Genetics of Susceptibility to Infectious Diseases

Genetic variations help to control responses to microbial pathogens and influence susceptibility to diseases that they cause

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We and microbial pathogens are often locked in a dynamic battle mediated by their virulence factors and our defenses. If we do not mount effective immune responses to pathogens, we will not overcome infections. However, if we overreact, host immune cascades may make those infections more severe. Both nonspecific factors, such as age and general health status, as well as specific genetic factors contribute to this balance. For instance, some individuals appear to be predisposed to certain infections whereas others are protected. By studying the immune response and genetic makeup of such individuals, we are gaining a better understanding of disease mechanisms and the interactive networks that either overcome infections or lead to serious pathologies.

We have only begun to uncover the genetic variations that control our response to microbial pathogens and influence our susceptibility to diseases caused by them. Inasmuch as all infectious diseases have a strong immunologic component, the early focus has been on genetic variations that control and regulate our immune responses to infectious agents. These immunogenetic polymorphisms appear to protect some but not all of us from specific infections. In some cases, genes that encode protection against a particular pathogen may increase the risk for another. Ultimately these variations ensure that some of us will survive a catastrophic biological threat, and help protect our species from extinction.

Many Types of Genetic Variations Influence Susceptibility to Pathogens

Although we know about many genetic variants that modulate responses to certain pathogens and diseases caused by them, we have a lot more to learn. Identifying genetic polymorphisms responsible for differential susceptibilities to infection is important, as is using this knowledge to understand the molecular mechanisms by which these polymorphisms affect outcome. This knowledge also helps our efforts to design specific therapeutic treatments to protect those who are most susceptible to some emerging and reemerging infections and to respond more effectively to a bioterrorism event.

Key genetic variations involve receptors and other components of the innate and adaptive immune system. In particular, polymorphisms in genes encoding Toll-like receptors (TLRs) of the human innate immune system partly explain individual variations in responses to pathogen-associated molecular patterns that affect susceptibility to certain infections or inflammatory conditions and influence responsiveness to particular vaccines (see ASM News, July 2004, p. 317).

For example, TLR2 gene polymorphisms are associated with reduced interleukin-12 (IL-12) production in leprosy patients, abrogation of nuclear factor-κB (NF-κB) activation and tumor necrosis factor-α (TNF-α) production in response to Mycobacterium tuberculosis stimulation, and increased risk of staphylococcal infections. Similarly, TLR4 variants are associated with risk for severe systemic disease caused by certain lipopolysaccharide (LPS)-producing bacteria. Specifically, the common TLR4 allele among Caucasians, TLR4B, triggers protective levels in response to LPS, whereas several other coding variants of TLR4 are associated with hyporesponsiveness to LPS and increased risk for severe infections.

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In addition to genetic variants that alter the structure and direct function of TLRs, several polymorphisms disrupt the ability of specific TLRs to recruit and interact with their cytoplasmic adaptor molecules. These changes can also result in aberrant signaling and defective responses to infections and may contribute importantly to susceptibility to bacterial infections such as anthrax and tuberculosis.

Further, polymorphisms in genes that encode complement components, their receptors, and proteins that regulate their functions are also linked to increased susceptibility to some infections. For example, inefficient variants of Fcy receptors and plasminogen activator inhibitor type 1 polymorphisms are linked to meningococcal disease susceptibility, which is also modulated by polymorphisms in the mannose-binding lectin (MBL) protein. For instance, critically ill patients in Denmark with certain mb12 allelic variants that confer low levels of MBL expression are at high risk of developing systemic inflammatory response syndrome and dying from it. Additional examples of inherited polymorphisms in complement component genes that affect bacterial disease susceptibility can be found at several websites, including www.complement-genetics.uni-mainz.de.

Similarly, researchers studying antimicrobial peptides are searching for genetic polymorphisms in these genes that affect susceptibility to infections. For instance, variants of human β-defensins are linked to variable susceptibility to Candida and possibly other oral infections, while other variants are associated with chronic obstructive pulmonary disease.

Yet another rapidly growing area in the field of infectious disease genetics involves polymorphisms in killer Ig-like receptors (KIR) that are expressed on natural killer (NK) and T cells. Members of the KIR family represent highly homologous immune receptors that regulate NK-cell responses through recognition of human leukocyte antigen (HLA) class I molecules on target cells. Heterogeneity of KIR genotypes and haplotypes is extensive, and some of these variants are associated with susceptibility to infections, including those by plasmodia, HIV, and hepatitis C virus (HCV).

**Cytokines Modulate Inflammatory Cascades and Responses to Pathogens**

Variations in genes encoding cytokines and chemokines, their receptors, or their inhibitors can affect whether individuals develop proinflammatory (Th1) or anti-inflammatory (Th2) cytokine profiles when responding to particular pathogens. At least 20 different infectious diseases and syndromes are firmly linked to polymorphisms in this class of genes. This linkage is not surprising because of the central role of these molecules in orchestrating immune responses. In some cases, a particular polymorphism increases an individual's susceptibility to certain infections while conferring protection against others.

Research on such polymorphisms dates to the discovery of the polymorphism in the FY/Duffy gene that encodes a chemokine receptor expressed on red blood cells that is also a receptor for vivax malaria. Thus, the null allele is present in nearly 100% of the African population in areas where malaria is endemic. Although rare in other populations, this allele protects individuals against malaria when homozygous and thus may explain its high frequency in this population.

Meanwhile, researchers learned that certain chemokines sometimes inhibit HIV-1 infection, particularly in those cases where the receptors are essential for cells to become infected. Indeed, at least six variants in chemokine, chemokine receptor, and cytokine genes contribute significantly to AIDS. For instance, CCR5, which is a coreceptor for HIV that facilitates its entry into CD4 cells, can significantly influence HIV infection outcomes. The specific genetic deletion variant CCR5Δ32, which results in an inactive protein and is found in 5–15% of Caucasians, and a tightly linked CCR2–641 variant are associated with slow progress of HIV disease in heterozygotes and protects against infection in individuals who are homozygotes.

However, these same alleles appear to confer risk to other infectious and autoimmune conditions, including systemic lupus erythematosus, insulin-dependent diabetes, pulmonary sarcoidosis (CCR5), and hypertension. In addition to CCR5, IL-10 and CCR2, variants of CCR5P, CCR2, stromal cell-derived factor (SDF-1), and RANTES also contribute to AIDS progression.

Cytokine and chemokine gene polymorphisms also can lead some individuals to develop unusually severe mycobacterial and viral diseases, even when they are infected with otherwise weakly pathogenic strains. Although these individuals had no known primary immu-
nodeficiency, investigators learned that they shared a defect in their interferon-γ (IFN-γ) responses that arise because of polymorphisms in genes encoding either IL-12, IL-12R, IFN-γ, IFN-γ-R, and/or STAT1, and that may occur within the coding region, promoter region, or in some cases outside genes encoding Th1 or Th2 cytokines, including IFN-γ, IL-10, IL-4, IL-6, and TNF-α.

Genetic variants of IFN-γ or its receptor that have opposing effects on the level of production or response to this cytokine may be differentially selected, depending on the types of pathogens that are prevalent in particular populations. Higher levels of IFN-γ usually promote microbial killing, but in some cases a high-producer phenotype can cause severe inflammation, leading to host-mediated pathogenesis. Thus, heterozygotes carrying both high- and low-producer alleles may be at an advantage because of their ability to balance their responses, producing enough to kill a pathogen but not enough to cause immunopathology.

Indeed, the heterozygous advantage is documented in studies that associate IFN-γ receptor genetic variants with either cerebral malaria or hepatic fibrosis that accompanies schistosomiasis. In the same context, variations in anti-inflammatory cytokines, such as IL-4 and IL-10, may affect outcomes of HCV infections, rates of
progression of HIV infections, and meningococcal infections. Genetically linked polymorphisms that result in high levels of IL-10 production are associated with adverse outcome of pneumococcal infection due to induced immunosuppression and impaired bacterial clearance. However, the same genotype is also associated with resistance to common herpesvirus infections and also protects against primary Epstein-Barr virus infections. Homozygosity versus heterozygosity in these TNF alleles, as well as genetic polymorphisms in other members of the TNF receptor superfamily, may also potentiate these effects. The issue is further complicated because these microsatellites are in linkage disequilibrium with themselves and as a part of extended haplotypes, including class I and class II HLA alleles.

**Major Histocompatibility Complex Variability Affects Responses to Diseases**

In humans, the major histocompatibility complex, also known as the HLA system, represents the most heterogeneous locus known. The three major classes of HLA, class I, class II and class III, located on chromosome 6, play a pivotal role in developing and regulating immune responses to pathogenic microbes.

Both class I and class II HLA molecules present antigenic peptides to T cells, which then mount antigen-specific immune responses. The HLA class I molecules are HLA-A, HLA-B, and HLA-C; the class II molecules are HLA DP, DQ, and DR. The class I molecules are expressed on all nucleated cells, whereas the class II molecules are expressed only on specialized cells, primarily those involved in antigen presentation to the CD4 T cells. Both class I and class II HLA molecules consist of α and β heterodimers. The class I β chain is the nonpolymorphic β2 microglobulin, whereas the α chains of HLA-A and HLA-B are highly polymorphic. With the exception of the α chain of the DR molecule, both α and β chains of class II molecules are highly polymorphic.

In fact, at the gene level, more than 950 class I alleles and over 650 class II alleles are known. This extensive polymorphism is not scattered throughout the molecule; rather, it is mostly condensed in the part of the molecule that binds the peptide antigen, known as the binding groove. Nonsynonymous polymorphisms resulting in conformational changes in the binding groove allow a highly diverse array of peptides to be bound by different class II allotypes. Individuals who are not capable of binding and presenting protective antigens from a particular pathogen will not be protected from that pathogen and may die. Others with an HLA variant that can effectively bind and present protective antigen will be able to mount an effective protective immune response to this pathogen and survive the infection.
Imagine there is only one type of HLA molecule that can bind and present to T cells only one protective antigen of a particular pathogen: we would all be protected from this but no other pathogen. If we were exposed to a different pathogen or if this pathogen mutated to where it would no longer bind to this single HLA type, none of us could mount protective immunity to it, and our species might vanish (Fig. 1). The same outcome could occur if there were only a limited number of HLA variants. The HLA class I and class II genes are, therefore, under a strong selective pressure from pathogens to maintain a high degree of polymorphism. The vast number of HLA allelic variation is believed to have evolved to protect our species from extinction, allowing some individuals to survive infections by different pathogens (Fig. 2).

Hence, it is not surprising that particular HLA types are associated with greater or reduced susceptibility to various infectious diseases, including hepatitis B virus, hepatitis C virus, human T-lymphotropic virus-I, HIV, leprosy, tuberculosis, and malaria. Typically the strongest associations are those that appear to protect against disease, although a number of associations lead to increased susceptibility.

**HLA Genetic Variation and Outcomes of Group A Streptococcal Infections**

Group A streptococci (GAS) provide an excellent model for studying host-pathogen interactions and how host factors contribute to clinical outcomes. These bacteria can cause a wide variety of human diseases, ranging from uncomplicated pharyngitis to severe and life-threatening illnesses such as rheumatic fever, streptococcal toxic shock syndrome (STSS), and necrotizing fasciitis (NF). Although there are over 100 serotypes of GAS, the same strain can be isolated from STSS, NF, and nonsevere bacteremia cases, or even from asymptomatic individuals. We reasoned that host factors must be contributing to these disease processes.

Hence, we focused on patients with invasive GAS infection cases that presented with severe systemic disease such as STSS or with nonsevere systemic disease such as mild bacteremia or cellulitis. All our patients were recruited through the active surveillance of invasive GAS infection conducted by the Ontario Streptococcal Study Group, which is headed by Donald Low of Mount Sinai Hospital Toronto, Ontario, Canada, and Allison McGeer of the University of Toronto. We determined the distribution of HLA class II haplotypes among these patients and also among matched, healthy individuals in Ontario. We looked first at the effects of HLA class II allelic variation because we had determined that the streptococcal superantigens (Strep SAgS) are major contributors to STSS pathogenesis and knew that HLA class II molecules also serve as receptors for superantigens. Superantigens bind directly to the HLA class II molecules and are presented to T cells in a less-restricted manner than regular antigens, usually...
causing at least $10^5$-fold-higher responses than conventional antigens. This response led Philippa Marrack to call this group of microbial toxins superantigens.

We proved that the super potent inflammatory response during an invasive GAS infection can lead to severe systemic disease with hypotension, organ failure, and shock. We also found the strongest HLA association to be with protection from STSS. Specifically, individuals with invasive GAS infection who carried the DRB1*1501/DQB1*0602 HLA class II haplotype were significantly protected from severe disease ($P = 0.005$). The frequency of this haplotype was also significantly higher in the nonsevere invasive cases compared to healthy controls ($P = 0.0007$). Two haplotypes seem to increase the risk for severe invasive disease, namely DRB*14/DQB1*0503 and the DRB1*07/DQB1*0201.

Meanwhile, the HLA class II associations with NF and STSS are different, a finding that pleases many infectious disease physicians who suspected that these are distinct manifestations of complicated invasive infections and thus require distinct patient management strategies. Even more interesting, patients who had NF were protected from developing additional STSS if they carried the STSS protective DRB1*1501/DQB1*0602 haplotype.

Because invasive GAS infections are relatively rare and it took a long time to enroll patients, we began investigating the molecular basis for the observed HLA class II associations with different outcomes. Moreover, patients who experienced severe systemic disease had a propensity, even in convalescence, to be high responders to the Strep SAgs produced by the isolate that infected them. By contrast, those who had nonsevere invasive disease were low responders. These results suggest that the propensity to be either a high or a low responder to streptococcal SAgs is a stable feature of the host response, controlled by immunogenetic factors, and that this propensity may contribute to differences in the severity of invasive GAS infection (Fig. 3).

We confirmed this notion in a series of functional studies in which healthy adults and recovered patients carrying either disease-protective or high-risk haplotypes were compared according to the magnitude of their response to the Strep SAgs from the highly prevalent M1T1 GAS serotype. Consistently, those who had the protective DRB1*1501/DQB1*0602 haplotype had a much-reduced cytokine and proliferative responses to the Strep SAgs as compared to those who had the high-risk DRB1*14/DQB1*0503 and DRB1*07/DQB1*0201 haplotypes. Moreover, those with the protective haplotype had lower responses than individuals with neutral (neither protective nor high-risk) haplotypes. To us, these findings suggest that the protective haplotype is immune suppressive. This notion was further confirmed by in vivo studies using HLA-transgenic mice carrying disease associated human HLA alleles.

Another receptor for SAgs is the Vβ element within the T-cell receptor (TCR) β chain. SAgs bind to Class II molecules and to specific Vβ elements on the TCR to bring the APC and T cells close enough to exchange activation signals that translate into inflammatory responses. Each SAg interacts with a characteristic set of TCR Vβ elements. Thus, variations in an individual’s TCR Vβ repertoire likely also affect the magnitude of the SAg response. However, we found that the contribution of the TCR Vβ repertoire variation to the overall SAg response is minimal compared to HLA class II allelic variation, which has a profound effect on the level and profile of the inflammatory response elicited by SAgs.

At one point we thought that the effect of the HLA class II variation on the Strep SAg response is related to differences in the affinity of binding of a given SAg to different class II alleles. Although we detected differences in SAg-Class II binding, they did not correlate with response levels. Hence, we are actively searching for something else that could control events ensuing from the interactions of Class II molecules, SAg, and TCR. Variations in genes other than the HLA Class II genes also may contribute to the host response to GAS virulence factors and to disease outcome. Candidates include the TNF microsatellites located within the HLA class III locus, cytokine genes, KIR genes, and other genes that control innate immune responses to streptococcal components. In addition, we have recently started to use novel mouse models, including fully “humanized” mice and complex trait recombinant inbred mice, to determine whether there are unexpected genes and pathways that may also participate in modulating host responses to GAS infection.

The advent of advanced biotechnology tools
and informatics will allow us to discover not only specific genetic associations with infectious diseases, but also the network of pathways that interact to modulate infectious disease phenotypes.

SUGGESTED READING


