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Rabies in wild and domestic carnivores of Africa: epidemiological and historical associations determined by limited sequence analysis

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ABSTRACT

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Virus isolates from three important reservoirs for rabies in Africa (domestic dogs, jackals and yellow mongooses) were compared by their reaction with a panel of monoclonal antibodies directed to the nucleocapsid protein and by the nucleotide sequence of a 200 base pair segment of the nucleocapsid gene. Although antigenically dissimilar, the variants commonly transmitted in dogs and jackals were very closely related by genetic analysis. Phylogenetic analysis and historical accounts support a common lineage for these variants in both past and present reservoirs for rabies in Europe. Two additional variants, distinct from the dog or jackal variant, were found in yellow mongoose samples and nucleotide sequence from these animals showed more divergence than any other group of samples. These variants and a third variant for which no host species could be identified, were shown to form two additional genetic groups only distantly related to each other. These three variants and a previously identified variant in Nigeria may be indigenous to African carnivores.

INTRODUCTION

Case surveillance and antigenic typing of virus isolates portray rabies as a disease maintained in nature as several distinct variants, each circulating in a particular animal reservoir or geographic area. These distinctions are useful markers of epidemiologically separate events, but analyses of this type tell us little of how these different variants arose or how a particular variant came to be associated with a particular ecological niche. Recent developments in molecular biology now permit an investigation of the origin of different outbreaks. Distinctive patterns of nucleotide substitution can be identified by direct sequencing of amplified genomic material from the polymerase chain reaction (PCR), and computer algorithms, originally developed for phylogenetic studies, can be used to determine epidemiological associations, and in some instances, historic relationships between outbreaks.

In this report, we present an antigenic and genetic analysis of rabies samples from South Africa in the context of rabies variants found in other areas of the world where rabies is enzootic in domestic and wild carnivores.

MATERIALS AND METHODS

The panel of nucleocapsid (N) protein specific monoclonal antibodies (Mab-N) and the procedure used

for analyzing virus samples is as described in Smith, Yager, Bigler & Hartwig (1990). With the exception of Mab-N T, all antibodies were prepared at the Centers for Disease Control from one spleen fusion from a mouse immunized with ERA rabies virus. Mab-N T was prepared by Dr L. Schneider, Tubingen using a street rabies virus as immunogen.

The procedures for RNA extraction from virus samples, reverse transcription of the RNA, amplification of cDNA by PCR, and direct sequencing of the PCR product is as described in Smith, Orciari, Yager, Seidel & Warner (1992). Reverse transcription of the nucleoprotein (N) gene was accomplished by extension of an oligonucleotide primer derived from sequence including the initial methionine codon of the N protein. The primer pair for PCR included this oligonucleotide and an oligonucleotide derived from sequence including the initial methionine codon of the phosphoprotein NS. The PCR product excised from an agarose gel was sequenced directly using the NS primer. Sequence including 200 bp of the carboxy terminus of the N protein was used for comparative analysis of all virus samples.

Initial analysis and alignment of the multiple nucleic and amino acid sequences obtained from this region of the N protein were done using computer algorithms in the PileUp and Distances Programs in the Sequence Analysis Software Package, Version 7 (GCG, Madison, WI). Phylogenetic analysis of samples was performed by the method of maximum likelihood analysis (Phylip, Version 3.4, J. Felsenstein, University of Wisconsin). Designation of genetic groups follows the format of previous studies (Fig. 1).

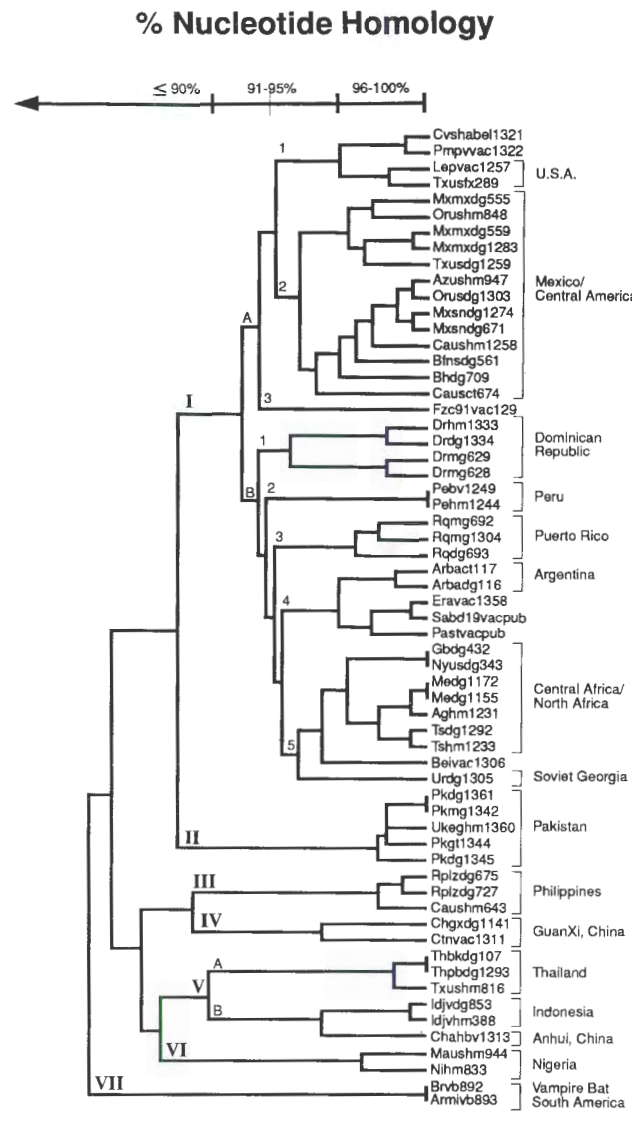

FIG. 1 Analysis and alignment of the multiple nucleic acid sequences obtained using computer algorithms in the PileUp and Distances Programs of the GCG genetic analysis software. The horizontal branch lengths are proportional to the similarity of sequences within and between clusters. Vertical lines are for clarity of presentation only. The numerical values for sequence divergence between pairs of samples were used to arbitrarily divide the sample clusters in the dendrogram into three categories. The first category consists of clusters of the most closely related samples. Members of these clusters differ from one another by < 5 % and, in general, were field samples collected from the same outbreak area. The second major category consists of joined clusters that differ from each other by < 10 %. Clusters of samples joined at this level tend to be related by historical, political, and cultural connections as well as by geographic proximity. Each node within this category was given an alpha-numeric designation so that the subgroups of samples could be more easily identified in the text. The third category consists of joined subgroups that differ from each other by 10 % or more. Subgroups of samples joined at this level were considered to be only distantly related. Each node in this category was designated by a Roman numeral and this designation was used to show the geographic origin of these samples in Fig. 5

RESULTS AND DISCUSSION

Fig. 2 depicts the results of antigenic typing of 95 rabies virus samples collected in 13 African countries. Five reaction patterns were identified and associated with the major vector species for rabies in Africa (domestic dog, *Canis familiaris*; black-backed jackal, *Canis mesomelas*; and yellow mongoose, *Cynictis penicillata*).

Reaction patterns 1 and 2 (dog and jackal rabies)

With one exception (a dog sample from Zimbabwe), all samples from dogs or other domestic animals and humans bitten by rabid dogs could be identified by reaction patterns 1 or 2 (Fig. 2).

Isolates identified by reaction pattern 1 originated in areas of uncontrolled or poorly controlled rabies in domestic dogs and were widely distributed in Africa (Fig. 3). This variant was also identified in all eight African human rabies cases in this study (two from

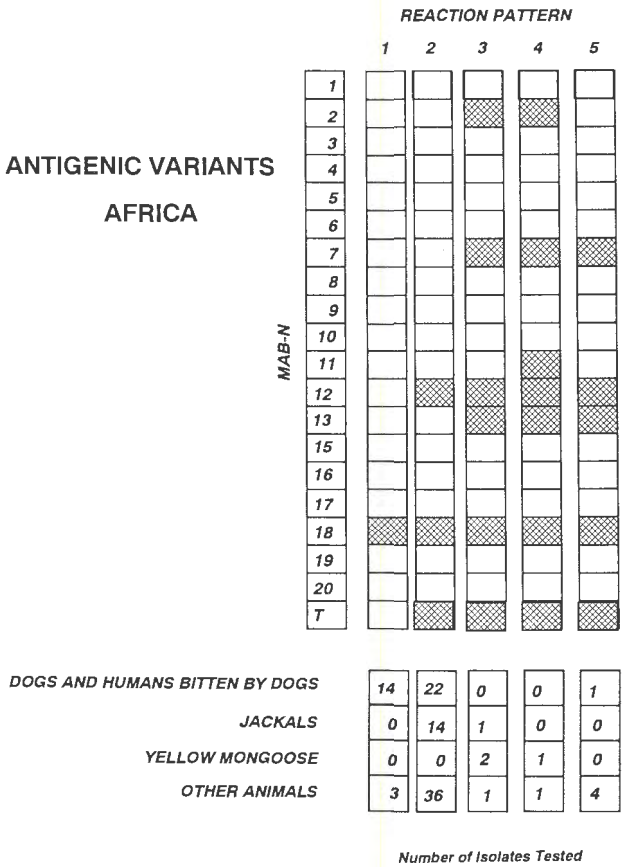

FIG. 2 Reaction pattern of 95 African rabies samples in an indirect immunofluorescence assay with a panel of 20 MAb-N. Open square, positive reaction; filled squares, negative reaction

FIG. 3 Distribution in Africa of a rabies variant identified by MAb reaction pattern 1 in Fig. 2; I, samples identified by genetic typing as group I; VI, samples identified by genetic typing as group VI

Nigeria, one from Kenya, one from Tunisia, one from Egypt, and three from Zimbabwe). In previous studies (Smith 1989), this antigenic variant was also found in dog rabies areas of Latin America, Asia, and eastern Europe. No isolate of this variant was made from a wild animal sample in Africa.

Alignment and comparison of the nt sequence of 11 samples from this group of antigenically identical isolates displays these samples as two separate genetic groups (Fig. 4). One group consisted of the two samples associated with dog rabies in Nigeria. In previous studies (Smith *et al.* 1992), these samples could not be related to dog rabies samples anywhere in the world and were designated as genetic group VI (Fig. 1). The remaining samples identified by reaction pattern 1 were included in a separate group and differed from the Nigerian samples by 10–16% (mean = 13%) (Fig. 4). Based on their approximately 94% nt homology with the Pasteur strain of rabies virus, isolates of this genetic variant were designated as genetic group I (Fig. 1). Following the format of Fig. 1, samples from Kenya and Tanzania were designated as a separate subgroup within genetic group I.

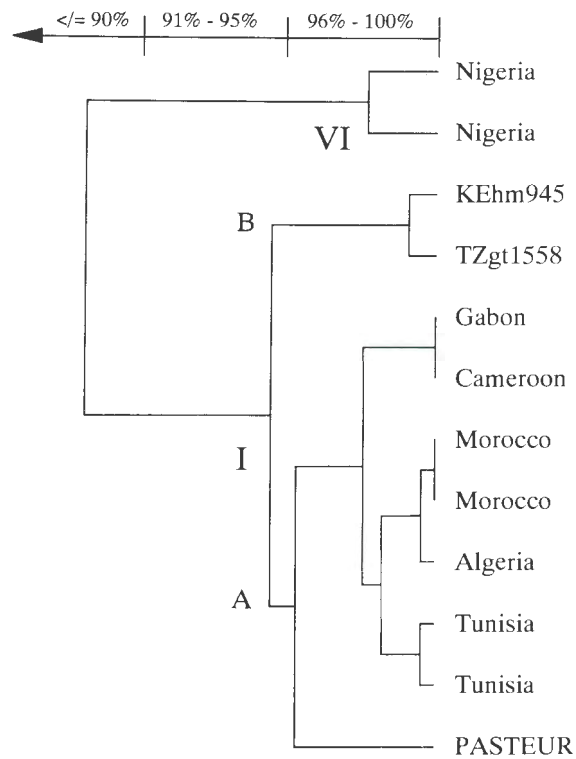

FIG. 4 Graphical representation of genetic relationships among 11 dog rabies associated rabies isolates identified by reaction pattern 1 in Fig. 2. KEhm945, isolate from American peace corp volunteer bitten by dog in Kenya; TZgt1558, isolate from goat bitten by dog in Zanzibar, Tanzania. Identification of all other samples is as in Smith *et al.* 1992. The Pasteur strain of rabies virus is included for comparison

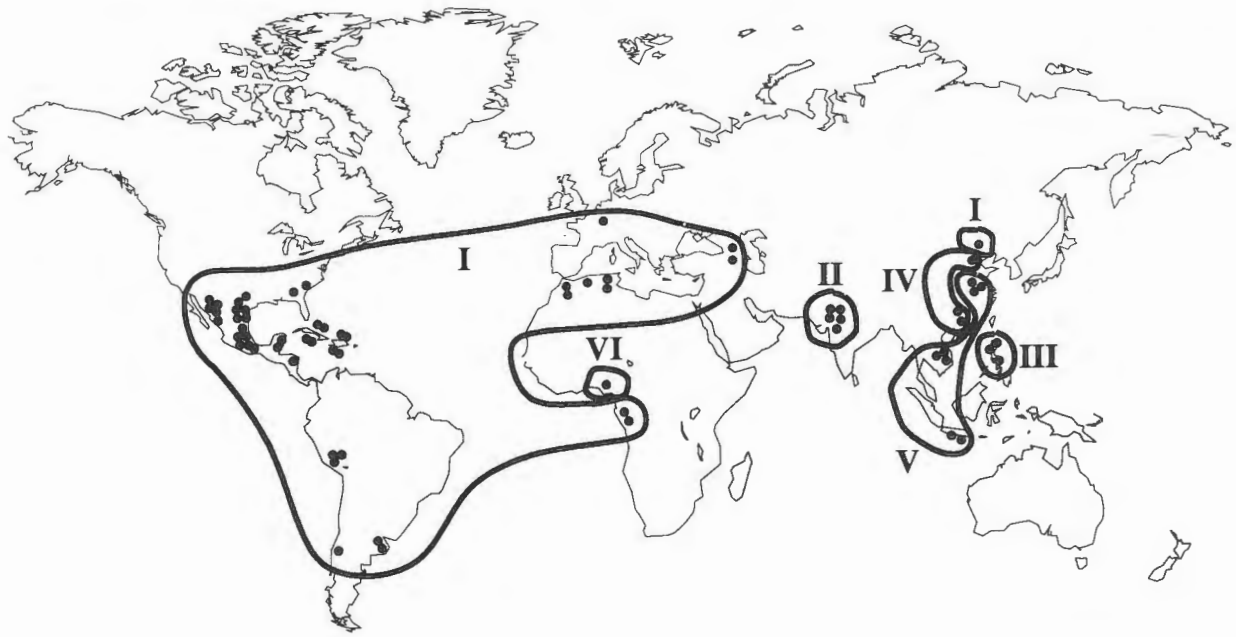

FIG. 5 Geographic distribution of unique genetic groups of rabies virus differing from each other by 10% or more as presented in the dendrogram in Fig. 1

Isolates of genetic group I rabies virus are common in areas of the world with uncontrolled or poorly controlled dog rabies (Fig. 5). These countries share a heritage of European colonization that often included outbreaks of rabies in dogs owned by European settlers (Smith *et al*. 1992), suggesting that the broad distribution of genetic group I viruses is the result of transport of rabies from Europe to other continents. The dog rabies associated variant in genetic group VI (Nigeria), did not share this lineage and may represent a virus indigenous to Africa.

A variant characterized by reaction pattern 2 was found in 14 of 15 samples from jackals (Fig. 2) and was the predominant variant found in areas of southern Africa where the jackal is thought to be the major vector for rabies (Fig. 6). This variant was also common in samples from dogs and other domestic animals in southern Africa but was found in only six samples from wild species other than the black-backed jackal (one water mongoose, *Atilax paludinosus*; two kudu, *Tragelaphus strepsiceros*; two bat-eared foxes, *Otocyon megalotis*; one honey badger, *Mellivora capensis*).

The nt sequences of rabies samples displaying reaction pattern 2 were very closely related (Fig. 7). Samples from jackals in South Africa differed by < 1% from samples from areas of Namibia and Zimbabwe enzootic for rabies in jackals. Samples from the bat-eared fox and water mongoose showed only a slightly greater divergence (2,5%) from each other

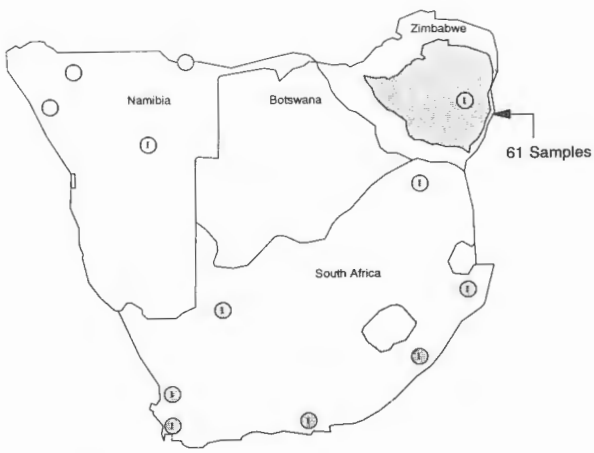

FIG. 6 Distribution in southern Africa of a rabies variant identified by reaction pattern 2 in Fig. 2; I, samples identified by genetic typing as group I

and from the jackal samples. As a group, the jackal rabies associated samples were included within the large genetic group I (Fig. 8) which also included the dog rabies associated variant identified by antigenic typing as reaction pattern 1. The dog rabies samples most closely related to the jackal samples were found in the subgroup of isolates from Kenya and Tanzania (97% homology).

As stated earlier, genetic group I includes not only dog rabies virus from Africa but also isolates collected

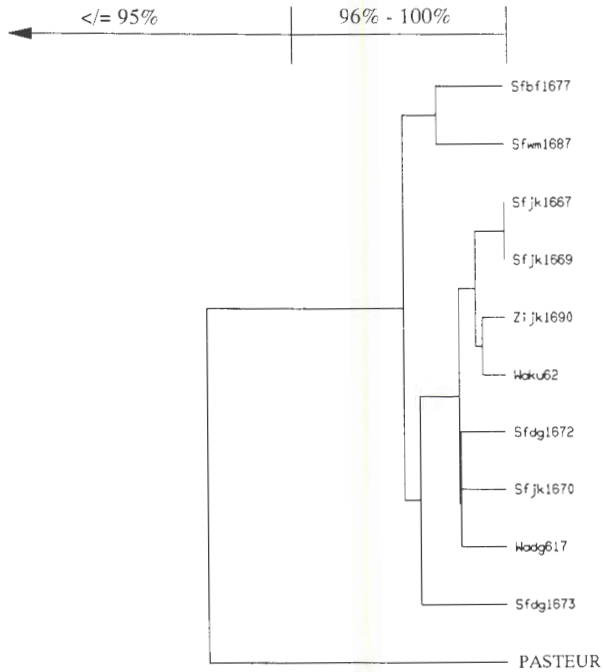

FIG. 7 Graphical representation of genetic relationships among rabies isolates identified by reaction pattern 2 in Fig. 2. Samples are identified by the country where the sample was collected (Sf, South Africa; Zi, Zimbabwe; Sf, Namibia), the animal origin (bf, bat-eared fox; wm, water mongoose; jk, jackal; ku, kudu; dg, dog), and a numerical designation for that sample in the rabies virus repository at the Centers for Disease Control

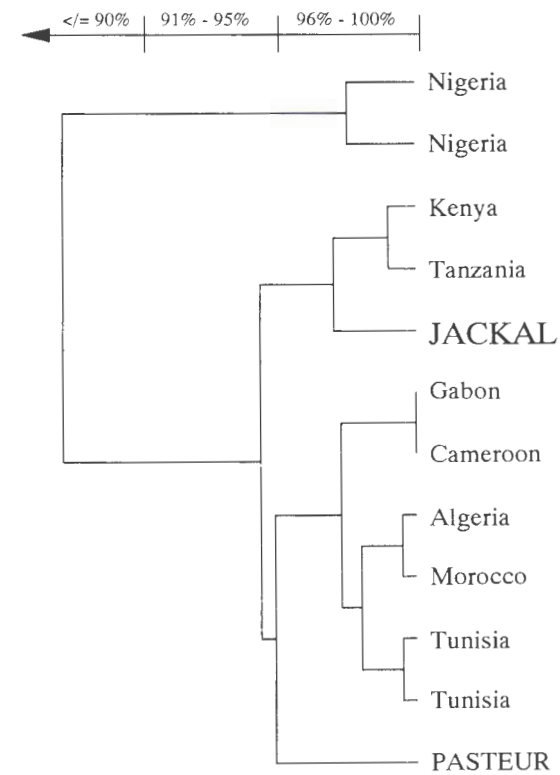

FIG. 8 Graphical representation of genetic relationships among rabies isolates from dogs (reaction pattern 1 in Fig. 2.) and jackals (reaction pattern 2 in Fig.2). Jackal sequence = Sfjk1670

on other continents in areas enzootic for rabies in dogs and economic or cultural ties to Europe. Phylogenetic analysis of sequence data from this geographically dispersed but genetically related group of viruses (Fig. 9) suggest two explanations for the placement of African jackal samples in this group.

1. The legacy in southern Africa of European colonization which included the introduction of dogs infected with rabies in Europe (e.g. Hutcheon 1894) may have been indirectly responsible for jackal rabies. Jackals would have been infected initially through contact with rabid dogs with subsequent independent maintenance of the variant within the jackal population. Similar historical accounts exist for the introduction of dog rabies with Europeans in the Americas (cited in Smith & Seidel 1993) with later transmission to and virus maintenance in wild canids such as foxes (Johnson 1945) and coyotes (Krebs, Holman, Hines, Strine, Mandel & Childs 1992). Additionally, transmission of rabies from dogs to foxes has been suggested as the source of the rabies outbreak which spread throughout western and central Europe after World War II (Blancou, Aubert & Artois 1991). The phylogenetic tree of these samples (Fig. 9) displays samples from jackals on a branch which includes the Pasteur rabies strain and samples from dog rabies in Africa and the Caribbean.
2. The jackal rabies variant is indigenous to Africa and related to the global reservoir of dog rabies only indirectly. In this explanation, the progenitor virus for genetic group I arose in wild canids in Europe or Africa, producing a variant with a northern range in a reservoir maintained by the red fox in Europe, a mid-range in desert foxes of the Arabian peninsula and a southern range in jackals in South Africa (Fig. 9). Sporadic transmission of this wild canid virus to dogs would have always occurred but a reservoir in domestic dogs would not have arisen until the last few centuries as dogs became popular as pets in Europe and the dog population had enlarged sufficiently to support enzootic transmission. Transport of dogs during European exploration and colonial expansion would have introduced the variant into new areas or reintroduced the variant back into areas where it had disappeared from wild canid populations. Isolates of the jackal variant differ at several nt positions from dog rabies isolates in genetic group I and form a separate subgroup of samples with samples from Kenya and Tanzania (Fig. 8 and Fig. 10). Although the distinctive nt

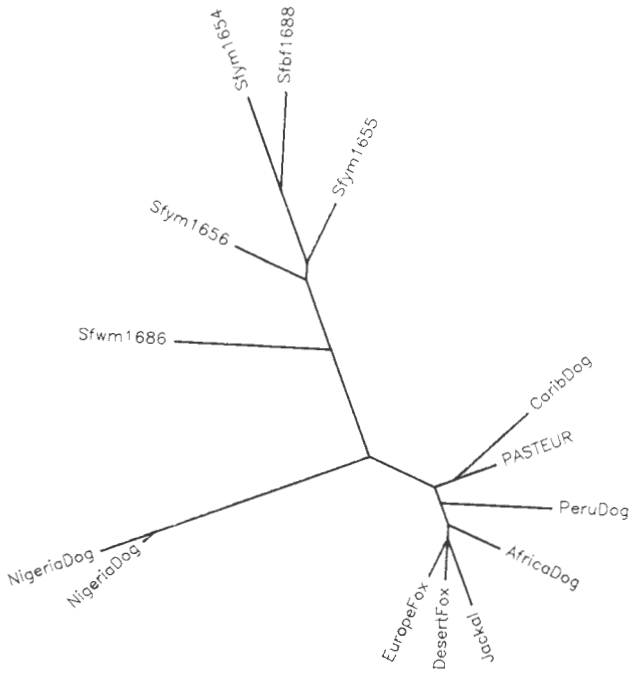

FIG. 9 Phylogenetic tree of rabies samples from genetic groups I, VI, VIII and IX analyzed by the method of maximum likelihood. Branch length is proportional to genetic distance between samples

substitutions characterizing this sub-group could have arisen from an independent introduction of dog rabies from a different area of Europe into this region of Africa, Kenyan natives recognized a rabies-like disease in jackals and dogs which predated the advent of Europeans (Hudson 1944) lending support to the argument for jackal rabies as indigenous to Africa.

Reconstruction of these events should be possible as more sequence data accumulate and historical accounts are reinterpreted to account for phylogenetic observations. Regardless of the original direction in which rabies moved into southern Africa, however, the public health implication of such closely related variants in jackals and dogs remains the same. Permanent elimination of rabies in domestic dogs in this area will require some measure of rabies control in wild canids.

Reaction patterns 3, 4, and 5 (mongoose rabies?)

Rabies virus samples from areas of South Africa where the yellow mongoose is the most important wildlife vector for rabies (Fig. 11) were heterogeneous in their reaction with the panel of MAb-N (reactions patterns 3, 4, and 5 (Fig. 2)) and formed two genetic groups differing from each other by 11 % and four sub-groups differing from each other by 6–8 % (Fig. 12). Samples identified by reaction pattern 4 (ground squirrel 1678 and wild cat 1679) were found in genetic group VIII. Samples identified by reaction pattern 3 were in two different subgroups of genetic group IX (group IXA and IXB) as were the two samples identified by reaction pattern 5 (genetic group IXC and IXD). Three samples from Zimbabwe not available for genetic analysis (dog, cow, honey badger) were also identified by reaction pattern 5.

Although the two genetic groups were clearly more related to each other than to any other group of African samples, the groups and even the sub-groups appear as several lineages (Fig. 9 and Fig. 12). With the exception of isolates of genetic group IXA which were made within a relatively small geographic area, there was no correlation between genetic relatedness and sample collection site (Fig. 11), nor was there a correlation between a particular animal species and variant. (For example, the four samples of rabies collected from yellow mongooses differed by as much as 8 % (Fig. 12).

The amount of genetic diversity within this group of samples and within samples from the yellow mongoose in particular was surprising in light of the small geographic area from which the samples originated. In previous genetic studies, nt sequence from the carboxy terminus of the N protein has shown a high degree of homology in samples from the same outbreak area or from related outbreaks and remained high in samples collected from a given outbreak over a long period of time, in samples from non-reservoir animals only incidentally involved in the outbreak, and in samples variously passaged in cell culture or in laboratory animals (Smith & Seidel 1993).

In light of these data, the number of genetically different variants identified in the small sample set of mongoose and presumably mongoose rabies associated samples in a relatively small geographic area indicate multiple independent cycles of virus transmission, several distinct reservoirs of disease, and an epidemiology much more complicated than that surmised from similar data for rabies in jackals or dogs in South Africa.

Sequence divergence of this magnitude may have arisen through several mechanisms. Discontinuous mongoose populations arising through topographic barriers to animal movement (mountain, rivers, etc.) or habitat isolation could give rise to different variants while maintaining the appearance of a homogenous reservoir for rabies. Alternately, the mongoose could simply be the indicator species for reservoirs of rabies in other, more reclusive species.

This latter hypothesis has important implications for the design of control measures. Immunization campaigns targeted solely on mongooses would reduce the number of cases reported to the public health community simply because mongoose cases are a large percentage of the total wildlife rabies cases

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Genetic group          10
Non-rabies     Mokola 1      --a---ag-a -g-----t-- taca-a-ct- --ggg-g-t- -agtg--a-c ct----a--t -----c--t- -g--ag---- a--t--g--a ----t-----
               Mokola 5      --a-----ga -g-----t-- taca-a-ct- --gga-g-t- gagta--g-c tt-g--c--t -----c--g- ----ag---- a--t--g--- ----t-----
               Duvenhage     --------g- -g-----c-- t--a-aa--a --ggcc--t- ---t---g-c t---t----t ---------- ----g----- c--------a ----tt----

VIII           Sfwc1679      ---------- --nnnnng-- t-----t--c -----a---- --------a- --t------- ---------- -------t-- ------c--- -----t----
               Sfwm1686      ---------- -------g-- t-----t--- --gt------ --------a- --t------- ---------- -------t-- ------a--- ----------

IX             Sfym1653      -------g-- --nnnn-g-- ------t--n n--t-a---- --g-c---ac aat------- ---------- ---------- c-----c--- ----------
               Sfym1655      ---------- -------g-- ------t--- -----a---- --------ac -at-t----- ---------- -------t-- c--t--c--- ----------
               Sfym1654      --------g- -------g-- ------t--- -----a---- ---a----ac aat------- ---------- ---------- ------c--- --t-------
               Sfbf1688      --------g- -------g-- ------t--- -----a---- ---ac----c -at------- --c------- ---------- c--t--nn-- ----------

VI             Nigeria Dog   --a------- -c-----g-- t-----a--- ---t------ -------t-- ---------- --c--c---- -c-------- ---t------ --t-----t-
               Nigeria Dog   --a------- -c-----a-- t---------- ---t----g- -------t-- ---------- -----c---- -c-------- ---t------ --t-----t-

I              Sfjk1670      ---------- -------a-- ---------- ---------- ---------- ----t----- ---------- ---------- ---a------ ----------
               Waku62        ---------- -------a-- ---------- ---------- ---------- ----t----- ---------- ---------- ---------- ----------
               Zijk1690      ---------- -------g-- ---------- ---------- -------nn- ----t----- ---------- ---------- ---------- ----------
               Kehm945       ---------- -------a-- ----a--s-- ---------- ---------- ----t----- ---------- ---------- ---------- ----------
               Europe Fox    ---------- -------a-- ---------- ---------- ---------- ----t----- ---------- ---------- ---------- ----------
               Desert Fox    ---------- -------a-- -----a---- ---------- ---------- ----t----- ---------- ---------- ---------- ----------
               Gabon Dog     ---------- -------a-- t--------- --------t- ---------- ----t----- ---------- ---------- ---t--c--- ----------
               Pasteur Virus ---------- -------a-- ---------- ---------- ---------- a--------- ---------- ---------- ------c--- ----------

               Consensus     GAGAAAGAAC TTCAAGA-TA CGAGGCGGCT GAACTGACAA AGACTGACGT GGCACTGGCA GATGATGGAA CTGTCAACTC TGACGATGAG GACTACTTCT

                                110
Non-rabies     Mokola1       -t--g----- ---------- -----g---- -c-g---g-- a--------- --t--caagt -g----aggt t--------c c-g--ca-ag ----g--c--
               Mokola5       -t--a----- t--------- -----a---- -c-g-a-g-- a--------c aac---aa-t -g----a-gt t--c-----t --g---a-tg -g--g-----
               Duvenhage     ----a--t-- a--------a ---------- -c--aa-g-- ---------c -----g--gt -----g-gg- a--c---a-a --g--c--c- -t--a-----

VIII           Sfwc1679      -a--c----- ------c--- ---------- ---------- t--------c -----ca--- -g-------- -------a-- --------t- -c-----c--
               Sfwm1686      -g--c--g-- ------c--- ---------- ---------- ---------c -----ca--- -g-------- -------a-- --------t- -c-----c--

IX             Sfym1653      -g-------- ------c--- ---------- ---------- ---------- ---------- ---------- a--c---a-- --------t- -c--------
               Sfym1655      -g--c----- ------c--- ---------- -----a---- -r-------c ---------- ---------- a--c---a-- --------t- ----------
               Sfym1654      -g--c----- -c----g--- -----c---- ----c----- ---------c ---------t ----a----- a--c---a-- --------t- -t--------
               Sfbf1688      -a--c----- -c----c--- -----g---- -c-------- ---------c ---------t ---------- a--c---a-- --------t- -t--------

VI             Nigeria Dog   -------g-- ---g------ -----c---- -------g-- ---------c -----g---- ---------- t--c---a-- --------t- ----------
               Nigeria Dog   -------g-- ---g------ ---------- -------g-- ---------c -----g---- ---------- t--c---a-- --------t- ----------

I              Sfjk1670      ---------- ---------a --------c- ----ca---- a--------- ---------- ---------- -s-------- --------c- ----------
               Waku62        ---------- ---------a --------c- ----ca---- a-n------- ---------- ---------- -s-------- --------c- ----------
               Zijk1690      ---------- ---------a --------c- ----ca---- a--------- ---------- ---------- -s-------- --------c- ----------
               Kehm945       ---------- ---------a --------c- -----a---- a---------- --------g- ------ag-- -n-------- --------c- ----------
               Europe Fox    ---------- ---------a ---------- -c-------- ---------- ---------- ----a----- c--------- --------c- ----------
               Desert Fox    ---------- ---g-----a -----c---- ---------- ---------- ---------- ---------- ---------- --------c- ----------
               Gabon Dog     ----g----- ---------a ---------- ---------- ---------- ---------- ---------- ---------- --------c- ----------
               Pasteur Virus -a-------- ---------g ---------- ---------- ---a------ ---------- -g-------- ---c------ --------c- ----------

               Consensus     CCGGTGAAAC CAGAAGTCCT GAAGCTGTTT ATACTCGAAT CATGATGAAT GGAGGTCGAC TAAAGAGATC GCATATACGG AGATATGT-T CAGTCAGTTC
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FIG. 10 cDNA sequences from the carboxy terminus of the rabies nucleoprotein. The consensus sequence represents the most common nucleotide at each position. Sequence identical to the consensus is indicated by –. Non-identical nucleotides are identified as A, adenosine; C, cytidine; G, guanosine; T, N, no base could be determined

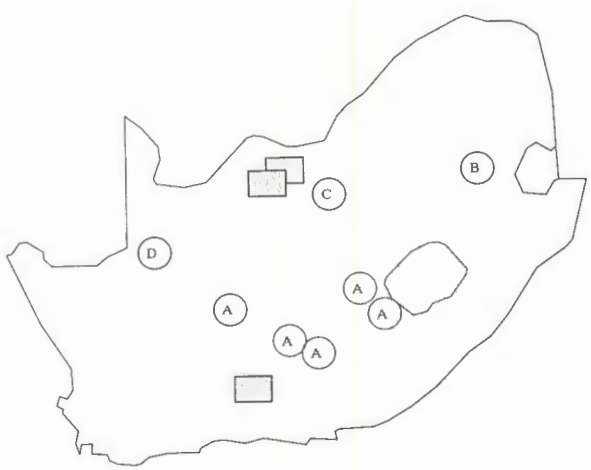

FIG. 11 Distribution in South Africa of rabies variant identified as genetic groups VIII and IX. 3,03 □, genetic group VIII, ○; A, genetic group IXA; B, genetic group IXB; C, genetic group IXC; D, genetic group IXD

identified in South Africa; however, if rabies in the yellow mongoose is incidental to a reservoir of rabies maintained in another or several other species, the effect will be short-term. Unless vaccination programs are continued indefinitely, herd immunity will eventually wane and new non-immune mongoose populations will again be susceptible to infection through contact with the reservoir species.

Genetic relatedness among members of the lyssavirus family of Rhabdoviruses

Because several articles in this volume will address antigenic and genetic typing of other lyssaviruses, Fig. 13 is intended to place the analyses presented in this report in the context of a phylogenetic analysis of non-rabies lyssaviruses. Despite significant sequence divergence in rabies variants transmitted in different reservoirs, rabies variants associated with domestic and wild carnivores are still much more closely related to each other than to isolates of Mokola, Duvenhage and the European bat lyssaviruses.

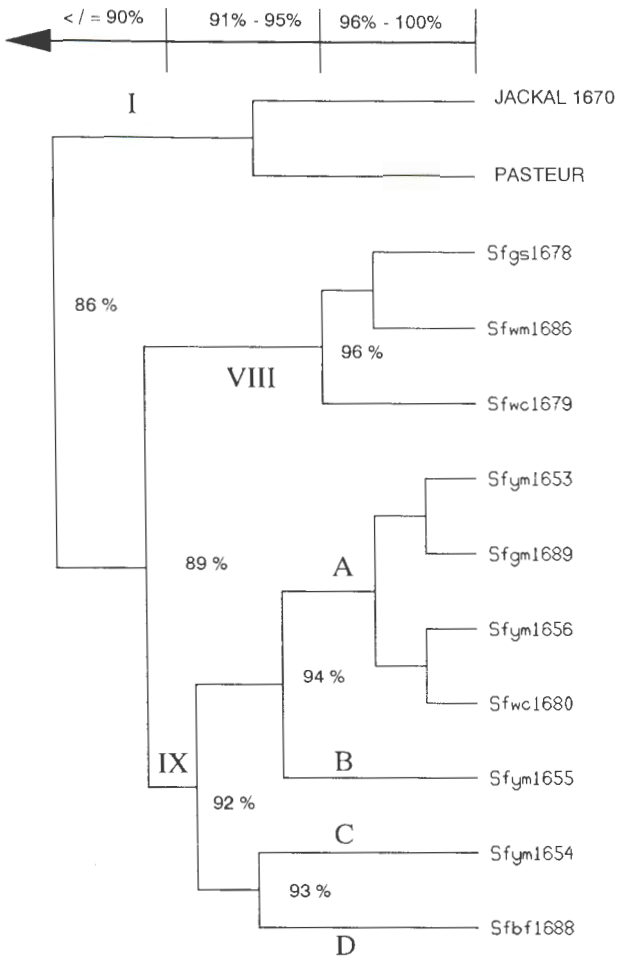

FIG. 12 Graphical representation of genetic relationships among rabies isolates identified by reaction patterns 3, 4, and 5 in Fig. 2. Sample identification is as before (gs, ground squirrel; wc, wild cat; ym, yellow mongoose; gm, gray mongoose)

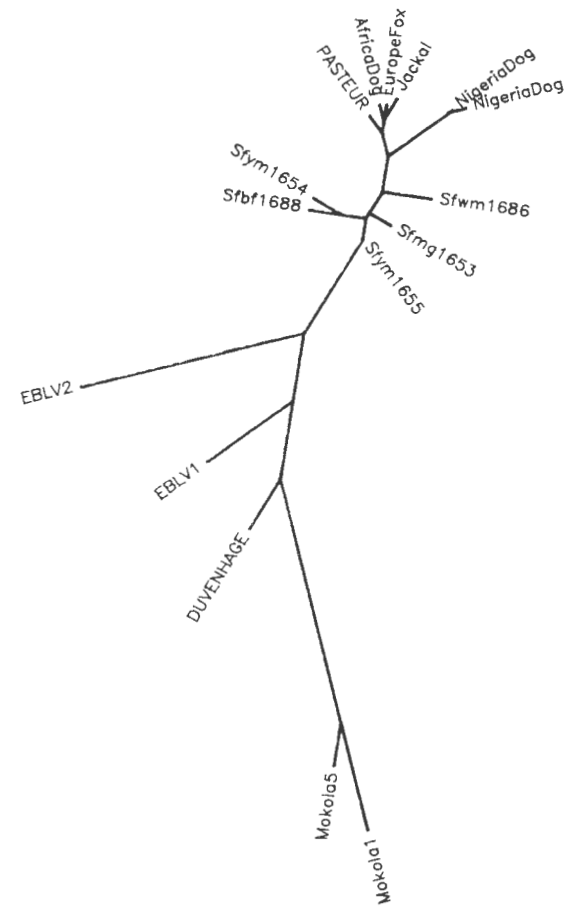

FIG. 13 Phylogenetic tree of rabies samples from genetic groups I, VI, VIII and IX and non-rabies lyssavirus samples analyzed by the method of maximum likelihood. Branch length is proportional to genetic distance between samples. Mokola 1, isolate from Nigeria; Mokola 5, isolate from Zimbabwe; EBLV1, isolate from Denmark; EBLV2, isolate from Holland

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REFERENCES

BLANCOU, J., AUBERT, M.F.A. & ARTOIS, M. 1991. Fox rabies, in *The natural history of rabies*, 2nd ed., edited by G.M. Baer. CRC Press: 258–289.

HUDSON, J.R. 1944. A short note on the history of rabies in Kenya. *East African Medical Journal*, 21:322–327.

HUTCHEON, D. 1894. *Reports of the colonial veterinary surgeon and assistant veterinary surgeons for the year 1893*. Cape of Good Hope: Department of Agriculture.

JOHNSON, H.N. 1945. Fox rabies. *Journal of the Medical Association of Alabama*, 14:268–271.

KREBS, J.W., HOLMAN, R.C., HINES, U., STRINE, T.W., MANDEL, E.J. & CHILDS, J.E. 1992. Rabies surveillance in the United States during 1991. *Journal of the Veterinary Medical Association*, 201:1836–1848.

SMITH, J.S. 1989. Rabies virus epitopic variation: Use in ecologic studies, in *Advances in Virus Research*, edited by K. Maramorosch, F.A. Murphy & A.J. Shatkin. San Diego: Academic Press: 215–254.

SMITH, J.S., YAGER, P.A., BIGLER, W.J. & HARTWIG, C. 1990. Surveillance and epidemiologic mapping of rabies variants in Florida. *Journal of Wildlife Diseases*, 26: 473–485.

SMITH, J.S., ORCIARI, L.A., YAGER, P.A., SEIDEL, H.D. & WARNER, C.K. 1992. Epidemiologic and historical relationships among 88 rabies virus isolates as determined by limited sequence analysis. *Journal of Infectious Diseases*, 166:296–307.

SMITH, J.S. & SEIDEL, H.D. 1993. Rabies: A new look at an old disease, in *Progress in Medical Virology*, edited by J. Melnick. Basel: Karger: 40:82–106.