A New Perspective for the Treatment of Renal Diseases?

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Asymmetric dimethylarginine (ADMA) is the most important endogenous inhibitor of nitric oxide synthase (NOS). It is an established uremic toxin (1,2) and one of the most intriguing risk markers for both cardiovascular (CV) complications (3) and renal disease progression (4). Accumulating evidence suggests that ADMA not only signals the risk of disease but that it is also causally implicated in the generation and/or progression of CV and kidney damage. Causal inference is a complex process (5) and suitable animal models as well as observational and experimental studies are needed for establishing causality. Importantly, the decisive proof of causality for risk factors related to human health is the healing or the improvement of the disease when the risk factor is eliminated or effectively countered, a proof still sorely lacking for ADMA in renal disease. In the last two years, a series of groundbreaking studies determined that methylarginine is a key determinant in animal models of CV and renal diseases, and as a result the interest in ADMA in the renal community is on the rise. The last paper of this series, by Matsumoto *et al.* (6), appears in this issue of *JASN* and provides perhaps decisive evidence on the causal role of ADMA in the progression of renal disease in the remnant kidney model (*i.e.*, the model that is currently considered as the most representative of chronic kidney disease in humans) (4). Highlighting the key components influenced by the experimental maneuver adopted in this study—namely ADMA and dimethylarginine dimethylaminohydrolase (DDAH)—is crucial for framing the novel information provided by this groundbreaking study (6).

ADMA: An Endogenous Inhibitor of NOS at the Crossroad of Cardiorenal Risk

ADMA and its enantiomer, symmetric dimethylarginine, were isolated in human urine by Kakimoto and Akazawa almost four decades ago (7). They speculated that both substances "may be important for the study of various pathologic states" (7). Twenty-two years later Vallance *et al.* provided initial evidence that the hypothesis of Kakimoto and Akazawa might be correct (8). On the basis of these seminal observations it was postulated that ADMA, by impairing NO synthesis, might contribute to the excess CV morbidity and mortality in CKD. ADMA levels are higher in dialysis patients with CV complications than in those without such complications (9). It correlates well with established clinical markers of CV burden such as intima-media thickness of the carotid artery (10) and left ventricular mass (11). There is also evidence that high ADMA concentration goes along with decreased number of circulating endothelial progenitor cells, thus impinging upon repair mechanisms (12). Furthermore, cohort studies both in the general population and in patients with CKD demonstrated a strong and independent link between ADMA, all-cause mortality, and CV events (13,14). Infusion of exogenous ADMA increases systemic vascular resistance (15,16), elevates mean arterial pressure (15,16), decreases heart rate (15,16), reduces cardiac output (15,16), and augments pulmonary vascular resistance (17) in men. In addition, ADMA administration impairs renal blood flow and sodium reabsorption in a dose-dependent manner (16). Exogenous ADMA also decreases cerebral perfusion and increases vascular stiffness (18). On the basis of the coherent evidence emerging in experimental and clinical studies, it is postulated that ADMA is not only a marker but also a potent mediator of endothelial dysfunction and atherosclerosis.

DDAH: The Key Step in the Regulation of ADMA Levels

DDAH is a key regulator of ADMA levels. Humans generate approximately 300 μmol ADMA (approximately 60 mg) per day (15). Only 20% of ADMA is excreted in the urine. The majority of ADMA, about 80%, is cleared by DDAH, which was first isolated from rat kidney. DDAH converts ADMA to citrulline and dimethylamine. Two isoforms of DDAH have been characterized and cloned: DDAH I is predominately found in tissues that express neuronal NOS, whereas DDAH II is predominately found in tissues expressing endothelial NOS (19). Colocalization of DDAH and NOS in various cell types including renal tubular cells supports the hypothesis that the intracellular concentration of ADMA is actively and cell-specifically regulated in NO-generating cells (20). By regulating ADMA

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levels, DDAH protects NOS activity. Such regulation is fundamental because NO, the final effector molecule of the system, is a most potent biologic vasodilator that inhibits monocyte adhesion, platelet aggregation, and vascular smooth muscle cell proliferation, which are all fundamental steps in the atherosclerosis process. Endothelial dysfunction as a result of reduced NO activity is an early step in the course of atherosclerotic vascular disease and evidence has accumulated that inhibition of NO synthesis by endogenous substances may be causally involved in this process.

The Dawn of a New Treatment for CKD?

CKD is now recognized as a major public health problem, as recently re-emphasized in a report by the Center for Diseases Control (21). Research on CKD is perceived as a priority because available treatments are still insufficient to effectively counter this epidemic. The hypothesis that lowering ADMA by enhancing DDAH activity ameliorates the progression of CKD was tested by Matsumoto *et al.* (6) using a rat model of 5/6 subtotal nephrectomy. Four weeks after subtotal nephrectomy the animals received either an adenoviral transfer of DDAH I or a control transfection. DDAH I gene transfer prevented further deterioration of renal function during the 2-wk follow-up. Moreover, increasing DDAH I activity alleviated tubulointerstitial fibrosis, which could at least in part be explained by a downregulation of TGF-β gene and protein expression. The positive effect of DDAH I gene transfer on morphologic changes was also reflected by the fact that DDAH I gene transfer prevented the increase in proteinuria. There are still many open questions. Why does overexpression of only one DDAH isoform have such profound biologic effects? Are there actions of DDAH beyond lowering ADMA? Are there adverse effects of a long-term increase in DDAH expression and/or activity? What is the importance of ADMA production/liberation in chronic renal disease? However, it is more important to point out that the experimental study goes along with observations in CKD and ESRD patients, as recently summarized (4). Furthermore, data by Matsumoto *et al.* (6) are in line with findings in DDAH transgenic mice, which exhibit low ADMA levels as a result of high DDAH I activity (22). These animals are less susceptible to vascular lesions (23), which is in part attributable to enhanced angiogenic potential (24), and they have a higher ability to regenerate endothelium (24). In contrast, knocking out this enzyme compromises ADMA degradation, which eventually results in inhibition of NO synthesis, increased vascular resistance and hypertension (25).

ADMA is increased early in the course of CKD, well before a reduction in GFR is measurable (26). Two recent studies have coherently shown that high ADMA predicts a faster rate of renal function loss (27,28). Therefore, lowering ADMA by increasing DDAH expression/activity might prevent progression in renal damage. Thus the study by Matsumoto *et al.* represents an important proof of concept that lowering ADMA may retard renal disease progression (6). Preclinical studies with drugs that lower ADMA by increasing DDAH transcription have already been published (29), which may open an entirely new opportunity for the treatment of CV and renal disease. Advancing risk factors research into the translational and clinical phases needs solid basic science, coherent experimental data, and epidemiologic and clinical observations. This is a multidimensional process that requires well-concerted efforts by a large number of research groups with different knowledge bases and expertise. Knowledge on ADMA has increased tremendously in recent years. Matsumoto *et al.* now provide a new and decisive piece of information for deciphering the role of ADMA in renal disease progression, and their study will certainly attract investigators and resources into this very promising research area (6).

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Disclosures

None.

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