

# Case of a 10-Year-Old Boy with Hyper-Immunoglobulin M Immunodeficiency Caused by Activation-Induced Cytidine Deaminase Deficiency

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We report the case of a 10-year-old boy, with a history of recurrent sinopulmonary infections, presenting with pneumococcal meningitis. Immune workup found low immunoglobulin G (IgG), IgA, and IgE and elevated IgM. Specific antibodies to pneumococcus, tetanus, and mumps were low. Genetic testing identified CD40L and uracil-DNA glycosylase as normal. Sequence analysis for mutations in activation-induced cytidine deaminase (AID) gene on chromosome 12p13 revealed a single missense mutation in exon 3 caused by a point mutation. AID is an enzyme that is specifically present in activated B cells and necessary for isotype switching. This case highlights the significance of AID deficiency in hyper-IgM immunodeficiency.

## Background

**H**YPER-IMMUNOGLOBULIN M (IgM) SYNDROME was first described in 1961 as a disorder characterized by low IgG, IgA, and IgE levels with normal or elevated IgM.<sup>1</sup> The initial defect discovered was the loss of CD40L, an essential component to helper T-cell/B-cell interaction. Without CD40L, the T cell cannot activate the B cell to allow for isotype class switching. This defect is x-linked recessive. An autosomal recessive form due to deficiency of CD40, the coreceptor for CD40L, was also discovered. These 2 defects are classified as combined T- and B-cell immunodeficiencies by the International Union of Immunological Societies Expert Committee on Primary Immunodeficiencies.<sup>2</sup>

Two other rare forms of hyper-IgM, better termed class switch recombination defects, which have been defined as antibody deficiencies, have been reported.<sup>2</sup> These defects are caused by mutations in nucleic acid-modifying enzymes called activation-induced cytidine deaminase (AID) and uracil nucleoside glycosylase.<sup>3</sup> These defects are largely autosomal recessive, but an autosomal dominant pattern has been also observed.<sup>4</sup>

## Case Presentation

A 10-year-old boy presented for immunology consultation after hospitalization for pneumococcal meningitis complicated by mastoiditis, bilateral otitis media, sinusitis, hearing loss, and cranial nerve palsy. His medical history is significant for recurrent ear infections from early infancy forward, which required 3 sets of pneumostomy tubes for

management. Even with the tubes in place, he had constant otorrhea unless he was taking oral antibiotics. He had chronic cervical lymphadenopathy. When he was 5 years old, he was hospitalized for 2 days for pneumonia. He had chicken pox at 6 years of age and shingles at 9 years of age.

He was the product of an uncomplicated full-term gestation. His neonatal course was unremarkable.

His family history is significant for a maternal uncle who died early in infancy because of pneumonia.

Initial immune workup revealed an IgG level of 2 mg/dL (768–1,632 mg/dL), IgA level <6 mg/dL (68–378 mg/dL), IgM level 889 mg/dL (60–263 mg/dL), and IgE level <1.0 mg/dL (0–120 mg/dL). Pneumococcal, Mumps, and Tetanus IgG antibodies were all nonprotective. Complement levels, complete blood count, and mitogen-induced T-cell studies were normal.

Genetic testing via flow cytometry and sequence analysis identified CD40L and uracil-DNA glycosylase as normal. Sequence analysis for mutations in AID revealed a single missense mutation in exon 3 caused by a point mutation. The gene encoding AID is located on chromosome 12p13, which includes 5 exons, and generates a 198-amino-acid protein.<sup>1</sup> The second allele was normal. Therefore, the patient is heterozygous for the mutation, making his mutation autosomal dominant. It is likely due to a *de novo* mutation because there is no family history of immunodeficiency in either the parents or half sibling. Homozygous mutations are more common and exon 3 is the most common location of the gene defect.<sup>1</sup> This genetic testing result is consistent with the diagnosis of AID deficiency. With genetic analysis we were able to rule out other causes of his immunodeficiency, such as combined

variable immunodeficiency, MHC II deficiency, or ataxia telangiectasia, which could give a similar presentation with high IgM and may be overlooked when only focusing on hyper-IgM syndrome.<sup>5,6</sup>

The patient was started on monthly intravenous immune globulin (IVIG) injections of 15 g and completed a full antibiotic course for meningitis. Levels of IgG normalized with trough levels between 600 and 700 mg/dL. Mild hearing loss remained. The patient showed a decrease in sinopulmonary infections on stable Ig infusions.

## Discussion

AID is an enzyme that is specifically present in activated B cells and necessary for isotype switching. The gene encoding this enzyme is the AID gene, which is located on the short arm of chromosome 12p13.<sup>6</sup> With gene sequencing, multiple genetic mutations were discovered.<sup>7</sup> Missense mutations, in-frame insertions or deletions, premature stop codons, and large deletions have been detected.<sup>8,9</sup> The majority of deletions are homozygous rather than heterozygous.<sup>9-12</sup> As described in the case presentation, our patient had a heterozygous missense mutation. The defect in AID was originally termed hyper-IgM type II, but more recently the nomenclature has changed and it is more appropriately termed Ig class switch recombination deficiency.<sup>13</sup>

The process of isotype switching requires both class switch recombination and somatic hypermutation. First, class switch recombination is the process by which the IgM can be converted to another Ig such as IgA, IgG, or IgE. This occurs when there is DNA recombination between 2 distinct switch regions. The first is located upstream of the constant  $\mu$ , which is the constant domain of IgM, and the second is a target constant region of another isotype such as  $\alpha$ ,  $\gamma$ , or  $\epsilon$ , which are the constant domains of IgA, IgG, and IgE, respectively. For the switch to occur, the DNA located between the 2 switch regions is removed.<sup>4</sup> This results in replacement of the  $\mu$  chain with another heavy chain ( $\alpha$ ,  $\gamma$ , or  $\epsilon$ ), allowing for production of different antibody types (IgA, IgG, and IgE). During this process, the variable chain remains unchanged, thus conserving the antibody specificity and affinity.<sup>4</sup> AID creates the cleavage by changing the cytidine residues to uracil. The uracil is subsequently removed by uracil-DNA glycosylase. This creates a point where the DNA is cleaved by an endonuclease, generating a single-stranded DNA break that is processed and repaired.<sup>4,8</sup> Following this model of class switch recombination, it is evident that uracil-DNA glycosylase deficiency would halt the process of class switch recombination as well, because both AID and uracil-DNA glycosylase are necessary for completion of the process.<sup>4</sup>

Second, somatic hypermutation is a process of mutation introduction into the variable region of the Ig to allow for selection of B cells with the highest antigen affinity. The process of class switch recombination and somatic hypermutation occur independently in the germinal centers of activated B cells.<sup>4</sup>

The gene encoding AID is located on chromosome 12p13, which includes 5 exons, and generates a 198-amino-acid protein.<sup>1</sup> Mutations have been discovered along the entire length of the gene. Research on AID-deficient mice has revealed that the location of the mutation determines the defect. The mice had multiple different mutations, allowing

researchers to test each mutation's effect on both class switch recombination and somatic hypermutation in an attempt to determine if there were specific separate regions encoding these processes. During these experiments, it was discovered that the C- and N-terminal domains have distinct functional differences. For example, mutations in the C-terminal domain of the gene results in exclusive impairment of class switch recombination while conserving somatic hypermutation function. On the other hand, mutations in the N-terminus are exactly opposite, conserving class switch recombination and affecting somatic hypermutation.<sup>14,15</sup> The genetic analysis in this patient demonstrated a missense mutation resulting in a deficiency of AID. The lack of IgG, IgA, and IgE demonstrates class switch recombination impairment. Testing to determine the function of somatic hypermutation in this patient was not performed.

Most AID-deficient patients present within the first 2 years of life with recurrent sinopulmonary infections, although late presentation is not rare. Opportunistic infections are also not a feature of AID deficiency, whereas they are seen in CD40L and CD40 deficiencies because of the related T-cell defect.<sup>1,13</sup> Lack of opportunistic infections may be one reason that late presentation is not uncommon in these patients.<sup>1</sup> As AID deficiency is an antibody deficiency, infections are typically due to encapsulated bacteria where antibody is needed for destruction.<sup>3</sup>

Gastrointestinal tract infections are the second most common features of AID deficiency.<sup>10</sup> Giardiasis infecting the gastrointestinal system, bacterial meningitis, viral encephalitis, and severe hepatitis B infections have been reported.<sup>10,11</sup> Lymphoid hyperplasia of peripheral nodes, as well as mesenteric nodes, tonsils, liver, and spleen, is seen in 1/2 to 2/3 of patients with AID deficiency and is related to the massive proliferation of B cells in the germinal centers.<sup>1,3,4</sup> There is presence of giant germinal centers filled with proliferating B lymphocytes coexpressing IgM, IgD, and CD38.<sup>1,13</sup> The theory is that the B cells are constantly stimulated to produce antibody by antigen but cannot successfully class switch.<sup>7</sup> Follicular hyperplasia of the small bowel mucosa-associated lymphoid tissue has been reported in AID-deficient mice. This is associated with increased colonization of anaerobic bacteria and could be due to antigen overload caused by ineffective isotype switching to allow for clearance of these pathogens. This resolves with use of antibiotics, and studies indicate that AID may be directly involved in maintaining gut flora homeostasis not exclusively related to the failure to class switch to IgA.<sup>16,17</sup> There has been evidence of an increase in autoimmune pathologies including diabetes mellitus, autoimmune hepatitis, rheumatoid arthritis, inflammatory bowel disease, and uveitis in about 20% of patients.<sup>8,18</sup> The link to autoimmunity has been observed but has not yet been explained.<sup>3</sup> A study in AID-deficient mice suggested that the autoimmunity could be a result of increased B-cell proliferation leading to loss of peripheral tolerance or increase in antigens that are unable to be cleared, resulting in increased B-cell activation.<sup>18</sup>

Laboratory results of these patients show decreased levels of IgG, IgA, and IgE with normal to elevated IgM, which is not unique to AID deficiency but is seen in CD40L, CD40, and uracil-DNA glycosylase deficiencies as well. IgG-specific antibody responses are nonexistent. Some patients have IgM isohemagglutinins. Normal numbers of T and B cells with normal T-cell function are seen in AID deficiency. Normal

numbers of memory B cells are seen, but they are exclusively IgM memory cells.<sup>4</sup> AID deficiency is usually inherited as an autosomal recessive trait. However, there is now evidence of an autosomal dominant pattern of inheritance. A dominant mutation was first described by Kasahara and colleagues in 2003. The defect was located in the N-terminus of the gene and is caused by a premature stop codon that results in a loss of the last 9 amino acids.<sup>19,20</sup> Subsequent studies have revealed more patients with heterozygous mutations consistent with autosomal dominant inheritance.<sup>20</sup> The 2 different hypotheses to explain this pattern propose that the mutant form creates a dominant effect on the normal signaling and activity.<sup>14</sup> Genetic tests for AID and uracil-DNA glycosylase are commercially available and can be used in the definitive diagnosis of these 2 class switch recombination defects.<sup>1,3</sup>

Treatment options are limited. IVIG will create passive immunity to decrease the risk of infection.<sup>21</sup> There is some conflict in the literature on the effect of IVIG on lymphoid hyperplasia.<sup>1,3,7</sup> When an infection develops, aggressive antimicrobial therapy is initiated. Treatment of autoimmunity requires anti-inflammatory and immunosuppressive agents.<sup>3</sup>

The only treatment that can impact the underlying immune defect is bone marrow transplantation with a human leukocyte antigen-matched donor. A retrospective analysis of 8 European countries revealed a 58% normalization of labs and resolution of symptoms after hematopoietic stem cell transplantation in 38 hyper-IgM patients. Of those transplants, 22 patients received stem cells from unrelated donors.<sup>22</sup>

### Author Disclosure Statement

The authors have no conflict of interest or disclosure to make regarding this case report.

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Received for publication December 15, 2009; accepted after revision September 7, 2010.

