All that Reacts with A1 Cells are not Anti A1: A Case Report on Red Cell Alloimmunisation in a Subgroup

Mili S¹, Meena D², Shaiji PS³

ABSTRACT

Introduction: When an A2/A2B individual has an antibody reacting only with A1 cells on reverse grouping, it is considered as anti-A1 antibody. However presence of an irregular antibody reacting with A1 reagent red cells, in an A2/A2B individual can mislead the diagnosis as A2/A2B with Anti A1

Case Report: Here we report a case of an A2B individual with anti E antibody which reacted with the pooled A1 reagent red cells, which led to a misdiagnosis.

Conclusion: Antibody screening must be done in addition to routine tests to confirm an anti A1 antibody to confirm the specificity

Keywords: A2B, Anti A1, Anti E, Placenta Praevia

INTRODUCTION

Apart from four main blood groups enlisted in ABO system namely A, B, AB and O by Landsteiner, there are subtypes of A and B antigen which are genetically and phenotypically distinct. Approximately, 20% of individuals with A antigen, belong to A2 and other weaker subgroups.¹ In India frequency of A2 is 0.8%– 3.0% while frequency A2B is 0.6 – 1.4%.² A1 and A2 antigens are distinguished with anti-A1 lectin (Dolichos biflorus). About 1 - 8% of A2 and 22 - 26% of A2B individuals possess anti-A1 in their serum, which occurs as a cold agglutinin and exclusively agglutinates A1 cells. The higher percentage of anti A1 in A2B is attributed to *R101 allele which is present in 41% of A2B individuals as against 1% in A2 group.³ These antibodies are considered clinically significant if they react at 37°C with A1 cells.

Prevalence of A2B in South India is approximately 10.50% with presence of anti-A1 in 3.75% of A2B individuals.⁴ Most of them agglutinate A1 cells only up to 25°C & are not significant. However, they can interfere in routine blood grouping and give incorrect typing results. When A2 individual has antibody reacting only with A1 cells on reverse grouping, it is considered as anti-A1 antibody and transfusion management planned accordingly. Here we encountered such a case which later turned out to be an irregular antibody reacting with reagent red cells. Proceeding without repeat testing and confirmation would have led to adverse effects in this case.

CASE REPORT

37yr old, G3P1L1A1, a case of Type IV Placenta Praevia at gestational age 37 wks 5days was referred to our centre, as the peripheral centre was unable to find a compatible unit with any of the three AB positive donors available. Emergency LSCS was warranted in view of patient’s clinical condition. Historical Group was AB Positive. Husband’s blood group was B positive. Request for cross matching 2 units was placed to the blood bank. Clinical and transfusion history derived no significant finding and no history of transfusions in the past. 1st pregnancy was 10 yrs back. 1st baby is AB positive. 2nd pregnancy was 8 yrs back which was terminated in first trimester due to personal reasons.

Blood Grouping was done using conventional tube technique for cell and serum grouping using Anti A, Anti B, Anti AB and Anti D antisera manufactured by Tulip diagnostics as well as 5% cell suspensions of pooled A1, B and O cells. There was an evident discrepancy between forward and reverse grouping. Sub grouping was done with anti-A1 lectin (Dolichos biflorus). The results are in Table 1. Patient seemed to be a case of A2B group with suspected anti-A1 in the serum causing agglutination of pooled A1 red cells. 2 units of B positive cells were selected for compatibility test with this patient.

But when blood grouping was repeated in a second sample, with a different set of pooled cells, reactivity had changed contrary to the expected pattern. This time, forward grouping was A2B while reverse grouping was A i.e. there was a 3 + reaction with B cells. Since the discrepancy was not resolved and the patient was taken for Emergency LSCS, 2 units of group O negative RBC were cross matched using Coombs gel technology & was found to be compatible. Daily QC reports of antisera and Reagent Red cells were verified. Clerical errors were ruled out.

A third set of EDTA and plain sample was obtained after LSCS for complete work up. Repeat Blood Grouping was done with a new sample and different pooled cells. Patient Serum showed repeated negative reaction with Pooled A1 cells, hence presence of Anti A1 was ruled out. Auto control at all temperatures were negative, thus auto

¹Assistant Insurance Medical Officer, ESI(CD)Mulangunnathukavu, Killannur, Kerala, ²Professor, Department of Transfusion Medicine, Government Medical College,Trivandum, Kerala, ³Assistant Professor, Department of Transfusion Medicine, Government Medical College, Trivandum, Kerala, India

Corresponding author: Shaiji PS, Assistant Professor, Department of Transfusion Medicine, Government Medical College, Trivandum, India

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Antibody was ruled out. Indirect antiglobulin test (with pooled O cells) were negative both in Tube Technique and Column Agglutination Technology using commercially available Matrix gel cards.

Testing of Saliva showed that patient is a Secretor with the presence of A and B Antigens in Saliva. Possibility of an alloantibody which reacts with the corresponding antigen which is randomly present on reagent red cells was strongly considered now. Antibody screening using 3 cell panel (Diacell, BioRad)) turned out to be Positive. Hence we proceeded with Antibody Identification with 11 cell panel (DiaPanel, BioRad) which showed the presence of Anti E reacting at the Immediate Spin phase and AHG phase (Figure 1).

On Antigen Typing – Patient was found to be E Negative and Husband was E and e Positive. Cross matching with Husband’s RBCs showed 3+ agglutination. Titration was done with Husband’s RBCs which yielded a titre of 16. Baby developed jaundice at postnatal 5th day. S. Bilirubin was 14g% which improved later with phototherapy. Baby’s Forward Grouping A2B Rh Positive. Direct antiglobulin test was Positive with 4+ grade.

**DISCUSSION**

Anti E antibody is an irregular antibody which is usually reactive at warm temperatures (37°C) and do not agglutinate saline-suspended RBCs carrying the corresponding antigen unless they have a major IgM component. Thus they usually do not create blood group discrepancies. But there are reports that anti E antibody which is naturally occurring reactive at room temperature.5,6 Here such an unusual case of anti E has lead to an ABO discrepancy. To add to the complexity, affected patient had an A2B blood group also, confusing the case as an A2B with anti A1.

Antibodies with anti-E specificity are detected in 14–20% of pregnant women. Anti-E can occur either as a natural IgM antibody without immune stimulation or as an IgG antibody in women with a history of blood transfusion or previous pregnancy. Van Loghem and Hart consider immunizations to the antigens E alone during pregnancy to be very exceptional.7 Malone and Dunsford in 1951 had reported a case of
multiparous female with placenta praevia found to have A2 blood group with saline reacting anti E. Huestis and Bates reported a similar case when an incompatible cross match drew attention to a previously unsuspected sensitization in a pregnant A group Rh-positive woman with an Rh-positive husband. Large studies showing association of subgroups of A antigen with placenta praevia or other cases of obstetric haemorrhage have not been extensively studied.

CONCLUSION

Certain points emphasised by this case are as follows:
• Performing antibody screening tests before or instead of a cross-match permits early recognition and identification of clinically significant antibodies and makes the decision about immediate-spin cross-match easier. It also decreases workload, reduces reagent costs, enables timely recognition of significant antibodies & helps in more effective use of blood inventory. In this case, if the policy was type and screen instead of cross match, the discrepancy could have been solved easily and quickly without unnecessary testing being performed
• Antibody Screening should be preferred over ICT using pooled cells because it yields better results
• From a clinician’s point of view, Antenatal antibody screening should be done in all pregnant women irrespective of the Rh (D) antigen status to detect red cell alloimmunization to other clinically significant blood group antigens also. This is essential for transfusion safety in mother and early management of HDFN
• Performing blood grouping at least twice before first transfusion- At the time of admission & before transfusion
• When serum of an individual contains anti A1, reactive at 37°C, further sub grouping for A2 or A2B group is required to select the appropriate blood for transfusion. But confirmation of the specificity by cell panels are also important.
• Routine antigen typing, at least for Rh antigens, to provide antigen negative blood is highly recommended

REFERENCES


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