Does the chain of Eastern Arc Mountains impede gene flow? Genetic evidence from the African savanna elephant and the Masai giraffe in Tanzania

George Lohay[1,2*] ()[§], **David Pearce**[3] (), **Monica L. Bond**[4,5] (), **Alex Lobora**[6] (),
Derek E. Lee[1,4] () & **Douglas Cavener**[1] ()

[1]*Department of Biology, The Pennsylvania State University, University Park, PA, U.S.A.*

[2]*Research and Innovation for the Serengeti Ecosystem, Grumeti Fund, Mara, Tanzania*

[3]*Department of Fisheries, Wildlife, and Conservation Sciences, Oregon State University, Corvallis, OR, U.S.A.*

[4]*Wild Nature Institute, Concord, NH, U.S.A.*

[5]*Department of Evolutionary Biology and Environmental Studies, University of Zurich, Zurich, Switzerland*

[6]*Ministry of Natural Resources and Tourism, Dodoma, Tanzania*

Received 10 February 2025, To authors for revision 12 March 2025, Accepted 12 April 2025

Abstract

An increase in the human population in recent years poses threats to the conservation of wildlife species. The expansion of human settlement and agricultural activities has led to the loss of wildlife corridors. Apart from the influence of humans on the gene flow of species between protected areas, natural features such as mountains, rivers, and hills can act as a barrier to gene flow. The African savanna elephant (*Loxodonta africana*) and the Masai giraffe (*Giraffa tippelskirchi*) are widely distributed in Tanzania. However, the Eastern Arc Mountains (EAM) and the Gregory Rift Valley (GRV) systems influence gene flow for some species. We conducted a study in southeastern Tanzania covering three major ecosystems: Ruaha–Rungwa, Katavi–Rukwa, and Selous–Mikumi to determine whether there is genetic differentiation between these ecosystems for giraffes and elephants. We analysed the mitochondrial DNA of 450 elephants and 100 giraffes. Our results show that (1) there is high genetic differentiation between populations found east and west of the EAM for both elephants and giraffes, (2) there is no female-mediated gene flow between these populations, (3) Populations found west of the EAM show high genetic connectivity suggesting historical gene flow between them, and (4) elephant populations from Ruaha share haplotypes with both Tarangire and Serengeti ecosystems suggesting historical connectivity between them. Our study reveals that the EAM plays a significant role in the gene flow of these species. However, the recent loss of miombo forests between Ruaha and Katavi, owing to anthropogenic activities, may reduce gene flow in the long run.

Keywords: Eastern Arc Mountains, genetic connectivity, mitochondrial DNA, conservation genetics, Masai giraffe, African savanna elephants.

Introduction

Habitat loss and fragmentation due to land conversion pose significant threats to biodiversity (Fahrig, 2013). To ensure successful conservation efforts, it is essential to maintain genetic connectivity through wildlife corridors (Noss *et al.*, 2012; Taylor, Fahrig & Merriam, 1993). While human activities are a primary driver of fragmentation (Newmark, 2008; Riggio, Jacobson, Hijmans & Caro, 2019), natural barriers such as rivers and mountains can also impede gene flow (Evans, Bliss, Mendel & Tinsley, 2011; Lohay, Weathers, Estes, McGrath & Cavener, 2020; Lohay *et al.*, 2023). Increased isolation can lead to genetic drift, inbreeding, and a higher risk of extinction. Conversely, maintaining connectivity can enhance population viability (Allendorf, Hohenlohe & Luikart, 2010).

In Tanzania, the Ruaha–Rungwa and Selous–Mikumi ecosystems are home to the country's largest elephant (*Loxodonta africana*) populations (Jones *et al.*, 2018; Mkuburo, Nahonyo, Smit, Jones & Kohi, 2020). Genetic studies reveal that habitat fragmentation has limited gene flow, particularly among younger elephants in the Ruaha–Rungwa ecosystems (Lobora *et al.*, 2018). The Eastern Arc Mountains (EAM) also restrict move-

*To whom correspondence should be addressed.
E-mail: georgel@grumetifund.org

ISSN 2410-8200 [Online only] — DOI: https://doi.org/10.3957/056.055.0408

ment for species like lions (*Panthera leo*) and sable antelope (*Hippotragus niger*) (Pitra, Hansen, Lieckfeldt & Arctander, 2002; Smitz *et al*., 2018). Moreover, the movement of giraffes between Selous and Ruaha is affected by the terrain slope (Crowhurst, Mullins, Mutayoba & Epps, 2013), and the patterns of elephant connectivity have shifted from being terrain-driven to being influenced by human settlements (Epps, Mutayoba, Gwin & Brashares, 2011).

This study investigates the genetic connectivity of African savanna elephants and Masai giraffes (*Giraffa tippelskirchi*) to evaluate how the EAM impact gene flow and genetic differentiation among the Selous–Nyerere, Ruaha–Rungwa, and Katavi–Rukwa ecosystems. We hypothesize that the EAM acts as a barrier, resulting in significant genetic differentiation between populations in these ecosystems. However, we also consider the possibility that some levels of genetic connectivity may still exist, indicating either historical or contemporary movement across the landscape.

We predict that genetic differentiation (F_{ST} values) will be higher among populations separated by the EAM compared to those within the same ecosystem. Furthermore, we expect mitochondrial DNA analyses to reveal distinct population clusters corresponding to geographic regions, reflecting limited gene flow. Conversely, if connectivity is present, individuals from different populations may share mitochondrial DNA haplotypes and exhibit lower genetic differentiation, suggesting ongoing movement between ecosystems despite potential barriers.

Methods

This study explores genetic connectivity in three Tanzanian ecosystems: Nyerere–Selous, Ruaha–Rungwa, and Katavi–Rukwa. The EAM may restrict species movement between Nyerere–Selous and Ruaha–Rungwa. The Ruaha–Rungwa ecosystem, which includes Ruaha National Park and Rungwa Game Reserve, is linked to Katavi–Rukwa through open areas that are not legally protected. The Rufiji River acts as a barrier for Masai giraffes, while increasing human activity and miombo woodland loss threaten wildlife movement (Lobora *et al*., 2018).

The research utilizes mtDNA analysis to complement previous microsatellite studies (Crowhurst *et al*., 2013; Lohay *et al*., 2020). Elephant faecal samples were collected from 2015 to 2017 and preserved in a buffer solution (Ahlering, Hailer, Roberts & Foley, 2011). Blood samples from the Tanzania Wildlife Research Institute (TAWIRI) were stored at –80°C, with DNA extraction performed using the Monarch Nucleic Acid Purification Kit (New England Biolabs). Giraffe faecal samples from Nyerere and Ruaha National Parks were collected between 2020 and 2022, following Lohay *et al*. (2023). DNA extraction was performed using established protocols for elephants (Lobora *et al*., 2018; Lohay *et al*., 2020) and (Lohay *et al*., 2023) for the Masai giraffes. PCR amplification and Sanger sequencing was done using established protocols (Lohay *et al*., 2020, 2023). To assess significant genetic differentiation between populations, we used *F*-statistics implemented in Arlequin version 3.5 (Excoffier & Lischer, 2010), with a significance threshold of $P \leq 0.05$. F_{ST} values closer to 1 indicate greater genetic divergence and less interbreeding.

The mitochondrial DNA (mtDNA) haplotypes obtained in this study were compared with previously published haplotypes, all of which clustered into three subclades: East-Central, Savanna-Wide, and Southeast Savanna (Lohay *et al*., 2020). These subclades were identified through the construction of a neighbour-joining phylogenetic tree using MEGA software (Kumar, Nei, Dudley & Tamura, 2008).

Results

We analysed 450 elephant and 100 giraffe mtDNA samples. We identified 22 elephant haplotypes and five Masai giraffe haplotypes. Only one haplotype was shared between elephant populations on either side of the EAM. Nyerere elephants had Southeast. No haplotypes were shared between the Nyerere and Ruaha Masai giraffe populations (Fig. 1).

Most elephants in the Eastern Arc Mountains (EAM) carry haplotypes common in Tanzania, including Serengeti and Tarangire. Rungwa elephants share haplotypes with those in Serengeti. Elephants from Nyerere and Selous exclusively carry haplotypes of the Southeast-savanna (SS) subclade, while nearly 50% of those in Katavi National Park (KNP) also have SS-related haplotypes. F_{ST} values between Selous (SGR)/Nyerere (NNP) and populations west of the EAM indicated limited female-mediated gene flow, with the highest value between Selous and Lukwati (*e.g*. RNP and PGR, $F_{ST} = 0.845$). In contrast, Ruaha (RNP) had the lowest F_{ST} values with Lukwati and Pigi (Fig. 2, $F_{ST} = 0.010$).

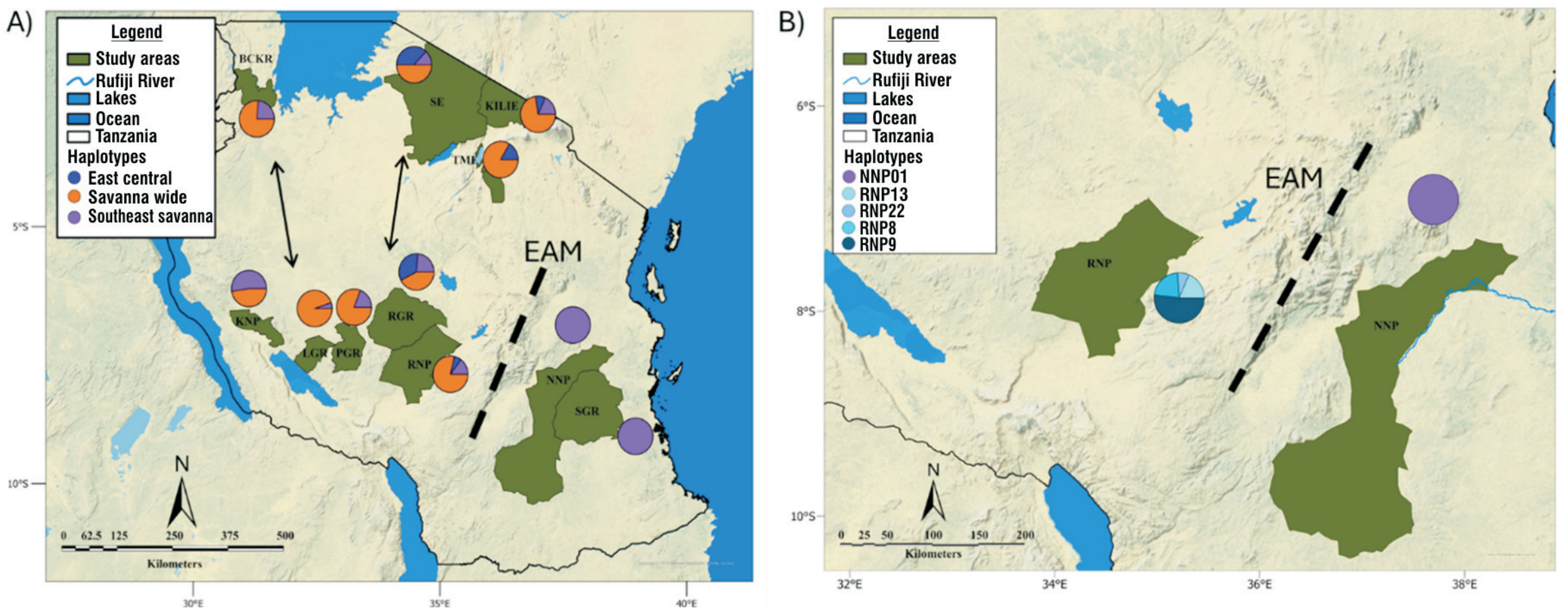

Fig. 1. Maps showing mitochondrial DNA (mtDNA) haplotype distribution in savanna elephants and Masai giraffes. (**A**) Elephants from Selous and Nyerere carried only Southeast-savanna (SS) subclade haplotypes, while those from Ruaha–Rungwa and Katavi–Rukwa had East-Central and Savanna-wide subclade haplotypes. (**B**) Masai giraffes in Nyerere had no shared haplotypes and belonged to two distinct subclades. (National Parks: KNP = Katavi, RNP = Ruaha, NNP = Nyerere, BCKR = Burigi Chato, Game Reserves: PGR = Pigi, LGR = Lukwati, RGR = Rungwa, SGR = Selous, Ecosystems: S.E. = Serengeti, KILIE = Kilimanjaro–Longido, TME = Tarangire Manyara)

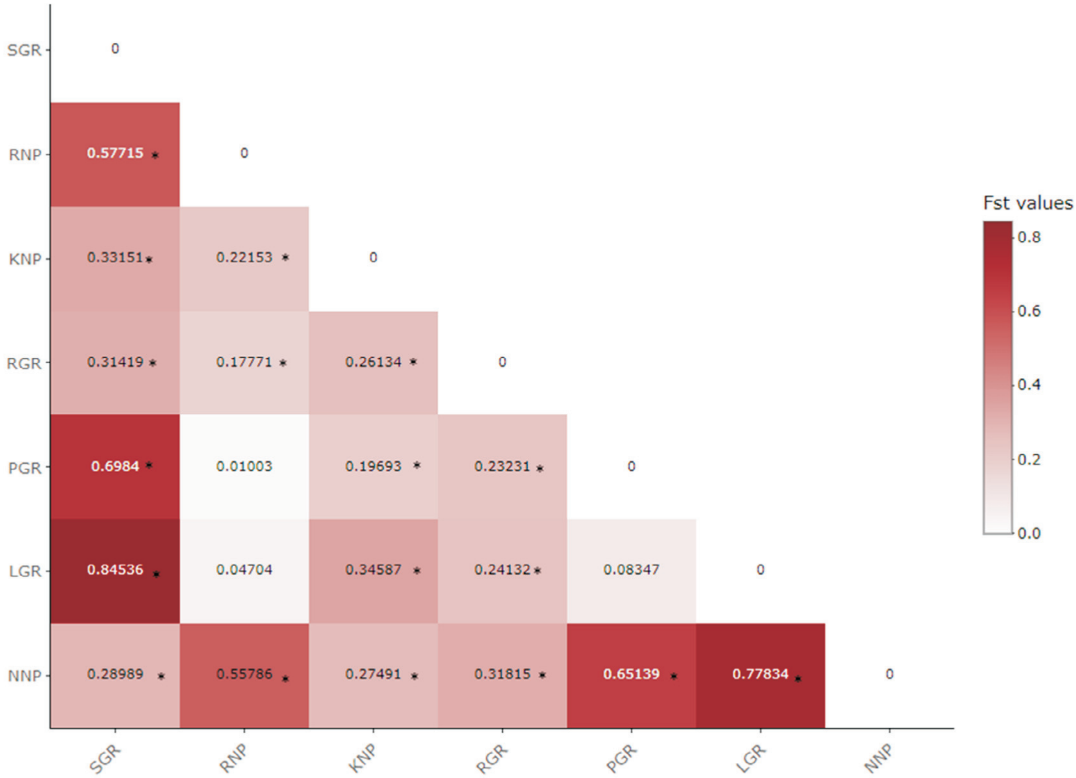

Fig. 2. Pairwise comparison of elephant mtDNA F_{ST} values, illustrating genetic differentiation, with darker shades representing higher F_{ST} values. Pairwise F_{ST} values that are statistically significant ($P \leq 0.05$) are indicated with an asterisk (*).

Most elephants in the EAM carry haplotypes that are widely distributed across Tanzania, including the Serengeti and Tarangire ecosystems. Our results also indicate that Rungwa (RGR) elephants share haplotypes with those in the Serengeti (Fig. 1A). Additionally, all elephants from Nyerere and Selous (Fig. 1A) exclusively carried haplotypes belonging to the Southeast-savanna (SS) subclade. However, nearly 50% of elephants in Katavi National Park (KNP) also carried haplotypes associated with the SS subclade.

The Masai giraffe population in Nyerere National Park (NNP) exhibited low haplotype diversity, with all individuals carrying a single haplotype, likely due to a low-diversity founder population. Genetic differentiation between Ruaha and Nyerere giraffes was high ($F_{ST} = 0.374$), with no shared haplotypes (Fig. 1B).

Discussion

Our results confirm that the EAM serves as a significant barrier to gene flow for both African savanna elephants and Masai giraffes, with a more pronounced effect observed in giraffes. This difference is likely due to the species-specific movement capabilities. Elephants can navigate steep terrain (Wall, Douglas-Hamilton & Vollrath, 2006), whereas giraffes are more restricted by slopes, leading to reduced dispersal in mountainous areas (Lohay *et al*., 2023).

Mitochondrial DNA patterns support this distinction. Only one elephant haplotype was found to be shared across the EAM, suggesting some historical gene flow mediated by females (Fig. 1). In contrast, giraffes showed no shared haplotypes between the Nyerere (NNP) and Ruaha (RNP) populations, indicating significant population subdivision ($F_{ST} = 0.374$). Moreover, giraffe haplotypes were found in distinct subclades, with Ruaha individuals clustering with populations west of the Rift, while Nyerere giraffes grouped with eastern populations (Lohay *et al*., 2023). This east–west genetic split reflects patterns seen in northern Tanzania, where the Gregory Rift Valley has separated giraffe lineages for about 290 000 years (Lohay *et al*., 2023). Elephants located west of the EAM, such as those in Ruaha, Rungwa, and Katavi, exhibited low F_{ST} values and shared haplotypes, indicating historical or ongoing gene flow supported by relatively intact corridors. However, increasing habitat loss, particularly of miombo woodlands, poses a growing threat to this connectivity (Lobora *et al*., 2018).

Giraffes, on the other hand, appear to be more sensitive not only to large barriers like mountains but also to finer-scale landscape features. Even in the absence of obvious physical divisions, genetic subdivision among giraffes is often influenced by slope, habitat type, and historical fragmentation (Brown *et al*., 2007; Crowhurst *et al*., 2013; Epps, Wasser, Keim, Mutayoba & Brashares, 2013). Similar genetic fragmentation has been observed in other species, including lions and sable antelopes (Smitz *et al*., 2018; Pitra *et al*., 2002). Our findings reinforce that giraffe populations can become genetically distinct, even over relatively short distances.

These results highlight the need to consider species-specific traits in habitat connectivity assessments. Elephants can navigate fragmented landscapes (Lohay *et al*., 2020, Ahlering *et al*., 2012), while giraffes face greater restrictions. These differences are crucial for conservation, especially amid ongoing land-use changes and deforestation affecting ecological corridors. To further clarify the timing and extent of divergence among giraffe populations, future research should incorporate nuclear DNA and broader geographic sampling. This will help build a more comprehensive understanding of population history and support targeted conservation strategies across Tanzania's key ecosystems.

Acknowledgements

Thanks to Penn State University and Wild Nature Institute for supporting this project. Special thanks to field assistants James Madeli, Emmanuel Kimaro, and the Tanzania Wildlife Research Institute for providing additional elephant tissue samples for this project. We are grateful to the Grumeti Fund for supporting the SAWMA conference. We gratefully acknowledge the Commission for Science and Technology (COSTECH) for granting research clearances #2020-185-NA-1990-172 and #2019-621-NA-2019-132, and Penn State University for approving our research using protocol IACUC#PROTO 201901219.

§ORCID iDs

G. Lohay: orcid.org/0000-0003-1129-8933
D. Pearce: orcid.org/0000-0003-2985-1020
M.L. Bond: orcid.org/0000-0001-8500-6564
A. Lobora: orcid.org/0000-0002-6067-4539
D.E. Lee: orcid.org/0000-0002-1042-9543
D. Cavener: orcid.org/0000-0002-1558-7137

References

Allendorf, F.W., Hohenlohe, P.A. & Luikart, G. (2010). Genomics and the future of conservation genetics. *Nature Reviews Genetics*, 11(10), 697–709. https://doi.org/10.1038/nrg2844

Ahlering, F.W.,Hailer, F, Roberts, M.T. & Foley, C (2011). A simple and accurate method to sex savannah, forest and Asian elephants using noninvasive sampling techniques. *Molecular Ecology Resources*, 11(5), 831–834. https://doi/full/10.1111/j.1469-1795.2010.00400.x

Ahlering, M.A., Eggert, L.S., Western, D., Estes, A., Munishi, L., Fleischer, R. & Maldonado, J.E. (2012). Identifying source populations and genetic structure for savannah elephants in human-dominated landscapes and protected areas in the Kenya-Tanzania borderlands. *PLOS ONE*, 7(12), e52288. https://doi.org/10.1371/journal.pone.0052288

Brown, D.M., Brenneman, R.A., Koepfli, K-P., Pollinger, J.P., Milá, B., Georgiadis, N.J., Louis, E.E., Grether, G.F., Jacobs, D.K. & Wayne, R.K. (2007). Extensive population genetic structure in the giraffe. *BMC Biology*, 5(1), 57. https://doi.org/10.1186/1741-7007-5-57

Crowhurst, R.S., Mullins, T.D., Mutayoba, B.M. & Epps, C.W. (2013). Characterization of eight polymorphic loci for Maasai giraffe (*Giraffa camelopardalis tippelskirchi*) using non-invasive genetic samples. *Conservation Genetics Resources*, 5(1), 85–*87.* https://doi.org/10.1007/s12686-012-9739-x

Epps, C.W., Mutayoba, B.M., Gwin, L. & Brashares, J.S. (2011). An empirical evaluation of the African elephant as a focal species for connectivity planning in East Africa. *Diversity and Distributions*. 17(4), 603–612. https://doi.org/10.1111/j.1472-4642.2011.00773.x

Epps, C.W., Wasser, S.K., Keim, J.L., Mutayoba, B.M. & Brashares, J.S. (2013). Quantifying past and present connectivity illuminates a rapidly changing landscape for the African elephant. *Molecular Ecology*, 22(6), 1574–1588. https://doi.org/10.1111/mec.12198

Excoffier, L. & Lischer, H.E.L. (2010). Arlequin suite ver 3.5: A new series of programs to perform population genetics analyses under Linux and Windows. *Molecular Ecology Resources*, 10(3), 564–567. https://doi.org/10.1111/j.1755-0998.2010.02847.x

Evans, B.J., Bliss, S.M., Mendel, S.A. & Tinsley, R.C. (2011). The Rift Valley is a major barrier to dispersal of African clawed frogs (*Xenopus*) in Ethiopia. *Molecular Ecology*, 20(20), 4216–4230. https://doi.org/10.1111/j.1365-294x.2011.05262.x

Fahrig, L. (2013). Rethinking patch size and isolation effects: The habitat amount hypothesis. *Journal of Biogeography*, 40(9), 1649–1663. https://doi.org/10.1111/jbi.12130

Giraffe Conservation Foundation website, https://giraffeconservation.org/wp-content/uploads/2019/04/Tanzanias-Giraffe-HR-c-GCF.pdf (accessed 16 March 2025).

Jones, T., Cusack, J.J., Pozo, R.A., Smit, J., Mkuburo, L., Baran, P., Lobora, A.L., Mduma, S. & Foley, C. (2018). Age structure as an indicator of poaching pressure: Insights from rapid assessments of elephant populations across space and time. *Ecological Indicators*, 88, 115–125. https://doi.org/10.1016/j.ecolind.2018.01.030

Kumar, S., Nei, M., Dudley, J. & Tamura, K. (2008). MEGA: A biologist-centric software for evolutionary analysis of DNA and protein sequences. *Briefings in Bioinformatics*, 9(4), 299–306. https://doi.org/10.1093/bib/bbn017

Lobora, A.L., Nahonyo, C.L., Munishi, L.K., Caro, T., Foley, C., Prunier, J.G., Beale, C.M. & Eggert, L.S. (2018). Incipient signs of genetic differentiation among African elephant populations in fragmenting miombo ecosystems in south-western Tanzania. *African Journal of Ecology*, 56(4), 993–1002. https://doi.org/10.1111/aje.12534

Lohay, G.G., Lee, D.E., Wu-Cavener, L., Pearce, D.L., Hou, X., Bond, M.L. & Cavener, D.R. (2023). Genetic evidence of population subdivision among Masai giraffes separated by the Gregory Rift Valley in Tanzania. *Ecology and Evolution*, 13(6), e10160. https://doi.org/10.1002/ece3.10160

Lohay, G.G., Weathers, T.C., Estes, A.B., McGrath, B.C. & Cavener, D.R. (2020). Genetic connectivity and population structure of African savanna elephants (*Loxodonta africana*) in Tanzania. *Ecology and Evolution*, 10(20), 11069–11089. https://doi.org/10.1002/ece3.6728

Mkuburo, L., Nahonyo, C., Smit, J., Jones, T. & Kohi, E. (2020). Investigation of the effect of poaching on African elephant (*Loxodonta africana*) group size and composition in Ruaha National Park, Tanzania. *Scientific African*, 9, e00490. https://doi.org/10.1016/j.sciaf.2020.e00490

Newmark, W.D. (2008). Isolation of African protected areas. *Frontiers in Ecology and the Environment*, 6(6), 321–328. https://doi.org/10.1890/070003

Noss, R.F., Robsob, A.P., Baldwin, R., Beier, P., Davis, C.R., Dellasala, D.A., Francis, J., Locke, H., Nowak, K., Lopez, R., Reining, C., Trombulak, S.C. & Tabor, G. (2012). Bolder thinking for conservation, *Conservation Biology*, 26(1), 1–4. https://doi.org/10.1111/j.1523-1739.2011.01738.x

Pitra, C., Hansen, A.J., Lieckfeldt, D. & Arctander, P. (2002). An exceptional case of historical outbreeding in African sable antelope populations. *Molecular Ecology*, 11(7), 1197–1208. https://doi.org/10.1046/j.1365-294X.2002.01516.x

Riggio, J., Jacobson, A.P., Hijmans, R.J. & Caro, T. (2019). How effective are the protected areas of East Africa? *Global Ecology and Conservation*, 17, e00573. https://doi.org/10.1016/j.gecco.2019.e00573

Smitz, N., Jouvenet, O., Ambwene Ligate, F., Crosmary, W-G., Ikanda, D., Chardonnet, P. & Michaux, J.R. (2018). A genome-wide data assessment of the African lion (*Panthera leo*) population genetic structure and diversity in Tanzania. *PLOS ONE*, 13(11), e0205395. https://doi.org/10.1371/journal.pone.0205395

Taylor, P.D., Fahrig, K.H. & Merriam, G. (1993). Connectivity is a vital element of landscape structure. *Oikos*, 68(3), 371–373.

Wall, J., Douglas-Hamilton, I. & Vollrath, F. (2006). Elephants avoid costly mountaineering. *Current Biology*, 16(14), R527–R529. https://doi.org/10.1016/j.cub.2006.06.049

Responsible Editor: L. Minnie