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Guideline

THE INVESTIGATION AND MANAGEMENT OF NEONATAL HAEMOSTASIS AND THROMBOSIS*

SUMMARY

These guidelines address developmental aspects of neonatal haemostasis and thrombosis, the laboratory investigation of the neonate, and the diagnosis and clinical management of haemostatic and thrombotic conditions occurring in this period (defined as the first 4 weeks of life following birth). Relevant scientific papers were identified by a systematic literature review from Medline 1975–2000 using index terms which incorporated the various component subjects of these guidelines. Further publications were obtained from the references cited and from reviews known to the members of the working party and to the Haemostasis and Thrombosis Task Force. Evidence and graded recommendations presented in these guidelines are in accordance with the US Agency for Health Care Policy and Research, as described in the *Appendix*. It will be noted that there is a lack of a strong evidence base for many of the recommendations suggested, as the appropriate clinical and laboratory trials have not been and perhaps never will be undertaken in neonates. Most of the recommendations are therefore of Grade C evidence levels IV: higher levels are mentioned specifically throughout the document when relevant.

INTRODUCTION

The human haemostatic system is dynamic and is profoundly influenced by age. Although considered immature in the newborn, it is a physiological system which results in few problems for the healthy term neonate, but may contribute to morbidity in the sick and preterm infant when additional acquired abnormalities may be present. Many of the procoagulants, anticoagulants and proteins involved in fibrinolysis are gestation dependent. The newborn's haemostatic system matures during the early weeks and months of life with most haemostatic parameters reaching adult values by 6 months of age. In addition, there is evidence of an accelerated maturation pattern in the premature infant with preterms showing similar levels of coagulation proteins to term infants by 6 months of age. Values for coagulation parameters in the infant are therefore dependent to a varying extent on both the gestational and postnatal age of the infant and are shown in Tables I–VI (Andrew *et al*, 1987, 1988). An understanding of these age-related ranges is essential to the interpretation of neonatal investigations.

Correspondence: Michael D. Williams, Department of Haematology, Birmingham Children's Hospital, Steelhouse Lane, Birmingham B4 6NH, UK. E-mail: mike.williams@bhamchildrens.wmids.nhs.uk

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1. INDICATIONS FOR HAEMOSTATIC OR THROMBOTIC TESTING OF THE NEONATE

Haemostasis

The majority of bleeding problems in the neonatal period are acquired but inherited coagulation disorders may also present at this time, particularly following iatragenic challenges. Generally, testing for defects of haemostasis is indicated in all sick neonates, which would include all admissions to the neonatal intensive care unit. More specifically, screening is carried out in the following circumstances:

- Any haemorrhagic neonate
- A family history of an inherited bleeding disorder (dependent on the coagulation factor defect, the severity of the deficiency and the likelihood of an accurate diagnosis in the neonate)
- Severe metabolic disease, severe respiratory distress syndrome, liver dysfunction or other predisposing factors for disseminated intravascular coagulation (DIC)
- Babies whose mothers were taking anticonvulsants, warfarin or antituberculous drugs at the time of delivery
- All neonates undergoing surgery or tissue biopsy who have had previous bleeding problems.

Thrombosis

Congenital thrombophilia should be considered in:

- Any child with a clinically significant thrombosis, including spontaneous thrombotic events, unanticipated or extensive venous thrombosis, ischaemic skin lesions or purpura fulminans
- A positive family history of neonatal purpura fulminans.

Asymptomatic neonates should not be investigated unless there is a significant medical history such as previous neonatal purpura fulminans or thrombosis. Parents need counselling before any investigations are performed either on themselves or their children so that the consequences of testing asymptomatic individuals can be fully explained.

2. LABORATORY INVESTIGATION OF NEONATAL HAEMOSTASIS, THROMBOSIS AND FIBRINOLYSIS

Laboratory investigation should identify neonates with inherited or acquired disorders of coagulation. The physiological deficiencies of procoagulant and anticoagulant

Table I. Reference values for coagulation tests in the healthy full-term infant during the first 6 months of life.

Tests	Day 1 (*n*)	Day 5 (*n*)	Day 30 (*n*)	Day 90 (*n*)	Day 180 (*n*)	Adult (*n*)
PT(s)	13·0 ± 1·43 (61)*	12·4 ± 1·46 (77)*†	11·8 ± 1·25 (67)*†	11·9 ± 1·15 (62)*	12·3 ± 0·79 (47)*	12·4 ± 0·78 (29)
APTT(s)	42·9 ± 5·80 (61)	42·6 ± 8·62 (76)	40·4 ± 7·42 (67)	37·1 ± 6·52 (62)*	35·5 ± 3·71 (47)*	33·5 ± 3·44 (29)
TCT(s)	23·5 ± 2·38 (58)*	23·1 ± 3·07 (64)†	24·3 ± 2·44 (53)*	25·1 ± 2·32 (52)*	25·5 ± 2·86 (41)*	25·0 ± 2·66 (19)
Fibrinogen (g/l)	2·83 ± 0·58 (61)*	3·12 ± 0·75 (77)*	2·70 ± 0·54 (67)*	2·43 ± 0·68 (47)*†	2·51 ± 0·68 (47)*†	2·78 ± 0·61 (29)
II(U/ml)	0·48 ± 0·11 (61)	0·63 ± 0·15 (76)	0·68 ± 0·17 (67)	0·75 ± 0·15 (62)	0·88 ± 0·14 (47)	1·08 ± 0·19 (29)
V (U/ml)	0·72 ± 0·18 (61)	0·95 ± 0·25 (76)	0·98 ± 0·18 (67)	0·90 ± 0·21 (62)	0·91 ± 0·18 (47)	1·06 ± 0·22 (29)
VII (U/ml)	0·66 ± 0·19 (60)	0·89 ± 0·27 (75)	0·90 ± 0·24 (67)	0·91 ± 0·26 (62)	0·87 ± 0·20 (47)	1·05 ± 0·19 (29)
VIII (U/ml)	1·00 ± 0·39 (60)*†	0·88 ± 0·33 (75)*†	0·91 ± 0·33 (67)*†	0·79 ± 0·23 (62)*†	0·73 ± 0·18 (47)†	0·99 ± 0·25 (29)
VWF (U/ml)	1·53 ± 0·67 (40)†	1·40 ± 0·57 (43)†	1·28 ± 0·59 (40)†	1·18 ± 0·44 (40)†	1·07 ± 0·45 (46)†	0·92 ± 0·33 (29)†
IX (U/ml)	0·53 ± 0·19 (59)	0·53 ± 0·19 (75)	0·51 ± 0·15 (67)	0·67 ± 0·23 (62)	0·86 ± 0·25 (47)	1·09 ± 0·27 (29)
X (U/ml)	0·40 ± 0·14 (60)	0·49 ± 0·15 (76)	0·59 ± 0·14 (67)	0·71 ± 0·18 (62)	0·78 ± 0·20 (47)	1·06 ± 0·23 (29)
XI (U/ml)	0·38 ± 0·14 (60)	0·55 ± 0·16 (74)	0·53 ± 0·13 (67)	0·69 ± 0·14 (62)	0·86 ± 0·24 (47)	0·97 ± 0·15 (29)
XII (U/ml)	0·53 ± 0·20 (60)	0·47 ± 0·18 (75)	0·49 ± 0·16 (67)	0·67 ± 0·21 (62)	0·77 ± 0·19 (47)	1·08 ± 0·28 (29)
PK (U/ml)	0·37 ± 0·16 (45)	0·48 ± 0·14 (51)	0·57 ± 0·17 (48)	0·73 ± 0·16 (46)	0·86 ± 0·15 (43)	1·12 ± 0·25 (29)
HMW-K (U/ml)	0·54 ± 0·24 (47)	0·74 ± 0·28 (63)	0·77 ± 0·22 (50)*	0·82 ± 0·32 (46)*	0·82 ± 0·23 (48)*	0·92 ± 0·22 (29)
XIIIa (U/ml)	0·79 ± 0·26 (44)	0·94 ± 0·25 (49)*	0·93 ± 0·27 (44)*	1·04 ± 0·34 (44)*	1·04 ± 0·29 (41)*	1·05 ± 0·25 (29)
XIIIb (U/ml)	0·76 ± 0·23 (44)	1·06 ± 0·37 (47)*	1·11 ± 0·35 (45)*	1·16 ± 0·34 (44)*	1·10 ± 0·30 (41)*	0·97 ± 0·20 (29)

*Values that do not differ statistically from the adult values.

†These measurements are skewed because of a disproportionate number of high values.

All values expressed as mean ± 1 SD. (*n*) = numbers studied. All factors except fibrinogen are expressed as U/ml where pooled plasma contains 1·0 U/ml. PK, Prekallikrein; HMW-K, high-molecular-weight kininogen.

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Table II. Reference values for the inhibition of coagulation in the healthy full-term infant during the first 6 months of life.

Inhibitors	Day 1 (*n*)	Day 5 (*n*)	Day 30 (*n*)	Day 90 (*n*)	Day 180 (*n*)	Adult (*n*)
AT	0·63 ± 0·12 (58)	0·67 ± 0·13 (74)	0·78 ± 0·15 (66)	0·97 ± 0·12 (60)*	1·04 ± 0·10 (56)*	1·05 ± 0·13 (28)
α_2-M	1·39 ± 0·22 (54)	1·48 ± 0·25 (73)	1·50 ± 0·22 (61)	1·76 ± 0·25 (55)	1·91 ± 0·21 (55)	0·86 ± 0·17 (29)
C1E-INH	0·72 ± 0·18 (59)	0·90 ± 0·15 (76)*	0·89 ± 0·21 (63)	1·15 ± 0·22 (55)	1·41 ± 0·26 (55)	1·01 ± 0·15 (29)
α_1-AT	0·93 ± 0·22 (57)*	0·89 ± 0·20 (75)*	0·62 ± 0·13 (51)	0·72 ± 0·15 (56)	0·77 ± 0·15 (55)	0·93 ± 0·19 (29)
HCII	0·43 ± 0·25 (56)	0·48 ± 0·24 (72)	0·47 ± 0·20 (58)	0·72 ± 0·37 (58)	1·20 ± 0·35 (55)	0·96 ± 0·15 (29)
Protein C	0·35 ± 0·09 (41)	0·42 ± 0·11 (44)	0·43 ± 0·11 (43)	0·54 ± 0·13 (44)	0·59 ± 0·11 (52)	0·96 ± 0·16 (28)
Protein S	0·36 ± 0·12 (40)	0·50 ± 0·14 (48)	0·63 ± 0·15 (41)	0·86 ± 0·16 (46)*	0·87 ± 0·16 (49)*	0·92 ± 0·16 (29)

*Values that do not differ statistically from the adult values.

All values expressed as mean ± 1 SD. (*n*) = numbers studied. AT, antithrombin; α_2-M, α_2-macroglobulin; C1E-INH, C1esterase inhibitor; α_1-AT, α_1-antithrombin; HC-II, Heparin Cofactor II.

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proteins can make distinguishing the pathological from the physiological difficult and the correct interpretation of such investigations is dependent on an awareness of the 'normal' levels of these proteins in the neonate and how these are influenced by gestational age, postnatal age and by external factors such as sepsis or vitamin K deficiency. In particular, the diagnosis of the majority of mild congenital factor deficiencies will need to be confirmed by repeat testing later in infancy: there is seldom any clinical indication to test for a mild deficiency in children less than 6 months old and investigation can therefore usually be deferred beyond this age. The gestation-dependent differences between neonates and adults, together with the evolving state of the newborn's coagulation system in the early weeks and months of life, necessitate sequential reference ranges which reflect the effects of gestational and postnatal age. As screening tests and factor assays are influenced by a number of variables including test methodology, reagents and instrumentation, laboratories should, wherever possible, establish their own normal ranges for neonates of different gestational ages (Hathaway & Corrigan, 1991): practically, the difficulty of recruiting suitable cohorts of neonates can preclude this approach, with laboratory reference ranges then being based on literature-derived data. The most frequently quoted data are those of Andrew *et al* (1987, 1988, 1990a) who used a standardized

Table III. Reference values for coagulation tests in healthy premature infants (30–36 weeks gestation) during first 6 months of life.

	Day 1		Day 5		Day 30		Day 90		Day 180		Adult	
Tests	M	B	M	B	M	B	M	B	M	B	M	B
PT(s)	13·0	(10·6–16·2)*	12·5	(10·0–15·3)*†	11·8	(10·0–13·6)*	12·3	(10·0–14·6)*	12·5	(10·0–15·0)*	12·4	(10·8–13·9)
APTT(s)_	53·6	(27·5–79·4)‡	50·5	(26·9–74·1)‡	44·7	(26·9–62·5)	39·5	(28·3–50·7)	37·5	(21·7–53·3)*	33·5	(26·6–40·3)
TCT(s)	24·8	(19·2–30·4)*	24·1	(18·8–29·4)*	24·4	(18·8–29·9)*	25·1	(19·4–30·8)*	25·2	(18·9–31·5)*	25·0	(19·7–30·3)
Fibrinogen (g/l)	2·43	(1·50–3·73)*†‡	2·80	(1·60–4·18)*†‡	2·54	(1·50–4·14)*†	2·46	(1·50–3·52)*†	2·28	(1·50 = 3·60)†	2·78	(1·56–4·00)
II (U/ml)	0·45	(0·20–0·77)†	0·57	(0·29–0·85)‡	0·57	(0·36–0·95)†‡	0·68	(0·30–1·06)	0·87	(0·51–1·23)	1·08	(0·70–1·46)
V (U/ml)	0·88	(0·41–1·44)*†‡	1·00	(0·46–1·54)	1·02	(0·48–1·56)	0·99	(0·59–1·39)	1·02	(0·58–1·46)	1·06	(0·62–1·50)
VII (U/ml)	0·67	(0·21–1·13)	0·84	(0·30–1·38)	0·83	(0·21–1·45)	0·87	(0·31–1·43)	0·99	(0·47–1·51)*	1·05	(0·67–1·43)
VIII (U/ml)	1·11	(0·50–2·13)*†	1·15	(0·53–2·05)*†‡	1·11	(0·50–1·99)*†‡	1·06	(0·58–1·88)*†‡	0·99	(0·50–1·87)*†‡	0·99	(0·50–1·49)
VWF (U/ml)	1·36	(0·78–2·10)†	1·33	(0·72–2·19)†	1·36	(0·66–2·16)†	1·12	(0·75–1·84)*†	0·98	(0·54–1·58)*†	0·92	(0·50–1·58)
IX (U/ml)	0·35	(0·19–0·65)†‡	0·42	(0·14–0·74)†‡	0·44	(0·13–0·80)†	0·59	(0·25–0·93)	0·81	(0·50–1·20)†	1·09	(0·55–1·63)
X (U/ml)	0·41	(0·11–0·71)	0·51	(0·19–0·83)	0·56	(0·20–0·92)	0·67	(0·35–0·59)	0·77	(0·35–1·19)	1·06	(0·70–1·52)
XI (U/ml)	0·30	(0·08–0·52)†‡	0·41	(0·13–0·69)‡	0·43	(0·15–0·71)‡	0·59	(0·25–0·93)‡	0·78	(0·46–1·10)	0·97	(0·67–1·27)
XII (U/ml)	0·38	(0·10–0·66)‡	0·39	(0·09–0·69)‡	0·43	(0·11–0·75)	0·61	(0·15–1·07)	0·82	(0·22–1·42)	1·08	(0·52–1·64)
PK (U/ml)	0·33	(0·09–0·57)	0·45	(0·26–0·75)†	0·59	(0·31–0·87)	0·79	(0·37–1·21)	0·78	(0·40–1·15)	1·12	(0·62–1·62)
HMWK (U/ml)	0·49	(0·09–0·89)	0·62	(0·24–1·00)‡	0·64	(0·16–1·12)‡	0·78	(0·32–1·24)	0·83	(0·41–1·25)*	0·92	(0·50–1·36)
XIIIa (U/ml)	0·70	(0·32–1·08)	1·01	(0·57–1·45)*	0·99	(0·51–1·47)*	1·13	(0·71–1·55)*	1·13	(0·65–1·61)*	1·05	(0·55–1·55)
XIIIb (U/ml)	0·81	(0·35–1·27)	1·10	(0·68–1·58)*	1·07	(0·57–1·57)*	1·21	(0·75–1·67)	1·15	(0·67–1·63)	0·97	(0·57–1·37)

*Values indistinguishable from those of adults.

†Measurements are skewed owing to a disproportionate number of high values.

‡Values different from those of full-term infants.

All factors except fibrinogen are expressed as U/ml, where pooled plasma contains 1·0 U/ml. All values are given as a mean (M) followed by lower and upper boundary encompassing 95% of the population (B). Between 40 and 96 samples were assayed for each value for newborns.

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Table IV. Reference values for inhibitors of coagulation in healthy premature infants during first 6 months of life.

	Day 1		Day 5		Day 30		Day 90		Day 180		Adult	
Tests	M	B	M	B	M	B	M	B	M	B	M	B
AT	0·38	(0·14–0·62)‡	0·56	(0·30–0·82)*	0·59	(0·37–0·81)‡	0·83	(0·45–1·21)‡	0·90	(0·52–1·28)‡	1·05	(0·79–1·31)
α2-M	1·10	(0·56–1·82)‡†	1·25	(0·71–1·77)*	1·38	(0·72–2·04)	1·80	(1·20–2.6)†	2·09	(1·10–3·21)†	0·86	(0·52–1·20)
C1E-INH	0·65	(0·31–0·99)	0·83	(0·45–1·21)	0·74	(0·40–1·24)†‡	1·14	(0·60–1·68)*	1·40	(0·96–2·04)†	1·01	(0·71–1·31)
α_1AT	0·90	(0·36–1·44)*	0·94	(0·42–1·46)‡	0·76	(0·38–1·12)‡	0·81	(0·49–1·13)*‡	0·82	(0·48–1·16)*	0·93	(0·55–1·31)
HCII	0·32	(0·00–0·60)‡	0·34	(0·00–0·69)*	0·43	(0·15–0·71)	0·61	(0·20–1·11)†	0·89	(0·45–1·40)*†‡	0·96	(0·66–1·26)
Protein C	0·28	(0·12–0·44)*‡	0·31	(0·11–0·51)*	0·37	(0·15–0·59)‡	0·45	(0·23–0·67)‡	0·57	(0·31–0·83)	0·96	(0·64–1·28)
Protein S	0·26	(0·14–0·38)‡	0·37	(0·13–0·61)*	0·56	(0·22–0·90)	0·76	(0·40–1·12)‡	0·82	(0·44–1·20)	0·92	(0·60–1·24)

*Values indistinguishable from those of adults.

†Measurements are skewed owing to a disproportionate number of high values. Lower limit which excludes the lower 2·5% of the population is given (B).

‡Values different from those of full-term infants.

All values are expressed in U/ml, where pooled plasma contains 1·0 U/ml. All values are given as a mean (M) followed by lower and upper boundary encompassing 95% of the population (B). Between 40 and 75 samples were assayed for each value for the newborn.

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Table V. Reference values for the components of the fibrinolytic system in healthy full-term infants during the first 6 months of life, compared with those in adults.

Fibrinolytic component	Day 1 mean (boundary)	Day 5 mean (boundary)	Day 30 mean (boundary)	Day 90 mean (boundary)	Day 180 mean (boundary)	Adults mean (boundary)
Plasminogen (U/ml)	1·95 (1·25–2·65)	2·17 (1·41–2·93)	1·98 (1·26–2·70)	2·48 (1·74–3·22)	3·01 (2·21–3·81)	3·36 (2·48–4·24)
TPA (ng/ml)	9·60 (5·0–18·9)	5·60 (4·0–10·0)*	4·10 (1·00–6·00)*	2·1 (1·0–5·0)*	2·8 (1·0–6·0)*	4·9 (1·4–8·4)
α_2AP (U/ml)	0·85 (0·55–1·15)	1·00 (0·70–1·30)*	1·00 (0·76–1·40)*	1·08 (0·76–1·40)*	1·11 (0·83–1·39)*	1·02 (0·63–1·35)
PAI (U/ml)	6·40 (2·0–15·1)	2·30 (0·0–8·10)*	3·40 (0·0–8·80)*	7·2 (1·0–15·3)	8·1 (6·0–13·0)	3·6 (0·0–11·0)

*Values indistinguishable from those of adults.

TPA, tissue plasminogen activator; α_2AP, α_2-antiplasmin; PAI, plasminogen activator inhibitor. For α_2AP, values are expressed as U/ml, where pooled plasma contains 1·0 U/ml. Plasminogen units are those recommended by the Committee on Thrombolytic Agents. Values for TPA are given as ng/ml. Values for PAI are given as U/ml, where one unit of PAI activity is defined as the amount of PAI that inhibits 1 international unit of human single-chain TPA. All values are given as a mean followed by the lower and upper boundary encompassing 95% of the population (boundary).

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Table VI. Reference values for the components of the fibrinolytic system in healthy premature infants during the first 6 months of life, compared with those in adults.

Fibrinolytic component	Day 1 mean (boundary)	Day 5 mean (boundary)	Day 30 mean (boundary)	Day 90 mean (boundary)	Day 180 mean (boundary)	Adults mean (boundary)
Plasminogen (U/ml)	1·70 (1·12–2·48)†	1·91 (1·21–2·61)†	1·81 (1·09–2·53)	2·38 (1·58–3·18)	2·75 (1·91–3·59)†	3·36 (2·48–4·24)
TPA (ng/ml)	8·48 (3·00–16·70)	3·97 (2·00–6·93)*	4·13 (2·00–7·79)*	3·31 (2·00–5·07)*	3·48 (2·00–5·85)*	4·96 (1·46–8·46)
α_2AP (U/ml)	0·78 (0·40–1·16)	0·81 (0·49–1·13)†	0·89 (0·55–1·23)†	1·06 (0·64–1·48)*	1·15 (0·77–1·53)	1·02 (0·68–1·36)
PAI (U/ml)	5·40 (0·0–12·2)*†	2·50 (0·0–7·1)*	4·30 (0·0–10·9)*	4·80 (1·0–11·8)*†	4·90 (1·0–10·2)*†	3·60 (0·0–11·0)

*Values indistinguishable from those of adults.

†Values that are different from those of the full-term infant.

All values are given as a mean followed by the lower and upper boundary encompassing 95% of the population (boundary).

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protocol and venous blood samples from healthy term and preterm infants who had received 1 mg of Vitamin K at least 12 h prior to the first blood sample being drawn (see Tables I–VI).

INVESTIGATION OF THE NEONATE

2A. Sampling

Sample collection. Present day automated coagulometers have reduced the need for laboratories to establish manual microtechniques for coagulation screening. One millilitre of blood is sufficient to perform a coagulation screen and an additional 1 ml will be necessary for subsequent procoagulant or anticoagulant assays.

Blood sampling from a neonate should avoid contamination with intravenous fluids, particularly heparin, and should also avoid activation of the coagulation process. Neonatal units should have established their own blood discard procedures to minimize the risk of contamination; activation is likely if sampling is slow through plastic tubing and will result in a shortening of the prothrombin time (PT), activated partial thromboplastin time (APTT) and thrombin time (TT). Platelet clumping is common in such samples, resulting in spurious thrombocytopenia. All neonatal samples should be inspected for fibrin strands and platelet clumps to help interpretation of results.

Sample anticoagulant. Sample tubes (1 ml) should be readily available to the Neonatal Unit. Ideally, the volume of anticoagulant in the sample tube should be based on the volume of plasma and not on the total volume of blood taken. It should therefore be reduced in proportion to the increased neonatal haematocrit in order to avoid dilution of coagulation factors and this is particularly pertinent for neonates with very high haematocrits (Corrigan, 1989). Laboratories and neonatal units usually take a more pragmatic approach to this problem and accept a degree of 'artefactual' prolongation of the coagulation times. Blood is therefore usually taken into 3·2% buffered sodium citrate, one part citrate to nine parts blood (National Committee for Clinical Laboratory Standards, 1998, *Grade C recommendation based on level IV evidence*) and the normal values detailed in Tables I–VI have been obtained using this methodology.

Overfilled or underfilled samples should not be analysed.

Sample processing. Twenty-four-hour access to laboratory support is essential, and this will include availability of factor assays.

2B. Laboratory investigation

i) Haemostasis. Screening tests of neonatal haemostasis and their interpretation are described in Table VII. The investigation and management of neonatal thrombocytopenia are not within the remit of these guidelines but have recently been the subject of Practice Guidelines (Letsky & Greaves, 1996) and of extensive review (Roberts & Murray, 1999).

Abnormal results require more specific testing of the various components of the haemostatic system and should include:

- Specific factor assays. Prolongation of the PT, APTT or TT which corrects in a 1:1 mix with normal plasma is indicative of either a single factor or multiple factor

Table VII. Screening tests of neonatal haemostasis.

Investigation	Result	Comment
1. Platelet count and morphology	$< 150 \times 10^9$/l is abnormal, see text	Platelet clumping secondary to activation is common. Always look for fibrin strands in sample. Morphology important for congenital platelet disorders such as Bernard Soulier, grey platelet syndrome
2. Prothrombin time (PT)	See text	Establish 'in-house' normal range* if possible. Prolonged by deficiencies of some Vitamin K-dependent factors.
3. Activated partial thromboplastin time (APTT)	See text	Establish 'in-house' normal range* if possible. Prolonged in healthy neonate because of relatively reduced levels of vitamin K-dependent factors and contact factors.
4. Thrombin clotting time (TCT)	See text	Prolonged compared with adult TCT because of fetal fibrinogen. Addition of calcium to the buffering system shortens time to adult range and increases sensitivity.†
5. Fibrinogen	See text	Equivalent to adult normal range but levels rise in the first week of life (NB: when screening for DIC). Discrepancies between functional and immune assays helpful in diagnosis of dysfibrinogenaemias.
6. Bleeding time (BT)	See text	Infrequently performed. Shorter than adult range. Modified template device for use in neonates.‡

*Results obtained are dependent on reagents and coagulometer used. It is therefore preferable to establish in-house normal ranges rather than rely on published data (see text).

†Will be sensitive to hypofibrinogenaemias, dysfibrinogenaemias and the presence of heparin but not to the effects of fetal fibrinogen (Ockelford & Carter, 1982).

‡Andrew *et al* (1990b).

deficiencies. The gestational age, postnatal age, vitamin K status and 'general well being' of the neonate should all be considered in the interpretation of factor assays (see later).

- Factor XIII screen. Factor XIII deficiency does not prolong any of the routine screening tests and requires specific investigation. Screening for this deficiency involves the use of calcium or thrombin to produce a fibrin clot and assessing the solubility of the clot in either 5 mol/l urea or 1% monochloracetic acid. The results of such screening tests can be variable with thrombin-based screens more sensitive to reductions in factor XIII (Jennings *et al*, 2000); factor XIII assays are not widely used in the UK but are more sensitive to lower levels of this protein (UK NEQAS for blood coagulation 1998, Survey 112, unpublished observation).
- Platelet function. The neonate's bleeding time is shorter than that of adults and of older children (Andrew *et al*, 1990b), and may be influenced by a number of variables: testing must therefore be standardized [*Grade B recommendations based on level IIa,b evidence* (Sutor, 1998)]. It is seldom indicated in the neonate and usually only performed when other more specific tests of haemostasis have failed to establish a cause of bleeding. Other 'global' tests of platelet function using instruments such as the Platelet Function Analyser (PFA)[100] may provide an alternative to the bleeding time and are currently being evaluated in neonates. Formal platelet aggregation testing in the neonate requires significant amounts of platelet-rich plasma (> 150 μl). Variable results have been reported in otherwise healthy neonates, making interpretation of results difficult and, in most cases, of little clinical significance.
- Platelet glycoprotein expression is fully developed in term and premature neonates and flow cytometry can establish the diagnosis of the congenital disorders Bernard Soulier disease and Glanzmann's Thrombasthenia using minimal amounts of blood.
- Investigation of the parents' platelet function may be helpful in selected cases.
- D-dimer. D-dimer estimation is a specific test of fibrin lysis by plasmin and is a sensitive marker of coagulation activation, including disseminated intravascular coagulation. There are very few data on normal D-dimer levels in the neonate, but these have been reported to be increased in cord blood samples even in the absence of DIC because of the activation of coagulation which occurs during delivery (Hudson *et al*, 1990).

ii) Thrombosis. The causative role of congenital deficiencies of anticoagulants in neonatal thrombosis is clear only in the homozygous/double heterozygous deficiencies of Protein C and Protein S which result in neonatal purpura fulminans: replacement of these proteins forms the basis of treatment for these conditions and the measurement of these proteins should be undertaken in all neonates who develop this condition. It is becoming increasingly recognized that there is a high prevalence of other thrombophilic defects in children who develop venous thrombosis (Junker *et al*, 1999; Lawson *et al*, 1999; Heller *et al*, 2000), although the causative nature of these defects and their influence on the natural history of the thrombotic event remain uncertain. Thrombophilic defects have also been reported in neonates with ischaemic stroke (Gunther *et al*, 2000), a condition which often accompanies perinatal asphyxia and, less commonly, hypertension and polycythaemia: again, the contributory role of such defects in these settings is uncertain. In order to justify the routine investigation of such defects, their demonstration should be shown to influence the treatment of an affected individual. At the present time this is not the case and so extensive thrombophilic screening of the neonate with venous thrombosis or cerebral infarction cannot be recommended as part of evidence-based guidelines. It is not possible, however, to make the assumption that the aetiology of thrombosis in children and adults is similar or that the response to treatment and long-term complications of thrombosis are the same in these two age groups. Clinicians should be aware that there are ongoing International and National Registries for Paediatric Thrombosis which will, in time, accumulate data and thereby inform the investigation and treatment of childhood thrombosis: the provision of thrombophilic information is likely to form an important component of such registries.

The normal ranges of anticoagulant proteins in the neonate are wide and can make interpretation of results difficult. With the provisos mentioned above, laboratory investigations may include the following, test methodology having been addressed in the British Society for Standards in Haematology (BCSH) guidelines for the investigation of thrombophilia (Walker *et al*, 2001).

- Protein C activity. Protein C is a Vitamin K-dependent protein and levels are physiologically reduced in the newborn. Homozygous protein C deficiency is usually easily diagnosed in the neonate with levels often undetectable at presentation. In contrast, the wide range of protein C levels seen in the normal neonate can make heterozygous protein C deficiency difficult to diagnose and assays may have to be repeated after 6 months to confirm a deficiency. Assays of other vitamin K-dependent coagulation proteins for comparison can be of help in the diagnosis, as can the measurement of parental levels of protein C
- Protein S. Total protein S levels in the neonate are low when compared with adult ranges but the protein is present almost totally in its free form due to the low levels of C4 b binding protein (Schwarz *et al*, 1988). Free protein S levels, however, are low when compared with adult values and increase to the adult range by 4 months of age, total protein S increasing similarly in the first 10 months of life. As with protein C, homozygous protein S deficiency is usually associated with low or undetectable levels of protein S in this age group and again, the heterozygous state can be difficult to diagnose definitively until a later age
- Anti-thrombin (AT). Functional AT levels are reduced in the term neonate and more so in the premature infant: they will remain reduced for at least the first 3 months of life
- Resistance to activated Protein C. The wide variations in neonatal factor VIII levels make this an inappropriate test in this age group and the Factor V Leiden genotype should be looked for directly

- Factor V Leiden
- Prothrombin^{20210A}.

Each of these single point mutations is associated with an increased risk of venous thrombosis in adults and children and is identified using polymerase chain reaction (PCR) techniques (Bertina *et al*, 1994; Poort *et al*, 1996). Approximately 4% and 2%, respectively, of Caucasians are heterozygous for these gene defects. Their causative role in neonatal thrombosis is unknown but they may have a contributory role in the pathogenesis of thrombosis in this particular age group.

Abnormalities of the fibrinolytic system. Plasminogen levels at birth are approximately 50% of adult values but homozygous plasminogen deficiency does not appear to be associated with thrombosis (Mingers *et al*, 1998) and it would therefore not seem justified to include this investigation in thrombophilia testing.

Other investigations of thrombophilia. Babies of women with systemic lupus erythematosus and/or antiphospholipid syndromes may infrequently develop thrombosis in the presence of associated autoantibodies (lupus anticoagulant, anticardiolipin antibodies). Testing thrombotic neonates for these disorders in the absence of a maternal history is not justified because of the rarity of such findings.

Associations of neonatal thrombosis and other thrombophilic defects such as the methylenetetrahydrofolate reductase (MTHFR) T677T genotype and increased serum levels of lipoprotein a have recently been reported (Heller *et al*, 2000).

3. THE MANAGEMENT OF SPECIFIC ACQUIRED DEFECTS OF HAEMOSTASIS

Vitamin K deficiency and disseminated intravascular coagulation (DIC) remain the major acquired haemostatic problems encountered in the neonate. The occurrence of intracranial and intraventricular haemorrhage in this age group has resulted in various prophylactic interventions being proposed but the lack of large controlled trials has made treatment recommendations difficult and a number of areas of controversy need to be resolved.

3A. Vitamin K deficiency

Levels of the vitamin K-dependent coagulation proteins (factor II, VII, IX, X) and the naturally occurring inhibitors protein C and protein S are physiologically low at birth and these proteins are functionally inactive in the absence of vitamin K.

Haemorrhagic disease of the newborn or, in current terminology, vitamin K deficiency bleeding (VKDB) can be classified as early, classical and late depending on the timing of presentation and the associated features (Sutor *et al*, 1999).

Diagnosis. Isolated prolongation of the PT is the earliest laboratory evidence of vitamin K deficiency followed by prolongation of the APTT. The diagnosis is confirmed by correction of these parameters by vitamin K1 or by assay of the specific factors and comparing with age-adjusted normal ranges. Other confirmatory tests include measurement of decarboxyprothrombin (PIVKA II), the Echis Prothrombin time ratio and measurement of vitamin K concentrations, but these assays are rarely available for routine laboratory use (Solano *et al*, 1990).

The use of vitamin K prophylaxis. i) In an attempt to reduce early VKDB, guidelines have been published on the management of pregnant women with epilepsy (Delgado-Escueta & Janz, 1992). In addition to giving advice about the choice of anti-convulsant, intramuscular vitamin K (1 mg) is recommended for all neonates together with antenatal administration of oral vitamin K (20 mg/d) during the last 4 weeks of pregnancy. The latter recommendation is based on the finding of absent PIVKA II in the cord blood of women who received antenatal oral prophylaxis (Cornelissen *et al*, 1993; *Grade B recommendation based on level IIa evidence*).

ii) Vitamin K requirements in a neonate are estimated to be around 1 μg/kg/d. Classical VKDB can be prevented by the postnatal administration of a single dose of vitamin K (1 mg) given orally or by intramuscular (i.m.) injection (Cornelissen *et al*, 1997). One i.m. dose of vitamin K, with rare exceptions, will also prevent late VKDB but the same is not true following a single oral dose, particularly in high-risk infants (McNinch & Tripp, 1991). Routine prophylaxis using parenteral vitamin K remains controversial following the report of an association between i.m. vitamin K and childhood cancer (Golding *et al*, 1992); a number of subsequent studies have failed to confirm this association and the current evidence has been reviewed recently (Zipursky, 1999).

Despite the absence of clear supporting data, the controversy surrounding i.m. vitamin K and the risk of childhood cancer seems unlikely to be resolved in the short term. This has resulted in the continued use of parenteral vitamin K and to the development of alternative oral regimens, leading to a wide variation in prophylaxis policy both in the UK and worldwide.

Currently, it is recommended that all neonates receive postnatal vitamin K prophylaxis for the prevention of VKDB (*Grade B recommendation based on level III evidence*, McNinch & Tripp, 1991). It is not possible at this time to make a firm recommendation on the optimal route or regimen to be used: however, in well babies vitamin K 1 mg by i.m. injection or oral vitamin K 1 mg at birth with an additional 25 μg/d thereafter for 3 months in infants who continue to be breast fed appear to be associated with the lowest risk of VKDB (*Grade B recommendation based on level III evidence*, Cornelissen *et al*, 1997). Alternative oral regimes have also been shown to be effective, such as vitamin K 2 mg at birth followed by 1 mg weekly for 3 months in breast-fed babies (*Grade B recommendation based on level III evidence,* Hansen & Ebbesen, 1996).

Management of vitamin K deficiency bleeding. Any infant suspected of VKDB should receive immediate intravenous vitamin K replacement: it is standard practice to administer a dose of 1 mg which will usually result in correction within a few hours (*Grade C recommendation based on level IV evidence*). Intravenous vitamin K can be associated with anaphylactoid reactions and should be administered by slow

intravenous injection; if venous access cannot be established it can be given subcutaneously, the intramuscular route being avoided in the presence of a coagulopathy (*Grade C recommendations based on level IV evidence*, Sutor *et al*, 1999).

In infants who are bleeding, fresh-frozen plasma (FFP) 10–15 ml/kg should be administered in addition to vitamin K (*Grade C recommendation based on level IV evidence*). This will raise the vitamin K clotting factors by 10–20 iu/dl: care should be taken to avoid increases in blood pressure secondary to rapid volume expansion. The use of Prothrombin Complex Concentrate (PCC) should be considered in the presence of life-threatening haemorrhage or intracranial haemorrhage when it is necessary to normalize the levels of the depleted coagulation factors. Whereas extrapolation of adult studies would suggest a dose of 50 u/kg, it should be noted that there is no direct data available for the use of these concentrates in the neonate.

3B. Disseminated intravascular coagulation (DIC)

DIC always occurs as a secondary event to another disease entity (Table VIII). The incidence is particularly high during the neonatal period, especially in preterm infants. The clinical spectrum of neonatal DIC varies greatly from apparently asymptomatic cases of low-grade compensated DIC to fulminant DIC characterized by bleeding and thrombosis. Intrapulmonary and intraventricular haemorrhage may be exacerbated in preterm infants by thrombocytopenia and an uncompensated coagulopathy.

Laboratory diagnosis of DIC. DIC is characterized by procoagulant activation, fibrinolytic activation and the consumption of anticoagulants together with biochemical evidence of end organ damage (Bick, 1995). Laboratory abnormalities may therefore include a prolongation of the routine coagulation screening tests [PT, APTT, thrombin clotting time (TCT)], a reduced plasma fibrinogen level, thrombocytopenia and increased D-dimers or fibrin/fibrinogen degradation products. Although not routinely performed for diagnostic purposes, reduced levels of procoagulant proteins and anticoagulants are also present, reflecting ongoing consumption. It should, however, be emphasized that the laboratory assessment of DIC can be difficult in the neonate and young infant because of the physiological coagulation changes which occur during this period. The use of selected factor assays (FV and FVIII) can be useful in differentiating DIC from other acquired coagulopathies in this age group.

Table VIII. Disorders associated with neonatal disseminated intravascular coagulation.

(a) Fetal/neonatal disorders	(b) Maternal/obstetric disorders
Hypoxia – acidosis; birth asphyxia, RDS	Dead twin
Infection – bacterial, viral, fungal, protozoal, parasitic	Placental abruption
Necrotizing enterocolitis	Severe pre-eclampsia
Meconium aspiration	
Aspiration of amniotic fluid	
Brain injury	
Hypothermia	
Haemolysis	
Giant haemangioma (Kasabach–Merrit syndrome)	
Homozygous ProteinC/S deficiency	
Malignancy	

Management of DIC. The most important aspect of management is reversal of the underlying disease process. Acidosis should be corrected, tissue perfusion maintained and the neonate well oxygenated. Beyond this there are no clear guidelines on the optimal management of neonatal DIC and a virtual absence of recent randomized controlled trials addressing the available treatment options.

All recommendations on the management of neonatal DIC are grade C based on level IV evidence.

Blood product replacement is indicated for the treatment of clinical bleeding in the presence of laboratory confirmation of DIC. FFP (10–15 ml/kg) provides procoagulant proteins and the naturally occurring inhibitors, AT, Protein C (PC) and Protein S. Cryoprecipitate 10 ml/kg contains a higher concentration of FVIII and fibrinogen per unit volume than FFP and is particularly useful in the presence of hypofibrinogenaemia. Platelet concentrates (10–15 ml/kg) may be necessary to maintain a platelet count of $> 50 \times 10^9$/l. Paediatric platelet packs contain 60 ml of concentrate and are therefore ideal for most neonatal transfusions: if such packs are not available an adult pack should be used. Volume reduction of adult packs may be clinically indicated but this process can result in platelet activation and aggregation, thereby reducing the *in vivo* efficacy of the platelets. Red cell concentrates may be required and exchange transfusion may be necessary to avoid volume overload.

Thrombosis can be as problematic as bleeding in DIC and heparin should be given in obvious thrombotic DIC. The administration of naturally occurring coagulation inhibitors such as AT and PC has been shown to be of benefit in specific cases of adult DIC associated with multi organ failure (Smith *et al*, 1997; Eisele *et al*, 1998), although it is not known if this is applicable to similarly affected neonates: in the absence of clinical trials heparin, AT or PC either alone or in combination cannot currently be recommended for the routine treatment of neonatal DIC.

3C. Intraventricular (IVH)/intracranial (ICH) haemorrhage

Periventricular-intraventricular haemorrhage is the most common form of ICH in preterm infants of low birth weight with an incidence of around 15–20% for infants less than 32 weeks gestation (Oh *et al*, 1996). The aetiology is multifactorial with alterations in cerebral blood flow, fragility of the immature germinal matrix vessels and endothelial ischaemia being more significant than impaired haemostasis.

Various therapeutic modalities have been used in an attempt to reduce the incidence of IVH, including measures to improve haemostasis. There has been no consistent

benefit achieved by the prophylactic use of FFP or platelets in high-risk infants (Northern Neonatal Nursing Initiative (NNNI) Trial Group, 1996a, b; Andrew *et al*, 1993) and these blood products should not therefore be routinely administered to such children (*Grade A recommendations based on level Ib evidence*). It is, however, common practice to transfuse platelets prophylactically when the platelet count falls below 30×10^9/l in an otherwise well infant or below 50×10^9/l in the sick preterm neonate.

Information is also limited on the optimal management of a neonates with a pre-existing IVH: it would, however, seem appropriate to correct a coagulation abnormality and maintain a platelet count of $> 50 \times 10^9$/l in order to prevent extension of the bleed (*Grade C recommendation based on level IV evidence*). An inherited coagulation disorder should always be excluded in any neonate with an apparently spontaneous ICH (*Grade C recommendation based on level IV evidence*).

4. THE MANAGEMENT OF INHERITED COAGULATION DEFICIENCIES

The vast majority of bleeding problems seen during the neonatal period are due to acquired haemostatic disorders. However, inherited coagulation disorders can present in the neonatal period and there may be no preceding family history to suggest the diagnosis.

Perinatal management. In the presence of a positive family history of an inherited coagulation deficiency, pregnancy and delivery should be managed in such a way as to reduce the potential risk of bleeding in both the mother and baby to a minimum. This should entail the close liaison of the Obstetric and Neonatal Units and the local Haemophilia Centre so that a management plan exists for the delivery and for the subsequent investigation and treatment of the neonate. The management of such women has been the subject of recent guidelines (Haemostasis and Thrombosis Task Force, 1994).

Rarely, factor replacement therapy may be required urgently following delivery and access to appropriate treatment should be arranged prior to delivery. At birth a cord blood sample should be obtained for the relevant coagulation factor investigations.

Historically the risk of intracranial haemorrhage in neonatal haemophilia A or B has appeared to be low. However, a recent literature review has described a cumulative incidence for intracranial and extracranial haemorrhage of 3·58% (Kulkarni & Lusher, 1999). Prophylactic administration of factor VIII concentrate to the neonate may reduce this incidence or modify the bleed (Buchanan, 1999), although no evidence currently exists to support this as routine practice. Whereas cranial ultrasound findings from large prospective series are lacking, scanning of the head should at the least be undertaken in the first hours after delivery if there has been any trauma during delivery and in other congenital deficiency states where the risk of ICH is higher, e.g. FXIII, FVII, FX, deficiency [*Grade C recommendation based on level IV evidence* (Girolami *et al*, 1985, Anwar & Miloszewski, 1999).

Clinical features. In the absence of a positive family history, the diagnosis may be suspected by the presence of abnormal bleeding, which usually occurs in the context of an otherwise healthy infant. The haemophilias are the commonest inherited bleeding disorders to present in the neonatal period. The pattern of bleeding observed in neonates is often iatrogenic in origin and can be characterized by continued oozing or excessive haematoma formation following venepuncture, heel stab sampling or the administration of intramuscular vitamin K. Significant haemorrhage can occur following circumcision. Umbilical bleeding is relatively uncommon in haemophilia and is typically associated with severe hypofibrinogenaemia and homozygous factor XIII deficiency. Likewise, ICH occurs infrequently in haemophiliacs but is a significant cause of morbidity and mortality in the severe forms of factor VII, factor X and factor XIII deficiency (Girolami *et al*, 1985).

Diagnosis. Factor VIII levels are within the normal adult range in both term and preterm infants and it is therefore possible to confirm a diagnosis of haemophilia A in the neonatal period regardless of gestational age and severity. This also applies to deficiencies of fibrinogen and factor V. The diagnosis of severe (< 2 iu/dl) and moderate (2–5 iu/dl) haemophilia B can also be confirmed in the neonatal period. However, confirmation of mildly (> 5 iu/dl) affected cases is problematic due to overlap with the normal range necessitating repeat testing at around 6 months of age.

von Willebrand disease (VWD) is caused by quantitative or qualitative defects of von Willebrand factor (VWF) (Sadler, 1994). This factor is an acute phase protein and physiological increases make the diagnosis of type 1 VWD difficult in the neonate. Type 2 VWD can be suspected from discrepancies in the plasma levels of VWF antigen and activity and some subtypes may be confirmed by analysis of VWF multimers, although the relative lack of VWF-cleaving protease in the neonate can make such analysis difficult. Type 3 von Willebrand disease can be diagnosed in neonates who have essentially a total deficiency of von Willebrand factor. Knowledge of the particular molecular defect occurring in the family will also be of value where the diagnosis is in doubt.

Homozygous deficiencies of factors II, VII, X and XI can be diagnosed in the neonatal period, whereas levels in heterozygotes may overlap with the normal range precluding confident identification at this stage. Exclusion of Factor XIII deficiency should be carried out in neonates having characteristic bleeding patterns accompanied by normal coagulation screening tests.

Management. Where there is clinically significant ongoing haemorrhage and a congenital factor deficiency is suspected but not confirmed, fresh-frozen plasma (10–15 ml/kg) may be administered while the results of laboratory investigations are awaited (*Grade C recommendation based on level IV evidence*). Guidelines for the treatment of hereditary coagulation disorders have recently been published by the UK Haemophilia Centres Doctors' Organization (UKHCDO, 1997) and management of these disorders should always be undertaken in conjunction with the local

Haemophilia Centre or Comprehensive Care Centre. Recombinant factor VIII and recombinant factor IX concentrates carry the lowest risk of transmitting viral infection and should therefore be given to neonates with haemophilia A or B who require factor replacement [*Grade B recommendation based on level III evidence* (UKHCDO, 1997)]. If recombinant products are not available, a high purity, viricidally inactivated plasma-derived concentrate should be used [*Grade B recommendation based on level III evidence* (UKHCDO, 1997)]. There is little information available on the pharmacokinetics of replacement therapy in neonates and dosing is therefore based on schedules used in older children and adults (Rickard, 1995).

Due to the risks of hyponatraemia and water intoxication, desmopressin (DDAVP) should not be used in the treatment of neonatal VWD. A viricidally treated intermediate purity factor VIII concentrate containing the high-molecular-weight multimers of von Willebrand factor remains the treatment of choice [*Grade C recommendation based on level IV evidence* (UKHCDO, 1997)].

The treatment of bleeding secondary to other inherited deficiency disorders should be with specific high purity factor concentrates where these products exist (fibrinogen, factor VII, factor XI, factor XIII) or, alternatively, with PCC for deficiencies of factor II or factor X and fresh-frozen plasma for factor V deficiency (*Grade C recommendation based on level IV evidence*). Although specific concentrates are usually plasma derived, recombinant factor VIIa may provide an alternative to plasma-derived factor VII for the treatment of inherited factor VII deficiency (Billio *et al*, 1997). Factor XI concentrate and PCCs should be used with care in the neonatal period because of the potential risk of thrombosis: there are few data available on the use of these products in neonates and FFP may provide a safer alternative. Neonates found to have homozygous factor XIII deficiency are at significant risk of ICH and are likely to benefit from routine prophylaxis that maintains plasma factor XIII levels above 3 iu/dl. Current regimens utilize a dose of 30 iu/kg administered once monthly. If factor XIII concentrate is not available, FFP 5–10 ml/kg can be used.

Prophylaxis during the neonatal period should also be considered in severe homozygous FVII and FX deficiency although even with appropriate replacement therapy significant haemorrhage may still occur (*Grade C recommendation based on level IV evidence*).

All infants who may require treatment with factor concentrates should be vaccinated against hepatitis B (*Grade C recommendation based on level IV evidence*). This should be administered subcutaneously with pressure applied over the injection site for 5 min. Hepatitis A vaccination should not be administered until the child is at least 1 year old.

5. NEONATAL THROMBOSIS

Neonates and infants less than 1 year of age account for the largest proportion of thrombotic events seen in the paediatric population (Andrew *et al*, 1994a). These events, however, remain relatively uncommon and often occur in sick term and preterm infants: the most important risk factor for the development of thrombosis during the neonatal period is the presence of an indwelling central line and consequently the vessels involved tend to be those most frequently used for catheterization. Other documented risk factors for the development of neonatal thrombosis include asphyxia, septicaemia, dehydration, maternal diabetes and cardiac disease.

Spontaneous, non-catheter-related thrombotic events are uncommon in the neonatal period and most frequently involve the renal vein. The majority of cases of renal vein thrombosis present during the first few days of life and, in approximately a quarter of these, the thrombosis is bilateral with a smaller proportion also developing extension into the inferior vena cava.

Predisposing factors. Acquired deficiencies of protein C and protein S, particularly in sick, preterm infants, may increase the risk of thrombosis (Manco-Johnson *et al*, 1991). At present, the impact of inherited prothrombotic defects on both catheter-related and spontaneous thrombotic events in the neonatal period remains poorly defined, although it seems likely that these will become increasingly recognized as contributory factors in this setting (Heller *et al*, 2000).

Neonatal thrombotic events have occasionally been reported in association with maternal systemic lupus erythematosus due to the transplacental passage of antiphospholipid antibodies (Tabbutt *et al*, 1994).

Diagnosis of neonatal thrombosis. Thrombosis must be confirmed objectively before undertaking thrombolytic treatment or anticoagulation given the significant risks of such treatment in this age group.

Doppler ultrasound methods are the most frequently used scanning techniques and provide readily available, non-invasive imaging giving valuable diagnostic information. Reliance on Doppler can fail to diagnose thrombosis in particular sites such as the aorta, right atrium and inferior vena cava in neonates with umbilical artery or venous catheters when compared with the use of contrast angiography (Vailas *et al*, 1986; Roy *et al*, 1997). Similar problems are likely to exist with imaging of the upper limb venous system and the most recent recommendation of the Scientific and Standardization Subcommittee on Neonatal Haemostasis is that contrast angiography remains the 'gold standard' imaging technique for the confirmation of thrombotic vessel occlusion, particularly when thrombolytic therapy or surgery is planned [*Grade B recommendation based on level III evidence* (Schmidt & Andrew, 1992)]. Linograms, when dye is injected via a central venous line, are not a substitute for venography and may fail to demonstrate extensive thrombosis.

Computerized tomography (CT) and magnetic resonance imaging (MRI) scanning are indicated for thrombosis-related problems of the central nervous system (CNS).

Management of neonatal thrombosis. The management of arterial and venous thromboembolic events in the neonatal period remains controversial and it is generally acknowledged that there is an urgent need for large multicentre studies on which to base recommendations. Treatment options include supportive care only, anticoagulant therapy

with heparin or low-molecular-weight heparin (LMWH), thrombolytic therapy and surgery. Warfarin is not indicated in the neonatal period because of the difficulties in establishing consistent levels of anticoagulation.

All recommendations on the management of neonatal thrombosis are Grade C, based on level IV evidence, unless otherwise stated.

Supportive therapy. In the absence of controlled studies indicating the efficacy of more aggressive therapy, supportive care alone may be appropriate management for clinically silent thrombosis, which will therefore include the majority of small, asymptomatic catheter-related events (Schmidt & Andrew, 1992). Regular objective monitoring should be performed to detect extension of the original thrombus and in the case of catheter-related events, catheter removal is recommended.

Anticoagulant therapy. In the presence of more extensive, clinically significant thrombosis, particularly when there is evidence of organ or limb dysfunction, consideration should be given to the use of anticoagulant therapy. Unfractionated heparin remains the most frequently used anticoagulant, although there is increasing experience with LMWH in this age group.

The use of heparin in the neonatal period is complicated by the physiological immaturity of the haemostatic system, with reduced levels of antithrombin resulting in relative heparin resistance (Schmidt *et al*, 1988).

A dosage regimen for unfractionated heparin has been suggested by Andrew *et al* (1994b) and is shown in Table IX [*Grade B recommendation based on level IIb evidence (*Andrew *et al*, 1994b)].

In the absence of a validated therapeutic range for the use of heparin in neonates, the APTT should be prolonged to a therapeutic range corresponding to an anti-Xa level of 0·35–0·7 units/ml, although the limitations of the anti-Xa level should be noted (it provides a guide to the pharmokinetics of heparin and only limited information on the anticoagulant and prohaemorrhagic effects *in vivo* of this drug. Also, standard anti Xa assays will tend to underestimate heparin unless the neonatal antithrombin deficiency is fully corrected in the test system).

Nomograms are available for dose adjustment (Andrew *et al*, 1994b), and can be modified by individual laboratories in order to achieve their own therapeutic APTT range.

Thrombocytopenia is common in the sick neonate and every attempt should be made to maintain a platelet count above 50×10^9/l during heparin therapy.

Table IX. Heparinization of neonates and dose adjustment.

Loading dose of heparin	: 75 iu/kg, then
Continuous infusion	: 28 iu/kg/h
Monitor using an APTT or heparin assay, taking first blood sample 4 h after loading dose. Use nomogram for dose adjustment.	

The optimal duration of anticoagulation remains undefined but short-term therapy (e.g. 10–14 d) is commonly used, with objective radiological monitoring performed both during and after completion of anticoagulant therapy. If there is evidence of a new or extending thrombus following completion of initial therapy, heparin should be recommenced.

Low molecular weight heparin is becoming increasingly used in the treatment of neonatal thrombosis. Dose-finding studies have been published and indicate that, as with standard heparin, dose requirements in the neonate are higher than in older children (Massicotte *et al*, 1996; Nohe *et al*, 1999). For example, the recommended dose of enoxaparin is 1·5 mg/kg/dose administered subcutaneously twice per day which should result in a therapeutic anti-Xa level of between 0·5 and 1·0 units/ml by chromogenic assay at 4 h post dose [*Grade B recommendation based on level IIb evidence (*Massicotte *et al*, 1996)].

Thrombolytic therapy. Thrombolytic therapy should be considered in the presence of extensive thrombosis with organ dysfunction or when limb viability is threatened. Such therapy should not be used within 10 d of surgery or in the presence of pre-existing bleeding problems.

Streptokinase, urokinase and tissue plasminogen activator (t-PA) have all been used in neonates but overall experience is relatively limited and results conflicting (Chalmers & Gibson, 1999). As with heparin, the response to thrombolytic agents in the neonate is significantly different from that seen in older children, reflecting physiologically reduced levels of plasminogen. *In vitro* studies have demonstrated that t-PA is more effective than streptokinase and similar to urokinase at lysing thrombi in plasmas with decreased concentrations of plasminogen. t-PA and urokinase are therefore the preferred agents for thrombolytic therapy in neonates. Recommended dosing regimens are shown in Table X [*Grade B recommendation based on evidence level IIB* (Leaker *et al*, 1996)].

There is no laboratory therapeutic range for thrombolytic agents and successful lysis is confirmed on clinical and radiological changes: monitoring should therefore be performed during thrombolytic therapy to assess the response to treatment and the duration of therapy will vary depending on this response. Failure to induce lysis of the thrombus may indicate the need for plasminogen supplementation with FFP.

In order to reduce the risk of bleeding in the neonate receiving fibrinolytics, it is recommended that fibrinogen is maintained at > 1 g/l using cryoprecipitate and the platelet count at $> 50 \times 10^9$/l by transfusion *(Grade C, level IV evidence).*

Table X. Thrombolytic regimes in the neonate.

Drug	Bolus	Maintenance	Duration
Urokinase	4400 u/kg	4400 u/kg/h	6–12 h
t-PA	None	0·1–0·6 mg/kg/h	6 h

The use of heparin either during or after completion of thrombolytic therapy is controversial: if given, a dose of 28 iu/kg/h is appropriate in the neonate.

Prophylactic anticoagulation. Heparin prophylaxis is recommended for neonates with indwelling umbilical artery catheters and those undergoing cardiac catheterization. Umbilical artery patency can be prolonged by the use of low-dose heparin (3–5 u/h) by continuous infusion [*Grade B recommendation based on level IIb evidence* (Sutor *et al*, 1997)]. Whether the incidence of clinically significant thrombi is also reduced is not clear. Thrombotic complications associated with cardiac catheterization can be reduced by the administration of a bolus of heparin (100–150 units/kg) as the femoral artery is catheterized (Freed *et al*, 1974). A second bolus may be required for prolonged procedures.

6. CONGENITAL HOMOZYGOUS PROTHROMBOTIC DISORDERS

Protein C and Protein S deficiencies

Homozygous (or compound heterozygous) deficiencies of protein C and protein S (plasma activities < 0·01 U/ml) are rare conditions and usually present as life-threatening disorders in the neonatal period (Marlar & Neumann, 1990). A less severe form of protein C deficiency in which the level of protein C, although reduced, remains detectable (0·02–0·23 U/ml), may present in the neonatal period, although does so more commonly in later life (Sharon *et al*, 1986).

Clinical features. Onset is usually within the first few days of life and can occur within hours of birth. The microcirculation is characteristically affected first with the development of purpura fulminans associated with laboratory evidence of DIC. Cerebral and renal vein thrombosis are common and can be presenting features. Ocular manifestations are also characteristic and it is likely that cerebral and ophthalmic thrombosis often occur as intrauterine events. Although purpura fulminans is almost always a feature, major vessel thrombosis occasionally occurs in isolation.

Diagnosis. In the acute untreated phase, baseline laboratory results are frequently indicative of DIC. The definitive diagnosis can be difficult in the neonate. Protein C and protein S levels are physiologically reduced at birth and are further reduced in the presence of DIC, during which protein C in particular can reach very low levels. The diagnosis is therefore based on finding undetectable protein C (or protein S) activity (< 0·01 U/ml) with heterozygous levels in the parents. When the molecular defect is known, prenatal diagnosis can be offered to families when there is a prior history of neonatal purpura fulminans.

Management. The most important aspect of management in the acute phase is the immediate and adequate replacement of the deficient inhibitor. Protein C deficient neonates should receive Protein C concentrate at a starting dose of 40 IU/kg, subsequent dosage being based on protein C recovery data. If concentrate is not immediately available, FFP 10–20 ml/kg should be used as a temporary measure. During the early stages of replacement therapy when DIC is ongoing, the half-life of the infused protein C may be as short as 2–3 h, necessitating frequent dosing. This usually improves to about 10 h once the DIC is controlled, enabling a single daily treatment to be given (Dreyfus *et al*, 1995). Protein C levels should remain in excess of 0·25 U/ml to prevent further thrombosis (Muller *et al*, 1996). Replacement therapy is required for an initial period of at least 6–8 weeks to facilitate resolution of clinical lesions.

There is no currently available protein S concentrate and FFP is therefore used for replacement therapy (10–20 ml/kg every 8–12 h) (*Grade C recommendation based on level IV evidence*).

Long-term management remains controversial but current regimens utilize oral anticoagulants with or without concomitant replacement therapy. It is difficult to warfarinize infants until their vitamin K-dependent factors have physiologically increased and affected babies should preferably receive replacement therapy. Older children receiving warfarin seem to require an International Normalized Ratio (INR) at the upper end of the therapeutic range (3·0–4·5) to prevent recurrent skin necrosis (*Grade C recommendation based on level IV evidence*) and therefore require careful monitoring. Where possible, dosing should be individualized to identify the minimum dose required to remain symptom free. In the event of recurrent problems during warfarin therapy, temporary or longer term reintroduction of protein C replacement may be required.

Similar problems have been reported in the management of homozygous protein S deficiency where, again, intermittent replacement with FFP may be required in addition to oral anticoagulants (Mahasandana *et al*, 1996).

Low-molecular-weight heparin may be a useful therapeutic option in the long term treatment of homozygous protein C deficiency in which plasma levels remain detectable but its place in the management of cases with undetectable levels has not been determined (Monagle *et al*, 1998).

Other homozygous defects

Homozygous AT deficiency has been recorded very infrequently. Type I defects are probably incompatible with life and the majority of reports relate to type II HBS defects (Chouwdury *et al*, 1994). Long-term anticoagulation has been successfully used in this disorder.

Homozygosity for the Factor V Leiden mutation rarely presents in childhood and a significant number of adults with this mutation also remain symptom free throughout their lives, the same being true for individuals homozygous for the PT^{20210A} mutation.

[1]*Birmingham Children's Hospital, Birmingham, and* [2]*Royal Hospital for Sick Children, Yorkhill, Glasgow, UK on behalf of the Haemostasis and Thrombosis Task Force, British Committee for Standards in Haematology*

Michael D. Williams[1]
Elizabeth A. Chalmers[2]
Brenda E. S. Gibson[2]

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RECOMMENDED REVIEW AND EXPIRY DATE OF THIS GUIDELINE

1. Review date: 5 years from publication
2. Expiry date: 6 months after review date.

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Keywords: neonate, developmental haemostasis, thrombosis, bleeding disorders, anticoagulation.

APPENDIX. GRADED RECOMMENDATIONS (US Department of Health & Human Services, 1992)

Grade of recommendation

A. (Evidence levels Ia, Ib)	Requires at least one randomized controlled trial as part of the body of literature of overall good quality and consistency addressing the specific recommendation.
B. (Evidence levels IIa, IIb, III)	Requires availability of well-conducted clinical studies but no randomized clinical trials on the topic of recommendation
C. (Evidence level IV)	Requires evidence from expert committee reports or opinions and/or clinical experience of respected authorities; indicates absence of directly applicable studies of good quality

Levels of evidence

Ia. Meta-analysis of randomized controlled trials.
Ib. At least one randomized controlled trial.
IIa. At least one well-designed controlled study without randomization.
IIb. At least one other type of well-designed quasi-experimental study.
III. Well-designed nonexperimental descriptive studies, such as comparative studies, correlation studies and case–control studies.
IV. Expert committee reports or opinions and/or clinical evidence of respected authorities.