4-8-2022

Lack of alloimmunization to the D antigen in D-negative orthotopic liver transplant recipients receiving D-positive red blood cells perioperatively

Anjanaa Vijayanarayanan

Lindsey Wlosinski

Jaber El-Bashir

Dragos Galusca

Shunji Nagai

See next page for additional authors

Authors

Anjanaa Vijayanarayanan, Lindsey Wlosinski, Jaber El-Bashir, Dragos Galusca, Shunji Nagai, Atsushi Yoshida, Marwan S. Abouljoud, and Zaher K. Otrock

Received: 3 January 2022 | Revised: 9 March 2022 | Accepted: 27 March 2022

DOI: 10.1111/vox.13282

ORIGINAL ARTICLE

Vox Sanguinis ISBT International Society of Blood Transfusion

Lack of alloimmunization to the D antigen in D-negative orthotopic liver transplant recipients receiving D-positive red blood cells perioperatively

Anjanaa Vijayanarayanan[1] | Lindsey Wlosinski[1] | Jaber El-Bashir[2] | Dragos Galusca[2] | Shunji Nagai[3] | Atsushi Yoshida[3] | Marwan S. Abouljoud[3] | Zaher K. Otrock[1]

[1]Department of Pathology and Laboratory Medicine, Henry Ford Hospital, Detroit, Michigan, USA

[2]Department of Anesthesiology, Pain Management and Perioperative Medicine, Henry Ford Hospital, Detroit, Michigan, USA

[3]Transplant and Hepatobiliary Surgery, Henry Ford Hospital, Detroit, Michigan, USA

Correspondence
Zaher K. Otrock, Department of Pathology, Wayne State University School of Medicine, Transfusion Medicine, Henry Ford Hospital, Department of Pathology—K6, 2799 West Grand Blvd, Detroit, MI 48202, USA.
Email: zotrock1@hfhs.org; zaherotrock@hotmail.com

Funding information
None.

Abstract

Background and Objectives: D-negative patients undergoing orthotopic liver transplantation (OLT) might require a large number of red blood cell (RBC) units, which can impact the inventory of D-negative blood. The blood bank might need to supply these patients with D-positive RBCs because of inventory constraints. This study evaluates the prevalence of anti-D formation in D-negative OLT patients who received D-positive RBCs perioperatively, as this will assist in successful patient blood management.

Materials and Methods: This was a retrospective study performed at a single academic medical centre. Electronic medical records for all 1052 consecutive patients who underwent OLT from January 2007 through December 2017 were reviewed. D-negative patients who were transfused perioperatively with D-positive RBCs and had antibody screening at least 30 days after transfusion were included.

Results: Of a total of 155 D-negative patients, 23 (14.8%) received D-positive RBCs perioperatively. Seventeen patients were included in the study. The median age was 54 years (range 36–67 years); 13 (76.5%) were male. The median number of D-positive RBC units transfused perioperatively was 7 (range 1–66 units). There was no evidence of D alloimmunization in any patient after a median serologic follow-up of 49.5 months (range 31 days to 127.7 months). The average number of antibody screening post OLT was 7.29.

Conclusion: Our study showed that transfusion of D-positive RBCs in D-negative OLT recipients is a safe and acceptable practice in the setting of immunosuppression. This practice allows the conservation of D-negative RBC inventory.

KEYWORDS
alloimmunization, blood management, D antigen, liver transplant, RBC transfusion

Highlights

- Owing to increased transfusion demand and inventory constraints, the blood bank might be forced to transfuse D-positive red blood cells (RBCs) to D-negative patients undergoing liver transplant.

This work was presented, in part, at the American Association of Blood Banks Annual Meeting, 13–16 October 2018.

- Our study showed that transfusion of D-positive RBCs to D-negative liver transplant recipients is a safe and acceptable practice perioperatively; none of the studied patients had alloimmunization to the D antigen.
- The practice of transfusing D-positive RBCs to D-negative liver transplant recipients allows the conservation of D-negative RBC units for other patients in greater need of this resource.

INTRODUCTION

Liver transplantation has significantly improved the outcomes and life expectancy of patients with end-stage liver disease. Transplant surgeries in general and liver transplants specifically have been associated with significant blood loss and often require intensive transfusion support with blood products [1]. Advances in surgical techniques and anaesthetic management have contributed to an overall reduction in the transfusion needs during and following liver transplantation. Despite blood management, liver transplantation is still associated with high blood transfusion requirements perioperatively because of abnormal haemostasis and bleeding complications [2, 3].

D-negative patients are routinely transfused with D-negative red blood cells (RBCs) because of the high immunogenicity of the D antigen. The rate of alloimmunization to the D antigen in healthy volunteers following transfusion of D-positive packed RBCs can be as high as 80% [4]. However, immunosuppressed antigen-negative patients (such as organ transplant recipients and oncology patients) are much less likely to produce an alloantibody when exposed to a foreign RBC antigen [5–7].

When D-negative patients undergo liver transplant surgeries, it may not be possible for the blood bank to provide the required number of D-negative units because of inventory constraints; thus, the blood bank might transfuse these patients with D-positive RBCs. Though the process of providing D-positive RBCs to D-negative transplant recipients is accepted, the incidence of alloimmunization to the D antigen in D-negative liver transplant recipients has not been well defined. With a very active liver transplant programme at our institution, studying the prevalence of anti-D formation in D-negative liver transplant recipients receiving D-positive RBCs perioperatively will assist in successful patient blood management.

MATERIALS AND METHODS

This was a retrospective study performed at a single large academic medical centre. The study was approved by our Institutional Review Board, and informed consent was waived because of the study design. Electronic medical records and blood bank files for all 1052 consecutive patients who underwent orthotopic liver transplantation (OLT) with or without other organ transplants at Henry Ford Hospital in Detroit, Michigan, from January 2007 through December 2017 were reviewed. Patients were identified through the Liver Transplant Institute database. The transfusion records of D-negative OLT patients were reviewed for perioperative RBC transfusions. The perioperative period was defined as the period spanning 1 week before OLT until 2 weeks following OLT. Patients who were transfused perioperatively with D-positive RBCs and had antibody screening at least 30 days after the first D-positive RBC transfusion were included in the analysis.

Data collected for each patient included demographic details, clinical history, perioperative RBC transfusion and follow-up antibody screening. Blood bank records were last reviewed in December 2021 for updated antibody screening.

Blood bank testing

ABO and RhD group testing were performed using the following immunohaematology analysers utilizing solid-phase technology: Galileo (available till 2012), Galileo NEO (2012 to February 2020) and NEO Iris (since March 2020). Manual tube testing was used to resolve discrepancies in ABO forward and reverse grouping. ABO blood group testing was performed using monoclonal anti-A, anti-B and anti-D antisera for forward typing (Immucor, Norcross, GA) and pooled A1 and B cells for reverse typing (Immucor). Antibody screening was performed by indirect antiglobulin test (IAT) using a two-cell screen on the same analyser. Positive samples were investigated for antibody identification with a 14-cell panel. All tests were performed according to manufacturers' instructions.

Immunosuppression and blood management

The detailed immunosuppression protocol has been described previously [2]. Briefly, induction immunosuppression included rabbit antithymocyte globulin or basiliximab. The maintenance immunosuppression regimen consisted of tacrolimus, mycophenolate mofetil and steroids.

As soon as the blood bank is notified of a potential liver transplant, the recipient's transfusion records are reviewed. If the current and previous antibody screening are negative for clinically significant alloantibodies to RBC antigens, 20 ABO-compatible RBC units will be allocated for the recipient. If clinically significant alloantibodies were identified historically or on the current antibody screening, 30 antigen-negative serologically cross-matched RBC units were allocated. Ideally, blood would be stocked 24 h in advance of the day of transplant; however, blood bank is notified 6–12 h before most scheduled liver transplants. Our blood bank utilized leukoreduced RBCs collected by our blood supplier; we would start issuing irradiated units on the

day of transplant. The ABO group of each RBC unit and the RhD type of units labelled as D-negative were confirmed by the Henry Ford Hospital blood bank before going into inventory. The decision to transfuse D-positive RBC units to D-negative patients was based on both demand and inventory of D-negative RBC units in the blood bank.

Intraoperative blood product transfusion was managed by the anaesthesiologist; transfusion was dictated by the patient's haemodynamic status, intraoperative course, blood loss and oozing from the surgical field, and laboratory testing, mostly haemoglobin, platelet count, prothrombin time and activated partial thromboplastin time. Cell salvage and antifibrinolytics were utilized, as needed. The criteria for pre- and post-operative blood transfusion were referenced from current AABB clinical practice guidelines [8–10].

RESULTS

A total of 1052 patients underwent 1099 OLT during the study period. The number of transfused D-positive and D-negative patients was 809/897 (90.1%) and 140/155 (90.3%), respectively. Of the total 155 D-negative patients, 23 (14.8%) received D-positive RBCs perioperatively. Two patients did not survive surgery, and four patients did not have serologic follow-up after 30 days following transfusion and thus were excluded. The characteristics of the included 17 patients are presented in Table 1. The median age was 54 years (range 36–67 years); 13 (76.5%) were male. Twelve patients were Caucasian. The median number of RBC units transfused perioperatively was 19 (range 2–77 units), while the median number of D-positive RBC units transfused was 7 (range 1–66 units). None of the patients received Rh immune globulin prophylaxis. At the time of OLT, none of the patients had clinically significant RBC alloantibodies through antibody screening or a history of alloimmunization to RBC antigens. There was no evidence of D alloimmunization in any patient after a median serologic follow-up of 49.5 months (range 31 days to 127.7 months). The average number of antibody screening post OLT was 7.29.

We had 14 D-negative patients (14/155 = 9.0% of all D-negative liver transplant recipients) who had D alloimmunization before transplant; none of these patients was transfused with D-positive blood at our institution before alloimmunization.

DISCUSSION

In this study, we reviewed the medical records of OLT patients to determine the prevalence of anti-D formation in D-negative liver transplant recipients receiving D-positive RBCs. During the perioperative period, 17 eligible patients were transfused with a median of 7 D-positive RBC units (range 1–66 units). After a median serologic follow-up of 49.5 months, none of the patients developed anti-D antibodies.

OLT has developed throughout the years and become the standard of care for advanced liver disease. Although the use of blood components during OLT has considerably decreased over the past two decades, transfusion demands are still significant, especially

TABLE 1 Characteristics of the 17 D-negative liver transplant recipients who received D-positive red blood cells (RBCs) perioperatively

Patient	Age (years)/gender	Ethnicity	Pretransplant diagnosis	No. of D-positive RBCs	No. of D– RBCs	Serological F/U[a]
1	51/female	AA	HCV cirrhosis	2	0	21.23 months
2	44/male	AA	HCV and alcoholic cirrhosis	3	19	79.43 months
3	36/male	AA	Alcoholic cirrhosis	1	18	31.07 months
4	64/male	White	Autoimmune hepatitis	9	2	8.17 months
5	67/female	White	Liver cirrhosis secondary to NASH	4	0	127.70 months
6	53/male	White	Alcoholic cirrhosis	30	2	58.87 months
7	48/male	White	HCV and alcoholic cirrhosis	4	20	53 days
8	56/male	White	Metastatic neuroendocrine tumour	9	24	113.13 months
9	67/male	White	Alcoholic cirrhosis	25	8	95.23 months
10	59/male	White	Alcoholic cirrhosis	32	4	80.30 months
11	61/female	AA	HCV cirrhosis with HCC	4	4	83.67 months
12	53/female	White	Liver cirrhosis secondary to NASH	10	7	14.20 months
13	63/male	White	HCV cirrhosis with HCC	5	10	54.10 months
14	63/male	White	Polycystic liver disease	17	16	5.07 months
15	52/male	AA	Liver cirrhosis secondary to NASH	7	9	49.47 months
16	48/male	White	Alcoholic cirrhosis	3	5	4.13 months
17	54/male	White	Alcoholic cirrhosis	66	11	31 days

Abbreviations: AA, African American; HCC, hepatocellular carcinoma; HCV, hepatitis C virus; NASH, non-alcoholic steatohepatitis.
[a]Time from first D-positive RBC transfusion to last antibody screening.

during the perioperative period [2]. There is also wide variation in transfusion rates between transplantation centres, which can be attributed to specific patient population, surgical techniques, differences in institutional transfusion practices, variable use of antifibrinolytics and intraoperative blood salvage, among other variables [11].

Ensuring an adequate blood supply can be challenging because of the increased demand, increased number of donor deferrals, and sometimes decreased supply due to weather conditions and pandemics. Inventory concerns become more pronounced with D-negative units, which comprise only 10%–15% of donors. The Rhesus (Rh) blood group is the second most clinically important system after the ABO system. The D antigen is one of the Rh blood group antigens; it is highly immunogenic in healthy subjects as well as in immunocompromised patients [12, 13]. Knowing the high immunogenicity of the D antigen and the risks of delayed haemolytic transfusion reactions, D-negative patients are not usually transfused with D-positive blood. However, blood banks might be forced to transfuse D-negative patients with D-positive blood to preserve their D-negative RBC units, which are a scarce resource. Although it is not a common practice to transfuse D-negative males and females beyond the childbearing age with D-positive blood, this is considered an acceptable approach when the inventory of D-negative blood is small; however, the risks of delayed serologic/haemolytic transfusion reactions should be discussed with the treating physicians.

An adequate perioperative supply of D-negative blood for D-negative patients undergoing OLT is not always possible. We had a total of 23 D-negative OLT patients who were transfused with D-positive blood during the study period, 17 of whom were included in the analysis. All these patients were either males or females who were beyond the childbearing age. The decision to transfuse these patients with D-positive blood was dictated by the increased transfusion needs of these patients (refer to Table 1 for transfusion needs) and the sub-optimal inventory levels of D-negative blood. This cohort of OLT patients was transfused with a wide range of D-positive RBC units among the total number of units transfused.

There is a multitude of factors that could have contributed to the decision to transfuse D-positive RBC units and to this variation in transfused units. First, this study was not limited to evaluating intraoperative blood transfusions in OLT. We, instead, reviewed blood transfusions perioperatively, for a period spanning 1 week before OLT until 2 weeks following OLT. Transfusion needs post OLT surgery can persist because of many factors including coagulopathy, which can extend for months following transplantation [14]. In addition, up to 11% of OLT patients would require re-operation for bleeding within 2 weeks following OLT, which adds to their transfusion needs [15]. Second, although OLT is mostly well planned, some cases are relatively urgent with short notification to the blood bank, thus limiting our preparedness to provide D-negative blood. Evaluating the clinical situation of D-negative OLT candidates is crucial in making the decision to accept these patients for surgery. The decision to switch them to D-positive RBCs is necessary when the D-negative inventory is limited. Third, the demand for large numbers of RBC units may arise with traumas, massive transfusions and emergency surgeries, along with OLT cases complicated with intraoperative bleeding. In these situations, the decision to switch D-negative OLT patients to D-positive RBCs in order to save D-negative RBCs for patients with known anti-D, females of childbearing age and patients with no blood type on file would be appropriate.

Ramsey et al. evaluated 19 D-negative liver, heart and heart-lung transplant recipients who were transfused with D-positive RBCs [16]. Anti-D was detected in three liver transplant recipients; however, anti-D antibodies were detected at 3, 11 and 15 days following transfusion, which was not convincing of a primary immune response. In another study by Burin des Roziers et al., 20 D-negative OLT recipients transfused with D-positive RBCs perioperatively were evaluated [17]. None of the patients developed anti-D antibodies using the IAT. Two patients showed weak and transient anti-D reacting only with papain-treated RBCs at 10 and 11 days without any signs of haemolysis. Other investigators also have supported the safety of transfusing D-positive RBCs in D-negative patients during OLT surgery [5, 6, 18]. Table 2 summarizes the studies that evaluated alloimmunization to the D antigen in D-negative OLT recipients who

TABLE 2 Summary of studies evaluating the rate of alloimmunization to the D antigen in D-negative orthotopic liver transplantation recipients transfused with D-positive red blood cells (RBCs)

Study (year published)	Type of transplant	No. of patients	No. of D-positive units transfused; median (range)	F/U period; median (range), months	Number of patients that developed anti-D
Tiwari et al. [18]	Liver	21	7 (2–20)	11 (6–90)	None
Burin des Roziers et al. [17]	Liver	20	7 (1–40)	30 (9–120)	2 patients at 10 and 11 days after transfusion, transient
Casanueva et al. [5]	Liver	17	19 (5–41)	15 (2–70)	None
Ramsey et al. [16]	Liver, heart, heart-lung	19	10 (3–153)	2.5–51; median NA	3 patients at 3, 11 and 15 days after transfusion
Yuan et al. [6]	Liver	15	9 (2–39)	3.6 (1.7–13.5)	None
Current study	Liver	17	7 (1–66)	49.5 (1–128)	None

Abbreviation: NA, not available.

were transfused with D-positive blood. The lack of alloimmunization in D-negative OLT patients receiving D-positive blood can be explained by the fact that patients are maintained on immunosuppressive protocols to prevent the risk of graft rejection. None of the patients in our study developed anti-D antibodies on last follow-up.

Transfusion of D-positive RBCs in D-negative patients is unavoidable in surgeries such as OLT, which put an enormous pressure on limited blood inventory. Transfusing D-positive units to these patients is essential to prevent postponing these lifesaving procedures. This also helps in managing inventory and allocating D-negative units for other patients in whom the risk of alloimmunization would be higher, for example, D-negative females of childbearing age. In conclusion, transfusion of D-positive blood to D-negative OLT recipients is acceptable and safe in the setting of immunosuppression due to the low risk of alloimmunization.

One of our study limitations relate to its retrospective nature and being dependent on medical record documentation. The study was done at a single tertiary-care centre; thus, the results might not be generalizable to other institutions. In addition, the lack of systematic screening of OLT patients might have contributed to the relatively small sample size and decreased serologic follow-up period of some patients.

In conclusion, this study showed that transfusion of D-positive RBCs in D-negative OLT recipients is a safe and acceptable practice perioperatively in the setting of immunosuppression; none of the patients had alloimmunization to the D antigen following transfusion of D-positive RBCs. This practice allows the conservation of D-negative RBC units for other patients in greater need of this resource.

ACKNOWLEDGEMENTS

Z.O. contributed to the study design; A.V., L.W. and Z.O. contributed to the acquisition of data; A.V., L.W. and Z.O. contributed to the drafting the paper or revising it critically; A.V., L.W., J.E., D.G., S.N., A.Y., M.A. and Z.O. contributed to the approval of the submitted and final version.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ORCID

Zaher K. Otrock https://orcid.org/0000-0002-5823-2694

REFERENCES

1. Jadlowiec CC, Taner T. Liver transplantation: current status and challenges. World J Gastroenterol. 2016;22:4438–45.
2. Uzuni A, El-Bashir J, Galusca D, Yeddula S, Nagai S, Yoshida A, et al. Transfusion requirements and alloimmunization to red blood cell antigens in orthotopic liver transplantation. Vox Sang. 2021;117:408-14.
3. Yokoyama APH, Kutner JM, Sakashita AM, Nakazawa CY, de Paula TAO, Zamper RPC, et al. Risk factors for transfusion after orthotopic liver transplantation. Transfus Med Hemother. 2019;46:431–9.
4. Gunson HH, Stratton F, Cooper DG, Rawlinson VI. Primary immunization of Rh-negative volunteers. Br Med J. 1970;1:593–5.
5. Casanueva M, Valdes MD, Ribera MC. Lack of alloimmunization to D antigen in D-negative immunosuppressed liver transplant recipients. Transfusion. 1994;34:570–2.
6. Yuan S, Davis R, Lu Q, Goldfinger D, Ziman AF. Low risk of alloimmunization to the D antigen in D− orthotopic liver transplant recipients receiving D+ RBCs perioperatively. Transfusion. 2008;48:2653–5.
7. Arora K, Kelley J, Sui D, Ning J, Martinez F, Lichtiger B, et al. Cancer type predicts alloimmunization following RhD-incompatible RBC transfusions. Transfusion. 2017;57:952–8.
8. Carless PA, Henry DA, Carson JL, Hebert PP, McClelland B, Ker K. Transfusion thresholds and other strategies for guiding allogeneic red blood cell transfusion. Cochrane Database Syst Rev. 2010;CD002042.
9. Carson JL, Grossman BJ, Kleinman S, Tinmouth AT, Marques MB, Fung MK, et al. Clinical Transfusion Medicine Committee of the AABB. Red blood cell transfusion: a clinical practice guideline from the AABB. Ann Intern Med. 2012;157:49–58.
10. Carson JL, Guyatt G, Heddle NM, Grossman BJ, Cohn CS, Fung MK, et al. Clinical practice guidelines from the AABB: red blood cell transfusion thresholds and storage. JAMA. 2016;316:2025–35.
11. Ozier Y, Tsou M-Y. Changing trends in transfusion practice in liver transplantation. Curr Opin Organ Transplant. 2008;13:304–9.
12. Frohn C, Dümbgen L, Brand JM, Görg S, Luhm J, Kirchner H. Probability of anti-D development in D− patients receiving D+ RBCs. Transfusion. 2003;43:893–8.
13. Yazer MH, Triulzi DJ. Detection of anti-D in D− recipients transfused with D+ red blood cells. Transfusion. 2007;47:2197–201.
14. Stravitz RT. Thrombosis and coagulopathy in the liver transplant candidate and recipient. Clin Liver Dis. 2017;9:11–7.
15. Thompson MA, Redden DT, Glueckert L, Smith AB, Crawford JH, Jones KA, et al. Risk factors associated with reoperation for bleeding following liver transplantation. HPB Surg. 2014;2014:816246.
16. Ramsey G, Hahn LF, Cornell FW, Boczkowski DJ, Staschak S, Clark R, et al. Low rate of Rhesus immunization from Rh-incompatible blood transfusions during liver and heart transplant surgery. Transplantation. 1989;47:993–5.
17. Burin des Roziers N, Ibanez C, Samuel D, Francoz C, Idri S, François A, et al. Rare and transient anti-D antibody response in D− liver transplant recipients transfused with D+ red blood cells. Vox Sang. 2016;111:107–10.
18. Tiwari AK, Arora D, Aggarwal G, Dara RC, Bhardwaj G, Sharma J, et al. Safety assessment of RhD-positive red cell transfusion in RhD-negative liver-transplant recipients: single-centre report from India. Indian J Med Res. 2020;152:662–6.

How to cite this article: Vijayanarayanan A, Wlosinski L, El-Bashir J, Galusca D, Nagai S, Yoshida A, et al. Lack of alloimmunization to the D antigen in D-negative orthotopic liver transplant recipients receiving D-positive red blood cells perioperatively. Vox Sang. 2022.