

COLLECTION OF BLOOD SAMPLE: CATTLE**1. PURPOSE**

To describe how to safely collect blood sample from cattle.

2. RESPONSIBILITY

- 2.1 Dairy Manager
- 2.2 Farm manager
- 2.3 Veterinarian
- 2.4 Researchers

*** NOTE**

- Demonstrator: The operators should be familiar with the correct techniques and the anatomy of cattle before attempting this procedure
- Students: The procedures must be clearly demonstrated before students attempt them. Students should be aware of the requirements for sterile technique.

3. MATERIAL

The type of material used will vary according to the collection method.

- 3.1 Disposable gloves
- 3.2 Halter
- 3.3 Clippers (optional)
- 3.4 Vacutainer collection:
 - 3.4.1 20G1" vacutainer needle: Coccygeal vein collection
 - 3.4.2 14G2" vacutainer needle: Jugular vein
 - 3.4.3 One vacutainer tube or bottle per animal (lavender, green, red)
 - 3.4.4 Vacutainer tube holder
- 3.5 Syringe Collection:
 - 3.5.1 20G1" needle: Coccygeal venipuncture
 - 3.5.2 14G2" needle: Jugular venipuncture
 - 3.5.3 1 – 5 cc syringe
- 3.6 Bleeding Tube Collection (> 100 ml)
 - 3.6.1 2 x 14G2" vacutainer needles
 - 3.6.2 500ml heparinized bottle (for collection of large volume e.g. 500ml)
 - 3.6.3 Heparin
 - 3.6.4 Bleeding tube (for collection of large volume, e.g. 500ml)
- 3.7 Antiseptic
- 3.8 Gauze
- 3.9 Sharps container
- 3.10 Biohazard box for syringe disposal

4. CONSIDERATIONS

- 4.1 Ensure that the animal is properly restrained to avoid injury to the animal and/or study personnel.
- 4.2 Blood can be collected from the jugular vein in cattle.

Blood Volume (mg/kg)	Total Blood volume (TBV), normal adult (ml)	Safe volume for single bled (ml)
60	27,000 - 36,000b (assumes adult weight 600-800kg)	2700 - 3600

FIGURE.1: NC3Rs Blood Sample Volumes

- 4.3 Minimal restraint is usually required to obtain blood samples from a cow. If restraint is necessary, it should be executed to ensure quick, easy and safe collection of the sample causing minimal distress.
- 4.4 Allow the animal some time to adjust to their restraint before beginning.
- 4.5 Operators should use gloves and disinfect them between animals to prevent the transmission of blood-borne diseases.
- 4.6 Equipment such as vacutainer holders must be cleaned between animals.
- 4.7 Use a fresh needle for each animal to eliminate risk of cross-contamination.

5. PROCEDURES

- 5.1 Use clean gloves to prevent the transmission of blood-borne diseases.
- 5.2 Use a fresh bleeding needle with every cow.
- 5.3 **JUGULAR VENIPUNCTURE USING VACUTAINER NEEDLE AND TUBES:** (<100 ml)
 - 5.3.1 Using the halter, position the animal's head so that it is slightly elevated and drawn to the side to expose the jugular vein. (Figure 2)



FIGURE 2: Restrain the animal to expose the jugular vein

- 5.3.2 Screw the 14G 2" Vacutainer needle onto the vacutainer holder.

- 5.3.3 Insert the vacutainer tube into the other end of the Vacutainer holder. DO NOT puncture the stopper. Hold these assembled materials in one hand.
- 5.3.4 Disinfect venipuncture site with alcohol.
- 5.3.5 Occlude jugular vein by applying pressure in the jugular groove located in the lower neck. (Figure 3)
- 5.3.6 Insert the needle into the distended jugular vein at a 45° angle cranial to the jugular groove. (Figure 3)



FIGURE 3: Occlude the vein and insert the needle

- 5.3.7 Once needle is positioned in the vein, insert a vacutainer tube into the needle to collect the blood.
- 5.3.8 You can collect more than one tube by replacing vacutainer tubes.
- 5.3.9 When the desired volume has been collected (5 ml minimum suggested) remove the occluding pressure from the vein.
- 5.3.10 Detach the tube from the needle and withdraw the needle from the jugular vein.

*** NOTE**

DO NOT PULL NEEDLE OUT OF VEIN WITH VACUTAINER TUBE STILL ATTACHED, AS THIS WILL RELEASE VACUUM IN VACUTAINER.

- 5.3.11 Apply pressure with gauze for 30 to 60 seconds in order to ensure adequate hemostasis.
- 5.3.12 Invert the tube several times to mix.

5.4 JUGULAR VENIPUNCTURE USING SYRINGE AND NEEDLE: (<100 ml)

- 5.4.1 Attach a 14G 2" needle to a 1 or 5 cc syringe.

- 5.4.2 Using the halter, position the animal's head so that it is slightly elevated and drawn to the side to expose the jugular vein. (Figure 2)
- 5.4.3 Disinfect venipuncture area with alcohol.
- 5.4.4 Occlude jugular vein by applying pressure in the jugular groove located in the lower neck. (Figure 3)
- 5.4.5 Pass the needle through the skin and into the vein by a firm thrust at an angle of 20° to the skin surface.
- 5.4.6 Withdraw the blood sample (Figure 4).



FIGURE 4: Occlude the vein, insert needle and withdraw blood

- 5.4.7 When the desired volume has been collected (5 ml minimum suggested), remove the occluding pressure from the vein. And remove the needle.
 - 5.4.8 Apply pressure with gauze for 30 to 60 seconds in order to ensure adequate hemostasis.
 - 5.4.9 Dispose of needle(s) in a Sharps container.
 - 5.4.10 Transfer the blood to the appropriate vacutainer tube.
 - 5.4.11 Invert the tube several times to mix.
 - 5.4.12 Serial samples can be taken by alternating sides, and by moving insertion sites cranially, as long as there is no hematoma formation.
- 5.5 JUGULAR VENIPUNCTURE USING BLEEDING TUBE (> 100 ml):**
- 5.5.1 Flush the Bleeding tube with heparin to prevent coagulation during collection.
 - 5.5.2 Attach a 14G 2" vacutainer needle on each end of the bleeding tube.



FIGURE 5: Attach vacutainer needle on each end of the bleeding tube

- 5.5.3 Disinfect the rubber seal of the heparinized bottle with alcohol.
- 5.5.4 Uncap the needle of one end of the bleeding tube and insert into the rubber top of the heparinized bottle.
- 5.5.5 Insert the needle on the other end of the tube into the distended jugular vein at a 45° angle cranial to the jugular groove. (see Figure 3 for positioning)
- 5.5.6 You can collect more than one bottle by transferring the needle on the bottle end to another bottle. Refer to [Figure 1 NC3Rs Blood Sample Volumes](#) for safe collections for a single bleed.
- 5.5.7 When the desired volume has been collected, remove the occluding pressure from the vein.
- 5.5.8 Detach the bleeding tube from the needle and withdraw the needle from the jugular vein.
- 5.5.9 Invert the bottle several times to mix.

*** NOTE**

DO NOT PULL NEEDLE OUT OF VEIN WITH VACUTAINER TUBE STILL ATTACHED, AS THIS WILL RELEASE VACUUM IN VACUTAINER.

- 5.5.10 Apply pressure with gauze for 30 to 60 seconds in order to ensure adequate hemostasis.
- 5.5.11 Serial samples can be taken by alternating sides, and by moving insertion sites cranially, as long as there is no hematoma formation.

5.6 COCCYGEAL VENIPUNCTURE USING VACUTAINER NEEDLE AND TUBES (<100 ml):

- 5.6.1 Restrain the cow to prevent her from moving away during the procedure.
- 5.6.2 Raise the tail vertically until it is horizontal with the ground and the ventral surface of the tail is accessible.
- 5.6.3 Locate the groove lying in the ventral midline of the tail. (~150 mm from the base of the tail). (Figure 6)



FIGURE 6: Coccygeal Vein

- 5.6.4 Screw the 20G 1" Vacutainer needle onto the vacutainer holder.
- 5.6.5 Insert the vacutainer tube into the other end of the Vacutainer holder. DO NOT puncture the stopper. Hold these assemblies material into one hand.
- 5.6.6 Disinfect venipuncture area with alcohol.
- 5.6.7 Midway along the body of a coccygeal vertebra, insert the 20G 1" needle attached to the vacutainer holder perpendicularly to the surface of the skin to a depth of a few millimeters.
- 5.6.8 Once needle is positioned, insert a vacutainer tube into the needle to collect the blood.
- 5.6.9 You can collect more than one tube by replacing vacutainer tubes.
- 5.6.10 When the desired volume has been collected (5 ml minimum suggested), remove the occluding pressure from the vein.
- 5.6.11 Detach the vacutainer tube from the needle and withdraw the needle from the jugular vein.

*** NOTE**

DO NOT PULL NEEDLE OUT OF VEIN WITH VACUTAINER TUBE STILL ATTACHED, AS THIS WILL RELEASE VACUUM IN VACUTAINER.

- 5.6.12 Apply pressure with gauze for 30 to 60 seconds in order to ensure adequate hemostasis.
- 5.6.13 Invert the tube several times to mix.

5.7 COCCYGEAL VENIPUNCTURE USING SYRINGE AND NEEDLE (<100 ml):

- 5.7.1 Restrain the cow to prevent her from moving away during the procedure.
- 5.7.2 Raise the tail vertically until it is horizontal with the ground and the ventral surface of the tail is accessible.
- 5.7.3 Locate the groove lying in the ventral midline of the tail. (~150 mm from the base of the tail). (Figure 6)
- 5.7.4 Disinfect venipuncture area with alcohol.
- 5.7.5 Midway along the body of a coccygeal vertebra, insert the 20G 1" needle perpendicularly to the surface of the skin to a depth of a few millimeters. (Figure 7)
- 5.7.6 Withdraw blood sample (Figure 8) and remove needle. In order to ensure adequate hemostasis, apply pressure with gauze for 30 to 60 seconds.



FIGURE 7: Insert the needle in the groove lying in the middle of the tail



FIGURE 8: Withdraw blood sample

- 5.7.7 Apply pressure with gauze for 30 to 60 seconds in order to ensure adequate hemostasis.
- 5.7.8 Transfer the blood to the appropriate vacutainer tube.
- 5.7.9 Invert the tube several times to mix.
- 5.8 Promptly dispose of used sharps in the sharps container. Note: Needle holders can be re-used.
- 5.9 Identify the tube(s) / bottle(s) with the animal's identification name or number and farm name and complete the sample form.
- 5.10 Place the tube(s) in an ice rack or refrigerator until it is shipped.

6. REFERENCES

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Document Status and Revision History

DATE	PREVIOUS VERSION	NEW VERSION
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