

Genetics and biology of asthma 2010: *La' ci darem la mano...*

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"*La' ci darem la mano*"—"There we'll be hand in hand"—sings Don Giovanni to Zerlina in Act 1 of the opera the Danish philosopher Søren Kierkegaard called "a work without blemish, of uninterrupted perfection."¹ As if taking cues from Mozart's masterpiece, the genetics and biology of asthma are finally beginning to intertwine hands. This issue of our journal proudly presents 3 cutting-edge articles²⁻⁴ that recapitulate the joys and sorrows of asthma genetics 2010: the exciting power of discovery the field has found in genome-wide association studies (GWASs), the complex challenges GWASs inevitably still involve, and the preeminent role of biology in deciphering what genetic findings reveal about asthma susceptibility.

Compared with traditional candidate gene and linkage studies, GWASs have the advantage of interrogating single nucleotide polymorphisms (SNPs) across the whole genome to identify novel disease susceptibility genes unrestrained by prior knowledge. However, this advantage has proved to be a double-edged sword because GWASs have oftentimes zoomed onto genes so unexpected that their identification has been called into question on grounds of poor biological plausibility. The case of *ORMDL3* (ORM1-like 3), the gene identified by the first asthma GWAS,⁵ is emblematic. Almost 3 years after publication of the study, the community is still debating whether the signal in chromosome 17q21 (which in the meantime has been widely replicated,⁶⁻⁹ although with subtle differences) is truly associated with variants in *ORMDL3* or rather derives from neighboring loci included within a block of strong linkage disequilibrium. The elusive biological relevance of *ORMDL3* to asthma pathogenesis significantly adds to this continued uncertainty.

The 3 articles we present in this issue eloquently illustrate the virtues of synergism between asthma genetics and asthma biology. In the first article Stephanie London's group took an approach that explicitly links high-throughput genome-wide analysis to biology.² Although human association studies and murine models have identified more than 200 asthma candidate genes, only some of these genes have been systematically tested for replication in human populations. This study comprehensively evaluated the association of previously implicated asthma

candidate genes with childhood asthma in Mexico City, thus focusing on an admixed population never previously studied with comparable depth. A GWAS was performed in 492 asthmatic children aged 5 to 17 years and both parents, typing more than 500,000 SNPs in each subject. Reasoning that preselecting SNPs from candidate genes and analyzing this prioritized subset of SNPs separately might improve the power of detecting a disease susceptibility locus in a GWAS,¹⁰ these investigators performed separate candidate gene analyses for 2,933 autosomal SNPs in 237 selected genes. Sixty-one of these genes had at least 1 SNP with a significant association with asthma. Multimarker analysis identified the genes encoding TGF- β 1 (*TGFB1*), IL-1 receptor-like 1 (*IL1RL1*), IL-18 receptor 1 (*IL18R1*), and dipeptidyl peptidase 10 (*DPP10*) as the genes most significantly associated with asthma.

In the second article a large group led by Kathleen Barnes reports the results of the first true GWAS for asthma in populations of African descent.³ Using 2 independent sets of samples, an African American case-control group from Baltimore–Washington, DC (n = 935), and 163 African Caribbean families from Barbados (n = 929), and genotyping more than 650,000 SNPs in each subject, this study identified 3 potential asthma genes: α -1B-adrenergic receptor (*ADRA1B*) on chromosome 5q33, prion-related protein (*PRNP*) on chromosome 20pter-p12, and *DPP10* on chromosome 2q12.3-q14.2.

Finally, Debbie Meyers, Gene Bleeker, and their collaborators performed a GWAS on a population of patients with severe or difficult-to-treat asthma to identify genes that are involved in asthma pathogenesis.⁴ In this study 292,443 SNPs were tested for association with asthma in 473 cases from The Epidemiology and Natural History of Asthma: Outcomes and Treatment Regimens (TENOR) population and 1,892 Illumina general population control subjects. Asthma-related quantitative traits (total serum IgE level, FEV₁, forced vital capacity [FVC], and FEV₁/FVC ratio) were also tested in identified candidate regions in 473 TENOR cases and 363 phenotyped control subjects without a history of asthma to further analyze GWAS results. Multiple SNPs in the *RAD50-IL13* region on chromosome 5q31.1 were associated with asthma, with the strongest signal in intron 2 of *RAD50*. The *HLA-DR/DQ* region on chromosome 6p21.3 was also associated with asthma.

Although all of these studies are technologically state of the art and novel in their conclusions, none of them is immune from problems. For instance, the relatively small size of the population samples precluded reaching results of robust statistical significance. Moreover, replication of results was sometimes problematic, especially in admixed populations, and when it was achieved, it was loose (ie, at the level of the gene but not of a specific SNP³). Despite these problems, biology lends credibility to the findings of these studies because the products of all the reported asthma-associated genes are plausible players in specific facets of allergic lung inflammation. As discussed in detail in the relevant articles,²⁻⁴ TGF- β 1 plays a central role in allergic

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airway inflammation and airway remodeling; IL-1 receptor-like 1 binds IL-33 and plays important roles in the regulation of T_H2 airway inflammation; IL-18 receptor 1 binds *IL18*, enhances T_H1 cell-driven immune responses in synergy with *IL12*, and can also induce T_H2 responses; DPP10, a member of the dipeptidyl peptidase family, can remove N-terminal dipeptides from chemokines and cytokines, and thus might modify their functional activities; the adrenergic α -1B-receptor is expressed in the lung and is involved in proinflammatory responses; prion-related protein, typically implicated in transmissible neurodegenerative spongiform encephalopathy, is also detected on human B and T lymphocytes, preferentially regulatory T cells; and *HLA-DR/DQ* is part of the HLA class II region, which is critical for antigen presentation.

Biology also helps make sense of a seemingly odd but tantalizing finding. Indeed, it would be difficult to understand how polymorphisms in the introns of *RAD50*, a DNA repair gene, could provide strong signals in an asthma GWAS if work in mice had not shown that these regions harbor regulatory elements that control the expression of the T_H2 cytokines *IL13* and *IL4*, even though these genes are located thousands of base pairs away.¹¹⁻¹⁴ Because of this peculiar topology, variants geographically in *RAD50* might have little effect on *RAD50* regulation and major effects on T_H2 cytokine expression.

However, if biology remains so crucial for the interpretation of GWASs and is in fact a measure of their success, should these studies (cumbersome, expensive, and logistically demanding as they are) still be viewed as the best approach for discovering disease susceptibility genes? More irreverently, can genetic determinants of common diseases be discovered by using purely genetic approaches? No simple answer can be provided for such a complex question. In several cases GWASs have been extremely informative, especially when analyzing diseases for which well-defined, quantitative phenotypic information is available.^{15,16} On the other hand, asthma is a very complex disease first and foremost because of its phenotypic heterogeneity. Thus moving away from simplistic, dichotomous phenotypes (asthma no/yes) and improving phenotypic assessments is a priority but one that comes in the way of the statistics-driven need to recruit larger and larger populations. The studies we present illustrate several other problems in need of urgent solutions. The fact that GWASs have only identified a modest number of common variants of relatively modest effect supports the notion that numerous rare functional SNPs are major contributors to susceptibility to common diseases.¹⁷⁻¹⁹ Although it is estimated that approximately 60% of SNPs in the human genome have minor allele frequencies of less than 5%, companies producing GWAS arrays are biased toward common tagging variants in support of the common disease-common variant hypothesis, and consequently, there are relatively few rare SNPs in coding and promoter regions in their SNP genotyping panels.²⁰ Moreover, the currently available commercial chips are inadequate in content for populations of African origin,³ and yet these populations often present with particularly severe disease. Finally, as we recently discussed elsewhere,²¹ it is difficult to assess how much environmental exposures affect the outcome of GWASs, but it is reasonable to surmise that

environmental heterogeneities might contribute significantly to discrepancies in GWAS results.

What is next? As Dermitzakis and Clark²² recently wrote, “the biggest opportunity for making serious progress in understanding chronic disease risk lies in developing a deeper biological awareness into genomic approaches to the study of complex disorders.” In other words, it is time for genetics and biology to truly go hand in hand; not only at the end, when results are interpreted, but also and most importantly early on, when studies are designed.

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