CLINICAL PRACTICE GUIDELINE

Blood group management & clinically significant antibodies: Rh D negative & Rh D positive women

This document should be read in conjunction with the Disclaimer

Contents

Aims

- To determine the woman's ABO and Rh(D) blood group and to detect the presence of atypical red cell antibodies, the primary aim of which is to:
  - identify women who are alloimmunised so they may undergo serological follow up and fetal assessment,
  - identify Rh(D) Negative women who may require the administration of prophylactic Rh(D) Immunoglobulin (RhD-Ig),
  - identify antibodies with potential to cause haemolytic disease of the fetus and newborn (HDFN)
  - provide selected, compatible blood for fetal and maternal transfusion when required.
- To administer Rh(D) Immunoglobulin (RhD-Ig) to non-sensitised Rh(D) Negative women in a timely manner so the risk of maternal sensitisation to fetal Rh(D) Positive red blood cells is reduced.
- To guide staff on the procedure for administration of RhD-Ig (Anti-D) prophylaxis, given to prevent Rh(D) isoimmunisation in non-sensitised Rh(D) Negative women antenatally, postnatally and with pregnancy loss or sensitising events.

Background

Blood Group incompatibility between a pregnant woman and her baby can lead to maternal antibody sensitisation and transfer of clinically significant antibodies across the placenta which may cause Haemolytic Disease of the Fetus and Newborn (HDFN).

The most common example of incompatibility is Rh(D) blood group incompatibility between a Rh(D) Negative woman and her Rh(D) Positive infant which may cause alloimmunisation against the Rh(D) antigen.[1] A sensitised woman may develop immune Anti-D, which crosses the placenta binding to, and destroying, fetal Rh(D) Positive blood cells.[1] This can result in anaemia and fetal hydrops.[1] Severe HDFN can result in oedema, hepatosplenomegaly, severe anaemia, jaundice and / or death.

Prophylactic RhD-Ig is a commercial preparation of human Anti-D.[1] The administration of RhD-Ig as soon as possible and within 72 hours of a fetomaternal haemorrhage (FMH) can remove fetal Rh(D) Positive cells from the maternal circulation so that sensitisation does not occur.[1] If RhD-Ig is not administered within 72 hours, a dose offered up to 9-10 days might still provide some protection.

Administration of RhD-Ig to a Rh(D) Negative woman within 72 hours of the birth of a Rh(D) Positive infant reduces the incidence of Rh isoimmunisation from about 13% to 2%. A small number of Rh(D) Negative women (1.5-1.8%) are still immunised by their Rh(D) Positive fetus despite RhD-Ig administration post-partum. Studies show this can be reduced to <0.2% if RhD-Ig is also given at 28 weeks and 34 weeks gestation[1].

The Kleihauer Test is used to identify women with a large FMH (>6 mL of packed fetal red cells) who may need **additional** doses of RhD-Ig to ensure clearance of fetal red cells. A Negative Kleihauer test indicates that one dose of RhD-Ig is sufficient.

A 625 IU dose of RhD-Ig can destroy the equivalent of a 6 mL bleed of packed fetal red cells. For Positive Kleihauer counts repeat the Kleihauer test 48 hours after administration of RhD-Ig as advised by The Transfusion Medicine Unit (TMU).

Blood Group & Antibody Screening in Pregnancy

Antenatal testing protocols

Table 1- Antepartum blood grouping & antibody screening at KEMH

		Gestation			
		1st visit 19-20 Weeks	**28-30 Weeks**	**36 weeks**	**On admission**
Rh(D) Positive	**ANC women**	G&S (if no current results)			G&S if appropriate*
	Shared care women	G&S (if no current results)			G&S if appropriate*
Rh(D) Negative	**ANC women**	G&S	G&S **Prophylactic RhD-Ig**	**Prophylactic RhD-Ig**	G&S*
	Shared care women	G&S (if no current results)	G&S (if no current results) **Prophylactic RhD-Ig**	**G&S (if not done in this pregnancy) Prophylactic RhD-Ig**	G&S*

* A pre-delivery Group and Screen (G&S) sample should be collected on admission to the Labour and Birth Suite or Family Birth Centre (or the Preadmission clinic if an elective Caesarean section birth is planned) if:

- **atypical red cell antibodies are present,**
- **the woman's serological history is unknown,**
- **prophylactic RhD-Ig has been given,**
- **there is increased risk of requiring a blood transfusion.**

Note: If blood for G&S is collected at an 'external' Pathwest collection centre, the pathology request form must request the sample be sent to KEMH. For additional background information Refer to Transfusion FAQ: What is a Group and Screen

Initial visit

At the first visit, current G&S results are required on **ALL** pregnant women, regardless of blood type. If a copy of this report does not accompany the woman to her first antenatal visit, a blood sample should be taken for this purpose before she leaves the clinic.

Subsequent visits

1. Refer to Clinical Significance of Antibodies (in this guideline) if clinically significant antibodies are detected.
2. Rh(D) Negative women should attend KEMH's antenatal clinic at 28-30 weeks gestation for administration of RhD-Ig unless administration is arranged by their GP. Shared care women who have had a G&S performed outside KEMH at 28 weeks, should have a copy of the report sent to TMU. Provided the outside G&S is current (within two weeks of the request for RhD-Ig) TMU will then issue RhD-Ig for administration in the clinic. If the woman's results are unavailable, a G&S should be sent to TMU where, upon receipt of the sample, RhD-Ig will be issued.
3. All Rh(D) Negative pregnant women attending KEMH at 34-36 weeks gestation will be offered RhD-Ig. This will be issued by TMU. A G&S request is only required if the patient has not had previous testing performed during the current pregnancy
4. It is essential that the following information is provided on the request form accompanying blood samples to TMU, including:
   - Previous history of transfusion or pregnancy
   - Previous history of antibodies, especially if reported at an outside facility
   - Gestation
   - Dates of any prophylactic RhD-Ig administered in the last 3 months, especially if given at an outside facility.

Clinical significance of antibodies

It is important to understand that both Rh (D) Negative and Rh(D) Positive women may produce clinically significant antibodies that may cause HDFN. Antibodies that cause HDFN are IgG and reactive by the Indirect Antiglobulin Test. The clinical significance of antibodies detected during routing prenatal testing should be assessed in association with the Maternal Fetal Medicine (MFM) Specialist and the TMU. Refer to table 7.4 page 44 ANZSBT Guidelines for Transfusion and Immunohaematology Practice. Antibody prevalence can vary between countries reflecting geographical or racial variations in gene frequencies.

Anti-D, Anti-c and Anti-K are most commonly implicated in causing HDFN severe enough to warrant prenatal intervention.

1. **HDFN caused by maternal anti-K (or other K system antibodies):**
   - previous obstetric history is not predictive of disease severity
   - Anti K impairs haemopoesis as well as causing haemolysis
   - Referral to a MFM specialist should occur regardless of the antibody titre
   - Anti-K titres do not correlate with disease severity.
   - Paternal K antigen status should be checked and if the phenotype is K Positive or unknown, fetal genotyping may be undertaken using fetal DNA obtained from maternal plasma on referral to ARCBS via TMU.

Principles of management of isoimmunisation

1. If clinically significant antibodies are detected at the first antepartum visit, these antibodies will be identified and a titration performed. Thereafter antibody investigation and titration should be repeated **every 4 weeks until 28 weeks** gestation and **then every 2 weeks thereafter** or as advised by TMU/ Australian Red Cross Blood Service (ARCBS) Red Cell Reference laboratory. The TMU will refer anti-D and anti-c antibodies to ARCBS for antibody quantitation.
2. A clinically significant rise in the antibody titre (or quantitation) will assist the clinician in determining when to initiate fetal monitoring, such as Doppler ultrasound and cordocentesis. Once fetal monitoring has been initiated the specialist will determine the frequency of further serological testing.
3. The MFM Specialists should perform an antepartum assessment of the severity of the HDFN. The following are indications for referral to a MFM Specialist:
    - An antibody that may cause HDFN with the titre reaching or exceeding 1:32. The MFM Specialist and Haematologist/ TMU will advise on antibody significance.
    - Anti-K at any titre when the paternal phenotype is K positive.
    - **All women who have had an infant previously affected by HDN. These women should be referred to a specialist as soon as possible and preferably before 20 weeks gestation irrespective of antibody level. The partner's blood group and genotype should be obtained as early as possible in the pregnancy.**

The Kleihauer Test

The Kleihauer test is performed on a maternal sample. It is used to assess the volume of a feto-maternal haemorrhage (FMH) and determine if additional doses of RhD-Ig are required. For a full summary of Kleihauer indications refer to WNHS Transfusion Protocols: The Kleihauer Test and Feto-Maternal Haemorrhage

Birth / Postpartum Kleihauer testing

Maternal sample

A pre-delivery G&S sample should be collected and sent to TMU on admission to the Maternal Fetal Assessment Unit/ Labour and Birth Suite (or Pre-Admission Clinic if an elective Caesarean section birth is planned) if:

- atypical red cell antibodies are present,
- the woman's serological history is unknown,
- prophylactic RhD-Ig has been given in the previous 3 months,
- there is an increased risk of requiring a blood transfusion.

Refer to WNHS Transfusion Protocols: The Kleihauer Test and Feto-Maternal Haemorrhage

Cord sample

A cord blood sample is collected from all babies born at KEMH and sent to TMU.

A request for blood group and a Direct Antiglobulin Test (DAT) should be made for all neonates born to a mother who:

- is Rh(D) Negative or,
- has known clinically significant antibodies or,
- has unknown maternal blood group and antibody status.

Where the cord sample is Rh(D) Positive and the mother is Rh(D) Negative, RhD-Ig will be supplied by TMU for administration to the mother.

A request for a blood group and DAT should be made for all infants with unexplained neonatal jaundice. If the DAT is positive, a bilirubin estimation and a haemoglobin level should be determined on a *peripheral blood sample taken from the infant. *Bilirubin may be performed on the cord sample ONLY if a cord sample in lithium heparin anticoagulant is supplied.

Note: When a Rh(D) Negative mother receives RhD-Ig during pregnancy, especially as routine prophylaxis at 28-30 and 34-36 weeks gestation:

- a Rh(D) Positive infant may be born with a positive DAT but no evidence of haemolysis
- the maternal sample may show Anti-D reactivity, as the half-life of passive RhD-Ig in the absence of FMH, is approximately 21 days.

The Haematologist can offer advice on the significance of these results.

Rh(D) Immunoglobulin (formerly Anti-D)

Administration of Rh(D) Immunoglobulin

- RhD-Ig should be administered by deep intramuscular injection (IMI). It should not be given subcutaneously.
- The deltoid muscle or anterolateral thigh is the preferred site. The buttocks should be avoided.
- For women with a Body Mass Index (BMI) of 30 or more, particular consideration should be given to factors which may impact on the adequacy of the injection, including the site of administration and the length of needle used.

Indications for the use of Rh(D) Immunoglobulin

RhD-Ig is issued from TMU on completion of appropriate serological testing. Refer to WNHS Transfusion Protocols: Rh D Immunoglobulin Products and Applications

Table 1

Summary of Rh(D) Immunoglobulin Indications

Event	RhD-Ig Dose
FIRST trimester sensitising events	CSL Minidose 250 IU (CSL 625 IU in multiple pregnancy)
Second and third trimester sensitising events	CSL 625 IU
Routine prophylaxis at 28-30 and 34-36 weeks	CSL 625 IU
Postpartum	CSL 625 IU
Rh(D) positive platelets	CSL Minidose 250 IU

Rh(D) Immunoglobulin at 28 and 34 weeks in antenatal clinics: Prophylactic administration

Instruction	Criteria	Role of the Midwife
Midwives working in the Antenatal clinics at KEMH may administer prophylactic RhD-Ig to Rh (D) Negative women at: • 28-30 and • 34-36 weeks gestation.	• All Rh(D) Negative women booked at KEMH who have had a current group and screen performed at KEMH, or has had a group and screen performed by an external laboratory within 2 weeks of the request for RhD–Ig and the results are available.	1. Identify that the woman has a Rh(D) Negative blood group. 2. Confirm the blood group with the **hard copy** of the results from pathology. Verbal confirmation by the woman or the blood group documented in the MPower is **not** appropriate. If a hard copy report is unavailable as the patient has only had a blood group and antibody screen performed on that visit, the results may be cross checked against the ICM computer report or verified with the Transfusion Scientist in TMU. This should be clearly documented in the Medical Record. 3. If there are no blood group results available or the results are from an external laboratory and they were processed more than 2 weeks prior to the request for RhD-Ig, complete a pathology form. Information that must be provided includes: • The maternal blood group (if known) • Any administration of RhD-Ig earlier during the pregnancy • A request for blood group and antibody screening 4. If the maternal blood group is available and current, telephone TMU, provide the woman's details and request RhD-Ig. 5. TMU will dispatch the vial via the electronic chute. 6. On arrival obtain informed consent; provide the woman with the brochure "Anti-D. You and Your Baby". Confirm patient identity with label on RhD-Ig vial. Ask the woman to state her name & date of birth & cross check details against ID band (if inpatient) as per Blood product checking procedure WNHS Transfusion Protocols: Checking Procedure Pre Administration of Blood Products 7. Complete the MR007 RhD (Anti D) Immunoglobulin Record. 8. The RhD-Ig should be administered as per Guideline: Pharmacy- Medication Safety: Administration & Checking Procedures by Nurse/Midwifery/Medical Staff & Students 9. The RhD-Ig must be given within 30 minutes of arrival at the clinic. If not required or delayed, it must be returned immediately to TMU. 10. Rh D-Ig must not be stored in any fridge other than TMU controlled blood fridges.

Rh(D) Immunoglobulin: Administration

Note: This guideline should be read in conjunction with the relevant protocols
WNHS Transfusion Protocols: Rh D Immunoglobulin Products and Applications

WNHS Transfusion Protocols: The Kleihauer Test and Feto-Maternal Haemorrhage

Key points

1. The administration of RhD-Ig to Rh(D) Negative women with no immune Anti-D antibodies, results in a significant reduction in the incidence of Rh(D) isoimmunisation.[1-5]
2. In general, a 250IU dose is used <13 weeks gestation and a 625IU dose is used ≥13 weeks gestation. The 250IU dose is increased to 625IU under the following conditions:
    - Uncertain gestational age with a possibility of being ≥13 weeks gestation.[2]
    - Twin/ multiple pregnancy.[2]
3. For all dosages refer to Rh D Immunoglobulin Products and Applications
4. A Kleihauer test for FMH is not required at less than 20 weeks gestation.[5] However the woman's blood group and antibody screen should be performed to confirm her Rh(D) group and check for immune Anti-D.[5] If the patient is Rh(D) Negative then RhD-Ig is indicated for sensitising events.
5. All Rh D Negative women with sensitising events ≥20 weeks gestation (e.g. miscarriage, ectopic pregnancy, termination of pregnancy, , cordocentesis, abdominal trauma considered enough to cause FMH, antepartum haemorrhage, external cephalic version) or postpartum indications for RhD-Ig should have the magnitude of the FMH assessed[5] via Kleihauer test. This will determine if **additional** doses of RhD-Ig are required.[1, 5] A 625IU dose is sufficient to remove up to 6ml of fetal RhD Positive red blood cells.[2]
6. It is **essential** that a G&S is performed before the first dose of RhD-Ig is administered (within 14 days prior to injection). This will identify women who have already been sensitised[3] and at risk of HDFN. Following injection, prophylactic Anti-D is detectable in the maternal circulation in serological tests for up to 10 weeks and is serologically indistinguishable from immune Anti-D. Testing can be omitted at 34 weeks gestation if prophylactic Anti-D was given at 28 weeks[3].
7. RhD-Ig is supplied by the TMU.
8. If a woman refuses RhD-Ig this must be documented in the medical notes and the Medical Officer and TMU notified. See also Clinical Guidelines O&G: Refusal of Blood Transfusion and Blood Products: Management and Transfusion Medicine Protocol: Blood Product Prescription Consent and Refusal
9. RhD-Ig can interfere with the response to live attenuated vaccines, therefore postnatal administration of such vaccines including poliomyelitis and measles (contraindicated or not recommended in pregnancy)[6] should occur at least 3 months after any RhD-Ig administration.[2]

Contraindications

Rh(D) immunoglobulin-VF should not be given to women who:

- Are Rh(D) positive or "weak" (D) positive[2]
- Have already been sensitised to the Rh(D) antigen[2] and have an immune Anti D.

  Warning: Following injection, prophylactic Anti-D is detectable in the maternal circulation for up to 10 weeks and is indistinguishable from immune Anti-D. If there is any doubt about whether the Anti-D is immune or prophylactic, consult with the obstetric team[3] and discuss with TMU who will offer individual advice. Prophylactic Anti-D gradually decreases in strength and usually disappears within 10 weeks of injection. **Note**: Although there is no benefit giving RhD-Ig to an already sensitised woman, there is no more risk than when given to a non-sensitised woman[2].
- Have isolated Immunoglobulin A (IgA) deficiency, unless these women have had testing that shows they do not have circulating Anti-IgA antibodies.[2]
- Have severe thrombocytopenia or any coagulation disorder that contraindicates IMI.[2]

Precautions[2]

Rh(D) immunoglobulin-VF:

- Must not be given intravenously (IV) due to potential anaphylactic reactions.

  See Administration (point 12) below for the Anti-D option available for IV use.
- Caution advised if the woman has a history of prior systemic allergic reactions after administration of human immunoglobulin preparations.
- Must not be given to the Rh(D) Positive postpartum infant.
- If the woman's Body Mass Index (BMI) is ≥30, product information recommends to confirm clearance of fetal cells and the presence of Anti-D post administration.[2] However, a consensus position statement recommends routine post-administration testing may not be required unless there has been a large fetomaternal haemorrhage[7] >6mLs.

Adverse effects[2]

- Local tenderness, erythema and stiffness may occur at the site for several hours, as with any intramuscular injection.
- Occasional: Mild pyrexia, malaise, drowsiness and urticaria may occur after immunoglobulins.
- Rarely: Skin lesions, headache, dizziness, nausea, generalised hypersensitivity reactions and convulsions.[2]

Administration of RhD Immunoglobulin

1. Ensure the woman's blood group is Rh(D) Negative and that she does not have confirmed immune Anti-D (already sensitised).[1, 3]
2. If the woman is postpartum, check infant is Rh(D) Positive, that she does require RhD-Ig[5] and that RhD-Ig and dose is prescribed on the woman's medication chart.
3. When a Kleihauer test is indicated, check the sample has been collected prior to RhD-Ig injection. This test is used to determine if **additional** doses of RhD-Ig are required. A negative Kleihauer test (reported as <1mL fetal red cells) indicates that one dose of RhD-Ig is sufficient. It does NOT mean that RhD-Ig is unnecessary.
4. Ensure the woman is informed and appropriately counselled as to the reasons for requiring RhD-Ig. Inform the woman that RhD-Ig is a blood product and provide an Anti-D patient information leaflet. Anti D leaflets 'You & Your Baby' (for antenatal and post natal use) and 'Important Information for Rh(D) Negative Women' (for early pregnancy loss) may be ordered from TMU. Complete and sign the verbal consent section on the RhD Immunoglobulin (Anti D) Record form (MR007).
5. Check the product as per Clinical Guideline Pharmacy: Medication Safety: Administration & Checking Procedure by Nursing/Midwifery/Medical Staff and Students. Confirm the patient's identity with the label on the RhD-Ig vial. Ask the woman to state her name and date of birth and also cross check the details against the ID band (if inpatient) as per WNHS Transfusion Medicine Protocols: Checking Procedure Pre Administration of Blood Products
6. Check the vial of RhD-Ig with the naked eye. If it appears turbid or contains sediment it must not be used[2] and should be returned to the TMU.
7. RhD-Ig must be brought up to room temperature before use.[2]
8. Cleanse the skin with alcohol and allow skin to dry.
9. Administer the RhD-Ig **slowly by deep intramuscular injection only**.[2, 3]

   The deltoid muscle or anterolateral thigh is the preferred site. The buttocks should be avoided.

   For women with a high BMI (e.g. 30 or more), particular consideration should be given to factors which may impact on the adequacy of the injection, including the site of administration, access to underlying muscle and the length of needle used.[3]
10. After administration of RhD-Ig, attach the peel off label to the RhD Immunoglobulin Record form (MR007) and complete all sections of the form in the woman's medical record. Records should be maintained for Anti-D immunoglobulin traceability[5]
11. Large doses (> 5mL) should be administered in divided doses at different sites.[2]
12. If a larger dose of RhD-Ig is required to cover a massive FMH, then Rhophylac may be issued from the TMU refer to Rh D Immunoglobulin Products and Applications. Rhophylac is administered IV.[3]
13. Any adverse events related to RhD-Ig use should be reported to the TMU

References

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Related WNHS policies, procedures and guidelines

KEMH Clinical Guidelines:

- WNHS Transfusion Protocols:
  - Blood Product Prescription Consent and Refusal
  - Checking Procedure Pre Administration of Blood Products
  - Rh D Immunoglobulin Products and Applications
  - The Kleihauer Test and Feto-Maternal Haemorrhage
- Obstetrics & Gynaecology
  - Standard Protocols: Pathology & Ultrasound Ordering by Midwife/Nurse: (Kleihauer: Requesting; Kleihauer, Postnatal: Requesting; Cord Blood Group: Requesting)
  - Complications of Pregnancy: Refusal of Blood Components and / or Products: Management

Other related documents

- Australian Red Cross (Rhophylac information; Anti D prophylaxis) Frequently asked questions about the use of Rh D immunoglobulin
- CSL Behring: Product Information & Consumer Medicine Information

NHMRC: Guidelines on the Prophylactic Use of Rh D Immunoglobulin (Anti-D) in Obstetrics

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