Journal of Mammalogy, 2026, **00**, 1–11
https://doi.org/10.1093/jmammal/gyag084
Research Article

Reassessing the diversity and distribution of African rope squirrels (*Funisciurus* Trouessart, 1880) in the Congo Basin

Pascal Baelo[1,†], Léa Fourchault[2,3,†], Ann Ceulemans[4], Nicolas Laurent[2,5], Claude Mande[1,6,7], Anne Laudisoit[6], Nicaise Amundala[7], Ganiyat Temidayo Saliu[2], Herwig Leirs[6], Guy-Crispin Gembu[1,7], Sophie Gryseels[2,6], Jan Hulselmans[6], Erik Verheyen[2,6,†], and Joachim Mariën[4,6,*,†]

[1]Biodiversity Monitoring Centre (CSB), University of Kisangani, Kisangani, Avenue Kithima 4, Kisangani, Tshopo, Democratic Republic of the Congo
[2]Vertebrate Group, Directorate Taxonomy and Phylogeny, Royal Belgian Institute of Natural Sciences, 29 Vautierstraat, Brussels, Brussels, 1000, Belgium
[3]Systems Ecology and Resource Management, Université Libre de Bruxelles, Boulevard du Triomphe, Brussels, Brussels, 1000, Belgium
[4]Department of Biomedical Sciences—Virus Ecology Group, Institute of Tropical Medicine Antwerp, Antwerp, Nationalestraat 155, Antwerp, Antwerp, 2000, Belgium
[5]Department of Environmental Sciences, Open Universiteit, Valkenburgerweg 177, Heerlen, Limburg, 6419, The Netherlands
[6]Evolutionary Ecology Group, Department of Biology, University of Antwerp, Antwerp, Universiteitsplein 1, Wilrijk, Antwerp, 2610, Belgium,
[7]Department of Ecology and Wildlife Management, University of Kisangani, Avenue Kithima 4, Kisangani, Tshopo, Democratic Republic of the Congo

*Corresponding author: Virus Ecology unit, Department of Biomedical Sciences, Institute of Tropical Medicine Antwerp, Kronenburgstraat 43, 2000 Antwerpen, Belgium. Email: jmarien@itg.be
†These authors contributed equally as first or last authors of this work.

Abstract

The taxonomy, distribution, and ecology of sub-Saharan squirrels are poorly documented. This study focuses on the diversity and distribution of rope squirrels (*Funisciurus* Trouessart, 1880) in the Congo Basin. We compiled the most comprehensive genetic (*n* = 397 sequences) and morphometric (*n* = 219 measured skulls) dataset to date, covering much of their geographical and taxonomic range. Analysis of mitochondrial markers recovered 10 MOTUs (Molecular Operational Taxonomic Unit), nuclear markers 2 MOTUs, and morphometrics 5 Operational Taxonomic Units. Integrating this evidence suggests at least six species-level lineages in our dataset: four allopatric taxa related to *F. anerythrus*; four genetically different taxa (of which two are sympatric species) that resemble *F.* cf. *bayonii*; one taxon corresponding to *F. pyrropus akka* de Winton, 1895; one morphologically distinct taxon that resembles *F.* cf. *congicus*; and one unidentified taxon. Their distributional ranges reveal that the Congo and Kasaï Rivers act as major biogeographical barriers, suggesting that they originated by allopatric speciation. Our findings provide a baseline for studying the effects of habitat loss on rope squirrel populations in the Congo Basin and inform research into their role as reservoirs of zoonotic pathogens, including mpox virus.

Keywords Sciuridae, Congo Basin, DNA-barcoding, Zoonoses, Rope squirrels, Mpox

With over 300 species in 51 genera, the Sciuridae are one of the most widespread mammalian families (Zachos et al. 2021; Steiner and Huettmann 2023). They play key roles as seed dispersers and ecosystem engineers and, in some regions, are important as a food source or in traditional medicine (Fa et al. 2002; Baelo et al. 2018; Fourchault et al. 2025). Additionally, squirrels can act as reservoirs of zoonotic pathogens (Bernd Hoffmann et al. 2015; Sklenovská and Van Ranst 2018; Schulze et al. 2020). Yet, despite their ecological and public health significance, their evolution and ecology remain poorly documented, particularly in Central Africa.

Central African squirrels comprise 7 genera, including the African pygmy (*Myosciurus pumilio*), rope (*Funisciurus*), sun (*Heliosciurus*), ground (*Xerus*), bush (*Paraxerus*), palm (*Epixerus*), and giant (*Protoxerus*) squirrels (Thorington et al. 2012; Kingdon and Happold 2013). The Protoxerini tribe (*Heliosciurus*, *Protoxerus*, *Paraxerus*, *Funisciurus*) is most widespread and occurs in rainforests and savannahs. Within Protoxerini, the genus *Funisciurus*, described first by Trouessart in 1880, is particularly well represented in museum collections and currently counts 10 recognized species (Kingdon and Happold 2013; mammaldiversity.org/, accessed on 22 January 2026). Recent work suggests that *Funisciurus* speciation is strongly influenced by major rivers, as shown for the Sanaga River in Cameroon (Curaudeau et al. 2025). This pattern raises the possibility that the large rivers of the Congo Basin may have similarly contributed to diversification within this group. However, support for this scenario remains tentative, as the currently inferred distributional ranges (primarily

Associate Editor: Deyan Ge
Received: March 20, 2025. **Accepted:** August 27, 2026
© The Author(s) 2026. Published by Oxford University Press on behalf of the American Society of Mammalogists.
This is an Open Access article distributed under the terms of the Creative Commons Attribution License (https://creativecommons.org/licenses/by/4.0/), which permits unrestricted reuse, distribution, and reproduction in any medium, provided the original work is properly cited.

Downloaded from academic.oup.com/jmammal/advance-article/doi/10.1093/jmammal/gyag084/8993869 by guest on 23 October 2026

based on museum specimens) may be unreliable due to uncertainty in morphological identifications of specimens (Monadjem et al. 2015).

Funisciurus squirrels have also been repeatedly linked to mpox virus (MPXV) circulation (Khodakevich et al. 1987; Chen et al. 2005; Curaudeau et al. 2023; Kinganda Lusamaki et al. 2025; Riutord-Fe et al. 2026). Mapping their distributions and phylogenetic relationships across sub-Saharan Africa could provide insights into virus–host co–evolution, as suggested by observations in Cameroon where distinct *Funisciurus* clades were associated with different MPXV lineages capable of infecting humans (Nakazawa et al. 2015; Curaudeau et al. 2025). Moreover, *Funisciurus* squirrels can attain high densities in human-modified landscapes, with up to 500 individuals per km² in palm oil plantations (Emmons 1980), increasing human–squirrel contact and potential MPXV spillover amid accelerating forest degradation and land–use change (Rimoin et al. 2017). Clarifying the taxonomy and distribution of *Funisciurus* species is therefore essential for understanding MPXV ecology and guiding targeted wildlife and public health surveillance across the region.

Within the genus *Funisciurus*, 8 species are currently reported from the Congo Basin, of which *F. anerythrus* is considered the most widespread (Kingdon and Happold 2013). Originally described from Uganda (Thorington and Hoffmann 2005), *F. anerythrus* is currently divided into 4 subspecies based on morphology: *F. a. anerythrus* (from Uganda to northern Angola); *F. a. bandarum* (from Chad to Central African Republic); *F. a. mystax* (from Cameroon to Gabon); and *F. a. raptorum* (restricted to Benin and Nigeria; Kingdon and Happold 2013). Other *Funisciurus* species reported in the Congo Basin are *F. carruthersi* (Rwenzori region), *F. bayonii* (Angola to DRC), *F. congicus* (DRC, Angola to Namibia), *F. pyrropus* (Senegal to DRC and Rwanda), *F. leucogenys* (from Ghana to Central African Republic), *F. isabella* (rainforest of Central Africa), and *F. lemniscatus* (rainforest of Central Africa). However, the current diversity and distribution of these species is mainly based on morphological records from museum specimens only, and have yet to be reassessed using integrative approaches that combine dense geographic sampling with molecular data.

In this study, we address these knowledge gaps by: (i) reassessing the taxonomic diversity of *Funisciurus* across the Congo Basin integrating molecular and morphometric approaches; and (ii) testing whether the Congo River acts as a major geographic barrier shaping their distributions.

Methods

Sample collection

We trapped and sampled squirrels between 2010 and 2022 at 17 localities across four provinces of the Democratic Republic of Congo (DRC: Tshopo, Bas-Uele, Ituri, and Tshuapa). Additional specimens from other localities in 3 provinces (Kwilu, Kinshasa, and Kongo Central), collected and preserved at the Royal Museum for Central Africa (RMCA, Tervuren, Belgium) were also included (Fig. 1).

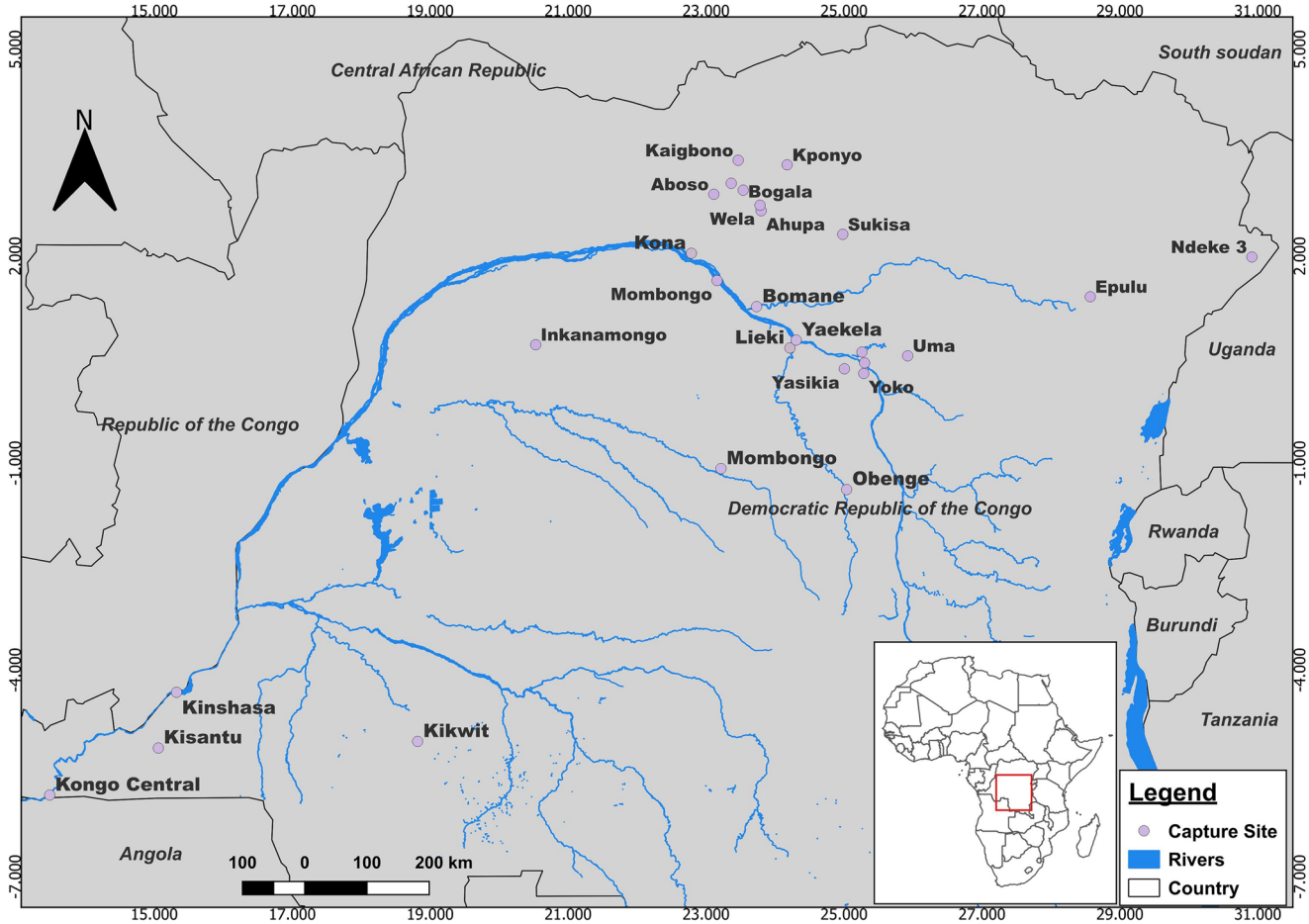


Fig. 1 Geographical location of collection sites. GenBank sequences were obtained from individuals captured near Kinshasa, Boende, and Kaigbono. Specimens for craniometric measurements and molecular analysis were obtained from the Biodiversity Monitoring Center in the DR Congo and the Royal Museum for Central Africa in Belgium.

Downloaded from academic.oup.com/jmammal/advance-article/doi/10.1093/jmammal/gyag084/8993869 by guest on 23 October 2026



Fig. 2 Traditional squirrel trap baited with palm nuts from DR Congo.

Squirrels were captured using traditional noose traps (Fig. 2) baited with ripe palm nuts and set in primary forest, secondary forest, and fallow land from ground level to 7 m above ground. Since standard rodent traps (pitfall, Havahart, flap, Sherman) were ineffective, we used traditional traps as the main method. These traps were constructed from stems, flexible vines or rods, rattan, wire fixed to branches, and fitted with 1 or 2 sliding nooses on a simple trigger, allowing capture from either direction.

Live-captured squirrels were euthanized using isoflurane in accordance with American Society of Mammalogists guidelines (Sikes et al. 2016). Morphological measurements were taken (see below), and specimens were dissected. Tissue samples (tongue, kidney, liver, lung, feces, heart, skin, and/or spleen) were preserved in 70–94% ethanol, RNAlater, or DNA/RNA shield solutions and shipped to Belgium for genetic analyses. Carcasses were fixed in 10% formalin and preserved either at the Centre de Surveillance de la Biodiversité in Kisangani (DRC) or at the University of Antwerp. A full specimen list is provided in Supplementary Data SD1.

Squirrels were initially identified in the field to genus or species level following Kingdon and Happold (2013) and Monadjem et al. (2015). We recorded external traits including coat characteristics and measurements of body, tail, hind foot, and ear. Sex and reproductive status were assessed externally: in females, nipple development (small or swollen) and vaginal state (closed or open); in males, testis position (abdominal or scrotal). Body measurements were taken on fresh specimens to the nearest millimeter using a Mitutoyo caliper (0.05 mm), and body mass (g) was measured with a Pesola spring balance (Supplementary Data SD2).

Adult squirrel skulls were dissected from a subset of the data ($n = 194$), and 23 standard craniometric variables were measured (codes and definitions in Supplementary Data SD3; Verheyen and Bracke 1966). Additionally, 25 skulls from the RMCA collections, identified as *F.* cf. *congicus*, were included.

DNA was extracted from squirrel tissues (tongue, liver, kidney, heart, skin, or lung) using either the NucleoSpin Tissue Mini Kit (Macherey-Nagel, Düren, Germany) or the Qiagen DNeasy Blood and Tissue Kit (Qiagen, Hilden, Germany), following the manufacturer's protocols. Mitochondrial cytochrome *b* (*Cytb*) fragments were amplified using primers L14723 with H15149 or H15915 (Montgelard et al. 2002), and nuclear apolipoprotein b (*ApoB*) fragments were amplified using primers ApoBf and ApoBr (Dubey et al. 2007). Amplicons were purified and sequenced by Macrogen (Amsterdam, The Netherlands) or the Flemish Institute of Biotechnology. Chromatograms were manually checked and edited, and sequences were aligned in Geneious Prime 2025.2 using the MAFFT alignment plugin.

The final *Cytb* and *ApoB* alignments included novel sequences from *Funisciurus*, *Paraxerus*, *Protoxerus*, and *Heliosciurus*, along with 2 outgroup sequences from GenBank (*Tamias* HQ698386.1 and *Marmota* KJ458055.1). We also incorporated all available Congolese *Funisciurus* sequences from GenBank (MF577066–MF577074, MF694312–MF694319, PV606393.1, PV606404.1, PV606400.1, PV606401.1, PV606405.1), yielding a final dataset of 397 mitochondrial *Cytb* and 62 nuclear *ApoB* sequences.

Research permits

Research permits were obtained through the Centre de Surveillance de la Biodiversité at the University of Kisangani. Samples exported to Belgium before the implementation of the Nagoya Protocol in the DRC were covered by material transfer agreements issued by the Centre de Surveillance de la Biodiversité, whereas those exported after 2018 were authorized under a Nagoya export permit issued by the Congolese Ministry of the Environment and Sustainable Development (MEDD): 003/ANCCB-RDC/SG-EDD/BTB/08/2019, 018/ANCCB-RDC/SG-EDD/BTB/07/2022, 021/SG-EDD/BTB/ANCCB-RDC/08/2023, 011/ANCCB-RDC/SG-EDD/BTB/2023. Permission to import the samples into Belgium was granted by the Federal Agency for the Safety of the Food Chain (FASFC): 2022DBP504, 2023DBP370, 2023DBP371, 2024DBP424.

Genetic species delimitation

To delineate species in our dataset, we integrated 5 complementary species delimitation methods (Nesi et al. 2011; Demos et al. 2014; Sloan et al. 2017).

We aligned all mitochondrial *Cytb* and nuclear *ApoB* sequences in Geneious using the MAFFT alignment plugin, and inferred a maximum-likelihood phylogeny in IQ-TREE (v3.0.1; Nguyen et al. 2015). The dataset included all Protoxerini sequences from our study, with *Marmota* and *Tamias* as outgroups. The best-fitting nucleotide substitution model was selected with ModelFinder (-m TEST), and node support was assessed using 10,000 ultrafast bootstrap replicates and 10,000 SH-aLRT replicates (Guindon et al. 2010; Hoang et al. 2018).

After assigning MOTUs to the specimens based on the IQTREE outcome, we tested whether craniometric and external morphological variation corresponded to mitochondrial MOTUs using multivariate analyses in R v4.2.3. A PERMANOVA test was first performed to test for significant sexual dimorphism within species. As no significant differences were observed, we calculated basic descriptive statistics (mean and SD; Supplementary Data SD4 and SD5) for all species. Subsequently, a discriminant analysis was performed on the full craniometric (23 variables) and morphometric (5 variables) data, with sequenced specimens explicitly indicated. Prior to these analyses, missing values in the craniometric variables were imputed using the mean of the corresponding MOTU to maximize sample retention, with remaining missing values replaced by the overall dataset mean. Group separation in multivariate space was visualized using the first 2 linear discriminant (LD) axes. Finally, differences among groups were formally tested using MANOVA with *Wilks' Γ*, followed by pairwise comparisons on PCA-reduced scores (to account for limited sample sizes in some group pairs) with adjusted *P*-values.

To reduce our dependence on explicit phylogenetic or craniometric reconstruction only, we further applied a computationally efficient, distance-based species delimitation approach (ASAP; Puillandre et al. 2021). ASAP infers species partitions by detecting

Downloaded from academic.oup.com/jmammal/advance-article/doi/10.1093/jmammal/gyag084/8993869 by guest on 23 October 2026

barcode gaps, *i.e.*, sharp contrasts between low intraspecific and high interspecific genetic distances. Unlike other barcode-gap approaches, ASAP does not require a user-defined maximum intraspecific divergence, which is particularly advantageous for poorly studied groups such as African rope squirrels. We applied ASAP to partitions based on both mitochondrial (*Cytb)* and nuclear (*ApoB*) gene fragments to obtain an independent, distance-based perspective on species boundaries. ASAP was run via the SpartExplorer web service (Miralles et al. 2022).

To further verify the robustness of our species delimitation methods, we applied a multi-rate Poisson Tree Processes (mPTP) method (Kapli et al. 2017). This tree-based approach uses single-locus phylogenetic branching patterns to infer species boundaries by testing whether the number of substitutions follows distinct Poisson processes within versus between species. An important advantage of mPTP is that it explicitly models variation in branching rates across lineages, making it less dependent on the presence of clear barcode gaps and complementary to purely distance-based methods such as ASAP. We used the IQ-TREE maximum-likelihood (ML) phylogeny as input for the mPTP analyses (Supplementary Data SD6 and SD7). We then visually compared the species partitions proposed by ASAP and mPTP with the topology of the mitochondrial ML tree and with the results of discriminant analyses based on morphometric data.

Finally, we calculated pairwise genetic distances from the aligned *Cytb* sequences in R using the dist.dna function in the "ape" R package (Paradis and Schliep 2019), applying the Kimura 2-parameter (K2P/K80) substitution model. Sites containing gaps or missing data were handled using pairwise deletion.

Results

Mitochondrial *Cytb* analyses recovered 10 *Funisciurus* MOTUs, all supported by ASAP, mPTP, K2P genetic distance, and the topology of the mitochondrial maximum-likelihood tree (Figs 3 and 4; Table 1). The optimal ASAP solution (ASAP score = 3.5; P =0.09) and the mPTP multi-rate model (log-likelihood = 954.567 *vs* single-species null = 717.972; 229/792 edges exceeding the branch-length threshold of 0.0001) delimited 3 MOTUs of *F. anerythrus* from the Congo River left bank (FaLB1 from Thsopo and Thsuapa, FaLB2 from Kikwit, and FaLB3 from Kinshasa); 1 *F. anerythrus* MOTU from the Congo River right bank (FaRB; including voucher specimen FMNH149422 from Epulu, DRC); 1 *F. pyrropus* MOTU (Fp, including FMNH148995 from Burundi); 1 MOTU representing an unidentified *Funisciurus* lineage captured in Yoko and Boende (Fx); and 4MOTUs within a left-bank lineage morphologically resembling *F.* cf. *bayonii* (FbLB1a, FbLB1b, FbLB1c, and FbLB2). From the latter, FbLB1a and FbLB2 occur sympatrically in Yoko. In agreement, pairwise *Cytb* K2P distances among the squirrel taxa ranged from 0.04 to 0.22, indicating mitochondrial divergence higher than expected intraspecific variation (Table 1). The 10 well-supported mitochondrial lineages occupy distinct geographic ranges, often delimited by major rivers (Fig. 5). Fx, FaLB1, and FbLB1/FbLB2 occur exclusively south of the Congo River; whereas FaRB and Fp are restricted to the north/right bank. Fc,

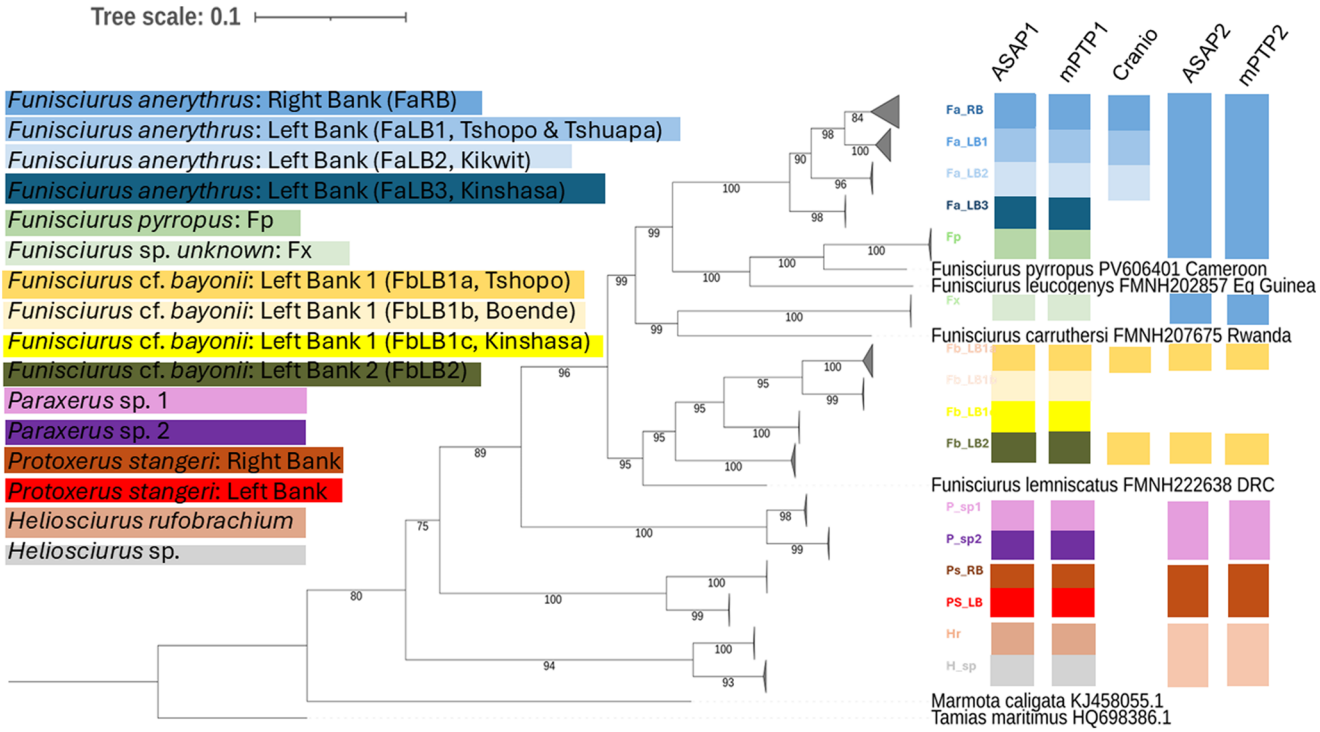


Fig. 3 Proposed Central African squirrel Molecular Operational Taxonomic Units using integrated taxonomic methods. Maximum Likelihood tree consisting of 397 cytochrome *b* sequences, rooted with *Tamias* as outgroup with 10,000 bootstrap iterations; ASAP, Assemble Species by Automated Partitioning, method applied to mitochondrial (ASAP1) and nuclear (ASAP2) sequences; mPTP, the multi-rate Poisson Tree Process, applied to mitochondrial (mPTP1) and nuclear (mPTP2) data; Cranio, a discriminant analysis applied to craniometric measures. RB, right bank of the Congo River; LB, left bank of the Congo River. Fx, previously undescribed *Funisciurus* species (n = 3 sequences); Fp, *F. pyrropus*; Fb, *F.* cf. *bayonii*; Fa, *F. anerythrus*. *Heliosciurus*, *Protoxerus*, and *Paraxerus* clades are shown for informative purposes but were not further analyzed due to the limited number of sequences available for these subclades.

Downloaded from academic.oup.com/jmammal/advance-article/doi/10.1093/jmammal/gyag084/8993869 by guest on 23 October 2026



Fig. 4 Different *Funisciurus* species captured in our study: (A) *Funisciurus* cf. *bayonii* (FbLB1a dorsal, Yoko); (B) *F.* cf. *bayonii* (FbLB1a ventral, Yoko); (C) *F. anerythrus* (FaRB, Epulu); (D) *F. anerythrus* (FaLB, Yoko); (E) *F.* sp. unidentified (Fx, Inkanamongo); (F) *F. pyrropus* (Fp, Epulu).

Table 1 Pairwise relationships among the identified *Funisciurus* molecular operational taxonomic units (MOTUs) inferred from cytochrome *b* sequences.

MOTU	FA RB	FA LB1	FA LB2	FA LB3	FB1a	FB1b	FB1c	Flem	FB LB2	FC	Fx	FP	Fleu	TM
FA RB		96	92	92	83	84	85	85	83	83	82	83	83	79
FA LB1	4		93	93	83	84	87	85	83	84	83	83	83	79
FA LB2	8	7			83	82		84	84	82	81	83	82	79
FA LB3	8	6			85	85	86	88	83	86	85	85	84	79
FB1a	18	19	19	16		92	90	85	86	82	82	81	82	78
FB1b	18	19	21	16	8		90	86	86	82	82	83	82	79
FB1c	17	14		15	10	10		88	88	86	83	82	85	80
F. lemniscatus	17	16	17	13	16	16	12		88	85	84	83	85	81
FB LB2	17	18	16	18	14	14	12	12		83	83	82	83	78
F. carruthersi	19	18	21	15	21	21	16	17	18		84	82	84	79
Fx	20	19	21	17	21	20	19	18	18	18		82	83	78
FP	19	19	18	16	22	20	21	19	20	20	21		85	78
F leucogenys	19	19	21	18	20	20	16	17	18	18	20	16		79
T. maritimus	24	24	24	25	25	25	23	22	25	24	25	25	24	

The lower diagonal shows Kimura 2-parameter (K2P) genetic distances, representing model-corrected mitochondrial sequence divergence, while the upper diagonal shows the percentage of identical nucleotide sites between sequences. *Tamias maritimus* was used as the outgroup.

FaLB2, and FaLB3 are confined to the left/south bank of the Kasaï River in our dataset.

The more slowly evolving nuclear *ApoB* marker recovered 2 lineages, a result that was consistent across ASAP and mPTP, and supported by the topology of the nuclear maximum-likelihood tree: *F. anerythrus* (including Fx, Fp, FaLB, FaRB) and *F.* cf. *bayonii* (including FbLB1a, FbLB1b, FbLB1c, and FbLB2; Fig. 3; Supplementary Data SD7). The best-supported ASAP partition (ASAP score = 1.0; P =0.12) and the mPTP multi-rate model (log-likelihood = 145.383 *vs* 131.911 under the single-rate model; 32/122 edges exceeding the branch-length threshold of 0.0001) delimited the same 2 MOTUs.

Linear discriminant analysis (LDA) of 23 craniometric variables showed strong differentiation among MOTUs (df = 6; *Wilks'* Γ = 0.009; P <0.0001). The first discriminant axis separated *F. anerythrus* from *F.* cf. *bayonii* and *F.* cf. *congicus* (Fig. 6), consistent with pairwise multivariate differentiation in the MANOVA fitted on PCA scores (Supplementary Data SD8). We note that *F.* cf. *congicus* is not molecularly confirmed; its assignment is provisional and based on the RMCA database identification and the species description in Mammals of Africa. FaRB and FaLB1 were separated along the second discriminant axis and were also significantly differentiated in pairwise comparisons (*Wilks'* Γ = 0.23; P < 0.001). The 2 *F.* cf. *bayonii* genotypes (FbLB1a

Downloaded from academic.oup.com/jmammal/advance-article/doi/10.1093/jmammal/gyag084/8993869 by guest on 23 October 2026

and FbLB2) could not be statistically distinguished, likely due to small sample sizes (*Wilks'* Γ = 0.15; P =0.59), although visual inspection did not suggest pronounced separation either. Craniometric data for *F. pyrropus* were sparse (n = 5) and largely overlapped with FaRB. Finally, 1 molecularly confirmed FaLB specimen from Kikwit (FaLB2, KIK675) fell outside the morphometric clusters of all other OTUs (including FaLB1), suggesting that it may represent a distinct OTU based on craniometry.

LDA based on external morphometrics (body mass, head–body length, tail length, hind foot length, and ear length) also indicated clear separation among OTUs (*Wilks'* Γ = 0.229, P <0.0001). As observed for craniometry, the first discriminant axis primarily distinguished *F. anerythrus* from *F.* cf. *bayonii* (Fig. 6), matching the pattern of pairwise multivariate differences from the MANOVA on PCA scores (Supplementary Data SD9). Differentiation between FaRB and FaLB1 was most evident along the second discriminant axis and remained significant in pairwise testing (Wilks' λ = 0.818, P = 0.0002). In contrast, the two *F. bayonii* genotypes (FbLB1 and FbLB2) were not separable statistically, likely reflecting limited sample sizes (*Wilks'* Γ = 0.919, P = .99), and visual inspection likewise suggested substantial overlap. External measurements for *F. pyrropus* were scarce (n = 5) and largely fell within the FaRB morphospace, while external morphometric data were not available for the remaining taxa. Finally, PERMANOVA on craniometric variables provided no evidence for sexual dimorphism within FaRB, FaLB, or FbLB (Supplementary Data SD10).

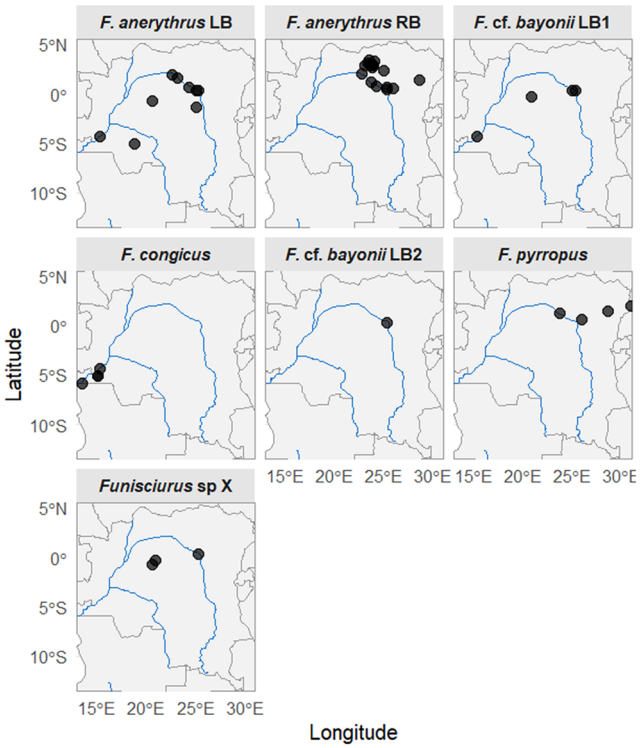


Fig. 5 Maps showing the distribution of the species and subspecies identified in relation to the Congo River and its tributaries.

Discussion

Using an integrative dataset of molecular and morphometric data, we reassessed *Funisciurus* taxonomic diversity in the Congo Basin. Our results reveal notable mismatches between recognized species and the genetic and morphometric patterns observed (Fig. 3), and show that the Congo and Kasaï Rivers sharply delimit OTU ranges (Fig. 5), highlighting the role of hydrological barriers and the need to revisit rope squirrel taxonomy and IUCN distribution maps. Combining evidence across genetic markers and craniometry, we propose the presence of

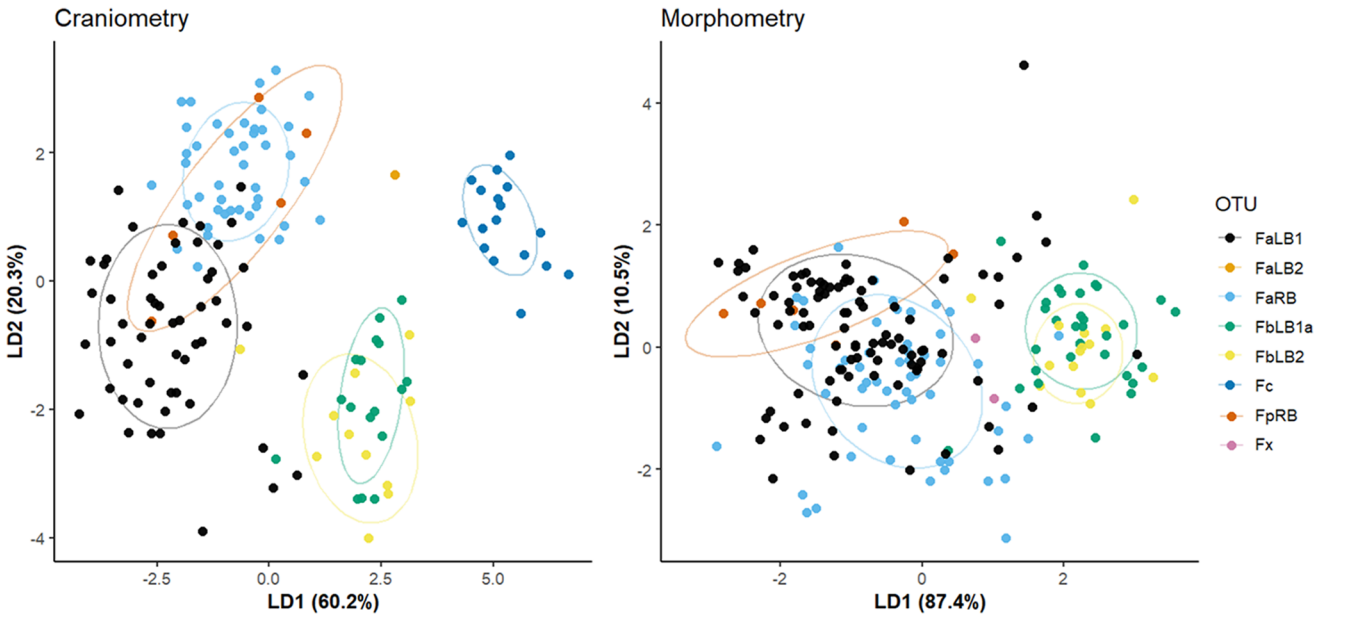


Fig. 6 Discriminant space of cranial and external morphometrics. Linear discriminant analysis (LDA) score plots showing separation among MOTUs based on (left) craniometric measurements (M-variables) and (right) external morphometrics (weight, head–body length, ear, foot, and tail). Points represent individual specimens and colors indicate mitochondrial MOTU assignments. Ellipses show 68% confidence regions (*t*-distribution) around group centroids. Axes are the first 2 discriminant roots (LD1 and LD2), with the percentage of discriminant variance explained indicated on each axis. Fx, previously undescribed *Funisciurus* species; Fp, *F. pyrropus*; Fb, *F.* cf. *bayonii*; Fa, *F. anerythrus*, Fc, *F.* cf. *congicus*; RB, right bank of Congo river; LB, left bank of Congo river.

Downloaded from academic.oup.com/jmammal/advance-article/doi/10.1093/jmammal/gyag084/8993869 by guest on 23 October 2026

at least 6 *Funisciurus* species in our dataset: *F. anerythrus*, 2 sympatric and noninterbreeding *F.* cf. *bayonii* taxa (FbLB1 and FbLB2), *F. pyrropus* (likely *F. p. akka* de Winton 1895), *F.* cf. *congicus* (based on craniometry only), and an as-yet unidentified *Funisciurus* species.

The inferred diversity of *Funisciurus* in the region depends strongly on the species-delimitation method applied. Although 4 rope squirrel species have historically been reported from our study area—*F. congicus* (Kuhl, 1820), *F. pyrropus* (F. Cuvier, 1833), *F. anerythrus* (Thomas, 1890), and *F. bayonii* (Bocage, 1890)—our least conservative delimitation (IQTREE, mPTP, or ASAP applied to mitochondrial data) recovers 10 MOTUs (Fig. 3): 4 that belong to *F. anerythrus* (FaRB, FaLB1, FaLB2, FaLB3); 1 *F. pyrropus* (Fp); 4 MOTUs that belong to a species that resembles *F.* cf *bayonii* (FbLB1a, FbLB1b, FbLB1c, and FbLB2); and 1 MOTU (Fx) that did not cluster closely with any available reference sequences of remaining *Funisciurus* species from Central Africa (*e.g.*, *F. leucogenys* or *F. lemniscatus*). This pattern was broadly consistent with pairwise *Cytb* Kimura 2-parameter (K2P) distances (Table 1). Under the reference framework proposed for mammals, *Cytb* distances below 2% are typically interpreted as intraspecific variation, values between 2% and 11% warrant additional evidence, and values above 11% are generally consistent with species-level divergence (Bradley and Baker 2001; Schrago and Mello 2020). In our context, MOTUs from *F. anerythrus* and *F. bayonii* 1 (FaLB1) would fall into the second category, whereas all other clearly suggest species-level divergence.

In contrast, our most conservative methods (mPTP or ASAP applied to nuclear data) recovered only 2 MOTUs: *F.* cf. *bayonii* and *F. anerythrus*. Under this scenario, *F. pyrropus* and Fx are included within the *F. anerythrus* clade, consistent with an interpretation as subspecies or geographically structured lineages rather than distinct species. In addition, craniometric and morphologic discriminant analyses suggest 4 separable OTUs: FaLB1, FaRB, FbLB, and Fc (for which we have no molecular data), whereas the limited number of measurable skulls for Fx, FaLB2, FbLB2, and Fp precludes conclusions regarding their cranial distinctiveness.

We consider the 4 mitochondrial *F. anerythrus* MOTUs as putative subspecies rather than distinct species, as the MOTUs are allopatrically separated by the Congo and Kasaï Rivers. Despite the cranial and external morphological differentiation observed in our dataset, this conservative approach is supported by nuclear *ApoB*, the K2P genetic distance, and by the fact that interbreeding cannot be ruled out if these river barriers were absent. A remaining challenge is to link the lineages identified here to named subspecies within the broadly distributed *F. anerythrus* complex. *Funisciurus anerythrus* was originally described from Buguera (Uganda) and is reported to occur widely across West and Central Africa, East Africa, and southern DRC (Thorington and Hoffmann 2005), including all major forest blocks of the central Congo Basin (Colyn 1991). In this context, it remains difficult to even confidently align our right-bank lineage (FaRB) with any currently recognized subspecies of *F. anerythrus* (*e.g.*, *F. a. anerythrus* Thomas, 1890; or *F. a. bandarum* Thomas, 1915) or with the synonymized name *F. a. niapu* Allen, 1922. At present, however, the lack of comparative cranial and/or sequence data from type or topotypic specimens precludes any further formal taxonomic assignment.

Interpreting the lineages assigned to *F.* cf. *bayonii* is even more challenging. Previous studies indicate that *F. bayonii* occurs mainly in south-western DRC and lacks described subspecies (Schouteden 1948; Nowak 1999; Thorington et al. 2012; Gerrie et al. 2016). It has also been characterized as a species of wooded savannah mosaics, occurring largely outside the Central African forest block (Thorington et al. 2012). In contrast, our data reveal 2 sympatric lineages (FbLB1 and FbLB2) that resemble *F. bayonii* yet occur at multiple evergreen-forest localities near the Congo River. Because genomic data for both *F. bayonii* and *F. congicus* voucher specimen are lacking, we cannot fully exclude that 1 of the *F.* cf *bayonii* taxa in our study actually represents *F. congicus*, a similar brown squirrel with a single white flank stripe that also occurs in the region. Nevertheless, we tentatively assigned both (FbLB1 and FbLB2) species to *F.* cf. *bayonii* complex because: (i) *F. congicus* specimens in our dataset are craniometrically clearly distinct from both FbLB1a and FbLB2 (Fig. 6); and (ii) head–body lengths of FbLB1 and FbLB2 are substantially larger than those reported for *F. congicus* in Mammals of Africa, and are more consistent with published measurements for *F. bayonii*. Finally, the strong mitochondrial divergence between the sympatrically occurring FbLB1 and FbLB2 supports treating them as separate species, as they have the theoretical opportunity to interbreed yet distinct mitochondrial genes. In contrast, the geographically structured sublineages within FbLB1 (FbLBa, FbLBb, and FbLBc) are sampled allopatrically, and are therefore more parsimoniously interpreted as subspecies.

Finally, we captured a species from south of the Congo River (specimen IDs: EBO87, YK125, MF577074) that does not cluster phylogenetically with any other *Funisciurus* species. Externally, these specimens resemble *F. lemniscatus*, notably in having 2 black lateral stripes separated by yellow on each flank **(**Fig. 4E). However, mitochondrial species–delimitation analyses (IQ-TREE, ASAP, mPTP) consistently place this lineage as a distinct, undescribed rope squirrel that is more closely related to *F. carruthersi* than to any other *Funisciurus* species currently represented in GenBank, including *F. lemniscatus*. Given that *F. carruthersi* is endemic to the Rwenzori Mountains, this relationship may be consistent with a scenario of ancient vicariance from a shared ancestor. Nevertheless, because our species does not resemble *F. carruthersi* morphologically, we refer to it here conservatively as *Funisciurus* sp. ("unidentified *Funisciurus*"). Overall, these patterns reinforce the need for a comprehensive taxonomic revision of the *Funisciurus* genus, as recently highlighted by Curaudeau et al. (2025).

Current IUCN rope squirrel distribution maps suggest that hydrological barriers play only a minor role in limiting rope squirrel distributions across the Congo Basin (Fig. 7, derived from the IUCN Red List; Cassola 2016a, 2016b; Gerrie et al. 2016). However, our results indicate that major rivers constitute strong barriers structuring *Funisciurus* ranges (Fig. 5). For example, our data suggest that *F. anerythrus* left bank (FaLB2) is restricted to areas southwest of the Kasaï river only (e.g., Kikwit), whereas FaLB1 presents its distribution only north of this river. Similarly, whereas the IUCN distribution map of *F. bayonii* is limited to south of the Kasaï river, the 2 sympatric MOTUs FbLB1a and FbLB2 are exclusively confined to the region between the Congo and the Kasaï rivers, with the Kasaï preventing southwestern expansion and the Congo limiting northeastern expansion. Likewise, the Congo River clearly partitions *F. anerythrus* into FaLB (restricted to the left bank) and FaRB (restricted to the right bank). *Funisciurus pyrropus* (Fp) is restricted to the eastern DRC (right bank of the Congo River) and the undescribed lineage Fx was detected only on the left bank of the Congo River (Fig. 5).

These patterns support the hypothesis that hydrological barriers play a pivotal role in promoting allopatric speciation in arboreal squirrels, as previously proposed for primates (Anthony et al. 2007; Harcourt and Wood 2012; Takemoto et al. 2017) and amphibians (Blackburn et al. 2021) in the Congo Basin, and for squirrels in Cameroon (Curaudeau et al. 2025). Similarly, studies on murid rodents and shrews suggest that the ecological barrier formed by the Congo River may limit gene flow between communities on opposite banks, likely facilitating divergence

Downloaded from academic.oup.com/jmammal/advance-article/doi/10.1093/jmammal/gyag084/8993869 by guest on 23 October 2026

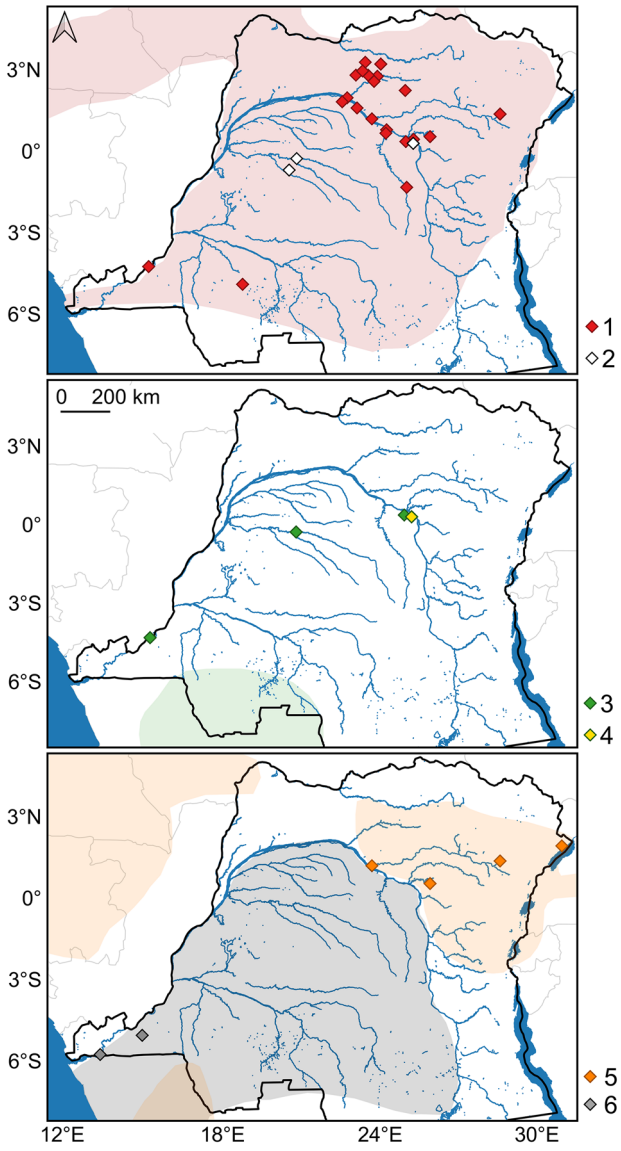


Fig. 7 Distribution of African rope squirrels in the Democratic Republic of the Congo related to novel findings. Square points indicate capture locations from the present study for: 1, *Funisciurus anerythrus* (red); 2, an as-yet unidentified *Funisciurus* species (white); 3, *F.* cf. *bayonii* 1 (green); 4, *F.* cf. *bayonii* 2 (yellow); 5, *F. pyrropus* (orange); 6, *F.* cf. *congicus* (grey). Colored polygons represent the known species ranges for the corresponding taxa, as reported by the IUCN Red List of Threatened Species, version 2025-2 (https://www.iucnredlist.org). Basemap imagery was obtained from Esri World Imagery.

and speciation (Katuala et al. 2008; Kennis et al. 2011). Comparable effects have also been reported for other rivers including the Lomami and Kasaï, which have been used to subdivide the southern Congo faunal region into distinct subregions (Colyn 1991). These observations indicate that IUCN distribution maps of rope squirrels in the Congo Basin could benefit from updates to better reflect potential river–associated structuring revealed by our dataset.

This study has several limitations that should be considered when interpreting our results. First, although we sampled many localities, sampling effort was uneven across the Congo Basin. As a consequence, we may underestimate *Funisciurus* diversity and we lack the resolution to evaluate barriers beyond the Congo and Kasaï Rivers. We therefore encourage the broader community to make morphological measurements and DNA sequences available for squirrels collected in southern DRC, Angola, and other undersampled parts of the Congo Basin. Second, samples were collected over more than 4 decades, across large distances, and were occasionally subject to variable field preservation, transport, and storage conditions. This pattern inevitably reduced the number of specimens with complete morphometric/craniometric datasets and resulted in heterogeneity in sequence length and quality. Despite these constraints, our dataset remains one of the most comprehensive assembled for Central African *Funisciurus*, including >397 *Cytb* sequences, 62 *ApoB* sequences, and 220 measured skulls, providing a robust foundation for the integrative patterns reported here. Finally, resolving the taxonomic status of several lineages will require targeted comparisons with name-bearing material. In particular, museum collections (including holotype and paratype specimens) represent a critical resource to link genetic lineages to formal taxonomic names and to reassess historically synonymized taxa. Where feasible, obtaining cranial measurements and generating DNA data from museum material should be prioritized to support a definitive revision of *Funisciurus* taxonomy and distribution.

In conclusion, our integrative morphometric and molecular analyses reveal substantial cryptic diversity in Congo Basin rope squirrels and support the presence of at least 6 *Funisciurus* species. In an integrated taxonomic framework, our results are most consistent with *F. anerythrus* comprising several allopatric, subspecies-level lineages (FaRB, FaLB1, FaLB2, FaLB3); whereas *F.* cf. *bayonii* comprises 2 sympatric, genetically distinct, and likely noninterbreeding species (FbLB1 and FbLB2). We further document *F. p. akka* de Winton, 1895 (restricted in our dataset to the right bank of the Congo River), identify a previously unidentified species (Fx) occurring exclusively on the left bank, and recover *F.* cf. *congicus* based on craniometric evidence. Across taxa, the Congo and Kasaï Rivers emerge as major hydrological barriers that sharply delimit (sub)species distributions. Because mpox virus has previously been detected in 2 specimens included here (*F. anerythrus* and *F.* cf. *bayonii*), our results provide an ecological and taxonomic baseline for interpreting phylogeographic host–virus associations. Improved resolution of squirrel taxonomy, phylogeography, and habitat use will therefore be important for future One Health surveillance studies of mpox virus–host relationships, and for assessing the potential contribution of *Funisciurus* and other arboreal Protoxerini (e.g., *Paraxerus*) to zoonotic spillover risk.

Acknowledgments

We thank the Faculty of Science and the Centre de Surveillance de la Biodiversité at the University of Kisangani for their supervision, and Emmanuel Gilissen and Marthys Rotonda for access to squirrel specimens from the MRAC-Tervuren collections.

Author contributions

Pascal Likangalele Baelo (Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Supervision, Writing—original draft, Writing—review & editing), Léa Fourchault (Conceptualization, Data curation, Formal analysis, Methodology, Writing—original draft, Writing—review & editing), Ann Ceulemans (Data curation, Formal analysis, Writing—review & editing), Nicolas Laurent (Data curation, Formal analysis, Investigation, Writing—review & editing), Claude Mande (Data curation, Investigation, Writing—review & editing), Anne Laudisoit (Conceptualization, Data curation, Investigation, Writing—review & editing), Nicaise Amundala (Data curation, Writing—review & editing), Ganiyat Temidayo Saliu (Data curation, Validation, Writing—review & editing), Herwig Leirs (Funding acquisition, Writing—review & editing),

Downloaded from academic.oup.com/jmammal/advance-article/doi/10.1093/jmammal/gyag084/8993869 by guest on 23 October 2026

Guy-Crispin Gembu (Data curation, Funding acquisition, Investigation, Supervision, Validation, Writing—review & editing), Sophie Gryseels (Formal analysis, Funding acquisition, Validation, Visualization, Writing—review & editing), Jan Hulselmans (Formal analysis, Methodology, Supervision, Validation, Visualization, Writing—review & editing), Erik Verheyen (Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Supervision, Validation, Writing—review & editing), and Joachim Mariën (Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Software, Supervision, Validation, Visualization, Writing—original draft, Writing—review & editing)

Supplementary data

Supplementary data are available at *Journal of Mammalogy* online.

Supplementary Data SD1. List of specimens examined and their sampling sites, including specimens from which skulls were extracted and those that underwent genetic analysis.

Supplementary Data SD2. Morphometric data of *Funisciurus* specimens examined, including ear, head-body, hind foot length (mm), and body weight (g).

Supplementary Data SD3. Craniometric data (M1 to M23) of *Funisciurus* specimens examined: M1, maximum length of the skull; M2, condilo-basal length; M3, henselion-basion length; M4, henselion-palation length; M5, length of the cleft palate; M6, length of the diastema; M7, distance between the anterior edge of the alveolus of M1 and the cutting edge of the upper incisor; M8, width of the interorbital narrowing; M9, width of the zygomatic arch; M10, minimum width of the palate; M11, length of the upper molar row; M12, external width of upper molar row at M1; M13, maximum molar width; M14, minimum width of zygomatic plate; M15, maximum width of nasals; M16, maximum length of nasals; M17, length of lower molar row; M18, length of the tympanic bulla; M19, maximum width of the cranial cavity; M20, depth of the upper incisors; M21, height of the rostrum at the anterior edge of the alveolus of M1; M22, maximum width of the rostrum; M23, distance between the extreme points of the coronoid and angular processes.

Supplementary Data SD4. Descriptive statistics including sample size (*n*), mean, and SD for craniometric measurements (M1–M23) of examined *Funisciurus* specimens, summarized by the delineated species groups. Measurements are defined as follows: M1, maximum skull length; M2, condylobasal length; M3, henselion–basion length; M4, henselion–palation length; M5, length of the cleft palate; M6, diastema length; M7, distance between the anterior edge of the alveolus of M1 and the cutting edge of the upper incisor; M8, interorbital constriction width; M9, zygomatic arch width; M10, minimum palatal width; M11, upper molar row length; M12, external width of upper molar row at M1; M13, maximum molar width; M14, minimum width of the zygomatic plate; M15, maximum nasal width; M16, maximum nasal length; M17, lower molar row length; M18, tympanic bulla length; M19, maximum width of the cranial cavity; M20, upper incisor depth; M21, rostrum height at the anterior edge of the alveolus of M1; M22, maximum rostrum width; M23, distance between the extreme points of the coronoid and angular processes. All measurements are reported in millimeters (mm).

Supplementary Data SD5. Descriptive statistics including sample size (*n*), mean, and SD for morphometric measurements of examined *Funisciurus* specimens, summarized by the delineated species groups. Body mass is reported in grams (g); all linear measurements (head–body length, ear length, hind foot length, and tail length) are reported in millimeters (mm). "NA" indicates that the SD could not be calculated.

Supplementary Data SD6. Maximum-likelihood phylogeny of Protoxerini based on cytochrome *b* (mitochondrial DNA) sequences from all taxa included in this study. The tree was inferred in IQ-TREE, with node support estimated from 10,000 bootstrap replicates. *Tamias* was used as the outgroup. Node labels indicate SH-aLRT support (%) and ultrafast bootstrap support (%), respectively.

Supplementary Data SD7. Maximum-likelihood phylogeny of Protoxerini based on apolipoprotein *b* sequences (nuclear) sequences from all taxa included in this study. The tree was inferred in IQ-TREE, with node support estimated from 10,000 bootstrap replicates. *Tamias* was used as the outgroup. Node labels indicate SH-aLRT support (%) and ultrafast bootstrap support (%), respectively.

Supplementary Data SD8. Pairwise multivariate differentiation in craniometrics among OTUs. Pairwise comparisons were performed using *Wilks*' λ from a MANOVA fitted on principal component scores derived from craniometric variables (M1-M23). For each OTU pair, the table reports the total sample size (*n*), the number of retained principal components (k), *Wilks*' λ, the approximate *F* statistic, the raw *P* value, and the adjusted *P* value (padj).

Supplementary Data SD9. Pairwise multivariate differentiation in morphometrics among OTUs. Pairwise comparisons were performed using *Wilks*' λ from a MANOVA fitted on principal component scores derived from morphometric variables. For each OTU pair, the table reports the total sample size (*n*), the number of retained principal components (k), *Wilks*' λ, the approximate *F* statistic, the raw *P* value, and the adjusted *P* value (padj).

Supplementary Data SD10. Results of PERMANOVA test to check for sexual dimorphism in craniometric and morphometric measurements of different *Funisciurus* species.

Supplementary Data SD11. MAFFT alignment of all cytochrome *b* sequences used to make the mitochondrial phylogenetic tree.

Supplementary Data SD12. MAFFT alignment of all apolipoprotein *b* sequences used to make the nuclear phylogenetic tree.

Funding

This research was supported by the Flemish Interuniversity Council South Initiative (VLIR-SI; to A.L. and E.V.), the Flemish Interuniversity Council (VLIR-CUI; to G.-C.G. and E.V.), the Research Foundation–Flanders (FWO grants G096222N to J.M. and H.L., and G051322N to S.G.), a FED-tWIN scholarship funded by the Belgian Federal Government (Prf-2019-prf004_OMEga; to S.G.), and the BiodivERsA3 ERA-Net COFUND program under the 2018–2019 BiodivERsA joint call (grant ANR-19-EBI3-0004). Additional support was provided by the CEBioS program of the Royal Belgian Institute of Natural Sciences for CSB fieldwork (P.B., C.M., G.-C.G.) and research stays in Belgium (P.B.); as well as by the Department of Work, Economy, Science, Innovation and Social Economy of the Flemish Government (WEWIS) and the DGD FA5 Synergy Fund to the Institute of Tropical Medicine Antwerp.

Conflict of interest

The authors declare no conflict of interest.

Data availability

The datasets generated and/or analyzed during this study will be made available for research purposes upon reasonable request to the corresponding authors. Sequence data will be deposited in the European Nucleotide Archive (ENA) upon acceptance of the manuscript under project accessions PRJEB112614 and PRJEB101311. The alignments used for phylogenetic analyses are also provided in the Supplementary Information: cytochrome *b* (Supplementary Data SD11) and apolipoprotein *b* (Supplementary Data SD12).

Downloaded from academic.oup.com/jmammal/advance-article/doi/10.1093/jmammal/gyag084/8993869 by guest on 23 October 2026

References

Anthony NM, Johnson-Bawe M, Jeffery K, Clifford SL, Abernethy KA, Tutin CE, Lahm SA, White LJT, Utley JF, Wickings EJ, et al. 2007. The role of Pleistocene refugia and rivers in shaping gorilla genetic diversity in central Africa. Proceedings of the National Academy of Sciences 104(51):20432–20436. https://doi.org/10.1073/pnas.0704816105

Baelo P, Asimonyio J, Amundala N, Akaibe D, Gambalemoke S, Erik V, Anne L, Bongo G, Koto-te-Nyiwa Ngbolua K. 2018. Preliminary inventory of squirrels (Sciuridae, Rodentia) of Kisangani Region in the Democratic Republic of the Congo. American Journal of Zoology 1(1):15–19.

Bernd Hoffmann DVM, Tappe D, Höper D, Herden C, Boldt A, Mawrin C, Niederstraßer O, Müller T, Jenckel M, van der Grinten E, et al. 2015. A variegated squirrel bornavirus associated with fatal human encephalitis. New England Journal of Medicine 373(2):154–162. https://doi.org/10.1056/NEJMoa1415627

Blackburn DC, Nielsen SV, Ghose SL, Burger M, Gonwouo LN, Greenbaum E, Gvoždík V, Hirschfeld M, Kouete MT, Kusamba C, et al. 2021. 2021. Phylogeny of African long-fingered frogs (Arthroleptidae: Cardioglossa) reveals recent allopatric divergences in coloration. Ichthyology and Herpetology 109(3):728–742. https://doi.org/10.1643/h2020165

Bradley RD, Baker RJ. 2001. A test of the genetic species concept: cytochrome-b sequences and mammals. Journal of Mammalogy 82(4):960–973. https://doi.org/10.1644/1545-1542(2001)082<0960:ATOTGS>2.0.CO;2

Cassola F. 2016a. *Funisciurus anerythrus*. The IUCN red list of threatened species 2016: e.T8756A22269868. [accessed 11 Nov 2025]. http://dx.doi.org/10.2305/IUCN.UK.2016-3.RLTS.T8756A22269868.en

Cassola F. 2016b. *Funisciurus congicus*. The IUCN red list of threatened species 2016: e.T8758A115088669. Gland, Switzerland: International Union for Conservation of Nature (IUCN) [accessed 11 Nov 2025]. https://dx.doi.org/10.2305/IUCN.UK.2016-3.RLTS.T8758A22270249.en

Chen N, Li G, Liszewski MK, Atkinson JP, Jahrling PB, Feng Z, Schriewer J, Buck C, Wang C, Lefkowitz EJ, et al. 2005. Virulence differences between monkeypox virus isolates from West Africa and the Congo Basin. Virology Journal 340(1):46–63. https://doi.org/10.1016/j.virol.2005.05.030

Colyn M. 1991. L'importance zoogéographique du bassin du fleuve Zaïre pour la spéciation: le cas des primates simiens. Annales-Musée royal de l'Afrique Centrale-Sciences zoologiques no. 264.

Curaudeau M, Besombes C, Nakouné E, Fontanet A, Gessain A, Hassanin A. 2023. Identifying the most probable mammal reservoir hosts for monkeypox virus based on ecological niche comparisons. Viruses 15(3):727. https://doi.org/10.3390/v15030727

Curaudeau M, Peterhans JK, Flanche T, Gilissen E, Granjon L, Barale L, Gessain A, Hassanin A. 2025. Squirrel reservoirs of monkeypox virus are sister species separated by the Sanaga River (Cameroon), as are the two main viral clades. One Health 21:101157. https://doi.org/10.1016/j.onehlt.2025.101157

Demos TC, Agwanda B, Hickerson MJ. 2014. Integrative taxonomy within the Hylomyscus denniae complex (Rodentia: Muridae) and a new species from Kenya. Journal of Mammalogy 95(1):E1–E15. https://doi.org/10.1644/13-MAMM-A-268

Dubey S, Salamin N, Ohdachi SD, Barrière P, Vogel P. 2007. July 2007. Molecular phylogenetics of shrews (Mammalia: Soricidae) reveal timing of transcontinental colonizations. Molecular Phylogenetics and Evolution 44(1):126–137. https://doi.org/10.1016/j.ympev.2006.12.002

Emmons LH. 1980. Ecology and resource partitioning among nine species of African rain forest squirrels. Ecological Monographs 50(1):31–54. https://doi.org/10.2307/2937245

Fa JE, Peres CA, Meeuwig J. 2002. Bushmeat exploitation in tropical forests: an intercontinental comparison. Conservation Biology 16(1):232–237. https://doi.org/10.1046/j.1523-1739.2002.00275.x

Fourchault L, Lamane A, Romaric NMD, Saliu GT, Gryseels S, Verheyen E, Dahdouh-Guebas F, Kreppel K. 2025. Public health risks of traditional zootherapeutic practices in Africa. One Health 21:101178. https://doi.org/10.1016/j.onehlt.2025.101178

Gerrie R, Kennerley R, Koprowski RJ. 2016. Funisciurus bayonii. The IUCN red list of threatened species. http://dx.doi.org/10.2305/IUCN.UK.2016-3.RLTS.T8757A22270331

Guindon S, Dufayard J-F, Lefort V, Anisimova M, Hordijk W, Gascuel O. 2010. New algorithms and methods to estimate maximum-likelihood phylogenies: assessing the performance of PhyML 3.0. Systematic Biology 59(3):307–321. https://doi.org/10.1093/sysbio/syq010

Harcourt AH, Wood MA. 2012. Rivers as barriers to primate distributions in Africa. International Journal of Primatology 33(1):168–183. https://doi.org/10.1007/s10764-011-9558-z

Hoang DT, Chernomor O, Haeseler A, Minh BQ, Vinh LS. 2018. UFBoot2: improving the ultrafast bootstrap approximation. Molecular Biology and Evolution 35(2):518–522. https://doi.org/10.1093/molbev/msx281

Hoffmann B, Tappe D, Höper D, Herden C, Boldt A, Mawrin C, Niederstraßer O, Müller T, Jenckel M, van der Grinten E, et al. 2015. A variegated squirrel bornavirus associated with fatal human encephalitis. New England Journal of Medicine 373(2):154–162. https://doi.org/10.1056/NEJMoa1415627

Kapli P, Lutteropp S, Zhang J, Kobert K, Pavlidis P, Stamatakis A, Flouri T. 2017. Multi-rate Poisson tree processes for single-locus species delimitation under maximum likelihood and Markov chain Monte Carlo. Bioinformatics 33(11):1630–1638. https://doi.org/10.1093/bioinformatics/btx025

Katuala P, Kennis J, Nicolas V, Wendelen W, Hulselmans J, Verheyen E, Van Houtte N, Dierckx T, Dudu A, Leirs H. 2008. The presence of Praomys, Lophuromys, and Deomys species (Muridae, Mammalia) in forest blocks separated by the Congo River and its tributaries (Kisangani region, Democratic Republic of Congo). Mammalia 72(3):223–228. https://doi.org/10.1515/MAMM.2008.044

Kennis J, Nicolas V, Hulselmans J, Katuala GBP, Wendelen W, Verheyen E, Dudu A, Leirs H. 2011. The impact of the Congo River and its tributaries on the rodent genus Praomys: speciation origin or range expansion limit? Zoological Journal of the Linnean Society 163(3):983–1002. https://doi.org/10.1111/j.1096-3642.2011.00733.x

Khodakevich L, Szczeniowski M, Manbu-ma-Disu M, Jezek Z, Marennikova S, Nakano J, Messinger D. 1987. The role of squirrels in sustaining monkeypox virus transmission. Tropical Geographical Medicine 39(2):115–122. https://doi.org/PMID: 2820094

Kinganda Lusamaki E, Amuri AA, Nuñez FN, Makangara JC, Pratt C, Vakaniaki EH, Hoff NA, Luakanda GN, Sabiti SN, Mulopo NM, et al. 2025. Clade I mpox virus genomic diversity in the Democratic Republic of the Congo, 2018–2024: predominance of zoonotic transmission. Cell Journal https://doi.org/10.1016/j.cell.2024.10.017

Kingdon J, Happold D. 2013. Mammals of Africa: volume 3: rodents, hares and rabbits. London: Bloomsbury Publishing Plc. https://doi.org/10.5860/choice.50-4188

Miralles A, Ducasse J, Brouillet S, Flouri T, Fujisawa T, Kapli P, Knowles LL, Kumari S, Stamatakis A, Sukumaran J, et al. 2022. SPART: a versatile and standardized data exchange format for species partition information. Molecular Ecology Resources 22(1):430–438. https://doi.org/10.1111/1755-0998.13470

Monadjem A, Taylor PJ, Denys C, Cotterill FPD. 2015. Family Sciuridae. In: Monadjem, A, Taylor PJ, Denys C, Cotterill FPD, editors. Rodents of sub-Saharan Africa: a biogeographic and taxonomic synthesis. Berlin (Germany): De Gruyter; p. 62–146.

Montgelard C, Sophie B, CT, Olivier, V, François, M. 2002. Systématique moléculaire de Sciurognathi (Rodentia): les gènes mitochondriaux du cytochrome b et de l'ARNr 12S soutiennent les Anomaluroidea (Pedetidae et Anomaluridae). Molecular Phylogenetics and Evolution 22(2):220–233. https://doi.org/10.1006/mpev.2001.1056

Downloaded from academic.oup.com/jmammal/advance-article/doi/10.1093/jmammal/gyag084/8993869 by guest on 23 October 2026

Nakazawa Y, Mauldin MR, Emerson GL, Reynolds MG, Lash RR, Gao J, Zhao H, Li Y, Muyembe J-J, Kingebeni PM, et al. 2015. A phylogeographic investigation of African monkeypox. Viruses 7(4):2168–2184. https://doi.org/10.3390/v7042168

Nesi N, Nakouné E, Cruaud C, Hassanin A. 2011. DNA barcoding of African fruit bats (Mammalia, Pteropodidae). The mitochondrial genome does not provide a reliable discrimination between *Epomophorus gambianus* and *Micropteropus pusillus*. Comptes Rendus Biologies 334(7):544–554. https://doi.org/10.1016/j.crvi.2011.05.003

Nguyen LT, Schmidt HA, von Haeseler A, Minh BQ. 2015. IQ-TREE: a fast and effective stochastic algorithm for estimating maximum likelihood phylogenies. Molecular Biology and Evolution 32(1):268–274. https://doi.org/10.1093/molbev/msu300

Nowak RM. 1999. Family Sciuridae. In: Nowak RM, editor. Walker's mammals of the world. 6th ed. Vol. 2. Baltimore (MD, USA): Johns Hopkins University Press; p. 1246–1306.

Paradis E, Schliep K. 2019. ape 5.0: an environment for modern phylogenetics and evolutionary analyses in R. Bioinformatics 35(3):526–528. https://doi.org/10.1093/bioinformatics/bty633

Puillandre N, Brouillet S, Achaz G. 2021. ASAP: assemble species by automatic partitioning. Molecular Ecology Resources 21(2):609–620. https://doi.org/10.1111/1755-0998.13281

Rimoin AW, Alfonso VH, Hoff NA, Doshi RH, Mulembakani P, Kisalu NK, Muyembe J-J, Okitolonda EW, Wright LL. 2017. Human exposure to wild animals in the Sankuru Province of the Democratic Republic of the Congo. EcoHealth 14(3):552–563. https://doi.org/10.1007/s10393-017-1262-9

Riutord-Fe C, Schlotterbeck J, Lagostina L, Kouadio L, Herridge HR, Jochum MJS, Noma NY, López-Morales A, Hoffmann D, Calvelage S, et al. 2026. Transmission of MPXV from fire-footed rope squirrels to sooty mangabeys. Nature 651(8104):185–190. https://doi.org/10.1038/s41586-025-10086-y

Schouteden H. 1948. Faune du Congo Belge et du Ruanda-Urundi. Vol. 1. Mammifères. Tervuren (Belgium): Musée du Congo Belge; 331 p. (Annales du Musée du Congo Belge, Série in-8°, Sciences zoologiques; vol. 1).

Schrago CG, Mello B. 2020. Employing statistical learning to derive species-level genetic diversity for mammalian species. Mammal Review 50(3):240–251. https://doi.org/10.1111/mam.12192

Schulze V, Lurz PW, Ferrari N, Romeo C, Steele MA, Marino S, Vittoria MM, Calvignac SS, Schlottau K, Beer M, et al. 2020. Search for polyoma-, herpes-, and bornaviruses in squirrels of the family Sciuridae. Virology Journal 17(1):42. https://doi.org/10.1186/s12985-020-01310-4

Sikes RS, The Animal Care and Use Committee of the American Society of Mammalogists. 2016. 2016 guidelines of the American Society of Mammalogists for the use of wild mammals in research and education. Journal of Mammalogy 97(3):663–688. https://doi.org/10.1093/jmammal/gyw078

Sklenovská N, Van Ranst M. 2018. Emergence of monkeypox as the most important orthopoxvirus infection in humans. Frontiers in Public Health 6(6):241. https://doi.org/10.3389/fpubh.2018.00241

Sloan DB, Havird JC, Sharbrough J. 2017. The on-again, off-again relationship between mitochondrial genomes and species boundaries. Molecular Ecology 26(8):2212–2236. https://doi.org/10.1111/mec.13959

Steiner M, Huettmann F. 2023. Taxonomy for the squirrels of the world: hairy classifications and conservation category games. In: Steiner, M, Huettmann, F, editors. Sustainable squirrel conservation: a modern reassessment of family sciuridae. Cham (Switzerland): Springer; p. 3–90. https://doi.org/10.1007/978-3-031-23547-4_1

Takemoto H, Kawamoto Y, Furuichi T. 2017. The formation of the Congo River and the origin of bonobos: a new hypothesis. In: Hare B, Yamamoto S, editors. Bonobos: unique in mind, brain, and behavior. Oxford (UK): Oxford University Press; p. 16. [accessed 11 Nov 2025]. https://doi.org/10.1093/oso/9780198728511.003.0016

Thorington JR, Koprowski JL, Steele MA, Whatton JF. 2012. Squirrels of the world. Baltimore (MD): Johns Hopkins University Press.

Thorington RWJr, Hoffmann RS. 2005. Family Sciuridae. In: Wilson DE, Reeder DM, editors. Mammal species of the world: a taxonomic and geographic reference. 3rd ed. Vol. 2. Baltimore (MD, USA): Johns Hopkins University Press; p. 754–818.

Verheyen W, Bracke E. 1966. The influence of aging on the craniometrical characters of Praomys Jacksoni (De Winton 1897). Annales du musée royal de l'Afrique Centrale de tervuren 144:91–101.

Zachos FE, Burgin CJ, Wilson DE, Mittermeier RA, Rylands AB, Lacher TE, Sechrest W. (chief editors). 2021. Illustrated checklist of the mammals of the world. Mammalian Biology 101(1):125–126. https://doi.org/10.1007/s42991-020-00088-w

Downloaded from academic.oup.com/jmammal/advance-article/doi/10.1093/jmammal/gyag084/8993869 by guest on 23 October 2026

© The Author(s) 2026. Published by Oxford University Press on behalf of the American Society of Mammalogists.
This is an Open Access article distributed under the terms of the Creative Commons Attribution License (https://creativecommons.org/licenses/by/4.0/), which permits unrestricted reuse, distribution, and reproduction in any medium, provided the original work is properly cited. Journal of Mammalogy 2026 00(00) 1–11
https://doi.org/10.1093/jmammal/gyag084
Research Article