Dermatology Diagnostics: Old and New!

Jason B. Pieper, DVM, MS, DACVD® Assistant Professor, Iowa State University

Cytology

The main goal of performing cytology is to evaluate for bacteria, yeast, and inflammatory cells. With the wide variability of clinical lesions to evaluate the skin, there are several different ways that you can take cytology samples. Some of them may be more ideal for certain situations.

<u>Impression Cytology</u>

This is one of the most common methods and is very beneficial for moist and exudative lesions or sampling a pustule.

How to Perform

You will just use the side of the microscope slide that you are planning to view under the microscope and apply it to the area of focus. You will allow the slide to dry completely prior to dipping it into all 3 stains of the Diff-Quik stain.

Tape Cytology

This is a method commonly done to sample dry and scaly areas of the skin.

How to Perform

You will apply clear or transparent tape to the area of focus on the skin several times and then partially adhere it to the slide for staining. Always avoid the fixative solution! You can then dip the tape into the eosinophilic solution I (Xanthene dye) followed by the basophilic solution II (Thiazine dye, Methylene blue) or only the basophilic solution II.

Bread and Butter

This is a method that some will use for lichenified and hyperkeratotic areas of the skin.

How to Perform

You will take a new scalpel blade and lightly scrape the area of interest similar to performing a skin scraping, without the oil. After you get some debris on the scalpel blade, you can then wipe the debris onto the microscope slide in a manner similar to putting butter on some bread or toast.

Cotton Swab

This is a method that is commonly used for areas that are moist and exudative, but also in delicate or tough to reach areas with a microscope slide.

How to Perform

You will take a cotton tipped applicator and roll it onto the skin of the affected area to obtain the sample. Then you will go ahead and roll it onto the microscope slide to apply the sample to the slide.

Skin Scraping

Skin scraping is a technique that people have used for quite some time to evaluate for parasites in the skin. The most important factor is knowing which parasite you are evaluating for and where it normally lives to determine whether to perform a superficial or deep skin scraping.

How to Perform

This is typically done by applying a drop of oil onto a microscope slide and putting the scalpel blade into the oil at first. If you are performing a superficial skin scraping you will perform broad strokes on the affected area without trying to attain any capillary oozing. For a deep skin scraping, you will want to squeeze the affected skin initially, then scrape in a more focused area until you get capillary oozing. With both methods you will want to scoop up the contents with your last scrape and then apply it to the slide and place a cover slip on it.

Tape Squeeze Method

This is a technique that has been described by some people to look for *Demodex* spp. that live in the hair follicle.

How to Perform

Initially you will place a small piece of clear acetate tape onto the affected area of the skin that you want to sample. The area is squeezed for 5 seconds in order to effectively squeeze the demodex mites out of the hair follicles and adhere to the tape. Then you will take the tape and put it onto a slide to evaluate for demodex mites.

Trichogram

A trichogram can also be used to evaluate for parasites when you are uneasy about using a scalpel blade near it. In addition, you can evaluate for hair stage, dermatophytes, trauma induced hair loss and color dilution alopecia.

How to Perform

You will take a microscope slide and put a small amount of oil on the slide. Then you can use hemostats or your fingers to pull the hairs out and place them onto the oil on the slide. Then you will place a cover slip on the slide in order to evaluate it.

Fungal culture

Fungal cultures are most commonly done to evaluate for dermatophytosis in the animal.

How to Perform

If you are evaluating for dermatophytosis, then you will want to take hair samples by plucking them in a sterile manner. If you are sending these hairs out for fungal culture, you ideally want to place them into a sterile tube or petri dish to allow oxygen to the sample. If you wish to perform the dermatophyte cultures in your own hospital, then you will apply the hairs onto the fungal culture media to start incubation. It is important to note that the fungal cultures do NOT need to be placed in a dark area. Fungal cultures should be grown for up to 21 days before being reported as negative.

Fungal PCR

Some laboratories are now performing PCR testing for dermatophytes. An advantage of performing PCR testing is that it is much quicker than traditional fungal culture. A disadvantage is that PCR is unable to detect whether the organisms are dead or alive since it just detects DNA from the organisms.

How to Perform

This is done by plucking at least 10-20 hairs and submitting in a red top sterile tube. Additionally, you can scrape some of the crusts and scale and place these into the tube for testing as well. Turnaround time for this test can be 1-3 days.

Skin Biopsy

Skin biopsies are performed most commonly to have the sample evaluated by a pathologist. Another purpose of performing a skin biopsy is for bacterial or fungal culture.

How to Perform

If you are going to submit the tissue for histopathology, then you don't want to disturb any of the skin you are sampling. Therefore, avoid clippers and scrubbing is important. Choose a large enough punch biopsy though!

If you are performing a biopsy for culture of the tissue, you will then clip the hair close and scrub the tissue prior to biopsying the skin.

Bacterial Culture and Susceptibility

This is a common diagnostic that we have used for some time, but we will discuss it in more detail in the pyoderma lecture.

Bacterial NGS/PCR Testing

This is a newer diagnostic looking at the molecular level of identifying bacteria, while not needing them to grow on a culture plate. This has historically been used quite often for microbiome studies in veterinary medicine, but is used in emergency medicine for humans due to the speed of results.

How to Perform

A culture swab is applied to the skin similar to a bacterial culture and susceptibility, but the companies usually provide a specific swab and transport system with them.