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MOLECULAR COLLECTIONS FOR BASIC RESEARCH: MUSEUMS, METHODS, AND MORALITY

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ABSTRACT

Many museums have established genetic resource collections (GRC's). These are collections of preserved tissues, blood, and molecular extracts (e.g., proteins and nucleic acids) that are intended for non-profit research in such fields as evolution, ecology, conservation, wildlife management, toxicology, epidemiology, and forensics. In establishing these collections, museums fulfill a moral imperative to conserve ex-situ as much information as possible about the genetic diversity in our world before it disappears. They also assume the responsibility to make this information accessible to the international, non-profit, research community.

Introduction

The traditional conservatories of ex-situ organismal biodiversity are museums, zoos, and botanical gardens. Until 20 years ago, museum collections consisted of whole specimens, either dried or chemically preserved. With the growth of comparative molecular methods, starting in the 1970s, many museums have assumed the additional role of conserving genetic biodiversity by establishing genetic resource collections or "GRC's" (Dessauer & Hafner, 1984; Dessauer *et al.* 1988, 1996). These collections consist of preserved tissues, blood, and molecular extracts (e.g., nucleic acids and proteins) that are used in genetic research. In establishing GRC's, museums have joined zoos, sperm banks, and type culture collections as genetic resource repositories (Gee, 1984; Ryder & Benirschke, 1984; Ryder *et al.* 2000). More recently, with the recognition of the potential of natural systems, such as rain forests, oceans, and deserts, to produce industrially important chemicals, more traditional collectors and collections have been joined by "bio-prospectors". These are individuals and institutions, such as pharmaceutical companies, interested in exploiting natural genetic products for profit. In doing so, they potentially preserve biodiversity *ex-situ*, while helping justify the preservation of biodiversity *in-situ* (e.g., Hoagland & Rossman, 1997). Finally, in addition to these genetic collections are the collections of pet and plant traders. These

individuals are sometimes interested in genetics (in the sense of breeding), but their fundamental goal is usually to make money.

This paper seeks to identify and elaborate on the role of museum-based GRC's and GRC collectors, to clarify their methods and goals, and to distinguish them from other types of genetic collections and collectors. GRC's are essential to modern research in biodiversity and conservation, and a clear definition of the role of GRC's will help administrators and politicians appreciate their importance. It will also help GRC curators improve the efficiency and service rendered by their collections.

In defining the museum's role in ex-situ preservation of biodiversity, I have highlighted issues in four basic categories: (1) Definition and purpose of museum-based GRC's; (2) Funding of museum-based GRC's; (3) Practical issues pertaining to museum-based GRC's; and (4) improving the effectiveness of museum-based GRC's. I have not produced a complete list of discussion topics, but have emphasized those that seem particularly important at the present time. Several issues will be approached by discussing the experiences and policies of the Collection of Genetic Resources at the Louisiana State University Museum of Natural Sciences (Sheldon & Dittmann, 1997). This collection is the oldest and probably largest museum-based vertebrate GRC. It was founded in 1979 and has its own curator, collection manager, and graduate research assistant. It contains about 50,000 specimens representing 4,500 species of birds, reptiles, amphibians, and mammals from around the world.

Discussion: GRC Management, Methods and Use

1. Definition and purpose of museum-based GRC's

Research purposes.—First and foremost, museum-based GRC's are non-profit repositories of genetic biodiversity. Their over-riding function is to serve as material sources for basic scientific research in genetics. To fulfill this function museums must constantly increase their specimen holdings via general collecting. General collecting serves two basic functions. First, genetic and whole specimens are available for comparative studies in such fields as evolutionary biology, wildlife management, toxicology, epidemiology, and forensics. Second, general collections provide a permanent, tangible, timeless record of an area's biodiversity (Stuebing & Wong, 2000).

Use of GRC's in basic (or pure) research emphasizes evolutionary biology and its constituent fields: systematics, population genetics, ecology, and conservation. Scientists working in these fields typically use GRC's to obtain proteins or nucleic acids, which they compare to one another to reconstruct the genetic histories (genealogies) of populations (phylogeography), species (phylogenetics), or molecules (molecular evolution). Once they understand the genetic history of the taxa or molecules, they consider ecological, behavioral, geographic, paleontological, chemical, or other factors that might have influenced that history.

By combining genealogical and these other data, scientists can reconstruct the evolutionary events that shaped modern of populations, species, or molecules (e.g., Brooks & McLennan, 1991, Harvey & Pagel, 1991). Further insight can be gained by considering the evolutionary histories of several taxa in an ecological community and taking into account modern climate, altitude, competition, predation, and other factors. This community-level approach yields common genealogical and ecological patterns that highlight the processes responsible for the creation of biodiversity. This is a particularly powerful tool for conservationists, because it provides an understanding of the forces that created and sustain biodiversity in a specific area. Thus, it provides the basic information required for preserving or reconstituting that diversity.

Museum collections play a smaller, but still critical, role in many other kinds of basic or non-profit applied research, such as toxicology, epidemiology, forensics, and wildlife management. Examples abound, but two are particularly dramatic. The most famous is the reconstruction of the historical pattern of the effect of DDT pollution on predatory birds from museum specimens (e.g., Peakall & Kiff, 1988; Kiff, 1991). In this case, populations of various species, including birds-of-prey and pelicans, were declining for unknown reasons. Analysis of museum specimens demonstrated a correlation between the introduction of DDT as an insecticide and egg-shell thinning, and the problem was solved by outlawing the use of DDT. A more recent, but equally remarkable example, is the role of museum GRC's in tracing the epidemiology of the Hanta virus. This mysterious virus has killed people in several countries. It was found to be infectious in small rodents, and its epidemiological history was reconstructed by examining hundreds of rodent tissue samples stored in GRC's at American universities (e.g., Baker, 1994). Without general collecting of specimens and tissue samples, these important toxicological and epidemiological patterns could never have been reconstructed. More examples of the usefulness of museum collections in general, and GRC's in particular, can be found in *Systematics Agenda 2000: Charting the Biosphere* (Anon, 1994).

An under-appreciated role of museum GRC's is their storage of basic genetic information for monitoring changes in in-situ genetic diversity. By assembling genetic samples continuously, museum collections can provide basic data on genetic variation of populations over historical time and, thus, a measure of the genetic "health" of those populations. For example, with genetic samples collected before and after an area is disturbed (e.g., by fire, logging, development, pollution, invading species, etc.), we can determine the impact of that disturbance on genetic variation in the area's populations. Such long-term genetic information would be particularly useful in protected areas, such as national parks and wildlife refuges (Stuebing & Wong, 2000).

Maximizing research potential. Museum-based GRC's consist of any material that can be used in comparative genetics. Most commonly, these materials are tissues and blood samples preserved by freezing or other methods. However, GRC's also commonly contain extract from tissues, such as DNA, RNA, proteins, and antisera.

An important function of GRC's is to help scientists locate preserved tissues, bloods, and genetic extracts that they may need in their research. As centralized repositories of genetic materials, GRC's simplify the sample searching process. During the last 20–30 years, many scientists have built personal collections of genetic materials in the course of their individual research. Unfortunately, as scientists' interests change, their personal tissue and extract collections are often relegated to the back of freezers and ultimately lost. This problem is exacerbated by the tendency of some museums not to recognize genetic materials as legitimate museum specimens. Not only must museums recognize the importance of genetic resources as museum specimens, they must be prepared to create collections to house and care for them. If researchers or museums do not have the wherewithal to create GRC's, they should donate genetic specimens to institutions that will take care of them. The advantage of gathering individual genetic collections together into centralized GRC's is that the samples receive proper care and researchers have a better chance of locating and using them.

Traditional museum collections as GRC's. As collecting new specimens becomes more difficult, scientists are turning to traditional museum specimens as sources of genetic information. Using the technology of the polymerase chain reaction (PCR), it is sometimes possible to obtain DNA from traditional museum specimens, especially more recently collected specimens. Such "destructive sampling" is not the optimal way to obtain DNA, as it requires intensive lab work, the DNA is often degraded and incomplete, and the museum specimen is harmed. Also, other types of macromolecules such as RNA and proteins are not preserved in traditional museum specimens. However, the use of traditional museum specimens as potential genetic sources must be recognized, and destructive sampling protocols need to be put in place to regulate use of traditional museum specimens for genetic research (see 3. *Loan policies*).

2. Funding of museum-based GRC's

The expense of GRC's. Genetic resource collections are expensive to equip and maintain. Because of the growth in molecular genetic research, GRC's are rapidly becoming the busiest museum collections, facing the greatest demands, but these pressures and needs are generally not recognized in the museum community. Many museums treat GRC's as part-time pursuits, tangential to their main role of building and curating traditional specimen collections. As a result, museums are not prepared to meet the demands of GRC's: care of small, easily lost and damaged specimens; development of computer databases to permit the tracking of specimens and vouchers; preparation of genetic loans that require finding, sub-sampling, and shipping samples in buffers or on dry ice (by courier); purchase and maintenance of ultracold freezers that break frequently and demand large amounts of electricity, or purchase and maintenance of storage tanks that use large amounts of liquid nitrogen; and development and oversight of destructive sampling policies that balance the needs of the museum with the needs of the research community (see Appendix 1).

As an example of GRC expenses, here is what it costs to run the LSU Collection of Genetic Resources for one year. The LSU museum has a full-time curator (US\$60,000), a collection manager/preparator (\$30,000), a graduate assistant (\$15,000), and an undergraduate student helper (\$5,000). Even with this labor force (\$105,000), we are completely overwhelmed with work, from administration and fund-raising, to preparing specimens, to maintaining equipment, to handling requests for lists of our specimen holdings, to evaluating the validity of tissue requests, to sampling and delivering tissues. In addition to labor, we have invested a great deal of money in equipment: 13 ultracold freezers (\$6,000 each), an alarm system that monitors the freezers (\$20,000), and many incidental pieces of equipment (e.g., liquid nitrogen dewars, standard freezers, generators, and computers). Finally, the supplies used by GRC's are copious and expensive: e.g., nunc tubes, storage boxes, freezer racks, dry ice, etc. We have at least \$50,000 invested in nunc tubes.

The LSU Collection is funded by two sources. LSU provides salaries, housing, and electricity, and the US National Science Foundation (NSF) supplies funds for equipment, equipment repair, and supplies. Fortunately, the NSF recognizes the value of LSU's collection. One measure of this value is the research benefit provided by the Collection. For example, during the last five years, the NSF provided LSU's Collection with \$135,000, and during that same period more than 250 theses and scientific papers were published using tissues from the Collection.

Selling tissues is not a funding option for museum GRC's. An alternative source of funding for GRC's might be to charge a fee for use of the tissues and extracts. But this approach is not acceptable for many reasons, of which here are two: First, because so few tissues are actually used in any period of time, the selling price of tissues would be prohibitive, especially for evolutionary biologists, who are traditionally underfunded. For example, LSU might provide 300 tissues to researchers in a year. It costs LSU about \$110,000 to \$150,000 per year to maintain its collection. The selling price of the tissues would, thus, have to be between \$350–\$500 per tissue sample, plus the cost of collecting, which is usually \$50–\$100 per sample. Second, and more importantly, museum tissues are collected for non-profit research. If they are sold, even at a loss, there is an impression that money is being made through museum collecting. This impression can quickly complicate the already difficult process of obtaining collecting permits. Moreover, the introduction of money into the process raises the issue of profit-sharing with the country of tissue-origin, even when there clearly are no profits to be made.

"Bio-piracy". As should be evident in the previous paragraphs, a clear distinction exists between collecting tissues for basic research and collecting for profit. GRC's are not a source of revenue for museums; they are a revenue drain—expensive to establish, maintain, and use. Moreover, museum collections are intended for use by the entire international non-profit research community. Consequently, scientists who collect for and curate research museums do not make money and are not in the profession for selfish reasons. Thus, they are not "bio-pirates". They are, in fact

serving society's best interests by preserving a record of the biodiversity of our natural world in the face of deforestation and other kinds of environmental degradation. In contrast, collections for the pet, orchid, or any other trades, or "bio-prospecting" for commercial products such as naturally occurring drugs, are profit-making ventures. As such, their potential for abuse of wildlife or unfairness in profit-sharing is substantial, and such for-profit ventures should be treated differently than museum collecting by government agencies.

3. Practical issues pertaining to museum-based GRC's

Curators of museum GRC's face many practical decisions on how to collect, preserve, and care for genetic samples. In making such decisions, they should follow this simple rule: Do whatever maximizes the potential of the collection. For each decision, there will be a proper way and a shortcut. The proper way is almost always more expensive, time-consuming, or difficult, but it produces the best result. The shortcut saves money, time, and labor, but produces a suboptimal result. Every decision must balance these two extremes, and there will be trade-offs. For example, it is certainly better to obtain a genetic sample via a shortcut than not to obtain a sample at all. In this section, I list four examples of important areas in which museum-based GRC collectors and curators must balance trade-offs. In view of the human tendency to take the easiest path, I stress the importance of setting the highest standards in collecting and curating genetic resources.

Field Preservation methods. The proper method for collecting genetic samples of vertebrates is submersion and storage of tissues in liquid nitrogen immediately upon death of the animal. By quick-freezing of tissue, DNA, RNA, and proteins are all preserved. Delay in freezing, or the use of alternative methods (e.g., buffer solutions, alcohol, and drying) reduces the potential use of the tissue. RNA and proteins are much more fragile than DNA and quickly denature or degrade without freezing (Dessauer *et al.* 1996). Unfortunately, because current research emphasizes PCR amplification and sequencing of DNA, and this methodology does not require frozen samples, many collectors have abandoned the use of liquid nitrogen in favor of other methods. This abandonment is understandable, as liquid nitrogen requires an expensive tank, is difficult to obtain in remote areas, upsets airline bureaucrats, and leads to excess baggage costs. However, by failing to use liquid nitrogen, collectors sacrifice the long-term potential of their samples for short-term convenience. RNA is essential for many types of molecular analyses and will become increasingly important for rapid sequencing of nuclear-genes (D. Pollock, pers. comm.). Similarly, new shotgun methods are being developed for rapid sequencing of entire mitochondrial DNAs. But, these methods require that mtDNAs be purified by ultracentrifugation, and the success of mtDNA by this purification method depends upon the preservation of circular mtDNA by freezing (Dowling *et al.* 1996). Almost certainly, failure to preserve tissues quickly in liquid nitrogen will obviate many analytical methods that have not yet been developed.

Museum preservation methods. The best method for storing tissues is in liquid nitrogen. This keeps the tissues much colder (-196° C) than ultracold freezers (*ca.*

-70° C) and is more likely to preserve fragile molecules, like RNA and proteins. Additional benefits of liquid nitrogen storage are that the tanks do not have mechanical parts and, thus, are less susceptible to breakdown than freezers with vulnerable compressors. Also, liquid nitrogen storage tanks do not rely on electricity, a great advantage in areas where the supply of electricity is unreliable. The major drawbacks to liquid nitrogen storage are the cost of liquid nitrogen, and their constrained storage space relative to freezers.

Voucher specimens. Methods of comparative molecular research have changed dramatically in the last 20 years, with substantial ramifications for museum collections. Before the advent of PCR amplification of DNA, large samples of tissues or blood were required to provide an adequate amount of DNA or protein for comparative analyses. Nowadays, small amounts of tissue or blood can provide DNA for a large number of molecular comparisons. This technological advance simplifies collection and storage of samples. It also makes possible the collection of DNA samples from living organisms, creating tremendous research advantages. For example, live-sampling permits genetics to be studied simultaneously with ecology and behavior (e.g., Westneat & Webster, 1994), and it allows threatened or endangered populations to be sampled without jeopardy (e.g., Zhi *et al.* 1996; Ryder *et al.* 2000).

However, live-sampling cannot replace the collection of traditional museum specimens, without which many scientific opportunities are squandered. Such lost opportunities include: (1) analyses of size, shape, development, diet, metabolism, sex, breeding condition, molt, life-cycles, and age of organisms; (2) analyses of distributional and ecological variation through time, based on specimen archives; and (3) follow-up studies of morphology inspired by discoveries made through genetic analysis of tissues. Apart from lost potential, failure to collect traditional specimens inevitably results in identification problems. On countless occasions, genetic analysis has led to anomalous results that call into question the field identification of the specimen. If a voucher specimen exists, the question of identity can be solved. Otherwise the genetic sample is useless and the research is compromised. Except in cases of on-going field studies or work on threatened or endangered species, traditional museum specimens should always be collected with genetic samples.

Loan policies. With the growth in molecular genetic analyses, GRC's are under tremendous pressure to provide tissues and extracts for researchers. This is a positive development, illustrating the important role assumed by GRC's. However, increased use creates competition, and competition, in turn, creates the need for a policy to guide GRC-management decisions. For example, is free and wide access to tissues, in itself, proper management of the collection? After all, the tissues can be used up. What if entire tissue samples were given to an unproductive researcher who never publishes his results, when they could have been given, at a later time, to a productive researcher doing particularly interesting science? And, should tissues be given to researchers who do not believe in scientific collecting, or are too

lazy to collect their own samples? Would it not be better to give tissues to collectors, rather than non-collectors, as an incentive to encourage further collecting, so that our natural world becomes better documented? And, what if two people want to work on the same tissues? And what if a well-financed researcher, with lots of equipment and technical assistance, is using a disproportionate amount of tissue, thereby restricting more poorly financed (slower-producing) researchers to a smaller proportion of the collection?

GRC curators wrestle with these kinds of questions constantly. Rather than try to answer them, as there are no absolute answers, I have included the Tissue Grant Policy of the LSU Museum as Appendix 1. This policy describes the main issues and how one museum deals with them. It is not the final word, but only a step in the process of balancing conflicting needs in a world where research directions change constantly.

4. Improving the effectiveness of museum-based GRC's

If we consider the basic purposes of museum-based GRC's, we can easily identify ways to improve the system. The purposes are (1) to conserve ex-situ as much of the world's genetic biodiversity as possible, without permanently harming in-situ biodiversity, and (2) to make this genetic resource available to as many researchers as possible. From these purposes, two fundamental needs are clear: (1) improved communication among collections and scientists as to what is stored in GRC's and (2) more coordinated efforts to obtain from the wild what is not stored in GRC's.

Improving communication. In the 1980s, during the inception of GRC's, researchers attempted to improve communication by holding symposiums (e.g., Dessauer & Hafner, 1984), and this approach is still an important method for idea exchange (e.g., Hoagland & Rossman, 1997; this symposium). More recently, electronic media have been brought to bear. A web site is planned at the San Diego Zoo, which with its "Frozen Zoo" was an early force in conserving the genetic diversity of endangered species (Ryder & Benirschke, 1984; Ryder *et al.* 2000). For information on that web site, email DNA_banks@sandiegozoo.org. At LSU, we have started a listserv called GENCOL, which is a medium for exchanging ideas about the management, loan policies, collecting, storage, permits, publicity, and other issues pertaining to all kinds of GRC's. GENCOL is open to all researchers, collection managers, government officials, and anyone else who is interested in GRC's. To join GENCOL send the following message to LISTSERV@LISTSERV.LSU.EDU. "subscribe GENCOL-L [your email address] [your name]". (Do not include quotes or brackets.) Alternatively, you can simply send an email to me (fsheld@lsu.edu). Finally, movement is afoot to link collection computer-databases at institutions having GRC's. Such links would simplify the process of locating genetic samples and ultimately save labor, as inquiries into collection holdings would be automatic rather than served by a collection manager. These links would also provide a simple way to assess the strengths and weaknesses of our GRC holdings.

Communication among GRC's should also improve communication among researchers. The need and advantages of improved communication among researchers is evident from the following examples. At present, at least five museum groups in four countries are attempting to reconstruct the phylogeny of babblers (Aves: Timaliidae). In some cases, these researchers have no idea that other groups are sequencing the DNA of the same species. (I know of the situation because these researchers have all requested babbler tissues from the LSU Collection.) Not only does this lack of communication foment unnecessary competition among researchers, but it is an inefficient use of genetic resources, funding, and intellectual energy. There are enough interesting questions in babbler systematics for all these researchers to work simultaneously and synergistically. A dramatic example of coordination in molecular systematics is the "Deep Green" program in plant phylogenetics (<http://ucjeps.berkeley.edu/bryolab/greenplantpage.html>). Dozens of researchers are working together to reconstruct the phylogeny of plant life. Each researcher or research group is responsible for a different branch or set of genes. Two spectacular products of this collaboration to date are Chase *et al.* (1993), with 42 authors, and Angiosperm Phylogeny Group (1998), with 26 authors.

Coordinating collecting efforts. Museums need more specimens from every area on earth. However, because of funding limitations, collecting efforts must emphasize underrepresented areas. Given good communication among collections, researchers could direct their efforts to these underrepresented areas. For example, the five groups working on babbler phylogenetics could identify the existing museum holdings of babbler samples, and then direct their collecting efforts to areas where the least amount of money would produce the most specimens.

Conclusion: Conservation, Morality and GRC's

We have an obligation to preserve as much of our natural heritage as possible before it disappears. This preservation process has been divided into two parts: *in-situ* preservation, which maximizes biodiversity through the development of parks and protected lands; and *ex-situ* preservation, which maximizes information on biodiversity through the collection of specimens and establishment of genetic banks and breeding populations. As emphasized in this and other papers in this symposium, *in-situ* preservation efforts cannot succeed without the perspective afforded by research on *ex-situ* specimens. Conversely, there is no way that *ex-situ* efforts can possibly document even a small proportion of the world's biodiversity before it disappears, unless there are permanent *in-situ* preserves protecting that diversity.

Efforts by conservationists to preserve biodiversity rely on a set of moral imperatives. That is, each type of conservationist—ecologist, taxonomist, park manager, wildlife manager, museum curator, lobbyist, politician, etc.—has an obligation to produce the most for his or her effort. For museum collectors, the objective is to obtain the maximum amount of information on biodiversity as possible. For the genetic resource collector, this objective can be expressed as a set of nested imperatives:

- I. Document as much biodiversity as possible.
- IA. Collect as widely as possible, without permanently harming populations.
- IA1 Collect each specimen from each individual organism in a way to maximize its usefulness.
- IA1a. Preserve as much data as possible from each specimen, including: exact collecting location, date, preservation time, habitat, altitude, breeding condition, soft-part colors, fat content, age information, molt condition, etc.
- IA1b. Preserve as museum specimens as much information as possible from each individual organism that is collected, including: skin, skeleton, dried parts, stomach contents, vocal recordings, photographs, videotapes, and genetic samples.
- IA1bi. For genetic specimens, obtain different tissues (blood, liver, muscle, heart, brain, etc.). Preserve them immediately upon the death of the organisms in liquid nitrogen.

The imperatives for the GRC curator are:

- I. Provide care for each specimen, such that its usefulness is maximized.
- IA. Store the specimen in liquid nitrogen if possible.
- IB. Catalog the specimen using an electronic database method to facilitate retrieval of data associated with it, including voucher information.
- II. Facilitate use of the specimen by the research community.
- IIA. Communicate with other collections, so that the research community knows that the specimen exists.
- IIB. Develop a clear set of guidelines for specimen use.

The imperatives for the scientist using the genetic resource collection are:

- I. Make maximum use of each specimen; it is a limited resource.
- II. Inform the GRC curator of research results, so that he or she can include the discoveries in the specimen database. Such cross-referencing is essential to achieve the maximum potential of the specimen.
- III. Acknowledge the contribution of the collector and the GRC in publications. Without such acknowledgment, the important role of collectors and GRC's in the scientific process is not appreciated, and their funding and public support will remain limited.
- IV. Return genetic extracts or unused samples to the GRC. Otherwise they may be lost to the research community.

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APPENDIX 1

Grant Policy and Procedures: Collection of Genetic Resources, Museum of Natural Science, Louisiana State University, Baton Rouge, Louisiana, USA.

The Collection of Genetic Resources at the Louisiana State University Museum of Natural Science (LSUMNS) consists of preserved tissues and tissue extracts of vertebrates. The Collection is a centralized repository, and we encourage its use by the international, non-profit, research community. This document describes procedures for obtaining tissues from LSUMNS and the reasons for those procedures.

Rationale of the Tissue Grant Policy

The policy governing the use of LSUMNS tissues is intended to provide fair access to the Collection. It is based on the following observations:

1. *Tissue collections differ from traditional museum collections.* Unlike most museum specimens, tissues are consumed by researchers. Thus, tissue "loans" are in fact grants of a limited resource. As a result, we treat tissue requests as we would grant applications, and our grant policy takes steps to prevent depletion of the Collection.
2. *The LSU Collection consists mainly of specimens collected by LSU researchers, but also specimens donated by researchers at other institutions.* In general, specimens collected by LSU personnel are intended for use by LSU researchers and their collaborators, although we commonly grant use of these tissues to non-LSU researchers. Donated specimens are for general community use, except when restricted by the donor.
3. *Tissues are expensive, difficult, time-consuming, and sometimes hazardous to collect.* Tissue samples are the limiting resource in molecular studies, yet the importance and difficulty of collecting tissues (and specimens in general) is not widely acknowledged in the scientific community.
4. *Tissue collections are expensive to maintain.* LSUMNS provides its collection with a full-time curator, half-time collection manager, half-time specimen preparator, part-time curatorial assistant, and substantial facilities (including 13 ultracold freezers monitored by a sophisticated alarm system). LSUMNS also has received funds from the NSF to computerize the Collection and purchase hardware for storage and maintenance. This level of support enables LSUMNS to care for a large number of specimens, to provide computerized lists of specimens to researchers, and to find, sample, and ship specimens efficiently.
5. *The value of tissues depends upon associated data.* Tissues without voucher specimens and collection data have limited value, because there is no way to

verify identification or perform complementary studies in anatomy, geographic variation, etc. We obtain vouchers for all samples we collect.

6. *The LSUMNS is interested in the fate of the tissues it grants.* We keep careful records of how tissues are used. This information may be required to fulfill obligations to the country of specimen origin, and it is useful for grant proposals.
7. *Tissue use may be governed by USDA and USF&WS rules:* The LSUMNS Collection operates in strict accordance with all relevant laws, rules, and regulations.

Tissue Grant Policy

Given the observations above, LSUMNS has set the following guidelines for granting tissue requests.

1. *Preference is given to researchers who collect specimens.* LSUMNS will not provide the majority of tissues for a project; we expect researchers to collect most of their specimens.
2. *Preference is given to foreign researchers seeking tissues from their own country.*
3. *Grantees must provide some evidence of reciprocal benefit to LSUMNS.* Examples of reciprocal benefit include: tissues offered in exchange for LSUMNS tissues; access for LSU researchers to substantial tissue holdings; help organizing collecting expeditions; funding for collecting expeditions. Reciprocal benefit is assumed for any foreign researcher requesting tissues of organisms from his/her own country.
4. *Preference is given to quality research.* Quality is judged by an LSUMNS committee (described below). The grantee must be a scientist who is likely to publish the results of research using LSUMNS tissues.
5. *Grants will be denied to researchers who have not made good use of samples in the past or who have not fulfilled grant requirements.*
6. *Grants of specimens may depend upon volume of tissue and rarity of taxon.*
7. *Grants are given only to researchers who agree to the following requests or obligations:*
 - a. Grantees *must* return unused tissues. Tissues may not be given to third parties without the permission of LSUMNS. This rule ensures maximal use

of tissues, prevents loss, prevents legal infractions, and provides a paper-trail.

- b. Grantees are *requested* to return a portion of the extract derived from granted samples (e.g., DNA). At present, few institutions are able to curate molecular extracts adequately, and once a study is completed extracts are usually lost. This provision is intended to prevent such losses and to increase extract availability to the research community. Extracts cannot be distributed to other researchers without LSUMNS permission.
- c. Grantees *must* acknowledge the “Louisiana State University Museum of Natural Sciences Collection of Genetic Resources” in all publications that use data generated from LSUMNS samples. Publications should include a table that lists tissue number, voucher number, and collector of each specimen. Such lists acknowledge the proportional contribution of the Collection and collector. Researchers also may need to acknowledge other institutions and grant numbers for samples processed through the LSUMNS Collection.
- d. Grantees *must* send a reprint of any publication benefiting from an LSUMNS tissue grant. It would be helpful if the grantee emailed the publication citation to the Curator or Collection Manager.
- e. Grantees *must* obtain, and bear costs associated with, permits to transport and house LSUMNS tissue samples. A USDA Transport Permit is required to receive imported bird and mammal tissues. A CITES Institutional permit is required to receive samples of CITES-listed Appendix I, II, or III taxa.
- f. Donated or exchanged tissues *must* be accompanied by either by a museum invoice with accession numbers or copies of appropriate permits (e.g., a U.S. collecting permit or a country-of-origin export permit).
- g. Researchers are responsible for shipping costs of tissue samples. This requirement includes most material returned or donated to LSUMNS. Exceptions will be made, however, for large donations and special cases.

Procedures for Requesting Tissues

Grant proposals to the LSUMNS Collection usually proceed in two steps. First, a list of specimens is requested from the Curator or Collection Manager. This request may be made by phone, email, or regular mail (email is preferred). Second, specific samples are requested in writing on letterhead from the Curator of Genetic Resources. Graduate students should submit a letter co-signed by their advisor, who will assume responsibility for use of the samples. If the tissue grant is approved, the letter represents a contract between the researcher and the LSUMNS.

The request will be evaluated by a committee consisting of the Curator of Genetic Resources, the curator of the vertebrate group in question, and the Manager of the Genetic Resources Collection. If the tissues have been donated to the Collection by a non-LSU researcher who wishes to be involved in the granting decision, we also obtain his/her opinion.

The proposal should provide the following information:

1. *A brief outline of the goals, methods, and time-frame of the project, justifying the use of the samples.*
2. *The total number of tissues to be used in the project, in addition to LSUMNS's contribution. Please specify the number of tissues that will be collected by the researcher and the number requested from other institutions.*
3. *A statement of reciprocal benefit.*
4. *A list of the LSUMNS specimens by name and number. Please specify the amount of tissue/extract required and the preferred method of delivery (e.g., on dry ice, in 95% EtOH, or in DMSO buffer).*
5. *A Federal Express (or comparable) charge number.*
6. *Copies of USDA permits (if required).*