



BAL Collection in Cows and Calves

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Calf Procedure

Bronchoalveolar lavage (BAL) is performed in sedated calves using a sterilized, flexible 10 French X 36 inch catheter with a 3-cc balloon cuff (Mila International, Inc. Medical Instrumentation for Animals, Florence KY). Five to 10 minutes after administration of 0.1 mg/kg xylazine IM, the sedated calf is restrained and the nostrils are cleaned with a dry 4X4" gauze sponge. The head and neck of the calf are extended to facilitate passage of the sterile BAL catheter by a person wearing surgical gloves. Prior to catheter introduction into the nostril, sterile saline is dripped into the catheter to lubricate the guide-wire stylette. The BAL catheter is introduced into the ventral meatus of the nose through which it is advanced until it encounters resistance in the caudal pharynx. At that point, the restrainer pushes the poll of the calf's head ventrally while simultaneously elevating the ventral mandible and the catheter is advanced down the trachea during the inspiratory phase of the respiratory cycle (see image below).



Repeated coughing is induced with proper catheter placement and it is rapidly advanced until resistance is met as it wedges in a cranial lung lobe bronchus. A failure to induce spontaneous coughing subsequent to passage beyond the pharynx usually implies passage into the esophagus. In the wedged position, the catheter is held firmly in place while the guide-wire stylette is removed. The balloon cuff is then inflated with 3 cc of air and 120-ml of sterile saline is infused using 60-ml syringes with a stopcock and catheter tipped adapter (company name) attached. Immediately after the 120 ml infusion, negative pressure is applied to aspirate fluid, a process that usually yields 10 to 40 ml of clear to mildly turbid, foamy fluid. The returned fluid sample is placed into a sterile 4-oz specimen cup. A second 120-ml infusion is introduced and aspirated as described and the pooled fluid is sealed in the specimen cup and preserved in a cooler until it can be processed. The fresh BAL fluid sample is processed within 2 hours of collection or refrigerated until it can be analyzed. A 5-ml aliquot of the pooled sample is used for bacterial and *Mycoplasma* cultures. The remaining fluid is submitted for cytological interpretation, which is based on routine staining of cytospin and direct smear preparations. BAL fluid that yields homogenous ($> 10^6$ CFU/ml) bacterial or positive *Mycoplasma bovis* culture is considered abnormal. A disproportionate lowering of macrophages ($\leq 60\%$) or elevation of neutrophils ($\geq 40\%$) provides evidence of an inflammatory response with or without a positive culture.



Procedure for Adult Cows

Bronchoalveolar lavage (BAL) is performed in sedated cows using a sterilized, flexible BAL tube (Bivona – 3 meter length, 11 mm outside diameter, 5-cc balloon cuff; call 800-258-5361 to order). Five to 10 minutes after administration of 0.08 mg/kg xylazine IM, the sedated cow is restrained and the nostrils are cleaned with a dry 4X4" gauze sponge. Prior to catheter introduction into the nostril, the integrity of the balloon is tested by introduction of 5-cc of air. The balloon is deflated and the sterile BAL catheter is introduced into the nostril and directed into the ventral meatus by a person wearing sterile gloves. The BAL catheter is advanced until it encounters resistance in the caudal pharynx. At that point, the restrainer maximally extends the head and neck of the cow while simultaneously keeping the neck straight. The catheter is passed into the trachea during the inspiratory phase of the respiratory cycle (see image below).



Repeated coughing is induced with proper catheter placement and it is then rapidly advanced until resistance is met as it wedges in a cranial lung lobe bronchus. Failure to induce spontaneous coughing subsequent to passage beyond the pharynx usually implies passage of the tube into the esophagus. In the wedged position, the balloon cuff is inflated with 5 cc of air, a 3-way stopcock is inserted into the end of the BAL tube and 240-ml of sterile saline is infused using 60-ml syringes. Immediately after the 240 ml infusion, negative pressure is applied to aspirate fluid, a process that can yield up to 120 ml of mildly turbid, foamy fluid. Mucus and purulent flecks may be visualized and some samples are slightly blood-tinged. The returned fluid sample is placed into sterile 4 to 8 oz specimen cups. A second 240-ml infusion is introduced and aspirated as described and the pooled fluid is sealed in the specimen cup and preserved in a cooler until it can be processed. When fluid collection is finished, the balloon is deflated completely and the catheter removed from the nose.

The fresh BAL fluid sample is processed as soon after collection as possible. If it is not processed within 2 hours of collection, it can be refrigerated until submission of samples. A 5-ml aliquot of the pooled sample is used for bacterial and *Mycoplasma* cultures. A similar fluid aliquot can be submitted for BRSV ELISA or PCR testing. The remaining fluid is used for cytological interpretation, which is based on routine staining of cytospin and direct smear preparations. BAL fluid that yields homogenous ($> 10^6$ CFU/ml) bacterial or positive *Mycoplasma bovis* culture is considered abnormal. A disproportionate lowering of macrophages ($< 80\%$) or elevation of neutrophils ($>20\%$) provides evidence of an inflammatory response with or without a positive culture.