HLA-DRB1*03:05 AT THE ANTIGEN EQUIVALENCY CROSSROADS

Nezar Eltayeb Elsheikh[1,2], Carrie L. Butler [1,2], Anh Du [1,2], Michelle Hickey[1,2], Nicole Valenzuela[1,2], Elaine F. Reed[1,2], Qiuheng Zhang[1,2], Rebecca Sosa[1,2]

[1]Department of Pathology and Laboratory Medicine and [2]Immunogenetics Center, University of California, Los Angeles, CA, USA

https://www.instagram.com/immunogenetics_la

Background

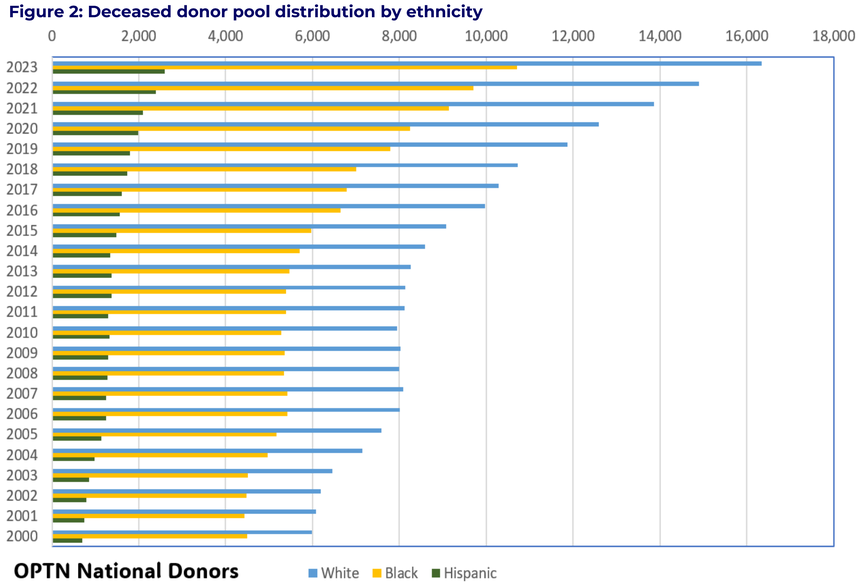
HLA molecular typing must be converted to antigen equivalency and entered into the United Network for Organ Sharing (UNOS) system. Two different serological equivalencies were assigned to the molecular typing of HLA-DRB1*03:05 (**Figure 1**) that could result in variable entries in UNOS.

Figure 1: HLA-DRB1*03:05 by HLA Dictionary

Confirmatory Data			
Sequence Status:	Complete CDS	Confirmed:	Confirmed
Cells:	5	Laboratories:	4
HLA Dictionary			
Expert Assigned Type:	DR3	WHO Assigned Type:	DR3
Neural Network Assigned Type:		DR17	

These discrepancies could result in variable entries into UNOS system. The 2023 OPTN national data shows that non-Hispanic Whites (CAU) represent the majority of the deceased donors followed by Blacks (AFA) and Hispanics (**Figure 2**). It was demonstrated that the most common carriers of DRB1*03:01 and DRB1*03:02 are CAU and AFA respectively. Blocking the DR3 broad group in UNET could potentially limit access to donors. While this may reduce risks for patients, it could also disadvantage recipients awaiting transplants by excluding the most common haplotypes in CAU and AFA populations.

Figure 2: Deceased donor pool distribution by ethnicity



AIM

We aimed to investigate the discrepancies in the HLA-DRB1*03:05 antigen equivalency report.

METHODS

We screened 705 samples with DR17 and DR18 reactivity using single antigen bead (SAB) class II test and found comparable MFI strengths amongst the two DR17 and DR18 beads. We identify a DR17 and DR18 positive serum with approximate MFI strengths of 7200 and 5100 respectively.

We performed SAB test for class II at neat, 1:2, and 1:4 dilutions to determine if they react differently.

We performed surrogate flow cytometry crossmatch (SXM) with an HLA-DRB1*03:05 donor and compared to results from donors with HLA typing of DRB1*03:01 and DRB1*03:02.

RESULTS

The mature protein sequence alignment for DRB1*03:05, DRB1*03:01 (DR17) and DRB1*03:02 (DR18) reveals that DRB1*03:05 differs from DR17 only at position 86 (glycine vs valine). In contrast, DR18 has phenylalanine, glutamic acid, and tyrosine at positions 26, 28, and 47 (**Figure 3**).

Figure 3: Alignment for HLA-DRB1*03:05, DR17, and DR18

```
AA Pos.                 10        20        30        40        50        60        70        80        90       100
DRB1*03:01:01:01  GDTRPRFLEY STSECHFFNG TERVRFLDRY FHNQEENVRF DSDVGEFRAV TELGRPDAEY WNSQKDLLEQ KRGRVDNYCR HNYGVVESFT VQRRVHPKVT
DRB1*03:02:01:01  ---------- ---------- -----F-E-- ---------- -----Y---- ---------- ---------- ---------- -----G---- ----------
DRB1*03:02:01:02  ---------- ---------- -----F-E-- ---------- -----Y---- ---------- ---------- ---------- -----G---- ----------
DRB1*03:05:01     ---------- ---------- -----F-E-- ---------- -----Y---- ---------- ---------- ---------- -----G---- ----------

AA Pos.                110       120       130       140       150       160       170       180       190       200
DRB1*03:01:01:01  VYPSKTQPLQ HHNLLVCSVS GFYPGSIEVR WFRNGQEEKT GVVSTGLIHN GDWTFQTLVM LETVPRSGEV YTCQVEHPSV TSPLTVEWRA RSESAQSKML
DRB1*03:02:01:01  ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
DRB1*03:02:01:02  ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
DRB1*03:05:01     ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

AA Pos.                210       220       230
DRB1*03:01:01:01  SGVGGFVLGL LFLGAGLFIY FRNQKGHSGL QPRGFLS
DRB1*03:02:01:01  ---------- ---------- ---------- -------
DRB1*03:02:01:02  ---------- ---------- ---------- -------
DRB1*03:05:01     ---------- ---------- ---------- -------
```

We found that the reactivity remained similar between the DR17 and DR18 beads across all dilutions (**Table 1**), implicating minimal variation between the two antigens.

Table 1: SAB and SXM Results

	SAB Neat	SAB 1:2 MFI	SAB 1:4 MFI
DR17	7,200	3,200	3,100
DR18	5,100	2,800	2,600
DRB1*03:05 T-cell	Negative	Negative	Negative
DRB1*03:05 B-cell	Positive	Negative	Negative
DRB1*03:01 T-cell	Negative	Negative	Negative
DRB1*03:01 B-cell	Positive	Positive	Negative
DRB1*03:02 T-cell	Negative	Negative	Negative
DRB1*03:02 B-cell	Positive	Positive	Negative

DISCUSSION

The antigen equivalency to HLA molecular typing must be entered into UNOS system based on the World Marrow Donor Association (WMDA) antigen table, the HLA Dictionary, UNOS-provided tables, and the Organ Procurement and Transplantation Network (OPTN) conversion guidelines. However, discrepancies in data from these sources can lead to variability in converting certain alleles.

Our results show that DR17 and DR18 are diluted out at a similar rate when tested with SAB II test. Crossmatch results have shown no discernible differences between DR17 and DR18 when surrogated with DRB1*03:05 cells. Although more testing is needed to draw a conclusion, these results are partially supporting the recent decision of the OPTN Histocompatibility Committee to make DR3 equivalent to itself. However, calling DR3 might block access to a large portion of donors causing more wait time and delayed transplantations.

Blocking the most common haplotypes among Caucasians (A1 B8 DR17) and African Americans (A30 B42 DR18) that are bearing DR17 and DR18 might largely minimize access to many donors.

CONCLUSION

Blocking the broad group antigen for DR17 and DR18 is considered safe for patients, but disadvantageous for minorities' recipients awaiting solid organ transplants. More research is needed to improve our testing tools to differentiate DR17 from DR18 to accurately convert molecular typing to antigen level.