Blood collection for diagnostic testing can be a challenge in exotic companion mammals, especially in smaller species. However, today’s technological advancements allow us to gain valuable information even with limited sample volume. In all cases, sample quality and minimal volume requirements must be taken into consideration.

**Laboratory Analysis**

**The Reference Laboratory**

Talk to your reference laboratory of choice to determine:

- Type of sample required for the desired test (whole blood versus plasma/serum)
- Minimum required volume.

In addition, preparation and sample storage are key to obtaining accurate results:

- **Serum** (separated from clotted blood) and **plasma** (separated from unclotted blood) samples may need to be kept frozen until ready for submission.
- **Anticoagulated whole blood** should be mixed with the proper type and volume of anticoagulant indicated for the test being run.
- **Whole blood samples** not sent out immediately should be refrigerated.
- **Blood smears** should be prepared immediately after the sample has been collected to minimize cell deterioration.

The anticoagulant of choice for a complete blood count (CBC) is ethylenediamine tetra-acetic acid (EDTA) because it best preserves the cellular components of blood and prevents platelet aggregation.

### In-house Analysis

Samples can also be run in-house, utilizing equipment, such as blood chemistry analyzers and microscopes.

<table>
<thead>
<tr>
<th>Circulating Blood Collected</th>
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<tbody>
<tr>
<td>7.5%</td>
<td>1 week</td>
</tr>
<tr>
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</tr>
<tr>
<td>15%</td>
<td>4 weeks</td>
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*Courtesy Iowa State University Institutional Animal Care and Use Committee*

**Facts at Your Fingertips**

- **Circulating blood volume; loss,**
  - 15% to 20%: Reduces cardiac output and blood pressure
  - 30% to 40%: Induces life threatening shock

- **Plasma:** Separated from unclotted blood
- **Serum:** Separated from clotted blood
- **Total Blood Volume:** In mammalian species, approximately 6% to 8% of body weight
- **Maximum Volume of Blood to Collect:** 10% of total blood volume

**Table 1. Approximate Recovery Times: Normal Packed Cell Volume Restoration After Sample Collection**

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*Courtesy Iowa State University Institutional Animal Care and Use Committee*
Our preference is the Vetscan analyzer (abaxis.com), which has the ability to produce and process a chemistry panel with only 0.15 mL of whole blood, serum, or plasma. This feature is extremely beneficial when taking samples from very small patients.

A single drop of blood collected in a heparinized hematocrit tube is enough to process a modified in-house CBC, which includes:

- Hematocrit
- Total serum solids
- Differential and estimated white blood cell (WBC) count
- Analysis of both red blood cell (RBC) and WBC morphology and characteristics.

**BLOOD: TOTAL & COLLECTION VOLUMES**

**Determining Volumes**

The volume of blood that can be safely removed from a patient without causing hypovolemia or other physiological stress depends on circulating blood volume.

- Carefully weigh each patient and calculate total blood volume: Total blood volume can vary among mammalian species, but in general is approximately 6% to 8% of the body weight.
- The volume of blood that can then be collected is 10% of the total blood volume.
- Therefore, a 100-gm patient has a blood volume of 6 to 8 mL and a maximum collection volume of 0.8 mL.
- Adjustments should be made for clinical condition; for example, collect smaller samples in patients that are hypovolemic or anemic.
- Regardless of the maximum amount available for collection, collect only what is required for testing.

In many mammals, restoration of circulating fluid volume after sample collection usually occurs within 24 hours; however, complete return to normal hematocrit and RBC volume may take 1 to 4 weeks, depending on the volume collected (Table 1). \(^1\)

**Potential Risks**

If too much blood is drawn too quickly without replacement, hypovolemic shock may develop. In many mammalian species:

- Loss of 15% to 20% of circulating blood volume will reduce cardiac output and blood pressure.
- Loss of 30% to 40% of circulating blood volume will induce life threatening shock.

**PRIOR TO COLLECTION**

**Sedation, Anesthesia, & Analgesia**

In small patients, safe and effective restraint for venipuncture is potentially dangerous due to patient movement and becomes increasingly difficult as patient size decreases. For these reasons, sedation or anesthesia may be the key to success.

**Sedation & Anesthesia.** In most cases, sedation is adequate and avoids the increased risk of general anesthesia.

- Keep in mind that inhalant anesthetics can produce artifacts in the CBCs of some species, such as lowering temporarily, but significantly, the hematocrit of a ferret.
- Various sedation protocols and methods can be considered depending on a number of factors, including overall patient condition.
- Ideally, a patient should be fasted for several hours before collecting blood if anesthesia is used. Even in patients that do not vomit, a temporary fast reduces the amount of food in the oral cavity, which could lead to aspiration during anesthesia.

**Analgesia.** The use of topical analgesia increases comfort and reduces patient movement at the moment of venipuncture.

- Safe dosages of lidocaine have not been determined in all species, but we have found 1 to 2 mg/kg to be safe and effective.

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**Figure 1.** Auricular vessels of the rabbit (A); any accessible vessel can be used for venipuncture. Collection of blood from the central auricular vessel in a rabbit using a 25-gauge needle and 1-mL syringe (B).

**Figure 2.** Venipuncture in a rabbit using the lateral saphenous vein. The vein is located laterally above the hock (A). The vessel is accessed using a 25-gauge needle and 1-mL syringe (B).
• Lidocaine can be used as a gel or injected by:
  - Rolling the skin away from the puncture site
  - Injecting the lidocaine
  - Allowing the skin to roll back into normal position.
• Wait 2 to 3 minutes before proceeding with venipuncture.

Choosing a Collection Site
The sample site chosen is based on experience and preference. Limiting factors include the size of the patient and ability to properly restrain. The correct site is the one that produces a high-quality sample without patient injury (Table 2).

Ferrets. Most ferrets can be safely restrained manually for blood collection by scruffing and stretching. An alternative is rolling the patient in a towel with the head and neck exposed.
• In our opinion, the ideal site for collection is the cranial vena cava, which is particularly safe in the ferret due to the distance between the manubrium and the heart. Follow the steps outlined in Notes on Collection from the Cranial Vena Cava (page 76), using a 25-gauge needle and a 1- to 3-mL syringe.
  • Other collection sites include the jugular vein and ventral tail vein, which is accessed using a 25-gauge needle and 1-mL syringe directed dorsally from the underside of the tail until bone is contacted. The needle is then retracted slightly and redirected until a flash of blood is obtained.

Rabbits. Restraint of rabbits is more difficult, as rabbits kick powerfully, which can result in luxation or subluxation of the vertebrae. Many handlers prefer a secure burrito-style towel wrap for restraint.
• Suggested collection sites are the central or marginal auricular vessels. For the purpose of blood collection, any large visible auricular vessel can be safely punctured for blood collection (Figure 1, page 73).
• Other collection sites include the lateral saphenous vein located along the lateral aspect of the leg above the hock (Figure 2, page 73) and cephalic veins.
• A 25- to 27-gauge needle with 1-mL syringe is recommended.

Guinea Pigs & Other Rodents. Guinea pigs and some larger rodents can simply be grasped around the thorax and shoulders for venipuncture restraint. Restraining smaller species is more difficult. For these patients, sedation can be extremely effective.
• Suggested collection sites include the cranial vena cava and lateral saphenous vein, following the steps outlined in Notes on Collection from the Cranial Vena Cava. We prefer the cranial vena cava in most rodents but the lateral saphenous in the guinea pig.
  • Other collection sites include the lateral tail vein for smaller rodents (rats) and, for larger rodents, the tarsal vein, which is often visualized along the dorsal aspect of the rear foot.
  • Laboratory collection techniques for rodents, including accessing the orbital sinus, are not recommended in pets.

Sugar Gliders & Hedgehogs. Some exotic pets, such as sugar gliders and hedgehogs, are challenging to restrain; sedation or anesthesia is required. It is also difficult to find an adequate venipuncture site.
• In both species, the cranial vena cava (Figure 3) is associated with a higher success rate for venipuncture.
• The ventral tail vein is sometimes useful in the sugar glider.

Pot-bellied Pigs. Pot-bellied pigs are difficult to restrain (vocalizing loudly and struggling) and have subcutaneous fat that complicates venipuncture. We prefer sedation and collection from the auricular vessels or the cranial vena cava (Figure 4)

**PROPER COLLECTION TECHNIQUE**
Improper collection techniques can produce poor-quality samples and cause patient harm. Proper precautionary measures should be taken to achieve the ideal sample.

- **Choose the correct size of needle and syringe** based on patient size and vessel being used. The ideal needle size is the largest that can be safely used in the patient. Very small needles or small needles used with larger syringes/excessive negative pressure will produce hemolysis.
- **Choose needles with clear hubs** to ideally detect the flash of blood when the vessel is punctured.
- **Preheparinize all needles and syringes** by pulling up a small amount of heparin and expelling it. We prefer this practice because it helps prevent micro clots in the needle and improves sample quality. Biochemistry values are not significantly impacted.
- **Properly restrain the patient**, either manually or ideally with sedation, if necessary.
- **Prepare the site** by clipping or cleaning with isopropyl alcohol.
- **Hold off the vessel** being utilized, if applicable.
- **Insert the needle**, bevel up, directly over the vessel. If you have entered the vessel correctly, you will see a small flash of blood in the hub of the needle.
- **Use minimal negative pressure** to acquire the proper sample volume needed.
- **Discontinue holding off the vessel** before removing the needle.
- **Consider entering the vessel with the needle only** in very small patients and collecting blood from the hub into heparinized hematocrit tubes.
- **Apply direct pressure** to the puncture site for 30 seconds after the needle has been removed.
- **Remove the top from the tube** selected for use.
- **Remove the needle from the syringe** and slowly dispense the sample into the collection tube, holding the tube at an angle and letting the sample roll down the side of the tube. Pushing the sample

<table>
<thead>
<tr>
<th>Species</th>
<th>Collection Site</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ferrets</td>
<td>Cranial vena cava</td>
<td>Vena cava approach especially easy and safe, even with manual restraint, due to ferret anatomy</td>
</tr>
<tr>
<td></td>
<td>Jugular</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Tail vein</td>
<td></td>
</tr>
<tr>
<td>Guinea Pigs</td>
<td>Lateral saphenous</td>
<td>Restraint relatively easy and located more proximally than expected; shaving enhances visualization</td>
</tr>
<tr>
<td></td>
<td>Cranial vena cava</td>
<td>Manual restraint for vena cava approach extremely stressful; not recommended</td>
</tr>
<tr>
<td></td>
<td>Cephalic vein</td>
<td>Reduced size makes adequate sample collection difficult</td>
</tr>
<tr>
<td>Pot Belly Pigs</td>
<td>Cranial vena cava</td>
<td>Subcutaneous fat makes venipuncture challenging; sedation or anesthesia recommended</td>
</tr>
<tr>
<td></td>
<td>Auricular vessels</td>
<td></td>
</tr>
<tr>
<td>Rabbits</td>
<td>Auricular vessels</td>
<td>Reported risk of vessel damage with auricular method; however, rare in our experience</td>
</tr>
<tr>
<td></td>
<td>Lateral saphenous</td>
<td>Restraint relatively easy</td>
</tr>
<tr>
<td></td>
<td>Medial femoral</td>
<td>Restraint relatively easy</td>
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</tr>
<tr>
<td>Rodents</td>
<td>Cranial vena cava</td>
<td>Sedation or anesthesia recommended; manual restraint for any collection site is difficult and not recommended</td>
</tr>
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<td>Sugar Gliders &amp; Hedgehogs</td>
<td>Cranial vena cava</td>
<td>Sedation or anesthesia recommended; manual restraint impossible in all but extremely debilitated patients</td>
</tr>
</tbody>
</table>
NOTES ON COLLECTION FROM THE CRANIAL VENA CAVA

The cranial vena cava is the largest accessible blood vessel in mammals; therefore, it is often extremely useful in small exotic companion animal patients.

- The access point is under the manubrium of the sternum.
- Depending on the species, the puncture site can be located just rostral to major cardiac vessels (small rodents) or separated from these vessels by a distance of 3 to 4 cm (ferrets).
- The patient must be kept absolutely still due to risk of significant bleeding if a vessel is lacerated. Some patients, like the ferret, can be manually restrained, but most require sedation or anesthesia. Extremely debilitated patients may not require sedation.
- The syringe and needle are inserted to the right, left, or just under the manubrium of the sternum; then directed toward the opposite hip.
- Negative pressure is applied until a flash of blood is obtained.

back through the needle or with excessive force will produce hemolysis.

COMPLICATIONS
As with any procedure, there are always associated risks that may lead to injury or death of the patient. Inexpert venipuncture can result in hematoma formation, infection and/or inflammation at the site, and potentially permanent destruction of the vessel. However, following the tips and suggestions presented in this article will help prevent negative consequences from taking place during venipuncture in your companion animal patients.

CBC = complete blood count; EDTA = ethylenediamine tetra-acetic acid; RBC = red blood cell; WBC = white blood cell

Reference

Suggested Reading