DISSOCIATION OF DAMAGE TO SPATIAL AND LUMINANCE CHANNELS IN EARLY LEBER'S HEREDITARY OPTIC NEUROPATHY MANIFESTED BY THE VISUAL EVOKED POTENTIAL

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SUMMARY

Leber's hereditary optic neuropathy (LHON) is a maternally inherited disorder characterised by the acute or subacute loss of central vision, leading to severe optic atrophy. The occurrence of a demyelinating disease such as multiple sclerosis (MS) has been reported mainly in females with the mitochondrial DNA 11778 mutation. They had a poor visual outcome. To investigate the damage to the optic nerve in an early stage of LHON as compared with optic neuritis (ON), including MS, we examined patients by testing their pattern visual evoked potential (PVEP) and flash VEP (FVEP). The mean amplitude and latency in the PVEP were reduced and delayed, respectively, in patients with LHON or ON. The mean amplitude in the FVEP was reduced in patients with LHON or ON, but the mean latency was not delayed in patients with LHON as compared with normal subjects or patients with ON. Luminance-related fibres may be less affected than are the spatial-vision-related fibres in patients with early LHON.

Leber's hereditary optic neuropathy (LHON) is a maternally inherited form of optic neuropathy, primarily of young males, that is characterised by the acute or subacute loss of central vision, leading to optic atrophy with cecocentral scotoma.¹ The maternal inheritance may be explained by the involvement of mitochondrial DNA (mtDNA). Wallace *et al.*² identified a mutation at nucleotide position (np) 11778 of mtDNA that is found exclusively in families with LHON. A number of other mtDNA mutations, especially primary mutations at positions 11778, 3460

and 14484, have been pathogenetically linked to LHON.³

The occurrence of a demyelinating disease such as multiple sclerosis (MS) or a MS-like disease, has been reported mainly in females who carry the mitochondrial DNA 11778 mutation.⁴⁻⁶ These patients had a poor visual outcome. In such cases, LHON can not be distinguished from MS without molecular genetic testing, especially in the absence of a family history. However, their clinical findings in an acute stage should also differ. Recent magnetic resonance imaging (MRI) studies have demonstrated Gd-DTPA enhancement in patients with acute optic neuritis, indicating the presence of inflammation.^{7,8} However, few, if any, characteristic abnormalities of the optic nerve signal have been reported in patients with early LHON.⁹

The visual evoked potential (VEP) is a useful test for detecting MS. A delayed latency in the VEP is commonly observed in patients with demyelinating disorders.^{10,11} The pattern VEP (PVEP) is reported to be attenuated, delayed or desynchronised in early LHON.^{12,13} However, a LHON patient with the 14484 mutation was reported in whom the PVEP was significantly delayed but the critical frequency of photic driving (CFPD), that is the highest frequency of photic driving in response to repetitive flash stimulation, was normal in an early stage.¹⁴ Thus, the spatial and luminance channels of patients with LHON may be dissociated. There is no report of both PVEP and flash VEP (FVEP) testing in patients with early LHON.

To investigate the dissociation of damage between spatial and luminance channels in an early stage of LHON, we examined patients with LHON by use of the PVEP and FVEP, and compared our findings with those in patients with optic neuritis (ON), including MS.

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SUBJECTS AND METHODS

Patients

We investigated 28 Japanese patients: 12 (18 eyes) with LHON and 16 (18 eyes) with ON, including 8 patients with MS. Of the group with LHON, 9 had the 11778 mutation, 2 had the 14484 mutation and 1 had the 3460 mutation. Eleven of the 12 patients with LHON were male and one was female. Their mean age at onset of the disease was 23.5 ± 8.1 years (range 11–37 years). PVEP and FVEP were recorded in the 28 patients within 1 month of disease onset. Of the group with ON there were 10 males and 6 females, with a mean age at onset of 28.6 ± 6.4 years (range 13–38 years). They showed central scotomas and decreased visual acuities from 0.06 to 0.3 in the affected eye, and were treated with steroids. Their vision then recovered to 1.0 or 1.2. They do not have mtDNA LHON mutations.

Thirty normal volunteers (30 eyes) served as controls: 20 males and 10 females, mean age 30.1 ± 6.0 years (range 20–39 years).

Visual Evoked Potentials

VEPs were derived monopolarly at 5% from theinion when theinion–nasion distance was 100%, as

described previously.^{15,16} Indifferent and ground electrodes (8 mm Ag–AgCl disc electrodes) were placed on separate earlobes. Evoked potentials were averaged 64 or 128 times. For recording the PVEP, chequerboard pattern reversal stimuli were displayed on a 20-inch television monitor. The size of the stimulus field subtended a visual angle of $14^\circ \times 11^\circ$ observed from a distance of 160 cm. Checks of four different sizes were used: 5', 10', 20' and 40'. The mean luminance was 50 cd/m² with a contrast of 80%. The temporal frequency of the stimulus pattern was 2 reversals per second. Latency time was

Fig. 1. Check size–amplitude curve (A) and check size–latency curve (B) in pattern visual evoked potentials (VEPs). Open circles and bars represent the mean $\pm$ SD in normal subjects (30 eyes). Filled circles and bars represent the mean $\pm$ SD in patients with Leber's hereditary optic neuropathy (LHON), and open squares represent the mean $\pm$ SD in patients with optic neuritis (ON). N shows the number of eyes. p values (*) represent one-factor ANOVA, Scheffé's F-test. (A) Mean amplitude was significantly reduced in both LHON and ON patients as compared with normal subjects in check sizes 10' (*1: $p = 0.0327$, *2: $p = 0.0202$, respectively), and 20' (*3: $p = 0.0056$, *4: $p = 0.0021$, respectively). There was no difference in amplitude between LHON and ON patients ($p > 0.05$). (B) In patients with LHON, mean latency was delayed as compared with normal subjects in two check sizes: 10' (**1: $p = 0.0476$) and 20' (**2: $p = 0.0246$). In patients with ON, mean latency was delayed as compared with normal subjects in check sizes 10', 20' and 40' (*1: $p < 0.0001$), and in patients with LHON in check sizes 20' (*1: $p = 0.0127$) and 40' (**2: $p = 0.0035$).

determined for the negative peak that appeared around 80 ms (N80). The early phase reportedly represents the specific part of the visual cortex response.¹⁷ The amplitude was measured from N80 to P100, which appears around 100 ms. The check size–latency curve and the check size–amplitude curve served as critical variables.

FVEPs were recorded at four different intensities of stimulus using a red filter (Fuji SC-64, Tokyo, Japan). The visual stimulus was a xenon light of 0.3 joule (J) and of 2.0 J. The four stimulus intensities were 0.3×10^{-2} J (-0.2 log unit), 0.3×10^{-1} J (-1.0 log unit), 0.3 J (0 log unit) and 2.0 J ($+0.8$ log unit). Recording sessions were started with a 2.0 log unit neutral density filter placed in front of the stimulus light source (0.3 J). The intensity–latency curve and the intensity–amplitude curve served as critical variables.^{16,18}

Statistical Analysis

Data are presented as the mean $\pm$ SD. Differences in amplitude or latency time in VEPs among the three groups (normal subjects, patients with LHON, patients with ON) were analysed by one-factor ANOVA, Scheffé *F*-test. A value of $p < 0.05$ was accepted as statistically significant.

RESULTS

The check size–amplitude curve and the check size–latency curve are shown in Fig. 1. PVEPs were recordable in 5 of the 12 patients with LHON and in 8 of the 16 patients with ON. In normal subjects, the check size–amplitude curve represented a maximum response produced by check sizes of 10' to 20',¹⁹ which suggests a band-pass function ($\cap$ -shaped curve). In patients with LHON and ON, however, the curve was linear. In LHON, as well as in ON, the mean amplitude was significantly reduced as compared with normal subjects, except for a larger check size of 40' (Fig. 1A). There was no difference in amplitude between patients with LHON and ON ($p > 0.05$). The latency time in LHON as well as in ON was significantly delayed as compared with normal subjects. The delay in the latter exceeded that in the former (Fig. 1B).

The intensity–amplitude curve and the intensity–latency curve are shown in Fig. 2. FVEPs were recordable in all 12 patients with LHON and in all 16 patients with ON. Mean amplitude was significantly reduced in both patients with LHON and ON as compared with normal subjects (Fig. 2A). There was no difference in amplitude between patients with LHON and ON ($p > 0.05$). Mean latency time was not delayed in LHON as compared with normal subjects ($p > 0.05$), but in ON the mean latency time was markedly delayed as compared with normal subjects as well as with LHON patients (Fig. 2B).

Fig. 2. Intensity–amplitude curve (A) and intensity–latency curve (B) in flash VEPs. Open circles and bars represent the mean $\pm$ SD in normal subjects (30 eyes). Filled circles and bars represent the mean $\pm$ SD in patients with LHON, and open squares represent the mean $\pm$ SD in patients with ON. N shows the number of eyes. p values (*) represent one-factor ANOVA, Scheffé's *F*-test. (A) Mean amplitude was significantly reduced in both LHON and ON patients as compared with normal subjects (*1: $p = 0.0027$, *2: $p = 0.0023$, *3: $p = 0.0003$, *4: $p = 0.0001$, *5: $p < 0.0001$). There was no difference in amplitude between LHON and ON patients ($p > 0.05$). (B) In patients with LHON, mean latency time was not delayed as compared with normal subjects ($p > 0.05$). In patients with ON, mean latency time was markedly delayed as compared with normal subjects (*1: $p = 0.0006$, *2: $p < 0.0001$), as well as with LHON patients (**1: $p = 0.242$, **2: $p < 0.0001$).

Fig. 3. Waveforms of pattern VEPs (PVEPs) from the right eye in a 23-year-old patient with LHON. In the presymptomatic stage (A), visual acuity was 25/20. Three days after the onset (B), visual acuity was 20/20. Two weeks after the onset (C), visual acuity was 20/30. Numerals represent latency time.

Fig. 3 shows PVEP waveforms in a patient with the 11778 mutation who developed LHON at the age of 23 years. VEPs were recorded in the right eye at the presymptomatic and symptomatic stages. PVEPs were not recorded in the left eye that had developed LHON 4 months earlier. In the presymptomatic stage (Fig. 3A), N80 was the upper limit of normal, P100 was slightly delayed, and the amplitudes were normal in three check sizes, except in check size 5'. Three days after the onset of LHON (Fig. 3B), the amplitude was markedly reduced, but N80 and P100 were unchanged. Two weeks after the onset (C), the amplitude was markedly reduced and N80 and P100 were further delayed. PVEPs were not recordable 1

month after the onset. During onset of LHON, there was a marked reduction in amplitude, followed by a delay in latency. Fig. 4 shows the waveforms of FVEPs in the same patient as shown in Fig. 3. During an acute stage, FVEPs were slightly attenuated but not delayed, although PVEPs deteriorated rapidly.

DISCUSSION

A recent histopathological report on the atrophic stage of LHON demonstrated a striking loss of ganglion cells and nerve fibre layers in the retina.²⁰ The optic nerve and tract appeared shrunken and atrophic with little evidence of inflammation. LHON supposedly affects primarily the retinal ganglion cells and their axons.

Fig. 4. Waveforms of flash VEPs (FVEPs) from the right eye in the same patient as shown in Fig. 3: presymptomatic stage (A), 3 days after the onset (B) and 2 weeks after the onset (C). Numerals represent latency time. Stimulus delay was 20 ms after a trigger signal (vertical arrows).

Patients with MS reportedly show no pathological changes in the retina, but do show plaques and periphlebitis in the optic nerve, chiasm and tract.²¹ While the axonal myelin sheaths undergo destruction in MS, the axons are often well preserved.^{22,23} LHON differs histopathologically from MS.

Few studies of VEP performed at an early stage of LHON have been reported. Dorfman *et al.*¹² studied PVEP in two brothers with early LHON. One month after the onset, the latency of VEP was normal. Then, the response developed prolonged latency and reduced amplitude with the waveform developing a bifid positivity in a W configuration. The pattern size used was a check of 1°50' of arc. Carroll and Mastaglia¹³ also found that PVEP in patients with LHON was less delayed in P100 latency, but showed a greater reduction in amplitude, and was disorganised. They speculated that these findings implied a more severe disturbance of conduction in the optic nerve, consistent with a combination of demyelination and the loss of nerve fibres.¹³ The pattern size used was a check of 12' of arc. PVEPs were normal in most of the asymptomatic individuals in a LHON family, but were absent in the symptomatic individuals.²⁴

We investigated optic nerve conduction using the PVEP and FVEP in 12 patients with LHON and in 16 patients with ON within 1 month after the onset. We recorded the PVEP and FVEP from a patient with LHON in both presymptomatic and symptomatic stages, and found the same result as had Carroll and Mastaglia¹³: a marked reduction in amplitude followed by latency delay in PVEP. Subsequently, the PVEP became non-recordable. However, FVEPs were less affected during such periods. In this study, the most striking manifestations of the effects on the VEP in LHON were those apparent on the FVEP. PVEPs were reduced and delayed, or not recordable in patients with LHON or ON. However, in the patients with LHON, the FVEPs were reduced but not delayed. This finding may suggest that most of the fibres in the luminance channels were less affected by LHON than were fibres in the spatial frequency channels. Latency time (N80) was less affected in LHON than in ON. In patients with demyelinating ON, a delay of latency was marked for both FVEP¹⁰ and PVEP.¹¹ The characteristics of FVEPs in the patients with LHON resemble those of patients with toxic neuropathy¹⁰ and with compression of the optic nerve.^{10,18} The early stage of LHON differs electrophysiologically from that of ON.

The patient with early LHON exhibits central or centrocecal scotomas. The optic disc is swollen and hyperaemic. Retinal arterioles on and around the disc are dilated, tortuous and telangiectatic. Fluorescein angiography shows no leakage of dye from the

telangiectatic vessels. The nerve fibre layer is also swollen. Several weeks after the onset, there is a reduction in size of the telangiectatic arterioles on the temporal side of the disc accompanied by nerve fibre layer 'dropout' (axonal loss) in a narrow channel in the papillomacular bundle.²⁵ Consequently, optic atrophy with nerve fibre layer defect is most pronounced in the papillomacular bundle. Nikoskelainen *et al.*²⁵ suggested that LHON causes intraocular, not retrobulbar neuropathy. Thus, the finding in patients with early LHON that the VEP was less delayed in latency but more markedly reduced in amplitude may be associated with selective axonal loss in the papillomacular bundle in the retina.

The critical frequency of photic driving (CFPD) was reportedly normal in a patient with LHON who showed a significant delay in the VEP response to pattern stimuli for a check of 56' of arc.¹⁴ Patients with demyelinating disease typically exhibit diminished CFPD values.²⁶ Another report found that latency of the flash-evoked blink reflex was normal in 10 of 11 patients with LHON, while 12 of 15 patients with other forms of ON showed delayed or extinguished response.²⁷ The direct pupillary response to light was reported to be preserved in LHON, as compared with ON.²⁸ The authors²⁸ speculated that in LHON, W cells functioning as a luminance detector and providing input to the pupillary light response were relatively intact, in contrast to an impairment of the X and Y cells. In an acute stage of LHON, luminance-related fibres or pupillary-reflex-related fibres may be less affected than are the spatial-frequency-related fibres, whereas in patients with ON all these types of fibres are damaged by inflammation or demyelination of the optic nerve.²⁶⁻²⁹

Key words: Leber's hereditary optic neuropathy, Multiple sclerosis, Visual evoked potential, Luminance, Latency.

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