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First report on the antibody verification of MICA epitopes recorded in the HLA epitope registry

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Summary

The International Registry of HLA Epitopes (http://epregistry.com.br) has been recently established as a tool to understand antibody responses to HLA mismatches. These epitopes are defined structurally by three-dimensional molecular modelling and amino acid sequence differences between HLA antigens. A major goal was to identify HLA epitopes that have been verified experimentally with informative antibodies. This report addresses the identification of MICA epitopes. Our analysis included published information about MICA antibody reactivity in sera from sensitized patients as well as data from our own laboratories. This report describes twenty-one MICA epitopes verified with antibodies which have primarily been tested in Luminex assays with single alleles. The epitopes correspond to distinct eplets that are often defined by single residues. The Registry is still a work-in-progress and will become a useful resource for HLA professionals interested in histocompatibility testing at the epitope level and investigating antibody responses to HLA mismatches in transplant patients.

Introduction

It is now well recognized that antibodies to MICA mismatches may induce allograft rejection and transplant failure (Mizutani *et al.*, 2006; Zou *et al.*, 2006, 2007; Panigrahi *et al.*, 2007; Suarez-Alvarez *et al.*, 2007; Terasaki *et al.*, 2007; Stastny *et al.*, 2009; Zou & Stastny, 2010; Cox *et al.*, 2011; Lu *et al.*, 2011; Lyu *et al.*, 2012; Chaudhuri *et al.*, 2013; Sanchez-Zapardiel *et al.*, 2013). Such antibodies recognize epitopes which can now be defined structurally by HLA molecular modelling and amino acid sequence comparisons. Under auspices of the 16th International Histocompatibility and Immunogenetics Workshop, we have developed a website http://epregistry.com.br (EpRegistry) for the International Registry of HLA Epitopes (Duquesnoy *et al.*, 2013). Its goal was to document a repertoire of HLA epitopes that have been verified with specific antibodies and therefore might be clinically relevant. EpRegistry has five epitope databases: HLA-ABC, HLA-DRB, HLA-DQ, HLA-DP and MICA. Each database is a list of potential epitopes defined by eplets which are small configurations of polymorphic amino acid residues in sequence locations on the HLA molecular surface. This is the first report on MICA epitopes which have been verified with informative antibodies and recorded in EpRegistry. Other reports describe current antibody-verified HLA-ABC and DRDQDP epitopes (Duquesnoy *et al.*, 2014a,b).

Methods

Update of the MICA eplet repertoire

EpRegistry has a MICA epitope database which has 58 eplets determined by molecular modelling of HLA structures and residues within 3.5 Ångstroms of surface-exposed polymorphic residues as previously described (Duquesnoy *et al.*, 2008). They are considered essential elements of MICA epitopes, and they are annotated by distinct sequence position numbers and residue descriptions with standard single-letter amino acid codes. Most MICA eplets are described by single residues in polymorphic sequence positions on the molecular surface. The original design of the MICA eplet repertoire was solely based on polymorphic residue differences within MICA itself (Duquesnoy *et al.*, 2013). It did not consider any interlocus comparisons with the structurally highly similar MICB

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locus which is expressed on endothelial cells and other tissues (Hankey *et al.*, 2002; Quiroga *et al.*, 2006; Stastny *et al.*, 2009; Wei *et al.*, 2010) and also polymorphic though not as much as MICA (Bahram *et al.*, 1996). The HLA-ABC eplet repertoires have always considered interlocus comparisons, and this has resulted in eplets shared by alleles encoded by two or three HLA-ABC loci (Duquesnoy, 2006). Certain eplets consist of residues that are polymorphic for one locus but monomorphic for another locus. Such eplets are excluded from the HLA-ABC repertoire because they are self and cannot be considered as mismatches eliciting HLA antibody responses.

We have reassessed the MICA eplet repertoire by considering interlocus comparisons between MICA and MICB residues. Table 1 shows the amino acid sequences for polymorphic residue positions of 28 MICA alleles selected from the panels in commercially available Luminex kits (Immucor, Life Codes; ThermoFisher, One Lambda) compared to residues on MICB virtually all of which are monomorphic. Eighty per cent of positions in the 1–273 sequence have identical residues, and this demonstrates a high sequence similarity between MICA and MICB. Twenty-eight MICA positions are polymorphic; they are generally defined by two different amino acid residues distinct for MICA. For instance, MICA has 14G and 14W, whereas all MICB alleles have the monomorphic 14Q. Other MICA positions have one distinct residue but the other residue is also present on MICB. As an example, position 24 has threonine (24T) on MICA*001, MICA*012 and MICA*018, whereas the other MICA alleles have an alanine (A) residue which is also present on all MICB alleles. Table 1 lists 26 polymorphic MICA residues that are monomorphic for MICB. The amino acids are shown with standard codes but with lower case letters. The question must be raised whether such residues could define eplets that might be considered mismatches and capable of inducing antibody responses.

With structural models of crystallized MICA and MICB molecules, we have used the Cn3D program (Hogue, 1997) to identify the amino acids within 3.5 Ångstrom radius of a polymorphic residue (shown with lower case font) and to determine whether the composition of such configuration has MICA-specific residues which are absent on MICB. Table 2 shows two patterns. In 14 cases, all residues within the 3.5 Ångstrom radius are identical for MICA and MICB. This means that such configuration is monomorphic for MICB and therefore must be considered as self. For instance, 91q (on all MICA alleles except MICA*017 which has 91R) is surrounded by residues in positions 89, 90, 109, 110 and 111, all of which are monomorphic at MICB. In other words, 89S + 90L + 91Q + 109H + 110F + 111Y must be considered as a true self-configuration. On the other hand, an arginine residue (R) in position 91 could define a MICA epitope. Similarly, 129v has corresponding 117F + 128T + 129V + 130P which cannot serve as a nonself eplet because all MICB alleles have this configuration. Again a methionine (M) in position 129 leads to a distinct MICA eplet.

The second pattern involves ten cases whereby there are residue differences between MICA and MICB in the 3.5 Ångstrom patch (Table 2). This means that a distinct residue present on MICA but not on MICB might contribute to an eplet defined by a polymorphic residue. For example, 36y is surrounded by residues in positions 23, 24, 35, 37, 40 and 41. All of them are the same on MICA and MICB except one in position 41 which has 41C on MICA but 41R on MICB. In other words, the combination of 36y and 41C is distinct for MICA but absent on MICB. This suggests the possibility of a MICA epitope represented by the 36Y eplet which can be described by $36Y + 41C^{m}$; the superscript indicates a monomorphic residue unique to MICA. As another example, 105r has a nearby 106S expressed by MICA but not MICB, so we can define the 105R eplet described by $105R + 106S^{m}$.

Residue 24a is polymorphic at MICA but monomorphic at MICB. It is surrounded by residues in positions 8, 9, 23, 25, 35 and 36 (Table 2). All of them are identical monomorphic residues on MICA and MICB except position 36 which is polymorphic for MICA. Residue 36C is only on MICA, and together with 24a it could constitute an eplet annotated as 24AC. On the other hand, 36y is monomorphic for MICB and this means that the 24a + 36y combination is self and cannot reflect a nonself eplet. Position 124 has the adjacent 125E or 125K which are absent from MICB and they can give rise to two eplets 124TE and 124TK (Table 2).

Altogether, we have identified 14 polymorphic MICA residues that define eplets which turn out to be monomorphic for MICB (Table 2). As such eplets cannot become mismatches, one might predict that they cannot be immunogenic. To date, we have not identified any antibodies or seen any reports of antibodies with specificities that correlate with MICA eplets that are monomorphic for MICB. They will be removed from the MICA eplet repertoire in the near future unless there is documentation of specific antibodies.

Antibody reactivity testing with MICA alleles

Antibody verifications of MICA epitopes have been mostly carried out by testing patient allosera and eluates of absorbed sera in Luminex assays using single-allele kits prepared by the laboratory or obtained from commercial vendors. These kits have only recently become available, and the allele panels varied considerably in size and quality. Altogether, we considered antibody reactivity patterns if the Luminex kits included ten or more informative alleles selected from the following group: MICA*001, *002*, 004, *005, *006, *007, *008, *009, *011, *012, *015, *016, *017, *018, *019, *024, *028, *029, *030, *033,

Table 1. Comparisons of amino acid sequence differences between selected MICA alleles and MICB

Position	A* 00 01	A* 00 02	A* 00 04	A* 00 05	A* 00 06	A* 00 07	A* 00 08	A* 00 09	A* 00 11	A* 00 12	A* 00 15	A* 00 16	A* 00 17	A* 00 18	A* 00 19	A* 00 24	A* 00 28	A* 00 29	A* 00 30	A* 00 33	A* 00 36	A* 00 37	A* 00 41	A* 00 42	A* 00 43	A* 00 46	A* 00 50	A* 00 51	M 1 CB
14	W	G	W	W	W	W	W	W	G	W	G	W	G	W	W	W	W	W	G	W	G	W	G	W	W	G	G	W	Q
24	T	a	a	a	a	a	a	a	a	T	a	a	a	T	a	a	a	a	a	a	a	a	a	a	a	a	a	a	A
26	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	g	V	V	V	V	V	V	V	G
36	C	C	y	y	y	C	y	y	C	C	C	y	C	C	y	y	y	C	C	y	C	C	y	C	C	C	C	y	Y
90	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	F	I	L
91	q	q	q	q	q	q	q	q	q	q	q	q	R	q	q	q	q	q	q	q	q	q	q	q	q	q	q	q	Q
105	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	K	r	r	r	r	r	r	r	R
114	g	g	g	g	g	g	g	g	g	g	R	g	g	g	g	g	g	g	g	g	g	g	g	g	g	g	g	g	G
122	I	I	V	I	V	I	I	V	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	L
124	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	S	t	t	t	t	t	t	t	t	T
125	K	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	Q
129	M	M	v	v	v	M	v	v	M	M	M	v	M	M	v	v	v	M	M	v	M	M	M	M	M	M	M	M	V
142	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	I	v	v	v	v	v	v	v	v	v	v	V
151	m	m	m	m	m	m	m	m	V	m	m	m	m	m	m	m	m	m	m	m	m	m	m	m	m	m	m	m	M
156	h	h	h	h	h	h	h	h	h	L	h	h	h	h	h	h	h	h	h	h	h	h	h	h	R	h	h	h	H
173	k	k	E	k	E	k	E	E	k	k	k	E	k	k	E	E	E	k	k	E	E	k	k	k	k	k	k	k	K
175	g	g	S	g	S	g	g	S	g	g	g	S	g	g	S	g	g	g	g	S	S	g	g	g	g	g	g	g	G
176	v	v	v	v	I	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	V
181	t	t	R	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	t	T
206	G	G	s	s	s	G	s	s	G	G	G	s	G	G	s	s	G	G	G	s	G	s	G	s	G	G	G	G	S
208	y	y	y	y	y	y	y	y	y	y	y	y	y	y	y	y	y	y	y	y	y	y	y	y	y	C	y	y	Y
210	W	W	r	r	r	W	r	r	W	W	W	r	W	W	r	r	W	W	W	r	W	r	W	r	W	W	W	W	R
213	t	t	t	t	t	t	I	t	t	t	t	I	t	t	I	t	t	t	t	I	t	I	t	I	t	t	t	t	T
215	S	S	t	t	t	S	t	t	S	S	S	t	S	S	t	t	S	S	S	t	S	t	S	t	S	S	S	S	T
221	v	v	v	v	v	v	v	v	v	v	v	L	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	v	V
251	q	q	q	R	q	q	R	q	q	q	q	R	q	q	R	q	q	q	q	R	q	R	q	R	q	q	q	q	Q
256	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	S	r	r	r	R
271	p	p	p	p	p	p	p	p	A	p	p	p	p	p	p	p	p	p	A	p	p	p	p	p	p	p	p	p	P

Table 2. Identification of MICA eplets defined by residues that are monomorphic at MICB but share other residues that are distinct for MICA

Residue	Exposure	Positions and residues within 3.5 Angstroms							Eplet	Residue description
24a	+/–	8	9	23	24a	25	35	**36y/C**	24AC (not 24ay)	24A + 36C
36y	+/–	23	24a	35	36y	37	40	**41C**	36Y	36Y + 41C^{m}
90l	+/–	73	89	90l	91q	111			None	
91q	+	89	90l	91q	109	110	111		None	
105r	++	96	97	99	104	105r	**106S**	123	105R	105R + 106S^{m}
114g	+	112	113	114g	115				None	
122l	+	**106S**	120	121	122l	123			122L	122L + 106S^{m}
124t	++	121	123	124t	**125E/K**	126			125T and 125K	124T + 125E and 124T + 125K
129v	++	117	128	129v	130				None	
142v	+	138	140	141	142v	**143R**	144	145	142V	142V + 145R^{m}
151m	++	147	148	149	150	151m	152		None	
156h	++	154	155	156h	157	**158H**	159	160	156H	156H + 158H^{m}
173k	++	**169R**	170	171	172	173k	174	175 g	173K	173K + 169R^{m}
175g	+	171	172	173k	174	175g	176	**177V**	175G	175G + 177V^{m}
181t	+	180	181t	182	208				None	
206s	+	182	205	206s	207	236	240	241	None	
208y	++	180	181	207	208y	209			None	
210r	++	209	210r	211	212				None	
213t		212	213t	214					None	
215t	++	214	215t	216	222	260			None	
221v	++	217	218	219	220	221v	222		None	
251q	++	**250C**	251q	252	253				251Q	251Q + 250C^{m}
256r	++	217	218	219	253	254	256r	257	None	
271p	++	180	270	271p	272				None	

*036, *037, *041, *042, *043, *046, *050 and *051. Early studies were often carried out with fewer than ten MICA alleles and this caused incomplete interpretations of many antibody reactivity patterns. Collaborative studies have also shown variability of MICA antibody reactivity patterns between laboratories (Stastny & Zou, 2013). There are also variations of the allele reactivity in the different Luminex kits, for instance MICA *001, *018 and *019 were highly reactive in one vendor's kit but not in other kits (Cox, 2011). Many published studies addressed antibody specificities against MICA epitopes, whereas other analyses only distinguished between reactive and non-reactive alleles. For the latter, we used HLAMatchmaker to determine epitope specificities of MICA reactive antibodies with informative reactivity patterns.

Antibody-verified epitopes can be classified as 'confirmed' or 'provisional' depending on the amount of information available including how often specific antibodies have been identified and the completeness of reactivity patterns with informative MICA panels. Special consideration has been given to the reactivity of eluates of selectively absorbed allosera and the availability of MICA types for the antibody producer and immunizer. Although many human monoclonal antibodies are HLA class I-specific and class II-specific, we are unaware of their specific recognition of MICA epitopes. Future studies may permit upgrades from 'provisional' to 'confirmed' status if additional experimental support becomes available. Conversely, there could be downgrades to 'questionable' status if new data contradict previous interpretations.

Results

This report summarizes our analysis of published data on antibody-verified MICA epitopes by four investigator groups as well as unpublished data in our own laboratories. Zou and Stastny (Dallas, TX) have pioneered this area (Zou & Stastny, 2009, 2010, 2011; Zou *et al.*, 2009; Stastny & Zou, 2013). Their studies included mouse cells transfected with different MICA alleles which were tested in absorption and elution experiments using patient sera with MICA antibodies (Zou *et al.*, 2009). During the 16th International Workshop, Stastny conducted a multilaboratory study on MICA antibodies (Stastny & Zou, 2013). El-Awar and Terasaki (Los Angeles, CA) studied MICA specificities of antibodies in sera and eluates from transplant recipients (El-Awar *et al.*, 2007). Suarez-Alvarez and Lopez-Larrea (Oviedo, Spain) tested MICA antibody-containing sera from transplant patients with Luminex panels and a synthesized library of overlapping MICA peptides (Suarez-Alvarez *et al.*, 2009). They identified antigenic regions with polymorphic residues, thereby suggesting the 'linearity' of several MICA epitopes. The Pittsburgh group analysed MICA epitope specificities of antibodies in sera from transplant patients (Duquesnoy *et al.*, 2008). The dissertation studies by Steven Cox have also addressed the antibody recognition of MICA epitopes (Cox,

2011). This report also includes unpublished data from co-author Mostecki who has carried out extensive analyses of antibody reactivity patterns with large MICA allele panels.

Studies by two different teams of investigators have identified two separate groups of MICA epitopes on alleles sharing distinct amino acid residues in six sequence positions (Duquesnoy *et al.*, 2008; Zou *et al.*, 2009). Group G1 (or supereplet CMGWS) is on alleles with the 36C, 129M, 173K, 206G, 210W and 215S configuration and the group G2 (or supereplet AYVE) is on MICA alleles which have 36Y, 129V, 173E, 206S, 210R and 215T. As these residues are widely distributed on the molecular surface, they cannot make contact with the same antibody and therefore cannot comprise a single epitope. These studies were based on antibody reactivity patterns with rather small Luminex panels, and their composition was insufficient for a distinction of separate epitopes within these groups. Zou *et al.* showed that absorptions with selected MICA alleles on transfected cells and the use of hybrid MICA molecules with residue substitutions in selected sequence positions (MICA*002 with K173E and MICA*008 with V129M) were helpful in the interpretation of certain G1 and G2 reactive antibodies (Zou *et al.*, 2009). Their findings suggested that 36C, 206G, 210W and 215S might be essential for G1 and that G2 would just reflect 206S, 210R and 215T. These MICA groups have several residues which as noted in Table 2, might be nonimmunogenic because they are monomorphic at MICB. Accordingly, G2 could be reduced to 36Y and 173E.

The Luminex panel assembled by co-author Mostecki has informative MICA alleles that can clarify the antibody specificity towards epitopes within the G1 and G2 groups. This panel consists of 28 alleles: MICA*001, *002, *007, *011, *012, *015, *017, *018, *029, *030, *041, *043, *046 and *050 have the CMKGWS motif, whereas MICA*004, *006, *008, *009, *016, *019, *024 and *033 have the YVESRT motif. However, none of these alleles can distinguish which residue configurations in these motifs are recognized by MICA-specific antibodies. We have added six alleles with variations from these motifs: MICA*005 (YVKSRT), MICA *028 (YVEGWS), MICA *036 (CMEGWS), MICA *037 (CMKSRT), MICA *042 (YMKSRT) and MICA *051 (YMKGWS), and they could be informative in the interpretation of epitope specificity of MICA-specific antibodies (Table 3A).

Table 3 shows five antibody reactivity patterns specific for MICA antibody epitopes within the CMKGWS and AYVESRT motifs. Panel 3B describes the 36C-specific alloserum GP-MIC-6. Positive reactions were seen with 36C-carrying alleles MICA*036 and MICA*037, while 36Y-carrying MICA*005, MICA*028, MICA*042 and MICA*051, which share other residues in the CMKGWS motif, were negative. Moreover, an eluate of MICA*037 absorbed serum showed the same 36C-specific reactivity pattern (data not shown). Conversely, Panel 3C shows the pattern of 36Y-specific antibodies; the 36Y-carrying MICA*005, MICA*028, MICA*042 and MICA*051 were reactive, whereas the 36C-carrying MICA*036 and MICA*037 were nonreactive although they share residues with the AYVESRT motif. The 129M epitope is in the CMKGWS motif, and panel 3D shows that alloserum GP-MIC-9 is specific for 129M as evidenced by the positive reactions with the informative MICA *036, MICA *037, MICA *042 and MICA *051 and the negative reactions with the 129V-carrying MICA*005 and MICA *028. The CMKGWS motif has also the 173K-defined and the 206GWS-defined epitopes which as shown in panels 3E and 3F have been antibody verified with these informative panels.

Table 4 lists the MICA epitopes verified so far with specific antibodies. Most epitopes correspond to eplets, and each one has a polymorphic residue description, a list of eplet-carrying alleles in Luminex panels, specific antibody sources (A: alloserum; E: serum eluate; M: mouse mAb) and cited publications. EpRegistry has more detailed documentation about the antibody verification of epitopes and their presence on Luminex and non-Luminex alleles.

At present, EpRegistry has twenty antibody-verified MICA epitopes which correspond to single eplets residing in sequence locations 14–221 (Table 4). Nine epitopes have confirmed status, but eleven epitopes must be considered provisional because of incomplete antibody reactivity information; 105R was defined only by a mouse monoclonal antibody. The antibody verification of these epitopes can be upgraded to confirmed status if additional antibodies have been tested with informative panels especially if MICA typing has been done on antibody producer and immunizer.

Six epitopes (125K, 151V, 156L, 176I, 181R and 221R) are on single MICA alleles, but all others are shared between multiple MICA alleles. There is also provisional evidence that one additional antibody-defined MICA epitope corresponds to a pair of residues 14W and 36C which are located about 18 Ångstroms apart on the molecular surface.

Discussion

This is the first report about antibody-verified MICA epitopes in the HLA Epitope Registry. It describes twenty-one MICA epitopes recognized by antibodies that have primarily been tested in Luminex assays with single alleles. Twenty epitopes correspond to distinct eplets that are often defined by single residues.

Needless to say, the current repertoire of antibody-verified epitopes must be considered incomplete, but we plan to update EpRegistry with new data. We invite HLA professionals to submit informative data about MICA reactive antibody reactivity patterns possibly specific for new and not so well-described epitopes. This applies also to publications on antibody-defined epitopes which have not been cited in this

Table 3. Antibody reactivity patterns specific for MICA epitopes in the G1 and G2 groups

A. Informative alleles for MICA epitopes in the G1 and G2 groups							B. Alloserum GP-MIC-6		C. Alloserum GP-MIC-4 eluate after absorption with MICA*051	
								GP MFI		GP MFI
MICA allele	36	129	173	206	210	215	Positive control	18 805	Positive control	12 577
G1 group A*001, etc.	C	M	K	G	W	S	Negative control	533	Negative control	96
G2 group A*004, etc.	Y	V	E	S	R	T	36C+ MICA*001	6943	36Y+ MICA*004	2613
A*005	Y	V	K	S	R	T	36C+ MICA*002	7311	36Y+ MICA*005	2418
A*028	Y	V	E	G	W	S	36C+ MICA*007	7997	36Y+ MICA*006	2158
A*036	C	M	E	G	W	S	36C+ MICA*011	5752	36Y+ MICA*008	2552
A*037	C	M	K	S	R	T	36C+ MICA*012	8141	36Y+ MICA*009	2600
A*042	Y	M	K	S	R	T	36C+ MICA*015	6774	36Y+ MICA*016	2377
A*051	Y	M	K	G	W	S	36C+ MICA*017	6458	36Y+ MICA*019	2635
							36C+ MICA*018	7367	36Y+ MICA*024	2474
							36C+ MICA*029	7796	36Y+ MICA*028	1692
							36C+ MICA*030	5996	36Y+ MICA*033	1971
							36C+ MICA*036	6204	36Y+ MICA*042	2377
							36C+ MICA*037	9205	36Y+ MICA*051	1860
							36C+ MICA*041	5934	36Y-negative alleles 375 ± 296	
							36C+ MICA*043	7465		
							36C+ MICA*046	8112		
							36C+ MICA*050	5855		
							36C_negative alleles	824 ± 344		

D. Alloserum GP-MIC-9		E. 16th IHIW alloserum S006 eluate after absorption with MICA*005		F. Alloserum #2 eluate after absorption with MICA*028	
	GP MFI		GP MFI		GP MFI
Positive control	23 684	Positive control	16 097	Positive control	11 713
Negative control	185	Negative control	209	Negative control	50
129M+ MICA*001	7606	173K+ MICA*001	3719	206GWS+ MICA*001	954
129M+ MICA*002	3472	173K+ MICA*002	6068	206GWS+ MICA*002	1618
129M+ MICA*007	10 112	173K+ MICA*005	4953	206QWS+ MICA*007	1401
129M+ MICA*011	1780	173K+ MICA*007	5456	206GWS+ MICA*011	1111
129M+ MICA*012	8258	173K+ MICA*011	4523	206GWS+ MICA*012	1100
129M+ MICA*015	3393	173K+ MICA*012	4588	206GWS+ MICA*015	1436
129M+ MICA*017	3443	173K+ MICA*015	5312	206GWS+ MICA*017	1309
129M+ MICA*018	8500	173K+ MICA*017	4784	206GWS+ MICA*018	1230
129M+ MICA*029	8011	173K+ MICA*018	5341	206GWS+ MICA*028	1137
129M+ MICA*030	3744	173K+ MICA*029	4974	206GWS+ MICA*029	1196
129M+ MICA*036	2297	173K+ MICA*030	4816	206GWS+ MICA*030	1229
129M+ MICA*037	8227	173K+ MICA*037	4978	206GWS+ MICA*036	927
129M+ MICA*041	1862	173K+ MICA*041	3342	206GWS+ MICA*041	1343
129M+ MICA*042	1768	173K+ MICA*042	4693	206GWS+ MICA*043	1343
129M+ MICA*043	9214	173K+ MICA*043	5012	206GWS+ MICA*050	1374
129M+ MICA*046	4453	173K+ MICA*046	5213	206GWS+ MICA*051	1448
129M+ MICA*050	3576	173K+ MICA*050	5410	206GWS-negative alleles	90 ± 101
129M+ MICA*051	2097	173K+ MICA*051	5156		
129M-negative alleles	563 ± 225	173K-negative alleles	310 ± 56		

report. The EpRegistry website has instructions of how to submit information about antibody reactivity patterns with HLA panels and the sensitizing event including, if possible, HLA types of antibody producer and immunizer. Moreover, additional data would be helpful such as absorption/elution studies with selected alleles and the use of mutated alleles with specific residue substitutions.

While still being a work-in-progress, the HLA Epitope Registry will become a valuable resource for researchers interested in MICA compatibility at the epitope level and investigating specific antibody responses in sensitized patients.

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Table 4. Antibody-verified MICA epitopes recorded in the HLA Epitope Registry as of 1 January 2014

MICA epitope * = provisional	Residue descriptions	Epitope-carrying Luminex alleles	Antibody source	References
14G	14G	MICA*002,*011,*015,*017,*030,*036,*041, *046,*050	A	Duquesnoy *et al.* (2008), Zou *et al.* (2009), Cox (2011), Mostecki and Balazs (2013)
14W	14W	MICA*001,*004,*005,*006,*007,*008,*009, *012,*016, *018,*019,*024,*028,*029, *033,*037,*042,*043,*051	A	Duquesnoy *et al.* (2008), Suarez-Alvarez *et al.* (2009), Stastny & Zou (2013)
14W+ 36C*	14W pair with 36C	MICA*001,*007,*012,*018,*029,*037,*043	A, E	Mostecki and Balazs (2013), Stastny & Zou (2013)
24T	24T26V36C	MICA*001,*012,*018	A, E	El-Awar *et al.* (2007), Duquesnoy *et al.* (2008), Zou *et al.* (2009), Cox (2011), Mostecki and Balazs (2013)
36C	36C	MICA*001,*002,*007,*011,*012,*015,*017,*018, *029,*030,*036,*037,*041,*043,*046,*050	A, E	Mostecki and Balazs (2013)
36Y	24A26V36Y	MICA*004,*005,*006,*008,*009,*016,*019, *024,*028,*033,*042,*051	E	Mostecki and Balazs (2013), Stastny & Zou (2013)
105R*	105R	All alleles except MICA*036	M	Mostecki and Balazs (2013)
122V*	122V124T	MICA*004,*006,*009	A	El-Awar *et al.* (2007), Zou *et al.* (2009), Cox (2011)
125E*	125E	All alleles except MICA*001	A	Zou *et al.* (2009), Mostecki and Balazs (2013)
125K	125K	MICA*001	A	Zou *et al.* (2009), Cox (2011), Mostecki and Balazs (2013)
129M*	129M	MICA*001,*002,*007,*011,*012,*015,*017,*018,*029,*030, *036,*037,*041,*042,*043,*046,*050,*051	A	Mostecki and Balazs (2013)
151V*	151V	MICA*011	A	Mostecki and Balazs (2013)
156H*	156H	MICA*001,*002,*004,*005,*006,*007,*008, *009,*011,*015,*016,*017,*018,*019,*024, *027,*028,*029,*030,*033,*036,*037,*041, *042,*046,*050,*051	A	Mostecki and Balazs (2013)
156L*	156L	MICA*012	A	Zou *et al.* (2009), Mostecki and Balazs (2013)
173K	173K175G	MICA*001,*002,*005,*007,*011,*012,*015, *017,*018, *029,*030,*037,*041,*042,*043, *046,*050,*051	A, E	Zou *et al.* (2009), Mostecki and Balazs (2013), Stastny & Zou (2013)
173E	173E	MICA*004,*006,*008,*009,*016,*019,*024,*028,*033,*036	M, A	Zou *et al.* (2009), Mostecki and Balazs (2013)
176I*	176I	MICA*006	A	Zou *et al.* (2009)
181R*	181R	MICA*004	A	El-Awar *et al.* (2007), Zou *et al.* (2009), Cox (2011)
206GWS*	206G210W215S	MICA*001,*002,*007,*011,*012,*015,*017,*018, *028,*029,*030,*036,*041,*043,*050,*051	A, E	Mostecki and Balazs (2013), Stastny & Zou (2013)
$206S_2$*	206S208Y210R (215T)	MICA*004,*005,*006,*008,*009,*016,*019, *024,*033,*037,*042	A	Zou *et al.* (2009)
213I	213I	MICA*008,*016,*019,*033,*037,*042	A, E	El-Awar *et al.* (2007), Zou *et al.* (2009), Cox (2011), Mostecki and Balazs (2013), Stastny & Zou, 2013)
221L*	221L	MICA*016	A	Mostecki and Balazs (2013)

A, alloserum; E, serum eluate; M, mouse monoclonal antibody.

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