Differential Mitochondrial Function in Remodeled Right and Nonremodeled Left Ventricles in Pulmonary Hypertension

ANISHA A. GUPTE, PhD,[1] ANDREA M. CORDERO-REYES, MD,[2] KEITH A. YOUKER, PhD,[2] RISË K. MATSUNAMI, PhD,[3] DAVID A. ENGLER, PhD,[3] SHUMIN LI, PhD,[1] MATTHIAS LOEBE, MD,[2,4] GUHA ASHRITH, MD,[2,5] GUILLERMO TORRE-AMIONE, MD, PhD,[2,5,6] AND DALE J. HAMILTON, MD, FACP[1,7]

Houston, Texas; and Nuevo Leon, Mexico

ABSTRACT

Objectives: Right ventricular failure is the primary reason for mortality in pulmonary hypertension (PH), but little is understood about the energetics of the failing right myocardium. Our aim was to examine mitochondrial function and proteomic signatures in paired remodeled right (RM-RV) and non-remodeled left (NRM-LV) ventricular tissue samples procured during heart-lung transplantation.

Methods and Results: Contractile dysfunction in RM-RV and preserved contractile function in NRM-LV were determined clinically and by echocardiography. Mitochondria were isolated from fresh paired RV and LV wall specimens of explanted hearts. Respiratory states in response to 4 substrates and an uncoupler were analyzed. Proteomic analysis on the mitochondrial isolates was performed with the use of liquid chromatography−mass spectrometry. The RM-RV mitochondria exhibited higher succinate state 4 levels with lower respiratory control ratio (RCR) compared with state 4 levels for pyruvate-malate (PM) and glutamate-malate (GM). RM-RV mitochondria also exhibited lower state 3 for palmitoyl-carnitine (PC) and state 4 for all complex I substrates compared with NRM-LV. The mean RCR were greater in RM-RVs than in NRM-LVs for PM and GM, which is consistent with tight coupling (low state 4 rates, higher RCRs); however, low RM-RV state 3 rates suggest concurrent substrate-dependent impairment in respiratory capacity. Mitochondrial proteomics revealed greater levels of mitochondrial ADP-ATP translocase and proteins of ATP synthesis, mitochondrial pyruvate and short branched chain acyl-CoA metabolism in RM-RV.

Conclusions: The mitochondrial respiration and proteomics in RM-RV are different from NRM-LV. These results have important implications in expanding our understanding of RV metabolism and future management of RV failure. (*J Cardiac Fail 2016;22:73−81*)

Key Words: Mitochondria, right ventricle, remodeling, pulmonary hypertension.

Pulmonary hypertension (PH) is a syndrome characterized by increased pulmonary arterial pressure that leads to adverse remodeling of pulmonary arterioles and the right ventricle (RV). There are several etiologies for PH,[1] but it is the decline in RV function that ultimately determines patient outcomes in PH.[2] Despite advances in the medical

From the [1]Bioenergetics Program, Houston Methodist Research Institute, Weill Cornell Medical College, Houston, Texas; [2]Methodist DeBakey Heart and Vascular Institute, Houston Methodist Research Institute, Houston, Texas; [3]Proteomics Programmatic Core Laboratory, Houston Methodist Research Institute, Houston, Texas; [4]Department of Cardiovascular Surgery, Houston Methodist Research Institute, Weill Cornell Medical College, Houston, Texas; [5]Department of Cardiology, Houston Methodist Research Institute, Weill Cornell Medical College, Houston, Texas; [6]Catedra de Cardiologia y Medicina Vascular, Tecnologico de Monterrey, Nuevo Leon, Mexico and [7]Department of Medicine, Houston Methodist Research Institute, Weill Cornell Medical College, Houston, Texas.

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Reprint requests: Dale J. Hamilton, MD, FACP, Houston Methodist Hospital and Research Institute, 6550 Fannin, Suite SM1001, Houston, TX 77030. Tel: +1-713-441-4452; Fax: +1-713-790-6617. E-mail: djhamilton@houstonmethodist.org

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management, the survival for PH patients 3 years after diagnosis continues to be only 67%.[3] Furthermore, a recent study reported that the adjusted hazard ratio for mortality was 2.41 in patients with PH and RV failure, even with current management standards.[4] Some PH patients require combined heart-lung transplantation, a procedure limited by the scarce availability of compatible donor organs.

During progression of PH, the RV initially responds to increased afterload by undergoing adaptive remodeling, but sustained increase in afterload leads to maladaptive remodeling associated with eccentric hypertrophy and diminished systolic and diastolic function, ultimately leading to RV failure.[5] The initial adaptive remodeling includes a shift from mitochondrial fatty acid oxidation to glycolysis, which then switches from aerobic to anaerobic metabolism during maladaptive remodeling.[6] However, these derangements have not been completely characterized in human RVs.[5,7–9] A better understanding of the energetics of the RV could offer new therapeutic strategies to enhance RV function and improve survival.

Our understanding of the metabolic and mitochondrial changes in the remodeled RV (RM-RV) is derived primarily from nonhuman animal studies, which have shown conflicting results depending on the severity of disease and nature of the model.[7] In human failing RVs, primarily surrogate markers of mitochondrial function, such as mitochondrial membrane potential, structure, and gene expression, have been assessed. It was shown that mitochondria from the failing RV are hyperpolarized and that the increased mitochondrial membrane potential correlated positively with hypertrophy.[10] Thus, a comprehensive understanding of the electron transport system function, especially in human RM-RV in PH and right heart failure, remains incomplete. Given the difficulty in safely procuring adequate amounts of fresh RV wall tissue for mitochondrial respiratory analyses, studies in human RV failure have been very few.[10] The aim of the present study was to characterize the mitochondrial function from freshly harvested human RM-RV and the paired non-remodeled left ventricular (NRM-LV) wall tissue from the same heart. We additionally characterized the proteomic profile of the same paired RM-RV and NRM-LV mitochondria. Results from these studies may have a significant impact on designing metabolically targeted therapeutic interventions to manage RV failure.

Methods

Patients and Tissue Collection

Paired RV and LV wall tissue were collected from 4 patients undergoing heart-lung transplantation at the Houston Methodist Hospital. Of the 9 patients undergoing heart-lung transplantation within the 2-year time frame of the study, only 4 patients met the criteria of remodeled RV with preserved LV function. Table 1 and Table 2 list the demographic, hemodynamic, and clinical characteristics, and Table 3 lists the medications of the 4 patients. The investigation conformed with the principles outlined in the Declaration of Helsinki[11,12] and was approved by the Houston Methodist Hospital's Institutional Review Board [IRB(2) 0511−0100]. Signed consent was obtained from each patient before enrollment for the study. Tissues collected in the operating rooms were immediately transported on ice-cold buffer A (220 mmol/L mannitol, 70 mmol/L sucrose, and 5 mmol/L MOPS)[13,14] to the laboratory for mitochondrial isolation.

Mitochondrial Isolation and Functional Assessment

Mitochondria were isolated from fresh human RV and LV wall tissue as previously described,[14] which yields primarily subsarcolemmal mitochondria with limited interfibrillar mitochondria. Approximately 100 μg mitochondria was used in each assay, and all readings were normalized to milligram of mitochondrial

Table 1. Demographic, Hemodynamic, and Clinical Data

Subject	Subject 1	Subject 2	Subject 3	Subject 4
Age (y)	47	17	51	66
Sex	F	F	F	F
Ethnicity/race	AA	AA	W	W
BMI (kg/m^2)	27.8	28.3	22.7	25.7
Diagnosis	WHO group I PH (idiopathic PH)	WHO group III PH (ARDS/lung necrosis)	WHO group III PH (pulmonary fibrosis)	WHO group multifactorial PH II/III (COPD/MVR)
Echocardiographic parameters				
RV function (qualitative)	Severely depressed	Moderately to severely depressed	Severely depressed	Mildly Decreased
RV Size	Enlarged	Enlarged	Enlarged	Enlarged
LVEF (%)	40−44	>70	60−64	65−69
LVIDd (cm)	4.4	2.6	3	4.5
LVIDs (cm)	3.2	1.3	1.8	2.8
Pulmonary arterial catheter parameters				
Right atrial pressure (mm Hg)	12	12	21	9
Pulmonary artery pressure (mean) (mm Hg)	74/30 (45)	76/28 (44)	120/33 (57)	65/25 (38)
Cardiac output (L/min)/cardiac index (L min^{-1} m^{-2})	4.7/2.6	4.4/2.3	2.8/1.6	5.7/3.2

F, female; AA, African American; W, white; WHO, World Health Organization; PH, pulmonary hypertension; ARDS, acute respiratory distress syndrome; COPD, chronic obstructive pulmonary disease; MVR, mitral valve replacement; RV, right ventricle; LV, left ventricle; LVEF, left ventricular ejection fraction; LVIDd, Left ventricular internal diameter at diastole; LVIDs, Left ventricular internal diameter at systole.