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Biodiversity biobanks: a landscape analysis

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ABSTRACT

Biobanks are curated collections of biological samples that are preserved at the molecular level, usually frozen, along with associated data, and managed to high scientific standards. We conducted a 'landscape analysis'—based both on a community survey and a literature review—to determine commonalities, information gaps, and challenges in the various workflows of biodiversity and environmental biobanks. The survey was completed by 55 institutions from more than 20 countries. Its results were compared to other collection-based surveys and complemented by literature research in the areas of general biobank management, staffing, sample handling, storage, (cryo-/)preservation, policies, databases, and networking. We illustrate strengths and weaknesses of biodiversity and environmental biobanks and provide some basic recommendations for improving biobank procedures. In general, we found that increased efforts are needed to standardise biobank workflows or individual workflow components. While general, organism-independent biobanking guidelines already exist, more detailed guidance documents to date mostly address only human biobanking, or a narrow range of biodiversity. We hope to start closing that gap by providing an overview of current protocols and practices in biodiversity and environmental biobanking in form of a handbook, to which the present work is directly related. The handbook is available open-access under https://doi.org/10.3897/ab.e101876.

Keywords

Biobanking, cryobank, biodiversity biobank, environmental specimen bank, collection, preservation, cryopreservation, cold storage

INTRODUCTION

Biodiversity is rapidly declining due to a range of factors including habitat loss, overexploitation, climate change, introduced species, and pollution. Biobanking is an essential complement to *in-situ* and other *ex-situ* conservation efforts, being amongst the most effective approaches to preserve species and their genetic diversity. Biobanking is also a complementary tool to many applications and branches of research, among others spatial and temporal biodiversity / environmental monitoring (Hildebrandt et al. 2021, Broders et al. 2022), reproductive strategies (Holt & Comizzoli 2021), and the recovery of lost genetic lineages (Singina et al. 2014, Borges & Pereira, 2019, Novak 2018).

Biodiversity biobanks and environmental biobanks (BEBs) are scientific collections comprising various types of (usually cold-)preserved samples from biological organisms (e.g., tissue, DNA, blood, somatic cells, germplasm, embryos, small organisms) or the environment (e.g., soil, water or filtrate,

air) with associated data that are managed to high scientific standards and follow standardised workflows (Comizzoli & Wildt 2017, NatSCA, https://www.isber.org/page/BPR, Campbell et al. 2018). The periodically updated ISBER (International Society for Biological and Environmental Repositories) document contains important information and recommendations on facilities and equipment, safety, quality control, biobank management, and liquid nitrogen handling. However, the ISBER Best Practices do not contain specialised biodiversity information, e.g., regarding culture collections, seed banking or environmental samples, and some of its content focusing exclusively on human biobanks is not relevant for the BEB community (Zimkus & Ford 2014a). As a consequence, several BEBs produce their own protocols, leading to different practices that are often not standardised. Standard operating procedures (SOPs) for collecting, storing, preserving, and using samples are crucial, as they ensure the correct functioning of the biobank, safeguarding DNA integrity or cell viability for extended timespans (Mullhall 2019). In addition, SOPs promote data comparability between different methods and aspects of the biobank workflow.

Biobanks vary widely in scope and are housed in different settings such as botanical gardens, zoos, natural history museums (Zika et al. 2011) and other agencies. BEBs focus on living collections (McCluskey et al. 2017a), DNA, RNA, and/or fixed tissues, all of which represent valuable genetic resources, usually for a wide range of species. Culture collections of microorganisms (protists, fungi, bacteria), livestock biobanks, and agricultural seed banks have been well-established for decades and procedures are consistent, as they must guarantee viability and availability of resources for use (Engels & Fassil 2009). Cold-preserved animal collections have formerly focused more exclusively on laboratory models (e.g., specific invertebrates, non-human primates, mice) for human disease research (McCluskey 2017). Recently, wildlife biobanks have been established, usually with well-defined collecting practices, but often lacking standardisation of sample processing and quality controls (Comizzoli & Wildt 2017). In addition, wild seedbanks have recently been established, although to date they still encompass a comparatively smaller spectrum of species than their agricultural counterparts (Paton et al. 2020). BEBs are as relevant as human biobanks, as they not only store valuable biodiversity and environmental samples that are difficult to access, but also because they are of use for health and sustainability research (MacKenzie-Dodds et al. 2013). Biodiversity samples are fundamental for breeding and population restoration programs along with prioritisation of species which are at risk of extinction, veterinary medicine, monitoring, and biotechnology, among others (Comizzoli & Wildt 2017, Breithoff & Harrison 2018).

Previous consultations and surveys conducted within the natural history collection community (Zimkus & Ford 2014b), plant genetic resources (Ashmore 1997, Andersson 2004, Andersson et al. 2006), plant-pathogenic taxa (e.g., viruses, microbes, fungi, arthropods, nematodes) (Korkaric & Beed 2010), livestock (Leroy et al. 2019, Passemard et al. 2020), aquatic model organisms (Hagedorn et al. 2019), and environmental specimen biobanks (Küster et al. 2014, Helbing & Hobbs 2019) have revealed the key practices carried out in different types of biobanks, as well as the challenges they are facing along the biobank workflow. Overall, the results showed that curation practices varied greatly. For instance, collection duplications for livestock germplasm are implemented, but this is not a common practice for livestock genomic collections (Andersson 2004, Passemard et al. 2020). The major challenges that all types of biobanks share lie in the following areas: general standardisation of practices, regulatory compliance, financial support, infrastructure, quality control and availability of trained staff (Pérez-Espona & CryoArks Consortium 2021). These issues have a higher impact in BEBs located in megadiverse developing countries (Angeles & Catap 2022).

This study was undertaken in the context of the SYNTHESYS+ project, carried out by a consortium of natural history collections (NHC) and botanical gardens, that aims to improve and expand access to NHC across Europe (Smith et al. 2019). SYNTHESYS' networked activity three (NA3) aims to develop

and disseminate standardised best practices for biobanking activities in the age of genomics. Led by the Global Genome Biodiversity Network (GGBN), work package NA3.1 engaged biodiversity repositories such as those at zoos and aquaria, natural history museums, culture collections, agriculture and livestock biobanks, palaeontological/archaeological collections, as well as environmental specimen collections, to obtain a broad overview of standards for sample collection, preservation, and storage, and for data management. Note that biorepositories can include living stock collections, DNA banks and specimen collections. The objective of the present work was to identify common practices, gaps, and challenges in the biobank processes, from field collection to data standards and sample use. We extracted a consensus based on information gathered from the available literature, expert-led communications and from questionnaire responses, covering mainly technical aspects and procedures. The results of the survey along with an extensive literature review have contributed to the development of a new handbook to biobanking practices that applies to a variety of organisms and palaeontological remains. Through SYNTHESYS, this handbook is available open-access under https://doi.org/10.3897/ab.e101876.

MATERIALS AND METHODS

We performed an online search taking into consideration national biobanks, and livestock germplasm collections as well as biobanks at zoos, aquaria, natural history museums, botanical gardens, and universities. Introductory emails were sent to request participation in the online questionnaire prepared in Google Forms (Annex 1). In addition, the survey was circulated among SYNTHESYS+ partners and GGBN members. The survey was open from November 17, 2019, to January 31, 2020. A list of participating institutions is given in Annex 2. We verified that multiple respondents belonging to the same institutions represented different collections, to avoid duplicate answers.

The survey included 45 questions grouped according to various biobank procedures (specimen collection, preservation methods, sample retrieval, material request and shipment, safety and security, data standards and policies) (Annex 1). No question was defined as mandatory to complete the survey. Most questions involved a list of items for which respondents were asked to indicate all applicable options. Some of the questions included multiple-choice and open answers. Therefore, the overall total for these questions did not add up to 100%. Survey results were exported from Google forms to Microsoft Excel for data visualisation. The responses were evaluated by applying simple summary statistics to determine the proportions of biobanks.

We also conducted a literature review on Google Scholar and ResearchGate between February 2021 and November 2022, using a combination of the following search terms: “biobank”, “collection”, “processing”, “storage”, “biodiversity”, “challenges”, “gaps”, “tissue”, and “DNA”. We considered both peer-reviewed and grey literature (e.g., theses, book chapters, institutional reports, poster presentations, conference and workshop proceedings) in English, Spanish, and Portuguese. We refined our search by excluding papers with the words “human”, “clinical”, “epidemiology”, and “cancer”. Papers were also excluded if they focused on strain descriptions, patents, biobank/collection history, catalogues, biobank counts worldwide, biobank services, monitoring, and genomic studies. Abstracts were manually checked to further confirm that articles dealt with biodiversity biobanking. The individual papers were read selectively with the aim of identifying any challenges in the biobank workflow that had not become apparent in the survey.

Furthermore, we personally contacted curators, biobank managers, and experts (Annex 3) to gain a deeper understanding of the work being done at their institutions, including specific methods such as cryopreservation (storage of viable material as, e.g., living cells) or procedures connected with sampling and storage during fieldwork and at biobank facilities.

RESULTS

We collected 55 responses from 38 institutions located in 21 different countries (Annex 3). Three institutes were represented by more than one response/collection: the Natural History Museum Vienna (7), the Royal Botanic Garden Edinburgh (3), and the Finnish Museum of Natural History (2).

Institution information. Most institutions belong to natural history collections (45%), followed by public research institutes (15%), universities (14%), botanical gardens/arboreta (11%), aquaria/zoos (7%), seed banks (4%) and private/contract research institutes (4%). To determine the level of organisation, we asked whether institutions have fully functional infrastructures with centralised storage locations that have been completed for more than 10 years (35%), between 5 and 10 years (24%), or less than 5 years (13%). Some institutions, however, were beginning the process to integrate their samples into a centralised facility (17%), whereas 11% were starting to be established. We also observed that several institutions classified themselves in different types of biobanks (Annex 3), meaning that they were not defined by a single specific kind of sample.

Some of the institutions (25%) adhere to defined best practices or standards, whereas in 7% these are in the process of being implemented, 2% did not know, 27% did not answer and 38% did not follow any standard. Diverse guidelines are being used such as the ones developed by ISBER, the Organisation for the Economics Cooperation and Development (OECD), the Global Genome Biodiversity Network (GGBN), the Food and Agriculture Organisation of the UN (FAO), the European Native Seed Conservation Network (ESCONET), the Millennium Seed Bank Partnership (MSBP), the Centre for Plant Conservation (CPC), the Society for the Preservation of Natural History Collections (SPNHC), the ICP Waters guidelines, those by CETAF, or they follow their own guidelines. Ancient human remain collections follow international forensic standards. Additionally, most biobanks are engaged in following practices regarding the different workflow procedures (Table 1).

Biobank facilities vary widely depending on the type of material stored. In general, facilities include laboratory refrigerators (4°C) (13), freezers (-12 to -30°C) (28), and/or ultracold mechanical freezers (-50 to 150°C) (23). As expected, museum and herbarium specimens were kept at room temperature (27). Only 21 institutions have liquid nitrogen cryovats (-80 to -196°C), including all those dealing with livestock germplasm. Furthermore, BEBs not only served as sample storage facilities, but also offer some add-on services (Table 2).

Sampling material. Different sample types are stored in the collections: tissues and fluids (28%), DNA (21%), living cells/germplasm (11%), living organisms/seeds (10%), environmental samples (10%), dried/fixed material (7%), DNA libraries (5%), PCR products (4%), RNA (3%) and pathological samples (1%). We observed that only 26 out of the 55 institutions are holding (genomic) DNA collections. Animal samples consisted mainly of blood, semen, tissue, hair, feathers, eggs, bones, and whole invertebrates. Plant samples consisted of pollen, seeds, fruits, flowers, cambium, and dried leaves. Microorganism samples came from plankton, cyanobacteria, fungi, spores, lichens, and oomycetes. Environmental samples covered were water, sediment, sewage sludge, soil, swabs, and faeces. Most of these samples were obtained from fieldwork/excursions (49), followed by living collections (in zoos, aquaria (16), botanical gardens (14)), museum sampling (22), individual laboratories (either in-house or out-house) (11), breed associations/farms (3), pathological/clinical departments (1), government agencies (1) and private collections (1). These samples are kept mainly to serve as backup storage for future use, such as captive breeding or propagation, as well as research in DNA-based and -omics techniques.

Specimen collection. Most of the institutions do follow collection protocols (74%). During fieldwork, the initial sample preservation methods include pure (flash-)freezing (36%), ethanol preservation

(27%), other chemical treatment (e.g., buffers, DMSO 13%), desiccants (10%), commercial reagents (8%), stabilization reagent kits (e.g., RNA later 3%) air dried (2%) and none (1%).

Most of the institutions (82%) have more than one labelling system for collected samples, comprising handwritten labels (33%), 2D barcodes (e.g., data matrix) (15%), linear barcodes (e.g., Code 128) (19%), self-adhesive cryolabels (30%), and printed straws (3%). Some NHCs use handwritten labels for old samples and barcodes for DNA samples and tissues. These labels may include a combination of the collector's number (30%), an accession number (33%), a unique biobank catalogue number (37%), or a combination. Some seedbank collections also include an additional seed lot number, since the accession number refers to the population. Often, printed labels are filled in by hand during fieldwork. Labelling is mainly made on vials (53%), whereas only 34% of the respondents do label both cap and label, and 13% did not answer this question.

Samples are usually collected by specialists during fieldwork, which helps to confirm species identification. At the biobank facilities, samples are morphologically examined and crosschecked with their corresponding voucher specimen before proceeding with DNA extractions. DNA barcoding/ karyotyping can also be performed, depending on the taxonomic group. Seed species confirmation is re-checked after germination, whereas domestic species have specific identification codes that allow species confirmation without performing morphological or genetic analyses (Fig. 1).

Preservation methods. Tissue samples are always (44%) or occasionally (29%) transferred to new vials. It is not commonly done in some institutions (27%), because appropriate vials were already used during field collection. In most of the cases (55%), starting preservation fluids were not removed or replaced, either because of time/human resources restrictions, uselessness, or not necessary, as samples were dried or special preservation fluids were used. 17% of the institutions mentioned that it was only done in case the ethanol concentration was too low, tubes were too full or if formalin or RNAlater had originally been used. Only 10% of the institutions include this step as standard practice before long-term storage. 18% of the respondents did not answer this question or did not know the answer.

Open flame (26%) and alcohol (24%) are the most common ways to sterilise instruments during sample transfer when processing a batch of samples. New material is always used for each sample in 22% of the cases, followed by bleach (7%) and others (5%) such as cyclohexane, hydrogen peroxide, hot bead sterilizer and steam sterilization in an autoclave. 1% mentioned that it was not necessary, as sealed plant samples are manually transferred to new vials.

When dealing with insufficient or rare material, less than half of the respondents (46%) pursue strategies to augment DNA quantities when needed (Fig. 2). We also observed comparable results regarding DNA quality control procedures, which are carried out mainly by request in half of the institutions (53%) (Fig. 3). Neither measure is a common practice at biodiversity biobanks.

Sample retrieval and material request. In the context of biobanks, DNA extractions are done most of the time after sample storage, usually on-demand. DNA extractions can also be done before storage, for instance, in the context of ongoing projects. 28% of the respondents mentioned that aliquots (backups) are produced to avoid damage through freeze/thaw cycles in the entire sample (Fig. 4). DNA is stored either in freezers or fridges, depending on when the DNA is going to be used. Another alternative given by a respondent to avoid freeze/thaw cycles is to not thaw the sample, by simply removing a piece of frozen tissue, which is brittle, while this remains on liquid nitrogen. However, this procedure is complicated for samples frozen in preservation fluid.

We received a diverse range of answers regarding the requested sample size or volume. In most cases, the curator, collection manager or grantee committee will advise users and finally decide on the sample size, depending on the DNA/tissue quality and quantity available. Moreover, other factors such as the likelihood of new requests, the ability to obtain similar samples from the wild, species rarity, research methodology and intended outcome are considered before determining the granted sample size. In general, plant biobanks grant 1-5 cuttings, between 10 and 50 seeds, and as much pollen as possible; livestock biobanks at least two straws per semen batch; cell culture/microorganism biobanks 15-20ml for cell suspensions or solid samples as cut pieces of agar in 2 ml cryotubes; wildlife biobanks between 5 to 15 ml blood or 1 cm^3 of frozen tissue; environmental biobanks between 1 and 18 kg soil (depending on the depth) and on average 2.5 kg for other environmental samples. Some NHC and botanical gardens simply grant the amount requested by the researcher.

The demand for biobank samples has increased over time in half of the institutions (51%) due primarily to 1) data being discoverable online (digitisation), enhancing visibility and global accessibility, 2) increasing number of research collaborations and projects, 3) collection growth, 4) breeders interested in storing samples according to breeding programmes, as well as increased interest by zoo/aquarium professionals and, 5) new techniques available for DNA/genome analyses. Two institutions reported a non-increasing sample demand due to the recent establishment of the biobank, lack of advertisement, or because it is not used optimally. So far, only 3% of the institutions have not received any shipping requests.

Varying approaches are used for shipping, depending on sample type. Most of the respondents send samples at room temperature, either in fixative/buffer (e.g., alcohol) (22%) or dried (26%). Seeds are sent in paper envelopes. Insulated containers including either cool packs (17%) or dry ice (12%) are also widely used. Vapour shippers (15%) and solutions containing DMSO (3%) are less frequently employed for shipping. Liquid cultures are sent in double packaging.

We also inquired about returns of requested samples to the biobank. The most common procedure is to keep returned material separate from the original samples (29%), followed by re-integration of the material together with the original sample (27%) and disposal of the material (20%). Returning leftover samples is not a universal practice, since 15% of the respondents mentioned that this has never been implemented in their facilities, and 7% do not accept returned samples.

Safety and security issues. Eight institutions (15%) allow only permanent trained staff to perform all biobank activities to protect the integrity and quality of the collection. In some cases (27%), students, interns and volunteers can assist with the banking processing (e.g., labelling), but are not allowed to be involved in initial sample preparation and subsampling (e.g., aliquoting), quality control, complex laboratory operations (e.g., making new solutions), liquid nitrogen safety, sample retrieval, using the collections management system, sample tracking, and core databasing, including the creation of new accession numbers, management and physical allocation of space, as well as storage organisation and, loans, including paperwork and shipment. One institute (2%) offers training to students before giving them access to the biobank, whereas the rest (34%) allow students to be involved in all biobanking procedures without restrictions. Biobanks provide a combination of training courses to staff, all of them in comparable proportions (Table 3).

Data standards. Regarding data storage, online databases are the most frequently used system by respondents (42%), followed by internal relational databases and Excel sheets (Fig. 5). Collection permits and export/import permits are not commonly stored in the same collection database (30%), whereas 6% are planning to incorporate permits into the database. Physical permits are mainly stored by the respective curator/museum department. In very few cases (12%), they are

electronically stored as multimedia files on their personal computers, on an internal server, or in an internal database/network/document management system. Otherwise, permits are referred to as absent/present in the database (4%) or links to permits are collected in a database (6%). Institutions that store permits in a databases (36%) have strict accessioning protocols, as well as associated paperwork attached to the accession record.

Half of the institutions (51%) provide field collectors with electronic data templates associated with vial labels to maintain consistency in data collection. These templates should become a common practice among institutions to harmonise data.

Transactions such as loans or requests are tracked in the database by most of the institutions (61%) to document sample use and/or remaining volume, as well as to obtain statistics of the collection's usage (Fig. 6). Institutes that do not follow this practice indicate as reasons: lack of staff, inability of the database to reflect this, or instead the use of an electronic document management system. Only 15% of the biobanks consistently follow and store the publications emerging from research carried out on their samples, whereas 58% do it sometimes. Almost a third of the respondents 27% are not aware of the research results obtained from their samples.

Regarding the use of LIMS (Laboratory Information Management Systems), only 12% of the institutions have their database (e.g., Specify, Arctos) linked to a LIMS. The information stored in LIMS includes, for instance, sample processing, DNA extraction procedures and DNA sequences, sample type, or children of samples.

At 29% of animal and plant repositories, it is mandatory to deposit (morphological) specimen vouchers along with banked tissue samples. However, in some cases (14%), photographs of the specimens (e-vouchers) are also accepted when there is no voucher available. Other institutions (36%) consider sometimes accepting samples without vouchers, depending on how rare/threatened the species are or depending on the country-specific permits (e.g., when countries do not allow whole specimens to be collected). 21% of the institutions accept samples without a voucher.

Database updates, regarding changing nomenclature that affects the taxonomic names of the samples, occur automatically (50%) in those institutions that have either a shared database or a database connected to the curated nomenclature database Yet, in 41% of the institutions, updates or changes must be done manually (Fig. 7).

Other topics/concerns mentioned in the survey included 1) handling costs and/or mailing fees, 2) ISO accreditation, which is often demanded but is not financially feasible for most biodiversity biobanks and can develop into a barrier to sharing and storing samples, 3) linking DNA samples with their respective specimen vouchers.

GGBN visibility. We observed there is still room for discoverability through the Global Genome Biodiversity Network for more than half of the institutions that took part in this survey. 31% of institutes are not aware of the existence of GGBN, whereas 41% do know it but are not members yet.

DISCUSSION

In the survey we conducted to evaluate procedures implemented at different biobank types, we found commonalities in sample collection practices. Comparable results were also obtained by Passemard et al. (2020) for livestock. Both Leroy et al. (2019) and Passemard et al. (2020) highlighted the need of sampling more at-risk/rare breeds along with preserving sufficient amounts of material

to allow breed reconstitution. We also realised that more capacity for sample collection aimed at animal cell culturing should be put into action (Ryder & Onuma 2018, Zimkus et al. 2018), as well as for collection of different tissue types per specimen (Zimkus et al. 2018, Bakker et al. 2020). Fungal diversity is still underrepresented in culture collections, due perhaps to the difficulties to isolate and culture most taxa (Paton et al. 2020). Additionally, more than 80% of described fungi are not deposited in collections, hampering their accessibility (Paton et al. 2020). Marine diversity banks and most environmental specimen banks have established sampling protocols, although these methods are difficult to standardise due to the diversity of studied sites and target taxa along with complicated technologies that have to be used (Küster et al. 2014, Rabone et al. 2019, Ruppert et al. 2019, Takahashi et al. 2023). However, preservation methods for this type of samples should be consolidated, allowing the reuse of samples without affecting the reproducibility factor (Paton et al. 2020).

Common practices also include sanitary and contamination controls throughout the biobanking process. Sanitary regulations for livestock can often be restrictive regarding conservation efforts of rare breeds (Passemard et al. 2020). Labelling (Miralles et al. 2020) varies greatly among collections. Significant progress has also been made in collection digitisation, web accessibility and compliance with the Nagoya Protocol, but much remains to be done, as indicated in Dunnum et al. (2018) and Paton et al. (2020).

We identified several common gaps and challenges classified into six topics, which are shown in Table 4. Although microbial culture collections were not included in our survey, staff working on this type of collections were contacted personally to have an insight into their practices. The survey results combined with the available literature helped us to recognise challenges and to provide some guidance where we considered it necessary.

Biobank management and facilities

Biobank standards, SOPs and defined workflows.— Both the survey results and the literature indicate that a number of collections does not adhere to any standards (De Vero et al. 2019). Various best practice guidelines for biobanks such as those released by OECD (2007), FAO (2012, 2014), ISBER (2018 and earlier editions), ISO 20387 (2018), WFCC (2010) provide technical, legal, and ethical recommendations and general requirements for the establishment and maintenance of biobanks. Other standards, such as the ISO FDIS 21899 (2020) or the ISO 21709 (2020) focus on validation procedures in the context of biotechnology operations. Biobanks should follow any of the mentioned guidelines or create their own set of SOPs, as high-quality standards should be met to collect, process, store, use and distribute biological samples, without affecting sample quality, functionality, and integrity (Zimkus & Ford 2014a). It is crucial that SOPs are carefully developed according to the organisms, sample types, and intended use to not only maximise the collection value, but also to guarantee consistency, comparability, reproducibility, and credibility (Korkaric & Beed 2010, Dunnum et al. 2018, Crop Trust 2020, Collins et al 2021, Angeles & Catap 2022). Recommendations and detailed information on which aspects to consider when establishing a SOP can be found in Zimkus & Ford (2014a).

We recognise that some specific guidelines are not easy to find due to limited distribution and visibility. Therefore, the creation of centralised document-sharing platforms such as Ocean Best Practices (Pearlman et al. 2019), FAANG (Harrison et al. 2021), the GGBN document library (Barker et al. 2019), the Museum genomics repository, and Protocols.io facilitates the retrieval of valuable protocols and literature, following the FAIR (Findable, Accessible, Interoperable and Reusable) principle (Collins et al. 2021).

Guidelines and protocols should also allow for flexibility so that they can be adapted to different key laboratory operations (e.g., identification, isolation, storage, growth), making processes cost-effective without losing the reproducibility factor (Ellis et al. 2018, Hagedorn et al. 2019).

Finally, staff and temporary personnel should understand and be aware of the active SOPs and guidelines at the BEB to guarantee that all polices are implemented (Zimkus & Ford 2014a).

Centralised facilities.— It is important that biological material and its by-products are stored in one central location that can be trusted by members of organisations or networks (Stackenbrandt et al. 2010). Thus, samples can be handled correctly for use and re-use (Staerk et al. 2018). However, biobanks are often managed decentrally (e.g. for individual departments only) within natural history collections or universities. This decentralisation can jeopardise sample quality in the long term, efficient sample accessibility and discoverability, and harmonisation of procedures within the institution.

Establishing central hubs is also possible when there are no biobank facilities at the research organisation, or if the BEBs do not include needed services, e.g. cryopreservation at -190°C. This type of arrangement allows for visits and training of personnel and use of resources following high standards (Hagedorn et al. 2019).

Lack of financial resources.—Biobank maintenance requires large budgets, often dependent on public or institutional funding (Smith et al. 2014, McCluskey 2017). Due to financial constraints BEBs may be closed down (Korkaric & Beed 2010, Boundy-Mills et al. 2020, Paton et al. 2020, Collins et al. 2021), especially when they start as personal collections or if collections are small or in disuse (Kang et al. 2006), unless they are taken over by other organisations (Boundy-Mills et al. 2020, Smith et al. 2020).

Ideally, BEBs should be fully and permanently funded, as human biobanks and biomedical model animal collections often are (McCluskey 2017, McCluskey et al. 2017a). Available financial resources are often too scarce to support the whole spectrum of BEB activities, organising expeditions, hiring sufficient staff, developing protocols, maintaining databases, and buying equipment (Ashmore 1997, Franco et al. 2006, Rice et al. 2006, Gostel et al. 2016, McCluskey 2017, Leroy et al. 2019, Collins et al. 2021, Perez Ortega 2022). Underfunding at collections like NHC has been addressed before (e.g., Dalton 2003, Kemp 2015, Astrin & Schubert 2017) and partly derives from the underrated value given to curatorial and taxonomic tasks in science (Collins et al. 2021). Additional financial sources are thus often needed for the unimpaired operation of BEBs. For instance, it has been suggested that user fees could be levied for identification, long-term repository and DNA-based services, and data mining (McCluskey 2017). Fees would depend on the difficulty, time and techniques used to preserve the samples (Smith et al. 2014). Public fund raising, contract research, grants, investments from charitable foundations are other options for increasing the biobank budget (Küster et al. 2014, Smith et al. 2014). Commercial use of samples is categorically ruled out for most biodiversity biobanks due to regulations applying to species trade (Comizzoli & Wildt 2017) and due to access regulations to genetic resources established through the Nagoya Protocol (see below) and other legal frameworks. Other possibilities for accessing additional financial resources include 1) establishing collaborations between BEBs in low-income and in high-income countries, providing funding and capacity building (de Vicente & Andersson 2006, Paton et al. 2020, Collins et al. 2021, Angeles & Catap 2022) 2) establishing collaborations with stakeholders and end-users (e.g., wildlife forensics) in funding proposals (Pérez-Espona & CryoArks Consortium 2021), and 3) the implementation of quantitative means that record the use of collections (e.g., publication, patent tracking) to attract attention of private and governmental institutions (McCluskey et al. 2017a).

Furthermore, long-term business plans for the sustainability and maintenance of biobanks should be established before curators retire, or research aims are adjusted (McCluskey 2017, Crop Trust 2020).

Personnel

Lack of personnel and dedicated staff.— Biobanks should employ sufficient personnel to carry out tasks related to sample processing, inventory control, quality management, distribution, database maintenance and development, and sustainability preparation (Vaught 2019). Smith et al. (2014), McCluskey (2017), and McCluskey et al. (2017) recommend having a minimum of three to six permanent staff for a medium size, at least for living microbe collections (e.g., 5000-10000 accessions), but if species/strain identification along with genomic/genotypic services and quality standards are to be implemented, additional staff would be required. Hence, not only a broad range of expertise is required to correctly handle the variety of samples stored in BEBs, but also a multidisciplinary team that can support crucial areas such as cryopreservation, tissue/cell culturing and specimen management, in some instances also taxonomy and molecular biology. Furthermore, the development of a staff succession plan would allow the transfer of knowledge when indispensable long-time employees retire (Crop Trust 2020).

BEBs are below the international guidelines' recommendations, as personnel is needed for collecting, sample accessioning, data management, viability tests, data analysis and web management , pathogen/strain/specimen identification and taxonomy, plant-pathology assessment and diagnostic assay development, , genetic resource management , (Kang et al. 2006, Walters & Hanner 2006, Xiangyu & Zhang 2006, Smith 2010, Ryan et al. 2019, Bakker et al. 2020, Crop Trust 2020, Broders et al. 2022). Reasons for staffing shortages in BEBs include insufficient funding, a shortage of trained individuals, elimination of positions, and difficulties in retaining qualified personnel (Korkaric & Beed 2010, Perez Ortega 2022).

Training programs.—Due to the lack of trained personnel, some biobank activities are carried out by students, interns and volunteers who may not have adequate qualifications (Zimkus & Ford 2014a). Ideally, training should be provided before giving them access to the biobank. Basic training should include biorisk management, equipment handling, laboratory procedures, and collection/data management. Building connections with universities is also an asset for training students, as already organised by MIRRI biobanks (Stackebrandt 2014). On the other hand, professional staff also need training for staying abreast of the latest technical procedures (Shivas et al. 2005, FAO 2012). Specific training courses should be developed particularly in data curation and management, specimen management (including isolation, storage, growth, and/or preservation of organisms), disease and zoonoses, species characterisation/taxonomy (Smith 2010, Ryan et al. 2019). Exchange visits with other biobanks can be useful for improving staff competences (Crop Trust 2020). Capacity building can also be improved by offering e-learning options (Fulton & Kresovich 2004, Bartels & Kotze 2006, Ebert et al. 2006), securing that knowledge and expertise remain available over time (Hagedorn et al. 2019). For instance, the Centre for Agriculture and Bioscience International (CABI) (Smith 2010) and the European, Middle Eastern and African Society for Biopreservation and Biobanking (ESBB) and ISBER continuously develop online courses or webinars regarding technical and practical aspects of working with collections. It is important to note that all training should be up to date, complying with good practices, and maintains process consistency (Zimkus & Ford 2014a).

Samples

Voucher accessioning and species identification.—Specimen vouchers as entire organisms are often deposited as (morphological) reference material in a public research collection or biobank for their long-term preservation and availability (Abd-Elsalam et al. 2010, Stackebrandt et al. 2014,

Groeneveld et al. 2016, McLean et al. 2016, Sikes et al. 2016, Ryan et al. 2019, Broders et al. 2022). Although specimen vouchering is common practice for animals and plants (Culley 2013, Clemann et al. 2014), it remains rare for microorganisms, plant-associated taxa and marine samples (Agerer et al. 2000, Korkaric & Beed 2010, Stackebrandt et al. 2014, Becker et al. 2019, Rabone et al. 2019, Angeles & Catap 2022). Microorganism genome sequences are frequently accessible online, but the strains are no longer extant, so reidentifications, further studies or discoveries are not possible (Boundy-Mills et al. 2020, Broders et al. 2022). A possible solution to increase accessioning rate is that funding agencies and journals request biobank accession numbers for all biomaterial collected and used for publication (Stackebrandt et al. 2014, McCluskey 2017, McCluskey et al. 2017a, Hobern et al. 2020).

Voucher material is used, among other aspects, as taxonomic and morphological reference to validate species identification, and it is essential for reproducibility of research (McCluskey et al. 2017a, Collins et al. 2021, González-Toral & Cires 2022). Because misidentifications and incorrect taxonomic status can lead to the waste of financial resources and detriment to the utility of specimens (Ryan et al. 2019, Broders et al. 2022), it is necessary to recur to online repositories (e.g., Fungal Names, Index Fungorum, MycoBank, WoRMS, Jstor database, Catalogue of Life, World Checklist of Vascular Plants, DynTaxa, VertNet) that can inform about current taxon names and correct nomenclature (Casaregola et al. 2016, Ryan et al. 2019, Bakker et al. 2020, Smith et al. 2020, Crous et al. 2021, Trujillo-Argueta et al. 2021).

Insufficient and rare material.—Samples are finite resources and will eventually deplete with usage (Collins et al. 2021). Hence, they should be used as efficiently as possible. Those coming from remote areas, rare species or that are underrepresented in the collection require special attention. Several options exist to replenish biomaterial (Dulloo et al. 2006). For instance, plant material can be grown in field genebanks or *in vitro* (de Vicente & Andersson 2006), and many microorganisms can be cultured. Animal cell culturing is also possible, but this technique is not yet widely used among BEBs, due to a lack of expertise and financial resources, and the need for rigorous standards for cell line authentication and contamination monitoring (McCluskey et al. 2017a, Hildebrandt et al. 2021). Alternatively, tissue samples can be aliquoted and stored to replenish DNA stocks (Andersson 2004) to prolong sample availability. BEBs should, however, consider restricting access to rare samples, if there is a risk that material will not be accessible in the future (Walters & Hanner 2006).

DNA banking.—DNA banking should be an essential component of every biodiversity biobank (Hodkinson et al. 2007, Gaudeul & Rouhan 2013, González-Toral & Cires 2022). However, we observed in our survey that only 47% of the respondents hold this type of collection. Similar results were found by Andersson (2004) and Passemard et al. (2020) for seed and livestock repositories, respectively. These repositories focus mainly on germplasm, and at least for livestock, ownership might be an issue for further sample usage (Passemard et al. 2020). Natural history collections, on the contrary, are expanding their genomic resource collections (Astrin et al. 2013), not least thanks to the technological advance in museomics (Card et al. 2021, Raxworthy & Smith 2021).

DNA banking can facilitate the establishment of core collections and the genetic characterisation of the collection (Ortiz & Engels 2004, Kang et al. 2006, Korkaric & Beed 2010, de Vero et al. 2019, Broders et al. 2022). It can also help to avoid exposing tissue samples repeatedly to freeze-thaw cycles. Moreover, extracting and storing genomic DNA and making it readily available for molecular applications is simpler, faster, and less expensive than culturing (de Vicente & Andersson 2006, Kang et al. 2006, McCluskey et al. 2017a), although it also lacks some of the benefits of culturing.

Back-ups and duplications.—Although this was not a survey question, we discovered that BEBs often do not have a backup site for their collections. Passemard et al. (2020) revealed that only 35% of

livestock germplasm collections and 11% of genomic collections have a duplication in a backup site, despite of holding unique material. Similar tendencies become apparent for wildlife (Comizzoli & Wildt 2017) and parasite repositories (Korkaric & Beed 2010), and some seedbanks (Crop Trust 2020).

Traditionally, biobanks generate backups by taking subsamples/aliquots of a sample and storing them in separate freezers. However, there are some potential risks associated with mechanical freezer breakdowns (Hanner et al. 2005) and especially with energy blackouts (Muller et al. 2016). The safer option, resources permitting, is safety duplication, which relies on the storage of a duplicated collection in a geographically separate location (e.g., mirror bank), to safeguard it as backup in case of physical disaster (Hodkinson et al. 2007, Uhlir 2011, FAO 2012, Comizzoli & Wildt 2017, CPC 2019, Boundy-Mills et al. 2020, Bakker et al. 2021, Hildebrandt et al. 2021, Priyanka et al. 2021). Backups are also helpful when the BEB is unable to provide adequate preservation (Korkaric & Beed 2010). In addition, animal germplasm can be backed up by storing somatic cells (FAO 2012). In plants (e.g., clonal crops), complementary backups are maintained in the field, *in vitro*, or cryopreserved (Dulloo et al. 2004, FAO 2014, Acker et al. 2017, Panis et al. 2020). For parasitic nematodes and microorganisms, two distinct preservation methods (e.g., cryopreservation and lyophilisation) are frequently combined to minimise the risk of loss (Korkaric & Beed 2010, De Vero et al. 2019).

An excellent example of a backup site is the Svalbard Global Seed Vault, which contains duplicates from most national agricultural seedbanks worldwide (Crop Trust 2020). The Seeds for Resilience project helps African seedbanks duplicate their crop collections at the Svalbard vault. Storage of animal genetic material is offered by the FrozenArk project (UK; Breithoff & Harrison 2018) and some institutional biobanks (e.g., the LIB Biobank at Museum Koenig, Bonn, Germany).

Quality control.—Several factors such as identity, authenticity and viability of the samples, as well as contamination, have to be monitored regularly to uphold sample quality (McCluskey et al. 2017a, ISBER 2018). Performing quality controls can save time and money, avoiding shifting the costs to later or further processes (Korkaric & Beed 2010, Hagedorn et al. 2019).

Increased demand of samples.—Thanks to digitisation, online discoverability, new genomic technologies, and growing numbers both of samples held at biobanks and of active scientists, loan requests have notably increased at most BEBs. This phenomenon was also observed in plant-pathogenic collections by Broders et al. (2022) and in plant and fungal collections by Paton et al. (2020). Preventive measures should be taken to mitigate impacts due to high sample demand. Biobanks should have adequate staffing, sustainability practices, and regulatory compliance (McCluskey 2017). Furthermore, granting DNA loans should be preferred over tissue loans (Ebert et al. 2006). DNA aliquots of small DNA quantities can be produced (Andersson 2004, Walters & Hanner 2006).

Returned samples—Re-integration of loaned material into the collection can lead to cross-contamination and other problems. When BEBs request that borrowed material should be returned after use, as it occurs in germplasm livestock collections (Passemard et al. 2020), samples should be stored separately, and accordingly documented.

Sample storage and preservation

The value of a sample depends on how it has been obtained and preserved (Walters & Hanner 2006, Comizzoli & Wildt 2017, Bakker et al. 2020) and on the quality and quantity of documentation. Zimkus et al. (2018) developed decision trees to assist researchers in selecting the type of sample

preservation method to use during fieldwork (on vertebrates) and considering options to preserve viable tissues. Samples should not be exposed to methanol, formalin, heat, and X-rays to avoid DNA damage (Bakker et al. 2020). Several measures have been described in Zimkus & Ford (2014a) to ensure the correct collection, processing, and storage of samples.

Samples are stored at various temperatures, ranging from room temperature to cryogenic (-196 °C) temperatures (Küster et al. 2014, Dunnum et al. 2018). Cryopreservation is the method of choice for long-term preservation of all types of organisms, regardless of the research goal (e.g., biodiversity conservation, artificial insemination) (Zimkus & Ford 2014a, Korkaric & Beed 2010, Breithoff & Harrison 2018, Dunnum et al. 2018, Passemard et al. 2020). However, there is no universal cryopreservation protocol that would hold for all species or genera (Brito et al. 2016, Hagedorn et al. 2019). Thus, protocols for the viable storage of material must be carefully adapted to the specificity of each taxon and cell type to maximise the potential for cryobanking (Lermen et al. 2009, Martínez-Páramo et al. 2016, De Vero et al. 2019, Cardoso et al. 2020, Pence & Bruns 2022).

Although numerous different methods exist to cryopreserve biomaterials (Hildebrandt et al. 2021), further protocols need to be developed or optimised for certain types of samples that are to date difficult or impossible to cryopreserve (Theilade & Petri 2003, Day et al. 2010, Comizzoli & Wildt 2017, De Vero et al. 2019, Kapoore et al. 2019, Leroy et al. 2019, Boundy-Mills et al. 2020, Campbell et al. 2020, Paton et al. 2020, Paredes et al. 2020, da Cruz et al. 2022, Pence & Bruns 2022), e.g., oocytes, avian and reptile sperm, local breeds, wild exceptional plants (e.g., tropical woody species) including recalcitrant and short-lived seeds, microalgae, basidiomycetes, and other recalcitrant microorganisms. Additionally, the limited use or poor success of some cryopreservation methods is also related to the scarcity of protocols to be implemented after recovery. For instance, species-specific protocols in plants are needed for tissue/cell culture, *in vitro* inoculation, germination, growth, propagation, and acclimatisation of plants (Engelmann 2000). In animals, further protocols are needed for recovering sperm motility (Clulow & Clulow 2016), assisted reproductive techniques (Saragusty & Arav 2011), and allotransplantation (Liptoi et al. 2020, Fujihara et al. 2022). These problems can only be overcome by understanding the organisms' physiology and the physiological conditions for recovery (Charlton et al. 2018, Paredes 2018, Hagedorn et al. 2019, Rain-Franco et al. 2021).

Several methods are used for the preservation and storage of microorganisms, including culturing, dehydration and freezing (Agarwal & Sharma 2006). Ryan et al. (2000) developed a decision-based key to help determine the most suitable preservation method, considering not only the species, structure, and water content (Ryan & Smith 2004), but also the user facilities and the economic factor.

Regarding the storage of viable plant material, conventional seed banking and cryopreservation of *in vitro* tissues are the most widely used methods. However, the former cannot be applied to desiccation-sensitive (recalcitrant) seeds, as they can undergo drying (FAO 2014, Paton et al. 2020, Ballesteros et al. 2020), and the latter is highly time-consuming, as it requires advanced production of gametophytes, shoot tips and somatic embryos, along with growing the tissues after retrieval (Pence & Bruns 2022). Cryopreservation of seeds, isolated embryos, embryonic axes, dormant buds, pollen, and spores of pteridophytes are other cost-effective or straightforward alternatives that do not require previous *in vitro* cultivation (Pence & Bruns 2022). Although feasible to cryopreserve, clonally propagated crops are often preserved in field genebanks or *in vitro* (Gueye et al. 2012), as there is still a lack of practical knowledge on how to replicate techniques without compromising the consistency of standard operations (Acker et al. 2017, Ellis et al. 2018). *In vitro* systems are a useful tool for the collection and propagation of plant germplasm, although there is an increased risk of contamination and somaclonal variation (Ruta et al. 2020).

Regarding the viable preservation of animal samples, there exist a number of protocols that allow live preservation of small invertebrates—whole or in parts (Smith & Chanley 1975, Smith et al. 2011, Thorp & Rogers 2015, Zinser et al. 2021, Roszkowska et al. 2021). Regarding the cryopreservation of cells, tissues, or embryos, considerably more research has gone into vertebrate taxa (but see, e.g., Paredes 2018, Gallichotte et al. 2021, Zhan et al. 2021, Aquino et al. 2022). Animal cryopreservation typically focuses on germplasm, but also on embryos/larvae and on somatic cells (Lermen et al. 2009, Singina et al. 2014, Martínez-Páramo et al. 2016).

In vitro cell cultures are an important tool for ex situ conservation and basic research due to their potential to maintain a renewable source of high-quality genetic material (e.g., genomic DNA) for in principle unlimited periods without affecting animal welfare (Houck et al. 2017, Sano et al. 2022). However, cryopreservation is an expensive method, especially in developing countries, as it requires special equipment, liquid nitrogen supply and specific maintenance (Theilade & Petri 2003, Gueye et al. 2012, Hildebrandt et al. 2021, Pérez-Espona & CryoArks Consortium 2021, da Cruz et al. 2022). Cryopreservation should be regarded as the best *ex situ* technique for conservation purposes. Nevertheless, additional preservation alternatives that are independent of freezing should be explored (Comizzoli & Wildt 2017, Anzalone et al. 2018).

Policies

The Nagoya Protocol (NP) on Access and Benefit Sharing (ABS) was created with the key aim of preventing biopiracy by providing a transparent framework for the fair and equitable access to genetic resources between provider and user countries, according to the Convention on Biological Diversity (Kursar 2011, Hurtado-Ortiz et al. 2019, Sherman & Henry 2020, Zimkus et al. 2021, Angeles & Catap 2022). Thus, NP gives sovereign rights to every state over their natural resources, making biodiversity conservation dependent on national legislations (Hurtado-Ortiz et al. 2019, Sirakaya 2021). In addition, NP has formalised sample exchange, especially in the microbial domain (Dedeurwaerdere 2010) and might help to increase public awareness of BEBs' limited financial resources (Rabone et al. 2019). Further details on ABS approaches can be found in Greiber (2013).

Although the principles behind NP are well-intentioned, the agreement has created an additional, complex layer of bureaucracy, which is complicated even more due to varying interpretations of NP by different countries (Watanabe 2015, McCluskey et al. 2017b, Prathapan et al. 2018, Staerk et al. 2018, Zimkus 2018, Hurtado-Ortiz et al. 2019, Boundy-Mills et al. 2020, Sherman & Henry 2020, Collins et al. 2021). NP has created difficulties to access genetic resources and obtain permits from signatory countries that still lack or are developing individual procedures for implementing the protocol (Prathapan et al. 2018). The number of accessions and loans have decreased due to NP ratification in certain countries (McCluskey et al. 2017b). International collaboration and national biodiversity research have been restricted, especially in developing countries (Kursar 2011, Prathapan et al. 2018). Long-term approaches are now needed to deal with the barriers to basic research introduced by NP (Pérez-Espona & CryoArks Consortium 2021, da Cruz et al. 2022). Although NP does not apply to marine biobanks that collect samples from regions outside national boundaries, NP can still add pressure for compliance of other agreements (e.g., biodiversity beyond national jurisdiction) (Rabone et al. 2019, Collins et al. 2021). The current classification of palaeontological and archaeological remains as genetic resources is debatable because it is impossible to predict in advance whether these samples will contain DNA (Lob & Botigué 2022). Historical collections also need to be aware whether NP is retrospective or prospective for implementation, as the temporal scope of NP varies (Sherman & Henry 2020). Further complications will arise should ABS regulations be extended to biodiversity data, as currently discussed (see von Wettberg & Khoury 2022 for further details).

Other protocols, such as The Cartagena Protocol on Biosafety and the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA) have also had an impact on the availability of living resources (McCluskey et al. 2017b). However, crops under ITPGRFA do not have to comply with NP and do not have to deal with bilateral agreements, which solves the issues connected to NP (Prathapan et al. 2018, Vogel et al. 2021). Several pleas for continuous uninterrupted access to genetic resources for livestock breeding and food production (Blackburn et al. 2014, Martyniuk et al. 2018), and taxonomy and conservation (Prathapan et al. 2018) have been made, as these research fields do not have a potential commercial value (e.g., in the form of patents).

Because BEBs facilitate the use of genetic resources, they must implement NP by 1) tracking and verifying that metadata and documents (e.g., permits, Prior Informed Consent, Material Transfer Agreements) are up to date for every sample/strain collected from October 2014 onwards (and also before where other regulations than NP apply), 2) contacting depositors to request further information, when incomplete, and 3) establishing compliance of newly accessed material (Hurtado-Ortiz et al. 2019). These tasks are laborious and require further human/financial resources, increasing biobanking costs (Hurtado-Ortiz et al. 2019, Collins et al. 2021)

Meanwhile, biobank networks have developed their own ABS guidelines (e.g., CETAF, GGBN, the step-by-step EMBRC guide, EMBRC guide to ABS, MIRRI, ECC, Davis & Borisenko 2017) to address these regulatory issues, providing support to biodiversity collections. Furthermore, to have a better understanding of NP, Zimkus (2018) recommends to 1) disseminate information on ABS, as in some cases their implications are still poorly known (Passemard et al. 2020), 2) educate staff on tracking procedures using databases, 3) use ABS clearing-House for identifying specific requirements per country, 4) apply for permits well in advance of starting field work, and 5) collaborate with local researchers.

Databases

We came across some general problems with databases that hinder standardisation processes. First, the use of Excel files or text documents for managing data, also observed in Casino et al. (2019), De Vero et al. (2019), and Passemard et al. (2020). We do not advise using these formats because they are not standardised, computationally demanding in requiring massively redundant data input (in comparison to a database), and frequently kept on personal computers where they may be lost, forgotten, or become obsolete (Yao et al. 2022). Hence, the use of relational database management software, either open-source, commercial or in-house-developed, is preferred (Hobern et al. 2022). Soon, graph databases may also be more intensively used in this context. Databases are necessary to make biobank data accessible and findable, using existing data standards, through international online platforms like GGBN or GBIF. A second common roadblock pertains to the lack of guidelines, time, and trained staff to manage and update databases. Rabone et al. (2019) exemplify this by the observation that data standards for marine biodiversity banks exist but are rarely employed. Third, we observed difficulties in the potential to re-use some of the available data for additional research. For instance, in the microbial domain, geographic information is often incomplete, hindering the understanding of the species' spatial distribution (Kang et al. 2006). Geographic information is a key component (alongside taxonomic information) in guiding international sampling and biobanking strategies, e.g. to concentrate collection efforts in underrepresented localities (Paton et al. 2020).

Biological databases are virtual infrastructures that provide accurate and relevant data about a particular biomaterial that can be easily retrieved (Stackebrandt 2014, Breithoff & Harrison 2018). These data (quantitative, qualitative, spatial, and imaging data, etc.) must be curated, regularly updated, and duplicated (Comizzoli & Wildt 2017), following the FAIR principles and specific

workflows to avoid database disuse (Howe et al. 2008, Wilkinson et al. 2016, Odell et al. 2017, Rabone et al. 2019). Ideally, each type of BEB ought to have its own, but interoperable set of standards to facilitate the accessibility and reuse of data. Examples of integrated databases are AgBioData (Odell et al. 2017), EFABISnet/CryoWEB (Duchev et al. 2010), and Genesys, providing database standards/templates and data sharing recommendations applied to the agricultural/livestock domain. Not all BEBs report their data in this type of integrated databases (Leroy et al. 2019).

Databases are a priority for all types of biobanks (Comizzoli & Wildt 2017). Hence, certain factors need to be considered when curating and managing databases (Kang et al. 2006, Deck et al. 2012, Stackebrandt 2014, Chase et al. 2016, Casaregola et al. 2016, McCluskey 2017, McCluskey et al. 2017a, Odell et al. 2017, Triebel et al. 2018, De Vero et al. 2019, Boundy-Mills et al. 2020, Damerow et al. 2021, Pérez-Espona & CryoArks Consortium 2021, Angeles & Catap 2022, Hobern et al. 2022, Yao et al. 2022):

- Data quality should be validated, manually and through the use of software packages such as CoordinateCleaner, used for geographical and temporal data cleaning (Zizka et al. 2019). Taxonomists should be involved in the validation of data (Paton et al. 2020).
- Standardised common metadata and appropriate semantics/ontologies/vocabulary to facilitate connections or interactions between databases and manual/automated data curation. Metadata should comply with the Darwin Core format standard, the ABCD (Access to Biological Collection Data 2005, Holetschek et al. 2012) standard, the ISO 21710 for microbial resources, the Multi-crop passport descriptors (Alercia et al. 2015), or the GGBN data standard (Droege et al. 2016).
- A globally unique identifier (GUID, or UUID for universally unique identifier) is highly relevant for sample tracking, particularly when material (e.g., strains, cultivated varieties, cell lines and animal resources) is shared among various collections, but accession numbers are not consistent. Using GUIDs helps to avoid duplications and data loss. In addition, subsampled tissues, viable cells, and derivatives (e.g., DNA) should also have unique identifiers, in addition to the source specimen ID, to define sample uses.
- Information on the sample (e.g., taxonomic identification, genetic data), its history (e.g., permits), and its practical management and usage (e.g., storage location) should all be included in the database/s.
- High-quality images of the specimens should be databased and made publicly accessible (Bakker et al. 2020, Miralles et al. 2020, Lawniczak et al. 2022), especially when destructive sampling is required.
- Mechanisms to restrict the publication of sensitive data should be in place, e.g., the geographic location of vulnerable populations, or data elements that fall under data protection.
- Data mining and visualisation tools (e.g., distribution maps) maximise the use of the data. The Paleobiology database exemplifies such data visualisation.
- Routine updates should be implemented in the biobank workflow to screen for inconsistencies and to add new information.

Discoverability and access to samples.—BEBs lacking an online presence are still common, mainly due to reduced Internet connectivity or use of data storage systems that are incompatible with digital resources (Hodkinson et al. 2007, Crop Trust 2020). Hence, these collections can only be discovered through personal contacts, conferences, or publications (De Vero et al. 2019). Small-size livestock biobanks holding a limited number of samples sometimes narrow down sample access for any but exceptional or urgent uses (Passemard et al. 2020). According to Küster et al. (2014) environmental specimen banks (ESB) have some publicly accessible data on their own websites, but

ESB data are still hardly visible and inaccessible for researchers. Digitisation processes are essential for data and sample discoverability, availability, and accessibility (Yao et al. 2022). Through digitisation, a collection can also efficiently and accurately document its use in research for funding organisations (Dunnum et al. 2018). Once a database is developed, it can gain visibility by using open-access web-based interfaces or through centralised databases or data aggregators (e.g., GGBN, GBIF) (Korkaric & Beed 2010, Casaregola et al. 2016, Zimkus & Ford 2014a, Paton et al. 2020). Many initiatives are taking place to digitise and characterise collections, although this task has to be further prioritised at natural history collections (Astrin & Schubert 2017, Hobern et al. 2020, Card et al. 2021). In addition, BEBs can become visible by being part of a network or registry comparable to GRSciColl, CETAF, Index Herbariorum, iDigBio, DiSSCo, DOE, GGBN, GRBio, or GRIN-Global (Rabone et al. 2019, Bakker et al. 2020, Crop Trust 2020, Hobern et al. 2020, Islam 2020, Collins et al. 2021, Damerow et al. 2021).

Sample traceability and sample use.— For transparency and ABS compliance, digital records of sample-associated permits and transactions should be tracked (Watanabe 2015, Casaregola et al. 2016, Zimkus 2018, Zimkus et al. 2021, Angeles & Catap 2022). A list of further ABS-relevant documents can be found on the Society for the Preservation of Natural History Collections (SPNHC) wiki page. If electronic storage of documents is not possible, links that direct to the permits should be created (see Zimkus et al. 2021 for further details).

In addition to permit tracking, the quantitative tracing of sample use makes it possible to evaluate the collections' impact (McCluskey & Wiest 2011, Smith et al. 2014, Dunnum et al. 2018, Paton et al. 2020) and to justify their funding. Ideally, the tracking system should integrate information regarding institutions that have requested material, publications, and GenBank/ENA data (de Vicente & Andersson 2006, Bakker et al. 2020). A possible tracing mechanism is the creation of a catalogue or a Google Scholar/ ResearchGate profile linking publication DOIs (digital object identifiers) with the samples that generated those results (Dunnum et al. 2018, Crop Trust 2020). The sedaDNA Scientific Society uses a comparable system to track literature regarding sedimentary ancient DNA. The tracking systems of CABRI or StrainInfo have been developed for tracing the exchange history of strains (Casaregola et al. 2016, McCluskey et al. 2017a). As mentioned above, the use and citation of persistent uniform identifiers (GUID) is important. GUIDs can link specimen data with the occurrence of the specimen in different datasets (McCluskey et al. 2017a, Rabone et al. 2019, Paton et al. 2020).

Use of LIMS.— LIMS are databases designed to maintain and to track, in a centralised manner, the different workflow stages concerning sample processing, experimental procedures (e.g., physiological tests, DNA analyses), and handling (Vu et al. 2012, Dolle et al. 2020), ensuring data integrity and system usability (Agilent SLIMS 2018). Moreover, some LIMS directly integrate freezer management functions for tracking the physical storage of not only the voucher specimens but also their by-products, i.e., molecular vouchers (Bilkhu et al. 2017). This type of database is widely used among zoos and aquaria (ZIMS: Zoological Information Management Software) as an easy and transparent practice to store and share sample records electronically (Hvilsom et al. 2022). ZIMS records data on animal health, species husbandry and ancestry, facilitating the association of phenotypic and genotypic data of each individual animal (Staerk et al. 2018). On the contrary, the use of LIMS at natural history collections and livestock collections is not yet very common and sometimes laboratory procedures are still recorded in spreadsheets, text documents or notebooks (Casino et al. 2019, Passemard et al. 2020).

Networking

Our survey revealed that most BEBs (72%) were not aware of or not yet members of the Global Genome Biodiversity Network (for which the survey was conducted). In their survey of livestock germplasm collections, Passemard et al. (2020) discovered similar results: only 33% of the respondents belonged to the EUGENA livestock network and 4% were part of any international networks. In addition, Hodkinson et al. (2007) pointed out that biobanks that are already organized in networks often continue operating incoherently. Zika et al. (2011) revealed that due to regulatory issues, sharing of samples among biobanks is limited. We advocate the need to connect with other BEBs for the following reasons: 1) to share knowledge (on collection management, biodiversity, etc.), 2) to foster and facilitate collaboration, 3) to establish common operating strategies for collecting, processing, and storing tissues, cells, and nucleotide samples in agreement with best practices, and to establish such best practices (e.g., ISBER), 4) to harmonise data management and exchange, 5) to implement common policies for legal and easy access to samples, and 6) to help establish or fund facilities in megadiverse countries (Zika et al. 2011, Smith et al. 2014, Seberg et al. 2016, Comizzoli & Wildt 2017, Angeles & Catap 2022). Additionally, networks can merge all collection data by using common online databases for sample accessibility and discoverability, thereby centralising the data, and optimising the use of collections (Zika et al. 2011, Droege et al. 2013. In the long run, global networks can promote and support the genetic characterisation of collections and the broadening of provided services to prevent further genetic diversity loss (Fauna Research Alliance).

Several global international network initiatives or consortia have been established (e.g., GGBN, ISBER, ESBB, and the Asian Network of Research Resource Centers ANRRC) and are actively engaging new and existing collections and other institutions (e.g., museums, zoos, universities, NGO's, conservation agencies) to become members.

Among the networks of microorganism collections are the World Federation for Culture Collections (WFCC), the Latin American network of culture collections (FELACC), the Asian Biological Resource Centers Network (ABRCN), the European Consortium of Microbial Resources Centres (EMbaRC), the European Culture Collections' Organisation (ECCO), the MIRCEN UNESCO Microbial Resources Centers, the Microbial Resource Research Infrastructure (MIRRI), the US Culture Collection Network (USCCN) and the US National Plant Diagnostic Network (NPDN). Among plant networks, mainly for seed banking, are the Millennium Seed Bank Partnership (MSBP), the Mid-Atlantic Regional Seed Bank (MARSB), and the Australian Seed Bank Partnership (ASBP). Livestock and agriculture networks comprise, among others, the Consultative Group on International Agricultural Research (CGIAR), the Aquaculture Infrastructures for Excellence in European Fish Research (AquaExcel), the European Genebank Network for Animal Genetic Resources (EUGENA), and the Brazilian Agricultural Research Corporation (EMBRAPA). Wildlife networks include, among others, FrozenArk, CryoArks, the Pan-Smithsonian Cryo-Initiative (PSCI), the Israel-German Ark of Life (IGAL), the Biodiversity Collections Network, the FAUNAbank, the Amphibia Bank, the Biodiversity Biobanks South Africa (SAS), and the future Global Wildlife Biobank. The IUCN SSC (Species Survival Commission) animal biobanking for conservation specialist group has recently been created to work as a global network for wildlife cell and tissue culturing. The International Environmental Specimen Banking Group (IESB) and the SedaDNA Scientific Society bring together environmental specimen collections and sedimentary ancient DNA collections, respectively. Networks for microbiomes are still lacking and should become a priority for biodiversity conservation (De Vero et al. 2019, Meisner et al. 2022).

A considerable number of network initiatives in Europe have been funded by the European Commission, both for human (e.g., BBMRI-ERIC, EuroBioBank, PHOEBE) and biodiversity (e.g.,

EMbaRC, MIRRI, SYNTHESYS+) biobanking. Networks increase the chances to obtain the funding needed for equipment, hiring staff, and developing databases (Comizzoli & Wildt 2017).

Conferences such as '*Harmonizing Biobank Research: Maximizing Value- Maximising Use*' (PHOEBE 2009), GGBN conferences (e.g., https://ggbn2023.weebly.com/), the ESBB and ISBER conference series, or the "*Plant and Animal Genome conference*" provide the chance to share accomplishments and identify problems and potential solutions, as well as new collaborations. Most importantly, these meetings can facilitate interactions between different biodiversity biobank communities that rarely work together (due to, e.g., different sample handling methods), but can also bring together the biodiversity and the human biobank communities, which share requirements and challenges in most basic biobanking processes (Comizzoli & Wildt 2017, Ryan et al. 2019, Angeles & Catap 2022). Such interactions can be instructive to biodiversity and environmental biobank staff, as technological progress often originates in the better-funded human domain. Hence, the organisation of network activities such as workshops, conferences, training, or mentorship strategies (Küster et al. 2014, Rabone et al. 2019) can help not only for clarifying or exchanging ideas on operating procedures, ethics, or legal issues but also for promoting connectivity, interaction, and network consolidation. These types of events can also assist in preventing inefficiencies and reiterations when establishing protocols within the biobank workflow (PHOEBE 2009). Creating a calendar of relevant meetings is useful to enhance networking (Hagedorn et al. 2019).

Further challenges

Aside from the issues addressed in the survey and the literature, ownership standards, intellectual property rights, and quality management certifications have received little attention so far (Comizzoli & Wildt 2017). BEBs in the public domain do not necessarily claim ownership rights on the stored biomaterials (Hurtado-Ortiz et al. 2019, Passemard et al. 2020), as commercial research is precluded in many biodiversity biobanks due to ABS regulations. Nonetheless, many public collections may require a transfer of ownership to reduce the bureaucratic burden introduced by custodianship, or to compensate for curation, storage, and data management expenses (Rabone et al. 2019). Ownership can become very relevant in livestock samples, where germplasm can belong to individual farmers and breeders, cooperatives, and international companies (Blackburn et al. 2014, Martyniuk et al. 2018).

When it comes to patents, copyrights, and trade secrets derived from organisms and their byproducts, intellectual property rights (IP) may be claimed (Chiarolla 2013). IP may impede the dissemination of knowledge and research (Vaught & Lockhart 2012). They can conflict with ABS regulations unless joint ownership of intellectual property is considered or if IP is excluded/assigned when negotiating bilateral PICs and MATs (CETAF 2019). Certain regulations, such as ITPGRFA, can also rule out IP (Chiarolla 2013).

Biobanks can implement a quality management system (QMS) and can obtain an accreditation that ensures that the biobank workflow is conducted in accordance with standardised procedures and includes mechanisms for self-improvement (Davis et al. 2012, Smith & Ryan 2012, Martins et al. 2017, González et al. 2018). Among biodiversity biobanks, this is most frequently implemented in biobanks focusing on microorganisms. The process of certification needs dedication of staff and resources, as well as the formulation of several documents, including SOPs, quality control manuals, management and planning programs, and data/records management standards (González et al. 2018). To date the most common certification is ISO 9001 (2015), which provides a framework for any type of organisation for describing, recording and controlling processes effectively (Martins et al. 2017), as well as facilitating training of new staff (Martins et al. 2017). ISBER has developed a self-

assessment tool for biobanks to determine not only how well best practices (e.g., ISBER) or standards (ISO 20387) are implemented, but also in which areas improvements can be made.

Why biobanks are important

Biobanks used to be considered purely storage facilities, but today they are seen as active collections and central research infrastructures (Breithoff & Harrison 2018). Biobanks are involved in the development of effective techniques for collecting, preserving, and storing biomaterials, including both specimen and molecular vouchers (Dunnum et al. 2018). They equally focus on metadata and data architecture. BEBs are essential infrastructures not only to preserve and provide samples from different groups of organisms but also to sustain innovation, food security, natural resource management, biotechnology, and biological research (Hanner & Gregory 2007, Martins et al. 2017, González et al. 2018, Breithoff & Harrison 2018, Bajerski et al. 2021). Furthermore, biobanks can be used to address aspects regarding wildlife forensics, conservation, environmental change, human and environmental health, zoonoses and plant pathogen incursion history and their respective biosecurity responses (Kang et al. 2006, Smith et al. 2014, Dunnum et al. 2018, Bakker et al. 2020, Pérez-Espona & CryoArks Consortium 2021, Angeles & Catap 2022, Broders et al. 2022). BEBs can also function as archives of samples collected in different environments at different temporal and spatial scales, upholding genetic variability data within taxa (Hildebrandt et al. 2021, Broders et al. 2022). For these reasons, biobank curators, collection managers, and stakeholders should increase public awareness of the potential and benefits of biobanks as decision-making tools in biodiversity conservation. Without biobanks, identification, and preservation of samples, as well as contamination control would be unreliable and suboptimal, and research reproducibility would be challenging (McCluskey et al. 2017a).

It is commonly recognized that further BEBs should be established, especially in regions with high biodiversity, to counteract biodiversity loss (Angeles & Catap 2022). To achieve this goal, issues concerning capacity, awareness, and financial resources to maintain biobanks need to be resolved.

Recommendations

We are aware that our survey results are limited in scope. However, they show a trend that is consistent with the literature review. Taken together, they provide insight into possible strategies that can be implemented in the biobanking workflows. As noted by the Crop Trust (2020), most challenges derive from flaws in operational procedures, equipment, and facilities. We therefore recommend the following to improve BEB workflows:

1. Add value to the collections through increased efforts in sample identification. Ideally, samples should be genetically characterised by using DNA barcodes, or SNP arrays for economically important species (e.g., livestock, hardwood species, crops) (IMAGE 2020, Bernhardsson et al. 2021, Keeble-Gagnère et al. 2021) without disregarding classical taxonomy.
2. Raise awareness of available best practices and adhere to them. Protocols and policies should be flexible so that they can adapt to different operating conditions without compromising standards and reproducibility. They should assist in harmonising processes and sharing knowledge, rather than creating obstacles. Guidelines should be available both in digital and printed form for biobank staff and new personnel.
3. Improve human resources by identifying further sources of funding and build capacity for current and new staff. Apart from third party funding and the provision of services, BEBs should request a commitment for appropriate funding from the host institution (e.g., museums, universities, or governmental research institutes) (Rice et al. 2006).

4. Promote positive outcomes (e.g., improvements in phytosanitary status, germplasm protection) (Ashmore 1997) and uses of collections as part of a financial strategy.
5. Embrace cryopreservation as it is by far the best method to maintain sample viability in the long term and to avoid spontaneous mutations (McCluskey et al. 2017a). Nonetheless, it may be worth exploring alternative preservation methods to freezing, as liquid nitrogen can be unavailable in certain situations (Comizzoli & Wildt 2017). Continued improvement in cryopreservation and *in vitro* techniques will further enhance sample preservation so that samples can be used for existing end emerging technologies in the future.
6. DNA banking should constitute a routine procedure in BEBs. *In vitro* cell or tissue culturing approaches are also essential for plant and animal genetic resources, especially for conservation plans and for applications that rely on high-quality material.
7. Set up a specimen management plan for compliance with the Nagoya Protocol and other regulations.
8. Implement the use of LIMS and freezer management solutions to maintain a comprehensive overview of sample use and storage.
9. Rely on (ideally open-access) relational databases or graph databases to store as much accurate metadata as possible, even if the data do not seem relevant at the time of collection (Broders et al. 2022).
10. Increase data compatibility and comparability by using metadata standards (e.g., GGBN data standard or DwC. When possible, automation should be favoured over manual data entry.
11. Increase communication and invest in the development of collaborative networks to establish common procedures, improve services, and create common data management systems to facilitate access to collections. Engaging with stakeholders such as NGOs, zoos, aquaria, farmers' organisations, seed producers and community seed banks (Koller et al. 2018), breeding and hunting associations, and other users can also be useful for the acquisition of new genetic resources and for supporting characterisation (CPSG 2019, Leroy et al. 2019, Crop Trust 2020, Paton et al. 2020, Hildebrandt et al. 2021). Involving stakeholders will improve conservation plans in the long-term (CPSG 2019).
12. Increase BEB visibility through network membership (e.g., GGBN).

"*The perfect biobank should have standardised operating procedures for the technical work, dependable funding to ensure the facilities can be maintained, and one database to log every existing sample so they can be fully utilised in research*" (Hales 2021). We recognise that any recommendations need to be flexible, as BEBs have different uses, objectives, and designs (da Cruz et al. 2022). This flexibility must nevertheless allow for comprehensive interoperability. Overall, our results and literature research indicate that BEBs need to further align with best practices and develop accurate SOPs. To fill these gaps at the technical level, we have developed a handbook explicitly aimed at the BEB community. This open access handbook on protocols and practices in biodiversity biobanking is available under DOI 10.3897/ab.e101876. It provides guidance and recommendations on field sampling, preservation, storage of biomaterials, and management procedures, and contributes to the standardisation of biodiversity and environmental biobanking practices. The handbook is meant to give a comprehensive overview of biobanking and cryopreservation procedures for protists, fungi, lichens, plants, and animals, as well as for environmental samples and palaeontological remains. We recommend using the handbook together with species- or group-specific information to adapt or improve protocols.

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Tables

Table 1. Overall overview and proportion of biobanks applying diverse standard practices within their workflows

Practice/policy	
General best practices and standards	14
Collection protocols	41
Quality control procedures	29
Viability controls	16
Sample use policy	27
Sample request standards	39
Emergency plans	38
Sanitary/ hygiene plans	36
Custody records	40
ABS policy	42

Table 2. Proportion of biobanks offering add-on services

Biobank Services	**Proportion**
Sample collection/ preservation kits	17
Technical advice	30
Sample storage	20
Sample Loans	6
Training	1
DNA extraction	12
Library preparation	2
PCR/ sequencing	8
Cell culturing	6
Education	2
Sampling	2
Archaeozoological analyses	1

Table 3. Proportion of biobanks offering training to staff in various practices

Training Course	**Proportion**
Toxic substances/ Biosecurity/ Zoonosis	30
Handling of liquid nitrogen	22
Electrical equipment and monitoring systems	25
Principles of care and maintenance	42
Policies and ethical issues	28
Databases	37

Table 4. Gaps and challenges found in the survey, established from communication with biodiversity biobank curators/managers and confirmed by the literature

Topic	Identified Gap
General biobank management	Lack of formally documented protocols/SOPs Unclear workflows No standardised protocols within same institution Not following biobank standards No centralised facility Lack of resources for expeditions/reagents/equipment
Personnel	Lack of financial resources, hence lack of human resources Lack of training programmes for students/interns/volunteers No dedicated staff for managing DNA collections and databases
Samples	No consistent deposition of specimen vouchers or molecular vouchers Limited options for insufficient or rare material Lack of backups and duplications Poor quality control No measures to deal with increased demand of samples Returned samples and contamination issue
Sample Preservation and Storage	Liquid nitrogen expenses Insufficient DNA banking
Policies	Nagoya protocol ISO accreditation
Databases	Digitisation backlogs Databases not often updated Lack of LIMS (custody recording) Permits are kept by collector, often not linked to specimen in DB Poor traceable transaction No traceable publications Collection not yet discoverable (i.e., through GGBN)

Figures

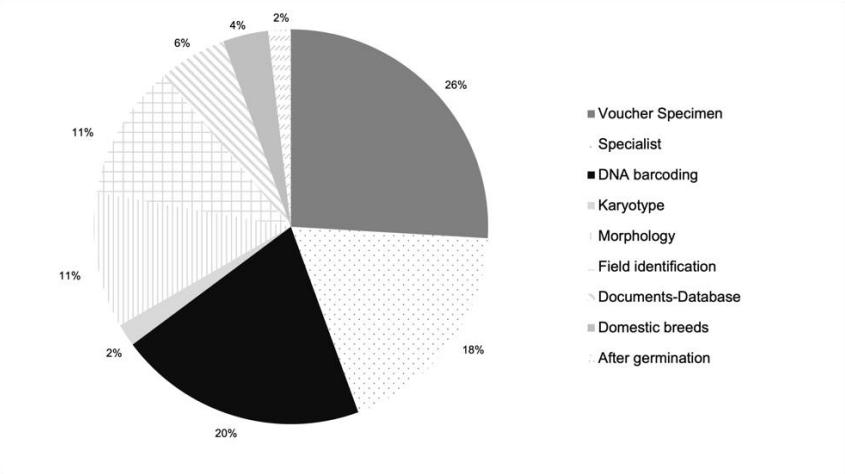

Fig 1. Confirmation of species identification measures after fieldwork has taken place.

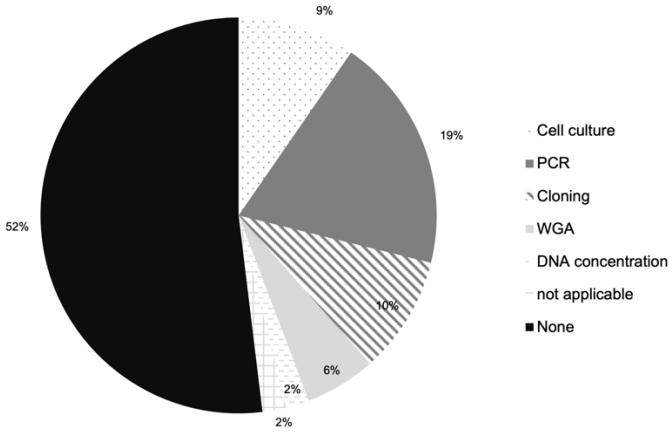

Fig 2. Procedures used to augment DNA quantities from insufficient and rare material

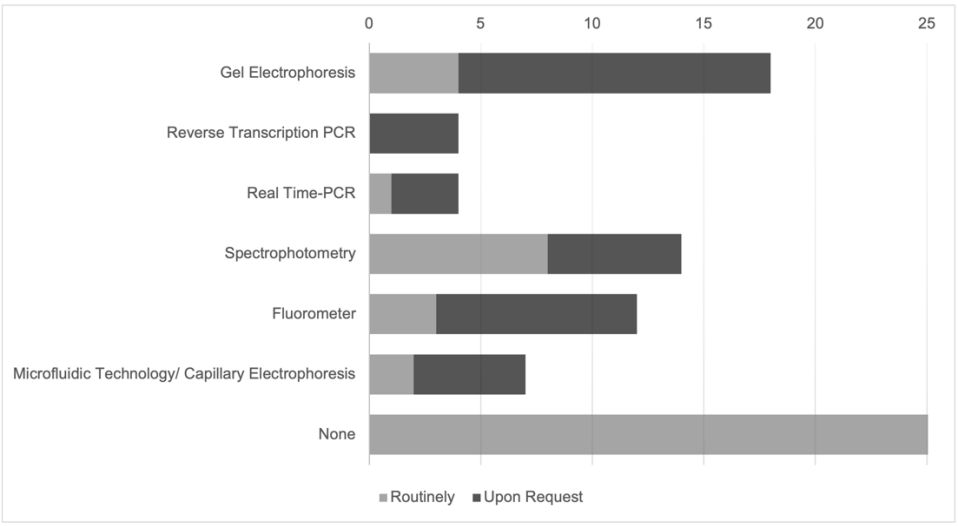

Fig 3. Quality control procedures that are performed routinely and upon request at different biodiversity biobanks

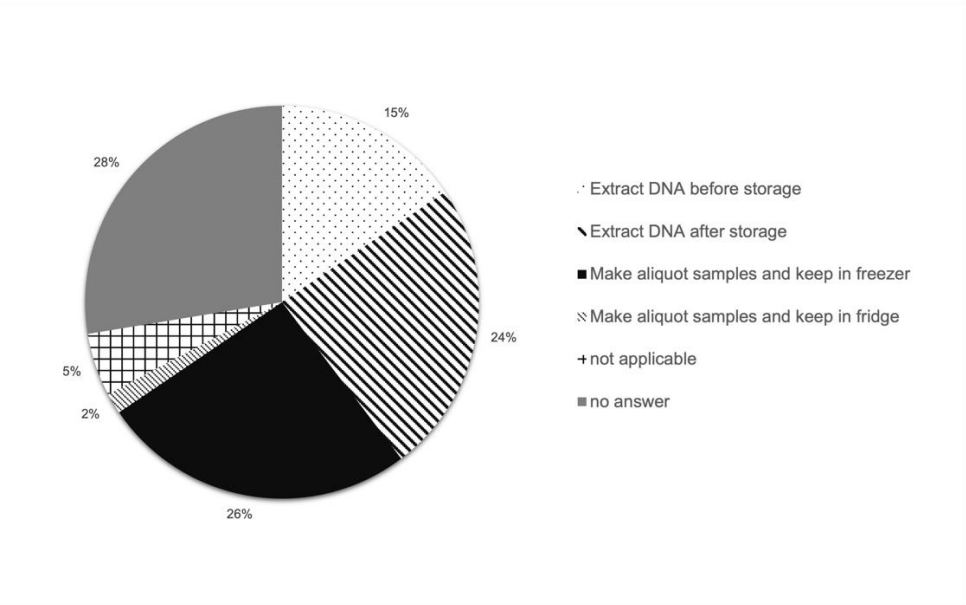

Fig 4. Measures taken to avoid freeze/thaw cycles

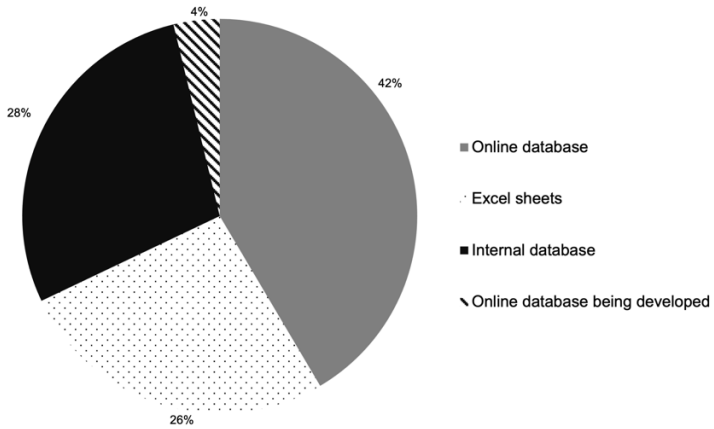

Fig 5. Types of databases (or workarounds) used at the biobanks

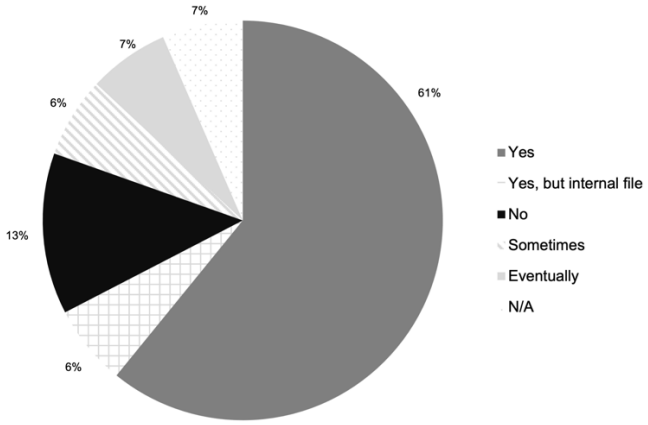

Fig 6. Sample transactions (loans/requests) tracked in the database

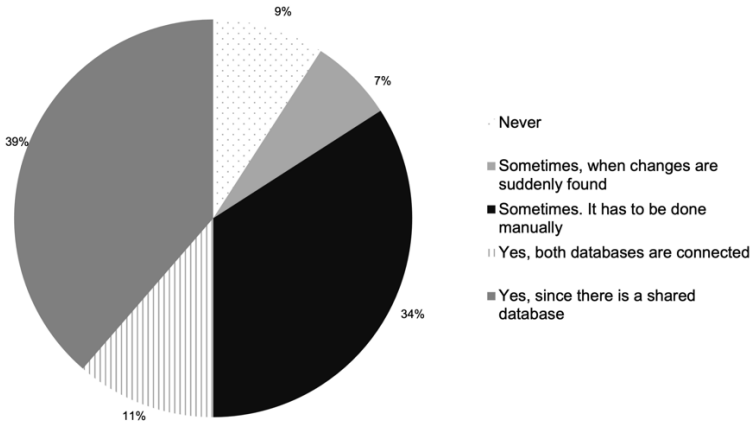

Fig 7. Database updates when voucher information has changed

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Annex 1. Questionnaire

SYNTHESYS+ (https://www.synthesys.info/) is a European Commission - funded project, creating an integrated European infrastructure for natural history collections.

Within SYNTHESYS+, subproject NA3.1 (run by the Global Genome Biodiversity Network) performs a landscape analysis of biodiversity and environmental biobanks and their standards and practices.

The present survey investigates biobank efforts and procedures in distinct domains of biodiversity/environmental biobanking (e.g., DNA/tissue storage methodologies, legal policies and ethical issues).

Your responses will provide us with technical details that will help to eventually compile a handbook on both existing and missing standard operating procedures (SOPs) specifically for biodiversity and environmental biobanks/repositories/molecular collections.

Thank you very much for your help!

Data protection statement: “the personal data you provide will not be shared with third parties. Your data will be stored by us and will be deleted after the survey storage is no longer necessary”

Institution’s name _____

Name, Role _____

Email address _____

GENERAL COLLECTION INFO

1. Type of institution (Check all that apply)
   - ☐ Natural History Museum
   - ☐ Herbarium
   - ☐ Botanical Garden/Arboretum
   - ☐ Aquarium/Zoo
   - ☐ Private institute/laboratory
   - ☐ Seed Bank
   - ☐ University
   - ☐ Other (please specify)

2. Type of biobank/repository (Check all that apply)
   - ☐ Seed bank/crop
   - ☐ Livestock/veterinary
   - ☐ Aquarium/Zoo
   - ☐ Living plant collection
   - ☐ Parasitology
   - ☐ Plant culture collection
   - ☐ Animal culture collection

- ☐ Microbial culture collection
- ☐ Environmental specimen bank (ESB)
- ☐ Natural history collection
- ☐ Wildlife and wood forensics
- ☐ Other_____

3. What type of material is found in your biobank/repository? (Check all that apply)
   - ☐ Tissue and/or blood
   - ☐ DNA
   - ☐ DNA libraries
   - ☐ PCR products
   - ☐ RNA
   - ☐ Proteins
   - ☐ Living cells (e.g. cultures)
   - ☐ Living organisms (including seeds)
   - ☐ Environmental samples
   - ☐ Other _____

4. Where did the samples originate from?
   - ☐ Excursions/Field work
   - ☐ Aquaria/Zoo
   - ☐ Botanical gardens
   - ☐ Museums
   - ☐ Pathological/clinical departments
   - ☐ Individual laboratories (either in-house or out-house)
   - ☐ Other _____

5. What is the current development status of your biobank?
   - ☐ Just starting, facility's infrastructure is not yet completed
   - ☐ Infrastructure is completed and samples are being integrated into the (centralized) facility

   Samples are already in permanent storage and...
   - ☐ ...facility's infrastructure is +/- complete since less than 5 years
   - ☐ ...facility's infrastructure has been +/- complete 5-10 years ago
   - ☐ ...facility's infrastructure has been +/- complete for more than 10 years ago

6. Which storage temperatures are being used in your biobank? (Check all that apply)
   - ☐ Room temperature storage (e.g. dried, sealed, ...)
   - ☐ Laboratory refrigerator (4°C)
   - ☐ Laboratory freezers (-12 to -30°C)
   - ☐ Ultracold mechanical freezers (-50 to -150°C)
   - ☐ Liquid nitrogen cryovats (-80 to -196°C)
   - ☐ Other_____

7. Which services are provided by the biobank? (Check all that apply)
   - ☐ Collection kits and sample preservation kits
   - ☐ DNA extraction
   - ☐ PCR and/or sequencing
   - ☐ DNA library preparation
   - ☐ "Trust" sample storage

- ☐ Cell culturing
- ☐ Technical advice on collecting practices
- ☐ Other_____

8. Does your biobank follow defined best practices or standards (e.g. ISBER 2018, ...)?
- ☐ Yes. Which one(s)?_____
- ☐ No

9. What is the purpose of your biobank?
- ☐ Backup storage for future use (e.g. captive breeding, propagation)
- ☐ Research: single-purpose (e.g. genomics)
- ☐ Research: multi-purpose (e.g. genomics, proteomics, transcriptomics, etc)
- ☐ Education
- ☐ Ex-situ biodiversity conservation
- ☐ Clinical (diagnostics & treatment)
- ☐ Commercial distribution

SPECIMEN COLLECTION

10. Do you follow routine collecting protocols?
- ☐ Yes
- ☐ No

11. What kind of sample / specimen is collected? (e.g., liver tissue, leaves, sediment, fibroblasts, micro-RNA…)

12. What kind of methods/fluids are routinely used for DNA/Tissue preservation during collection? (Check all that apply)
- ☐ Pure freezing
- ☐ Ethanol
- ☐ Desiccants (Silica gel, DriertiteTM)
- ☐ Stabilization reagent kits (i.e. Qiagen)
- ☐ Chemical treatment. Which one?_____(i.e. buffers, FTA paper, formalin)
- ☐ Other, which one_____

13. How are samples labeled? (Check all that apply)
- ☐ Unique biobank catalogue number
- ☐ Collectors' number (e.g. field number)
- ☐ Accession number
- ☐ Linear barcode
- ☐ 2D barcode
- ☐ Handwriting
- ☐ Self-adhesive cryolabel
- ☐ Other_____

PRESERVATION METHODS

14. Are samples transferred to new vials for final storage?
- ☐ Yes
- ☐ No
- ☐ Sometimes. Why?_____

15. Do you remove preservation fluids before storage?

- ☐ Yes. Why?_____
- ☐ No. Why?_____
- ☐ Sometimes. Why?_____

16. To minimize the amount of freeze/thaw cycles, do you: (Check all that apply)

- ☐ Extract DNA and/or other compounds before storage
- ☐ Make aliquot samples and keep them frozen
- ☐ Make aliquot samples of DNA and keep some of them in the fridge
- ☐ Extract DNA only after storage (i.e. when a loan has been granted)
- ☐ Other _____

17. What kind of DNA quality control is performed at the biobank? (Please specify which ones are done routinely (R) and which ones upon request (U))

- ☐ None
- ☐ Gel electrophoresis
- ☐ Reverse transcription PCR
- ☐ Real Time-PCR
- ☐ Spectrophotometry (NanoDrop, ...)
- ☐ Fluorometer (Qubit, Quantus, ...)
- ☐ Microfluidic technology or capillary electrophoresis (Bioanalyzer, Fragment Analyzer, ...)
- ☐ Northern blot analysis

18. When dealing with insufficient or rare material, do you produce appropriate quantities of DNA by: (Check all that apply)

- ☐ PCR
- ☐ Whole genome amplification (WGA)
- ☐ Cloning
- ☐ Cell Culture
- ☐ None of them
- ☐ Other _____

19. Do you have protocols to verify living material viability?

- ☐ Yes. How often do you test for this viability? _____
- ☐ No
- ☐ Doesn't apply

20. ***Only for seed banks*****:** How do you control for humidity? what are the levels of relative humidity used at your repository to store seeds?

SAMPLE RETRIEVAL, MATERIAL REQUESTS, AND SHIPMENT

21. What kind of sample size is considered for sample requests?

- ☐ The size of a rice grain for tissue samples
- ☐ Depending on DNA/tissue quality and quantity. Please type the amount_____
- ☐ As requested by the researcher
- ☐ Other _____

22. Has the demand for biobank samples increased over time? Please explain

23. Do you have policies and standards to request samples from your biobank?

- ☐ Yes
- ☐ No

24. How do you ship samples? (Check all that apply)

- ☐ In fixative/buffer at ambient temperature
- ☐ Dried at ambient temperature
- ☐ In insulated container with cool packs
- ☐ In insulated container, on dry ice
- ☐ Dry/vapor shipper
- ☐ DMSO
- ☐ Other _____

EMERGENCY PLANS

25. Do you have disaster/emergency plans in place (e.g., power failure, fire, etc.)? If so, Which ones?

SAFETY AND SECURITY

26. Are both cap and vial labeled per sample?

- ☐ Yes
- ☐ No

27. How are instruments sterilized/decontaminated during sample transfer when processing a batch of samples?

- ☐ Heat/flame
- ☐ Use new material per sample
- ☐ Alcohol
- ☐ Bleach
- ☐ Hydrogen peroxide
- ☐ Other_____

28. What is the process to confirm correct species identification?

29. Do you record the custody and handling of samples from collection to loans/disposal?

- ☐ Yes
- ☐ No

30. Do your protocols contain certain workflows that cannot be performed by students, interns, volunteers? If so, which?

31. If requested samples are returned, do you

- ☐ Discard the material
- ☐ Keep the material in a special area, separate from the original sample
- ☐ Re-integrate the material and keep together with all other samples
- ☐ Other_____

32. Do you follow any biological and chemical hygiene plans? Please describe how material and chemical reagents are discarded (biohazard waste bag, incineration, regular waste…)

33. Training is provided to biobank staff for:
- ☐ Handling of liquid nitrogen
- ☐ Biosecurity
- ☐ Biohazards
- ☐ Zoonotic pathogens
- ☐ Toxic substances
- ☐ Electrical equipment and/or monitoring systems
- ☐ Databases
- ☐ Policies and ethical issues
- ☐ Principles of care and maintenance

DATA STANDARDS

34. Do you have a database that can be accessed over the Internet?
- ☐ Yes
- ☐ No, but an internal database is used (i.e. FileMaker)
- ☐ Excel sheets are used
- ☐ Other_____

35. Are collection and permits (e.g. export/import) collected in the database? If not, where are permits stored?
- ☐ Always
- ☐ Never. Where else?_____
- ☐ Sometimes. Where else?_____

36. Does the biobank offer (to collectors) electronic data templates associated with vials/samples?
- ☐ Always
- ☐ Never

37. Are all uses of preserved samples tracked as transactions (loans/gifts) in the database?
- ☐ Always
- ☐ Never. Why?_____
- ☐ Sometimes. Why?_____

38. Are results and publications derived from samples tracked in the database?
- ☐ Always
- ☐ Never
- ☐ Sometimes

39. Is your sample database linked to a LIMS (laboratory information management system)? Please add a description of stored data

POLICIES

40. What are the acquisition policies at your biobank? (Specific criteria for receiving samples in the biobank)

41. ***Only for animal/plant repositories***: Are tissue samples accessioned into the collection accompanied by a specimen voucher?
- ☐ Yes, there must be always a backup voucher

☐ Yes, but pictures of the specimens are also accepted when there is no voucher available ('e-vouchers')
☐ Sometimes, depending on how rare/threatened the species are.
☐ No. Why?_____

42. Is the database updated when there are changes made to voucher specimens?
☐ Yes, since there is a shared database
☐ Yes, both databases are connected
☐ Sometimes. Why? _____ (i.e. has to be done manually, changes are suddenly found)
○ d. Never. Why?_____

43. Is there a written policy on sample use? (categories to restrict or allow the use of the samples)

44. Have your ABS policies undergone drastic change after the introduction of the Nagoya Protocol?
☐ Yes
☐ No
☐ The Nagoya protocol has not been implemented/ratified in my country

45. Any other relevant information regarding standards or practices in your biobank that was not examined in this survey?

YOUR CONSENT

Would you like your institution to be listed on the biobanks handbook for more visibility?
☐ Yes
☐ No

Have you heard of the Global Genome Biodiversity Network (GGBN), a consortium of biodiversity biobanks, before? If so, is your institution already a member of GGBN?
☐ Yes. My institution is already a member
☐ Yes, but my institution is not yet a member
☐ No

Would you like to receive info on GGBN (once)?
☐ Yes
☐ No

Would you like to receive regular updates on GGBN developments (to the email specified above)?
☐ Yes
☐ No

Would you like to be updated on activities concerning this questionnaire?
☐ Yes
☐ No

Annex 2. List of institutions that participated in the survey

Institution	Country	Type institution	Type biobank/collection
Australian Frozen Zoo	Australia	University	Livestock/veterinary, Aquarium/Zoo
CSIRO National Research Collections	Australia	Public Institution	Seed bank/crop, Living plant collection, Parasitology, Plant culture collection, Animal culture collection, Microbial culture collection, Natural history collection, Wildlife and wood forensics
Haus des Meeres	Austria	Aquarium/Zoo	Aquarium/Zoo
NHM Vienna (Anthropology Unit)	Austria	NHM	Natural history collection, human remains
NHM Vienna (Hymenoptera Unit)	Austria	NHM	Natural history collection
NHM Vienna (Laboratory of Molecular Systematics)	Austria	NHM	Natural history collection, DNA and tissue bank
NHM Vienna (Herpetology Unit)	Austria	NHM	Natural history collection
NHM Vienna (Evolutionary Biology Unit)	Austria	NHM	Natural history collection
NHM Vienna (Crustacea Unit)	Austria	NHM	Natural history collection
Naturhistorisches Museum Wien (Mammalogy Unit)	Austria	NHM	Natural history collection
University of Vienna (Archaeozoology Unit)	Austria	NHM	Natural history collection, osteological remains
INBO	Belgium	Public Institution	DNA-collections
Meise Botanic Garden (Molecular Lab)	Belgium	Botanical Garden/Arboretum	DNA-collections
Meise Botanic Garden (Seed Bank)	Belgium	Botanical Garden/Arboretum	Seed bank/crop
Royal Belgian Institute of Natural Sciences	Belgium	NHM	animal tissue and DNA
University of Zagreb, Faculty of Science	Croatia	University	University
National Museum in Prague	Czechia	NHM, Herbarium	Natural history collection
Finnish Museum of Natural History Luomus (DNA laboratory)	Finland	NHM	Natural history collection
Finnish Museum of Natural History Luomus (Botany unit)	Finland	Herbarium, Botanical Garden/Arboretum, Seed Bank	Seed bank/crop
University of Helsinki	Finland	University	Microbial culture collection
Andra	France	Public institution	Environmental specimen bank (ESB)

French Nationale Cryobank (Cryobanque Nationale)	France	Private/public national genebank	Domestic animals
Botanic Garden and Botanical Museum Berlin	Germany	NHM, Herbarium, Botanical Garden/Arboretum, Seed Bank, University	Seed bank/crop, Living plant collection, Microbial culture collection, Environmental specimen bank (ESB), Natural history collection
Fraunhofer IME, Schmallenberg	Germany	Contract Research Institute	Environmental specimen bank (ESB)
State Museum of Natural History Stuttgart	Germany	NHM	Natural history collection
Zoological Research Museum A. Koenig	Germany	NHM	Animal culture collection, Environmental specimen bank (ESB), Natural history collection
Hungarian Natural History Museum	Hungary	NHM, Herbarium	Parasitology, Natural history collection
MTA-DE "Lendület" Evolutionary Phylogenomics Research Group (a research group of the Hungarian Academy of Sciences based at the University of Debrecen)	Hungary	University	Natural history collection, DNA-bank
Israel Gene Bank, Agricultural Research Organization Volcani center	Israel	Public Organisation	Seed bank/crop, Living plant collection, Plant culture collection, Environmental specimen bank (ESB)
The Hebrew University	Israel	University	Livestock/veterinary, Living plant collection, Parasitology, Natural history collection, Wildlife and wood forensics
European Association of Zoos and Aquaria	Netherlands	Zoo/Aquarium membership organization	Aquarium/Zoo
University of Ibadan	Nigeria	University	Aquarium/Zoo
Natural History Museum, University of Oslo	Norway	NHM	Natural history collection, Wildlife and wood forensics
Norwegian Institute for Water Research (NIVA)	Norway	Private institute/laboratory	Environmental Specimen Bank
The Kostrzyca Forest Gene Bank (Poland)	Poland	Herbarium, Botanical Garden/Arboretum, Seed Bank	Seed bank/crop, Living plant collection, Plant culture collection, Animal culture collection
Plant Gene Bank, Directorate for National Reference Laboratories, Ministry of Agriculture, Forestry and Water Management, Republic of Serbia	Serbia		Seed Bank
South African National Biodiversity Institute	South Africa	Aquarium/Zoo, Biodiversity	Aquarium/Zoo, Parasitology, Animal culture collection,

		Biobank	Wildlife and wood forensics, Also Veterinary but not livestock
IRIAF-CERSYRA de Valdepeñas (Regional Institute for Research and Development in Agri-food and Forestry in Castilla-La Mancha -- Regional Center for Animal Selection and Reproduction	Spain	Public Research Institute	Livestock/veterinary
SERIDA	Spain	Public Institution	Livestock/veterinary
Banco Nacional de Germoplasma Animal (BNGA)- MAPA-IMIDRA	Spain	Public Institution	Livestock/veterinary
Gothenburg University	Sweden	University	Microbial culture collection
Swedish Museum of Natural History (NRM)	Sweden	NHM	Environmental specimen bank (ESB)
NHMLondon	UK	NHM	Livestock/veterinary, Aquarium/Zoo, Parasitology, Animal culture collection, Environmental specimen bank (ESB), Natural history collection, Wildlife and wood forensics, Plant Material
Royal Botanic Garden Edinburgh (Cryptogamic Section)	UK	Herbarium	Natural history collection
Royal Botanic Garden Edinburgh	UK	Herbarium	Natural history collection
Royal Botanic Garden Edinburgh	UK	Botanical Garden/Arboretum	Living plant collection, Natural history collection
Royal Botanic Gardens Kew	UK	Seed Bank	Seed Bank/wild species
M.V. Zubets Institute of Animal Breeding and Genetics	Ukraine	Public Institution	Farm animal genetic resources bank
Biodiversity Institute, University of Kansas	USA	Natural History Museum	Natural history collection
Missouri Botanical Garden	USA	Herbarium	Natural history collection
Museum of Comparative Zoology, Harvard University	USA	Natural History Museum	Natural history collection
National Tropical Botanical Garden	USA	Botanical Garden/Arboretum	Seed bank
North Carolina Museum of Natural Sciences	USA	Natural History Museum	Natural history collection
San Diego Zoo Global	USA	Aquarium/Zoo	Animal cell culture collection
University of Alaska Museum	USA	Natural History Museum, University	Natural history collection

ANNEX 3. Institutions that were personally contacted.

- Manaaki Whenua Landcare Research
- Ocean genome legacy
- Frozen Tissue Collection at the Australian Centre for Wildlife Genomics
- BIOTECH Philippine National Collection of Microorganisms
- Culture Collection of Cryophilic Algae CCCryo, Fraunhofer Institute for Cell Therapy and Immunology at its Branch for Bioanalytics and Bioprocesses (IZI-BB)
- Archaeological animal DNA facility (ISRaDNA)
- National Animal Germplasm Program, USDA
- Manter Lab of parasitology
- IVB Genetic Bank
- Steinhardt museum (Fungi section)
- Fundação Oswaldo Cruz (Protozoa section)
- University of Kansas (Ichthyology section)
- Denver Botanic Gardens
- Rio de Janeiro Botanical Garden
- Institute of Vertebrates Biology
- Intertryp / Cirad
- GABI / Inrae
- CIRM-CFBP
- Institut Pasteur (microorganisms' section)
- Pacific Center for Molecular Biodiversity
- Museum of Zoology, Pontificia Universidad Católica del Ecuador
- National Biodiversity Cryobank of Canada
- Yukon palaeontology program
- Species 360