Intraventricular haemorrhage and haemostasis defects

D W BEVERLEY, G W CHANCE, M J INWOOD, M SCHAUS, AND B O'KEEFE

Departments of Paediatrics, Obstetrics and Gynaecology, and Haematological Services, St Joseph's Hospital, London, Ontario, Canada

SUMMARY Twenty five of 106 preterm infants of 34 weeks' gestation or less developed intraventricular haemorrhage within the first 48 hours of life. A comparison of infants with and without intraventricular haemorrhage showed no significant differences in their haemostatic parameters at birth. At age 48 hours the group with intraventricular haemorrhage showed a prolonged activated partial thromboplastin time and reduced factor II, VII, and X activity. There was a significant correlation between the severity of intraventricular haemorrhage and the degree of haemostasis abnormality both in cord blood and in blood obtained at age 48 hours. Those infants sustaining grade IV intraventricular haemorrhage had a significantly prolonged activated partial thromboplastin time, reduced factor II, VII, and X activity; and a decreased fibrinogen concentration at birth. At age 48 hours these defects were accompanied by reduced platelet counts and an increased megathrombocyte index. Although intraventricular haemorrhage is multifactorial, we postulate that correction of haemostasis abnormalities at birth may prevent progression to more severe grades of haemorrhage.

Many factors have been implicated in the formation of intraventricular haemorrhage¹⁻³ and the aetiology is most probably multifactorial. Ninety two per cent of all intraventricular haemorrhages occur in infants of less than 35 weeks' gestation.⁴ Haemostasis defects have been associated with the development of intraventricular haemorrhage. Published reports⁵⁻⁸ have suggested that vitamin K₁ coagulation dependent factors are reduced in infants who have had intracranial haemorrhages. Treatment with fresh frozen plasma has been recommended to correct the coagulation deficit; this suggestion has not been pursued vigorously and it is unclear if treatment has altered mortality. More recently defects in platelet function have also been described.⁹ The present study was designed to examine prospectively the relation between defects in the coagulation, plasminolytic, and platelet systems and the occurrence of intraventricular haemorrhage in infants of 34 weeks' gestation or less.

Patients and methods

Patients. A total of 106 infants of 34 weeks' gestation or less born in St Joseph's Hospital were studied after obtaining informed parental consent.

The birthweight of the study population was mean (SD) 1.67 (0.58) kg (range 0.6 to 3.49 kg) and the gestational age was mean (SD), 31.2 (3.12) weeks (range 24 to 34 weeks). Infants were excluded if there was a maternal history of ingestion of drugs such as aspirin and warfarin that might have altered the coagulation studies. Infants weighing less than 1500 g and those weighing 1500 g or more received vitamin K₁ intramuscularly in doses of 0.5 mg and 1 mg respectively shortly after birth. None of the infants received indomethacin during the study period. Blood products including fresh frozen plasma, albumin, and whole blood were given according to the infants' clinical status. When arterial catheters were used the infusion fluid contained 1 U/ml heparin and infusion rates did not exceed 1 ml/hour.

Detection of intracranial haemorrhages. Intraventricular and periventricular haemorrhages were detected using a Diasonics 20S ultrasound detector (Diasonics, Canada) fitted with a 7.5 MHz transducer. Serial craniosonograms were performed at 8 hour intervals in the first 24 hours of life, at 12 hour intervals in the second 24 hours, and weekly thereafter. Haemorrhages were graded according to

the classification of Papile¹⁰ as follows: grade I, subependymal; grade II, intraventricular without dilatation; grade III, intraventricular with dilatation; and grade IV, intracerebral.

Blood sample collection. Blood was collected at birth. The umbilical cord was double clamped and 2.25 ml blood was drawn immediately from the umbilical vein; 0.45 ml of blood was added to 0.05 ml of 1.8% ethylenediaminetetra-acetic acid, mixed gently, and used for platelet studies; the remaining 1.8 ml of blood was added to 0.2 ml of 0.11 M sodium citrate and mixed gently. The latter sample was centrifuged at 1520 g for 15 minutes; the plasma was then removed and frozen at -70°C for batch analysis.

At age 48 hours a further 2.25 ml of blood was obtained from the neonates by direct radial artery puncture except in infants who had an indwelling arterial catheter. In these infants the sample was collected from the catheter after taking the following precautions to avoid contamination with heparin. The catheter was first flushed with 2 ml of 0.9% saline which was then withdrawn and discarded. Three ml of blood were drawn, a separate plastic syringe was then used to withdraw 0.45 ml of blood into ethylenediaminetetra-acetic acid, and another syringe was used to withdraw a further 1.8 ml of blood into sodium citrate. The first 3 ml of blood was then returned to the infants. These 48 hour samples were anticoagulated and separated in the same way as the cord venous sample. For technical reasons it was not possible to obtain cord blood in 7 infants and in 11 infants a sample was not taken at age 48 hours. Thus paired samples were obtained from 20 infants who developed intraventricular haemorrhage and 68 infants who did not.

Haemostasis investigations. The ethylenediaminetetra-acetic acid sample was used for platelet studies. A total platelet count was performed using a Coulter S Plus counter (Coulter Electronics, Canada) and verified by a manual count. The mean platelet volume was derived from the Coulter counter. The platelet size was determined using an ocular micrometer and the megathrombocyte index was calculated after the method of Garg *et al.*¹¹

The activated partial thromboplastin time was determined using an automated reagent (General Diagnostics, Canada) and clot formation was detected using a semi-automated clot timer (Dual Fibrometer Systems, USA). The combined factor II, VII, X assay was performed using commercially available factor II, VII, and X deficient plasma (Sigma Chemicals Company, USA) and converted

to percentage activity from a previously prepared calibration curve. A clottable protein fibrinogen assay was determined using thrombin (Parke Davis, Canada) and a previously prepared calibration curve.

The α_2 antiplasmin and plasminogen activity were both performed using a proteolytic enzyme detection system (Dade Division, USA). The α_2 antiplasmin was measured indirectly by the percentage of plasmin activity inhibited by the patient's plasma while the plasminogen was directly assayed.

In infants who had a prolonged activated partial thromboplastin time, a thrombin time and reptilase time were performed to exclude heparin contamination.

Statistical methods. Basic distributional characteristics of each of the study variables were examined. The primary statistical techniques for analysis were the Student's *t* test for group comparisons and the analysis of variance for a one way classification.

Results

Twenty five of the 106 infants studied sustained 38 intraventricular haemorrhages. The haemorrhage was unilateral in 12 infants and bilateral in 13 and was first detected during the first 24 hours of life in 17 infants. Eight infants died, 6 of whom had intraventricular haemorrhage; all deaths occurred after age 48 hours. The six infants who died in the group with intraventricular haemorrhage sustained either grade III or grade IV haemorrhages. There were no statistical differences between the groups in any of the clotting factors measured at birth, however, at age 48 hours those with intraventricular haemorrhages showed significant prolongation of the activated partial thromboplastin time and reduced factor II, VII, X activity (Table 1).

There were five infants with grade I, two with grade II, 13 with grade III, and five with grade IV intraventricular haemorrhages. For the purpose of analysis the results for the two infants with grade II haemorrhages were combined with those of the 13 with grade III haemorrhages. The infants with grade III and IV haemorrhages were significantly smaller and more immature than those who had no intraventricular haemorrhage or grade I haemorrhage (Table 2). Analysis of the results according to grade of haemorrhage is shown for cord venous blood and for the blood sample obtained at age 48 hours (Table 3). The infants with more severe haemorrhages had evidence of a coagulopathy at birth. There was prolongation of the activated partial thromboplastin time, reduced activity of factors II,

Table 1 Coagulation studies at birth and age 48 hours in infants with and without intraventricular haemorrhage (IVH)

	APTT (sec)	Factor II, VII, X (% activity)	Fibrinogen (g/l)	α_2 antiplasmin (% inhibition)	Platelets ($\times 10^9/l$)	MPV (fl)	MI (%)	Plasminogen (CTA U/ml)
<i>Cord blood sample</i>								
No IVH (n=81)								
Mean	54.4	41.6	1.71	73.7	226	8.05	19.8	0.90
(SD)	(13.4)	(10.0)	(0.9)	(16.7)	(56.3)	(0.95)	(12.7)	(0.33)
No investigated	74	74	74	75	75	75	74	75
IVH (n=25)								
Mean	62.8	38.5	1.92	74.0	209	8.29	23.9	0.81
(SD)	(26.7)	(13.9)	(1.19)	(14.7)	(103.3)	(0.83)	(13.8)	(0.27)
No investigated	23	24	24	24	24	23	23	24
<i>48 hour sample</i>								
No IVH (n=81)								
Mean	42.4*	50.2†	2.18	81.6	246	8.17	20.6	0.92
(SD)	(7.5)	(12.8)	(0.67)	(16.9)	(89.5)	(0.88)	(12.5)	(0.34)
No investigated	72	74	74	74	73	74	73	74
IVH (n=25)								
Mean	60.0*	41.9†	1.93	79.6	188	8.5	27.0	0.82
(SD)	(23.9)	(21.1)	(1.13)	(19.6)	(126)	(0.93)	(13.7)	(0.33)
No investigated	21	21	21	21	21	20	20	21

* $t=5.5$, $P<0.001$; † $t=2.25$, $P<0.05$.

APPT=activated partial thromboplastin time; MPV=mean platelet volume; MI=megathrombocyte index.

Conversion—SI to traditional units: fibrinogen 1 g/l=100 mg/100 ml.

Table 2 Gestational age and weight of study population according to severity of intraventricular haemorrhage (IVH)

Grade of IVH	No of patients	Gestational age* Mean (SD) (wks)	Weight† Mean (SD) (kg)
No IVH	81	32.3 (2.1)	1.83 (0.49)
Grade I	5	31.6 (2.14)	1.80 (0.58)
Grades II and III	15	27.1 (2.63)	1.00 (0.43)
Grade IV	5	28.7 (2.5)	1.23 (0.32)

* $F=36.15$, $P<0.001$; † $F=18.39$, $P<0.001$.

VII, and X, and decreased concentrations of fibrinogen in the cord blood. At age 48 hours these abnormalities persisted and in addition there was a fall in the platelet count and an increase in the megathrombocyte index. Abnormalities in the haemostatic parameters could not be accounted for completely by gestational age effects as the values obtained were outside our reported normal range.¹²

There were no changes in the mean platelet volume, α_2 antiplasmin, and plasminogen with severity of intraventricular haemorrhage at either birth or age 48 hours. We were unable to show a relation between the timing of intraventricular haemorrhage and the severity of the coagulopathy present.

Discussion

Coagulopathy has been associated with the occurrence of intraventricular haemorrhage.⁵⁻⁸ This study

shows that at age 48 hours there was prolongation of the activated partial thromboplastin time and decreased activity of factors II, VII, and X in those infants who sustained haemorrhages. Prolongation of the activated partial thromboplastin time may be attributed to various causes:

(1) Infants who sustained intraventricular haemorrhages were very immature, and hepatic unresponsiveness to vitamin K₁ has been described in similar infants^{13 14} resulting in prolonged activated partial thromboplastin time.

(2) Many of the infants had indwelling arterial catheters which were anticoagulated with heparin and it is possible that the sick neonate is unable to clear heparin as efficiently as more mature healthy infants and so becomes mildly heparinized.¹⁵

(3) Prolongation of the activated partial thromboplastin time may be caused by a consumptive process secondary to the haemorrhage or due to disseminated intravascular coagulation.¹⁶

These results also show that in infants who developed intraventricular haemorrhages the severity correlated with the degree of coagulopathy. This coagulopathy was present at birth and was detectable in cord blood. The abnormal haemostasis results detected at birth could have been caused by fetal illness compounded by hepatic immaturity, or alternatively the haemostasis abnormalities could have reflected pathology within the placenta which gave rise to disseminated intravascular coagulation in utero.

By age 48 hours the coagulation abnormalities

Table 3 Coagulation studies (mean (SD)) at birth and at age 48 hours: relation to grade of intraventricular haemorrhage (IVH)

Investigation	Time	No IVH (n=81)	Grade of IVH		II & III (n=15)	IV (n=5)	Significance
			I (n=5)				
Activated partial thromboplastin time (sec)	Birth	54.4 74	43.7 4	(10.7)	62.3 14	79.4 5	F=4.61; P<0.01
	48 h	42.4 72	52.0 5	(19.4)	50.0 11	89.8 5	F=34.1; P<0.001
Factors II, VII, X (% activity)	Birth	41.6 74	48.8 5	(15.6)	38.8 14	27.4 5	F=3.92; P<0.025
	48 h	50.2 74	51.6 5	(25.0)	42.1 11	31.6 5	F=3.26; P<0.05
Fibrinogen (g/l)	Birth	1.71 74	2.85 5	(1.49)	1.99 14	0.78 5	F=4.5; P<0.01
	48 h	2.18 74	3.01 5	(1.33)	1.97 11	0.77 5	F=8.5; P<0.001
α_2 antiplasmin (% inhibition)	Birth	73.7 75	83.2 5	(8.8)	73.0 14	67.8 5	NS
	48 h	81.6 74	95.6 5	(9.0)	78.5 11	66.2 5	NS
Platelets ($\times 10^9/l$)	Birth	226 75	232 5	(127.5)	222 14	149 5	NS
	48 h	246 73	281 5	(132.2)	195 11	79.0 5	F=6.05; P<0.001
Mean platelet volume (fl)	Birth	8.05 75	8.62 5	(1.4)	8.17 13	8.28 5	NS
	48 h	8.17 74	8.65 4	(1.56)	8.34 11	8.76 5	NS
Megathrombocyte index (%)	Birth	19.8 74	20.3 4	(9.3)	24.3 14	25.6 5	NS
	48 h	20.6 73	19.0 5	(9.0)	23.1 10	42.8 5	F=5.26; P<0.01
Plasminogen (CTA U/ml)	Birth	0.9 75	0.86 5	(0.41)	0.87 14	0.62 5	NS
	48 h	0.92 74	0.66 5	(0.29)	1.01 11	0.60 5	NS

Figures in italics are the number of infants in whom each investigation was undertaken.
NS = not significant.

had progressed to a consumptive process with a fall in peripheral platelet count and an increase in the megathrombocyte index. We were unable to determine from our results whether the changes present at age 48 hours were a continuation of the process initiated before birth and detectable in cord blood or whether they were secondary phenomena caused by the intraventricular haemorrhage itself. The fact that infants with grade I haemorrhage tended to have a more efficient haemostatic mechanism than those infants who had no haemorrhage supports our belief that coagulopathy itself is not the cause of intraventricular haemorrhage. We suggest that in those infants sustaining intraventricular haemorrhage its subsequent size will be determined partly by the ability of the coagulation cascade to restore vascular integrity.

Follow up studies show that infants who survive grade III or IV intraventricular haemorrhages will have a risk of permanent brain damage of approximately 40%.^{17,18} The presence of a coagulopathy at birth in infants who subsequently develop grade III and IV intraventricular haemorrhages suggests that amelioration may be possible through correction of the coagulation disorder. Results of intervention studies reported to date^{6,7} have been equivocal but these were performed before the ultrasound era when accurate diagnosis of intraventricular haemorrhage was difficult. We hypothesise that if the abnormalities of coagulation present at birth can be corrected, extension of the intraventricular haemorrhage may be limited, thereby decreasing its resultant mortality and morbidity.

We thank Dr G Wells for statistical assistance, the consultant obstetrical and neonatal staff of St Joseph's Hospital for permission to study their patients, and the nursing staff of the delivery room and the neonatal intensive care unit for patient cooperation. DWB was funded by St Joseph's Hospital Foundation.

References

- Volpe JJ. Neonatal periventricular hemorrhage: past, present and future. *J Pediatr* 1979;**92**:693-6.
- Tarby TJ, Volpe JJ. Intraventricular hemorrhage in the premature infant. *Pediatr Clin North Am* 1982;**29**:1077-104.
- Volpe JJ. Neonatal intraventricular hemorrhage. *N Engl J Med* 1981;**304**:886-91.
- Garcia-Pratts JA, Procianny RS, Adams JM, Rudolph AJ. The hyaline membrane disease. Intraventricular hemorrhage relationship in the very low birth weight infant: perinatal aspects. *Acta Paediatr Scand* 1982;**71**:79-84.
- Gray OP, Ackerman A, Fraser AJ. Intracranial haemorrhage and clotting defects in low birth weight infants. *Lancet* 1968;**i**:545-8.
- Thomas DB, Burnard ED. Prevention of intraventricular haemorrhage in babies receiving artificial ventilation. *Med J Aust* 1973;**1**:933-6.
- Hambleton G, Appleyard WJ. Controlled trial of fresh frozen plasma in asphyxiated low birth weight infants. *Arch Dis Child* 1973;**48**:31-5.
- Gupta JM, Starr H, Fincher P, Lam-Po-Tang PRLC. Intraventricular haemorrhage in the newborn. *Med J Aust* 1976;**2**:338-40.
- Setzer ES, Webb IB, Wassenaar JW, Reeder JD, Mehta PS, Eitzman DV. Platelet dysfunction and coagulopathy in intraventricular hemorrhage in the premature infant. *J Pediatr* 1982;**100**:599-605.
- Papile L, Burstein J, Burstein R, Koffler H. Incidence and evolution of subependymal and intraventricular hemorrhage: a study of infants with birth weights less than 1500 grams. *J Pediatr* 1978;**92**:529-34.
- Garg SK, Amorosi EL, Karparkin S. Use of megathrombocyte as an index of megakaryocyte number. *N Engl J Med* 1971;**284**:11-7.
- Beverley DW, Inwood MJ, Chance GW, Schaus M, O'Keefe B. 'Normal' haemostasis parameters: a study in a well-defined inborn population of preterm infants. *Early Hum Dev* (in press).
- Oski FA, Naiman JL, Stockman JA, Pearson HA. *Hematological problems in the newborn*. Philadelphia: WB Saunders, 1982.
- Aballi AJ Lopez Banus V, De Lamerens S, et al. Coagulation studies in the newborn period. I. Alterations of thromboplastin gestation and effects of Vitamin K on full term and preterm infants. *Am J Dis Child* 1957;**94**:594-601.
- Galant SP. Accidental heparinization of a newborn infant. *Am J Dis Child* 1967;**114**:313-9.
- Woods GW, Luban NLC, Hilgartner MW, Miller DR. Disseminated intravascular coagulation in the newborn. *Am J Dis Child* 1979;**133**:44-6.
- Krishnamoorthy KS, Shannon DC, DeLong GR, Todres ID, Davis KR. Neurologic sequelae in the survivors of neonatal intraventricular haemorrhage. *Pediatrics* 1979;**64**:233-7.
- Shankaran S, Slovis TL, Bedard MP, Poland RL. Sonographic classification of intracranial hemorrhage. A prognostic indicator of mortality, morbidity and short term neurologic outcome. *J Pediatr* 1982;**100**:469-75.

Correspondence to Dr D Beverley, Department of Paediatrics, Leeds General Infirmary, Leeds LS1 3EX.

Received 23 January 1984