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Non-HLA Antibody Induced Agonism on the Angiotensin II Type 1 Receptor in Renal Allograft Vascular Injury

Duska Dragun

Department of Nephrology and Intensive Care Medicine Campus Virchow-Klinikum and Center for Cardiovascular Research Medical Faculty of the Charité Berlin, Berlin, Germany

Abstract

The rising incidence of steroid-refractory rejections is a challenging problem in renal allotransplantation. Etiological studies implicate either an overwhelming T-cell response or, more frequently, involvement of alloantibodies. Alloantibodies induce a spectrum of histologic tubulointerstitial and vascular changes paralleled with immunohistochemical positivity for C4d along peritubular capillaries. The degree of vascular involvement seems to be a more important prognostic determinant. Fibrinoid necrosis of the arteries with secondary thrombotic occlusions is C4d negative in 50% of cases and has a worst prognosis among all allograft vascular lesions. Apart from donor-specific human leukocyte antigen antibodies, non-human leukocyte antigen antibodies reacting to arterial antigens have been speculated to be responsible for rejections in some patients. We recently reported the presence of agonistic antibodies against the angiotensin II type 1 receptor (AT_1R-AA) in 16 recipients of renal allografts who had severe vascular rejection and malignant hypertension, but who did not have anti-human leukocyte antigen antibodies. The AT_1R-AA appear to be non-complement-fixing autoantibodies targeting the second extracellular loop of AT_1R. The AT_1R-AA act as allosteric activators on AT_1R and induce mediators of inflammation and thrombosis. Transfer of AT_1R-AA into rats with kidney allografts induced vasculitis and hypertension, supporting the notion that AT_1R-AA are not an epiphenomenon. Removal of AT_1R-AA by plasmapheresis in combination with pharmacologic AT_1R blockade leads to improved renal function and graft survival in AT_1R-AA-positive patients. We have shown that the analysis of the subtle diagnostic and mechanistic differences may help to identify patients at particular risk and improve outcome of steroid-refractory rejections with vascular pathology. **(Trends in Transplant. 2007;1:113-20)**

Corresponding author: Duska Dragun, duska.dragun@charite.de

Key words

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Correspondence to:
Duska Dragun
Department of Nephrology and Intensive Care Medicine
Charité Campus Virchow Clinic
Augustenburger Platz 1
13353 Berlin, Germany
E-mail: duska.dragun@charite.de

Introduction

The successful developments in immunosuppressive modalities that were designed to target T-cell-mediated immune response and prevent destruction of tubular epithelia have resulted in the reduction of acute rejection episodes and improved overall allograft survival[1]. On the other hand, humoral presensitization emerges as one of the major risk factors for renal allograft rejection and allograft loss[2], despite the fact that most of the patients with antibody mediated rejection (AMR) have negative crossmatch. Generally, AMR has a worse prognosis than the T-cell-mediated rejection, which forced significant research efforts during the last decade[3]. Antibody mediated rejection remains a diagnostic and therapeutic challenge and can occur with all immunosuppression regimens, even in the context of profoundly depletional therapy[4]. It is not possible to distinguish AMR from T-cell-mediated rejection on simple clinical grounds. However, when AMR occurs in organ transplants, the process resists conventional treatment approaches and frequently leads to allograft loss[5]. The morphologic features apply to a wide spectrum of tubulointerstitial and vascular lesions in the allograft also, including severe changes like thrombosis, fibrinoid necrosis of the arteries, and endarteritis[6,7]. Neutrophils in the capillaries are characteristically but not always found[6]. Among different morphologic features, vascular lesions carry the worst prognosis[8]. The association of antidonor humoral reactivity against human leukocyte antigen (HLA) class I antigens and vascular rejection has been documented in studies by Halloran, et al. more than a decade ago[9]. Donor-specific anti-HLA alloantibodies initiate rejection through complement-mediated and antibody dependent cell-mediated cytotoxicity[10]. The diffuse staining of the complement degradation product C4d affecting the surface of peritubular capillaries is generally regarded as a marker for HLA antibody mediated alloresponse and is associated with inferior graft survival[11]. Nevertheless, 40-50% of rejections with severe vascular changes such as fibrinoid necrosis are C4d negative, implicating involvement of non-complement-fixing antibodies against undefined targets[12]. Clinical, etiological, and histopathologic heterogeneity of AMR emphasizes the necessity for better recognition of the subtle etiologic differences between affected patients. Unknown immune targets and consecutive lack of detection methods make non-HLA AMR particularly difficult to diagnose and treat.

Relevance of non-HLA antibody response

Putative pathogenic antibodies that are not directed against the HLA system were considered in recipients who rejected HLA-identical kidneys more than three decades ago[13]. However, characterization of non-HLA antibodies remains very poor; many appear to be autoantibodies[14]. In renal allograft rejection, the presence of antibodies to non-HLA antigens has been associated with antibodies against endothelial cells, tubular epithelial cells, podocytes, mesangial cells, and monocytes[14]. Most of the efforts in the past were focused on anti-endothelial cell antibodies[15]. Anti-endothelial cell antibodies are a heterogeneous group of antibodies directed against a variety of antigenic determinants, but the existence of a common polymorphic non-HLA antigen system in endothelial cells could not be confirmed by biochemical identification of the relevant antigens[16]. They have been reported in a variety of autoimmune diseases featuring vasculitis as a denominator[15]. The most comprehensive evidence about their biologic relevance is derived from studies using immunoglobulins isolated from patients with systemic sclerosis[17]. Anti-endothelial cell antibodies are especially common in renal transplant recipients who are pre-sensitized

against a panel of HLA antigens[18]. Non-HLA-reactive anti-endothelial cell antibodies seem to recognize endothelial cell antigens that can be induced upon tumor necrosis factor-α (TNFα) and interferon-γ (IFNγ) stimulation, implicating a permissive role of endothelial activation that may be a prerequisite for the pathogenicity of anti-endothelial cell antibodies[18].

Similarly to some autoimmune diseases, non-HLA antibodies may be diagnostic of disease, but they may not necessarily be an effector mechanism. For this reason, it is important to identify non-HLA antibodies, determine their antigen specificity and pathogenicity, and clarify the mechanisms by which they contribute to rejection or other forms of allograft injury.

Clinical manifestations of AT_1R-AA related vascular rejections

We reported the presence of agonistic antibodies against the angiotensin II type 1 receptor (AT_1R-AA) in 16 recipients of renal allografts who had severe vascular rejection and malignant hypertension, but who did not have donor-specific anti-HLA antibodies[19]. These AT_1R-AA have also been associated with preeclampsia and malignant hypertension[20,21]. Pregnancies complicated by preeclampsia and graft rejection bear some immunologic similarities[22]. The decision to seek and isolate AT_1R-AA was instigated by the observation that the first patient we studied developed accelerated vascular rejection refractory to steroids and anti-lymphocyte antibody preparations in a "zero-mismatch" kidney. As this patient developed malignant hypertension with seizures during the rejection process, the clinical picture was so reminiscent of eclamptic crisis in pregnancy, a condition that she had developed two decades before transplantation, that we started to prospectively look for patients with similar clinical features. We detected our further 15 patients primarily based on severe vascular pathology, absence of donor-specific antibodies, hypertensive crisis accompanied by seizures in three other patients, and lack of response to steroids or anti-lymphocyte preparations. All patients in our study had primary graft function.

Clinicopathologic features of AT_1R-AA-related process in patients were noticed between day 5 and 14 posttransplantation, with minor interindividual differences concerning temporal occurrence of maximal allograft injury and increase in blood pressure. Most of patients (13/16) did not have hypertension before vascular rejection occurred, implying that the posttransplant hypertension was most likely secondary to rejection. Some of the patients developed thrombopenia together with other signs of microangiopathic hemolysis such as an elevated lactic dehydrogenase and presence of schistocytes. Reactivation or *de novo* cytomegalovirus infection was excluded in all patients. Autoimmune and hereditary causes of thrombophilia were also ruled out.

Causes of end-stage renal disease (ESRD) were not primarily attributed to hypertensive nephrosclerosis, but instead to a variety of tubulointerstitial or glomerular diseases. None of the patients in our study was reported to have ESRD due to renal involvement in a systemic autoimmune disease or hemolytic uremic syndrome/thrombotic thrombocytopenic purpura. We believe that the specific cause of ESRD was not relevant for the development of AT_1R-AA in our patients. The frequency of AT_1R-AA-positive rejections was equal between both female and male renal transplant recipients. The AT_1R-AA-related vascular rejections occurred during the first week after transplantation. These rejections had significantly shorter allograft survival and more severe histology, irrespective of the

treatment, compared to donor-specific anti-HLA-antibody associated rejections[19].

Morphologic features of AT_1R-AA-related vascular rejections

The majority of AT_1R-AA-positive patients detected in the initial study developed Banff type III rejection with fibrinoid arterial wall necrosis and secondary thrombotic occlusion with allograft infarction. In some of the patients, first biopsies were presented with Banff type II rejection, transmural arteritis (transplant endarteritis), and transplant glomerulitis. Apart from arterial and capillary changes, we also noticed tubulitis and interstitial infiltrates, characteristic for acute cellular rejection. Thus, some biopsies seem to fall into the category of "mixed rejection". However, unlike so-called "mixed AMR"[23], our patients did not respond to aggressive T-cell depletion therapies. In few patients, hyperacute rejection manifested as fibrinoid arterial wall necrosis, and interstitial hemorrhage with frank necrosis of tubular epithelia developed during the first three posttransplant days. Thus, AT_1R-AA-positive rejections tend to share many morphologic features with both "pure" and "mixed" HLA-antibody mediated AMR. Patients with AT_1R-AA-positive rejections had worse vascular scores and Banff grade compared to those with HLA-mediated AMR. In contrast to HLA antibodies, AT_1R-AA seem to operate through complement-independent mechanisms, as C4d was detected in biopsy specimens from only five of our 16 patients[19].

Diagnostic and therapeutic implications

A very short period from transplantation to rejection episode implicated a possible relevance of preformed antibodies. Retrospective analysis of historic sera from our patients obtained before transplantation showed positivity for AT_1R-AA, which confirms that preformed and not *de novo* produced AT_1R-AA were likely responsible for vascular rejection. The AT_1R-AA are low-titer IgG1 and IgG3 (complement-fixing subclass) antibodies, yet they do not form immunocomplexes with the antigen. Detection of AT_1R-AA activity initially relied on the bioassay that measures the chronotropic responses to AT_1R-IgG-mediated stimulation of cultured cardiomyocytes coupled with receptor-specific antagonists. The dose/response relationship between AT_1R-AA concentration and the chronotropic response is linear. High costs and a time-consuming test setting precluded screening of larger patient cohorts by the bioassay at the time when the initial study was performed. Only patients with suggestive clinical features and biopsy findings, and not all patients with allograft dysfunction, underwent bioassay test.

In our initial study, seven of 16 patients with AT_1R-AA were treated with a combination of plasmapheresis, intravenous immune globulin infusions, and the AT_1R-blocker, losartan (100 mg daily). This combination treatment led to improved renal function and graft survival, compared to the outcomes amongst patients with AT_1R-AA who received standard treatment for AMR and rapidly lost their allografts[19]. None of the patients received angiotensin converting enzyme (ACE) inhibitors or AT_1R blockers prior to rescue protocol, as the practice of our transplant center was not to use them in the posttransplant period. Among seven treated patients, four remained rejection free and AT_1R-AA negative (longest follow-up, seven years). They still continuously receive 50 mg losartan daily. One patient died six months after successfully treated AT_1R-AA rejection with functioning graft due to herpes simplex virus encephalitis. Two patients who were initially rescued from AT_1R-AA rejection developed *de novo* anti-HLA class I directed against A29 and A8 loci of the donor-

specific antibody and C4d positive glomerular type of humoral rejection without affection of arterial vessels after a four- and six-month period of stable allograft function, respectively. They required rituximab rescue therapy due to plasmapheresis and intravenous immunoglobulin resistance.

Subsequent serum samples were obtained from five of eight patients who were not treated with our losartan-including rescue protocol. Two of these patients became AT_1R-AA negative approximately two months after transplant nephrectomy (removal of antigen). However, three other patients remain AT_1R-AA positive despite transplant nephrectomy. Interestingly, all these three patients previously lost two transplants due to undefined accelerated vascular rejections. We speculate that these patients may have altered plasma cell memory repertoire in terms of long-lived plasma cells that are not responsive to removal of antigen compared to other patients.

We are aware that the small number of patients in our initial study may limit the degree to which the results can be generalized, and that bioassay testing under a suggestive indication provided left-censorship bias. However, we believe our findings, even with their limitations, are highly significant. Due to high costs and our time-consuming bioassay, larger studies were initially not feasible. We have now established and validated a cell-based ELISA in collaboration with biotech partners for detection of AT_1R-AA in serum[24]. The ELISA currently has 100% specificity and 88% sensitivity. Variability between assays is 12%[24]. Pretransplantation screening for AT_1R-AA detects a subset of ESRD patients who are similar but not identical to patients with anti-HLA-panel reactivity. Pretransplantation screening of recipients for AT_1R-AA may help to improve individual risk assessment and offer patients with AT_1R-AA preemptive specific treatment. Whether AT_1R-AA acts as a progression factor during native renal disease, or as an independent factor of cardiovascular comorbidity, remains to be determined. Whether all AT_1R-AA-positive patients who will be continuously treated with AT_1R blockade will develop milder or no vascular rejection is the subject of current studies. Some transplant nephrologists are the only remaining clinicians skeptical about the use of anti renin-angiotensin system (RAS) drugs due to the concern of interference with renal allograft perfusion. According to reported beneficial effects of blockade of RAS on early outcomes of renal transplants, this view seems to be outdated[25]. Moreover, AT_1R antagonists may exert a clinically relevant immunomodulatory role by blocking IFNγ production by T-cells[26]. Pharmacologic action of AT_1R antagonists is based on inverse agonism, which implicates that reactive upregulation of AT_1R on target cells may, in case of therapy discontinuation, increase detrimental responses[27]. Given this consideration, perioperative discontinuation of ACE inhibitors or AT_1R blockers may thus predispose for AT_1R-AA-related pathologies. For example AT_1R-AA-positive patients who receive continuously AT_1R blockers or ACE inhibitors together with intensified immunosuppression (depletional antibody induction, tacrolimus, mycophenolate mofetil, and steroids) and are recipients of living-donor kidneys seem not to be prone to development of fulminant AT_1R-AA-related rejection[28].

Pathophysiologic consequences of antibody mediated AT_1R stimulation

The AT_1R-AA seem to induce vascular and tubulointerstitial pathology via mechanisms independent from complement activation that are distinct from those in patients with HLA antibodies. We raised and confirmed the hypothesis that AT_1R-AA may act in similar manner as an endogenous agonist for the AT_1R, angiotensin II, and exert direct effects

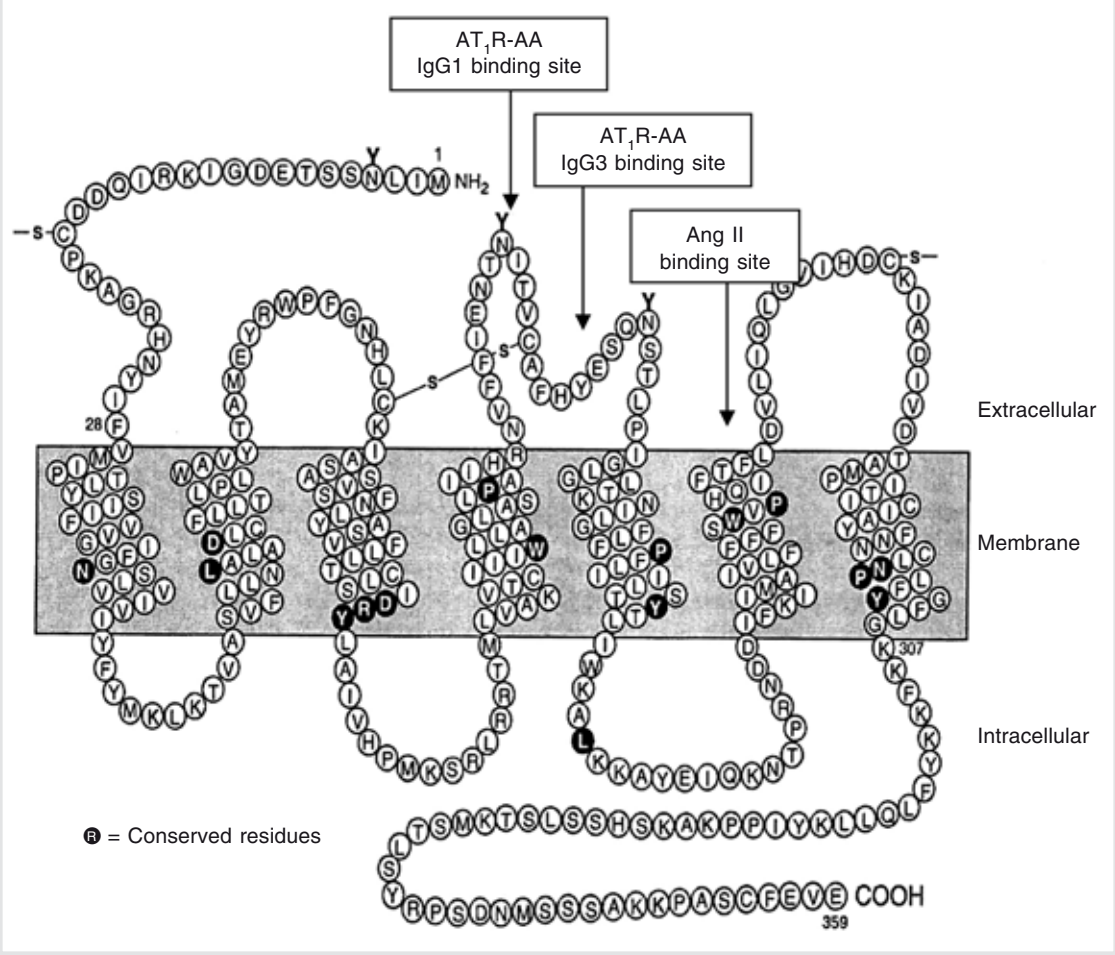

Figure 1. *Secondary structure and consensus sequence of the mammalian angiotensin II type 1 receptor. The amino acid sequence shown is based on the derived sequences of five individual cloned mammalian AT_1 receptors. The amino acid residues that are highly conserved among G-protein coupled receptors are indicated by bold letters. The positions of the three extracellular carbohydrate chains, and of the two extracellular disulfide bonds, are also indicated. IgG1 and IgG3 AT_1R-AA bind to the second extracellular loop of the receptor, while a natural agonist angiotensin II binds to transmembrane "pockets" close to the plasma membrane.*

on endothelial and vascular smooth muscle cells[29]. The responses elicited by AT_1R stimulation are context dependent and specific for target-cell lineage[30]. According to our working concept, AT_1R-AA bind to the second extracellular loop of AT_1R (Fig. 1) and act as an allosteric receptor agonist. The AT_1R-AA/AT_1R interaction initiates signal transduction cascades by inducing extracellular signal-regulated kinase 1/2 phosphorylation in endothelial and vascular smooth muscle cells. Consequent increasing DNA binding activity of transcription factors activated activator protein 1 (AP-1) and nuclear factor-κB (NFκB) is responsible for increased expression of their target genes involved in inflammatory responses and coagulation. Increased synthesis of chemokines MCP-1 and RANTES may probably explain intravascular inflammatory cell infiltration, while augmented activity and expression of tissue factor may account for thrombotic angiopathy. Although we have documented that AT_1R-AA belong to complement-fixing IgG1 and IgG3 antibodies, our

findings suggest that genes regulated by AT_1R-triggered transcription factors and not complement-directed cytotoxicity act as an effector pathway of the vascular injury. The illustrative example is that AT_1R-AA enhanced promoter activity of tissue factor, an initiator of extrinsic coagulation pathway and a target gene for AP-1 and NFκB *in vitro*. Tissue factor mediates clotting abnormalities associated with hyperacute and xenograft rejection, as well as in antiphospholipid antibody syndrome[31]. Accordingly, renal transplant biopsy specimens obtained during an AT_1R-AA-mediated rejection episode revealed intense diffuse tissue factor staining of epithelial, endothelial, and mesangial cells in absence of complement activation. Binding of AT_1R-AA to AT_1R expressed on target cells is a critical step for activating the downstream cascade and inducing damage to the allograft. However, we have not yet proven whether AT_1R-AA function only through pro-coagulatory and chemotactic activity, or whether they also act by means of innate and specific immune responses and increased vascular reactivity. Direct effects of AT_1R-AA on immune response are likely, since human T-cells are fully equipped with functioning components of the renin-angiotensin system and express AT_1R on their surface[32]. Our current working hypothesis is that factors surrounding the organ transplantation process may lead to increased expression of AT_1R and thereby affect the overall reactivity of the vascular cells to AT_1R-AA. Passive transfer of human IgG containing AT_1R-AA induced a transmural arteritis similar to the human situation and led to increased blood pressure in otherwise non-rejecting and normotensive transplanted animals. These findings provided further evidence that AT_1R-AA may have a causative role. Similar to stimulation of the AT_1R by its natural ligand, angiotensin II[29], agonistic receptor activation mediated by AT_1R-AA could play a key role in the initiation and amplification of pathobiological events that lead to transplant vasculitis and hypertension.

Unresolved questions

We have not explained whether or not AT_1R-AA-related pathology represents a "true-rejection" or an autoimmune phenomenon that becomes overt in dependence of permissive factors related to allogeneic environment and not yet elucidated factors related to the transplant procedure itself. An allogeneic background, brain death-associated "cytokine storm", reperfusion injury to the transplant, and/or use of calcineurin inhibitors or steroids are probably permissive factors responsible for an increased AT_1R density on target cells. For example, in heart transplantation, systemic upregulation of AT_1R could be found in donors with spontaneous intracerebral hemorrhage that was associated with subsequent development of cardiac vasculopathy[33]. However, the relative individual contribution of considered permissive factors needs to be further elucidated in order to better understand and prevent AT_1R-AA-related clinical syndrome. Another important question is, why do AT_1R-AA develop in patients with preeclampsia and ESRD and what is the role of antigen mimicry (cross-reactivity with bacterial or viral antigens) or genetic predispositions?

Conclusion

We provide a novel concept in the pathogenesis of accelerated vascular rejection process where autoimmune-mediated receptor activation is linked to a severe vascular pathology in the situation of allogeneic transplantation. At present we believe that pretransplantation testing of recipients for AT_1R-AA may help to improve individual risk assessment of patients with AT_1R-AA-preemptive specific treatment. We are also just beginning to learn more about pathophysiologic mechanisms and optimization of diagnostic and therapeutic modalities for AT_1R-AA-positive patients.

References

1. Meier-Kriesche HU, Schold JD, Kaplan B. Long-term renal allograft survival: Have we made significant progress or is it time to rethink our analytic and therapeutic strategies. Am J Transplant 2004;4:1289-95.
2. Terasaki P. Humoral theory of transplantation Am J Transplant 2003;6:665-73.
3. Colvin RB. Antibody-mediated renal allograft rejection: Diagnosis and pathogenesis. J Am Soc Nephrol 2007; 18:1045-56.
4. Lorenz M, Regele H, Schillinger M, et al. Risk factors for capillary C4d deposition in kidney allografts: Evaluation of a large study cohort. Transplant 2004;78:447-52.
5. Crespo M, Pascual M, Tolkoff-Rubin N, et al. Acute humoral rejection in renal allograft recipients: Incidence, serology and clinical characteristics. Transplant 2001;71:652-8.
6. Mauiyyedi S, Crespo M, Collins AB, et al. Acute humoral rejection in kidney transplantation: II. Morphology, immunopathology, and pathologic classification. J Am Soc Nephrol 2002;13:779-87.
7. Racusen LC, Colvin RB, Solez K, et al. Antibody-mediated rejection criteria – an addition to the Banff 97 classification of renal allograft rejection. Am J Transplant 2003;3:708-14.
8. Nickeleit V, Vamvakas EC, Pascual M, Poletti BJ, Colvin RB. The prognostic significance of specific arterial lesions in acute renal allograft rejection. J Am Soc Nephrol 1998; 9:1301-8.
9. Halloran PF, Schlaut J, Solez K, Srinivasa NS. The significance of the anti-class I response II. Clinical and pathologic features of renal transplants with anti-class I-like antibody. Transplant 1992;53:550-5.
10. Vongwiwatana A, Tasanarong A, HIdalgo LG, Halloran PF. The role of B cells and alloantibody in the host response to human organ allograft. Immunol Rev 2003;196:197-218.
11. Böhmig G, Exner M, Habicht A, et al. Capillary C4d deposition in kidney allografts: A specific marker of alloantibody-dependent graft injury. J Am Soc Nephrol 2002;13:1091-9.
12. Nickeleit V, Mihatsch MJ. Kidney transplants, antibodies and rejection: is C4d a magic marker? Nephrol Dial Transplant 2003;18:2232-9.
13. Ahern AT, Artruc AB, Della Pelle P, et al. Hyperacute rejection of HLA A-B identical renal allografts associated with B lymphocytes and endothelial reactive antibodies. Transplant 1982;33:103-5.
14. Rose M. Role of MHC and non-MHC antibodies in graft rejection. Curr Opin Organ Transplant 2004;9:16-23.
15. Praprotnik S, Blank M, Meroni PL, Rozman B, Eldor A, Shoenfeld Y. Classification of anti-endothelial cell antibodies into antibodies against microvascular and macrovascular endothelial cells. Arthritis and Rheum 2001;44:1484-94.
16. Yard B, Spruyt-Gerritse M, Claas F, et al. The clinical significance of allospecific antibodies against endothelial cells detected with an antibody-dependent cellular cytotoxicity assay for vascular rejection and graft loss after renal transplantation. Transplant 1993;55:1287-93.
17. Lunardi C, Bason C, Navone R, et al. Systemic sclerosis immunoglobulin G antibodies bind the human cytomegalovirus late protein UL94 and induce apoptosis in human endothelial cells. Nature Med 2000;6:1183-6.
18. Bas Bernardet S, Hourmant M, Coupel S, et al. Non-HLA type endothelial cell reactive alloantibodies in pretransplant sera of kidney recipients trigger apoptosis. Am J Transplant 2003;3:167-77.
19. Dragun D, Müller DN, Bräsen JH, et al. Angiotensin II type 1-receptor activating antibodies in renal allograft rejection. N Engl J Med 2005;352:558-69.
20. Wallukat G, Homuth V, Fischer T, et al. Patients with preeclampsia develop agonistic antibodies against the angiotensin AT1 receptor. J Clin Invest 1999;1103:945-52.
21. Fu MI, Herlitz H, Schulze W, et al. Autoantibodies against the angiotensin receptor (AT1) in patients with hypertension. J Hypertens 2000;18:945-53.
22. Mellor AL, Munn DH. Immunology at the maternal-fetal interface: Lessons for T-cell tolerance and suppression. Annu Rev Immunol 2000;18:367-91.
23. Nickeleit V, Andreoni K. The classification and treatment of antibody mediated rejection: Where do we stand? Kidney Int 2007;71:7-11.
24. Slowinski T, Suker D, Schönemann C, et al. 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 [abstract].
25. Heinze G, Mitterbauer C, Regele H, et al. Angiotensin-converting enzyme inhibitor or angiotensin II type 1 receptor antagonist therapy is associated with prolonged patient and graft survival after renal transplantation. J Am Soc Nephrol 2006;17:889-99.
26. Weidanz JA, Jacobson LM, Muehrer RJ, et al. AT_1R blockade reduces IFN-γ production in lymphocytes *in vivo* and *in vitro*. Kidney Int 2005;67:2134-42.
27. Bond RA, Ijzerman AP. Recent developments in constitutive receptor activity and inverse agonism, and their potential for GPCR drug discovery. Trends Pharmacol Sci 2006;27:92-6.
28. Scornik JC, Guerra G, Schold JD, Srinivas TR, Dragun D, Meier-Kriesche HU. Value of the posttransplant antibody tests in the evaluation of patients with renal allograft dysfunction. Am J Transplant 2007 [in press].
29. Touyz R. Molecular and cellular mechanisms in vascular injury and hypertension: role of angiotensin II. Curr Opin Nephrol Hypert 2005;14:125-31.
30. Kenakin TP. Recombinant roulette versus the apparent virtues of "natural" receptor systems: receptor genotypes vs. phenotypes. Trends Pharmacol Sci 2002;23:403-4.
31. Jurewicz M, McDermott DH, Sechler JM, et al. Human T and natural killer cells posses a functional renin-angiotensin system: Further mechanism of angiotensin II-induced inflammation. J Am Soc Nephrol 2007;18:1093-102.
32. Dobado Berrios P, Lopez-Pedrera C, Velasco F, Cudrado MJ. The role of tissue factor in the antiphospholipid syndrome. Arthritis Rhem 2001;44:2467-76.
33. Yamani MH, Cook DJ, Tuzcu EM, et al. Systemic up-regulation of angiotensin II type 1 receptor in cardiac donors with spontaneous intracerebral hemorrhage. Am J Transplant 2004;4:1097-102.