G_{M1} Ganglioside Concentration in the Cerebrospinal Fluid of Neonates and Children.

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SUMMARY

G_{M1} ganglioside concentration was measured by radioassay technique in individual samples of lumbar cerebrospinal fluid from 20 neonatal and 17 older pediatric patients. The lumbar CSF G_{M1} ganglioside concentration of neonates (76.6 ± 27.4 ng/ml) is greater than that of older infants and children (31.9 ± 22.2 ng/ml). The lower range of G_{M1} ganglioside concentration of CSF from older pediatric patients is similar to the previously reported adult CSF values. The mean CSF G_{M1} ganglioside concentration in pediatric patients with active neurologic disease (53.1 ± 30.0 ng/ml) is greater than that of children without central nervous system pathology. The temporal evolution and magnitude above baseline values of lumbar CSF G_{M1} ganglioside concentration in three neonates was correlated with the clinical status of these patients.

SPECULATION

The development of a rapid, sensitive radioassay for G_{M1} ganglioside concentration in small aliquots of CSF permits the timely determination of CSF G_{M1} ganglioside concentration in individual patients. The preliminary data suggest that altered temporal patterns of neonatal CSF G_{M1} ganglioside concentration during a period of acute central nervous system injury may be a sensitive indicator of active central nervous system pathology.

INTRODUCTION

Gangliosides are sialoglycosphingolipids that are predominately located in neurons (11,14). The human cerebral cortex ganglioside dry weight concentration is approximately five-fold greater than that of white matter (9). In contrast, white matter has a larger concentration of myelin basic protein than does grey matter. This myelin associated protein is unique to central nervous system tissue (2).

The magnitude of cerebrospinal fluid myelin basic protein activity has been used as an indicator of both severe myelin damage (8) and of active demyelination (3). Studies of brain gangliosides in glycosphingolipidoses suggest a correlation between ganglioside concentration or pattern and the presence of neuronal pathology (14).

Previous studies of cerebrospinal fluid or serum ganglioside content have been limited by assay techniques that require large specimen volumes and often necessitate the use of pooled samples (1,5,7,9,12,14). We have recently developed a radioassay for the monosialoglycosphingolipid G_{M1} (6). The assay is based on the high affinity binding of G_{M1} ganglioside to cholera toxin (4) and permits the rapid measurement of G_{M1} ganglioside concentration in twenty to fifty microliter aliquots of cerebrospinal fluid.

This paper reports the radioassay measurement of G_{M1} ganglioside concentration in the cerebrospinal fluid of individual neonates and children. Correlation of this preliminary data with the clinical status of a small group of newborn infants suggests that the temporal pattern of cerebrospinal fluid G_{M1} ganglioside concentration during a period of injury may be a sensitive indicator of the severity of acquired central nervous system damage.

MATERIALS AND METHODS

Samples. Cerebrospinal fluid samples were collected in glass or plastic tubes and stored at -20°C or -60°C until the time of assay. Prior to assay, samples were thawed, mixed, and centrifuged (2000 X g, 5 minutes).

G_{M1} Ganglioside Radioassay. The radioassay for G_{M1} ganglioside has been described elsewhere in detail (6). Briefly, after addition of 20-50 µl of an appropriate dilution of ganglioside standard or CSF to 12 X 75 mm plastic tubes, the volume was adjusted to 150 µl with freshly prepared 0.1 M sodium phosphate buffer (pH 7.4) that contained 1% (w/v) bovine serum albumin. Fifty microliters of ¹²⁵Iodinated cholera toxin (approximately 10 ng/50 µl; 500,000 cpm/50 µl) was added to the assay tube, the solution mixed for 2-3 seconds, and incubated for a total of forty minutes (after the addition of iodotoxin). Fifty microliters of liver membrane suspension (protein concentration of 1.5 mg/ml) was added and the incubation at 23°C was continued for 10 minutes with intermittent shaking of the mixture. Incubation was terminated by the addition of 2 ml of distilled-deionized water. The diluted mixture was filtered through a Millipore EGWP 25 mm Cellotape® membrane, and the membrane subsequently transferred to a disposable glass or plastic tube for counting (1 minute) in a gamma counter.

Myelin Basic Protein Radioimmunoassay. The radioimmunoassay for myelin basic protein has been described elsewhere in detail (2,3). Cerebrospinal fluid samples were stored at -20°C until the time of assay.

Clinical Population. Single samples of cerebrospinal fluid were obtained from 17 neonates and 17 older pediatric patients ranging in age from 5 weeks to 18 years. Lumbar punctures had been performed for evaluation of suspected sepsis, fever associated seizures, or failure to thrive. Serial samples were available from three additional neonates whose detailed clinical courses are described.

CASE REPORTS

Case 1 - This Black male infant was the 3060 gram product of a gravida 1, healthy 17 year old. The family history was unremarkable. No antenatal abnormalities were known. Labor commenced spontaneously at term and terminated 12 hours later via a face presentation. The APGAR scores were 7 (1 min) and 9 (5 min). During the first day of life he had a poor cry, deficient suck, and was noted to be icteric. The infants vigor improved by day 2. On day 5, post feeding, he experienced transient respiratory distress that was associated with a 20 second period of apnea. Eupnea returned after brief ambu-bag resuscitation. His subsequent course was uneventful. Relevant laboratory data are shown in Table 3. Blood, urine and CSF cultures as well as CSF counterimmunoelectrophoresis was normal. No specific pathologic etiology for the patient's icterus was found. No neurobehavioural abnormalities were apparent upon discharge at age 10 days, after completion of a

9 day course of ampicillin and kanamycin treatment. Outpatient department assessment at age 8 weeks documented normal growth and development.

Case 2 - This Black female was the 2280 gram product of a gravida 2, nulliparous 31 year old vegetarian. The family history was unremarkable. No antenatal abnormalities were known. Labor commenced spontaneously at 35 weeks gestational age and terminated 12 hours later in a vertex presentation. Slight meconium staining occurred. The APGAR scores were 3 (1 min) and 5 (5 min). Immediate endotracheal resuscitation at the time of birth was associated with a prompt response and adequate respiratory function. However, upon admission to the newborn nursery the infant developed cyanosis, bradycardia, apnea, and convulsions. These symptoms were associated with hypoglycemia (serum glucose: 22 mg/dl). Prompt parenteral glucose administration was effected. No bacterial or viral etiology for these symptoms was found. In subsequent weeks, not withstanding antibiotic therapy, the baby continued to have episodes of bradycardia, temperature instability, seizures, and failure to thrive. At age three weeks, the serum T₄ concentration was found to be 3.3 µg/dl (normal range: 5-13 µg/dl) and administration of a thyroid hormone preparation was initiated at that time; by 10 weeks of age the T₄ concentration had normalized.

The EEG on day 12 showed frequent paroxysmal generalized and synchronous slow wave activity with associated underlying symmetrical spike activity. A repeat EEG on day 22 was noted to be slightly improved from the record ten days earlier. A CTT scan of the head at 6 weeks of age was within normal limits.

Neurobehavioural assessment at age 7 weeks noted an inactive, poorly developed infant who continued to require isolette care for thermal stability. Body weight was 2245 grams (3rd percentile). The infant slept when she was not stimulated. During arousal the visual axes were conjugate, a vacuous stare was noted, and pupillary light reflexes were present. However, the infant did not fix on a human face, regard a rattle, or follow a light. Oculo-vestibular stimuli evoked tonic conjugate eye deviations without a fast corrective phase. Excessive extremity abduction and hypotonia were present. There was neither positive weight support nor automatic walking. Inverted suspension and traction responses were deficient. Marked head lag was noted. The deep tendon reflexes were normally brisk but no fasciculations were observed. No rooting could be elicited but there was a prominent snout reflex. Weak palmar and plantar grasp responses were elicited on the left; right grasp responses were absent. Babinski's response was present bilaterally. Placing responses, the Moro reflex, and tonic neck attitudes were absent. Nociceptive stimulation led to a brief, weak, monotonous, cry. The head and chest were not elevated in ventral recumbency. During the brief periods of arousal, relatively frequent, transient periods of lip-smacking, conjugate eye deviation, as well as tonic, torsional, extensor, body and extremity posturing were witnessed.

Case 3 - This Hispanic male was the 3360 gram product of a gravida 1, healthy 15 year old. The family history was unremarkable. No antenatal abnormalities were known. Labor commenced spontaneously at term and terminated 14 hours later via a vertex presentation. Meconium stained amniotic fluid was noted intrapartum. The APGAR scores were 1 (1 min) and 4 (5 min). Notwithstanding prompt tracheal aspiration and intubation, the neonate never respired spontaneously. Upon admission to a special care nursery he exhibited scalp edema, absent pupillary light reflexes, immobility, and hypotonia. Neurologic examination at age 2 days failed to document neural activity in any intracranial structure. Mechanical ventilation, antibiotic therapy and parenteral fluids failed to prevent terminal bradycardia and hypotension at 9 days of age.

RESULTS

The relevant clinical data and CSF G_{M1} ganglioside concentrations of 17 neonates and 17 older pediatric patients are shown in Tables 1 and 2, respectively. Table 3 describes the longitudinal evolution of CSF G_{M1} ganglioside and myelin basic protein concentrations as well as relevant clinical information on three neonates on whom serial lumbar punctures were performed.

In the neonates discharged as healthy babies but with diverse perinatal complications (cases 1, 4-20), the mean CSF G_{M1} ganglioside concentration was 76.6 ± 27.4 ng/ml ($\bar{X} \pm$ SD; range 35.8-129.6 ng/ml). The range of initial lumbar CSF G_{M1} ganglioside concentrations in three neonates, in which subsequent lumbar punctures were performed (cases 1-3), was 53.1-80.1 ng/ml. In cases 2 and 3 the measured CSF G_{M1} ganglioside concentration subsequently increased and remained elevated above the initial values. No serial increase in CSF G_{M1} ganglioside concentration was noted in case 1. In these three neonates, CSF myelin basic protein was detectable only in the terminal sample from case 3 (6.4 ng/ml).

The mean lumbar CSF G_{M1} ganglioside concentration in 9 pediatric age patients without nervous system pathology (cases 21-29) and in 8 children with active neurologic disease (cases 30-37) were 31.9 ± 22.2 ng/ml ($\bar{X} \pm$ SD; range: 4.8-70.0 ng/ml) and 53.1 ± 30.0 ng/ml ($\bar{X} \pm$ SD; range: 6.1-94.5 ng/ml) respectively.

No correlation was found between CSF G_{M1} ganglioside concentration and CSF erythrocyte, leukocyte, total calcium, glucose or total protein or serum bilirubin contents. The addition of buffer washed (0.1 M sodium phosphate, pH 7.4) adult human erythrocytes and leukocytes to CSF (hematocrit of approximately 6.4%) reduced measured G_{M1} ganglioside concentration by 7.9%. Prior storage, -20°C, of this sanguinous CSF yielded only a 4.8% decrease of measured G_{M1} ganglioside concentrations. The addition of serum to CSF (radioassay incubation solution total protein of 800 mg/dl) reduced radioassay measured G_{M1} ganglioside concentration by only 13%.

DISCUSSION

This study of cerebrospinal fluid G_{M1} ganglioside concentration in newborn infants and children, as presented in Tables 1 and 2, indicates the versatility of the G_{M1} ganglioside radioassay. This radioassay permits the rapid measurement of G_{M1} ganglioside concentration in small aliquots of cerebrospinal fluid from individual patients. Data from studies of adult patients with diverse neurological disorders has indicated that the increase of CSF G_{M1} ganglioside concentration is not specific for a single disease process (6). Our lower range values of CSF G_{M1} ganglioside concentration in children older than 28 days of age, who had fever associated seizures and/or suspected pyogenic leptomeningitis, agree with the lower range of CSF G_{M1} ganglioside concentration found in adults. The present study does not document a "true" normal CSF G_{M1} ganglioside concentration at all ages. The specimens of CSF that we studied were obtained from patients with diverse toxic-neuropath-

ological processes. However, the lower range of pediatric and adult CSF GM₁ ganglioside concentration determined by radioassay is similar to the previously reported normal adult CSF values (1,6,9,12). We can thus conclude that certain disease states result in minimal, if any, elevation above normal values of single, isolated measurements of CSF GM₁ ganglioside concentration.

In our study, the mean cerebrospinal fluid GM₁ ganglioside concentration of neonatal subjects (Table 1) is greater than that of older infants and children (Table 2). Ledeen and Yu (10) as well as Svennerholm (14) have reviewed published concentrations and patterns of ganglioside sialic acid in human brains at different ages. The reviewed data indicate that the GM₁ ganglioside sialic acid concentration in human infant brain is greater than that in human adult brain. However, the adult and newborn values of brain total ganglioside sialic acid content do not differ. Ledeen and Yu (9) also concluded that the lumbar CSF ganglioside pattern is similar to that of brain. Thus, we conclude that the increase in CSF GM₁ ganglioside concentration in neonates and in patients with neuropathologic processes indicates that the CSF GM₁ ganglioside concentration is probably a function of both central nervous system tissue ganglioside content and turnover.

We found no prognostic significance of single CSF GM₁ ganglioside concentration determinations in individual newborn infants who were being evaluated for possible sepsis, seizures, or failure to thrive (Table 1). However, the temporal evolution above baseline values of CSF GM₁ ganglioside concentration in three neonates did correlate with the clinical status of these patients (Table 3).

Single assays of CSF myelin basic protein concentration in individual newborn infants and children may indicate severe brain tissue destruction (8). No prognostic significance was noted in our individual or serial measurements of CSF myelin basic protein in the three neonates whose temporal pattern of CSF GM₁ ganglioside concentration was studied (Table 3). Cerebrospinal fluid myelin basic protein concentration is a less sensitive indicator of acute neonatal central nervous system injury than serial CSF GM₁ ganglioside concentrations. This may be accounted for by the relative paucity of central nervous system myelin in the human newborn.

The data reported in this communication suggest that alterations of newborn central nervous system ganglioside concentration and/or turnover can be observed by determining the temporal evolution of neonatal CSF GM₁ ganglioside concentration. During the acute phase of injury, altered temporal patterns of CSF GM₁ ganglioside concentration may be a sensitive indicator of central nervous system damage. To further test these hypotheses, future studies of larger populations of newborn infants with diverse, acute, potentially encephalopathic events are necessary.

REFERENCES

1. Balasubramanian, A.S., Raman, P.T., and Taori, G.M.: Free and bound N-Acetyl neuraminic acid in the cerebrospinal fluid in various neuro-

- logical disorders. *Indian J Med Res*, 62: 781-787 (1974).
2. Cohen, S.R., McKhann, G.M., and Guarneri, M.: A radioimmunoassay for myelin basic protein and its use for quantitative measurements. *J. Neurochem*, 25: 371-376 (1975).
3. Cohen, S.R., Herdon, R.M., and McKhann, G.M.: Radioimmunoassay of myelin basic protein in spinal fluid. An index of active demyelination. *N Engl J Med*, 295: 1455-1457 (1976).
4. Cuatrecasas, P.: Interaction of *Vibrio cholerae* enterotoxin with cell membranes. *Biochem*, 12: 3547-3558 (1973).
5. Evans, J.E., and McCluer, R.H.: High pressure liquid chromatography of neutral glycosphingolipids. *Biochem Biophys Acta*, 270: 565-569 (1972).
6. Ginns, E.I., and French, J.H.: A radioassay for GM₁ ganglioside concentration in cerebrospinal fluid. *J Neurochem*, (in press for Oct. 1980).
7. Iwamori, M., and Nagai, Y.: A new chromatographic approach to the resolution of individual gangliosides. *Biochem Biophys Acta*, 528: 257-267 (1978).
8. Kohlschütter, A.: Myelin basic protein in cerebrospinal fluid from children. *Eur J Pediatr*, 127: 155-161 (1978).
9. Ledeen, R.W., and Yu, R.K.: Gangliosides of CSF and plasma: their relation to the nervous system, in Volk, B.W. (ed): *Sphingolipids, Sphingolipidoses and Allied Disorders* (Plenum Publishing Corporation, New York, 1972) pp. 77-93.
10. Ledeen, R.W., and Yu, R.K.: Gangliosides of the nervous system, in Witting, L. (ed): *Glycolipid Methodology* (Am Oil Chem Soc, 1976) pp. 187-214.
11. Ledeen, R.W.: Ganglioside structures and distribution: Are they localized at the nerve ending? *J Supramol Struct*, 8: 1-17 (1978).
12. Nagai, Y., Kanfer, J.N., and Tourtellotte, W.W.: Preliminary observations of gangliosides of normal and multiple sclerosis cerebrospinal fluid. *Neurology*, 23: 945-948 (1973).
13. Svennerholm, L.: Gangliosides, in Lajtha, A. (ed): *Handbook of Neurochemistry*, Vol 3. Metabolic Reactions in the Nervous System, (Plenum Press, New York, 1970) pp. 425-452.
14. Yu, R.K., and Ledeen, R.W.: Gas liquid chromatographic assay of lipid bound sialic acids: Measurement of gangliosides in brain of several species. *J Lipid Res*, 11: 506-516 (1970).
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TABLE 1
LUMBAR CEREBROSPINAL FLUID GM₁ GANGLIOSIDE CONCENTRATION OF NEONATES

PATIENT SEX/WT. GEST. AGE APGAR		NURSERY STATUS	LUMBAR CEREBROSPINAL FLUID					DISCHARGE STATUS
			PROTEIN (mg/dl)	GLUCOSE (mg/dl)	WBC (mm ⁻³)	RBC (mm ⁻³)	GM1 GANGLIOSIDE CONCENTRATION (ng/ml)	
Case 4 F/2860 40 wks	8/10	Amnionitis Lethargy Meconium Stain	18	76/90	NA	NA	58.7	Well
Case 5 F/1750 34 wks.	8/9	Prematurity Metabolic Acidosis Hyperbilirubinemia	220	47/55	5L	3600	70.7	Well
Case 6 M/2080 36 wks	9/10	Maternal Diabetes 30 hr Prom.	132	60/NA	2L	160	55.2	Well
Case 7 F/3560 40 wks	8/9	Amnionitis 48 hr Prom. Fever	108	58/NA	9L,2P	TNTC	99.4	Well
Case 8 F/4525 40 wks	1/8	Resp. Distress Meconium Stain Fever, C-Section	72	38/NA	4L,36P	350	97.2	Well
Case 9 M/3840 40 wks	8/10	34 hr. Prom.	127	60/66	2L,6P	40	110.4	Well
Case 10 M/2380 NA	NA	Tachypnea, Acidosis Cyanosis, Jittery Hypoglycemia Post-Mature	70	67/74	1L	0	46.4	Well

TABLE 1 (continued part 2)
LUMBAR CEREBROSPINAL FLUID G_{M1} GANGLIOSIDE CONCENTRATION OF NEONATES

PATIENT SEX/WT. GEST. AGE	APGAR	NURSERY STATUS	LUMBAR CEREBROSPINAL FLUID					DISCHARGE STATUS
			PROTEIN (mg/dl)	GLUCOSE (mg/dl)	WBC (mm ⁻³)	RBC (mm ⁻³)	G _{M1} GANGLIOSIDE CONCENTRATION (ng/ml)	
Case 11 M/4160 40 wks	NA	Fever Hypocalcemia	80	44/66	NA	NA	90.7	Well
Case 12 F/NA NA	NA	Physiological Jaundice	88	81/83	7L	7	90.7	Well
Case 13 F/2640 NA	8/9	Prom Maternal Cervical Strep	480	39/80	NA	TNTC	129.6	Well
Case 14 M/3540 NA	9/10	Jaundice Heart Murmur	56	80/NA	NA	NA	59.2	Asymptomatic Heart Murmur
Case 15 M/2880 38 wks	7/9	Cyanosis, Tachypnea Hypocalcemia Inguinal Hernia	204	39/104	NA	290	58.3	(L) Inguinal Hernia
Case 16 M/3060 38 wks	9/10	Tachypnea Cardiac Murmur CHF	42	52/91	0	2	35.8	Controlled CHF

TABLE 1 (continued part 3)
LUMBAR CEREBROSPINAL FLUID G_{M1} GANGLIOSIDE CONCENTRATION OF NEONATES

PATIENT SEX/WT. GEST. AGE	APGAR	NURSERY STATUS	LUMBAR CEREBROSPINAL FLUID					DISCHARGE STATUS
			PROTEIN (mg/dl)	GLUCOSE (mg/dl)	WBC (mm ⁻³)	RBC (mm ⁻³)	G _{M1} GANGLIOSIDE CONCENTRATION (ng/ml)	
Case 17 F/2420 36 wks	7/9	Prematurity, Apnea Hyponatremia, Dehydration, Hyperbilirubinemia	104	42/NA	4P	66	40.6	Well
Case 18 M/3459 NA	NA	Fever	N1	N1	0	0	73.8	Well
Case 19 M/3856 40 wks	NA	Urinary Tract Infection	90	63/103	3L	0	108.9	Well
Case 20 F/3720 40 wks	8/9	C-Section Fever Oral Thrush	110	40/78	4L	40,000	100.0	Well

All lumbar punctures were obtained prior to 4 days of age except those of cases 18, 19 and 20 which were at 12, 26 and 27 days of age, respectively. Abbreviations: Wt. weight (gm); Gest. Age, gestational age by exam; APGAR, expressed as 1 minute/5 minute values; Prom, premature rupture of membranes; L, lymphocytes; and P, polymorphonuclear leukocytes. Glucose is expressed as CSF/serum values. NA, not available; N1, normal.

TABLE 2
LUMBAR CEREBROSPINAL FLUID G_{M1} GANGLIOSIDE CONCENTRATION OF PEDIATRIC PATIENTS

PATIENT AGE/SEX	CLINICAL DATA	LUMBAR CEREBROSPINAL FLUID					DISCHARGE STATUS
		PROTEIN (mg/dl)	GLUCOSE (mg/dl)	WBC (mm ⁻³)	RBC (mm ⁻³)	G _{M1} GANGLIOSIDE CONCENTRATION (ng/ml)	
Case 21 5 wk/M	Fever Respiratory Infection	58	43/100	6L	0	14.0	
Case 22 6 wk/M	Pneumonia	84	51/89	0	0	18.0	
Case 23 2 mo/F	Fever, Bronchiolitis	84	71/106	2	7200	70.0	
Case 24 3 mo/M	Pneumonia, Anemia, Hypothyroidism	34	55/82	3L,1P	12	63.0	
Case 25 7 mo/M	S/P Patent Ductus Repair Pneumonia	28	79/NA	3	1	17.3	
Case 26 10 mo/M	Febrile Fit, Cough, Fever (T=106°F)	10	91/90	NA	NA	34.0	
Case 27 13 mo/M	Febrile Fit, Fever (T=104°F) Bilateral Otitis	28	86/90	2L,1P	160	28.0	
Case 28 3 yr/F	Fever (T=104°F), Otitis Media Abdominal Pain	28	55/100	0	0	4.8	
Case 29 18 yr/F	Viral Syndrome, Headache, Fever	N1	N1	0	0	37.6	
Case 30 3 mo/F	Moebius Syndrome	22	56/NA	0	0	44.0	

TABLE 2 (continued part 2)
LUMBAR CEREBROSPINAL FLUID G_{M1} GANGLIOSIDE CONCENTRATION OF PEDIATRIC PATIENTS

PATIENT AGE/SEX	CLINICAL DATA	LUMBAR CEREBROSPINAL FLUID				
		PROTEIN (mg/dl)	GLUCOSE (mg/dl)	WBC (mm ⁻³)	RBC (mm ⁻³)	G _{M1} GANGLIOSIDE CONCENTRATION (ng/ml)
Case 31 3 mo/M	Tonic-Clonic Seizures S/P Subarachnoid Hemorrhage	244	44/80	20L	100	94.5
Case 32 3 mo/M	Rubinstein-Taybi Syndrome Irritability, Hypertonia	34	78/91	NA	NA	79.6
Case 33 3.5 mo/F	Viral Meningitis Fever, Irritability	20	54/84	37L	3	65.9
Case 34 15 mo/M	Akinetic Seizures Wide based Gait	22	55/58	4L	3	6.1
Case 35 3 yr/M	Tonic-Clonic Seizures	22	76/92	1L	0	71.0
Case 36 6 yr/F	Mental Retardation, Motor Delay, Progressive Tremor	10	63/NA	1L	0	22.3
Case 37 7 yr/M	Recurrent Meningismus, Head- ache, Lethargy, Diplopia, Ataxia, Elevated CSF IgG and MBP	16	56/NA	17L	18	41.2

Glucose is expressed as CSF/serum values. Abbreviations: L, lymphocytes; P, polymorphonuclear leukocytes; NA, not available.

TABLE 3
SERIAL G_{M1} GANGLIOSIDE AND MYELIN BASIC PROTEIN CONCENTRATIONS IN THE LUMBAR CSF OF NEONATES

PATIENT	AGE (DAYS)	LUMBAR CEREBROSPINAL FLUID					MYELIN BASIC PRO- TEIN CONCENTRATION (ng/ml)
		PROTEIN (mg/dl)	GLUCOSE (mg/dl)	WBC (mm ⁻³)	RBC (mm ⁻³)	G _{M1} GANGLIOSIDE CONCENTRATION (ng/ml)	
Case 1	1	102	58	NA	NA	53.1	<2
	5	90	47	9	6	52.2	<2
Case 2	1	175	30	7	3780	65.2	<2
	3	490	56	20	9000	ND	ND
	6	96	43	70	8	113.4	ND
	12	104	18	11	0	102.6	<2
	18	75	100	0	4800	ND	ND
	24	84	40	4	49	75.6	<2
	43	113	46	6	600	77.3	ND
Case 3	1	540	64	ND	ND	80.1	<2
	5	ND	ND	27	23,600	157.5	6.4

Abbreviations: NA, not available; ND, not assayed.