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Rapid Survey and Assessment of the Northern Sportive Lemur, *Lepilemur septentrionalis*, in Northern Madagascar

Jean Freddy Ranaivoarisoa¹, John R. Zaonarivelo², Runhua Lei³, Steig E. Johnson⁴, Tracy M. Wyman⁴, Russell A. Mittermeier⁵ and Edward E. Louis Jr.^{1,3*}

¹Madagascar Biodiversity Partnership, Antananarivo, Madagascar

²Sciences of Life and Environment Department, Faculty of Sciences, University of Antsirana, Madagascar

³Center for Conservation and Research, Omaha's Henry Doorly Zoo and Aquarium, Omaha, NE, USA

⁴Anthropology Department, University of Calgary, Calgary, Canada

⁵Conservation International, Arlington, VA, USA

Abstract: The northern sportive lemur, *Lepilemur septentrionalis*, faces imminent danger of extinction, more so than any other lemur in Madagascar. The population estimates for this sportive lemur remain unknown because of habitat loss and ongoing human encroachment, but they are unlikely to number more than a few hundred individuals. We present the results of extensive surveys conducted in 2010, 2011 and 2012 of known habitat, the Sahafary and Analalava classified forests, and confirm the species designation of the sportive lemur observed in Montagne des Français in 2007. Six *L. septentrionalis* individuals were examined in Sahafary in 2011, along with eight individuals during the 2010 and 2011 expeditions to Montagne des Français. A July 2012 survey in extended forest fragments of Montagne des Français identified another 10 individuals. Surveys of the Analalava forest in 2011 detected no northern sportive lemurs, despite documenting their presence in 2005. One individual was recorded in the July 2012 survey. Although the morphological data of the sportive lemurs at Montagne des Français was comparable to that of Sahafary, the sportive lemur at Montagne des Français was subsequently verified as *L. septentrionalis* with mitochondrial DNA D-loop sequence data analyses. The confirmation of the northern sportive lemur at Montagne des Français is significant since it establishes additional habitat for this species. However, sustained human encroachment from Antsirana continues to finance the production of charcoal and collection of sand; activities that are threatening this population. Habitat loss and hunting continue to be the principal threats to the long-term survival of the northern sportive lemur. With only 19 known individuals, we urge immediate conservation action for this Critically Endangered species.

Keywords: northern sportive lemur, *Lepilemur septentrionalis*, Sahafary, Montagne des Français

Introduction

The northern sportive lemur, *Lepilemur septentrionalis*, a small sportive lemur, inhabits dry forests of northern Madagascar (Rumpler and Albignac 1975; Junger and Rumpler 1976; Rumpler *et al.* 2001). The distribution of this nocturnal lemur was initially believed to extend from the Montagne d'Ambre region in the north to the Mahavavy River in the west (Hawkins *et al.* 1990; Ratsirason and Rumpler 1998; Rumpler *et al.* 2001). Rumpler and Albignac (1975) originally described *L. septentrionalis* with four subspecies (*L. s. andrafiamenensis*, *L. s. ankaranensis*, *L. s. sahafarensis*, and *L. s. septentrionalis*). Based on the parameter of geographic separation, Groves (2001) reduced the four subspecies to two, *L. s. septentrionalis* (*L. s. sahafarensis* as a junior

synonym) and *L. s. ankaranensis* (*L. s. andrafiamenensis* as a junior synonym). Rumpler (2004) and Ravaoarimanana *et al.* (2004) subsequently elevated these two subspecies to species based on cytogenetic and molecular data. The taxonomic status of *L. septentrionalis* has continued to be supported by more detailed cytogenetic, morphological and molecular data (Rumpler *et al.* 2001; Ravaoarimanana *et al.* 2004; Andriaholinirina *et al.* 2006; Louis *et al.* 2006; Lei *et al.* 2008; Ramaromilanto *et al.* 2009). Due to the taxonomic revision confirming *L. septentrionalis* and *L. ankaranensis* as distinct species, the perceived range of the northern sportive lemur was drastically reduced; limited to a few degraded patches of dry forest in the Sahafary region just south of Antsirana (Fig. 1). Considered the southern geographic extent for this species, the Irodo and Bobakindro rivers, combined with

the limestone plateau and tsingy formation of Ankarana and Analamerana, could be acting as a significant barrier to dispersal (Louis *et al.* 2008).

Recognized as Critically Endangered on the IUCN Red List of Threatened Species and ranked as one of the world's 25 most endangered primates (Mittermeier *et al.* 2008; Mittermeier *et al.* 2009; IUCN 2012), the northern sportive lemur is in imminent danger of extinction, more so than any other lemur (Mittermeier *et al.* 2010). Expeditions carried out by the Madagascar Biodiversity Partnership and Omaha's Henry Doorly Zoo and Aquarium in 2005, recorded the northern sportive lemur in the classified forests of Analalava and Sahafary; its identity subsequently verified through molecular genetics analysis (Louis *et al.* 2006). Analalava classified forest persists as a very small scrubby and degraded habitat of about 80 ha (Figs. 1–3). Sahafary classified forest is embedded in a *Eucalyptus* plantation that is routinely used for charcoal production, and there is less suitable habitat even than in Analalava. An independent survey led by R. A. Mittermeier in 2005 visited the forest fragments of Andrahona and Tsaratanana near Ankarongana. They also went to Madirobe, mentioned by Y. Rumpler (*pers. comm.*) as a possible site for *L. septentrionalis*, but no forest was found there and

the local people seemed not to know anything about the animal. Andrahona is a sacred mountain, located 36 km south of Diego-Suarez. The forest there was in reasonably good condition, especially around the peak, but there were many trails and significant numbers of saplings were being taken out for construction. Local people said that sportive lemurs occurred there, but none were found. Further surveys there in September 2012, led by E. E. Louis, recorded only *Microcebus tavaratra* and *Eulemur coronatus*. The forest at Tsaratanana was a tiny tract of degraded, heavily used forest in the middle of a large *Eucalyptus* plantation, 5.4 km north-west of the town of Ankarongana. A guide from Ankarongana was familiar with the animal and a single *L. septentrionalis* was found (Fig. 4). It had a small "V"-shaped cut in its ear, so was perhaps an animal that had been studied by Y. Rumpler's team. The chances of protecting the forest there were considered very remote.

Due to the political instability created by the 2009 coup, little if any security is being provided for any protected or non-protected area in Madagascar. According to a survey in 2007, approximately one hundred northern sportive lemurs were estimated in the Andrahona, Ankarakataova and Sahafary regions, none of which are protected (Mittermeier *et*

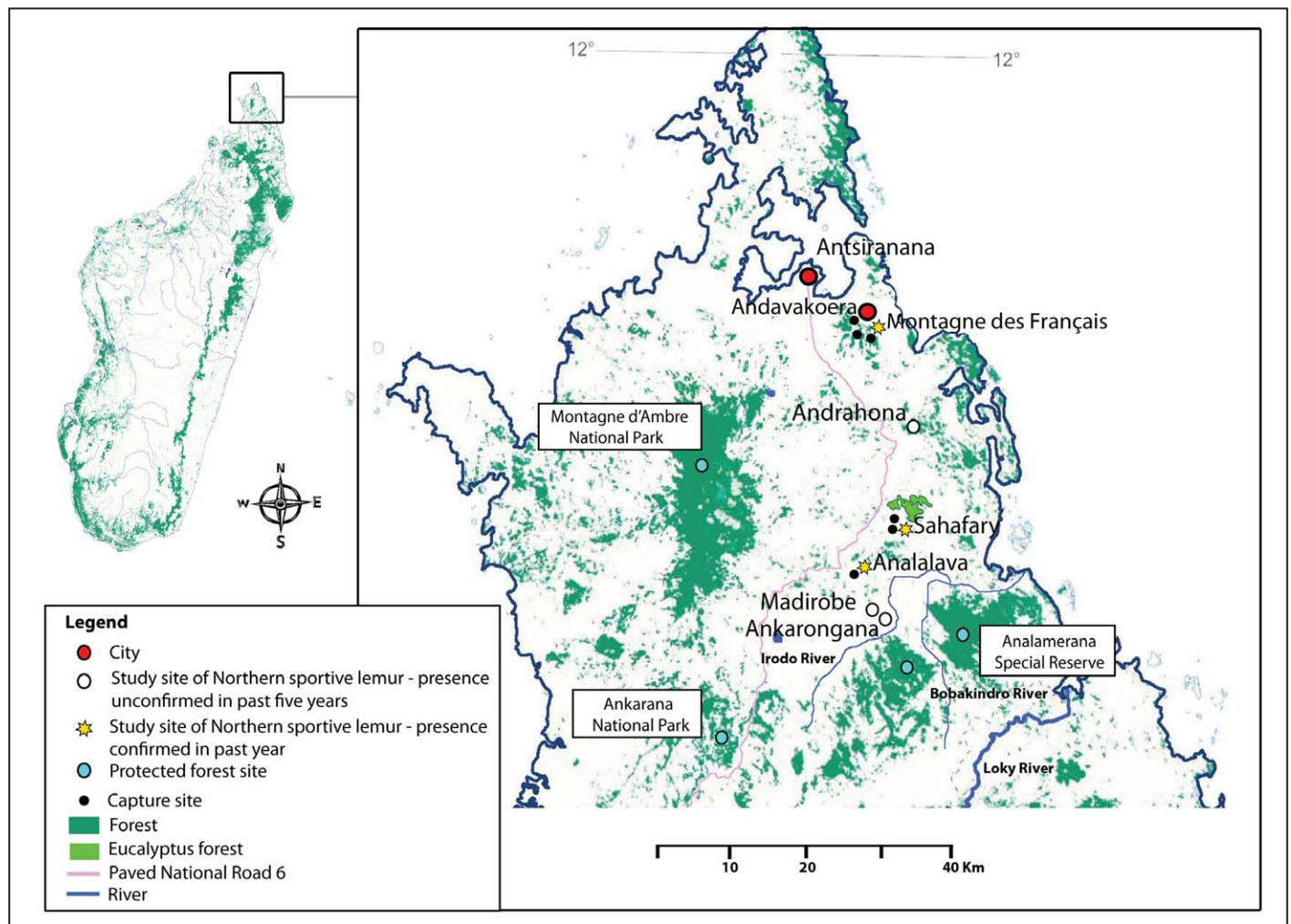


Figure 1. Distribution range and capture localities of the northern sportive lemur, *Lepilemur septentrionalis*.

al. 2008; Mittermeier *et al.* 2010). During a comprehensive survey of amphibians and reptiles at Montagne des Français, D’Cruze *et al.* (2007) photographed what they presumed to be *L. septentrionalis*. However, without a reference to indicate the relative dimensions in the photograph, direct confirmation could not be made without a clear-cut comparison to the nominal species in the region, *L. septentrionalis* and *L. ankaranensis*. Here, we present the results of three extensive surveys conducted in 2010, 2011 and 2012 that revisited the Sahafary and Analalava forests, and determined the species status and range of the unspecified sportive lemur noted at Montagne des Français (Fig. 3). We also suggest conservation measures for the remaining populations of the Critically Endangered northern sportive lemur.



Figure 2. Landscape of the Analalava region showing the remaining forest in the valley (center of photograph). Loss of habitat is evident throughout the horizon. Introduced *Eucalyptus* forests can be seen on the ridge above the Analalava forest (left side of photograph), along with the effects of erosion from long-term slash-and-burn practices creating a massive ridge. Photograph taken by Jean Freddy Ranainarisoa.



Figure 3. Landscape of the Sahafary region showing the tremendous loss of habitat and crater-like landscape created by effects of erosion from long-term slash-and-burn practices. Photograph taken by Jean Freddy Ranainarisoa.

Methods

Study area

The northern sportive lemur is found in dry deciduous forest fragments, restricted to the extreme northern region of Madagascar (Fig. 1). Research was conducted at Montagne des Français (12°20' 02.7"S, 49°21' 21.9"E) in 2010, 2011 and 2012, and in Sahafary (12°36'20.7"S, 49°26'32.8"E) and Analalava (12°39'25.6"S, 49°24'36.5"E) in 2011 and 2012 (Figs.1–3; Table 1). This forest belt is found south-east of the seaport city of Antsiranana, in the district of Diego I, near the commune of Ramena, extending south into the district of Diego II, near the communes of Sadjoavato and Ankarongana.

Sample collection

All lemurs investigated in this study were wild-caught, free-ranging individuals immobilized with a CO₂ projection rifle or blowgun with 10 mg/kg of Telazol® (Fort Dodge Animal Health; Overland Park, Kansas; Table 1). Four 2.0 mm biopsies and 1.0 cc per kilogram of whole blood were collected from each sedated animal and immediately stored in room temperature storage buffer (Seutin *et al.* 1991). Fecal samples were collected and stored immediately in 10 ml of RNALater® (Life Technologies, Grand Island, New York). A HomeAgain® microchip (Schering-Plough Veterinary Corp.; Kenilworth, New Jersey) was placed subcutaneously between the scapulae of each lemur. This procedure was used to field-catalog each animal with a unique recognition code in order to re-identify all captured individuals during any future immobilizations. Morphometric measurements were also taken following Louis *et al.* (2006). For presentation purposes, we summarize all measurements by population in this publication following the guidelines of Smith and Jungers (1997). Genomic DNA was extracted from samples using a phenol-chloroform/isoamyl extraction (Sambrook *et al.* 1989).

Data generation

From these samples, the displacement loop or control region of mitochondrial DNA (530-553 base pairs) was amplified (D-loop; Baker *et al.* 1993; Wyner *et al.* 1999) under the conditions described in other studies (Louis *et al.* 2006; Lei *et al.* 2008; Ramaromilanto *et al.* 2009). The samples were electrophoresed on a 1.2% agarose gel to verify the PCR product and purified with Exonuclease I and shrimp alkaline phosphatase (EXOSAP; Silva *et al.* 2001).

The purified products were cycle sequenced using a BigDye® terminator sequencing kit (Life Technologies, Carlsbad, CA). The sequences were analyzed by capillary electrophoresis with an Applied Biosystems 3130xl genetic analyzer. The PCR and sequencing primer suite from Louis *et al.* (2006) were used to generate the D-loop fragment sequences. The sequences were aligned to generate a consensus contig using Sequencher® 4.10 (Gene Codes Corporation; Ann Arbor, MI). All sequences have been deposited in GenBank with accession numbers JQ771832-JQ771834. We used accessioned GenBank D-loop sequences for sportive lemur

species to establish reference baselines (Louis *et al.* 2006; Lei *et al.* 2008; Ramaromilanto *et al.* 2009).

Phylogenetic analysis

Maximum likelihood (ML) analyses for the D-loop sequence data were performed with PhyML 3.0 software under HKY+I+G model selected by Modeltest 3.7, along with bootstrap percentages computed after 1000 replicates (Guindon and Gascuel 2003; Guindon *et al.* 2005; Posada and Crandall 1998). The best-scoring ML-trees were searched and saved in PAUP* 4.0b10 (Swofford 2001). Bayesian inference analysis was conducted using MrBayes 3.0b4 (Huelsenbeck and Ronquist 2001; Ronquist and Huelsenbeck 2003). A Markov Chain Monte Carlo (MCMC) run with four simultaneous chains and 1,000,000 generations was performed with HKY+I+G model selected for the ML inferences by using MrModeltest 2.2 (Nylander 2004). Every hundredth generation, the tree with the best likelihood score was saved, resulting in 10,000 trees. These were condensed in a majority rule consensus tree using PAUP* 4.0b10 (Swofford 2001) and clade posterior probabilities (PP) were computed. MEGA 4.0 (Tamura *et al.* 2007) was used to calculate uncorrected pairwise distances ('p') and absolute differences.

Results

Six *Lepilemur septentrionalis* were captured in Sahafary in 2011 (Table 1). Ten individuals were sampled in Montagne des Français: five in 2010 and five in 2011, along with two recaptured individuals identified by previously implanted microchips. Unfortunately, no northern sportive lemur was detected during the 2011 expedition into the Analalava forest despite its noted presence in 2005, but one individual was documented during the July 2012 expedition (Table 1). Furthermore, only one solitary female crowned lemur, *Eulemur coronatus*, was

seen during the week-long field survey at this site in 2011, but none in 2012. Morphological data are presented in Table 2. The northern sportive lemur individuals from Montagne des Français are slightly larger in average size than the individuals from Sahafary (650 g and 630 g, respectively).

The uncorrected pairwise differences among all recognized *Lepilemur* species for D-loop sequences ranged from

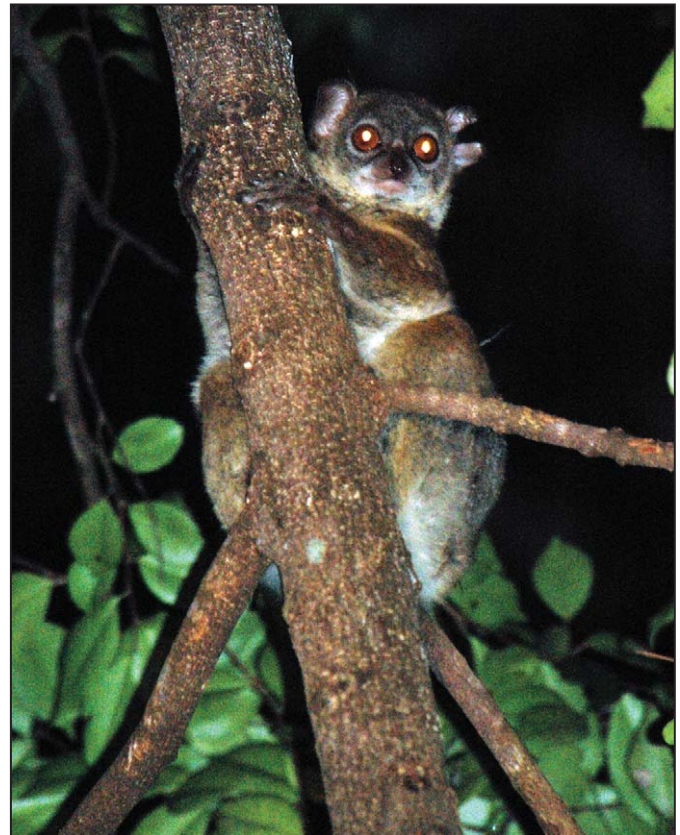


Figure 4. Sahafary sportive lemur (*Lepilemur septentrionalis*), Tsaratanana Forest, north-west of Ankarongana village. Photo by R. A. Mittermeier.

Table 1. List of northern sportive lemur, *Lepilemur septentrionalis*, examined during this study.

ID Number	Location	Sex	Microchip ID	Weight (kg)	Global Positioning System Coordinates		Survey Year
MDF10.1	Montagne des Français	Female	47311E676C	0.58	S12°20'07.4"	E:049°21'17.4"	2010
MDF10.2	Montagne des Français	Female	460B1D4B08	0.73	S12°20'04.3"	E:049°21'16.2"	2010, 2011
MDF10.3	Montagne des Français	Male	473110605D	0.63	S12°20'04.6"	E:049°21'19.0"	2010
MDF10.4	Montagne des Français	Female	46703A4210	0.58	S12°20'05.9"	E:049°21'18.2"	2010, 2011
MDF10.5	Montagne des Français	Female	460A1C2344	0.68	S12°20'05.3"	E:049°21'19.3"	2010
MDF11.1	Montagne des Français	Female	4732703611	0.73	S12°20'02.7"	E:049°21'21.9"	2011
MDF11.2	Montagne des Français	Female	46702C5F16	0.72	S12°20'04.2"	E:049°21'26.3"	2011
MDF11.3	Montagne des Français	Male	4730466E37	0.70	S12°20'05.0"	E:049°21'25.4"	2011
MDF11.4	Montagne des Français	Female	4731203336	0.65	S12°20'59.2"	E:049°21'34.5"	2011
MDF11.5	Montagne des Français	Female	460COC2B45	0.56	S12°20'03.5"	E:049°21'16.3"	2011
FARY11.1	Sahafary (Madirobe)	Male	467025447A	0.47	S12°36'20.7"	E:049°26'32.8"	2011
FARY11.2	Sahafary (Madirobe)	Female	476D115C41	0.70	S12°36'25.2"	E:049°26'34.2"	2011
FARY11.3	Sahafary (Madirobe)	Female	4773303F28	0.69	S12°36'35.7"	E:049°26'35.5"	2011
FARY11.4	Sahafary (Madirobe)	Male	47682A370D	0.67	S12°36'35.7"	E:049°26'35.5"	2011
FARY11.5	Sahafary (Madirobe)	Male	47732C6D2C	0.61	S12°36'34.2"	E:049°26'35.1"	2011
FARY11.6	Sahafary (Madirobe)	Female	4777511349	0.68	S12°36'32.4"	E:049°26'32.7"	2011

2.3% to 15.5% (Tables 3a and 3b). The uncorrected pairwise differences for D-loop sequences between *L. septentrionalis* and the other 25 nominal sportive lemur species ranged from 5.1% to 13.4%. The uncorrected pairwise differences representing interspecific variation between *L. septentrionalis* from Montagne des Français and Sahafary/Analalava were 1.4%. In the maximum likelihood and Bayesian phylogenetic tree reconstructions, similar tree topologies were obtained (Fig. 6). Three individual northern sportive lemurs from Montagne des Français, MDF10.2, MDF10.4 and MDF10.5, clustered closely with the one haplotype of *L. septentrionalis* from Sahafary/Analalava (Fig. 6).

Discussion

In 2005, the only known populations of the northern sportive lemur were reconfirmed as originally recorded by Rumpler and Albignac (1975; Louis *et al.* 2006). Both morphometric and molecular analyses confirm that the sportive lemur at Montagne des Français is indeed *L. septentrionalis*, extending the distribution from the Sahafary and Analalava regions (Fig. 6; Tables 1–3). The confirmation of the northern sportive lemur is significant since it establishes a new locality for this species. The expeditions in 2010 and 2011 verified the continuing existence of the northern sportive lemur in Sahafary classified forest, but not a single animal was detected during these expeditions in the Analalava forest where it formerly existed in 2005. During the July 2012 expedition, however, one individual was reported when Analalava was revisited. The northern sportive lemurs from Montagne des Français were slightly larger in average size than those from Sahafary (0.65 kg and 0.63 kg, respectively), but this difference could be attributed to the level of an individual's maturity (Tables 1–2). Another possible explanation for the slight size difference could be severe habitat degradation in Sahafary versus higher quality forest forage in Montagne des Français (Fig. 3).

In 2008, Service d'Appui à la Gestion de l'Environnement (SAGE; <www.MadagascarSAGE.org>) promoted the designation of Montagne des Français as a newly protected area, and supported the development of a Vondron'Olona Ifototra (VOI) in Andavakoera, the primary village of this mountain forest. A VOI is a local government organization at the village level that manages the region's resources. Montagne des Français is the most pristine forest available for the northern sportive lemur, providing natural quality habitat for this Critically Endangered species.

SAGE estimates that approximately 20,000 people are exploiting this forest even though Andavakoera has only 200 residents. Business interests from the nearby seaport of Antsiranana finance the exploitation of the remaining Montagne des Français habitat through the production of charcoal and collection of sand for city and port construction projects (D'Cruze *et al.* 2007). Habitat loss from uncontrolled long-term slash-and-burn practices, regrowth of non-endemic invasive plants, and the desertification effects of deforestation and erosion has resulted in a mosaic landscape of forest fragments, grasslands, and craters (Figs. 2–3). Human encroachment is compounded by debilitating poverty and exacerbated by the political instability of the past four years. Moreover, opportunistic hunting and illegal logging over the past seven years has impacted all lemur populations throughout the region. During the previous 2005 expedition in the Analalava forest, numerous northern sportive lemurs were noted, along with multiple family groups of crowned lemurs, *Eulemur coronatus*, and Sanford's lemurs, *E. sanfordi*. During the 2011 week-long survey, however, only one solitary female crowned lemur was observed and the majority of the forest was gone (Fig. 2). No other lemurs were documented in the 2012 survey except for one *L. septentrionalis*. Even in the Sahafary region, habitat loss has been severe due to the effects of erosion during this same period. The folivorous diet of the northern sportive lemur and this species' predilection for leaves complicates any attempts or plans to maintain it in captivity. There is no record of a sportive lemur held in any zoological park, as all known attempts to maintain them in captivity have failed on average within one week of capture. *In situ* conservation



Figure 5. Northern sportive lemur, *Lepilemur septentrionalis*, from Montagne des Français. Photograph taken by Edward E. Louis, Jr.

Table 2. Average morphometric data (cm) summarized from northern sportive lemur, *Lepilemur septentrionalis*, populations.

	HC	Body L.	Tail L.	F-H	F-UR	F-Hd	F-LD	F-Tb	H-F	H-T	H-Ft	H-LD	H-Tb
MDF	7.6±0.5	20.7±1.1	26.7±1.0	6.2±1.1	6.6±0.5	5.4±0.3	2.6±0.2	1.4±0.2	10.1±0.8	9.0±0.8	7.8±0.5	2.6±0.2	2.7±0.3
FARY	7.6±0.2	20.7±2.9	23.9±1.6	5.4±0.2	6.3±0.6	4.9±0.4	2.6±0.2	1.3±0.1	10.5±0.6	9.5±0.8	6.4±0.8	2.8±0.2	2.3±0.3

Note: F-H: Humerus, F-Hd: Hand, F-LD: longest digit (Forelimb), F-Tb: Thumb (forelimb), F-UR: Ulna/radius, HC: head crown, H-F: Femur, H-Ft: foot, H-LD: longest digit (Hindlimb), H-T: Tibia, H-Tb: Thumb (Hindlimb), L.: length, MDF: From Montagne des Français, RATY: From Sahafary (Madirobe)

Table 3a. Genetic distance matrix for D-Loop fragment sequence data for the 26 nominal sportive lemur species. The numbers represent the following *Lepilemur* species: 1. *L. ankaraensis*, 2. *L. milanoii*, 3. *L. tymerlachsonorum*, 4. *L. dorsalis*, 5. *L. ahmansorum*, 6. *L. sahamalazensis*, 7. *L. septentrionalis*, 8. Population from Montagne des Français, 9. *L. edwardsi*, 10. *L. grewcockorum*, 11. *L. otto*, 12. *L. microdon*, 13. *L. leucopus*, 14. *L. petteri*, 15. *L. randrianasoloi*, 16. *L. aeeclis*, 17. *L. ruficaudatus*, 18. *L. hubbardorum*, 19. *L. fleuretiae*, 20. *L. mustelinus*, 21. *L. betsileo*, 22. *L. jamesorum*, 23. *L. wrightae*, 24. *L. seali*, 25. *L. hollandorum*, 26. *L. scottorum*. Genetic distances based on absolute differences are displayed above the diagonal, and genetic distances based as a percentage are displayed below the diagonal.

	1	2	3	4	5	6	7	8	9	10	11	12	13
1		12.7	19.7	30.8	31.2	29.7	34.0	33.8	33.0	45.2	44.7	48.8	43.7
2	2.6±0.6		11.6	24.4	30.2	28.0	29.2	27.0	35.8	42.5	48.1	47.5	43.7
3	4.0±0.8	2.3±0.6		23.5	29.0	21.0	28.0	25.0	37.0	41.5	45.3	50.3	44.0
4	6.2±1.0	4.9±0.9	4.8±0.9		25.8	16.8	29.5	29.5	32.8	35.2	40.2	49.8	49.0
5	6.3±1.0	6.1±1.0	5.9±1.0	5.2±0.9		26.0	37.0	34.0	27.0	37.0	38.0	44.8	45.4
6	6.0±1.0	5.7±1.0	4.3±0.8	3.4±0.7	5.3±0.9		33.0	33.0	28.0	34.5	37.7	44.3	45.2
7	6.9±1.0	5.9±1.0	5.7±1.0	6.0±1.0	7.5±1.1	6.7±1.0		7.0	39.5	41.5	47.3	51.3	40.4
8	6.8±1.0	5.5±0.9	5.1±0.9	6.0±1.0	6.9±1.1	6.7±1.0	1.4±0.5		40.5	40.5	47.7	52.3	42.6
9	6.7±1.0	7.2±1.1	7.5±1.1	6.6±1.0	5.5±1.0	5.7±1.0	8.0±1.1	8.2±1.2		27.5	22.0	33.5	31.4
10	9.1±1.1	8.6±1.2	8.4±1.2	7.1±1.1	7.5±1.1	7.0±1.0	8.4±1.2	8.2±1.2	5.6±0.9		29.5	37.8	42.7
11	9.0±1.2	9.7±1.3	9.2±1.2	8.1±1.1	7.7±1.1	7.6±1.1	9.6±1.2	9.6±1.3	4.5±0.8	6.0±0.9		39.4	44.1
12	9.9±1.2	9.6±1.2	0.2±1.3	0.1±1.2	9.1±1.2	9.0±1.2	10.4±1.3	10.6±1.3	6.8±1.0	7.6±1.1	8.0±1.1		42.3
13	8.8±1.2	8.9±1.2	8.9±1.2	9.9±1.2	9.2±1.2	9.1±1.2	8.2±1.1	8.6±1.2	6.4±1.0	8.6±1.2	8.9±1.1	8.6±1.2	
14	9.1±1.2	9.2±1.2	9.1±1.2	8.8±1.2	10.0±1.2	8.0±1.1	7.6±1.1	8.2±1.2	6.6±1.0	8.6±1.2	9.0±1.1	9.0±1.2	2.9±0.6
15	9.0±1.2	9.4±1.3	9.7±1.3	10.2±1.3	9.2±1.2	10.2±1.3	8.6±1.2	8.5±1.2	8.5±1.1	9.0±1.2	9.9±1.2	10.1±1.2	5.9±1.0
16	10.5±1.2	10.7±1.3	10.4±1.3	10.4±1.3	9.5±1.2	10.1±1.2	9.7±1.2	10.3±1.3	8.2±1.0	8.9±1.2	9.8±1.2	9.8±1.2	8.4±1.1
17	10.9±1.3	11.2±1.3	11.0±1.3	10.8±1.3	11.3±1.3	9.9±1.3	9.9±1.2	10.0±1.2	10.1±1.3	10.4±1.3	10.0±1.2	11.0±1.3	9.3±1.2
18	10.1±1.2	10.3±1.3	10.3±1.3	9.9±1.2	10.4±1.3	10.8±1.3	9.3±1.2	9.1±1.2	9.5±1.2	9.5±1.2	9.8±1.2	11.3±1.3	8.3±1.2
19	12.6±1.4	12.4±1.4	13.2±1.4	13.2±1.4	11.4±1.3	12.1±1.4	13.2±1.4	12.3±1.4	12.4±1.4	12.9±1.4	12.9±1.4	13.1±1.4	13.9±1.5
20	14.0±1.4	13.3±1.4	13.3±1.4	13.4±1.4	13.3±1.4	12.7±1.4	13.3±1.4	13.6±1.4	13.1±1.3	14.1±1.4	13.9±1.3	12.7±1.3	13.3±1.4
21	12.1±1.3	11.6±1.3	11.7±1.3	12.4±1.4	12.2±1.4	11.2±1.4	13.2±1.4	12.6±1.4	12.2±1.4	13.1±1.4	12.9±1.4	11.8±1.3	13.6±1.5
22	13.2±1.4	12.8±1.3	13.2±1.4	13.3±1.4	12.5±1.3	12.3±1.4	13.0±1.4	12.4±1.3	13.1±1.4	14.0±1.4	13.3±1.3	13.1±1.4	14.4±1.4
23	13.2±1.4	12.7±1.4	11.6±1.3	11.9±1.3	11.2±1.3	11.8±1.3	12.3±1.3	11.8±1.3	11.7±1.3	11.1±1.3	11.5±1.3	12.2±1.4	13.6±1.4
24	12.8±1.4	12.4±1.4	13.2±1.5	13.7±1.4	13.0±1.4	13.4±1.4	13.4±1.4	13.2±1.4	13.1±1.4	12.4±1.4	13.9±1.4	12.3±1.3	15.3±1.5
25	13.1±1.3	13.5±1.4	12.3±1.4	13.9±1.4	14.2±1.4	12.6±1.3	12.8±1.4	12.4±1.4	13.5±1.4	14.3±1.4	13.7±1.4	13.7±1.3	14.7±1.4
26	12.4±1.4	12.4±1.4	13.0±1.4	13.5±1.4	12.6±1.4	12.6±1.4	13.4±1.4	12.8±1.4	12.8±1.4	14.1±1.5	14.7±1.5	14.7±1.4	16.0±1.5

Table 3b. Genetic distance matrix for D-Loop fragment sequence data for the 26 nominal sportive lemur species. The numbers represent the following *Lepilemur* species: 1. *L. ankaraensis*, 2. *L. milanoii*, 3. *L. tymerlachsonorum*, 4. *L. dorsalis*, 5. *L. ahmansorum*, 6. *L. sahamalazensis*, 7. *L. septentrionalis*, 8. Population from Montagne des Français, 9. *L. edwardsi*, 10. *L. grewcockorum*, 11. *L. otto*, 12. *L. microdon*, 13. *L. leucopus*, 14. *L. petteri*, 15. *L. randrianasoloi*, 16. *L. aeeclis*, 17. *L. ruficaudatus*, 18. *L. hubbardorum*, 19. *L. fleuretiae*, 20. *L. mustelinus*, 21. *L. betsileo*, 22. *L. jamesorum*, 23. *L. wrightae*, 24. *L. seali*, 25. *L. hollandorum*, 26. *L. scottorum*. Genetic distances based on absolute differences are displayed above the diagonal, and genetic distances based as a percentage are displayed below the diagonal.

	14	15	16	17	18	19	20	21	22	23	24	25	26
1	44.8	44.5	51.9	53.6	49.7	62.1	69.0	60.0	65.2	65.4	63.4	64.6	61.5
2	45.3	46.6	53.0	55.2	51.0	61.4	65.5	57.2	63.1	62.5	61.4	66.5	61.2
3	44.8	47.8	51.5	54.2	50.8	65.0	65.9	58.0	65.0	57.5	65.0	61.0	64.0
4	43.4	50.6	51.4	53.4	49.0	65.2	66.1	61.2	65.7	58.8	67.5	68.7	66.5
5	49.6	45.4	46.8	56.0	51.5	56.5	65.7	60.2	61.8	55.5	64.0	70.0	62.0
6	39.4	50.2	50.0	48.7	53.2	60.0	62.6	55.3	60.7	58.5	66.0	62.0	62.0
7	37.8	42.6	48.0	48.8	45.8	65.0	65.6	65.0	64.3	60.8	66.0	63.3	66.0
8	40.6	42.0	51.0	49.5	45.2	61.0	67.2	62.0	61.3	58.5	65.0	61.3	63.0
9	32.5	41.9	40.5	49.7	46.8	61.5	64.7	60.5	64.5	58.0	64.5	66.5	63.0
10	42.7	44.7	43.8	51.5	47.2	63.5	69.5	64.5	69.2	54.8	61.5	70.5	69.5
11	44.7	48.8	48.2	49.4	48.4	63.7	68.9	63.7	65.7	56.8	68.7	67.6	72.7
12	44.4	49.8	48.5	54.6	55.6	64.8	62.5	58.3	64.6	60.4	60.8	67.8	72.5
13	14.4	28.9	41.4	45.8	40.9	68.6	65.9	67.2	71.1	67.4	75.8	72.6	78.8
14		31.7	41.1	43.5	43.6	66.6	62.5	64.1	68.1	67.0	70.2	68.3	75.0
15	6.4±1.0		30.6	37.3	28.9	66.4	66.5	66.6	67.0	64.0	70.6	67.7	74.8
16	8.3±1.1	6.2±1.0		41.7	39.0	73.0	66.1	65.0	73.0	64.4	65.0	70.0	73.3
17	8.8±1.1	7.6±1.0	8.4±1.1		31.9	62.2	56.9	62.2	60.8	67.5	66.7	66.9	76.5
18	8.8±1.2	5.8±1.0	7.9±1.1	6.5±1.0		67.3	63.0	61.1	64.6	66.1	68.3	73.5	74.2
19	13.5±1.5	13.4±1.4	14.8±1.5	12.6±1.4	13.6±1.4		37.7	42.0	37.0	50.2	45.0	52.7	52.0
20	12.7±1.4	13.5±1.4	13.4±1.4	11.5±1.3	12.7±1.4	7.6±1.0		26.2	27.4	46.3	44.5	52.9	50.8
21	13.0±1.4	13.5±1.4	13.2±1.4	12.6±1.4	12.4±1.4	8.5±1.2	5.3±0.8		26.6	44.8	46.0	50.0	47.7
22	13.8±1.4	13.6±1.4	14.8±1.4	12.3±1.3	13.1±1.3	7.5±1.0	5.5±0.8	5.4±0.8		43.5	47.7	45.7	45.3
23	13.6±1.4	13.0±1.4	13.0±1.3	13.7±1.4	13.4±1.4	10.2±1.2	9.4±1.1	9.1±1.2	8.8±1.1		49.3	49.9	56.7
24	14.2±1.5	14.3±1.5	13.2±1.5	13.5±1.4	13.8±1.4	9.1±1.2	9.0±1.2	9.3±1.3	9.6±1.2	10.0±1.3		35.7	29.0
25	13.8±1.4	13.7±1.4	14.2±1.4	13.5±1.4	14.9±1.4	10.7±1.3	10.7±1.3	10.1±1.2	9.2±1.1	10.1±1.2	7.2±1.1		30.7
26	15.2±1.5	15.1±1.5	14.8±1.5	15.5±1.6	15.0±1.5	10.5±1.3	10.3±1.3	9.6±1.3	9.2±1.2	11.5±1.4	5.9±1.0	6.2±1.0	

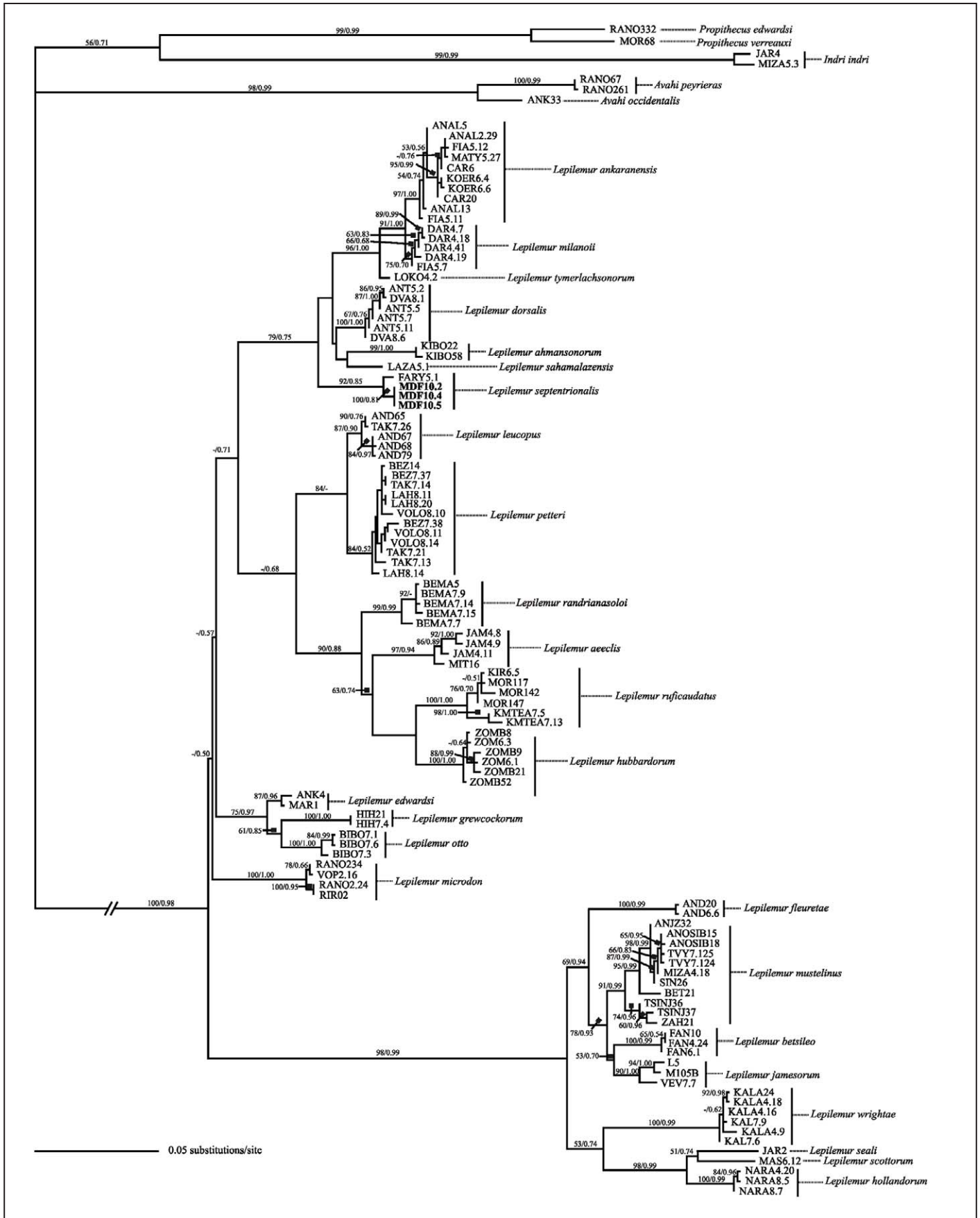


Figure 6. Phylogenetic relationships between *Lepilemur* species inferred from the maximum likelihood and Bayesian approaches of D-loop sequences from 108 sportive lemur individuals with seven outgroup taxa. BP/PP represent bootstrap support (BP) and posterior probability support (PP) values, respectively. We obtained the maximum likelihood phylogram (-ln likelihood = 6522.06) from the D-loop alignment from a transition/transversion ratio of 4.37 and a shape parameter of 0.73.

programs and community-based interactions are therefore the only viable solutions.

The development of an interconnected program between the VOI, the local gendarme and community can provide the platform for reducing the illegal charcoal activities and habitat loss in the last remaining vestige of habitat for the northern sportive lemur. Conservation strategies that emphasize education, monitoring, reforestation, and research should be implemented to leverage the biodiversity and rebuild the fragile ecosystem in northern Madagascar. This multi-layered program should be linked to sustainable alternative agricultural practices and the introduction of fuel-efficient rocket stoves and briquette technology. The best measures of success will be greater accessibility to food and increased income to the region, both of which are interconnected with habitat restoration and the long-term survival of the northern sportive lemur.

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- Authors' addresses:*
Jean Freddy Ranaivoarisoa, Madagascar Biodiversity Partnership, Antananarivo 101, Madagascar, **John R. Zaonarivelo**, Sciences of Life and Environment Department, Faculty of Sciences, University of Antsirana, Antsirana 201, Madagascar, **Runhua Lei**, Center for Conservation and Research, Omaha's Henry Doorly Zoo and Aquarium, Omaha, NE 68107, USA, **Steig E. Johnson** and **Tracy M. Wyman**, Anthropology Department, University of Calgary, Calgary, AB T2N 1N4, Canada, **Russell A. Mittermeier**, Conservation International, 2011 Crystal Drive Arlington, VA 22202, USA, and **Edward E. Louis, Jr.**, Madagascar Biodiversity Partnership, Antananarivo 101, Madagascar, and Center for Conservation and Research, Omaha's Henry Doorly Zoo and Aquarium, Omaha, NE 68107, USA. *Corresponding author:* Edward E. Louis, Jr., e-mail: <edlo@omahazoo.com>.