Heart of the Matter: Divergent Roles of Hypoxia-Inducible Factors in Hypoxia-induced Right Ventricle Hypertrophy

Since their discovery in the early 1990s, hypoxia-inducible factors (HIFs) have been cemented as fundamental regulators of organismal response to changes in oxygen. HIF family members, hypoxia-inducible factor 1 and 2 (HIF-1 and HIF-2, respectively), exist as heterodimers with a ubiquitous β subunit (HIF-1β) and oxygen-sensitive α subunit (HIF-1α or HIF-2α), the latter determining activity of the HIF-1 or HIF-2 transcriptional complex. HIF-1 and HIF-2 have been extensively studied and confirmed to transcriptionally activate hundreds of genes, with both overlapping and distinct targets. HIF signaling controls a diverse set of physiologic processes, including development, erythropoiesis, energy and metabolism, angiogenesis, and cell proliferation and migration. HIFs also participate in the pathogenesis of disease, protecting against peripheral artery disease, cardiac ischemia, and rejection following transplant and promoting cancer (1, 2).

Over the past two decades, numerous studies have explored the role of HIFs in pulmonary hypertension (PH), with sometimes conflicting results (3–13). Studies include global inhibition or deletion of HIFs or gain or loss of HIF function in the pulmonary circulation. In this issue of the *Journal*, Smith and colleagues (pp. 652–664) investigated the contribution of HIF-1α and HIF-2α expressed within cardiomyocytes to chronic hypoxia-induced PH (14). In an elegant and comprehensive set of studies, the authors used a series of transgenic mice with tamoxifen-inducible knockdown of HIF-1α and/or HIF-2α in cardiomyocytes or HIF-1α knockdown in smooth muscle and evaluated the hemodynamic response, right ventricle hypertrophy (RVH) and RNA-sequencing (RNA-seq) analysis in the heart tissue. The authors rigorously confirmed cell-specific knockdown of each HIF by crossing mice with a membrane targeted global reporter mouse (15). Several key differences in the response to chronic hypoxia were observed depending on the paralog or cell type, with nonuniform effects between RV systolic pressure (RVSP), RVH, and cardiac output. The most striking findings were that deletion of HIF-1α worsened RV remodeling but not RVSP and that RVH was prevented by simultaneous knockdown of HIF-2α. RNA-seq analysis highlighted distinct genes modulated in response to chronic hypoxia in each strain; HIF-1α knockdown increased pathways associated with hypertrophic remodeling, including G-protein coupled receptor and ion channel function, whereas HIF-2α knockdown predominantly modulated inflammatory and immune pathways.

Several interesting discussion points emerge from this work. Perhaps first and foremost is the striking importance of considering the role of specific HIFs, cell type, and type of injury. This study provides novel insight into RV responses to chronic hypoxia, demonstrating how systemic hypoxemia, through HIF signaling, can have a direct effect on the myocardium. In this setting, HIF-1α in cardiomyocytes appears protective. The role of HIF-1α in hypoxia-induced PH has been the subject of debate, with studies showing protective (10), detrimental (3, 13), or limited (8) roles. Factors proposed to contribute to differential results across studies have included sex, constitutive versus inducible and/or global versus cell-type–specific deletion, or timing of measurements. Interestingly, the current finding with respect to HIF-1α knockdown in cardiomyocytes is consistent with work from Kim and colleagues describing worsened PH in mice with constitutive loss of HIF-1α in smooth muscle driven by SM22-α promoter–linked Cre. Of note, SM22-α is also expressed in cardiomyocytes (12), raising the question as to whether these new results may also explain, in part, why SM22-α–driven deletion of HIF-1α increased RVH and RVSP. The RNA-seq data are also compelling. The implication of HIF-1α in the regulation of cardiomyocyte ion channels is consistent with early work demonstrating a role for HIF-1α in ion channel regulation (4), whereas the inflammatory and immune pathways regulated by HIF-2α are reminiscent of the recent work by Hu and colleagues showing a role for HIF-2α in lung macrophage activation (8). There is limited work on the inflammatory response within the RV in PH or cross-talk between different cardiac, vascular, and lung cells, and this is fertile ground for future research.

Although it provides new insights, this study does have some limitations worth discussing and raises additional questions. Notably, the results represent a snapshot in time, and analysis of other studies suggests that the role of HIFs in PH pathogenesis varies not only by cell type but also temporally. Measurements in this study were made at an intermediate time point (4 wk), and the genes altered, or effects on RVSP or RVH, might differ earlier or later in the disease process. RNA-seq was only performed on one cell type, and new methodologies with single-cell RNA-seq could provide further insight into effects of cardiomyocyte knockdown on other cardiac cells. It will be important to validate the effects of HIF signaling not just on RNA profiles but also on proteins and metabolites. Finally, it remains unclear whether RVH during chronic hypoxia, and thus the effect of HIF-1–induced repression of RVH, is beneficial (i.e., compensatory) or detrimental. For example, reduced RVH and RVSP are often interpreted as decreased PH but could also signal a lack of appropriate compensatory hypertrophy, maybe as a consequence of impaired angiogenesis. The exact mechanisms involved in RVH also require further elucidation. The enhanced RVH could be a direct result of hypoxia on intrinsic

Originally Published in Press as DOI: 10.1165/rcmb.2020-0325ED on August 13, 2020

cardiomyocyte signaling or hypoxia-induced cytokine production and reduced recruitment of immune cells by other cell types. These questions highlight the importance of studies focused on elucidating the signaling responsible for RV responses to chronic hypoxia and provide strong rationale to continue this line of investigation.

In summary, this manuscript illustrates the importance of considering the cell type and HIF family member contributing to pathology. With the previous focus on the role of HIFs in the lung with respect to hypoxia-induced PH, this study provides new insight into the distinct gene targets mediated by each HIF in cardiac responses to prolonged hypoxic stress. The gene set identified will provide a strong foundation for their group and others upon which to base new studies. Perhaps most importantly, this work will inform ongoing studies aimed at targeting HIFs for PH and will be a critical factor for ultimate tailoring of therapies targeting dysregulated or excess HIF signaling in disease while preserving the protective effects of HIFs. The current work serves as a reminder that the heart should not be considered simply a bystander in CH-induced PH and clues gleaned from the differential HIF pathways activated may provide insight into other cardiac functions. ■

Author disclosures are available with the text of this article at www.atsjournals.org.

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