

PREVENTION OF RH ALLOIMMUNIZATION

This guideline has been reviewed by the Maternal-Fetal Medicine Committee and the Genetics Committee, with input from the Rh Program of Nova Scotia, and approved by the Executive and Council of the Society of Obstetricians and Gynaecologists of Canada.

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Abstract

Objective: To provide guidelines on use of anti-D prophylaxis to optimize prevention of rhesus (Rh) alloimmunization in Canadian women.

Outcomes: Decreased incidence of Rh alloimmunization and minimized practice variation with regards to immunoprophylaxis strategies.

Evidence: The Cochrane Library and MEDLINE were searched for English-language articles from 1968 to 2001, relating to the prevention of Rh alloimmunization. Search terms included: Rho(D) immune globulin, Rh iso- or allo-immunization, anti-D, anti-Rh, WinRho, Rhogam, and pregnancy. Additional publications were identified from the bibliographies of these articles. All study types were reviewed. Randomized controlled trials were considered evidence of highest quality, followed by cohort studies. Key individual studies on which the principal recommendations are based are referenced. Supporting data for each recommendation is briefly summarized with evaluative comments and referenced.

Key Words

Rhesus, alloimmunization, fetus, anemia

Values: The evidence collected was reviewed by the Maternal-Fetal Medicine and Genetics Committees of the Society of Obstetricians and Gynaecologists of Canada (SOGC) and quantified using the Evaluation of Evidence guidelines developed by the Canadian Task Force on the Periodic Health Exam.

Recommendations:

1. Anti-D Ig 300 µg IM or IV should be given within 72 hours of delivery to a postpartum nonsensitized Rh-negative woman delivering an Rh-positive infant. Additional anti-D Ig may be required for fetomaternal hemorrhage (FMH) greater than 15 mL of fetal red blood cells (about 30 mL of fetal blood). Alternatively, anti-D Ig 120 µg IM or IV may be given within 72 hours of delivery, with testing and additional anti-D Ig given for FMH over 6 mL of fetal red blood cells (12 mL fetal blood). (I-A)
2. If anti-D is not given within 72 hours of delivery or other potentially sensitizing event, anti-D should be given as soon as the need is recognized, for up to 28 days after delivery or other potentially sensitizing event. (III-B)
3. There is poor evidence regarding inclusion or exclusion of routine testing for postpartum FMH, as the cost-benefit of such testing in Rh mothers at risk has not been determined. (III-C)

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4. Anti-D Ig 300 µg should be given routinely to all Rh-negative nonsensitized women at 28 weeks' gestation when fetal blood type is unknown or known to be Rh-positive. Alternatively, 2 doses of 100–120 µg may be given (120 µg being the lowest currently available dose in Canada): one at 28 weeks and one at 34 weeks. (I-A)
5. All pregnant women (D-negative or D-positive) should be typed and screened for alloantibodies with an indirect antiglobulin test at the first prenatal visit and again at 28 weeks. (III-C)
6. When paternity is certain, Rh testing of the baby's father may be offered to all Rh-negative pregnant women to eliminate unnecessary blood product administration. (III-C)
7. A woman with "weak D" (also known as D^u-positive) should not receive anti-D. (III-D)
8. A repeat antepartum dose of Rh immune globulin is generally not required at 40 weeks, provided that the antepartum injection was given *no earlier than 28 weeks' gestation*. (III-C)
9. After miscarriage or threatened abortion or induced abortion during the first 12 weeks of gestation, nonsensitized D-negative women should be given a minimum anti-D of 120 µg. After 12 weeks' gestation, they should be given 300 µg. (II-3B)
10. At abortion, blood type and antibody screen should be done unless results of blood type and antibody screen during the pregnancy are available, in which case antibody screening need not be repeated. (III-B)
11. Anti-D should be given to nonsensitized D-negative women following ectopic pregnancy. A minimum of 120 µg should be given before 12 weeks' gestation and 300 µg after 12 weeks' gestation. (III-B)
12. Anti-D should be given to nonsensitized D-negative women following molar pregnancy because of the possibility of partial mole. Anti-D may be withheld if the diagnosis of complete mole is certain. (III-B)
13. At amniocentesis, anti-D 300 µg should be given to nonsensitized D-negative women. (II-3B)
14. Anti-D should be given to nonsensitized D-negative women following chorionic villous sampling, at a minimum dose of 120 µg during the first 12 weeks' gestation, and at a dose of 300 µg after 12 weeks' gestation. (II-B)
15. Following cordocentesis, anti-D Ig 300 µg should be given to nonsensitized D-negative women. (II-3B)
16. Quantitative testing for FMH may be considered following events potentially associated with placental trauma and disruption of the fetomaternal interface (e.g., placental abruption, blunt trauma to the abdomen, cordocentesis, placenta previa with bleeding). There is a substantial risk of FMH over 30 mL with such events, especially with blunt trauma to the abdomen. (III-B)
17. Anti-D 120 µg or 300 µg is recommended in association with testing to quantitate FMH following conditions potentially associated with placental trauma and disruption of the fetomaternal interface (e.g., placental abruption, external cephalic version, blunt trauma to the abdomen, placenta previa with bleeding). If FMH is in excess of the amount covered by the dose given (6 mL or 15 mL fetal RBC), 10 µg additional anti-D should be given for every additional 0.5 mL fetal red blood cells. There is a risk of excess FMH, especially when there has been blunt trauma to the abdomen. (III-B)
18. Verbal or written informed consent must be obtained prior to administration of the blood product Rh immune globulin. (III-C)

Validation: These guidelines have been reviewed by the Maternal-Fetal Medicine Committee and the Genetics Committee, with input from the Rh Program of Nova Scotia. Final approval has been given by the Executive and Council of the Society of Obstetricians and Gynaecologists of Canada.

Sponsors: The Society of Obstetricians and Gynaecologists of Canada.

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INTRODUCTION

Anti-D immunoprophylaxis has made erythroblastosis fetalis caused by sensitization to the D-antigen a preventable disease, and perinatal deaths from alloimmunization have fallen 100-fold.¹ Prevention of Rh alloimmunization by immunoprophylaxis has been primarily responsible for the dramatic reduction in the incidence of the mortality from this disease, although changes in family size and the quality of perinatal care have also contributed.² Anti-D IgG has been licensed for routine postpartum prophylaxis since 1968 in Canada, and routine antepartum prophylaxis was introduced in 1976.³ Maternal alloimmunization still occurs in 0.4 per 1000 births^{4,5} or approximately 1% to 2% of D-negative women in Canada and the United Kingdom,^{5,6} usually from failure to administer anti-D immune globulin to eligible pregnant and postpartum women or because of inadequate dosing schedules.⁷

Supporting data for each recommendation is briefly summarized with evaluative comments and referenced (Table 1). The quality of evidence and classification of recommendations reported in these guidelines has been described using the Evaluation of Evidence criteria outlined in the Report of the Canadian Task Force on the Periodic Health Exam (Table 2).⁸

ANTI-D IMMUNE GLOBULIN

Anti-D immune globulin G is a blood product containing a high titre of antibody to Rh antigens of red blood cells. It is obtained from human plasma and is effective in the prevention of active rhesus alloimmunization.^{9,10} In Canada, the product is manufactured by Cangene Corporation in Winnipeg under the trade name "WinRho." Routes of administration for this product include intravenous (IV) or intramuscular (IM). The duration of action of either route is the same.⁵ After IV administration, the initial serum levels of anti-D are higher in the first week but similar thereafter until 3 months. Highly circulating levels of anti-D might be of benefit when the timing of FMH is known (e.g., postpartum, third trimester bleed), but are not relevant to antenatal prophylaxis at 28 weeks.⁵

Following administration of anti-D, a positive antibody screen will be found in the woman.¹¹ This response is typically of low titre and weakly reactive. Anti-D crosses the placenta and binds to fetal red blood cells, without causing hemolysis,

TABLE I SYNOPSIS OF QUALITY OF EVIDENCE REGARDING ANTI-D IMMUNOPROPHYLAXIS*		
Recommendations	Strength of Recommendations	Quality of Evidence
Postpartum Prophylaxis		
• Anti-D 120–300 µg within 72 hours of delivery	A	I
• Anti-D up to 28 days after delivery	B	III
• Routine FMH testing after delivery	C	Insufficient
Antepartum Prophylaxis		
• Anti-D 300 µg at 28 weeks	A	I
• Repeat antibody screening at 28 weeks	C	III
• Routine paternal testing	C	III
• Anti-D for “weak D” (e.g., D ^u)	D	III
• Repeat anti-D at 40 weeks	C	III
Early Pregnancy Loss and Termination		
• Anti-D 120–300 µg after spontaneous/induced abortion	B	II-3
• Antibody screening prior to anti-D after abortion	B	III
• Ectopic pregnancy: 120–300 µg Rh immune globulin	B	III
• Molar pregnancy: 120–300 µg Rh immune globulin	B	III
Invasive Fetal Procedures		
• Amniocentesis: 300 µg Rh immune globulin	B	II-3
• CVS: 120–300 µg Rh immune globulin	B	II
• Cordocentesis: 300 µg Rh immune globulin	B	II-3
APH, Abdominal Trauma, ECV, FMH		
• Quantitative FMH testing	B	III
• Anti-D 120–300 µg following placental trauma	B	III
Consent		
• Informed consent prior to administration of anti-D	C	III
*CVS: chorionic villous sampling; APH: antepartum hemorrhage; ECV: external cephalic version; FMH: fetomaternal hemorrhage.		

anemia, or jaundice.¹² In one study, 20% of the Rh-positive babies, born to mothers receiving 2 antepartum doses of anti-D immune globulin, had a positive direct antiglobulin test, but their hemoglobin and bilirubin levels were no different from those of Rh-negative babies.¹³

Canadian preparations of anti-D immune globulin have never been associated with blood-borne infections such as HIV or hepatitis B or C. Donors are strictly screened.¹⁴ All plasma units undergo quarantine and repeated testing to demonstrate that they are nonreactive to syphilis, hepatitis B surface antigen, anti-HCV, HIV-1 p 24 antigen, anti-HIV-1, and anti-HIV-2.¹⁴ Anion-exchange chromatography is used to extract pure IgG. Anti-D IgG is then filtered (35 nm virus filter) and subjected to solvent-detergent to inactivate possible residual viruses. Finally, the solvent-detergent mixture is removed by reverse-phase chromatography, to yield the current product version, WinRho SDE.¹⁵ Other (not WinRho) anti-D preparations have been associated with epidemics of hepatitis C in Ireland (1977 and early 1990s)¹⁶ and Germany.¹⁷ Transmission of HIV has not been reported.¹⁷

Reports of adverse drug reactions from administration of prophylactic anti-D to Rh-negative women are rare and usually mild, manifesting as local swelling, headache, or chills.¹⁵ The rare hypersensitivity reaction manifesting as urticaria, itching, or maculopapular rash may be treated with antiurticarial agents. Anaphylaxis occurs rarely, but warrants the availability of epinephrine when administering anti-D IgG. The Royal College of Obstetricians and Gynaecologists concluded that no serious adverse reactions have been reported in women receiving intramuscular anti-D IgG.¹

POSTPARTUM PROPHYLAXIS

If Rh-negative mothers do not receive postpartum anti-D IgG prophylaxis after an Rh-positive baby, the incidence of sensitization during the next pregnancy is 12% to 16%, compared to 1.6% to 1.9% in mothers receiving postpartum prophylaxis.^{3,18,19} A meta-analysis of postpartum anti-D prophylaxis was carried out by Crowther and Middleton,⁹ including 6 randomized trials involving over 10 000 women who received either postpartum

<p>TABLE 2</p> <p>QUALITY OF EVIDENCE ASSESSMENT⁸</p>	<p>CLASSIFICATION OF RECOMMENDATIONS⁸</p>
<p>The quality of evidence reported in these guidelines has been described using the Evaluation of Evidence criteria outlined in the Report of the Canadian Task Force on the Periodic Health Exam.</p> <p>I: Evidence obtained from at least one properly randomized controlled trial.</p> <p>II-1: Evidence from well-designed controlled trials without randomization.</p> <p>II-2: Evidence from well-designed cohort (prospective or retrospective) or case-control studies, preferably from more than one centre or research group.</p> <p>II-3: Evidence obtained from comparisons between times or places with or without the intervention. Dramatic results in uncontrolled experiments (such as the results of treatment with penicillin in the 1940s) could also be included in this category.</p> <p>III: Opinions of respected authorities, based on clinical experience, descriptive studies, or reports of expert committees.</p>	<p>Recommendations included in these guidelines have been adapted from the ranking method described in the Classification of Recommendations found in the Canadian Task Force on the Periodic Health Exam.</p> <p>A. There is good evidence to support the recommendation that the condition be specifically considered in a periodic health examination.</p> <p>B. There is fair evidence to support the recommendation that the condition be specifically considered in a periodic health examination.</p> <p>C. There is poor evidence regarding the inclusion or exclusion of the condition in a periodic health examination, but recommendations may be made on other grounds.</p> <p>D. There is fair evidence to support the recommendation that the condition not be considered in a periodic health examination.</p> <p>E. There is good evidence to support the recommendation that the condition be excluded from consideration in a periodic health examination.</p>

anti-D prophylaxis or no treatment. Anti-D administered within 72 hours of birth lowered alloimmunization to D antigen, detected at six months postpartum (relative risk [RR] 0.04) and during a subsequent pregnancy (RR 0.12). Higher doses (100–200 µg) of anti-D were more effective than lower doses (up to 50 µg) in preventing D alloimmunization in a subsequent pregnancy. No evidence was seen that 100 µg anti-D was substantially less effective than a higher dose, although the number of immunizations were few. Crowther and Middleton concluded that the cost-effectiveness of smaller doses of anti-D immune globulin, combined with screening for the degree of fetomaternal hemorrhage (FMH) and administering additional anti-D as necessary, should be compared with the use of larger doses of anti-D without laboratory testing for FMH.

TIMING OF POSTPARTUM ANTI-D ADMINISTRATION

It is recommended that anti-D be given within 72 hours of potential maternal exposure to fetal cells, because in the initial experiments, in which men were the subjects, anti-D was administered 72 hours after exposure,^{20,21} and clinical trials of postpartum prophylaxis specified that time limit.^{10,22} However, if this window is missed, there may still be some protection when anti-D is given up to 13 days²⁰ or even 28 days²³ after a potentially sensitizing event.

POSTPARTUM TESTING FOR FETOMATERNAL HEMORRHAGE

The need for and cost-effectiveness of testing for fetomaternal hemorrhage depends on the frequency of the volume of FMH

being in excess of the volume covered by the administered anti-D dose. The amount of anti-D (20 µg) needed to protect against 1 mL of D-positive red blood cells (about 2 mL fetal blood) was established by experiments in D-negative men injected with D-positive cells and anti-D IgG.¹⁰ Anti-D 300 µg protects against 30 mL fetal blood, and 120 µg protects against 12 mL. Sebring and Polesky²⁴ summarized studies in postpartum women of the volume of FMH measured by acid elution testing. Of 20 234 women, 99.67% had a volume of FMH less than 25–30 mL (range, 99.0%–99.67%). That is, about 3 per 1000 (range, up to 10 per 1000) D-negative women with D-positive babies would be inadequately protected with a 300 µg dose of anti-D, and about 1 per 1000 (up to 3 per 1000) would be alloimmunized as a result.²⁴⁻³⁰ The risk of alloimmunization is lower than the risk of the volume of FMH being over 30 mL, since not all women mount an immune response to exposure to D-positive blood, particularly when there is also ABO incompatibility between mother and baby. Bowman⁵ pointed out that alloimmunization due to massive FMH postpartum (about 0.07% of deliveries) is 20 times less common than third-trimester alloimmunization when antepartum anti-D is omitted (1.8% of Rh-negative pregnancies). Risk factors for FMH volume over 30 mL have been cited,³¹ but more than half of the cases of FMH over 30 mL occur in women without identified risk factors.²⁴⁻²⁶ Women undergoing Caesarean section may have a higher risk of large FMH (2.5% of 199 women studied),³² as well as women whose baby is stillborn, particularly without obvious cause (4.5% to 9.5%).^{24,33} Ness *et al.*²⁵

found that 5.6% of women had an FMH volume over 11 mL, so when a 120 µg dose of anti-D is used postpartum, FMH testing is likely indicated. The American Association of Blood Banks 1998 standard includes anti-D 300 µg and postpartum screening for FMH for all D-negative women with D-positive babies.³⁴ If a full dose of anti-D has been given within 21 days before delivery, there is no need to repeat it after birth if excess FMH has been excluded.¹¹

RECOMMENDATIONS

- 1. Anti-D Ig 300 µg IM or IV should be given within 72 hours of delivery to a postpartum nonsensitized Rh-negative woman delivering an Rh-positive infant. Additional anti-D Ig may be required for FMH greater than 15 mL of fetal red blood cells (about 30 mL of fetal blood). Alternatively, anti-D Ig 120 µg IM or IV may be given within 72 hours of delivery, with testing and additional anti-D Ig given for FMH over 6 mL of fetal red blood cells (12 mL fetal blood). (I-A)**
- 2. If anti-D is not given within 72 hours of delivery or other potentially sensitizing event, anti-D should be given as soon as the need is recognized, for up to 28 days after delivery or other potentially sensitizing event. (III-B)**
- 3. There is poor evidence regarding inclusion or exclusion of routine testing for postpartum FMH, as the cost-benefit of such testing in Rh mothers at risk has not been determined.^{34,35} (III-C)**

ANTEPARTUM PROPHYLAXIS

Without antenatal anti-D prophylaxis, 1.6% to 1.9% of Rh-negative women at risk become sensitized.³⁶ Routine antenatal prophylaxis reduces the rate of sensitization during pregnancy to 0.2%, as shown by at least 9 clinical studies of antenatal prophylaxis with anti-D immune globulin.³⁷ Antenatal prophylaxis at 28 to 29 weeks is recommended by the Canadian Task Force on Preventive Health Care,³⁸ American College of Obstetricians and Gynecologists,³³ and the US Preventive Services Task Force.³⁹ A meta-analysis of antenatal anti-D prophylaxis carried out by Crowther⁴⁰ included 2 trials. In the Hutchet trial, anti-D 100 µg at 28 and 34 weeks' gestation led to a clear reduction in immunization at 2 to 12 months after giving birth, in women who had received anti-D: alloimmunization was reduced from 4/360 to 0/363 (OR 0.13).⁴¹ Data were not given for the risk of Rh D alloimmunization in a subsequent pregnancy. This approach is the standard of care in the United Kingdom, adopted by the Royal College of Obstetricians and Gynaecologists.¹ It may achieve a higher circulating concentration of anti-D immune globulin as term approaches than does the single larger dose, but the cost of an extra injection may mitigate against this protocol and this is currently not the standard of care in Canada.³⁸ In Manitoba cohort studies (nonrandomized controls), a single 300 µg dose in first

pregnancies was associated with alloimmunization in 1.8% (62/3533) of controls and 0.18% (19/9609) in the treatment group.²² Bowman²² documented in prospective cohort studies that giving anti-D 300 µg at 28 and at 34 weeks (0.1% alloimmunization) did not confer significantly greater protection compared to one 300 µg dose at 28 weeks (0.2% alloimmunization [19/9609], half of whom were already alloimmunized by 28 weeks).²² Trolle¹² also found no alloimmunization with anti-D 300 µg, compared to 1.9% sensitized with no anti-D at 28 weeks. Tovey *et al.*⁶ compared anti-D 100 µg at 28 and 34 weeks to a cohort without antepartum prophylaxis, and found 0.2% (4/2069) treated and 1.4% (29/2000) untreated women became alloimmunized later. Robson *et al.*³⁷ comprehensively reviewed the important studies (not just randomized controlled trials) of antepartum prophylaxis and concluded in favour of routine administration of anti-D immunoglobulin at 28 and 34 weeks' gestation.

ANTENATAL ANTIBODY SCREENING

All women should have a blood type and antibody screen with an indirect antiglobulin test at the first prenatal visit, since 1.5% to 2% of pregnant women show atypical blood group sensitization.⁴² Opinions are divided on whether a repeat anti-D antibody screen at 28 weeks is indicated. The rationale for a repeat screen at 28 weeks is to identify the 0.18% or fewer women who have become alloimmunized since the first prenatal screen, in order to care for the fetus. It has been suggested that a second blood sample should be sent even from the Rh-positive woman at 27 to 28 weeks' gestation "to confirm that she is Rh-positive and that atypical blood group antibodies have not developed."⁴³ On the other hand, Barss *et al.*⁴⁴ pointed out that the cost of repeat irregular antibody screening in the third trimester exceeds US\$600 000 per perinatal death averted. Jackson and Branch³¹ recommend in Gabbe's text that "before administration of 300 µg of Rh-immune globulin at the beginning of the third trimester, it is probably unnecessary to obtain a second antibody screen to ensure that the patient is not already sensitized and actively producing anti-D. Similarly, a repeat antepartum antibody screen at 35 to 36 weeks' gestation is unwarranted."³¹ The American Society of Clinical Pathology's (ASCP's) practice parameters include "unexpected antibody" testing before antenatal anti-D is given, but omit repeat Rh testing if 2 documented test results are on the record.¹¹

PATERNAL TESTING

Rh testing of the baby's father may be offered to all Rh-negative pregnant women to eliminate unnecessary blood product administration. If the pregnant woman volunteers and confirms in private that her partner is indeed the biological father, and he is *documented* to be Rh-negative, then anti-D may be omitted.⁴⁵ Partners should not be routinely tested without this private confirmation of paternity. This may avoid creating a potential

conflict for the pregnant woman between privacy in the relationship and the well-being of the fetus.

MANAGEMENT OF "WEAK D"

A test for weak D phenotype (e.g., D^u) must be performed in women who initially test Rh-negative.⁴⁶ These women are genetically Rh-positive and are at low risk of producing anti-D antibodies and at very low risk of having an affected fetus.^{11,33,34} There is not universal agreement about this policy.⁴⁶⁻⁴⁹

In a survey including 3500 institutions, at least one patient with the weak D phenotype anti-D alloantibody formation was observed during a 12-month period by 31.8% of transfusion services.⁴⁸

REPEAT DOSING AT 40 WEEKS

Twelve weeks after injection of anti-D IV or IM, the mean residual circulating anti-D is 0.6 ng/mL to 1.0 ng/mL (representing 5 µg to 8 µg of anti-D), and some women have no residual anti-D. This is not enough to protect against a volume of FMH of greater than 1 mL.^{5,29} Bowman and Pollock⁵ noted that 3 of 9 failures of antenatal prophylaxis occurred in women delivering at least 13-5/7 weeks after the antenatal dose of anti-D. If the woman remained undelivered, they recommended a second dose of anti-D at 12-3/7 weeks after the previous antenatal dose, with no further postpartum dose unless transplacental hemorrhage was documented.⁵ Manitoba guidelines (1999) mandate a 39 to 40 week dose; ASCP practice parameters say it may be done.¹¹ There is insufficient evidence at this time to make a recommendation for or against administering another dose of anti-D to an unsensitized D-negative woman who remains undelivered at 40 weeks.

RECOMMENDATIONS

4. **Anti-D Ig 300 µg should be given routinely to all Rh-negative nonsensitized women at 28 weeks' gestation when fetal blood type is unknown or known to be Rh-positive. Alternatively, 2 doses of 100–120 µg may be given (120 µg being the lowest currently available dose in Canada): one at 28 weeks and one at 34 weeks. (I-A)**
5. **All pregnant women (D-negative or D-positive) should be typed and screened for alloantibodies, with an indirect antiglobulin test at the first prenatal visit and again at 28 weeks. (III-C)**
6. **Where paternity is certain, Rh testing of the baby's father may be offered to all Rh-negative pregnant women to eliminate unnecessary blood product administration. (III-C)**
7. **A woman with "weak D" (also known as D^u-positive) should *not* receive anti-D. (III-D)**
8. **A repeat antepartum dose of Rh immune globulin is generally not required at 40 weeks, provided that the antepartum injection was given *no earlier than 28 weeks' gestation.* (III-C)**

EARLY PREGNANCY LOSS OR TERMINATION, ECTOPIC PREGNANCY, HYDATIFORM MOLE

THREATENED AND INDUCED ABORTION

The D antigen is detectable on embryonic red blood cells by 38 days from conception, or 7-3/7 weeks' gestational age.⁵⁰ Fetal erythrocytes can be found in maternal circulation after spontaneous abortion in up to one-third of women at risk,⁵¹ and fetal red cells >0.05 mL can be detected in 26% of women,⁵¹ while the risk of alloimmunization following spontaneous abortion is 1.5% to 2%.³³ The risk of alloimmunization following induced abortion is 4% to 5%.³³ Even 0.1 mL of D-positive red blood cells can sensitize 3% of D-negative women,²⁷ so anti-D is indicated.

The total fetoplacental blood volume at 12-week pregnancy is 3 mL; that is, 1.5 mL fetal red cells.⁵² A 120 µg dose of anti-D would be protective.

For miscarriage or induced abortion beyond 12 weeks' gestation, anti-D 300 µg is indicated.^{11,39} The evidence in favour of anti-D prophylaxis for threatened abortion is weak. Von Stein⁵³ found that 11% of women with threatened abortion (not confirmed by ultrasound as to viability of embryo or fetus) had a positive acid elution test (over 0.07% acid-resistant cells) compared to 4% of controls of similar gestational age.

If an Rh-negative woman has had a negative anti-D antibody screen during this pregnancy, antibody screening need not be repeated before giving anti-D at abortion: only 1 of 9303 pregnant women followed by Bowman developed Rh immunization before 16 weeks' gestation, so almost no anti-D treatment would be avoided by repeat screening.⁵ If a blood type and antibody screen have not been done in this pregnancy, it should be done at the time of abortion.

ECTOPIC PREGNANCY

Alloimmunization has been reported after ectopic pregnancy.¹¹ Twenty-five percent of women with a ruptured tubal pregnancy have a significant number of fetal red blood cells in their circulation, suggesting that anti-D is indicated.¹¹

MOLAR PREGNANCY

Due to absent or incomplete vascularization of villi in complete hydatidiform mole, and the probable absence of D antigen on the villous trophoblast,⁵⁴ the risk of Rh alloimmunization in molar pregnancy is minimal. Anti-D Ig may be omitted when complete mole is diagnosed in nonsensitized Rh-negative mothers. Anti-D may not be omitted for partial mole or uncertain diagnosis.

RECOMMENDATIONS

9. **After miscarriage or threatened abortion or induced abortion during the first 12 weeks of gestation, nonsensitized D-negative women should be given a minimum anti-D of 120 µg. After 12 weeks' gestation, they should be given 300 µg. (II-3B)**

10. At abortion, blood type and antibody screen should be done, unless results of blood type and antibody screen during the pregnancy are available, in which case antibody screening need not be repeated. (III-B)
11. Anti-D should be given to nonsensitized D-negative women following ectopic pregnancy. A minimum of 120 µg should be given before 12 weeks' gestation and 300 µg after 12 weeks' gestation. (III-B)
12. Anti-D should be given to nonsensitized D-negative women following molar pregnancy because of the possibility of partial mole. Anti-D may be withheld if the diagnosis of complete mole is certain. (III-B)

INVASIVE FETAL DIAGNOSTIC PROCEDURES

AMNIOCENTESIS

Even with sonographic placental localization, a potentially immunizing volume of FMH (>0.1 mL) occurs in at least 2% of pregnancies undergoing amniocentesis,²³ and immunoprophylaxis with anti-D 300 µg is recommended. A small non-randomized trial found that 5.2% of women not treated with anti-D at amniocentesis became alloimmunized, whereas none of the treated women became sensitized following this invasive procedure.⁵⁵ If results of blood type and antibody screen done during pregnancy are available, antibody screening need not be repeated before giving anti-D at amniocentesis.⁵ Otherwise, blood type and antibody screen should be done.

CHORIONIC VILLOUS SAMPLING

As 14% of first-trimester sampling of chorionic villi results in FMH,⁵⁶ immunoprophylaxis is recommended, although estimates of the risk of subsequent alloimmunization are imprecise. Since the total fetoplacental blood volume is 3 mL at 12 weeks, anti-D 50 µg is sufficient at 12 weeks' gestation or less. For procedures carried out later in gestation, 300 µg of anti-D should be used. The minimum dose available commercially is 120 mg.

CORDOCENTESIS

Fetomaternal hemorrhage can occur following cordocentesis, particularly if a transplacental route is chosen. The prevalence of FMH following cordocentesis exceeds that following amniocentesis.⁵⁷

RECOMMENDATIONS

13. At amniocentesis, anti-D 300 µg should be given to nonsensitized D-negative women. (II-3B)
14. Anti-D should be given to nonsensitized D-negative women following chorionic villous sampling, at a minimum dose of 120 µg during the first 12 weeks' gestation, and at a dose of 300 µg after 12 weeks' gestation. (II-B)

15. Following cordocentesis, anti-D Ig 300 µg should be given to nonsensitized D-negative women. (II-3B)

ANTEPARTUM HEMORRHAGE, ABDOMINAL TRAUMA, EXTERNAL CEPHALIC VERSION, FETOMATERNAL HEMORRHAGE

Clinical conditions associated with potential placental trauma or disruption of the fetomaternal interface (e.g., placental abruption, external cephalic version, blunt trauma to the abdomen, placenta previa with bleeding) can lead to sensitizing FMH.²⁴ Fetomaternal hemorrhage has been identified in 1% to 6% of attempted or successful external cephalic version attempts.⁵⁸⁻⁶⁰ Blunt abdominal trauma in pregnancy has also been documented to cause large FMH.⁶¹ Since these conditions may be more likely to cause fetomaternal hemorrhage in excess of 30 mL, measurement of FMH volume is prudent.

RECOMMENDATIONS

16. Quantitative testing for FMH may be considered following events potentially associated with placental trauma and disruption of the fetomaternal interface (e.g., placental abruption, blunt trauma to the abdomen, cordocentesis, placenta previa with bleeding). There is a substantial risk of FMH over 30 mL with such events, especially with blunt trauma to the abdomen. (III-B)
17. Anti-D 120 µg or 300 µg is recommended in association with testing to quantitate FMH following conditions potentially associated with placental trauma and disruption of the fetomaternal interface (e.g., placental abruption, external cephalic version, blunt trauma to the abdomen, placenta previa with bleeding). If FMH is in excess of the amount covered by the dose given (6 or 15 mL fetal RBC), 10 µg additional anti-D should be given for every additional 0.5 mL fetal red blood cells. There is a risk of excess FMH, especially when there has been blunt trauma to the abdomen.⁶² (III-B)

CONSENT

Informed consent must be obtained prior to administration of any blood product. Verbally informing the woman about the source and safety of anti-D immunoglobulin is likely sufficient.⁶³ If the woman refuses anti-D, explain the potential consequences. (A patient information brochure is available from the manufacturer. Some centres keep a copy of this, signed by the patient, on the medical record to document that the information has been given, or alternatively obtain formal written consent.) Strategies may differ between centres in keeping with existing institutional policies governing consent to treat for transfusion or other blood products.

RECOMMENDATION

18. Verbal or written informed consent must be obtained prior to administration of the blood product Rh immune globulin. (III-C)

REFERENCES

1. Urbaniak S. The scientific basis of antenatal prophylaxis. *Br J Obstet Gynaecol* 1998;105:11–8.
2. Joseph K, Kramer M. The decline of Rh hemolytic disease: should Rh prophylaxis get all the credit? *Am J Public Health* 1998;88:209–15.
3. Bowman J, Pollock J. Antenatal prophylaxis of Rh isoimmunization: 28 weeks gestation service programme. *Can Med Assoc J* 1978;118:627–30.
4. Armson BA, Parson ML. The Rh program of Nova Scotia 1964–1994. Halifax: Atlantic Society of Obstetricians and Gynaecologists; 1995.
5. Bowman J, Pollock J. Failures of intravenous Rh immune globulin prophylaxis: an analysis for the reasons of such failures. *Transf Med Rev* 1987;1:101–12.
6. Tovey L, Townley A, Stevenson B, Tavener J. The Yorkshire antenatal anti-D immunoglobulin trial in primigravidae. *Lancet* 1983;2:244–6.
7. Urbaniak S. Consensus conference on anti-D prophylaxis, April 7 & 8, 1997: final consensus statement of the Royal College of Edinburgh/Royal College of Obstetricians and Gynaecologists. *Transfusion* 1998;38:97–9.
8. Woolf SH, Battista RN, Angerson GM, Logan AG, Eel W. Canadian Task Force on the Periodic Health Exam. Ottawa: Canada Communication Group; 1994. p. xxxvii.
9. Crowther C, Middleton P. Anti-D administration after childbirth for preventing Rhesus alloimmunization. *The Cochrane Library* (4). 2001. Oxford, Update Software.
10. Stern K, Goodman H, Berger M. Experimental iso-immunization to hemo-antigens in man. *J Immunol* 1961;87:189–98.
11. Hartwell E. Use of Rh immune globulin. *ASCP Practice Parameter*. *Am J Clin Pathol* 1998;110:281–302.
12. Trolle B. Prenatal Rh-immune globulin prophylaxis with 300 mcg immune globulin anti-D in the 28th week of pregnancy. *Acta Obstet Gynecol Scand* 1989;68:45–7.
13. Maayan-Metzger A, Schwartz T, Sulkes J, Merlob P. Maternal anti-D prophylaxis during pregnancy does not cause neonatal haemolysis. *Arch Dis Child Fetal Neonatal Ed* 2001;84:F60–2.
14. Hong F, Ruiz R, Price H, Griffiths A, Malinoski F, Holoski M. Safety profile of WINRHRO Anti-D. *Sem Hematol* 1998;35:9–13.
15. Canadian Pharmacists Association. WinRho SDF™ Rh (D) immune globulin (human) passive immunizing agent. In: *Compendium of pharmaceuticals and specialties (CPS)*. 36th edition. Toronto: Canadian Pharmacists Association; 2001. p. 1714–6.
16. Murdoch A. Irish mothers called for hepatitis C virus screening. *Br J Med* 1994;308:613–4.
17. Power J, Lawlor E, Davidson F, Yap P, Kenny-Walsh E, Whelton M, et al. Hepatitis C viraemia in recipients of Irish intravenous anti-D immunoglobulin. *Lancet* 1994;344:1166–7.
18. Bowman J, Chown B, Lewis M, Pollock J. Rh isoimmunization during pregnancy: antenatal prophylaxis. *Can Med Assoc J* 1978;118:623–30.
19. Bowman J. Preventing Rh sensitization during pregnancy. *Perinat Care* 1978;2:24–32.
20. Samson D, Mollison P. Effect on primary Rh immunization of delayed administration anti-Rh. *Immunology* 1975;28:349–57.
21. Gorman J, Freda V, Pollack W. Prevention of sensitization to Rh with anti-Rh gamma globulin. *Fred Proc* 1964;23:238.
22. Bowman J. Antenatal suppression of Rh alloimmunization. *Clin Obstet Gynecol* 1991;34:296–303.
23. Bowman J, Pollock J. Transplacental fetal haemorrhage after amniocentesis. *Obstet Gynecol* 1985;66:749–54.
24. Sebring E, Polesky H. Fetomaternal hemorrhage: incidence, risk factors, time of occurrence and clinical effects. *Transfusion* 1990;30:344–57.
25. Ness P, Baldwin M, Niebyl J. Clinical high-risk designation does not predict excess fetal-maternal hemorrhage. *Am J Obstet Gynecol* 1987;156:154–8.
26. Stedman C, Baudin J, White C, Cooper E. Use of the erythrocyte rosette test to screen for excessive fetomaternal hemorrhage in Rh negative women. *Am J Obstet Gynecol* 1986;154:1363–9.
27. Zipursky A, Israels L. The pathogenesis and prevention of Rh immunization. *Can Med Assoc J* 1967;97:1245–57.
28. Zipursky A. Rh hemolytic disease of the newborn – the disease eradicated by immunology. *Clin Obstet Gynecol* 1977;20:759–72.
29. Haemolytic disease of the fetus and newborn due to anti-Rh D (Rh D haemolytic disease). In: Mollison P, Engelfriet C, Contreras M, editors. Boston: Blackwell Scientific Publications; 1993. p. 546–80.
30. Bowman J. The prevention of Rh immunization. *Trans Med Review* 1988;2:129–50.
31. Jackson J, Branch D. Isoimmunization in pregnancy. In: Gabbe S, Niebyl J, Simpson J, editors. New York: WB Saunders; 1996. p. 899–932.
32. Feldman N, Skoll A, Sibai B. The incidence of significant fetomaternal haemorrhage in patients undergoing cesarean section. *Am J Obstet Gynecol* 1990;163:855–8.
33. American College of Obstetricians and Gynecologists. Prevention of Rh D isoimmunization. *ACOG Practice Bulletin*. 1999.
34. Snyder EL, Shoos Lipton K. Prevention of hemolytic disease of the newborn due to anti-D. In: *Bulletin #98-2*. Bethesda (MD): American Association of Blood Banks; February 16, 1998. p. 1–6.
35. Vengelen-Tyler V. Technical manual. Bethesda (MD): American Association of Blood Banks; 1996. p. 12.
36. Hadley A, Kumpel B. The role of Rh antibodies in haemolytic disease of the newborn. *Baillieres Clin Haematol* 1993;6:423–44.
37. Robson S, Lee D, Urbaniak S. Anti-D immunoglobulin in RhD prophylaxis. *Br J Obstet Gynaecol* 1998;105:129–34.
38. Beaulieu M-D. Screening for D (Rh) sensitization in pregnancy. *Canadian Guide to Clinical Preventive Health Care*; 1994. p. 116–24.
39. Diguiseppe C. Screening for D (Rh) incompatibility. *US Preventive Services Task Force. Guide to Clinical Preventive Services* (2). 1996.
40. Crowther C. Anti-D administration in pregnancy for preventing Rhesus alloimmunization. *The Cochrane Library* (4). 2001. Oxford, Update Software.
41. Hutchet J, Dallemagne S, Hutchet C, Brossard Y, Larsen M, Parnet-Mathieu F. The antepartum use of anti-D immunoglobulin in rhesus negative women. Parallel evaluation of a multicentre study carried out in the region of Paris. *J Gynecol Obstet Biol Reprod (Paris)* 1987;16:101–11.
42. Weinstein L. Irregular antibodies causing hemolytic disease of the newborn: a continuing problem. *Clin Obstet Gynecol* 1982;25:321–32.
43. Bowman J. Hemolytic disease (erythroblastosis fetalis). In: Creasy R, Resnik R, editors. Philadelphia: WB Saunders Co.; 1999. p. 736–68.
44. Barss V, Frigoletto F, Konugres A. The cost of irregular antibody screening. *Am J Obstet Gynecol* 1988;159:428.
45. Mitchell R, Bowell P, Letsky E, de Silva M, Whittle M. Guidelines for blood grouping and red cell antibody testing during pregnancy. *Transfusion Med* 1996;6:71–4.
46. Mayne K, Allen D, Bowell P. “Partial D” women with anti-D alloimmunization in pregnancy. *Clin Lab Haematol* 1991;13:239–44.
47. Konugres A, Polesky H, Walker R. Rh immune globulin and the Rh-positive, D^u variant mother. *Transfusion* 1982;22:76–7.
48. Domen R. Policies and procedures related to weak D phenotype testing and Rh immune globulin administration. Results from supplementary questions to the Comprehensive Transfusion Medicine Survey of the College of American Pathologists. *Arch Pathol Lab Med* 2000;124:1118–21.
49. Gorman J. New applications of Rh immune globulin: effect on protocols. In: Frigoletto F, Jewett J, Konugres A, editors. Boston: GH Hall Medical Publishers; 1981. p. 199–210.
50. Bergstrom H, Nilsson L, Nilsson L, Ryttinger L. Demonstration of Rh antigens in a 38-day-old fetus. *Am J Obstet Gynecol* 1967;99:130–3.
51. Litwak O, Taswell H, Banner E. Fetal erythrocytes in maternal circulation after spontaneous abortion. *J Am Med Assoc* 1970;214:531–4.

-
52. De Crespigny L, Davison G. Anti-D administration in early pregnancy: time for a new protocol. *Aust N Z J Obstet Gynecol* 1995;35:385-7.
 53. Von Stein G, Munsick R, Stiver K, Ryder K. Fetomaternal haemorrhage in threatened abortion. *Obstet Gynecol* 1992;79:383-6.
 54. van't Veer MB, Overbeeke MA, Geertzen HG, van der Lans SM. The expression of Rh-D factor in human trophoblast. *Am J Obstet Gynecol* 1984;150:1008-10.
 55. An assessment of the hazards of amniocentesis. Report to the Medical Research Council by their Working Party on Amniocentesis. *Br J Obstet Gynaecol* 1978;85:1-41.
 56. Brambati B, Guercilena S, Bonacchi I, Oldrini A, Lanzani A, Piceni L. Feto-maternal transfusion after chorionic villus sampling: clinical implications. *Hum Reprod* 1986;1:37-40.
 57. Bowman JM, Pollock JM, Peterson LE, Harman CR, Manning FA, Menticoglou SM. Fetomaternal haemorrhage following funipuncture: increase in severity of maternal red-cell alloimmunization. *Obstet Gynecol* 1994;84:839-43.
 58. Marcus R, Crewe-Brown H, Krawitz S, Katz J. Fetomaternal hemorrhage following successful and unsuccessful attempts at external cephalic version. *Br J Obstet Gynaecol* 1975;82:578-80.
 59. Nord E, Blaschke E, Green K, Thomassen P. 100 cases of external cephalic version with special reference to fetomaternal transfusion. *Acta Obstet Gynecol Scand* 1989;68:55-8.
 60. Lau T, Stock A, Rogers M. Fetomaternal hemorrhage after external cephalic version at term. *Aust N Z J Obstet Gynecol* 1995;35:173-4.
 61. Pearlman M, Tintinalli J, Lorenz R. Blunt trauma during pregnancy. *N Engl J Med* 1990;323:1609-13.
 62. Pollack W, Ascari W, Crispen J, O'Connor R, Ho T. Studies on Rh prophylaxis: II. *Transfusion* 1971;11:340-4.
 63. Manitoba Rh Program. Manitoba guidelines for prenatal antibody screening: indications for administration of Rh immune globulin [newsletter]. May 1999. *Coll Physicians Surg Manitoba* 1999;35(3):1.