Clinical/Scientific Notes

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TWO NOVEL *HTRA1* MUTATIONS IN A EUROPEAN CARASIL PATIENT

Cerebral autosomal recessive arteriopathy with subcortical infarcts and leukoencephalopathy (CARASIL) is a hereditary, nonhypertensive cause of recurrent lacunar stroke and cognitive decline associated with alopecia, spondylosis deformans, and lumbago.[1] The disease has been linked to mutations in the *HTRA1* gene, encoding for serine protease HTRA1, loss of which causes dysregulation of transforming growth factor-β signaling.[2]

Since the first description,[3] most affected patients have been reported in Japanese and Chinese pedigrees with typical and atypical phenotypes. No strong genotype–phenotype correlation has yet been found, and homozygous patients with nonsense or missense mutations may be clinically indistinguishable.[2] The recent unique report of a Caucasian case suggests that the disease is not limited to Asia.[4]

Here we report a new European patient with a peculiar phenotype carrying 2 novel mutations of the *HTRA1* gene. The heterozygous proband's parents showed variable leukoencephalopathy but no clinical signs of the disease.

Case report. The proband was a 29-year-old Romanian female with unrelated parents. Family history was unremarkable. She complained of chronic lumbar and cervical pain since the age of 14 years. She had 2 ischemic strokes with left hemiparesis and dysarthria at ages 24 and 29, respectively.

Neurologic examination showed ataxic gait, gaze-evoked nystagmus, dysmetria, hypoactive deep tendon reflexes at legs, and no alopecia. Cognitive status was normal. Brain MRI showed severe diffuse leukoencephalopathy, including subcortical infarcts and evidence of microbleeds, while spine MRI showed degenerative disc disease (figure 1A). Several idiopathic and metabolic causes of ischemic strokes were excluded (MELAS [mitochondrial encephalomyopathy, lactic acidosis, and stroke-like episodes], Fabry disease, rheumatologic diseases, and vasculitis). Cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy (CADASIL) was ruled out by automatic sequencing of exons 2–24 of the *NOTCH3* gene and no evidence of granular osmiophilic material in skin and muscle biopsy. The parents were neurologically normal: the father, age 61 years, had negative history for neurologic and cardiovascular diseases and hypertension; the mother, age 60 years, reported mild hypertension under treatment since the age of 54 years without other vascular risk factors. After genetic diagnosis of the daughter, the parents underwent brain MRI. The father showed mild supratentorial leukoencephalopathy (figure 1, B1, B3, B5, B7) and the mother diffuse infratentorial and supratentorial leukoencephalopathy (figure 1, B2, B4, B6).

Methods and Results. With institutional review board approval and informed patient consent, we directly sequenced exons 1–9 of the *HTRA1* gene in the proband. Two novel heterozygous mutations were identified: the c.961G>A in exon 4 (inherited from the father) and a G deletion (c.126delG) in exon 1 (inherited form the mother). The missense mutation in exon 4 results in substitution of a highly conserved alanine residue with threonine (p.Ala321Thr). This mutation is predicted to be pathogenic by PolyPhen-2, SIFT, and Mutation Taster software. The deletion in exon 1 causes a frameshift, altering the amino acid sequence from position 42 (p.Glu42fs), and introduces a stop codon at position 214 (figure e-1 on the *Neurology*® Web site at Neurology.org). Neither mutation was found in public databases (http://browser.1000genomes.org/index.html; http://www.ncbi.nlm.nih.gov/projects/SNP/, build 137) or 320 control chromosomes.

Discussion. We here report a compound heterozygosity with 2 new mutations of the *HTRA1* gene causing CARASIL in a patient with unrelated parents. The deletion (c.126delG) in exon 1, causing a frameshift of the protein with a truncated product, was inherited from the mother. The missense mutation in the protease domain of the *HTRA1* gene (p.Ala321Thr) was inherited from the father. The patient did not have alopecia, usually considered a phenotypic marker of the disease. However, 3 women and 1 man without alopecia have been described among CARASIL subjects.[2,5] If further confirmed, this observation could suggest a sex-linked difference in the presence of alopecia in the CARASIL population. The number of female patients would be underestimated, misrepresenting the male: female ratio of 3:1 in clinically defined CARASIL cases.[1]

Our patient had severe leukoencephalopathy with temporal lobe involvement, the latter an overlapping

Supplemental data at Neurology.org