

Enhancing Sample Collection for the Dermatology Patient

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Abstract

On an almost daily basis, a patient is presented with dermatologic issues as the chief complaint for the visit such as a “hot spot”, an ear infection or pruritus (itch). Technicians are often taking a history for the veterinarian while simultaneously obtaining a TPR (temperature, pulse, respiration). Collecting an ear cytology or skin cytology is something that a technician could easily do at this time to increase the efficiency of the appointment. With practice, reviewing the samples can be accomplished in a short amount of time. The veterinarian could then have results of these simple in-house diagnostics when they enter the exam room to have a more focused discussion on what therapies are needed and differentials to address with the client. This is good medicine, increases income for the hospital and documents a complete medical record. This discussion will focus on technique for sample collection, interpretation of findings and differentials based on signalment and physical exam.

Keywords: atopy, pruritus, folliculitis

Discussion

Sample collection for a dermatology patient is likely a daily occurrence in animal clinics. Citing being too busy or that it is not necessary to take samples does not outweigh the positive reasons for taking the time to do so. Having accurate results provides the patient with excellent medical

care, yields a definitive diagnosis, at recheck can assess response to therapy (very important if patient is not better—cannot assess if didn't know what was there in the first place), creates a more defined list of differentials for the underlying cause, and is financially beneficial to clinic, staff and client.

Sample collection implements, aside from the microscope, are not expensive and most clinics use some of the materials in their day to day use (scalpel blades for surgery for example). Set up a “derm caddy” that contains all the materials needed to take samples on the dermatology patient. The caddy should contain: microscope slides, cover slips, mineral oil, number 10 scalpel blades (sterile), clear acetate tape, cotton-tipped applicators, Diff-Quik stain #3 in a syringe and a marker. Additional items needed are: Diff-Quik stain, immersion oil, lighter to heat fix, and +/- gram stain. Please wear gloves when handling patients with dermatology conditions to protect yourself as well as other patients.

Pruritus (itch) is a very common presenting complaint of our patients. Pruritus is present from an underlying cause and pet may have infection secondary to this underlying cause which exacerbates the pruritus. Treatment of infection can improve pruritus however not addressing underlying cause will result in that patient continuing to suffer from recurrent infections and pruritus. A patient may present with an ear infection but knowing where else to look can give some clues to the underlying cause.

Dermatology patients often have folliculitis (macule, papule, pustule, epidermal collarettes, alopecia) that accompany the pruritus. The three rule outs for folliculitis are: bacteria (most common), dermatophyte and *Demodex*. A further discussion of various sampling methods and techniques will be reviewed to rule in or rule out these differentials.

Dermatophyte culture

Although a positive Wood's lamp is indicative of *Microsporum canis*, it does not fluoresce for all *M. canis* (approximately 50%) cases and does not fluoresce at all for *Microsporum gypseum* or *Trichophyton mentagrophytes*. It is important to identify the source of the dermatophyte in order to give recommendations for the client to treat all affected pets, hygiene and why pet was not able to self-limit the infection. Identification of dermatophyte can be done in hospital or sent to an outside laboratory.

Collect samples from the leading margin of lesion as a false negative may result if taking from the center of a suspect area as it resolves. Culture media is kept in the refrigerator and should be allowed to come to room temperature before plating the samples. Hairs may be plucked using fingers or a hemostat. Using a new toothbrush to comb pet head to tail (lesional areas last) is recommended when rechecking cultures or trying to find asymptomatic carriers. Use sterile scissors to cut the tips of the toothbrush—do not force into the agar. Samples should be evaluated daily for growth as well as color change on media. Samples should be kept warm and with increased humidity. Acetate tape is applied to the fungal colony and examined under 10X on the microscope for identification. Color change and growth do not mean it is necessarily positive for dermatophyte but contaminant from the environment which is why taking the next step of identifying the type of conidia is important.

Skin Cytologies

The more often you perform and evaluate skin cytologies, the more efficient and confident in your results you will become. There are various techniques in obtaining skin cytology that can be utilized.

Direct impression smears are best when lesion is a pustule, is hemorrhagic or exudative (moist) and haired skin areas that are able to be held with thumb and forefinger. Hard to do in between small paws and may be dangerous to do near an eye. These are to be stained and evaluated on oil immersion (100X).

Acetate tape preparations are for dry lesions, in between digits of the paws, near eyes and for areas difficult to obtain good contact with the slide. The tape is then placed on a slide that has been prepared by placing Diff-Quik stain #3 on it. Use a paper towel to press tape onto slide and also squeezes excess stain out. Slide is ready to be evaluated. Immersion oil is placed directly on top of the tape.

Nose ropes and tail folds a cotton-tipped applicator is used to get material in the crevices and is rolled on to a slide like an ear cytology.

Ear cytology

Cotton-tipped applicators (CTA) are used to collect material from the ear canal. Gently swab the canal to collect exudate and roll onto slide. Break the CTA to make an “L” if unable to roll out onto a slide. If thick, take a second CTA and thin the sample out otherwise will be difficult to assess. If using slides that are frosted, left and right. Non-frosted, consider making an “L” with the material. Consider making two identical slides when dealing with patients that have chronic otitis. If rods are noted, a slide is made from same material for gram stain and patient does not have to be restrained to gather another sample which may be uncomfortable or painful for the pet. Ear cytology slides are heat fixed to melt the waxy material so the stain penetrates better. A slide that was not heat fixed will not come off but particularly yeast organisms may not pick up the stain as well and may be missed because they are not the dark purple. Always collect

samples from both ear canals even if one does not seem to be posing an issue. This will document that infection is either not present at the visit, there is an infection that would not have been detected if not sampled and is excellent medical record documentation.

Claw bed

Paronychia is the inflammation of the claw bed with or without secondary infection. Infection from around the claw can be different than what is detected between the digits. Inspect the claw for crusting or exudate around where the claw comes into contact with the skin. Often claws are discolored brown secondary to the infection and they will grow out gradually normally if infection is appropriately addressed. The wooden end of the CTA is used to scrape material down the claw as pushing up can injure the pet and cause more discomfort. The end is aggressively rubbed onto the slide to transfer the material which is generally waxy in nature requiring sample to be heat fixed and then stained like ear cytology samples.

Skin Scrapings

Skin scrapings are done superficially to look for *Sarcoptes*, *Notoedres* and *Demodex gatoi*. Deep skin scrapings are to look for *Demodex canis*, *D. injai* and *D. cati*. Use a new or sterilized (cost efficient!) number 10 scalpel blade for each patient to eliminate contamination from between pets for blood borne pathogens. Superficial scrapings are messy! Apply mineral oil to the area on the pet to be scraped. Superficial mites will be caught in the oil. Remove the oil, scale, and crust on affected area down to “normal skin” and put on slide that also has mineral oil on it. For deep scrapes, pinch the area to extrude the mites, scrape in one direction with blade perpendicular to the skin in the grain of hair growth ideally scraping towards you. Inspect blade occasionally for evidence of blood on the blade. Once this is noted, squeeze affected area one

more time and do a final swipe with the blade to collect the blood, debris and oil and transfer to slide that has been prepared with mineral oil. It is helpful to apply a cover slip to ensure the entire slide is examined for presence of mites. While the mites can be visualized at 4X, 10X makes it much more obvious and less likely to miss. The condenser should be lowered down to enhance contrast (as you would when reading a fecal sample).

An alternative to the traditional skin scrape with a blade is a trichogram. Hairs are plucked in the direction of hair growth and place on a slide prepared with a heavy drop of mineral oil. This technique is best for areas that a blade should not be near such as the periorbital region. Also over bony areas such as paws. If *demodex* mites are present, they will come out with the hair. Many clinicians have changed to this as their only method of evaluating for *demodex*. Additionally, hair can be examined for fungal hyphae indicating presence of dermatophyte.

Lastly, another method is to squeeze the skin as previously described when preparing to scrape but apply acetate tape to area. Tape is then applied to a slide directly—no mineral oil or stain is needed. Mites will adhere to the tape and evaluate with condenser down on 10X.

Biopsy

If you have done the dermatology basics (cytology, scrape and dermatophyte culture) and despite appropriate therapy and addressing the underlying cause, the next diagnostic step may be to collect biopsy samples usually using punch biopsies. These samples can be for culture and/or histopathology. Knowing what the samples are to be used for determines how you prepare them.

Hair is trimmed away from the lesion by using scissors or clippers but clip with grain of hair and not surgically smooth. Do not “shave off” the top of the lesion—the crust and scale are important for the histopathologist to evaluate in addition to the skin.

Have clinician choose biopsy sites. Marking around the area with a Sharpie is helpful. Areas to be biopsied are blocked with lidocaine. Calculate dose of maximum amount of lidocaine for each patient to avoid lidocaine toxicity. May dilute with saline. Inject using 25g needle subcutaneously under lesion. Give it time to take effect!! Skin samples that are for culture are prepped with chlorhexidine. Let dry before sampling. Areas to be sampled for histopathology are not prepped with scrub—the sample should be taken in all of its glory!

A 6 or 8 mm punch biopsy tool is usually adequate to collect most samples. Close with simple interrupted or a cruciate pattern with doctor's choice of suture.

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