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Pre-transplant angiotensin II type 1 receptor antibodies: a risk factor for decreased kidney graft function in the early post-transplant period?

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ABSTRACT

Angiotensin II type 1 receptor antibodies (AT_1Rab) are associated to a significantly lower graft survival and a higher risk of acute rejection after kidney transplantation. This study aimed to evaluate graft function and BPAR during the 1st year post-transplant (PT) in adult kidney transplant recipients (KTR), between 03/2009 and 08/2012. Pre-KT sera were screened for AT_1Rab (ELISA) and HLA-DSA (Luminex). Three groups were analyzed: AT_1Rab only (n = 13); HLA-DSA only (n = 8); and no AT_1Rab or HLA-DSA (n = 90). No differences were observed in clinical characteristics across groups. A higher percentage of BPAR was observed in the AT_1Rab positive group, but this difference was not significant. KTR with AT_1Rab had a lower mean eGFR (20 mL/min/1.73m^2) when compared to KTR with no Abs at 12 months. The significant difference in eGFR was observed since the 1st month PT. Multivariate analysis showed 4 factors independently and significantly associated with eGFR at 12mos PT: BPAR (-18.7 95%, CI -28.2 to -9.26, p<0.001), AT_1Rab (-10.51, CI -20.9 to -0.095, p = 0.048), donor age (-0.42, CI -0.75 to -0.103 p = 0.010), and recipient age (-0.36, CI -0.67 to -0.048, p = 0.024). In this study AT_1Rab in pre-transplant sera from KTR, was an independent and significant risk factor contributing to a lower eGFR 12 months. PT. This finding deserves to be confirmed in a larger KTR population.

Anticuerpos contra el receptor 1 de angiotensina II pretrasplante: ¿un factor de riesgo para la disminución de la función del injerto en el periodo postrasplante temprano?

RESUMEN

Los anticuerpos dirigidos contra el receptor 1 de angiotensina II (anti-AT_1R) han sido asociados a una supervivencia significativamente menor del injerto y a un mayor riesgo de rechazo agudo en receptores de trasplante renal. El objetivo de este estudio fue evaluar la función renal y los episodios de rechazo agudo confirmados por biopsia (RACB) durante el primer año de evolución postrasplante (PT) en receptores de trasplante renal (RTR) adultos, efectuados entre 03/2009 y 08/2012. El suero pretrasplante fue analizado para detectar la presencia de anti-AT_1R (ELISA) y para anticuerpos HLA donante específico (ADE-HLA) (Luminex). Se conformaron tres grupos para el análisis: presencia exclusiva de anti-AT_1R (n = 13); presencia exclusiva de ADE-HLA (n = 8); y ausencia de anti-AT_1R y de ADE-HLA (n = 90). No se observaron diferencias en las características clínicas entre los grupos. Un porcentaje mayor, no significativo, de RACB fue observado en el grupo anti-AT_1R positivo. Los RTR con anti-AT_1R tuvieron una menor tasa de filtrado glomerular estimada (TFGe) promedio (20 mL/min/1.73m^2) a 12 meses PT cuando se compararon con los RTR sin anticuerpos. Esta diferencia significativa en la TFGe se observó desde el primer mes PT. El análisis multivariado mostró cuatro factores independiente y significativamente asociados con TFGe a 12 meses PT: RACB (-18.7, 95% IC -28.2 a -9.26, p < 0.001), anti-AT_1R (-10.51, IC -20.9 a -0.095, p = 0.048), edad del donante (-0.42, IC -0.75 a -0.103, p = 0.010) y edad del receptor (-0.36, IC -0.67 a -0.048, p = 0.024). En este estudio la presencia de anti-AT_1R en suero pretrasplante de RTR fue un

factor de riesgo independiente y significativo para TFGe reducida a 12 meses PT. Este hallazgo requiere ser confirmado en una población más numerosa de RTR.

INTRODUCTION

In recent years, the production of antibodies (Abs) against non-HLA related antigens has been described in renal transplant recipients; they can either be alloantibodies or autoantibodies.[1-7] Some of the Abs whose implications on renal graft survival are beginning to be understood, are angiotensin type I receptor (AT$_1$R) antibodies (AT$_1$Rab).

The gene for AT$_1$R is on chromosome 3 and via 4 exons, it encodes a G protein coupled to a receptor with seven transmembrane domains. This receptor is responsible for most angiotensin II-mediated physiological cardiovascular responses, including blood pressure regulation and fluid and electrolyte balance.[8]

Anti-AT$_1$Rab recognize a structure constituted by the second extracellular loop and act as receptor agonists fostering vascular inflammatory processes.[8,9] AT$_1$Rab may mediate graft vasculopathy by endothelial activation.[10] AT$_1$Rab have been shown to have direct effects on endothelial cells as well as smooth muscle cells by inducing Erk 1/2 mediated signal transduction. These Abs can also activate transcription factor activator protein-1 (AP-1) downstream from Erk 1/ 2, increase the binding activity of nuclear factor-κB (NF-κB) transcription factor as well as the expression of NF-κB pro-inflammatory target genes for chemokines such as MCP-1 and RANTES.[7,11]

A relationship has been established between the presence of AT$_1$Rab and hypertension,[12] cardiac hypertrophy[13] and renal fibrosis;[14] these all result from excessive activity of the angiotensin II-AT$_1$R complex and lead to increased cardiovascular morbility and mortality.[15]

There are multiple pathways by which AT$_1$Rab may develop, including both autoreactive and alloreactive responses. Autoantibodies can arise as a result of molecular mimicry and cross-reactivity.[10] Inflammatory events might lead to de novo expression of autoantigens, proteins that become accessible after injury or as a result of the transplant process itself.[3,16] AT$_1$Rab were initially described in women with pre-eclampsia and malignant hypertension.[17-20]

Due to the receptor's protein nature, it is polymorphic and some of its variants have been linked to an increased response to angiotensin II and therefore, to renal and cardiovascular disease.[21] The most studied polymorphism is A1166C that, when present in the donor, leads to an increased risk of reactivity with the recipient's antibodies since this polymorphism is associated to an increased expression of the AT$_1$R.[4,10] Patients may also develop Abs to AT$_1$R polymorphisms through the usual mechanisms of allosensitization (pregnancy, transfusions and transplants)[4] as well as upon immunosuppressant therapy withdrawal or non-adherence to the treatment.[10,22]

Although AT$_1$Rab have been shown to belong to the IgG1 and IgG3 sub-classes that fix complement, some reports have underscored the low frequency of C4d positivity in biopsies from patients with these Abs and graft rejection; this suggests a different pathogenesis to that due to the effect of donor specific HLA Abs (DSA).[7,23] In this regard, It has been suggested that genes regulated by AT$_1$R triggered transcription factors and not complement-dependent cytotoxicity to act as the effector pathway leading to vascular injury.[10]

The association of AT$_1$Rab (pretransplant or of *de novo* synthesis) with acute rejection has been well documented, particularly in humoral responses;[22-24] it has been described as a severe vascular rejection, resistant to therapy even in transplants of grafts obtained from identical HLA relatives.[7] The development of acute rejection refractory to steroid therapy and/or anti-lymphocyte Abs foretells the risk for an aggressive clinical course and/or graft loss.[25-27] Furthermore, recent studies have described an association between AT$_1$Rab and graft loss regardless of the development of HLA-DSA.[23,24] We must underscore the fact that if AT$_1$Rab and HLA-DSA coexist, the risk of graft loss is increased when compared to only the presence of HLA-DSA.[24,28]

Angiotensin receptor blockers such as losartan block the effects mediated by AT$_1$Rab and have been used to decrease the Ab titers as have plasmapheresis and intravenous immunoglobulin.[7,29]

The purpose of this study was to evaluate the renal graft's function and the acute rejections events documented by biopsy, during the first year post

renal transplant in patients with AT_1Rab before transplant, and compare them to kidney transplant recipients with pre-transplant donor specific HLA Abs (DSA) only, and patients with neither anti-AT1R Abs nor HLA-DSA before transplant.

MATERIAL AND METHODS

This is a retrospective, cohort study of living donor renal transplant recipients (RTR) at the Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán (Mexico City), transplanted between March 2009 and August 2012. We systematically store pre-transplant serum samples from the donor and the recipient for future studies in the transplantation laboratory. They are stored at -70 °C until used that in this study, was September 2013. AT1Rab were determined by ELISA (CellTrend GMBH, Luckenwalde, Germany) and titers $\geq$ 17 IU were considered positive.[30,31] HLA-DSA determinations were performed with Luminex Labscreen Single Antigen beads class I and class II (One Lambda, Canoga Park, CA, USA); a MFI $\geq$ 500 was considered positive. HLA-DSA determination is systematically conducted as part of the patient's immunological risk evaluation in all renal transplant candidates in the Institute, as well as in the post-transplant period in those with biopsy-documented acute rejection (cellular or humoral) and 12 months after transplant coinciding with the graft's protocol biopsy. If acute rejection is suspected, defined as a $\geq$ 25% verified increase in serum creatinine when compared to baseline and in the absence of obvious causes (infection, urinary tract obstruction, dehydration, CNI toxicity), a percutaneous biopsy of the renal graft is performed.

Kidney transplant recipients (KTR) were divided into three groups according to their pre-transplant serological status:

*   KTR with positive AT_1Rab.
*   KTR with positive HLA-DSA, and
*   KTR with neither AT_1Rab nor HLA-DSA.

Patients with both anti-AT_1R and HLA-DSA were excluded.

Demographic variables were obtained from the donor and recipient, their history of pre-transplant cardiovascular disease and sensitization, the risk of CMV, ischemia duration, induction and maintenance immunosuppressive therapy.

The analyzed outcomes included: renal function during the first year after transplant (MDRD equation); incidence of acute rejection documented by graft biopsy; development of de novo HLA-DSA; and development of hypertension in that same time period.

Statistical analysis

Categorical variables are shown as frequencies and proportions whereas continuous variables were analyzed with Kolmogorov-Smirnov test in order to determine their distribution. Those variables with normal distribution are shown as mean ± standard deviation and median with range for those with abnormal distribution. Between-group comparisons were established by χ^2 or Fisher's exact test for categorical variables and with ANOVA or Kruskal-Wallis' test for continuous variables (distribution depending). Multivariate analysis with linear regression was used to determine which variables affected the eGFR one year after transplant. All possible associated factors to an eGFR < 70 mL/min/1.73 m^2 one year post-transplant, were also analyzed by logistic regression. P < 0.05 was considered statistically significant.

RESULTS

Of 174 renal transplants performed during the study period, 120 kidneys were obtained from a living related donor and of these, 114 had available sera for AT_1Rab determination.

According to the Ab determination in pre-transplant sera, 13 patients were included in the AT_1Rab group, 8 patients in the DSA group, and 90 patients in the group with neither AT_1Rab nor HLA-.DSA antibodies. Three patients having both AT_1Rab and HLA-DSA were excluded from the analysis.

Demographic characteristics and the relevant variables analyzed are shown in table 1. No differences were observed in donor eGFR, donor/recipient gender/age, recipient body mass index, kidney transplant number, haplotype-matches, warm/cold ischemia duration in all groups, and there were no cases of delayed graft function. Tacrolimus trough levels during the first year were similar in the 3 groups, as well as ACE inhibitor/AT_1R blocker use or hypertension development during the first year post-transplant.

There were differences in terms of the induction immunosuppressant therapy used between groups, particularly that of thymoglobulin in patients with no Abs. However, this was due to the fact that 3 patients in that group underwent a second transplant and 4 had a very high %PRA. Regardless, the %PRA was lower in the serologically negative group of pa-

Table 1. Demographic characteristics of donors and recipients.

KTR characteristics (n = 111)	AT$_1$Rab (n = 13)		HLA DSA (n = 8)		No Abs (n = 90)		P value
Female gender recipient	6	(46.15%)	3	(37.5%)	41	(45.56%)	0.75
Female gender donor	9	(69.2%)	5	(62.5%)	53	(58.8%)	0.76
Recipient age at KT (years)	26	(25-35)	26	(25-44)	28	(22-40)	0.8
Donor age at KT (years)	37	(29-51)	29.5	(25-37)	41	(29-45)	0.22
Second kidney transplant	0		0		3	(3.3%)	0.68
High risk for CMV infection	0		1	(12.5%)	14	(15.56%)	0.30
Warm ischemia time (min)	5	(4-6)	3	(3-8)	4	(3-5)	0.10
Cold ischemia time (hours)	1	(1-2)	1	(1-2)	1	(1-2)	0.75
Induction IS therapy:							
None	2	(15.3%)	0		11	(12.3%)	0.63
Anti-CD25	11	(84.6%)	2	(25%)	72	(79.7%)	0.01
Thymoglobulin	0		6	(75%)	7	(7.8%)	< 0.001
Class I % PRA	1.23	(0-7)	18.4	(0-70)	4.35	(0-76)	0.01
Class II % PRA	5.9	(0-37)	25	(0-83)	2.4	(0-74)	< 0.001
0 Haplotypes	3	(23%)	4	(50%)	29	(32.5%)	0.57
1 Haplotype	8	(61.5%)	4	(50%)	48	(53.3%)	0.87
2 Haplotypes	2	(15.3%)	0		12	(13.3%)	0.48
Donor pre-N eGFR* (ml/min/1.73 m^2)	106	(88-119)	108	(105-119)	108	(94-117)	0.83
PreTx hypertension	6	(50%)	6	(75%)	72	(80.9%)	0.057
PreTx use of ACEIs or ARBs	4	(33.3)	4	(50%)	46	(52.27%)	0.49
BMI pre KT	25.1	(21.6-25.6)	24.05	(22.5-25.3)	23	(21-26.5)	0.92
BMI at 1st year PT	23.9	(21.6-26)	27	(24.7-29)	23.5	(21-26)	0.46
Median tacrolimus levels during 1st year (ig/dL)	10.3	(9.5-10.8)	10	(9.1-11.14)	9.8	(8.8-10.7)	0.46

KT: kidney transplantation. Pre-N: pre-nephrectomy. *eGFR: estimated glomerular filtration rate by CKD-EPI. PreTx: pre-transplant. ACEIs: angiotensin-converting enzyme Inhibitors. ARBs: angiotensin receptor blockers. BMI: body mass index.

tients (Table 1). During follow-up, all patients were on triple immunosuppressive therapy with tacrolimus, mycophenolate mofetil and steroids.

Main outcomes

Table 2 shows renal function evolution for a year after transplant and comparing all three KTR groups. A decreased eGFR one year post-transplant is evident in patients with AT$_1$Rab (51.7, r 43-63.6 mL/min) when compared with KTR and positive HLA-DSA (58.3, r 52.8-72.8 mL/min) and the KTR group with no antibodies (71.5 r 56.3-82.6 mL/min), p = 0.03. Only the difference between AT$_1$Rab group and no antibodies group was significant (Bonferroni test: p = 0.049). The eGFR difference between patients with HLA-DSA versus the other two groups was not significant. We must emphasize that from the first month post-transplant, the eGFR was significantly lower in the group of patients with AT$_1$Rab when compared with no antibodies group (p = 0.01). When comparing all three groups in terms of the eGFR delta between the first and twelfth month post-transplant, no statistically significant differences were established (Table 2).

Despite a higher percentage of biopsy-proven acute rejection in the AT$_1$Rab group, there was no significant difference between the groups. We must mention that the acute rejection events in the AT$_1$ab group, tend to occur at a latter post-transplant time period when compared to those documented in the other two groups.

In terms of de novo DSA appearance and the frequency of hypertension one year after transplant, no difference between groups was detected (Table 2).

Lineal regression multivariate analysis showed four factors independently and significantly associated with this outcome:

- Biopsy proven acute rejection (β = -18.7, 95%CI -28.2 to -9.26; p < 0.001).
- AT$_1$Rab (β = -10.51, CI -20.9 to -0.095; p = 0.048).
- Donor age (β = -0.42, CI -0.75 to -0.103; p = 0.01) and recipient age (β = -0.36, CI -0.67 to -0.048; p = 0.024).

Table 2. Main outcomes.

KTR characteristics (n = 111)	anti-AT$_1$R (n = 13)	anti-HLA DSA (n = 8)	Without Abs	P value
1st month PT SCr (mg/dL)	1.35 (1.13-1.47)	1.11 (0.9-1.4)	1.03 (0.86-1.26)	0.04
1st month PT eGFR (mL/min/1.73 m^2)	62.9 (49.6-69.8)	81.96 (53.5-95.5)	78.55 (61.25-90.8)	0.01
3rd month PT SCr (mg/dL)	1.36 (1.14-1.54)	1.18 (1.13-1.2)	1.09 (0.99-1.34)	0.1
3rd month PT eGFR (mL/min/1.73 m^2)	58.6 (50.9-68.4)	65.7 (55-69.9)	74.1 (59.1-85.6)	0.08
1st year PT SCr (mg/dL)	1.48 (1.34-1.6)	1.21 (1.15-1.47)	1.14 (0.99-1.42)	0.03
1st year PT eGFR (mL/min/1.73 m^2)	51.7 (43-63.6)	58.4 (52.8-72.8)	71.5 (56.3-82.6)	0.03
Δ eGFR 1st year - 1st month PT	2 (-0.53 a 10.26)	8.74 (-3.17 a 18.31)	4.3 (-8.5 a 13.8)	0.77
Hypertension 1 year PT	4 (33.3%)	2 (33.3%)	41 (39%)	0.72
Use of ACEIs or ARBs during 1st year PT	1 (8.3%)	1 (14.2%)	19 (22%)	0.7
Systolic arterial pressure	115 (100-125)	120(110-130)	120 (110-120)	0.35
Diastolic arterial pressure	72.5 (65-80)	80 (70-80)	76 (70-80)	0.69
PT de novo HLA DSA	3 (23%)	0	15 (16.8%)	0.33
PT de novo HLA DSA time to detection (months)	7.8 (7.6-10.3)	0	11.4 (4.1-12)	0.91
KTR with evidence of acute rejection	4* (30.7%)	1 (11.1%)	12** (13.4%)	0.25
PT time to 1st AR episode (months)	6.1 (3.9-10.3)	0.16	1.2 (0.2-5.4)	0.09
Acute cellular rejection events	3 (23.1%)	0	7 (7.8%)	0.12
Acute humoral rejection events	2 (15.3%)	1 (11.1%)	7 (7.8%)	0.65

* 1 patient with evidence of both cellular and humoral rejection ** 2 patients with evidence of both cellular and humoral rejection. KTR: kidney transplant recipient. PT: post-transplant. SCr: serum creatinine. eGFR: estimated glomerular filtration rate by MDRD. ACEIs: angiotensin-converting enzyme inhibitors. ARBs: angiotensin receptor blockers. PT: post-transplant. AR: acute rejection.

Logistic regression multivariate analysis

When analyzing factors relating to an eGFR below 70 mL/min after a one-year post-transplant follow-up, five variables remained significant: biopsy proven acute rejection, OR 6.27 (95%CI 1.18-33.06, p = 0.039), AT$_1$Rab, OR 6.17 (1.09-34.79, p = 0.035), de novo DSA, OR 4.49 (1.11-18.13, p = 0.035), recipient age, OR 1.04 (1.005 -1.09, p = 0.027) and donor age, OR 1.06 (1.01 - 1.11, p= 0.009).

DISCUSSION

The documented association between AT$_1$Rab and graft function deterioration in the early post-transplant period is interesting, particularly because the Abs effect on this variable was independent of other possible causes that could explain the decreased glomerular filtration rate observed in the AT$_1$Rab positive KTR group, even during the first month post-transplant. Although no other study has, to date, reported AT$_1$Rab as a cause of early graft dysfunction –in the absence of acute rejection–, our results could partly explain their negative impact on graft survival; two very recent studies on AT$_1$Rab (pre-transplant or de novo) have clearly demonstrated the presence of these Abs representing an independent risk factor.[23,24] Giral, et al., screened 599 KTR before transplantation to determine the degree of pre-sensitization against AT$_1$R as a risk factor for graft survival and acute rejection (AR). They established a threshold of AT$_1$Rab levels of 10 IU based on the time period to graft failure. AT$_1$Rab > 10 IU were detected in 283 patients (47.2%) before transplant. KTR who had AT$_1$Rab > 10 IU had a 2.6-fold higher risk of graft failure, 3 years PT and subsequently (p = 0.0005).[23] Moreover, Taniguchi, et al., tested PT AT$_1$Rab and HLA-DSA in sera from 351 KTR; 134 with BPR and/or lesions constituted the abnormal biopsy group (ABG) and 217 patients, the control group (CG). The ABG's rate of AT$_1$Rab was significantly higher than in the CG (18 vs. 6%, p < 0.001), and 79% of ABG patients with positive AT$_1$Rab (> 15 IU) lost their grafts vs. 0% in the CG. In Tanguchi's, et al., study, patients with both AT$_1$Rab and HLA-DSA had lower graft survival than those with HLA-DSA alone (log-rank p = 0.007).[24] The synergistic negative effect of AT$_1$Rab and HLA-DSA on kidney transplants was initially described in a patient with accelerated kidney transplant rejection and hypertensive encephalopathy.[28] In our study, a higher threshold for AT$_1$Rab positivity (≥ 17 IU) was used than that in the description of acute rejection and graft loss;[23,24] this could have added strength to the association of AT$_1$Rab with worse graft function.

As previously mentioned, we determined that renal function deterioration begins during the first

month post-transplant, as reflected by a decreased eGFR in the AT_1Rab group when compared with the HLA-DSA positive group and that with no antibodies (62.9 mL/min *vs.* 81.9 mL/min *vs.* 78.5 mL/min, respectively). However, the renal function deltas at one and twelve months post-transplant, were similar in all three groups. Despite the limited study sample, this finding suggests that the main change in kidney function developed in the early post-transplant period and that the AT_1Rab could be responsible, since all three groups are most similar when comparing all possible variables that could compromise early eGFR between groups (Table 1), including the donors' eGFR. We must emphasize that the documented acute rejections in the AT_1Rab positive group developed after the eGFR was documented in the first post-transplant trimester. Although this study does not allow the establishment of a causal relationship between positive AT_1Rab and graft dysfunction, the fact that these Abs promote transplant vasculopathy by increasing the effects of angiotensin II has been well-documented; they promote cell growth, apoptosis, fibrosis, extracellular matrix remodeling and inflammation.[32-34] In any case, our findings suggest initial hemodynamic injury mediated by AT_1Rab, although it apparently is not progressive during the first year. This contrasts with the injury mediated by HLA-DSA that tends to progress after initial injury, as documented in various studies. Several pathways have been suggested by which AT_1Rab may develop.[3,10,16] The transplant process itself may lead to increased AT_1R expression, thus exacerbating the effects of antibodies on the receptors of activated vascular cells. Ischemia and reperfusion may also lead to oxidative stress, triggering an alloantigen-mediated immune response.[10,35,36] Others have postulated that the endothelium's natural balance may be altered, thus increasing its susceptibility to an antibody attack by AT_1Rab and generating a cascade of events leading to vascular rejection. AT_1Rab may amplify local inflammation, increase antigen expression and the production of Th1 cytokines and inflammatory chemokines.[10]

In terms of acute rejection episodes, our study found an increased proportion of rejection, both humoral and cellular, in the positive AT_1Rab group although this was not statistically significant. Whether the Abs are directly responsible for injury or are only a marker of the inflammatory milieu leading to the vascular injury and culminating in graft loss, remains to be determined.

It is important to increase our study sample and evaluate our patients by measuring post-transplant AT_1Rab titers in order to determine their behavior over time, as well as follow the patients' renal function for a longer time period to establish whether the same findings persist.

AT_1Rab may represent a relevant link in the chain of factors compromising renal graft injury. If subsequent research provides similar results, we would expect that the determination of these Abs will be part of the routine evaluation of patients in the pre-transplant period, in the estimation of their immunological risk.

In summary, anti-AT_1R Abs in KTR pre-transplant sera was an independent and significant risk factor for decreased eGFR early in the post-transplant period. This finding deserves to be confirmed in a larger KTR population.

ACKNOWLEDGEMENTS

REFERENCES

1. Sumitran-Karuppan S, Tyden G, Reinholt F, et al. Hyperacute rejections of two consecutive renal allografts and early loss of the third transplant caused by non-HLA antibodies specific or endothelial cells. *Transpl Immunol* 1997; 5: 321.
2. Sumitran-Holgersson S, Wilczek HE, Holgersson J, et al. Identification of the nonclassical HLA molecules, MICA, as targets for humoral immunity associated with irreversible rejection of kidney allografts. *Transplantation* 2002; 74: 268.
3. Dragun D. Humoral responses directed against non-human leukocyte antigens in solid-organ transplantation. *Transplantation* 2008; 86: 1019.
4. Amico P, Honger G, Bielmann D, et al. Incidence and prediction of early antibody-mediated rejection due to non human leukocyte antigenantibodies. *Transplantation* 2008; 85: 1557.
5. Rose ML. Role of MHC and non-MHC antibodies in graft rejection. *Curr Opin Organ Transplant* 2004; 9: 16.
6. Aguilera I, Alvarez-Marquez A, Gentil MA, et al. Anti-glutathione Stransferase T1 antibody-mediated rejection in C4d-positive renal allograft recipients. *Nephrol Dial Transplant* 2008; 23: 2393.
7. Dragun D, Muller DN, Brasen JH, et al. Angiotensin II type 1-receptor activating antibodies in renal allograft rejection. *N Engl J Med* 2005; 352: 558.
8. Hunyadi L, Catt KJ. Pleiotropic AT1 receptor signaling pathways mediating physiological and pathogenic actions of angiotensin II. *Mol Endocrinol* 2006; 20: 953.
9. Cheng ZJ, Vapaatalo H, Mervaala E. Angiotensin II and vascular inflammation. *Med Sci Monit* 2005; 11: RA194-RA205.
10. Reinsmoen NL. Role of angiotensin II type 1 receptor-activating antibodies in solid organ transplantation. *Hum Immunol* 2013; 74: 1474.
11. Dragun D. The role of angiotensin II type 1 receptor activating antibodies in renal allograft vascular rejection. *Pediatr Nephrol* 2007; 22: 911.

12. Schiffrin EL, Touyz RM. Multiple actions of angiotensin II in hypertension: benefits of AT1 receptor blockade. *J Am Coll Cardiol* 2003; 42: 911.
13. Hunyadi L, Turu G. The role of the AT1 angiotensin receptor in cardiac hypertrophy: angiotensin II receptor or stretch sensor? *Trends Endocrinol Metab* 2004; 15: 405-08.
14. Maric C, Zheng W, Walther T. Interactions between angiotensin II and atrial natriuretic peptide in renomedullary interstitial cells: the role of neutral endopeptidase. *Nephron Physiol* 2006; 103: 149-56.
15. Dzau VJ. Tissue angiotensin and pathobiology of vascular disease: A unifying hypothesis. *Hypertension* 2001; 37: 1047.
16. Dragun D, Philippe A, Catar R. Role of non-HLA antibodies in organ transplantation. *Transplantation* 2012; 17: 440.
17. Fu ML, Herlitz H, Schulze W, et al. Autoantibodies against the angiotensin receptor (AT1) in patients with hypertension. *J Hypertens* 2000; 18: 945.
18. Wallukat G, Homuth V, Fischer T, et al. Patients with preeclampsia develop agonistic autoantibodies against the angiotensin AT1 receptor. *J Clin Invest* 1999; 103: 945.
19. Zhang S, Zheng R, Yang L, et al. Angiotensin type 1 receptor autoantibody from preclamptic patients induces human fetoplacental vasoconstriction. *Jour Cell Phys* 2012; 228: 142.
20. LaMarca B, Parrish MR, Wallace K. Agonistic autoantibodies to the angiotensine II type I receptor cause pathophysiologic characteristics of preeclamsia. *Gend Med* 2012; 9: 139.
21. Miller JA, Scholey JW. The impact of renin-angiotensin polymorphisms on physiological and pathophysiological processes in humans. *Curr Opin Nephrol Hypertens* 2004; 13: 101.
22. Reinsmoen NL, Lai CH, Heidecke H, et al. Anti-angiotensin type 1 receptor antibodies associated with antibody mediated rejection in donor HLA antibody negative patients. *Transplantation* 2010; 90: 1473.
23. Giral M, Foucher Y, Dufay A, et al. Pretransplant sensitization against angiotensin II type 1 receptor is a risk factor for acute rejection and graft loss. *Am J Transpl* 2013; 10: 2567.
24. Taniguchi M, Rebellato LM, Cai J, et al. Higher risk of kidney graft failure in the presence of anti-angiotensin II type 1 receptor antibodies. *Am J Transpl* 2013; 13: 2577.
25. Van Saase J, Van der Woude FJ, Thorogood J, et al. The relation between acute vascular and interstitial renal allograft rejection and subsequent chronic rejection. *Transplantation* 1995; 59: 1280.
26. Zhang G, Wang H, Wang F, et al. Gene polymorphism of the renin-angiotensin-aldosterone system and angiotensin II type 1-receptor activating antibodies in renal rejection. *Tohoku J Exp Med* 2007; 213: 203.
27. Dragun D, Bräsen JH, Schönemann C, et al. Patients with steroid refractory acute vascular rejection develop agonistic antibodies targeting angiotensin II type 1 receptor. *Transplant Proc* 2003; 35: 2104.
28. Kelsch R, Everding AS, Kuwertz-Bröking E, et al. Accelerated kidney transplant rejection and hypertensive encephalopathy in a pediatric patient associated with antibodies against angiotensin type 1 receptor and HLA class II. *Transplantation* 2011; 92: e57.
29. Dragun D. Agonistic antibody-triggered stimulation of angiotensin II type 1 receptor and renal allograft vascular pathology. *Nephrol Dial Transplant* 2007; 22: 1819.
30. Slowinski T, Suker D, Schonemann C. Screening of patients on waiting list for a renal transplant for agonistic non-HLA antibodies targeting angiotensin II type 1 receptor. *J Am Soc Nephrol* 2006; 17: 393A.
31. Dragun D. The detection of antibodies to the angiotensin II-type 1 receptor in transplantation. *Methods Mol Biol* 2013; 1034: 331.
32. Yousufuddin M, Cook DJ, Starling RC, et al. Angiotensin II receptors from peritransplantation through first-year-post-transplantation and the risk of transplant coronary artery disease. *J Am Coll Cardiol* 2004; 43: 1565.
33. Ruiz-Ortega M, Lorenzo O, Rupérez M, et al. Role of renin-angiotensin system in vascular disease. *Hypertension* 2001; 38: 1382.
34. Dragun D, Catar R, Kusch A. Non-HLA-antibodies targeting angiotensin type 1 receptor and antibody mediated rejection. *Hum Immunol* 2012; 73: 1282.
35. Win TS, Pettigrew GJ. Humoral autoimmunity and transplant vasculopathy: when allo is not enough. *Transplantation* 2010; 90: 113.
36. Lukitsch I, Kehr J, Chaykovska L, et al. Renal ischemia and transplantation predispose to vascular constriction mediated by angiotensin II type 1 receptor-activating antibodies. *Transplantation* 2012; 94: 8.

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