

# Adenosine Deaminase 2 Deficiency: More Than Monogenic Vasculitis

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The phrase *autoinflammatory disease* is a relatively new term that describes a group of disorders characterized by early onset recurrent or persistent inflammation, seemingly unprovoked and emerging without an infection or other apparent cause [1]. The inflammation occurs in the absence of autoantibodies or autoreactive antigen-specific T cells, a feature that distinguishes these disorders from the more common and widely known autoimmune conditions [2]. Our understanding of the biologic basis of autoinflammatory diseases has advanced significantly since Kastner proposed this term in 1999 shortly after the discovery of a genetic cause for familial Mediterranean fever [3]. Monogenic defects of genes involved in innate immunity comprise, and are associated with, most of the autoinflammatory diseases. The field of study focusing on autoinflammatory disorders is expanding rapidly.

Several classifications of autoinflammatory diseases have been proposed according to either underlying immunologic mechanism or clinical phenotype based on the major organ or system involved. One clinically defined subgroup comprises disorders formerly referred to as periodic or episodic fever syndromes. Among periodic fevers, the most common is familial Mediterranean fever (FMF) [4,5]. A new subgroup of autoinflammatory diseases includes inherited vasculopathies with autoinflammatory features.

Deficiency of adenosine deaminase 2 (DADA2) is a new autoinflammatory disorder described at 2014 by two independent groups [6,7]. During the last two decades, Israeli rheumatologists followed a group of children of Georgian-Jewish origin with familial polyarteritis nodosa (PAN) vasculitis. With recent

advances in scientific tools and using whole exome sequencing, an autosomal recessive loss of function mutation in the *cat eye syndrome chromosome region, candidate 1 (CECR1)* gene, which encodes adenosine deaminase 2 (ADA2), was associated with this vasculitis in a cohort of 24 patients [6]. All 19 Georgian-Jewish patients were homozygous for a mutation encoding a Gly47Arg substitution. Their unaffected relatives were either heterozygous for the mutation or did not carry it. In a random 246-person cohort of unrelated controls of Georgian-Jewish ancestry, 25 were heterozygous and none were homozygous for this variant, yielding a carrier frequency of 0.1 in this isolated, ancient community. This mutation was not present in more than 8000 exome sequences (public and in-house databases). The enzymatic activity of ADA2 was significantly reduced in the serum of the patients.

The age at disease onset varied widely from 2 months to 59 years. Most patients were diagnosed with polyarteritis nodosa before the age of 10 years. In six children with severe disease, the diagnosis was concluded before 1 year of age. Skin manifestations were the most common finding in Georgian-Jewish patients, and livedo reticularis, specifically, was sometimes the sole manifestation of the disease. Necrotizing arterial vasculitis, characteristic of polyarteritis nodosa, was found in some skin biopsies. Pathologic findings consistent with

**Recessive loss of function mutations in the *cat eye syndrome chromosome region, candidate 1 (CECR1)* gene causing Adenosine Deaminase 2 (ADA2) deficiency was associated with familial polyarteritis nodosa vasculopathy, an autoinflammatory disease with highly varied clinical expression**

non-specific leukocytoclastic vasculitis, panniculitis and livedoid vasculitis were seen in the remainder of the specimens. Recalcitrant leg ulcers were common, and in some patients they were the

most prominent feature, usually recurrent and healing very slowly. This condition is most probably under-diagnosed, as more patients with such ulcers are treated in dermatologic clinics and not diagnosed with DADA2. Of interest, a report from 1986 described four young Georgian-Jewish patients with recurrent leg ulcers, most of them familial and without systemic disease [8]. Visceral involvement was of great importance in the Georgian-Jewish cohort: seven patients had severe difficulty controlling hypertension without glomerulonephritis. In seven

different cases the gastrointestinal tract was involved, leading to bowel resection in some cases. Neurologic disease, which occurred in 60% of patients, affected the peripheral nervous system more commonly than the central nervous system, and consisted mainly of sensory neuropathy and cranial nerve palsy. Of great significance were brain strokes (usually ischemic), the most common central nervous system manifestation. Fever and musculoskeletal manifestations were common initial manifestations of the disease. Overall, disease severity was highly variable, even within a family. Few patients had a mild disease limited to the skin with no symptoms or signs of systemic vasculitis. The majority had severe, often nervous system and visceral polyarteritis nodosa, which was fatal for three of the patients [6].

Zhou and colleagues [7] found mutations in the *CECR1* gene in a cohort of nine patients with recurrent fevers, livedo racemosa and early onset of stroke. Functional studies proved the deleterious consequences of these mutations. Both studies [6,7] were published in the same issue of the *New England Journal of Medicine*.

In recent years, several cohorts and dozens of patients with DADA2 have been reported [9-13]. The clinical presentations of the original series described are varied and range from a vasculitis disease limited to the skin to a systemic and sometimes a fatal form of vasculopathy, which includes early onset stroke, lacunar infarctions, childhood polyarteritis nodosa, livedo reticularis, lacunar infarctions, fever, portal hypertension and Sneddon syndrome. Recently it was suggested to classify ADA2 deficiency as a vasculitis associated with a probable cause, similar to hepatitis B-associated vasculitis [12]. Additional studies have shown the disease to be extremely heterogeneous with a wide spectrum of symptoms without vasculopathy, including various hematologic, neurologic, skin and other manifestations. Patients with isolated bone marrow failure, lympho-proliferation and spastic paraplegia were included as well [14].

Recently, Ben-Ami and colleagues [9] described five Caucasian children from four unrelated Arab families, all born to consanguineous parents, who were referred to the pediatric hematology clinic at Hadassah-Hebrew University Medical Center for evaluation of diverse hematologic disorders. Using whole exome sequencing, all patients were diagnosed with ADA2 deficiency. All five patients were found to have low levels of serum ADA2 activity. Interestingly, none of the children had any manifestations of vasculitis or personal or family history suggestive of immune or inflammatory dysregulation. One patient was an 8 year old girl who presented with acute, severe hemolytic anemia (hemoglobin, 2.3 g/dl) and splenomegaly. Two patients

had severe macrocytic anemia and reticulocytopenia from early infancy, requiring regular blood transfusions. Their clinical manifestations, laboratory findings and bone marrow biopsies were typical of classic Diamond-Blackfan anemia. Their workup showed that pure red cell aplasia (PRCA) was the only mani-

### **Deficiency of adenosine deaminase 2 (DADA2) was associated with different hematologic disorders and immunodeficiency of the B-cell compartment**

festation of ADA2 deficiency. Refractory PRCA was reported in a 4 year old boy with mild immunodeficiency, alopecia areata and hepatosplenomegaly,

who was diagnosed with ADA2 deficiency. None of these five patients developed vascular manifestations, such as livedo reticularis, or experienced strokes or aneurisms during follow-up [9]. Following failure of treatment with several immuno-suppressive agents he developed progressive bone marrow failure and underwent a hematopoietic stem cell transplantation (HSCT) [15].

In a cohort of 181 patients from different countries with common variable immunodeficiency, compound heterozygous or homozygous mutations in the *CECR1* gene were found in 6% of cases [10]. The clinical spectrum of the patients ranged from isolated antibody deficiency to different inflammatory manifestations, including recurrent strokes. Compromised B-cell compartment with low memory B-cells and correlation between B-cell function and inflammatory parameters were observed.

Clinical manifestation and other disease characteristics were described in a series of nine patients with ADA2 deficiency from the Netherlands and Belgium, all with an identical homozygous R169Q mutation in the *CECR1* gene [13]. This mutation is most common in North Europe with a carrier rate of 1 in 500. All patients presented before age of 8 years. ADA2 enzyme activity was low in patients and correlated with the presence of strokes. A large phenotypic variability in patients homozygous for identical mutation was observed, as it was reported previously in patents of Georgian-Jewish ancestry. This series differed from previously reported studies. In addition to manifestations of vasculopathy, most patients had hepatosplenomegaly, cytopenia and hypogammaglobulinemia. Three patients underwent a HSCT that restored ADA2 activity and resolved the clinical symptoms [13]. In a cohort of 15 patients with ADA2 deficiency, most evaluated in England, a wide range of disease severity was described, from limited cutaneous vasculitis to severe multi-system vasculitis.

### **Therapy with anti-TNF agents is effective in treating deficiency of adenosine deaminase 2 (DADA2) and warrants genetic screening of siblings for proper diagnosis, follow-up and timely treatment**

Almost half of the patients had lymphopenia and/or hypogammaglobulinemia. One-third of the cases were asymptomatic, never having received treatment. Nanthapaisal and colleagues [11]

recommended screening of all unaffected siblings of index cases by Sanger sequencing of *CECR1* and measurement of ADA2 enzyme activity in the serum.

The pathogenesis of ADA2 is not yet fully understood. ADA2 is an intra- and extra-cellular enzyme involved in the

purine pathway, secreted mainly by monocytes and macrophages. Very low blood levels of ADA2, observed in ADA2 deficient patients, are used to confirm the condition. ADA2 functions also as a growth factor for endothelial cells and macrophages, essential for endothelial stability and inducing T-cell-dependent differentiation of monocytes into anti-inflammatory M2 macrophages [7,16,17]. ADA2 deficiency is associated with upregulation of neutrophils-expressed genes and production of inflammatory cytokines.

Although high doses of systemic corticosteroids are beneficial for many patients, most immunosuppressive medications are not effective in the treatment of more severe patients with ADA2 deficiency. Since the original report from our group [6], the beneficial effect of tumor necrosis factor (TNF) blockers was observed in most ADA2 deficient patients. Few patients were reported to be cured by successful HSCT [18].

**CONCLUSION**

In conclusion, recessive loss of function mutations of the *CECR1* gene and ADA2 deficiency were found to be a cause of autoinflammatory disease and familial polyarteritis nodosa vasculopathy with highly varied clinical expression. Recently DADA2 was associated with different hematologic disorders and immunodeficiency of the B-cell compartment.

Genetic analysis of the *CECR1* gene should be considered in various clinical conditions, including inflammatory vasculopathy with early onset or familial disease, early stroke, bone marrow failure and hypogammaglobulinemia. Therapy with anti-TNF agents is effective in treating DADA2 and warrants genetic screening of siblings for proper diagnosis, follow-up and timely treatment. HSCT should be considered in patients with severe cytopenia and bone marrow failure.

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**References**

1. Padeh S, Berkun Y. Auto-inflammatory fever syndromes. *Rheum Dis Clin North Am* 2007; 33: 585-623.
2. Berkun Y, Eisenstein EM. Update on auto-inflammatory diseases and familial Mediterranean fever. *IMAJ* 2016;18 (3-4): 221-4.
3. International FMF Consortium. Ancient missense mutations in a new member of the RoRet gene family are likely to cause familial Mediterranean fever. *Cell* 1997; 90: 797-807.
4. Padeh S, Berkun Y. Familial Mediterranean fever. *Curr Opin Rheumatol* 2016; 28(5): 523-9.
5. Gül A. Treatment of familial Mediterranean fever: colchicine and beyond. *IMAJ* 2014; 16: 281-4.
6. Navon Elkan P, Pierce SB, Segel R, et al. Mutant adenosine deaminase 2 in a polyarteritis nodosa vasculopathy. *N Engl J Med* 2014; 370: 921-31.
7. Zhou Q, Yang D, Ombrello AK, et al. Early-onset stroke and vasculopathy associated with mutations in ADA2. *N Engl J Med* 2014; 370: 911-20.
8. Suster S, Ronnen M, Bubis JJ, Schewach-Millet M. Familial atrophic blanche-like lesions with subcutaneous fibrinoid vasculitis. The Georgian ulcers. *Am J Dermatopathol* 1986; 8: 386-91.
9. Ben-Ami T, Revel-Vilk S, Brooks R, et al. Extending the clinical phenotype of adenosine deaminase 2 deficiency. *J Pediatr* 2016; 177: 316-20.
10. Schepp J, Proietti M, Frede N, et al. Screening of 181 patients with antibody deficiency for deficiency of adenosine deaminase 2 sheds new light on the disease in adulthood. *Arthritis Rheumatol* 2017.
11. Nanthapisal S, Murphy C, Omoyinmi E, et al. Deficiency of adenosine deaminase type 2: a description of phenotype and genotype in fifteen cases. *Arthritis Rheumatol* 2016; 68: 2314-22.
12. Batu ED, Karadag O, Taskiran EZ, et al. A case series of adenosine deaminase 2-deficient patients emphasizing treatment and genotype-phenotype correlations. *J Rheumatol* 2015; 42: 1532-4.
13. Van Montfrans JM, Hartman EA, Braun KP, et al. Phenotypic variability in patients with ADA2 deficiency due to identical homozygous R169Q mutations. *Rheumatology (Oxford)* 2016; 55: 902-10.
14. Caorsi R, Penco F, Schena F, Gattorno M. Monogenic polyarteritis: the lesson of ADA2 deficiency. *Pediatr Rheumatol Online J* 2016; 14: 51.
15. Hashem H, Egler R, Dalal J. Refractory pure red cell aplasia manifesting as deficiency of adenosine deaminase 2. *J Pediatr Hematol Oncol* 2017; 39 (5): e293-6.
16. Zavalov AV, Engström A. Human ADA2 belongs to a new family of growth factors with adenosine deaminase activity. *Biochem J* 2005; 391: 51-7.
17. Zavalov AV, Gracia E, Glaichenhaus N, Franco R, Lauvau G. Human adenosine deaminase 2 induces differentiation of monocytes into macrophages and stimulates proliferation of T helper cells and macrophages. *J Leukoc Biol* 2010; 88: 279-90.
18. Van Eyck L, Hershfield MS, Pombal D, et al. Hematopoietic stem cell transplantation rescues the immunologic phenotype and prevents vasculopathy in patients with adenosine deaminase 2 deficiency. *J Allergy Clin Immunol* 2015; 135: 283-7.e5.

**Capsule**

**Liver T cells in obesity-associated diabetes**

Obesity is associated with an increased risk of developing a range of disorders, including cardiovascular diseases and type 2 diabetes. In obese individuals, accumulation of fat in the liver (known as nonalcoholic fatty liver disease or NAFLD), has been linked to the development of insulin resistance. Ghazarian and colleagues found that type I interferon-driven activation of CD8+ T cells in the liver correlated with insulin

resistance in patients with NAFLD and in mouse models of obesity. Furthermore, the gut microbiome played an important role in driving inflammation in the livers of obese mice. These findings add to the growing recognition of the immune axis in metabolic disorders associated with obesity.

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