DIAGNOSTIC TECHNIQUES IN SMALL ANIMAL DERMATOLOGY

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Dermatologic conditions among pets are frequent causes of visits to a veterinarian. Distinguishing among these disorders can be a daunting task for a general practitioner. Confirmatory diagnosis requires performing different diagnostic techniques working through diagnostic flow charts and a process of elimination. The most common dermatologic problems include pruritus, alopecia, crusting and scaling, otitis, non-healing wounds, nodules and tumors, and ulcerative disorders.

SIGNALMENT AND HISTORY TAKING

A careful dermatologic history is critical to interpret the physical examination findings and choose appropriate diagnostic tests. A complete general history should be obtained, including information about prior illnesses, vaccinations, husbandry (housing, feeding practices, etc), changes in attitude and food consumption, elimination practices, exposure to other animals, and travel within the past 6–12 mo. This should be followed by a detailed dermatologic history. Use of a preprinted history form can be very useful for chronic or complicated cases. A good history is important, because many skin diseases that look similar are differentiated based on interpreting clinical signs and historical patterns.

When you develop a standardized medical history questionnaire and then use it consistently in your veterinary practice, important historical pieces of information are much less likely to be missed or overlooked. Important questions to have your medical team discuss with the client include the following:

The following information should be obtained:

1) the primary complaint;
2) length of time the problem has been present;

   In general, patients with atopy or inhalant allergies tend to start scratching in an early phase of life and then become progressively worse each allergy season. Food and flea allergies can develop at any point in life. In some regions of the country, flea allergies are seasonal.

3) Signalment

a) age at which the skin disease started

Always consider the age of onset when developing a list of differential diagnoses for a cutaneous condition. Distinct age predilections are seen in many diseases, eg, demodicosis and dermatophytosis in pediatric animals. In general, allergic disease begins in young to middle-aged dogs, whereas most patients with an endocrinopathy may be somewhat older. Most atopic dogs first show clinical signs between the ages of 1 and 3 years.

b) breed

Breed predilections include a predisposition of Cocker Spaniels to primary disorders of keratinization, and of most retrievers and terriers, Lhasa apsos, Shih Tzus, cocker spaniels, Irish setters, German shepherds and English bulldogs to atopic dermatitis.

Food allergy and parasite hypersensitivities (e.g., fleas, *Sarcoptes* or *Cheyletiella* species) can and will occur at any age and in any breed.

4) presence and severity of pruritus

The primary concern of most clients of allergy patients is scratching, licking, rubbing, chewing and discomfort. Information about whether the pruritus is seasonal or year-round, where the pruritus predominantly occurs (e.g., head and neck, sides and interdigital region) and how often the patient truly is scratching should be collected.

5) Onset of pruritis

Onset of pruritis should be recorded. Pruritus after development of rash may be due to infectious, parasitic or immune-mediated causes, whereas allergic patients often scratch first and develop a secondary infection with associated lesions.

6) how the disease started and its progression

Diseases that begin with pruritus may lead to self-trauma and subsequent development of secondary skin lesions such as alopecia and seborrhea or infections such as bacterial or yeast pyoderma

7) type and progression of lesions noted by the owner;

8) evidence of seasonality

9) area on the body the problem was first noticed

Regional patterns are seen in atopic dermatitis where typically the face and feet are affected, in cheyletiellosisit is primarily dorsal, in scabies it is primarily ventral,

and in endocrine hair loss it usually involves the trunk and spares the head and leg;

10) any previous treatments and the responses to such

Antibiotic-responsive skin diseases suggest a bacterial cause while pruritus that responds to small doses of glucocorticoids, antihistamines, or essential fatty acids suggests allergic dermatitis.

11) frequency of bathing and when the last bath was given

Recent bathing may obscure or change important clinical lesionseg., flea dirts will be washed of when given bath. Further excessive bathing and wetting of the skin can predispose to skin disease.

12) presence of fleas, ticks, or mites;

13) other contact animals

Evidence of contagion should be recorded as it suggests fleas, scabies, cheyletiellosis, or dermatophytosis.

14) the environment of the animal

Housing changes can influence the development of certain skin diseases, eg, contact dermatitis, contagious diseases

15) physical signs or reports of systemic illness

Change in appetite, increase in water consumption or urination or exercise intolerance may suggest other underlying diseases or disorders, as can as a patient seeking or avoiding heat. Endocrine [eg, hypothyroidism and hyperadrenocorticism] disorders and metabolic diseases [eg, diabetes mellitus, renal disease, liver disease] should be noted, because the skin can be the first place signs of systemic illness are noted).

16) Patient's medical history

It is important to fully review all past diagnostics and therapies and the success or failure of those therapies.

I. SKIN SCRAPINGS

INDICATIONS: For diagnosis of external parasites like Sarcoptes, Demodex, and Cheyletiella. False-negative results are possible.

PROCEDURE:



Superficial skin scraping

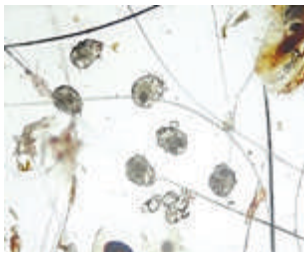
i) To identify **Sarcoptes mites** superficial scrapings of crusted, papular, or alopecic lesions on elbows, pinnal margins, and the ventral trunk are ideal. However, as Sarcoptes mites live in the stratum corneum and are often few in number false negative scrapings are common. So any animal with pruritus consistent with scabies should be trial-treated with appropriate acaricidal therapy.

a

ii) In cats superficial scrapes are used to detect *Demodex gatoi* and *Notoedres cati*

Deep skin scraping

Deep scrapes are largely used to diagnose dogs with Demodex canis mites and cats with Demodex cati mites



b

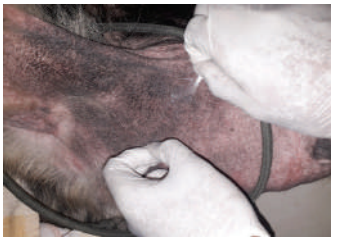
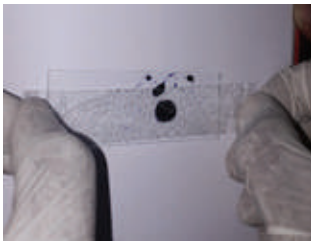
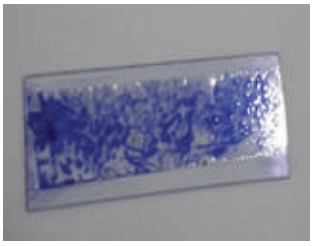
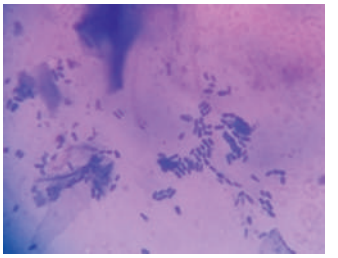
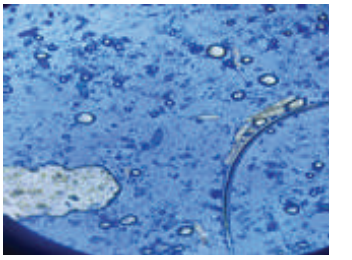
a. **Demodex spp.** in a canine deep skin scrapping

b. **Notoedres mites (adults and eggs)** in a feline skin scrapping

II. TAPE IMPRESSION

INDICATIONS: Acetate tape impressions is indicated when Cheyletiella mites, Demodex mites or Malassezia dermatitis are suspected. It can be used to sample dry, lichenified, and interdigital areas.

PROCEDURE:

Malassezia identified as round to oval budding yeasts under high-power field

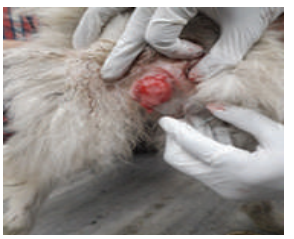
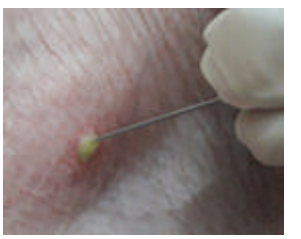
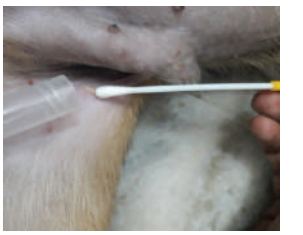
Demodex mites observed in a canine tape impression stained with LCB.

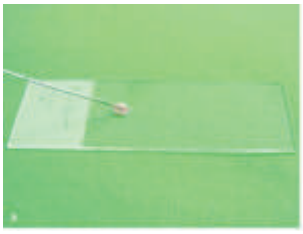
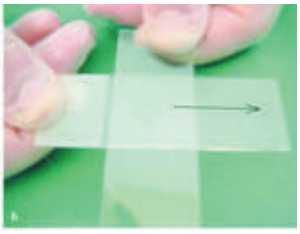
III. SKIN CYTOLOGY

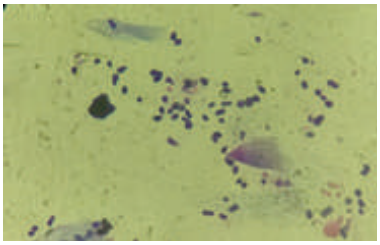
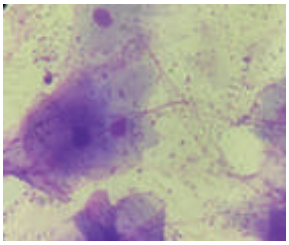
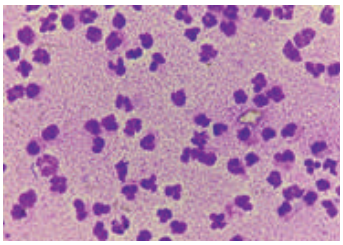
Sample collection : Direct impression, swab collection or fine needle aspiration (depending on the type of lesion)

INDICATIONS: Skin cytology stained with Diff-Quick, can be used to obtain information on bacterial or Malassezia infection, as well as to characterize inflammatory infiltrate of skin and ear affections. In addition, swab samples can be sent for culture and sensitivity testing.

PROCEDURE :

Fungal spore in a ear swab stained with Diff-Quick

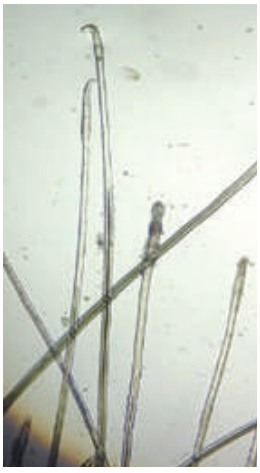
Keratinized cells surrounded by bacterial cocci

Neutrophils surrounded by bacterial cocci

IV. TRICHOGRAM

INDICATIONS: minimally invasive procedure in confirming self-trauma, especially in cases of feline alopecia; detecting Demodex mites and dermatophytosis; determining hair growth cycles; investigating hair follicle and shaft disorders

PROCEDURE:




Growing, anagen hairs - Anagen-phase bulbs are rounded, smooth, shiny, glistening and soft so the root may bend. They are fairly tightly attached to the surrounding skin.

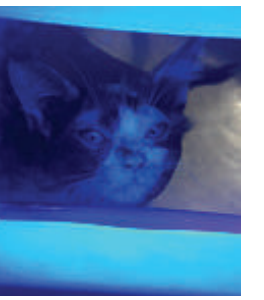
Resting, telogen hairs - Telogen bulbs are club- or spear-shaped with a rough surface. More loosely attached to the skin and epilate more easily. A sample with exclusively or mostly telogen hairs points to an endocrine disorder or follicular arrest.

If the animal is pruritic and licks the hair off (or if the hair shafts are damaged due to dermatophytes), the tips of the hairs are broken off. If the hair falls out for other reasons, the tips are tapered.

V. WOOD'S LAMP EXAMINATION

INDICATIONS: Wood's lamp evaluation for dermatophytosis is an easy and useful tool. It is a good screening test, although false-positive and false-negative results often occur. Only 30% to 80% of *Microsporum canis* isolates fluoresce, and *Microsporum gypseum* and *Trichophyton mentagrophytes* do not fluoresce under a Wood's lamp.

PROCEDURE:



INTREPRETATION

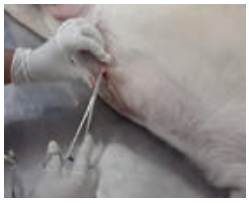
Hairs invaded by M. canis may show a yellow-green fluorescence. This fluorescence runs along the hair shafts rather than fluorescing on discrete, individual, occasional scales, as may be seen in normal animals and humans.

VI. SKIN PUNCH BIOPSY

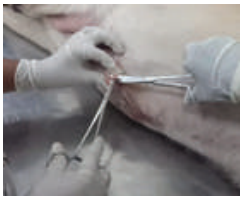
INDICATIONS: Acute and severe lesions, Neoplasia (nodule, chronic non-healing ulcerative lesion), Unusual skin lesions, no response to an appropriate therapy.

PROCEDURE : Mark the required area. Trim hair with scissors or clippers. For Local anesthetic effect 0.5 to 1ml of 2% lidocaine/site can be injected into the subcutis under or around the biopsy site. General anesthesia may be necessary when taking biopsies from mucocutaneous junctions, nasal planum, pawpad and pinna. Allow 3 to 5 minutes for the local anesthetic to have effect. Use a 6 or 8 mm punch biopsy on most cases.

Position the punch biopsy instrument at right angles to the surface of the skin over the center of a lesion. With firm continuous pressure the punch is rotated in one direction until the dermis is free from its underlying attachment. The punch is removed and any blood should be carefully blotted.



Support the tissue at the base and cut free with iris scissors. Blot on a gauze to remove excess blood.

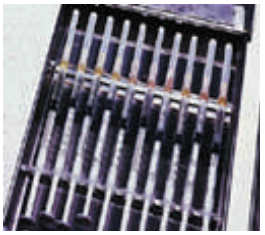


The biopsy sample should be placed in 10% formalin immersion. Close with one cruciate or two simple interrupted sutures.

VII. INTRADERMAL TESTING

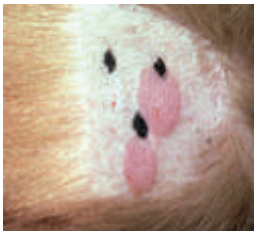
INDICATIONS: After a clinical diagnosis of atopic dermatitis has been made and you want to identify the relevant allergens to include in an allergy vaccine, as an adjunct test to confirm flea allergy in a dog

PROCEDURE : If necessary sedate the dog with medetomidine or propofol. Clip the hair from the lateral thorax. Mark the sites with a permanent marker pen. Inject approximately 0.05 ml of the negative and positive control controls as well as each allergen. Reaction sites are examined 10 to 20 minutes later and evaluated for size, erythema and turgor and graded from 0 to 4+. This test will not diagnose the disease but should only be used for those patients where immunotherapy is going to be used to treat atopic dermatitis. Most dogs with flea allergy will show an immediate reaction but some will only have a delayed (24-48 hour) reaction.

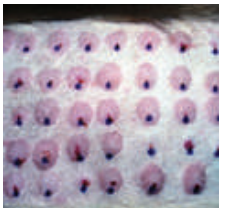


Multiple syringes filled with allergens for intradermal testing

Injecting allergen into the dermis



Positive intradermal test to flea allergen ((-) and (+) controls on the top row)



Multiple positive reactions in a dog with atopic dermatitis

VIII. FUNGAL CULTURE

Indication

Fungal cultures are an important diagnostic tool that can be easily performed in a general practice setting for differential diagnoses of patients with clinical signs consistent with fungal skin infection (eg, multifocal patchy alopecia, crusts, scales, pustules, papules), as well as for patients with similar clinical signs (ie, demodicosis, pyoderma) that fail to respond to appropriate therapy.

Procedure Sample collection

Hair pluck

Using a sterile hemostat, pluck hairs (≈10-20) from the periphery of lesional skin; the hair bulb and root must be intact. Gently place plucked hair on the culture medium. Ensure the plucked hair remains in place and in contact with the agar by applying gentle pressure with the sterile haemostat

Mackenzie Toothbrush technique

Brush a sterile toothbrush through the animal's hair coat, against the direction of growth, for approximately 30 strokes. Lightly press the bristles onto the culture medium

Incubation in culuture media

Incubate the closed culture plate in a warm (ie, 75°F-86°F [24°C-30°C]), dark area for 21 days. Place a small dish of water nearby to provide humidity and prevent the medium from drying out. Check the medium daily for fungal growth.

Interpretation

Macroscopic fungal colony morphology

Microsporum and *Trichophyton*species—the most common dermatophytes in dogs and cats—are white, light yellow, tan, or buff- colored cottony-to-powdery-appearing colonies

Dermatophyte colonies are never black, green, or gray.

Microscopic identification

Gently touch a small piece of clear acetate tape to the surface of the fungal colony.

Apply the tape to a glass slide over a drop of blue stain (methylene blue, lactophenol cotton blue, or the blue Diff-Quik solution [basophilic thiazine dye]).

Examine the slide under 100X to 400X magnification to identify the characteristic dermatophyte macroconidia.

Microsporum canis spores are large, spindle-shaped, and thick- walled with six or more internal cells and often have a terminal knob. If *M. canis* is identified, then other animals in the household should be screened via dermatophyte culture using the toothbrush technique to determine whether they are asymptomatic carriers. All pets with positive culture results should be treated with topical antifungal therapy, with or without systemic treatment. Culture- positive animals should be isolated from culture-negative animals if possible.

Microsporum gypseum produces large spindle-shaped spores with thin walls, no terminal knob, and six or fewer internal cells.

Trichophyton mentagrophytes produces long cigar-shaped macroconidia with thin walls. Spiral-shaped hyphae and numerous small grapelike clusters of microconidia are also characteristic of *Trichophyton* species.

IX. COAT BRUSHINGS

Indication

To rule out parasitic infestation like Flea, Furmite and Cheiliteilla sp

Procedure

- ❖ Gently brush the coat with a tootbrush or flea comb and collect the material in a clean sheet.
- ❖ Place the debris collected on to a slide wit mineral oil and view under 10x

X. DIET TRIAL TO IDENTIFY FOOD ALLERGIES

Diagnosis of an Adverse food reactions (AFRs) goes hand in hand with treatment because confirmation of disease is based on response to therapy. Confirmation of an AFR depends on reduction or resolution of clinical signs while the animal is being fed a strict elimination diet, recurrence of clinical signs when the animal is challenged with the original diet (and anything else given orally), and resolution of signs after the elimination diet has been reinstated.

A complete diagnostic elimination-challenge diet trial (ECDT) can be considered a 4-phase process—**eliminate, challenge, confirm, and identify**.

Phase 1: Eliminate

This initial phase involves strictly feeding only the trial diet for up to 12 weeks while monitoring for reduction of clinical signs; no other treats, supplements, capsules, toothpastes, dental chews, outdoor hunting, or scavenging are allowed.

Phase 2: Challenge

This phase involves reintroducing the previous diet while monitoring for a flare of clinical signs. If the pet has an AFR, signs usually recur within 2 to 3 days but may take up to 2 weeks.

Phase 3: Confirm

This phase involves restarting the strict elimination diet; resolution of clinical signs confirms the diagnosis of an AFR. Confirmation may take 2 to 4 weeks while clients monitor for reduction of clinical signs. If the elimination diet is appropriate and balanced for the patient, it may be continued indefinitely.

Phase 4: Identify

This final phase of the ECDT is intended to identify the specific ingredients that cause a flare of signs and that should be avoided. The patient continues eating the strict elimination diet while being offered previously fed ingredients (usually proteins, which are the most commonly problematic) as treats or diet toppers. Small amounts (<10% of caloric intake) of 1 ingredient at a time are offered for up to 2 weeks while the pet is monitored for flare of signs. If no signs are noted, the ingredient can be fed; if signs are noted, the ingredient should be avoided. Completing phase 4 helps determine problematic foods to avoid, which significantly improves prognosis.

XI. DIAGNOSIS OF CUSHINGS SYNDROME

- **Clinical signs :** Dermatological signs : bilateral symmetric endocrine alopecia and thinning of skin, recurrent pyoderma, calcinosis cutis. Systemic signs : Polyuria and polydipsia, abdominal distention and panting
- **History:** Prolonged usage of glucocorticoids, either as injectable, orally or topically?
- **Endocrine Diagnostics : Low-dose dexamethasone suppression (LDDS)**
- LDDS demonstrates decreased pituitary sensitivity to negative feedback from glucocorticoids. Screening test of choice for hyperadrenocorticism unless the history suggests iatrogenic causes, in which case ACTH stimulation is preferred.

Procedure: Blood samples are obtained (1) before, (2) 4 hours after, and (3) 8 hours after dexamethasone administration (0.01 mg/kg IV). Diagnosis of HAC is based on lack of suppression of cortisol concentration 8 hours after dexamethasone administration.

Interpretation

- Normal dogs: Causes rapid and prolonged suppression of cortisol secretion (< 1.1 µg/dL)
- Criteria for identifying dogs with PDH included:
- 4-hour post LDDS cortisol concentrations < 1.1 µg/dL or < 50% of basal cortisol concentration
- 8-hour post LDDS cortisol concentrations < 50% of the basal cortisol concentration and >1.4µg/dL
- Patients with an adrenal tumor : At any dosage does not suppress cortisol secretion

ACTH Stimulation

ACTH stimulation assesses adrenocortical reserve. Due to greater purity and quality control, use of synthetic ACTH is recommended.

Indications. ACTH stimulation is the gold standard for diagnosis of iatrogenic HAC and spontaneous Addison's disease (hypoadrenocorticism). Diagnosis of HAC is based on finding an elevated cortisol concentration (post ACTH administration) based on the reference range established by the laboratory.

XII, DIAGNOSIS OF HYPOTHYROIDISM

Dermatologic changes are the most common sign of canine hypothyroidism, which includes bilaterally symmetrical, non-pruritic alopecia, mainly affecting the trunk; dry and lustreless coat; seborrhea (greasy, scaly and malodorous); secondary bacterial and fungal infections; thickening of skin and facial folds (resulting in the well-described "tragic" facial expression); clipped areas fail to re-grow properly; poor wound healing and easy bruising. Other signs related to decreased metabolic rate include lethargy or dull mentation, inactivity or unwillingness to exercise, weight gain, cold intolerance or heat seeking

Endocrine Diagnostics

Total T4 concentrations up to 1.0 µg/dL may be consistent with hypothyroidism.

Free T4, the biologically active form of T4, most accurately reflects thyroid function. It can be used as the initial diagnostic test for hypothyroidism or to further evaluate thyroid function when the TT4 is borderline. (normal range: 0.6 – 3.0 ng/dL)

Concentrations of **TSH** are increased (>0.6ng/ml) in most of the hypothyroid dogs, however a significant proportion will have normal TSH levels