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Case Report

Autosomal dominant Hashimoto's thyroiditis with a mutation in *TNFAIP3*

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Abstract. Hashimoto's thyroiditis (HT) is an autoimmune disease thought to involve a combination of genetic and environmental factors, but its detailed pathogenesis is unknown. We present a family with haploinsufficiency of the gene encoding tumor necrosis factor α-induced protein 3 (*TNFAIP3*, also known as *A20*) and show a link with HT in a three-generation pedigree. Currently, *TNFAIP3* polymorphisms are associated with several autoimmune diseases, and haploinsufficiency of A20 was recently observed in families with an early-onset autoinflammatory disease resembling Behçet's disease. However, HT has not been linked with *TNFAIP3* variants. We analyzed *TNFAIP3* and human leukocyte antigen (*HLA*) in the family showing HT as an autosomal dominant trait, and identified a novel heterozygous c.2209delC mutation of *TNFAIP3* in the members with HT. The known HLA haplotypes linked to HT could not be identified. Based on our analysis of this pedigree, we consider HT as a possible phenotype of A20 haploinsufficiency.

Key words: Hashimoto's thyroiditis, hypothyroidism, TNFAIP3, A20, haploinsufficiency of A20

Introduction

Hashimoto's thyroiditis (HT) is an autoimmune thyroid disease thought to develop through a combination of genetic and environmental factors; however, its detailed pathogenesis remains unknown. Recently Zhou *et al*. revealed that a familial early-onset autoinflammatory disease resembling Behçet's disease (BD) inherited in an autosomal dominant manner was caused by haploinsufficiency of the gene encoding tumor necrosis factor α-induced protein 3 (*TNFAIP3*, also known as *A20*) (1). A20 is a negative regulator of multiple intracellular immune signaling pathways, including tumor necrosis factor-α (TNF-α) signaling. In this study, patients from a family with haploinsufficiency of A20 (HA20) who developed HT by autosomal dominant inheritance are reported.

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Case Presentation

Patient 1 (P1) is the proband (Fig. 1) who suffered from recurring febrile episodes with severe abdominal pain, vomiting, and bloody stools from the age of 6 mo. Family histories show

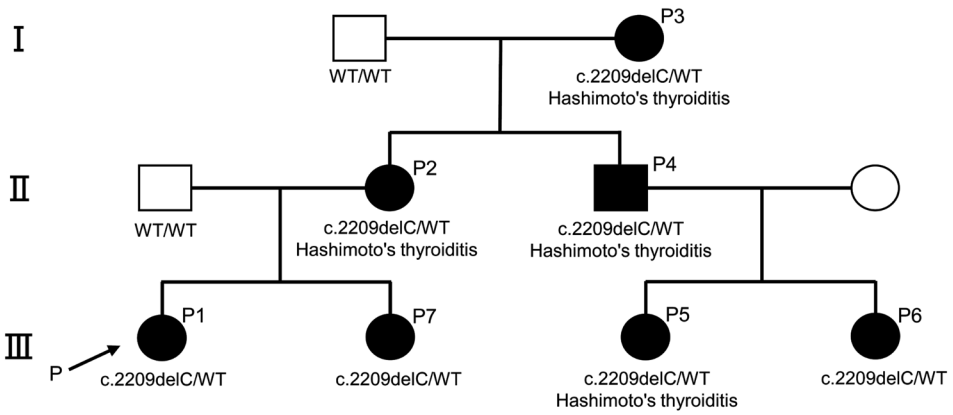

Fig. 1. Pedigree of the patients.

that individuals P2–P6 also experienced some episodes suggestive of an autoinflammatory disease, especially P2 who is P1's mother. P2 had recurrent episodes of fever and cervical lymphadenitis diagnosed as subacute necrotizing lymphadenitis and chronic mild hepatitis in her childhood. At 21 and 22 yr of age, she suffered from two febrile episodes with diarrhea and elevated C-reactive protein levels, suggestive of inflammatory bowel disease. Colonoscopy revealed possible Crohn's disease, but these symptoms were resolved spontaneously. Since childhood, P3, P1's maternal grandmother, and P4, P1's maternal uncle, have suffered from recurring episodes of stomatitis. At 21 yr of age, P4 was diagnosed with malignant lymphoma (Hodgkin's disease) and at the age of 33 yr, he was diagnosed with a brain tumor (craniopharyngioma). P5, P1's maternal cousin, experienced several episodes of fever from the age of 1 yr, whereas P6, P1's maternal cousin, suffered four episodes of fever before the age of 1 yr. Moreover, P7, P1's younger sister, had multiple perineal fistula possibly associated with inflammatory bowel disease at the age of 1 mo.

Among these patients, P2, P3, P4, and P5 were diagnosed with HT. P2 was diagnosed with severe HT at 28 yr of age (Table 1). Ultrasonography revealed a mild degree of diffuse swelling in her thyroid. She only exhibited increased weight (body mass index, 35.4 kg/

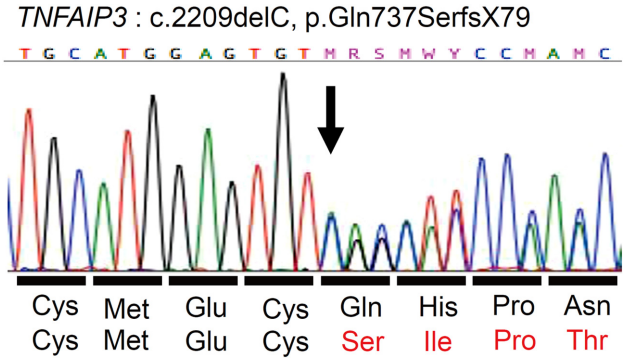

Fig. 2. Results of *TNFAIP3* analysis.

m^2) as a major clinical finding characteristic of hypothyroidism. P3 was diagnosed with HT at the age of 27 yr with symptoms such as fatigability. She has been treated with oral levothyroxine and her current anti-thyroid autoantibody levels are shown in Table 1. P4 was also diagnosed with HT during a hormone evaluation at his craniopharyngioma diagnosis. His levels of anti-thyroid autoantibodies at 34 yr of age are shown in Table 1. P5 had no clinical findings characteristic of hypothyroidism, but ultrasonography showed a mild degree of diffuse swelling in her thyroid and elevated thyrotropin (TSH) and anti-thyroglobulin antibody levels at the age of 9 yr (Table 1). P1, P6, and P7, currently 6-, 2-, and 1-yr-old, respectively, have normal thyroid functions and no anti-thyroid autoantibodies, but are too young for this to be

Table 1. Results of pre-treatment thyroid function tests and anti-thyroid autoantibodies

	P2	P3	P4	P5	Normal values
Age at HT diagnosis (yr)	28	27	34	9	
TSH (μIU/mL)	163.18	N/A [a]	55.96	6.50	0.35–4.94
Free T4 (ng/dL)	0.27	N/A [a]	0.37	1.00	0.70–1.48
TPOAb (IU/mL)	381	(18) [b]	123	10	< 16
TgAb (IU/mL)	13	(1050) [b]	126	284	< 28
TSBAb (%)	0	(0) [b]	0	0	≤ 31.7

HT; Hashimoto's thyroiditis, TPOAb; thyroid peroxidase antibody, TgAb; thyroglobulin antibody, TSBAb; TSH-stimulation blocking antibody. [a] Pre-treatment thyroid function tests are not available (N/A). [b] Anti-thyroid autoantibody levels at 67 yr of age are shown.

Table 2. Results of *HLA* allele typing

	HLA-A	HLA-B	HLA-C	HLA-DRB1
P2	02:01 24:02	40:02 54:01	01:02 03:04	04:03 14:03
P3	24:02 26:03	35:01 54:01	01:02 03:03	11:01 14:03
P4	02:06 24:02	40:02 54:01	01:02 –	04:06 14:03
P5	11:01 24:02	54:01 55:02	01:02 –	04:05 14:03

Common *HLA* haplotypes among the patients are underlined. HLA; Human leukocyte antigen.

apparent.

We analyzed *TNFAIP3* in the present family and identified a novel heterozygous c.2209delC mutation that was present in all the HT members tested (Figs. 1 and 2) (RefSeq transcript NM_006290.2). We also analyzed human leukocyte antigen (*HLA*) alleles in P2, P3, P4, and P5 who were diagnosed with HT (Table 2). All HT members appeared to have an A*24:02-B*54:01-C*01:02-DRB1*14:03 haplotype.

A brief case report mentioning this family was previously described by Kadowaki *et al*. as part of a study examining HA20 patients in nine independent Japanese families (2).

Discussion

A20 is encoded by *TNFAIP3* and inhibits the tumor necrosis factor-induced transmission pathway for nuclear factor-κB (NF-κB) signals. *TNFAIP3* polymorphisms are associated with several autoimmune diseases including Graves' disease (GD) (3). However, the pathogenic contribution of *TNFAIP3* variants to HT has not been revealed. The major clinical presentations of HA20 patients include some BD-like symptoms such as mucous membrane lesions, including recurrent stomatitis, ophthalmic lesions, dermal lesions, and articular lesions (1, 2, 4, 5). Moreover, these reports also showed clinical heterogeneity among the members of the same family. We, therefore, analyzed *TNFAIP3* in the present family (P1–P7) and identified a heterozygous c.2209delC mutation that was present in all members tested (Figs. 1 and 2). *In vitro* functional analysis of this variant was previously reported as a pathogenic mutation (2). Hence, P1–P7 were all diagnosed with HA20.

Little is known about the exact pathogenesis of HT. However, considering that certain families are more susceptible to this disease than others, it is likely that genetic factors are involved.

Previous reports suggest the possible association of HT with polymorphisms of *HLA*, cytotoxic T-lymphocyte-associated protein 4 (*CTLA4*), protein tyrosine phosphatase non-receptor type 22 (*PTPN22*), or thyroglobulin (*Tg*) genes (6). In particular, the *HLA* gene is associated with a number of autoimmune diseases. An HA20 patient demonstrating very early-onset psoriatic arthritis, spondylitis, and aortic insufficiency was reported. This patient also had another possible genetic factor, HLA-B27, which is well known to cause psoriatic arthritis and ankylosing spondylitis (2). Thus, we considered that HA20 might drive other genetic factors associated with autoimmune disorders such as *HLA*. HT is linked to particular HLA antigens or *HLA* alleles such as A2 (7), A*02:07 (8), DR3 (9), DRB1*03:01 (10), DR4 (DR53, DRB4) (7-9), DR5 (11), and DRB1*11:04 (12). Furthermore, because both the *TNFAIP3* and *HLA* are located on chromosome 6, we speculate that an association exists between *TNFAIP3* and *HLA*. As a result, P2, P3, P4, and P5 all have the DRB1*14:03 allele, which has been demonstrated as the susceptible *HLA* allele for GD in the Japanese population, but not for HT (13). To our knowledge, the identified common *HLA* allele is not reported to be associated with HT. Hence, in the present family, it is considered that *TNFAIP3* dominantly contributes to the pathogenesis of HT without any relation to *HLA*. Nonetheless, this study is limited because we cannot exclude other genetic effects such as *CTLA4*.

Moreover, some HA20 patients have been reported to show systemic as well as endocrinological autoimmune disorders, including HT, GD, and insulin-dependent diabetes (14). Additionally, a case showing GD in the previously summarized cohort of Japanese HA20 had a TSH-receptor antibody (TRAb) as well as a thyroid peroxidase antibody (TPOAb) and a thyroglobulin antibody (TgAb) (2). Furthermore, an HA20 patient accompanied with HT was reported recently (15). Excessive differentiation of T helper 17 (Th17) cells was observed in HA20 patients (1, 2). Takagi *et al*. reported an HA20 patient who presented symptoms similar to the autoimmune lymphoproliferative syndrome and whose double-negative T (DNT) cell counts were increased (4). Th17 and DNT are well known subsets of T lymphocytes that play a role in autoimmunity (16, 17). Thus, it is possible that HA20 patients have autoimmune diseases as well as autoinflammatory diseases. In addition, particular immune dysregulation disorders in primary immunodeficiency syndromes (for example, signal transducer and activator of transcription 3 (*STAT3*) gain-of-function mutations or CTLA4 deficiency) are known to complicate various autoimmune diseases including autoimmune thyroid disease (AITD) (18, 19). As mentioned above, A20 is one of the immunoregulators, and disruption of these regulators may readily cause AITD. Therefore, it is possible that *TNFAIP3* mutation contributes to the onset of AITD much more than other genetic effects such as *HLA* or *CTLA4* polymorphisms, considering the existence of this family case and other sporadic cases with AITD and the etiology. For all these reasons, HT can be considered as one of the many symptoms to be found in HA20.

This is the first familial case report of HA20 associated with HT. We propose that if HT patients demonstrate an autosomal dominant inheritance pattern and have BD-like clinical presentations such as recurrent stomatitis, a diagnosis of HA20 should be considered.

Conflicts of Interest: The authors declare that they have no conflicts of interest.

Acknowledgements

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