

Collection of Swabs for Diagnosis of Respiratory Pathogens by PCR

PCR is a very sensitive diagnostic tool for identification of nucleic acid of respiratory pathogens on swabs. Care should be taken in collection of swabs to avoid contamination by nucleic acids on hands. ***Clean exam gloves should be worn during collection of swabs and the gloves should be changed between EACH ANIMAL to avoid cross contamination.***

1. Ideally, the swabs should be collected from animals in the acute phase of clinical disease (<7 days) and before antibiotics are administered when possible since the amount of some pathogens in the upper respiratory tract can decrease after this time period, especially canine influenza and canine respiratory coronavirus. To increase diagnostic accuracy, ***swabs should be collected from several animals, including those with clinical disease and those exposed but without clinical signs.***
2. Always collect samples from the healthy animals first and the animals with respiratory signs second to avoid cross-contamination.
3. Two to three swabs should be collected from each animal and pooled together in one dry sterile sample tube to maximize detection of any respiratory pathogens. Dacron swabs are preferred when available.
 - a. ***For each cat***, collect a conjunctival swab and deep pharyngeal swab and place both swabs in the same dry sterile tube to be processed together.
 - b. ***For each dog***, collect a conjunctival swab, a deep nasal swab, and a deep pharyngeal swab and place all three swabs in the same dry sterile tube to be processed together.
 - c. For the conjunctival swab, pull a lower eyelid away from the eyeball to expose the conjunctival sac. Insert the tip of the swab into the sac and firmly rub the inside of the eyelid to remove the epithelial cells and to moisten the swab tip. Most animals tolerate this procedure very well. Open a clean sterile tube, insert about one-third of the swab stick, then snap the plastic handle so that it breaks and the swab tip falls into the tube. Close the tube tightly.
 - d. For the deep pharyngeal swab, ask an assistant to open the mouth. Then vigorously swab the back of the oropharynx near the tonsils, taking care to avoid the tongue and heavy saliva contamination. Add this swab to the same tube with the conjunctival swab.
 - e. For the deep nasal swab (best sample for canine influenza virus), insert the swab tip as far into a nostril as possible and rub to remove epithelial cells. Most dogs will tolerate this once, but may not hold still for more attempts. Add this swab to the other two already in the tube.
4. A single bacterial culturette from a deep pharyngeal swab should also be submitted. This will undergo culture and sensitivity if *Bordetella* or *Streptococcus zooepidemicus* are detected.
5. To avoid cross-contamination, label each tube with the animal's ID using a permanent marker such as a Sharpie pen, snap the cap tightly, and drop the pair of tubes (one for PCR and one for culture) into that dog's individual baggie before changing gloves and moving on to the next animal. Label the outside of baggie as well.



6. Packing and shipping the samples

- a. Once the swabbing is complete, the individual sealed baggies should be placed together in a larger baggie and stored in the refrigerator (not frozen) pending shipping. No liquids should be added to the swabs or tubes.
- b. The samples should be packed in a Styrofoam shipping container with frozen ice packs to keep the samples cool but not frozen.
- c. The form should be sealed in its own baggie to protect it from moisture before placing it in the box with the samples.
- d. The package should be shipped by overnight courier.

