

Standard Operating Procedure: Bone Marrow Aspiration and Biopsy in Dogs and Cats

These SOPs were developed by the Office of the University Veterinarian and reviewed by Virginia Tech IACUC to provide a reference and guidance to investigators during protocol preparation and IACUC reviewers during protocol review. They can be used as referenced descriptions for procedures on IACUC protocols. However, it is the sole responsibility of the Principal Investigator to ensure that the referenced SOPs adequately cover and accurately represent procedures to be undertaken in any research project. Any modification to procedure as described in the SOP must be outlined in each IACUC protocol application (e.g. if the Principal Investigator plans to use a needle size that is not referenced in the SOP, simply state that alteration in the IACUC protocol itself).

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I. Procedure Summary & Goal

- a. The collection and analysis of material from a patient's bone marrow provides a great deal of diagnostic and prognostic information.
 - i. Diagnosing cause of cytopenia unresponsive to therapy, bicytopenia, pancytopenia, or markedly high blood cell counts.
 - ii. Visually detect histoplasma, leishmania, cytauxzoon, etc.
 - iii. Detect occult neoplasia
 - iv. Fever of unknown origin
 - v. Evaluate iron stores
- b. This information can be used to guide veterinarians and clients towards appropriate treatment protocols.

II. Personal Protective Equipment & Hygiene

- a. Hands should be washed thoroughly or sanitized before and after the procedure.
- b. Personal protective equipment appropriate to the setting should be used.

III. Supply List

- a. 16-gauge Rosenthal or Illinois aspiration needle (large dogs)
- b. 18-gauge Rosenthal or Illinois aspiration needle (small dogs / cats)
- c. Jamshidi marrow biopsy needle -12-13-gauge for most dogs -14-gauge (pediatric) for small dogs
- d. Electric clippers
- e. #40 Clipper blade
- f. Surgical scrub (betadine or chlorhexidine) and alcohol
- g. 2% lidocaine, 1 ml
- h. Syringe(s), 12 ml
- i. Sterile EDTA solution
- j. Petri dish
- k. Surgical gloves (sterile)

- Glass microscope slides
- m. Glass or plastic pipette
- n. #11 Bard-Parker scalpel blade
- o. Tissue fixative (Formalin or Zenker's solution) for biopsy
- p. Tissue cassettes for biopsy
- q. Sterile hypodermic needle, 20-22 gauge, ¾ to 1½ inch length
- r. Sterile drape (optional)

IV. Detailed Procedure

- a. Pre-operative Preparation
 - i. Fast the animal
 - 1. All patients will be fasted for 12-18 hours prior to the procedure.
 - 2. No medications will be administered prior to the pre-anesthetics.
- b. Pre-medicate your patient
 - i. See Sedation and Anesthesia SOP
- c. Place an intravenous catheter (see Peripheral Intravenous Catheter Placement SOP).
 - i. Start intravenous fluids
 - 1. Isotonic electrolyte solution (LRS or 0.9% NaCl)
 - 2. Rate: two times maintenance
- d. Place the patient under general anesthesia
 - i. See Sedation and Anesthesia SOP
- e. Palpate the anatomic landmarks at indicated aspiration site.
 - i. Common sites for bone marrow aspiration
 - 1. Dorsal iliac crest (dog)
 - 2. Trochanteric fossa medial to greater trochanter of the femur (dog and cat)
 - 3. Lateral aspect of the humeral head (dog and cat)
- f. Prepare skin at the aspiration site as described in the Sterile Scrub SOP.
- g. Local analgesia
 - i. Prior to final scrub
 - 1. Dogs: Infiltrate the skin, subcutaneous tissue, and periosteum over the needle insertion site with 1ml of 2% lidocaine.

- 2. Cats: Infiltrate the skin, subcutaneous tissue, and periosteum over the needle insertion site with 0.5ml of 2% lidocaine
- h. Complete final scrub
- i. Put on sterile gloves, saving the glove packaging as a sterile field.
- j. Aseptically prepare your bone marrow aspiration needle
 - i. Draw up 1-2 ml of EDTA anticoagulant into a 12ml syringe.
 - ii. Remove the stylet from the bone marrow aspiration needle and set it on the sterile field (sterile glove packaging).
 - iii. Flush 1-1.5 ml of the EDTA through the bone marrow needle into a petri dish and replace the stylet into the bone marrow needle.
- k. Make a 3-mm stab incision in the skin over the aspiration site
- I. Hold the needle with the back of the needle pressed at the junction between your fingers and palm.
 - i. Advance the bone marrow needle with stylet in place through the incision down to the level of the periosteum.
 - ii. Further advance the aspiration needle by applying a clockwise-counterclockwise rotating motion and steady pressure.
 - iii. Upon penetration of the near cortex of the desired bone, a significant decrease in resistance should be felt.
- m. Remove the stylet and make sure the needle can stand in place after the stylet is removed.
- n. Attach the 12 ml syringe containing EDTA to the aspiration needle and apply negative pressure by forcefully pulling back on the plunger to aspirate the marrow fluid.
 - i. To avoid dilution of the marrow sample with peripheral blood, less than 1ml of marrow should be aspirated.
- o. Once the marrow fluid is obtained, leave the aspiration needle in place, detach the syringe and eject the marrow contents into the petri dish containing EDTA.
 - i. The sample can be examined grossly for marrow particles (spicules) or an assistant can rapidly make a cytologic preparation to confirm that marrow has been obtained.
- p. Following successful sample collection, remove the aspiration needle by rotating and applying gentle traction on the needle.
- q. Bone marrow biopsy
 - i. Advance the needle an additional 1-2 cm after penetration into the bone cortex.
 - ii. Loosen the biopsy sample by alternate rotation and wobbling of the bone marrow needle.
 - iii. Partially withdraw the needle, redirect it at a different angle, and advance it into a new area of hone
 - iv. Again, loosen the biopsy sample by alternating rotation & wobbling of the needle.

- v. Remove the biopsy needle using rotation and gentle traction and place the biopsy sample in a tissue cassette in formalin or other tissue fixative.
- r. Administer analgesia upon recovery, and as needed thereafter. (Optional)
 - i. Dogs
 - Morphine; 0.25-1.0 mg/kg, hydrocodone; 0.1-0.2 mg/kg, buprenorphine; 0.005-0.03 mg/kg, IM or SQ, once upon recovery
 - 2. Ketoprofen: 2mg/kg IV, once upon recovery
 - 3. Carprofen: 1.0 2.2 mg/kg by mouth, every 12 hours up to three days post procedure
 - ii. Cats
 - 1. Buprenorphine; 0.01-0.03 mg/kg; hydrocodone; 0.05-0.2 mg/kg, IM, or SQ

V. Potential Adverse Effects, Mitigation, or Treatment

- a. Infection at the site, but this is rare
- b. Trauma to soft tissue structures such as muscle, nerves, and potentially joint surfaces
- c. Hemorrhage at the site
- d. Post procedure pain
- e. Avoidance Measures:
 - i. Pay careful attention to sterile technique; supervision of laboratory instructors
 - ii. Pay careful attention to landmarks, have skeleton/bone specimens available to use to appropriately find the landmarks and avoid unintended trauma to surrounding tissues; supervision of laboratory instructors
 - iii. Apply direct pressure to the site. Hemorrhage into the bone marrow does not cause problems because the blood is absorbed quickly.
 - iv. Pain is usually brief and not lasting after the procedure. Analgesics are used as pre-meds for sedation/anesthesia and are usually adequate, but may be used post-operatively if needed.

VI. Variations

- a. Contraindications
 - i. DIC
 - ii. Exposure to anticoagulant rodenticides
 - iii. Severe liver failure
 - iv. Severe anemia (transfuse prior to sedation)

VII. Links to Multimedia Aids and References

a. http://www.vetmed.wsu.edu/resources/techniques/bm.aspx

b.	http://veterinarymedicine.dvm360.com/vetmed/Diagnostic+Center/Skills-Laboratory-How-to-collect-diagnostic-bone-m/ArticleStandard/Article/detail/174492