PEE IS THE KEY

HOW TO UNLOCK YOUR CASES WITH PERFECT-PRACTICE URINALYSIS

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GROSS APPEARANCE

PARAMETER	APPEARANCE	POSSIBLE CAUSES
COLOR	Orange	Bilirubinuria
	Red	Erythrocytes, free haemoglobin, myoglobin (beetroot) o Haematuria: supernatant clear and red pellet blood
	Brown / Black	Methemoglobinuria, some drugs
	Green	Methylene blue, Dithiazanine, Methocarbamol, Urate crystalluria, Indigo blue, Evan's blue, Bilirubin, Biliverdin, Riboflavin, Thymol, Phenol, Triamterene, Amitriptyline, Anthraquinone, Green food dye
CLARITY	Cloudy	High concentration, WBC, RBC, Crystals, bacteria, mucus, lipid or contaminants (foreign debris)
	Clear	-

DIPSTICK

How useful is the dipstick?

- Very useful when read at the correct time interval
- Very sensitive to **moisture, heat** and **light** so keep container closed when not in use
- Cold urine may inhibit chemical reactions on the dipstick- so warm it up if refrigerated

GLUCOSURIA

- Stress related, dextrose administration
- Diabetes Mellitus (hyperglycaemia)
- Proximal renal tubular dysfunction (acute tubular necrosis, pyelonephritis, Fanconi's syndrome, primary renal glucosuria, certain treats)
- In presence of azotaemia can indicates acute renal injury (leptospirosis, toxin)

False Negative	False Positive
Cold urine, Ascorbic Acid, Formaldehyde	Contamination with hydrogen peroxide, chlorine, or bleach

PROTEINURIA: Interpret in relation to the USG

- Transient in acute disease
- Correlate with collection technique
- UPC ratio: impacted by inflammation as measures both Alb + Glob
- More sensitive to albumin and less to globulins/Bence Jones proteins (protein fractions)

False Negative	False Positive
-	Blood, Pyuria, Bacteriuria, Alkaline pH, Use of disinfectants

KETONES: More sensitive to acetoacetic acid

- Diabetes Mellitus
- Prolonged starvation (excessive lipid catabolism)
- High protein, low carb diet
- Does not detect BOH

False Negative	False Positive
Pad on dipstick exposed to light, heat, humidity, very acidic urine	Large amounts of bilirubin (discolored urine)

What can you trust and what can you discount on dipstick?

NITRITES	LEUCOCYTES
Unreliable ○ While some bacteria are able to convert nitrate to nitrite, it is **not a reliable marker of bacteriuria** in animals	Unreliable (people still look at them anyway) ○ Detects **esterases** found in the granules of neutrophils, basophils and eosinophils ○ **Useless in cats** due to high false positive range ○ **Sensitive but not specific in dogs** (A positive test is positive but a negative test is inconclusive)

What affects the results?

ACIDURIA	Gram-negative bacteria Ingestion of meat Respiratory and metabolic acidosis Depletion of chloride due to severe vomiting Severe diarrhoea Starvation Pyrexia Urinary acidifiers
ALKALINE URINE	Gram-positive bacteria often associated with alkaline urine (urease production which metabolizes urea to ammonia), *Proteus, Staphylococcus* Recent meal Administration of bicarb/citrate Renal tubular acidosis Diet with elevated veggies/cereals Metabolic and respiratory alkalosis

RBCs

- Small number of intact RBCs will cause **spotting** of the test pad
- Haemoglobin, myoglobin, or large number of intact RBCs will cause a **homogenous color change**
  - About 0.5 mL blood per liter of urine (approx. 2500 RBC/mL) is necessary for visual detection of hematuria
  - RBCs will lyse in dilute (USG 1.008) or alkaline urine

FALSE NEGATIVE	FALSE POSITIVE
Due to RBCs that settled on the bottom of the sample leading to a disparity between the dipstick and sediment evaluation	Due to contamination with oxidizing agents in disinfectants

- Haemoglobin vs. haematuria: **centrifuge** the sample
  - Dipstick cannot differentiate between haemoglobin and myoglobin
  - Icteric or discoloured plasma suggests haemoglobinuria as myoglobin does not cause a colour change in plasma.

HAEMOGLOBINURIA / MYOGLOBINURIA	HAEMATURIA
Red or brown supernatant	Clear supernatant

USG: On centrifuged urine (supernatant)

- If specific gravity is off the scale, **dilute 1:1 the urine with distilled water** and read, **doubling** the last 2 decimal places (*e.g.* 1.024 = 1.048)

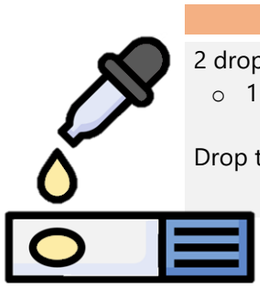
DIFFERENT CAUSES OF USG CHANGE	
Glucosuria	Solutes affect the reading
Very high protein levels	Solutes affect the reading
Sepsis	Endotoxaemia (pyothorax, pyometra) will block the action of ADH and result in lower than expected USG
Hyperadrenocorticism	Due to lack of aldosterone
Cold urine	Result in false USG increase
Other conditions	Liver disease, glucocorticoids, hypercalcaemia, hypokalaemia, hypertension, polycythemia

- **Gram-negative infected specimens** are reported to have a **lower USG** than Gram-positive infected specimens and culture-negative specimens
  - USG is higher in specimens infected with *S. felis* and lower in specimens infected with *E. coli* than in those infected with other uropathogens

SEDIMENT CYTOLOGY

Tips for looking at urine to look for bacteria (techniques, smear techniques etc.)

- Dipstick testing on un-spun urine
- Centrifuge after dipstick
  - USG on the supernatant
  - Slides: decant or siphon off most of the supernatant



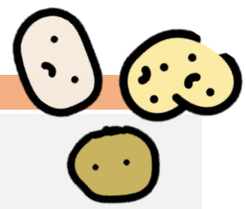
WET MOUNT	DRY MOUNT
2 drops of sediment, coverslipped - 1 plain and 1 stained with methylene blue Drop the condenser down on the microscope	Make 2 air-dried smears of urine sediment at the time of collection - 1 using Diff-Quick - 1 using Gram Stain or no stain to send to the commercial lab

Bacteria vs. Gunk: How do we differentiate under the microscope?

- Check ASAP for crystals as these may **form and dissolve** in storage and transit
- Check for crystals, leucocytes, bacteria, red blood cells, casts

CAUSES OF ERROR OR FALSE POSITIVE ON WET MOUNT MICROSCOPIC EXAMINATION	
Lipid droplets Debris Stain artifact	Stain contamination Brownian motion Inexperience looking at sediment

Do animals with stones always have crystals? The answer is no.



TYPE OF STONES	POSSIBLE CAUSES
Struvite (Magnesium Ammonium Phosphate)	Infection in dogs (urease-forming bacteria) Sterile in cats
Calcium Oxalate Dihydrate	Neutral to acidic urine Veggies high in oxalates
Calcium Carbonate	Normal in horses, rabbits Sustained hypercalcaemia Abnormal renal handling of filtered calcium
Cysteine	Seen in acidic urine. Presence of cystine crystals represents a proximal tubular defect in amino acid reabsorption. Dachshunds, Newfoundlands, English Bulldogs, and Scottish Terriers have a high incidence of cystine urolithiasis
Ammonium Biurate	Normal in Dalmatians (processing of purines) Portosystemic shunt Never normal in cats
Sulfa crystals	-

What about casts?
Few, but not as important as many.

TYPE OF CAST	POSSIBLE CAUSES
Hyaline	Benign - dehydrated animals
Granular / Waxy	Plasma proteins/albumin and associated with renal disease
White Cell	Pyelonephritis / renal inflammation
Epithelial	Tubular injury (leptospirosis)

Are there any other 'surprises' you'll find on urine that will point you in a new direction?

1. **Eosinophils**: eosinophilic cystitis
   - Horses & Dogs: fibroblastic proliferation within hyperplastic urothelial cells of the bladder with foci of eosinophils and accompanying ulceration and haemorrhage
2. **Renal parasitism**: *Halicephalobus gingivalis* (aberrant migration)
3. **Epithelial cells**: dysplasia vs. neoplasia
   - Submit an aliquot of **urine or urine sediment in EDTA** for cytospin or direct smear cytology at commercial lab
4. **Neoplasia**
5. **Yeast, Fungi**
6. **Sperm**

URINE CULTURE

QUANTIFIED URINE CULTURE	QUALITATIVE URINE CULTURE
Identify bacteria, number of bacteria, and determine sensitivity	Isolation of something, anything via any method

Voided vs. Catheterization vs. Cystocentesis
Is there any point of sending free flow for culture?

- **Clean cystocentesis: gold-standard**
- Voided: make sure to clip and clean external genitalia, if necessary

To fridge or not fridge for culture?

- Cold urine: alter USG (false increase), promotes crystal formation, may inhibit chemical reactions, inhibits bacterial overgrowth

How long can it sit for before going to the lab, especially for culture and for crystal analysis?

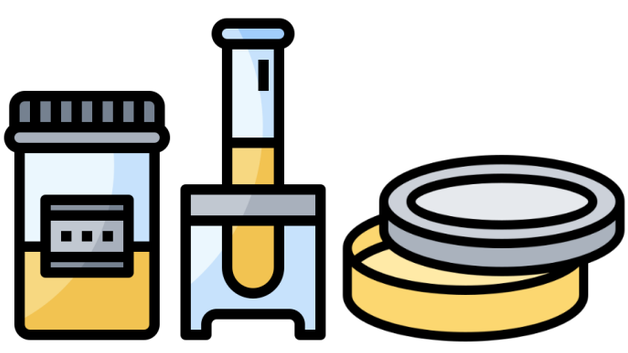
- Processing quickly is ideal, at least **within 6 hours**
- At room temperature bacterial counts can double **every 30 minutes**
- Multiplication or destruction of bacteria can occur **within 1 hour** of collection

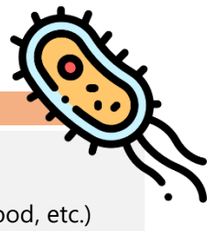
What will affect the ability of bugs to grow on culture?

- Patients currently on antibiotics can inhibit growth without killing organisms
- **False negative results**: iatrogenic bactericidal events (storage, handling, transport)

If it's on antibiotics, could it still grow anything?

- Negative bacterial culture result during antibiotic administration indicates successful eradication of infection
- Negative growth with persistence of clinical signs suggests **another underlying cause** for the clinical signs





POSITIVE CULTURE RESULTS: INTERPRETATION & RECOMMENDATIONS	
Same organism, susceptible to current antibiotics	Antibiotic not reaching the site of infection ○ Owner compliance ○ Impairment of intestinal absorption (given with food, etc.) ○ Biofilms ○ Nidus of infection (abscess, necrosis, uroliths)
Same organism but different susceptibility pattern	Emerging antibiotic resistance Change based on susceptibility results
Different organism	Original bug + antibiotic therapy effective but allowed colonization with another more resistant organism Can occur with indwelling catheters, surgical procedures which allow exposure to the environment Sort out the underlying problem

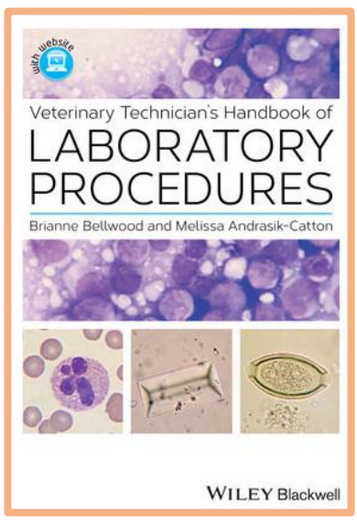
What's the sensitivity – *i.e.* does negative mean negative, for example, pyelonephritis?

Because concentration of antimicrobials (except nitrofurantoin) in paper disks is comparable to typical serum concentration of the drug, **drugs that are found to be resistant by the agar disk diffusion method** may be **effective in the urinary tract** if they are excreted in **high concentrations** in urine (*e.g.* ampicillin, cephalexin).

MIC is defined as the lowest antimicrobial concentration (or highest dilution) that allows no visible bacterial growth.

- The MIC is **several dilutions lower** than the minimum bactericidal concentration of drugs.
- In general, antimicrobial agents are likely to be effective if they can achieve a urine concentration that is **4 times the MIC**.
- Many antimicrobial drugs that are renally excreted reach concentrations in urine that are **10 to 100 times greater** than serum concentrations.

SUGGESTED REFERENCES



KRISTEN'S BOOK RECOMMENDATION

Bellwood, B. and Andrasik-Catton, M. (2013). *Veterinary Technician's Handbook of Laboratory Procedures*. John Wiley & Sons.

https://bit.ly/patholgybook

ONLINE MICROSCOPY IMAGES

https://eclinpath.com/urinalysis/crystals/