University of Birmingham

Birmingham Women's NHS
NHS Foundation Trust

Fetal Red cell alloimmunization :
Modern management & controversies

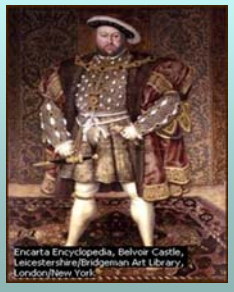
Mark. D Kilby.
Centre for Women's & Children's Health, University of Birmingham &
Fetal Medicine Centre, Birmingham Women's Foundation Trust.

Jornadas sobre Medicina Fetal, *5th March 2015*

HOSPITAL ITALIANO de Buenos Aires

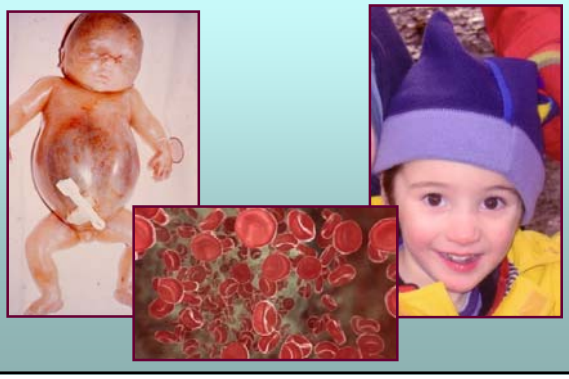
Rhesus Disease

Henry VIII married Katherine of Aragon (1509-1533).



One child (Mary) and many 'hydropic' stillbirths.

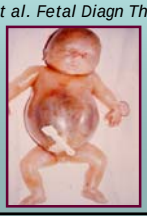
Red cell alloimunisation

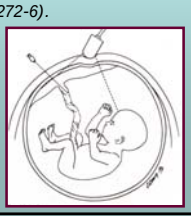


Fetal therapy : Outcome

% Live births:	Overall	Non-hydrops	Hydrops
BWH, 1999-2003	92%	97%	89%
Literature (1998)	84%	94%	74%

(Somerset et al. Fetal Diagn Ther. 2006;21(3):272-6).





Rhesus disease

- Rhesus alloimmunisation between 10.6 - 53.5 cases per 10,000 total births *(Hughes et al,1994)*
- 400 - 500 new cases per year (mainly D).

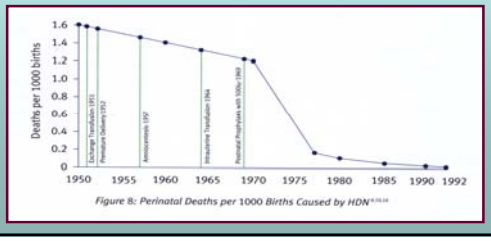
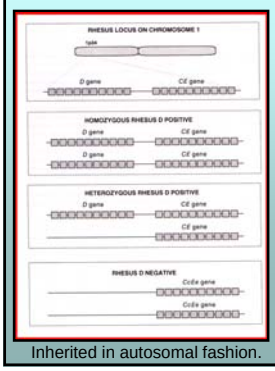


Figure 8: Perinatal Deaths per 1000 Births Caused by HDN

Rhesus disease

- Introduction of erythrocytes (fetal) with 'foreign' antigens to the mother entering the maternal circulation.
- Common RBC antigens that cause in-utero anaemia are:
  - Rhesus family (Anti-D, C, E, c and e)
  - Anti-Kell.
  - Anti-U
- In UK, 15% D negative (Black 8%, Chinese <1%).

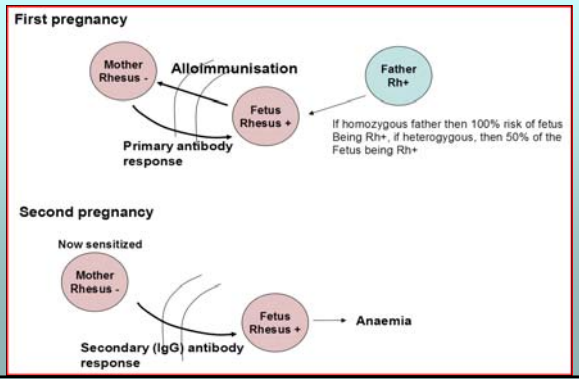
Genetics of common Rhesus Disease



Inherited in autosomal fashion.

- 1932 - Diamond indicated that hydrops, icterus Gravis and kernicterus same aetiology.
- 1938 - Darrow noted that RBC destruction aetiology of disease.
- 1940 - Lansteiner & Weiner characterise 'Rhesus blood group' system.
- 1941 - Levine made link between antibodies and HDN.
- 1991 - Colin cloned human D locus
- 1994 - Exons 4,5 and 6 of D locus appear critical for reactivity.

Pathogenesis of 'Rhesus disease'

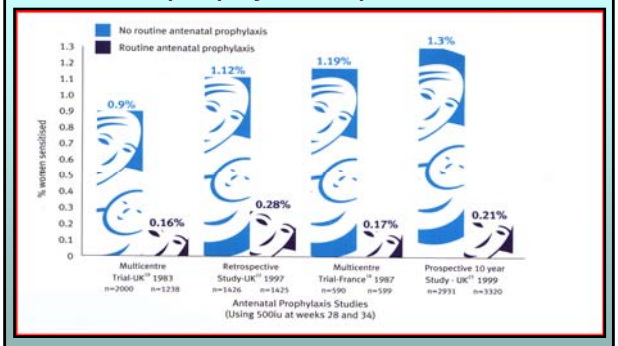


Potentially sensitising obstetric events

'Targeted' Prophylaxis :
- at least 250 iu anti-D before 20 weeks.
- at least 500 iu anti-D after 20 weeks.

- Delivery of Rh(D) positive fetus
- Prenatal diagnosis (i.e. amniocentesis, CVS)
- Revealed or concealed APH
- External version of fetus
- Closed abdominal trauma
- Intrauterine death
- [Threatened] Miscarriage (at or after 12 wks.)
- Instrumental delivery
- Administrative failure.

"Routine' prophylactic prenatal Anti-D

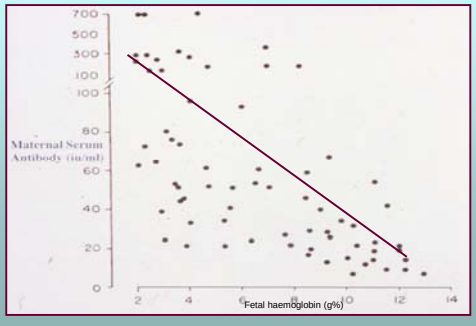


In most centres in the UK, RAADP is given as a single dose of 1500 IU at 28 weeks of gestation *(NICE, 2008)*.

Prenatal detection of alloimunisation

- Rhesus negative women screened in the WM Region at
- booking, 20 and 28 weeks.
- Indirect immunoglobulin (Coombs) test and titre.
- Serial quantification using auto-analyser.
- Paternal genotype
- If anti-D titre < 4iu/ml. Repeat 2 wkly.
- If anti-D titre is between 4-10 iu/ml : risk of anaemia
- relatively low but central referral. Ultrasound +/- other tests.
- Recent data indicates that anti-D threshold of >6iu is
- 'predictive' of prenatal intervention ()
- If anti-D titre > 10 iu/ml. Careful follow up.
- Role of cell-mediated cyto-toxicity test (ADCC) may have
- some role *(Oepkes et al,2001)*

Maternal anti-D titres, potency & fetal anaemia



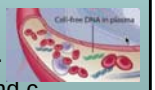
(Bowell PJ et al, Br J Obstet Gynaecol. 1988;95(8):759-64)

Paternal genotyping.

- For anti-D, if father Rh(D) positive there is a 56% chance he is heterozygous.
- 8% males are Kell positive (97% heterozygous)
- For c, 80% males will be positive and 60% will be heterozygous.
- If father is heterozygous then fetal genotyping by NIPD.

Fetal genotyping

- During pregnancy, cell free fetal DNA is present in maternal blood.
- The No: copies of fetal DNA increase with advancing gestation.
- Real-time PCR distinguish *RHD* from *RHDΨ* (in black Africans). Also Y chromosome associated SRY sequences.



- 100% accuracy in 94 women (first pilot).
- Probes and primers designed for Kell and c.

(Finning et al,2002; Avent,2001)

Prediction of Fetal RhD status

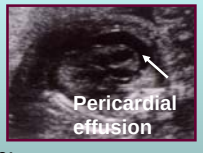
Gestation (weeks)	n	Predicted RhD-positive		Predicted RhD-negative		Accuracy of RHD typing
		Male	Female	Male	Female	
≤ 14	19	8	5	2	4	100%
15 - 28	77	27	24	12	14	100%
29 - 42	28	9	11	6	2	100%
Unknown	13	7	3	2	1	100%
Total	137	51	43	22	21	100%

K.M. Finning, P.G. Martin, P.W. Soothill, N.D. Avent. Prediction of fetal D status from maternal plasma: introduction of a new noninvasive fetal *RHD* genotyping service. Transfusion 2002, 42, 1079-82

Management of alloimunisation

Ultrasound assessment:

- Regular biometry
- Check for effusions and ascites.



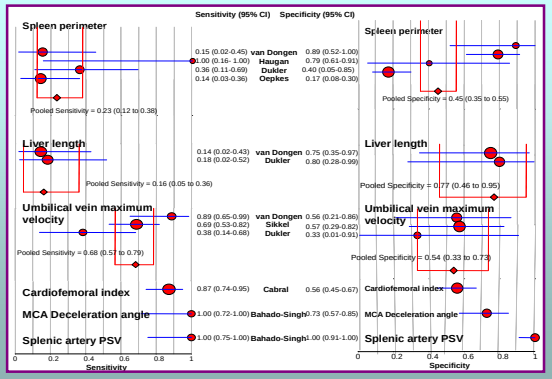
- Frequency dependent upon:
  - Absolute anti-D titre
  - Rate of rise
  - previous history
  - Development of anaemia / IUD

Anti-Kell alloimmunisation

- Anti-Kell may cause serious fetal anaemia and the severity is not influenced by previous obstetric history. 0.1 - 0.2% of all pregnancies.
- Rare. Between 1984-1996 in WM Region there were 18 Kell positive (affected) fetuses born to women with anti-K.
- These were pregnancy-related in 66.6% (12/18) and 33.3% (6/18) transfusion-induced. Same outcome in groups.
- 50% (9/18) had severe disease *(Walker et al,1971)*
- All demonstrated suppressed erythropoiesis.
- Amniocentesis (OD450) not helpful in the management.

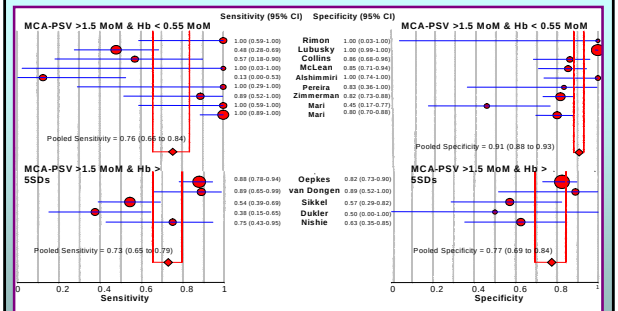
(Grant et al,2000)

Evidence for screening for fetal anaemia



(Pretlove S et al, BJOG. 2009;116(12):1558-67)

Middle cerebral artery Peak systolic velocity

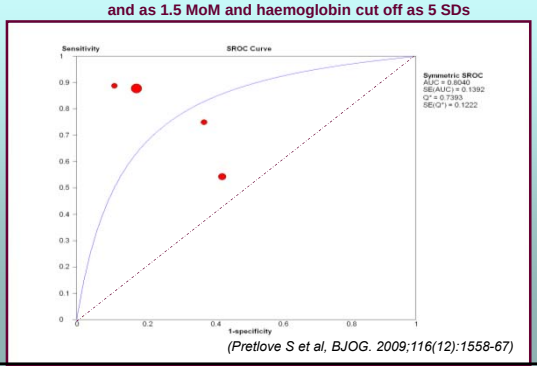


Sensitivities and specificities of middle cerebral artery peak systolic velocity in detection of severe anaemia stratified according to definition of severe anaemia

(Pretlove S et al, BJOG. 2009;116(12):1558-67)

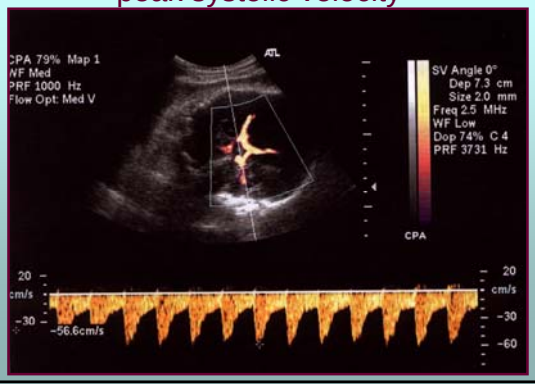
Pooled ROC : MCA PSV

Graphic representation of sensitivity as : SROC using cut off for MCA-PSV and as 1.5 MoM and haemoglobin cut off as 5 SDs



(Pretlove S et al, BJOG. 2009;116(12):1558-67)

Middle cerebral artery Doppler insonation: 'peak systolic velocity'



Middle cerebral artery Doppler insonation: 'peak systolic velocity'

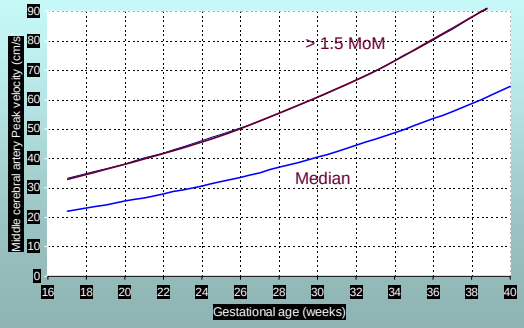


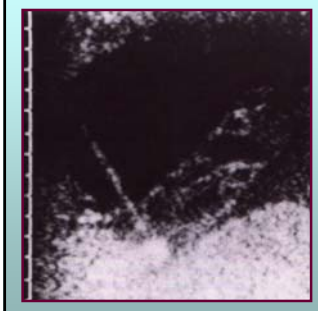
Table 1. Peak Systolic Velocity of Blood Flow in the Middle Cerebral Artery Measured by Doppler Ultrasonography and Amniotic-Fluid ΔOD_{450} Classified According to Liley Zones as Predictors of Anemia.*

Measurement	Severe Fetal Anemia†	No Anemia or Mild Anemia	Total
	number of pregnancies		
MCA blood flow			
>1.5 MoM	65	16	81
≤1.5 MoM	9	75	84
	88%	**17%**	
Total	74	91	165
Amniotic-fluid ΔOD_{450}			
Liley zone 2c or 3	56	21	77
Liley zone 1 or 2	18	70	88
	75%	**23%**	
Total	74	91	165

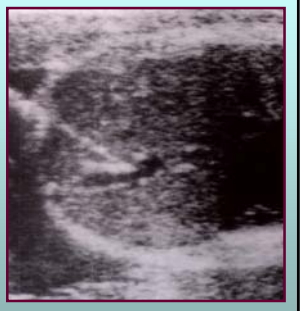
* MCA denotes middle cerebral artery, and MoM multiples of the median. Liley's method is described in Liley.[4]
† Severe fetal anemia is defined as a hemoglobin level at least 5 SD below the mean for gestational age.

Oepkes et al, NEJM. 2006.355:156-164

Fetal blood sampling: 'gold standard'

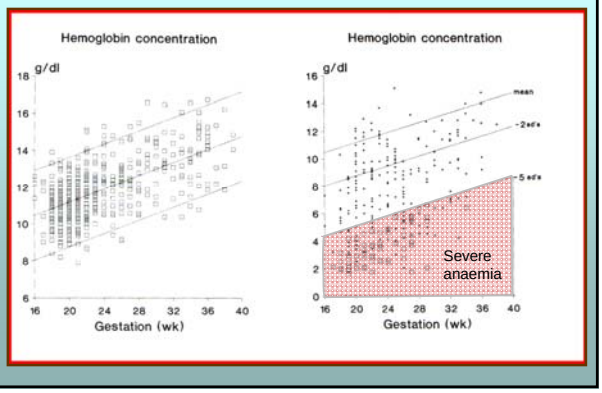


Cordocentesis (25%)

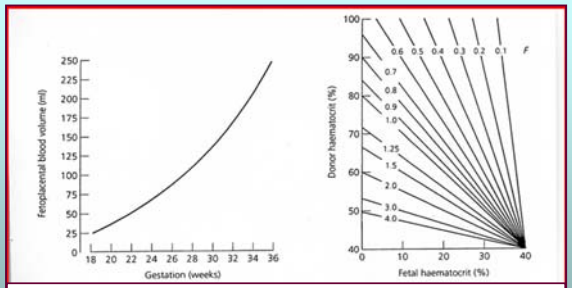


Intrahepatic vein puncture (75%)

Fetal Anaemia

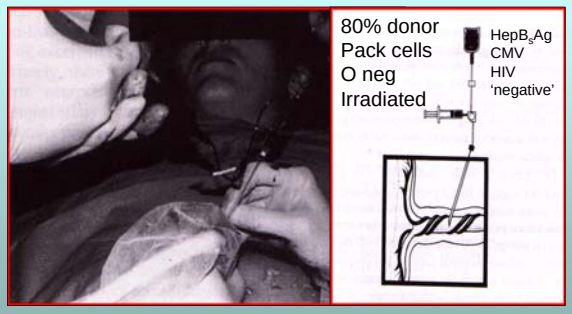


Calculated volume of transfusion



Vol. of transfused blood = $\frac{\text{Donor Hct - Desired Hct}}{\text{Desired Hct - fetal Hct}}$ X fetal blood vol.

Intravascular transfusion (1)

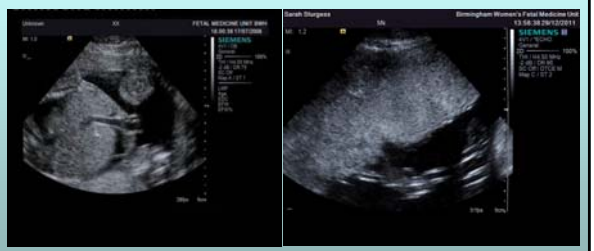


Percutaneous fetal blood sampling and transfusion

Fetal intravascular transfusion

Intrahepatic vein puncture

Placental cord root

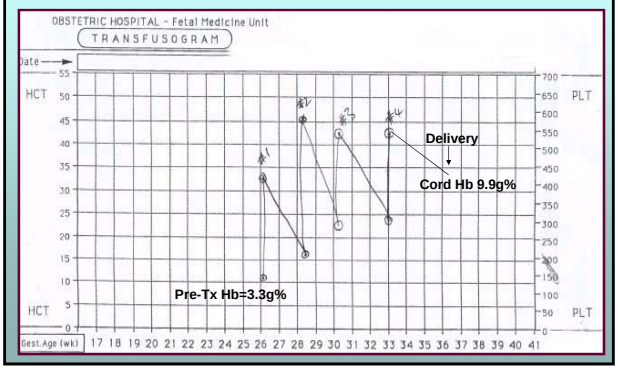


Safe & low risk of vessel leak.

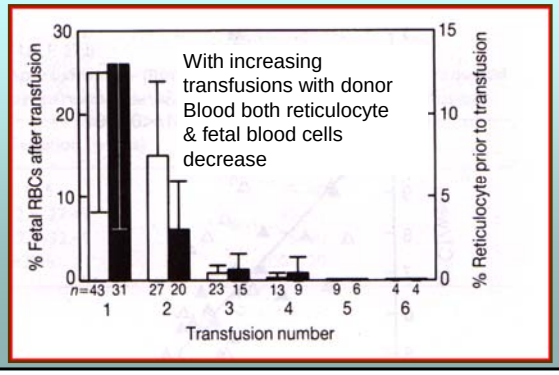
Ease of access/risk of 'tamponade'

Superior & safe results by transfusion via IHV. *(Fetal Diagn Ther. 2006;21(3):272-6)*

Transfusion progress



Suppression of endogenous erythropoiesis



Complications of IUT:

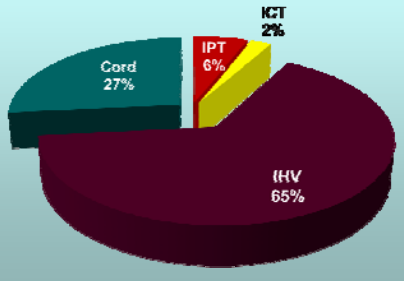
General :

- Risk of miscarriage /preterm delivery.
- Aminorrhexis
- Infection

Specific :

- Cord accidents
- Haemoperitoneal extravasation
- Cardiac ‘overload’
- Death

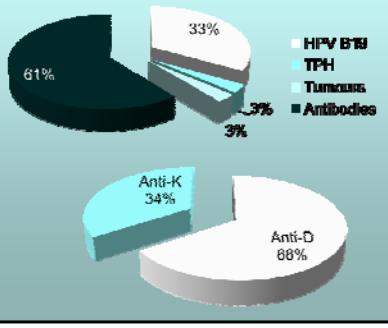
Fetal blood transfusions at Fetal Medicine Centre at Birmingham Women’s Hospital



Between March 2004 – September 2014, 357 IUTs performed on 105 pregnancies

Underlying aetiology of fetal anaemia

357 IUTs performed on 105 pregnancies



Complications : in most common approach

March 2004 – September 2014, 357 IUTs

	IHV	Cord	IPT	ICT
Live neonate	225 (96.6%)	91 (95.7%)	19 (90.4%)	6 (75%)
Miscarriage	6	3	2	2
Stillbirth	1	1	0	0
NND	1	0	0	0
Transient bradycardia	1 (0.4%)	8 (8.4%)	1 (5.2%)	3 (37.5%)

Over a ten year period the miscarriage and perinatal loss was 4.5%. Transient bradycardia in 3.6%/
Overall Survival is 95.5%.

(Somerset DA et al, Fetal Diagn Ther. 2006;21(3):272-6)

Intraperitoneal transfusion

- Initially described by Liley,1963.
- Donor red cells placed into peritoneal cavity.
- Absorbed over 24 - 72 hours via subdiaphragmatic lymphatics / thoracic duct.
- Amount of blood = (GA – 20) X 10mls.

Useful when:

1. History of early-onset anaemia (<20 weeks)
2. If fetus in difficult position.

- Study of 6 pregnancies with severe alloimmunisation, with all fetuses in previous pregnancies having Hb<5g% prior to 20 weeks (66% [4/6]mortality). IPT & IVIG (0.8g/Kg) from 16-22weeks, then IVT. Survival in 83% [6/7].

(Fox CE et al, Fetal Diagn Ther. 2008;23(2):159-63)

Intra-cardiac transfusion (2004 – 2014)

- Eight cases (2.2% of all IUTs).
- All with hydrops fetalis : 62.5% HPV B19.
- All maternal BMI>30 kg/m^2

Table 1. Demographic data relating to the eight subjects undergoing fetal intracardiac transfusion

Patient	Maternal age, years	Parity	Cause of fetal anemia	GA (weeks + days) at first IUT	First Hb, g/dl	Post-IUT Hb, g/dl	Number of IUTs per pregnancy	Outcome	GA (weeks + days) at delivery
1	32	4	Parvovirus	19+2	3.0	11.4	2	IUD at 19+2	–
2	36	2	Parvovirus	17+5	1.8	10.4	1	Live-birth	38+2
3	34	2	Parvovirus	20+0	1.3	11.1	1	Live-birth	38+4
4	31	3	D antibodies (61 IU)	18+1	6.7	12.2	8	Live-birth	33+1
5	30	1	Kell (very high)	18+5	2.3	12.8	8	Live-birth	34+2
6	28	5	Parvovirus	23+1	0.4	10.1	1	IUD at 23+1	–
7	32	1	Parvovirus	20+1	6.1	12.3	1	Live-birth	38+0
8	43	2	D antibodies (125 IU)	17+1	2.2	10.2	7	Live-birth	35+2

GA = Gestational age; Hb = hemoglobin; IUT = intrauterine transfusion.

- Neonatal survival is 75% (6/8). Miscarriages within 48 hr

(Mackie F et al, Fetal Diagn Ther. 2015;27(3):272-6)

Long term morbidity (1)

TABLE 2
Long-term neurodevelopmental outcome in 291 long-term survivors after intrauterine transfusions

Variable	
Age at follow-up, y[a]	8.2 (2-17)
Isolated severe development delay, n (%)	5 (1.7)
Isolated cerebral palsy, n (%)	2 (0.7)
Isolated bilateral deafness, n (%)	3 (1.0)
Cerebral palsy and severe developmental delay, n (%)	4 (1.4)
Neurodevelopmental impairment,[a] n (%)	14 (4.8)

[a] Neurodevelopmental impairment is defined as at least one of the following: cerebral palsy, severe development delay (< −2 SD), bilateral deafness, or blindness.

Lindenburg. Long-term outcome fetal transfusions. Am J Obstet Gynecol 2012.

Long term morbidity (2)

TABLE 4
Analysis of potential risk factors for neurodevelopmental impairment (NDI)

Variable	NDI (n = 14)	No NDI (n = 277)	*P* value univariate analysis	OR (95% CI) univariate analysis	*P* value multivariate analysis[a]	OR (95% CI) multivariate analysis[a]
Hydrops, n (%)	9 (64)	66 (24)	.002	5.8 (1.9–17.8)	.11	3.3 (0.76–14.5)
Hemoglobin at first IUT,[b] g/dL	4.2 ± 1.9	5.6 ± 2.4	.032	1.3 per g/dL decrease (1.0–1.7)	—	—
Z hemoglobin (SDs)	−8.1	−7.3	.13	1.3 per SD decrease (0.6–1.1)	—	—
Number of IUTs[c]	4 (1–5)	3 (1–6)	.018	1.7 per IUT (1.1–2.5)	.02	2.3 per IUT (1.1–4.6)
GA at birth <32 wks, n (%)	2 (14)	4 (1)	.006	12.8 (2.1–79.5)	.54	2.3 (0.17–31.1)
Perinatal asphyxia, n (%)	1 (7)	10 (4)	.51	2.0 (0.2–17.1)	.19	5.8 (0.4–81.3)
Severe neonatal morbidity,[d] n (%)	6 (43)	16 (6)	< .001	13.1 (4.0–42.4)	< .001	85.6 (9.7–755.3)

GA, gestational age; *IUT*, intrauterine transfusion; *OR*, odds ratio; *SD*, standard deviation.

[a] Including parental education as a possible confounder; [b] Value given as mean ±SD; [c] Value given as median (range); [d] Severe neonatal morbidity is defined as at least 1 of the following: respiratory distress syndrome, intraventricular hemorrhage ≥ grade 3, periventricular leukomalacia ≥ grade 2, necrotizing enterocolitis ≥ grade 2, and sepsis.

Lindenburg. Long-term outcome fetal transfusions. Am J Obstet Gynecol 2012.

Conclusion

- Fetal Anaemia is still a common problem requiring treatment in Fetal Therapy Centres.
- Maternal alloimmunisation is still a major cause, although prenatal prophylaxis has reduced the risks of anti-D alloimmunisation.
- Care prenatal surveillence is required to time in-utero therapy.
- Such therapy has good outcomes with at least 95% survival.

Jornadas sobre Medicina Fetal

HOSPITAL ITALIANO de Buenos Aires

SADIPT

Thank you for the invitation to give the lecture and for your attention!