

Approach to Kidney Transplant in Sensitized Potential Transplant Recipients

Antoine Barbari, Souodod Abbas, Mahassen Jaafar

Abstract

More than one-third of patients on waiting lists for kidney transplant are sensitized. Most have previously formed donor-specific and non-donor-specific serum antibodies and/or positive crossmatch by complement-dependent cytotoxicity and/or flow cytometry. Two categories of alloantibodies include antibodies against major histocompatibility complex human leukocyte antigen class 1 and class 2 and antibodies against minor histocompatibility complex. A current positive crossmatch is an absolute contraindication for transplant. Positive historical panel reactive antibody and/or donor-specific antibodies (human leukocyte antigen and minor histocompatibility complex), even in the absence of a historical positive crossmatch, are associated with an increased risk for allosensitization, antibody-mediated rejection, and accelerated graft failure. Desensitization protocols are numerous, complex, and expensive. It is recommended to perform a systematic determination of historical and current panel reactive antibody, donor-specific antibodies (human leukocyte antigen and minor histocompatibility complex), and crossmatch by the most sensitive assays. The risk of sensitization may be estimated from the combined results of the crossmatch with the donor and those of the recipient's panel reactive antibody and donor-specific antibodies at baseline. The adoption of a scoring

system for risk stratification may facilitate the task of organ allocation for sensitized patients. Recipients with an estimated sensitization risk $\geq$ high may be referred preferably to the national waiting priority list and informed about the financial and the medical risks that may incur with future transplant. Sensitized patients at high risk for antibody-mediated rejection may benefit from a structured monitoring process involving systematic and regular immunologic, histologic, and functional assessments of the graft after transplant. We recommend the adoption and regular updating of these approaches to ensure safe and appropriate therapeutic standards in these sensitized patients, in accordance with best clinical practice.

Key words: Renal, Alloimmunity, Human leukocyte antigen, Antibody

Introduction

A systematic search for donor-specific antibodies is performed before any solid-organ transplant. These donor-specific antibodies are categorized into 2 major groups: antibodies against the major histocompatibility complex (MHC) human leukocyte antigen (HLA) and antibodies against the non-HLA minor histocompatibility complex mHC. The MHC HLA antibodies involve anti-HLA class 1 (A, B, and C) and class 2 (DP, DQ, and DR) antibodies. The non-HLA antibodies are directed against antigens that are expressed on endothelial cells, epithelial cells, keratinocytes, dendritic cells, and monocytes, but not on peripheral blood lymphocytes.¹ They include antibodies against MHC class 1 chain-related protein A (MICA) and MHC class 1 chain related protein B (MICB) which are encoded by genes within the MHC and are genetically linked to HLA-B.² They also include anti-platelet, anti-endothelial cell, anti-

From the Renal Transplantation Unit, Rafik Hariri University Hospital, Bir Hassan, Beirut, Lebanon

Acknowledgements: The authors declare that they have no conflicts of interest to declare. Dr Abbas has wrote the Introduction and the section on anti-HLA antibodies detection techniques; Miss Jaafar collected all relevant articles and created 2 Tables (Nos. 1 and 3) in addition to collecting the data she needed to make the cost calculation in Table 3; and Dr Barbari wrote the remainder of the manuscript and created Table 2 and all Figures.

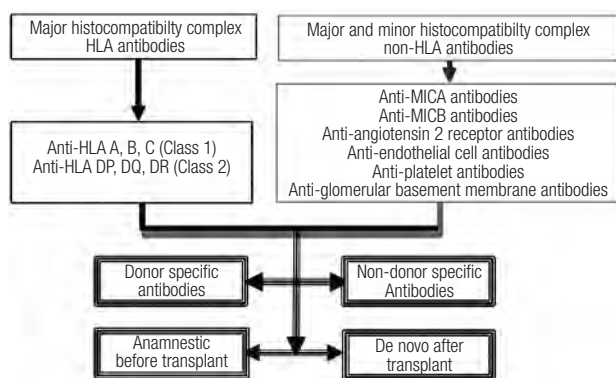
Corresponding author: Antoine Barbari, Renal Transplantation Unit, Rafik Hariri University Hospital, Bir Hassan, Beirut, Lebanon

Phone: +961 1 832040 **Fax:** +961 1 832041 **E-mail:** barbariantoine@gmail.com

Experimental and Clinical Transplantation (2012) 5: 419-427

angiotensin 2 receptor, and anti-basal membrane antibodies.³ These antibodies could be made in the recipient before transplant in an anamnestic fashion after a sensitization process related to a previous exposure to foreign antigen during blood transfusion, pregnancy, or previous organ transplant, or to an autoantigen in an autoimmune disease. Alloantibodies also could occur *de novo* after transplant as a result of allosensitization induced by the grafting process (Figure 1).

Figure 1. Alloantibodies in Solid-Organ Transplant



These antibodies are directed against human leukocyte antigen (HLA) or non-HLA antigens and may be donor-specific or non-donor-specific. They may be detected in the recipient before transplant or develop *de novo* after transplant. **Abbreviations:** MICA, major histocompatibility complex class 1 alpha chain-related protein A MICA; MICB, major histocompatibility complex class 1 alpha chain-related protein B

Currently, 35% patients on the transplant waiting list in the United States are sensitized with panel reactive antibody (PRA) levels > 0%, and 15% patients are highly sensitized with PRA levels > 80%.⁴ Approximately 33% of transplant recipients have donor-specific antibodies, with approximately 75% anti-HLA and 25% anti-MICA antibodies.⁵ The presence of HLA antibodies before transplant, or the *de novo* occurrence of HLA antibodies after transplant, is a major risk factor for antibody-mediated rejection (ABMR) and for major graft dysfunction with subsequent accelerated graft loss.⁴ Recent studies have demonstrated an association between the strength of donor-specific antibodies and risk of developing ABMR, response to treatment of ABMR, allograft survival, and success of desensitization protocols.^{1, 4, 6, 7}

Several recent reports have described the role of MICA and other non-HLA antibodies in the pathogenesis of ABMR in renal and cardiac transplants.⁷⁻¹⁰ Several factors affect the likelihood of developing ABMR and subsequent early graft failure. These factors include the presence and peak strength of

class 1 and class 2 donor-specific antibodies before transplant, previously formed MICA antibodies, positive historical crossmatch (either isolated or associated with any of the anti-HLA antibodies), and anti-non-HLA donor-specific antibodies.^{4, 7, 9, 11-13} The presence of these antibodies (anti-HLA and anti-non-HLA), even in the absence of a positive historical crossmatch, is associated with an increased risk for allosensitization, accelerated renal arteriosclerosis, and antibody-mediated graft failure.^{12, 14, 15} This risk is amplified with the coexistence of donor-specific antibodies and positive historical crossmatch.¹¹ In the past, a positive current crossmatch represented an absolute contraindication for transplant.^{4, 11, 12} Recent published reports demonstrate the possibility of ABMR leading to accelerated and late graft loss in HLA identical recipients with positive PRA, undetectable donor-specific antibodies, and negative B- and T-cell flow cytometry crossmatch on the day of transplant. The existence of anti-MICA antibodies before transplant may be responsible for graft loss.^{3, 15}

Current recommendations include the systematic determination of historical and current PRA, donor-specific antibodies (HLA and MICA), and complement-dependent cytotoxic and flow cytometry crossmatch by the most sensitive assays in every potential recipient before transplant. These recommendations are irrespective of the relation of the recipient with the donor, and they apply to recipients with the highest risk for sensitization such as patients with revision transplant, multigravida women, and recipients with a history of transfusion.^{1, 4, 15-17} The different types of desensitization protocols have been reviewed,⁴ and the present review will focus on the practical approach to these sensitized patients who are potential candidates for kidney transplant. This approach relies on an integrated analysis of several medical, economic, and logistic parameters that will enable the transplant team and the National Organization for Organ Sharing and Transplant (NOOST) in the decision-making about the suitability and feasibility of kidney transplant in these high-risk patients.

Anti-human leukocyte antigen antibodies: detection techniques

Several techniques are available to detect anti-HLA antibodies. In contrast with anti-HLA antibodies, anti-MICA antibodies are neither easily nor routinely evaluated in most transplant centers. With recent

evidence about the negative effect of anti-MICA antibodies on graft outcome, more transplant centers are measuring these antibodies before and after transplant, especially in previously sensitized patients or recipients with unexplained graft dysfunction.^{4, 5, 9, 10} Anti-HLA antibodies are determined by measuring PRA by complement-dependent cytotoxic techniques or a solid-phase assay using either enzyme-linked immunosorbent assay or flow cytometry, which is a more sensitive technique. Although PRA level, measured by enzyme-linked immunosorbent assay or flow cytometry, is an estimate of previously formed HLA antibodies, flow cytometry (Luminex, Austin, TX USA) single antigen donor-specific antibody assay, a multiple beads-based technology that is not yet available in many countries, offers a more sensitive and accurate tool to assess the type and the strength of each donor-specific antibody.^{4, 9, 11, 17} A shortcoming of the Luminex test is the lack of information about whether antibodies identified by Luminex can activate complement and harm the allograft.⁴ Crossmatches of both B and T cells between recipient-donor pairs are determined with 2 complementary techniques: complement-dependent cytotoxic and flow cytometry assays (the latter being more sensitive) provide a negative cutoff and allow the assessment of crossmatch strength.⁴

Approach to transplant from living donor

Every potential kidney transplant recipient from a living donor has a baseline quantitative PRA determined by flow cytometry screening or, preferably, Luminex technique; dual crossmatch by complement-dependent cytotoxic and flow cytometry techniques; and measurement of donor-specific antibodies by flow cytometry (preferably Luminex) before transplant.^{1, 4, 15-17} Historical crossmatch and/or donor-specific antibody titers are out of any type of immunologic conditioning such as statin, intravenous immunoglobulin, or anti-cluster of differentiation 20 (anti-CD20). The positive result of either test (crossmatch and donor-specific antibodies) is an absolute contraindication for transplant and an indication for desensitization, which is applied according to the level of risk for ABMR (Tables 1 and 2; Figure 2).^{4, 6, 11}

Those highly sensitized patients should preferably be transplanted from a deceased donor, rather than a living donor, because of the increased risk of ABMR and early graft failure and loss.^{4, 6, 11-13, 18} Panel

reactive antibodies in combination with crossmatch results and donor-specific antibody (HLA and MICA) levels are used to stratify immunologic risk according to sensitization status (Table 2; Figure 3).^{4, 11} Patients who are at extremely high risk (positive for both donor-specific antibodies and crossmatch) are not

Table 1. Estimated Immunologic Risk For Kidney Transplant Graft Rejection Upon Registration on the National Waiting List*

Panel Reactive Antibody Class I and Class II			Scoring For Each Individual Anti-HLA Antibody Class	Transplant Mode
Average [†]	Percentage [‡]	Index (%) [§]		
0 to 2	< 10	< 10	Low Risk	Living or deceased donor
2 to 4	10 to 20	< 10	Medium risk	
4 to 6	20 to 50	< 10	High risk	Preferably deceased donor
	10 to 20	> 10		
4 to 6	20 to 50	> 10	Very high risk	Refer to national priority list
6 to 8	> 50	> 10	Extremely high risk	

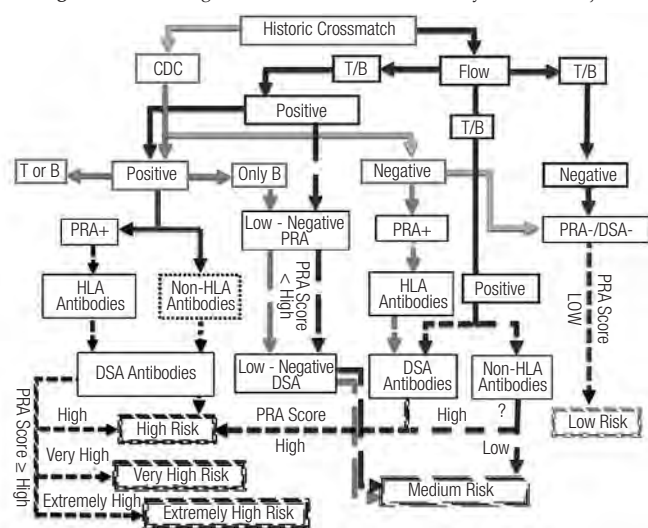
*Based on historical panel reactive antibody (PRA) scoring and the establishment of a virtual crossmatch. The scoring system relies on the combined PRA results in relation to average, percentage, and index. The sensitizing antigens, against which immune dominant antibodies are detected, are designated as the forbidden antigens for the patient. Only donors who do not carry these forbidden antigens are considered acceptable for this sensitized recipient.

[†]Average represents the overall avidity of the PRA reaction.

[‡]Percentage reflects the affinity (level or strength) of each individual antibody against human leukocyte antigen (HLA).

[§]Index is an assessment of the avidity and affinity of antibody-antigen binding combined.

Figure 2. Immunologic Risk Stratification For Antibody-Mediated Rejection



The assessment of the estimated risk relies on the combination of the panel reactive antibody (PRA) score, level of antibody against human leukocyte antigen (HLA) and non-HLA, donor-specific antibodies (DSA), and historic B and T cell crossmatch (complement-dependent cytotoxicity-CDC and flow cytometry) results. Patients are stratified into 5 risk groups: low, medium, high, very high, and extremely high. All except the extremely high risk group may receive transplant with specific desensitization and/or immunosuppressive strategies adjusted according to the risk level (Table 2 and Figure 3).

Table 2. Estimated Immunologic Risk for Kidney Transplant Graft Rejection at Time of Transplant*

Peak Panel Reactive Antibody			Crossmatch		DSA		Scoring	Transplantation Mode
Average [†]	Percentage [‡]	Index [§] (%)	Historical CDC	Current Flow	Current CDC	Flow and/or HLA MICA		
0-2	< 10	< 10	Neg	Neg	Neg	Neg	Low risk	Yes, living or deceased donor
2-4	10-20	<10	Neg	T/B Pos	Neg	Neg	Medium risk	Yes, living or deceased donor with antithymocyte globulin or anti-CD20 induction + intravenous immunoglobulin
			B Pos	Neg	Neg	MFI <3000		
4-6	20-50	< 10	T or B Pos	Neg	Neg	Neg	High risk	Yes, preferably deceased donor with desensitization and antithymocyte globulin induction (Figures 3 and 4)
	10-20	> 10	Neg	T/B Pos	Neg	Neg		
4-6	20-50	> 10	Pos	Pos	Neg	Neg	Very high risk	
6-8	> 50	> 10	Pos	Pos	Pos	Pos	Extremely high risk	No, waiting list Desensitization

Abbreviations: B, B cell; MFI, mean fluorescence intensity; MICA, MHC class 1 chain-related protein A; Neg, negative; Pos, positive; T, T cell

*Based on historical panel reactive antibody (PRA) scoring, historical and current crossmatch results obtained by complement-dependent cytotoxic-CDC and flow cytometry techniques, and the levels of current anti-human leukocyte antigen (HLA) and non-HLA donor-specific antibodies (DSA). This risk stratification should enable the transplant team in the decision-making about the transplant, desensitization, and immunosuppression.

[†]Average represents the overall avidity of the panel reactive antibody reaction.

[‡]Percentage is the affinity (level or strength) of each individual anti-HLA antibody.

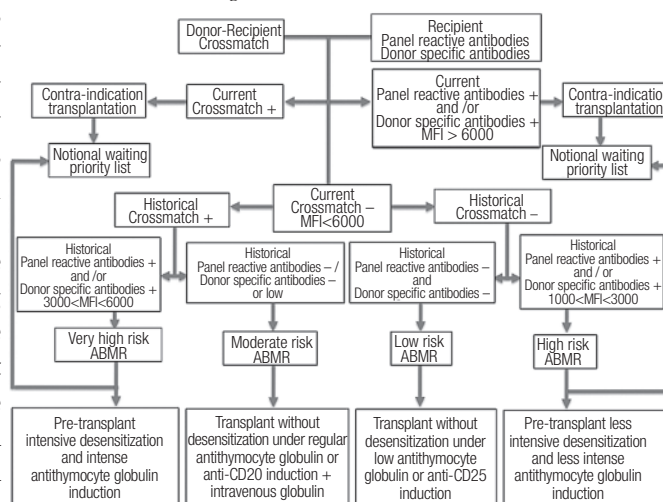
[§]Index is an assessment of the avidity and affinity of antibody-antigen binding combined.

transplanted from the specific donor, but may seek other living donors who do not carry the unacceptable antigens⁴ or are referred to the national waiting list where they are registered on the priority list. In cases of transplant from a deceased donor with a negative current crossmatch, desensitization may be started at the time of transplant.^{4,6}

Transplant from living donors in these highly sensitized patients may be considered only with the possibility of desensitization before transplant. This may require specific complex, lengthy, and very costly protocols, followed by intensive induction therapy based on antithymocyte globulin,^{4,6,12,13,18} that may not be covered by third party payers in many countries (Figures 3 and 4). These approaches are discouraged because of the high risk of early graft failure, high cost, and financial burden on the recipients, and they are used only for emergency indications and after ensuring adequate financial support. Patients who are considered at medium risk may receive a transplant from a living donor without desensitization before transplant, but they receive intensive induction therapy with antithymocyte globulin or anti-CD20 in cases with only a positive B-cell complement-dependent cytotoxic crossmatch, and intravenous immunoglobulin, as currently recommended by United Network for Organ Sharing.⁴ Low-risk patients with low PRA reactivity, negative test for donor-

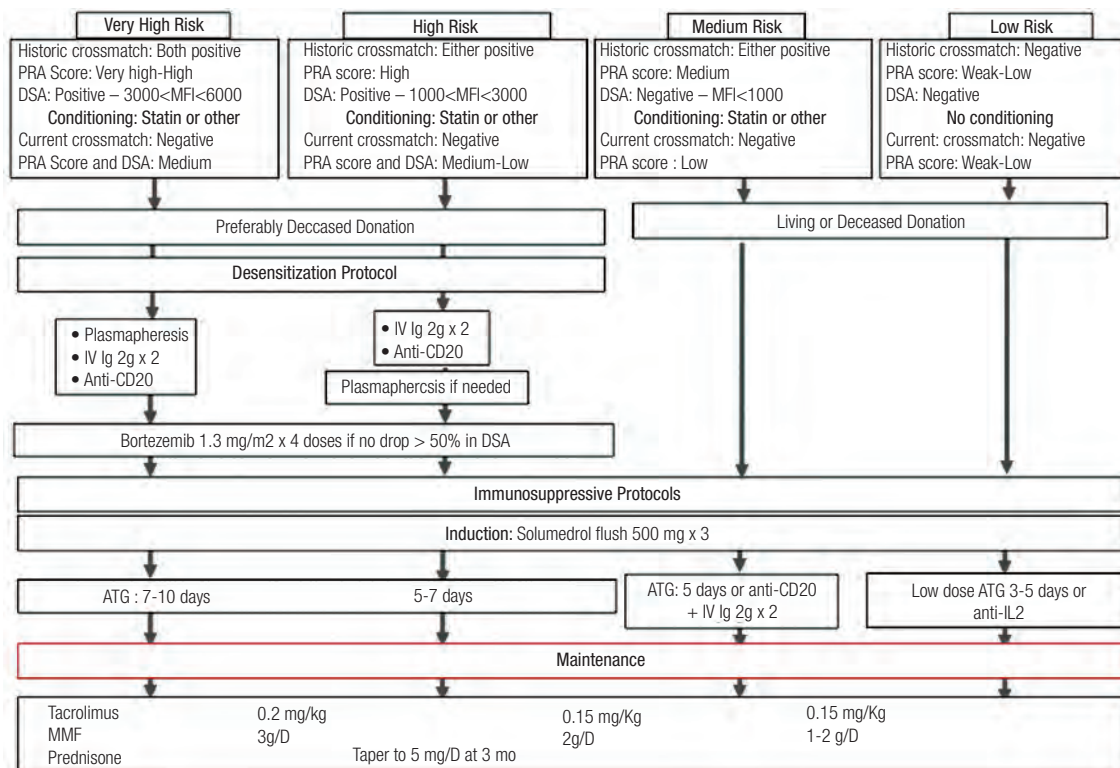
specific antibody (HLA and MICA), and negative crossmatch may be transplanted safely from a living donor using anti-CD25 induction therapy or a minimal amount of antithymocyte globulin.

Only 3% living kidney transplants in the United States are performed in highly sensitized patients with a PRA status > 80%, and 13% living kidney

Figure 3. Recommended Approach for Kidney Transplant for Sensitized Patients With a Living Donor

The decision-making is based on the estimated risk for occurrence of antibody-mediated rejection. It also depends on the capacity of the recipient to cover the financial expense after transplant in the absence of availability of financial coverage by either private or public third party payers.

Abbreviations: ABMR, antibody-mediated rejection; MFI, mean fluorescence intensity

Figure 4. Most Commonly Used Desensitization and Immunosuppressive Strategies According to the Estimated Risk for Possible Antibody-Mediated Rejection

Highly sensitized patients (risk $\geq$ high) receive transplants preferably from deceased donors. Irrespective of the donor type, desensitization is carried out before transplant. In case of transplant from a deceased donor with a negative current crossmatch, desensitization also may be done upon and after transplant.^{4, 6}

Abbreviations: ATG, antithymocyte globulin; DSA, donor-specific antibody; IV Ig, intravenous immunoglobulin; MFI, mean fluorescence intensity; MMF, mycophenolate mofetil; PRA, panel reactive antibody

transplants in the United States are performed in highly sensitized patients with a PRA status 10%. A greater percentage of these highly sensitized patients (15% for PRA status > 80%; 20% for PRA status 10%) are transplanted from a deceased donor.¹⁹ Such needed information is lacking in many countries of the Middle East. It is only through the creation of a national registry that these data will become available. These findings justify a mandatory approach, at the time of approval for the transplant, of a potential sensitized kidney graft recipient by the transplanting center and by the NOOST-governed Medical and Ethical Committees from all types of living donors. The final decision to proceed with transplant depends on the combined results of the current and historical crossmatch, donor-specific antibody and PRA obtained at the time of selection for transplant and guaranteed availability of adequate financial coverage (Table 2, Figures 2 and 3). Transplanting highly sensitized patients without these precautions may cause disastrous consequences for both graft and recipient survival.

Approach to transplant from deceased donor

Potential kidney transplant recipients with no available living donor have a baseline quantitative PRA obtained by flow cytometry screening (preferably by Luminex) before acceptance on the national waiting list. The information available from the assay may allow establishing a virtual crossmatch by a scoring system that may define the level of the potential risk for sensitization (Table 1).^{4, 11} Sensitized patients with a PRA score $\geq$ high or donor-specific antibodies mean fluorescence intensity (MFI) > 3000 by Luminex^{4, 5, 11} are placed as a priority by the NOOST on the national waiting list while defining, if possible, the unacceptable antigens against which they are sensitized.⁴ These highly sensitized patients may be desensitized by whatever means the transplant center elects to use according to different desensitization protocols (Figure 4).^{4, 6, 12, 13, 18, 20} Aside from statins,²¹ these protocols are extremely expensive²² and are not part of current practice in many countries.

There is no single universally confirmed strategy for desensitization. The aim of desensitization is to reduce the PRA level or donor-specific antibody

strength and to convert the results of a positive historical crossmatch to a current negative crossmatch at the time of transplant. Despite desensitization, these highly sensitized patients remain at a high risk to develop ABMR and early graft failure after transplant. Desensitization in highly sensitized patients who have previously formed donor-specific antibodies with negative crossmatch, using an intensive prophylactic immunosuppressive strategy combining intravenous immunoglobulin, anti-CD20, and plasmapheresis, may be carried out at the time of transplant. This may provide an acceptable 1-year: frequency of ABMR (17%), graft function (54 mL/min/1.73 m²), graft histology (13% with chronic and 7% with subclinical acute ABMR), and graft and patient survival frequencies at 1 year (graft, 89%; patient, 95%).^{4, 6}

The risk of ABMR after transplant in desensitized patients varies according to different studies and protocols from 10% to almost 100%.^{4, 12, 23} Donor-specific antibody strength and type before transplant may vary despite desensitization treatment, and peak donor-specific antibody level before transplant is a better predictor of subsequent development of ABMR than peak donor-specific antibody level on the day of transplant. Furthermore, patients with pre-existing donor-specific antibodies have worse graft outcome compared with patients with non-donor-specific anti-HLA antibodies or those with no prior sensitization. This decrease in graft survival also is observed in sensitized patients with donor-specific antibodies before transplant who do not develop ABMR after transplant.^{6, 11, 12, 15}

These findings justify a systematic approach by the NOOST at the time of the selection of a potential sensitized kidney graft recipient and organ allocation from a deceased donor (Figure 5). The current and historic crossmatch, PRA, and donor-specific antibodies may be available at the time of selection, and this may minimize time. The decision to allocate the kidney may depend on the combined results of these 6 parameters. A current positive crossmatch is an absolute contraindication for transplant. The systematic performance of these laboratory tests at the time of allocation incurs additional cost to the allocation and transplant process that may be covered by either the sensitized potential recipient or a third public or private party payer if the current crossmatch is positive. In the latter situation, and in the absence of a reimbursement mechanism by a public organization, the extra cost may be divided equally among all organs

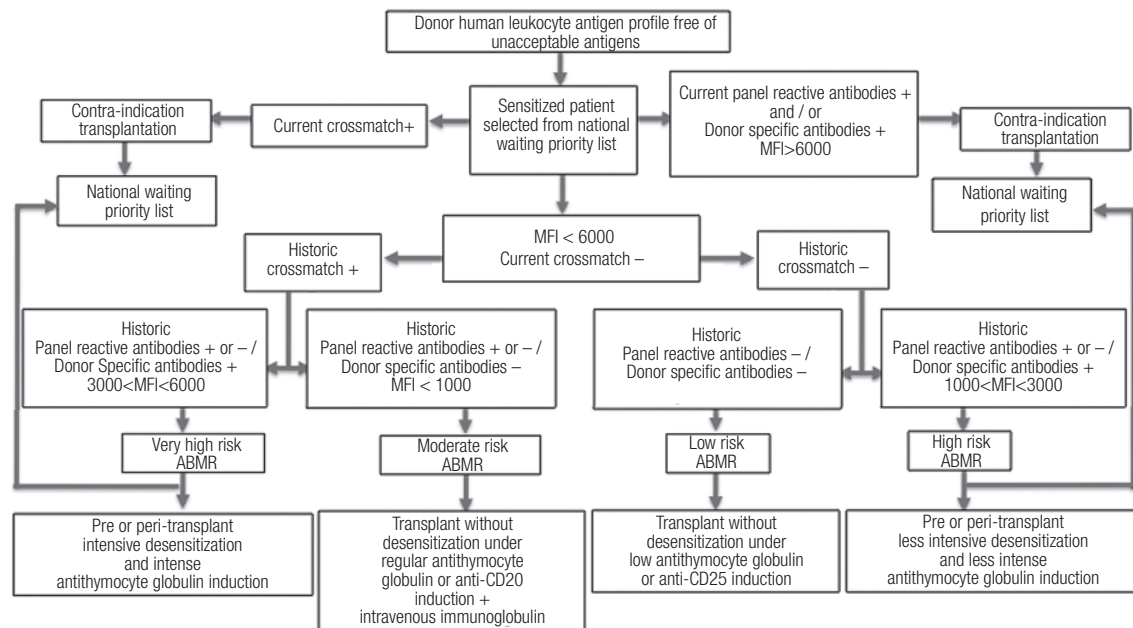
recipients and added to the individual baseline cost charged to each recipient by the NOOST.

Sensitized recipients should be equally informed about the associated risks and the high cost of desensitization strategies and immunosuppressive protocols before they accept to proceed with the transplant. These protocols are not covered by most public sector third party payers in many countries; therefore, these patients may be asked to provide a proof of adequate financial coverage during the registration process for the national waiting list, to ensure good quality care in accordance with international good practice standards. Sensitized patients that are identified as being at high risk should be informed initially by the transplant team about the risks and the estimated cost of the entire transplant procedure and the NOOST when they are accepted on the national list. Early information is essential to give them the opportunity to prepare financially before accepting the risk of transplant. If those patients cannot fulfill these requirements, they may be denied organ allocation and placed back on the priority list or be removed from the list.

Every patient registered on the national list may have an updated flow sheet in his or her medical file, at NOOST and the transplant center, that summarizes the baseline or historical and current immunologic parameters at the time of transplant. In case of blood transfusions, the PRA is repeated in potential recipients 1 month after the transfusion and reported to NOOST by the transplant center. After a sensitized patient is identified, it is preferable to have the serum specimens stored quarterly in the immunology laboratory. This will allow tracing the dynamic and multidirectional variations in PRA or donor-specific antibody levels spontaneously or after desensitization before transplant.^{6, 11} This practice will facilitate the task for NOOST of organ allocation for those sensitized patients on the priority list at time of procurement.

Monitoring sensitized patients after transplant

Highly sensitized patients at high risk for ABMR may be subject to a well structured monitoring process, with regular systematic immunologic, histologic, pharmacologic, and functional assessments of the graft after transplant.^{5, 6, 12, 14, 24, 25} This could be achieved with biopsies at the time of transplant; at 90 days, 1 year, and later after transplant;¹⁴ and as indicated clinically. In addition to measuring drug blood levels, crossmatch and donor-specific antibodies may be

Figure 5. Recommended Approach to Sensitized Patients on the Priority National Waiting List at the Time of Organ Allocation From a Deceased Donor

The donor human leukocyte antigen (HLA) profile should not contain any forbidden antigens against which the potential recipient is sensitized. Only patients with negative current crossmatch and mean fluorescence intensity (MFI) < 6000 are considered for transplant. The estimated risk for antibody-mediated rejection (ABMR) is determined according to an integrated analysis of historic crossmatch results, donor-specific antibodies, and panel reactive antibodies levels. Accordingly, the National Organization for Organ Sharing and Transplant and the transplant team will inform the patient about the immunologic risk and the high cost that may occur after transplant. The decision to proceed with the transplant procedure will depend on the ability of the recipient to cover the financial cost that may occur after transplant in the absence of financial coverage by either private or public third party payers.

determined monthly or bimonthly during this period, because the levels of donor-specific antibodies (HLA and MICA) after transplant may follow a dynamic and multidirectional mode similar to that observed before transplant.^{6, 12, 24, 25} This may require the preservation of blood and tissues from the donor spleen in the immunology laboratory. Any central laboratory accredited by the NOOST may have a storage facility in accordance with best practice guidelines. This approach may ensure early diagnosis of acute ABMR. Early therapeutic intervention and rapid reversal of the rejection is associated with improved graft survival compared with late intervention that may lead to refractory rejection and irreversible kidney damage.²⁶ Several factors affect the therapeutic response: time of ABMR diagnosis; type of desensitization and immunosuppressive antirejection regimens; level of graft dysfunction; other confounding factors such as coexisting or previous cellular rejection; transplant glomerulopathy; additional renal pathology such as focal segmental glomerulosclerosis, membranoproliferative glomerulonephritis, and thrombotic microangiopathy; and poor drug compliance.^{20, 24-27} This may explain the observed variation between centers about reported success frequencies of therapeutic response and graft outcomes.^{6, 18, 24-26}

Therapeutic regimens for ABMR are similar to desensitization protocols and vary at different transplant centers.^{4, 6, 18, 19, 23, 24} They are costly and complex (Table 3).^{4, 6, 19, 22} The cost may vary with the type of strategy adopted in different centers, total dosage of each treatment given, and therapeutic response and evolution of ABMR. These therapies are targeted at (1) antibody removal (plasmapheresis or immunoadsorption); (2) antibody neutralization (intravenous immunoglobulin); (3) B-cell depletion and inhibition (anti-CD20 [rituximab] and anti-CD52 [alemtuzumab] antibodies and antithymocyte globulin); (4) plasmacyte depletion (bortezomib); and (5) blockage of the activation of terminal complement (anti-C5 [eculizumab]) (Figure 6).^{4, 18, 20, 27-31} The therapeutic effect of these strategies differs according to the ABMR phenotype and the type of immune cells involved in the pathogenetic mechanism of the rejection episode. The identification of the mechanism of injury may be important for future therapy.²³⁻³¹ These expensive regimens are not readily available in many countries and frequently not covered by most third party payers.

Therefore, it is important that the transplant team and NOOST inform high-risk patients, early in the

Table 3. Calculated Cost During the Initial 5 Months After Transplant for Treatment of a Sensitized Patient with Acute Antibody-mediated Rejection*

Procedure	Test/Treatment	Quantity	Unit Price USD \$	Total Price USD \$
Transplant and desensitization				
Extracorporeal plasma exchange†	Plasmapheresis/immunoadsorption†	9	1000	9000
	Antithymocyte globulin†	30	464	13 920
	Human Immunoglobulin†			
Medications	6 g/amp	24	800	19 200
	Rituximab, 500 mg†	1	3100	3100
	Proteasome inhibitor: bortezomib†	4	1750	7000
	Valcyte tabs, 450 mg	90	51	4590
Immunology assays	Tacrolimus level	14	35	493
	Mycophenolic acid level	5	55	257
	Human leukocyte antigen (class 1 and 2)	1	350	350
	Panel reactive antibody (class 1 and 2)†	6	300	1800
	Crossmatch (complement-dependent cytotoxic and flow cytometry)†	6	100	600
	Donor-specific antibodies†	3	300	900
	Cerebrovascular disease panel	1	140	140
	Cell count†	5	80	400
Biopsy	Graft	1	600	600
			Total	62 350
Follow-up				
Extracorporeal plasma exchange	Plasmapheresis/immunoadsorption†	4	1000	4000
Medications	Proteasome inhibitor: bortezomib†	4	1750	7000
Biopsy	Graft†	1	600	600
	Panel reactive antibody (class 1 and 2)†	2	300	600
Immunology assays	Crossmatch†(complement-dependent cytotoxic and flow cytometry)	2	100	200
	Donor-specific antibodies†	2	300	600
	Cell count†	2	80	160
			Total	13 460
Grand total (desensitization and follow-up)			Total	75 810

*In United States dollars (USD \$). Excludes the cost of 30 days of the initial hospitalization period averaging USD \$25 000. This cost may vary according to the type of strategies adopted in different centers and the total dosage of each treatment that is given within the specific strategy, such as the number of plasmapheresis or immunoadsorption sessions, the total amount of thymoglobulin or antithymocyte globulin and rituximab, the number of cycles of proteasome inhibitor and the total amount of intravenous immunoglobulin, defined by the number of courses and dose per course. This cost could be heightened by the occurrence of potential complications in these patients.

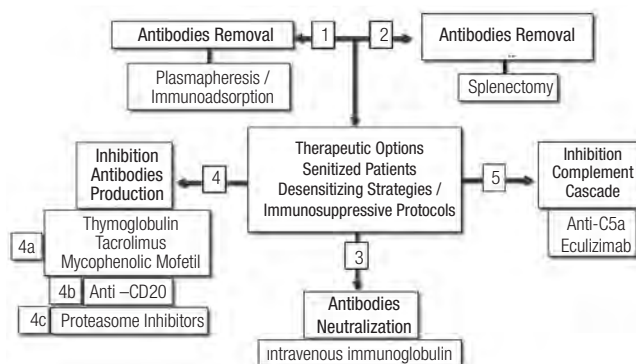
†Procedure, assay, or treatment that may be repeated > once, depending on the therapeutic response and the clinical course of ABMR.

preparation for transplant, about the potential risks and financial burden of ABMR.

Conclusions

Sensitized patients are at varied risk of developing ABMR. The risk differs with the degree of sensitization before desensitization. The strength of sensitization is additive and a function of the type and load or number of sensitization events (revision transplant > pregnancy > transfusion > autoimmune disease). The estimated risk of sensitization for patients with a living donor is determined from the combined results of the crossmatch (complement-dependent cytotoxic and flow cytometry) with the donor and those of the recipient's PRA and donor-specific antibodies (Table 2 and Figure 3).

Highly sensitized recipients may be referred to the national waiting priority list and informed about the financial and medical risks that may occur with transplant. The level of risk may guide the national medical and ethical committees in the approval or

Figure 6. Recommended Approach to Sensitized Patients on the Priority National Waiting List at the Time of Organ Allocation From a Deceased Donor

These strategies (1, 2, 3, and 4a) have been used previously, individually or collectively, in the desensitization process before transplant and in the treatment of antibody-mediated rejection (ABMR). Currently, more aggressive prophylactic strategies (1, 2, 3, 4a, and 4b) are used in sensitized patients with preformed antibodies at the time of transplant from a deceased donor. Newer drugs such as proteasome inhibitors (bortezomib) and anti-C5a drugs (eculizumab) (4c and 5) have been developed as additional potential treatment for desensitization and/or anti-rejection regimens, primarily in patients refractory to other therapies for ABMR. All these approaches are targeted at the immunologic mechanisms involved in the pathogenesis of the allosensitization process and the resulting humoral rejection.

refusal of transplant for genetically non-related donors and the transplant team for treatment before and after transplant (Figure 3). When medical approval is obtained for kidney transplant from a living donor, patients should undergo desensitization before transplant. For patients with no available living donor, the risk of sensitization may be estimated upon registration on the national waiting list, with a scoring system that is based on the potential recipient's PRA test results, to establish the virtual crossmatch. Patients are stratified according to immunologic status and estimated risk of developing ABMR (Table 1). High-risk patients may be placed on a priority list and informed about the potential risks and the need for financial preparation early upon registration (Table 3). When organs are allocated by NOOST from donors that do not carry the forbidden antigens, the risk for ABMR may be re-evaluated before transplant, in case of negative current crossmatch, using the historical crossmatch (complement-dependent cytotoxic and flow cytometry) and historical and current donor-specific antibody results (Table 2 and Figure 5). The results of the new risk reassessment, and a guarantee of financial coverage by a third party payer and/or the recipient, will dictate the proper decision to proceed with transplant and the specific required treatment after transplant (Figure 4).

In the absence of randomized controlled trials, these recommendations are formulated based on the most recent scientific evidence available. They could be adopted, implemented, and continuously updated by the NOOST and transplant centers to ensure safe and appropriate standards of patient care in accordance with best clinical practice.

References

1. Zou Y, Stastny P, Süsal C, Döhler B, Opelz G. Antibodies against MICA antigens and kidney-transplant rejection. *N Engl J Med*. 2007;357(13):1293-1300.
2. Champsaur M, Lanier LL. Effect of NKG2D ligand expression on host immune responses. *Immunol Rev*. 2010;235(1):267-285.
3. Narayan S, Tsai EW, Zhang Q, Wallace WD, Reed EF, Ettenger RB. Acute rejection associated with donor-specific anti-MICA antibody in a highly sensitized pediatric renal transplant recipient. *Pediatr Transplant*. 2011;15(1):E1-E7.
4. Marfo K, Lu A, Ling M, Akalin E. Desensitization protocols and their outcome. *Clin J Am Soc Nephrol*. 2011;6(4):922-936.
5. Terasaki PI, Ozawa M, Castro R. Four-year follow-up of a prospective trial of HLA and MICA antibodies on kidney graft survival. *Am J Transplant*. 2007;7(2):408-415.
6. Loupy A, Suberbielle-Boissel C, Zuber J, et al. Combined posttransplant prophylactic IVIg/anti-CD 20/plasmapheresis in kidney recipients with preformed donor-specific antibodies: a pilot study. *Transplantation*. 2010;89(11):1403-1410.
7. Li L, Chen A, Chaudhuri A, et al. Compartmental localization and clinical relevance of MICA antibodies after renal transplantation. *Transplantation*. 2010;89(3):312-319.
8. Reinsmoen NL, Lai CH, Heidecke H, et al. Anti-angiotensin type I receptor antibodies associated with antibody mediated rejection in donor HLA antibody negative patients. *Transplantation*. 2010;90(12):1473-1477.
9. Cox ST, Stephens HA, Fernando R, et al. Major histocompatibility complex class I-related chain A allele mismatching, antibodies, and rejection in renal transplantation. *Hum Immunol*. 2011;72(10):827-834.
10. Zhang Q, Cecka JM, Gjertson DW, et al. HLA and MICA: targets of antibody-mediated rejection in heart transplantation. *Transplantation*. 2011;91(10):1153-1158.
11. Lefaucheur C, Loupy A, Hill GS, et al. Preexisting donor-specific HLA antibodies predict outcome in kidney transplantation. *J Am Soc Nephrol*. 2010;21(8):1398-1406.
12. Loupy A, Hill GS, Suberbielle C, et al. Significance of C4d Banff scores in early protocol biopsies of kidney transplant recipients with preformed donor-specific antibodies (DSA). *Am J Transplant*. 2011;11(1):56-65.
13. Haririan A, Nogueira J, Kukuruga D, et al. Positive cross-match living donor kidney transplantation: longer-term outcomes. *Am J Transplant*. 2009;9(3):536-542.
14. Hill GS, Nochy D, Bruneval P, et al. Donor-specific antibodies accelerate arteriosclerosis after kidney transplantation. *J Am Soc Nephrol*. 2011;22(5):975-983.
15. Loupy A, Hill GS, Jordan SC. The impact of donor-specific anti-HLA antibodies on late kidney allograft failure. *Nat Rev Nephrol*. 2012;8(6):348-357.
16. Opelz G; Collaborative Transplant Study. Non-HLA transplantation immunity revealed by lymphocytotoxic antibodies. *Lancet*. 2005;365(9470):1570-1576.
17. Hyun J, Park KD, Yoo Y, et al. Effects of different sensitization events on HLA alloimmunization in solid organ transplantation patients. *Transplant Proc*. 2012;44(1):222-225.
18. Raghavan R, Jeroudi A, Achkar K, et al. Bortezomib in kidney transplantation. *J Transplant*. 2010;2010. pii: 698594.
19. Jordan SC, Toyoda M, Kahwaji J, Vo AA. Clinical aspects of intravenous immunoglobulin use in solid organ transplant recipients. *Am J Transplant*. 2011;11(2):196-202.
20. Walsh RC, Alloway RR, Giritla AL, Woodle ES. Proteasome inhibitor-based therapy for antibody-mediated rejection. *Kidney Int*. 2012;81(11):1067-1074.
21. Nurhan Ozdemir F, Akcay A, Sezer S, et al. Effect of simvastatin in the treatment of highly sensitized dialysis patients: the pre and post-renal transplantation follow-up outcomes. *Transpl Immunol*. 2004;13(1):39-42.
22. Woodle ES, Walsh RC, Alloway RR, Giritla A, Brailey P. Proteasome inhibitor therapy for antibody-mediated rejection. *Pediatr Transplant*. 2011;15(6):548-556.
23. Jordan SC, Reinsmoen N, Peng A, et al. Advances in diagnosing and managing antibody-mediated rejection. *Pediatr Nephrol*. 2010;25(10):2035-2048.
24. Waiser J, Budde K, Schütz M, et al. Comparison between bortezomib and rituximab in the treatment of antibody-mediated renal allograft rejection. *Nephrol Dial Transplant*. 2012;27(3):1246-1251.
25. Flechner SM, Fatica R, Askar M, et al. The role of proteasome inhibition with bortezomib in the treatment of antibody-mediated rejection after kidney-only or kidney-combined organ transplantation. *Transplantation*. 2010;90(12):1486-1492.
26. Walsh RC, Brailey P, Giritla A, et al. Early and late acute antibody-mediated rejection differ immunologically and in response to proteasome inhibition. *Transplantation*. 2011;91(11):1218-1226.
27. Ramos EJ, Pollinger HS, Stegall MD, Gloor JM, Dogan A, Grande JP. The effect of desensitization protocols on human splenic B-cell populations in vivo. *Am J Transplant*. 2007;7(2):402-407.
28. Stegall MD, Gloor JM. Deciphering antibody-mediated rejection: new insights into mechanisms and treatment. *Curr Opin Organ Transplant*. 2010;15(1):8-10.
29. Clatworthy MR. Targeting B cells and antibody in transplantation. *Am J Transplant*. 2011;11(7):1359-1367.
30. van den Hoogen MW, Hilbrands LB. Use of monoclonal antibodies in renal transplantation. *Immunotherapy*. 2011;3(7):871-880.
31. Farkash EA, Colvin RB. Diagnostic challenges in chronic antibody-mediated rejection. *Nat Rev Nephrol*. 2012;8(5):255-257.