Genetic Resource Collections Associated with Natural History Museums: A Survey and Analysis to Establish a Benchmark of Standards¹

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ABSTRACT

In today's modern natural history museum, genetic resources are a standard and integral part of traditional collections. These collections require significant time and resources to amass and specialized training to build and maintain, and, unlike traditional museum specimens, their repeated use eventually results in their complete consumption. Although standards exist for the management of traditional natural history collections, there are few guidelines available for the documentation, arrangement, and housing of this new type of museum research collection. To establish a benchmark of current practices for collecting and storing genetic resources, we distributed an online survey that included 57 questions to 45 independent collections at 39 different institutions in nine different countries. Survey results revealed that procedures varied widely among collections with samples being preserved, processed, stored and distributed using a number of different methods. Variances in practice were a function of institutional differences, including size, budget, personnel and storage locations. The vast majority of surveyed collections had written internal policies in relation to genetic resources; however, it was questionable whether internal guidelines were adequate given that published resources were not used to inform the majority of policies. In addition, published guidelines did not address some of the unique historical and practical issues pertinent to natural history collections, including tracking data associated with both voucher specimens and their associated genetic samples, and processing loans/gifts. Ultimately, we hope these findings establish a starting point that initiates a broader discussion regarding the standardization of curation for genetic resources to preserve their integrity for long-term use.

KEY WORDS: Best Practices, cryopreservation, DNA, genetic, natural history collections, RNA, tissues

Historically, natural history collections and other repositories of preserved biological material have focused on the preservation of whole-organism specimens. During the past decade, genetic resources have become a standard and integral part of these traditional collections. These diverse genomic collections most often include frozen tissues, chemically preserved tissues, and/or associated extracts

and, unlike typical museum specimens, genetic resources are consumptive. These types of collections are also costly and time-consuming to amass and require specialized training to build and maintain; genetic resources must be carefully collected, transported, stored and monitored in a low temperature environment if they are to remain useful for molecular analyses. These are just some of the numerous rea-

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sons why genetic resources present a number of unique management problems for natural history collections. In addition, despite more standardized methods for the management of traditional natural history collections, there seemingly is no best-practice standard for the documentation, arrangement, and housing for this new type of museum research collection.

10

To learn more about the current protocols, challenges and considerations associated with genetic resources, we surveyed natural history museums and other collections with genetic samples. By surveying a representative sample of collections and developing an overall view of curation procedures, this analysis addresses the following major objectives: 1) to document the current state of curation procedures across different institution types; 2) to establish a benchmark of current practices and define the advantages and disadvantages of the varying standards; 3) to examine the practices and determine the feasibility for standardizing some or all of the curation procedures in the future, helping move collections towards a uniform best-practice standard; and 4) to compare the curation procedures for genetic resources with procedures in other natural history disciplines, as a means to standardize the curation of preparation types and bridge the gaps among different collections.

RESEARCH METHODS AND SURVEY PROCESS

A list of potential genetic resource collections to survey was compiled using the Registry of Biological Repositories (http://www. biorepositories.org), appendices included in Prendini et al. (2002) and additional online research of established public and private genetic resource collections. After a list of museum collections and their contacts was finalized, introductory letters were sent via email to request participation in an online survey hosted by the iCommon Poll Tool of Harvard University. In addition to sending direct requests for participation, a link to the online poll was advertised twice in the Society for the Preservation of Natural History Collection (SPNHC) newsletter. Follow-up letters were sent via email to remind those who had not completed the survey after 60 days. Online surveys were conducted between January and May 2012. A listing of the collections and their associated institutions that completed the survey is given in Table 1. Due to the fact that institutions may have genomic collections housed in separate locations, multiple respondents completed the survey for some institutions. It was verified that those institutions with multiple respondents completing the survey represented different collections or departments so as to not duplicate answers.

The online survey included 57 questions, which consisted of both multiple-choice and short answer (Appendix 1). Most multiplechoice questions allowed users to select more than one answer choice by asking respondents to check all answers that applied; therefore, the overall total for a given question frequently did not add up to 100%, rather each individual answer choice within a specific question would total 100%. In addition, most multiple-choice questions allowed respondents to writein responses using the "Other" selection if the given multiple-choice answers were not adequate.

The results of the survey were divided into five separate areas of curation that included: 1) general collection and institution information (institution type, budget, holdings, storage locations, personnel, policy and guidelines for curation and management, facility functions, equipment and genetic sample types); 2) processing of samples (initial collection, sample transfer and final storage); 3) sample labeling, tracking and data; 4) security and safety (security of collections, personnel

safety measures and back-up precautions for equipment); and 5) use of collections (loan/ gift frequency and policies, processing and shipment of loans/gifts, and requirements of researchers). The results from each of these areas are presented independently.

RESULTS GENERAL COLLECTION AND INSTITUTION INFORMATION

Institution type and budget

The first six survey questions collected information regarding the respondents and their institutions (Questions 1 through 6; Appendix 1). Respondents from 45 independent collections within 39 different institutions completed the online survey, representing nine countries: Australia, Brazil, Canada, Germany, Ecuador, Norway, Spain, U.K. and U.S. (Table 1). Three institutions (Denver Museum of Science and Nature, Kansas University Biodiversity Institute and Royal Ontario Museum) had respondents from two different collections or departments completing the survey, and a single institution (Department of Biology and the M. L. Bean Life Science Museum, Brigham Young University) was represented by four collections. Many institutional factors varied considerably among the genetic resource collections surveyed, including the institution type and size, as well as the degree of organization and infrastructure of these collections. The majority of the surveyed institutions were self-categorized as natural history museums (69%), and additional institution types included universities/colleges (53%), botanical gardens/herbaria (13%), science museums/science centers (11%) and seed/spore banks (2%). The "Other" category was used by 9% of respondents to describe their institution. Many respondents (44%) selected multiple categories to describe their

institution, and a large percentage of this group (31%) were natural history museums associated with a university or college. No participating institutions chose any of the following four offered choices to describe their institution: cell line center, culture or stock center, private institute/laboratory or zoo.

Respondents were asked to provide an annual budget for the curation of their genetic resources (Question 45; Appendix 1). Approximately one-half of the collections surveyed (51%) provided an approximate budget. Annual budgets for these 23 genetic resource collections were highly variable, ranging from \$0 to \$300,000 (mean \$29,227; median \$5,500) with the majority (61%) being less than \$10,000. The survey did not ask respondents to specify employees' salaries in addition to general operating costs, but it was clear that some estimates included salaries, while other estimates did not. For some collections surveyed (20%), the cost of curating genetic samples was included in the overall budget for the institution or department, and there was no separate budget for the curation of genetic resources. Some collections could not provide a budget for the curation of genetic resources (16%), and a few collections (7%) reported that the funds used to pay for the curation of genetic samples came from personal research budgets.

Holdings

The total number of genetic samples present in the collections surveyed, as well as the taxonomic scope of these collections, varied widely among the 45 independent collections within the 39 different institutions surveyed (Questions 9 and 10; Appendix 1). The holdings within the collections surveyed ranged from 250 to 2,000,000 samples (mean 98,866; median 30,000; Table 1). The majority of collections surveyed (62%) included approximately 15,000 to 150,000 samples.

The total number of voucher specimens was used as an estimate of the total number of genetic samples in some cases (16%) because these respondents did not know the total number of genetic samples present in their collections. Many of the respondents that used the total number of voucher specimens as an estimate of the total number of genetic samples noted that there were often multiple genetic samples associated with a single voucher specimen, so their total holdings were likely underestimated. The taxonomic groups represented in those collections also varied widely with the vast majority of collections surveyed (89%) including multiple taxonomic groups at the Class level or above. For example, some large collections within institutions with a regional focus had representatives from many taxonomic groups, whereas other collections focused on a single taxonomic group.

Storage locations

12

To assess the degree of organization of genetic resource collections within an institution, the survey asked respondents whether their respective institutions had formed a centralized repository for genetic resources and whether those centralized repositories were fully functional (Questions 14 and 18; Appendix 1). The majority of institutions (58%) designated that they had a centralized repository for genetic resources (Table 1). To characterize the functionality of the centralized repositories, respondents were asked to describe the state of their facility's infrastructure (i.e., building/remodeling or outfitting the repository with equipment) and the status of transferring genetic samples to their centralized storage locations. The majority of those institutions with centralized facilities (62%) reported that infrastructure was complete and samples resided in permanent storage locations. More than one-quarter of the centralized facilities (27%) stated that infrastructure was not yet complete, and a minority (12%) had completed infrastructure projects but were in the process of moving samples to the facility's permanent storage. In addition to determining whether centralized repositories were fully functional, the survey inquired if any outside funding was received to help build infrastructure or facilitate projects, such as sample labeling (Question 46; Appendix 1). More than one-half (53%) of centralized repositories reported that they had received outside funding either to assist in building a genetic resources facility or to finance projects involving the organization and/or labeling of genetic samples.

Three questions within the survey were focused on learning about the collections present at institutions without centralized repositories (Questions 15, 16 and 17; Appendix 1). Those institutions without centralized repositories (42%) reported that their collections were stored in 1 to 12 different locations, and the methods of physical storage of genetic samples within these institutions (without centralized repositories) was highly variable (Table 1). For example, respondents at institutions without centralized repositories reported that samples were stored in a single room or freezer but managed individually by discipline, stored in multiple rooms within the same building, or stored within different buildings. This information, unfortunately, could not be clearly quantified because the respondents gave these examples as part of their answer to the survey question regarding the number of separate physical locations (i.e., laboratories or rooms) where genetic resources were stored (Question 15, Appendix 1). The collections that reported that their institutions did not have centralized repositories (42%) cited a number of reasons why samples remained in separate collections or laboratories, including lack of space (44%), lack of funding (35%), and lack of personnel

(30%). Only 4% of those respondents cited a paucity of samples as a reason why their institution did not have a centralized facility. Three additional reasons why centralized repositories were not built at these institutions were cited under "Other" (19%), including accessibility of samples for researchers or those working in distributed collections (13%), ownership or control of samples (4%) and lack of equipment (2%). More than one-third (39%) of the collections surveyed without centralized repositories (42%) noted that their institution had plans to build a centralized repository within 1 to 3 years (22%) or in more than 3 years (17%).

Personnel

Two questions within the survey were focused on those personnel working with genetic resources (Questions 11 and 19; Appendix 1). The majority of collections had curators/professors with higher degrees (80%) or collection managers with higher degrees (51%) maintaining collections. In addition, other personnel included paid student assistants (31%), collection managers without higher degrees (27%), staff or technical assistants (22%), unpaid student assistants (16%) and volunteers (7%). The majority of collections (77%) responded that 2 to 6 different categories of personnel worked with genetic resource collections, but the survey results could not determine the extent of work completed by the various types of personnel. For those institutions with centralized repositories (58%), curators/professors with higher degrees (50%) and collection managers with higher degrese (43%) most often maintained the genetic resource collections. Additional workers included paid or unpaid student assistants (34%), staff or technical assistants (27%), collection managers without higher degrees (21%) and volunteers (16%).

Policy and guidelines for curation and management

One question within the survey was focused on the personnel determining the policy that concerned genetic resources (Question 13; Appendix 1). Among the 45 independent collections surveyed, policies regarding genetic resources were most often determined by curators/professors (84%), but governing boards (30%) or collection managers (34%) were also often involved in writing policy. In 5% of those collections surveyed, collections managers without higher degrees determined policy involving genetic resources. Using respondents' answers from an additional question (Question 14; Appendix 1), the survey results determined that policy for centralized repositories (58%) was most often determined by curators/ professors (88%), but governing boards (46%) and collection managers (39%) were also involved in determining policies within these collections.

The survey included a single question that assessed the guidelines used for the curation and management of genetic resource collections (Question 12; Appendix 1). The vast majority of genetic resource collections (78%) had written internal guidelines to inform those working with samples on the correct storage and use of samples, while 18% of those surveyed did not use any internal written guidelines. For the collections with written department or institutional guidelines (78%), the majority (52%) responded that only internal guidelines were used in the curation or management of genetic resources, while less than one-half (48%) used at least one of the four published guidelines presented in the survey (Question 12; Appendix 1). The publication most often used (20%) was the Best Practices for Repositories published by the International Society for Biological

and Environmental Repositories (ISBER, 2012). Fewer collections used Prendini et al. (2002; 7%), the National Cancer Institute Best Practices for Biospecimen Resources (NCI et al., 2011; 4%), or the Organisation for Economic Cooperation and Development Best Practice Guidelines for Biological Resource Centres (OECD, 2007; 2%).

Facility functions and equipment

14

Two separate questions within the survey assessed the main functions of centralized repositories for genetic resources and determined the equipment available within them (Questions 20 and 21; Appendix 1). The main functions of the 26 institutions surveyed with centralized repositories (58%) involved sample storage/tracking (67%) and tissue sub-sampling and/or processing loans/gifts (61%). In addition, some of the centralized repositories performed additional genetic laboratory functions, including DNA/RNA extraction (16%), PCR (16%), and sequencing (14%). All the centralized repositories performing these additional functions (except one), performed all three additional laboratory functions; whereas, the single exception completed DNA/RNA extractions and PCR (but not sequencing) within their facility. The various types of cold storage equipment used in these centralized collections are discussed in the *Processing of samples: Final storage* section. Additional equipment present within these centralized repositories included biosafety cabinets (28%), fume hoods (23%), centrifuges (19%), gel electrophoresis equipment (19%) and vortexes (17%).

Sample types

Collection representatives were asked about the types of genetic resources stored in their repositories (Question 7; Appendix 1). The majority of collections stored frozen tissues (91%), DNA samples (76%), PCR products (53%) and chemically preserved tissues (42%; Fig. 1). Many also stored RNA (24%), while a minority preserved proteins (11%), cell cultures (9%), cell lines (9%), antisera (7%), and bacterial artificial chromosomes (BACs; 4%). The "Other" category (6%) consisted of single collections storing one of the following

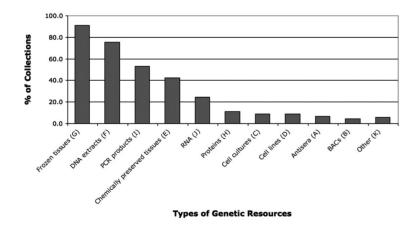


Figure 1. Types of genetic resources stored in 45 independent collections at 39 different institutions, represented in descending order of percentage except for "Other" category (shown last). For reference to the survey, the letter choice of each answer in Question 7 (Appendix 1) is shown in parentheses after the selection. Responses in the "Other (K)" category included bacterial isolates, dried scales, and silica gel dried plant tissue.

sample types: bacteria isolates (2%), dried scales (2%), and silica gel dried plant tissue (2%). Details regarding how samples were initially preserved (flash-frozen or using various preservatives) by collections surveyed are recorded in the *Processing of samples: Initial collection and storage* section.

PROCESSING OF SAMPLES

Initial collection and storage

The survey determined the origins of genetic resources and the methods used during the initial collection of genetic samples (Questions 8 and 22; Appendix 1). Genetic samples within the 45 collections surveyed originated from in-house researchers (93%) and sources outside their institutions, including gifts and/or donations (80%) and affiliates of their institutions (76%). To characterize the methods used during the initial collection of genetic resources, collection representatives were asked to select the processes used when first preserving samples. A number of different methods were used by the 45 collections surveyed during initial preservation of

genetic samples (Fig. 2). Most collections (80%) stored samples that were initially preserved in 2 to 5 different ways. The vast majority of those surveyed indicated that their collections included samples preserved with ³ 95% ethanol (90%) or those that were flashfrozen (69%). Respondents also reported their collections included samples initially preserved in dimethylsulfoxide (DMSO; 38%) or RNAlater® (registered trademark of Ambion; 33%). Few collections (7%) used Allprotect Tissue Reagent (Qiagen) as an initial preservative. A number of respondents reported that they used additional methods of preservation using the "Other" category (11%; Fig. 2), including lysis buffers (5%), buccal swabs (2%), silica gel dessicant (2%), and FTA paper® (2%). Of the two respondents using lysis buffer, one reported that Queen's lysis buffer was used, while the other did not specify the type of lysis buffer.

Two questions within the survey extracted additional details about the initial storage of genetic samples, including the types of vials used and the methods used for labeling those vials (Questions 23 and 24; Appendix 1). The

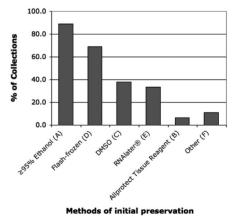


Figure 2. Methods and preservatives used in the initial preservation of genetic samples in 45 independent collections at 39 different institutions, represented in descending order of percentage except for "Other" category (shown last). For reference to the survey, the letter choice of each answer in Question 22 (Appendix 1) is shown in parentheses after the selection. Responses in the "Other (F)" category included lysis buffers, buccal swabs, silica gel dessicants, and FTA paper".

vials used during initial collection included four separate types, which were categorized by the location of the threads (internally or externally) on the vial opening, and the presence or absence of gaskets within the vial caps (Fig. 3). Many collections reported that they used externally threaded vials with gaskets (39%), internally threaded vials with gaskets (36%) and externally threaded vials without gaskets (34%). Internally threaded vials without gaskets were less common (23%). The majority of collections (69%) reported that multiple types of vials were used for initial

storage with approximately one-third (32%) of collections accepting any type of vial. Vials were most often initially labeled with a collector/field number (64%) or institution catalog number (53%). Barcodes were also used to label vials during initial collection but in far fewer cases (20%). A number of collections used the "Other" category (16%) to describe a pre-determined and dedicated series of numbers used to label genetic samples, and a single collection reported that they included taxonomic information on the vials.

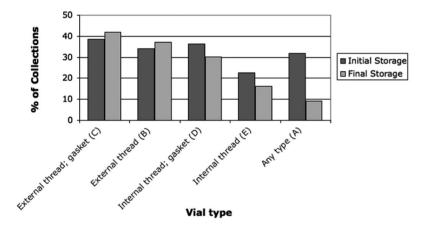


Figure 3. Vials used during initial collection (dark grey) and final storage (light grey) of genetic samples in 45 independent collections at 39 different institutions, represented in descending order of frequency of use in final storage. Variation in the types of vials included location of the threads internally or externally on the vial opening, and vials caps with or without gaskets. For reference to the survey, the letter choice of each answer in Questions 23 and 27 (answer choices identical; Appendix 1) is shown in parentheses after the selection.

Sample transfer

Respondents were asked about curation of genetic resources before their final deposition in the collection, including the transfer of samples into new vials and the removal of liquids used in the preservation process (Questions 25 and 38; Appendix 1). The majority of collections surveyed (64%) reported that select samples were transferred into new vials before final storage. In a fewer cases (9%), vials were always transferred, regardless of the type of vial used in initial collection. Some

collections (27%) did not transfer samples into new vials, simply depositing samples into the collection as received. Of those collections surveyed that either always or selectively transferred samples into new vials (73%), respondents cited a number of reasons why these select vials were transferred: if the vial was not the proper size for the final storage container (35%), if the vial was not rated for ultra-cold temperatures (35%), if the vial was compromised (cracked, broken, or the seal had failed; 17%), if labeling on the vial was

difficult to read (7%), or if the vial was not the preferred type of the collection (7%). A separate question (Question 38; Appendix 1) determined whether collections using liquid nitrogen poured off liquids before deposition in the collection. The highest percentage of collections (42%) poured off preservatives and/or buffers on a case-by-case basis, while one-third (33%) never removed the original preservatives or buffers from samples, and onequarter (25%) always poured off liquids before deposition in the collection.

The survey asked respondents to select the methods of decontamination or sterilization used during the sample transfer process (Question 26; Appendix 1). A number of different decontamination or sterilization techniques were used by the collections surveyed to sterilize instruments (Fig. 4). The most common practices included ethanol (51%), heat/flame (44%) and bleach (33%). Those collections that reported that "Other" (16%) decontamination or sterilization methods were being utilized included the use of detergent/ soap with water (7%), autoclave (5%), UV light (2%) and water (2%). Approximately

one-quarter of collections surveyed (24%) used multiple methods of decontamination or sterilization. The survey could not determine if the type of decontamination or sterilization was correlated to a specific use; for example, if some methods were used to sterilize instruments while working with a sequence of independent samples, while other techniques were used before or after processing a batch of

Respondents were asked to characterize the vials used for final storage of their genetic samples (Questions 27 and 28; Appendix 1). Similar to the vials used for initial storage, vials used for final storage included four separate types, which were categorized by the location of the threads (internally or externally) on the vial opening, and presence or absence of a gasket within the vial caps (Fig. 3). Many collections surveyed reported that they used externally threaded vials with gaskets (42%), externally threaded vials without gaskets (37%), and internally threaded vials with gaskets (30%). Internally threaded vials without gaskets were less common than other vial types (16%). The majority of collections

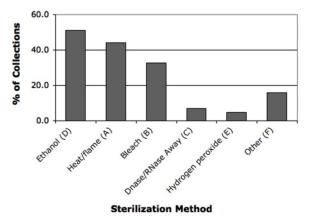


Figure 4. Methods of sterilization used during the sample transfer process in 41 independent collections at 39 different institutions, represented in descending order of final storage in collection except for "Other" category (shown last). For reference to the survey, the letter choice of each answer in Question 26 (Appendix 1) is shown in parentheses after the selection. The percentage of alcohol in the "Ethanol (D)" answer was not included in the question. Responses in the "Other (F)" category included detergent/soap with water, autoclave, UV light and water.

(63%) reported that only a single vial type was used for final storage, while more than one-third (37%) accepted multiple types of vials for final storage. Only 9% of collections reported that they used all vial types for final storage.

Final storage

18

Respondents were asked several questions regarding how genetic samples were stored for long-term preservation, which included room temperature and cold storage (Questions 37, 39 and 40; Appendix 1). Temperature ranges were provided in the survey (Question 37; Appendix 1) to clarify the cold storage types: general-purpose or laboratory freezers (-12°C to -30°C), ultracold freezers (approximately -50°C to -86°C) and standard liquid nitrogen cryovats (below -110°C). The most commonly used type of cold storage within those collections surveyed was ultracold freezers (78%; Fig. 5; Table 1). Many collections used liquid nitrogen cryovats (27%, which

included both standard (25%) and isothermal (2%) cryovats) or general purpose and/or laboratory freezers (24%). Fewer collections (13%) stored samples at room temperature, while the "Other" category (4%) consisted of refrigeration at 4°C. Within those institutions with centralized repositories (58%; Question 14; Appendix 1), the majority of collections included ultracold freezers (61%), while fewer collections included general-purpose or laboratory freezers (37%) and standard liquid nitrogen cryovats (26%). For those repositories that used liquid nitrogen in either standard or isothermal cryovats (27%; Question 37; Appendix 1), most (58%) had liquid nitrogen delivered and stored in dewars that were attached to the liquid nitrogen cryovats. One-third of these collections (33%) had liquid nitrogen delivered and stored in bulk tanks. Lastly, a single collection (8%) reported that they produced their own liquid nitrogen on site.

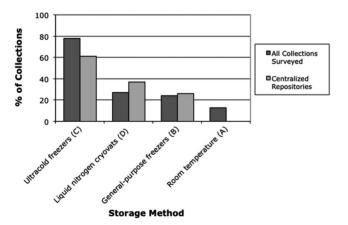


Figure 5. Storage methods used by genetic resource collections in 45 independent collections at 39 different institutions represented in decreasing percentage of use for all collections surveyed (dark grey). Results for centralized repositories are also shown (light grey). Liquid nitrogen cryovats (D) included both standard (25%) and isothermal (2%) models. For reference to the survey, the letter choice of each answer in Question 37 (Appendix 1) is shown in parentheses after the selection. Responses to Question 14 (Appendix 1) were used to identify institutions with centralized repositories.

SAMPLE LABELING, TRACKING AND DATA

GENETIC RESOURCE COLLECTIONS SURVEY

Sample labeling

Collection representatives were asked to characterize the methods used to label genetic resources (Questions 29, 30 and 32; Appendix 1). The majority of collections (57%) wrote information by hand directly onto vials, which included both information written on the vial by the original collector and additional information written after receipt by the genetic resource collection. Fewer collections used self-printed labels that wrapped around vials (22%), paper tags placed into vials (7%) or laser-etched vials (5%). Barcodes were also a commonly used method of sample labeling and, in particular, pre-printed or self-printed barcode labels that wrapped completely around vials were most commonly used (32%). Vials that included directly-printed barcodes (vials manufactured with barcodes) on the sides or bottoms of vials were used less often (12%) than self-applied barcode labels. Self-applied barcodes placed onto vial caps were seldom used (7%). Two different types of barcodes were used by the 45 collections surveyed, including linear or onedimensional (1D) barcodes (54%) and twodimensional (2D) barcodes (46%). Of those collections surveyed that used 2D barcodes (46%), formats included both DataMatrix (75%) and QRCode (25%). Radio-frequency identification (RFID) tags were not used by any of the surveyed collections to label vials.

Sample tracking

To characterize the specimen retrieval process, respondents were asked how samples were organized within their collections (Questions 31 and 36; Appendix 1). The majority of collections surveyed used a rack and cell box system (82%), while a minority (18%) did not utilize this method of sample organization. Of the 82% of surveyed

collections that used a rack and cell box system, one-quarter (25%) additionally used a barcode system to track samples. A single collection used a rack and cell box system in conjunction with RFID tags attached to the racks and boxes, although, as previously mentioned, no collection used RFID to label sample vials. The majority of collections (60%) also reported that genetic samples were additionally organized by department or division (36%), taxonomic group (22%) or research project (2%).

Databasing genetic sample data

Collection representatives were asked about the methods used to track data associated with genetic resources (Questions 33 and 34; Appendix 1). The majority of collections (63%) directly associated genetic sample data with voucher specimen data, while one-third (33%) used independent databases to track genetic sample and voucher specimen data, respectively. Four different platforms were used to make sample data available online: noncommercial collection management software (44%), internally written applications (35%), web delivery through data aggregators (18%), and commercial collection management software (6%). Those collections that used non-commercial collection management software (44%) used one of two applications developed for museum and herbaria research data: Arctos (27%) and Specify (17%). Those collections that utilized internally written applications to make data available online (35%) included use of FileMaker Pro (17%), Microsoft Access (6%), internally designed spreadsheets (6%) and unspecified methods (6%) to track data. Those collections surveyed that used web delivery via data aggregators (18%) employed GBIF (6%), HerpNET (6%) or ORNIS (6%); two of the three collections using data aggregators reported that they tracked genetic data using internal

databases. The single collection using a commercial database management application (6%) purchased RURO FreezerPro, software developed for frozen sample management. A separate question (Question 35; Appendix 1) determined if taxonomic or other data changes associated with the original voucher specimen (from which the genetic resource was sampled) were also applied to the genetic specimen. The vast majority of collections (70%) always updated data associated with the genetic samples when changes were made to voucher specimens, while more than onequarter (26%) reported that sometimes these changes were made, and a minority (5%) never made these update changes.

SECURITY AND SAFETY

Access to collections

20

To characterize the security of genetic resources, collection representatives were asked about access to rooms and freezers where genetic resource collections were stored (Questions 41 and 42; Appendix 1). The majority of collections surveyed (64%) reported that samples were stored in locked rooms or facilities, and more than one-third (34%) were stored in locked freezer units. Almost one-third (30%) stored samples in a room or facility that remained unlocked during the day and, of those collections storing samples in rooms that remained unlocked, the majority (62%) did not store samples in locked freezer units. Only a single collection (2%) reported that samples were stored in an unlocked room or facility, although samples were stored in locked freezers. The majority of collections surveyed (64%) reported that only trained personnel had access to genetic resources, while more than one-third (37%) reported that all those with access to the room where the genetic resources were stored also had access to the samples. Only a single collection (2%) reported that all those who had access to the building had access to genetic resources.

The 45 genetic resource collections

ZIMKUS AND FORD

Back-up precautions

surveyed were asked to determine what backup supply systems and equipment monitoring were being used to reduce the possibility of sample loss (Question 43; Appendix 1). Using respondents' answers from two additional questions (Questions 14 and 37; Appendix 1), the survey also ascertained the back-up precautions for both centralized repositories and those collections using liquid nitrogen storage equipment. Audible or visual alarms were used in 60% of all surveyed collections; alarms were used in one-half of centralized repositories (50%) and slightly more than one-half of those collections storing samples in liquid nitrogen (55%; Fig. 6). Battery back-up systems were present in less than one-quarter of all collections surveyed (21%); battery backup systems were present in more than one-half of collections using liquid nitrogen (55%) and were less common within centralized repositories (15%). Uninterruptible power supplies were present in one-third of all collections surveyed (33%); this type of backup power was present in more than one-third of the centralized repositories (35%) and more than one-quarter of collections using liquid nitrogen (27%). External monitoring of equipment by outside sources, including university controls or external contracts, was used in the majority of collections surveyed (55%); outside monitoring of equipment was used in more than one-half of centralized repositories (54%) and less than one-half of collections using liquid nitrogen (46%).

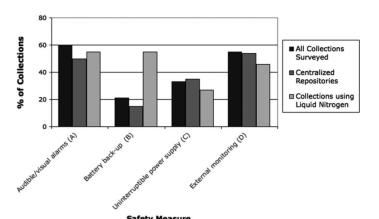


Figure 6. Safety measures used by genetic resource collections in 45 independent collections at 39 different institutions represented in increasing vigilance of back-up method or monitoring. Results are shown for all collections surveyed (dark grey), centralized repositories (medium grey) and collections using liquid nitrogen (light grey). For reference to the survey, the letter choice of each answer in Question 43 (Appendix 1) is shown in parentheses after the selection. Responses to Questions 14 and 37 (Appendix 1) were used to determine safety measures for institutions with centralized repositories and collections using liquid nitrogen, respectively.

Personnel safety

To characterize safety practices used in relation to liquid nitrogen equipment, representatives were asked about the presence of monitoring devices and specialized ventilation (Question 44; Appendix 1). For those collections surveyed that used liquid nitrogen in either standard or isothermal cryovats (27%), the majority (75%) reported that oxygen monitors were present in collection space to alert occupants of unsafe oxygen levels. Among these collections, twothirds (66%) also used specialized exhaust systems to maintain an adequate supply of air. Two collections (17%) did not have either oxygen monitors or specialized ventilation but noted that their equipment was present in a room with sufficient volume of air so that this was not a concern. A single collection (8%) had an oxygen monitor but no specialized ventilation and noted that their liquid nitrogen equipment was located near an exterior door that could be opened if oxygen levels were depleted.

GENETIC RESOURCE COLLECTIONS SURVEY

USE OF COLLECTIONS

Loan/gift frequency and policies

Surveyed collections were asked about whether their institutions had loan/gift programs that included genetic resources and were asked to estimate the average number of transactions processed annually (Questions 47 and 48; Appendix 1). The vast majority of the 45 collections surveyed (93%) had active loan/gift programs, sending genetic samples to researchers at other institutions. Only 7% of collections surveyed reported that loans/gifts of genetic material were not made to other institutions. The total number of loan/gift transactions processed annually was highly variable across those collections surveyed. When all respondents' data were accumulated, the range of loans/gifts processed was from zero to 2,000 loans with the majority of collections (84%) reporting that they processed 50 or fewer loan/gift transactions per year.

Respondents were also asked about their loan/gift policies and the details of their

approval process (Questions 49, 50 and 51; Appendix 1). Surveyed collections reported that varying types of information must be submitted in a genetic sample request, including a description of the proposed research (95%), number of samples requested (95%), qualifications of investigator (83%), scientific value of research (63%), feasibility of the project (59%) and evidence of sufficient lab facilities and funding (47%). Among the collections surveyed, incoming loan/gift requests were received by the departments/ collections where samples originated (54%), individual researchers (44%) or centralized genetic resource collections (29%). The survey revealed that those who ultimately approved the genetic sample request often differed from those to whom the incoming request was sent. Genetic sample requests were approved by the departments/collections where samples originated (51%), centralized genetic resource collections (37%) and individual researchers (27%).

Processing and shipment of loans/gifts

The survey asked respondents to describe the amount or size of genetic samples normally processed for loans/gifts (Question 52; Appendix 1). None of the collections surveyed sent the entire aliquot or sample, but rather sent only a sub-sample; however, a single collection (2%) reported that they often provided the complete voucher specimen with a specific destructive sampling policy. One-quarter of collections surveyed (25%) provided sub-samples that were the approximate amount needed for two DNA extractions, while fewer (9%) sent the amount needed for three DNA extractions. Some collections surveyed quantified the amount sent using cubic millimeters (14%), which ranged from 1 to 6 mm³, while others gave an approximate weight (7%), which ranged from 10 mg to 2 grams. Some collections surveyed

(7%) compared the average size of their tissue sub-samples to that of a grain of rice. A single collection (2%) reported aliquots of lysis buffer (50 ml) were sent in addition to tissue samples. Some collections surveyed (7%) reported that there was no standard amount provided, rather the amount sent was specific to the request or dependent on the amount of a given sample present in the collection. A small percentage of collections (5%) sent the specific amount or volume requested and, thus, could not report an average amount since it varied with each loan/gift request.

Respondents were asked about how loans/gifts of genetic material were shipped, including how shipments were made and if those receiving genetic samples were asked to offset the costs associated with processing and shipping (Questions 53 and 54; Appendix 1). The majority of surveyed collections shipped samples at ambient temperature using ethanol (83%; percentage of ethanol not specified), and many shipped samples frozen using dry ice (54%). More than one-quarter of the collections surveyed (27%) also sent samples stored in DMSO as a preservative. More than one-third of those collections surveyed (38%) requested that researchers offset costs of processing and/or shipment in some way. Of those collections surveyed that asked researchers to offset costs, the majority (80%) requested that the researcher pay shipping costs, while one-third (33%) asked that the researcher pay a processing fee, which was assessed either per loan transaction or per sample processed. Only a small percentage (13%) required that researchers pay both processing fees and shipping costs. Of those collections that requested that the researcher pay shipping costs (80%), some (17%) noted that this only applied to those requesting international loans/gifts, which may have included fees associated with importation or exportation of the samples, and others (17%)

noted that some institutions or researchers were exempt from these charges. Those institutions or researchers that were exempt from shipping charges included institutions that maintained their own collections, researchers that contributed to the lending collection and institutional collaborators.

Requirements of research users

Collections representatives were asked about their loan policy in regards to unused genetic material and the publication of data resulting from use of the genetic material (Questions 55 and 56; Appendix 1). When a researcher completed a project, more than one-half of the collections surveyed (55%) asked that any unused sample be returned to the loaning collection for future use. A large percentage (45%) reported that the loanee was requested to dispose of the unused portion of the sample, while fewer (18%) asked that the loanee return the sample to the loaning collection to be disposed of by staff. Regardless of what was done with the genetic sample upon the completion of the project, the majority of collections (82%) reported that unused genetic material was handled in a consistent manner. In contrast, some collections (18%) indicated that they handled these unused portions using any of the three options (noted above), depending on the circumstances of the loan.

Upon completion of the research, most collections (78%) asked the researcher to supply reprints and/or citations of papers that resulted from the use of their genetic resource samples. The majority of collections (69%) also requested that researchers submit data to a public genetic sequence database, such as GenBank. More than one-half of the collections (54%) asked that researchers inform the collection of the accession numbers assigned by the public genetic sequence database to the submitted sequence data, so

they could track the genomic sequence data.

Additional relevant information

The last question in the survey (Question 57; Appendix 1) asked respondents to report any additional information regarding their collection that would be relevant or important to understanding genetic resource collections associated with natural history collections. Of the 45 surveyed collections, 17 collections (38%) used this opportunity to report additional information, sometimes regarding multiple topics. The majority of those that responded (76%) reported additional details about their respective collections (e.g., holdings, personnel, future plans). Approximately one-third of these respondents (29%) discussed general issues associated with genetic resource collections (e.g., funding, ownership, sample tracking), while about onequarter of the respondents (24%) outlined the importance of genetic resource collections (e.g., the use and value of genetic resources in research).

DISCUSSION AND CONCLUSIONS

Natural history collections have unique issues related to sample storage, voucher data, security and loans. A major goal of this project was to elucidate the current state of curation procedures used by genetic resource collections among institutions. Survey results revealed that practices varied widely across all aspects of curation and maintenance with respect to how samples are collected, processed, stored and distributed. Variance in practice was associated with basic differences among the institutions surveyed, including collection size, budget, personnel and storage locations. Ultimately, we hope that these results will stimulate discussion regarding the standardization of curation procedures and the future of genetic resource collections.

GENERAL COLLECTION INFORMATION

Although the collections surveyed were found in various types of institutions and were often stored in multiple locations within those institutions, it was clear that the centralization of genetic resource collections has become a common goal. More than onehalf had centralized repositories and, of those collections without centralized repositories (42%), more than one-third had plans to form a centralized facility in the near future, even though they also reported that lack of space, funding, and personnel were major obstacles. Another result illuminated by the survey was that the transfer of genetic samples out of individual laboratories or collections into centralized facilities led to increased organization and curation. Institutions with centralized facilities provided more precise information regarding sample locations and total holdings when compared to institutions with collections located in multiple locations.

Genetic resource collections are most often acquired as a result of time-consuming, expensive and specialized field collection by researchers. In addition, genetic samples must be stored in a low temperature environment if their products are to remain useful for molecular analyses. Owing to the investment in these collections and their inherent value, trained staff is needed to monitor cold storage equipment, work with data associated with samples and process samples for use in research. The survey found that the majority of collections had curators/professors with higher degrees (80%) or collection managers with higher degrees (51%) maintaining collections. Additional personnel included paid student assistants, collection managers without higher degrees, staff or technical assistants, unpaid student assistants and volunteers. Survey results could not determine the specific duties

of staff members or whether personnel had the appropriate education or training to work with these sample types. The majority of genetic resource collections surveyed (77%) had from 2 to 6 people working in them, which suggests that curators, professors or collection managers are overseeing collections, either by managing or training other staff members, while students (paid or unpaid), staff or technical assistants, or volunteers are completing day-to-day procedures. As a large portion of these collections were within or associated with colleges or universities, it is likely that many of these collections rely heavily on student assistants to complete the detailed tasks of organizing, labeling and databasing collections.

An important part of this survey was to determine what policies were present in relation to the curation and management of genetic resources and to determine what published guidelines, if any, are being used to inform these policies. All genetic resource collections at minimum should have internal guidelines to ensure both short- and long-term availability and integrity of these irreplaceable resources. The survey results revealed that the majority (78%) of those collections surveyed did have written internal guidelines to inform those working with samples of their collection policy regarding the correct storage and use. Institutional guidelines often give broad generalizations of how specimens in all collections within a museum or institution should be curated, but these guidelines generally are not specific enough to address the special curation needs of genetic samples. Interestingly, three-quarters of the collections surveyed without internal guidelines were associated with collections that were not centralized.

Collection representatives were asked whether published guidelines were used in

the curation or management of their genetic resources because it was impossible to know if internal guidelines were formulated from published information. Respondents were presented with four publications commonly used as resources for biorepositories, asked to select if any were relevant and requested to write in any additional publications used (Question 12; Appendix 1). The survey revealed that the more than one-half of collections with written departmental or institutional guidelines did not use published guidelines in the curation or management of their genetic resource collections. Since common principles apply to all biospecimen types, the presented guidelines published by ISBER (2012), NCI (2011) and OECD (2007) are useful for basic information regarding biorepositories that all genetic resource collections would find helpful. It should be noted, however, that these published guidelines do not address some unique issues for genomic collections within natural history collection. Prendini et al. (2002) gives detailed information on how to obtain, store and archive samples in zoological and botanical studies. Although this reference is more applicable to genetic resources in natural history museums, it was used by less than ten percent of those collections surveyed, possible because the resource is now over ten years old. Nagy (2010) presents a useful update of the methods listed by Prendini et al. (2002), but this paper is focused on available preservation methods and does not address many other aspects of the long-term care and use of genetic resources in natural history collections. The survey did not determine whether those that utilize the ISBER (2012) guidelines also use this organization's online self-assessment resource, which assists repository operators in determining how well their collection follows the ISBER Best Practices guidelines.

PROCESSING OF SPECIMENS

The initial methods of collecting genetic samples and the subsequent changes in sample storage before a sample's final deposition in a biorepository are important to document. The type of storage medium and the temperature at which a tissue is maintained can affect the future ways that it may be used for molecular analysis. For example, the proper collection and preservation of tissue samples is essential for isolating DNA, RNA and proteomes suitable for genomics, as these macromolecules are susceptible to rapid postmortem degradation. Flash-freezing samples at extremely low temperatures is the ideal way to initially preserve samples because it maximizes the research potential of that sample. It may be difficult or impossible, however, for those collecting samples in the field to keep samples cold using wet ice, dry ice or liquid nitrogen given possible time constraints and/or issues with transporting these substances. The survey results revealed that a number of different preservation techniques that do not require refrigeration were being used for the initial collection of genetic samples (Fig. 2). Ultimately, the initial method of sample collection may be influenced by a number of factors, including the initial sampling location, type of tissue that comprises the genetic sample and the eventual intended research use. The majority of those 45 collections surveyed indicated that their collections included samples preserved with ³ 95% ethanol or those that were flashfrozen (Fig. 2). Respondents also reported that samples were initially preserved using DMSO, RNAlater®, Allprotect Tissue Reagent, lysis buffers, buccal swabs, silica gel dessicant and FTA paper®.

The less-commonly used preservation techniques vary in their components and uses. DMSO is used in a number of different buffers

as an alternative to cryogenic preservation of tissue and is commonly included in a saltsaturated solution (20% DMSO, 0.25 M EDTA, pH 7.5, NaCl saturated; Seutin et al., 1991). RNAlater® solution and a similar product, Allprotect Tissue Reagent, are used for the stabilization of DNA, RNA, and protein in tissue samples, minimizing the need to immediately process and/or freeze tissue samples. Queen's lysis buffer was the only lysis buffer reported by name, and it is most often used for the preservation of DNA in nonmammalian vertebrates that possess nucleated red blood cells (Seutin et al., 1991). Buccal swabs are used to collect cheek cells from the inside of the mouth. These swabs can be kept at room temperature for a period of time before placing them in a freezer for long-term storage. Silica gel beads, made from sodium silicate, are a dessicant commonly used by botanical collections; samples can be stored at room temperature as long as the material is regularly checked for dryness (Chase and Hills, 1991). FTA paper® (patented by L. A. Burgoyne, Whatman) is impregnated filter paper that was developed for the collection and storage of DNA at room temperature (Smith and Burgoyne, 2004); fluid and small tissue samples are spotted on the filter and airdried.

26

Unfortunately, at this time, the manner by which samples are initially preserved during the sampling process may not be optimal for long-term storage or compatible with practices of the collection where they are deposited. To maximize long-term storage, samples may need to be transferred into new vials or their preservatives may need to be poured off before their final storage in a genetic resource collection. Numerous aspects of the sample transfer process, as well as how genetic samples were stored long-term, varied among the collections surveyed. The vials used for final storage of genetic samples by the 45 collections surveyed included externally or internally threaded vials with or without silicon gaskets, and there was not a single type of vial that was overwhelmingly preferred by these collections (Fig. 3). The survey also revealed that the majority of collections (69%) used multiple types of vials for initial storage, while most (63%) used a single vial type for final storage.

It was clear from the survey results that methods utilized to sterilize instruments during the sample transfer process were highly variable and, for a small subset of collections. may have yielded unintentional consequences. For the majority of respondents, ethanol, heat/flame and bleach were commonly used to sterilize equipment (Fig. 4). For a small group of collections (16%), additional methods were used to decontaminate or sterilize, including detergent/soap with water, autoclave, UV light and water. Some of these additional methods could be counterproductive to research objectives by leading to sample contamination if used as the sole method of sterilization. The potentially negative outcomes associated with improper sterilization of instruments stem from confusion between decontamination and sterilization and/or a questionable sample-transfer workflow. Decontamination renders instruments or surfaces safe for staff to touch, but these methods may not sterilize, which destroys all biological matter; instrument sterilization is needed to prevent contamination between different samples. For example, washing instruments with water or scrubbing instruments using detergent and water will likely decontaminate but will not sterilize. In addition, some techniques, such as heat sterilization in an autoclave and use of UV light, can decontaminate, but these techniques are likely only used before and/ or after processing a batch of samples and are likely not used while processing a number of independent samples. It was unclear if the

lack of consensus among the respondents in relation to the instrument sterilization process was related to lack of policy that addresses these procedures or lack of training for those working with these types of samples.

GENETIC RESOURCE COLLECTIONS SURVEY

Another procedural inconsistency in relation to sample processing concerned the removal of the preservation fluids in the vials before storage in collections that used liquid nitrogen. Of the surveyed collections that used liquid nitrogen (27%), many collections (42%) poured off preservatives and/or buffers on a case-by-case basis. One-third of the collections never poured off liquids before final storage in a liquid nitrogen collection, which is potentially owing to the time involved with such a task, while one-quarter of collections always removed the original preservatives or buffers from samples. The lack of consensus in regards to this practice may also be related to the fact that no studies have been undertaken to investigate the long-term effects of many preservatives, including DMSO, RNAlater®, and Allprotect Tissue Reagent, on genetic sample quality.

One of the major goals of this survey was to determine the methods of storage being used by genetic resource collections associated with natural history museums. Cryogenic storage at extremely cold temperatures is the most efficient way of preserving genomic material because biological activity substantially slows at temperatures less than approximately -136 °C (Engstrom et al., 1990; Kilpatrick, 2002; Mutter et al., 2004; Corthals and DeSalle, 2005). Although cryovats cooled by liquid nitrogen are the best means to achieve this storage efficiency for genetic samples, the survey results indicated that natural history collections stored genetic resources in a number of ways (Fig. 5). Variability in cold storage of genetic samples is clearly linked to aspects of the institution where genomic collections reside, including collection size,

budget, personnel and space. Interestingly, the survey results found that centralized facilities were more likely to store samples in ultracold freezers rather than liquid nitrogen, which is likely a result of both the costs associated with liquid nitrogen equipment and personnel needed for monitoring the supply of liquid nitrogen.

The survey results indicated those collections using liquid nitrogen equipment obtained liquid nitrogen in various ways. Most repositories (using liquid nitrogen) had the product delivered and stored in dewars (58%), while one-third had liquid nitrogen delivered and stored in bulk tanks. Bulkstorage tanks have the ability to supply a number of cryovat units, but owing to their large size must be house outside of buildings. It is important to note that expensive, vacuum jacketed cryogenic pipes are needed to deliver the liquid nitrogen from the bulk tank to the point of use, so larger bulk tanks must be located as close to the collection space as possible. A single collection (8% of collections using liquid nitrogen) reported that they produced their own liquid nitrogen on site with a liquid nitrogen plant, which generates nitrogen by separating it from the other components of the air. The use of liquid nitrogen plants is not a feasible option for most natural history museums due to the amount of liquid nitrogen needed to operate several cryovats, as well as the associated energy costs and required maintenance.

SAMPLE LABELING, TRACKING AND DATA

The ability to retrieve samples from a collection is an integral part of the curation and management of any natural history collection. The manner by which the physical locations of genetic samples are tracked allows users to maximize storage capacities, reduce the amount of time needed to locate samples,

and minimize the potential of sample loss. The use of databases has improved the efficiency of specimen retrieval by recording precise location data and, in addition, the use of rack and box storage systems has been instrumental in improving the general organization of genetic samples. The majority of collections surveyed (82%) used a rack and box system, and collections also often organized samples by department/division (36%) or taxonomic group (22%). The potential risk to the collections without a rack and box system of organization is greater owing to the practice of stacking boxes on top of one another within freezers, which forces users to remove boxes from a freezer to access boxes underneath or behind them.

28

Barcodes are now commonly being used to facilitate sample tracking and retrieval, especially for large collections. Although more than one-half of collections surveyed wrote information directly onto vials by hand, preprinted or self-printed barcode labels that wrap completely around vials were also used in almost one-third of the collections surveyed. Barcodes printed on wrapping labels have the benefit of allowing users to view information previously written on vials if a portion of the label is transparent and positioned over the writing. Vials that include directly-printed barcodes (vials manufactured with barcodes) on the sides or bottoms of vials were not common (12%), which is not surprising given that additional information cannot be hand-written on these types of vials. Barcodes inserted into vial caps were rarely used (7%), and RFID tags, which tracks using intelligent barcodes, are not being used to label vials by any surveyed collections, which is likely due to their high cost.

Genetic samples in natural history museums are generally housed separate from the vouchers from which they were originally sampled because of their different storage and conservation needs. Given the independent storage of the physical voucher and genetic samples, collections were asked how data were tracked for these two specimen types. The survey results found that the majority of collections (63%) directly associated geneticsample data with voucher-specimen data using an internal database, internal spreadsheet or online database, while one-third used independent internal or online databases to track the genetic and voucher-specimen data, respectively. For the collections using separate systems to track the data associated with these two preparation types, it is likely that some data changes are not made, owing to the effort needed to coordinate two separate data systems. In addition, more than onequarter of collections surveyed reported that taxonomic or other data changes associated with the original voucher specimens were only sometimes made, while 5% reported that these updates were never made.

SECURITY AND SAFETY

The use of ultra-cold temperatures generally requires additional safety precautions and equipment specifically rated for cryogenic conditions. By the nature of these requirements, access and use of genetic resource collections must be limited to those with the appropriate training. In addition, genomic collections have often been amassed as a result of numerous, costly collection trips, making them extremely valuable. These samples should, therefore, be stored in secure locations to minimize theft or inadvertent loss, and the appropriate back-up power systems and/or monitoring should be present in case of freezer failure. The survey revealed that almost one-third of collections surveyed stored samples in a room or facility that remained unlocked during the day and, of those collections storing samples in rooms that remained unlocked, the majority (62%)

also did not store samples in locked freezer units. As many genetic resource collections are research collections stored within college and/ or university laboratories, it is likely that doors are left unlocked during the day so students may have easy access. In addition, the survey found that more than one-third of collections surveyed reported that individuals with access to the room where genetic resources were stored also had access to samples, which likely included individuals not trained to work with these types of collections.

GENETIC RESOURCE COLLECTIONS SURVEY

There are clear advantages and disadvantages to the types of cold storage currently being used by genetic resource collections in relation to maintenance and monitoring. Liquid nitrogen cryovats can maintain ultra-cold temperatures without external power but require consistent and dedicated maintenance. In contrast, samples stored in electric freezers require less scheduled maintenance but are at risk of loss in the event of mechanical breakdown or power disruption. Even a short power outage may be detrimental to samples being stored in a mechanical freezer, and there have been documented cases of large collections of samples being compromised due to freezer failures (Hanner et al., 2005). The survey was insightful in regards to the use of safety measures among all surveyed collections, as well as centralized repositories and those that use liquid nitrogen equipment (Fig. 6). In general, audible or visual alarms, uninterruptible power supplies, and outside monitoring were present in similar percentages across all surveyed collections when compared centralized repositories and those collections storing samples in liquid nitrogen. The survey results indicated that battery backup systems were present in more than one-half of collections using liquid nitrogen but far less common in centralized repositories (15%), which is noteworthy for two reasons. First, a battery back-up system within a collection

using liquid nitrogen would function only to power the equipment used to supply the cryovat with liquid nitrogen as the unit itself would stay cold without power. Second, since the survey found that most centralized repositories (61%) included ultracold freezers, one can infer that many collections using ultracold freezers do not have battery back-up systems.

USE OF COLLECTIONS

Almost all of the 45 collections surveyed had active loan/gift programs and, due to the consumptive nature of genetic resources, it seems prudent that repositories develop clear policies in relation to the distribution of material to justify that a loan/gift is warranted. Unlike traditional natural history specimen loans, the survey revealed that those to whom the incoming request was submitted for processing often differed from those who ultimately approved the loan/gift request. This difference in the approval process is likely due to the location of the samples (i.e., in the laboratory of an individual researcher, department/collection centralized or repository) or policies regarding who retains custodianship and/or rights to approve loans/ gifts.

Genetic resources often require special means of shipment to preserve the integrity of the samples, which can become cost prohibitive to those institutions sending large numbers of loans/gifts. The survey found that genetic resources were shipped using numerous methods and most commonly were mailed at ambient temperature using ethanol or frozen using dry ice. Differences among shipping methods used by these collections are likely linked to shipping restrictions and costs, as well as the intended use of the samples. For example, shipping genetic samples on dry ice is more expensive than shipping at

room temperature but may be required when working with specific types of macromolecules, such as RNA. The survey also revealed that the costs associated with processing and/or shipping samples was an issue for many of the institutions as more than one-third requested that researchers offset costs of processing and/or shipment in some way. Of those collections surveyed that offset costs of processing and/or shipment, the majority (80%) asked that researchers pay shipping costs, while one-third asked that researchers pay processing fees, which were assessed either per transaction or per sample processed.

Unlike traditional natural history specimens, which are returned after the completion of a project, genetic samples are most often completely consumed by the researcher. The survey revealed that aspects of processing loans/gifts differed among the collections surveyed, which is likely linked to the consumptive use of genetic material. Although none of the collections surveyed sent the entire aliquot or sample, no clear consensus was found in respect to the amount or size of the sub-sample or aliquot sent. In addition, when portions of loans are leftover after a project has been completed, the sample can be disposed of by the user or returned to the lending collection. More than one-half of the collections surveyed asked that any unused sample be returned to the original collection for future use. Using returned samples for future loans obviously raises the question whether samples were compromised given that they could have been contaminated or mislabeled while being used on loan. The survey did not determine if the history of a returned genetic sample was tracked, which would allow collections to advocate the use of a returned loan only as a last resort. Alternatively, less than one-half of collections surveyed reported that researchers were requested to dispose of the unused portion of the sample themselves.

A minority of collections surveyed (18%) asked that the researcher return the sample to the loaning collection to be disposed of by staff. The majority of collections (82%) dealt with unused genetic material in a single way, suggesting their collection has a policy in relation to the unused portions of loaned genetic samples. However, some collections (18%) handled these unused portions in two or three different ways, suggesting that either there is no collection policy in relation to leftover genetic samples or decisions regarding loan returns are made on a case-by-case basis.

Most genetic resource collections requested that researchers supply information regarding publications and genetic sequence data resulting from the use of their material. Similar to many other natural history collections, most genetic resource collections (78%) asked that reprints and/or citations of publications resulting from the use of their samples be supplied to the originating collection. One major difference between genetic resource collections and other natural history collections is that genetic data resulting from use of the samples can be submitted to a public genetic sequence database, such as GenBank. The majority of collections (69%) requested that researchers submit data to a public genetic sequence database. In addition, most collections (54%) asked that researchers inform the collection of the genetic sequence accession numbers assigned to the sequences by these public genetic sequence databases, suggesting that most genetic resource collection want to track the end-products of research (genetic sequences) and possibly link them to the original genetic samples and/or voucher specimens.

DEVELOPING BEST PRACTICE STANDARDS

The development of standardized methods for genetic resource collections within natural

history museums is clearly needed to ensure the long-term, consistent availability of highquality genetic samples for research purposes. With the establishment of centralized repositories, institutions must develop internal administration, ownership, management, and associated policies to address the growing compliance and regulatory issues associated with importing, exporting, utilizing, and housing genetic resources. It is clear from the results of this survey, however, that not all institutions may have the funding, space or personnel needed to create a centralized repository. In addition, many may want to keep collections within the laboratory environment because of an active research program. We, therefore, advocate the creation of guidelines for the curation of genetic resources that include tiers or levels of best practice standards wherever possible. Institutions can use these guidelines as a point of reference and tailor them to fit the parent institution's needs, goals and research programs.

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Appendix 1. Survey questionnaire as posted online using Harvard University's iCommons Poll Tool. Multiple-choice questions that allowed more than one selection were indicated in the question by informing respondents to check all that apply. Comments in square brackets convey how many characters were available for text responses.

Survey Questionnaire: Developing best practices for genetic resources associated with natural history collections

This research study is about the current protocols, challenges, and concerns associated with genetic resource collections. You will be asked questions about how your institution currently deals with genetic resources, what you consider the value of genetic resources to be, and what challenges you see in curating these types of collections. You will also be asked for demographic information, such as the type and size of your institution, and your role at that institution. This research project is being conducted by Dr. Breda Zimkus (bzimkus@oeb. harvard.edu), Project Manager of the Museum of Comparative Zoology Cryogenic Collection, and is funded by CollectionsWeb (http://collectionsweb. org/). Feel free to contact Breda via email with any questions that you may have.

Question 1: Name [Text response limited to 250 characters]

Question 2: Title [Text response limited to 250 characters]

Question 3: Email Address [Text response limited to 250 characters]

Question 4: Institutional Address [Text response limited to 250 characters]

Question 5: Please give a brief description of your position's duties. [Text response limited to 500 characters]

An increasing number of universities and natural history museums have established genetic resource facilities that are shared by various divisions and disciplines to combine resources and share in the maintenance and processing of standard collection functions, such as research loans. We would like to know more about your individual institution to

understand how genetic resources are managed and utilized.

Question 6: Type of Institution (Check all that apply.)

- A. Botanical garden or herbarium
- B. Cell line center
- C. Culture or stock center
- D. Natural history museum
- E. Private institute or laboratory
- F. Seed or spore bank
- G. Science museum or science center
- H. University/college
- I. Zoo
- J. Other; please describe [Text response limited to 250 characters]

Question 7: What types of genetic resources does your institution have? (Check all that apply.)

- A. Antisera
- B. Bacterial artificial chromosome (BAC)
- C. Cell cultures
- D. Cell lines
- E. Chemically preserved tissues
- F. DNA
- G. Frozen tissues
- H. Proteins
- I. PCR products
- J. RNA
- K. Other; please describe [Text response limited to 250 characters]

Question 8: Where do genetic samples originate from? (Check all that apply.)

- A. Researchers from institution
- B. Affiliates of institution
- C. Outside researchers/institutions (i.e., gifts, donations)
- D. Other; please describe [Text response limited to 250 characters]

Question 9: Approximately how many samples are present at your institution? Specify whether the number of samples refers to the number of actual vials or the approximate number of specimens with an associated genetic sample. If you only have knowledge of a subset of the total samples at your institution (i.e., personal research samples

or departmental samples), please also specify this in your answer. [Text response limited to 500 characters]

Question 10: Briefly describe the taxonomic groups that are represented in your collection. Listing the phyla/classes are sufficient for diverse collections, while orders/families are all that is needed for smaller collections. If you only have knowledge of a subset of the total samples at your institution (i.e., personal research samples or departmental samples), please specify this in your answer. [Text response limited to 500 characters]

Question 11: Who maintains the genetic resources at your institution? (Check all that apply.)

- A. Curator/Professor with higher degree
- B. Collection Manager with higher degree
- C. Collection Manager without higher degree
- D. Staff or Technical Assistant without higher degree
- E. Unpaid student assistant (i.e. undergraduate or graduate student)
- F. Paid student assistant (i.e., undergraduate or graduate student)
- G. Volunteer
- H. Other; please describe [Text response limited to 250 characters]

Question 12: What guidelines are used for the curation and management of genetic resources? (Check all that apply.)

- A. International Society for Biological and Environmental Repositories (ISBER) Best Practices for Repositories
- B. National Cancer Institute (NCI) Best Practices for Biospecimen Resources
- C. Organisation for Economic Co-operation and Development (OECD) Best Practice Guidelines for Biological Resource Centres
- D. Prendini, L., Hanner, R. and DeSalle, R. Obtaining, storing and archiving specimens and tissue samples in molecular studies
- E. Departmental or institutional guidelines
- F. No written guidelines are used
- G. Other; please describe [Text response limited to 250 characters]

Question 13: Who determines policy that involves genetic resources? (Check all that apply).

- A. Board or governing group
- B. Curator/Professor
- C. Collections Manager with higher degree
- D. Collections Manager without higher degree
- E. Staff or Technical Assistant without higher degree

Question 14: Is there a centralized repository for genetic resources at your institution?

- A. No
- B. Yes

Question 15: PLEASE LEAVETHIS QUESTION BLANK IF THERE IS A CENTRALIZED REPOSITORY FOR GENETIC RESOURCES AT YOUR INSTITUTION. If there is NOT a centralized repository for genetic resources, in how many separate physical locations (i.e., laboratories or rooms) are genetic resources stored? [Text response limited to 500 characters]

Question 16: PLEASE LEAVETHIS QUESTION BLANK IF THERE IS A CENTRALIZED REPOSITORY FOR GENETIC RESOURCES AT YOUR INSTITUTION. If there is NOT a centralized repository for genetic resources, please select the major reasons why. (Check all that apply.)

- A. Lack of funding
- B. Lack of space
- C. Lack of personnel
- D. Too few samples
- E. Other; please describe [Text response limited to 250 characters]

Question 17: PLEASE LEAVETHIS QUESTION BLANK IF THERE IS A CENTRALIZED REPOSITORY FOR GENETIC RESOURCES AT YOUR INSTITUTION. If there is not a centralized repository for genetic resources, is there a future plan for one?

- A. No
- B. Yes, within 1-3 years
- C. Yes, 3+ years

Question 18: If your institution has a centralized repository for genetic resources, what is its current status?

- A. Infrastructure is complete and samples are in their permanent storage locations
- B. Infrastructure is complete and samples are being moved to permanent storage
- C. Infrastructure is not yet complete
- D. Not applicable- no centralized repository
- E. Other; please describe [Text response limited to 250 characters]

Question 19: Who works in the centralized genetics resources facility? (Check all that apply.)

- A. Curator/Professor with higher degree
- B. Collection Manager with higher degree
- C. Collection Manager without higher degree
- D. Staff or Technical Assistant without higher degree
- E. Student assistant
- F. Volunteer
- G. Not applicable no centralized repository
- H. Other; please describe [Text response limited to 250 characters]

Question 20: What type of work is completed in the centralized genetic resources facility? (Check all that apply.)

- A. DNA/RNA extraction
- B. Organization of samples (i.e., storage and tracking)
- C. PCR
- D. Sequencing
- E. Tissue sub-sampling and loans
- F. None of the above
- G. Not applicable- no centralized repository
- H. Other; please describe [Text response limited to 250 characters]

Question 21: If there is a centralized repository, what equipment is present in it? (Check all that apply.)

- A. Biosafety cabinet
- B. Centrifuge
- C. Fume hood
- D. Gel electrophoresis equipment)
- E. General-purpose or laboratory freezers (between -12° and -30°C)

- F. Liquid nitrogen cryovats (below -110°C)
- G. Ultracold freezers (between -50°C and -86°C)
- H. Vortex
- I. Not applicable- no centralized repository
- J. Other; please describe [Text response limited to 250 characters]

SAMPLE STORAGE AND TRACKING

Question 22: How are samples initially preserved by researchers? (Check all that apply.)

- A. 95% ethanol
- B. Allprotect Tissue Reagent
- C. Dimethylsulfoxide (DMSO)
- D. Flash frozen at -80 degrees celsius or below
- E. RNALater Stabilization Reagent
- F. Other; please describe [Text response limited to 250 characters]

Question 23: What types of vials are used by researchers when collecting genetic samples? (Check all that apply.)

- A. Any type of vial
- B. Externally threaded vial with silicon gasket
- C. Externally threaded vial without silicon gasket
- D. Internally threaded vial with silicon gasket
- E. Internally threaded vial without silicon gasket
- F. Other; please describe [Text response limited to 250 characters]

Question 24: How are samples labeled when received by the centralized genetic resources facility or laboratory where genetic resources are stored? (Check all that apply.)

- A. Barcode
- B. Catalog number
- C. Collector number or field number
- D. Other; please describe [Text response limited to 250 characters]

Question 25: Are samples transferred to different vials for final storage?

- A. Always
- B. Never
- C. Sometimes; briefly describe some reasons why samples are transferred (i.e., vials

are changed if not rated for cryogenic conditions) [Text response limited to 250 characters]

Question 26: How are instruments sterilized/ decontaminated during the sample transfer process?

- A. Heat/flame
- B. Bleach (sodium hypochlorite; NaClO)
- C. DNase Away, RNase Away or similar surface decontaminant
- D. Ethanol
- E. Hydrogen peroxide
- F. Other; please describe [Text response limited to 250 characters]

Question 27: What types of vials are used for final specimen storage? (Check all that apply.)

- A. Any type of vial
- B. Externally threaded vial with silicon gasket
- C. Externally threaded vial without silicon gasket
- D. Internally threaded vial with silicon gasket
- E. Internally threaded vial without silicon gasket
- F. Other; please describe [Text response limited to 250 characters]

Question 28: Is a specific brand or type of vial recommended by the institution or genetic resources facility for the collection of samples? [Text response limited to 500 characters]

Question 29: Please describe if additional information is written on tubes or cap inserts after receipt by the genetic resources facility and before final storage in the collection (e.g., catalog number, locality data). [Text response limited to 500 characters]

Question 30: How are genetic samples labeled/tracked? (Check all that apply.)

- A. Barocode inserted into vial cap
- B. Laser etched vials
- C. Numbers written directly on vials with marker
- D. Pre-printed/self-printed barcodes that wrap around vials

- E. Pre-barcoded vials (barcode on side of vial
- F. Pre-barcoded vials (barcode on bottom of tube
- G. Radio frequency identification (RFID) tags
- H. Other; please describe [Text response limited to 250 characters]

Question 31: How are samples physically tracked? (Check all that apply).

- A. Vial boxes only; no rack or barcode system
- B. Vial boxes and rack storage without samples barcoded
- C. Vial boxes and rack storage with samples barcoded
- D. Vial boxes and rack storage with radio frequency identification (RFID) tags
- E. Other; please describe [Text response limited to 250 characters]

Question 32: If barcodes are used, please describe the type.

- A. 1D
- B. 2D: DataMatrix
- C. 2D: ORCode
- D. Not applicable
- E. Other; please describe [Text response limited to 250 characters]

Question 33: How are the data associated with genetic samples recorded? (Check all that apply.)

- A. Internal database
- B. Internal spreadsheet
- C. Online database; please name platform [Text response limited to 250 characters]

Question 34: How are the data associated with the voucher specimens (from which the genetic resources were sampled) recorded?

- A. Different internal database from genetic samples
- B. Different online database as genetic samples
- C. Different spreadsheet from genetic samples D. Same internal database as genetic samples
- E. Same online database as genetic samples
- F. Same spreadsheet as genetic samples
- G. Other; please describe [Text response limited to 250 characters]

Question 35: Are taxonomic changes made to the original voucher specimens also applied to the genetic samples?

- A. Always
- B. Never
- C. Sometimes; please give a brief explanation [Text response limited to 250 characters]

Question 36: Are samples organized by departments or major taxonomic groups?

- A. No
- B. Yes; please give brief explanation [Text response limited to 250 characters]

Question 37: How are samples stored long-term? (Check all that apply.)

- A. General-purpose or laboratory freezers (-12°C to -30°C)
- B. Liquid nitrogen cryovats (below -110°C)
- C. Room temperature using preservative
- D. Ultracold freezers (-50°C to -86°C)
- E. Other; please describe [Text response limited to 250 characters]

Question 38: PLEASE LEAVE THIS QUESTION BLANK IF LIQUID NITROGEN STORAGE IS NOT USED. If samples are stored in liquid nitrogen cryovats, are preservatives or buffers (if present) poured off before storage?

- A. Always
- B. Never
- C. Sometimes; briefly describe what types of liquids are poured off [Text response limited to 250 characters]

Question 39: PLEASE LEAVE THIS QUESTION BLANK IF LIQUID NITROGEN STORAGE IS NOT USED. If liquid nitrogen-cooled cryovats are used, how is the liquid nitrogen most often obtained and stored?

- A. Liquid nitrogen delivered and stored in dewars
- B. Liquid nitrogen delivered and stored in bulk tank
- C. Liquid nitrogen made on site with production plant and stored in dewars
- D. Other; please describe [Text response limited to 250 characters]

Question 40: PLEASE LEAVETHIS QUESTION BLANK IF LIQUID NITROGEN STORAGE IS NOT USED. If a bulk storage system is used, briefly describe the system and approximate size of the tank. [Text response limited to 500 characters]

Question 41: How are genetic resources secured? (Check all that apply.)

- A. In room or facility that is not locked
- B. In room or facility that is locked overnight but accessible during day
- C. In room or facility that is always locked
- D. In locked units (freezers, etc.)

Question 42: In addition to trained personnel, who has access to genetic resources? (Check all that apply.)

- A. No others have access; only trained personnel
- B. All those who have access to building
- C. All those who have access to the room where samples are stored; please give a brief list of untrained personnel who would have access to samples (i.e., post-doctoral fellows, graduate students, undergraduate students)

Question 43: Are there back-up systems in place for cold storage units? (Check all that apply.)

- A. Audible or visual alarm only
- B. Battery back-up to ensure continuous supply of electrical power
- C. Uninterruptible power supply
- D. Monitoring by outside security (e.g., university controls or external contract)

Question 44: If liquid nitrogen cryovats are used, are there safety devices and/or specialized exhaust systems to protect users from low-oxygen levels? (Check all that apply.)

- A. Oxygen monitor
- B. Specialized exhaust system
- C. Not applicable
- D. Other; please describe [Text response limited to 250 characters]

Question 45: What is the annual budget for the centralized genetic resources facility or laboratory

where genetic resources are stored? [Text response limited to 500 characters]

Question 46: Has your institution received outside funding to assist in building a genetic resources facility or completing barcoding projects (e.g., NASA, NSF, NSERC)?

- A. No
- B. Yes; type of funding

Question 47: PLEASE LEAVE ALL REMAINING QUESTIONS BLANK IF LOANS ARE NOT MADE TO OTHER INSTITUTIONS). Are loans of genetic material made to other institutions?

- A. No (SURVEY IS COMPLETE)
- B. Yes

Question 48: What is the average number of gifts and/or loans sent out per year? [Text response limited to 500 characters]

Question 49: To whom are genetic resource requests submitted? (Check all that apply.)

- A. Individual researcher
- B. Department/collection from which sample originated
- C. Centralized genetic resources collection
- D. Other; please describe [Text response limited to 250 characters]

Question 50: Who approves loan requests? (Check all that apply.)

- A. Individual researcher
- B. Department/collection where specimen stored
- C. Genetic resources collection
- D. Other; please describe [Text response limited to 250 characters]

Question 51: What information is submitted with loan requests? (Check all that apply.)

- A. Description of research
- B. Evidence of sufficient lab facilities and funding
- C. Feasibility of the project
- D. Number of individuals or specimens requested
- E. Scientific value of research

- F. Qualifications of investigator
- G. Other; please describe [Text response limited to 250 characters]

Question 52: What is the standard amount of genetic resource sent out on loan (e.g., specific amount or volume, amount sufficient for 2 extractions)? [Text response limited to 500 characters]

Question 53: Are there any costs charged for loans to outside requestor?

- A. No
- B. Yes; please describe (give amounts if possible) [Text response limited to 250 characters]

Question 54: How are loans shipped? (Check all that apply.)

- A. Shipped frozen using dry ice
- B. Shipped via at ambient temperature with ethanol
- C. Shipped via at ambient temperature with DMSO)
- D. Other; please describe [Text response limited to 250 characters]

Question 55: What is required from the loanee? (Check all that apply.)

- A. Deposition of sequences to GenBank or other public database
- B. Communication of GenBank or other publish database accession numbers
- C. Reprints of papers resulting from use of samples
- D. Return unused portion of loan
- E. Other; please describe [Text response limited to 250 characters]

Question 56: What is done with unused portions of the loan? (Check all that apply.)

- A. Disposed of by loanee
- B. Returned to collection to be used for other loans
- C. Returned to collection to be disposed of by staff
- D. Other; please describe [Text response limited to 250 characters]

38 Zimkus and Ford

Question 57: Please include any additional information regarding your collection that may be relevant or important to understanding genetic resources associated with natural history museums. [Text response limited to at least 1500 characters, depending on font]

Table 1. List of 45 collections within 39 institutions surveyed in an online poll of collections with genetic resources (survey questions in Appendix 1). "Genomic Samples" refers to genetic resources, whereas "Voucher Specimens" refers to the original (whole) specimens from which the genetic resources were sampled. Additional details given by respondents in relation to the number of genetic samples or voucher specimens present in their collections are indicated with footnotes at the end of the table. Responses for "Collection Represented" were determined by answers given in Questions 2 and 10 (respondent's title and taxonomic groups represented in collection, respectively). Respondents provided details regarding "Type of Institution" in Question 6 (10 answer choices available, including "Other" that allowed a text response). Responses for "Locations of Samples" were determined using answers provided for Questions 14 and 15 (existence of centralized repository at institution and physical locations of specimens, respectively). Respondents provided details regarding "Status of Centralized Repository" in their responses to Question 18, and information regarding "Storage Types" was obtained from answers given in Question 37. "No. of Genomic Samples" and "No. of Voucher Specimens" were determined by the responses given to Question 9. "N/A" indicates answers that are not applicable for the corresponding survey question, and "—" indicates information not given in response to the survey question.

Institution	Collection Represented	Type of Institution	Locations of Samples	Status of Centralized Repository	Storage Types	No. of Genomic Samples	No. of Voucher Specimens
Academy of Natural Sciences (Philadelphia, PA, USA)	Ornithology Collection	natural history museum	1 location (centralized repository)	infrastructure complete; samples in storage	ultracold freezers	125,000	—
Australian National Fish Collection, CSIRO Marine and Atmospheric Research (Hobart, Tasmania, Australia)	Australian National Fish Collection	natural history museum	1 location (centralized repository)	infrastructure complete; samples in storage	general purpose or laboratory freezers; ultracold freezers; cold room (4°C)	15,000	_
Bell Museum of Natural History University of Minnesota (St. Paul, MN, USA)	Genetic Resource Collection	botanical garden or herbarium; natural history museum; university/college	5 locations	N/A	ultracold freezers	42,100	
Burke Museum of Natural History (Seattle, WA, USA)	Genetic Resource Collection	natural history museum	1 location (centralized repository)	infrastructure complete; samples in storage	general purpose or laboratory freezers; ultracold freezers	45,000	45,000
California Academy of Sciences (San Francisco, CA, USA)	Center for Comparative Genomics/ CryoCollection	natural history museum; science museum or science center	1 location (centralized repository)	infrastructure complete; samples in storage	ultracold freezers	24,500	<u> </u>
Carnegie Museum of Natural History (Pittsburg, PA, USA)	Section of Invertebrate Zoology	natural history museum	2 locations	N/A	ultracold freezers	17,000	_
Carnegie Museum of Natural History (Pittsburg, PA, USA)	Section of Mammals	natural history museum	2 locations	N/A	ultracold freezers	_	14,400*

Cincinnati Museum Center (Cincinnati, OH, USA)	Zoology Collection	natural history museum; science museum or science center	1 location (centralized repository)	infrastructure complete; samples in storage	ultracold freezers	5,000	2,500
Conner Museum School of Biological Sciences, Washingtor State University (Pullman, WA, USA)		natural history museum; science museum or science center; university/college; other (vertebrate museum; research collection; public natural history exhibit)	I location (centralized repository)	infrastructure not yet complete	general purpose or laboratory freezers; ultracold freezers (within 2 years)		250*
Cornell University (Ithaca, NY, USA)	The Cornell Lab of Ornithology	natural history museum; university/college	partially centralized (Museum of Vertebrates)	infrastructure complete; samples in storage	ultracold freezers	6,000	_
Denver Museum of Science and Nature (Denver, CO, USA)	Department of Vertebrate Zoology	science museum or science center	1 location (centralized repository)	infrastructure complete; samples in storage	ultracold freezers	2,650	_
Denver Museum of Science and Nature (Denver, CO, USA)	Health Sciences Department	science museum or science center	2 locations	N/A	ultracold freezers	1,800	—
Department of Biology and the M. L. Bean Life Science Museum, Brigham Young University (Provo, UT, USA)	Crustacean Collection/Research Collection	natural history museum; university/college	10 locations (approx.)	N/A	ultracold freezers	8,000	_
Department of Biology and the M. L. Bean Life Science Museum, Brigham Young University (Provo, UT, USA)	Herbarium of Nonvascular Cryptogams	university/college	12 locations	N/A	general purpose or laboratory freezers; ultracold freezers	108,000	_
Department of Biology and the M. L. Bean Life Science Museum, Brigham Young University (Provo, UT, USA)	Ichthyology Collection	natural history museum; university/college	4 locations	infrastructure not yet complete	room temperature using preservative; ultracold freezers	25,000	<u> </u>

Department of Biology and the M. L. Bean Life Science Museum, Brigham Young University (Provö, UT, USA)	Research Collection	natural history museum; university/college	3 locations	infrastructure not yet complete	room temperature using preservative; ultracold freezers	36,000	<u> </u>
Florida Museum of Natural History, University of Florida Gainesville, FL, USA)	Genetics Resources Repository	botanical garden or herbarium; natural history museum; seed or spore bank; university/college	1 location (centralized repository); 1 additional collection (Lepidoptera)	infrastructure complete; samples in storage	liquid nitrogen cryovats; ultracold freezers	33,294	<u>—</u>
Kansas University Biodiversity nstitute (Lawrence, KS, USA)	Herpetology Collection	university/college	1 location (centralized repository)	infrastructure not yet complete	liquid nitrogen cryovats; ultracold freezers	35,000	_
Kansas University Biodiversity Institute (Lawrence, KS, USA)	Ichthyology Collection	natural history museum; university/college	1 location (centralized repository)	infrastructure complete; samples being moved	liquid nitrogen cryovats	15,989	10,298
Museo de Zoología, Pontificia Jniversidad Católica del Ecuador (Quito, Ecuador)	Amphibian Collection	natural history museum; university/college	1 location (centralized repository)	infrastructure complete; samples in storage	ultracold freezers	35,000	
Museum of Comparative Zoology (Cambridge, MA, USA)	Cryogenic Collection	natural history museum	1 location (centralized repository)	infrastructure complete; samples being moved	liquid nitrogen cryovats	61,500 ¹	<u>—</u>
Museum of Southwestern Biology and Department of Biology, University of New Mexico (Albuquerque, NM, JSA)	Division of Genomic Resources	university/college	1 location (centralized repository)	new infrastructure needed	ultracold freezers	400,000	
Museum of Vertebrate Zoology Berkeley, CA, USA)	Genetic Research Collection	natural history museum	1 location (centralized repository)	infrastructure complete; samples in storage	liquid nitrogen cryovats; ultracold freezers	100,000	90,000
Museum of Zoology, University of Michigan (Ann Arbor, MI, USA)	Mammal Collection	natural history museum; university college	4 locations	N/A	isothermal liquid nitrogen freezers	61,000	<u>—</u>
Natural History Museum of Los Angeles County	Mammal Collection	natural history museum	10 locations (approx.)	N/A	general purpose or laboratory freezers	4,500	

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National Museum of Natural History (Smithsonian Institution, Suitland, MD, USA)	National Museum of Natural History Biorepository	natural history museum	1 location (centralized repository)	infrastructure not yet complete	liquid nitrogen cryovats; ultracold freezers	300,000 ²	
Natural Sciences Museum of Barcelona (Barcelona, Spain)	Natural History museum	natural history museum	2 locations	N/A	room temperature using preservative	4,800	
North Carolina Museum of Natural Sciences (Raleigh, NC, USA)	Amphibian and Reptile Collection	natural history museum	1 location (centralized repository)	infrastructure complete; samples being moved	ultracold freezers	40,719	_
Ocean Genome Legacy (Ipswich, MA, USA)	Marine Genome Resource Bank and Data Collection	other (non-profit marine research center)	1 location (centralized repository)	infrastructure complete; samples in storage	liquid nitrogen cryovats; ultracold freezers	32,000	
Plant Pest Diagnostics Center, California Department of Food & Agriculture (Sacramento, CA, USA)	Plant Pest Diagnostic Program (Entomology and Botany)	botanical garden or herbarium; natural history museum	5 locations	N/A	ultracold freezers	15,000 ³	—
Queensland Museum (Brisbane, Australia)	Biodiversity and Geosciences Program	other (state museum that includes natural history, cultures and history)	(centralized	infrastructure complete; samples in storage	ultracold freezers	8,000	
Royal Ontario Museum (Toronto, ON, Canada)	Herpetology	natural history museum	1 location (samples managed by discipline)	N/A	ultracold freezers	30,000	—
Royal Ontario Museum (Toronto, ON, Canada)	Green Plant Herbarium (TRT)	botanical garden or herbarium; other (general museum)	2 locations	N/A	general purpose or laboratory freezers	1,500	
Sam Noble Museum, University of Oklahoma (Norman, OK, USA)	Genomic Resources	natural history museum university/college	1 location (centralized repository)	infrastructure complete; samples in storage	general purpose or laboratory freezers; room temperature using preservative; ultracold freezers	26,184	10,045

South Australian Museum (Adelaide, Australia)	Australian Biological Tissue Collection	natural history museum	1 location (centralized repository)	infrastructure complete; samples in storage	ultracold freezers	_	140,000*
Sternberg Museum of Natural History, Fort Hays State University (Sternberg, KS, USA)	Zoological Collections	natural history museum; university/college	2 locations	N/A	room temperature using preservative	_	12,000*
Texas Natural Science Center, Γhe University of Texas at Austin (Austin, TX, USA)	Herpetology and Ichthyology	science museum or science center; university/college	1 location (centralized repository)	infrastructure complete; samples in storage	liquid nitrogen cryovats	30,000	_
	Genetic Resources Collection	university/college	1 location (centralized repository)	infrastructure complete; samples in storage	general purpose or laboratory freezers; liquid nitrogen cryovats; ultracold freezers;	—	87,400*
The Natural History Museum (London, UK)	Molecular Collections	botanical garden or herbarium; natural history museum	1 location (centralized repository)	infrastructure not yet complete	general purpose or laboratory freezers; liquid nitrogen cryovats; ultracold freezers	2,000,000 ¹	
The University of Texas at El Paso (El Paso, TX, USA)	Department of Biological Sciences	university/college	3 locations	N/A	ultracold freezers	3,500	_
Jniversidade de Brasília Brasília, Brazil)	Departamento de Zoologia	university/college	1 location (centralized repository)	infrastructure complete; samples in storage	ultracold freezers	—	20,000*
University of Alaska Museum of the North (Fairbanks, AK, USA)		natural history museum; university/college	1 location (centralized repository)	infrastructure complete; samples in storage	liquid nitrogen cryovats	170,000	100,000
University of Central Oklahoma (Edmond, OK, USA)	Department of Biology	university/college	1 location (centralized repository)	infrastructure not yet complete	ultracold freezers	_	890

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University of Oslo Natural	Natural History	botanical garden or	1 location	infrastructure not	general purpose or	150,000	
History Museum (Oslo, Norway)		,	(centralized repository)	yet complete	laboratory freezers; ultracold freezers; refrigerator (4°C)		
Zoological Institute, Technical University of Braunschweig (Braunschweig, Germany)	Division of Evolutionary Biology		2 locations	N/A	general purpose or laboratory freezers; ultracold freezers	20,000	_

¹Total number of samples based on inventory completed before construction of centralized repository.

²Total number of samples estimated; inventory to be completed as samples moved into centralized repository.

³Total number of samples estimated includes Entomology and Botany samples; samples from Plant Pathology and Nematology not included in estimation.

*Total number of samples unknown, but multiple samples were most often present for each specimen.