

Boosting the Exhausted Vasculature—SIRT3 (to the) Rescue

Perturbations of metabolic pathways are an overarching theme spanning numerous pathological conditions and diseases, including pulmonary hypertension (PH). The majority of metabolic disturbances in PH can be linked with underlying mitochondrial dysfunction (1). Pulmonary artery cells from patients with PH have higher glucose uptake but lower oxidative glucose metabolism in the mitochondria (2–4). Patients with PH display higher lipid accumulation in the heart (5) and lower mitochondrial fatty acid oxidation in the pulmonary artery cells (4), similar to altered glucose handling. Although metabolic disturbances are unlikely to be the causative factor in the development of PH, they give diseased cells a competitive growth and survival advantage, such as the reported increase in glutaminolysis (6) and one-carbon or glycine metabolism in patients with PH (7). Ultimately, the mitochondrial dysfunction leads to more inefficient ATP production through glycolysis. This potential energy deficit is, in part, compensated through higher glucose uptake and supported by increased glutaminolysis. Additionally, increased glutaminolysis and one-carbon metabolism support increased demand for biochemical building blocks such as purine nucleosides in proliferating pulmonary artery cells. Finally, the shifting energy demands and metabolic pathways also affect cellular redox balance, with downregulation and diminished activity of mitochondrial superoxide dismutase (SOD2) being a hallmark of PH-associated oxidative stress (1). This vicious cycle centered on dysfunctional mitochondria can thus coordinately sustain an altered cellular phenotype associated with disease progression. It is interesting that these disturbances are preserved in cultured cells (8, 9) and cannot be reversed by fuel shunting back into the mitochondrial tricyclic acid cycle (10).

It is becoming clear that maintaining such a persistent disease phenotype requires more profound and deeper mechanisms that go beyond transcription factor-controlled gene expression changes. Epigenetic reprogramming was recently described as an underlying mechanism that orchestrates expression changes in human adventitial fibroblasts from patients with PH (11). In this issue of the *Journal*, Li and colleagues (pp. 570–583) link diminished mitochondrial function and increased proliferation of adventitial fibroblasts with increased acetylation of mitochondrial proteins and lower SIRT3 protein levels in PH samples (12). SIRT3 is a NAD⁺-dependent mitochondrial protein deacetylase that acts as a crucial regulator of mitochondrial function (13). Loss of SIRT3 is associated with decreased mitochondrial respiratory capacity and higher oxidative stress (13). SIRT3 regulates the activity of mitochondrial enzymes involved in the formation of the epigenetic modulators acetyl-coenzyme A and β -hydroxybutyrate (14). SIRT3 is, therefore, a central regulator, linking mitochondrial metabolic processes with cellular health, survival, and phenotypic characteristics.

Li and colleagues show lower SIRT3 expression in bovine PH lung samples and correspondingly higher acetylation of mitochondrial proteins, including SOD2. They further show lower SIRT3 expression in human and bovine PH-adventitial fibroblasts. Quantifying the acetylation of select mitochondrial proteins such as MPC1 and SOD2 as a surrogate measure of SIRT3 activity, the authors provide further evidence of diminished SIRT3 expression and activity levels in PH fibroblasts. Taking advantage of the distinctive fluorescent properties of endogenous metabolites, the authors applied a novel fluorescence lifetime imaging microscopy technique to subcellularly resolve metabolic activities and show diminished NAD⁺ levels in PH fibroblasts. They go on to show that genetic or pharmacological approaches that have been used to restore SIRT3 levels or boost its activity improve mitochondrial health and normalize the adventitial fibroblast phenotype. In particular, a combination of SIRT3 overexpression and NAD⁺ supplementation was the most effective in normalizing mitochondrial protein acetylation, increasing oxidative phosphorylation, and normalizing fibroblast proliferation and apoptosis rates.

This study puts another piece of the puzzle in place and establishes SIRT3 as a crucial novel therapeutic target for the treatment of PH. Studies by Paulin and colleagues have shown the beneficial effects of SIRT3 restoration in human pulmonary artery smooth muscle cells and animal models of PH (15). Li and colleagues extend the concept that modulation of SIRT3 levels and activity could be beneficial for patients with PH by investigations using adventitial fibroblasts. However, a study by Waypa and colleagues showed that the loss of SIRT3 does not exacerbate chronic hypoxia-induced PH (16). Nevertheless, on closer inspection of the underlying data, the authors show a clear trend in shortening of the normalized pulmonary artery acceleration time and increased arterial wall thickness in the hypoxic SIRT3-knockout mice compared with hypoxic wild-type mice, indicating a potential PH augmentation (16). It is interesting that the discordance was attributed possibly to mouse strain-specific differences in underlying metabolism. Although, in their current work, Li and colleagues did not address potential metabolic differences between different cell types and their effect on SIRT3-controlled processes, their study shows that reduced SIRT3 levels or diminished activity is a recurring feature in PH pathogenesis that is shared across multiple cell types.

Restoration of SIRT3 activity by boosting NAD⁺ levels was shown as a promising therapeutic approach in a cardiovascular disease setting. Oral supplementation with nicotinamide, an NAD⁺ precursor, normalized myocardial metabolism and function, lowered blood pressure, and improved mortality in animal models of heart failure (17). It is intriguing that certain exercise regimes were associated with tissue-specific increases in NAD⁺ levels (18).

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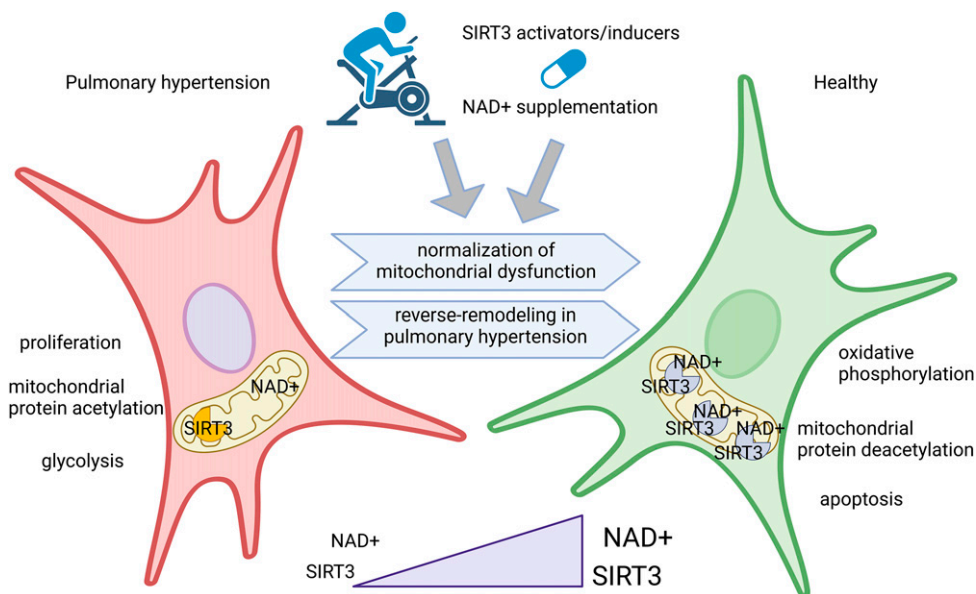


Figure 1. *SIRT3*-centered approach of rebalancing mitochondrial health as a novel reverse-remodeling strategy for pulmonary hypertension treatment. Nutritional supplementation with *SIRT3* cofactor NAD^+ , or with its precursor nicotinamide, or exercise regimes could be combined with pharmacological treatments that increase *SIRT3* expression levels. The synergistic effect of concomitant increase in enzyme and cofactor levels is shown in the present study to normalize major metabolic disturbances associated with pulmonary hypertension and reprogram the diseased cells toward a homeostatic phenotype. This illustration was created using BioRender.com.

A well-balanced and controlled level of physical activity through regular exercise was also shown to be beneficial for patients with PH (19).

The combined use of over-the-counter dietary supplements with personalized physical activity modalities could represent an innovative approach in the management of PH (Figure 1). This opens a window into potential cost-effective novel treatment strategies centered on *SIRT3*- NAD^+ . However, before wider use or uncontrolled self-medication, more research such as the study by Li and colleagues is needed to carefully and systematically investigate all aspects and long-term changes associated with mechanisms that target epigenetic reprogramming. ■

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Senka Ljubojevic-Holzer, Ph.D.
Division of Cardiology
and
Division of Molecular Biology
Medical University of Graz
Graz, Austria

Slaven Crnkovic, Ph.D.
Division of Physiology
Medical University of Graz
Graz, Austria

Institute for Lung Health
Giessen, Germany

Cardiopulmonary Institute
Giessen, Germany

and

Ludwig Boltzmann Institute for Lung Vascular Research
Graz, Austria

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