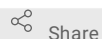


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Sample Collection: Primate Feces for DNA/RNA

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1 Works for me



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ABSTRACT

All of the methods and protocols for this project have been reviewed and approved by the Otterbein University Institutional Animal Care and Use Committee, Otterbein University's Environmental Health and Safety Officer, the AZA Prosimian Taxonomic Advisory Group, and the individual collaborating AZA Institutions.

The pygmy slow loris (*Nycticebus pygmaeus*) captive population is declining at a rate that jeopardizes their future in the Species Survival Plan (SSP). Experts agree that diet is somehow compromising reproduction, and this study will investigate dietary estrogenicity. Many plants produce chemicals known as phytoestrogens that activate estrogen receptors. The phytoestrogen-fertility relationship is well-documented in livestock, rhinoceroses, cheetahs, and pandas, and this study will characterize the pattern in slow lorises. We will adapt existing biomedical methods to investigate dietary estrogens and reproductive outcomes in captive pygmy slow lorises. Gene expression profiles will serve as proxies for in vivo estrogen receptor activity, and our heterologous cell-based receptor reporter assay will quantify in vitro estrogen receptor activation. Generalized linear mixed-effects models will integrate these parameters with reproductive outcomes to test the effect of gene expression profiles and diet estrogenicity on fertility. This long-term project will ultimately provide one of the first deep physiological examinations of phytoestrogen's proximate and ultimate influences on primate reproduction.

PROTOCOL CITATION

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<https://protocols.io/view/sample-collection-primate-feces-for-dna-rna-bq3bmyin>

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GUIDELINES

Thank you in advance for agreeing to participate in this project on Pygmy Slow Lorises, Dietary Estrogens, and Reproductive Function. This study aims to identify endocrine-active compounds in pygmy slow loris foods, which may be contributing to reproductive complications in captivity. We know that everyone helping us collect these samples is already incredibly busy, and truly appreciate your contribution to what we hope will benefit zoos and the lorises.

If you have any questions about this protocol or the long-term study, or you would like to inquire about more formal collaboration on the project, please contact the principal investigator, Dr. Alicia Rich, an Assistant Professor at Otterbein University (rich2@otterbein.edu).

MATERIALS TEXT

PPE:

- Facial covering
- Nitrile examination gloves

Sample Storage:

- Sterile RNase-free tubes prefilled with stabilization solution.

 DNA/RNA Shield Zymo

- **Research Catalog #R1100-50** Step 3

SAFETY WARNINGS

Follow all of your standard procedures at your institution for handling animal feces.

DISCLAIMER:

Monitor subjects during any and all sample collection procedures for signs of distress. For example, slow lorises emit a distinctive “chitter” vocalization when they are in distress. If collection causes an animal to repeatedly exhibit signs of distress, immediately suspend the protocol and reach out to the PI on this project to reassess the methods. Do not collect any samples from an animal if it will risk aggravating any existing wound, illness, or other condition. Let the PI know that you will need to exclude that animal from the study.

BEFORE STARTING

We will provide all of the necessary materials for sample collection. Make sure that nothing is missing from your kit and you understand these guidelines.

Sample Collection

- 1 Gather fresh feces (< 1 hour old) from an animal as soon as possible. Make sure you are certain which animal deposited the sample and that the sample has not been contaminated with other material. We prefer that you gather the feces as close as possible to the hair samples (on the same day is best), but we realize this may not be possible.

If it is not possible to gather fresh feces indirectly from a reliably identified individual, then follow the rectal swab directions in the direct sample collection procedure.



Always wear a facial covering and nitrile examination gloves while collecting samples.

- 2 Label one of the sterile tubes provided (pre-filled with Zymo® DNA/RNA Shield) with your initials, the date (YYYYMMDD), the time (HH:MM:SS), animal code (PSL for pygmy slow loris), sample type (feces), and animal name or ID. Open the tube but take care not to touch the inside of the tube, lid, or the outside of the attached scoop to your glove or any other surface. Use the scoop to gather ~0.5-1 ml of feces and deposit it into the tube.

- 3  DNA/RNA Shield Zymo

Make sure there is just enough **Research Catalog #R1100-50** to submerge the entire sample in the tube. If there is too much sample for one tube and you have an extra, then you can fill a secondary tube and label it as such. We will use matched tubes like this for validation in the lab.

Close the lid as tightly as possible and use parafilm (included) to reinforce the outside of the closure.

Sample Storage

- 4 Place this tube inside of a clearly labeled sample bag (also provided), and put that into the envelope with postage, already addressed to Alicia Rich at Otterbein University. Please drop this envelope in the mail as soon as possible (within 24-48 hours of sample collection).



Remove your gloves inside out and safely dispose of them in the proper area. Always wash your hands thoroughly after collecting a specimen.