Should Epitope-Based HLA Compatibility Be Used in the Kidney Allocation System?

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Abstract

The new kidney allocation system (KAS) still applies donor-recipient HLA compatibility at the antigen level and some four-digit alleles have been added recently. This system is used to record unacceptable mismatches for sensitized transplant candidates with serum HLA antibodies. Since the reactivities of such antibodies are specifically associated with epitopes rather than HLA antigens, a more scientifically accurate assessment of mismatch acceptability could be based on epitopes. HLA class I and class II epitope specificity analyses can now be readily performed with serum antibody assays with single allele panels. This report describes an epitope-based HLA compatibility system for KAS and involves recipient and donor HLA typing at the four-digit allele level. It focuses on sensitized patients who have serum antibodies specific for HLA epitopes that can be entered as unacceptable mismatches in the transplant candidate database. Newly developed software programs could readily identify compatible HLA types.

Introduction

After many years of deliberation and consensus-building, a new kidney allocation system (KAS) was implemented late 2014 by the United Network for Organ Sharing (UNOS) (1). Its goals are to improve longevity matching between donor kidneys and recipients and promote transplant access for historically disadvantaged subpopulations including highly sensitized patients. The probability of a HLA-compatible donor for such candidates can be assessed with a Calculated Panel-Reactive Antibody (CPRA) which reflects a percentage of potential donors with unacceptable antigens (2).

During the first year of KAS, the annual number of deceased donor transplants increased by 4.6% over the previous year (3). KAS has increased access to transplantation for recipients disadvantaged by antibody sensitization. About 8% of patients on the waiting list have 99–100% CPRA. Before KAS, they received just 2.5% of transplants, but this rose to 13.4% after KAS. This resulted in a decrease of 1000 such highly sensitized candidates on the waiting list. Transplant rates changed little for CPRA 95–98% recipients but declined for CPRA 80–89% recipients, from 6.8% to 2.7%. The remaining CPRA groups

showed moderate declines. After KAS, zero-ABDR mismatched transplants fell from 8.2% to 4.7% and zero-DR mismatched kidneys decreased from 19.8% to 16.8% (3). Of course, a much longer time is needed to evaluate the KAS effect on transplant outcome.

HLA Mismatch Acceptability Strategies in KAS

The HLA matching algorithm of UNOS is based on a list of HLA antigens recognized by the World Health Organization (WHO) Nomenclature Committee. Over a period of more than three decades, HLA compatibility criteria continued to change because of improved serological definitions of HLA antigens and the identification of "splits" or subtypes of HLA antigens. They led to new criteria for the compatibility algorithm including so-called matching equivalences between antigenic types. Molecularly based HLA typing methods improved the definitions of serologically defined antigens and recently, the HLA compatibility algorithm began to include selected four-digit alleles believed to represent unique antigens not recognized by the WHO serologic nomenclature. For instance, A9 was split into A23 and A24 whereas two A24 alleles A*24:02 and A*24:03 are now being used for matching purposes. DR2 was split into DR15 and DR16 whereas three DR15 alleles are listed: DRB1*15:01, DRB1*15:02 and DRB1*15:03.

KAS has updated criteria for HLA compatibility which besides the traditional HLA-A, HLA-B and HLA-DR loci now also include HLA-C, HLA-DQA,B and HLA-DPB. For each locus, the WaitList^{SM} entry page of the UNOS database has tables on which one can record unacceptable antigens for sensitized patients. They can be selected from lists of broad and split HLA antigens and selected 4-digit HLA alleles. For each of them there are so-called Donor Equivalent Antigens that must be avoided in the HLA types of potential donors. Supplemental Tables 1-3 describe details.

For instance, HLA-A1 listed as an unacceptable antigen the table shows A1 as the donor equivalent antigen and for an unacceptable HLA-A2 it lists A2, A0201, A0202, A0203, A0205 and A0206 as donor equivalents. An unacceptable A0201 has as donor equivalents A0201 plus the A2 antigen. The system becomes more complicated for A9 for which A9, A23, A24, A2402 and A2403 are listed as unacceptable donor equivalent antigens and for B5 for which B5, B51, B5101 and B5102 are listed (surprisingly, B52 is not listed although this antigen together with B51 is a split of B5). The HLA-DR table lists DR1, DR0101 and DR0102 but not DR0103 as donor equivalent antigens for the unacceptable DR1. Altogether, the system of antigen equivalents is confusing and needs to be revised.

Exclusive Use of HLA Alleles in Unacceptable Mismatching

The criteria for unacceptable HLA antigen equivalences are based on a mixture of old and new HLA typing information. Current molecularly based HLA typing data at the antigen level do not consider anymore broad specificities such as A10, A19, A28, B5 and DR6 because their splits can now be readily determined. Therefore, one must raise the question whether they should be maintained on the list of unacceptable antigens. Moreover, currently used serum testing methodologies determine antibody-reactivity with single four-digit HLA alleles.

For instance, many single allele panels used for antibody testing have just one A1 allele, namely A*01:01. A positive reaction would render A1 as unacceptable but in reality A*01:01 should be listed. The antibody testing kit might have several A2 alleles including A*02:01, A*02:03 and A*02:06. A positive reaction with all of them might suggest that the A2 antigen is an unacceptable mismatch but would it be better to list just the reactive A2 allele as unacceptable? Moreover, some alleles corresponding to the same HLA antigen react with a given serum but others are non-reactive. Typical examples are the A*24:02 and A*24:03 of A24 and B*44:02 and B*44:03 of B44. Such cases raise uncertainties about the mismatch acceptability of a HLA antigen.

Since mismatch acceptability is now determined with modern serum screening methods with four-digit allele panels, the antigen equivalences tables should list alleles rather than antigens. This means that A1 becomes A*01:01, the five A2 alleles *A*02:01, A*02:02, A*02:03, A*02:05 and A*02:06 remain but the A2 antigen would be excluded, A3 becomes A*03:01, etc. Antigens such as A9 should be removed because there are no A9-annotated alleles. A*23:01 (rather than A23), A*24:02 and A*24:03 should be used instead. There are also no A10-annotated alleles and the associated A25, A26, A34 and A66 antigens should be replaced with A*25:01, A*26:01, A34:01, A*34:02, A*66:01 and A*66:02; the latter four alleles are already listed in the antigen equivalences table for HLA-A.

Supplemental Tables 1-3 demonstrate how a replacement of the current system based on HLA antigen equivalents by an allele-based system will make the unacceptable mismatch algorithm much easier to manage. Such system is consistent with the concept that sensitized recipients and potential donors be typed at the four-digit allele level (4). The HLA types of such donors may consist of alleles that are listed in the unacceptable mismatch tables. Accordingly, it seems quite easy to distinguish which alleles are acceptable or unacceptable.

However, the unacceptable mismatch tables would be limited to alleles that are used in the antibody screening assays. For instance, most HLA-ABC kits have fewer than 100 alleles although more 8000 class I alleles have been identified and the list is still growing. Given the increasing racial and ethnic diversity of the US population, one would expect more frequent occurrences of non-panel HLA alleles in transplant donors. How does one determine the mismatch acceptability of an untested donor allele not listed in the unacceptable mismatch table? This question can especially create a dilemma if two or more alleles corresponding to a given HLA antigen have been assigned differently in terms of mismatch acceptability.

HLA Epitope-Specific Antibody Analysis

This problem can be solved by applying the principle that HLA antibodies are specific for epitopes (5, 6). Each allele consists of a string of epitopes that can be classified by eplets, i.e. small configurations of polymorphic amino acid configurations on the HLA molecular surface. Certain epitopes specifically associated with antibody reactivity are solely defined by single eplets and others require combinations of eplets with nearby residue configurations, they are referred as eplet pairs.

The clinical relevance of epitope-based matching should of course, only apply to epitopes that have been experimentally verified with informative antibodies. The HLA Epitope Registry (http://www.epregistry.ufpi.br) has for each locus a list of epitopes specifically associated with antibody reactivity . The website now includes a downloadable PDF file “EpiPedia of HLA” which describes the experimental evidence of HLA epitopes specifically associated with antibody reactivity. In our experience, the HLAMatchmaker antibody analysis programs (Version 2.0 downloadable from www.HLAMatchmaker.net) have shown that the reactivity of more than 90% of post-pregnancy sera can be explained with epitopes that have been specifically associated with antibody reactivity (7-9). Thus, considerable progress has been made in defining HLA epitope repertoires but more studies are needed. The www.HLAMatchmaker.net website has now a downloadable Excel document “Five Maps of HLA Epitopia” which describe the sequence locations of eplets that correlate with antibody specificity and polymorphic residues as potential candidates defining additional epitopes. These maps can be used in navigating the continents of HLA Epitopia while searching for newly antibody-defined epitopes (10).

Antibody-Reactive Epitopes and Mismatch Acceptability at the Allele Level

Any HLA allele that carries an epitope specifically associated with antibody reactivity of a pre-transplant serum must be called an unacceptable mismatch. One could also call an epitope that is specifically associated with antibody reactivity as an unacceptable mismatch and this consideration offers several

advantages in the management of the sensitized transplant candidate. The identification of epitope specificities will increase our understanding of complex serum reactivity patterns often seen for highly sensitized patients. Certain sera should be further investigated with absorption-elution studies with selected alleles. The HLA typing information of the patient and preferably the immunizing donor(s) will greatly facilitate the epitope specificity analysis.

Since most sera have antibody reactivities associated with small numbers of epitopes, it would be rather easy for histocompatibility testing laboratories to enter such unacceptable epitopes into the WaitList[SM] entry page of the UNOS database. A dedicated software program could readily determine from spreadsheets of alleles with strings of antibody-associated epitopes which alleles are acceptable or unacceptable mismatches. This approach will enhance the so-called virtual crossmatch test because donor HLA alleles not used in antibody screening can be readily included because they have the unacceptable epitope information.

Table 1 has examples how matching at the epitope level can differentiate between mismatch acceptability and unacceptability of HLA alleles. Epitopes have been selected for four sets of HLA-A and four sets of HLA-B alleles. Each set has one or more HLA alleles generally used in antibody testing assays; they are marked with asterisks. Such panel alleles give positive reactions if they carry the epitope specifically recognized by antibody and this means that they carry an unacceptable epitope mismatch.

For instance, A*11:01 and A*11:02 give positive reactions with antibodies specific for each of the five epitopes selected for Table 1 whereas three epitope-specific antibodies (62EE, 82LR and 144KR) give positive reactions with A*24:02 and A*24:03. Conversely, 166DG-specific antibodies react with A*24:02 but not with A*24:03. This means that A*24:02 is an unacceptable mismatch but A*24:03 is acceptable for recipients with 166DG-specific antibodies.

The primary purpose of Table 1 is to illustrate how epitope-based HLA compatibility can determine the mismatch acceptability of HLA alleles not used in antibody assays. Their epitope repertoires can be readily defined from amino acid sequence comparisons and molecular modeling. For instance, five of the six untested A11* alleles have the 90D eplet; they can be considered unacceptable mismatches for patients with 90D-specific antibodies. In contrast, A*11:12 lacks 90D (it has 90A instead) and would be an acceptable mismatch. A*11:05 is an acceptable mismatch for patients with 144KR-specific antibodies. Table 1 has many more examples of a given epitope specifically reacting with antibody, that alleles not tested in serum screening assays can be reported as acceptable or as unacceptable mismatches. After identifying all epitopes specifically associated with a transplant candidate's antibodies it becomes possible to classify all potential donor alleles in terms of mismatch acceptability.

Epitope-Based Compatibility for HLA-C, HLA-DRB3/4/5, HLA-DQ and HLA-DP

The above considerations apply also to the products of other HLA loci. KAS has additional tables with corresponding unacceptable antigen equivalences. Although HLA-C unacceptable antigen equivalences can be readily converted it should be noted that HLA-C alleles have certain epitopes that are shared with HLA-A and/or HLA-B alleles; examples are 80N, 90D and 163EW shown in Table 1. Altogether, class I epitope compatibility should be applied to alleles encoded by the combined HLA-A, -B, -C loci.

At present, KAS has tables for the Bw4 and Bw6 antigens on groups of equivalent HLA-B antigens and alleles. These antigens are defined by polymorphic residues in sequence positions 79-83 and they represent distinct groups of epitopes, including the Bw4 epitope 82LR which is also present on A23, A24, A25 and A32 alleles and Bw6 epitope 80N also shared with many HLA-C alleles. These tables are unnecessary because the class I epitope-based compatibility tables already include the Bw4- and Bw6-assocated epitopes.

The KAS tables have unacceptable antigen equivalences for DR51, DR52 and DR53 and they display already four-digit DRB5, DRB3 and DRB4 alleles that are generally used in DRB antibody testing. Each allele has epitopes that are locus-specific but many others are shared with DRB1 alleles. A DRB epitope compatibility algorithm should be used for the combination of all four DRB loci.

KAS has separate tables for DQA and DQB unacceptable antigen equivalences. The recently proposed DQA table has a rather long list of DQA1 alleles but many of them are not used in the kits used to determine antibodies and therefore, they can never be reported as unacceptable mismatches. The DQB table reflects a DQB1 allele panel commonly used in antibody testing assays.

Table 2 illustrates how class II matching at the epitope level can differentiate between mismatch acceptability and unacceptability for three DRB1 allele groups, namely DRB1*04, DRB1*11 and DRB1*14. Alleles generally used in antibody testing assays are marked with asterisks. From their positive and negative reactions with antibodies specific for selected epitopes one can readily determine which untested alleles are acceptable or unacceptable mismatches. Table 2 shows how three DQB epitopes specifically associated with antibody reactivity affect the mismatch acceptability of different DQB1*06 alleles.

Although DQA1 and DQB1 alleles have distinct repertoires of epitopes it should be noted DQA and DQB are parts of the same molecule referred to as the DQ heterodimer. It is possible that certain DQ epitopes are defined by pairs of DQA1 and DQB1 eplets (11, 12).

KAS has now included HLA-DPB compatibility because there is ample evidence that DP antibodies affect transplant outcome. DPB panels commonly used to test antibody reactivity have relatively small numbers of epitopes specifically associated with antibody reactivity.

Discussion

HLA antibodies are primary causes of transplant failures and it is now widely accepted that their reactivity is associated with specific epitopes. Accordingly, HLA matching at the epitope level provides better opportunities to identify suitable donors for transplant patients. Traditionally, HLA matching criteria have been based on serologically defined HLA antigens. Large collaborations during the early international HLA workshops resulted in the identification of distinct serum clusters with highly correlated reactivity patterns. This led to an arbitrary notation system of serological antigens such as HLA-A1, -A2, -B7, -B8, -DR1 and -DR2, etc. Since we know now that HLA antibodies are specific for epitopes rather than HLA antigens it seems obvious that HLA typing sera must recognize distinct epitopes uniquely present on serologically defined antigens. An HLAMatchmaker analysis has shown that many HLA antigens have distinctly unique eplets (13). For Instance, anti-A1 antibodies are specifically associated with an epitope defined by the 163RG eplet which is only found on A1 molecules, whereas anti-B7 antibodies are specific for 177DK unique on B7 and anti-DR1 antibodies specifically react with 12LKF_2 unique on DR1. Accordingly, the HLA antigen matching effect on transplant outcome can be reinterpreted as actually demonstrating the influence of matching for epitopes although rather small numbers of epitopes were considered in these graft survival association studies.

Many HLA antibodies have complex reactivity patterns some of them could be translated to so-called Cross-Reacting Groups (CREGs) of HLA antigens; the A2-CREG and B7-CREG are common examples. Each CREG has so-called public determinants shared between certain groups of antigens and so-called private determinants limited to a given serologically defined antigen within the CREG (14). Most highly sensitized patients have antibodies against public epitopes which can now be defined structurally. As an example, the A2-CREG which includes A2, A23, A24, A68, A69 and B17 has several epitopes corresponding to public determinants including the 127K eplet on A2+A23+A24+A68+A69, 144TKH on

A2+A68+A69, 107W on A2+A69 and 62GE on A2+B17. Altogether, HLA class antigens have private and public determinants which actually represent epitopes that can be structurally defined.

After elucidation of the molecular structure and amino acid sequences of HLA molecules during the late nineteen-eighties, the application of DNA-based technologies permitted accurate HLA antigen typing results. Many serologically identical antigens showed amino acid sequence variations annotated as four-digit alleles that had often enough different epitope repertoires. It is now recognized that HLA compatibility is better determined at the allele than at the antigen level (4).

This report describes an epitope-based HLA compatibility system proposed for KAS and involves recipient and donor HLA typing at the four-digit allele level. It focuses on sensitized patients who have serum antibodies that need to be tested for their epitope specificities. Eurotransplant has incorporated HLAMatchmaker in the Acceptable Mismatch program to identify donors for highly sensitized patients (15, 16). The availability of single allele panels for antibody analysis will permit the identification of antibody-reactive epitopes as unacceptable mismatches. Such epitopes should be avoided on donor HLA types and HLAMatchmaker can readily determine the unacceptable alleles including those that have not been tested.

A KAS implementation of an epitope-based HLA compatibility program for sensitized transplant candidates deserves some logistical considerations. First, HLA typing should be done at the four-digit allele level. This can be readily accomplished for all transplant candidates and live donors but there might be time constraints for certain deceased donors unless more rapid DNA typing methods become available. Second, several methods can be used for antibody testing and there are different assessments of positive reactions. While this report cannot address these issues, the goal of any antibody analysis should be a reliable identification of epitopes specifically recognized by transplant candidate antibodies. Third, histocompatibility laboratories would enter such unacceptable epitopes on the WaitList^{SM} entry page of the UNOS database. This would be much easier than entering reactive HLA alleles which for highly sensitized patients, would constitute long lists. There will be a need for a new software program with a spreadsheet of epitope repertoires of potential donor alleles so that the entered epitope information will convert alleles as acceptable or unacceptable mismatches. It could also perform a much more accurate virtual cross-match test between a transplant candidate and a potential donor.

HLA matching at the epitope level can also benefit transplant outcome in non-sensitized patients who have no donor-specific HLA antibodies before transplantation. Numerous studies have demonstrated significant correlations between eplet loads of HLA mismatches and the development of donor-specific class I and class II antibodies as well as rejection incidence and allograft outcome (17-31). These findings have recently led to the implementation of new donor selection strategies for non-sensitized transplant candidates especially younger patients (32-34). HLA mismatches with low eplet loads can be expected to improve transplant outcome. Even if the first allograft rejected, retransplant candidates might become less highly sensitized thereby making it easier to find acceptable mismatches.

In conclusion, epitope-based HLA compatibility testing offers a new strategy in the clinical transplant setting. It relies on HLA typing at the four-digit allele level and permits a more comprehensive identification of acceptable mismatches for sensitized patients and an effective permissible mismatch strategy for non-sensitized patients. Our understanding of HLA epitopes has made steady progress and new investigative approaches address the mechanisms of epitope immunogenicity and how they could affect mismatch permissibility (35-42).

Conflict of interest statement

The author has no conflicts of interest to disclose as described by Human Immunology

References

1. Israni AK, Salkowski N, Gustafson S, al. e. New national allocation policy for deceased donor kidneys in the United States and possible effect on patient outcomes. J Am Soc Nephrol 2014;25:1842–1848.
2. Cecka JM, Kucheryavaya AY, Reinsmoen NL, Leffell MS. Calculated PRA: Initial results show benefits for sensitized patients and a reduction in positive crossmatches. Am J Transplant. 2011;11:719-724.
3. Stewart DE, Kucheryavaya AY, Klassen DK, Turgeon NA, Formica RN, Aeder MI. Changes in Deceased Donor Kidney Transplantation One Year After KAS Implementation. Amer J Transplant. 2016;16:1834-1847.
4. Duquesnoy RJ, Kamoun M, Baxter-Lowe LA, Woodle ES, Bray RA, Claas FHJ, Eckels DD, Friedewald JJ, Fuggle SV, Gebel HM, Gerlach JA, Fung JJ, Middleton D, Nickerson P, Shapiro R, Tambur AR, Taylor CJ, Tinckam K, Zeevi A. Personal Viewpoint: Should HLA Mismatch Acceptability for Sensitized Transplant Candidates Be Determined at the High-Resolution rather than the Antigen Level? Am J Transplant. 2015;20:1-6.
5. Duquesnoy RJ. HLA epitope based matching for transplantation. Transplant Immunology. 2014;31:1-6.
6. Duquesnoy RJ. Human leukocyte antigen epitope antigenicity and immunogenicity. Current Opinion in Organ Transplantation. 2014;19:428-435.
7. Duquesnoy R, Hönger G, Hösli I, Marrari M, Schaub S. Detection of newly antibody-defined epitopes on HLA class I alleles reacting with antibodies induced during pregnancy. Int J Immunogenetics. 2016;43:200-209.
8. Duquesnoy R, Hönger G, Hösli I, Marrari M, Schaub S. Antibody-defined epitopes on HLA-DQ alleles reacting with antibodies induced during pregnancy and the design of a DQ eplet map. Hum Immunol. 2016; 77: 824-831.
9. Duquesnoy RJ, Honger G, Hosli I, Marrari M, Schaub S. Identification of epitopes on HLA-DRB alleles reacting with antibodies in sera from women sensitized during pregnancy. Hum Immunol. 2015; 77: 214-222
10. Duquesnoy RJ. Five Maps for HLA Epitopia: Their usefulness in searches for newly antibody-defined epitopes.See HLAMatchmaker website www.epitopes.net 2016.
11. Tambur AR, Rosati J, Roitberg S, Glotz D, Friedewald JJ, Leventhal JR. Epitope analysis of HLA-DQ antigens: what does the antibody see? Transplantation. 2014;98:157-166.
12. Tambur A. HLA-DQ antibodies: are they? Are they relevant? Why so many? Curr Opin Organ Transplant. 2016;21:xx.
13. Duquesnoy RJ. Reflections on HLA Epitope-Based Matching for Transplantation. Frontiers in Immunology. 2016;In Press.
14. Rodey GE, Fuller TC. Public epitopes and the antigenic structure of the HLA molecules. Critical Reviews in Immunology. 1987;7:229-267.
15. Claas FHJ, Dankers MK, Oudshoorn M, van Rood JJ, Mulder A, Roelen DL, Duquesnoy RJ, Doxiadis IIN. Differential immunogenicity of HLA mismatches in clinical transplantation. Transplant Immunol. 2005;14:187-191.
16. Doxiadis IIN, Duquesnoy RJ, Claas FHJ. Extending options for highly sensitized patients to receive a suitable kidney graft. Curr Opin Immunol. 2005;17:536-540.
17. Dankers MKA, Witvliet MD, Roelen DL, De Lange P, Korfage N, Persijn GG, Duquesnoy RJ, Doxiadis IIN, Claas FHJ. The Number of Amino Acid Triplet Differences between Patient and Donor is Predictive for the Antibody Reactivity Against Mismatched HLA Antigens. Transplantation. 2004;77:1236-1239.

18. Goodman RS, Taylor CJ, O'Rourke CM, Lynch A, Bradley JA, Key T. Utility of HLAMatchmaker and single-antigen HLA-antibody detection beads for identification of acceptable mismatches in highly sensitized patients awaiting kidney transplantation. Transplantation. 2006;81:1331-1336.
19. Kosmoliaptsis V, Bradley JA, Sharples LD, Chaudhry A, Key T, Goodman RS, Taylor CJ. Predicting the immunogenicity of human leukocyte antigen class I alloantigens using structural epitope analysis determined by HLAMatchmaker. Transplantation. 2008;85:1817-1825.
20. Duquesnoy RJ, Awadalla Y, Lomago J, Jelinek L, Howe J, Zern D, Hunter B, Martell J, Girnita A, Zeevi A. Retransplant Candidates Have Donor-Specific Antibodies that React with Structurally Defined HLA-DR,DQ,DP Epitopes. Transplant Immunology. 2008;18:352-360.
21. Kosmoliaptsis V, Chaudhry AN, Sharples LD, Halsall DJ, Dafforn TR, Bradley JA, Taylor CJ. Predicting HLA class I alloantigen immunogenicity from the number and physiochemical properties of amino acid polymorphisms. Transplantation. 2009;88:791-798.
22. Duquesnoy RJ, Marrari M. Detection of Antibodies against HLA-C Epitopes in Patients with Rejected Kidney Transplants. Transplant Immunol. 2011;24:164-171.
23. Honger G, Fornaro I, Granado C, Tiercy JM, Hosli I, Schaub S. Frequency and determinants of pregnancy-induced child-specific sensitization. American Journal of Transplantation. 2013;13:746-753.
24. Wiebe C, Nickerson P. Posttransplant monitoring of de novo human leukocyte antigen donor-specific antibodies in kidney transplantation. Current Opinion in Organ Transplantation. 2013;18:470-477.
25. Wiebe C, Nickerson P. Acceptable mismatching at the class II epitope level: the Canadian experience. Curr Opin Organ Transplant. 2014;19:442–446.
26. Wiebe C, Nevins TE, Robiner WN, al. e. The synergistic effect of class II HLA epitope mismatch and nonadherence on acute rejection and graft survival. Am J Transplant. Am J Transplant. 2015;15:doi: 10.1111/ajt, 13341.
27. Filippone EJ, Farber JL. Humoral immunity in renal transplantation: epitopes, Cw and DP, and complement activating capability – an update. Clin Transplant. 2015;29:279-287.
28. Sapir-Pichhadze R, Tinckam K, Quach K, al. e. HLADR and -DQ eplet mismatches and transplant glomerulopathy: a nested case-control study. . Am J Transplant 2015;15:137.
29. Singh P, Filippone E, Colombe B, Shah A, Zhan T, Harach M, Gorn C, Frank A. Sensitization trends after renal allograft failure: the role of DQ eplet mismatches in becoming highly sensitized. Clin Transplant. 2015;3DOI: 10.1111/ctr.1266.
30. Sullivan P, Warner P, Kemna M, Albers E, Law S, Weiss N, Law Y. HLA molecular epitope mismatching and long-term graft loss in pediatric heart transplant recipients. J Heart and Lung Transplant. 2016;34:950-957.
31. Walton D, Hiho S, Cantwell LS, Diviney M, Wright S, Snell G, Paraskeva M, Westall G. HLA Matching at the Eplet Level Protects Against Chronic Lung Allograft Dysfunction. Am J Transplant. 2016;20:1-9.
32. Bryan C, Chadha V, Warad B. Donor selection in pediatric kidney transplantation using DR and DQ eplet mismatching: A new histocompatibility paradigm. Pediatric Transplantation. 2016;20 in press.
33. Kausman JY, Walker AM, Cantwell LS, Quinlan C, Sypek MP, Ierino FL. Application of an epitope-based allocation system in paediatric kidney transplantation Pediatric Transplantation. 2016;20 in press.
34. Duquesnoy RJ. The Eplet Load Concept in Clinical Transplantation (Editorial). Pediatric Transplantation. 2016;20:In Press.
35. Duquesnoy RJ. The antibody response to an HLA mismatch: a model for nonself-self discrimination in relation to HLA epitope immunogenicity. Int J Immunogenetics. 2011;39:1-9.
36. Duquesnoy RJ. Humoral Alloimmunity in Transplantation: Relevance of HLA Epitope Antigenicity and Immunogenicity. Frontiers in Transplantation and Alloimmunity. 2011;2:59 (published online).

37. Kosmoliaptsis V, Sharples LD, Chaudhry AN, Halsall DJ, Bradley JA, Taylor CJ. Predicting HLA class II alloantigen immunogenicity from the number and physiochemical properties of amino acid polymorphisms. Transplantation. 2011;91:183-190.
38. Mallon DH, Bradley JA, Taylor CJ, Kosmoliaptsis V. Structural and electrostatic analysis of HLA B-cell epitopes: inference on immunogenicity and prediction of humoral alloresponses. Current Opinion in Organ Transplantation. 2014;19:420-427.
39. Mallon DH, Bradley JA, Winn PJ, Taylor CJ, Kosmoliaptsis V. Three-dimensional structural modelling and calculation of electrostatic potentials of HLA Bw4 and Bw6 epitopes to explain the molecular basis for alloantibody binding: toward predicting HLA antigenicity and immunogenicity. Transplantation. 2015;99:385-390.
40. Otten HG, Calis JJA, Kesmir C, van Zuilen AD, Spierings E. Predicted indirectly recognizable HLA epitopes presented by HLA-DR correlate with the de novo development of donor-specific HLA IgG antibodies after kidney transplantation. Human Immunology. 2013;74:290-296.
41. Geneugelijk K, Honger G, van Deutekom H, Thus K, Kesmir C, Hosli I, Schaub S, Spierings E. Predicted Indirectly ReCognizable HLA Epitopes presented by HLA-DRB1 (PIRCHE-II) are related to HLA-antibody formation during pregnancy Amer J Transplant. 2015; *15:3112-22*
42. Duquesnoy RJ. The Antibody Response to a HLA Mismatch: Putting Together the Pieces of a Puzzle. Am J Transplant. 2015;15:3019-3020.

Table 1 Examples of HLA-A and HLA-B allele mismatch acceptability determined by epitopes specifically associated with serum antibody reactivity

Epitope	90D	144KR	150AAH	151AHA	163RW
A*11:01*	Positive	Positive	Positive	Positive	Positive
A*11:02*	Positive	Positive	Positive	Positive	Positive
A*11:03	UnAcc	UnAcc	Acc	Acc	UnAcc
A*11:04	UnAcc	UnAcc	UnAcc	UnAcc	Acc
A*11:05	UnAcc	Acc	UnAcc	UnAcc	UnAcc
A*11:06	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
A*11:08	UnAcc	UnAcc	UnAcc	Acc	UnAcc
A*11:12	Acc	UnAcc	UnAcc	UnAcc	UnAcc

Epitope	62RR	66NV	76ANT	163RW
A*26:01*	Positive	Positive	Positive	Positive
A*26:02	UnAcc	UnAcc	UnAcc	UnAcc
A*26:03	UnAcc	UnAcc	Acc	UnAcc
A*26:05	UnAcc	UnAcc	Acc	UnAcc
A*26:06	UnAcc	UnAcc	Acc	UnAcc
A*26:07	Acc	Acc	UnAcc	UnAcc
A*26:08	UnAcc	UnAcc	UnAcc	UnAcc
A*26:09	UnAcc	UnAcc	UnAcc	Acc
A*26:12	UnAcc	UnAcc	UnAcc	UnAcc
A*26:16	UnAcc	UnAcc	UnAcc	UnAcc
A*26:17	UnAcc	UnAcc	UnAcc	UnAcc
A*26:20	UnAcc	UnAcc	UnAcc	UnAcc

Epitope	62RR	144TKH	150AAH
A*68:01*	Positive	Positive	Positive
A*68:02*	Positive	Positive	Positive
A*68:03	UnAcc	UnAcc	UnAcc
A*68:04	UnAcc	UnAcc	UnAcc
A*68:05	UnAcc	UnAcc	UnAcc
A*68:06	UnAcc	UnAcc	UnAcc
A*68:07	UnAcc	UnAcc	UnAcc
A*68:08	UnAcc	UnAcc	UnAcc
A*68:10	Acc	UnAcc	UnAcc
A*68:12	UnAcc	Acc	UnAcc
A*68:15	UnAcc	UnAcc	UnAcc
A*68:16	UnAcc	UnAcc	Acc
A*68:17	UnAcc	UnAcc	UnAcc
A*68:19	UnAcc	UnAcc	UnAcc
A*68:20	UnAcc	UnAcc	UnAcc
A*68:23	UnAcc	UnAcc	UnAcc
A*68:25	UnAcc	UnAcc	UnAcc
A*68:31	UnAcc	UnAcc	UnAcc

Epitope	71TTS	76ESN	80N	156DA	66IF+163TEW
B*08:01*	Positive	Positive	Positive	Positive	Positive
B*08:02	Acc	Acc	Acc	UnAcc	UnAcc
B*08:03	Acc	Acc	Acc	UnAcc	UnAcc
B*08:04	UnAcc	UnAcc	UnAcc	UnAcc	Acc
B*08:05	UnAcc	UnAcc	UnAcc	UnAcc	Acc
B*08:06	Acc	Acc	UnAcc	Acc	UnAcc
B*08:07	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*08:09	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*08:12	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*08:13	UnAcc	UnAcc	UnAcc	Acc	UnAcc
B*08:20	UnAcc	UnAcc	UnAcc	Acc	UnAcc

Epitope	41T	113H	163EW	180E
B*40:01*	Positive	Positive	Positive	Positive
B*40:02*	Positive	Positive	Positive	Negative
B*40:06*	Positive	Positive	Positive	Negative
B*40:03	UnAcc	UnAcc	UnAcc	Acc
B*40:04	UnAcc	UnAcc	UnAcc	Acc
B*40:05	UnAcc	UnAcc	Acc	Acc
B*40:07	UnAcc	UnAcc	UnAcc	UnAcc
B*40:08	UnAcc	UnAcc	UnAcc	Acc
B*40:09	UnAcc	Acc	UnAcc	Acc
B*40:10	UnAcc	UnAcc	UnAcc	UnAcc
B*40:11	UnAcc	UnAcc	UnAcc	Acc
B*40:12	Acc	UnAcc	UnAcc	UnAcc
B*40:13	UnAcc	UnAcc	UnAcc	Acc

Epitope	62EE	82LR	144KR	166DG
A*24:02*	Positive	Positive	Positive	Positive
A*24:03*	Positive	Positive	Positive	Negative
A*24:04	UnAcc	Acc	UnAcc	UnAcc
A*24:05	UnAcc	UnAcc	Acc	UnAcc
A*24:06	UnAcc	UnAcc	UnAcc	UnAcc
A*24:07	UnAcc	UnAcc	UnAcc	UnAcc
A*24:08	Acc	UnAcc	UnAcc	UnAcc
A*24:10	UnAcc	UnAcc	UnAcc	Acc
A*24:13	UnAcc	UnAcc	UnAcc	UnAcc
A*24:14	UnAcc	UnAcc	UnAcc	UnAcc
A*24:15	UnAcc	UnAcc	UnAcc	UnAcc
A*24:17	UnAcc	UnAcc	UnAcc	UnAcc
A*24:18	UnAcc	UnAcc	UnAcc	Acc
A*24:20	UnAcc	UnAcc	UnAcc	UnAcc
A*24:22	UnAcc	UnAcc	UnAcc	Acc
A*24:23	UnAcc	UnAcc	UnAcc	Acc
A*24:25	UnAcc	UnAcc	UnAcc	UnAcc
A*24:26	UnAcc	UnAcc	UnAcc	UnAcc
A*24:28	UnAcc	Acc	UnAcc	UnAcc
A*24:30	UnAcc	UnAcc	UnAcc	UnAcc
A*24:32	UnAcc	UnAcc	UnAcc	UnAcc
A*24:33	UnAcc	UnAcc	UnAcc	Acc
A*24:53	UnAcc	UnAcc	UnAcc	UnAcc

Epitope	65QIA	76ESN	113H	163EW	180E
B*07:02*	Positive	Positive	Positive	Positive	Positive
B*07:03	Acc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:04	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:05	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:06	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:07	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:08	Acc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:09	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:10	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:12	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:13	Acc	Acc	UnAcc	UnAcc	UnAcc
B*07:14	UnAcc	UnAcc	Acc	UnAcc	UnAcc
B*07:16	Acc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:18	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:19	UnAcc	UnAcc	UnAcc	Acc	UnAcc
B*07:20	UnAcc	UnAcc	UnAcc	Acc	UnAcc
B*07:22	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:23	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:24	UnAcc	UnAcc	UnAcc	Acc	UnAcc
B*07:26	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*07:33	UnAcc	UnAcc	UnAcc	UnAcc	Acc
B*07:36	UnAcc	Acc	UnAcc	UnAcc	UnAcc

Epitope	65QIA	71ATD	80TLR	131S	163EW
B*27:05*	Positive	Positive	Positive	Positive	Positive
B*27:08*	Positive	Negative	Negative	Positive	Positive
B*27:01	UnAcc	Acc	UnAcc	UnAcc	UnAcc
B*27:02	UnAcc	Acc	Acc	UnAcc	UnAcc
B*27:03	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*27:04	UnAcc	Acc	UnAcc	UnAcc	UnAcc
B*27:06	UnAcc	Acc	UnAcc	UnAcc	UnAcc
B*27:07	UnAcc	UnAcc	UnAcc	Acc	UnAcc
B*27:09	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*27:10	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*27:12	Acc	Acc	Acc	UnAcc	UnAcc
B*27:14	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
B*27:15	UnAcc	Acc	UnAcc	UnAcc	Acc
B*27:18	Acc	Acc	Acc	UnAcc	UnAcc
B*27:20	UnAcc	Acc	UnAcc	Acc	UnAcc

For each epitope listed for a given allele group it shows the alleles that are acceptable (Acc) or unacceptable (UnAcc) mismatches. Alleles marked with asterisks are commonly used in assays

Table 2 Examples of HLA-DRB1 and HLA-DQB1 allele mismatch acceptability determined by epitopes specifically associated with serum antibody reactivity

Epitope	31FYY	70DA	70QT	67LQ+60Y
DRB1*04:01*	Positive	Negative	Positive	Positive
DRB1*04:02*	Positive	Positive	Negative	Negative
DRB1*04:03*	Positive	Negative	Positive	Positive
DRB1*04:04*	Positive	Negative	Positive	Positive
DRB1*04:05*	Positive	Negative	Positive	Positive
DRB1*04:06	Acc	Acc	UnAcc	UnAcc
DRB1*04:07	UnAcc	Acc	UnAcc	UnAcc
DRB1*04:08	UnAcc	Acc	UnAcc	UnAcc
DRB1*04:09	UnAcc	Acc	UnAcc	UnAcc
DRB1*04:10	UnAcc	Acc	UnAcc	UnAcc
DRB1*04:11	UnAcc	Acc	UnAcc	UnAcc
DRB1*04:13	UnAcc	Acc	UnAcc	UnAcc
DRB1*04:15	UnAcc	UnAcc	Acc	Acc
DRB1*04:17	UnAcc	Acc	UnAcc	UnAcc
DRB1*04:18	UnAcc	UnAcc	Acc	Acc

Epitope	11STS	31FYY	47F	70DA	73A	77T
DRB1*11:01*	Positive	Positive	Positive	Positive	Positive	Positive
DRB1*11:04*	Positive	Positive	Positive	Positive	Positive	Positive
DRB1*11:02	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*11:03	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*11:05	Acc	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*11:06	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*11:07	UnAcc	UnAcc	UnAcc	Acc	Acc	Acc
DRB1*11:08	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*11:09	UnAcc	Acc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*11:10	UnAcc	Acc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*11:11	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*11:13	UnAcc	Acc	UnAcc	Acc	UnAcc	UnAcc
DRB1*11:14	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*11:15	UnAcc	Acc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*11:17	UnAcc	Acc	Acc	Acc	UnAcc	UnAcc
DRB1*11:19	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc

Epitope	11STS	16H	70QT	67LQ+60Y	67LQ/R+73AT
DRB1*14:01*	Positive	Positive	Negative	Negative	Positive
DRB1*14:02*	Positive	Positive	Positive	Positive	Positive
DRB1*14:54*	Positive	Positive	Negative	Negative	Positive
DRB1*14:03	UnAcc	UnAcc	Acc	Acc	Acc
DRB1*14:04	Acc	Acc	Acc	Acc	UnAcc
DRB1*14:05	UnAcc	Acc	Acc	Acc	UnAcc
DRB1*14:06	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*14:07	UnAcc	UnAcc	Acc	Acc	UnAcc
DRB1*14:08	UnAcc	UnAcc	Acc	Acc	UnAcc
DRB1*14:10	Acc	UnAcc	Acc	Acc	UnAcc
DRB1*14:11	Acc	Acc	Acc	Acc	UnAcc
DRB1*14:12	UnAcc	UnAcc	Acc	Acc	Acc
DRB1*14:15	Acc	Acc	Acc	Acc	Acc
DRB1*14:16	UnAcc	UnAcc	Acc	Acc	Acc
DRB1*14:17	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*14:18	UnAcc	UnAcc	Acc	Acc	UnAcc
DRB1*14:19	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*14:20	UnAcc	UnAcc	UnAcc	UnAcc	UnAcc
DRB1*14:24	UnAcc	UnAcc	UnAcc	Acc	Acc

Epitope	56PV	85VA	87F
DQB1*06:01*	Negative	Positive	Positive
DQB1*06:02*	Negative	Positive	Positive
DQB1*06:03*	Negative	Positive	Positive
DQB1*06:04*	Positive	Negative	Negative
DQB1*06:09*	Positive	Negative	Positive
DQB1*06:05	UnAcc	Acc	Acc
DQB1*06:06	UnAcc	Acc	Acc
DQB1*06:07	Acc	Acc	UnAcc
DQB1*06:08	UnAcc	UnAcc	Acc
DQB1*06:10	Acc	UnAcc	UnAcc
DQB1*06:11	Acc	UnAcc	Acc
DQB1*06:12	UnAcc	Acc	UnAcc
DQB1*06:13	UnAcc	UnAcc	UnAcc
DQB1*06:14	Acc	UnAcc	Acc
DQB1*06:15	Acc	Acc	UnAcc
DQB1*06:16	Acc	UnAcc	Acc
DQB1*06:17	UnAcc	Acc	UnAcc
DQB1*06:18	UnAcc	UnAcc	UnAcc
DQB1*06:19	Acc	UnAcc	UnAcc

For each epitope listed for a given allele group it shows the alleles that are acceptable (Acc) or unacceptable (UnAcc) mismatches. Alleles marked with asterisks are commonly used in assays