A SIX MONTH OLD GIRL WITH HEMOLYTIC ANEMIA

Case study by Jim Perkins, M.D. (©2009)

CLINICAL PRESENTATION

A six month old girl presented with lethargy after four days of cough, rhinorrhea, and fever, and one day of vomiting. She had been born at term by a normal delivery after prolonged rupture of membranes. Her early infancy was uncomplicated with apparently normal development, but she weighed 5.7 kilograms (5th percentile weight for age = 5.9 kg). She had not been transfused.

On examination the infant was pale and drowsy with sunken eyes, poor skin turgor, and dry mucous membranes. Her temperature was 99.8° F, her pulse 150 (upper limit of normal 160), and her respiratory rate was 40. There was no icterus. Cardiac examination was notable only for a systolic murmur, and the lungs were clear. The spleen was palpable 4 cm below the costal margin; hepatomegaly was not apparent.

The hemoglobin was 1.7 gm/dL and the hematocrit was 5.2%, with 23% reticulocytes, 14 nucleated RBCs per 100 nucleated cells, and a mean RBC volume of 106. The RBC morphology was strikingly abnormal with polychromasia, poikilocytosis, and microspherocytes present, in addition to the nucleated RBCs noted above. There were 11,600 WBC/□L with a differential count of 33% neutrophils, 3% band forms, and 63% lymphocytes. The platelet count was 175,000. Electrolyte and cerebrospinal fluid profiles were normal. Blood and other cultures were obtained.

Pretransfusion testing demonstrated a positive direct antiglobulin test with a warm autoantibody in the eluate. The antibody screen was also positive and the serum contained a warm autoantibody as well as alloanti-M (see below).

A diagnosis of warm autoimmune hemolytic anemia (WAIHA) was made; her physicians also determined to rule out sepsis.

SEROLOGIC TESTING

ABO and Rh Typing

Anti-		Red Cells		Anti-			Interp
A	B	A1*	B	D	Control	CCC	
0	4+	4+	0	4+			B Pos

*A1 reverse typing cell was M positive

Antibody Screen, LISS enhancement

	I.S.	37°, 30'	AHG	CC
OI	4+	4+	4+	
OII	4+	4+	4+	
AC	0	0	3+	

Direct Antiglobulin Test

	Poly	>IgG	>C3
Test	4+	4+	w+
CCC			

Raw serum panel

	Rh					MNSs				P	Lewis		Kell		Duffy		Kidd		Lutheran			LISS				Enzyme		
	D	C	E	c	e	M	N	S	s	P1	Le^a	Le^b	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b		I.S.	37°	AHG	CC	37°	AHG	CC
1. R1R1	+	+	0	0	+	+	+	0	0	+	+	0	0	+	0	0	0	+	0	+	1	4+	4+	3+		4+	4+	
2. R1wR1	+	+	0	0	+	+	0	+	+	+	0	+	+	+	+	+	0	+	0	+	2	4+	4+	3+		4+	4+	
3. R2R2	+	0	+	+	0	0	+	0	+	+	0	+	0	+	0	+	0	+	0	+	3	0	0	3+		4+	4+	
4. Ror	+	0	0	+	+	+	0	0	+	0	0	0	0	+	0	0	+	+	0	+	4	4+	4+	3+		4+	4+	
5. r'r	0	+	0	+	+	+	0	0	+	0	+	0	0	+	+	+	0	+	0	+	5	4+	4+	3+		4+	4+	
6. r"r	0	0	+	+	+	+	0	+	+	+	0	+	0	+	0	+	+	+	0	+	6	4+	4+	3+		4+	4+	
7. rr	0	0	0	+	+	0	+	+	+	+	0	0	+	+	0	+	+	+	0	+	7	0	0	3+		4+	4+	
8. rr	0	0	0	+	+	0	+	+	+	+	+	0	0	+	+	0	+	0	+	+	8	0	0	3+		4+	4+	
9. rr	0	0	0	+	+	+	+	0	+	0	0	+	+	0	+	+	+	0	0	+	9	4+	4+	3+		4+	4+	
10. rr	0	0	0	+	+	+	0	+	0	+	0	+	0	+	0	+	0	+	0	+	10	4+	4+	3+		4+	4+	
Pt																			0	+	Pt	0	0	3+		4+	4+	

Eluate from patient's cells

	Rh					MNSs				P	Lewis		Kell		Duffy		Kidd		Lutheran			LISS				Enzyme		
	D	C	E	c	e	M	N	S	s	P1	Le^a	Le^b	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b		I.S.	37°	AHG	CC	37°	AHG	CC
1. R1R1	+	+	0	0	+	+	+	0	0	+	+	0	0	+	0	0	0	+	0	+	1			3+				
2. R1wR1	+	+	0	0	+	+	0	+	+	+	0	+	+	+	+	+	0	+	0	+	2			3+				
3. R2R2	+	0	+	+	0	0	+	0	+	+	0	+	0	+	0	+	0	+	0	+	3			3+				
4. Ror	+	0	0	+	+	+	0	0	+	0	0	0	0	+	0	0	+	+	0	+	4			3+				
5. r'r	0	+	0	+	+	+	0	0	+	0	+	0	0	+	+	+	0	+	0	+	5			3+				
6. r"r	0	0	+	+	+	+	0	+	+	+	0	+	0	+	0	+	+	+	0	+	6			3+				
7. rr	0	0	0	+	+	0	+	+	+	+	0	0	+	+	0	+	+	+	0	+	7			3+				
8. rr	0	0	0	+	+	0	+	+	+	+	+	0	0	+	+	0	+	0	+	+	8			3+				
9. rr	0	0	0	+	+	+	+	0	+	0	0	+	+	0	+	+	+	0	0	+	9			3+				
10. rr	0	0	0	+	+	+	0	+	0	+	0	+	0	+	0	+	0	+	0	+	10			3+				
Pt																			0	+	Pt							

An attempt was made to chloroquine treat the patient's RBCs so that a warm autoadsorption and antigen phenotyping could be performed. However, her RBCs proved too fragile, undergoing lysis in this reagent, and indeed, on standing overnight. Instead, an extended blood group antigen phenotype was determined for both the father and mother, and an alloadsorption with selected donor RBCs was attempted.

Antigen Phenotype

	D	C	E	c	e	K	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	S	s	M	N
Patient; "saline" antisera	w+	**0**	0	w+											**0**	
Positive control		**4+**	**4+**	**4+**											**4+**	
Negative control	**0**	**0**	**0**	**0**											**0**	
Father	**0**	**0**	**0**	+	+	**0**	**0**	**0**	+	**0**	+	**0**	+	+	**0**	+
Mother	+	**0**	+	+	**0**	**0**	**0**	**0**	**0**	+	+	**0**	**0**	+	**0**	+
Projected patient phenotype	+	**0**	+	+	+	**0**	**0**	**0**	+	+	+	**0**	**?**	+	**0**	+

Patient serum alloadsorbed x8 with ZZAP treated cells; adsorbing cell phenotype C, E, S, K, Fy^a, Fy^b, Jk^a negative, M positive

	Rh					MNSs				P	Lewis		Kell		Duffy		Kidd		Lutheran			LISS				Enzyme		
	D	C	E	c	e	M	N	S	s	P1	Le^a	Le^b	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b		I.S.	37°	AHG	CC	37°	AHG	CC
1. R1R1	+	+	**0**	**0**	+	+	+	**0**	**0**	+	+	**0**	**0**	+	**0**	**0**	**0**	+	**0**	+	**1**	**4+**	**4+**	**2+**		**0**	**0**	**2+**
2. R1wR1	+	+	**0**	**0**	+	+	**0**	+	+	+	**0**	+	+	+	+	+	**0**	+	**0**	+	**2**	**4+**	**4+**	**2+**		**0**	**0**	**2+**
3. R2R2	+	**0**	+	+	**0**	**0**	+	**0**	+	+	**0**	+	**0**	+	**0**	+	**0**	+	**0**	+	**3**	**0**	**0**	**1+**		**w+**	**3+**	
4. Ror	+	**0**	**0**	+	+	+	**0**	**0**	+	**0**	**0**	**0**	**0**	+	**0**	**0**	+	+	**0**	+	**4**	**4+**	**4+**	**2+**		**0**	**0**	**2+**
5. r'r	**0**	+	**0**	+	+	+	**0**	**0**	+	**0**	+	**0**	**0**	+	+	+	**0**	+	**0**	+	**5**	**4+**	**4+**	**2+**		**0**	**0**	**2+**
6. r"r	**0**	**0**	+	+	+	+	**0**	+	+	+	**0**	+	**0**	+	**0**	+	+	+	**0**	+	**6**	**4+**	**4+**	**2+**		**w+**	**3+**	
7. rr	**0**	**0**	**0**	+	+	**0**	+	+	+	+	**0**	**0**	+	+	**0**	+	+	+	**0**	+	**7**	**0**	**0**	**0**	**2+**	**0**	**0**	**2+**
8. rr	**0**	**0**	**0**	+	+	**0**	+	+	+	+	+	**0**	**0**	+	+	**0**	+	**0**	+	+	**8**	**0**	**0**	**0**	**2+**	**0**	**0**	**2+**
9. rr	**0**	**0**	**0**	+	+	+	+	**0**	+	**0**	**0**	+	+	**0**	+	+	+	**0**	**0**	+	**9**	**4+**	**4+**	**2+**		**0**	**0**	**2+**
10. rr	**0**	**0**	**0**	+	+	+	**0**	+	**0**	+	**0**	+	**0**	+	**0**	+	**0**	+	**0**	+	**10**	**4+**	**4+**	**2+**		**0**	**0**	**2+**
Pt																					**Pt**							

Eluate from the alloadsorbing cell

	Rh					MNSs				P	Lewis		Kell		Duffy		Kidd		Lutheran			LISS				Enzyme		
	D	C	E	c	e	M	N	S	s	P1	Le^a	Le^b	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Lu^a	Lu^b		I.S.	37°	AHG	CC	37°	AHG	CC
1. R1R1	+	+	0	0	+	+	+	0	0	+	+	0	0	+	0	0	0	+	0	+	1			4+				
2. R1wR1	+	+	0	0	+	+	0	+	+	+	0	+	+	+	+	+	0	+	0	+	2			2+				
3. R2R2	+	0	+	+	0	0	+	0	+	+	0	+	0	+	0	+	0	+	0	+	3			4+				
4. Ror	+	0	0	+	+	+	0	0	+	0	0	0	0	+	0	0	+	+	0	+	4			4+				
5. r'r	0	+	0	+	+	+	0	0	+	0	+	0	0	+	+	+	0	+	0	+	5			2+				
6. r"r	0	0	+	+	+	+	0	+	+	+	0	+	0	+	0	+	+	+	0	+	6			3+				
7. rr	0	0	0	+	+	0	+	+	+	+	0	0	+	+	0	+	+	+	0	+	7			3+				
8. rr	0	0	0	+	+	0	+	+	+	+	+	0	0	+	+	0	+	0	+	+	8			3+				
9. rr	0	0	0	+	+	+	+	0	+	0	0	+	+	0	+	+	+	0	0	+	9			3+				
10. rr	0	0	0	+	+	+	0	+	0	+	0	+	0	+	0	+	0	+	0	+	10			3+				
11. U-								0	0															3+				
12. Js^b-																								3+				
Pt																					Pt			*				

A valid autocontrol could not be performed because DAT negative patient RBCs were not obtainable.

Because of her unknown future need for transfusion, an attempt was made to provide the patient with RBCs that were negative for C, K, Fy^a, and Fy^b in addition to M and E, but this was not always possible. Before transfusion repeat alloadsorption with the same cell shown above was performed in an attempt to detect further alloimmunization.

RESPONSE TO TREATMENT

The patient was hydrated and placed on a broad spectrum antibiotic which was discontinued when the cultures were reported negative. Prednisone was started, initially at a dose of 3 mg/kg/D, decreased to 2 mg/kg/D at discharge. The patient was transfused with RBCs delivered in multiple small aliquots over the course of 24 hours to a total of 20 ml per kg. This elevated the hematocrit to 25.5%, but it fell to 21.6% 48 hours later (hemoglobin 8.4 g/dL and 7.1 g/dL respectively). The patient was discharged 3 days after admission to be followed with daily blood counts.

Severe anemia (hemoglobin 3.07 g/dL, hematocrit 11.6%) requiring readmission recurred 5 days after the initial transfusion, and again 4 days later, in spite of transfusion and an increase in the dose of prednisone. Anti-nuclear and HIV antibody tests were negative, and complement levels were normal. Ten days after the initial diagnosis intravenous immune globulin (IVIG) was started at a dose of 1 gm/kg/day for 2 days.

Her hemoglobin level remained stable for 10 days, but she subsequently required transfusion every 2 to 4 days in spite of five additional doses of IVIG over the course of 4 weeks. She was then started on cyclosporine. Her requirement for transfusion gradually decreased over the following month (see table), after which she became transfusion *in*dependent for 5 weeks. Unfortunately, however, there were problems maintaining adequate cyclosporine levels, and she followed a relapsing/remitting course during which her requirement for transfusion appeared to correspond to low cyclosporin levels or to acute infectious episodes.

QUESTIONS:

1. Why wasn't an autoadsorption done in this case?

2. Why were the parental phenotypes determined? Why might the E antigen typing using "saline" anti-E and that predicted from the parents phenotypes have been discrepant?

3. What allo- and/or autoantibodies are present in this case?

4. Comment on the patient's clinical presentation, treatment, and course.