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SYNTHESIS

Multinational evaluation of genetic diversity indicators for the Kunming-Montreal Global Biodiversity Framework

Alicia Mastretta-Yanes[1,2,3] | Jessica M. da Silva[4,5] | Catherine E. Grueber[6] | Luis Castillo-Reina[7] | Viktoria Köppä[8] | Brenna R. Forester[9] | W. Chris Funk[10,11] | Myriam Heuertz[12] | Fumiko Ishihama[13] | Rebecca Jordan[14] | Joachim Mergeay[7,15] | Ivan Paz-Vinas[10,16] | Victor Julio Rincon-Parra[17] | Maria Alejandra Rodriguez-Morales[18] | Libertad Arredondo-Amezcua[19] | Gaëlle Brahy[12] | Matt DeSaix[10] | Lily Durkee[11,20] | Ashley Hamilton[21,22] | Margaret E. Hunter[23] | Austin Koontz[22] | Iris Lang[24] | María Camila Latorre-Cárdenas[25] | Tanya Latty[6] | Alexander Llanes-Quevedo[26] | Anna J. MacDonald[27] | Meg Mahoney[11,28] | Caitlin Miller[10,11] | Juan Francisco Ornelas[29] | Santiago Ramírez-Barahona[30] | Erica Robertson[10,11] | Isa-Rita M. Russo[31] | Metztli Arcila Santiago[32] | Robyn E. Shaw[33] | Glenn M. Shea[34,35] | Per Sjögren-Gulve[36] | Emma Suzuki Spence[37] | Taylor Stack[38] | Sofía Suárez[32,39] | Akio Takenaka[13] | Henrik Thurfjell[40] | Sheela Turbek[10] | Marlien van der Merwe[41] | Fleur Visser[42,43] | Ana Wegier[39] | Georgina Wood[44,45] | Eugenia Zarza[1,46] | Linda Laikre[8] | Sean Hoban[21,22]

Correspondence
Jessica M. da Silva, Centre for Ecological Genomics and Wildlife Conservation, Department of Zoology, University of Johannesburg, Auckland Park 2006, Johannesburg, South Africa.
Email: jessicads@uj.ac.za

Alicia Mastretta-Yanes, Departamento de Ecología de la Biodiversidad. Instituto de Ecología, Universidad Nacional Autónoma de México. Av. Ciudad Universitaria 3000, 04510 Coyoacán, Ciudad de México, Mexico.
Email: amastretta@iecologia.unam.mx

Present address
Alicia Mastretta-Yanes, Departamento de Ecología de la Biodiversidad, Instituto de Ecología, Universidad Nacional Autónoma

Abstract

Under the recently adopted Kunming-Montreal Global Biodiversity Framework, 196 Parties committed to reporting the status of genetic diversity for all species. To facilitate reporting, three genetic diversity indicators were developed, two of which focus on processes contributing to genetic diversity conservation: maintaining genetically distinct populations and ensuring populations are large enough to maintain genetic diversity. The major advantage of these indicators is that they can be estimated with or without DNA-based data. However, demonstrating their feasibility requires addressing the methodological challenges of using data gathered from diverse sources, across diverse taxonomic groups, and for countries of varying socio-economic status and biodiversity levels. Here, we assess the genetic indicators for 919 taxa, representing 5271 populations across nine countries, including megadiverse countries and developing economies. Eighty-three percent of the taxa assessed had data available to calculate at least one indicator. Our results show that although the majority of species maintain most populations, 58%

Alicia Mastretta-Yanes and Jessica M. da Silva co-first authors, contributed equally to this work.

Linda Laikre and Sean Hoban co-senior authors.

For affiliations refer to page 15.

de México, Av. Ciudad Universitaria 3000, 04510, Coyoacán, Ciudad de México, Mexico

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of species have populations too small to maintain genetic diversity. Moreover, genetic indicator values suggest that IUCN Red List status and other initiatives fail to assess genetic status, highlighting the critical importance of genetic indicators.

KEYWORDS
biodiversity indicators, Convention on Biological Diversity, COP15, effective population size, populations maintained, Red List

INTRODUCTION

In December 2022, the United Nations' Convention of Biological Diversity (CBD; www.cbd.int) Kunming-Montreal Global Biodiversity Framework (GBF) was adopted. The GBF sets the pathway to achieve the vision of a world living in harmony with nature, with significant progress by 2030 (CBD, 2022a). The conservation of genetic diversity in the GBF is categorically different from previous commitments (Carroll et al., 2023) and is the first to aim for conserving genetic diversity of all species, not just economically valuable or domesticated. Until now, the genetic diversity of non-economically important species has been neglected by previous CBD strategies and other national and global conservation policies (Hoban et al., 2020; Laikre, 2010; Laikre et al., 2010). This was largely due to the complexity and expense associated with genetic information, communication barriers and lack of indicators to track genetic change to inform policy (Hoban et al., 2024; Hoban, Bruford, et al., 2023; Laikre et al., 2020; Taylor et al., 2017; Vernesi et al., 2008). To address this gap, three genetic indicators were proposed to monitor different aspects of genetic diversity, namely (i) maintaining genetically distinct populations, (ii) populations being large enough to retain genetic diversity, and (iii) the number of species with DNA-based monitoring of genetic diversity programmes (Hoban et al., 2020). The first two are based on processes leading to the loss of genetic diversity; by focusing on the underlying process, they can be estimated using both genetic and non-genetic data (Hoban et al., 2020; Hoban et al., 2024; Hoban, Paz-Vinas, et al., 2021; Laikre et al., 2020). These two indicators were adopted in the GBF (CBD, 2022a, 2022b), which means that Parties will use these indicators to report on their progress over the next decade.

The genetic diversity indicators were developed using SMART (specific, measurable, achievable, realistic, and timely) criteria (see table 2 in Hoban, Bruford, et al., 2021) and were designed to be relevant to Goal A ("The genetic diversity *within* populations of wild and domesticated species is maintained, *safeguarding their adaptive potential*") and Target 4 (…"to maintain and restore the genetic diversity *within and between* populations of native, wild and domesticated species to maintain their adaptive potential, including through *in situ* and *ex situ* conservation and sustainable management practices,"…) of the GBF (CBD, 2022a). The indicator that measures whether genetic diversity *between* populations is maintained was adopted as a complementary indicator (CBD, 2022b). It focuses on the loss of genetically distinct (e.g., harbouring genetic variants not found or rare elsewhere), presumably locally adapted, populations, and it is estimated as the number of populations that currently exist divided by the number of populations that originally existed (i.e., the proportion of maintained populations within species; PM indicator hereafter; see Materials and methods for detailed definitions and baseline time periods). A PM indicator value of 0 means that all populations within that species and within a given country have been lost, and a value of 1 means that all populations are maintained (the desired value). To estimate this indicator, it is necessary to spatially define and count populations, which is noted as one of the scientific challenges to estimating the indicators (Hoban et al., 2024).

The indicator that measures if genetic diversity is maintained *within* populations was adopted as headline indicator A.4 (mandatory for countries to report; CBD, 2022b). It focuses on populations being large enough to retain genetic diversity, and it is estimated as the proportion of populations within species in a given country with an effective population size (Ne) greater than 500 (*Ne 500 indicator* hereafter). A Ne 500 indicator value of 0 means that all populations within a species have an Ne below 500, and a value of 1 means that all populations are above 500 (the desired value). This indicator leverages established theory and empirical data in population genetics: when populations are below approximately Ne 500, loss of genetic diversity accelerates (Gilpin & Soulé, 1986; Jamieson & Allendorf, 2012). When Ne exceeds 500, evolutionary potential is expected to remain nearly stable (e.g., a very slow rate of loss) for fitness traits because there is a dynamic equilibrium between genetic drift (reducing diversity), mutation (adding diversity), and the efficiency of natural selection on additive genetic variance (Frankham et al., 2014; Franklin, 1980).

It should be noted that some have argued for an Ne >1000 threshold (Frankham, 2021). Importantly, in the absence of genetic data, the Ne of a population can be approximated using the census population size of mature individuals (Nc) and a ratio between Ne and Nc. The Ne/Nc ratio varies depending on the species' variance in reproductive success, breeding strategy, sex ratio and other life history traits (Frankham, 1995; Waples, 2002), so it can be adjusted by taxonomic group or even by population. If the ratio is unknown, a conservative ratio of 0.1 (i.e., Ne being equivalent to 10% of Nc) can be used (Frankham, 2021; Frankham et al., 2017; Hoban, Paz-Vinas, et al., 2021; Palstra & Ruzzante, 2008).

Other processes that can affect genetic diversity, such as undesired gene flow with introduced species, populations, or genetically modified organisms, inbreeding, or changes in frequency of selected genes, do require genetic data to be monitored (O'Brien et al., 2022). For these situations, a third indicator was proposed (Hoban et al., 2020; Laikre et al., 2020), which is the number of species in which genetic diversity has been or is being monitored using DNA-based methods for at least one population (*DNA-based genetic monitoring* indicator hereafter). This indicator is not included in the CBD monitoring framework, but countries can voluntarily report it (Pearman et al., 2024).

To assist Parties to the CBD and other stakeholders in compiling relevant data and quantifying these indicators, we developed a standardized, reproducible and flexible workflow (Hoban, da Silva, et al., 2023). However, concerns remained over the feasibility of reporting on these indicators for a large number of species, especially for biologically rich, developing economy nations where financial resources for biodiversity conservation and monitoring are generally more limited and where biological data (genetic or non-genetic) are perceived to be less readily available. Furthermore, some methodological concerns remained, including the baseline for assessing population extinction, how to delimit population boundaries, and the feasibility of using different sources of data to estimate the indicators (Hoban et al., 2024).

In this study, we address these concerns through a multinational application of the workflow described in Hoban, da Silva, et al. (2023). We conduct the first multi-country assessment of genetic diversity status, with emphasis on the PM and Ne 500 indicators. Nine countries across six continents, varying in economic status and biodiversity richness, were included: Australia, Belgium, Colombia, France, Japan, Mexico, South Africa, Sweden and the United States of America. Five of these countries are megadiverse (Australia, Colombia, Mexico, South Africa, and the USA; Mittermeier et al., 2005) and three are developing economy countries (Colombia, Mexico and South Africa: WorldData.info). Within each country, teams of researchers and conservation practitioners from academia, government institutions and non-governmental organizations undertook the assessments. Our specific objectives were to (i) quantify data availability across countries, taxonomic groups and indicators; (ii) assess whether methods for defining populations influence indicator values; (iii) quantify the distribution of indicator values across taxonomic groups and conservation threat status; and (iv) provide guidance to facilitate the calculation and uptake of the genetic diversity indicators at a global scale.

MATERIALS AND METHODS

The study design and methods were developed collaboratively among co-authors in an iterative manner. This resulted in the production of a project guidance document to help harmonize the project methods and ensure all project participants understood the principles and aims; and a standardized set of questions needed to calculate the indicators. A questionnaire was then developed using KoboToolBox (https://www.kobotoolbox.org/; a free and open-source tool for data collection and management) and used by participants to conduct their assessments (see Supporting Information S1). The resulting dataset was then downloaded as a.csv file and processed in R v. 4.2.1 using custom functions and a processing pipeline specifically developed for this study for quality checking, indicator calculation and subsequent analyses. All resources (questionnaire, pipeline and R code) used for this study are available from https://github.com/AliciaMstt/GeneticIndicators.

Species selection, alternative assessment of data sources, and data availability

Each country team aimed to assess 50–100 species, subspecies or similar (hereafter referred to as taxa) that represented different taxonomic groups, ecosystems, distributional range sizes (i.e., range-restricted or wide-ranging), conservation status and life history traits. As a means of quality control and to minimize assessor bias, each team incorporated some degree of multi-person calibration by reviewing a portion of the assessments. Alternatively, some teams adopted a multi-person approach for some taxa. While all countries followed the same principles and answered the same questions, discretion was given in the specific approach used in selecting taxa and country-relevant data sources (Table S1). For each taxon, metadata were recorded, including taxonomic group, ecosystem type (freshwater, marine, terrestrial), habitat type, range type (widespread, restricted), rarity, endemism, Global Red List category and several life history traits (fecundity, reproductive strategy, age at maturity, maximum or median lifespan).

In cases of variation in the number of populations or population sizes within a given taxon (either due to

uncertainties in the method used to define populations or differences in different data sources), more than one assessment was submitted for some taxa, referred to here as "alternative assessments".

For each country, the number and type of species assessed, the methods used to define populations within each, and the availability of population-level data (Ne or Nc) was explored. For species with population data, indicator values were then calculated. The indicator values from each taxon's *alternative assessments* were averaged, and a single value for each indicator was recorded. This method was chosen for standardization and simplicity; however, there may be other ways of utilizing and reporting alternative assessments.

Defining extinct and extant populations

To calculate the PM indicator, data on the number of extant (still in existence) and extinct populations is needed, thus needing a reference baseline time period. The monitoring framework of the GBF recommends Parties "use the period from 2011 to 2020, where data is available, as the reference period, unless otherwise indicated, for reporting and monitoring progress" (paragraph 2 of CBD, 2022b). However, the framework also notes that "baselines, conditions and periods used to express desirable states or levels of ambition in goals and targets should, where relevant, take into account historical trends". For this reason, we explored population extinction, considering a baseline period before the industrial era. Since the exact period representing this varies by country and may depend on the species and data availability, we suggested using a relatively broad baseline time period of 50–200 years ago for data retrieval and allowing for more specific baselines to be defined by countries. The origin of the population (e.g., natural, introduced; see Table S2) was considered, acknowledging that it is challenging to define populations when re-introductions or translocations have been done. Fragmentation of a once widespread range can also present challenges for defining the current and historic number of populations. For the Ne 500 indicator, population size data focused on the most recent available data per population. Because the data on the number of populations and their size may have been captured at different time points, the year(s) these data are associated with were recorded separately.

A checklist of six different methods typically used to define populations (a similar categorization is used by the IUCN Green Status for defining spatial units or populations: IUCN, 2020), plus an option to include additional approaches, was used: (1) Genetic clusters/clades, (2) Geographic boundaries, (3) Ecological or Biogeographic proxies, (4) Traits (e.g., behavioural, morphological, physiological), (5) Management Units, (6) Dispersal Buffers, and (7) Other (see Table S2 for definitions of each). Participants could select all methods that applied to each taxon and were required to accompany this with a brief narrative explaining how populations were defined.

Estimating the PM indicator

For species where the number of extinct and extant populations could be defined, the proportion of the number of maintained populations (currently present; extant) against the total number of known populations (sum of extant and extinct) was determined (i.e., PM indicator). For species where the number of extinct populations was classified as unknown, the PM indicator was not calculated. All subsequent analyses involving this indicator were conducted on this reduced dataset.

Estimating the Ne >500 indicator

Population size data were provided as Nc point estimates (i.e., count data), as semi-qualitative measures or as a range (e.g., "1000–2000", "<5000 by much"), or as Ne estimated from genetic data. For species with census or Ne data for at least one population, the Ne 500 indicator was calculated as a proportion of the number of populations with Ne>500 against the total number of extant populations for a species. When population sizes were known for some but not all populations, only populations with Ne or Nc data were considered in calculating the indicator.

For Nc data provided as a semi-qualitative measure or as a range, generalized Nc values were assigned to facilitate the computation of Ne, and hence the Ne 500 indicator. Populations noted as having slightly more than 5000 individuals were allocated a census size of 5500. This value is 10% above the minimum Ne threshold (i.e., Ne 500; Nc 5000, assuming a 0.1 Ne/Nc ratio). Populations classified as being substantially larger than 5000 individuals were assigned a value of 10,000 (double the minimum threshold). For populations estimated to have a census size of just under 5000, a value of 4500 was assigned, being 10% below the minimum threshold; and populations with considerably fewer than 5000 individuals were given a value of 500, by similar logic, indicating their increased risk of losing genetic diversity in the short term (corresponding to Ne = 50).

When Ne data were lacking but Nc data were available, a Ne/Nc conversion ratio of 0.1 was applied to roughly estimate contemporary Ne from Nc. While some species have been documented as having higher or lower conversion ratios, 0.1 has been found to be a conservative minimum threshold covering 95% of plants and 77% of animal species, indicating its applicability for most species (Frankham, 2021; Frankham et al., 2017; Hoban, Paz-Vinas, et al., 2021; Palstra & Ruzzante, 2008). For species with a known conversion ratio, this alternative

ratio was also applied to estimate Ne, and the results were compared.

Number of species being monitored using DNA-based methods

For each country, the third proposed genetic indicator (number of species being monitored using DNA-based methods: Hoban, Bruford, et al., 2021; Hoban et al., 2020; Laikre et al., 2020) was quantified. This is a count of the number of species for which DNA-based studies for at least one population are conducted, using data from two or more time periods (or planning to do so, with a dedicated budget) to investigate changes in the genetic diversity of species' populations.

Testing alternatives to the Ne/Nc 0.1 ratio

The Ne/Nc ratio of 0.1 has been recommended as a pragmatic rule of thumb that will work for most species, although median Ne/Nc values for some taxa are closer to 0.2. To explore the extent to which results depend on the use of the 0.1 Ne/Nc ratio, Ne was recalculated, assuming other Ne/Nc ratios (0.2 and 0.3) for the taxa where Nc point estimates were available.

Effect of method on defining populations and on indicator values

Considering the varied methods employed by each country to define populations and the expectation that wide-ranging species are likely to have more populations than species with narrow ranges, we assessed the impact of method and range type on the number of populations identified and on indicator values, using random intercepts to control for variation among countries. Specifically, generalized linear mixed models (glmer) and generalized linear mixed models via template model builder (glmmTMB) were conducted using the packages lme4 v. 1.1-31 (Bates et al., 2015) and glmmTMB v. 1.1.7 (Brooks et al., 2017). Details of each model and the code line used to run them are available in Supporting Information S3, Tables S6–S20.

In all of our analyses, we controlled for variation among countries in the mean value of our response variables (either number of populations or indicator values), as these are likely to vary in important ways due to the choice of taxa targeted.

Quantifying national indicator values

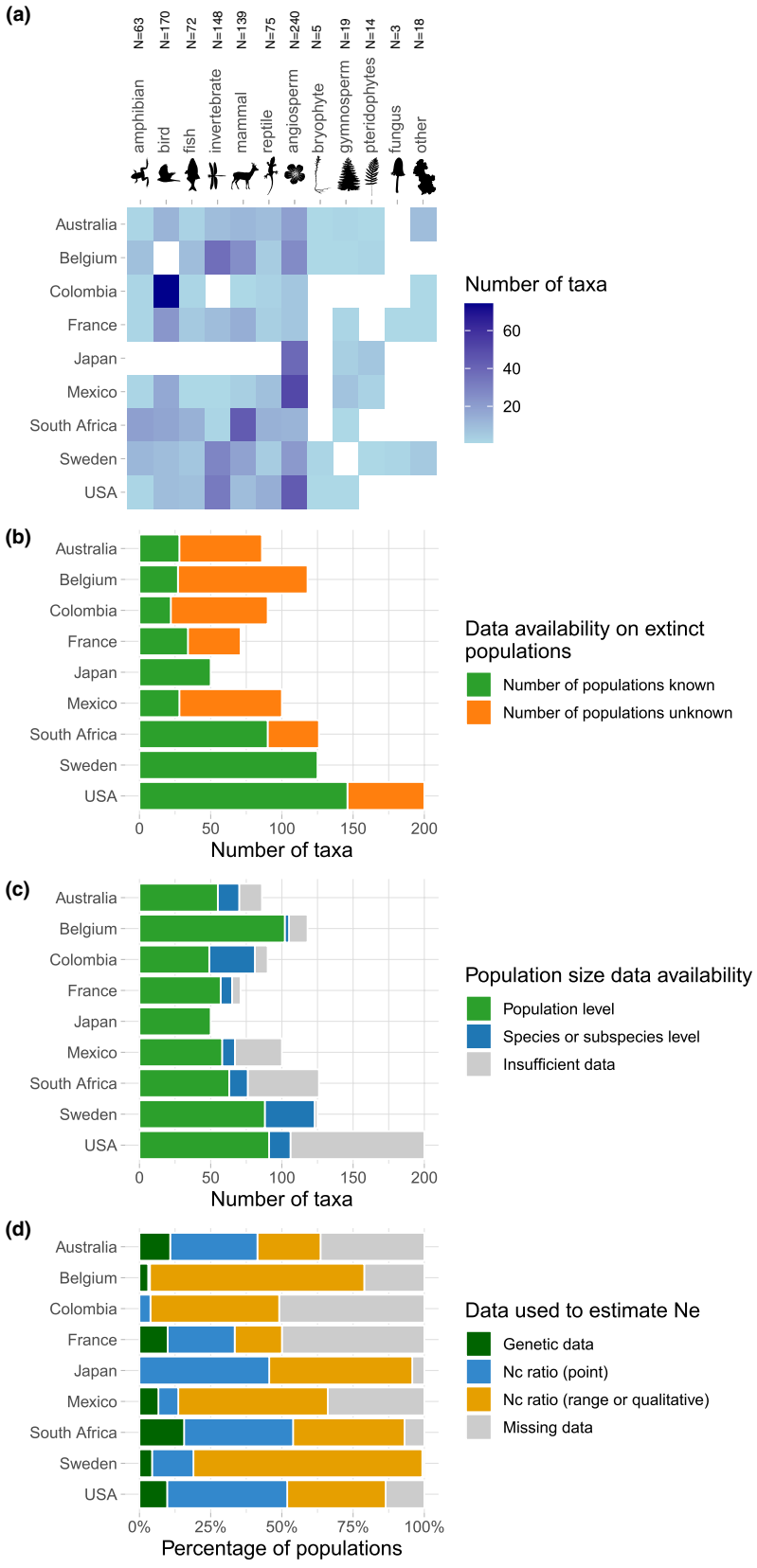
To illustrate how national indicator values can be obtained, for each of the indicators, we first averaged all available taxa within a given taxonomic group (simple equation [Equation 1] in Hoban, da Silva, et al., 2023) and then averaged these values, providing equal weight among taxonomic groups to the national level indicator (Equation 3 in Hoban, da Silva, et al., 2023). This latter equation accounts for the possible unequal representation of taxonomic groups (see Supporting Information S1 for an example).

RESULTS

Species selection, alternative assessment of data sources, and data availability

Discretion was given to country teams in the specific approach used for selecting taxa. For example, the Japanese and Colombian teams predominantly focused on a single taxonomic group (plants and birds, respectively; Figure 1a) to examine if they could leverage on-going monitoring projects, their most up-to-date curated data and informatics pipelines to estimate the genetic diversity indicators. Three general levels of data availability were observed across all countries: (i) data are stored in a centralized database, with little to no knowledge gaps; (ii) data are not stored in a centralized database but are available from various resources with some knowledge gaps; or (iii) little to no data are available. On average, a single taxon assessment took 3 ± 1.7 h to complete for most taxa (including time to find data). Relatively 'easy' taxa (i.e., where information was readily available and where populations were well defined geographically) took on average 2 ± 1.4 h to complete, while more difficult taxa took approximately 5.5 ± 3 h.

A total of 966 assessments spanning over 11 taxonomic groups (animals: amphibians, reptiles, birds, mammals, fish, invertebrates; plants: angiosperms, gymnosperms, bryophtyes, pteridophytes; fungi), representing 919 taxa (50 to 160 taxa assessed per country: Figures 1 and Figure S1a), identified 5652 populations (5271 after addressing alternative assessments, see below), with a mean of 628 ± 590 populations by country. The discrepancy between the number of assessments and their populations comes from 44 taxa that had alternative assessments conducted (91 in total; Figures S1b and S2). These alternative assessments stemmed from different sources of information reporting different values. For example, multiple assessments from the USA are the result of different analytical scales generally being applied in Species Status Assessments under the Endangered Species Act – "analytical or resiliency units" (AUs) and "adaptive or representation units", which are at a broader scale than AUs (e.g., representing ecoregions). Accordingly, alternative assessments for a single taxon were submitted to ensure that all data were considered and uncertainty incorporated (Figure S2). Alternative assessments per taxon ranged from 2 to 4, with a mode of 2.



Overall, 83% of taxa and assessments (765/919 and 802/966, respectively) had data to report on at least one of the two indicators (Figure 1b,c, Table S5). Approximately 57% of assessments (550/966) had data to report on the PM indicator (Figure 1b, Table S5). We found that variation in data availability is not attributable to taxonomic groups or the method used to define populations (Figure S3). Importantly, assessments from Sweden and

FIGURE 1 Taxa assessed and data availability by country. (a) A heat map showing the number of species or subspecies (taxa) assessed for a given taxonomic group within each country, counting taxa with alternative assessments once. (b) Total number of taxa with and without data on the number of extinct populations within a taxon, as needed for the PM indicator. Alternative assessments of a given taxon were counted separately. (c) Availability of population size data as needed for the Ne 500 indicator. Data were considered available if effective population size (Ne) or census population size (Nc) data was present for at least one population of a taxon (green bars), for the taxon as a whole ("species or subspecies level"; blue bars). Taxa without any Ne or Nc data were classified as having "insufficient data" (grey bars). Alternative assessments of the same taxon were counted separately. (d) Proportion of populations within each country with data on population size. The Nc ratio (point) represents count estimates or point approximate values (e.g., capture–recapture study found 3120 individuals). "Nc ratio (range or qualitative)" is more generic estimates of census size, either represented by quantitative ranges or qualitative descriptions of population size (e.g., "a few hundreds", ">5000 by much").

Japan focused on available data, and therefore, if no explicit evidence of population extinction was found, they assumed no population loss (Figure 1b). For the Ne 500 indicator, 613 assessments (~63%) had population size data (Nc or Ne) for at least one population within a species (Table S5; Figure S4), while 130 assessments (13%) only had census size data for the taxon as a whole (Figure 1c). While census size for an entire taxon within a country was not used in this study, the information was recorded as it could prove valuable. For example, if a census size of less than 5000 individuals is reported for a species, it can be inferred that each of its populations falls below the Ne 500 threshold. Additionally, the size might actually be based on aggregated data from individual populations that could not be located during the course of this study. Further efforts could then be made to disaggregate the data. Population size data were more commonly available for some taxonomic groups than others. For example, angiosperms, mammals and birds had more data available compared to invertebrates (Figure S5).

Census data made up the vast majority of population size information used to quantify Ne (22% and 53% of 4240 populations assessed for point and for range or semi-qualitative estimates, respectively; Table S5). In contrast, only 6% of populations within species had Ne estimates that were based on genetic data (349/5652 populations assessed; Figure 1d, Table S5).

Defining extinct and extant populations

Populations were defined using a variety of methods across taxonomic groups for all nine countries (Figures 2 and 3a). Because of this, and realizing species range size could affect the number of populations identified and the associated indicator values, we assessed the interactions of these variables while controlling for variation among countries (see Supporting Information S3, Tables S6–S20). Wide-ranging taxa were found to have significantly more populations than range-restricted taxa when controlling for the method used to define populations (Supporting Information S3, Figure S6 and Tables S6–S8). Of the methods used to define populations, "genetic clusters" tended to identify a smaller number of populations that encompassed larger geographical areas compared to all other methods (Figure S6, Table S6).

Effect of the method on defining populations and on indicator values

Taxa where the "genetic clusters" method was used to define populations (either alone or in combination with other methods) had a significantly higher PM indicator value compared to when other methods were used ($p=0.039$; Figure 3b, Table S9). After controlling for species range, the method used to define populations was no longer a statistically significant predictor of the PM indicator (Figure 3d,e, Table S14).

Like the PM indicator, taxa where the "genetic clusters" method was used to define populations (either alone or in combination with other methods) had a significantly higher Ne 500 indicator value compared to when this method was not used ($p=0.028$; Figure 3c, Table S15). However, in contrast to the PM indicator, the "genetic clusters" method still produced higher values for the Ne 500 indicator even after controlling for species range (Table S20). No other consistent relationships between methods and indicator values were found.

Estimating the PM indicator

Globally, we found that 41% of taxa ($n=211/518$) for which we could estimate the PM indicator have lost at least 1 out of every 10 of their populations (PM indicator <0.9) during the timescale of the study (e.g., last 50–200 years) and 3% ($n=15/518$) have lost 3 out of 4 or more populations (PM indicator <0.25; Figure 4a,b).

Estimating the Ne 500 indicator

Of the 4589 populations assessed with Ne data (either through proxies [Nc point or range estimates] or actual measures of Ne), 84% were below the Ne 500 threshold. Of the taxa with Ne data, 58% ($n=330/568$) had all of their populations below the threshold (Ne 500 indicator=0; Figure 4d), and less than 19% ($n=106$) had all of their populations above Ne 500 (Ne 500 indicator=1; Figure 4d). Similar trends for both PM and Ne 500 indicators were found for all taxonomic groups (Figure 4a,c). The values of both indicators (from 0 to 1) are heterogeneously distributed across seven of the IUCN Red List categories reported in this study, from Least Concern to

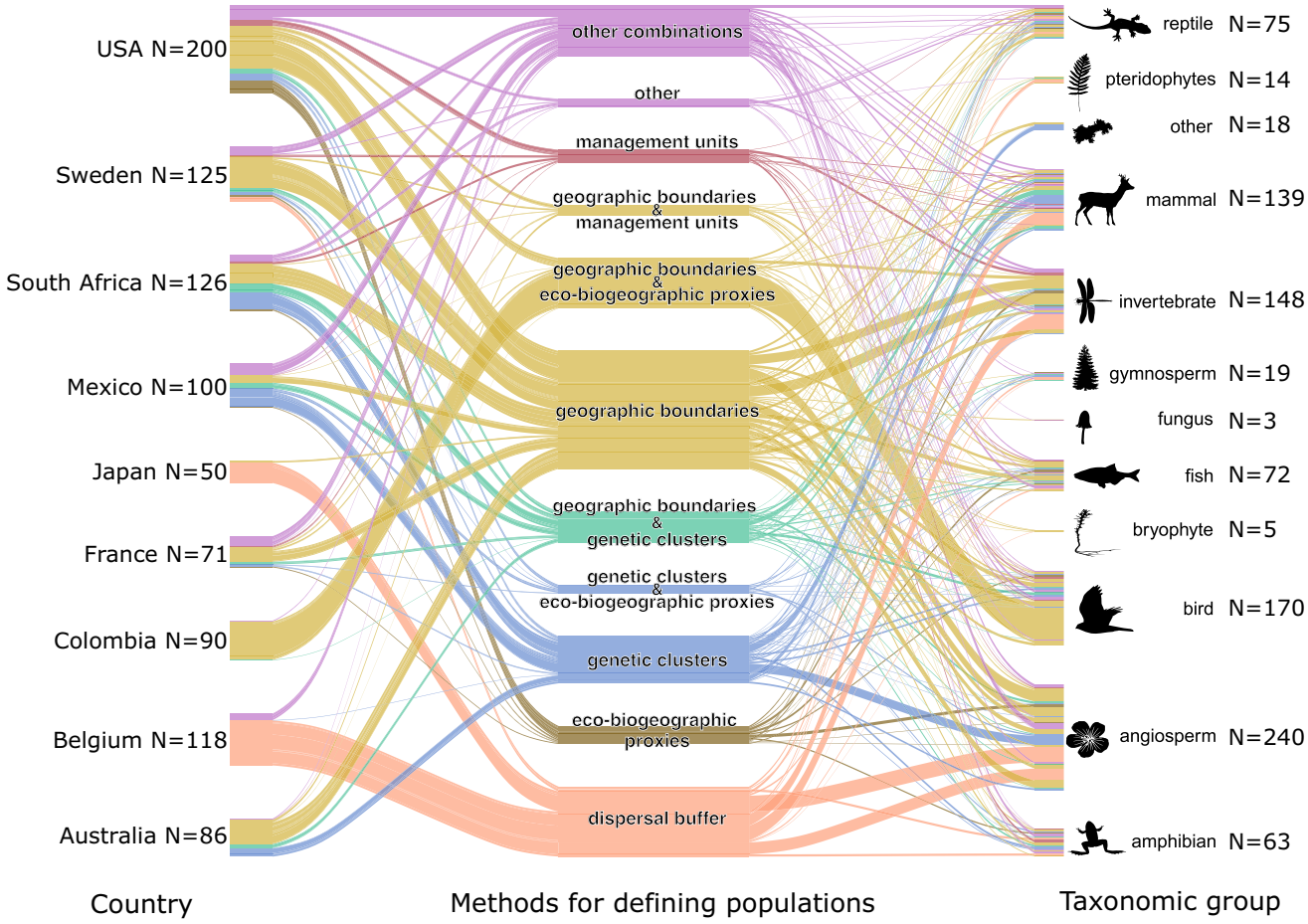


FIGURE 2 Alluvial plot illustrating the various methods used to define populations for each participating country and taxonomic group. Each method is assigned a unique colour (centre), and every species or subspecies (i.e., taxa) assessed is represented by a single (thin) line. Starting from the centre (method), each line (taxa) can be followed to the left to show its assessment country, and to the right to indicate its taxonomic group. Therefore, the height of any given category (country, method, taxonomic group) is indicative of the number of taxa falling within it, and the number of colours present within each category illustrates the variation in methods used by a country or for a taxonomic group. For example, 'geographic boundaries' was the most used method for defining populations and was employed by all but one country and used in all taxonomic groups. Also, some taxonomic groups were assessed by a diversity of methods, like reptiles, while others were dominated by a certain method, like geographic boundaries in birds.

Critically Endangered (Figure 5). No particular Red List category had only very high or very low values. For example, indicator values at or near 0 were not confined to Endangered taxa, but were also found in Least Concern taxa (Tables S21 and S22).

Number of species being monitored using DNA-based methods

The genetic monitoring indicator was reported by eight of the nine countries in this study, with 5–20 species currently being monitored genetically by those countries (Figure 6b).

Testing alternatives to the Ne/Nc 0.1 ratio

For the 12 species with a known Ne/ Nc ratio, indicator values did not change compared to when the 0.1 ratio was used, except for one species, *Synercus caffer caffer* (Cape Buffalo), where the 0.1 ratio overestimated the proportion of populations below Ne 500 (Table S23). For the 197 assessments and 1303 populations with an Nc point estimate where Ne was recalculated assuming less conservative Ne/Nc ratios (0.2 and 0.3; Figure S8), the distribution of indicator values shows only small changes. Thus, the main conclusion that most species have an Ne 500 indicator value of 0 and the large majority have indicator values below 0.5 holds (Figure S8).

Quantifying national indicator values

Overall, the aggregated indicator values suggest that for most of the participating countries, the majority of taxa have not lost many populations (PM >0.90; less than 10% loss); however, a large percentage of the populations remaining are too small to maintain genetic diversity

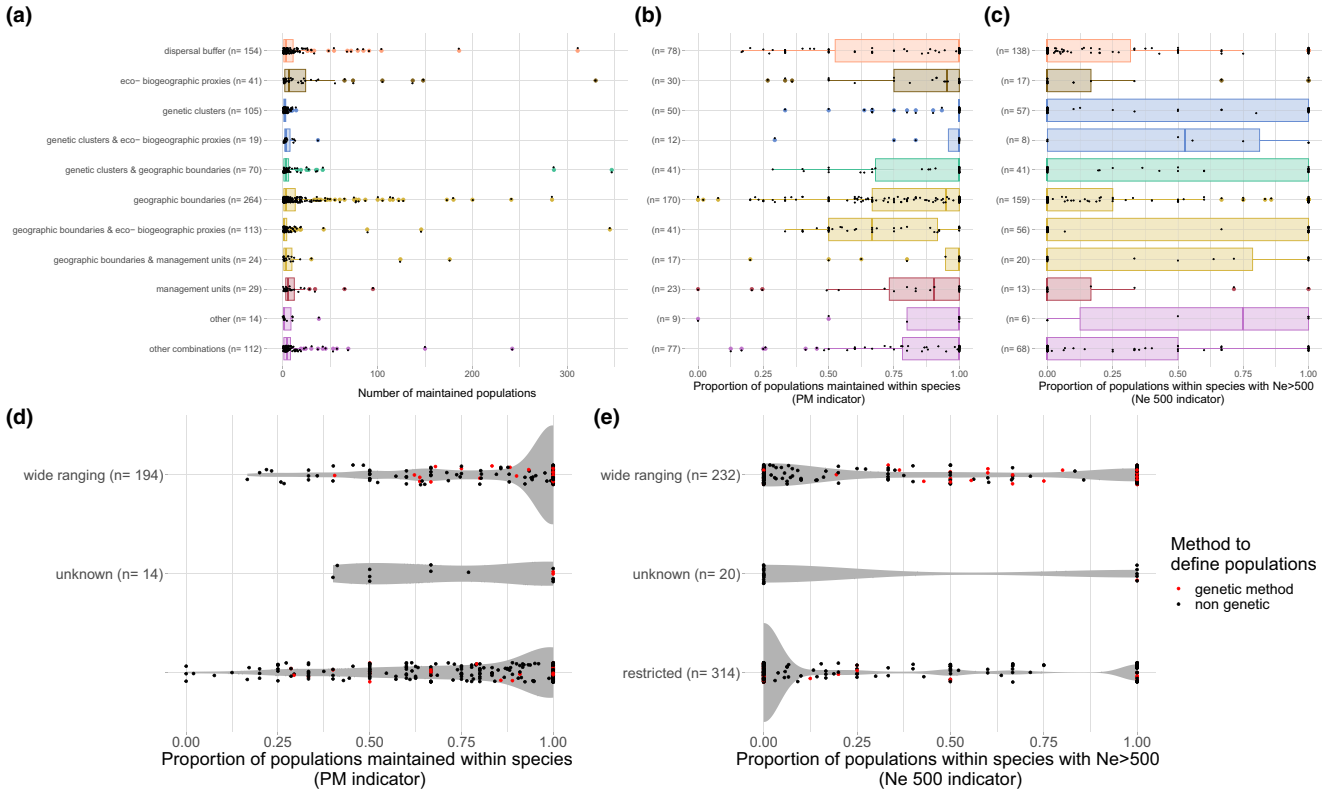


FIGURE 3 Aggregated results across all nine countries examining associations between the methods used to define populations, the number of populations maintained for any given taxon, and the indicator values within a taxon. (a) Boxplot showing the spread in the number of extant populations for each method applied; (b, c) Boxplots showing the range in indicator values across each of the methods applied, for the PM and Ne >500 indicator, respectively; and (d, e) Violin plots showing the range in indicator values across species range types, for the PM and Ne >500 indicator, respectively. In all plots, each dot represents the indicator value for a single assessment. Red dots highlight taxa where genetic methods, alone or in combination with others, were used to define populations. *n*, sample size, is shown to the left of each plot. Outliers with more than 500 populations were removed from these plots and statistical analyses.

(Ne 500 for all countries <0.42; Figure 6B). Moreover, the majority of countries were found to conduct temporal genetic monitoring studies on at least a few taxa (Figure 6). While the numbers reported are seemingly low, all countries are conducting other genetic types of studies (Figure S9). All of these indicator values can be disaggregated by taxonomic group, as illustrated by the South African example (Figure 6c), to investigate which groups are influencing these values and need more urgent attention.

DISCUSSION

Data are available and it is feasible to report on the genetic diversity indicators

In less than a year, we assessed the conservation status of genetic diversity in 919 taxa from nine countries, finding data to report on either the PM or Ne 500 indicator in 83% of taxa (Figure 1b,c). This demonstrates that data are available and that it is feasible to report on the genetic diversity indicators of the adopted GBF.

For the PM indicator, we found that country taxa selection and the exact baseline time period determine whether the number of extinct populations is available and, consequently, the number of taxa for which this indicator could be calculated. The baseline period to measure biodiversity loss has been the subject of intense debate because baselines represent technical and political decisions that can have a profound influence on outcomes and perceived responsibility (Donadio Linares, 2022). It has been shown that scientific efforts are best suited to assess biodiversity loss using baselines as distant as possible, especially considering the periods when drastic environmental changes started in each region (Donadio Linares, 2022). Based on our data availability findings, countries may struggle to report the PM indicator when considering more historical periods without significant effort. Meanwhile, evaluating the PM indicator using recent data could greatly help to establish a current baseline to prevent further genetic diversity loss through population extinctions.

Regarding data availability for the Ne 500 indicator, we found that relatively few populations have Ne estimated from genetic data. While this is unsurprising, we were still able to estimate Ne for 4240 populations using

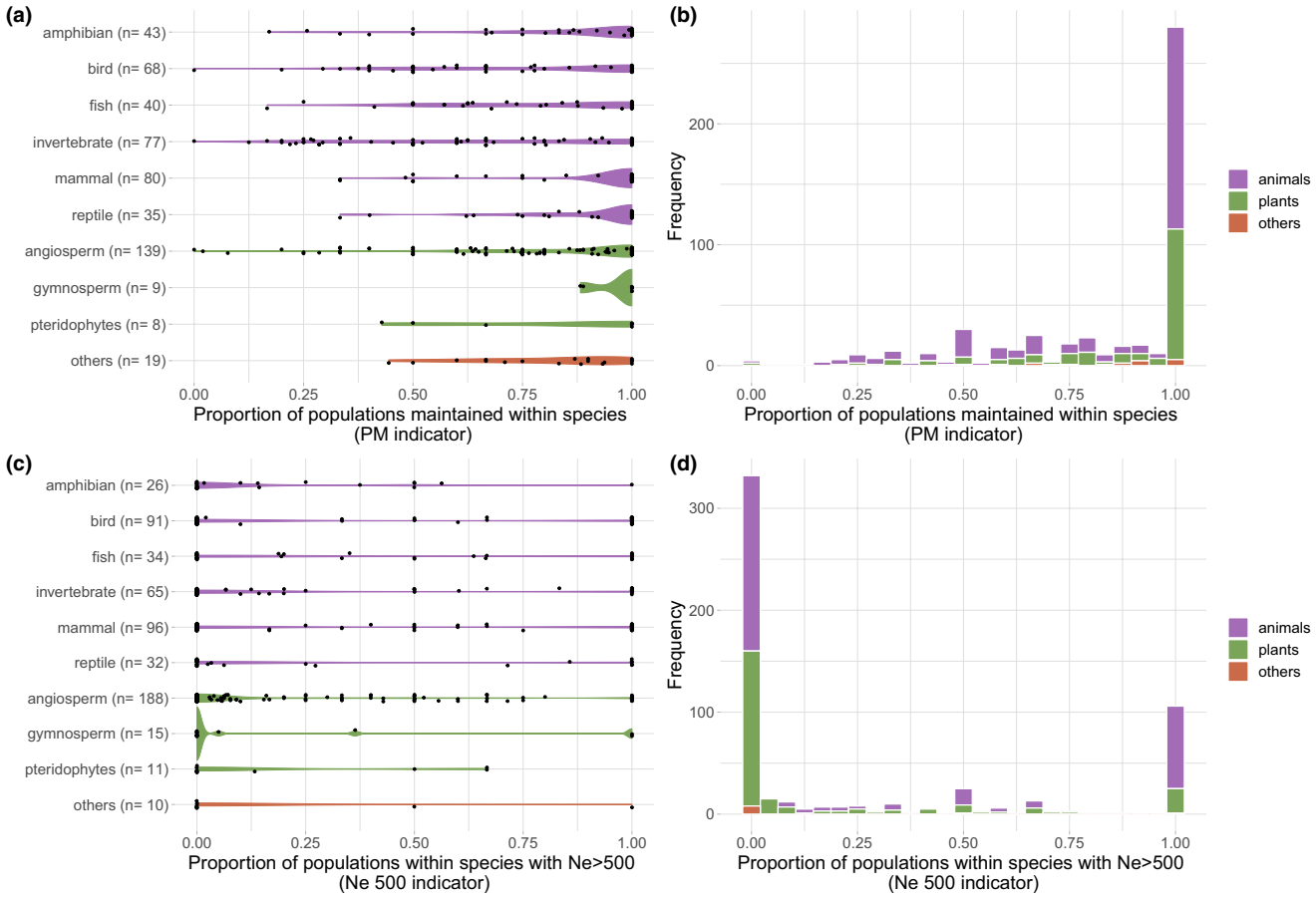


FIGURE 4 Aggregated results across all nine countries show the indicator values across taxonomic groups. The spread in indicator values is shown in the violin plots for the (a) PM indicator and (c) Ne >500 indicator across taxonomic groups, as well as the frequency barplots, grouped according to Kingdom (b, d). In (a) and (b), each dot represents the indicator value for a single assessment, with the sample size, *n*, for each taxonomic group provided.

Nc data and a Ne/Nc ratio of 0.1, an acceptable proxy when the ratio is unknown (see Table S23). To put these numbers into perspective, a recent review of genetic studies measuring Ne found that 712 papers published between 2006 and 2020 estimated Ne in around 3500 populations (Clarke et al., 2024). In other words, our proxy-based assessment, which was completed in less than one year, obtained more estimates of Ne than a decade of hundreds of DNA-based studies. We acknowledge that Ne quantified using genetic data may, in some cases, be more accurate than that estimated using proxy data, and that genetic data is crucial for assessing the effect of genomic erosion (e.g., Femerling et al., 2023). However, using proxies, we have shown that estimates of Ne can be obtained in a rapid and efficient manner, which can enable the tracking and reporting of genetic information on large scales in all countries. Further work examining the relationship between proxy and DNA-based data will help refine the indicator. We therefore emphasize that both DNA-based and proxy data are important and useful for genetic conservation action and should be used to complement one another. We also note that Ne or Nc data, when available for a taxon, was sometimes not available for all populations. An indicator based on only some populations of a species is an imperfect summary, though it is hard to determine whether this could create a significant systematic bias in the data.

An important feature of our dataset is that it includes range or semi-qualitative Nc data. This not only increases data availability but also allows local knowledge holders, including indigenous peoples and local communities, to contribute to these assessments. As one of many examples, the Nc range data for several Mexican plants were obtained by consulting with park rangers, botanists or citizen scientists who are active at the local level. Such population size estimates may not be quantitative enough for conventional ecological and evolutionary analyses, but they may be sufficient to ascertain if a population is above or below Ne 500. Moreover, because these estimates are at the population level (i.e., individual populations that locals know well), they may be more robust than size estimates encompassing an entire species' range, which typically entail greater inherent uncertainty and assumptions (Jędrzejewski et al., 2018; Wilson et al., 2011). Additionally, incorporating local knowledge into the genetic indicators assessments may

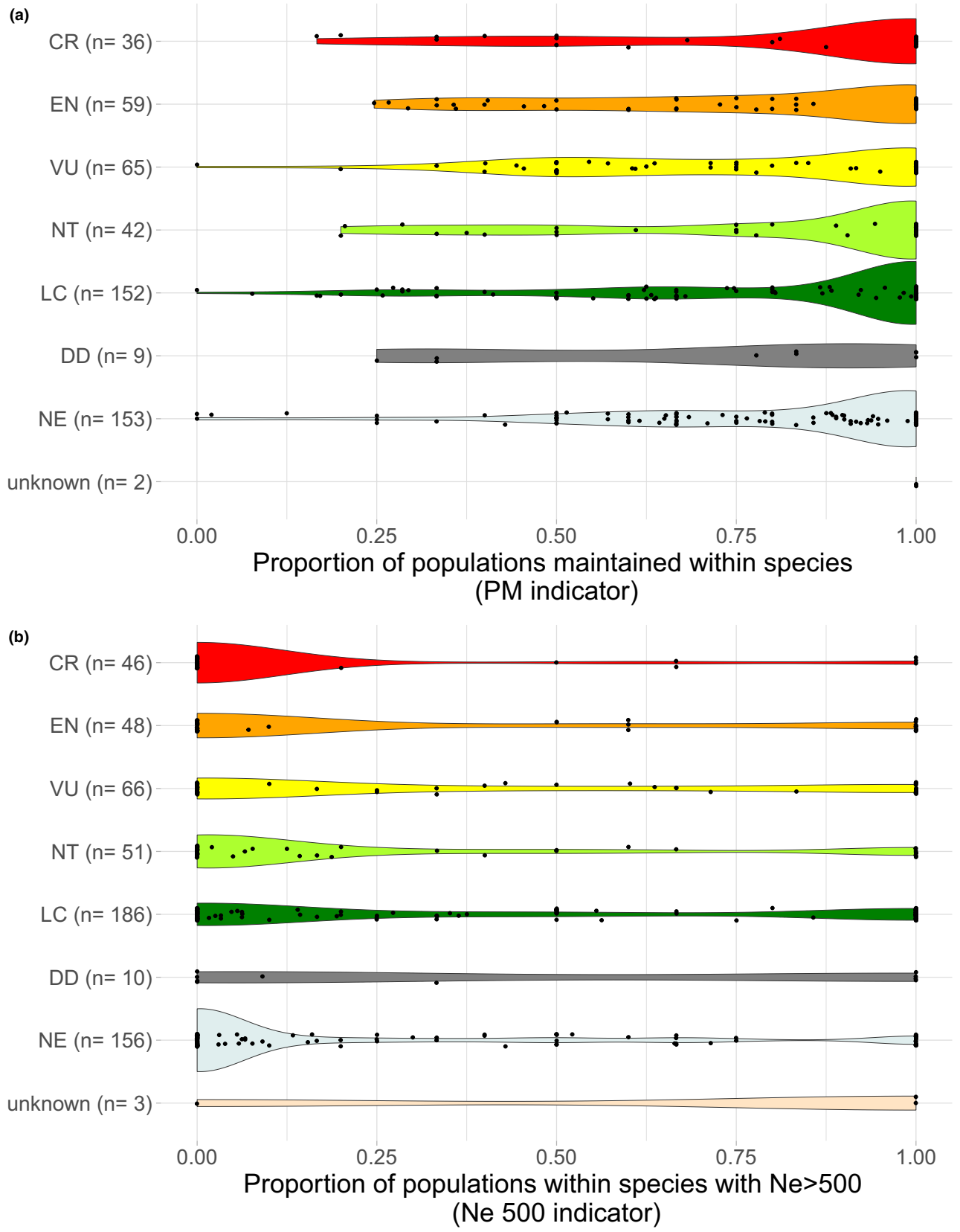


FIGURE 5 Violin plots illustrating the spread in (a) PM and (b) Ne >500 indicator values across IUCN Red List categories. Species were classified by their Global Red List status. Abbreviations reflect official IUCN Red List categories. Sample sizes (*n*) are provided for each threat category.

(a)

	Indicator value = 0 (worst scenario)	Indicator value = 1 (best scenario)
PM	all population lost within a species	all populations within a species are maintained
Ne 500	all populations within a species are below Ne 500	all populations within a species are above Ne 500

(b)

Country	PM	Ne 500	Genetic Monitoring
Australia	0.90 ± 0.18 (28)	0.17 ± 0.30 (47)	10
Belgium	0.45 ± 0.22 (27)	0.25 ± 0.38 (101)	10
Colombia	0.60 ± 0.17 (22)	0.33 ± 0.47 (43)	*Not evaluated*
France	0.85 ± 0.28 (34)	0.42 ± 0.47 (55)	7
Japan	0.93 ± 0.15 (50)	0.08 ± 0.18 (50)	0
Mexico	0.94 ± 0.14 (28)	0.22 ± 0.35 (47)	7
South Africa	0.95 ± 0.16 (90)	0.42 ± 0.48 (61)	5
Sweden	0.78 ± 0.27 (120)	0.19 ± 0.33 (83)	20
USA	0.79 ± 0.24 (117)	0.35 ± 0.41 (79)	6

(c)

Taxonomic group	PM	Ne 500	Genetic Monitoring
Amphibian	0.92 ± 0.17 (18)	0.13 ± 0.25 (4)	2
Bird	1.00 ± 0.00 (11)	0.33 ± 0.47 (11)	1
Fish	1.00 ± 0.00 (9)	0.30 ± 0.48 (4)	0
Mammal	0.99 ± 0.04 (32)	0.61 ± 0.48 (31)	2
Reptile	0.87 ± 0.25 (7)	1.00 (1)	0
Angiosperm	0.83 ± 0.28 (12)	0.06 ± 0.19 (10)	0

FIGURE 6 Summary of values for the three genetic diversity indicators for the taxa assessed. (a) Conceptual table showing extreme values for the proportion of maintained populations within species (PM indicator) and the proportion of populations within species with an effective population size (Ne) greater than 500 (Ne 500 indicator). (b) Example of how countries can summarize their national indicator values for three genetic diversity indicators for the species assessed here. Mean indicator values±standard deviations are provided for the PM and Ne 500 indicators. Sample sizes used to quantify these two indicators are provided in brackets. (c) Demonstration of disaggregation by taxonomy with indicator values for South Africa broken down by taxonomic group.

help strengthen community-based conservation efforts by highlighting the value of local action. For example, Nc data from one of the two remaining populations of *Xenospiza baileyi*, an endangered bird from Mexico, comes from participatory monitoring run by "Brigada de Monitoreo Biológico Milpa Alta", a local community organization of San Pablo Oztotepec (Hoban et al., 2024).

Our findings also show that a variety of methods can be used to practically delineate populations; however, species range type should be considered when interpreting the results. This is because populations spanning larger geographic areas tend to have more individuals compared to populations occupying smaller areas. This had a minor effect on the Ne 500 indicator value when the "genetic clusters" method was used due to this method sometimes identifying larger geographic areas as a single population. The fewer populations identified may stem from influences such as recent human-induced fragmentation (hence may not be detectable genetically) and our use of diverse approaches to identify populations, which could result in an overestimation of distinct populations compared to those identified based on genetic data. A potential solution to improve the representation of genetic diversity in widely distributed taxa when no genetic evidence is available is to account for uncertainty by defining populations with different methods, such as occurrence over different life zones (e.g., Khoury et al., 2019; Tobón-Niedfeldt et al., 2022), and subsequently calculating averages or displaying confidence intervals. In practice, countries will need to document the chosen method transparently so that the same approach can be applied when re-evaluating the taxa over time. Initiating monitoring programmes now could enable countries to detect future changes in population structure. We also note that assessments of genetic clusters should consider whether genetic differences are due to recent bottleneck effects, which can make small populations appear genetically distinct (Clarke et al., 2024; Liddell et al., 2021; Weeks et al., 2016).

The genetic diversity indicators reveal a loss of diversity otherwise unnoticed

This first multinational assessment of genetic diversity indicators has shown that 41% of the assessed taxa have lost at least one-tenth of their populations and that in 58% of taxa, all populations are too small to sustain genetic diversity (Figure 4). These aspects of genetic diversity loss may go unnoticed under other species-level assessment criteria (e.g., endangered species lists or IUCN Red Lists; Figure 5); we found numerous taxa with Least Concern or Near Threatened Red List status but low genetic indicator values.

With each extinct population, unique genetic diversity may have disappeared, so even if the species is re-introduced to an area at a later stage, the genetic diversity of the species would likely not be fully recovered. Loss of populations can also affect the biotic interactions within ecosystems, which can have profound

cascading consequences ranging from co-extinctions to the loss of ecosystem services (Young et al., 2016). Early estimates suggested that population loss in tropical forests could occur 3–8 orders of magnitude more rapidly than species loss (Hughes et al., 1997), and yet this loss of diversity is seldom reported. The PM indicator allows tracking of these losses and can inform corrective and preventative actions. Our results are less severe than the range losses reported by Ceballos et al. (2017), which found that all 177 mammals examined had lost at least 30% of their range size, though the authors acknowledge that most of their species were medium- to large-sized. However, we found that 53% of taxa ($n = 277/518$) still maintain all of their populations (PM indicator = 1; Figure 4b), suggesting that many species may retain a substantial amount of range-wide adaptive capacity for now, though much genetic diversity within populations has been lost (as noted below). However, note that the number of extinct populations (Figure 1b) was unknown for 43% of taxa, which means that we could be underestimating population loss, especially when considering older time baseline periods. Also, note that the PM indicator complements other metrics of species' decline, such as changes in the area of occupancy.

Although the findings of the PM indicator suggest population stability for some taxa, the Ne 500 indicator shows that the vast majority of populations analysed are below a threshold for maintaining genetic diversity and may have already lost substantial diversity (Ne threshold = 500; Figure 4c,d). Importantly, even in wide-ranging taxa, the Ne 500 indicator is skewed towards lower values (Figure 3e). This is worrisome because wide-ranging species are thought to be of less conservation concern (Staude et al., 2020). For instance, the maize wild relative *Zea mays* ssp. *parviglumis* was listed as Least Concern on the IUCN Red List; however, there are populations of high concern within it (Rivera-Rodríguez et al., 2023).

While Ne 500 does not signal the immediate decline of a species' genetic health, it is the point at which genetic erosion starts to accelerate and adaptive capacity declines (Crow & Kimura, 1970; Frankham et al., 2014; Jamieson & Allendorf, 2012), and hence the point at which management intervention could prevent any further loss of genetic diversity. A more consequential threshold occurs at a lower Ne (e.g., 100 or 50), where inbreeding becomes pronounced in the short term (with associated risk of inbreeding depression and extinction; Frankham et al., 2014; Franklin, 1980). Of the 1615 populations with Ne, or point estimates of Nc (Table S5), 57% had Ne <50, which indicates most of the populations had substantially low genetic diversity levels. Despite these consequential findings, we acknowledge that some taxa and their associated populations may have survived in small populations without significantly compromising their genetic health (e.g., through the purging or limited accumulation of deleterious alleles), such as the Ethiopian wolves *Canis simensis* (Mooney., 2018). However, to date, these cases are few, and small populations typically remain at higher risk of extinction due to stochastic processes and catastrophes (Kardos et al., 2023; Lande, 1993) or struggle to increase in size due to inbreeding depression (Kardos et al., 2023).

Considering that the success of species conservation depends on local decisions affecting each population (Collen et al., 2011; Ehrlich & Daily, 1993), the genetic diversity indicators are not only useful to report on the genetic status of species, but may also be able to help inform and prioritize action and policy for populations, species or even geographic regions with high conservation needs. For instance, genetic data are helping inform current and future management of the numbat *Myrmecobius fasciatus* (a small Australian marsupial) across remnant and translocation sites (Northover et al., 2023).

Towards addressing genetic diversity conservation at a global scale

Our results show it is feasible and affordable to estimate the GBF genetic indicators for varied countries, for a wide range of taxonomic groups and using existing data. Our dataset does not necessarily represent what the participating countries will report in their National Reports to the CBD. Instead, it illustrates the practical scale of effort needed for such reporting. The collaborative experiences and insights obtained throughout this project can be useful to other nations for integrating genetic diversity into their national reporting and policies (e.g., National Reports and National Biodiversity Strategy and Action Plans) and can hopefully inspire and facilitate further collaborative processes across stakeholder groups, which have been shown to effectively expedite and support species conservation (Lees et al., 2021).

CBD Parties have asked what species should be included to assess the indicators at the country level and how long the assessments take (Hoban et al., 2024). Here, we have shown that it is indeed feasible to assess more than 100 species per country that reflect diverse ecosystems, taxonomic groups, range types and life history traits in a fairly short period of time. While national metrics were calculated for each of the nine countries, these values are based on the species chosen for this study. For official CBD reporting, the species selected by countries could focus on genetic diversity monitoring (Hvilsom et al., 2022) or be a subset of other lists that countries already use for monitoring or conservation priority-setting. Some bias in species selection is expected and may be acceptable as long as countries clearly document the rationale behind their selection. Concerns about bias should not prevent initial assessments of the indicators since the first effort helps a country set up the infrastructure and methods of data gathering and analysis. Parties can summarize

the indicators at an overall national level or aggregate them according to taxonomic group, ecosystem type or conservation status (as demonstrated in Figure 6). Although we did not include domesticated species (we did include crop wild relatives, managed, and semi-domesticated species), the indicators are also applicable to domesticated taxa and breeds, so they could be used by countries to evaluate the loss of breeds and the sufficient Ne of breeds. We have shown that assessing non-threatened species is critical because these indicators could reveal genetic diversity loss that might otherwise go unnoticed. We note that genetic indicators could be used for other purposes, such as delimiting Key Biodiversity Areas, within-country species prioritization, tracking changes after management interventions (as noted by Hoban et al., 2024) or to be included in IUCN's Green Status by informing and increasing the sensitivity of the Species Recovery Score.

Importantly, although the PM indicator was adopted as a complementary (non-mandatory) indicator, we encourage Parties to report this indicator jointly with the Ne 500 indicator. In order for Goal A and Target 4 to be fully achieved with respect to genetic diversity – maintain and restore the genetic diversity *within* and *between* populations […] to maintain their adaptive potential – it is required that both Ne 500 and PM indicators be reported. If the Ne 500 indicator is used alone, it must be adjusted to incorporate local population loss (see detailed discussion at Hoban et al., 2024). The Ne 500 indicator is estimated at the population level, which implicitly involves population delimitation. Therefore, the effort to estimate the PM indicator when data is available for estimating the Ne 500 would involve minimal additional work.

The fact that genetic studies are not needed to estimate these indicators does not mean genetic data are not needed or desired. Both are complementary to each other (see discussion in Hoban et al., 2024). The PM and Ne 500 indicators can point countries towards which species or populations need genetic examination, either because census data shows they are too small and hence genetic studies are needed to guide management (e.g., identifying which individuals should be used in breeding programmes or translocations) or because other processes not covered by the PM and Ne 500 indicators may be affecting the genetic diversity of these populations (e.g., gene flow between crops, crop wild relatives and genetically modified organisms, Wegier et al., 2011). Consequently, genetic studies will remain an important source of information and are critical to the management of species. For this reason, we also recommend reporting on the DNA-based genetic monitoring indicator (Hoban et al., 2020; Hoban, Bruford, et al., 2021; Hoban, da Silva, et al., 2023), as was done here (Figure 6b). While this indicator is not currently listed among the complementary and component indicators in the global monitoring framework, it is useful to track the efforts being undertaken by countries to use DNA-based methods to monitor genetic diversity, as it can help inform adaptive species management and conservation policy (Posledovich et al., 2021; Schwartz et al., 2007). In Mexico, for example, genetic monitoring focuses on crop wild relatives where gene flow with genetically modified organisms and improved varieties is a concern (Rivera-Rodríguez et al., 2023; Rojas-Barrera et al., 2019; Wegier et al., 2011). In South Africa, genetic monitoring programmes typically focus on threatened species (e.g., da Silva & Tolley, 2018; Labuschagne et al., 2016; Stephens et al., 2022) or species of cultural and/or economic interest (e.g., de Jager et al., 2020; Miller et al., 2020). Moreover, Sweden recently initiated a national genetic monitoring programme (Andersson et al., 2022; Johannesson & Laikre, 2020) with species such as cod, salmon and moose, which are heavily harvested, to help prevent their collapse (Dussex et al., 2023; Johannesson & Laikre, 2023). Reporting on this monitoring indicator could help incentivize more such studies within a country, as well as incentivize others to start implementing DNA-based monitoring (Hoban et al., 2024). Indeed, genetic studies exist for many of the taxa assessed in this study, which could form the basis for future genetic monitoring (Figure S9).

Practical considerations

Based on the results of this study, we estimate that the three genetic diversity indicators could be assessed for 100 species in around 300–400 h (around 3 h/ species). This is orders of magnitude faster than what it takes to perform conventional genetic studies. However, if coordinated with other processes, it is anticipated that this time could be further reduced. For instance, of the 136 countries that submitted the 6th CBD national report, 61 have a national Red List for at least one taxonomic group and 62 other nations are currently in the process of establishing one (Raimondo et al., 2023). If the experts are already gathered for Red Listing workshops and the relevant data, similar to the data we employed in this study, is accessible to them, we estimate that assessing the genetic diversity indicators may only take an additional 10–20 mins per species. Additionally, the tools and resources developed and used here were improved following the lessons learned from this study and will be a dynamic, collaborative resource with updates provided as needed at https://ccgenetics.github.io/guidelines-genetic-diversity-indicators/ (Mastretta-Yanes et al., 2024).

Further advances could link existing biodiversity databases, species spatial predictions of density and distribution (Jędrzejewski et al., 2018) and earth observation data (Schuman et al., 2023), in a semi-automated process to further reduce the time needed

for these assessments, depending on a country's existing capacities and infrastructure. Capacity-building needs will depend largely on what data are available within a country. Available data should be considered broadly, including citizen science, grey literature, local experts' knowledge, and informal data held by small NGOs and local communities, and not only data coming from scientific studies (Hoban et al., 2024).

In closing, we have shown that the calculation of these genetic indicators is feasible and invaluable in monitoring a level of biodiversity otherwise unnoticed. The genetic diversity indicators may provide data that can contribute to cutting-edge avenues for research in ecology and evolution, including the effects of past demographic history on how species cope with small contemporary Ne, as well as the genetic basis of how populations adapt to changing conditions. Meanwhile, in the context of increasing environmental changes in the Anthropocene, the present assessment highlights a consequential trend—that although the majority of species assessed have so far maintained the majority of their populations, many species around the world are on the precipice of decline in genetic diversity because their populations are not sufficiently large. Yet, this trend also presents an opportunity to safeguard and enhance genetic diversity to protect and maintain species and populations around the world.

AUTHOR CONTRIBUTIONS

SH, JMdS, AM-Y and LL conceived the idea. SH, JMdS, and AM-Y coordinated the overall project. While the study design and methods (including project guidance documents, an instruction manual and a questionnaire) were developed collaboratively in an iterative manner among country leads and other core team members (MEH, AJM, PS-G), JMdS and AM-Y developed the initial drafts of the online data collection tool and manuscript. Data collection was coordinated by country leads. Data analysis and interpretation was coordinated by AM-Y, JMdS, CEG, LC-R and VK, with AM-Y leading the development of the pipeline and R code. All authors completed at least 10 assessments, as well as reviewed and/or contributed to revisions of the manuscript. Country leads were: CEG and RJ (Australia); JM and LC-R (Belgium); VJR-P and MAR-M (Colombia); MH and IP-V (France); FI (Japan); AM-Y (Mexico); JMdS (South Africa); LL and HK (Sweden); BRF, WCF, and SH (USA).

AFFILIATIONS

[1]Consejo Nacional de Humanidades Ciencias y Tecnología (CONAHCYT), Ciudad de México, Mexico
[2]Comisión Nacional para el Conocimiento y Uso de la Biodiversidad (CONABIO), Ciudad de México, Mexico
[3]Wildlife Ecology and Management, University of Freiburg, Freiburg im Breisgau, Germany
[4]South African National Biodiversity Institute, Kirstenbosch Research Centre, Cape Town, South Africa
[5]Centre for Ecological Genomics and Wildlife Conservation, University of Johannesburg, Johannesburg, South Africa
[6]Faculty of Science, School of Life and Environmental Sciences, The University of Sydney, Sydney, New South Wales, Australia
[7]Ecology, Evolution and Biodiversity Conservation, Department of Biology, KU Leuven, Leuven, Belgium
[8]Stockholm University, Stockholm, Sweden
[9]U.S. Fish and Wildlife Service, Fort Collins, Colorado, USA
[10]Department of Biology, Colorado State University, Fort Collins, Colorado, USA
[11]Graduate Degree Program in Ecology, Colorado State University, Fort Collins, Colorado, USA
[12]BIOGECO, INRAE, University of Bordeaux, Cestas, France
[13]National Institute for Environmental Studies, Tsukuba, Ibaraki, Japan
[14]CSIRO Environment, Sandy Bay, Tasmania, Australia
[15]Research Institute for Nature and Forest, Geraardsbergen, Belgium
[16]Université Claude Bernard Lyon 1, LEHNA UMR 5023, CNRS, ENTPE, Villeurbanne, France
[17]Alexander von Humboldt Biological Resources Research Institute, Bogotá, Colombia
[18]Department of Biology, Faculty of Science, Javeriana University, Bogotá, Colombia
[19]Independent Mountain Ecology Researcher, Michoacan, Mexico
[20]Department of Agricultural Biology, Colorado State University, Fort Collins, Colorado, USA
[21]Committee on Evolutionary Biology, University of Chicago, Chicago, Illinois, USA
[22]Center for Tree Science, The Morton Arboretum, Illinois, USA
[23]U.S. Geological Survey, Wetland and Aquatic Research Center, Gainesville, Florida, USA
[24]Conservatoire d'espaces Naturels d'Occitanie, Montpellier, France
[25]Instituto de Investigaciones en Ecosistemas y Sustentabilidad, Universidad Nacional Autónoma de México, Ciudad de Mexico, Mexico
[26]Museo de Zoología "Alfonso L. Herrera", Departamento Biología Evolutiva, Facultad de Ciencias, Universidad Nacional Autónoma de México Ciudad de México, Ciudad de Mexico, Mexico
[27]Australian Antarctic Division, Department of Climate Change, Energy, the Environment and Water, Kingston, Tasmania, Australia
[28]Department of Ecosystem Science and Sustainability, Colorado State University, Fort Collins, Colorado, USA
[29]Departamento de Biología Evolutiva, Instituto de Ecología, A.C. (INECOL), Xalapa, Veracruz, Mexico
[30]Departamento de Botánica, Instituto de Biología, Universidad Nacional Autónoma de México, Ciudad de Mexico, Mexico
[31]School of Biosciences, Cardiff University, Cardiff, UK
[32]Facultad de Ciencias, Universidad Nacional Autónoma de México, Ciudad de México, Mexico
[33]Division of Ecology and Evolution, Research School of Biology, The Australian National University, Canberra, Australian Capital Territory, Australia
[34]Sydney School of Veterinary Science, B01, University of Sydney, Sydney, New South Wales, Australia
[35]Australian Museum Research Institute, The Australian Museum, Sydney, New South Wales, Australia
[36]Nordic Chapter of the Society for Conservation Biology, Uppsala, Sweden
[37]Department of Public and Ecosystem Health, Cornell University, Ithaca, New York, USA
[38]Department of Fish, Wildlife, and Conservation Biology, Colorado State University, Fort Collins, Colorado, USA
[39]Laboratorio de Genética de la Conservación, Jardín Botánico, Instituto de Biología, Universidad Nacional Autónoma de México, Ciudad de México, Mexico
[40]Swedish Species Information Center, Swedish University of Agricultural Science, Uppsala, Sweden
[41]Research Centre for Ecosystem Resilience, Botanic Gardens of Sydney, Sydney, New South Wales, Australia
[42]Conservation Genetics Laboratory, University of Liège, Liège, Belgium
[43]Zoology & Entomology Department, University of Pretoria, Pretoria, South Africa
[44]UWA Oceans Institute, University of Western Australia, Crawley, Western Australia, Australia
[45]School of Biological Sciences, University of Western Australia, Crawley, Western Australia, Australia
[46]Departamento de Ciencias de la Sustentabilidad, El Colegio De La Frontera Sur, Tapachula, Chiapas, Mexico

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PEER REVIEW

The peer review history for this article is available at https://www.webofscience.com/api/gateway/wos/peer-review/10.1111/ele.14461.

DATA AVAILABILITY STATEMENT

Kobo forms and scripts used to collect and analyse data are available on the GitHub repository https://github.com/AliciaMstt/GeneticIndicators (Zenodo: https://zenodo.org/doi/10.5281/zenodo.10620306).The data that support the findings of this study are available in DRYAD (https://doi.org/10.5061/dryad.bk3j9kdkm). Some data that could lead to the geographic identification of endangered species have been obscured.

ORCID

Alicia Mastretta-Yanes https://orcid.org/0000-0003-2951-6353
Jessica M. da Silva https://orcid.org/0000-0001-8385-1166
Catherine E. Grueber https://orcid.org/0000-0002-8179-1822
Luis Castillo-Reina https://orcid.org/0009-0008-3477-5097
Brenna R. Forester https://orcid.org/0000-0002-1608-1904
W. Chris Funk https://orcid.org/0000-0002-6466-3618
Myriam Heuertz https://orcid.org/0000-0002-6322-3645
Fumiko Ishihama https://orcid.org/0000-0001-8515-5914
Rebecca Jordan https://orcid.org/0000-0002-4048-6792
Joachim Mergeay https://orcid.org/0000-0002-6504-0551
Ivan Paz-Vinas https://orcid.org/0000-0002-0043-9289
Victor Julio Rincon-Parra https://orcid.org/0000-0001-8910-0072
Maria Alejandra Rodriguez-Morales https://orcid.org/0000-0002-4075-294X
Libertad Arredondo-Amezcua https://orcid.org/0009-0004-2426-4737
Gaëlle Brahy https://orcid.org/0009-0002-9854-6054
Lily Durkee https://orcid.org/0000-0002-7328-0256
Margaret E. Hunter https://orcid.org/0000-0002-4760-9302
Austin Koontz https://orcid.org/0000-0002-6103-5894
Iris Lang https://orcid.org/0000-0001-5329-5023
María Camila Latorre-Cárdenas https://orcid.org/0000-0002-9555-2769
Tanya Latty https://orcid.org/0000-0002-7469-8590
Alexander Llanes-Quevedo https://orcid.org/0000-0002-1790-4204
Anna J. MacDonald https://orcid.org/0000-0003-2972-200X
Juan Francisco Ornelas https://orcid.org/0000-0002-1124-1163
Santiago Ramírez-Barahona https://orcid.org/0000-0003-3999-6952
Erica Robertson https://orcid.org/0000-0002-0869-6300

Isa-Rita M. Russo https://orcid.org/0000-0001-9504-3633
Robyn E. Shaw https://orcid.org/0000-0002-7899-1743
Glenn M. Shea https://orcid.org/0000-0002-0052-4205
Per Sjögren-Gulve https://orcid.org/0000-0001-6584-128X
Emma Suzuki Spence https://orcid.org/0000-0003-1595-0167
Sofía Suárez https://orcid.org/0009-0005-3516-5184
Henrik Thurfjell https://orcid.org/0000-0001-9261-7654
Sheela Turbek https://orcid.org/0000-0003-1080-9375
Marlien van der Merwe https://orcid.org/0000-0003-1307-5143
Fleur Visser https://orcid.org/0000-0001-5135-2530
Ana Wegier https://orcid.org/0000-0003-4400-8111
Georgina Wood https://orcid.org/0000-0003-3855-0932
Eugenia Zarza https://orcid.org/0000-0001-7702-8579
Linda Laikre https://orcid.org/0000-0001-9286-3361
Sean Hoban https://orcid.org/0000-0002-0348-8449

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