

# Don't fear the yucky ear! Using in house diagnostics to target treatment of otitis externa.

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#### **Conflict of Interest Disclosure:**

I have financial interest, arrangement or affiliation with: IDEXX honorarium for this lecture

All photographs and microscope images are from my personal collection or are from the Texas A&M Veterinary Teaching Hospital collection.

IDEXX inVueDX™ Cellular Analyzer images are courtesy of IDEXX.



## **Conflict of Interest Disclosure:**

The information contained herein is intended to provide general guidance only. As with any diagnosis or treatment you should use clinical discretion with each patient based on a complete evaluation of the patient, including history, physical exam and presentation, and laboratory data. With respect to any drug therapy or monitoring program, you should refer to applicable product insert(s) for complete description of dosage, indications, interactions, and cautions. Diagnosis, treatment, and monitoring should be patient specific and is the responsibility of the veterinarian providing primary care. (2024)



# Learning Objectives

Recognize the significance of common otic pathogens and inflammatory cells.

- -Relate cytology results to the otic examination
- -Design a treatment plan for otitis externa using cytology and the otic examination
- -Consider cases that may benefit from advanced diagnostics and referral



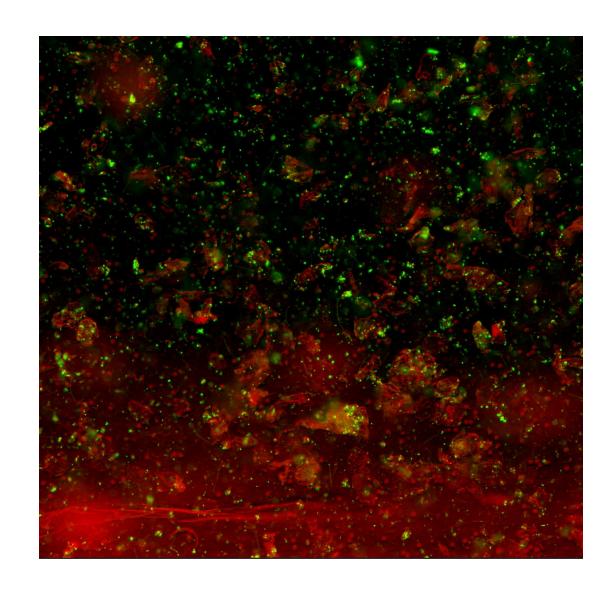
## **Disclosures**

This lecture is sponsored by IDEXX and Merck Animal Health.

I have previously given sponsored lectures for Dechra and Vetoquinol. I will discuss ototopicals that are manufactured by these companies.

All photographs and microscope images are from my personal collection or are from the Texas A&M Veterinary Teaching Hospital collection.

IDEXX inVueDX<sup>™</sup> Cellular Analyzer images are courtesy of IDEXX.



# Ear Cytology Collection and Staining

#### Sample Collection

Cotton tip applicator (CTA) placed into the ear canal until you reach the end of the vertical canal Spin the CTA along the long axis gently, then remove

\*\*In cases of inflammatory otitis, where otoscopic examination is impossible or impractical, you can still collect an ear cytology

#### Staining

Diff Quick ® modified Romanowsky stain

- 1. Fixative (light blue)-Methanol (30 sec)
- 2. Eosinophilic(red)-Xanthene dye (60 sec)
- 3. Basophilic(dark blue/purple)- Methanol Blue/Azure A (60-90 sec)



# Microscope Set up

## Ear Cytology Evaluation

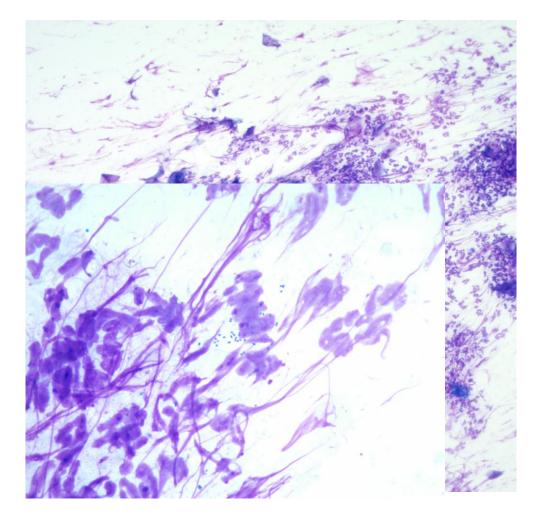
Clean, dedicated microscope used for fine needle aspirates, ear cytology, and skin cytology

Start on 10x objective with the condenser raised and the light at a medium intensity

Locate the area of interest on the slide by scanning for clumps of blue/purplish stain.

Move to 100x objective using immersion oil, keep the condenser raised, and increase light intensity if needed.

Scan through ~10 fields on 100x objective to evaluate for yeast, bacteria, fungal hyphae, and inflammatory cells



10x objective, neutrophils, keratinocytes

## The Normal Ear Canal

The normal ear canal is temperature, pH, and humidity regulated.

Normal ear canals rely on the lipid content of cerumen, host defense(antimicrobial) peptides, and regulated desquamation to maintain homeostasis.



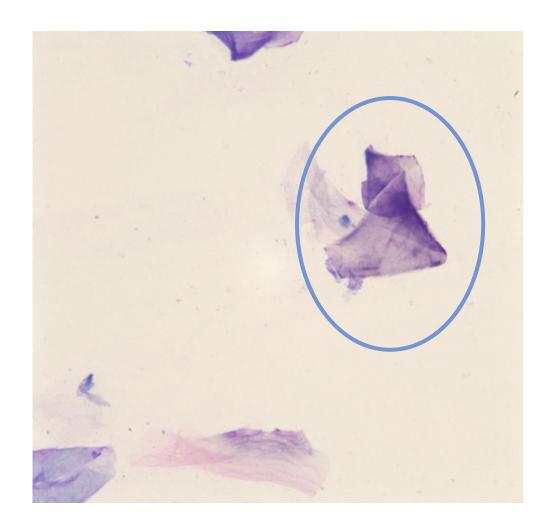
# Impression Cytology of the Normal Ear

Unstained samples will appear nearly clear and slightly greasy

After staining it is still nearly clear since the lipid does not take up much stain

Few anucleated keratinocytes(squamous cells) will pick up purple stain

\*\*\*A few yeast or cocci adhered to squamous cells is within the realm of normal



100x objective, few keratinocytes

## **Melanin Granules**

Ovoid/rectangular

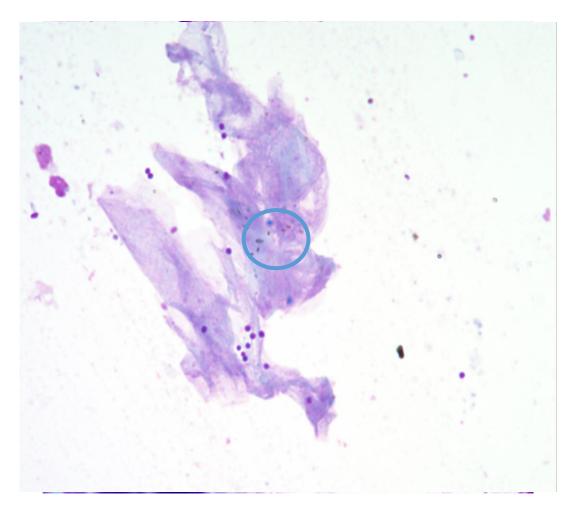
(Staphylococcal bacteria are perfect circles)

Melanin granules are brown/gold and slightly refractile

(Staphylococcal bacteria are blue/purple)

Melanin granules are always inside keratinocytes

(Staphylococcal bacteria may be adhered to keratinocytes or free in cerumen)



100x objective, melanin granules, cocci bacteria

# Primary problem- What started this mess?

#### Part 1

- Allergic skin and ear disease
- Endocrine disease
- Disorders of keratinization
- Ectoparasites
- Immune mediated diseases
- Foreign bodies/ Masses

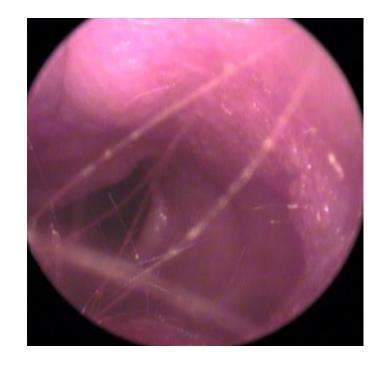
\*\*Dogs and cats can have more than one primary cause for their otitis

# **Allergic Otitis**

"The Negative Cytology"



Erythema, stenosis



Erythema, stenosis, nodular hyperplasia

Erythema, stenosis, epithelial hyperplasia

## Inflamed ear canal

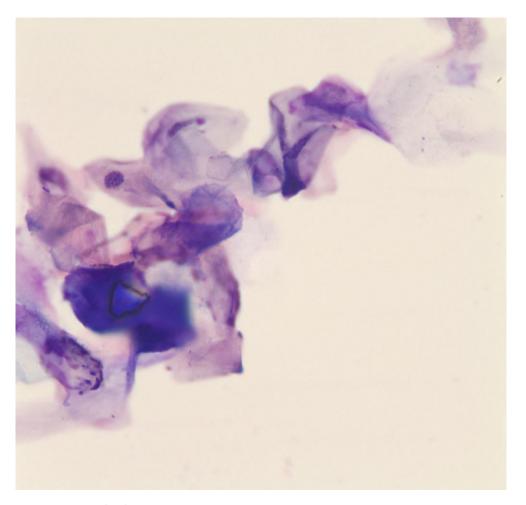
'Nothing to see here'

Increased number of keratinocytes

- anucleate and nucleated keratinocytes

The cerumen has a lower lipid content -potential for more blue/purple stain uptake

\*\*More chronic cases may have low numbers neutrophils or proteinaceous debris



Variably stained keratinocytes

# Allergic Otitis-flea, food, atopy (environmental)

## Diagnosis

- Is the patient on excellent year-round flea/ectoparasite control?
- Are the clinical signs(otitis, body itch etc) year-round, seasonal, sporadic?
- If ANY signs of itch are non seasonal, has a strict prescription diet trial been performed?
- -Dogs/Cats with no response to prescription diet trial while on flea/ectoparasite control likely have environmental based allergy.

## **Treatment**

Flea allergy = year-round treatment for affected pet and house mates

Food allergy = continue diet that does not lead to flares

# **Allergic Otitis**

## Systemic Treatment

Corticosteroids-Best for acute flares and management of nodular hyperplasia

Modified Cyclosporine- Good for management of chronic inflammation along with ototopical

Oclacitinib – mixed response for management of chronic inflammation along with ototopical

## **Ototopical Treatment**

Higher Potency Corticosteroids

- -Fluocinolone acetonide 0.01 % and DMSO 60%
- -Hydrocortisone aceponate
- -Mometasone
- -Dexamethasone SP 4mg/ml

Lower Potency corticosteroids

- -Burrows Solution with Hydrocortisone
- -acetic acid 1%, 0.15% ketoconazole, 1% hydrocortisone flush

## Keratinaceous/Ceruminous otitis

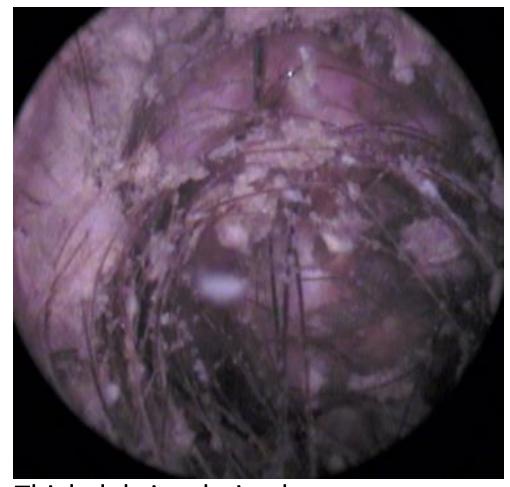
## Increase in keratinocytes without inflammatory cells

#### Keratinization disorders

- Vitamin A-responsive Dermatosis (Cocker Spaniels)
- -Primary seborrhea
- Sebaceous Adenitis (Poodles, Akitas, Vizsla)

#### **Endocrine**

- -Hypothyroidism
- -Hyperadrenocorticism



Thick debris admixed with hair

## **Keratinaceous/Ceruminous Otitis**

## Diagnosis

- The clinical signs do not support allergic skin and ear disease OR allergic disease is well controlled
- Endocrine testing performed when the patient is not inflamed or infected is consistent with hypothyroidism or hyperadrenocorticism
- Breed, clinical signs +/- skin biopsy have diagnosed sebaceous adenitis or Vit A responsive dermatosis

## **Treatment**

#### Keep the pH of the ear canal acidic

- -Acetic acid based branded flushes
- -1:1 water/ white vinegar(5-6%)

# Soften/break down waxy accumulations

- Phytosphingosine
- Propylene glycol (sebaceous adenitis)
- -Squalene (severe build up of debris)

## **Secondary and Predisposing Factors**

#### Part 2

- Secondary infectious otitis externa is most often caused by Malassezia yeast, cocci bacteria (Staphylococcus, Streptococcus), and rod bacteria (Pseudomonas, Proteus, Corynebacterium)
- Noninfectious secondary factors include Dry overcleaning (either with dry cotton or gauze), wet overcleaning (water-based cleaners), ear canal irritants(low pH products, alcohol)

Predisposing factors such as hair type/amount in canals, pendulous ears, humidity, and water
exposure increase the chance of secondary infectious otitis externa

## Malassezia Yeast

## Malassezia pachydermatis

Opportunistic pathogen that is part of the normal flora

Bowling pin, snowman, peanut shaped when budding

Stain deep blue/purple with Diff Quick®

#### Treatment:

Terbinafine

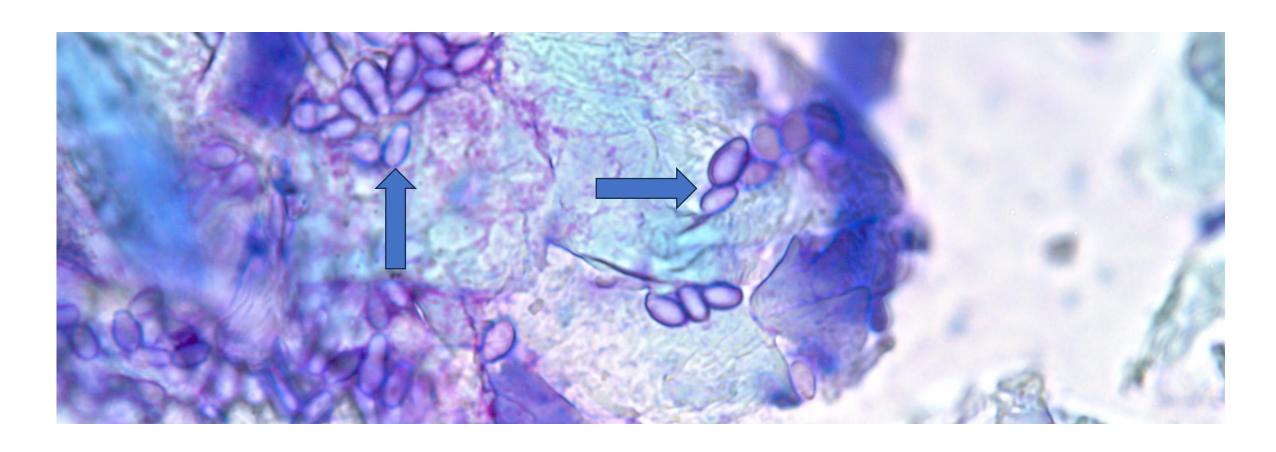
Azoles (Posaconazole, thiobendazole, miconazole, clotrimazole)

Corticosteroid Ototopical



# Boo....it's a ghost (yeast)

Malassezia pachydermatis



## **Cocci Bacteria**

Staphylococcus pseudintermedius

Opportunistic pathogen that is part of the normal flora

Deeply blue/purple perfect circles when stained with Diff Quick®

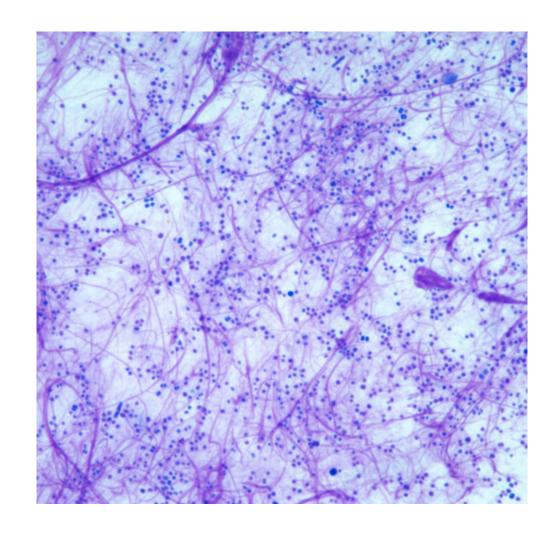
#### Treatment:

Aminoglycoside (gentamicin, neomycin)

Florfenicol

+/-Fluroquinolone (enrofloxacin)

Corticosteroid Ototopical



# Rod Shaped bacteria

## Pseudomonas aeruginosa

- -Pseudomonas and other rod shaped bacteria are never 'normal' on otic cytology
- Deeply blue/purple rods
- -Pseudomonas requires inflammation + predisposing factor(increased humidity, increased pH) to overgrow
- -Rod shaped otitis is more common in ears that have chronic changes from previous inflammatory and infectious (cocci, yeast) insults.

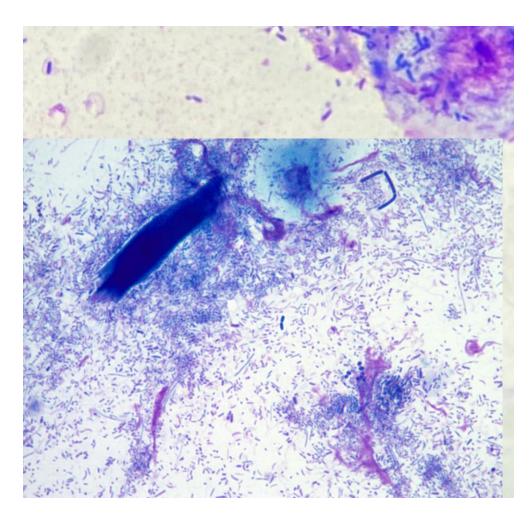
#### **Treatment:**

Polymixin B

Flouroquinolone (enrofloxacin)

Silver Sulfadiazine

Aminoglycoside (Amikacin)



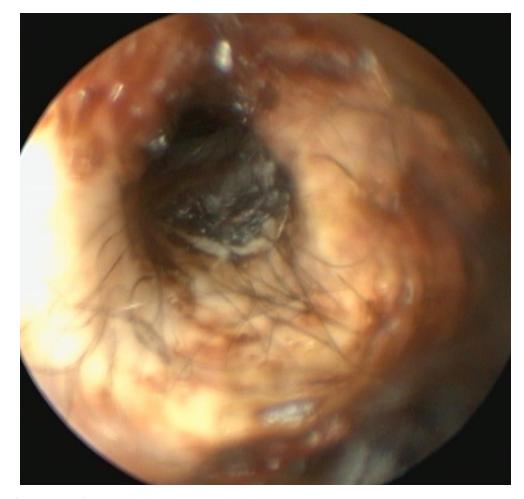
# Case Presentation -2 year old GSD Mix

#### First Time Offender

HX: Went swimming in the pond twice last weekend and is now shaking head.

No prior ear infection, mild pedal pruritus in the spring and summer, monthly isoxazoline flea/tick control. Presumed healthy animal.

PE: No skin erythema, no ectoparasites, BCS 5/9 Both ear canals similarly affected. TM are intact



Minimal erythema, moderate dark brown waxy debris

# Case Presentation- 2 year old GSD Mix

#### First Time Offender

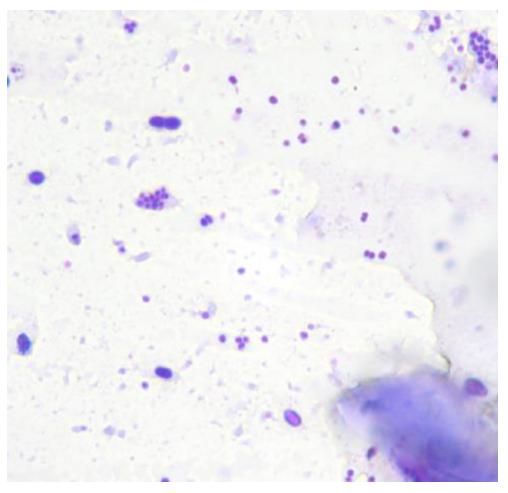
Cytology results have evidence cocci bacteria and *Malassezia* overgrowth

**Primary Problem**: Allergic skin and ear disease (minimal to mild)

**Predisposing factor**: Wet ear canals from swimming

**Secondary factor**: cocci bacteria and Malassezia overgrowth

**Treatment**: Corticosteroid ototopical and cleanser OR leave in terbinafine + antibacterial ototopical OR azole ototopical + antibacterial ototopical +/- cleanser



Moderate cocci and fewer yeast

## First Time Offender

#### Recheck 2 weeks later

HX: Applied medications as directed, head shaking has resolved

PE: No skin erythema, no ectoparasites, BCS 5/9

Ear canals have minimal erythema, but otherwise normal on otoscopic examination

Treatment: monitor for new clinical signs, cerumenolytic and antimicrobial cleanser after swimming



Few anucleate keratinocytes

# Case Presentation- 2 year old GSD Mix

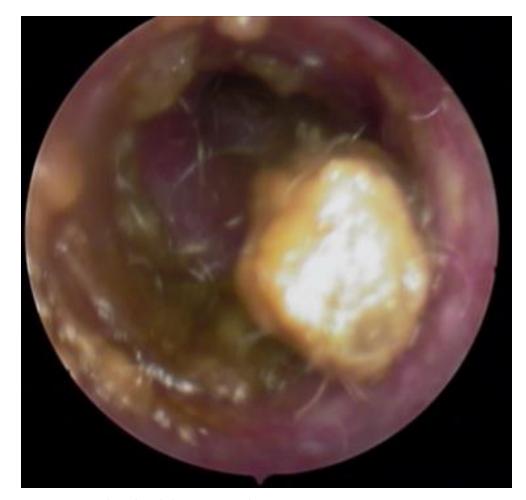
## Repeat Offender

HX: Went swimming in the pond twice last weekend and is now shaking head.

4 prior ear infections, moderate spring-fall pruritus of paws and trunk, monthly isoxazoline flea/tick control.

PE: Salivary staining on paws, excoriations on the axilla with occasional papules on axilla and inguinal region, no ectoparasites, BCS 5/9

Both ear canals similarly affected. TM are intact



Erythema, epithelial hyperplasia, ceruminous debris, ceruminolith

# **Case Presentation – 2 year old GSD Mix**

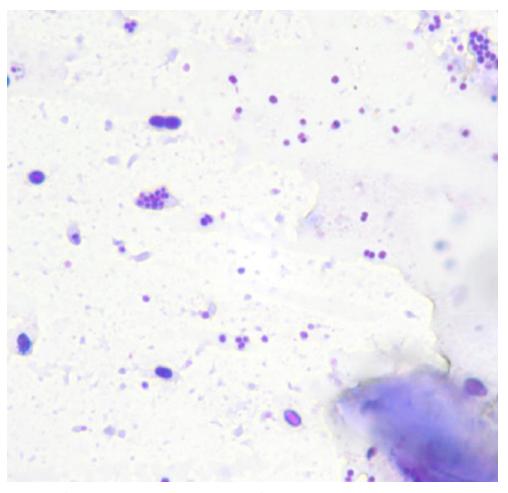
## Repeat Offender

Cytology results have evidence cocci bacteria and *Malassezia* overgrowth

**Primary Problem**: Allergic skin and ear disease (moderate, relapsing)

Predisposing factor: Wet ear canals from swimming

**Secondary factor**: cocci bacteria and Malassezia overgrowth



Moderate cocci and fewer yeast

# Case Presentation- 2 year old GSD Mix

#### **Treatment**

#### **Topical Plan**

Corticosteroid ototopical and cerumenolytic cleanser

OR

azole antifungal ototopical + topical antibiotic and ceruminolytic cleanser

OR

terbinafine ototopical + topical antibiotic and ceruminolytic cleanser

#### Systemic Plan

Oral corticosteroid 0.5-1mg/kg per day until resolution of inflammatory changes, then taper

\*\*\* modified cyclosporine OR oclacitinib would only be chosen if there was a significant contraindication for a corticosteroid.

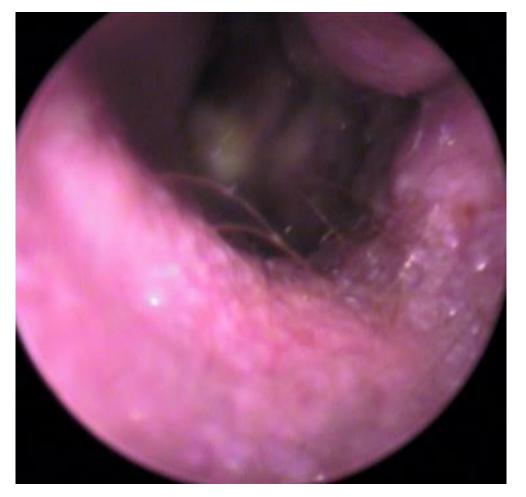
# Repeat Offender- 2 year old GSD Mix

#### Recheck 2 weeks later

HX: Owner administered medications as directed, head shaking is improved but not resolved

PE: Reduced salivary staining on paws, papules and excoriations resolved.

Ear examination shows minimal debris with erythematous slightly irregular ear canals and focal stenosis



Erythema, epithelial and glandular hyperplasia, mild stenosis

# Case Presentation -2 year old GSD Mix

#### Maintenance Plan

#### Topical Plan

Ototopical steroid daily until inflammation resolves +/- a few times per week longer term

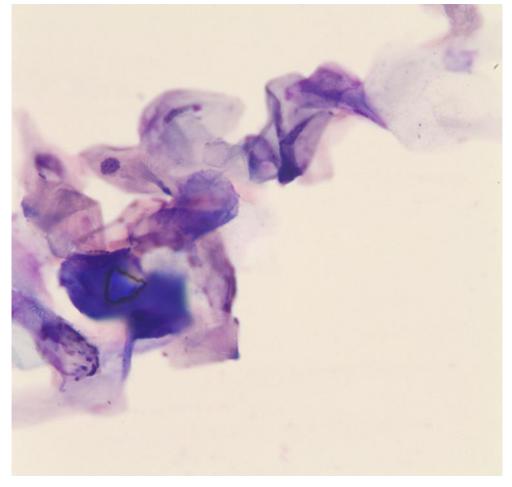
Ceruminolytic and antimicrobial flush 1-2 times per week and after swimming

#### Systemic Plan

Modified cyclosporine

Oclacitinib

Allergy specific immunotherapy (+ CsA or oclacitinib)



Variably stained keratinocytes

# Ear Flushing and Otitis

#### Ceruminous

Oil based cleansers

Alcohol based cleansers

#### **Purulent**

Water based cleanser
Detergent based cleanser

#### **Antimicrobial**

Low pH products including acetic acid based

Isopropyl alcohol

Chlorhexidine

Hypochlorous acid

## Adjunctive Antimicrobial

Triz- EDTA

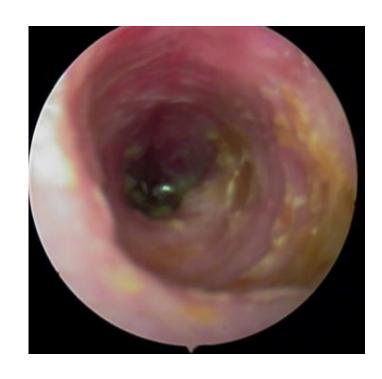
Monosaccharides, polysacaccharides

## Now let's make it even more complicated

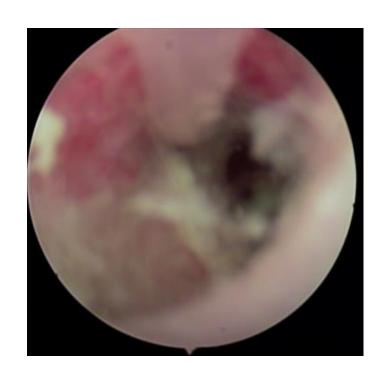
## Perpetuating factors- Part 3

- Ulcerative ear canals (edema, lack of normal glandular function)
- Calcification
- Nodular hyperplasia
- Worsening Stenosis
- Otitis media +/- osteomyelitis

# Perpetuating factor: Ulcerated ear canals



Ceruminous/seborrheic to purulent discharge



Purulent discharge



Thick and slimy purulent discharge, probable biofilm

# **Perpetuating Factors**



Otitis Media, ulcerations of canal



Nodular glandular and epithelial hyperplasia, moderate stenosis



Heavy debris admixed with ear packing material, ceruminolith

# **Inflammatory Cells**

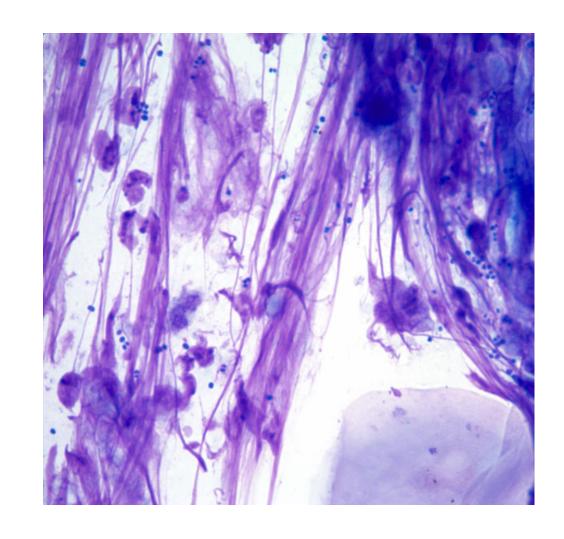
## Neutrophils

Inflammatory cells are not present in acute otitis

Chronic otitis externa, especially with secondary bacterial infection will have neutrophils and occasionally macrophages

Neutrophils and neutrophilic streaming may be an indicator of otitis media

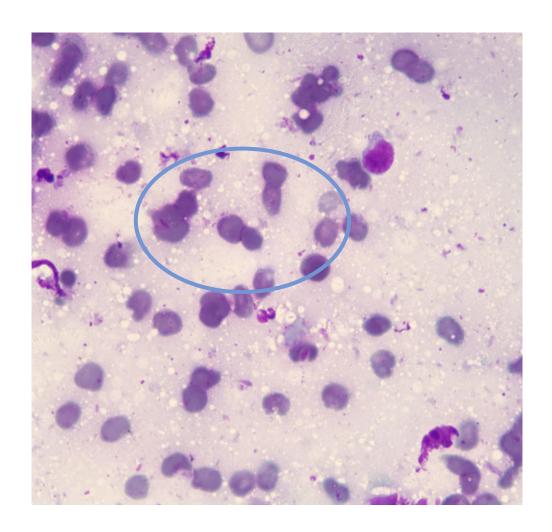
Neutrophils and macrophages may be present in cases of polyp/tumor in the ear canal



## **Red Blood Cells**

RBC's may be present in ulcerated ear canals

RBC's may be present in cases of polyp/masses



## Referral cases- Older Feline and ear mass

#### Recurrent otitis externa



Mass covered by ceruminous debris



Erythematous mass surrounded by keratin debris

Bleeding otic mass

## Referral cases- Older Feline and ear mass

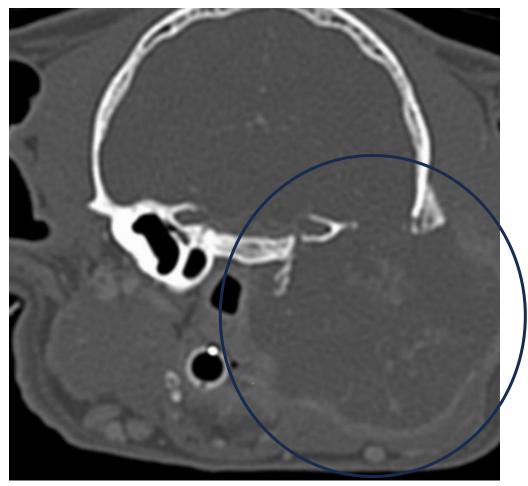
#### Malignant Neoplasia

Early referral and CT imaging is recommended if there is concern for a mass

Malignant tumor types include: ceruminous adenocarcinoma and squamous cell carcinoma

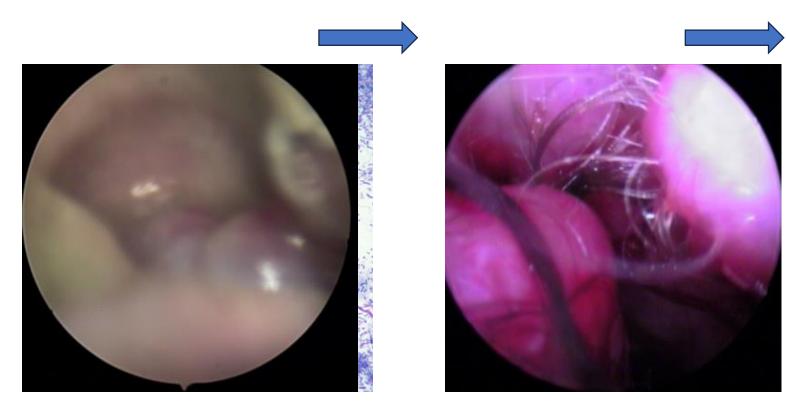
Without gross extension beyond the canal, total ear canal ablation +/- radiation therapy

With extension outside the ear canal, palliative care +/- radiation therapy are remaining options



Expansive and lytic malignant mass

# Chronic hyperplastic otitis – cocker spaniel



Slimy, purulent exudate with stenosis and nodular hyperplasia

Mixed rod otitis and nuclear streaming

Nodular hyperplasia and stenosis after treatment of infection

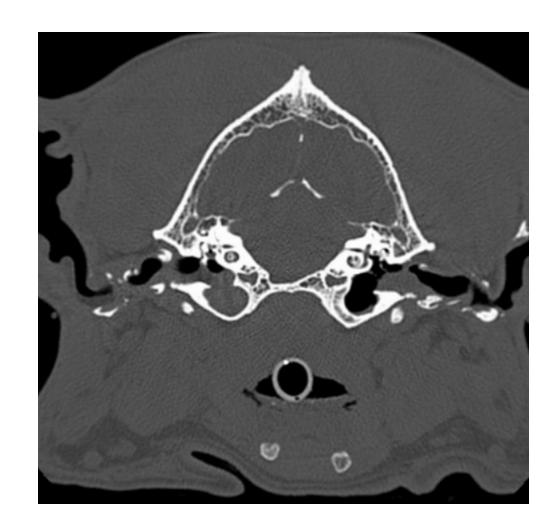
# **Chronic Hyperplastic Otitis- Cocker Spaniel**

### End Stage Ear Disease

Calcified Ear canals and nodular hyperplasia

Medical management is unlikely to be successful

Total ear canal ablation and bulla osteotomy (TECA-BO) is recommended



# **Otitis Media**



Large rupture of tympanic membrane, purulent otitis externa



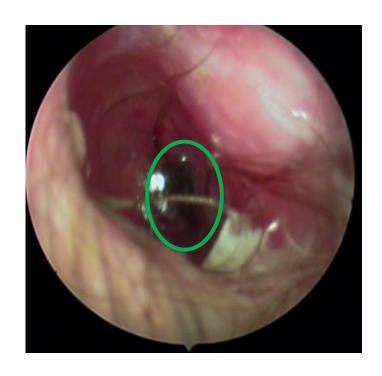
Otitis media, focal calcification of external ear canal



Small rupture of tympanic membrane, purulent otitis externa

# Otitis media- Brachycephalic dogs

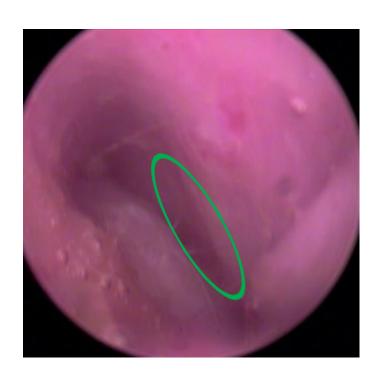
#### French Bulldog



Stenotic horizontal ear canal and purulent otitis externa



Marked stenosis of horizontal canal



Marked stenosis of the horizontal canal

# **New Technology**

#### IDEXX inVue Dx™ Cellular Analyzer

Sample Collection

Cotton tip applicator (CTA) placed into the ear canal until you reach the end of the vertical canal

Spin the CTA along the long axis gently, then remove

The sample is placed in the reagent and agitated for 10-15 seconds

The reagent mixed with sample are placed onto the cartridge and the cartridge is inserted into the analyzer

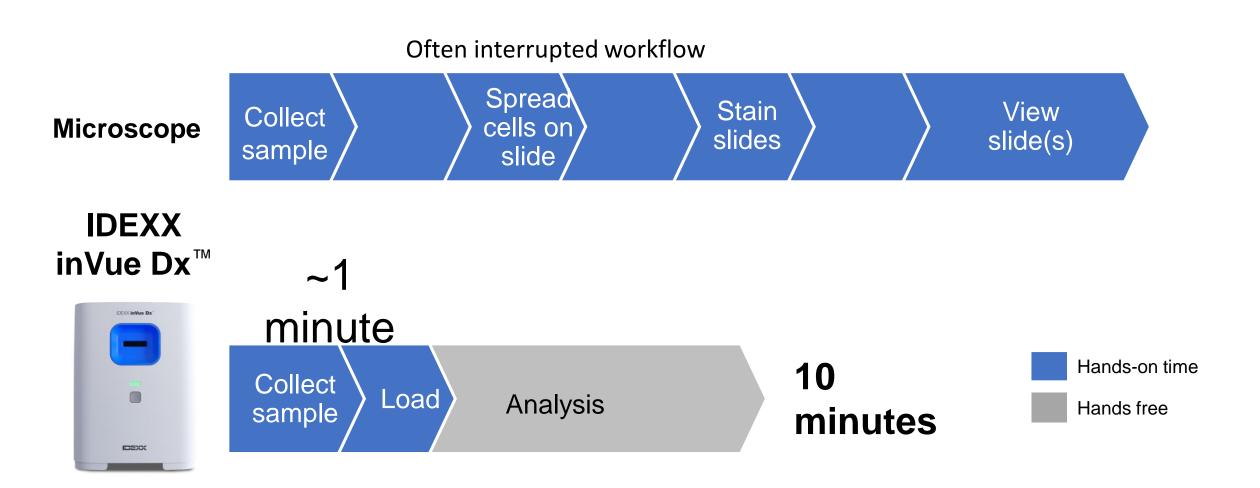


Put sample in the reagent.

Drop sample into cartridge.

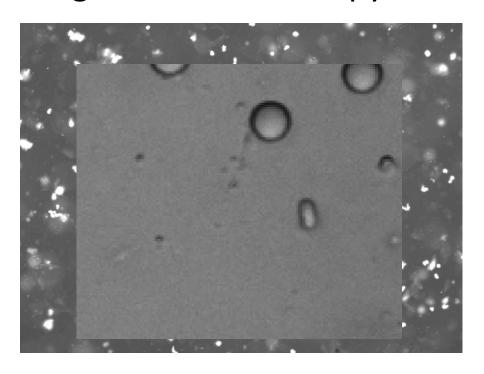
Insert and press
the Start button

# IDEXX inVue Dx<sup>™</sup> Cellular Analyzer's novel slide-free workflow gives time back to practices

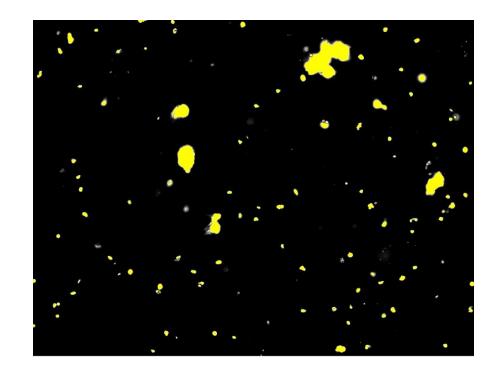


# But how does it work???

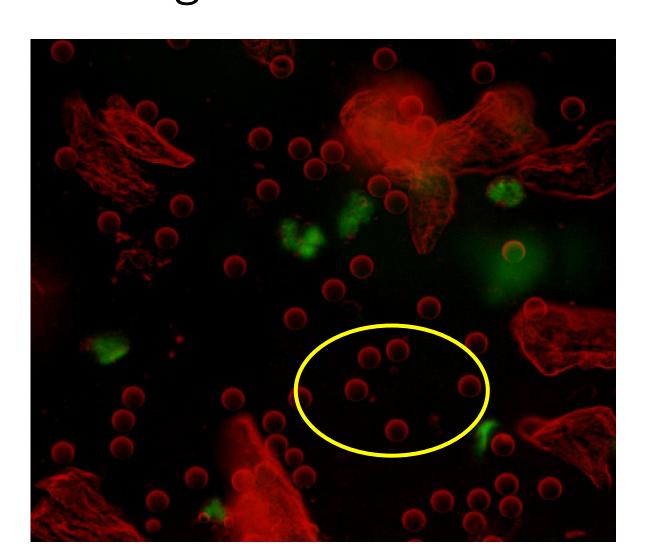
#### **Bright Field Microscopy**



#### Fluorescent Microscopy

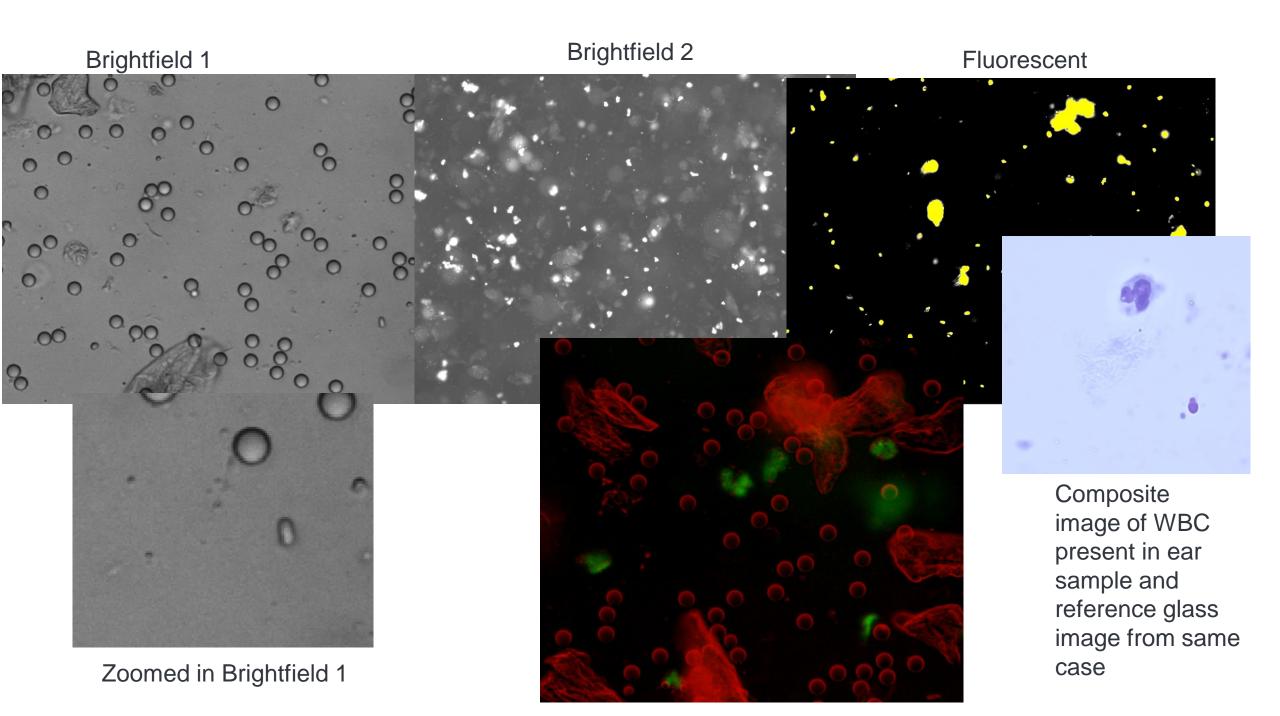


# Blending of these images leads to a final image

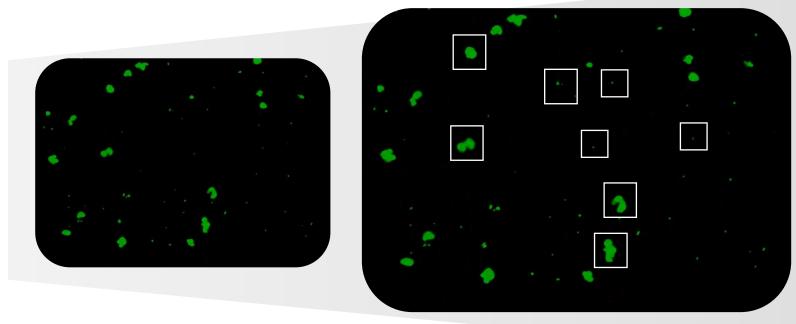


The yellow circle is showing the focus beads

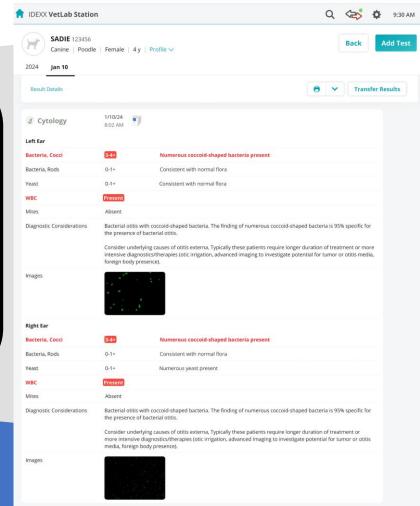
Focus beads tell the analyzer how 'deep' it is looking in the sample and gives the analyzer the ability to look at multiple planes.



IDEXX inVue Dx<sup>™</sup> analyzer: automated quantification, classification, and interpretation of ear cytology



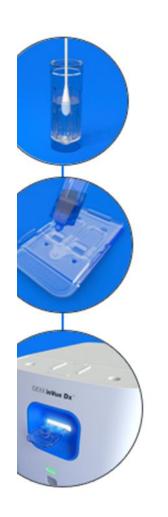
Deep-learning models deliver reference labquality results in a real-time environment



## **Future Directions**

Slide microscopy and IDEXX inVue DX™ require the clinician to correlate the inflammatory cells or infection with the patient's exam findings.

This technology may be especially useful to large mobile units that already have blood analyzers, shelter situations, ER practices, and high-volume private practice



Put sample in the reagent.

Drop sample into cartridge.

Insert and press

Perform ear cytology on all cases of otitis

Encourage clients to make recheck appointments

Work through the primary, secondary, and perpetuating factors in repeat offenders

Don't hesitate to refer chronic otitis cases to a Dermatologist

Key Points



## References

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