Strategic Plan for Lung Vascular Research:

An NHLBI-ORDR Workshop Report

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Online Data Supplement

Supplementary Material:

The Workshop presentations included many excellent slides of published and new data, overviews, and schematics and illustrations. We provide a brief description of select slides, the session from which they were derived, and the presenting authors. The selected slide sets represent major concepts that emerged from the open discussions during the Workshop. The S1-S2 set is overview material. Slides S3-S7 depict the identified need for continued research focused on the lung vasculature itself using novel tools and approaches, such as high-throughput technologies, pharmacogenomics, systems biology, metabolomics, and vascular proteome mapping. The S8-S9 set represents the identified need to "un-silo" lung vascular research and begin defining integrated lung vascular biology, i.e. research on circulating elements' effects on lung vascular development and function as well as identifying systemic vascular bed-lung vascular "crosstalk." The last set (S10-S15) represents the identified need to define disease in terms of the entire lung vascular-cardiac axis and thereby design approaches to clinical research inclusive of right heart measures.

Figure S1:

Overview of the Lung's Vasculature: Lung vascular structures include the pulmonary circulation proper, the bronchial vessels, and the lymphatics. Normal and integrated function of these vascular beds is essential for healthy airway function, gas exchange, and fluid balance. Disease arising in lung airways, such as or COPD and emphysema, or parenchymal tissues, such as fibrosis, will impact vascular cells and tissues. Diseases and injuries of the vasculature itself, particularly the pulmonary circulation, compromise local lung function, cardiac performance, and produce dysfunction in overall cardiovascular homeostasis severe enough to cause significant morbidity and mortality. Advances in treating lung vascular diseases will be accomplished with increased knowledge of lung vascular biology, including

defining lung vascular regeneration potential and pathways (from “Workshop Introduction and Charge to Participants;” provided by S. Erzurum).

Figure S2:

Differing Structures-Different Functions: Bronchial microcirculation vessels (left panel) demonstrate the typical systemic vascular arrangement of arteriolar vessels feeding into simple capillaries to deliver oxygen and nutrients to the airway tissues that drain back into venules returning blood to the large veins and heart. The pulmonary circulation architecture (right panel) is designed to optimize gas exchange; low resistance arterioles supply vast networks of capillaries engulfing alveoli which collectively supply oxygen rich blood to pulmonary venules and veins. The microanatomy of the pulmonary circulation is essentially networks of endothelial cells suspended in air (from “Vascular Crosstalk between Pulmonary and Systemic Circulations;” provided by D. McDonald).

Figure S3:

Understanding Genes-to-Function Using Systems Biology: High-throughput techniques have allowed for large datasets to be acquired for entire genomes. The genome and its resultant proteome/metabolomes give rise to the phenome and in the setting of environmental perturbations, the pathophenome. A tool for interpreting the genome-phenome relationship and alterations in disease is systems biology. Given emerging opportunities to define the lung vascular genome/proteome/metabolome, systems biology will become useful in advancing our understanding of lung vascular diseases (from “Integrating –omics and Systems Biology Approaches;” provided by J. Loscalzo).

Figure S4:

Systems Schema for Pulmonary Arterial Hypertension (PAH): Mutations in TGF-β signaling pathway genes Alk-1, BMPR2, and Endoglin are associated with PAH. However, disease manifestation is dependent upon input by other "disease-modifying" genes and environmental determinants. The integration of these inputs against the primary mutation background can produce intermediate phenotypes and/or the pathophenotype. Systems Biology modeling defines the network of factors that produce disease around a gene mutation identified by targeted sequencing and/or genome-wide associated studies (GWAS) and by repeat testing with variable modification (from "Integrating –omics and Sytems Biology Approaches;" provided by J. Loscalzo).

Figure S5:

Manifestation of Disease: These are lesions which represent the culmination of genotype-environment interactions, producing vasculopathy and causing fatal pulmonary hypertension. The lesions are characterized by endothelial cell abnormalities leading to occlusion of the vascular lumen and producing profound alterations in pulmonary blood flow. There are no therapies available that specifically target these lesions (from "Discovery of Novel PAH Treatments;" provided by R. Tuder).

Figure S6:

Utilizing -omics for Therapeutics: While discovery and understanding of the networks comprising lung vasculopathy development is one challenge that may be addressed with the systems biology tool, an additional challenge becomes utilizing that information to design therapies. Pharmacogenomic GWAS approaches are promising for matching designer therapies, i.e. drug effects, to specific clinical phenotypes. As lung vascular research progresses, there will be opportunities to test disease-modifying treatments (from "Discovery of Novel PAH Treatments;" provided by R. Weinshilboum).

Figure S7:

Routing to the Right "Zip Code": Designing the right disease-modifying, drug based upon –omics technologies and model testing through a systems biology approach, ultimately may be accomplished. However, drug delivery could still be limiting in successful treatment of lung vascular disease. Additional utility of proteomic approaches will be to define the molecular diversity of tissue vasculature between organs, within an organ, and importantly, between healthy and diseased vascular loci (from "Vascular Crosstalk between Pulmonary and Systemic Circulations;" provided by R. Tuder).

Figure S8:

Future Topics for Investigation in Circulating Elements-Lung Vascular Research: Self-explanatory slide which identifies research questions related to improving our understanding of progenitor cells and lung vascular interactions. Emphasis is placed on lung endothelium as an integration site for bone marrow-derived proangiogenic precursor cell signaling (from "Circulating elements;" provided by K. Asosingh).

Figure S9:

Liver-Lung: Clues on remodeling programs and pathways in the pulmonary circulation may be derived from increasing our understanding of systemic vasculature pathologies causing lung vascular dysfunction. An example includes hepatopulmonary syndrome (HPS) versus porto-pulmonary hypertension (PPHTN) in which the pulmonary microcirculation becomes characteristically vasodilated or the pulmonary resistance vessels become constricted and remodeled, respectively. Mechanisms have not been elucidated (from "Vascular Crosstalk between Pulmonary and Systemic Circulations;" provided by M. Fallon).

Figure S10:

Large Vessel Disease is Impactful In Addition to Small Vessels Disease: Proximal pulmonary artery health as reflected by compliance is recognized to have an independent effect on mortality in aging. The underlying pathogenesis causing stiffening is not known, but proximal stiffening leads to high pulsatility in distal vessels which can alter shear effects on small vessel lumen. Stiffening also increases right heart afterload. Clinical measures, such as impedance, can be taken to assess pulmonary arterial stiffening and may be used to better estimate lung vascular health and disease (from "Improving PAH Care through Human Subjects Studies;" provided by K. Stenmark).

Figure S11:

Modulating Pulmonary Arterial Stiffness: Self-explanatory slide listing interventions and therapies known to reduce arterial stiffness and thereby presumably reduce age-associated morbidity and mortality (from "Improving PAH Care through Human Subjects Studies;" provided by K. Stenmark).

Figure S12:

Pulmonary Vascular Stiffness Responsiveness Coupling to Right Ventricular Responsiveness: Exercise, which reduces pulmonary arterial stiffness (FIG S11), appears to also affect right ventricular (RV) adaptive responses. Data being gleaned from the Multi-Ethnic Study of Atherosclerosis (MESA) shows a relationship of exercise independently on RV end-diastolic mass in normal, healthy subjects (from "Improving PAH Care through Human Subjects Studies;" provided by S. Kawut).

Figure S13:

Impacting Right Ventricular Function Regardless of Large or Small Vessel Disease Etiology:

Whether the primary disease arises within the large vessels of microcirculation, right ventricular (RV) dysfunction leading to failure is the result of increased afterload. The cause of mortality in PAH is RV failure. The point at which RV compensation is lost and failure ensues is not known, nor are the mechanisms responsible (from "Improving PAH Care through Human Subjects Studies;" provided by K. Stenmark).

Figure S14:

The Right Ventricle versus the Left Ventricle: Afterload responses, hypertrophic programs, HDAC inhibition responses, ANP expression, norepinephrine responses, phenylephrine challenge, and failure markers differ between the right ventricle (RV) and the left. The extent of differences between the two chambers has not been studied and the impact of lung vascular disease on RV susceptibility to targeted therapeutics is unknown. RV research opportunities are emerging (from "The Lung Vascular-Cardiac Axis;" provided by N. Voelkel).

Figure S15:

Support for the Right Ventricle: Understanding how to best manage right ventricular (RV) afterload in PAH may help prolong survival. While ultimately finding vascular disease-modifying therapies is the goal, therapeutics could be gauged to RV supportive end-points. It is clear that if the RV is unable to compensate in the setting of increased afterload, rapid mortality from PAH results (from "The Lung Vascular-Cardiac Axis;" provided by N. Voelkel).