CHOROID PLEXUS, AGEING OF THE BRAIN, AND ALZHEIMER'S DISEASE

Jean-Marie Serot [1, 2], Marie-Christine Béné [2], Gilbert C Faure [2]

[1] Hotel-Dieu 54350 Mont Saint Martin, France, [2] Laboratoire d'Immunologie, Faculte de Medecine, 54500 Vandoeuvre les Nancy, France

TABLE OF CONTENTS

1. ABSTRACT

Choroid plexus tissues are intraventricular structures composed of villi covered by a single layer of ciliated, cuboid epithelium. The plexuses secrete cerebrospinal fluid (CSF), synthesize numerous molecules, carry nutrients from the blood to CSF, reabsorb brain metabolism by-products and participate in brain immunosurveillance.

During ageing, atrophy of epithelium occurs along with thickening of basement membranes. Enzymatic activities of epithelial cells decrease significantly. CSF secretion decreases as much as 50%. These modifications are concurrent with subnormal brain activity.

In Alzheimer's disease, epithelial atrophy, thickening of basement membrane and stroma fibrosis are even more prominent. Ig and C1q deposition along the basement membrane can be frequently detected, suggesting immunological processes. Synthesis, secretory, and transportation functions are significantly altered resulting in decreased CSF turnover, reduced beta-amyloid clearance, and increased glycation phenomena as well as oxidative stress. Such modifications may favour fibrillary transformation of beta-amyloid protein and tau protein polymerisation.

2. INTRODUCTION

Alzheimer's disease (AD) afflicts 5% of people more than 65 years old and at least 35% of people older than 85. This disease is altogether a personal drama, a familial "catastrophe", a major public health problem and a therapeutic challenge.

Lesions are characterized by the association of diverse abnormalities that can be encountered with a lower intensity and slightly different topography in non-demented elderly subjects. Cortical atrophy occurs mainly in parieto-temporal areas. Cerebral cortex contains numerous senile plaques (SP). These broadly round structures are made of extra-cellular β-amyloid deposits surrounded usually by a neuritic corona containing intra-neuronal fibrillar inclusions, neuro-fibrillar tangles (NFT) (1). Despite the development of neuropsychological diagnostic criteria, a definitive diagnosis is only made by pathological analysis of the brain (2) and no specific peripheral blood marker or preventive treatment is yet available.

Notwithstanding the progress of research, the pathogenesis of AD is not fully elucidated. Protein β-amyloid fibrillotransformation and NFT appear to be the final common pathway of various metabolic disorders. Overproduction or decreased clearance of β-amyloid peptides are assumed to be responsible for progressive accumulation and aggregation in brain interstitial fluid. Various mutations of amyloid precursor protein (APP) and presenilins 1 and 2, by modifying APP catabolism, induce hereditary forms through protein β-amyloid hyperproduction (3). In sporadic AD, which represents 99 % of cases, there is neither APP hyperexpression nor mutation of the above mentioned 3 genes (3); however the brain contains high amounts of neurotoxic soluble oligomers of β-amyloid protein (4) which are small aggregates that are soluble and neurotoxic. Major oxidative stress also occurs in the brain (5) which might explain fibrillotransformation of amyloid protein and NFT (6). Age, apoE4 phenotype and menopause are risk factors truly defined. Age plays the major role. This suggests that modifications related to ageing, associated with other genetic and environmental factors, might modify amyloid protein metabolism (7, 8).

Brain is immersed in CSF, which confers mechanical protection and diminishes apparent weight according to Archimede's principle (9). CSF should not be considered as an inert and static fluid. It is renewed several times a day and is a medium for transportation of many molecules. It is used for elimination of by-products of cerebral catabolism and toxic compounds. CSF is similar to brain interstitial fluid. It is almost completely secreted by choroid plexus (CP). CPs are intraventricular epithelial formations specialized in CSF secretion. They are contiguous with the ependyma. CP secretions are critical for maintaining water balance and therefore the brain volume (9). CP undergoes important morphological modifications during ageing. Recent reports indicate that the structure and functions of CP are significantly altered in AD.

3. STRUCTURE AND FUNCTIONS OF CHOROID PLEXUS

3.1. Structure of choroid plexus

Three CP regions are described: CP of the 3rd ventricle or Vicq d'Azyr plexus, is a median ribbon in the choroid network on the roof of 3rd ventricle; it is subdivided in 2 "cordons" which reach right and left foramen of Monro and run on the internal surface of each lateral ventricle. There are also 2 small CPs on the roof of 4th ventricle. Most studies analyze the lateral ventricle CPs which are more readily accessible. Weight of human CP is about 3 g (10).

Choroid tissues are composed of villi covered by an unistratified epithelium with a central vascular axis. Epithelial cells are cuboid with a rounded central or basal nucleus (11). Mitochondria are more numerous at the basal and apical poles, occupying 15 % of cytoplasm in primates (12). Epithelial cells present at the apical pole numerous microvilli of uniform diameter, enmeshed with each other, and a few cilia. The basolateral membrane contains numerous interdigitations. Transmission electron microscopy reveals light and dark cells as ultrastructurally similar. The dark aspect could be due to dehydration related to hypofunctionality (13). In man, between epithelial cells, near the basement membrane, dendritic cells can be found (< 1 % of epithelial cells). They appear to secrete IL-10 (14).

Epithelial cells are about 15 μm high. Their total number has been estimated at 10^8. They lie on an epithelial basement membrane surrounding a thin stroma with numerous collagen fibers, scarce dendritic cells, macrophages, fibroblasts and large capillaries with a fenestrated endothelium (11). CPs are richly innervated, receiving adrenergic, cholinergic, peptidergic and serotoninergic fibers. The distribution of nervous fibers varies widely according to species (15). Choroid blood flow is high, between 4 and 7 times greater than brain flow depending upon species (16).

3.2. Functions of choroid plexus

Choroid epithelial cells keep some properties of ependymal cells. For instance, they transport glucose from blood to CSF by GLUT1, a glucose transporter (17). They immunostain for presenilin-1 (18) and lipocortin-1, an anti-inflammatory corticosteroid (19). By the synthesis of PLTP, i.e. phospholipid transfer protein (20), they are implicated in brain lipoprotein metabolism.

CP functions are numerous. Choroid tissues belong to the hemato-encephalic barrier. Metabolic activity of CPs is estimated to be half that of the kidneys (10). The plexuses secrete about 90 % of CSF; the remaining 10 % comes from brain interstitial fluid drainage. CSF formation is an active osmotic process regulated by 2 enzymes: carbonic anhydrase and Na^+K^+-ATPase. Choroid epithelial cells synthesize numerous proteins such as transthyretin, transferrin, ceruloplasmin, cytokines and growth factors, e.g. TGFα, TGFβ, bFGF, TNF, and IGF II (21, 22). Protein synthesis varies according to species. The CPs actively transport from blood to CSF numerous molecules such as folate, glucose, vitamins B6, B12, C and probably vitamin E (23, 24). They are able to reabsorb and eliminate from CSF various by-products of brain metabolism, i.e., organic anions and cations (25, 26).

Immunohistochemistry has shown that epithelial cells are polarized. OATP1 (organic anion transport polypeptide) is located at the apical pole while OATP2 is basolateral. P-glycoprotein (MDR gene product) is located at the apical pole while MRP (multidrug resistant protein) (27) and GLUT1 (glucose transporter) (28) are situated basolaterally.

3.3. Composition of cerebrospinal fluid

CSF composition is different from plasma (29), but similar to brain interstitial fluid (30). CSF pH is slightly acid. Compared to plasma, the levels of Na^+, K^+, Ca^{++}, HCO_3^-, proteins and glucose are lower, but Cl^- and Mg^{++} levels are higher (29). Folate levels are 2 to 3 times higher in CSF than in plasma (23). Transthyretin (TTR) represents 25 % of proteins synthesized by CP and 5 % of CSF proteins. The CSF level of TTR is lower than plasma, i.e., 15 mg/ml vs. 200. CSF TTR is almost exclusively synthesized by CP (31). These data support the idea that CSF is not a plasma ultrafiltrate but an active secretion by CP.

4. AGEING OF CHOROID PLEXUS

4.1. Anatomical modifications in ageing

Reports of physiological CP ageing have described epithelial atrophy, weight increase (32) and slightly different modifications according to species.

In elderly rats (33), epithelial cells lose 15 % of their normal height. At 6 mo of age, they are 12.5 μm high. At 18 mo, choroid cells have an average height of 11.5 μm and their morphology is modified, presenting a dome-like appearance. By 30 mo, the epithelial cells are flattened (only 10.5 μm high) and have an irregular and elongated nucleus, significantly shortened microvilli and lipid vacuoles. Concomitantly, there is an irregular fibrosis of stroma and thickening of basement membranes. The epithelial basement membrane (EBM) is typically 97 nm

thick in 6 mo-old-rats, and becomes 214 nm thick by 30 mo. The endothelial basement membrane thickens similarly, however in lower proportions: 73 nm at 6 mo and 87 nm at 30 mo (33). In elderly mice, the number of dark epithelial cells increases, and the size of microvilli decreases (34).

In man, the height of epithelial cells decreases with age by about 10 %. On average, they are 15 µm high in newborns and 13.7 µm in the elderly (35). Cytoplasm of old epithelial cells contains fibrillar inclusions called Biondi bodies and lipofuchsin deposits, the presence of which probably alters cell function (36). Nuclei are irregular and flattened, and the basement membrane is thickened. EBM appears regular in newborns and is 95 nm thick, but it becomes irregular and thickened, i.e., 116 nm, in aged humans (35). The stroma is thicker and contains collagen fibers, hyaline bodies, calcifications and psammomas. Sometimes large and/or numerous cystic formations can be seen. Arterial walls are also thicker, at media and adventia levels, the elastic fibers being fragmented (37).

4.2. Functional alterations in ageing

CPs synthesize numerous enzymes, e.g., the lactic dehydrogenase and succinate-dehydrogenase involved in anaerobic glycolysis and the Krebs cycle. In ageing rats, the dehydrogenase activities decrease, respectively, by 9 and 26 % (38) thereby altering metabolisms dependent on glucose catabolism. The number of epithelial cells deficient in cytochrome C oxidase, a mitochondrial enzyme, increases with age thereby decreasing the cell production of ATP by altering the respiratory mitochondrial chain (39). Reductions in ageing rats also occur in Na^+K^+-ATPase (40) as well as the Na^+K^+-$2Cl^-$ cotransporter (41).

The anatomic and enzymatic modifications of CPs related to ageing are probably responsible for the drastic diminution of CSF secretion. In animal models, CSF secretion decreases as much as 45 % during ageing. In rats, it has been evaluated as 1.2 µl/min at 3 mo and 0.65 µl/min at 30 mo (42). In young sheep, CSF secretion has been measured as 125 µl/min/g CP and in old sheep as 72.5 µl/min/g CP (32). Due to the decreasing secretion and the simultaneously increased CSF volume caused by brain atrophy, the CSF turnover takes longer (42) in elderly rats (7.9 h) than in young rats (2.2 h). In man, CSF secreted volume diminishes with age, from 0.41ml/min at 28 yr of age to 0.19 ml/min at 77 yr (43). As the result of cerebral atrophy due to ageing (44), the turnover of CSF is estimated to occur 6 times a day in young adults compared to 1.7 times daily in elderly subjects.

4.3. Modification of cerebrospinal fluid in ageing

During ageing CSF composition does not vary much. Simultaneous, balanced alterations of the synthesis, secretion, reabsorption, and transportation functions of CP might explain the stable concentration of CSF components in healthy, elderly subjects. Nevertheless, leptomeninges fibrosis (45) and decreased secretion might alter the epuration capacities of CSF. For example, in rats, clearance of β-amyloid protein injected intraventricularly decreases considerably with age, from 10.4 µl/min at 3 mo to 0.71 µl/min at 30 mo; consequently, the amyloid protein brain content increased significantly (7 % at the end of CSF perfusion in young rats and 49 % in old animals (42)). Therefore, the increase of β1-40 and β1-42 amyloid protein levels in elderly humans could be related to decreased clearance from the CNS.

The CSF concentration of some molecules synthesized by CP (TTR) or transported from plasma and of high molecular weight (α2-macroglobulin, IgG) increases slightly with age. This could be due to altered clearance rather than compromised brain-barrier permeability (42, 46, 47). Overall, however, these slightly-modified transport phenomena do not seem to disturb brain homeostasis in healthy elderly subjects.

CP demonstrates a capacity for sequestering toxic heavy metals and metalloid ions (48). For instance, a patient with argyria had silver deposits in the basal lamina of CP, but the brain was free of silver (49). Moreover, in hemochromatosis, MRI detected an iron overloading in CP (50).

5. CHOROID PLEXUS AND ALZHEIMER'S DISEASE

5.1. Anatomical alterations in Alzheimer's disease

In AD, CSF composition is considered subnormal for common biological parameters (2). However, some molecules such as pyruvate and lactate are at elevated levels, which favours a brain oxidative phosphorylation deficit (51). Other substances are decreased suggesting anatomical and/or functional alterations of the CPs.

CPs present abnormalities similar to those observed in ageing, although greatly enhanced. Epithelial atrophy is significant: cell height decreases by 22 % to 10.5 µm compared to controls of the same age (13.7 µm), and by 30 % compared to newborns (15 µm) (35). Epithelial cells contain numerous lipofuchsin vacuolaes and Biondi bodies. The percentage of epithelial cells containing Biondi bodies is significantly increased in AD patients (36). There are also numerous Biondi inclusions in the ependymal layer (52). Epithelial basement membranes are very irregular and thickened; their thickness increases by 28 % compared to controls of the same age, 352 nm vs. 274 nm, and by 270 % compared to newborns (95 nm) (35). Stroma of villi is irregularly fibrotic; its thickness can reach several tenths of a µm (35, 53). CP contains vessels with thickened walls, hyaline bodies, calcifications, and psammomas mainly at the glomus level. Immunohistology reveals many linear deposits of IgG, IgM and C1q along the epithelial basement membrane suggesting intervention of immunological processes (54).

5.2. Cerebrospinal fluid in Alzheimer's disease

These anatomical abnormalities suggest a decrease of functional capacities of CPs. According to their localisation, secretory capacities are difficult to appreciate but they seem to decrease drastically. Indeed, during

isotopic scinticisternographies, there is a major delay of reabsorption as well as transitory ventricular contamination phenomena which intimates a decrease in CSF turnover (55-57) and epuration capacities. CSF production has been measured (58) by using an intraventricular catheter in patients with Parkinson's disease or AD. The mean CSF production rate decreased significantly in AD patients. It is worth noting that in Down's syndrome the CSF production does not differ from that of normal subjects (59). Due to cerebral atrophy and the consequent ventricular dilatation, the CSF turnover is about 36 h in AD patients.

The decrease of CSF production could favor the glycation of proteins and the formation of β-amyloid protein oligomers. AD brain contains 3 times more advanced glycation products (AGE) than controls (60). Diffuse deposits of amyloid protein, senile plaques and NFT are known to contain AGE (6). Glycation of proteins promotes their aggregation, the polymerisation of tau proteins with formation of filaments, and protein β-amyloid protein fibrillotransformation (60, 61). *In vitro*, fibrillotransformation is accelerated in the presence of AGE, probably through initiation of nucleation phenomena (62). The first stage of protein glycation induces spontaneous formation of Amadori bodies in a few hours or days. In ageing, levels of CSF proteins, glucose and Amadori bodies remain stable, the CSF turnover reduction being insufficient to increase protein glycation. On the contrary, in AD the Amadori bodies increase significantly while CSF protein and glucose levels remain stable. Increased contact time between glucids and proteins relates to a substantial decrease of CSF turnover (63).

Amyloid protein clearance, which is significantly reduced in elderly animals (42), is still unknown in AD patients. Amyloid protein monomers do not seem to have brain toxicity. *In vitro*, they self-aggregate and form spontaneously soluble, apparently neurotoxic oligomers, the level of which increases with time (64). Amyloid protein oligomers can also be formed and secreted from cells (65). The brain from AD patients contains 12 times as many oligomers as controls (4). AD severity appears to be correlated more to amyloid protein oligomer level than to senile plaque number (64). The decrease of CSF turnover, the increase of protein glycation and the diminution of β-amyloid protein clearance could altogether induce oligomer formation and retention.

CPs are involved in the regulation and secretion of several growth factors that can affect CSF dynamics (22). Experimentally, ventricular enlargement and fibrosis of arachnoid villi were produced in adult rats by infusing bFGF (66). In AD, hydrocephalus ex vacuo and arachnoid fibrosis are usual (67) and increased bFGF levels in brain parenchyma could be due to local sequestration (22).

Importantly, epithelial atrophy suggests a decrease of enzymatic activities due to altered protein synthesis. *In vitro*, CSF inhibits amyloid protein fibrillotransformation probably through sequestration (68). Levels of CSF transthyretin, a CP synthesized molecule which associates with β-amyloid protein to form complexes (69), are 13 % lower in AD (47, 70, 71).

Transport functions are also impaired. CSF folate and vitamin B12 are actively transported by CP to CSF and are involved in methylation processes, particularly of myelin, proteins, phospholipids, nucleic acids, catechnolamines and acetylcholine. The levels of these two vitamins are significantly lower in AD (72-74). In the plasma, homocysteine levels were correlated with vitamin B12 and folate depletion. Homocysteine, which mediates lipid peroxidation and increases the production of toxic HNE, has an increased level in AD CSF (74).

Ascorbic acid and α-tocopherol levels, the two major scavengers of free radicals of CSF, are decreased in AD (75, 76). These abnormalities could explain the major oxidative stress of AD brain (5) and the decrease of CSF antioxidant capabilities in AD (76).

The reduced levels of these molecules, in the face of decreased CSF turnover, means an even more diminished availability for brain. These abnormalities could cause methylation problems, increase oxidative stress and lipid peroxidation, modify amyloid protein clearance, and facilitate tau protein polymerisation, as well as formation of amyloid protein oligomers and fibrilloformation.

6. CONCLUSION

CP tissues, despite their small weight, appear to play a major role in brain homeostasis. Ageing of CP is characterized by epithelial atrophy, a decrease of enzymatic activities, thickening of basement membranes (mainly epithelial BM), and irregular stroma fibrosis. These morphological modifications imply untoward effects on brain functions, in part due to compromised CSF epuration capacities.

During AD, these ultrastructural abnormalities are significantly augmented. They are frequently associated with immunoglobulins and C1q deposits along epithelial basement membrane, reflecting immunological processes. These morphological modifications are associated with diminished secretion, synthesis and transport functions, inducing a slower turnover of CSF, a decrease of protein amyloid sequestration, an increase of glycation processes and of oxidative stress. Altogether, these conditions may facilitate fibrillotransformation of amyloid protein and tau protein polymerisation.

Other ageing-induced alterations, such as leptomeningeal and arachnoid villi fibrosis (67), capillary network modifications induced by brain hypovascularisation (77), and CP hypoperfusion secondary to heart failure (78), may also potentiate CP dysfunction and its consequences.

7. ACKNOWLEDGEMENTS

8. REFERENCES

1. Selkoe D. Translating cell biology into therapeutic advances in Alzheimer's disease. *Nature* 399 S, A23-A31 (1999)

2. Mirra S.S., A. Heyman, D. McKeel, S.M. Sumi, B.J. Crain, L.M. Brownlee, F.S. Vogel, J.P. Hughes, G. van Belle & L. Berg. The consortium to establish a registry for Alzheimer's disease (CERAD). Part II. Standardization of the neuropathologic assessment of Alzheimer's disease. *Neurology* 41, 479-486 (1991)

3. Davies P. A very incomplete comprehensive theory of Alzheimer's disease. *Ann NY Acad Sci* 924, 8-16 (2000)

4. Kuo Y.M., M.R. Emmerling, C. Vigo-Pelfrey, T.C. Kasunic, J.B. Kirkpatrick, G.H. Murdoch, M.J. Ball & A.E. Roher. Water-soluble Aβ(N-40, N-42) oligomers in normal and Alzheimer disease brains. *J Biol Chem* 271, 4077-4081 (1996)

5. Lovell M.A., S.P. Gabbita & W.R. Markesbery. Increased DNA oxidation and decreased levels of repair products in Alzheimer's disease ventricular CSF. *J Neurochem* 72, 771-776 (1999)

6. Smith M.A., L.M. Sayre, V.M. Monnier & G. Perry. Oxidative posttranslational modifications in Alzheimer disease. A possible pathogenic role in the formation of senile plaques and neurofibrillary tangles. *Mol Chem Neuropath* 28, 41-48 (1996)

7. Katzman R. Alzheimer's disease as an age-dependent disorder. *Ciba Foundation Symposium* 134, 69-85 (1988)

8. Soto C., G.P. Saborio & B. Permanne. Inhibiting the conversion of soluble amyloid-β peptide into abnormally folded amyloidogenic intermediates: relevance for Alzheimer's disease therapy. *Acta Neurol Scand* S176, 90-95 (2000)

9. Johanson C.E. Ventricles and cerebrospinal fluid. In: Neuroscience in Medicine. Eds : Conn P.M., Lippincott J.B., Philadelphia, 171-196 (1995)

10. Zheng W. Choroid plexus and metal toxicity. In: Toxicology of metals. Eds: Chang L.W., CRC Lewis Publishers, London, 609-626 (1996)

11. Dohrmann G.J. The choroid plexus: a historical review. *Brain Res* 18, 197-218 (1970)

12. Cornford E.M., J.B. Varesi, S. Hyman, R.T. Damian & M.J. Raleigh. Mitochondrial content of choroid plexus epithelium. *Exp Brain Res* 116, 399-405 (1997)

13. Dohrmann G.J. Dark and light epithelial cells in the choroid plexus of mammals. *J Ultrastruct Res* 32, 268-273 (1970)

14. Serot J.M., M.C. Béné, B. Foliguet & G.C. Faure. Monocyte-derived IL-10-secreting dendritic cells in choroid plexus epithelium. *J Neuroimmunol* 105, 115-119 (2000)

15. Nilsson C., R. Ekman, M. Lindvall-Axelsson & C. Owman. Distribution of peptidergic nerves in the choroid plexus, focusing on coexistence of neuropeptide Y, vasoactive intestinal polypeptide and peptide histidine isoleucine. *Regul Pept* 27, 11-26 (1990)

16. Faraci F.M., W.G. Mayhan & D.D. Heistad. Effect of serotonin on blood flow to the choroid plexus. *Brain Res* 478, 121-126 (1989)

17. Vannucci S.J., R.R. Clark, E. Koehler-Stec, K. Li, C.B. Smith, P. Davies, F. Maher & I.A. Simpson. Glucose transporter expression in brain: relationship to cerebral glucose utilization. *Dev Neurosci* 20, 369-379 (1998)

18. Kim K.S., J. Wegiel, V. Sapienza, J. Chen, H. Hong & H.M. Wisniewski. Immunoreactivity of presenilin-1 in human, rat, and mouse brain. *Brain Res* 757, 159-163 (1997)

19. Johnson M.D., J.M. Kamso-Pratt, W.O.Jr Whetsell & R.B. Pepinsky. Lipocortin-1 immunoreactivity in the normal human central nervous system and lesions with astrocytosis. *Am J Clin Pathol* 92, 424-429 (1989)

20. Gander R., P. Eller, S. Kaser, I. Theurl, D. Sauper, A. Ritsch, J.R. Patsch & B. Foger. Molecular characterization of rabbit phospholipid transfer protein: choroid plexus and ependyma synthesize high levels of phospholipid transfer protein. *J Lipid Res* 43, 636-645 (2002)

21. Aldred A.R., C.M. Brack & G. Schreiber. The cerebral expression of plasma protein genes in different species. *Comp Biochem Physiol* 111B: 1-15 (1995)

22. Stopa E.G., T. M. Berzin , S. Kim, P. Song, V. Kuo-Leblanc, M. Rodriguez-Wolf, A. Baird & C.E. Johanson. Human choroid plexus growth factors: what are the implications for CSF dynamics in Alzheimer's disease. *Exp Neurol* 167, 40-47(2001)

23. Spector R. Vitamin homeostasis in the central nervous system. *New Eng J Med* 296, 1393-1398 (1977)

24. Fand I. & W.P. McNally. Whole-body localization of 14C-tocopherylacetate in the rat following oral administration. *Arch Int Pharmacodyn Ther* 250, 4-17 (1981)

25. Ghersi-Egea J.F., B. Leininger-Muller, G. Suleman, G. Siest & A. Minn. Localization of drug-metabolizing enzyme activities to blood-brain interfaces and circumventricular organs. *J Neurochem* 62, 1089-1096 (1994)

26. Stoll J., K.C. Wadhwani & Q.R. Smith. Identification of the cationic amino acid transporter (System y+) of the rat blood-brain barrier. *J Neurochem* 60, 1956-1959 (1993)

27. Gao B., P.J. Meier. Organic anion transport across the choroid plexus. *Microsc Res Tech* 52, 60-64 (2001)

28. Dwyer K.J. & W.M. Pardridge. Developmental modulation of blood-brain barrier and choroid plexus GLUT1 glucose transporter messenger ribonucleic acid and immunoreactive protein in rabbits. *Endocrinology* 132, 558-565 (1993)

29. Segal M.B. The choroid plexuses and the barriers between the blood and the cerebrospinal fluid. *Cell Mol Neurobiol* 20, 183-196 (2000)

30. Felgenhauer K. The blood-brain barrier redefined. *J Neurol* 233, 193-194 (1986)

31. Weisner B. & H.J. Roethig. The concentration of prealbumin in cerebrospinal fluid (CSF), indicator of CSF circulation disorders. *Eur Neurol* 22, 96-105 (1983)

32. Wilson M.R., J.E. Preston, S.A. Thomas & M.B. Segal. Altered cerebrospinal fluid secretion rate and ^{125}I-labelled β-amyloid transport in the aged sheep isolated perfused choroid plexus. *J Physiol* 515P, 7-8P (1999)

33. Serot J.M., M.C. Béné, B. Foliguet & G.C. Faure. Choroid plexus and ageing: a morphometric and ultrastructural study. *Eur J Neurosci* 14, 794-798 (2001)

34. Sturrock R.R. An ultrastructural study of the choroid plexus of aged mice. *Anat Anz* 165, 379-385 (1988)

35. Serot J.M., M.C. Béné, B. Foliguet & G.C. Faure. Morphological alterations of the choroid plexus in late-onset Alzheimer's disease. *Acta Neuropathol* 99, 105-108 (2000)

36. Wen G.Y., H.M. Wisniewski & R.J. Kascsak. Biondi ring tangles in the choroid plexus of Alzheimer's disease and normal aging brains: a quantitative study. *Brain Res* 832, 40-46 (1999)

37. Shuangshoti S. & M.G. Netsky. Human choroid plexus: morphologic and histochemical alterations with age. *Am J Anat* 128, 73-96 (1970)

38. Ferrante F. & F. Amenta. Enzyme histochemistry of the choroid plexus in old rats. *Mech Ageing Dev* 41, 65-72 (1987)

39. Cottrell D.A., E.L. Blakely, M.A. Johnson, P.G. Ince, G.M. Borthwick & D.M. Turnbull. Cytochrome oxidase deficient cells accumulate in the hippocampus and choroid plexus with age. *Neurobiol Aging* 22, 265-272 (2001)

40. Kvitnitskaia-Ryzhova T.I. & A.L. Shkapenko. A comparative ultracytochemical and biochemical study of the ATPases of the choroid plexus in aging. *Tsitologiia* 34, 81-87 (1992)

41. Preston J.E. Age-related reduction in rat choroid plexus chloride efflux and CSF secretion rate. *Soc Neurosci* 25, P697-698 (1999)

42. Preston J.E. Ageing choroid plexus-cerebrospinal fluid system. *Microsc Res Tech* 52, 31-37 (2001)

43. May C., J.A. Kaye, J.R. Atack, M.B. Schapiro, R.P. Friedland & S.I. Rapoport. Cerebrospinal fluid production is reduced in healthy aging. *Neurology* 40, 500-503 (1990)

44. Tanna N.K., M.I. Kohn, D.N. Horwich, P.R. Jolles, R.A. Zimmerman, W.M. Alves & A. Alavi. Analysis of brain and cerebrospinal fluid volumes with MR Imaging: Impact on PET data correction for atrophy. Part II. Aging and Alzheimer dementia. *Radiology* 178, 123-130 (1991)

45. Albeck M.J., C. Skak, P.R. Nielsen, K.S. Olsen, S.E. B∅rgesen & F. Gjerris. Age dependency of resistance to cerebrospinal fluid outflow. *J Neurosurg* 89, 275-278 (1998)

46. Kunicki S., J. Richardson, P.D. Mehta, K.S. Kim & E. Zorychta. The effects of age, apolipoprotein E phenotype and gender on the concentration of amyloid-β (Aβ) 40, Aβ42, apolipoprotein E and transthyretin in human cerebrospinal fluid. *Clin Biochem* 31, 409-415 (1998)

47. Serot J.M., D. Christmann, T. Dubost & M. Couturier. Cerebrospinal fluid transthyretin: aging and late onset alzheimer's disease. *J Neurol Neurosurg Psychiatry* 63, 506-508 (1997)

48. Zheng W. Toxicology of choroid plexus: special reference to metal-induced neurotoxicities. *Microsc Res Tech* 52, 89-103 (2001)

49. Goebel H.H. & J. Muller. Ultrastructural observations on silver deposition in the choroid plexus of a patient with argyria. *Acta Neuropathol* 26, 247-251 (1973)

50. Kira R., S. Ohga, H. Takada, K. Gondo, F. Mihara & T. Hara. MR choroid plexus sign of iron overload. *Neurology* 55, 1340 (2000)

51. Parnetti L., Reboldi G.P. & V. Gallai. Cerebrospinal fluid pyruvate levels in Alzheimer's disease and vascular dementia. *Neurology* 54, 735-737 (2000)

52. Miklossy J., R. Kraftsik, O. Pillevuit, D. Lepori, C. Genton & F.T. Bosman. Curly fiber and tangle-like inclusions in the ependyma and choroid plexus--a pathogenetic relationship with the cortical Alzheimer-type changes? *J Neuropathol Exp Neurol* 57, 1202-1212 (1998)

53. Jellinger K. Neuropathological aspects of dementias resulting from abnormal blood and cerebrospinal fluid dynamics. *Acta Neurol Belg* 76, 83-102 (1976)

54. Serot J.M., M.C. Béné & G.C. Faure. Comparative immunohistochemical characteristics of human choroid plexus in vascular and Alzheimer's dementia. *Hum Pathol* 25, 1185-1190 (1994)

55. Coblentz J.M., S. Mattis, L.H. Zingesser, S.S. Kasoff, H.M. Wisniewski & R. Katzman. Presenile dementia.

Clinical aspects and evaluation of cerebrospinal fluid dynamics. *Arch Neurol* 29, 299-308 (1973)

56. Serot J.M., A. Bertrand, F. Penin, C. Hocquard, M.C. Mietkiewicz & G. Cuny. La cysternographie isotopique dans les détériorations mentales et les démences du sujet âgé. *Med Hyg* 36, 1928-1930 (1978)

57. Brusa G., F. Claudiani, A. Piccardo, N. Pizioo & R. Stoehr. Scinticisternography in presenile and senile degenerative disease. *Ital J Neurol Sci* 11, 43-47 (1990)

58. Silverberg G.D., G. Heit, S. Huhn, R.A. Jaffe, S.D. Chang, H. Bronte-Stewart, E. Rubenstein, K. Possin & T.A. Saul. The cerebrospinal fluid production rate is reduced in dementia of the Alzheimer's type. *Neurology* 57, 1763-1766 (2001)

59. Atack J.R., S.I. Rapoport & M.B. Schapiro. Cerebrospinal fluid production is normal in Down syndrome. *Neurobiol Aging* 19, 307-309 (1998)

60. Vitek M.P., K. Battacharaya, J.M. Glendening, E. Stopa, H. Vlassara, R. Bucala, K. Manogue & A. Cerami. Advanced glycation end products contribute to amyloidosis in Alzheimer disease. *Proc Natl Acad Sci USA* 91, 4766-4770 (1994)

61. Troncoso J.C., A. Costello, A.L. Watson Jr & G.V.W. Johnson. *In vitro* polymerization of oxidized tau into filaments. *Brain Res* 613, 313-316 (1993)

62. Münch G., R. Schinzel, C. Loske, A. Wong, N. Durany, J.J. Li, H. Vlassara, M.A. Smith, G. Perry & P. Riederer. Alzheimer's disease--synergistic effects of glucose deficit, oxidative stress and advanced glycation endproducts. *J Neural Transm* 105, 439-461 (1998)

63. Shuvaev V.V., I. Laffont, J.M. Serot, J. Fujii, N. Taniguchi & G. Siest. Increased protein glycation in cerebrospinal fluid of Alzheimer's disease patients. *Neurobiol Aging* 22, 397-402 (2001)

64. Klein W.L., G.A. Krafft & C.E. Finch. Targeting small Aβ oligomers: the solution to an Alzheimer's disease conundrum? *Trends Neurosci* 24, 219-224 (2001)

65. Walsh D.M., I. Klyubin, J.V. Fadeeva, M.J. Rowan & D.J. Selkoe. Amyloid-β oligomers: their production, toxicity and therapeutic inhibition. *Biochem Soc Trans* 30,552-557 (2002)

66. Johanson C.E., J. Szmydynger-Chodobska J, A. Chodobski, A. Baird, P. McMillan & E.G. Stopa. Altered formation and bulk absorption of cerebrospinal fluid in FGF-2-induced hydrocephalus. *Am J Physiol* 277, R263-R271 (1999)

67. Bellur S.N, V. Chandra & L.W. McDonald. Arachnoid cell hyperplasia. Its relationship to aging and chronic renal failure. *Arch Pathol Lab Med* 104, 414-416 (1980)

68. Schwarzman A.L. & D. Goldgaber. Interaction of transthyretin with amyloid β-protein: binding and inhibition of amyloid formation. *Ciba Foundation Symposium* 199, 146-164 (1996)

69. Tsuzuki K., R. Fukatsu, H. Yamaguchi, M. Tateno, K. Imai, N. Fujii & T. Yamauchi. Transthyretin binds amyloid beta peptides, Abeta1-42 and Abeta1-40 to form complex in the autopsied human kidney-possible role of transthyretin for Abeta sequestration. *Neurosci Lett* 281, 171-174 (2000)

70. Riisoen H. Reduced prealbumin (transthyretin) in CSF of severely demented patients with Alzheimer's disease. *Acta Neurol Scand* 78, 455-459 (1988)

71. Dorta-Contreras A.J., M. Barshatzky, E. Noris-Garcia & T. Serrano-Sánchez. Niveles de transtirretina en suero y en liquido cefalorraquideo apoyan la hipótesis nutrio-virica de la neuropatia epidémica cubana. *Rev Neurol 31*, 801-804 (2000)

72. Ikeda T., Y. Furukawa, S. Mashimoto, K. Takahashi & M. Yamada. Vitamin B12 levels in serum and cerebrospinal fluid of people with Alzheimer's disease. *Acta Psychiatr Scand* 82, 327-329 (1990)

73. Serot J.M., D. Christmann, T. Dubost, M.C. Béné & G.C. Faure. CSF-folate levels are decreased in late-onset AD patients. *J Neural Transm* 108, 93-99 (2001)

74. Selley M.L., D.R. Close & S.E. Stern. The effect of increased concentrations of homocysteine on the concentration of (E)-4-hydroxy-2-nonenal in the plasma and cerebrospinal fluid of patients with Alzheimer's disease. *Neurobiol Aging* 232, 383-388 (2002)

75. Tohgi H., T. Abe, M. Nakanishi, F. Hamato, K. Sasaki & S. Takahashi. Concentrations of ? -tocopherol and its quinone derivative in cerebrospinal fluid from patients with vascular dementia of the Binswanger type and Alzheimer type dementia. *Neurosci Lett* 174, 73-76 (1994)

76. Schippling S., A. Kontush, S. Arlt, C. Buhmann, H.J. Stürenburg, U. Mann, T. Müller-Thomsen & U. Beisiegel. Increased lipoprotein oxidation in Alzheimer's disease. *Free Rad Biol Med* 28, 351-360 (2000)

77. De La Torre J.C. Impaired cerebromicrovascular perfusion: summary of evidence in support of its causality in Alzheimer's disease. *Ann NY Acad Sci* 924, 136-152 (2000)

78. Rubenstein E. Relationship of senescence of cerebrospinal fluid circulatory system to dementias of the aged. *Lancet* 351, 283-285 (1998)

Send correspondence to: Dr Jean-Marie Serot, Laboratoire d'Immunologie, BP 184, 54500 Vandoeuvre les Nancy, France, Tel: 33-383 68 36 60, Fax: 33-383 44 60 22, E-mail: faure@grip.u-nancy.fr