Morphologic aspects of antibody-mediated rejection in renal allografts

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Abstract

Recent studies have illustrated the important role of alloantibodies in mediating renal allograft rejection. Compared to T-cell mediated-rejection, antibody-mediated rejection is usually more refractory to conventional anti-rejection treatment and more likely to contribute to allograft failure. Antibody-mediated rejection can occur in three principal forms: hyperacute, acute, and chronic active. This brief review aims to characterize the main pathological features attributed to antibody-mediated rejection including light microscopy manifestations, ultrastructural alterations, and immunopathologic markers.

Keywords antibody-mediated rejection; C4d; donor-specific antibody; transplant glomerulitis; peritubular capillaritis

Antibody-mediated rejection (AMR) is characterized by humoral allograft injury caused by antibodies targeted against donor alloantigens. AMR is a significant barrier for long-term kidney allograft survival and a considerable cause of allograft loss. Because of this important adverse impact, understanding the mechanisms and the pathologic manifestations of AMR has become a major focus of interest in the field of kidney transplantation over the last couple of decades.

The importance of humoral immunity in allograft prognosis has only been recently appreciated. In 1990, Halloran and colleagues noted that the detection of circulating donor-specific antibodies (DSA) was associated with severe rejection characterized by pronounced microcirculation injury, high serum creatinine values, and increased risk of allograft failure.[1] Shortly afterwards, Feucht et al. found that C4d deposition along the peritubular capillaries is often associated with poor allograft prognosis.[2] Later on, Dr. Robert Colvin's group has uncovered the significant association between C4d staining and the presence of DSA.[3]

It has been postulated that AMR process starts with the presence of circulating DSA which typically activate complement leading to C4d deposition in the tissue, tissue damage, graft dysfunction, and ultimately graft failure if not treated properly.[4] Therefore, the three cardinal features of AMR include the serologic evidence of DSA, the immunohistochemical evidence of C4d staining along the peritubular capillaries, and the morphologic evidence of tissue injury (Table 1).[5]

Grossly, in acute AMR, the kidneys are swollen, and, in severe cases, they appear congested and may show foci of haemorrhage or even cortical necrosis with mottled appearance of the affected parenchyma. In contrast, chronically rejected kidneys are usually small and show granular surface. Urinalysis often reveals some degree of proteinuria. In some cases, red blood cells in the urine can also be appreciated reflecting destruction of microcirculation and/or vascular injury.

C4d is a cleavage product of C4 that covalently binds to tissues at the site of C4 activation and therefore can be detected for a relatively long period of time following the activation of complement cascade via classical or lectin pathways. C4d is assessed in the peritubular capillaries using either the immunofluorescence or immunoperoxidase techniques, and it is marked as diffusely or focally positive when present in >50% or 10–50% of the tissue surface, respectively (Figure 1a–b). In addition to C4d, tissue microarray studies demonstrate upregulation of several transcripts associated with endothelial activation and NK cells.[6,7] Small preliminary studies have also suggested that cytokine levels in the peripheral blood could potentially predict acute AMR during episodes of renal dysfunction.[8] The evolution of our understanding of AMR in renal allografts is best illustrated by the changes observed in the internationally used Banff classification schema for renal allograft pathology over time; while the nomenclature of T-cell-mediated rejection (TCMR) has remained largely unmodified, the classification of AMR underwent several revisions and additions.[5,9,10] In the original version of the Banff schema, AMR included two types – immediate (hyperacute) and delayed (accelerated acute).[9] The most recent version of the Banff classification schema includes three principle AMR subcategories – the C4d deposition without morphologic evidence of active rejection, acute AMR, and chronic active AMR.[11]In this manuscript, we review the morphologic aspects of these different forms of AMR in kidney allografts.

Hyperacute AMR

Although hyperacute AMR has been practically eliminated by the current protocols and pre-transplant serologic screening and work-up, it is important to review the pathologic features of this most aggressive type of AMR. This form of rejection is associated with pre-existing DSA and it is characterized by the development of severe cyanosis of the graft, often observed in the operating room within minutes following completion of vascular anastomosis. On light microscopy, the major findings include interstitial oedema, neutrophil and platelet aggregates admixed with fibrin deposits and/or microthrombi in glomerular and peritubular capillaries, fibrinoid necrosis of the vessel walls and/or glomerular tufts, and acute tubular necrosis. C4d staining is typically detected in the peritubular capillaries. The extent of cortical necrosis present in the tissue and the overall histopathologic findings largely depend on the interval time between the transplantation and the tissue sampling. Most of the cases result in immediate graft loss requiring surgical removal.[12]

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Diagnostic features of AMR; if at least one finding in each of the three cardinal features is present, the diagnostic criteria for AMR are met, and if two out of three cardinal criteria are met, the findings can be considered "suspicious for AMR"

Cardinal features	Acute AMR	Chronic active AMR
Morphologic evidence	Peritubular capillaritis and/or glomerulitis, with neutrophils or mononuclear cells; glomerular or vascular microthrombi; arterial fibrinoid necrosis; acute tubular injury	Glomerular basement membrane reduplication, peritubular capillary basement membrane multilayering, interstitial fibrosis and tubular atrophy, and/or fibrous intimal thickening of the arterial walls
Immunohistologic evidence	C4d staining by immunofluorescence or immunoperoxidase techniques along the peritubular capillaries	
Serologic evidence	Circulating donor-specific antibodies	

Table 1

Acute AMR

Both hyperacute and acute AMR are characterized by the presence of pan-capillary endothelial cell injury, mediated by DSA. Unlike the hyperacute rejection, acute AMR occurs days, weeks, or sometimes even years post-transplantation.[5] In acute AMR, endothelial cell injury is characterized by cell swelling, degenerative changes, loss of endothelial cell fenestrations, subendothelial widening of the glomerular capillary walls by accumulation of fluffy electron lucent material, and aggregates of platelets and fibrin thrombi in the capillaries. Acute AMR can manifest as glomerulitis and peritubular capillaritis with marginated neutrophils and/or mononuclear leukocytes, vascular fibrinoid necrosis and/or thrombosis, acute tubular injury, and in very severe cases, cortical necrosis (Figure 2a–b).[1] None of these changes are specific for AMR; many other forms of a primary vascular injury and procoagulant states can also result in similar changes when affecting the kidney, including scleroderma renal crisis, anti-phospholipid antibody syndrome, typical or atypical haemolytic-uraemic syndrome, and other forms of acute thrombotic microangiopathies, including the toxic endothelial injury related to immunosuppressant drug regimen.[13] Peritubular capillaritis and glomerulitis can also be encountered in other conditions such as pure TCMR, interstitial nephritis, and glomerulonephritis, although the combination of both is highly suggestive of AMR. Therefore, the morphologic features alone are not sufficient to diagnose acute AMR and the demonstration of both DSA as well as the immunologic evidence of tissue injury by antibodies (e.g., complement fixation and C4d deposits) are necessary to establish such diagnosis.

Three grades of acute AMR are recognized by the Banff classification schema; (I) acute tubular necrosis with minimal inflammation, (II) capillary margination and/or thromboses (peritubular capillaritis/glomerulitis), and (III) arterial transmural inflammation and fibrinoid necrosis.[5] These three grades are based on morphologic features in kidney samples meeting criteria of positive C4d staining in the peritubular capillaries and the detection of circulating DSA. Interstitial inflammation and tubulitis are not features of AMR, but are important findings of TCMR. Not infrequently, TCMR and AMR may coexist.[5,9,11]

Chronic active AMR

Chronic active form of AMR was introduced to the Banff classification schema at the 2005 meeting.[10] The morphologic features of chronic active AMR result from an ongoing and repetitive endothelial cell injury which leads to chronic changes manifested as glomerular basement membrane reduplication, peritubular capillary basement membrane multilayering (PTCBMML), arterial fibrointimal thickening, and interstitial fibrosis and tubular atrophy (Table 1).[10] The glomerular changes with endothelial cell injury and a membranoproliferative pattern in the absence of immune type or other deposits are known as chronic transplant

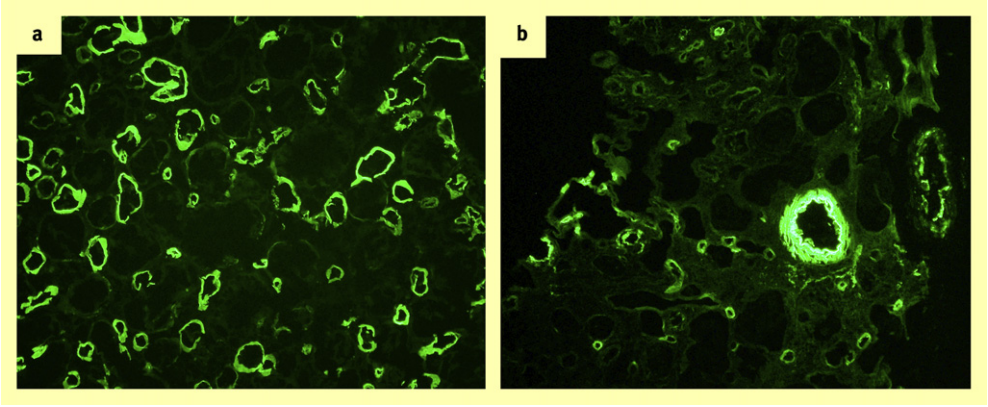

Figure 1 Diffuse (**a**) versus focal (**b**) C4d staining along peritubular capillaries (monoclonal C4d staining by immunofluorescence technique). Note the linear to very finely granular pattern of staining. In contrast to peritubular capillaries, arterial staining for C4d is considered non-specific (**b**).