

  
**Diagnostic Tests**
  
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### Dermatologic Diagnostic Tests


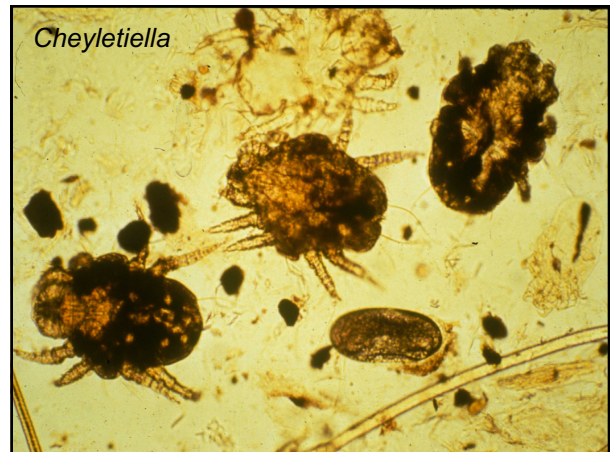
- Flea combing
- Skin scraping
- Trichogram
- Wood's lamp evaluation
- Fungal culture
- Skin cytology
- Ear cytology
- Bacterial culture and sensitivity
- Skin biopsy

### Diagnostic Screening Tests

<u>Scraping</u>	<u>Cytology</u>	<u>Culture</u>
<b>Mites</b>	<b>Bacteria, yeast, cells</b>	<b>Bacteria + fungi</b>
Deep	Surface	Bacterial
Superficial	Cotton swab	Mycobacterial
	Tape	Fungal
	Impression	
	FNA	

### Flea Combing

- Indications: pruritus +/- alopecia
- Identify fleas, flea "dirt," *Cheyletiella*
- Technique
  - Comb with grain of hair to collect scale and debris
  - Place dark material on damp paper towel to examine for blood
  - Place contents into a drop of mineral oil and examine at 10x with condenser down

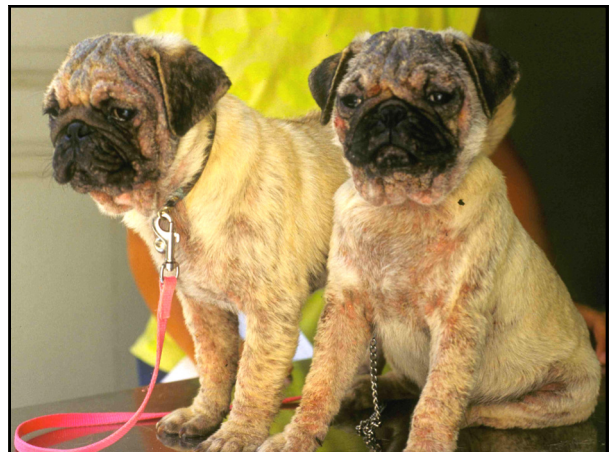
## Skin Scraping - Materials

- Indications: alopecia +/- pruritus
- Identify *Demodex*, *Sarcoptes*, other parasites
- Scissors or clippers (if long coated)
- Scalpel blades (#10) or spatula
- Mineral oil
- Glass slides
- Cover slips
- Marker or pencil



## Skin Scraping - Deep

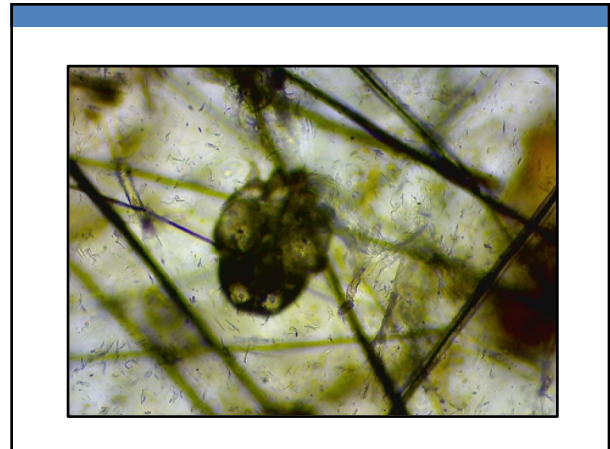
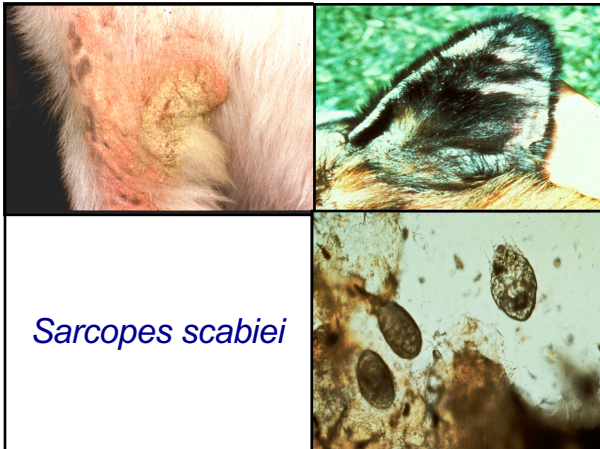
- 3-4 sites
- Representative sites
- Newer lesions
- Avoid excoriated areas
- Small focal area
- Pinch and release the skin
- Induce capillary hemorrhage
- Place material into a drop of mineral oil and examine at 10x with condenser down





## Skin Scraping - Superficial

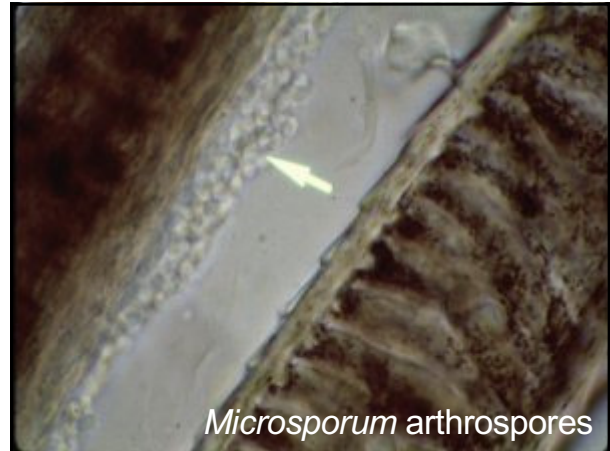
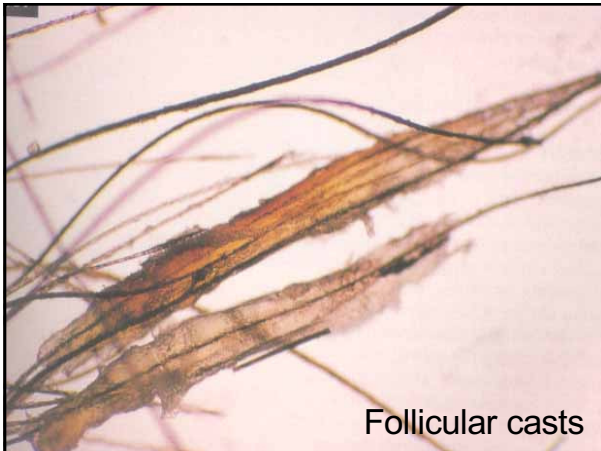
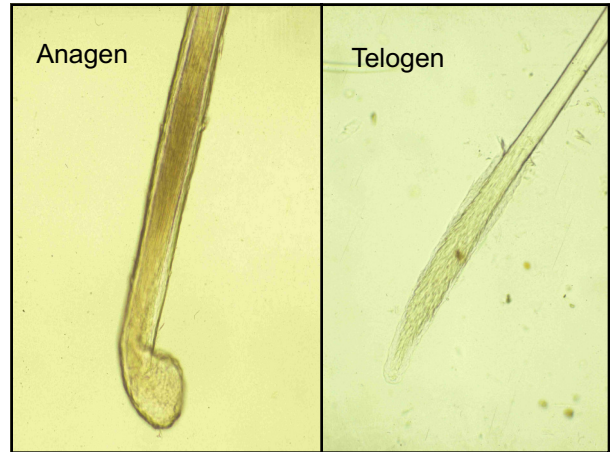
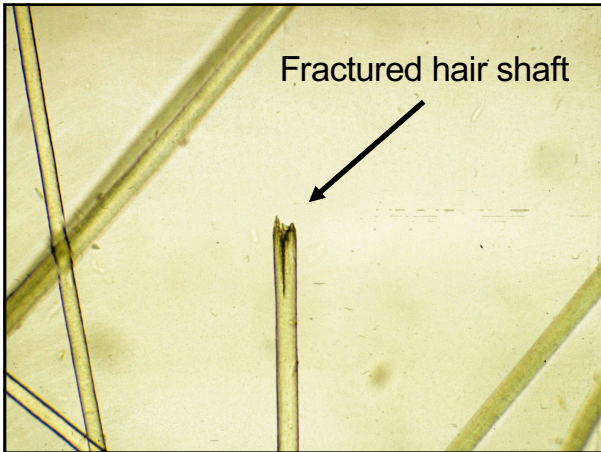
- Same general technique as deep scrapings
- Use broader strokes
- Don't need to pinch and don't need blood
- Best technique for finding *Sarcoptes*, *Cheyletiella*, lice



## Trichogram – “Hair Pluck”

- Indications: hair loss, abnormal hair coat
- Identify self trauma, growth phase, arthrospores (ringworm), parasites (*Demodex*, *Cheyletiella*, lice)
- Technique
  - Pluck hairs from periphery of lesion, place in mineral oil, and examine at 10-40x with the condenser down
- Look at hair bulb, distal end of hair, as well as hair shaft





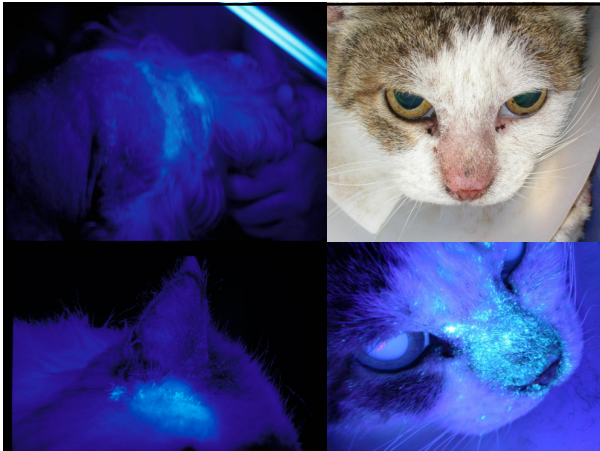
- ### Wood's Lamp
- Indications: dermatophytosis suspect
  - UV light lamp of 360 nm wavelength
  - Plug in model with magnification preferred
  - Darken room
  - Examine at distance of 4-10 cm
  - Must take time to thoroughly examine hair coat
  - Need to be trained to recognize positive hairs

Burton UV  
Wood's light  
with  
magnification



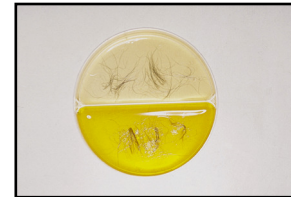
## Wood's Lamp

- *M. canis* strains **commonly** fluoresce apple or blue green at shaft
- False positives
- Identify infected sites that are not visible on gross clinical examination
- Valuable to select hairs for direct examination and culture
  - Positive hairs examined by direct examination may **confirm** an infection



## Fungal Culture

- Indications:
  - alopecia,
  - folliculitis,
  - furunculosis,
  - human infection
- Identify dermatophytes
- Definitive test

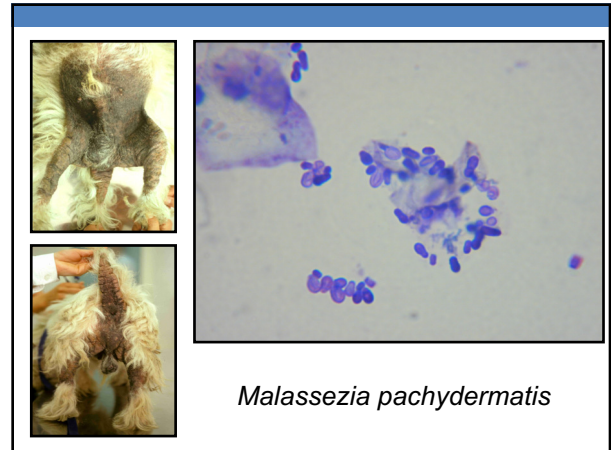
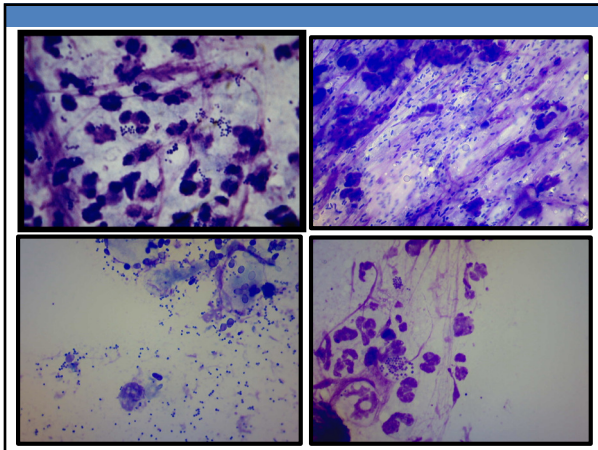


## Skin Cytology Impression Smear

- Indications: alopecia, pruritus, erythema, exudate etc.
- Identify inflammatory cells and organisms that will aid in diagnosis
- Touch slide directly to lesion if greasy or moist
- Use acetate tape if lesion is dry
- Briefly heat fix (if greasy), stain with Diff-Quik®, and examine at 100x

## Skin Cytology Acetate Tape Prep

- Indications: pruritus, alopecia, erythematous, moist, dry or greasy skin surface, skin folds
- Identify *Malassezia* > bacteria, inflammatory cells
- Technique
  - Apply sticky side of clear acetate tape repeatedly to lesion
  - Apply 1-2 drops of Diff-Quik® basophilic (#3) stain beneath tape and stick to slide or stain like a glass slide
  - Examine at 100x

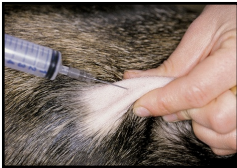


### Fine Needle Aspirate

- Indications: nodules or plaques
- Identify inflammatory and neoplastic cells as well as infectious organisms
- Technique: aspiration vs. needle "core biopsy"

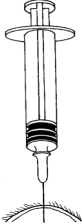
Aspiration

- 6 ml syringe with 20-22G needle
- Insert needle into the lesion

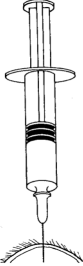


- Aspirate about 1 ml
- Move needle in the lesion without extracting it
- Release vacuum, extract needle from lesion

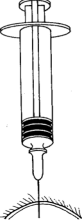
**A**



**B**



**C**



(A) insert  
(B) aspirate  
(C) release (repeat)

*Do not aspirate air!*

### Fine Needle Aspiration

- Remove needle and add ~2 cc of air into the syringe
- Replace needle and push contents on a clean glass slide



### Fine Needle Aspiration

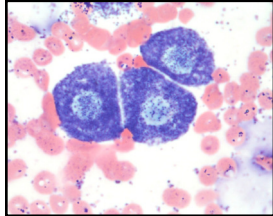
Needle core technique

- Insert the needle into the lesion
- Rotate needle on axis and redirect while within the lesion
- Repeat insertion
- Add ~2 cc of air to a 3 or 6 cc syringe
- Place needle and push contents on a clean glass slide



## Fine Needle Aspiration

- Stain in Diff-Quik®
- Scan at low magnification to identify “busy areas” of slide
- Examine at 100x

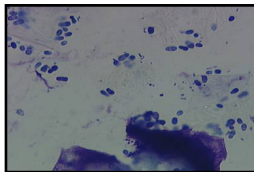
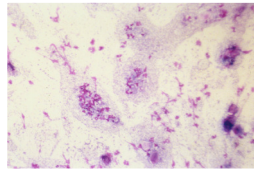


## Ear Cytology



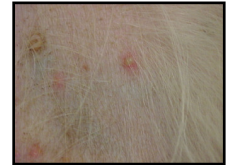
## Ear Cytology

- Scan at low magnification (10x) for keratinocytes and/or inflammatory cells
- Evaluate further at 100x



## Bacterial Culture and Sensitivity

- Why Perform?
  - Identify the agent causing infection
  - Perform antimicrobial susceptibility to evaluate for drug resistance and select the appropriate therapy



## Culture: Indications

- Recurrent pyoderma
- Pyoderma previously treated with multiple antibiotics
- Pyoderma unresponsive to empirical treatments
- Deep pyoderma
- Pyoderma with cytological evidence of rods
- Nodular lesions with inflammatory cytology



## Culture: Materials

- Sterile swabs with transport medium
- Needles, syringes, sterile vials
- Sterile saline solution (0.9% NaCl)
- Sterile surgical set including scalpel blades
- Submission sheet for the lab
- Deliver to lab within 48 hours

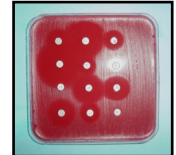


## Lab Choice and Advice

- Use a veterinary lab with ability to isolate methicillin resistant Staph bacteria
- Inform the lab if the dog is currently treated with antibiotics
  - Longer incubation in enriched culture media
- Inform the lab of the cytology findings (cocci, rods, mixed infection)
  - Influences culture media and selection of relevant colonies for susceptibility testing

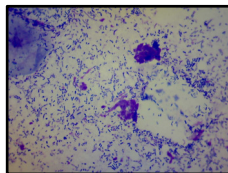
## Susceptibility Testing

- Methodology
  - Kirby-Bauer diffusion disks (acceptable)
  - Minimum inhibitory concentrations (better)
- Information needed from laboratory
  - Bacterial speciation
  - Methicillin/oxacillin resistance or susceptibility



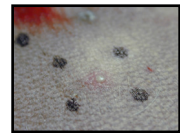
## Skin Culture

- Always correlate results with cytology
  - Cocci bacteria → *Staphylococcus pseudintermedius*
  - Rod bacteria → *Pseudomonas* sp or *Proteus* sp



## Skin Culture

- Superficial skin lesions
  - Pustules
  - Epidermal collarettes
- Deep skin lesions
  - Draining tracts
  - Dermal or subcutaneous nodules/plaques
  - Fluctuant dermal or subcutaneous lesions



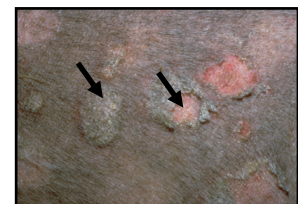
## Culture - Superficial Lesions

Pustules are the ideal lesion to culture!



## Culture - Epidermal Collarettes

- Clip the hair if necessary
- Find a newly developed epidermal collarette
- Do not clean lesion
- Rub a dry sterile cotton swab across the collarette 3 to 4 times
- Place swab in transport media
- Sensitivity: 81.8%
- Specificity: 100%



White et al. JAVMA 2005

## Culture - Deep Lesions

- Three methods
  - Culture fresh exudate
  - Culture dermal/subcutaneous tissue
  - Culture pocketed dermal/subcutaneous fluid by fine needle aspiration



## Culture - Exudate from a Draining Lesion



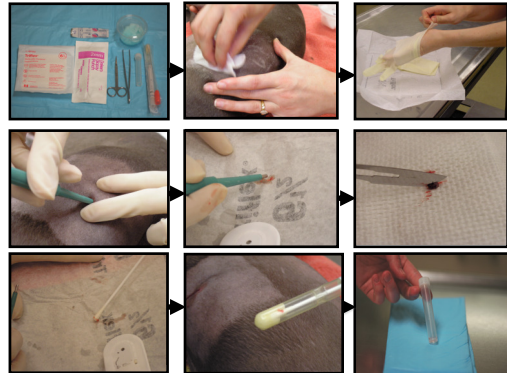
- Clip the hair if necessary
- Clean the skin surface
- Fold and pinch the skin to extrude fresh exudate from the deep dermis
- Use mini-tip swab to sample
- Place sample in transport media

## Culture – Deep Tissue



Ideal method when suspect a deep skin infection

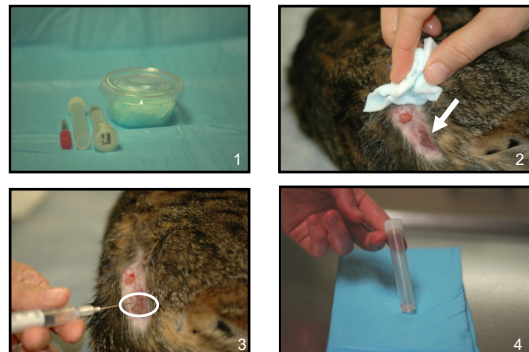
## Culture: Deep Tissue Biopsy



## Culture - Tissue Fluid

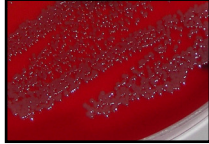


## Culture: Tissue Fluid via FNA



## Culture - Limitations

- Problems with transportation → bacterial death
- Selection of non-pathogenic bacteria
- Errors in sampling
  - Residential flora or contaminants
  - Concurrent systemic and/or topical antibacterial therapy

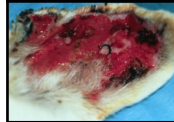


## Culture - Key Points

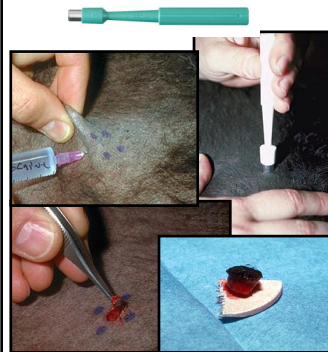
1. Clip the hair
2. Clean deep lesions and adjacent skin gently with 70% alcohol or diluted chlorhexidine
3. Do not clean skin prior to sampling superficial lesions (pustules, epidermal collarettes)
4. Use a mini-tip culturette to avoid touching the surrounding non-lesional skin
5. Use a transport media to keep the organisms viable
6. Do not use local lidocaine when collecting tissue samples as it may inhibit microbial growth
7. FNA should be used for dermal or subcutaneous lesions with pockets of fluid

## Skin Biopsy

- Why perform?
  - Tissue sample for histopathology
  - Tissue sample for culture
- Indications
  - Neoplasia suspect
  - Ulcerative disease
  - Severe acute generalized disease
  - Mucosal lesions
  - Footpad lesions
  - Nodular lesions
  - Lack of diagnosis after a complete workup



## Biopsy - Techniques for Histopathology



- Use largest punch possible
- DO NOT prep, scrub or disturb the lesions
- Gently clip the hair
- Local infusion with 2% lidocaine and bicarbonate
- Handle sample as carefully as possible

## Biopsy – Key Points

1. Treat infection before performing biopsy
2. Discontinue glucocorticoids: oral 3-4 weeks; repository: 6-8 weeks
3. Gently clip the hair if needed – do not scrub!
4. Preferably use a 6 mm or 8 mm biopsy punch and rotate in one direction only
5. Inject lidocaine subcutaneously; may use ring block depending on location
6. Collect multiple samples that represent various lesion stages including crusts

## Histopathology – Key Points

1. Immediately fix in 10% formalin (1:10 tissue to formalin volume ratio)
2. Use pathologist with interest in dermatology
3. Provide written details
  - a. Signalment
  - b. History
  - c. Lesion description
  - d. Your differential diagnoses

