2023 BLOOD SAMPLING PROTOCOL

The Bird Genoscape Project is in the process of building genoscapes for several species of concern and need blood samples from the following taxa:

- American Robin (AMRO)
- Western Grebe (WEGR)
- Kentucky Warbler (KEWA)
- Western Tanager (WETA)
- Loggerhead Shrike (LOSH)
- Yellow-billed Cuckoo (YBCU)

Sampling Kits – The following items are included in the sampling kit:
- Cryotubes prefilled with ~300μL of blood lysis buffer
- 26G ½” Needles
- A box of 75μL microcapillary tubes (non-heparinized*) and bulb dispenser
  * Heparin is a PCR-inhibitor so please only use non-heparinized tubes
- Alcohol wipes
- Cotton swabs

Specific Procedure for Brachial Venipuncture
1. Separate feathers and clean the area on the underside of the wing using an alcohol wipe.
2. Locate brachial vein.
3. Remove the cap from the needle.
4. Approach the brachial vein with the needle from a 45° angle. A light prick should be adequate to generate enough blood.
5. Once a drop of blood has formed, lightly touch the end of the capillary tube to the drop. If the blood is not flowing easily, rotate the bird to a vertical position so that gravity helps the flow of blood into the capillary tube.
6. Stop collecting blood when the tube is 3/4 full (~50μL).
7. Place pressure on punctured area with clean, dry cotton swab until bleeding has ceased.
8. Empty contents of capillary tube into lysis buffer tubes using the bulb dispenser and shake the blood-lysis mixture well.
9. Label all tubes per labeling instructions below.

Tube Labeling - Please make sure each tube contains the following information clearly printed.
- Band number
- Species code
- Date
- Location

Metadata
Please email the metadata for all the samples as an excel file to Christine Rayne (cerayne@rams.colostate.edu). See preferred template below:

<table>
<thead>
<tr>
<th>Species</th>
<th>Band #</th>
<th>Date Collected</th>
<th>Recapture</th>
<th>Sex</th>
<th>Age</th>
<th>Country</th>
<th>State</th>
<th>Near Town</th>
<th>Lat</th>
<th>Long</th>
<th>Sample Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>American Robin</td>
<td>XXXX-XXXX</td>
<td>dd/mm/yyyy</td>
<td>Y or N</td>
<td>M, F, or U</td>
<td>If known</td>
<td>USA</td>
<td>MD</td>
<td>Silver Spring</td>
<td>39.01</td>
<td>77.02</td>
<td>Blood &amp; Feather</td>
</tr>
</tbody>
</table>

**Sample Storage**

Blood samples should be stored in **a cool dark space at room temperature**. (Blood samples can be stored at room temperature for up to 6 months per Seutin et.al. 1991.) **Please do NOT freeze** samples as we have found that it lowers the final DNA yield.

**Sample Shipment**

When you’re ready to mail your samples, please email Jacob Job (birdgenoscape@gmail.com) with the following information and she will email you a prepaid FedEx or UPS label:

- Approximate weight of package
- Preference for UPS or FedEx label
- Return address
- Contact number + email

If using your own label, please mail the samples to the following address:

ATTN: Jacob Job  
Colorado State University, Biology  
200 W. Lake Street  
Campus Delivery 1878  
Fort Collins, CO 80523

**Copies of the following Permits must be included in the shipment**

For domestic shipments **of samples collected within the USA**, please include:
1. A copy of your (the collector’s) federal bird banding permit
2. A copy of your (the collector’s) state scientific collecting permit
3. A copy of the importer’s USDA domestic transport permit #138961*

For domestic shipments **of samples previously imported to the USA**, please include:
1. A copy of your (the collector’s) federal bird banding permit
2. A copy of your (the collector’s) state scientific collecting permit
3. A copy of the importer’s USDA domestic transport permit #138961*

For shipments from **outside of the USA** please include:
1. A copy of the importer’s USDA international transport permit #138972*
2. A copy of the importer’s USFWS Migratory Bird import/export permit*
3. A copy of your (the collector’s) federal bird banding permit
4. A copy of your (the collector’s) provincial scientific collecting permit (for Canada)
5. USFWS Form 3-177 (Declaration of importation or exportation of fish or wildlife) – can also be filled online at https://edecs.fws.gov/
6. FedEx Declaration of Biological Shipments
7. FedEx Commercial Invoice
8. CITES export permit as necessary
   *Forms provided by the Bird Genoscape Project

Thank you very much for your participation in The Bird Genoscape Project! If you would like to collaborate on a particular species or learn more about the current sample distribution for our target taxa, please visit our website:

www.birdgenoscape.org
SAMPLE DISTRIBUTION MAP

The following maps show the current sample distribution for our target species.

**American Robin**

**Canada Warbler**

**Kentucky Warbler**

**Western Tanager**