TUBB4A de novo mutations cause isolated hypomyelination

ABSTRACT

Objective: We present a series of unrelated patients with isolated hypomyelination, with or without mild cerebellar atrophy, and de novo TUBB4A mutations.

Methods: Patients in 2 large institutional review board–approved leukodystrophy bioregistries at Children’s National Medical Center and Montreal Children’s Hospital with similar MRI features had whole-exome sequencing performed. MRIs and clinical information were reviewed.

Results: Five patients who presented with hypomyelination without the classic basal ganglia abnormalities were found to have novel TUBB4A mutations through whole-exome sequencing. Clinical and imaging characteristics were reviewed suggesting a spectrum of clinical manifestations.

Conclusion: Hypomyelinating leukodystrophies remain a diagnostic challenge with a large percentage of unresolved cases. This finding expands the phenotype of TUBB4A-related hypomyelinating conditions beyond hypomyelination with atrophy of the basal ganglia and cerebellum. TUBB4A mutation screening should be considered in cases of isolated hypomyelination or hypomyelination with nonspecific cerebellar atrophy.

GLOSSARY

DYT4 = dystonia type 4; H-ABC = hypomyelination with atrophy of the basal ganglia and cerebellum; TUBB4A = tubulin, beta 4A class IVa.

Hypomyelinating leukodystrophies remain a diagnostic challenge with a large percentage of unresolved cases.1 Herein, we report on a series of unrelated patients with isolated hypomyelination, with or without mild cerebellar atrophy, and de novo TUBB4A mutations.

Mutations in TUBB4A (MIM 602662) are known to cause either dystonia type 4 (DYT4 [MIM 128101]) or hypomyelination with atrophy of the basal ganglia and cerebellum (H-ABC [MIM 612438]).2,3 In DYT4, an autosomal dominant mutation (c.4C>G [p.Arg2Gly]) in TUBB4A was identified in patients presenting with a “whispering” dysphonia, generalized dystonia, and gait ataxia, but normal MRI features.2 In H-ABC, a cohort of 11 individuals were found to have a common de novo mutation at c.745G>A (p.Asp249Asn) in TUBB4A.3

H-ABC is a rare leukodystrophy diagnosed on the basis of distinctive MRI findings including hypomyelination, cerebellar atrophy, and absence or disappearance of the putamen at an early age.5,5 Individuals with H-ABC present with developmental delay, extrapyramidal movement disorders (dystonia, choreoathetosis, rigidity, opisthotonos, and oculogyric crises), ataxia, and spastic tetraplegia with variable onset and in some cases seizures.4,5

Herein, we describe novel de novo mutations in TUBB4A in 5 patients belonging to 4 families with hypomyelinating leukodystrophy, and who lack the full complement of features associated with H-ABC. This finding expands the phenotype of TUBB4A-related hypomyelinating conditions beyond H-ABC and suggests that TUBB4A should be considered in cases of isolated hypomyelination.

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Go to Neurology.org for full disclosures. Funding information and disclosures deemed relevant by the authors, if any, are provided at the end of the article.
METHODS Standard protocol approvals, registrations, and patient consents. This study was performed under the approval of institutional review boards at Children’s National Medical Center or Montreal Children’s Hospital. Blood was collected and DNA extracted with informed consent from all subjects and their parents. The DNA was analyzed using exome sequencing as previously described. Of note, these cases were tested as part of a larger cohort analysis in patients with unsolved leukodystrophies, and not due to any prior suspicion of TUBB4A-related disorders. Analysis was performed on trio or great family groups in all cases. Sanger sequencing was used to validate and perform segregation analysis of all candidate mutations.

RESULTS In all 5 cases, we identified novel de novo mutations in TUBB4A (table). All of the patients have similar MRI features including hypomyelination but did not present with severe basal ganglia involvement characteristic of H-ABC (figure 1). These 5 patients presented with a diverse clinical spectrum (see supplemental data on the Neurology® Web site at Neurology.org) as well as a broad range of neuroradiologic features, some of which are also seen in H-ABC (table). Patient 4 presented with cerebellar atrophy from a young age while patients 3 and 5 have isolated hypomyelination. Patients 1 and 2 have global atrophy, as often seen in patients with long-standing hypomyelinating disorders.

DISCUSSION These patients were ascertained as cases of unsolved hypomyelination without any radiologic or clinical features permitting a more specific diagnosis. These cases unexpectedly all had de novo TUBB4A mutations distinct from the original H-ABC–related mutation, at c.745G>A (p.Asp249Asn). The c.845G>C variant shared by patients 1 and 2 was not identified in either parent when tested by exome or Sanger sequencing. We hypothesize that there is likely low-level parental mosaicism, as was identified in the sibling group in the original description of TUBB4A mutations in H-ABC. Unfortunately, because of the older age of these patients (in their fifth decade), it was no longer possible to obtain new parental samples to test this hypothesis. In the other families, parental testing by exome and standard sequencing did not identify these variants in the parents.

Missense mutations in genes that code for α- and β-tubulin proteins, essential for assembly of neuronal microtubules, have been shown to cause a group of neurologic disorders characterized by abnormal neuronal migration, differentiation, axon guidance, and maintenance. Mutations in TUBA1A (MIM 605742), TUBA8 (MIM 605742), TUBB2B (MIM 612850), TUBB3 (MIM 602661), and TUBB4A have all contributed to this spectrum of disorders.

The clinical variability between DYT4 and H-ABC (late-childhood or juvenile-onset dystonia vs severe pediatric-onset dystonia and quadriplegia) suggests...
that the TUBB4A gene is associated with a spectrum of clinical manifestations. Other genes associated with hypomyelinating leukodystrophies are also known to cause a disease spectrum, varying from severe spastic quadriparesis with limited functional ability to mild spastic paraplegia. For example, PLP1 (MIM 300401) mutations cause Pelizaeus-Merzbacher disease (MIM 312080) as well as spastic paraplegia 2 (MIM 312920). Similar examples exist for Pol III–related leukodystrophies (4H or Hypomyelination with Hypogonadotropic Hypogonadism and Hypodontia) and Pelizaeus-Merzbacher–like disease caused by GJC2 mutations. It is therefore reasonable to predict that TUBB4A mutations could present similar variability in the context of hypomyelinating leukodystrophy.

TUBB4A is a neuronally expressed member of the β-tubulin protein family, and forms heterodimers with α-tubulins. The αβ heterodimers polymerize to form microtubules, an essential component of the cytoskeleton. The previously described residues associated with H-ABC (p.Asp249) and DYT4 (p.Arg2) both sit near the intradimer interface between α- and β-tubulin (figure 2). The c.763G>A de novo mutation found in patient 4 results in the amino acid change p.Val255Ile; this residue is located in the same α-helix as p.Asp249 (figure 2). Val255 is exposed to the intradimer interface. Mutation at this position may affect heterodimer formation or stability.

The c.845G>C variant shared by patients 1 and 2 results in the amino acid change p.Arg282Pro and is located in the structure known as the M-loop (figure 2). The M-loop extends from the side of tubulin monomers and is largely responsible for stabilizing the lateral contact between adjacent tubulin protofilaments. The conformational change resulting from the introduction of a proline into the middle of the M-loop is likely to destabilize this interaction. The p.Q292K change caused by the c.874C>A mutation in patient 3 is located in the α-helix H9; this helix is also believed to participate in the lateral contacts between adjacent protofilaments. Finally, the c.1172G>A mutation in patient 5 results in the amino acid change p.Arg391His. This residue is located near the interdimer interface.
Thus this mutation may affect the polymerization of heterodimers. Overall, these mutations are hypothesized to have a similar impact of tubulin polymerization and stability as the previously described c.745G>A (p.Asp249Asn) in TUBB4A identified in H-ABC. Although all patients have hypomyelination with or without cerebellar atrophy, as seen in H-ABC, no patients in the cohort have putamen atrophy, despite nearly 5 decades of disease progression in patients 1 and 2. This suggests that mutations in TUBB4A other than p.Asp249Asn can result in a phenotype of isolated hypomyelination, albeit with or without nonspecific cerebellar atrophy.

Further monitoring is required to determine the full clinical spectrum of these mutations because it is difficult to ascertain what may develop over time, in particular for the younger 2 patients. However, even if it is possible that these patients could develop additional imaging features consistent with H-ABC over time, it remains important, based on these findings, to consider TUBB4A mutation screening in cases of isolated hypomyelination or hypomyelination with nonspecific cerebellar atrophy.

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