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Mannose binding lectin 2 gene polymorphisms in patients after renal transplantation with acute graft rejection

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ABSTRACT

Mannose binding lectin 2 (*MBL2*) is one of the pattern-recognition soluble receptors and is responsible for complement activation via the lectin pathway, so it plays an important role in kidney transplantation.

The aim of the study was to examine the association between *MBL2* gene polymorphisms and acute rejection of the kidney allograft. This study enrolled 266 Caucasian deceased-donor renal transplant recipients – 69 with diagnosed acute graft rejection, 197 with stable graft function.

A 969 bp DNA fragment, from chromosome 10, including promoter region and exon 1 of *MBL2* gene was sequenced. The DNA fragment obtained contained 122 SNPs accordingly to the NCBI dbSNP database. Of this number only nine showed variation within our population (rs36014597, rs5030737, rs1800450, rs7095891, rs11003123, rs7096206, rs7084554, rs11003124, rs11003125), and only these were subjected to further analysis.

Among the studied polymorphisms in the *MBL2* gene only rs1800450 polymorphism was statistically significantly associated with kidney allograft rejection. The AA genotype was significantly associated with an increased risk of acute kidney allograft rejection. (AA vs GA + GG: OR = 9.29 (95%CI: 1.83–47.17), p = 0.005).

The results of our study indicate that *MBL2* gene rs1800450 polymorphism may be associated with the risk of acute kidney allograft rejection. The AA genotype, associated with lower MBL2 expression, may be associated with an increased risk of acute kidney allograft rejection.

1. Introduction

Kidney transplantation is a common therapy for patients with renal insufficiency. Unfortunately, the transplantation of kidney allograft induces the activation of a patient's immune system, leading to acute graft rejection (AR). Both innate immunity as well as humoral and cellular immune responses are involved in these processes. The complement system plays an important role in the immune response in patients with kidney allografts [1]. The complement system is an important part of the innate immune system, involved in the defence against various pathogens [2]. Previous studies have indicated that kidney transplantation activates the innate immune system especially during ischemia–reperfusion injury [3]. The cells of innate immunity express pattern recognition receptors that, after activation, are involved in the maturation of antigen-presenting cells producing various mediators that play an important role in immune response. Mannose binding lectin 2 (MBL2) belongs to the pattern-recognition soluble receptors and is responsible for the complement system activation via the lectin pathway [4]. MBL2 thus has an important role in the defence against various microorganism activating macrophages, and other cells involved in the phagocytosis of pathogens [5]. Previous studies have indicated that MBL2 also plays important role in ischemia–reperfusion after kidney transplantation, in kidney allograft rejection, and in long term kidney allograft function [4,6,7].

The *MBL2* gene is located on chromosome 10, and is responsible for the synthesis of MBL2 protein [8,9]. Several polymorphisms have been detected in the *MBL2* gene. Some may affect *MBL2* gene expression and alter the synthesis of MBL2 protein. These polymorphisms were in-

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Table 1
Clinical characteristics of the studied group of renal graft recipients.

Parameter	Value
Age [years]	47.17 ± 12.25
Sex	164M/102F
HD before Tx	94%
DGF	31.6%
Biopsy confirmed AR	25.9%
Five-year patients' survival rate	97.8%
Five-year allografts' survival rate	82.3%
Return to dialysis	8%
Steroids	91%
Cyclosporine A	75%
Cyclosporine A [ng/ml]	150–200
Tacrolimus	24%
Tacrolimus [ng/ml]	7–12

HD - hemodialysis, Tx – transplantation, DGF – delayed graft function, AR – acute rejection.

vestigated in various diseases, where innate immunity plays an important role, especially in bacterial infections, tuberculosis, rheumatic fever, sepsis, lupus erythematosus [10–14].

The aim of this study was to examine the association between kidney allograft recipients *MBL2* gene polymorphisms and acute rejection of kidney allograft.

2. Materials and methods

This study enrolled 266 Caucasian deceased-donor renal transplant recipients – 69 with diagnosed acute graft rejection, 197 with stable graft function. The transplantation procedures were performed in 1999–2004. All kidneys were received from deceased donors. The duration of follow-up was five years. The first renal allograft recipients were consecutively included, after giving their consent to participate in the study. Patients were excluded if they had received more than one renal transplant, if their graft had been functioning for less than six months, or if they failed to provide consent. Clinical characteristics of renal graft recipients are presented in Table 1.

The histories of the patients were analyzed, taking into account delayed graft function (DGF), acute rejection and the risk of graft loss/dialysis after transplantation. DGF was defined as the need for hemodialysis within the first 7 days after transplantation. Acute graft rejection (AR) was diagnosed clinically and confirmed by biopsy. AR diagnoses were classified as T cell-mediated rejection. Some patients had signs of antibody-mediated rejection, however in all patients dominated the signs of T cell mediated rejection.

All biopsies were reviewed by a renal pathologist using the Banff working classification criteria [15]. All patients received a standard immunosuppressive protocol with triple drug therapy including a calcineurin inhibitor (cyclosporine A in 75% of patients and tacrolimus in 24%), azathioprine (55%) or mycophenolate mofetil (37%), and steroids (91%). Informed consent was obtained from all patients. The local ethics committee of the Pomeranian Medical University in Szczecin, Poland approved the study protocol.

2.1. Methods

DNA was isolated from peripheral blood using the Genomic Mini AX Blood 1000 Spin kit (A&A Biotechnology, Gdańsk, Poland). A 969 bp DNA fragment including promoter region and exon 1 of *MBL2* gene was amplified with use of the following primers: Forward 5′-GGGGAATTC CTGCCAGAAAGT-3′, Reverse 5′-CATATCCCCAGGCAGTTTCCTC-3′, accordingly to Wu et al. [16]. The cycling conditions were 94 °C for 4 min, 35 cycles of 94 °C for 30 s, 56 °C for 30 s, 72 °C for 30 s and finally 72 °C for 5 min. The PCR products were separated by agarose gel electrophoresis and then subjected to DNA sequencing. The DNA was sequenced, using forward and reverse primers, BigDye Terminator v3.1 cycle sequencing and the obtained sequences were analysed using an ABI 3130 DNA Analyser (Applied Biosystems).

2.2. Statistical analysis

The consistency of the genotype distribution was assessed using the Hardy-Weinberg equilibrium exact test. A χ^2-square test and Fisher's exact test were used to compare genotype and allele distributions between the groups. Multivariate logistic regression analysis was performed to find independent predictors of acute rejection. Cox proportional hazards regression model was used to analyse the risk of graft loss/return to dialysis after transplantation. P-values < 0.05 were considered statistically significant.

3. Results

A 969 bp DNA fragment from chromosome 10, including the promoter region and exon 1 of *MBL2* gene was sequenced. The DNA fragment obtained contained 122 SNPs according to the NCBI dbSNP database. Of this number only nine showed variation within our population (rs36014597, rs5030737, rs1800450, rs7095891, rs11003123, rs7096206, rs7084554, rs11003124, rs11003125), and only these were subjected to further analysis. The location of the polymorphisms studied is presented in Table 2.

The distributions of studied polymorphisms were in HWE. Among the polymorphisms studied in the *MBL2* gene, only the rs1800450

Table 2
List of non-coding and coding SNPs as having functional effect.

SNP ID	Chromosome position	Alleles	Functional consequence	Localization	
rs1800450	Chromosome 10: 52771475	G > A	Missense	Exon 1	+225
rs5030737	Chromosome 10: 52771482	C > T	Missense	Exon 1	+218
rs7095891	Chromosome 10: 52771701	G > A	-[a]	5′ near gene	−1
rs11003123	Chromosome 10: 52771774	G > A	-[a]	5′ near gene	−74
rs7096206	Chromosome 10: 52771925	G > C	-[a]	5′ near gene	−225
rs36014597	Chromosome 10: 52772040	A > G	-[a]	5′ near gene	−340
rs7084554	Chromosome 10: 52772053	T > C	-[a]	5′ near gene	−353
rs11003124	Chromosome 10: 52772131	T > G	-[a]	5′ near gene	−431
rs11003125	Chromosome 10: 52772254	G > C	-[a]	5′ near gene	−554

[a] Intron variant.

Table 3
The association between *MBL2* rs36014597, rs5030737, rs1800450 genotypes and acute rejection.

	Acute rejection		Without acute rejection		p-value[a]		p-value[b]	OR (95% CI)
	n	%	n	%				
MBL2 rs36014597								
Genotype								
AA	47	68.11	129	65.48	0.86	GG + AG vs AA	0.77	0.89(0.49–1.59)
AG	18	26.09	58	29.44		GG vs AG + AA	0.76	1.15(0.35–3.80)
GG	4	5.80	10	5.08		GG vs AA	1.00	1.10(0.33–3.67)
						AG vs AA	0.64	0.85(0.46–1.59)
						GG vs AG	0.74	1.29(0.36–4.61)
MBL2 rs36014597								
Allele								
A	112	81.16	316	80.20				
G	26	18.84	78	19.80		G vs A	0.90	0.94(0.57–1.54)
MBL2 rs5030737								
Genotype								
CC	60	86.96	174	88.32	0.24	TT + CT vs CC	0.83	1.13(0.50–2.59)
CT	8	11.59	23	11.68		TT vs CT + CC	1.00	–
TT	1	1.45	0	0		TT vs CC	1.00	–
						CT vs CC	1.00	1.01(0.43–2.37)
						TT vs CT	1.00	–
MBL2 rs5030737								
Allele								
C	128	92.75	371	94.16				
T	10	7.25	23	5.84		T vs C	0.54	1.26(0.58–2.72)
MBL2 rs1800450								
Genotype								
GG	47	68.11	136	69.04	0.004*	AA + GA vs GG	0.88	1.04(0.58–1.88)
GA	16	23.19	59	29.95		AA vs GA + GG	0.005*	9.29(1.83–47.17)
AA	6	8.70	2	1.01		AA vs GG	0.006*	8.68(1.69–44.50)
						GA vs GG	0.53	0.78(0.41–1.49)
						AA vs GA	0.004*	11.06(2.04–60.13)
MBL2 rs1800450								
Allele								
G	110	79.71	331	84.01				
A	25	20.29	63	15.99		A vs G	0.29	1.34(0.82–2.19)

MBL2 rs36014597 HWE: p = 0.17.
MBL2 rs5030737 HWE: p = 1.
MBL2 rs1800450 HWE: p = 0.83.
[a] χ^2 test.
[b] Fisher exact test.
* $p < 0.05$.

polymorphism was statistically significantly associated with kidney allograft rejection (Tables 3–5). The AA genotype was significantly associated with an increased risk of acute kidney allograft rejection, AA vs GA + GG, OR 9.29, 95%CI (1.83–47.17), p = 0.005 (Table 3). The association remains significant after Bonferroni correction for multiple testing (χ^2 test p = 0.004, while Bonferroni-corrected p-value threshold is 0.05/9 = 0.0056, since 9 polymorphisms were studied).

This association was confirmed in multivariate regression analysis taking into the account recipient's sex, age and *MBL2* rs1800450 AA genotype. In this analysis recipient's *MBL2* rs1800450 AA genotype was statistically significantly associated with increased risk of acute rejection (Table 10).

Tables 6–8 present the associations between studied polymorphisms and DGF. These associations were statistically non significant.

In Table 9 is shown univariate Cox regression analysis for the risk of graft loss/dialysis after transplantation. There were no statistically significant associations between studied polymorphisms and the risk of graft loss/dialysis after transplantation.

4. Discussion

In this study we examined the associations between polymorphisms

Table 4
The association between *MBL2* rs7095891, rs11003123, rs7096206 genotypes and acute rejection.

	Acute rejection		Without acute rejection		p-value[a]		p-value[b]	OR (95% CI)
	n	%	n	%				
MBL2 rs7095891								
Genotype								
GG	46	66.67	126	63.96	0.90	AA + GA vs GG	0.77	0.89(0.50–1.58)
GA	19	27.53	60	30.46		AA vs GA + GG	1.00	1.04(0.32–3.38)
AA	4	5.80	11	5.58		AA vs GG	1.00	1.00(0.30–3.28)
						GA vs GG	0.76	0.87(0.47–1.61)
						AA vs GA	1.00	1.15(0.33–4.03)
MBL2 rs7095891								
Allele								
G	111	80.43	312	79.19				
A	27	19.57	82	20.81		A vs G	0.81	0.93(0.57–1.50)
MBL2 rs11003123								
Genotype								
GG	46	66.67	126	63.96	0.86	AA + GA vs GG	0.77	0.89(0.50–1.58)
GA	19	27.53	61	30.96		AA vs GA + GG	0.76	1.15(0.35–3.80)
AA	4	5.80	10	5.08		AA vs GG	1.00	1.10(0.33–3.67)
						GA vs GG	0.65	0.85(0.46–1.58)
						AA vs GA	0.74	1.28(0.36–4.57)
MBL2 rs11003123								
Allele								
G	111	80.43	313	79.44				
A	27	19.57	81	20.56		A vs G	0.90	0.94(0.58–1.53)
MBL2 rs7096206								
Genotype								
CC	45	65.22	111	56.34	0.19	GG + CG vs CC	0.21	0.69(0.39–1.22)
CG	19	27.53	77	39.09		GG vs CG + CC	0.37	1.63(0.53–5.05)
GG	5	7.25	9	4.57		GG vs CC	0.56	1.37(0.44–4.31)
						CG vs CC	0.14	0.61(0.33–1.12)
						GG vs CG	0.18	2.25(0.68–7.50)
MBL2 rs7096206								
Allele								
C	109	78.99	299	75.89				
G	29	21.01	95	24.11		G vs C	0.49	0.84(0.52–1.34)

MBL2 rs7095891 HWE: p = 0.19.
MBL2 rs11003123 HWE: p = 0.26.
MBL2 rs7096206 HWE: p = 1.
[a] χ^2 test.
[b] Fisher exact test.

in the *MBL2* gene in kidney allograft recipients and kidney allograft acute rejection. We found an association between rs1800450 AA genotype and increased risk of acute rejection. *MBL2* gene rs1800450 was studied in various diseases, particularly including bacterial and viral infections, tuberculosis, rheumatoid arthritis, type 2 diabetes and diabetic nephropathy, and systemic lupus erythematosus. Previous studies have shown that this polymorphism changes the *MBL2* gene expression and MBL2 protein synthesis [14,17,18,19]. Subjects with the AA genotype have lower transcriptional activity and lower MBL2 protein synthesis.

Several studies investigated the role of MBL in kidney transplantation. The studies examined the role of MBL2 expression in ischemia-reperfusion injury, in kidney graft rejection and the effect of MBL on kidney allograft function [6,7,20], however the results are inconsistent.

Some studies suggest that the increased expression of MBL2 and complement system activation has a negative effect on kidney graft outcome and survival, however other studies indicate a positive and protective effect from MBL2 and the complement system on kidney graft function.

Berger et al. have shown that kidney graft survival was significantly longer in recipients with low MBL2 levels [6,20]. Conversely, Bay et al.

Table 5
The association between *MBL2* rs7084554, rs11003124, rs11003125 genotypes and acute rejection.

	Acute rejection		Without acute rejection		p-value[a]		p-value[b]	OR (95% CI)
	n	%	n	%				
MBL2 rs7084554								
Genotype								
TT	47	68.11	126	63.96	0.67	CC + TC vs TT	0.56	0.83(0.46–1.49)
TC	18	26.09	62	31.47		CC vs TC + TT	0.75	1.29(0.38–4.32)
CC	4	5.80	9	4.57		CC vs TT	0.75	1.19(0.35–4.05)
						TC vs TT	0.54	0.78(0.42–1.45)
						CC vs TC	0.50	1.53(0.42–5.56)
MBL2 rs7084554								
Allele								
T	112	81.16	314	79.70				
C	26	18.84	80	20.30		C vs T	0.80	0.91(0.56–1.49)
MBL2 rs11003124								
Genotype								
TT	47	68.11	126	63.96	0.74	GG + TG vs TT	0.56	0.83(0.46–1.49)
TG	18	26.09	61	30.96		GG vs TG + TT	0.76	1.15(0.35–3.80)
GG	4	5.80	10	5.08		GG vs TT	1.00	1.07(0.32–3.59)
						TG vs TT	0.54	0.79(0.42–1.48)
						GG vs TG	0.73	1.36(0.38–4.84)
MBL2 rs11003124								
Allele								
T	112	81.16	313	79.44				
G	26	18.84	81	20.56		G vs T	0.71	0.90(0.55–1.47)
MBL2 rs11003125								
Genotype								
GG	32	46.38	89	45.18	0.78	CC + GC vs GG	0.89	0.95(0.55–1.65)
GC	26	37.68	82	41.62		CC vs GC + GG	0.55	1.25(0.58–2.68)
CC	11	15.94	26	13.20		CC vs GG	0.68	1.18(0.52–2.65)
						GC vs GG	0.76	0.88(0.48–1.60)
						CC vs GC	0.52	1.33(0.58–3.06)
MBL2 rs11003125								
Allele								
G	90	65.22	260	65.99				
C	48	34.78	134	34.01		C vs G	0.92	1.03(0.69–1.56)

MBL2 rs7084554 HWE: p = 0.34.
MBL2 rs11003124 HWE: p = 0.25.
MBL2 rs11003125 HWE: p = 0.13.
[a] χ^2 test.
[b] Fisher exact test.

have indicated that low MBL2 serum levels are associated with reduced kidney graft survival [21]. Similarly, Ibernon et al. have shown that subclinical rejection in renal transplants is associated with low serum MBL2 levels, and low pre-transplant serum MBL2 levels are associated with more severe inflammation and increased apoptosis in early surveillance renal allograft biopsies, suggesting that MBL2 modulates renal inflammation after transplantation [22,23]. These authors have also indicated that low serum mannose-binding lectin is a risk factor for new onset diabetes mellitus after renal transplantation [24].

In the study by Damman et al., *MBL2* gene polymorphisms of the donor and recipient did not affect graft outcome after kidney transplantation [25]. Golshayan et al. investigated *MBL2* gene polymorphism in patients after kidney transplantation [26]. These authors analysed polymorphisms in the *MBL2* gene with MBL2 serum levels and serum levels in kidney transplant recipients. Low MBL2 serum levels and deficient *MBL2* genotypes were associated with a higher incidence

Table 6
The association between *MBL2* rs36014597, rs5030737, rs1800450 genotypes and delayed graft function (DGF).

	DGF		Without DGF		p-value[a]		p-value[b]	OR (95% CI)
	n	%	n	%				
MBL2 rs36014597								
Genotype								
AA	56	66.67	120	65.94	0.69	GG + AG vs AA	1.00	0.97(0.56–1.67)
AG	25	29.76	51	28.02		GG vs AG + AA	0.59	0.58(0.16–2.12)
GG	3	3.57	11	6.04		GG vs AA	0.56	0.58(0.16–2.18)
						AG vs AA	0.88	1.05(0.59–1.87)
						GG vs AG	0.54	0.56(0.14–2.17)
MBL2 rs36014597								
Allele								
A	137	81.55	291	79.95				
G	31	18.45	73	20.05		G vs A	0.72	0.90(0.57–1.44)
MBL2 rs5030737								
Genotype								
CC	70	83.33	164	90.11	0.18	TT + CT vs CC	0.15	1.82(0.86–3.87)
CT	14	16.67	17	9.34		TT vs CT + CC	1.00	–
TT	0	0	1	0.55		TT vs CC	1.00	–
						CT vs CC	0.10	1.93(0.90–4.13)
						TT vs CT	1.00	–
MBL2 rs5030737								
Allele								
C	154	91.67	345	94.78				
T	14	8.33	19	5.22		T vs C	0.18	1.65(0.81–3.38)
MBL2 rs1800450								
Genotype								
GG	61	72.62	122	67.03	0.41	AA + GA vs GG	0.40	0.77(0.43–1.36)
GA	22	26.19	53	29.12		AA vs GA + GG	0.44	0.30(0.04–2.49)
AA	1	1.19	7	3.85		AA vs GG	0.44	0.29(0.03–2.37)
						GA vs GG	0.56	0.83(0.46–1.49)
						AA vs GA	0.43	0.34(0.04–2.96)
MBL2 rs1800450								
Allele								
G	144	85.71	297	81.59				
A	24	14.29	67	18.41		A vs G	0.27	0.74(0.45–1.23)

[a] χ^2 test.
[b] Fisher exact test.

of acute cellular rejection during the first year. In contrast, there was no significant association with rates of antibody mediated rejection, patient death, early graft dysfunction or loss. The authors concluded that *MBL2* gene polymorphisms result in low MBL2 serum levels and are associated with acute cellular rejection after kidney transplantation.

The mechanisms of the effect of MBL2 on the kidney allograft and process of acute rejection are very complex. MBL2 is important in the defence against various microorganisms, including bacterial, viral and fungal infections, in patients after kidney transplantation [7,27]. On the other hand studies have shown that MBL2 could activate the complement system during ischemia-reperfusion, leading to ischemia-reperfusion injury, and may have a cytotoxic effect on renal tubules [28,29].

Experimental studies have indicated that MBL2 has positive effects

Table 7
The association between *MBL2* rs7095891, rs11003123, rs7096206 genotypes and delayed graft function (DGF).

	DGF		Without DGF		p-value[a]		p-value[b]	OR (95% CI)
	n	%	n	%				
MBL2 rs7095891								
Genotype								
GG	54	64.29	118	64.84	0.69	GG + AG vs AA	1.00	0.97(0.56–1.67)
GA	27	32.14	52	28.57		GG vs AG + AA	0.59	0.58(0.16–2.12)
AA	3	3.57	12	6.59		GG vs AA	0.56	0.58(0.16–2.18)
						AG vs AA	0.88	1.05(0.59–1.87)
						GG vs AG	0.54	0.56(0.14–2.17)
MBL2 rs7095891								
Allele								
G	135	80.36	288	79.12				
A	33	19.64	76	20.88		G vs A	0.72	0.90(0.57–1.44)
MBL2 rs11003123								
Genotype								
GG	54	64.29	118	64.84	0.18	TT + CT vs CC	0.15	1.82(0.86–3.87)
GA	27	32.14	53	29.12		TT vs CT + CC	1.00	–
AA	3	3.57	11	6.04		TT vs CC	1.00	–
						CT vs CC	0.10	1.93(0.90–4.13)
						TT vs CT	1.00	–
MBL2 rs11003123								
Allele								
G	135	80.36	289	79.40				
A	33	19.64	75	20.60		T vs C	0.18	1.65(0.81–3.38)
MBL2 rs7096206								
Genotype								
CC	48	57.14	108	59.34	0.41	AA + GA vs GG	0.40	0.77(0.43–1.36)
CG	33	39.29	63	34.62		AA vs GA + GG	0.44	0.30(0.04–2.49)
GG	3	3.57	11	6.04		AA vs GG	0.44	0.29(0.03–2.37)
						GA vs GG	0.56	0.83(0.46–1.49)
						AA vs GA	0.43	0.34(0.04–2.96)
MBL2 rs7096206								
Allele								
C	129	76.79	279	76.65				
G	39	23.21	85	23.35		A vs G	0.27	0.74(0.45–1.23)

[a] χ^2 test.
[b] Fisher exact test.

via complement system activation on the clearance of necrotic and apoptotic cells, increasing the phagocytosis by macrophages and dendritic cells [30,31]. Nauta et al. have indicated that MBL2 is involved in non-inflammatory sequestration of apoptotic and necrotic cells interacting with structures exposed on apoptotic and necrotic cells and stimulating the production of IL-6, IL-10, and TNF-α [30,31].

These studies show the multidirectional action of MBL2 in transplanted organs. Some studies suggest the protective role of MBL2 on kidney allografts, however others indicate the negative effect of MBL2 and complement system activation on kidney grafts.

The results of our study suggest that *MBL2* gene rs1800450 polymorphism in kidney allograft recipients may be associated with the risk of acute kidney allograft rejection. The AA genotype, associated with lower MBL2 expression, may be associated with an increased risk of acute kidney allograft rejection.

Table 8
The association between *MBL2* rs7084554, rs11003124, rs11003125 genotypes and delayed graft function (DGF).

	DGF		Without DGF		p-value[a]		p-value[b]	OR (95% CI)
	n	%	n	%				
MBL2 rs7084554								
Genotype								
TT	55	65.48	118	64.84	0.79	CC + TC vs TT	1.00	0.97(0.56–1.67)
TC	26	30.95	54	29.67		CC vs TC + TT	0.59	0.58(0.16–2.12)
CC	3	3.57	10	5.49		CC vs TT	0.56	0.58(0.16–2.18)
						TC vs TT	0.88	1.05(0.59–1.87)
						CC vs TC	0.54	0.56(0.14–2.17)
MBL2 rs7084554								
Allele								
T	136	80.95	290	79.67				
C	32	19.05	74	20.33		C vs T	0.72	0.90(0.57–1.44)
MBL2 rs11003124								
Genotype								
TT	55	65.48	118	64.84	0.69	GG + TG vs TT	0.15	1.82(0.86–3.87)
TG	26	30.95	53	29.12		GG vs TG + TT	1.00	–
GG	3	3.57	11	6.04		GG vs TT	1.00	–
						TG vs TT	0.10	1.93(0.90–4.13)
						GG vs TG	1.00	–
MBL2 rs11003124								
Allele								
T	136	80.95	289	79.40				
G	32	19.05	75	20.60		G vs T	0.18	1.65(0.81–3.38)
MBL2 rs11003125								
Genotype								
GG	34	40.48	87	47.80	0.41	CC + GC vs GG	0.40	0.77(0.43–1.36)
GC	39	46.43	69	37.91		CC vs GC + GG	0.44	0.30(0.04–2.49)
CC	11	13.09	26	14.29		CC vs GG	0.44	0.29(0.03–2.37)
						GC vs GG	0.56	0.83(0.46–1.49)
						CC vs GC	0.43	0.34(0.04–2.96)
MBL2 rs11003125								
Allele								
G	107	63.69	243	66.76				
C	61	36.31	121	33.24		C vs G	0.27	0.74(0.45–1.23)

[a] χ^2 test.
[b] Fisher exact test.

Table 9
Univariate Cox regression analysis for the risk of graft loss/dialysis after transplantation (*MBL2* rs36014597, rs1800450, rs7095891, rs11003123, rs7096206, rs7084554, rs11003124, rs11003125)

Risk factor	HR (95% CI)	p
rs36014597 *MBL2* (one G allele)	0.45(0.15–1.34)	0.15
rs1800450 *MBL2* (one A allele)	1.10(0.44–2.73)	0.83
rs1800450 *MBL2* (AA vs GA + GG)	1.69(0.23–12.59)	0.61
rs7095891 *MBL2* (one A allele)	0.43(0.14–1.27)	0.13
rs7095891 *MBL2* (AA vs GA + GG)	0.87(0.12–6.50)	0.89
rs11003123 *MBL2* (one A allele)	0.43(0.14–1.27)	0.13
rs11003123 *MBL2* (AA vs GA + GG)	0.95(0.13–7.09)	0.96
rs7096206 *MBL2* (one G allele)	0.67(0.27–1.67)	0.39
rs7084554 *MBL2* (one C allele)	0.43(0.15–1.29)	0.13
rs7084554 *MBL2* (CC vs TC + TT)	1.05(0.14–7.79)	0.97
rs11003124 *MBL2* (one G allele)	0.43(0.15–1.29)	0.13
rs11003124 *MBL2* (GG vs TG + TT)	0.95(0.13–7.09)	0.96
rs11003125 *MBL2* (one C allele)	1.80(0.73–4.47)	0.20

HR – hazard ratio.

Table 10
Multivariate logistic regression analysis of association between *MBL2 rs1800450* and acute rejection as the dependent variable.

Independent variables	OR (95% CI)	p
Sex (male vs female)	1.63 (0.88–3.01)	0.12
Recipient's age (years)	0.98 (0.95–1.00)	0.035
MBL2 rs1800450 (AA carriers)	8.66 (1.65–45.53)	0.010

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