10 Abdominocentesis and Peritoneal Fluid Analysis

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Indications

Abdominocentesis refers to the technique of puncture of the abdominal wall and peritoneal cavity with a needle (or teat cannula) and collection of a sample of peritoneal fluid for analysis. Abdominocentesis is performed to obtain peritoneal fluid in cases of colic or enterocolitis as a means of assessing intestinal damage, or horses with a fever of unknown origin to diagnose peritonitis. Neoplastic cells can be seen using cytological evaluation of the peritoneal fluid in approximately 50% of gastrointestinal neoplasia cases and, therefore, peritoneal fluid analysis can also be useful in horses with unexplained weight loss, inappetence, and chronic intermittent colic.

Peritoneal fluid analysis is considered part of the minimum database for colic patients at many

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Table 10.1 Things you will need for abdominocentesis.

Needle technique	• Halter and lead rope • ± Stocks • ± Sedation (xylazine/butorphanol) • Clippers • Povidone-iodine or chlorhexidine scrub • Alcohol • Sterile gloves • 3–4 18 gauge 1 1/2 in. needle • EDTA tube (cytology) • Plain tube (bacterial culture and sensitivity testing)
Teat cannula technique	• Halter and lead rope • ± Stocks • ± Sedation (xylazine/butorphanol) • Clippers • Povidone-iodine or chlorhexidine scrub • Alcohol • 3 mL 2% lidocaine in a syringe and 22–25 gauge needle • Sterile gloves • #15 blade • Sterile 4 × 4 gauze sponges • Teat cannula • EDTA tube (cytology) • Plain tube (bacterial culture and sensitivity testing)

referral hospitals. Abdominocentesis and peritoneal fluid analysis is particularly useful when the decision to manage the horse medically versus surgically is not readily apparent (see Chapter 15 on Medical versus Surgical Management of the Horse with Colic [p. 164]) and for monitoring horses that are being managed medically.

Abdominocentesis is generally not performed on the initial examination for colic, particularly with horses showing mild signs (see Chapter 5 on Management of Mild Colic [p. 45]), because it is unlikely to yield results that would alter management and the procedure is not without the potential for complication. The decision to perform abdominocentesis should be made with caution in horses with severe abdominal distention or suspected sand impaction because of the risk for enterocentesis (p. 89). It is also not recommended to perform the procedure on horses with severe uncontrollable pain because of the risk of injury to the veterinarian and in such cases surgery or euthanasia is often indicated.

Indications for abdominocentesis prior to referral include horses with suspected gastrointestinal tract rupture/perforation and horses with clinical signs consistent with a strangulating obstruction where the owners do not chose to pursue referral or surgical treatment. Abdominocentesis and peritoneal fluid analysis in these cases can be used to support the indication for euthanasia.

Preparation

A "List of Things You Will Need" is provided in Table 10.1. Abdominocentesis is performed with the horse restrained in stocks or in a stall with a halter and lead rope. An area of 10 cm × 10 cm to the right of midline at the most dependent aspect of the ventral abdomen is clipped and aseptically prepared using povidone-iodine or chlorhexidine scrub and alcohol. Alternatively, the area on the cranial midline where the pectoral muscles create a V-shape can be used. Sonography (p. 116) can be used to locate abdominal structures that should be avoided (e.g., spleen and colon) and identify an area of peritoneal fluid accumulation; however, it is not particularly sensitive for the latter and abdominocentesis should still be performed when indicated even if peritoneal fluid is not identified sonographically. Abdominocentesis should not be performed to the left of midline because of the potential for splenic injury and subsequent hemoabdomen. Large subcutaneous vessels should be avoided to prevent sample contamination, hematoma formation, and hemorrhage.

Procedure

Abdominocentesis can be performed using an 18 gauge needle or a teat cannula. The use of an 18 gauge needle (see below) is easier and quicker and if an enterocentesis is performed the hole into the intestine is likely to be smaller; however, fluid is obtained more often with a teat cannula because of the more appropriate length and larger bore and proponents suggest that enterocentesis is less likely because of the blunt tip. The needle or teat cannula is manipulated for several minutes in an attempt to obtain a sample from an area of fluid accumulation. The sample is collected into an ethylenediaminetetraacetic acid (EDTA) tube for cytology and a

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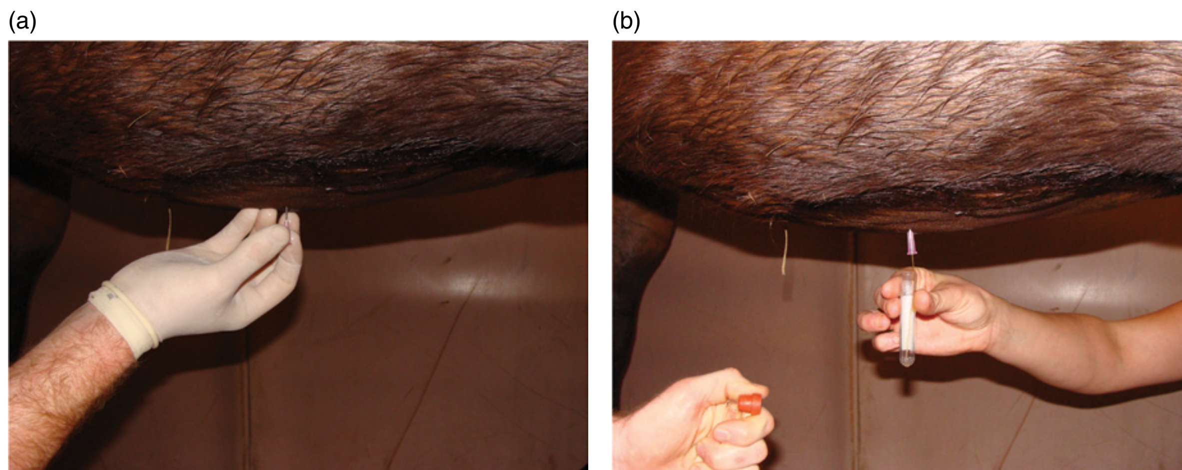

Figure 10.1 Abdominocentesis using the needle technique (a) with collection of peritoneal fluid into an EDTA tube (b).

sterile tube, syringe, or culture vial for bacterial culture and sensitivity testing.

Needle technique

Abdominocentesis using an 18 gauge needle is performed by carefully inserting the needle through the skin, subcutaneous tissue, body wall, and into peritoneal cavity (Figure 10.1). It can be difficult to determine when the tip of the needle is within the peritoneal cavity and in some cases an 18 gauge 1 ½ in. needle may be of insufficient length. Spontaneous movement of the needle is associated with intestinal movement and can be an indication that the needle is correctly positioned. Multiple needles are often needed to obtain a sample.

Teat cannula technique

Abdominocentesis using a teat cannula requires infiltration of the subcutaneous tissue and body wall with 3 mL of 2% lidocaine. A stab incision is made using a #15 blade through the skin and body wall. Gauze sponge is wrapped around the teat cannula to prevent sample contamination. The teat cannula is pushed through the body wall and into the peritoneal cavity; quite a bit of force is usually necessary. Correct positioning of the teat cannula within the peritoneal cavity can be confirmed by filling a 10 mL syringe with air and injecting it through the teat cannula; if the teat cannula is not in the peritoneal cavity the air will be heard rushing back out of the teat cannula.

Complications

The main complication with abdominocentesis is enterocentesis, which should be suspected when green-brown fluid is obtained in a horse with clinical signs inconsistent with gastrointestinal tract rupture/perforation. In the vast majority of cases, enterocentesis is without clinically apparent consequence. Inadvertent enterocentesis has a reported frequency rate of 2–5%.[4] The incidence of complications directly related to inadvertent enterocentesis was reported to be 0.5% (4/850 abdominocentesis samples).[25] When an enterocentesis occurs, the needle or teat cannula should be removed. Although prophylactic antimicrobial drug administration is recommended, the necessity is unknown. Localized cellulitis and peritonitis are rare consequences of enterocentesis and probably more likely to occur in horses with compromised bowel.

Injury to the spleen can occur and may happen more frequently in horses with a nephrosplenic entrapment whereby the spleen is moved ventrally and across midline. There is usually no consequence except that the results of peritoneal fluid analysis cannot be interpreted.

Omental herniation is a complication of abdominocentesis in foals using the teat cannula

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technique and occurs with such a frequency that many clinicians will avoid this procedure in neonates. Omental herniation can be corrected by sedating the foal and placing it in dorsal recumbency, aseptically preparing the area, exteriorizing an additional centimeter of the omentum, ligating the exteriorized omentum (if necessary), and placing it back in the abdomen with hemostats or a teat cannula. The skin is apposed using absorbable suture material or a skin staple. Abdominocentesis in foals should be performed with sonographic guidance and using a needle.

Peritoneal fluid analysis

The fluid that bathes the abdominal cavity is an ultrafiltrate of plasma that functions to reduce friction by lubrication. The constituents of peritoneal fluid are affected by the integrity of the mesothelial lining, changes in vascular permeability and lymphatic flow, plasma oncotic pressure, and capillary hydraulic pressure. Thus, changes in the character of the fluid can be attributed to specific disease processes and may yield information in the diagnosis, treatment, and/or prognosis of horses with colic.

Nonrepresentative sampling

When evaluating peritoneal fluid, it is important to determine whether the sample is representative. Two common causes of nonrepresentative peritoneal fluid sampling include blood contamination and enterocentesis. Blood contamination may occur when a peripheral vein or abdominal organ (usually the spleen) is punctured. During abdominocentesis, blood contamination is obvious when the initial sample is clear and becomes bloody or is bloody then clears. As little as 0.05 mL blood in 1 mL of peritoneal fluid (5% contamination) can result in fluid red blood cell (RBC) counts up to 449 000/μL, which is nearly ten times the upper limit of RBC reference values.[4,18] However, white blood cell (WBC) counts and total protein (TP) concentration remain within reference intervals with up to 17% blood contamination.[18] Enterocentesis may manifest grossly as a green to brown discoloration of peritoneal fluid, but contaminated fluid may also appear grossly normal with contamination only evident upon cytologic evaluation. Peritoneal fluid evaluation alone cannot distinguish between enterocentesis and peracute intestinal leakage or perforation. This distinction is best accomplished in the context of the patient's clinical assessment (p. 171) and sonographic evaluation (p. 133).

Gross characteristics

Volume

In equids, peritoneal fluid volume in health typically ranges from 100 to 300 mL,[1] although it has been estimated that up to 2 L may be present.[4] Although the equine peritoneal cavity normally contains a large volume of fluid, abdominocentesis typically yields <10 mL of fluid and most commonly 3–5 mL.[24] An effusion is defined as an increase in the normal volume of peritoneal fluid, which may or may not have increased protein or cell concentrations. The amount of fluid obtained from abdominocentesis does not necessarily correlate with the presence or absence of effusion. In seriously ill horses with effusion confirmed surgically or postmortem, abdominocentesis may yield only a small volume.[24] Diagnosis of mild to moderate effusions in equids, especially adults, can be difficult due to the constraints on palpation and a large abdominal space that accommodates even moderate effusion volumes. Sonography is often necessary to diagnose effusion (p. 133). In horses, it is convention to classify abnormal peritoneal fluid (i.e., increased protein concentration and/or cell count) as an effusion regardless of whether an increased volume is documented. In contrast, a fluid with a normal cell count and protein concentration is only classified as an effusion if there is a confirmed increase in volume.

Color and clarity

The physical characteristics of peritoneal fluid may provide hints as to its cellularity or biochemical composition. Normal peritoneal fluid is yellow and transparent. Discoloration of the fluid may reflect hemorrhage (orange to red), gastrointestinal tract rupture/perforation (green or brown), bile peritonitis (dark green), or vascular compromise

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(orange to reddish brown). Abnormal abdominal fluid color has been used in studies to predict the need for surgical treatment. The sensitivity and specificity of subjectively discolored fluid for predicting the need for surgical intervention varies in different reports: 92% and 74%,[19] 78% and 48%,[10] and 51% and 95%, respectively.[27] Weimann *et al*. used hemoglobin concentration as an objective measurement of discoloration from hemolysis and found that when peritoneal hemoglobin concentration was >0.01 mmol/L (0.02 g/dL), the test was 80% sensitive and 82% specific for selecting surgical treatment.[27] While discolored fluid, especially serosanguinous, supports the need for surgical treatment,[10,19,27] an interpretation of discolored fluid should only be made when the possibility of enterocentesis or blood contamination has been excluded and should be formulated in the context of clinical findings.

The clarity of abdominal fluid in health reflects low cellularity. Increased turbidity is usually reported in terms of cloudy/hazy or opaque, and suggests increased cellularity, presence of plant material, or, rarely, lipid (i.e., chyloabdomen). While turbidity is abnormal, measurement of cell numbers and cytologic evaluation of the fluid are necessary to determine its cause.

Biochemical evaluation

Many peritoneal fluid biochemical parameters have been assessed to determine their use in diagnosis and prognosis. Only tests that are readily available are discussed.

Protein concentration

Because peritoneal fluid is an ultrafiltrate of plasma, protein concentrations are much lower than plasma. Protein can be rapidly and accurately measured by handheld refractometers. Protein measured by refractometer for body cavity fluids is linearly related to protein measured by biochemical methods and results are accurate to at least 0.6 g/dL.[13] Many references cite the reference interval for normal peritoneal fluid protein concentration as <2.5 g/dL,[4] which is the lowest protein reading on many refractometers. To report values below the lower end of the protein scale, the value measured using either refractive index, refraction, or urine specific gravity (SG) scales can be converted to TP with published conversion tables.[12] When abdominal fluid protein was calculated using conversion tables or measured biochemically, normal values were typically ≤2.0 g/dL.[1,14,17] Increases in peritoneal fluid protein concentration above reference values indicate either increased permeability of the capillaries due to inflammatory mediators or increased hydraulic pressure within hepatic sinusoids. An increase in protein concentration due to inflammatory mediators is typically accompanied by an increase in nucleated cell count (NCCs).

Because refractometers measure protein via a total solids (TS)-based technique, the total dissolved solids in the sample affect light refraction.[12] In addition to protein, TS include electrolytes, glucose, urea, and lipids. While the altered refraction of body fluid is mostly due to protein content, increases in lipid, glucose, or urea content interfere with refractometric protein measurements. However, marked increases in urea or glucose (273 and 649 mg/dL, respectively) are needed to increase protein measurement by 0.4–0.5 g/dL.[20] Unlike glucose and urea, lipid does not freely diffuse into peritoneal fluid from plasma; increased concentrations of lipid in peritoneal fluid only occur in cases of chylous effusion (chyloabdomen).

Another potential cause of erroneous refractometer readings is the addition of EDTA from K_3EDTA anticoagulant tubes. At the standard concentration of EDTA (5 µmol/mL), K_3EDTA by itself has minimal effect on the fluid's refraction (≤0.1 g/dL increase). At higher concentrations of EDTA (10 and 20 µmol/mL), EDTA can increase refractometer total protein (TP_{Ref}) by 0.9–1.0 g/dL. Underfilling of EDTA tubes has the effect of increasing the EDTA concentration and will cause spurious increases in the TP_{Ref}. Some commercial tubes with K_3EDTA anticoagulant may also contain additives to prevent crystallization of the EDTA. Tubes that contain the additive may increase TP_{Ref} readings by up to 0.9 g/dL, even when properly filled.[9] It is recommended that TP_{Ref} readings should be performed on either fluid collected in serum tubes or in properly filled K_3EDTA tubes containing no additives. While sodium heparin anticoagulant has no effect on TP_{Ref},[9] heparin has deleterious effects on cellular morphology and is not recommended for samples that will be evaluated cytologically.

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The term "total solids" has caused much confusion in the reporting of refractometric protein results. TP and TS are not synonymous. Currently the majority of all refractometers incorporate a conversion factor in their design so that the scales report TP and not TS. Contributing to the confusion is the fact that at least one refractometer is named the "TS meter" (AO Corporation) when it is in fact calibrated to report TP.[12] Today nearly all refractometers report TP and not TS, so comparisons between peritoneal fluid samples are appropriate.

(SG) has also been used to estimate protein concentration in equine peritoneal fluid. However, use of the refractometer SG scale will yield erroneous results because the SG scale is calibrated specifically for urine. Urea, not protein, is the principal constituent of urine, thus SG calibrated for urine gives falsely high results for body fluids.[12] A caveat applies to low protein peritoneal fluid (<1.0 g/dL); in this instance, the urine SG scale more accurately estimates true SG because the fluid's refractive index is more similar to urine.[23]

Lactate and glucose concentration and pH

Lactate is the end product of anaerobic glycolysis and has been used as a marker for ischemia in colic. Normal peritoneal fluid lactate concentrations vary with respect to methodology, but are usually <1.0 mmol/L and the ratio of peritoneal fluid: plasma lactate concentration <1:1.[5,17] Peritoneal lactate, TP, NCC, and glucose concentrations were significantly increased in peritoneal fluid of horses with colic compared to clinically normal horses.[5,17] Peritoneal lactate concentrations were significantly higher in horses with strangulating compared to nonstrangulating lesions,[5,17] with the exception of strangulating small colon obstructions.[17] Increases in fluid lactate, decreases in fluid pH, and abnormal fluid color and/or turbidity were most strongly correlated with ischemic strangulating lesions. Peritoneal fluid lactate concentrations were shown to be more sensitive than blood lactate concentrations in detecting early ischemic lesions.[5,17] The prognosis becomes progressively poorer for horses with strangulating and nonstrangulating lesions as peritoneal fluid lactate concentration increases. One study showed the probability of death of horses with nonstrangulating lesions to be 63% for peritoneal lactate concentrations of 12 mmol/L and 82% for lactate concentrations of 16 mmol/L; the same concentrations of peritoneal fluid lactate had 82% and 92% probabilities of death, respectively, in strangulating lesions.[5] An increase in the peritoneal fluid lactate concentration with serial measurements several hours apart has been associated with the need for surgical treatment.[21]

Peritoneal glucose concentrations have been shown to be useful as a relatively rapid test for distinguishing septic from nonseptic exudates.[26] Positive bacterial culture or cytologic identification of intracellular bacteria in abdominal fluid is a definitive indicator of abdominal sepsis, but false-negatives do occur, especially with antimicrobial therapy. In one study of 36 peritonitis cases, peritoneal fluid glucose concentration and pH were significantly lower in horses with septic versus nonseptic peritonitis.[26] Serum to peritoneal fluid glucose concentration differences >50 mg/dL were most diagnostically useful in confirming sepsis.

D-dimer concentration

Mesothelial cells play an important role in the initiation and resolution of inflammation by secretion of immunomodulatory mediators, which impact coagulant and fibrinolytic factors such as D-dimer.[3,6] D-dimer concentrations in normal peritoneal fluid are reported to be <88 ng/mL,[6,7] whereas in horses with colic, median peritoneal fluid D-dimer concentrations ranged from 2023 to 24301 ng/mL depending upon diagnosis. In horses with colic, median fluid D-dimer concentrations were highest with enteritis, ischemic lesions, and septic peritonitis (8028, 16181, and 24301 ng/mL, respectively) compared with large colon obstructions (2023 ng/mL).[6] Blood contamination <20% does not alter peritoneal fluid D-dimer concentrations.[7] These studies show that peritoneal fluid D-dimer concentration appears to be a useful indicator of fibrinolytic activity and higher concentrations are correlated with greater disease severity.

Cells and cell counts

Cytologic evaluation of peritoneal fluid can yield important information that may even be diagnostic

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(e.g., large cell lymphoma, carcinoma, uroabdomen, septic inflammation). However, cytology should be interpreted in the context of clinical signs, cell counts, and biochemical evaluation.

Erythrocytes

Erythrocytes are not normally present in peritoneal fluid in health, but blood contamination during abdominocentesis results in RBC counts that range from <5 000/μL to <43 200/μL in healthy horses.[4,18,23] Visual assessment of discoloration attributable to RBCs was not detected for samples with <40 000 RBCs/μL,[15] thus typical amounts of blood contamination associated with abdominocentesis should not change the fluid color. Erythrocyte counts will increase above reference values because of minor blood vessel damage associated with necrosis and/or inflammation. See Hemorrhagic effusions (p. 97).

Nucleated cells

Total NCCs for abdominal fluid are performed with automated blood count analyzers or manually with a hemocytometer. Generally, NCC reference values in normal peritoneal fluid are reported as <10 000/μL,[4,23] and often <5 000/μL.[1,4,26] Many equine clinicians and clinical pathologists consider values <5 000/μL normal, values between 5 000 and 10 000/μL to be ambiguous, and values >10 000/μL as abnormal. Reference values for foals are <1 500/μL.[14] The cells normally present within the peritoneal cavity comprise leucocytes and mesothelial lining cells. Nucleated cells in cytologic differentials are categorized as neutrophils, lymphocytes, and large mononuclear cells (monocytes/macrophages and mesothelial cells).

In horses, mature neutrophils comprise the predominant cell in the differential evaluation (typically 50–80% of nucleated cells). Because neutrophils do not return to the vasculature, but age and die in body cavity fluid, pycnotic and hypersegmented neutrophils are normally noted in a proportion of the population. Increased numbers of neutrophils enter the peritoneal cavity in response to chemotactic inflammatory mediators. During inflammation, the ability of neutrophils to respond to stimuli and migrate into tissue increases with maturation.

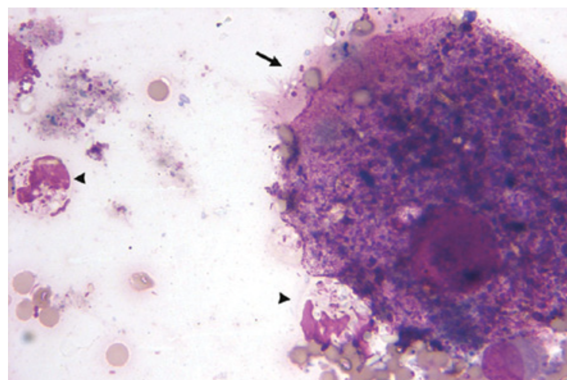

Figure 10.2 Peritoneal fluid from a horse with a septic abdomen. Neutrophils are degenerate and contain many cytoplasmic bacterial rods and cocci (arrowheads). The presence of a large ciliated protozoal organism indicates that there has been intestinal rupture or leakage (arrow).

Consequently, mature neutrophils arrive first and immature forms (i.e., bands, metamyelocytes) are only present in tissue when mature neutrophils have been severely depleted. Thus, the presence of band or earlier forms of neutrophils in peritoneal fluid indicates severe inflammation; a caveat would be if the immature forms originate from blood and there is significant blood contamination of the sample. If blood contamination is not apparent, the presence of immature neutrophils in peritoneal fluid is associated with a poorer prognosis (i.e., need for surgical intervention and increased mortality rates).[11]

Degenerate change in neutrophils is characterized by swollen, pale-staining chromatin and loss of membrane integrity (Figure 10.2). While degenerate change is not pathognomonic for infection, the presence of degenerate neutrophils warrants a more thorough search for organisms. These changes occur *after* the cell has left the blood and indicate release of neutrophil enzymes and/or exposure to bacterial cytotoxins. In contrast, "toxic" neutrophils are characterized by the presence of cytoplasmic basophilia (often with Dohle bodies), foamy cytoplasm, less condensed chromatin, nuclear hyposegmentation, and larger cell size. These changes reflect maturational defects in the bone marrow that are associated with rapid neutropoiesis due to a strong systemic inflammatory stimulus. Toxic change exists in neutrophils *before* they enter the peritoneal cavity. Toxic change is

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often associated with severe bacterial infections or significant tissue necrosis.

Large mononuclear cells encompass both monocytes/macrophages and mesothelial cells. When monocytes exit the vasculature, they differentiate into macrophages; both forms will be present in body cavities (Figure 10.3). Macrophages will occasionally contain senescent neutrophils. As with neutrophils, increases in monocyte/macrophage concentrations in fluid are attributable to inflammation and usually accompany neutrophil increases; neutrophil proportions typically exceed those of monocyte/macrophages in acute inflammation, whereas mononuclear cells may predominate with chronic effusions and certain disease processes (e.g., fungal infection).

Mesothelial cells are readily identified when they are present in cohesive aggregates (Figure 10.4), and in horses they are rare in both normal fluid and effusions. Although included in the large mononuclear cell group, they comprise only a minor fraction of the large mononuclear cell differential.

The lymphocytes in normal peritoneal fluid are small to medium sized (7–10 μm diameter) and constitute the smallest proportion of the three main cell types in normal peritoneal fluid. Increases in lymphocytes may be associated with chronic inflammatory conditions, parasitism, or chyloabdomen. In chronic inflammation, the lymphocyte population is often a mixture of small lymphocytes, medium lymphocytes (prolymphocytes), reactive lymphocytes, and plasma cells. Reactive lymphocytes are usually slightly larger than small lymphocytes (9–11 μm diameter) and are characterized as having deeply basophilic cytoplasm and a mature chromatin. Lymphoblasts are large lymphocytes (12–16 μm diameter) whose nuclei have immature chromatin and usually contain a nucleolus or nucleoli. The presence of lymphoblasts is abnormal and is usually associated with large cell lymphoma. However, in the context of reactive lymphocytes and plasma cells, rare lymphoblasts may be part of the inflammatory response.

Other cell types such as eosinophils, basophils, and mast cells do not contribute to the cellular differential in normal abdominal fluid. The largest increases in eosinophil numbers can be associated with parasitic infections (including larval migration), but smaller increases may be seen with inflammation due to any etiology. Eosinophilia in peritoneal fluid (Figure 10.5) may also be seen with focal eosinophilic enteritis[22] and, rarely, lymphoma.[16] Basophils and mast cells can increase concurrently with eosinophils, though both are rare findings, even in inflammation.

Neoplastic cells can occasionally be found in peritoneal fluid. Lymphoma (Figure 10.6) and carcinoma (Figure 10.7) are the most common neoplasms identified in peritoneal effusions.

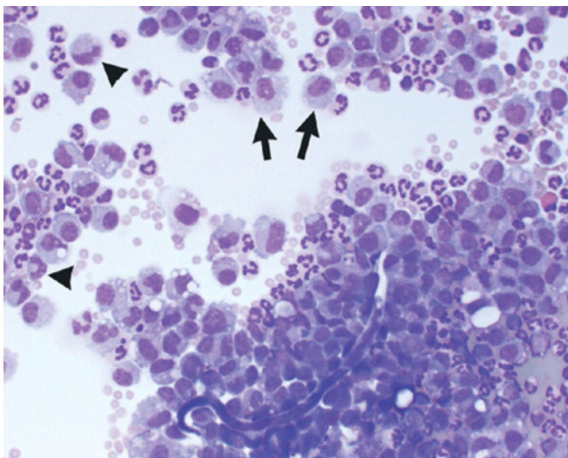

Figure 10.3 Concentrated preparation of normal peritoneal fluid containing both monocytes (arrowheads) and macrophages (arrows). Note the presence of a microfilarial organism in the bottom center. *Setaria* spp. are an incidental finding, especially in horses that have not been routinely dewormed.

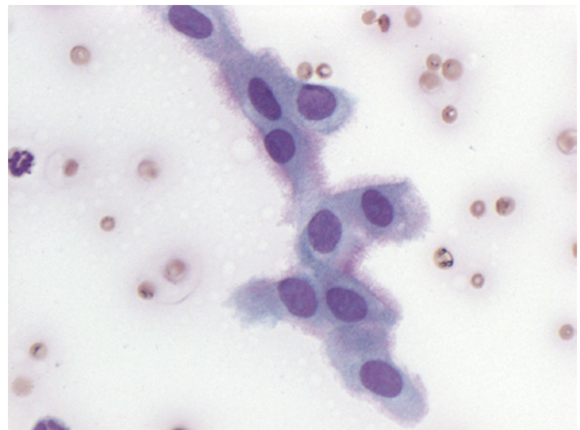

Figure 10.4 Mesothelial cell cluster. Mesothelial cells are round to polygonal, uniform, and cohesive.

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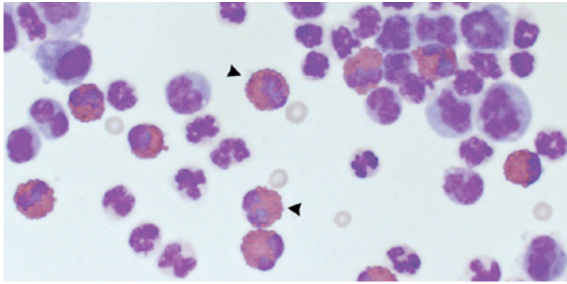

Figure 10.5 Eosinophilic inflammation in peritoneal fluid. Neutrophils comprise the predominant leukocyte, but significant numbers of eosinophils are present (arrowheads).

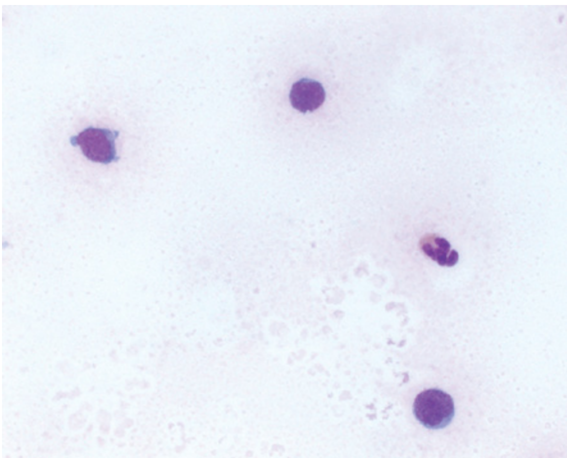

Figure 10.6 Peritoneal fluid from a horse with large cell lymphoma. Three large immature lymphocytes are present; note the size of the three lymphocytes compared to the neutrophil.

Interpretation of findings

The main categories of effusion are formulated to provide insight into the general pathophysiologic mechanism responsible for an increase in the volume of body cavity fluid. Effusions are caused mainly by transudative, exudative, or hemorrhagic processes (Table 10.2). A fourth category, which is uncommon in horses, is lymphorrhagic effusion caused by leakage of lymph from lymphatic vessels (e.g., chyloabdomen).[23]

Transudates

Transudates are caused by increased hydraulic pressure or increased hydraulic and decreased oncotic pressure. These effusions have low protein concentrations and NCCs. In horses, normal peritoneal fluid is distinguished from a transudate only by documenting an increase in fluid volume. The most common causes of transudates are the per-acute/acute phase of any lesion causing decreased venous/lymphatic drainage in the portal system (e.g., volvulus/torsion, neoplasia, granuloma), lymphatic obstruction, acute uroabdomen, and protein-losing nephropathies and enteropathies. Diagnosis of uroabdomen in the horse is facilitated by the characteristic presence of calcium carbonate crystals in equine urine, which are present extracellularly and within neutrophils (Figure 10.8). Suspicion of uroabdomen should be confirmed by measuring fluid creatinine concentration.

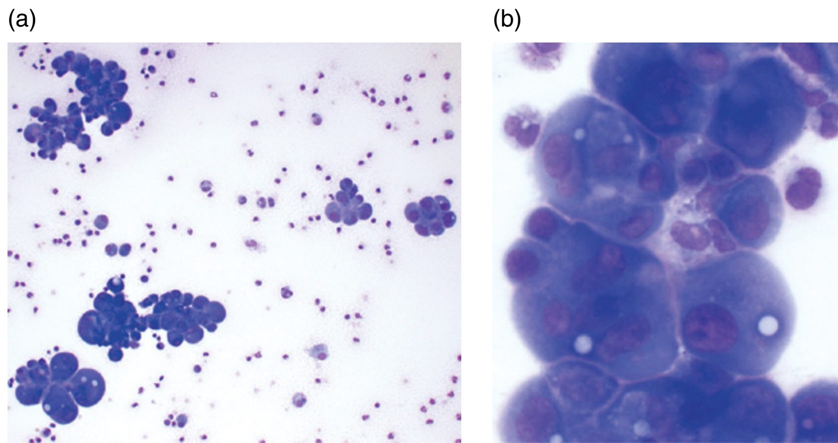

Figure 10.7 (a) Many clusters of large, cohesive cells in abdominal fluid. (b) The marked variability in cell and nuclear sizes, multinucleation, and prominent nucleoli are criteria of malignancy. The cohesive nature implies either an epithelial (carcinoma) or mesothelial (mesothelioma) phenotype.

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Table 10.2 Effusion classifications.

Classification	TP_{Ref}	NCC	RBC	Mechanism	Disorder
Transudate	<2.0	$<5\times10^3$	$<4\times10^4$	↑Hydraulic and ↓oncotic pressure	• Nephrotic syndrome • Cirrhosis • Acute uroabdomen
High-protein transudate ("modified")	≥2.0	$<5\times10^3$	$<4\times10^4$	↓Hydraulic and ↓oncotic pressure in postsinusoidal or sinusoidal hepatic vessels	• Congestive heart failure • Portal hypertension (postsinusoidal)
Exudate	>2.0	$>5\times10^3$	4×10^4–5×10^5	↑Vascular permeability due to inflammatory mediators	*Septic*: • Bacterial, fungal, or parasitic infection *Nonseptic*: • Neoplasia • Ischemic necrosis • Pancreatitis • Bile peritonitis • Chronic uroabdomen
Hemorrhage (acute)	≥2.0	≥ or $\leq5\times10^3$	$\geq1\times10^6$	Loss of blood from vessels	• Trauma • Neoplasia • Hemostatic defects • Uterine artery rupture

TP_{Ref}, total protein by refractometer (g/dL); NCC, nucleated cell count/μL; RBC, red blood cells/μL.

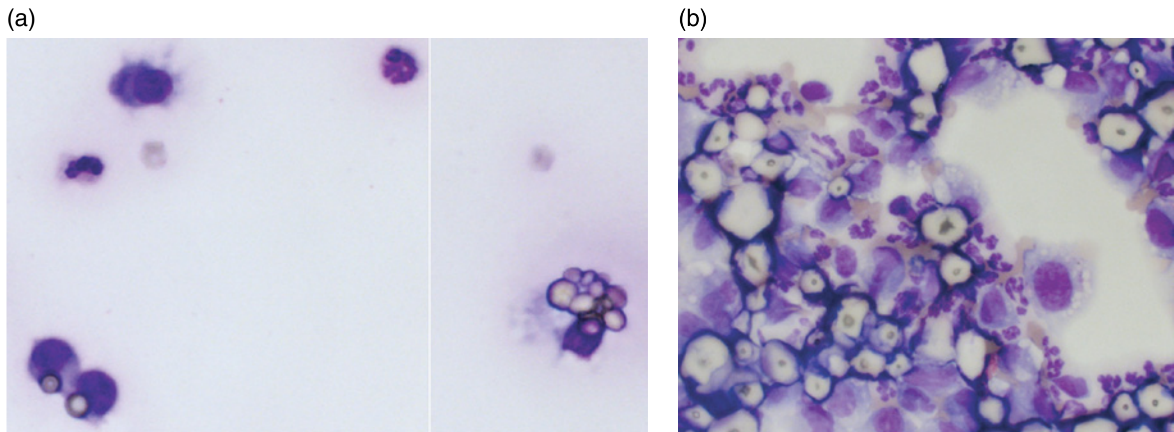

Figure 10.8 (a) Peritoneal fluid from an adult male horse with colic. Note the presence of round to oval calcium carbonate crystals within three macrophages. (b) Concentrated preparation of normal peritoneal fluid. There are many round to polygonal crystals with a characteristic central divet consistent with starch granules from surgical gloves. These are common contaminants that should not be mistaken for calcium carbonate crystals.

Plasma protein permeability of vessels within the hepatic sinuses is higher than that for vessels elsewhere in the peritoneal cavity. Consequently, increases in hydraulic pressure that involve the hepatic sinuses (i.e., sinusoidal or postsinusoidal increases) will produce a higher protein transudate with a normal NCC. Congestive heart failure and any lesion that produces postsinusoidal portal

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hypertension are the most common causes of high-protein transudates in the peritoneal cavity.

Exudates

Exudates are characterized by increased vascular permeability due to inflammatory mediators and therefore are attributable to inflammation. Inflammation may be caused by infectious or non-infectious processes (Table 10.2). The absence of microorganisms on cytologic examination does not preclude an infectious process. If clinical findings are suspicious of an infectious process, biochemical evaluation of the fluid (for lactate, glucose, and pH) and bacterial culture are warranted.

Hemorrhagic effusions

Uniformly hemorrhagic specimens or consistently hemorrhagic specimens from different abdominocentesis sites suggest true abdominal hemorrhage, which may be confirmed via cytology and/or sonographic evaluation of the abdomen (p. 134). Because platelets (and coagulation factors) are consumed during hemorrhage, hemorrhagic fluid contains no platelets and does not clot, whereas fluid with blood from peripheral or splenic vessels will contain platelets and will clot. A hemorrhagic effusion is defined by the fluid RBC count (Table 10.2); acute hemorrhagic effusions usually have RBC counts >1 000 000/μL and packed cell volumes >3%.[23] Erythrocytes and plasma from hemorrhage will be resorbed by lymphatics so the fluid TP and RBC counts vary with time; in dogs ~65% of RBCs are resorbed within 2 days of hemorrhage.[2]

Cytologic evaluation of abdominal fluid can be helpful in determining whether there is true hemorrhage. In fluids with high RBC counts, the presence of platelets indicates a component of blood contamination since platelets are not present in hemorrhagic effusions. However, the absence of platelets does not preclude peripheral blood contamination if the sample was collected into a non-anticoagulant tube (e.g., red top tube) and subsequently clotted *in vitro*. Cytologic findings that support hemoabdomen include the presence of erythrocytes and/or hemosiderin within macrophages. If there is a delay in sample processing (>3–4 h), RBC phagocytosis can occur *in vitro* in the transport tube. Conversely, erythrophagocytosis can be absent in samples from true hemorrhage if there has been insufficient time for RBC phagocytosis within the peritoneal cavity (i.e., peracute hemorrhage). In general, it takes several hours for erythrophagocytosis to occur; subsequent RBC breakdown into hemosiderin usually takes at least 3 days.[8] Thus, cytologic interpretations of hemorrhagic fluid should be undertaken with knowledge of the type of collection tube used as well as the amount of time before the sample was processed.

Application of peritoneal fluid analysis

Abdominocentesis is a valuable clinical tool whose sensitivity and specificity are highest when all components of fluid evaluation are considered together in conjunction with clinical findings and initial response to treatment (Clinical scenarios 5, 7–9, located in Appendix A). All fluid evaluations should include assessment of gross appearance, TP concentration, cell counts (or estimation of cellularity), and cytology. If this is not possible, the minimum testing for fluid evaluation should include measurement of TP and measurement (or estimation) of cellularity, because effusions are defined by these parameters.

References

1. Brownlow, M.A., Hutchins, D.R. & Johnston, K.G. (1981) Reference values for equine peritoneal fluid. *Equine Veterinary Journal*, **13**, 127–130.
2. Clark, C.H. & Woodley, C.H. (1959) The absorption of red blood cells after parenteral injection at various sites. *American Journal of Veterinary Research*, **20**, 1062–1066.
3. Collatos, C., Barton, M.H., Prasse, K.W. & Moore, J.N. (1995) Intravascular and peritoneal coagulation and fibrinolysis in horses with acute gastrointestinal tract diseases. *Journal of the American Veterinary Medical Association*, **207**, 465–470.
4. DeHeer, H.L., Parry, B.W. & Grindem, C.B. (2002) Peritoneal fluid. In: *Diagnostic Cytology and Hematology of the Horse* (eds R.L. Cowell & R.D. Tyler), 2nd edn., pp. 127–162. Mosby, St. Louis, Missouri.
5. Delesalle, C., Dewulf, J., Lefebvre, R.A., *et al*. (2007) Determination of lactate concentrations in blood plasma and peritoneal fluid in horses with colic by an Accusport analyzer. *Journal of Veterinary Internal Medicine*, **21**, 293–301.

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6. Delgado, M.A., Monreal, L., Armengou, L., Ríos, J. & Segura, D. (2009) Peritoneal D-dimer concentration for assessing peritoneal fibrinolytic activity in horses with colic. *Journal of Veterinary Internal Medicine*, **23**, 882–889.
7. Delgado, M.A., Monreal, L., Armengou, L., Segura, D. & Ríos, J. (2009) Effects of blood contamination on peritoneal D-dimer concentration in horses with colic. *Journal of Veterinary Internal Medicine*, **23**, 1232–1238.
8. Epstein, C.E., Elidemir, O., Colasurdo, G.N. & Fan, L.L. (2001) Time course of hemosiderin production by alveolar macrophages in a murine model. *Chest*, **120**, 2013–2020.
9. Estepa, J.C., Lopez, I., Mayer-Valor, R., Rodriguez, M. & Aguilera-Tejero, E. (2006) The influence of anticoagulants on the measurement of total protein concentration in equine peritoneal fluid. *Research in Veterinary Science*, **80**, 5–10.
10. Freden, G.O., Provost, P.J. & Rand, W.M. (1998) Reliability of using results of abdominal fluid analysis to determine treatment and predict lesion type and outcome for horses with colic: 218 cases (1991–1994). *Journal of the American Veterinary Medical Association*, **213**, 1012–1015.
11. Garma-Avina, A. (1998) Cytology of 100 samples of abdominal fluid from 100 horses with abdominal disease. *Equine Veterinary Journal*, **30**, 435–444.
12. George, J.W. (2001) The usefulness and limitations of hand-held refractometers in veterinary laboratory medicine: An historical and technical review. *Veterinary Clinical Pathology*, **30**, 201–210.
13. George, J.W. & O'Neill, S.L. (2001) Comparison of refractometer and biuret methods for total protein measurement in body cavity fluids. *Veterinary Clinical Pathology*, **30**, 16–18.
14. Grindem, C.B., Fairley, N.M., Uhlinger, C.A. & Crane, S.A. (1990) Peritoneal fluid values from healthy foals. *Equine Veterinary Journal*, **22**, 359–361.
15. Hunt, E., Tennant, B.C. & Whitlock, R.H. (1985) Interpretation of peritoneal fluid erythrocyte counts in horses with abdominal disease. In: *Proceedings of the Second Equine Colic Research Symposium*, University of Georgia, Athens, Georgia, pp. 168–174.
16. La Perle, K.M.D., Piercy, R.J. & Blomme, E.A.G. (1998) Multisystemic, eosinophilic, epitheliotropic disease with intestinal lymphosarcoma in a horse. *Veterinary Pathology*, **35**, 144–146.
17. Latson, K.M., Nieto, J.E., Beldomenico, P.M. & Snyder, J.R. (2005) Evaluation of peritoneal fluid lactate as a marker of intestinal ischaemia in equine colic. *Equine Veterinary Journal*, **37**, 342–346.
18. Malark, J.A., Peyton, L.C. & Galvin, M.J. (1992) Effects of blood contamination on equine peritoneal fluid analysis. *Journal of the American Veterinary Medical Association*, **201**, 1545–1548.
19. Matthews, A., Dart, A.J., Reid, S.W., Dowling, B.A. & Hodgson, D.R. (2002) Predictive values, sensitivity and specificity of abdominal fluid variables in determining the need for surgery in horses with an acute abdominal crisis. *Australian Veterinary Journal*, **80**, 132–136.
20. McSherry, B.J. & Al-Baker, J. (1976) Comparison of total serum protein determined by T/S meter and biuret technique. *Bulletin of the American Society of Veterinary Clinical Pathologists*, **3**, 4–12.
21. Peloso, J.G. & Cohen, N.D. (2012) Use of serial measurements of peritoneal fluid lactate concentration to identify strangulating intestinal lesions in referred horses with signs of colic. *Journal of the American Veterinary Medical Association*, **240**, 1208–1217.
22. Southwood, L.L., Kawcak, C.E., Trotter, G.W., Stashak, T.S. & Frisbie, D.D. (2000) Idiopathic focal eosinophilic enteritis associated with small intestinal obstruction in 6 horses. *Veterinary Surgery*, **29**, 415–419.
23. Stockham, S.L. & Scott, M.A. (2008) Cavitary effusions. In: *Fundamentals of Veterinary Clinical* (eds S.L. Stockham & M.A. Scott), 2nd edn., pp. 831–868. Blackwell Publishing, Ames, Iowa.
24. Swansick, R.A. & Wilkinson, J.S. (1976) A clinical evaluation of abdominal paracentesis in the horse. *Australian Veterinary Journal*, **52**, 109–116.
25. Tulleners, E.P. (1983) Complications of abdominocentesis in the horse. *Journal of the American Veterinary Medical Association*, **182**, 232–234.
26. Van Hoogmoed, L., Rodger, L.D., Spier, S.J., Gardner, I.A., Yarbrough, T.B. & Snyder, J.R. (1999) Evaluation of peritoneal fluid pH, glucose concentration, and lactate dehydrogenase activity for detection of septic peritonitis in horses. *Journal of the American Veterinary Medical Association*, **214**, 1032–1036.
27. Weimann, C.D., Thoefner, M.B. & Jensen, A.L. (2002) Spectrophotometric assessment of peritoneal fluid haemoglobin in colic horses: An aid to selecting medical vs. surgical treatment. *Equine Veterinary Journal*, **34**, 523–527.

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