Mineralocorticoid Receptor Antagonism: A Promising Therapeutic Approach to Treat Ischemic AKI

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Key Words

Acute renal injury · Aldosterone · Endothelial nitric oxide synthase · Endothelin 1 · Nitric oxide

Abstract

Acute kidney injury (AKI) is a common complication in hospitalized patients. One of the leading causes of AKI is renal ischemia/reperfusion (IR). In spite of all the progress made in acquiring knowledge about the mechanisms involved in AKI, no pharmacologic approach has yet become successful in clinical trials. Recent evidence suggests that mineralocorticoid receptor (MR) antagonism may be a useful strategy to prevent or treat AKI induced by IR. Here, we summarize the experimental work that supports MR antagonism as a potential approach to treat this disease. We also review the evidence that identifies a critical mechanism participating in the sustained vasoconstriction during kidney IR and uncovers that this mechanism is targeted by MR antagonists, thus explaining their beneficial effects.

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Acute kidney injury (AKI) is a frequent complication in hospitalized patients with higher incidence rates in intensive care unit patients. It is associated with unfavorable outcomes such as increased short- and long-term mortality rates, longer hospital stay, cardiovascular complications and chronic kidney disease (CKD) development [1]. Since many clinical situations can lead to a partial or complete reduction in the renal blood flow, an important cause of AKI is renal ischemia/reperfusion (IR) [2]. Ischemic injury is a complex pathophysiological entity that involves several alterations in the kidney. The reduction in the oxygen delivery leads to endothelial cell death, capillary loss, increase in capillary permeability, tubular cell death and to the activation of the surviving endothelial cells. Especially, direct injury in S2 and S3 segment of the proximal tubule epithelium due to reduced oxygen delivery and reactive oxygen species (ROS) generation during reperfusion take place [3]. In addition, an imbalance in vasoactive factors occurs and favors an increase in vasoconstriction, thus maintaining and prolonging the initial reduction in the renal blood flow [4]. All these alterations facilitate the initiation of an inflammatory process that will propagate the initial injury [4]. Unfortunately, even though the knowledge of AKI mechanisms is increasing, there is no effective pharmacological approach to treat or prevent AKI that has been successful in clinical trials.

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MR Antagonism Protects against Ischemic AKI in Rodents

During the past decade, evidence indicating that mineralocorticoid receptor (MR) antagonism (MRA) may be a useful strategy to protect against AKI has been accumulating. In 2007, a study by Mejía-Vilet et al. [5] showed that in a rat model of bilateral renal IR, the prophylactic administration of spironolactone (Spiro) prevented the acute renal dysfunction and tubular injury induced by IR. Spiro beneficial effect was associated with the preservation of a normal renal blood flow and reduction in oxidative stress. In this study, we suggested that the prevention of the hypo-perfusion induced by IR was mediated by acting on the eNOS/NO pathway, since the levels of urinary nitrates and nitrites were restored in Spiro treated rats. Moreover, this group presented increased eNOS expression and reduced phosphorylation in the Threonine 495 of eNOS, a key residue that when it is phosphorylated, it prevents eNOS from producing NO. In a subsequent study by Sánchez-Pozos et al. [6], it was demonstrated that Spiro is also able to treat kidney IR injury efficiently when administered immediately or up to 3 h after reperfusion. The benefit of Spiro at 6 h after reperfusion was partial and was lost when administered after longer periods of time. The renal hypo-perfusion was also prevented by Spiro treatment, thus suggesting a key role for MR activation in mediating vasoconstriction during renal IR. This study reported that renal vasoconstriction was associated with a reduction in the expression of the endothelin (ET)-B receptor, which mediates vasodilator effects in endothelial cells, and an increase in ET-A receptor, that mainly mediates vasoconstrictor effects. This imbalance in ET-A/ET-B receptor expression was not observed in rats receiving Spiro, thus gaining some insights into the possible mechanisms responsible for the beneficial effects of MR inhibition during kidney IR.

Aldosterone as a Key Mediator of AKI

To further explore if activation of MR by aldosterone is responsible for the deleterious effects observed during IR, we studied rats that underwent adrenalectomy and dexamethasone supplementation and were subjected to renal IR. These rats were efficiently protected from all the alterations induced by the ischemic insult in a similar way as Spiro administration does, leading to the conclusion that aldosterone plays a key role in the vasoconstriction and therefore in the deleterious effects of renal IR [7].

Non-Steroidal MRA Protects against AKI

Although the blockade of MR seems to be a promising therapeutic approach to treat AKI, the use of Spiro in renal ischemia is restricted due to its potassium-sparing effects, especially in patients with compromised renal function. In this sense, it is important to mention about the recent development of novel non-steroidal MR antagonists such as BR-4628 or finerenone that retain the beneficial effects of previously existing MR antagonists at a lower administration dose and with a lower risk of hyperkalemia [8, 9]. Indeed, finerenone has reduced the deleterious effects on plasma potassium levels as compared to Spiro in patients with heart failure and mild renal disease in spite of similar benefits in B-type natriuretic peptide levels [10].

In a recent study [11], we tested the efficacy of a novel, non-steroidal MRA, BR-4628, to protect against renal injury induced by IR. We showed that BR-4628 administration prevents and treats renal dysfunction, tubular injury and oxidative stress induced by IR. In order to provide new insights into the mechanism of kidney protection conferred by MRA, we studied the effects of renal IR and MR inhibition on the ET-1/ET-A/ET-B receptor pathway. We identified a critical mechanism participating in the sustained vasoconstriction during kidney IR and provide evidence that this mechanism is targeted by MRA, thus explaining the beneficial effects of MR antagonists. We demonstrated that the protective effect of BR-4628 on renal function, hypo-perfusion and tubular injury was lost when a selective ET-B antagonist was co-administered, pointing out the importance of the ET-B receptor in mediating the protection conferred by MR antagonists. It was previously reported that in pulmonary endothelial cells, aldosterone induces an increase in ROS generation that in turn allows a sulfenic modification of the Cys-405 residue in the ET-B receptor avoiding the binding to heterotrimeric G-protein beta-gamma-subunits, which signal to AKT [12]. Therefore, AKT reduction results in decreased eNOS activating phosphorylation. We therefore analyzed if this modification also happened in the kidney during IR. We showed for the first time in vivo and confirmed that in the ischemic kidney, the ET-B receptor undergoes a sulfenic acid modification of a critical cysteine residue, decreasing eNOS activation and affecting renal hemodynamics. The post-translational modification of the ET-B receptor and its consequences on eNOS signaling and NO production were efficiently prevented by MRA, suggesting that sulfenic modification of the Cys-405 residue in the ET-B occurs as a consequence of in-

Fig. 1. Mechanisms contributing to sustained vasoconstriction after renal IR. Renal IR increases aldosterone and ET-1 levels. Activation of MR by aldosterone may promote an augmentation in the expression of ET-A receptor (mediates ET-1 induced vasoconstriction) and ROS; effects blunted by MRA. ROS generation in turn induces a sulfenic acid modification in ET-B receptor, which accounts for the reduced activation of eNOS and NO production. The reduction in the bioavailability of NO, together with the increased expression of ET-A receptor shifts the balance toward vasoconstriction. The sustained vasoconstriction induces hypoxia in the kidney tissue leading to tubular injury. Injured tubules may go through maladaptive repair and produce CKD development.

creased oxygen reactive species after IR. The fact that ET-B receptor is inactivated during kidney IR may promote vasoconstriction by 2 different pathways: first, by a reduction of eNOS activation due to ET-B receptor sulfenic modification; second by shifting the balance to ET-1 mediated vasoconstriction by ET-A receptor (fig. 1). As commented by Juncos and Juncos [13], the fact that MRA acts through different mechanisms in order to prevent vasoconstriction makes MR a target that can be effective in treating AKI.

MRA Prevents the Transition from AKI to CKD

MRA holds promise not only to prevent the acute consequences of IR but also to avoid the progression to CKD induced by renal IR. Previously we showed [14] that a single episode of bilateral renal IR in rats induced CKD after a 9-month follow-up. The main alterations observed were renal dysfunction, proteinuria, extensive tubular dilation, glomerular hypertrophy and sclerosis and interstitial fibrosis development. Importantly, prophylactic MR inhibition for a few days before AKI was able to prevent the development of all these alterations.

Spirolactone Reduces Oxidative Stress in Kidney Transplantation

In support of all these experimental evidence showing a beneficial effect of MRA in ischemic kidney injury, we performed a pilot study to analyze the effect of Spiro administration in preventing adverse outcomes after living-kidney transplantation. Renal function was already well preserved in these patients receiving living-donor grafts, but it was not improved compared with untreated renal transplant patients. Interestingly, the patients receiving Spiro presented lesser levels of urinary hydrogen peroxide, a marker of oxidative stress [15]. Whether MRA may be beneficial in patients transplanted with grafts with expanded criteria that are more susceptible to IR injury and delayed graft function will be tested in the ongoing EPURE TRANSPLANT clinical trial (NCT02490904).

In conclusion, MRA, especially with novel non-steroidal antagonists, is a promising therapeutic strategy to prevent or limit kidney injury induced by IR both in acute setting and in AKI-mediated CKD. The current evidence calls for future research to test MRA efficiency in patients at high risk of developing AKI such as those in intensive care units, kidney transplant and cardiac surgery patients.

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