After years of effort, investigators at Vanderbilt and Columbia found the gene for heritable pulmonary hypertension (PH) nearly simultaneously in the year 2000.1,2 Mutations in the so-called bone morphogenetic protein receptor type 2 (BMPR2) are now known to be responsible for about 75% of cases of heritable pulmonary arterial hypertension (PAH).3 In the other 25% of families, either the mutation remains unknown, or it is in the ALK-1 or endoglin genes, SMAD 8 or caveolin 1.4 Undoubtedly, other rarer mutations that cause disease will be found. In most registries, known heritable PAH accounts for about 6% of the population enrolled, so 6 out of every 100 cases.5,6 However, surveys of mutation status in idiopathic PAH have revealed a prevalence of about 10%-20% of BMPR2 mutation carriers.7 Thus, as shown in Figure 1, the largest number of patients with the gene raising the risk of PH is in the idiopathic group, and these actually outnumber the known family cases. One of the scientists vital to the discovery of BMPR2 mutations, William Nichols, PhD, at the University of Cincinnati, now has National Institutes of Health (NIH) funding to obtain DNA and RNA from a large number of patients with idiopathic PAH from multiple centers, to discover the actual prevalence of BMPR2 mutations in a USA cohort, and to look for other genetic associations with PH.

The next phase in our journey with heritable PAH was to look for genetic/genomic associations that might help explain the incomplete penetrance of BMPR2 mutations. For many years, based on discussions at the Evian World Health Organization (WHO) Pulmonary Hypertension Conference, the penetrance of the BMPR2 gene has been estimated to be about 20%. This means that having the mutation is not enough, and that environmental, genetic, or biological differences must have a role. Newer data from the Vanderbilt PAH Registry suggests that the penetrance is about 27%, with a 3:1 female to male ratio, giving females about a 42% penetrance and males 14%.8 Nonetheless, the reduced penetrance gave rise to several years of difficult experiments designed to discover a major second gene that might explain penetrance. Although multiple genes were discovered to have a minor role, no major gene has been found. Genes that have been found to possibly have influence on the likelihood of disease include serotonin transporter, Cyp, angiotensin converting enzyme, and nitric oxide synthase. Undoubtedly, a large number of interacting gene products will have major influence on the development of PAH. Part of the problem is that PAH is not one disease and therefore does not have one etiology. This is a very complex problem to address. It also must include epigenetic and other somatic gene modifications that are not discoverable by germline approaches. The problem is summarized in Figure 2. Although basic important work is progressing on mechanisms that regulate BMPR2 expression and function, the modifier gene approach must await newer modalities such as whole exome and genome sequencing to discover rare variants that may influence development of pulmonary vascular disease. This approach will also discover rare variants that have a Mendelian effect on the development of PH in families that remain with undiscovered mutations.

The current thrust of our work is along 4 lines. The first is the study of the gender difference and sex hormones in PH. The greatest risk for development of either idiopathic or heritable PH is female gender, 3:1 over males. Eric Austin in our laboratory is studying estrogen metabolites and the role of cytochrome enzymes in determining circulating levels of estrogen metabolites and their impact on PH risk. Others are following similar experimental approaches. These studies will certainly yield insights into development of PH and perhaps lead to preventive or management therapies.
The second set of experiments is to determine the relationship of insulin resistance and PAH. These experiments are driven by the increasing recognition that hyperglycemia and features of the metabolic syndrome are common in PAH. Either the pulmonary circulation is remodeled or primed for vascular damage by insulin resistance/hyperglycemia, or PAH may lead to metabolic abnormalities. These experiments are led by Anna Hemnes.

The third approach is to determine how BMPR2 expression is regulated. Because of the increasing data that BMPR2 is downregulated in all form of PAH, Rizwan Hamid in our group is performing experiments to determine the role of splice variants, the role of the normal wild-type allele in PAH, and he is beginning to explore the possibility of stimulating BMPR2 expression to normalize the vasculature in PAH or those at risk of PAH.

Finally, James West is heading a translational study looking at the enzyme ACE2. Preliminary data show that ACE2, which converts angiotensin 2 to angiotensin 1-7 is protective in the mouse model of BMPR2-driven PAH, and normalizes cell culture function of cytoskeletal abnormalities. Angiotensin 1-7 is an antiproliferative protein that has generally opposite effects of angiotensin 2. These studies will include a small Phase II human trial.

Thus, our journey at Vanderbilt can be divided into 3 phases. The first was the struggle to identify the genetic mechanism for inherited PH. The second was to assess associations with genes that might influence BMPR2 expression as full-blown clinical PH. The third, current phase is the study of intracellular pathways that are intimately involved in PH. These pathways are sex hormones, glucose metabolism, and neurohumoral effects of the angiotensin system. In addition we continue to investigate what regulates BMPR2 expression in hopes of ultimately modifying it to treat disease.

References