Wildlife Health Protocol -- Genetic Sampling: Blood Card Collection 04.28.11

I. Goal: Blot whole blood onto card for DNA preservation.

II. Previous Training Required: Only personnel trained in venipuncture, syringe and needle handling, and wildlife behavior during blood collection should use this protocol for live animals.

III. Items you will need:
- 5ml or larger syringe with 16 or 18 gauge needle (1 set per animal)
- Whatman DNA blood card (1 per animal)
- Sample card labeling (e.g.sharpie or preprinted label)
- Sharps disposal container
- 3 or 4 clipboards
- Large plastic bag or cardboard box

IV. Procedures
1. Before bleeding, open and clip 2 cards onto clipboard such that the 4 black circles are exposed for blood application.
2. Ensure that animal is properly restrained, without restriction to the airway, and other personnel are aware of your sampling. Venipuncture frequently causes live animals to respond and potentially injure unaware personnel.
3. Collect at least 5 ml blood in excess of other needs using proper venipuncture technique.
4. Either:
   a. Carefully carry the syringe and needle to the DNA card area; or
   b. Recap the needle using proper technique and hand the sample to designated personnel for blood card collection. If sufficient personnel are available, this is the preferred technique.
5. Hold the syringe and needle at a 25 - 45 degree angle, bevel down, with the needle touching the bloodcard inside a circle.
6. Using your index finger, gently press on the syringe plunger to slowly express a small amount of blood onto the paper inside a circle with slow circular movements.
7. Move from circle to circle and fill all four circles. It is OK to go outside the circles. Fill enough to penetrate through the preservative paper, but don’t over saturate the paper or it won’t dry well.
8. If blood clots or becomes difficult to express through the needle:
   a. Carefully squirt a tiny amount of blood into a trash receptacle to clear the needle, retaining as much blood as possible, or insert the needle into a vacuum tube to pull the clot through and clear the needle if you have sufficient blood for both the tube and blood card. Don’t force the syringe plunger!
   b. If neither technique works, carefully remove the needle and continue applying blood from the tip of the syringe.
9. CHECK the back of the card to confirm that blood has soaked through the preservative paper.
10. Properly label the card.
11. After 2 cards are filled, place clipboard out of rain and direct sunlight to allow cards to dry. Check that cards are dry on the back side of the preservative paper before storing. Once dry, fold the card and place in large Ziploc or other storage container out of direct sun/rain.

Tips:
- If preservative paper is stuck to backing paper, the card will not dry quickly; position card to allow air to circulate to both surfaces of preservative paper.
• If you have difficulty getting cards to dry (humid or rainy day), use more clipboards. You can also fold partially dried cards up and then set them out indoors on a large table to dry overnight after the day’s sampling is complete.