Monitoring status and trends in genetic diversity for the Convention on Biological Diversity: an ongoing assessment of genetic indicators in nine countries

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ABSTRACT

Recent scientific evidence shows that genetic diversity must be maintained, managed, and monitored to maintain biodiversity and nature's contributions to people. Three genetic diversity indicators, two of which do not require DNA-based studies, were previously proposed for reporting to the Convention on Biological Diversity and other conservation and policy initiatives. These indicators allow an approximation of the status and trends of genetic diversity to inform policy, using existing demographic and geographic information.

Application of these indicators has been initiated and here we describe ongoing efforts in calculating these indicators with examples. We specifically describe a project underway to apply these indicators in nine countries, provide example calculations, address concerns of policy makers and other challenges, and describe a roadmap for further development and deployment with the incorporation of feedback from the broader community. We also present guidance documents and data collection tools for calculating indicators. We demonstrate that Parties can successfully and cost-effectively report these genetic diversity indicators with existing biodiversity observation data, and in doing so, better conserve the Earth's biodiversity.

MAIN TEXT

Reporting genetic diversity change is vital and is feasible

Genetic diversity is variation at the DNA level, including differences among individuals and populations of each species. Because it contributes to the traits and survival of organisms, this intraspecific diversity is vital for helping species adapt to changing environments (including climate, pests, habitat changes, and disease). It also contributes to the stability and resilience of ecosystems including after extreme climate events[1,2], and helps ensure success of ecological restoration[3], including in forest, grassland, streams, coral, and seagrass ecosystems[4–6]. *Unfortunately, genetic diversity has already declined substantially due to habitat loss, habitat fragmentation, over-harvest, and other human activities*[7,8].

Although it is recognized as one of three basic units of biodiversity, genetic diversity has been neglected in public policy and management[9–11]. Vague or imprecise wording in policy language, insufficient indicators to track progress, expensive technology, and metrics that are inaccessible to non-geneticists has resulted in weak conservation action and minimal reporting on genetic diversity status and trends[12–15]. For instance, a recent analysis of 57 national biodiversity reports revealed that countries primarily use indicators which are not well connected to genetic erosion, primarily focus on breeds of economically important species or crop wild relatives, neglect genetic monitoring, and focus on ex situ rather than in situ gene conservation[16].

It is a critical time to conserve genetic diversity, particularly through the upcoming United Nations Convention on Biological Diversity (CBD) post 2020 Global Biodiversity Framework (GBF), whose final negotiations occur in December 2022. Reporting biodiversity indicators is an obligation under the Convention; indicators at national scale and disaggregated by taxonomic groups, habitats or other categories also helps countries understand and mitigate biodiversity loss. In CBD negotiations over the past two years, disagreement and confusion over genetic diversity concepts has been apparent. Truly safeguarding genetic diversity will require clear, precise, science-informed wording in CBD Goals and Targets, and affordable, accessible, and relevant genetic diversity indicators[17,18].

In this paper we summarize recent significant advancements in indicators that assess status and trends in genetic diversity, and their application at national scales. Specifically, we

- reiterate the need and purpose of three indicators, two of which do not require DNA-based analysis
- summarize and address concerns from policy makers
- describe ongoing deployment of indicators in 9 countries on 6 continents
- specify indicator calculation, including with examples
- address other challenges and describe a roadmap for uptake and use of genetic diversity indicators, including current and future support resources.

We are confident all nations can successfully report these genetic diversity indicators, and in doing so, better conserve the world's biodiversity.

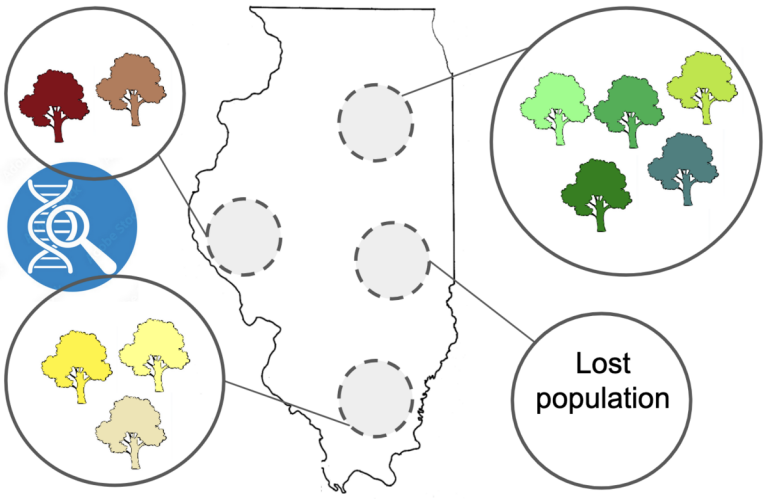
Need and purpose for each indicator

In 2020, three genetic diversity indicators were proposed and discussed (Figure 1)[18–21]. The indicators have several motivations: to assess or approximate genetic diversity status without requiring new DNA data; to be affordable and feasible with existing data and with limited time investment; to use simple calculations; to allow for easy translation into policy and management of species; and to be applicable and relevant in all

countries, taxonomic groups, and ecosystems. It is also desirable to describe genetic erosion with concepts that are intuitive or accessible to non-geneticists (e.g. genetic losses due to small populations and loss of populations). Assessing genetic status without DNA-based genetic data is vital since relatively few species have DNA-based studies, especially in biodiversity hotspots.

The proposed indicators relate to central conservation genetics concepts:

(1) conserving genetic diversity within large populations for rapid response to changing environmental conditions, (2) conserving genetic diversity among populations to provide diverse 'options' for the future adaptation of the species (e.g option value,[22]), (3) assessing genetic data directly to guide conservation actions and sustainable use.



Indicator 1. Populations large enough to maintain genetic diversity	Indicator 2. Populations still exist (not lost)	Indicator 3. Populations monitored with DNA data
0.33	0.75	1

Figure 1. Example of the three genetic diversity indicators, for four hypothetical populations in Illinois, USA. One tree = 1,000 plants (five trees = 5,000 plants). Colors illustrate genetic variation within and among populations. In 2020, 2 of 3 extant populations are Nc<5,000 (Ne<500 considering an effective to census size ratio of Ne/Nc = 0.1) and thus too small to maintain genetic diversity (indicator 1). Three of four historic populations are maintained (indicator 2). DNA-based methods have been used to monitor genetic diversity in two populations (indicator 3 - a value of 1 means that one or more populations of the species is monitored with DNA-based methods).

Indicator 1 is "the proportion of populations within species with a genetically effective size, Ne, greater than 500." Effective population size is a concept that quantifies the rate of genetic erosion within a population (genetic erosion is the loss of genetic diversity, increase of inbreeding, and reduction of population ability to adapt). Past CBD indicators do not reflect genetic erosion within populations (though they did reflect genetic erosion in agricultural breeds)[10]. Large Ne can help avoid population's and species' extinction. Specifically an effective size of 500 is well regarded as a *minimum* threshold for populations to maintain genetic diversity[23,24](though see[23]). Using a ratio of effective to census size, Ne/Nc (0.1 to 0.3 for most

species[20] translates to comparing census population size to a threshold of about 5000. *In this way, demographic information - census size - is translated to information on genetic status.* For example, *Cupressus abramsiana* , a USA endemic gymnosperm, has five populations, and two exceed a census of 5000. Indicator 1 for this species is 0.4.

Indicator 2 is "the proportion of populations within species which are maintained." This indicator is needed because past CBD indicators do not reflect loss of genetic distinctiveness of each population. *Each population may hold traits and genetic adaptations supporting species' survival*[25,26]. This concept is already recognized in distinct agricultural breeds, which are analogous to populations, each with unique traits or characters. Genetic, geographic and ecological variation allows future options for adaptation. Losses of species' populations and geographic range change are often quantifiable[27,28]. For example, *Capensibufo rosei* , endemic to the Cape Peninsula of South Africa, is known from five historical populations; only one exists today, along with a newly discovered population. Indicator 2 for this species is 0.33 (2 of 6 populations maintained)[29].

Indicator 3, is "the number of species and populations being genetically monitored within a country." Although not common in all countries at present, DNA-based genetic monitoring, when available, can guide conservation actions and policy[30,31]. For example, a DNA-based monitoring program of mountain pygmy possums *(Burramys parvus*) revealed inbreeding, very low Ne of ˜10 and the loss of over 80% of the population's genetic diversity. Based on this knowledge, a genetic rescue program introduced six genetically healthy males from the closest known population[32]. This indicator includes any monitoring program using DNA data to help managers assess genetic status and choose appropriate actions, including studies of genetic connectivity, hybridization, adaptation, etc.[33].

These indicators more directly assess genetic erosion than the Living Planet Index or Red List Index. Neither of these focuses on the threshold of Ne 500 within populations; below Ne500, genetic erosion is exponentially faster. Also, the RLI typically assesses the entire species, which may be safe from extinction but still suffer losses of genetic diversity. Meanwhile the LPI does not consider losses before the 1970s, though many populations were lost or declined prior to this time.

Addressing concerns from policy makers

The Coalition for Conservation Genetics[34] presented these indicators to policy and management personnel globally through 10+ webinars (Supplemental Material). Five concerns about indicator uptake were common (Table 1): (1) necessity of DNA data, (2) feasibility of obtaining sufficient amounts of data, (3) realistic limitations on time, (4) limitations on skills or knowledge, and (5) concerns over data sharing, particularly Digital Sequence Information (DSI)[35]. We address these concerns in Table 1, highlighting that: *no new DNA collection is needed, most countries have sufficient data for reporting, one person can complete analysis in a fraction of one year without specialized expertise, and no DSI is shared in indicator reporting.*

Table 1. Resolving concerns regarding genetic diversity indicators

Concerns	**Solutions or clarifications**
Data needed: Is genetic data (DNA-based analysis) needed?	**Indicators 1 and 2 *do not need* genetic data (DNA sequencing), genetic expertise or laboratory facilities to generate data**. Genetic data can be used to estimate Ne, but *in most cases existing census size data* (counts of individuals using camera traps, surveys, etc.) along with an Ne/Nc ratio can be applied to obtain a valid proxy of Ne, and similar records for measuring 'populations maintained'.

Concerns	Solutions or clarifications
Achievable: Is there enough census or geographic data?	Yes, many countries monitor priority species, maintain biodiversity databases or citizen science tools, and contribute Red List assessments which may have geographic and census data. Data collection can include local knowledge or expert consultation, or categorical or imperfect data. **Genetic diversity indicator reporting is not for all species- it would be for a relatively small, representative sample** of species per country (we recommend >?100) [18,20,21]. Representative = from a diversity of ecosystems, taxonomic groups, rarity, and lifespan
Realistic: Is indicator calculation too time-consuming?	Collection of census sizes and number of populations is similar in scope to compiling other information for CBD National Reports. From pilot tests, 3-5 persons could complete analyses on a total of 100 species in 2-4 weeks, or 1 person could do it in 2-4 months per country.
Realistic: Does indicator calculation require special skills or tools?	No. Basic biology training should be sufficient for gathering data. Our guidance documents explain how to choose representative species, how to find and record data, and how to resolve challenges (Supplemental Material see also [36]). A data collection device allows for standard recording and storage of data, and analysis (Supplemental Material).
Data sharing Does reporting genetic indicators require reporting or sharing genetic data or Digital Sequence Information (DSI)?	**Reporting on genetic indicators *does not* involve sharing or reporting any DSI.** Indicators 1 and 2 will typically be based on demographic and geographic data (e.g. census sizes, population distributions), which do not use DNA data (DSI) at all. If DNA data were used to calculate Ne or define population boundaries, only the count of populations and estimates of effective size would be recorded- *no DSI is reported or shared*. Even raw census data and population data could be retained by the country, reporting only the proportions for the indicators. Although indicator 3 does assess genetic studies in the country, it is only a count of studies- it does not assess DNA data or share DSI. Indicating DNA data availability is at the discretion of each country[35].

Towards indicator deployment

While the indicators were being discussed by the CBD (CBD/WG2020/3/5, CBD/ID/OM/2022/1/2), a trial was initiated by the Swedish Environmental Protection Agency. Examining Swedish national Red List assessments, this trial (1) assessed data quality/ availability for 22,000 species, and (2) calculated indicator values for 79 species[37]. Approximately 33% of species had census estimates and 20% of species had historic

and modern population information- data which are the basis for indicators 1 and 2. The trial also found that 70% of herptile and 49% of mammal populations likely had Ne > 500 while 32% of herptiles and 84% of mammals are maintaining their geographic range. This study concluded that

Data for the indicators are likely available for thousands of species

A large proportion of species are already experiencing genetic erosion within and among populations.

A new project described here is testing the indicators in nine countries across six continents, in order to:

(1) create and refine a standard workflow, definitions, methodology, and data collection device, and (2) for each country, evaluate >100 species to: (2a) determine how many species have necessary data, (2b) extract data when possible and perform indicator calculation, and (2c) identify challenges encountered so that guidance and calculations can be improved for use by more countries.

The project will also highlight taxonomic groups and regions where data are insufficient. *We describe the approach to demonstrate that data are available to calculate the indicators in countries around the world, and to show that data collation is practical, achievable and adaptable.*

Gathering data on populations of species can be challenging because there is no global, standard database of population census size or changes in population distribution. The Living Planet Index for example does not measure full population census sizes. However, population level data of many species is available in different reports, atlases, and databases, and with local and expert knowledge holders. Publicly available data, e.g. from the citizen science, databases, and the Red list can also be used.

We identified three approaches to gathering data, which suit different countries' needs and can be used in combination (Figure 2).

- "Manual data extraction" involves reviewing national or subnational documents (management or recovery plans, status assessments, environmental impact reports), scientific literature, country flora or natural history descriptions, IUCN Red List assessment or NatureServe assessment text, etc.
- "Expert consultation" involves facilitated discussion and knowledge sharing among local experts and traditional knowledge holders with firsthand (but perhaps unpublished) information- similar to Red List assessment workshops which elicit quantitative information on species.
- "Automated data extraction" is possible from existing databases on national surveys, where species' occurrences or population sizes are regularly assessed and stored, often in a gridded spatial database (common in some fish, plants, birds, and mammals), as with stock assessments, some National Red List databases, or forest inventories. Population presence can also be obtained from citizen science databases (e.g. iNaturalist). Automated analysis can compare census sizes for each population to the Ne (or Nc) threshold, and compare historic data, atlases, or range maps to current population presence or projections of habitat change to assess "populations maintained".

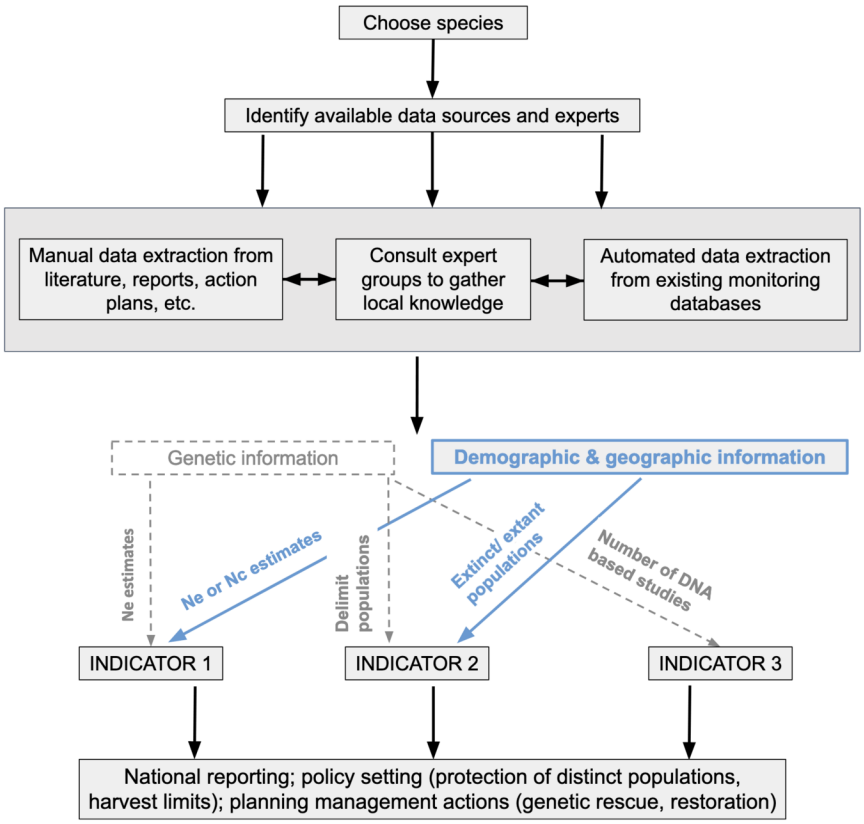


Figure 2: Process of assessing indicators for a set of species, using data from different sources. Note that genetic information is optional for indicators 1 and 2 as shown in grey dashed lines. For indicator 1, census sizes (Nc) can be converted to effective sizes (Ne) by applying a species-specific effective to census size ratio, and/or a ratio of Ne/Nc = 0.1

Examples from different countries illustrate the diverse options available. Recovery plans for dozens to thousands of threatened species are mandated by national legislation (Australia- the Environment Protection and Biodiversity Conservation Act, https://www.dcceew.gov.au/environment/epbc; South Africa- Biodiversity Management Plans, https://www.dffe.gov.za/content/management_plans/biodiversity; USA- the Endangered Species Act, https://www.fws.gov/law/endangered-species-act). These documents typically detail species biology and demographic status. In Japan, many threatened vascular plants have been surveyed for census size for over two decades by the Japanese Society for Plant Taxonomy, while for common trees, statistical estimates for population size[38]were estimated from vegetation survey data. In Mexico, taxonomic experts who recently helped validate distribution models for crop wild relatives will be consulted for indicator values[39] . In France, Belgium and Sweden, much biodiversity data from experts and diverse sources are collected in easy to access web-based portals (France- INPN, Belgium - www.observations.be, Sweden- Swedish Species Information Centre, Artdatabanken). In Colombia, the Biodiversity Information System (SIB) repository compiles species surveys from throughout the country (https://biodiversidad.co/), which is mandated by many public and private organizations. These data are reviewed by national experts for validation and used to create freely available species distribution models (http://biomodelos.humboldt.org.co/), and for conservation prioritization.

Table 2: Countries participating in testing of genetic diversity indicators, showcasing variation in overall approach

Country	Number of people	Taxonomic groups*	Approach	Sources**
South Africa	7 or 8	B, F, H, I, M, Ma, P	Manual + Expert	AH, EGI, NRL, GRL, SMP
USA	10	B, F, H, I, M, P	Manual	AH, GRL, SMP
Japan	4 or 5	P	Manual + Auto	AH, FG, ND, NRL
Mexico	10	B, CW, F, H, M, P	Expert + Auto	AH, EGI, ND, NRL
Australia	8	B, F, H, I, M, P	Manual	AH, GRL, ND, SMP
Sweden	2+	B, F, H, I, M, P	Manual + Expert	AH, ND, NRL, SMP
Belgium	2+	B, I, M, P	Manual + Expert	AH, NRL, SMP
Colombia	3+	B, H, M, P	Manual + Expert	AH, EGI, GRL, ND, NRL
France	4	B, F, H, I, M, P	Manual	AH, ND, SMP, NRL

*B=Birds, CW=crop wild relatives, F=freshwater fish, H=herptiles, I=invertebrates, M=mammals, Ma=marine, P=plants

**AH=ad hoc/ other (websites, email experts, scientific literature), EGI- Expert group input, FG=field guide, GRL=Global Red List, ND=National dataset or database of species information and/or occurrences, NRL=National Red List, SMP=species specific management, action or recovery plan

Calculating indicator values at national levels

Genetic indicator calculation is straightforward. First, indicators are calculated for each species. When data are available as a range (e.g. Nc is 10 000 to 20 000), the mean is used. Qualitative data such as "a few hundred" or "at least 5000" are sufficient for comparison to the Nc 5000 threshold. In the case of differing estimates from multiple sources, either the most recent source is used, or a mean of values based on the different sources is taken (see Table 3, rows 2 and 3).

While indicator 3 is a simple sum of all species for which one or more populations are being monitored using DNA methods, the country indicator value for indicators 1 and 2 is the mean of values across species (a median could be used for skewed distributions). If taxonomic groups are not represented evenly, the indicator value is the mean of each taxonomic group's means, which down-weights overly represented taxonomic groups, e.g. mammals. Additionally, each species can be weighted by the proportion of its geographic range in the country, from 0 to 1, to reflect national responsibility, with full weight for endemic species[40]. Transboundary populations can be weighted similarly.

Equation 1: simple indicator value (IV) mean across species (s)

$$\frac{\sum_{s=1}^{s=100} IV_s}{s}$$

Equation 2: indicator value (IV) weighted by proportion of geographic range in country (W)

$$\frac{\sum_{s=1}^{s=100} IV_s W_s}{\sum_{s=1}^{s=100} W_s}$$

Equation 3: indicator value (IV) giving equal weight to birds (b, 20 species), plants (p, 30 species) and mammals (m, 50 species)

$$\frac{\sum_{s=1}^{s=100} IV_s W_s}{\sum_{s=1}^{s=100} W_s}$$

Table 3: Example indicator values for select species

Species	**Taxonomic group**	**Country**	**Indicator 1 (Ne)**	**Indicator 2 (populations)**	**Indicat**
Pelobates fuscus	Amphibian	Belgium	0	0.33	1
Rana arvalis	Amphibian	Belgium	0.18	0.84	1
Angelica heterocarpa	Angiosperm	France	0.5	1	1
Zingel asper	Fish	France	0.2	1	1
Tetrao urogallus	Bird	France	0.33**	0.6	0
Taraxacum yuparense	Angiosperm	Japan	0	0.5	0
Carex cinerascens	Angiosperm	Japan	0.5	0.8	0
Capensibufo rosei	Amphibian	South Africa	0.5	0.33	1
Clinus spatulatus	Fish	South Africa	N/A***	1	0
Syncerus caffer caffer	Mammal	South Africa	0.3	1	1
Alces alces	Mammal	Sweden	1	1	1
Silurus glanis	Fish	Sweden	0	0.5	1
Cupressus abramsiana	Gymnosperm	USA	0.4	1	0
Rana muscosa	Amphibian	USA	0	0.76**	1
Charadrius melodus	Bird	USA	0.39**	1	1
Erigeron maguirei	Angiosperm	USA	0.5	1	0

*Indicator 3 is binary for each species, 0 or 1 (1=one or more populations of the species is monitored with DNA-based method; 0=no DNA-based monitoring for the species)

**Different reports suggest different values or different interpretations of population boundaries; these values are means of the different interpretations

*** Indicator 1 could not be calculated because no Nc or Ne data are available.

Overcoming challenges

In trials of the methodology outlined above we have resolved several challenges: biases, uncertainty in data, and difficulty in defining populations.

Biases: Ideally, the 100+ species assessed by a country reflect diverse ecosystems, taxonomic groups, rarity categories, and life history (e.g. lifespan). In reality, biases exist due to a country's habitat types, biogeography, latitude, and investment priorities in biodiversity monitoring and thus data availability. Similar biases are well known in other indicators such as the Red List Index and Living Planet Index[41,42]. Weighting by taxonomy or ecosystem can help address bias (previous section). Additionally, biases should be noted by displaying counts per category in a matrix as is common in Red List summary tables.

Uncertainty or qualitative information : Sometimes, census sizes are recorded as a range of values rather than point values; using vague wording such as "several hundred" or "very small"; or census at the species but not population level. Our draft guidance (Supplemental Materials) provides the assessor with advice on translating these into quantitative information, and recording degree of uncertainty. In addition, if desired to show variation, the indicator can be calculated and reported, with and without such species.

In addition, for indicator 1, if census size is not available, it may be possible to use known occupied area multiplied by mean density (number of individuals supported per unit area) = estimated number of individuals. This allows evaluating whether an area is capable of housing a population Nc larger than the threshold; if the area is smaller, the Ne will likely be smaller than 500[43]. For indicator 2, if information on the number of historic populations is not known, the assessor may record overall decline in area (which is more common), which will ultimately result in lost populations and genetic diversity[7].

Definition of population boundaries (required for indicators 1 and 2) . Ecologists and geneticists have worked for decades to understand population distinctions[44–46]. For the indicators, available knowledge can be used to assess genetic distinction, typically less than one migrant per generation from other populations[47], such as: relying on population designations from the report or experts consulted, which reflect knowledge of the species biology, history and dispersal; using discrete patches such as forest or lakes; using ecoregions, geographic (and migration) barriers such as mountains/ valleys, or hydrological zones, which may promote local adaptation; synthesis of phylogeographic studies ("i.e. proxies of genetic differentiation"[48]); or use of grid cells based on species' dispersal.

Roadmap

We have presented the purpose and straightforward methodology of each indicator, addressed concerns of policy makers, showed that data are available and usable, and described ongoing work in nine countries. We have demonstrated that genetic diversity indicators are ready for use in biodiversity conservation and reporting, with existing data.

Increased uptake of these indicators by Parties to the CBD and other users (environmental agencies, wildlife managers, national legislation, etc.) will require further successful demonstrations, published step-by-step workflows, and training workshops, ideally in multiple languages[34]. We have created an online data collection form using Kobotoolbox (*www.kobotoolbox.org/*) and a guidance document (Supplemental material) for anyone to use. By sharing these on github we are promoting interactive engagement with stakeholders who can offer suggestions or ask questions.

Future development of online resources can enhance data storage, managing, and sharing. An online portal

could accept and store submitted data (Ne or Nc values, population designations, references) over multiple cycles of CBD reporting, which would make completion of the indicators easier with each cycle and increase transparency. Other advantageous cyber infrastructure would be connections to resources like species distribution models or Map of Life[28,49], which could help with defining populations, calculating Ne based on area and density, loss of distinct populations etc. Finally, as noted by others[37,50,51] *it would be advantageous for all Red List assessment workshops to include gathering of raw data and information needed for calculation of the indicators, including population boundaries, during the workshop.*Red List assessors scour literature and consult experts for demographic and geographic information, but currently this is not recorded in any systematic way in the Red List database.

Lastly, we note that the indicators presented here are complementary to other indicators which are very useful in certain situations. The "genetic scorecard" assesses genetic diversity threats such as hybridization and poor recruitment[52,53], while another indicator assesses the extent to which geographic ranges are protected in situ or ex situ[54,55]. Meanwhile, direct assessments of genomic health (e.g. genetic load) based on DNA data are available for some species[56]. In the future, genetic indicators could be synthesized together for comprehensive genetic assessment[18,57].

Closing remarks

The indicators support and enable the following post-2020 CBD Goal A and Target 4 wording, presented previously by the conservation genetic community,[10,18,21]

"....all genetically distinct populations are maintained, [at least 97% of] genetic diversity within populations is maintained, and large effective population sizes and appropriate genetic exchange are ensured."

"Ensure active management actions to enable the recovery and conservation of species and of genetically depleted populations, and the long-term maintenance and protection of genetic diversity within and among all populations."

We close by reiterating that scientific evidence shows that genetic diversity is a basic pillar of all biodiversity that must be*maintained (not lost), protected (via legislation and policy), managed (through interventions such as restoring gene flow and genetic rescue), and monitored (with DNA-based and non-DNA-based metrics like the indicators)*[6,17].These four elements are needed to enable all species to adapt to environmental change, ensure resilient ecosystems, and benefit humanity.

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Supplemental Material: Single Word document containing (1) list of webinars held for policy makers, and links to relevant CBD documents (2) complete guidance document for collecting data underlying the indicators, (3) Several examples of the species selection matrix for three countries, (4) Screenshots of the full data collection form in Kobo, (5) detailed guide to completing Kobo collection form

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