National Human Genome Research Institute
Understanding of the Molecular Mechanisms of Cardiovascular Diseases

This innovative laboratory team seeks to identify the molecular, cellular, and genetic mechanisms that cause vascular disorders. In particular, their research focuses on defining the pathways that regulate cell growth in the vasculature, remodel the vasculature after injury, and lead to genetic susceptibility to vascular diseases. Taken together, these studies focus on the molecular genetics of vascular diseases, with an emphasis on cell cycle regulation of proliferation, inflammation, and apoptosis.

Lead Agency:

National Human Genome Research Institute (NHGRI)/

National Institutes of Health

Agency Mission:

The National Human Genome Research Institute (NHGRI) led the National Institutes of Health's (NIH) contribution to the International Human Genome Project, which had as its primary goal the sequencing of the human genome. This project was successfully completed in April 2003. Now, the NHGRI's mission has expanded to encompass a broad range of studies aimed at understanding the structure and function of the human genome and its role in health and disease.

To that end NHGRI supports the development of resources and technology that will accelerate genome research and its application to human health. A critical part of the NHGRI mission continues to be the study of the ethical, legal and social implications (ELSI) of genome research. NHGRI also supports the training of investigators and the dissemination of genome information to the public and to health professionals.

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General Description:

Understanding the Molecular Mechanisms of Cardiovascular Diseases
Cardiovascular diseases are the leading cause of morbidity and mortality in industrialized countries. Most cardiovascular diseases result from complications of atherosclerosis, which is a chronic and progressive inflammatory condition characterized by excessive cellular proliferation of vascular smooth muscle cells, endothelial cells, and inflammatory cells that leads to occlusive vascular disease, myocardial infarction, and stroke. Recent

studies have revealed the important role of cyclins, cyclin-dependent kinases (CDKs), and cyclin-dependent kinase inhibitors (CKIs) in vascular and cardiac tissue injury, inflammation, and wound repair. This research seeks to understand the circuitry of the cyclin-CDK-CKI interactions in normal physiology and disease pathology, providing a better understanding of the molecular mechanisms of cardiovascular diseases. This approach will hopefully lead to the rational design of new classes of therapeutic agents.

Given role of cyclins in vascular health, a major focus of the study is CKIs, which are primarily involved in inhibiting the proliferation of a variety of normal cell types. Previous research identified a particular CKI, known as $p27^{Kip1}$, as a major regulator of vascular cell proliferation during arterial remodeling. In one set of studies, her group found that $p27^{Kip1}$ plays a major role in cardiovascular disease through its effects on the proliferation of bone marrow-derived immune cells that migrate into vascular lesions. To demonstrate whether $p27^{Kip1}$ regulates arterial wound repair, NHGRI Investigators recently subjected $p27^{-/-}$ (homozygous knockout), $p27^{+/-}$ (heterozygous knockout), and $p27^{+/+}$ (wild-type) mice to a wire injury in the femoral artery and examined subsequent cell proliferation and lesion formation. Cell proliferation was significantly increased in the innermost lining of the blood vessels of $p27^{-/-}$ mouse arteries compared with $p27^{+/+}$ arteries. Arterial lesions also were markedly increased in the $p27^{-/-}$ mice compared with those of $p27^{+/+}$ mice. The heterozygous knockout mice ($p27^{-/+}$) had an intermediate phenotype. These findings suggest that vascular repair and regeneration are regulated by the proliferation of hematopoietically and nonhematopoietically derived cells through a $p27^{Kip1}$-dependent mechanism, with immune cells largely mediating these effects.

A related area of study focuses on the structural and functional analysis of a serine-threonine kinase called kinase interacting stathmin, or KIS. A nuclear protein that binds the C-terminal domain of $p27^{Kip1}$, KIS phosphorylates a serine residue at position 10 (Ser 10) in the sequence and thereby promotes its export to the cytoplasm. KIS is activated by mitogens during G0/G1, and expression of KIS overcomes growth arrest induced by $p27^{Kip1}$. Depletion of KIS with small interfering RNA (siRNA) inhibits Ser 10 phosphorylation and enhances growth arrest. In addition, treating $p27^{-/-}$ cells with KIS siRNA causes them to grow and progress to S/G2, similar to control-treated cells, implicating $p27^{Kip1}$ as the critical target for KIS. Previous research cloned and characterized the gene encoding this kinase and is studies are now examining its structure and function, including the transcriptional control of the KIS promoter, the phenotypic consequences of knockout out the KIS gene in mice, and the effect of knock-in mutations at different phosphorylation sites of p27.

NHGRI investigators are also involved in a clinical study of the genetics of restenosis, which is the recurrence of a blockage in an artery after it has been manually reopened with an artificial stent. Restenosis is a major limitation of stent therapy for coronary artery disease. In this study, the investigators are following patients who have received bare metal stents for the treatment of a blocked coronary artery and then comparing the genetic profiles of patients with restenosis with those of patients with no restenosis. The genetic analyses include gene expression profiling, serum proteomics, and genotyping using candidate gene and genome-wide scanning approaches. The goal is to identify

gene, RNA, and protein profiles of patients with recurrent restenosis, so as to advance our understanding of the pathogenesis of this problem and to target potential therapies.

Excellence: What makes this project exceptional?

This project utilizes both cardiovascular and genetic medicine to create innovative therapeutic targets for conditions that affect millions worldwide.

Significance: How is this research relevant to older persons, populations and/or an aging society?

Cardiovascular disease remains the leading cause of death and disability in the elderly population, and cardiovascular risk increases steadily with age.

Effectiveness: What is the impact and/or application of this research to older persons?

Understanding the molecular pathophysiology of vascular diseases, such as in-stent restenosis, is critical to the design and development of novel therapeutics.

Innovativeness: Why is this research exciting or newsworthy?

This research has the potential to identify key genetic variants responsible for cardiovascular inflammation, a wide-spread condition, with the aim of eventually tailoring therapies specifically for each group.