Guano Collection for Genetics Protocol (Adapted from Northern Arizona University, College of Engineering,

Forestry and Natural Sciences, School of Forestry and Center for Microbial Genetics and Genomics)

Required Materials:

- RNAlater[™] solution (available upon request)
- 15mL conical tubes (mass collection) or 1.5mL tube (single species (1-3 pellets)
- Nitrile gloves for sample collection
- Permanent marker for labeling
- Extra Ziploc bags (quart or gallon)
- Trash bag/bin for storing discarded gloves and contaminated
- Cooler with ice for sample storage in the field (if possible)
- Tweezers (optional)

Avoid cross-contamination between samples:

- Use new gloves, tweezers, scoops when collecting a new sample or moving to a new site
- Do not set collection equipment down on any non-sterile surfaces
- Use Ziplock bags to separate samples from different sites
 Please report any potential cross-contamination that could have occurred

Sampling Protocol

- 1. Open tube, making sure the cap does not touch anything. Either hold the cap between two unused fingers or have another gloved person hold it. Avoid setting it down.
- 2. With clean gloves on, collect the **freshest** looking guano and place in the 15mL (or 1.5ml) tube. Freshly deposited guano contains the highest quantities of target DNA. **Carefully search for fresh guano deposits for collection**.
- 3. Fill tube with 1 part guano to 5 or more parts RNALater. Do not overfill tube.
- 4. Close tube tightly.
- 5. Invert the tube several times to submerge all contents with RNAlater solution and to test its seal for shipment.
- 6. Clearly write a unique label with a permanent marker on the tube. If there are multiple replicates from the same sample, ensure that all samples are given a replicate ID (A, B, C or 1, 2, 3, etc.).
- 7. Place tubes from same collection location within a ziplock.
- 8. Store collections in a sample cooler if possible OR immediately refrigerate upon returning from field sampling.
- 9. Fill out the <u>sample submission form</u> and follow the mailing instructions listed in the Shipping Protocol found at genidags.com

If collecting wing punches:

- 1. While wearing gloves, use sterile tweezers to transfer wing punch to the 1.5mL tube.
- 2. Ensure that the tissue is submerged in the RNAlater[™] (1 part tissue to 5 or more parts RNALater[™]).
- 3. Place only one tissue per tube.
- 4. Use new gloves/tweezers between each sample. Tweezers can be cleaned with 10%bleach and rinsed with DNA-free water between samples.
- 5. Ice samples in the field if possible.