First population estimates of two Critically Endangered frogs from an isolated forest plateau in Madagascar

Izabela M. Barata, Jary H. Razafindraibe, Rojo Nadrianina Ravelojaona
Etienne Ralovarisoa, Katherine E. Mullin
Michael A. Hudson and Jeff Dawson

Abstract In the largely deforested areas of Madagascar, small forest fragments remain as last refuges for amphibian diversity. Isolated populations of the Critically Endangered *Anodonthyla vallani* and *Anilany helenae* persist in the fragmented forest of Ambohitantely but little information is available to inform their management and any conservation interventions. We generated estimates of population size and occupancy for both species in the largest fragment of Ambohitantely Special Reserve using acoustic survey data collected from 84 sites along 12 transects in December 2018. We used a single-season occupancy model to estimate detection and occupancy and a Royal–Nichols model to estimate abundance and population size. *Anilany helenae* and *A. vallani* had high occupancy rates (80 and 93%, respectively) whereas their detection rates differed (34 and 55%, respectively). Abundance and occupancy were best explained by vegetation structure whereas detection was influenced by time of survey and rainfall. For our sampled sites the estimated population sizes of males were 855 for *A. vallani*, with an estimated density of 52 individuals/ha, and 388 for *A. helenae*, with an estimated population density of 23 individuals/ha. Given their relatively low densities, small population sizes and restricted ranges, any further habitat loss could have drastic consequences for these populations. Our results provide guidance for future species-focused studies, and can inform conservation management at the local scale. Our work will help to improve species monitoring in Madagascar and elsewhere, especially for range-restricted non-charismatic amphibians.

Izabela M. Barata (Corresponding author, orcid.org/0000-0002-8955-2607, izabela.barata@durrell.org), Michael A. Hudson* (orcid.org/0000-0002-7640-1885) and Jeff Dawson (orcid.org/0000-0002-1563-9849) Durrell Wildlife Conservation Trust, Les Augrès Manor, Trinity, Jersey, JE3 5BP, Channel Islands

Jary H. Razafindraibe, Rojo Nadrianina Ravelojaona and Etienne Ralovarisoa University of Antananarivo, Antananarivo, Madagascar

Katherine E. Mullin (orcid.org/0000-0002-7816-3083) Cardiff School of Biosciences, Cardiff University, Cardiff, UK

*Also at: Institute of Zoology, Zoological Society of London, Regent's Park, London, UK

Received 18 February 2021. Revision requested 27 May 2021.
Accepted 27 July 2021. First published online 29 September 2022.

Keywords Acoustic monitoring, amphibian conservation, *Anilany helenae*, *Anodonthyla vallani*, hotspot, population management, small population size

Supplementary material for this article is available at doi.org/10.1017/S0030605321001034

Introduction

The tropical forests of Madagascar are one of the highest-priority areas for biodiversity conservation because of their high levels of species richness and increasing threats (Myers et al., 2000). The fauna and flora of Madagascar have a unique evolutionary history as a result of the separation of Madagascar from Africa and India 165 and 88 million years ago, respectively (Ali & Aitchison, 2008). Multiple processes have shaped the current endemism rates of many taxa, resulting in a large number of restricted-range species (Goodman & Benstead, 2005; Pearson & Raxworthy, 2009). Approximately 9% of species in Madagascar have been driven to extinction by deforestation between 1950 and 2000 (Allnutt et al., 2008). Estimates show that 44% of natural forest cover of Madagascar has been lost over 6 decades (1953–2014) and c. half of the remaining forest is within 100 m of the forest edge (Vieilledent et al., 2018).

As deforestation accelerates in many areas of Madagascar (Allnutt et al., 2008; Vieilledent et al., 2018), small fragments remain as last refuges for biodiversity (Andreone et al., 2008a). Comprising > 40 fragments of multiple sizes, Ambohitantely Special Reserve contains some of the last remaining forest in the central plateau of Madagascar and is considered the last refuge for multiple range-restricted endemic frogs. Despite high levels of amphibian richness and endemicity (Goodman & Benstead, 2005; Andreone et al., 2007), no extinctions of Malagasy amphibians have been detected (Andreone et al., 2008a). However, amphibian diversity is probably underestimated (Vieites et al., 2009) and species losses could occur before species are discovered. Species occurring in small fragments are under heavy anthropogenic pressure (Vallan, 2000b; Lehtinen & Ramanamanjato, 2006) and the mid- to long-term stability of these amphibian communities is uncertain (Andreone et al., 2007). Additionally, little information is available on the biology and abundance of most Malagasy frogs (Andreone & Luiselli, 2003; Andreone et al., 2008b), resulting in poor baselines from which to elucidate population trends.

Isolated populations of two Critically Endangered frogs, *Anodonthyla vallani* and *Anilany helenae*, persist in Ambohitantely Special Reserve but little information is available to inform their management and any conservation interventions. Both species are endemic to a few forest fragments in and around Ambohitantely Special Reserve, the largest of which is 1,284 ha and is located within the protected area. They are restricted to high-elevation habitats (*A. vallani* recorded at 1,590 m and *A. helenae* at 1,500 m), with an estimated extent of occurrence of 29 km^2 for both species (IUCN SSC Amphibian Specialist Group, 2016, 2020). Here we provide a first estimation of population sizes and occupancy rates for both species, using acoustic surveys, and examine the ecological relationships driving these key parameters. These baseline population estimates will support establishment of conservation priorities and examination of extinction probabilities (Andreone et al., 2008b).

Study area

Ambohitantely is a Special Reserve, created in 1982, on the central plateau of Madagascar (Fig. 1a), 135 km north-east of the capital Antananarivo. The Reserve covers 5,600 ha, less than half of which is natural forest. Forest remnants are distributed patchily and consist of c. 42 fragments that vary in shape and size (range 0.16–1284 ha; Fig. 1b). Together these forest fragments encompass an area of 1,627 ha (based on recent satellite data). Altitude range is 1,300–1,650 m. The subhumid climate is typical of a montane forest, with two defined seasons (wet and dry) and mean temperatures of 12.5–23 °C (Goodman et al., 2018). The area has a mean annual rainfall of 1,461 mm, of which 86% falls in the warm season (November–April).

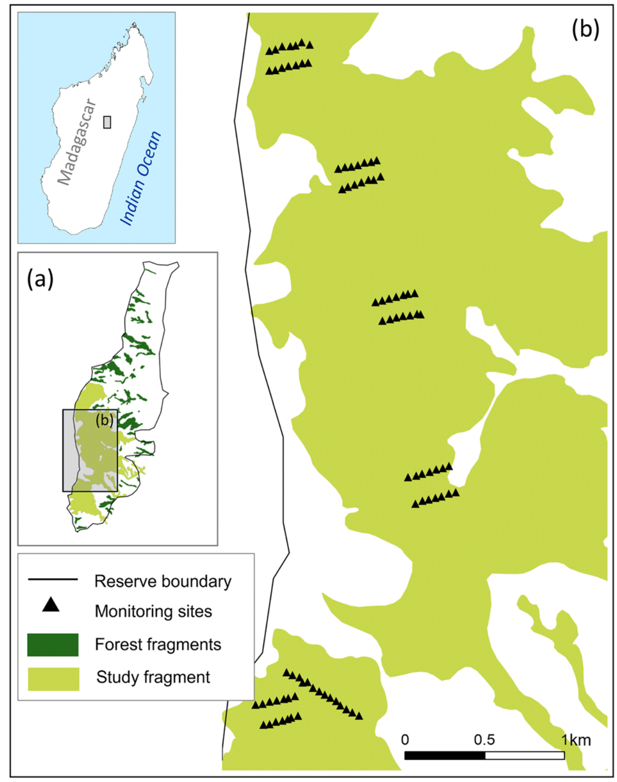

Fig. 1 (a) Ambohitantely Special Reserve in north-east Madagascar, with the forest fragments within the Reserve, and (b) the largest forest fragment (1,284 ha) where we conducted our study to estimate the population sizes of two Critically Endangered frogs (*Anodonthyla vallani* and *Anilany helenae*; Plate 1). Monitoring sites were in six blocks, each with two transects; each transect was 300 m long and had seven monitoring sites.

Methods

Survey design

Although *A. vallani* and *A. helenae* have been recorded in five and four fragments, respectively, in and around Ambohitantely Special Reserve (Vallan, 2000b; K.E. Mullin, unpubl. data, 2019, 2020), we collected data only in the largest fragment (1,284 ha), a forest remnant that is large enough to contain a representative number of individuals of both species (Vallan, 2000b). In this fragment we placed six blocks of two transects each (12 transects in total; Fig. 1). The distance between blocks was 120–1,500 m. Each transect was 300 m long and contained seven monitoring sites, each 50 m apart, resulting in a total of 84 sites across the study area (Fig. 1). At each monitoring site we carried out a 5-minute acoustic point count survey during the night (18:00–23:00) and recorded the presence or absence of *A. vallani* and *A. helenae* (Plate 1a,b). Acoustic surveys were carried out on 8–28 December 2018, during the wet season, and each monitoring site was visited at least three times (with a 5-minute acoustic point count survey at each visit), to create a detection history.

In our study area both target species could potentially be misidentified as *Platypelis pollicaris* (K.E. Mullin, pers. obs., 2019; Plate 1c), but they can be differentiated by an experienced observer (J.H. Razafindraibe, pers. obs., 2018). To ensure that target species were being correctly identified and to control for variation in detection, a single experienced observer (JHR, who has > 6 years of amphibian survey experience in Madagascar) performed the acoustic surveys and species identifications. Additionally, recordings of *A. vallani* and *A. helenae* were available for comparisons in the field; JHR listened to these prior to each acoustic point count and when there was uncertainty regarding species identification. Spectrograms are available for *A. vallani*, *A. helenae* and

P. pollicaris, illustrating the differences in their calling patterns (Supplementary Fig. 1).

We estimated distance to target species call as either near ($\leq$ 10 m) or far ($>$ 10 m) and we recorded the direction to the target using a compass (the observer always stood facing north). We did this to minimize the risk of double counting of individuals from separate points; if two calls could potentially belong to the same individual (e.g. both being recorded as far and in the same relative direction), one record was discounted from the analysis. At each monitoring site we recorded time, maximum and minimum temperatures (°C), maximum and minimum relative humidity (%) and rainfall (categorical: 0 no rain, 1 light rain, 2 heavy rain) at the beginning and end of every 5-minute acoustic point count survey, and elevation, distance from water and vegetation structure (bamboo and *Pandanus* numbers within a 5-m radius of the observer, and canopy cover within a 1-m radius of the observer).

Data analysis

We estimated occupancy and detection probability using a single-season occupancy model (MacKenzie et al., 2002), and we used the Royle–Nichols model to estimate species abundance and population size (Royle & Nichols, 2003). We modelled species separately (i.e. using a single-species modelling framework). Occupancy models account for detection rates (i.e. chances of detecting a species at a site if present) and estimate the proportion of area occupied by a species, whereas the Royle–Nichols model estimates the occupancy rate when heterogeneity in the detection probability exists as a result of variation in animal abundance. The Royle–Nichols model estimates abundance (i.e. number of individuals at each site) using presence/absence data from repeated occasions, and detection probability represents the likelihood of recording all individuals at a site at a given time (Royle & Nichols, 2003). Statistical analyses were performed in *R 3.4.2* (R Foundation for Statistical Computing, Vienna, Austria) using package *unmarked* (Fiske & Chandler, 2011).

We built models using a stepwise approach, using the Akaike information criterion (AIC) to rank candidate models and to select covariates for final model fitting. In the first step we fitted detection covariates using the variables time of survey, rainfall and maximum temperature. In the second step we fitted covariates for occupancy and abundance using the variables elevation, vegetation structure (bamboo, *Pandanus* and canopy) and distance from water. In the final step we combined the covariates detection probability and abundance/occupancy to build the final models. We ranked the models based on their AIC (the model with the lowest AIC having the best fit) and weighted them by the probability of being the best model in the set. We considered models with ΔAIC $<$ 2 to have strong support (Burnham & Anderson, 2002). We also used model weights to estimate the relative importance of predictors, given by the cumulative weight of the best-fitted models in which the predictors appear, and interpreted this as the probability of being a component of the best model (Symonds & Moussalli, 2011). We averaged all candidate models to account for model selection uncertainty.

Because we used an acoustic survey, detectability refers to the probability of hearing the species in a site if the species is both present and active (i.e. calling). Similarly, population size refers to the number of males present at the surveyed sites in the largest fragment in Ambohitantely Special Reserve. We followed the method of Kéry & Royle (2016) to calculate population size and used a parametric bootstrap method that generates a sampling distribution of the population based on the best-fitted model. We estimated density within our study fragment (i.e. number of adult males/ha) by dividing the estimated population size by the total surveyed area (each sampled site of 25 m radius and 1,963 m^2; total surveyed area of 16.5 ha). This was then extrapolated across the entire fragment (1,284 ha) to estimate the population beyond the surveyed sites.

Results

We obtained 126 acoustic records (i.e. presence) for *A. vallani* and 69 for *A. helenae*. We detected individuals of *A. vallani* in 71 sites (naïve occupancy 0.84) and individuals of *A. helenae* in 51 sites (naïve occupancy 0.61). Details of model fitting and of the covariates influencing species occupancy, abundance and detectability are presented in Supplementary Material 1. Overall, occupancy of both species was best explained by features in the vegetation structure: canopy cover for *A. vallani* and bamboo numbers for *A. helenae*. Detection probability was influenced by time of survey (for *A. vallani*) and rainfall (for *A. helenae*) (Fig. 2, Supplementary Table 1). Based on model-averaged estimates occupancy of the two species was similar, whereas detection probabilities differed (Supplementary Fig. 2). Occupancy estimates for both species were high (0.93 ± 0.07 for *A. vallani*; 0.80 ± 0.09 for *A. helenae*). Species detection rates differed, with *A. helenae* having a 34 ± SE 0.05% chance of being detected if present, which was lower than that of *A. vallani*, with a probability of detection of 55 ± SE 0.05%.

For both species abundance was best explained by vegetation structure, whereas detection was best explained by time of survey for *A. vallani* and rainfall for *A. helenae* (Supplementary Table 1). The abundance of *A. vallani* was best explained by bamboo number and canopy cover, whereas the abundance of *A. helenae* at each site was best explained by the numbers of both bamboo and *Pandanus* (Fig. 2). There was an estimated abundance of 10 adult male *A. vallani* at each monitoring site, with a large standard error (range = 4–24, SE = 7). Overall, for our sampled sites

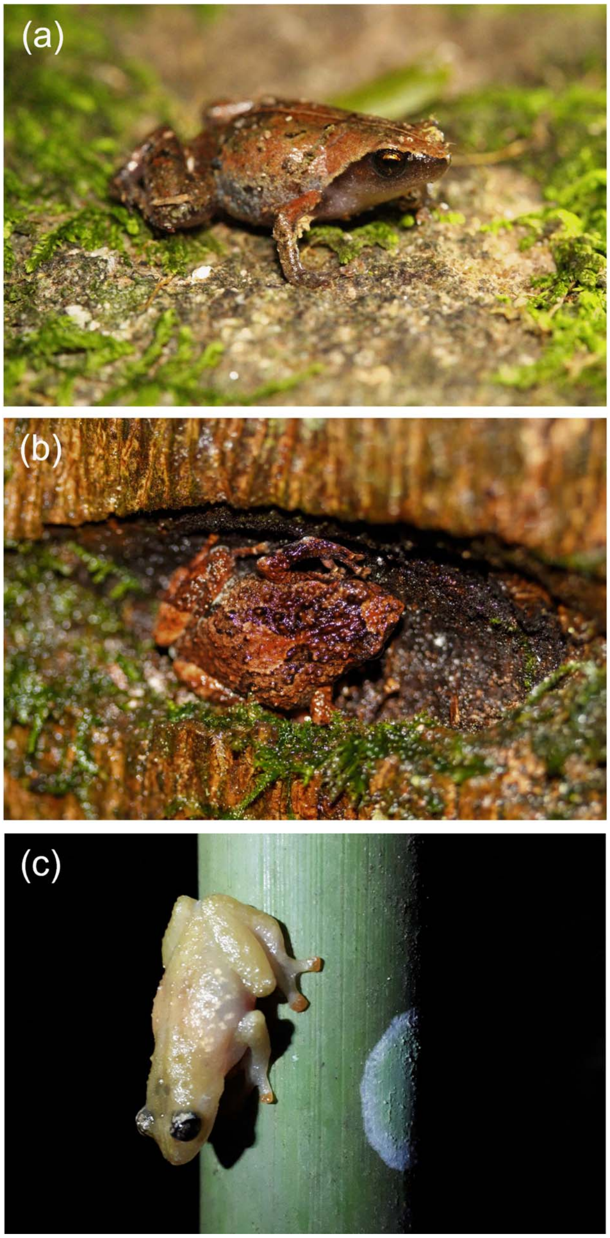

PLATE 1 The two Critically Endangered amphibian species studied: (a) *Anilany helenae* and (b) *Anodonthyla vallani*. Both species could potentially be misidentified as (c) *Platypelis pollicaris* but they can be differentiated visually and acoustically by an experienced observer (see spectrograms showing the differences in their calling patterns; Supplementary Fig. 1).

the estimated population size of male *A. vallani* was 855 (95% CI = 250–1,052), giving an estimated population density of 52 individuals/ha. The estimated abundance of adult male *A. helenae* was four individuals per site (range = 1–8, SE = 2). The estimated population size of male *A. helenae* in the studied sites was 388 (95% CI = 128–580), giving an estimated density of 23 individuals/ha. For both species population numbers appear to be overestimations of the sampled distribution of the population based on the best-fitted model (Supplementary Fig. 3). Extrapolating the estimated density across the whole fragment (1,284 ha) gives estimated male population sizes of 66,534 for *A. vallani* and 30,193 for *A. helenae*.

Discussion

Using presence and absence data obtained from multiple acoustic surveys, we provide the first estimates of population size for *A. vallani* and *A. helenae*, two Critically Endangered amphibian species from the central plateau of Madagascar. Both species are known to live in only a small number of forest fragments, one of which is outside Ambohitantely Special Reserve and has been subject to much deforestation since these frogs were discovered there (Vallan, 2000a). It is unknown whether these species are able to survive in the smaller forest fragments within the Reserve, given the different microhabitats and microclimates these smaller fragments probably have. For example, tree holes, which are an important reproduction site for *A. vallani*, may be absent in heavily logged forest fragments (K.E. Mullin, pers. obs., 2019, 2020). The reproduction of both species relies on high humidity and dew points, which could be lower in smaller forest fragments that are more affected by edge effects. Although forest fragments of various sizes are important to support a diverse amphibian community in Madagascar (Riemann et al., 2015), forest fragmentation in Ambohitantely Special Reserve decreases amphibian population sizes and increases species vulnerability by reducing the heterogeneity of the microhabitats available to frogs (Vallan, 2000b). Therefore, our estimates of population sizes refer to the single largest forest fragment in Ambohitantely Special Reserve, which is probably the largest and most significant population refuge for these threatened frogs.

Both species had moderate to high occupancy rates in the studied forest fragment but we recorded relatively low density estimates compared to similar studies of leaf litter frogs: 470 individuals/ha of *Mantella milotympanum* in Madagascar (Vieites et al., 2005), 78–4,285 individuals/ha of four *Sooglossus* species in Seychelles (Gerlach, 2007) and 20–960 individuals/ha of multiple tree frog species in the Brazilian Atlantic Forest (Siqueira et al., 2009). Although we consider our estimates of population size at the surveyed sites to be robust, extrapolations to the whole fragment based on density do not account for heterogeneity in occupancy outside the surveyed area or variation in habitat suitability/availability across the fragment. Despite these uncertainties, such density estimations and extrapolations beyond surveyed sites provide valuable baselines for future monitoring and assessments of population trends.

The acoustic surveys allowed us to collect a large dataset over a short period of time, facilitating the rapid assessment

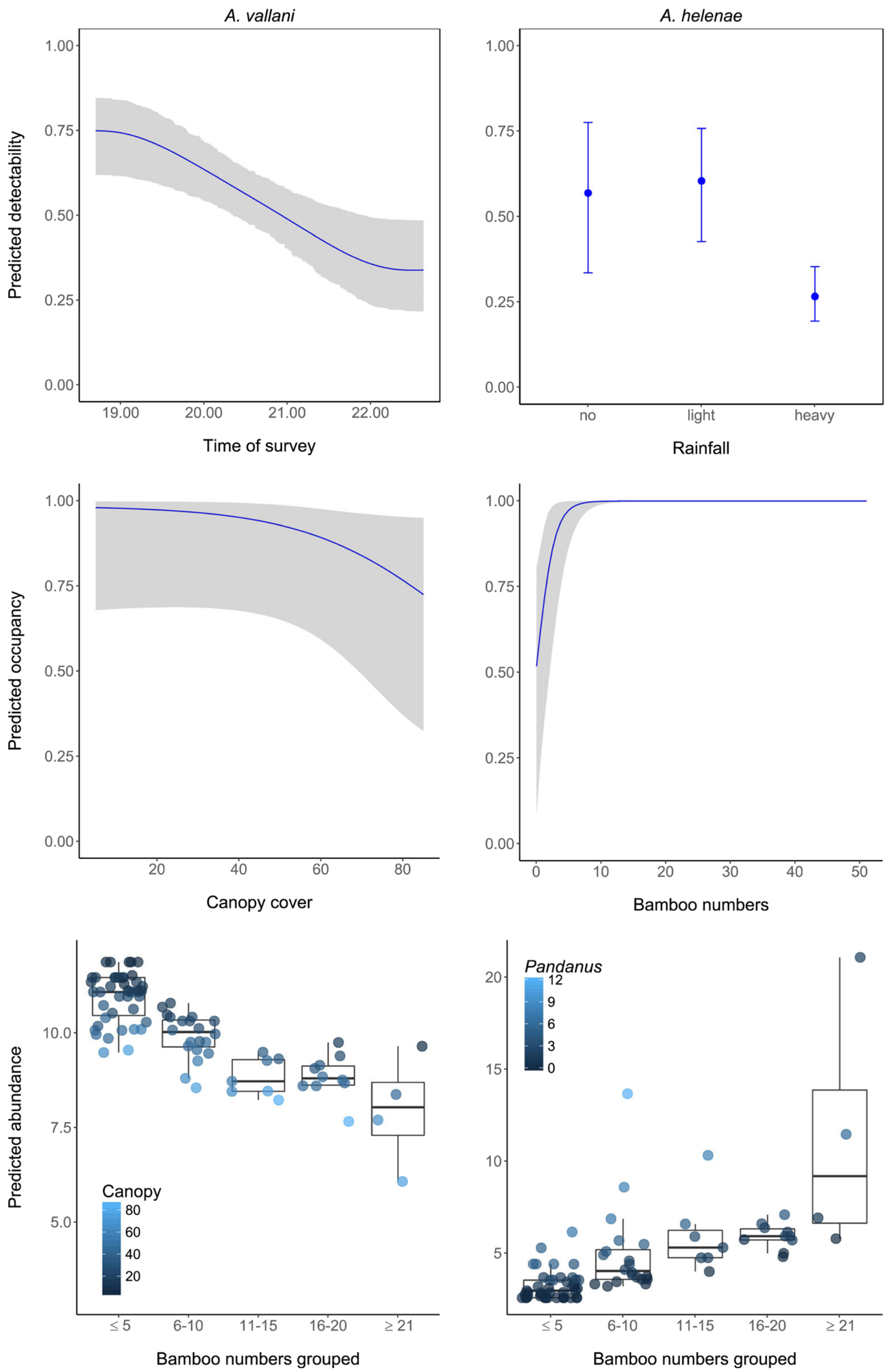

FIG. 2 Predicted values of detection probability, occupancy, and abundance (with 95% confidence intervals), based on model averaging. For *A. vallani*, predictions are based on time of survey (detectability, top left), canopy cover (occupancy, middle left), and number of bamboo and canopy cover (abundance, bottom left). For *A. helenae*, predictions are based on rainfall (detectability, top right), number of bamboo (occupancy, middle right), and numbers of bamboo and *Pandanus* (abundance, bottom right) at a given site.

of the status of two relatively understudied species. We found that the ideal conditions for the acoustic surveying of *A. helenae* involve no/moderate rain, whereas acoustic surveying of *A. vallani* should occur at 18.40–20.40 as this is when the likelihood of detection is highest. Although acoustic surveys can optimize data collection for cryptic species, they should be used with caution when species with similar calls are sympatric, and only if the observer has sufficient experience to distinguish the calls of target and non-target species. Additionally, by using acoustic surveys, estimates will only account for, and will therefore be limited to, the number of active males in the population, which can potentially introduce bias into inferences to the whole population, especially when the sex ratio is unknown (a 50:50 sex ratio is a naïve assumption). Alternative methods are available that could be used to improve parameter estimates, such as using automated recording devices (Measey et al., 2017) or environmental DNA (Lopes et al., 2017), which has since been trialled for *A. vallani* at Ambohitantely (Mullin et al., 2021b). Both of these tools eliminate the requirement for the observer to be at the site at the same time as the species, enabling data collection on much larger spatial and temporal scales and improving the robustness of any inferences made, although both tools require significant technical expertise.

Overall, occupancy and abundance were strongly associated with vegetation structure. *Anilany helenae* is terrestrial and has been recorded in the leaf litter of riparian forests, with its breeding behaviour possibly involving ground nests (Vallan, 2000a; IUCN SSC Amphibian Specialist Group, 2016). *Pandanus* leaf axils and bamboo leaves on the forest floor could influence species presence by creating hiding places and benefitting the behaviour of this cryptic species. *Anodonthyla vallani* is active at night, with males calling from tree trunks at heights of 2–3 m. It is presumed to reproduce through larval development in water-filled tree holes (Vences et al., 2010; IUCN SSC Amphibian Specialist Group, 2020). Given this requirement for trees and tree holes, the negative relationship between abundance and canopy cover could be explained by the proportion of such habitat available. Given this narrow habitat requirement, any further habitat degradation could have drastic consequences for the studied populations.

Our findings indicate that a mosaic of vegetation cover is an important feature for the persistence of these species, and probably many others, providing a diversity of habitats that regulates ecological processes and population sizes. However, wildfires (caused by human activity outside the Reserve) are a continuing threat to the forest in Ambohitantely Special Reserve and these forest-dependent species. In Ambohitantely Special Reserve forest fragmentation decreases amphibian population sizes and increases species vulnerability to local extinction by reducing heterogeneity in microhabitats (Vallan, 2000b). Positively engaging and working with local communities to address the pressures leading to habitat degradation are key conservation actions required. For example, management authorities often work with communities in protected areas in Madagascar to establish and maintain firebreaks (J. Dawson, pers. obs., 2020) and this is already being done in Ambohitantely. Similarly, to reduce forest exploitation at the nearby Ankafobe Private Reserve, plantations have been established outside the Reserve for people to use for firewood and charcoal in return for protecting the forest (K.E. Mullin, pers. obs., 2020).

As the annual deforestation rate increases in Madagascar (Vieilledent et al., 2018), persistent isolation is becoming a threat to amphibian species that are rare, habitat specialists and/or intolerant of matrix habitats, making them particularly prone to local extinction (Lehtinen & Ramanamanjato, 2006). Although *A. helenae* occurs elsewhere in the surrounding landscape (Mullin et al., 2021a), the status of both target species in other fragments is largely unknown and the forest fragment we surveyed is probably a key refugium of the species. Studies of amphibian population connectivity and viability in isolated fragments in the central plateau of Madagascar are essential, contributing to our understanding of the effects of fragmentation and providing evidence to support future land-use planning. *Anodonthyla vallani* and *A. helenae* are both categorized as Critically Endangered based on criteria B1ab(iii) because of their reduced extent of occurrence (B1), severe fragmentation (a) and the continued decline (b) in the quality of their habitat (iii) (IUCN SSC Amphibian Specialist Group, 2016, 2020). *Anilany helenae* was assessed in 2016 and would benefit from an update to its assessment. Our findings do not contradict the current assessment but provide a baseline from which to assess any potential future changes in population size (IUCN Red List criterion A) or continuing declines (IUCN Red List criteria C). Despite Madagascar currently having 370 described frog species (AmphibiaWeb, 2021), population and/or abundance baselines are only available for a few charismatic species (e.g. *Mantella* spp.). Research is needed to increase this knowledge as these are key metrics for assessing the impacts of threats as well as conservation actions, especially for range-restricted species. Highly threatened frogs with small population sizes could be close to extinction, highlighting the need to preserve remaining forest fragments if we are to conserve the high number of range-restricted species that have so far survived deforestation in Madagascar.

Acknowledgements We thank the Malagasy authorities for permitting this research and the Director of Ambohitantely Special Reserve, Hery Lala Ravelomanantsoa, for facilitating our work; Tovo Raditra for being our forest guide and the other Madagascar National Parks staff at Ambohitantely Special Reserve for providing support; Steve Buckland for advice on developing this project; Serge Ndriantsoa and Tsanta Rakotonanahary for logistical support in organizing fieldwork and securing permissions; and the staff at Durrell Madagascar's head office in Tana for logistical support, especially our drivers Bruno and Franckly. Funding was provided through a Mohammed bin Zayed Conservation Fund grant to Durrell Wildlife Conservation Trust.

Author contributions Study design: JD, JHR; fieldwork: JHR, RNR, ER; data analysis: IMB, with input from MAH; writing: IMB, with equal contributions from all other authors.

Conflicts of interest None.

Ethical standards This research abided by *Oryx* guidelines on ethical standards. Surveys were undertaken under a research permit issued by the Ministry of Environment, Ecology and Forests (now Ministry of Environment and Sustainable Development) and with permission from Madagascar National Parks (permit number 259/19/MEEF/SG/DGF/DSAP/SCB).

References

Ali, J.R. & Aitchison, J.C. (2008) Gondwana to Asia: plate tectonics, paleogeography and the biological connectivity of the Indian sub-continent from the Middle Jurassic through latest Eocene (166–35 Ma). *Earth-Science Reviews*, 88, 145–166.

Allnutt, T.F., Ferrier, S., Manion, G., Powell, G.V.N., Ricketts, T.H., Fisher, B.L. et al. (2008) A method for quantifying biodiversity loss and its application to a 50-year record of deforestation across Madagascar. *Conservation Letters*, 1, 173–181.

AmphibiaWeb (2021) *The AmphibiaWeb Database*. amphibiaweb.org [accessed 22 June 2021].

Andreone, F. & Luiselli, L.M. (2003) Conservation priorities and potential threats influencing the hyper-diverse amphibians of Madagascar. *Italian Journal of Zoology*, 70, 53–63.

Andreone, F., Carpenter, A.I., Cox, N., Du Preez, L., Freeman, K., Furrer, S. et al. (2008a) The challenge of conserving amphibian megadiversity in Madagascar. *PLOS Biology*, 6, e118.

Andreone, F., Cox, N.A., Glaw, F., Kohler, J., Rabibisoa, N.H.C., Randriamahazo, H. et al. (2008b) Update of the Global Amphibian Assessment for Madagascar in light of species discoveries, nomenclature changes, and new field information. *Monografiae del Museo Regionale di Scienze Naturali di Torino*, 45, 419–436.

Andreone, F., Vences, M., Glaw, F. & Randrianirina, I.E. (2007) Remarkable records of amphibians and reptiles on Madagascar's central high plateau. *Tropical Zoology*, 20, 19–39.

Burnham, K.P. & Anderson, D.R. (eds) (2002) *Model Selection and Multimodel Inference*. Springer, New York, USA.

Fiske, I. & Chandler, R. (2011) Overview of *unmarked*: an *R* package for the analysis of data from unmarked animals. cran.r-project.org/web/packages/unmarked/vignettes/unmarked.pdf [accessed 18 November 2021].

Gerlach, J. (2007) Distibution and status of the Seychelles frogs (Amphibia: Anura: Sooglossidae). *Herpetological Journal*, 117, 115–122.

Goodman, S.M. & Benstead, J.P. (2005) Updated estimates of biotic diversity and endemism for Madagascar. *Oryx*, 39, 73–77.

Goodman, S.M., Raherilalao, M.J. & Wohlauser, S. (2018) *The Terrestrial Protected Areas of Madagascar: Their History, Description and Biota*. Association Vahatra, Antananarivo, Madagascar.

IUCN SSC Amphibian Specialist Group (2016) *Rhombophryne helenae*. In *The IUCN Red List of Threatened Species* 2016. dx.doi.org/10.2305/IUCN.UK.2016-1.RLTS.T58010A84183331.en.

IUCN SSC Amphibian Specialist Group (2020) *Anodonthyla vallani* (amended version of 2016 assessment). In *The IUCN Red List of Threatened Species* 2020. dx.doi.org/10.2305/IUCN.UK.2020-3.RLTS.T190944A176030045.en.

Kéry, M. & Royle, J.A. (2016) *Applied Hierarchical Modeling in Ecology*. Elsevier, London, UK.

Lehtinen, R. & Ramanamanjato, J.-B. (2006) Effects of rainforest fragmentation and correlates of local extinction in a herpetofauna from Madagascar. *Applied Herpetology*, 3, 95–110.

Lopes, C.M., Sasso, T., Valentini, A., Dejean, T., Martins, M., Zamudio, K.R. & Haddad, C.F. (2017) eDNA metabarcoding: a promising method for anuran surveys in highly diverse tropical forests. *Molecular Ecology Resources*, 17, 904–914.

MacKenzie, D.I., Nichols, J.D., Lachman, G.B., Droege, S., Andrew, J. & Langtimm, C.A. (2002) Estimating site occupancy rates when detection probabilities are less than one. *Ecology*, 83, 2248–2255.

Measey, G.J., Stevenson, B.C., Scott, T., Altwegg, R. & Borchers, D.L. (2017) Counting chirps: acoustic monitoring of cryptic frogs. *Journal of Applied Ecology*, 54, 894–902.

Mullin, K.E., Rakotomanga, M.G., Razafiarimanana, M.L.C., Barata, I.M.B, Dawson, J., Hailer, F. & Orozco-terWengel, P. (2021a) First amphibian inventory of one of Madagascar's smallest protected areas, Ankafobe, extends the range of the Critically Endangered *Anilany helenae* (Vallan, 2000) and the Endangered *Boophis andrangoloaka* (Ahl, 1928). *Herpetology Notes*, 14, 521–531.

Mullin, K.E., Barata, I.M., Dawson, J. & Orozco-terWengel, P. (2021b) First extraction of eDNA from tree hole water to detect tree frogs: a simple field method piloted in Madagascar. *Conservation Genetic Resources*, 2021, 1–9.

Myers, N., Mittermeier, R.A., Mittermeier, C.G., da Fonseca, G.A.B. & Kent, J. (2000) Biodiversity hotspots for conservation priorities. *Nature*, 403, 853–858.

Pearson, R.G. & Raxworthy, C.J. (2009) The evolution of local endemism in Madagascar: watershed versus climatic gradient hypotheses evaluated by null biogeographic models. *Evolution*, 63, 959–967.

Riemann, J.C., Ndriantsoa, S.H., Raminosoa, N.R., Rödel, M.O. & Glos, J. (2015) The value of forest fragments for maintaining amphibian diversity in Madagascar. *Biological Conservation*, 191, 707–715.

Royle, J.A. & Nichols, J.D. (2003) Estimating abundance from repeated presence-absence data or point counts. *Ecology*, 84, 777–790.

Siqueira, C.C., Vrcibradic, D., Almeida-Gomes, M., Borges-Junior, V.N.T., Almeida-Santos, P., Almeida-Santos, M. et al. (2009) Density and richness of leaf litter frogs (Amphibia: Anura) of an Atlantic Ranforest area in the Serra dos Órgãos, Rio de Janeiro State, Brazil. *Zoologia*, 26, 97–102.

Symonds, M.R.E. & Moussalli, A. (2011) A brief guide to model selection, multimodel inference and model averaging in behavioural ecology using Akaike's information criterion. *Behavioral Ecology and Sociobiology*, 65, 13–21.

Vallan, D. (2000a) A new species of the genus *Stumpffia* (Amphibia: Anura: Microhylidae) from a small forest remnant of the central high plateau of Madagascar. *Revue Suisse de Zoologie*, 107, 835–841.

Vallan, D. (2000b) Influence of forest fragmentation on amphibian diversity in the nature reserve of Ambohitantely, highland Madagascar. *Biological Conservation*, 96, 31–43.

Vences, M., Glaw, F., Köhler, J. & Wollenberg, K.C. (2010) Molecular phylogeny, morphology and bioacoustics reveal five additional species of arboreal microhylid frogs of the genus *Anodonthyla* from Madagascar. *Contributions to Zoology*, 79, 1–32.

Vieilledent, G., Grinand, C., Rakotomalala, F.A., Ranaivosoa, R., Rakotoarijaona, J.R., Allnutt, T.F. & Achard, F. (2018) Combining global tree cover loss data with historical national forest cover maps to look at six decades of deforestation and forest fragmentation in Madagascar. *Biological Conservation*, 222, 189–197.

Vieites, D.R., Rabemananjara, F.C., Bora, P., Razafimahtratra, B., Ravoahangimalala O.R. & Vences, M. (2005) Distribution and population density of the black-eared Malagasy poison fog, *Mantella milotympanum* Staniszewski, 1996 (Amphibia: Mantellidae). In *African Biodiversity: Molecules, Organisms, Ecosystems* (eds B.A. Huber, B.J. Sinclair & K.-H. Lampe), pp. 197–204. Springer Nature, Dordrecht, The Netherlands.

Vieites, D.R., Wollenberg, K.C., Andreone, F., Köhler, J., Glaw, F. & Vences, M. (2009) Vast underestimation of Madagascar's biodiversity evidenced by an integrative amphibian inventory. *Proceedings of the National Academy of Sciences of the United States of America*, 106, 8267–8272.