OBSERVATION

Mitochondrial Transfer RNA^{Phe} Mutation Associated With a Progressive Neurodegenerative Disorder Characterized by Psychiatric Disturbance, Dementia, and Akinesia-Rigidity

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Background: Mitochondrial diseases are characterized by wide phenotypic and genetic variability, but presentations in adults with akinetic rigidity and hyperkinetic movement disorders are rare.

Objectives: To describe clinically a subject with progressive neurodegeneration characterized by psychosis, dementia, and akinesia-rigidity, and to associate this phenotype with a novel mitochondrial transfer RNA^{Phe} (tRNA^{Phe}) (*MTTF*) mutation.

Design, Setting, and Patient: Case description and detailed laboratory investigations of a 57-year-old woman at a university teaching hospital and a specialist mitochondrial diagnostic laboratory.

Results: Histopathological findings indicated that an underlying mitochondrial abnormality was responsible for the subject's progressive neurological disorder, with mitochondrial genome sequencing revealing a novel m.586G>A *MTTF* mutation.

Conclusions: The clinical phenotypes associated with mitochondrial disorders may include akinesia-rigidity and psychosis. Our findings further broaden the spectrum of neurological disease associated with mitochondrial tRNA^{Phe} mutations.

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MITOCHONDRIAL DNA (mtDNA) mutations are associated with markedly heterogeneous phenotypic and genetic profiles, are not uncommon, and are probably underdiagnosed.[1] Mitochondria may play a critical role in the etiopathogenesis of Parkinson disease, and mutations in several nuclear-encoded genes linked with inherited Parkinson disease have been hypothesized to result in mitochondrial dysfunction.[2] In contrast, presentations of primary mitochondrial gene defects as extrapyramidal movement disorders in adults appear to be rare. Nelson et al[3] described a 53-year-old patient with suspected Huntington disease (HD) who on histochemical grounds was found to have a complex I defect associated with a heteroplasmic mitochondrial transfer RNA^{Trp} (tRNA^{Trp}) (m.5549G>A) mutation. There are additional single case reports of patients with proven or suspected mitochondrial cytopathy who have manifested chorea.[4,5] Herein, we describe a subject with a progressive neurodegenerative disorder characterized by akinesia-rigidity, abnormal movements, dementia, and psychiatric disorder associated with a novel mtDNA mutation (m.586G>A) in the tRNA^{Phe} (*MTTF*) gene (GenBank NC_012920).

Video available online at www.archneurol.com

METHODS

PATIENT

The proband is a 57-year-old, right-handed, white woman who was referred to psychiatric services with self-neglect, poor mobility, and weight loss. Twenty years previously, she had been involved as a passenger in a serious motor vehicle crash with loss of consciousness and flail ribs but had made a rapid recovery to apparent full health. She had seen an ear-nose-throat specialist for worsening deafness 12 years previously, but no action had ensued. Over a period of 8 years, she had developed progressively worsening mobility and an increasing number of falls. Over the last 2 years, frequent facial grimacing had been noted. During the previous year, she had been able to walk only short distances, holding on to furniture with a stooped posture. Additionally, her short-

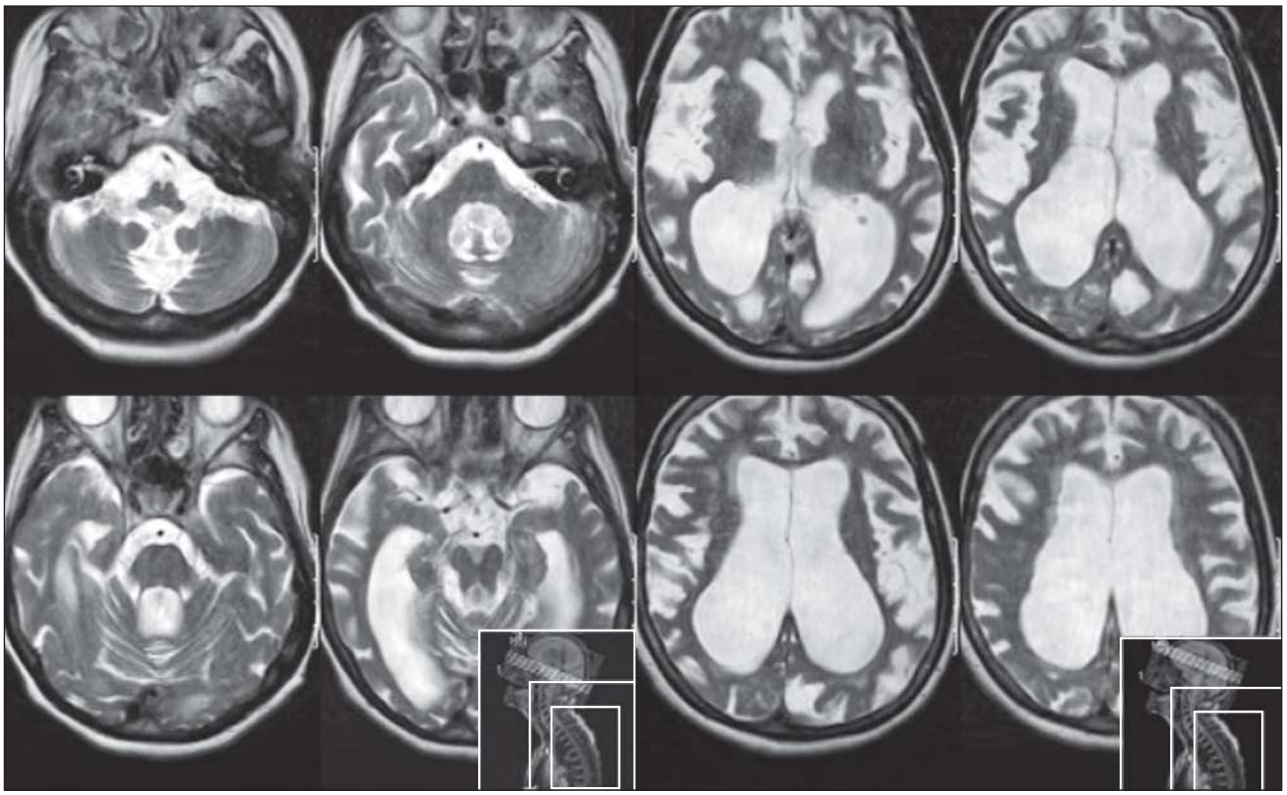

Figure 1. Magnetic resonance imaging scan of the brain (axial sections, T2-weighted) showing brain stem, cerebellar, and widespread cortical atrophy.

term memory and word-finding ability had deteriorated markedly in recent months, and she had paranoid delusions and second-person auditory hallucinations. There was no history of medicine use (including phenothiazines), alcohol consumption, or smoking. She was an only child whose father had died of multiple strokes and whose mother had recently died, in her early 80s, of metastatic breast cancer. She had previously worked as a doctors' receptionist, having gone to secretarial college, but she had been unemployed for many years at the time of presentation. She had 1 child, a son who at the time of presentation was well and in his early 20s.

On examination, she was stooped in posture and wheelchair-bound and had intermittent facial grimacing. She had a reactive affect with distractibility. She experienced auditory hallucinations (her son's voice talking to her) during the examination and did not fulfill criteria for clinical depression. The Mini-Mental State Examination score was 23/30 with deficits in orientation, recall, and repetition. She was unable to complete the Luria 3-step hand sequences, although no primitive reflexes were elicited. Cranial nerve examination results were normal except for profound sensorineural deafness left worse than right clinically and slurred speech with phonemic paraphasias. There was a mild symmetrical proximal pattern limb weakness with wasting of the quadriceps. She had spastic legs with brisk reflexes and bilaterally up-going plantar responses. Ataxia was evident both on finger-nose testing and on gait assessment (video, http://www.archneurol.com).

Formal neuropsychology testing revealed considerably slow responses (bradyphrenia). She was partly oriented in time and place and answered questions of current, recent, and past events variably, suggesting uneven memory skills. She presented with speech production and word retrieval difficulties. On selected tests of executive function (semantic and phonemic word fluency, interpretation of common proverbs, cognitive estimates), she consistently performed very poorly and showed perseverative tendencies. Her overall neuropsychological profile was thought to be consistent with predominantly frontal lobe and/or subcortical cerebral dysfunction and was also suggestive of a degree of temporal involvement.

Results of the following investigations were normal or negative: urea and creatinine levels, electrolyte levels, full blood cell count, liver enzyme tests, thyroid function tests, C-reactive protein level, antinuclear antibodies, thyroid peroxidase antibodies, treponemal serology, anti-Yo/anti-Hu/anti-Ri antibodies, and serum ceruloplasmin level. The creatine kinase level was mildly elevated at 164 IU/L (reference range <145 IU/L) (to convert to microkatals per liter, multiply by 0.0167). Magnetic resonance imaging of the brain revealed generalized cerebral, cerebellar, and brainstem atrophy (**Figure 1**). The venous lactate level was 14.32 mg/dL (reference range 4.50-19.82 mg/dL) (to convert to millimoles per liter, multiply by 0.111), and the cerebrospinal fluid (CSF) lactate level was elevated at 25.95 mg/dL (reference range 10.81-18.92 mg/dL). Other CSF studies revealed less than 1 white blood cell and 14 red blood cells per cubic millimeter, normal cytological examination results, and negative culture results for pathogens. The CSF protein level was 0.061 g/dL (reference range <0.045 g/dL) (to convert to grams per liter, multiply by 10), and the CSF glucose level was normal. Identical oligoclonal bands were seen in CSF and blood.

Molecular genetic testing for the *SCA1*, *SCA2*, *SCA3*, *SCA6*, *SCA7*, dentatorubral-pallidoluysian atrophy, and HD mutations all proved negative.

ANALYSIS

Needle muscle biopsy of the left quadriceps was undertaken, and standard histological and histochemical analyses were performed. In addition, total DNA extraction was performed on muscle and blood using standard techniques, and direct sequencing of the entire mitochondrial genome was performed.

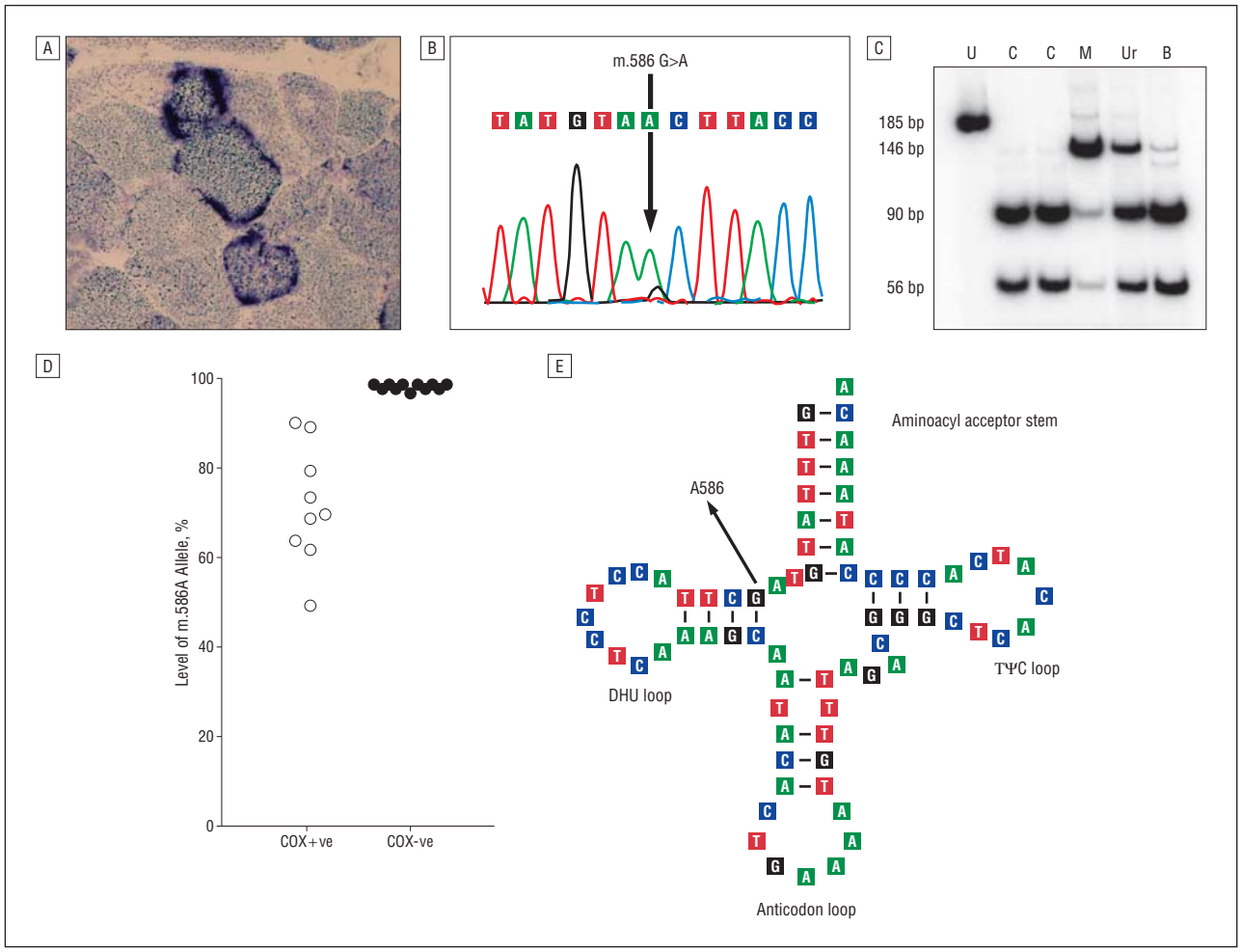

Figure 2. Identification and characterization of a novel mitochondrial DNA mutation. A, Histochemical demonstration of succinate dehydrogenase activity showing strongly staining succinate dehydrogenase–positive fibers with evidence of mitochondrial accumulation. B, Sequencing electropherogram demonstrating the heteroplasmic m.586G>A transition in the patient's muscle. C, Polymerase chain reaction–restriction fragment length polymorphism analysis in different tissues, showing the highest levels of the mutation (146–base pair [bp] product) in muscle (M) and lower levels in urinary epithelial cells (Ur) and blood (B). U indicates uncut polymerase chain reaction product; C, control. D, Graphical representation of single-fiber polymerase chain reaction analysis, clearly demonstrating the mutation segregating with cytochrome-*c* oxidase (COX) deficiency in isolated muscle fibers. COX+ve indicates COX positive; COX-ve, COX deficient. E, Schematic representation of the mitochondrial transfer RNA^{Phe} cloverleaf structure, illustrating the position of the mutation that affects a Watson-Crick base pair within the D-stem. DHU indicates dihydrouridine.

RESULTS

Histological and histochemical analyses of the muscle biopsy revealed ragged-red fibers on modified Gomori trichrome and succinate dehydrogenase staining (**Figure 2**A), with sequential cytochrome-*c* oxidase (COX) and succinate dehydrogenase histochemistry revealing marked numbers (approximately 30%) of COX-deficient fibers (not shown). Although insufficient muscle tissue was available for respiratory chain enzyme studies, we felt that the histological and histochemical changes observed were consistent with a diagnosis of mitochondrial myopathy prompting further investigations.

Long-range polymerase chain reaction of muscle DNA excluded mtDNA rearrangements, whereas sequencing of the entire mitochondrial genome detected a previously unrecognized m.586G>A transition in the *MTTF* gene (Figure 2B). Polymerase chain reaction–restriction fragment length polymorphism analysis showed that the mutation was heteroplasmic and present at the highest level in the patient's muscle (85% mutant load), with lower levels in urinary epithelia (29%) and blood (3%) (Figure 2C). Single muscle fiber analysis was performed to determine whether the amount of mutated mtDNA correlated with the observed biochemical phenotype in individual cells. Significantly higher levels of the m.586G>A mutation were present in COX-deficient fibers (mean [SD], 99.4% [0.2%]; n=11) than in COX-positive fibers (mean [SD], 72.1% [8.5%]; n=9) ($P<.001$, 2-tailed *t* test), confirming segregation of the m.586A genotype with respiratory chain dysfunction and thus establishing pathogenicity (Figure 2D).

The patient's mother had died 2 years earlier of metastatic breast carcinoma. She had no history of deafness, psychiatric illness, or any type of neurological disease. Following ethical approval, it proved possible to extract DNA from the paraffin-embedded breast tissue samples. This revealed the same m.586G>A mutation at a level of 13%, indicating maternal transmission of the mitochondrial genetic defect.

COMMENT

We consider the described m.586G>A mtDNA mutation to be pathological for the following reasons: the histopathological features were strongly suggestive of mtDNA involvement and no other mutation was detected on sequencing the entire mitochondrial genome; the mutation was heteroplasmic and present at higher levels in postmitotic skeletal muscle than mitotic cells; it is not listed as a polymorphic variant in large, publicly available databases of human mtDNA sequences; single muscle fiber analysis clearly demonstrated that the mutation segregates with COX-deficient fibers (Figure 2D); and the mutation affects a nucleotide that is phylogenetically conserved within the $tRNA^{Phe}$ structure (http://mamit-trna.u-strasbg.fr) located in the D-stem, thus predicting disruption of a Watson-Crick base pair (Figure 2E). Furthermore, the identical variant has been previously documented as a somatic mtDNA mutation in aging, COX-deficient human colonic epithelia.[6]

Of particular clinical interest here is the association of a pathological mtDNA mutation with an extrapyramidal phenotype. Our patient's clinical presentation with psychiatric disturbance, dementia, and akinesia-rigidity was otherwise unexplained, and previous clinical considerations had included a late effect of her traumatic brain injury and a neurodegenerative disorder such as progressive supranuclear palsy or HD. She had developed abnormal facial movements and rigidity without exposure to any antipsychotic medication. From a neuropsychological perspective, her presentation was indicative of predominant frontal lobe dysfunction, which would have been in keeping with progressive supranuclear palsy or HD (subcortical dementia). The development of bilateral sensorineural hearing impairment was a potential red flag for a mitochondrial cytopathy.

The current case most resembles the patient described by Nelson et al.[3] Their patient too had become symptomatic in his 40s, but his phenotype (comprising personality change and/or aggression, dementia, and widespread chorea) was suggestive of HD. Additionally, their patient had bilateral sensorineural deafness, cerebellar ataxia, and extensor plantar responses, like our subject. Computed tomography revealed widespread atrophy in that case too, and at autopsy diffuse neuronal loss and gliosis throughout the brain were confirmed. While mitochondrial dysfunction may play a central role in the development of Parkinson disease and possibly other extrapyramidal disorders, this clinical phenotype has rarely been associated with mitochondrial genetic defects. In this context, the current observations may be of pivotal importance in our understanding of parkinsonism, and greater scrutiny in patients with otherwise unexplained akinetic-rigid and other movement disorders for mitochondrial dysfunction may be warranted.

Intrinsic mitochondrial genetic factors such as heteroplasmy and mitotic segregation lead to variable mutation loads between different tissues and between individuals within the same family. Here the proband's mother appears to have been clinically unaffected, presumably due to the low levels of the m.586G>A mutation.

In conclusion, our case expands the range of clinical phenotypes associated with mtDNA mutations and contributes to a growing list of pathogenic mtDNA mutations now described in the mitochondrial $tRNA^{Phe}$ gene.[7]

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REFERENCES

1. Tuppen HA, Blakely EL, Turnbull DM, Taylor RW. Mitochondrial DNA mutations and human disease. *Biochim Biophys Acta.* 2010;1797(2):113-128.
2. Schapira AH. Mitochondria in the aetiology and pathogenesis of Parkinson's disease. *Lancet Neurol.* 2008;7(1):97-109.
3. Nelson I, Hanna MG, Alsanjari N, Scaravilli F, Morgan-Hughes JA, Harding AE. A new mitochondrial DNA mutation associated with progressive dementia and chorea: a clinical, pathological, and molecular genetic study. *Ann Neurol.* 1995;37(3):400-403.
4. Nakagaki H, Furuya J, Santa Y, Nagano S, Araki E, Yamada T. A case of MELAS presenting juvenile-onset hyperglycemic chorea-ballism [in Japanese]. *Rinsho Shinkeigaku.* 2005;45(7):502-505.
5. Caer M, Viala K, Levy R, et al. Adult-onset chorea and mitochondrial cytopathy. *Mov Disord.* 2005;20(4):490-492.
6. Taylor RW, Barron MJ, Borthwick GM, et al. Mitochondrial DNA mutations in human colonic crypt stem cells. *J Clin Invest.* 2003;112(9):1351-1360.
7. Deschauer M, Swalwell H, Strauss M, Zierz S, Taylor RW. Novel mitochondrial transfer RNA^{Phe} gene mutation associated with late-onset neuromuscular disease. *Arch Neurol.* 2006;63(6):902-905.