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ScienceDirect
The Veterinary Journal 172 (2006) 422–431
The Veterinary Journal
www.elsevier.com/locate/tvjl

Review

The function, composition and analysis of cerebrospinal fluid in companion animals: Part I – Function and composition

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Abstract

Cerebrospinal fluid (CSF) is a clear, colourless ultrafiltrate of plasma with low protein content and few cells. The CSF is mainly produced by the choroid plexus, but also by the ependymal lining cells of the brain's ventricular system. CSF flows through the ventricular system and then into the subarachnoid space and it is subsequently absorbed through the subarachnoid villi into the venous system. CSF has several functions in the nervous system. It protects the brain during blood pressure fluctuations, regulates the chemical environment of the central nervous system and it is a vehicle for intracerebral transport. This two-part article reviews CSF function, physiology, analytical techniques and interpretations in disease states of companion animals. This first part will address the function and composition of CSF in companion animals.

Keywords: Cerebrospinal fluid; Nervous system; Function; Blood–brain barrier; Composition

1. Introduction

Accurate analysis of cerebrospinal fluid (CSF) provides a wide range of information about the neurological health of the patient. Similar to a complete blood count, CSF analysis has good sensitivity (for the detection of disease), however, these changes rarely suggest a specific diagnosis; rather they allow individual diseases to be grouped in categories (inflammatory, neoplastic, metabolic) according to their ability to create similar changes. (Tipold, 1995; Bailey and Vernau, 1997). The possible abnormalities of CSF are relatively limited compared to the varieties of neurological disease that exist. The CSF is not always abnormal with some of these diseases (Braund, 1994), but occasionally it will help to provide a specific diagnosis. For these reasons accurate anamnesis, physical and neurological examinations, imaging studies and other diagnostic tests are essential for an accurate and correct interpretation of CSF changes in the context of an individual case (Chrisman, 1992).

2. Function of cerebrospinal fluid

2.1. Regulation of intracranial pressure (ICP)

The CSF has a protective role. In particular, the CSF protects the brain from changes in arterial and central venous pressure fluctuations associated with posture, respiration and exertion (Bailey and Vernau, 1997). CSF helps to modulate the normal variation in intracranial pressure (ICP) and together with cerebral blood flow helps to regulate ICP itself. The skull is a rigid structure with a fixed volume containing three components: brain tissue, intracranial vascular volume and intracranial CSF. Only the CSF and blood can be displaced to maintain constant intracranial volume and

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doi:10.1016/j.tvjl.2005.07.021

pressure without causing damage to the brain tissue. Thus, in the normal situation, the CSF volume and the blood volume vary inversely to maintain the ICP within normal limits (Herndon and Brumback, 1989a).

Physiological fluctuations of CSF pressure occur spontaneously with cardiac function and variations in respiratory activity. For example, in people, a pressure fluctuation of 1–2 mm H_2O is normally associated with the arterial pulse; during inspiration there is a fall in the CSF pressure which rises during expiration, probably due to the changes in the intracranial venous pressure associated with breathing. The amplitude of respiratory fluctuation is extremely variable, usually in the range of 2–5 mm H_2O with normal breathing and 5–10 mm H_2O with deep breathing (Herndon and Brumback, 1989b). In pathological ICP elevations, the fluctuations in CSF pressure become very marked (Lundberg et al., 1975). Additionally, there are different compensatory mechanisms: if there is a sudden increase in intracranial blood volume, CSF can be momentarily accommodated into the cervical subarachnoid space because of the elasticity of the dura mater. Chronic changes may be compensated for by increased absorption or decreased CSF formation. When these compensatory mechanisms fail, ICP rises, and cerebral blood flow falls. The following equation explains the relationship between ICP and cerebral blood flow (Rowland et al., 1991).

Cerebral perfusion pressure
= Mean arterial blood pressure − ICP.

Several types of abnormalities and pathological conditions, such as brain oedema and hydrocephalus, can also be accommodated by changes in CSF volume (Fishman, 1992; Rosenberg, 1990; Milhorat, 1987).

2.2. Regulation of the chemical environment of the central nervous system (CNS)

Cerebrospinal fluid has a role in the excretion of the potentially toxic by-products of cerebral metabolism. This is necessary to ensure that the fluids bathing the brain cells have a well regulated chemical composition because neurons require a consistent ionic composition in the extracellular space and are much less tolerant of changes than are other cell types.(Davson and Oldendorf, 1967; Katzman and Pappius, 1973). The brain lacks a lymphatic system, therefore substances such as proteins primarily leave the brain's tissue via the perivascular spaces, as well as by direct diffusion through the pia mater, into the subarachnoid space. On reaching the subarachnoid space, the protein flows in the CSF to eventually be absorbed through the arachnoidal villi into the cerebral veins (De Lahunta, 1983). Thus, the perivascular spaces serve as a modified transport to the lymphatic system for the brain.

In addition to transporting fluid and proteins, the perivascular spaces also transport extraneous particles, such as cells and even bacteria, from the brain into the subarachnoid space. For example, if an infection occurs within the brain, white blood cells are carried away from the brain through the perivascular spaces (Guyton and Hall, 2000). Other sites of CSF absorption and drainage include the veins and the lymphatic vessels around the spinal nerve roots, the spinal nerves, and the first two cranial nerves when they leave the skull (De Lahunta, 1983).

CSF also has a filtration function allowing movement of water-soluble substances from the brain parenchyma into the CSF. In this way, solutes entering the brain through the blood–brain barrier, as well as those synthesised by the brain, diffuse freely from the brain interstitial fluid into the CSF.

2.3. Intracerebral transport

CSF is a vehicle for the intracerebral transport of biologically active substances. For example, hormone-releasing factors (thyrotropin releasing-hormone, corticotropin releasing-hormone, grow hormone releasing-hormone, gonadotropin releasing-hormone), formed in the hypothalamus and discharged into the CSF of the third ventricle, may be carried in the CSF to their effective sites in the median eminence. The CSF can also be the vehicle for intracerebral transport of opioids and other neuroactive substances from the systemic circulation throughout the brain (Fishman, 1992; Milhorat, 1987). CSF is also known to carry neurotransmitters and neuropeptides, which may be altered in diseases such as epilepsy and spinal chord compression (Vaughn et al., 1988; Podell and Hadjiconstantinou, 1997; Ellenberger et al., 2004).

3. Anatomy and composition

3.1. Anatomy of the ventricular system

The ventricular system consists of two lateral ventricles within the cerebral hemispheres, each of which communicates through an interventricular foramen of Monro with the single midline third ventricle (Fig. 1) (De Lahunta, 1983; Herndon and Brumback, 1989a; Braund, 1994). Each lateral ventricle cavity can be divided into five regions – the anterior (frontal) horn, the body, the posterior (occipital) horn, the temporal (inferior) horn, and the collateral trigone (atrium). The third ventricle communicates through the cerebral aqueduct of Sylvius, or mesencephalic aqueduct with the midline fourth ventricle. The cerebral aqueduct of Sylvius running through the dorsum of the midbrain narrows after its origin from the third ventricle, then

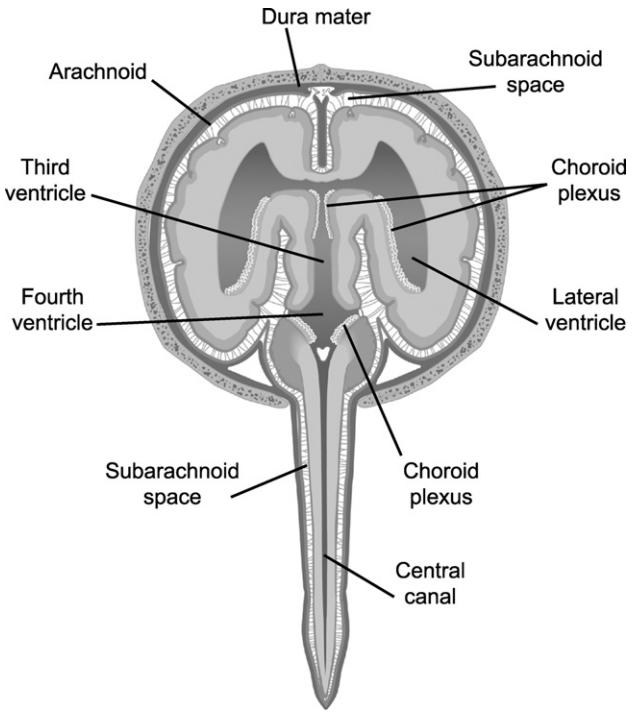

Fig. 1. The ventricular system and the pathway of cerebrospinal fluid flow from the choroid plexus in the lateral ventricles to the arachnoid villi. Modified from De Lahunta (1983). Reproduced from the Manual of Canine and Feline Neurology, 3rd edition, Edited by S. Platt and N. Olby (2004) with permission of the illustrator Allison Wright and BSAVA.

dilates into the ampulla (the ventricle of the midbrain) before narrowing again into the pars posterior, which dilates to open into the lateral apertures of the fourth ventricle (De Lahunta, 1983). The fourth ventricle is the triangular cavity of the medulla oblongata communicating with the third ventricle through the mesencephalic aqueduct and with the subarachnoid space through three openings in the roof of the fourth ventricle (a single median foramen of Magendie and two lateral foramina of Luschka). The cavity of the fourth ventricle extends into the central canal of the upper cervical spinal cord (Milhorat, 1987; Herndon and Brumback, 1989a; Fishman, 1992; Braund, 1994; Davson and Segal, 1996).

3.2. Fluid interfaces

The CNS (brain and spinal cord) is uniquely isolated in many ways from the body and the systemic circulation. There are several interfaces (Fig. 2) between brain tissue and systemic fluids, and there are three extracellular fluid compartments in the cranial cavity associated with brain parenchyma: plasma, CSF, and interstitial fluid (IF). Cells forming the interfaces, or barriers, between the rest of the body and the neural tissue, control the composition of the CSF and the interstitial fluid. These semi-permeable interfaces (the blood–brain barrier (BBB), the blood–CSF barrier and the CSF–brain barrier), control the production and absorption of the CSF and provide a fluid environment that is relatively stable despite changes in the composition of the blood (Andrews, 1998). These barriers are necessary because brain cell survival depends on a careful regulation of the local fluid environment. When the barriers separating the blood from the brain are injured, toxic substances from the blood invade the brain (Guyton and Hall, 2000).

The movement of molecules from blood to brain is regulated by the above-mentioned interfaces formed by epithelial and arachnoid cells, tightly joined together forming a series of epithelial sheets. The junctions allowing the exchange to occur join the epithelial cells lining the ventricles and covering the brain. These barriers exist both in the choroid plexus and at the tissue capillary membranes in essentially all areas of the brain parenchyma except for some areas of the hypothalamus, the pituitary gland, and the area postrema, where substances diffuse with ease into the tissue spaces (Miller and Leslie, 1994; Harding et al., 1985). The area postrema is located on the dorsal surface of the medulla oblongata at the caudal end of the fourth ventricle, and it is implicated as a chemoreceptor trigger zone for emesis. It is anatomically positioned to detect emetic toxins in the blood as well as in the CSF (Bailey and Vernau, 1997). The area postrema of the brain helps to monitor and control changes in body fluid components, such as plasma osmolality, glucose concentration, and electrolyte concentration (De Lahunta, 1983; Fishman, 1992; Braund, 1994).

A connection exists between the scala tympani and the subarachnoidal space by the cochlear aqueduct. The pressure in the three compartments of the cochlea fluctuates in parallel to changes in the CSF in several animal species, but the actual rate of flow between the CSF and the perilymph via the cochlear aqueduct is unknown. It is possible that the endolymph duct may be important in the spread of infection between the inner ear and the meninges (Oldendorf, 1977).

3.2.1. Blood–brain barrier

The blood–brain and blood–spinal cord interfaces exist between the plasma and the IF at the level of capillaries (Fig. 3) (Andrews, 1998). The BBB consists of non-fenestrated endothelial cells with interendothelial tight junctions (Mitic and Anderson, 1998). These junctions are complexes of protein (zonula occludens-associated proteins, and cingulin) (Butt, 1995; Aubrey et al., 2000), which are responsible for the lack of permeability of the BBB and its high electrical resistance (5000–8000 Ω/cm^2 for the parenchymal vasculature), when compared with most epithelia of the rest of the body (less than 100–200 Ω/cm^2). The high electrical resistance indicates the tight nature of the BBB and its low ionic

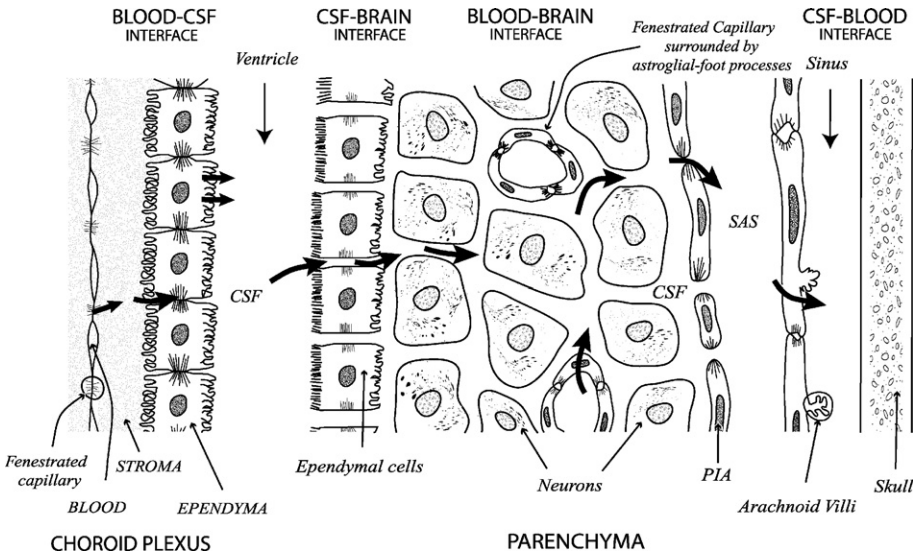

Fig. 2. Schematic diagram of interfaces between blood, brain and cerebrospinal fluid (CSF) (see text for description). Arrows ➡ indicate formation of CSF at the choroid plexus, and interstitial fluid at the brain capillary. The cerebrospinal fluid is absorbed in to blood at the aracnoid villi. SAS, subarachnoid space. (R. Di Terlizzi, modified from Rosemberg, 1983).

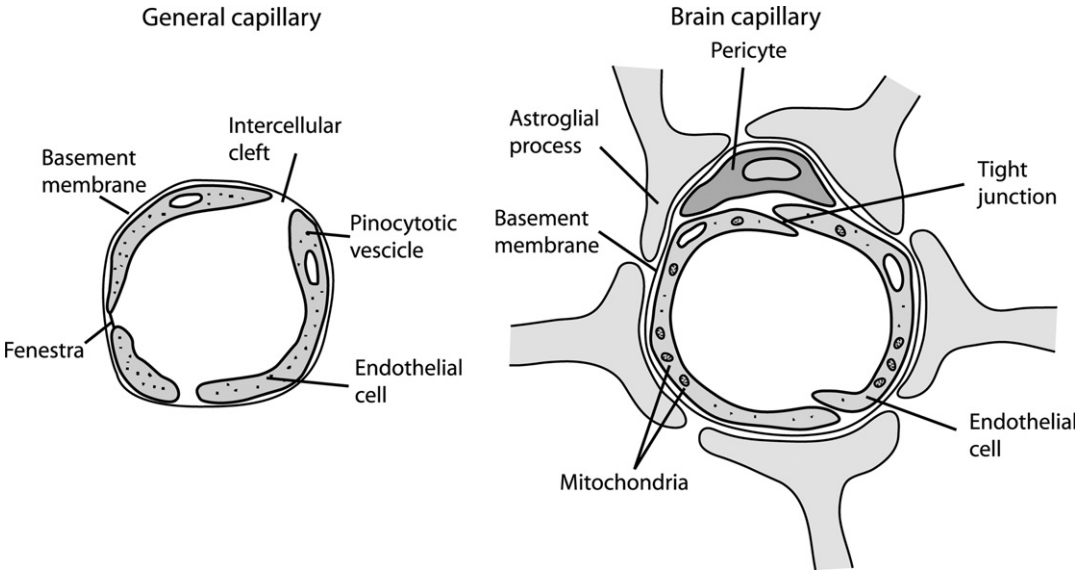

Fig. 3. Blood–brain barrier. Unlike most capillaries of the body, cells of brain capillary walls are joined by tight junctions. These junctions regulate the passage of solutes from the blood. As a result, the exchange of blood solutes is highly selective. (R. Di Terlizzi, modified from Rowland et al., 1991).

permeability (Milhorat, 1987; Rosenberg, 1990; Davson and Segal, 1996; Aubrey et al., 2000). CNS endothelial cells represent the physical component of the barrier and are the most important structures responsible for maintaining the neuronal environment on account of their large surface area in comparison to the other interfaces. The barrier formed by the interendothelial tight junctions provides a mechanism of compartmentalising the CNS from the systemic circulation but this separation is often compromised in a variety of inflammatory conditions (Braund, 1994). The capillaries are surrounded by different cells: pericytes, perivascular macrophages and astrocytes (Lewis, 1976).

3.2.2. Blood–CSF barrier

The blood–CSF barrier exists between plasma and CSF at the choroid plexus and consists of two cell layers separated by a thin basal membrane. The vascular endothelial layer is a monolayer of epithelial fenestrated tissue (Rosenberg, 1990; Milhorat, 1987; De Lahunta, 1983). The choroidal epithelium consists of specialised tall columnar cells, similar to other secretory epithelia.

The surface of the choroid plexus has frond-like villous processes which markedly increase the surface area exposed to the ventricular cavity. Each villus is covered by a single layer of cuboidal epithelium and has a central core consisting of a capillary surrounded by a small amount of loose connective tissue. The epithelial cells have microvillus brush borders and numerous basal and lateral in-foldings. Occasional epithelial cells have cilia on their apical surfaces. Apical tight junctions between adjacent epithelial cells provide a barrier to the passage of macromolecules. These cells have numerous mitochondria, a Golgi complex, endoplasmic reticulum, and an abundance of lysosomes (Andrews, 1998). This kind of epithelium, characteristic of the circumventricular organs (four choroid plexi, the median eminence, the neural lobe of hypophysis) borders the brain ventricles and is involved in specific secretory activities that require direct contact with the plasma (Rosenberg, 1990). The blood–brain barrier is lacking here, so the hypothalamic cells are exposed to the circulating blood (Rosenberg, 1990).

The extracellular space of the median eminence is exposed to substances in the blood that can modulate release of the hypothalamic releasing factors. To compensate for the absence of the BBB, the ependymal layer over the hypothalamic region of the third ventricle has tight junctions that limit the movement of substances between the hypothalamic nuclei and the CSF. Thus, substances that enter into the brain in the hypothalamic region are restricted from moving into the CSF and confined within the brain parenchyma (De Lahunta, 1983). The final site where the blood and the CSF meet is at the arachnoid membrane of the arachnoid villi (Fig. 4). These villi are microscopic evaginations of the arachnoid membrane into the lumen of the dural sinuses. The barrier nature of the cells is due to their tight junctions.

An important function of the arachnoid villi is to prevent blood in the venous sinus from entering the CSF whilst allowing CSF components to enter the vascular space. This is accomplished with valve-like channels that collapse when pressure is applied from the venous side and open when the CSF pressure increases (Rosenberg, 1990). Even when the sinus pressure exceeds that in the CSF, there is no reversal of flow. The arachnoid villi therefore act as one-way valves (Milhorat, 1987; Bell, 1995). Arachnoid villi are not limited to intracranial venous sinuses, but are also present at the spinal nerve roots penetrating into the spinal veins (Lewis, 1976). These structures have a semi-permeable membrane between the plasma and the CSF; carbon dioxide (CO_2) readily passes between plasma and CSF, whereas bicarbonate exchange is slow owing due to the relative impermeability of this barrier to bicarbonate (Page et al., 1979; De Lahunta, 1983; Braund, 1994).

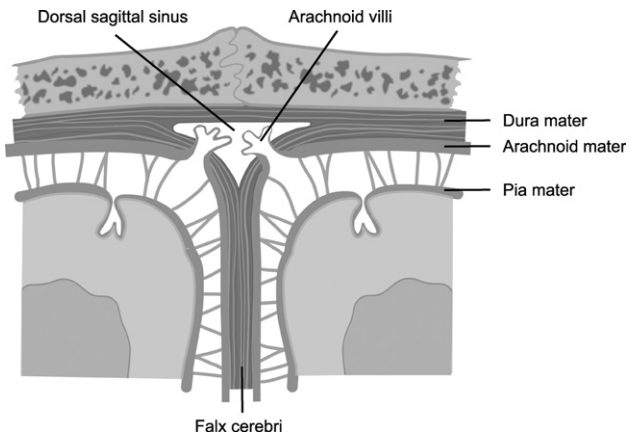

Fig. 4. Cerebrospinal fluid is absorbed into the venous system through the arachnoid villi. Reproduced from the Manual of Canine and Feline Neurology, 3rd edition, Edited by S. Platt and N. Olby (2004) with permission of the illustrator Allison Wright and BSAVA.

3.2.3. CSF–brain/spinal cord interface

The CSF-IF barrier occurs over the outer surface of the brain and in the ventricles. The surfaces of the ventricles are lined with a cuboidal epithelium called ependyma, which usually lies above a subependymal layer of glial cells. At this location, an exchange between ventricular fluid and the adjacent subependymal extracellular fluid of the brain occurs. The ependymal cells are epithelial cells that have different forms depending on the region in which they are found. In some regions, the epithelial cells of the ependyma have cilia in contact with the CSF (Davson and Segal, 1996). On the floor of the third ventricle, there is a region called the median eminence or infundibulum which possesses specialised ependymal cells called 'tanycytes'. The median eminence connects the hypothalamic nuclei with the ventricular surface.

The brain and the spinal cord are surrounded by the leptomeninges. They are divided into the arachnoid mater and the inner layer, the pia mater. The two membranes contain the subarachnoid space filled with the extraventricular CSF. The leptomeninges contain few capillaries. The pia mater forms the outer surface of the perivascular space through 'invagination' into the nervous system and tissue derived from the arachnoidea contributes to the inner wall of this space (De Lahunta, 1983). Both the ependyma and the pia mater, a single layer of cells, are extremely permeable. For this reason, all the substances that enter into the CSF can also diffuse readily into the superficial areas of the brain's interstitial fluid. However, substances in the interstitial fluid can often diffuse in the other direction; in this way, the CSF and the nervous system interstitial fluid are contiguous (Rosenberg, 1990; Fishman, 1992; Bagley, 1996).

3.3. CSF formation (Fig. 1)

The majority of CSF is formed principally by the choroid plexi at the rate of approximately 0.047 mL/

min in dogs (dependent on the size of the animal), 0.017 mL/min in cats, 0.002 mL/min in rats and 0.35 mL/min in people (De Lahunta, 1983; Milhorat, 1987; Davson and Segal, 1996). The rate of formation is closely related to the weight of the choroid plexi and to the rate of sodium and bicarbonate ion exchange. In people, the daily production rate is about three to four times as much as the total volume of fluid in the entire CSF system (Milhorat, 1987; Davson and Segal, 1996). Probably two thirds or more of this fluid originates as a secretion from the choroid plexi in the four ventricles, and mainly in the two lateral ventricles (Bering and Sato, 1963; Lorenzo et al., 1970).

The ependymal lining of the ventricular system, the external pial–glial membrane of the brain surface, and blood vessels in the pia-arachnoid secrete additional amounts of fluid (Speake et al., 2001). Experimental studies in dogs have demonstrated that 35% of CSF is derived from the third and lateral ventricles, 23% from the fourth ventricle, and 42% from the subarachnoid space (Rosenberg, 1990). The choroid plexi are small, cauliflower-like growths that project into all four ventricles and consist of small capillaries covered by a thin epithelial layer. Both ultra-filtration of plasma across the choroidal capillary endothelial wall and active secretion by the choroidal epithelial cells are involved in the formation of the CSF. The secretion of CSF, like the aqueous humour, depends primarily on the active transport of sodium ions into the ventricles (Fishman, 1992).

Sodium–potassium ATPase in the membrane of the choroidal epithelial cells plays an important role in the formation of CSF. Sodium is actively transported out of the choroidal epithelial cell into the ventricular cavity, with water molecules and chloride and bicarbonate ions following through by way of facilitated transport (Herndon and Brumback, 1989a; Bailey and Vernau, 1997). The enzyme carbonic anhydrase, which catalyses the reaction of carbon dioxide and water to form carbonic acid (which then dissociates into hydrogen and bicarbonate ions), also seems to be involved in formation of CSF in the choroidal epithelium (De Lahunta, 1983). Compared with plasma ultrafiltrate, CSF concentrations of chloride, sodium, and magnesium are slightly higher, and potassium, total calcium and glucose are slightly lower; additionally there is significantly less protein in the CSF (Guyton and Hall, 2000).

Because CSF formation is in part an active, energy-dependent process, its formation is very constant and independent of either CSF pressure or blood pressure. The villi function like 'valves', allowing the fluid and its contents to flow readily into the venous blood of the sinuses while not allowing the blood to flow backward in the opposite direction (Cutler et al., 1968). Normally, this one-way valve action of the villi allows CSF to begin to flow into the blood when CSF pressure is about 2 cm H_2O greater than the pressure of the blood in the sinuses (Herndon and Brumback, 1989b). As the CSF pressure rises higher, the valve opens further, which means that, under normal conditions, the CSF pressure almost never rises more than a few cm H_2O more than the pressure in the venous sinuses. Acute changes in plasma osmolality may directly affect the filtration process; hypo-osmolality increasing CSF formation and hyperosmolality reducing it. The relationship is almost linear, with a nearly 7% change in the rate of CSF formation with each 1% change in plasma osmolality in experimental animals (Bell, 1995). Acute increases in intraventricular pressure up to about 30 cm H_2O have minimal effect on the rate of the CSF formation, but chronic increases in pressure, such as in some forms of hydrocephalus, apparently reduce CSF formation and may result in atrophy of choroid plexi (Andrews, 1998).

Absorption through arachnoid villi occurs transcellularly through micropinocytotic vesicles, but may also occur through an endothelium-lined, intercellular cleft. This mechanism is variable among species (Rosenberg, 1990). Particles ranging in size from colloids (0.2 μm diameter) to human erythrocytes (7.5 μm diameter) can pass from the subarachnoid space into the venous sinus, while larger particles are excluded (Rosenberg et al., 1980).

The arachnoidal cells in the villi are joined by tight junctions forming a continuous lining of the subarachnoid surface, and they also contain giant vacuoles. These vacuoles, some of which have openings on both the basal and apical surfaces of the cell and provide a transcellular channel, are responsible for the mechanism of bulk transport of fluid and particulate matter into the venous system (Fishman, 1992; Milhorat, 1987). CSF absorption in people begins at an average CSF pressure of 7 cm of water (1 cm of $H_2O = 7.5$ mm Hg) and increases linearly with increasing pressure up to 25 cm H_2O at which point the absorption rate is around 90 mL/h. Equilibrium between the rates of formation (constant at 20 mL/h over a wide range of CSF pressures) and absorption is achieved at the CSF pressure of approximately 11 cm H_2O (Higgins et al., 1977; De Lahunta, 1983). No information on these values could be found for dogs and cats. In disease conditions, accumulation of large particles (e.g., protein, erythrocytes, leukocytes, etc.) within the villi may impair absorption and cause hydrocephalus (Wilson and Stevens, 1977; Cook and DeNicola, 1988; Chrisman, 1992). Similarly, inflammation of villi in CNS inflammatory processes may cause occlusion leading to hydrocephalus (Bailey and Higgins, 1985; Rand et al., 1990).

3.4. CSF composition

CSF is a product of plasma filtration and membrane secretion, and so its composition is different from that of plasma. CSF, in general, is clear, colourless, nearly acel-

lular, and has a low protein concentration. Various ions, enzymes, and other substances are also found in normal CSF. In healthy animals, as in people, the CSF composition is maintained relatively constant by the various membrane interfaces, but some fluctuation can occur with fluctuations in plasma composition.

Erythrocytes and nucleated cells: CSF normally does not contain erythrocytes (Bailey and Higgins, 1985) and the normal nucleated cell count is variable in different species. Normal range for total nuclear cell count (TNCC) is reported to be from 0 to 5 cells/μL in dogs and from 0 to 8 cells/μL in cats with the majority of animals having 0–2 cells/μL (Meinkoth and Crystal, 1999).

Protein concentration of normal CSF is very low. The lumbosacral samples tend to have slightly higher levels compared with the cisternal samples (Fishman and Chan, 1980). Typical reference ranges for dogs and cats are 10–40 mg/dL compared to 5–7 g/dL in the serum (Crone, 1965). Albumin is the main protein in the CSF (50–70%), and normally γ-globulin levels are very low (5–12%) (Fishman, 1992). When the BBB is damaged, protein leaks into the CSF. Albumin enters the CSF in the greatest quantities because the concentration of these proteins is highest in the blood. However, in some diseases there is an increase in γ-globulins that are secreted by B lymphocytes present in the nervous system.

Glucose in the CSF is derived essentially from the plasma and carried into the brain by facilitated transport or diffusion; only a limited amount of glucose enters the brain by diffusion (Fishman, 1992). When the concentration of glucose in the blood is low, it is more avidly transported across the capillary by the carrier mechanism. However, high concentrations of glucose saturate the carrier molecules. Therefore, the concentration of CSF glucose depends on the blood glucose concentration, the rate of glucose transport into the CSF, and the metabolic rate of the central nervous system. The normal CSF glucose concentration is about 60–80% of the blood glucose concentration, reflecting in part the high metabolic rate of the central nervous system (Davson and Pollay, 1963; Rosenberg, 1990). In people, a glucose gradient has been demonstrated to exist along the neuraxis; the concentration decreases from ventricular to lumbar fluid (Fishman, 1959).

Sodium is the most abundant ion in the CSF, being important in transport and osmoregulation. CSF and plasma sodium concentration are closely related (Crone and Christensen, 1981). Some studies reported that acetazolamide, an inhibitor of carbonic anhydrase, slows the entrance of sodium into the CSF, and vasopressin enhances the movement of sodium from blood to brain (Bradbury and Davson, 1965). The electrical charge across a membrane is determined by the distribution of charged ions on either side of the membrane. In epithelial membranes, the Na^+, K^+-ATPase pump, regulates the electrical charge. Tight-junctioned epithelial sheets have a high electrical resistance, whereas those with more porous junctions have a lower resistance (Katzman and Pappius, 1973). These levels of resistance give an indication of the tightness of the junctions.

Potassium concentration is critical for neuronal function and the release of neurotransmitters. Potassium ion concentration is lower in CSF than in plasma and it is maintained within a very narrow margin. The normal CSF potassium concentration is 3 mmol/L. Changes in plasma potassium concentration have little effect on the CSF potassium levels (Rosenberg, 1990). Even with very high potassium plasma concentrations, the CSF potassium concentration remains within the normal range (Murphy et al., 1986), because its transport across the BBB is limited. Choroid plexus epithelium has a lower permeability to potassium than to sodium, and the reverse is true at the capillary level (Rosenberg, 1990). When potassium levels are increased in the CSF, sodium is exchanged with potassium by an active transport mechanism.

Total calcium in the CSF is normally in the range between 1 and 1.5 mmol/L (plasma levels: 2–2.75 mmol/L). Calcium is secreted from the choroid plexi and the amount that enters from blood into the CSF is independent of the concentration of calcium in the plasma. The low CSF concentration of calcium is maintained by transport mechanisms between blood and CSF. This active transport mechanism exists at the BBB to maintain calcium homeostasis. Both cerebrovascular endothelium and the choroid plexi participate in this active process (Wood, 1983; Benga et al., 1985). Acute and chronic changes in plasma calcium concentration have little effect on brain calcium levels (Fenstermacher and Rall, 1972).

Magnesium and chloride are found at slightly higher concentrations in CSF than in plasma and both are known to play an important role in neuronal conduction (Maren, 1992). The transport of these ions between the blood and CSF does not occur exclusively by passive transport (Wood, 1983). Some studies (Wilson and Stevens, 1977; Rand et al., 1990; Jackson et al., 1996) have reported that the intravenous administration of acetazolamide in cats causes a proportional reduction in the rate of CSF formation as well as the entry of chloride into the CSF. This finding suggests that the movement of chloride ions from blood to CSF is closely linked to CSF production, but the exact mechanism is obscure (Banik et al., 1983).

Enzymes. Numerous enzymes have been found in CSF of domestic animals (Fishman, 1992; Kjeldsberg and Knight, 1986) and people (Kramer and Hoffmann, 1997). These enzymes have three different sources: (1) blood, (2) neural tissue or neural tumours, (3) cells within the CSF (Indrieri et al., 1980; De Lahunta, 1983;

Raicevic et al., 2000). Creatine Kinase (CK) is a dimer composed of two subunits (B or brain, M or muscle) limited to cardiac and skeletal muscle (isoenzyme CK-2 oMB and CK-3 o MM) and nervous tissue (isoenzyme CK-1 o BB), (Indrieri et al., 1980; Wakim and Fleisher, 1956). An elevation in serum CK often reflects muscle disease, whereas elevations in CSF may reflect nervous system disease. The concentration of CK in CSF is usually independent of that in serum and may reflect a variety of neurological diseases with guarded to poor prognosis (Vaagenes et al., 1988).

Aspartate transaminase (AST) and CK may increase in the CSF when extensive myelin degeneration has occurred (De Lahunta, 1983). Also CSF activities of CK, AST, and lactate dehydrogenase (LDH) are decreased at 6 h after cardiac arrest in humans with the most severe neurological damage (Abate et al., 1998). LDH activity is increased in lymphoma of the central nervous system (Yin et al., 2002); inflammatory disorders are often characterised by an increase in LDH activity, while in non-inflammatory disorders (hydrocephalus and spinal cord neoplasia) no variation in LDH activity is detected (Sugi et al., 1975; Braund, 1994).

A recent study (Head et al., 2002) demonstrated decreased activity in CSF enzymes after subarachnoid haemorrhage in dogs. The authors measured the changes in glucose, glutamate, lactate and pyruvate concentrations in CSF during cerebral vasospasm. Changes in lactate and pyruvate concentrations may be found in some cases with mitochondrial disease. The lactate:pyruvate ratio reflects the oxidative state of the brain (Davis, 1990). The concentration of lactic acid in the brain is dependent upon its rate of production and independent of blood lactate concentration (Vaughn et al., 1988; Bailey and Vernau, 1997).

Neurotransmitters. Neurotransmitters are produced by neurons and they have been studied in humans in different neurological diseases (Loescher and Schwartz-Porsche, 1986; Podell and Hadjiconstantinou, 1997). The concentration of several neurotransmitters and their metabolites have been measured in the CSF of different animals (Ellenberger et al., 2004).

Gamma amino-butyric acid (GABA) is a major inhibitory neurotransmitter in the brain and in the spinal chord. Low levels of GABA are found in CSF of dogs with epilepsy (Meldrum, 1994; Ellenberger et al., 2004).

Glutamate (GLU) is the major excitatory neurotransmitter in the CNS. It has been found to play an important role in the epileptic activity (Spranger et al., 1996; Koutsilieri et al., 1999), and that the GLU to GABA ratio may be a useful indicator of genetic epilepsy (Olby et al., 1999). Glutamate is also considered to be a mediator of secondary tissue damage and elevated concentrations are found in several diseases (De Lahunta, 1983). Chronic and acute compressive spinal cord lesions in dogs due to intervertebral disc herniation are associated with elevation in CSF glutamate concentration (Frier et al., 1974; De Lahunta, 1983).

3.5. Pressure

In normal animals, CSF pressure is regulated primarily by its absorption through the arachnoid villi. The rate of CSF formation is very constant, and so this process is rarely a factor in pressure control. Most dogs have a pressure of <170 mm H_2O and most cats <100 mm H_2O under general anaesthesia. An obstruction of the venous return from the head causes CSF pressure to rise almost immediately.

There are many diseases in which the villi became blocked by large particulate matter, by fibrosis or an excess of plasma protein molecules in the CSF. Examples of these conditions include: (1) space-occupying lesions (tumours and haemorrhage), in these cases there is compression of the venous sinuses that prevent the CSF absorption at the arachnoid villi; (2) cerebral oedema (usually associated with brain injuries); (3) meningoencephalitis; (4) vitamin A deficiency causing poor absorption of CSF due to atrophy of the villi (De Lahunta, 1983).

Acknowledgements

The authors are grateful to Dr. Steve Stockham, veterinary clinical pathology professor at Kansas State University College of Veterinary Medicine for his advices, guidance, suggestions, and critical review of this manuscript.

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