

Rabies in the Eastern Cape Province of South Africa – Where are we going wrong?

S J van Sittert^{a*}, J Raath^b, G W Akol^c, J M Miyen^d, B Mlahlwa^a and C T Sabeta^d

ABSTRACT

Rabies is a growing problem in the Eastern Cape Province of South Africa. This study investigated dog ecology, vaccination coverage and rabies neutralising antibody levels in 203 randomly selected dogs within a local municipality in the former Transkei area. Responses to vaccination were also evaluated in 80 of these dogs. The population was remarkably uniform in size, breed and condition. Slightly over 1/5th of the population was between 6 weeks and 1 year of age, while very few dogs reached 10 years or older. According to owner responses, the Animal Health Technicians achieved a total vaccination coverage of 65 % of owned dogs over several years, but only 56 % within the previous 12 months. Only 32 % of dogs had adequate circulating rabies virus neutralisation antibodies ($\geq 0.5\text{IU/l}$). After vaccination, 83 % had seroconverted to this level. The magnitude of seroconversion was independent of body condition or age. This study proposes a different approach to vaccination strategies than those currently employed in certain areas of the province.

Keywords: dog, Eastern Cape, ecology, lyssavirus, rabies, seroconversion, South Africa, Transkei, vaccination.

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INTRODUCTION

Rabies is a fatal disease caused by a virus of the family *Rhabdoviridae*, genus *Lyssavirus*. The severe nature of the estimated 55 000 human deaths a year caused by the disease, mainly in Africa and Asia²⁴, underline its impact. *Lyssavirus* currently includes 11 genotypes recognised by the International Committee on Virus Taxonomy¹⁹, 4 of which have been isolated in South Africa to date (Rabies virus, Mokola virus, Lagos bat virus and Duvenhage virus)³¹. For the Rabies virus genotype, the domestic dog is the major reservoir and vector and is responsible for most human cases worldwide³¹. However, different Rabies virus biotypes within particular species and geographical areas can exist⁴⁰. For example, in southern Africa 2 biotypes (canine and mongoose) are

recognised. Mongoose rabies is adapted to and circulates within herpestid species, particularly the yellow mongoose (*Cynictis penicillata*). The cosmopolitan canid biotype, on the other hand, circulates within members of the family Canidae: dogs (*Canis familiaris*), black backed jackals (*C. mesomelas*) and bat-eared foxes (*Otocyon megalotis*)^{31,32,35}. While mongoose rabies is considered to be indigenous to the area, canine rabies appears to have been introduced during modern times³¹.

The 1st confirmed diagnosis of rabies in South Africa was in 1893 following an outbreak of the disease in the Eastern Cape from an imported dog^{6,40}. This outbreak was brought under control, however, and the spatial spread of rabies was then closely associated with wildlife species particularly the yellow mongoose. By 1940, canine rabies had begun to spread south of the Zambezi River so that by 1950 it appeared in South Africa in the northern Limpopo Province, where it is still present^{6,26,40}. Subsequent to the 1950 Limpopo outbreak, canine rabies spread to Mozambique from where it entered KwaZulu-Natal in 1961^{25,40}. Although the KwaZulu-Natal outbreak was brought under control, the disease reappeared in 1976 following an influx of refugees from Mozambique⁴⁰. This outbreak could not

be brought under control and marked the start of the incessant southward spread of canine rabies. In 1986, the disease reached the Eastern Cape, when it was confirmed in the northern areas of Transkei (Maluti and Umzimvubu local municipalities of the Eastern Cape Province)². Over the next 4 years, canine rabies continued to spread throughout Transkei and by the early 1990s had reached East London. Currently, rabies is a re-emerging public health problem in the Eastern Cape, evident by the fact that from 2008 to 2009 more human cases were reported than from any other province⁵.

Although rabies has been confirmed in wild and domestic animal species in the Eastern Cape, canine rabies was the main driver of the observed temporal trends over the period 1986–2009 (Fig. 1). The mongoose biotype in the Eastern Cape is maintained especially in the Karoo and adjoining areas (Cacadu, Western Chris Hani and Ukhahlamba District municipalities), but is absent in the former Transkei area³². Among domestic animals in the Eastern Cape, rabies has been diagnosed predominantly in dogs (52 %) followed by cattle (34 %), goats (6 %), sheep (4 %), domestic cats (2 %) and other domestic animals (2 %) over the period 1986–2009 (Eastern Cape Department of Agriculture, Forestry and Fisheries, unpubl. data). The high incidence of rabies in cattle and other dead-end hosts compared to the rest of South Africa¹⁸ seems to indicate that surveillance coverage is still insufficient, and that the burden of rabies in the province may be underestimated.

Control of rabies through targeted vaccination of at least 70 % of the dog population is the standard method of controlling rabies and has been used successfully in developed countries^{10,42}, although a reciprocal increase (whether real or relative) of the disease in wildlife species may occur³². Vaccination of dogs and cats has been implemented in the Eastern Cape Province on an annual basis since 1986, but has neither curbed the spread nor prevented the rise in the number of canine rabies cases and dead-end host species, probably because less than 70 % of the population was reached². The problem is compounded by the lack of information

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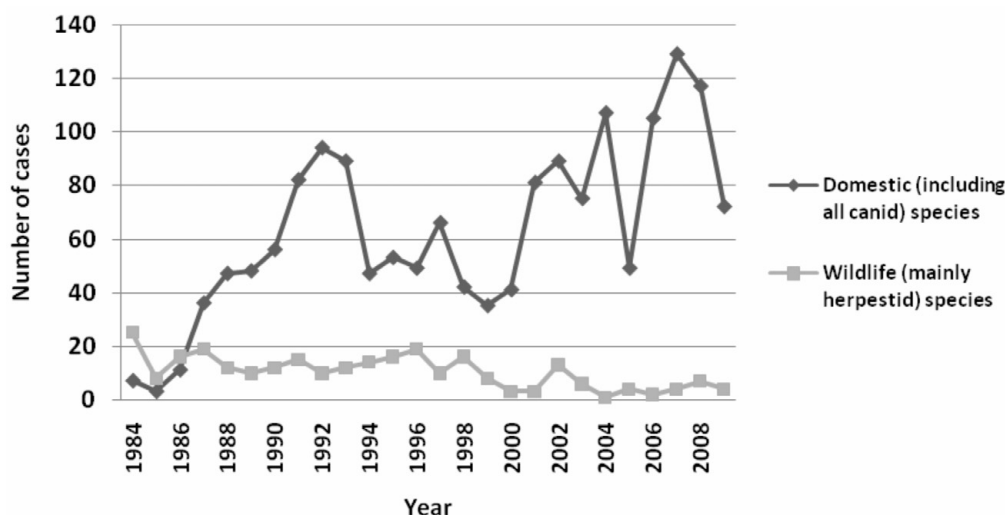


Fig. 1: Trends of rabies in domestic (including all canids) and wildlife (predominantly herpestids) species in the Eastern Cape Province during the period 1984–2009. Rabies cases reported as domestic prior to 1986 (typing results not available) were cases that occurred in 2 bat-eared foxes (1984), 1 bovine (1984), 1 canine (1984), 2 feline (1984 and 1985) and 1 caprine (1985), all of which occurred in the western part of the province that is predominantly endemic to herpestid rabies. (Eastern Cape Department of Agriculture, Forestry and Fisheries, Veterinary Services, unpubl. data).

on the ecology of the dog population in this province. Indeed, information on dog population ecology appears to be very limited in most parts of Africa³³. In order to develop an effective approach to rabies control it is essential to understand the dog ecology and establish monitoring measures for assessing the effectiveness of the vaccination campaigns given the shortcomings highlighted above. This paper outlines preliminary findings of a study aimed at identifying some of the key ecological factors that appear to influence the successful immunisation of dogs against rabies in a communal farming area in the province. The study further investigated factors that may influence the ability of dogs to respond effectively to rabies vaccines.

MATERIALS AND METHODS

Study area

This study was conducted in the Emalahleni Local Municipality, which is administered from the central town of Lady Frere (27°14'E; 31°42'S), and is part of the greater Chris Hani District Municipality in the Eastern Cape Province of South Africa (Fig. 2). The majority (3317 km²) of the 3551 km² area comprises what was formerly Transkei where raising livestock on communal grazing is the main farming activity. In the northernmost area the people farm livestock on a commercial level. Apart from 3 small towns, the mountainous area consists of rural villages of varying population sizes. The housing in the district is generally traditional round huts and more modern brick houses, especially in the towns. The total human population in the Emalahleni municipality was estimated at

125 288 in 2007 (Statistics South Africa, pers. comm.). No formal census data of dog numbers in Emalahleni are available, and thus the dog population could only be estimated as ranging from 8348 to 25 058, based on a human to dog ratio of 1:5 to 1:15²³. During 2008–2009 10 cases of laboratory confirmed rabies (8 dogs, 1 bull and 1 goat) were reported in Emalahleni. These cases were separated temporally throughout the 2 years and spatially across the district, and it was not established if it comprised 1 or many outbreaks. Every year a rabies vaccination campaign is launched throughout the municipality, where dogs and cats are vaccinated free of charge through a street to street or farm to farm vaccination strategy. This proactive vaccination campaign is conducted by animal health technicians (AHTs) in addition to additional vaccinations in the face of an outbreak.

Sampling methods

The dog population density is proportional to the human population density and is often remarkably similar across different African communities^{9,20,28}. As a sampling strategy, 203 GPS coordinates were randomly generated throughout the municipality. In order to reflect the expected spread of the canine population, however, the coordinates were weighted/focused towards areas of higher human population density. This was carried out by selecting 1.4 %, 2.9 %, 7.4 %, 14.7 %, 29.4 % and 44.1 % of the GPS coordinates to fall in human population densities of 0–5, 5–10, 10–25, 25–50, 50–100 and >100 people per square kilometre respectively (Fig. 3). ArcGIS® software (ESRI Inc., Redlands, USA) was used to randomly

allocate biased coordinates throughout the municipality based on data from the South African Geo-referenced Information System (www.agis.agric.za). These coordinates were located with a Garmin® nüvi® 220W series GPS tracking device (Garmin Distribution Africa, Honeydew, South Africa). The closest household to each set of coordinates that owned a dog or dogs was used as a sampling point.

Sample collection and storage

The most recent vaccination campaign preceding the study was from October to December 2008. Sampling took place from November 2008 to May 2009. Sampling that coincided with the 2008 campaign (i.e. November and December 2008) was done within areas that were already covered during the 2008 campaign. At each sampling point, 1 dog of at least 6 weeks of age was randomly selected. With the owner's consent, blood was collected from the cephalic, lateral saphenous or jugular veins. The blood was allowed to clot before being centrifuged and the serum stored at –20 °C within 12 hours. On the day of blood collection, the dog was injected subcutaneously with 1 ml of the same vaccine used during the 2008 rabies campaign, namely Rabdomun® (Intervet/Schering-Plough, Isando, RSA (batch A667B2)). Each ml of vaccine had an antigenic value of ≥5.0, as determined by the modified NIH test, and at least 10^{7.3} MLD₅₀ Flurry LEP strain virus grown on Baby Hamster Kidney (BHK) 21 cell culture, inactivated with an aziridine compound and absorbed into aluminium hydroxide (package insert). A questionnaire administered in isiXhosa, Afrikaans or English was completed through an owner interview for each dog

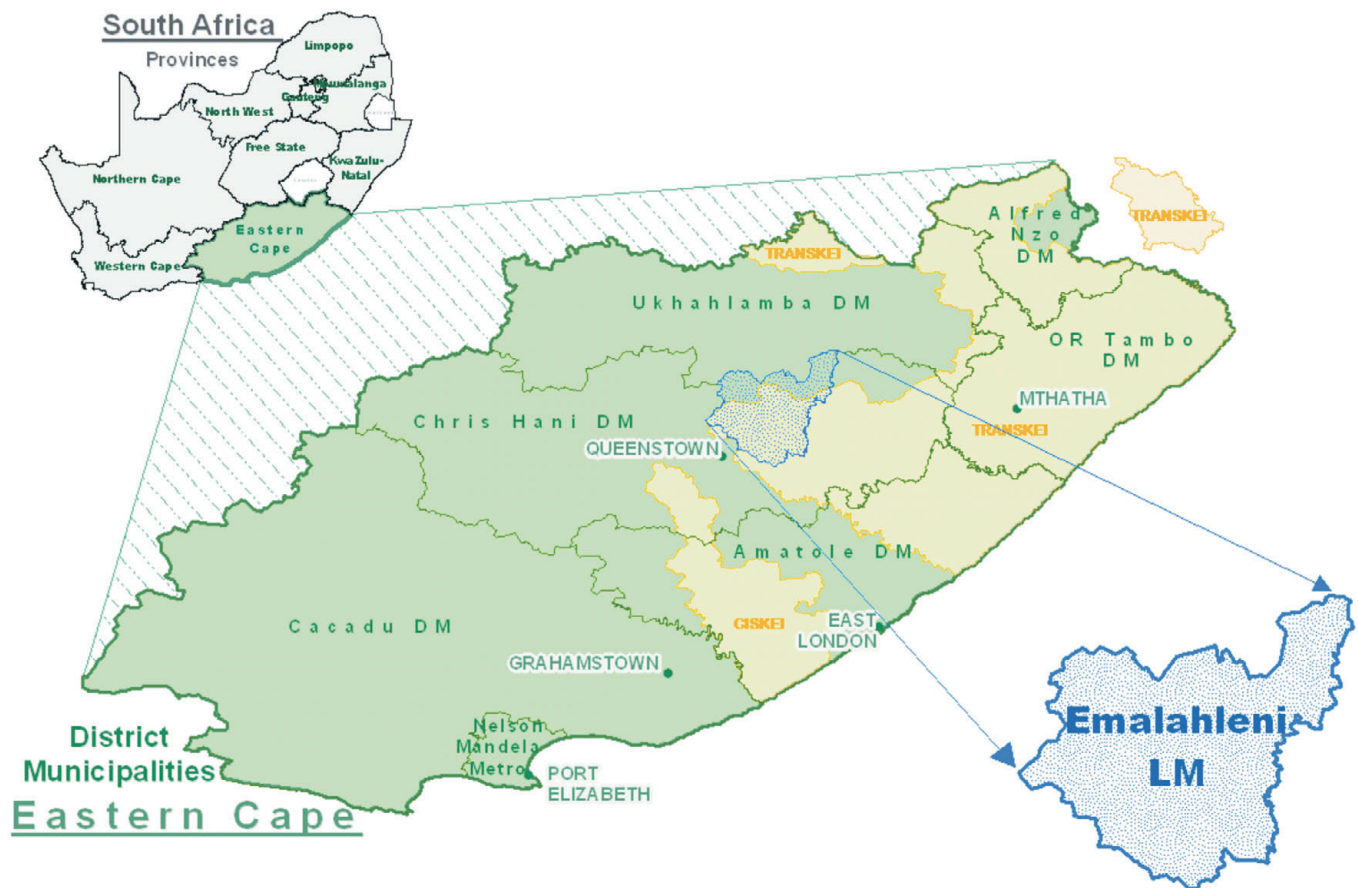


Fig. 2: Location of the study area within the Eastern Cape, South Africa. The map indicates the former Transkei (North) and Ciskei (South) areas, as well as the Emalahleni local municipality, which formed part of the westernmost part of the former Transkei area.

in the study sample. The questionnaire noted the GPS coordinates of the household, the name of owner and the name of the locality, i.e. village/farm. Data recorded about the dog included name, age, sex (male/female/sterilised/lactating), breed, size, condition, rabies vaccination history, function (pet/security/hunting/herding) and any noteworthy external characteristics (external parasites/injuries/external tumours). The dog in question was classified as confined (i.e. permanently confined) or roaming freely (including semi restriction, i.e. access to the general dog population some of the time without owner supervision). Rabies vaccination history was ascertained by asking if the dog was previously vaccinated (yes/no/unsure) and if previously vaccinated, when last (less than 12 months ago/more than 12 months ago/unsure). In most cases, however, the vaccination history could not be verified. The condition of the dog was subjectively classified as 'poor', 'thin', 'good' or 'overweight': dogs with mange, hair loss, high external parasite burden or which were emaciated were placed into the 'poor' category; 'thin' dogs had a low muscle and fat mass, a well kept coat with no or few external parasites; 'good' condition dogs had an adequate muscle and fat mass, a

well kept coat with no or few external parasites and overweight dogs had a well kept coat with no or few external parasites and a fat mass which made palpation of bony prominences on the spine and pelvic area difficult. Interviews and condition scoring were conducted by the local state veterinarian. It should be emphasised that parasite burden was judged only on the presence of adult ticks and fleas seen or felt on the dog's head and external pinnae, dorsal and lateral body wall or tail. No attempt was made to collect or count these parasites. Any dog additions or losses to the household in the past 2 years were recorded as well as the current number of dogs living at the house. A post-vaccination blood sample was taken between 30 and 60 days (mean = 52 days) after the 1st sample from 80 of the sampled dogs. The selection of dogs for the post-vaccination sample was done randomly.

Determination of rabies antibody titres

Rabies antibody titres were assessed using a standard 48 hour fluorescent antibody virus neutralisation test (FAVNT) as described previously⁷. Briefly, 3-fold serum dilutions were incubated with a 100 TCID₅₀/l of challenge rabies virus (ATCC VR959, CVS-II) and any un-neutralised

challenge virus allowed to grow on susceptible Baby Hamster Kidney cells (BHK C13-ATCC: CL-10, Diagnostic Hybrids, USA). Virus growth on BHK cells was detected by acetone-fixation of the monolayer and stained with fluorescein isothiocyanate (FITC)-labelled anti-rabies monoclonal hyperimmune serum (Onderstepoort Veterinary Institute, Pretoria, South Africa). The microtitre plates were examined under a fluorescence microscope to detect infected cells and 50 % endpoint titres calculated using the Spearman-Kärber method^{21,38,41}. The FAVN tests were performed at the Onderstepoort Veterinary Institute, an OIE rabies reference laboratory.

Data analysis

Data were analysed with SPSS[®] statistics 17.0 (SPSS Inc, Chicago, USA). Proportions of responses were evaluated through a chi-square test for an $r \times k$ contingency table. Where proportions of previously vaccinated dogs were evaluated, 'unsure' responses ($n = 5$) were omitted. Serum titre results in International Units per millilitre (IU/l) were assessed for normality through normal probability plots. Since the distribution of serum titres were skewed to the right, data were log transformed. The difference between the 1st

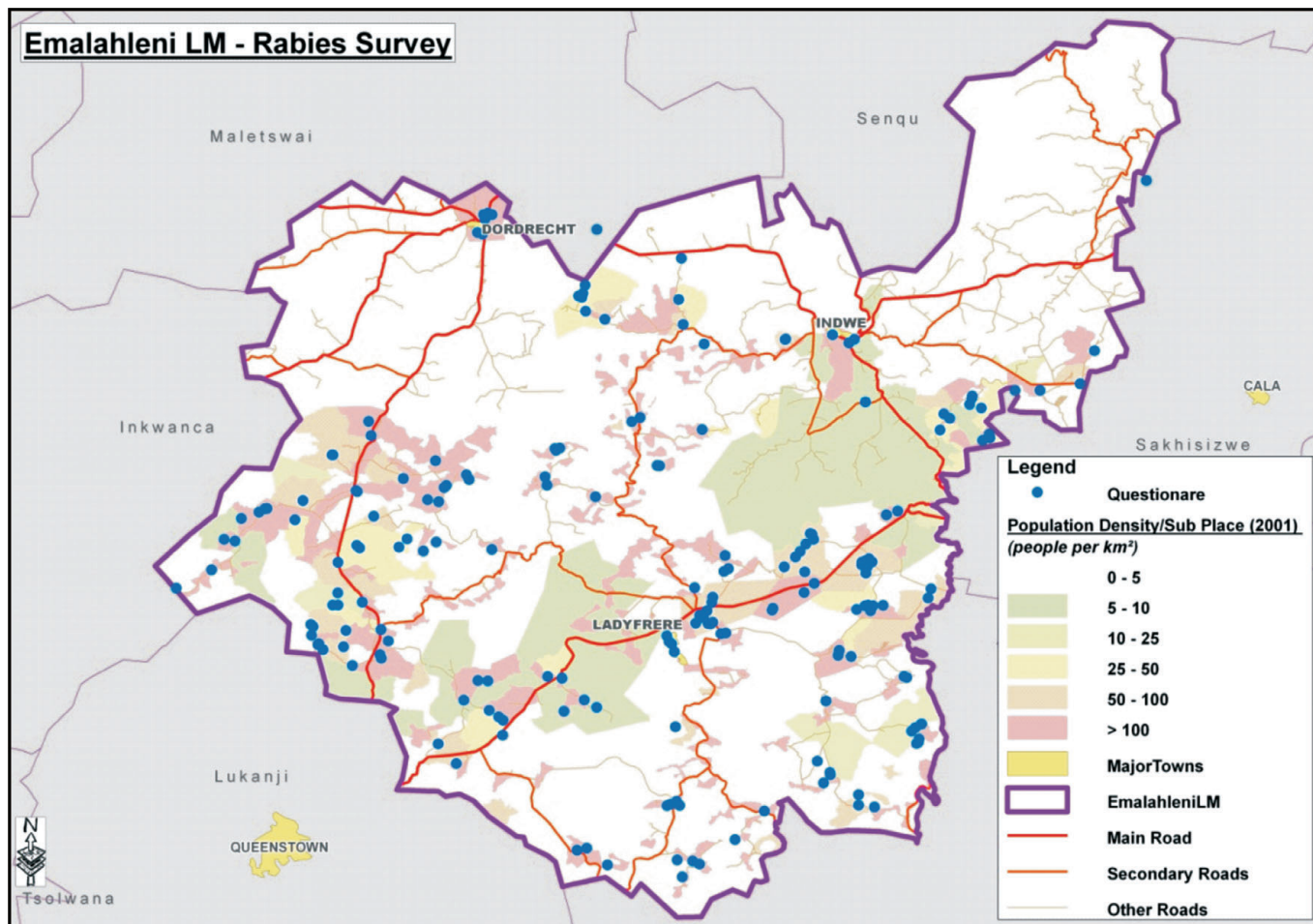


Fig. 3: Emalahleni Local Municipality indicating population density and the sampling points.

and 2nd (30–60 days post-vaccination) serum rabies antibody titre were evaluated through a paired *t*-test. A repeated measures analysis of variance (or mixed design ANOVA) with 'age group' and 'condition score' as between subject factors, was used to evaluate the interaction effects of these factors with the repeated serum titre measurements. For purposes of this mixed design ANOVA, the age group factor was pooled into the following categories: less than 1 year, 1–3 years and more than 3 years. The body condition factor was grouped into the following categories: poor, thin and good/overweight. The level of significance was set at $P < 0.05$.

RESULTS

Description of the canine population

The majority (96 %) of the dogs were classified as a medium-sized dog, while large dogs represented 3 % and small dogs 1 %. The breed most represented in the study could be classified as Africanis, or the local landrace¹⁷, comprising 93 %, while cross-breeds represented 4 % and other dog breeds 3 % of the population. The male:female ratio was 1.7:1. Owners very often did not know the exact age of their dogs, but were able to classify them

into the age groups specified in the questionnaire. The dogs in the sample population had the following age group distribution: 6–12 weeks = 3 %, 3–12 months = 18 %, 1–3 years = 43 %, 3–10 years = 35 % and more than 10 years = 1 %. The dogs were generally in a 'good' condition with 3 %, 76 %, 14 % and 7 % of the sample population classified into 'overweight', 'good', 'thin' and 'poor' categories respectively.

Nearly a quarter of the households (23 %) reported that they use dogs for hunting/herding animals in addition to keeping them as pets or for security reasons. Most of the dogs in the study (77 %) were not confined to the house and roamed freely. The mode of dogs per household that owned dogs was 2 (minimum = 1; 1st quartile = 2; median = 3; 3rd quartile = 4; maximum = 10). The median number of dogs entering the household (born/bought/adopted) during the 2 years prior to the study was 1 (range 0 to 18) and for dogs leaving the household (died/sold/given away) the median was 0.5 (range 0–18). However, by many owners' admission this was an estimate and they could not exactly recall the dog turnover rate at their house over 2 years.

Animal health technician efficacy

Animal health technicians are responsible for vaccinating as many dogs and cats as possible during the annual vaccination campaigns and in Emalahleni municipality there are currently 8 AHTs. The effectiveness of the previous years' vaccination coverages was evaluated with the question: 'Has the dog sampled been vaccinated before?' (Table 1). Of the 203 respondents, 65 % reported 'yes', 2 % were unsure, and 33 % reported 'no'. Of the dogs that had been vaccinated previously, 64 % had been vaccinated once before, 20 % twice, 10 % 3 times and 1 % 4 times (5 % were unsure as to how many times the dog was vaccinated previously). Slightly more than half of the dogs (56 %) had been vaccinated within the 12 months before being sampled, while 5 % of owners were not sure when last their dogs had been vaccinated. The effect of the population density group the owner resided in was not significant with regard to the reported vaccination status of his/her dog ($\chi^2_{(5)} = 2.97, P = 0.71$). Individual AHT efficacies were compared by grouping the responses in terms of the geographical areas for which each AHT was responsible. Nine geographical areas were identified, of which 8 AHT each has

Table 1: Summary of the dogs sampled in the Emalahleni municipality. Dogs were divided into groups based on sex, condition, age and rabies vaccination status. In each group, dogs were further divided into previous rabies vaccination status.

Classification		Previous vaccination status			Total
		Yes	No	Unsure	
Total no. of dogs in study					203
Sex:	Male	86	39	2	127
	Female	45	28	3	76
Condition:	Poor	7	6	1	14
	Thin	21	6	2	29
	Good	98	55	1	154
	Overweight	6	0	2	6
Age group:	6 weeks to 3 months	0	5	0	5
	3–12 months	18	18	1	37
	1–3 years	54	32	1	87
	3–10 years	56	12	3	71
	More than 10 years	3	0	0	3
Previously vaccinated		131	67	5	203

had his/her own area, while a single area was shared between 2 AHTs. The reported vaccination status of dogs was significantly associated with the technician responsible for its vaccination ($\chi^2_{(8)} = 18.08, P = 0.02$). Twenty-three per cent of owners reported that they used their dogs for hunting or herding as opposed to using them solely as pets or for security. Of the dogs used for hunting/herding, 72 % were reported as previously vaccinated while 62 % of dogs which are used purely as pets or for security had been vaccinated before according to their owners. The relationship between a dog's use and its reported vaccination status was not significant, however ($\chi^2_{(1)} = 1.05, P = 0.31$). There were also no significant relationships between a dog's reported vaccination status and its sex ($\chi^2_{(1)} = 1.05, P = 0.31$) or body condition ($\chi^2_{(3)} = 5.37, P = 0.15$). Conversely, a dog's reported vaccination status was significantly associated with its age ($\chi^2_{(4)} = 23.91, P < 0.001$) (Table 1).

Serological description of the dogs sampled

Rabies-neutralising antibody titres were determined in 203 dogs. The geometric mean rabies titre was 0.38 IU/l (1st quartile = 0.15 IU/l (lowest measured point); median = 0.21 IU/l; 3rd quartile = 0.65 IU/l). Of the 203 dogs sampled, 51 % had titres above 0.2 IU/l, while only 32 % had titres of ≥ 0.5 IU/l – an animal with a titre of 0.5 IU/l and above is highly likely to be protected against rabies virus infection in the face of challenge^{3,42}. When considering only those dogs whose owners reported previous vaccination, 43 % had rabies-neutralising antibody titres of 0.5 IU/l and above. On the other hand, when considering only dogs with adequate

neutralising antibody titres, 11 % had not received a previous vaccination according to their owners. There was a highly significant relationship between a dog's reported vaccination status and whether or not it had an adequate rabies-neutralising antibody titre ($\chi^2_{(1)} = 21.3, P < 0.001$). There was no significant relationship between a dog's body condition and whether or not it had a titre of ≥ 0.5 IU/l ($\chi^2_{(3)} = 3.85, P = 0.28$), but there was sufficient evidence to conclude that titres of 0.5 IU/l and above were associated with increasing age ($\chi^2_{(4)} = 26.64, P < 0.001$).

Response to vaccination

Eighty of the 203 dogs originally sampled were retested 30–60 days later to determine the response to vaccination. Of the 80 dogs that were resampled, 83 % had titres that indicated successful seroconversion. Of those dogs that previously had rabies antibody titres below 0.5 IU/l, 64 % seroconverted to titres above 0.5 IU/l (range: 0.5–23.38 IU/l). The geometric mean titre of the retested dogs was 1.44 IU/l (1st quartile = 0.64 IU/l; median = 1.30 IU/l; 3rd quartile = 2.61 IU/l). A paired-samples *t*-test indicated that the resampled dogs had a significantly greater titre 30–60 days after the rabies vaccination (Fig. 4a, $t_{(79)} = 11.3, P < 0.001$) (1st quartile of difference = 0.26 IU/l; median = 0.7 IU/l; 3d quartile of differences = 2.14 IU/l). There seemed to be minor differences in the magnitude of response to vaccination of the different factor groups (Fig. 4b,c,d), although neither the 'age group \times vaccination interaction' ($F_{(2,72)} = 0.65, P = 0.53$) nor the 'condition \times vaccination interaction' ($F_{(2,72)} = 1.52, P = 0.225$) was significant. However, there was a significant main effect of 'age group' ($F_{(2,72)} = 5.74, P =$

0.005. This effect tells us that at least 1 of the age groups differed in their serum titres.

DISCUSSION

The estimation of the canine population size in Emalahleni could be narrowed down further to c. 16 500, based on 2008's vaccination figures in the municipality (unpubl. records) and vaccination coverage of about 56 % a year (see above). The canine population in the study area was remarkably homogeneous in size, breed, and condition. Interestingly, however, only a small proportion of dogs was assigned to the 'poor condition' category (7 %). The proportion of dogs classified as in good condition (76 %) is slightly higher than similar studies in rural southern Africa (56 %³⁰ and 67 %³⁴) and rural Zambia (60 %), but similar to urban dogs in Zambia (74 %)¹¹. It could be argued that a high proportion of these dogs, by virtue of not having similar access to veterinary services or balanced diets as dogs in more developed areas of the country, should be prone to high parasite burdens and/or malnutrition, and thus be overrepresented in the 'poor condition' category. This contrary finding might be ascribed to the subjective nature of the assessment, but may also be ascribed to an inherent resistance to parasites and disease of the local land-race, and the ability to look for additional sources of food in the rural environment¹⁷. It is important to remember, however, that the rural 'village' scenario should be distinguished from that which occurs in townships/shanty towns in South Africa, as the finding about dog condition and breed might not be the same in both²⁷. Furthermore, if a more objective approach is taken through measurement of clinical parameters, counting internal and external parasites and serological screening for various diseases, it may be that a larger proportion of animals will be identified as in need of veterinary attention^{30,34}.

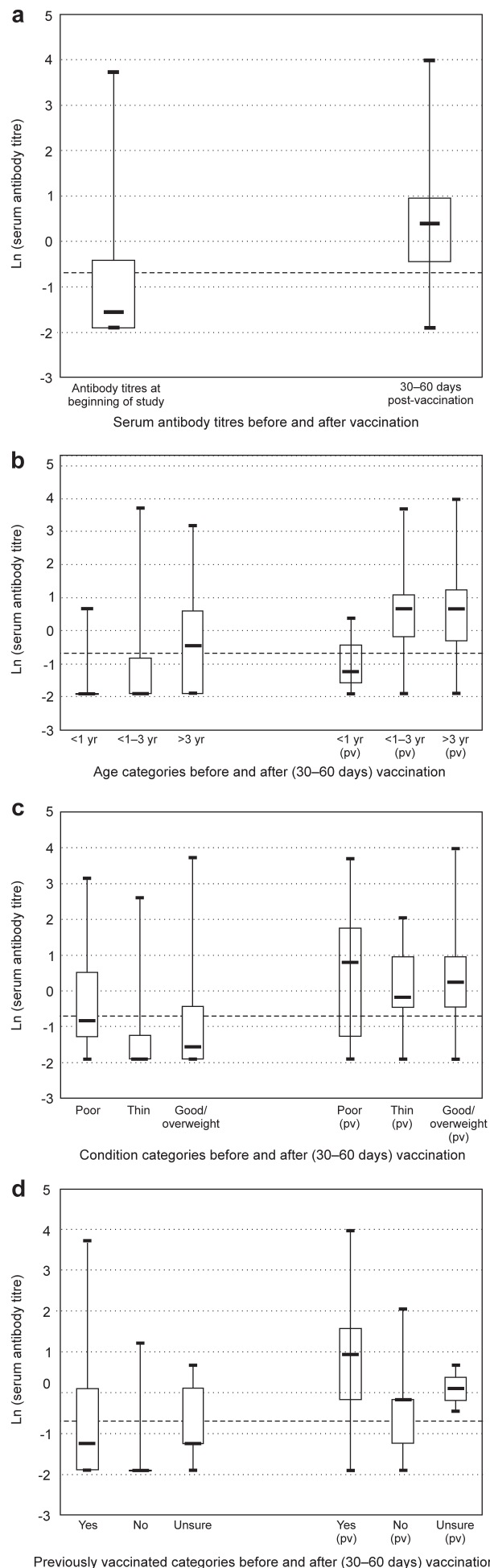
As in similar studies in Africa^{23,34,37}, there were more males than females. This could be ascribed to better care of male animals due to preference for using male animals during hunting²³, although in Emalahleni only 23 % of owners reported using their animals for either hunting or herding. As sterilisation services are either too expensive or difficult to arrange (only 7 (3 %) of the owners reported their dog as sterilised, of which 6 were males), it might also be that owners prefer to keep males rather than females as a form of population control at home.

Slightly over 1/5th (21 %) of the dogs in the study sample were between 6 weeks and 1 year of age, an observation similar

to a study in Tunisia³⁷, where 28 % of the dogs were less than 1 year old (including animals younger than 6 weeks). On the other hand, a study in a district in Kenya found that more than 50 % of the dog population was less than 1 year old²³. The proportion of dogs aged less than 1 year is crucial in rabies eradication as this age group is very often unvaccinated³⁹ or shows a smaller proportion of seroconversion (≥ 0.5 IU/l) than older dogs³⁷. Moreover, only a small proportion (1 %) of dogs were reported to reach 10 years or more. This is similar to a previous study in which the authors found no dog over the age of 8 years in a town in former Bophuthatswana (in present-day North West Province, South Africa)³⁴. This indicates that although many dogs may not seem to be in a poor condition based on this study's condition scoring system, more dogs may be in need of veterinary attention or improved care in order to improve their longevity. Further studies on the causes of dog mortality within these communities are thus needed.

The finding that 77 % of the owned dog population is free-roaming is in accordance with observations in other rural African communities^{17,23}. It is important to note that the feral (or ownerless) dog proportion is often very small in these communities, seldom exceeding 5 %²⁰. Thus eradication of unrestricted or free-roaming animals as an adjunct in the control of rabies is not practical in this kind of community because of the difficulty in distinguishing between owned and feral animals. Even if feral animals were to be eliminated from the population, the majority of owned dogs would still roam freely. Furthermore, it has been found that dog control in the form of quarantine, movement control and dog removal had been counterproductive in some cases in eastern and southern Africa in recent years, provoking public antipathy towards rabies control campaigns³³. Therefore, although free-roaming dogs are an important factor in the epidemiology of rabies outbreaks, we can expect this trend to continue within many African communities. The experience of field work in Emalahleni has shown, however,

Fig. 4: Box and whisker plots of antibody titres (logarithmically transformed) at the beginning of the study ($n = 203$) and 30–60 days post-vaccination ($n = 80$). The dashed line denotes the antibody titre which is considered as successful seroconversion (0.5 IU/l). a, Antibody titres pooled before and after vaccination; b, antibody titres grouped according to age categories; c, antibody titres grouped according to condition; d, antibody titres grouped according to previous vaccination status reported by owners.



that dogs tended not to wander far from their homes unless accompanied by their owners, and were generally available for vaccination when requested, especially when extension work and public education has been thorough.

Although the figures for this study were higher than the 29 % reported in a similar study in Kenya²³ it is of concern that less than 70 % of dogs had been reported as vaccinated previously, as it has been shown that 70 % vaccination coverage would prevent outbreaks of the disease 96.5 % of the time¹⁰. Even more concerning is the proportion of dogs reported by their owners to have been vaccinated within the previous 12 months (56 %). In the Emalahleni municipality a dog's reported vaccination status was significantly associated with the technician responsible for the area in which the dog resided, indicating differing campaign efficacies between AHTs. Furthermore, there was no significant relationship between a dog's vaccination status and the dog's condition, sex or use in hunting or herding. It thus seems that a dog is vaccinated ultimately because an animal health technician was present within his street or village during a campaign, and not whether the dog was important to the owner or not, or if the dog received adequate care. In other words, it is unlikely that a dog owner will travel with his/her dogs to the local state veterinary office for a free rabies vaccination, irrespective of the function or importance of the dog to the owner, or the care level that the dog receives.

The immune mechanisms through which an animal is protected against challenge from rabies virus still needs elucidation, but is likely to involve both cellular and humoral immune responses¹⁴. Nevertheless, the correlation between high levels of neutralising antibody and protection against challenge is high, as has been reported in an extensive review on the subject³. The World Health Organisation has subsequently recommended that 0.5 IU/l be regarded as the minimum neutralising antibody titre for a dog to be considered adequately protected against the disease⁴².

The annual vaccination campaigns in Emalahleni resulted in only 32 % of dogs sampled in this study having adequate protective antibodies, which, interestingly, is the same proportion found in dogs in a similar study in urban Tunisia³⁷. Rabies-neutralising antibody titres tend to wane rapidly after primary vaccination³. Furthermore, the vaccine type and manufacturer can also have an influence on the peak antibody levels reached and the longevity of the antibody response. In

this regard a study that compared 2 different commercial vaccines in laboratory dogs it was found that after a single vaccination the percentages of dogs with titres above 0.5 IU/l after 1 week were 93 % and 100 %, and after 1 month 93 % and 67 %, respectively²⁹. Even though there is large individual variation in the neutralising antibody responses, laboratory dogs do show better antibody responses than pet dogs and stray dogs³. In this study, 43 % of dogs whose owners reported previous vaccination had antibody titres of 0.5 IU/l or above. This response is lower than the percentage in other communities: a study in Spain found it to be 58 %¹²; in urban Bolivia it was also found to be 58 %³⁹; in rural and peri-urban Kenya it was 48 %²³ and in urban Botswana 52 %³⁶. Considering that the owner's responses regarding the vaccination status of their dogs could not be verified, it becomes difficult to evaluate the low proportion of dogs with adequate neutralising antibody titres in Emalahleni, but it may, amongst others, be attributable to: low vaccination coverage by AHTs in the preceding rabies campaign, a rapid drop in antibody titre after being vaccinated in the preceding campaign and/or incorrect handling and administration of vaccines during campaigns. As this study found that the magnitude of seroconversion in Emalahleni was independent of body condition and age they could be eliminated as contributing factors to such a low level of animals protected in the sample. Titres of 0.5 IU/l and above were associated with increasing age, however, which could indicate that the older dogs received more than 1 vaccination and therefore maintained antibody levels above the threshold level longer than younger dogs. As the extent to which maternally derived rabies antibodies interferes with vaccination in pups is controversial, we concur with other authors^{23,37} that vaccination of dogs younger than 3 months of age should be considered to increase the proportion of dogs immunised and to increase the proportion of dogs that will receive 2nd and 3rd booster vaccinations. Importantly, however, owner education should be thorough so as to prevent owners from associating the usually high neonatal and juvenile morbidity and mortality rate in these rural settings²³ with rabies vaccination.

Similar to a previous finding²³, 11 % of dogs had rabies-neutralising antibody titres of ≥ 0.5 IU/l despite owners reporting no previous vaccination. As all these dogs were older than 1 year, this cannot be ascribed to maternally derived antibodies. This finding could thus either be

due to owner ignorance of the vaccination status of his/her dog, a possible false positive reaction of the FAVN test, or even exposure to lyssaviruses in nature. Indeed, there are reported cases of rabies antibody titres being detected in previously unvaccinated but clinically healthy animals where there was a long incubation period after infection, where animals were clinically unaffected carriers of rabies virus or had recovered from clinical infection^{14,13-16}. If clinically healthy dogs were able to maintain and spread the infection, this might play a significant role in the epidemiology of rabies in the area.

Comparison of antibody titres 30–60 days post-vaccination in this study ($n = 80$) showed that the majority (83 %) of dogs seroconverted adequately. In a similar study in urban Tunisia³⁷, seroconversion varied between 72 % 1 month after vaccination to 36 % 12 months after vaccination. On the other hand, the proportion of dogs that seroconverted to levels of ≥ 0.5 IU/l after vaccination in this study is slightly less than the 85 % and >90 % of dogs responding adequately to 1 or more than 1 rabies vaccination, respectively, in a study of sera from pet dogs vaccinated in veterinary practices in France⁸. This could indicate that the dogs in Emalahleni do not seroconvert as efficiently as dogs that have better access to veterinary care. The geometric mean titre 30–60 days post-vaccination within this study (1.44 IU/l) compares favourably with other studies^{29,37,39}. However, as the number of vaccinations a dog had received before the study could not be verified, further interpretation of this result was not attempted.

Vaccination campaigns tend to get coverage in the major and minor media in the area, and considerable effort is made to design and distribute information about the disease in the province. Furthermore, the vaccination campaign is launched at major community meetings across the province every year. According to this evaluation of technician efficacy a breakdown in the campaign occurs afterwards, at field level, with the delivery of vaccine within communities. This could be attributed to difficulty in supervising the process and lack of motivation of AHTs to complete the campaign in very often demanding environments, as well as a relatively young dog population. Ideally dogs in the area should be vaccinated twice a year, as this could also theoretically lower the proportion of dogs that need to be vaccinated in order to block the spread of disease²². Unfortunately, however, the Eastern Cape rabies eradication campaign competes with other important disease control pro-

grammes like anthrax, sheep scab, bovine brucellosis and tuberculosis, Newcastle disease and other emergency disease outbreaks (for example the recent classical swine fever and Rift Valley fever outbreaks). Consequently, twice a year vaccination would be impractical and unsustainable with the current number of AHTs available. There is thus an urgent need to find a balance between the 'gold standard' in rabies control and what is repeatedly practical in Emalahleni.

In response to this specific challenge, the 2009 rabies campaign in Emalahleni was approached by pooling AHTs in an area rather than scattering their manpower throughout the district. All the AHTs were thus concentrated in a village for a shorter time than would be the case with individual efforts before moving on to the next village/area. This approach had several advantages: greater visibility of the campaign, improved supervision during the campaign and an increase in proportion of animals vaccinated to 72 % (based on our canine population estimate). Disadvantages included greater transport costs (as AHTs now had to travel to areas other than those for which they had been responsible), and a longer period to conclude the campaign in the municipality (4 months instead of 3 months). Although certain models do predict 70 % vaccination coverage to be adequate in blocking the spread of disease^{10,22}, they often assume that a single injection will confer lifelong immunity, and that each vaccinated animal has seroconverted successfully. As we have seen in this study, 83 % of animals vaccinated during the study seroconverted successfully after 30–60 days. Thus, models should also be developed to test whether a 70 % vaccinated proportion would be adequate in blocking the spread of infection given the successful seroconversion proportion above, also taking into account the number of dogs receiving 2 or more vaccinations.

CONCLUSION

Rabies is an emerging public health problem in the Eastern Cape Province, notwithstanding the fact that every year a vaccination campaign is launched to vaccinate as many dogs and cats as possible. This study highlights some of the areas where this worrying trend might originate. Problems identified during previous dog ecology studies in Africa were confirmed in Emalahleni, and we can assume that these trends will continue owing to various socio-economic factors as listed previously⁴⁰. However, in South Africa, we are fortunate to have adequate government funding in terms of rabies

disease surveillance and provision of vaccines. It is thus important that during planning of pro-active rabies vaccination campaigns, work and supervision at field level is not neglected. This can be achieved by active involvement of supervisors during the campaign who will experience first-hand the difficulties met during vaccine administration at ground level, and attending to staff shortages of AHTs and state veterinarians.

This study's preliminary findings prompt similar investigation into other rural and urban municipalities in the Eastern Cape and in the rest of South Africa, as findings about dog ecology and vaccination efficacy may differ regionally. Importantly, more studies should focus on the evaluation of different rabies control and vaccination strategies, and why current methods are failing in South Africa. Rabies vaccination strategies in the face of an outbreak also deserve special attention.

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REFERENCES

1. Aghomo H O, Oduye O O, Tomori O, Ibe M 1989 Isolation of rabies virus from clinically healthy and previously unvaccinated dogs. *Bulletin of Animal Health and Production in Africa* 37: 131–135
2. Akol G, Lwanga-Iga I, Amaral L A, Kroll-Lwanga-Iga S 1993 Rabies control in Transkei South Africa. *Proceedings of the Southern and Eastern African Rabies Group International Symposium, Pietermaritzburg, South Africa*, 29–30 April 1993: 81–91
3. Aubert T F A 1992 Practical significance of rabies antibodies in cats and dogs. *Revue scientifique et technique, Office International des Épidémiologies* 11: 735–760
4. Bigler W J, McLean R G, Trevino H A 1973 Epizootologic aspects of raccoon rabies in Florida. *American Journal of Epidemiology* 98: 326–335
5. Blumberg L, Leman P, Paweska J, Weyer J 2010 Human rabies in South Africa, 2009. *Communicable Diseases Surveillance Bulletin of the National Institute of Communicable Diseases* 8: 16–17
6. Brown K In press. Rabid epidemiologies: the emergence and resurgence of rabies in twentieth century South Africa. *Journal of the History of Biology*. DOI 10.1007/s10739-010-9241-9

7. Cliquet F, Aubert M, Sagné L 1998 Development of a fluorescent antibody virus neutralisation test (FAVN test) for the quantitation of rabies-neutralising antibody. *Journal of Immunological Methods* 212: 79–87
8. Cliquet F, Verdier Y, Sagné L, Aubert M, Schereffer J L, Selve M, Wasniewski M, Servat A 2003 Neutralising antibody titration in 25 000 sera of dogs and cats vaccinated against rabies in France, in the framework of the new regulations that offer an alternative to quarantine. *Revue scientifique et technique, Office International des Épidémiologies* 22: 857–866
9. Cohen C, Sartorius B, Sabeta C, Zulu G, Paweska J, Mogoswane M, Sutton C, Nel L H, Swanepoel R, Leman P A, Grobelaar A A, Dyason E, Blumberg L 2007 Epidemiology and molecular virus characterization of reemerging rabies, South Africa. *Emerging Infectious Diseases* 13: 1879–1886
10. Coleman P G, Dye C 1996 Immunization coverage required to prevent outbreaks of dog rabies. *Vaccine* 14: 185–186
11. De Balogh K K, Wandeler A I, Meslin F X 1993 A dog ecology study in an urban and a semi-rural area of Zambia. *Onderstepoort Journal of Veterinary Research* 60: 437–443
12. Delgado S, Cármenes P 1997 Immune response following a vaccination campaign against rabies in dogs from northwestern Spain. *Preventive Veterinary Medicine* 31: 267–261
13. Fekadu M 1991 Latency and aborted rabies. In Baer G M (eds) *The natural history of rabies*. CRC Press, Boca Raton, Florida: 191–198
14. Fekadu M 1993 Canine rabies. *Onderstepoort Journal of Veterinary Research* 60: 421–427
15. Fekadu M 1988 Pathogenesis of rabies virus infection in dogs. *Reviews of Infectious Diseases* 10 Suppl 4: S678–683
16. Fekadu M 1972 Atypical rabies in dogs in Ethiopia. *Ethiopian Medical Journal* 10: 79–86
17. Gallant J 2002 *The story of the African dog*. University of Natal Press, Pietermaritzburg
18. Gummow B, Roefs Y A A, de Klerk G 2010 Rabies in South Africa between 1993 and 2005 – what has been achieved? *Journal of the South African Veterinary Association* 81: 16–21
19. International Committee on Taxonomy of Viruses 2010 ICTV master species list 2009 version 5. Online at: http://talk.ictvonline.org/files/ictv_documents/m/msl/1231.aspx (accessed 5 August 2010)
20. Kaare M, Lembo T, Hampson K, Ernest E, Estes A, Mentzel C, Cleaveland S 2009 Rabies control in rural Africa: evaluating strategies for effective domestic dog vaccination. *Vaccine* 27: 152–160
21. Kärber G 1931 Beitrag zur kollektiven Behandlung pharmakologischer Reihenversuche. *Naunyn-Schmiedeberg's Archiv für Experimentelle Pathologie und Pharmakologie* 162: 480–483
22. Kitale P M, McDermott J J, Coleman P G, Dye C 2002 Comparison of vaccination strategies for the control of dog rabies in Machakos District, Kenya. *Epidemiology and Infection* 129: 215–222
23. Kitale P M, McDermott J J, Kyule M, Gathuma J, Perry B, Wandeler A 2001 Dog ecology and demography information to support the planning of rabies control in Machakos District, Kenya. *Acta Tropica* 78: 217–230
24. Knobel D L, Cleaveland S, Coleman P G, Fèvre E M, Meltzer M I, Miranda M E G, Shaw A, Zinsstag J, Meslin F 2005

- Re-evaluating the burden of rabies in Africa and Asia. *Bulletin of the World Health Organization* 83: 360–368
25. Mansvelt P R 1962 Rabies in South Africa. Field control of the disease. *Journal of the South African Veterinary Medical Association* 33: 313–319
 26. Mansvelt P R 1956 Rabies in the northern Transvaal (1950) outbreak. *Journal of the South African Veterinary Medical Association* 27: 167–178
 27. McCrindle C M E, Gallant J, Cornelius S T, Schoeman H S 1999 Changing roles of dogs in urban African society: A south African perspective. *Anthrozoos* 12: 157–161
 28. Meltzer M I, Rupprecht C E 1998 A review of the economics of the prevention and control of rabies: Part 2: Rabies in dogs, livestock and wildlife. *PharmacoEconomics* 14: 481–498
 29. Minke J M, Bouvet J, Cliquet F, Wasniewski M, Guiot A L, Lemaitre L, Cariou C, Cozette V, Vergne L, Guigal P M 2009 Comparison of antibody responses after vaccination with two inactivated rabies vaccines. *Veterinary Microbiology* 133: 283–286
 30. Minnaar W N, Krecek R C 2001 Veterinary needs of dogs in two resource limited communities in the Gauteng and North West Provinces in South Africa. *Journal of the South African Veterinary Association* 72: 209–213
 31. Nel L H, Markotter W 2007 Lyssaviruses. *Critical Reviews in Microbiology* 33: 301–324
 32. Nel L H, Sabeta C T, Von Teichman B, Jaftha J B, Rupprecht C E, Bingham J 2005 Monogoose rabies in southern Africa: A re-evaluation based on molecular epidemiology. *Virus Research* 109: 165–173
 33. Perry B D 1993 Dog ecology in eastern and southern Africa: implications for rabies control. *Onderstepoort Journal of Veterinary Research* 60: 429–436
 34. Rautenbach G H, Boomker J, De Villiers I L 1991 A descriptive study of the canine population in a rural town in Southern Africa. *Journal of the South African Veterinary Association* 62: 158–162
 35. Sabeta C T, Bingham J, Nel L H 2003 Molecular epidemiology of canid rabies in Zimbabwe and South Africa. *Virus Research* 91: 203–211
 36. Sebunya T K, Ndabambi N, Mpuchane S 2007 A serosurvey of rabies antibodies in dogs in Gaborone, Botswana. *Journal of Animal and Veterinary Advances* 6: 549–552
 37. Seghaier C, Cliquet F, Hammami S, Aouina T, Tlatli A, Aubert M 1999 Rabies mass vaccination campaigns in Tunisia: Are vaccinated dogs correctly immunized? *American Journal of Tropical Medicine and Hygiene* 61: 879–884
 38. Spearman C 1908 The method of ‘right and wrong’ cases (‘constant stimuli’) without Gauss’s formulae. *British Journal of Psychology* 227–242
 39. Suzuki K, González E T, Ascarrunz G, Loza A, Pérez M, Ruiz G, Rojas L, Mancilla K, Pereira J A C, Guzman J A, Pecoraro M R 2008 Antibody response to an anti-rabies vaccine in a dog population under field conditions in Bolivia. *Zoonoses and Public Health* 55: 414–420
 40. Swanepoel R 2004 Rabies. In Coetzer J A W, Tustin R C (eds) *Infectious diseases of livestock* (2nd edn) Vol. 2. Oxford University Press Southern Africa, Cape Town: 1123–1182
 41. Thrusfield M 2007 Diagnostic testing. In Thrusfield M (ed.) *Veterinary epidemiology* (3rd edn). Blackwell Science, Oxford: 307–308
 42. World Health Organization 2005 WHO expert consultation on rabies (2004: Geneva, Switzerland). *WHO Technical Report Series* 931: viii + 88