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Cerebrospinal Fluid and Central Nervous System Cytology

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CEREBROSPINAL FLUID

Cerebrospinal fluid (CSF) is present within the ventricular system of the brain, the central canal of the spinal cord, and the subarachnoid space (SAS) between the pia mater and the arachnoid mater. CSF is a component of, and is continuous with, the interstitial fluid of the central nervous system (CNS). It is separated from the bloodstream and from the CNS parenchyma by an intricate barrier system comprising ependymal epithelium, choroid plexus epithelium, the leptomeninges, areas of modified leptomeninges, and the arachnoid villi (the reader is referred elsewhere for a thorough discussion of these barriers and their transport mechanisms).[1,2] CSF has mechanical (protection) and metabolic (transport, excretion) functions. Sampling of the CSF is an important part of the minimum database for patients with neurological signs and may be useful in monitoring response to therapy in CNS inflammatory disease. When performed correctly, acquisition is a rapid, inexpensive, and technically simple method of sampling the local environment of the CNS extracellular space for evidence of inflammatory, neoplastic, traumatic, or degenerative disease. It is not without risk, however, and should be performed judiciously, that is, when clinical indication exists and no contraindications are present. This chapter will review the biology of CSF, methods for collection, and causes for abnormalities in parameters, such as protein concentration and nucleated cell count.

Limitations of Cerebrospinal Fluid Analysis

Analysis of CSF is an important adjunctive diagnostic tool in the workup of patients with CNS disease and must be interpreted within the context of the patient's history, clinical signs, clinicopathological data, imaging studies, and other ancillary diagnostics. Rarely is CSF solely used to provide an etiological diagnosis (exceptions include cytological visualization of infectious agents or overtly neoplastic cells), but analysis may significantly narrow the field of pathophysiological differentials, guiding further diagnostic and therapeutic options. CSF analysis is most sensitive in detecting inflammatory disease.[3] Positive findings in CSF tend to be more diagnostically helpful compared with negative findings but are often nonspecific because many different diseases may cause a common CSF pathology (e.g., neutrophilic pleocytosis).[4] Occasionally, the magnitude of change within the CSF may be as instructive as the character of the change (e.g., a marked increase in protein concentration raising diagnostic concern for feline infectious peritonitis, marked neutrophilic pleocytosis raising diagnostic concern for steroid-responsive meningitis–arteritis in a young dog in pain).[4] More frequently, however, specific disease etiologies will present with CSF changes of variable character and magnitude. CSF that falls within laboratory reference intervals should never be used to rule out a differential diagnosis because negative findings may represent early or mild disease, disease suppressed or masked by therapeutic intervention, or a disease process that does not present within the particular area of the extracellular space being sampled. CSF analysis may or may not correlate with imaging studies; a retrospective study of 92 cats receiving magnetic resonance imaging (MRI) for spinal signs showed that abnormal CSF was not a predictor for abnormal MRI.[5] In another study, approximately 25% of dogs with intracranial signs and inflammatory CSF had normal brain MRI results.[6]

Formation and Movement of Cerebrospinal Fluid

The conventionally accepted theory of CSF secretion and transport is based on the concept of active transport of ions within the ventricular ependymal cells and choroid plexi, subsequent passive flow of fluid, and circulation and drainage of CSF into dural venous sinuses. These ideas have recently come under scrutiny as potentially simplistic and inconsistent with the past 100 years of experimental evidence.[7] Analysis of past experiments, coupled with new data, supports a "global production" hypothesis—that instead of exclusive formation within the ventricles, CSF is continually created and reabsorbed diffusely by cerebral capillaries that have slight variances in hydrostatic and osmotic pressure. Canine studies have documented CSF production within the ventricular system and the SAS.[2]

Contraindications to Acquisition of Cerebrospinal Fluid

CSF should not be collected from patients with unacceptable anesthetic risk or with suspected coagulopathy, severe cervical trauma, or increased intracranial pressure secondary to edema, hemorrhage, hydrocephalus, or a large neoplasm.[8] CSF collection in the presence of elevated intracranial pressure may cause brain herniation and death secondary to compression of respiratory centers.[9] Signs of increased intracranial pressure may include stupor, coma, bradycardia, systemic hypertension, cranial nerve deficits, rigid paresis, or all of these.[10,11] Mannitol and hypertonic saline are the first-line medical therapies for elevated intracranial pressure. Head elevation, modest hyperventilation, administration of drugs to slow brain metabolism, and craniectomy with durectomy are sometimes used in cases refractory to traditional treatments. Advanced imaging before CSF collection, especially in patients presenting with intracranial neurological signs, may be useful in identifying contraindications. Imaging, in particular MRI, is exquisitely helpful in providing structural data that may be correlated to CSF results.

Collection Techniques

Collection Sites

CSF can be collected from the cerebromedullary cistern (at the atlanto-occipital space) or from the lumbar cistern in the L5-L6

interarcuate space. The cerebromedullary cistern is used more commonly because a larger volume of CSF with lower risk for blood contamination can be reliably collected.

Cerebromedullary Cistern Versus Lumbar Cistern

A study of 158 dogs with focal, noninflammatory disease showed that in cases of spinal lesions, CSF was more likely to be abnormal if collected from the lumbar cistern, that is, caudal to the lesion.[12] This observation may be explained by presupposing cranial to caudal flow of CSF, but the traditionally held theory of CSF flow has recently been contested.[7] In canines, CSF collected from the cerebromedullary cistern generally has lower microprotein concentrations compared with samples collected from the lumbar cistern.[13] Blood contamination may be more pronounced in lumbar collection, as the desired subarachnoid space is more difficult to enter and yields a smaller volume of fluid that tends to flow more slowly.[9,10] Moreover, hemodilution may contribute to increased measured protein concentration.[13]

Rare instances of CSF contamination with hematopoietic precursors have only been reported from lumbar sites.[14] A low, but potentially catastrophic, risk for puncturing the cervical spinal cord or caudal brainstem exists during cerebromedullary collection. Because the spinal cord length is variable, spinal cord puncture is a possibility during lumbar collection, but it is associated with less severe adverse effects compared with injury following cisternal puncture. In a case series of four accidental cisternal parenchymal punctures (documented by using MRI), three of the four patients suffered neurological decompensation and subsequently had to be euthanized.[15]

Equipment

The following equipment should be assembled: anesthesia and monitoring equipment, clippers, aseptic preparation materials for the skin, sterile gloves, and a spinal needle with stylet. For cerebromedullary cistern collection in dogs weighing less than 25 kg and for cats, a 22-gauge, 1.5-inch spinal needle is usually adequate, and a 22-gauge, 2.5-inch spinal needle is recommended for dogs weighing greater than 25 kg. For lumbar puncture, a 22-gauge spinal needle up to 6 inches long may be required for obese or extremely large patients. If available, fluoroscopic equipment may aid in the acquisition of cisternal or lumbar CSF. At the authors' institution, fluoroscopy is often used before cisternal CSF acquisition in toy-breed dogs to exclude the possibility of subclinical atlantoaxial subluxation.

Cerebrospinal Fluid Acquisition

The anesthetized patient is placed in lateral recumbency (it is generally easier for a right-handed clinician to have the patient in right-lateral recumbency, and vice versa), with the neck and back flush to the edge of a sturdy table. For collection from the cerebromedullary cistern, the neck is flexed such that the dorsum of the muzzle is 90 degrees to the long axis of the body (if needed, stabilizing the endotracheal tube to prevent kinking and deflating the cuff to prevent tracheal trauma), and the snout is propped up slightly, if necessary, to keep it parallel with the table and not angulated from the sagittal plane.[10] A wide area (3–5 cm) around the atlanto-occipital joint (beyond atlas wings and axis spinous process and to the external occipital protuberance) is shaved and aseptically prepared, and landmarks are palpated with a gloved, nondominant hand.[9] The needle is inserted at the intersection of two imaginary perpendicular lines that run (1) along the dorsal midline (dividing the patient sagittally) from the occipital protuberance to the cranial spinous process of the axis (C2) and (2) across the craniolateral aspects of the wings of the atlas (C1) (dividing the patient craniocaudally).

For lumbar collection, the pelvic limbs are brought forward into full flexion, and the needle is inserted cranial and parallel to the dorsal spinous process of L6 for dogs and L7 for cats, advancing the needle until the ventral aspect of the vertebral canal is encountered; the needle is then retracted slightly and CSF is collected from the ventral SAS.[10,16] The pelvic limbs may be kicked or may twitch slightly during collection because of irritation of the cauda equina or spinal cord parenchyma.

For either location, once landmarks are palpated, the needle is held stably with the dominant hand and very slowly advanced, stylet in place. The heel of the dominant hand may be supported against the table. For cisternal collection, it is important to advance the needle toward the point of the nose without angulation. The stylet is removed with the nondominant hand every 2 to 3 mm to check for fluid within the needle hub, waiting a few seconds. It is common to feel a decrease in resistance to forward needle movement once the thecal space is entered. If bone is hit or frank hemorrhage is observed from the needle, it should be withdrawn slowly and collection reattempted.[9] If clear or slightly blood-tinged fluid is observed, advancement of the needle is stopped, and open tubes are placed directly under the needle hub to collect freely falling drops. CSF is collected passively and should not be aspirated.

There are no significant objective data regarding the maximal amount of CSF that may be collected in dogs. Several authors claim that it is safe to collect 0.2 milliliters (mL) of CSF per kilogram of body weight (1 mL/5 kg); in other species much higher volumes of CSF per body weight are acquired standardly.[17] In general, 0.5 to 1 mL of CSF is adequate for routine diagnostic tests, including cell counts, protein concentration, and cytological analysis. Larger volumes are necessary for additional diagnostics (cultures, titers, polymerase chain reaction [PCR], flow cytometry, protein electrophoresis, etc.).

Two sets of tubes should be readied and ideally handled by an assistant. An ethylenediaminetetraacetic acid (EDTA)–treated (purple-top) tube is used for cell counts, flow cytometry, and PCR testing for organisms, and plain (red-top) tubes are used for protein concentration, culture, or immunologic assays.[10] Some sources indicate that plain tubes are recommended, as EDTA could increase protein concentration. If CSF analysis will occur rapidly (within 1 hour), collection into a plain tube is adequate, whereas preservation of cells may be improved with collection into EDTA if analysis will be delayed. If low volume is present, priority is given to the EDTA tube. If CSF appears red, then iatrogenic hemorrhage (puncture of a dural vessel) or actual CNS hemorrhage has occurred. In this instance, the first few drops are allowed to collect into the first set of tubes, and the second set of tubes are reserved for the latter portion of the sample, as iatrogenic hemorrhage tends to clear over time. If the hemorrhage does clear, a decision may be made about discarding the first set of tubes or keeping them for ancillary testing not affected by the hemorrhage. After collection, the needle is withdrawn without the stylet, and the CSF within the needle is allowed to drip into one of the tubes or is placed in an additional plain tube and saved for culture.

CEREBROSPINAL FLUID PROCESSING AND ANALYSIS

As with other clinicopathological and cytological samples, evaluation of a fresh specimen is preferred to minimize cellular degradation, to which CSF is particularly vulnerable because of its relatively low protein concentration. Sample degradation will affect cell differential count to a greater extent than the total nucleated cell count or the protein concentration.[18] A study of 30 canine CSF samples with pleocytosis concluded that delay of analysis up to 8 hours was unlikely to alter interpretation, especially in samples with protein concentrations above 50 milligrams per deciliter (mg/dL).[18] Preservative should be added to low protein samples unless analysis is to be completed within

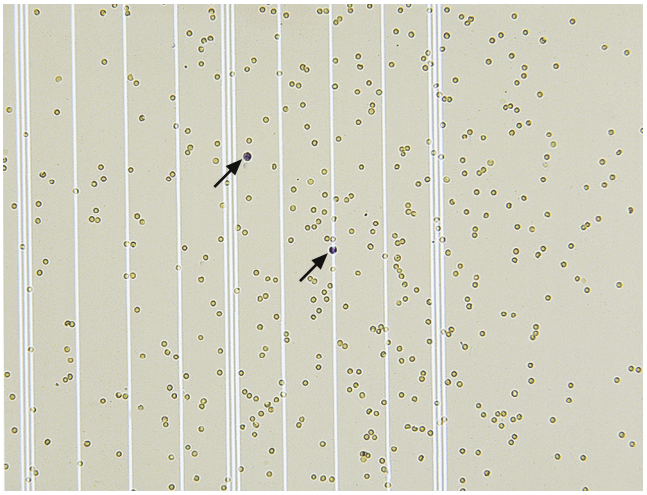

Fig. 14.1 Numerous erythrocytes and two leukocytes present on a hemacytometer. The two nuclei of the two leukocytes in the center of the field stain dark purple *(arrows)* (new methylene blue stain, original magnification 50×).

60 minutes (see next section), and a dilutional effect must then be factored into cell counts.[18] Samples to be shipped to a reference laboratory overnight should be kept at refrigeration temperature and shipped with ice packs for analysis within 48 hours.[9,16] The reference laboratory should be prenotified to ensure prompt analysis.

If analysis is likely to be delayed by more than 1 hour and the CSF sample has a protein concentration less than 50 mg/dL, one of the following may be added as a protein source to maintain cellular integrity: (1) hetastarch (add 1:1 volume), (2) fetal calf serum (3.7 g/dL protein; add 20% by volume), or (3) autologous plasma or serum (fresh or frozen; 11% by volume ≡ one drop from 25-gauge needle (approximately 0.03 mL) mixed into 0.25 mL CSF).[10,19,20] The sample should be labeled with the protein source and amount added to the sample. One study demonstrated better preservation of mononuclear cells in canine samples when fetal calf serum was used instead of hetastarch.[18] All samples should be refrigerated at 4°C to minimize cellular degradation.

Cell Counts

A hemocytometer may be employed in practice to count nucleated cells and erythrocytes. Both sides of the cover-slipped hemocytometer are loaded with unstained CSF, which is then placed in a humidified container for 10 to 15 minutes to allow cells to settle on the glass. Because the fluid is unstained, the microscope condenser is lowered to improve contrast. Erythrocytes and nucleated cells are differentiated by size, refraction, granularity, and smoothness of plasma membrane.[21] Some laboratories stain CSF samples with new methylene blue (NMB), as leukocytes will take up stain, whereas erythrocytes remain unstained, making differentiation of leukocytes (specifically small lymphocytes) and erythrocytes easier (Fig. 14.1).[22] A small volume of CSF is drawn into a capillary tube coated with NMB or a tube that has a small volume of NMB followed by an air pocket.[22] The tube containing NMB and CSF is gently rocked back and forth, allowing the cells to take up some stain without diluting the CSF with a volume of NMB.[22] The hemocytometer is then loaded, and each population is counted and totals are calculated, as follows: Neubauer chamber: (1) both areas of large nine squares are counted, and the average of the number of leukocytes and erythrocytes is found; (2) the average is multiplied by 9 to get the cells per microliter (cells/μL).[10]

The ADVIA 120 (Siemens Medical Solution, Fernwald, Germany) hematology instrument has been validated for analyzing canine CSF samples and shows excellent correlation with manual methods used in dogs with increased total cell counts (pleocytosis), but the instrument may overestimate the cell count in samples without pleocytoses and has not been validated for the identification of eosinophils.[23] The automated differential count is also more accurate at higher cell numbers and thus should be compared with a traditional manual differential. The ADVIA 2120 hematology analyzer displayed satisfactory agreement with the standard hemocytometer method.[24] Validation experiments using 67 canine samples showed a sensitivity of 100% and specificity of 89% for accurately identifying samples with pleocytosis when manual counting was considered the gold standard (>5 cells/μL).[24] The instrument tended to be less accurate at lower (within reference interval) nucleated cell counts.[24] Erythrocytes may be a source of interference, as a red blood cell (RBC) count of 250 cells/μL was shown to elevate the nucleated cell count.[24] With regard to differential cell count, the instrument performed better in the presence of pleocytosis, whereas monocytes were overcounted at lower nucleated cell counts.[24] Automated cell counts thus should not replace a manual differential but may be used as another level of quality control. Automated instruments cannot recognize altered cell types, such as atypical neoplastic cells.

Measurement of Microprotein Concentration

Measurement of CSF specific gravity is not considered to be helpful because of low sensitivity for detecting abnormalities.[12] CSF microprotein may be semiquantitatively measured by using urine dipsticks that detect albumin. This assay has a lower detection limit of 100 mg/dL; therefore, it has low sensitivity for mild to moderate CSF protein concentration elevations (30 mg/dL to 100 mg/dL). False-positive or false-negative reactions may occur if the dipstick reads at trace or 1+, but this method is useful if other techniques are not available.[11] Reference laboratories apply a similar but more sensitive methodology to measurement of CSF microprotein as that of serum protein, using the trichloroacetic acid method, the Ponceau S red dye–binding method, or the Coomassie brilliant blue method.[22]

CSF globulin production is typically screened for with the Pandy reaction. In this test, a few drops of CSF are added to 1 mL of 10% carbolic acid solution, and the resulting turbidity is graded 0 to 4+. Any Pandy score above zero is considered elevated. Globulin concentration below 50 mg/dL will be undetectable with either test.[10,21]

Protein electrophoresis and immunoelectrophoresis may be performed on CSF and serum for maximum fractionation.[25] The utility of protein electrophoresis or immunoelectrophoresis of CSF lies in discriminating altered blood–brain barrier (BBB) permeability from increased localized production of immunoglobulin, which may be suggestive of (but not specific for) a disease entity for which an electrophoretic pattern has been established.

Cytological Slide Preparation

Cytological analysis is a critical component of CSF evaluation because the differential count (percentages) of cells may be abnormal, even if the total nucleated count is within reference interval. Cytology also enables examination for neoplastic cells, infectious agents, and evidence of prior hemorrhage. It may also serve as a quality control point, allowing for correlation between observed cellularity and the total count generated by a hemocytometer or an automated analyzer. Because of its low cellularity, CSF must be concentrated before cytological smear preparation.

Use of an in-house sedimentation chamber (Sörnäs procedure) may be very useful and preserves cell-free fluid for ancillary testing.[10] This technique will recover approximately 60% of total cells, which is sufficient for analysis.[16] A syringe barrel (with the tip and needle aseptically

removed with a scalpel blade) is turned upside down and the smooth, top side is placed in warm petroleum jelly and then onto a clean slide. Once a seal has formed, fresh CSF (at least 0.5 mL) is placed in the syringe and allowed to sit for 30 minutes.[16,21] Then, the supernatant is aspirated carefully with a pipette so as not to disturb the bottom layer contacting the slide. The syringe barrel is removed, and any excess CSF is carefully absorbed with a small piece of filter paper or paper towel. The slide is completely and rapidly air-dried without heat (inadequate drying results in cellular distortion), excess petroleum jelly removed with a scalpel blade, and the slide is stained with routine Romanowsky stains (e.g., Diff-Quik).

If CSF is sent to a reference laboratory, a cytological slide will likely be prepared using cytocentrifugation (500–1000 revolutions per minute [rpm] for 5–10 minutes, either onto a slide coated with albumin or with the addition of 0.05 mL of 30% albumin for improved cell capture) for maximal concentration of nucleated cells onto one slide.[16] Cytocentrifuged cytology may show excellent cellular detail, but the preparation may enlarge cells slightly and create an artifactual foamy or vacuolated appearance.[16] Slides are air-dried and stained with conventional Romanowsky stains. Multiple cytospin preparations may be made to yield 200 intact nucleated cells for classification.

Additional Cerebrospinal Fluid Testing

Culture

As it is rare for etiologic agents to localize only within the CNS, all cases of suspected infection may be aided diagnostically by fine-needle aspiration (FNA) cytology, biopsy with histopathology, culture of nonneural lesions, or all of these.[21] Bacterial culture and sensitivity testing of CSF is recommended for most cases of neutrophilic pleocytosis, given the appropriate clinical index of suspicion for a septic lesion. Even when organisms are visualized on CSF cytology, speciation and susceptibility testing may help guide prognostic and treatment decisions. Alternatively, bacterial or fungal culture may be negative regardless of cytological observation of organisms.[10,20] It must be remembered that bacterial CNS infection is highly uncommon in dogs and cats compared with other domestic animal species.[26]

Titers and Polymerase Chain Reaction Testing for Infectious Agents

Advanced techniques for neurological disease diagnosis are expanding rapidly. Enzyme-linked immunosorbent assay (ELISA)–based assays for antibody detection and PCR-based assays for nucleic acid detection of several medically important microbes have been developed for use on CSF and may be instructive in the diagnosis of viral, rickettsial, protozoal, or fungal diseases.[20] A large canine study that included a subset of 16 dogs with neoplastic or inflammatory disease showed that CSF titer provided diagnosis in 25% of cases.[3] Antibody assays should be interpreted cautiously because the presence of antibody may indicate prior exposure or vaccination rather than active infection. Moreover, compromise to the BBB in states of inflammation may translate to the presence of antibodies within the CSF without local production. Occasionally cross-reactive antibodies may be present that do not represent presence of the disease agent under assessment. Similarly, specimens for PCR should be submitted to a laboratory with strict quality control to minimize false-negative and false-positive results. Poor collection technique may result in false-positive results, especially for bacterial species that are ubiquitous in the environment.[27] As with other aspects of CSF analysis, a negative PCR result does not definitively rule out the presence of a pathogen because of the sampling limitation of a small portion of the extracellular space.[20]

Enzymes, Neurotransmitters, and Other Molecules

CSF contains glucose, electrolytes, neurotransmitters, and enzymes, but these substances are not measured routinely, although this measurement represents a rapidly expanding area of research in the effort to give clinicians better tools for diagnosing patients and determining prognoses. CSF enzymes originate from the bloodstream, the CNS, or cells within CSF.[10] One study of 34 cats with noninflammatory CNS disease showed that measurement of CSF activities of lactate dehydrogenase (LDH), aspartate aminotransferase (AST), and creatine kinase (CK) were not diagnostically sensitive but may be useful in detection of acute injury.[28] Multiple studies have correlated elevations in CSF CK activity with poor prognosis in dogs with neurological disease or spinal cord injury.[29,30] Immunoassays for vascular endothelial growth factor (VEGF) and S-100 calcium-binding protein have shown elevations of both molecules in the CSF of experimentally induced hypothyroid dogs, suggesting endothelial and glial contribution to increased BBB permeability in this population.[31] Myelin basic protein (MBP) has been found to be elevated in lumbar CSF in dogs with degenerative myelopathy, supporting the conclusion that it is a demyelinating lesion.[32] MBP concentration is elevated in the CSF of dogs affected by intervertebral disk herniation (IVDH) and has been found to be an independent predictor of poor prognosis.[33] Beta-2-microglobulin, a major histocompatibility complex I (MHC-I)–associated molecule, has been assayed by using ELISA and found to be elevated in the CSF of dogs with IVDH and inflammatory disease and also positively correlated with normal total nucleated cell count (TNCC).[34] The amino acids tryptophan and glutamine have been found to be elevated in the CSF of dogs with portosystemic shunts because of abnormal ammonia metabolism.[35] One study found increased oxytocin in the CSF of dogs with spinal cord compression, where it is believed to have an analgesic effect.[36] Gamma-aminobutyric acid (GABA) and glutamate neurotransmitter concentrations have been measured in dogs with epilepsy.[37]

NORMAL CEREBROSPINAL FLUID PARAMETERS

Gross Examination

Normal CSF is clear and colorless, with few cellular elements and a protein concentration approximately 200 to 300 times less than that of plasma or serum. Red or yellowish coloration indicates prior lesional hemorrhage or iatrogenic hemorrhage during collection. In the latter case, a pellet of RBCs will be present after centrifugation. True xanthochromia (yellowish color of hemoglobin breakdown products) that does not clear on centrifugation, cytological evidence of erythrophagia, or both indicate prior hemorrhage into the subarachnoid space.[20] Increased bilirubin leakage into the SAS or high concentrations of CSF protein (>100–150 mg/dL) may cause xanthrochromia.[21] Increased turbidity of the sample may be caused by increased number of cells present (>400 RBCs/μL or >200 nucleated cells/μL) but is usually not affected by mild changes.[10,11]

Cell Counts

TNCC is fewer than 5 cells/μL in the dog and fewer than 8 cells/μL in the cat, and elevation above this range is termed *pleocytosis*.[10] Grading of pleocytosis is somewhat subjective: In one reference, "mild" was defined as 6 to 50 cells/μL; "moderate" as 51 to 1000 cells/μL; and "marked" as more than 1000 cells/μL.[4]

Microprotein Concentration

Depending on laboratory-specific reference intervals, normal protein concentration is usually less than 25 to 30 mg/dL for cisternal CSF and less than 45 mg/dL for lumbar CSF.[10,20] Approximately 80% to 95% of CSF protein is albumin, and 5% to 12% of CSF total protein comprises gammaglobulins.[2] Eighty percent of CSF protein is transferred from plasma, with the remainder produced within the CNS. The latter

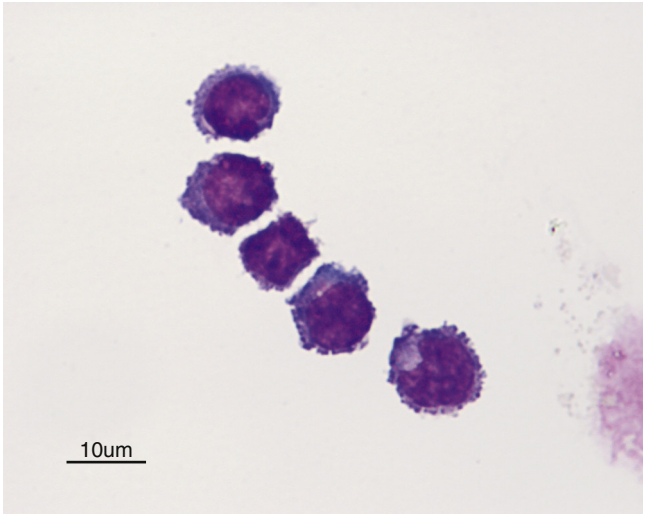

Fig. 14.2 Small lymphocytes in a cerebrospinal fluid sample (Wright-Giemsa stain).

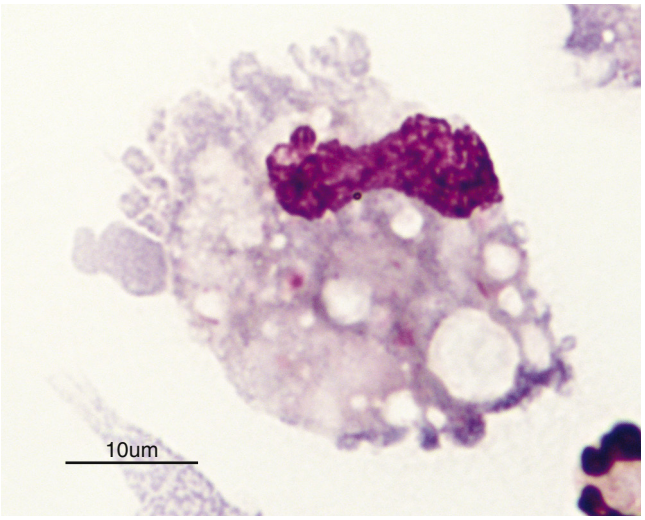

Fig. 14.4 Large mononuclear cell with cytoplasmic vacuolation in a cerebrospinal fluid sample (Wright-Giemsa stain).

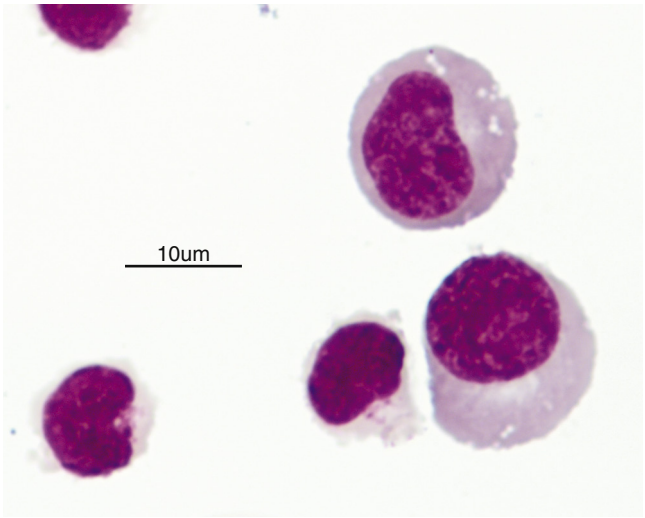

Fig. 14.3 Small lymphocytes in a cerebrospinal fluid sample. The two cells to the right have slightly increased amounts of cytoplasm (Wright-Giemsa stain).

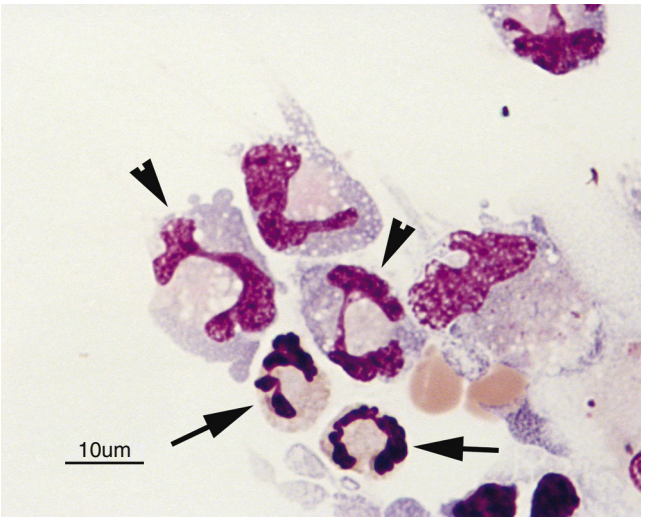

Fig. 14.5 Numerous large foamy mononuclear cells and two neutrophils *(arrows)* in a sample of cerebrospinal fluid. Large mononuclear cells may have nuclei that are similar in shape to band cells or neutrophils, only larger *(arrowheads)* (Wright-Giemsa stain).

population includes molecules also produced by other organs and proteins unique to the CSF that may potentially be used as markers of CNS tissue damage. Experimental evidence and earlier literature support a gradient of increasing protein concentration from cranial to caudal within the subarachnoid space, which has been attributed to slower flow and greater blood–CSF permeability caudally.[12]

Normal Cytology

Normal CSF is acellular or contains small numbers of small lymphocytes (Figs. 14.2 and 14.3) and large mononuclear cells (macrophages, ependymal lining cells, meningothelial lining cells, choroid plexus cells) (Figs. 14.4 and 14.5). Large mononuclear cells may be vacuolated and contain phagocytized material (Fig. 14.6). A low frequency of nondegenerate neutrophils (<25%), which are usually indicative of blood contamination during collection, may be present.[38] A study of 359 samples of canine CSF found a 7.5% incidence of meningeal, choroid plexus, ependymal, endothelial cells, or all of these.[39] No correlation existed between the presence of these cells and the presence of pleocytosis, elevated protein concentration, or the primary disease etiology.[39] Thus it is postulated that the presence of these cells is an artifact of collection and should not be overinterpreted. The authors recommended the term "surface epithelial cells" for the combined grouping (which cannot be distinguished cytologically), although not all of these cells (meningeal, endothelial) are of epithelial origin.[39] Occasionally, anucleate superficial squamous epithelial cells may be seen; these may be caused by contamination from the skin (Fig. 14.7).

Other Parameters

Occasionally, small amounts of granular, foamy extracellular material are present and are consistent with myelin or myelin-like material, which will stain positively with Luxol fast blue stain. This material may consist of myelin fragments, which are generated from demyelination, or may consist of myelin figures (a nonspecific term for layered phospholipids exfoliated from damaged cells).[40] The two cannot be distinguished with light microscopy. The significance of this material remains unclear because it may be observed in samples from patients with no discernible cause. A study of 98 canine cerebromedullary and

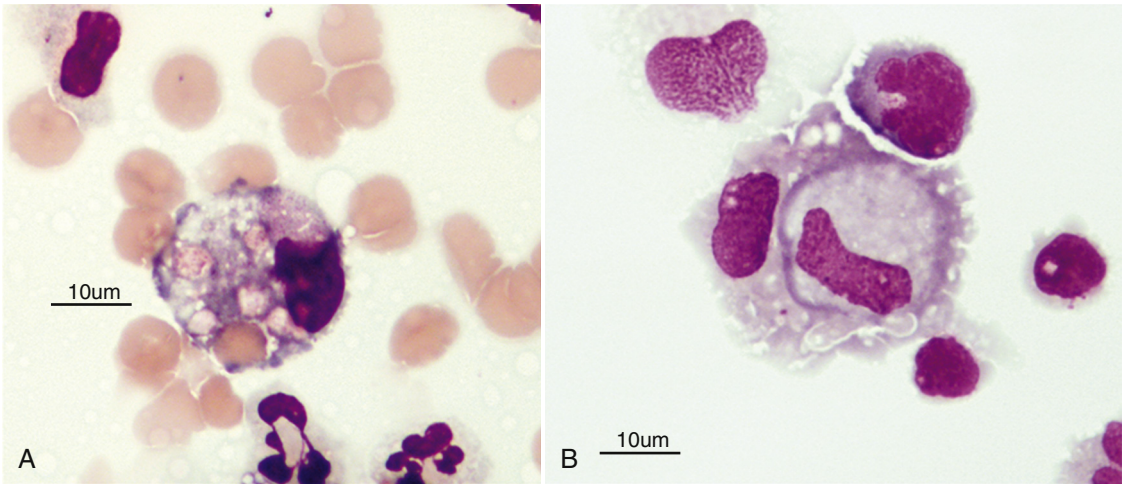

Fig. 14.6 Large mononuclear cells in cerebrospinal fluid showing evidence of (A) erythrophagia and (B) leukocytophagia (Wright-Giemsa stain).

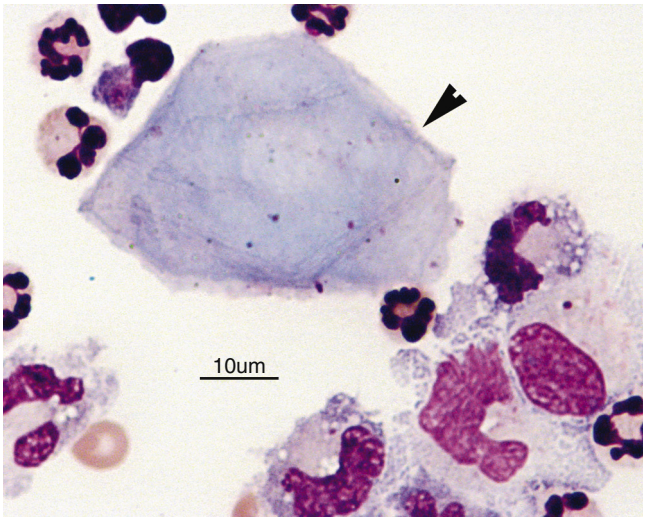

Fig. 14.7 Cerebrospinal fluid sample from a dog. A single large keratinized epithelial cell is present *(arrowhead)*. Squamous epithelial cells represent cutaneous contamination (Wright-Giemsa stain).

lumbar CSF samples showed 20% incidence of myelin-like material, with a higher percentage in samples from the lumbar cistern or from small dogs (<10 kg).[41] The presence of the material was not correlated with case outcome.[41] Similarly, in a study of 61 Cavalier King Charles Spaniels with Chiari-like malformations, myelinlike material was observed in 57% of lumbar CSF collections and 12% of cerebromedullary collections.[42] Thus myelin-like material may be a procedural artifact or may be consistent with a demyelinating (e.g., canine distemper virus, degenerative myelopathy) or potentially necrotizing disorder (e.g., IVDH, other spinal trauma, or a necrotic neoplasm).[40,41]

INTERPRETATION OF ABNORMAL CEREBROSPINAL FLUID

Blood Contamination and Hemorrhage

Normal CSF should not contain erythrocytes, but hemodilution is a common occurrence. Varying reports on the effect of blood contamination on TNCC, leukocyte differential, and protein concentration have been published.[43-46] Deciding whether increased TNCC or protein concentration is the result of hemodilution alone or a significant change concurrent with hemodilution necessarily remains, to an extent, a subjective assessment and must be critically evaluated in light of the magnitude of CSF findings along with the other pertinent facts of the case. Correction formulas for CSF parameters in the face of hemodilution (e.g., adding 1 nucleated cell/μL per 100 or 500 RBCs/μL) are unreliable.[45,46] In a recent study of 106 canine CSF samples without pleocytosis (TNCC <5/μL) but containing at least 500 RBCs/μL, the mean percentage of neutrophils (45.2% versus 5.7%), percentage of samples with eosinophils present (36.8% versus 6.8%), and mean protein concentration (40 mg/dL versus 26 mg/dL) were found to be significantly increased in the samples with blood contamination when compared with controls.[47] Significant RBC contamination warrants repeat sampling, if possible. Marked hemorrhage or evidence of prior hemorrhage (erythrophagocytosis, xanthochromia, hemosiderin-laden macrophages) may be useful in the diagnosis of CNS trauma, which may be accompanied by neutrophilic to mixed cell pleocytosis and mild increase in protein concentration.[4]

Elevated Microprotein Concentration

Elevated protein concentration in CSF (>30 mg/dL) may occur with or without pleocytosis, and in the absence of pleocytosis is termed *albuminocytological dissociation* (ACD). High protein concentration may be the result of leakage of plasma or cellular proteins across the BBB, localized production of immunoglobulin, localized tissue damage or necrosis, decreased clearance of protein into the venous sinuses, obstruction of CSF circulation, or all of the above. As such, it is a nonspecific change that indicates CNS damage or hyperproteinemic disease and is consistent with disease of any etiology (e.g., trauma, metabolic, infectious, inflammatory, degenerative, or neoplastic). Caution should be exercised when diagnosing ACD if the sample is hemodiluted (>500 RBCs/μL).[47] As is true for pleocytosis, inflammation of the meninges and superficial regions of parenchyma will result in greater CSF protein elevations than for lesions that are more remote from the SAS.

Alterations of Leukocyte Percentages Without a Pleocytosis

Occasionally, an abnormal leukocyte differential (shifted from mononuclear predominance to neutrophil predominance) without pleocytosis occurs. This may only be detected if cytological analysis (after sedimentation or cytocentrifugation of CSF) is performed. Increased percentages of neutrophils may occur in early or mild inflammatory

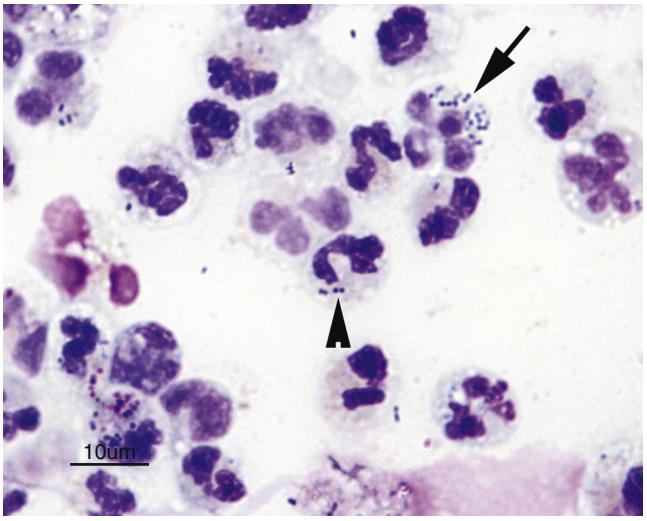

Fig. 14.8 Septic meningoencephalitis. Note the degenerate neutrophils and the presence of bacteria *(arrow)* (Wright-Giemsa stain).

disease, noninflammatory CNS disease, disease that is remote from the SAS or sampling site, or in cases of hemodilution. An increased proportion of neutrophils is present when neutrophils comprise greater than 25% of all nucleated cells, and increased percentage of eosinophils occurs when eosinophils comprise greater than 1% of the differential.[10]

Increased Neutrophil Percentage

When present (with or without increased TNCC), neutrophils should be evaluated for toxic change, degenerative change, and intracellular organisms or other inclusions (Fig. 14.8). Increased percentage of neutrophils without pleocytosis has been associated with healthy dogs, blood contamination, degenerative disk disease, neoplasia, cerebrovascular accident, fracture, CNS aspergillosis, and fibrocartilaginous embolism (FCE).[10,42,48] A study of 61 Cavalier King Charles Spaniels with Chiari-like malformation documented that those with syringomyelia were more likely to have an increased percentage of neutrophils, but it was not reported whether this subpopulation also had a concurrent pleocytosis.[42] In another study, cats with CNS neoplasia had increased percentage of neutrophils or lymphocytes without a pleocytosis.[28] Although not a classic pattern, infectious or inflammatory disease should not be ruled out if increased neutrophils are visualized without pleocytosis.

Increased Eosinophil Percentage

Increased percentage of eosinophils has been reported in parasitic and protozoal diseases, such as *Neospora caninum* infection.[38] One cat with eosinophilic meningoencephalitis (EME) of unknown etiology had an increased percentage of eosinophils and lymphocytes without pleocytosis.[49]

Increased Nucleated Cell Counts (Pleocytoses)

The specific diseases mentioned in the next section on various categories of pleocytosis are a survey of the current literature and meant to be a helpful starting point in the generation of particular differential diagnoses. Thus disease entities are listed in the section under which they are most commonly present, but it is important to note that for all disease entities, variability in the nature and the magnitude of pleocytosis may emerge in a particular patient at a particular point in time. Wherever possible, other categories of pleocytosis that have been reported for a disease have been mentioned. Generally, pleocytoses are defined by the cell type that comprises 70% or more of the nucleated cell population. If all cell types are 50% or less, the pleocytosis is classified as a mixed cell pleocytosis. And if, for example, lymphocytes are greater than 50% but less than 70%, some pathologists will classify the pleocytosis as mixed cell, lymphocyte predominant. A pleocytosis will be classified as eosinophilic if eosinophils compose at least 10% to 20% of the nucleated cell population.[11]

Neutrophilic Pleocytosis

Infectious conditions

Bacterial meningoencephalomyelitis. Bacterial infections of the CNS are unusual and represent a small portion of neutrophilic pleocytoses. Typically, this pleocytosis is severe (could be over 1000 cells/μL), neutrophilic, and accompanied by significantly elevated protein concentration, but the cell population may change to mononuclear during the course of treatment.[10,20,50] Rare instances of brain abscessation secondary to sepsis (which may be a sequela of iatrogenic immunosuppression) may result in marked neutrophilic pleocytosis, markedly elevated protein concentration, visualization of bacterial organisms (see Fig. 14.8), and abnormal MRI findings.[51] *Staphylococcus intermedius* was cultured from the CSF of a dog presenting with a retrobulbar abscess and neurological signs.[52] The CSF showed a moderate neutrophilic pleocytosis (75 cells/μL) and borderline elevation in protein concentration (30 mg/dL).[52] Local extension of severe otitis interna resulting in meningoencephalitis and ventriculitis in a dog has been reported.[53] This patient exhibited a severe neutrophilic pleocytosis (3672 cells/μL) and protein elevation (>400 mg/dL).[53] *Pasteurella multocida* meningoencephalomyelitis in a kitten was characterized by marked neutrophilic pleocytosis (981 cells/μL) with mild protein elevation (31 mg/dL) and rare extracellular and intracellular bacterial rods.[54] Bacterial culture and susceptibility testing are recommended but may yield false-negative results if organisms are not circulating in the extracellular space or if prior antibiotic therapy had been given. Serology and CSF-PCR (using organism-specific or universal bacterial [UB] PCR) are recommended.[27,54]

Cryptococcosis in dogs. *Cryptococcus* spp. are a large genus of systemic dimorphic fungi with a predilection for CNS tissue, which is infected hematogenously or via direct penetration of the cribriform plate. Only two species at this time are medically important: (1) *Cryptococcus neoformans* (var. *neoformans* and var. *grubii*) and (2) *Cryptococcus gattii*. In a recent study of 31 dogs with cryptococcosis, 68% had CNS infection, with neurological signs being the most common reason for presentation.[55] Dogs and cats with cryptococcosis typically have pleocytoses and elevated protein concentrations, but pleocytoses may be variably neutrophilic, eosinophilic, mononuclear, or mixed. In a recent study of 15 dogs with CNS cryptococcosis, organisms were found in 11 of 15 CSF samples (Figs. 14.9 and 14.10).[56] All affected dogs had pleocytoses that were mixed to mononuclear, whereas cats tended to have neutrophilic pleocytoses.[56] Of the samples, 11 of 12 also had increased protein concentrations (mean 494 mg/dL), which were significantly higher than in cats in the same study (mean 45 mg/dL).[56] Capsular antigen latex agglutination testing on serum or CSF is highly sensitive and specific and is recommended if cryptococcosis is suspected but organisms are not visualized cytologically.[57] This test may yield negative results if disease is present but localized (i.e., within the respiratory tract), so appropriate clinical signs should guide testing. Culture of CSF may also be helpful and may distinguish *C. neoformans* from *C. gattii* with the use of selective media. The finding of inflammatory foci on MRI may be supportive of the presence of fungal disease; cryptococcosis may result in mass lesions, meningitis, or pseudocyst formation.

Cryptococcosis in cats. Cryptococcosis is the most common systemic fungal disease of cats and is believed to infect the CNS less

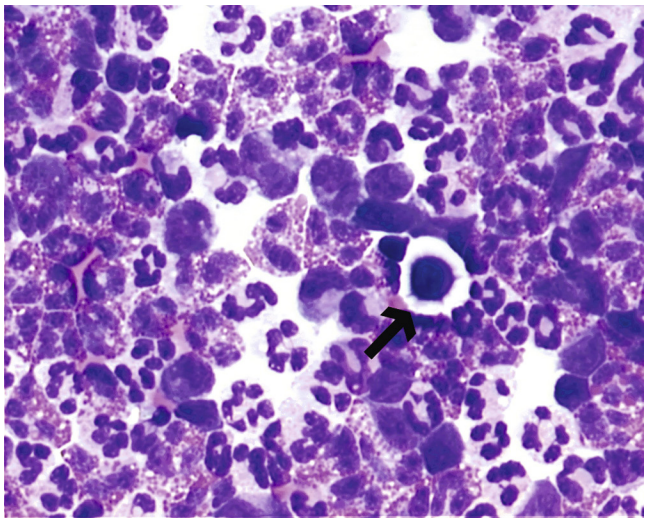

Fig. 14.9 Cryptococcosis. Note the presence of *Cryptococcus* spp. *(arrow)* and the presence of numerous eosinophils (modified Wright stain, original magnification 500×).

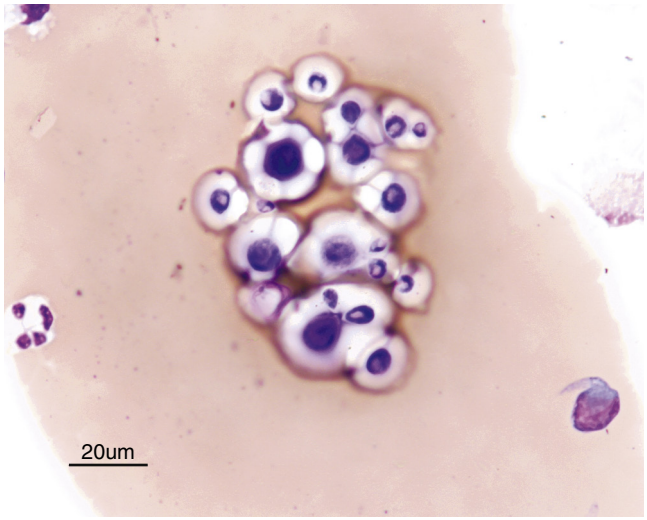

Fig. 14.10 Cryptococcosis. Numerous yeasts show a thick clear capsule. Narrow-based budding is also evident (Wright-Giemsa stain).

frequently than in the dog. A recent study found that 42% of 62 cats with cryptococcosis had CNS infection, but respiratory signs were still a more common reason for presentation.[55] Mild to marked neutrophilic or mononuclear pleocytosis may occur, with variable and occasionally normal protein concentrations.[4] A study of cats with CNS cryptococcosis showed organisms in 9 of 11 of the CSF samples, and a majority of cases (9 of 10) had neutrophilic pleocytosis and increased protein concentration (8 of 10).[56] Eosinophilic pleocytosis may also occur. Capsular antigen latex agglutination testing on serum or CSF is recommended for confirmation of *Cryptococcus* spp. infection, with rare false-negative reactions if disease is highly localized.

Histoplasmosis. *Histoplasma capsulatum* is a systemic dimorphic fungus that has been visualized in canine CSF and may be extracellular or within leukocytes.[58] A case report of an extradural *H. capsulatum* granuloma overlying spinal segments T11-L1 in a cat was associated with no cisternal CSF abnormalities.[59]

Aspergillosis. A study of dogs with systemic aspergillosis reported 4 of 8 CSF samples with neutrophilic pleocytosis (magnitude unspecified) and 1 of 8 with mononuclear reactivity.[60] Protein concentrations were not reported.[60] A more recent study focused on dogs with CNS aspergillosis, with 4 of 6 dogs demonstrating a neutrophilic pleocytosis (range: 20–1450 cells/μL) accompanied by protein elevation (38–1682 mg/dL).[48]

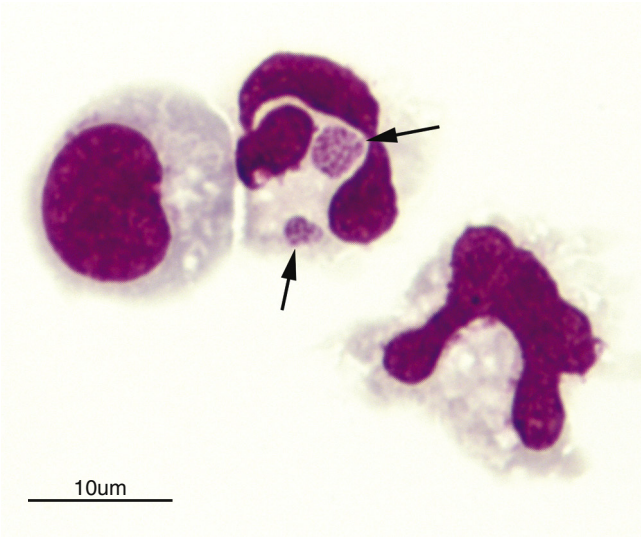

Fig. 14.11 Cerebrospinal fluid from a dog with ehrlichiosis. Two *Ehrlichia* morulae are evident in the central cell *(arrows)*.

Phaeohyphomycosis. Phaeohyphomycosis represents a group of darkly pigmented (typically brown, using routine stains) hyphal fungi, including neurotropic *Cladophialophora* spp. (formerly named *Cladosporidium* spp.) and *Xylohypha* spp. Acute infection may be characterized by mild to moderate neutrophilic pleocytosis and mild to moderately elevated protein concentration.[57]

Ehrlichiosis. Neutrophilic pleocytosis has been reported in cases of granulocytic *Ehrlichia* spp. in dogs (Fig. 14.11).[61] Neurological signs are uncommon in this disease, and affected dogs may display features ranging from ataxia to seizures.

Feline infectious peritonitis. Feline infectious peritonitis (FIP) has been traditionally linked to marked CSF changes, but the current literature paints a somewhat more varied picture. One study of natural FIP infection showed neutrophilic pleocytosis (as defined by >50% neutrophils) in the majority (7 of 11) of cases, with fewer cases of mononuclear (3 of 11; as defined by >80% mononuclear cells) and mixed cell (1 of 11) pleocytosis, all of variable severity.[4] Most cases (7 of 9) also had differing degrees of elevated protein concentrations.[4] Diagnosis was confirmed by histopathology or suggested by elevated feline coronavirus antibody titers and reduced albumin-to-globulin ratios in both serum and body cavity effusions.[4] A slightly older study of 16 CSF samples (natural and experimental infections) showed pleocytosis in 2 of 16 cases (neutrophilic and lymphocytic) and elevated protein concentration in 4 of 16 cases.[62] In a larger study of 67 cats with FIP or non-FIP disease, incidence of pleocytosis was highest in the neurological FIP group, but 20% of these patients did not have a pleocytosis.[63] Additionally, protein concentrations were variably elevated and not statistically different in FIP compared with non-FIP neurological disease.[63] Another study of 12 cats with CNS FIP showed 8 of 12 with unspecified pleocytosis and 3 of 12 with elevated protein concentration.[64] In cats with CNS disease, sensitivity of feline coronavirus (FeCoV) immunoglobulin G (IgG) in CSF for the diagnosis of FIP was 60%, and specificity was 93%, with a positive predictive value of 75% and a negative predictive value of 87% (FIP prevalence in this population was 25.6%).[63] Definitive diagnosis of this disease remains challenging, with virus

identification (PCR or immunohistochemistry) accompanied by pyogranulomatous inflammation in tissues being the gold standard. Hypergammaglobulinemia, elevated serum α_1-acid glycoprotein (AGP), MRI abnormalities (typically involving the ventricular lining and meninges), and positive feline coronavirus IgG titer or PCR from serum, tissue, or CSF are supportive but not specifically diagnostic, and negative findings do not rule out disease.[20,63,65]

Toxoplasmosis in cats. Cats are the definitive hosts for *Toxoplasma gondii* and may be subclinically infected; thus, diagnostics should only be performed on patients with appropriate clinical signs. Cats typically present with mild neutrophilic or mononuclear pleocytosis and normal to mildly elevated protein concentration, but marked protein elevation may occur.[4] Mild lymphocytic pleocytosis is also reported.[65] Diagnosis may be confirmed by direct visualization of organisms in CSF, aspirates of other inflammatory foci, histopathology of affected tissues, or fecal examination. Serology must be interpreted cautiously because IgG may remain elevated for up to 6 years after exposure. Therefore, paired serum IgM-IgG titers, indicating acute exposure, or documentation of rising serum IgG titers are more useful, but the latter is difficult to document in the advanced state of disease.[65,66]

Spinal epidural empyema in dogs. Epidural empyema is an uncommon disease in dogs, resulting from pyogenic infection in the epidural space. One study showed 4 of 5 dogs with neutrophilic pleocytosis of variable magnitude (11–342 cells/μL).[67] No organisms were visualized on any of the samples.[67] Except for one case with a lumbar CSF protein concentration of 726 mg/dL, protein elevations were modest.[67] Three CSF samples were cultured with no growth, and two dogs for which follow-up CSF was obtained showed resolution of pleocytosis.[67] These results are not surprising, as the dura likely provides a barrier to prevent infection extending from the epidural space to the subarachnoid space.

Other infections. A case of *Sarcocystis* spp. infection has been reported in a young cat with a marked neutrophilic pleocytosis with intracellular and extracellular merozoites observed on CSF cytology.[68] Diagnosis was confirmed with decreasing paired serologic titers, and speciation to the level of *Sarcocystis dasypi* or *Sarcocystis neurona* was conducted with PCR from blood.[68] A case of systemic *Acanthamoeba* spp. infection in a young Boxer, diagnosed post mortem, had antemortem CSF with marked neutrophilic pleocytosis (4956 cells/μL), marked increase in protein concentration (259 mg/dL), and subnormal CSF IgA concentration (33 mg/dL; reference interval 35–270 mg/dL).[69] Postmortem PCR for the organism was positive on extraneural tissue but not on CSF or spinal cord.[69] The patient had been deliberately immunosuppressed on the basis of a preponderance of evidence of steroid-responsive meningitis arteritis at initial presentation and thus may have been infected either before or opportunistically after treatment.[69] Another case report of canine cerebellar *Balamuthia mandrillaris* infection (diagnosed post mortem with immunohistochemistry) displayed a marked neutrophilic pleocytosis (234 cells/μL), but other cases with lymphocytic pleocytosis have been reported.[70] Because of tissue encystment, it is suggested that extraneural tissue be used for immunohistochemistry or PCR for antemortem confirmation of amoebic infection; PCR of CSF may be diagnostic but is not widely available.[69,70] Two dogs with aberrant spinal migration of *Spirocirca lupi* nematodes had moderate to marked neutrophilic to mixed or eosinophilic pleocytoses (800 cells/μL with 91% neutrophils; 180 cells/μL with 60% neutrophils, 30% eosinophils).[71]

Noninfectious conditions

Steroid-responsive meningitis arteritis. Steroid-responsive meningitis arteritis (SRMA) is presumptively an immune-mediated disease of mainly young, medium-and large- breed dogs: Beagles, Boxers, Bernese Mountain Dogs, Weimaraners, and Nova Scotia Duck Tolling Retrievers are overrepresented.[20] CSF analysis is important in diagnosis and typically features a moderate to marked neutrophilic pleocytosis (a left shift may be present) and markedly elevated protein concentration. Chronically, pleocytosis may change to a more mononuclear or mixed population (Fig. 14.12) and may become mild or even fall into reference intervals.[72] A study of 20 affected dogs showed neutrophilic pleocytosis in 12 of 20 cases and mononuclear pleocytosis in 8 of 20 cases.[72] Concurrent elevations of serum and CSF IgA titers (elevated IgG and IgM fractions may be present), serum concentration of cross-reactive protein (CRP), or serum α_2-macroglobulin is diagnostically supportive but not specific.[20,50] Increases in IgA have been linked to a T-helper 2 (Th2)–dominated immune response driven by elevated interleukin-4 (IL-4) and decreased IL-2 and interferon-gamma (IFN-γ).[73] Serum amyloid A (SAA), serum AGP, and serum haptoglobin may also be elevated.[74] Another study of 36 dogs with SRMA reported statistically significant elevations of CSF and serum CRP, but not serum α_2-macroglobulin, in dogs with SRMA compared with other neurological diseases.[75] In a study of 20 dogs, serum CRP was positively correlated with CSF TNCC.[72] Additionally, serum haptoglobin and serum and CSF IgA remained increased throughout successful treatment, indicating that these parameters are more useful for diagnosis than for monitoring therapy.[72] Serum and CSF concentrations of CRP and SAA have been documented to fall significantly during treatment, and repeat measurement of serum CRP or SAA may be used to guide therapy and predict relapse, which is less invasive and more sensitive than repeat CSF sampling.[72,74,75] Rare cases have been documented in cats with marked mononuclear or mixed pleocytosis and mild to moderate protein concentration elevations.[4]

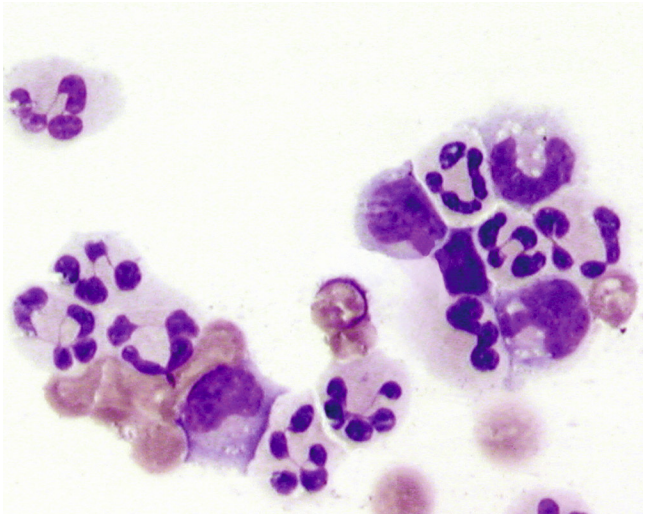

Fig. 14.12 Steroid-responsive meningitis in a Bernese Mountain Dog. Mixed inflammation with nondegenerate neutrophils and large mononuclear cells (modified Wright stain, original magnification 500×).

Intervertebral disk herniation. CSF from patients with IVDH may be extremely variable; data indicate that CSF findings correlate with location of sampling, disk herniation location, chronicity of the lesion, and severity of spinal cord injury. Bearing this in mind, it is no surprise that some reports in the literature state that neutrophilic, lymphocytic, mixed, and mononuclear pleocytoses are most common in dogs with IVDH.[12,30,76] A study of 423 cases of IVDH showed 51% with pleocytosis, of which 31% were neutrophilic, 41% were lymphocytic, 20% were mixed, and 7.4% were mononuclear.[76] Of all cases, 71% had elevated protein concentrations.[76] Interestingly, a

larger number of cases of lymphocytic pleocytosis were observed in the samples analyzed more than 7 days after onset of clinical signs.[76] The magnitude of pleocytosis, in general, was also shown to decrease with increasing time between clinical onset and sampling, and this observation has been corroborated by other studies.[12,76] Prior treatment with corticosteroids was observed to reduce the number of observed lymphocytes in CSF.[76] The authors also found a higher incidence of pleocytosis in thoracolumbar disease (61%) compared with cervical disease (23%), but this may have been caused by exclusive sampling of lumbar CSF closer to the lesion.[76] IVDH is rare in cats and has been reported to feature mild mixed cell pleocytosis and elevated protein concentration.[4]

Ischemic myelopathy caused by fibrocartilaginous embolism. Patients typically present with nonpainful, progressive, asymmetrical neurological signs. As only histopathology is confirmatory, it is a multimodal diagnosis of exclusion. A study of 32 dogs with presumptive FCE, based on history, clinical signs, imaging, and outcome, showed 53% with normal CSF, 25% with ACD, and 19% with mild to moderate pleocytosis (7–84 cells/μL; median 12/μL).[77] Pleocytoses were neutrophilic or mixed.[77] One study of 36 confirmed cases in dogs showed that 64% had normal CSF and the remainder displayed mild changes.[78] Another study looking at five dogs suggested that pleocytosis may be marked, up to 529 cells/μL.[3]

FCE is much less common in cats. In general, the disease process and clinical signs are similar to those in dogs, with the exception that the disease presents in cats in middle or older age, usually with cervical spinal cord signs. A case series of five cats showed CSF ranging from normal to marked neutrophilic pleocytosis with moderately elevated protein concentration and variable correlation to clinical outcome.[79] The case with the most severe CSF changes had extensive myelomalacia at necropsy.[79] It was suggested in this study that CSF is more likely to be abnormal if collected closer to the lesion and that MRI is helpful for localization and in supporting the diagnosis.[77,79]

Thiamine deficiency in cats. Thiamine deficiency is a rare nutritional disorder of patients fed noncommercial, misformulated commercial, or irradiated diets. Two case reports showed increased percentage of neutrophils or mild neutrophilic pleocytosis, presumptively from cerebrocortical necrosis.[4] Diagnosis is based on history, response to treatment, MRI features compatible with the disease (cortical and brainstem hyperintensities), or histopathology.[80]

Chiari-like malformation. A study of 61 Cavalier King Charles Spaniels with Chiari-like malformation showed that 40% of dogs with concurrent syringomyelia and cisternal CSF sampling had mild (up to 15 cells/μL) pleocytoses and increased percentages of neutrophils compared with the subpopulation without syringomyelia, but it was not specifically documented whether pleocytoses were, in fact, neutrophilic or mixed with an increased percentage of neutrophils.[42] A positive correlation was also seen to exist between TNCC and syrinx size.[42]

Neoplasia. It is important to perform CSF in neurology patients with suspected neoplasia, as definitive diagnosis may be achieved if neoplastic cells are directly observed via cytology. Inflammatory pleocytoses or elevated protein concentrations are common in patients with cancer, tend to be mild to moderate in magnitude, and may represent paraneoplastic inflammation, compromise of the BBB, lesional necrosis, or all of these.[28] Normal CSF is also a common finding in cases of neoplasia. Moreover, in the absence of overtly neoplastic cells, no defined patterns connect specific tumors with specific types of inflammatory pleocytoses. Neutrophilic pleocytosis of unspecified magnitude was found in the CSF of 2 of 11 cats with spinal lymphoma and in 3 of 7 cats with nonlymphoma spinal neoplasia (astrocytoma or osteosarcoma).[81] Additionally, the remaining four cats with nonlymphoma spinal tumors (meningioma, peripheral nerve sheath tumor, plasma cell tumor) had either normal CSF or ACD of unspecified magnitude.[81] Metastatic tumors to the CNS should also be considered in a patient with neurological signs.

Eosinophilic Pleocytosis

Eosinophilic meningoencephalitis of dogs. EME is an idiopathic diagnosis of exclusion that is typically steroid responsive and is postulated to be triggered by an underlying hypersensitivity, allergy, or self-limiting infection. The disease may be overrepresented in Rottweilers and Golden Retrievers.[82] A study of 23 dogs with eosinophilic pleocytosis (defined by >20% eosinophils) showed 16 cases of idiopathic EME, 4 cases of infectious disease (*C. neoformans*, *N. caninum*, *Baylisascaris procyonis*), and 3 cases of IVDH.[83] The magnitude of pleocytosis or the percentage of eosinophils could not be used to distinguish infectious versus EME cases, although IVDH cases tended to have milder pleocytoses (<84 cells/μL).[83] In about half the EME cases, MRI showed abnormal findings.[83] Peripheral eosinophilia may or may not be present.

Infectious and other conditions. Eosinophilic pleocytosis is highly suggestive of protozoal (toxoplasmosis, neosporosis), fungal (cryptococcosis), parasitic (including *Cuterebra* spp., dirofilariasis), and algal (protothecosis) infections and also rarely in cases of canine distemper and rabies viruses.[10,84] Eosinophils have also been found in cases of granulomatous meningoencephalomyelitis (GME).[85] Eosinophilic pleocytosis has been documented in bacterial encephalitis as well.[21]

Lymphocytic Pleocytosis

Infectious conditions

Toxoplasmosis in dogs. Dogs with clinical indications for *T. gondii* infection tend to have neurological or neuromuscular signs. Case reports are sporadic; documentation of mild ACD (58 mg/dL) and also a report of mild lymphocytic or eosinophilic pleocytosis (35 cells/μL) with an elevated protein concentration of 77 mg/dL exist in the literature.[86] It is important to rule out other potential causes of the neurological signs because immunocompetent dogs tend to clear subclinical infections, and therefore paired serum IgM-IgG titers or sequential serum IgG titers are preferable to a single serum IgG titer. To the author's knowledge, no data on the life span of canine IgG antibodies exist. Reports in the literature are conflicting with regard to the cross-reactivity of *T. gondii* antibodies to other agents, such as *N. caninum*.[20,86] PCR testing for *Toxoplasma* in serum, tissue, or CSF is diagnostic.[86,87]

Rabies. Pleocytoses may be lymphocytic and of varying severity. Ancillary antemortem diagnostics include viral PCR on saliva or CSF and the saliva antigen latex agglutination test. In a study of 15 dogs under quarantine for suspected natural infection (subsequently confirmed positive), 13 of 15 were saliva-PCR positive, and 4 of 15 (27%) were CSF-PCR positive.[88] All animals with positive results on CSF were also positive on saliva, and interestingly 100% correlation was seen between positive CSF-PCR and the dull clinical presentation (all aggressive clinical presentations were CSF-PCR negative).[88] Negative testing should never exclude diagnosis because viral load is highest within salivary glands and brain parenchyma.[88]

Acute canine distemper virus infection. Antemortem diagnosis of canine distemper virus (CDV) infection is difficult and is frequently made by exclusion when coupled with appropriate clinical signs. CSF and MRI findings are variable and may be normal in the acute stage of disease before inflammation has peaked.[89] In a study of 32 dogs with noninflammatory distemper, half (15 of 32) had normal

CSF.[85] A study of eight dogs with natural infection (confirmed by CNS tissue–PCR and histopathology) showed lymphocytic pleocytosis in all samples and normal protein concentrations.[90] Another case (confirmed by tissue–PCR and CSF-PCR) in a 7-month-old dog displayed marked (554 cells/μL) lymphocytic pleocytosis and a normal protein concentration.[89] Because this is a demyelinating disease, myelin-like material, which is amorphous, granular, pink, foamy, and stains positively with Luxol fast blue, may be present.[40] PCR testing of CSF, serum, urine, epithelial or tonsillar tissue is available, and immunohistochemistry on biopsy specimens of nasal mucosa, haired skin, or footpad is 88% to 96% sensitive for detection of viral antigen.[91]

Chronic canine distemper virus infection. If a pleocytosis is present, it is likely to be lymphocytic. Pleocytoses are typically mild to moderate, but severe lymphocytic pleocytoses have been reported.[89] Main differential diagnoses include other viral diseases, GME, or chronic bacterial infection. Extranigral signs related to the gastrointestinal or the respiratory system, if present, may be helpful in distinguishing this disease from GME.[89] In a study comparing four dogs with chronic CDV, six dogs with acute CDV, and controls, dogs with chronic CDV had markedly elevated CSF IgG concentration.[92] The IgG region was polyclonal, including a population of neutralizing antibodies for CDV.[92]

Coccidioidomycosis. *Coccidioides immitis* is a dimorphic fungus acquired through inhalation, and most cases in the United States are observed in the southwestern region of the country. Signs tend to involve respiratory or skeletal systems, and CNS involvement is rare. One dog had a mild to moderate lymphocytic pleocytosis.[57] Complement fixation (detecting IgG) or tube precipitation (detecting IgM), or agar-gel immunodiffusion serological testing is recommended for confirmation.

Noninfectious conditions

Necrotizing meningoencephalitis. Meningoencephalitis has been subcategorized as necrotizing meningoencephalitis (NME) and necrotizing leukoencephalitis (NLE) on the basis of histopathological appearance. Both NLE and NME are believed to have an immune-mediated basis, and recent data support that in Pugs with NME, canine leukocyte antigen gene aberrations exist.[93] Meningoencephalitis is rapidly progressive and affects a variety of generally young to middle-aged toy-breed dogs, including the Pug, Shih Tzu, Papillon, Maltese, Chihuahua, Yorkshire terrier, French bulldog, Pekingese, West Highland White terrier, Boston terrier, Japanese Spitz, and Miniature Pinscher breeds.[20,94] A study of CSF from 14 Pugs with NME showed 12 of 14 with pleocytoses of varying severity (mean 120 cells/μL).[95] Of these dogs, 66% had a lymphocytic pleocytosis, 17% had a mononuclear pleocytosis, and 17% had a mixed cell pleocytosis (Fig. 14.13)[95]; 11 of 14 dogs had elevated protein concentrations (mean 88.4 mg/dL).[95] Another study of three dogs showed one with ACD and two with moderate to marked (40–220 cells/μL) neutrophilic to lymphocytic pleocytosis.[94] MRI findings may help support a diagnosis, but only histopathological examination of lesions provides definitive proof.

Other. Four cats with ischemic encephalopathy had mild (<10 cells/μL) lymphocytic pleocytosis.[28] Another study of feline ischemic encephalopathy reported one cat with normal CSF and another with mononuclear to mixed pleocytosis (26 cells/μL).[96] A report of two cats with cerebrovascular disease (infarction or stroke) showed one with mononuclear pleocytosis and the other with ACD.[97] Cerebrovascular disease was correlated in several other cases (without CSF data) to hepatic lipidosis or FIP.[97] A prospective study of CSF from 17 Pembroke Welsh Corgis with familial degenerative myelopathy showed normal CSF in 15 samples and hemodilution in 2 samples.[98] Various degenerative myelopathies have been described in German Shepherds, Afghan Hounds, Rottweilers, Jack Russell Terriers, and Smooth-Haired Fox Terriers and have been associated with mild CSF changes.[11]

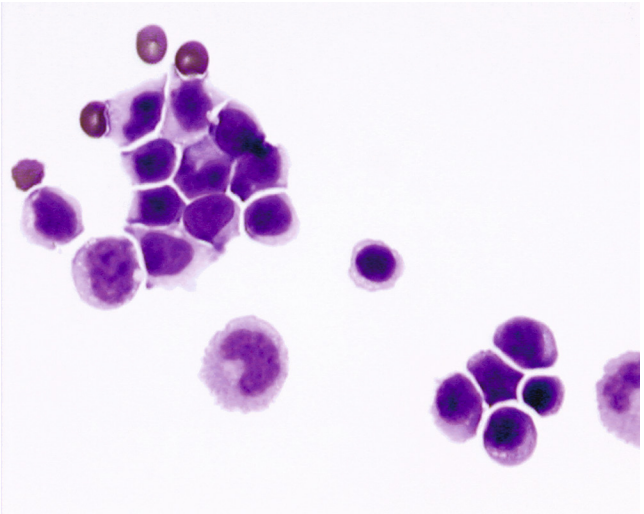

Fig. 14.13 Necrotizing meningoencephalitis in a Pug. Mixed lymphocytic and large mononuclear inflammation (modified Wright stain, original magnification 500×).

Mononuclear Pleocytosis

Infectious conditions

Neosporosis. Dogs are the definitive host of *N. caninum*. Most clinical cases are young animals that present with CNS (especially cerebellar) signs, neuromuscular signs, or both. Two recent case reports have described protozoal tachyzoites directly visualized in the CSF intracellularly and extracellularly.[99,100] Both patients had been previously treated with glucocorticoids.[99,100] One case had a marked mixed to mononuclear pleocytosis (1450 cells/μL) with marked protein elevation (992 mg/dL), and the other case had a marked eosinophilic pleocytosis (298 cells/μL) with a markedly elevated protein concentration (392 mg/dL) protein at the time of diagnosis.[99,100] Diagnosis was confirmed in both cases by postmortem immunohistochemistry of tissue.[99,100] Another study showed six of seven naturally infected dogs demonstrating a mild to marked mononuclear pleocytosis (12–300 cells/μL) with moderately to markedly elevated protein concentrations observed in four of six dogs (60–290 mg/dL).[101] A majority of these cases displayed cerebellar neurologic signs.[101] Antemortem immunohistochemistry of CNS or muscular tissue is diagnostic but may not be practical. Increased serum liver or muscle enzymes and electromyography (EMG) may be helpful if concordant signs are present.[99] A variety of serological techniques to assay for the presence of antibody are available, and a titer greater than 1:64 is supportive of diagnosis. Antibody detection in serum or CSF, coupled with PCR of CSF or other potentially affected tissues, is currently recommended for the definitive diagnosis of CNS neosporosis.[20]

Other. A case report of a Pug with a mild mononuclear pleocytosis (8 cells/μL), mild elevation in protein concentration (89 mg/dL), evidence of hemorrhage, and direct visualization of *Angiostrongylus vasorum* helminth larvae is found in the literature.[102] Eosinophilia was not observed within the CSF or peripheral blood.[102] The organism is endemic in Europe and Canada among foxes and canids, mainly causing respiratory signs or coagulopathy; neurological signs are typically caused by hemorrhage.[102] Two dogs with paraparesis and pyogranulomatous lumbar masses (one intradural, one extradural) had lumbar CSF with

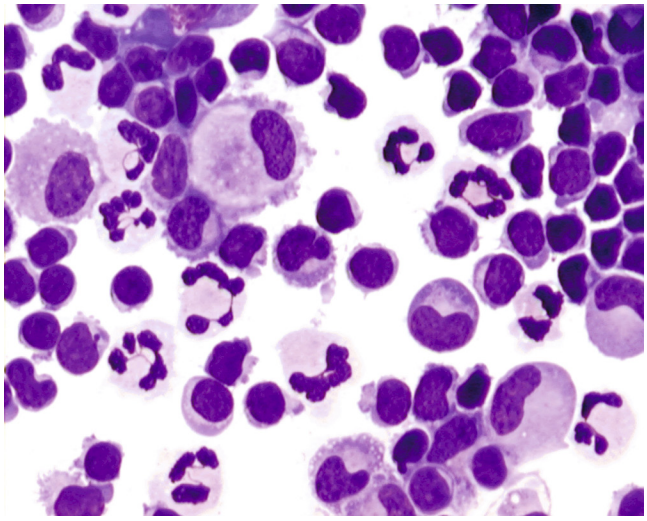

Fig. 14.14 Granulomatous meningoencephalitis. Mixed mononuclear and neutrophilic inflammation (modified Wright stain, original magnification 500×).

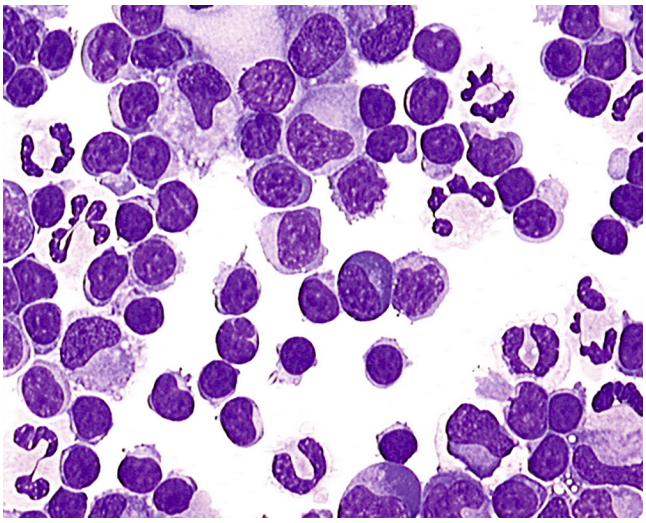

Fig. 14.15 Granulomatous meningoencephalitis. Mixed mononuclear and neutrophilic inflammation. Note the presence of a plasma cell in the center (modified Wright stain, original magnification 500×).

mild mixed cell pleocytosis (lymphocytes and nondegenerate neutrophils) or lymphocytic pleocytosis.[103] These patients were serologically PCR positive for *Bartonella vinsonii* subsp. *berkhoffi* and presented with a nodular dermatosis.[103] A dog with neurological signs and *Hepatozoon canis* infection showed marked lymphocytic pleocytosis (243 cell/μL) with mildly elevated protein concentration (37 mg/dL).[104] Organisms were not visualized in CSF but were found on cytology of peripheral blood, lymph node, bone marrow, and bony lesions. Serology and positive PCR from a bone marrow sample were diagnostic.[104]

Noninfectious conditions

Intrathecal contrast administration. Contrast media or pharmacological agents, such as epidural anesthetics, may introduce preanalytical error into CSF samples, artificially raising TNCC and protein concentrations.[105] In a study of 17 healthy dogs given either iopamidol or metrizamide for EMG, same-day CSF sampling showed that 8 of 17 developed a mild to moderate mononuclear to mixed mononuclear or neutrophilic pleocytosis (6 of 8 were from iopamidol).[106] In the same study, 3 of 17 (all metrizamide) developed mild protein elevation, but mean protein concentration for both groups stayed within the reference interval.[106] In dogs given metrizamide, 7 of 8 had an increased Pandy score after EMG, which was considered a false-positive result because of the contrast agent.[106] These data, plus histopathology from the same population, showed that the contrast agents caused low-grade leptomeningeal inflammation with no statistical difference between the two agents studied.[106] Another similar study over 30 days showed that post-EMG CSF changes reversed after approximately 5 days.[107]

Granulomatous meningoencephalitis. GME is a progressive immune-mediated disease that is overrepresented in females, Toy-breed dogs, and Terriers.[20] It is a diagnosis of exclusion and has clinical presentations and MRI findings that may be similar to various infectious and neoplastic diseases. CSF may be unaffected or may display a mononuclear to mixed pleocytosis and protein concentration elevations, both of varying severity (Figs. 14.14 and 14.15). In a study of 188 CSF samples from dogs with inflammatory neurological diseases, marked pleocytosis (>1000 cells/μL) was found in cases of SRMA, bacterial encephalitis, or GME.[85] Pleocytosis may also be lymphocytic or neutrophilic.[10] CSF protein electrophoresis may be helpful, as several cases have been shown with increased β-globulin and gammaglobulin fractions.[108]

Mixed Cell Pleocytosis

Most of the diseases described previously in this chapter may manifest as mixed cell pleocytoses, depending on the time interval between disease onset and CSF sampling, disease severity, and previous treatment administered. A mixed cell pleocytosis would be expected to occur during transition between different phases of the inflammatory response, where certain cells may predominate at specific times after injury.

Blastomycosis. Infection of the CNS by *Blastomyces dermatiditis* is typically rare and may involve chorioretinitis or focal cerebral granuloma in the cat.[109] A study of two dogs with systemic blastomycosis and neurologic signs showed mild mixed cell pleocytosis (8 cells/μL, mononuclear predominant; and 15 cells/μL, lymphocytic predominant).[110] Using CSF cytology or culture to diagnose the organism may be unrewarding. Agar-gel immunodiffusion serologic testing has high sensitivity and specificity for canine antibodies and is recommended if appropriate clinical signs (respiratory signs or lymphadenopathy) are present.[57] Agar-gel immunodiffusion testing is less sensitive (25%–33%) in the cat, as indicated by a limited number of reports.[57] Urine antigen enzyme immunoassay (EIA) has good sensitivity for dogs and has been used successfully on at least one cat.[109] EIA may also be performed on CSF. Cytology of nasal, pulmonary, or dermal lesions is more likely to yield direct visualization of organisms.

Neoplasia

Lymphoma. Lymphocytic pleocytosis of inflammatory origin may be difficult to distinguish from lymphoma exfoliating into the CSF (Fig. 14.16). The size of lymphocytes and morphological atypia may be helpful, although these may be challenging to differentiate from artifactual morphological changes secondary to cytospin preparation. Cats with neoplasia may have lymphocytic pleocytoses (suggestive of lymphoma), mild to moderate mononuclear to mixed cell pleocytoses (suggestive of nonlymphoma tumors), or normal CSF. One study examined six cases of feline CNS or multifocal lymphoma, which displayed pleocytoses of variable magnitude, absent to mildly elevated protein concentrations, and neoplastic cells visualized in 5 of 6 of the CSF samples.[4] In this study, eight cats with CNS signs that were ultimately diagnosed with nonlymphoma tumors (e.g., meningioma, carcinoma, nerve sheath tumor) had mild CSF protein elevations and either normal TNCC (1 of 8) or mild to moderate mononuclear or mixed cell

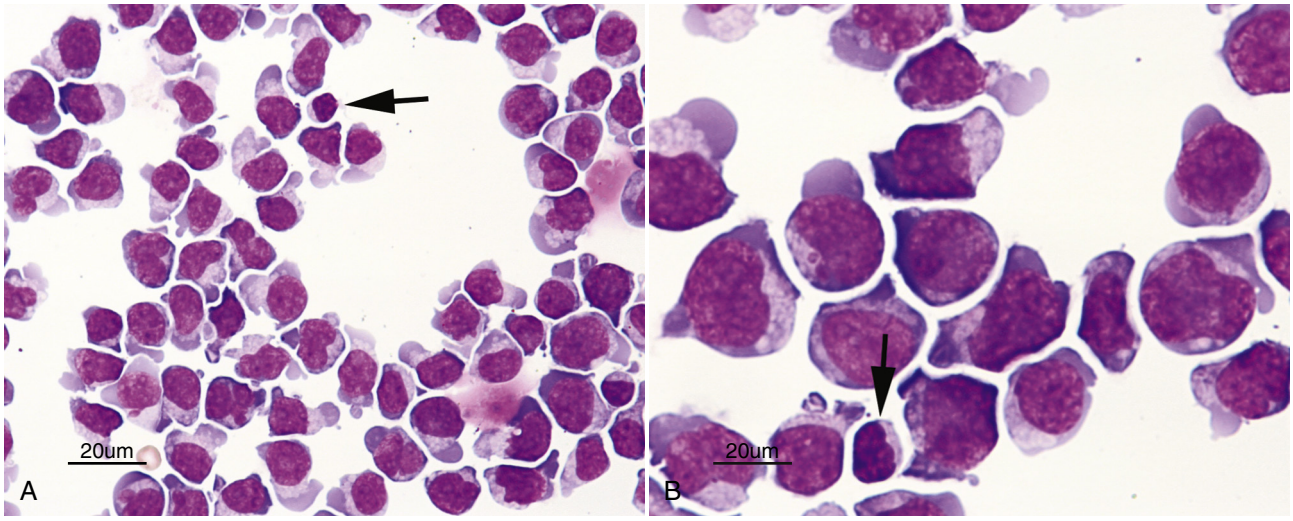

Fig. 14.16 Cerebrospinal fluid samples from a dog with central nervous system lymphoma. (A) Low magnification shows a cellular slide consisting almost entirely of large, pleomorphic lymphoid cells. A mature lymphocyte with condensed, mature chromatin is present *(arrow)*. (B) Higher magnification of the same slide. Lymphoid cells are large with pleomorphic nuclei and immature chromatin. Some cells have distinct nucleoli. A mature lymphocyte with condensed chromatin is present *(arrow)* (Wright-Giemsa stain).

pleocytosis (7 of 8).[4] Another study of 11 cats with spinal lymphoma showed neoplastic cells visualized in one case and hemodilution, ACD, or neutrophilic pleocytosis in the remainder of cases.[81] A case report of feline multiple myeloma involving lumbar vertebrae and associated soft tissues exhibited cisternal CSF with an elevated protein concentration of 290 mg/dL and mild pleocytosis (8 cells/μL) consisting of a majority of neoplastic plasma cells.[111] Diagnosis was further confirmed by abnormal urine protein electrophoresis and bone marrow aspiration.[111]

Histiocytic malignancies. Malignant histiocytosis or histiocytic sarcoma tumor cells in canine CSF have been documented in two recent case reports; CSF cytology displayed marked mononuclear pleocytoses (>500 cells/μL) and mild to moderately elevated protein concentrations (<135 mg/dL).[112,113] Tumor cells phenotypically resembled macrophages, displayed multiple criteria of malignancy, and reacted positively to CD1c on immunocytochemistry, compatible with interstitial dendritic cell origin.[112,113] Necropsy was confirmatory and found no evidence of neoplasia outside of the CNS.[112,113] A case report of a gliomatosis cerebri (GC) neoplasm in a middle-aged poodle showed CSF with a mild lymphocytic pleocytosis (20 cells/μL) and protein concentration elevation.[114] On histopathology, lymphocyte-like perivascular cuffing and meningitis were noted. Other case studies of canine GC have reported normal CSF or mild ACD.[115]

Meningioma. In a study of 56 dogs with intracranial meningioma, in which CSF analysis was performed, 29% had normal CSF, 45% had ACD, and 27% had pleocytosis (2 of 3 of these neutrophilic pleocytosis; 1 of 3 unspecified), with the overall incidence of neutrophilic pleocytosis at 18%.[116] In this study, a positive correlation existed between elevated TNCC and anatomical localization of the lesion to the caudal (versus middle or rostral) portion of the cranial fossa, and no association between pleocytosis and necrosis within the lesion was found.[116] These findings contradict prior reports of a high percentage of abnormal CSF findings in meningioma, and the authors reported that concurrent glucocorticoid therapy in some of the patients may have negatively biased the data.[11,116] A study of 26 dogs with spinal meningioma showed no cases with exfoliating tumor cells, 62% with mild pleocytosis up to 47 cells/μL (mean 11 cells/μL), and normal or variably elevated protein concentrations up to 836 mg/dL (mean 212 mg/dL).[117] Both cisternal and lumbar CSF samples were evaluated in this study and not found to be significantly different.[117] Interestingly, tumors of the lumbar region displayed higher mean TNCC and protein concentrations compared with tumors of the cervical area (24 versus 4 cells/μL and 158 versus 98 mg/dL, respectively), which the authors postulated may be reflective of a higher number of lumbar CSF samples with proximity to the lesion.[117]

Other neoplasms. A case report of canine CSF with 240 cells/μL was characterized by atypical neoplastic round cells that were confirmed on immunocytochemistry and immunohistochemistry to be from a metastatic mammary carcinoma.[118] Inflammatory cells were of low numbers and were of a mixed population.[118] A study of CSF from 25 dogs with choroid plexus tumors showed direct observation of tumor cells in 47% of the cases of carcinoma.[119] Mild to moderate mixed-cell pleocytosis was present in all cases of papilloma and in half of the carcinomas; when pleocytosis was present, no difference in magnitude existed between benign and malignant tumors.[119] All cases had elevated protein concentrations, with median concentration for carcinoma being significantly higher (108 mg/dL) than median concentration for papilloma (34 mg/dL).[119] A cutoff protein concentration of 80 mg/dL yielded a sensitivity of 67% and a specificity of 100% for detection of choroid plexus carcinomas.[119] Another case report of canine choroid plexus carcinoma had a mononuclear pleocytosis of 165 cells/μL, mildly elevated protein concentration of 30 mg/dL, and numerous tumor cells visualized.[120]

CENTRAL NERVOUS SYSTEM CYTOLOGICAL EVALUATION

A rise in the availability of stereotactic brain biopsy has facilitated increased cytological assessments of CNS lesions. This technique offers several advantages, although significant equipment investment and time to perfect techniques is required. Stereotactic biopsy often offers application accuracy for targeting lesions that approximate 3 mm or less in all directions. In one study, diagnostic accuracy of stereotactic biopsy specimens submitted for histopathology (i.e., agreement with specimens obtained via open approaches) exceeded 90%.[121] In experienced hands, stereotactic biopsy is believed to be a relatively low-morbidity procedure.

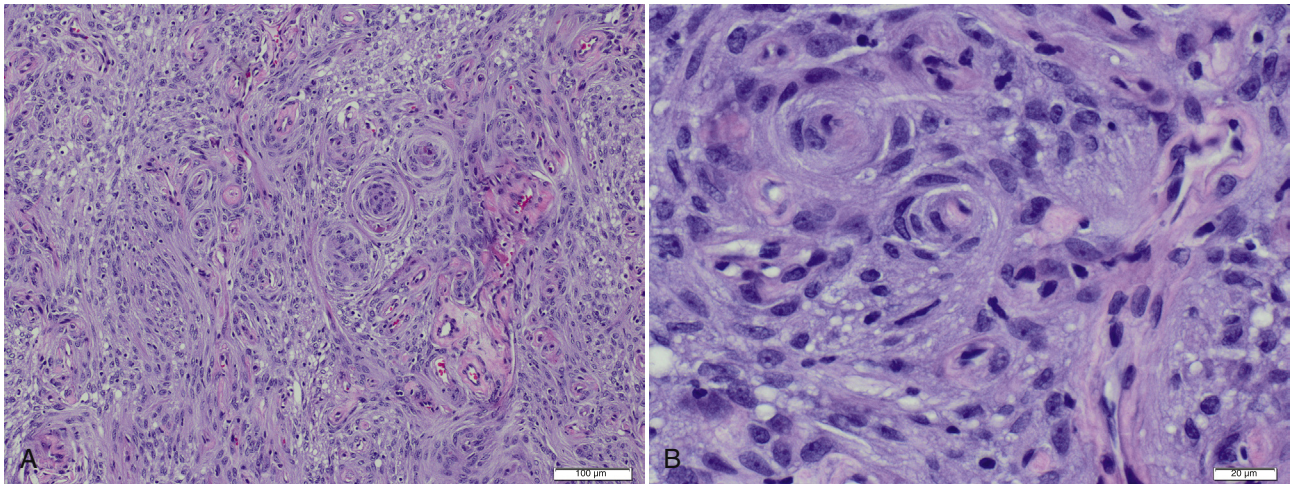

Fig. 14.17 Meningioma. (A) Low magnification shows spindle-shaped cells arranged in whorls and interweaving bundles (hematoxylin and eosin [H&E] stain, original magnification 100×). (B) High magnification of spindle-shaped cells whorling around and weaving through bright pink supporting matrix (H&E, original magnification 400×).

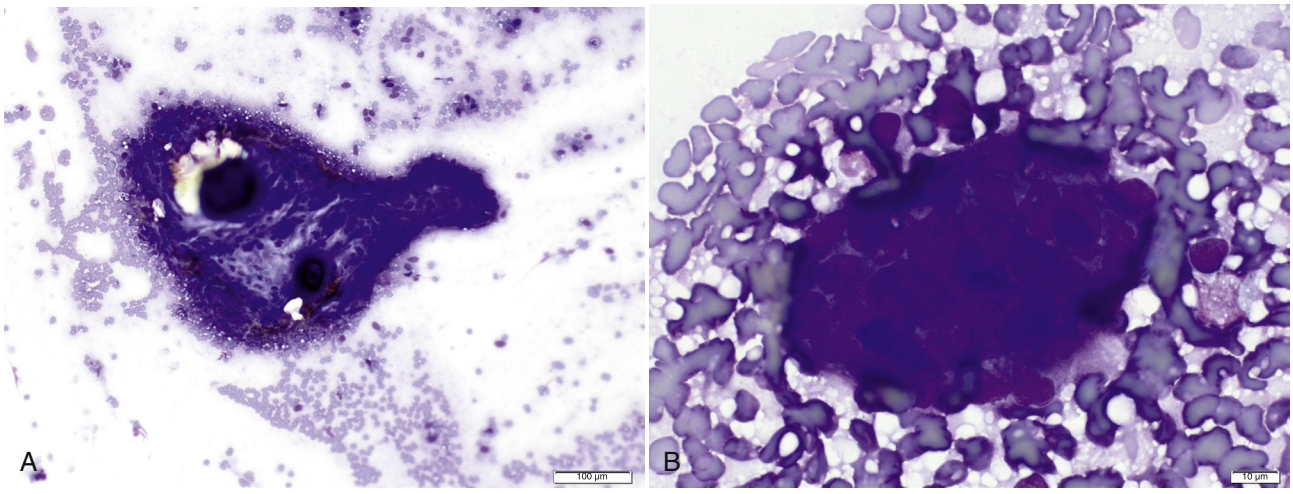

Fig. 14.18 Meningioma. (A) Low-magnification view of spindle-shaped cells arranged in whorls around centrally located mineralized material (presumed Psammoma body) (Diff-Quik, original magnification 100×). (B) Spindle cells appear to pile up on one another (Diff-Quik, original magnification 600×).

Cytological interpretation of brain biopsy specimens acquired via stereotaxy or open approaches may be challenging and does require a tumor that exfoliates well, a surgeon willing to provide multiple samples, and a cytologist with expertise in this area.[122] A study of 42 canine and feline cases of biopsy- or necropsy-confirmed CNS lesions showed squash-prep smear cytology to have 76% sensitivity in accurately determining diagnosis, with an additional 14% of cases having partial correlation between cytology and histopathology. For the remaining 10% of cases, cytological interpretation did not correlate with final diagnosis.[123] Cytological interpretation of CNS lesions may be very difficult, and biopsy with histopathological examination is recommended to confirm all diagnoses.

It is important for cytological samples to be prepared in the same manner each time to avoid introducing additional cytological variations that the pathologist has to read through. Some authors recommend wet-fixation of tissues followed by staining with hematoxylin and eosin (H&E).[122] At the authors' institution, CNS cytological samples are air-dried and stained with Diff-Quik or a modified Wright stain. The reader is referred elsewhere for a complete discussion of normal CNS cytology.[124] Clinical imaging findings, and signalment, should be considered carefully and may help the pathologist to formulate a list of potential differential diagnoses. It must be kept in mind that primary tumors may metastasize to the CNS, and these should be included in the differential diagnoses, where appropriate.

Meningioma

Meningiomas are composed of neoplastic cells arising from the meningothelial cells of the leptomeninges of the CNS.[125] These tumors are the most common primary CNS tumors of dogs and cats.[126] Histologically, these neoplasms are classified into at least nine subtypes based on appearance, and some tumors may be characterized by more than one pattern (Fig. 14.17).[125] Cytologically, smears are often characterized by spindle-shaped cells draped around vessels and arranged in large whorling structures (Fig. 14.18). Some cells may contain nuclei that display intranuclear cytoplasmic pseudoinclusions, but this is not a feature reliably seen on a majority of tumors (Fig. 14.19).[127]

Glial Tumors

As a whole, this group represents the second most common primary CNS neoplasm seen in dogs and cats.[126] Glial tumors are more

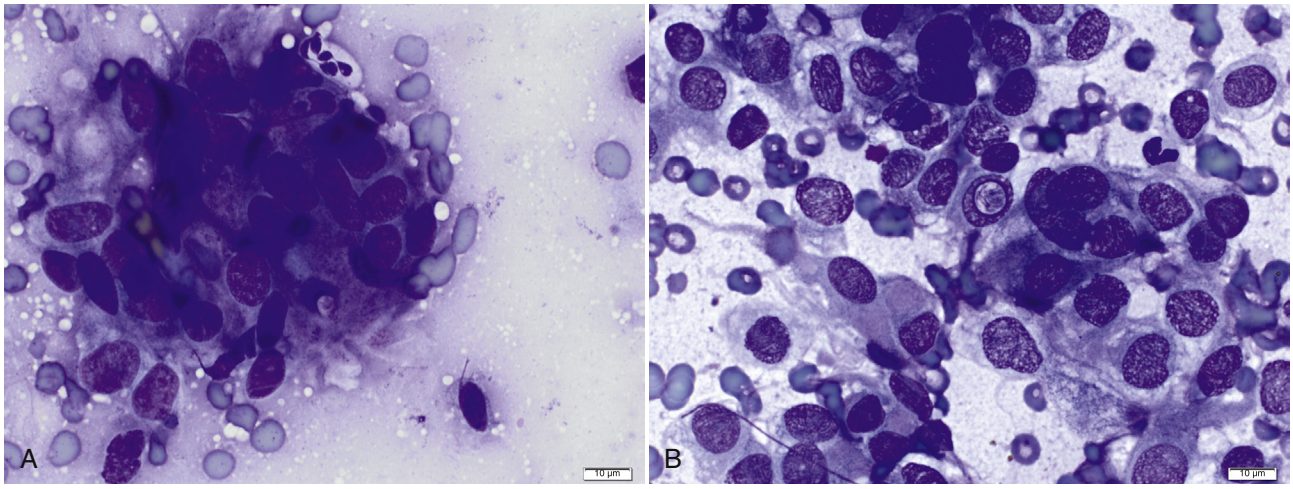

Fig. 14.19 Meningioma. (A) Cells contain plump oval to elongate nuclei, and neutrophils are occasionally observed adjacent to cell aggregates (Diff-Quik, original magnification 600×). (B) Infrequent cells *(center)* display intranuclear cytoplasmic pseudoinclusions (Diff-Quik, original magnification 600×).

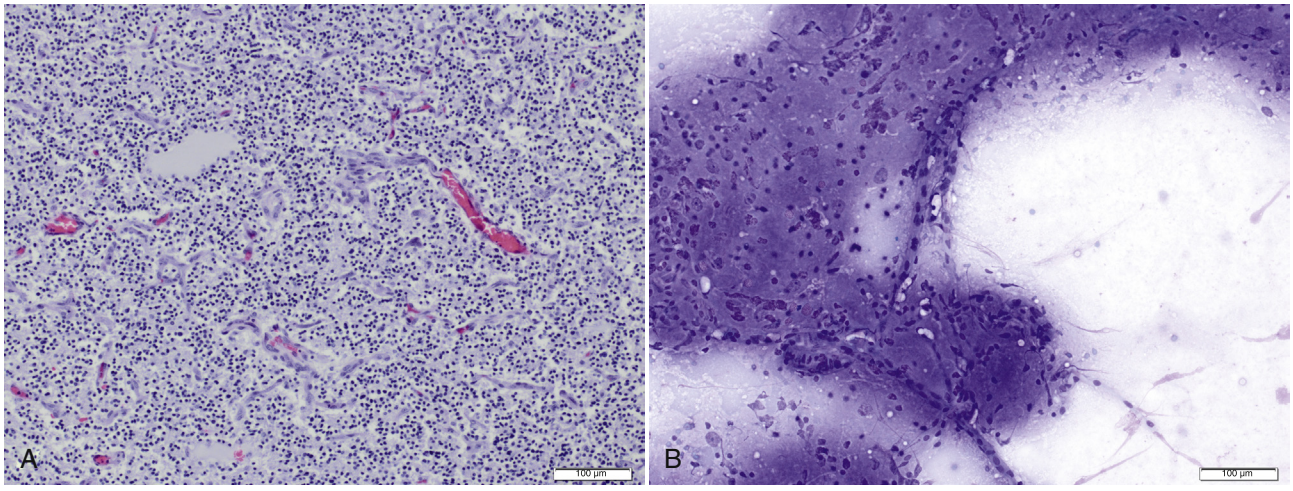

Fig. 14.20 Oligodendroglioma. (A) Tissue section showing round hyperchromatic nuclei surrounded by clear space admixed with branching capillaries and occasional lakes of basophilic mucin (H&E stain, original magnification 100×). (B) Numerous bare nuclei are seen admixed with purple fibrillar background material and branching capillary structures (Diff-Quik, original magnification 100×).

common than meningiomas in brachycephalic breeds.[126] Glial tumors arise from the supporting cells of the CNS. Astrocytomas are found most frequently in the cerebral hemispheres, although they have been reported to occur in various locations throughout the CNS.[125]

Astrocytoma

Astrocytomas arise from transformed astrocytes and are characterized cytologically by high cellularity, a high degree of nuclear pleomorphism, and fibrillar cytoplasmic processes.[125] Tumor cells will stain positively for glial fibrillary acid protein (GFAP).[125]

Oligodendroglioma

Oligodendrogliomas are derived from transformed oligodendrocytes and are found within the gray or white matter of the CNS, with the highest incidence in the cerebral hemispheres.[125] Cytological preparations are characterized by large numbers of blood vessels surrounded by neoplastic cells (Fig. 14.20).[125] Neoplastic oligodendrocytes have small amounts of eosinophilic cytoplasm surrounding uniformly round nuclei.[125]

Ependymoma

Ependymomas are derived from the ependymal lining cells found on the surface of the ventricular system of the brain and central canal of the spinal cord.[125] These tumors are rare and are found most often in the lateral ventricles.[125] Cytologically, smears are characterized by neoplastic cells palisading around branching vascular structures.[125] Cells are cuboidal to columnar in shape with high nuclear-to-cytoplasmic (N:C) ratios and eccentrically placed nuclei.[125]

Choroid Plexus Tumor

Choroid plexus tumors arise from the modified ependymal lining cells that contribute to the production of CSF. They are more common in dogs than in cats.[125] Papillomas and carcinomas have a very similar cytological appearance and may only be reliably differentiated on the basis of histopathological examination.[123] Cytological preparations contain polygonal cells arranged in rafts, columns, or papillary projections around capillary structures (Fig. 14.21).[125]

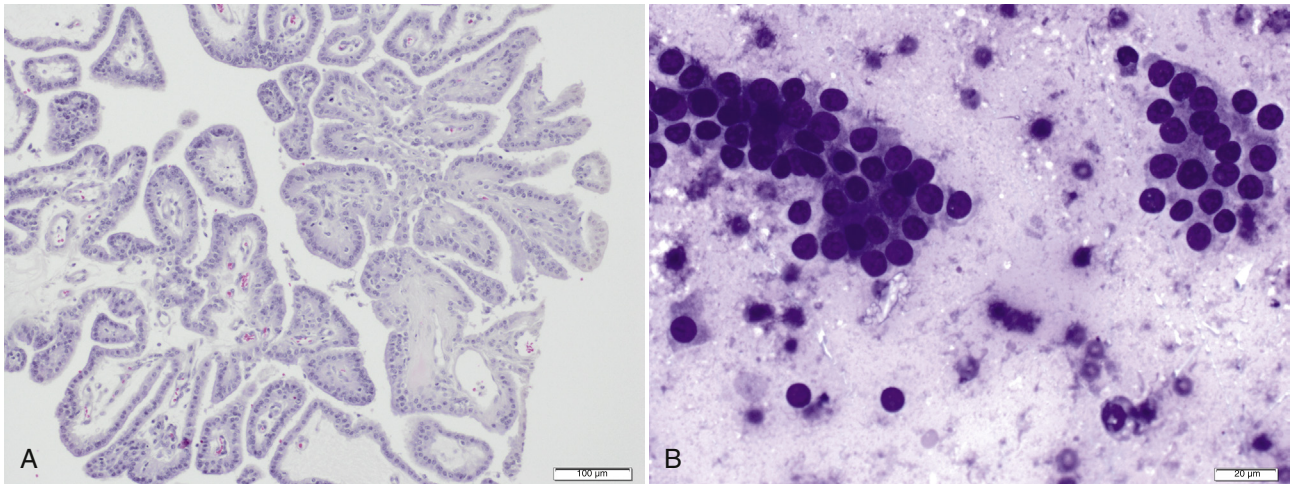

Fig. 14.21 Choroid plexus papilloma. (A) Cuboidal to columnar epithelial cells are arranged in papillary projections, which often contain small vessels (H&E stain, original magnification 100×). (B) Cuboidal epithelial cells are arranged in small sheets and papillary-like projections (Diff-Quik, original magnification 400×).

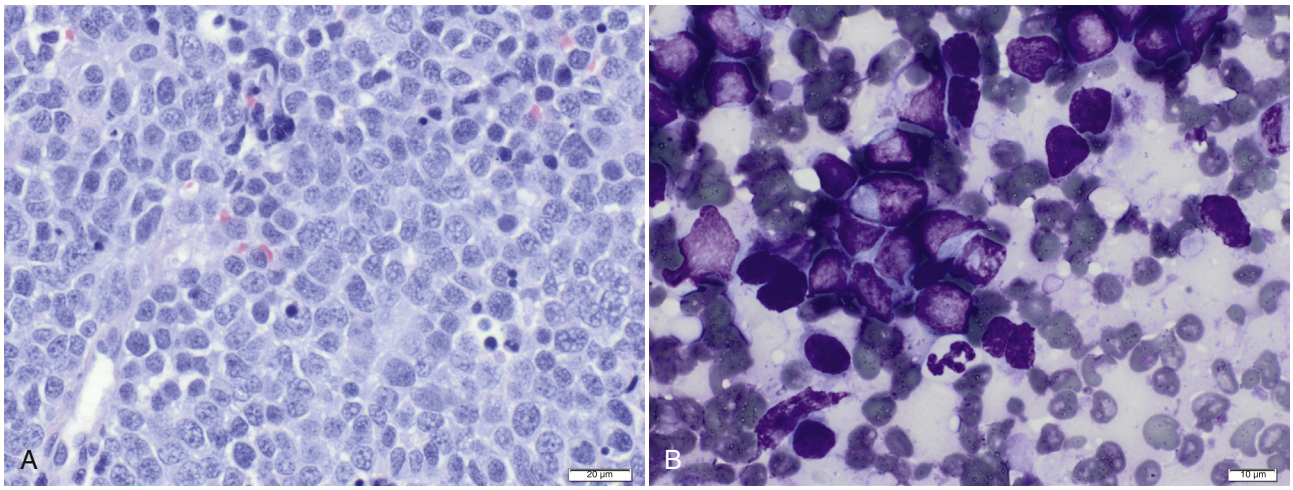

Fig. 14.22 Medulloblastoma. (A) Round cells are arranged in sheets (H&E stain, original magnification 400×). (B) Cells are large and round in shape with a high nucleus-to-cytoplasm (N:C) ratio and distinct cell borders (Diff-Quik, original magnification 600×).

Medulloblastoma

Medulloblastoma arises within the cerebellum and is a type of primitive neuroectodermal tumor derived from a germinal neuroepithelial cell.[125] Cytologically, preparations are highly cellular, composed of individual round cells that are large in size and have moderate to high N:C ratios. The appearance of these cells is reminiscent of large lymphocytes or histiocytes (Fig. 14.22).

Nephroblastoma

Nephroblastoma is a unique tumor arising in the spinal cord of young dogs (under age 4 years), usually between the T10 and L2 spinal cord segments.[125] The cytological appearance of this tumor has been described in a recent report and is characterized by three populations of cells: (1) high N:C ratio blastemal cells, (2) spindle-shaped mesenchymal cells, and (3) cuboidal epithelial cells.[128]

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