CSF concentration gradients of monoamine metabolites in patients with hydrocephalus

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Abstract

Concentration gradients of homovanillic acid (HVA), 5-hydroxyindoleacetic acid (5-HIAA), and 3-methoxy-4-hydroxyphenylglycol (MHPG), were assessed in 762 successive CSF fractions (2 ml lumbar CSF) from 15 patients with the adult hydrocephalus syndrome (AHS) and 11 patients with hydrocephalus of other causes (mixed group). A mean volume of 49.6 (SD 11.8) ml CSF was removed in the AHS group and 56.4 (10.2) ml in the mixed group. The CSF was collected with a specially designed carousel fraction collector and the corresponding CSF dynamics were continuously registered by a constant pressure CSF infusion method. Pronounced gradients in CSF HVA and CSF 5-HIAA were seen in both patient groups in the first 25 ml of CSF removed. The concentration curves levelled off, despite the removal of larger amounts of CSF and stabilised at about twice the initial concentrations. This phenomenon has not been described before. Concentrations of HVA and 5-HIAA in the first CSF fraction correlated strongly with concentrations in fractions up to about 40 ml. A positive correlation between the first fraction of CSF HVA and CSF 5-HIAA concentrations and CSF outflow conductance was found in the AHS group. There was no gradient in MHPG. It is suggested that the rostrocaudal gradients in CSF HVA and 5-HIAA may be explained by a downward flow of CSF along the spinal cord with absorption of metabolites occurring during passage. Mixing of CSF from different CSF compartments, extraventricular production sites of CSF, clearance of metabolites to venous blood or extracellular fluid, and CSF outflow conductance are probably important determinants of the plateau phase in patients with hydrocephalus. It is concluded that lumbar CSF does not exclusively reflect the concentrations of HVA, 5-HIAA, or MHPG in the ventricles. It should be noted that these results obtained in patients with hydrocephalus may not be applicable to other groups of patients or normal subjects.

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Many neurological and psychiatric disorders

are associated with disturbances in the neurotransmission of biogenic amines.¹⁻⁶ Treatment of certain disorders is performed by stimulating neurotransmitter synthesis or by stimulating or blocking presynaptic or postsynaptic monoamine receptors. The monoamine systems are located at different anatomical sites in the human brain. The highest concentrations of dopamine are found in the basal ganglia, of noradrenaline in the hypothalamus, and of serotonin in the subcortical regions.⁷ There is a correlation between transmitter concentration at these specific topographic origins and ventricular CSF.^{7,8} In humans, however, the use of ventricular CSF, brain biopsy, or microdialysis of the extracellular fluid are possible only during certain neurosurgical operations or at necropsy.

Lumbar puncture is the most important and widely used diagnostic tool in the study of monoamine metabolite concentrations and findings in lumbar CSF are commonly assumed to be similar to those in ventricular CSF. This assumption is supported by rostrocaudal CSF gradients⁹⁻¹²—that is, the finding of decreasing concentrations of substances in the spinal subarachnoid space the further from the brain the CSF is removed. Rostrocaudal gradients of HVA and 5-HIAA, and to a lesser extent, of MHPG, have been described (table 1).¹³⁻²⁵ Bulk flow of CSF is directed from the ventricles towards the cerebral subarachnoid space via the basal cisternae but the direction and amount of spinal CSF flow are unclear, and CSF may even be a “stagnant backwater.”²⁶ Thus the assumption that monoamine metabolite concentrations found in lumbar CSF represent those in ventricular CSF may not hold true.

In this study, a method for standardised sampling of large volumes of lumbar CSF was used in a prospective series of 26 patients, investigated because of hydrocephalus. The rostrocaudal gradients for HVA, 5-HIAA, and MHPG are presented as well as factors that may contribute to differences in the metabolite concentrations between ventricular and lumbar CSF. Different CSF spinal flow options are discussed.

Materials and methods

PATIENTS AND CLINICAL INVESTIGATION

During February 1990–May 1991, 146 patients were studied in a CSF hydrodynamic investigation²⁷ at the neurological department. Of these, 27 (17 men, 10 women; mean age (SD) 72.6 (6.4) years) satisfied the inclusion

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Table 1 Earlier published studies concerning CSF gradients of monoamine metabolites. Monoamine metabolite concentrations are presented as a ratio between the concentration in the first and last CSF fraction

Reference No	CSF removed (ml)	Fractions (No)	ml/fraction	Bedrest	Fasting	Diagnosis	MHPG (ratio)	5-HIAA (ratio)	HVA (ratio)	Body position	Patient (No)	Others
13	40	4	10	NM	NM	VND	1.0	1.2	1.7	Sitting	9	
14	12	4	3	Yes	Yes	VPD	≈1.0	≈1.7	≈1.9	Sitting	28	
15	60	2	6	NM	Yes	VND	—	1.3	—	NM	72	PEG
16	a 20 b 20	10	2	NM	NM	MS + controls	1.1	1.75	1.5	LRP	40	
17	38	3	5	Yes	NM	VND	1.1	—	—	LRP	15	
18	40–50	5	10	NM	NM	VND	—	Increase	Increase	NM	8	Neurosurgical patient during anaesthesia
19	15–24	4	2.5	NM	NM	VND, VPD	—	Increase	Increase	Sitting	3	PEG
20	42	7	6	Yes	Yes	VND	—	2.4	Increase	NM	14	PEG
21	19	3	2	Yes	NM	VPD	—	1.8	1.8	NM	18	
22	36	3	12	Yes	NM	Schizophrenia	1.1	1.56	1.87	LRP	NM	
23	15	2	8	NM	NM	VND	—	2.1	3.1	Sitting	10	
24	NM	2	NM	Yes	No	VND	—	2	—	NM	NM	PEG
25	NM	2	3–4	NM	Yes	VND	—	≈1.4	≈1.5	Sitting	35	PEG
Present study	28–82	14–41	2	No	No	Hydrocephalus	1	2	2	Lying, Supine	26	

NM = Not mentioned; VND = various neurological diseases; VPD = various psychiatric diseases; LRP = lateral recumbent position; PEG = pneumoencephalography; MS = multiple sclerosis.

criteria for this study—namely, (1) hydrocephalus verified by CT; (2) clinical suspicion of the adult hydrocephalus syndrome (AHS); (3) no obvious contraindications to an extensive CSF tap.

At the time of inclusion, 24 of the 27 patients had disturbance of gait, alone or in combination with dementia or incontinence, and two had focal neurological signs, two complained of headache, one of vertigo, and two were asymptomatic.

All patients underwent the following laboratory investigations to determine the diagnosis and to exclude reversible and treatable causes of dementia: full blood count; urine analysis; erythrocyte sedimentation rate; serum electrolytes; tests of hepatic, renal, and thyroid function; blood glucose; antinuclear antibodies; serum cholesterol and triglyceride concentrations; syphilis and Borrelia serology; plasma protein electrophoresis; serum B12/folic acid; CSF protein and cell count. CT and, usually, MRI were also included.

After investigation, the 27 patients were categorised as follows: the adult hydrocephalus syndrome⁵ (gait disturbance, dementia and/or incontinence, dilated ventricles, and no obvious cause such as subarachnoid haemorrhage, meningitis, or trauma), 15 patients; multi-infarction dementia,²⁸ four; Alzheimer dementia (NINDS-ARDRAS criteria),²⁹ one; idiopathic intracranial hyperten-

sion (increased CSF pressure, unknown cause),³⁰ one; alcoholic dementia (daily abuse for many years, no other cause), one; hydrocephalus of unknown cause, five. According to the diagnosis, patients were categorised as two groups: AHS (15 patients) or mixed (12 patients) groups. One patient in the mixed group was excluded because of a non-functioning lumbar puncture needle.

Table 2 summarises the characteristics of the two groups. There were no significant differences concerning physical, clinical, or CSF hydrodynamic variables between the AHS and mixed patient groups (Student's *t* test). Two patients were taking benzodiazepines, two antidepressants, and another two anticholinergic drugs (terodiline). None was on neuroleptic medication. Data on CSF from these patients did not differ from the rest of the study group.

Consent for all aspects of the study was obtained from all the patients after explanation of the investigation procedure and possible risks.

CSF HYDRODYNAMIC STUDIES

The CSF hydrodynamic studies were performed according to Ekstedt.²⁷ Briefly, at 0800, after 12 hours of bed rest but without fasting, two needles were inserted in the L3-L4 interspace by a very experienced investigator. The patient was then allowed to lie supine with the zero pressure reference level at the cranial sagittal centre. The needles were connected to pressure transducers by means of plastic tubes filled with artificial CSF. At application, loss of CSF was between 0.5–2.0 ml in each patient. Pressure of CSF was continuously recorded via one of the needles. The other needle could be used for infusion of artificial CSF or drainage and CSF sampling via a three way tap (see later). The CSF resting pressure (*P*_d) was measured when the resting recording had been stable for at least 10 minutes. Thereafter, the CSF sampling started (see later). The examination ended with determination of the conductance (*G*_{op}; the inverse of resistance to CSF outflow). This was done by applying a number of different

Table 2 Characteristics of the AHS and mixed patient groups

	AHS (n = 15)	Mixed (n = 11)	All patients (n = 26)
Physical measurements:			
Age (y)	73.6 (3.9)	71.2 (8.8)	72.0 (8.3)
Sex (male/female)	10/5	7/4	17/9
Length (m)	1.7 (0.1)	1.7 (0.1)	1.7 (0.1)
Weight (kg)	72.3 (11.5)	63.4 (14.4)	68.7 (13.2)
Body mass index (kg/m ²)	25.7 (3.4)	22.6 (3.3)	24.4 (3.6)
Clinical variables:			
Duration of disease (months)	27 (18)	35 (19)	31 (19)
Mini-mental state ⁵² (points)	22.9 (5.4)	20.8 (9.2)	21.7 (7.3)
Haschinski ischaemic scale ⁵³ (points)	2.4 (2.1)	4.3 (5.1)	3.3 (3.6)
CSF hydrodynamic variables:			
CSF removal (ml)	49.6 (11.8)	56.4 (10.2)	52.5 (11.5)
CSF pressure (kPa)	1.6 (0.4)	1.8 (0.8)	1.7 (0.6)
Conductance (mm ³ /kPa × s)	9.8 (3.5)	13.7 (7.3)	11.7 (5.5)
CSF formation rate (mm ³ /s)	7.1 (2.1)	8.5 (2.0)	7.7 (2.2)

Values are means (SD).

pressures to the CSF space while recording the resulting inflow of artificial CSF. Thus within a few minutes a stable flow at a stable pressure was obtained. Usually, three different pressure/flow values were sought for each patient. The volume accounting method was used to calculate the pressure/flow relation. The slope for the pressure/flow values is equal to G_{op} .

CSF SAMPLING

The needle was connected to a plastic tube that ended with an injection needle, the point of which was fixed at a height of the zero reference level for the pressure measurements. The point of the needle was adjusted to 1 cm above one of the 12 test tubes of a specially designed carousel fraction collector, placed on a digital precision balance. An electronic device rotated the carousel 1/12 of a turn each time the balance recorded a preset weight increase from the moment of the last change of test tube. A 2 g (= 2 ml) weight increase was used but any value could be chosen. Immediately a test tube was filled, it was placed in the freezer (-70°C). For the first test tube a value equal to the amount of artificial CSF in the plastic tubing was chosen and this fraction of CSF was discarded. The CSF pressure was about 0.2 kPa during the collection period, well below the pressure in the sagittal sinus. Thus when the CSF pressure had stabilised at this level, no CSF could escape through the arachnoidal villi into the sagittal sinus. The rate of CSF outflow thus corresponded to the CSF formation rate.

ASSESSMENT OF MONOAMINE CONCENTRATIONS

Concentrations of 3-methoxy-4-hydroxyphenylglycol (MHPG), homovanillic acid (HVA), and 5-hydroxyindoleacetic acid (5-HIAA) were determined simultaneously by reversed phase isocratic liquid chromatography with electrochemical detection essentially as described elsewhere.^{31,32} Briefly, to each 90 μl of CSF, 10 μl of 1M perchloric acid, 9 mM L-cysteine, and 0.5 mM isoproterenol (internal standard) were added. The samples were vortexed, centrifuged at 48 000 g for 30 minutes at 4°C and 20 μl was then directly injected into the chromatographic system. The isocratic mobile phase, with a flow rate of 0.28 ml/minute consisted of 100 mM Na_2EDTA , 0.334 mM octylsulphate: 5.5/94.5% (v/v) acetonitrile/water at a pH of 2.35. The stationary phase consisted of a 250×2.0 mm stainless steel narrow bore column packed with Nucleosil C-18.5 μm from Macherey-Nagel (Düren, Germany), the column temperature was controlled by means of a plastic jacket coupled to a water thermostat that was set at 25°C . The substances were detected electrochemically with a BAS 3 electrochemical detector (Bioanalytical systems, West Lafayette, IN, USA), operated at a potential of 0.75 mV *v* an Ag/AgCl reference electrode. The detector was coupled to an integrator (model SP 4400, Spectraphysics, San Jose, CA, USA). The coefficient of varia-

tion (CV) showed an average value of 1.8% for standards and 2.1% for CSF samples. A linear response was found for the substances in the range of 0.1–256 pmol ($r > 0.999$). The detection limit, defined as a ratio of peak height to noise greater than 2, varied between 2 and 5 pmol with the equipment used. Variation (CV) showed an average value of 1.8% for standards and 2.1% for CSF samples.

STATISTICAL ANALYSIS

Statistical analyses were conducted with the JMP program for the Macintosh computer.³³ Comparisons of physical, clinical, and CSF hydrodynamic variables (table 2) between the AHS and the mixed groups were evaluated by Student's unpaired *t* test. Linear regression analyses between physical, clinical, and CSF hydrodynamic variables *v* the first CSF fraction of the CSF metabolites were performed where the regressors were tested in the models with a *t* test ($H_0 = 0$) and regarded as statistically significant when the corresponding *p* value was below 0.05. The concentrations of transmitter metabolites were plotted against CSF fractions with 95% confidence intervals.

Results

There were no serious adverse events during the CSF hydrodynamic investigation or removal of CSF. The duration of the CSF pressure recording was 43.0 (SD 10.9) minutes in the AHS and 44.9 (14.9) minutes in the mixed group. The duration of the drain, where the CSF pressure is lowered by passive removal of CSF, was 66.8 (20.4) minutes in the AHS and 63.7 (21.2) minutes in the mixed group. The estimation of CSF formation rate started when the CSF pressure had stabilised at 0.2 (0.2) kPa in both groups. The duration of the CSF formation rate appraisal was 35.4 (16.7) minutes in the AHS and 42.2 (19.7) minutes in the mixed group. 49.6 (11.8) ml CSF was collected in the AHS and 56.4 (10.2) ml in the mixed group. There were no significant differences in these variables between the AHS and mixed groups.

Mean values of CSF homovanillic acid (HVA) concentrations were plotted against CSF fractions in the AHS (fig 1A) and mixed group (fig 1B). CSF concentrations are presented as a ratio of the first CSF fraction to compensate for interindividual variations of initial CSF HVA values. A similar pattern of increasing HVA concentrations was found during the first 25 ml in both the AHS and mixed groups. In subsequent (27–59 ml) fractions, a plateau was reached with no further increase in CSF HVA concentration.

Similarly, increased concentrations of 5-HIAA were seen in the first 25 ml in patients in both the AHS and mixed groups, although no further increase was seen in fraction 27–59 ml (fig 1C,D). No gradient of CSF MHPG concentration was seen in either the AHS or mixed groups (fig 1E,F).

Figure 2A shows the relation between the first fraction of CSF HVA concentration and

Figure 1 CSF gradients of HVA, 5-HIAA, and MHPG in the AHS ($n = 15$) and mixed patient ($n = 11$) groups. Concentrations are presented as a ratio of the first CSF fraction (mean value and 95% confidence intervals).

(A) HVA in AHS group; the median and ranges in CSF fraction Nos 1 and 15 were 1: 200.0 (100.0–919.0) nmol/l and 15: 432.0 (329–991) nmol/l; (B) HVA in mixed group; 1: 239.5 (91.0–469.9) nmol/l and 15: 410 (172.4–1353.4) nmol/l; (C) 5-HIAA in AHS group; 1: 79.6 (25.7–238.0) nmol/l and 15: 103.4 (33.4–262.0) nmol/l (D) 5-HIAA in mixed group; 1: 64.0 (40.7–100.8) nmol/l and 15: 128.0 (57.5–358.6) nmol/l (E) MHPG in AHS group; 1: 43.1 (21.7–91.5) nmol/l and 15: 44.2 (26.4–90.0) nmol/l; (F) MHPG in mixed group; 1: 45.3 (22.3–160.8) nmol/l and 15: 42.9 (20.9–108.6) nmol/l.

CSF outflow conductance. There was a positive correlation in the AHS group (fig 2A; linear regression; $r = 0.64$, $p = 0.014$) but not in the mixed group. There were also a positive correlation between CSF HVA concentration and CSF pressure (linear regression; $r = 0.63$, $p = 0.017$) in the AHS group only.

There was a positive correlation between the first fraction of CSF 5-HIAA concentration and CSF outflow conductance in the AHS group (fig 2B; linear regression; $r =$

0.62 , $p < 0.03$), but not in the mixed group. A trend was found between CSF 5-HIAA and CSF pressure (linear regression; $r = 0.49$, $p = 0.068$) in the AHS group.

There were no significant correlations between concentration of any of the monoamine metabolites in the first CSF fraction *v* the physical or clinical variables described in table 2 in either the AHS or the mixed group. There were no significant correlations between the values for individual slope

Figure 2 (A) Linear correlation between the first CSF fraction of HVA and CSF outflow conductance in the AHS group; (B) Linear correlation between the first CSF fraction of 5-HIAA and CSF outflow conductance in the AHS group.

Table 3 Linear correlations between concentrations of HVA and 5-HIAA in the first CSF fractions and sequential fractions

CSF fraction (ml CSF)	Correlation coefficient		Patients (No)
	HVA	5-HIAA	
2	0.95***	0.81***	22
4	0.86***	0.62***	25
6	0.75***	0.40*	26
8	0.73***	0.22	25
10	0.87***	0.73***	25
12	0.85***	0.62***	24
14	0.89***	0.69***	23
16	0.75***	0.51**	22
18	0.68***	0.36	22
20	0.69***	0.54**	25
22	0.78***	0.55**	25
24	0.50***	0.24	24
26	0.51***	0.30	26
28	0.62***	0.37	24
30	0.72***	0.46*	22
32	0.49*	0.09	23
34	0.57**	0.08	21
36	0.36	0.14	20
38	0.34	0.14	21
40	0.60	0.17	20
42	0.73***	0.48*	20
44	0.34	0.03	21
46	0.18	0.02	21
48	0.23	0.24	18
50	0.52*	0.10	15
52	0.47	0.10	15
54	0.93***	0.14	14
56	0.72	0.22	10
58	0.43	0.56	6

*p < 0.05; **p < 0.01; ***p < 0.001.

gradients of monoamine metabolites and CSF hydrodynamics, or the physical or clinical variables described in table 2.

As there were no apparent differences in the concentration curves in the AHS and mixed groups (fig 1A, 1C v 1B, 1D), patients were pooled in to a single group for further calculations. Table 3 shows the correlations between concentrations of HVA and 5-HIAA in the first 2 ml of CSF removed and each subsequent CSF fraction. Correlations of HVA were high until about 34 ml CSF had been removed. Highly significant correlations of 5-HIAA were noted until about 24 ml of CSF had been removed and thereafter there were no correlations.

Discussion

The major finding in this prospective study of patients with communicating hydrocephalus was a rostrocaudal CSF gradient of the acidic monoamine metabolite (HVA and 5-HIAA) concentrations that levelled off to a plateau after withdrawal of more than 25 ml of CSF (Fig 1 A-D). Also we confirmed our previous report of a positive linear relation between lumbar HVA and 5-HIAA concentrations v CSF outflow conductance.⁵ A rostrocaudal gradient of HVA and 5-HIAA has been reported previously (table 1), but with a less pronounced slope of 5-HIAA compared with HVA in some instances. The plateau phase has not been described before, probably because of methodological faults in previous studies (table 1). Also, a limited number of patients, inconsistencies in diagnosis, removal of small quantities of CSF, or interference with analyses because of injection of gas, and inconsistencies in body position during lumbar puncture (table 1) may have influenced the results.

The following discussion (fig 3A-C) is based on the fact that CSF HVA is formed mainly in the basal ganglia⁷ and the assumption that CSF HVA is an indicator of CSF flow. One has to take into consideration, however, the influence of formation, mixing, and clearance of CSF (as discussed in the second section), which are plausible confounding mechanisms in the model to be discussed.

GRADIENTS EXPLAINED BY SPINAL CSF FLOW?

Little is known about the existence of an upward or downward spinal CSF flow in humans; this is probably the main obstacle in the interpretation of concentration gradients in the subarachnoid spinal space. According to fig 3, there are three principle possibilities of the spinal flow direction: downwards, upwards or to and fro (fig 3A-C):

CSF flow downward (fig 3A)

If the spinal CSF flow is directed downwards, CSF HVA from the ventricles moves via the basal cisternae towards the cortical space as well as towards the spinal compartment. This implies the existence of CSF absorption at the spinal level. A structure that closely resembles the arachnoid villi associated with the spinal

Figure 3 Directions of CSF flow. Schematic drawings of hypothetical models of CSF flow explaining different types of concentration curves. For further explanation, see discussion.

nerve roots has been suggested.³⁴ Besides the venous drainage, a "lymphatic pathway" has been proposed.²⁶ A CSF stream directed downward together with clearance of HVA along the way, will result in a rostrocaudal concentration gradient according to fig 3A.

CSF flow upwards (fig 3B)

Assuming that CSF is produced by the spinal cord and no absorption occurs in the perispinal space, a flow will be directed upwards and CSF eliminated by the cortical arachnoid villi. No HVA will be found in the initial fractions when CSF is removed by a lumbar CSF tap. When cisternal CSF reaches the lumbar needle, however, there will be an immediate increase in HVA concentration (fig 3B). Sato *et al*³⁵ have shown formation of CSF by the spinal cord in dogs but Lux *et al*³⁶ could not confirm this for the monkey. No data are available on the formation of CSF from the spinal cord parenchyma in humans.

CSF flow to and fro (fig 3C)

In the absence of CSF spinal formation and spinal CSF absorption, there will be no net flow through the foramen magnum and the concentration curve of CSF HVA will be close to zero in the first fractions. Because of diffusion and the impelling forces of the spinal CSF (perpetual variations in volume during cardiac systole, respiration, talking, coughing, sneezing, straining, or changing body position),^{26, 37} there will be a to and fro motion of CSF between the spinal and supraspinal compartments. From zero, the concentrations of CSF HVA would steadily increase in ensuing fractions (fig 3C).

In conclusion, the rostrocaudal gradients of CSF HVA and CSF 5-HIAA may be predicted by a spinal CSF flow directed downwards, CSF absorption, and clearance of acidic metabolites at the spinal level. There was no rostrocaudal gradient of MHPG, which was an expected finding considering that the neutral glycol MHPG diffuses freely and rapidly across the blood-brain and blood-CSF barriers.^{3, 38-40}

THE PLATEAU

Most studies report a striking similarity between the ratio of maximum concentrations *v* the base line concentrations of CSF HVA and CSF 5-HIAA. In the present study the ratio was about 2 in both 5-HIAA and HVA, which agrees with previous results (table 1). In studies in which monoamine concentrations in lumbar CSF have been compared with concentrations in CSF removed simultaneously from the ventricles, HVA and 5-HIAA concentrations in the lateral ventricles have been estimated to be up to threefold to 10-fold those in lumbar CSF.^{9, 38, 40-42} The ventriculolumbar gradients were 4:1 and 5:1, respectively, in nine patients with AHS.³⁸ The plateau phase in later CSF fractions in this study, using a lumbar CSF tap technique, with a lower maximum ratio of CSF HVA and 5-HIAA compared with studies with simultaneous measurement from lumbar and ventricular CSF may be explained by some of the following mechanisms. These variables may also explain the increasing confidence intervals in the later CSF portions (fig 1A-F).

Mixing of CSF from different compartments

When CSF pressure is lowered by CSF removal, as in this study, CSF in the system as well as newly formed CSF will flow towards the lumbar puncture needle. The initial fractions are composed of CSF from adjacent regions, both caudal and rostral to the lumbar puncture needle. As the drain continues, the spinal subarachnoid space is gradually emptied and the net CSF volume is replaced by dilatation of the extradural veins⁴⁴ and CSF from the intracranial compartments. During this phase the CSF pressure declines and will fall below the sagittal sinus pressure. When this happens, CSF from the ventricles as well as CSF in the cortical subarachnoid space will flow towards the lumbar region. This implies a mixture of CSF from different

compartments containing different concentrations of CSF monoamines in the lumbar spinal space.²⁵ Emptying of the cortical sulcal CSF volume during the lumbar puncture is possible.⁴⁵

Different sites of CSF production

CSF is generally considered to be mainly formed by the choroid plexus. Recent studies suggest, however, that up to 70% of the CSF may be formed in extraventricular compartments.^{46,47} This implies that CSF formed in the ventricles containing high concentrations of the monoamine metabolite HVA would be "diluted" when reaching the cisternae and the spinal compartment by CSF containing low concentrations.

Clearance of metabolites to venous blood

CSF HVA and CSF 5-HIAA are cleared by a probenecid sensitive system.⁴⁸ The site of this system is unknown but the cortical subarachnoid space⁴⁹ and to a lesser extent the fourth ventricle⁵⁰ and the spinal subarachnoid space⁵¹ have been proposed. Monoamine metabolites circulating with the CSF bulk flow will be continuously absorbed resulting in a gradual decrease in transmitter concentrations as the CSF travels down the spinal subarachnoid space.

Clearance of metabolites to extracellular fluid

Metabolite concentrations may also be lowered by diffusion from CSF to extracellular fluid along the CSF circulating pathway. The spinal cord has an extensive surface area across which diffusional loss of a monoamine metabolite may occur.

CSF outflow conductance

Disturbed hydrodynamics of CSF affect formation, mixing, and direction of flow of CSF and interrupt formation and clearance of monoamine metabolites.^{5,10} As confirmed in this study, a low CSF outflow conductance may facilitate the clearance of acidic substances.⁵

Clinical implications

In the present study there was good correlation between the initial lumbar and subsequent concentrations of CSF HVA and 5-HIAA respectively until 40 ml of CSF had been removed (table 3). A similar result was described by Banki and Molnár,²⁰ who removed CSF via the lumbar route and found a high correlation coefficient ($r = 0.73$) between the first and last fraction (42 ml CSF removed). In the present study, the positive correlations disappeared in the last CSF fractions. These fractions are not solely representative for ventricular CSF, and the impact of mixing and dilution of CSF and clearance of acidic metabolites on lumbar CSF HVA and 5-HIAA concentrations, as earlier discussed, could provide a reasonable explanation for this finding. A lumbar CSF tap could not be used to remove ventricular CSF exclusively; however, lumbar CSF HVA and 5-HIAA

seem to be derived from the same sources as the fluid next to the brain, although not defined to any specific supraspinal compartment. Lumbar CSF MHPG does not reflect brain neurochemistry.

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