Simultaneous exposure to nitric oxide inhibition and angiotensin II overload: is it a murine model of mitochondrial dysfunction in nonischemic heart failure?

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CARDIAC ENERGY DEPRIVATION is the consequence of a complex alteration of all bioenergetic cellular components of the dysfunctional myocardium and is considered to be a critical contributor to the progressive development of heart failure (HF) of different origins (16). The study of bioenergetics of nonischemic systolic failing heart associated with elevated systemic blood pressure is a clinically relevant pathophysiological topic as it may aid in detecting biomarkers of early HF onset in patients with hypertension (17). In particular, this typical form of acute HF is characterized by severely reduced ejection fraction and better outcome (1, 21).

The progressive mitochondrial damage in hypertensive cardiomyopathy is likely due to high levels of angiotensin II (ANG II), a potent prooxidant and vasoconstrictor hormone, and deficiency of nitric oxide (NO), a soluble gas with vasodilatory properties. The loss of mitochondrial function plays a key role in leading the energy depletion up to the onset of severe decay of contractile function in hearts with coronary patency (2, 18), but translation of this knowledge to early prevent HF associated with chronic high blood pressure is far from straightforward.

This limitation is due to both limited clinically relevant animal models and the poor understanding of regulatory mechanisms activated by complex microenvironment. Considerable clinical evidence has recently demonstrated that cardiac bioenergetics is unaffected by oxygen supplementation in patients with nonischemic HF (9). Data from the abovementioned study (9) and others (19) strongly support the hypothesis that mechanonenergetics of hypertensive failing heart is more dependent on metabolic efficiency of mitochondrial apparatus rather than oxygen supply to stressed cardiomyocytes.

Hence, it is plausible to suppose that the simultaneous exposure of cardiomyocytes to the deficiency of NO production by an overactive NADPH oxidase system might early impair the mitochondrial efficiency even in the presence of an oxygenated tissue. Similar mechanisms have been identified in advanced stages of HF when diminished oxygen delivery impairs substrates metabolism and increases oxidative stress, which uncouples endothelial NO synthase leading to impaired reserve for mitochondrial oxidative flux (26).

Indeed, recent proteomics studies have demonstrated that the downregulation of different mitochondrial oxidative pathways characterizes the phenotype of nonischemic HF (4, 15). Further analysis of the interplay between mitochondrial function, active species production, and substrate metabolism is crucial for the understanding of the molecular basis of cardiomyocytes chronically stressed by hypertensive microenvironment and for revealing novel treatment strategies. For this purpose, there is a clear need for new animal models in this clinically important area.

In their article published in the American Journal of Physiology-Heart and Circulatory Physiology, Hamilton and co-workers (12) describe a original murine model of hypertensive HF induced by long-term treatment with N^G^-nitro-L-arginine methyl ester (L-NAME, 0.3 mg/ml with 1% NaCl po), a potent inhibitor of NO synthesis, and ANG II (0.7 mg·kg^{-1}·day^{-1} sc). Interestingly, the detection of elevated systolic blood pressure in cotreated mice with reduced ejection fraction is accompanied by larger amount of fibrosis and number of hypertrophic cardiomyocytes, higher degree of mitochondrial impairment, and oxidative stress compared with those parameters in hearts of mice single-treated with ANG II or L-NAME. However, it is unclear which of the two hits primarily impairs myocardial mitochondrial function up to onset of HF.

Studies in both animal models (23-24) and human HF (3) have indicated that reduced NO bioavailability contributes to progression of cardiac mechanonenergetics dysfunction. Chronic L-NAME seems to exacerbate the myocardial mitochondrial dysfunction due to ANG II-induced NO deficiency (12). Indeed, the ANG II-induced activation of NADPH oxidase increases reactive oxygen species production (10), which leads to the uncoupling of NO synthase, the NO neutralization by superoxide, and peroxynitrite formation (14). Taken together, the authors argue that simultaneous exposure of myocardium to both pathogenic factors is necessary to cause mitochondrial-based HF.

Despite similar ejection fraction, the systemic blood pressure and cardiac fibrosis in mice treated with ANG II is higher than those in mice treated with L-NAME. These data confirm that ANG II-induced oxidative stress promotes hypertensive mitochondrial injury (25). Unfortunately, the lack of control group treated with an alternative vasoconstrictive agent cannot exclude whether hypertension, per se, can determine mitochondrial damage.

To unravel the bioenergetics mechanisms, the authors have conducted a detailed in vitro characterisation of interfibillar mitochondria (IFM), which provide most of the energy for the contractile apparatus and subsarcolemmal (SSM) cardiac mitochondria (13). The function of SSMs was lower than cardiac IFM of L-NAME + ANG II-treated mice. Interestingly, similar dysfunction is detectable in patients with type 2 diabetes (8), who are simultaneously exposed to high ANG II-induced oxidative stress and NO deficiency (22).
Myocardial metabolic pathways regulating energy production, as oxidative phosphorylation, mitochondrial function, fatty acid oxidation, AMPK signaling, and tricarboxylic acid cycle, are more altered in t-NAME + ANG II-treated mice. Moreover, complexes I, II, IV and V are downregulated. Similarly, pathways regulating cardiac hypertrophy, hypoxia signaling, oxidative stress, and calcium signaling are strongly deregulated during combined treatment.

The animal model was previously designed by Oestreicher et al. (20), but there are still some clinically relevant limitations. In fact, we cannot exclude that mitochondria damage may occur in t-NAME + ANG II-treated mice with hypertension and preserved ejection fraction.

Even if typical hallmarks of myocardial stiffness (i.e., fibrosis, hypertrophy) are detectable in t-NAME + ANG II-treated mice, proper evaluation of diastolic function would help to identify the onset of diastolic failure at earlier stages. Interestingly, the magnitude of cardiac mitochondrial oxidative phosphorylation dysfunction in the proposed model is not detectable in human chronic heart failure (6). It is conceivable that 28 days of combined treatment are not enough to reproduce mitochondrial abnormalities of chronic heart failure in patients. Deeper multimodal validation of this model at long-term follow-up and in large animals (i.e., dogs, pigs) would be recommended.

Despite the above limitations, this model may improve our understanding on the complex role of mitochondrial dysfunction at early stages of HF and provide a new tool to test targeted therapeutic strategies.

REFERENCES


