Eplet Matching - Kidney Now or Later

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Overview

- HLA eplets
  - Premise
  - State of field
  - Potential application
  - Health Disparities

Lim Kid Int 2017
Structure and Polymorphism of HLA

- HLA molecules present peptides to T cells

- HLA class I is formed by a continuous polypeptide encoded by one gene
  - Expressed on every nucleated cell

- HLA class II proteins are heterodimers formed by the gene products of alpha (α) and beta (β) genes
  - Only expressed on antigen presenting cells and activated/inflamed cells like endothelium

**Numbers of HLA Alleles**

<table>
<thead>
<tr>
<th>Type</th>
<th>Number</th>
</tr>
</thead>
<tbody>
<tr>
<td>HLA Class I Alleles</td>
<td>25,228</td>
</tr>
<tr>
<td>HLA Class II Alleles</td>
<td>10,592</td>
</tr>
</tbody>
</table>
Sources of HLA Allosensitization

Other documented routes:
- Homograft (cardiac repair)
- Skin grafting
- Allogeneic cell therapy

Nonsensitizing clinical events:
- Immunization
- Infection
- Surgery
- Changes in immunosuppression
Evolution of HLA Typing and Compatibility

DNA (intermediate)

Antigen level
A36 vs. A2

Cross-reactive groups
A1 CREG vs. A2 CREG

Serology

What an antibody sees

<table>
<thead>
<tr>
<th></th>
<th>A</th>
<th>B</th>
<th>C</th>
<th>DRB1</th>
<th>DQB1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient</td>
<td>1</td>
<td>8</td>
<td>7</td>
<td>17</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>24</td>
<td>45</td>
<td>6</td>
<td>14</td>
<td>7</td>
</tr>
<tr>
<td>Donor</td>
<td>1</td>
<td>8</td>
<td>7</td>
<td>17</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>64</td>
<td>8</td>
<td>1</td>
<td>5</td>
</tr>
</tbody>
</table>

Mismatch: 3/6 ABDR Mismatch

14th Annual Living Donation Conference
Presented by the American Foundation for Donation and Transplantation
Association of antigen level mismatching with outcome

Gjertson NEJM 1991

Collaborative Transplant Study; Williams Transplant 2016
Evolution of HLA Typing and Compatibility

DNA (intermediate)

Antigen level
A36 vs. A2

DNA (high resolution)
Allele level
A*36:01 vs. A*02:01 vs. A*02:05

Serology
Cross-reactive groups
What an antibody sees

A1 CREG vs. A2 CREG

What a T cell sees

What a discriminating antibody sees
Degrees of difference

vs

vs
Degrees of difference

vs

vs
What are HLA Eplets?

HLA eplets are amino acid residues that are different between two alleles, that *may* represent a target for an antibody or T cell.

Tambur and Claas AJT 2015
HLA Allosensitization

**Self Immunizer**

- B*57:01:01:01
- B*07:02:01:01
- B*27:05:02:01
- B*40:01:01
- B*56:01:01:01

**Self B57**

- B*07:02
- B*07:03
- B*08:01
- B*14:01
- B*14:02
- B*14:05
- B*14:06
- B*15:03
- B*15:10
- B*15:18
- B*27:03
- B*27:05
- B*27:08
- B*38:01
- B*39:01
- B*39:05
- B*42:01
- B*48:01
- B*55:01
- B*56:01
- B*59:01
- B*67:01
- B*73:01
- B*81:01
- B*82:01
- B*82:02

**Paternal Antigen B7**

**Anti-HLA-B7**

Luminex Alleles of Epitope: 45EE

- B*07:02
- B*07:03
- B*08:01
- B*14:01
- B*14:02
- B*14:05
- B*14:06
- B*15:03
- B*15:10
- B*15:18
- B*27:03
- B*27:05
- B*27:08
- B*38:01
- B*39:01
- B*39:05
- B*42:01
- B*48:01
- B*55:01
- B*56:01
- B*59:01
- B*67:01
- B*73:01
- B*81:01
- B*82:01
- B*82:02
HLA Eplet vs. Antigen Mismatching

B57 vs. B7
1 antigen mismatch
= 4 eplet mismatch
AA Pos.
B*57:01:01:01
B*07:02:01:01

B57 vs. B62
1 antigen mismatch
= 3 eplet mismatch
AA Pos.
B*57:01:01:01
B*15:01:01:01

B57 vs. B58
1 antigen mismatch
= 2 eplet mismatch
AA Pos.
B*57:01:01:01
B*58:01:01:01
Clinical Utility of HLA Eplet Mismatching
HLA Eplet Mismatching: Keep in mind

Multiple algorithms
❖ Some in silico, others based on antibody verification
❖ Literature is a mix of approaches
❖ No consensus yet on which is the “best”

Requires high resolution typing
❖ Can be imputed from intermediate but imperfect accuracy, especially in non-Europeans [Engen AJT 2021]
❖ Larger studies from SRTR are based on imputation

Thresholds and relative risk are not fully defined
❖ How many is too many?
❖ Which mismatches are worse?
❖ Which outcome is more important?
Clinical Utility of HLA Eplet Mismatching

**Pre-transplant**
- equitable access
- DSA cross-reactivity
- unacceptable antigen

**Post-transplant**
- immunosuppression optimization
- rejection risk
- dnDSA risk
- graft longevity

**Retransplant**
- cPRA after graft failure

14th Annual Living Donation Conference
Presented by the American Foundation for Donation and Transplantation
Improving access to transplant

**chance of finding a compatible donor** for highly sensitized patients based on 0-2 triplet mismatches was much higher compared to matching on the antigen level [Duquesnoy Transpl 2003]

**reduced transplant wait time** for highly sensitized patients by 50% with comparable outcomes as non-sensitized patients [Eurotransplant; Lemieux Int Immuno 2021; Heidt Transpl 2019]

eplet based method of antibody analysis had a very high degree of **correlation with cell-based crossmatches** [Norin Hum Immun 2022]
Improving post-transplant outcomes

DR or DQ eplet mismatch was significantly correlated with production of dnDSA to that locus (OR 2.50 and 2.00 per 10MM) [Wiebe JASN 2017]

antibody-verified eplet mismatch load was associated with any type of rejection; but no threshold below which the risk of dnDSA occurrence was absent [Senev JASN 2020]

low-immunological risk recipients (0-2 ABDR mismatched kidneys) but with high eplet mismatches (≥20) had 2-fold increased risk of acute rejection [Nguyen Transpl Dir 2016]

significant relationship between eplet mismatches and graft failure; greater effect in unsensitized recipients [Sapir-Pichhadze Kid Int 2020]

discriminative performance for graft failure was low [Senev JASN 2020]

after dnDSA development in kidney transplant recipients, eplet mismatches did not correlate with ABMR or allograft loss [Wen Hum Immun 2021]
Tailoring immunosuppression based on risk

in CTOT-09 DQ mMM (>17) predicted dnDSA on **TAC minimization** despite patients being “low risk” (per sensitization) first 6 months post-transplant [Hricik JASN 2015]

recipients with high eplet mismatch load were **less likely to tolerate low tacrolimus** levels without developing de novo DSAs [Davis AJT 2021]

patients with high-risk DQ eplet mismatch score more frequently developed **acute rejection**, even if no pre-formed T cell alloimmunity was detected [Bestard AJT 2021]
Proposed Risk Stratification Model

<table>
<thead>
<tr>
<th>Pretransplant donor-recipient HLA laboratory evaluation</th>
<th>CDC crossmatch</th>
<th>Flow crossmatch</th>
<th>Single antigen bead</th>
<th>History of sensitization</th>
<th>HLA molecular MM</th>
<th>HLA identical</th>
<th>Immune risk assessment</th>
</tr>
</thead>
<tbody>
<tr>
<td>DSA positive</td>
<td>DSA positive</td>
<td>DSA positive</td>
<td>DSA positive</td>
<td></td>
<td></td>
<td></td>
<td>Active memory and at risk for hyperacute rejection</td>
</tr>
<tr>
<td>Negative</td>
<td>DSA positive</td>
<td>DSA positive</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Active memory and at risk for ABMR and TCMR</td>
</tr>
<tr>
<td>Negative</td>
<td>Negative</td>
<td>DSA positive</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Active memory and at risk for ABMR and TCMR</td>
</tr>
<tr>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
<td>Pregnancy or prior sensitization to repeat MM</td>
<td></td>
<td></td>
<td></td>
<td>At risk for latent memory with a recall B and T cell response</td>
</tr>
<tr>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
<td>cPRA with unknown repeat MM</td>
<td></td>
<td></td>
<td></td>
<td>Potential risk for latent memory with a recall B and T cell response</td>
</tr>
<tr>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
<td>No</td>
<td>High</td>
<td></td>
<td></td>
<td>Increased risk for de novo alloimmune response</td>
</tr>
<tr>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
<td>No</td>
<td>Low</td>
<td></td>
<td></td>
<td>Baseline risk for de novo alloimmune response</td>
</tr>
<tr>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
<td>No</td>
<td>0</td>
<td>Yes</td>
<td></td>
<td>Low risk for de novo alloimmune response</td>
</tr>
</tbody>
</table>

MM, Mismatch; DSA, donor-specific antibody; ABMR, antibody-mediated rejection; TCMR, T cell-mediated rejection.

“There is a body of evidence in support of the utility of HLA mMM score as a basis for primary alloimmunity risk stratification”
Proposed prognostic and predictive biomarker for clinical trials (under review by FDA)

The FDA Center for Drug Evaluation and Research agreed to evaluate the potential role of HLA-DR/DQ eplet mMM score as a strategy for enrichment or risk stratification in phase 2 and 3 kidney transplant clinical drug development trials and as a prognostic biomarker for de novo DSA, graft rejection, and graft failure.
Decreasing breadth of sensitization after graft failure

- cPRA after graft failure

highly sensitized patients had **lower rates of re-transplantation**, and higher rates of nephrectomy/graft intolerance syndrome [Singh Clin Transpl 2016]

after graft failure, **non-European recipients have a greater cPRA** than those of European ancestry [Tambur Kid Int 2021]
Decreasing breadth of sensitization after graft failure

clear relationship between the immunogenicity of donor HLA class I and class II mismatches and the development of HLA-specific antibodies after graft failure and relisting for transplantation [Kosmoliaptsis AJT 2016]

molecular mismatch scores were independently associated with degree of sensitization after graft failure [Kosmoliaptsis AJT 2016]

HLA eplet mismatch burden associated with higher risk of higher cPRA after renal allograft failure [Singh Clin Transpl 2016]
Equitable allocation and addressing health disparities

Higher rates of antigen-level mismatching in non-European recipients

Models eliminating antigen-level matching predicted increased rates of transplantation for non-European ancestry patients

…but quite offset by greater risk of rejection
Enhancing HLA matching for non-European patients

The current allocation system inadvertently matches Black patients to donors with significantly higher immunogenic transplants compared to other races.

Using race-adjusted immunogenicity thresholds in allocation would result in a net gain of thousands additional kidney life-years for all races [Bekbolsynov Front Immun 2022]

HLA class I eplet load greater than 70 resulted in a greater risk of rejection in the race-mismatched pediatric transplants vs. race-matched [Philogene Ped Nephrol 2019]
Selecting a “better matched” mismatched donor

By molecular analysis, the patient’s DR18 is more similar to DR17, which is very common across donor populations

- Better chance at finding a good “match”
- Lower risk of dnDSA
- Better graft survival
- Lower breadth of sensitization if graft fails
Practical Applications of HLA Eplet Mismatching
Epitopes for Antibody Analysis: Unacceptable Antigen Entry and VXM

Donor: DPB1*105:01

Recipient antibodies: DPB1*02:01, 04:01, 04:02

Predict DSA to DPB1*105:01 based on crossreactive epitopes
Eplets for Allocation?

NKR is currently employing high-resolution genotyping to improve molecular matching and blood typing (for A subtyping), and these efforts may be particularly important for compatible pairs entering paired exchange.
Current Consensus on HLA eplet mismatching

1. Approaches need to be optimized and algorithms standardized and locked before implementation in clinical practice.

2. Thresholds for risk categories need to be established and the impact of other factors on these thresholds need to be accounted for. Formal evaluation, in prospective clinical trials, should be performed.

3. Tools to prospectively determine donor/recipient HLA specific immunogenicity beyond the mismatch load (given that DSA can be developed in some patients with low HLA mMM score) should be developed.

4. This is essential before considering evaluation and implementation of immunogenicity analysis as a guide to organ allocation schemes.
Takeaways

Future Applications:

- Individualized immunologic risk assessment in subpopulations to achieve maximum survival benefit
  - Highly sensitized
  - Pediatric/allograft longevity
- Equitable allocation
- Desensitization efficacy
- Optimization of immunosuppression for retransplant candidates
Thank you

Questions?

“I'D TAKE THE AWE OF UNDERSTANDING OVER THE AWE OF IGNORANCE ANY DAY”

Douglas Adams
Excellent Reviews

• https://www.sciencedirect.com/science/article/pii/S0198885921002925
## Evidence for Clinical Utility

<table>
<thead>
<tr>
<th>Application</th>
<th>Correlation</th>
<th>Reference</th>
</tr>
</thead>
</table>
| **Predict dnDNA risk**                          | DR or DQ eplet mismatch was significantly correlated with production of dnDSA to that locus (OR 2.50 and 2.00 per 10MM)  
There was no threshold below which the risk of dnDSA occurrence was absent | Wiebe JASN 2017  
Senev JASN 2020 |
| **Identify compatible donors for highly sensitized candidates** | Reduced transplant wait time by 50% with comparable outcomes as non-sensitized patients | Eurotransplant; Lemieux Int Immuno 2021; Heidt Transpl 2019 |
| **Predict graft loss**                          | Graft survival comparable 0-2 triplet mismatch recipient and 0 AB antigen mismatches  
1.23-1.41 HR per 10 MM                                                                                   | Eurotransplant  
Sapir-Pichhadze Kid Int 2020 |
| **Predict rejection risk**                      | primarily eplet mismatch load on the DQ molecule (OR 1.06)  
discriminative performance for graft failure was low                                                   | Senev JASN 2020 |
| **Risk of rejection after dnDSA formed?**       | once a patient has developed de novo DSA, eplet mismatches did not correlate with ABMR or death-censored allograft loss | Wen Hum Immun 2021 |
| **Breadth of sensitization after graft failure**| HLA eplet mismatch burden associated with higher risk of higher cPRA after renal allograft failure    | Kosmoliaptsis AJT 2016  
Singh Clin Transpl 2016 |
| **Risk stratification and/or biomarker in clinical trials** | FDA agreed to evaluate DR-DQ eplet mMM score for enrichment or risk stratification in phase 2 and 3 transplant clinical trials and as a prognostic biomarker for de novo DSA, graft rejection, and graft failure. | Wiebe AJT 2019 |
Session Survey

Nicole Valenzuela, PhD, Suzanne McGuire, RN, BSN, CCTC, and David Serur, MD | April 20th 8:45 AM-9:30 AM