EDITORIAL COMMENT

Nitroso-Redox Imbalance Affects Cardiac Structure and Function*

Vasileios Karantalis, MD,†
Ivonne Hernandez Schulman, MD,†‡
Joshua M. Hare, MD†

Miami, Florida

Cardiovascular diseases (CVDs) and their treatment pose a huge economic and social burden in Western societies and in the developing world (1). Thus, there is significant interest in developing new therapeutic targets, and it is of particular interest that an old class of drugs, inhibitors of xanthine oxidoreductase (XO), may be repositioned to treat heart failure and left ventricular (LV) dysfunction. In this context, there is growing awareness that nitroso-redox balance may represent a new therapeutic target for heart failure (2). However, whether XO inhibitors fully address nitroso-redox imbalance in all patients remains controversial.

The hypothesis that XO inhibition could treat heart failure was tested in the OPT-CHF (Oxypurinol Therapy for Congestive Heart Failure) trial (3). In that study, the primary endpoint was not statistically different in the population of symptomatic heart failure patients enrolled; however, patients with hyperuricemia appeared to be a responsive population. While hyperuricemia is classically associated with gout and nephropathy, it is clearly also a biomarker in heart failure populations (4–6). Uric acid (UA) is the end product of purine metabolism, involving the conversion of hypoxanthine to xanthine and then to UA in reactions catalyzed by XO (Fig. 1) (7). In most mammals including rodents, UA is further degraded to allantoin via the enzyme urate oxidase (UO), resulting in low UA serum levels. In humans and great apes, however, this final step does not occur because of a single point mutation inactivating UO, resulting in substantially higher serum UA levels. Whether UA plays beneficial or deleterious roles remains controversial, and some arguments can be made that the impact of UA on vascular tone has played an evolutionary role (4). However, in hypertension, type 2 diabetes mellitus, coronary artery disease (CAD), heart failure, and chronic kidney disease (CKD), high UA levels correlate with an increased risk of stroke and CVDs (4,6,8–10).

As UA levels represent a biomarker of XO activity, the OPT-CHF results suggest that patients with elevated XO activity responded preferentially (3). XO is a key oxidase participating in nitroso-redox imbalance in the heart, producing superoxide as a byproduct of purine metabolism (Fig. 1). Thus, the epidemiological association of high UA levels with worse prognosis in a wide cohort of patients with CVDs likely reflects the fact that UA levels rise with increased XO activity (4–10).
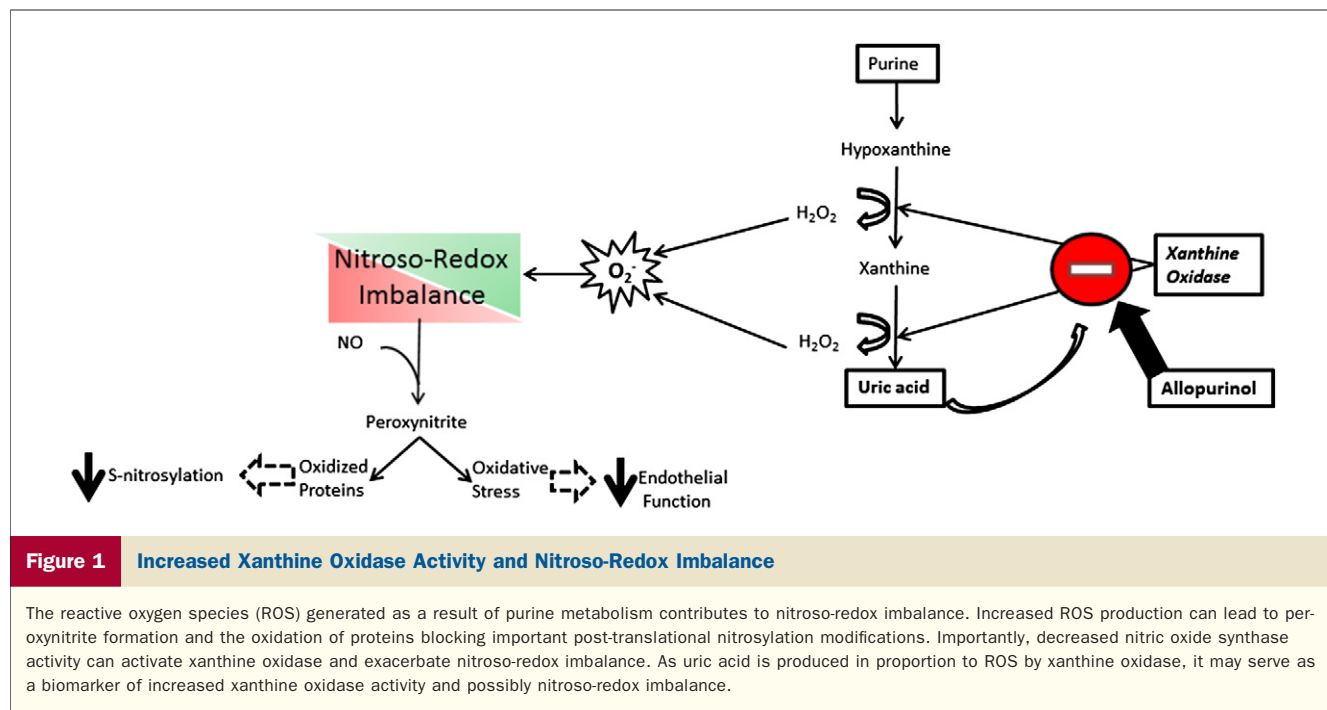
XO and Nitroso-Redox Imbalance

In the heart, XO localizes to the sarcoplasmic reticulum (SR), interacting with neuronal nitric oxide (NO) synthase (nNOS, NOS1) and the ryanodine receptor (RYR2) (11,12). Depressed NOS1 activity or abundance in the SR augments XO activity (12,13), elevating oxidative stress in the SR (14–16). The functional consequences of this imbalance are evident in patients with heart failure, where inhibition of XO with allopurinol improved myocardial efficiency (reversing mechanoenergetic uncoupling) (17,18). The mechanism of this effect is attributable, at least in part, to post-translational modification of thiol moieties on proteins (19,20). In the case of the SR, nitrosylation of the RYR2 modulates the open probability of this Ca^{2+} pore. With nitroso-redox imbalance, formation of reactive oxygen species predominates, and in turn also activates the RYR2, but in an irreversible manner, leading to Ca^{2+} leak (11,12,16).

This colocalization of an oxidase with a NO synthase and their signaling interactions underlies the basis for nitroso-redox imbalance as a key pathophysiologic signaling mechanism (2). In this regard, we and others have demonstrated that the NOS1-deficient mouse has increased mortality, LV remodeling, and ventricular arrhythmias after myocardial infarction, associated with increased XO activity and decreased S-nitrosylation of Ca^{2+} handling proteins (21–23). This phenotype is further substantiated in NOS1 overexpressing mice where specific myocardial NOS1 overexpression protects from remodeling and preserves Ca^{2+} cycling (24).

In the failing heart, NOS1 translocates from the SR to the cell membrane (25,26), an effect that is protective from Ca^{2+} overload. Increased NOS1 in sarcolemmal caveolae increases S-nitrosylation of the L-type calcium channel, leading to a decreased inward Ca^{2+} current. In turn, the reduced Ca^{2+} influx within the cardiac myocyte prevents

Figure 1 **Increased Xanthine Oxidase Activity and Nitroso-Redox Imbalance**

The reactive oxygen species (ROS) generated as a result of purine metabolism contributes to nitroso-redox imbalance. Increased ROS production can lead to per-oxynitrite formation and the oxidation of proteins blocking important post-translational nitrosylation modifications. Importantly, decreased nitric oxide synthase activity can activate xanthine oxidase and exacerbate nitroso-redox imbalance. As uric acid is produced in proportion to ROS by xanthine oxidase, it may serve as a biomarker of increased xanthine oxidase activity and possibly nitroso-redox imbalance.

Ca^{2+} overload-induced injury (23,27,28). While this mechanism is largely protective, probably acutely (29), it also results in SR nitroso-redox imbalance over the long term, which as described above can potentiate SR Ca^{2+} leak, arrhythmias, and contractile dysfunction. The disruption of physiological nitrosylation of SR proteins has been shown to be deleterious (19,20,30,31).

Cardiac Function and Structure

Struthers and collaborators have made a series of contributions regarding the role of XO inhibition and cardiovascular morbidity and mortality. They have shown that allopurinol in a dose-dependent fashion (32) decreases both cardiovascular events and mortality (33), improves endothelial function in patients with CAD (34), and regresses LV mass in patients with CKD (10). In the study published in this issue of the *Journal* (35), the authors further address a key issue, that of the regression of LV hypertrophy (LVH) in patients with CAD. Indeed, a key surrogate for clinical efficacy in disorders of LV dysfunction is the amelioration of LVH (36,37). They randomized 66 patients with CAD and LVH to receive either 600 mg/day allopurinol or placebo on a background of optimal medical therapy. LV mass was reduced by 5.2 ± 5.8 g for the high-dose allopurinol cohort versus 1.3 ± 4.48 g for the placebo group (p = 0.007). Moreover, allopurinol significantly reduced LV end-systolic volume, improved endothelial function, and reduced augmentation index, an independent predictor of LV mass regression (38). The beneficial effect of XO inhibition on LVH regression is supported by experimental animal models of heart failure (39). Furthermore, the importance of these findings are highlighted by fact that in the LIFE study

(40), LVH regression alone was associated with reduced all-cause mortality (by 28%), cardiovascular mortality (by 38%), sudden cardiac death (by 19%), myocardial infarction (by 15%), new heart failure (by 36%), new onset atrial fibrillation (by 12%), and stroke (by 24%).

The importance of this small single-center study lies in the fact that it adds further support for the development of XO inhibitors in the treatment of heart failure. As with angiotensin-converting enzyme inhibitors, beta-blockers, and the combination of isosorbide dinitrate and hydralazine, the regression of LVH augurs well for the possibility that XO inhibitors could have clinical benefits in an appropriately selected heart failure population. Thus, this work adds another piece of evidence supporting the development of therapeutic strategies targeting myocardial nitroso-redox imbalance.

Reprint requests and correspondence: Dr. Joshua M. Hare, Interdisciplinary Stem Cell Institute, University of Miami Miller School of Medicine, Biomedical Research Building, 1501 N.W. 10th Avenue, Room 824, P.O. Box 016960 (R125), Miami, Florida 33101. E-mail: jhare@med.miami.edu.

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