Lecture 4

Registry of HLA epitopes and HLAMatchmaker analysis for antibody-verified epitopes

16th Workshop Project: International HLA Epitope Registry

GOALS:

- Develop a notation system for epitopes encoded by HLA-ABC, DRB1/3/4/5, DQAB, DPAB and MICA
- Establish a database of antibody-verified HLA epitopes
- Offer clinically relevant search functions

The registry does not address so-called cellular HLA epitopes defined by alloreactive T-cells

The Website-Based International Registry of Antibody-Verified HLA Epitopes

Int J. Immunogenetics, 40: 54-59

http://www.epregistry.com

Developed and managed by

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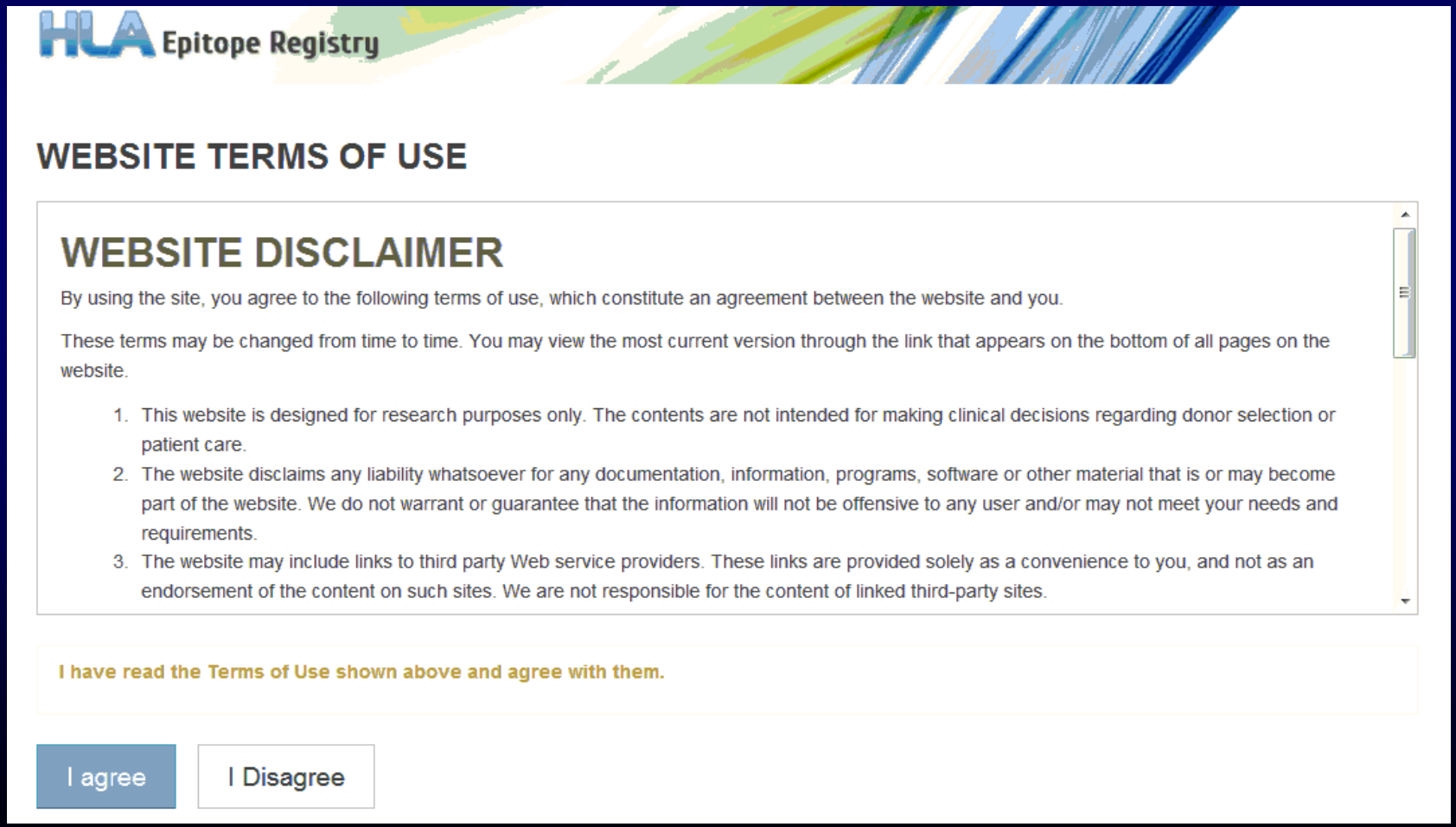
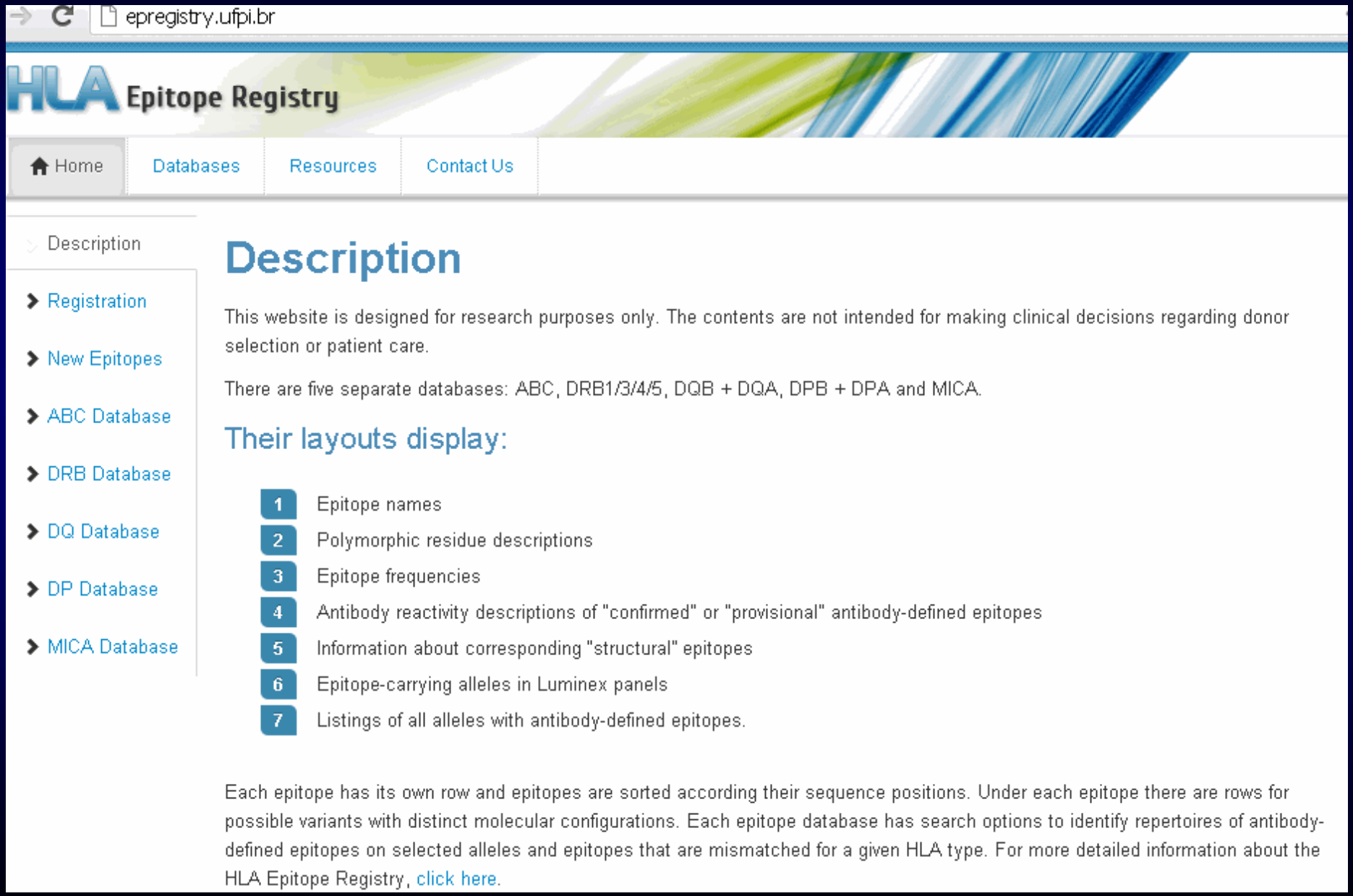
HLA Epitope Registry: www.epregistry.ufpi.br

(can be viewed in Microsoft Internet Explorer,
Google Chrome or Mozilla Firefox browsers)

- Website information is for research use only. Users must agree to terms of use before using website:



Registry Home Page



Class I Eplet Examples

Eplet	Description	Eplet-Carrying Alleles in Luminex Panel
62GE	62G,63E,65R	A*02:01/02/03/05/06,B*57:01/03,*58:01
166DG	166D,167G	A*01:01,*23:01,*24:02,*80:01,B*15:12
163EW	162G163E166E167W	A*66:02,B*07:02*13:01/02,*27:05/08,*40:01/02/06,*47:01,*48:01,*73:01,*81:01,C*02:02,*17:01
$44KM_3$	43Q44K45M46E / 149A150V151H152A / 158V	A*01:01, A*36:01

Locus	Eplet	Description	Eplet-Carrying Alleles in Luminex Panel
MICA	24T	24T, 26V,36C	A*001,*012,*018
MICA	175S	173E,175S	A*004,*006,*009,*016,*019,*033,*036

Class I ABC Database

Databases

ABC | DRB | DQB + DQA | DPB + DPA | MICA

Filters

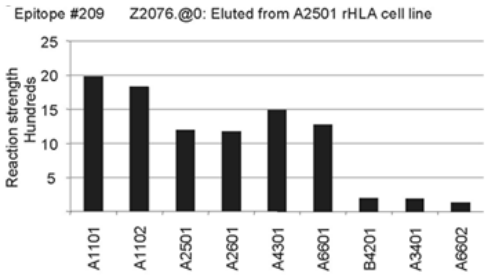
Epitope	Polymorphic Residues	Frequency	Antibody Reactivity	Struct Epitope	Luminex Alleles	All Alleles
163LE	163L166E				B*15:01, B*15:02, B*15:03, B*15:10, B*15... More	A*26:04, B*07:2(B*07:24, B*07:6(B*08... More
163R	163R		Provisional	View	A*01:01, A*11:01, A*11:02, A*25:01, A*26... More	A*01:01, A*01:0: A*01:03, A*01:0(A*01... More
163RD	162G163R166D167G		Confirmed	View	A*01:01 More	A*01:01, A*01:0: A*01:03, A*01:0(A*01... More
163RE	162G163R166E167W		Confirmed	View	A*11:01, A*11:02, A*25:01, A*26:01, A*43... More	A*02:101, A*02:135, A*02:154, A*02:38, A...

Antibody Reactivity Report for 163RE

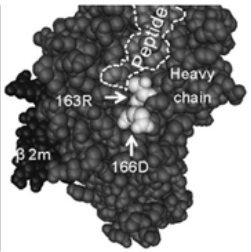
Epitope:	163RE (confirmed)
Luminex single allele reactivity:	**Human monoclonal antibody VP5G3 (IgM) from Arend Mulder (Leiden):** Sensitization due to pregnancy. Antibody producer types as HLA-A24, A32; B18, B60; Cw10, Cw7. Immunizing haplotype: HLA-A26; B55 (Bw6); Cw1 Data supplied by Marilyn Marrari (Pittsburgh):

PANEL	OL MFI	GP MFI
Positive control	nt	nt
Negative control	9	5
163RE+ A*11:01	11150	8163
163RE+ A*11:02	12567	4246
163RE+ A*25:01	13029	7194
Immunizing allele: 163RE+ A*26:01	12521	9514
163RE+ A*43:01	7510	1165
163RE+ A*66:01	12498	11044
Self Alleles	21 ± 11	9 ± 4
Other 163RE-negative alleles	84 ± 278	9 ± 3

Data from El-Awar et al, *Human Immunology* 68: 170-180, 2007: Serum eluate Z2076.@0 (absorbed with A*25:01 cells), no information on HLA types of antibody producer/immunizer



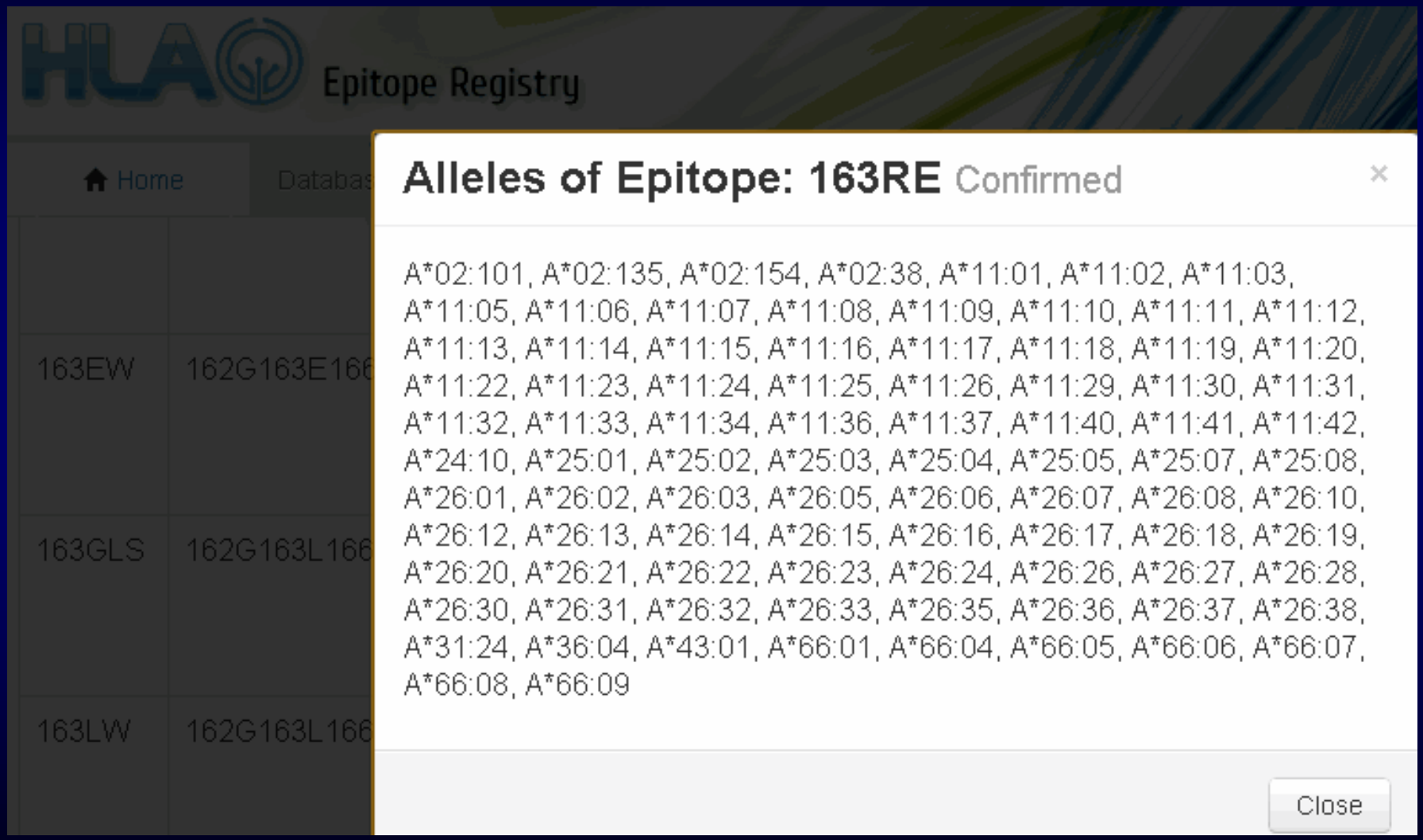
Antigen	aa position 163	166
A0101	R	D
A1101	R	E
A1102	R	E
A2501	R	E
A2601	R	E
A4301	R	E
A6601	R	E
B4201	T	E
A3401	T	E
A6602	E	E



Other antibody assays:	**Data from Mulder et al, *Tissue Antigens* 52: 393-396, 1998:** VP5G3 reacts with HLA-A11, A25, A26, A66 in CDC assays; A43 not tested. **Data from Fernandez-Vina et al, in Immunobiology of the Human MHC vol. I, John A. Hansen (Ed), pp 890-931, 2006:** VP5G3 (13-WS 0061) positive with HLA-A*11:01, *11:02, *11:03, *24:10, *25:01, *25:02, *26:01, *26:02, *26:03, *26:08, *26:15, *66:01 in CDC assays in 13th IHWS. No HLA-A43 cells were tested.
Comments:	Equivalent to TerEp #209. Epitope is not shared by HLA-A*66:02, but is shared by *24:10, which was positive in CDC assays.

Epitope-sharing Luminex alleles

All Alleles that Share Epitope



Select a Database to View - DRB

Databases

ABC | **DRB** | DQB + DQA | DPB + DPA | MICA

› Filters

Epitope	Polymorphic Residues	Frequency	Antibody Reactivity	Struct Epitope	Luminex Alleles	All Alleles
4Q	4Q		Provisional		DRB1*07:01, DRB1*09:01, DRB4*01:01, D... More	DRB1*07:01, DRB1*07:09, DRB1*09:01, D... More
4R	4R				DRB1*01:01, DRB1*01:02, DRB1*01:03, D... More	DRB1*01:01, DRB1*01:02, DRB1*01:03, D... More
6C2	6C(157I)				DRB5*02:02 More	DRB1*13:112, DRB1*13:128, DRB5*02:02,... More
6R2	6R(157T)				DRB1*01:01, DRB1*01:02, DRB1*01:03, D... More	DRB1*01:01, DRB1*01:02, DRB1*01:03, D... More
12TSE	12T13S14E		Confirmed		DRB1*03:01, DRB1*03:02, DRB1*03:03, D... More	DRB1*03:01, DRB1*03:02, DRB1*03:03, D... More
13FE	13F14E		Provisional		DRB1*01:01, DRB1*01:02, DRB1*01:03, D... More	DRB1*01:01, DRB1*01:02, DRB1*01:03, D... More
13FEL	13F14E26L		Provisional		DRB1*01:01, DRB1*01:02, DRB1*01:03, D... More	DRB1*01:01, DRB1*01:02, DRB1*01:03, D... More

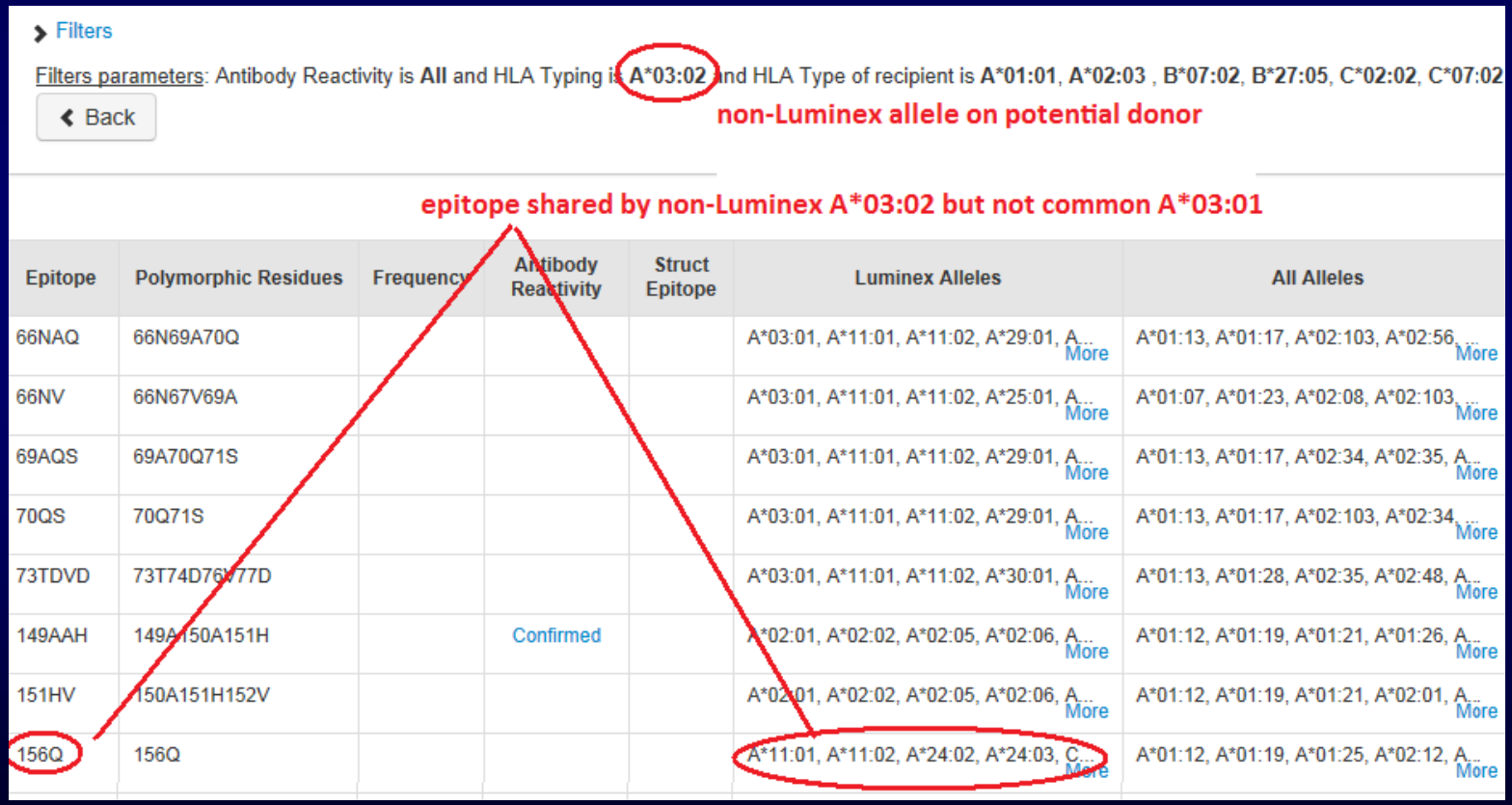
Epitope Database Search Options

1. Identify the epitope repertoire of any selected allele (Luminex or non-Luminex)
   - Choose antibody-verified or all epitopes
2. Identify all mismatched epitopes for a given HLA phenotype
   - Choose antibody-verified or all epitopes

These options can be combined to identify a donor allele's repertoire of epitopes that are mismatched for a recipient's HLA type

Identifying epitopes on non-Luminex alleles mismatched for a patient

Enter the non-Luminex allele of interest (e.g., donor) in the “Search by allele” field and the patient HLA type in the “HLA type of recipient” field; results show one epitope on non-Luminex A*03:02 that is not shared by the common A*03:01 found in the Luminex panels.

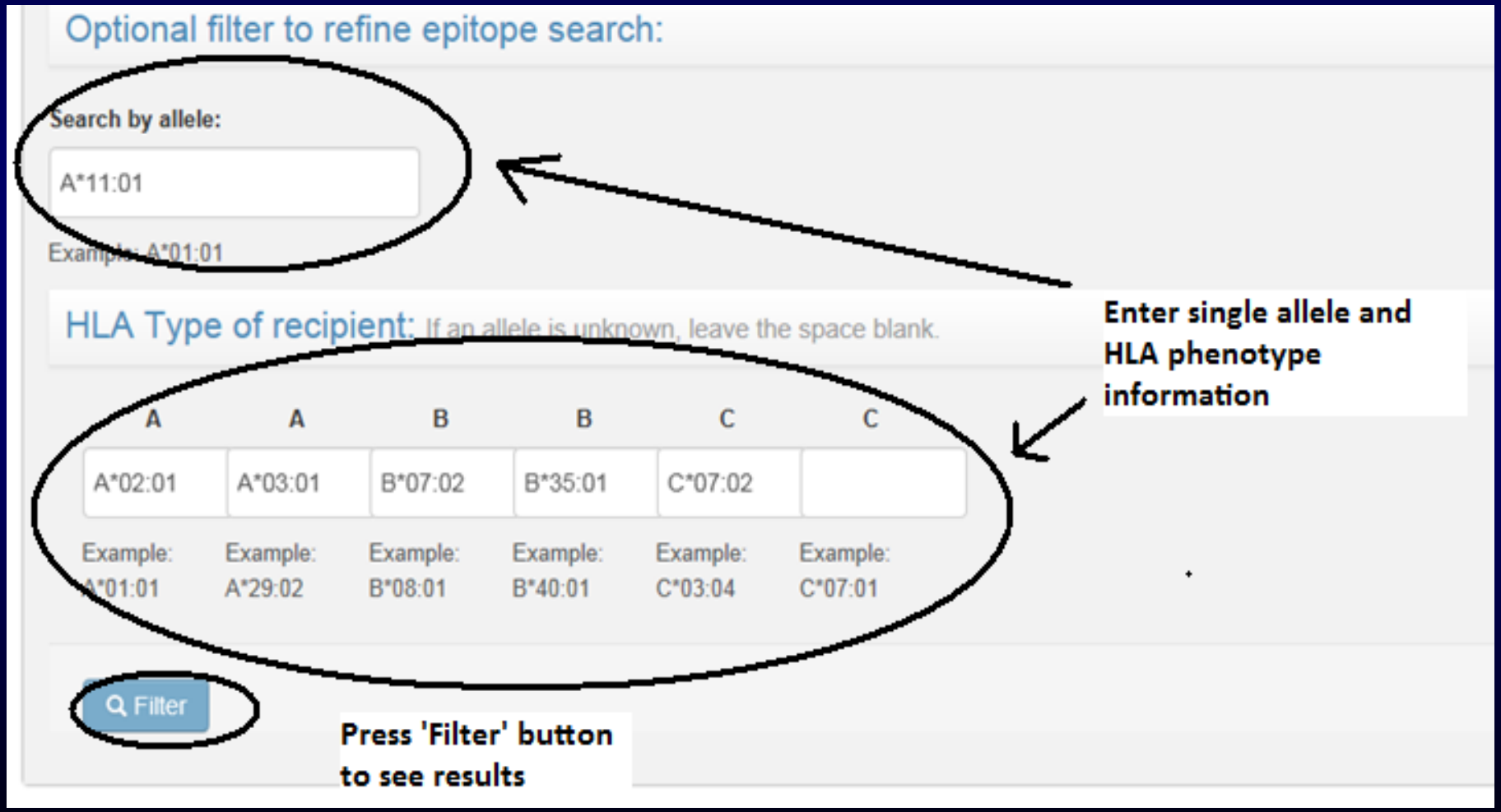


Epitope	Polymorphic Residues	Frequency	Antibody Reactivity	Struct Epitope	Luminex Alleles	All Alleles
66NAQ	66N69A70Q				A*03:01, A*11:01, A*11:02, A*29:01, A... More	A*01:13, A*01:17, A*02:103, A*02:56, ... More
66NV	66N67V69A				A*03:01, A*11:01, A*11:02, A*25:01, A... More	A*01:07, A*01:23, A*02:08, A*02:103, ... More
69AQS	69A70Q71S				A*03:01, A*11:01, A*11:02, A*29:01, A... More	A*01:13, A*01:17, A*02:34, A*02:35, A... More
70QS	70Q71S				A*03:01, A*11:01, A*11:02, A*29:01, A... More	A*01:13, A*01:17, A*02:103, A*02:34, ... More
73TDVD	73T74D76V77D				A*03:01, A*11:01, A*11:02, A*30:01, A... More	A*01:13, A*01:28, A*02:35, A*02:48, A... More
149AAH	149A150A151H		Confirmed		*02:01, A*02:02, A*02:05, A*02:06, A... More	A*01:12, A*01:19, A*01:21, A*01:26, A... More
151HV	150A151H152V				A*02:01, A*02:02, A*02:05, A*02:06, A... More	A*01:12, A*01:19, A*01:21, A*02:01, A... More
156Q	156Q				A*11:01, A*11:02, A*24:02, A*24:03, C... More	A*01:12, A*01:19, A*01:25, A*02:12, A... More

If screening results show antibody reactivity towards epitope 156Q, A*03:02 is likely an unacceptable allele!

Combining Options

Use Filters 2 and 3 in combination to display epitopes on an allele that are mismatched for a given HLA type, e.g., to identify epitope mismatches for a patient's HLA phenotype that are found on a specific donor allele.



Results of combined options

› Filters

Filters parameters: Antibody Reactivity is **All** and HLA Typing is **A*11:01** and HLA Type of recipient is **A*02:01, A*03:01, B*07:02, B*35:01, C*07:02**

‹ Back

Epitope	Polymorphic Residues	Frequency	Antibody Reactivity	Struct Epitope	Luminex Alleles	All Alleles
151AHA	150A151H152A		Confirmed		A*11:01, A*11:02 More	A*01:26, A*02:27, A*11:01, A*11:02, A... More
151HA	151H152A				A*01:01, A*11:01, A*11:02, A*36:01 More	A*01:01, A*01:02, A*01:03, A*01:06, A... More
156Q	156Q				A*11:01, A*11:02, A*24:02, A*24:03, C... More	A*01:12, A*01:19, A*01:25, A*02:12, A... More
163R	163R		Provisional		A*01:01, A*11:01, A*11:02, A*25:01, A... More	A*01:01, A*01:02, A*01:03, A*01:06, A... More
163RE	162G163R166E167W		Confirmed	View	A*11:01, A*11:02, A*25:01, A*26:01, A... More	A*02:101, A*02:135, A*02:154, A*02:38... More

We found 5 epitopes in the database.

The HLA Epitope Registry is a Work-In-Progress

- As of July 2013, 85 HLA-ABC, 54 DRB1/3/4/5, 21 DQ (18 DQB1, 3 DQA1), 7 DPB1 and 24 MICA antibody-verified epitopes are listed
- Each database will be updated on a quarterly basis
- Currently working on a tool to allow users to print or create and download pdf files of results

New Antibody-Verified Epitopes

- An important aspect is the participation by HLA professionals who have identified antibodies specific for new and not-so-well described epitopes
- From the website one can download instructions for how to submit information about epitope-specific antibody reactivity

HLAMatchmaker Analysis of Sera from Sensitized Patients

Use of antibody-verified class I epitopes recorded in the International HLA Epitope Registry

Analysis of Epitope Specificity of HLA Antibody

HLA information about the sensitizing event

- High-resolution HLA typing of patient and the immunizing HLA antigen
- Provides information about patient's exposure to mismatched eplets
- Identification of immunizer eplets on alleles in the Luminex panel

Everybody Likes the Luminex Assay with Single Alleles

but

It’s Not a Perfect Test!

Potential Limitations of Single Allele Luminex Assays

- Some Luminex kits have certain alleles with significant MFI values unrelated to allosensitization events (technical problems or do they reflect “natural” antibodies?)

Unexplained high MFI values with 129 first pregnancy sera with no antibodies against paternal HLA class I alleles (Study with Stefan Schaub, Basel, Switzerland)

	Nr of cases	Mean MFI + SD (Range)
Self alleles	129	6 ± 7 (1-55)
Paternal alleles	299	9 ± 17 (1-108)

Luminex allele	Nr of cases	Adjusted MFI*	Luminex allele	Nr of cases	Adjusted MFI*
A*11:02	6	221,254,428,1033,1122,1493	B*14:02	1	835
B*15:12	5	201,209,314,391,709	B*40:02	1	365
A*29:01	4	237,251,519,3874	B*42:01	1	333
A*29:02	4	204,236,1864,3819	B*15:16	1	317
B*45:01	3	305,526,1718	B*67:01	1	310
A*68:02	3	304,889,1141	B*13:02	1	300
B*73:01	3	177,320,629	B*44:03	1	290
B*58:01	2	222,342	B*54:01	1	258
A*80:01	2	880,1726	B*18:01	1	225
B*57:01	2	203,229	B*44:02	1	225
B*82:01	2	272,578	A*26:01	1	224
C*17:01	1	2113	C*07:02	1	211
C*03:02	1	922	A*25:01	1	203

*Adjusted MFI = observed MFI - (Mean MFI with self + 3 StDev) Only >200 values are shown

Antibody Verification of HLA Class I Epitopes

- Duquesnoy RJ, Marrari M, Mulder A, da M. Sousa LCD, Da Silva AS, do Monte SJH. 'First Report on the Antibody Verification of HLA-ABC Epitopes Recorded in the HLA Epitope Registry'. *Tissue Antigens* 83:391-400, 2014
  - 97 HLA-ABC antibody-verified epitopes have been recorded, 62 correspond to eplets and 35 are defined by eplets paired with other residue configurations

Serum Analysis with HLAMatchmaker

Three types of epitopes in the International Registry:

1. Antibody-verified epitopes defined by eplets*
2. Antibody-verified epitopes defined by eplet pairs*
3. Theoretical epitopes with distinct residue structures (might become antibody-verified?)

*Duquesnoy RJ, Marrari M, Mulder A, da M. Sousa LCD, Da Silva AS, do Monte SJH. 'First Report on the Antibody Verification of HLA-ABC Epitopes Recorded in the HLA Epitope Registry'. *Tissue Antigens* 83:391-400, 2014

97 HLA-ABC antibody-verified epitopes have been recorded, 62 correspond to eplets and 35 are defined by eplets paired with other residue configurations

HLAMatchmaker Analysis for Antibody-Verified Epitopes

- Class I SAB data on 28 first pregnancy sera provided by Stefan Schaub (Basel, Switzerland)
- High-resolution types of mother and paternal haplotype of child
- Analysis only done for sera whereby at least one immunizing allele had MFI>2500

Case	HLA Type of Antibody Producer			MFI self	MFI Cut-off	MFI Pos Cont	Mismatched Immunizer Alleles MFI Values			Number of Reactive Alleles HLA-A MFI Range	HLA-B MFI Range	HLA-C MFI Range	cPRA
#186	A*11:01	A*24:02	B*07:02				A*03:02	B*27:05	C*01:02	1	7	2	32%
#186	B*39:06	C*07:02	C*07:02	7±5	100	7002	nd (01=286)	8953	7245	286	104-8953	691-7245	
#254	A*01:01	A*03:01	B*40:02				A*31:01	B*27:05	C*01:02	0	2	8	56%
#254	B*57:01	C*02:02	C*06:02	4±3	100	8855	2	11	3025		2182-3061	210-3202	
#247	A*03:01	A*03:01	B*15:01				A*02:01	Match	C*03:03	13	3	0	71%
#247	B*35:01	C*04:01	C*07:02	15±29	200	9736	4347		1	314-5951	724-1042		
#233	A*02:01	A*23:01	B*15:18				Match	B*40:02	C*02:02	2	31	1	71%
#233	B*51:01	C*07:04	C*15:02	86±39	200	8285		9647	430	7976	226-11608	430	
#373	A*02:01	A*11:01	B*35:03				A*29:01	B*08:01	C*07:02	25	32	3	73%
#373	B*35:01	C*04:01	C*04:01	1±0	100	8695	8649	10629	5	559-10276	117-10629	112-144	
#129	A*01:01	A*23:01	B*15:17				A*30:02	B*13:02	C*06:02	28	27	7	75%
#129	B*49:01	C*07:01	C*07:01	3±3	100	9719	10888	8982	5712	118-11108	103-8982	256-7653	
#355	A*32:01	A*32:01	B*15:01				A*11:01	B*51:01	C*15:02	1	13	9	76%
#355	B*38:01	C*03:03	C*12:03	46±45	200	7861	47	8272	11977	583	243-8272	1075-11977	
#354	A*01:01	A*02:01	B*35:03				A*30:01	B*51:01	C*14:03	8	20	0	77%
#354	B*40:01	C*01:02	C*07:01	8±6	100	8353	5769	5500	nd (02=3)	2609-7792	102-7297		
#100	A*11:01	A*11:01	B*35:01				A*24:02	B*55:01	C*03:03	2	27	2	78%
#100	B*40:02	C*02:02	C*04:01	5±4	100	8341	8	11270	14	132-169	102-11270	186-471	
#387	A*03:01	A*03:01	B*07:02				A*02:05	B*51:01	C*16:02	15	33	3	87%
#387	B*57:02	C*07:02	C*18:01	3±3	100	8180	nd (01=8498)	10242	nd (01=1)	155-8988	132-10242	218-568	
#334	A*01:01	A*03:01	B*08:01				A*23:01	B*27:02	C*02:02	7	28	0	88%
#334	B*35:03	C*07:01	C*07:02	4±7	100	9685	8407	nd (05=5821)	9	105-8978	121-8693		
#188	A*01:01	A*03:01	B*08:01				A*02:01	B*51:01	C*15:02	12	31	1	90%
#188	B*37:01	C*06:02	C*07:01	8±9	100	11215	9506	10487	47	165-11355	282-10994	135	
#369	A*26:01	A*30:01	B*18:01				A*02:01	B*15:01	C*03:04	9	32	3	90%
#369	B*38:01	C*07:01	C*12:03	4±2	100	9197	2879	12245	2848	172-3194	107-12245	2848-4268	
#85	A*02:01	A*30:01	B*13:02				A*68:01	B*35:01	C*07:02	0	11	11	91%
#85	B*14:02	C*06:02	C*08:02	8±3	100	9641	18	270	3383		134-293	163-12308	
#177	A*01:01	A*02:01	B*40:02				A*03:01	B*07:02	C*07:02	0	11	10	92%
#177	B*57:01	C*02:02	C*06:02	30±41	200	9001	8	3379	3045		469-13249	606-11695	
#87	A*01:01	A*68:01	B*40:01				A*02:01	B*35:01	C*04:01	3	28	7	93%
#87	B*44:02	C*03:04	C*07:04	2±2	100	11120	358	6814	183	216-358	130-6951	162-2563	
#376	A*31:01	A*32:01	B*27:05				A*02:01	B*51:01	C*15:02	13	25	0	94%
#376	B*40:01	C*01:02	C*03:04	7±5	100	8580	2947	2034	42	159-3949	218-2441		
#189	A*24:02	A*32:01	B*35:03				A*01:01	B*08:01	C*07:01	27	3	7	95%
#189	B*44:29	C*05:01	C*12:03	1±1	100	8614	11998	1800	ıd (02=7107	241-13734	486-14956	256-10851	
#316	A*02:01	A*03:01	B*07:02				A*24:02	B*40:01	C*03:04	7	39	11	95%
#316	B*18:01	C*07:01	C*07:02	67±91	400	8538	8916	11888	9587	3434-11850	441-12522	539-10443	
#228	A*01:01	A*24:02	B*08:01				A*02:01	B*15:01	C*03:04	28	42	7	96%
#228	B*40:02	C*02:02	C*07:01	117±131	500	7311	10395	9061	6174	814-12334	629-13490	1120-8945	
#159	A*01:01	A*68:01	B*44:02				A*02:01	6421	C*06:02	4	33	4	96%
#159	B*51:01	C*14:02	C*14:03	3±3	100	8709	5136	6421	489	165-6456	108-6421	264-497	
#353	A*30:02	A*30:02	B*18:01				A*02:01	B*49:01	C*07:01	12	43	1	97%
#353	B*18:01	C*05:01	C*05:01	80±133	500	7792	3720	9367	nd (02=1)	542-9855	553-13957	824	
#70	A*11:01	A*68:01	B*07:02				A*02:05	B*50:01	C*06:02	0	46	4	98%
#70	B*07:02	C*07:02	C*07:02	15±24	100	9299	nd (01=1)	7891	124		135-7891	124-288	
#49	A*02:01	A*02:01	B*15:01				A*01:01	B*58:01	C*07:01	25	15	4	98%
#49	B*51:01	C*01:02	C*03:03	33±78	300	10440	11818	6305	ıd (02=1418	5698-13117	455-13108	857-6987	
#24	A*24:02	A*32:01	B*07:02				A*02:01	B*44:02	C*05:01	26	22	1	99%
#24	B*41:02	C*07:02	C*17:03	6±9	100	8627	7609	6742	376	529-7928	108-12019	376	
#275	A*02:01	A*02:01	B*44:05				A*03:02	B*18:01	C*07:01	27	41	8	100%
#275	B*57:01	C*02:02	C*06:02	2±2	100	8573	nd (01=8487)	7916	nd (02=39)	157-6798	130-7916	489-2139	
#384	A*03:01	A*33:01	B*14:02				A*24:02	B*40:06	C*15:02	15	38	12	100%
#384	B*15:18	C*07:04	C*08:02	11±7	100	8644	6122	8106	8227	206-8802	107-11983	114-8426	
#392	A*11:01	A*32:01	B*35:03				A*02:01	B*49:01	C*07:01	10	46	15	100%
#392	B*35:01	C*04:01	C*04:01	270±96	600	9291	11153	7810	d (02=12577	5963-12732	772-12611	2295-12908	

Epitope Analysis Approach

1. Determine antibody specificities to antibody-verified epitopes recorded in the Registry

   What proportion reactive alleles carry such epitopes?

2. Analyze the remaining antibody reactivity and look for informative alleles expressing eplet-defined epitopes
3. Such epitopes might be classified as being newly antibody-verified

Case	Immunizing Allele Antibody-Verified Epitopes on Reactive Alleles	Numbers of Antibody-Verified Epitope-Carrying Reactive Alleles				
		HLA-A	HLA-B	HLA-C	Total	
#186	71ATD,161D	1/1	1/7	0/2	2/10	20%
#254	73TVS,76VS+152RE	0/0	1/2	8/8	9/10	90%
#247	127K, 62GE	9/13	3/3	0/0	12/16	75%
#233	41T, 163EW	1/2	17/31	0/1	18/34	53%
#373	62LQ,66IF+163TEW,76ANT,180E	7/25	8/32	0/3	15/60	25%
#129	56R,80K,144QL	3/28	2/27	7/7	12/62	19%
#355	44RT+69TNT,80K	0/1	8/13	7/9	15/23	65%
#354	56R, 80I,82LR	8/8	19/20	0/0	27/28	96%
#100	65QIA,69AA,163TEW+65QI	0/2	24/27	0/2	24/31	77%
#387	44RT+69TNT,127K,144TKH,163LW+65QI	9/15	18/33	0/3	27/51	53%
#334	65GK,80I,82LR,163EW+73TE	5/7	26/28	0/0	31/35	89%
#188	80I,131S+163LW,144TKH	11/12	22/31	0/1	33/44	75%
#369	44RMA,127K,144TKH,163LW,173K	9/9	23/32	3/3	35/44	80%
#85	44RT,62GE,80K,163TW+65QI,267QE	0/0	1/11	2/11	3/22	14%
#177	65QIA+76ESN,76VRN,267QE	0/0	11/11	10/10	21/21	100%
#87	62GE,44RT,80K,131S+163LW	3/3	22/28	7/7	32/38	84%
#376	131S+163LW,150AAH	11/13	20/25	0/0	31/38	82%
#189	90D,76ANT,138MI+79GT,267QE	21/27	1/3	5/7	27/37	73%
#316	41T,73TVS,82LR,166DG,219W	7/7	27/39	9/11	43/57	75%
#228	44RMA,76VS+152RE,144TKH,144K+76VDT,163LW,173K	9/28	24/42	6/7	39/77	51%
#159	62GE,76ESN,107W	4/4	31/33	0/4	35/41	85%
#353	82LR, 41T,131S+163LW,144TKH	11/12	34/43	0/1	45/56	80%
#70	41T, 69TNT, 131S+163LW	0/0	40/46	0/4	40/50	80%
#49	65RNA+80I,138MI+79GT,144KR,166DG check c ep	25/25	5/15	4/4	34/44	77%
#24	62GE,142TKH,80T,166ES	26/26	12/22	1/1	39/49	80%
#275	73TS, 76ESN, 44RT+69TNT,138MI,144KR,163TEW+65QI	22/27	37/41	7/8	66/76	87%
#384	21H,41T,80K, 82LR,127K,163EW,167DG	14/15	31/38	10/12	55/65	85%
#392	1C,41T,62GE,76VRN,82LR+138T,127K,267QE	10/10	27/46	15/15	52/71	73%

Which additional epitopes might be recognized by antibodies in these first pregnancy sera?

For what proportions of reactive alleles can we identify specific epitopes?

Case	Additional Epitopes on Reactive Alleles	Total Number of Epitope-Carrying Reactive Alleles		Numbers of Remaining Reactive Alleles (MFI)*
#186	6K,71KA, 76ED	7/10	70%	3 (104-108; C*14:02=691)
#254	None	9/10	90%	1 (B*73:01=3061)
#247	None	12/16	75%	4(314-384; A*29:01=650; A*29:02=620)
#233	80N+66I	33/34	97%	1 (430)
#373	62RN+163TW,71STN,245AS	42/60	70%	18 (117-365;A*03:01;B*37:01=690;B*40:02=831;A*30:01=4947)
#129	66NV, 80N+45E (TP?)163TEW+182T	61/62	98%	1 (297)
#355	21H94I+91G	18/23	78%	5(243-467;A*33:01=583;B*14:02=1062;B*15:02=1859)
#354	None	27/28	96%	1 (101)
#100	None	24/31	77%	7 (102-186;C*03:02=471)
#387	69TNT+41A,131S+163LW+62R	41/51	80%	10(132-400; A*33:01=1658;B*54:01=2440)
#334	71KA	32/35	91%	3(105-121)
#188	170RH	38/44	70%	6 (186-345; B*39:01=447,B*67:01=493; B*15:12=510)
#369	None	35/44	80%	9 (107-353; B*40:06=602)
#85	80N+138T,163LW+65QI+70N74Y171Y	22/22	100%	0
#177	None	21/21	100%	0
#87	131S	34/38	89%	4(130-420; B*54:01=750; B*15:12=1732)
#376	144AH+161E	33/38	87%	5 (121-343)
#189	142TKH (TP?)	33/37	86%	4(156-256; B*15:12=486;C*15:02=606)
#316	63EI	49/57	86%	8(441-646;B*15:11=916;B*82:01=1111;B*15:02=1368;B*15:10=1708)
#228	131S, 166EW+182T	69/77	89%	8 (297-758;A*80:01=4483)
#159	80K+14R+16G+138T?	39/41	95%	2(333; B49:01=5167)
#353	131S+170RY	51/56	91%	5(542-718;B*07:02=1310;B*42:01=1639)
#70	131S	46/50	92%	4 (124-288)
#49	138MI,69AA+180Q,166ES (TP)	43/44	98%	1 (B*15:13=867)
#24	None	55/65	85%	10 (108-263; B*15:12=8306)
#275	66NV,69TNT+41A	76/76	100%	0
#384	None	55/65	85%	10 (106-308;B*42:01=404;B*08:01=458;B*82:01=512)
#392	14R,193PI+183D	70/71	99%	1 (772)

*List only alleles with MFI values more than 300 above cut-off values

Conclusion

The HLAMatchmaker version with antibody-verified class I epitopes works quite well

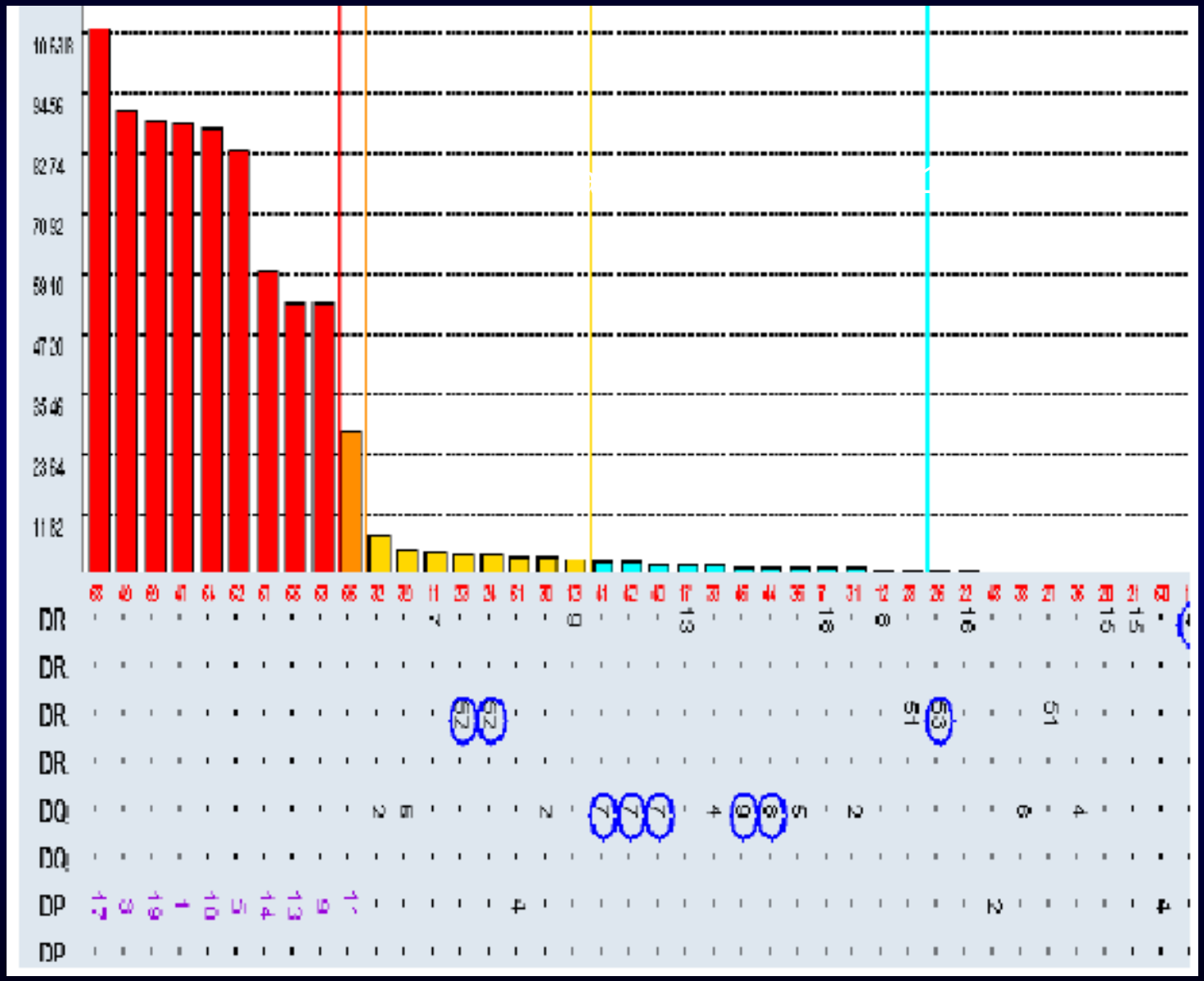
but

more antibody-verified epitopes need to be added

Principles of HLAMatchmaker-based antibody analysis programs

Two examples:
HLA-DP
HLA-C

Luminex Example of Anti-DP Reactivity (Patient Type: DPB1*0201,-



Anti- HLA-DP Reactivity of Serum Group B Pt 43, Patient types as DPB1*0201,-

CON3	x	x	76	
CON1	x	x	107	
CON2	x	x	121	
P 77	x	x	7274	
P 112	DPB1*0101	DPA1*0103	2596	
P 113	DPB1*0101	DPA1*0201	5228	
P 114	DPB1*0101	DPA1*0202	5932	
P 115	DPB1*0101	DPA1*0301	2603	
P 116	DPB1*0201	DPA1*0103	425	SELF
P 117	DPB1*0301	DPA1*0103	2957	
P 118	DPB1*0401	DPA1*0103	894	
P 119	DPB1*0401	DPA1*0201	3807	
P 120	DPB1*0401	DPA1*0202	3650	
P 121	DPB1*0401	DPA1*0301	741	
P 122	DPB1*0402	DPA1*0103	727	
P 123	DPB1*0402	DPA1*0301	832	
P 124	DPB1*0501	DPA1*0103	2958	
P 125	DPB1*0501	DPA1*0201	5863	
P 126	DPB1*0501	DPA1*0202	4790	
P 127	DPB1*0501	DPA1*0301	2714	
P 128	DPB1*1301	DPA1*0201	5080	
P 129	DPB1*1401	DPA1*0201	5812	
P 130	DPB1*1701	DPA1*0201	5104	
P 131	DPB1*1801	DPA1*0103	423	
P 132	DPB1*1901	DPA1*0201	4878	
P 133	DPB1*1901	DPA1*0301	2233	
P 134	DPB1*2801	DPA1*0202	3814	

Anti- HLA-DP Reactivity of Serum Group B Pt 43 Patient types as DPB1*0201,-

CON3	x	x	76	
CON1	x	x	107	
CON2	x	x	121	
P 77	x	x	7274	
P 112	DPB1*0101	DPA1*0103	2596	
P 113	DPB1*0101	DPA1*0201	5228	
P 114	DPB1*0101	DPA1*0202	5932	
P 115	DPB1*0101	DPA1*0301	2603	
P 116	DPB1*0201	DPA1*0103	425	SELF
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P 125	DPB1*0501	DPA1*0201	5863	
P 126	DPB1*0501	DPA1*0202	4790	
P 127	DPB1*0501	DPA1*0301	2714	
P 128	DPB1*1301	DPA1*0201	5080	
P 129	DPB1*1401	DPA1*0201	5812	
P 130	DPB1*1701	DPA1*0201	5104	
P 131	DPB1*1801	DPA1*0103	423	
P 132	DPB1*1901	DPA1*0201	4878	
P 133	DPB1*1901	DPA1*0301	2233	
P 134	DPB1*2801	DPA1*0202	2814	

Questions:

Do the antibodies react with DPB or DPA or both ?
What epitopes are recognized?
Which DP antigens are acceptable mismatches ?
Which DP antigens are unacceptable?

HLAMatchmaker Analysis of Anti- HLA-DP Reactivity of Serum Group B Pt 43

Step 1: Show all eplets in the panel that are mismatched for this patient
(Note the eplets on DPA1*0103 only the “SELF” DPB1*0201)

CON3	x	x	76				
CON1	x	x	107				
CON2	x	x	121				
P 77	x	x	7274		mmEp	DPB Eplets	DPA Eplets
P 112	DPB1*0101	DPA1*0103	2596		15	8V,,,,35YA,,56AE,,65DIK,69IKR,70GKR,76V,84DE,87AV,,,,,,,	18P,28E,51QA,83T,111K,127L,,
P 113	DPB1*0101	DPA1*0201	5228		15	8V,,,,35YA,,56AE,,65DIK,69IKR,70GKR,76V,84DE,87AV,,,,,,,	18P,28E,51RA,83A,111R,127P,,
P 114	DPB1*0101	DPA1*0202	5932		15	8V,,,,35YA,,56AE,,65DIK,69IKR,70GKR,76V,84DE,87AV,,,,,,,	18P,28E,51RA,83A,111R,127P,,
P 115	DPB1*0101	DPA1*0301	2603		15	8V,,,,35YA,,56AE,,65DIK,69IKR,70GKR,76V,84DE,87AV,,,,,,,	18P,28E,51QA,83T,111K,127L,,
P 116	DPB1*0201	**DPA1*0103**	425	SELF	6	,,,,,,,,,,,,,,,,,,,,,,,,	**18P,28E,51QA,83T,111K,127L,,**
P 117	DPB1*0301	DPA1*0103	2957		15	8V,11L,,,,,56ED,64DL,65DLK,,70GKR,76V,84DE,87AV,,,,,,,,,	18P,28E,51QA,83T,111K,127L,,
P 118	DPB1*0401	DPA1*0103	894		11	,,,,35FA,,56AE,,65DIK,69IKR,70GKR,,,,,,,,,,,,,	18P,28E,51QA,83T,111K,127L,,
P 119	DPB1*0401	DPA1*0201	3807		11	,,,,35FA,,56AE,,65DIK,69IKR,70GKR,,,,,,,,,,,,,	18P,28E,51RA,83A,111R,127P,,
P 120	DPB1*0401	DPA1*0202	3650		11	,,,,35FA,,56AE,,65DIK,69IKR,70GKR,,,,,,,,,,,,,	18P,28E,51RA,83A,111R,127P,,
P 121	DPB1*0401	DPA1*0301	741		11	,,,,35FA,,56AE,,65DIK,69IKR,70GKR,,,,,,,,,,,,,	18P,28E,51QA,83T,111K,127L,,
P 122	DPB1*0402	DPA1*0103	727		9	,,,,,,,,65DIK,69IKR,70GKR,,,,,,,,,,,,,	18P,28E,51QA,83T,111K,127L,,
P 123	DPB1*0402	DPA1*0301	832		9	,,,,,,,,65DIK,69IKR,70GKR,,,,,,,,,,,,,	18P,28E,51QA,83T,111K,127L,,
P 124	DPB1*0501	DPA1*0103	2958		13	,,,,35LV,,56AE,,65DIK,69IKR,70GKR,,84DE,87AV,,,,,,,,,,	18P,28E,51QA,83T,111K,127L,,
P 125	DPB1*0501	DPA1*0201	5863		13	,,,,35LV,,56AE,,65DIK,69IKR,70GKR,,84DE,87AV,,,,,,,,,,	18P,28E,51RA,83A,111R,127P,,
P 126	DPB1*0501	DPA1*0202	4790		13	,,,,35LV,,56AE,,65DIK,69IKR,70GKR,,84DE,87AV,,,,,,,,,,	18P,28E,51RA,83A,111R,127P,,
P 127	DPB1*0501	DPA1*0301	2714		13	,,,,35LV,,56AE,,65DIK,69IKR,70GKR,,84DE,87AV,,,,,,,,,,	18P,28E,51QA,83T,111K,127L,,
P 128	DPB1*1301	DPA1*0201	5080		13	8V,11L,,,35YA,,56AE,,,,,76I,84DE,87AV,,,,,,,,,,	18P,28E,51RA,83A,111R,127P,,
P 129	DPB1*1401	DPA1*0201	5812		15	8V,11L,,,,,56ED,64DL,65DLK,,70GKR,76V,84DE,87AV,,,,,,,,,	18P,28E,51RA,83A,111R,127P,,
P 130	DPB1*1701	DPA1*0201	5104		11	8V,11L,,,,,56ED,,,,,,84DE,87AV,,,,,,,,,,	18P,28E,51RA,83A,111R,127P,,
P 131	DPB1*1801	DPA1*0103	423		11	8V,,,,,,,,65DIK,69IKR,70GKR,,84VG,,,,,,,,,,,	18P,28E,51QA,83T,111K,127L,,
P 132	DPB1*1901	DPA1*0201	4878		10	,,,,,,56AE,,,,,76I,84DE,87AV,,,,,,,,,,	18P,28E,51RA,83A,111R,127P,,
P 133	DPB1*1901	DPA1*0301	2233		10	,,,,,,56AE,,,,,76I,84DE,87AV,,,,,,,,,,	18P,28E,51QA,83T,111K,127L,,
P 134	DPB1*2801	DPA1*0202	3814		11	,,,,35FA,,,64DL,65DLK,,70GKR,,84VG,,,,,,,,,,,	18P,28E,51RA,83A,111R,127P,,

HLAMatchmaker Analysis of Anti- HLA-DP Reactivity of Serum Group B Pt 43

Step 2: After recording DPA1*0103 as negative in the program, identify other negative alleles and enter them in the program

CON3		x	x	76			
CON1		x	x	107			
CON2		x	x	121			
P 77		x	x	7274		mmEp	DPB Eplets	DPA Eplets
P 112	DPB1*0101	DPA1*0103		2596		9	8V,,,,35YA,,56AE,,65DIK,69IKR,70GKR,76V,84DE,87AV,,,,,,,	,,,,,,,
P 113	DPB1*0101	DPA1*0201		5228		13	8V,,,,35YA,,56AE,,65DIK,69IKR,70GKR,76V,84DE,87AV,,,,,,,	,,51RA,83A,111R,127P,,
P 114	DPB1*0101	DPA1*0202		5932		13	8V,,,,35YA,,56AE,,65DIK,69IKR,70GKR,76V,84DE,87AV,,,,,,,	,,51RA,83A,111R,127P,,
P 115	DPB1*0101	DPA1*0301		2603		9	8V,,,,35YA,,56AE,,65DIK,69IKR,70GKR,76V,84DE,87AV,,,,,,,	,,,,,,,
P 116	DPB1*0201	DPA1*0103		425	SELF	0	,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 117	DPB1*0301	DPA1*0103		2957		9	8V,11L,,,,,56ED,64DL,65DLK,,70GKR,76V,84DE,87AV,,,,,,,,,,	,,,,,,,
P 118	DPB1*0401	DPA1*0103		894		5	,,,,35FA,,56AE,,65DIK,69IKR,70GKR,,,,,,,,,,,,,	,,,,,,,
P 119	DPB1*0401	DPA1*0201		3807		9	,,,,35FA,,56AE,,65DIK,69IKR,70GKR,,,,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 120	DPB1*0401	DPA1*0202		3650		9	,,,,35FA,,56AE,,65DIK,69IKR,70GKR,,,,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 121	DPB1*0401	DPA1*0301		741	NEG	5	,,,,35FA,,56AE,,65DIK,69IKR,70GKR,,,,,,,,,,,,,	,,,,,,,
P 122	DPB1*0402	DPA1*0103		727	NEG	3	,,,,,,,,65DIK,69IKR,70GKR,,,,,,,,,,,,,	,,,,,,,
P 123	DPB1*0402	DPA1*0301		832	NEG	3	,,,,,,,,65DIK,69IKR,70GKR,,,,,,,,,,,,,	,,,,,,,
P 124	DPB1*0501	DPA1*0103		2958		7	,,,,35LV,,56AE,,65DIK,69IKR,70GKR,,84DE,87AV,,,,,,,,,,	,,,,,,,
P 125	DPB1*0501	DPA1*0201		5863		11	,,,,35LV,,56AE,,65DIK,69IKR,70GKR,,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 126	DPB1*0501	DPA1*0202		4790		11	,,,,35LV,,56AE,,65DIK,69IKR,70GKR,,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 127	DPB1*0501	DPA1*0301		2714		7	,,,,35LV,,56AE,,65DIK,69IKR,70GKR,,84DE,87AV,,,,,,,,,,	,,,,,,,
P 128	DPB1*1301	DPA1*0201		5080		11	8V,11L,,,35YA,,56AE,,,,,76I,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 129	DPB1*1401	DPA1*0201		5812		13	8V,11L,,,,,56ED,64DL,65DLK,,70GKR,76V,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 130	DPB1*1701	DPA1*0201		5104		9	8V,11L,,,,,56ED,,,,,,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 131	DPB1*1801	DPA1*0103		423	NEG	5	8V,,,,,,,,65DIK,69IKR,70GKR,,84VG,,,,,,,,,,,	,,,,,,,
P 132	DPB1*1901	DPA1*0201		4878		8	,,,,,,56AE,,,,,76I,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 133	DPB1*1901	DPA1*0301		2233		4	,,,,,,56AE,,,,,76I,84DE,87AV,,,,,,,,,,	,,,,,,,
P 134	DPB1*2801	DPA1*0202		3814		9	,,,,35FA,,,64DL,65DLK,,70GKR,,84VG,,,,,,,,,,,	,,51RA,83A,111R,127P,,

HLAMatchmaker Analysis of Anti- HLA-DP Reactivity of Serum Group B Pt 43

Step 3: Remaining eplets after entering the negative DP alleles in the program

CON3	x	x	76				
CON1	x	x	107				
CON2	x	x	121				
P 77	x	x	7274		mmEp	DPB Eplets	DPA Eplets
P 112	DPB1*0101	DPA1*0103	2596		4	,,,,35YA,,,,,,,76V,84DE,87AV,,,,,,,,,,	,,,,,,,
P 113	DPB1*0101	DPA1*0201	5228		8	,,,,35YA,,,,,,,76V,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 114	DPB1*0101	DPA1*0202	5932		8	,,,,35YA,,,,,,,76V,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 115	DPB1*0101	DPA1*0301	2603		4	,,,,35YA,,,,,,,76V,84DE,87AV,,,,,,,,,,	,,,,,,,
P 116	DPB1*0201	DPA1*0103	425	SELF	0	,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 117	DPB1*0301	DPA1*0103	2957		7	,11L,,,,,56ED,64DL,65DLK,,,76V,84DE,87AV,,,,,,,,,,	,,,,,,,
P 118	DPB1*0401	DPA1*0103	894		0	,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 119	DPB1*0401	DPA1*0201	3807		4	,,,,,,,,,,,,,,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 120	DPB1*0401	DPA1*0202	3650		4	,,,,,,,,,,,,,,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 121	DPB1*0401	DPA1*0301	741	NEG	0	,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 122	DPB1*0402	DPA1*0103	727	NEG	0	,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 123	DPB1*0402	DPA1*0301	832	NEG	0	,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 124	DPB1*0501	DPA1*0103	2958		3	,,,,35LV,,,,,,,84DE,87AV,,,,,,,,,,	,,,,,,,
P 125	DPB1*0501	DPA1*0201	5863		7	,,,,35LV,,,,,,,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 126	DPB1*0501	DPA1*0202	4790		7	,,,,35LV,,,,,,,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 127	DPB1*0501	DPA1*0301	2714		3	,,,,35LV,,,,,,,84DE,87AV,,,,,,,,,,	,,,,,,,
P 128	DPB1*1301	DPA1*0201	5080		9	,11L,,,35YA,,,,,,,76I,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 129	DPB1*1401	DPA1*0201	5812		11	,11L,,,,,56ED,64DL,65DLK,,,76V,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 130	DPB1*1701	DPA1*0201	5104		8	,11L,,,,,56ED,,,,,,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 131	DPB1*1801	DPA1*0103	423	NEG	0	,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 132	DPB1*1901	DPA1*0201	4878		7	,,,,,,,,,,76I,84DE,87AV,,,,,,,,,,	,,51RA,83A,111R,127P,,
P 133	DPB1*1901	DPA1*0301	2233		3	,,,,,,,,,,76I,84DE,87AV,,,,,,,,,,	,,,,,,,
P 134	DPB1*2801	DPA1*0202	3814		6	,,,,,,,64DL,65DLK,,,,,,,,,,,,,,,	,,51RA,83A,111R,127P,,

HLAMatchmaker Analysis of Anti- HLA-DP Reactivity of Serum Group B Pt 43

Step 4: determine what eplets are shared between reactive alleles

CON3	x	x	76				
CON1	x	x	107				
CON2	x	x	121				
P 77	x	x	7274		mmEp	DPB Eplets	DPA Eplets
P 112	DPB1*0101	DPA1*0103	2596		4	,,,,35YA,,,,,,,76V,**84DE,87AV**,,,,,,,,,,,	,,,,,,,
P 113	DPB1*0101	DPA1*0201	5228		8	,,,,35YA,,,,,,,76V,**84DE,87AV**,,,,,,,,,,,	,,**51RA,83A**,111R,127P,,
P 114	DPB1*0101	DPA1*0202	5932		8	,,,,35YA,,,,,,,76V,**84DE,87AV**,,,,,,,,,,,	,,**51RA,83A**,111R,127P,,
P 115	DPB1*0101	DPA1*0301	2603		4	,,,,35YA,,,,,,,76V,**84DE,87AV**,,,,,,,,,,,	,,,,,,,
P 116	DPB1*0201	DPA1*0103	425	SELF	0	,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 117	DPB1*0301	DPA1*0103	2957		7	,11L,,,,,56ED,64DL,65DLK,,,76V,**84DE,87AV**,,,,,,,,,,	,,,,,,,
P 118	DPB1*0401	DPA1*0103	894		0	,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 119	DPB1*0401	DPA1*0201	3807		4	,,,,,,,,,,,,,,,,,,,,,,,,	,,**51RA,83A**,111R,127P,,
P 120	DPB1*0401	DPA1*0202	3650		4	,,,,,,,,,,,,,,,,,,,,,,,,	,,**51RA,83A**,111R,127P,,
P 121	DPB1*0401	DPA1*0301	741	NEG	0	,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 122	DPB1*0402	DPA1*0103	727	NEG	0	,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 123	DPB1*0402	DPA1*0301	832	NEG	0	,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 124	DPB1*0501	DPA1*0103	2958		3	,,,,35LV,,,,,,,,**84DE,87AV**,,,,,,,,,,	,,,,,,,
P 125	DPB1*0501	DPA1*0201	5863		7	,,,,35LV,,,,,,,,**84DE,87AV**,,,,,,,,,,	,,**51RA,83A**,111R,127P,,
P 126	DPB1*0501	DPA1*0202	4790		7	,,,,35LV,,,,,,,,**84DE,87AV**,,,,,,,,,,	,,**51RA,83A**,111R,127P,,
P 127	DPB1*0501	DPA1*0301	2714		3	,,,,35LV,,,,,,,,**84DE,87AV**,,,,,,,,,,	,,,,,,,
P 128	DPB1*1301	DPA1*0201	5080		9	,11L,,,35YA,,,,,,,76I,**84DE,87AV**,,,,,,,,,,	,,**51RA,83A**,111R,127P,,
P 129	DPB1*1401	DPA1*0201	5812		11	,11L,,,,,56ED,64DL,65DLK,,,76V,**84DE,87AV**,,,,,,,,,,	,,**51RA,83A**,111R,127P,,
P 130	DPB1*1701	DPA1*0201	5104		8	,11L,,,,,56ED,,,,,,**84DE,87AV**,,,,,,,,,,	,,**51RA,83A**,111R,127P,,
P 131	DPB1*1801	DPA1*0103	423	NEG	0	,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,
P 132	DPB1*1901	DPA1*0201	4878		7	,,,,,,,,,,76I,**84DE,87AV**,,,,,,,,,,	,,**51RA,83A**,111R,127P,,
P 133	DPB1*1901	DPA1*0301	2233		3	,,,,,,,,,,76I,**84DE,87AV**,,,,,,,,,,	,,,,,,,
P 134	DPB1*2801	DPA1*0202	3814		6	,,,,,,,64DL,65DLK,,,,,,,,,,,,,,,	,,**51RA,83A**,111R,127P,,

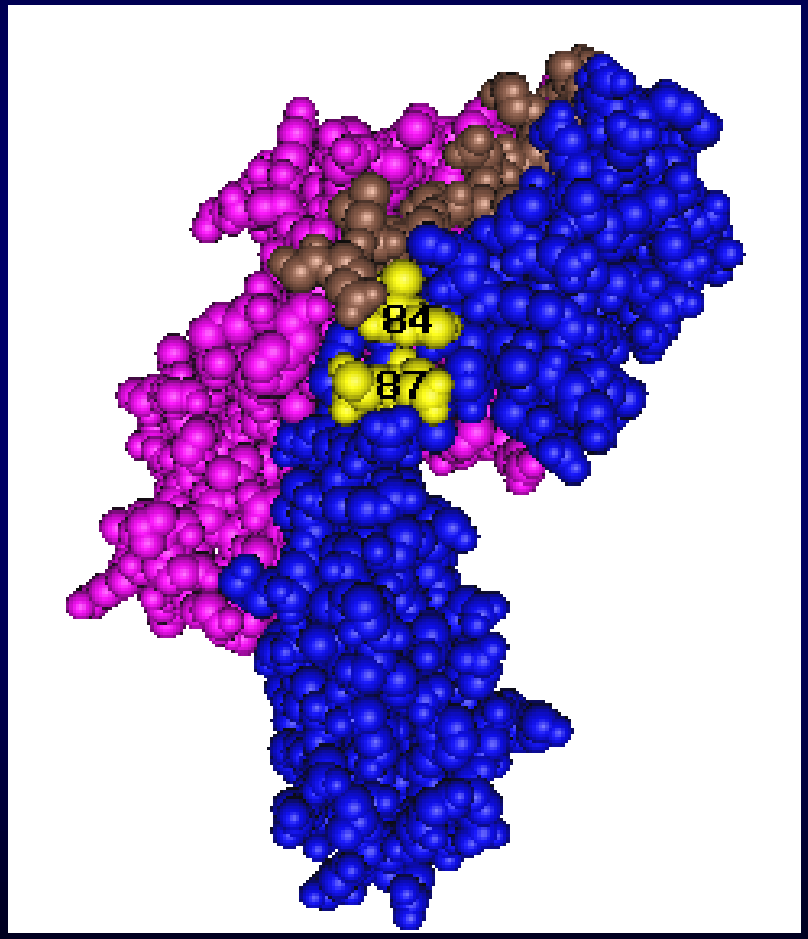
HLAMatchmaker-Predicted Unacceptable DPB Alleles not Present in the Luminex Panel

DPB1*0601	8V,11L,,,,,56ED,64DL,65DLE,,,,**84DE,87AV**,,
DPB1*0801	,,,,,,,,,,76V,**84DE,87AV**,,
DPB1*1601	,,,,,,,,,,,**84DE,87AV**,,
DPB1*2001	8V,11L,,,,,56ED,64DL,65DLK,,,,**84DE,87AV**,,
DPB1*2101	8V,11L,,,35LV,,,,,,,,**84DE,87AV**,,
DPB1*2201	,,,,35LV,,,,,,,,**84DE,87AV**,,
DPB1*2501	8V,11L,,,,,,64DL,65DLK,,,76V,**84DE,87A**V,,
DPB1*2601	8V,11L,,,35YA,,,,,,,76V,**84DE,87AV**,,
DPB1*2701	8V,11L,,,35YA,,,,,,,,**84DE,87AV**,,
DPB1*2901	8V,11L,,,,,56ED,64DL,65DLE,,,76V**,84DE,87AV**,,
DPB1*3001	8V,11L,,,,,,,,,,**84DE,87AV**,,
DPB1*3101	,,,,,,,64DL,65DLK,,,,**84DE,87AV**,,
DPB1*3501	8V,11L,,,,,56ED,,,,,76V,**84DE,87AV**,,
DPB1*3601	8V,11L,,,35LV,,,,,,,,**84DE,87AV**,,
DPB1*3701	8V,11L,,,,,,,,,,76V**,84DE,87AV**,,
DPB1*3801	,,,,35LV,,,,,,,,**84DE,87AV**,,
DPB1*4401	8V,11L,,,35LV,,56ED,64DL,65DLE,,,76V,**84DE,87AV**,,
DPB1*4501	8V,11L,,,,,,64DL,65DLK,,,76V,**84DE,87AV**,,
DPB1*5001	8V,,,,,,56ED,64DL,65DLK,,,76V,**84DE,87AV**,,
DPB1*5201	8V,11L,,,,,,64DL,65DLK,,,76V,**84DE,87AV**,,
DPB1*5401	8V,11L,,,,,,,,,,76V**,84DE,87AV,**,,
DPB1*5501	8V,11L,,,,,,,,,,,**84DE,87AV**,,
DPB1*5601	8V,11L,,,,,,64DL,65DLK,,,76V,**84DE,87AV**,,
DPB1*5701	,,,,,,56ED,64DL,65DLK,,,76V,**84DE**,,,
DPB1*5801	8V,11L,,,35LV,,,,,,,,**84DE,87AV**,,
DPB1*6301	,,,,35LV,,,,,,,,**84DE,87AV**,,
DPB1*6501	,,,,35YA,,,,,,,76V,**84DE,87AV**,,
DPB1*6701	8V,11L,,,,,,64DL,65DLK,,,76V,**84DE,87AV**,,
DPB1*6801	,,,,,,,,,,,76V,**84DE,87AV**,,
DPB1*6901	8V,11L,,,,,56ED,64DL,65DLR,,,,**84DE,87AV**,,
DPB1*7001	8V,11L,,,,,56ED,64DL,65DLK,,,76V,**84DE,87AV**,,
DPB1*7601	8V,11L,,,,,56ED,64DL,65DLK,,,76V,**84DE,87AV**,,
DPB1*7801	8V,11L,,,,,56ED,64DL,65DLK,,,76V,**84DE,87AV,**,,
DPB1*7901	8V,11L,,,,,,,,,,76V,**84DE,87AV**,,
DPB1*8401	,,,,,,,,,,,76V,**84DE,87AV**,,
DPB1*8501	8V,11L,,,35YA,,,,,,,,**84DE,87AV**,91H,
DPB1*8701	8V,11L,,,,,,64DL,65DLK,,,,**84DE,87AV**,,
DPB1*8801	8V,11L,,,,,56ED,,,,,76V,**84DE,87AV**,,
DPB1*8901	8V,,,,35YA,,,,,,,,**84DE,87AV**,,
DPB1*9001	8V,,,,,,,,,,,76V,**84DE,87AV**,,
DPB1*9101	8V,11L,,,,,56ED,64DL,65DLK,,,,**84DE,87AV**,,
DPB1*9201	8V,11L,,,,,56ED,64DL,65DLK,,,76V,**84DE,87AV**,,
DPB1*9301	8V,11L,,,,,,,,,,,**84DE,87AV**,,
DPB1*9701	,,,,35LV,43WR,,,,,,,**84DE,87AV**,,
DPB1*9801	8V,11L,,,,,56ED,,,,,,**84DE,87AV**,,

HLAMatchmaker-Predicted Acceptable DPB Alleles not Present in the Luminex Panel

DPB1*0202	1	,,,,35LV,,,,,,,,,,
DPB1*1501	6	8V,,,33Q,35YA,,,64DL,65DLR,,,,84VG,,,
DPB1*1801	2	8V,,,,,,,,,,,84VG,,,
DPB1*2301	0	,,,,,,,,,,,,,,
DPB1*2401	0	,,,,,,,,,,,,,,
DPB1*2801	3	,,,,,,,64DL,65DLK,,,,84VG,,,
DPB1*3201	1	,,,,,,56EV,,,,,,,,
DPB1*3301	0	,,,,,,,,,,,,,,
DPB1*3401	4	,,,,35LV,,,64DL,65DLK,,,,84VG,,,
DPB1*3901	1	,,,,35YA,,,,,,,,,,
DPB1*4001	2	,,,,35YA,,,,,,,,84VG,,,
DPB1*4101	3	,,,,,,,64DF,65DFE,69FER,,,,,,
DPB1*4601	1	,,,,,,56ED,,,,,,,,
DPB1*4701	0	,,,,,,,,,,,,,,
DPB1*4801	1	,,,,35LV,,,,,,,,,,
DPB1*4901	1	,,,,35YA,,,,,,,,,,
DPB1*5101	0	,,,,,,,,,,,,,,
DPB1*5301	2	,,,,35YA,,,,,,,,84VG,,,
DPB1*6001	1	,,,,,,,64DN,,,,,,,,
DPB1*6201	2	,,,,35LV,,,,,,,,84VG,,,
DPB1*6601	2	8V,11L,,,,,,,,,,,,,
DPB1*7101	0	,,,,,,,,,,,,,,
DPB1*7201	2	,,,,,,,64DL,65DLK,,,,,,,
DPB1*7301	3	,,,,,,,64DL,65DLK,,,76V,,,,
DPB1*7401	7	8V,11L,,33Q,35YA,,,64DL,65DLR,,,,84VG,,,
DPB1*7501	1	,,,,,,,,,,,76V,,,,
DPB1*7701	0	,,,,,,,,,,,,,,
DPB1*8001	1	,,,,,,56ED,,,,,,,,
DPB1*8101	0	,,,,,,,,,,,,,,
DPB1*8201	1	,,,,,,,,,,,,,,91H,
DPB1*8301	2	,,,,,,,64DF,65DFK,,,,,,,
DPB1*8601	3	8V,11L,,,,,56ED,,,,,,,,
DPB1*9401	1	,,,,,43GW,,,,,,,,,,
DPB1*9501	3	,,,,35LV,,,64DL,65DLE,,,,,,,
DPB1*9601	2	,,,,35YA,,,64HI,,,,,,,,
DPB1*9901	1	,,28D,,,,,,,,,,,,

Molecular Locations of Positions 84 and 87



84DE+87AV Corresponds to an Antibody-Defined Epitope

HLA-DP Epitope Typing Using Monoclonal Antibodies

William H. Marshall, Sheila Drover, Dianne Codner, Jane Gamberg, M. Douglas Copp, Hong-Wei Liu, Lang-Tuo Deng, and H. Banfield Younghusband

Human Immunology 59, 189–197 (1998)

TABLE 4 Proposed specificities of the monoclonal antibodies

NFLD.M66	IgG1	DPB1:84–87:DEAV
NFLD.M126	IgG1	DPB1:84–87:DEAV
NFLD.M120	IgG2a	DPB1:85–87:GPM
NFLD.M123	IgM	DPB1:84–87:GGPM

Frequency of Anti-84DE,87AV Reactive Antibodies

Our experience:

- 78/355 (22%) HLA class II-reactive patient sera have anti-HLA-DP antibodies
- 33/78 (42%) react with 84DE,87AV

Frequency of Anti-84DE,87AV Reactive Antibodies

Our experience:

- 78/355 (22%) HLA-reactive patient sera have anti-HLA-DP antibodies
- 33/78 (42%) react with 84DE,87AV

Danny Youngs (Seattle) ASHI Quarterly 2004

29/67 (43%) of patients with anti-DP antibodies react with DEAV at position 84-87

HLAMatchmaker Determination of Unacceptable DPA Alleles

	Eplet	Unacceptable Alleles
DPA	51RA	*0108*0201*0202*0203*0401
DPA	83A	*0105*0201*0202*0203*0401
DPA	111R	*0201*0202
DPA	127P	*0201*0202*0401

HLA-C Antibodies

Example of Luminex Screen for Antibodies Specific for HLA-C (1)

	HLA-C Type
Patient	Cw*0602
Patient	Cw*0702
Imm Donor	Cw*0102
Imm Donor	Cw*0701

Bead	Allele	MFI	RX
1	NegCont	880	
2	PosCont	12591	
83	Cw*0102	9850	**IMM**
84	Cw*0202	1054	
98	Cw*0302	2118	
97	Cw*0303	5498	
99	Cw*0304	4991	
85	Cw*0401	1393	
86	Cw*0501	1330	
87	Cw*0602	813	SELF
88	Cw*0702	624	SELF
89	Cw*0801	2465	
90	Cw*1203	767	
91	Cw*1402	7769	
92	Cw*1502	738	
93	Cw*1601	4305	
94	Cw*1701	798	
95	Cw*1802	1099	

What is the cut-off point to distinguish between positive and negative reactions?

Example of Luminex Screen for Antibodies Specific for HLA-C (2)

		HLA-C Type
	Patient	Cw*0602
	Patient	Cw*0702
	Imm Donor	Cw*0102
	Imm Donor	Cw*0701
	Cut Off	1400

Bead	Allele	MFI	RX
1	NegCont	880	
2	PosCont	12591	
83	Cw*0102	9850	**IMM**
84	Cw*0202	1054	NEG
98	Cw*0302	2118	**POS**
97	Cw*0303	5498	**POS**
99	Cw*0304	4991	**POS**
85	Cw*0401	1393	NEG
86	Cw*0501	1330	NEG
87	Cw*0602	813	SELF
88	Cw*0702	624	SELF
89	Cw*0801	2465	**POS**
90	Cw*1203	767	NEG
91	Cw*1402	7769	**POS**
92	Cw*1502	738	NEG
93	Cw*1601	4305	**POS**
94	Cw*1701	798	NEG
95	Cw*1802	1099	NEG

Antibody react with Cw*0102 of donor
Reactivity with donor's Cw*0701? No allele in panel
What HLA-C epitopes are recognized?

Do HLAMatchmaker analysis
What are the mismatched eplets?

Example of Luminex Screen for Antibodies Specific for HLA-C (3)

	HLA-C Type	
Patient	Cw*0602	
Patient	Cw*0702	
Imm Donor	Cw*0102	,1C,6K,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,219W,,,,,
Imm Donor	Cw*0701	,,,,,,,65QNR,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,

Bead	Allele	MFI	RX	Mismatched Eplets
	Cut Off	1400		
1	NegCont	880		,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
2	PosCont	12591		,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
83	Cw*0102	9850	**IMM**	,1C,6K,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,219W,,,,,
84	Cw*0202	1054	NEG	,,21H,,,,,,,69QRQT,,,,,77TVN,,,,,,,,,,,,,,,,,211T,,,,,
98	Cw*0302	2118	**POS**	,,21H,,,,,,,69QRQT,,,,,77TVS,,,,,94II,,,,,,,,,,163LW,173K,,,219W,,,,,
97	Cw*0303	5498	**POS**	,,21H,,,,,,,69QRQT,,,,,77TVS,,,,,94II,,,,,,,,,,163LW,173K,,,219W,,,,,
99	Cw*0304	4991	**POS**	,,21H,,,,,,,69QRQT,,,,,77TVS,,,,,94II,,,,,,,,,,163LW,173K,,,219W,,,,,
85	Cw*0401	1393	NEG	,,17WR,,,,,,,,,,,,,,,,,,,113YN,116F,,,,,,,,,,219W,,,275KP,,
86	Cw*0501	1330	NEG	,,35Q,,,46QGE,,,,69QRQT,,,,,77TVN,,,,,,,,113YN,116F,138K,,,,,,177KT,,,,,,275GP,,
87	Cw*0602	813	SELF	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
88	Cw*0702	624	SELF	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
89	Cw*0801	2465	**POS**	,,35Q,,,46QGE,,,,69QRQT,,,,,77TVS,,,,,,,,113YN,116F,,,,,152RT,,177KT,,,,,,275GP,,
90	Cw*1203	767	NEG	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
91	Cw*1402	7769	**POS**	,,,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,219W,,,,,
92	Cw*1502	738	NEG	,,21H,,,,,65QNR,,69QRQT,,,,,77TVN,,,,,94II,,,,,,,,,,,,,,,,,,
93	Cw*1601	4305	**POS**	,,,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,193LV,,,,,,
94	Cw*1701	798	NEG	,,,,,,,,,,,,,,,,,,,,,113YN,116F,143ISQ,,,,,,,184R,,270C,,,275KP,,
95	Cw*1802	1099	NEG	,,,,,,,,,,,,,,,,,,,,,113YN,116F,,,,,,,,,,219W,,,275KP,,

These are the eplets on HLA-C alleles of donor and the panel
Cw*1203 has no mismatched eplets for this patient
One or more eplets on donor's Cw*0102 must react with antibody
Any reactivity with 65QNR of donor's Cw*0701?

Example of Luminex Screen for Antibodies Specific for HLA-C (4)

	HLA-C Type	
Patient	Cw*0602	
Patient	Cw*0702	
Imm Donor	Cw*0102	,1C,6K,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,219W,,,,,
Imm Donor	Cw*0701	,,,,,,,**65QNR**,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,

	Cut Off	1400		
Bead	Allele	MFI	RX	Mismatched Eplets
1	NegCont	880		,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
2	PosCont	12591		,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
83	Cw*0102	9850	**IMM**	,1C,6K,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,219W,,,,,
84	Cw*0202	1054	NEG	,,21H,,,,,,,69QRQT,,,,,77TVN,,,,,,,,,,,,,,,,,211T,,,,,
98	Cw*0302	2118	**POS**	,,21H,,,,,,,69QRQT,,,,,77TVS,,,,,94II,,,,,,,,,,163LW,173K,,,219W,,,,,
97	Cw*0303	5498	**POS**	,,21H,,,,,,,69QRQT,,,,,77TVS,,,,,94II,,,,,,,,,,163LW,173K,,,219W,,,,,
99	Cw*0304	4991	**POS**	,,21H,,,,,,,69QRQT,,,,,77TVS,,,,,94II,,,,,,,,,,163LW,173K,,,219W,,,,,
85	Cw*0401	1393	NEG	,,17WR,,,,,,,,,,,,,,,,,,,113YN,116F,,,,,,,,,,219W,,,275KP,,
86	Cw*0501	1330	NEG	,,35Q,,,46QGE,,,,69QRQT,,,,,77TVN,,,,,,,,113YN,116F,138K,,,,,,177KT,,,,,,275GP,,
87	Cw*0602	813	SELF	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
88	Cw*0702	624	SELF	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
89	Cw*0801	2465	**POS**	,,35Q,,,46QGE,,,,69QRQT,,,,,77TVS,,,,,,,,113YN,116F,,,,,152RT,,177KT,,,,,,275GP,,
90	Cw*1203	767	NEG	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
91	Cw*1402	7769	**POS**	,,,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,219W,,,,,
92	Cw*1502	738	NEG	,,21H,,,,,**65QNR**,,69QRQT,,,,,77TVN,,,,,94II,,,,,,,,,,,,,,,,,,
93	Cw*1601	4305	**POS**	,,,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,193LV,,,,,,
94	Cw*1701	798	NEG	,,,,,,,,,,,,,,,,,,,,,,113YN,116F,143ISQ,,,,,,,184R,,270C,,,275KP,,
95	Cw*1802	1099	NEG	,,,,,,,,,,,,,,,,,,,,,,113YN,116F,,,,,,,,,,219W,,,275KP,,

No reactivity with 65QNR of donor's Cw*0701
No reactivity with the eplets of the negative alleles
Cw*0202, Cw*0401, Cw*0501, Cw*1502, Cw*1701 and Cw*1801
Record these negative alleles in the HLAMatchmaker program

Example of Luminex Screen for Antibodies Specific for HLA-C (5)

	HLA-C Type	
Patient	Cw*0602	
Patient	Cw*0702	
Imm Donor	Cw*0102	,1C,6K,,,,,,,69QRQT,,,,,**77TVS**,,,,,,,,,,,,,,,,,,219W,,,,,
Imm Donor	Cw*0701	,,,,,,,65QNR,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,

Bead	Allele	MFI	RX	Mismatched Eplets	Eplets on Reactive Alleles
	Cut Off	1400			
1	NegCont	880		,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	
2	PosCont	12591		,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	
83	Cw*0102	9850	**IMM**	,1C,6K,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,,219W,,,,,	,1C,6K,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,,,,,,,,,,
84	Cw*0202	1054	NEG	,,21H,,,,,,,69QRQT,,,,,77TVN,,,,,,,,,,,,,,,,,,211T,,,,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
98	Cw*0302	2118	**POS**	,,21H,,,,,,,69QRQT,,,,,77TVS,,,,,94II,,,,,,,,,,163LW,173K,,,219W,,,,,	,,,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,,163LW,173K,,
97	Cw*0303	5498	**POS**	,,21H,,,,,,,69QRQT,,,,,77TVS,,,,,94II,,,,,,,,,,163LW,173K,,,219W,,,,,	,,,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,,163LW,173K,,
99	Cw*0304	4991	**POS**	,,21H,,,,,,,69QRQT,,,,,77TVS,,,,,94II,,,,,,,,,,163LW,173K,,,219W,,,,,	,,,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,,163LW,173K,,
85	Cw*0401	1393	NEG	,,17WR,,,,,,,,,,,,,,,,,,,113YN,116F,,,,,,,,,,219W,,,275KP,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
86	Cw*0501	1330	NEG	,,35Q,,,46QGE,,,,69QRQT,,,,,77TVN,,,,,,,,113YN,116F,138K,,,,,,177KT,,,,,,2	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
87	Cw*0602	813	SELF	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
88	Cw*0702	624	SELF	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
89	Cw*0801	2465	**POS**	,,35Q,,,46QGE,,,,69QRQT,,,,,77TVS,,,,,,,,113YN,116F,,,,,152RT,,177KT,,,,,,	,,,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,152RT,,,,,,,,,,
90	Cw*1203	767	NEG	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
91	Cw*1402	7769	**POS**	,,,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,,219W,,,,,	,,,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,,,,,,,,,,
92	Cw*1502	738	NEG	,,21H,,,,,65QNR,,69QRQT,,,,,77TVN,,,,,94II,,,,,,,,,,,,,,,,,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
93	Cw*1601	4305	**POS**	,,,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,193LV,,,,,,	,,,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,,,,,193LV,,,,,,
94	Cw*1701	798	NEG	,,,,,,,,,,,,,,,,,,,,,113YN,116F,143ISQ,,,,,,,184R,,270C,,,275KP,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
95	Cw*1802	1099	NEG	,,,,,,,,,,,,,,,,,,,,,113YN,116F,,,,,,,,,,219W,,,275KP,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,

The 77TVS eplet of donor's Cw*0102 is present on all reactive alleles

77TVS is an unacceptable mismatch

Example of Luminex Screen for Antibodies Specific for HLA-C (6)

	HLA-C Type
Patient	Cw*0602
Patient	Cw*0702
Imm Donor	Cw*0102 ,1C,6K,,,,,,,69QRQT,,,,,**77TVS**,,,,,,,,,,,,,,,,,,219W,,,,,
Imm Donor	Cw*0701 ,,,,,,,65QNR,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,

Cut Off 1400

Bead	Allele	MFI	RX	Mismatched Eplets	Eplets on Reactive Alleles
1	NegCont	880		,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	
2	PosCont	12591		,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	
83	Cw*0102	9850	IMM	,1C,6K,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,,219W,,,,,	,1C,6K,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,,,,,,,,,,,
84	Cw*0202	1054	NEG	,,21H,,,,,,,69QRQT,,,,,77TVN,,,,,,,,,,,,,,,,,,211T,,,,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
98	Cw*0302	2118	POS	,,21H,,,,,,,69QRQT,,,,,77TVS,,,,,94II,,,,,,,,,,163LW,173K,,,219W,,,,,	,,,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,,163LW,173K,,
97	Cw*0303	5498	POS	,,21H,,,,,,,69QRQT,,,,,77TVS,,,,,94II,,,,,,,,,,163LW,173K,,,219W,,,,,	,,,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,,163LW,173K,,
99	Cw*0304	4991	POS	,,21H,,,,,,,69QRQT,,,,,77TVS,,,,,94II,,,,,,,,,,163LW,173K,,,219W,,,,,	,,,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,,163LW,173K,,
85	Cw*0401	1393	NEG	,,17WR,,,,,,,,,,,,,,,,,,,113YN,116F,,,,,,,,,,219W,,,275KP,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
86	Cw*0501	1330	NEG	,,35Q,,,46QGE,,,,69QRQT,,,,,77TVN,,,,,,,,113YN,116F,138K,,,,,,177KT,,,,,,2	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
87	Cw*0602	813	SELF	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
88	Cw*0702	624	SELF	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
89	Cw*0801	2465	POS	,,35Q,,,46QGE,,,,69QRQT,,,,,77TVS,,,,,,,,113YN,116F,,,,,152RT,,177KT,,,,,,	,,,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,152RT,,,,,,,,,,
90	Cw*1203	767	NEG	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
91	Cw*1402	7769	POS	,,,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,,219W,,,,,	,,,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,,,,,,,,,,,
92	Cw*1502	738	NEG	,,21H,,,,,65QNR,,69QRQT,,,,,77TVN,,,,,94II,,,,,,,,,,,,,,,,,,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
93	Cw*1601	4305	POS	,,,,,,,,,69QRQT,,,,,77TVS,,,,,,,,,,,,,,,,,193LV,,,,,,	,,,,,,,,,,,,,,**77TVS**,,,,,,,,,,,,,,,,,,193LV,,,,,,
94	Cw*1701	798	NEG	,,,,,,,,,,,,,,,,,,,,,113YN,116F,143ISQ,,,,,,,184R,,270C,,,275KP,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,
95	Cw*1802	1099	NEG	,,,,,,,,,,,,,,,,,,,,,113YN,116F,,,,,,,,,,219W,,,275KP,,	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,

Acceptable Alleles

Cw*0202 *0401*0403*0404*0405*0501*0602*0603*0604*0701*0702*0704*0706*0707*0710*1202*1203*1204*1502*1503*1505*1506*1509*1701*1801

Eplet	**Unacceptable Alleles**
77TVS	**B*4601**Cw*0102*0103*0302*0303*0304*0305*0306*0310*0801*0802*0803*0804*1402*1403*1601*1604