Pre-Transplant Panel Reactive Antibodies and HLA-A*/DQB1* Mismatches Associated With the Development of de novo Donor Specific Antibody After Lung Transplant

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Background

- Major histocompatibility complex (MHC) determines the compatibility of donors for organ transplant.
- In humans, the MHC complex is known as the human leukocyte antigen (HLA), and it is responsible for discriminating between self and non-self cells within the human body.
- HLA genes have a high degree of polymorphism including the six main loci HLA-A*/B*/C*/DQ*/DR*/DP*, each with several distinct alleles
- The main cause of graft failure is antibody mediated rejection (or AMR), which is caused by the presence of *de novo* donor specific antibodies (dDSA), or antibodies that do not preexist but develop post-transplant to fight against foreign cells from the transplanted organs.

Hypothesis and Purpose

- The incidence of HLA dDSA after lung transplant is prevalent and related to negative transplant outcomes such as antibody mediated rejection.
- Once a patient is highly sensitized with DSA, it is very difficult to treat even with immunosuppressant drugs.
- In the first large cohort study of its kind, we aimed to assess pre-transplant predictors that were associated with the development of dDSA after lung transplants. If a reliable pre-transplant factor could be identified, then it could also be used to more safely and effectively match organ donations to potential recipients.
- We hypothesized that at least one pre-transplant factor would be related to the appearance of dDSA.

Methodology

- We retrospectively recorded the development of de novo DSA among 422 recipients who underwent lung transplants between October of 2012 and December of 2016.
- Data were collected about recipient demographic characteristics such as gender, age, and race.
- Data on more immunological characteristics were also collected such as blood type, panel reactive antibody (PRA), the mean fluorescence intensity (MFI) of dDSA at the time of pretransplant, and donor/recipient mismatches of 0, 1, and 2 in each of HLA-A, B, C, DQ, DR, and DP loci were analyzed for the development of DSA.

Data

DSA	Sex	Race	Age	ABO	Pre-Tx cPRA Total	Pre-Tx cPRA Total St	Pre-Tx cPRA CI	Pre-Tx cPRA CI St	Pre-Tx cPRA CII	Pre-Tx cPRA CII St	Pre-Tx MFI Class I	Pre-Tx MFI Class II	A DSA Y or N	# of A mm 0/1/2	B DSA Y or N	# of B mm 0/1/2	C DSA Y or N	# of C mm 0/1/2	DR DSA Y or N	# of DR mm 0/1/2	DQB1 DSA Y or N	# of DQB1 mm 0/1/2
no	F	WHITE	48	O	0	0	0	0	0	0	NA		n		y	2	y	2	n	2	y	2
no	M	WHITE	64	A	0	0	0	0	0	0			n	na	n	na	n	na	n	na	n	na
yes	F	WHITE	57	A	92	20	91	2	19	19	NA		y	2	y	2	n	2	n	2	n	1
yes	F	WHITE	38	A	0	0	0	0	0	0	NA		y	2	n	2	n	2	n	1	y	1
no	M	WHITE	55	A	0	0	0	0	0	0			n	1	n	2	n	2	n	2	n	2
no	M	WHITE	60	O	22	22	0	0	22	22			n	1	n	2	n	0	n	0	n	0
yes	M	WHITE	66	O	40	0	40	0	0	0	NA		n	2	n	1	n	1	n	2	y	2
yes	M	WHITE	65	O	0	0	0	0	0	0	NA		n	2	n	1	n	1	n	2	y	2
yes	M	WHITE	62	O	0	0	0	0	0	0		G0	n	2	n	2	n	2	n	2	y	2
no	M	WHITE	66	A	23	0	23	0	0	0			n	1	n	1	n	1	n	2	n	1
no	M	WHITE	73	O	22	0	5	3	19	0			n	2	n	2	n	2	y	2	y	2
yes	M	WHITE	66	O	0	0	0	0	0	0		G1	n	2	n	2	n	2	n	2	y	2
yes	M	WHITE	70	O	0	0	0	0	0	0	G1	G1	y	2	n	2	n	2	n	2	y	2
yes	M	WHITE	66	A	0	0	0	0	0	0		G1	n	1	n	2	n	2	y	2	y	2
yes	M	WHITE	73	O	0	0	0	0	0	0	G1	G1	n	2	y	2	n	1	n	2	y	2
no	M	WHITE	51	O	43	0	43	0	0	0			n	0	n	2	n	1	n	2	n	1
yes	M	WHITE	23	A	3	0	3	0	0	0	G1	G0	n	1	y	2	n	2	n	2	y	2
yes	M	WHITE	66	A	13	0	0	0	13	0		G1	n	1	n	2	n	2	n	2	y	2
no	F	WHITE	30	O	0	0	0	0	0	0			n	1	n	2	n	2	n	2	n	2
no	M	WHITE	53	A	0	0	0	0	0	0			n	0	n	1	n	0	n	2	n	2
yes	M	BLACK	63	O	5	0	5	0	0	0	G1		y	2	n	2	n	2	n	2	n	2
no	M	WHITE	75	AB	21	0	21	0	0	0			n	2	n	2	n	2	n	1	n	1
yes	M	WHITE	58	O	0	0	0	0	0	0	G0	G0	n	2	y	1	n	1	n	2	y	2
no	M	WHITE	68	A	0	0	0	0	0	0			n	2	n	2	n	2	n	1	n	1
no	F	WHITE	72	O	0	0	0	0	0	0			n	0	n	2	n	2	n	2	n	2
no	F	WHITE	61	O	17	0	17	0	0	0			n	0	n	2	n	1	n	2	n	1
no	M	WHITE	70	B	0	0	0	0	0	0			n	1	n	2	n	2	n	0	n	0
no	M	WHITE	65	A	0	0	0	0	0	0			n	2	n	1	n	1	n	1	n	1
no	F	WHITE	53	A	0	0	0	0	0	0			n	1	n	2	n	2	n	2	n	1
no	F	WHITE	59	A	0	0	0	0	0	0			n	1	n	2	y	1	n	2	n	2
no	F	WHITE	58	O	0	0	0	0	0	0			n	2	n	2	n	2	n	1	n	0
yes	M	WHITE	67	A	50	0	0	0	50	0		G0	n	2	n	2	n	2	n	2	y	2
no	F	WHITE	21	O	10	2	10	2	0	0			n	0	n	2	n	2	n	2	n	1

*Partial Data. Full data set is available in Allogen Lab database upon request.

- Data collection included 422 patients, but due to the large size of the dataset, only part of the data sheet is shown.
- Over 26 factors were recorded for each patient on each locus of HLA (the size of the data sheet spanned 233 columns wide).

Results

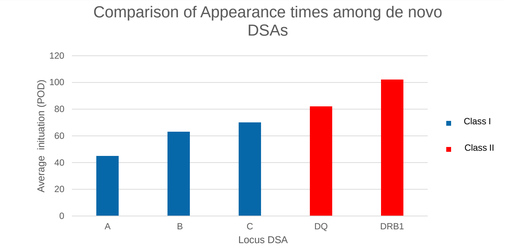

Figure 1. The comparisons of the average post operation days for dDSA occurrence. Among all loci evaluated, HLA-A was the quickest to appear, and HLA-DR was the slowest.

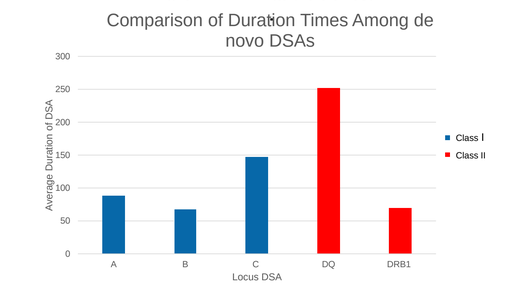

Figure 2. The comparisons of the average post-operation days for dDSA to be resolved. Among all loci evaluated, HLA-B was the quickest to be resolved, and HLA-DQ was the slowest.

Table 1. Association of Recipient Demographic Characteristics and Pre-Transplant Histocompatibility Predictors to the Development of De Novo Donor Specific Antibody

	Total (n= 422)	*dDSA Positive (n=222)	dDSA Negative (n=200)	p value
Gender				
Male [n (%)]	300 (71.1)	155 (69.8)	145 (72.5)	
Female [n (%)]	122 (28.9)	67 (30.2)	55 (27.5)	
Race				
Caucasian [n (%)]	376 (89.1)	192 (86.5)	184 (92.0)	
African American [n (%)]	32 (7.6)	20 (9.0)	12 (6.0)	
Others [= (%)]	14 (3.3)	10 (4.5)	4 (2.0)	
Recipient Age				
<40 (n, x±SD of years)	(55, 28±5)	(33, 29±6)	(22, 26±5)	
40-65 (n, x±SD of years)	(218, 57±6)	(119, 56±6)	(99, 57±6)	
>65 (n, x±SD of years)	(149, 69±3)	(70, 69±3)	(79, 70±3)	0.053
Pre-transplant Total **PRA ≥ 30% [n (%)]	62 (15.0)	40 (18.4)	22 (11.0)	<0.05
Blood Group (A/B/AB/O)	164/48/19/191	83/24/8/107	81/24/11/84	
***MFI of dDSA at Pre-transplant Class I				
0-500 MFI [n (%)]	387 (91.7)	197 (92.1)	190 (94.1)	
> 500 < 1000 [n (%)]	29 (6.9)	17 (7.9)	12 (5.9)	
Missing [n (%)]	6 (1.4)	NA	NA	
***MFI of dDSA at Pre-transplant Class II				
0-500 MFI [n (%)]	335 (79.3)	179 (81.7)	156 (79.6)	
> 500 < 1000 [n (%)]	80 (19.0)	40 (18.3)	40 (20.4)	
Missing [n (%)]	7 (1.7)	NA	NA	
***HLA Mismatches (0/1/2 MM)				
HLA-A* [Total n (n of 0/1/2 MM)]	412 (9/99/304)	85 (0/5/80)	327 (9/94/224)	<0.001
HLA-B* [Total n (n of 0/1/2 MM)]	419 (1/55/358)	69 (0/5/64)	350 (1/50/294)	
HLA-C* [Total n (n of 0/1/2 MM)]	418 (5/106/305)	38 (1/3/34)	380 (6/103/271)	
HLA-DR81* [Total n (n of 0/1/2 MM)]	420 (8/69/343)	44 (1/2/41)	376 (7/67/302)	
HLA-DQB1*[Total n (n of 0/1/2 MM)]	418 (23/109/286)	155 (2/6/147)	263 (21/103/139)	<0.001
HLA-DPB1* [Total n (n of 0/1/2 MM)]	242 (167/15/60)	1 (1/0/0)	241 (166/15/60)	

* De novo Donor Specific Antibody
** Panel Reactive Antibody
*** Some individuals missing data for scoring

Results

- Previous studies showed that dDSAs that appear earlier and last longer were the most harmful to transplants.
- Among 6 studied loci, HLA-A* and DQB1* mismatches were remarkably associated with the occurrence of dDSA ($p<0.001$).
- dDSA development from the recipients with ≥30% pre-transplant PRA was significantly more than that from the individuals with <30% PRA regardless of dDSA MFI values at pre-transplant time ($p<0.05$).
- A weak relationship between recipient age and dDSA presence was observed, but was not statistically significant.
- No obvious correlations were found between dDSA and other evaluated factors (Table 1).

Conclusions

- Mismatches in HLA-A and HLA-DQ loci were associated with dDSA production after LuTx.
- PRA>30% was associated with dDSA production after LuTx.
- Both of these factors should be considered as risk factors and warrant closer post transplant monitoring.

Recommendations

- This is a single center study, and the study results warrant multiple center study results to further confirm our own.

References

- Heeger, Peter S, and Rajani Dinavahi. “Transplant Immunology for Non-Immunologist.” *Mount Sinai Journal of Medicine*, vol. 79, 2012, pp. 376–387.
- Rodey, Glenn E. *HLA Beyond Tears: Introduction to Human Histocompatability*. 2nd ed., Durango, CO, De Novo Inc., 2000.
- Sun, Qiquan, et al. “Immunology of Transplantation.” *Primer on Transplantation*, 3rd ed., Columbus, OH, 2011, pp. 1–17.