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LIQUID GOLD PART 2 - 'WHY AND HOW TO FILTER THE GOLDEN INFORMATION TO MAXIMIZE THE VALUE OF THE MICROSCOPIC EXAMINATION OF URINE'

Introduction

Complete urinalysis should be performed every time a need to evaluate the renal system exists. However, the complete urinalysis is not only of value for evaluation of the renal system. The urinalysis can provide information about other body organ systems like the liver as well as help characterize underlying systemic acid-base abnormalities, the severity of diabetes and diabetic ketoacidosis as well as assisting in characterizing various hematologic abnormalities like hemolytic disease.

Routine urinalysis can easily and accurately be performed in a veterinary practice. There are different components of a complete urinalysis. The physical examination of the urine collected includes crude but important gross inspection of the urine. The chemical examination primarily consists of multiple tests performed on the classic dry reagent strip testing commonly known as the "Dip Stick" test. The microscopic examination consists of evaluation of the formed elements of a urine specimen and is something that should be performed in a short time following the collection of the urine specimen.

Urinalysis - microscopic examanation

Microscopic examination of urine sediments requires the availability of a good quality microscope. If one is not available accurate assessment of the formed elements of the urine will not be possible. Both a good 10x and 40x objective is required on the microscope. When examining the urine sediment microscopically, most people use a bright field microscope that is used for routine hematology and cytology specimens. When examining the urine specimen, putting the light out of bright field microscopy settings by lowering the substage condenser and partially closing the iris on the microscope may prove helpful in making various formed elements easier to visualize. The preparation of the sediment requires only a simple tabletop centrifuge with a relatively slow speed to allow sedimentation of the formed elements without mechanical damage to these elements. Most people examine the sediment on unstained preparations, but there are a series of commonly used stains including some developed specifically for urine sediment examination (Sedi Stain) or routine Romanovsky stains commonly used in hematology or cytology specimen analysis (Diff Quik, Wright's stain, etc.). The initial examination of an unstained specimen will be difficult. However, learning how to read these specimens precludes the observation of stain precipitate and mis-identifying this as an organism or crystal and prevents the bacterial or fungal contamination of the stains with subsequent overgrowth and mis-interpretation of the presence of these organisms. Practice with some pseudo urine specimens by using a clean urine specimen spiked with various "formed elements" commonly seen in urine sediment specimens. Epithelial cells from the buccal mucosa (spatula gentle scraping of the oral cavity), leukocytes and erythrocytes from anticoagulated whole blood originally submitted for hematology evaluation, and potential neoplastic or various inflammatory cell populations from other specimens (fine needle aspirates of solid masses, sediment smears of effusions, etc.). Some of the more commonly encountered formed elements in urine are briefly discussed below.

Cells

Erythrocytes: Red blood cells in the urine, hematuria, can originate from any region of the urinary tract but most commonly is associated with local bleeding in the urinary bladder associated with cystitis or it is iatrogenic during the collection process. In fresh urine, the cells are non-nucleated and close examination reveals their biconcave shape. In dilute urine, erythrocytes swell and lyse resulting in hemoglobinuria rather than hematuria. In concentrated urine, erythrocytes typically crenate and resemble crenated erythrocytes in the peripheral blood with multiple short uniform sharp cytoplasmic projections. Practice in the identification of these cells in urine is relatively simple. Place a drop of EDTA anticoagulated blood into dilute, non-concentrated and concentrated urine samples and see the effect on the erythrocytes.

Leukocytes: White blood cells are commonly seen with cystitis or any other type of inflammatory disease in the urinary tract. Typically only very few leukocytes are seen in

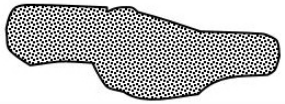
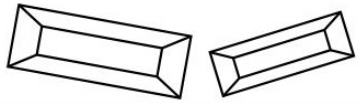
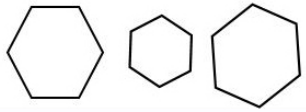
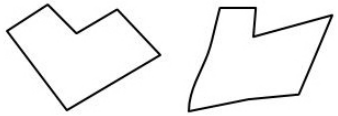
normal urine (less than 2 WBC / HPF). These cells are usually spherical and in contrast to the erythrocytes, nuclei are visualized with careful inspection at 40x objective magnification field of view. Neutrophils are the most common leukocyte seen in urine and the lobed nucleus is usually very easy to identify even in unstained preparations. Mononuclear leukocytes, monocytes, macrophages and lymphocytes, have rounded to slightly indented nuclei and are slightly larger than the neutrophil. If there is ever a question regarding the identification of leukocytes or recognizing what type of leukocyte is present, preparation of a concentrated sediment air-dried specimen of the urine sediment and staining with a stain you use for hematology is recommended. This will allow direct detailed visualization of nuclear and cytoplasmic morphology similar to evaluation of a blood film.

Epithelial cells: Differentiation between renal tubular, transitional and squamous epithelial cells from different regions of the urinary tract may take some practice. These cells are generally larger than the leukocytes and they typically have more cytoplasm (lower nuclear/cytoplasmic ratios) than leukocytes. Squamous epithelial cells are commonly larger than transitional epithelial cells (2-4 times larger than leukocytes) and renal tubular epithelial cells are generally only slightly larger than leukocytes. Transitional epithelial cells may take on a polygonal or irregular shape with variably prominent projections from the cytoplasm. Close inspection even in an unstained preparation should allow visualization of the relatively centrally located round to oval nucleus commonly seen in these epithelial cells. They may be present individually or in small cohesive clusters. Collection of relatively large numbers of transitional epithelial cells with catheterized specimens is common due to the mechanical sloughing during the placement of the catheter.

Crystals: Most crystals found commonly in the urine of animals are not clinically significant. Fresh, un-refridgerated urine samples generally do not have crystals or have only very few. If a urine sample is allowed to stand for any period of time, crystals will form if the correct compounds are present, particularly in a supersaturated specimen. Most crystals have typical morphologic appearances that allow identification. Knowledge of the dependency of pH on the formation of crystals may prove helpful also. Certain crystals will form only under certain pH situations (see table below).

Crystal	Acid Urine	Alkaline Urine
Amorphous urates	+	-
Bilirubin	+	-
Calcium oxalates	+	+/-
Cholesterol	+	-
Cystine	+	-
Hippuric acid	+	+/-
Leucine	+	-
Sodium urate	+	-
Sulfonamides	+	-
Tyrosine	+	-
Uric acid	+	-
Ammonium biurates	+/-	+
Amorphous phosphates	-	+
Calcium carbonate	-	+
Calcium phosphates	-	+
Triple phosphates	-	+

DIAGNOSTIC TESTING

Few Common Urine Crystals	
Calcium oxalates (dihydrate)	
Calcium oxalates (monohydrate)	
Amorphous phosphates	
Triple phosphates	
Cystine	
Cholesterol	
Bilirubin	

Casts: Casts are formed in the tubules of the kidney as a result of clumping of cells or other material within a protein matrix. These formed elements of the urine sediment prove to be helpful indicators of renal disease. Except for protein casts, renal tubular casts indicate tubular damage. Accurate identification of the various casts may be difficult because there is actually a transition between the different types of casts. In addition, they may have different formed elements including cells, granular proteinaceous cellular debris, protein, and fat. Casts are identified by several common features. These cylindrical structures have parallel sides and typically one end of the cast is irregularly ragged and the other end is potentially smooth and blunted. The ragged end represents the end of the cast that has broken off any remaining cast material within the renal tubules. The smooth end represents the end of the renal tubular thrombus, which represents the cast that eventually is sloughed and potentially identified in the urine sediment.

Miscellaneous components of the urine sediment: A variety of components may be found in the urine of normal and abnormal animals. Bacteria identified as uniform small round (coccal) to short rod-shaped structures that elicit "Brownian Motion" because of their small sample size. If there is ever a question related to the accurate identification of bacteria, an air-dried sediment of the urine stained with the stain commonly used in hematology can be helpful in characterizing these small particles. Yeasts and other structures including contaminating fungal forms from the environment may be found in urine also. Sperm may be seen and are typically easy to recognize based upon the distinctive structure and the fact that many will be viable and motile in a fresh urine specimen. Structures such as filter / fiber fragments and mucus strands may prove interfering with the accurate identification of casts. Glove powder must not be confused with crystals that are truly formed in the urine.