GLUTATHIONE S-TRANSFERASES (GSTs) are multifunctional enzymes that are perhaps best known to participate in cell detoxification by catalyzing the conjugation of reduced glutathione to a wide variety of xenobiotics that have electrophilic centers. Production of these adducts promotes cytoprotection and further increases their subsequent cellular excretion (10). As understanding of these enzymes has broadened, it is now apparent that GSTs participate in S-glutathionylation reactions involved in a wide range of cellular functions that include the regeneration of S-thiolated proteins and catalysis in metabolic pathways not associated with detoxification (11).

GSTs critically contribute to the cellular defense against reactive oxygen species (ROS) (11). As a consequence of existing in an oxygen-rich environment, intracellular ROS, as well as reactive nitrogen species, are constantly produced. Their presence shifts the intracellular balance between oxidation and reduction; the subsequent changes in redox homeostasis alter cell function. GSTs are integrally involved in the metabolism of these reactive compounds and oxidized products. For example, GSTs catalyze the glutathionylation of peroxidized lipid products such as the toxic electrophile 4-hydroxynonenal (4-HNE) (9). GSTs also regulate signal transduction events activated by ROS and mediated through the JNK pathway (11). GSTs are regulated in part by nuclear factor-erythroid 2-related factor 2 (Nrf2), which stimulates transcription by binding to the cis-acting antioxidant response element (ARE) present in the promoter regions of GSTs (12).

Fig. 1. Production of 4-hydroxynonenal (4-HNE) by the per-oxidation of fatty acids allows for the oxidation of cysteine residues within Keap1. Cysteine oxidation changes the conformation of Keap1, permitting the release of Nrf2 into the cytosol. Unbound nuclear factor-erythroid 2-related factor 2 (Nrf2) is phosphorylated and translocates into the nucleus, where it binds to the cis-acting antioxidant response elements (ARE), leading to transcription/translation of GSTM1 and other antioxidant genes. GSTM1 inhibits the further production of 4-HNE and also directly glutathionylates 4-HNE, preventing oxidation of Keap1. A subsequent reduction in 4-HNE levels permits decreased production of reactive oxygen species (ROS) within the mitochondria. Thus GSTM1 potentially plays a central role in the prevention of a positive feedback loop involving 4-HNE and ROS, as well as providing feedback to inhibit further Nrf2 activation.

Several lines of evidence suggest that GSTs participate integrally in vascular biology and function. GSTM1(0), the GSTM1 null allele, is associated with a more rapid increase in common carotid intima media thickness in smokers (2). Reductions in GSTM1 levels in vascular smooth muscle cells (VSMC) in vitro result in the production of ROS and cellular proliferation (14). Rats subjected to acute hypotension demonstrate upregulation of GSTM1 in the kidney, potentially leading to an increased degradation of ROS (13). Reductions in
GSTM1 and an associated increase in oxidative stress have been documented in the kidneys of stroke-prone spontaneously hypertensive rats (7), although a follow-up study by the same investigators did not identify an association between single nucleotide polymorphisms in GSTM1 genes and hypertension in humans (3). Although changes in GST protein content have been shown to alter cellular redox homeostasis, the data regarding an association with hypertension and kidney injury in particular have not been definitive.

In an issue of the *American Journal of Physiology-Renal Physiology*, Chang et al. (1) show that clinical outcomes for study participants in the African American Study of Kidney Disease and Hypertension (AASK) trial were affected by the GSTM1 genotype. Participants who had hypertensive nephropathy and were homozygous or heterozygous for the GSTM1 null allele GSTM1(0) displayed accelerated disease progression by decreased time to the composite outcome of a glomerular filtration rate (GFR) event, dialysis, or death compared with those patients who were homozygous for the GSTM1 active allele. Additional studies showed that mice that had undergone 5/6 nephrectomy to generate a murine model of chronic kidney disease (CKD) developed an increase in Nrf2 and GSTM1 as well as an associated increase in 4-HNE adduct formation in the remnant kidney. Knockdown of GSTM1 in cultured VSMC increased intracellular levels of 4-HNE adducts, while overexpression of GSTM1 decreased 4-HNE adducts. Finally, incubation of VSMC with 4-HNE appeared to produce a dose-dependent increase in Nrf2 and GSTM1. These studies suggested that the GSTM1 genotype, the presence of CKD, and oxidative stress regulate the renovascular content of GSTM1.

Based upon these novel findings and previous literature, one can hypothesize that GSTM1 is normally upregulated in hypertensive patients with CKD as a protective response to increased oxidative stress. Conversely, reductions in GSTM1 levels, most notably from the common allelic variant GSTM1(0) lead to reduced protective capacity and accelerated kidney disease progression. This loss of kidney function may be due to an increase in ROS and subsequent lipid peroxidation in the kidney vasculature of hypertensive CKD patients. Increased levels of 4-HNE lead to an increase in Nrf2, as shown by this paper, and cytoplasmic translocation to the nucleus (5), most likely due to the disruption of key cysteine residues in Keap1, freeing Nrf2 from ubiquitination and subsequent proteolysis. Once free of the Keap1/Cul3 ubiquitination system, Nrf2 translocates from the cytoplasm to the nucleus and binds to the cis-acting ARE in the upstream promoter region of many antioxidant genes, including GSTM1, and initiates gene transcription (6) (Fig. 1). Increasing GSTM1 promotes a decrease in lipid peroxidation and 4-HNE levels, leading to a reduction in ROS production in VSMC and the associated functional consequences that include VSMC proliferation and migration. The hypothesis that hypertensive nephrosclerosis is due to oxidative stress in the vasculature awaits confirmation, but the present study adds intriguing new findings that merit additional work in this area.

**REFERENCES**


