

Frequently Asked Questions about the use of Rh (D) immunoglobulin

Abbreviations used in this section:

ANZSBT: Australian and New Zealand Society of Blood Transfusion

FMH: Fetomaternal Haemorrhage

National Blood Authority (2003) [Approved by the NHMRC]: *Guidelines on the prophylactic use of Rh D immunoglobulin (Anti-D) in obstetrics*

Testing Questions and Answers

Introduction

The following answers have been prepared and endorsed by ANZSBT. The recommendations are supported by the literature review and are in accordance with ANZSBT Guidelines: *Laboratory Assessment of Fetomaternal Haemorrhage* (2002) and, *Guidelines for Blood Grouping & Antibody Screening in the Antenatal & Perinatal Setting, 2nd Edition* (2003).

Testing Questions

1. FMH screening

- 1.1 *When should I perform a FMH screen?*
- 1.2 *What tests are available to quantify a FMH?*
- 1.3 *What is the best test to quantify a FMH?*
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- [1.5](#) *What is the average size of a FMH?*

2. Red cell antibody screening

- 2.1 *What test should be performed to detect red cell antibodies?*
- 2.2 *When should red cell antibody screening be performed?*
- 2.3 *Do you still need to perform a red cell antibody screen if Rh (D) immunoglobulin is given prophylactically?*
- 2.4 *When should the blood sample for the red cell antibody screen be collected in relation to the administration of Rh (D) immunoglobulin at 28 and 34 weeks?*
- 2.5 *How should the results of a red cell antibody screen be interpreted if antenatal prophylaxis has been previously given?*
- 2.6 *Is it possible to distinguish between passively-acquired and actively-acquired Anti-D?*

2.7 *From a laboratory perspective, sometimes information is missing regarding whether a patient has received Rh (D) immunoglobulin prophylactically. What would you recommend?*

2.8 *At 28 weeks presentation, is antibody screening advised as well as assessment of FMH?*

3. **Other**

3.1 *Should I quantitate the amount of Rh (D) antibody that has been given to check if there is enough antibody?*

QUESTIONS & ANSWERS

1. FMH Screening

1.1 *When should I perform a FMH screen?*

For potentially sensitising events that occur after the first trimester.

In this circumstance a maternal sample should be taken prior to administration of Rh (D) immunoglobulin, to assess the volume of FMH. However, at no time should a single dose of Rh (D) immunoglobulin be withheld based upon, or pending, the results to quantitate FMH.

1.2 *What tests are available to quantify a FMH?*

There are a number of tests available to assess the volume of FMH and allow additional Rh (D) immunoglobulin to be given where appropriate. The main tests used are:

- **The Kleihauer Acid Elution Test** — which is widely used but relies on subjective interpretation. Kleihauer testing appears to be precise only in small volumes of transplacental haemorrhage. It gives quantitative results but is open to interpretation by the laboratory staff performing the test, which has resulted in a number of cases of inaccurate results. Consequently the experience of the laboratory staff performing the test plays a major role in the success of the test. In addition, Kleihauer testing involves identification of haemoglobin F (HbF), which may lead to false positive results in the presence of inherited conditions resulting in elevated levels of HbF in the adult circulation.
- **Flow Cytometry** — which is reliable and accurate, but not widely available outside metropolitan areas.

1.3 *What is the best test to quantify a FMH?*

Flow cytometry is accepted as the most accurate quantitative test for FMH and is the method of choice for quantitation if readily available.

However, until flow cytometry becomes more widely available the NHMRC Report (2003) has stated that the following recommendations must be ensured:

- laboratories undertaking quantitative assessment of FMH by any method (such as Kleihauer-Betke test) must show acceptable performance in internal and external quality assurance programs and have clearly defined test methods, continuing

assessment protocols and documented staff training programs to ensure accuracy and reproducibility of results;

- results should be reported in a format that allows easy correlation with product inserts of locally available Rh (D) immunoglobulin;

1.4 What should I do if the FMH quantitation shows that the FMH is greater than that covered by the dose of Rh (D) immunoglobulin already administered?

Where FMH quantitation shows that fetomaternal haemorrhage greater than that covered by the dose already administered has occurred, administration of an additional dose(s) of Rh (D) immunoglobulin sufficient to provide immunoprophylaxis must be administered and preferably within 72 hours.

For large bleeds follow up testing should be performed on a sample collected 48 hours post Rh (D) immunoglobulin administration, to determine if further dosing is required. Supplemental Rh (D) immunoglobulin should be administered if:

- FMH is still positive; and
- Rh (D) immunoglobulin is not detected in maternal plasma by IAT (Indirect Antiglobulin Test).

1.5 What is the average size of a FMH?

About:

- 99% of fetal bleeds are less than 5 mL of red blood cells.
- 50 % of bleeds are less than 0.05 mL
- 0.6% of bleeds may be higher than 30 mL.

2. Red cell antibody screening

2.1 What test should be performed to detect red cell antibodies?

In the absence of evidence to the contrary, antibody screening using an indirect antiglobulin test [IAT] test should be used.

2.2 When should red cell antibody screening be performed?

When no clinically significant antibodies are detected at the first trimester the value of repeat testing in all pregnancies at 28 weeks or later has been questioned [Judd 2001]. However repeat testing of RhD negative women prior to administering RhD immunoprophylaxis at 28 weeks is becoming the accepted protocol in most Australian centres, as is the elimination of the antibody screen at 34-36 weeks.

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2.3 Do you still need to perform a red cell antibody screen if Rh (D) immunoglobulin is given prophylactically?

Yes, as other clinically significant red cell antibodies may be detected.

2.4 *When should the blood sample for the red cell antibody screen be collected in relation to the administration of Rh (D) immunoglobulin at 28 and 34 weeks?*

It is paramount that the blood sample is taken **before** the administration of the antenatal Rh (D) immunoglobulin, and it is acceptable for the Rh (D) immunoglobulin to be given immediately after the blood has been taken, before the results are available. This is because the vast majority of Rh(D) negative women will not be sensitised and this is the most practical approach for optimising patient care.

2.5 *How should the results of a red cell antibody screen be interpreted if antenatal prophylaxis has been previously given?*

If the clinical notes clearly state that Rh (D) immunoglobulin has been given prophylactically, the ANZSBT recommend that this be reported as ‘Anti-D detected, likely to be consistent with administration of passive (prophylactic) anti-D given at 28 or 34 weeks.’

If no clinical notes are provided, ANZSBT recommend that this be reported as ‘Anti-D antibody alone is detected in this antenatal sample from this Rh (D) negative woman. This may be indicative of either passively administered prophylactic anti-D or active allosensitisation. If this antibody is not the result of prophylaxis then further antenatal serology should be performed at 4 weekly intervals through to 34 weeks gestation and thereafter at 2 weekly intervals until term. If the antibody titre is more than or equal to 1:32, referral to a maternal-fetal medicine specialist is recommended.’

See 'Topics in Transfusion Medicine', vol 10, no 1, May 2003. ANZSBT. Also at www.anzsbt.org.au

2.6 *Is it possible to distinguish between passively-acquired and actively-acquired Anti-D?*

No, that is why the patient’s history and clinical notes on request forms are so important.

2.7 *From a laboratory perspective, sometimes information is missing regarding whether a patient has received Rh (D) immunoglobulin prophylactically. What would you recommend?*

It is important to emphasise to the clinician to fill in the information on the request form. You could introduce a special box on the request form to indicate whether Rh (D) immunoglobulin has been given prophylactically.

2.8 *At 28 weeks presentation, is antibody screening advised as well as assessment of FMH?*

Antibody screening should be done at 28 weeks. There is no requirement for FMH screening at 28 weeks unless there is a concern over a potential sensitising event.

3. Other

3.1 Should I quantitate the amount of Rh (D) antibody that has been given to check if there is enough antibody?

Quantitation of the presence of Rh (D) immunoglobulin is not a reliable approach to determining if sufficient Rh (D) immunoglobulin has been given. This is why it is important to quantitate the magnitude of the fetal maternal haemorrhage (FMH). Additional doses of Rh (D) immunoglobulin may be required if the bleed is greater than that covered by the dose of Rh (D) immunoglobulin already given.