PeliCase 2

Batch No.:	8000457790	Expiry date: 2022-12-05
Name:		Hospital:
Signature:		Ward:

Case:
A 34-year-old woman gave birth to her second child, a daughter, in hospital. Because she formed an anti-C and anti-K during her first pregnancy, she is being monitored by a gynaecologist and gave birth in a hospital. These were the only antibodies which showed up in the antibody screening during her pregnancy. At birth, the baby's haemoglobin and bilirubin levels were 9.9 mmol/l and 100 micromol/l respectively. Her bilirubin level increased from 131 to 234 micromol/l between Day 2 and Day 5.

We have been asked to select blood for a transfusion.

What are your findings and what do you recommend?

Results:

Erythrocytes (mother): (PeliCase cells)

Anti-A	Anti-B	Anti-A,B	Anti-D (1)	Anti-D (2)	Control

Serum (mother): (PeliCase serum)

Serum counter-control				ABO/RhD
A1 test erythrocytes	A2 test erythrocytes	B test erythrocytes	O test erythrocytes	

Antibody Screening				
Screening cell:	Column Technique	LISS/IAT	PEG/IAT	Coombs Control Cells
1				
2				
3				

	Antigen Typing							DAT			
	C	E	c	e	K			Poly	IgG	C3d	
typing erythrocytes (mother)											
typing erythrocytes (daughter)											

Erythrocytes (baby): (PeliCase cells)

Anti-A	Anti-B	Anti-A,B	Anti-D (1)	Anti-D (2)	Control

Serum (baby): (PeliCase serum)

Serum counter-control				ABO/RhD
A1 test erythrocytes	A2 test erythrocytes	B test erythrocytes	O test erythrocytes	

Antibody Screening				
Screening cell:	Column Technique	LISS/IAT	PEG/IAT	Coombs Control Cells
1				
2				
3				

Questionnaire

Specificity of antibody/antibodies:

1. Discuss the clinical relevance of the given situation in this case:

2. What possible explanations can be given for this?

3. What additional tests should be carried out to solve this case?

4. What is the transfusion recommendation?

PeliCase 2 is produced by Sanquin Reagents B.V.
If you have any questions regarding this panel, please contact Sanquin Reagents B.V.'s Marketing & Sales Department (tel.: +31 (0)20 5123599/email: reagents@sanquin.nl).

Reagent Documentation

Reagents for typing erythrocytes:

Reagent	*Producer*	*Batch No.*	*Expiry Date*
Anti-A			
Anti-B			
Anti-A,B			
Anti-D (1)			
Anti-D (2)			
Anti-C			
Anti-c			
Anti-E			
Anti-e			
Anti-K			
Anti-Fy^a			
Anti-Fy^b			
Anti-Jk^a			
Anti-Jk^b			
Anti-S			
Anti-s			
……..			
……..			
……..			
……..			
……..			
Monoclonal Control			
……			
A1 cells			
A2 cells			
B cells			
O cells			

Reagents for the antibody screening and identification/DAT

Reagent	*Producer*	*Batch No.*	*Expiry Date*
Polyspecific AHG			
Anti-IgG			
Anti-C3d			
Screening Panel			
Identification Panel			
Enzyme Panel			
Albumin			
LISS			
PEG			
…….			
…….			
…….			
…….			

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I. Results

The following antibodies were found in her serum:

PeliCase cells (mother)	**PeliCase serum (mother)**	**Eluate**
ABO determination: A RhD phenotyping: ccdee Typing: K negative DAT: Negative in Biorad LISS and Ortho kolom	Screening: posive Irregular antibodies: anti-C, anti-D en anti-K	nvt
PeliCase cells (baby)	**PeliCase serum (baby)**	**Eluate**
ABO determination: A RhD phenotyping: ccDEe Typing: K negative DAT: positive (IgG0 in Biorad LISS, tubes and Ortho kolom	Screening: positive Irregular antibodies: Anti-C, anti-D and anti-K	Anti-D and anti-K

The results in serum of **the mother**, using the different techniques are as follows:

Technique	**Reaction Strengths**			
	Depending on antigen expression			
	Anti-C	Anti-D	Anti-K	
KT Albumine	-	(+)	(+)	
37°C Albumine	2+	2+	+	
IAT Albumine	+	2+	2+	
PEG IAT	2+	2+	2+	
Bromeline 2 staps	2+	3+	2+	
Biorad LISS kolom	-	2+	2+	
Biorad papaïne	4+	4+	-	
Ortho Biovue	-	2+	2+	
Cellbind	-	3+	3+	

The results in serum of **the baby**, using the different techniques are as follows:

Technique	**Reaction Strengths**			
	Depending on antigen expression			
	Anti-C	Anti-D	Anti-K	
KT Albumine	-	-	-	
37°C Albumine	-	3+	+	
IAT Albumine	+	2+	2+	
PEG IAT	2+	2+	2+	
Bromeline 2 staps	nvt	nvt	nvt	
Biorad LISS kolom	(+)	3+	2+	
Biorad papaïne	4+	4+	-	
Ortho Biovue	-	2+	2+	
Cellbind	-	+	3+	

The results in <u>eluat</u> of **the baby**, using the different techniques are as follows:

Technique	**Reaction Strengths**			
	Depending on antigen expression			
	Anti-C	Anti-D	Anti-K	
KT Albumine	-	-	-	
37°C Albumine	-	3+	-	
IAT Albumine	-	2+	-	
PEG IAT	-	3+	-	
Bromeline 2 staps	nvt	nvt	nvt	
Biorad LISS kolom	-	4+	-	
Biorad papaïne	nvt	nvt	nvt	
Ortho Biovue	-	4+	-	
Cellbind	-	4+	(+)	

	Crossmatch IAT				
	Bovine/Albumine	PEG	LISS	Ortho	Cellbind
mother	neg	neg	neg	neg	neg
Baby	neg	neg	neg	neg	neg

II. Discussion

The mother's irregular antibodies can pose a threat to her and her baby. These antibodies may have resulted from previous pregnancies or transfusions. For the mother, this means that matching blood should always be found for transfusions necessary at a later stage. For the baby, this means that haemolysis can occur during and immediately after pregnancy.

If a reactivity pattern of anti-(C + D) is found, anti-G should also be considered.
The G antigen is an important Rhesus antigen discovered by Allen & Tippett in 1958. The G antigen is present on almost all D-positive or C-positive erythrocytes but is absent on D-negative and C-negative erythrocytes.
Although it is not necessary to know the difference between
anti-D and anti-C in relation to anti-G in a standard transfusion, this is important during pregnancy. This is important since anti-G can be disguised as anti-D and anti-C

in the standard identification. Because of this false presence of anti-D, patients do not receive the anti-D immunoglobulin which they should receive. Pregnant women with anti-D or anti-G run the risk of developing a haemolytic disease of the foetus and new-born and should therefore be monitored during pregnancy. In pregnancies with anti-G, alloimmunization with anti-D can be prevented through treatment with anti-D prophylaxis.
The antibody should be properly identified. This can be done through differential adsorption and elution, using R_2R_2 and r'r cells to see the difference between anti-G or anti-D and anti-C.
Your immunohematology reference laboratory can carry out this test.

III. RIVM (National Institute for Public Health and the Environment) Blood Group Antibodies and Pregnancy

All pregnant women are offered a blood test in or around week 12 to examine their blood groups and/or to determine whether they have irregular antibodies. They are also checked for hepatitis B, HIV and syphilis.

a) If they test negative for irregular antibodies but are Rhesus D negative, another blood test is carried out in Week 27.
   The unborn child's DNA in the blood of the Rhesus negative mother can be used to determine his/her Rhesus blood group.
   If the baby is Rhesus positive, an anti-D injection is given in Week 30 to prevent the formation of antibodies. This injection is given again after childbirth.
b) If the mother tests negative for irregular antibodies and the Rhesus-c antigen, her blood is again tested for antibodies in Week 27. If she tests positive, additional blood tests and ultrasounds are carried out.
c) If the mother tests positive for irregular antibodies, further tests should be carried out and another blood sample should be taken. The mother's antibodies can only break down the baby's blood if they are targeting the baby's blood group. To determine the baby's blood group, it may be necessary to determine the father's blood group.

To determine how active the antibodies in the mother's blood are and how likely they are to break down the baby's blood, further tests need to be carried out. Depending on test results, a pregnant woman can either continue to be monitored by her midwife or GP (ADCC <10%) or be referred to a gynaecologist (between 30% and 50%) for additional check-ups. They will use ADCC assays to monitor the mother's blood and to determine whether the baby is developing anaemia (haemolytic disease of the foetus and new-born (HDFN)) and, if so, how serious this is.

If there are signs that the baby is developing a serious form of anaemia, it may be necessary to induce labour or to perform a C-section. It is sometimes necessary to treat a baby before birth with intrauterine transfusions (IUTs). These can be given from the 16th week of pregnancy.

A standard blood transfusion may need to be given after birth to increase the haemoglobin concentration. The bilirubin level should also be reduced. This can be done by placing the baby under an ultraviolet (UV) light. If this doesn't help sufficiently, a replacement transfusion may be considered, replacing a significant part of the baby's blood with donor blood. This is done to prevent brain damage.

Considerations

If a pregnant woman has formed irregular IgG-antibodies, it is advisable to test her blood and that of her baby immediately after birth with:

1. The baby's ABO and Rhesus D
2. DAT on the baby's erythrocytes
3. Elution of the baby's erythrocytes
4. Titre and specificity of the antibodies in the baby's serum
5. Titre and specificity of the antibodies in the mother's serum
6. If necessary, look for IgG-antibodies anti-A or anti-B in the mother's serum
7. Look for low-frequency antibodies in the mother's serum
8. ADCC assay.

IV. Blood Transfusion Guidelines (Dutch Institute for Healthcare Improvement CBO, 2011, module 3.7.1. including the revisions of 15-10-2020)

Selection of ABO-RhD-compatible units.
The blood for a replacement transfusion is pooled blood from a filtered ABO-compatible EC (both mother and baby), less than 72 hours old + AB plasma. The donor erythrocytes do not have the antigens which the mother developed antibodies against.

In case of a transfusion for a neonate less than 90 days old, an IAT crossmatch needs to be carried out with the mother's plasma. This is done to look for low-frequency antibodies which are not detected with a panel of test erythrocytes. The main reasons for this are that the titre of the erythrocyte antibodies is more elevated in the mother and that the material is more readily available. Obtaining material from the neonate to carry out crossmatches can worsen his/her aneamia and is not necessary if there is blood available from the mother. If possible, the mother's material should be obtained within 3 days of childbirth.

According to the guidelines, women below the age of 45 should be matched for the c and K antigen, obviously taking account of the existing antibodies.

V. Transfusion Recommendation

Select units of blood groups A, Rh D(-), C(-) and K(-)

Additional information:

1. Issitt PD. Applied blood group serology. 4th edition. Durham, NC: Montgomery Scientific Publications, 1998.

2. Mollison's, Blood transfusion in clinical medicine. HG Klein, DJ Anstee. 12th edition. Blackwell Publishing 2014.

3. The blood group antigen factsbook. Marion E. Reid, Christine Lomas-Francis and Martin L. Olsson, London: Academic Press, 2012 3rd edition.

4. Sanquin Erythrocyte Serology.

5. Dutch Institute for Healthcare Improvement CBO Blood Transfusion Guideline.

6. National Institute for Public Health and the Environment Rhesus Blood Group during pregnancy 2020.

7. Indian J Hematol Blood Transfus. 2017 June; 33(2): 259-263