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Review Articles

The Expression of Neuronal Nitric Oxide Synthase in the Kidney and Its Role for Renal Functions

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The present work reviews the distribution of neuronal nitric oxide synthase in the kidney and its role in various renal functions. A review of the literature shows that most authors have focused on the endothelial isoform as the primary source of nitric oxide production in kidneys. Nowadays, there is convincing evidence that renal neuronal nitric oxide synthase plays a role in the control of the regulatory mechanisms of the kidney such as tubuloglomerular feedback mechanism and pressure natriuresis. It seems that the increased immunoreactivity of neuronal nitric oxide synthase in both renal cortex and medulla has renoprotective role under hypertension and weakens the development of target organ damage.

Key words: neuronal nitric oxide synthase (nNOS), kidney, spontaneously hypertensive rats (SHR), Wistar rats

Nitric oxide (NO) is a signaling molecule, which takes part in the regulation of various renal functions – maintenance of glomerular and medullary hemodynamics, sodium homeostasis, renin production, influence on the renal sympathetic activity. It is synthesized by the three isoforms of the enzyme nitric oxide synthase (NOS), which are all expressed in the kidney [7, 13]. Endothelial NOS (eNOS) is found mainly in the intrarenal vascular endothelium, the cells of the collecting ducts and distal tubular segments [11], while inducible NOS (iNOS) is usually expressed under pathological conditions [5]. There is convincing evidence that neuronal nitric oxide synthase (nNOS) has a primary role for the maintenance of blood pressure and vascular tone. This isoform is mainly expressed in macula densa cells, cells of the thick ascending limb of the loop of Henle, Bowman's capsule, cells of the collecting ducts and efferent arterioles [11]. Indeed, nNOS is responsible for various regulatory functions depending on its localization in the renal structure. For example, in the inner medulla, nNOS participates in the longterm regulation of blood pressure – the selective inhibition of the isoform in rats

on high salt diet results in arterial hypertension [12]. On the other hand, macula densa-derived NO is associated with regulation of the tubuloglomerular feedback (TGF) and renin production [11, 12]. Several hypotheses suggest the effects of macula densa-derived NO – 1. it influences macula densa cells and acts as an autacoid; 2. NO affects the juxtaglomerular mesangial cells; 3. NO reaches the afferent arteriole via diffusion process and induces vasodilatation [17]. The final effect of macula densa-derived NO is to attenuate TGF. Many hormones and molecules such as prostaglandins and angiotensin II influence the TGF sensitivity. For example, angiotensin II enhances TGF, while the atrial natriuretic peptide attenuates this autoregulatory mechanism [17]. It seems that the salt intake, intracellular pH, angiotensin II and the protein inhibitor of nNOS are important regulatory factors for nNOS activity and renin production [18, 21]. The influence of salt intake on nNOS expression in macula densa cells shows contradictory results. Some studies have shown increased nNOS activity in rats on low salt diet [20]. On the other hand, an increased NO production has been described in macula densa cells on high salt intake [23]. The activity of nNOS depends on the intracellular pH and is most prominent at pH 8 [22]. The protein inhibitor of nNOS is mainly expressed in the endothelial cells of the glomerular capillary tufts, the cells of the collecting ducts, but it is not found in macula densa cells [18].

The decrease of average blood pressure together with the increased renal blood flow and glomerular filtration rate during pregnancy are usually associated with higher levels of NO. The observed hemodynamic changes can be explained by increased activity of nNOS [2]. Undoubtedly, this isoform is important for normal pregnancy, but the mechanisms that affect the renal hemodynamics are still misunderstood. The short-term use of a selective inhibitor of nNOS in non-pregnant rats doesn't cause significant changes in the renal hemodynamics, while in pregnant rats it is accompanied by a 25% decrease in glomerular filtration rate and 60% increase in intrarenal vascular resistance [1]. In addition, the short-term selective inhibition of nNOS doesn't lead to changes in average blood pressure, while chronic inhibition provokes arterial hypertension [4].

The altered NO production may have a crucial role in the development of renal morphological changes in various pathological conditions that lead to end stage kidney disease. Many studies have demonstrated reduced plasma NO levels in patients with chronic kidney disease compared to control groups. The mechanism for the decreased production of NO is still poorly understood, but factors such as decreased l-arginine levels and accumulation of NOS inhibitors have been discussed [16].

Over the years, most researches have focused on the role of eNOS in the maintenance of blood pressure and the regulation of various renal functions. In addition, the altered production of NO is usually associated with changes in systemic blood pressure and the deficiency of NO is a possible mechanism for the development of arterial hypertension [8]. The application of NOS inhibitors in healthy rats causes hypertension and chronic kidney damage [3]. Some authors have studied the influence of the selective inhibition of nNOS by 7-nitroindazole (7-NI) on blood pressure, glomerular filtration rate and TGF. The results show that the acute application of 7-NI doesn't affect blood pressure, but it increases TGF. An elevated blood pressure and intensive TGF are observed in the treated rats after one week. The chronic inhibiton of nNOS for four weeks is associated with pronounced hypertension in the experimental group rats compared to control animals. This data indicates that the altered production of NO by nNOS may have a significant role in the development of arterial hypertension [15]. There is limited and contradictory information regarding the changes in the expression of renal nNOS under hypertensive conditions. The spontaneously hypertensive rat (SHR) is a good experimental model for demonstration of the consequences of prolonged untreated essential hypertension [24]. In this strain, the primarily enhanced TGF has been discussed

in the pathogenesis of elevated blood pressure. The main function of the macula densa-derived NO is to attenuate this autoregulatory mechanism and the changes in the expression of nNOS may play a crucial role in the progress of hypertension in SHR. Some authors have established a higher activity of the constitutive isoforms of renal NOS – eNOS and nNOS in SHR compared to Wistar rats [9, 14]. Some studies have described higher expression of nNOS in macula densa cells together with NADPH-oxidase. The increased activity of NADPH-oxidase is associated with production of superoxide ions, which further react with NO and decrease its bioavailability. The authors conclude that nNOS expression is closely associated with the balance between NO and superoxide ions [21]. In addition,the reduction of superoxide ions leads to vasodilatation of the afferent arterioles in SHR, while such effect is not observed in Wistar rats [10].

Fernandez et al. [6] have studied the expression of nNOS in age-matched Wistar rats and SHR. It was found that the immunoreactivity of nNOS is high positive in the inner papillary region and lacking in the outer medulla in the normotensive group. In the renal cortex, the isoform was mainly found in macula densa cells, as well as the parietal and visceral layers of Bowman's capsule. In SHR, the inner medulla showed a higher immunoreactivity and positive expression of the isoform was found in the outer medulla [6]. Our previous study also demonstrated a higher expression of nNOS in the renal medulla and the structural elements of the renal corpuscles in SHR compared to normotensive Wistar rats [19]. The increased expression of NOS in both renal cortex and medulla is associated with compensatory changes in TGF and pressure natriuresis [11]. These findings suggest that nNOS plays a renoprotective role in arterial hypertension and influences the development of hypertensive structural alterations such as glomerulosclerosis, and tubulointerstitial fibrosis, described by the term hypertensive nephrosclerosis.

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