A Novel Presenilin-1 Mutation (Leu85Pro) in Early-Onset Alzheimer Disease With Spastic Paraparesis

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Background: Early-onset familial Alzheimer disease is caused by mutations in the amyloid precursor protein (APP), presenilin-1 (PSEN1), or presenilin-2 (PSEN2) genes. Phenotypic diversity has been reported to be associated with various mutations in PSEN1. Various mutations of PSEN1 have been reported in cases of early-onset Alzheimer disease with spastic paraparesis.

Objective: To describe a novel mutation in the PSEN1 gene associated with early-onset Alzheimer disease with spastic paraparesis.

Patient and Methods: The patient was a 27-year-old man who developed early-onset dementia with spastic paraparesis. We examined sequences of the PSEN1, PSEN2, and APP genes from the patient and his family. To detect a possible mutation effect on the production of amyloid-β peptide (Aβ), transfected HEK293 cells were examined for Aβ42 and Aβ40 production.

Results: We found a novel mutation (Leu85Pro) in PSEN1. This mutation influenced the production of Aβ, resulting in a 2-fold elevation of Aβ42 production and of the Aβ42/40 ratio.

Conclusion: To our knowledge, this is the first report of very early-onset Alzheimer disease with spastic paraparesis and with the visual variant form of the disease, which is associated with visuospatial cognitive disorder. The Leu85Pro mutation in PSEN1 was pathogenic.

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EARLY-ONSET FAMILIAL ALZHEIMER disease (AD) is caused by mutations in the amyloid precursor protein (APP), presenilin-1 (PSEN1), or presenilin-2 (PSEN2) genes. Phenotypic diversity has been reported to be associated with various mutations in PSEN1. Studies of early-onset AD with spastic paraparesis (SP) have previously identified 8 missense mutations, 2 deletions, and 1 insertion in PSEN1. It remains unclear how the mutation is involved in the pathological cascade, particularly in the production of amyloid-β peptide 42 (Aβ42).

In this article, we report a case exhibiting early-onset AD with SP accompanied by a novel PSEN1 mutation (Leu85Pro). Cells cultured to express PSEN1 with this mutation produced a 2-fold elevation of Aβ42 production and an increase in the Aβ42/40 ratio.

REPORT OF A CASE

A 27-year-old Japanese man was admitted to our hospital for evaluation because of the difficulties he was experiencing in coping with his daily life. He had lived in the United States since the age of 12 years. He was a good basketball player and captain of his high school team. After he entered a university in California, he gradually became withdrawn and unmotivated. Eventually, he gave up basketball and other forms of exercise. Despite his condition, he finally managed to graduate from the university at the age of 25 years, taking 2 years longer than usual to complete his degree. At the age of 26 years, he had difficulty driving a car and sometimes became lost while driving. Around this time, he also became aware of hand tremor and memory impairment. By the age of 27 years, he found that he was unable to cope with living alone, and he returned to Japan. His parents noticed that he could not cook, use the telephone, or bathe himself. He did not have a history of alcohol or drug abuse. No other family members showed similar signs or symptoms.

On hospital admission, he was alert and oriented. Neurological examination showed that he was neither interested in his condition nor aware of its deteriora-
Genomic DNA was examined in the coding regions of the PSEN1, PSEN2, and APP genes, using an ABI PRISM model 310 sequencer (PerkinElmer, Fremont, Calif). APOE genotypes were determined as described previously. All materials were obtained with informed consent after an appropriate consultation. This study received prior approval from the institutional ethics committee of Osaka City University Medical School, Osaka, Japan. To examine the effect of the novel mutation (described later) on Aβ production by APP, we compared 2 PSEN1 complimentary DNA constructs with or without the mutation. The mutant PSEN1 complimentary DNA (T254C) was prepared by site-directed mutagenesis and introduced into the pCI mammalian expression vector (Promega). Wild-type and mutant PSEN1 complimentary DNA were cotransfected with mutant APP (Swedish) complimentary DNA into human embryonic kidney 293 (HEK293) cells by lipofection (Lipofectamine; Life Technologies, Gaithersburg, Md). Two days after transfection, conditioned media were collected and assayed for Aβ40 and Aβ42 by enzymelinked immunosorbent assay (BioSource International, Camarillo, Calif). To examine the protein expression level of each transfected cell, cells were lysed in 1% Triton-X100 saline containing protease inhibitors (Sigma, St Louis, Mo) and centrifuged at 10000 g for 15 minutes at 4°C. The supernatant was subjected to Western blotting for APP and actin and also immunoprecipitated with anti-PSEN1 C-terminal fragment antibody (a rabbit polyclonal antibody recognizing the C-terminal fragment of APP) for Western blotting of PSEN1. The samples were electrophoresed on NuPAGE 4% to 12% Bis-Tris gels (Invitrogen, Purchase, NY) and transferred onto polyvinylidene difluoride membranes. The membrane was blocked and subsequently incubated in the primary antibody solution (a rabbit polyclonal antibody that recognizes the C-terminus of APP, a mouse monoclonal antibody that recognizes the C-terminal fragment of PSEN1, or an antiactin antibody purchased from Sigma). Bound antibody was visualized using horseradish peroxidase–conjugated secondary antibody and ECL Plus (Amersham Pharmacia Biotech Inc, Piscataway, NJ). Amyloid-β40 and Aβ42 in cerebrospinal fluid and serum of the patient and the control were assayed by enzyme-linked immunosorbent assay as described earlier.

Sequence analysis showed that the patient had a novel PSEN1 mutation in exon 4, at nucleotide position 254 (T to C) in 1 PSEN1 allele, indicating an amino acid change from leucine to proline at position 85 (Leu85Pro) (Figure 2A). There was no other mutation detected in the coding regions of PSEN1, PSEN2, or APP. No other family members, including the patient’s parents and 2 siblings, had any mutations in PSEN1, PSEN2, or APP.

Figure 1. Neuroimaging study of the patient. A, Axial T1-weighted magnetic resonance imaging of the brain showed mild diffuse cortical atrophy by the standards for his age. Single-photon emission computed tomography (B) and positron emission tomography (C) showed hypoperfusion and hypometabolism in the bilateral occipital and temporal lobes.
We found a novel heterozygous mutation in PSEN1 (T254C, resulting in Leu85Pro within the first transmembrane domain) of a patient with very early-onset AD with SP. Because the mutation detected in this case resulted in a marked increase in Aβ42 production and in the Aβ42/40 ratio, it is almost certainly causative. Several previous studies have shown other PSEN1 mutations to be causative in familial AD with SP. Only 1 mutation is associated with SP in the same domain.3 Unfortunately, we could not examine the paternity, because the family members refused consent to test it. The spontaneous mutation rates in the disease range from 10⁻⁴ to 10⁻⁷ per locus per generation,19 although the rates would vary according to their gene sizes. If the mutation of our case were de novo, this would be a rarity.

Neuropsychological evaluation of the patient revealed a complex visual problem in addition to impairment of intelligence and memory. Single-photon emission computed tomography and positron emission tomography examinations showed bilateral hypoperfusion and hypometabolism in the occipital and temporal lobes. These findings are compatible with the diagnosis of the visual variant of AD, as previously described by Levine et al.20 rather than typical AD. Although neuropathological confirmation was not available in this patient, his symptoms and neuroimaging data were consistent with the visual variant form of AD. We are able to exclude other neurological diseases leading to juvenile dementia. The present case is noteworthy because the patient was younger than other patients with visual variant AD, and this is the first report, to our knowledge, of early-onset AD with SP and with the visual variant form of the disease. The relationship between the Leu85Pro PSEN1 mutation and the unique visuospatial cognitive disorder requires further study.

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