Editorial

Genomics and Complex Neonatal Disorders: Maybe We're Getting Somewhere

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Slowly, but surely, the promise of the genomic revolution is being fulfilled in neonatal medicine. Several recent observations have shown linkages between genome-based discoveries and a deeper and more specific understanding of the pathophysiology of complex disorders arising in the neonatal period. Several recent reports have identified single-nucleotide polymorphisms (SNPs) occurring in either the coding part of a gene of putative interest or in its promoter region. These identified SNPs are then associated with an increased or decreased likelihood of developing a particular disorder, or predicting a specific response to a given therapy. It is gratifying to begin to unravel clinical mysteries, but it is even better that the results may yield more specific therapies, or therapies more specifically used, than those currently available.

Lorenz et al.,[1] wondered if the known risk factor of maternal urogenital infection for the problem of preterm birth could be related to variability in the innate immune response that is mounted against Gram-negative bacteria. Utilizing a known polymorphism in the gene that encodes for the Toll-like Receptor-4 (TLR-4) protein, which mediates the innate immune response to Gram-negative infection, the investigators sought to identify an association between this polymorphism, the presence of which leads to a decreased expression of TLR-4 protein, and preterm birth. Focusing on a population of premature infants in the relatively homogenous population of Finland, they demonstrated that 24% of singleton premature infants and their mothers, compared with 15% of term infants and their mothers, carried the TLR-4 variant. The rationale for their hypothesis was that these women and their fetuses may have had reduced capacity to fend off Gram-negative ascending urogenital infections with a resultant increased risk for preterm labor. The positive and negative predictive values arising from this single association are poor, but if similar genomic variations could be linked to preterm labor and delivery, it might then be possible at the beginning of pregnancy to identify women at particularly high risk of spontaneous preterm birth associated with low-grade infections and manage their pregnancy differently. Although not addressed by Lorenz et al.,[1] it will be interesting to determine if the same allelic variation in the TLR-4 gene is nonrandomly associated with other conditions in premature infants, such as chronic lung disease or Gram-negative infection.

Mutations in the surfactant protein-B (SP-B) promoter region provide another example of a linkage between a neonatal disorder and genomic variation. Mutations have been identified that result in reduced expression or an inability to increase the production of this crucial protein. These mutations result in a deficiency that is less severe than the rare complete deficiency of SP-B, a lethal disorder. Some preterm or borderline-term infants may develop respiratory distress syndrome (RDS) because of the presence of a mutation conferring a decreased, inactive, or unstable form of an SP-B. Animal models that are heterozygous for SB-P deficiency show a reduced ability to withstand a ''second hit'' of hyperoxia. Hyperoxic exposure contributed to a diminished total SP-B and resulted in more rapid mortality in heterozygotes than in wild-type controls.[2] Within the large population of preterm and near-term infants who develop RDS, some of the risk for developing RDS could be explained by genomic-based variation in the capacity to regulate the production of SP-B. Findings in the animal models also amplify the results of interesting case reports linking severe deficiency in SP-B to persistent clinical respiratory distress in both term and preterm infants.[3–5]

Persistent pulmonary hypertension of the newborn was described more than 35 years ago,[6] but a specific and unique treatment for this disorder, inhaled nitric oxide (INO), was approved only about 2 years ago. This occurred after several multicenter trials tested the benefits of administration of INO, presumably to overcome a local pulmonary deficiency in production or availability of endogenous nitric oxide (NO) to induce postnatal pulmonary vascular relaxation. These studies demonstrated a significantly decreased need for extracorporeal membrane oxygenation. Pearson et al.[7] demonstrated that infants with PPHN have low plasma concentrations of arginine, the precursor of nitric oxide, and of total plasma nitric oxide metabolites. These findings support the hypothesis that inadequate pulmonary production of NO contributes to the pathogenesis of neonatal pulmonary hypertension. An analysis of the multicenter studies[8] demonstrated a very consistent dichotomous pattern of response to INO, ranging from trivial or no response to, at the other extreme, a dramatic increase in arterial PO_2, presumably

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induced by improved total pulmonary blood flow and its more proper intrapulmonary distribution resulting from pulmonary vascular relaxation. A testable hypothesis is that dramatic responders to INO are deficient in their endogenous production or availability of NO. Recent insights may explain why some infants are responsive to INO and some are not. Wechsler et al.[9] demonstrated that, in patients with asthma, much of the variation in endogenous NO pulmonary excretion is explained by a single SNP in the promoter region of the neuronal or Type 1 nitric oxide synthase. A similar result was found in patients with cystic fibrosis.[10] Similar data are lacking in neonates at risk for PPHN, but exploring a possible linkage between SNPs and the clinically observed range of responses to INO should be fruitful.

The dichotomous response to INO allows application of the technique of extreme phenotypic discordance.[11] Given the pattern of results in response to INO in PPHN, it should be possible, with relatively small numbers of patients, to determine whether or not deficiencies in the production or upregulation of one or more of the nitric oxide synthase (NOS) isozymes is an important predictor of response. Such findings would be important, not only for the perinatal period but also for the follow-up of survivors as they move into adult life. Sartori et al.[12] demonstrated that adults who as infants had neonatal pulmonary hypertension continued to manifest exaggerated increases in pulmonary artery pressure when subjected to a hypoxic stimulus. These results are compatible with the presence of genetic variation in NO production under stressful circumstances and, hence, in variation in the regulation of pulmonary vascular resistance both in the neonate and in the adult. The technique of extreme phenotypic discordance has been utilized to demonstrate an association between a meaningful mutation in the 5′ flanking region of the Type 3 NOS gene in adults with coronary spasm.[13] A T to C mutation in the Type 3 gene promoter region reduces endothelial NO synthesis and predisposes patients with this mutation to the clinical problem of coronary spasm. A similar approach in neonates also may yield clinically meaningful results.

The above examples demonstrate the power and also the seductiveness of linking increasingly easily performed genomic evaluations with complex disorders. Linkages are associations at best, and statistically significant nonrandom associations with neonatal disorders are not the same as clinically meaningful predictions, allowing modifications and individualization in therapy. This warning has been clearly articulated by Gambaro et al.,[14] who summarize some potential errors in inappropriate extrapolation of results of linkage studies. For example, sampling bias and poorly described or unrecognized confounding factors in the population being studied are well known pitfalls. The interactions between environmental factors and polymorphisms of particular genes, the role of imprinting and, of course, post-translational changes in the protein encoded by the particular gene can occur. Each factor then limits the familiar paradigm of altered gene producing an abnormal protein producing a specific clinical disorder. Workers linking SNPs to severity or response to therapy of clinical disorders in neonates should heed these limitations.

The early 21st century may come to be seen in later days as a golden moment in the history of medical research. There is increasing sophistication in the knowledge base about the regulation of genes, the production and post-production modifications of proteins, and the relationship of their altered gene expression and protein production to important medical disorders. In adult and pediatric medicine, much of the low-hanging fruit has already been picked; the more difficult clinical problems still need to be attacked with imagination and with an approach that is informed by insights from genomic analysis. There is a rapidly increasing understanding about the basis for, and the extent of, the variability or occurrence of severity of similar diseases between one human being and the next, including babies. Because of these new insights, there is a rising and realistic public expectation and public faith that there will be improved — meaning safer and more efficacious — and more individualized, medical care. Because of the extraordinary expense and lifelong implications of undertreatment, overtreatment, or maltreatment of important disorders arising in the perinatal period, it is particularly crucial for us to get it right. Those engaged in basic, translational, and clinical outcomes-based research, and who thereby serve as the stewards of this public trust, must deliver.

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