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Variable phenotypic expression of mutations in genes of the immune system

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Commentary

Discovery of mutated genes that cause various types of primary immunodeficiencies has significantly advanced our understanding of the pathogenesis of these diseases and of the functions of normal gene products. However, it is becoming abundantly clear that the phenotypic presentation of mutations in a given gene can be quite different, depending upon the location and type of mutation but also probably upon other genetic factors and environmental influences. In this issue of the *JCI*, de Villartay et al. describe a third phenotype for mutations in*recombination activating gene 1* (*RAG1*), in addition to the already known phenotypes of SCID and Omenn syndrome.

G protein–mediated signals may be useful. Agents with pathway-selective activities might represent a potentially new generation of drugs. More studies that elucidate the physiologic roles of novel 7TMR signaling mechanisms, such as those reported here by Zhai et al. (3), will be necessary to lay the foundation for such developments.

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Variable phenotypic expression of mutations in genes of the immune system

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Discovery of mutated genes that cause various types of primary immunodeficiencies has significantly advanced our understanding of the pathogenesis of these diseases and of the functions of normal gene products. However, it is becoming abundantly clear that the phenotypic presentation of mutations in a given gene can be quite different, depending upon the location and type of mutation but also probably upon other genetic factors and environmental influences. In this issue of the *JCI*, de Villartay et al. describe a third phenotype for mutations in *recombination activating gene 1* (*RAG1*), in addition to the already known phenotypes of SCID and Omenn syndrome (see the related article on page 3291).

Human primary immunodeficiency diseases have been recognized for only a little over a half century (1). The spectrum of such diseases has grown at an extremely rapid pace during the past 50 years, with currently more than 120 different syndromes having been described (2). For most of that period, the conditions were identified by their clinical and immunologic presentations. However, for the past 12 years, they have been defined extensively at a molecular level. Because making a firm diagnosis by clinical and immunologic criteria has always been problematic due to variability in presentation, it was thought that molecular testing would remove this ambiguity. The report by de Villartay et al. in this issue of the *JCI* clearly indicates that is not always the case (3). In the pre-molecular diagnostic period, there had been examples within sibships of clinical variability in expression of primary immunodeficiency. One of these was in a family reported by de Saint-Basile et al. (4) in which one sibling had Omenn syndrome and another had SCID. Subsequently, the explanation for this was found when mutations in *recombination activating genes 1* and *2* (*RAG1* and *RAG2*, respectively) were found to cause SCID (5), and later hypomorphic

Nonstandard abbreviations used: ADA, adenosine deaminase; BTK, Bruton tyrosine kinase; CD3ε, CD3 ε chain; *RAG1*, *recombination activating gene 1*; SAP, signaling lymphocyte activation molecule–associated (SLAM-associated) protein; WASP, Wiskott-Aldrich syndrome protein.

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Table 1
Mutated immune system genes with variable phenotypic expression

Mutated gene	Normal gene product	Classic syndrome	Variant syndromes
RAG1	Recombination activating gene 1	SCID (4, 5, 7, 15)	1. Omenn syndrome (4, 6, 7) 2. Oligoclonal γ/δ T cells, autoimmune disease, and CMV infection (3)
BTK	Bruton tyrosine kinase	Agammaglobulinemia (8)	Polysaccharide antibody deficiency (9)
WASP	Wiskott-Aldrich syndrome protein	Wiskott-Aldrich syndrome (10)	X-linked thrombocytopenia (10)
SH2D1A	Slam-associated protein (SAP)	Fatal infectious mononucleosis (11)	1. Common variable immunodeficiency (11) 2. Hemophagocytic lymphohistiocytosis (12)
CD3ε	CD3 ε chain	SCID (14, 21)	Moderate susceptibility to infection (13)
IL2RG	Common cytokine receptor γ chain (γc)	SCID (15)	Moderate combined immunodeficiency (16)
ADA	Adenosine deaminase	SCID (15, 17)	Moderate combined immunodeficiency (17)
JAK3	Jak3	SCID (15)	Moderate combined immunodeficiency (18)

References are shown in parentheses.

mutations in these genes were found to cause Omenn syndrome (6). It was then found that identical mutations in these genes could cause either syndrome (7). De Villartay and his colleagues (3) now show that another distinct clinical syndrome can be caused by hypomorphic mutations in *RAG* genes. In contrast to infants with SCID or Omenn syndrome due to *RAG* mutations, the infants in the current report had autoimmune cytopenias, B cells, normal Ig levels, clonal T cells with γδ TCRs, normal T cell proliferation, and only slightly elevated NK cell levels. Because all of the infants had disseminated CMV infections, the authors suggest that the expansion of Tiγδ T cells (where Ti represents the TCR expressing the γδ heterodimer) was due to stimulation by the CMV virus. As the authors point out, there is mounting evidence that T cells with γδ receptors have a role in host defense against several microbial pathogens, and they have been found to be increased in immunocompromised humans infected with CMV after transplantation. Thus, it is possible that environmental or other genetic factors modify the clinical manifestation of such mutations.

Other genes with phenotypic variability

Other genes of the immune system in which mutations result in variable phenotypes include those that encode Bruton tyrosine kinase (BTK), Wiskott-Aldrich syndrome protein (WASP), signaling lymphocyte activation molecule–associated (SLAM-associated) protein (SAP), CD3 ε chain (CD3ε), common γ chain (γc), adenosine deaminase (ADA), and Jak3 (Table 1). These genes encode proteins essential for: (a) B cell development and function (BTK); (b) T cell development and signaling (CD3ε); (c) T cell function and survival (ADA); (d) cytokine receptor signaling (γc and Jak3); (e) regulation of cell signaling (SAP); and (f) intracellular signaling and actin polymerization (WASP). Usually, but not always, the phenotypic variability is due to the type and location of the mutation, with hypomorphic mutations causing a less complete immunodeficiency. In the CBA/N mouse, a mutation in the *Btk* gene results in polysaccharide antibody deficiency, whereas such mutations in humans usually cause agammaglobulinemia (8). However, there is a report of a human with only polysaccharide antibody deficiency who had a mutation in this gene (9). Mutations in the *WASP* gene result in Wiskott-Aldrich syndrome but also in X-linked thrombocytopenia (10). *SH2D1A* mutations have been shown to cause variously fatal infectious mononucleosis, common variable immunodeficiency (11), or hemophagocytic lymphohistiocytosis (12). Mutations in CD3ε were first reported to cause a relatively mild form of immunodeficiency (13), but more deleterious mutations were later found to cause SCID (14). Mutations in the genes encoding γc, ADA, and Jak3 usually result in SCID (15) but have also been shown to cause less severe forms of immunodeficiency (16–18).

Clinical ambiguity also arises from the fact that seemingly identical clinical syndromes can be caused by mutations in different immune system genes. For example, it was recently shown that Omenn syndrome can also be caused by mutations in the *Artemis* gene that encodes a DNA repair factor (19). Other conditions include the hyper-IgM syndrome that can be caused by mutations in at least 6 different genes (20) and SCID, known to be caused by mutations in at least 10 different genes (15, 21). The most striking example of this will likely be the syndrome of common variable immunodeficiency that promises to have many different molecular causes, including mutations in genes that encode SAP (11), inducible costimulator (ICOS) (22), transmembrane activator and calcium modulator and cyclophilin ligand interactor (TACI) (23), and the CD40 ligand (20).

Conclusion

The above examples are likely only the tip of the iceberg. Discovery of the molecular bases of many primary immunodeficiency diseases has been of major importance in understanding their pathogenesis and inheritance as well as in elucidating the functions of the genes in the immune system. It is hoped that these discoveries will eventually be of importance for gene therapy. The recognition by de Villartay and his colleagues (3) of a new syndrome caused by *RAG1* mutations should alert all physicians who care for patients with recurrent infections that atypical presentations may occur when genes of the immune system are mutated. However, from a diagnostic standpoint, until routine molecular testing for genes known to be mutated in primary immunodeficiency is implemented, most of these conditions will go undetected, and the full spectrum of phenotypic and genotypic heterogeneity will not be known. The routine use of molecular testing is unfortunately probably a few years off, considering that it is currently available primarily only in research laboratories and that newborn screening by available immunologic methods is not even performed for any of these conditions. Indeed, there is no screening

for any of these conditions at any time of life anywhere in the world. Until this happens, there will still be patients who present with recurrent infections who have undiagnosed, genetically determined immunodeficiency, the basis of which is unidentified. Failure to make these diagnoses early in life results in high rates of mortality and morbidity that could be prevented.

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New insights into nNOS regulation of vascular homeostasis

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An important physiological response to changes in local or systemic oxygenation is the modulation of vascular tone, which is mediated in part by changes in the activities of the 3 NO synthase (NOS) isoforms. In arterial smooth muscle cells, acute hypoxia induces increased vascular tone, which is attenuated if hypoxia persists. In this issue of the *JCI*, Ward et al. demonstrate that changes in O_2 concentration have effects on neuronal NOS enzymatic activity and gene expression that contribute to vascular homeostasis under conditions of acute and chronic hypoxia (see the related article beginning on page 3128).

Every cell in the human body is dependent upon the delivery of adequate concentrations of O_2 to maintain normal cellular functions, which are principally powered by ATP derived from mitochondrial oxidative phosphorylation. The anatomical matching of O_2 delivery to demand is determined by the production of secreted factors that stimulate blood vessel growth, most notably VEGF (1). Every step of its biogenesis, from transcription of *VEGF* gene sequences in the nucleus and protection of the resulting mRNA against degradation to the ribosomal translation and folding of VEGF protein in the endoplasmic reticulum and transport via the Golgi system to the plasma membrane for secretion, is O_2 regulated, as is the expression of its cognate receptor on vascular endothelial cells (1–3).

Anatomical responses to changes in O_2 demand occur on a scale of days, whereas other physiological responses resulting in alterations in O_2 delivery occur on a scale of seconds. Systemic responses are mediated by chemoreceptor cells in the carotid body that depolarize in response to reduced arterial O_2 tension, leading to reflex changes in ventilation, heart rate, and vascular tone (4). The vasculature within tissues also responds to acute regional hypoxia by dilation of arterioles that control the flow of blood into each capillary bed, as in the case of increased O_2 consumption in skeletal muscle during exercise (5). In contrast, when systemic hypoxia occurs as a result of vascular hypotension (shock), the adrenergic nervous system directs redistribution of blood flow to maintain the perfusion of

Nonstandard abbreviations used: HIF-1, hypoxia-inducible factor–1; nNOS, neuronal NOS; NOS, NO synthase.

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