

Comparative profiling of HLA-DR and HLA-DQ associated factor VIII peptides presented by monocyte-derived dendritic cells

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Supplemental informations

Reagents

The hybridoma producing the anti-HLA-DQ monoclonal antibody (SPV-L3, IgG2a)²¹ was a kind gift from Prof. Dr. H. Spits (Academic Medical Center, Amsterdam). The hybridoma producing the HLA-DR-specific monoclonal antibody (L243, IgG2a) was purchased from ATCC (Wesel, Germany). Full-length recombinant FVIII was a kind gift from Dr. Birgit Reipert (Baxalta Innovations, Vienna, Austria). Antibodies were purified from hybridoma supernatant using protein A Sepharose (GE Healthcare) and coupled to CNBr-activated Sepharose 4B at a final concentration of 2 mg/ml according to manufacturer's recommendation (Amersham Biosciences, Buckinghamshire, UK).

Generation of monocyte-derived dendritic cells

Buffy coat were obtained from healthy volunteers in accordance with Dutch regulations after approval from the Sanquin Ethical Advisory Board, in accordance with the Declaration of Helsinki. Peripheral blood mononuclear cells (PBMCs) were isolated using a Ficoll density gradient. Subsequently, centrifugal elutriation was performed to separate the monocytes, yielding 70-80% purity.²² Monocyte derived dendritic cells (moDCs) were generated by incubating monocytes (0.84×10^6 /ml) in 6-well plates in Cellgro medium in the presence of 1000 U/mL IL-4 and 800 U/mL GM-CSF (both from Cellgenix).

HLA typing

DNA was extracted using Qiagen DNeasy Blood & Tissue isolation kit (Hilden, Germany) according to manufacturer's protocol. Sanger sequence-based typing for HLA-DR and HLA-DQB was performed in house using SBTessenz (GenDx, Utrecht, The Netherlands) according to manufacturer's protocol. For HLA-DQA, sequencing was performed by GenDX (Utrecht, The Netherlands) using the NGSgo system for amplification and library preparation. Sequencing was performed on Illumina with 2x 150 base pair chemistry. Results were analyzed with NGSengine (GenDX).

Endocytosis of FVIII by moDCs and purification of HLA-DR and HLA-DQ complexes

After 5 days of differentiation, 5×10^6 moDCs were re-plated in a 24-well plate in 1 mL of conditioned medium, supplemented with 100 nM full-length FVIII. After 5 hours of incubation

at 37°C, 5% CO₂, 1µg/mL lipopolysaccharide (Sigma-Aldrich) was added together with 1% fetal calf serum (FCS) for additional 19 hours. The up-regulation of CD40, CD80, CD83, CD86 and HLA-II molecules was assessed by flow cytometry to confirm the mature phenotype of moDCs.

Legend to Supplementary Figure 1

For each group of overlapping FVIII peptide, the NETMHCIIpan3.1prediction tool was run taking into account the two DRB1 alleles in the case of HLA-DR and the four DQA1/DQB1 combination (2 possible alpha and beta chains) in the case of HLA-DQ. Values corresponding to the highest affinity value are depicted in red.

Donor A

HLA-DRB1*07:01 - 14:54

HLA-DQA1*01:04 - 02:01

HLA-DQB1*03:03 - 05:03

A1

a1

A2

B

a3

A3

C1

C2

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*07:01	DRB1*14:54			
KRTLFVEFTDHLF ---LFVEFTDHL-	445.67	869.08	66	78	13
KASEGAEYDDQTSQREKEDDRVFPQGSHTY -----VFQGSHTY	6791.46	10945.36	126	155	30
RESVDQRGNQIMSKRNWILF -----RGNQIMSKRNWIL-	1759.22	792.24	575	595	21
-----IMSKRNVI---			147	155	
DSYEDISAYLLSKNNAIEPR ---EDISAYLLSKNNAIEPR ---DISAYLLSKNNAIEPR ---ISAYLLSKNNAIEPR -----YLLSKNNAI---	22.04	142.46	740	759	20
-----YLLSKNNAI---			583	591	
KMLFLSTRQNVVGS KMLFLSTRQNVG- ---FLSTRQNV---	105.25	119.86	1244	1258	15
-----FLSTRQNV---			1244	1257	14
-----FLSTRQNV---			1246	1254	
QTSATNSVYKRVENVLKPDLPR ---TNSVYKRVENVLKPDLPR -----NSVYKRVENVLKPDL -----YKRVENVL-----	139.00	1010.38	1482	1506	25
-----YKRVENVL-----			1486	1506	21
-----YKRVENVL-----			1487	1503	17
-----YKRVENVL-----			1491	1499	
TSGKVELLPKRVHIYQK -SGKVELLPKRVHIYQK -SGKVELLPKRVHIYQ- ---VELLPKRVHI---	463.81	275.50	1507	1522	16
---VELLPKRVHI---			1508	1522	15
---VELLPKRVHI---			1508	1521	14
---VELLPKRVHI---			1511	1519	
ETTRTTLQSDQREID -ITRRTLQSD-----	7063.96	6386.65	1668	1682	15
-ITRRTLQSD-----			1669	1677	
AQDQRIHWY AQDQRIHWY	18949.16	19761.59	1954	1962	9
AQDQRIHWY			1954	1962	
HYSIRSTLRMEL -YSIRSTLRM--	86.51	156.19	2174	2185	12
-YSIRSTLRM--			2175	2183	
LRTHPQSW ND		ND	2325	2332	8

Peptide	Predicted Affinity (nM)				Start	Stop	Length
	DQA1*01:04/DQB1*03:03	DQA1*01:04/DQB1*05:03	DQA1*02:01/DQB1*03:03	DQA1*02:01/DQB1*05:03			
KRTLFVEFTDHLF ---LFVEFTDHL-	3563.34	323.47	3100.65	339.58	66	78	13
KASEGAEYDDQTSQREKEDDRVFPQGSHTY -----VFQGSHTY	5739.64	2697.56	1765.57	803.92	126	155	30
RESVDQRGNQIMSKRNWILF -----RGNQIMSKRNWIL-					575	595	21
-----IMSKRNVI---					147	155	
DSYEDISAYLLSKNNAIEPR ---EDISAYLLSKNNAIEPR ---DISAYLLSKNNAIEPR ---ISAYLLSKNNAIEPR -----YLLSKNNAI---					740	759	20
-----YLLSKNNAI---					583	591	
KMLFLSTRQNVVGS KMLFLSTRQNVG- ---FLSTRQNV---					1244	1258	15
-----FLSTRQNV---					1244	1257	14
-----FLSTRQNV---					1246	1254	
QTSATNSVYKRVENVLKPDLPR ---TNSVYKRVENVLKPDLPR -----NSVYKRVENVLKPDL -----YKRVENVL-----					1482	1506	25
-----YKRVENVL-----					1486	1506	21
-----YKRVENVL-----					1487	1503	17
-----YKRVENVL-----					1491	1499	
TSGKVELLPKRVHIYQK -SGKVELLPKRVHIYQK -SGKVELLPKRVHIYQ- ---VELLPKRVHI---					1507	1522	16
---VELLPKRVHI---					1508	1522	15
---VELLPKRVHI---					1508	1521	14
---VELLPKRVHI---					1511	1519	
ETTRTTLQSDQREID -ITRRTLQSD-----					1668	1682	15
-ITRRTLQSD-----					1669	1677	
AQDQRIHWY AQDQRIHWY					1954	1962	9
AQDQRIHWY					1954	1962	
HYSIRSTLRMEL -YSIRSTLRM--					2174	2185	12
-YSIRSTLRM--					2175	2183	
LRTHPQSW ND					2325	2332	8
ND							

LSDLQEAKEYTFSDD ---LQEAKEYTF---	4777.13	2520.99	2824.35	1226.06	824	838	15
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Donor B

HLA-DRB1*04:04 - 15:01

HLA-DQA1*01:02 - 03:01

HLA-DQB1*03:02 - 06:02

A1

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*04:04	DRB1*15:01			
AEVYDTVITLKNMASHPV			97	116	20
--EVYDTVITLKNMASHPV			98	116	19
---YVYDTVITLKNMASHPV			99	116	18
----YDTVITLKNMASHPV			100	116	17
-----YTVITLKNMASHPV			100	115	16
-----DTVITLKNMASHPV			101	116	16
-----TIVITLKNMASHPV			101	115	15
-----TVITLKNMASHPV			102	116	15
-----VVITLKNMASHPV			102	115	14
-----VIVITLKNMASHPV			103	115	13
-----VITLKNMAS----	29.76	55.54			

a1

DDNSPSFIQIRSVAKKHPK			380	399	20
--DDNSPSFIQIRSVAKKHPK			381	399	19
---DNSPSFIQIRSVAKKHPK			382	399	18
----NSPSFIQIRSVAKKHPK			383	399	17
-----SPSFIQIRSVAKKHPK			384	399	16
-----SPSFIQIRSVAKKHP-			384	398	15
-----FIQIRSVAR----	43.14	53.69			

A2

EVGDLLIIFKNQASRPYNI			475	494	20
--EVGDLLIIFKNQASRPYNI			475	493	19
---VGDLLIIFKNQASRPYNI			476	493	18
----VGDLLIIFKNQASRPYNI			476	494	19
-----VGDLLIIFKNQASRPY--			476	492	17
-----GDLLIIFKNQASRPY--			477	492	16
-----DILLIIFKNQASRPY--			478	492	15
-----DILLIIFKNQASRPYNI-			478	493	16
-----LIIFKNQAS-----	30.89	39.06			

B

GPSFRQLVSLGPEKSV			1142	1159	18
---GPSFRQLVSLGPEKSV	547.55	1377.99			
STELEKRIIVDDTSTQNSK			1343	1361	19
---LEKRIIVDDTSTQNSK			1346	1361	16
-----IIVDDTSTQ--	1347.98	4244.58			
TAFKKTILSLNACESHAI			1609	1630	22
---KKTILSLNACESHAI			1612	1630	19
----KKTILSLNACESHAI			1612	1628	17
-----KKTILSLNACESHAI-			1612	1629	18
-----KKTILSLNACESH-			1613	1627	15
-----KKTILSLNACESH-			1613	1628	16
-----KKTILSLNACESHAI			1613	1631	19
-----KKTILSLNACESHAI-			1613	1629	17
-----KKTILSLNACESHAI-			1613	1630	18
-----KKTILSLNACESH-			1614	1627	14
-----ILSLNACES-----	91.27	302.99			

a3

EITRTTLLQSDQREID			1668	1682	15
---ITRTTLLQSDQREID	5389.30	11549.92			

A3

VEDNIMVTFRNQASRPYS			1786	1803	18
---VEDNIMVTFRNQASRPYS			1786	1801	16
----VEDNIMVTFRNQASRPYS			1786	1802	17
-----VEDNIMVTFRNQASRPY-			1787	1802	16
-----EDNIMVTFRNQASRPY-			1787	1802	16
-----EDNIMVTFRNQASRPYS			1787	1803	17
-----DNIMVTFRNQASRPYS			1788	1802	15
-----MVTFRNQAS----	83.12	99.79			

C1

LLAPMIHGKIQGARQ-			2094	2110	17
---LAPMIHGKIQGARQ-			2095	2110	16
----LAPMIHGKIQGARQ-			2095	2111	17
-----LAPMIHGKIQGARQ-			2096	2110	15
-----LAPMIHGKIQGARQ-			2096	2111	16
-----LAPMIHGKIQGARQ-			2096	2109	14
-----IHGKIQGA----	196.82	199.23			
ISQFIIMYSLDGKK			2117	2130	14
---FIIMYSLDG--	39.57	70.22			

Peptide	Predicted Affinity (nM)				Start	Stop	Length
	DQA1*01:02/DQB1*03:02	DQA1*01:02/DQB1*06:02	DQA1*03:01/DQB1*03:02	DQA1*03:01/DQB1*06:02			
VYDTVITLKNMASHPV					99	116	18
--YDTVITLKNMASHPV					100	116	17
---YTVITLKNMASHPV					100	115	16
----DTVITLKNMASHPV					101	115	15
-----DTVITLKNMASHPV					101	116	16
-----TIVITLKNMASHPV					101	114	14
-----TVITLKNMASHPV					102	115	14
-----VITLKNMAS----	772.91	340.10	3725.3	985.22			

RASEGAEYDDQTSQREEDRVFPGSHTY					126	155	30
---YDDQTSQRE-----	5177.02	6416.15	1659.83	1437.26			

SPSFIQIRSVAKKHPK					384	399	16
---SPSFIQIRSV-----	1495.49	588.29	7509.19	2079.44			

DTLLIIFKNQASRPY					478	492	15
---LIIFKNQAS----	1460.90	850.44	6127.86	1743.19			

EITRTTLLQSDQREID					1668	1682	15
---ITRTTLLQSDQRE--					1668	1680	13
----ITRTTLLQSDQREI-					1668	1681	14
-----ITRTTLLQSDQRE---					1668	1679	12
-----TLLQSDQRE--	1378.70	1820.63	1033.17	671.86			

VEDNIMVTFRNQASRPY-					1786	1802	17
---VEDNIMVTFRNQASRPYS					1786	1803	18
----VEDNIMVTFRNQASRPY-					1787	1802	16
-----VEDNIMVTFRNQASRPY-					1788	1802	15
-----MVTFRNQAS----	1001.90	570.61	3428.55	930.04			

LAPMIHGKIQGARQ-					2095	2110	16
---LAPMIHGKIQGARQ-					2096	2110	15
----LAPMIHGKIQGARQ-					2096	2111	16
-----LAPMIHGKIQGARQ-					2096	2111	16
-----IHGKIQGA----	656.56	205.32	7275.10	1220.29			

Donor C

	HLA-DRB1*01:01 - 15:01					HLA-DQA1*01:01 - 04:01					HLA-DQB1*05:01 - 06:02				
	Peptide	Predicted Affinity (nM)		Start	Stop	Length	Peptide	Predicted Affinity (nM)				Start	Stop	Length	
		DRB1*01:01	DRB1*15:01				DQA1*01:01/DQB1*05:01	DQA1*01:01/DQB1*06:02	DQA1*04:01/DQB1*05:01	DQA1*04:01/DQB1*06:02					
A1	DTVVITLKNMASHPVSLHA --VVITLKNMASHPVSLHA --VVITLKNMASHPVSLHA -----LKNMASHPV-----	4.66	47.86	101	119	19									
a1	DDNSPSFIQIRSVAKKHPK --DDNSPSFIQIRSVAKKHPK --DNSPSFIQIRSVAKKHPK ---NSPSFIQIRSVAKKHPK ----SPSFIQIRSVAKKHPK -----SPSFIQIRSVAKKHPK -----FIQIRSVAR----	7.02	53.69	380	399	20	NSPSFIQIRSVAKKHPK --SPSFIQIRSVAKKHPK ---PSFIQIRSV-----	10016.53	483.28	5117.88	685.29	383	399	17	
A2	VGDLLIIFKNQASRPYNI VGDLLIIFKNQASRPYNI VGDLLIIFKNQASRPYNI -GDLLIIFKNQASRPYNI --DILLIIFKNQASRPYNI ---DILLIIFKNQASRPYNI -----FNQASRPYNI-----	13.19	39.06	476	494	19									
a2							ISAVLLSKNNNAIEPR ---YLLSKNNNAI----	6459.97	1079.43	3758.62	1150.22	745	759	15	
B	SDTEFKVYFPLIHDR ---FKVYFPLIHDR---	29.55	186.14	1054	1068	15									
	EMLFLPESARWIQRT EMLFLPESARWIQRT ---FLPESARWIQRT---	13.86	147.88	1117	1131	15									
	GPSFRQLVSLGPEKSVGG -----LVSLGPEKSVGG-----	30.15	1377.99	1142	1159	18									
	KKKDTILSNACESNHA -KKDTILSNACESNHA -KKDTILSNACESNHA -KDTILSNACESNHA ---DILSNACESNHA -----ILSNACESNHA-----	34.63	720.43	1612	1628	17									
a3	EITRTTLQSDQREID -ITRTTLQSD-----	5810.81	11549.92	1668	1682	15	EITRTTLQSDQREI -ITRTTLQSD-----	4190.55	1770.77	1088.90	996.42	1668	1681	14	
	KTRHYVIAAVERLMD-- KTRHYVIAAVERLMDYG -TRHYVIAAVERLMD-- ---YVIAAVERLMD-----	9.58	104.21	1713	1727	15									
A3	EVDNIMVTFNQASRPY- -VDNIMVTFNQASRPY- -VDNIMVTFNQASRPY- -VDNIMVTFNQASRPY- -EDNIMVTFNQASRPY- -EDNIMVTFNQASRPY- -EDNIMVTFNQASRPY- -DNIMVTFNQASRPY- -----FNQASRPY-----	24.95	99.79	1785	1802	18									
	IWRVECLIGEHLHAG ---VECLIGEHLHAG---	65.12	314.18	2014	2028	15									
C1	ISQFIIMYSLDGKRW ISQFIIMYSLDGKRW ---FIIMYSLDGKRW---	14.08	63.02	2117	2131	15	APMIIRGIKTQGAROK APMIIRGIKTQGAROK ----IRGIKTQGA----	13223.73	259.37	7849.72	448.59	2096	2111	16	
C2	HPQSWVHQIALRMEVL HPQSWVHQIALRMEVL HPQSWVHQIALRMEVL ---VWVHQIALRMEVL---	5.89	70.99	2328	2343	16									

Donor D

HLA-DRB1*01:01 - 08:01

HLA-DQA1*01:01 - 04:01

HLA-DQB1*04:02 - 05:01

A1

a1

A2

a2

B

a3

A3

C1

C2

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*01:01	DRB1*08:01			
SPSFIQIRSVAKKHPK ---FIQIRSVAK---	7.17	16.63	384	399	16
HPKTVVHYIAAEEDW HPKTVVHYIAAEEDW ---WVHYIAAE---	440.10	541.47	397	412	16
SGYTFKHMVYEDTLT SGYTFKHMVYEDT-- -GYTFKHMVYEDT-- ---YTFKHMVY----	489.99	230.44	673	688	16
DISAYLLSKNNAIEPRS DISAYLLSKNNAIEPR- -ISAYLLSKNNAIEPR- --ISAYLLSKNNAIEPR- ---SAYLLSKNNAIEPR- ----AYLLSKNNAIEPR- -----YLLSKNNAI----	7.74	128.99	744	760	17
SDTEFKKVTPLIHDRML SDTEFKKVTPLIHDR-- SDTEFKKVTPLIHDR-- SDTEFKKVTPLIHDRM- SDTEFKKVTPLIHDR-- ----FKKVTPLIHDR--	26.09	38.12	1054	1070	17
EMFLPESARWIQRTH EMFLPESARWIQR-- ---FLPESARWI---	13.97	143.71	1117	1132	16
GPSFRQLVSLGPEKSVESG ----LVSLGPEKSVESG---	30.15	530.95	1142	1159	18
IFKKEKSKQKSPK ----WKSQKSPK---	10172.05	6845.46	1594	1608	15
SPKTAKKKDTILSLN ----FKKDTILSLN---	64.30	142.82	1605	1621	17
KKKDTILSLNACESHNA- -KKDTILSLNACESHN- --KDTILSLNACESHNAI- ---KDTILSLNACESHNA- ----KDTILSLNACESHN- -----ILSLNACESH---	32.83	358.57	1612	1628	17
EITRTTLLSQDQREID -ITRTTLLSQD-----	5810.81	2555.92	1668	1682	15
KTRHYFIAAVERLWD -KTRHYFIAAVERLWD --TRHYFIAAVERLWD ---YFIAAVERL---	9.78	119.71	1712	1727	16
KPKETKTYFKVQHMAFT -----FKVQHMAFT---	41.6	36.59	1827	1845	19
ISQFIIMYSLDGKRW ISQFIIMYSLDGK-- ---FIIMYSLDG---	14.08	125.95	2117	2131	15
ASSYFTNMFATWSPEK ---FTNMFATWS---	43.84	308.78	2211	2226	16
HPQSNVHQIALRMEVL HPQSNVHQIALRMEV- HPQSNVHQIALRMEV- ---WVHQIALRMEV---	5.89	66.19	2328	2343	16

Peptide	Predicted Affinity (nM)				Start	Stop	Length
	DQA1*01:01/DQB1*04:02	DQA1*01:01/DQB1*05:01	DQA1*04:01/DQB1*04:02	DQA1*04:01/DQB1*05:01			
VVITLKNMASHPVVS ----LKNMASHPVV--	6083.99	14141.70	11565.18	9331.33	103	116	14
ISAYLLSKNNAIEPR ----YLLSKNNAI---	3416.59	6459.97	6174.05	3758.62	745	759	15
EITRTTLLSQDQREID ----TLLSQDQRE---	1066.28	3153.29	956.72	766.04	1668	1682	15
SFTPVVNSLD -FTPVVNSLD-	7535.97	19955.18	6718.08	9559.53	2308	2317	10

Donor F

HLA-DRB1*01:01 - 11:01

HLA-DQA1*01:01 - 05:05

HLA-DQB1*03:01 - 05:01

A1

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*01:01	DRB1*11:01			
DTVVITLKNMASHPVSLHA			101	119	19
DTVVITLKNMASHPVSLHA			101	116	16
--VVITLKNMASHPVSLHA			103	119	17
-----LKNMASHPV-----	4.66	34.47			
TLHKFILLFAVFDGK			210	225	16
TLHKFILLFAVFDGKS			210	226	17
-----FILLFAVFD-----	274.95	1010.31			

A2

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*01:01	DRB1*11:01			
YGEVGDILLIFKNQASRPYNIYPHG			473	498	26
--GEVGDILLIFKNQASRPYNIYPHG			474	493	20
-----EVGDILLIFKNQASRPYNIYPHG			475	493	19
-----EVGDILLIFKNQASRPYNIYPHG			475	492	18
-----VGDILLIFKNQASRPYNIYPHG			476	492	17
-----VGDILLIFKNQASRPYNIYPHG			476	493	18
-----DTLLIFKNQASRPYNIYPHG			478	493	16
-----DTLLIFKNQASRPYNIYPHG	10.89	29.76			
-----LIFKNQASRPYNIYPHG					
YSSFYNERDLASG			552	565	14
YSSFYNERDLASGLIG			552	568	17
YSSFYNERDLASG			552	565	14
-----FYNERDLASG	79.74	218.31			
ENIQRFLEPNPAGVLEDFE			608	626	19
--NIQRFLEPNPAGVLEDFE			609	626	18
-----NIQRFLEPNPAGVLEDFE			609	623	15
-----NIQRFLEPNPAGVLEDFE			609	624	16
-----IQRFLEPNPAGVLEDFE	16.52	101.01			

B

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*01:01	DRB1*11:01			
HSDMVFTEPESGLQLRNEK			862	881	20
-----FTEPESGLQLRNEK	37.88	1446.59			
SDTEPKVTFPIIHDR			1054	1068	15
-----PKVTFPIIHDR	29.55	34.57			
KMLFLPESARWIQRTH			1117	1132	16
-----KMLFLPESARWIQRTH			1117	1131	15
-----KMLFLPESARWIQRTH			1117	1130	14
-----FLPESARWIQRTH	13.86	139.38			
GFSPRQLVSLGPEKSVVGG			1142	1159	18
GFPEKSVVGGFLSEKMKVYVGG			1152	1174	23
GFPEKSVVGGFLSEKMKVYVGG			1152	1172	21
-----FLSEKMKVYVGG	29.87	249.2			
KKKDTILSNACENHAI			1612	1629	18
-----KKKDTILSNACENHAI			1612	1628	17
-----KKKDTILSNACENHAI			1613	1627	15
-----KKKDTILSNACENHAI			1613	1628	16
-----KDTILSNACENHAI			1614	1628	15
-----KDTILSNACENHAI			1614	1627	14
-----ILSNACENHAI	32.83	906.6			

A3

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*01:01	DRB1*11:01			
SPQKTRHYFIAAVERLMDYGMSSPHVLRN			1709	1739	31
-----SPQKTRHYFIAAVERLMDYGMSSPHVLRN			1710	1727	18
-----KTRHYFIAAVERLMDYGMSSPHVLRN			1712	1727	16
-----KTRHYFIAAVERLMDYGMSSPHVLRN			1712	1729	18
-----KTRHYFIAAVERLMDYGMSSPHVLRN			1713	1729	17
-----KTRHYFIAAVERLMDYGMSSPHVLRN			1713	1727	15
-----KTRHYFIAAVERLMDYGMSSPHVLRN			1713	1728	16
-----TRHYFIAAVERLMDYGMSSPHVLRN			1714	1727	14
-----TRHYFIAAVERLMDYGMSSPHVLRN			1714	1729	16
-----RHYFIAAVERLMDYGMSSPHVLRN			1715	1729	15
-----YFIAAVERLMDYGMSSPHVLRN	6.75	43.32			
KPKHETKTFKVVQHMAFT			1827	1845	19
-----PKVVQHMAFT	41.60	28.37			
AGIWRVECLIGEHLHA			2012	2027	16
-----AGIWRVECLIGEHLHA			2013	2026	14
-----AGIWRVECLIGEHLHA			2013	2028	16
-----IWRVECLIGEHLHA			2014	2027	14
-----VECLIGEHLHA	61.68	709.38			
HEPFSWIKVLLAPMIHGINT			2084	2105	22
-----VLLAPMIHGINT	17.25	220.43			

C1

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*01:01	DRB1*11:01			
YISQFIIMYSLDGKKW			2116	2131	16
-----YISQFIIMYSLDGKKW			2116	2131	16
-----YISQFIIMYSLDGKKW			2117	2131	15
-----YISQFIIMYSLDGKKW			2117	2131	15
-----YISQFIIMYSLDGKKW			2117	2132	16
-----YISQFIIMYSLDGKKW			2117	2130	14
-----YISQFIIMYSLDGKKW			2117	2132	16
-----FIMYSLDGKKW	14.08	82.99			
ASSYFTNMFATWSPSK			2211	2226	16
-----ASSYFTNMFATWSPSK			2211	2227	17
-----FTNMFATWSPSK	33.43	161.12			
SPSKARLHLQGRSNARFQ			2223	2241	19
-----LHLQGRSNARFQ	170.60	160.56			

C2

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*01:01	DRB1*11:01			
IHPQSWVHQIALRMEVL			2327	2343	17
-----IHPQSWVHQIALRMEVL			2327	2342	16
-----IHPQSWVHQIALRMEVL			2328	2341	14
-----IHPQSWVHQIALRMEVL			2328	2342	15
-----IHPQSWVHQIALRMEVL			2328	2343	16
-----IHPQSWVHQIALRMEVL			2328	2340	13
-----IHPQSWVHQIALRMEVL			2328	2343	16
-----IHPQSWVHQIALRMEVL			2328	2342	15
-----IHPQSWVHQIALRMEVL			2328	2344	17
-----VHQIALRMEVL	5.89	49.88			

Peptide	Predicted Affinity (nM)				Start	Stop	Length
	DQA1*01:01/DQB1*03:01	DQA1*01:01/DQB1*05:01	DQA1*05:05/DQB1*03:01	DQA1*05:05/DQB1*05:01			
TLHKFILLFAVFDGK					210	225	16
-----TLHKFILLFAVFDGK	2628.73	182.3	2031.65	89.34			
DTLLIFKNQASRPYNIYPHG					478	493	16
-----DTLLIFKNQASRPYNIYPHG	1229.03	2944.14	624.25	553.01			
ISQFIIMYSLDGKKW					2117	2131	15
ISQFIIMYSLDGKKW					2117	2131	15
ISQFIIMYSLDGKKW					2117	2130	14
ISQFIIMYSLDGKKW					2117	2130	14
-----ISQFIIMYSLDGKKW	1570.67	668.02	1192.95	171.26			
IHPQSWVHQIALRMEVL					2328	2342	15
IHPQSWVHQIALRMEVL					2328	2343	16
-----IHPQSWVHQIALRMEVL	278.11	1422.51	333.23	207.5			

Donor G

HLA-DRB1*01:01 - 11:01

HLA-DQA1*01:01 - 05:05

HLA-DQB1*03:01 - 05:01

	Peptide	Predicted Affinity (nM)		Start	Stop	Length	Peptide	Predicted Affinity (nM)				Start	Stop	Length
		DRB1*01:01	DRB1*11:01					DQA1*01:01/DQB1*03:01	DQA1*01:01/DQB1*05:01	DQA1*05:05/DQB1*03:01	DQA1*05:05/DQB1*05:01			
A1	DTVVITLKNMASHPV			101	116	16								
	-----LKNMASHPV-	5.81	34.47											
	TLRKFILLFAVFEGR			210	225	16								
	----FILLFAVFD---	274.95	1185.56											
A2	EVGDILLIFKNQASRPYN			475	493	19								
	-VGDILLIFKNQASRPYN			476	493	18								
	-VGDILLIFKNQASRPY-			476	492	17								
	----LIFKNQAS----	16.73	41.19											
	YSSFVNERDLASG			552	565	14								
	---FVNERDLA---	142.54	218.31											
	ENIQRFLEPAGVQLEDFE			608	626	19								
	-NIQRFLEPAGVQLEDFE			609	626	18								
	-NIQRFLEPAGVQLED--			609	624	16								
	---IQRFLEPDA-----	18.44	112.51											
a2	ISAVLLSKNNAIEPR			745	759	15								
	---YLLSKNNAI---	7.74	82.32											
	SQTEFKVYFPLIHDR			1054	1068	15								
	----FKVYFPLIH---	29.55	34.57											
B	KKDTILSLNACSNH-			1613	1627	15								
	KKDTILSLNACSNHA			1613	1628	16								
	KDTILSLNACSNH-			1614	1627	14								
	---ILSLNACSNH---	34.63	1034.52											
A3	WTRHYFIAAVERLNDYG			1713	1729	17								
	WTRHYFIAAVERLND--			1713	1727	15								
	-WTRHYFIAAVERLND--			1714	1727	14								
	----YFIAAVERLND---	9.58	67.77											
C1	ISQFIIMYSLDGKRW			2117	2131	15						2117	2131	15
	ISQFIIMYSLDGK--			2117	2130	14								
	---FIIMYSLDG---	14.08	85.91				1570.67	668.02	1192.95	171.26				
C2	ASSYPTMFAWSPSK			2211	2226	16								
	-SSYPTMFAWSPSK			2212	2226	15								
	----PTMFAWSPSK---	40.43	295.73											
	HPQSNVHQIALRMEV			2328	2340	13						2328	2340	13
	HPQSNVHQIALRMEV			2328	2342	15						2328	2340	13
	HPQSNVHQIALRME-			2328	2341	14								
	----VHQIALRME---	6.61	57.29				278.11	1422.51	333.23	207.5				

Donor H

HLA-DRB1*04:01 - 11:01

HLA-DQA1*03:01 - 05:05

HLA-DQB1*03:01 - 03:02

A1

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*04:01	DRB1*11:01			
DTVVITLKNMASHPVS ---VITLKNMAS---	41.45	34.47	101	116	16
TKNSLMQDRDASARA-- TKNSLMQDRDASARAWP ---LMQDRDAS---	650.65	3552.60	231	246	16

A2

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*04:01	DRB1*11:01			
GEVGDILLIIFRNQASRFYN -EVGDILLIIFRNQASRFYN -EVGDILLIIFRNQASRFY- --VGDILLIIFRNQASRFYN --VGDILLIIFRNQASRFY- ---GDILLIIFRNQASRFYN -----LIIIFRNQAS----	42.37	41.19	474	493	20
YSSFVNMERDLASG ---FVNMERDLA--	388.22	218.31	552	565	14
ENIQRFLEPNFAGVLEDFE ENIQRFLEPNFAGVLEDFE -NIQRFLEPNFAGVLEDFE -NIQRFLEPNFAGVLEDFE ---IQRFLEPNFAGVLEDFE	77.99	101.01	608	626	19

B

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*04:01	DRB1*11:01			
LSDLQAKYVETFSDD -SDLQAKYE----	7142.28	8730.43	824	838	15
ESDTEFKKVTPLIHDM -SDTEFKKVTPLIHDR- ----FKKVTPLIH--	142.17	33.21	1053	1069	17
LEKRIIVDDSTQWSKN -LEKRIIVDDSTQWSKN --KRIIVDDSTQWSK- ----IIVDDSTQ---	626.93	4929.66	1345	1362	18
KVFLRVATESSAK ---FLRVATESS--	27.52	62.03	1562	1575	14

a3

Peptide	Predicted Affinity (nM)				Start	Stop	Length
	DQA1*03:01/DQB1*03:01	DQA1*03:01/DQB1*03:02	DQA1*05:05/DQB1*03:01	DQA1*05:05/DQB1*03:02			
EITRTTLDSDQEE ----TTLQSDQEE	3885.56	1625.18	5669.03	1696.01	1668	1680	13

A3

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*04:01	DRB1*11:01			
WTRHYTAAVERLMDYG ----YFIAAVERL----	125.64	75.32	1713	1729	17
DNIMVTFRNQSREYS ----FRNQSREYS--	62.00	52.28	1788	1803	16

C1

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*04:01	DRB1*11:01			
APMIHGKIQGARQ ---IHGKIQGA--	185.00	198.88	2096	2110	15
YISQPIIMYSLDGKRW-- YISQPIIMYSLDGKRWQT -ISQPIIMYSLDGKRW-- -ISQPIIMYSLDGKRWQ- -ISQPIIMYSLDGKRW- ----FIIMYSLDG----	136.62	82.99	2116	2131	16

Peptide	Predicted Affinity (nM)				Start	Stop	Length
	DQA1*03:01/DQB1*03:01	DQA1*03:01/DQB1*03:02	DQA1*05:05/DQB1*03:01	DQA1*05:05/DQB1*03:02			
ISQPIIMYSLDGKRW ISQPIIMYSLDGKRW -SQPIIMYSLDGKRW--	1749.71	1807.85	1192.95	854.14	2117	2131	15
ISQPIIMYSLDGKRW ISQPIIMYSLDGKRW -SQPIIMYSLDGKRW--					2117	2130	14

C2

Peptide	Predicted Affinity (nM)		Start	Stop	Length
	DRB1*04:01	DRB1*11:01			
SPKARLHLQGRSNARFQ ----LHLQGRSNA----	1019.80	160.56	2223	2241	19
YVKEFLISSQDGHWT YVKEFLISSQDGHQ-- YVKEFLISSQDGHQW- --KEFLISSQDGHQ-- --KEFLISSQDGHQW- ----FLISSQDGH----	87.13	482.04	2275	2291	17

