

Bone marrow testing is used to diagnose and monitor bone marrow diseases and CBC abnormalities. Full evaluation of bone marrow requires a CBC the day of the bone marrow collection, bone marrow aspiration, and a bone marrow core biopsy.

Indications for bone marrow evaluation:

- Persistent or unexplained neutropenia, nonregenerative anemia, and/or thrombocytopenia
 - Generally, patients with marked thrombocytopenia tolerate bone marrow aspirate/core biopsies well
- · Persistent or unexplained marked leukocytosis and/or thrombocytosis
- Abnormal blood cell morphology such as immature cells or dysplastic changes
- Further evaluation of hyperproteinemia
- Detection of metastasis to bone marrow (lymphoma, carcinoma, etc.)

Helpful tips:

- If there is pathology directly related to the bone (bony lysis, bony mass, periosteal reaction, etc.), a bone aspirate/biopsy is the appropriate test (link to bone biopsy sheet)
- Depending on clinical concerns, a bone marrow aspirate or bone marrow core biopsy may be performed independently of one another, though both are heavily reliant on comparison with a current CBC for interpretation

Bone Marrow Aspirate Procedure

Supplies needed

- Sterile gloves
- Scalpel blade
- 16- or 18-gauge needle with removable stylet (Jamshidi, Illinois sternal bone marrow aspiration needle, etc.), 1-1.5 inches in length
- Syringe (10-20 mL, ideally containing EDTA anticoagulant)
- · Glass slides
- EDTA (purple top) tube
- Petri dish (optional)

Collection

- The most common bone marrow collection sites are the proximal humerus for adult dogs and cats and the sternum or proximal ribs for large animals
 - Other sites include the proximal femur (for small cats/toy breed dogs) or the iliac crest (medium to large dogs)
- Collection site is clipped and scrubbed for sterility, using sterile gloves for the procedure
- Inject local anesthetic from the skin to the overlaying periosteum
- Make a small skin incision and insert bone marrow aspiration needle through incision
- Apply moderate pressure to needle, rotating the needle in alternating clockwisecounterclockwise directions until the needle is firmly embedded into the marrow cavity

- The tip of the needle must pass through a layer of cortical (cancellous) bone before entering the medullary cavity, which contains bone marrow
- Once needle is seated, remove stylet and attach syringe to needle
- Apply vigorous negative pressure rapidly to increase chance of collecting bone marrow particles, which may appear as small yellow specks entering the syringe
- · Quickly remove syringe and make 4-5 smears
 - Placing only a small drop of sample (about the same amount needed for a blood smear) is recommended, as smears that are too thick greatly reduce visibility of the cells
 - If syringe does not contain EDTA, samples should be made quickly, or clotting of the sample can markedly degrade/lyse hematopoietic cells
- · Empty contents of syringe into an EDTA tube
- If no bone marrow particles are observed on the slides, the stylet can be reinserted and redirected

Helpful tips

- Images of diagnostic bone marrow aspirates are shown below (Figure 1A and 1B)
- If no particles are identified, the contents of the EDTA tube can be placed in a petri dish, and particles may stick to the dish when sample is swirled. These particles can be collected and placed on a glass slide for smearing. This procedure will be done at TVMDL if provided bone marrow aspirate slides do not yield marrow particles (Figure 2)
- Please keep aspirate slides away from formalin and formalin vapors
 - · Put slides away before opening formalin jar
 - Ship bone marrow aspirates and core biopsies in separate containers if possible)



Figure 1: A) Diagnostic bone marrow aspirate preparation, unstained. The blue arrows point to areas with bone marrow particles. B) Diagnostic bone marrow aspirate preparation, stained. The dark blue stained area signifies abundant bone marrow particles. **Images provided with collaboration with Purdue University (Camila Benaduce E. Mello, DVM, and Craig A. Thompson, DVM, MS).*



Figure 2: Bone marrow particles observed as white flecks sticking to the petri dish. These can be collected and placed on a slide for cytologic evaluation.

Bone Marrow Core Biopsy Procedure

Supplies needed

- Sterile gloves
- Scalpel blade
- 11-13-gauge Jamshidi needle 3-4 inches long
- Formalin container

Collection

- The most common bone marrow collection site is the proximal humerus for adult dogs and cats and the sternum or proximal ribs for large animals
 - The head of the femur is not used for bone marrow biopsies, as the sampled core material often does not stay in the needle when removed
 - · Ideally, bone marrow aspirates and core biopsies are sampled from two separate locations
- · Collection site is clipped and scrubbed for sterility, using sterile gloves for the procedure
- · Inject local anesthetic from the skin to the overlaying periosteum
- Make a small skin incision and insert needle through incision. The stylet should be locked in place to prevent excess movement
- Apply moderate pressure to needle, rotating the needle in alternating clockwise-counterclockwise directions until the needle is firmly embedded into the bone
 - The tip of the needle must pass through a layer of cortical (cancellous) bone before entering the medullary cavity, which contains bone marrow
 - Larger and older dogs will have a thicker layer of cortical bone than smaller and younger dogs
- Once needle is seated, remove the stylet and continue advancing the needle with alternating clockwise-counterclockwise motions, at least one inch if possible



Figure 3: The needle stylet (Jamshidi biopsy needle depicted) is inserted into the proximal humerus near the point of the shoulder (A). The distal limb (B) can be held with the elbow joint at a 60-90° angle to facilitate needle advancement.

- Remove the needle and use the provided thin, hooked wire to push the collected bone marrow out of the **handle end** of the needle
 - Forcing the marrow out the tip end of the needle can lead to crush artifact, as the tip of the needle is thinner than the body
- Place core in formalin fixative

Helpful tips

- Ideally, before placing the bone marrow core biopsy in fixative, the bone marrow sample can be rolled across a glass slide and sent with the core. This is called a roll preparation
- Please take caution to prevent formalin artifact in aspirates or roll preparations when also collecting bone marrow core biopsies. Formalin exposure to slides (the liquid and the vapors of formalin) can irreversibly alter staining properties. Slides meant for cytological review should be shipped in separate containers