CLINICAL/SCIENTIFIC NOTE OPEN ACCESS

Biallelic COX10 Mutations and PMP22 Deletion in a Family With Leigh Syndrome and Hereditary Neuropathy With Liability to Pressure Palsy

Yasuko Kuroha, MD, PhD,* Takanobu Ishiguro, MD, PhD,* Mari Tada, MD, PhD, Norikazu Harada, MD, MPH, Kei Murayama, MD, PhD, Izumi Kawachi, MD, PhD, Kensaku Kasuga, MD, PhD, Akinori Miyashita, MD, PhD, Arika Hasegawa, MD, PhD, Tetsuya Takahashi, MD, PhD, Nae Matsubara, MD, PhD, Osamu Onodera, MD, PhD, Akiyoshi Kakita, MD, PhD, Ryoko Koike, MD, PhD, and Takeshi Ikeuchi, MD, PhD

Neur Online Genet 2022;8:e200030. doi:10.1212/NXG.0000000000200030

Correspondence
Dr. Ikeuchi
ikeuchi@bri.niigata-u.ac.jp

Abstract

Objectives
Leigh syndrome is a progressive encephalopathy characterized by symmetrical lesions in brain. This study aimed to investigate the clinicopathologic and genetic characteristics of a family with Leigh syndrome and hereditary neuropathy with liability to pressure palsy (HNPP).

Methods
Data from a Japanese family's clinical features, MRIs, muscle biopsy, and an autopsy were analyzed. A whole-exome sequence was performed, as well as real-time PCR analysis to determine copy number variations and Western blot analyses.

Results
The proband and her 2 siblings developed spastic paraplegia and mental retardation during childhood. The proband and her sister had peripheral neuropathy, whereas their father developed compression neuropathy. Leigh encephalopathy was diagnosed neuropathologically. Brain MRI revealed changes in cerebral white matter as well as multiple lesions in the brainstem and cerebellum. Muscle biopsy revealed type 2 fiber uniformity and decreased staining of cytochrome c oxidase. The COX10 missense mutation was identified through whole-exome sequence. A 1.4-Mb genomic deletion extending from intron 5 of COX10 to PMP22 was detected.

Discussion
These findings suggest that in this family, Leigh syndrome is associated with a mitochondrial respiratory chain complex IV deficiency caused by biallelic COX10 mutations coexisting with HNPP caused by heterozygous PMP22 deletion.

*These authors contributed equally to this work as co-first authors.

From the Department of Neurology (Y.K., A.H., T.T., N.M., R.K.), Nishinigata Chuo Hospital, Niigata; Department of Molecular Genetics (Y.K., T. Ishiguro, N.H., K.K., A.M., T. Ikeuchi), Department of Neurology (T. Ishiguro, I.K., O.O.), and Department of Pathology (M.T., A.K.), Brain Research Institute, Niigata University; and Department of Metabolism (K.M.), Chiba Children's Hospital, Japan.

Go to Neurology.org/NG for full disclosures. Funding information is provided at the end of the article.

The Article Processing Charge was funded by the AMED.

This is an open access article distributed under the terms of the Creative Commons Attribution-NonCommercial-NoDerivatives License 4.0 (CC BY-NC-ND), which permits downloading and sharing the work provided it is properly cited. The work cannot be changed in any way or used commercially without permission from the journal.
Leigh syndrome is a group of clinically heterogeneous conditions with widely varying clinical manifestations. More than 75 causative genes have been identified in Leigh syndrome. The diagnosis of Leigh syndrome is challenging due to the wide range of clinical manifestations. We present the clinicopathologic and genetic characteristics of a family with Leigh syndrome coexisting with hereditary neuropathy with liability to pressure palsy (HNPP).

**Clinical Characteristics**

Table 1 summarizes the family's clinical characteristics. Patients 1 (III-1, eFigure 1, links.lww.com/NXG/A545) and 3 (III-3) presented with spastic paraplegia, intellectual disability, cerebellar ataxia, and respiratory failure. Brain MRI for patient 1 revealed diffuse high-intensity lesions with coarsening and vacuolization in the white matter (eFigure 2, A–F) as well as multiple lesions in the brainstem and cerebellum (eFigure 2, G–I). Patients 1 and 3 had marked peripheral neuropathy. Patient 4 (II-5) developed compression neuropathy after surgery (eTable 1).

**Histology and Respiratory Chain Enzyme Activity of Biopsied Muscle**

A biopsy specimen from patient 1 revealed variation in muscle fiber diameter (eFigure 3A, links.lww.com/NXG/A545). A moderate increase in lipid was found in muscle fibers (eFigure 3B). Modified Gomori trichrome staining revealed no ragged-red fibers (eFigure 3C). COX activity was found to be reduced in a widespread and severe manner (eFigure 3, D and E). The uniform staining pattern of type 2 fibers was revealed by ATPase histochemistry (eFigure 3F).

Measurement of respiratory chain complex enzyme activity in biopsied muscle revealed that the levels of all enzymes were lower than those in normal tissues (eTable 2, links.lww.com/NXG/A545). The enzyme activity in fibroblast revealed a selective decrease in complex IV (eTable 2). The results of the enzymes were consistent with complex IV deficiency.

**Autopsy Findings**

An autopsy was performed on patient 3 (III-3) at age 9 years. The brain showed diffuse and severe white matter degeneration (Figure 1, A and B). The loss of myelin and axons appeared to be similar in extent in the degenerated white matter (Figure 1, C and D), and vacuolar changes of the neuropil accompanied by gliosis were observed (Figure 1E). Atrophy and myelin pallor was also evident in the cerebellar white matter, middle cerebellar peduncles, and pontine base (Figure 1F). Spotty demyelinating lesions were found in the left medial lemniscus (Figure 1F) and the reticular formation of the medulla oblongata (Figure 1, G and H). Axons were relatively preserved in both lesions, as well as the ventral part of the medulla oblongata, despite the significant loss of myelin (Figure 1, I and J). Olivary hypertrophy with severe gliosis (Figure 1K), Purkinje cell loss in the cerebellar cortex, and gliosis in the dentate nucleus (Figure 1L) were evident.

**Genetic Analysis**

Whole-exome sequence analysis showed COX10 variants of p.Arg159Gln and novel p.Pro295Leu in patients 1 and 3 (Figure 2A). An in silico analysis suggested that the p.Pro295Leu variant was pathogenic, whereas the p.Arg159Gln was

---

**Table 1** Summary of Clinical and Genetic Findings of the Affected Member of the Family

|                  | Patient 1 (III-1) | Patient 2 (III-2) | Patient 3 (III-3) | Patient 4 (II-5) |
|------------------|-------------------|-------------------|-------------------|------------------|
| Clinical diagnosis| Leigh syndrome    | Cerebral palsy    | Leigh syndrome    | Pressure palsy   |
| Age at onset     | 4 y               | 15 mo             | 2 y               | 79 y            |
| Age (current status) | 54 y (alive) | 4 y (deceased)   | 9 y (deceased)    | 80 y (alive)    |
| Initial symptoms | Gait disturbance and mental and motor retardation | Gait disturbance and mental and motor retardation | Gait disturbance and mental and motor retardation | Numbness and weakness of upper extremities |
| Other symptoms   | Spastic tetraplegia, nystagmus, vertigo, cerebellar ataxia, dysphagia, and respiratory failure | N/A | Spastic tetraplegia, dysphagia, and respiratory failure | None |
| Peripheral neuropathy | Present | N/A | Present | Present |
| Serum lactic acid (mg/dL) | Normal (17.4) | N/A | Normal (12.4) | N/A |
| CSF lactic acid (mg/dL) | Elevated (43.8) | N/A | N/A | N/A |
| **COX10 mutation(s)** | p.P295L/partial deletion | N/A | p.P295L/partial deletion | Partial deletion |
| **PMP22 mutation** | Deletion | N/A | Deletion | Deletion |

Abbreviation: N/A = data not available.
predicted to be benign (eTable 3, links.lww.com/NXG/A545). CNV analysis of COX10 revealed a decreased gene dosage, indicating the presence of a deletion in patients 1, 3, and 4 (Figure 2B). Furthermore, CNV analysis revealed a decreased gene dosage in PMP22 (Figure 2C). A DNA microarray analysis revealed an approximately 1.4-Mb deletion spanning from intron 6 of COX10 to PMP22 (Figure 2D).

Protein Analysis

Proteins were extracted from the frozen tissue (cerebral cortex and white matter) in patient 3. In both mitochondrial and total lysate fractions, Western blot analysis revealed decreased expression levels of the COX1/MT-CO1, COX2/MT-CO2, and COX4 complex IV subunits (eFigure 4, A and B, links.lww.com/NXG/A545).

Discussion

In this study, we identified a family with Leigh syndrome coexisting with HNPP caused by biallelic COX10 mutations and heterozygous PMP22 deletion. An enzyme activity analysis revealed that mitochondrial respiratory chain complex IV deficiency is the cause of Leigh syndrome in this family, which is most likely caused by biallelic COX10 mutations composed of novel missense mutation and partial deletion (eFigure 5, links.lww.com/NXG/A545).
Peripheral neuropathy was observed in patients 1 and 3, and compression neuropathy was noted in their father (patient 4). We identified a deletion of PMP22, which is cosegregated with neuropathy in the family (eFigure 5, links.lww.com/NXG/A545). We report the patients of Leigh syndrome with neuropathy associated with PMP22 deletion and COX10 mutation. COX10 is located in the vicinity of PMP22 and has a CMT1A-repeat sequence that is highly similar to that of PMP22 (eFigure 6). The presence of such a sequence suggests a mechanism for homologous recombination. Genetic rearrangements resulting in HNPP deletion disrupted 1 copy of COX10 on the recombinant allele. If the pathologic mutation of COX10 coexists in the other allele, a patient with HNPP could develop Leigh syndrome, as was observed in our family.

Acknowledgment
The authors thank the patients and their family members whose participation made this work possible. The authors also thank Drs. Shuichi Igarashi, Motoyoshi Yamazaki, and Yoji Onishi for kindly providing valuable clinical information.

Study Funding
This study was partly supported by AMED JP22dk0207060 and KAKENHI 22H02980 (to T.I.) and AMED JP21ek0109468 and JP19ek0109273 (to M.K.).

Disclosure
The authors report no disclosures relevant to the manuscript. Go to Neurology.org/NG for full disclosures.

Publication History
Received by Neurology: Genetics April 6, 2022. Accepted in final form July 26, 2022.

Appendix Authors

| Name                  | Location                                                                 | Contribution                                                                 |
|-----------------------|---------------------------------------------------------------------------|------------------------------------------------------------------------------|
| Yasuko Kuroha, MD, PhD| Department of Neurology, Nishinigata Chuo Hospital, Niigata; Department of Molecular Genetics, Brain Research Institute, Niigata University, Japan | Drafting/revision of the manuscript for content, including medical writing for content; major role in the acquisition of data; study concept or design; and analysis or interpretation of data |
| Takanobu Ishiguro, MD, PhD | Department of Molecular Genetics, and Department of Neurology, Brain Research Institute, Niigata University, Japan | Drafting/revision of the manuscript for content, including medical writing for content; major role in the acquisition of data; study concept or design; and analysis or interpretation of data |
| Mari Tada, MD, PhD    | Department of Pathology, Brain Research Institute, Niigata University, Japan | Drafting/revision of the manuscript for content, including medical writing for content; major role in the acquisition of data; and analysis or interpretation of data |
| Norikazu Hara, PhD    | Department of Molecular Genetics, Brain Research Institute, Niigata University, Japan | Major role in the acquisition of data and analysis or interpretation of data |
| Kei Murayama, MD, PhD | Department of Metabolism, Chiba Children's Hospital, Japan                | Major role in the acquisition of data and analysis or interpretation of data |
References

1. Leigh DJ. Subacute necrotizing encephalomyelopathy in an infant. J Neurol Neurosurg Psychiatry. 1951;14(3):216-221. doi:10.1136/jnnp.14.3.216.

2. Rahman S, Blok RB, Dahl HH, et al. Leigh syndrome: clinical features and biochemical and DNA abnormalities. Ann Neurol. 1996;39(3):343-351. doi:10.1002/ana.410390311.

3. Baertling F, Rodenburg RJ, Schaper J, et al. A guide to diagnosis and treatment of Leigh syndrome. J Neurol Neurosurg Psychiatry. 2014;85(3):257-265. doi:10.1136/jnnp-2012-304426.

4. Lake NJ, Compton AG, Rahman S, et al. Leigh syndrome: one disorder, more than 75 monogenic causes. Ann Neurol. 2016;79(2):190-203. doi:10.1002/ana.24551.

5. Ogawa E, Shimura M, Fushimi T, et al. Clinical validity of biochemical and molecular analysis in diagnosing Leigh syndrome: a study of 108 Japanese patients. J Inherit Metab Dis. 2017;40(5):685-693. doi:10.1007/s10545-017-0042-6.

6. Reiter LT, Murakami T, Koeuth T, et al. A recombination hotspot responsible for two inherited peripheral neuropathies is located near a mariner transposon-like element. Nat Genet. 1996;12(3):288-297. doi:10.1038/ng0396-288.

7. Reiter LT, Murakami T, Koeuth T, et al. The human COX10 gene is disrupted during homologous recombination between the 24 kb proximal and distal CMT1A-REPs. Hum Mol Genet. 1997;6(9):1595-1603. doi:10.1093/hmg/6.9.1595.