Access this article online

Quick Response Code:

Website: www.ajts.org

DOI: 10.4103/ajts.ajts_132_20

Red cell antibodies resulting in false-positive complement-dependent cytotoxicity cross-match: A unique case

Rajesh B. Sawant, Sharad Sheth[1], Pooja Mehta[2], Deepali Naker[2]

Abstract:

A false-positive complement-dependent cytotoxicity cross-match (CDC XM) has a negative impact in donor selection process obliterating healthy, donor compatible population. A 47-year-old male with chronic kidney disease was planned for ABO-compatible renal transplantation from his sister. CDC and donor-specific antibody (DSA) lysate XM were negative 10 days before transplant. The pretransplant CDC XM showed 40% positivity. DSA lysate XM and HLA antibody screen were negative. Patient's Indirect antiglobulin test (IAT) was positive and anti-M antibody (IgG + IgM) was identified. Therapeutic plasma exchange, intravenous immunoglobulin, and rituximab were used for desensitization. Decrease in positivity of CDC XM and anti-M titer was seen. The transplant was performed successfully. Red cell alloantibody should be considered in differential diagnosis of a positive CDC XM. The utility of DSA lysate XM as a pretransplant monitoring tool is immense in such situations. Institutional policies regarding plan of action in the event of positive CDC XM and negative DSA lysate XM and vice versa should be formed.

Keywords:

Complement-dependent cytotoxicity, cross-match, HLA, positive, renal transplant

Department of Transfusion Medicine and Histocompatibility and Immunogenetics, [1]Nephrology and [2]Histocomptibility and Immunogenetics, Kokilaben Dhirubhai Ambani Hospital and Medical Research Institute, Mumbai, Maharashtra, India

Address for correspondence:
Dr. Rajesh B. Sawant, Department of Transfusion Medicine and Histocompatibility and Immunogenetics, Kokilaben Dhirubhai Ambani Hospital and Medical Research Institute, Andheri West, Mumbai-400053, Maharashtra, India.
E-mail: rbshil@gmail.com

Submitted: 29-08-2020
Revised: 10-09-2020
Accepted: 14-03-2021
Published: 12-06-2021

Introduction

Antibody-mediated rejection in cases of kidney transplantation is associated with the presence of preformed donor-specific antibodies (DSAs). Hence, recipients are screened before transplantation. The conventional complement-dependent cytotoxicity cross-match (CDC XM) is performed to confirm compatibility between the donor and recipient.

The presence of antilymphocyte antibodies in the recipients' serum (HLA and non-HLA antibodies) targeting donor antigens results in positive CDC XM. Previous sensitization events in recipients such as pregnancy, blood transfusion, and previous transplant can result in a positive CDC XM. To avoid rejection, an unexpected positive CDC XM must be immediately reported in a prospective setting.

We report here an unexpected positive CDC XM due to the presence of a RBC alloantibody with anti-M specificity.

Case Report

A 47-year-old male patient with chronic kidney disease due to acute pyelonephritis was considered for kidney transplantation with his sister as a prospective donor. The patient had a history of Packed red blood cells (PRBC) transfusion (day: −38). The blood group of the patient and donor was O Rh (D) positive. The low-resolution HLA

How to cite this article: Sawant RB, Sheth S, Mehta P, Naker D. Red cell antibodies resulting in false-positive complement-dependent cytotoxicity cross-match: A unique case. Asian J Transfus Sci 2021;15:94-6.

typing was performed by polymerase chain reaction SSO method revealing a 5/6 mismatch [Table 1].

The initial CDC XM and the DSA lysate XM performed during the pretransplant work-up were negative; hence, the patient was posted for transplant. Since the initial CDC XM was negative, auto-CDC XM and dithiothreitol (DTT)-modified CDC XM were not performed. As per our institutional protocol, CDC XM is done 48 h before transplant. The pretransplant routine CDC XM (day: −14) showed 40% positivity. To confirm the result, DTT-modified CDC XM assay was performed which also showed 40% positivity. This indicated the presence of IgG antibodies in the recipient's serum; hence, the transplant was put on hold. The recipient's sample was sent for blood grouping, antibody screening, and blood reservation. The alloantibody screening revealed the presence of anti-M alloantibody with a titer of 256 and having both IgM (RT) and IgG (37°C) components [Table 2].

The patient was managed with therapeutic plasma exchange (TPE) followed by intravenous immunoglobulin treatment as a desensitization protocol. TPE (1.2× plasma volume) was performed using a continuous flow cell separator (day: −10). Following the treatment on day −8, DSA lysate XM and DTT CDC XM were performed. We found that DSA lysate XM was negative for both HLA Class I (MFI-544) and II (MFI-227) and DTT CDC XM showed 15% positivity, indicating the persistence of IgG antibodies. He was further managed with rituximab and second sitting of TPE (day: −8). This treatment resulted in the reduction of anti-M titer to 8 on day −7 and negative DTT CDC XM on day −3.

The transplant was performed; posttransplant day +3, DSA monitoring was done which was found to be negative. The patient accepted the graft well. Further follow-up was possible till 10 months posttransplant and the graft was functioning well at that time.

Table 1: HLA typing results of donor and recipient on Luminex 100/200 platform

HLA alleles	Donor		Recipient	
HLA A	A*03	A*68	A*02	A*26
HLA B	B*44	B*51	B*08	B*15
HLA DRB1	DRB1*03	DRB1*15	DRB1*03	DRB1*14

Table 2: Patient management plan summary

Investigations	Days	Results		
CDC XM	−27	Negative		
CDC XM	−14	Routine CDC-40% positive		
		DTT modified-40% positive		
Alloantibody screening, identification, and titer	−13	Anti-M antibody detected, total (IgG+IgM) titer=256		
Therapeutic plasma exchange performed and IVIG administered				
CDC XM	−8	Routine CDC-15% positive		
		DTT treated-15% positive		
DSA XM	−8	Class I	Class II	Result
		544	227	Negative
Therapeutic plasma exchange performed and rituximab administered				
Antiglobulin titer	−7	Total (IgG+IgM) titer=8		
CDC XM	−3	Negative		
Day: 0 – transplant done				
DSA XM	+3	Class I	Class II	Result
		248	403	Negative

DTT=Dithiothreitol, IVIG=Intravenous immunoglobulin, CDC=Complement-dependent cytotoxicity, CDC XM=CDC cross-match

Discussion

The conventional CDC XM technique was established as the prototype of cross-matching in the late sixties and has unquestionably improved the outcome of transplantations since hyperacute and acute rejections have been reduced.[1,2] Positive CDC XM can be observed in recipient with an autoimmune disease or preexisting antibodies not detected by single-antigen bead assay (SAB) due to complement interference or due to previous desensitization protocols.[3] Although false-positive CDC XM due to autoimmune diseases has been mentioned in the literature, it lacks concrete examples which demonstrate different possibilities or reasons for CDC XM positivity (non-HLA antibodies).[4]

Whenever a CDC XM is positive, we follow an algorithm; where CDC XM is repeated with freshly drawn samples, CDC XM with and without DTT is done to confirm the positivity. The results are correlated clinically and with the results of DSA lysate XM. If the discrepancy is not resolved, it is further referred for SAB testing and flow cross-match. Since the discrepancy was resolved on red cell alloantibody screening, it was not tested further by SAB and flow XM.

In the present study, CDC XM showed 40% positivity before transplant; even with DTT treatment, the cross-match was positive, due to which transplant was postponed. Initially, the antibody levels were reduced by desensitization protocol, but positivity still persisted. To confirm the positivity, DSA lysate XM was performed which was negative for both Class I (MFI-544) and II (MFI-227), indicating false or doubtful positivity in CDC results. Simultaneously, the patient's antiglobulin test revealed the presence of anti-M alloantibodies which was reactive for both IgG (37°C) and IgM (RT) type.

In this case, alloantibody may have formed after transfusion of the PRBC unit on day −38. The antibody screening was performed before transfusion which was negative then.

Anti-M antibody is commonly IgM type, but there are case reports available where it has been reported to occur with both IgM and IgG components, as was seen in this case.[5] When such alloantibody exists in recipient's serum and is of IgG subtype, it is likely to be detected in CDC XM as well (like any other IgG type antibody, e.g., rituximab). In India, the prevalence of alloantibodies has been reported in range of 3.4%–9.8%.[6,7] This case also emphasizes that although the addition of DTT and Anti human globulin (AHG) reagent has improved the efficiency of CDC XM with time, still the probability of counterfactual results persists with this technique. Tests like DSA lysate XM, Red Cell antibody screening and titre, HLA antibody screening are suggested as a prognostic tool in kidney transplantation. This will reduce our dependence on the result of single gold standard test that is CDC XM. Thus, this case highlights that multidisciplinary approach and healthy communication between the transplant and laboratory services is essential for the proper management of transplant cases.

Conclusion

Red cell alloantibody should be considered in differential diagnosis of a positive CDC XM. The utility of DSA lysate XM as a pretransplant monitoring tool is immense in such situations.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

References

1. Kissmeyer-Nielsen F, Kjerbye KE. Lymphocytotoxic microtechnique. Purification of lymphocytes by flotation. In: Curtoni ES, Mattiuz PL, Tosi RM, editors. Histocompatibility Testing. Copenhagen: Munksgaard; 1967. p. 381-3.
2. Patel R, Terasaki PI. Significance of the positive crossmatch test in kidney transplantation. N Engl J Med 1969;280:735-9.
3. Desoutter J, Apithy MJ, Bartczak S, Guillaume N. False positive B-cells crossmatch after prior rituximab exposure of the kidney donor. Case Rep Transplant 2016;2016:1:Article Id-4534898.
4. Schlaf G, Pollok-Kopp B, Schabel E, Altermann W. Artificially positive crossmatches not leading to the refusal of kidney donations due to the usage of adequate diagnostic tools. Case Rep Transpl 2013;2013:4:746395.
5. Shah SP, Kalgutkar SM, Sawant RB, Deshpande AS. Anti-M antibodies: Biphasic (reactive at room temperature and at 37°C): A case series. Asian J Transfus Sci 2016;10:159-60.
6. Thakral B, Saluja K, Sharma RR, Marwaha N. Red cell alloimmunization in a transfused patient population: A study from a tertiary care hospital in north India. Hematology 2008;13:313-8.
7. Shukla JS, Chaudhary RK. Red cell alloimmunization in multi-transfused chronic renal failure patients undergoing hemodialysis. Indian J Pathol Microbiol 1999;42:299-302.