

More Than Just a Hematocrit Gaining Information from the Hematocrit Tube

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Laboratory testing in the emergency and critical care veterinary setting has several purposes.

- To confirm, establish or rule out a diagnosis,
- To monitor a patient's response to therapy (management guide)
- To establish prognosis
- To screen for or detect disease.

As an important contributing member of the veterinary team, the nurse/technician should become familiar with normal values for each laboratory test, recognize what clinical signs can be seen with abnormal results and know when to rule-out human or mechanical error (e.g. uncalibrated equipment, mislabeled or mishandled sample).

The intensive care unit (ICU) should utilize instrumentation that performs most of its analysis on heparinized blood. This minimizes the turnaround time of results. (NOTE: Tests that are performed on serum require that the sample sit for at least 1 hour prior to spinning down). Laboratory equipment should be purchased based not only on cost effectiveness but also on ease of use, minimum volume of blood required per test, turn around time for results and dependability.

Stat laboratory testing performed in the emergency and critical care setting should include: packed cell volume (PCV), total protein (TP), blood glucose utilizing a glucometer, blood urea nitrogen (BUN) reagent strip, blood electrolytes (sodium, potassium, chloride and ionized calcium), blood gas analysis, urine specific gravity, urine dipstick, platelet count or estimate, activated clotting time (ACT), buccal mucosal bleeding time, white blood cell count (WBC) or estimation, plasma osmolality and blood ammonia. Other tests that can be made available are complete blood cell count (CBC) with differential, certain chemistries (i.e. creatinine, albumin), urine sediment, ethylene glycol test and cytology of abdominal and thoracic fluid.

The Hematocrit Tube

Blood should be obtained from a clean stick to avoid red cells hemolysis. Several hematocrit tubes should be filled, with one or more allowed to sit for at least 10 minutes. The tube should then be examined to see if there is evidence of agglutination of red cells, seen as red cell clumping, most typically in red colored plasma. The tube is then spun down and each layer examined.

Red Cell Layer - Packed Cell Volume

The PCV is used to determine red blood cell mass. After centrifugation, red blood cell (RBC) sedimentation in the hematocrit tubes will occur in order of cell age, with the oldest cells occupying the lower end of the tube closest to the clayed end and the youngest ones towards the top. This is useful when assessing for blood parasites, which are found in the layer of older more fragile red cells. Parasites that can be detected in the RBCs include Hemobartenella, Cytauxzoon (cats) and Babesia (dogs).

Causes for decreased PCV in the ICU include: acute/chronic blood loss, immune mediated disease, overzealous fluid therapy, and obtaining the sample from an area with a hematoma.

Causes for increased PCV include: the early stage shock in the dog (due to splenic contraction),



polycythemia and dehydration.

Buffy coat

The thin white layer in the centrifuged hematocrit tube, lying between the red cell layer and the plasma or serum, is the buffy coat. The white blood cells and platelets are in this space. A thicker buffy coats implies that there are either more white cells and platelets than normal, or that there are more immature white cells, which are most often larger than older white cells. A slide can be made from this layer to stain and examine with the microscope to detect white cell morphology and any white cell inclusions or abnormalities.

Plasma or serum layer - Total Protein

After spinning the hematocrit tube, an assessment is made of the coloration of the serum or plasma. Normal plasma and/or serum should be “straw colored”, or very pale yellow in coloration, and clear. Darker yellow coloration is called icterus, and implies that there is bilirubin in the plasma. This is abnormal and requires that you notify the veterinarian immediately. Yellow coloration of the plasma is often one of the earliest signs of icterus, seen by the technician before the bilirubin blood values greatly elevate or before the animal has yellow coloration to their gums or skin. Icterus can result from the excessive breakdown of red blood cells, as might occur in an autoimmune hemolytic anemia. Other causes include liver disease, where the liver can not process or metabolize the bilirubin that is delivered to it, or obstruction or rupture of the biliary tract. Each cause of icterus is serious and requires additional testing and aggressive therapeutics for the underlying cause.

Bright red serum can be a result of free hemoglobin in the blood from rapid intravascular hemolysis of red blood cells or myoglobin from acute muscle breakdown. Both causes are serious and require immediate diagnostics and aggressive therapeutics to treat the underlying cause. Diuresis is required to protect the kidneys since free hemoglobin can cause acute renal failure. When hemolysis is due to a difficult venipuncture, the serum is only lightly red colored. Another sample may be necessary with a cleaner stick to rule out hemolysis.

White serum or plasma indicates lipemia. These fats in the serum can be the result of metabolic alternations or can appear after eating a meal. It is important that the veterinarian knows that there is lipemia, since other data base tests, such as serum sodium, will be considered in light of the lipemia (hyperlipemia and hyperproteinemia can cause pseudohyponatremia). Determination of total protein (TP) is important during initial screening. Disease processes such as edema, ascites, infections, coagulopathies, diarrhea, weight loss and renal and hepatic disease will alter TP. Albumin, globulin's and fibrinogen constitute the TP.

Decreases in TP can be due to: liver disease, exudation due to severe skin lesions (burns), protein losing enteropathies/nephropathies, vasculitis, excessive fluid therapy and hemorrhage. Serum protein level, because of the lack of fibrinogen, will be lower than plasma protein levels. Increases in TP can be due to: drug therapy, dehydration, feline infectious peritonitis (FIP), chronic inflammation and other disease process such as multiple myelomas. The plasma or serum from the hematocrit tube can also be used to determine the blood glucose and blood urea nitrogen.

Blood Glucose

Glucose is required as an energy source by all body cells, and is therefore essential to maintain an adequate level in the plasma. The glucose obtained after a meal is absorbed from the small intestine as an end product of carbohydrate digestion, which is then stored away as glycogen in the liver and muscles. Other times, in order to maintain normal plasma concentration, glucose can be released from the muscle although not as rapidly. Other means of maintaining plasma glucose involve the conversion of the liver glycogen (glycogenolysis), as



well as that, which is derived from non-carbohydrate sources (by hepatic gluconeogenesis). During periods of starvation, glucose is obtained from the breakdown of fats and proteins (primarily muscle) through gluconeogenesis in the liver and the kidneys.

Plasma glucose is regulated by the hormones glucagon and insulin. Glucagon serves to raise glucose level, whereas insulin lowers it. Glucagon, glucocorticoids (i.e. cortisol), adrenaline (epinephrine), growth hormone and progesterone all raise plasma glucose levels by increasing gluconeogenesis or glycogenolysis, or by interfering with the utilization of glucose. Insulin lowers glucose levels by inhibiting gluconeogenesis and increasing cellular uptake.

Glucose is filtered from the blood by the renal glomeruli and then totally absorbed in the tubules. When there is an excess in the glomerular filtrate, glucose will appear in the urine (glycosuria). Renal threshold in the dog is 180mg/dL, and in the cat 290mg/dL.

Increased plasma glucose levels can be due to increased production or release (fear, excitement, stress, severe trauma, post-prandial, seizures) and in conditions of inadequate or decreased usage as seen in diabetes mellitus.

Decreases in plasma glucose can be attributed to excess insulin (endogenous or iatrogenic), cortisol deficiency, hepatic disorders (vascular shunts, and glycogen storage diseases), impaired ability to produce glucose and/or excessive utilization (neoplasm, sepsis, polycythemia, and puppies).

Falsely elevated plasma glucose levels can be due to recent administration of injectables containing dextrose or carbohydrates, hemolysis or lipemia. Metronidazole can falsely elevate glucose when values are obtained using the hexokinase or oxidase method.

Falsely decreased levels can be due high levels of vitamin C (by inhibiting glucose oxidase/oxidase method, and failure to separate plasma or serum from the red cells (glycolytic enzymes in red cell continue to utilize glucose).

Glucose levels obtained from whole blood samples by the use of reagent strips (i.e. Ames) or reflectance meter (i.e. Glucometer) can be falsely low. As plasma is absorbed from the drop of whole blood, hemoconcentration of cells, proteins, and other less permeable particles occurs at the test strip surface, blocking diffusion. The reagent strip thus measures glucose content of the absorbed fluid rather than glucose concentration from the sample.

Blood Urea Nitrogen (BUN)

Urea is synthesized in the liver from ammonia, most of which comes from the breakdown (catabolism) of amino acids derived from tissue or dietary proteins. Also some ammonia is absorbed from the bowel, where it is formed by the action of bacteria on dietary amino acids and re-circulated endogenous urea, and carried to the liver. In the kidney, urea is freely filtered through the glomeruli and then passively reabsorbed in the tubules; normally about half is reabsorbed and reconverted to urea. One quarter of it is excreted by the gut, converted to ammonia, absorbed and then re-converted to urea.

Blood strip tests do not give precise values but can indicate whether or not urea levels are significantly increased. The presence of ammonia salts (i.e. ammonium oxalate as anticoagulant) will falsely elevate readings and ammonia vapor (or tobacco smoke) will falsely affect test strips (i.e. Azostix or Urastrat).

Causes for decreased plasma urea levels are low protein diet, anabolic steroids (diverting proteins from catabolism to tissue formation), liver failure, porto-systemic shunt, diabetes insipidus, psychogenic polydipsia and primary hyperammonemia.

Increases in urea can be due to: Pre-renal, renal or post-renal conditions.

Pre-renal Azotemia can be due to increased protein breakdown secondary to high protein diet, carbohydrate deficiency, intestinal hemorrhage, fever, necrosis, hyperthyroidism, prolonged



exercise, catabolic drugs, reduced anabolism and decreased renal perfusion..

Renal Azotemia is due to primary renal failure. Causes for renal azotemia include; acute or chronic interstitial nephritis, acute tubular necrosis, acute or chronic renal failure, chronic glomerulonephritis, chronic amyloidosis, chronic pyelonephritis, diffuse nephrocalcinosis, and neoplasia.

Post-renal Azotemia is usually seen with obstruction to urinary flow to due congenital disorders, calculi, neoplasms, blood clots, feline urological syndrome, surgical ligation, and prostatic disorders.

Post-renal azotemia can also be seen in-patient with a ruptured bladder or torn ureter.

