STANFORD-DUKE

CARDIOVASCULAR RESEARCH SYMPOSIUM

November 29-30, 2018
Li Ka Shing Conference Center
Paul Berg Hall
Stanford University

Stanford
Cardiovascular Institute

Duke Cardiovascular Research Center
Duke University School of Medicine

THURSDAY, NOVEMBER 29, 2018

INTRODUCTORY REMARKS

1:00 pm *Welcome*
Joseph C. Wu, MD, PhD
Director, Stanford Cardiovascular Institute
Simon H. Stertzer, MD, Professor, and Professor of Medicine & Radiology
Stanford University

Howard Rockman, MD
Edward S. Orgain Professor of Cardiology
Professor of Medicine, Cell Biology, and Molecular Genetics
Duke University

Sean M. Wu, MD, PhD
Associate Professor of Medicine and, by courtesy, Pediatrics
Stanford University

1:05 pm *Introduction*
Lloyd Minor, MD
The Carl and Elizabeth Naumann Professorship for the Dean of the School of Medicine
Professor of Otolaryngology - Head and Neck Surgery, and, by courtesy, of Neurobiology
and Bioengineering
Stanford University

OPENING KEYNOTE

1:10 pm *Heart & Brain Sciences, Health Equity and the AHA*
Ivor Benjamin, MD
President, American Heart Association
Director, Cardiovascular Center
Professor, Department of Medicine, Division of Cardiology
Medical College of Wisconsin

GOOGLE BASELINE PROJECT: A STANFORD DUKE COLLABORATION

Session Chair: **Francois Haddad, MD**, Clinical Associate Professor, Cardiovascular Medicine, Stanford University

1:45 pm *Project Baseline: Convergence of Data & Healthcare*
David Maron, MD
Director of Preventive Cardiology
Clinical Professor, Cardiovascular Medicine
Stanford University

2:05 pm *Project Baseline: Return of Results - Seek and Ye Shall Find*
Svati Shah, MD, MHS
Vice-Chair of Translational Research
Associate Professor of Medicine
Duke University

2:25 pm *Project Baseline: Participants as Partners - Engage, Educate, Empower*
Ken Mahaffey, MD
Director of Stanford Center for Clinical Research
Vice Chair Clinical Research, Department of Medicine
Stanford University

2:45 pm **Coffee Break**

G PROTEIN-COUPLED RECEPTOR BIOLOGY ACROSS THE CONTINENT

Session Chair: **Helen Blau, PhD**, Donald E. and Delia B. Baxter Foundation Professor, Director, Baxter Laboratory for Stem Cell Biology, Stanford University

3:05 pm *Ubiquitin-dependent regulation of β-adrenergic receptor trafficking and signaling*
Sudha Shenoy, PhD
Associate Professor in Medicine and in Cell Biology
Duke University

3:30 pm *New Paradigms for Arrestin-Mediated Signaling at GPCRs and Other Receptors*
Sudar Rajagopal, MD, PhD
Assistant Professor in Cardiology
Co-Director of Duke Pulmonary Vascular Disease Center
Duke University

3:50 pm *β-receptor Subtype Regulation of Cardiotoxicity/Cardioprotection: Just When You Thought it was Safe to go Back in the Water*
Daniel Bernstein, MD
Alfred Woodley Salter and Mabel G. Salter Endowed Professor of Pediatrics
Stanford University

4:10 pm *Biased GPCR Signaling*
Howard Rockman, MD
Edward S. Orgain Professor of Cardiology
Professor of Medicine, Cell Biology, and Molecular Genetics
Duke University

4:30 pm **Conclusion of First Day and Departure**

FRIDAY, NOVEMBER 30, 2018

7:30 am **Arrival and Breakfast**

8:00 am *Welcome*
Robert Harrington, MD
Chair, Department of Medicine
Arthur L. Bloomfield Professor of Medicine
Stanford University

KEYNOTE LECTURE

Chair: **Howard Rockman, MD**, Edward S. Orgain Professor of Cardiology, Professor of Medicine, Cell Biology, and Molecular Genetics, Duke University

8:05 am *Structure-aided Drug Discovery for G Protein Coupled Receptors*
Brian Kobilka, MD
Professor of Molecular and Cellular Physiology
2012 Nobel Laureate
Stanford University

BASIC & TRANSLATIONAL APPROACHES TO VASCULAR & MYOCARDIAL BIOLOGY

Session Chair: **Marlene Rabinovitch, MD**, Dwight and Vera Dunlevie Professor in Pediatric Cardiology, Stanford University

8:50 am   *Genomic Approaches to Discover Novel Drug Targets for Obesity and Insulin Resistance*
Erik Ingelsson, MD, PhD
Professor of Medicine
Stanford University

9:10 am   *TREK-1 Affects the Cardiac Injury Response by Modulating Intercellular Crosstalk*
Dennis Abraham, MD
Assistant Professor of Medicine
Director of Duke Cardiovascular Physiology Core
Duke University

9:30 am   **Coffee Break**

9:40 am   *Genetic Mechanisms of Coronary Artery Disease: What Can we Learn From GWAS?*
Thomas Quertermous, MD
William G. Irwin Professor in Cardiovascular Medicine
Stanford University

10:00 am   *The Long and the Short of it: Telomere Length as a Hallmark of Cardiac Failure*
Helen Blau, PhD
Donald E. and Delia B. Baxter Foundation Professor
Director, Baxter Laboratory for Stem Cell Biology
Stanford University

10:20 am   *Unfolding the Misfolded: Molecular Insights into Cardiac Amyloidosis*
Ronglih Liao, PhD
Professor of Medicine
Co-Director, Stanford Cardiac Amyloid Center
Stanford University

10:40 am   **Coffee Break**

HEART FAILURE THERAPIES- PERCUTANEOUS, SURGICAL, AND REGENERATIVE APPROACHES

Chair: **Sharon Hunt, MD**, Professor Emerita of Medicine (Cardiolovascular), Stanford

10:50 am   *Innovative Therapies for Ischemic Cardiomyopathy*
Y. Joseph Woo, MD
Norman E. Shumway Professor
Chair, Department of Cardiothoracic Surgery
Stanford University

11:10 am   *Man vs. Machine: Will Mechanically Assisted Circulation Achieve Its Goals?*
Joseph Rogers, MD
Professor of Medicine
Chief Medical Officer, Duke University

11:30 am   *Personalized Medicine in Heart Transplantation*
Kiran Khush, MD, MAS
Associate Professor of Medicine
Stanford University

11:50 am   *Vascular Approaches to Innate Heart Regeneration*
Ravi Karra, MD
Assistant Professor of Medicine
Duke University

12:10 pm   **Lunch and Poster Viewing / Meet the Professors Lunch**

GENOME, TRANSCRIPTOME, AND PRECISION HEALTH

Chair: **Nigam Shah, MBBS, PhD**, Associate Professor of Medicine (Biomedical Informatics) and of Biomedical Data Science, Stanford University

1:10 pm   *Pharmacogenomics for Precision Cardiovascular Health*
Latha Palaniappan, MD, MS
Professor of Medicine
Stanford University

1:30 pm   *Unexpected Biology of snoRNAs in the Heart*
Chris Holley, MD, PhD
Assistant Professor of Medicine and Molecular Genetics and Microbiology
Duke University

1:50 pm *Population, Medical, and Functional Genomics in the Personal Genome Era*
Carlos Bustamante, PhD
Professor of Biomedical Data Science, of Genetics and, by courtesy, of Biology
Stanford University

2:10 pm **Coffee Break**

STATE-OF-THE-ART APPROACHES TO THE DETECTION AND TREATMENT OF EP DISEASES

Chair: **Sanjiv Narayan, MSc, MD**, Professor of Medicine (Cardiovascular Medicine), Stanford University

2:20 pm *Wearables and Arrhythmias: Where are we Headed?*
Mintu Turakhia, MD, MAS
Executive Director, Stanford Center for Digital Health
Associate Professor of Medicine
Palo Alto Veterans Affairs Health Care System

2:40 pm *Innovation and the Future of Arrhythmia Therapy*
Paul Wang, MD
Professor of Medicine
Stanford University
Editor of *Circulation: Arrhythmia and Electrophysiology*

BEST ABSTRACT TRAINEE PRESENTATIONS

Chair: **Elan Burton, MD**, Clinical Assistant Professor, Cardiothoracic Surgery, Stanford

3:00 pm *Molecular Mechanisms of Angiotensin Receptor Activation and Biased Signaling*
Laura Wingler
Duke University - Lefkowitz Lab

3:13 pm *Parental Disease and Over-transmission of Genetic Risk for Diabetes are Related to Congenital Heart Disease in Offspring*
Catherine Tcheandjieu
Stanford University - Priest Lab

3:26 pm *On the Intertwined Nature of Endothelium and Muscle: Defining New Signaling Paradigms in Peripheral Artery Disease*
Hasan Abbas
Duke University - Kontos Lab

3:38 pm *Modeling hypertrophic cardiomyopathy caused by mutations in beta-myosin using human induced pluripotent stem cell derived cardiomyocytes (iPSC-CMs)*
Alison Schroer
Stanford University - Bernstein Lab / Pruitt Lab (UCSB)

3:50 pm **Coffee Break**

POPULATIONS, QUALITY, AND OUTCOMES IN CARDIOVASCULAR CARE

Chair: **Paul Heidenreich, MD**, Professor of Medicine (Cardiovascular), and, by courtesy, of Health Research and Policy, Palo Alto Veterans Affairs Health Care System, Stanford

4:00 pm *Evaluating Precision Medicine*
Mark Hlatky, MD
Professor of Health Research and Policy and of Medicine
Stanford University

4:20 pm *Closing the Gaps in Care: What's Next in Implementation Science?*
Eric Peterson, MD
Fred Cobb Distinguished Professor of Medicine
Duke University

4:40 pm *Controversies in Economics of Precision Medicines*
Kevin Schulman, MD
Professor of Medicine
Associate Chair of Business Development and Strategy
Stanford University

5:00 pm **Wine and Cheese Reception and Poster Session with Presentation of Poster Awards**

Ivor Benjamin, MD

Ivor Benjamin, MD, is the Director of the Cardiovascular Center (CVC) and a co-leader of the CVC program in Precision Cardiovascular Medicine at the Medical College of Wisconsin. He received his bachelor's degree from Hunter College in New York and his MD from Johns Hopkins University School of Medicine. Dr. Benjamin is a certified specialist in internal medicine and cardiology. His research interests are in cardiology, inheritable heart failure, and myocardial infarctions, with a focus on the genes encoding heat shock proteins and oxido-reductive stress-response pathways and their relationship to genetic forms of heart disease, cardiotoxicological science, and precision medicine.

Dr. Benjamin is an Established Investigator of the American Heart Association (AHA). He has received the AHA Award of Merit, the Daniel Savage Memorial Service Award from the Association of Black Cardiologists, and the NIH Director's Pioneer Award from the National Heart, Lung, and Blood Institute. Dr. Benjamin serves as Editor-in-Chief of Cecil Essential Medicine 9th Edition. He is also the founding member of the J*ournal of the American Heart Association*, and serves on the editorial boards of *Circulation* and *Circulation Research*.

Brian Kobilka, MD

Brian Kobilka, MD, received his bachelor's degree in Biology and Chemistry from the University of Minnesota and his MD from Yale University School of Medicine. After internal medicine training at Washington University School of Medicine, Dr. Kobilka was a research fellow at Duke University, where he later became an Assistant Professor Medicine. He then moved to Stanford University School of Medicine, where he is now the Helene Irwin Fagan Chair in Cardiology, a Professor in the Department of Molecular and Cellular Physiology, and, by courtesy, of Chemical and Systems Biology. In 2012, Dr. Kobilka was awarded the Nobel Prize in Chemistry for his seminal work on G protein-coupled receptors. He has authored numerous peer-reviewed articles on his work characterizing the structure and mechanism of activation of GPCRs. He is a member of the National Academy of Sciences, the National Academy of Medicine, and the American Academy of Arts and Sciences.

David Maron, MD

David Maron, MD, is the Director of Preventive Cardiology and is board certified in Internal Medicine, Cardiovascular Disease, and Clinical Lipidology. He received his undergraduate degree at Stanford, his MD from the University of Southern California, and completed his residency in internal medicine at UCLA. Dr. Maron then completed a cardiology fellowship and a research fellowship in Cardiovascular Disease Epidemiology and Prevention at Stanford University. He faculty at Vanderbilt for 20 years before returning to Stanford in 2014. He is currently the Co-Chair and Principle Investigator of the International Study of Comparative Health Effectiveness with Medical and Invasive Approaches (ISCHEMIA) trial, a large, international, NIH-funded study.

Svati Shah, MD, MHS

Svati Shah, MD, is a physician scientist and Vice-Chief of Translational Research in the Division of Cardiology, Department of Medicine, and a faculty member and Co-Director of Translational Research in the Duke Molecular Physiology Institute (DMPI) and Duke Clinical Research Institute (DCRI). Her research focuses are metabolic and genetic pathways of cardiometabolic diseases, and integrating diverse genomic, metabolomic and proteomic techniques for identification of novel mechanisms of disease and biomarkers. Dr. Shah also collaborates closely with the DCRI for biomarker discovery in biospecimens from clinical trials and she is the Duke Principle Investigator for the Verily Project Baseline study. Dr. Shah is also Director of the Duke Adult Cardiovascular Genetics Clinic where she cares for patients and their families who have, or at risk for, cardiovascular genetic disorders. Her training includes receiving a MHS in Epidemiology from Johns Hopkins School of Public Health, a master's degree in Medical Genomics from Duke University, and a research fellowship in Genetic Epidemiology at the Duke Center for Human Genetics.

Ken Mahaffey, MD

Ken Mahaffey, MD, is the Vice Chair of Clinical Research in the Department of Medicine at Stanford University and the Director of the Stanford Center for Clinical Research (SCCR). SCCR is an academic research organization whose goal is to support researchers as they design and conduct clinical research studies, and to enroll Stanford patients in clinical trials. Dr. Mahaffey's own research focuses on the design and conduct of multicenter clinical trials and analyses of important clinical cardiac issues using large patient databases. He also studies the methodology of clinical trials including the standardization of the definition of MI used in clinical trials, the adjudication of clinical endpoints, and the evaluation of evidenced-based operations. He works with the FDA and chaired the Myocardial Infarction and Death Definitions Working Group in the Standardized Data Collection for Cardiovascular Trials Initiative.

Sudha Shenoy, PhD

Sudha Shenoy, PhD, received her bachelor's and master's degrees in India, studying Zoology, and her PhD at Oklahoma State University in Biochemistry and Molecular Biology. Dr. Shenoy continued at Oklahoma State University for her first postdoctoral fellowship, before completing her second fellowship at Duke University in Receptor Biology. She is an Associate Professor in Medicine and Cell Biology at Duke University School of Medicine and is a member of the Duke Cancer Institute. Dr. Shenoy research focuses on receptor biology, the biochemical characterization of ubiquitination, receptor trafficking, and signaling. Her research aims to understand how ubiquitination of G protein-coupled receptors (GPCRs) and the adaptor proteins β-arrestins 1 and 2 affect receptor endocytosis, signal transduction, and desensitization.

Sudar Rajagopal, MD, PhD

Sudar Rajagopal, MD, PhD, received his bachelor's degree in Chemistry from the University of Chicago. He continued at the University of Chicago for his medical degree and his doctorate in Biochemistry and Molecular Biology. He is an Assistant Professor in Cardiology at Duke University Medical Center, and is Co-Director of Duke Pulmonary Vascular Disease Center. He was named a Mandel Foundation Scholar by the Duke Cardiovascular Research Center and received the Jeremiah Stamler Distinguished Young Investigator Research Award from the Northwestern Cardiovascular Young Investigator Forum. His research focuses on the role of G protein-coupled receptor signaling in pulmonary arterial hypertension (PAH), with the goal of understanding how these receptors contribute to the pathophysiology and development of PAH.

Daniel Bernstein, MD

Daniel Bernstein, MD, received his bachelor's degree from MIT in Massachusetts and his medical degree from New York University School of Medicine. He is an Alfred Woodley Salter and Mabel G. Salter Endowed Professor of Pediatrics at Stanford University and is the Associate Dean for Curriculum and Scholarship at Stanford University School of Medicine. In addition, Dr. Bernstein received the Best Lecture Award from Stanford School of Medicine and the Stanford Stole (fellowship mentorship award) from Stanford University Pediatric Cardiology. His research focuses on regulation of cardiovascular function in both normal physiological states and in disease states such as cardiomyopathy. Current work includes using iPSC-derived cardiomyocytes to better understand heart failure and congenital heart disease and the role of mitochondrial dynamics, structure, and function in normal and diseased cardiac physiology.

Howard Rockman, MD

Howard Rockman, MD, received his bachelor's degree in Biochemistry and his medical degree from McGill University. He did his medical residency at Montreal General Hospital and a Cardiology Fellowship at the University of California, San Diego. He was then an Assistant Professor at the University of California, San Diego, before moving to the University of North Carolina at Chapel Hill, and finally to Duke University where he is now a Professor of Medicine, Cell Biology, and Molecular Genetics as well as an Edward S. Orgain Professor of Cardiology. Dr. Rockman is Editor-in-Chief of the *Journal of Clinical Investigation Insight* and is the Director of Duke Cardiovascular Research Center. His research focuses on understanding the molecular mechanisms of cardiac hypertrophy and heart failure.

Abraham Verghese, MD, MACP

Dr. Verghese is the Linda R. Meier and Joan F. Lane Provostial Professor and Vice Chair for the Theory and Practice of Medicine at Stanford University. He is also an infectious disease physician, best-selling author, and a popular speaker. His research seeks to understand what is conveyed to a patient by the physician's presence and technique at the bedside from an educational point of view, and also from ethnographic studies of how rituals impact patient-physician relationships. He launched the Stanford Medicine 25 initiative that is designed to showcase and teach fundamental physical exam skills and their diagnostic benefits. Dr. Verghese obtained his medical degree at the University of Madras, did his residency at East Tennessee State University, College of Medicine, and competed a fellowship in Infectious Disease at Boston University School of Medicine. He also earned a Master of Fine Arts degree at Iowa Writers Workshop. He is a member of the National Academy of Sciences and received the National Humanities Medal from President Obama.

Erik Ingelsson, MD, PhD

Erik Ingelsson, MD, PhD, received his medical degree and his PhD in Epidemiology from Uppsala University, in Uppsala, Sweden. He then completed a research fellowship at the Framingham Heart Study, Boston University School of Medicine in Cardiovascular Medicine. Dr. Ingelsson is a Professor of Medicine and, by courtesy, of Health Research and Policy. His research focuses on cardiology with a special focus on the role of obesity and insulin resistance in the development of subclinical and clinical cardiovascular disease. Dr. Ingelsson's work is translational and interdisciplinary, combining big data approaches with gene editing in functional model systems to obtain new biomarkers for risk prediction and for the discovery of novel targets for drug development.

Dennis Abraham, MD

Dennis Abraham, MD, received his bachelor's degree in Premedicine from Pennsylvania State University and his medical degree from Sidney Kimmel (Jefferson) Medical College. He completed an internship and his residency in Internal Medicine at Mount Sinai in New York, and was a postdoctoral research fellow at Columbia University Medical Center. Dr. Abraham then moved to Duke University Medical Center, where he is now an Assistant Professor of Medicine in the Division of Cardiovascular Medicine. He is also Director of the Duke Cardiovascular Physiology Core. Dr. Abraham's work is aimed at understanding how heart failure develops and the development of new drug therapies.

Thomas Quertermous, MD

Thomas Quertermous, MD, competed his clinical training in Cardiology at Massachusetts General Hospital and his research training in Molecular Genetics in the Department of Genetics at Harvard Medical School. He first established an independent laboratory at Massachusetts General Hospital before moving first to Vanderbilt University and then to Stanford University as a leader of the Division of Cardiovascular Medicine. He is currently a William G. Irwin Professor in Cardiovascular Medicine at Stanford University. Research in Dr. Quertermous's lab focuses on the use of genetic approaches to study vascular disease. Current efforts involve large-scale human genetics and genomics to better understand the genetic basis of atherosclerosis and its related risk factors.

Helen Blau, PhD

Helen Blau, PhD, received her bachelor's degree in Biology from the University of York, England, and her master's degree and Ph.D. in Biology at Harvard University in Massachusetts. She completed her postdoctoral research fellowship in the Division of Medical Genetics, Department of Biochemistry and Biophysics at the University of California, San Francisco before becoming an Assistant Professor in the Department of Pharmacology at Stanford University. Dr. Blau is currently a Donald E. and Delia B. Baxter Foundation Professor at Stanford University. She is also Director of the Baxter Laboratory for Stem Cell Biology, and of the Institute for Stem Cell Biology and Regenerative Medicine. Dr. Blau is a member of the National Academy of Sciences, Pontifical Academy of Inventors, American Institute for Medical and Biological Engineering, and the American Philosophical Society. Her research focuses on cellular reprogramming, therapeutic interventions to enhance stem cell function in muscle regeneration, and cell rejuvenation.

Ronglih Liao, PhD

Ronglih Liao, PhD, received her bachelor's degree in Chemistry from Tamkang University, Taiwan, and her PhD at the University of Alabama at Birmingham. She then became an Assistant Professor in the Department of Medicine at Boston University School of Medicine before moving to Harvard Medical School as an Associate Professor. She next moved to Stanford University School of Medicine where she is a Professor of Medicine and co-Director of the Stanford Cardiac Amyloid Center. She is also a visiting Professor of Medicine at Brigham and Women's Hospital, Harvard Medical School. She was the first female Council Chair of the AHA Basic Cardiovascular Science Council. Dr. Liao's research aims to understand the mechanisms that underlie the pathophysiology of heart failure and to develop novel treatments to combat heart failure.

Joseph Woo, MD

Joseph Woo, MD, is the Norman E. Shumway Professor and Chair of the Department of Cardiothoracic Surgery at Stanford University School of Medicine and holds a courtesy appointment in the Department of Bioengineering. He received his undergraduate degree from the Massachusetts Institute of Technology and his medical degree from the University of Pennsylvania, where he also conducted his postgraduate surgical training in General and Cardiothoracic Surgery and completed a postdoctoral research fellowship. Dr. Woo has an active clinical practice of 300 pump cases per year focusing on complex cardiac valve repair, aortic surgery, cardiopulmonary transplantation, and minimally-invasive surgery, and has advanced these fields by developing several innovative operations. He also directs basic research on stem cells, angiogenesis, tissue engineering, and biomechanics.
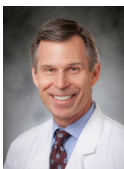
Joseph Rogers, MD

Joseph Rogers, MD, is a Professor of Medicine at Duke University. He obtained his bachelor's degree from the University of Kansas and attended medical school at the University of Nebraska. He performed his Internal Medicine residency at Nebraska prior to research and Clinical Cardiology training at Washington University in St. Louis. Following his fellowship, he remained on faculty at Washington University and directed their Cardiac Transplant program for ten years prior to moving to Duke. He also served as Medical Director of the Cardiac Transplant and Mechanical Circulatory Support programs at Washington University and Duke. Dr. Rogers's clinical interests are directed toward the management of patients with advanced heart failure. His research focuses on the clinical application of mechanical circulatory support devices, with an emphasis on palliative care intervention in advanced heart failure.

Kiran Khush, MD, MAS

Kiran Khush, MD, MAS, is an Associate Professor of Medicine in the Division of Cardiovascular Medicine at Stanford University School of Medicine. She obtained her bachelor's degree from Stanford University and her medical degree from Harvard Medical School. She completed her medical residency at the University of California, San Francisco (UCSF), followed by fellowships in General Cardiology, Echocardiography, and Advanced Heart Failure and Transplant Cardiology. Dr. Khush became an Assistant Professor at UCSF while also completing her master's degree in Clinical Research and Epidemiology. She then moved to Stanford University. Her clinical and translational research program focuses on heart transplantation.

Ravi Karra, MD

Ravi Karra, MD, received his medial degree from Duke University School of Medicine and completed his residency in Internal Medicine at Brigham and Women's Hospital and a Cardiology fellowship and an Advanced Heart Failure fellowship at Duke University Medical Center. He is currently an Assistant Professor in the Department of Medicine, Division of Cardiology, Section of Advanced Heart Failure at Duke University School of Medicine. He leads a research group focused on translating regenerative biology to patients with heart failure. His group uses state-of-the-art functional screening approaches to identify key mediators of heart regeneration. In addition, through collaboration with the Department of Biomedical Engineering, he has a focus on the design of biopolymers for delivering regenerative compounds to the heart. He is also a practicing cardiologist, specializing in recovery from heart failure.

Latha Palaniappan, MD, MS

Latha Palaniappan, MD, MS, received her bachelor's and medical degrees from the University of Michigan. She also obtained her master's degree in clinical epidemiology from Stanford University. Dr. Palaniappan is currently a Professor of Medicine and, by courtesy, of Health Research and Policy at Stanford University Medical Center. Her research focuses on addressing the gap in knowledge of health in Asian subgroups and other understudied racial and ethnic minorities. She co-founded PRANA, a South Asian Wellness program, and the Center for Asian Health Research and Education (CARE). Her current work examines the clinical effectiveness of structured physical activities for diabetes management and the best exercise regimes for normal-weight diabetics.

Christopher Holley, MD, PhD

Christopher Holley, MD, PhD, received his bachelor's degree in Biology from Duke University. He also obtained his MD and PhD in Pharmacology from Duke University. Dr. Holley then completed his residency in internal medicine and a cardiovascular clinical fellowship and cardiovascular research fellowship at Washington University School of Medicine / Barnes-Jewish Hospital in St. Louis, Missouri. He is now an Assistant Professor of Medicine, in the Sections for Heart Failure, Transplantation, and Mechanical Circulatory Support at Duke University School of Medicine as well as Assistant Professor of Molecular Genetics and Microbiology. His research focuses on the role of non-coding RNA in cardiovascular health and disease, with a focus on small nucleolar RNA (snoRNA).

Carlos Bustamante, PhD

Dr. Carlos D. Bustamante is an internationally recognized leader in the application of data science and genomics technology to problems in medicine, agriculture, and biology. He received his Ph.D. in Biology and MS in Statistics from Harvard University (2001), was on the faculty at Cornell University (2002-9), and was named a MacArthur Fellow in 2010. He is currently Professor of Biomedical Data Science, Genetics, and (by courtesy) Biology at Stanford University. Dr. Bustamante has a passion for building new academic units, non-profits, and companies to solve pressing scientific challenges. He is Founding Director of the Stanford Center for Computational, Evolutionary, and Human Genomics (CEHG) and Inaugural Chair of the Department of Biomedical Data Science.

Mintu Turakhia, MD, MAS

Mintu Turakhia, MD, MAS, is a cardiac electrophysiologist, outcomes researcher, and clinical trialist. He is the Executive Director of the Stanford Center for Digital Health and Associate Professor of Medicine at the Palo Alto Veterans Affairs Health Care System. Dr. Turakhia has an active, highly-funded multidisciplinary program in heart rhythm research, where he uses big data, biostatistical, and data science approaches to examine quality, outcomes, and risk of heart rhythm disorders such as atrial fibrillation. He is a principal investigator of several multi-center trials to test digital health tools and wearable devices in the detection and treatment of heart disease. Dr. Turakhia is an elected member of the American Society of Clinical Investigation and Fellow of the American Heart Association, American College of Cardiology, and Heart Rhythm Society.

Paul Wang, MD

Dr. Paul Wang is a Professor of Medicine at the Stanford University School of Medicine and co-Director of Bioengineering Scholarly Concentration. He received his bachelor's degree from Harvard University and his MD at the College of Physicians and Surgeons Columbia University. Dr. Wang is an expert in the treatment of cardiac arrhythmias, was co-inventor of catheter cryoblation, and has pioneered new techniques in the management of heart rhythm issues. He has co-authored numerous textbooks and book chapters and is past Chair of the American Heart Association Council on Clinical Cardiology ECG and Arrhythmias Committee. In addition, Dr. Wang founded the annual Stanford Biodesign New Arrhythmia Technologies Retreat, focusing on new technological advances in arrhythmia management and diagnosis. He is Editor-in-Chief of *Circulation:Arrhythmia* and *Electrophysiology*.

Mark Hlatky, MD

Mark Hlatky, MD, is a Professor of Health Research and Policy and of Medicine at Stanford University, and is Director of Health Services Research Master's Degree Program. He received his bachelor's degree in Physics from Massachusetts Institute of Technology and his medical degree from the University of Pennsylvania. His interests are in outcomes research, evidence-based medicine, and cost-effectiveness analysis. He pioneered the collection of data on economic and quality of life outcomes as part of randomized trials. He has also developed decision models to assess the efficacy and cost-effectiveness of clinical strategies. Dr. Hlatky has been awarded both the Distinguished Scientist Award from the American Heart Association and the Distinguished Scientist Award from the American College of Cardiology.
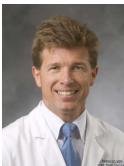
Eric Peterson, MD

Dr. Eric Peterson is a Fred Cobb Distinguished Professor of Medicine in the Division of Cardiology at Duke University. He received his medical degree from the University of Pittsburg, completed his residency at Children's Hospital Boston, was a Fellow in General Internal Medicine at Harvard University, and a Fellow in Cardiology at Duke University. Dr. Peterson is the Principal Investigator of the National Institute of Health, Lung, and Blood Institute Coordinating Center for Education and Research on Therapeutics. He received the American Heart Association Meritorious Achievement Award and was voted one of the world's top 400 most influential researchers in biomedicine. With over 800 peer-reviewed publications, Dr. Peterson is recognized as a leader in outcomes and quality research.

SPEAKER BIOS

Kevin Schulman, MD

Kevin Schulman, MD, is Professor of Medicine at Stanford University School of Medicine and, by courtesy, Professor of Economics at Stanford Graduate School of Business. He is also Associate Chair of Business Development and Strategy in the Department of Medicine and Director of Industry Partnerships and Education for the Clinical Excellence Research Center (CERC) at Stanford University School of Medicine. Dr. Schulman's research interests include organizational innovation in health care, health care policy, and health economics. He is the Founding President of the Business School Alliance for Health Management, which consists of the leading business schools offering health management programs. Dr. Schulman is an elected member of the American Academy of Pediatrics and the American Society for Clinical Investigation.

ORGANIZING COMMITTEE

Joseph C. Wu MD, PhD
Simon H. Stertzer, MD, Professor, and Professor Medicine & Radiology
Director, Stanford Cardiovascular Institute

Howard Rockman, MD
Edward S. Orgain Professor of Cardiology
Professor of Medicine, Cell Biology, and Molecular Genetics
Duke University

Sean M. Wu, MD, PhD
Associate Professor of Medicine, and by courtesy, of Pediatrics
Stanford University

1. Proteomics of Right Heart Failure in Patients with Pulmonary Arterial Hypertension

Myriam Amsallem MD MS[1,2,3*], Andrew J. Sweat MD[4*], Jennifer Arthur Ataam PhD[2,3], Edda Spiekerkoetter MD[4], Marlene Rabinovitch MD PhD[5], Elie Fadel MD PhD[3], Olaf Mercier MD PhD[3], François Haddad MD[1,2*] and Roham Zamanian MD PhD[4,5*]

[1]Div. of Cardiovascular Medicine , Stanford University School of Medicine, CA, USA; [2]Stanford Cardiovascular Institute, Stanford University School of Medicine, CA, USA; [3]Research and Innovation Unit, INSERM U999, Marie Lannelongue Hospital, Paris Sud University, France; [4]Div. of Pulmonology and Critical Care, Stanford University School of Medicine, CA, USA; [5]Vera Moulton Wall Center at Stanford University School of Medicine, CA, USA; *Both first and senior authors contributed equally to the study.

Background: Inflammatory features have been reported in pressure-overloaded right ventricle. This study sought to determine the circulating immune proteomic profile associated with right heart maladaptive phenotype (RHMP) in patients with pulmonary arterial hypertension (PAH).

Methods: This study included a discovery cohort (n=121, from 2008 to 2011) and a validation cohort (n=76, from 2011 to 2014), who underwent plasmatic proteomic profiling using 48-plex flow cytometry multiplex Luminex® (including interleukins, chemokines and growth factors). RHMP was defined using the Mayo right heart score (based on right ventricular RV longitudinal strain, NYHA class and NT-proBNP) and the Stanford right heart score (based on RV end-systolic remodeling index, NYHA class and NT-proBNP). The association between cytokines and RHMP was assessed using partial least square regression analysis.

Results: The median age of the discovery cohort was 50 [39 – 59] years, with a majority of female (74%), and 33% with connective tissue disease. Patients from the validation cohort had more severe features (lower six minute walk test distance, lower cardiac index and higher levels of NT-proBNP) than patients from the discovery cohort, with similar resistance levels and right heart echocardiography metrics. High levels of hepatic growth factor (HGF), stem cell growth factor beta (SCGFβ) and nerve growth factor (NGF) were significantly associated with worst Mayo and Stanford scores but not with pulmonary vascular resistance or mean pulmonary arterial pressure, in both cohorts.

Conclusion: High plasmatic levels of HGF, SCGFβ and NGF are associated with right heart adaptive phenotypes beyond pulmonary disease severity in patients with PAH.

2. Multi-Echo Flow-encoded Rosette (MELROSE) enables velocity and T2* assessment of both extravascular tissue and intravascular blood for motion robust, quantitative cardiovascular blood flow and oxygenation mapping

Adam Bush, Christopher Sandino, Marcus Alley, Shreyas Vasanawala

Cardiovascular Cardiac catheterization is an invasive albeit common procedure performed in children with congenital heart disease for intrathoracic oxygen saturation assessment, exposing patients to anesthesia and risk of infection and complication. Prior MRI based intrathoracic oximetry methods have been limited due to partial volume contamination of the blood pool with surrounding tissue and motion corruption. Recently, subtractive MRI oximetry methods have demonstrated reliability and robustness but

are limited to Cartesian strategies in the brain. In this work we use a subtractive, velocity encoded, non-Cartesian rosette trajectory for quantitative, motion robust, extra and intravascular flow and T2* mapping entitled Multi Echo fLowencoded ROSEtte (MELROSE). We validate flow and T2* values in a flow phantom and present preliminary results in a healthy subject. Theory Rosette trajectories are flower-like k-space traversal patterns first described by Noll. Rosette trajectories have several advantages over spiral and radial sequences, including higher average gradients and improve incoherence for compressed sensing application yet remain largely unused. In this work, we use a novel rosette shape parameterization, q. Each repetition time, a "flower" is acquired, representing a highly undersampled k-space acquisition. By performing a multi-shot sequence and incrementing successive flowers by the golden angle (137.5°) a fully sampled data set can be acquired. Recombination of individual petals, or samples between temporally adjacent center crossings, allows for multi-echo reconstructions... Abstract truncated

3. Induced pluripotent stem cell modeling of insulin resistance and endothelial dysfunction

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Background: Cardiovascular disease (CVD) is the number one cause of death globally, with 17.5 million deaths per year. Insulin resistance is a precursor to type 2 diabetes and patients with this condition are more likely to develop CVD. The genetic causes of insulin resistance and effects on the vascular system are poorly understood. Our goal is to elucidate the molecular mechanisms of how insulin resistance causes vascular dysfunction using patient-specific induced pluripotent stem cells (iPSCs) differentiated into endothelial cells. Unlike previous models, iPSC-derived endothelial cells (iPSC-EC) are ideal because they generate abundant patient-specific tissue sample.

Hypothesis: iPSC-EC derived from patients with insulin resistance are dysfunctional

Methods & Results: Insulin resistant and control patient peripheral blood mononuclear cells were reprogrammed into iPSCs and subsequently differentiated into endothelial cells as confirmed by qPCR and flow cytometry. iPSC-EC stimulation with tumour necrosis factor TNFα was used to model insulin resistance. When treated with TNFα, iPSC-EC had increased cell adhesion molecule expression and dysfunctional insulin signaling, implying endothelial dysfunction and genes involved in the insulin signaling were downregulated. At baseline, iPSC-EC derived from insulin resistant patients had increased endothelial nitric oxide synthetase (eNOS) expression and phosphorylation and paradoxically, stimulation with insulin did not increase eNOS phosphorylation, suggesting endothelial dysfunction may be caused by an abnormality in this pathway. Indeed, insulin resistant iPSC-EC have a relative impairment of NO release, impaired angiogenesis and increased reactive oxygen species production under conditions of hyperglycemia and inflammation mimicked by TNFα.

Conclusion: iPSC endothelial cells model insulin resistance and endothelial dysfunction. eNOS expression and endothelial cell function is abnormal in insulin resistant patients. The underlying mechanisms merits further investigation.

4. Accelerated aging in lethal dilated cardiomyopathy

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Introduction: Duchenne muscular dystrophy (DMD) is a lethal X-linked recessive disease that results from mutations in the dystrophin gene and is the most common myopathic disease in humans with a prevalence of one in every 3500 males. Dystrophin is crucial for the formation of a dystrophin-glycoprotein complex, which connects the cytoskeleton of a muscle fiber to the surrounding extracellular matrix in both skeletal and cardiac muscles. In the heart, loss of dystrophin leads to increased fibrosis and death in the third decade of life due to dilated cardiomyopathy. Previously we showed that cardiomyocytes in patients with DMD had telomeres 50% the length of unaffected controls. Notably, shortening was not observed in smooth muscle cells.

Hypothesis: We hypothesize that contractile defects due to dystrophin deficiency drive telomere shortening and result in metabolic compromise and cell death due to inhibition of mitochondrial function and biogenesis.

Method: To study telomere shortening in the course of disease progression, we differentiated DMD human induced pluripotent stem cell line into beating cardiomyocytes (hiPSC-CMs) as a model.

Results: We observed aberrant calcium handling and decreased contractility using bioengineered micropatterned hydrogel traction force microscopy. Here we present new evidence where aberrant contraction results in telomere deprotection and resection in in non-dividing hiPSC-CMs. Induction of DNA damage response culminated in mitochondrial dysfunction and apoptosis.

Conclusions: Patient hiPSC-CMs recapitulate in 30 days the telomere shortening that occurs in 30 years of life in DMD patients and this technology enables the study of cause and effect and tests of interventions.

5. Deep learning of cardiac MRI data shows genome-wide associations for bicuspid aortic valve in the UK Biobank

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With a prevalence of 1-2% in the general population, bicuspid aortic valve (BAV) is the most common congenital heart disease (CHD) and accounts for more morbidity and mortality than all other CHDs

combined. Although reported heritability estimates are as high as 89%, specific molecular genetic markers of BAV risk remain to be discovered.

Here, 9,802 magnetic resonance imaging (MRI) sequences from the UK Biobank were analyzed to classify aortic valves as either BAV or normal (tricuspid) using a deep learning algorithm (https://www.biorxiv.org/content/early/2018/06/05/339630). A genome-wide association study was conducted on the subset of unrelated European-ancestry participants (595 BAV, 9207 tricuspid aortic valve) using PLINK. External validation of the genetic findings was performed on imputed data from a case-control study of up to 2594 cases representing eight CHD types and 5159 healthy subjects from the Wellcome Trust Case Control Consortium 2 (WTCCC2).

Markers at three loci displayed statistically significant associations with BAV, including a variant on chromosome 12 near IGF1 and LINC00485 (rs146357447, 12:103025165, MAF=1.3%, odds ratio (OR): 3.2, p=6.1e-9), an intronic locus on MIR28 (rs550423221, 3:188508236, MAF=0.2%, OR=9.6) and a marker on chromosome 2 (rs192377594, 2:140363901, MAF=0.6%, OR=4.1). In the external dataset rs146357447 was associated with risk for atrial septal defect (OR=1.9, p=0.033), and the MIR28 marker displayed an association with non-specific/mixed CHD (OR=1.9, p=0.013).

The results suggest novel candidate loci as determinants of genetic risk for BAV in the general population, and indicate a shared genetic architecture with different types of CHD.

6. Mechanisms leading to telomere shortening in Duchenne muscular dystrophy cardiomyopathy

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Duchenne muscular dystrophy (DMD) is caused by a lack of dystrophin, and DMD patients face muscle degeneration that culminates in loss of respiratory muscle strength and dilated cardiomyopathy. Dystrophin serves several distinct functions, including maintenance of cell membrane integrity, structural support to the extracellular matrix, and protection against oxidative stress. While the function of dystrophin is well understood, the molecular events that lead to cardiomyocyte cell death remains to be explored. A severe limitation in the field is that the mouse model lacking functional dystrophin (mdx) does not exhibit cardiac symptoms seen in humans. For unknown reasons, mice maintain much longer telomeres than humans. When our lab generated mice with "humanized" telomeres, by crossing mdx mice with mice lacking telomerase, dilated cardiomyopathy as seen in patients was recapitulated. Preliminary data suggests that contractile stress due to the lack of dystrophin leads to a pathogenic condition of oxidative stress, telomere shortening, and mitochondrial dysfunction. Using human iPS cells derived from DMD patients, we observe telomere shortening in human DMD cardiomyocytes compared to CRISPR-corrected controls. Understanding the earliest molecular events that trigger pathogenesis will enable identification of strategies for intervention to ameliorate all forms of DMD caused by a wide range of mutations in dystrophin.

7. Lipid Peroxidation Decreases Mitochondrial Dynamics and Impairs Bioenergetics in Children with Right Ventricular Failure due to Congenital Heart Disease

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Background: In complex congenital heart disease patients such as those with tetralogy of Fallot, the right ventricle is subject to pressure overload stress leading to right ventricular hypertrophy (RVH) and eventually right ventricular failure (RVF). The role of chronic oxidative stress, in particular lipid peroxidation, in RVH and RVF in congenital heart disease is unknown.

Methods: Oxidative stress, mitochondrial structure, dynamics and respiration were assessed in the right ventricle of patients with congenital heart disease and in a murine model of RVH and RVF. The effect of 4-hydroxynonenal (4HNE; byproduct of lipid peroxidation) and carvedilol on mitochondrial dynamics and respiration was assessed in cardiomyocytes.

Results: Increased lipid peroxidation was associated with lower maximal respiration in patients with RVF [RVH 390.2±20.17 vs. RVF 204.1±34.73 pmol/(sec*ml), p=0.0032]. Our murine model of RVH and RVF mimicked the patient data and also demonstrated (i) decreased mitochondrial fission (DRP1, MFF) and fusion (OPA1) protein expression; (ii) decreased mitochondrial DNA content by 61%; and (iii) fragmented mitochondrial network in RVF. Cardiomyocyte treatment with 200 μM 4HNE decreased mitochondrial dynamics protein expression, increased leak respiration by 33%, and abolished ADP-mediated respiration. The β-blocker and antioxidant, Carvedilol prevented DRP1 and MFF from decreasing in response to 4HNE.

Conclusion: Mitochondria are the largest source and target of lipid peroxidation products. Lipid peroxidation in RVF is associated with impaired mitochondrial dynamics and membrane damage leading to reduced energy generation. Carvedilol improved mitochondrial fission, raising the potential for its use in RVF in children with congenital heart disease.

8. Impaired Bioenergetics in Right Ventricular Failure is associated with Lipid Peroxidation and Decreased Mitochondrial Dynamics

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Background: The right ventricle (RV) is uniquely at risk in patients with complex congenital heart disease (CHD). Despite successful repair, the RV is subject to pressure overload stress, leading to right ventricular hypertrophy (RVH) and eventually right ventricular failure (RVF). The critical role of mitochondrial dynamics in the development of RVH and RVF in CHD is unknown. As a major source of reactive oxygen species, mitochondria are susceptible to oxidative damage. We hypothesized that impaired energy generation in pressure overload-induced right ventricular failure is accompanied by blunted mitochondrial dynamics and increased lipid peroxidation.

Methods: Mitochondrial structure and function were assessed in RV tissue resected from patients with CHD and a murine model of RVH and RVF. The role of oxidative stress was assessed in the development of mitochondrial dysfunction.

Results: Patients with RV outflow tract obstruction leading to RVF demonstrated lower maximal respiration than those with moderate RVH. [390.2±20.17 vs. 204.1±34.73 pmol/(sec*ml), p=0.0032]. To understand the mechanism of impaired respiration, we used a murine model of RVH and RVF. RVF was characterized by decreased maximal respiration [Sham 744.3±49.3 vs. moderate RVH 513.2±112.6 vs. RVF 306±40.18, p<0.0001 (vs. Sham) and p=0.0394 (vs. RVH)], and mitochondrial fission (DRP1, MFF) and fusion (OPA1) compared with RVH. RVF exhibited increased lipid peroxidation and irregularly shaped mitochondria.

Conclusion: Pressure overload-induced RVF has impaired mitochondrial respiration and dynamics. These were associated with increased lipid peroxidation, which may promote the dysfunction.

9. Modeling the Mitral Valve

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This work is concerned with modeling and simulation of the mitral valve, one of the four valves in the human heart. The valve is composed of leaets, the free edges of which are supported by a system of chordae, which themselves are anchored to the papillary muscles inside the left ventricle. First, we examine valve anatomy and present the results of original dissections. These display the gross anatomy and information on fiber structure of the mitral valve. Next, we build a model valve following a design-based methodology, meaning that we derive the model geometry and the forces that are needed to support a given load, and construct the model accordingly. We incorporate information from the dissections to specify the fiber topology of this model. We assume the valve achieves mechanical equilibrium while supporting a static pressure load. The solution to the resulting differential equations determines the pressurized configuration of the valve model. To complete the model we then specify a constitutive law based on a stress-strain relation consistent with experimental data that achieves the necessary forces computed in previous steps. Finally, using the immersed boundary method, we simulate the model valve in uid in a computer test chamber. The model opens easily and closes without leak when driven by physiological pressures over multiple beats. Further, its closure is robust to driving pressures that lack atrial systole or are much lower or higher than normal.

10. Identifying the Novel Role of a Presenilin-2 Mutation in Arrhythmogenicity using Patient Specific Induced Pluripotent Stem Cells Derived Cardiomyocytes

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Arrhythmia is a major cause of sudden cardiac death and affects more than 14 million Americans. In familial cases, disease-causing mutations are expected to be found in genes encoding proteins that regulate membrane potential or calcium kinetics. Through genetic testing, we identified a ventricular fibrillation patient with family history of cardiovascular diseases that does not carry any disease-causing mutation in the arrhythmia-related genes. This patient, however, carries a previously reported dilated cardiomyopathy mutation (S130L) in presenilin-2 (PSEN2). To understand if this mutation can contribute to arrhythmia, the beating regularity and action potential morphology of cardiomyocytes derived from the patient-specific induced pluripotent stem cells (hiPSC-CMs) were assessed by

fluorescence-based membrane potential imaging. Up to 30% of these hiPSC-CMs demonstrated delayed after-depolarizations (DAD) and irregular beating pattern, which were prevented by correcting this PSEN2 mutation through CRISPR/Cas9 genome editing. Interestingly, we were unable to recapitulate the arrhythmic propensity by introducing this mutation into two healthy control hiPSC-CM lines, until we inserted another modulator mutation in histidine-rich calcium binding protein (HRC) that was also found in the patient, suggesting PSEN2 mutation is providing the substrate for arrhythmia induction. Mechanistically, compromised intracellular calcium removal was detected in S130L-PSEN2 hiPSC-CMs, which was concordant with a reduction in SERCA protein expression. Compromised calcium removal also led to elevated diastolic calcium and activated calcium/calmodulin-dependent protein kinase II (CAMKII), indicated by its enhanced phosphorylation. As a result, ryanodine receptor was hyper-phosphorylated at the CAMKII site (ser2814), which could facilitate calcium leakage from the ryanodine receptor and contribute to the occurrence of DAD. Collectively, our findings reveal a previously unknown function of PSEN2 in cardiomyocyte function and suggest that this PSEN2 mutation can compromise normal intracellular calcium cycling and contribute to arrhythmia through activating CAMKII.

11. Novel Alpha-actinin 2 mutations are associated with cardiomyopathy and hypertrophy in human cardiac tissue and iPSC-derived cardiomyocytes

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In cardiac and skeletal muscle, alpha-actinins are critical cytoskeletal proteins that anchor actin filaments within the sarcomere. Mutations in ACTN2 have been associated with cardiac abnormalities. However, the mechanisms behind how ACTN2 mutations lead to cardiac dysfunction remain poorly understood. The aim of this study was to investigate the effects of two novel ACTN2 mutations on cardiac and skeletal muscle phenotypes in human tissue and patient-specific iPSC-derived cardiomyocytes.

We identified patients in the Stanford Center for Inherited Cardiovascular Disease database with rare or novel ACTN2 variants using a custom mutation pipeline optimized for rare variant discovery. We identified one patient homozygous for a stop-gain mutation (p.Q860X) in ACTN2 and a family with an exon 8-10 deletion. In heart transplant tissue of the homozygous patient, we observed mild hypertrophy and interstitial fibrosis. There was no variation in ACTN2 protein expression, indicating absence of nonsense mediated decay. siRNA knock down of ACTN2 in neonatal rat ventricular cardiomyocytes and a human myoblast cell line resulted in dramatic changes in cell size and morphology. Patient-derived iPSC-cardiomyocytes were hypertrophic, displayed sarcomeric structural disarray and had a slower contractile velocity. Using Co-Immunoprecipitation for ACTN2, followed by mass-spectrometry, we identified a missing protein-protein interaction with AKAP9 in the patient with the truncated ACTN2 variant.

The molecular effects of ACTN2 on a cellular level and how it causes cardiomyopathy has not been fully elucidated. Here, we provide evidence that two loss of function genetic variants in ACTN2 are associated to contractile dysfunction and lead to cardiac abnormalities.

12. Development Of A Genome Base Editing Approach For The Treatment Of Genetic Dilated Cardiomyopathy In Vivo

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Rationale: The recently developed CRISPR–Cas-based genome editing holds great promise for targeting genetic disorders, such as cardiomyopathies. Adenine base editors (ABE) enable efficient adenine-to-guanine base (A·T to G·C) conversion in post-mitotic cells independent of dsDNA break formation and homology-directed repair (HDR). Familial dilated cardiomyopathy arising from C-to-T point mutations, such as TNNT2R173W, can potentially be corrected by an ABE base editing system in vivo.

Objective: As precise correction of disease-causing mutations in adult tissues in vivo is challenging, we are establishing a versatile adeno-associated viral (AAV) platform for ABE-dependent base editing in adult animals.

Methods and Results: We engineered a dual trans-splicing AAV vector system encoding the newly described xCas9 and SpCas9-NG variants that recognize a wider range of protospacer adjacent motif (PAM) sequences that is compatible with gRNA-directed targeting of the TNNT2R173W mutation. This system allows splitting of the fusion ABE-Cas9 protein into two parts, thereby circumventing the limited cargo capacity of AAV vectors. Combined with an AAV vector expressing a targeting gRNA, a three-vector base editing system was developed and validated in vitro. In silico analysis of the TNNT2R173W site has shown that it is amenable to adenine base editing with multiple gRNAs. We are currently testing this system in patient-specific induced pluripotent stem cell-derived cardiomyocytes (iPSC-CMs) harboring the TNNT2R173W mutation in vitro, and in a transgenic mouse model of dilated cardiomyopathy carrying the same mutation in vivo.

Conclusions: The therapeutic potential offered by this AAV-ABE system holds promise in future testing on transgenic mice models of dilated cardiomyopathy. Our system addresses two key challenges in therapeutic base editing, namely target recognition and in vivo delivery of the large ABE-Cas9 fusion gene, by introducing two engineered AAV-ABE variants with relaxed PAM recognition and a modified split-AAV platform respectively.

13. Regulatory Mechanism of LMOD1 Association with Coronary Artery Disease

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Atherosclerotic coronary artery disease (CAD) continues to be the leading cause of mortality and morbidity worldwide, with an estimated 40% of one's lifetime risk attributed to genetic factors. Meta-analysis of genome-wide association studies implemented to identify this risk in human populations, has now identified rs2820315 (P=7.7E-10; OR=1.05), in the smooth muscle cell-restricted gene, Leiomodin 1 (LMOD1), as the leading genetic polymorphism associated with CAD. However, the causal mechanism by which this polymorphism is responsible for predisposition to CAD remains to be identified. Expression quantitative trait loci (eQTL) mapping in GTEx and STARNET databases revealed that carriers of the risk allele at rs2820315 display significantly attenuated LMOD1 expression

in vascular tissues than carriers of the ancestral allele. Epigenomic profiling and fine-mapping analyses identified rs34091558 as a top candidate causal variant in high linkage disequilibrium (r2=0.94) with the lead variant. To determine the mechanism responsible for reduced LMOD1 expression, we performed position weight matrix (PWM) motif analyses and found that rs34091558 disrupts the binding site of a transcription factor called forkhead box O3 (FOXO3). Subsequent chromatin immunoprecipitation and reporter assays demonstrated reduced FOXO3 binding and transcriptional activity by the risk allele in cultured HCASMCs. Platelet-derived growth factor BB (PDGF-BB) stimulation also significantly reduced LMOD1 expression coincident with FOXO3 knockdown. Finally, both gain and loss-of-function for FOXO3 and LMOD1 in HCASMC delineated a regulatory circuit by which LMOD1 regulates SMC proliferation, migration and contraction, characteristic features of atherosclerotic lesion progression. Taken together, these results provide compelling functional evidence that: 1) rs3091558 is associated with reduced LMOD1 expression, 2) this reduction appears to be mediated through the inhibition of FOXO3 binding and 3) changes in vessel wall processes through LMOD1 dysregulation may partially explain the heritable risk for CAD.

14. Exosomal miRNA Profiles of Endothelial Cells and Pericytes in Pulmonary Niche on an Organ-on-a-chip Model

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Pulmonary arterial hypertension (PAH) is a disorder effecting pulmonary circulation. PAH is a result of reduced blood flow due to partial or entire elimination of microvessels because of attenuated recruitment of pericytes (Pc), in addition to genetic aberrations and environmental factors including endothelial cell (EC) death. To understand the cross-talk between cells, extracellular vesicles (EVs), are being increasingly investigated since their shedding from cells occurs in response to global and local changes in their environment and can be used to monitor cargo molecules carried by them, in response to these conditions in vitro and in vivo. Exosomes involve in this communication, which will impact the recipient cell's fate, packed with microRNAs (miRNAs).

Motivated by this strong relationship between exosomes and their regulatory behaviors, we hypothesize that shear stress on ECs can trigger exosome mediated Pc recruitment during PAH progression through WNT family. The objective of this work is to determine molecular changes during Pc recruitment of ECs leading to PAH via deciphering exosomal miRNA profiling.

We cultured patient derived healthy and PAH EC and Pc cell lines in static conditions as monocultures to draw a baseline of exosomal miRNA profiles. We profiled secreted exosomes from these cultures. And collected RNA for sequencing, where PAH EC and Pc cultures had more exosomes and exosomal RNAs, compared to healthy donor cultures (n=3). We will report on cultures in static co-culture models, and on an in-house developed dynamic organ-on-a-chip model to decode their exosomal cross-talk through exosomal miRNA profiling.

15. Modelling microenvironmental mechanical properties in Duchenne Muscular Dystrophy iPSC-derived cardiomyocytes

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Duchenne Muscular Dystrophy (DMD) is an X-linked disease affecting ~1:3500 boys per year that culminates in heart failure in early adulthood. DMD results from >200 possible genetic mutations on dystrophin. The lack of dystrophin disrupts the anchoring of the cell sarcomere to the extracellular matrix (ECM), affecting cardiomyocyte contraction. With disease progression, tissue increases in stiffness due to fibrosis and changes in ECM composition in accordance with a dilated cardiomyopathy phenotype. We hypothesize that this entails a positive feedback loop involving multiple mechanosensing pathways. Here, we use a single-cell platform to model the fibrotic remodelling in DMD. We measure the force production of single human induced pluripotent stem cell derived cardiomyocytes (hiPSC-CMs) on hydrogel substrates with a stiffness matching that of healthy or fibrotic tissue. Furthermore, we enhance the hiPSC-CMs structural maturity and standardize our measurements by patterning single iPSC-CMs in an elongated 1:7 aspect ratio using microcontact printing of ECM proteins. We compute the contractile strength as a function of bead displacement in the hydrogel substrate using Digital Image Correlation (DIC) and Fourier Transform Traction Cytometry (FTTC). We show that DMD hiPSC-CMs have a dramatically reduced ability to produce force on stiffer substrates compared to their isogenic controls. This loss of function correlates with an increase in reactive oxygen species (ROS) and mitochondrial dysfunction. The effect of stiffness in this difference in contractile function uncovers a potent role of mechanosignaling mediated by the dystroglycan complex. This platform will increase our understanding of the biophysics underlying cardiomyocyte mechanosensing.

16. High-throughput phenotypic screening using induced pluripotent stem cell derived cardiomyocytes identifies compounds that rescue genetic dilated cardiomyopathy contractility performance

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Introduction: Familial dilated cardiomyopathy (DCM) is a leading cause of heart failure. To date, there is still a large gap in our understanding of the molecular events and signaling pathways that lead from a mutation to diverse disease phenotypes, and disease-modifying therapies are lacking. The development of induced pluripotent stem cell (iPSC) technology has enabled new opportunities to identify disease-modulating therapeutics and empowers comparatively rapid drug screening for human genetic diseases such as DCM.

Methods: iPSCs were generated from three DCM patients harboring a pathogenic mutation in the TNNT2 gene (p. Arg173Trp; TNNT2R173W) and differentiated towards cardiomyocytes (iPSC-CMs). We performed a primary phenotypic screening using high-throughput contractility assays in iPSC-CMs monolayers, and further validated our finding at the single cell and 3D engineered heart tissue levels.

Results: We tested a small molecule library of 200 well-characterized protein kinase inhibitors and identified two compounds that rescued the contractile deficit of TNNT2R173W iPSC-CMs. We pursued

two hits for further studies and demonstrated that these two kinase inhibitors when combined provided a synergistic effect.

Conclusions: Here we determined the feasibility of performing a primary phenotypic screen in DCM iPSC-CMs and demonstrated that small-molecule discovery using an iPSC-based disease model can identify candidate drugs for potential therapeutic intervention. The identification of compounds that increase contractility in DCM iPSC-CMs could yield novel therapies for genetic DCM.

17. Stanford Center for Undiagnosed Diseases: Unusual Cardiovascular Phenotypes and Precision Medicine

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Introduction: The Stanford Center for Undiagnosed Diseases (CUD) is a clinical site of the Undiagnosed Diseases Network (UDN). The CUD enrolls patients with rare, undiagnosed diseases across various clinical indications including those with undiagnosed cardiovascular disease. The mission of the UDN is to improve knowledge of the molecular etiology of disease and to develop bioinformatic tools to support precision medicine. Since 2015, the CUD has enrolled 153 of 359 patients who applied (42.6%). The primary phenotype is cardiovascular in twelve patients (7.8%). To date, evaluation in the CUD has yielded a confirmed diagnosis in two of twelve patients with cardiovascular phenotypes and 29 of 106 patients overall.

Case Report: A 32-year-old female presented with a 10-year history of persistent intermittent chest pain and troponin elevation (>20 mg/dL) of unclear etiology, borderline left ventricular (LV) ejection fraction (55%), hypokinesis of the LV wall, mesocardial myocardial fibrosis, mild endothelial dysfunction, and sinus tachycardia. Repeated coronary catheterizations showed no epicardial coronary artery disease and PET CT showed no evidence of myocardial inflammation. Her extensive clinical workup had failed to identify a unifying diagnosis. Prior clinical exome sequencing was recommended, but coverage was denied by the patient's insurance provider. The patient was thus referred to the CUD. Evaluation in the CUD included clinical exome sequencing. A heterozygous pathogenic nonsense variant was identified in the DSP gene (c.1273C>T; p.Arg425Ter). Pathogenic variants in DSP are associated with both dilated cardiomyopathy and arrhythmogenic right ventricular cardiomyopathy. While the patient's cardiac imaging did not reveal any LV enlargement or right ventricular involvement, case reports of patients with DSP nonsense variants have described similar presentations with elevated troponin, chest pain, and fibrosis in the absence of ventricular enlargement. Thus, we considered the DSP variant to be diagnostic. The patient has since established care with a heart failure cardiologist and electrophysiologist for ongoing surveillance and sudden death risk stratification.

18. Modeling hypertrophic cardiomyopathy caused by mutations in beta-myosin using human induced pluripotent stem cell derived cardiomyocytes (iPSC-CMs)

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Hypertrophic cardiomyopathy affects 1:500 Americans and is commonly caused by mutations in beta-myosin heavy chain, the main motor protein responsible for contraction of human ventricles. This protein is arranged in hierarchical structures called sarcomeres and myofibrils that allow for coordinated contraction of cardiomyocytes. We have used CRISPR-Cas9 gene editing to insert mutations (P710R and D239N) into hiPSCs that we subsequently differentiate into cardiomyocytes. We used micropatterning techniques to promote cell and myofibril alignment and contractile function. Transmission electron microscopy has confirmed microstructural changes in the sarcomeres and myofibrils of cells containing these mutations, and traction force microscopy has revealed differences in force generation at the single cell level. We have also measured altered signaling through MAPK pathways that may regulate hypertrophy. These cellular level experiments provide an important complement to molecular studies of these mutations, and we are developing models that will allow us to predict how changes in force and kinetics might translate across scales and contribute to cellular and tissue remodeling.

19. Parental disease and over transmission of genetic risk for diabetes are related to Congenital Heart Disease in offspring

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Maternal diabetes and elevated blood glucose during pregnancy are the most recognized risk factor for Congenital heart disease (CHD). However, the contribution or relationship to other maternal conditions such as cancer, lung, and neurological diseases, in addition to any paternal contribution to CHD risk in offspring are largely unexplored. The relationship of parental conditions was assessed using the 500,000 participants of the UK-biobank among which, 2006 adults have CHD. in the UK-biobank, a survey uniformly ascertained the Self-reported parental history of diseases (including diabetes, heart disease (unspecified), chronic bronchitis, cancer, Parkinson and Alzheimer disease, and severe depression) at the time of enrollment.

In a separate study of 850 trios (2550 individuals) with array genotyping data from the Pediatric Cardiology Genomic Consortium (PCGC), the deviation in the transmission of the genetic risk from parent to child for each condition was tested using the transmission disequilibrium test based on the Polygenic risk score (pTDT). In the UK Biobank CHD risk was associated with history of heart diseases in both parents [OR=1.41(1.19–1.66), p=5x10-05], chronic bronchitis/emphysema in mother or father [OR=1.22(1.07–1.39), p=2.8x1003], and Alzheimer in mother [OR=1.26(1.08 – 1.47), p=2.2x10-03] but not with the parental history for diabetes. Among the trios, we found a deviation in the transmission of PRS from parent to child for diabetes [mean(pTDT)=0.15(0.05–0.25) p=3x10-03], chronic obstructive pulmonary diseases (COPD) [mean(pTDT)=0.13(0.02–0.24) p =0.02], and Alzheimer disease [mean(pTDT)=0.11(0.06 – 0.16), p=0.02]. Our findings suggest that lifetime risk of heart, lung

and Alzheimer diseases in the parents may be related to the risk of CHD in offspring. Additionally, risk alleles for diabetes, COPD, and Alzheimer in the parents were observed to be over-transmitted from parent to child, suggesting that these novel inherited risk alleles for CHD may not be limited to a maternal or in utero metabolic effect.

20. Transgenic Mice Lacking BMPR2 in Smooth Muscle Cells have Persistent Pulmonary Hypertension Related to Impaired Contractility, Heightened Proliferation and Resistance to Apoptosis of PASMC

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Mutations in bone morphogenetic protein receptor 2 (BMPR2) are associated with idiopathic pulmonary arterial hypertension (PAH), but the link between loss of BMPR2 and the pathogenesis of PAH remains unclear. To understand the role of BMPR2, we generated mice lacking BMPR2 in vascular smooth muscle cells (SMC) with reporter (SM22 Cre+R26R+Bmpr2-/-). The penetrance of a ventricular septal defect was 50%. SM22 Cre-Bmpr2-/- survivors were compared to controls in room air, three weeks of hypoxia and following four weeks recovery in room air (n = 4-8). While female groups were similar under all conditions, mutant males showed reduced hypoxia-induced vasoconstriction and developed less severe PH following chronic hypoxia, judged by right ventricular systolic pressure. There was, however, more persistent PH following recovery, associated with sustained muscularization of distal pulmonary arteries (PA). We found that PASMCs from male SM22 Cre-Bmpr2-/- mice vs. controls were less contractile in response to angiotensin II (4μM) and showed heightened proliferation and resistance to apoptosis. We observed a similar phenotype in human PASMC where BMPR2 was knocked down by siRNA, related to increased β-arrestin2 and active β-catenin and reduced active RhoA and Rac1. Reducing β-arrestin2 restored the contractile phenotype and attenuated the heightened proliferation phenotype. Interestingly, tissue staining revealed heightened expression of β-arrestin2 and active β-catenin in PASMCs of PAH patients with BMPR2 mutation vs. controls. Our study relates loss of BMPR2 in PASMC to impaired PA contractility and heightened PASMC proliferation. The mechanism is consistent with dysregulation of tandem β-catenin and RhoA signaling in response to BMPR2 stimulation.

21. Endothelial expression of constitutively active Notch4 initiates brain arteriovenous malformation involving a nitric oxide synthase-mediated molecular mechanism

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Arteriovenous (AV) malformations (AVMs) are characterized by a nidus of enlarged, tangled vessels that shunt blood directly from arteries to veins, displacing intervening capillaries. Mechanisms underlying AVM pathogenesis are poorly understood, hindering therapeutic development. Endothelial expression of constitutively active Notch4 (Notch4*) initiates brain AVMs in mice through enlargement of capillary-like vessels without an increase in endothelial cell number or proliferation. Instead, initial enlargement of AV shunts correlates with area expansion of individual endothelial cells, suggesting that aberrant vasodilation may play a role in early stages of AV shunting. We hypothesized that Notch4* disrupts endothelial nitric oxide synthase (eNOS) signaling, permitting vessel enlargement and AV shunting. Consistent with this, pharmacological inhibition of nitric oxide synthase (NOS) by administering the NOS inhibitor NG-nitro-L-arginine (L-NNA) or

eNOS gene deletion decreased brain AV shunt diameter, severity of brain AVM-associated pathologies, and illness progression in mice expressing endothelial Notch4*. Furthermore, pial arteries isolated from Notch4* mice exhibited decreased arterial tone compared to controls, and this was abolished by L-NNA. Interestingly, endothelial Notch4* expression did not result in detectable changes in nitric oxide (NO) production by 4-Amino-5-Methylamino-2',7'-Difluorofluorescein (DAF-FM) Diacetate in the brain or by cyclic guanosine monophosphate (cGMP) production, a surrogate for aortic NO production. Instead, NOS-dependent superoxide production was elevated in Notch4* brains at the initial stages of AV shunting formation, as assessed by dihydroethidium (DHE). Administering the superoxide dismutase mimetic 4-Hydroxy-2,2,6,6-tetramethylpiperidine 1-oxyl (Tempol) decreased brain AV shunt diameter, severity of brain AVM-associated pathologies, and illness progression in mice expressing endothelial Notch4*, mirroring the effects of NOS inhibition and eNOS deletion. Our data suggest that endothelial Notch4*-induced brain AVM involves an eNOS-dependent molecular mechanism that upregulates superoxide production.

22. Single cell analysis of smooth muscle cell phenotypic modulation in vivo reveals a critical role for coronary disease gene TCF21 in mice and humans

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In response to various stimuli, vascular smooth muscle cells (SMCs) can de-differentiate, proliferate and migrate in a process known as phenotypic modulation. However, the phenotype of modulated SMCs in vivo during atherosclerosis and the influence of this process on coronary artery disease (CAD) risk are not established. Using single cell RNA sequencing, we comprehensively characterized the transcriptomic phenotype of modulated SMCs in vivo in both mouse and human arteries and found that these cells transform into unique fibroblast-like cells that we term "fibromyocytes". SMC-specific knockout of TCF21, a causal CAD gene, markedly inhibited SMC phenotypic modulation in mice, leading to fewer fibromyocytes within the lesion and the protective fibrous cap. TCF21 expression was also strongly associated with SMC phenotypic modulation in diseased human coronaries. In human CAD-relevant tissues, TCF21 expression was associated with decreased CAD risk, establishing a protective role for both TCF21 and SMC phenotypic modulation in this disease.

23. Anti-aging effects of growth hormone-releasing hormone agonist on cardiovascular system in old mice

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Aim: Diastolic dysfunction is a characteristic of aged hearts. The agonists of growth hormone-releasing hormone (GHRH-A) exhibit several favorable effects on heart function. Here we assessed the effects of GHRH-A MR409 on heart function and systemic parameters in aged mice.

Methods and Results: Starting at the age of 15 months, mice were subcutaneously injected daily with MR409 (100ug/mouse) or vehicle (n=7 each). Echocardiography and body weights were measured at baseline and 5 months after treatment. Mice treated with MR409 for 5 months showed significantly improved ejection fraction and attenuated hypertrophy, increased exercise activity, and healthier hair growth in comparison with the controls. No changes in body weight were observed after the treatment. In studies in vitro, senescence levels were detected with β-gal staining in the doxorubicin-treated H9C2 cardiac myoblasts, neonatal cardiomyocytes (NRCM) or endothelial cells (EC) after 2 or 10 passages, respectively. When cultured with MR409, significantly fewer β-gal positive cells were observed as compared with control cells. Cell cycle associated protein p21 was reduced in all MR409-treated cells (H9C2, NRCM and EC). Reactive oxygen species in Dox-treated H9C2 cells were reduced when cultured with MR409. Electron microscopy showed that mitochondrial morphology was preserved in the Dox-treated H9C2 cells upon cultures with MR409 while it was damaged in control cells after Dox treatment. MR409 also improved cellular ATP production and oxygen consumption rate of Dox-treated H9C2.

Conclusion: GHRH-A can rejuvenate aged mice in aspects of heart function, exercise capacity, hair growth, cellular energy production and senescence biomarkers.

24. Pericytes differentiate into smooth muscle cells through CXCL12 activation in hypoxia induced pulmonary hypertension

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Background: Pericytes are specialized perivascular cells that directly interact with endothelial cells. They are also multipotent cells-like. Little is known regarding the contribution of pericytes to pulmonary vascular diseases such as pulmonary arterial hypertension (PAH), a disorder associated with abnormal vascular remodeling of microvessels secondary to smooth muscle cell (SMC) accumulation. It is also unclear whether the overabundant SMCs are of pericyte-origin and what underlying mechanisms drive that transition.

Materials and Methods: Transcriptome analysis was performed on human healthy and PAH pericytes and compared against healthy and PAH SMC. In order to trace pericyte lineage during hypoxia induced PH, we utilized fate mapping using the NG2-tdTomato(NG2-tdT) murine model which can selectively mark pericytes with red florescence. FACS was used to sort tdT positive cells followed by bulk and single cell RNA-seq analysis.

Results: Transcriptomic analysis demonstrated that genetic landscape of PAH pericytes was homologically similar to that of PAH SMCs. Analysis of NG2-tdT murine lung sections revealed that pericytes relocate from alveolar capillaries to the precapillary arterioles and expressed smooth muscle myosin heavy chain (SMMHC), a marker of mature SMCs. Bulk RNA-seq analysis of pericytes sorted from 21-day hypoxia revealed strong upregulation of cell motility related genes compared to normoxia. Further analysis using single cell RNA-seq revealed that pericytes under hypoxia can be distinguished into 8 different clusters. Among them, the SMC-like cluster was the most abundant and within this cluster, individual cells had elevated level of C-X-C motif chemokine 12 (CXCL12). Analysis of in both human PAH serum samples (N=83) and pericytes demonstrated a significant elevation in CXCL12. Moreover, overexpression of CXCL12 in healthy human lung pericytes produced a SMC like phenotype associated with greater contractility and reduced association to endothelial cells in matrigel tube formation assay.

Conclusion: Our results suggest that pericytes contribute to muscularization of distal precapillary vessels in response to hypoxia by differentiating into SMCs via a CXCL12 associated pathway. Our findings contribute to a better understanding of pericyte biology and identify pericytes as a potential therapeutic target in both hypoxia induced PH and PAH.

25. GDF 11 is essential for maintaining cardiac function under pressure overload

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Objective: Growth differentiation factor 11 (GDF11) is a member of transforming growth factor β superfamily. The physiological and pathological functions of GDF11 in cardiomyocytes and heart remain unclear. Here we sought to elucidate the cardiomyocyte-specific roles and mechanism of GDF11 in pathological cardiac hypertrophy.

Methods and Results: GDF11 expression was increased in human hearts with dilated cardiomyopathy (DCM) and myocardial infarcted (MI), which was confirmed in mouse models of hearts after transverse aortic constriction (TAC) and MI. GDF11 in heart was mainly derived from cardiomyocytes. Cardiac specific GDF11 conditional knockout (CKO) and control Cre mice were subjected to TAC-mediated pressure overload or MI. Deficiency of GDF11 accelerated cardiac dysfunction and left ventricular dilatation after TAC or MI. More fibrosis and fewer vasculatures were detected in the hearts of CKO mice after TAC or MI as compared with controls. GDF11 overexpression with cardiac injection of AAV9-GDF11 during TAC procedure rescued the detrimental cardiac function of CKO mice. In vitro culture, GDF11 overexpression in CMs resulted in more VEGF secretion. The conditioned medium from GDF11-overexpressed CMs stimulated significantly more tube formation of endothelial cells, which could be blocked by VEGF neutralizing antibody. GDF11 overexpression promoted the phosphorylation of Smad2/3 and Akt/protein kinase B (AKT) in CMs. Inhibition of TGF-β/Smad signal pathway by TGF-β receptor inhibitor (SB431542) blunted the GDF11-induced CM's paracrine effect.

Conclusions: GDF11 functions as an injury-induced cardiokine that stimulates paracrine effect of CMs to protect myocardium from injury.

NOTES

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