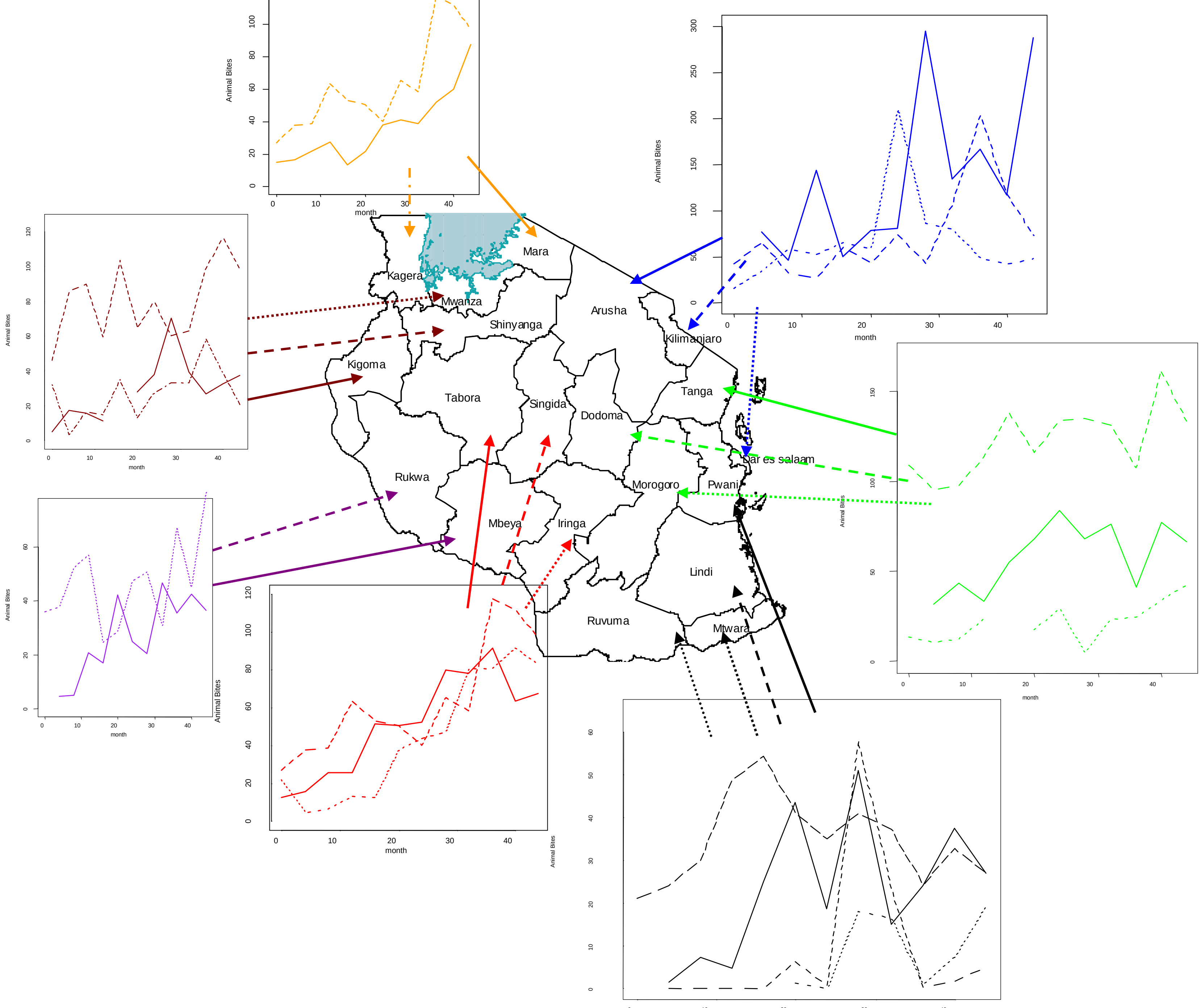


Animal bite records as index of rabies incidence

LARGE-SCALE SPREAD IN TANZANIA



2003: reported from 17 out of 21 regions



Animal Bites
month
Mara
Kagera
Mwanza
Shinyanga
Arusha
Kilimanjaro
Kigoma
Tabora
Singida
Dodoma
Tanga
Dar es salaam
Rukwa
Morogoro
Pwani
Mbeya
Iringa
Lindi
Ruvuma
Mtwara

Rabies Control Strategies in Rural Tanzania

Magai Kaare s0234607@sms.ed.ac.uk

Tiziana Lembo, Katie Hampson, Ernest Eblate, Craig Packer,
Andy Dobson, Sarah Cleaveland

Why do we need different control strategies?

- Required vaccination coverage must be achieved
- Dog accessibility for parenteral vaccination differ between settings
- No one strategy is effective for all communities
    - different cultural attitudes
    - different population distributions
    - different dog densities
- The cost effectiveness of vaccine delivery strategies will differ in different community settings

Examples: Low density pastoralist communities

Examples: High density Agro-pastoralist communities

STRATEGIES

- Central Point
- House to house
- Use of Community-Based Animal Health Workers (CAHWs)

STRATEGIES

- Central Point
- House
- Use of
  Anima

STRATEGIES

- Central Point
- House to house
- Use of Community-Based Animal Health Workers (CAHWs)

STRATEGIES

- Central Point
- House to house
- Use of Community-Based Animal Health Workers (CAHWs)

COMPARISONS

- COST EFFECTIVENESS
- COVERAGE

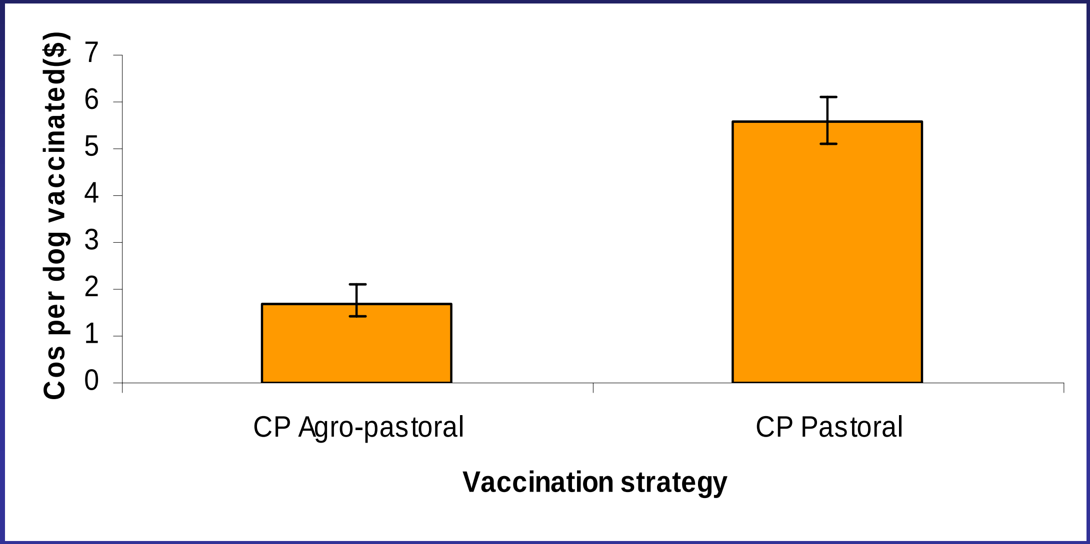
- Between agro-pastoralist and pastoralist communities for central point

- Within pastoralist communities between all strategies (CP,CAHW,HH and their combination)

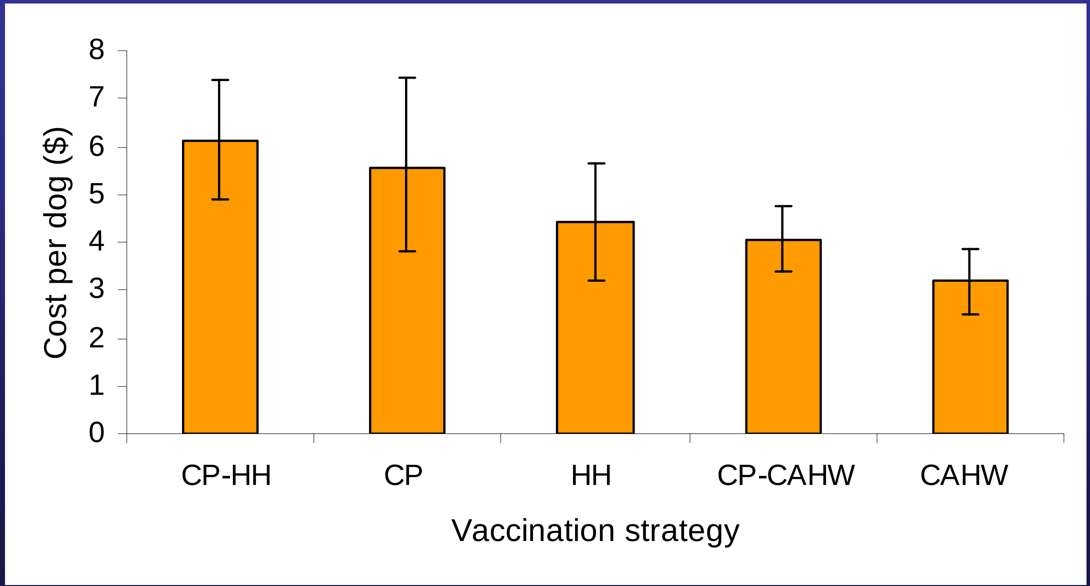
- Central point within agro-pastoralist communities dependant on socio-economic, ethnic and spatial factors

Cost effectiveness

Central point was cost-effective in Agropastoralist but NOT in pastoralist communities



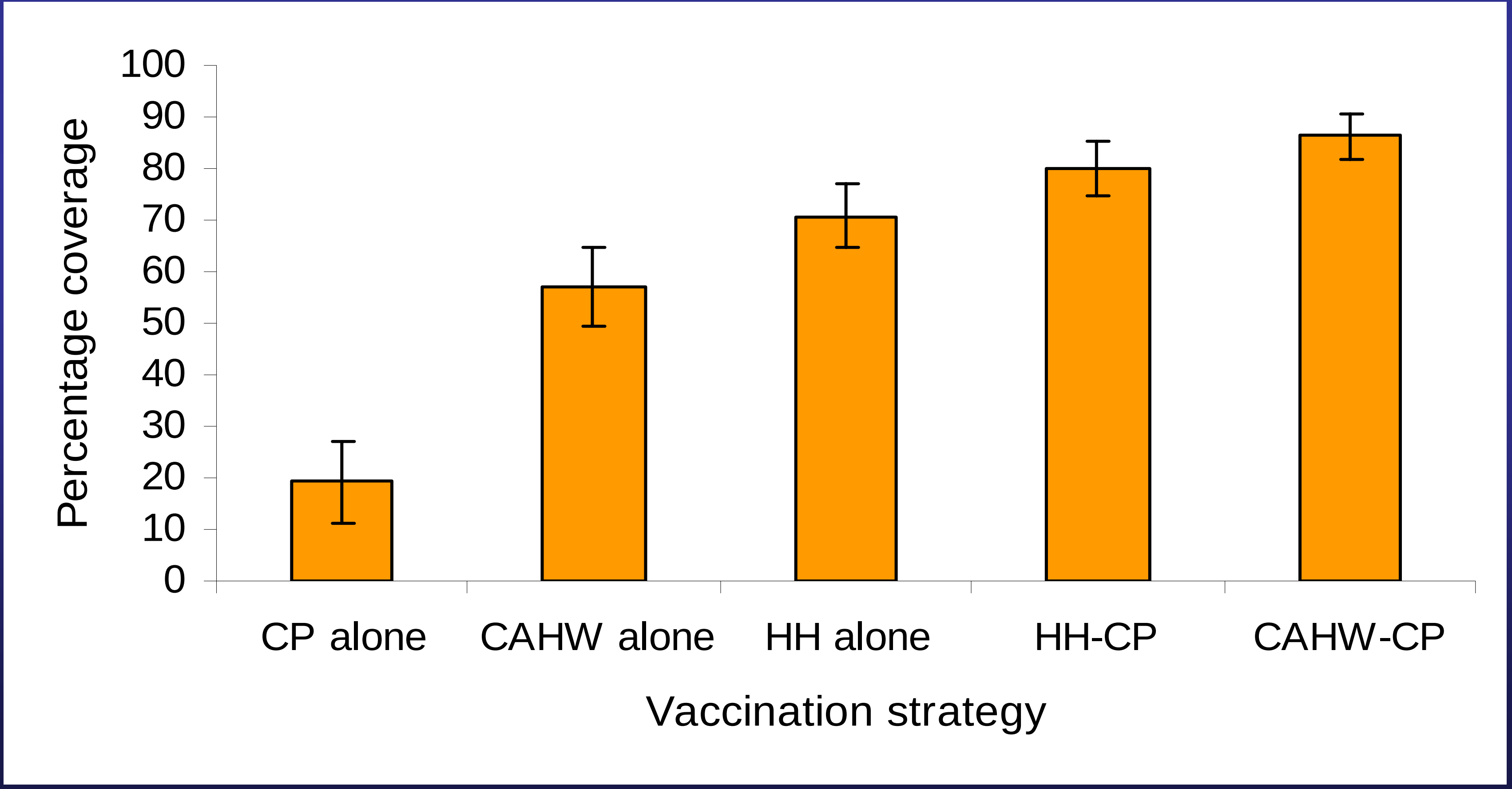
CAHWs and combined Central Point and CAHW strategies were most cost effective in pastoralist communities



COVERAGE

Combined CAHWs/Central point and Central point/house to house strategies achieved highest coverage in pastoralist communities

Central point alone resulted in the lowest vaccination coverage



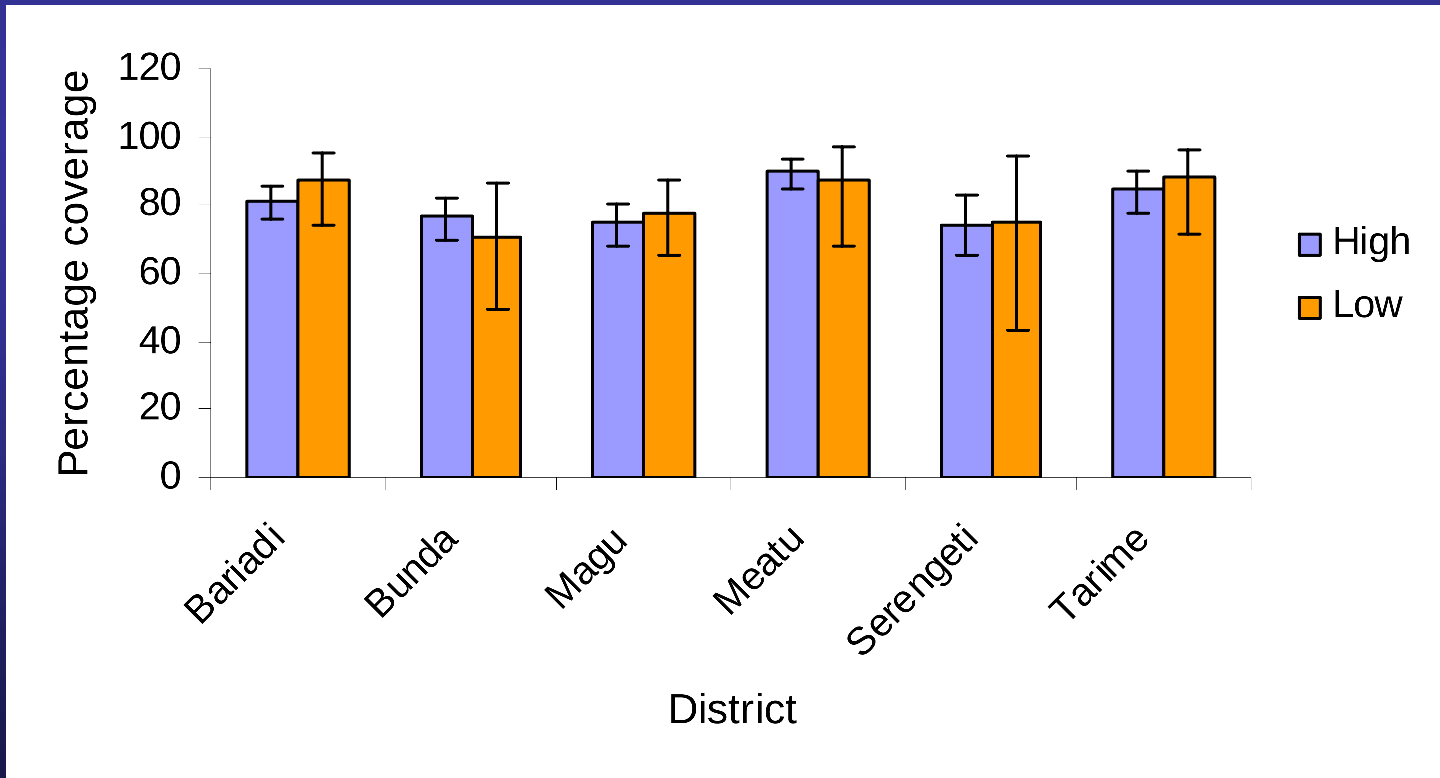
COVERAGE

Central point in Agro-pastoralist communities according to different variables

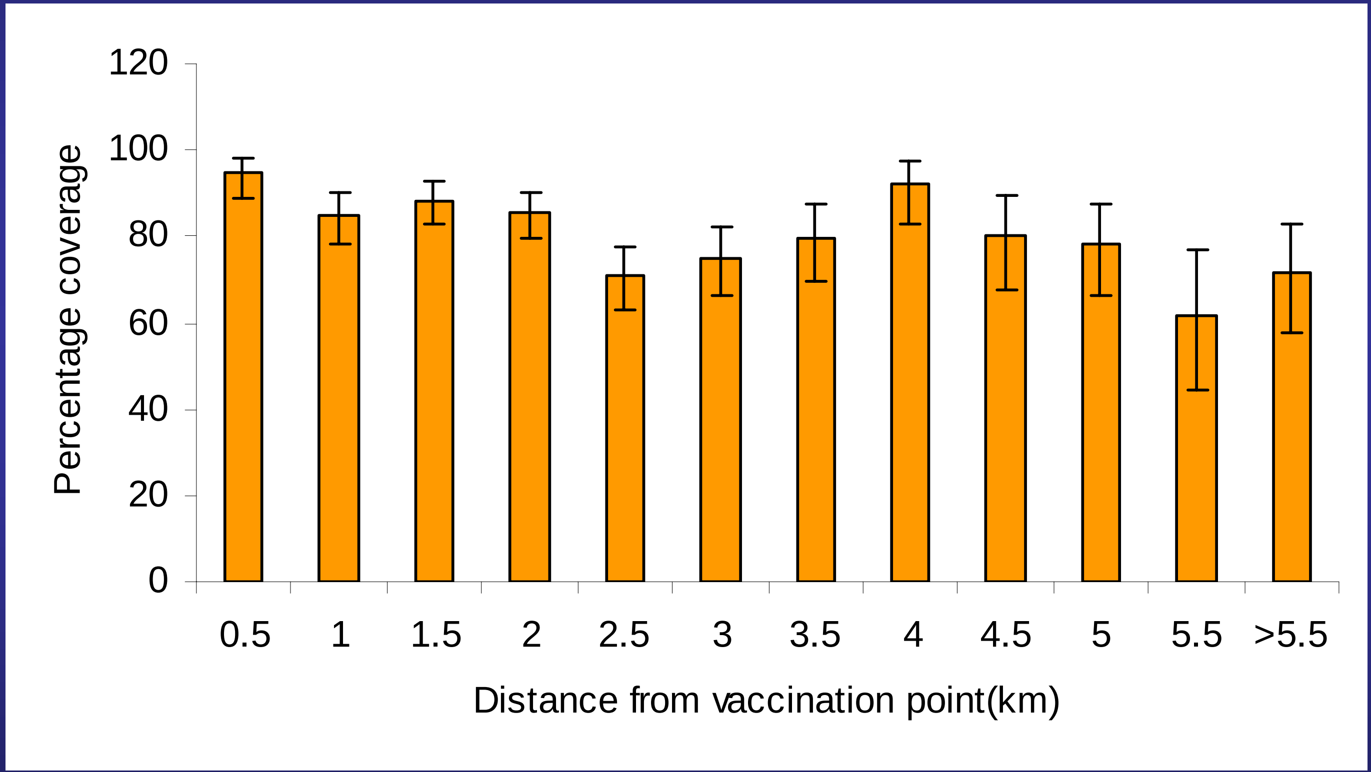
Coverage was high and not significantly different

- in a range of ethnic groups
- irrespective of distance to nearest hospital or district headquarters

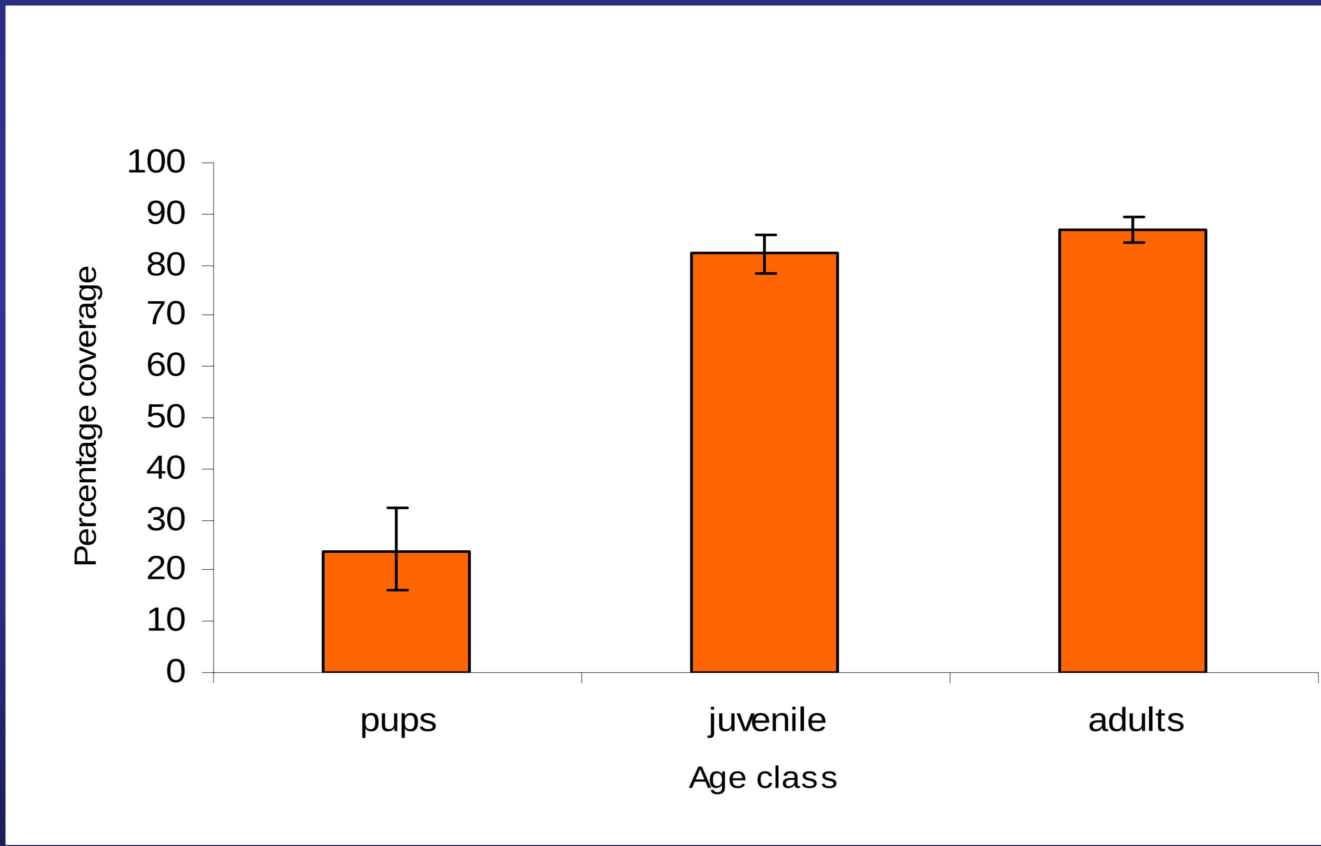
Coverage was high irrespective of socioeconomic status but was more variable among those of low socio-economic status



Coverage was high but declined slightly with distance from the central point



Coverage was high in adult and juvenile dogs, but lowest in puppies



CONCLUSIONS

- Central-point vaccination was more effective in agro-pastoralist than pastoralist communities and achieved the theoretical coverage (70%) sufficient to prevent rabies outbreaks

- The high vaccination coverage achieved in agro-pastoralist communities using central point vaccination was robust to a wide range of social, cultural, spatial and economic factors

- Conventional central point vaccination was ineffective as a strategy for rabies control in highly dispersed and remote pastoralist communities

- Combined strategies involving CP and CAHW were most effective in terms of coverage and cost, with 86% coverage and the combination is therefore recommended as the strategy of choice for rabies control in remote and dispersed pastoralist communities

VISION FOR TANZANIA

- Rabies control is feasible through CP mass vaccination of domestic dogs in most of agro-pastoralist rural cultural and economic settings
- Different strategies will be required for remote pastoralist communities
- Rabies day has been successful in Brazil and is a potential approach for Tanzania
- HOWEVER SUSTAINED AND DEDICATED POLITICAL COMMITMENT WILL BE CRUCIAL!

Acknowledgements

- Ministry of Water and Livestock Development
- Ministry of Regional administration and Local Governments
- INTERVET
- TANAPA
- Ministry of Health
- USA-NSF/NHI (grant no. 0225453)
- CDP
- University of Edinburgh
- FZS

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Application of molecular epidemiology: case studies in Africa, Sudan, Botswana and Namibia

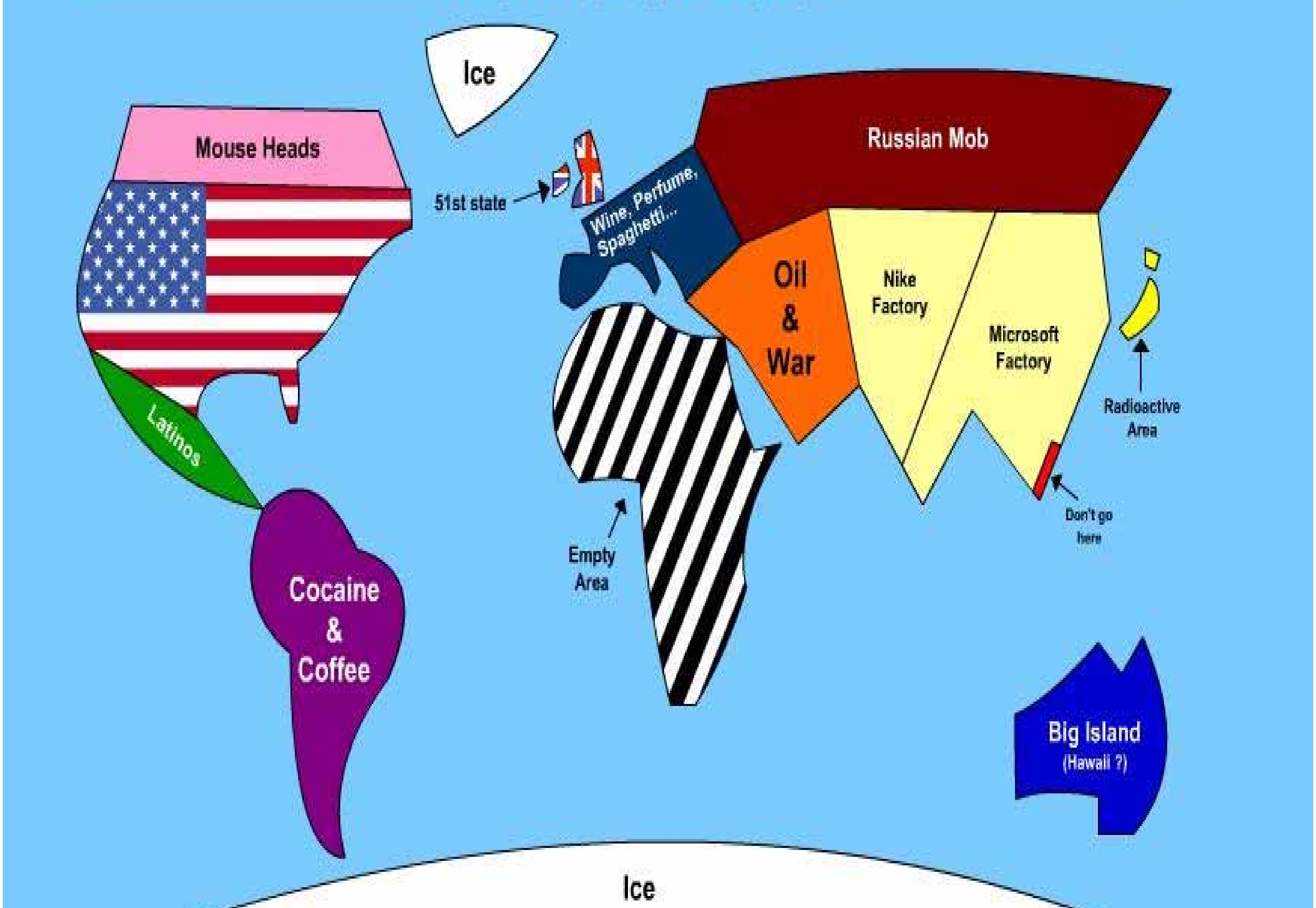
Dr. Anthony R. Fooks

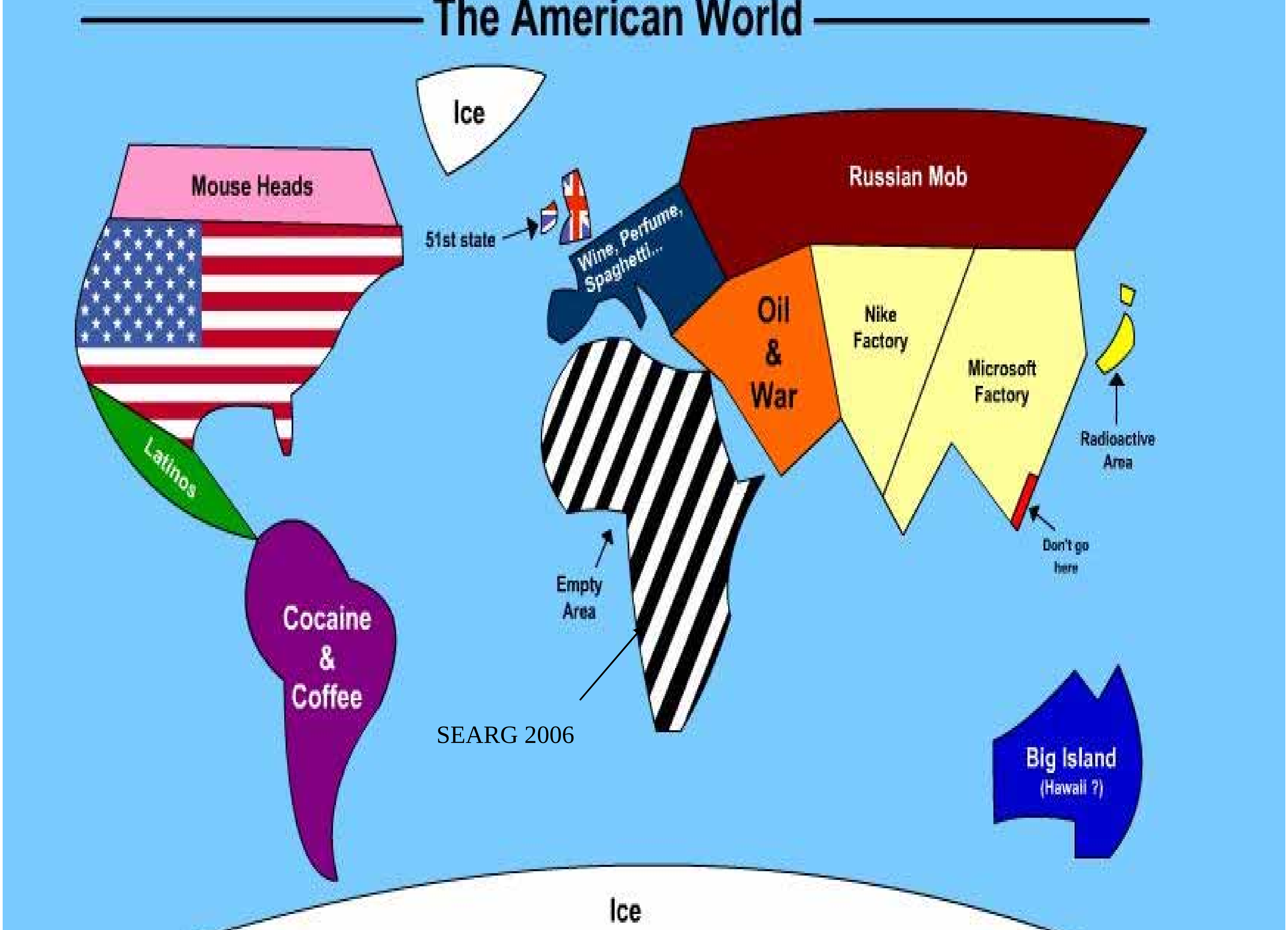
Rabies & Wildlife Zoonoses Group

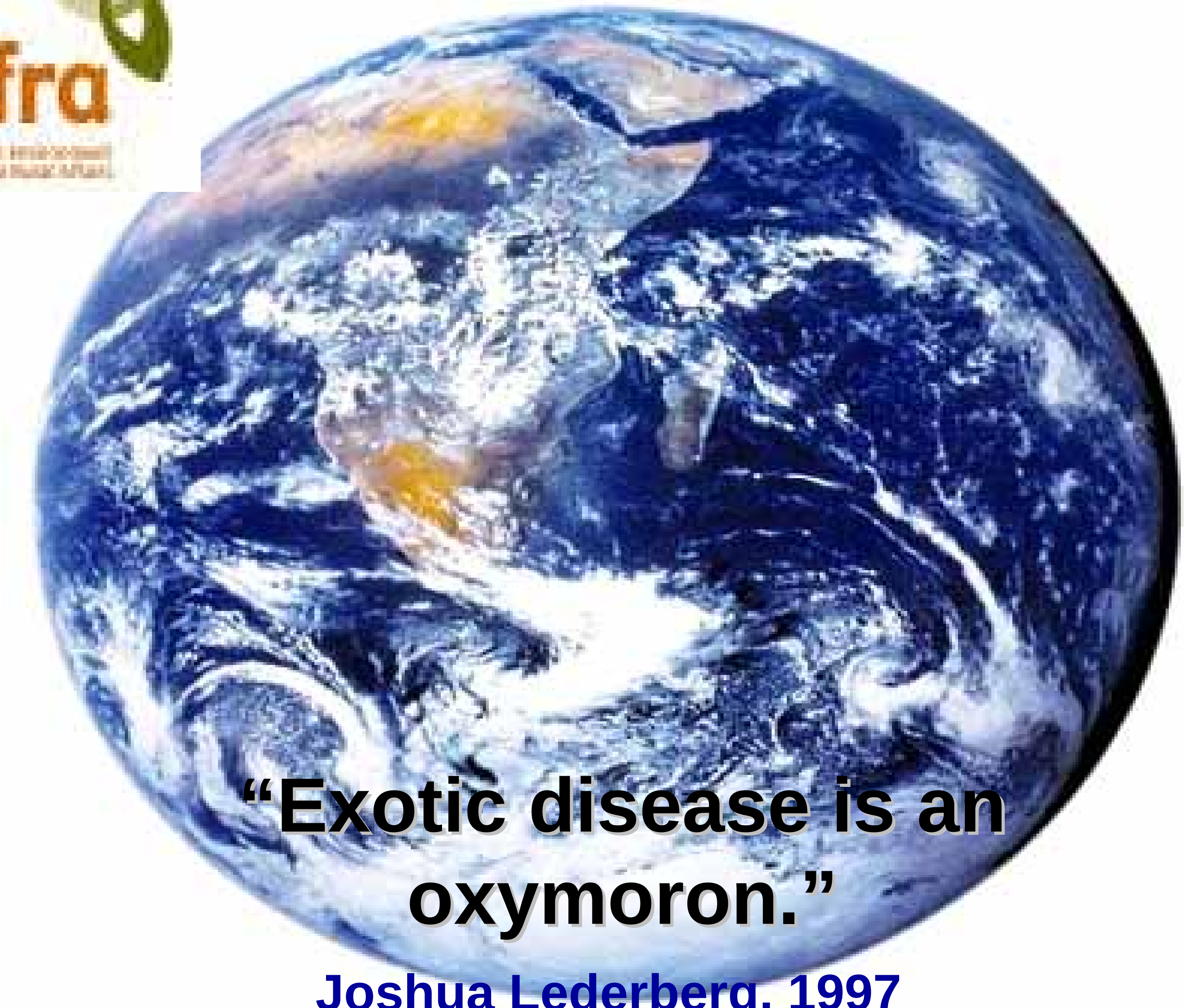
Veterinary Laboratories Agency – Weybridge, UK

8th SEARG Meeting, Namibia

The American World




The American World
Ice
Mouse Heads
51st state
Wine, Perfume, Spaghetti...
Russian Mob
Oil & War
Nike Factory
Microsoft Factory
Radioactive Area
Don't go here
Latinos
Empty Area
Cocaine & Coffee
SEARG 2006
Big Island (Hawaii ?)
Ice



“Exotic disease is an oxymoron.”

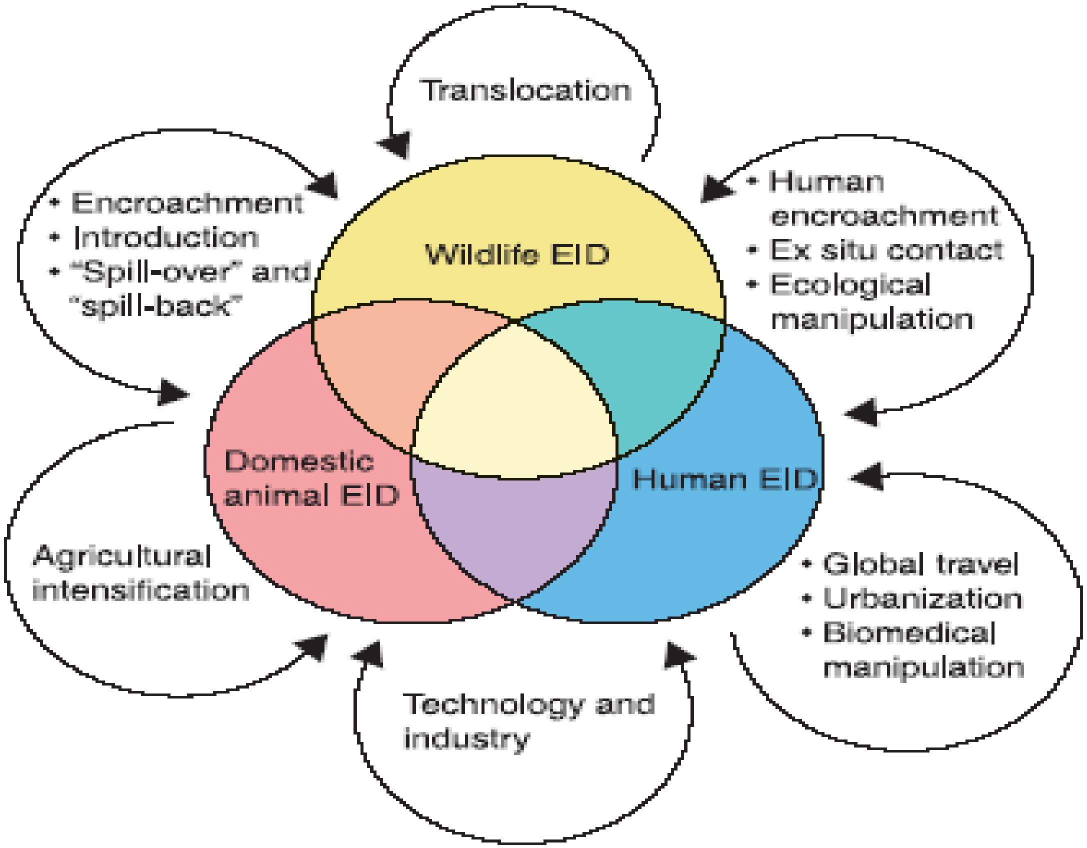
Joshua Lederberg, 1997

Rabies epidemiology in Africa

- Molecular epidemiology assists in our understanding of the transmission of rabies virus from the reservoir host to a spillover host, including man
- Movement of rabies across geographical areas
- Identification of the primary source of spillover infections can then inform control measures or attempts at rabies elimination in a region

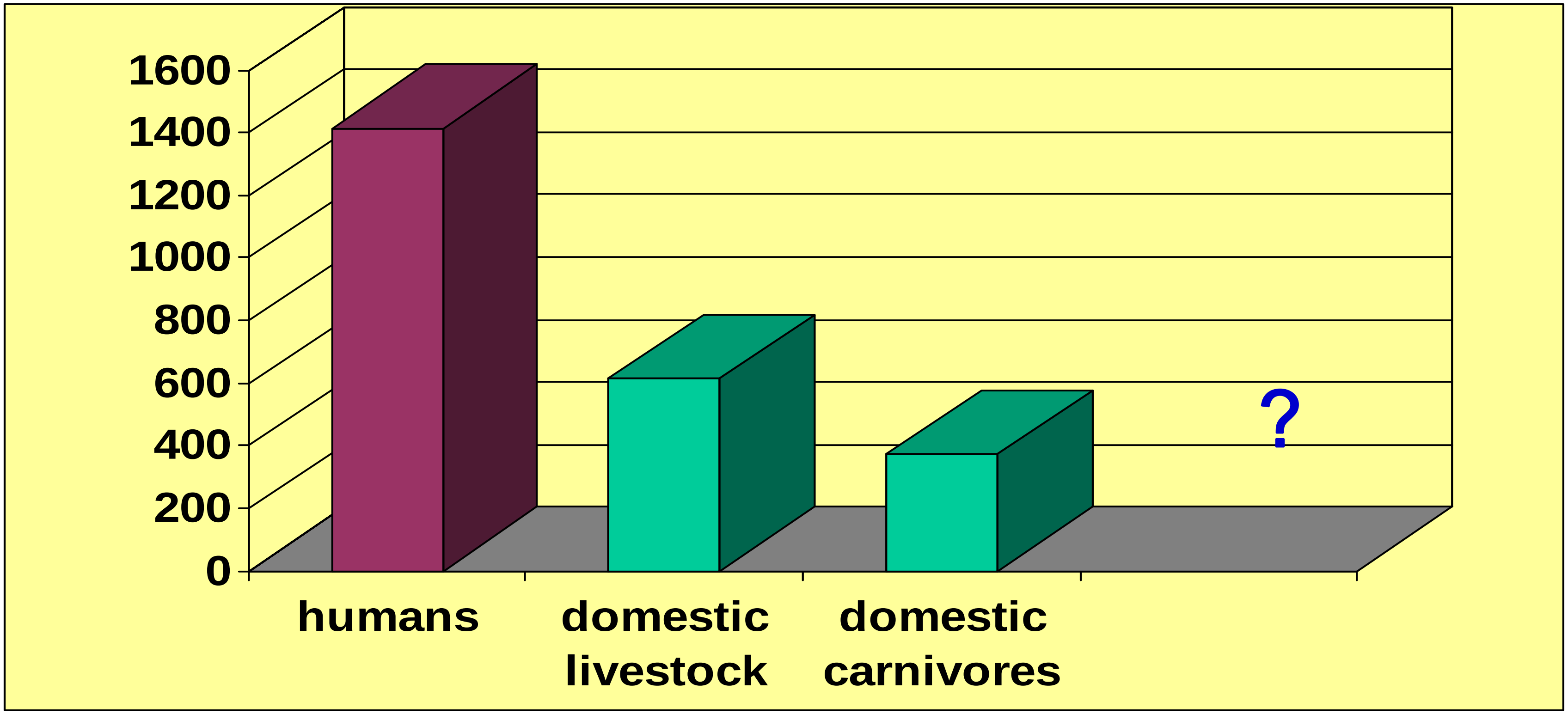
Smith, 1989; Smith et al., 1992; Kissi et al., 1995; Bourhy et al., 1999; De Mattos et al., 2000; Nadin-Davis et al., 2001; Badrane and Tordo, 2001

Rationale

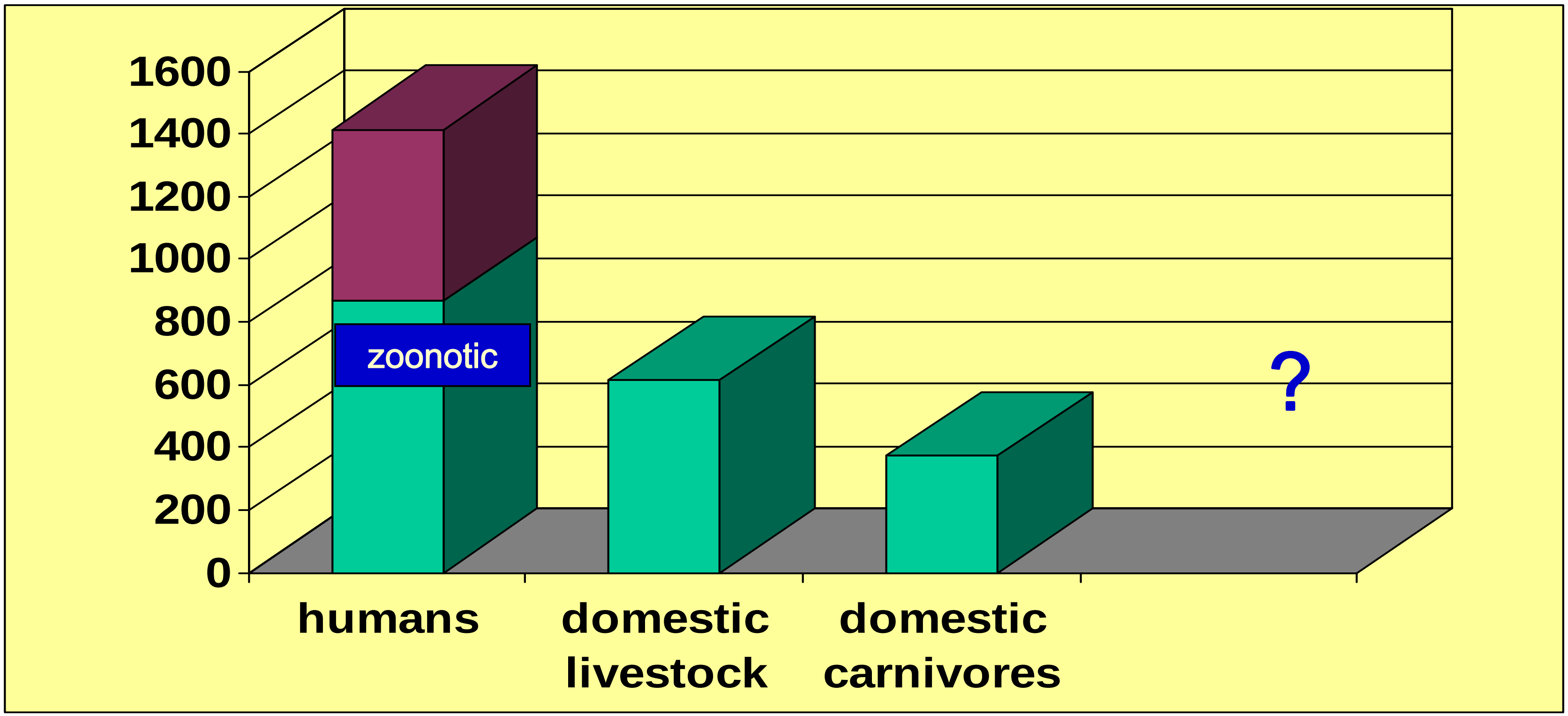


From: Daszak, P., A. A. Cunningham, et al. (2000). "Emerging Infectious Diseases of Wildlife- threats to biodiversity and human health." Science **287**: 443-448.

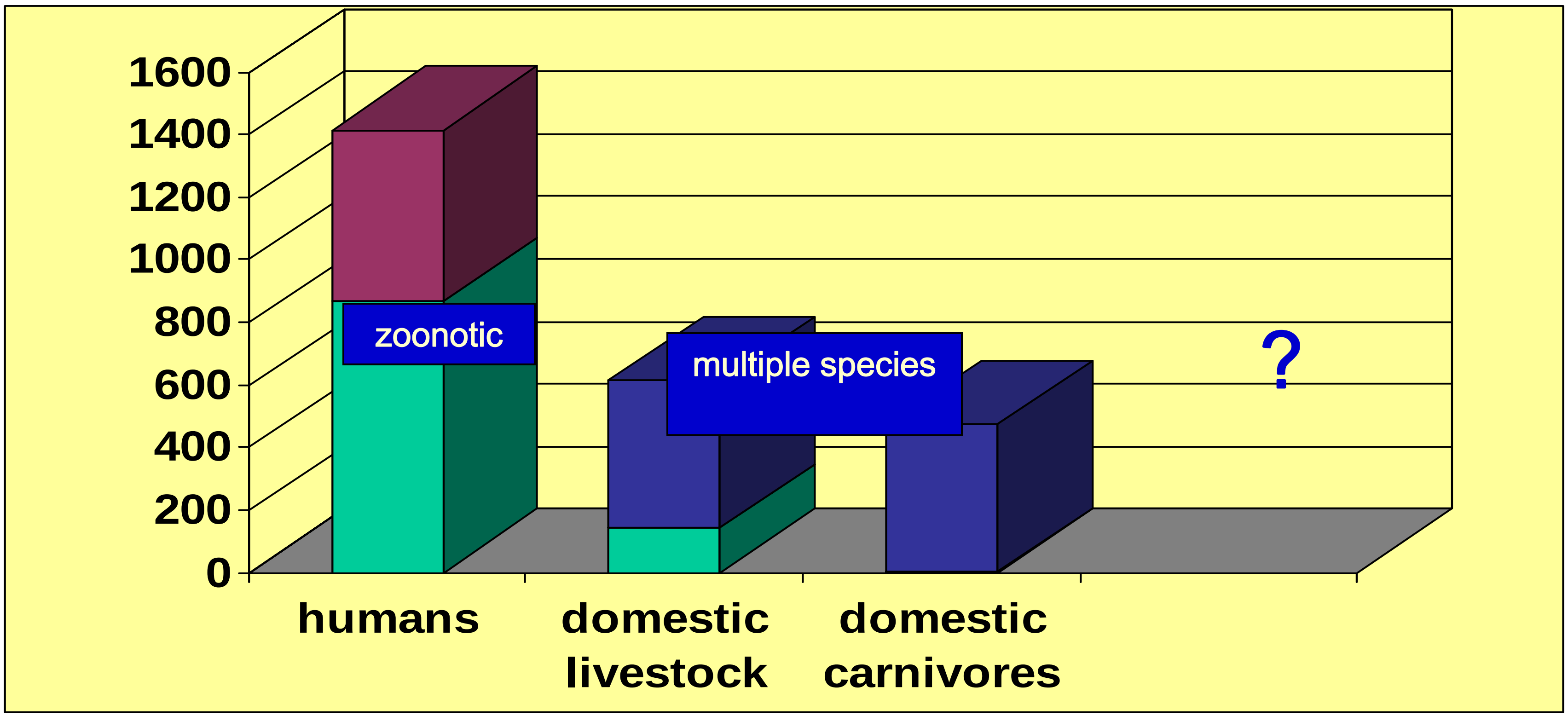
Numbers of Known Pathogens



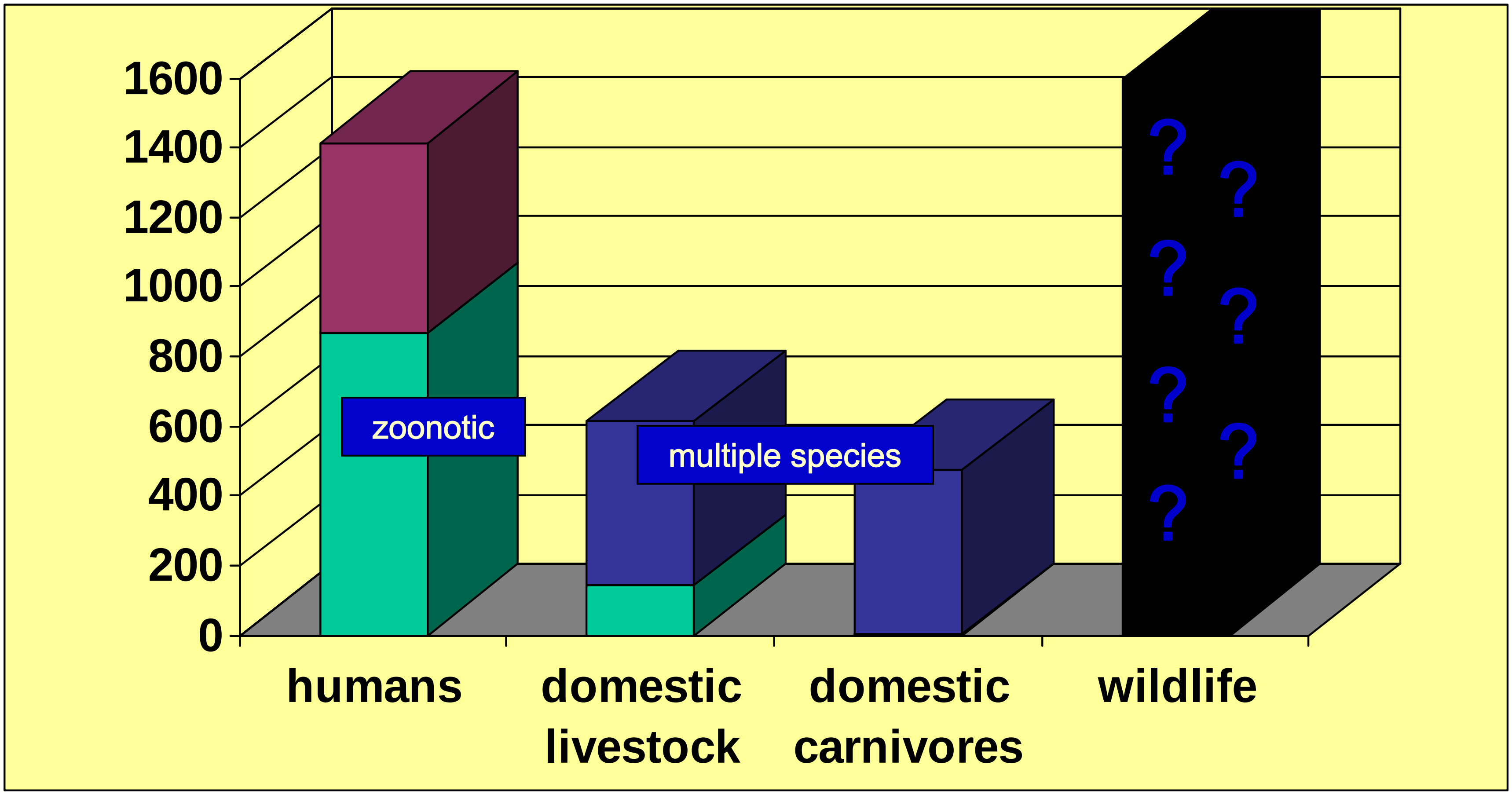
Numbers of Known Pathogens



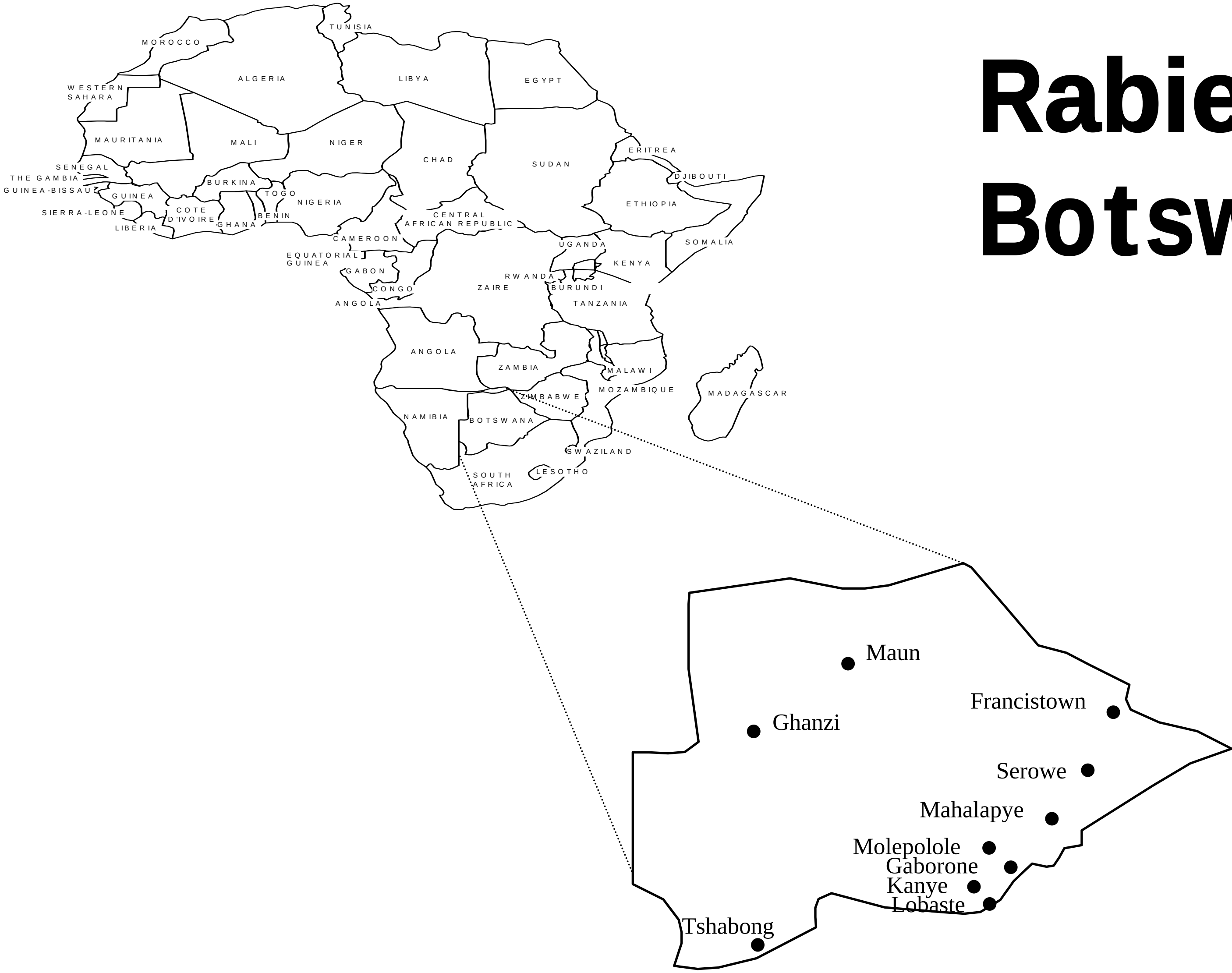
Numbers of Known Pathogens



Numbers of Known Pathogens



Rabies in Botswana



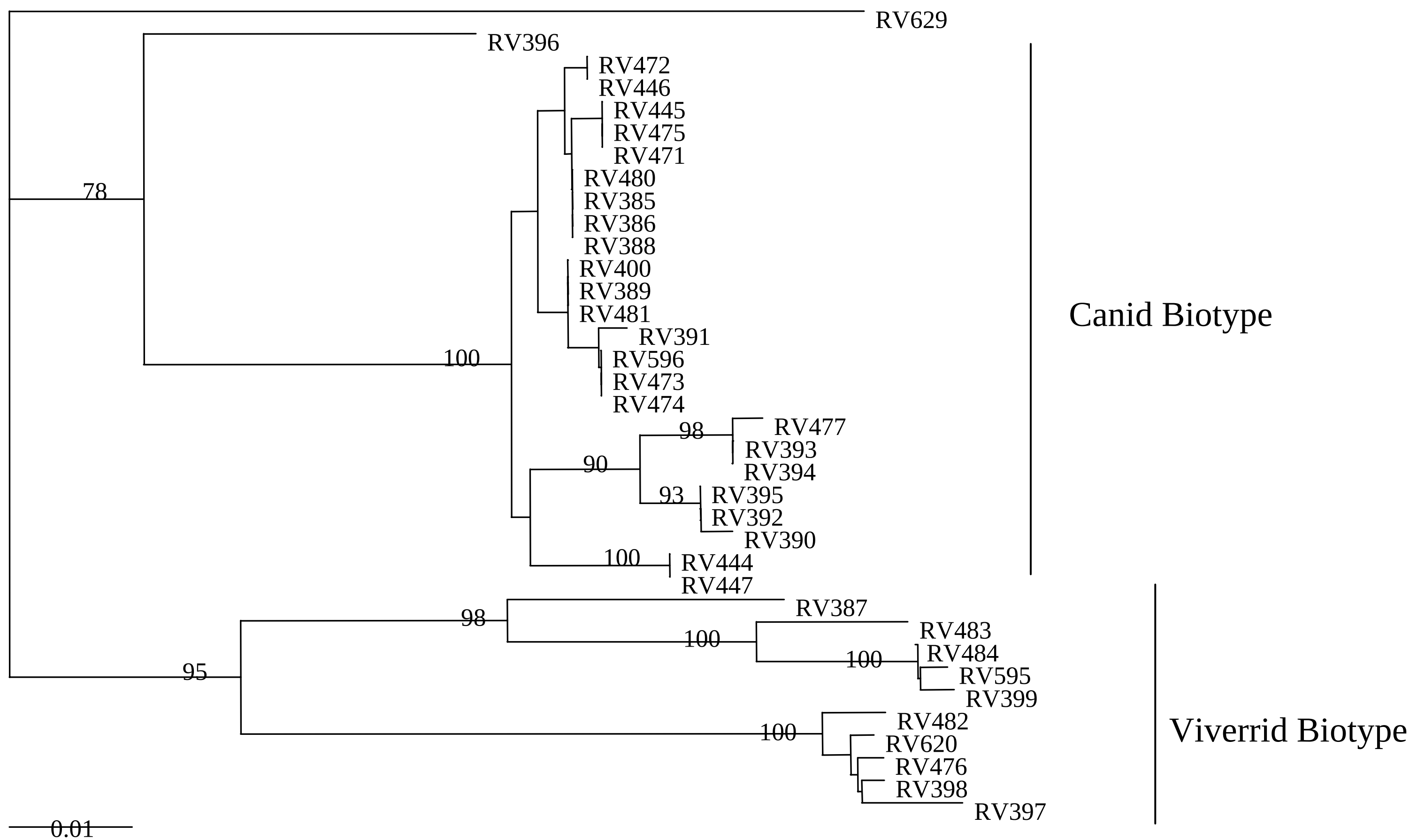
Johnson, N., Letshwenyo, M., Baipoledi, E.K., Thobokwe, G. and A.R. Fooks. (2004). Molecular epidemiology of rabies in Botswana: a comparison between antibody typing and nucleotide sequence phylogeny. *Veterinary Microbiology*. **10;101(1):**31-8.

Botswana

- Hosts include:
  - Domestic dogs, Black-backed jackal, yellow mongoose, small-spotted genet
- *n=35* (samples from 1988 – 1992)
  - Jackal (n=5); Mongoose (n=1); Dog (n=5); Goat (n=6); Human (n=1); Other (n=17)
- Two dominant groups:
  - Gp 1: Domestic dog
  - Gp 2: Wildlife species

Mab binding - Botswana

RABV Biotype	M2	DB1	DB3	DB4	DB11	DB14	L3	L4	L23	L25	L26	L28
Canid	+	+	+	+	+	+	+	+	+	+	+	+
Viverrid	+	+	+	+	-	-	+	-	-	-	+	-


RV629
RV396
RV472
RV446
RV445
RV475
RV471
RV480
RV385
RV386
RV388
RV400
RV389
RV481
RV391
RV596
RV473
RV474
RV477
RV393
RV394
RV395
RV392
RV390
RV444
RV447
RV387
RV483
RV484
RV595
RV399
RV482
RV620
RV476
RV398
RV397
78
100
98
90
93
100
98
100
100
95
100
0.01
Canid Biotype
Viverrid Biotype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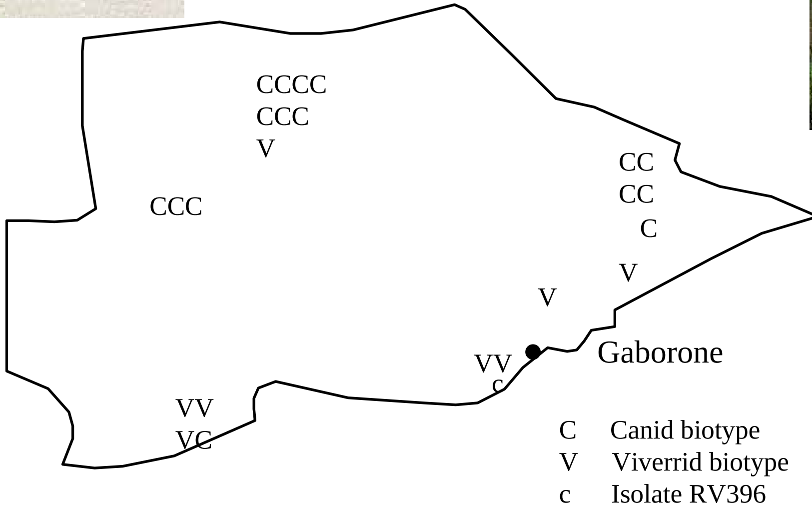
CCCC
CCC
V
CCC
CC
CC
C
V
V
Gaborone
VV
c
VV
VC
C Canid biotype
V Viverrid biotype
c Isolate RV396


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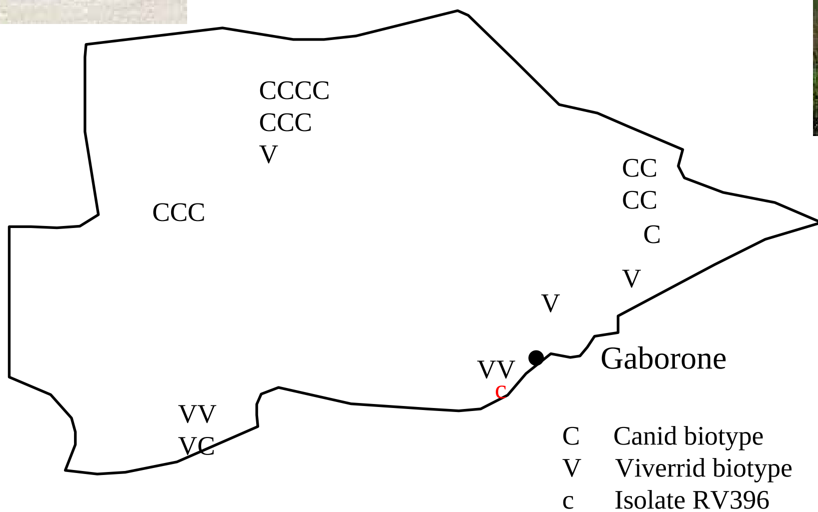
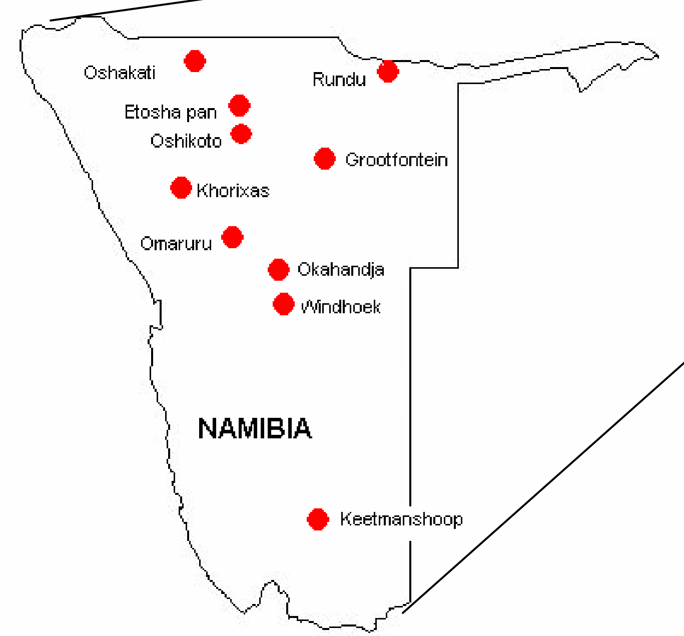

CCCC
CCC
V
CCC
CC
CC
C
V
V
VV
c
Gaborone
VV
VC
C Canid biotype
V Viverrid biotype
c Isolate RV396


www.corbis.com

Conclusions

- Two distinct biotypes (von Teichman *et al*., 1995; Nel *et al*., 1997; Sabeta *et al*., 2003)
  - Wildlife-associated group more phylogentically diverse
    - Isolated in SE Botswana
  - Canine-associated group
    - Dominated north Botswana
  - Main population centres in SE
  - Suggests vaccination campaigns targeting urban areas in south are successful
- Single outlier (RV396) – domestic cat
  - Recovered from a town nr to RSA border
  - Introduced by transboundary incursions
- Some minor discrepancies between Mab typing and genetic analysis
  - Sub-groups <u>not</u> identified by Mab typing
- Little sequence variation within groups
- Little evolutionary pressure
- Spillover infections of domestic animal rabies
  - Control strategies for dog-mediated rabies must be continued

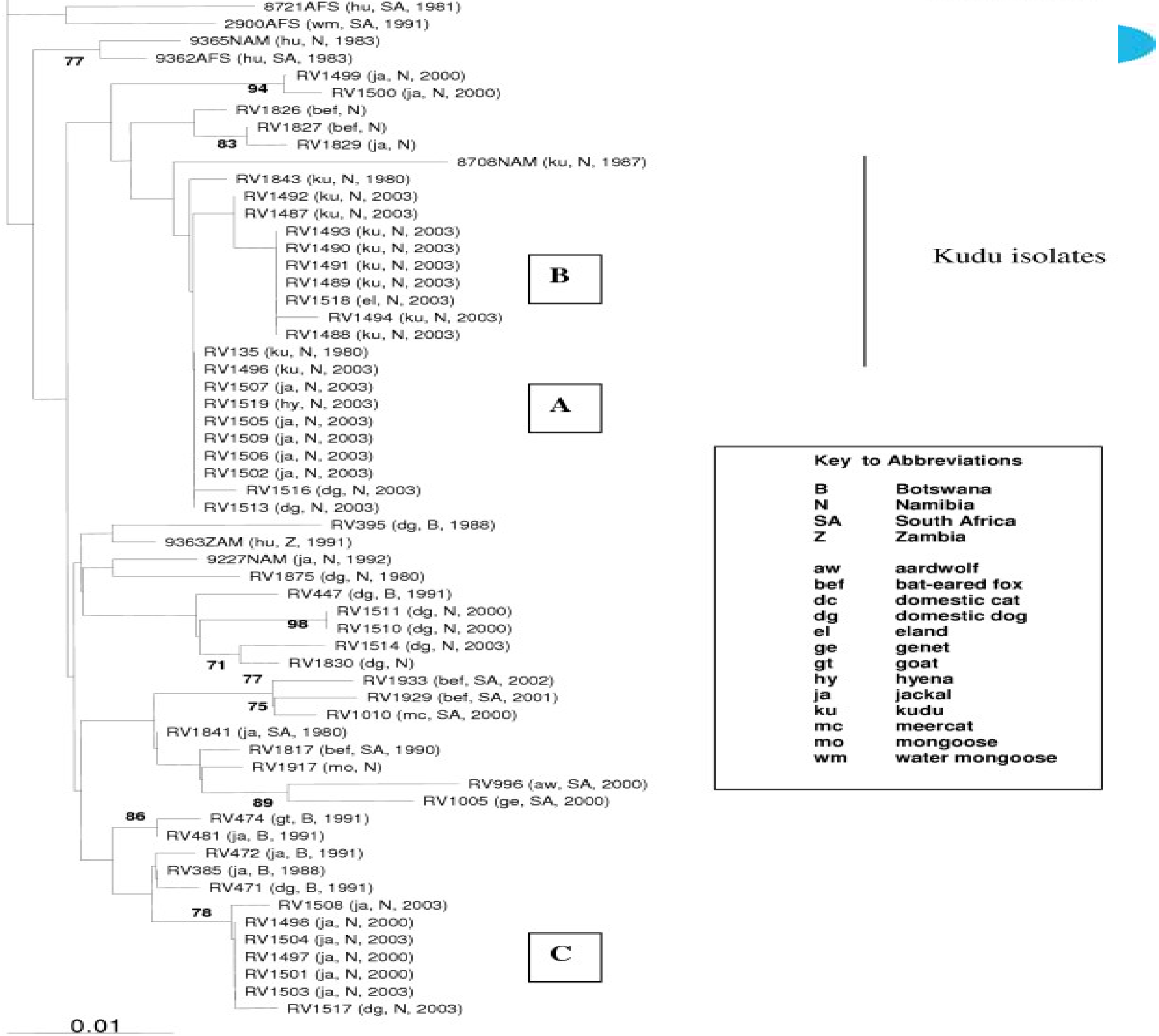
Rabies in Namibia



Mansfield K., McElhinney, L.M., Hübschle, O., Mettler, F., Sabeta, C., Nel, L.H. and A.R. Fooks. (2006). A Molecular Epidemiological Study of Rabies Epizootics in Kudu (*Tragelaphus strepsiceros*) in Namibia. *BMC Veterinary Research.* 13; *2(1):2*

Namibia

- Two distinct biotypes
  - *Canid*
  - *Wildlife – mongoose (viverrid)*
- *n=37* (samples from 1980 – 2003)
  - Jackal (*n=6*); Kudu (*n=10*); Dog (*n=2*); BEF (*n=2*); Other *(n=17*)


8721AFS (hu, SA, 1981)
2900AFS (wm, SA, 1991)
9365NAM (hu, N, 1983)
9362AFS (hu, SA, 1983)
77
RV1499 (ja, N, 2000)
94
RV1500 (ja, N, 2000)
RV1826 (bef, N)
RV1827 (bef, N)
83
RV1829 (ja, N)
8706NAM (ku, N, 1987)
RV1843 (ku, N, 1980)
RV1492 (ku, N, 2003)
RV1487 (ku, N, 2003)
RV1493 (ku, N, 2003)
RV1490 (ku, N, 2003)
RV1491 (ku, N, 2003)
RV1489 (ku, N, 2003)
RV1518 (el, N, 2003)
RV1494 (ku, N, 2003)
RV1488 (ku, N, 2003)
B
Kudu isolates
RV135 (ku, N, 1980)
RV1496 (ku, N, 2003)
RV1507 (ja, N, 2003)
RV1519 (hy, N, 2003)
RV1505 (ja, N, 2003)
RV1509 (ja, N, 2003)
RV1506 (ja, N, 2003)
RV1502 (ja, N, 2003)
RV1516 (dg, N, 2003)
RV1513 (dg, N, 2003)
A
RV395 (dg, B, 1988)
9363ZAM (hu, Z, 1991)
9227NAM (ja, N, 1992)
RV1875 (dg, N, 1980)
RV447 (dg, B, 1991)
RV1511 (dg, N, 2000)
98
RV1510 (dg, N, 2000)
RV1514 (dg, N, 2003)
71
RV1830 (dg, N)
77
RV1933 (bef, SA, 2002)
RV1929 (bef, SA, 2001)
75
RV1010 (mc, SA, 2000)
RV1841 (ja, SA, 1980)
RV1817 (bef, SA, 1990)
RV1917 (mo, N)
RV996 (aw, SA, 2000)
89
RV1005 (ge, SA, 2000)
86
RV474 (gt, B, 1991)
RV481 (ja, B, 1991)
RV472 (ja, B, 1991)
RV385 (ja, B, 1988)
RV471 (dg, B, 1991)
RV1508 (ja, N, 2003)
78
RV1498 (ja, N, 2000)
RV1504 (ja, N, 2003)
RV1497 (ja, N, 2000)
RV1501 (ja, N, 2000)
RV1503 (ja, N, 2003)
RV1517 (dg, N, 2003)
C
0.01
Key to Abbreviations
B Botswana
N Namibia
SA South Africa
Z Zambia
aw aardwolf
bef bat-eared fox
dc domestic cat
dg domestic dog
el eland
ge genet
gt goat
hy hyena
ja jackal
ku kudu
mc meercat
mo mongoose
wm water mongoose

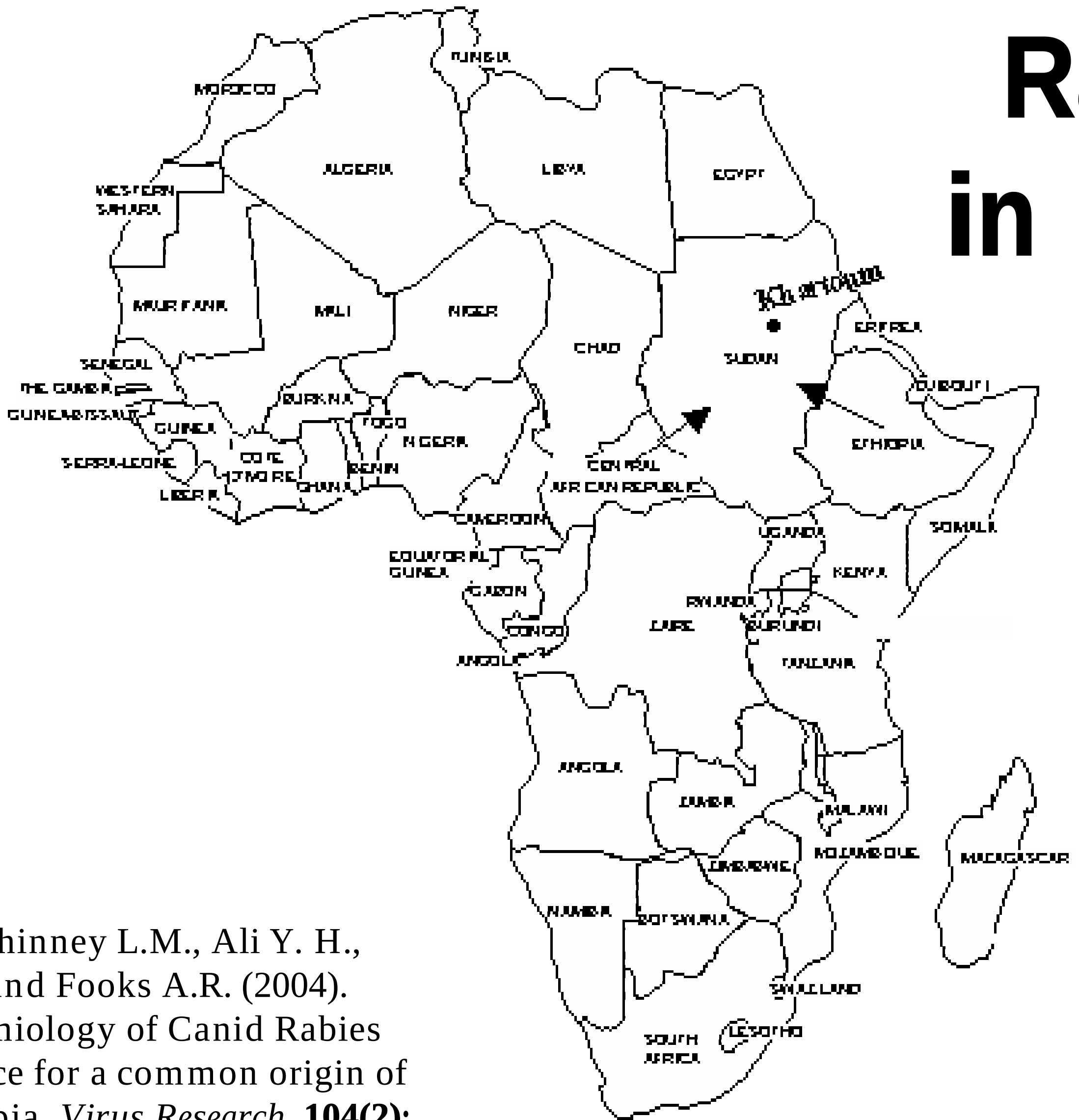
Results

- Group A
  - Northern districts of Grootfontein and Etosha
  - Dog, jackal and kudu isolates
- Group B
  - Central Namibia
  - Mainly kudu isolates
- Group C
  - Jackal isolates from Windhoek
  - Kudu
  - Domestic dogs shared identical sequences

Conclusions

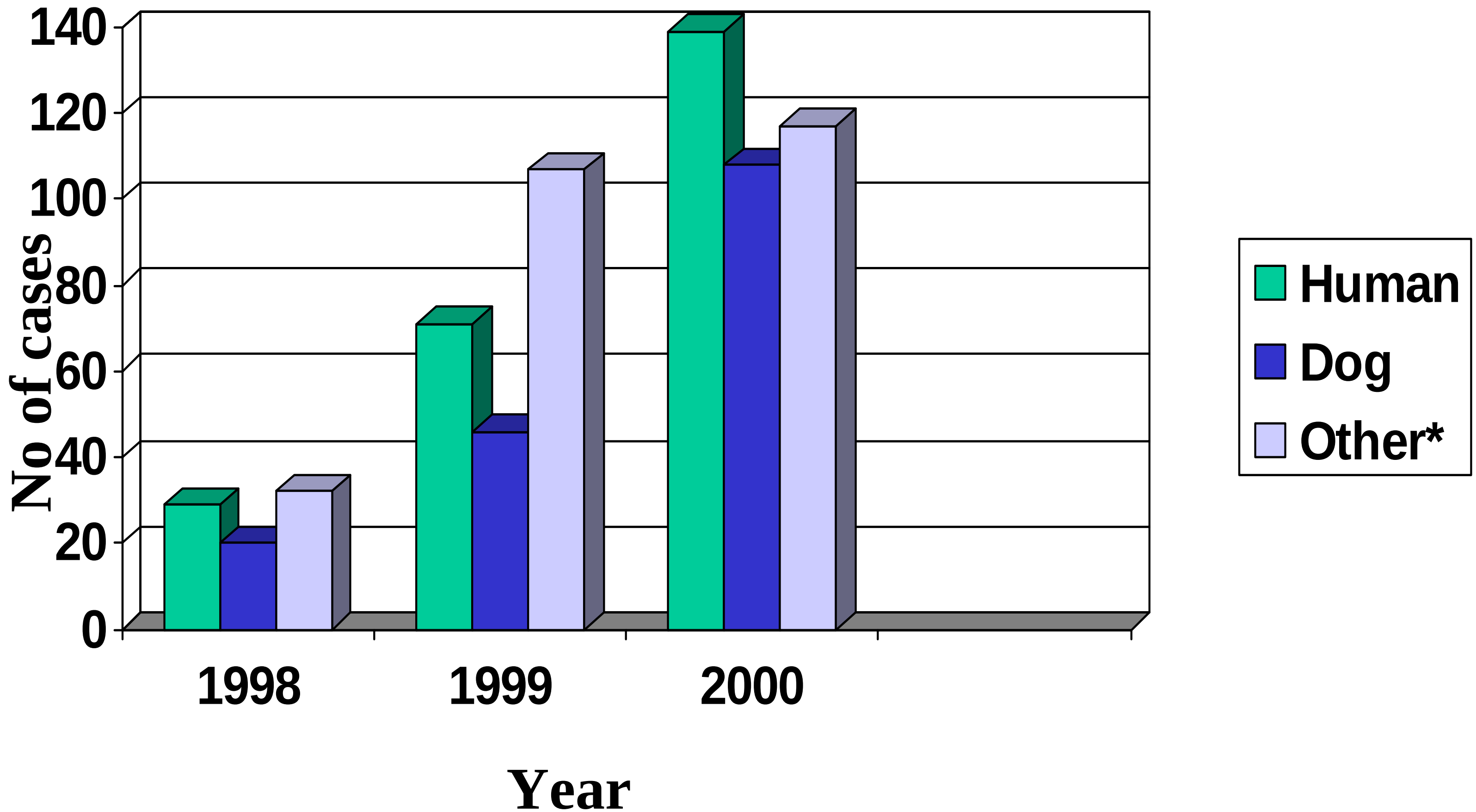
- Two distinct biotypes
  - *Canid*
  - *Wildlife*
- Canid biotype already firmly established in southern Africa
  - Poses a threat to domestic livestock and man
- Jackal and kudu may form part of the same epidemiological cycle of rabies in Namibian wildlife
  - Both groups share 97 – 100% sequence homology
- Control strategies will be difficult to implement due to the large variation in species
  - Focus on the domestic dog

Rabies in Sudan



Johnson N., McElhinney L.M., Ali Y. H., Intisar K. Saeed. and Fooks A.R. (2004). Molecular Epidemiology of Canid Rabies in Sudan: Evidence for a common origin of rabies with Ethiopia. *Virus Research.* **104(2);** 201-205.

Rabies in Sudan



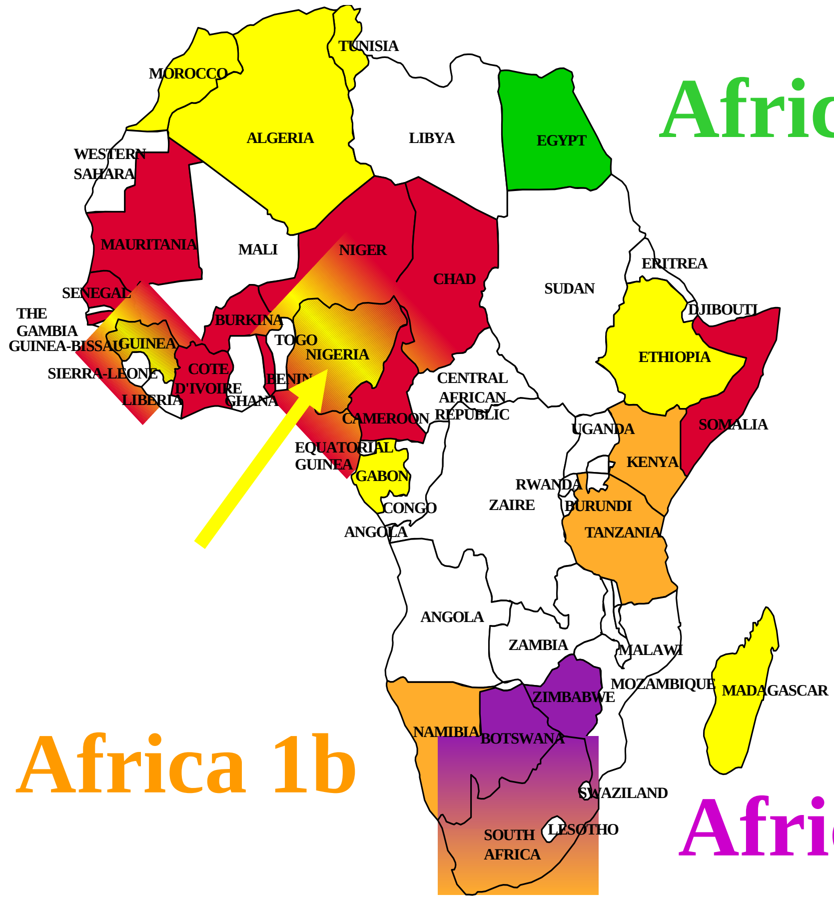
(Source: Central Veterinary Research Laboratory, Khartoum).

* Other species include cats, goats, sheep, horses, cattle, camels and monkeys.

Rabies - Africa

Africa 1a

Africa 4



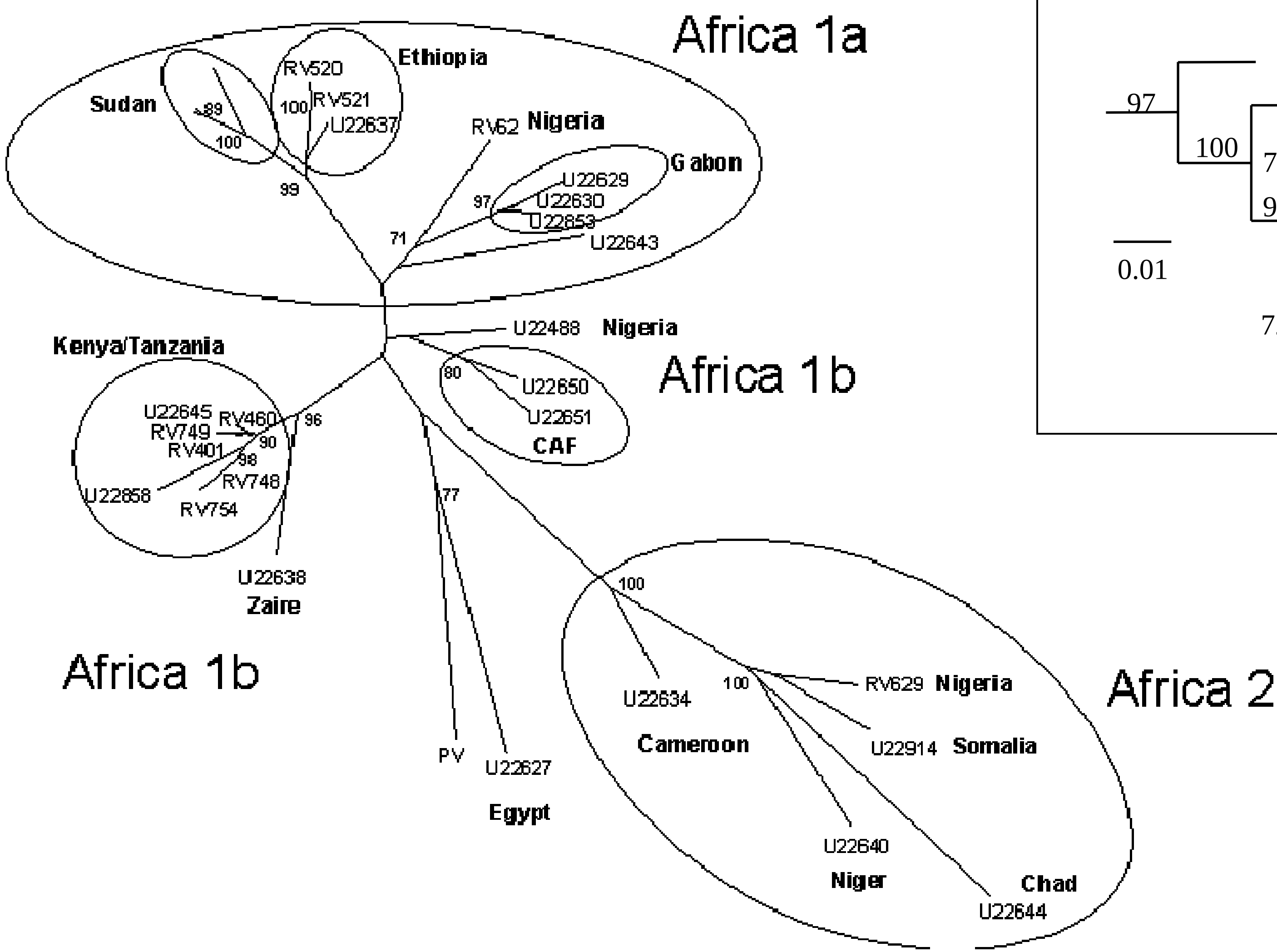
Africa 2

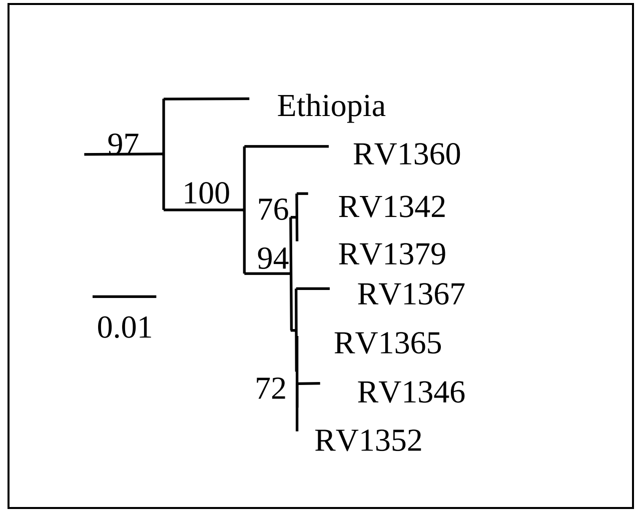
Africa 1b

Africa 3

Canid rabies in Sudan: Evidence for a common origin of rabies with Ethiopia

- Small panel of Sudanese canid RABV samples was studied (*n=7*)
- RABV in Sudan clustered within the Africa 1 group of viruses
- Common sequence homologies between RABV circulating in Sudan and Ethiopia
- Numerous species implicaed - role in rabies movement across national boundaries


Africa 1a
Sudan
Ethiopia
RV520
RV521
U22637
89
100
100
99
RV62 Nigeria
Gabon
U22629
U22630
U22853
97
71
U22643
U22488 Nigeria
Africa 1b
80
U22650
U22651
CAF
Kenya/Tanzania
U22645
RV749
RV401
RV460
96
90
98
RV748
U22858
RV754
U22638
Zaire
Africa 1b
77
PV
U22627
Egypt
100
U22634
Cameroon
100
RV629 Nigeria
U22914 Somalia
U22640
Niger
Chad
U22644
Africa 2


97
Ethiopia
100
RV1360
76
RV1342
94
RV1379
RV1367
0.01
RV1365
72
RV1346
RV1352

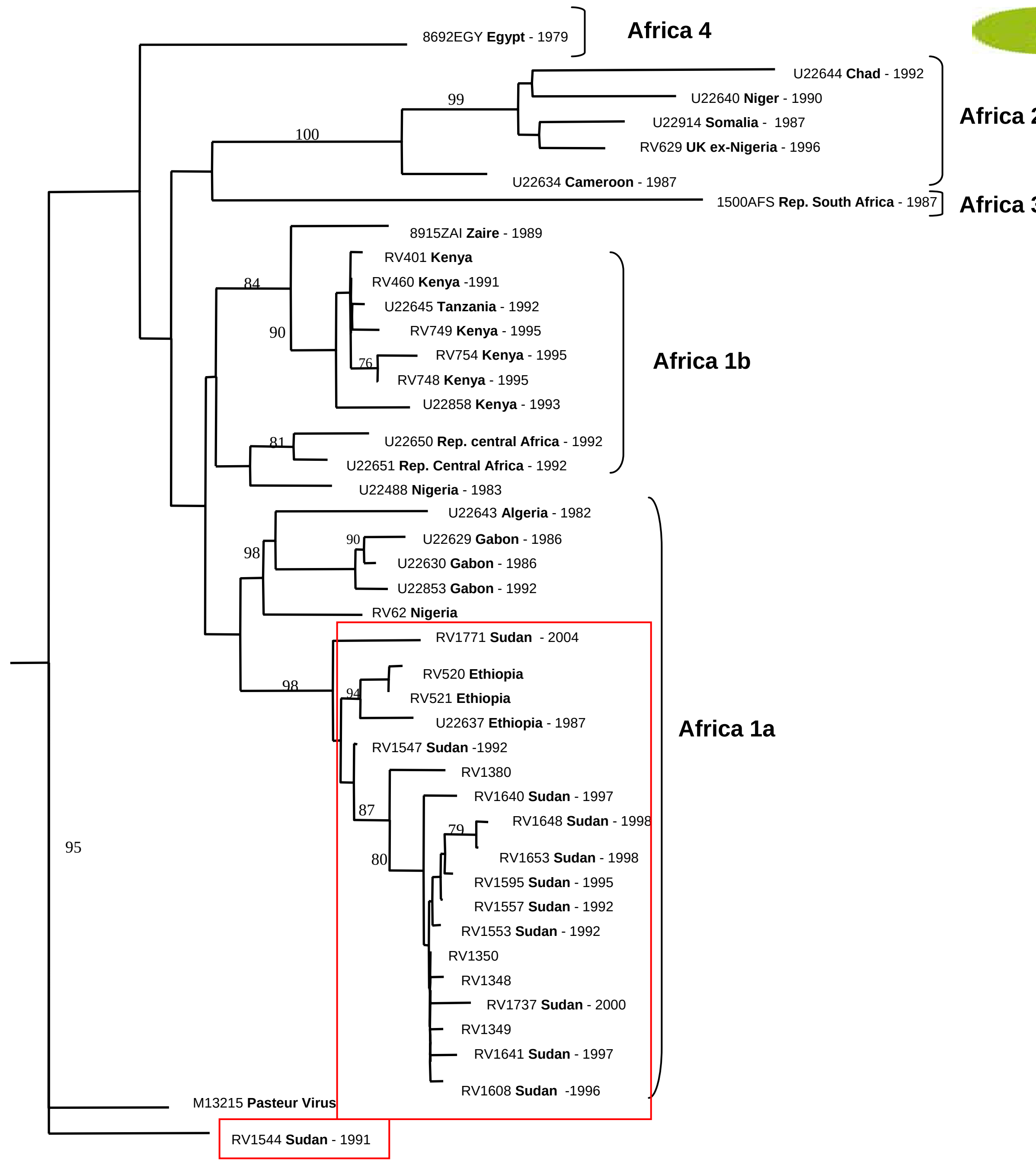
Sudan

- North – East Africa
- Landlocked
- Largest country in Africa

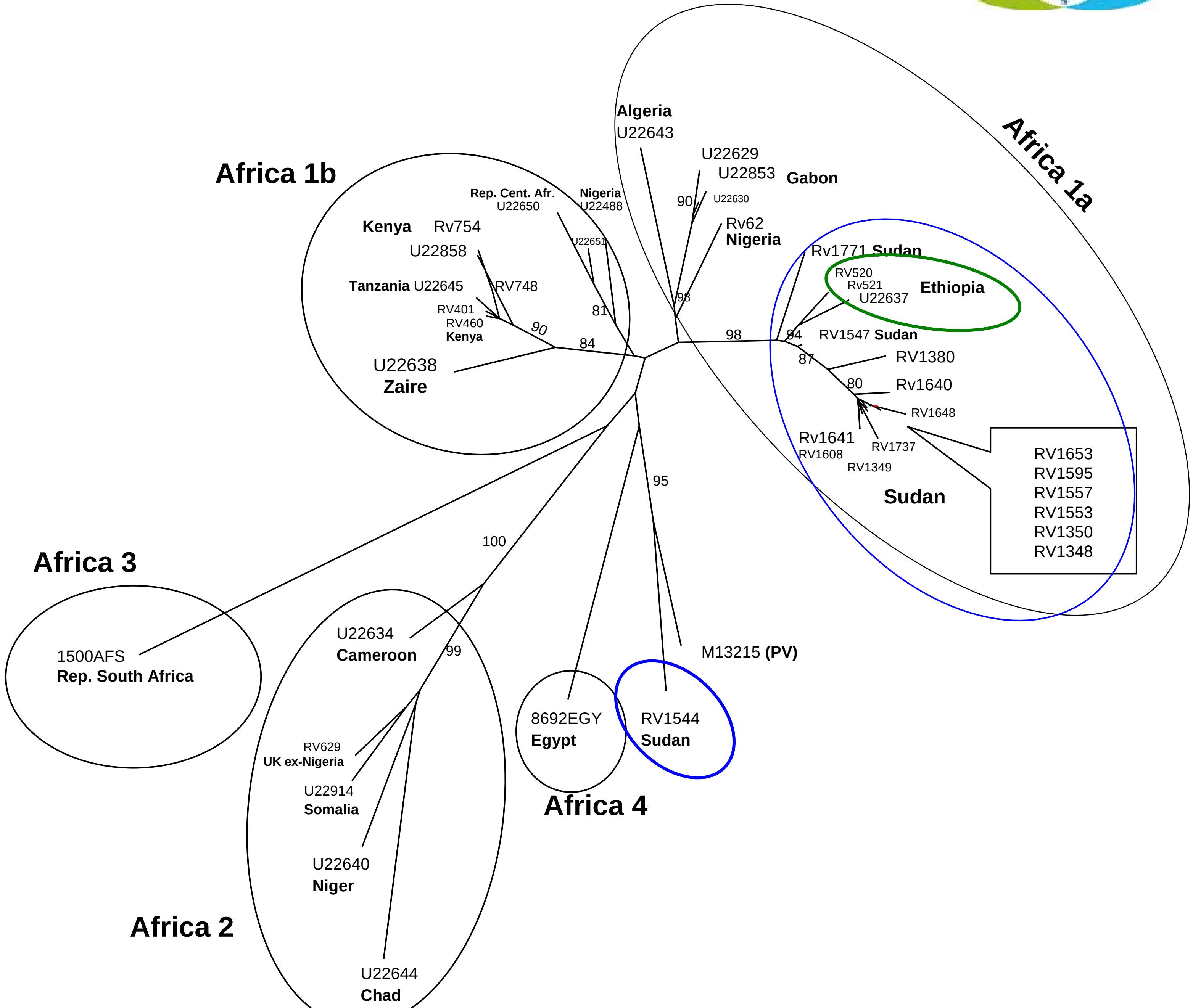
N-gene analysis

- A panel of 50 selected Sudanese RABV isolates:cat, dog, cattle, donkey, goat, camel (1991 – 2004)
- Canid Sudanese RABV isolates *(Johnson et al., 2004)* (1979-2001) & Non-dog isolates (Africa group 1a and 1b)
- Sequence of RABV isolates from group 3 and 4 (1979 – 1996)

  (Kissi, Tordo and Bourhy, 1995)

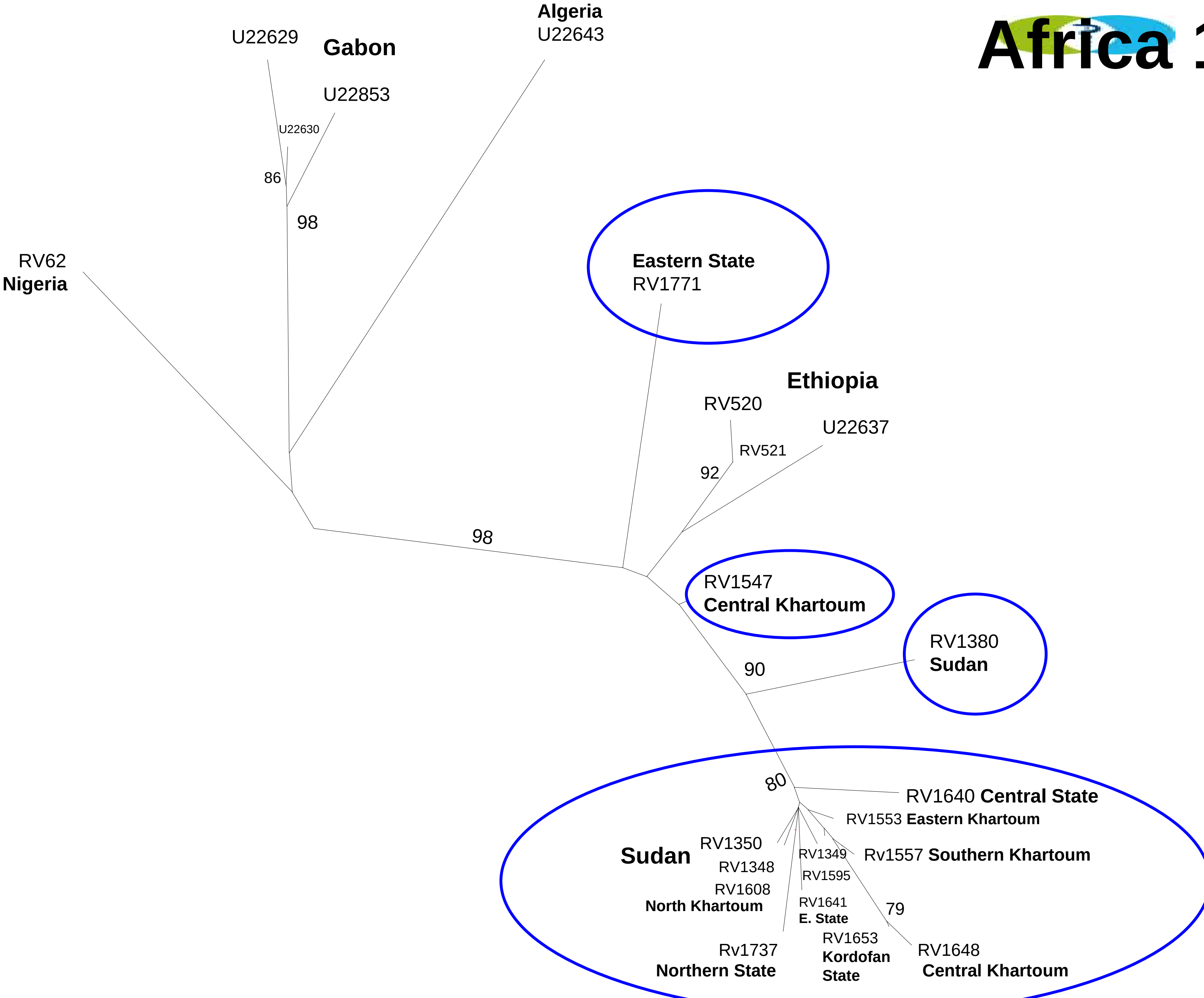


8692EGY Egypt - 1979
Africa 4
U22644 Chad - 1992
U22640 Niger - 1990
U22914 Somalia - 1987
RV629 UK ex-Nigeria - 1996
U22634 Cameroon - 1987
Africa 2
1500AFS Rep. South Africa - 1987
Africa 3
8915ZAI Zaire - 1989
RV401 Kenya
RV460 Kenya -1991
U22645 Tanzania - 1992
RV749 Kenya - 1995
RV754 Kenya - 1995
RV748 Kenya - 1995
U22858 Kenya - 1993
U22650 Rep. central Africa - 1992
U22651 Rep. Central Africa - 1992
U22488 Nigeria - 1983
Africa 1b
U22643 Algeria - 1982
U22629 Gabon - 1986
U22630 Gabon - 1986
U22853 Gabon - 1992
RV62 Nigeria
RV1771 Sudan - 2004
RV520 Ethiopia
RV521 Ethiopia
U22637 Ethiopia - 1987
RV1547 Sudan -1992
RV1380
RV1640 Sudan - 1997
RV1648 Sudan - 1998
RV1653 Sudan - 1998
RV1595 Sudan - 1995
RV1557 Sudan - 1992
RV1553 Sudan - 1992
RV1350
RV1348
RV1737 Sudan - 2000
RV1349
RV1641 Sudan - 1997
RV1608 Sudan -1996
Africa 1a
M13215 Pasteur Virus
RV1544 Sudan - 1991
100
99
84
90
76
81
98
90
98
94
87
79
80
95



Africa 1a
Africa 1b
Africa 2
Africa 3
Africa 4
Algeria
U22643
U22629
U22853
Gabon
90
U22630
Rv62
Nigeria
98
Rep. Cent. Afr.
U22650
Nigeria
U22488
U22651
81
84
Kenya
Rv754
U22858
Tanzania U22645
RV748
RV401
RV460
Kenya
90
U22638
Zaire
98
Rv1771 Sudan
RV520
Rv521
U22637
Ethiopia
94
RV1547 Sudan
87
RV1380
80
Rv1640
RV1648
Rv1641
RV1608
RV1737
RV1349
Sudan
RV1653
RV1595
RV1557
RV1553
RV1350
RV1348
95
100
M13215 (PV)
1500AFS
Rep. South Africa
U22634
Cameroon
99
RV629
UK ex-Nigeria
U22914
Somalia
U22640
Niger
U22644
Chad
8692EGY
Egypt
RV1544
Sudan

Africa 1a



Algeria
U22643
U22629
Gabon
U22853
U22630
86
98
RV62
Nigeria
Eastern State
RV1771
Ethiopia
RV520
U22637
RV521
92
98
RV1547
Central Khartoum
RV1380
Sudan
90
80
RV1640 Central State
RV1553 Eastern Khartoum
Sudan
RV1350
RV1348
RV1608
North Khartoum
RV1349
RV1595
Rv1557 Southern Khartoum
RV1641
E. State
79
RV1653
Kordofan
State
Rv1737
Northern State
RV1648
Central Khartoum

Results

- Sudanese isolates average sequence identity %
  - nt: 95.5 to 99.8%
  - aa: 95.6 to 99.3%
- Almost all Sudanese isolates → Africa group 1a except RV1544
  - Sudanese RABV can be divided into 4 main clusters (5 with RV1544)
  - RV1544 (isolated from cattle in Central Khartum, 1991)
  - Closely related to 8692EGY (human isolates, Egypt, 1979) – Africa 4
  - Vaccine-related strain? Genetic homology with PV

Discussion

- Results further support the link between RABV in Sudan and Ethiopia

- Relationship between
RV1544 (Sudan) & 8692EGY (Egypt)
geographical factor – cattle trade?

- Wide range of host species
contribute to movement of rabies
in Sudan

- Dog-mediated rabies poses the largest problem

- Control strategies should focus on controlling rabies in domestic dogs

Acknowledgements

- **Botswana:** Letshwenyo M., Baipoledi, E.K., Thobokwe, G
- **Sudan:** Ali Y. H., Intisar K. Saeed
- **Namibia:** Hübschle, O., Mettler, F.,
- **RSA:** Sabeta, C., Nel, L.H.


Thank you

World Health Organization

From Gro Harlem Brundtland:

“In a modern world, bacteria and viruses travel almost as fast as money. With globalization, a single microbial sea washes all of humankind. There are no health sanctuaries.”

Classical rabies diagnostics and new developments

Wanda Markotter

University of Pretoria
South Africa

University of Pretoria

Diagnosis of rabies must be rapid and accurate in order for timely administration of PEP

Diagnosis of rabies must be rapid and accurate in order for timely administration of PEP

Diagnosis of rabies aid in defining epidemiological patterns of disease and this aid in designing control programs

Diagnostic tests

- **Antigen detection**
- **Antibody detection**

Methods
for antigen detection

- Fluorescent antibody (FA) test
- Virus isolation
- ELISA
- Histopathological examination
- Immunohistochemistry
- Direct rapid immunohistochemistry test (dRIT)
- Molecular methods – RT PCR

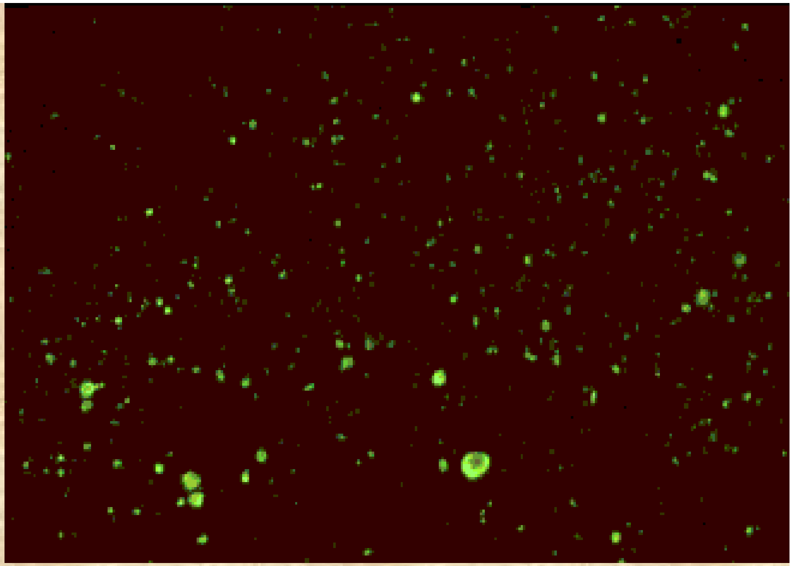
Methods
for antibody detection

Rapid fluorescent focus inhibition test (RFFIT)

Fluorescent antibody virus neutralization test (FAVN)

ELISA

Gold standard for Rabies diagnostics

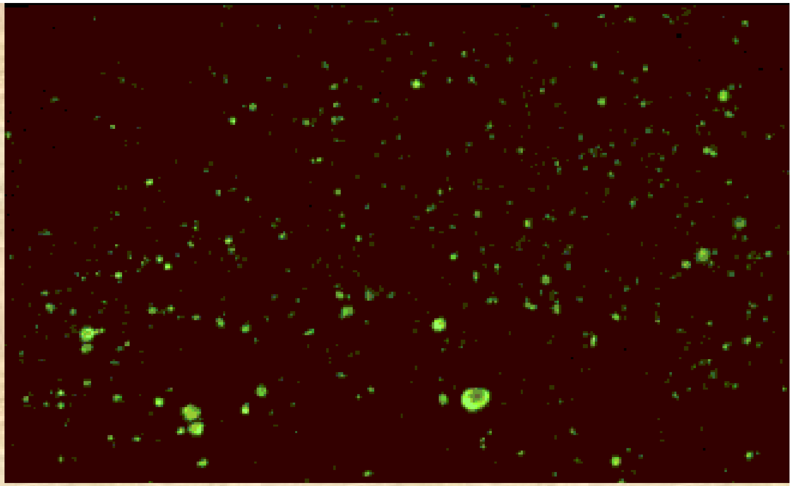


- **Fluorescent antibody technique (FA test)**

Principle

Microscopic examination of fixed brain impressions after being treated with anti-rabies serum or globulin conjugated to fluorescein isothiocyanate

Gold standard



- Fluorescent antibody technique (FA test)

- Success of the test depends upon:
- Expertise of the examiner
- Quality of the conjugate
- The fluorescent microscope

Conjugates

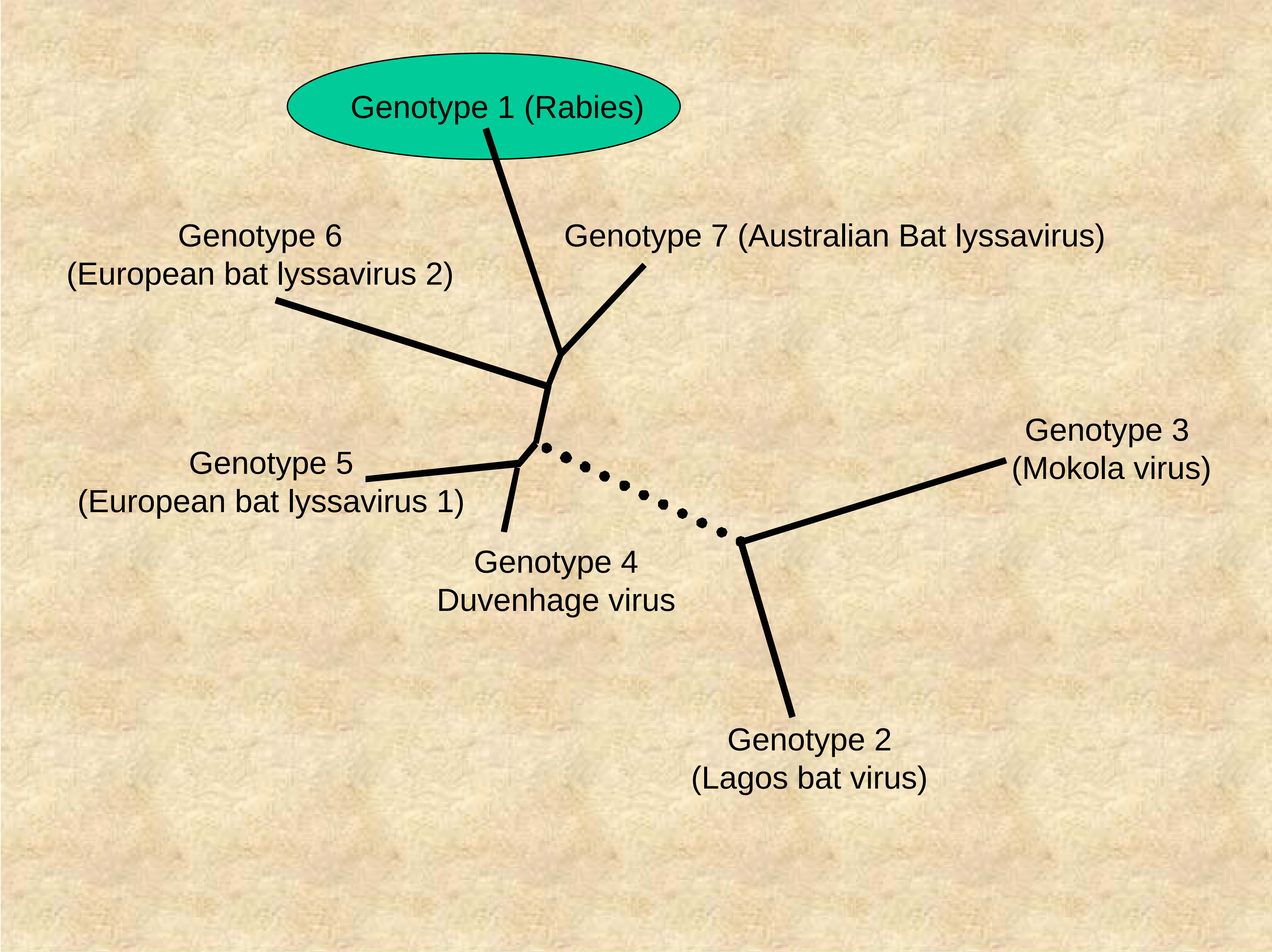
- Quality

-- Detect all lyssavirus genotypes

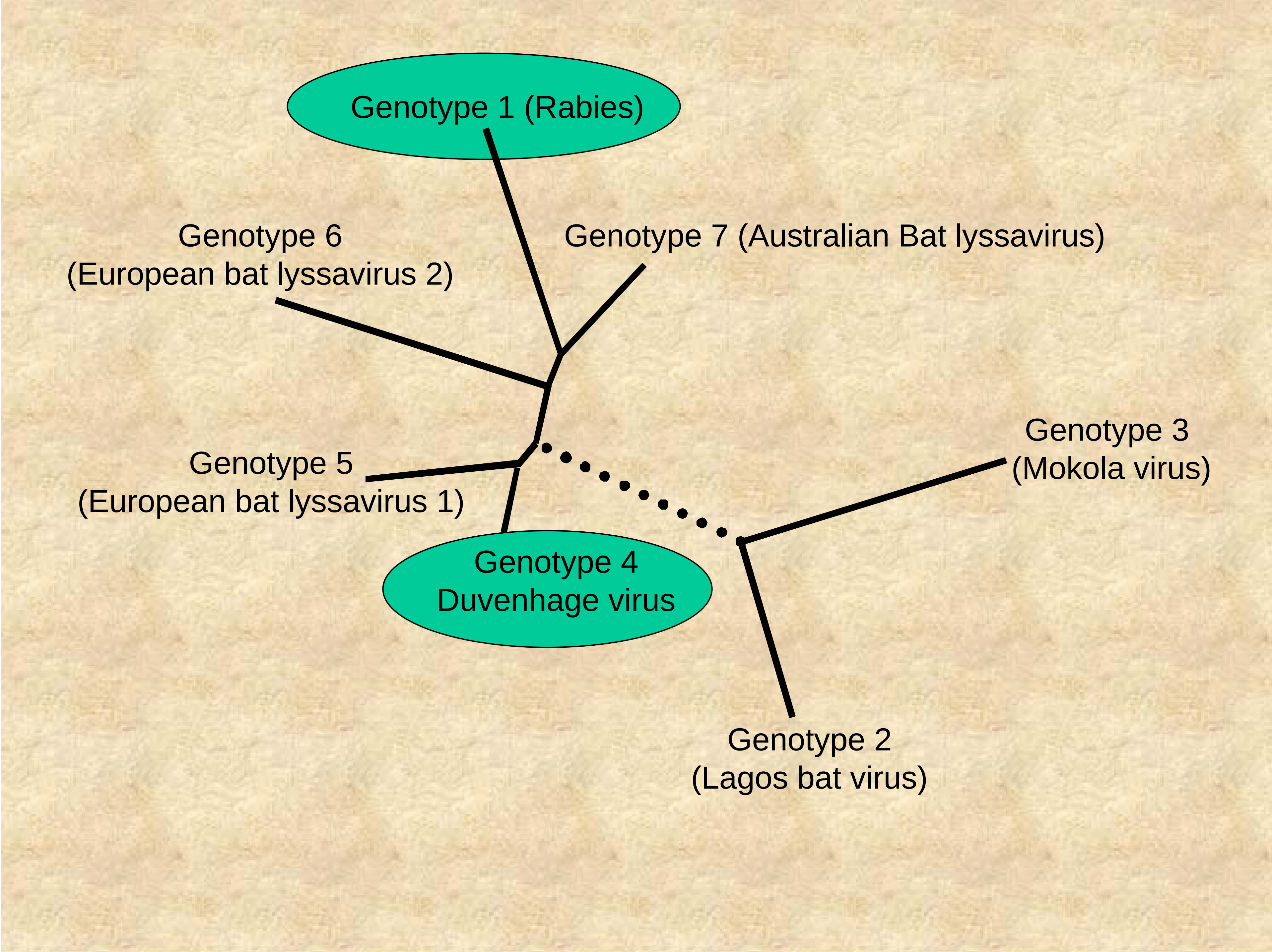
Conjugates

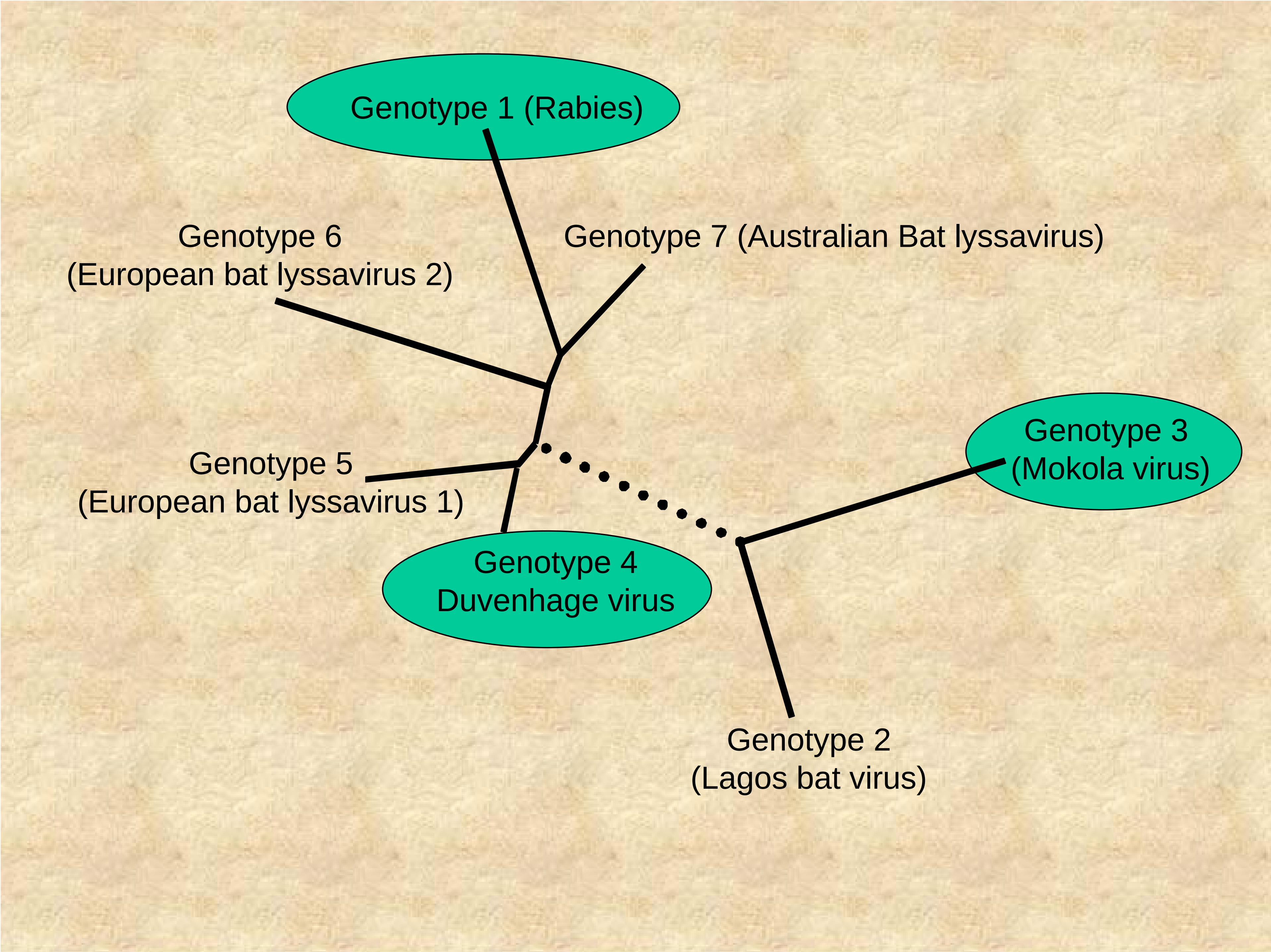
- Commercially available
- polyclonal and monoclonal
- Detect all lyssavirus genotypes

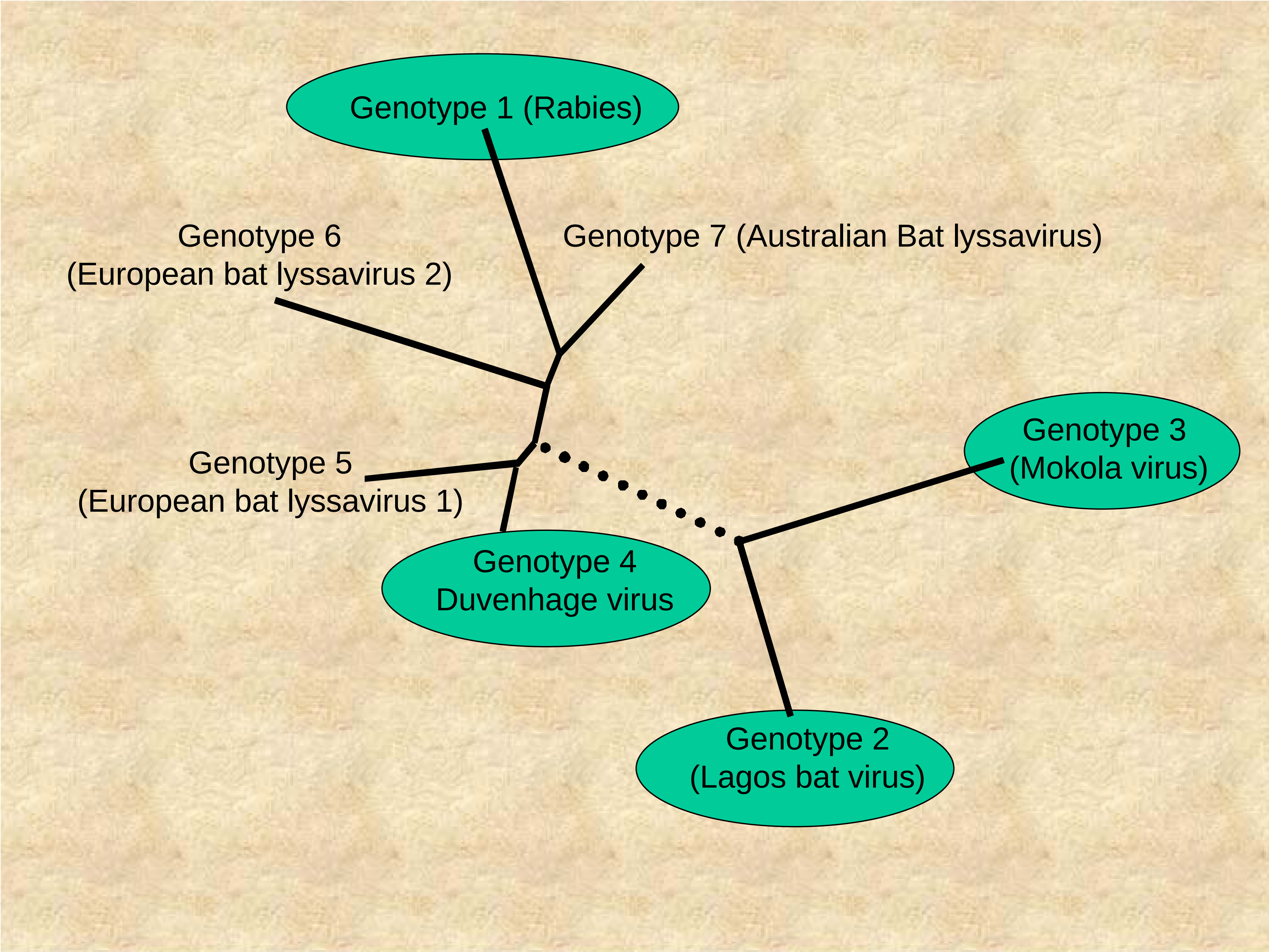
- Self manufactured
- Test the quality
- Ensure that it detect all genotypes



Genotype 1 (Rabies)
Genotype 6
(European bat lyssavirus 2)
Genotype 7 (Australian Bat lyssavirus)
Genotype 3
(Mokola virus)
Genotype 5
(European bat lyssavirus 1)
Genotype 4
Duvenhage virus
Genotype 2
(Lagos bat virus)


Genotype 1 (Rabies)
Genotype 6
(European bat lyssavirus 2)
Genotype 7 (Australian Bat lyssavirus)
Genotype 5
(European bat lyssavirus 1)
Genotype 3
(Mokola virus)
Genotype 4
Duvenhage virus
Genotype 2
(Lagos bat virus)


Genotype 1 (Rabies)
Genotype 6
(European bat lyssavirus 2)
Genotype 7 (Australian Bat lyssavirus)
Genotype 3
(Mokola virus)
Genotype 5
(European bat lyssavirus 1)
Genotype 4
Duvenhage virus
Genotype 2
(Lagos bat virus)


Genotype 1 (Rabies)
Genotype 6
(European bat lyssavirus 2)
Genotype 7 (Australian Bat lyssavirus)
Genotype 5
(European bat lyssavirus 1)
Genotype 3
(Mokola virus)
Genotype 4
Duvenhage virus
Genotype 2
(Lagos bat virus)

Virus isolation

Confirm or further characterization

Cell culture (Neuroblastomas) or IC mice inoculation

Cell culture is more rapid

Samples

- Rabies diagnosis in animals
- Rabies diagnosis in humans

Rabies diagnosis in animals

- Brain material – post mortem
- Can perform the FA test

Rabies diagnosis in humans

Diagnose rabies before death - More than one test necessary

Clinical diagnosis is unreliable

Any secretions and body fluids can be used to diagnose rabies during lifetime

Negative result does not rule out rabies

Perform the FA test on skin biopsies

Virus isolations in mice or cell culture

RT-PCR on secretions and body fluids

Test for neutralizing antibodies in serum or spinal fluid

Post mortem – Perform FA on brain material

Characterization of viruses

- Epidemiological data
- Provide information about the genotype (species) of the virus
- Provide information about the variant of the virus

Characterization of viruses

- Monoclonal antibodies
- RT-PCR and DNA sequencing

	Canid Biotype (GT1)	Mongoose Biotype (GT1)	Lagos bat virus (GT2)	Mokola virus (GT3)	Duvenhage virus (GT4)
1	-	-	-	-	-
2	+++	var	-	-	-
3	+++	var	-	-	-
4	var	var	-	-	-
5	+++	+++	+++	+++	+++
6	-	-	+++	-	-
7	-	-	-	-	+++
8	-	var	-	-	+++
9	+++	-	-	-	+++
10	-	-	-	+++	-
11	-	var	-	var	-
12	-	+++	-	-	-
13	-	var	-	-	-
14	++	var	-	-	-
15	++	var	-	-	-
16	-	var	-	-	+++

RT-PCR and DNA sequencing

- Primers sequences available in the literature
- DNA sequencing information will allow phylogenetic analysis
- Provide information for control programs

Formalin fixed tissue

- Formalin not recommended
- Limited diagnostic tests
- Transfer samples to absolute ethanol in less than seven days
- Can perform the FA test after proteolytic digestion

Example

- Mongoose involved in a possible vaccine failure in 2004 in South Africa
- Mongoose brain was in formalin for nine months before transferred to ethanol
- Could obtain DNA sequence using RT-PCR and DNA sequencing techniques

References

- www.cdc.gov
- WHO Laboratory techniques in rabies
- OIE manual for diagnostic tests and vaccines for terrestrial animals

Acknowledgements

- Prof Louis Nel (UP)
- Dr. Charles Rupprecht (CDC)
- Dr. Alex Wandeler (Canadian Food Inspection Agency)
- Pamela Yager (CDC)
- Lilian Orchiaro (CDC)
- Dr. Claude Sabeta (OVI)

Validation of a less invasive blood-sampling technique in rabies serology using reduviid bugs

Ad Vos [1], Thomas Müller [2], Larissa Neubert [1] & Christian Voigt [3]

1) IDT GmbH, PSF 214, 06855 Rosslau – GERMANY

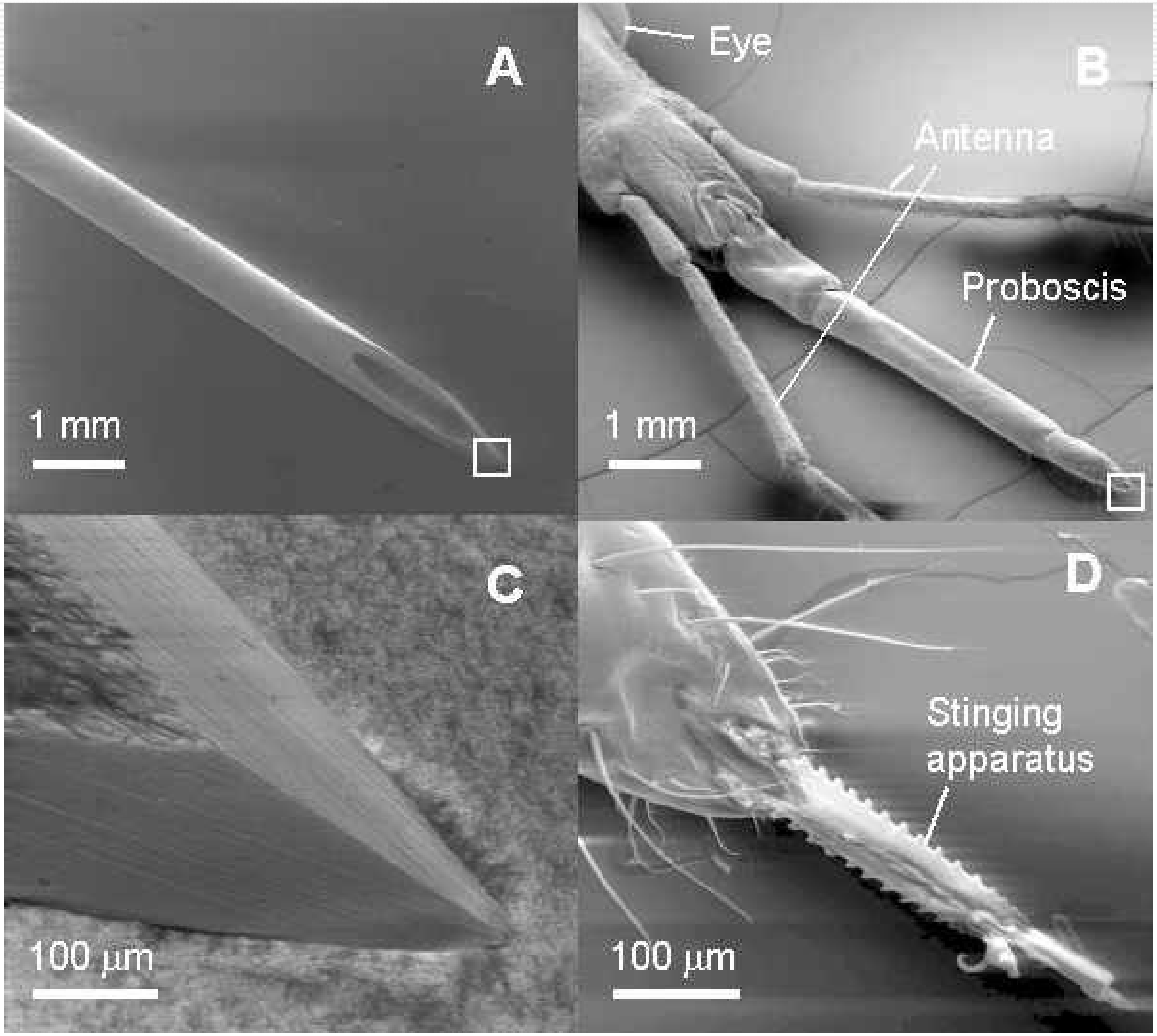
2) FLI, Seestrasse 55, 16868 Wusterhausen – GERMANY

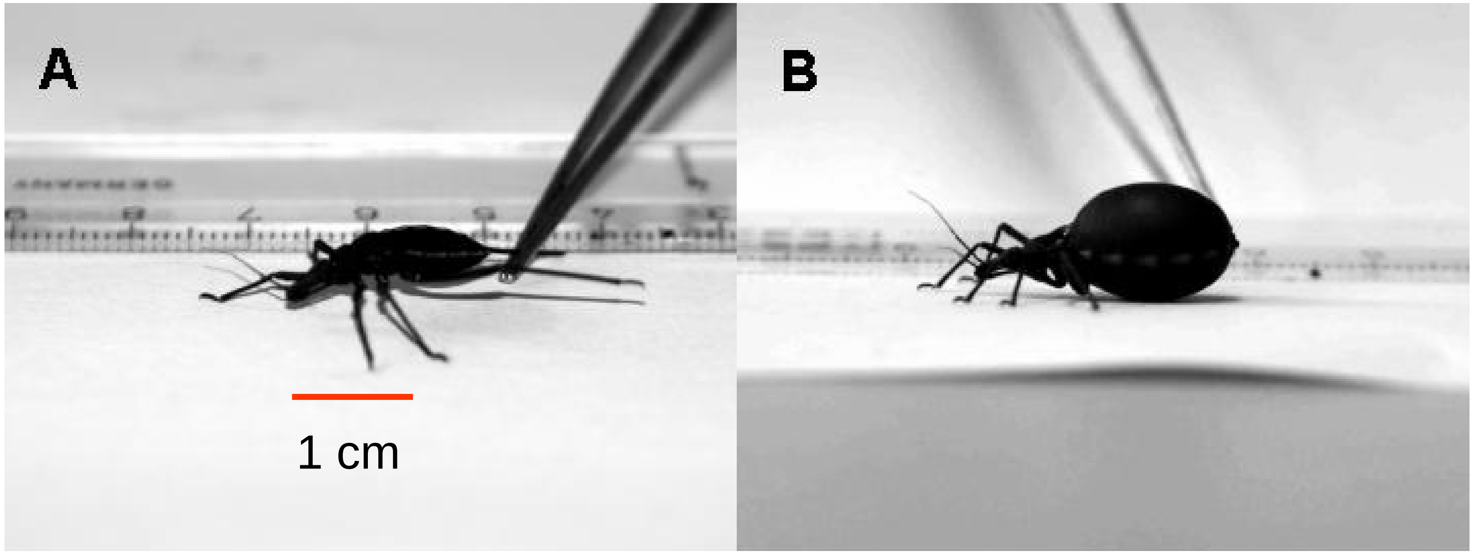
3) IZW, Alfred-Kowalke Strasse 17, 10315 Berlin - GERMANY

Major advantages:

- possibility to obtain blood from animals with limited accessibility (conventional methods)
- reduction of stress (and injuries)
- possible elimination of anaesthesia

Already used for endocrinological (progestrone, testerone) and serological studies (RHDV antibodies) in mammals (incl. bats).


A
B
C
D
Eye
Antenna
Proboscis
Stinging
apparatus
1 mm
1 mm
100 µm
100 µm



Two species tested:

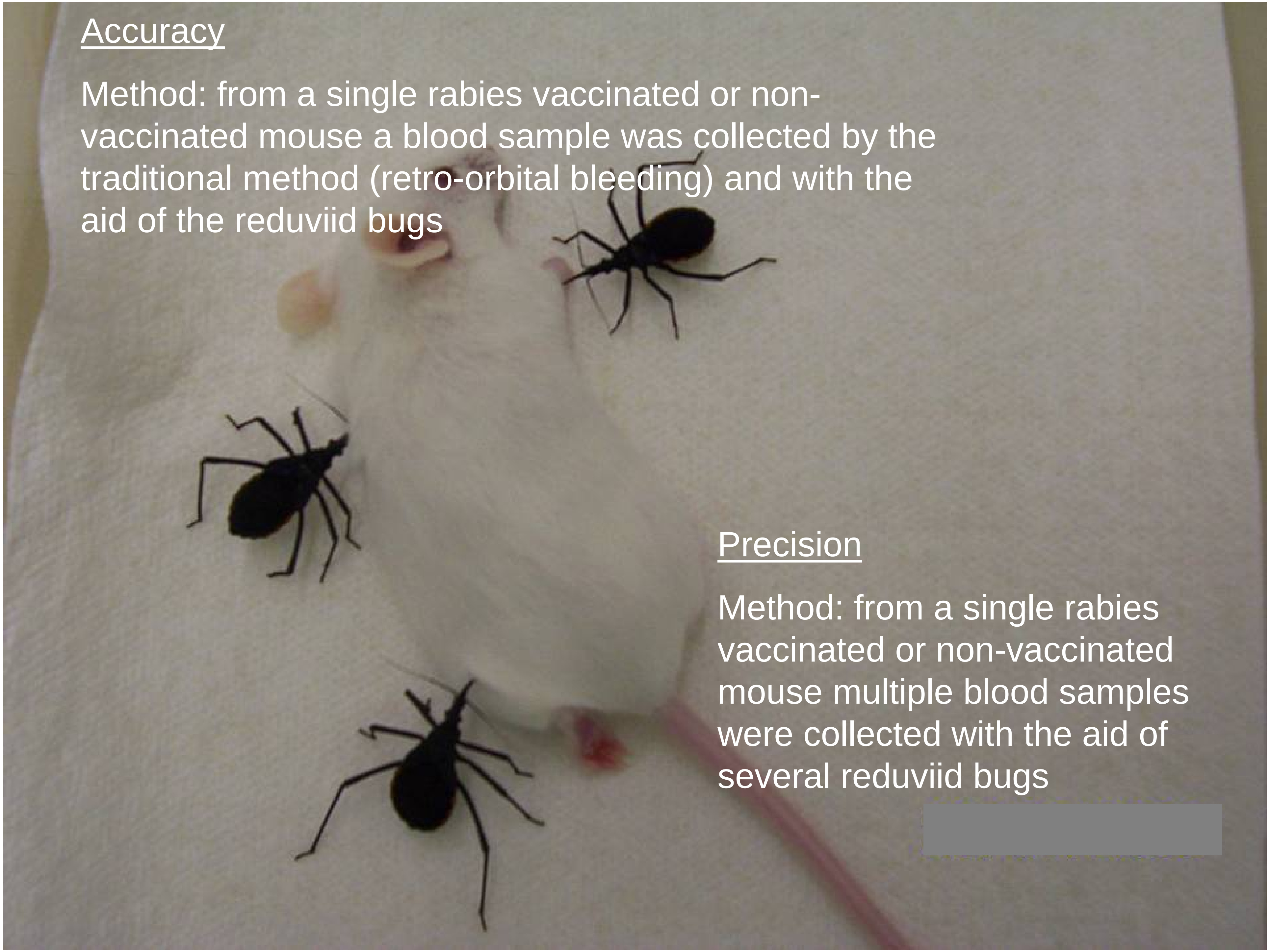
Dipetalogaster maximus & Rhodnius prolixus

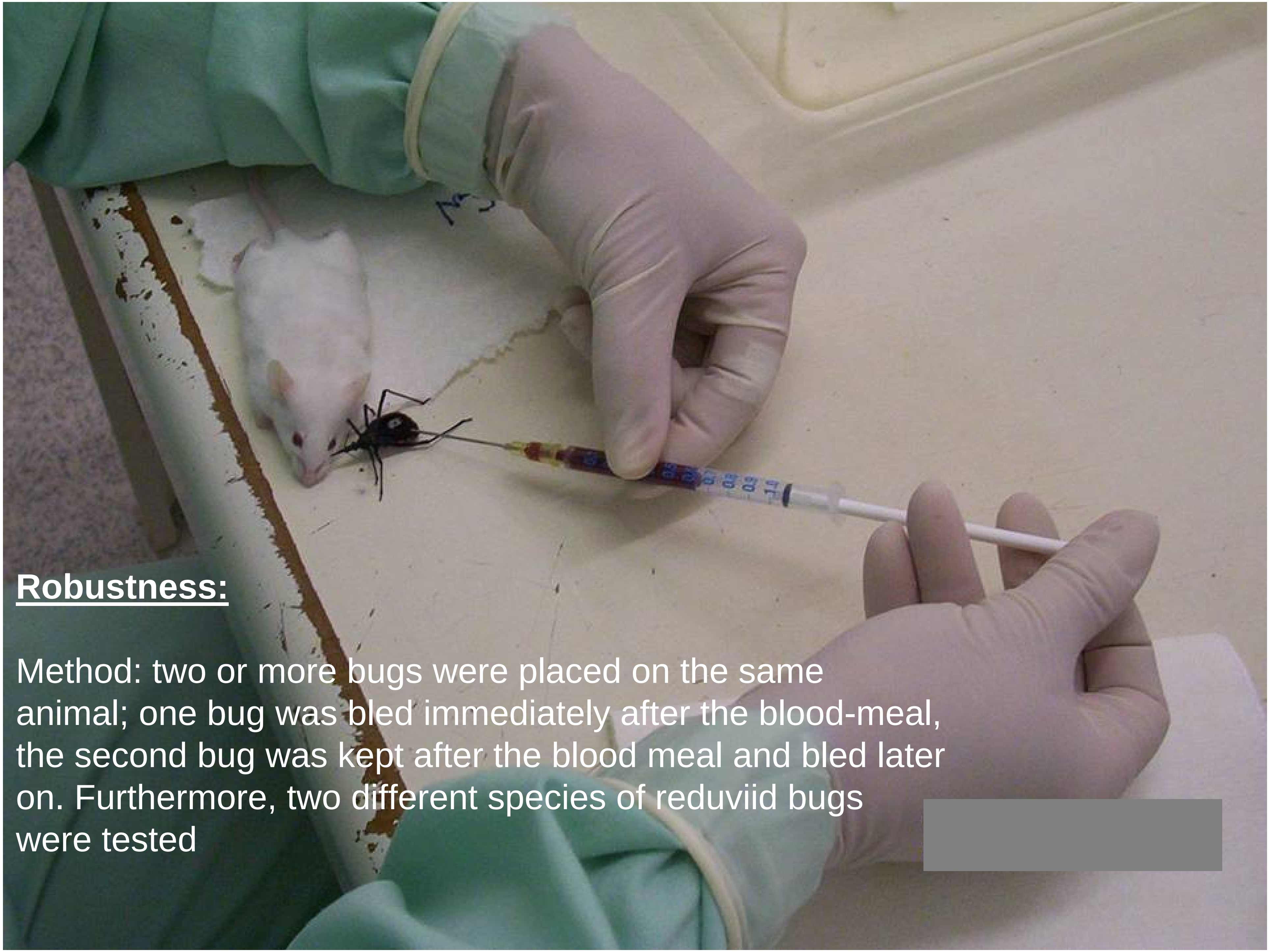
Validation parameters for analytical procedures
(adapted after ICH Q2A)

- **accuracy**
  - expresses the closeness of agreement between the value obtained using a conventional accepted method and the value found with the method under investigation

- **precision**
  - expresses the closeness of agreement between a series of measurements obtained from multiple sampling of the same homogeneous sample

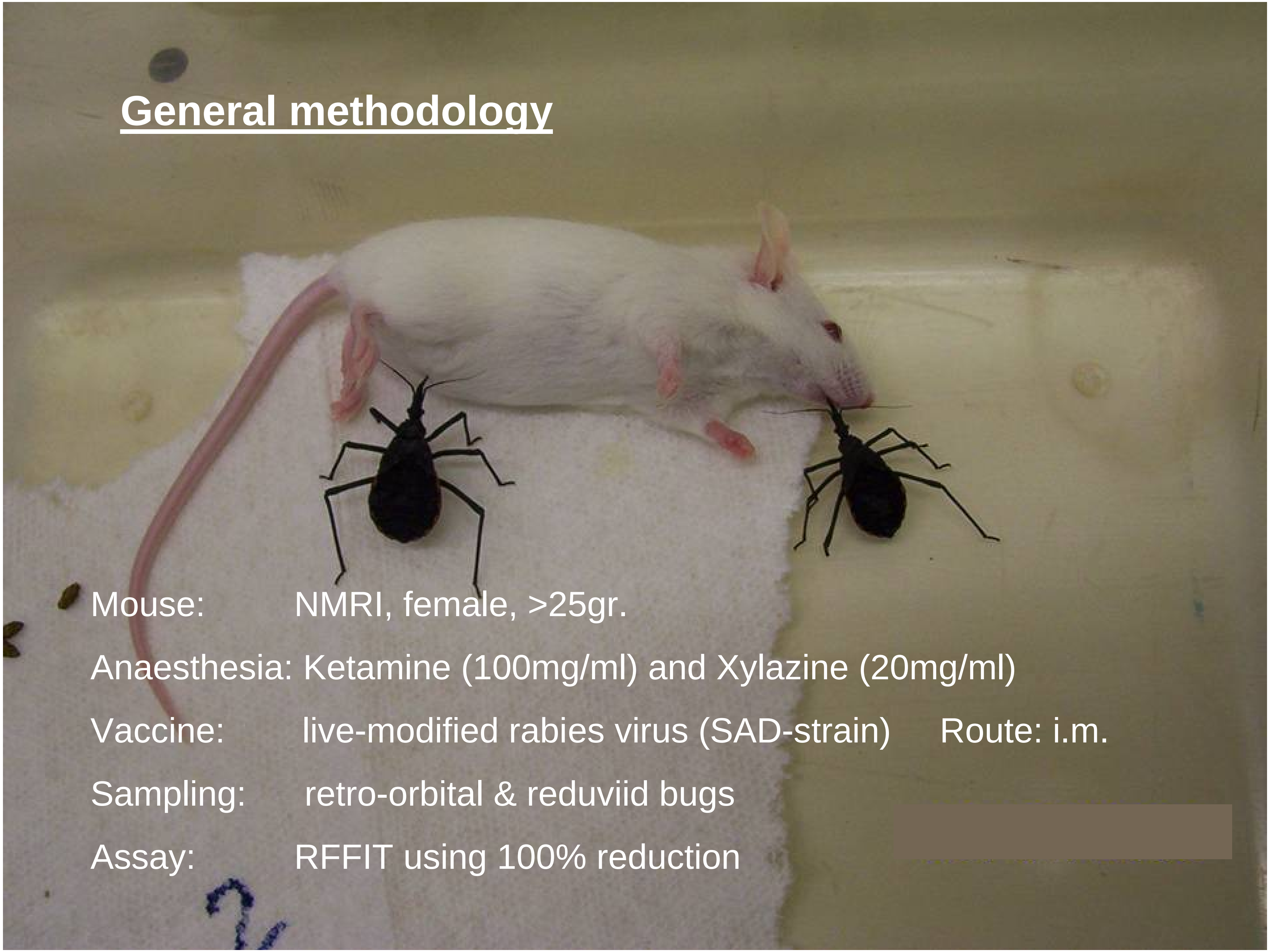
- **robustness**
  - is a measure of the capacity of an analytical procedure to remain unaffected by small deliberate variation in method parameters


Accuracy
Method: from a single rabies vaccinated or non-vaccinated mouse a blood sample was collected by the traditional method (retro-orbital bleeding) and with the aid of the reduviid bugs
Precision
Method: from a single rabies vaccinated or non-vaccinated mouse multiple blood samples were collected with the aid of several reduviid bugs



Robustness:

Method: two or more bugs were placed on the same animal; one bug was bled immediately after the blood-meal, the second bug was kept after the blood meal and bled later on. Furthermore, two different species of reduviid bugs were tested



General methodology

Mouse: NMRI, female, >25gr.

Anaesthesia: Ketamine (100mg/ml) and Xylazine (20mg/ml)

Vaccine: live-modified rabies virus (SAD-strain) Route: i.m.

Sampling: retro-orbital & reduviid bugs

Assay: RFFIT using 100% reduction

Results: Accuracy

Non-vaccinated mice – titre (IU/ml)

Nr.	r.o.	bug*	Nr	r.o.	bug*
1	<0.5	<0.5	7	<0.5	<0.5
2	<0.5	<0.5	8	<0.5	<0.5
3	<0.5	<0.5	9	<0.5	<0.5
4	<0.5	<0.5	10	<0.5	<0.5
5	<0.5	<0.5	11	<0.5	<0.5
6					.

* - sometimes more than one bug was placed on a mouse. In this case, the first bug removed was used for this comparison.

Results: Accuracy

Vaccinated mice – titre IU/ml

Nr.	r.o.	bug*	Nr	r.o.	bug*
1	2.0	2.0	10	8.0	8.0
2	4.0	2.0	11	1.0	1.0
3	2.0	2.0	12	16.0	30.0
4	0.5	<0.5	13	8.0	8.0
5	8.0	4.0	14	2.0	2.0
6	8.0	8.0	15	1.0	2.0
7	2.0	2.0	16	2.0	1.0
8	2.0	2.0	17	16.0	30.0
9	4.0	2.0	18	4.0	2.0 .

* - sometimes more than one bug was placed on a mouse. In this case, the first bug removed was used for this comparison.

Results: Precision

Mice – titre IU/ml

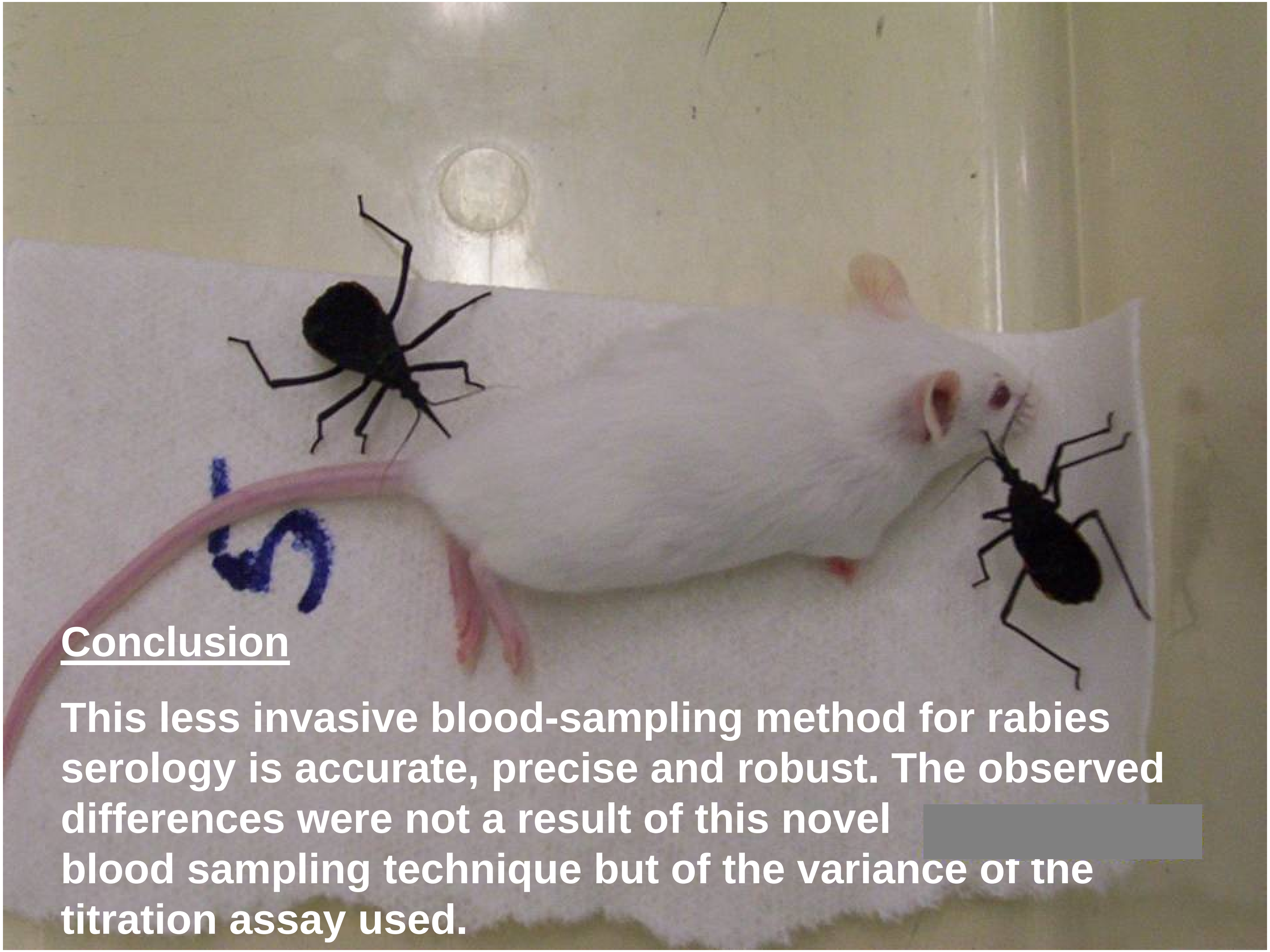
Nr.	bug 1	bug 2	bug 3	bug 4	Nr.	bug 1	bug 2	bug 3	bug 4
1	<0.5	<0.5			10	8.0	8.0		
2	<0.5	<0.5			11	30.0	8.0		
3	<0.5	<0.5			12	8.0	16.0		
4	<0.5	<0.5			13	2.0	2.0	1.0	
5	<0.5	<0.5	<0.5		14	2.0	4.0		
6	4.0	8.0	4.0		15	2.0	<0.5*		
7	8.0	8.0			16	1.0	2.0	1.0	2.0
8	2.0	4.0			17	30.0	30.0		
9	2.0	2.0			18	2.0	2.0	2.0	4.0 .

* - about half of liquid drawn from bug was body fluid

Results: Robustness
Mice – titre IU/ml

Nr.	bug 1 0 hr.	bug 2 0.5 hr.	bug 3 2 hr.	bug 4 4 hr.
1			2.0	4.0
2			<0.5	<0.5
3	0.5		<0.5	
4	<0.5		<0.5	
5	2.0		2.0	
6	2.0		4.0	
7	8.0		8.0	
8	1.0		2.0	
9	8.0	2.0		
10	2.0	1.0		.

IDT



Conclusion

This less invasive blood-sampling method for rabies serology is accurate, precise and robust. The observed differences were not a result of this novel blood sampling technique but of the variance of the titration assay used.

Oral Immunization

Alexander I. Wandeler
Centre of Expertise for Rabies
Canadian Food Inspection Agency

Wildlife Rabies Control by Oral Immunization

oral rabies vaccination program

↓

percentage of individuals resisting infection increased

(increased HERD IMMUNITY)

↓

effective reproductive rate of disease
reduced below unity

HERD IMMUNITY

- required levels unknown
- 70% immunized individuals sufficient to control dog rabies
- different simulation models suggest levels between 50% and 100%
- varies with disease transmission dynamics in particular species and populations

oral rabies vaccines
presently applied for wildlife immunization

- ERA/SAD: live attenuated
  significant residual pathogenicity
  applied in Europe (1978) and Ontario (1989)

- SAG: live attenuated, escape mutant of SAD_{Berne}
  reduced pathogenicity
  applied in Switzerland and France

- VRG: live, vaccinia rabies-glycoprotein recombinant
  applied in Belgium (1988), France (1989),
  USA (1990), and Canada (1999)

Recombinant Vaccines

- Incorporation of rabies virus G-gene into vector virus genome
- Vector viruses: vaccinia
  human adenovirus type 5
  avian poxviruses
- G-protein expressed by cell infected with recombinant virus
- Vaccinated animal responds to G-protein and vector virus proteins

vaccine efficacy in target species

- small differences between species when vaccine given by intra-muscular injection
- significant differences between species when vaccine given by oral route
- immunity resulting from oral immunization protects against all rabies (genotype 1) virus variants

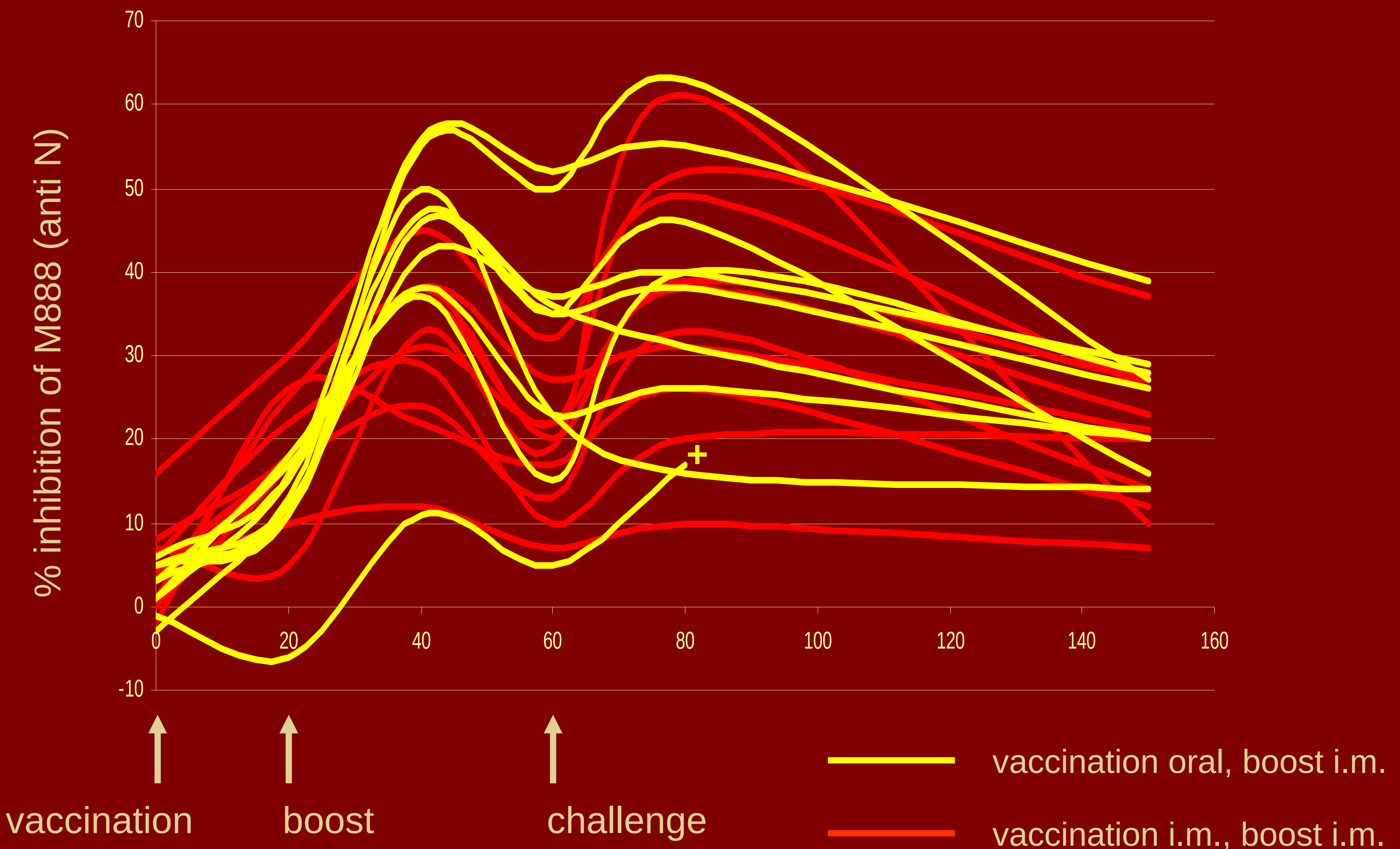
vaccine efficacy in target species

	ERA/SAD	vaccinia recombinants	adenovirus recombinants
Vulpes vulpes	+++	+++	++
Mephitis mephitis	+	+	+++
Procyon lotor	+	++	+++
Canis familiaris	+	++	?

Possible future developments

- Genetically engineered to improve the efficacy, target specificity, residual pathogenicity, …
- Combination with other antigens
- Killed vaccines and systems that facilitate their proper presentation to the immune system
- Not recommended: self-propagating recombinants

Immunization of skunks with recombinant HAV-5 rabies N

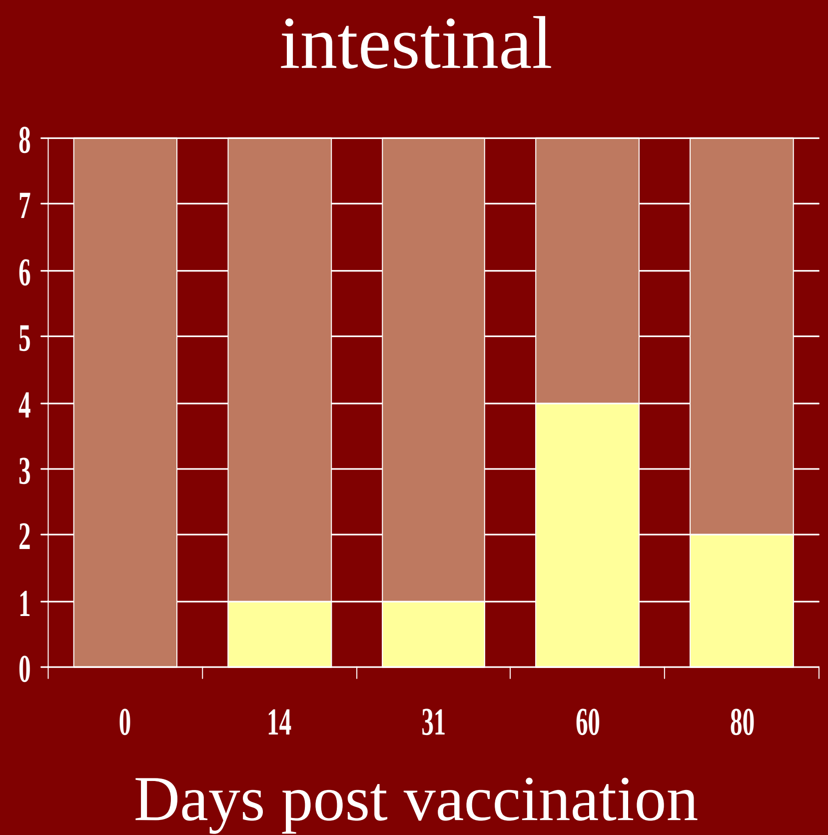


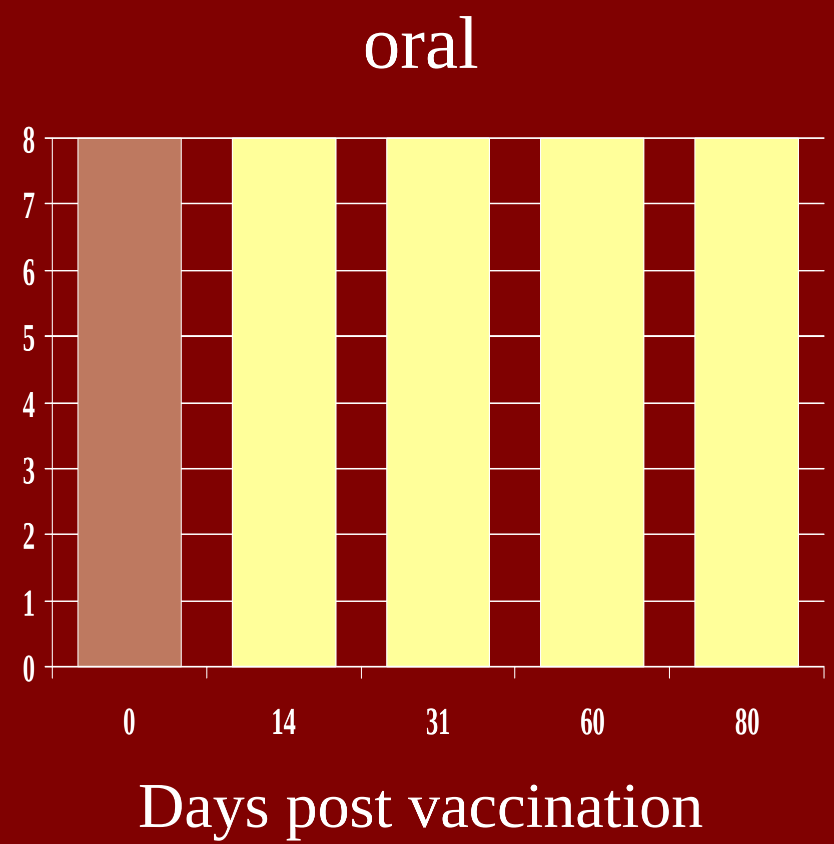
vaccine release onto target tissue of bait consumer

factors:
- live attenuated and recombinant vaccines must infect host tissues for proper antigen expression and presentation
- all live attenuated and some recombinant vaccines are inactivated by low pH and proteolytic enzymes

possible target tissues :
- oro-pharyngeal mucosa
- tonsils
- intestinal mucosa and Peyer's patches

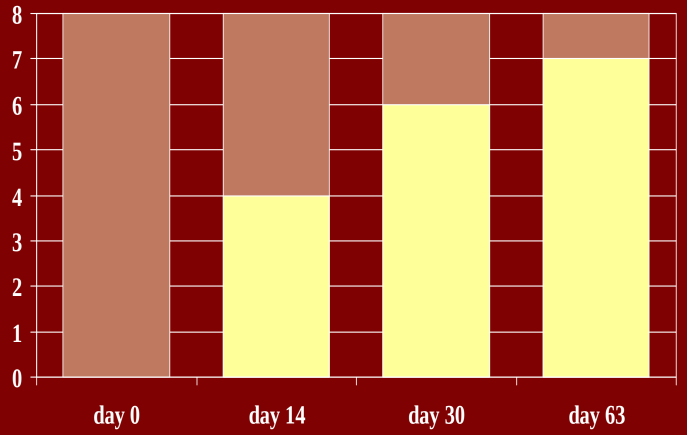
Seroconversion of skunks after application of $10^{9.5}TCID_{50}$ of Ad-RG1



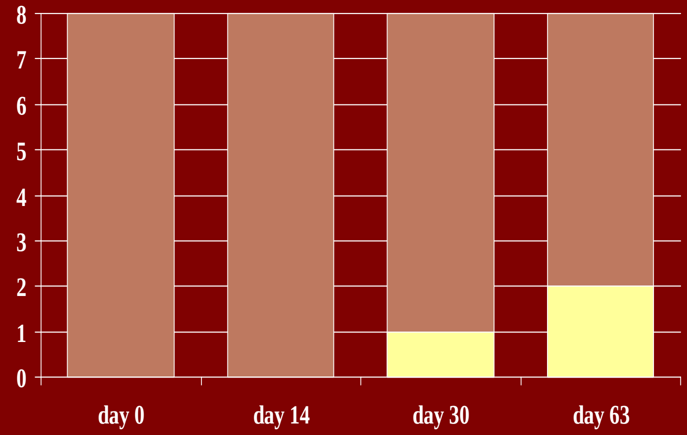


Seroconversion in skunks after exposure to Ad-RG1

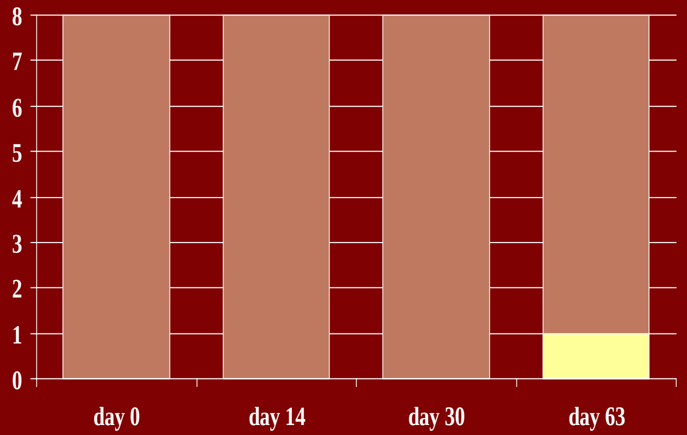
2ml by oral instillation



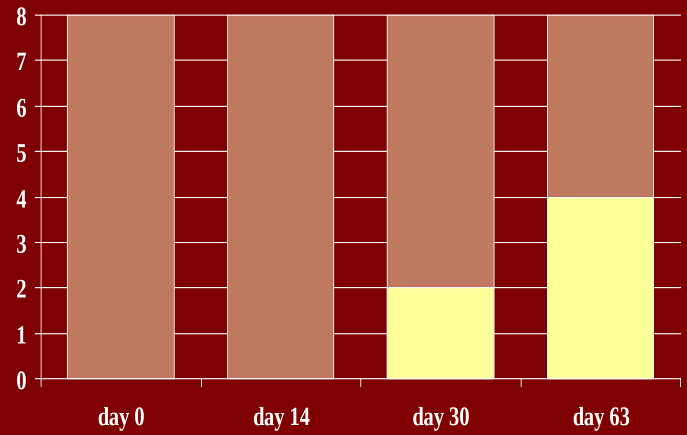
2ml in blister pack bait

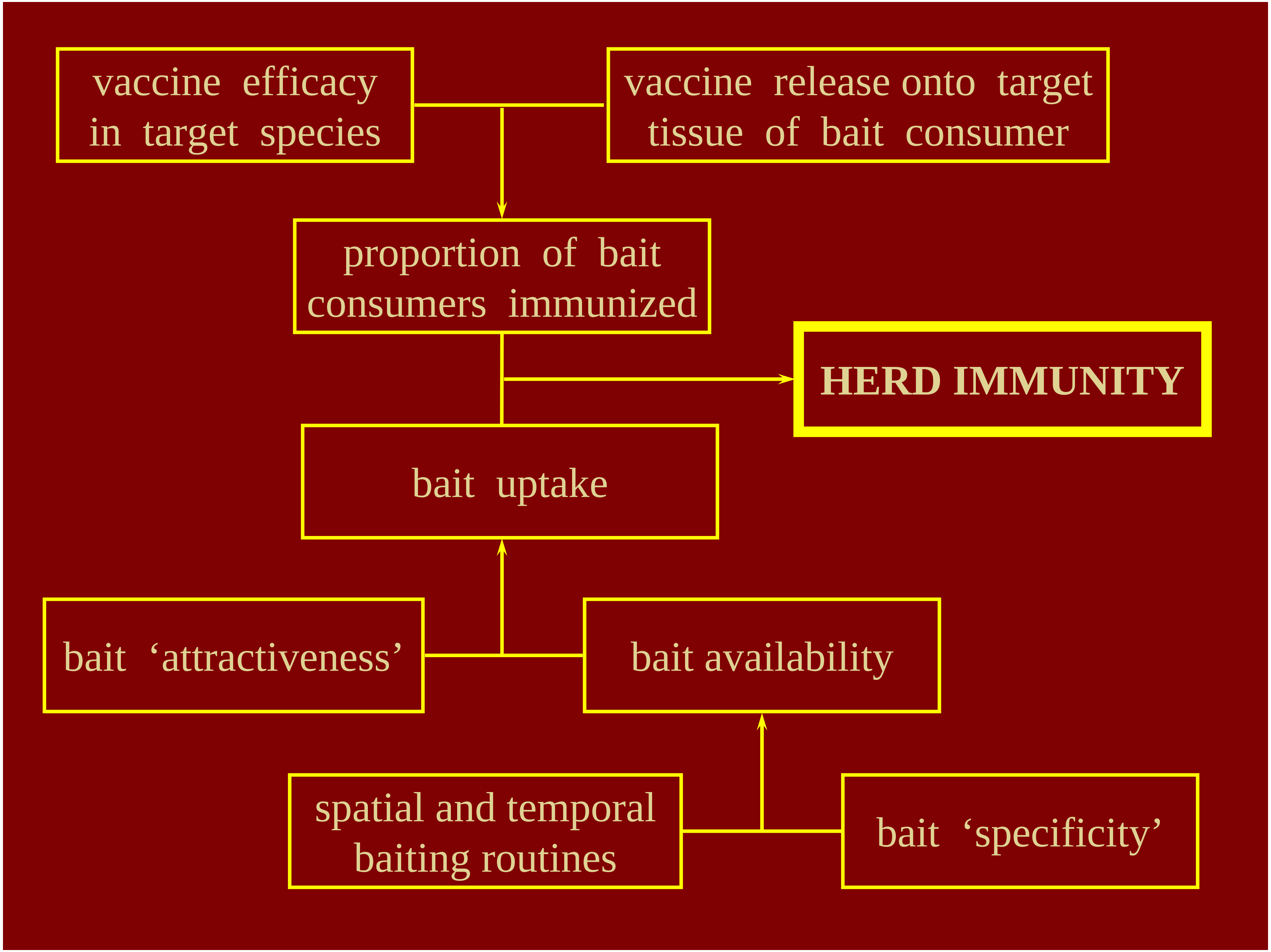


2ml in gelatin in Dupont bait



2ml in sponge bait

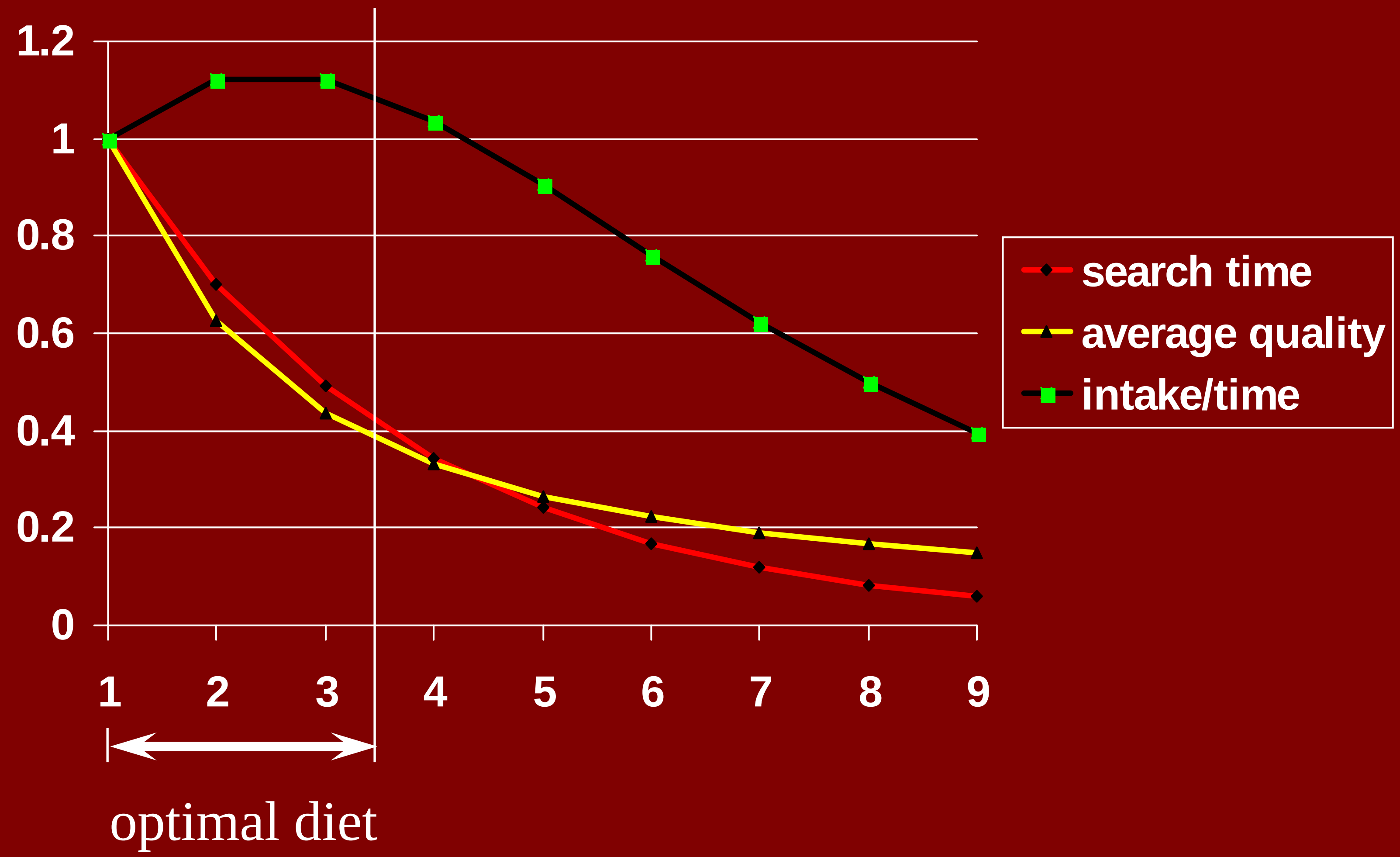



vaccine efficacy in target species
vaccine release onto target tissue of bait consumer
proportion of bait consumers immunized
HERD IMMUNITY
bait uptake
bait 'attractiveness'
bait availability
spatial and temporal baiting routines
bait 'specificity'

Thank you for your attention

bait ‘attractiveness’

and the concept of optimal foraging



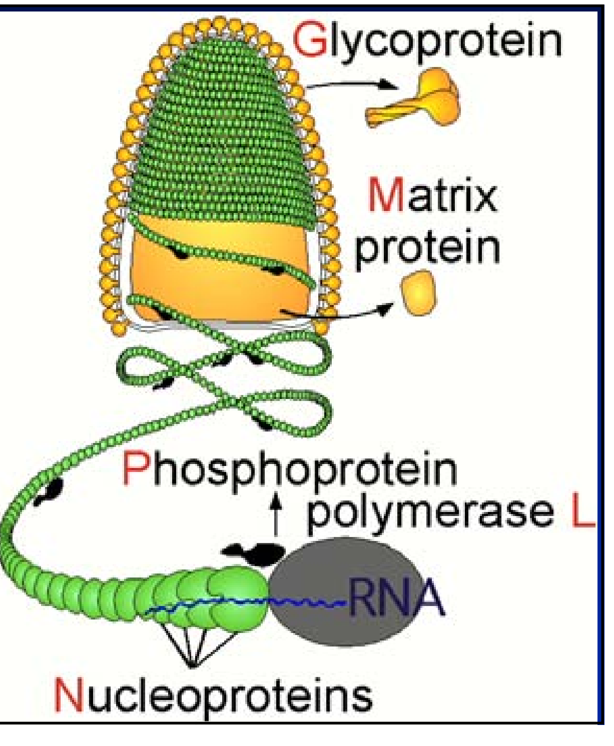
The value of research on new vaccines

Noël TORDO

INSTITUT PASTEUR

Rabies virus: the antigen

- Genus *Lyssavirus*
- Family *Rhabdoviridae*
- Order *Mononegavirales*
  - envelope
  - non-segmented (-) RNA
  - template: (RNA + N) = RNP
  - RNA-dependent RNA polymerase (L + P)

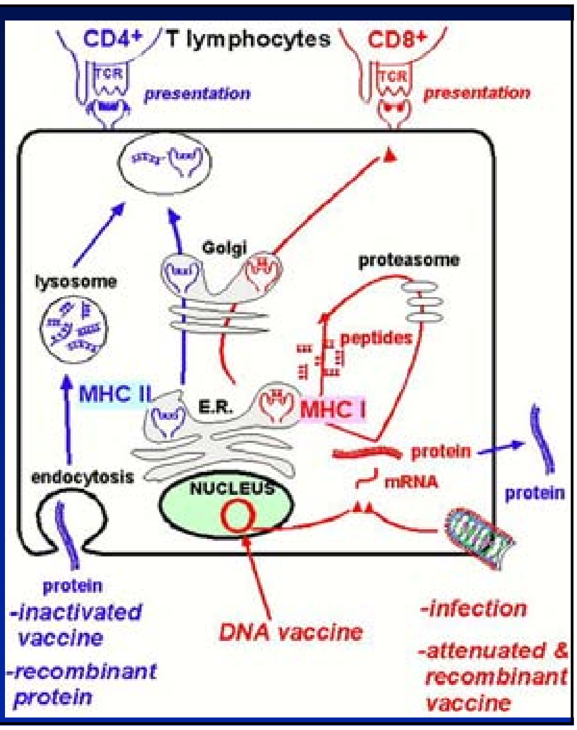


Immunogenicity of lyssavirus components

Antigen	*Neutralis. Ab*	*cell immunity* *Thelper*	*Tcytotox*	*Protection* *IC*	*IM*
VIRUS	+	+	+	+	+
RNP		+	+		+
Glyco G	+	+	+	+	+
Matrix M			+		
Nucleo N		+	+		+
Phospho P		+	+		
Polym L	?	?	?	?	?

Presentation of T antigens to lymphocytes:

- *« Replicative » antigens are more potent vaccine*



Tools for prevention/therapy

Pasteur's vaccine

rabid rabbit spinal cord
-> dessiccated

in 2005

human vaccines (prevention + therapy)

Not recommended
by WHO

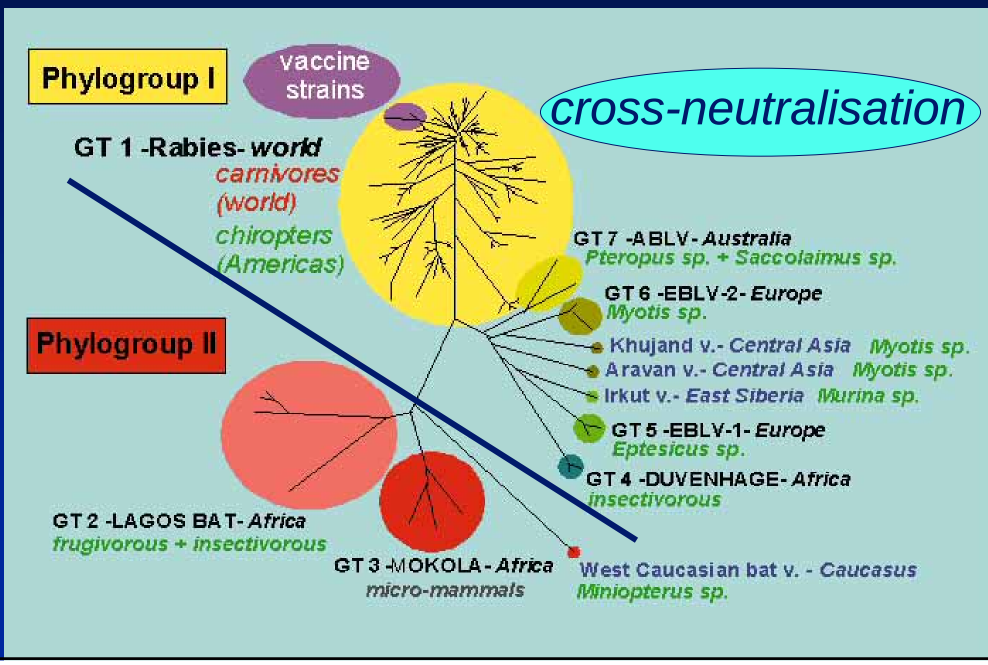
↓ cell culture: safe + efficient (expensive ?)

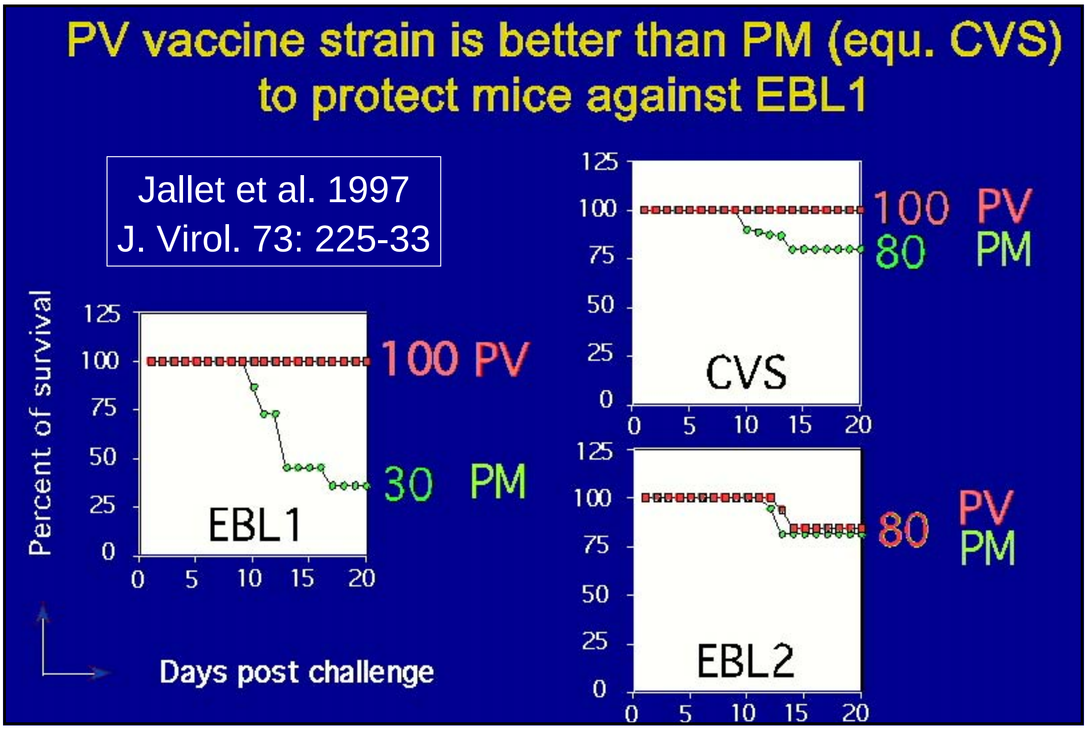
animal vaccines (prevention)

↓ nervous tissue (injection)
↓ cell culture (injection)
↓ attenuated/recombinant (oral, wildlife)

No efficient antiviral

Diversity of the *Lyssavirus* genus





Rabies vaccinology: what to improve ?

1. No Research needed

- Optimize availability of cell culture vaccines where needed
- Optimise vaccine delivery (save money)
  - Improve i.d. schedules, …

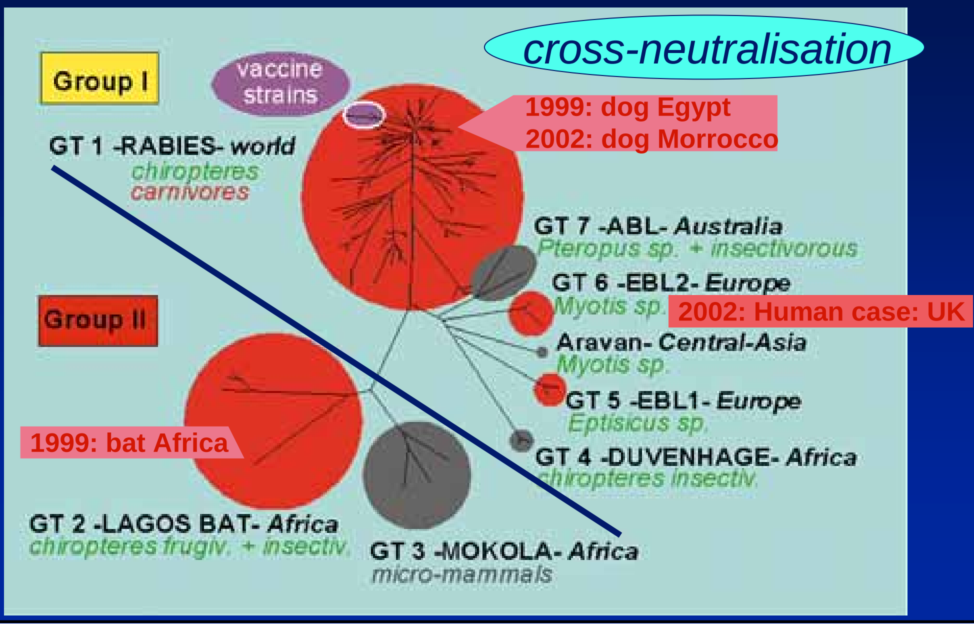
2. Research needed !

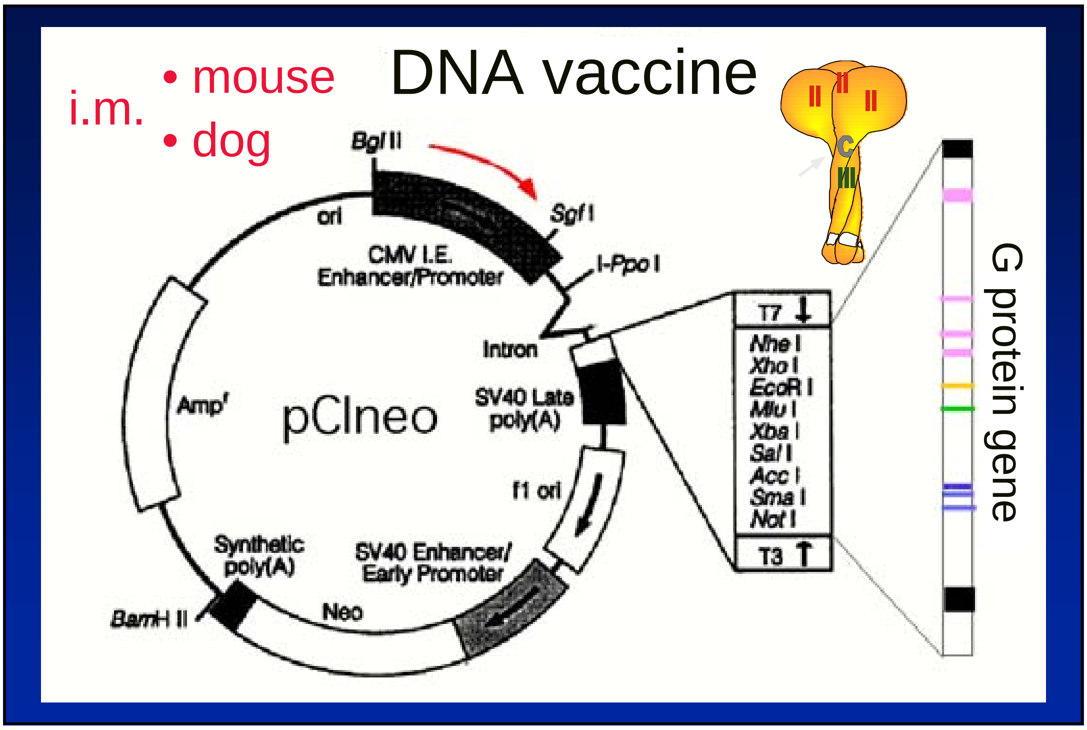
- Increase vaccine spectrum: rabies + rabies-related lyssavirus
- Oral route (wildlife vaccination): needs « replicative » antigen
  - attenuated lyssaviruses (SAD, SAG, reverse genetics)
  - recombinant viruses: poxviruses, adenoviruses expressing G protein
- Combined protection (more than one antigen)
- Marked vaccines (tagged by reverse genetics)
  - difference between infected and vaccinated animals
- Adjuvants (improving the immune response)

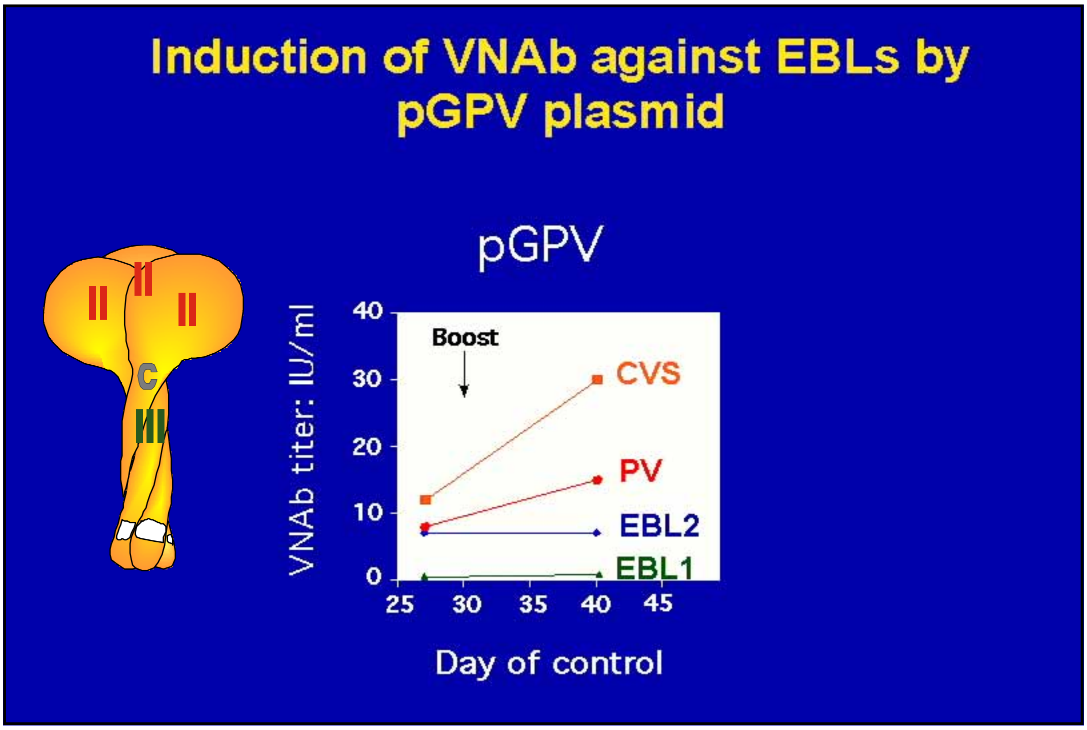
Trials to open the vaccine spectrum

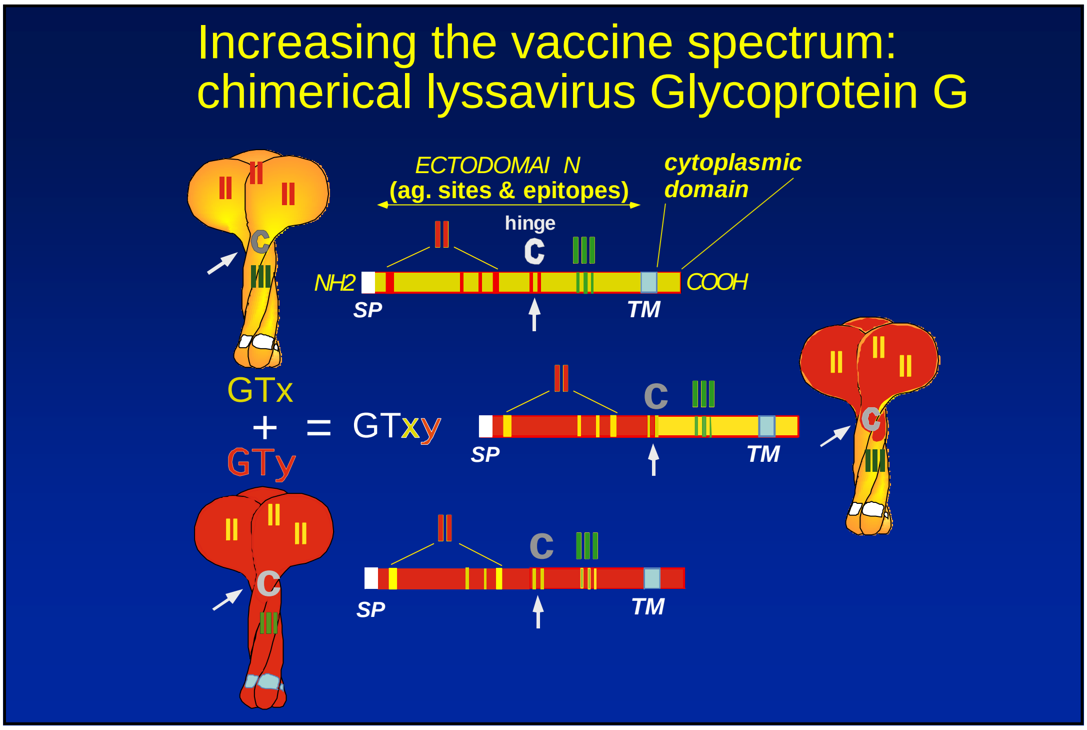
anti-rabies -> anti-lyssavirus

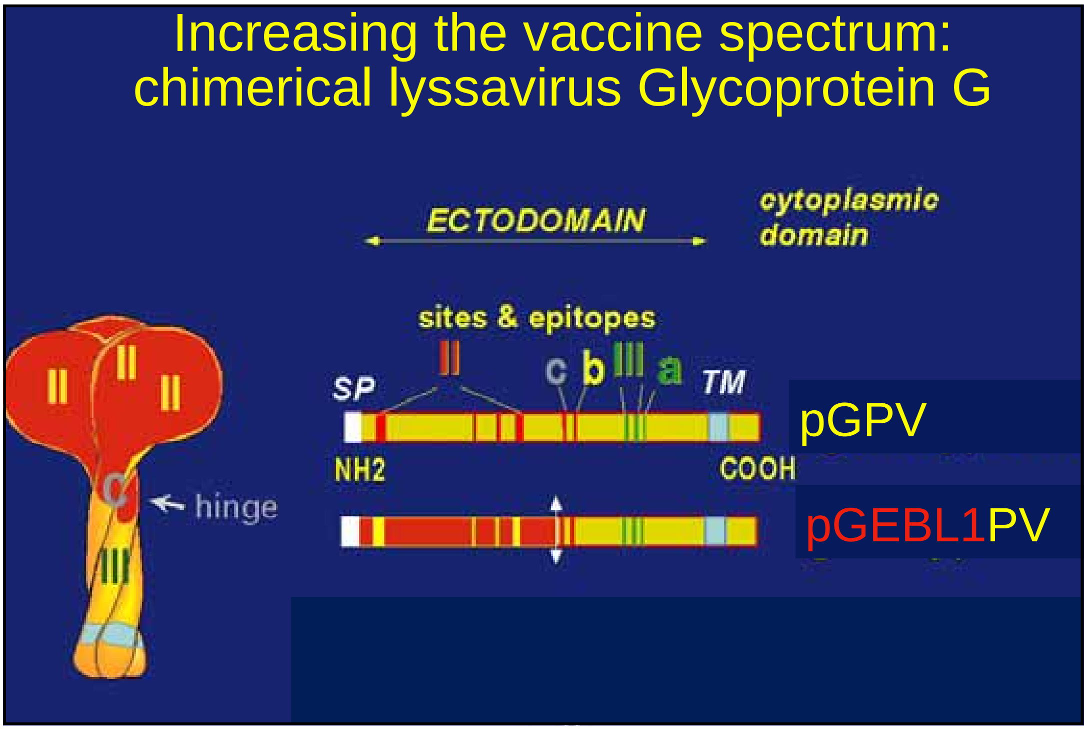
Rabies in Western Europe today



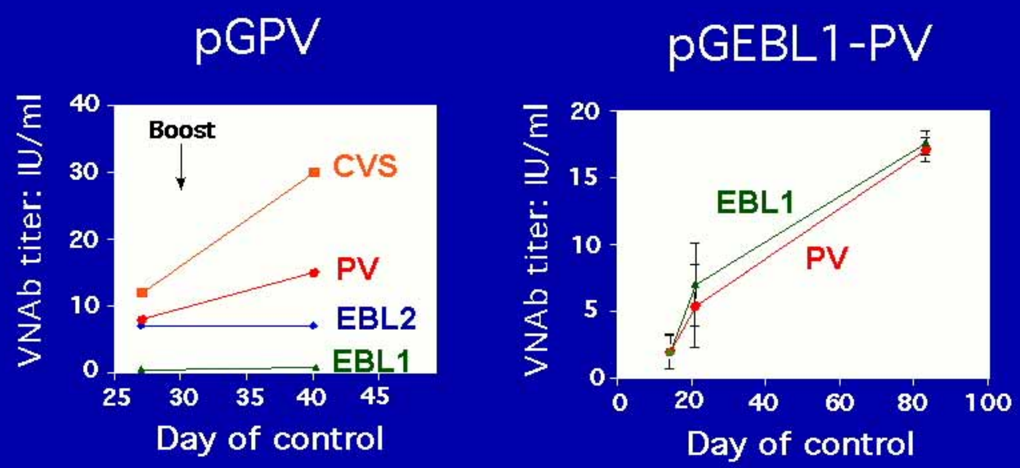

i.m.
• mouse
• dog
DNA vaccine
pCIneo
Bgl II
Sgf I
I-Ppo I
ori
CMV I.E. Enhancer/Promoter
Intron
SV40 Late poly(A)
f1 ori
SV40 Enhancer/ Early Promoter
Neo
Synthetic poly(A)
BamH II
Amp^r
T7
Nhe I
Xho I
EcoR I
Mlu I
Xba I
Sal I
Acc I
Sma I
Not I
T3
G protein gene
II
II
II
C
III


Induction of VNAb against EBLs by pGPV plasmid
pGPV
VNAb titer: IU/ml
Boost
CVS
PV
EBL2
EBL1
0
10
20
30
40
25
30
35
40
45
Day of control
II
II
II
C
III


Increasing the vaccine spectrum:
chimerical lyssavirus Glycoprotein G
ECTODOMAI N
(ag. sites & epitopes)
cytoplasmic
domain
hinge
NH2
COOH
SP
TM
GTx
+ = GTxy
GTy
SP
TM
SP
TM

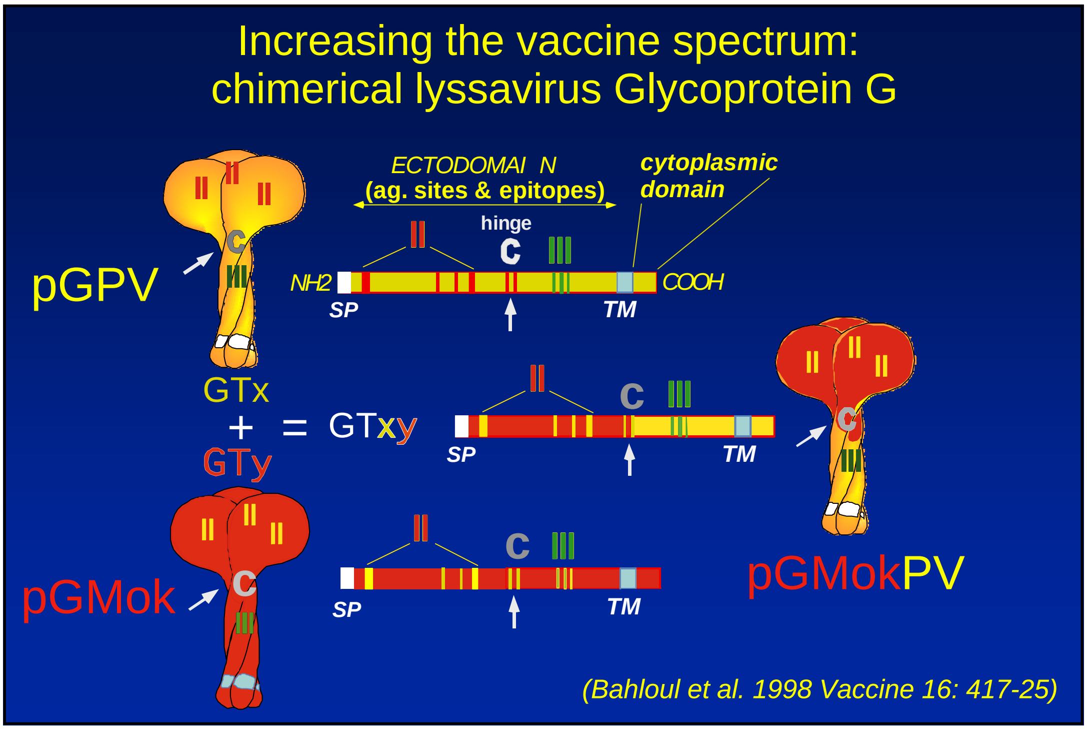

Increasing the vaccine spectrum:
chimerical lyssavirus Glycoprotein G
ECTODOMAIN
cytoplasmic
domain
sites & epitopes
SP
c b III a
TM
pGPV
NH2
COOH
hinge
pGEBL1PV

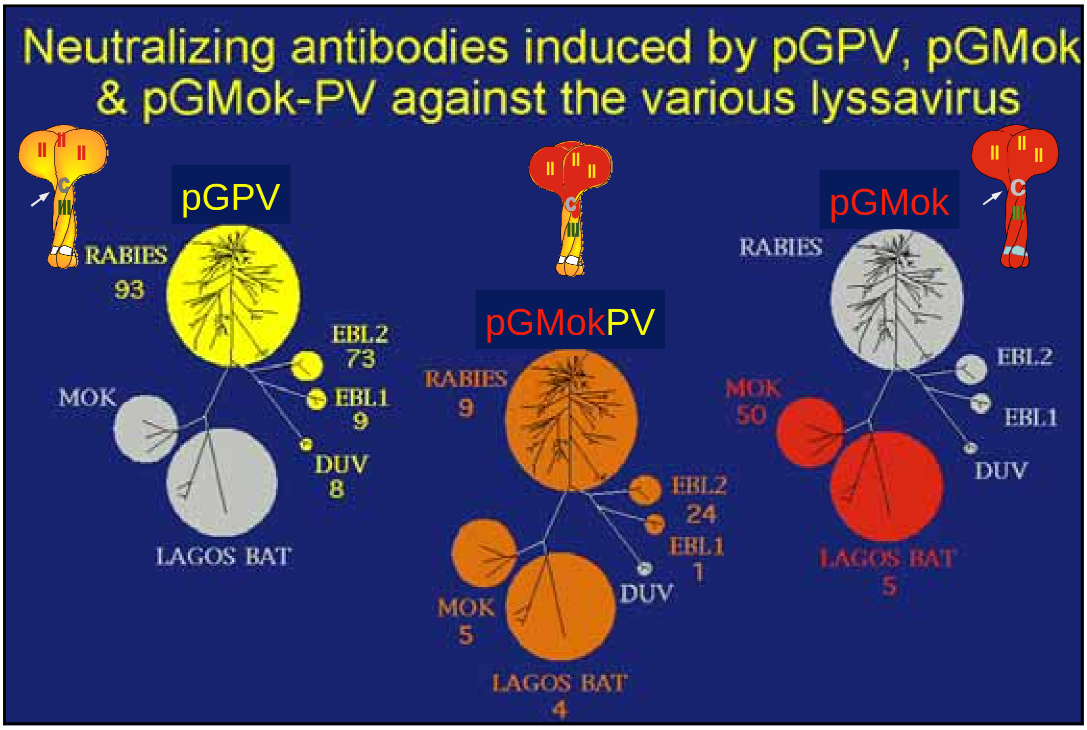
Induction of VNAb against EBLs by pGPV and pGEBL1-PV plasmids



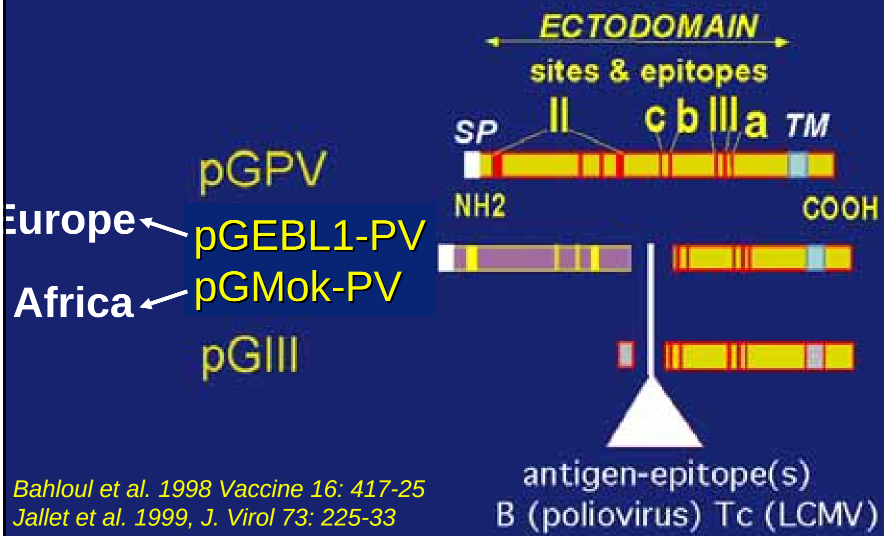
Protection induced by pGPG and pEBL1-PV plasmids against EBLs

plasmid	*% survival against :*		
	CVS	*EBL1*	*EBL2*
pGPV	**80**	**45**	**65**
pGEBL1-PV	**75**	**75**	**80**
pCIneo	**0**	**0**	**0**


Increasing the vaccine spectrum:
chimerical lyssavirus Glycoprotein G
pGPV
ECTODOMAI N
(ag. sites & epitopes)
cytoplasmic
domain
hinge
NH2
COOH
SP
TM
GTx
+ = GTxy
GTy
pGMok
pGMokPV
(Bahloul et al. 1998 Vaccine 16: 417-25)


Neutralizing antibodies induced by pGPV, pGMok
& pGMok-PV against the various lyssavirus
pGPV
RABIES
93
EBL2
73
EBL1
9
DUV
8
MOK
LAGOS BAT
pGMokPV
RABIES
9
EBL2
24
EBL1
1
DUV
MOK
5
LAGOS BAT
4
pGMok
RABIES
EBL2
EBL1
DUV
MOK
50
LAGOS BAT
5

Multivalent vaccinology: chimerical G protein carrying foreign epitopes/antigen



Bahloul et al. 1998 Vaccine 16: 417-25
Jallet et al. 1999, J. Virol 73: 225-33
Desmezières et al, 1999 JGV: 2343-51

CONCLUSIONS

Lyssaviruses originate in bats from which they regularly spill over into carnivores

- Rabies eradication will be difficult, rabies contol is possible.

In countries having controled carnivoran rabies by parenteral (dog) or oral (fox in W. Europe) vaccination

- Bat rabies is epidemiologically emerging.

Several Lyssavirus genotypes (EBLs, Mokola, Lagos bat, new) are genetically and functionaly divergent from vaccine strains

- Increasing the vaccine spectrum has been demonstrated rabies -> lyssavirus vaccine.

Lyssavirus chimerical G can carry foreign epitopes-antigens

- multivalent vaccination (1 animal, multiple pathogens).

Institut Pasteur, Paris

Unit Antiviral Strategies

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Alternative Recombinant Poxvirus Vaccines for Rabies: Experiences with a recombinant MVA rabies vaccine

Principle investigator:

Jacqueline Weyer

University of Pretoria

Molecular Virology

Co-authors:

Dr Louis H. Nel, University of Pretoria

Dr Charles E. Rupprecht, CDC, USA

Dr Gerrit J. Viljoen, ARC-OVI, SA *(now FAO/IAEA, Austria)*

University of Pretoria

V-RG

- Recombinant vaccinia virus rabies glycoprotein;
- Commercial vaccine for oral vaccination of wildlife in United States and some European Countries;
- Disadvantages:

  Safety considerations

  - wide host range, possible spread to non-targets;
  - potency in immunosuppressed subjects

Recombinant poxviruses as vaccines

Features:

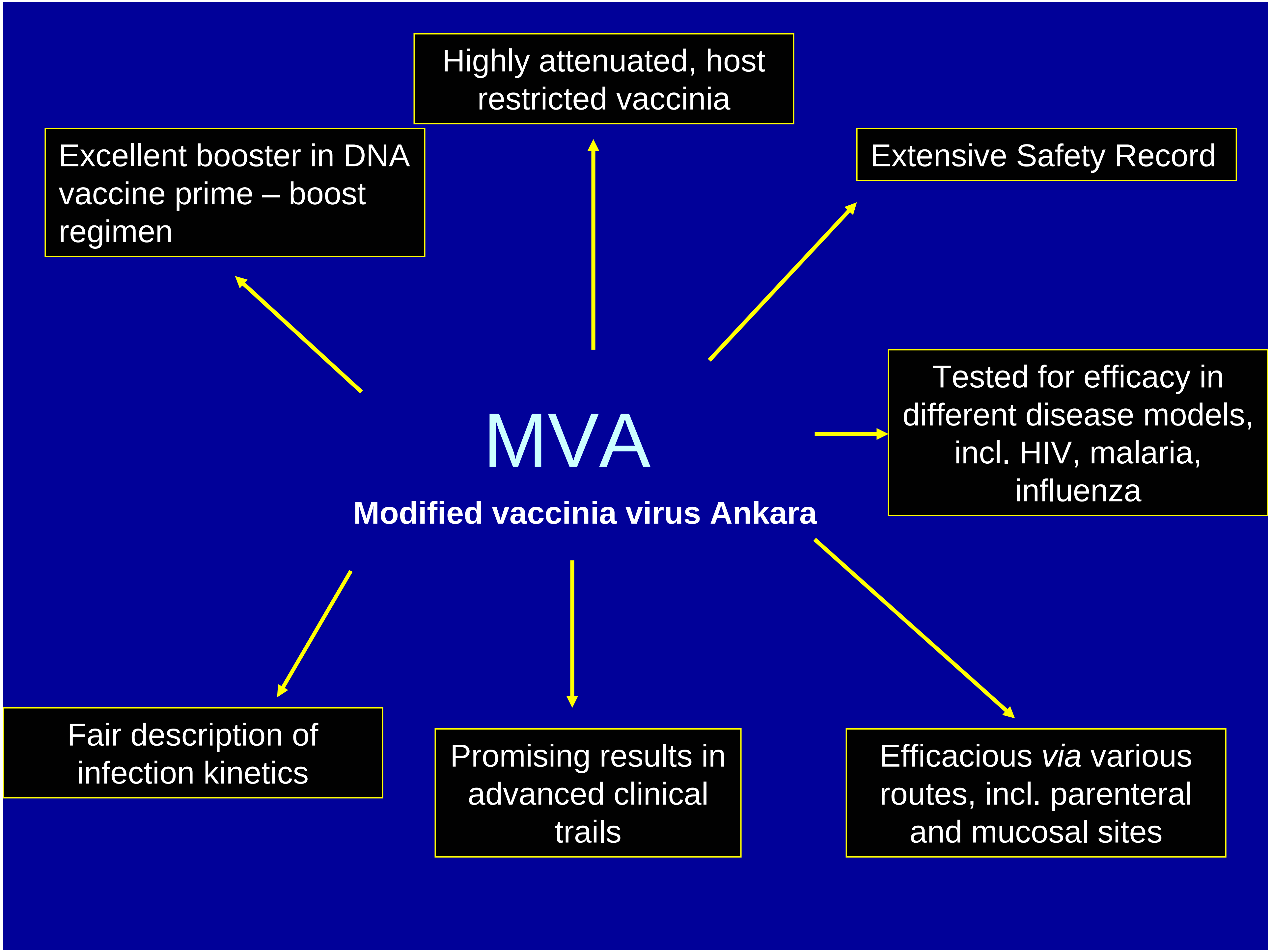
- Relative heat stability/stability of lyophilized material
- Relative ease/low cost of production/administration
- Tolerate large inserts (< 30 Kb)
- Trigger both sides of the immune response
- Choice of many different vectors such as vaccinia and vaccinia virus derivatives

A useful rabies vaccine:

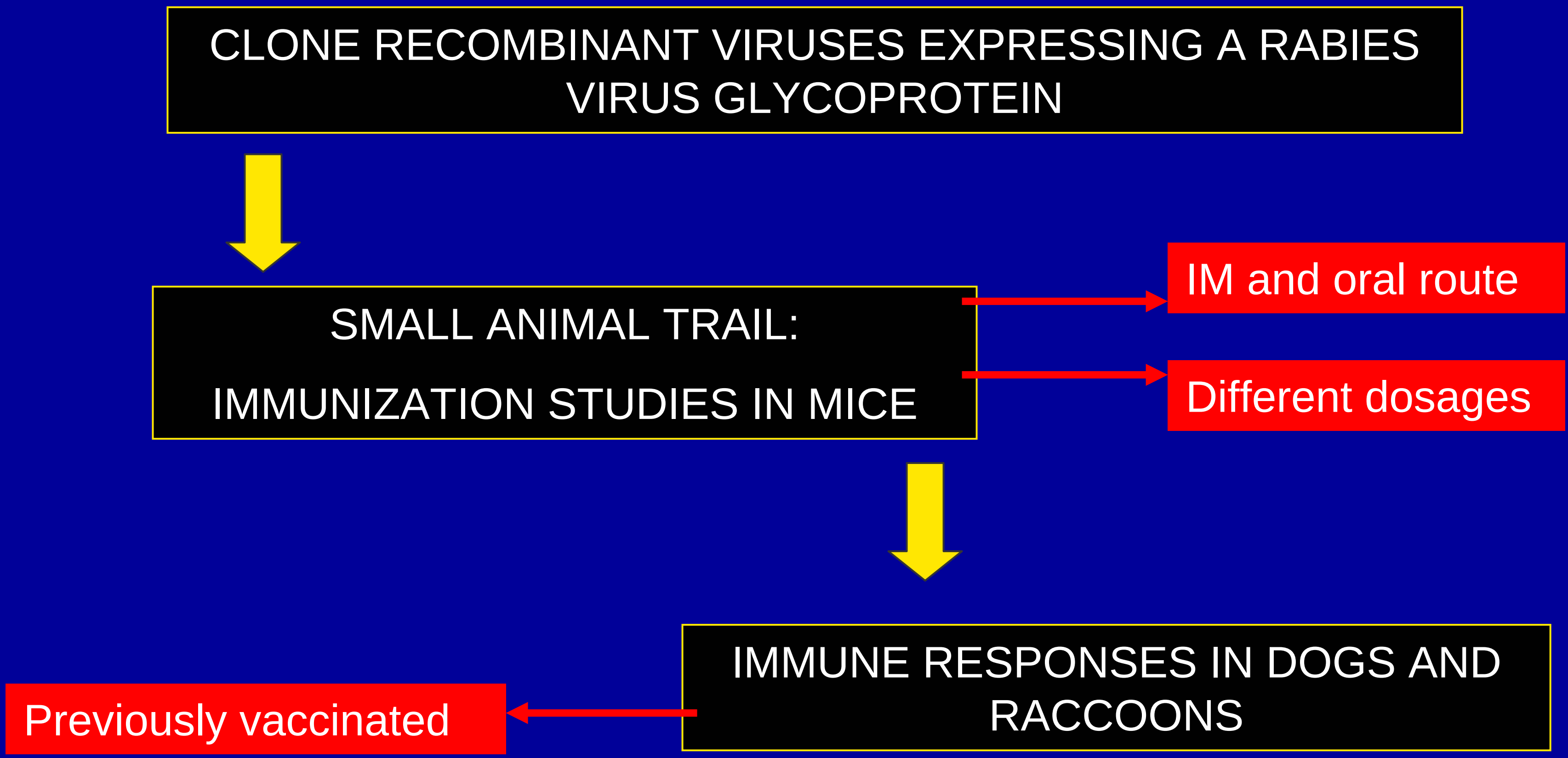
- *oral administration;*
- *suitability for distribution in baits;*
- *undoubted safety*

Hypothesis

Modified vaccinia virus Ankara **(MVA)** expressing a full-length rabies virus glycoprotein will provide an safer alternative vaccine, with improved safety, compared to a experimental recombinant vaccinia glycoprotein

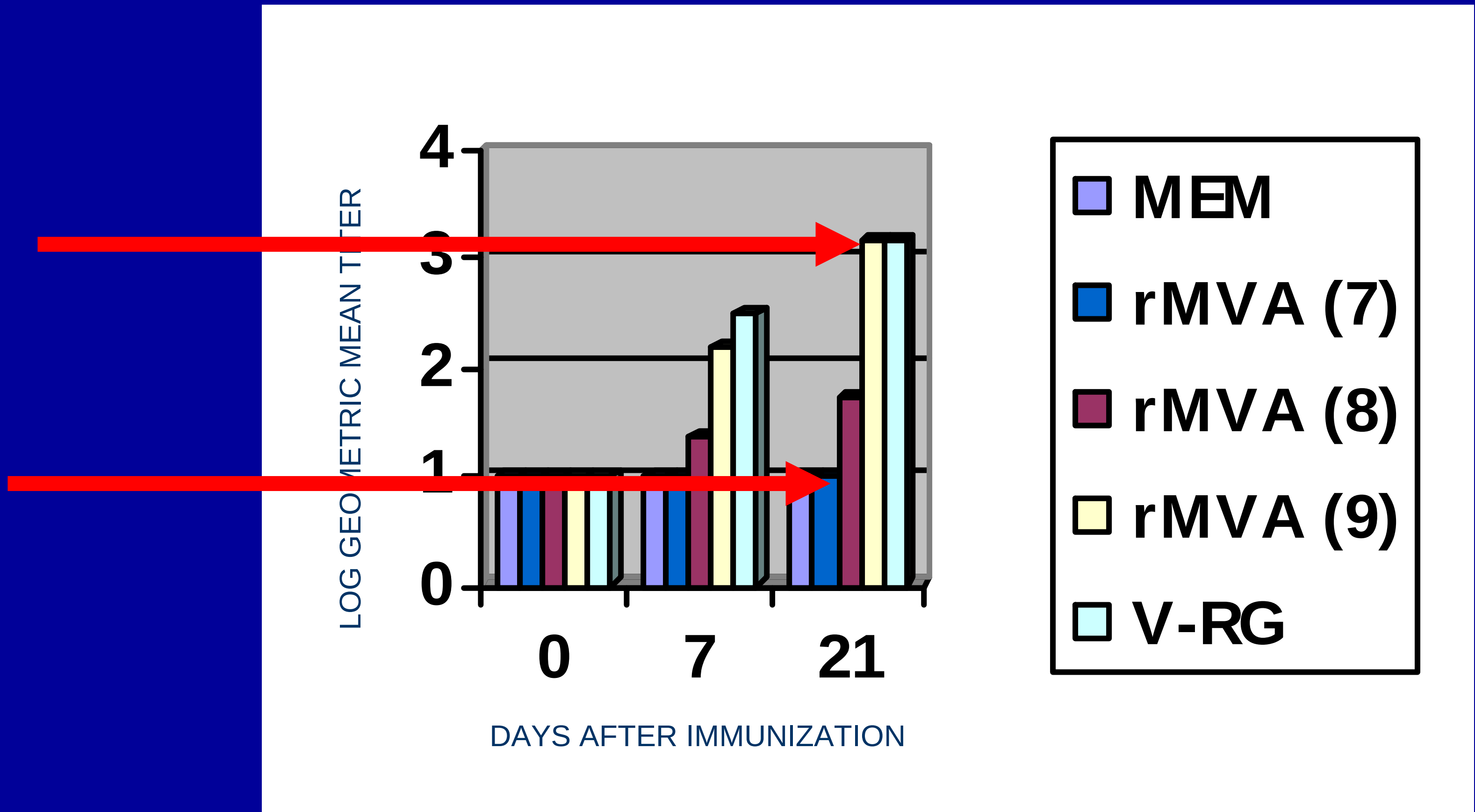

Highly attenuated, host restricted vaccinia
Excellent booster in DNA vaccine prime – boost regimen
Extensive Safety Record
MVA
Modified vaccinia virus Ankara
Tested for efficacy in different disease models, incl. HIV, malaria, influenza
Fair description of infection kinetics
Promising results in advanced clinical trails
Efficacious *via* various routes, incl. parenteral and mucosal sites

Experimental Outline



Testing of rMVA-RG in murine model

Log 10 geometric mean virus neutralizing antibody titer in mice that received recombinant vaccinia viruses intramuscularly



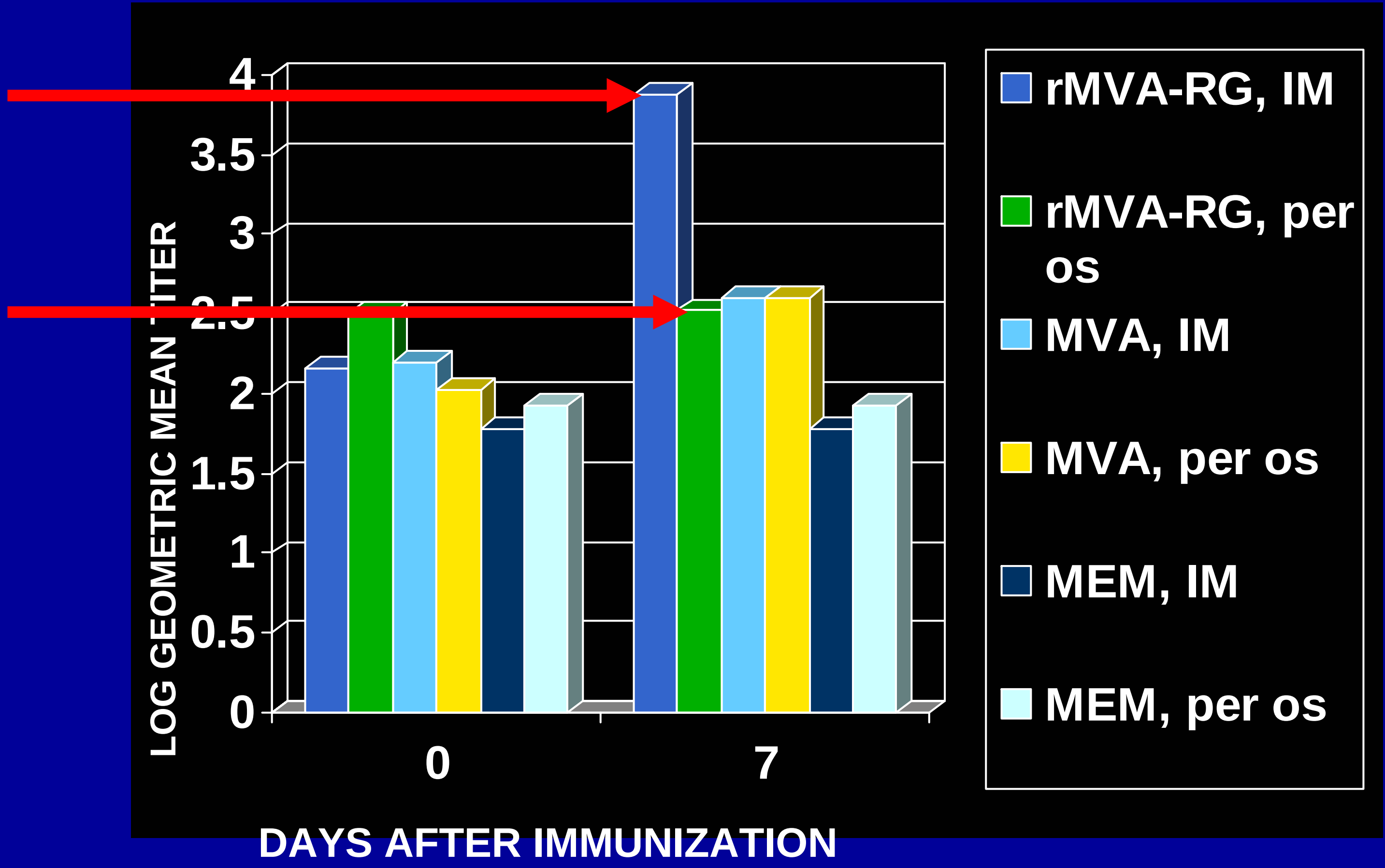
Testing of rMVA-RG in murine model

Log 10 geometric mean virus neutralizing antibody titer in mice that received recombinant vaccinia viruses orally

GROUP		DAY 0[A]	DAY 7[B]	DAY 21[C]	% SURVIVORSHIP (NUMBER ANIMALS)
rMVA-RG	boost	< 1.0	< 1.0	< 1.0	0 % (0/10)
V-RG	boost	< 1.0	1.14 (<1.0 – 2.18)	1.86 (<1.0 – 2.23)	40 % (4/10)
wt MVA	boost	< 1.0	< 1.0	< 1.0	0 % (0/10)
Vacc Cop	boost	< 1.0	< 1.0	< 1.0	0 % (0/10)
MEM	boost	< 1.0	< 1.0	< 1.0	0 % (0/10)

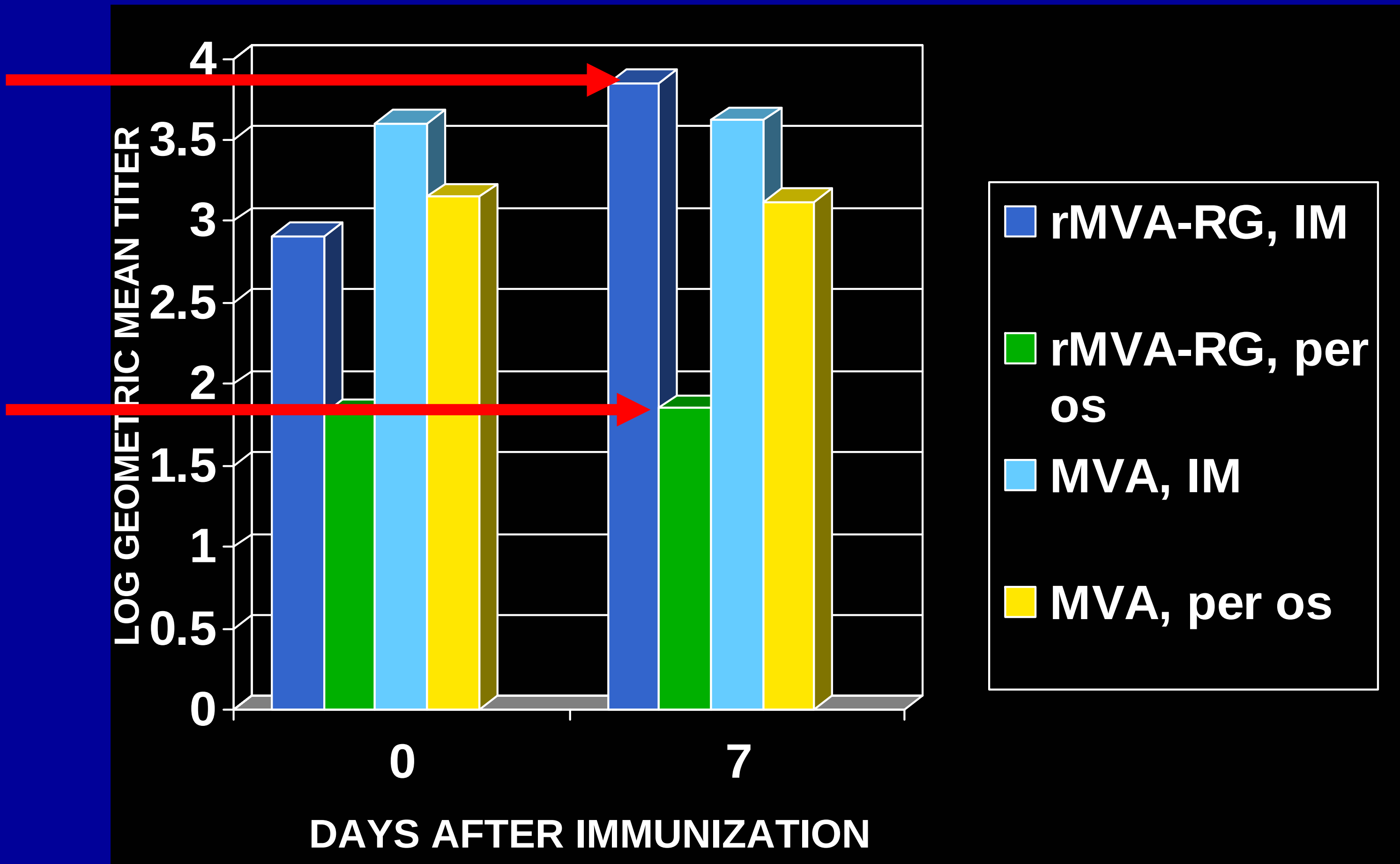
Anamnestic responses in previously vaccinated dogs

Log 10 geometric mean virus neutralizing antibody titer in dogs that received recombinant vaccinia viruses orally



Anamnestic responses in previously vaccinated raccoons

Log 10 geometric mean virus neutralizing antibody titer in raccoons that received recombinant vaccinia viruses orally



Results

- rMVA elicited humoral responses upon IM administration – dose dependant
- rMVA elicited anamnestic responses in previously vaccinated dogs/raccoons;
- rMVA failed to elicit humoral immune responses upon oral administration

Conclusion

- Severe attenuation influence efficacy of recombinant MVA:

  - higher dosages required;

  - failure of immune responses *per os*

Acknowledgements

- University of Pretoria, SA:

  Wanda Markotter
- CDC, USA:

  Josh Self

  Michael Niezgoda
- ARC-OVI, SA:

  Dr Claude Sabeta

  Janet Mans *(now NIH, US)*

DNA VACCINES FOR RABIES: AN OVERVIEW

Nobantu Phalatsi
Prof. LH Nel
Jacqueline Weyer
University of Pretoria

INTRODUCTION

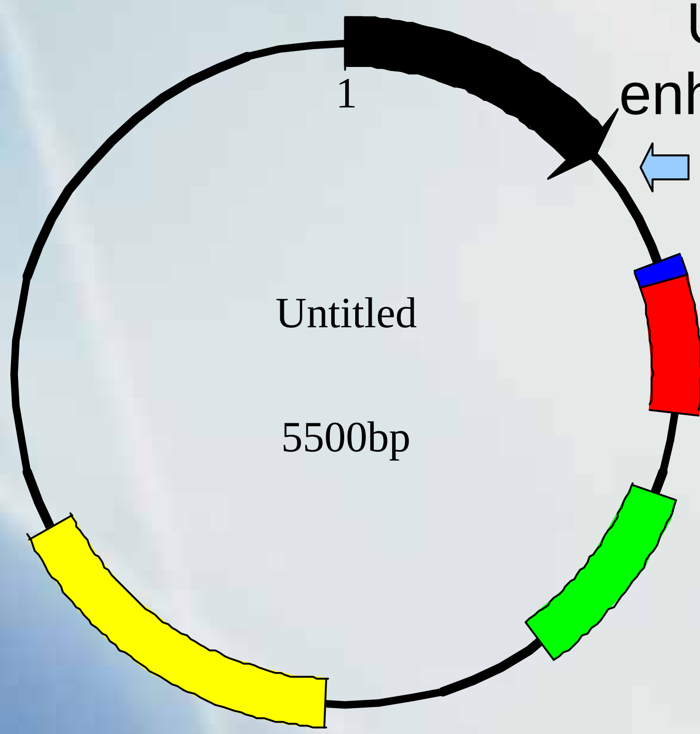
WHAT ARE DNA VACCINES?

DNA vaccines are mammalian expression vectors that carry foreign gene(s)

May be in the form of DNA or mRNA

Able to direct *in vivo* expression, leading to induction of antigen-specific immune responses (genetic immunization)

▪ MAIN FEATURES OF DNA VACCINES



Promoter

Usually I/E promoter/ enhancer from pathogenic virus

Intron - Upstream of coding region

Augmented expression depends on position of introns within transcription unit

MCS - site in which target gene is inserted

Polyadenylation signal - for enhanced mRNA stability & tln

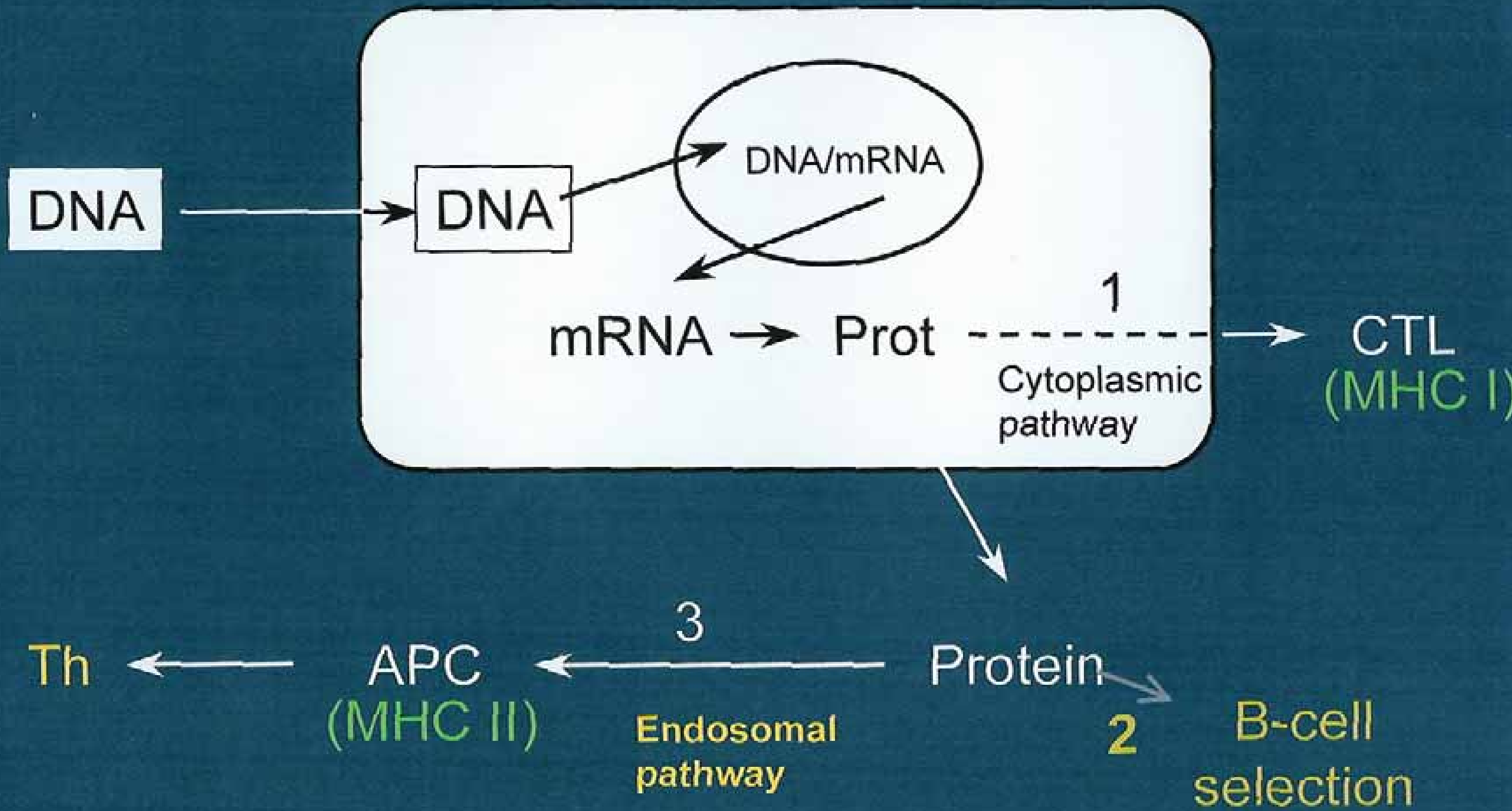
ori - replication origin usually ColE1 (best characterized)

Marker gene

i) Drug resistance gene

ii) auxotrophic gene

Immunology of DNA vaccines



WHY USE DNA VACCINES?

- DNA vaccines consist of only plasmid DNA, unlike recombinant or attenuated virus vaccines ∴ safer
- Able to induce similar immune responses as live or attenuated virus, engaging both MHC-I and -II pathways.
- Relatively inexpensive to manufacture

- Easy to generate, modify and purify
- Expressed proteins folded and processed in similar manner as live virus
- Can easily adapt existing vaccine models for a pathogen for other similar pathogens (versatile) - emerging epidemics
- High temperature stability

Shortcomings of DNA vaccine technology

- Immune response elicited by DNA vaccines develop relatively slowly with current methods of administration
- Wariness about introducing foreign genetic material into human hosts

Optimization strategies

- **Improvement of antigen expression**

- Optimization of elements of backbone
- Modification of the expressed antigen
- Optimization of transfection
- Modification of the vaccination regimen

SAFETY CONSIDERATIONS

1. Integration into the host genome
2. Induction of tolerance
3. Development of autoimmunity
4. Induction of inflammatory responses to Ag-producing cells

RECENT DEVELOPMENTS

- DNA vaccines for rabies have been used:

- **in different animal models:**
  - mice
  - dogs
  - cats
  - non-human primates

- **targeting different antigens:**
  - glycoprotein
  - chimeric glycoprotein
  - nucleoprotein

- **to different sites:**
  - mainly i.m.
  - other routes: i.d., i.p., subcutaneous, i.n.

- **against different lyssaviruses:**
  - rabies virus
  -mokola virus

- DNA vaccines have been administrated in
- **different formulations:**
  - naked
  - carrier-mediated

Have been investigated in prime-boost strategy (excellent primer)

Other application: Use in production of MAb (Barry *et al.*, 1994)

- **2003:** Single inoculation of G-expressing DNA vaccine into ear pinnae of dogs provides long-lasting protection

- **2005:** Immunogenicity of transmembrane domain of G protein.

- Utility of DNA vaccines as research tools to study vaccine models has been widely demonstrated

MSc PROJECT

- To evaluate the effects of the G-, N-, and M-protein of RV on efficacy of a DNA vaccine by delivering singly, and in combination.
- G and N proteins have been investigated as immunogens for rabies
- M protein never been investigated in the context of a vaccine
- Genetic immunization offers perfect system

- M protein is a major structural protein shown to be vital for virus assembly and budding
- For VSV, M is able to induce budding alone, pinching off membranes autonomously
- Interaction between G and M facilitates more efficient budding of nascent virions

- Budding of G protein-expressing particles may further stimulate Th-cell or antibody responses.

- Expression of G,M and N proteins (structural proteins) may facilitate production of virus-like particles, which can augment immune responses

- Vaccines expressing G,M and N proteins of rabies virus will be co-expressed in different combinations:
  - G + N
  - G + M
  - G + M + N

- Efficacy based on induction of protective immunity.

ACKNOWLEDGMENTS

- Prof LH Nel
- Jackie Weyer
- Dr. Sabeta
- Dr. Shumba
- Wanda Markotter
- Students in Virology lab (Univ. of PTA)

Oral Rabies Vaccine

Developing a safer and efficacious oral vaccine for dogs

Nico Visser, Teshome Mebatsion and Sreenivas Kilari

Rabies in developing countries

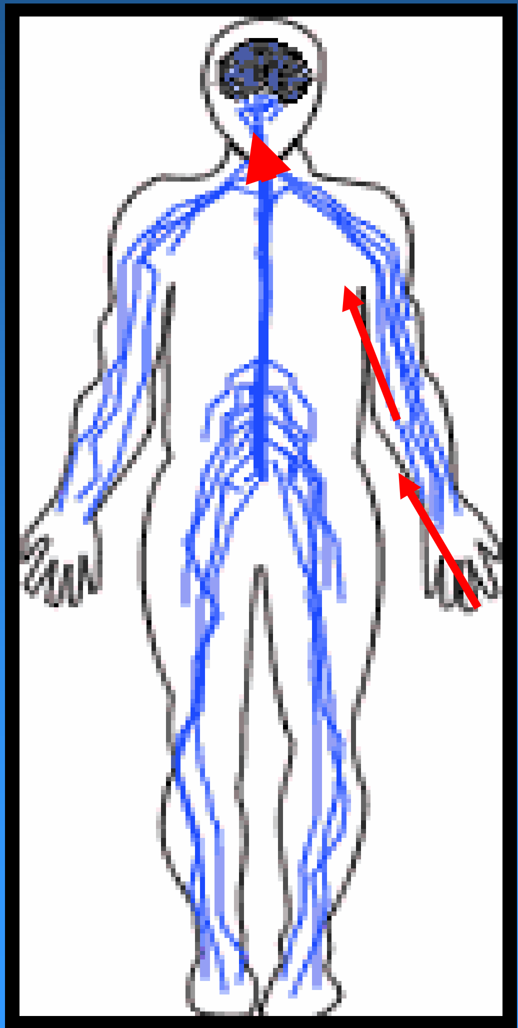
- Annually in India alone, about 23 000 people die of rabies. Recently WHO estimated in Africa ~24 000 cases
- About 95 % of rabies is due to bites from dogs which are mostly stray and ownerless
- Over 70 % of the victims are children younger than 15 years of age

Solution = Oral immunization of stray dogs

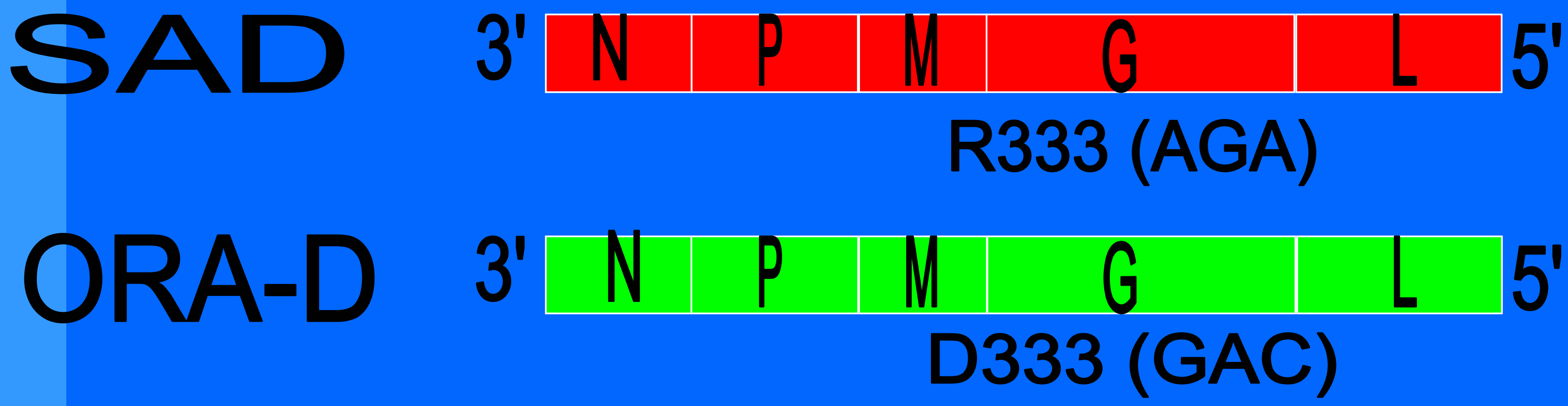
in addition to routine parenterally vaccination

Rabies

Neurotropic fatal infection

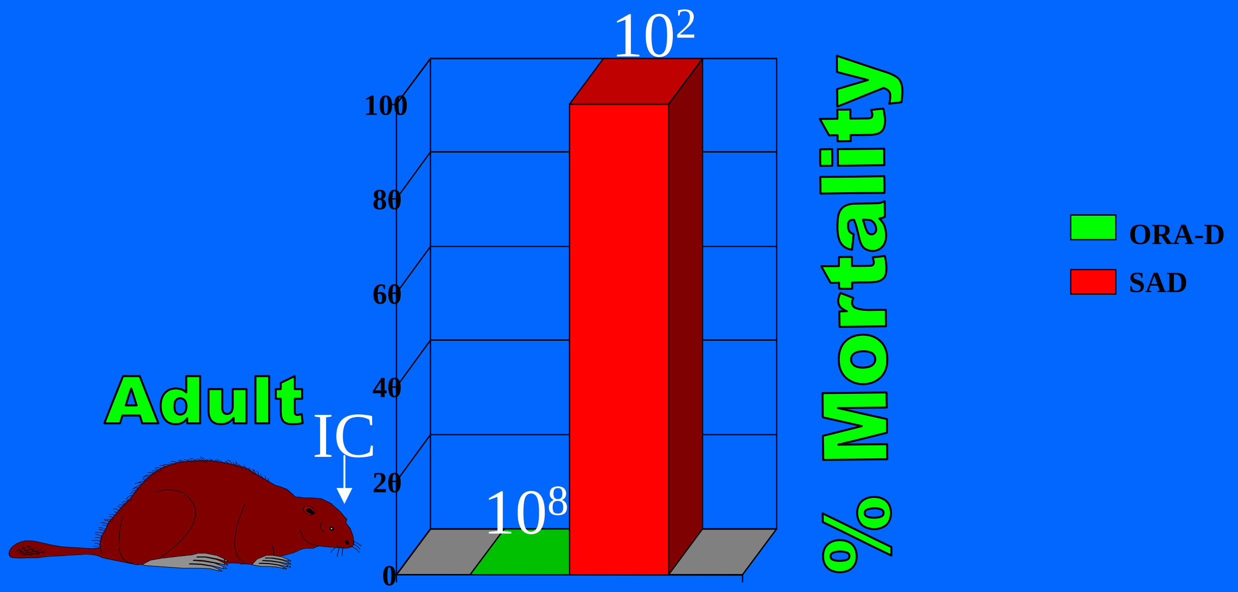


Genetically stable engineered rabies virus



The triple mutant ORA-D is genetically stable in suckling mice passages and in more than 25 cell culture passages in the absence of any Mab

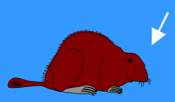
ORA-D with Arg333 substitution is safe in adult mice

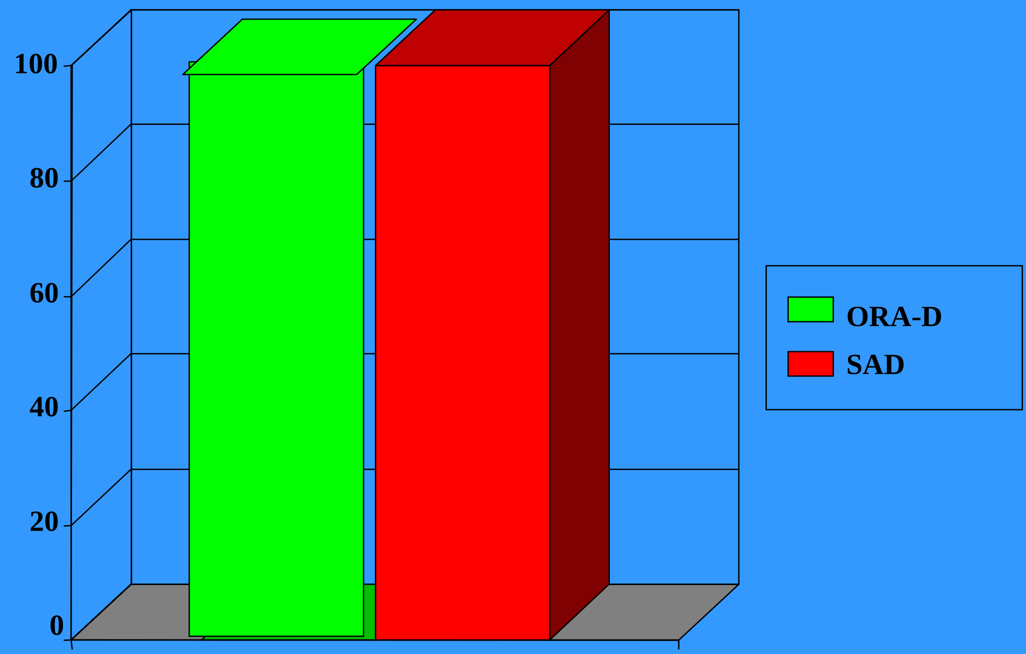


In baby mice the ARG substitution is not sufficient

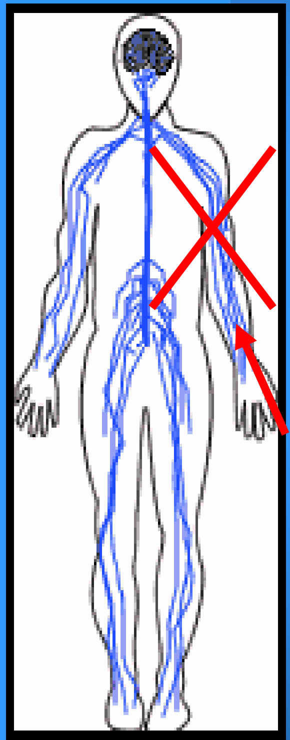
2-day-old

baby mice





Dynein Light Chain (LC8)



- LC8 protein is involved in intracellular movement
- Rabies P binds to LC8 (at AA 138-172)
- LC8 is probably responsible for in the axonal transport of rabies virus along neuron cells
- **Thus the goal of the next step of attenuation is to block this transport**

(Raux, et al., J. Virol. Nov. 2000), (Jacob, et al., J. Virol. Nov. 2000)

Deletion in P gene of rabies virus

A

SAD-L16 3' N P M G L 5'

R333

SAD-L16 GKSSEDKSTQTTG

L- Δ P7 GKSSE-------G

B

ORA-D 3' N P M G L 5'

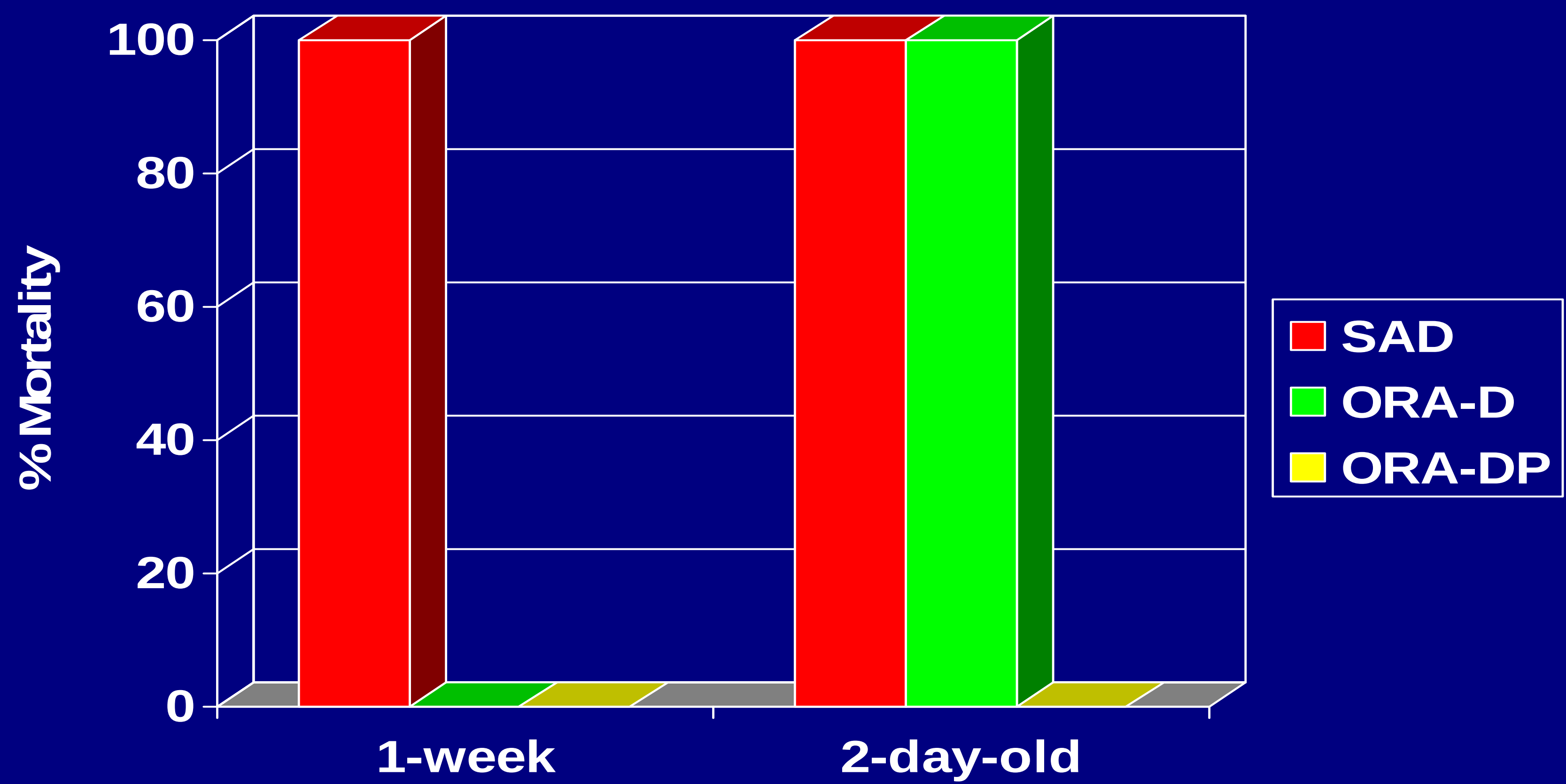
D333

D333 GKSSEDKSTQTTG

ORA-DP D - Δ P7 GKSSE-------G

Mebatsion, 2001, J. Virol., 75, 11496

Pathogenicity of Adapted Rabies Viruses in Suckling Mice

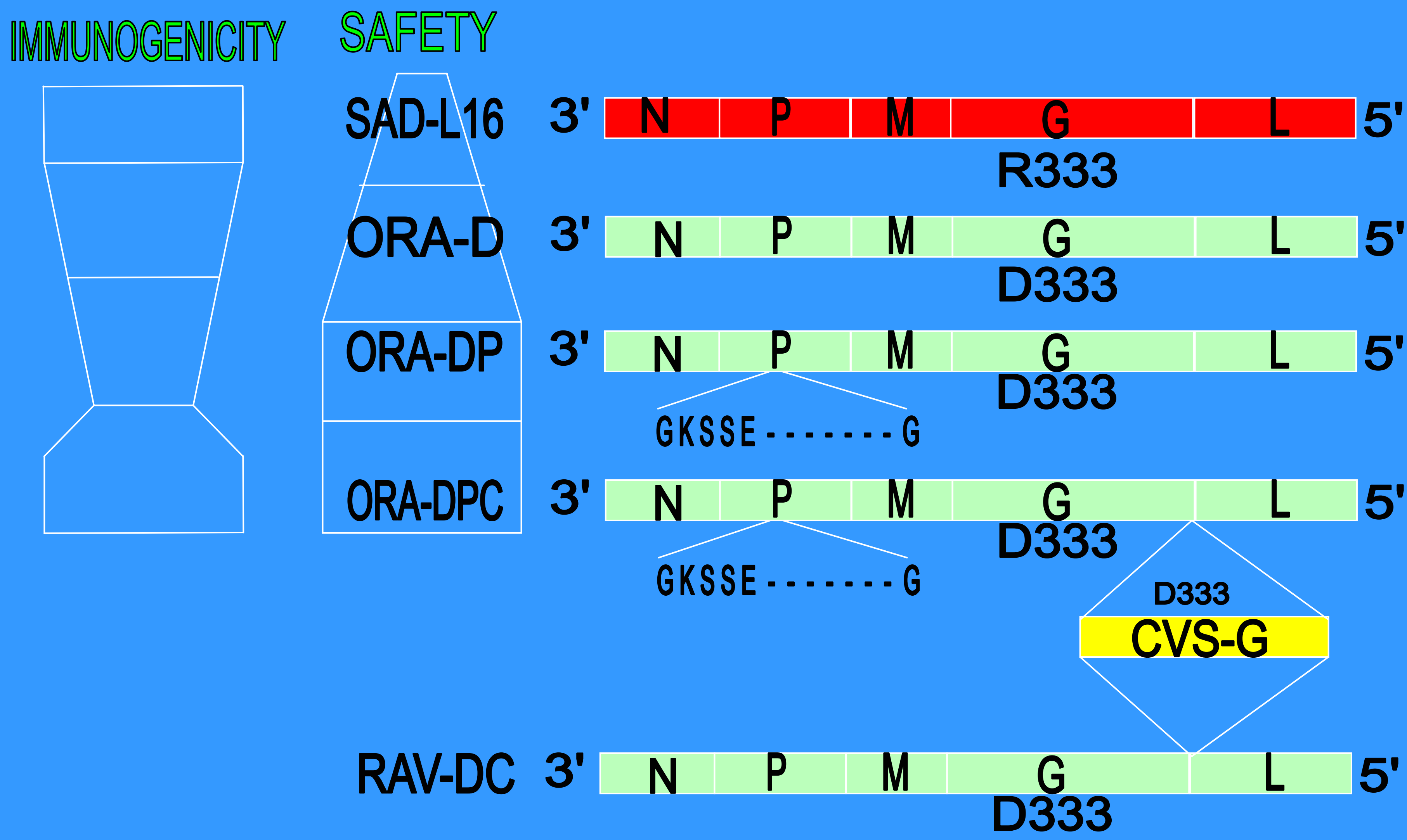


Deletion in P is synergistic to Arg substitution

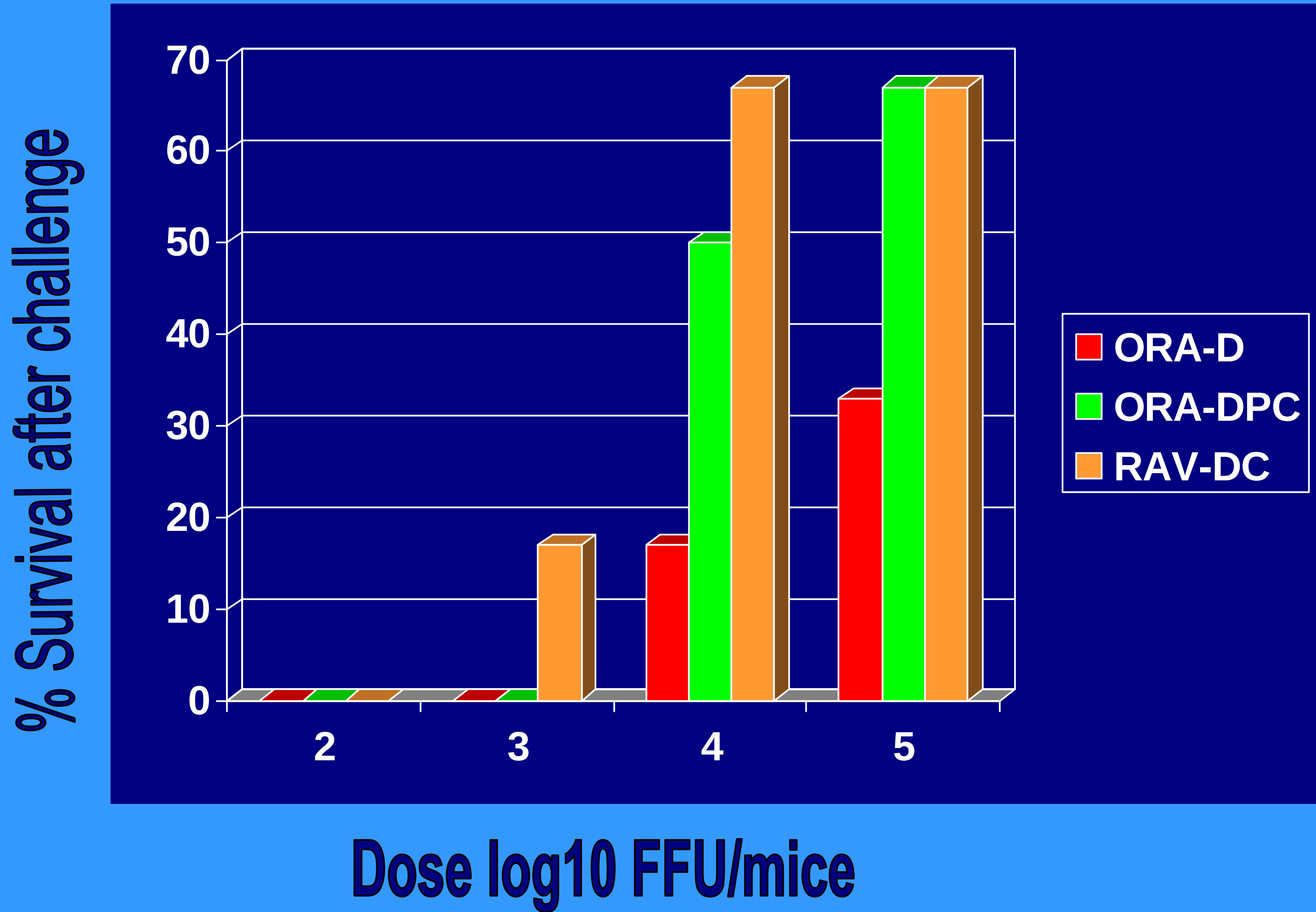
- Elimination of P-binding is only effective in the context of ORA-D. Thus after substitution of Arg333, this is adding to attenuation and thus safety
- But not as single deletion in the SAD B19

Mebatsion, 2001, J. Virol., 75, 11496

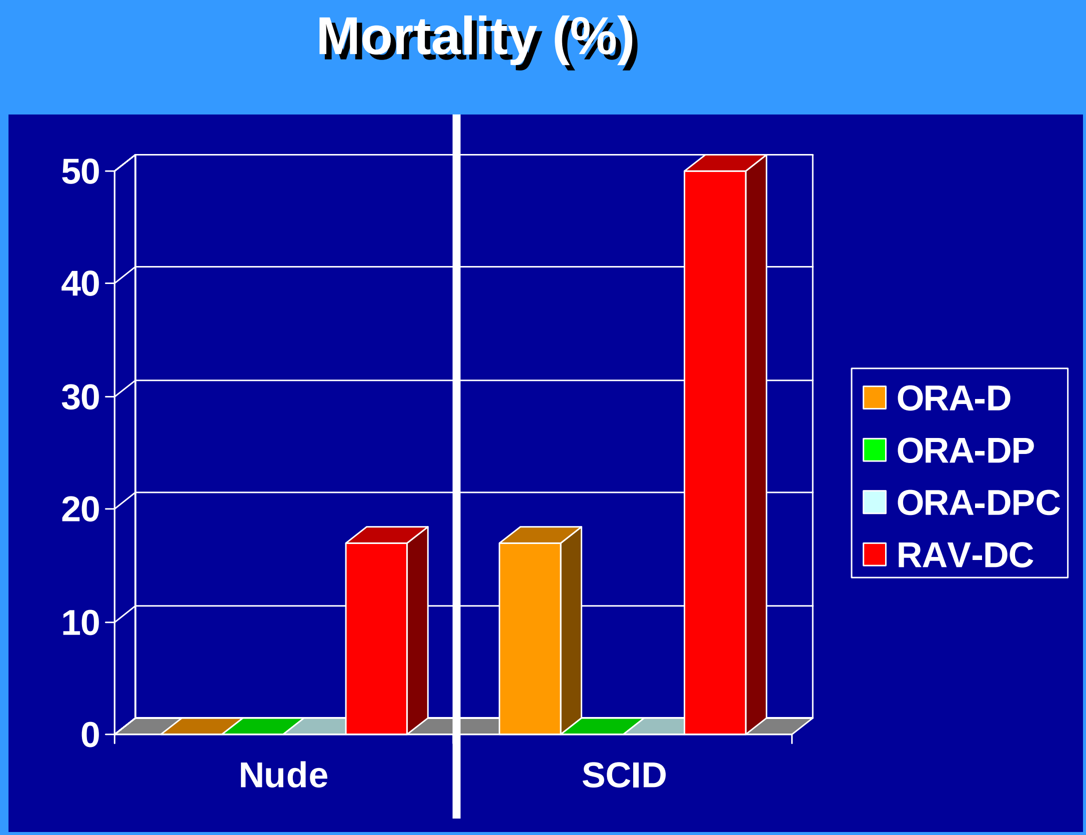
Increasing IMMUNOGENICITY without decreasing SAFETY



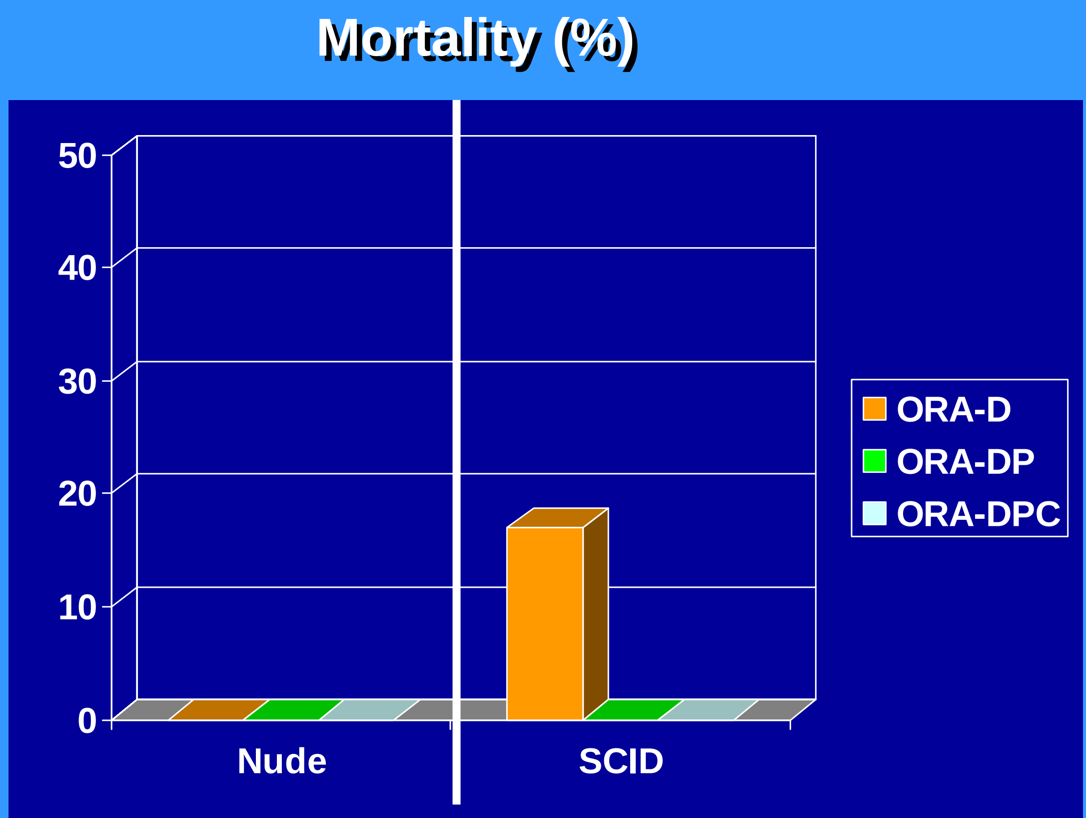
Efficacy of Rabies Viruses in Mice



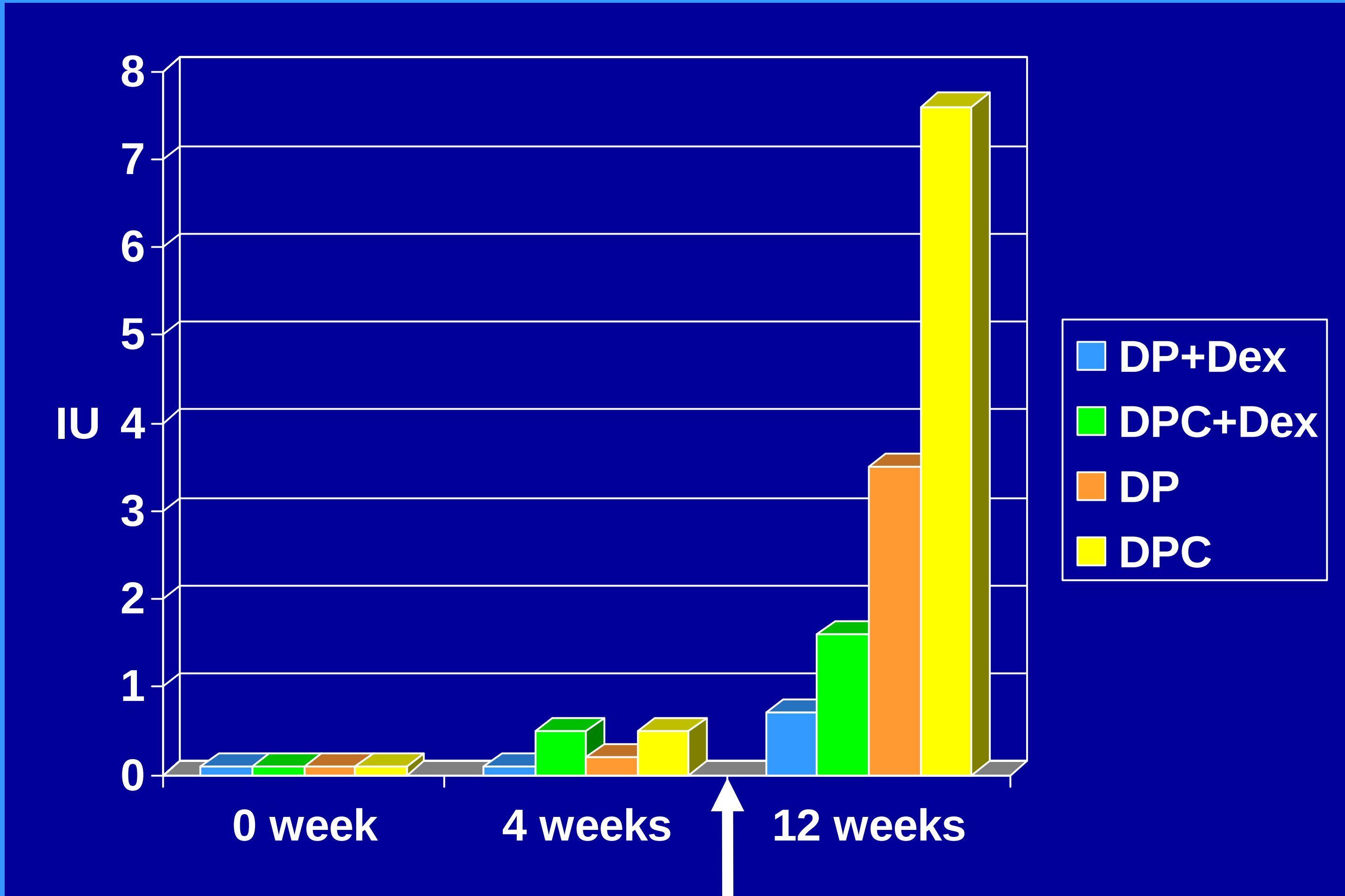
Pathogenicity of the adapted viruses in Nude and SCID mice



Pathogenicity of the adapted viruses in Nude and SCID mice



Safety and Efficacy of ORA-DP/-DPC in dogs (- or + Dexadresson, 0.5 mg/kg bw)



Booster dose 8 weeks after the 1st immunization

Vaccine application

- Preferable by “throwing” the baits towards the dogs
- Aiming that each dog has (at least) one bait
- Take a time and place where the dogs usually group together
- Re-collect the non-punctured containers from the site after the distribution

Vaccination schedule

- **Preferable Two vaccinations 4-8 weeks apart**
  - Advantages are having
  1) a higher chance of reaching an animal at least one time
  2) a high titer in those animals that accepted the bait both times

- **Revaccinations each time there are new litters**
  - Advantage is that the number of sensitive animals is as small as possible for the shortest period of time

Conclusions

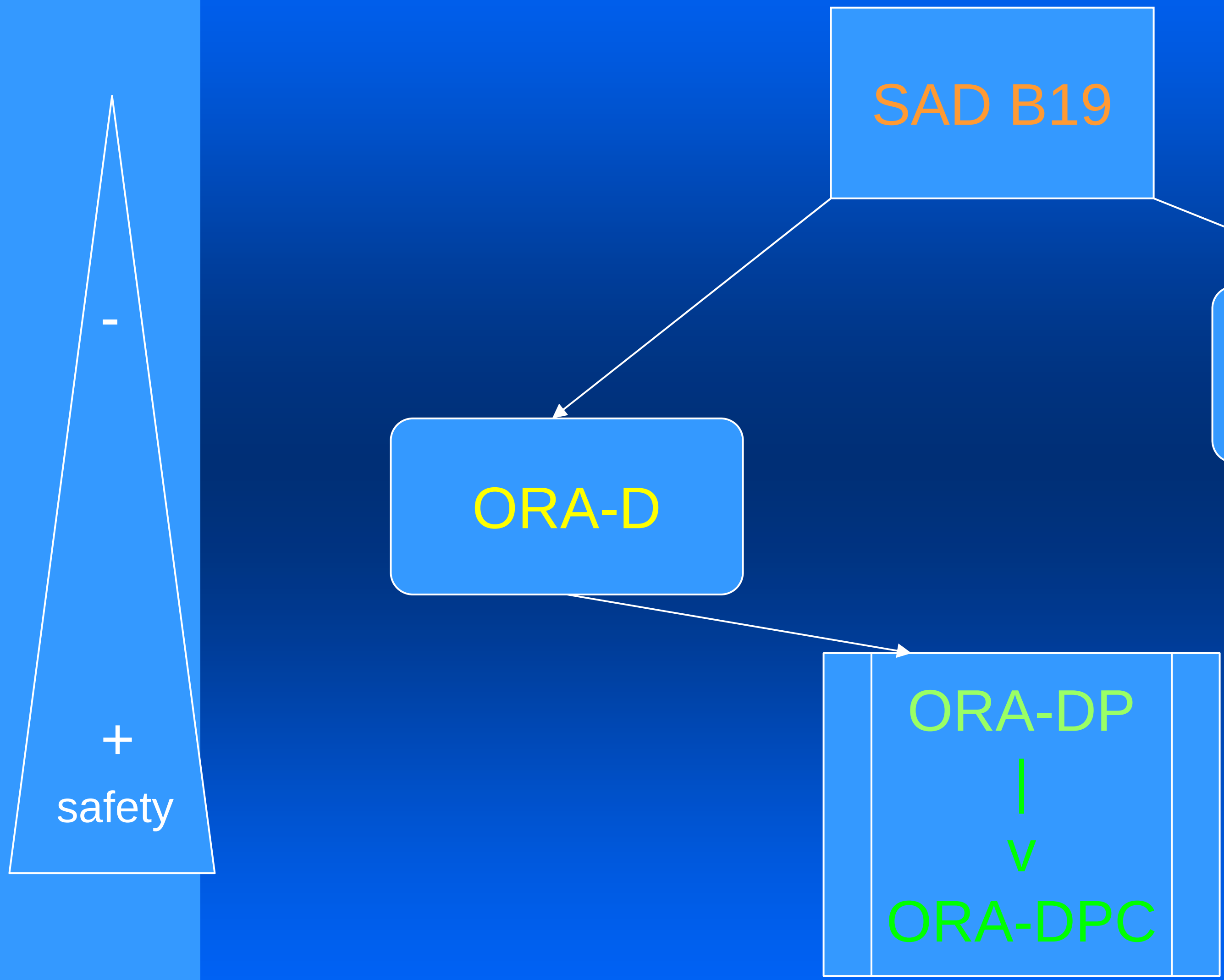
- **Positive effect on efficacy:**
  - Additional G-protein
  - Guided delivery of bait

- **Positive effect on safety:**
  - Start with attenuated vaccine strain
  - Stable replacement of Arg333
  - Deletion in P-protein
  - Additional G-protein
  - Removal of left baits

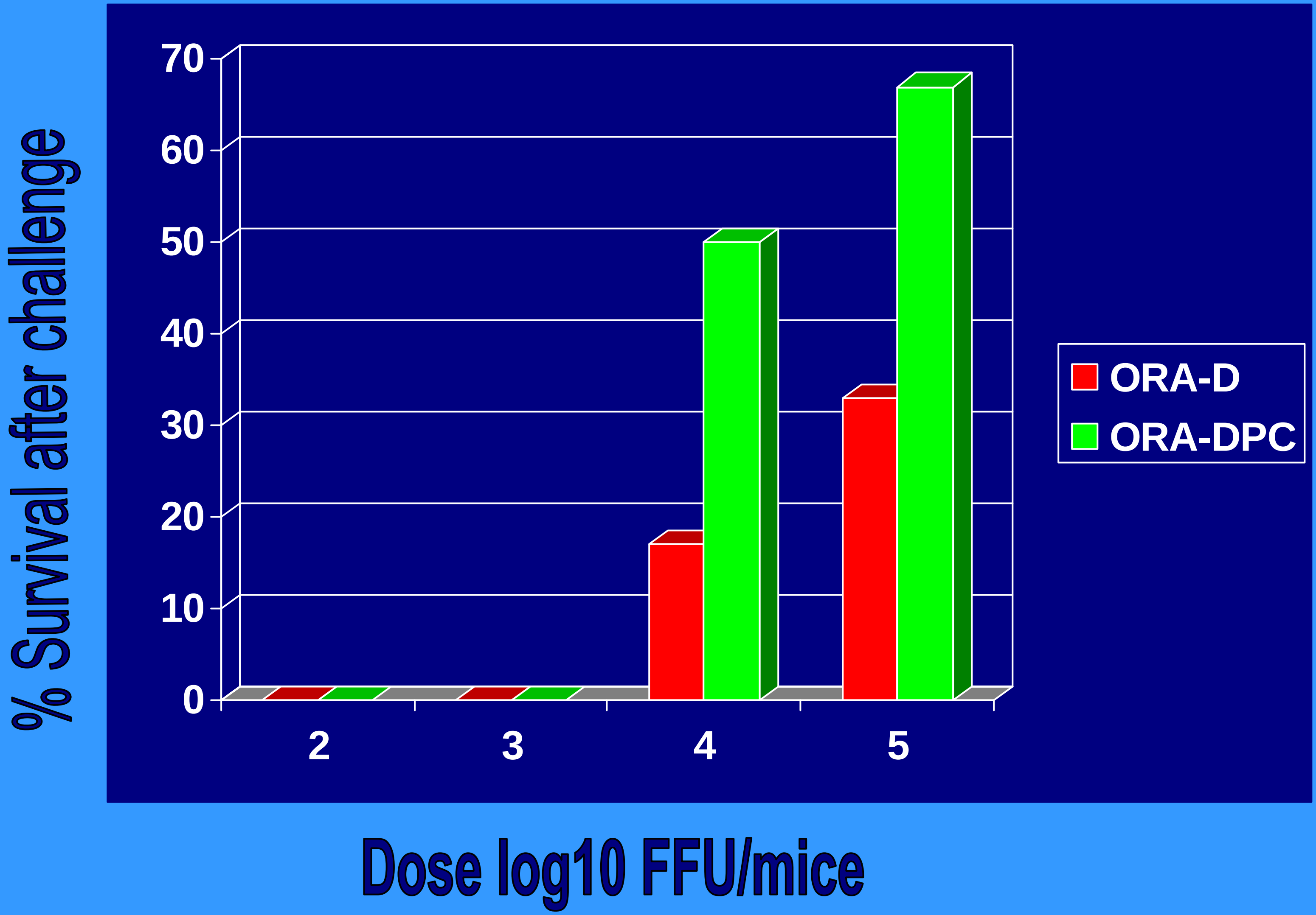
Thank you !!

www.rabies-vaccination.com

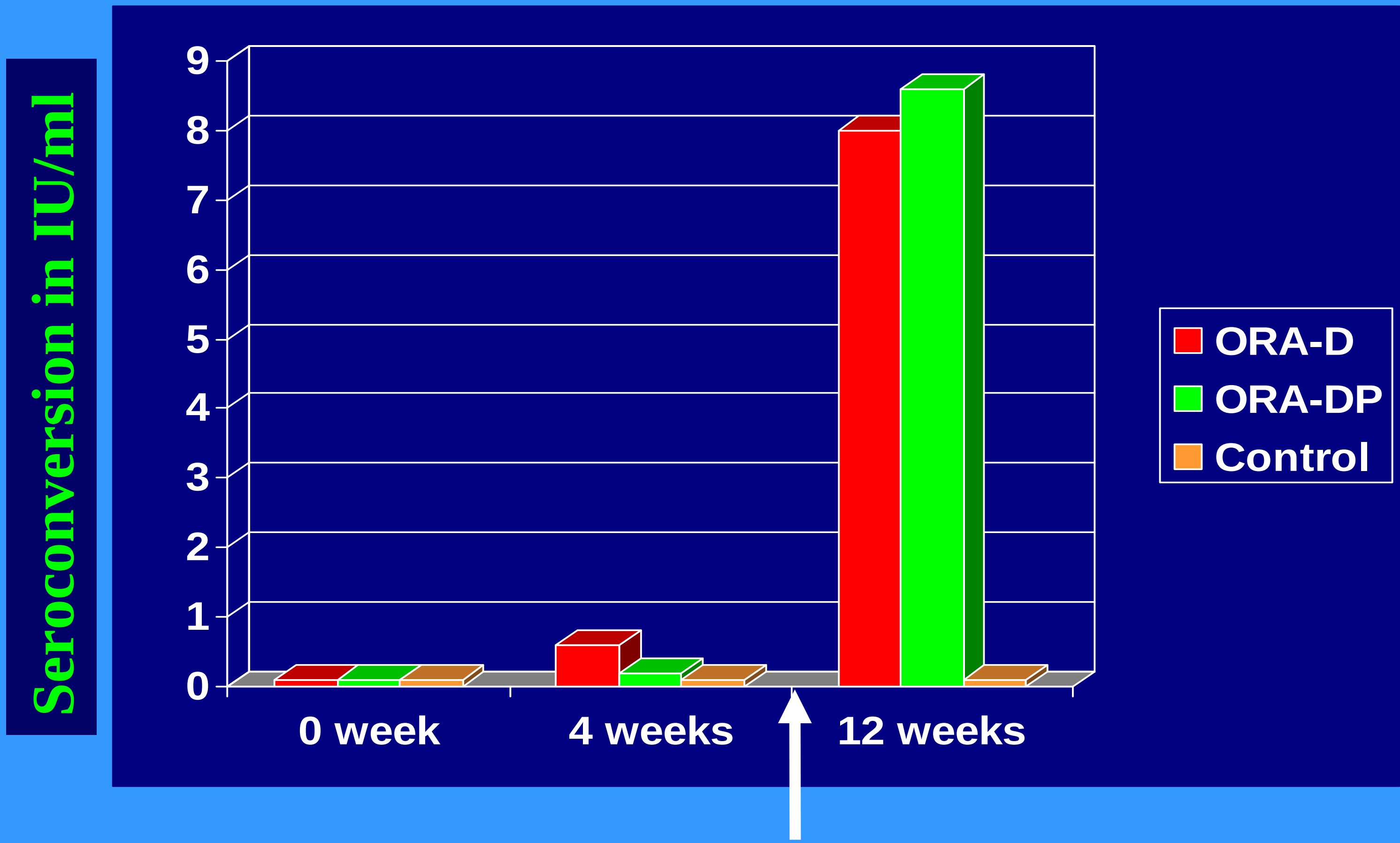
Relation between vaccine strains



Efficacy of Rabies Viruses in Mice

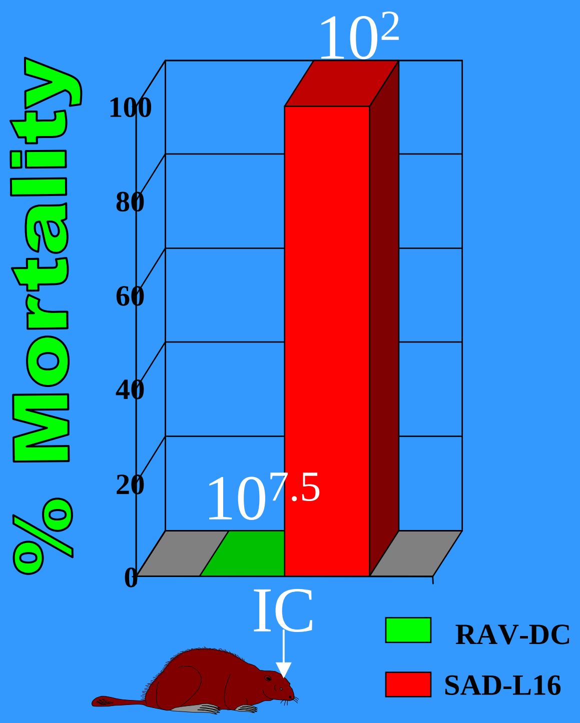


Efficacy of further attenuated Oral Rabies Vaccine Candidates in Dogs



Booster dose 8 weeks after the 1st immunization

Safety of RAV-DC in 3-week- old mice after i.c. inoculation



Safety and Efficacy Evaluation of the Oral Rabies Vaccine SAG2 in Indian Stray Dogs

J. Barrat[1], E. Picard[1], H.K. Pradhan[2], B. Pattnaik[2], S.S. Patil[2], P.R. Vanamayya[2], Rich Sood[2], Ripudaman Singh[3], J.P. Gurbuxani[3], F.X. Meslin[4], A. Régnault[5] and F. Cliquet[1]

[1] AFSSA – LERRPAS, Malzeville, France
[2] HSADL, Indian Veterinary Research Institute, Bhopal, India
[3] Petswill, Ludhiana Punjab, India
[4] WHO, Geneva, Switzerland
[5] Virbac, Carros, France

Several partners in the trial

- Coordinated :
  - Between WHO (Geneva) and VIRBAC (France)
- Executed and conducted :
  - By WHO Collaborating Centre for Research and Management in Zoonoses Control of Malzeville (France)
  - At High Security Animal Disease Laboratory, Bhopal (India)
- Other collaborator :
  - Petswill, 3 Kesar Complex, Malhar Road, Gurudev Nagar, Ludhiana Punjab (India)

Rabies in India

- Reported from all states of India. More than 20,000 deaths per year
- Approximately 95% of human rabies cases are due to stray dogs bites (jackals 1,7%, cats 0,8%, foxes 3%, …)
- Inadequate laboratory diagnostic facilities
- Estimated dog population : 27 millions ; dog/human ratio = 1/40
- Majority of dogs are stray, unowned and unprotected

Objectives of the trial

To evaluate in Indian stray dogs both the safety and the efficacy of a lyophilised SAG2 (DBL2) bait

- **Safety / innocuity:**
  - To assess the absence of pathogenecity of SAG2 for dogs even immunodepressed ;
  - To assess the absence of salivary excretion of the vaccinal virus
- **Efficacy:**
  - To assess the protection induced by the vaccine in dogs

Lyophilised DBL2 bait
(SAG2, Virbac, France)
Selected from SAD Bern strain and two successive mutations of the Arginin 333 codon

Final objective:

To associate parenteral vaccination of owned dogs and oral vaccination of stray dogs in a rabies control project

Animals used in the study (1)

- Dogs aged 6-12 months, all originating from Bhopal. Till challenge, all animals were kept in a traditional animal facility

- Identification with collar & numbered tag

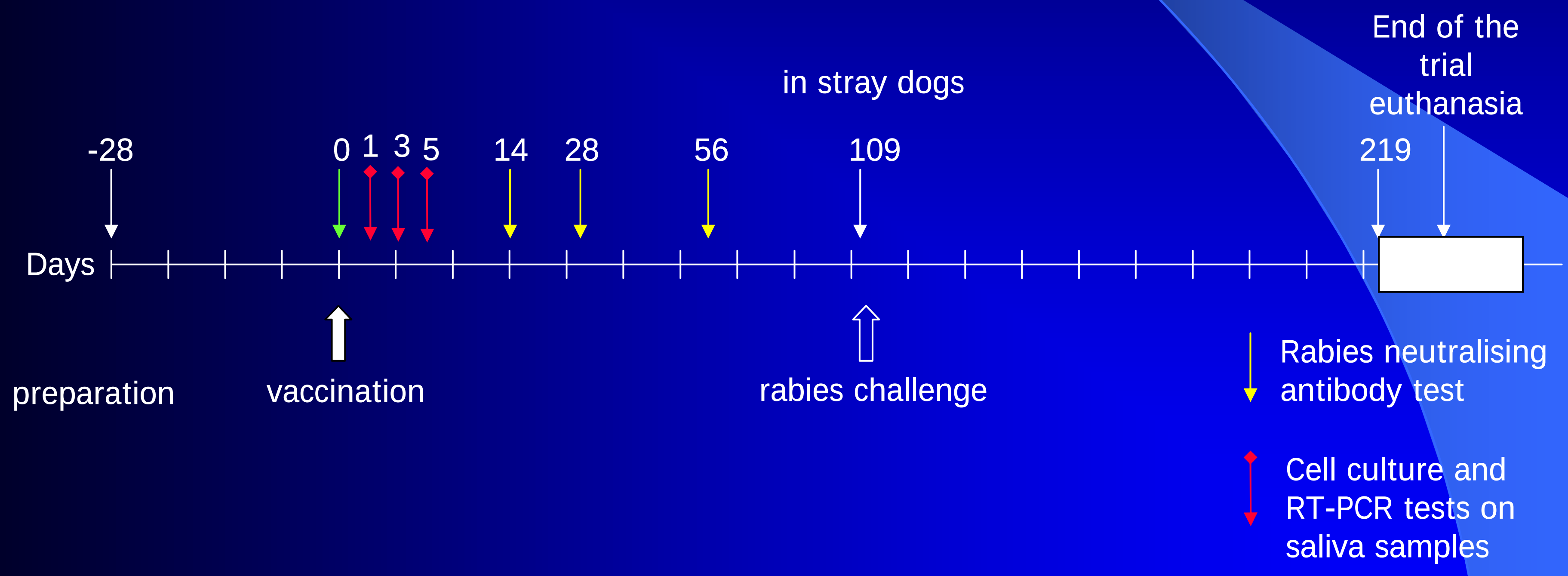
- Dewormed and vaccinated against canine dispemper, Rubarth hepatitis, parvovirosis and leptospirosis

Initial experimental design

		group	number of dogs	vaccine	route	frequency
safety	vaccinated group	I	5	Rabidog[R] bait	oral	single bait
safety	vaccinated group	II	5 (immuno-depressed)	Rabidog[R] bait	oral	single bait
safety	control group	III	5	-	-	-
efficacy	vaccinated group	IV	11	Rabidog[R] bait	oral	single bait
efficacy	control group	V	5	-	-	-

Comparison of the bait acceptance, efficacy and safety of lyophilised SAG2 bait

Schematic representation of the experimental protocol



Adaptation

- Groups I ("normal" vaccinees of safety trial) and IV (vaccinnees of efficacy trial) have no treatment between D0 (vaccination) and D109 (challenge).
- The control groups III and V have been treated identically till D109.

- Salivary excretion, health and serological response have been monitored on 12 dogs (with 8 controls)

Vaccination and challenge

- **Vaccination**
  - Titre of lyophilized Rabidog[R] vaccine bait : $10^{8.50}$ $TCID_{50}$/dose
- **Rabies challenge**
  - Intramuscular challenge with the supernatant of homogenised sub-maxillary salivary glands of naturally rabid dogs titrating $10^{6.5}$ $MICLD_{50}$/ml
  - Strain: Street rabies virus, Tunisian origin, passaged once in dogs
  - Injection of 100 $MICLD_{50}$ of virus per animal at D109 (more severe test than 28 days post-baiting)
- **Clinical observation**
  - Daily observation all along the experiment and any unusual sign recorded.
  - 110 days post-challenge, euthanasia of all surviving dogs (D219)

Results

Bait acceptance and selection of dogs

		group	number of dogs	VNA titre > 0,5 IU/ml at D0	Bait uptake	Time (min)
safety	vaccinated group	I	5	* 1/5	5/5	<3 (2/5) <20 (3/5)
	vaccinated immunodepressed group	II	5	0/5	5/5	<3 (5/5)
	control group	III	5	* 2/5	-	-
efficacy	vaccinated group	IV	11	* 2/11	11/11	<2 (11/11)
	control group	V	5	0/5	-	-

- all vaccinated dogs had eaten totally their bait in less than 20 min (70% < 3 min)
- * 5 dogs have been rejected (virus neutralizing activity > 0.5 IU/ml at D0)

Safety trial

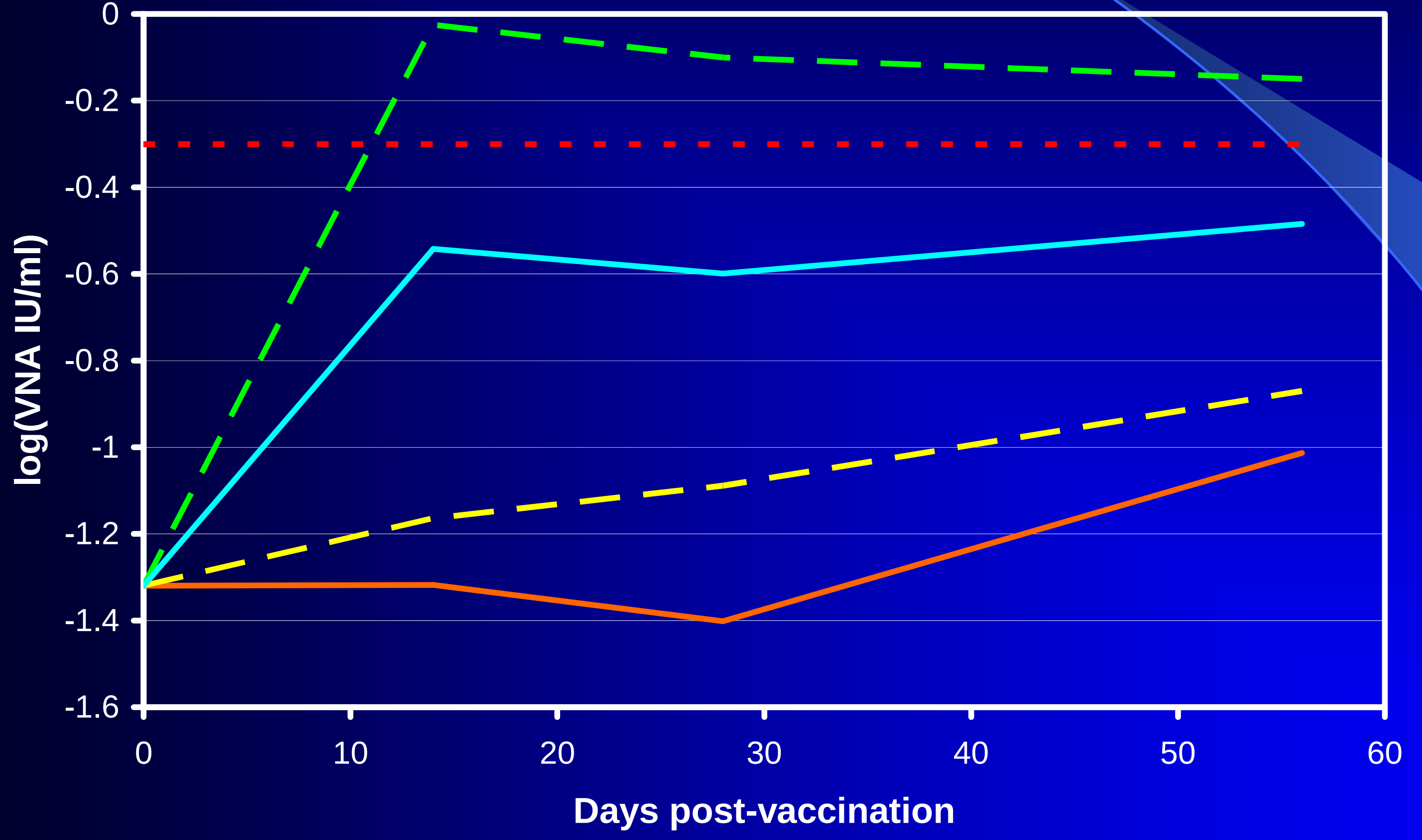
	group	number of dogs	dead	clinical signs	Salivary excretion (CC and PCR) D0 before, after baiting D1, D3, D5	Rabies diagnosis (FAT) at D219
vaccinated groups	I and IV	4 + 8	0/12	no	Neg (0/12)	Neg (0/12)
vaccinated immunodepressed group	II	5	0/5	no	Neg (0/5)	Neg (0/5)
control groups	III and V	3 + 5	0/8	no	Neg (0/8)	Neg (0/8)

- Clinical observation : all animals remained healthy all along the 219 days observation period

- No salivary excretion by using cells culture inoculation and no specific RNA detection using RT-PCR

Efficacy trial

Rabies virus neutralising antibodies (VNA)



8 controls

13 vaccinees

7 positive vaccinees

6 negative vaccinees

Threshold 0.5IU/ml

vaccinated dogs (groups 1 and 4) : blood samples have been taken at D 14, 28, 56

Survival after challenge

	group	Number of dogs	Number of positive for VNA	dead	Mortality delay (days)	Clinical phase (days)	Rabies diagnosis (FAT)
Vaccinated group	IV	9	5/9	0/9	-	-	Neg (9/9)
Control group	V	5	0/5	5/5	25 - 85	6 - 8	Pos (5/5)

FAT: performed on brain and salivary glands on D219, i.e. 110 days post-challenge

- 9/9 vaccinated dogs resisted a virulent challenge that killed 5/5 controls

Conclusions of safety trial

- No salivary excretion of infective particle or virus RNA
- No vaccinal rabies
- Absence of adverse symptoms even in imuno depressed dogs
- Absence of replication of SAG2 strain in brain and salivary glands of all vaccinated dogs

☞ *The innocuity of SAG2 vaccine is then established in dogs*

Conclusions of efficacy trial

- Bait acceptability: 100 % in less than 20 min

- Protection:
  - *Serological testing shows that 50% of vaccinated dogs can be considered protected*
  - *Resistance to a virulent challenge that killed unvaccinated controls assesses the protection induced by SAG2 vaccine*

☞ ***The efficacy of SAG2 vaccine is then established***

- The protective effect of SAG2 vaccine had been already demonstrated on laboratory and field dogs and on other species
- Efficacy established on:
  - jackals in Zimbabwe (Bingham *et al.*, 1999)
  - captive artic foxes (Follman *et al.,* 2004)
  - …
- Safety and efficacy shown in Tunisian dogs and South African puppies (Schumacher *et al.*, 1999)
- Establishment of the safety:
  - on Tunisian animals (Hammami *et al.*, 1999) :
    - Dogs (adult and young)
    - Domestic cats
    - Local animal species susceptible to consume vaccine baits: jackal, jerboa, merion, gerbil

afssa

Final conclusion

- SAG2 confirmed to be:
  - Safe for people and dogs because
    - neither salivary excretion
    - nor adverse clinical signs
    - or replication of virus were observed in vaccinated dogs
  - Efficient for vaccination
    - as half of vaccinated dogs seroconverted and
    - as all vaccinees resisted a virulent challenge

☞ SAG2 vaccine may be used in the field to orally vaccinate Indian stray dogs for rabies control

To finish: some pictures…
"Outside" quarantine facility of HSADL,
before challenge

Bait acceptance

- All tested dogs had totally eaten the baits :
  - 16/21 in less than 3 minutes
  - 21/21 in less than 20 minutes

Acknowledgments

- This work was supported by WHO
- F.X. Meslin from WHO (Geneva), H.K. Pradhan from HSADL (India) and J.P. Gurbuxani from Petswill (India) for his unfailing support
- MJ. Barrat, A.Verdot, A. Servat, JL. Schereffer, A. Hamen, G. Farré from AFSSA for expert technical assistance
- André Aubert

Thank you for your attention.

Elimination of canine rabies in an isolated area: A pilot project

Number of human rabies deaths per country, Asia, year 2001

Proposed project

1. Who?
2. What?
3. Where?
4. When?
5. Why?
6. How?

Who would be involved?

- Global involvement of rabies experts and rabies centers that can help make the program a success
- National government
- WHO/OIE/NGOs etc

What would the project involve?

- To design, write, and eventually submit a proposal for a large amount of money to eliminate canine rabies on an island
- To use this project as a “showcase” for other countries and funding organizations to prove that canine rabies can be eliminated in a geographic area

Where would it be located?

- Isolated geographic region
  - Need for specific ‘borders’ for surveillance, to prevent re-entry, to stepwise vaccinated and eliminate canine rabies
  - Island
- Need for governmental support
- Need for KOL to be in place
- Past experience would be a bonus

Where?

- Several islands were discussed
- A review of Sri Lanka – Why?
  - Big enough to prove that it can be done
  - KOLs are in place and have been trained in various WHO CC around the world
  - Previous history, and ongoing projects involving the use of oral canine rabies
  - Intradermal use of PEP for humans
  - Tsunami relief efforts – international awareness

Where?

- Evaluation visit by a team from WHO
- November visited Sri Lanka
- Field visits, met National Health Authorities
- Will it be feasible?...

Sri Lanka – considerations

- De-centralized government
- New government
- Tamil tigers in the north and east
- Centralized diagnostic lab
- Need to involve veterinarians

Sri Lanka - considerations

- Dedicated professionals
- Funding support from government
- KOLs that have connections with government
- 70 % of dogs are accessible

When?

- Proposal needs:
  - To be well designed
  - Well written to capture attention of foundation
  - √ 'Buy-in' globally from rabies experts
  - Background information on key persons to be contacted to help support the project internally

WHEN will it be ready? 2006

Why?

- Ongoing discussion among rabies experts
- Need for “showcase” for proof of concept
- To secure financial support from a large foundation
- Historical evidence that this is possible
- Recent success story in Latin America

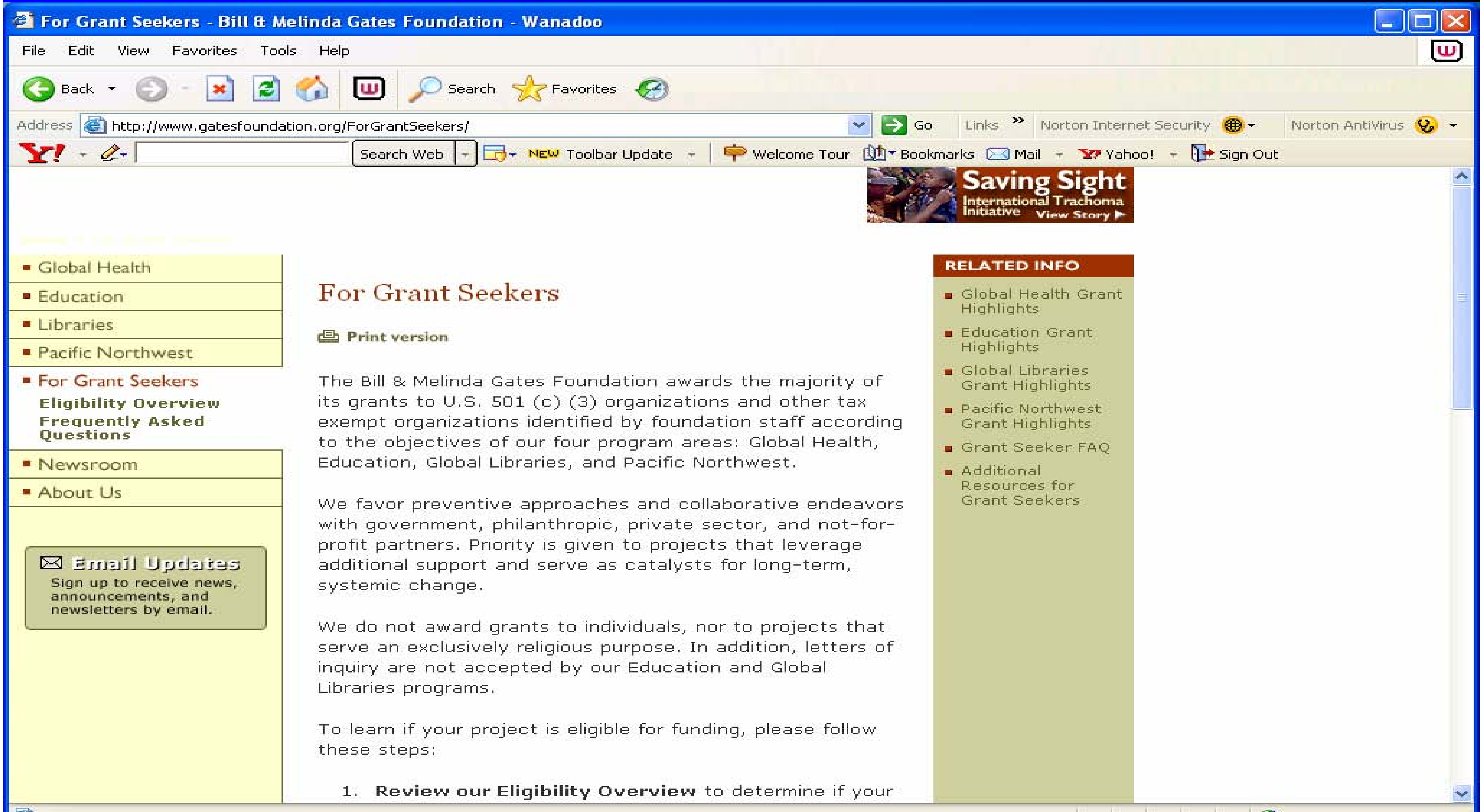
How?

- How much will it cost?
- MILLIONS!!!
- Who has THAT much money?
- GATES – and a few others

How?

- Chose the right location
- Research funding organization
- Commitment from national government
- Organize joint funding from several sources

GATES foundation



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Saving Sight International Trachoma Initiative View Story

- Global Health
- Education
- Libraries
- Pacific Northwest
- For Grant Seekers
  - **Eligibility Overview**
  - **Frequently Asked Questions**
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For Grant Seekers

Print version

The Bill & Melinda Gates Foundation awards the majority of its grants to U.S. 501 (c) (3) organizations and other tax exempt organizations identified by foundation staff according to the objectives of our four program areas: Global Health, Education, Global Libraries, and Pacific Northwest.

We favor preventive approaches and collaborative endeavors with government, philanthropic, private sector, and not-for-profit partners. Priority is given to projects that leverage additional support and serve as catalysts for long-term, systemic change.

We do not award grants to individuals, nor to projects that serve an exclusively religious purpose. In addition, letters of inquiry are not accepted by our Education and Global Libraries programs.

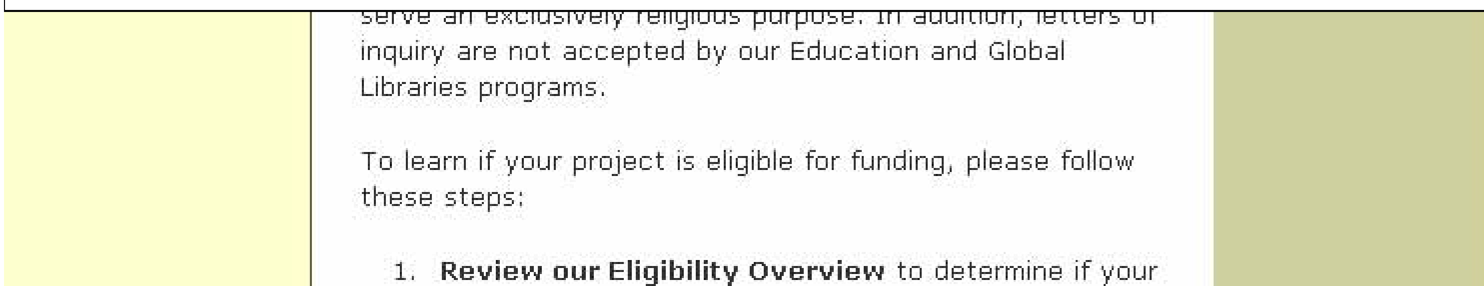
To learn if your project is eligible for funding, please follow these steps:

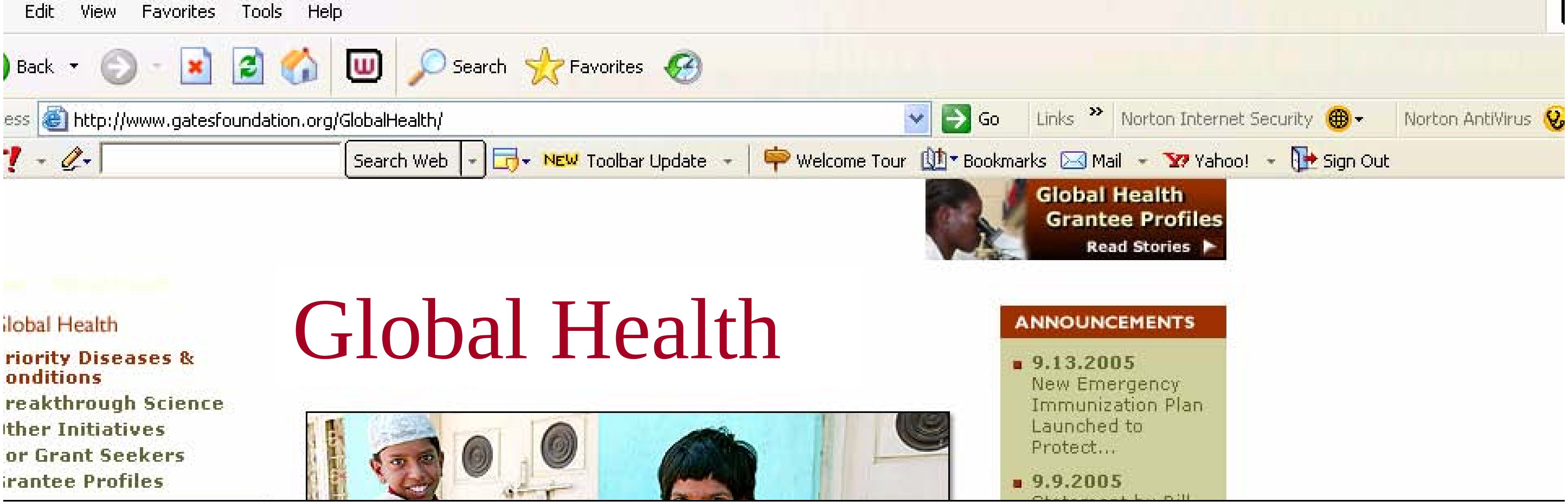
1. **Review our Eligibility Overview** to determine if your

RELATED INFO

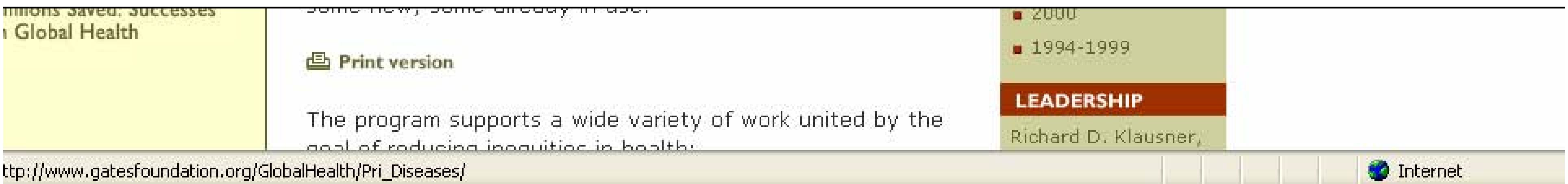
- Global Health Grant Highlights
- Education Grant Highlights
- Global Libraries Grant Highlights
- Pacific Northwest Grant Highlights
- Grant Seeker FAQ
- Additional Resources for Grant Seekers

"We favor preventative approaches and collaborative endeavors with government, philanthropic, private sector, and not-for-profit partners. Priority is given to projects that leverage additional support and serve as catalysts for long-term systemic change."





"The mission of our Global Health program is to ensure that people in the developing world have the same chance for good health as people in the developed world. We see a tremendous opportunity to save millions of lives through the development and distribution of health tools and strategies – some new, some already in use."



Priority Diseases and Conditions

"We support efforts to prevent and treat diseases and conditions that meet three criteria: (1) they cause widespread illness and death in developing countries; (2) they represent the greatest inequities in health between developed and developing countries; (3)they receive inadequate attention and funding."

Our priority diseases are:

Acute Diarrheal Illness

The foundation helps fund work aimed at preventing and treating infectious diarrhea, which contributes to the deaths of 2 million to 3 million young children each year.

Read more ▶

Acute Lower Respiratory Infections

Respiratory illnesses like pneumonia kill about 2 million young children every year. The foundation aids efforts to improve diagnosis and develop better vaccines against these common infections.

Read more ▶

Child Health

Much of the work on specific diseases supported by the foundation benefits children. In addition, we assist in broader efforts to improve child survival, by supporting efforts to prevent millions of deaths among newborns.

Read more ▶

HIV/AIDS

To slow the global spread of HIV, the foundation supports the development of vaccines and other tools and strategies with the potential to prevent tens of millions of infections and deaths. We also fund comprehensive initiatives that include both prevention and treatment.

Read more ▶

include both prevention and treatment.

Read more ▶

Malaria

The foundation's grantees are working in many areas to reduce the burden of malaria on the world's poorest countries. Their work includes the development of vaccines to prevent the disease and large-scale efforts to control malaria by making better use of existing tools.

Read more ▶

Poor Nutrition

The foundation assists efforts to improve nutrition, including developing foods and crops that are high in essential vitamins and minerals, and ensuring that healthy foods get to those who need them most.

Read more ▶

Reproductive and Maternal Health

To improve the health of women in the developing world, the foundation supports efforts to reduce deaths and illness related to pregnancy and prevent unintended pregnancies.

Read more ▶

Tuberculosis

The foundation supports work in the prevention and treatment of TB, including the search for improved vaccines, better tools for diagnosis, and new drugs to treat active TB.

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Vaccine preventable diseases

immunization, an innovative partnership that has helped save hundreds of thousands of lives by increasing the number of children who receive basic vaccines.

Read more ▶

Other Infectious Diseases

Developing countries are disproportionately affected by other common diseases, including sexually transmitted infections, infections involving multicellular organisms such as worms, and those caused by parasites transmitted through insects.

Read more ▶

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QUICK LINKS

Global Health	Infectious Diseases | HIV/AIDS & TB | Reproductive & Child Health
Education	Increasing U.S. Graduation Rates | Scholarships | Research & Evaluation
Global Libraries	U.S. Library Program | International Library Initiatives
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Children's health

Rabies is a disease of children 40 to 60 % of victims are children less than 15 years

Photo: M Warrell

Photo: BJ Mahendra

Photo: S Scholand

Where do we stand?

- Very preliminary proposal ready
- Have ‘buy in’ from global rabies experts
- Have completed Sri Lanka visit
- Waiting for a response from the Sri Lankan authorities
- Have identified potential funding partners


Finally
We must take control of our own destiny

Human rabies prophylaxis: basis and new challenges

Yolande Rotivel

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Rabies is a viral zoonosis

Negative strand, bullet shaped RNA virus

Transmission from animals to humans

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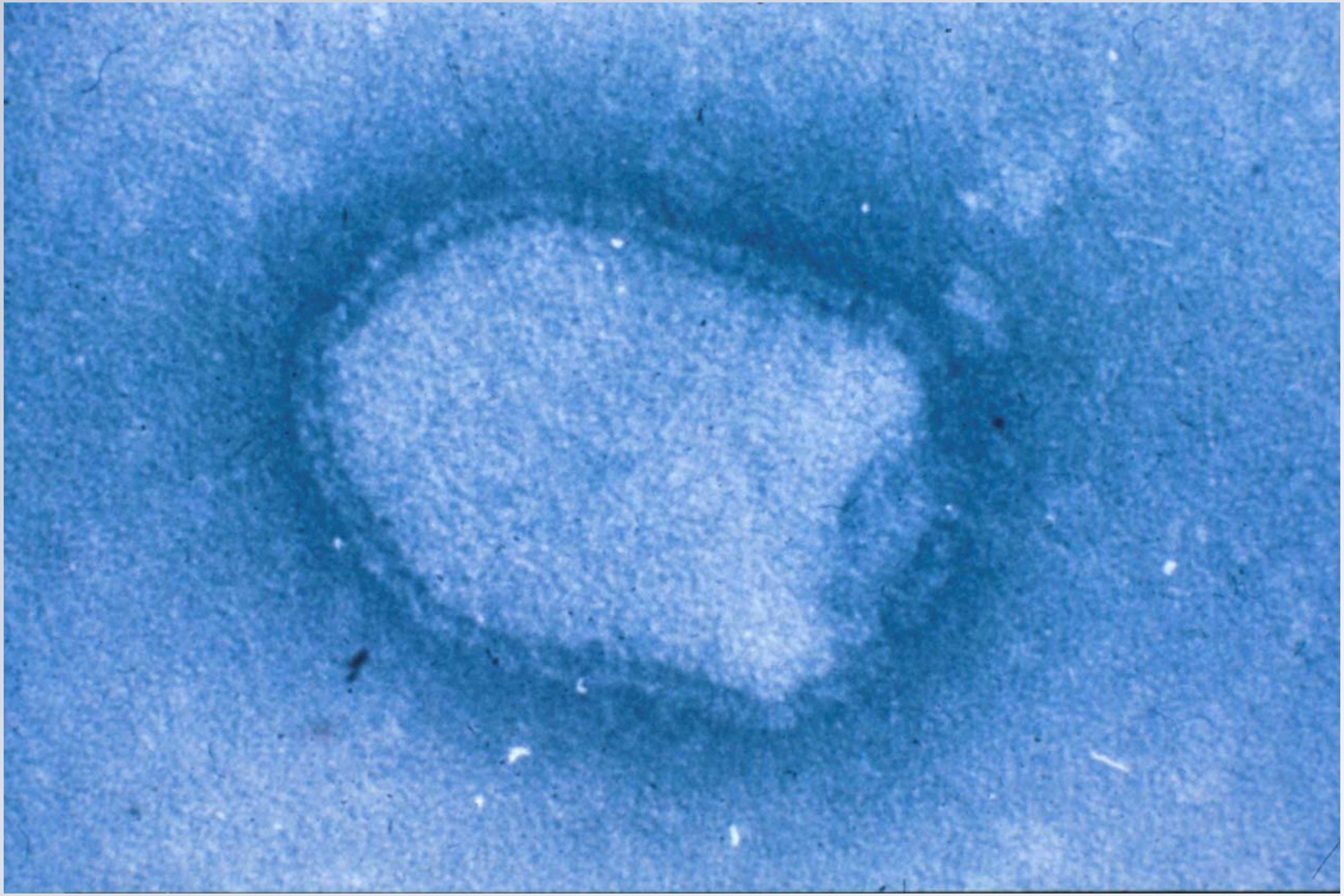

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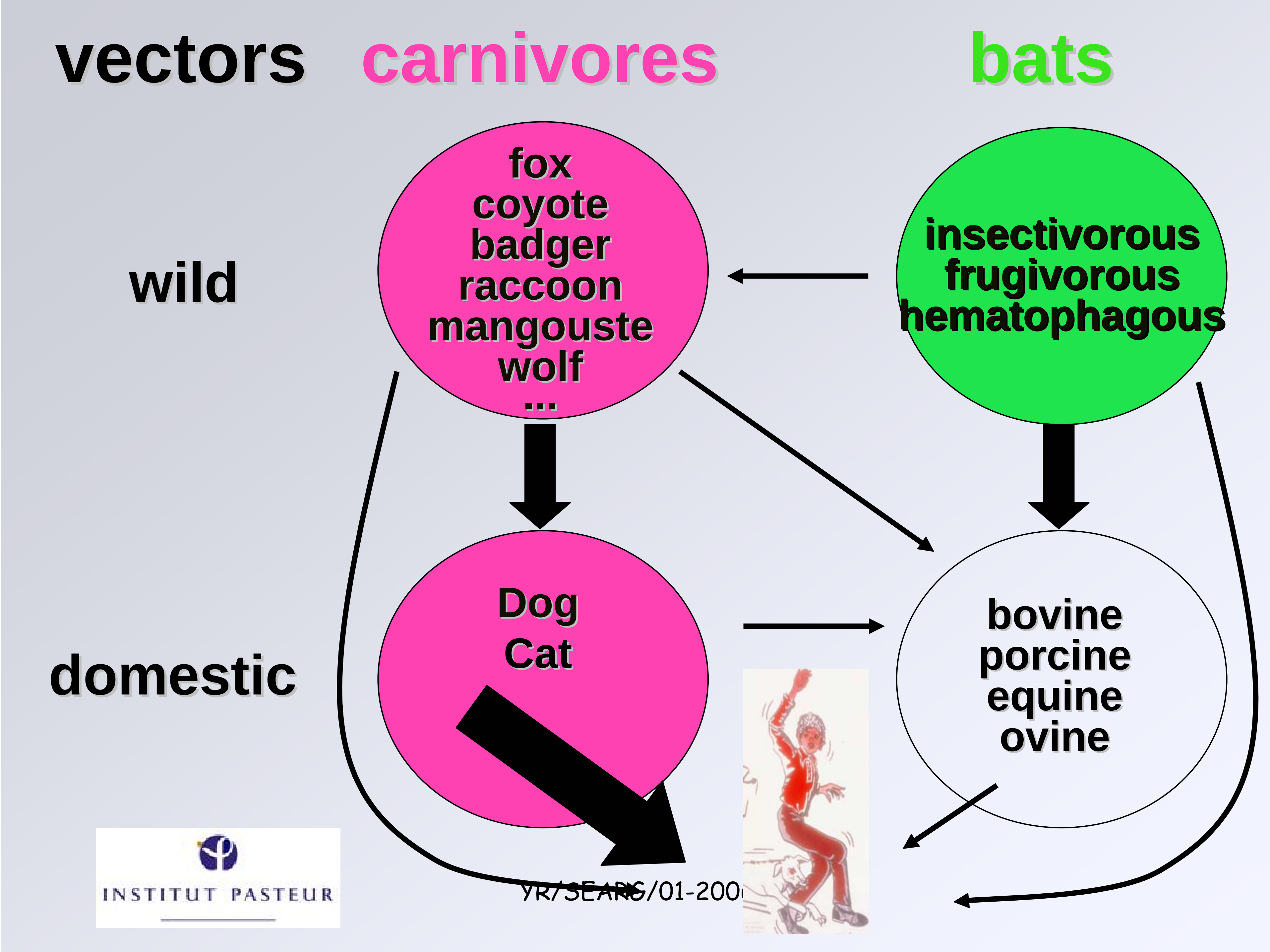
Transmission from animals to humans

Dogs : 90%
But homeotherms mammals
Carnivorous either domestic or wild :
Cats
Monkeys
Foxes
Mongooses
Wolves
Raccoons…
Bats


vectors
carnivores
bats
wild
domestic
fox
coyote
badger
raccoon
mangouste
wolf
...
insectivorous
frugivorous
hematophagous
Dog
Cat
bovine
porcine
equine
ovine
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YR/SEARG/01-200

Canine rabies, Street rabies

Selvatic rabies

DOG

WILD CARNIVOROUS

CHIROPTERS

Rabies free

?

USA

Latin America

Asia, India
Middle-East
North Africa, Eastern Europe
Sub-Saharian Africa

Western Europe
Australia

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Human rabies prophylaxis: Basis

- Avoiding exposure
  - What is an exposure?
  - Non specific treatment of exposure
- Postexposure treatment and preexposure vaccination
  - Vaccines
  - RIG
  - Regimens

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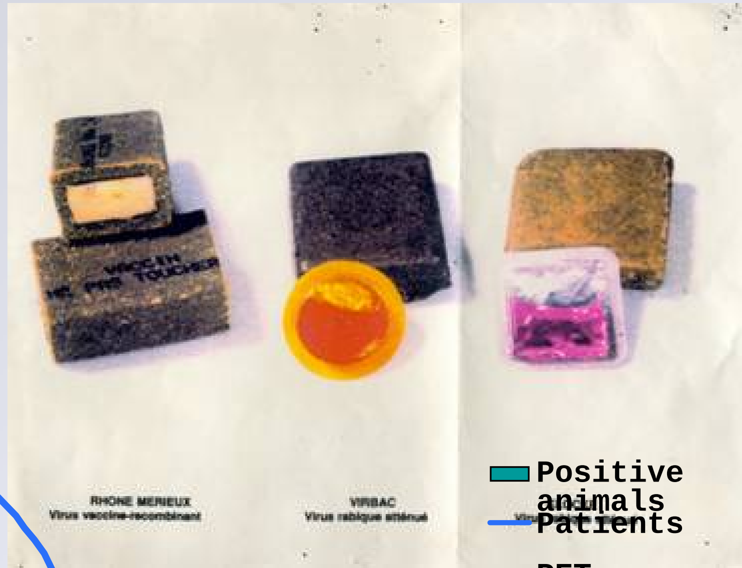
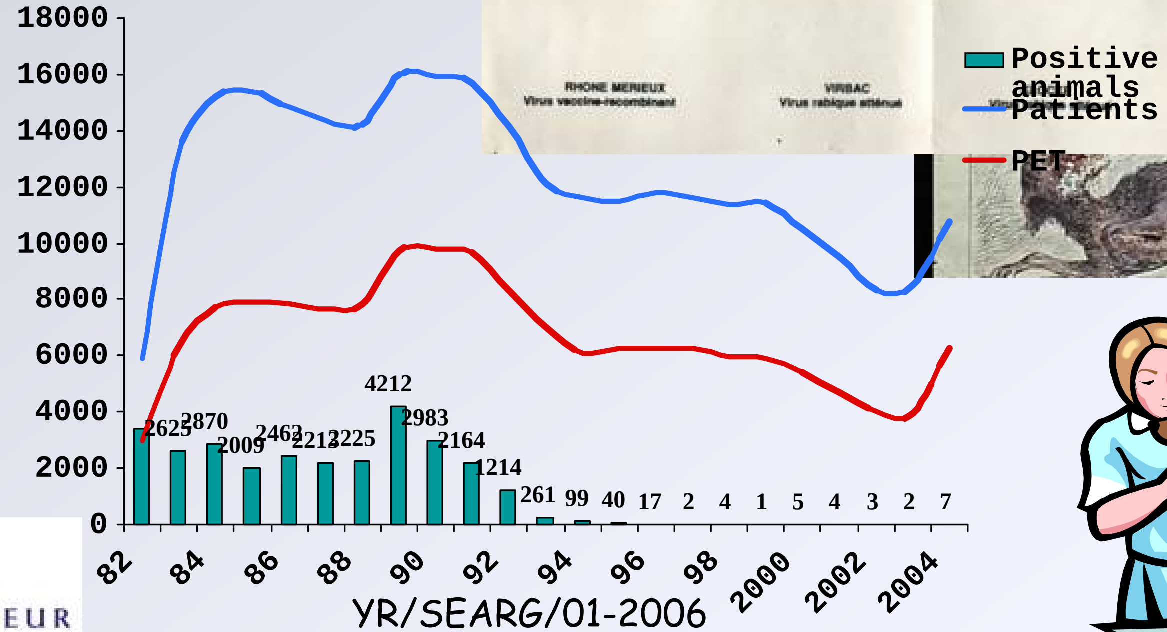
Avoiding exposure

- Vaccination
  - Pets
  - Wild fauna





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Avoiding exposure

- **Safe procedures**
  - **In rabies laboratory**
  - **For veterinarians**
  - **Animals handlers**

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Exposures

WHO 2004

- **Category I : touching or feeding of animals. Licks on intact skin**
- **Category II : nibbling on uncovered skin.Minor scratches or abrasions without bleeding**
- **Category III : single or multiple transdermal bites or scratches, licks on broken skin. Contamination of mucous membranes with saliva (licks). Exposures to bats**

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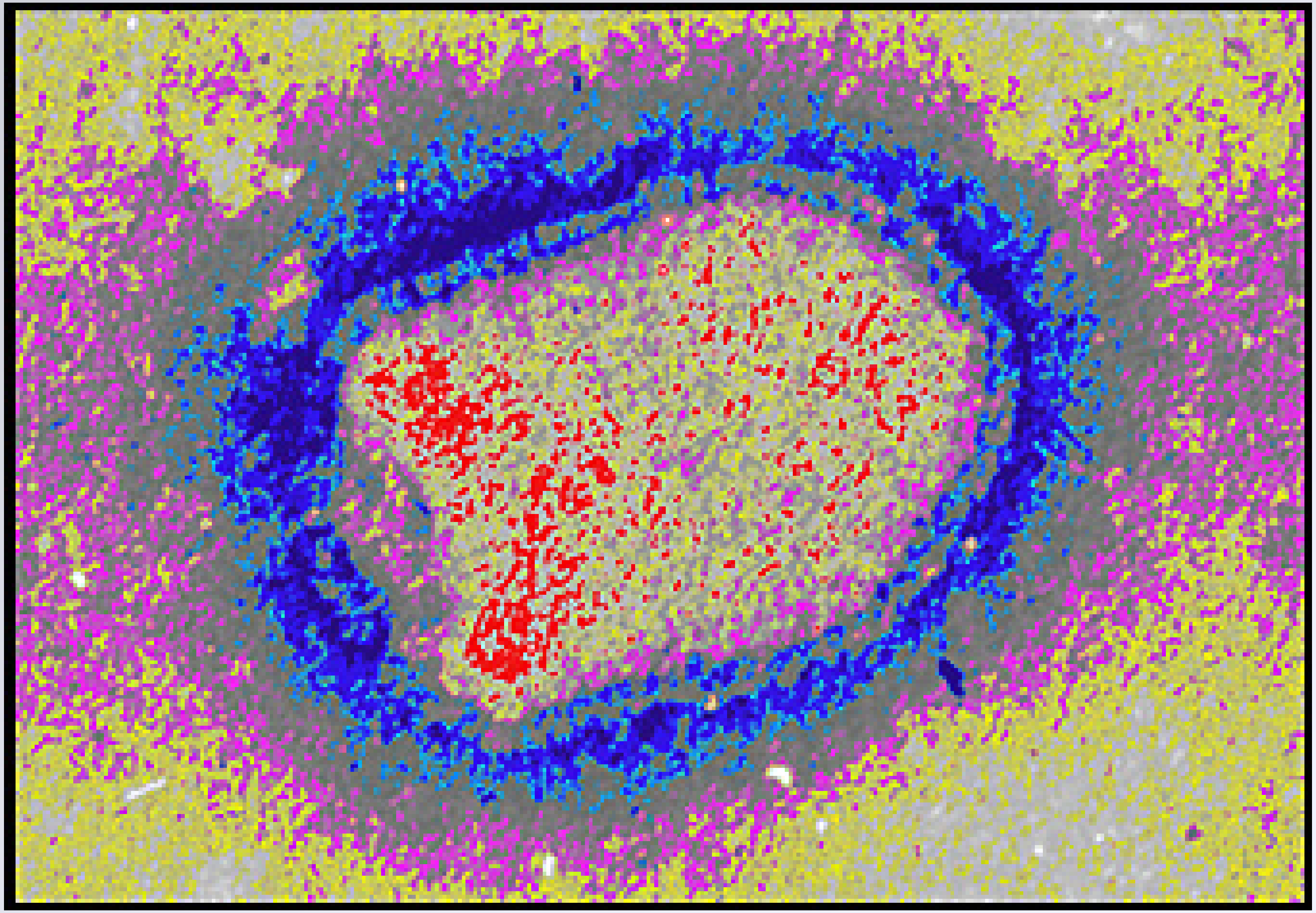
Is the animal excreting rabies virus?



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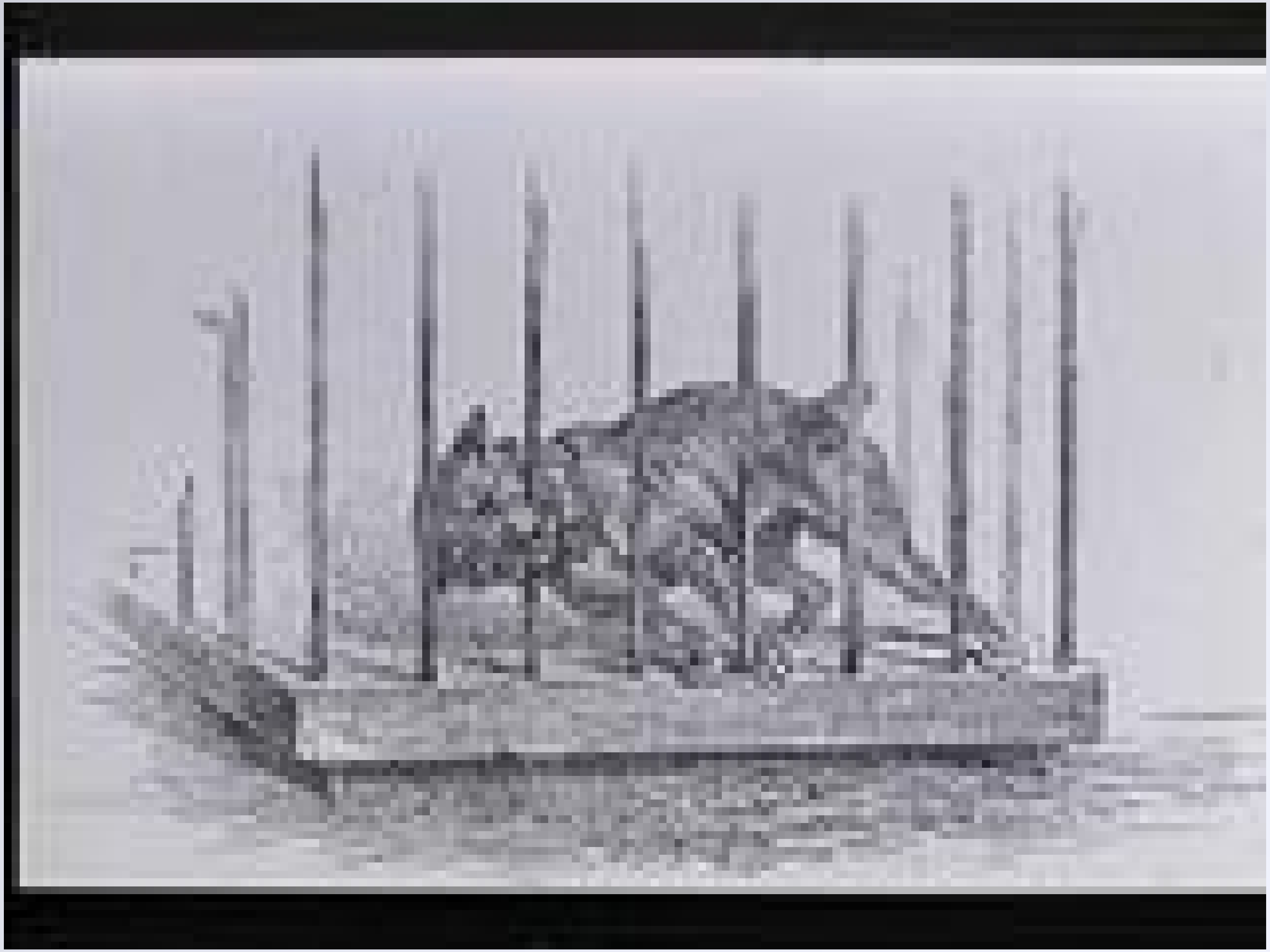

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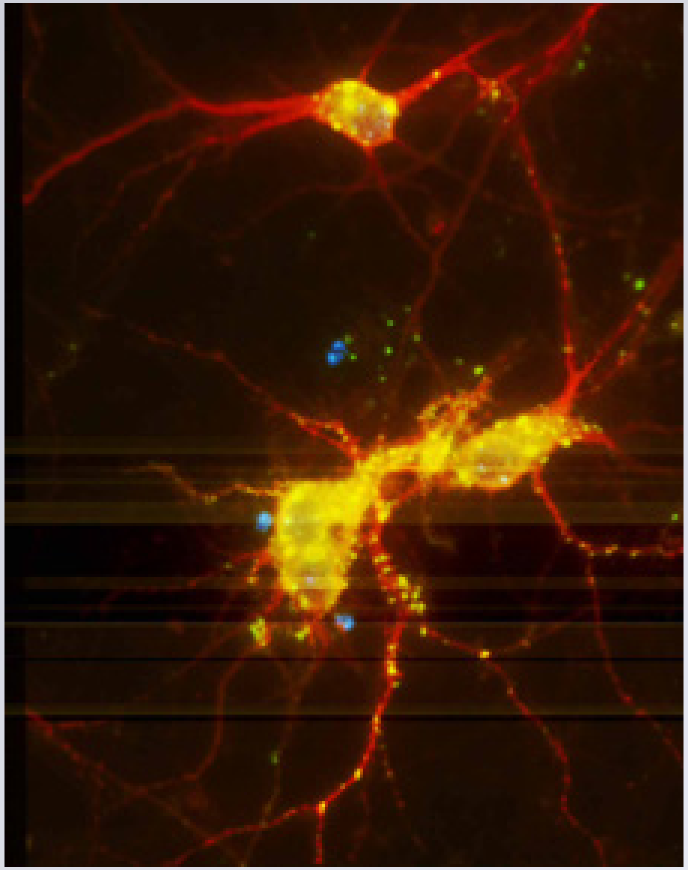

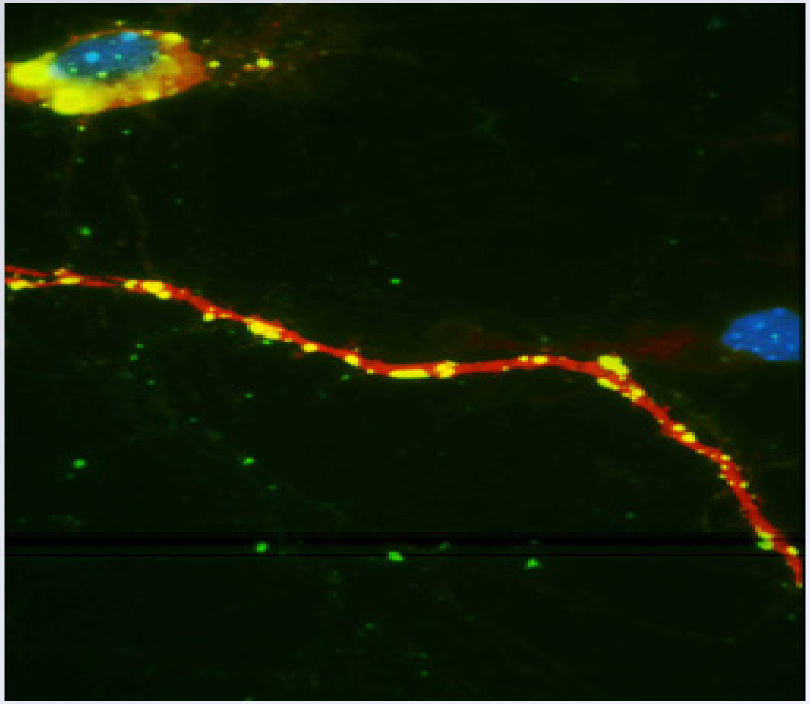

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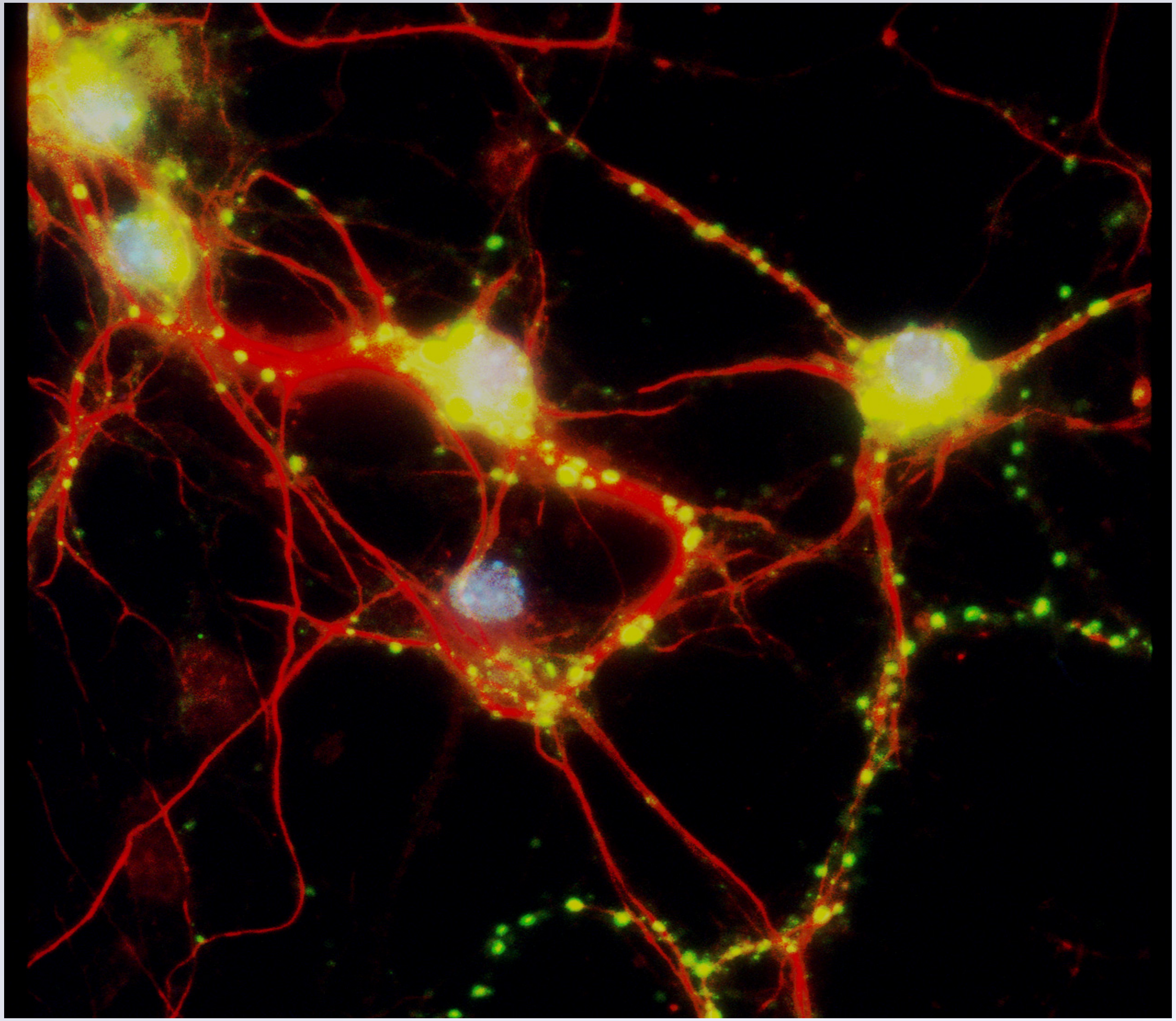

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PE Ceccaldi, Institut Pasteur

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If exposure…

Limiting virus amount

- Immediate and thorough washing of the wound with soap and water, detergent
- Delay suturing
- Antibiotics
- Tetanus toxoid

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WHO expert consultation on rabies
WHO, Geneva 2004
First report
WHO Technical Report Series 931

http://www.who.int/rabies/931/en/index.html

Type of contact, exposure and recommended post-exposure prophylaxis

Category	Type of contact with asuspect or confirmed rabid domestic or wild[a] animal, or animal unaivalable for testing	Type of exposure	Recommended post-exposure prophylaxis
I	Touching or feeding of animals Licks on intact skin	None	None, if reliable case history is available
II	Nibbling of uncovered skin Minor scratches or abrasions without bleeding	Minor	Administer vaccine immediately[b] Stop treatment if animal remains healthy throughout an observation period of 10 days[c] or if animal is proven to be negative for rabies by a reliable laboratory using appropriate diagnostic techniques
III	Single or multiple transdermal bites or scrtches, licks on broken skin. Contamination of mucous membrane with saliva (i.e. licks) Exposure[d] to bats	Severe	Administer rabies immunoglobulin and vaccine immediately. Stop treatment if animal remains healthy throughout an observation period of 10 days or if animal is found to be negative for rabies by a reliable laboratory using appropriate diagnostic techniques

[a] Exposure to rodents, rabbits and hares seldom requires specific anti-rabies post-exposure prophylaxis.
[b] If an apparently healthy dog or cat in or from a low-risk area is placed under observation, the situation may warrant delaying initiation of treatment.
[c] This observation period applies only to dogs and cats. Except in the case of threatened or endangered species, other domestic and wild animals suspected as rabid should be humanely killed and their tissues examined for the presence of rabies antigen using appropriate laboratory techniques.
[d] Post-exposure-prophylaxis should be considered when contact between a human and a bat has occurred unless the exposed person can rule out a bite or scratch, or exposure to a mucous membrane.

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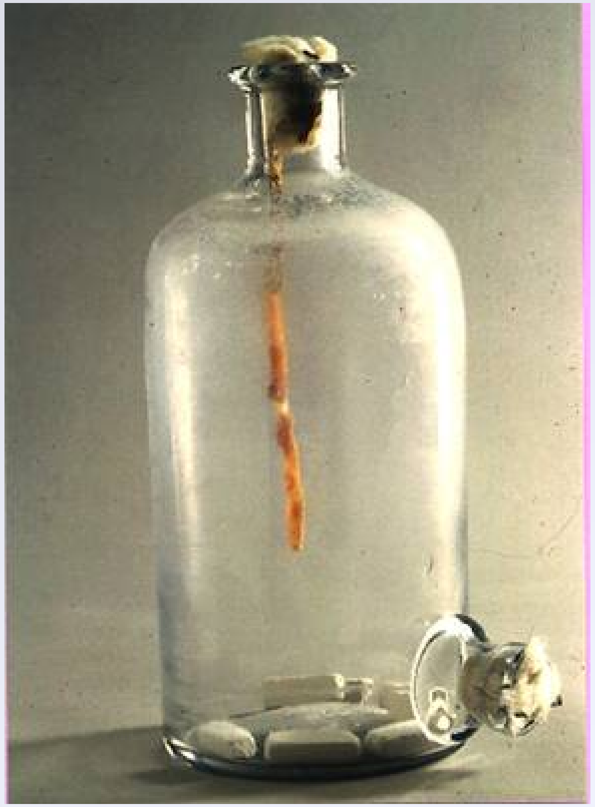
Pasteur's PET

- **Daily injections of a vaccine made of dessicated (partly? Inactivated) spinal cord**



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Modern rabies vaccines

- Produced on cell culture or embryonated eggs
- >2.5 IU/dose
- Safe : no neurological and immunological adverse reactions
- Increased immunogenicity : less injections

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Rabies immunoglobulin

- **Equine Rabies ImmunoGlobulin or F(ab')2**
  - **40 UI/Kg**
- **Human Rabies ImmunoGlobulin**
  - **20 UI/Kg**

Locally infiltrated around the wound(s)

(IM if no feasible only)

0n day 0, no later than day 7

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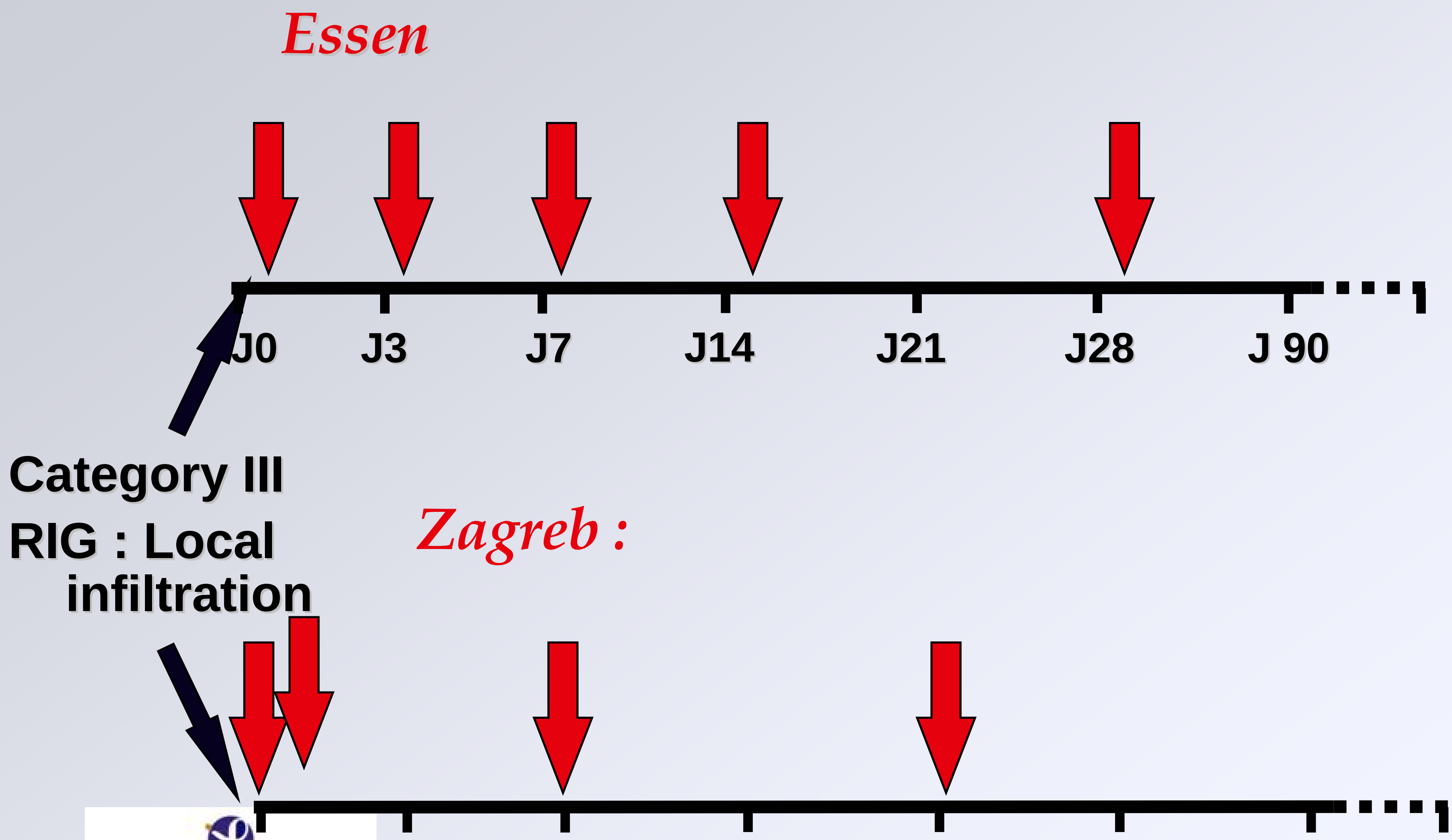
IM regimens WHO 2004



IM injection
in the deltoid muscle

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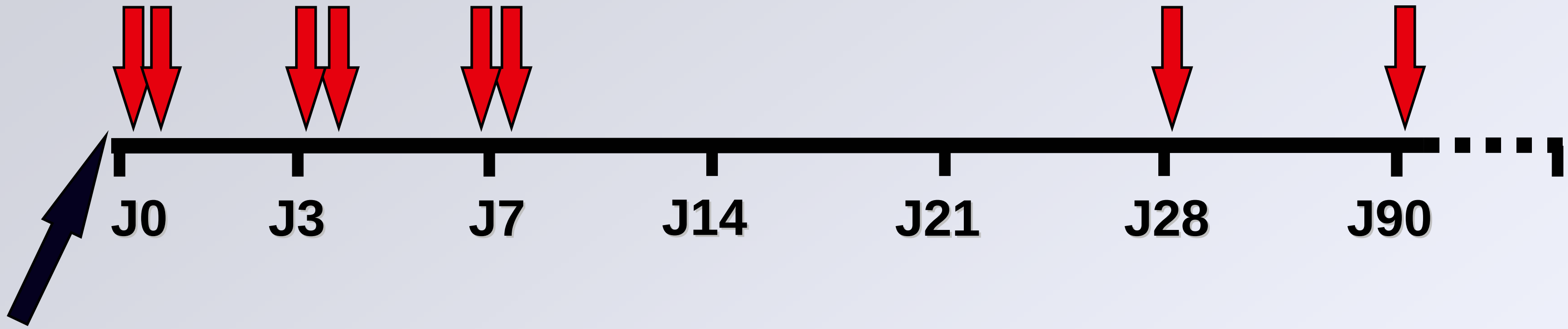
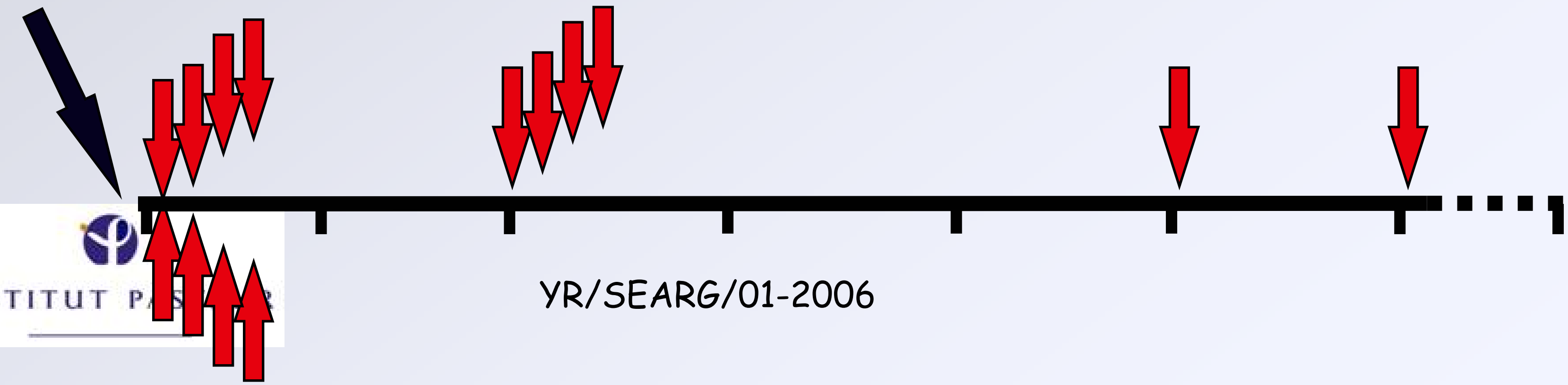
Intradermal regimens WHO 2004
0.1 ml/injection

Thai Red Cross



Category III
RIG :Local
infiltration

Oxford: 1,4 - 2,8 ml



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CORRECT PROCEDURE FOR INTRADERMAL VACCINE INJECTION

Inserting needle intradermally

Injecting vaccine intradermally against resistance

Final blanched papule

Appearance on day 7 of 2-site id vaccine course, showing two previous injection sites

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Benefits IM/ID

- IM
  - Immunogenicity

  No failure
  - Easy to perform

  No failure
  - Visa of health authorities
  - Conservation

- ID
  - 50% to 80% cost reduction
  - Immunogenicity

  Multisite

  ID route

  But
- Number of patients
- Experimented nurses

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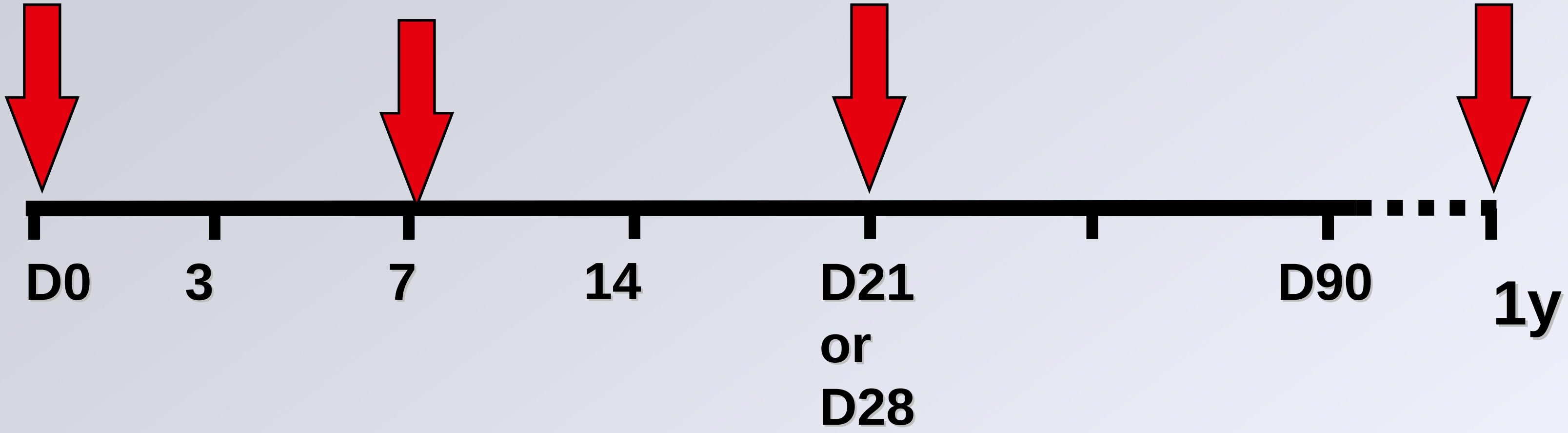
Pre-exposure immunization
WHO 2004



In case of exposure

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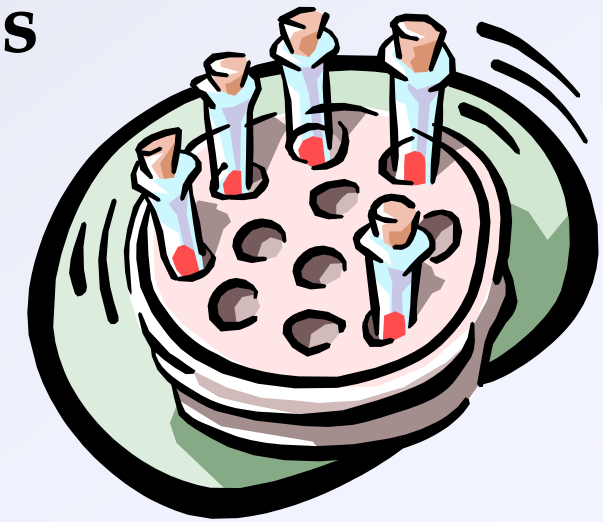
Serological testing

- According to the severity of exposure
  - Laboratory workers : twice a year
- Immunocompromized patients
- If virus neutralizing antibodies titer <0.5 IU/ml a booster dose is recommended



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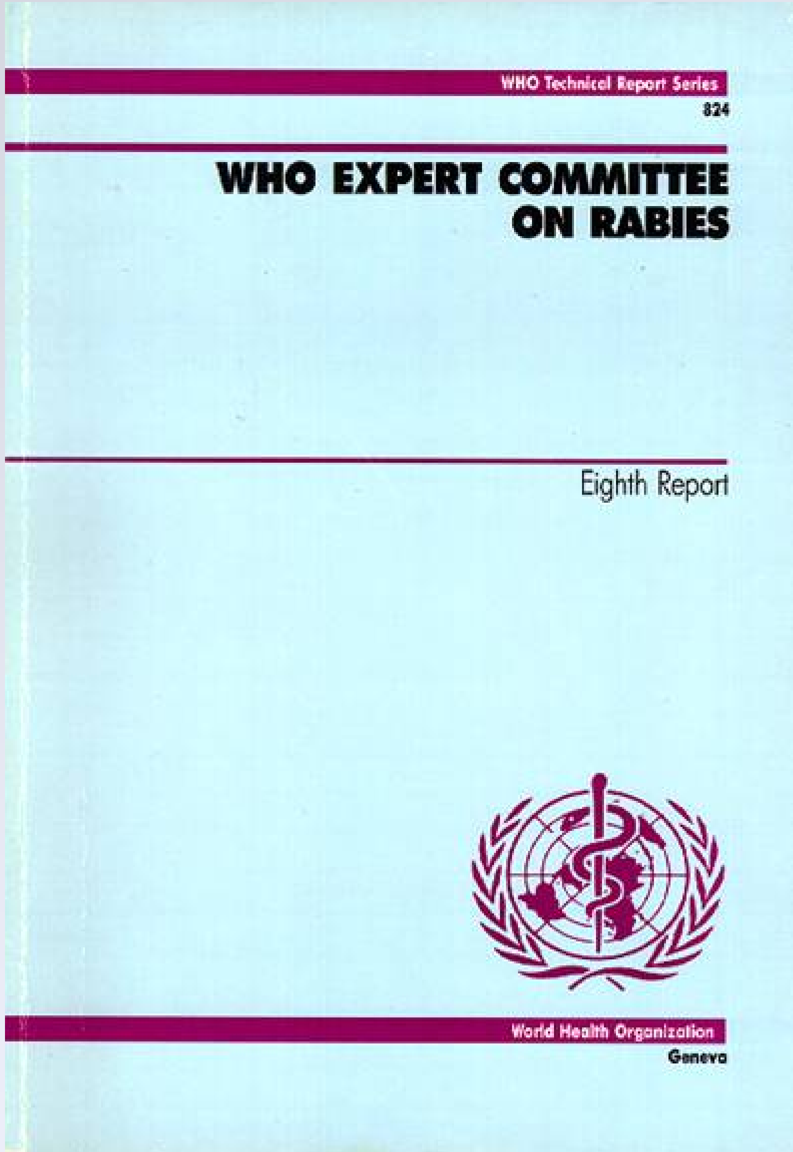
http://www.who.int/

http://www.who.int/rabies/931/en/index.html

Who Expert Consultation on Rabies

October 2004



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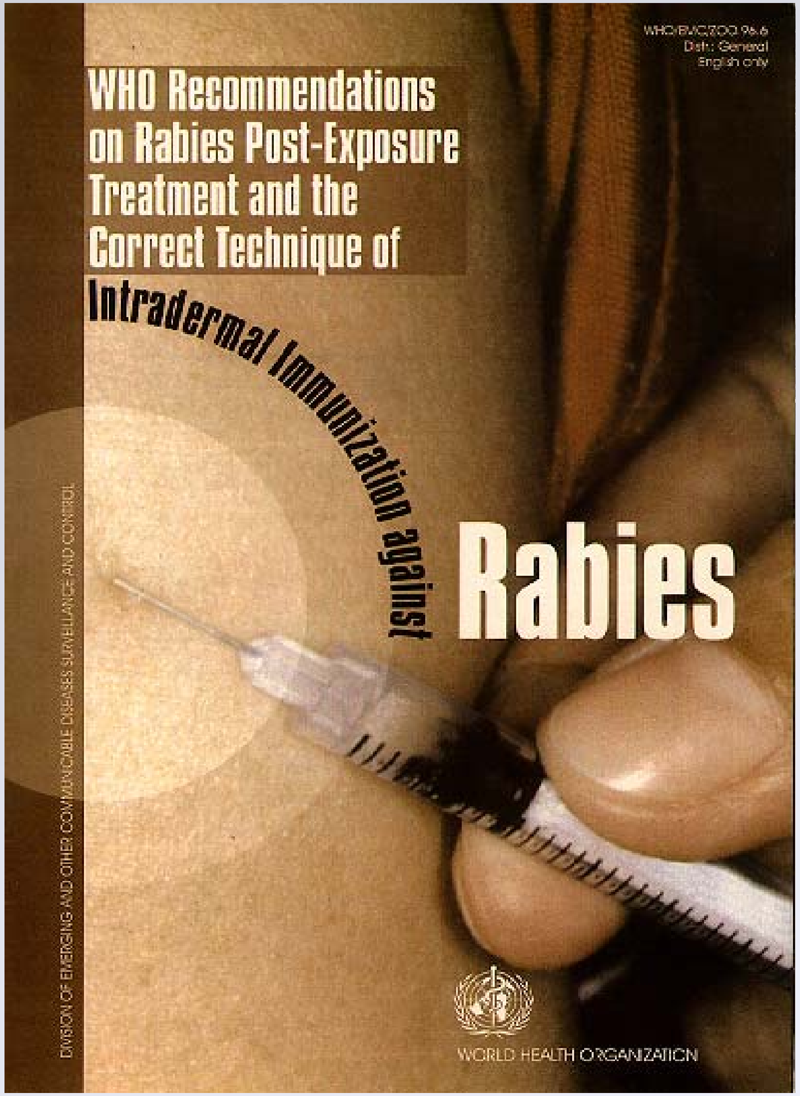

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Despite effective biologicals and performant regimens,

Every year, an estimation of 50 000 deaths occurr, due to

- ignorance of the disease
- lack of biologicals
- lack of education/training
- new epidemiological schemes
- new lyssaviruses

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Current rabies prophylaxis is effective before the virus enters the PNS

- **Washing… must be** immediate
- **PET must be applied** asap
- **PET must be applied** even months or years after the exposure **as the incubation period can be long**

(Smith, NEJM, 1992)

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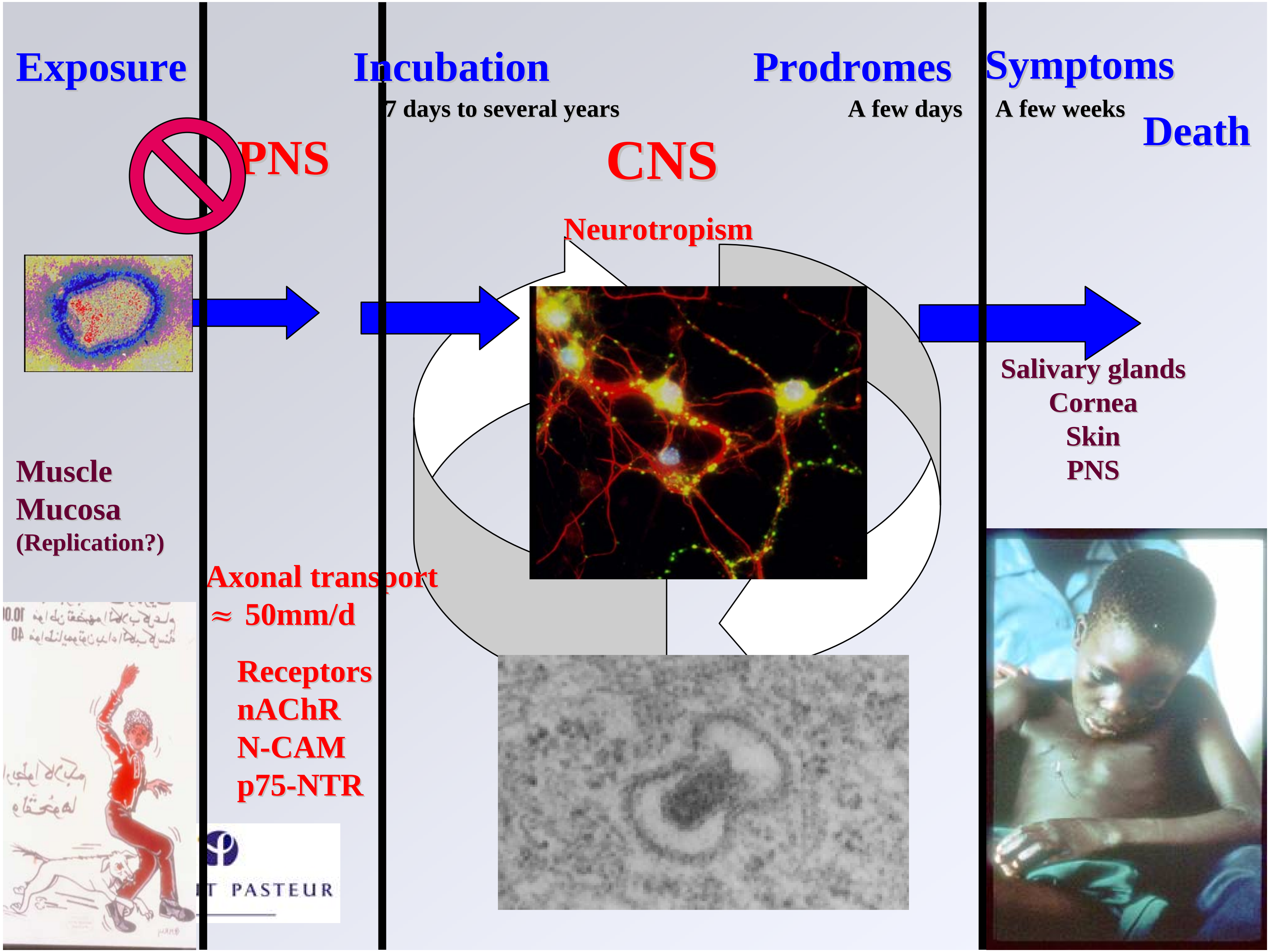
Exposure
Incubation
7 days to several years
Prodromes
A few days
Symptoms
A few weeks
Death
PNS
CNS
Neurotropism
Muscle
Mucosa
(Replication?)
Axonal transport
≈ 50mm/d
Receptors
nAChR
N-CAM
p75-NTR
Salivary glands
Cornea
Skin
PNS

Ignorance

- Of the disease
- Of the mode of transmission
- Of the exposure
  - Bat bite
- Of what to do in case of exposure

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No access to biologicals

Non available
Too expensive
Bad quality

Lack of biologicals

- Lack of RIG
  - Only 1% of PET associate RIG +vaccine (WHO, 1999)
  - Category III exposures : 73% (France 2004)
- Brain tissue vaccines/tissue culture vaccines : 40% of vaccines used were of Brain Tissue Origin in 1999
- Vaccines against new Lyssaviruses: African, European, Caucasian …or studies on efficacy of current biologicals on these viruses.

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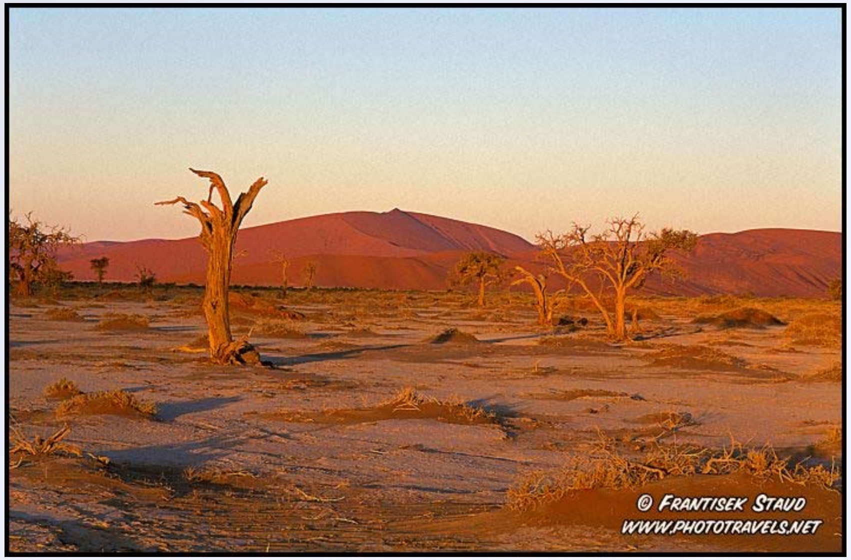
No access to medical cure

Too far

Too expensive in terms of travel, loss of working days…

Multiple medical facilities or centralized?



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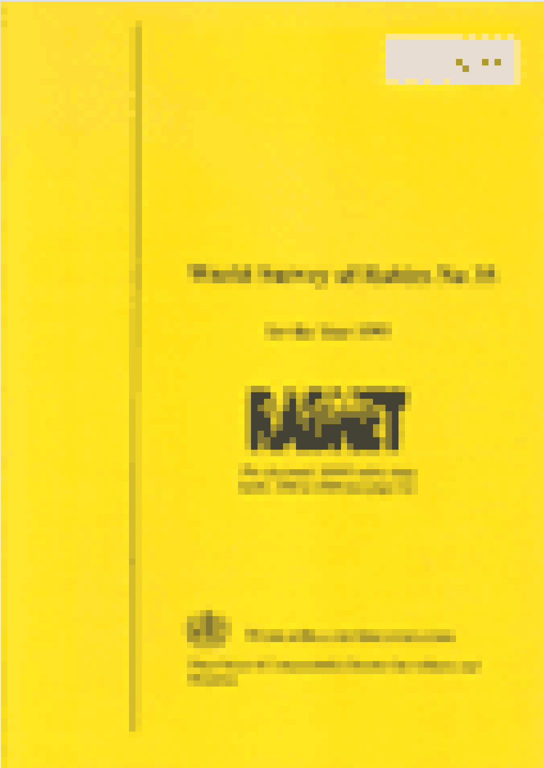
Ignorance of the disease

- Data collection
  - **Human rabies cases**
  - **Exposures: circumstances, population, animals…**
  - **Postexposure treatments : vaccine, serum, regimens…**



https://www.voozanoo.net/epiconcept/

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Treatment failures

- Delayed PET
- Inappropriate : guide-lines not followed
- Poor conservation of biologicals : unsustainable cold chain
- Lyssaviruses or rabies virus strains too distant from the vaccine/RIG strain

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New aspects of epidemiolgy

Human to human transmission

- **Mother to child**
- FEKADU M, ENDESHAWT, ALEMU W, *et al.* Possible human-to-human transmission of rabies in Ethiopia. Ethiop Med J 1996 ; 34 : 123-7.
- **Organ transplantation**
  - SRINAVASAN A, BURTON EC, KUEHNERT MJ, *et al*. Transmission of rabies virus from an organ donor to four transplant recipients. N Engl J Med 2005 ; 352 (11) : 1103-11.
  - JOHNSON N, BROOKES SM, FOOKS AR, *et al*. Review of human rabies cases in the UK and Germany. Vet Rec 2005 ;November 26 :715.

Prolonged survival

- WILLOUGHBY RE, TIEVES KS, HOFFMAN GM, *et al*. Survival after treatment of rabies with induction of coma. N Engl J Med 2005 ; 352 (24) : 2508-14.

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New aspects of epidemiology

- Importation of animals
- Travels
- Outbreaks of rabies due to vampire bats
- New vectors: bats, raccoon dogs, new pets…

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Our weapons against rabies challenges

- Vaccines and RIG safe and efficacious
- Decreasing costs
- Validated PET regimens
- Serological monitoring
- Technology transferts
- Pre-exposure vaccination…
- Physiopathology research
- Epidemiosurveillance
- Monoclonal antibodies
- Education
- Training
- Survival…

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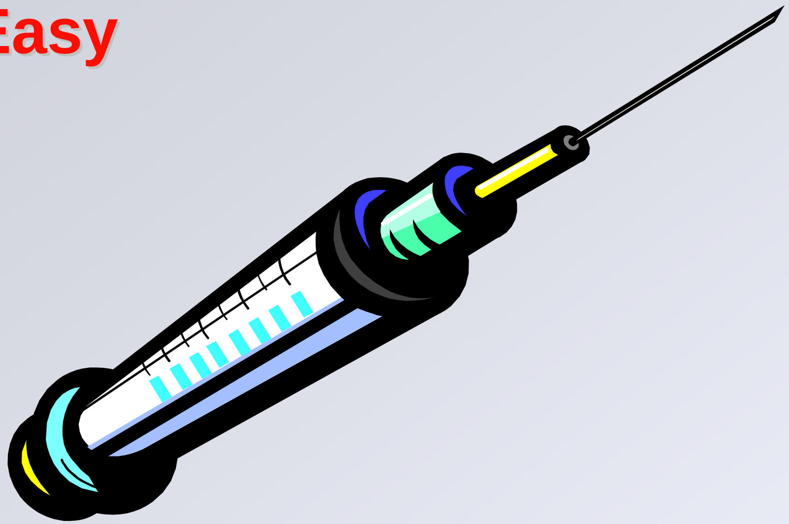
Pre-exposure vaccination

Is

Safe
Easy



- Laboratory workers
- Bat handlers
- Travelers
- Children in enzootic areas
- Animal handlers
- …

Should be

Available
Affordable

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Disease

- **Nowadays, we can only be efficient before the virus enters the CNS**
- **What of the survival case?**
- **New possibilities???**

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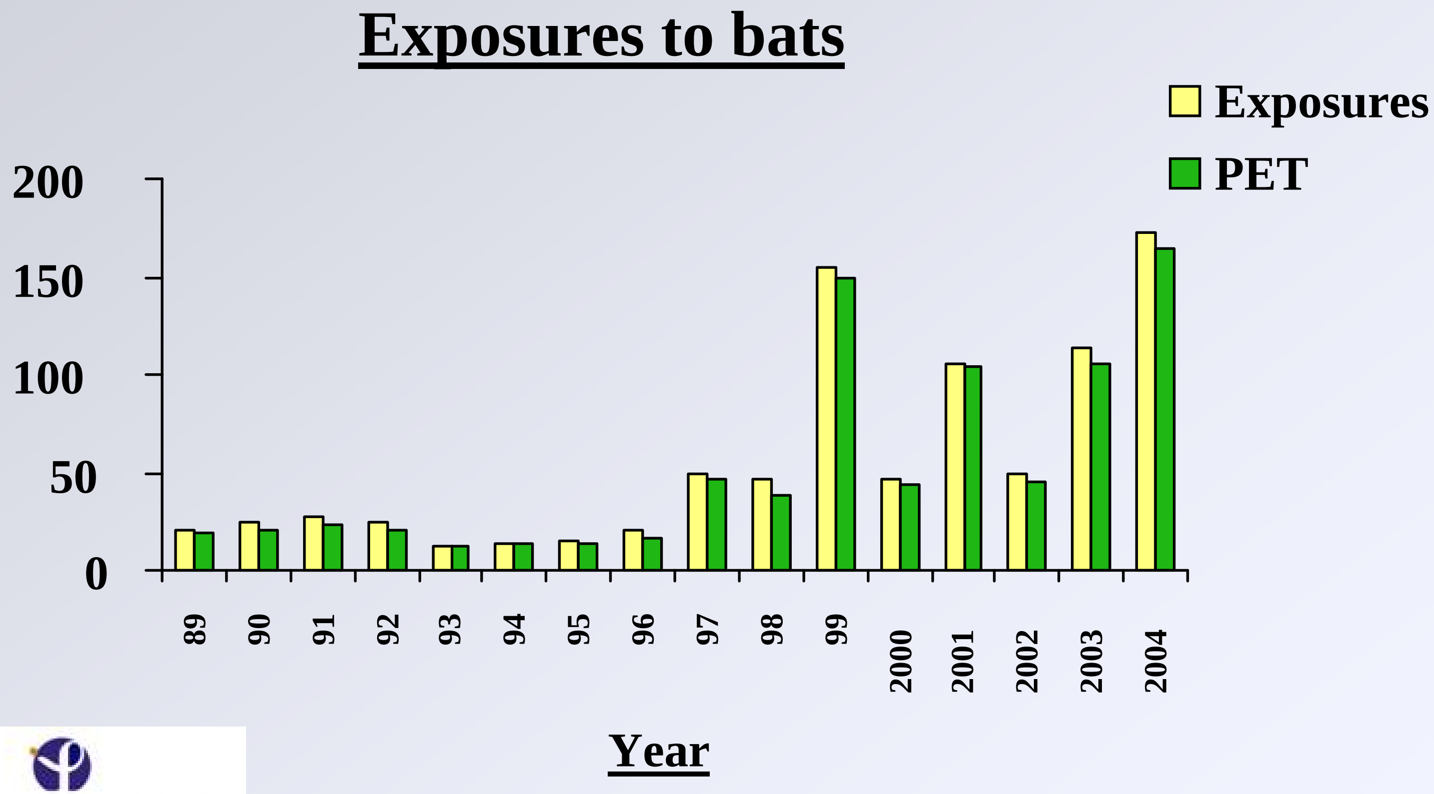
Bat exposures

93% of the patients exposed to bats are given PET



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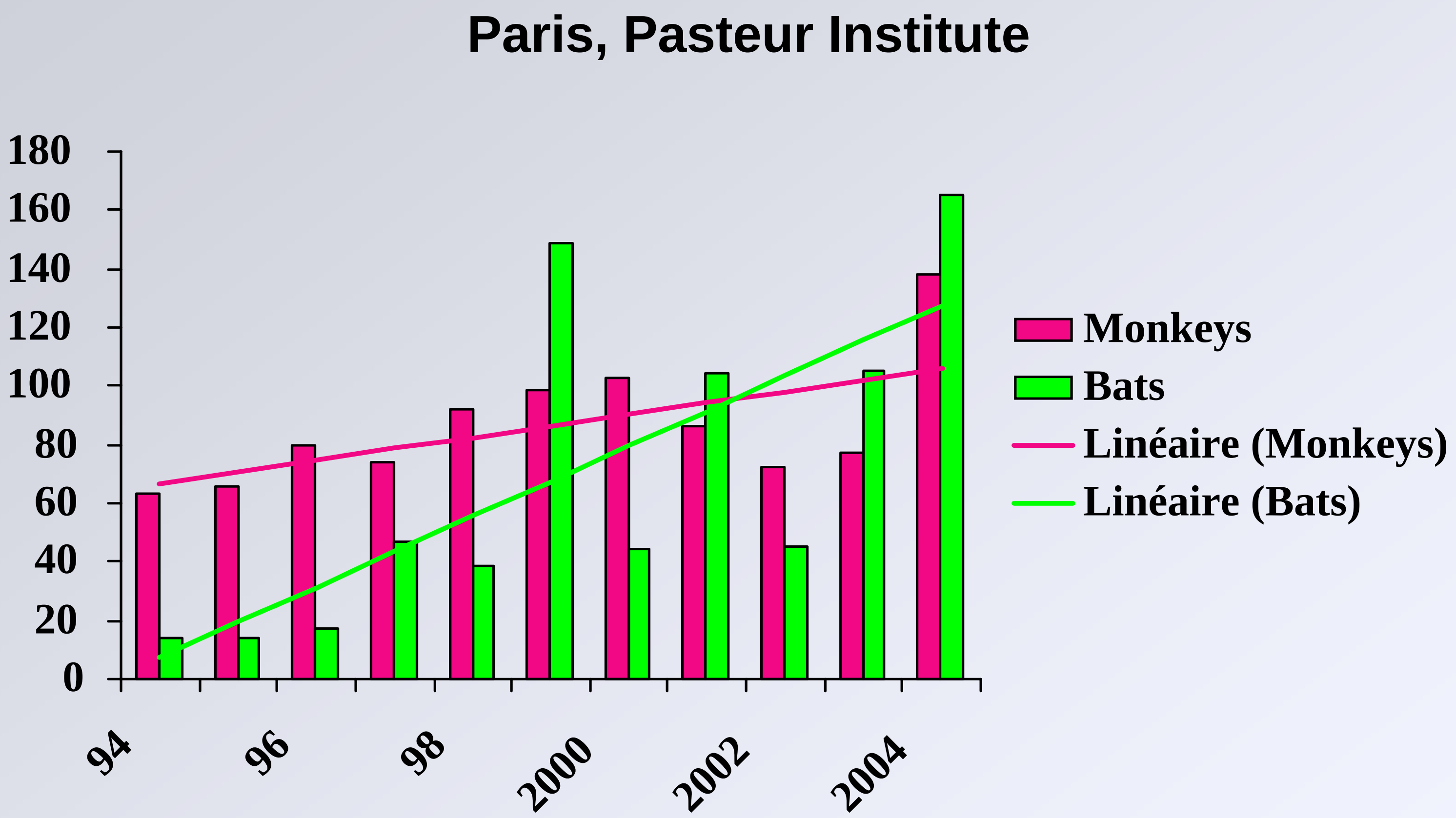
PET initiated after monkey and bat exposures



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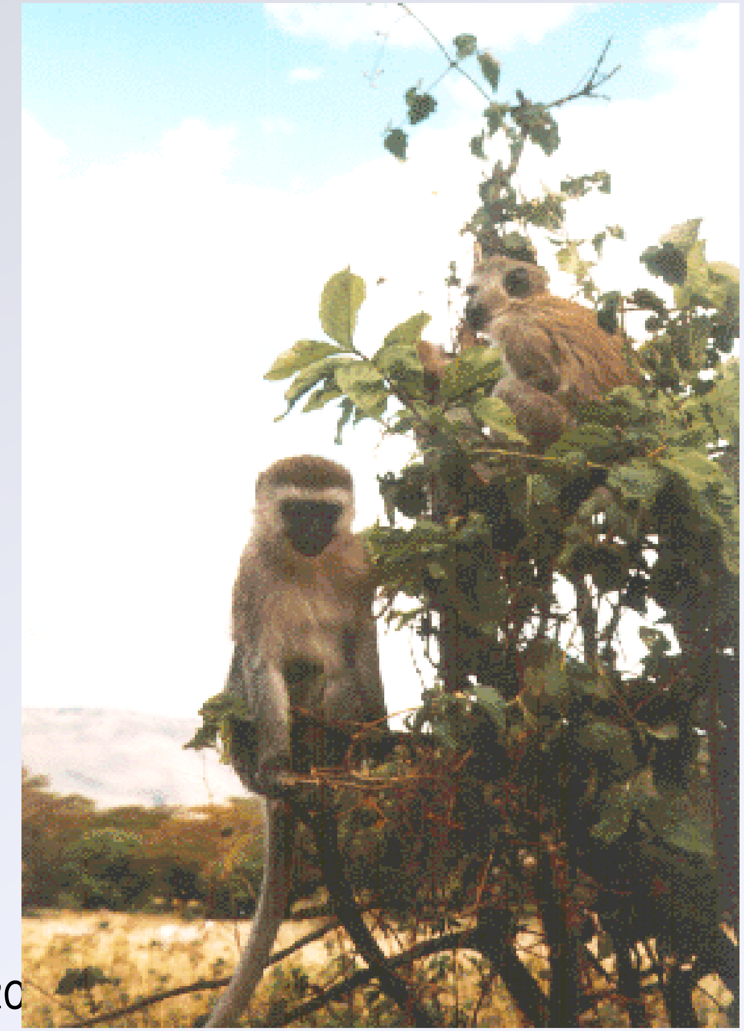
Monkeys

- **Monkey exposures :**
  - **France**
  - **Travel**



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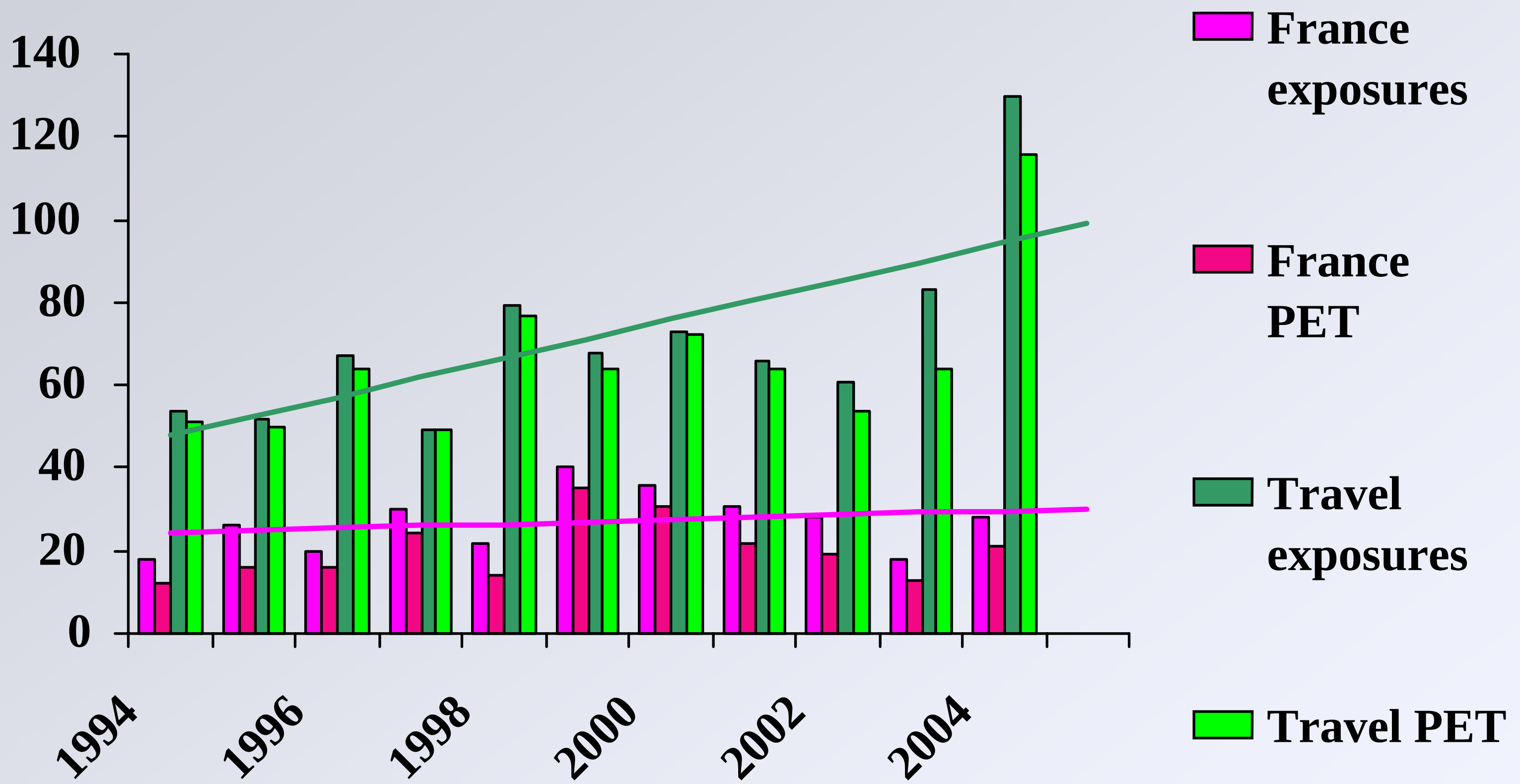
Monkey exposures and PET



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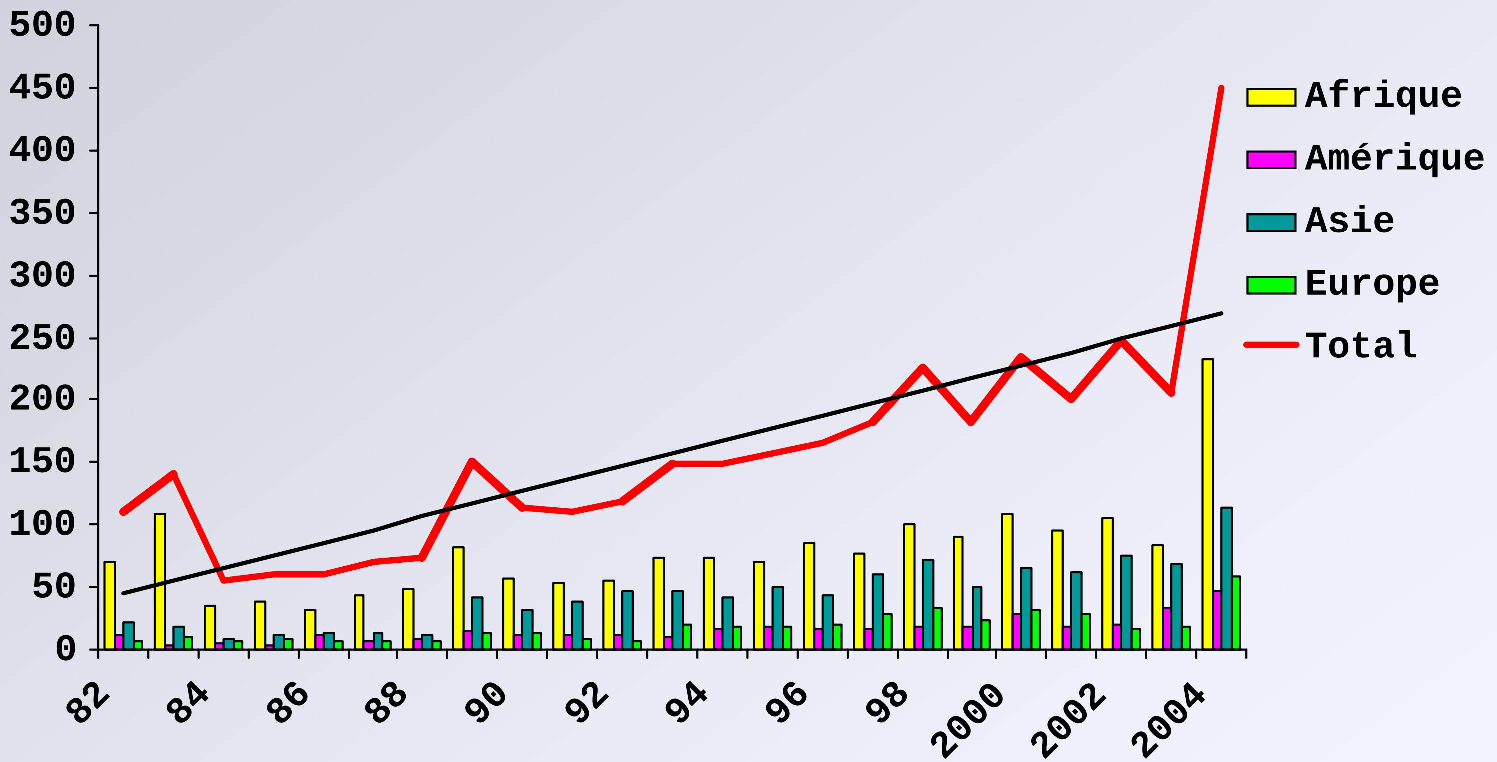
YR/SEARG/01-2006

PET in travelers
Institut Pasteur-1982-2004



Indirect effect of sensibilization due to the dogs Imported from Morocco?

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YR/SEARG/01-2006

Human Rabies
France 1970-2004

- 20 cases all imported
- 8 children, less than 5 years old
- 18 males, 2 women
- 3 treatment failures (Fuenzalida Vaccine used in Africa)
- 1 treatment failure : no RIG
- diagnosis : 9 ante mortem (AM), 11 *post mortem* (PM).
- 17 dogs, 1 cat, 1 UK, 1 corneal graft

- Gabon x 4
- Mali x 1
- Madagascar x 1
- Niger x 1
- Senegal x 1
- Algeria x 5
- Tunisia x 1
- Moroccox 2
- Egypt x 1 + corneal graft x 1
- India x 1
- Mexico x 1

Africa 18

Last case 2003

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The prospects of antivirals in rabies phophylaxis

Noël TORDO

INSTITUT PASTEUR

Rabies and antivirals

Advantages

- no therapeutics (vaccine, RIG are preventive)
- long incubation period (weeks to years: 2 months)

Disadvantages

- virus in the neuron (difficult to access)
- neglected disease (poor countries)

Rabies and antivirals, previous trials

(Reviewed in Jackson et al, 2003)

- α-interferon: (+) in monkey, (-) in human
  *Weinmann et al 1979 Infect Immun **24**:24-31Merigan et al 1984 Ann Neurol **16**:82-7*
  *Warrell et al, 1989 Br Med J **299**:830-3*
- ribavirin (purine analogs, AraC): (+) in vitro, (-) in mouse/fox/human
  *Bussereau et al 1983 Annales Virol (Inst Pasteur) **134**:127-34; 487-506*
  *1988 Acta Virol **32**:33-49*
- interferon & vidarabine: (+/-) in animals
  *Dolman & Charlton 1987 Can J Neurol Sci **14**:162-5*
- ketamine (antagonist NMDA receptor): (+) in vitro (+) rat stereotax. inj.
  *Lockhart et al 1992 Antimicrob. Agents Chemother. **36**, 1750-1755*
  *1991 Antiviral Chem Chemother **2**:9-15*
- heteropolyanions: (+/-) in fox
  *Pepin & Blancou 1985 Archiv. Virol **83**: 327-329.*
- corticosteriods: (-) in mouse
  *Enright et al 1970 Can J Microbiol **16**:667-75*

Rabies therapy: a case report (2005)
(Willoughby et al, N Engl J Med. 2005, 352: 2508-14)

- Scholar-athlete aged 15
- Future veterinarian
- Picked up downed bat
- Laceration on L index finger

No post-exposure prophylaxis

- One month incubation

Crisis management (2-3 hours): Internet; Key article

ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, Aug. 1992, p. 1750–1755
0066-4804/92/081750-06$02.00/0
Vol. 36, No. 8

Inhibition of Rabies Virus Transcription in Rat Cortical Neurons with the Dissociative Anesthetic Ketamine

BRIAN PAUL LOCKHART,† NOEL TORDO, AND HENRI TSIANG*
Rabies Unit, Pasteur Institute, 25, Rue du Dr. Roux, 75724 Paris, Cedex 15, France

Received 15 October 1991/Accepted 5 May 1992

In a previous study (B. P. Lockhart, H. Tsiang, P. E. Ceccaldi, and S. Guillemer, Antiviral Chem. Chemother. 2:9–15, 1991), we demonstrated an antiviral effect of the general anesthetic ketamine for rabies virus in neuronal cultures and in rat brain. This report describes an attempt to determine at what level ketamine acts on the rabies virus cycle in rat cortical neuron cultures. Immunofluorescence and [^{35}S]methionine labelling of infected neurons showed that ketamine (1 to 1.5 mM) inhibited viral nucleoprotein and glycoprotein syntheses. Northern (RNA) blots of total RNA from drug-treated neurons, hybridized with ^{32}P-labelled oligonucleotide probes for rabies virus nucleoprotein, matrix protein, and glycoprotein genes, showed a marked reduction (5- to 11-fold) in the levels of rabies virus mRNAs, relative to those in untreated neurons. No significant change in the levels of cellular β-actin mRNA were detected in ketamine-treated cells. A similar antiviral effect was observed with MK-801; however, no inhibition of rabies virus synthesis was observed with the general anesthetic chloral hydrate. The antiviral effect was not complete; a time-dependent recovery of viral transcription and rabies virus protein synthesis was observed, but no infectious virus was released into the culture supernatant. The lack of any modification of cellular protein or mRNA synthesis by ketamine suggests an antiviral mechanism acting at the level of rabies virus genome transcription.

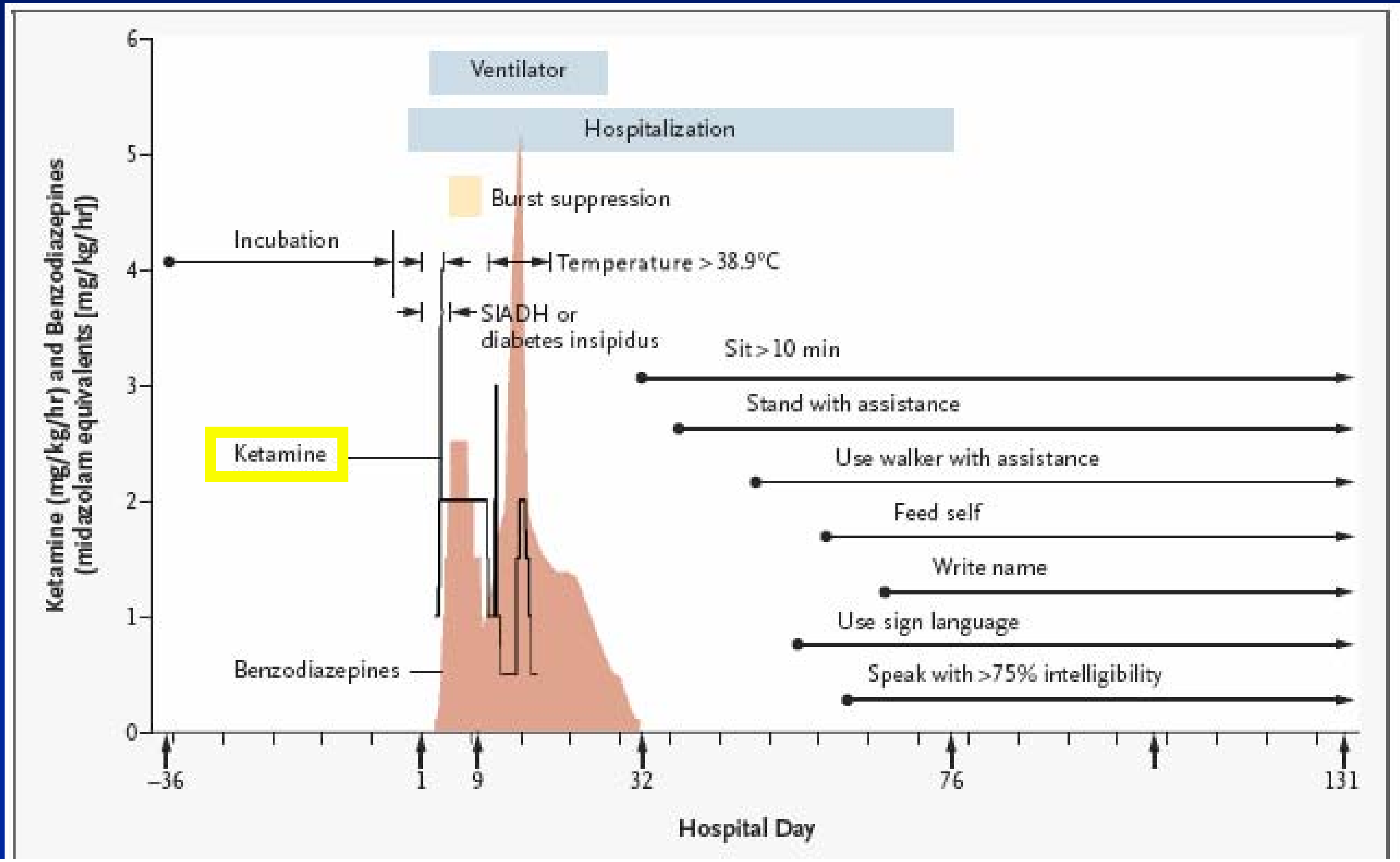
Effect was virus-specific!

Survival after treatment of rabies with induction of coma

(Willoughby et al. 2005, N. Eng. J Med 352:2508-14)

Amantadin

Ribavirin



Conclusion:

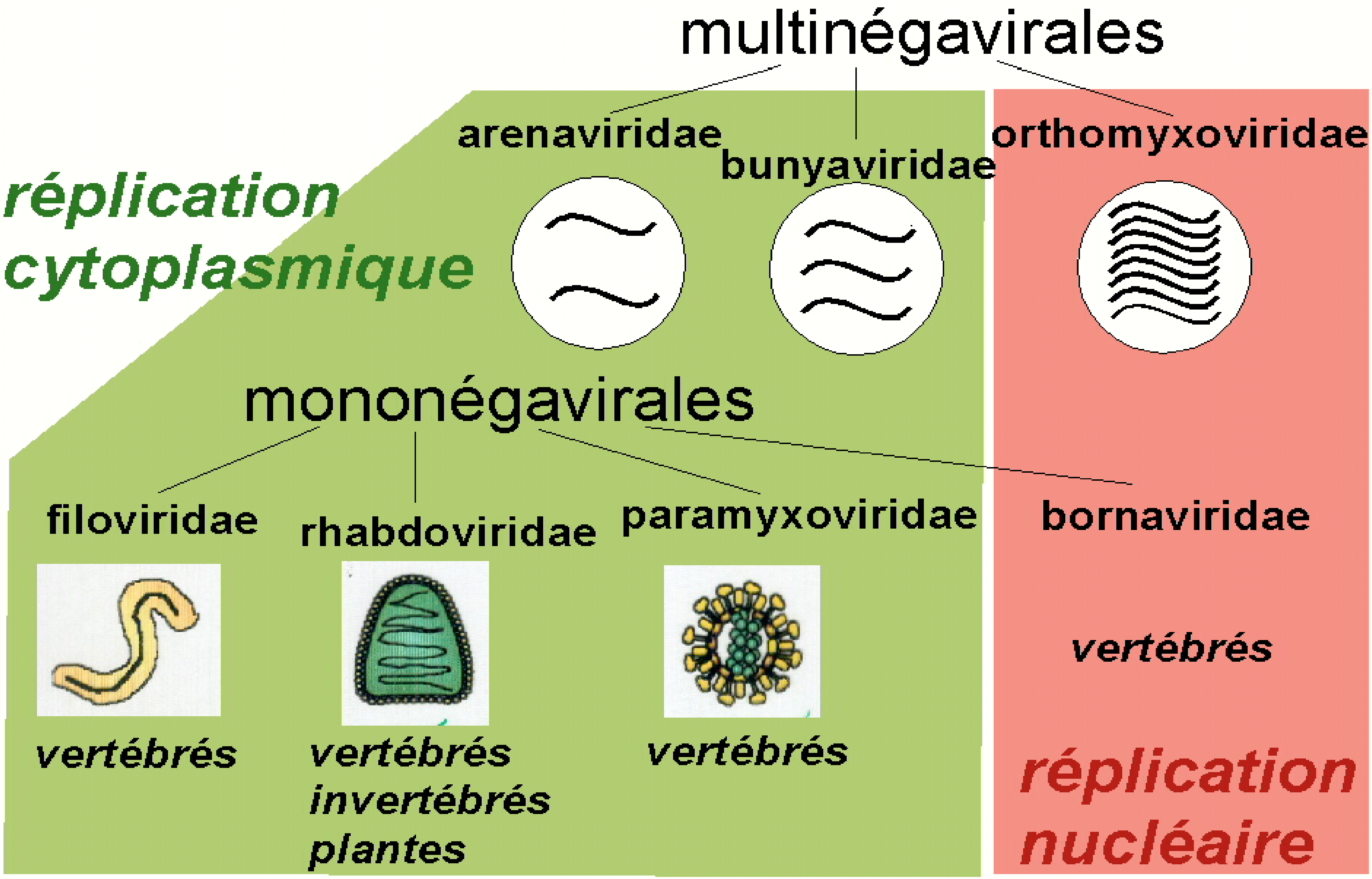
An antiviral approach against rabies is possible…

…but commercially difficult (neglected disease)

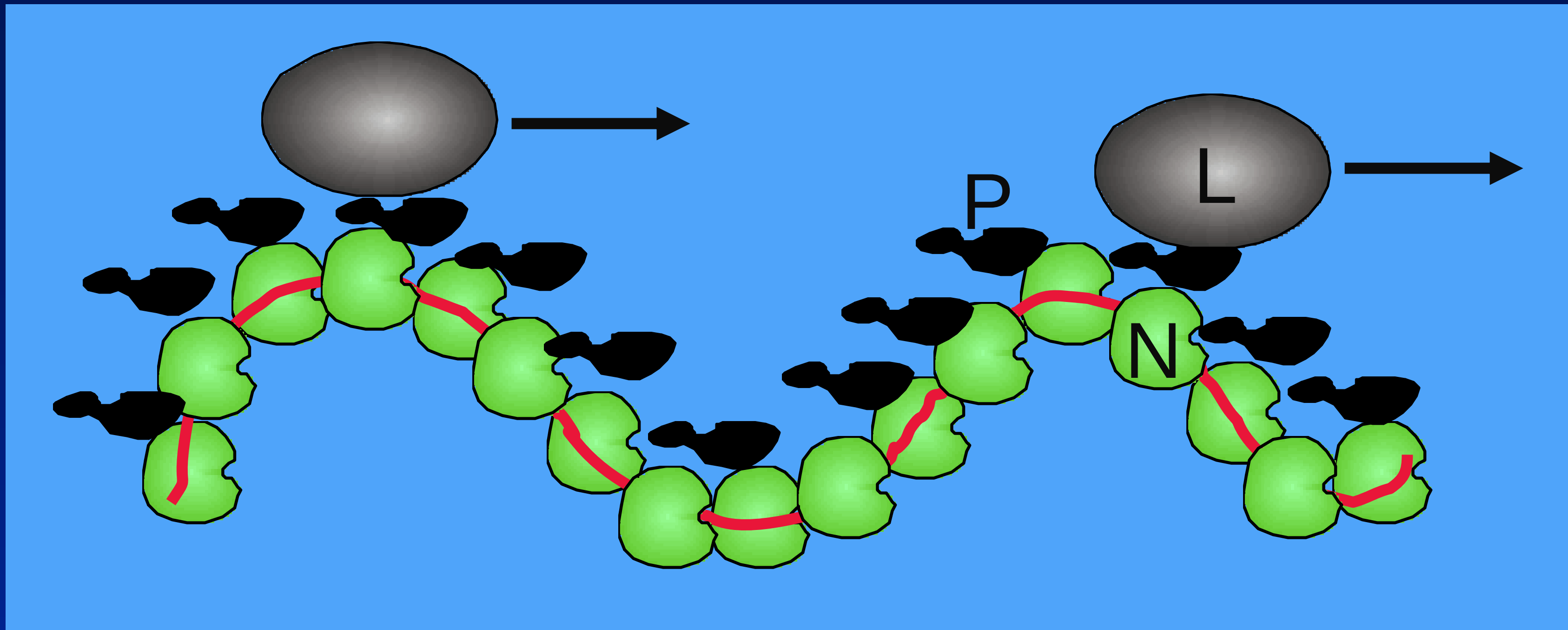
Concept: “Unit Antiviral Strategies”

Development of large spectrum antivirals targeting commom targets of negative strand RNA viruses

virus à ARN négatifs



transcription/replication complex (RNP)



Template

- RNA genome
  + nucleoprotein N

Enzymes

- RNA polymerase L
- phosphoprotein P (cofactor)

Unit Antiviral Strategies:
inhibiting the transcription/replication complex of negative strand RNA viruses (Mononegavirales)

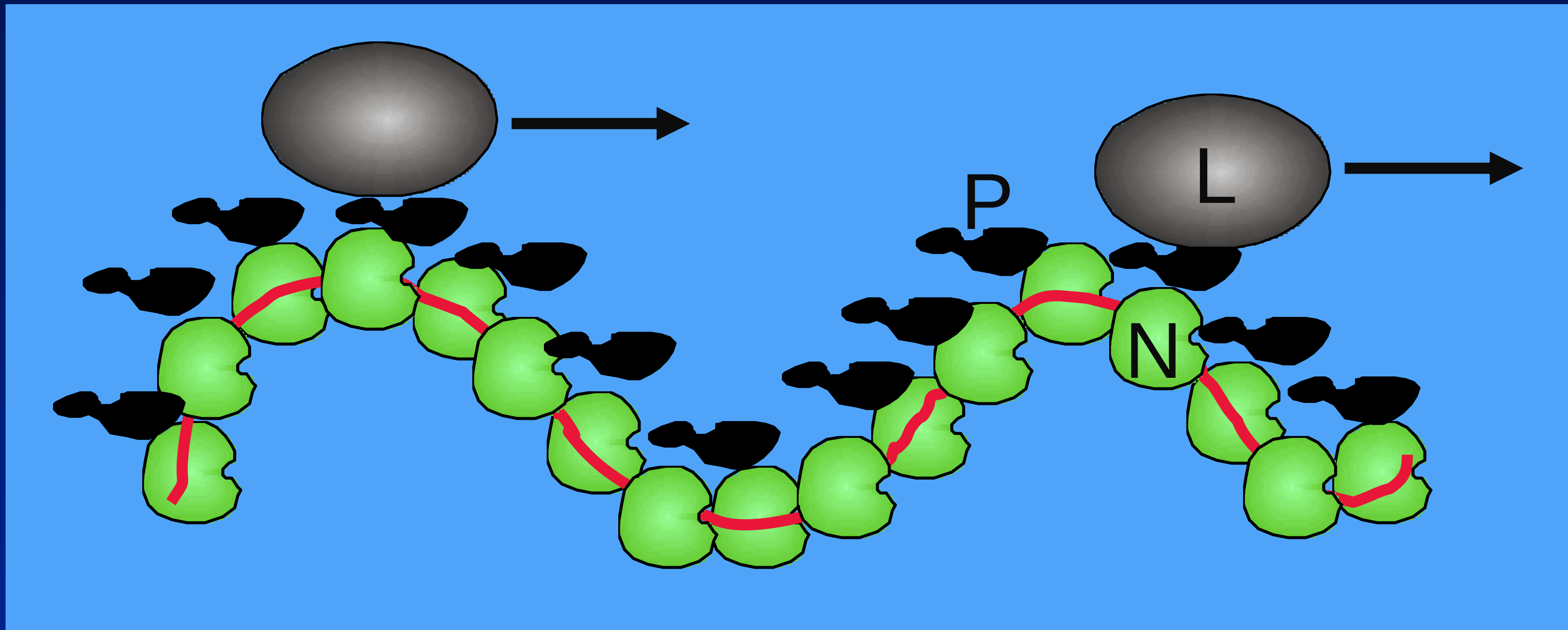
Cognitive approach →

- structure/function of N, P, L proteins
- interactome with host factors
  2-hybrid, coIP, cristal structure, site directed mutagenesis

Random approach →

- Highthrouput screening of molecule (peptide) libraries
  2-hybrid, phage display, ex vivo screening

Target: P phosphoprotein



Template

- RNA genome
  + nucleoprotein N

Enzymes

- RNA polymerase L
- phosphoprotein P (cofactor)

combinatory library of auto-contraint peptides in *S. cerevisiae (2-hybrid)*

Toxins: conotoxins (mollusques); defensins (mammals)

constraint through disulfures bridges (cysteines)

Cys library (26 a.a) $1x10^7$ independent peptides

C-2x-C-5x-C-6x-C-5x-C-2x-C- Gal4 AD

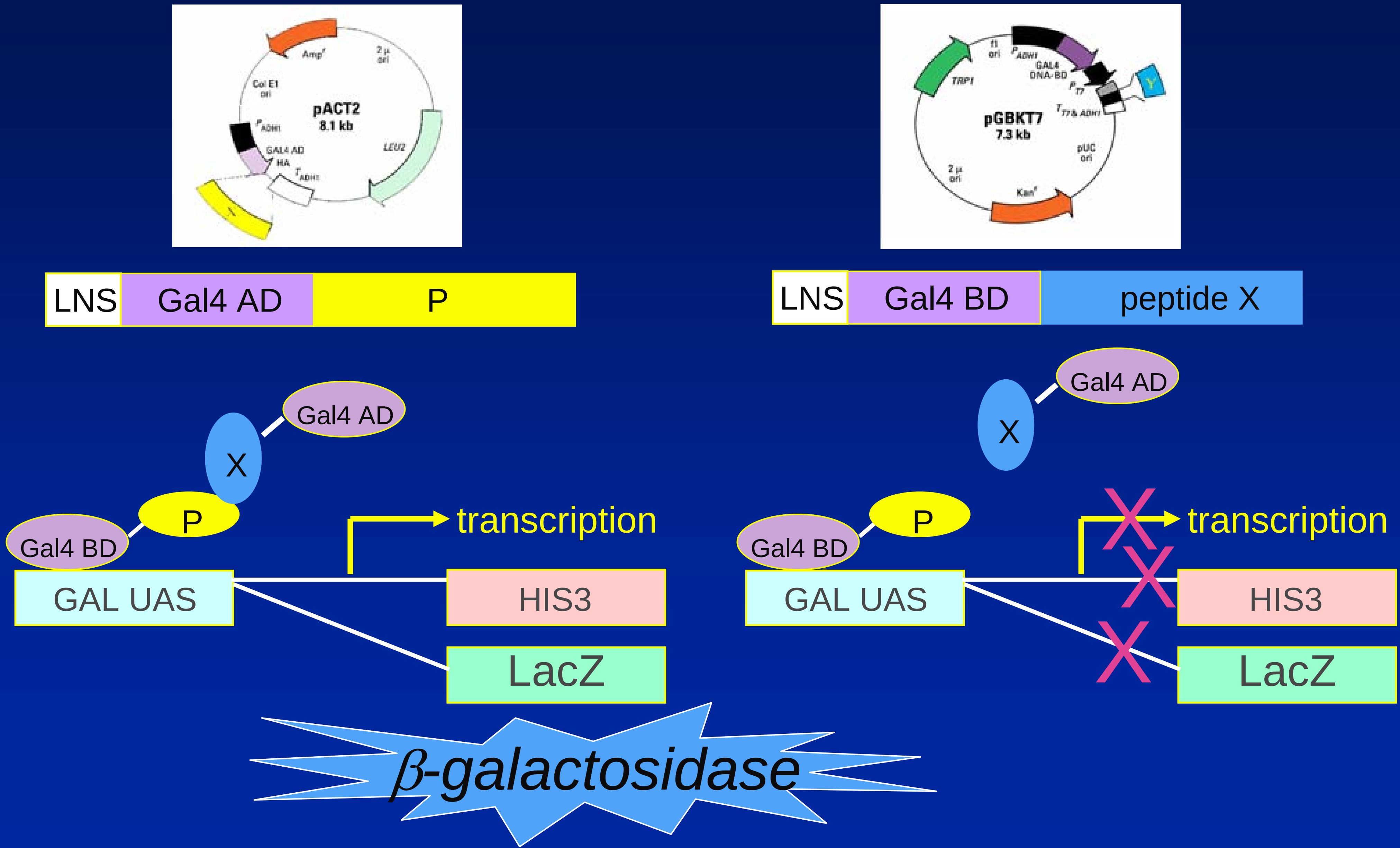
Antimicrobial properties: lebocines, apidaecines (insects)

conformational constraint through prolines (turns)

Pro library (29 a.a) $3x10^7$ independent peptides

PP-5x-P-5x-PPP-5x-P-5x-PP - Gal4 AD

Yeast two-hybrid screening (*S. Cerevisiae*)



Selection step 1: screening in yeast 2-hybrid

peptide libraries

2-hybrid screening

P-PV partners

46.5% identity

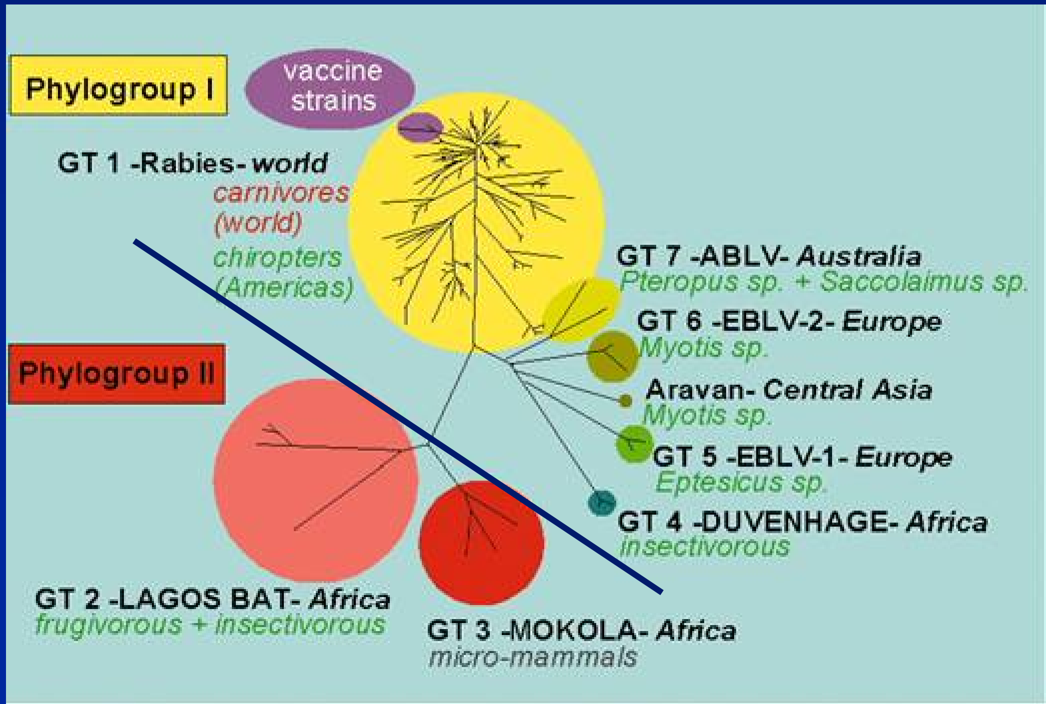
P-Mok partners

in silico analysis (phylogeny, groups)

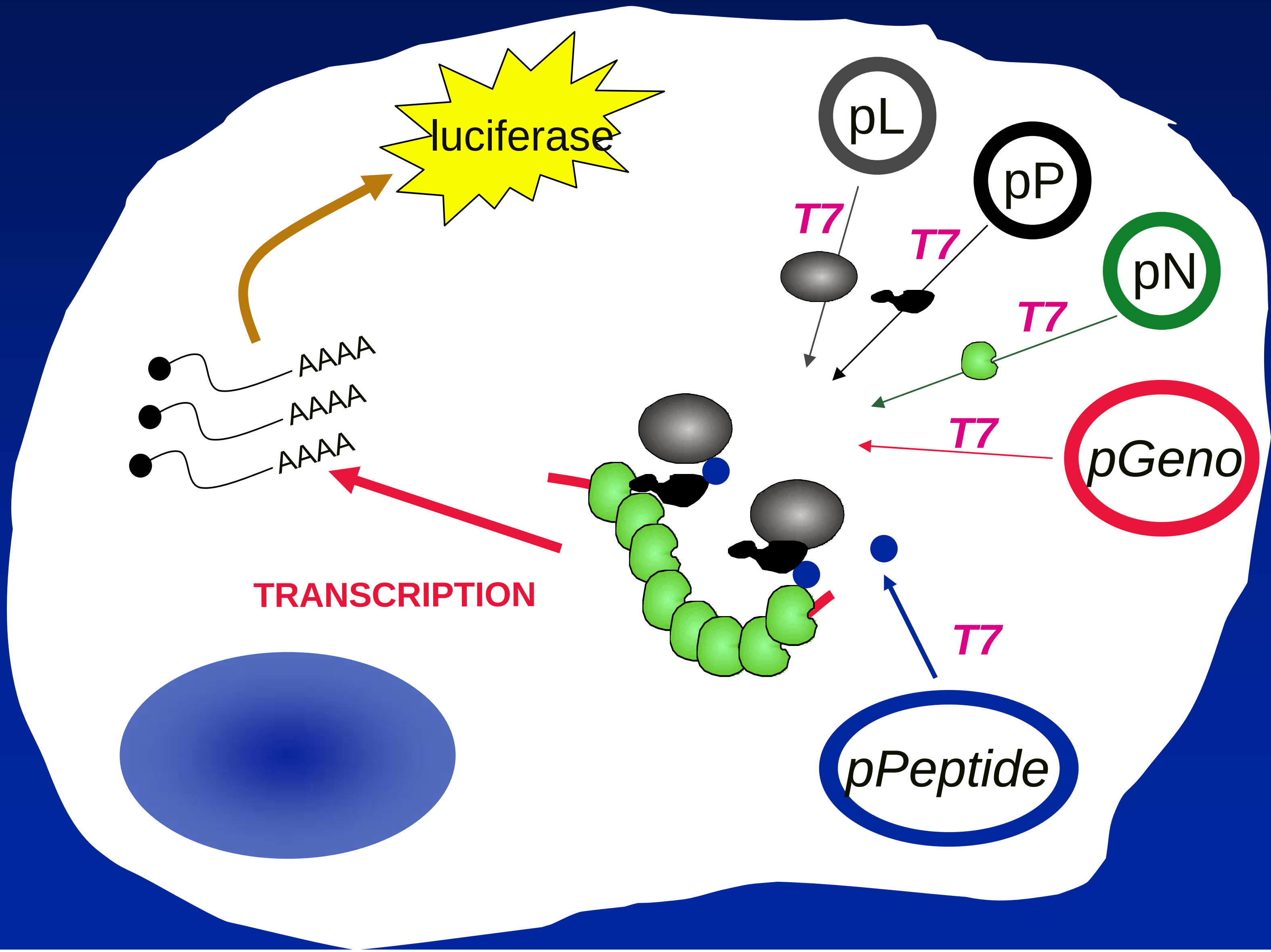
29

8,55 10^7

755

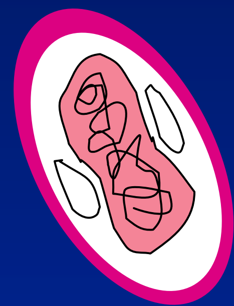


Selection step 2: inhibition of RNP transcription *(mini-replicons)*



1. Infection

vaccinia virus / T7 polymerase



2. Transfection

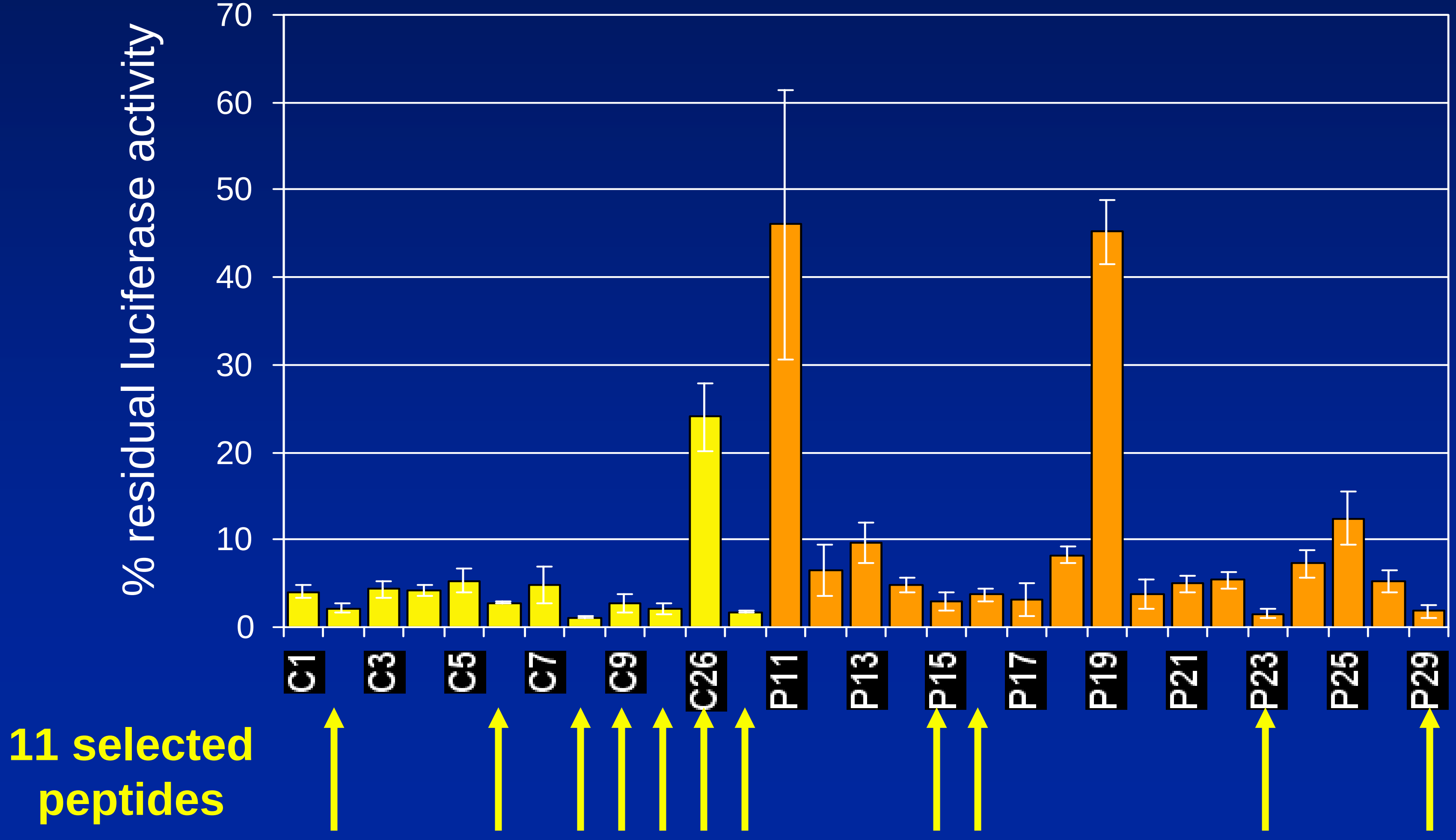
plasmides

- N, P, L proteins
- mini(geno)me

reporter: luciferase

Selection step 2: inhibition of RNP transcription
(reverse genetics)

12 Cys peptides + 17 Pro peptides



Selection step 2: inhibition of RNP transcription

peptide libraries — *8,55 10^7*

2-hybrid screening

P-PV partners | P-Mok partners — *755*

in silico analysis (phylogeny, groups) — *29*

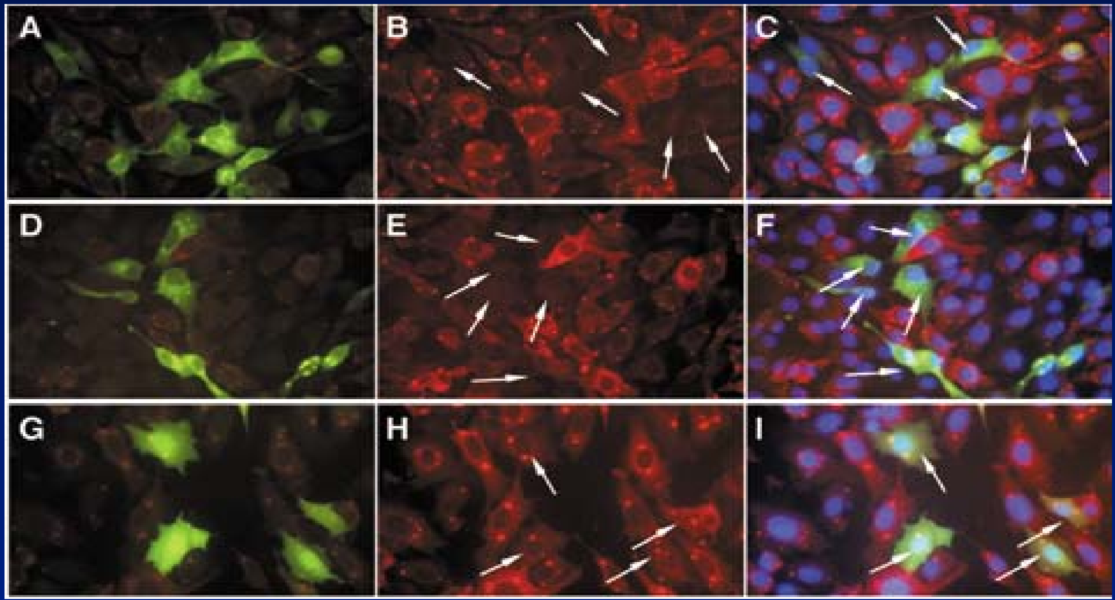
inhibition of RNP transcription (minireplicons) — ***11***

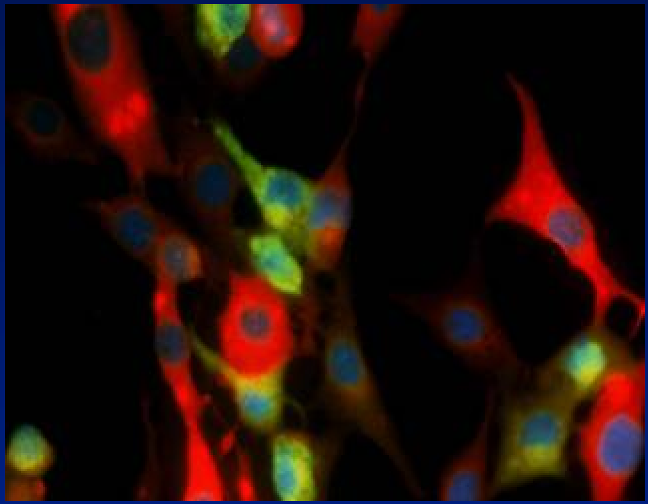
Selection step 4: inhibition of Neuro-2A cell infection

C2-GFP

P16-GFP

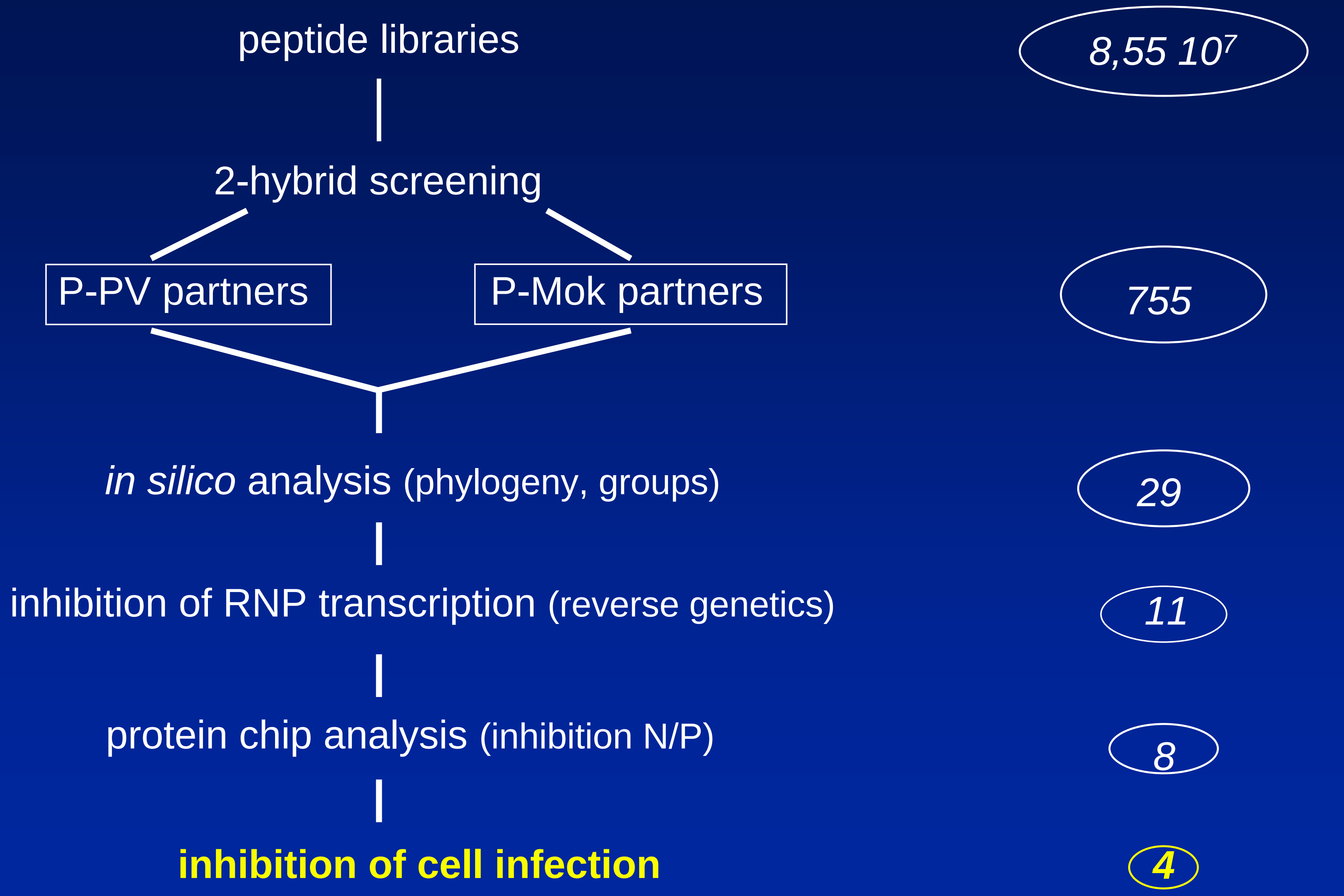
GFP





Peptide	% inhibition infection	ratio P/N peaks	% luciferase activity
No	0	1,05	100
C2	**89**	**1,70**	**2,1**
C6	**83**	**1,64**	**2,8**
C8	**78**	**1,52**	**1,1**
C10	57	2,53	2,1
C26	5	1,09	24,0
C27	22	1,00	1,7
P16	**71**	**2,49**	**3,7**
P29	Toxic	1,60	1,8

Selection step 4: inhibition of cell infection



Conclusions

We are developing rational approaches to design peptides interfering with and inhibiting the replication complex (RNP)

- 2-hybrid: P protein (rabies and Mokola) -> *Lyssaviruses*

Successive screenings for:

- protein-protein interactions (2-hybrid)
- functional inhibition (minigenome expression, infection)

➢ 4 candidates inhibitory peptides against P (Lyssaviruses)

Réal et al, 2004, J. Virol, ***78*** *: 7410-7*

Perspectives

Improvement of the inhibitory effect of selected peptides

- synthesis in fusion with sequences favouring transmembrane passage for cell delivery;
  (cell penetrating peptide: penetratin, TAT, Antenapedia…)
- Optimisation of size/affinity/stability
  (D-peptides, peptido-mimetism =>« drug-design», …)

Screening peptide libraries:

- on other targets of the replication complex (N, L)
- on other negative strand RNA viruses (segmented genomes)
- for antiviral effect *in vivo*

Institut Pasteur, Paris

Unit Antiviral Strategies

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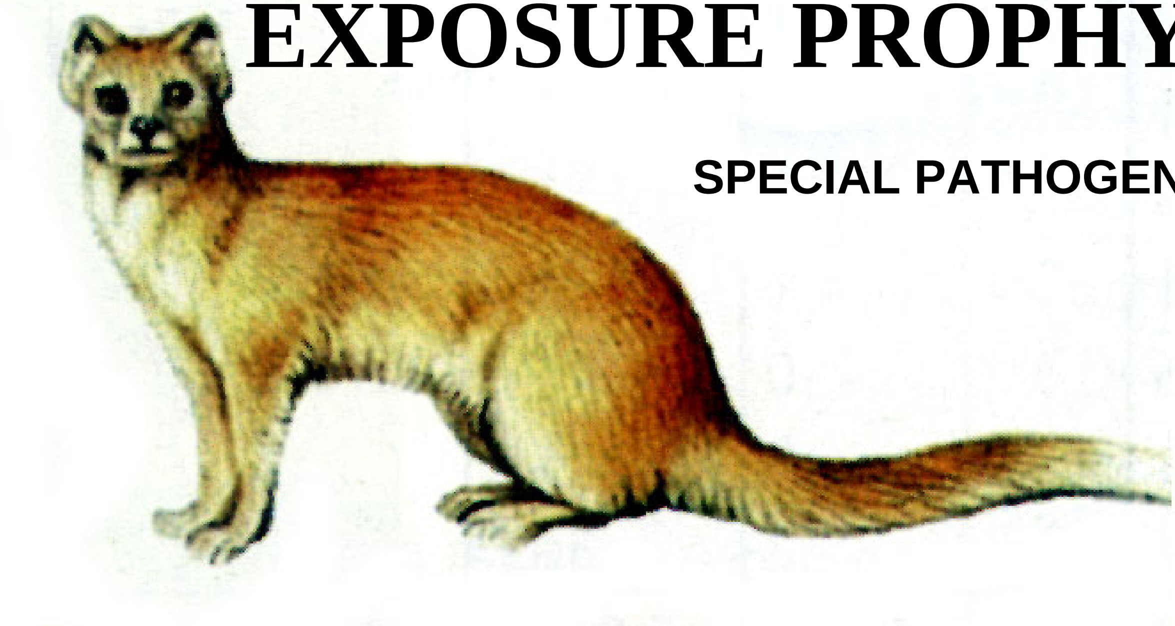
Jean-Francois ELEOUET

FAILED RABIES POST-EXPOSURE PROPHYLAXIS?



SPECIAL PATHOGENS UNIT, NICD

NICD

19 year old, previously healthy soldier

- **Category 3** exposure on finger -rabid yellow mongoose
- **13 hours :** wound toilet, human diploid cell vaccine IMI into gluteus, RIG (20IU/kg) 1ml into wound, rest into deltoid
- **Vaccine course:** days 3, 7, 14
- **Day 21: onset of rabies- like illness,** neg rabies Ag on saliva and CSF, rabies IgM 1:8 in serum (IFA)
- **Day 37: died,** serum rabies IgM 1:256 (IFA), rabies virus isolated from brain
- Shill NEJM

Reasons for failure of PEP?

- Vaccine and RIG potency adequate
- Cold chain maintained
- **Possible causes of failure:** administration of vaccine into the gluteus muscle, inadequate local wound administration of RIG, underlying immune deficiency?
- Shill NEJM

57 year old, previously healthy man

- **Category 3** exposure on hand -water mongoose
- **3.5 hours:** cell culture **vaccine** IMI into deltoid
- **5 hours: RIG** (20IU/kg) into wounds +wound cleaning with Eusol
- **Vaccine course:** days 3, 7, 14
- **Day 24: onset of rabies- like illness,** pos rabies PCR (saliva), rabies IgM 1:256, IgG 1:512 in serum (IFA)
- **Day 30: died**, rabies PCR and FA pos on brain, culture negative

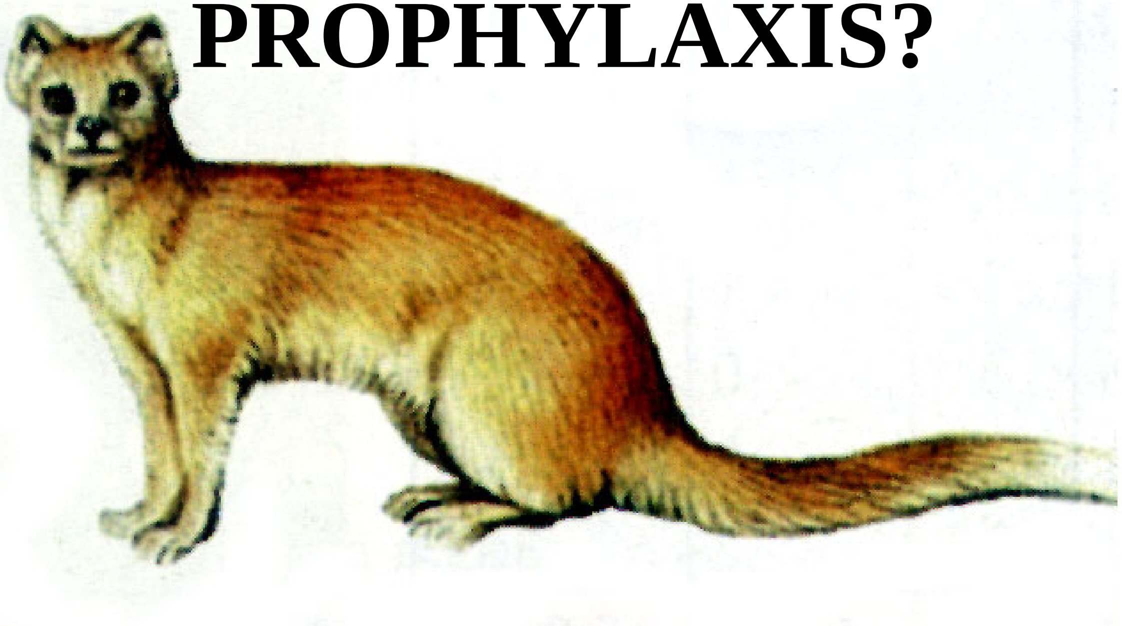
NICD communique 2004

Reasons for failure of PEP?

- Correct PEP procedure
- Vaccine and RIG potency investigation
- Cold chain maintained
- No immune problems in patient
- **Possible causes of failure:** inadequate wound cleaning, penetrating wound with direct introduction of virus into nerve tissue?
-

NICD communique 2004

FAILED POST-EXPOSURE PROPHYLAXIS?



Special Pathogens Unit –NICD, Sandringham-Johannesburg – the referral center for human rabies in southern Africa

- On average the SPU confirms 7-10 cases annually and the majority of these cases are generally believed to be from contact with rabid dogs in the KwaZulu Natal Province
- In many instances patients in SA receive no PEP, partial or inadequate PEP and in some instances no history is available to determine if appropriate vaccination was provided

Rabies – uniformly fatal viral infection generally transmitted by the bite of infected animals

- Despite the availability of effective vaccines annual human deaths ~ 60, 000 worldwide; ~ 98% of these preventable fatalities in Africa, Asia and Latin America - animal control, vaccination programs and effective human post-exposure prophylaxis are either not widely available, or not effectively applied

- Rabies virus strains circulating in specific species undergo genetic adaptation and evolve into distinct biotypes that differ in antigenicity and pathogenicity

- Two biotypes of rabies virus in southern Africa
  - canid viruses (*Canis familiaris, C. mesomelas, C. adustus, Otocyon megalotis)*
  - “*viverrid viruses”* (historical term) currently postulated to be named “mongoose” biotype or mongoose rabies virus (*Cynictis penicillata, Galerella sanguinea*)

Mongoose biotype

- There is a considerable antigenic and genetic diversity within isolates of mongoose biotype in comparison to the isolates of canid biotype which is closely related to the European wild (WR56) or /vaccine rabies strains (PV/ERA).
- Based on pseudogene nucleotide sequence 3 canid isolates (5/91 - jackal; 421/92 – dog; 127/91 bat-eared fox) were showed to belong to mongoose rabies isolates group suggesting that biotypes may jump species boundaries
- In South Africa, historical records show that mongoose rabies may have been described since the early 1800s, long before the introduction of canine rabies
- First cases of confirmed rabies related to the bite by a yellow mongoose were reported in 1928 in two children in Wolmaransstad district in the NWP.

Is mongoose rabies in humans an important health issue?

There have been two recorded instances in which patients were bitten by rabid mongoose and despite receiving PEP they contracted the disease and died - apparent vaccine failure?

The mongoose biotype belongs the same rabies ("dog") virus genotype

The vaccines strains are usually of the dog "cosmopolitan" type

The vaccines are said to protect reasonable well against all of the Group 1 lyssaviruses, that is, all the serogroup 1 rabies, EBL1, EBL2, Duvenhage and ABLV) … and would be expected to offer very good protection against the serogroup 1 viruses

Is then the "apparent" vaccine failure due to antigenic heterogeneity?

Would be sequence analysis, particularly of the neutralizing epitope sites of the glycoprotein useful to address it?

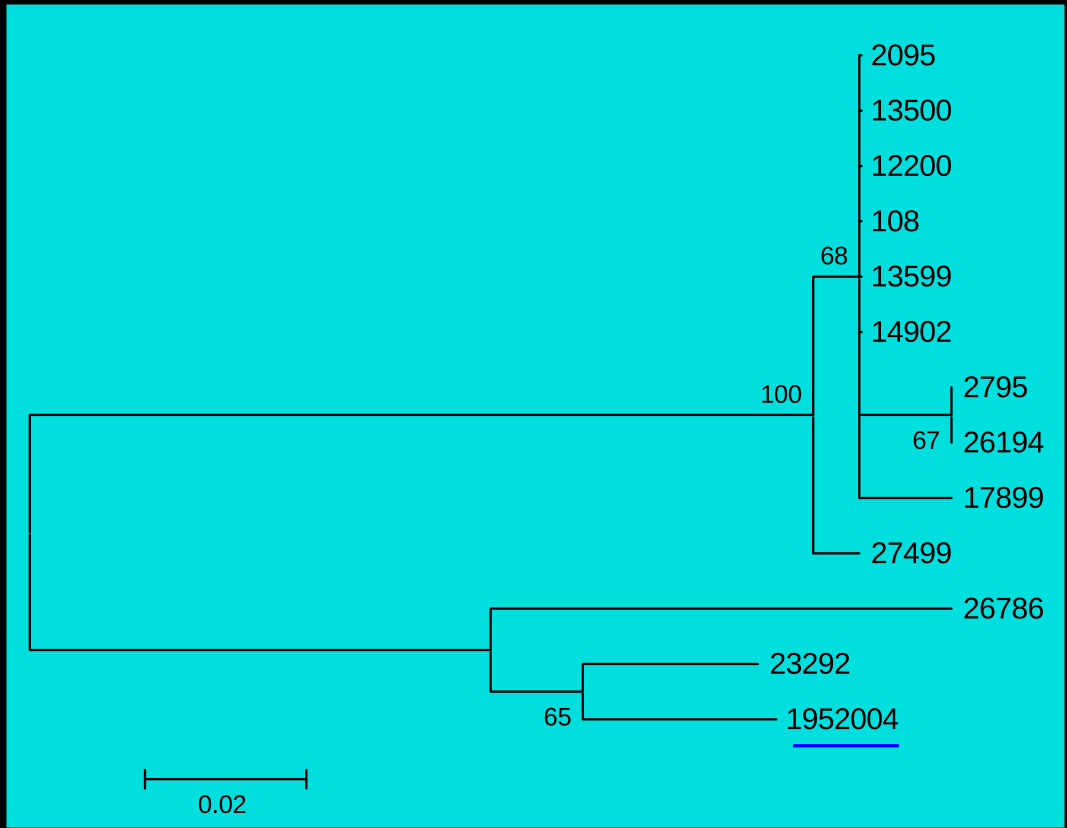
What about other factors, for example, a very rapid incubation period (less than 10 days) often caused by severe bites to the head or hands ….or marginal vaccine potency (are the batches available for potency testing?)

..… these incidences have prompted us to investigate the efficacy of rabies vaccine against mongoose biotype

Methods

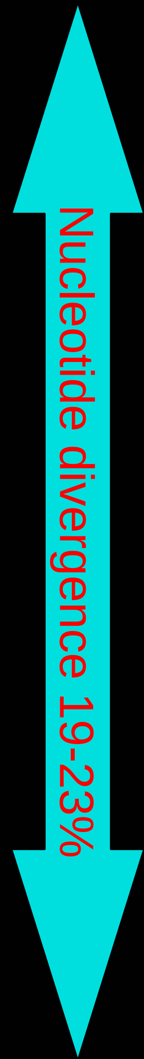
- Characterization of rabies isolates to confirm the biotype will be performed using partial nucleotide sequencing
- The potency of rabies HDCV against viverrid isolates will be determined using the potency test in mouse model

Phylogenetic tree illustrating genetic distance within the South African isolates of rabies isolates recovered from patients with history of dog (A) and mongoose (B) bite



A - rabies isolates from patients with history of dog bite

B - rabies isolates from patients with history of mongoose bite



+PCR from saliva and brain but live virus could not be recovered

Case 195/2004 (Standerton, Mpumalanga) related to bite by *Atilax paludinosus; is it the same strain as 878/92 (Harrismith); 668/92 (Albert); 610/96 (Somerset East); 113/91 (Beaufort West)*

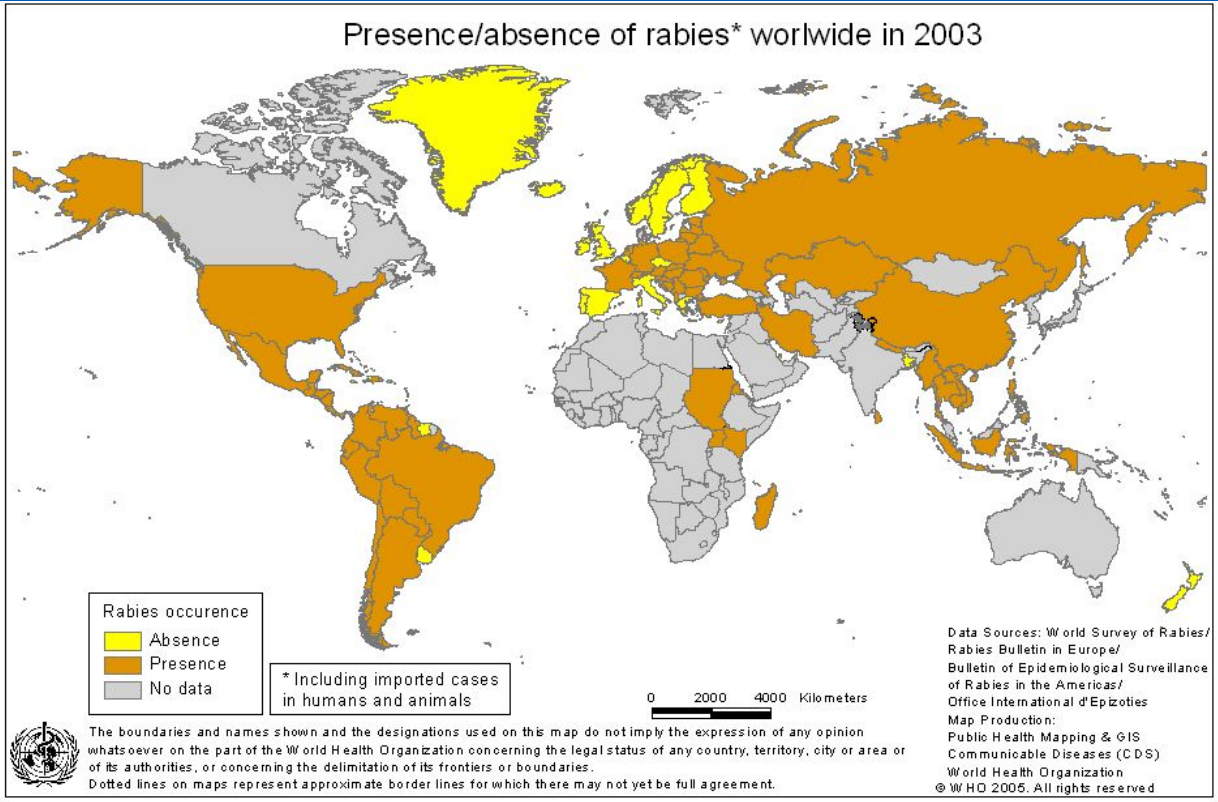
Development of a murine rabies monoclonal antibody cocktail for PEP

T. Müller, C. Rupprecht, A. Wandeler,
H. Ertl, N. Tordo, T. Fooks,
B. Dietzschold, F. X. Meslin, M.P. Kieny

FLI, Wusterhausen, Germany
CDC, Altanta, USA
ADRI, Ottawa, Canada
Wistar Institute, Philadelphia, USA
VLA, Weybridge, UK
Pasteur Institute, Paris, France
TJU, Philadelphia, USA
WHO, Geneva, Switzerland

FRIEDRICH-LOEFFLER-INSTITUT
FLI
Bundesforschungsinstitut für Tiergesundheit
Federal Research Institute for Animal Health

Rabies in the world



WHO guidelines for PEP

Category	Type of contact with a suspect or confirmed rabid animal	Type of exposure	Recommended PEP
I	Touching or feeding of animals Licks on intact skin	None	None, if reliable case history is available
II	Nibbling of uncovered skin Minor scratches or abrasions without bleeding	Minor	Administer vaccine immediately
III	Single or multiple transdermal bites or scratches, licks on broken skin Contamination of mucous membrane with saliva (i.e. licks) Exposures to bats	Severe	Administer rabies immunoglobulin and vaccine immediately

Rabies immunoglobulins

Classes of rabies biologicals

- human rabies immunoglobulin (HRIG)
- equine rabies immunoglobulins (ERIG)
- highly purified F(ab´)2 products produced from ERIG.

WHO recommendations for production and control
WHO guide for PEP – precautions for use

Rabies immunoglobulins

Developing countries

- In areas with endemic dog rabies: 25 - 35% of patients requiring PEP should receive both passive and active immunization
- < 1% of all PEP are comprised of vaccine and RIG
- HRIG: confidential quantities on specific markets but too expensive for most people
- ERIG: cheaper and safe (purified) but limited quantities, inaccessible to those that need it most

Rabies immunoglobulins

Additional problems

- ERIG production:
  - discontinuation by most international manufacturers
  - where it has been initiated it remains limited and hardly satisfies national needs
  - animal protection groups condemn animal rearing

WHO Initiative

WHO consultation May 2002

- accelerated research on and development of products which could be used as alternatives to RIG
  - MAbs cocktail
  - Transfectomas
  - plant expression systems

WHO Initiative

Anti-G MAbs for PEP

- Active principle in RIG is constituted by antibodies specific to rabies glycoprotein (G)
- Availability of human and murine MAbs
- Neutralizing activity *in vitro & in vivo* demonstrated
- Production could potentially reach large quantities at low cost
- Quality control easier than for polyclonal serum
- Technology transfer to selected developing countries

WHO Initiative

↓

WHO Project

- joint endeavour of two different programmes
  - Communicable Diseases (CDS)
  - Family and Community Health (HTP) – Initiative for Vaccine Research (IVR)

WHO Project

Objectives

- discover a unique murine MAb cocktail from available panels
- make a product which
  - can be used broadly in developing countries
  - should be available at the lowest possible reasonable price to the public sector
- phase 1: selection of MAbs & validation both *in vitro & in vivo*
- phase 2 selection of a production technology & transfer

WHO murine MAb cocktail

Selection criteria

- Biological activity
  - Neutralizing potency: minimum of 100 IU/ml
  - Breadth of neutralisation: broad spectrum
  - Production stability: loss 10% up to 30 passages
- Affinity binding sites I, II, III
- Immunglobuline isotype preferably IgG1, 2a & (3).
- History of hybridomas contamination (FMDV, TSE)

WHO murine MAb cocktail

Candidate MAbs

Institution	N° MAbs	designation
Wistar Institute, USA	1	1112
CDC, USA	1	6271-3
ADRI, Canada	2	M777-16 M727-5
FLI, Germany	1	E559.1.14

WHO murine MAb cocktail

Candidate MAbs

	E559.9.14	1112-1	62-713	M727-5-1	M777-16-3
Strain of mouse	Balb/c	Balb/c	Balb/c	Balb/c	Balb/c
Antigen	ERA G	ERA G	whole ERA	whole ERA	whole ERA
contamination risk (FMDV, TSE)	very low	very low	none	none	none
IgG subtype	IgG 1	IgG 1	IgG 2b	IgG2a	IgG 1
Antigenic site on G	II	II c	III (?)	?	?
Escape mutant	Yes	Yes	No	No	No
IU per ml	62.5	3	30-60	22-32	11-32
Production stability	no loss	not known	no loss	slight instability	slight instability

WHO murine MAb cocktail

In vitro studies

genotype	E559.9.14	1112-1	62-71-3	M727-5-1	M777-16-3
RABV (N = 22)	3	5	2	7	4
Mokola					
Lagos Bat	n.d.	n.d.	n.d.	n.d.	n.d.
Duvenhage					
EBLV-1					
EBLV-2					
ABLV					
Aravan					
Khujand					
Irkut					
WCBV					
N° lyssaviruses not neutralised	5	8	6	9	6

WHO murine MAb cocktail

Escape mutants

MAb	Virus	G - Protein
1112-1	CVS	change of Gly to Glu in AA 53
E559.1.14	SAD B19	change of Leu to Arg in AA 57 change of Lys to Glu in AA 217

WHO murine MAb cocktail

E559.1.14 escape mutant

	IU/ml								
MAb	10	5	2	1	0.5	0.25	0.125	0.063	0.03
E559.9.14									
62-71- 3									
727-5									
777-16									
1112-1									
D1									

WHO murine MAb cocktail

Next steps

- Testing of 5 formulations (mini-cocktails)
  - CDC MAb 62-71-3 associated with each of the 4 others plus a CDC/CDC MAb 62-71-3 combination
  - *In vivo* – CDC hamster model
  - *In vitro* – neutralisation FLI

WHO murine MAb cocktail

In vivo studies

hamsters

- preliminary tests (mini-cocktails)

chimpanzees

- final preparation by instillation of the product
  - safety
  - half-life

macaques

- challenge
- best treatment protocol
  - vaccine
  - administration (route, regimen…etc)

Other projects on MAbs

Human MAb cocktail

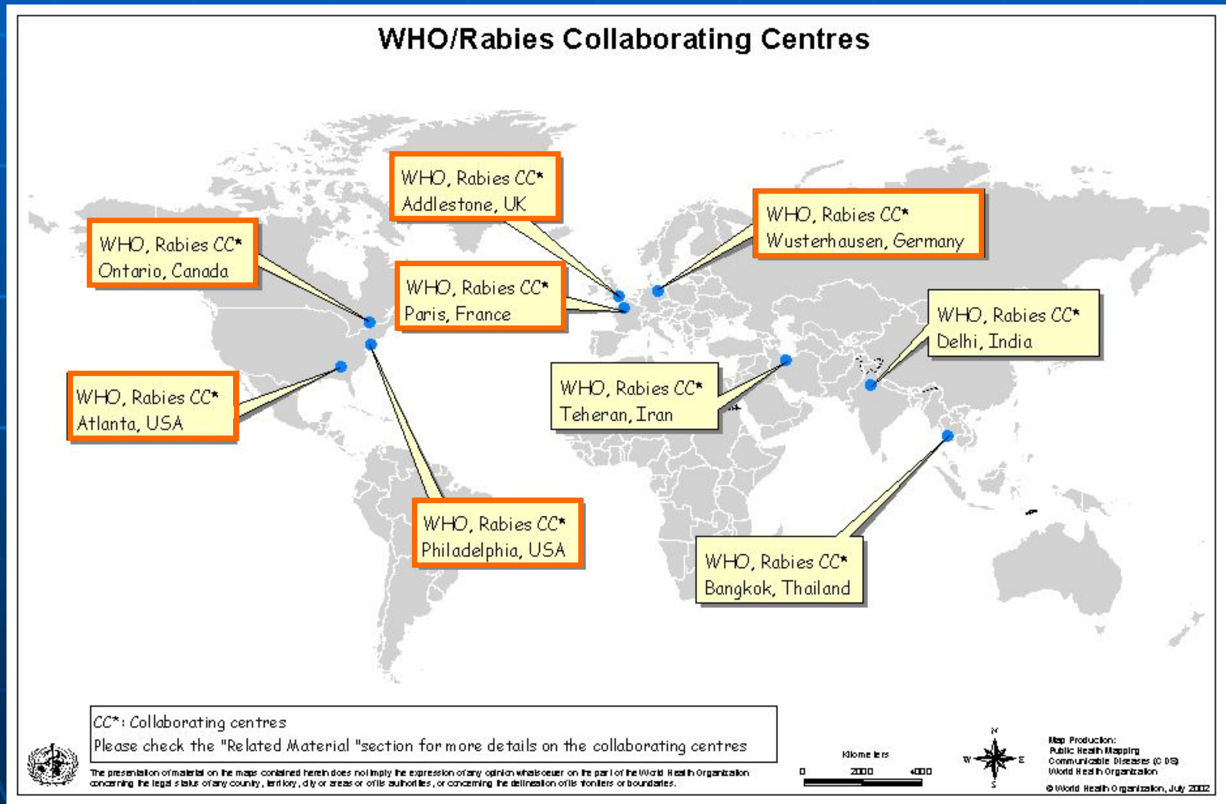
Prosniak et al. (2003): Development of a cocktail of recombinant expressed human rabies virus-neutralizing monoclonal antibodies for postexposure prophylaxis of rabies. Infect Dis. 188(1):53-6.

WHO murine MAb cocktail
Human MAb cocktail

- human MAbs for rabies PEP would be preferential
- however:
  - murine MAbs can be humanized easily
  - uniqueness of the WHO project consists in the preferential conditions under which the product would have to be made available to the public sector of rabies infected countries of the developing world

WHO murine MAb cocktail

Participating institutions



HUMAN RABIES IN AFRICA; THE UGANDA PERSPECTIVE

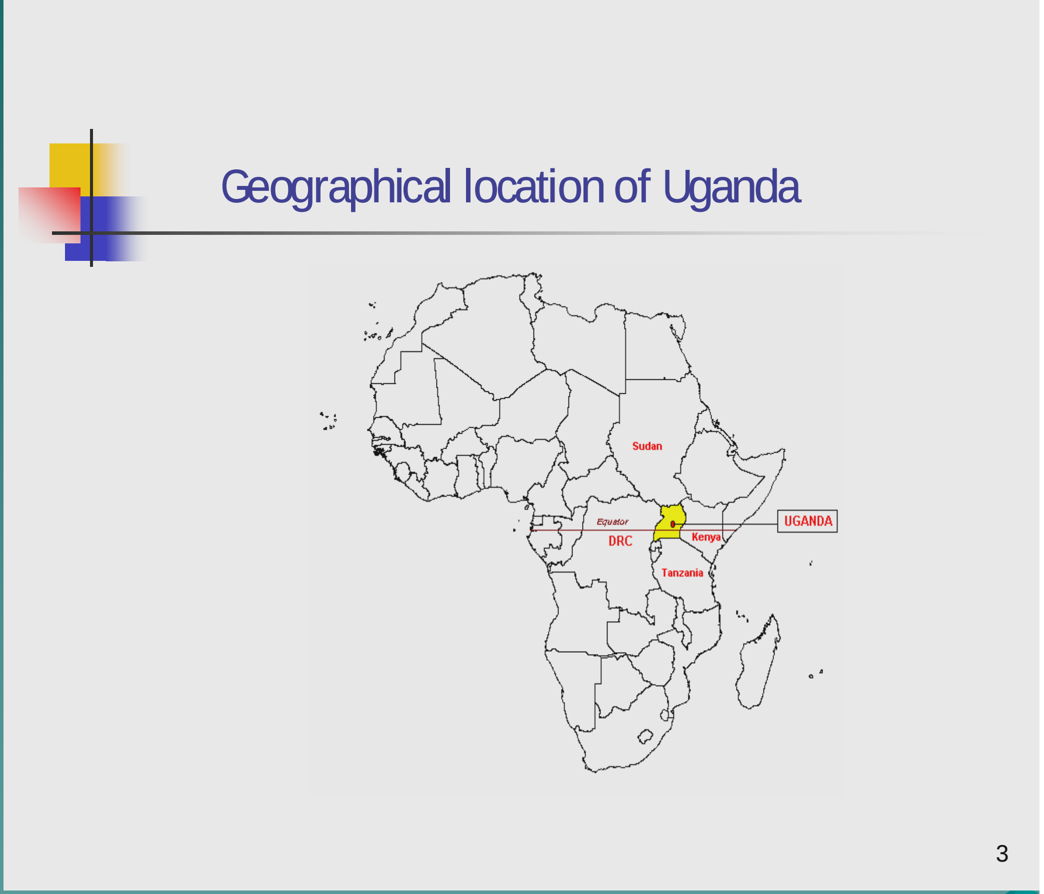
Dr. Winyi Kaboyo,
Ministry of Health
Uganda

1. Historical background

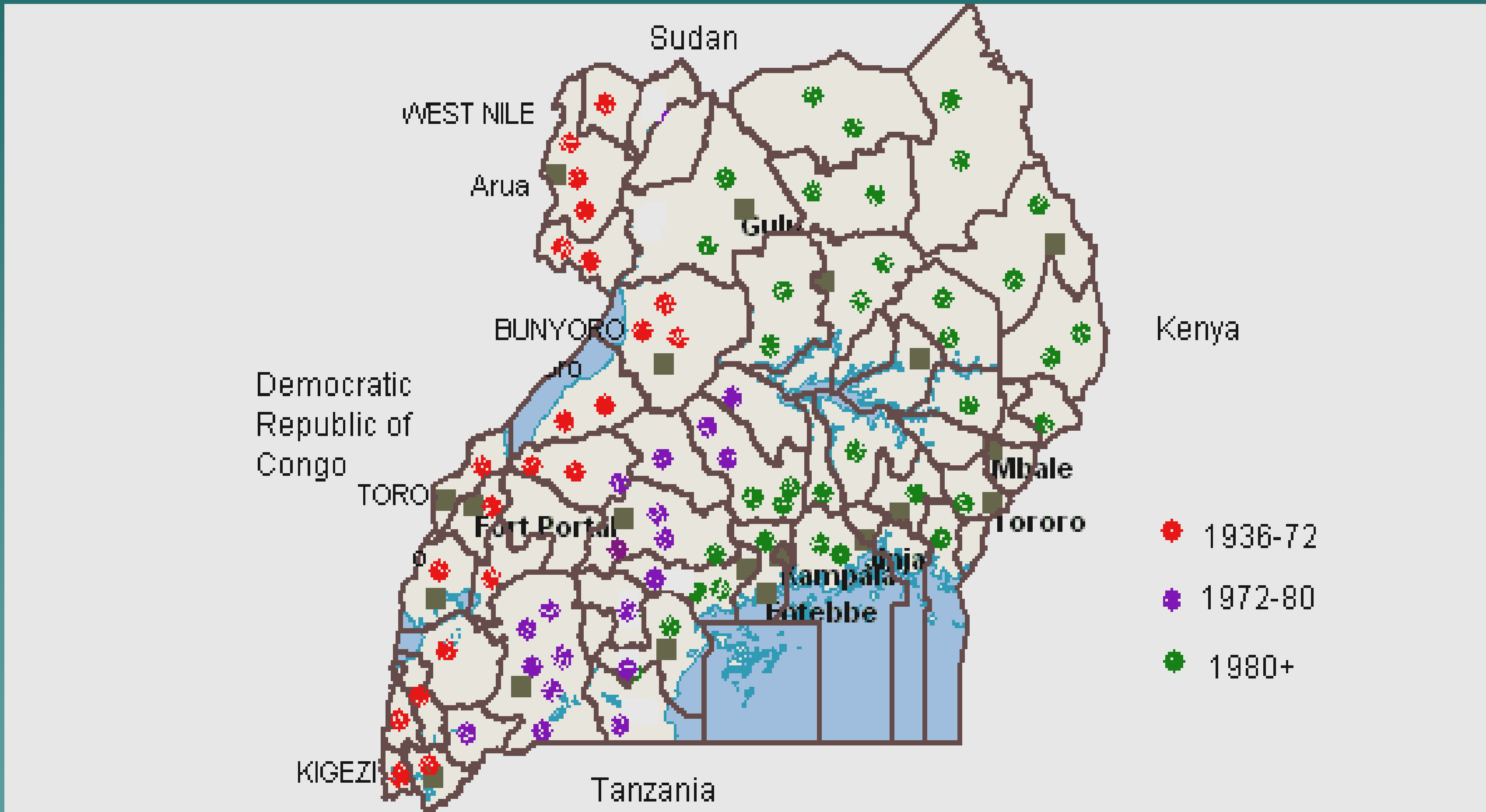
- The existence of rabies in Africa can be traced back many centuries.
- The disease has been described for centuries in Ethiopia and in many other African cultures as “Mad Dog Disease” (*Iraro ly’Embwa* in Rutoro language).
- In Ethiopia medical books of the 17th century have recommended treatments for people bitten by rabid dogs,
- The first officially recorded epidemic was in 1903 in Addis Ababa.

In other countries

- South Africa, rabies was first confirmed in 1892 in a dog brought from England;
- Sudan 1904,
- Mozambique in 1908,
- Kenya 1912
- Tanzania 1932
- Uganda 1936.



Geographical distribution of Human Rabies Cases in Uganda, 1936-1980+.



2. Epidemiology

- Annual global human rabies mortality 40,000 to 60,000 and for Africa is 4,000 people (WHO estimate)
- Uganda btw 1992-2004; average of 3,800 bite victims/yr got PET
- incidence of 19 animal bites per 100,000 population.
- 24 rabies mortality; incidence rate of 0.12 deaths per 100,000 population (mid-term pop 20M)

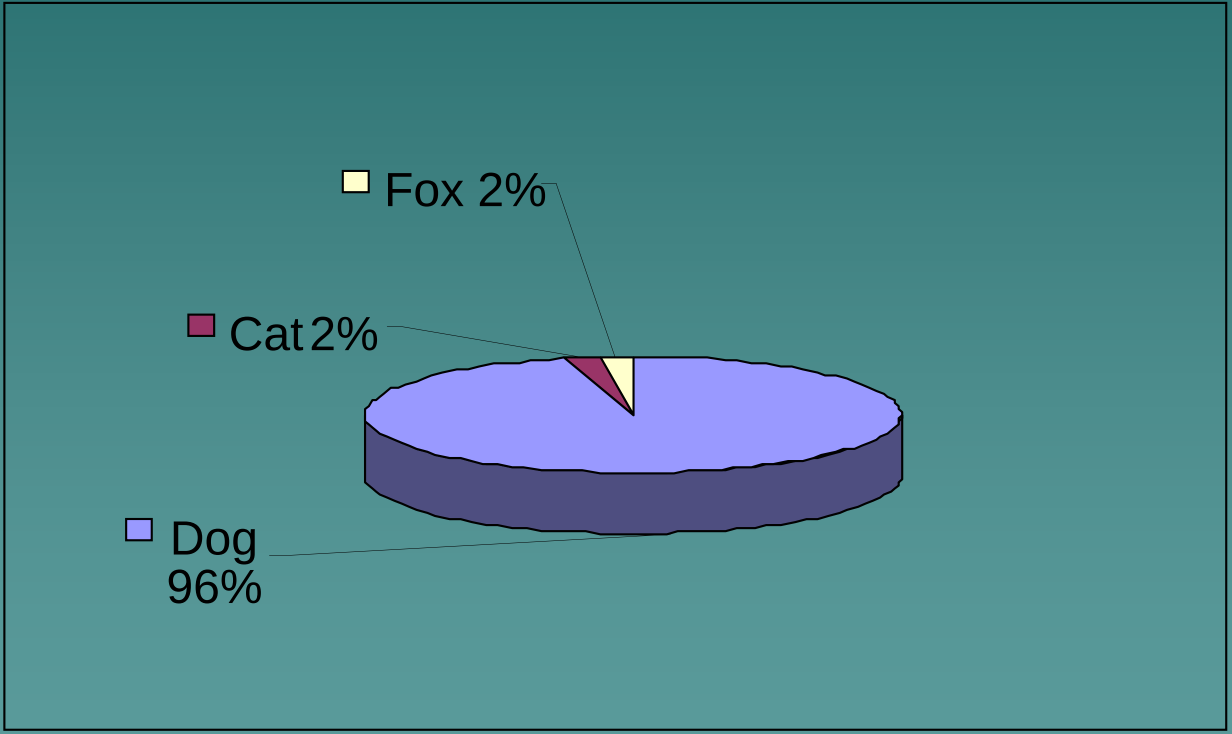
Reported Human Rabies Data, Uganda (1992- 2004).

Year	Rabies vaccine doses	Estimated cost (US$)	No. of post exposure treatments	No. of rabies clinical cases/ deaths
1992	3,976	35,000	766	50
1993	5,720	52,000	1,518	23
1994	8,298	74,700	2,614	15
1995	13,623	122,000	3,222	14
1996	16,000	144,000	1,698	9
1997	16,000	144,000	2,916	10
1998	16,000	144,000	3,112	10
1999	10,000	90,000	4,537	5
2000	10,000	90,000	6,037	12
2001	19,570	119,371	6,577	35
2002	15,133	98,365	4,789	12
2003	19,500	128,600	6,929	105
2004	10,550	79.000	4,628	18
Total	**164,370**	**1,321,036**	**49,343**	**318**

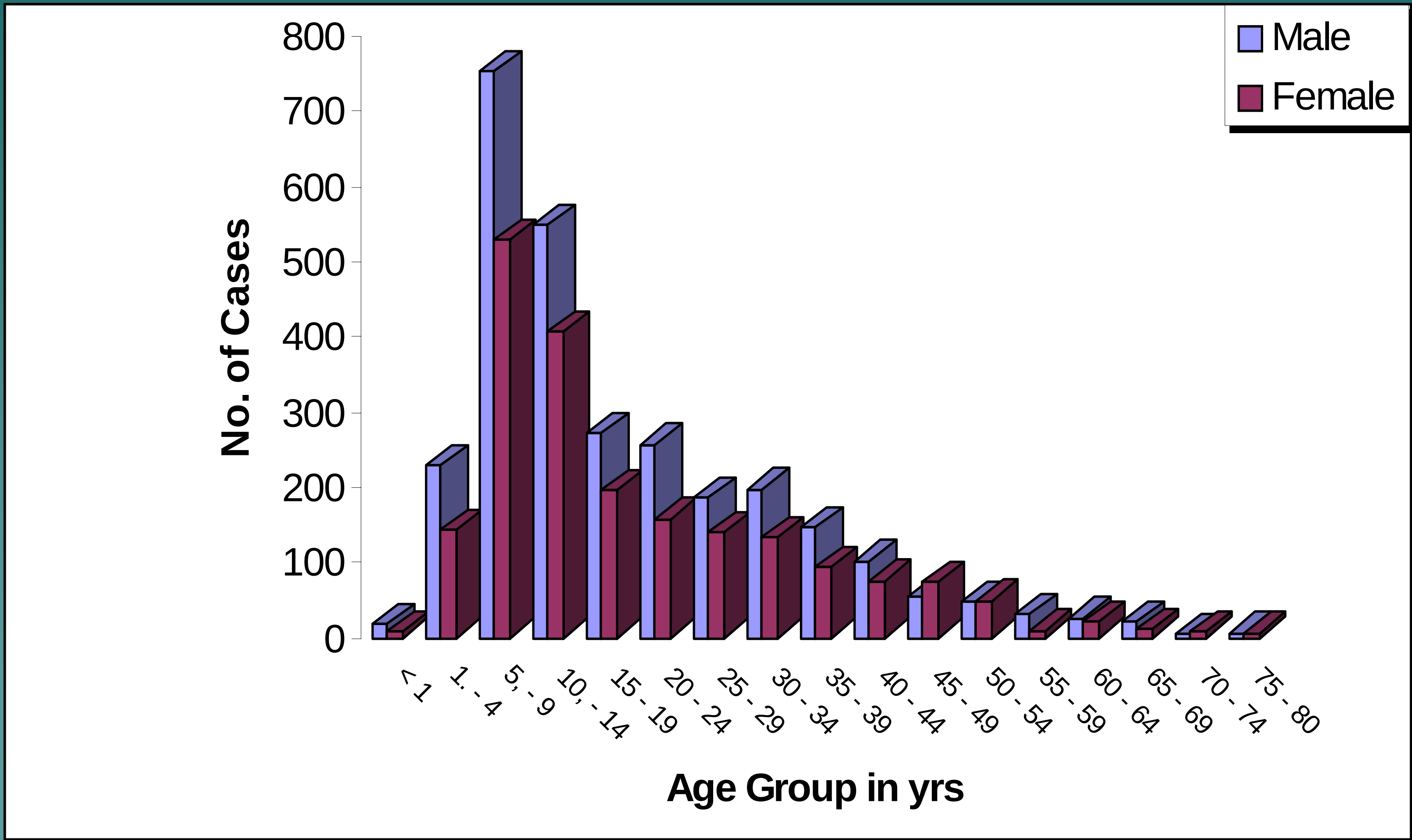
Human rabies transmission:

- In Africa: “urban cycle” dog to human transmission is responsible for over 95% of all human exposures
- e.g in Nigeria: 98.8%
- Ghana: 98.7% and in
- Uganda: dogs responsible for 95.8% of all human exposures

Reported Human Rabies deaths by Source of Exposure; 2003, Uganda.

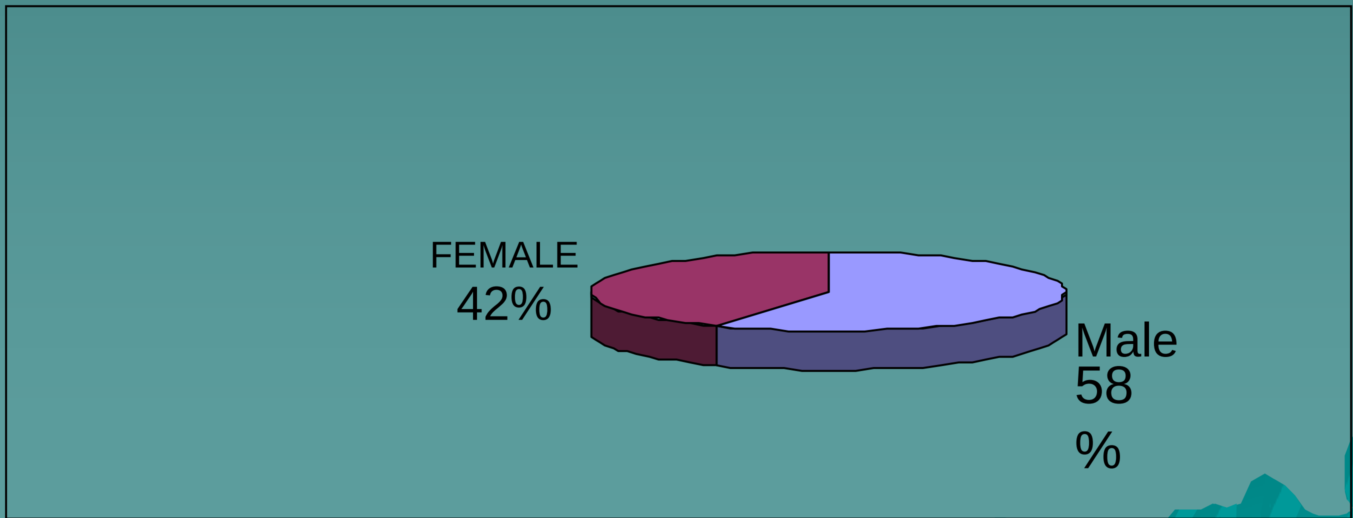


Age and Sex distribution of Bite cases given rabies PET, Uganda, 1990-1994.



Sex distribution of reported human rabies deaths in Uganda, 2003.

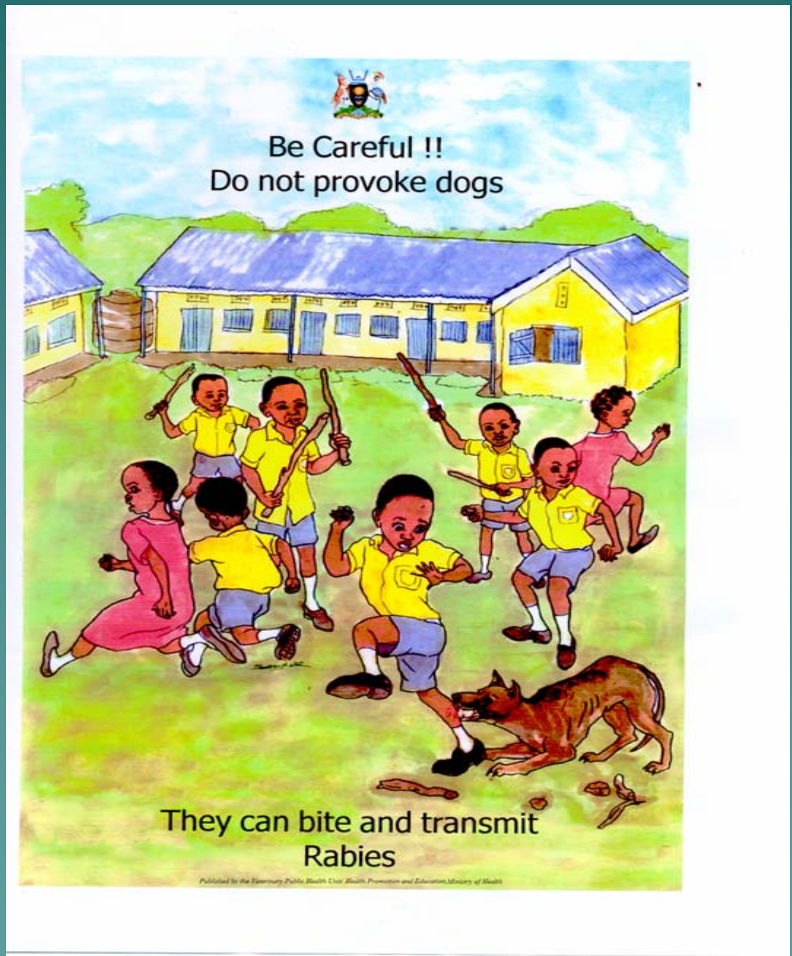
- 105 people who died from rabies, 60 were males and 44 females
- Ghana 257 deaths from rabies 153 (60%) Males and 104 (40%) were Females

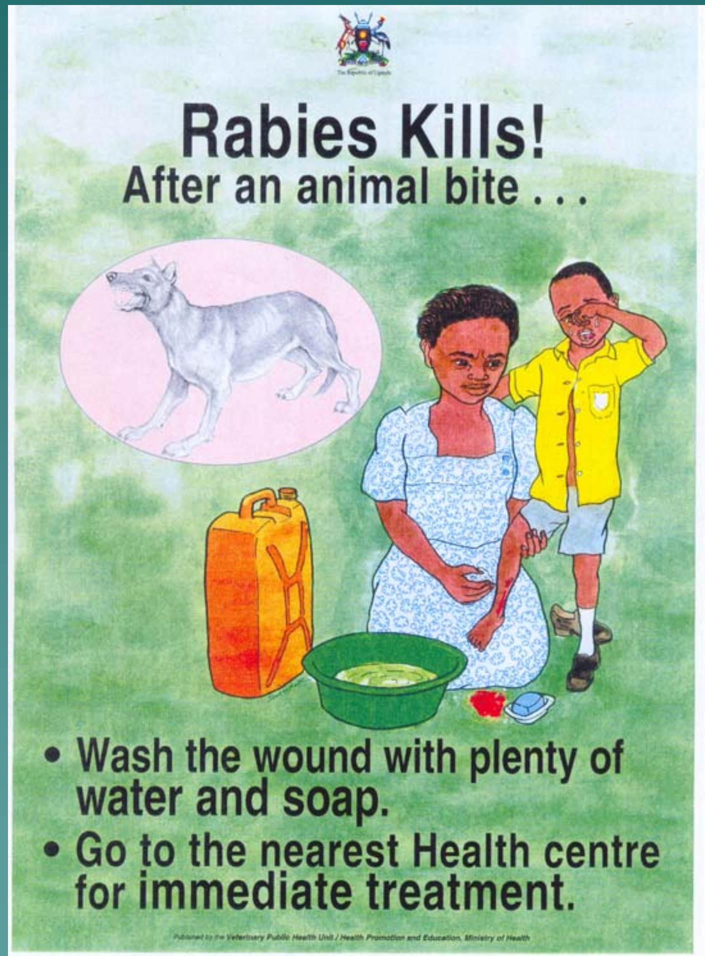


Strategies for human rabies Prev & Control

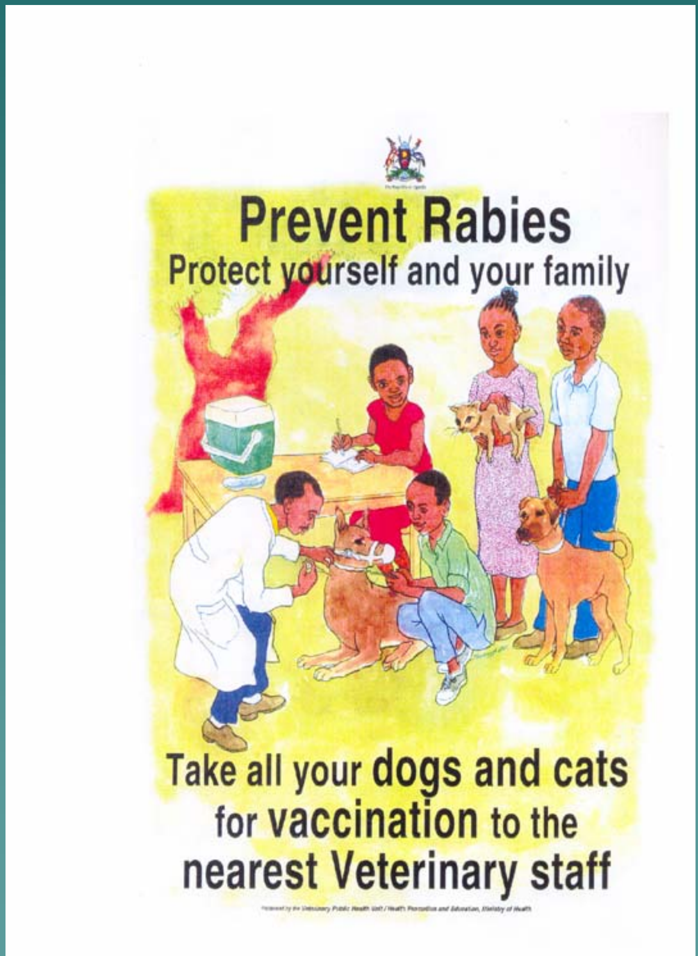
- Animal (dog & cat) vaccination this is not regularly done coverage is still about 20%
- Private vaccination at a fee US$ 3-5/pet; vast majority of pets not protected
- PET: im and recently introduced id treatment of suspected human exposures.
- Health education/School Health Services /advocacy/ target mainly children (5-19yrs), community leaders
- Develop & use of IEC materials

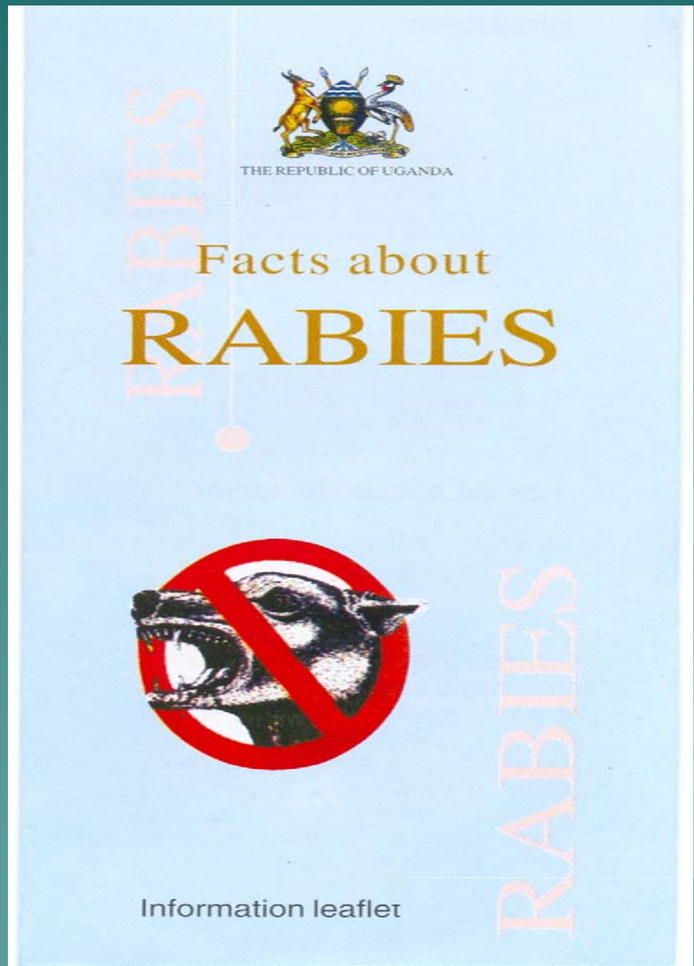
Health Education Posters





IEC materials



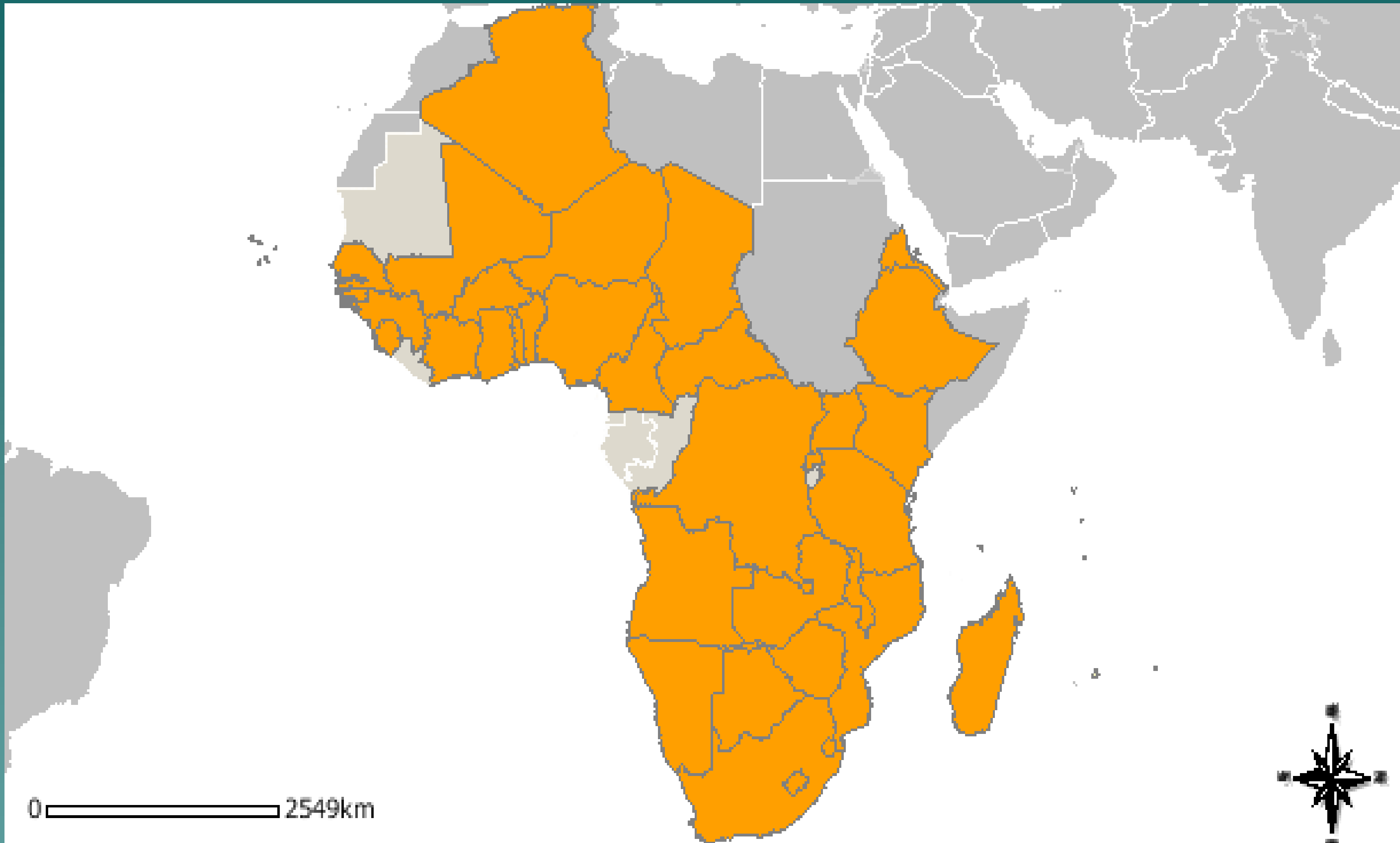


Challenges

- 1. low priority & commitment given to rabies compared to:

  -Malaria, HIV/AIDS, etc rabies cases/deaths are fewer ("not seen")
- 2 little or no funding
- 3.Lack of reliable data/ under-reporting to no reporting cf. (WHO/RABNET)

Reported presence of rabies 1993



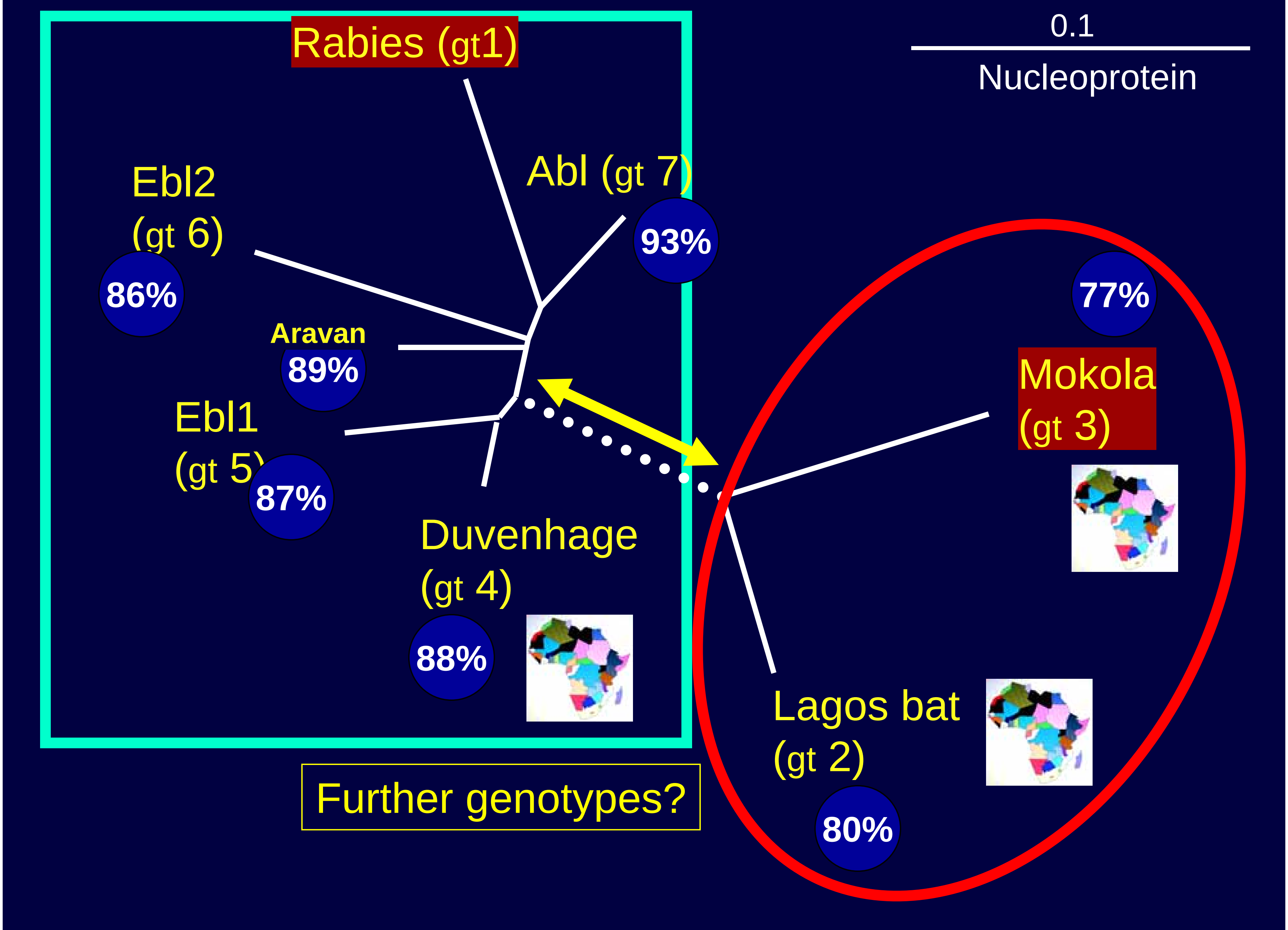
Reported presence of Rabies 2003

Conclusion & Way forward

- Human rabies still a public health problems but not regarded as a priority (except by SEARG ??)
- Need for improved surveillance , and reporting to highlight the burden of disease/economic impact/ DALYS for rabies cf to other public health priorities
- More funding of prev & control activities
- Research; develop new faster diagnositic techniques, curative drug?, simpler to admn.& affordable vaccine, introduce cost-effective PET regimen e.g. 2-1-1 im and id schedules.

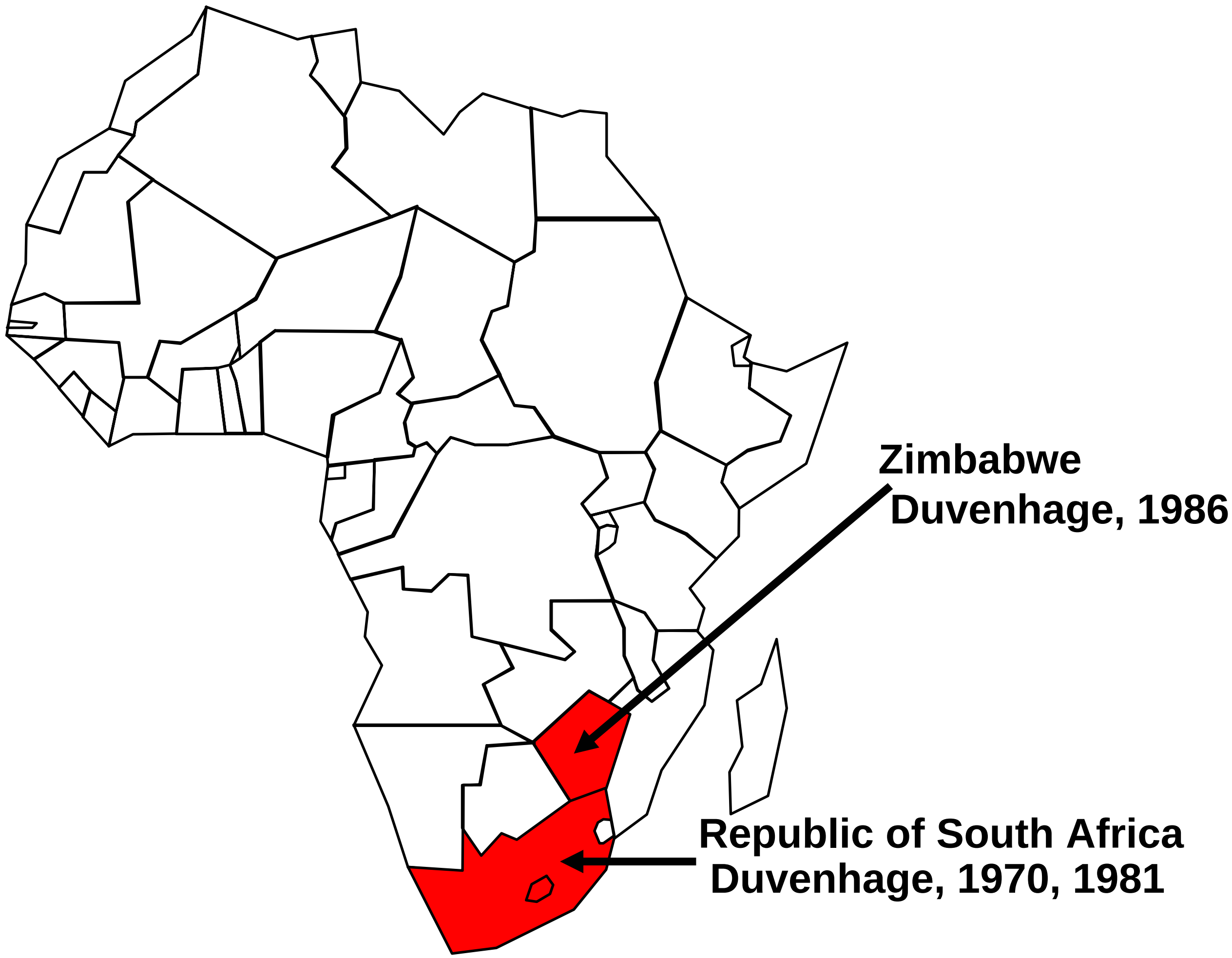
Acknowledgement

- The SEARG Committee for the invitation and opportunity to make this presentation.

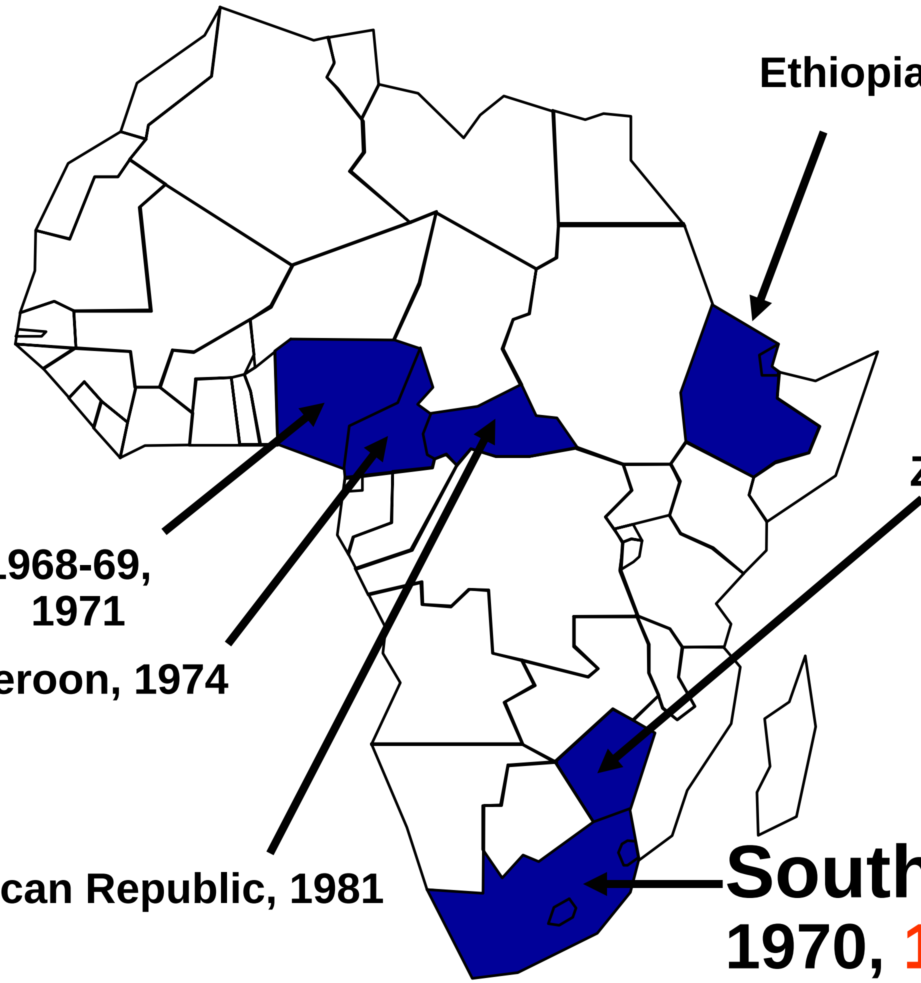


0.1
Nucleoprotein
Rabies (gt1)
Ebl2
(gt 6)
86%
Abl (gt 7)
93%
Aravan
89%
Ebl1
(gt 5)
87%
Duvenhage
(gt 4)
88%
77%
Mokola
(gt 3)
Lagos bat
(gt 2)
80%
Further genotypes?

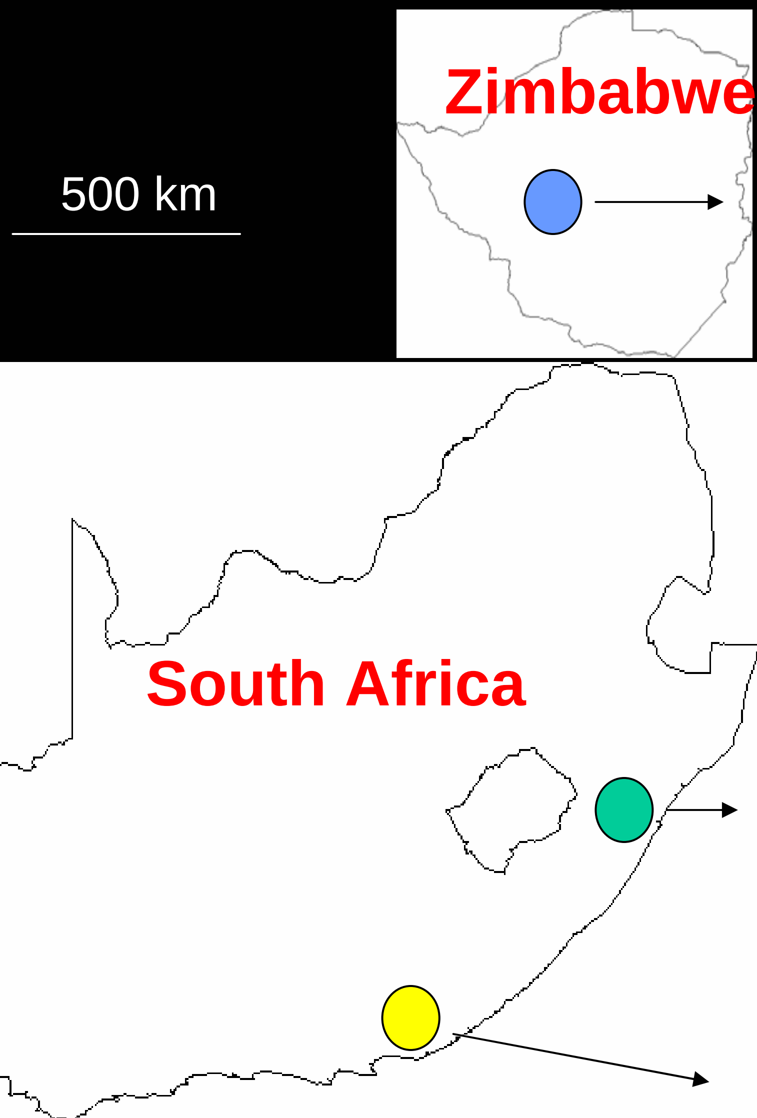
Isolation of African Duvenhage virus



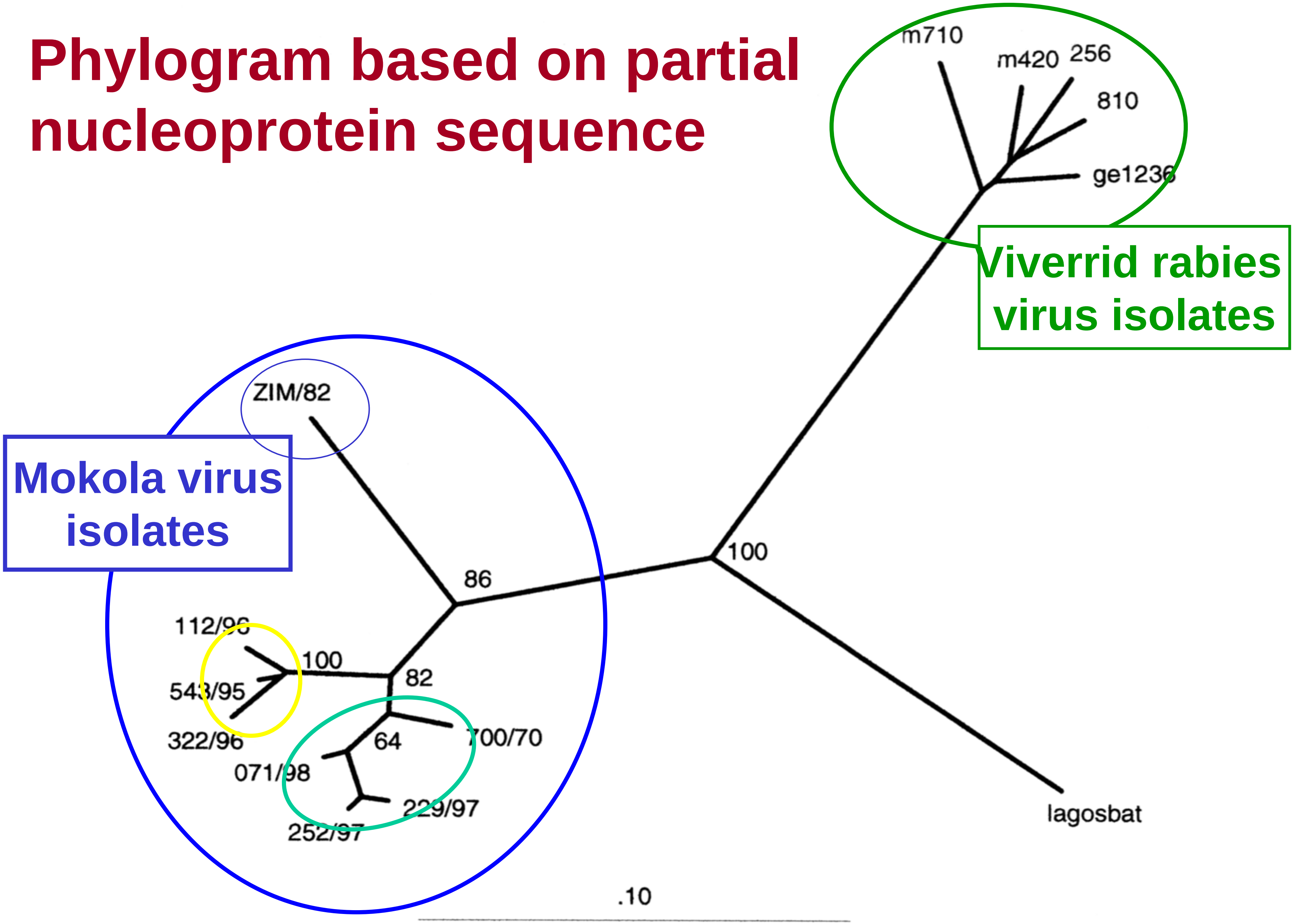
Isolation of African Mokola virus



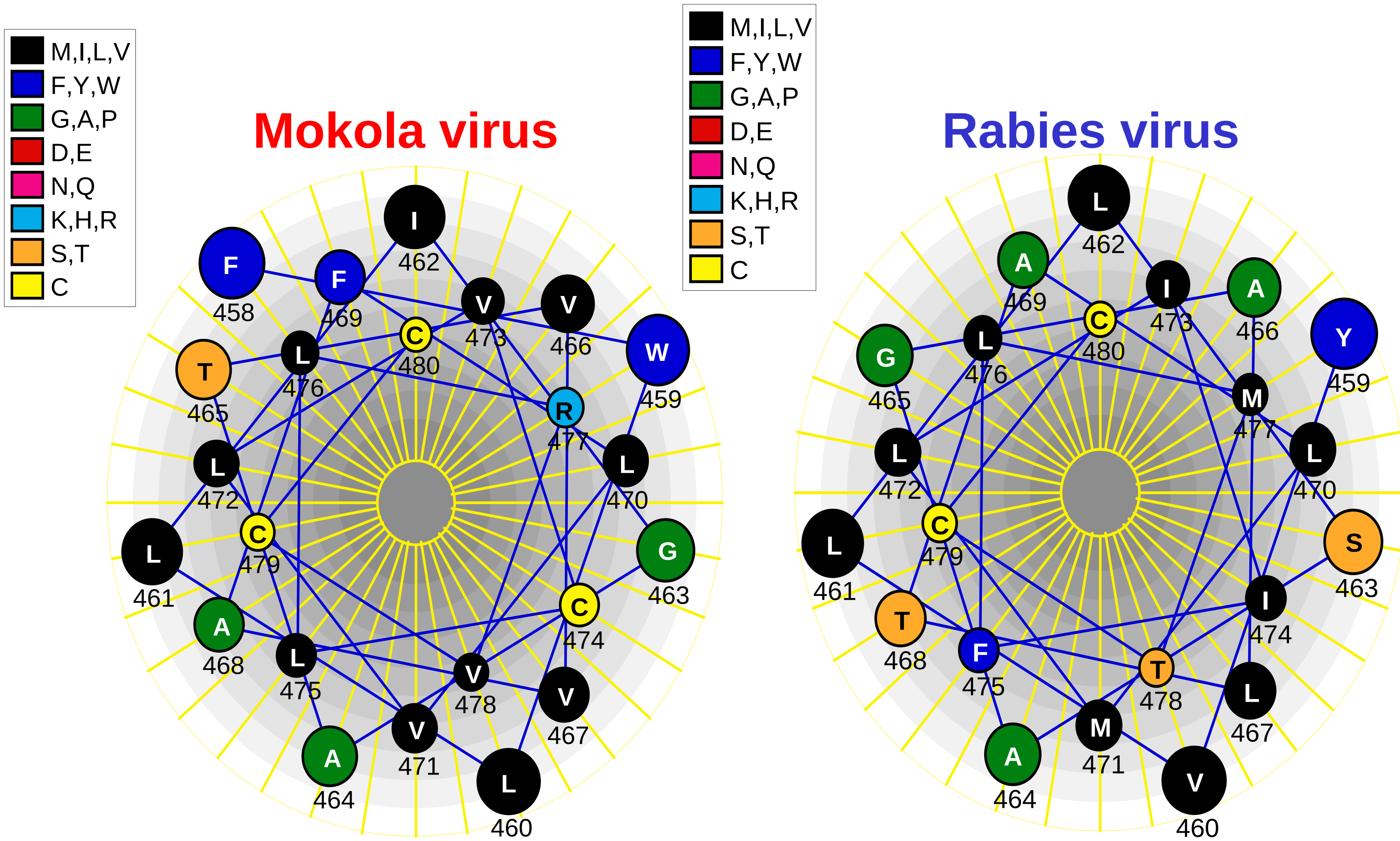
Mokola virus isolation sites in southern Africa



Phylogram based on partial nucleoprotein sequence



Comparison of the helical configuration of the glycoprotein transmembrane domains of Rabies and Mokola viruses



Major antigenic site III

	aa 330-338
mokola SA1	KRVDRWAD
mokola SA3	KRVDKWAD
mokola SA7	KRVDRWAD
mokola Zimbabwe	KRVDKWAD
mokola Ethiopia	KRVDRWAD
rabies (SAD)	KSVRTWNE

Cross-protection of mice against Lyssaviruses by Rabies Vaccines

Vaccine	Virus	Mouse Protection *
PM	Rabies (Dog)	30/30
PM	Lagos Bat	10/30
PM	Mokola	0/30
V-RG	Rabies (Dog)	12/12
V-RG	Rabies (Bat)	12/12
V-RG	Mokola	0/12

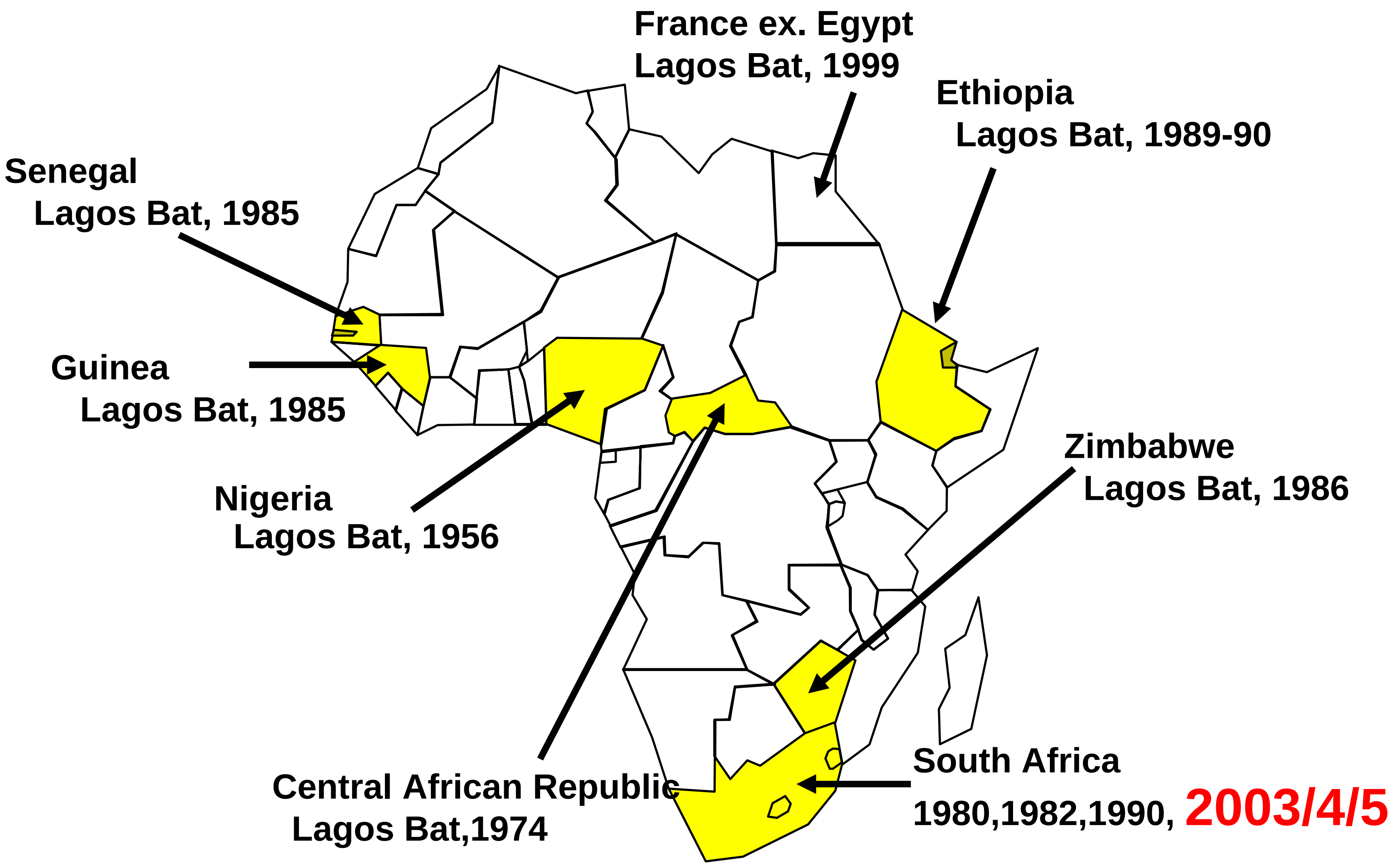
- *Number of survivors/number inoculated
- *Mice challenged IC 2 months post-vaccination

Cross-neutralization of Lyssaviruses by human sera following rabies vaccination

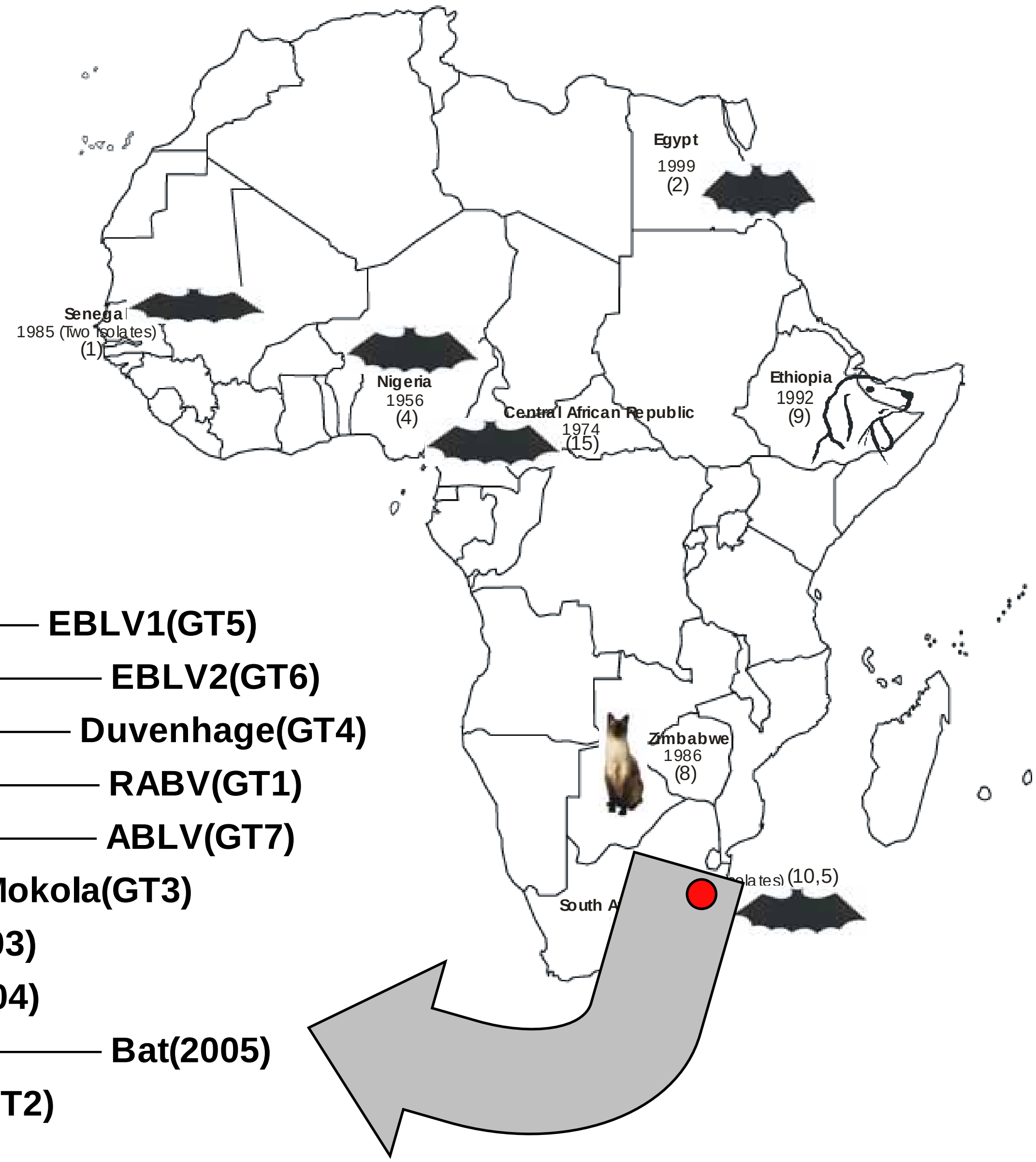
Virus	HRIG*	Serum 1	Serum 2
Rabies	940	>1400	200
ABV	625	>1400	250
EBV1	125	1300	36
EBV2	125	1200	19
Duvenhage	125	540	10
Mokola	5	33	<5
Lagos bat	12	170	45

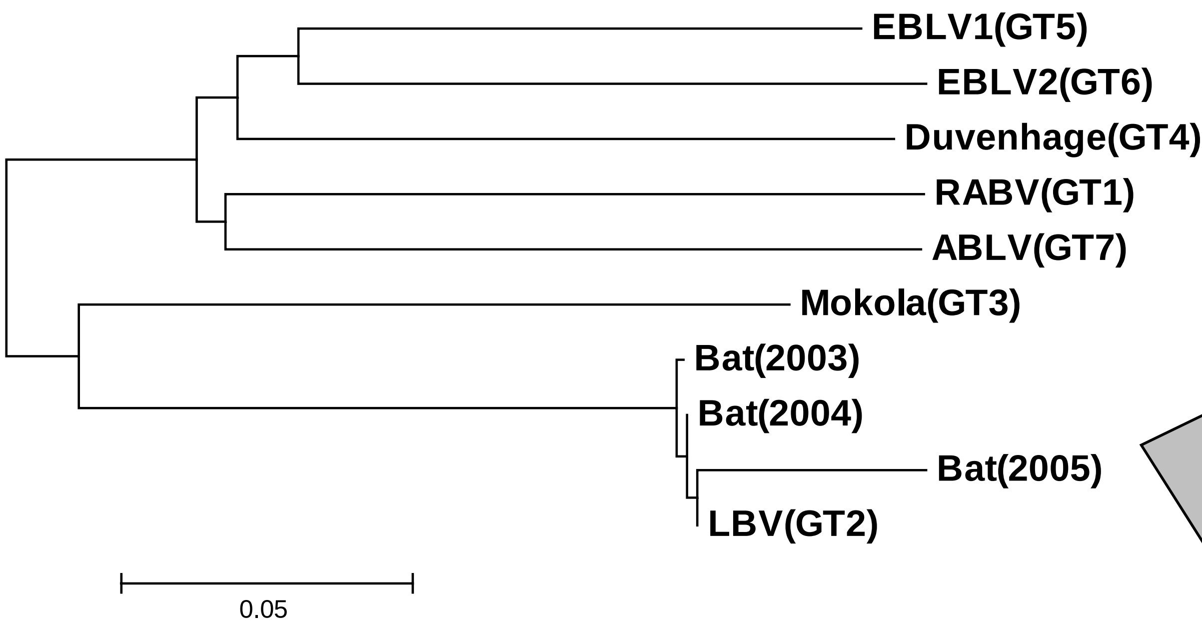
* Dilution of human rabies immune globulin or human test sera which neutralized ~50 TCID of virus in vitro

Isolation of African Lagos Bat virus



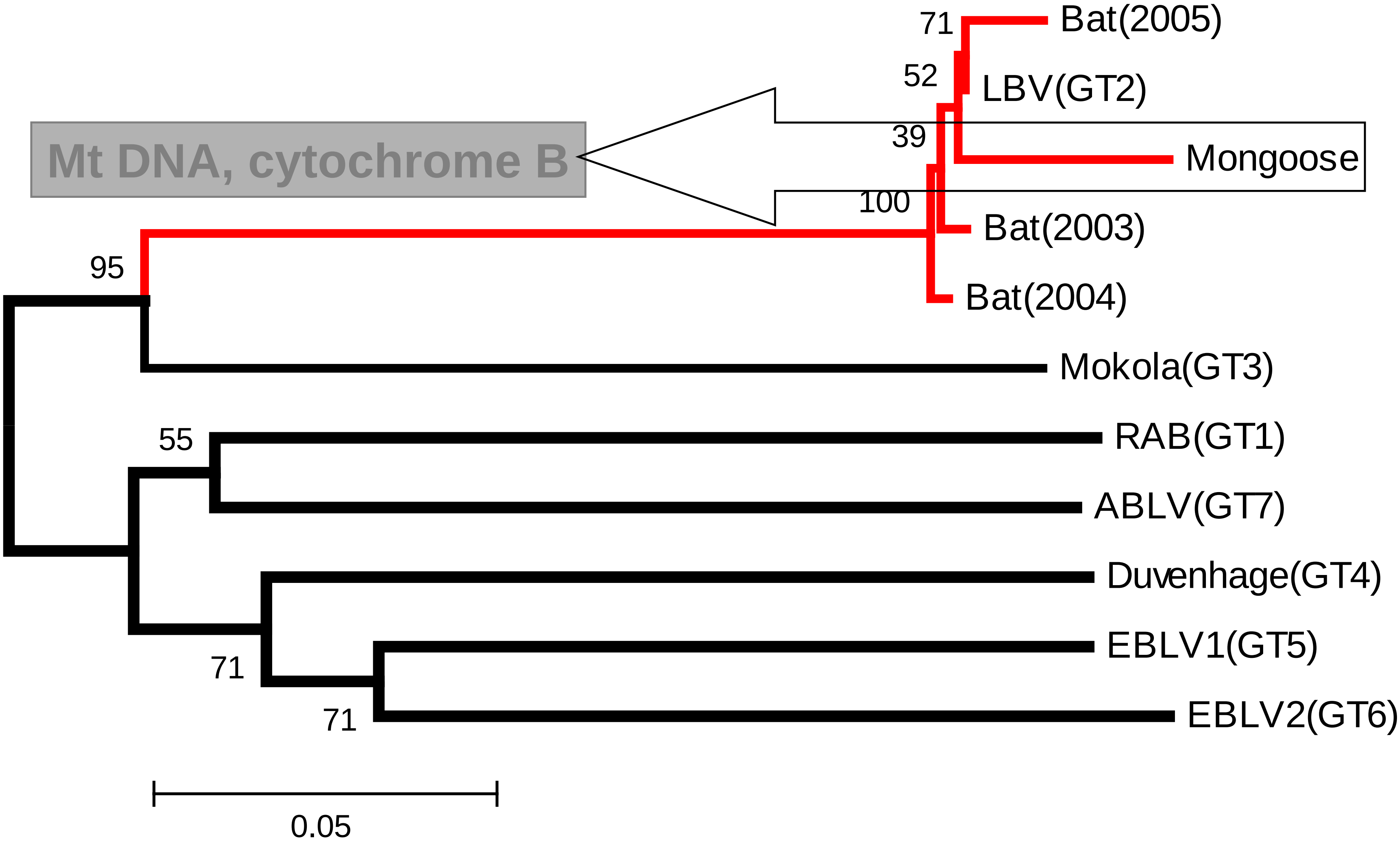
LBV isolated in KZN 2003-2005



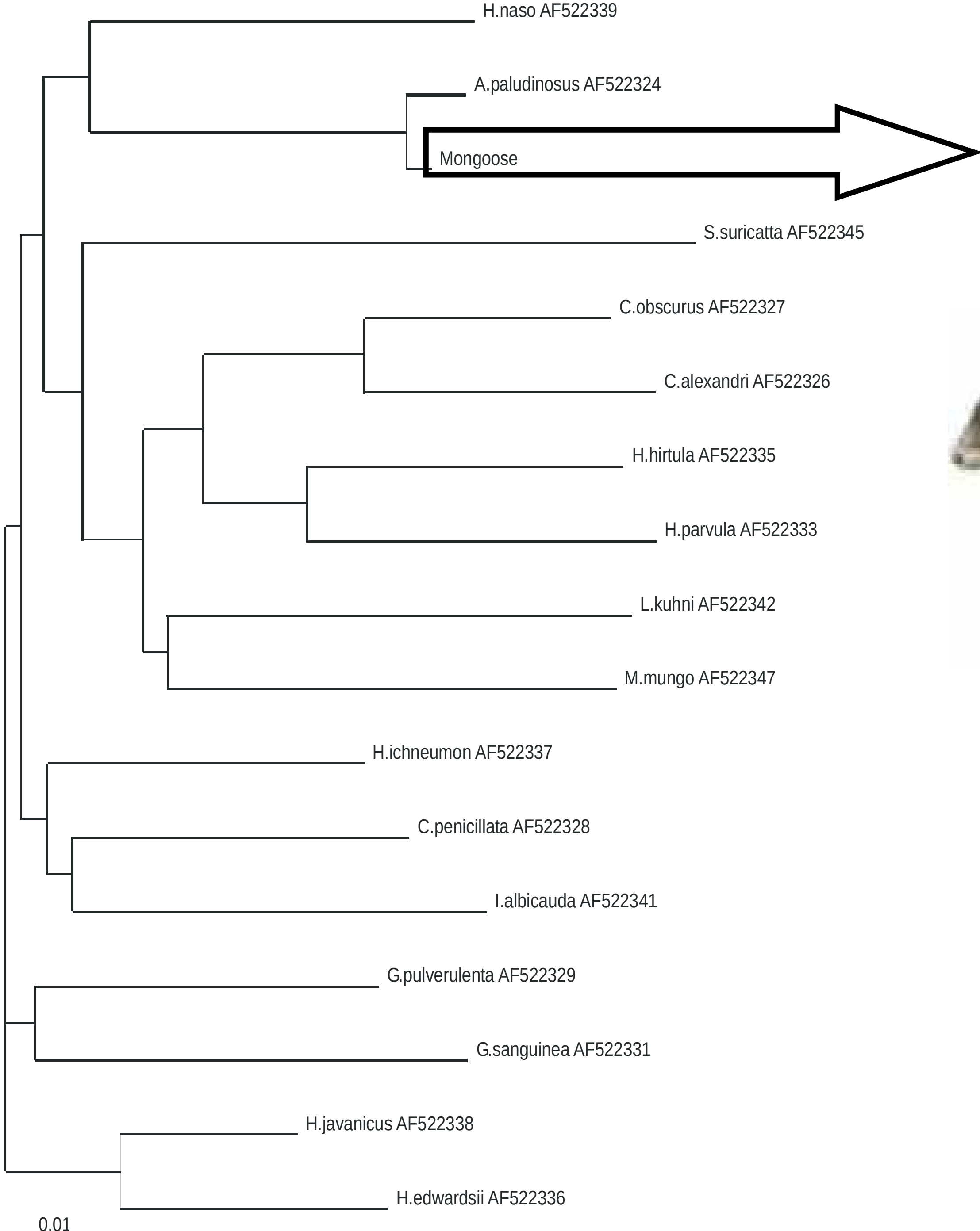


Conclusions?

- First isolation of LBV from SA in 13 yrs
- Sporadic? - due to inadequate surveillance!
- Suggest that LBV is persistently present in Megachiroptera



Bat(2005)
71
52
LBV(GT2)
Mt DNA, cytochrome B
39
Mongoose
100
Bat(2003)
95
Bat(2004)
Mokola(GT3)
55
RAB(GT1)
ABLV(GT7)
Duvenhage(GT4)
EBLV1(GT5)
71
EBLV2(GT6)
71
0.05

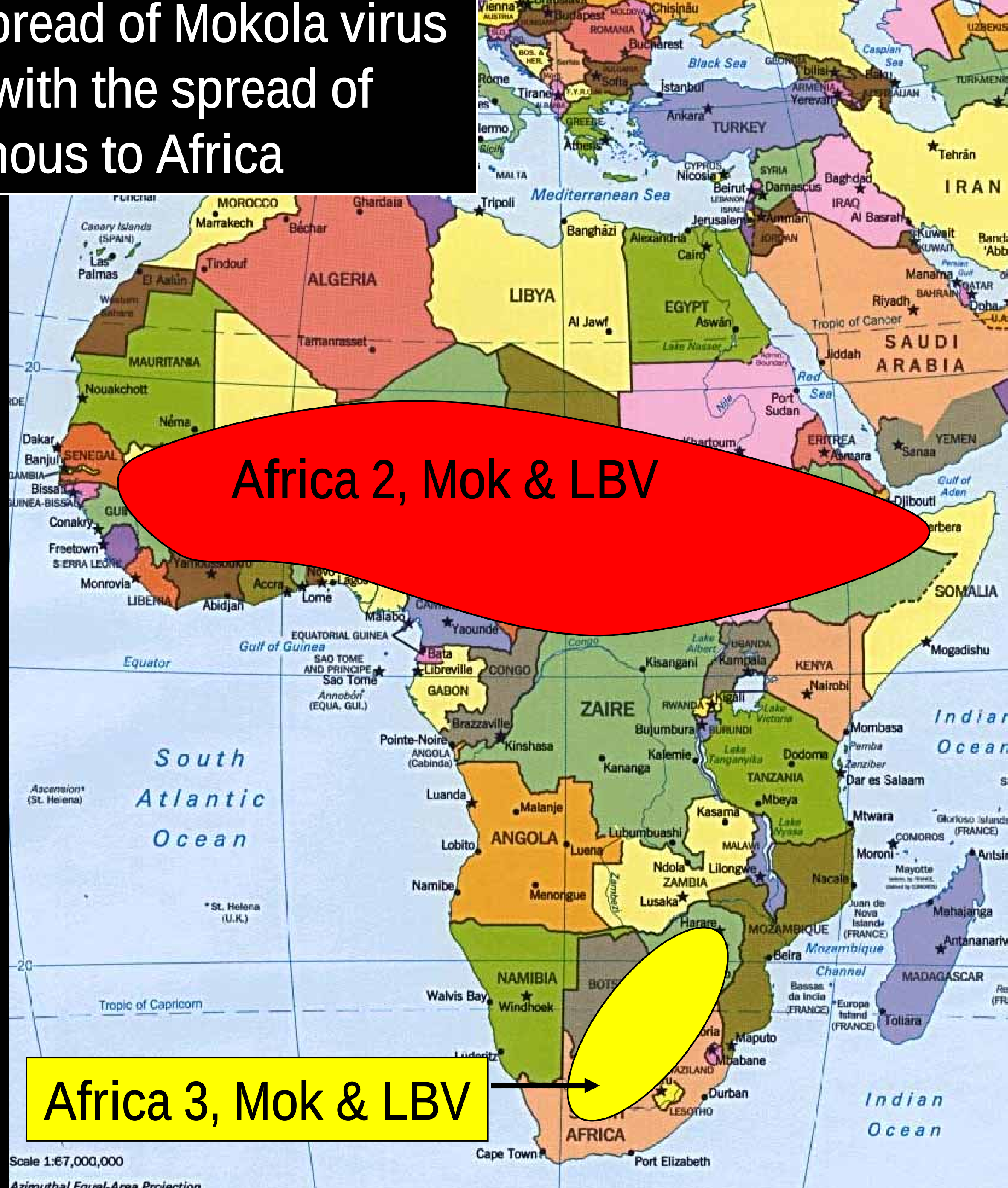


Water or marsh mongoose *(Atilax paludonsis)*

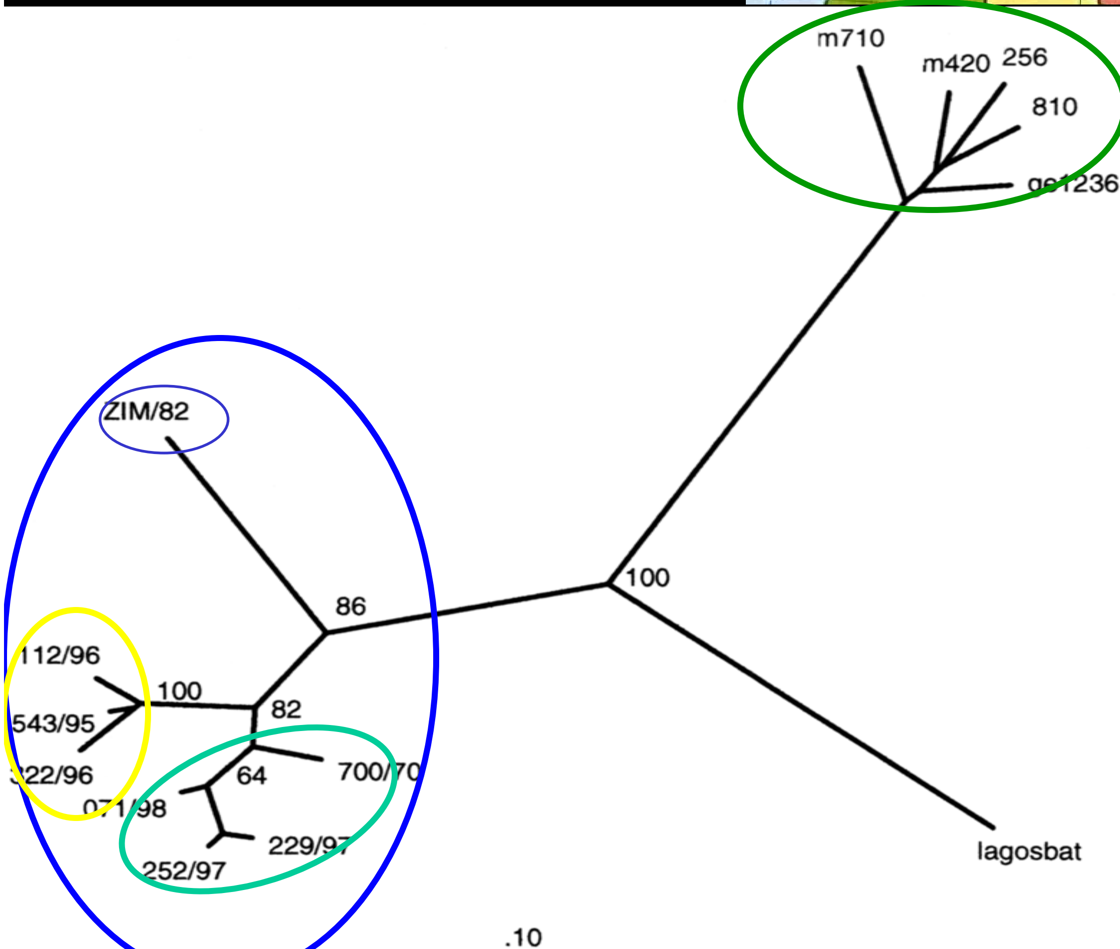
Conclusions?

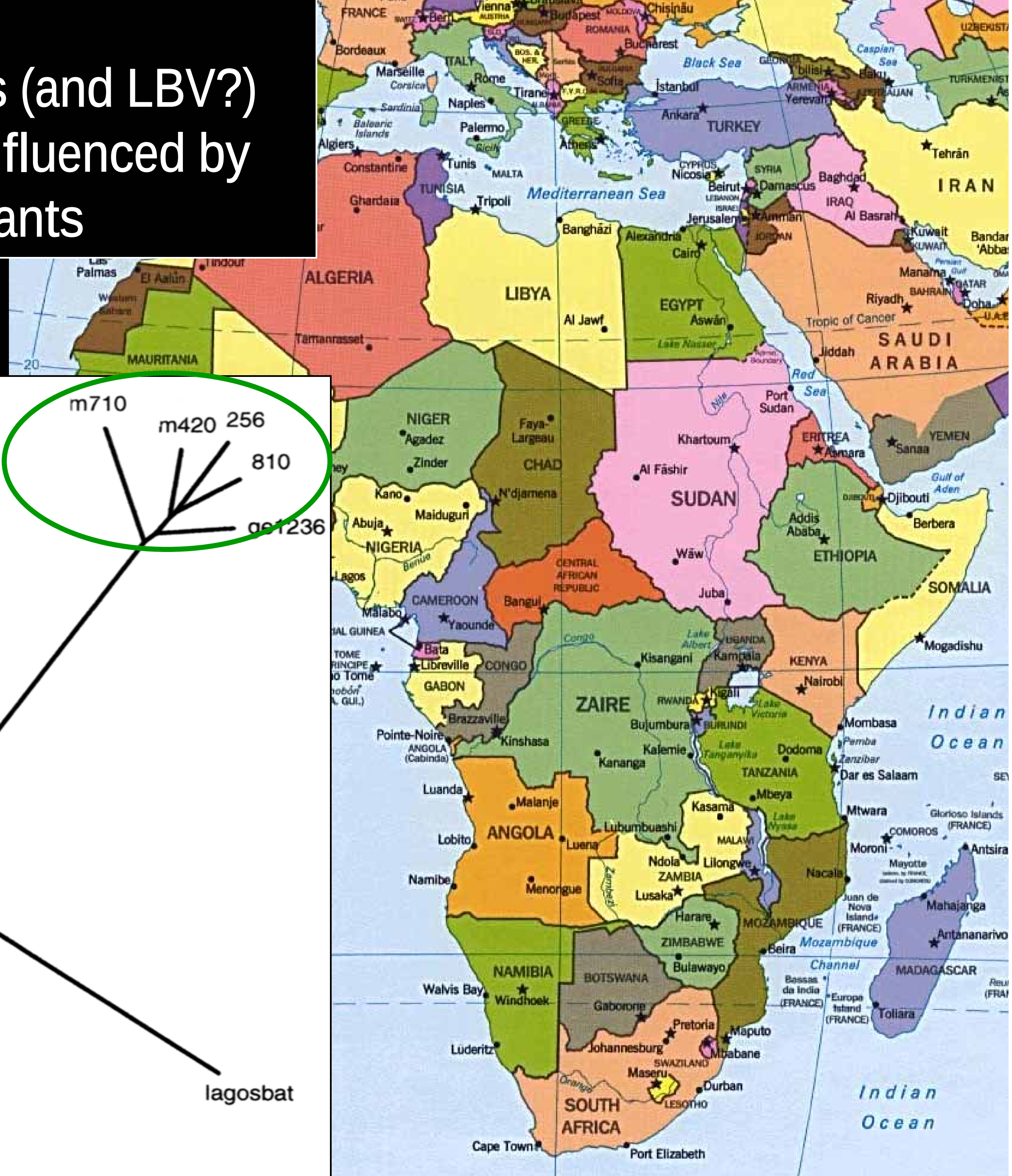
- LBV from terrestrial wildlife
- Lyssavirus (nonrabies) from mongoose sp.
- Genetic variation
- Maintenance and persistence
- Urban proximity and rabid behaviour (wildlife carnivore)
- “Vaccine failure” (domestic dog)

1. The geographical spread of Mokola virus and LBV coincides with the spread of rabies types indigenous to Africa



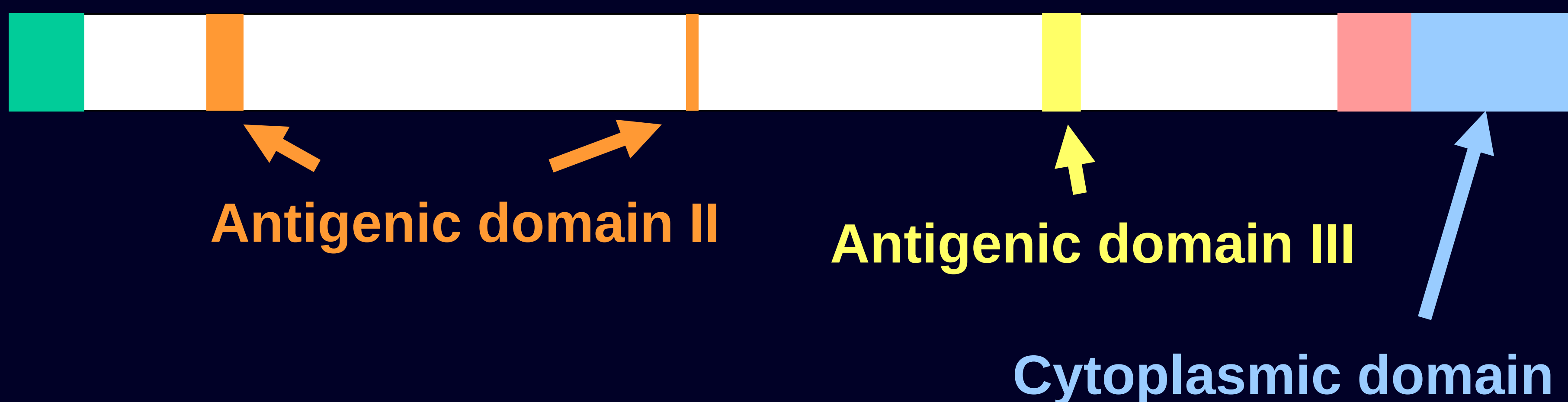
2. Pattern of Mokola virus (and LBV?) evolution is strongly influenced by geographical determinants





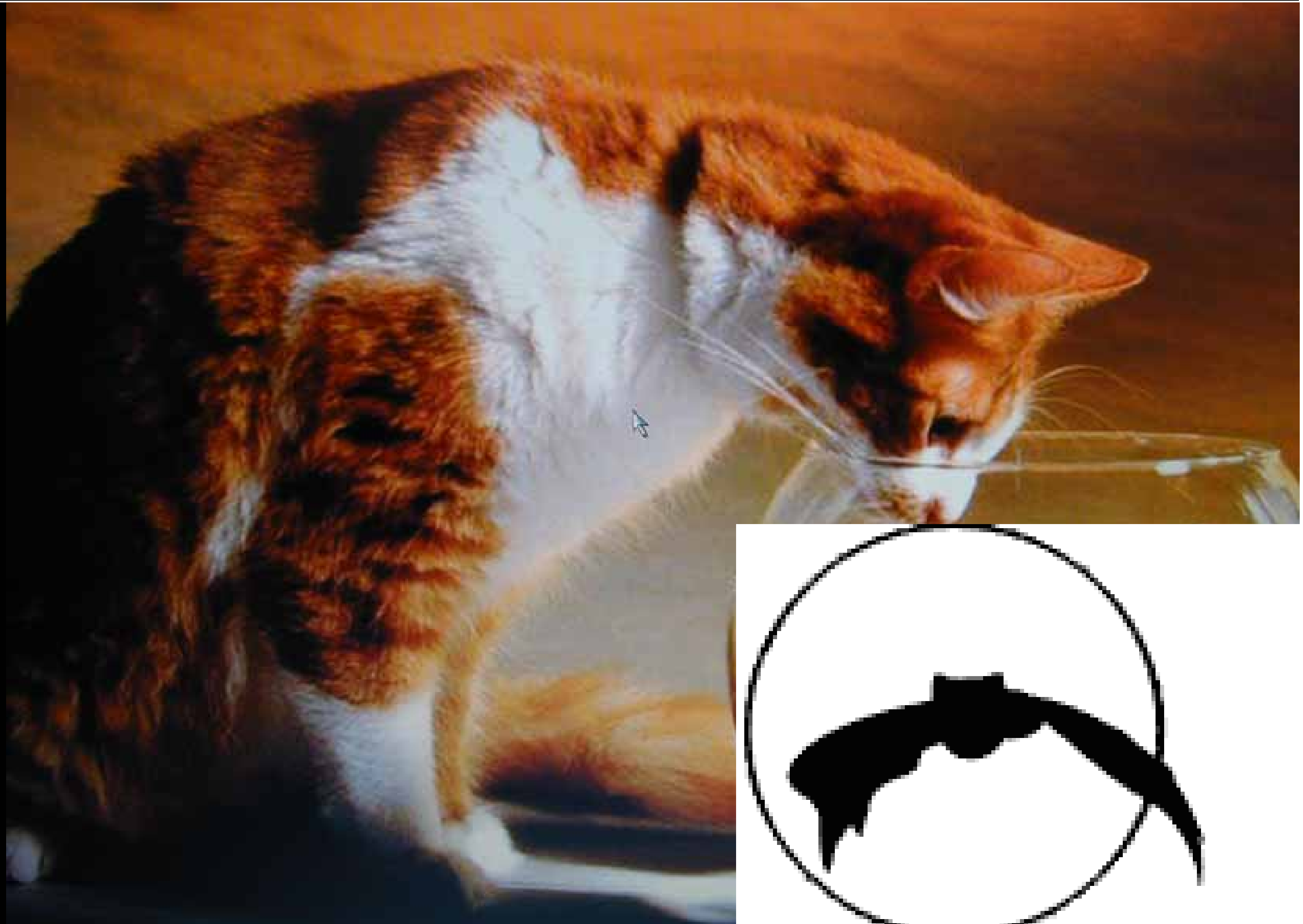
3. Similar genetic diversity **within** the Mokola and Rabies genotypes (based on full-length glycoproteins)

4. Important dissimilarities in the antigenic/cytoplasmic domains of Mokola/LBV and Rabies virus glycoproteins



5. Rabies vaccines do not protect against Mokola and LBV

6. The reservoir species for Mokola and many aspects of the epidemiology and pathogenicity of the African lyssaviruses are unknown



Key issues

•Nonrabies (rabies-related) lyssavirus infections are rare, although the viruses may be readily encountered where appropriate surveillance is applied.

•Several new nonrabies lyssaviruses were discovered and the incidence of others increased in recent years.

•Nonrabies lyssaviruses cause acute encephalitis, clinically indistinguishable from rabies, but their epidemiology is obscure

•Cross-reactive lyssavirus vaccines may never be commercially viable.

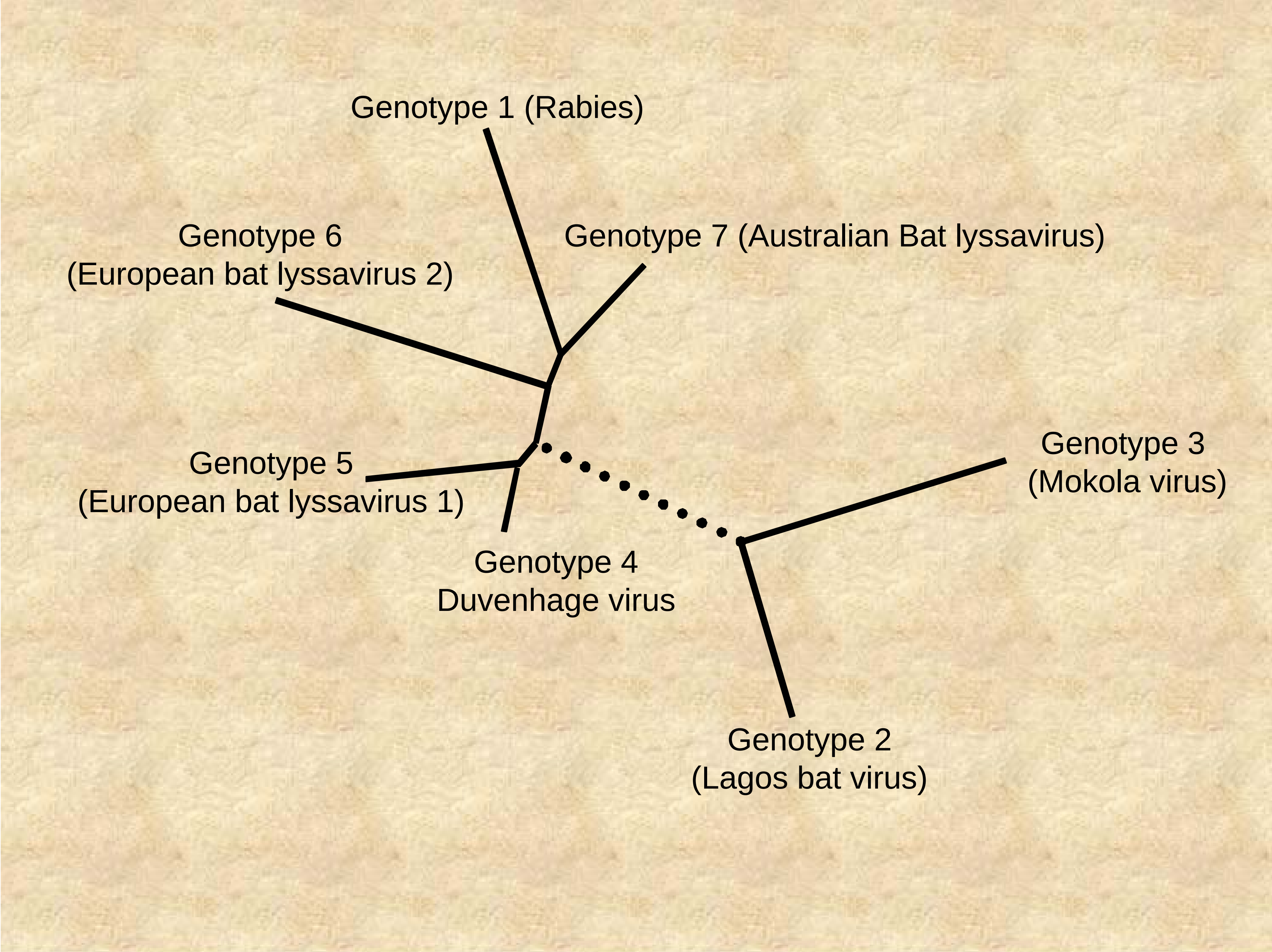
Rabies in Africa

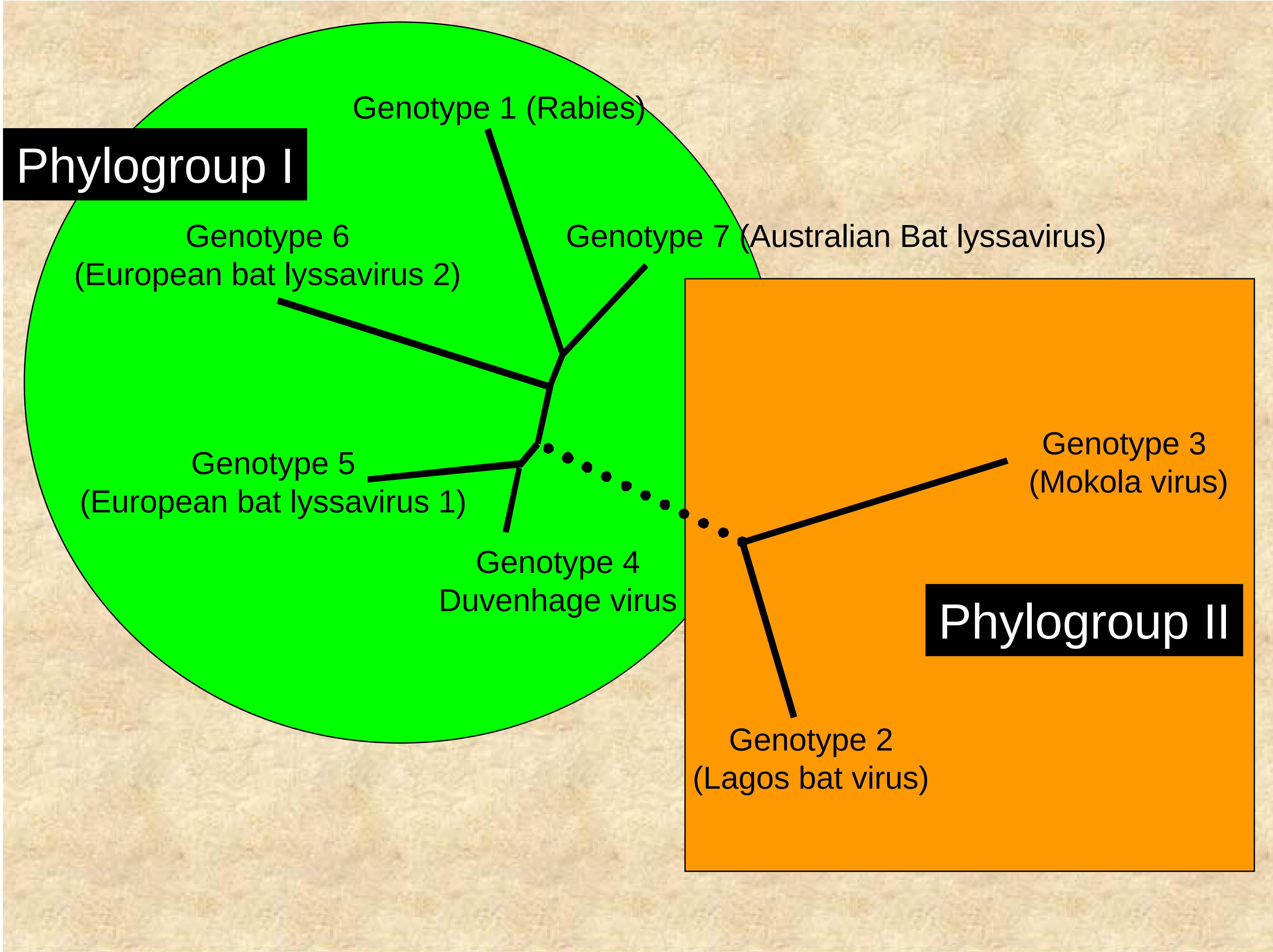
An overview and recent discoveries

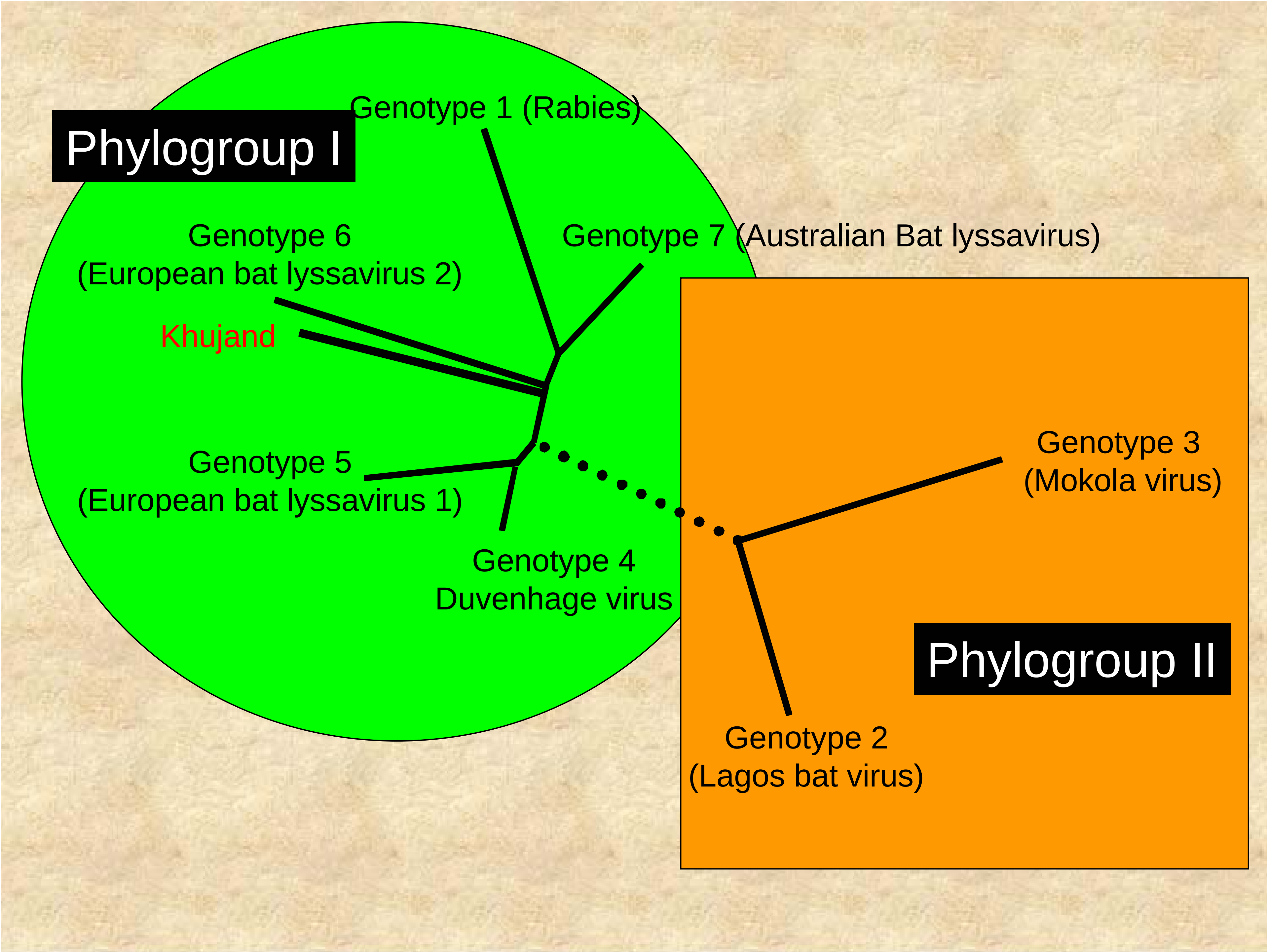
Wanda Markotter

PhD candidate
University of Pretoria
South Africa

University of Pretoria


Genotype 1 (Rabies)
Genotype 6
(European bat lyssavirus 2)
Genotype 7 (Australian Bat lyssavirus)
Genotype 5
(European bat lyssavirus 1)
Genotype 3
(Mokola virus)
Genotype 4
Duvenhage virus
Genotype 2
(Lagos bat virus)


Genotype 1 (Rabies)
Phylogroup I
Genotype 6
(European bat lyssavirus 2)
Genotype 7 (Australian Bat lyssavirus)
Genotype 5
(European bat lyssavirus 1)
Genotype 4
Duvenhage virus
Genotype 3
(Mokola virus)
Phylogroup II
Genotype 2
(Lagos bat virus)


Phylogroup I
Genotype 1 (Rabies)
Genotype 6
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Khujand
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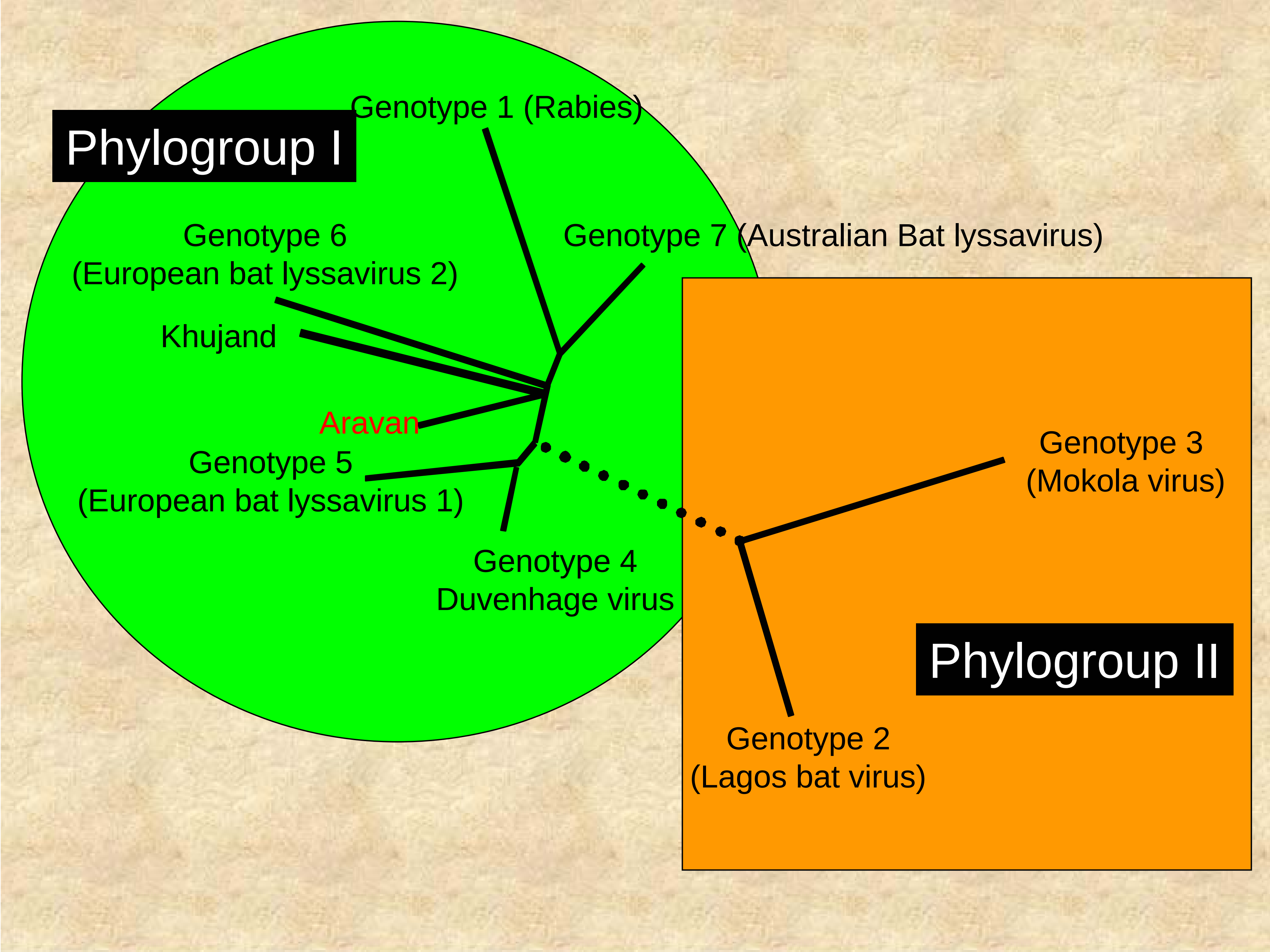

Phylogroup I
Genotype 1 (Rabies)
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(European bat lyssavirus 2)
Genotype 7 (Australian Bat lyssavirus)
Khujand
Aravan
Genotype 5
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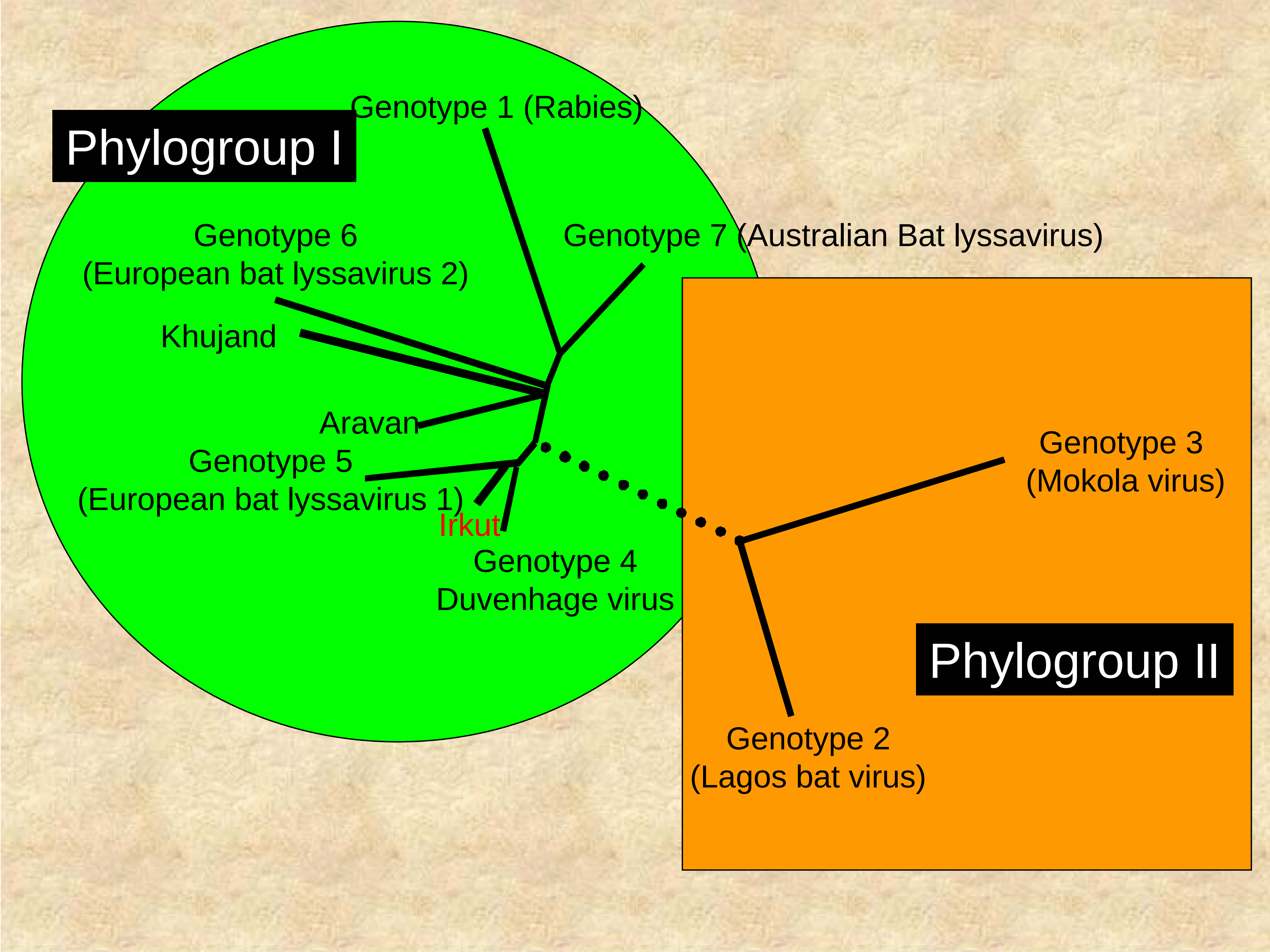
Phylogroup II
Genotype 2
(Lagos bat virus)


Phylogroup I
Genotype 1 (Rabies)
Genotype 6
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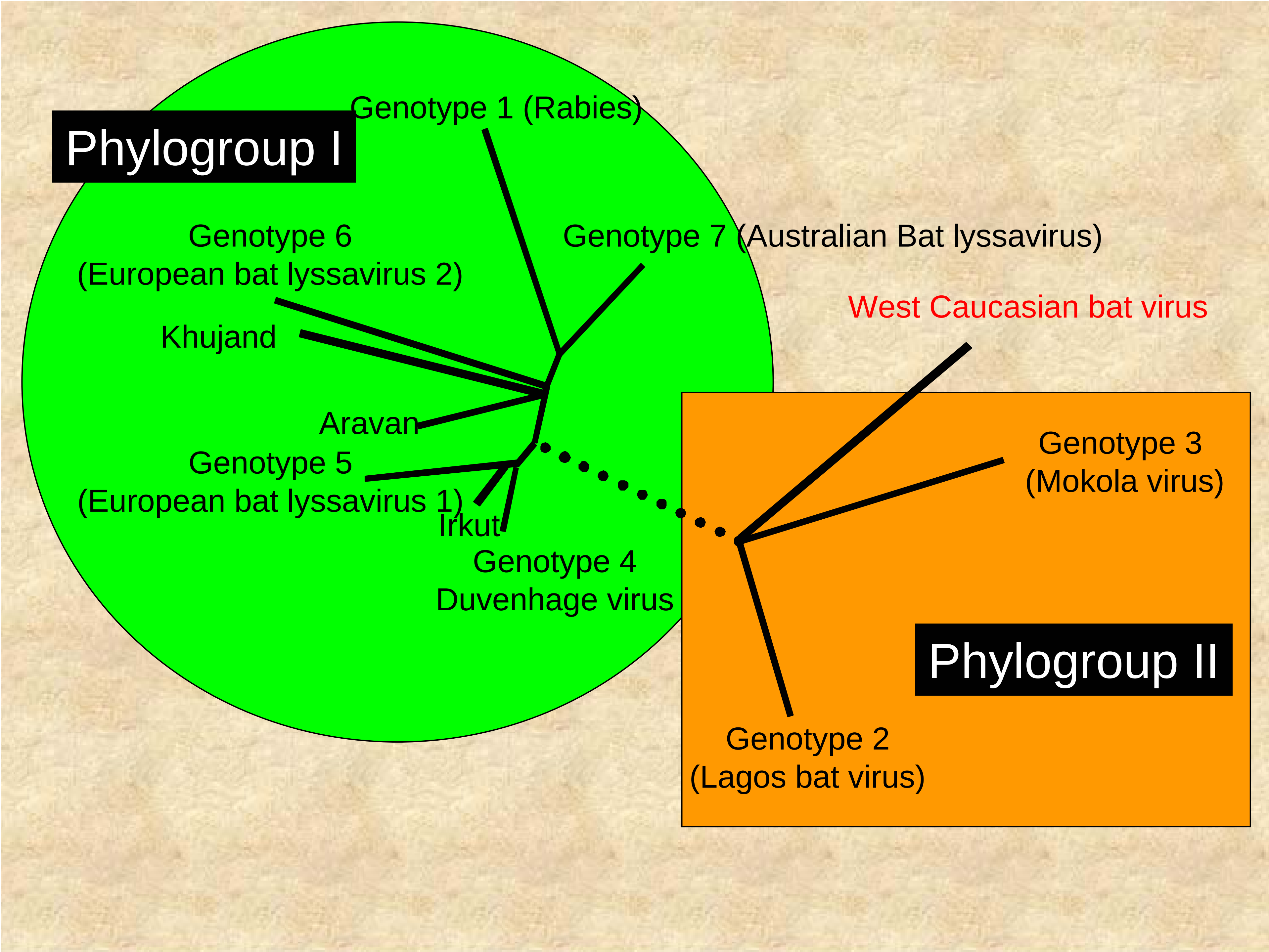
Genotype 7 (Australian Bat lyssavirus)
Khujand
Aravan
Genotype 5
(European bat lyssavirus 1)
Irkut
Genotype 4
Duvenhage virus
Genotype 3
(Mokola virus)
Phylogroup II
Genotype 2
(Lagos bat virus)


Phylogroup I
Genotype 1 (Rabies)
Genotype 6
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Genotype 7 (Australian Bat lyssavirus)
West Caucasian bat virus
Khujand
Aravan
Genotype 5
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Irkut
Genotype 4
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Genotype 3
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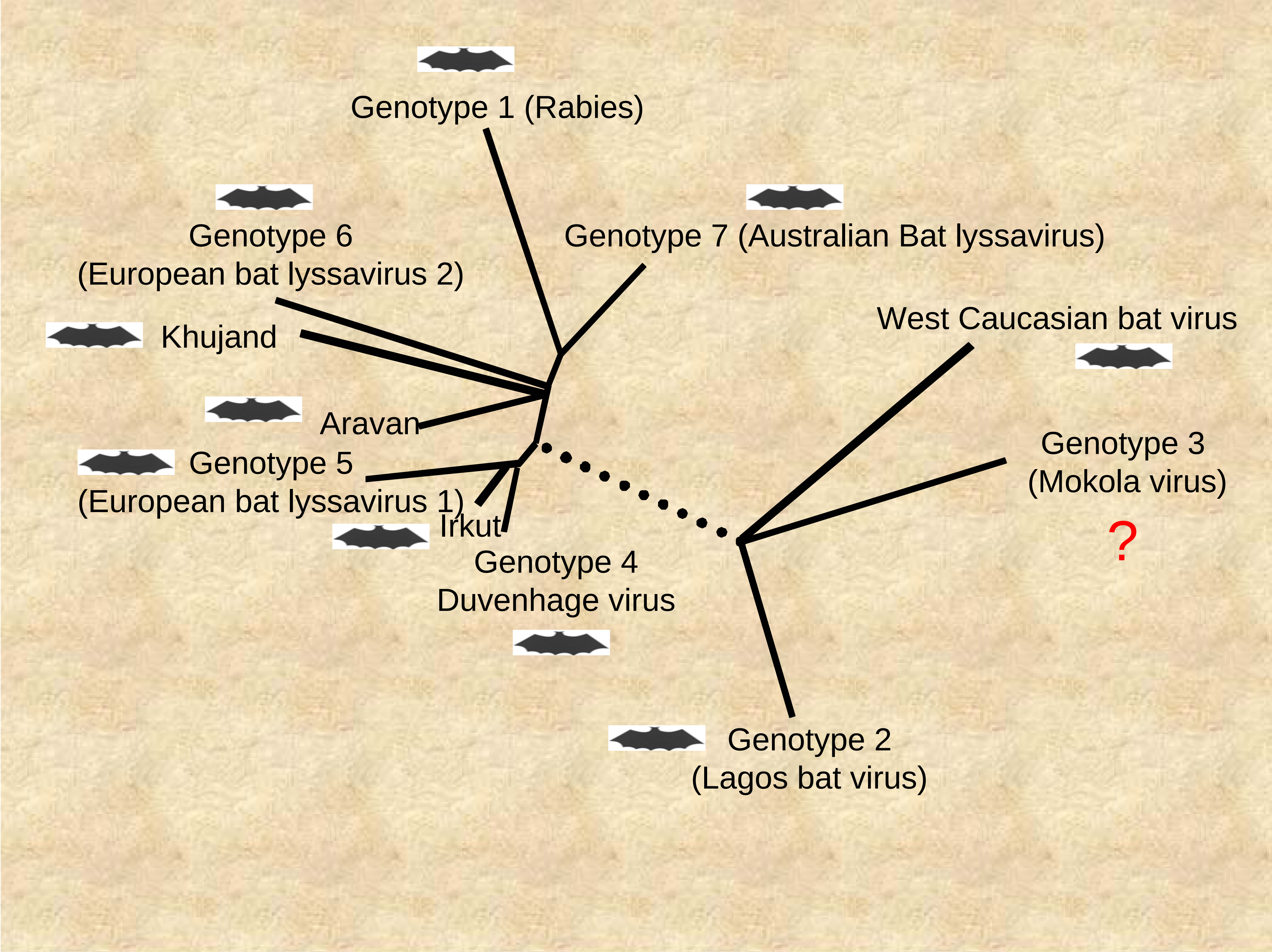
Phylogroup II
Genotype 2
(Lagos bat virus)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Irkut
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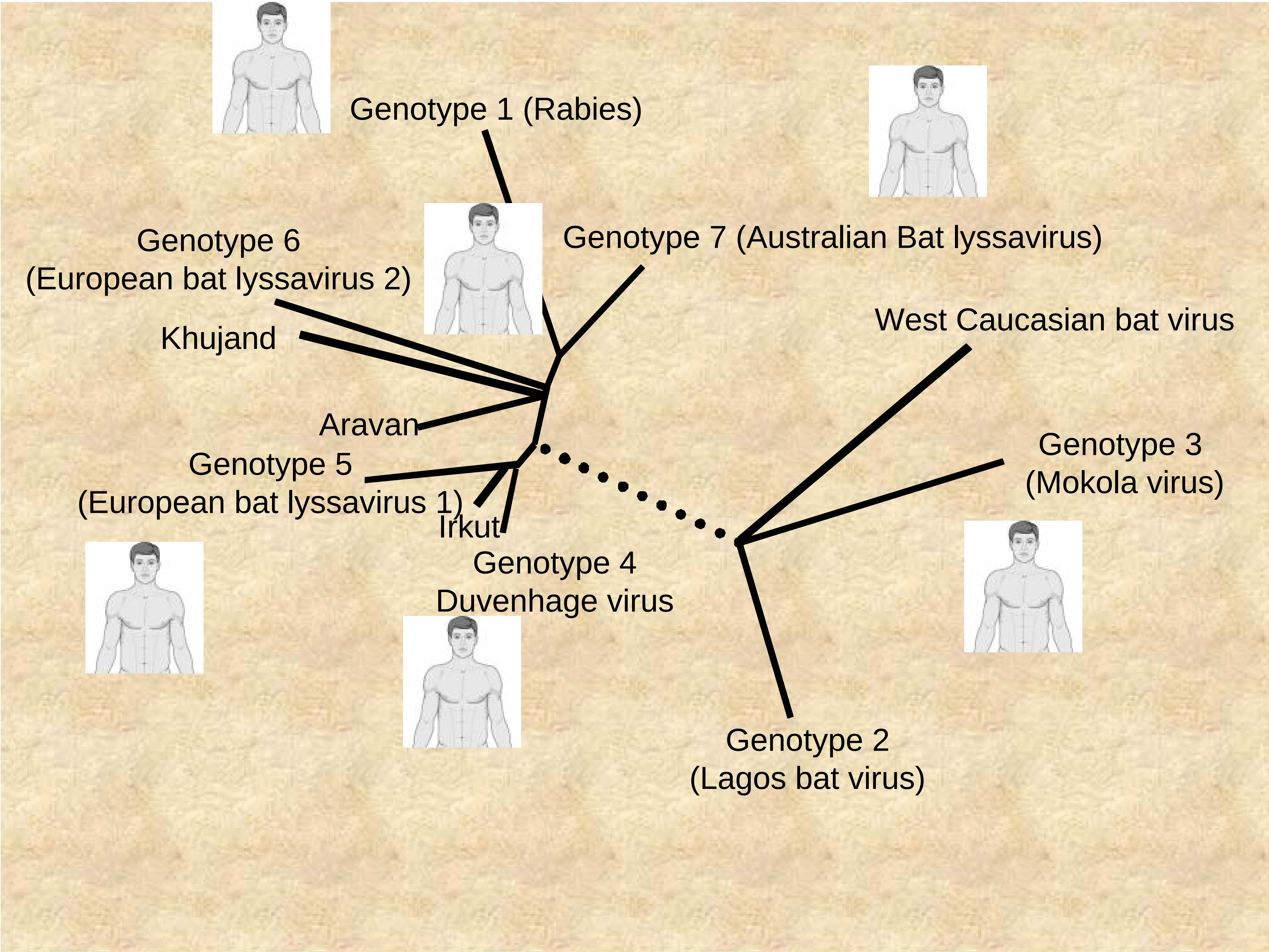
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Duvenhage virus
Genotype 2
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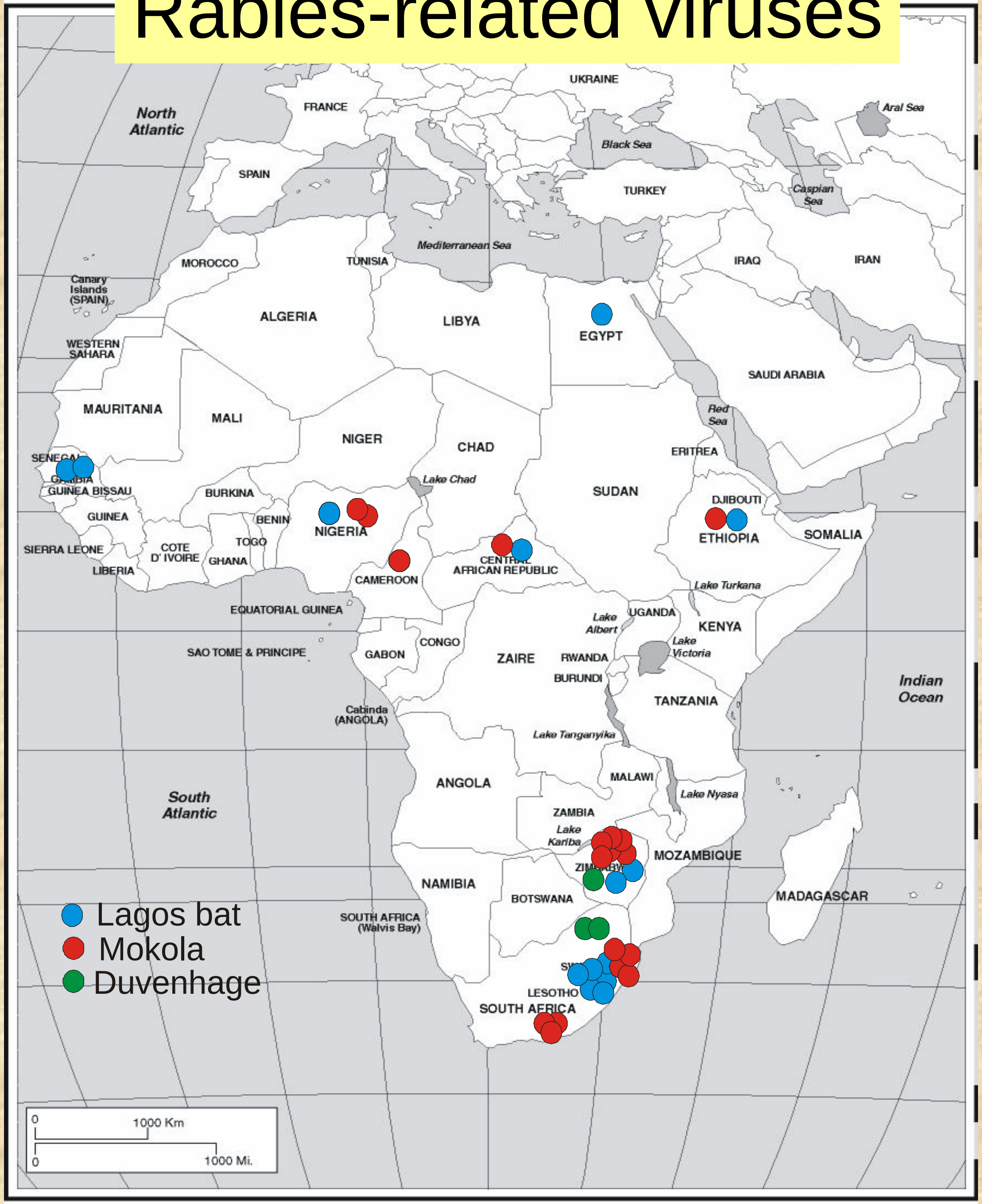
Rabies-related viruses



Produced by the Cartographic Research Lab
University of Alabama

South Africa – Rabies related viruses

Surveillance in South Africa

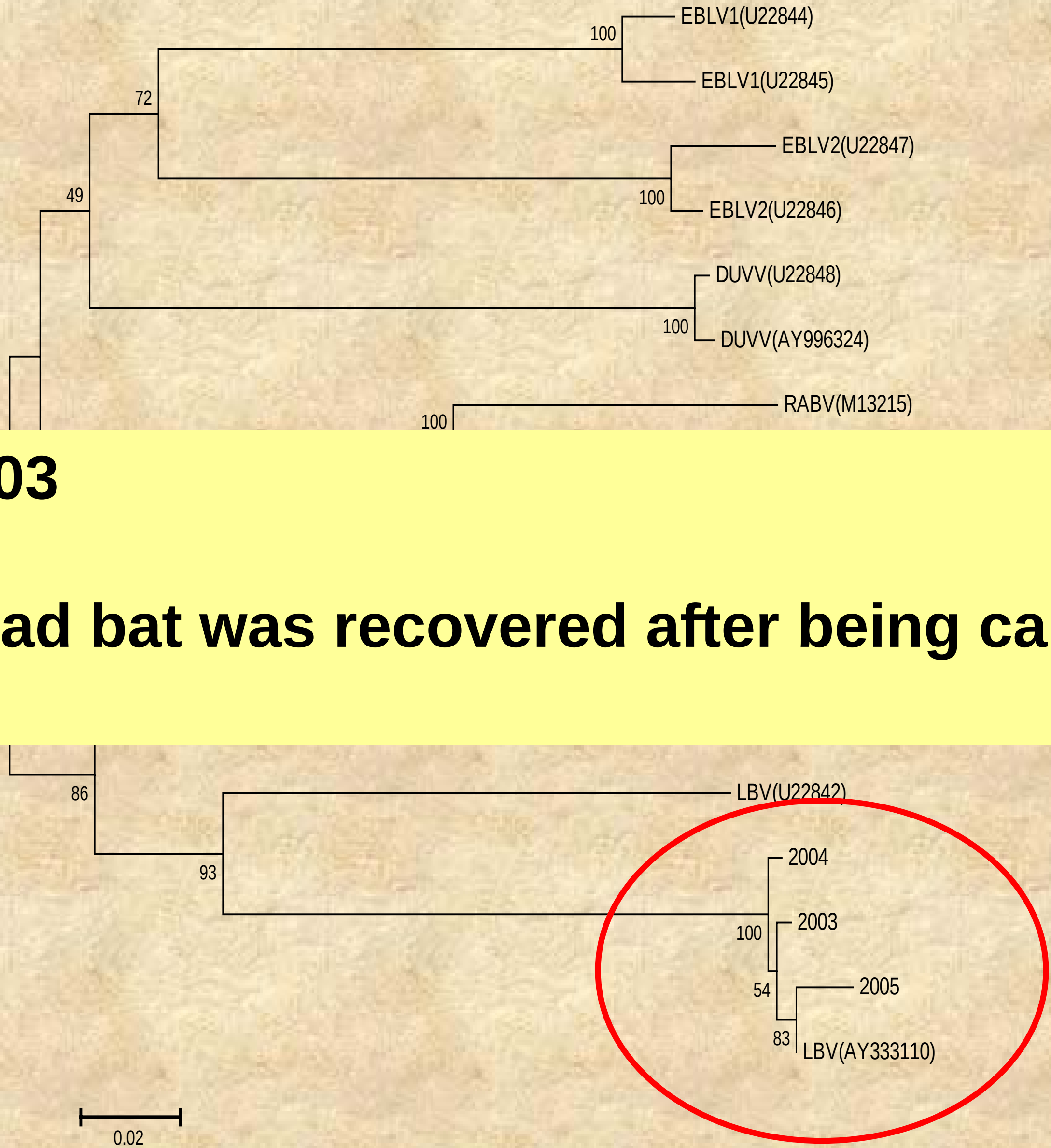
- Started with a passive surveillance program in 2003

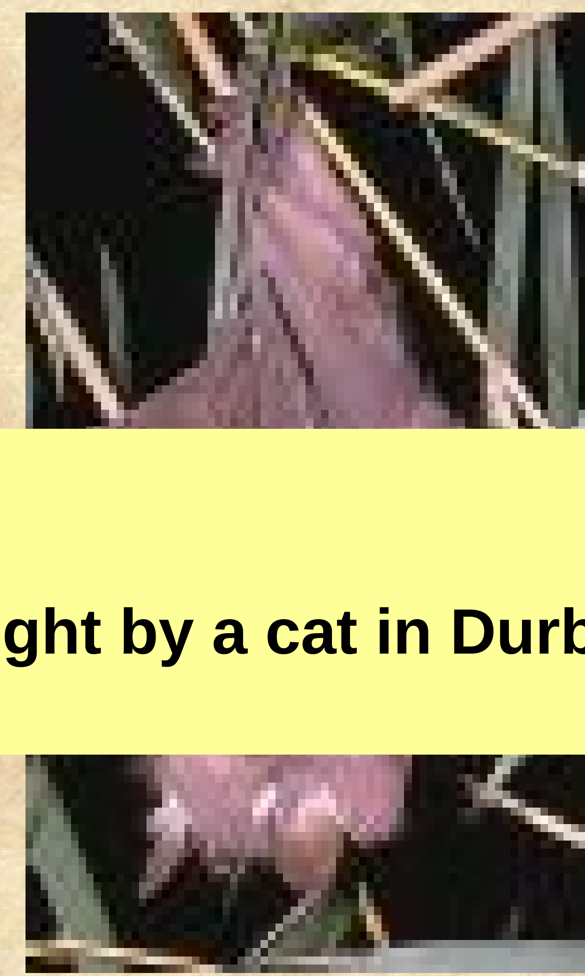
- Involved the following groups:
- Bat rehabilitators
- Bat interest group of Kwazulu Natal
- Durban Science museum

Surveillance in South Africa

- Three new cases of Lagos bat virus in bats
- First Lagos bat virus identified in a mongoose
- Lagos bat virus in a vaccinated dog

LBV bat isolates

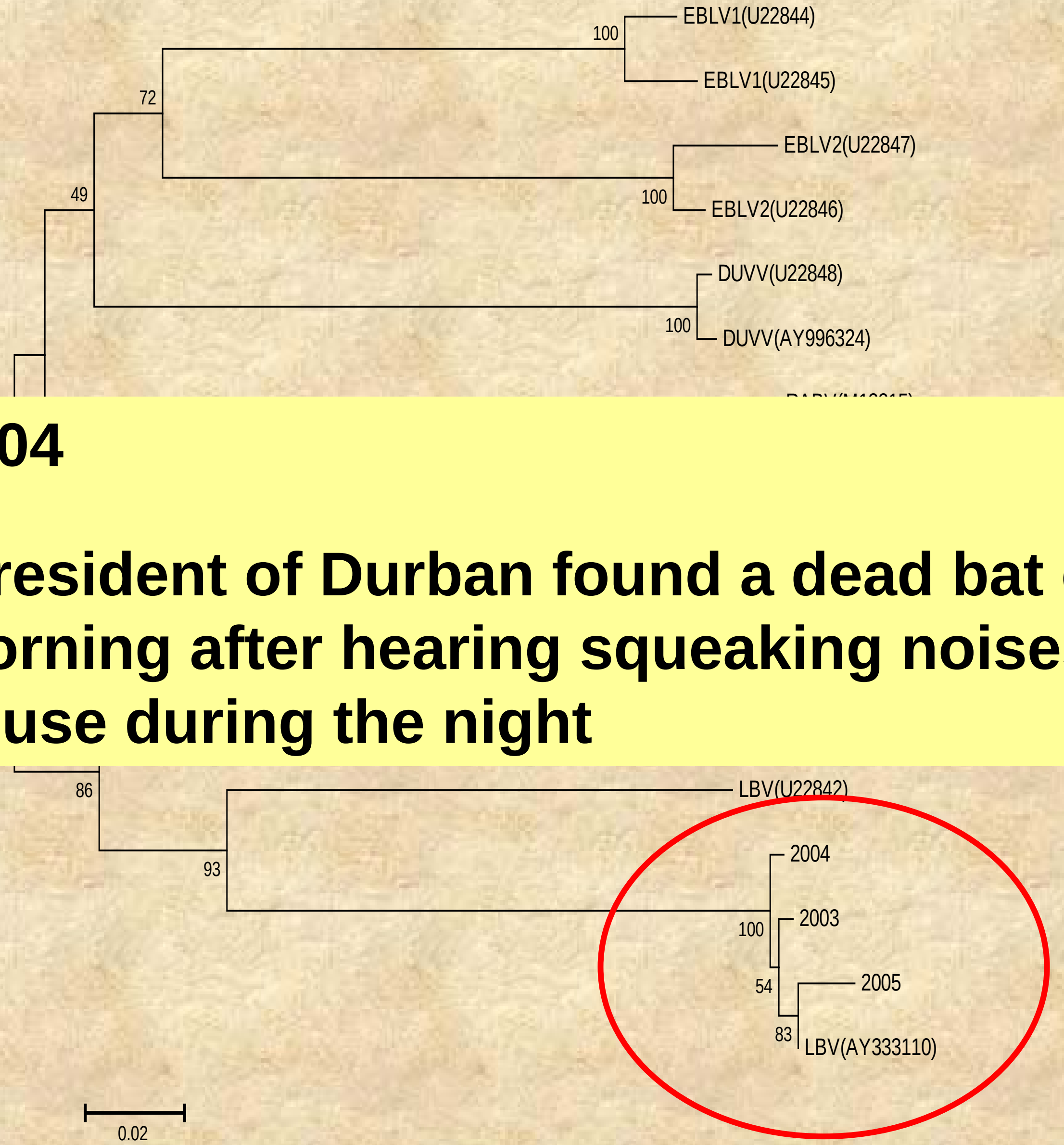


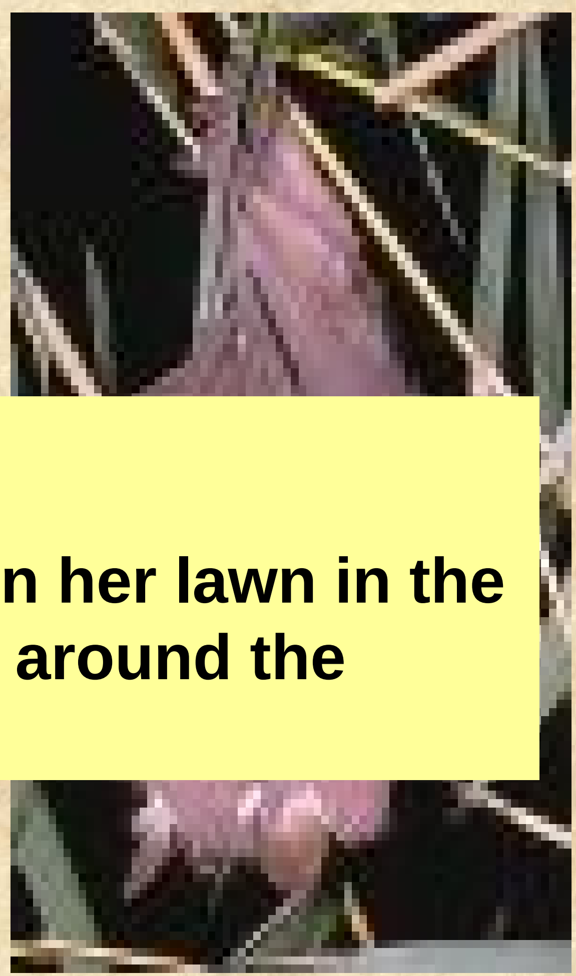


2003

Dead bat was recovered after being caught by a cat in Durban

LBV bat isolates

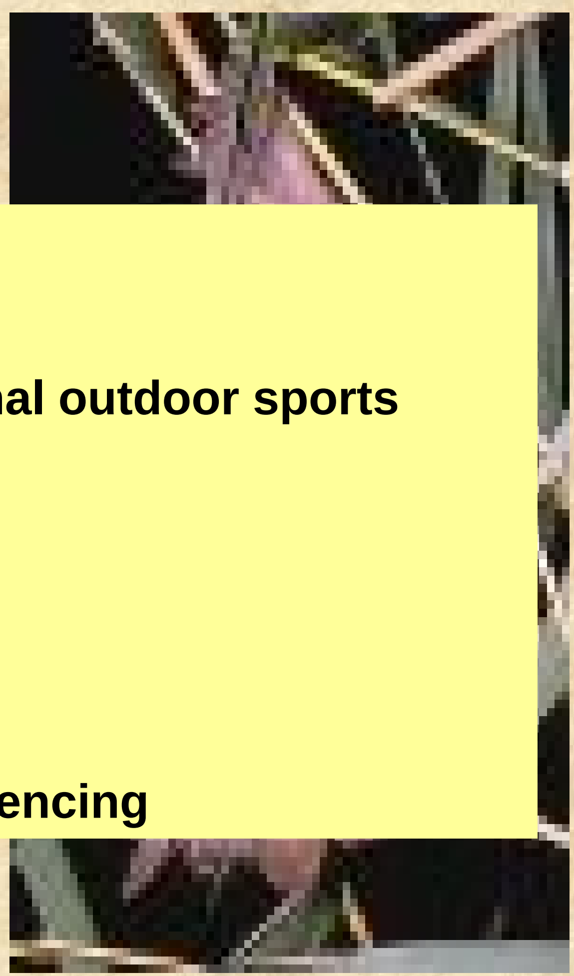




2004

A resident of Durban found a dead bat on her lawn in the morning after hearing squeaking noises around the house during the night

LBV bat isolates



2005

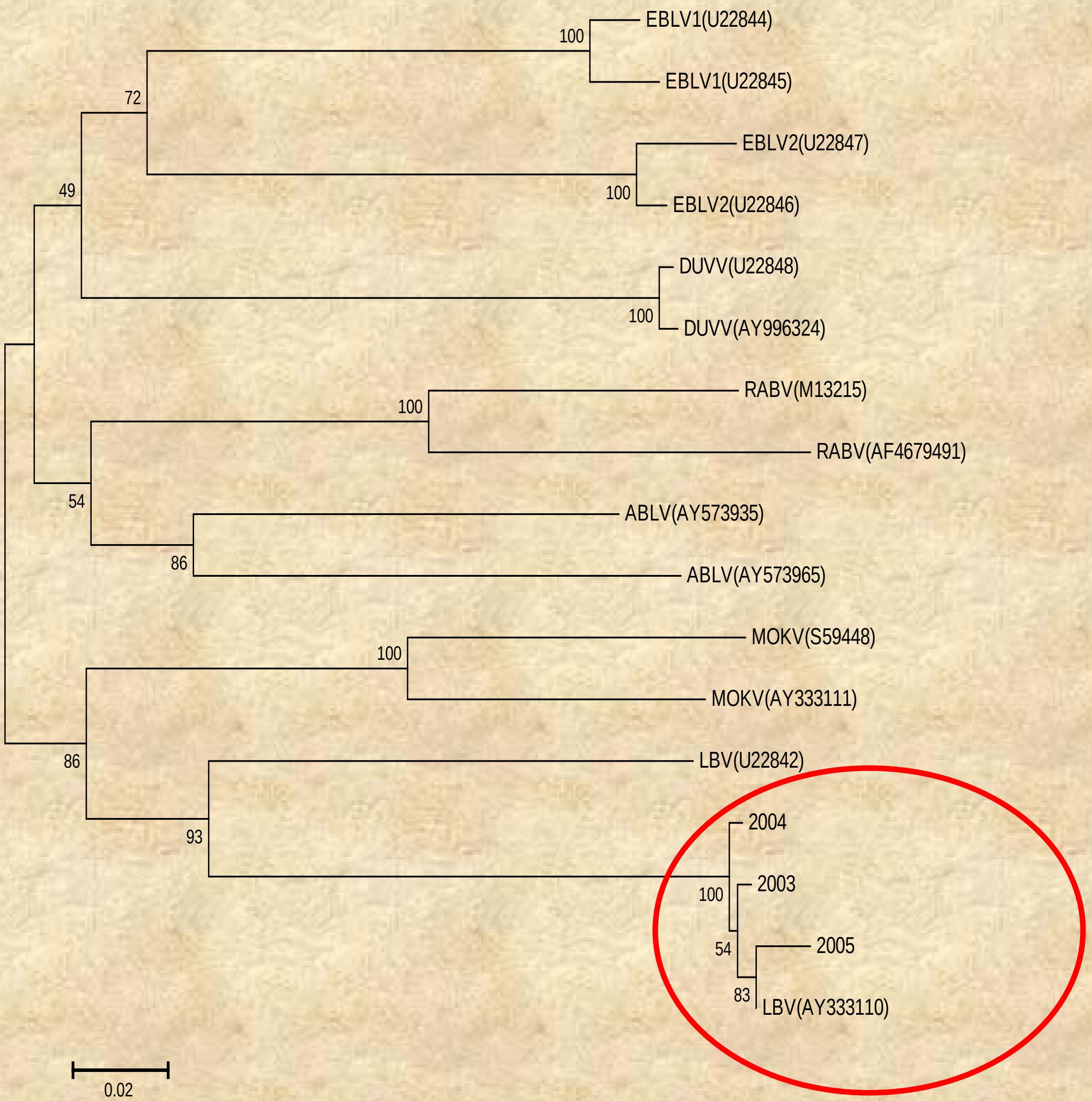
Caretaker found a bat with pup attached at a communal outdoor sports complex

Cat was playing with it

Mum died, pup survived but died a few days later

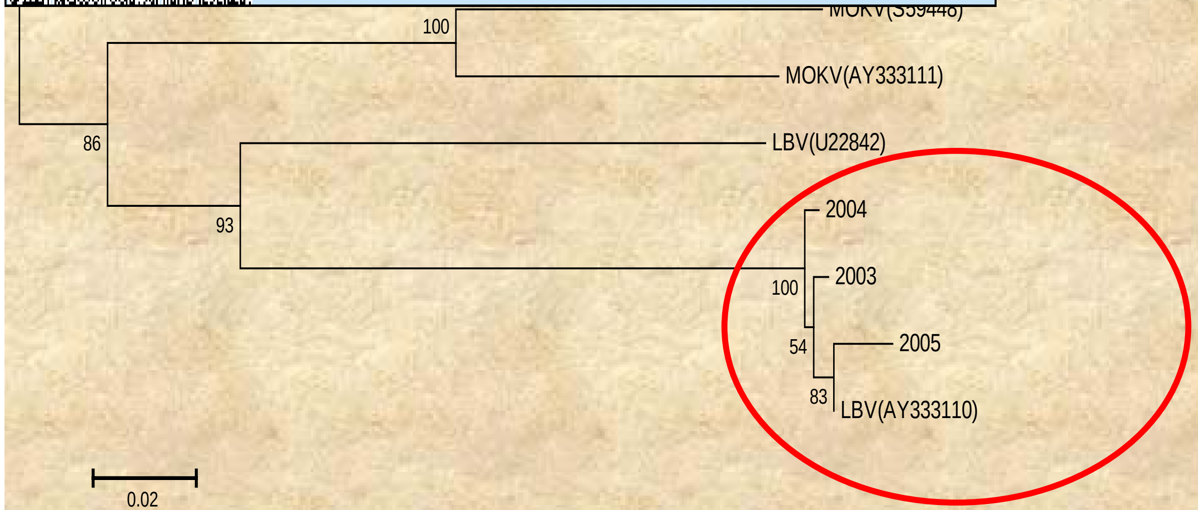
Virus was only identified through PCR and DNA sequencing

LBV isolates from bats

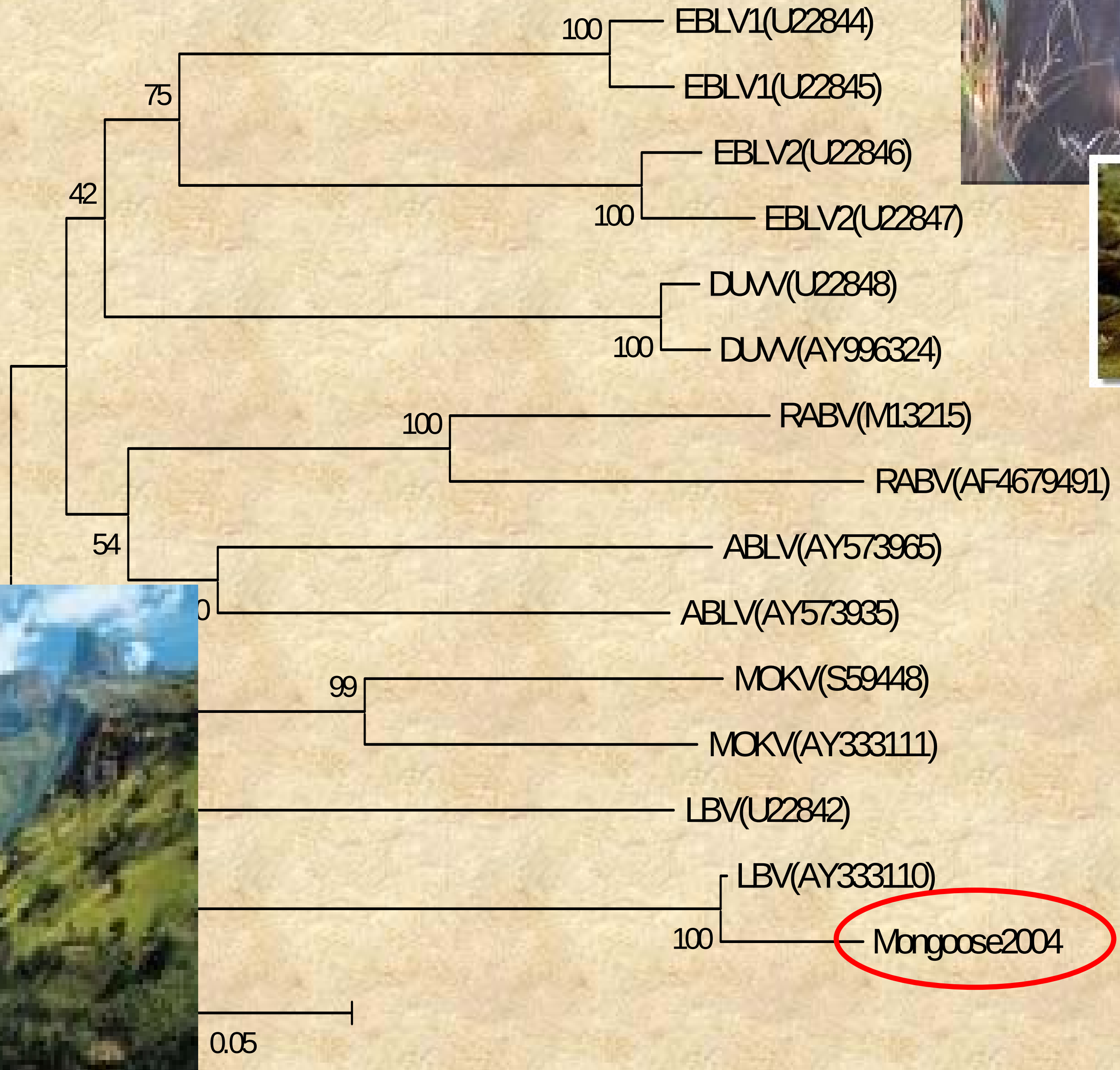


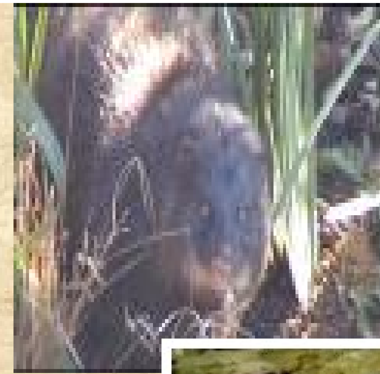
isolates

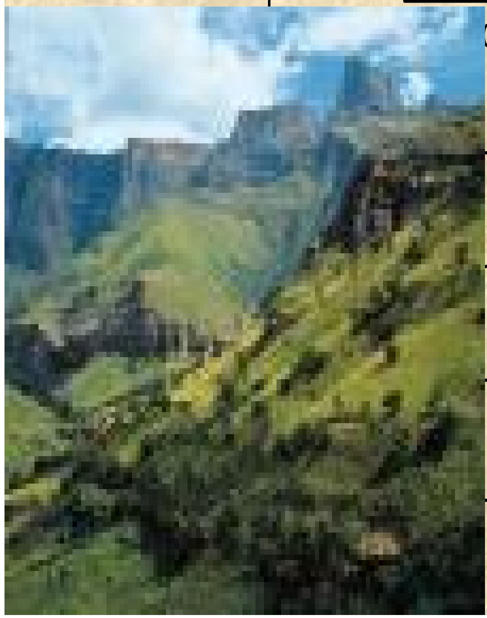
679491)



LBV Mongoose isolate







LBV Mongoose isolate

2004

Mongoose was captured by on of Cruelty to animals (SPCA)

Waterland in a residential area in Westville near Durban, KwaZulu-Natal

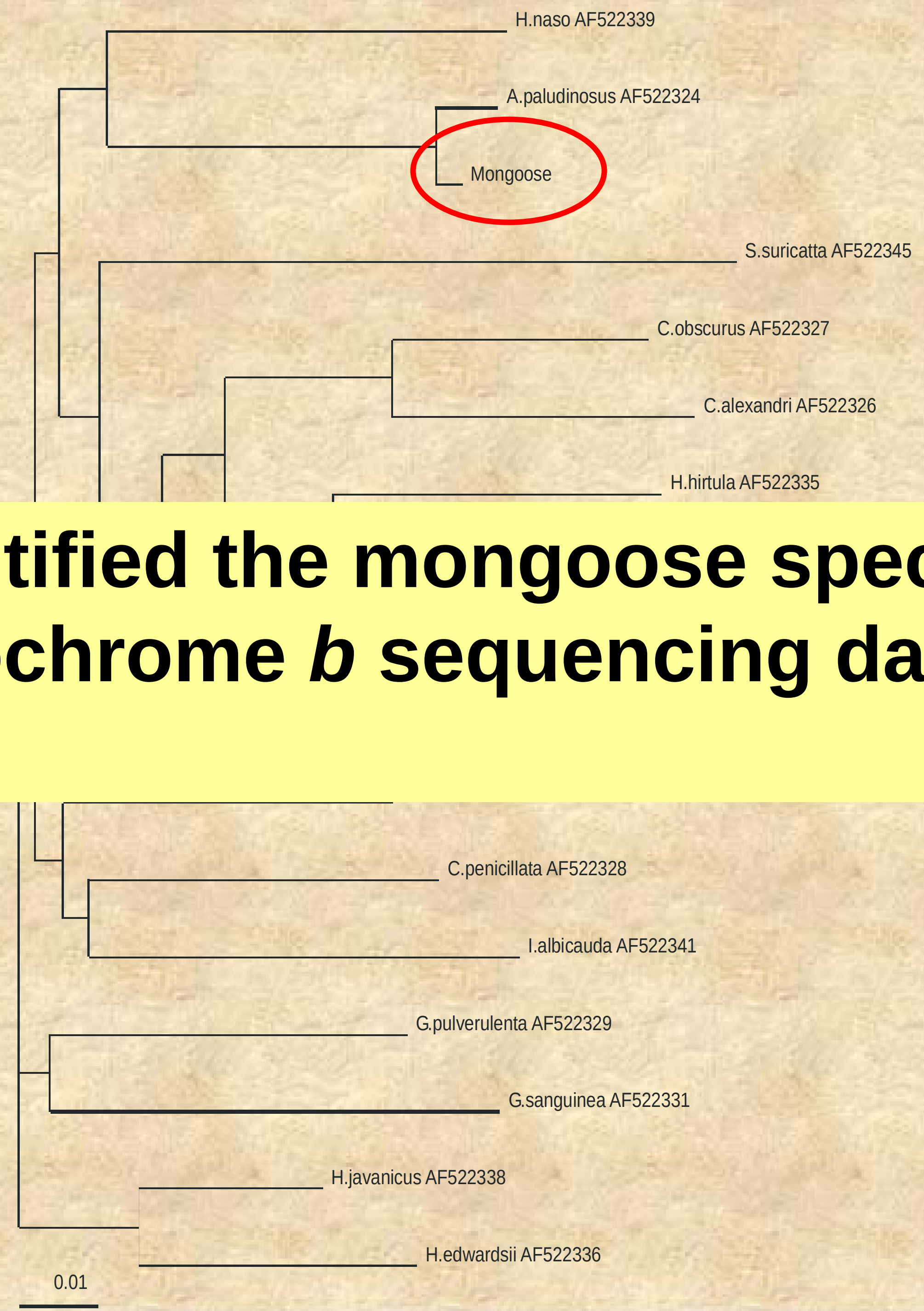
Mongoose was behaving strangely.

It was disorientated, attacking inanimate objects and its behavior alternate between friendly and aggressive.

Only the brain was submitted for testing and the carcass wasn't preserved.

The mongoose specie was subsequently not identified.

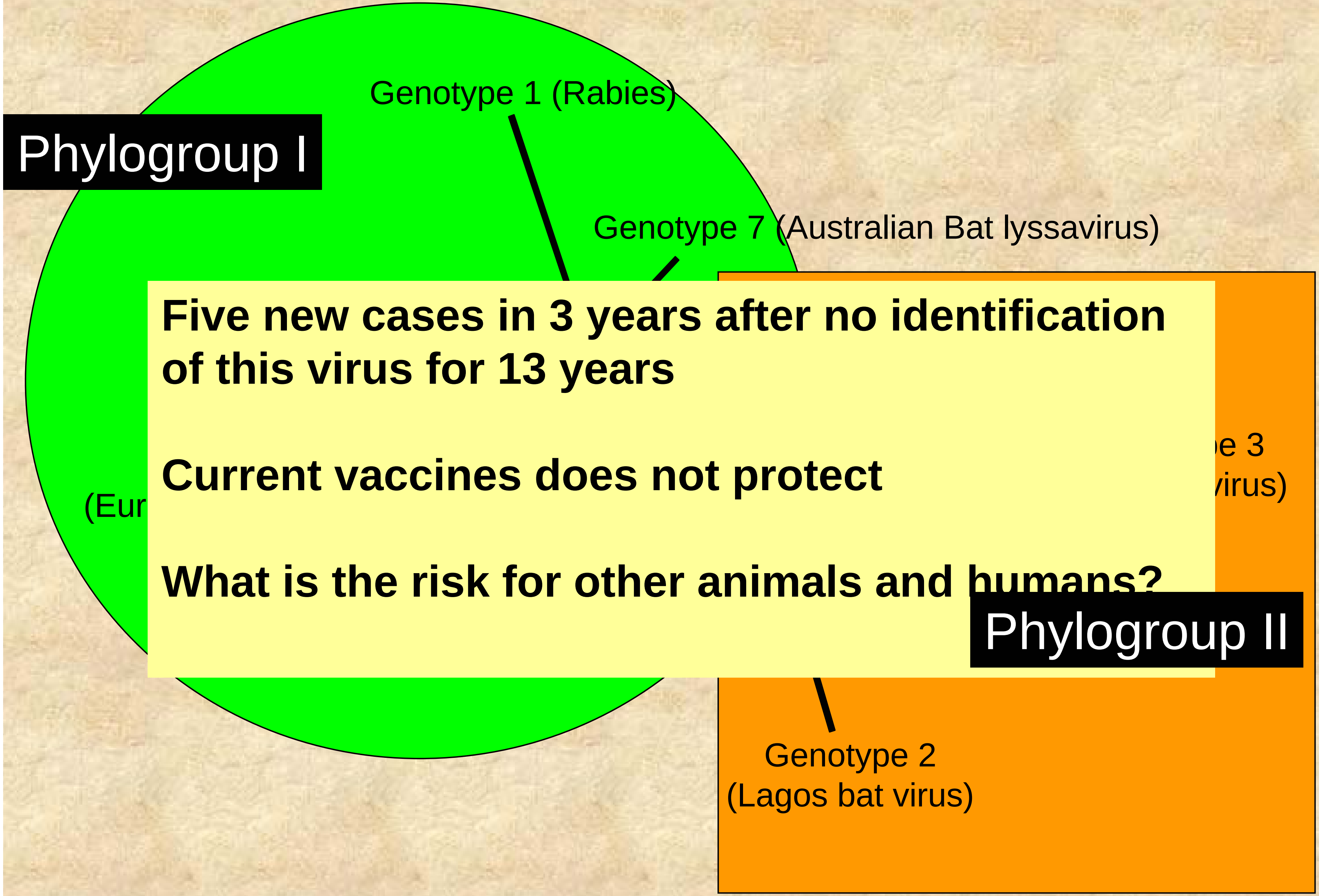
Lagos bat virus was identified



Identified the mongoose species by using cytochrome *b* sequencing data

Vaccine failure in a dog

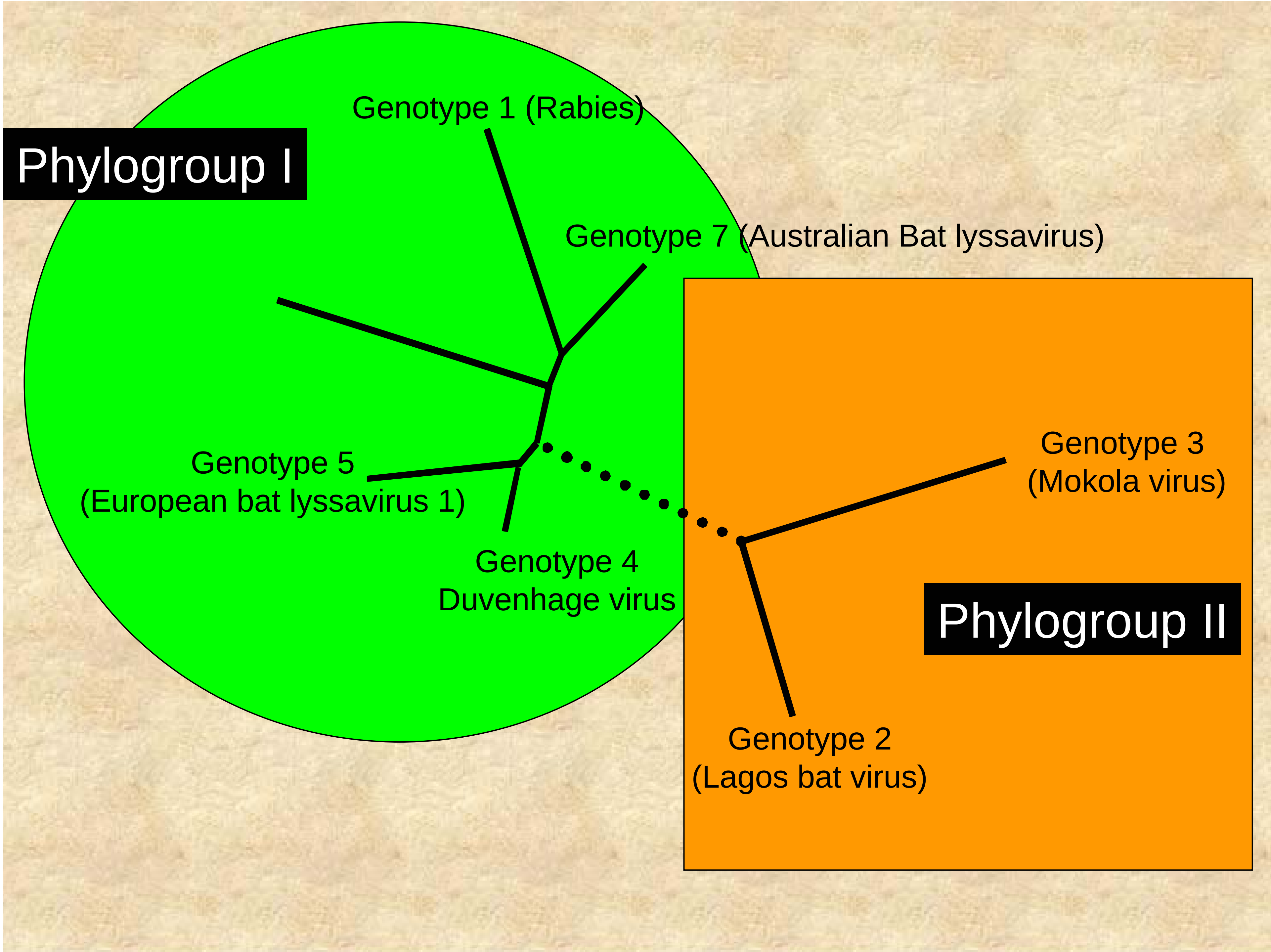
- In 2003 a dog died of rabies symptoms in the Kwazulu Natal province
- The dog had a vaccination record
- Family pet
- DNA Sequence analysis indicated that it was infected with Lagos bat virus



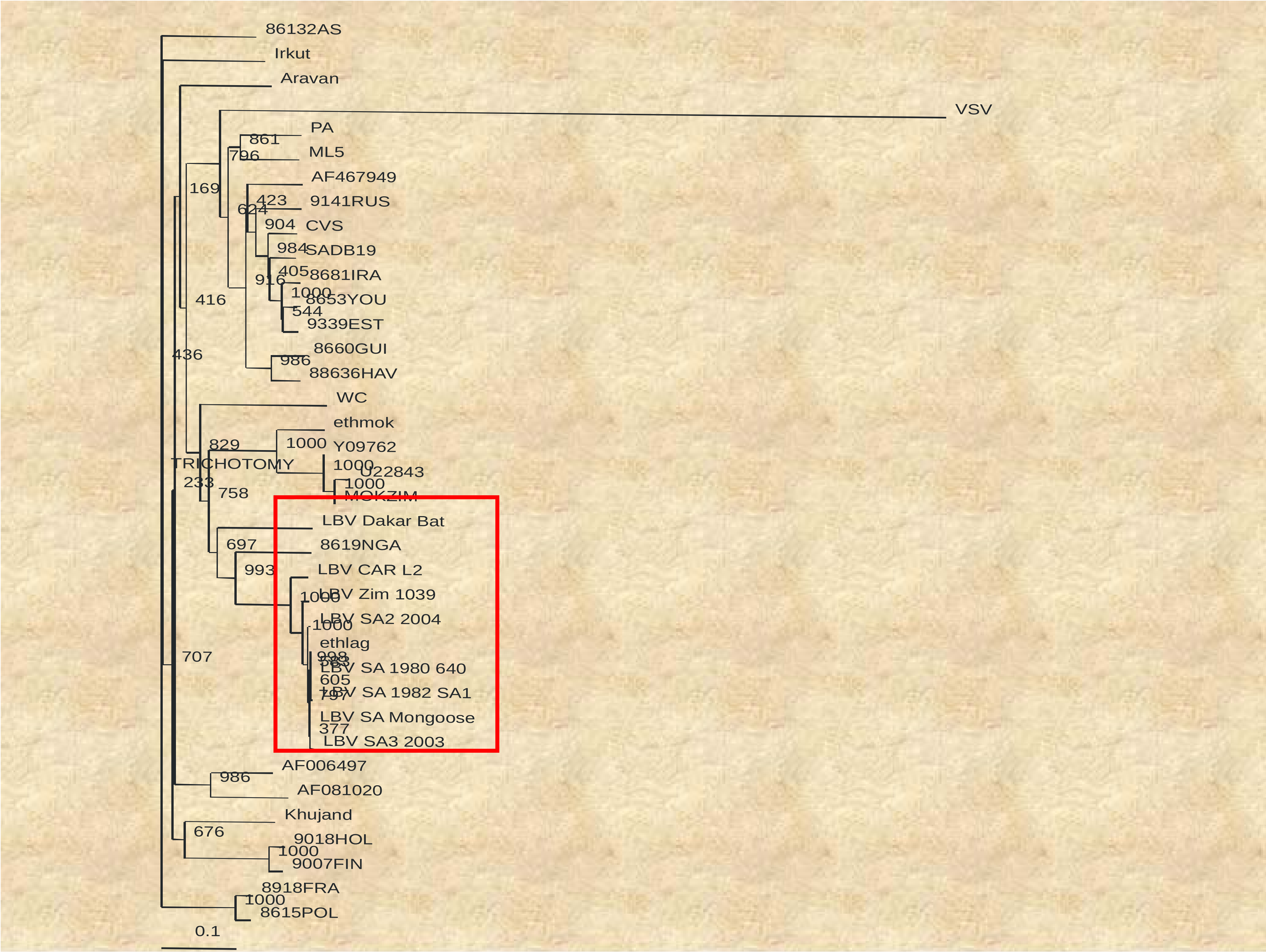
Five new cases in 3 years after no identification of this virus for 13 years

Current vaccines does not protect

What is the risk for other animals and humans?

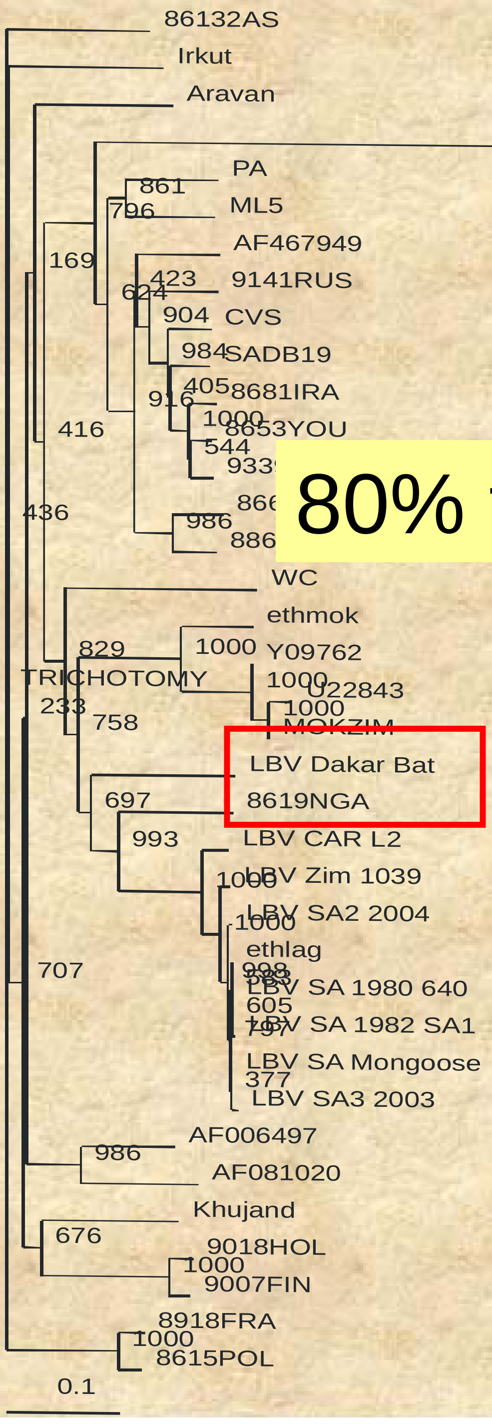

Phylogroup I
Genotype 1 (Rabies)
Genotype 7 (Australian Bat lyssavirus)
Genotype 5
(European bat lyssavirus 1)
Genotype 4
Duvenhage virus
Genotype 3
(Mokola virus)
Phylogroup II
Genotype 2
(Lagos bat virus)

Egypt
1999
(2)
Senegal
1985 (Two isolates)
(1)
Nigeria
1956
(4)
Central African Republic
1974
(15)
Ethiopia
1992
(9)
Zimbabwe
1986
(8)
1980(3 isolates) (10,5)
1982 (5)
South Africa
1990 (3)


86132AS
Irkut
Aravan
VSV
PA
861
ML5
796
AF467949
169
423
9141RUS
624
904
CVS
984
SADB19
405
8681IRA
916
1000
8653YOU
416
544
9339EST
8660GUI
436
986
88636HAV
WC
ethmok
829
1000
Y09762
TRICHOTOMY
1000
U22843
233
1000
758
MOKZIM
LBV Dakar Bat
697
8619NGA
993
LBV CAR L2
1000
LBV Zim 1039
1000
LBV SA2 2004
ethlag
707
998
LBV SA 1980 640
605
LBV SA 1982 SA1
797
LBV SA Mongoose
377
LBV SA3 2003
AF006497
986
AF081020
Khujand
676
9018HOL
1000
9007FIN
8918FRA
1000
8615POL
0.1

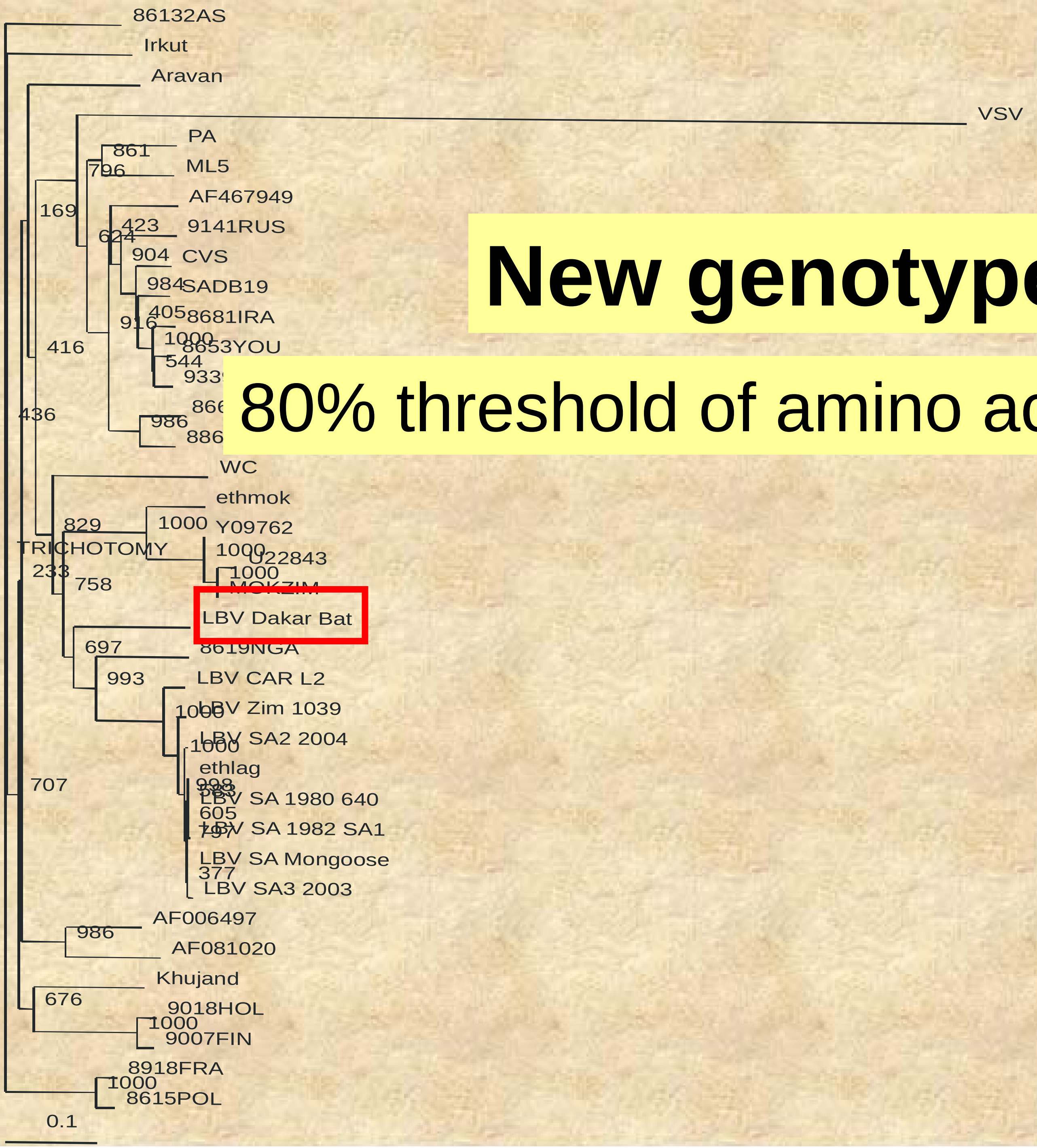
New genotype:

80% threshold of amino acid identity



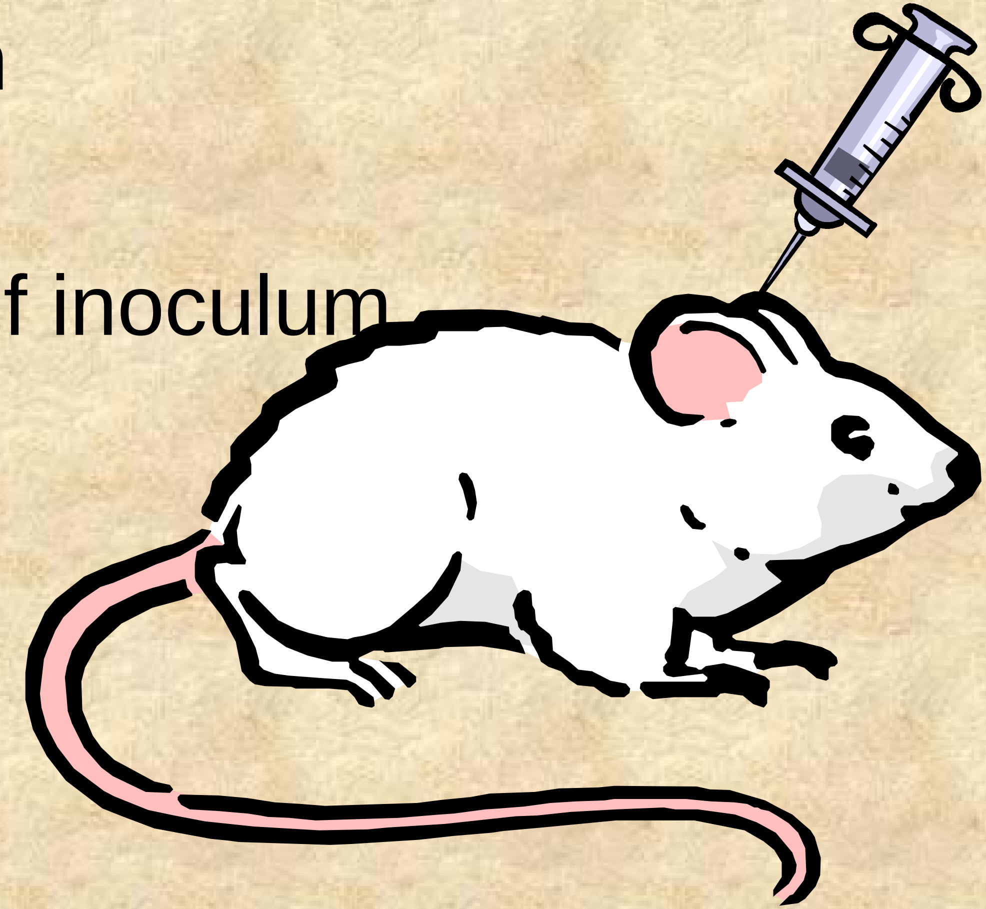
New genotype:

80% threshold of amino acid identity



Pathogenesis of Lagos bat virus

- Route of inoculation
- Effect of the Dose of inoculum



Comparison of dose of inoculum vs. route of inoculation

	Lagos Bat SA2003	Lagos Bat Dakar	Lagos Bat SA2004	Lagos Bat SA1982	Lagos Bat CAR	Lagos Bat Nigeria	Lagos Bat Zimcat	Mokola MOKSA2	Bat variant (GT1)	Mongoose SA 2004
Virus titre (LD50)	5.1	7.1	4.3	5.5	5.3	2.5	4.9	4.1	5.1	5.1
Intracerebral inoculation: 1000 LD 50 Dose	(5/5)	(5/5)	(5/5)	(4/5)	(5/5)	(2/5)	(5/5)	(5/5)	5/5	5/5
Intracerebral inoculation: LD 50 Dose	(4/5)	(4/5)	(4/5)	(5/5)	(3/5)	(1/5)	(4/5)	(5/5)	5/5	3/5
Intracerebral inoculation: 10^{-2} LD 50 Dose	(1/5)	(4/5)	(5/5)	(3/5)	(2/5)	(0/5)	(2/5)	(5/5)	4/5	4/5
Intramuscular inoculation Maximum dose	(1/4)	(5/5)	(3/5)	(0/5)	(1/5)	0/5	(1/5)	1/5	5/5	1/5
Intramuscular inoculation: 10 000 LD 50 Dose	()0/4	(3/5)	(2/5)	(0/5)	(0/5)	0/5	(0/5)	(0/5)	1/5	1/5

Comparison of dose of inoculum vs. route of inoculation

	Lagos Bat SA2003	Lagos Bat Dakar	Lagos Bat SA2004	Lagos Bat SA1982	Lagos Bat CAR	Lagos Bat Nigeria	Lagos Bat Zimcat	Mokola MOKSA2	Bat variant (GT1)	Mongoose SA 2004
Virus titre (LD50)	5.1	7.1	4.3	5.5	5.3	2.5	4.9	4.1	5.1	5.1
Intramuscular inoculation Maximum dose	(1/4)	(5/5)	(3/5)	(0/5)	(1/5)	0/5	(1/5)	1/5	5/5	1/5
Intramuscular inoculation: 10 000 LD 50 Dose	()0/4	(3/5)	(2/5)	(0/5)	(0/5)	0/5	(0/5)	(0/5)	1/5	1/5

Lagos bat virus and Mokola virus can kill mice when inoculated intramuscularly

Comparison of dose of inoculum vs. route of inoculation

	Lagos Bat SA2003	**Lagos Bat Dakar**	**Lagos Bat SA2004**	**Lagos Bat SA1982**	**Lagos Bat CAR**	**Lagos Bat Nigeria**	**Lagos Bat Zimcat**	**Mokola MOKSA2**	**Bat variant (GT1)**	**Mongoose SA 2004**
Intramuscular inoculation: 10 000 LD 50 Dose	()0/4	(3/5)	(2/5)	(0/5)	(0/5)	0/5	(0/5)	(0/5)	1/5	1/5

Lagos bat virus and Mokola virus can kill mice when inoculated intramuscularly

Mortality is related to the dose of virus inoculums

Conclusions

- Passive surveillance in South Africa identified five new cases of rabies-related viruses in a short period.
- Surveillance and diagnostic procedures of rabies and rabies-related viruses in Africa must be improved
- Improved surveillance and molecular characterization of lyssaviruses in Africa will provide a better understanding of lyssavirus epidemiology in general
- Assessment of the pathogenesis of African lyssaviruses in animal models other than mice is needed to plan future vaccine strategies for the African continent

Acknowledgements

Prof. Louis Nel (UP)
Dr. Peter Taylor (Durban Science museum)
Dr. Charles Rupprecht (CDC)
Ivan Kuzmin (CDC)
Claude Sabeta (OVI)
Dr. Alex Wandeler (Canadian Food inspection agenc
Dr. Jenny Rhandles (Allerton Veterinary laboratory)
Bat rehabilatators from Kwazulu Natal (Wendy and Kate)
Bat interest group of Kwazulu Natal
Prof Bob Swanepoel (NICD)

Funding:
University of Pretoria
National Research foundation of South Africa
Centers for Disease control and Prevention, Rabies unit, USA

Rabies in Namibian cheetahs

Susanne Schulze
SEARG Meeting 22-26 January 2006

Namibia as a study site

- thought to be host of the largest cheetah population
- most cheetahs live on commercial farmland
- absence of other large carnivore species
- presence of domestic dogs
- high offtake of cheetahs by people

Ecology of wildlife diseases

- susceptibility

Ecology of wildlife diseases

- susceptibility

- host density

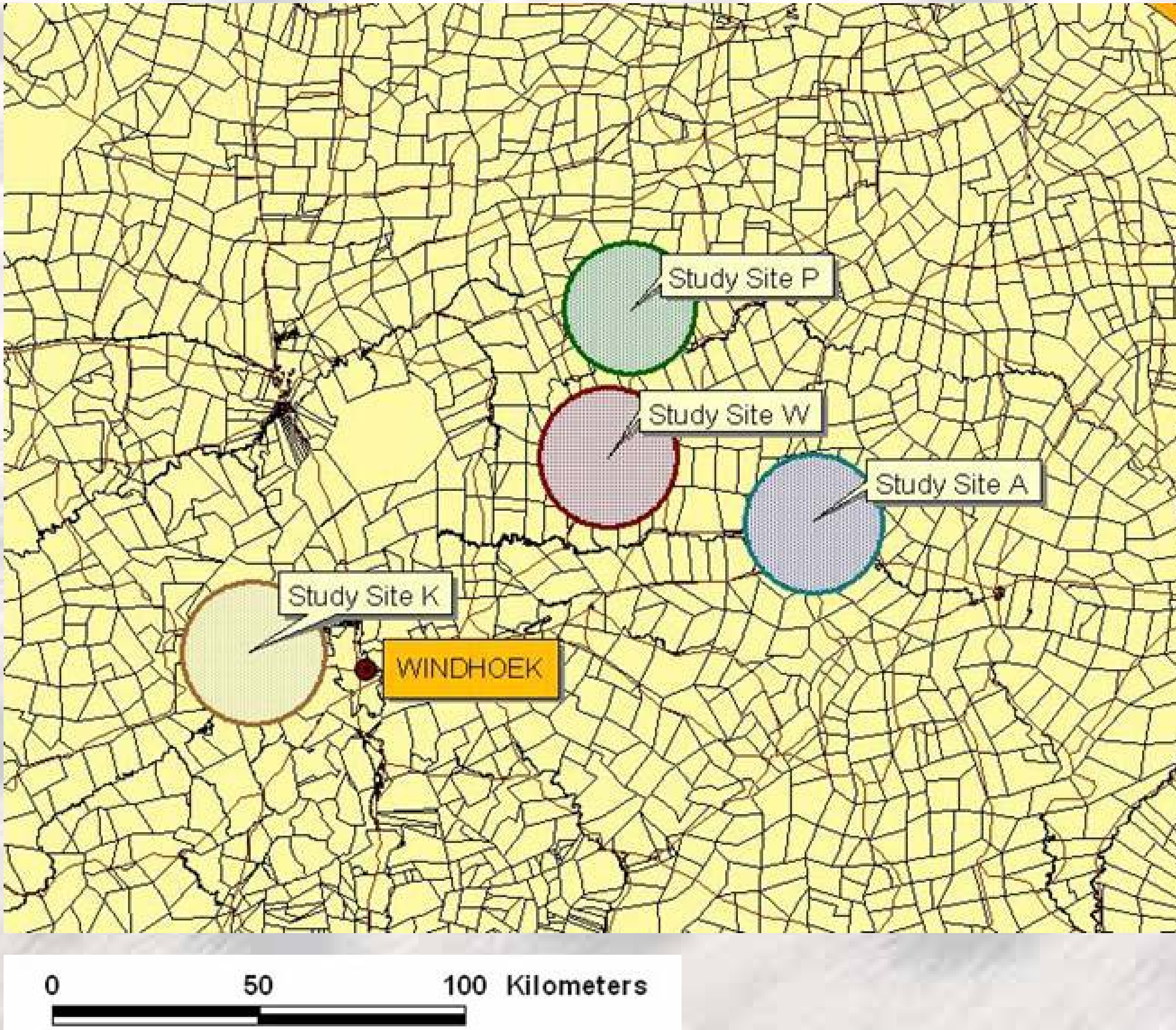
Ecology of wildlife diseases

- susceptibility
- host density
- social behaviour

Ecology of wildlife diseases

- susceptibility
- host density
- social behaviour
- reservoir hosts

project study sites



Capture and immobilisation

Immobilisation and sample collection

Material & methods

	free- ranging	captive	method
serum samples	42	12	RFFIT (rapid fluorescence inhibition test)
brain tissue	6	1	RT-PCR (reverse transcriptase polymerase chain reaction)

Results RFFIT free-ranging cheetahs

positive	negative
N=2 (0.5IU/ml)	N=40

Cheetah sightings with other carnivore species

- 52 radio-collared study animals
- 64 aerial tracking flights
- Jackal 4.6%
- Bat-eared fox 0.4%
- Domestic dog 0.0%
- Caracal 0.4%
- Leopard 0.4%

Prey species of free-ranging cheetahs

- Greater Kudu
- Hartebeest
- Oryx
- Springbok
- Duiker
- Waterbuck
- Warthog
- Suricate
- Springhare
- Treemouse

- Cattle
- Goat
- Sheep

Prey species of free-ranging cheetahs

- Greater Kudu*
- Hartebeest*
- Oryx
- Springbok*
- Duiker*
- Waterbuck
- Warthog
- Suricate*
- Springhare*
- Treemouse

- Cattle*
- Goat*
- Sheep*

Results RFFIT captive cheetahs

vaccinated		non-vaccinated	
positive	negative	positive	negative
N=6	N=0	N=0	N=6

Antibody titers in captive cheetahs

animal #	Vacc. status	Titer (IU/ml)
1	yes	0.5
2	yes	0.8
3	yes	0.5
4	yes	12.5
5	yes	104.2
6	yes	62.5

Acknowledgements

Institute for Zoo and Wildlife Research (IZW), Berlin, Germany

Prof Heribert Hofer
Dr Bettina Wachter
Dr Marion L. East and Dr Oliver Höner

Federal Research Centre for Virus diseases of Animals, Tübingen, Germany

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Friedrich- Loeffler- Institut, Wusterhausen, Germany

Dr Thomas Müller

Namibia

Seeis Conservancy and Hochfeld Conservancy
Dr Ulf Tubessing, Dr Hans-Otto Reuter, Dr Mark Jago
Okatumba Wildlife Research
The Ministry of Environment and Tourism

Messerli Foundation, Switzerland

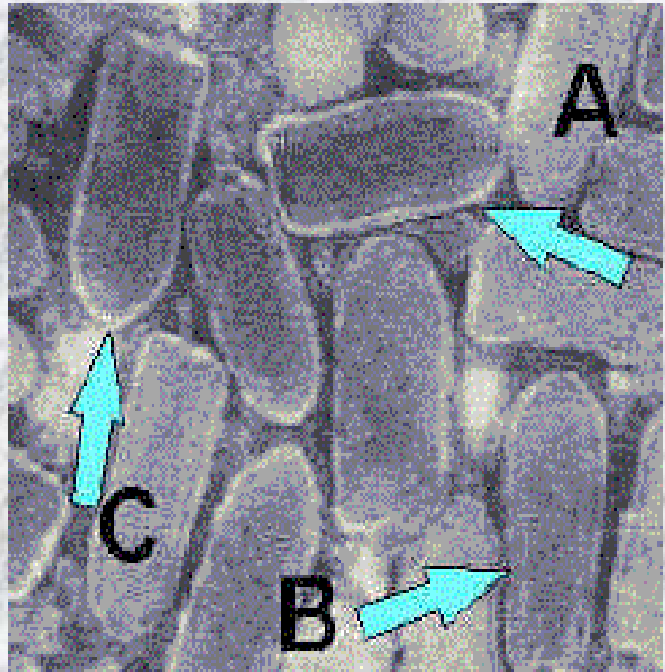
Thank you for your attention!

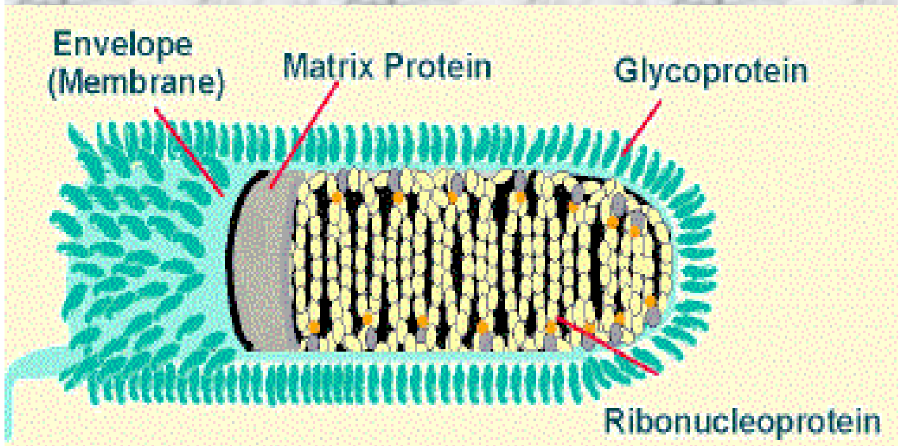
Rabies in bat-eared foxes in South Africa

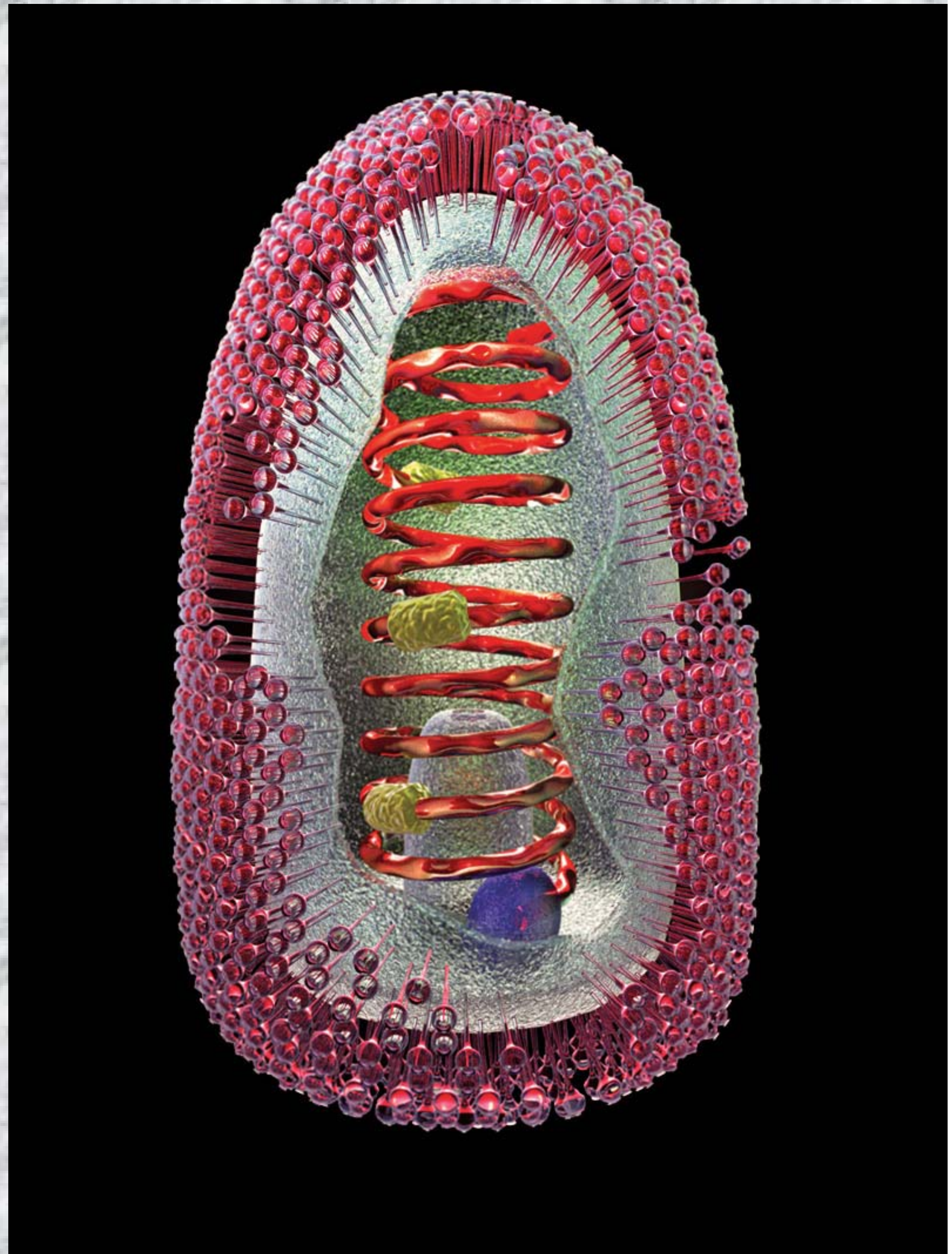
Claude Sabeta

Presentation

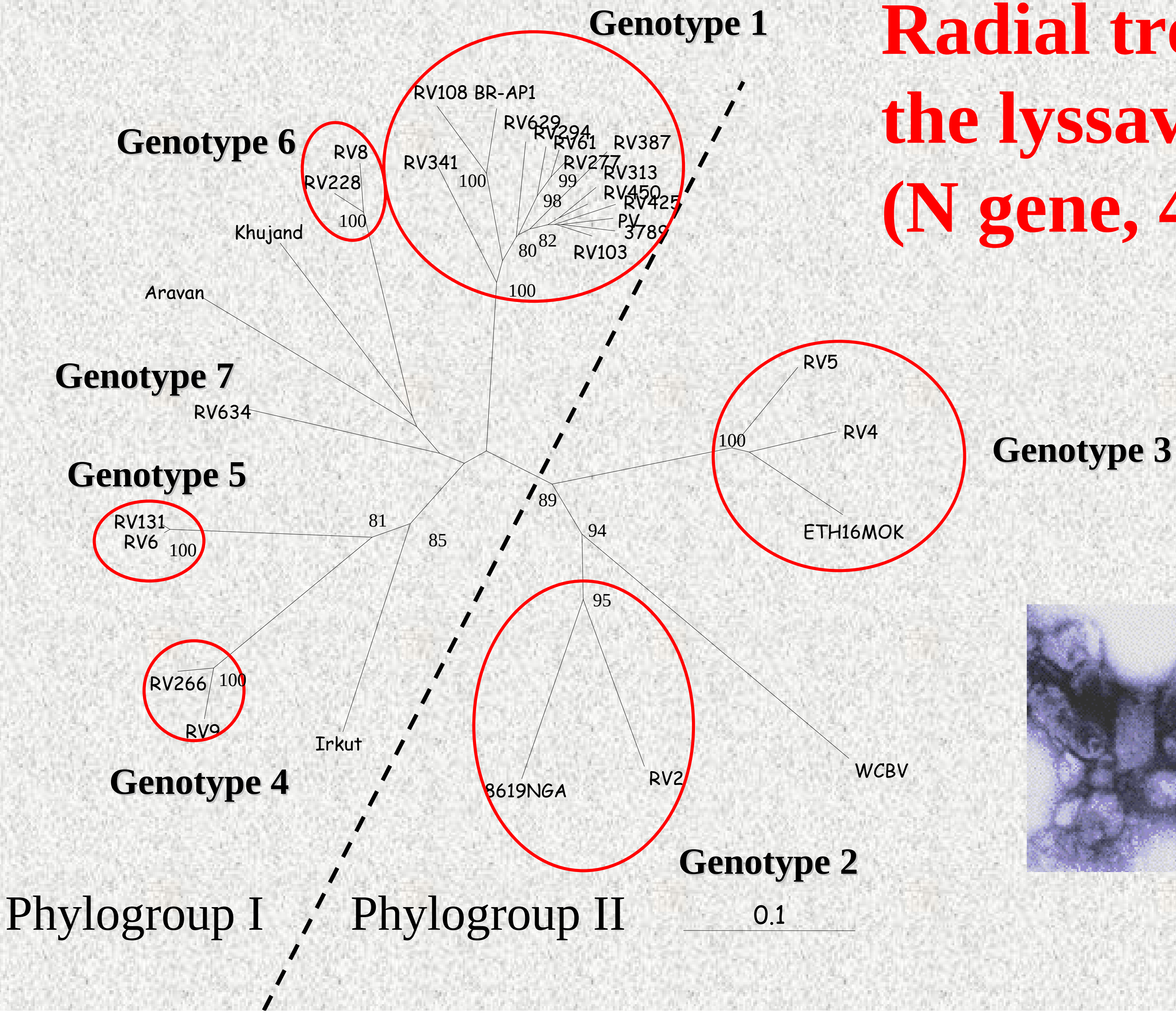
- Background to rabies in southern Africa
- Rabies in bat-eared foxes: history
- Surveillance data (1994-2004)
- Monoclonal antibody typing data
- Molecular characterisation
- Implications for disease control in wildlife

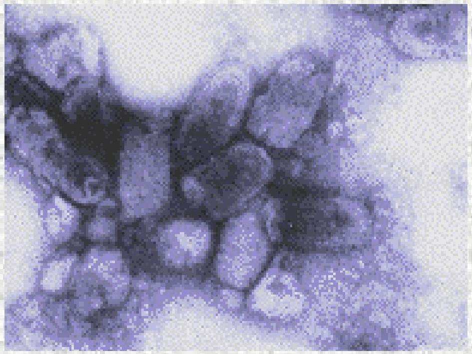

A
C
B


Envelope
(Membrane)
Matrix Protein
Glycoprotein
Ribonucleoprotein



Radial tree of the lyssaviruses (N gene, 405bp)

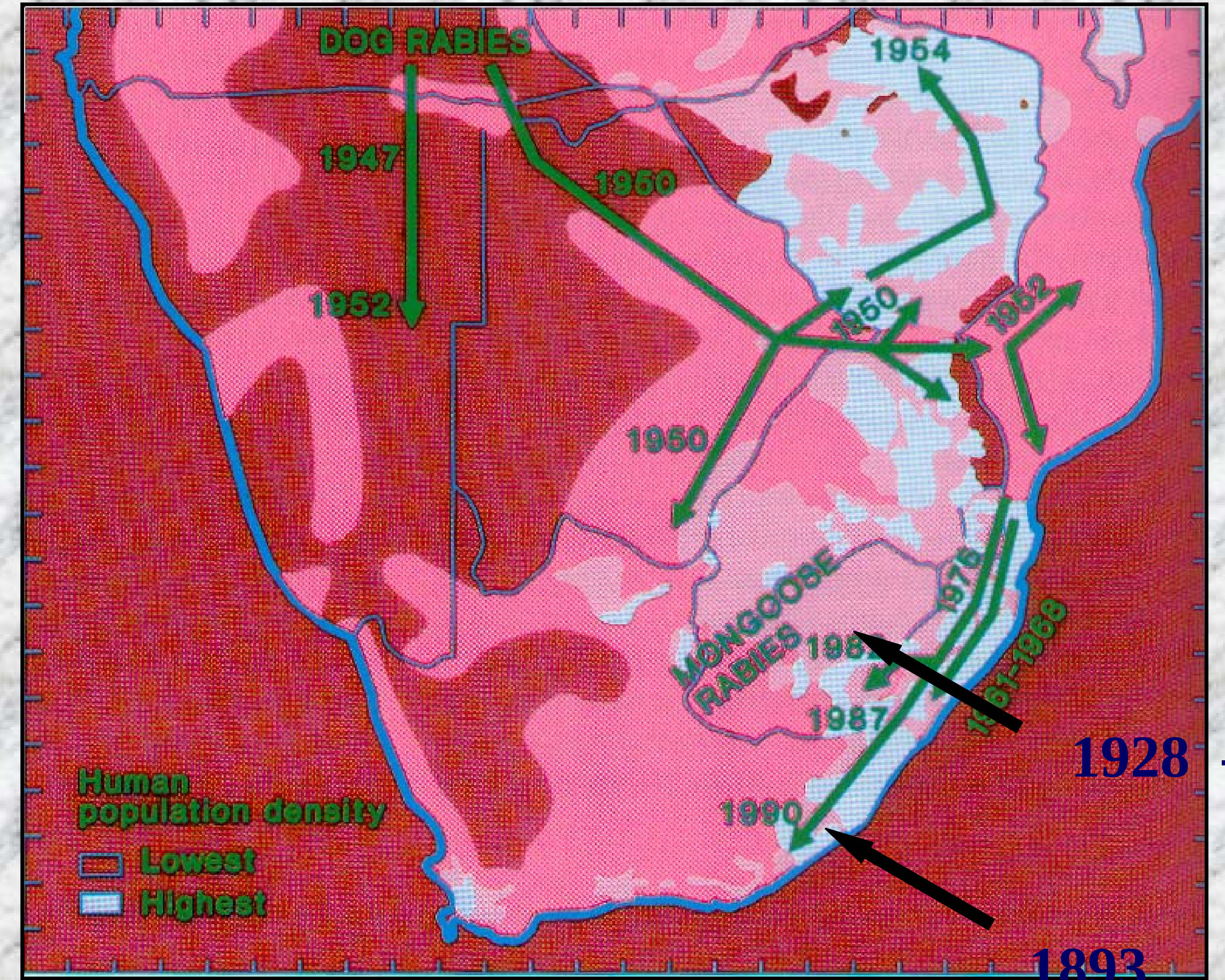




Rabies in South Africa: additional facts

- Canid rabies only became common in South Africa after 1950
- Rabies diagnosed in wild carnivore species in 1908
- Yellow mongoose *Cynictis penicillata* frequently diagnosed with rabies (70%).

Canid rabies in southern Africa: the history

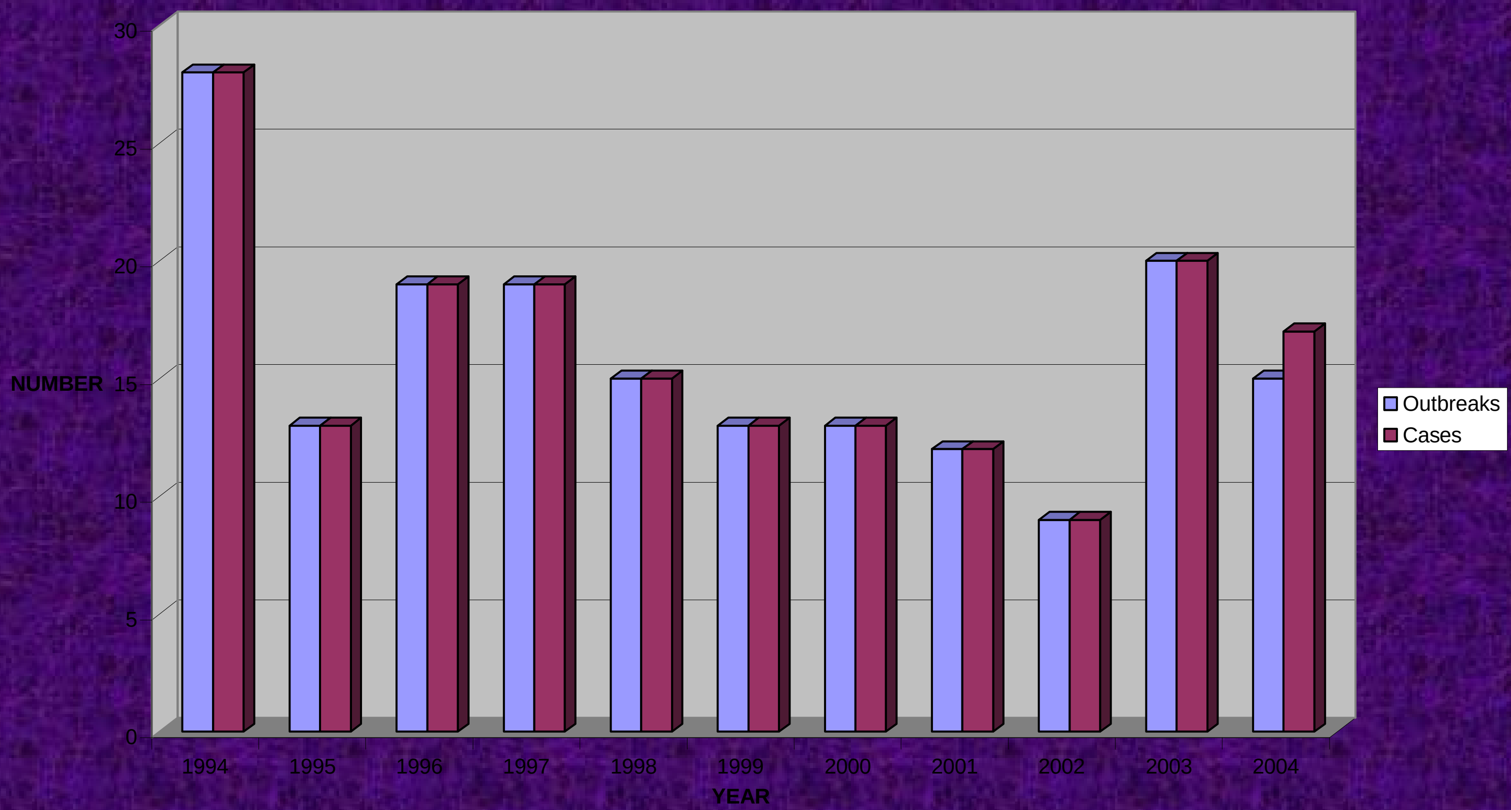


1928 - 2 human cases

1893

Source:

Rabies cases in bat-eared fox (1994-2004)



Compiled by Animal Health, Department of Agriculture

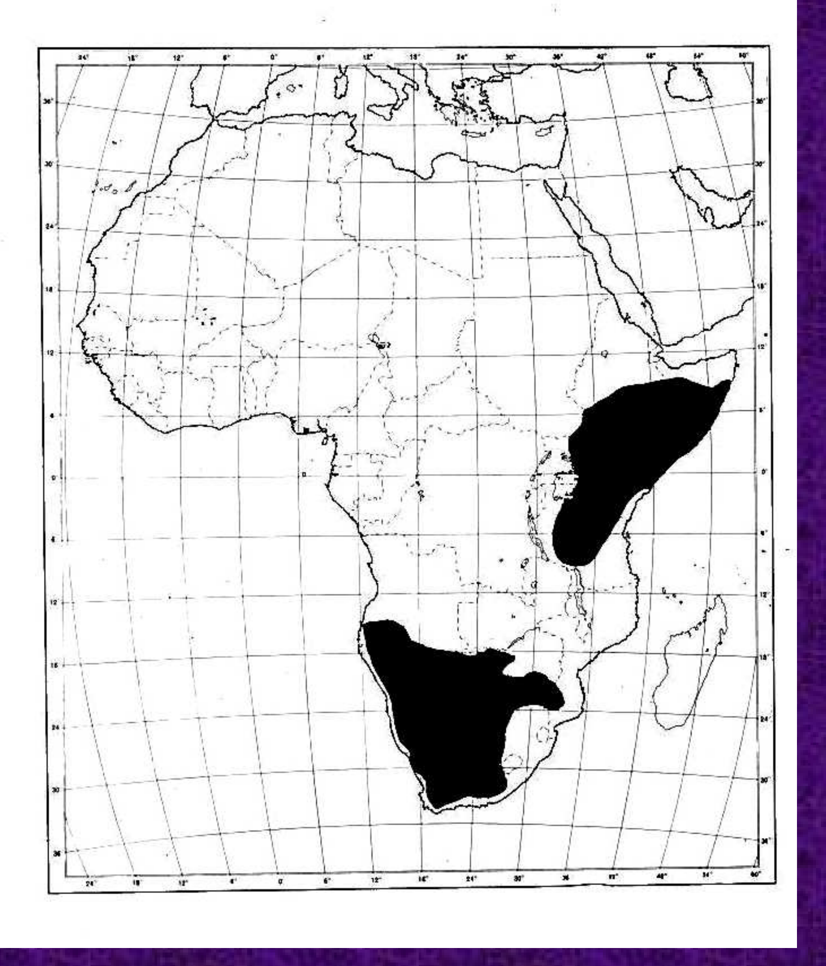
The bat-eared fox - *Otocyon megalotis*

- Small nocturnal canid
- Ears relatively large in proportion to the head
- Bat referred to is the Egyptian slit-faced bat (*Nycteris thebaica*), common and widespread in the region.
- Afrikaans name *bakoorjakkals* or *draaijakkals* often used although they are not jackals.

Source: The mammals of the southern African sub region 1990

Bat-eared fox distribution

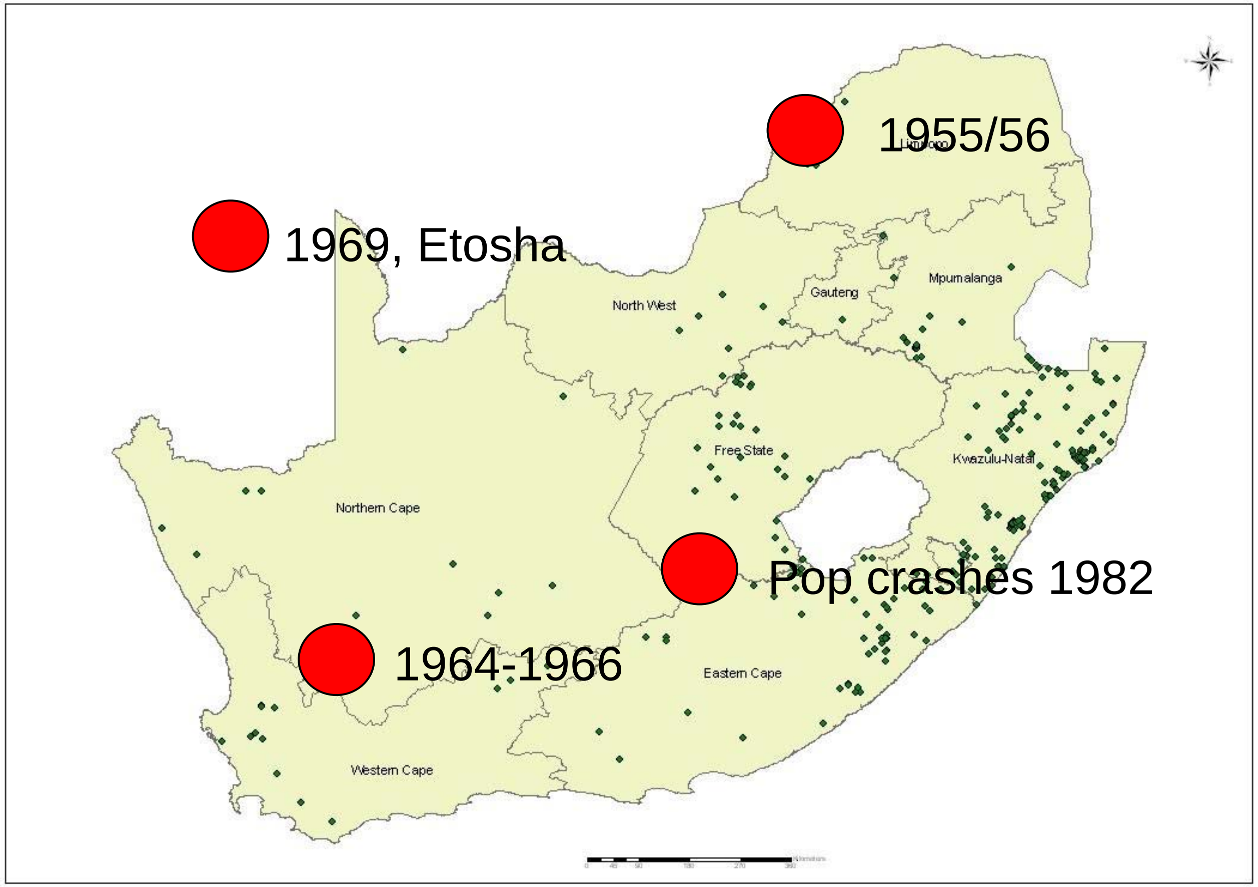
Two disjunct populations in southern (Kalahari, Namib desert, Botswana, SW Angola and East Africa (Tanzania)



Source: The mammals of the southern African sub region 1990

Maintenance host species for canid rabies in southern Africa

Rabies cases in bat-eared foxes

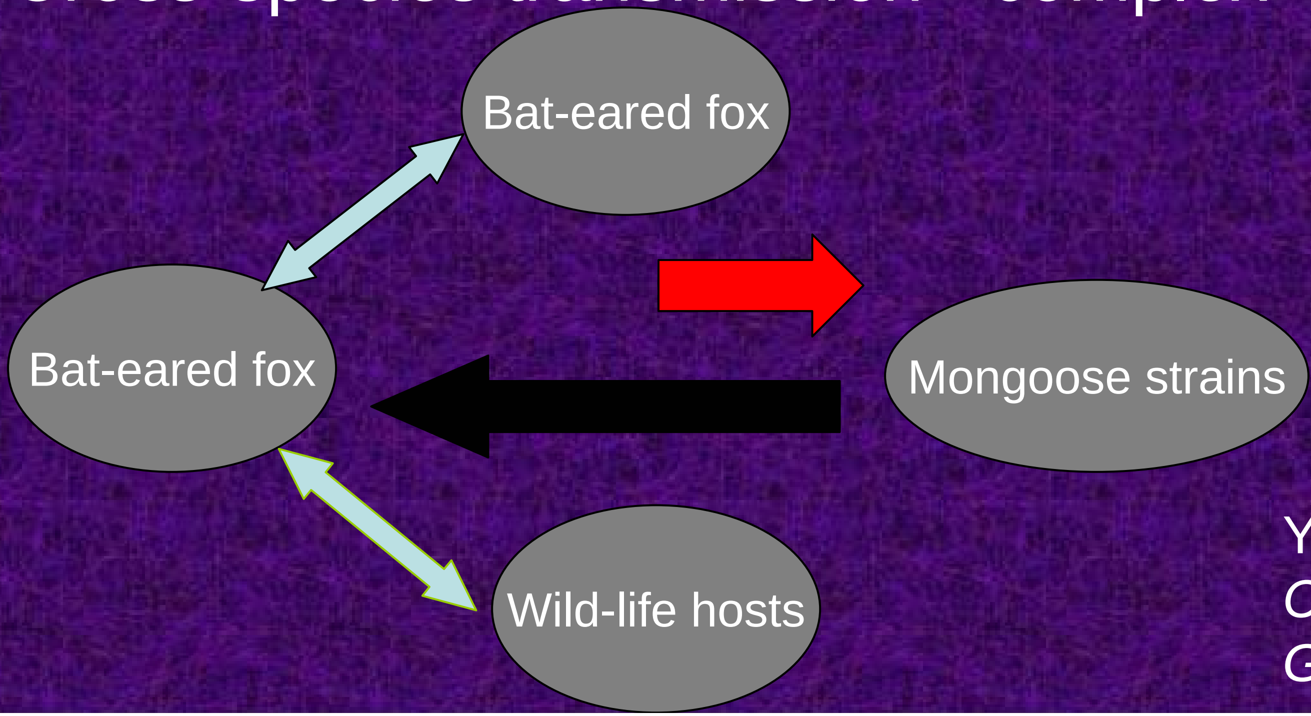


Questions

- Recent appearance of rabies in bat-eared foxes has raised the following questions;
  - Does *O. megalotis* maintain rabies cycles independent of other species?
  - Rabies in *O. megalotis* – result of spillover from domestic dog/arrived spread of an infection in this species from Namibia/Botswana?
  - Is there any threat to the dog population in the Western Cape region?

Results and discussion

- Antigenic typing: *O. megalotis* maintains the canid rabies biotype
- Mongoose strains in bat-eared fox are a result of spillover
- Cross-species transmission - complex



Yellow mongoose
Cynictis penicillata
Genets, felids

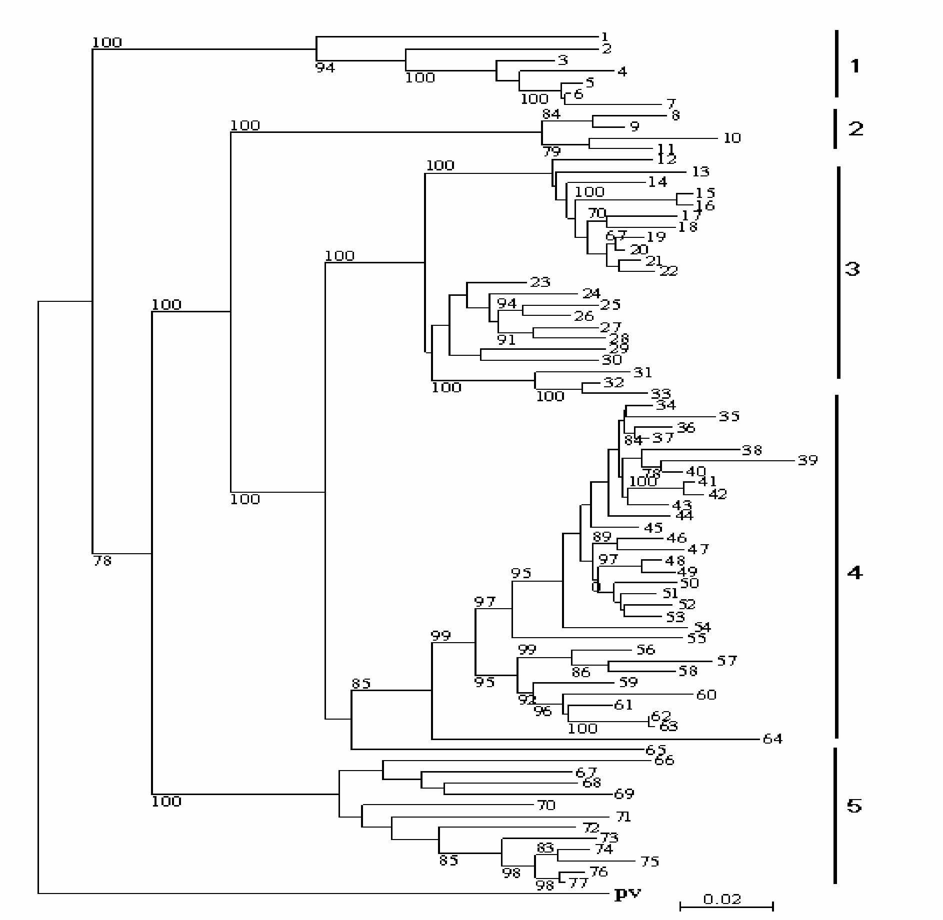
Monoclonal antibody reaction

	1	2	3	4	5	6	7	8	9	10	11	12	
C	+	-	+	+	+	+	-	+	+	+	-	+	X
	+	-	+	+	-	+	-	+	+	+	-	+	
V	+	-	+	+	-	+	-	-	-	-	-	+	X
	+	-	+	+	+	+	-	-	-	-	-	+	
	+	-	+	-	-	+	-	-	-	-	-	-	X
	+	+	+	+	-	+	-	-	-	-	-	+	
	+	+	+	+	-	+	-	-	-	-	-	+	
	+	+	+	+	-	+	-	-	-	-	-	-	

Rabies control program

Two aspects of epidemiology, surveillance and knowledge of the distribution of the antigenic and genetic variants are essential components ➔ economical programs

Mongoose rabies in Zimbabwe and South Africa

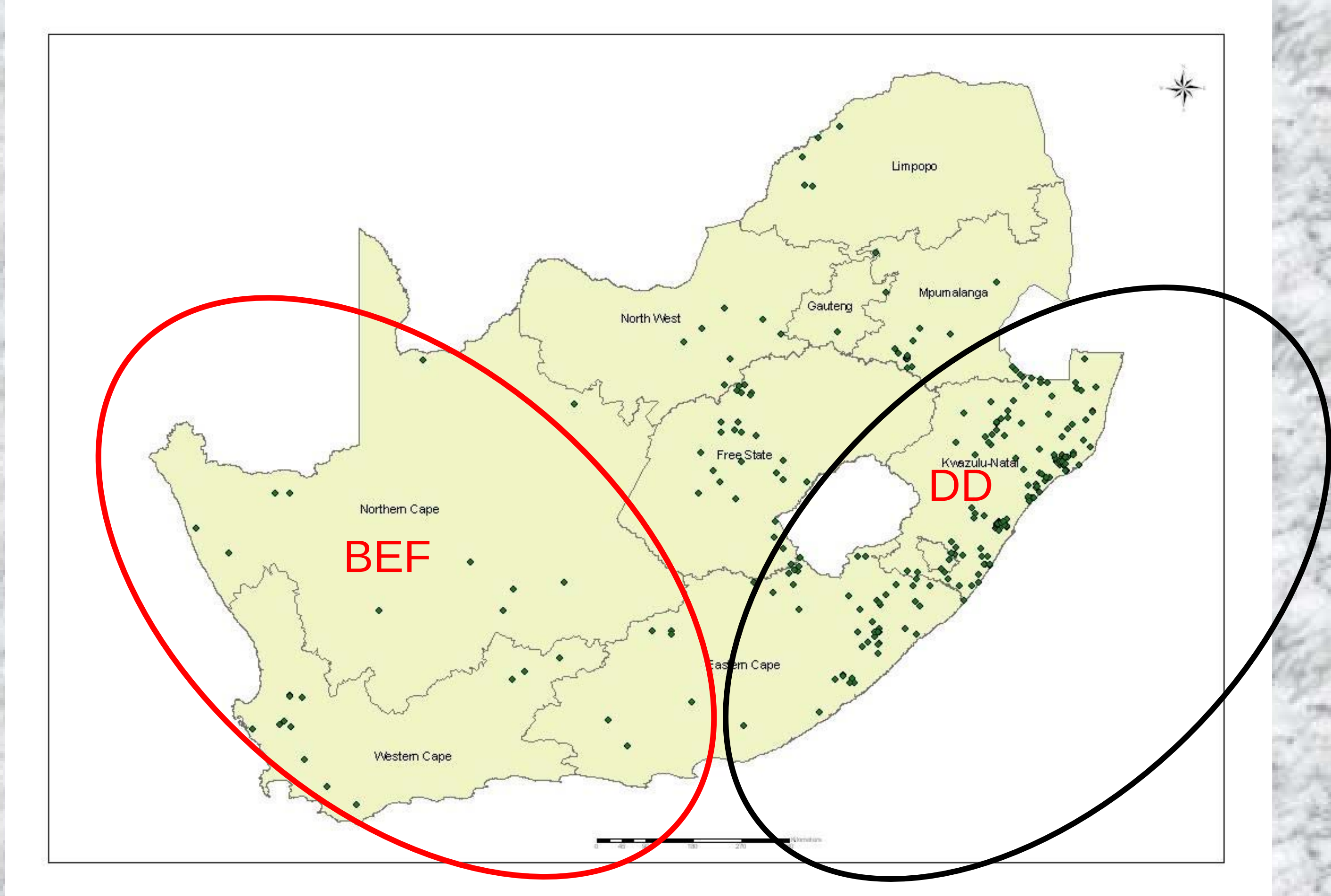


Nel et al., Virus Research 2005

Results and discussion

- In addition to the domestic dog (KZN and Limpopo provinces) and *C. mesomelas* (Limpopo province, canid rabies has now spread to the western part of South Africa.

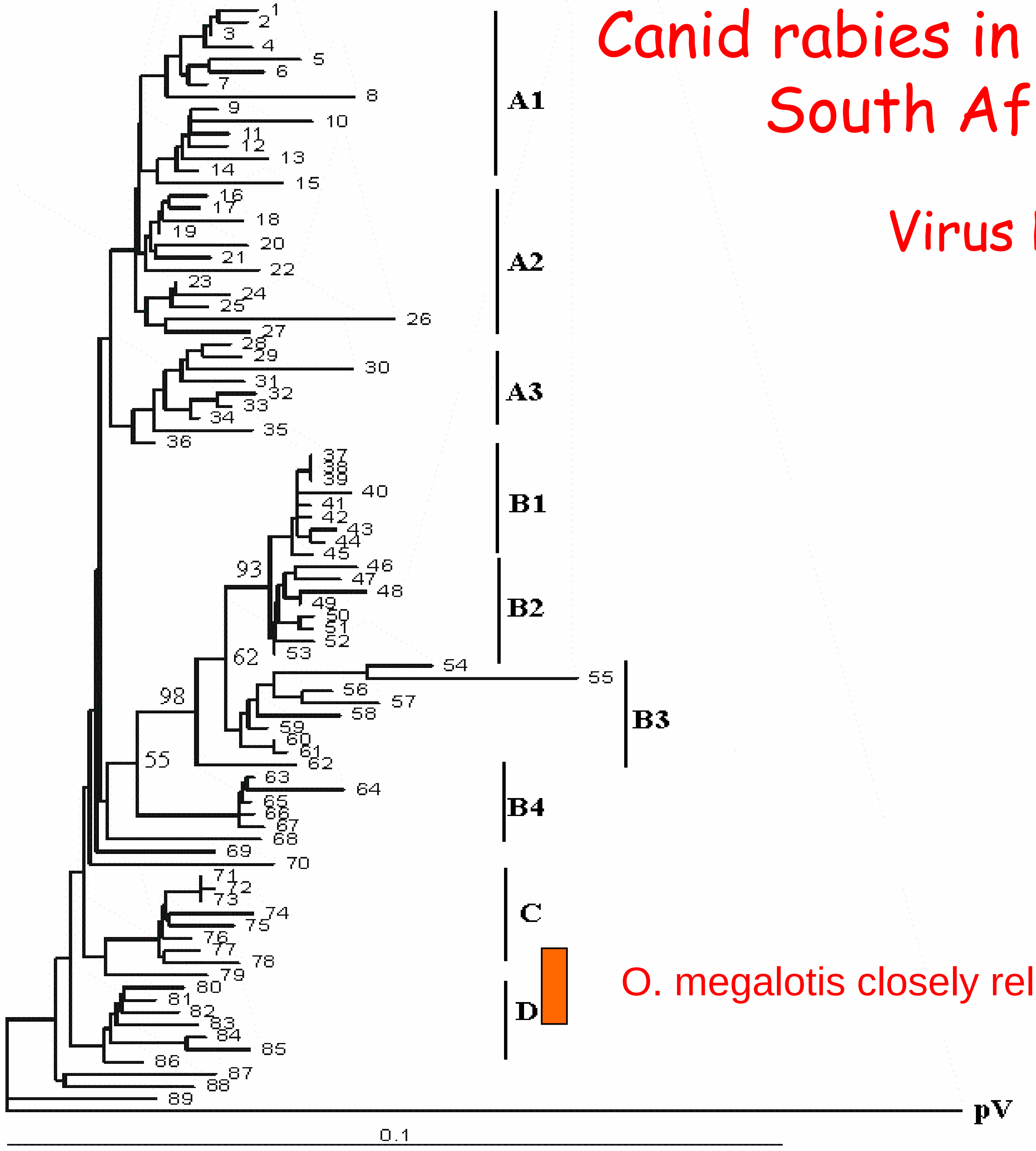
Rabies cases in South Africa (2004)



Compiled by Animal Health, Department of Agriculture

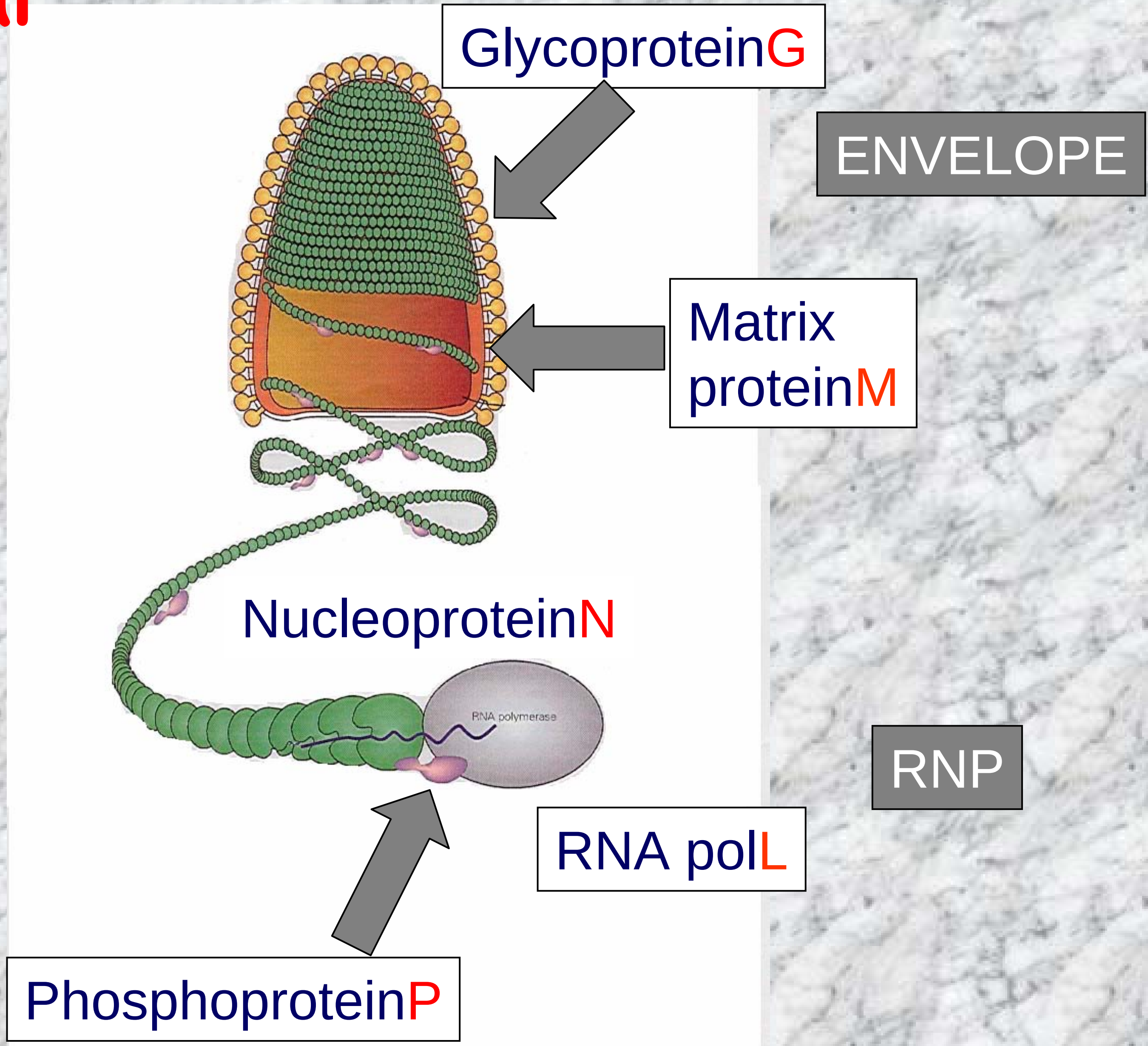
Canid rabies in Zimbabwe and South Africa

Virus Research, 2003



O. megalotis closely related but distinct

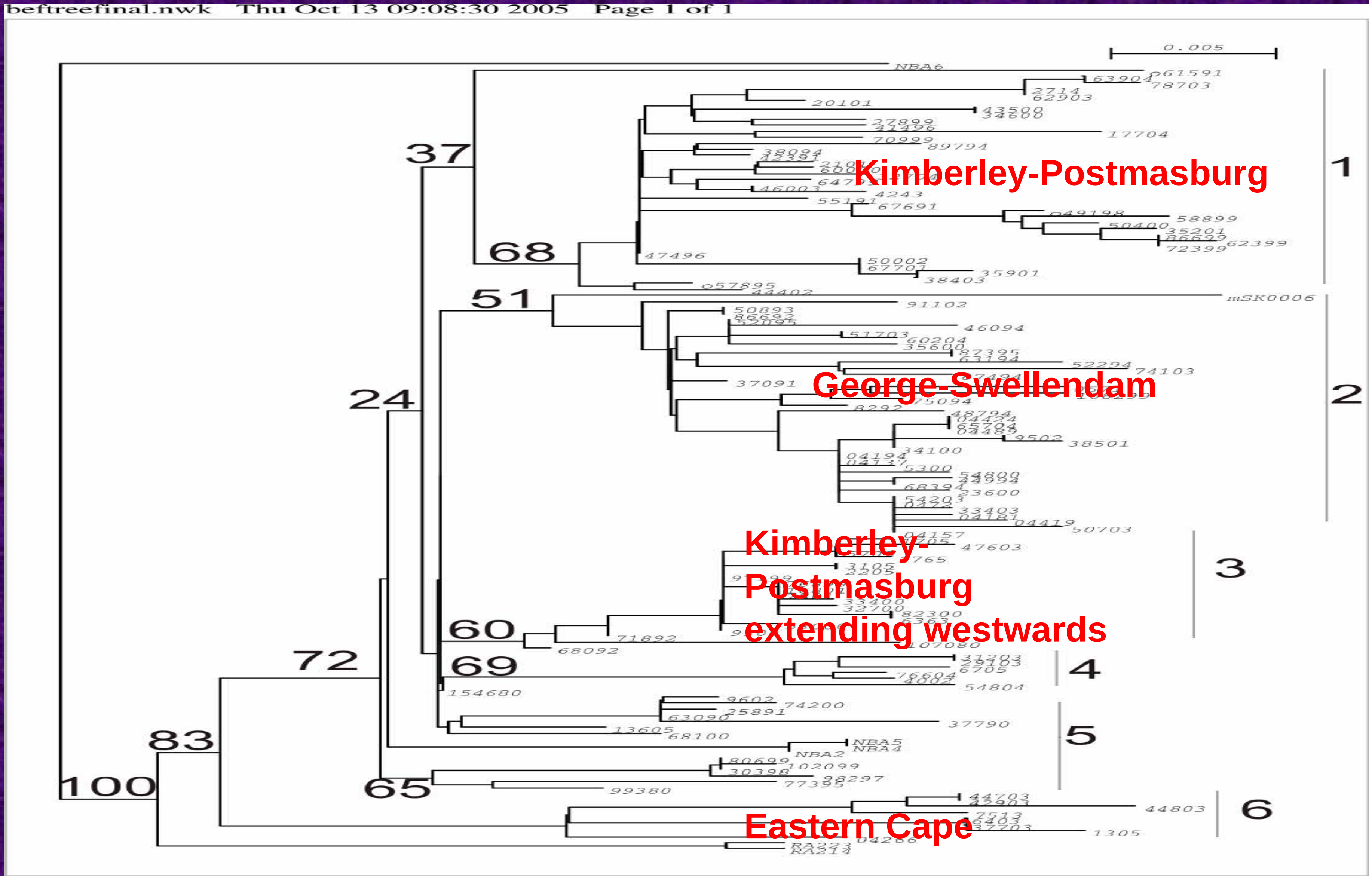
Rabies viral structure



Genetic analysis

- N = 129
- Amplification of the N and G-L intergenic regions done according to standard protocols (Heaton et al., 1997; Von Teichman et al., 1995).
- Sequence analysis (DNA star software package)
- Rabies viruses from bat-eared foxes very closely related, despite genomic region studied [G-L = 97%; N = 98.2%].

Genetic analysis



Genetic analysis

Genetic analysis

- Tree topologies similar (relatively low bootstrap support for the N tree).

- One clade: virus strains from the early 1990s; an old strain responsible for recent epidemics.

Conclusions

- Broader coverage of rabies variants more easily determined by antigenic typing
- Genetic methods are finer, resolve cyles in specific foci
- Bat-eared fox supports rabies cycles in the western part of the country (wildlife rabies)
- Interaction with domestic dogs (Eastern Cape)
- The viruses are very closely related
- Social dynamics need further investigation
- Other tools such as modelling could be utilised

Acknowledgements

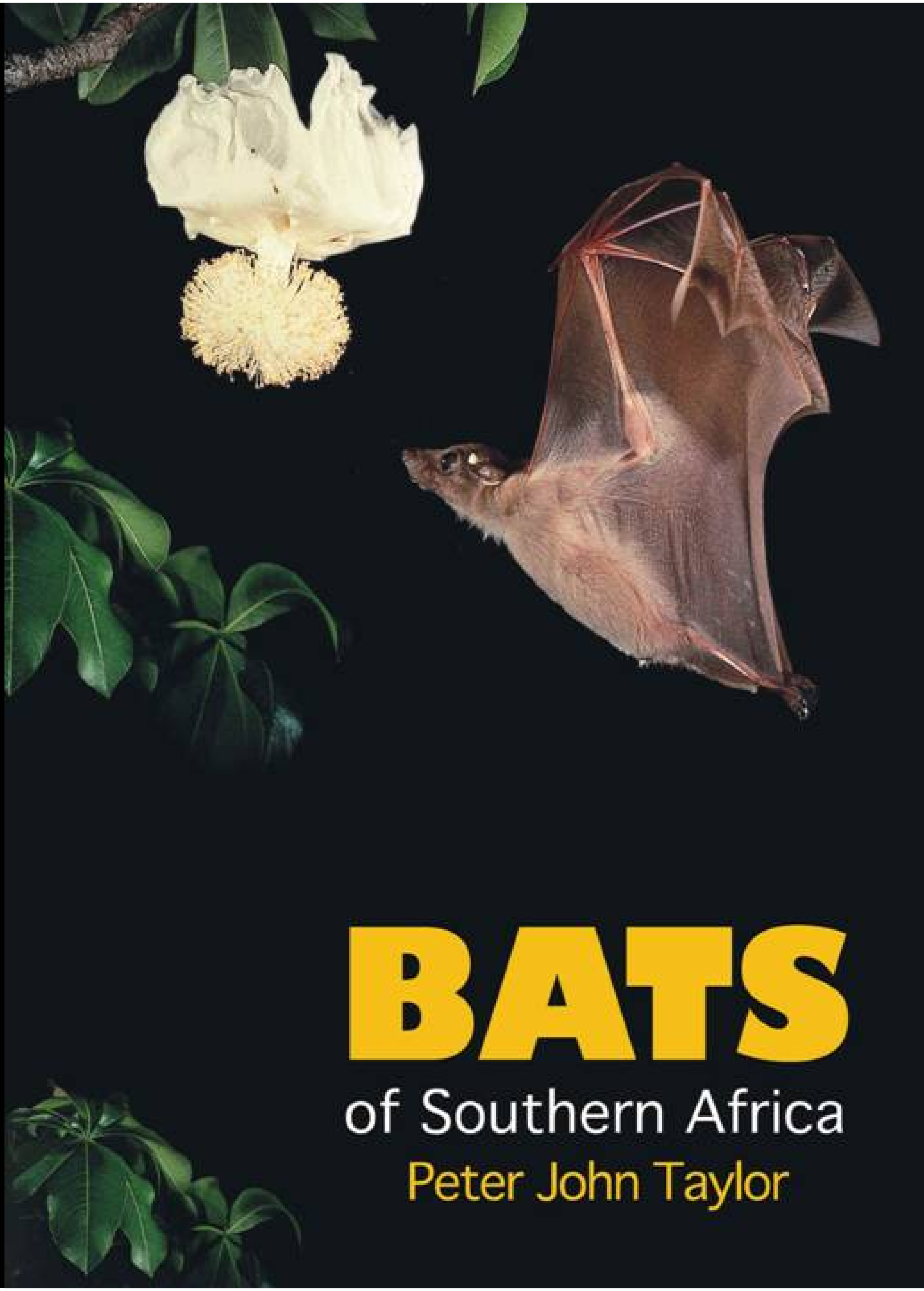
- J Bingham
- A Liebenberg
- A Wandeler
- J Randles
- L Nel
- T Fooks
- D Mohale

BATS OF AFRICA:

Diversity, misconceptions, truths, threats

Peter John Taylor

Durban Natural Science Museum



BATS
of Southern Africa

Peter John Taylor

New Scientist 5 Dec 1998

"Bats out of hell"

"Death on the wing - three species of fruit bat carry the lethal megamyxovirus"

"In the past four years, Australia has seen three new diseases makes the leap from bats to humans, two of them with fatal consequences"

VICTIMS OF BAD PRESS!!

(All) "Bats carry rabies!"

"After the first fatal case of ABL hit the news, there was a spate of indiscriminate killings"

"Ebola in Australia? ... the disease could turn up anytime , courtesy of a roving fruit bat"

"… in the past four years only three people have died from bat-borne diseases, far fewer than the annual toll from snake or spider bite"

"Bats should not be considered public enemy number one. Fruit bats play a vital ecological role"

THE RESPONSE!

BATS Magazine - Summer 1999, Fall, 2000
RABIES UPDATE: The Media Blitz that Threatens Bats,
M. D. Tuttle (Founder of Bat Conservation Int.)

“To the general public, it all sounds very scary, and typically, these hypotheses become fact in the public mind long before scientists can test them”

“The intrinsic conflict between the mandated need to inform the public of potential health hazards and the need of the media for sensational headlines is threatening to compromise the scientific process.” *Nature* June 11, 1996

“In recent months, leading newspapers, medical publications, and even a children’s magazine have published greatly exaggerated warnings that bats are the primary source of human rabies in America, that bites can easily occur without being noticed, and that medical advice should be sought immediately if one has been near a bat. The September 29, 2000, issue of The New York Times, in addition to repeating these scary warnings, noted that individual New York families are now paying up to $5,000 or more to rid their premises of bats.”

“State Department of Health policies are causing a whopping $1 million to be spent locally every three years on a disease that has never struck a resident of Ontario County: rabies contracted from bats.”

“Human Mortality in the United States
Annual Deaths*
900 from bicycle accidents
150 from accidents caused by deer
20 from dog attacks
18 from power lawn mower accidents
4 from accidents on playground equipment
1.2 from bat rabies
***average annual rates per population of 267,000,000**
from the International Classification of Diseases and the National Highway Traffic Safety Administration “

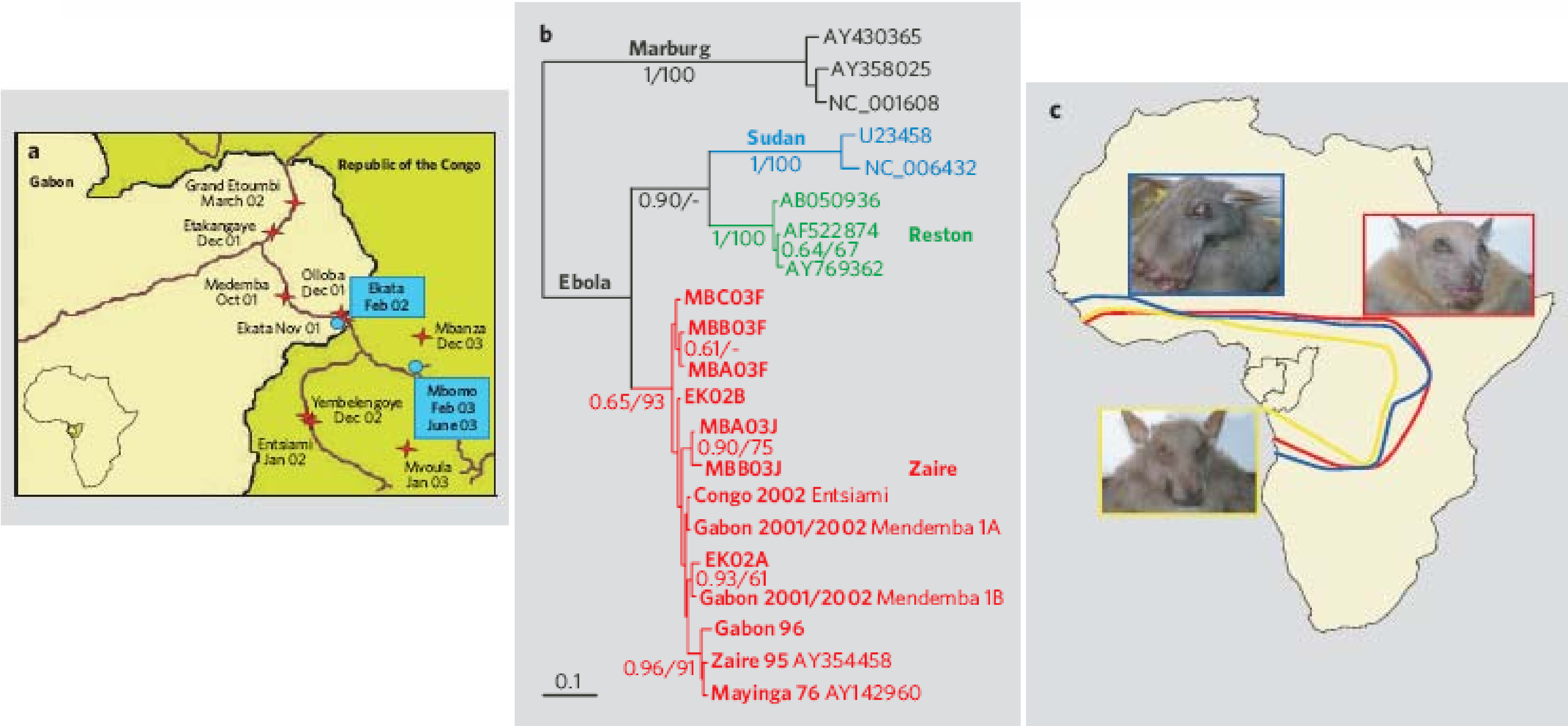
“Are claims that bats are aggressive, and that undetected bat bites are an important source of human rabies, justified? Some 250 bat researchers from the United States, Canada and Mexico say no. “

“..in its October 23, 1999, issue the New Scientist ran a four paragraph update admitting that ‘researchers hunting for the source of the Ebola virus have been looking in the wrong place.’ They concluded that ground-dwelling mammals, not bats, are now the prime suspects.”

BRIEF COMMUNICATIONS

Fruit bats as reservoirs of Ebola virus

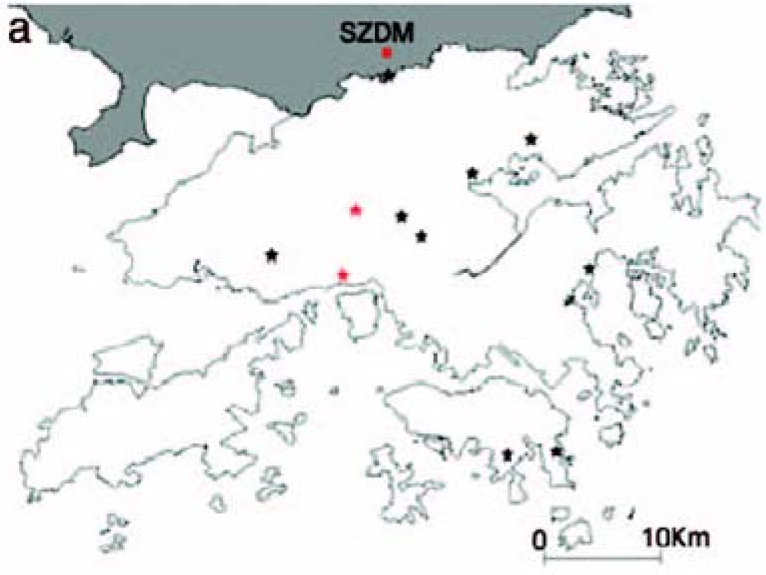
Bat species eaten by people in central Africa show evidence of symptomless Ebola infection.



Severe acute respiratory syndrome coronavirus-like virus in Chinese horseshoe bats

Susanna K. P. Lau*†‡§, Patrick C. Y. Woo*†‡§, Kenneth S. M. Li*, Yi Huang*, Hoi-Wah Tsoi*, Beatrice H. L. Wong*, Samson S. Y. Wong*†‡, Suet-Yi Leung¶, Kwok-Hung Chan*, and Kwok-Yung Yuen*†‡§

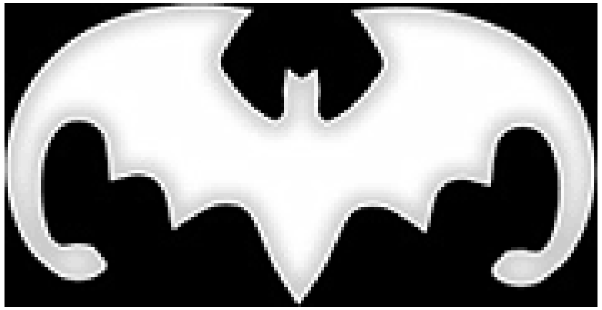
14040–14045 PNAS **September 27, 2005** vol. 102 no. 39



"The pathogenicity and host range of bat-SARS-CoV remain to be determined".

Mythology!

USUALLY NEGATIVE
BUT . . .

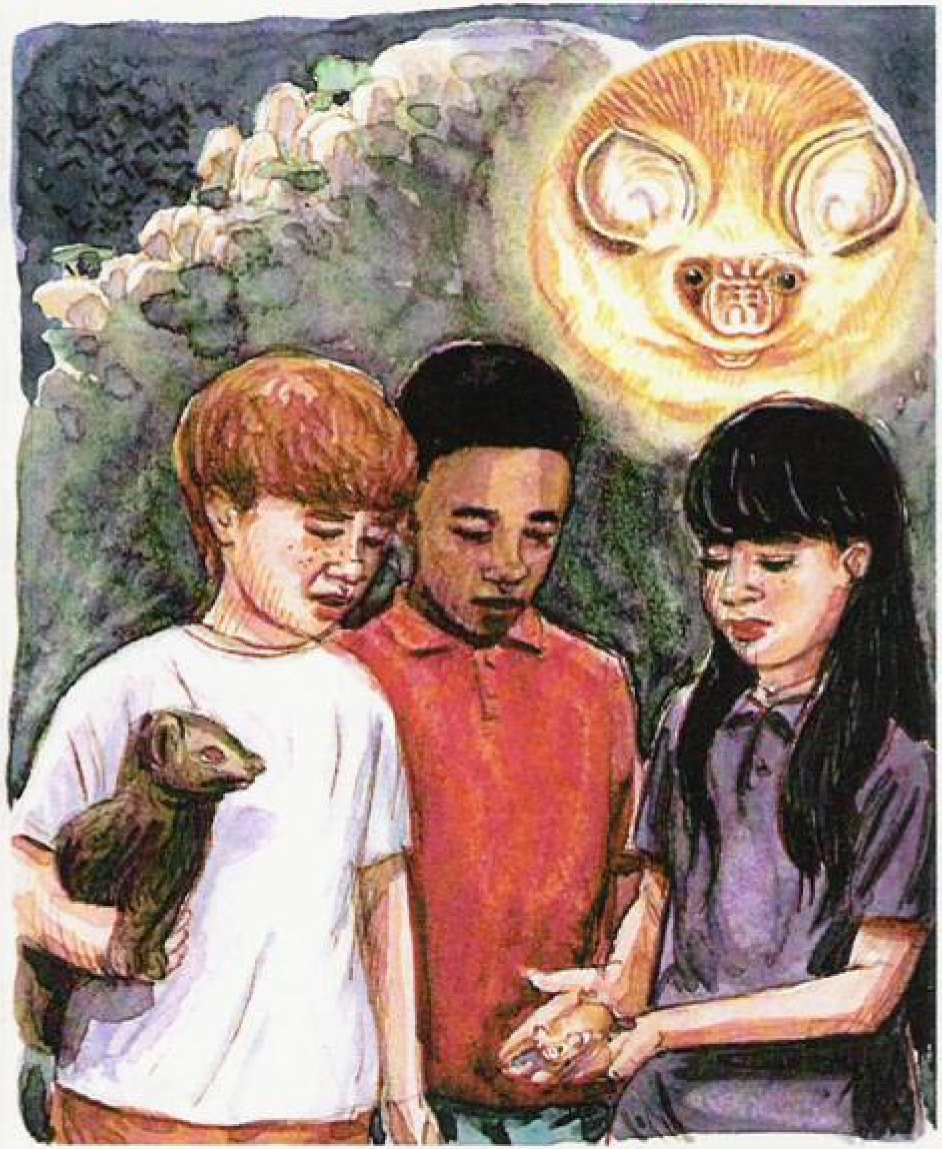


Symbol of happiness
("fu") in China

And good luck emblem
For Bacardi, founded
In 1862

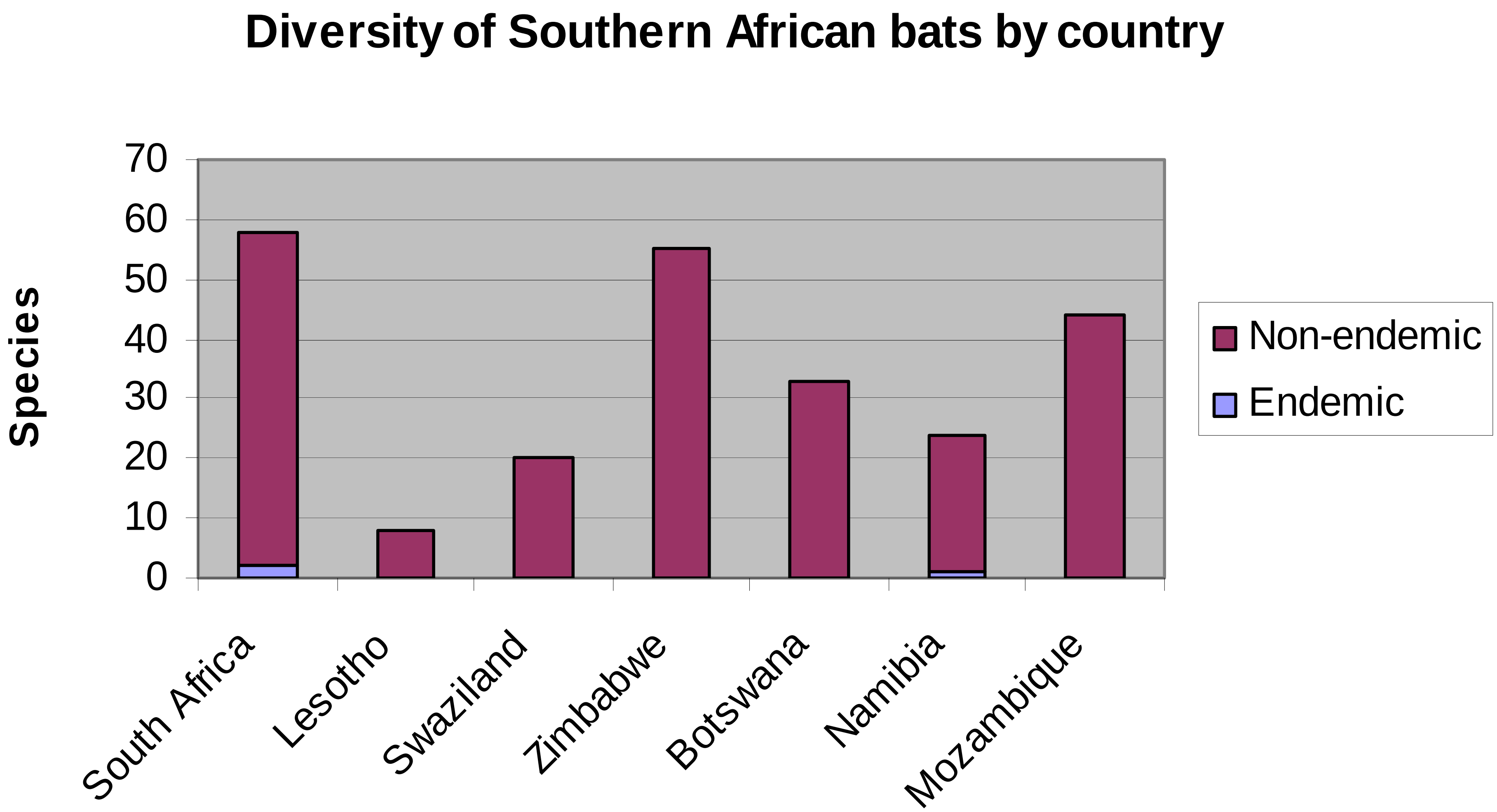
BACARDI
ND 279-657

ELIJAH
AND THE MESSENGERS OF DEATH

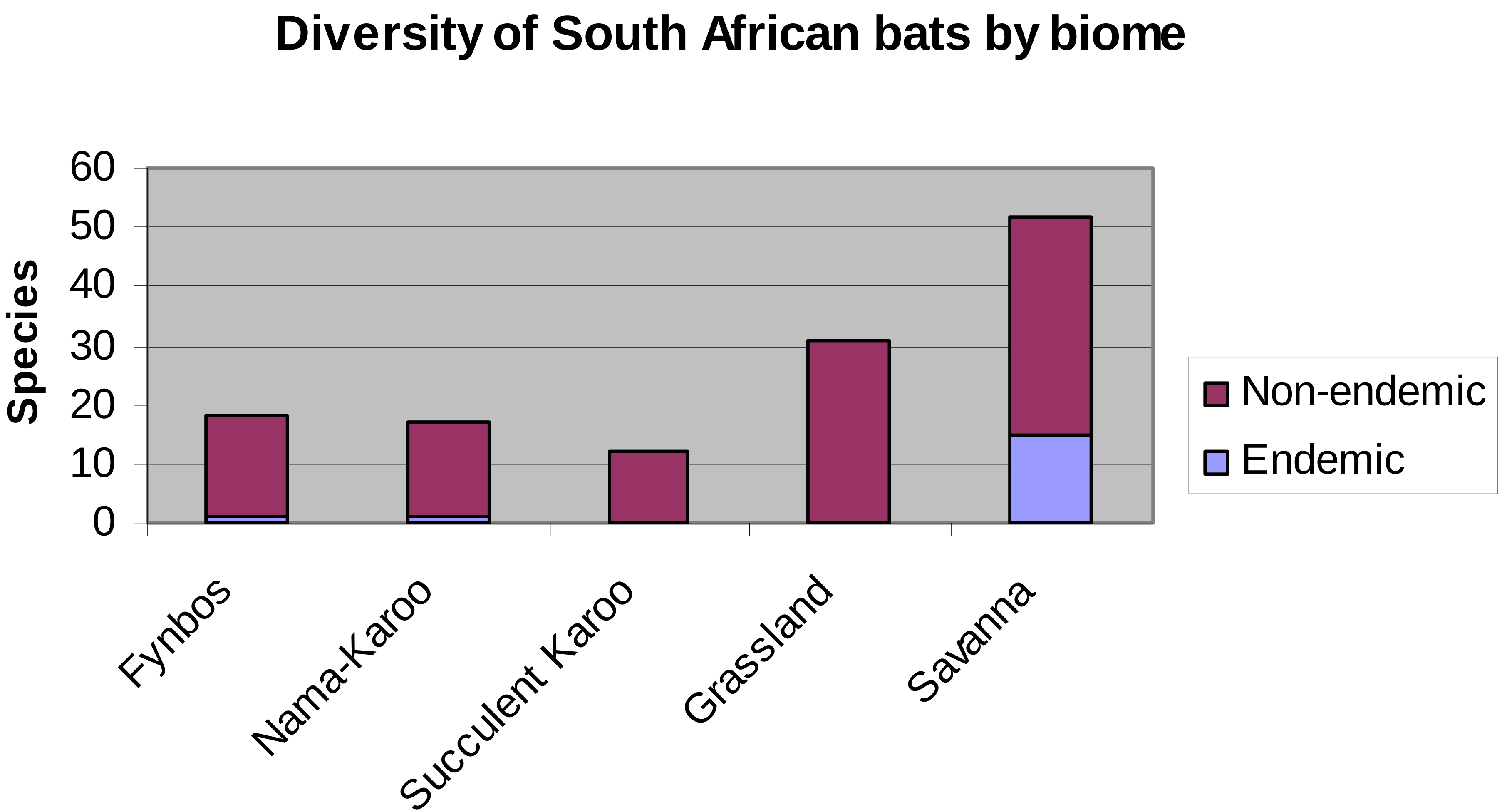


PETER JOHN TAYLOR

Bats of Southern Africa



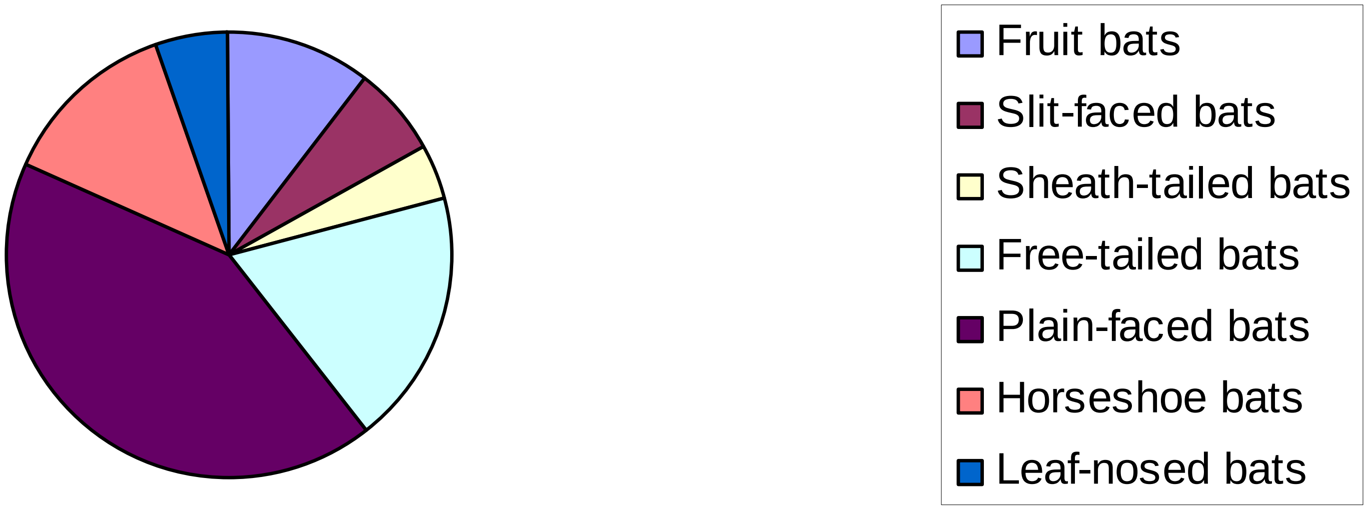
Diversity of Southern African bats by country
Species
70
60
50
40
30
20
10
0
South Africa
Lesotho
Swaziland
Zimbabwe
Botswana
Namibia
Mozambique
Non-endemic
Endemic


Diversity of South African bats by biome
Species
60
50
40
30
20
10
0
Fynbos
Nama-Karoo
Succulent Karoo
Grassland
Savanna
Non-endemic
Endemic

Diversity of Southern Africa bats (Chiroptera): Families

Total no. of species = 76

(Two suborders: Mega- and Microchiroptera or Yinpterochiroptera and Yanchiroptera)



fruit bats

8 species in southern Africa

STRAW-COLOURED FLYING FOX

WAHLBERG'S EPAULETTED FRUIT BAT

EGYPTIAN FRUIT BAT

Slit-faced bats

5 species in southern Africa

Sheath-tailed bats

3 species in southern Africa

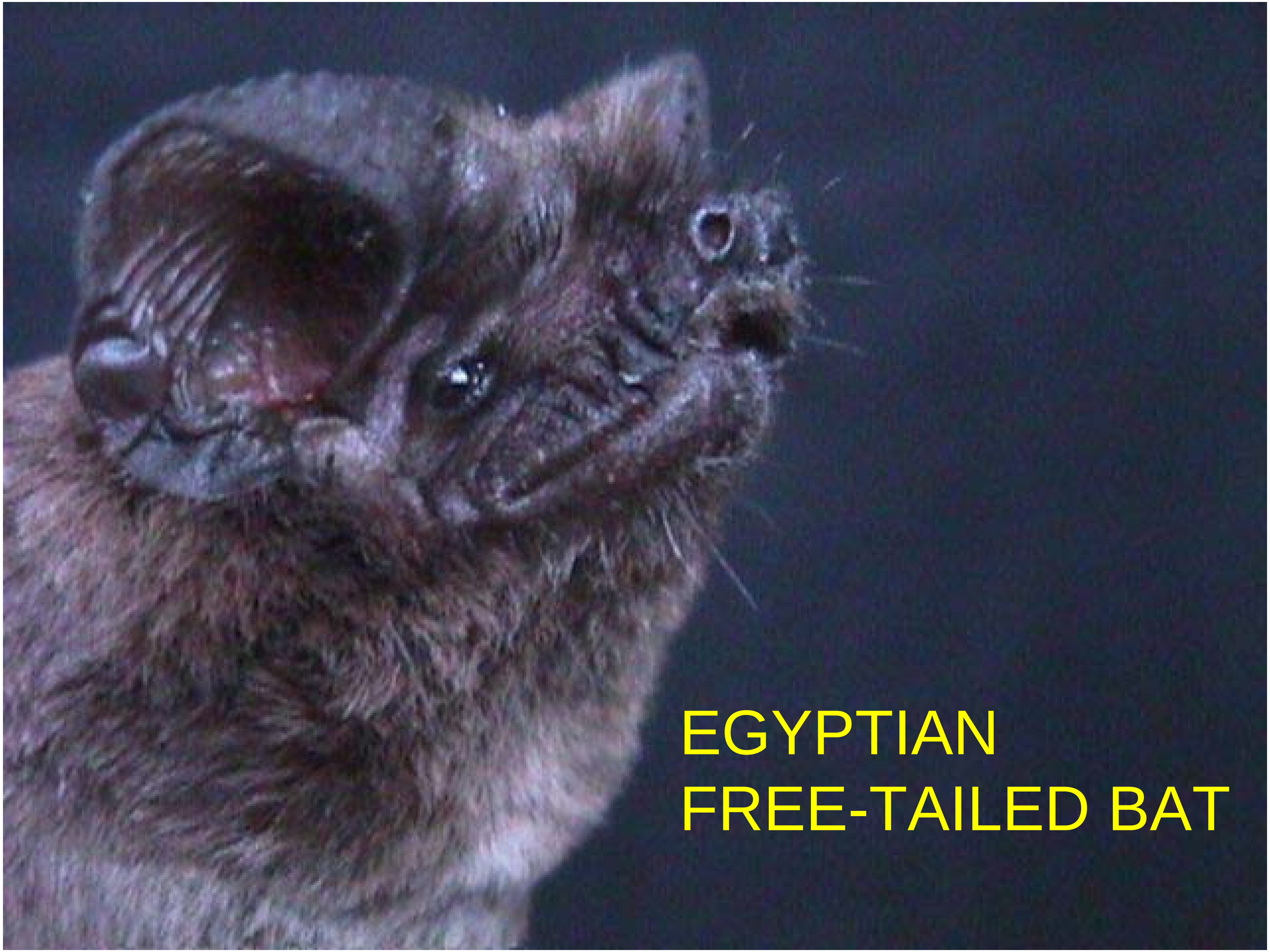
MAURITIAN
TOMB BAT


C. Grant
1999

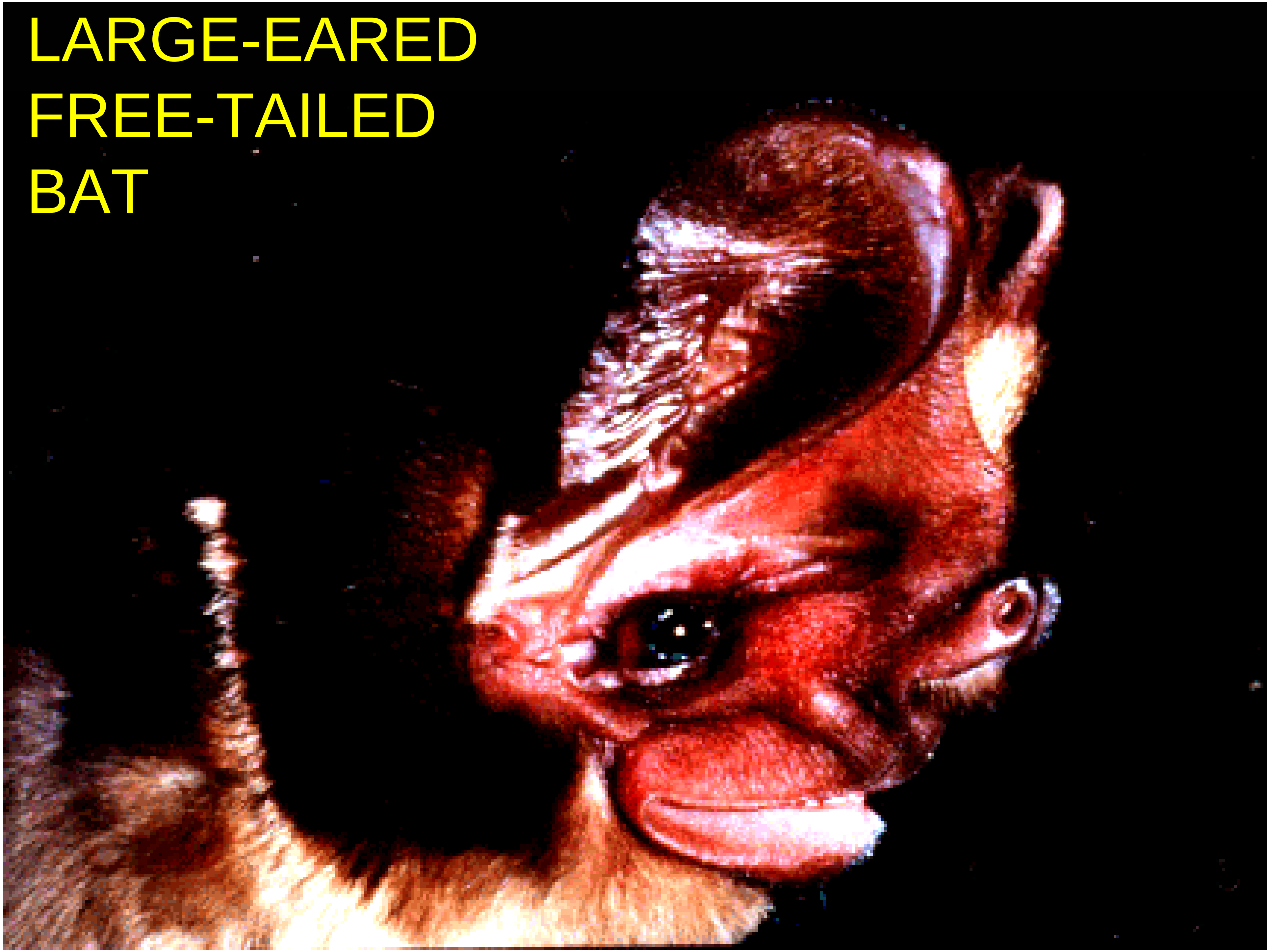
FREE-TAILED BATS

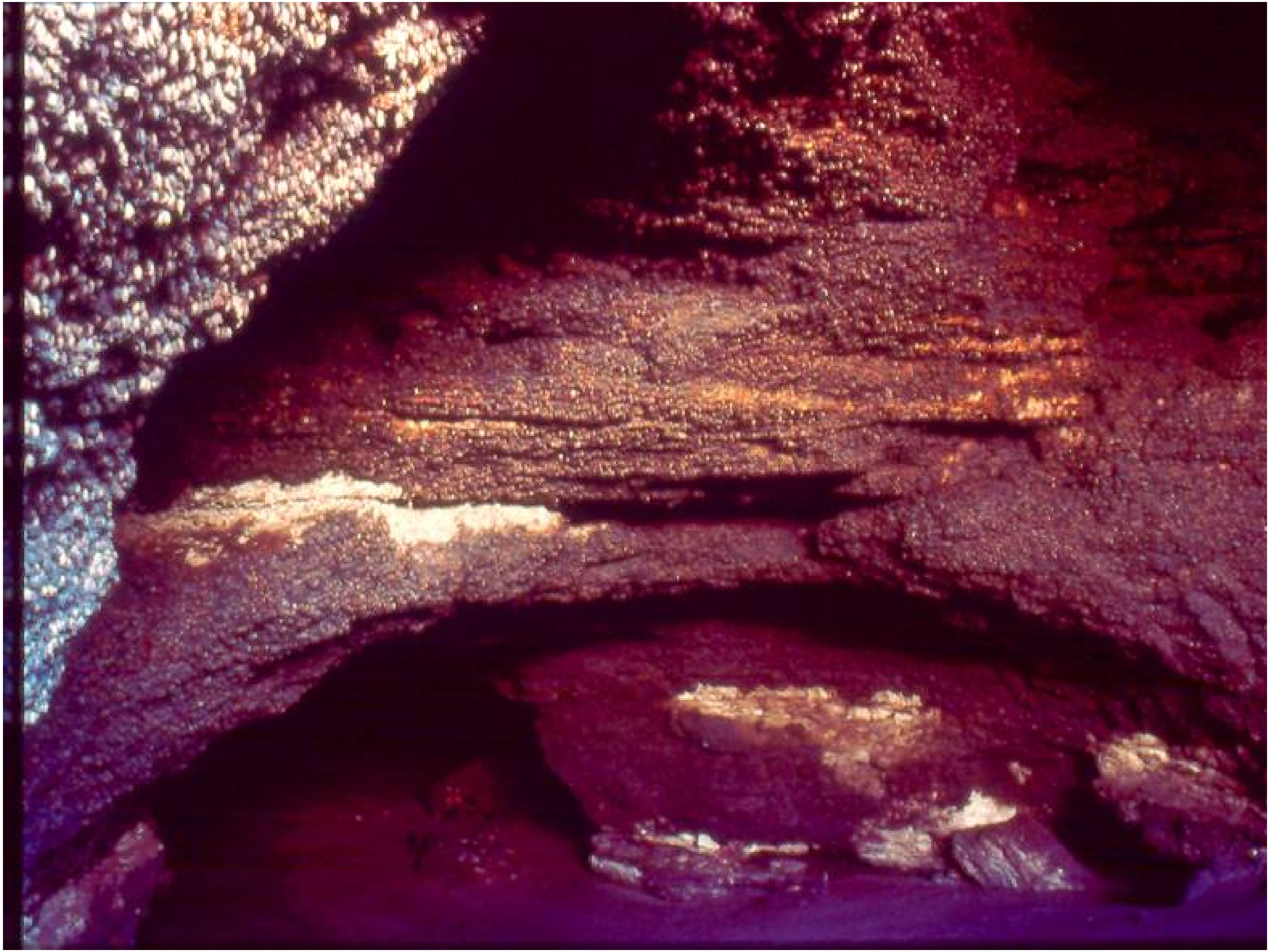
14 species in southern Africa

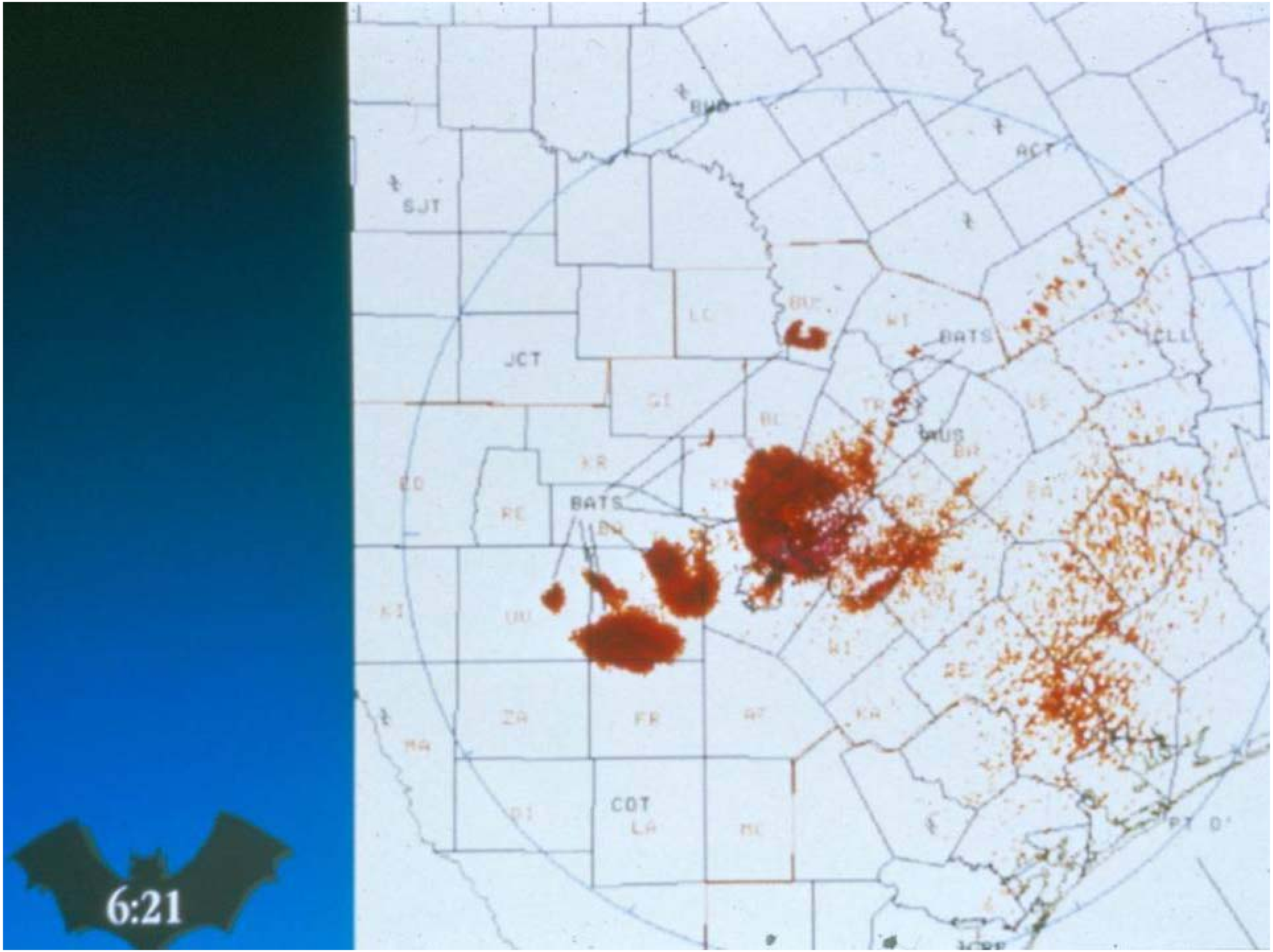
ANGOLAN FREE-TAILED BAT

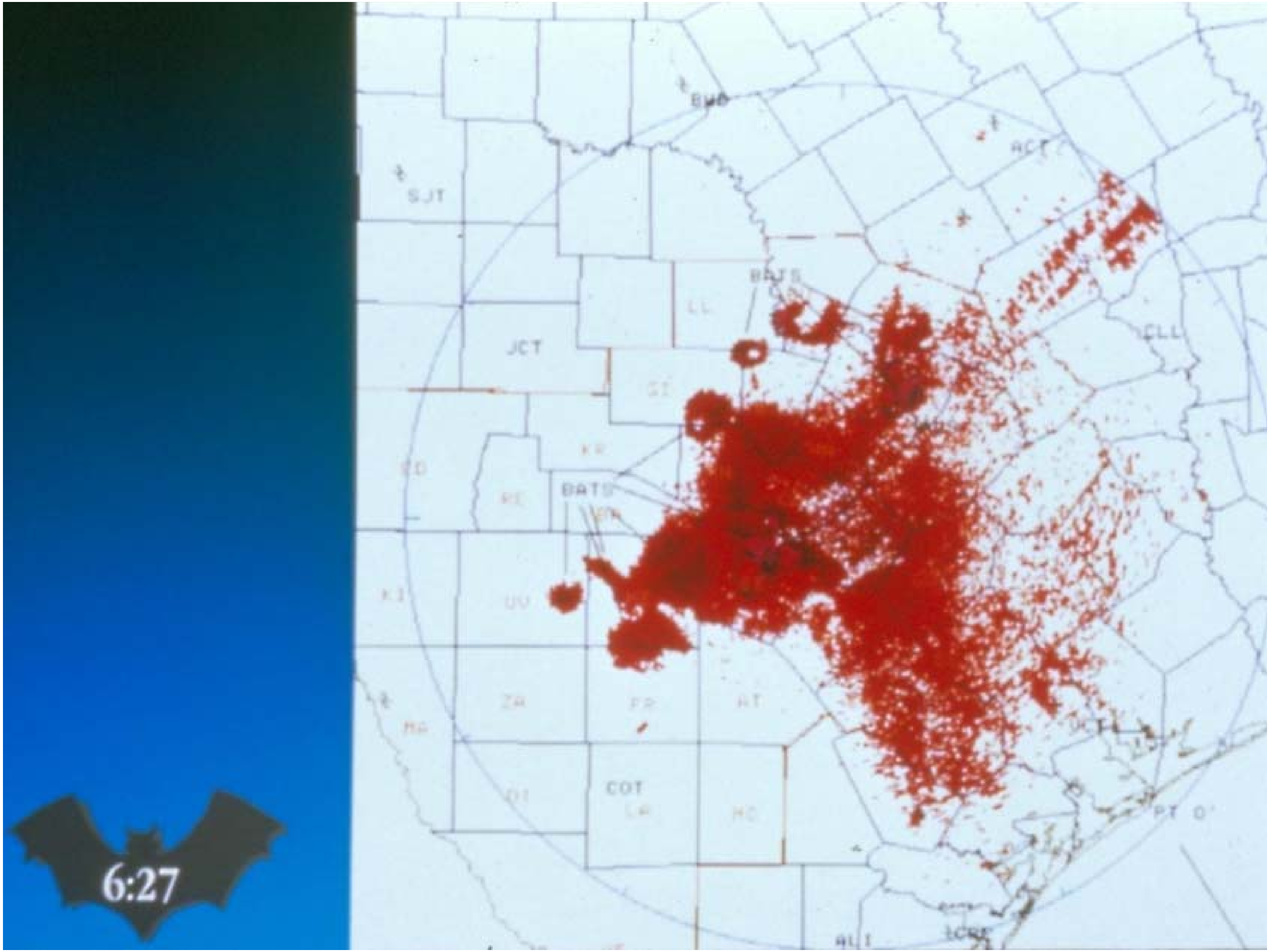

EGYPTIAN
FREE-TAILED BAT


LITTLE FREE-TAILED BAT


LARGE-EARED
FREE-TAILED
BAT


6:21
SJT
BWD
ACT
JCT
BATS
CLL
AUS
BATS
COT
PT O'


6:27
SJT
BWD
ACT
BATS
LL
CLL
JCT
BATS
COT
PT O'
ALI
CRP

PLAIN-FACED (VESPER) BATS

32 species in southern africa

YELLOW HOUSE BAT


BANANA BAT


C. Grant
2000

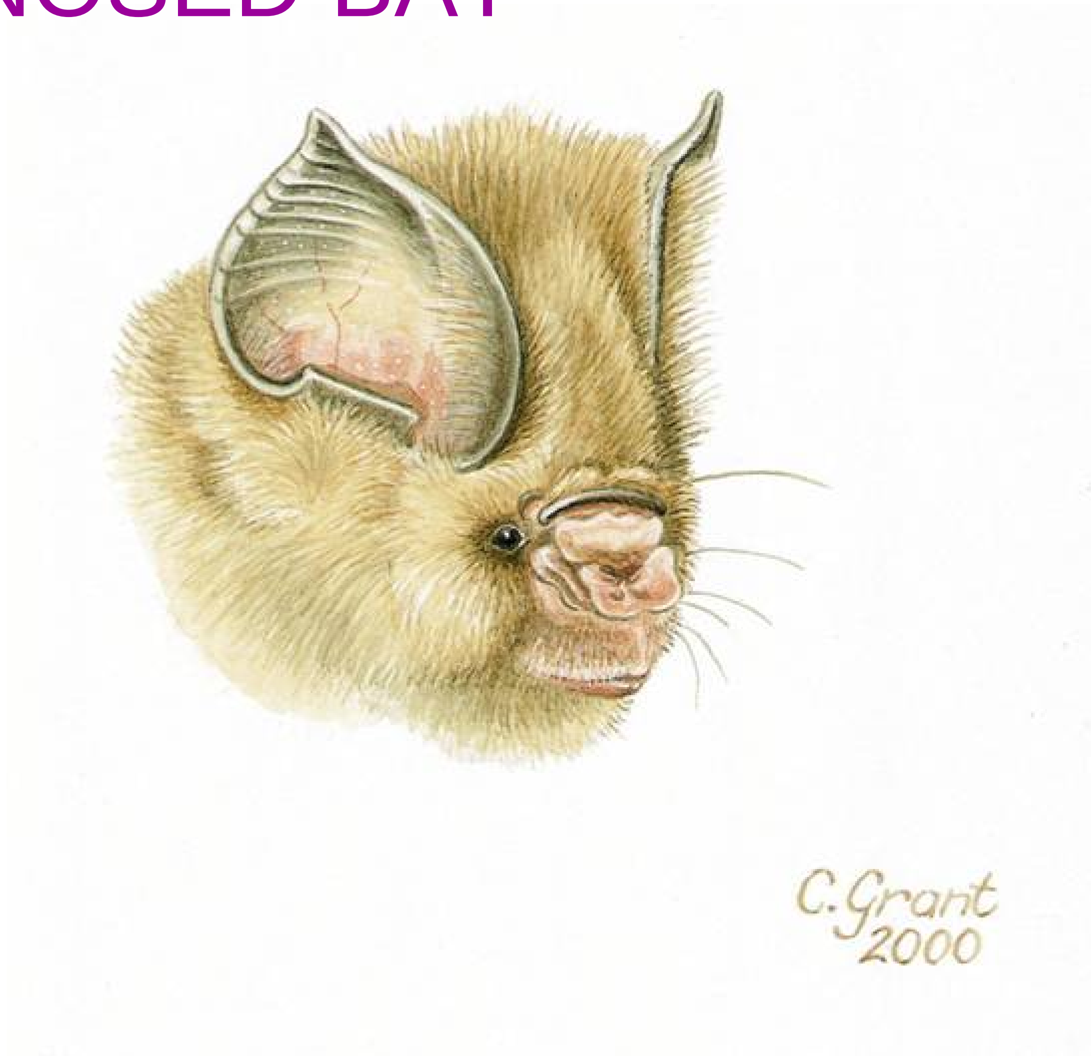
BUTTERFLY BAT

LEAF-NOSED BATS

4 species in southern Africa


COMMERSON'S
LEAF-NOSED BAT

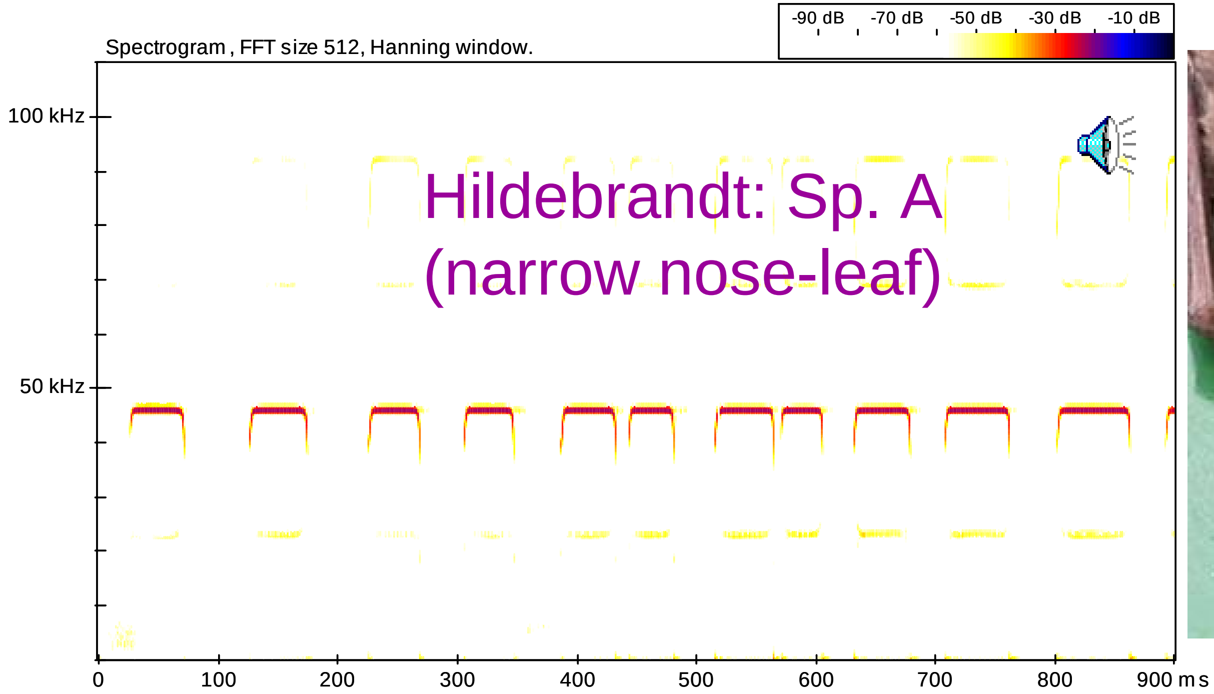
SUNDEVALL'S LEAF-NOSED BAT



HORSESHOE BATS

10 species in southern Africa


HILDEBRAND'S
HORSESHOE BAT

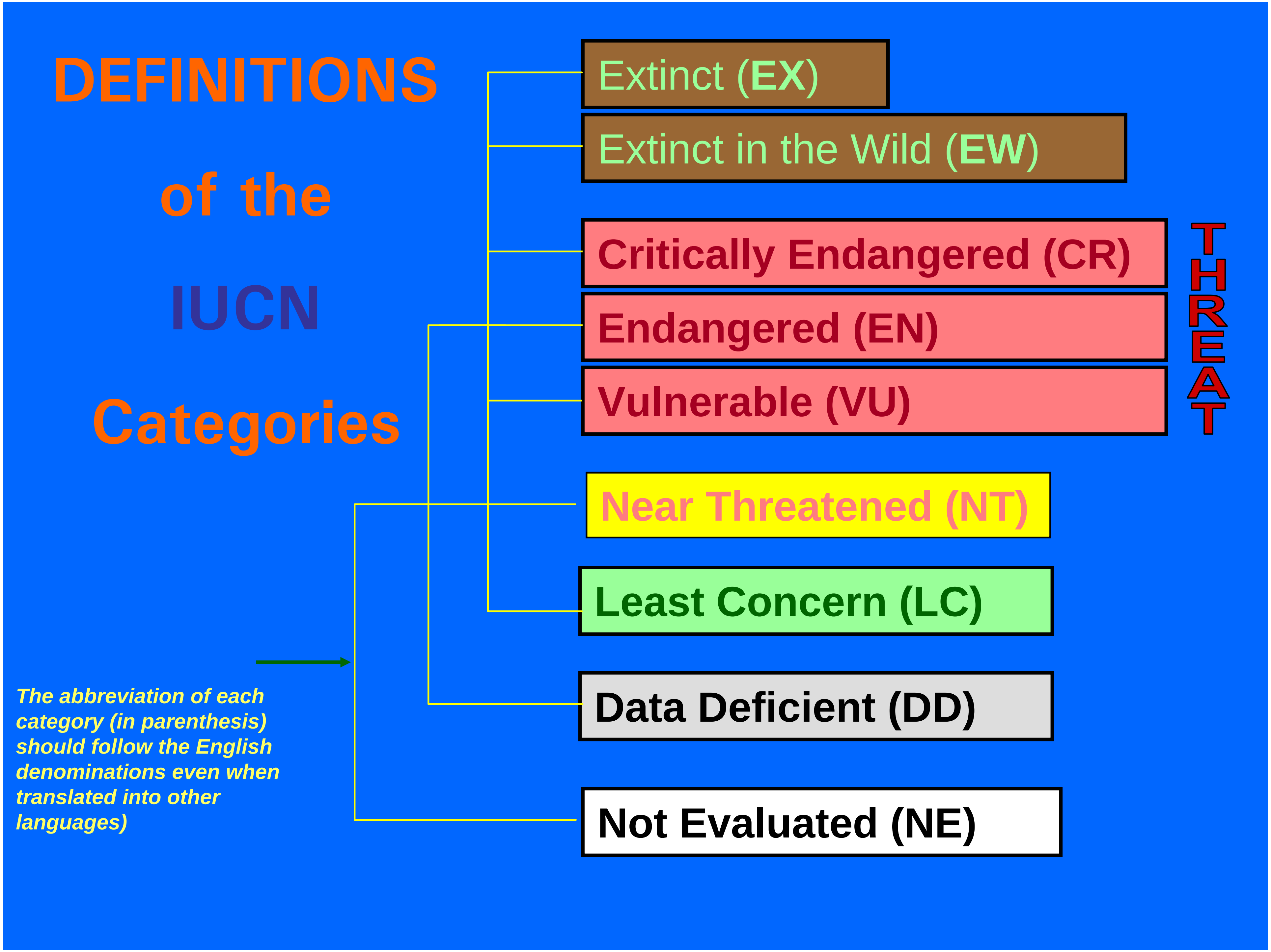


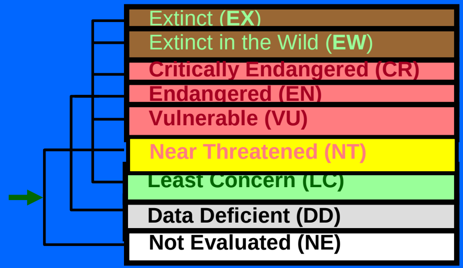
-90 dB -70 dB -50 dB -30 dB -10 dB
Spectrogram , FFT size 512, Hanning window.
100 kHz
50 kHz
Hildebrandt: Sp. A
(narrow nose-leaf)
0 100 200 300 400 500 600 700 800 900 ms

-90 dB -70 dB -50 dB -30 dB -10 dB
Spectrogram , FFT size 512, Hanning window.
100 kHz
50 kHz
Hildebrandt: Sp. B
(wide nose-leaf)
0 100 200 300 400 500 600 700 800 900 ms

How endangered

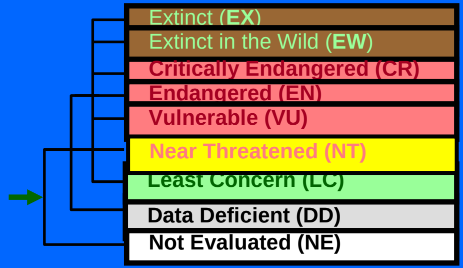
Are our bats?


DEFINITIONS
of the
IUCN
Categories
Extinct (EX)
Extinct in the Wild (EW)
Critically Endangered (CR)
Endangered (EN)
Vulnerable (VU)
THREAT
Near Threatened (NT)
Least Concern (LC)
Data Deficient (DD)
Not Evaluated (NE)
The abbreviation of each category (in parenthesis) should follow the English denominations even when translated into other languages)



Data Deficient (DD)

A taxon is Data Deficient when there is inadequate information to make a direct, or indirect, assessment of its risk of extinction based on its distribution and/or population status.

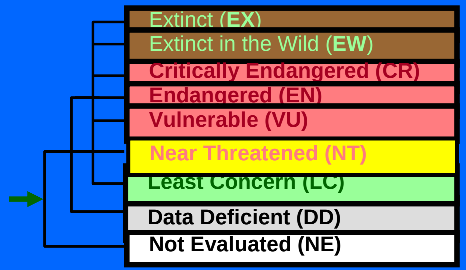


Extinct (EX)

A taxon is Extinct when there is no reasonable doubt that the last individual has died.

A taxon is presumed Extinct when exhaustive surveys in known and/or expected habitat, at appropriate times (diurnal, seasonal, annual), throughout its historic range have failed to record an individual. Surveys should be over a time frame appropriate to the taxon's life cycle and life form.

Raphus cucullatus = *Dodo*

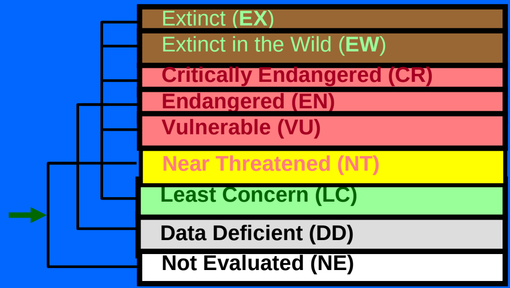


Extinct in the Wild (EW)

A taxon is Extinct in the Wild when it is known only to survive in cultivation, in captivity or as a naturalized population (or populations) well outside the past range.

A taxon is presumed Extinct in the Wild when exhaustive surveys in known and/or expected habitat, at appropriate times (diurnal, seasonal, annual), throughout its historic range have failed to record an individual. Surveys should be over a time frame appropriate to the taxon's life cycle and life form.

Equus ferus przewalskii = Przewalski's horse

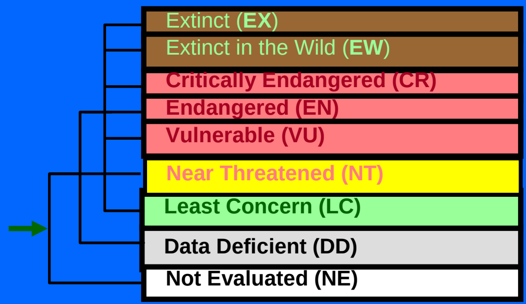


Least Concern (LC)

A taxon is Least Concern when it has been evaluated against the criteria and does not qualify for Critically Endangered, Endangered, Vulnerable or Near Threatened.

Widespread and abundant taxa are included in this category.

Bulbucus ibis = *Cattle egret*



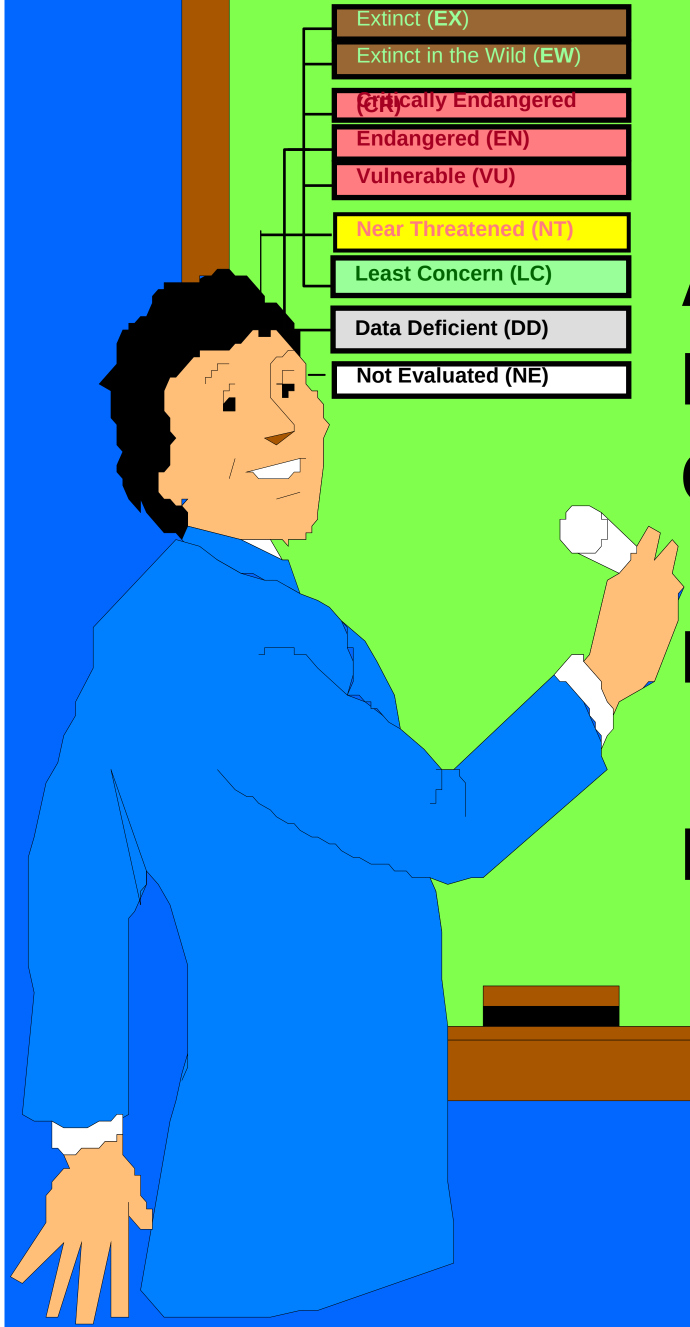
Near Threatened (NT)

A taxon is Near Threatened when it has been evaluated against the criteria but does not qualify for Critically Endangered, Endangered or Vulnerable now, but is close to qualifying for or is likely to qualify for a threatened category in the near future.

- Extinct (EX)
- Extinct in the Wild (EW)
- Critically Endangered (CR)
- Endangered (EN)
- Vulnerable (VU)
- Near Threatened (NT)
- Least Concern (LC)
- Data Deficient (DD)
- Not Evaluated (NE)

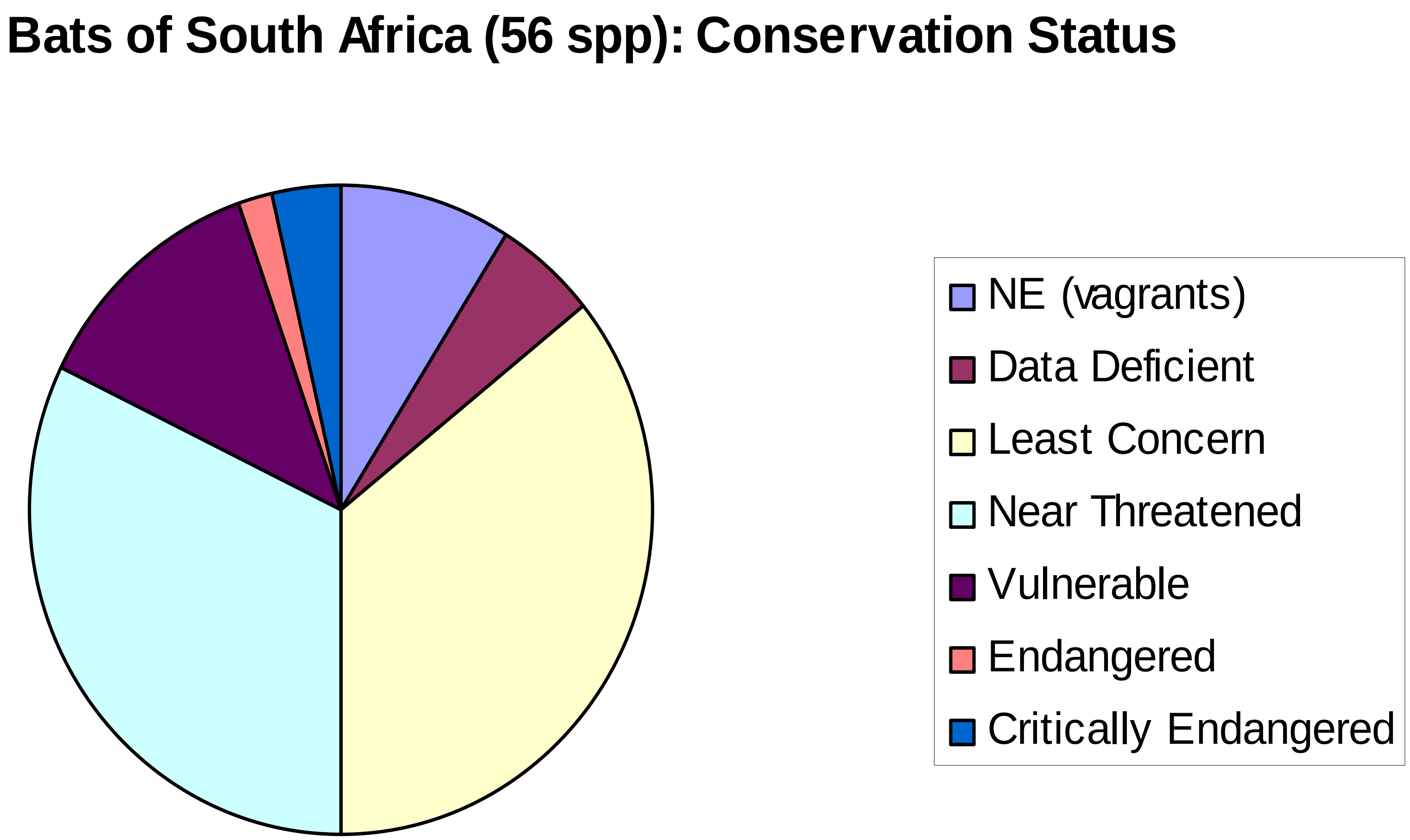
Categories of Threat

A taxon is CR; EN or VU when the best available evidence indicates that it meets any of the criteria A to E, and it is therefore considered to be facing an extremely high; very high or high risk of extinction in the wild.



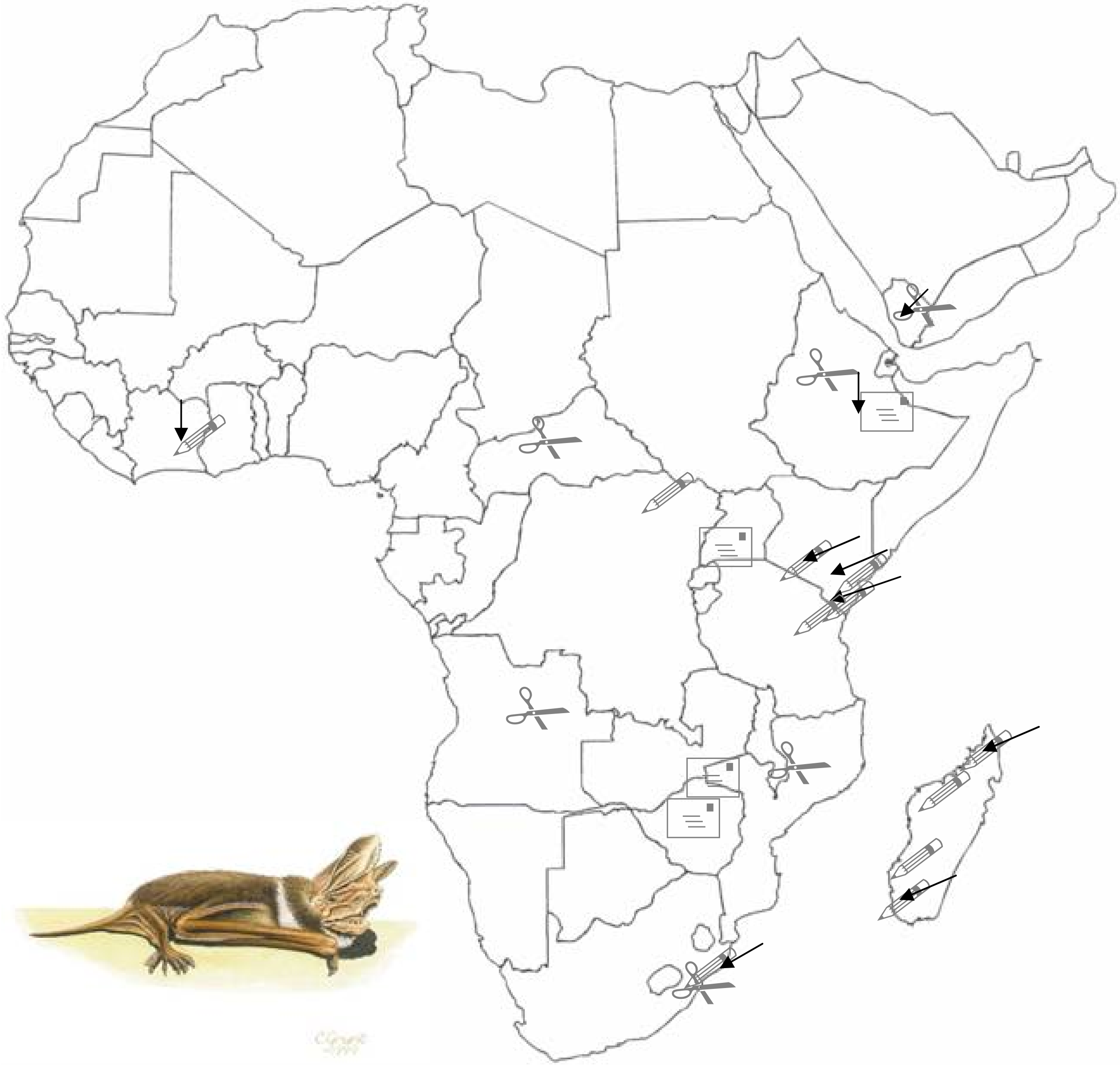
Criteria

A. Reduction in Population Size
B. Geographic Range
C. Small Population Size (+ decline)
D. Small Population Size (or Restricted Range)
E. Quantitative Analysis

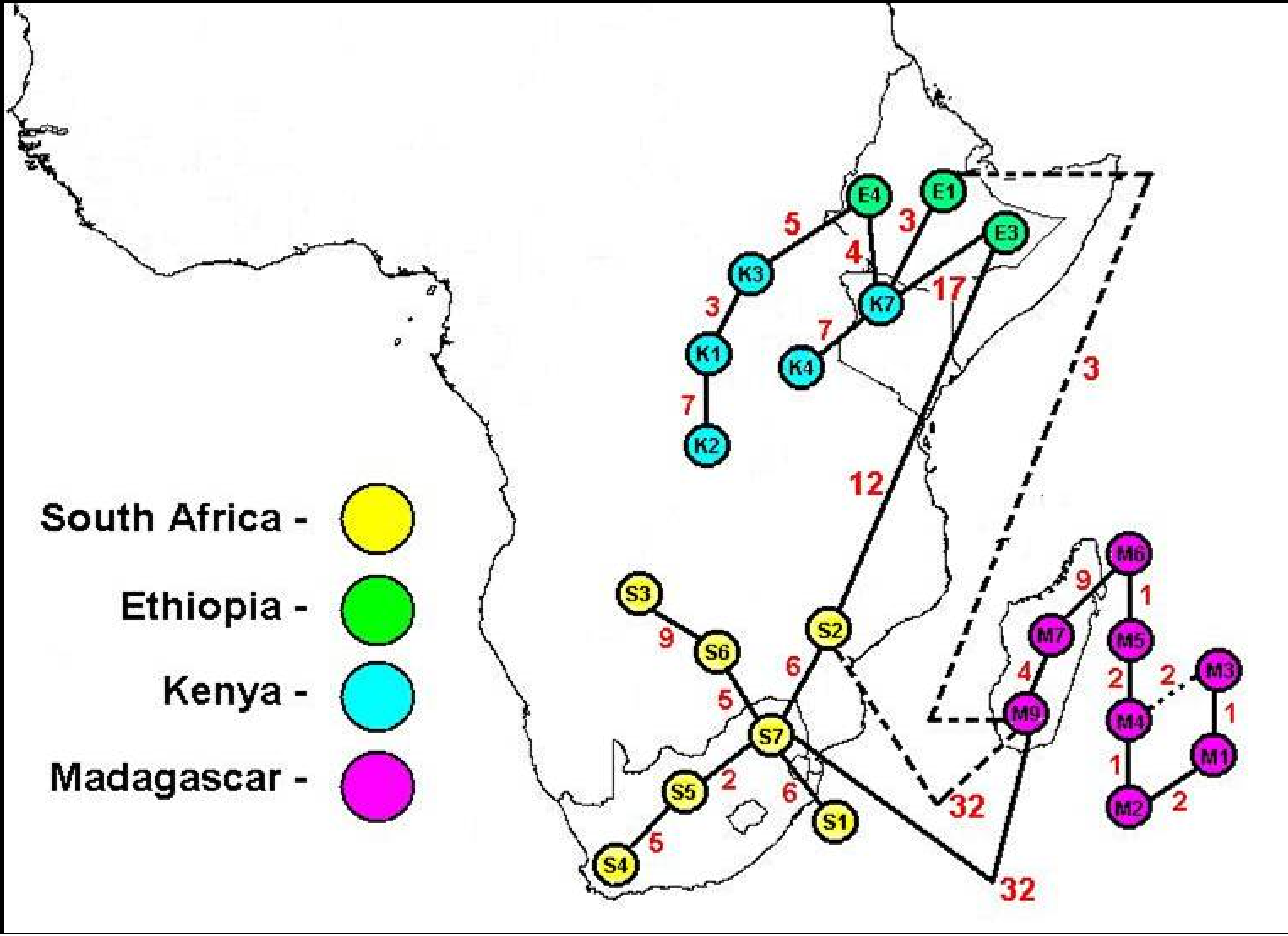

Bats of South Africa (56 spp): Conservation Status
NE (vagrants)
Data Deficient
Least Concern
Near Threatened
Vulnerable
Endangered
Critically Endangered


CR (B2,a,biii): Rendall's serotine

VU (D2): Large-eared free-tailed bat

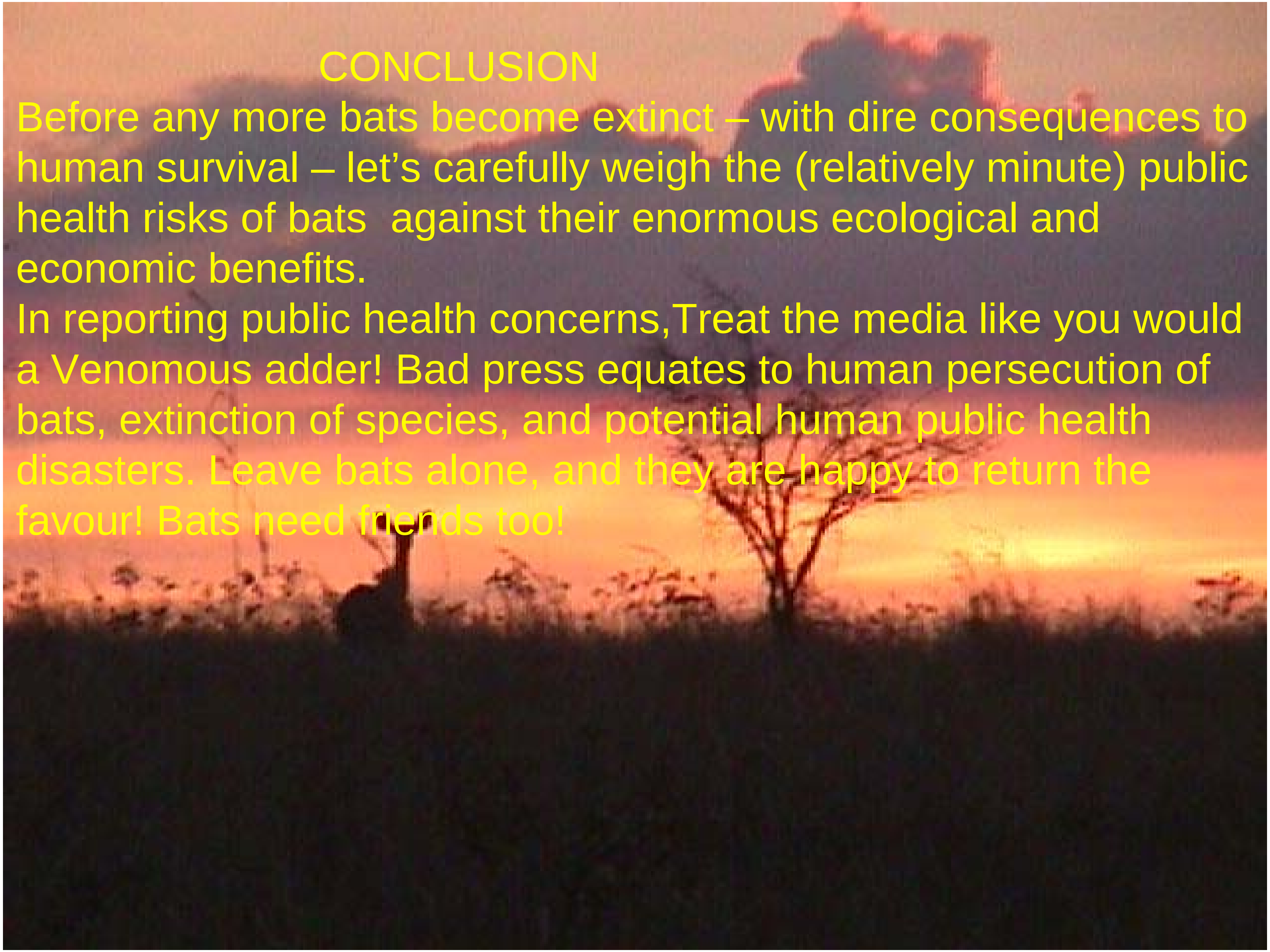


Minimum Spanning Network – Cytochrome b



VU (D2): Thomas' house bat

Extinct? Natal free-tailed bat



CONCLUSION

Before any more bats become extinct – with dire consequences to human survival – let’s carefully weigh the (relatively minute) public health risks of bats against their enormous ecological and economic benefits.

In reporting public health concerns,Treat the media like you would a Venomous adder! Bad press equates to human persecution of bats, extinction of species, and potential human public health disasters. Leave bats alone, and they are happy to return the favour! Bats need friends too!

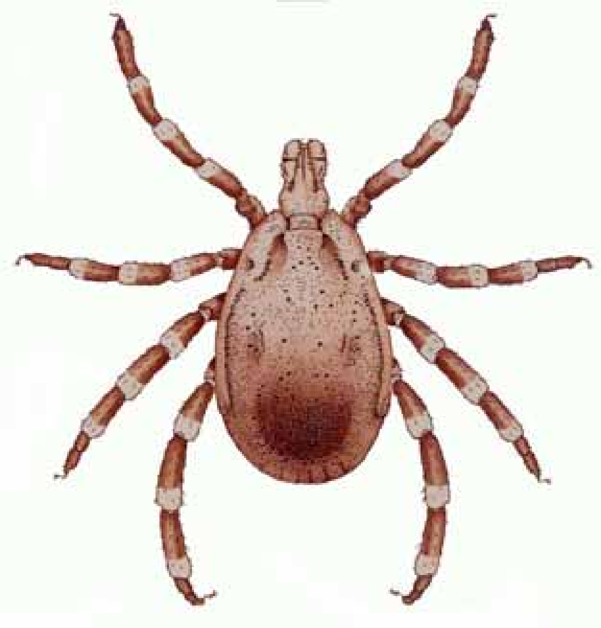
Acknowledgements

IMAGE CREDITS:

Bat Interest Group of KwaZulu-Natal
George Del Corral
Dr M. Brock Fenton
Christine Grant
Dr Ara Monadjem
Kate Richardson
Dr Merlin Tuttle / Bat Conservation International
Umsinsi Press
Proc. of the National Academy of Sciences
Nature

FILOVIRUSES AND BATS

R SWANEPOEL
SPECIAL PATHOGENS UNIT
NATIONAL INSTITUTE FOR COMMUNICABLE DISEASES
SOUTH AFRICA



EBOLA FEVER OUTBREAKS

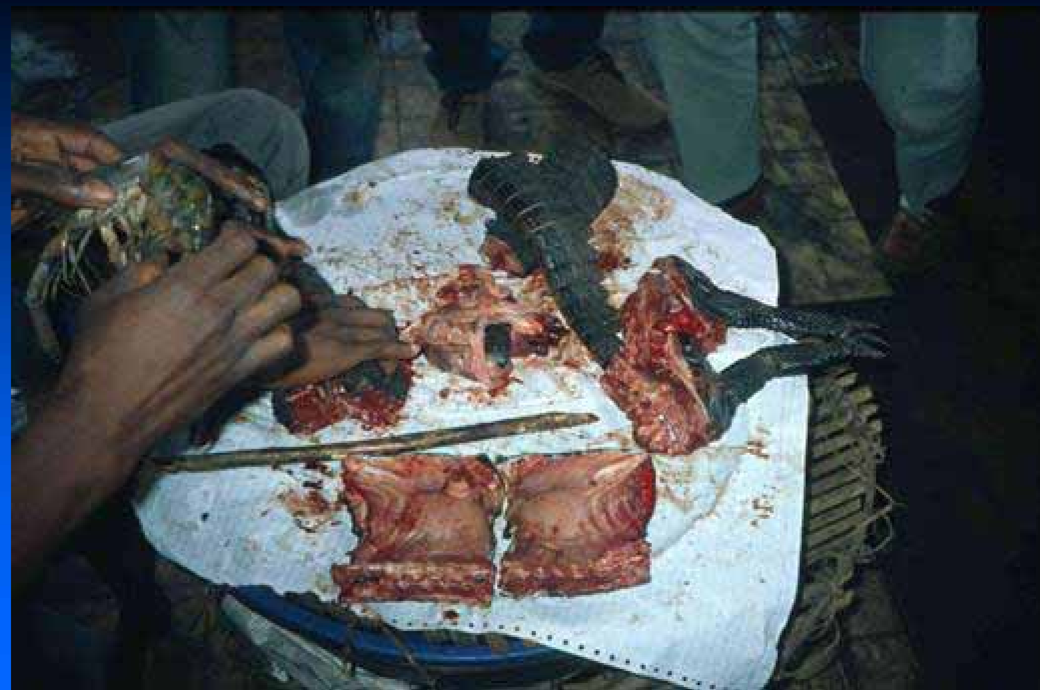
(EXCLUDING RESTON VIRUS)

YEAR	LOCATION	VIRUS TYPE	DEATHS/CASES (%)
1976	YAMBUKU, ZAIRE	EBOLA ZAIRE	280/318 (88%)
1976	NZARA, SUDAN	EBOLA SUDAN	151/284 (53%)
1979	MARIDI, SUDAN	EBOLA SUDAN	22/34 (65%)
1994	TAI FOREST, IVORY COAST	EBOLA IVORY COAST	0/1
1994	NOUNA RIVER, GABON	EBOLA ZAIRE	29/44 (66%)
1995	KIKWIT, ZAIRE	EBOLA ZAIRE	245/317 (77%)
1996	MAYIBOUTH, GABON	EBOLA ZAIRE	21/37 (57%)
1996	BOOUE, GABON	EBOLA ZAIRE	45/60 (75%)
1996	JOHANNESBURG, SOUTH AFRICA	EBOLA ZAIRE	1/2
2000-1	GULU, UGANDA	EBOLA SUDAN	224/425 (53%)
2001-3	GABON & CONGO-BRAZZAVILLE	EBOLA ZAIRE	306/386 (79%)
2005	CONGO-BRAZZAVILLE	EBOLA ZAIRE	12/13

















MARBURG DISEASE OUTBREAKS

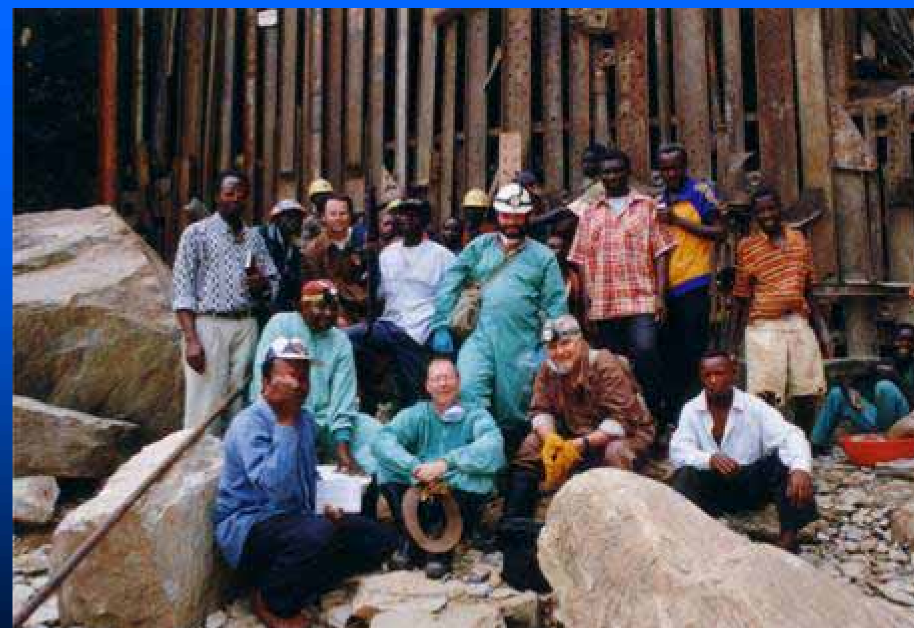
YEAR	LOCATION	DEATHS/CASES
1967	GERMANY, YUGOSLAVIA	7/32
1975	ZIMBABWE, S AFRICA	1/3
1980	W KENYA	1/2
1987	W KENYA	1/1
1998-2000	N-E DEM REP CONGO	126/154
2005	ANGOLA	329/374









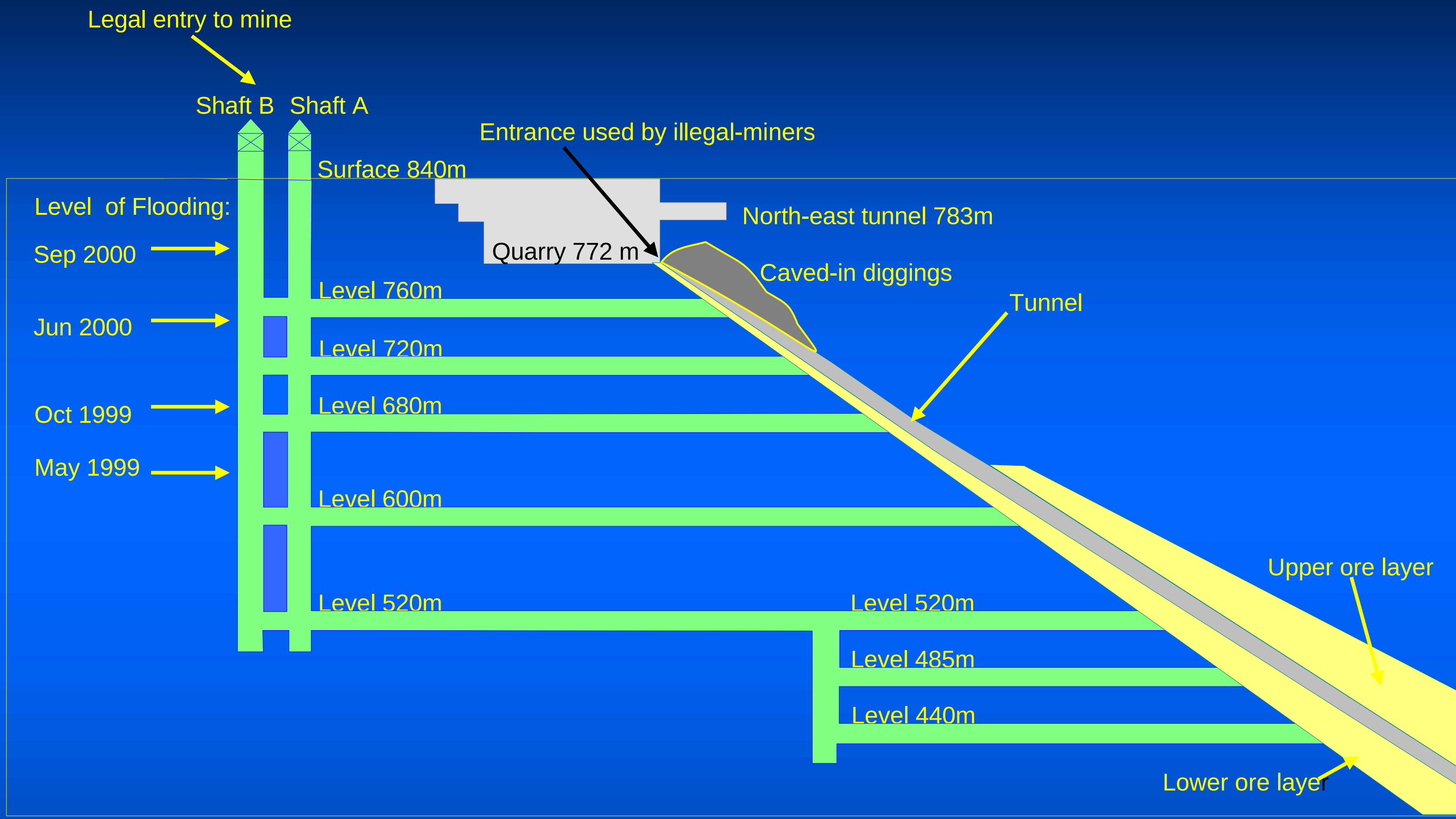
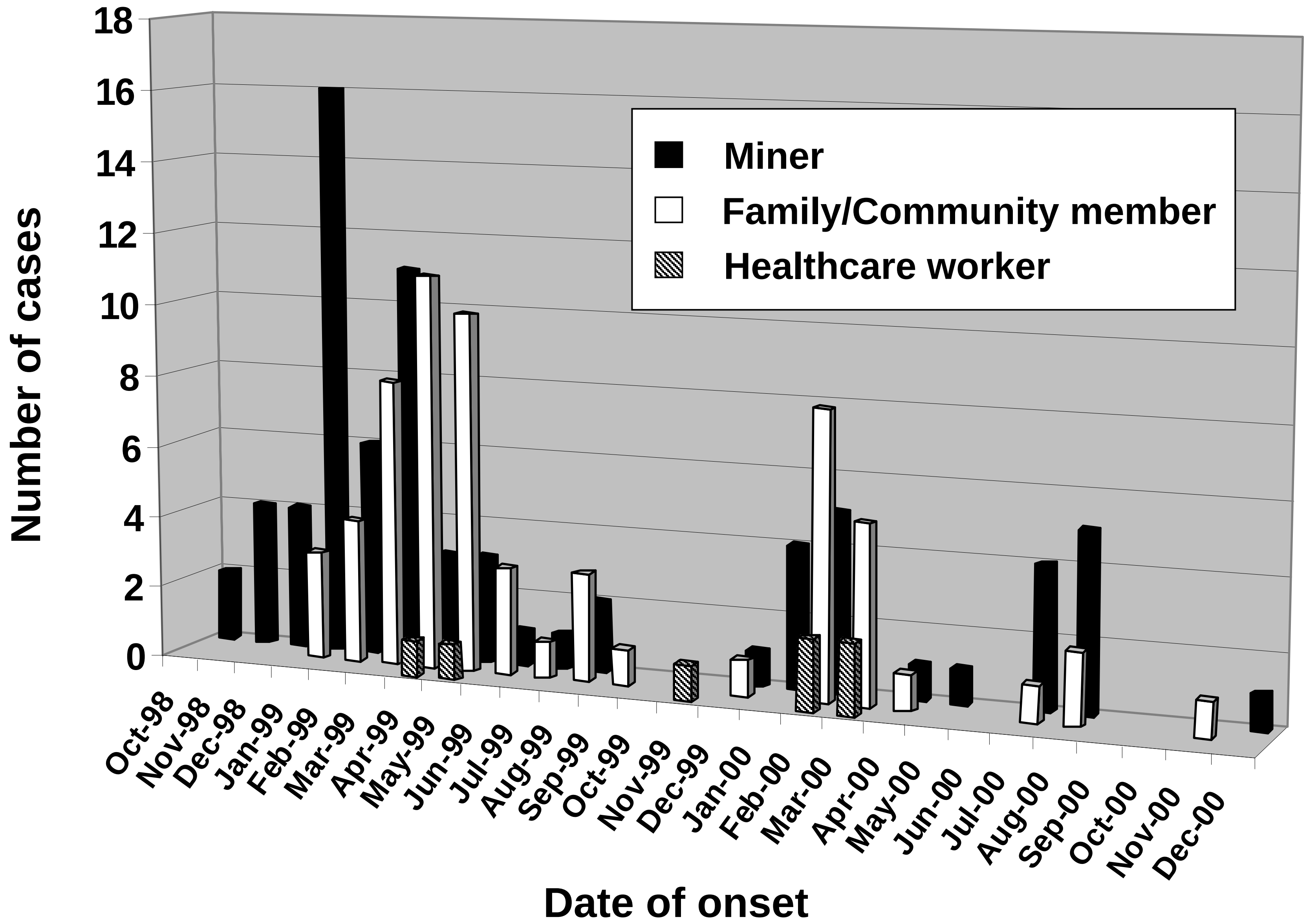
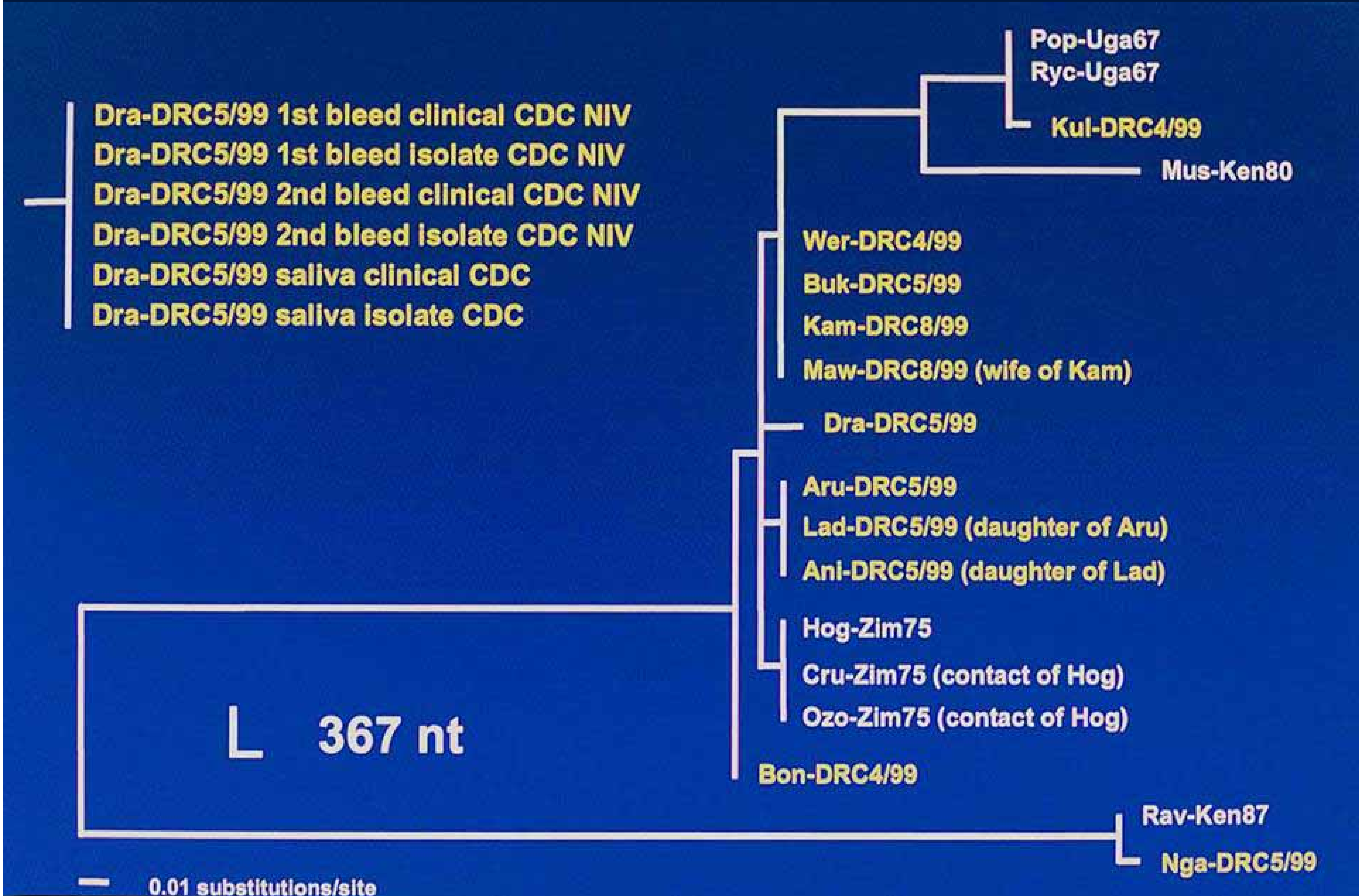


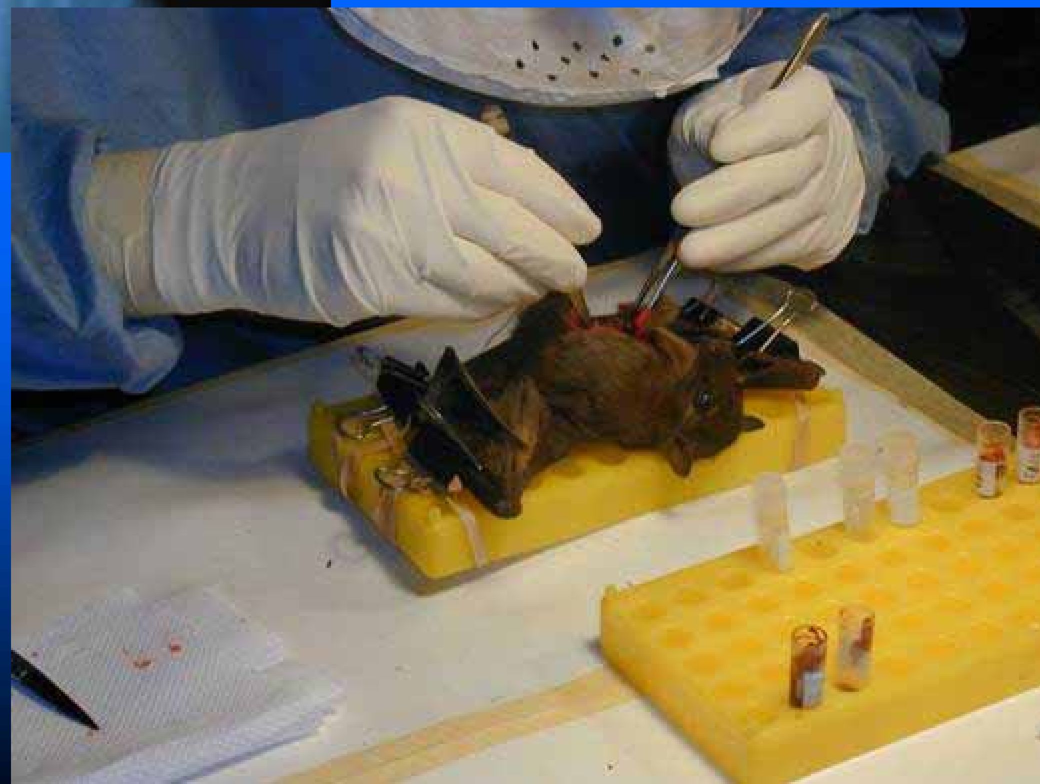
Diagram of Gorumbwa mine. Depth levels are in meters above sea-level.

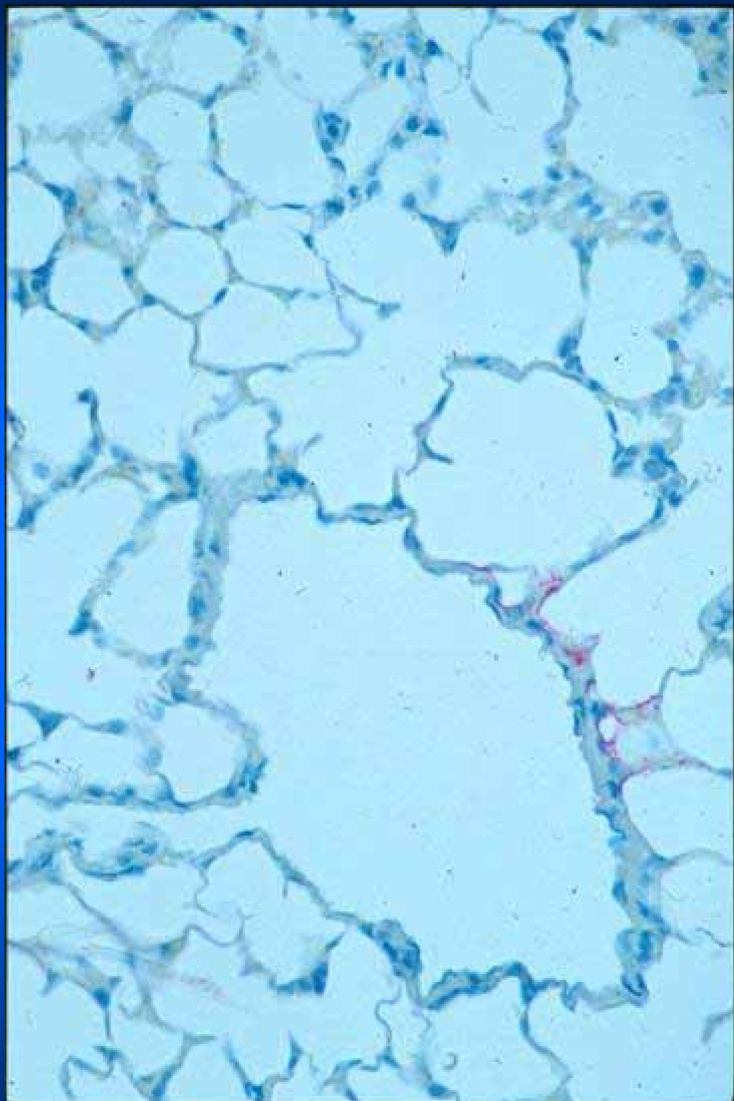


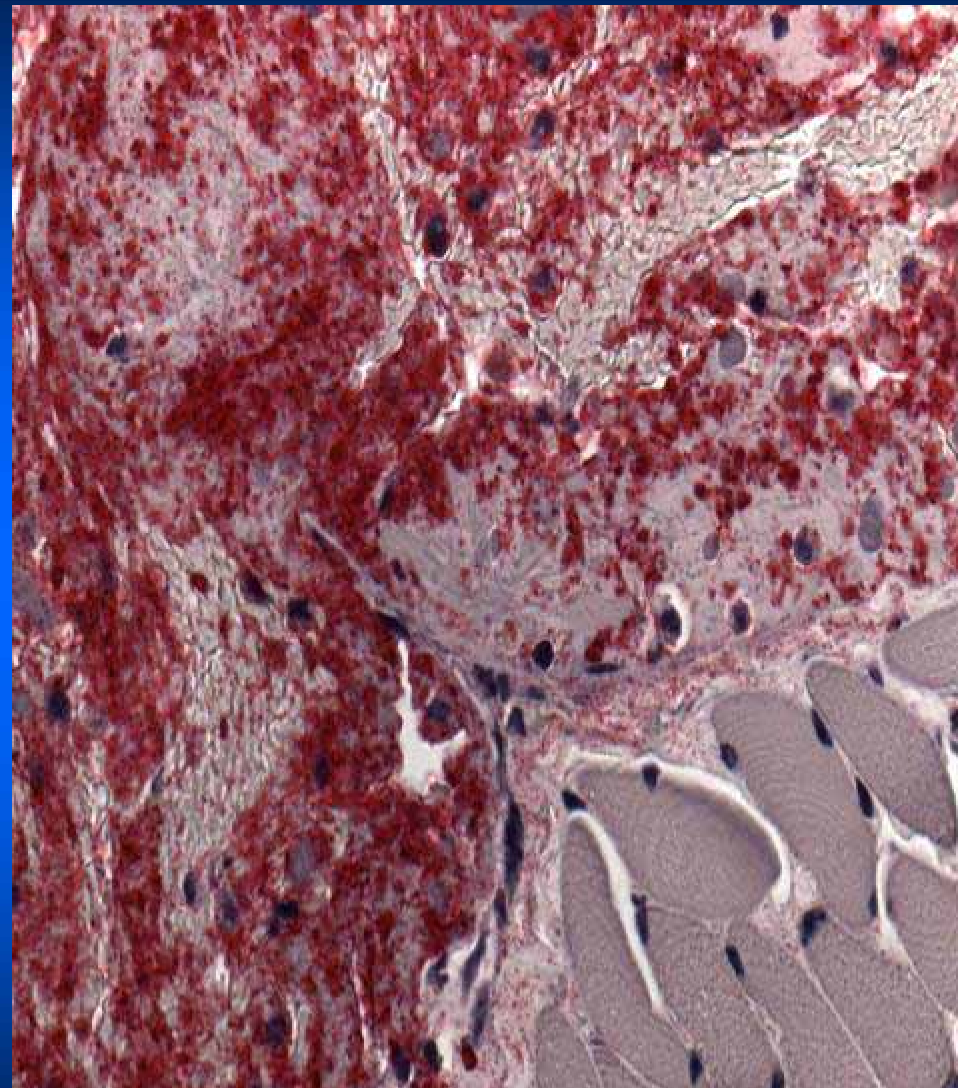
Miner
Family/Community member
Healthcare worker
Number of cases
0
2
4
6
8
10
12
14
16
18
Oct-98
Nov-98
Dec-98
Jan-99
Feb-99
Mar-99
Apr-99
May-99
Jun-99
Jul-99
Aug-99
Sep-99
Oct-99
Nov-99
Dec-99
Jan-00
Feb-00
Mar-00
Apr-00
May-00
Jun-00
Jul-00
Aug-00
Sep-00
Oct-00
Nov-00
Dec-00
Date of onset


Pop-Uga67
Ryc-Uga67
Kul-DRC4/99
Mus-Ken80
Dra-DRC5/99 1st bleed clinical CDC NIV
Dra-DRC5/99 1st bleed isolate CDC NIV
Dra-DRC5/99 2nd bleed clinical CDC NIV
Dra-DRC5/99 2nd bleed isolate CDC NIV
Dra-DRC5/99 saliva clinical CDC
Dra-DRC5/99 saliva isolate CDC
Wer-DRC4/99
Buk-DRC5/99
Kam-DRC8/99
Maw-DRC8/99 (wife of Kam)
Dra-DRC5/99
Aru-DRC5/99
Lad-DRC5/99 (daughter of Aru)
Ani-DRC5/99 (daughter of Lad)
Hog-Zim75
Cru-Zim75 (contact of Hog)
Ozo-Zim75 (contact of Hog)
L 367 nt
Bon-DRC4/99
Rav-Ken87
Nga-DRC5/99
0.01 substitutions/site


GEOLOGIE





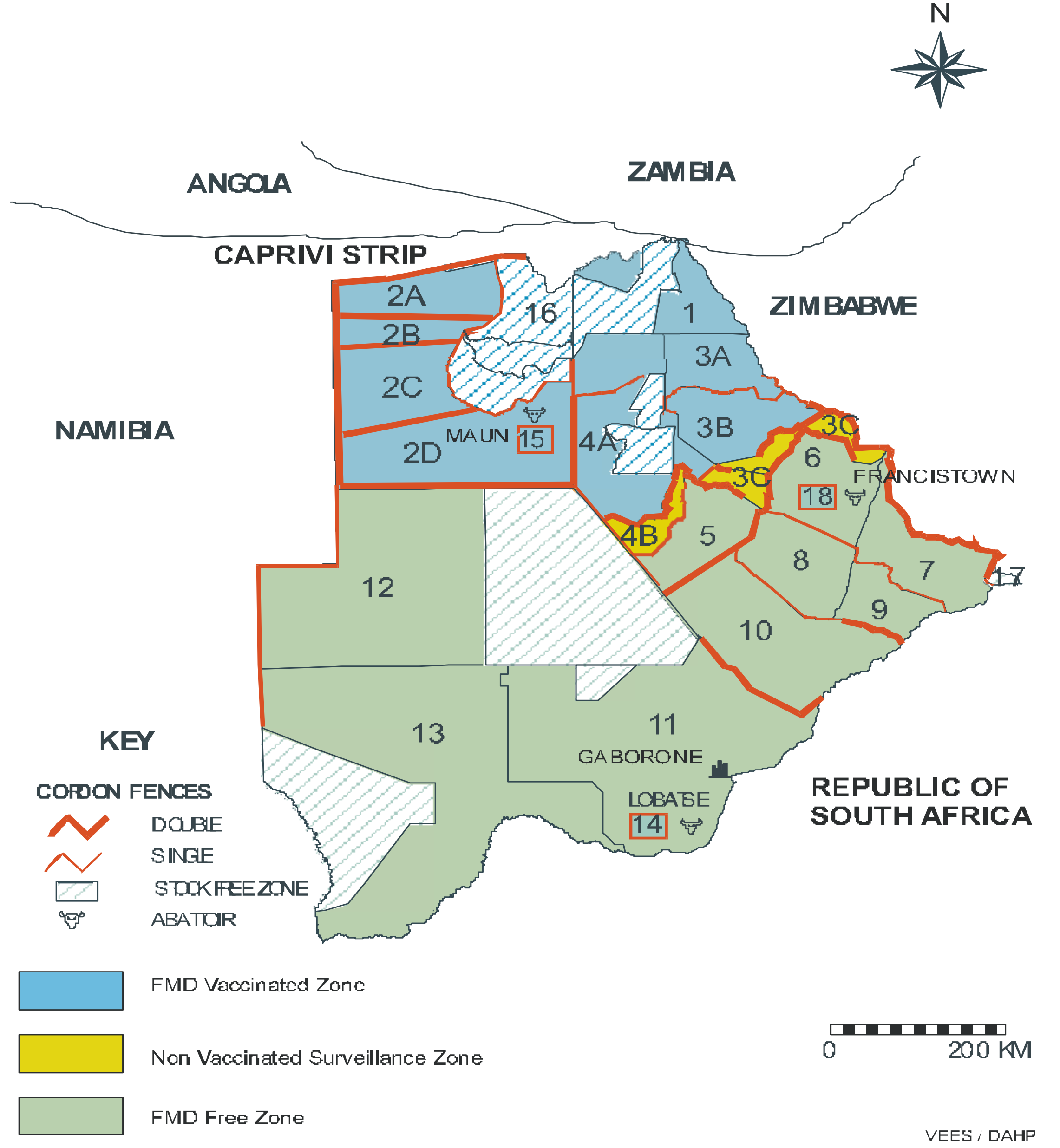
RABIES REPORT 2004 – 2005: BOTSWANA

Dr G Thobokwe

Botswana National Veterinary Laboratory

8th SEARG MEETING – NAMIBIA, January 22 – 26, 2006

Botswana Veterinary Disease Control Zones



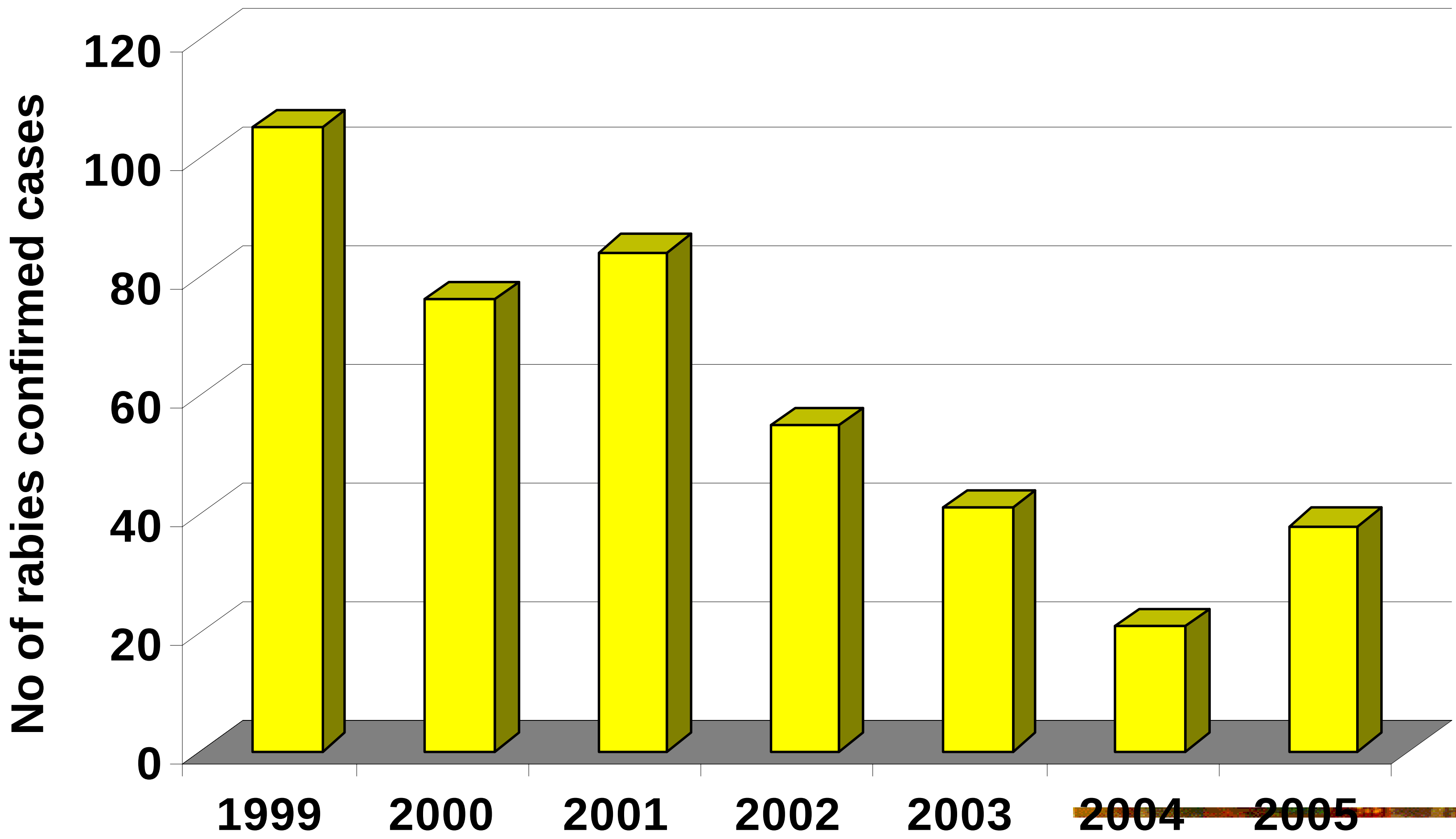
Intro

- 6 Regions and 17 Veterinary districts
- Veterinary districts sending samples to BNVL
- Distinctive red box used for rabies specimens
- In red box ½ brain(incldg brainstem) in 50% glycerol saline and ½ in 10% formalin
- Rabies notification form – species details,location human contacts

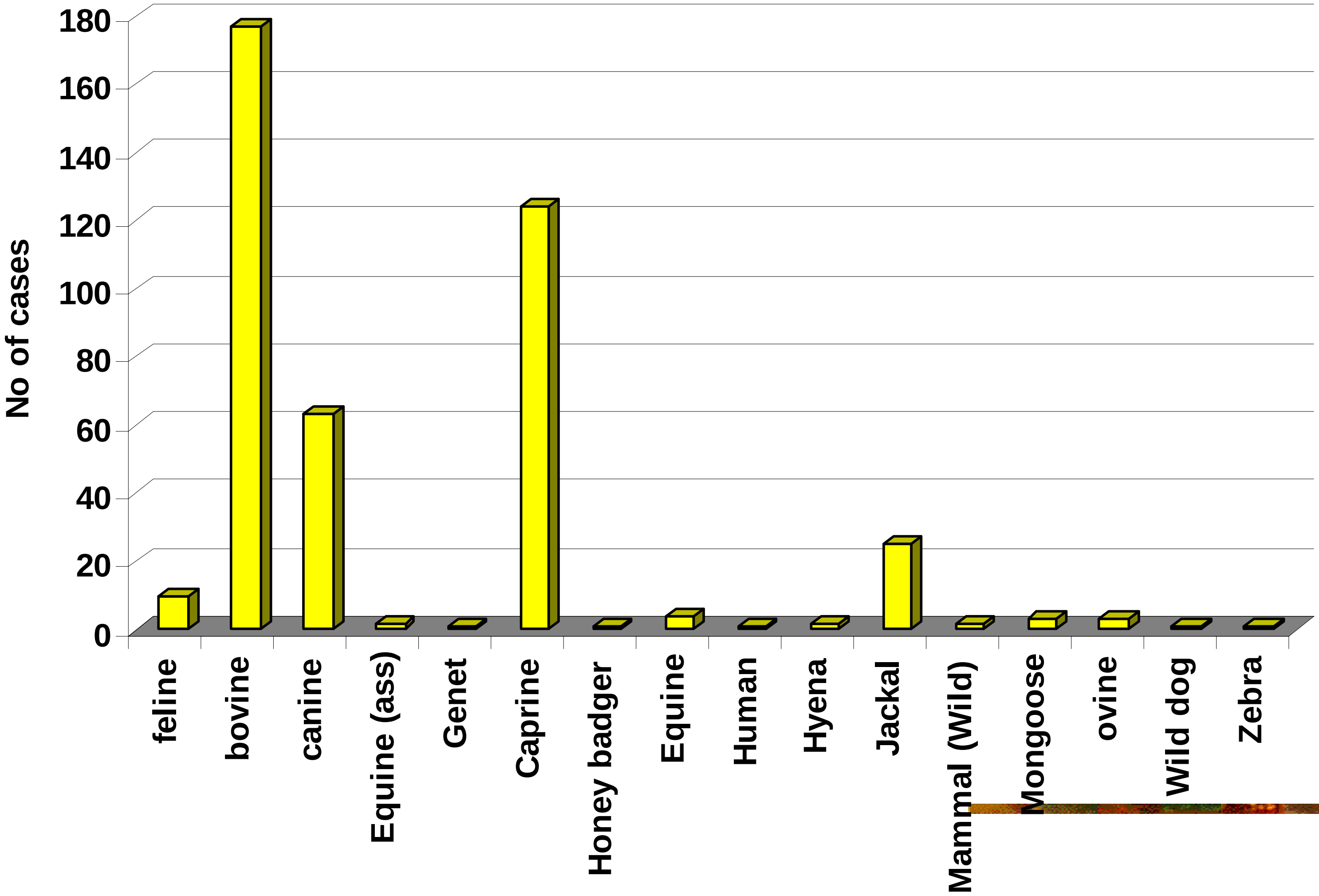
Diagnosis

- Std test is FAT – demonstrate rabies virus antigen by means of immunoflorescence
- Supplementary test – Mouse inoculation test (IC mouse inoculation)
- In addition – Histopathology mainly for Ddx and BSE in case of bovines

Overview 1999 to 2005 - cases



Species distribution – 99 to 2005



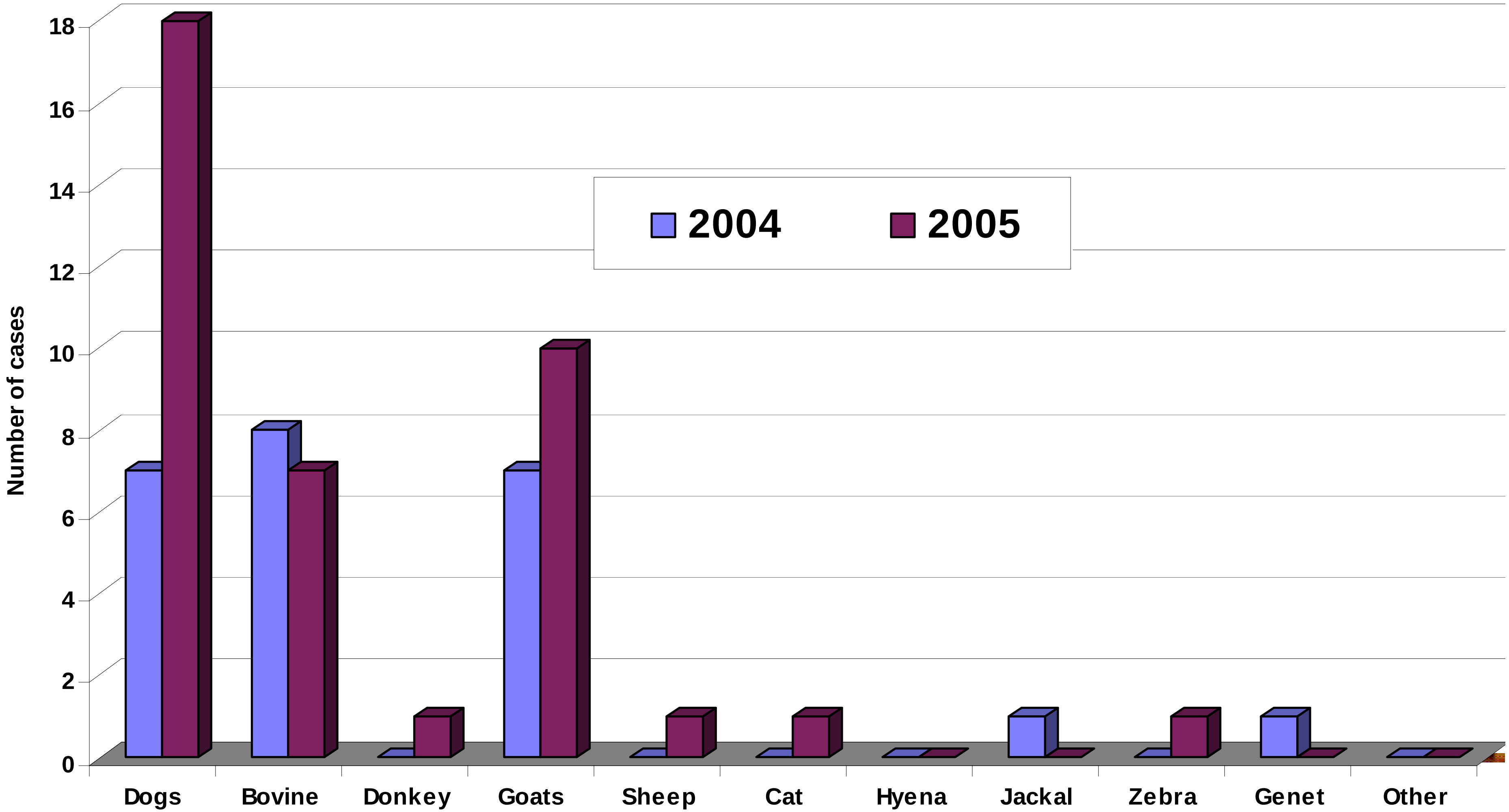
Overview 1999 to 2005

- cases have been significantly decreasing from 1999 to 2004
- slight peak of cases in 2005
- Bovine 45.1%, Caprine 29.5%, Canine 15%, Jackal 6% and Cats 2.4%
- Mongoose, genet, hyena and wild dogs
- Stray dog or dogs with no owners

2004 and 2005 cases

	2004			2005		
Species	Rabies pos	Rabies neg	RF of Positive cases (%)	Rabies pos	Rabies neg	RF of Positive cases (%)
Dogs	7	39	29.2%	18	19	46.2%
Bovine	8	19	33.3%	7	22	17.9%
Donkey	0	1	0.0%	1	0	2.6%
Goats	7	6	29.2%	10	4	25.6%
Sheep	0	1	0.0%	1	0	2.6%
Cat	0	0	0.0%	1	1	2.6%
Hyena	*0*	*4*	*0.0%*	*0*	*0*	*0.0%*
Jackal	*1*	*0*	*4.2%*	*0*	*1*	*0.0%*
Zebra	*0*	*0*	*0.0%*	*1*	*0*	*2.6%*
Genet	*1*	*1*	*4.2%*	*0*	*0*	*0.0%*
Other	*0*	*1*	*0.0%*	*0*	*1*	*0.0%*
Total	24	72		39	48	
%	25.0%	75.0%		44.8%	55.2%	

Species distribution 2004 to 2005



2004 and 2005

- Slight increase of rabies cases in 2005 compared to 2004 (mainly eastern part)
- Canine cases increased significantly in 2005, by more than 50%, while others remained relatively same
- Vaccination coverage? Stray dogs? Or Both

Genotypes

- No typing done from recent samples
- anti-nucleocapsid monoclonal antibodies and molecular phlogenetic typing done before (King et al 1993 and Johnson et al 2004).
- isolates in Botswana belong to only the classical rabies virus (genotype 1).

Genotypes ctd

- two biotypes of the lyssavirus 1 in Botswana, canid and viverrid
- Canid – dog and jackal?
- Viverrid – mongoose, genet cat, wild cat

Species ditribution of biotypes

Species	Type
Dog	*canid and viverrid*
Bovine	*canid and viverrid*
Goat	*canid and viverrid*
Horse	*canid*
Cat	*viverrid*
Human	*viverrid*
Mongoose	*viverrid*
Jackal	*canid*
Genet	*viverrid*
Duiker	*viverrid*
Wildcat	*viverrid*

Geographical distribution

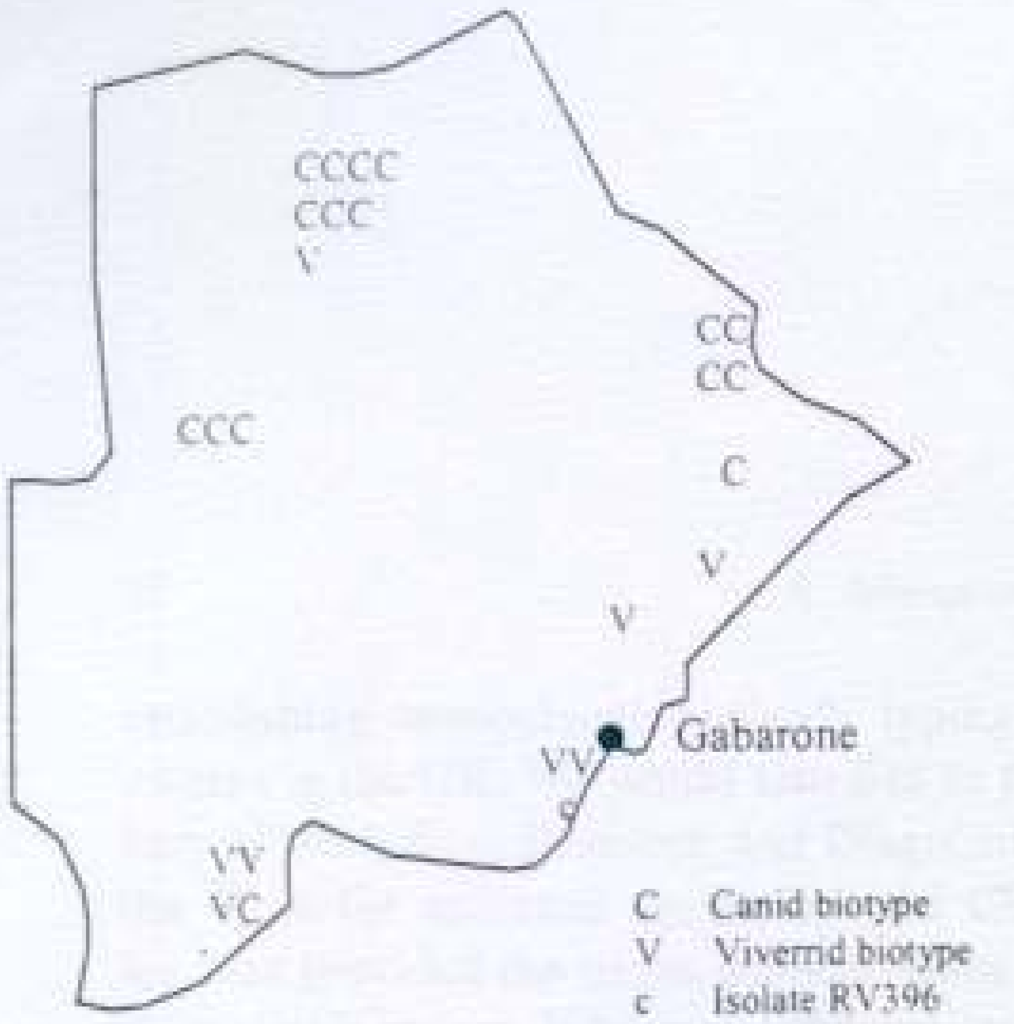


Fig. 3. Geographical distribution of canid and viverrid RABV biotypes within Botswana.

Bioytpes

- Canid – north and west
- Viverrid – south east
- Counterintuitive since population in south east
- Good vaccination coverage?
- More reporting of wild life?
- History – more movement of people and animals in north and west
- Jackals??

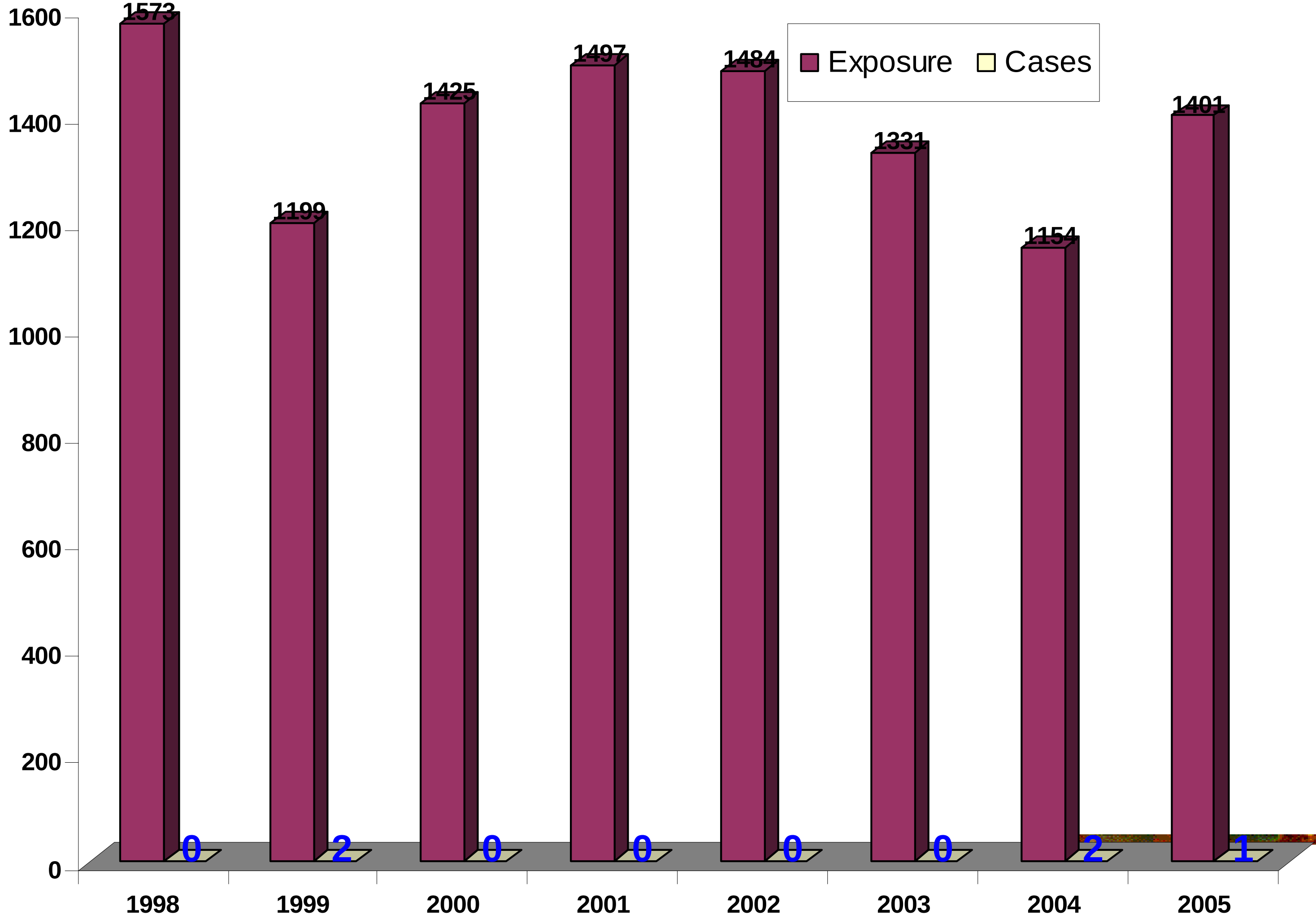
Human rabies

- Policy is if exposed, go to health facility, vet in ext area notified then investigation carried out
- Animal reported sick, rabies suspected, history of contacts taken and medical help advised
- Exposure – scratch, bite, handling and consumption of meat from/of suspected rabid animal

Human deaths 2004 to 2005

	2004	2005
deaths confirmed by laboratory tests	0	1
deaths diagnosed on clinical grounds only	2	0

Human rabies Exposures, 98-05



Exposures 2004 and 2005

	2004	2005
(bite) *exposure* cases reported during this period?	**1154**	**1401**
Total Number of people (bitten) *exposed (*by) *to* suspect (dogs) *animals* per/100 000:	**67**	**80**

Exposure by species

	2004		2005	
Species	**No cases**	**No human contacts**	**No cases**	**No human contacts**
Dogs	*7*	*11*	*16*	*30*
Cows	*10*	*57*	*15*	*69*
Sheep	*3*	*7*	*0*	*0*
Goats	*4*	*8*	*4*	*10*
Jackal	*0*	*0*	*4*	*7*
Donkey	*0*	*0*	*2*	*4*

Vaccines

	Animal Vaccine		Human Vaccine	
	2004	**2005**	**2004**	**2005**
0rdered	180 000	200 000		35000
Cost (P)	369000.00	410000.00		1 499 388.00
distributed	148730	178730		
Type	CC		CC	
Source	RSA		RSA	

Vaccines

- All people “exposed” given PEP
- Min of Health have an “IgG” for severe cases – deepbites, head and neck bites
- Although not monitored vaccine failure not experienced

Vaccination returns

	2004	**2005**
Dogs	114568	125608
Cats	4661	8758
Bovine	NA	177
sstock	NA	21
donkeys	NA	86
“Humans”	1154	1401

Comments

- Low number of cases (low prevalence) implies Vaccination effective
- Stray dog may be a problem
- Collaboration between Min of Health and DAHP to be enhanced
- Public education

Thank You – God bless

KENYA

COUNTRY REPORT EIGHTH SEARG BIANNUAL MEETING NAMIBIA, WINDHOEK, 22-26 JANUARY 2006

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Virology Section

Central Veterinary Laboratories, Kabete

%age of total submitted that were positive per species

SPECIES	NO. POSITIVE		NO. NEGATIVE		TOTAL	
	2004	2005	2004	2005	2004	2005
Canine	44 (60.3%)	28 (38.9%)	24	18	68	46
Bovine	18 (24.7%)	37 (51.4%)	3	0	21	37
Ovine	2 (2.7%)	1 (1.4%)	0	0	2	1
Caprine	3 (4.1%)	2 (2.8%)	0	0	3	2
Feline	1 (1.4%)	1 (1.4%)	1	0	2	1
Equine	5 (6.8%)	2 (2.8%)	4	1	9	3
Misc	0	1 (1.4%)	2	0	2	1
TOTALS	73	72	34	20	107	92

Percentage Positive per samples submitted for each species

SPECIES	NO. POSITIVE		TOTAL	
	2004	2005	2004	2005
Canine	44 (64.7%)	28 (60.9%)	68	46
Bovine	18 (85.7%)	37 (100%)	21	37
Ovine	2 (100%)	1 (100%)	2	1
Caprine	3 (100%)	2 (100%)	3	2
Feline	1 (50%)	1 (100%)	2	1
Equine	5 (55%)	2 (66.7%)	9	3
Misc	0	1 (100%)	2	1
TOTALS	73 (68.2%)	72 (78.3%)	107	92

1.1.: RABIES PREVALENCE

- The number of canine rabies cases decreased from 44 in 2004 to 28 in 2005.This decrease is probably due to the decrease in the samples submitted (46 in 2005 compared to 68 in 2004 rather than a decrease in canine rabies).

- There was an increase in the prevalence of bovine rabies (37 in 2005 compared to 18 in 2004). This is again probably due an increase in the number of bovine cases submitted which in turn may probably have arisen from the high economic value of the cattle in the affected farms.

1.2.: RABIES GENOTYPES

- Genotyping of the lyssavirus has not been attempted in this country and therefore the genotypes that exist are not known. Proposals are in place to develop mechanisms to carry out antigenic and genetic characterization of rabies virus isolates that occur in Kenya.

2. HUMAN RABIES

- Although diagnostic facilities available at CVL – Kabete
- Human rabies almost always diagnosed on clinical grounds only
- Very poor record keeping even for the clinical cases.
- Figures of human dog bites very scanty, where available.
- Plans for more collaboration between between veterinary and human health institutions underway to enable the collection of such reports in future and to help improve on the number of cases confirmed in the laboratory.
- Few or no samples submitted to the lab for rabies testing from areas with significant dog bite reports and hence the suspected gross underreporting.

HUMAN RABIES contd.

2.1.: Source of Human Rabies

- The most likely source of human rabies in the country is the dog- usually stray. Nearly all the human cases are as a result of dog bite.
- At least one person is known to have been bitten by a hyena that was positive for rabies.

2.2.:Contribution of Stray Dogs

- The stray (unrestricted) dog is the major contributor of rabies both to other domestic animals and to man.

3. VACCINE USE

3.1.: Vaccine Production

- No veterinary or human vaccine is produced in the country.
- All the animal vaccine used in the country is imported, mainly from South Africa

3.2.: Animal Rabies Vaccine Usage

- The main rabies vaccine distributor imported 250,000 doses in 2004 and 350,000 in 2005, and supplies about 70% of the vaccine (personal communication).

Vaccine type	2004	Cost (USD)	2005	CostUSD
Tissue culture	350,000doses	110,000	500,000doses	160,000

VACCINE USE contd.

3.3.: Dog population and Vaccination dynamics:

- Actual figures of animal vaccinations are difficult to obtain.

- If all the vaccine imported was used, then in 2004, 350,000 dogs were vaccinated and 500,000 dogs in 2005.

- Estimated total dog population using a dog:human population ratio of 1:7.7 is about 4.3million.

VACCINE USE contd.

3.4.: Post Exposure Treatment:

- The figures for persons who received vaccine for post exposure treatments is not available. Figures from the Department of Public Health in the Ministry of Health indicate an average of 10,000 doses of human anti-rabies vaccine per year.

- WHO estimates that 11 persons are bitten by dogs in every 1 million people, which translates to 343 dog bites using a human population of 33 million Kenyans.

VACCINE USE contd.

3.5: No reports of vaccine failures have been received.

3.6: The obstacles to rabies vaccination include:

- Dwindling of financial resources (The government issued only 25,000 doses in 2005 ,100,000 doses in 2004– personal communication)
- High number of stray dogs.
- Dog population dynamics (very high puppy turnover).
- Little attachment to dogs by most owners..

ACKNOWLEDGEMENTS

- Veterinary field staff (public & private)
- Staff, Central Veterinary Laboratories, Kabete
- University of Nairobi (Dr. Kitala)
- Southern and Eastern African Rabies Group
- Director of Veterinary Services - Dr. J. Musaa
- Dr. Joseph M. Macharia, CVL Kabete
- Prof. Louis Nel, SEARG
- Dr. Claude T. Sabeta , SEARG

THANKS

RABIES SITUATION IN MOZAMBIQUE

2004 -2005

ANIMAL RABIES

- METHODS FOR DIAGNOSIS
  - SLIDE TEST (SELLER´S STAINING)
  - IMMUNOLOGICAL (FAT)
  - BIOLOGICAL (MOUSE INOCULATION TEST)
- NUMBER OF SPECIMENS
  - 2004 (77)
  - 2005 (84)

NUMBER OF SPECIMENS OF DOMESTIC ANIMALS SUBMITTED

Domestic animals	2004		2005	
	Rabies pos.	Rabies neg.	Rabies pos.	Rabies neg.
Dogs	14	58	16	54
Cats	2	2	2	7
Ruminants	1		1	1
Swine			1	2
Total (%)	17 (22%)	60 (77.9%)	20 (23.8%)	64 (76.2%)

NUMBER OF WILD ANIMALS SUBMITTED

Wild animals submitted	2004		2005	
	Rabies pos.	Rabies neg.	Rabies pos.	Rabies neg.
Monkeys			1	
Total (%)				

Animal rabies prevalence trend

- There was a slight increase in 2005 compared to the previous year
- This can be attributed to the low vaccination coverage

LYSSAVIRUS GENOTYPES

- The virus isolates cannot be assigned to the respective lyssavirus genotypes, because the genotype classification has not done yet been done.
- However, the isolates are being kept for that purpose

Human rabies deaths

Total nº of human deaths confirmed by laboratory tests (if any)	2004	2005
	-------	--------
Total nº of human rabies deaths diagnosed on clinial grounds only	29	35

SOUCES OF HUMAN RABIES CASES

	2004	2005
Domestic dogs		
Other domestic animals were involved	------	------
Wild carnivore species were involved	------	------
Bats		
Bite cases were reported	1369	2443
Total number of people bitten by suspect dogs per/100 000	0.014	0.024

Contribution of stray dogs to the overall rabies cases

- The system of data collection does not permit the information on the contribution of stray dogs to the overall rabies cases to be obtained
- However the majority of bitten people receive post-exposure treatment because the responsible animals can not be kept under observation

Vaccine use in the control of rabies

- The vaccine used is imported (animal & human)
- The type of vaccine is Rabisin-inactivated
- The main institutions responsible for the importation are: Ministry of Agriculture and Ministry of Health

Rabies vaccine imported and respective cost

	2004		2005	
	Human	Animal	Human	Animal
Doses imported	20,000	79,000	1,000*	45,000
Cost (US Dollars)	-----	30,810	37,531	17,550

Dog population and vaccinated ones

	2004	**2005**
Vaccinated dogs	66,995	43,494
Estimated dog population	805,000	805,000
Vaccination (%)	8.3	5.4

Main problems faced in vaccination campaigns

- Weak involvement of the municipalities authorities
- Weak public awareness
- Low numbers of vaccinators in the rural areas
- High numbers of stray dogs
- There is not any sanction in relation in relation to the dogs which do not show up in the vaccination campaigns
- Financial constraints

Post-exposure treatment and bites

	2004	**2005**
Post-exposure treatment	1369	2443
Bites	1369	2443

Vaccine failures

- Across the country there is no occurrence of vaccine failure has been recorded
- If it does happen, it can be attributed to the difficulties in keeping the vaccine in cold places in remote areas

Other obstacles to rabies vaccination

- In some remote areas some village hunters refuse to have their dogs vaccinated due to a belief that the vaccine would impact negatively on the dog´s hunting abilities.

THANK YOU

SEARG

2004 - 2005

NAMIBIA

COUNTRY REPORT

RABIES: DOMESTIC ANIMALS

Species	2004				2005			
	No. of specimens	Pos.		Neg.	No. of specimens	Pos.		Neg.
		Number	%			Number	%	
Canine	**186**	**72**	**45.6**	**114**	**373**	**179**	**59.5**	**194**
Feline	**32**	**19**	**12.0**	**13**	**22**	**8**	**2.7**	**14**
Bovine	**128**	**53**	**33.5**	**75**	**162**	**79**	**26.2**	**83**
Caprine	**17**	**9**	**5.7**	**8**	**40**	**23**	**7.6**	**17**
Ovine	**8**	**2**	**1.3**	**6**	**22**	**10**	**3.3**	**12**
Equine	8	1	0.6	7	5	1	0.3	4
Porcine	3	2	1.3	1	5	1	0.3	4
TOTAL	**382**	**158**	**100.0**	**224**	**629**	**301**	**100.0**	**328**

RABIES: WILDLIFE

Species	2004				2005			
	No. of specimens	Pos.		Neg.	No. of specimens	Pos.		Neg.
		Number	%			Number	%	
Jackal	**14**	**10**	**26.3**	**4**	**21**	**15**	**33.3**	**6**
Honey Badger	2	1	2.6	1	2	1	2.2	1
Bat ear Fox	4	4	10.5	0	0	0	0.0	0
Cheetah	2	1	2.6	1	0	0	0.0	0
Leopard	0	0	0.0	0	1	0	0.0	1
Wild Dog	0	0	0.0	0	1	1	2.2	0
Meercat	9	0	0.0	9	2	1	2.2	1
Suricate	0	0	0.0	0	4	0	0.0	4
Mouse	0	0	0.0	0	3	0	0.0	3
Baboon	2	0	0.0	2	2	0	0.0	2
Kudu	**30**	**21**	**55.3**	**9**	**48**	**26**	**57.8**	**22**
Eland	2	1	2.6	1	1	1	2.2	0
Springbok	0	0	0.0	0	3	0	0.0	3
Steenbok	0	0	0.0	0	1	0	0.0	1
Damara dikdik	0	0	0.0	0	1	0	0.0	1
Zebra	0	0	0.0	0	1	0	0.0	1
TOTAL	**65**	**38**	**100.0**	**27**	**91**	**45**	**100.0**	**46**

RABIES: ANIMALS, SUMMARY

Species	2004				2005			
	No. of specimens	Pos.		Neg.	No. of specimens	Pos.		Neg.
		Number	%			Number	%	
Domestic	**382**	**158**	**80.6**	**224**	**629**	**301**	**87.0**	**328**
Wild	**65**	**38**	**19.4**	**27**	**91**	**45**	**13.0**	**46**
TOTAL	**447**	**196**	**100.0**	**251**	**720**	**346**	**100.0**	**374**

COMMENTS: PREVIOUS TABLE

- SHARP INCREASE IN DOGS IN BIG TOWNS - WINDHOEK, OSHAKATI, RUNDU. MANY LOW INCOME RESIDENTS + UNEMPLOYED.
- INCREASE IN CATTLE DUE TO WILDLIFE CYCLE: JACKAL/SURICATES

RABIES IN HUMANS

	2004	2005
Total no. of human rabies deaths confirmed	17	24
Domestic dogs: bites by rabid dogs.	28	32
Other animals involved	UNKNOWN	
Bats??	UNKNOWN	
Bite cases reported	54	60
Total Number of people bitten by suspect dogs per 100 000:	1.6	1.8

PROTOCOL AND PROBLEMS

- ANNUAL AWARENESS CAMPAIGNS.

  (IS IT EFFECTIVE ?)
- DOGS & CATS VACCINATED FREE OF CHARGE BY GOVERNMENT.

  (CHANGE PROTOCOL ?)

- STRAY DOGS NOT VACCINATED.
- SOME PEOPLE MINIMAL INTEREST IN CARING FOR THEIR PETS: NEED TO CHANGE ATTITUDE OF PEOPLE.

DOG POPULATION & VACCINATIONS

Dog population	Dogs vaccinated	
	2004	2005
250000	16827	63266

VACCINE IMPORTED

	Vaccine type	No. of doses	Cost N$	Cost US$ (N$1=US$6.5)
Imported for animals	Rabisin	270000	784000	120600
	Rhabdomune	40000	400000	61600
	Canigen LR	6000	290000	44600
	Urican	7600	36000	6000
TOTAL: ANIMALS		323600	1510000	232000
Imported for HUMANS	Verorab	6100	2136000	330000

OBSTACLES

- STRAY DOGS: NOT VACCINATED.
- ATTITUDE OF SOME PEOPLE: LOW VACC %.
- CATS DO NOT LIKE CARS: LOW VACC %.
- GAME FARMS LARGE: CARCASSES DECOMPOSED – NO DIAGNOSIS.
- COLD CHAIN DIFFICULT IN REMOTE AREAS.
- EFFECT OF ORAL VACCINE ON NON-TARGET ANIMALS / PEOPLE ? CONTROL OF DOSAGE ?
- FUNDS FOR VACCINE.

NAMIBIAN KUDU HISTORY

- Rabies outbreak in kudu **1978**, central & north western Namibia: 4% mortalities in kudu. **1980** = 44% of kudu population died in this area.
- 1980 to 1983 rabies swept through southern & eastern Namibia. Details unknown.
- National game census in 1982 indicated approximately 30 000 to 50 000 decrease in kudu population.
- 1991 kudu population returned to normal.

[Animal Health and Veterinary Medicine in Namibia
Herbert P. Schneider, 1994]

SUMMARY

- ACTIVE CYCLE IN KUDU IS ONGOING.
- WILDLIFE CYCLE ONGOING.
- SPILL OVER TO DOMESTIC ANIMALS.
- URBAN CYCLE ONGOING
- SPILL OVER TO HUMANS.

ACKNOWLEDGMENT

- Dr. Jack Vries: Ministry of Health and Social Services.
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- Mrs. Julia Shimwino: CVL

END

Presented by:
Dr. Frans Joubert

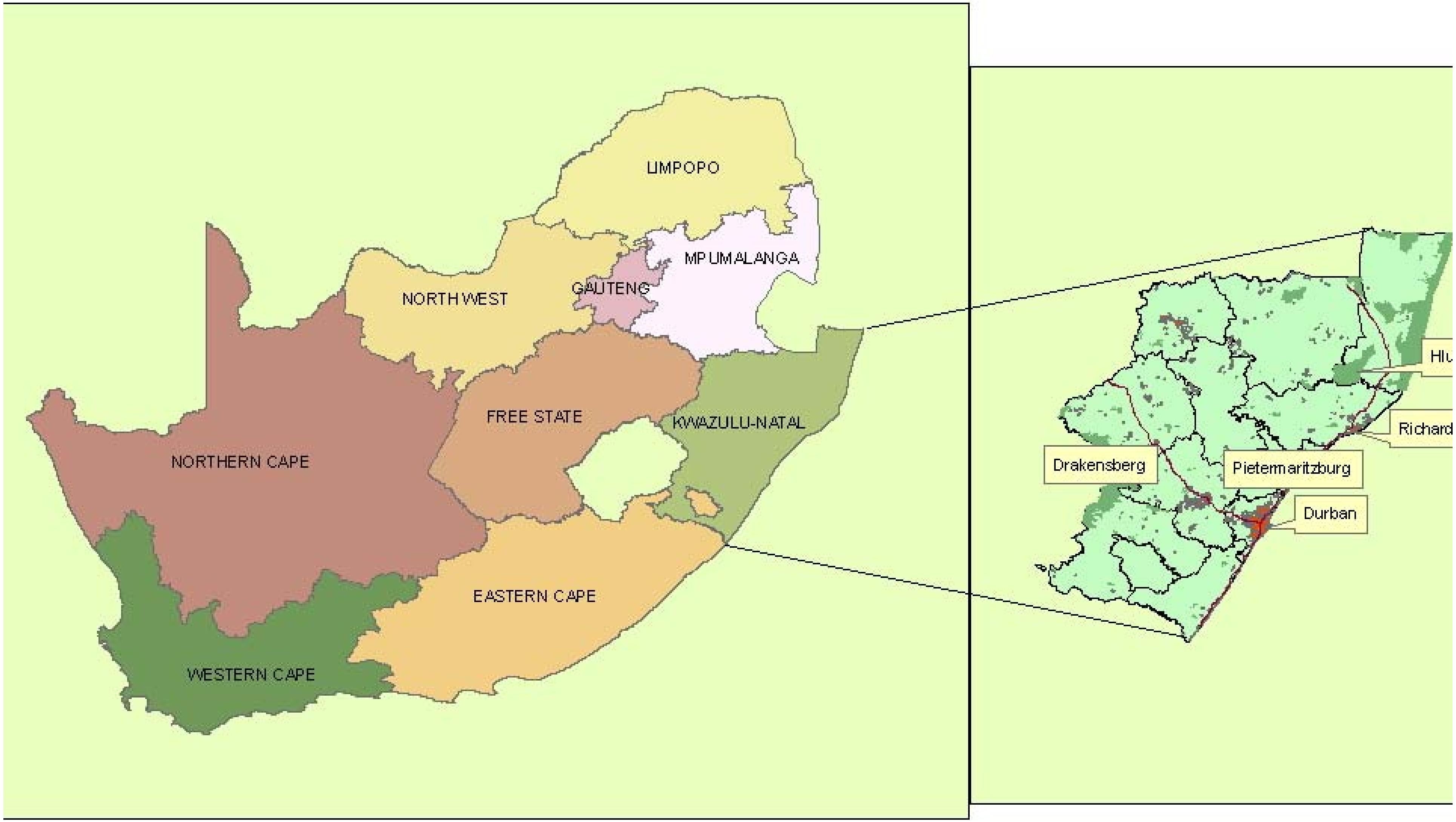
Directorate of
Veterinary Services
NAMIBIA

RABIES UPDATE

KWAZULU NATAL

JAN – DEC 2005

Dr.K.Perrett


LIMPOPO
MPUMALANGA
NORTH WEST
GAUTENG
FREE STATE
KWAZULU-NATAL
NORTHERN CAPE
EASTERN CAPE
WESTERN CAPE
Drakensberg
Pietermaritzburg
Durban

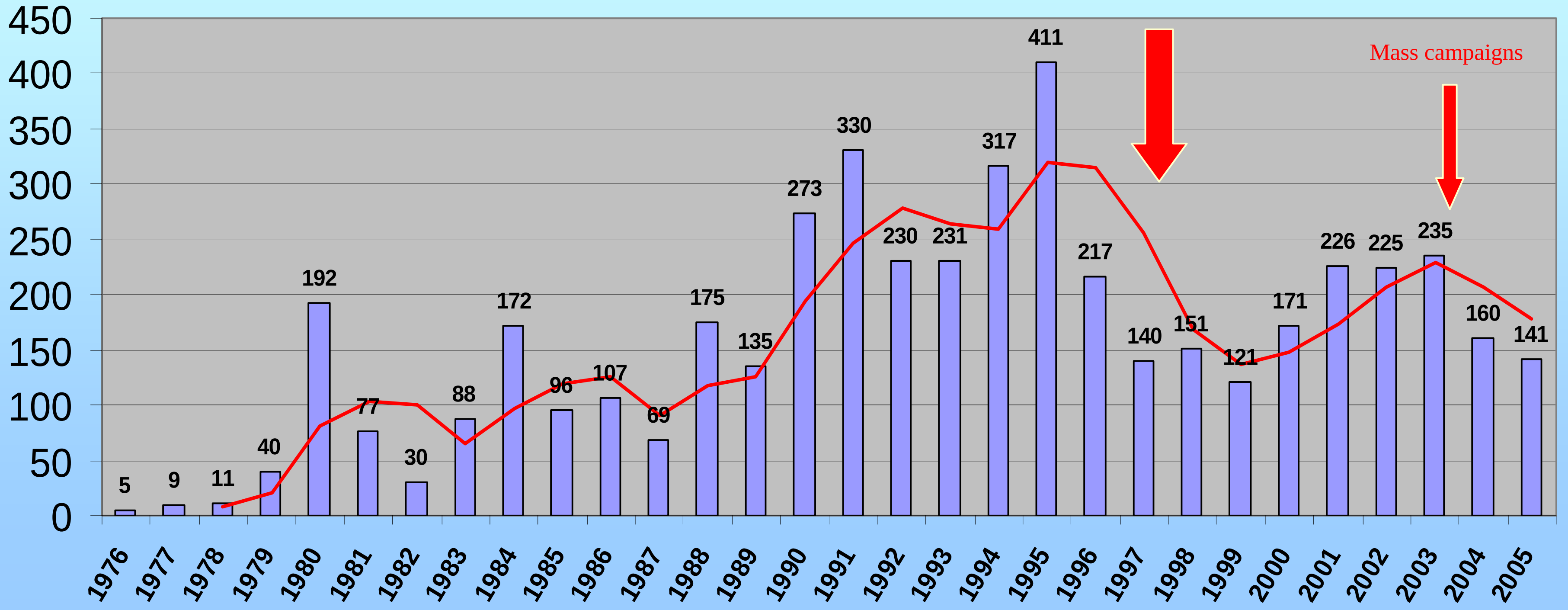
KZN STATISTICS

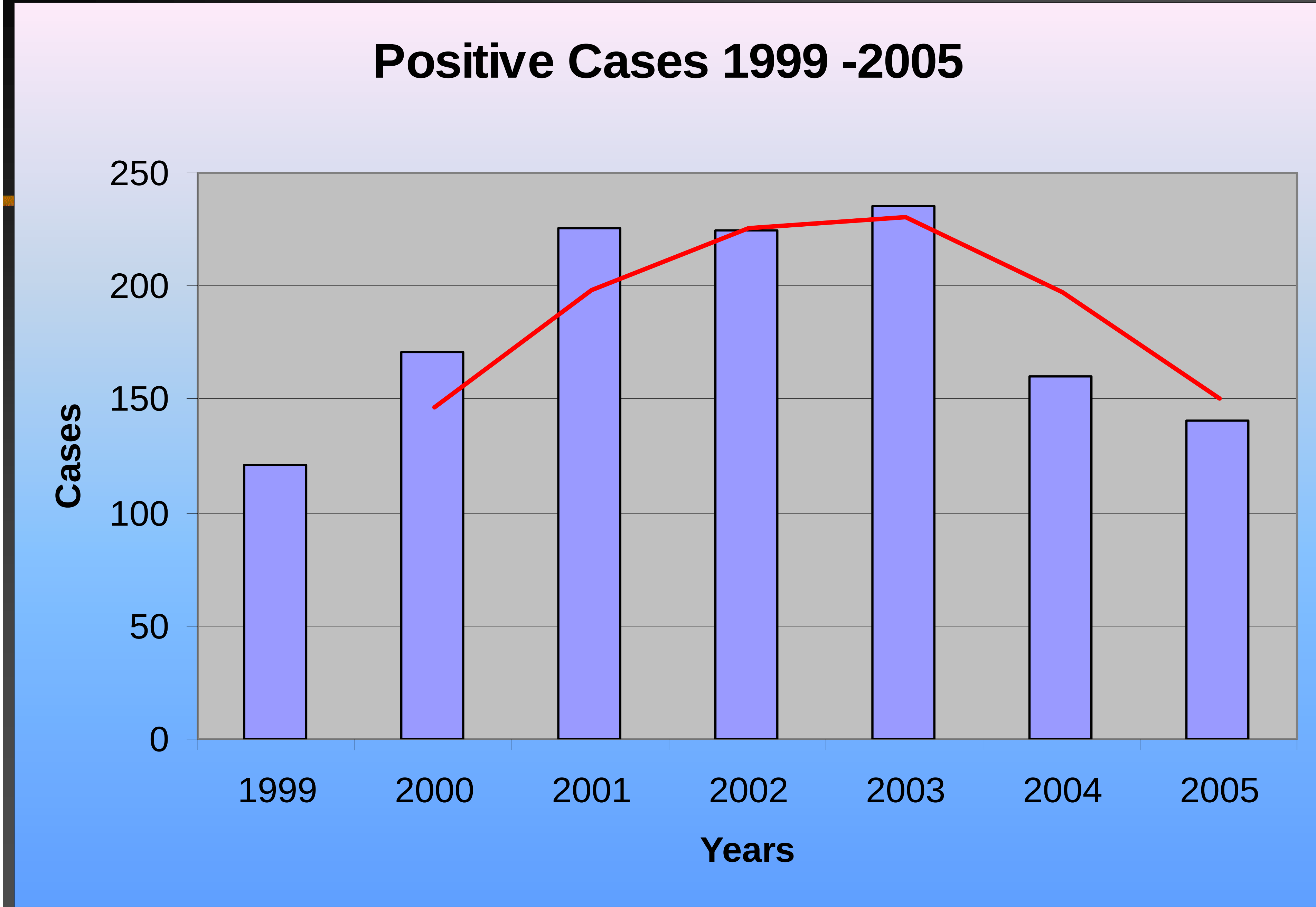
- Area: 92,100 km^2
- Population : 9,426,017
- Population growth: 12.0% (2001 census)
  - (Negative growth rate 2004 ??)
- Highest HIV/AIDS infection in RSA

RABIES UPDATE

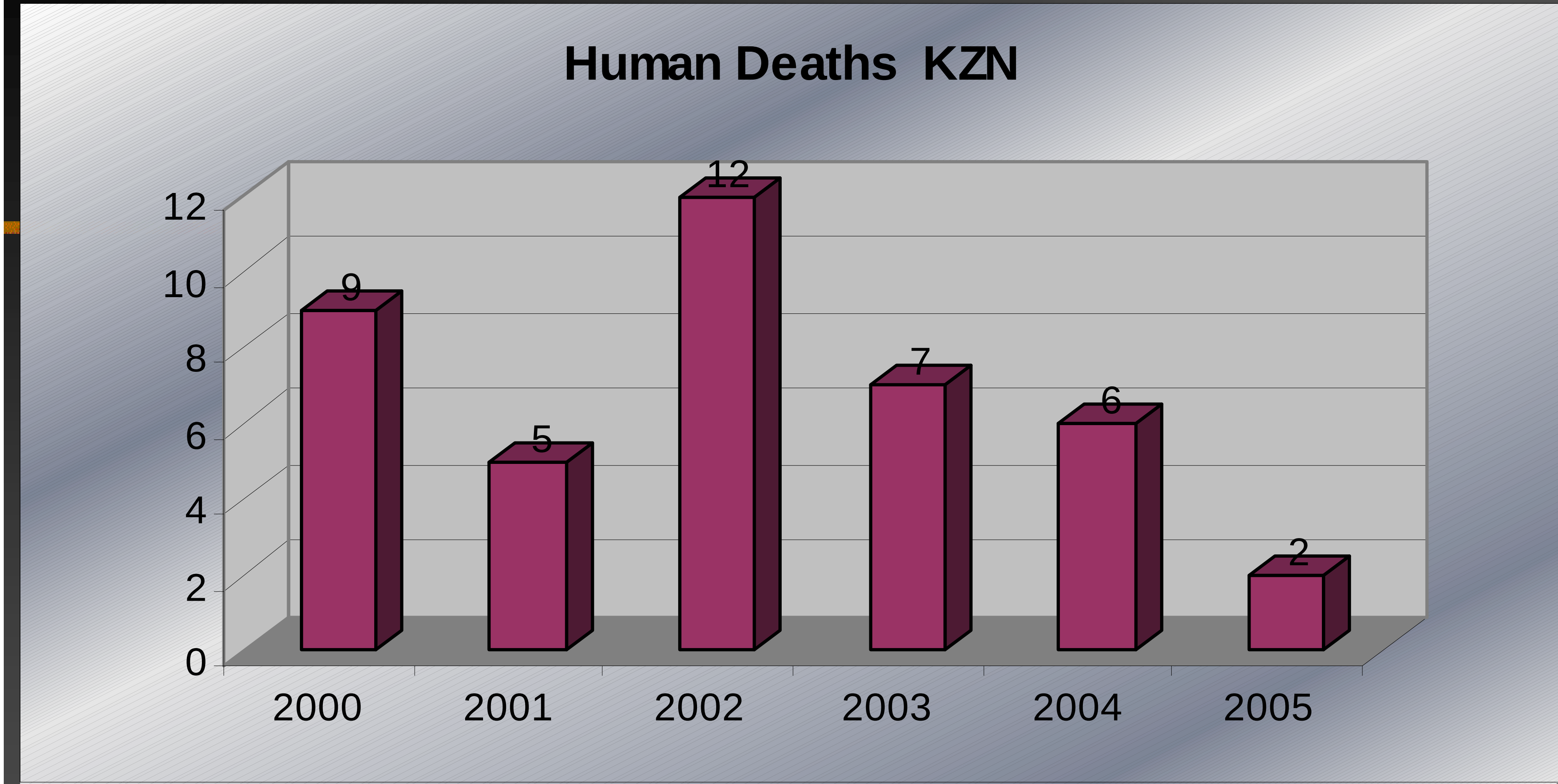
- Historical data
- Monthly/ quarterly data for last 3 years
- Species composition of positive cases
- Spatial distribution of cases
- Laboratory Submissions
- Rabies vaccinations

Positive Rabies Cases

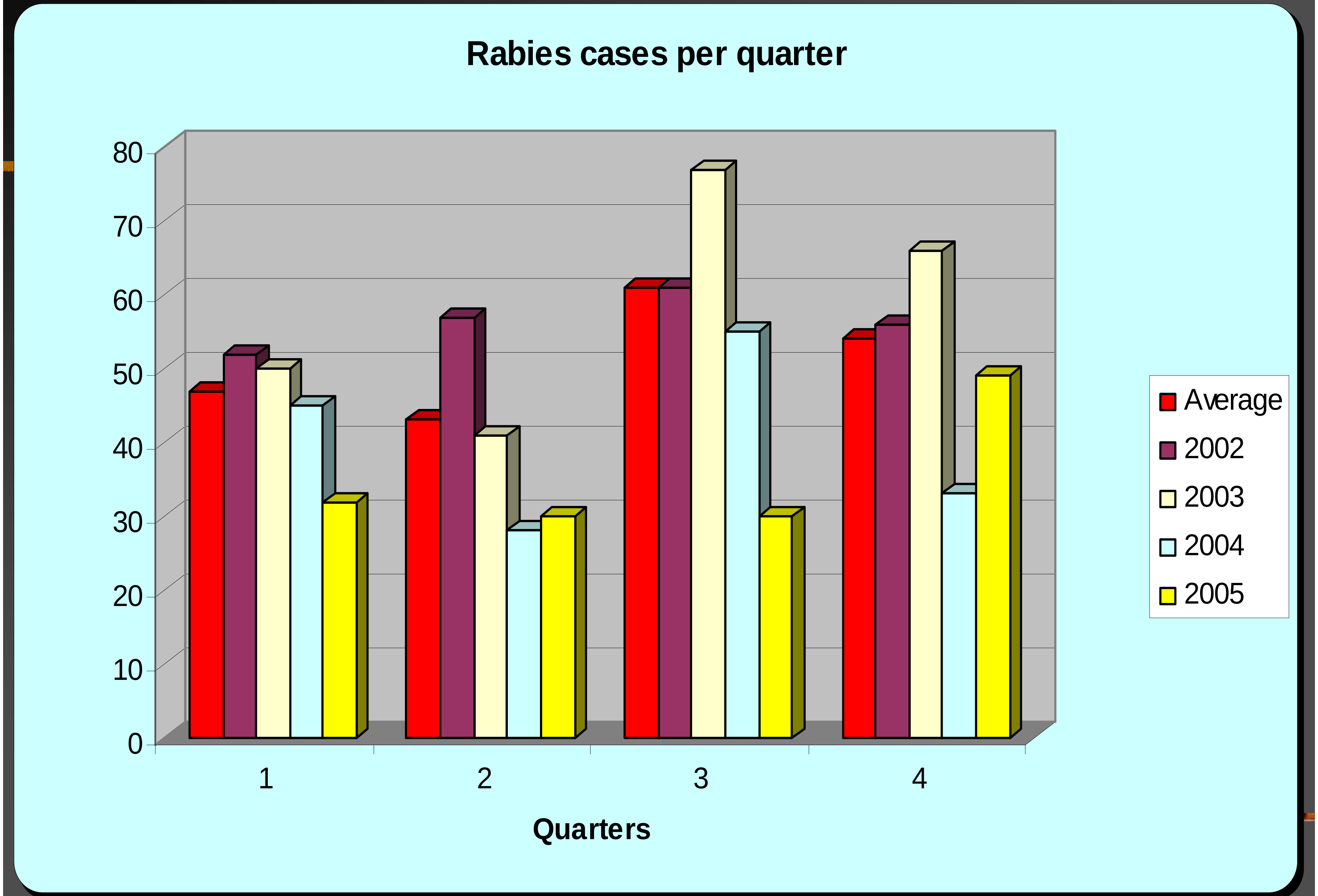


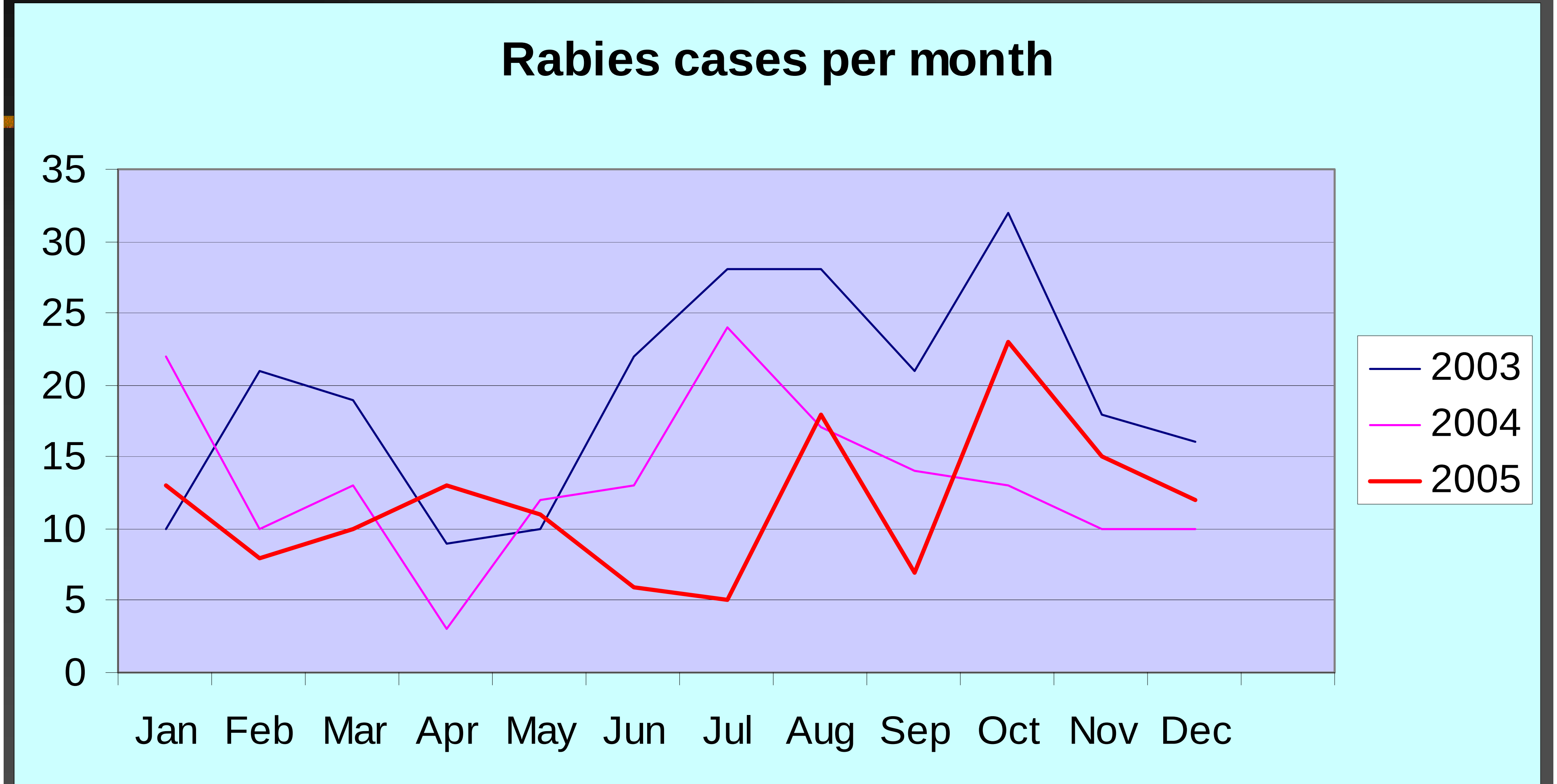


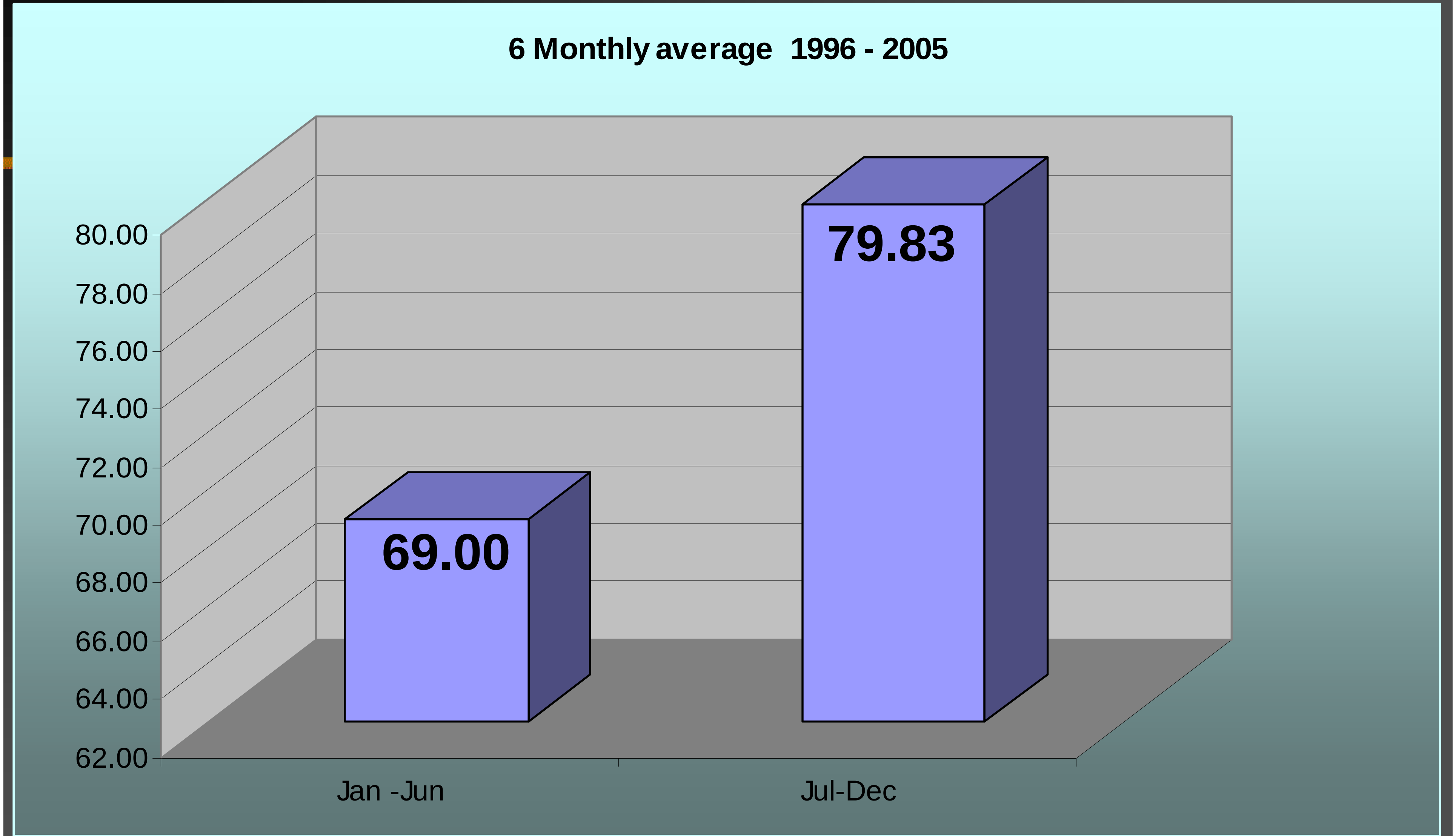
Positive Cases 1999 -2005
Cases
250
200
150
100
50
0
1999
2000
2001
2002
2003
2004
2005
Years



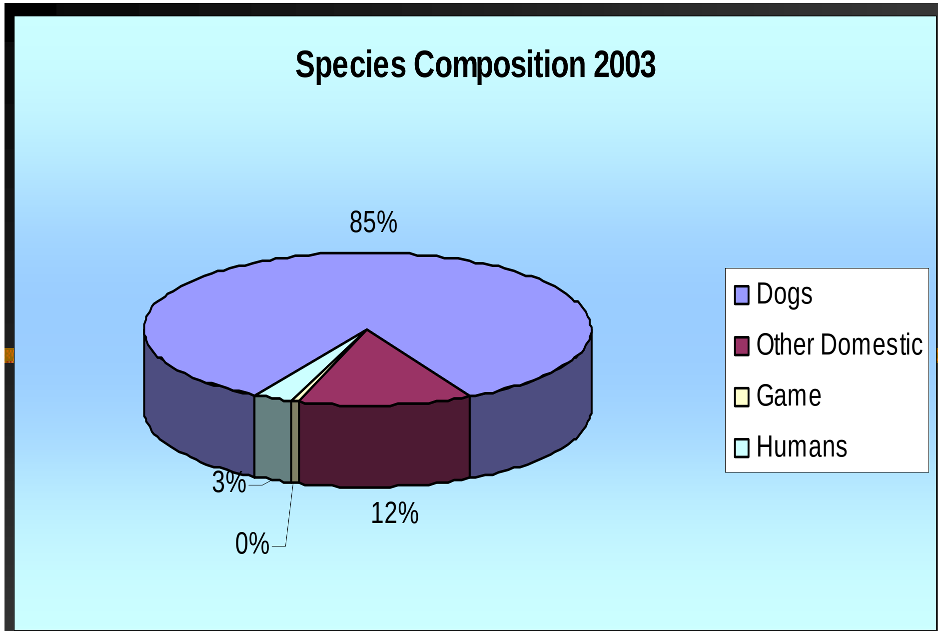
FIRST CONFIRMED DEATH
IN 2005 OCCURRED
IN NOVEMBER

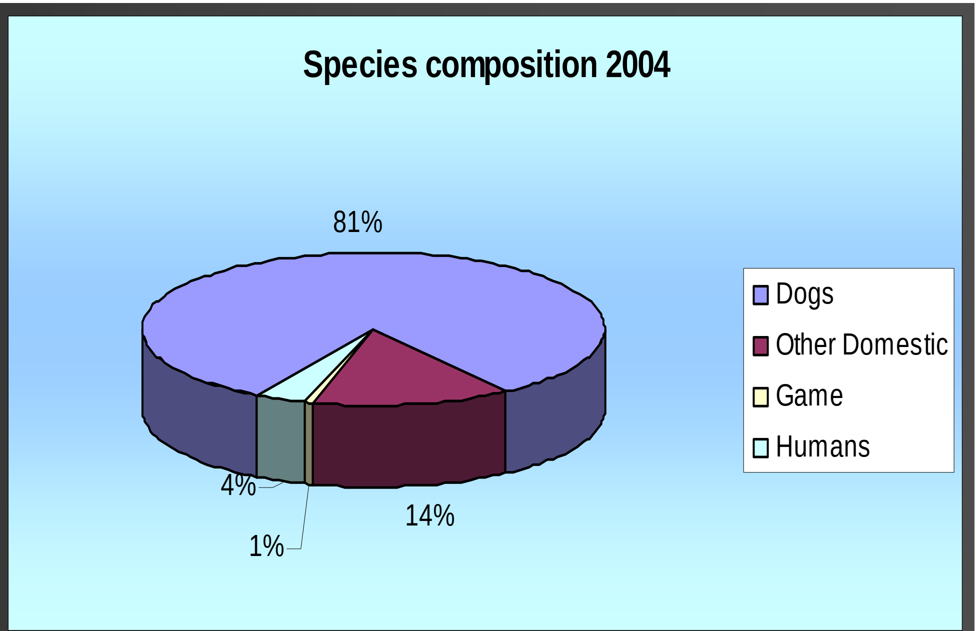

Rabies cases per quarter
0
10
20
30
40
50
60
70
80
1
2
3
4
Quarters
Average
2002
2003
2004
2005

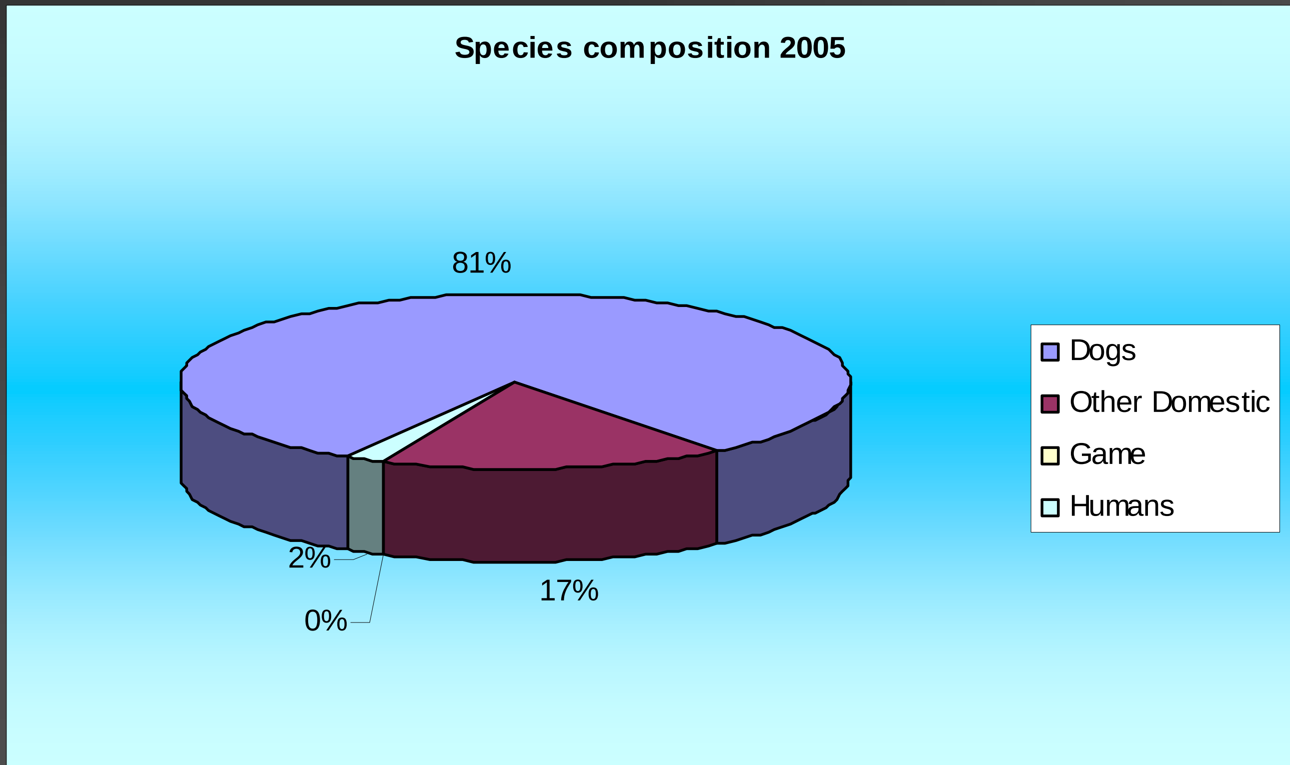

Rabies cases per month
35
30
25
20
15
10
5
0
Jan
Feb
Mar
Apr
May
Jun
Jul
Aug
Sep
Oct
Nov
Dec
2003
2004
2005



6 Monthly average 1996 - 2005
80.00
78.00
76.00
74.00
72.00
70.00
68.00
66.00
64.00
62.00
69.00
79.83
Jan -Jun
Jul-Dec


Species Composition 2003
85%
3%
0%
12%
Dogs
Other Domestic
Game
Humans


Species composition 2004
81%
4%
1%
14%
Dogs
Other Domestic
Game
Humans

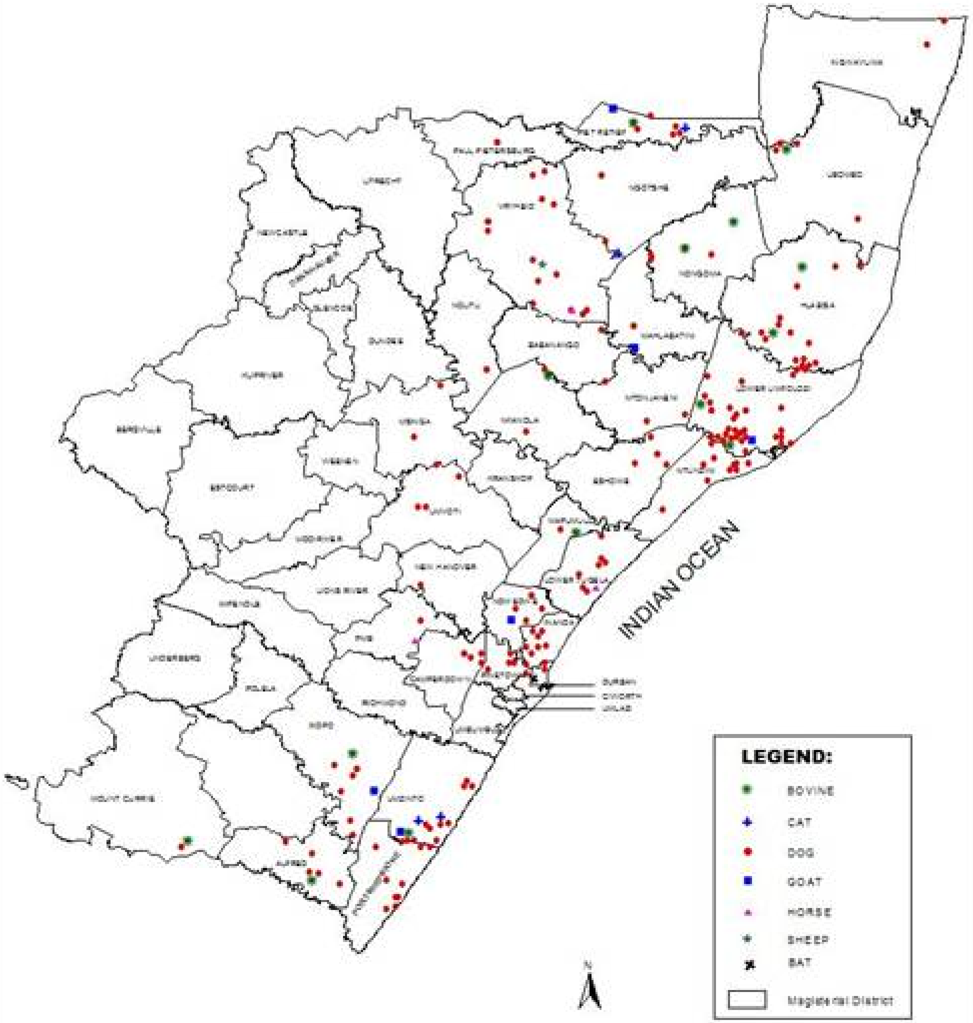

Species composition 2005
81%
2%
0%
17%
Dogs
Other Domestic
Game
Humans

KWAZULU - NATAL RABIES CASES

JANUARY - DECEMBER, 2003

234 cases

(203 dogs, 16 bovine, 6 goats, 4 cats, 3 horses, 1 bat and 1 sheep)

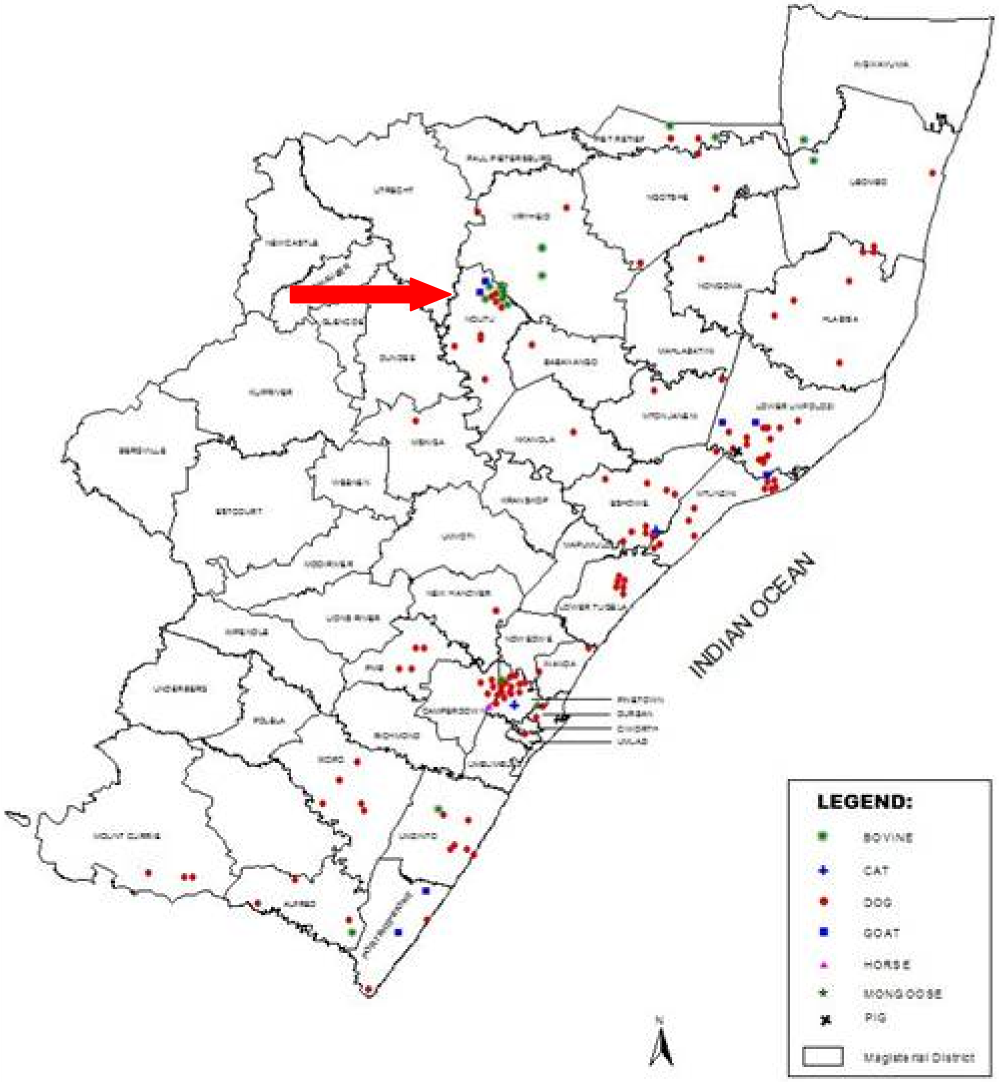


KWAZULU - NATAL RABIES CASES

JANUARY - DECEMBER, 2004

160 cases

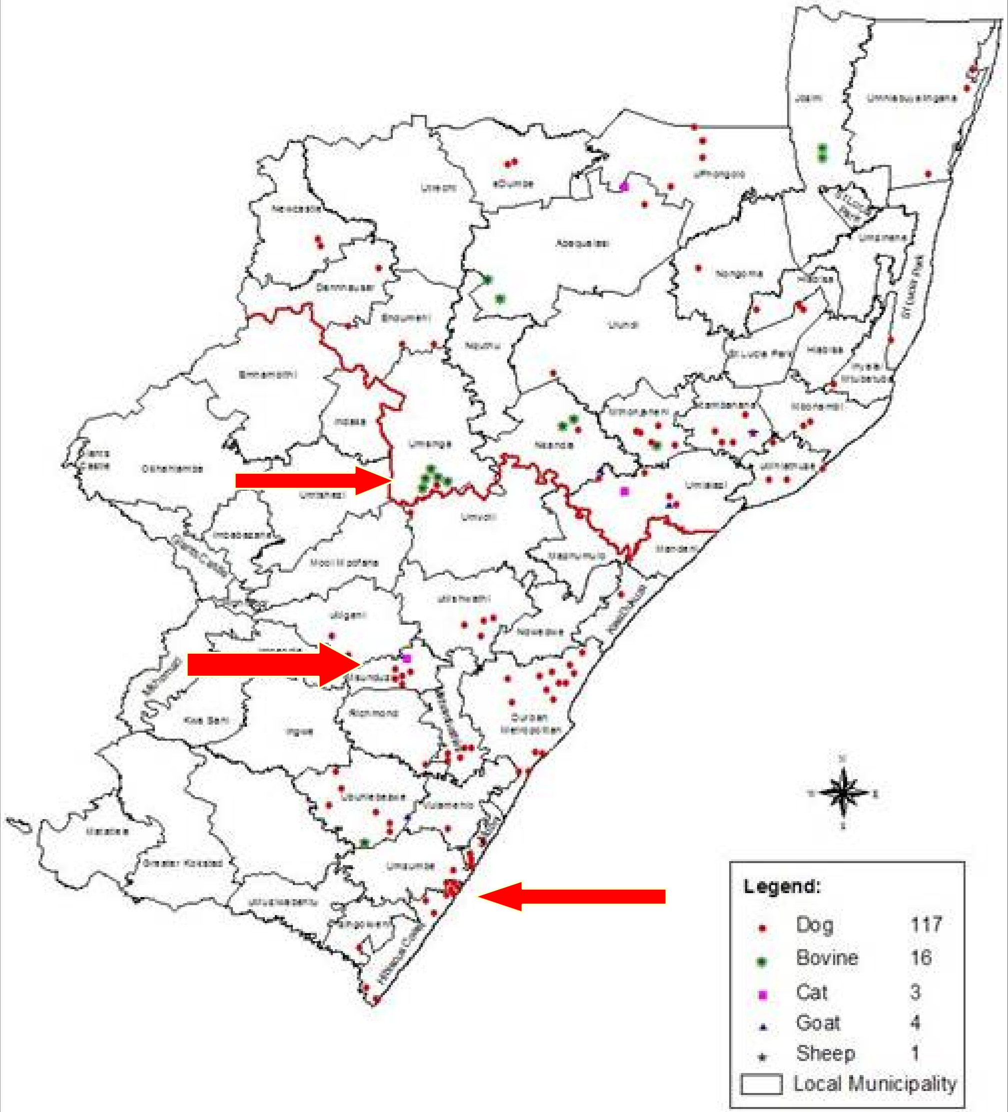
(132 dogs, 16 bovines, 2 cats, 7 goats , 1 horse, 1 mongoose and 1 pig)

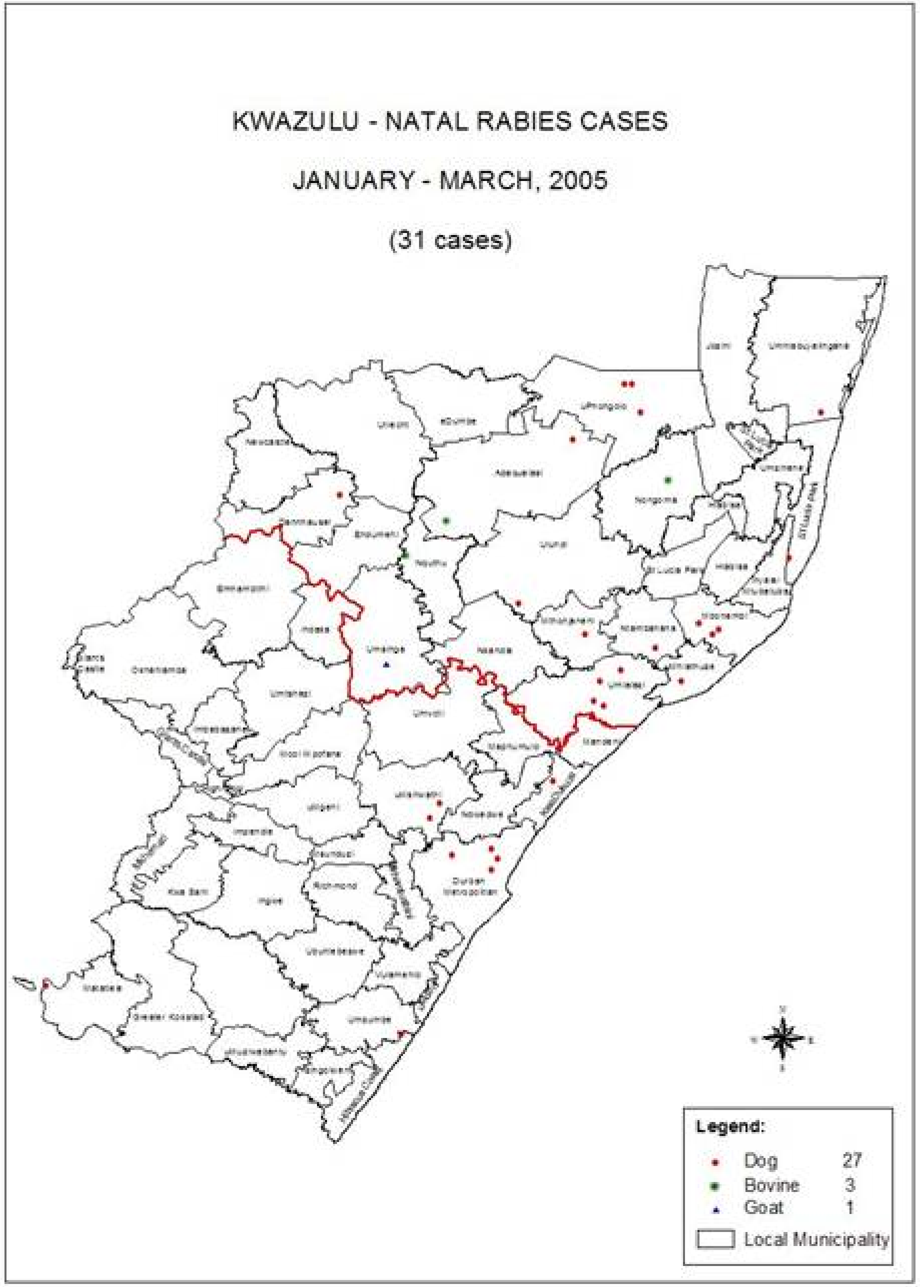


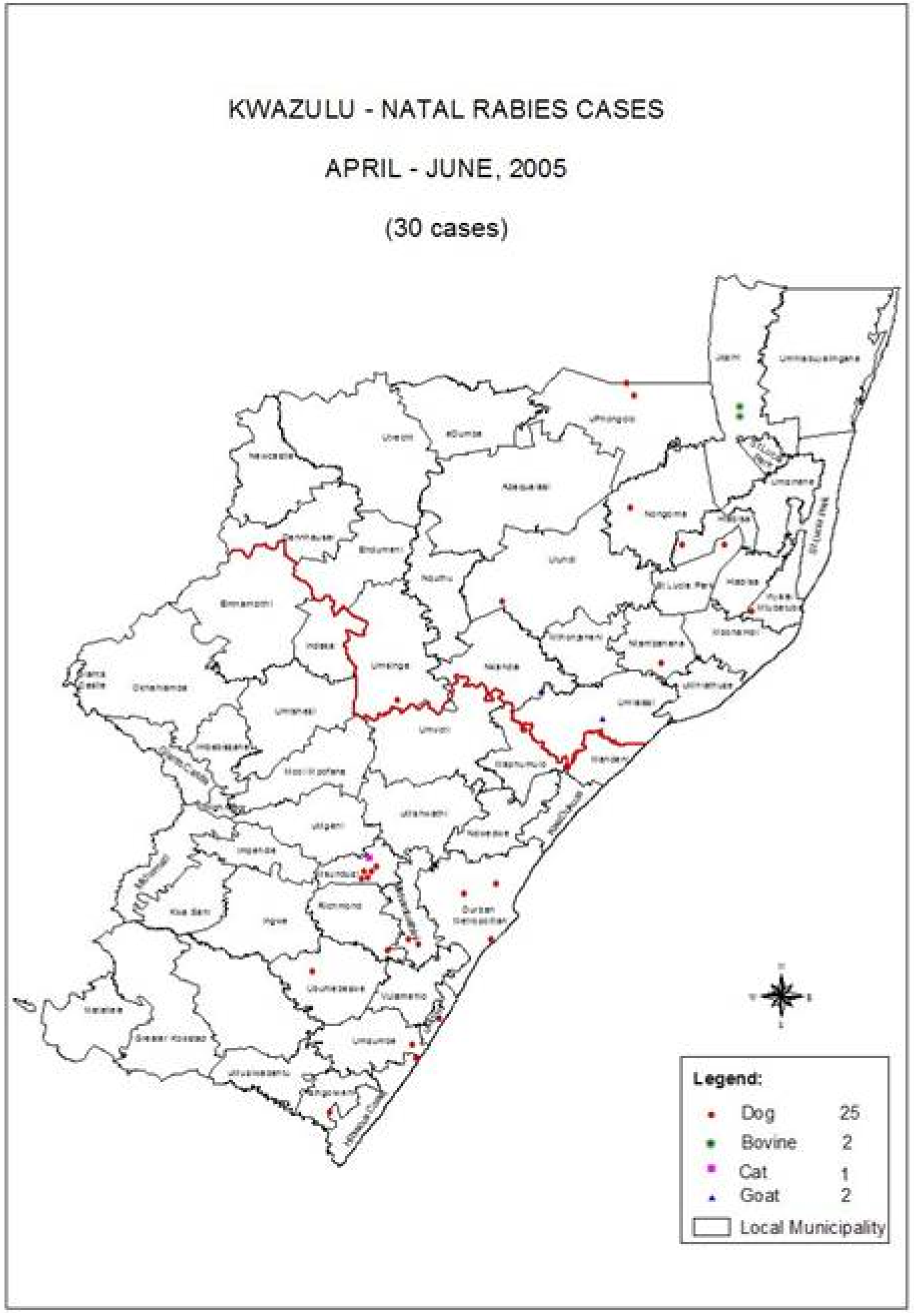
KWAZULU - NATAL RABIES CASES

JANUARY - DECEMBER, 2005

(141 cases)



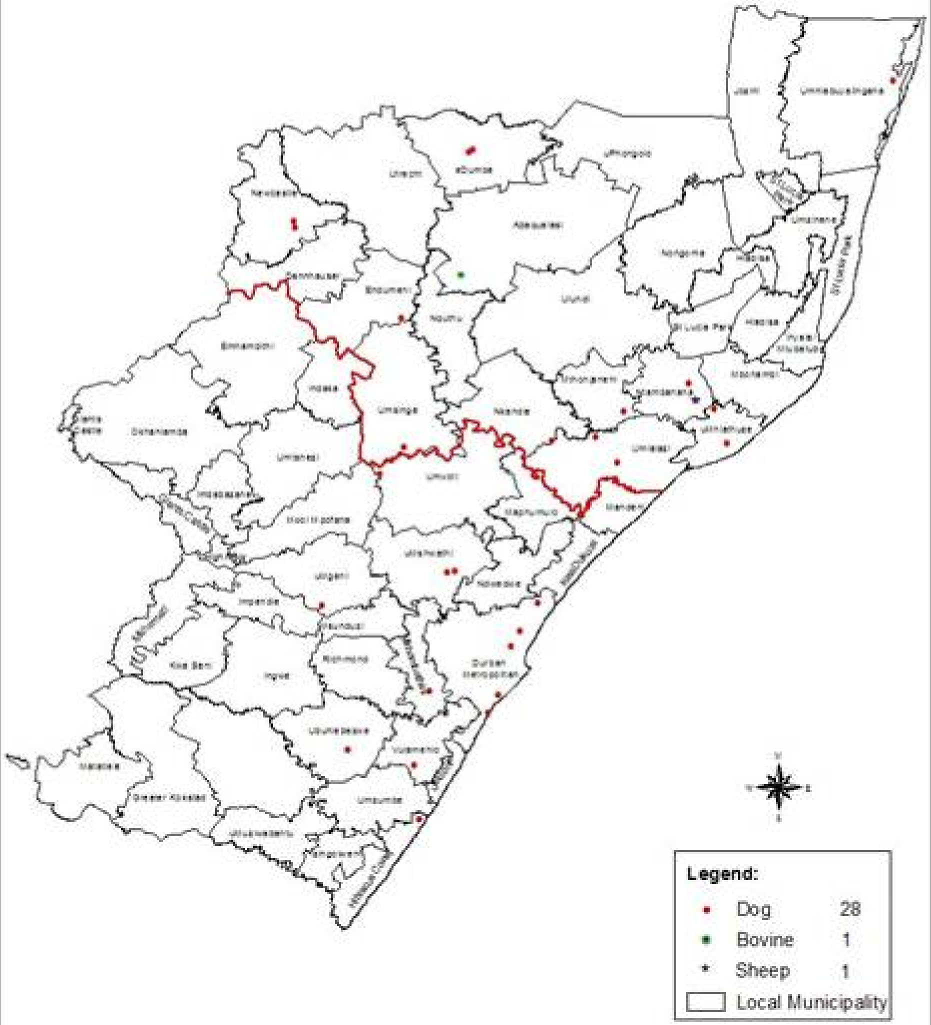

KWAZULU - NATAL RABIES CASES
JANUARY - MARCH, 2005
(31 cases)
Legend:
Dog 27
Bovine 3
Goat 1
Local Municipality


KWAZULU - NATAL RABIES CASES
APRIL - JUNE, 2005
(30 cases)
Legend:
Dog 25
Bovine 2
Cat 1
Goat 2
Local Municipality

KWAZULU - NATAL RABIES CASES

JULY - SEPTEMBER, 2005

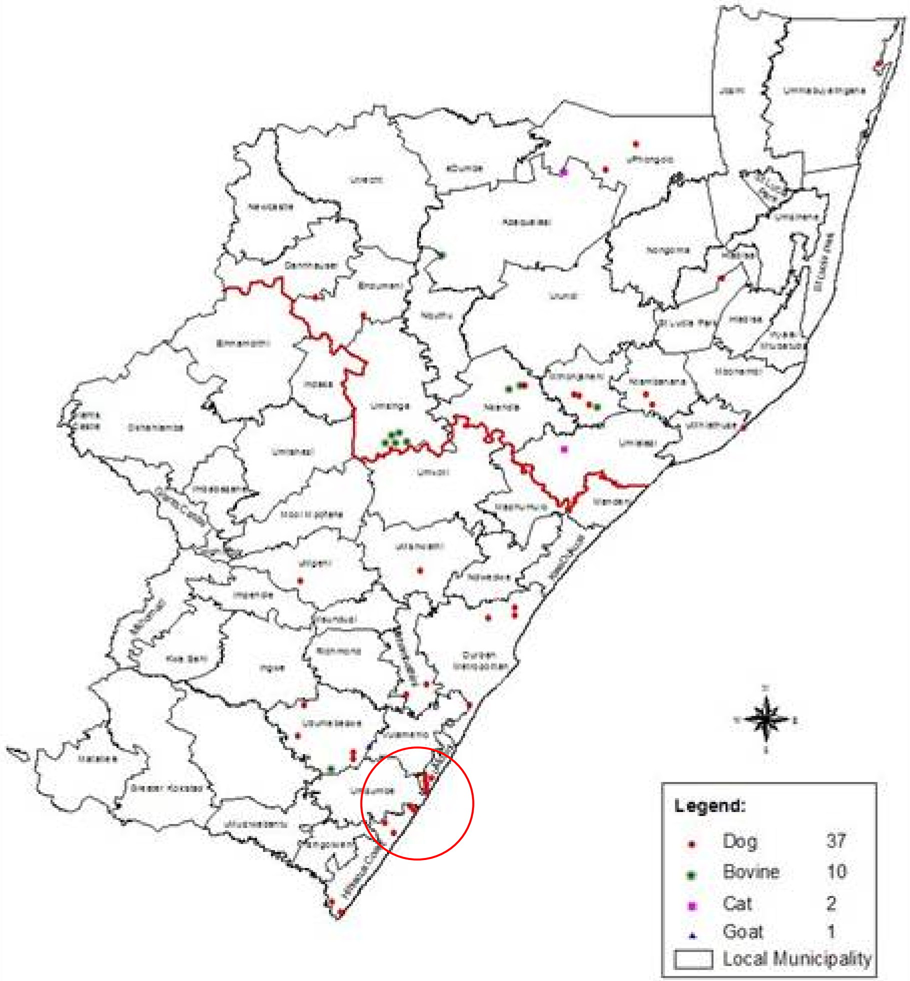
(30 cases)



KWAZULU - NATAL RABIES CASES

OCTOBER - DECEMBER, 2005

(50 cases)

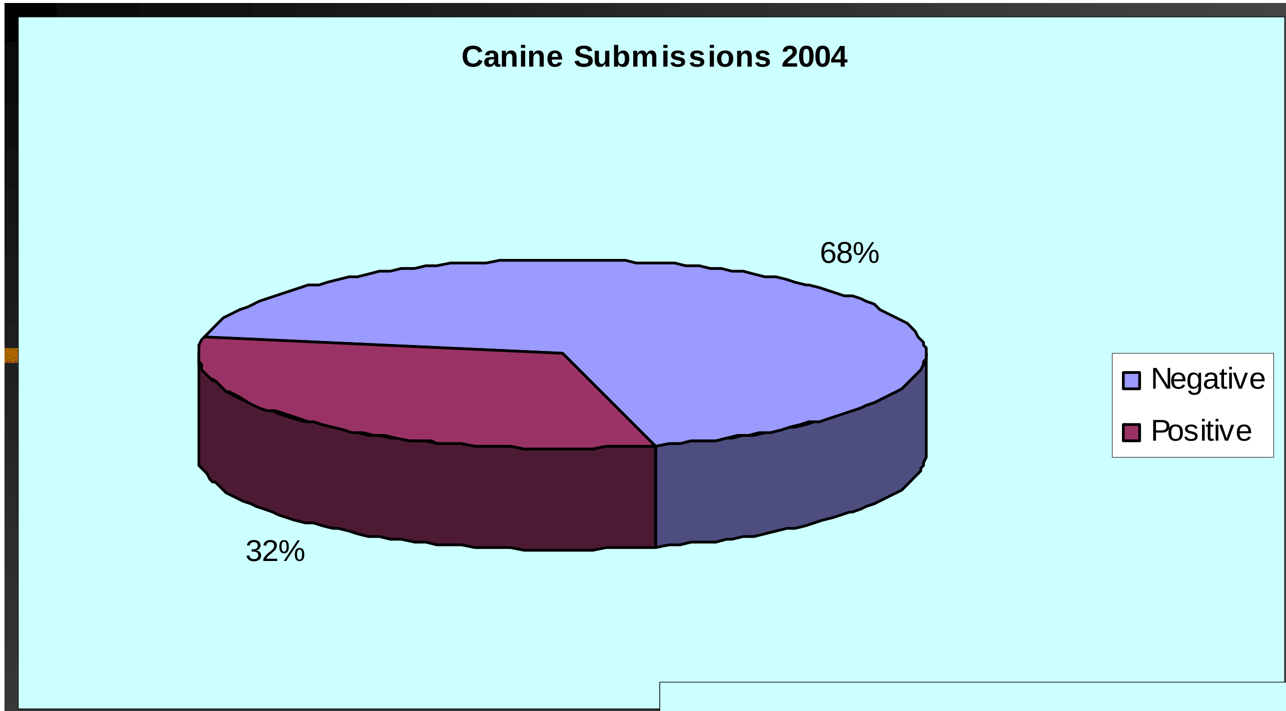


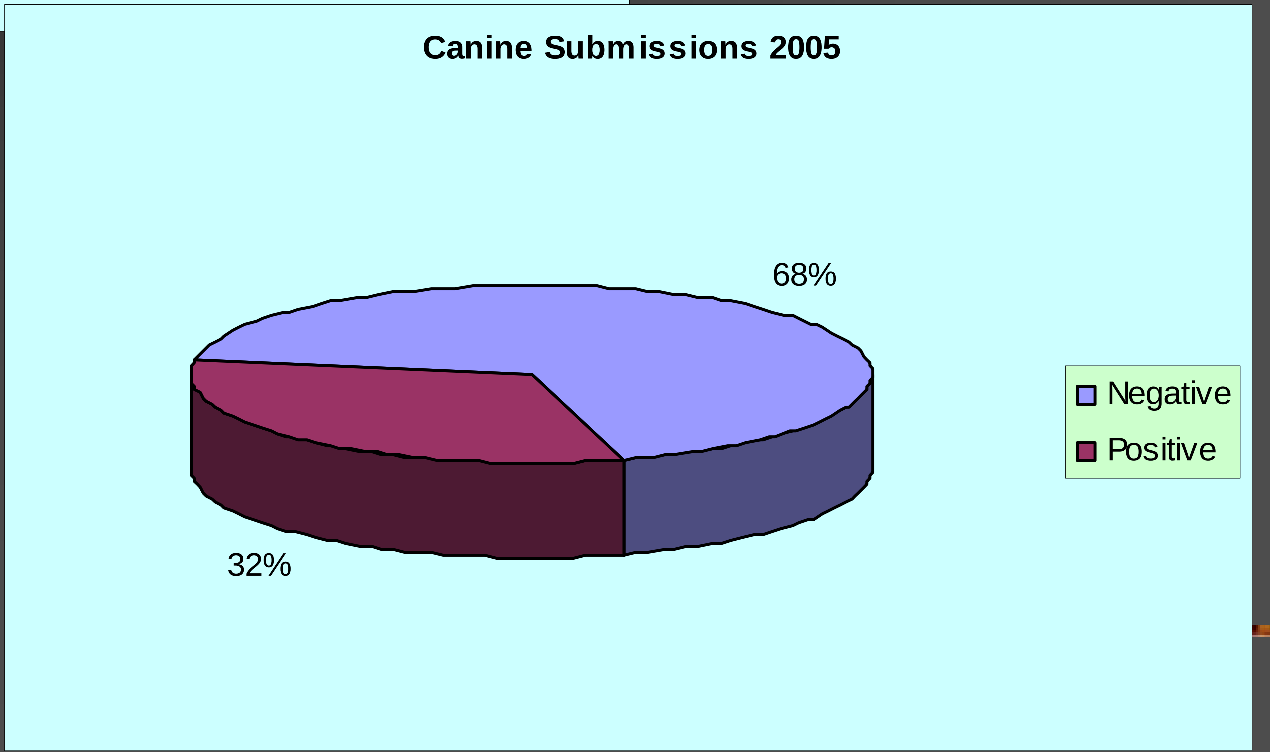
LABORATORY SUBMISSIONS 2004

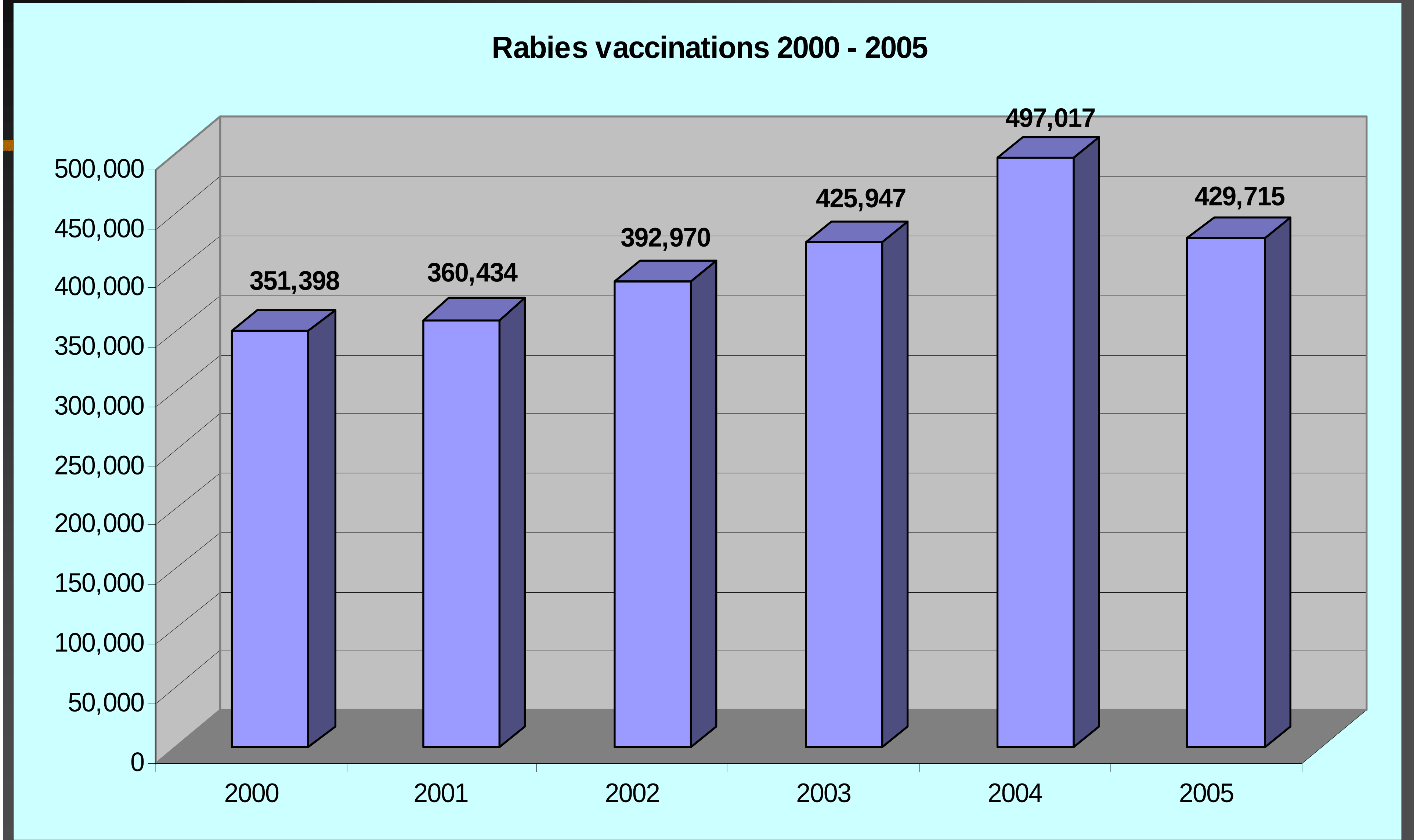
		Submissions	Negative	Positive	% Positive
KZN					
	Canine	418	285	133	31.82
	Bovine	32	16	16	50.00
	Caprine	9	2	7	77.78
	Human	6	0	6	100.00
	Feline	65	63	2	3.08
	Equine	4	3	1	25.00
	Porcine	3	2	1	33.33
	Mongoose	7	6	1	14.29
	Bat	1	0	1	100.00
	Other	36	36	0	0.00
		581	413	168	28.92

LABORATORY SUBMISSIONS 2005

		Submissions	Negative	Positive	% Positive
KZN					
	Canine	364	247	117	32.14
	Bovine	25	9	16	64.00
	Caprine	5	1	4	80.00
	Human	4	2	2	50.00
	Feline	47	44	3	6.38
	Equine	2	2	0	0.00
	Porcine	1	1	0	0.00
	Mongoose	6	6	0	0.00
	Bat	2	2	0	0.00
	Sheep	2	1	1	50.00
	Bushbaby	3	3	0	0.00
	Bushbuck	1	1	0	0.00
	Dassie	4	4	0	0.00
	Duiker	1	1	0	0.00
	Genet	2	2	0	0.00
	Giraffe	1	1	0	0.00
	Jackal	6	6	0	0.00
	Meercat	2	2	0	0.00
	Monkey	3	3	0	0.00
	Rabbit	1	1	0	0.00
	Game	2	2	0	0.00
		484	341	143	29.55


Canine Submissions 2004
68%
32%
Negative
Positive


Canine Submissions 2005
68%
32%
Negative
Positive


Rabies vaccinations 2000 - 2005
500,000
450,000
400,000
350,000
300,000
250,000
200,000
150,000
100,000
50,000
0
351,398
360,434
392,970
425,947
497,017
429,715
2000
2001
2002
2003
2004
2005

South Africa country report 2004-2005

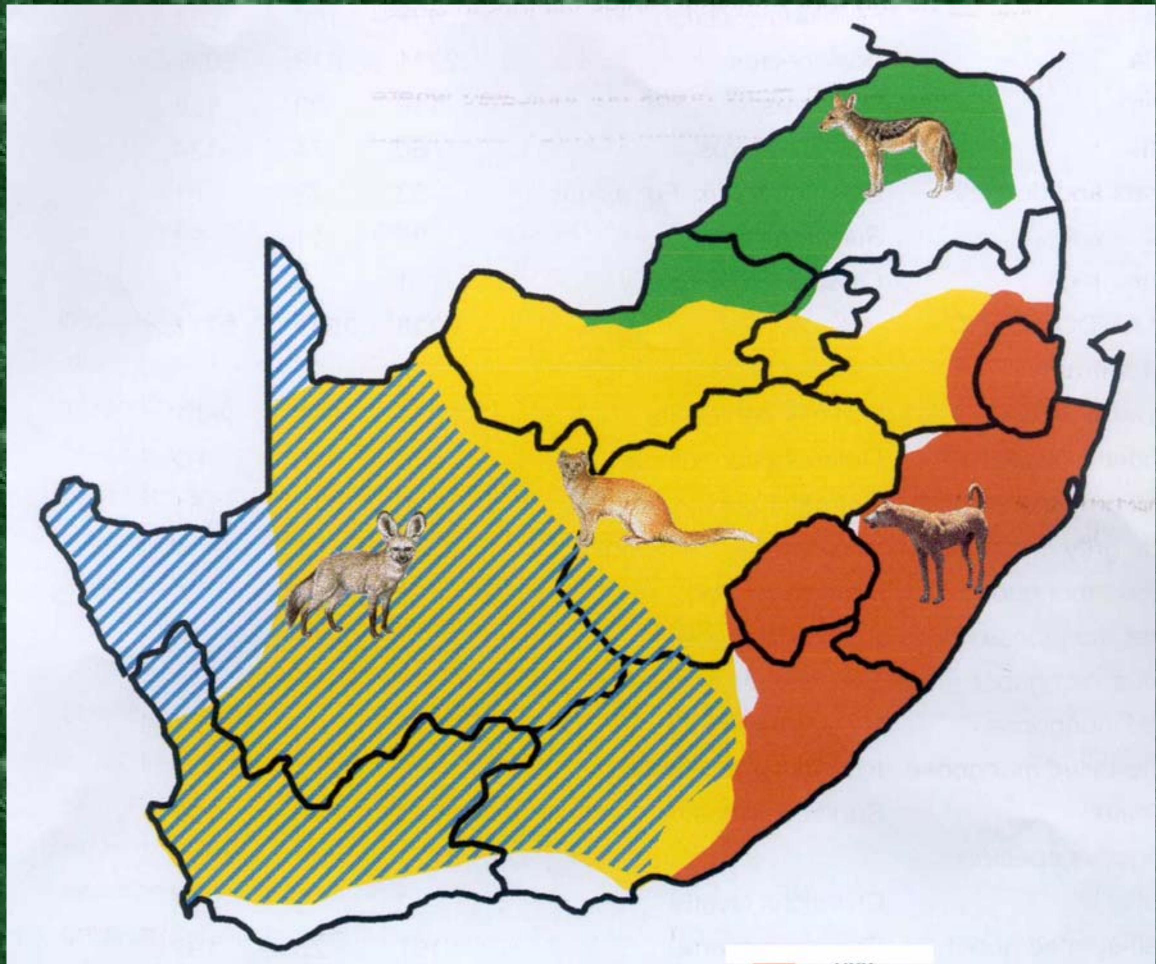
OIE Reference laboratory
Onderstepoort Veterinary Institute
SabetaC@arc.agric.za

ARC • LNR

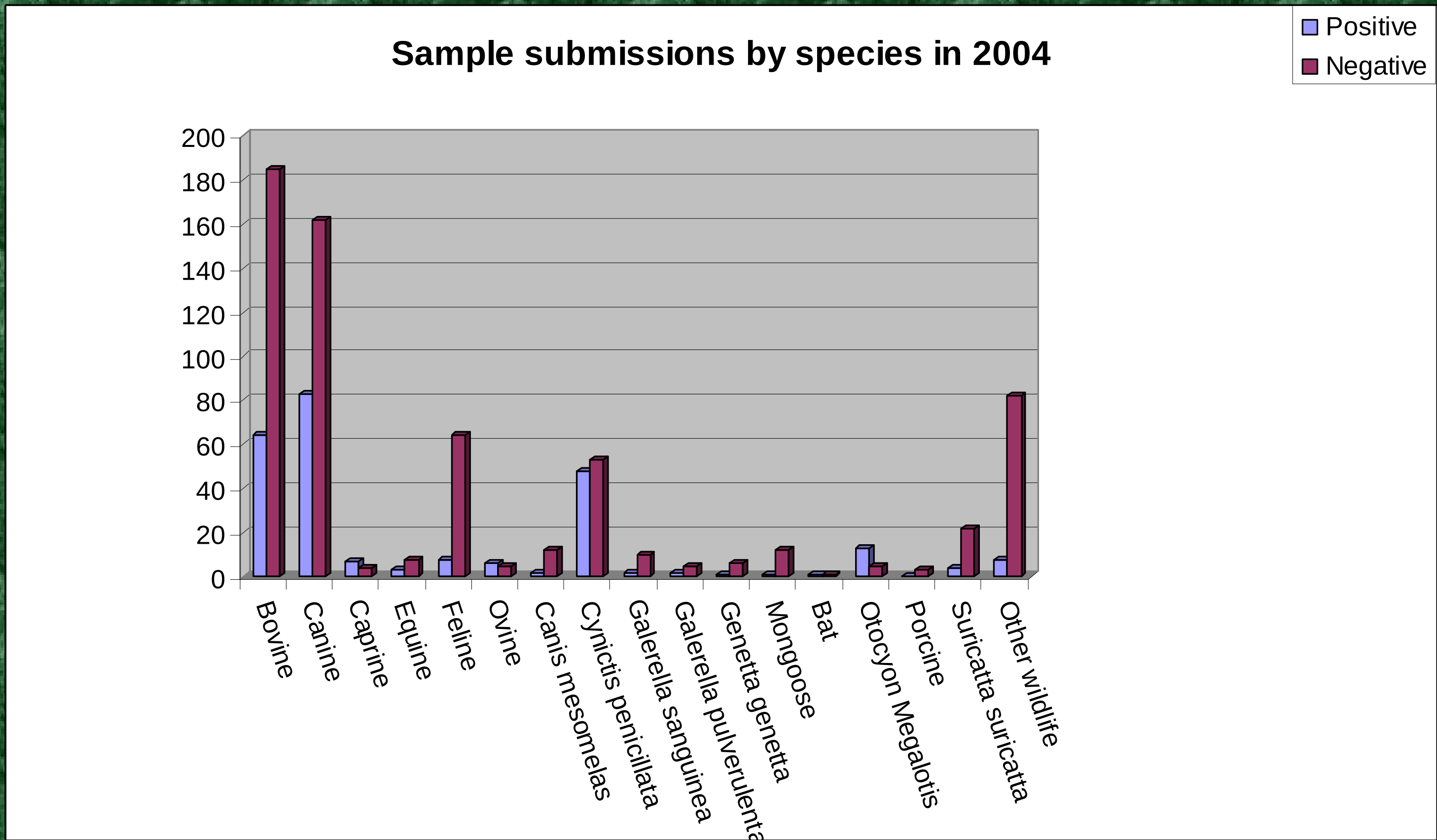
Rabies diagnosis - methods used.

- Fluorescent antibody test (FAT)
- Virus isolation in suckling mice (MIT)
- Virus isolation in neuroblastoma cell cultures (RTCIT)
- Immunohistochemical test (sub-contract to UP)
- Fluorescent antibody virus neutralisation test (FAVNT) – export of pets

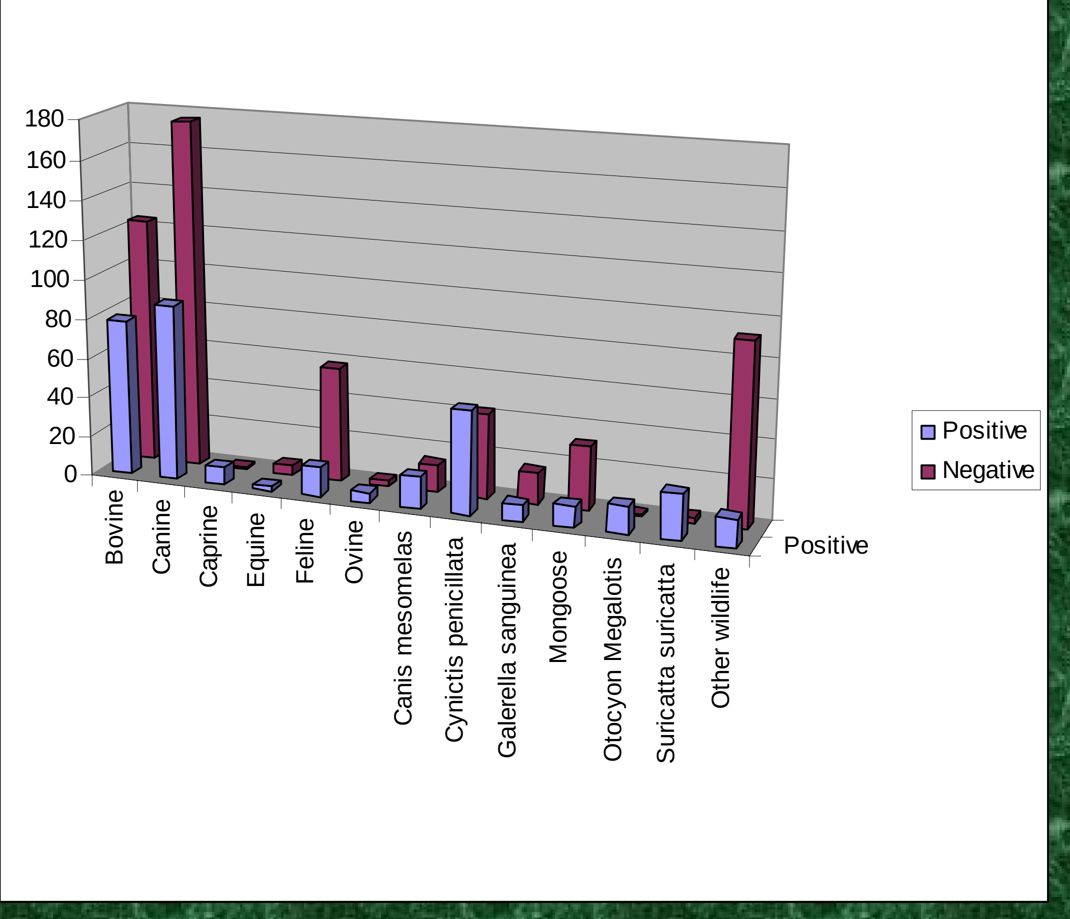
Maintenance host species, South Africa



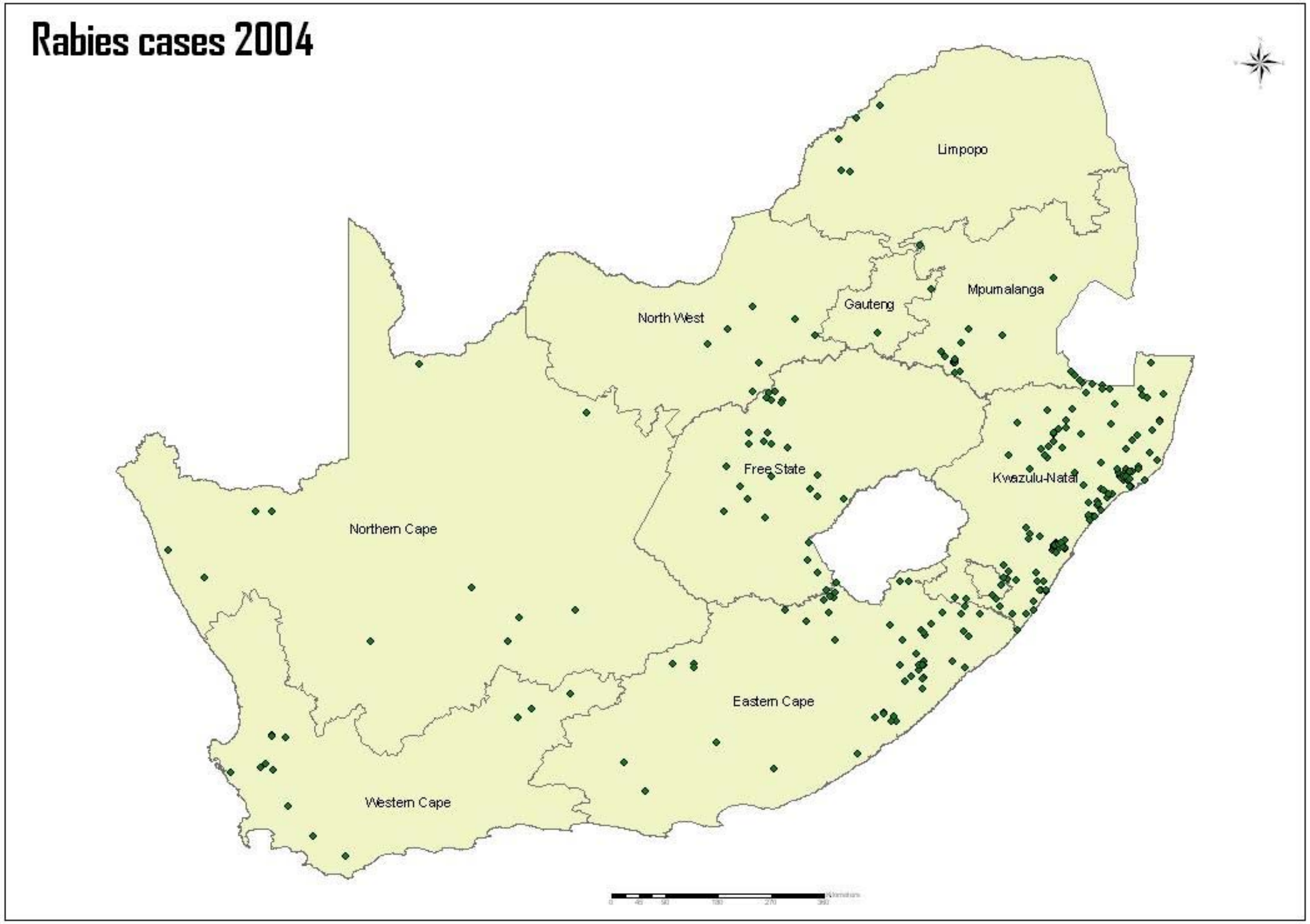
Rabies submissions: 2004



Sample submission 2005



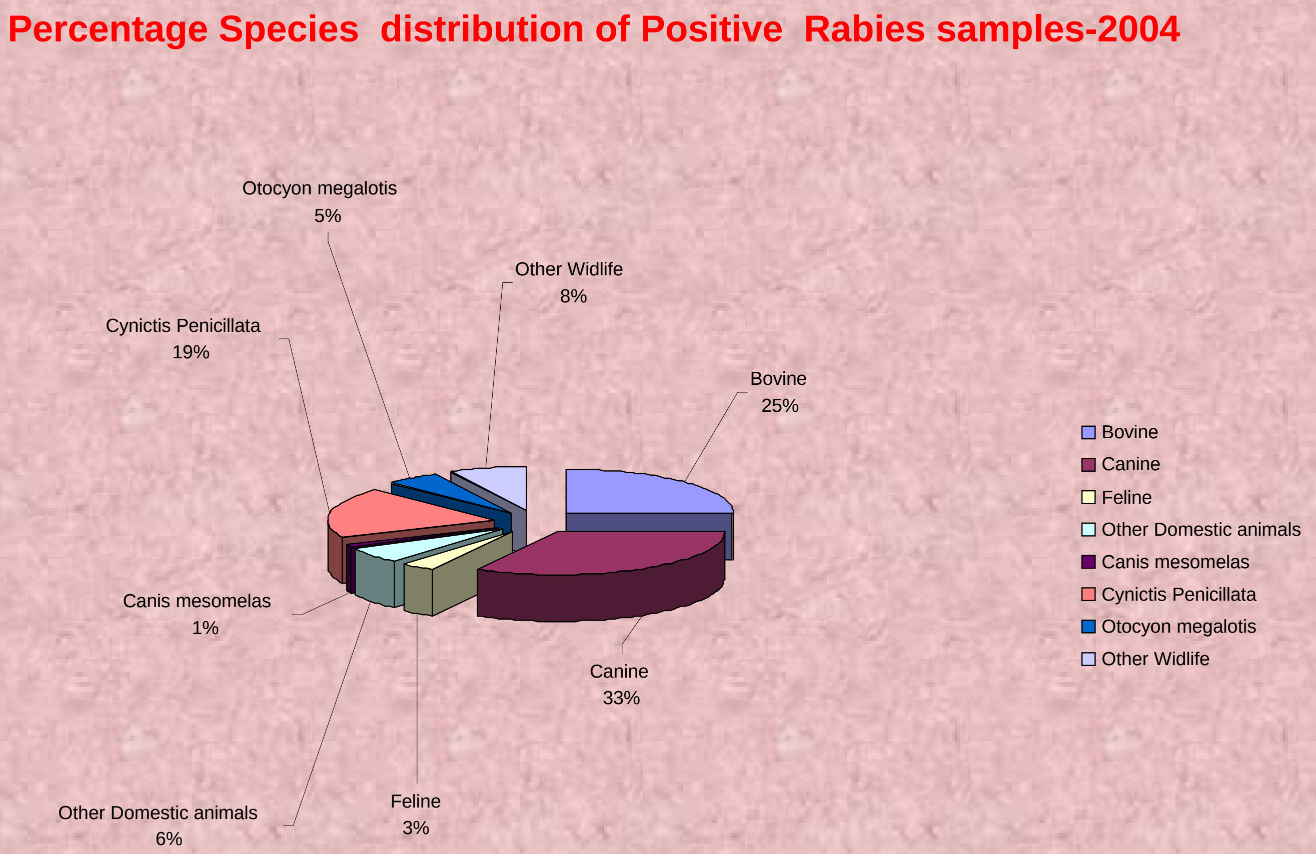
Summary rabies cases 2004



Trends 2004 - 2005

- No evidence of decline in the number of rabies cases
  - Failure to control rabies in wildlife animals
  - Low vaccination coverage
  - Canine and bovine (domestic) ; Cynictis penicillata and O. megalotis (wildlife)

Typing of positive rabies cases



Human Rabies

- Human rabies cases are handled by the National Institute of Communicable Diseases

- 10 (2004) and 9 (2005)

- Types of exposure: bites/scratches, touching/feeding animal, licking of broken skin

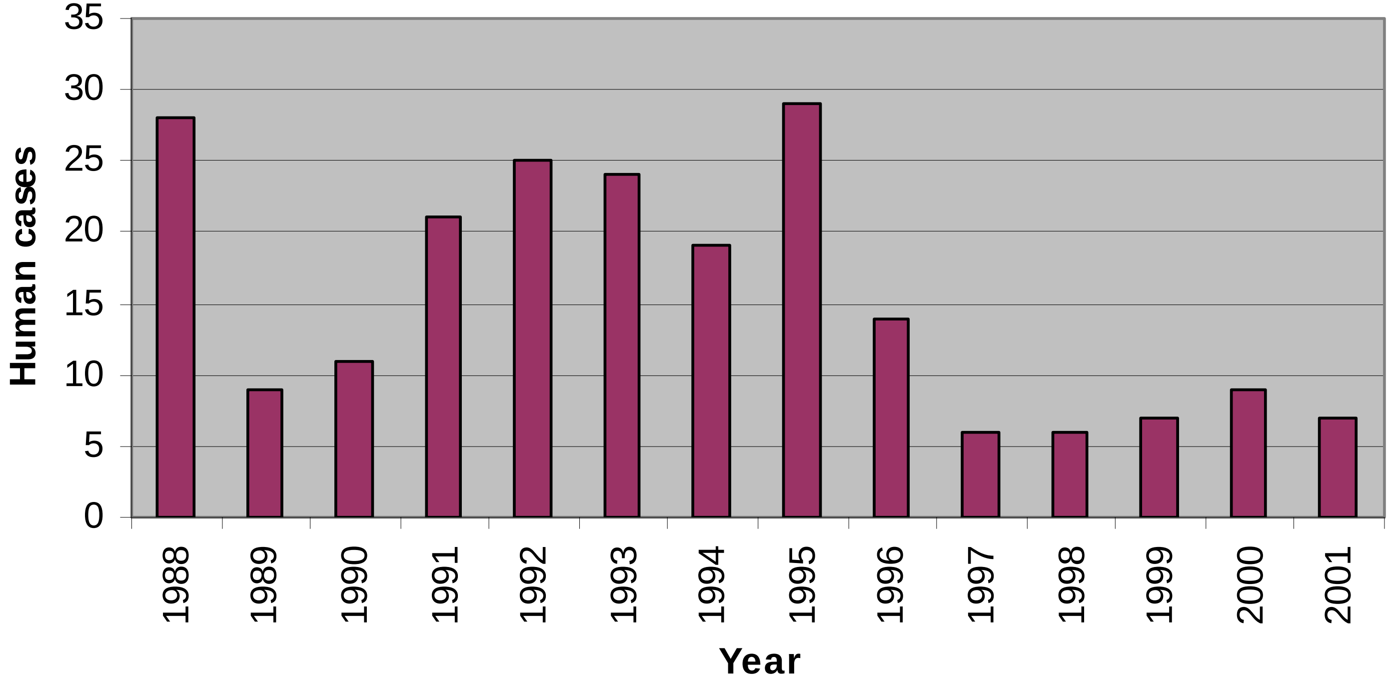
Human rabies sources 2004

Species	Total bites from suspect cases	Total bites from confirmed cases
Domestic dogs	82	33
Feline	34	7
Jackal *C. mesomelas*	6	3
Cyncitis penicillata	3	3
O. megalotis	3	3
Felis lybica	3	2
Other wildlife	36	0
TOTAL	157	51

Human rabies sources

Species	Total bites from suspect cases	Bites from confirmed cases
Domestic dog	80	20
Domestic cat	20	9
Jackal C. mesomelas	2	1
Cynictis penicillata	6	2
Other Wildlife	9	0
TOTAL	117	32

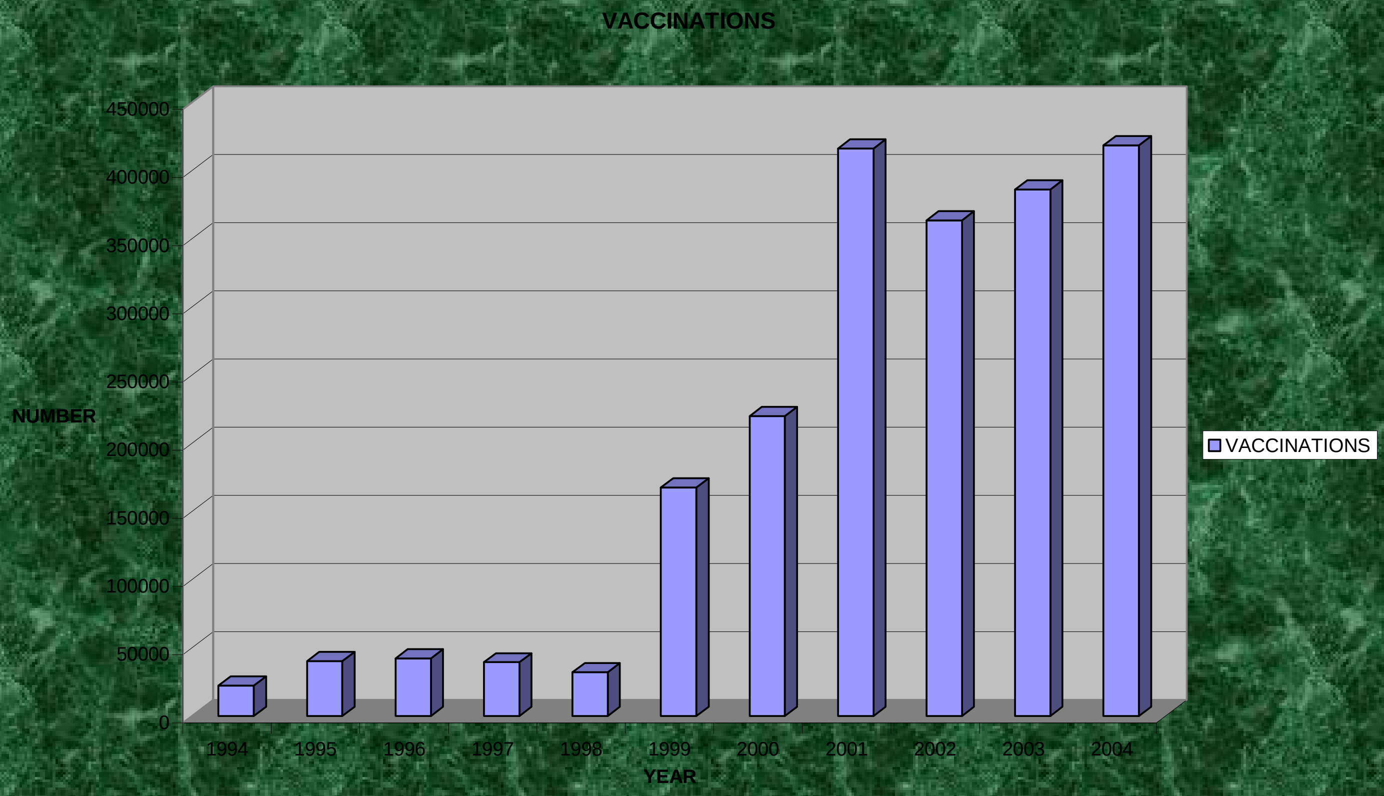
Laboratory confirmed human rabies



Vaccine use in the control of rabies

- Prophylactic human vaccines are ordered by the Ministry of Health
- Number of persons who received post-exposure treatment not known

Vaccinations 1994-2004



2005 770 000 – 800 000 for animal use

Dog population densities

- 2003 : 4 659 236, 2004: 4 588 404

  "I must stress that this information is supplied to us by the Provinces, and I can give no guarantee on the correctness of it. We usually consult with the Directorate of Agricultural Statistics on the correctness of the figures for production animals, but for dogs and cats these are the only figures I know of".

- Survey conducted in KwaZulu/Natal (Dr Bagnall's presentation)

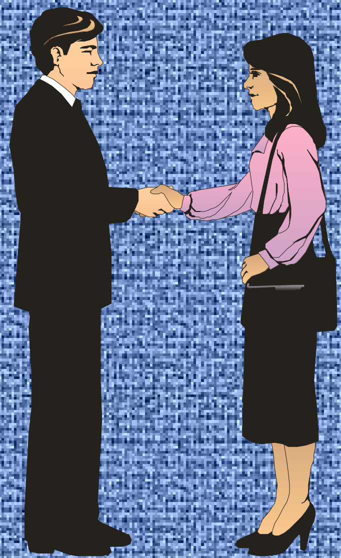
Conclusions

- Unnecessary use of post-exposure prophylaxis
- Dog densities need to be assessed more accurately.

Conclusions

- The principal lyssavirus responsible for rabies encephalitis is genotype 1.
- Domestic dog rabies continues to pose a threat to wildlife host species, livestock and human public health.
- Despite the greater specificity of genetic typing methods, broad coverage is more readily accomplished by the simpler protocols of antigenic typing (unusual lyssavirus strains).
- Two biotypes of rabies exist in South Africa.

Thank you for your attention!!!!



Rabies in Sudan

By

Dr. Yahia Hassan Ali

INTRODUCTION

- Rabies is existing in Sudan in human and animals.
- Rabies incidence during 2002-2003 was decreased and increased in 2004.
- Animal Rabies:
- Reported suspected rabid animals in Sudan during 2003-2005 were 252, 662 and 43., respectively. (Table 1,2).

Table 1: Species of reported rabies suspected animals in Sudan (2003-2005)

year	Dog	Cat	Monkey	Goat	Sheep	Donkey	Horse	Cattle	Camel	Total
2003	175	7	3	16	4	39	-	4	4	252
2004*	323	76	8	71	41	73	-	18	52	662
2005**	11	4	-	10	-	10	3	5	-	43
Total	509	87	11	97	45	122	3	27	56	957

*Data for 80% of the provinces

**Data for 60% of the provinces and up to September only

Table 2: Rabies suspected animals in different states

Year	Khartoum	Central	Northern	Eastern	Kordofan	Darfur	Total
2003	38	1	19	9	180	5	252
2004*	15	4	-	-	540	103	662
2005**	36	7	-	-	-	-	43
Total	89	12	19	9	720	108	957

*Data for 80% of the provinces

**Data for 60% of the provinces and up to September only

Laboratory Diagnosis:

Using FAT, 1 of 4, 3 of 8 and 9 of 10 samples were found positive during 2003, 2004 and 2005, respectively (Table 3).

Rabies Control :

The routinely applied strategy is vaccination of susceptible animals (using tissue culture vaccine) and destruction (by shooting) of stray and unvaccinated dogs

(Table 4).

Table 3: Results of rabies laboratory diagnosis using FAT (2003-2005)

Species	Result	2003	2004	2005	Total	% +ve
Dog	Positive	-	-	1	1	50%
	Negative	-	1	-	1	
Cat	Positive	-	-	-	-	0%
	Negative	2	2	-	4	
Goat	Positive	-	-	2	2	50%
	Negative	-	2	-	2	
Equine	Positive	-	-	5	5	83.3%
	Negative	-	-	1	1	
Cattle	Positive	-	2	1	3	50%
	Negative	1	2	-	3	
Camel	Positive	1	1	-	2	66.7%
	Negative	-	1	-	1	
Total	Positive	1	3	9	13	52%
	Negative	3	8	1	12	

Table 4: Rabies control measures in Sudan (2003 – 2005)

Year		Khartoum	Central	Northern	Eastern	kordofan	Darfur	Total
2003	Vaccination	234	37	7751	205	966	2676	11869
	Destruction	45	11	20	13	43	10	142
2004*	Vaccination	780	-	1482	-	-	27	2289
	Destruction	4144	-	500	-	-	159	4803
2005**	Vaccination	854	-	-	-	-	-	854
	Destruction	-	-	-	-	-	-	-

*Data for 80% of the provinces

**Data for 60% of the provinces and up to September only

Human Rabies:

The reported human rabies post-exposure treatments and deaths of the disease in Sudan during 2003-2005 were 9184/ 4, 9893/ 6, 12922/ 6., respectively (Table 5).

Vaccines:

Available human rabies vaccines are locally produced Goat brain vaccine, Duck embyo and Vero cell culture vaccines (Table 6).

Table 5: Human rabies post-exposure treatments and Deaths of the disease in Sudan (2003-2005)

State		2003	2004	2005	Total
Khatoum	Post-exposure	5062	4948	5378	15388
	Deaths	-	-	1	1
Central	Post-exposure	1148	1410	1645	4203
	Deaths	2	5	4	11
Northern	Post-exposure	141	228	117	486
	Deaths	-	-	-	-
Eastern	Post-exposure	406	377	739	1522
	Deaths	2	-	-	2
Kordofan	Post-exposure	794	1322	2000	4116
	Deaths	-	1	1	2
Darfur	Post-exposure	1445	1413	2630	5488
	Deaths	-	-	-	-
Southern	Post-exposure	188	195	418	801
	Deaths	-	-	-	-
Total	Post-exposure	9184	9893	12922	31999
	Deaths	4	6	6	16

Table 6: Locally produced and imported human rabies Vaccines in Sudan: (2003-2005)

Type of vaccine/ doses	2003	2004	2005	Total
Goat brain (locally produced)	47310	38740	47280	133330
Duck embryo (imported)	5000	-	-	5000
Tissue culture (imported)	25000	25000	15000	65000
Total	77310	63740	62280	203330

Conclusion:

Animal Rabies:

- Marked increase in rabies incidence in 2004.
- Dogs are the main animals involved in rabies.
- Very low animal rabies vaccination coverage.
- Weak rabies reporting system in most States.

Human Rabies:

- Increased No. of rabies post-exposure treatments in 2004, 2005.
- Most of cases are reported at Khartoum State.
- No laboratory confirmation is applied.

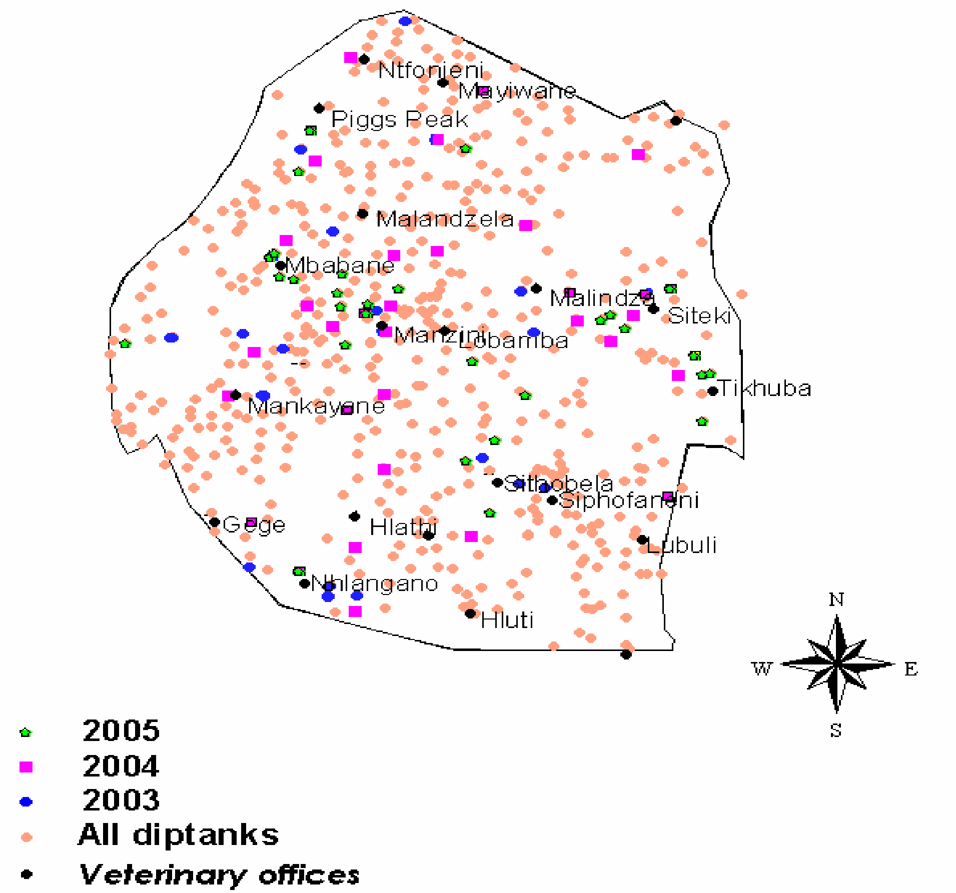
THANK YOU FOR ATTENTION


The Serial Killer

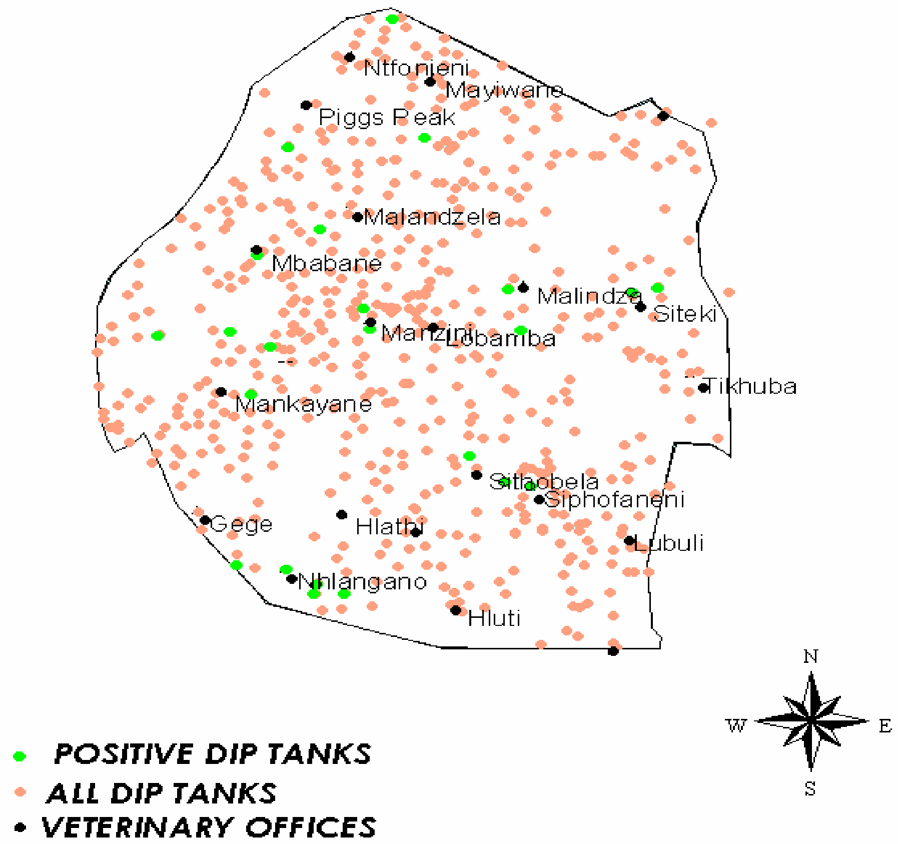
Table1: Animal Rabies Cases (2003- 2005)

Species	2003		2004		2005	
	Positive	Negative	Positive	Negative	Positive	Negative
Canine	30	6	38	15	26	22
Bovine	6	0	10	6	12	2
Caprine	2	1	4	2	9	2
Equine	0	0	0	1	0	2
Feline	0	0	0	1	0	3
Porcine	0	0	0	1	0	0
Other	0	0	0	0	0	0
Total	38	7	52	26	47	31

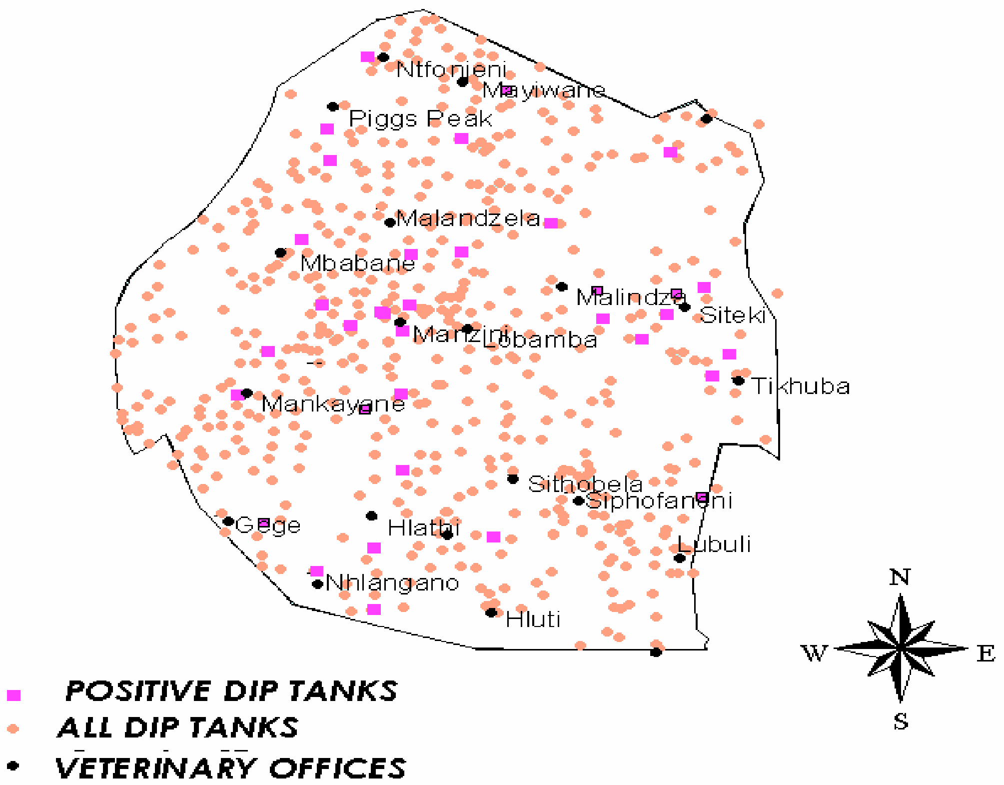
Rabies spatial distribution in Swaziland for the years 2003, 2004, 2005



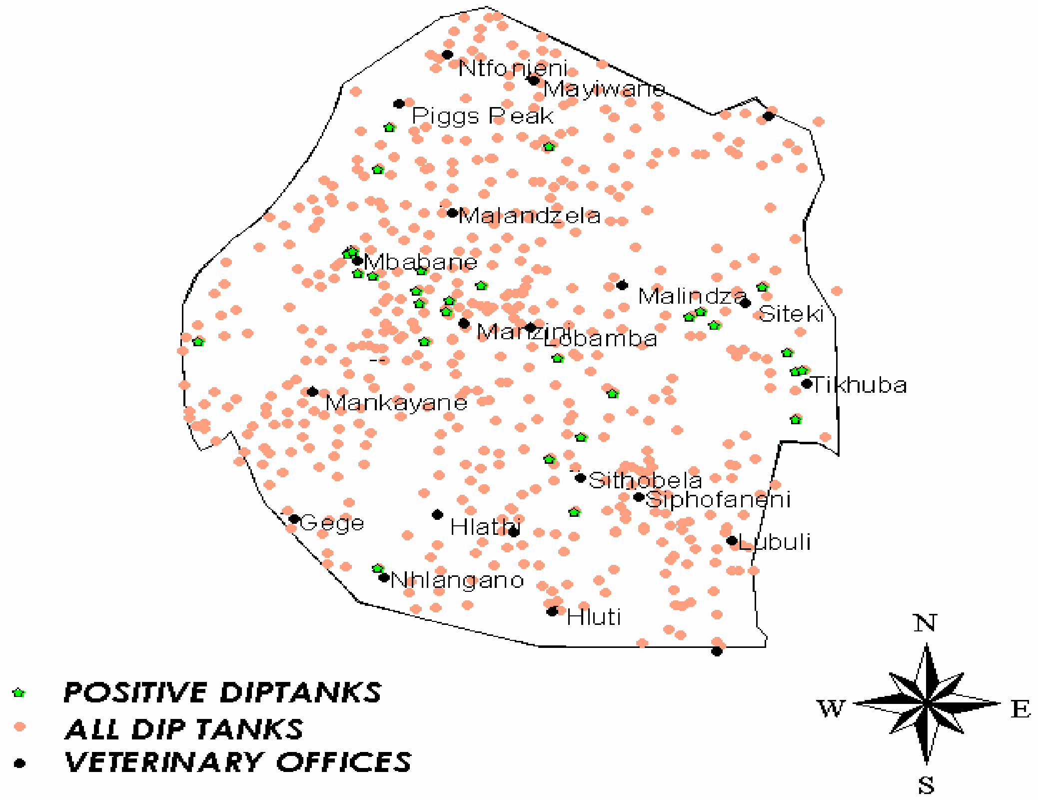
RABIES SPATIAL DISTRIBUTION (2003)



Rabies spatial distribution 2004



Rabies spatial distribution 2005



RABIES STATUS IN UGANDA

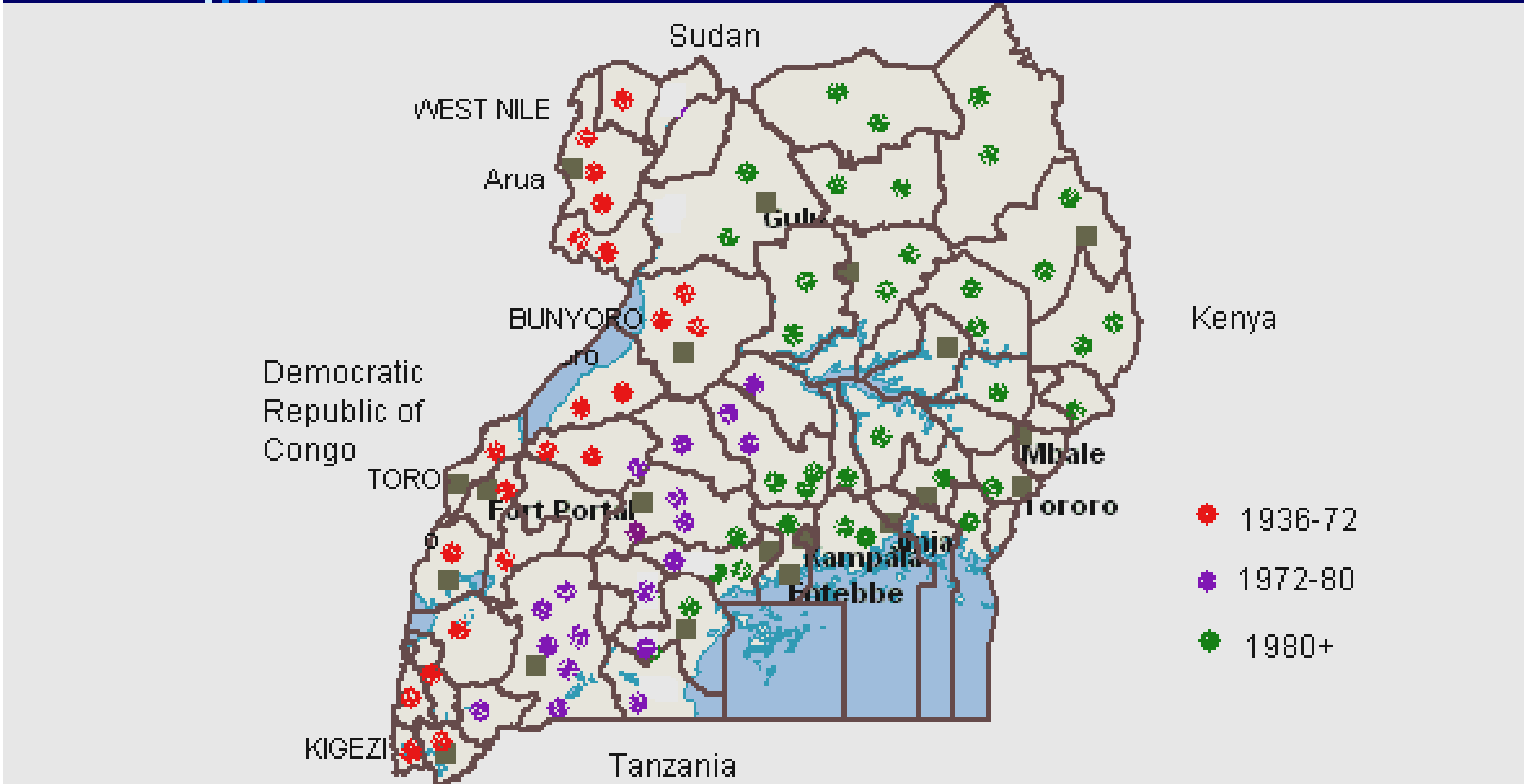
8th SEARG Conference, 22nd -26th January 2006, Windhoek-Namibia.

C. S. Rutebarika[1] , K. Mugabi[1], W. Kaboyo[2].

Introduction

- Rabies is endemic throughout the country.
- Both canine and sylvatic forms exist.
- Vaccination is major strategy for control.
- Ownerless and stray dogs remain a major constraint.
- Government has improved funding for rabies control.
- TECOR remains a constant pressure group.

The spread of rabies



Human rabies

- Cases recorded.
  - 105 in 2003
  - 18 in 2004.
  - 6 in 2005 (so far)
  - Under reporting and misdiagnosis are major constraints of status report.
  - 11,846 doses of anti rabies vaccine (ARV) were procured in2004/05.(Ugx 160m)
    - 7,750 doses were utilised for PET.
  - PET is free and intradermal route is a policy along with im.

Human rabies ctd

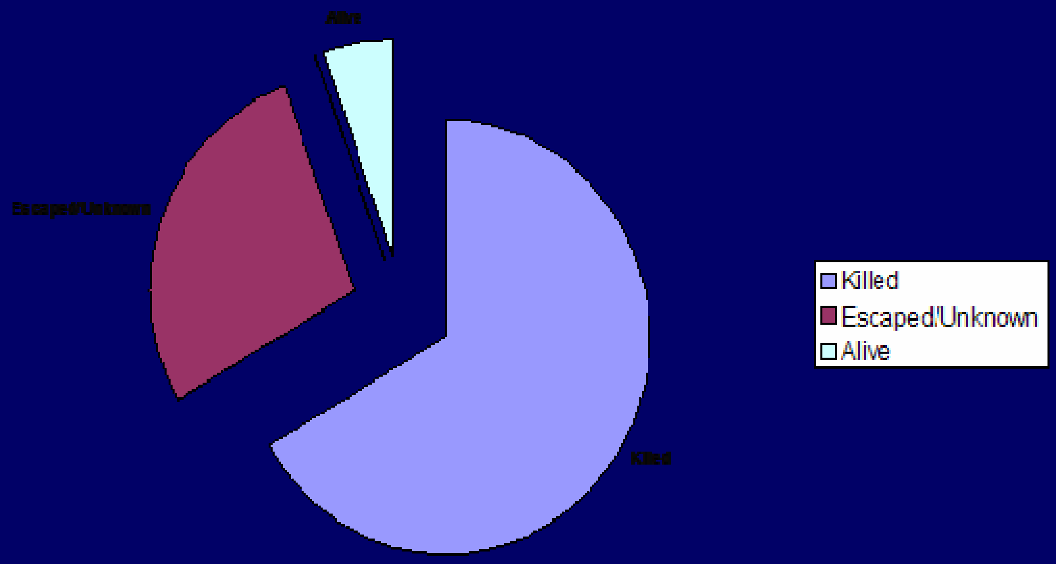
- Dog is biggest offender (95%).
- Male-Female ratio 50:50.
- Age group-5-19 years.(56%)
- 60% of offending dogs killed.

Human deaths from rabies in 2004

Month	Age (yrs)	Sex	Animal species	Fate of animal	Fate of patient
June	6	M	Dog	Killed	Died
June	8	F	Dog	Unknown	Died
June	20	M	Dog	Unknown	Died
July	26	M	Fox	Killed	Died
August	32	F	Dog	Died	Died
September	10	M	Dog	Killed	Died
September	13	F	Dog	Killed	Died
September	23	F	Dog	Unknown	Died
September	10	M	Dog	Killed	Died
September	23	F	Dog	Escaped	Died
October	48	M	Dog	Killed	Died
October	2	F	Dog	Killed	Died
October	5	F	Dog	Killed	Died
November	43	M	Dog	Unknown	Died
December	8	M	Dog	Killed	Died
December	17	M	Fox	Killed	Died
December	19	F	Dog	Alive	Died
December	42	F	Dog	Killed	Died
Total		**F = 9** **M = 9**	**18**		**18**

Source: Veterinary Public Health Unit, MoH-Uganda.

Fate of offending animals.

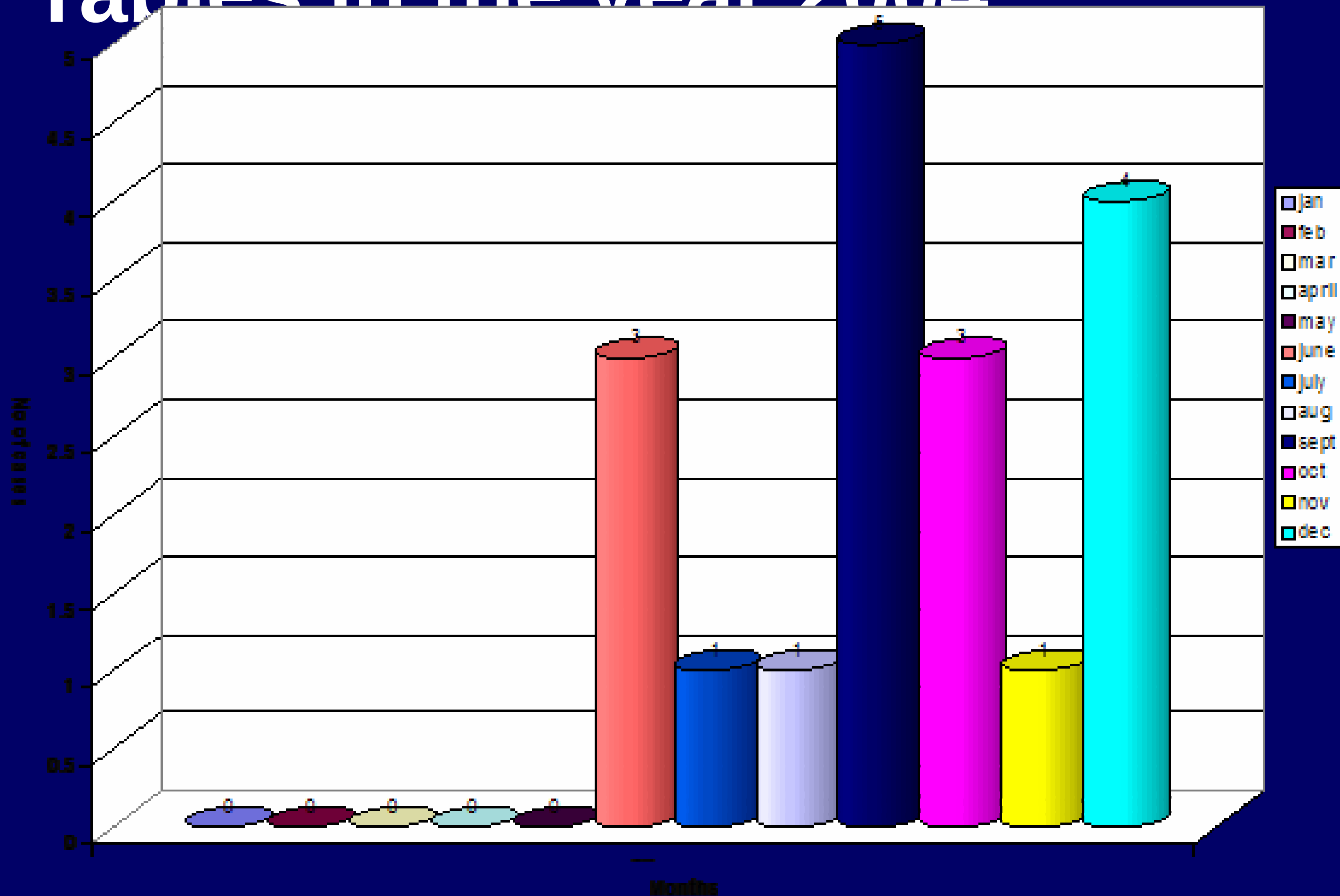


Human Rabies data (1992- 2005)

Year	No. of post exposure treatments	ARV doses (imported)	No. of rabies cases/deaths	Estimated cost (US$)
1992	766	3,976	50	35,000
1993	1,518	5,720	23	52,000
1994	2,614	8,298	15	74,700
1995	3,222	13,623	14	122,000
1996	1,698	16,000	9	144,000
1997	2,916	16,000	10	144,000
1998	3,112	16,000	10	144,000
1999	4,537	10,000	5	90,000
2000	5,398	10,000	12	90,000
2001	6,577	19,570	35	119,371
2002	4,789	15,133	12	98,365
2003	6,929	19,500	105	128,600
2004	4,628	10,550	18	79,000
2005*	7,750	11,846	6	88,000
Total	**56454**	**176,216**	**324**	**1,409,036**

* **Reports for 2005 are incomplete.**

Distribution of fatal human rabies in the year 2004



Animal Rabies control

- Government procured 540,000 doses ARV
- 296,358 doses utilised.
- Logistical support provided to local govts.
- Improved reporting due to PACE effort.
- 4,150 clinical suspected cases were reported.
- 5,615 stray dogs and cats were destroyed.
- USPCA's data on population mgt and rabies control not available yet.

Funding for rabies control, diagnosis and reporting

- GoU major funding agency.
- Diagnostic lab provided with FAT reagents (PACE and GoU).
- USPCA, PVPs and tertiary institutions procure own vaccines.
- No donor contribution (vaccines) yet.

Use of dog population reduction

- Population mgt
  - Poisoning
  - Shooting
  - Hunting
  - Pills, castration & spaying.

Constraints to Rabies control

- Irregular and insufficient funding
- Control of stray dog populations.
- Insufficient public awareness on Rabies.
- Civil service reform.
- Minimal role by the private sector.
- Civil strife in certain areas of the country.

Acknowledgements

- Dr. N. Kauta.
- Dr. S. Okware.
- Dr. R O Ademun.
- Dr. N. Nantima.
- Mr. J. Barisigara.

Thank you for attention

Country report SEARG 2006

Zimbabwe

Dr Munyaradzi. F. Chigiji

Introduction

- The Central Veterinary Laboratory in Harare is currently carrying out Rabies diagnosis in both animal and human samples.
- There is a close working relationship between the Ministry of Health and Child Welfare and the Veterinary Department in tackling issues of zoonotic diseases, rabies included.

Animal Rabies

- **Method used for diagnosis**
  - a) Fluorescent Antibody Test (FAT)
  - b) Mouse inoculation

Samples received and test results for the years 2004 and 2005

	2004		2005	
Number of domestic animal samples submitted	Rabies pos.	Rabies neg	Rabies pos.	Rabies neg
Dogs	211	129	135	97
Goats	28	6	13	6
Mule	0	1	0	0
Cattle	64	58	58	39
Pigs	0	2	3	1
Cats	8	12	3	8
Horses	1	2	1	1
Donkeys	4	1	8	3
Sheep	2	4	1	1
Total	318	215	222	156
% of tested	(59.7%)	(40.3%)	(58.7%)	(41.3)

Samples received and test results for the years 2004 and 2005

Number of wild animal samples submitted	2004 Rabies Pos.	Neg.	2005 Rabies Pos.	Neg
Jackals	7	2	6	6
Impala	0	1	0	0
Rat	0	1	0	0
Mangoose	0	1	0	0
Honey badger	0	0	0	1
Lemur	0	0	0	1
Elephants	0	0	0	1
Hyena	0	0	1	2
Bat	0	0	0	2
Total % of tested	7 (58.3%)	5 (41.7%)	7 (35%)	13 (65%)

Samples received and test results for the years 2004 and 2005

The Central Veterinary Laboratory does not have the capacity to assign the virus isolates into their respective lyssavirus genotypes.

Currently most experienced staff left the department and the research officers are in need of training.

- Without a base dog population it is impossible to calculate prevalence. Therefore the comparison is based on absolute numbers not proportion.
- There has been a decrease in the number of positive rabies cases from a total of 325 to 229 domestic and wild animals.
- The cause of this decline may have been:

1. Our surveillance system has compromised its' efficiency as a result of transport problems caused by the fuel problems encountered in 2005.
2. Reduced numbers of samples were delivered to the Central Veterinary Laboratory as most of the new farmers are still to be educated on the importance of laboratory diagnosis.

Human Rabies

	2004	2005
Total number of human rabies deaths **confirmed by laboratory tests** (Fluorescent Antibody Test and Mouse inoculation). *Figures obtained from (VLRDB).*	5	3
Total number of human rabies deaths **diagnosed on clinical grounds only** (i.e. without any further specimen examination by a laboratory*) Figures obtained from (MoHCW):*	38	7

Sources of human rabies.

	2004	2005
Domestic dog bites	4	2
Unknown source of rabies infection	1	1
Number of bites reported	4756	6326

Human Rabies

The majority (62.5%) of human rabies cases have been attributed to stray dog bites. This is mainly as a result of dogs being the major household pets in Zimbabwe. As a result of owners failing to feed their dogs and having inadequate space, the dogs begin to roam the streets. The dogs in the commercial farms are a high-risk group as they come into contact with the jackals which are considered the main reservoir of rabies amongst wild animals in Zimbabwe.

Vaccine use in the control of rabies

The vaccines for animal use imported into the country in the year 2004 were:

- ❖ Rabisin - 10 000 doses
- ❖ Nobivac - 5 000 doses
- ❖ Rabdomun - 112 000 doses

- Number of human rabies vaccines imported in the year 2004 was 18 000(6 000 of these were donated by UNICEF) and in the year 2005, 12000 vaccines were imported.
- In 2004 the number of people who received post-exposure treatment was 948.
- In 2005 the number of people who received post-exposure treatment was 610.

Conclusion

Challenges faced by Zimbabwe with regards to dog vaccinations are:

- The population of stray dogs is very high.
- Dog amenability to vaccination by the parenteral route coupled with the unavailability of an oral vaccine for dogs.
- Transport problems
- Vaccine shortages occur as a result of foreign currency shortages.
- Support services
- Education
- Land resettlement

Rabnet database

Nicolette van Zyl

University of Pretoria

HISTORY OF RABNET

- WHO collected data on rabies since 1959.
- Late 1990's: web-based version accessible through Rabnet.
- **Rabnet:** interactive information system able to generate maps and graphs of rabies data.
- Rabnet version 2: keeps information resources such as ready-made maps, rabies-related documents etc.

RABNET WEB ADDRESS

- http://gamapserver.who.int/globalatlas/home.asp
- Usernames and passwords available.

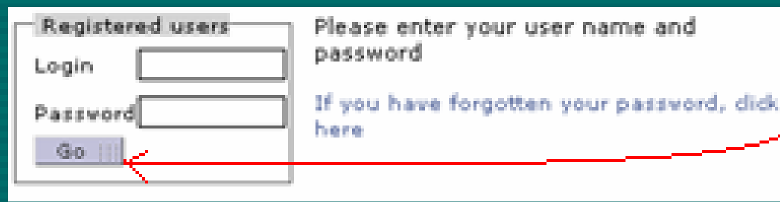
Welcome to the new Rabnet Questionnaire!

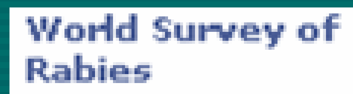
On the left side of the page you will find:

Registered Users Login

- Type in your specific country password and login, then click on 

to proceed

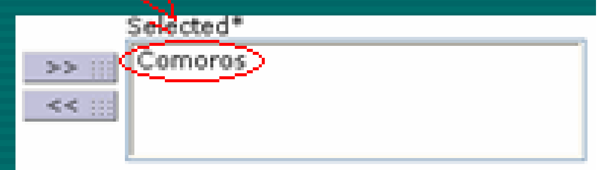


- Click on the 

textbox

- Now you have entered the questionnaire.

- Your country has already been selected. Should the wrong country appear, please contact us at: Rabnet@who.int



- Select the year you want to add data for.

Time Period

Year
From: 2004
To: 2004

- Click Go on the right bottom side of the page
- Now you will be able to choose the province for which you want to add data.

Province	Year	Date of Submission	Title	Name	Institution	Insert	Update	Delete
Baden-Wuerttemberg	2004					Insert		
Bayern	2004					Insert		
Berlin	2004					Insert		
Brandenburg	2004					Insert		
Bremen	2004					Insert		

- Click on "insert" and the questionnaire will appear!

Welcome to the WHO
Global Atlas of infectious diseases

- If you want to enter data for a province that has already be entered and submitted. Use the "Update" function.

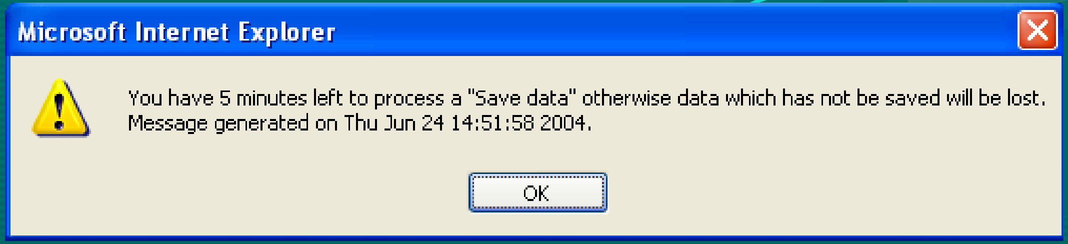
Province	Year	Date of Submission	Title	Name	Institution	Insert	Update	Delete
BERAT	2004					Insert		
DIBËR	2004					Insert		

- A country summery (total) will be automatically generated.

In this questionnaire you will see the time warning in red

IMPORTANT: In order to avoid "Session Timeout" (loss of data), you need to save the data at least every 20 minutes by using the "Save data" buttons at the end of each section of the form.

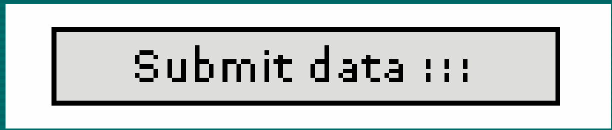
In addition, a pop-up textbox appears after 15 minutes:



To prevent being "timed-out" there is a textbox on the right bottom of every page. Every time you click on this button, the data is saved.

Save data :::

- Once you have entered all the data press:



on the **left bottom** of the page!

Welcome to the WHO Global Atlas of infectious diseases

- On behalf of the Rabnet team we thank you in anticipation of your kind cooperation!
- For any remark, question, other, please do not hesitate to contact us at: rabnet@who.int

Proposal for a SEARG website and electronic publication of proceedings

J. Barrat[1], MJ Duchene[1] and A. Barrat[2]

[1] AFSSA – LERRPAS, Malzeville, France
[2] ESIAL – Henri Poincaré University, Nancy, France

The CD

- First one done in 2001 in Lilongwe.
- Goal:
  - To facilitate the replication of proceedings
  - To have a cheap unprotected and free to copy medium
- Financially
  - Much cheaper than books
- On each CD :
  - Acrobat reader (for install and live CD) / Adobe reader
  - All the proceedings published since the beginning
  - Diagnosis manual

What can we update or change?

- "Principle" of the CD:
  - Cheap medium to collect all what has been presented so far during our meetings
  - One new version every meeting
- Do we update the diagnosis manual?
- Any other idea is very welcome

SEARG website

- Why?
  - Possibility to use a 20GB space free for 3 years (till end 2008)
  - With 227 papers presented so far in SEARG meetings, it may be useful to have an indexation system.
- How was this done?
  - Our librarian has keyworded these papers.
  - A database has been prepared with
    - Titles
    - Authors
    - Key words
  - The pre-version is ready

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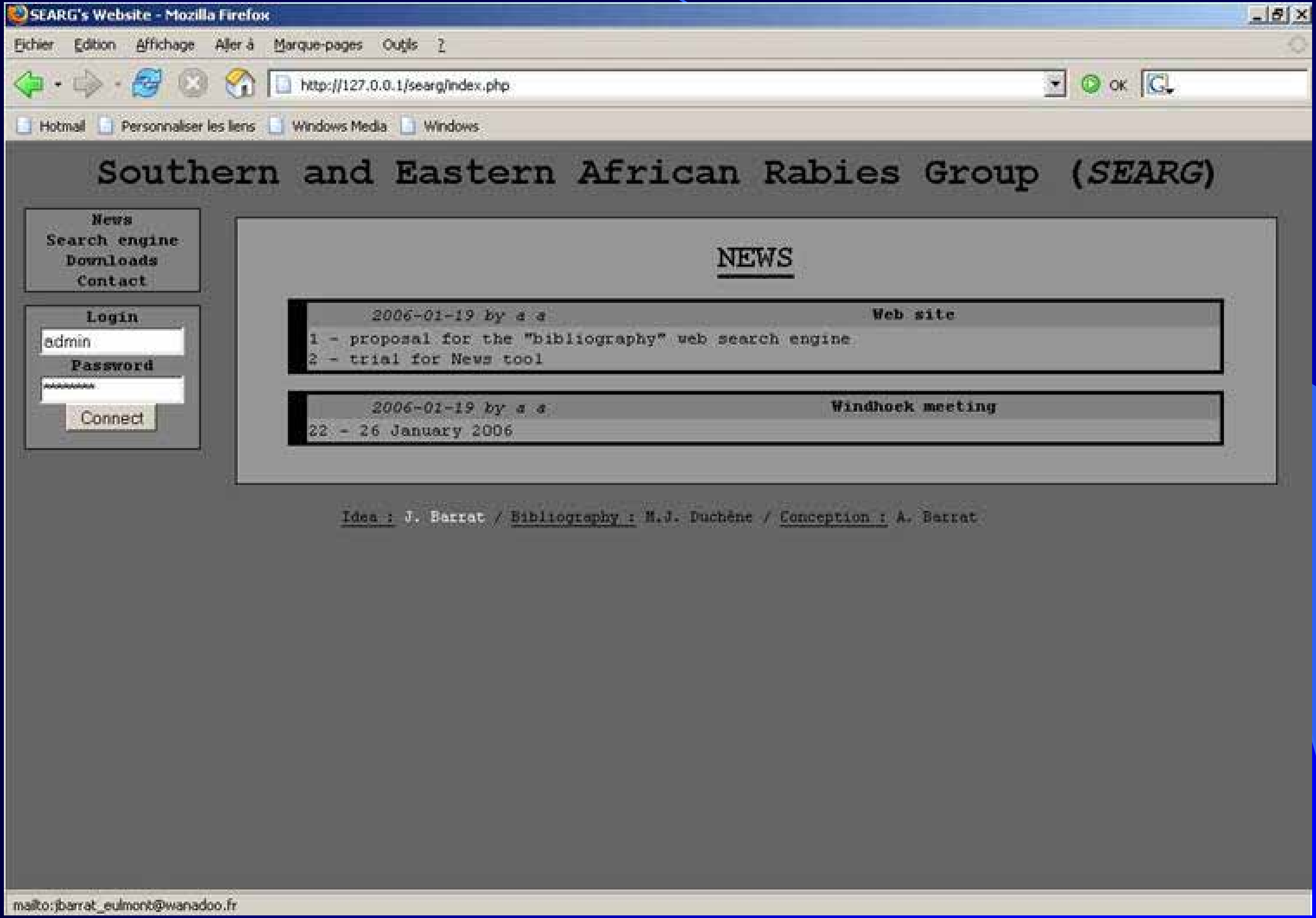

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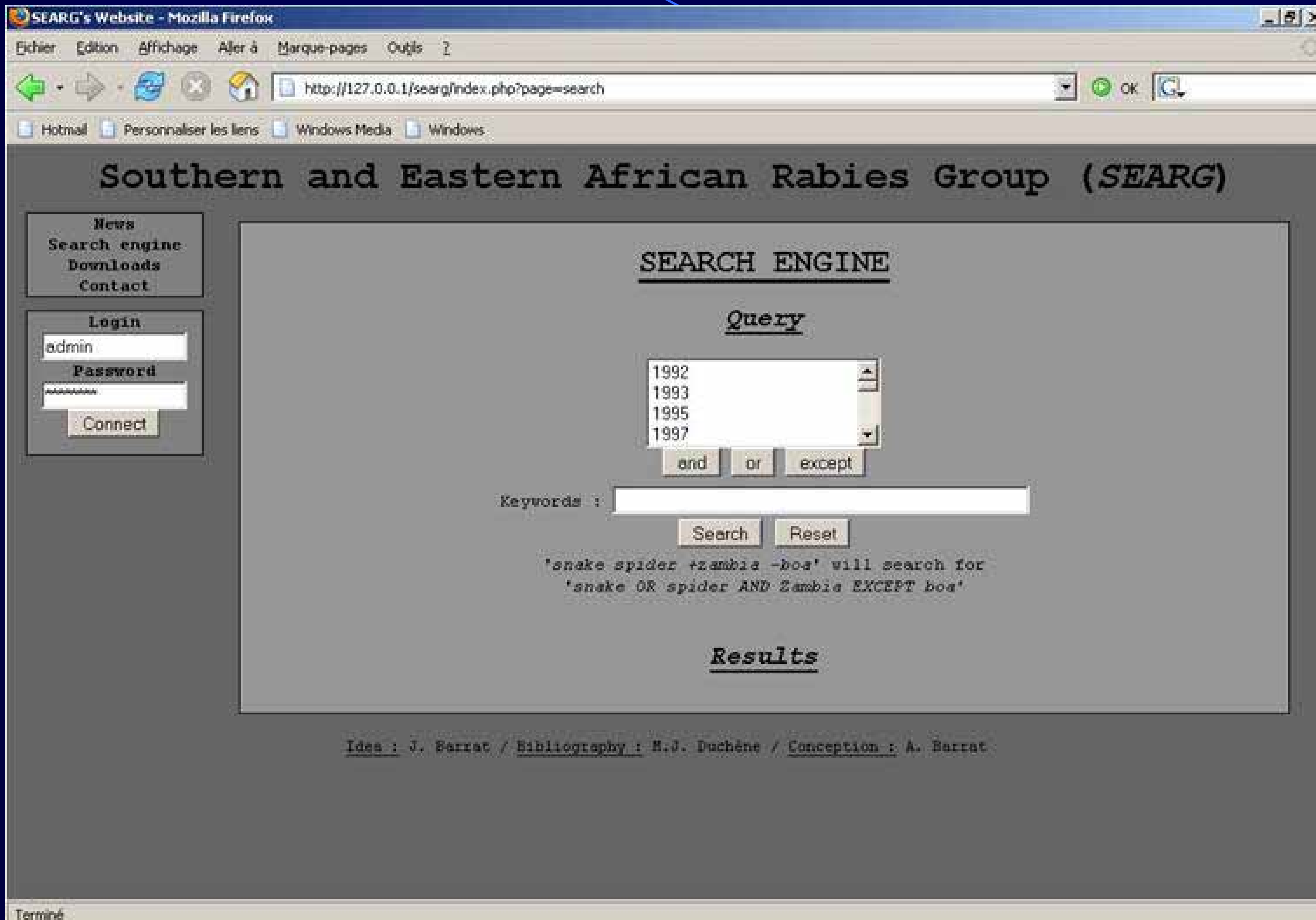
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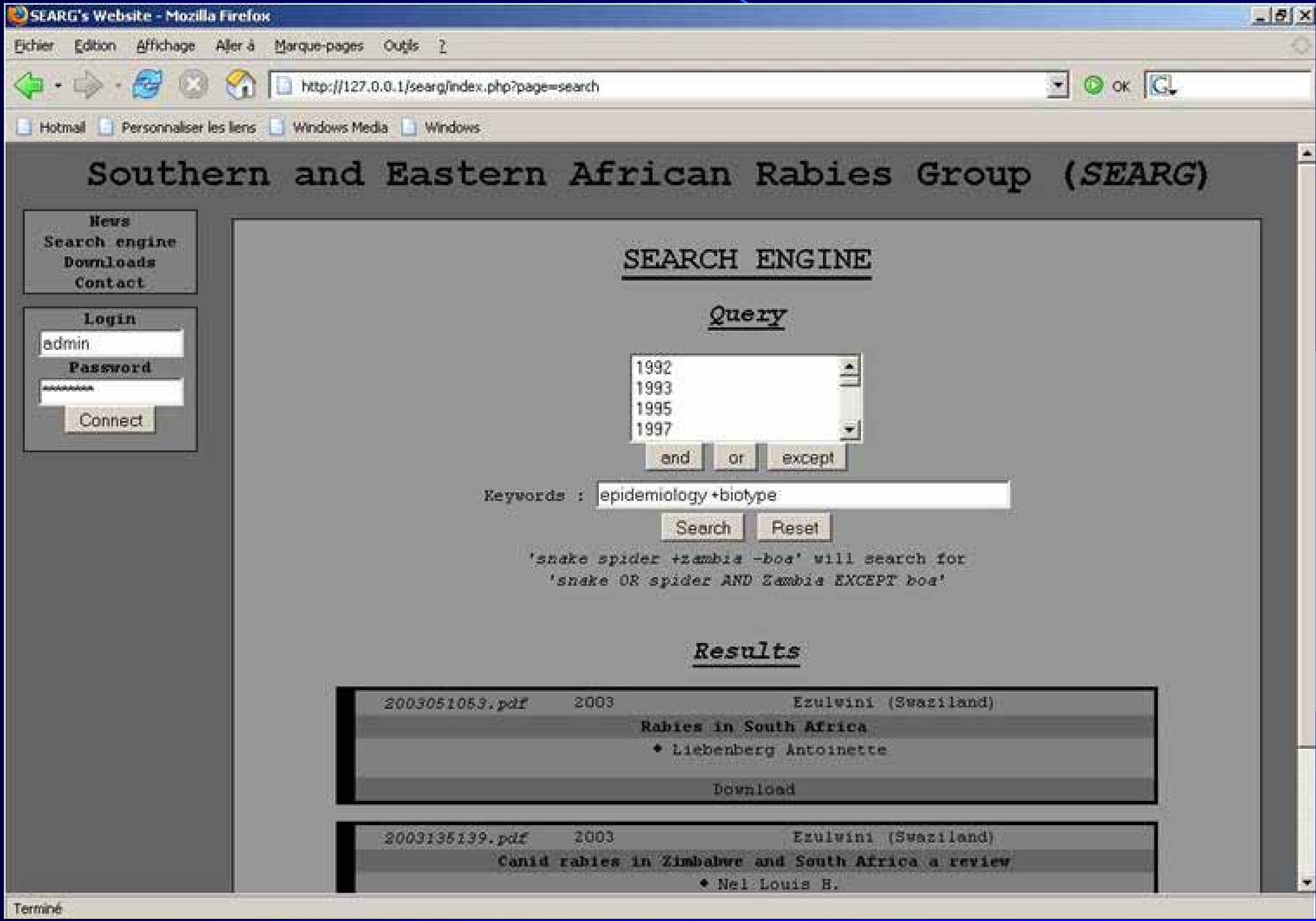
Result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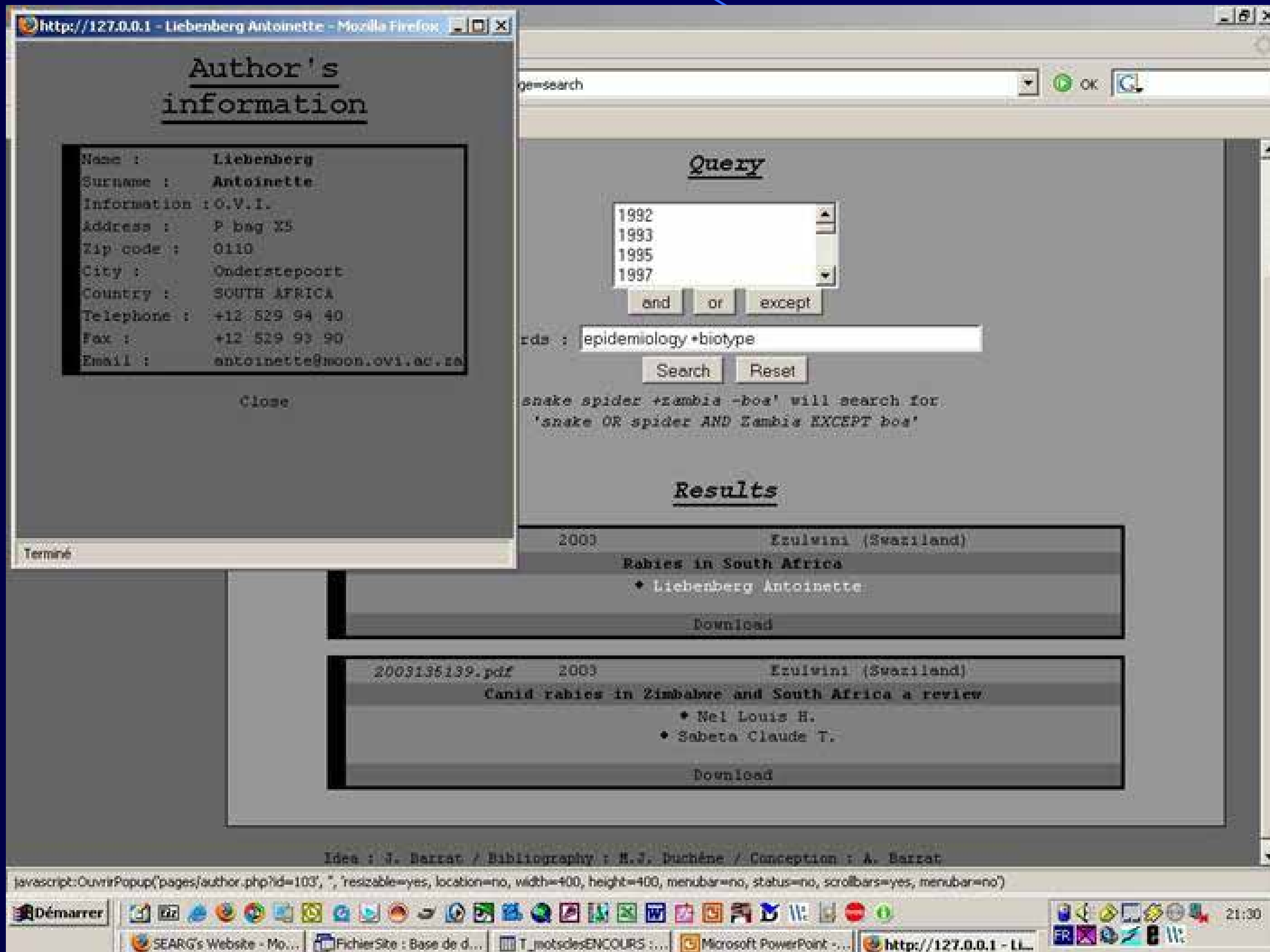

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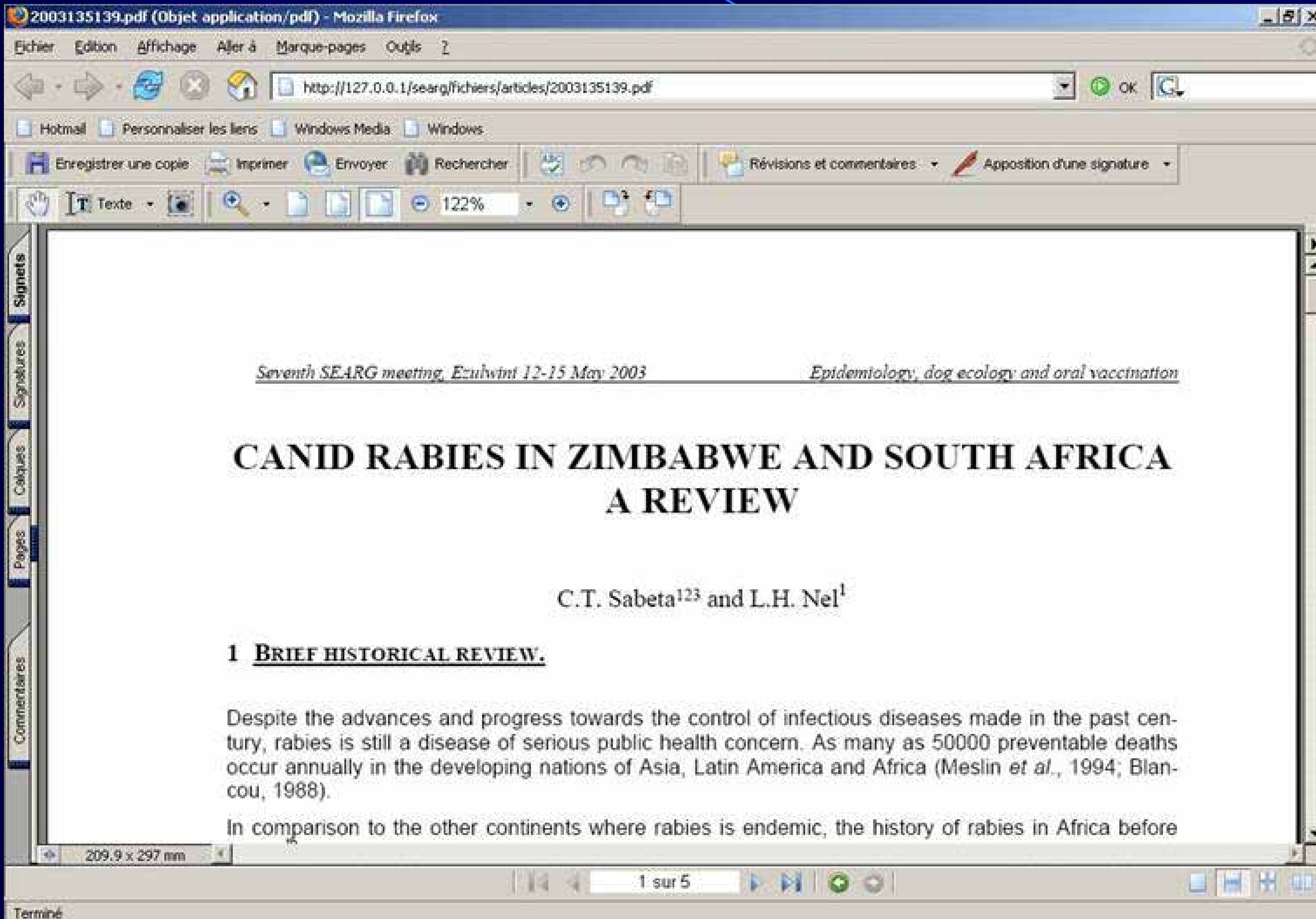

CANID RABIES IN ZIMBABWE AND SOUTH AFRICA A REVIEW

C.T. Sabeta[123] and L.H. Nel[1]

1 BRIEF HISTORICAL REVIEW.

Despite the advances and progress towards the control of infectious diseases made in the past century, rabies is still a disease of serious public health concern. As many as 50000 preventable deaths occur annually in the developing nations of Asia, Latin America and Africa (Meslin *et al.*, 1994; Blancou, 1988).

In comparison to the other continents where rabies is endemic, the history of rabies in Africa before

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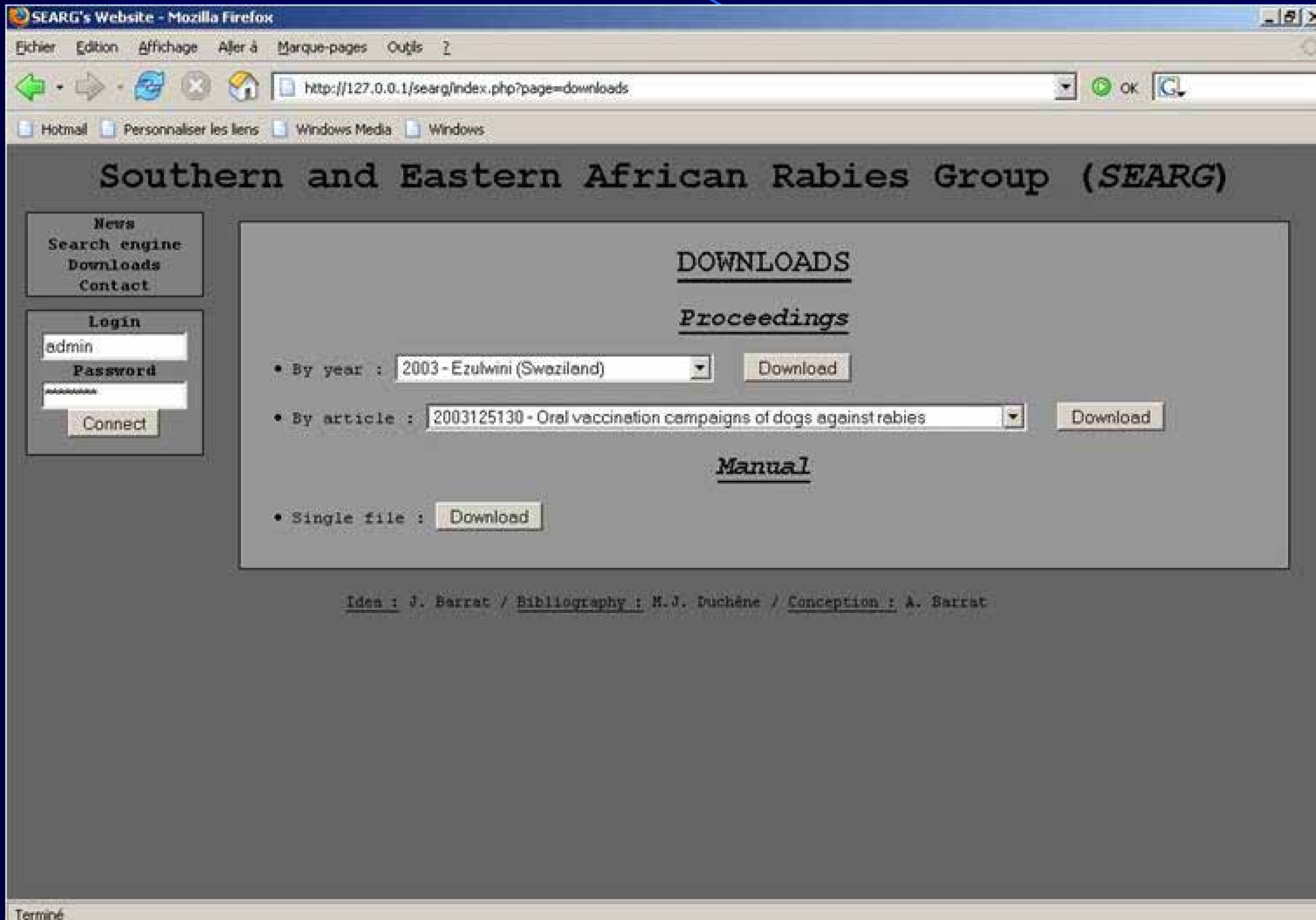
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What has to be decided now :

- The background : SEARG with the logo
- An information system should be added to exchange epidemiological data
  - Controlled by a supervisor (quite necessary) or not?
  - Used to provide epidemiological situation?
- A technical forum may also be made
  - Controlled by a supervisor (quite necessary) or not?
- Which quality of the documents?
  - The good quality used now means big files ➔long time for download
  - Lower quality ➔shorter files ➔shorter time for download

And now?

- Everything could be done for this summer

- When everything works at a good speed, the hyperlink can be given to WHO and OIE and ...

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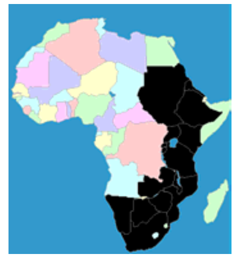

Dear friends and colleagues,

It is with pleasure that I write to you - on behalf of the conference organizers, in the wake of our 8th International SEARG meeting in Windhoek, Namibia.

Having had the opportunity to stand back and reflect after the inevitable hectic scramble of the meeting, I wish to sincerely thank Dr Otto Huebschle and our Namibian hosts as well as each of you for your interest and participation. You have made this a successful, pleasant and memorable meeting. I am also happy to announce that, courtesy of Jackie Weyer (the designer) and Nantu Phalatsi & others (photographers), you can view pictures of the meeting at the following website: http://www.seargwindhoek.photosite.com

I would also like to remind you that your country reports and all the other contributions for the proceedings should reach me by 21 March 2006. In this regard - I have some news: The Onderstepoort Journal of Veterinary Research (http://www.journals.co.za/ej/ejour_opvet.html) has agreed to consider the publication of selected articles in a supplementary or special edition. Therefore, if agreed and all goes according to plan, such articles for publication in the Onderstepoort Journal of Veterinary Research will be specifically invited from the pool of contributions that were submitted for the proceedings. Authors will have the opportunity to accept or decline the invitation.

Over the years we have made much progress with SEARG and I would like to use this opportunity to highlight what, in my opinion, are a few important issues:

1. SEARG is an important organization – the only one of its kind in Africa. We are recognized as an official body with a firm mandate by international organizations such as the OIE, WHO, IAEA/FAO and others. It is on this premise and based on the quality and composition of our programs that our donors have been willing and able to support us morally and financially.
2. SEARG has captured the interest and attention of the rest of the rabies world and we have assumed an international presence similar to our sister organizations in the Americas and in Europe. The emphasis is focused on the continent, but with a clear intention not to exclude anyone who cares to make a contribution. Rabies should not be addressed in isolation and need not be politicized.

3. A country report format has been agreed upon and has already led to an improvement of the quality of information shared and has allowed comparative evaluation and better analyses of the country situations.
4. Our delegate numbers have been growing steadily and this 8th meeting was one of the most well supported to date. Nineteen delegates from South Africa and an almost equal number from Namibia were financially supported by their respective governments. This indicates a tremendous commitment by these authorities and serves as an example of what can be achieved.
5. A constitution has been adopted and will lead the way forward – with clear guidelines ensuring the continued involvement of our members.
6. While SEARG has clear objectives and our meetings are specifically geared to these, there have been many additional benefits: Such benefits to the host countries include international exposure and recognition, press coverage and interest in the activities of the associated government departments. Also, satellite meetings of people who otherwise have to specifically travel around the globe to meet, have been taking place. Of much importance has also been the opportunity to discuss infectious diseases of animals and humans in the broader sense as most of our members are concerned with the control of multiple diseases in their respective countries.
7. Proceedings have been published regularly and an innovative website and opportunity for continued sharing of information via this dedicated SEARG website has been realized, thanks to the tremendous efforts of Dr Jacques Barrat.

There are, in my opinion, also a few areas in need of attention:

1. We should not underestimate the commitment and hard work that has brought us here, nor the dedication and hard work that will be required from the entire SEARG steering committee, its adopted members and all regular members to make this organization self-sustained.
2. Our members and country delegates should not create the impression that they are unwilling to seek financial support from their governments and departments toward representing their very own countries. We have to make some contributions to our own affairs and the reality is that we will have to move beyond a total dependence on being fully sponsored by external organizations. We owe it to our many friends, donors and sponsors to show progress and a willingness to reciprocate. Ideally, we will be able to once again reach the goal of having representation from both the Agricultural and Health sectors from each of our member countries.
3. Whereas in the past SEARG meetings depended on a few individuals, SEARG should evolve beyond this. Therefore, we should actively and continually seek, encourage and involve new and upcoming leaders.

Finally, I am very pleased to inform you that the Director of Animal Health and Production of Botswana has agreed to host the next scheduled meeting of SEARG in 2008 (details to follow in due course). By agreement, Mozambique is now first in line as potential hosts for 2010. I would like to thank the Departmental directors of both these countries for their prompt reactions, following the 2006 Namibian meeting.

I look forward to a productive two years and until we meet again, in the beautiful Botswana, I thank you again and wish you all the best.

Louis Nel
Pretoria
7 February 2006