Detection of Anti-D and Anti-G in a Pregnant Woman: A Case Report

Gebe Kadında Anti-D ve Anti-G Saptanması: Bir Olgu Sunumu

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ABSTRACT

Antibody identification in Rh-negative pregnant women is usually done to detect RhD alloimmunisation. The G antigen is part of the Rh blood group and is ubiquitous on most D-positive red cells. The detection of anti-G however is complicated. The objective of this case report is to highlight the importance of identifying anti-G correctly especially in managing antenatal patients. We herein report a case of a 30-year-old pregnant woman, who was thought to have anti-D and anti-C on initial antibody identification, was subsequently found out to have anti-G and anti-D on further testing.

Keywords: Pregnancy, anti-D, anti-G, haemolytic disease of fetus and newborn.

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ÖZET

Rh negatif hamile kadınlarda antikor tanımlaması genellikle RhD alloimmünizasyonunu saptamak için yapılır. G antijeni Rh kan grubunun bir parçası ve çoğu D pozitif kırmızı hücrede her yerde bulunur. Ancak anti-G’nin tespiti karmaşıktır. Bu vaka raporunun amacı, özellikle antenatal hastaların yönetiminde anti G’nin doğru tanılanmasını önemini vurgulamaktır. Burada, ilk antikor tanımlamasında anti-D ve anti-C olduğu düşünülen, daha sonra ileri testlerde anti-G ve anti-D olduğu saptanan 30 yaşında bir hamile kadın vakasını sunuyoruz.

Anahtar Sözcükler: Gebelik, anti-D, anti-G, fetüs ve yeni doğanın hemolitik hastalığı.

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INTRODUCTION

The G antigen is present on most D-positive cells and all C-positive red blood cells. (1) They are absent on red blood cells that are D and C-negative. The G antigen is encoded by RHD and by the C allele of the RHCE gene hence anti-G often emerge as similar serologically to anti-C and anti-D.

Identifying anti-G is not important in routine transfusion practice, however, it is vital in transfusion management of pregnant women as anti-G can cause haemolytic disease of fetus and newborn (HDFN). It is also important in decision regarding anti-D administration in Rh negative women and for medicolegal purposes, for example, in paternity determination. (2) It is of less importance in routine transfusion practice. We report a case of anti-D and anti-G in a pregnant woman.

CASE REPORT

A 30-year-old gravida 5, Para 3+1 female was grouped as AB with rr (dce/dce) phenotype. Her husband’s blood group was O with R1R1 (CDe/CDe) phenotype. During the first two pregnancies, the antibody identifications showed anti-D with titers ≥1:214. Direct Coombs test (DCT) was negative. During her first pregnancy, an extra reaction was detected only at enzyme phase, however, this was not investigated further.

During the second pregnancy, the antibody identification was performed only at LISS phase as there was insufficient sample. She suffered a miscarriage during her third pregnancy. Along the course of her pregnancies, she was never given anti-D immunoglobulin, as it was assumed that she was alloimmunised. All her newborns had mild neonatal jaundice.

Antibody screening (3-cell panels, Bio-Rad ID-DiaCell I-II-III Asia, gel technique) was positive (4+) in all cells. Antibody identification (11 cell panel, Bio-Rad ID DiaPanel-P, gel technique) revealed specificity towards anti-D and anti-C hence anti-G is suspected.

Double absorption and elution

Further testing by double adsorption and elution were performed using O positive R2R2 (cDe/cDe) and O negative r'r (dce/dCe) cells. The double adsorption and elution technique was performed via tube technique.

The patient’s serum was adsorbed with R2R2 cells to adsorb anti-D and anti-G and leave anti-C in the post adsorbed serum. The first post adsorbed serum was checked with the following known cells: R2R2 (D+G+C-), r'r (G+D+C+) and rr (D-G-C-) to check for the presence of anti-C, whilst the first eluate from the adsorbed R2R2 cells was also similarly tested.

Subsequently, the previous eluate was adsorbed with r'r cells to adsorb any anti-G if present and leave unbound anti-D in post adsorbed serum. The r'r cells were then eluted and tested in the same manner to confirm the presence of anti-G. This multistep process is simplified in Figure 1 and the results are summarised in Table 1.

Figure 1. Multistep process

| Step | Description |
|------|-------------|
| 1. | Patient’s serum: First adsorption with R2R2 cells (D+G+C-) |
| 2. | Post adsorbed R2R2 cells |
| 3. | Eluate from post adsorbed R2R2 cells (Post adsorbed eluate) |
| 4. | Check with known cells to confirm presence of unbound anti-C (M) |
| 5. | Second adsorption with r'r (D-G+C+) |
| 6. | Post adsorbed r'r cells |
| 7. | Check with known cells to confirm presence of anti-D (O) |
| 8. | Post adsorbed serum. Check with known cells to confirm presence of anti-G (P) |
| 9. | Eluate from post adsorbed r'r cells |
| 10. | Check with known cells to confirm presence of anti-G (P) |
The result showed that this patient had anti-G and anti-D. The strength of the anti-G reaction is lower (+1) than anti-D reaction (+4), as the anti-G titer is much lower. The combined antibodies (anti-D and anti-G) titer is 1:152. The patient gave birth to a healthy baby boy at 38 weeks of pregnancy, who had mild neonatal jaundice.

DISCUSSION
Anti-G identification is important for the provision of anti-D immunoglobulin and predicting outcome of fetus. It is known that both anti-D and anti-C can cause HDFN. The role of anti-G in HDFN is unclear as the titer of anti-G rarely reaches as high titer as anti-D and anti-C.(3) However, infants of mothers with high anti-G titer can develop moderate or severe HDFN.(4) Women with anti-G+C must be given anti-D to prevent HDFN due to anti-D alloimmunisation.(2) Mothers who had dual antibody specificity (such as anti-D + anti-C) can have infants with moderate to severe HDFN, whilst mothers with anti-G alone, can also develop severe HDFN needing exchange transfusion.(5) In our case study, the patient developed anti-D with low titre of anti-G and her infant only developed mild jaundice that needed phototherapy.

Identification of anti-G can assist in resolving medico-legal issues. For example, it can provide explanation regarding the development of anti-D in Rh (D) negative patients who were given D-negative products. This is because anti-G and anti-C can be found in the serum of recipients of Rh (D) negative products, which mimic anti-D. Also, it can aid in paternity issues, for example in infant with HDFN due to apparent anti-D when both parents are Rh (D) negative. The antibody implicated could be anti-G or anti-G+C. This will exclude anti-D in Rh (D) negative women whom partners are Rh (D) negative as well.(3)

CONCLUSION
This case demonstrates the difficulties in eliciting anti-G. In our setting, the process took approximately six to seven hours. The availability of donor cells (R2R2 and r’r) is also limited. The titer of anti G can fluctuate or be very low that it might be lost during the washing process. Also, antibody identification needs to be done at both LISS and enzyme phase to not miss the presence of anti-C and/or anti-G.

Conflict of interest
No conflict of interest was declared by the authors.

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| Sample | R2R2(D+G+ C-) | r’r (D- G+ C+) | rr (D- G- C-) | Antibody Interpretation |
|--------|---------------|--------------|--------------|------------------------|
| M      | 0             | 0            | 0            | No anti-C Present      |
| N      | 4+            | 1+           | 0            | Either: Anti-G present OR anti-G +anti-D present |
| O      | 4+            | 0            | 0            | Presence of anti-D     |
| P      | 4+            | 1+           | 0            | Presence of anti-G and anti-D |

Table 1. The results of the multistep process.