Fibrin-fibrinogen degradation products in cerebrospinal fluid

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SYNOPSIS An increase in low molecular weight fibrin-fibrinogen degradation products (FDP) was demonstrated in cerebrospinal fluid (CSF) from 17 of 18 patients with bacterial or viral meningitis compared with 29 patients without meningitis. The CSF also showed an increase in coagulation proteins of molecular weight less than 90 000 (factors VII, IX, and plasminogen) but did not contain fibrinogen (MW 340 000) or plasminogen activator. It is concluded that low molecular weight FDP in the CSF in infective meningitis result from leakage through a damaged blood-CSF barrier rather than from local digestion of fibrin deposited on the meninges.

Fibrin-fibrinogen degradation products (FDP) are not found in normal cerebrospinal fluid (CSF) but have been demonstrated following subarachnoid haemorrhage (Tovi, 1972), meningococcal meningitis (Brueton *et al*, 1974), and in patients with raised CSF protein (Hunter *et al*, 1974). The source of origin of the FDP is not clear although this is an important point in view of the potential diagnostic value of raised FDP and also in relation to the treatment of subarachnoid haemorrhage by fibrinolytic blockade (Mullan and Dawley, 1968; Gibbs and Corkill, 1971; Tovi, 1972).

Normal CSF does not contain plasminogen activator (Tovi, 1972) although the meninges and choroid plexus are rich in this fibrinolytic activator. The release of an increased amount of activator from these tissues could therefore result in the local digestion of fibrin-fibrinogen to give rise to FDP in the CSF. Alternatively, FDP could leak into the CSF via a damaged blood-CSF barrier. The latter would be analogous to the appearance of low molecular weight FDP in the urine when glomerular basement membrane damage results in highly selective proteinuria (Hall *et al*, 1975).

A study has therefore been made of coagulation-fibrinolytic proteins of differing molecular weight in the CSF and blood from 47 patients undergoing diagnostic lumbar puncture in order to determine the source of origin of FDP in the CSF in meningitis.

Patients and Methods

Fifty-eight specimens of CSF were obtained from 47 patients undergoing diagnostic lumbar puncture.

The 47 patients (42 were children) included 13 patients with bacterial and five with viral meningitis (meningitis group). The remaining 29 patients (non-menigitis group) comprised 10 with acute lymphoblastic leukaemia (three with meningeal involvement), five with febrile convulsions, and eight with infections such as pneumonia which did not involve the CNS but were associated with meningism; the remaining six patients had a variety of conditions affecting their level of consciousness but without evidence of CNS infection. Patients with bacterial meningitis were initially treated for two weeks with either parenteral chloramphenicol, penicillin, and sulphadimidine, or intravenous ampicillin.

Lumbar CSF (1·0 ml) was taken directly into a plastic tube containing 0·1 ml of 3·8 % sodium citrate for estimation of plasminogen, fibrinogen, and factors VII and IX, and 0·5 ml was taken into 250 units of aprotinin in a dry container for estimation of FDP. The specimens were transported immediately to the laboratory and centrifuged at 0-4°C. Thrombin (0·1 ml of a 50 unit/ml concentration) was added to the supernatant of the specimen containing aprotinin and, after incubation at 37°C for 15 minutes, the specimen was assayed for FDP using Thrombo-Wellcotest (Wellcome Reagents Ltd) and Diagen (Diagnostic Reagents Ltd) latex slide tests and also the tanned red cell haemagglutination inhibition immunoassay (TRCHII) of Merskey *et al* (1966).

Venous blood (1·0 ml) was taken into a Thrombo-Wellcotest kit sample tube (containing thrombin and aprotinin) for FDP estimation by the three methods.

Plasminogen in CSF was estimated by the method of De Vreker (1965), factor IX by one-stage assay (Hardisty and Ingram, 1965), and factor VII by a one-stage method (Stefanini, 1950) substituting fac-

Test	Result		Units
Thrombo-Wellcotest	<8	(34)	mg/l
Diagen	<4	(34)	mg/l
TRCHII	<5	(34)	mg/l
Plasminogen	<10	(24)	units
Fibrinogen	0	(32)	g/l
Factor VII	0	(7)	%
Factor IX	0	(7)	%
Fibrin plate lysis	<60	(8)	mm²
Protein	<0·5	(31)	g/l

Table I   *Range of values obtained from 34 CSF specimens from 29 patients without meningitis*
The number of CSF samples studied for each test is stated in parentheses.

tor VII-deficient plasma and omitting adsorption with aluminium hydroxide. Plasminogen activator was measured using human fibrin plates (Astrup and Müllertz, 1952; Cash and Allan, 1967). Red and white cell counts on CSF were performed using an improved Neubauer counting chamber, and identification of leukaemic blast cells was confirmed by cytocentrifugation. CSF protein was estimated according to King and Haslewood (1936). Citrated samples of CSF for fibrinogen estimation were tested undiluted and diluted 1/1-1/8 in glycylglycine buffer and then reacted with Diagen test reagent which incorporates an antiserum against whole fibrinogen.

Statistical significance was determined using Student's *t* test and correlation coefficient. Molecular weight values for coagulation factors were obtained from Hardisty (1974).

Results

NON-MENINGITIS GROUP
The range of CSF values obtained for this group is given in table I. Of the 29 patients studied, only two showed FDP values in the CSF (by the Thrombo-Wellcotest method) and in low concentration (<8 mg/l); the remaining 27 patients had no detectable FDP by all three methods studied. None of 24 CSF samples contained detectable plasminogen; the lower limit of sensitivity of the test was 10 units. None of the CSF samples tested contained demonstrable fibrinogen, factor VII or factor IX, or showed plasminogen activator activity.

MENINGITIS GROUP
The results obtained for the 18 patients are given in table II. The FDP values are stated for the Thrombo-Wellcotest method only since this was the most sensitive of the three methods studied; it gave a raised value (16 mg/l or above) in 17 of the 18 patients, and in 10 patients it was the only test of the three to give a raised FDP value. The FDP values obtained by the Thrombo-Wellcotest were consistently higher than the values obtained by the two other methods. The increased reactivity of the low molecular weight FDP fragments FgD and FgE (MW 50-83000) in the Thrombo-Wellcotest compared with the Diagen latex test has previously been reported (Donati *et al*, 1973).

Plasminogen values in the CSF were raised in 11 of 13 patients studied, factor IX was detected in five of seven CSF samples, and CSF protein was raised in 14 of the 18 patients. None of the samples showed any increase in plasminogen activator activity by the fibrin plate method or contained detectable fibrinogen.

CORRELATION WITH CSF ERYTHROCYTE COUNT
Nine of the 29 patients without meningitis showed a

FDP (mg/l)		Plasminogen (units)	Factor IX (%)	Factor VII (%)	Protein (g/l)	Erythrocytes (× 10⁶/l)	Leucocytes (× 10⁶/l)
CSF	Serum						
16— 32	—	70	—	—	1·0	6	30
16— 32	—	180	—	—	1·0	4 000	0
32— 64	—	100	—	—	3·0	2 800	3 400
128—256	16— 32	70	—	—	0·6	500	2 500
32— 64	256—512	—	—	—	3·0	770	2 100
128—256	32— 64	120	14	5	2·5	1 000	12 000
32— 64	<8	—	—	—	0 3	0	282
16— 32	—	0	0	0	3·0	120	8 000
32— 64	16— 32	24	6	0	0·5	200	1 400
64—128	32— 64	80	42	30	1·5	95	24 500
64—128	8— 16	100	4	0	2·5	0	13 000
16— 32	<8	—	—	—	0·2	6	8
128—256	8— 16	10	—	—	0·4	1 200	25
128—256	16— 32	320	20	15	3·7	210	5 600
16— 32	<8	—	—	—	0·9	0	700
16— 32	<8	—	—	—	0·8	160 000	114
64—128	<8	80	0	0	1·3	1 000	20
4 — 8	<8	20	—	—	0·3	120	160

Table II   *CSF results for the 18 patients in the meningitis group: serum FDP levels given for comparison*

raised CSF erythrocyte count (range 20-2 400 × 10^6/l) but there were no detectable FDP (Thrombo-Wellcotest) in eight of the nine; the ninth patient showed FDP of only 4-8 mg/l. Of 12 patients in the meningitis group with 120-160 000 × 10^6 erythrocytes/l CSF, there was an increase in FDP in 11; conversely, five patients with raised FDP levels in the CSF showed no erythrocytes or only 6 × 10^6/l. In patients with both a raised FDP level and erythrocyte count in the CSF there was no significant correlation between the individual values (r = 0·09, P > 0·5). Similarly, there was no significant correlation between CSF erythrocyte count and factor IX level (r = 0·014, P > 0·5).

RELATIONSHIP BETWEEN FDP IN THE SERUM AND CSF
Of the 17 patients with raised FDP (Thrombo-Wellcotest) in the CSF, the serum FDP level was measured in 14 and found to be raised (range 16-512 mg/l) in six (table II). The patient with the very high serum FDP level (256-512 mg/l) also suffered from septicaemia.

SEQUENTIAL CSF STUDY
Serial samples of CSF were obtained from three patients in the meningitis group with persistent or progressive neurological signs. In a child who developed hydrocephalus, FDP were still present in the CSF after five weeks, and in an infant with severe diffuse cerebral damage they were still present at six weeks. The latter patient also developed bilateral subdural effusions which contained a high concentration of FDP (1024 mg/l). In a third patient, who developed a hemiplegia, the FDP level was still rising after two weeks. In all three patients, serum FDP were very low or undetectable.

Discussion

This study has shown that the concentration of low molecular weight FDP, and also coagulation proteins of molecular weight less than 90 000, was either very low or undetectable in CSF from 29 patients with no demonstrable inflammatory disease of the meninges. In contrast, 17 of 18 patients with bacterial or viral meningitis showed an increase in low molecular weight FDP in the CSF when measured by a latex test incorporating antibody against FgD and FgE (MW 50-83 000). The FDP were usually accompanied by plasminogen (MW 89 000), protein, and factors VII (MW 35-63000) and/or IX (MW 50 000) but not by fibrinogen (MW 340 000) or plasminogen activator.

The individual levels of FDP and factor IX did not correlate with the number of erythrocytes in the CSF so that the selective increase in low molecular weight proteins is unlikely to have resulted from contamination with blood at the time of lumbar puncture. It seems more likely that the leakage resulted from inflammatory damage to the blood—CSF barrier. Since the half-life of FDP is approximately 5-15 hours (Hardisty, 1974) and since the CSF is probably renewed every 6-8 hours (Milhorat, 1972), the persistence of small molecular size FDP and other proteins in the CSF for several weeks in three patients with a local complication of meningitis is suggestive of continued leakage of this type. Fibrin deposition on the meninges and its subsequent lysis could also give rise to FDP in the CSF; in the present study, however, there was no demonstrable increase in CSF plasminogen activator as might be expected if this were a major mechanism.

Raised FDP levels in the CSF were sometimes found in the absence of raised serum FDP. The cerebral vessels in meningitis may show areas of arteritis with occlusion (Thomas and Hopkins, 1972) and also vasodilatation (Leeds and Goldberg, 1971) so that local fibrin deposition and breakdown could result in a high local concentration of FDP with direct leakage into CSF. The accompanying rise in FDP in the serum of some patients may be secondary to this local production in the cerebral vessels or arise systemically as a consequence of transient viraemia, bacteraemia or endotoxaemia.

FDP may also appear in the CSF following subarachnoid haemorrhage (Tovi, 1972) and this has been explained on the basis of secondary fibrinolytic digestion of the fibrin clot. This explanation is the rationale for the use of fibrinolytic blockers such as epsilon aminocaproic acid to prevent re-bleeding (Mullan and Dawley, 1968; Gibbs and Corkill, 1971; Tovi, 1972). Patients with subarachnoid haemorrhage do not, however, show increased plasminogen activator activity in the CSF (Tovi *et al*, 1972), as would be expected with increased fibrinolysis, and the present study of meningitis has indicated an alternative mechanism for the occurrence of FDP in CSF. Further studies of this type are therefore indicated in subarachnoid haemorrhage, and also in patients with raised CSF protein of non-inflammatory origin (Hunter *et al*, 1974) and in infants with birth asphyxia who show excess protein in ventricular CSF and are prone to intraventricular haemorrhage (Cole *et al*, 1974).

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