JACC: BASIC TO TRANSLATIONAL SCIENCE
VOL. 7, NO. 1, 2022
ISSN 2452-302X

Letter

RESEARCH LETTER

HIF-1α Cardioprotection in COVID-19 Patients

Myocardial injury with sustained cardiac involvement is noted in more than half (58%) of patients and can be detected months after SARS-CoV-2 infection.[1] SARS-CoV-2 infection directly triggers respiratory illness and frequently leads to both pulmonary and cardiovascular tissue hypoxia.[2] Hypoxia-inducible factor (HIF)-1α is a master transcriptional regulator of tissue hypoxia and has been reported to decrease the expression of angiotensin-converting enzyme 2 (ACE2), which may potentially attenuate SARS-CoV-2 cell entry and lessen the severity of COVID-19. Of note, decreased pathogenicity of SARS-CoV-2 has been reported at high-altitude regions, possibly secondary to reduced ACE2 in lungs of local inhabitants who are acclimatized to hypoxia.[3]

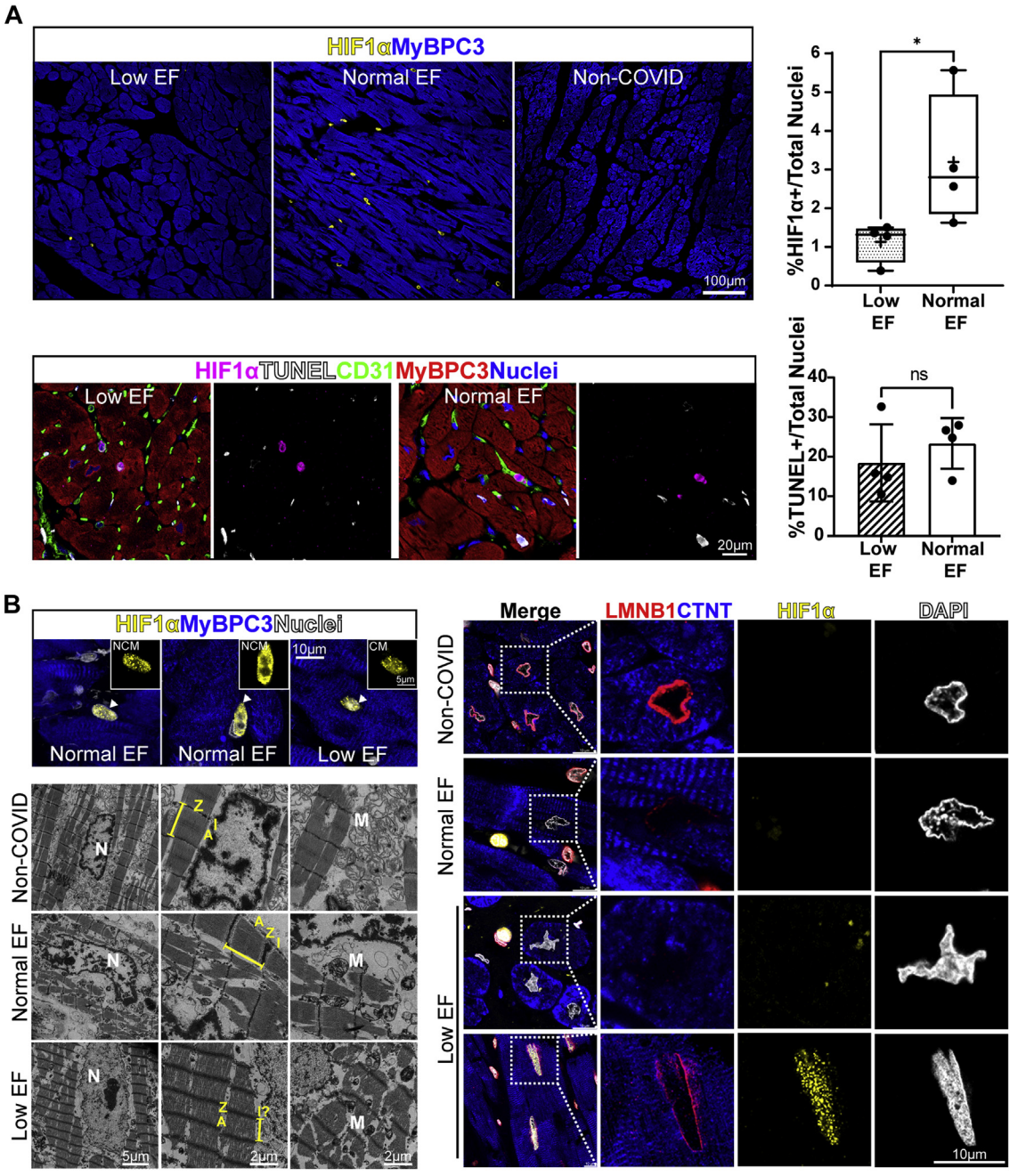
In this study, we examined markers of tissue hypoxia in post-mortem samples of 8 patients (age range 39 to 76 years; median age 64 years) with known SARS-CoV-2 infection. The COVID-19 patients were divided into 2 groups based on left ventricular ejection fraction (LVEF). Patients with LVEF >50% were categorized as "normal LVEF" (range 58% to 63%), and patients with LVEF <45% as "low LVEF" (range 33% to 43%). Three non-COVID hearts from donors of noncardiac cause of death served as control hearts (age range 59 to 79 years, median age 60 years). HIF-1α expression, which was determined by immunohistochemistry, was significantly higher in the hearts of patients with normal LVEF in comparison to the low LVEF group, whereas scant HIF-1α$^+$ cells were observed in non-COVID hearts (**Figure 1A**). HIF1α was immunolocalized to CD31$^+$ endothelial cells in both the low and normal LVEF groups. Notably, all HIF-1α$^+$ cells in the heart were terminal deoxynucleotidyl transferase dUTP nick end labeling (TUNEL)-negative (**Figure 1A**). These findings suggest, but do not prove, that HIF-1α up-regulation confers cytoprotective responses in endothelial cells in the hearts of COVID-19 patients.

In normal LVEF patients, HIF-1α was predominantly expressed in nonmyocytes, with either nuclear or perinuclear localization (**Figure 1B**). By contrast, in low LVEF patients, HIF-1α accumulated in a speckled pattern in cardiomyocyte nuclei (**Figure 1B**). We then examined nuclear and sarcomeric ultrastructure by transmission electron microscopy. Similar to non-COVID control hearts, the nuclear envelope appeared thick and dense in the normal LVEF hearts (**Figure 1B**). In low LVEF hearts, the nuclear envelope was thinner, without disruption of the lamin membrane. Sarcomeric damage was noted in the low LVEF heart as evidenced by swollen Z lines, smeared I bands, contracted sarcomeres, and distorted myofibril arrangements, whereas sarcomere disarray was not apparent in the normal LVEF group or in the non-COVID control hearts. Fewer mitochondria were detected in the hearts of low and normal LVEF groups of COVID-19 as compared with the hearts of control subjects. The mitochondria were scattered among myofibrils, rather than regularly aligned rows, as noted in control hearts. Additionally, mitochondria in low LVEF hearts were smaller, with swollen cristae.

We also examined heart sections by coimmunostaining with nuclear envelope marker lamin B1 and HIF-1α. In non-COVID hearts, lamin B1 was continuously expressed in all cells (**Figure 1B**). In COVID-19 cardiomyocytes that lacked HIF-1α expression, lamin B1 expression was decreased in normal LVEF hearts, whereas it was undetectable in low LVEF hearts. Nuclei of the HIF-1α$^-$ myocytes appeared to be enlarged or irregularly shaped. This observation is in line with a recent report of loss of nuclear DNA in SARS-CoV-2–infected induced pluripotent stem cell-derived cardiomyocytes in vitro.[4] Surprisingly, lamin B1 was found to be preserved in cardiomyocytes with nuclear HIF-1α accumulation, with healthy spindle-shaped nuclear morphology. HIF-1α clustered into speckles when it accumulates in cardiomyocyte nuclei, a unique distribution pattern of endogenous HIF-1α, HIF-2α, and HIF-1β previously seen in immortal cell lines.[5] Together, this evidence supports a protective role of HIF1α in maintaining DNA stability in cardiomyocytes. Although descriptive, these studies may shed insights into mechanistic pathways that lead to the development of cardiac dysfunction in SARS-CoV-2 cardiomyopathy.

Bingyan J. Wang, PhD
Sangeetha Vadakke-Madathil, PhD
Lori B. Croft, MD
Rachel I. Brody, MD, PhD
***Hina W. Chaudhry, MD**

FIGURE 1 **HIF-1α Cardioprotection in COVID-19 Patients**



(A) Top: HIF-1α expression in COVID-19 left ventricles with low LVEF (<45%), normal LVEF (>50%), and non-COVID control hearts. Percentage of HIF-α^+ cells over total cells counted indicating a clear increase in number of HIF-$1\alpha^+$ cells with higher LVEF. **Horizontal bar** = median. **Plus sign** = mean. *P = 0.029 by unpaired t-test, nonparametric, Kolmogorov-Smirnov. n = 4. **Bottom:** Coexpression of HIF1α and CD31 showing endothelial cells have increased expression of HIF-1α. HIF-$1\alpha^+$ cells were TUNEL− in both LVEF groups. There was no difference in the number of TUNEL$^+$ cells between the LVEF groups. n = 4. **(B) Top left:** In normal LVEF hearts, HIF-1α was primarily expressed in noncardiac myocytes in the nuclear/perinuclear area. In low LVEF hearts, HIF-1α was expressed exclusively in cardiomyocyte nuclei. n = 4. **Bottom left:** Transmission electron microscopy of COVID-19 and non-COVID hearts showing striking differences in nuclear ultrastructure, sarcomere structure/myofibril arrangement, and mitochondria between the 2 LVEF groups. Due to sample scarcity, representative images are from 1 heart per group (5 to 8 scans per heart). **Right:** Lamin B1 in non-COVID hearts shows intact nuclear envelope in healthy cardiomyocytes. Cardiomyocytes with no nuclear HIF-1α display low/undetectable lamin B1 expression, whereas cardiomyocytes with nuclear HIF-1α exhibiting intact lamin B1 and compact nuclear morphology. 4 of non-COVID control hearts/n = 4 of COVID patients per LVEF group. A = A band; EF = ejection fraction; I = I band; LVEF = left ventricular ejection fraction; M = mitochondria; N = nucleus; TUNEL = terminal deoxynucleotidyl transferase dUTP nick end labeling; Z = Z line.

*Cardiovascular Regenerative Medicine
Icahn School of Medicine at Mount Sinai
1 Gustave Levy Place
New York, New York 10029, USA
E-mail: hina.chaudhry@mssm.edu

https://doi.org/10.1016/j.jacbts.2021.12.001

All studies and protocols were reviewed and exempted by the institutional review board at Mount Sinai Hospital to be "not human subjects." Dr Chaudhry is supported by Empire State Stem Cell Board IIRP grant, contract number C32608GG and NIH grant R01HL150345. Dr Chaudhry is the founder and equity holder of VentriNova, Inc. All other authors have reported that they have no relationships relevant to the contents of this paper to disclose.

The authors attest they are in compliance with human studies committees and animal welfare regulations of the authors' institutions and Food and Drug Administration guidelines, including patient consent where appropriate. For more information, visit the Author Center.

REFERENCES

1. Huang L, Zhao P, Tang D, et al. Cardiac involvement in patients recovered from covid-2019 identified using magnetic resonance imaging. *J Am Coll Cardiol Img*. 2020;13(11):2330–2339.

2. Thachil J. Hypoxia-an overlooked trigger for thrombosis in COVID-19 and other critically ill patients. *J Thromb Haemost*. 2020;18:3109–3110.

3. Arias-Reyes C, Zubieta-DeUrioste N, Poma-Machicao L, et al. Does the pathogenesis of SARS-CoV-2 virus decrease at high-altitude? *Respir Physiol Neurobiol*. 2020;277:103443.

4. Perez-Bermejo JA, Kang S, Rockwood SJ, et al. SARS-CoV-2 infection of human iPSC-derived cardiac cells reflects cytopathic features in hearts of patients with COVID-19. *Sci Transl Med*. 2021;13(590):eabf7872.

5. Taylor SE, Bagnall J, Mason D, Levy R, Fernig DG, See V. Differential sub-nuclear distribution of hypoxia-inducible factors (HIF)-1 and -2 alpha impacts on their stability and mobility. *Open Biol*. 2016;6(9):160195.