의학 석사 학위논문

Association of donor-specific anti-MHC class I-related chain A (MICA) antibodies with renal allograft outcome

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A thesis of the Degree of Master of Science in Medicine

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Abstract

Association of donor-specific anti-MHC class I-related chain A (MICA) antibodies with renal allograft outcome

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Background. Association of anti-MHC class I-related chain A (MICA) with kidney allograft rejection was suggested. However, associations of donor-MICA specific antibodies (dsMICA) and strength of dsMICA with allograft outcome were not elucidated yet.

Methods. From November 2009 to June 2016, 125 remaining sera from renal transplantation recipients with no HLA antibody and biopsy-proven acute rejection (AR) (n = 13), AR + Interstitial Fibrosis and Tubular Atrophy (IFTA) (n = 13), IFTA only (n = 12),

IFTA + borderline change (n = 15), borderline change only (n = 18) and normal biopsy (n = 54) were screened for MICA antibody. MICA antibody identification was performed with LABScreen MICA (One Lambda, USA) on sera with positive MICA screening. Patients with positive MICA screening with available residual donor DNA for MICA genotyping (n = 12) were also analyzed for dsMICA.

Results. Among 125 sera, 19 were positive for MICA antibody (15.2%). dsMICA was positive on 5 out of 12 sera analyzed (41.7%). Neither MICA antibodies nor dsMICA was associated with acute rejection (AR). However, IFTA was significantly associated with MICA positivity (OR = 3.84; 95% CI = 1.34 - 9.98, P = 0.009). The MFI value of dsMICA was significantly higher in patients with IFTA II or III (n = 3, median ± SE, 21919.0 ± 2581.0) compared to patients without IFTA II or III (n = 9, median ± SE, 500.0 ± 155.8) (P = 0.009).

Conclusion. MICA was associated with IFTA in patients without HLA antibody. The association of strength of dsMICA with IFTA severity was suspected. MICA or dsMICA was not associated with acute rejection in patients without HLA antibody. The MICA antibody is useful as a predictor of chronic renal damage and as a useful indicator for future treatment strategy.

Keywords: anti-MHC class I-related chain A (MICA) antibodies, graft rejection, renal transplantation

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CONTENTS

INTRODUCTION

The presence of preformed anti-HLA antibodies is a risk factor for antibody mediated rejection and is associated with reduced clinical outcomes in solid organ transplantation, especially in kidney transplantation. The anti-HLA antibodies are well established to risk factor for antibody mediated rejection and graft failure, and laboratory tests such as crossmatching for detection of preformed anti-HLA to donor organ antigen are routinely performed as a part of preparatory tests [1]. Detection and identification of specific anti-HLA antibodies is one of the most important factors of hyperacute rejections. However, a number of other clinically relevant non-HLA antigenic determinants have been reported to be associated with graft outcome in the absence of detectable HLA antibodies [2-5]. Allograft failures may happen without detectable lymphocytotoxic antibodies, supposing that non-HLA antigenic systems may also play a role in acute kidney rejections [2]. These include angiotensin type 1 receptor, endothelial cell antigens, glutathione S-transferase T1, vimentin, and especially MHC class I-related chain A (MICA) [2, 3]. Recent reports revealed that about 10% to 23% of recipients are presensitized to non-HLA antigens [6], and 38% of graft failure is due to non-HLA antigens [5].

Comparing to the fact that association of donor HLA specific antibodies with renal allograft outcomes is well established, studies regarding non-HLA antigen are now gaining attention. The new polymorphic families of MHC class I-related genes, MICA and MICB

were firstly investigated in 1994 [7]. MICA and MICA B gene are located on Chromosome 6p21.3, close proximity to the HLA-B locus. MICA encodes a 62 kDa cell-surface glycoprotein that functioning as innate and adaptive immunity [4, 7]. MICA is expressed in epithelial cells in gastrointestinal tract and keratinocytes, endothelial cells, skin-derived fibroblasts, and monocytes, whereas MICA is not expressed in resting lymphocytes or immature dendritic cells. This is often presumed to be the cause of graft rejection in patients with negative results in conventional crossmatching tests [8]. The structure of MICA is similar to that of HLA class I, it has three extracellular domains, and shows 30% sequence homology with HLA class I. Meanwhile, MICA does not bind to β2 microglobulin unlike HLA class I [9]. Considering that MHC class I and II react with CD8 and CD4, respectively, MICA polymorphic present immune responses due to ligands for the immunostimulatory C-type lectin-like receptor, natural killer group 2, member D (NKG2D). These MICA-NKG2D interactions may activate NK cells, produce cytokines, or induce cytotoxicity through cytotoxic T lymphocytes, which may be responsible for the innate and adaptive immune response of kidney allografts [4, 9].

In several previous studies, association of anti-MICA antibodies with kidney allograft outcome was suggested [4, 5, 10, 11]. Several studies have demonstrated that MICA lowers graft survival in pretransplantation or posttransplantation and is a risk factor for AMR [1, 5, 10, 11 , 12-14]. However, another study insisted that MICA is not statistically related to graft failure or AMR [15-17]. The sensitization event of MICA Ab was identical to that of antibody

generation in HLA antigen before transplantation [15], and MICA antibody lowered the transplantation survivals only accompanying with HLA antibody [14]. A few researches have studied MICA donor specific antibodies, but the effect of MICA antibodies on solid organ transplantation is not clear yet. Also, the roles of donor specific MICA-antibodies (dsMICA) and the strength of dsMICA were also not elucidated.

Considering previous studies, the results of several studies tend to be inconsistent. In particular, the studies of dsMICA have not been studied so far and show contradictory conclusions. Furthermore, since the study was conducted in different races and patient groups [18], it is necessary to study the Korean population in order to apply the impact of MICA antibody in Koreans. Therefore, we analyzed the association of dsMICA antibodies with renal allograft rejection and chronic allograft injury in patients without detectable dsHLA antibodies in Korean renal transplantation patients.

MATERIALS AND METHODS

Subjects

A total of 125 remaining sera during period November 2009 to June 2016 in clinical laboratory of Seoul National University Hospital and from renal transplantation recipients with no HLA antibody and biopsy-proven acute rejection (AR) (n = 13), AR + Interstitial Fibrosis and Tubular Atrophy (IFTA) (n = 13), IFTA only (n = 12), IFTA + borderline change (n = 15), borderline change only (n = 18) and normal biopsy (n = 54) were retrospectively examined. All of the remaining sera were screened for MICA antibody, and the medical records including age, gender, day after kidney transplantation and acute rejection, serum creatinine level, and causes of end stage renal disease were investigated.

MICA antibody identification was performed with LABScreen MICA (One Lambda, USA) on sera with positive MICA screening. Twelve patients who were positive MICA screening and negative HLA class I and class II antibody in LABscreen Mixed assay (One lambda, Canoga Park, CA, USA) and available residual donor DNA for MICA genotyping were additionally analyzed for dsMICA.

This study was conducted under the approval of Institutional Review Board of Seoul National University Hospital (H-1511-027-716) and according to Declaration of Helsinki.

MICA and HLA Antibodies Screening

Screening for MICA and HLA antibodies was performed using LABscreen Mixed assay (One lambda, Canoga Park, CA, USA) with LABScan 100^{TM} Flow analyzer (LuminexCorp., Austin, Texas, USA) according to the manufacturer's instructions. LABscreen Mixed assay find out the presence of HLA Class I/II and MICA antibodies. Utilizing the microbeads coated with purified HLA Class I/II or MICA antigens and pre-optimized reagents, the panel reactive antibody (PRA) test can detect HLA and MICA antibodies in human serum.

After incubate the 20 uL of test serum with 5 uL of LABscreen beads and proceeds the washing step, add the 100uL of PE-conjugated anti-human IgG to each test to label the beads. Any HLA and MICA antibodies present in the test serum bind to the antigen on the beads. LABScan 100^{TM}Flow analyzer detects the fluorescent emission of PE and dye signature from each bead, simultaneously. Acquired data is analyzed to assign positivity of MICA and HLA antibody screening, and specify the antibody. To calculate the reactivity of test serum, non-specific binding is corrected by negative control beads and background values. Normalized background ratio (NBG ratio) is determined by following calculations (Figure 1). Score HLA class I/II and MICA reaction separately, in accordance with reactivity strength of the patient sample for each bead set. Positive reactions are determined by NBG ratio over 2.2 in the LABScreen Mixed assay. If anyone beads in the test is positive, then the results are assigned to positive.

MICA Antibody Identification and donor MICA genotyping

MICA antibody identification was performed with LABScreen MICA assay (One Lambda, Canoga Park, CA, USA) on sera with positivity for MICA screening. Directions for MICA antibody identification assay are similar to the LABScreen Mix assay. After proceed the incubation, washing, and binding of PE-conjugated anti-human IgG process, data acquisition and analysis is conducted. The PRA test can discriminate the specificities of MICA antibodies against the MICA antigen in each LABScreen MICA panel. The reaction pattern of the test serum is compared to the lot specific worksheet defining the antigen array. MICA single antigen assay are used to confirm the antibody specificities suggested by a PRA MICA screening assay. Criteria for determining the positivity or negativity of assay is same as the LABScreen Mixed assay.

For MICA genotyping of donors, genomic DNA was extracted from the peripheral bloods using QuickGene-Mini80 DNA isolation system (Fujifilm, Tokyo, Japan). MICA sequence-specific oligonucleotide (SSO) genotyping kit (One Lambda, USA) was used according to the manufacturer's instructions. The target DNA is amplified by polymerase chain reaction (PCR), and denaturation/neutralization process is conducted. After coupling with hybridization and labeling, sample data is acquired by detection in a single reaction mixture. The positive and negative reactions are defined by the percent of positive and negative value for the probe, higher or lower than the preset cut off value for the probe, respectively. MICA allele of the

serum is determined by matching the pattern of positive and negative bead, using the software provided by the manufacturer, or information in the MICA SSO worksheet.

Statistical Analysis

The difference of frequency of MICA antibody and dsMICA antibody between patients with acute rejection and patients without acute rejection was analyzed using Chi-square test and Fisher's exact test as appropriate. Odd ratio (OR) and 95% confidence intervals (95% CI) for predicting results of renal biopsy after transplantation were calculated. The MFI value of dsMICA between two groups was compared using Mann-Whitney test. Statistical significance was set at $P < 0.05$. All statistical analyses were performed using the SPSS software for Windows version 16.0 (SPSS Inc., Chicago, IL, USA).

RESULTS

Among 125 sera, 19 patients were positive for MICA antibody (15.2%). dsMICA was positive on 5 out of 12 sera analyzed (41.7%). Neither MICA antibodies nor dsMICA was associated with acute rejection (AR). Clinical characteristics of the patients with MICA antibody positive group and MICA antibody negative group are given in Table 1. There was no statistical difference between patients with MICA and patients without MICA in age, periods after acute rejection, donor age and sex, number of HLA mismatch, deceased donor, and diagnosis of original disease causing end stage renal disease.

Among patients with MICA positivity, 6 patients (31.6%) showed acute rejection in renal biopsy after transplantation, and 20 patients among MICA negativity (18.9%) showed no evidence of acute rejection (Table 2). The frequencies of MICA antibodies were not different between patients with AR and patients without AR. There was no difference of MICA positivity, when AR was analyzed by subdivision to AMR and ATR. However, IFTA showed statistically significantly association with MICA positivity (OR = 3.84; 95% CI = 1.34 − 9.98, P = 0.009). IFTA I did not show difference, but IFTA stage over II showed significant difference on frequency of MICA positivity (OR = 0.037; 95% CI = 1.02−11.5, P = 0.037). In univariable logistic regression analysis showed associations of MICA antibody, acute rejection and transplantation from deceased donor with IFTA.

In multivariable analysis, MICA positivity was an independent risk factor for IFTA (OR = 2.84, 95% CI = 1.12 - 7.19, P = 0.028) (Table 3).

dsMICA was analyzed for 12 patients whose residual donor DNA was available for MICA genotyping. The characteristics of 12 patients dsMICA tests were performed on were shown in Table 4. Four of them showed IFTA I and three of them showed IFTA grade II or III (Banff '05 classification). Two patients with dsMICA had no evidence of rejection or IFTA on renal biopsy, but showed low levels of dsMICA MFI (MFI value of 1128 and 1835).

The frequencies of dsMICA antibodies were significantly higher in patients with IFTA II or III than that of patients without IFTA II or III (P = 0.045, Table 5A). Three patients were positive for dsMICA in patients with IFTA II or III group (among 5 patients tested), and no patient in patients without IFTA II or III (among 7 patients tested). In the analysis of dsMICA with strong intensity (MFI > 10,000), the frequency of dsMICA positive patients in the IFTA II or III group was significantly higher than that of patients without IFTA II or III group (P = 0.005, OR = 133.0, 95% CI 2.19 − 8082.5) (Table 5B). Among total 12 patients with dsMICA analysis, the MFI value of dsMICA was significantly higher in patients with IFTA II or III (n = 3, median ± SE, 21919.0 ± 2581.0) compared to patients without IFTA II or III (n = 9, median ± SE, 500.0 ± 155.8) (P = 0.009) (Figure 2).

Regarding the risk factors which predict increased serum creatinine

(sCr) at the time of recent follow-up, significant univariable associations with abnormal sCr were observed with acute rejection (P = 0.017), IFTA (P = 0.017), and donor age (P = 0.004). Donor age was independently associated with increased sCr on multivariable linear regression analysis (OR = 1.04; 95% CI = 1.01-1.08, P = 0.018) (Table 6). Regarding the risk factors which predict graft failure, in univariable analysis, acute rejection (P = 0.030) and re-transplantation (P = 0.012) were associated with graft failure. In multivariable analysis, there was no significant risk factor for predicting graft failure (Table 7).

DISCUSSION

This is the first study to demonstrate the association of MICA antibody with IFTA II or III in renal transplantation patients without detectable dsHLA antibodies. Also, the association of MICA or dsMICA with IFTA severity was suspected. However, there was no statistically significant association of MICA or dsMICA with acute allograft rejection.

In previous studies, the results of the association of MICA in renal allograft rejection and survival were not consistent. Zou et al. conducted relatively large patients (n = 1910), multicenter, and prospective cohort study of association of renal allograft outcomes with preformed MICA antibody. This study revealed that MICA antibodies were positive for 11.4% patients before kidney, and MICA positive recipients showed decreased one-year survival than MICA negative recipients [11]. Another studies also showed higher antibody mediated rejection and low graft survival in presensitized MICA positive recipients before kidney transplantation [2, 10]. However, Lemy et al. studied the effects of MICA antibodies on graft survival over a relatively long period of time. And they concluded that sensitizing events for MICA antibody are the same as for human leukocyte antigen antibodies, and MICA antibody did not show any significant difference in graft outcome[15].

There have also been a number of studies on allograft outcomes according to de novo MICA antibodies after transplantation. These studies have shown differences in effect of MICA antibodies on

kidney transplantation, depending on whether or not the research has been carried out including the dsMICA. Cox et al. analyzed 442 renal transplantation recipients and revealed that only coexistence of dsMICA and HLA antibody were significantly associated with ACR (not with AMR) [14]. They also revealed that presence of dsMICA alone was associated with reduced eGFR after 2 years, which implicate possible pathogenic role of dsMICA on premature graft loss resulting from chronic damage and somewhat similar with our study. Meanwhile, Solgi et al. revealed no association of MICA or dsMICA with AR in the absence of HLA antibody [19].

The reasons for these inconsistent conclusions are, firstly, the research design difference such as small number of patients, not considering confounding factors for graft survival analysis, and lack of standardized tests. In addition, few studies analyzed dsMICA [8, 14, 19, 20] and designing the role of MICA antibody without HLA antibody is difficult. Second, the expression level of MICA depending on the transplanted organ or extent of stress according to the patient underlying disease, may cause a difference in clinical outcome. Previous researches have examined the expression of MICA using immunocytochemistry and flow cytometry on biopsy of various transplantation organs [20–23]. MICA is constitutively limited to express on epithelial cells of gastrointestinal tract in normal tissue and induced expression on other epithelial cells or endothelial cells by stress [21]. Compared to renal epithelial cells, MICA is poorly expressed in the heart endothelial cells, which may be less effect on clinical outcomes than renal kidney transplantation [22]. Moreover, the fact that the patient's underlying disease and stress levels have not been corrected for in several studies may affect the outcome of the

transplant. MICA is a stress-induced molecule that is associated with immune surveillance. Ischemia reperfusion injury and cytokines such as IL-2, IL-4, and IL-15 can upregulate the expression of MICA on the endothelium or epithelium of the graft [4]. Post-transplantation events such as AR, infection, and allo-recognition of compartment-specific antigens can induce overexpression of MICA antigens. MICA antigen expression may be increased by a variety of factors that have little relation on transplantation, and MICA antibody may also show positive.

In this study, patient with high MFI value (23,881) of pre-transplantation dsMICA (patient #7, re-transplantation) also had vascular thrombosis at the time of nephrectomy. Complement-independent mechanism of graft damage of MICA antibody by inducing prothrombotic phenotype and resulting in a vascular thrombosis was suggested [2]. Patient #7 received 1st renal transplantation 19 years ago, but IgA nephropathy recurred. Patient #11 and #12 with increased MFI of dsMICA showed AMR and IFTA II. However, interestingly, patient #8 ~ #10 showed ATR and/or AMR without detectable dsMICA after 2~3 months post-transplantation.

Taken together, dsMICA might not be the direct cause of renal graft damage, but can be secondary findings resulting from allogeneic response to overexpressed donor MICA antigens due to various injury, such as infection, ischemia reperfusion and allogeneic response to HLA antigen. IFTA is a common pathological finding of chronic kidney disease caused by chronic renal allograft dysfunction due to AMR [24]. According to a recent study, the IFTA phenotype is strongly enriched for the dysregulated gene pathway, which is highly

shared with the AR biopsy profile. All of the IFTA phenotype could impact on immune mediated injury and worse long term outcomes [25]. These findings suggest that MICA positivity may play an important role as a predictor of worse outcome in long-term renal transplant outcomes. The predictability of IFTA is significant in that it can be established early in the diagnosis and treatment strategy for the survival of the final renal allograft. Recently, attempts have been made to treat or prevent IFTA through early detection of IFTA through blood or urine monitoring of IFTA, anti-EMT agent, antioxidant therapy, tubular epithelial repair, and mesenchymal stem cell therapy. These attempts are expected to improve long term survival and prevent to return to hemodialysis [24, 26].

This study has some limitations. First, this is a retrospective study. Second, the number of patients was small. In particular, the remaining donor DNA required for the identification of dsMICA was insufficient, making it difficult to obtain samples. In the future, the causal relationship of dsMICA and chronic allograft injury or IFTA should be further analyzed with larger number of patients with serial analysis of dsMICA, dsHLA and protocol biopsies.

In conclusion, the association of dsMICA with IFTA was suggested. This study has significance in that it is the first study of dsMICA antibody in Koreans. In addition, it is meaningful that MICA has significance as a secondary finding related to chronic allograft survival. The dsMICA antibody is expected to be used to predict the prognosis of future chronic graft as a serial pattern rather than a cross-sectional view.

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TABLES AND FIGURES

Table 1. Clinical characteristics of the study population

Clinical characteristics	MICA Ab pos (n = 19)	MICA Ab neg (n = 106)	*P*
Age*	46 ± 3.6	51 ± 1.6	
Male:Female	15:4	65:41	
Days after kidney transplantation*	379.0 ± 107.5	357.0 ± 31.7	
Days after acute rejection*	14.0 ± 33.0	78.0 ± 15.6	
Serum Cr (Recent F/U)*	1.29 ± 0.67	1.12 ± 0.07	0.035
Serum Cr (at 1 yr)*	1.30 ± 0.14	1.17 ± 0.06	
Serum Cr (at 3 yr)*	1.16 ± 0.15	1.12 ± 0.04	
Serum Cr (at 5 yr)*	2.57 ± 1.03	1.34 ± 0.10	
Graft failure (%)	3 (15.8%)	1 (0.9%)	0.001
Donor age	48 ± 2.6	46 ± 1.5	
Donor Sex (M:F)	12:9	56:50	
Deceased donor	9 (47.4%)	33 (31.1%)	
No. of HLA mismatch			
0−3	8 (42.1%)	60 (56.6%)	
4−6	10 (52.6%)	36 (34.0%)	
Causes of ESRD			
Glomerular diseases	12 (63.2%)	59 (55.7%)	
Tubulointerstitial diseases	2 (10.5%)	6 (5.7%)	
Vascular diseases	0 (0.0%)	8 (7.5%)	
Cystic and congenital diseases	0 (0.0%)	14 (13.2%)	
Unknown	5 (26.3%)	19 (17.9%)	

*Values are presented as the median ± SE (standard error).

Abbreviation: Cr, creatinine; MICA, MHC class I-related chain A; ESRD, end stage renal disease

Table 2. Prevalence of MICA antibodies according to renal biopsy after transplantation

	MICA (positive)*	MICA (negative)*	*P*	OR	95% CI
AR (n = 26)	6 (31.6%)	20 (18.9%)	0.209		
AMR (n = 13)	2 (10.5%)	11 (10.4%)	0.984		
ATR (n = 19)	5 (26.3%)	14 (13.2%)	0.148		
Borderline (n = 33)	4 (21.1%)	29 (27.4%)	0.566		
IFTA (n = 40)	11 (57.9%)	29 (27.4%)	0.009	3.84	1.34–9.98
IFTA I (n = 25)	6 (31.6%)	19 (17.9%)	0.171		
IFTA II or III (n = 15)	5 (26.3%)	10 (9.4%)	0.037	3.42	1.02–11.5
Normal (n = 54)	6 (31.6%)	48 (45.3%)	0.267		

*Frequency and percentage of patients with MICA positivity and negativity

Abbreviation: MICA, MHC class I–related chain A; AR, acute rejection; AMR, Antibody–Mediated Rejection; ATR, Acute T–cell mediated Rejection; IFTA, Interstitial Fibrosis and Tubular Atrophy

Table 3. Logistic regression results for factors significantly associated with IFTA

	n	Univariable analysis			Multivariable analysis		
		P	OR	95% CI	*P*	OR	95% CI
MICA positivity	16	0.013	2.88	1.25−6.62	0.028	2.84	1.12−7.19
Acute rejection	13	0.030	2.67	1.10−6.47	0.073		
HLA mismatch ≥4	17	0.123					
Deceased donor	24	< 0.001	5.58	2.46−12.67	< 0.001	5.97	2.51−14.19

Abbreviation: MICA, MHC class I−related chain A; IFTA, Interstitial Fibrosis and Tubular Atrophy; HR, hazard ratio

Table 4. Twelve patients who were positive MICA screening and negative HLA class I /II antibody, and available residual donor DNA for MICA genotyping

No.	Sex/Age	Donor MICA allele	Pre-tpl dsMICA	Post-tpl dsMICA	MFI of dsMICA	Origin of ESRD	Kidney biopsy
1	M/36	*002, *010	NT	Pos	1128	MPGN	Normal
2	M/49	*008, *011	NT	Pos	1835	IgAN	Normal
3	M/61	*004,*010	NT	Neg	< 500	FSGS	Normal
4	M/58	*010, *027	Neg	Neg	< 500	MN	Borderline change
5	M/34	*002, *010	NT	Neg	< 500	Unknown	IFTA I
6	F/37	*008, *012	NT	Neg	< 500	IgAN	IFTA I
7	M/68	*002, *045	Pos*	Pos	23296	IgAN	IFTA II/III
8	M/52	*002, *010	NT	Neg	< 500	DM ESRD	ATR
9	F/31	*009, *010	Neg	Neg	< 500	LN	ATR, IFTA I
10	M/30	*009, *010	Neg	Neg	< 500	FSGS	ATR, IFTA I
11	M/51	*009, *012	Neg	Pos	21919	Unknown	AMR, IFTA II
12	M/31	*010, *012	Neg	Pos	14957	Unknown	ATR, AMR, IFTA II

Abbreviation: dsMICA, donor-specific MHC class I-related chain A; AMR, antibody-mediated rejection; IFTA, interstitial fibrosis and tubular atrophy; ATR, acute T-cell mediated rejection; Neg, negative; NT, not tested; Pos, positive; tpl, transplantation

*The MFI value of pre-transplantation dsMICA was 23881.

Table 5. Prevalence of dsMICA antibodies according to renal biopsy after transplantation

(A) dsMICA

Renal biopsy	dsMICA (positive, n=5)*	dsMICA (negative, n=7)*	*P*	OR	95% CI
AMR	2 (40.0%)	0 (0.0%)	0.127		
ATR	1 (20.0%)	3 (42.9%)	0.408		
IFTA	3 (60.0%)	4 (57.1%)	0.921		
IFTA I	0 (0.0%)	4 (57.1%)	0.064		
IFTA II or III	**3 (60.0%)**	**0 (0.0%)**	**0.045**	**21.0**	**1.0−565.0**
Normal	2 (40.0%)	1 (14.3%)	0.310		

(B) dsMICA with strong intensity (MFI > 10,000)

Renal biopsy	dsMICA (positive, n=3)*	dsMICA (negative, n=9)*	*P*	OR	95% CI
AMR	**2 (66.7%)**	**0 (0.0%)**	**0.045**	**31.7**	**1.0−1039.0**
ATR	1 (33.3%)	3 (33.3%)	> 0.999		
IFTA	3 (100.0%)	4 (44.4%)	0.091		
IFTA I	0 (0.0%)	4 (44.4%)	0.157		
IFTA II or III	**3 (100.0%)**	**0 (0.0%)**	**0.005**	**133.0**	**2.19−8082.5**
Normal	0 (0.0%)	3 (33.3%)	0.248		

Table 6. Logistic regression results for factors significantly associated with increased serum creatinine

	Univariable analysis			Multivariable analysis		
	P	OR	95% CI	*P*	OR	95% CI
MICA positivity	0.115	2.04	0.84–4.95			
dsMICA positivity	0.327	0.25	0.02–4.00			
Acute rejection	0.017	3.09	1.22–7.79	0.171	2.03	0.74–5.60
IFTA	0.017	2.80	1.20–6.54	0.085	2.23	0.90–5.54
Deceased donor	0.684	1.20	0.51–2.82			
Donor age	0.004	1.05	1.02–1.09	0.018	1.04	1.01–1.08
Female donor	0.775	0.89	0.39–2.03			
HLA mismatch	0.581	0.93	0.72–1.21			
Re-transplantation	0.999	0	NA			

Abbreviation: MICA, MHC class I-related chain A; dsMICA, donor-specific MHC class I-related chain A; IFTA, Interstitial Fibrosis and Tubular Atrophy; HR, hazard ratio

Table 7. Logistic regression results for factors significantly associated with graft failure.

	Univariable analysis			Multivariable analysis		
	P	OR	95% CI	*P*	OR	95% CI
MICA positivity	0.055	9.52	0.95-95.05			
dsMICA positivity	0.999	1.08	N/A			
Acute rejection	0.030	12.78	1.28-128.56	0.997	1.4	N/A
IFTA	0.997	1.80	N/A			
Deceased donor	0.116	6.31	0.64-62.6			
Donor age	0.823	1.01	0.94-1.08			
Female donor	0.858	0.83	0.11-6.11			
No, of HLA mismatch	0.388	1.39	0.66-2.90			
Re-transplantation	0.012	54.0	2.42-1204.35	0.996	1.6	N/A

Abbreviation: MICA, MHC class I-related chain A; dsMICA, donor-specific MHC class I-related chain A; IFTA, Interstitial Fibrosis and Tubular Atrophy; HR, hazard ratio

Figure 1. Equation for calculation of LABScreen panel reactive antibody or LABScreen single antigen. Normalized background ratio is determine by correcting with negative control beads and background values obtaining by negative control serum

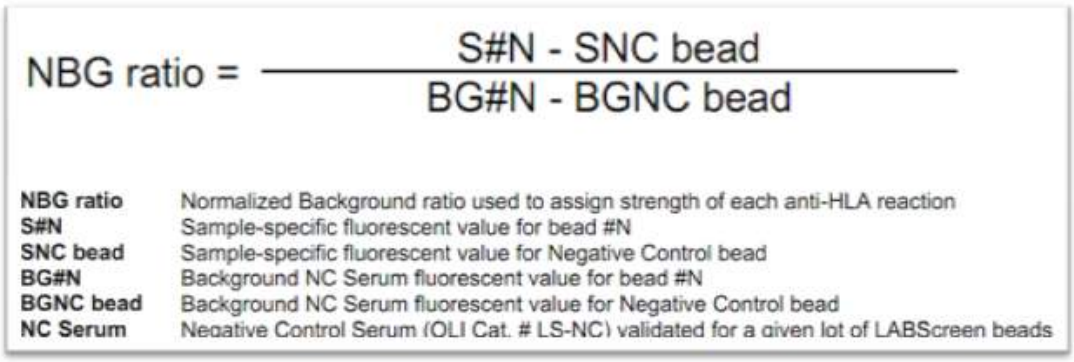
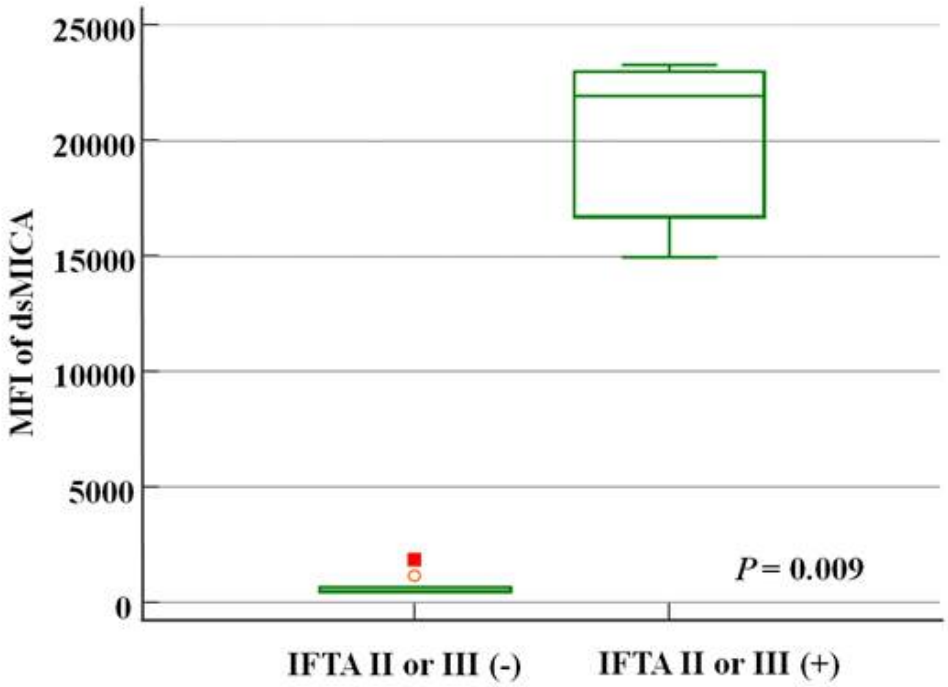


Figure 2. The MFI value of donor MICA-specific antibody (dsMICA) according to presence of interstitial fibrosis and tubular atrophy (IFTA) grade II or III. The MFI value of dsMICA was significantly higher in patients with IFTA II or III (n = 3, median MFI 21,919) compared to patients without IFTA II or III (n = 9, median MFI 500) (P = 0.009).



요약(국문초록)

한국인에서 공여자 특이 항-MICA 항체와 이식신 거부반응과의 연관성

이누리

의학과

서울대학교 대학원

배경. 이식신거부반응의 주된 원인은 공여자특이항체(Donor specific HLA antibody, DSA)이다. 그러나, HLA-DSA가 존재하지 않는 경우에도 이식신거부반응이 발생할 수 있으며, 가능한 원인 항원으로 MHC class I-related chain A (MICA)가 제시되고 있다. 하지만 이식 성적에 있어, 공여자 특이 MICA (dsMICA) 항원과의 관련성은 아직 명확히 알려진 바가 없다.

방법. 2009년 11월부터 2016 년 6월까지 HLA 항체가 없으면서 이식 후 조직검사에서 비정상소견을 보인 신장 이식 환자 71명과 (급성거부반응 13명, 급성거부반응 + Interstitial Fibrosis and Tubular Atrophy (IFTA) 13명, IFTA only 12명, IFTA + borderline change 15명, borderline change only 18명) 정상 조직소견 54명에 대하여 MICA 항체 선별검사 LABScreen MICA (One Lambda, USA)를 시행하였다. MICA 항체

선별검사 양성인 환자 중 공여자특이 MICA 항체분석을 위한 공여자 잔여 DNA가 있는 12명을 대상으로 MICA 유전자형을 분석하였고, 환자 혈청으로 MICA 항체 동정검사를 시행하여 공여자특이 MICA 항체를 분석하였다.

결과. 125명 중 19명이 MICA 항체 (15.2 %) 양성이었다. 공여자특이 MICA 항체는 12명 중 5명 (41.7 %)이 양성이었다. MICA 항체 또는 공여자특이 MICA 항체 모두 급성 거부 반응과 관련이 없었으나, IFTA는 MICA 항체와 유의한 연관이 있었다 (OR = 3.84, 95 % CI = 1.34 – 9.98, P = 0.009). 또한 공여자특이 MICA 항체의 MFI (median fluorescence intensity) 값은 조직생검 소견이 IFTA II 또는 III인 환자에서 (n = 3, median ± SE, 21919.0 ± 2581.0)에서 IFTA II 또는 III가 아닌 환자(n = 9, median ± SE, 500.0 ± 155.8)에 비해 유의하게 높았다 (P = 0.009).

결론. MICA 항체는 HLA 항체가 없는 환자에서 IFTA 조직 소견과 유의한 연관을 보였으며, 공여자특이 MICA 항체의 강도는 IFTA의 중증도와 연관이 있었다. 그러나, MICA 또는 공여자특이 MICA 항체는 HLA 항체가 없는 환자에서 급성 거부 반응과 통계적으로 유의한 연관성이 없었다. MICA 항체는 만성적 신손상의 예측 및 추후 치료방향을 결정하는 유용한 지표로써 의미가 있다.

주요어: anti-MHC class I-related chain A (MICA) 항체, 이식 거부 반응, 신장 이식

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