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An exploratory study to evaluate efficacy and safety of frequent Transcutaneous Electrical Stimulation for Leber Hereditary Optic Neuropathy

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Electrical stimulation (ES) may be effective for intractable retinal or optic nerve diseases. We studied frequent transcutaneous ES in a single-center, single-arm prospective study in patients with Leber hereditary optic neuropathy (LHON) who carry the mitochondrial (mt) 11778 G > A mutation. A 30-min ES was applied to either eye every other day for 12 weeks. The primary outcome was the difference in the logarithm of the minimum angle of resolution (LogMAR) at baseline and 1 week after completion of ES treatment. The secondary outcomes included changes in visual field; LogMAR; critical flicker frequency; and inner retinal thickness. Safety endpoints included the corneal endothelial cell density and complications during ES. Fourteen patients participated in the study; four dropped out. The median (interquartile range) LogMAR values before stimulation and 1, 4, and 8 weeks after ES were 1.60 (1.45–1.80), 1.70 (1.35–1.80), 1.60 (1.43–1.73), and 1.50 (1.43–1.73), respectively, indicating no significant improvement (primary outcome: Wilcoxon's signed rank test, $p = 1.000$, secondary outcome: Friedman test, $p = 0.229$). There were no improvements in any secondary efficacy endpoints and no complications. In conclusion, frequent transcutaneous ES did not improve visual acuity in patients with LHON carrying the mt11778 G > A mutation.

Keywords Electrical stimulation, Leber Hereditary Optic Neuropathy, Mitochondrial disease, Prospective study, Visual field

Leber hereditary optic neuropathy (LHON), a maternally inherited intractable optic neuropathy, was the first human disease proven to be associated with mitochondrial (mt) DNA mutations[1]. Three major missense mutations, mt3460G > A, mt11778G > A, and mt14484T > C, are detected in more than 90% of patients. The estimated prevalence is 1 in around 50,000[2–4].

LHON is caused by sudden apoptosis in retinal ganglion cells (RGCs). The clinical manifestations include subacute progression of visual loss and visual field defects in one eye, and subsequent involvement of the contralateral eye after several weeks[5], culminating in final visual acuity of 0.1 or below—often as low as 0.01—in both eyes[6]. There is usually a central scotoma in the visual field, but the peripheral vision remains intact. The patient is able to adapt and compensate for the purposes of mobility and general activities of daily living; however, activities such as reading and performing precisions tasks are impaired[7].

In some cases, visual function improves spontaneously[6]. However, the degree of improvement rarely reaches the point at which daily life is not inconvenienced. Therefore, in most cases, therapeutic intervention to improve visual function is applied. Idebenone is one of the representative therapies for LHON[8]. Idebenone acts as a carrier of electrons in the mitochondria under certain concentrations and has a compensatory effect on the electron transfer system. A large Randomized Controlled Trial (RCT) reported that oral administration of idebenone at 900 mg/day for 24 weeks significantly improved visual function[9]. Gene therapy is another important treatment option. It introduces the normal ND4 gene into the nucleus by allotropic expression, resulting in the expression of intact electron transfer complex function. Several RCTs have reported a small but significant improvement in visual function[10–13].

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However, these are not sufficiently effective to significantly improve quality of life.

Electrical stimulation (ES), a potential treatment strategy for intractable retinal or optic nerve diseases, attempts to restore visual function by activating the function of the remaining photoreceptor cells or RGCs. The efficacy of ES has been reported in several clinical studies. In our previous study in which 10 patients with LHON received six ES treatments every 2 weeks[14], the average logarithm of the minimum angle of resolution (LogMAR) was significantly improved 1 week after the ES treatment, meeting the primary outcome. The study was conducted safely with no complications. However, the size of the effect was small, and stimulation was applied infrequently. Continuous ES is reported to be more cytoprotective in both the basic and clinical research[15,16].

Hence, we designed a prospective and exploratory clinical trial to determine the effect of ES on patients with LHON carrying the mt11778 G > A mutation. We evaluated if a repeated and longer-term stimulation protocol resulted in a greater improvement in visual function without affecting safety.

Results

Historical data of the patients

Patient demographics are presented in Table 1. The average age of the patients was 50.2 ± 10.4 years old. Fourteen patients with the mt11778 G > A mutation participated in this study, of which four were registered in our previous study. The median (interquartile range (IQR)) duration of the disease was 37.5 (26.3–123) months. Ten patients completed the whole study, and four patients discontinued: one patient forgot the ES schedule and interrupted treatment and three patients had interrupted hospital visits during the study.

Efficacy of the study

Figure 1 presents the LogMAR after 0, 13, 16, and 20 weeks on this study. The median (IQR) LogMAR values were 1.60 (1.45–1.80), 1.70 (1.35–1.80), 1.60 (1.43–1.73), and 1.50 (1.43–1.73); these results did not meet the criteria for the primary or secondary outcome of this study (Primary outcome; Wilcoxon's signed rank test, $p = 1.000$, secondary outcome; Friedman test, $p = 0.229$). In the subgroup analysis, we compared BCVA between three patients who participated in our previous study (patient 1, 2, 14) and other patients who completed the study. There were no differences in visual acuity between the two groups before and after the study (Table S1, mixed-effect model and global analysis, $p = 0.67$).

Table 2; Fig. 2, Figure S1, and Table S2 present the analysis of the visual field data. Table 2 shows the number of points at which sensitivity improved or decreased by > 5 dB after the ES treatment at each of the measurement points in the visual field test. In six cases, there were more improved points than decreased points in both ES-treated and untreated eyes. Figure 2 presents the sum of the sensitivity of each area of the visual field. There were no significant changes in sensitivity for any of the evaluation areas. The p values of the Friedman rank sum test were 0.194, 0.519, 0.093, 0.499, 0.039, 0.943, 0.645 in the small central (4 points), large central (16 points), total, nasal-upper, nasal-lower, temporal-upper, and temporal-lower areas, respectively. In the nasal-lower area, the p value was below 0.05; however, the post-hoc analysis revealed no significant difference at any time for any evaluation points. We also compared the variation in visual field sensitivity during this study in the treated and untreated eyes. Figure S1 and Table S2 present the results of the mixed-effect model comparing changes in the sum of the sensitivities of each visual field area in treated and untreated eyes over the course of the study. No significant difference between the trends in sensitivity in the two groups was detected.

Safety of the study

Table 3 presents the analysis of secondary outcome data, including CFF (Table 3a), RNFL and GCC thickness (Table 3b), and CED (Table 3c). Neither a significant difference nor a 20% decrease in the thickness of the OCT result was observed.

No.	Age	Duration	Sex	Eye	Previous Study	Status	Reason
1	32	168	M	R	Y	C	
2	49	372	M	R	Y	C	
3	54	348	M	R	Y	D	Forgotten ES schedule
4	52	18	M	L	N	C	
5	42	30	M	R	N	D	Interruption of hospital visits
6	52	33	M	L	N	C	
7	40	34	M	R	N	D	Interruption of hospital visits
8	63	16	F	L	N	C	
9	44	24	M	L	N	C	
10	65	44	M	L	N	C	
11	51	41	M	L	N	D	Interruption of hospital visits
12	38	48	M	R	N	C	
13	71	27	M	L	N	C	
14	50	468	M	R	Y	C	

Table 1. Patient demographics. M, male; F, female; R, right; L, left; Y, yes (participated in our previous study); N, no; C, completed; D, dropped out; ES, electrical stimulation.

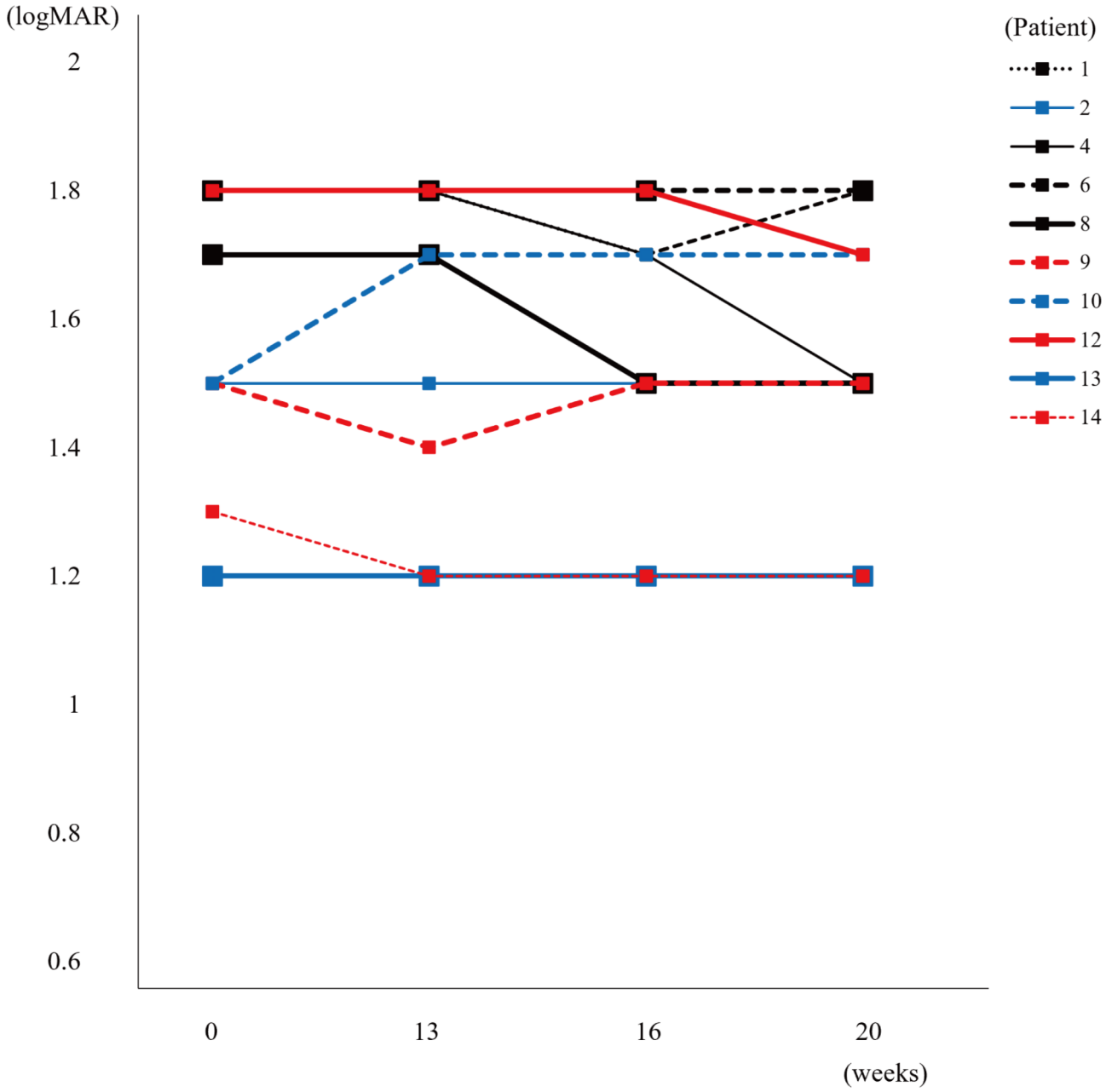

Fig. 1. LogMAR at 0, 13, 16, and 20 weeks of the patients.

Phosphene of the patients

Figure 3 presents the phosphene thresholds every 10 days during the study. Seven patients perceived phosphene and recorded it accurately. The median phosphene thresholds for these patients were 350 (217–706), 303 (203–440), 350 (253–603), 454 (200–549), 274 (132–400), 300 (106–372), 249 (207–367), 300 (120–450), and 249 (141–451) μA, as evaluated on days 0, 10, 20, 30, 40, 50, 60, 70, and 80, respectively. There was no significant decrease in phosphene thresholds (Friedman rank sum test, 0.0928).

Discussion

In theory, ES can inhibit neuronal apoptosis and microglial activation and promote neuronal regeneration. In rodents, ES increased the production of insulin-like growth factor-1 (IGF-1), brain-derived neurotrophic factor, ciliary neurotrophic factor, and Bcl-2 from Müller cells, which may be the cause of the cytoprotective effects of ES. Other animal studies have also found that, after transection of the optic nerve, ES protects the retina and optic nerve architecture and function by inhibiting RGC cell death and expressing ICG-1 depending on the intensity of the current[17–20]. ES is also reported to increase retinal blood flow, which is expected to reflect the stimulation of RGCs[21–23]. In addition, continuous ES is thought to have an enhanced neuroprotective effect. Morimoto et al.[15] reported a significantly increased survival rate of RGCs after four consecutive ES treatments

	ES			control		
Pt	0_13wk	0_16wk	0_20wk	0_13wk	0_16wk	0_20wk
1	I	I	D	I	D	D
2	I	I	I	I	I	I
4	NA	NA	NA	NA	NA	NA
6	I	I	I	I	I	I
8	I	I	I	I	I	I
9	I	D	I	I	D	I
10	D	D	D	D	I	I
12	I	I	I	I	I	D
13	NA	NA	NA	NA	NA	NA
14	I	I	I	I	I	I

Table 2. Cross-table indicating whether each patient showed $\geq$ 5 dB improvement or deterioration in visual field sensitivity after electrical stimulation. Cases with ≥5 dB improvements than ≥5 dB deteriorations are described as I (improved), whereas cases with more ≥5 dB deteriorations than ≥5 dB improvements are described as D (deteriorated). I: Improved, D: deteriorated, NA: not applicable.

every 3–4 days compared with a single treatment. Tagami et al.[16] showed that continuous transcorneal ES for 12 days significantly increased the survival rate of RGCs compared with a single stimulation.

Based on the evidence supporting ES obtained from basic research, numerous clinical trials have studied intractable degenerative diseases of the retina and optic nerve. Transcorneal ES was reported to improve visual acuity and visual field in patients with old traumatic optic neuropathy or nonarteritic anterior ischemic optic neuropathy[24]. In a randomized, controlled, multicenter study of patients with retinitis pigmentosa, transcorneal six consecutive weekly ES treatments improved the electroretinogram (ERG) and visual field[25]. Continuous ES was also efficacious in other types of retinal or neuroophthalmological diseases, such as open-angle glaucoma[26], age-related macular degeneration[27], and central retinal artery occlusion[28].

These basic and clinical reports suggest that ES is effective in intractable diseases of the retina and optic nerve by activating residual cellular function[29]. Moreover, ES is a potential treatment for LHON. However, the present study did not identify any promising results, although our previous study reported an improvement in visual function. Although the methods of analysis in the previous study and the current study differed in some respects, there was no significant difference in the age of the patients or in the course of visual function between the two studies. In the previous study, the median (IQR) logMAR at baseline and 1 / 4 / 8 weeks after the last ES in the previous study were 1.80 (1.70–1.80), 1.75 (1.52–1.80), 1.75 (1.50–1.80), and 1.75 (1.52–1.80), respectively[14], that were found no significant difference with the current study. We speculate that the reason for the significant improvement in visual function with ES in the previous study is that there was one patient who responded well to ES, which may have influenced the results of the study.

In this study, the analysis of visual acuity did not meet either the primary or secondary outcomes. In some patients, the sensitivity of the visual field met the secondary outcome. However, as the significance of an improvement in sensitivity of 5 dB or more, which was defined as a secondary outcome, depends on the sensitivity of each measurement point at the beginning of the study, an analysis that only compares the number of points with improved sensitivity may be insufficient. Therefore, we converted the sensitivity measured in dB to asb and reanalyzed the variation in sensitivity. There was no significant improvement in visual field sensitivity, and the ratio of variation in the sensitivity analyzed by the mixed-effects model was not significantly different from that of the untreated eye.

The OCT measurements met the criteria for secondary outcome because there was not a decrease of more than 20% in both RNFL and GCC thickness. However, the inner retinal layers were extremely thin before ES treatment, producing a phenomenon similar to the floor effect[30]. Therefore, it is difficult to confirm if ES maintained the inner retinal layer thickness.

Phosphene is a pseudophotosensory perception produced by the activation of visual cortical functions by electrical stimulation of the retina and may be one indicator of improved visual function[22]. In the present study, we analyzed the variation in recorded power every 10 days in seven patients who were aware of phosphenes during the ES treatment period and were able to record the smallest power that produced a phosphene. The phosphene threshold varied widely, depending on the patient, even though there appeared to be little difference in visual function. Although ES tended to decrease the power at which phosphenes were perceived, no significant differences were detected. This result was consistent with a lack of significant fluctuations in other measures of visual function.

In addition, as a subgroup analysis, we examined if the changes in visual acuity in patients who participated in our previous study different from other patients[14], but found no variation. As there was an interval of approximately 2 years between the previous study and the present study, we believe this indicates that the results of the present study were not affected by the previous study.

Figure 2appears to show some improvement in visual field sensitivity during the study, although the difference was not significant. We used visual sensitivity in two different methods to analyze visual field. As established as a secondary endpoint, we used 5 dB value, the sensitivity at which the visual grayscale color tone

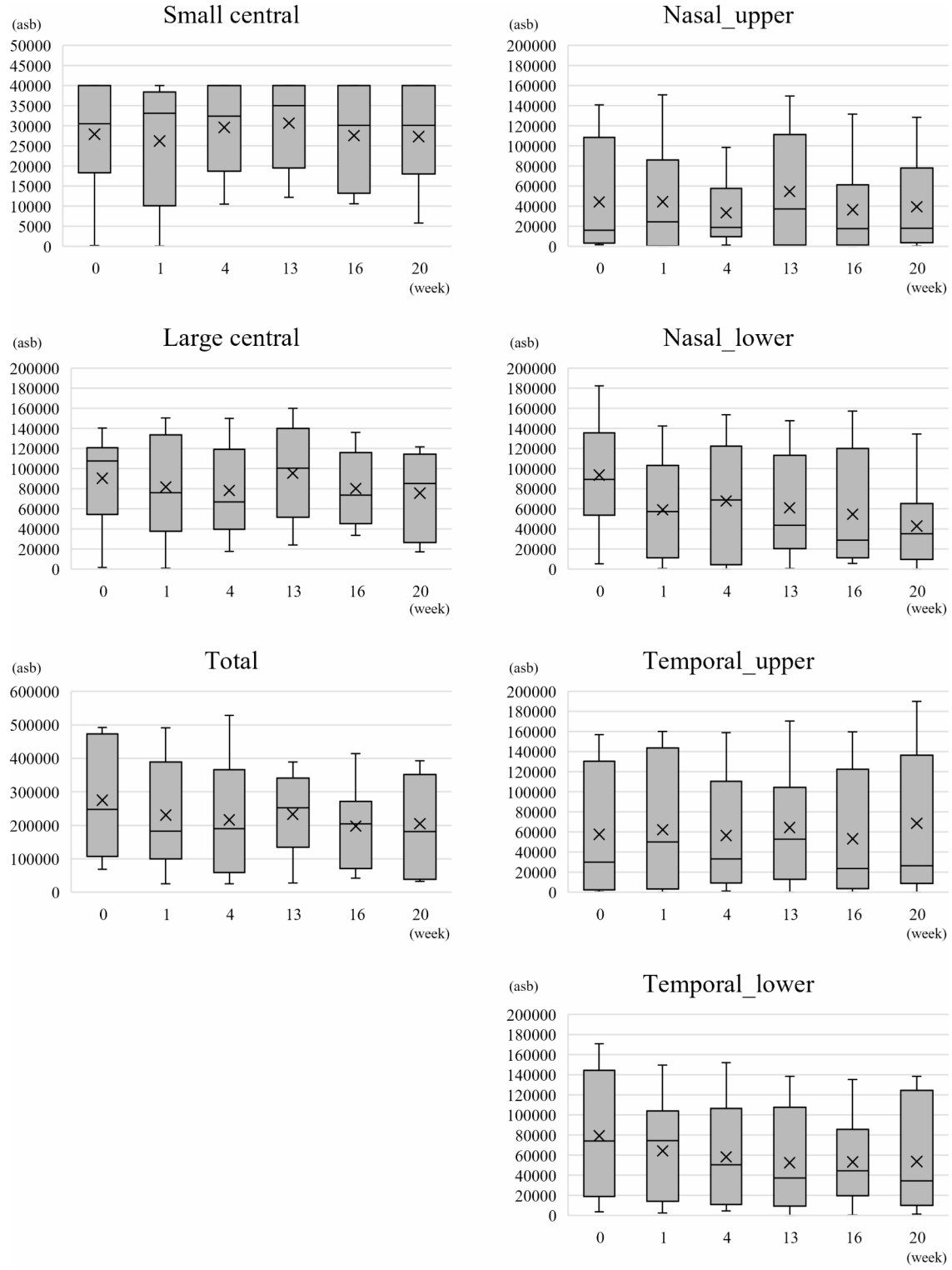

Fig. 2. Sum of the sensitivity of each visual field at 0, 1, 4, 13, 16, and 20 weeks. asb: apostils.

changes, as the criterion for visual field improvement. In addition, we converted dB to asb to perform statistical analysis. Our group previously analyzed the correlation between visual acuity and visual field and found that inferior nasal residual visual field was related to visual acuity[31]. Therefore, we analyzed visual field by region, including the whole visual field, to evaluate which area was most relevant if ES affected visual function. As mentioned in the introduction section, spontaneous recovery has been reported in patients with LHON, with the degree of recovery varying from substantial improvement in visual acuity to limited improvement in the visual field[32]. Following gene therapy, it has been reported that treatment of one eye also improves the visual

a					
wk	**0**	**13**	**16**	**20**	**I**
Disappear (Hz)	20.7 (17.7–37.7)	22.7 (19.3–43.7)	21 (19.7–32.0)	23.5 (19.9–39.7)	0.843
Appear (Hz)	14.3 (11.7–35.8)	21.3 (15.0–35.3)	22.3 (13.3–27.7)	20.5 (11.4–40.0)	0.266
b					
wk	**0**	**1**	**12**	**20**	**I**
RNFL (μm)	58.5 (54.8–64.3)	58.0 (56.0–62.5)	58.5 (55.8–64.3)	58.5 (55.5–62.5)	0.22
GCC (μm)	48.0 (46.5–51.3)	48.0 (46.5–50.5)	49.0 (46.0–51.5)	48.0 (46.3–50.8)	0.775
c					
wk	**0**	**1**	**12**	**20**	**I**
CED	2709 (2360–2909)	2767 (2429–2974)	2892 (2618–3072)	2819 (2388–3133)	0.541

Table 3. CFF(3a), RNFL GCC thickness (3b), CED (3c) during the study. All data are described as the median (interquartile range). Statistical analysis was performed using Friedman's test. CFF, critical flicker frequency; wk, weeks after electrical stimulation; RNFL, retinal nerve fiber layer; GCC, ganglion cell layer; CED, corneal endothelial cell density.

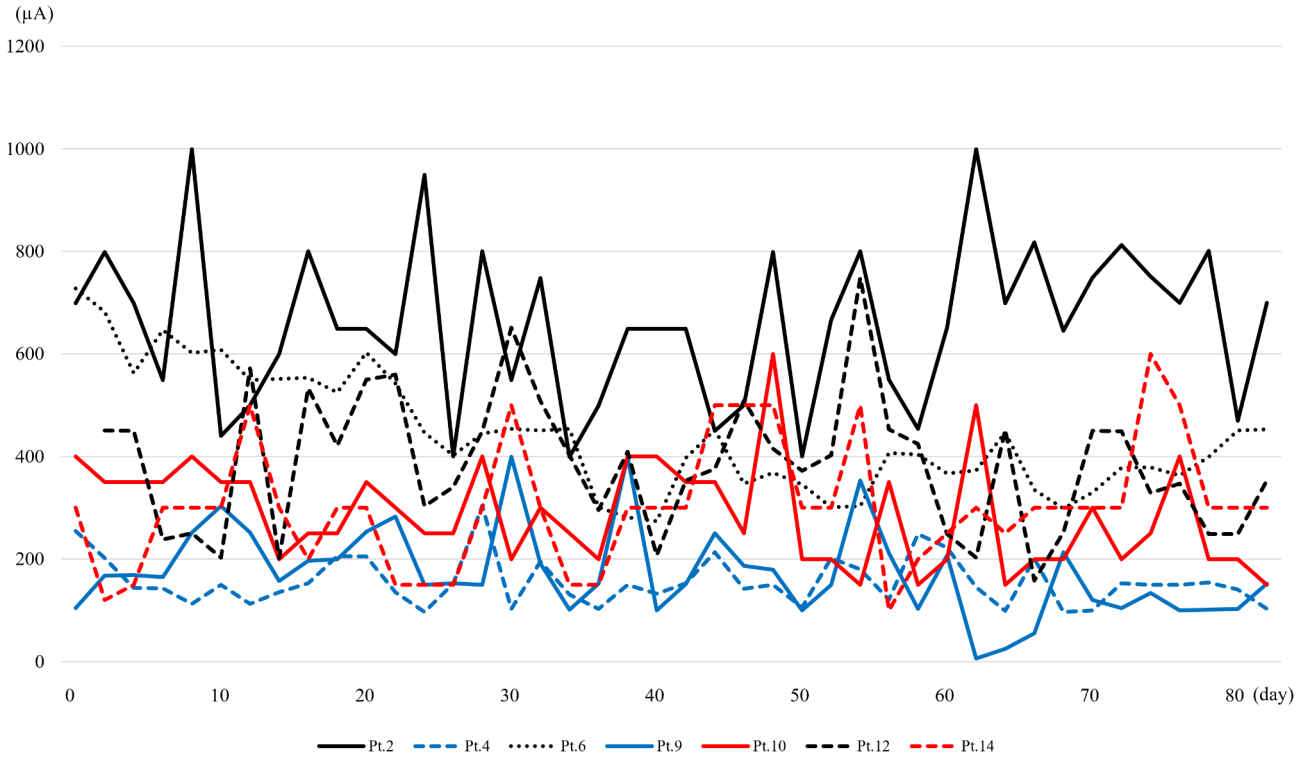

Fig. 3. Variability of phosphene perception in seven patients.

function of the untreated eye[10], but some of these cases include this spontaneous recovery. Therefore, effective treatment for LHON must follow a strategy that significantly improves visual function beyond this degree of spontaneous recovery. In this study, we did not set up control in this study. However, the current that is applied to one eye is almost undetectable in the contralateral eye. In other words, this ES instrument has little effect on the contralateral eye. Therefore, we tried to validate the effect of ES by comparing the results with those of the contralateral eye, as the substitute of control. Consequently, it must be said that this study identified only slight natural changes in visual function, with no effect of ES on visual field improvement observed. Supporting this result, as shown in Table 3, there were an equal number of cases in both the ES-treated and untreated eyes in which the number of points with more than 5 dB improved sensitivity exceeded those with decreased sensitivity.

We used transcutaneous ES equipment in this study and our previous study because transcutaneous ES is safer than the transcorneal approach that is widely applied. As the direct placement of electrodes on the cornea is a burden to the subject and may induce corneal epithelial damage or dry eye[33], many recent studies have examined transcutaneous ES[34,35].

No complications, including CED depletion, were observed in this clinical study. This result is consistent with those of other studies and may be an indicator of equipment safety.

There are several limitations in this study. First, although the sample size was calculated using statistical methods, it was very small. In addition, there is no control group. To evaluate the effect of ES on LHON patients more detail, we should adjust the study design in the future. In addition, the range of disease duration for the patients was very wide in this study. As shown in Table 1, this study includes cases with a long duration of time since onset. Such cases may not respond to therapeutic intervention, because in such cases the condition is fixed, and the optic nerve is completely atrophied. Considering that spontaneous recovery occurs within approximately two years[36], a strict disease duration could affect the results of the study. In other words, earlier intervention may change response to ES treatment.

In conclusion, prolonged transcutaneous ES can be safely administered in patients with LHON but does not significantly improve visual function. There is a need in LHON for new treatment modalities that improve visual function and perhaps visual structure.

Methods

Study design and subjects

This was a single-arm, non randomized, exploratory, prospective study to evaluate the efficacy and safety of frequent transcutaneous ES treatments for patients with LHON carrying the mt11778 G > A mutation. This clinical trial was approved by the Kobe University Clinical Research Ethical Committee, Japan (No. C190030) and registered with the Japan Registry of Clinical Trials (No. jRCTs052200033, https://jrct.niph.go.jp/, the first registration date was 06/07/2020). The research protocol was based on our previous studies and is published elsewhere[14,37].

In brief, 14 patients with LHON participated in this study. Written informed consent was obtained from all patients. For patients who met the inclusion criteria and did not meet any exclusion criteria, either the left or right eye was selected for study. The inclusion criteria were: (1) age ≥ 16 and < 80 years; (2) diagnosis of LHON based on the diagnosis guideline authorized by the Japan Ophthalmological Society and the Japan Neuro-ophthalmological Society[3,38] ; (3) best-corrected decimal visual acuity between 0.01 and 0.1, inclusive; (4) written informed consent from participants or legal representatives; (5) stable condition for > 8 months after the onset of LHON; (6) presence of mt11778 G > A mutation; and (7) could be supported to use the stimulation device by someone with normal visual function. The exclusion criteria were: (1) smoking history within half a year before the initiation of the study; (2) use of electronic devices such as a pacemaker; (3) history of intraocular surgery within a year; (4) ocular complications other than early cataract or intraocular lens; (5) history of idebenone treatment within a year; (6) ongoing treatment with ethambutol, chloramphenicol, linezolid, erythromycin, streptomycin, antiretroviral drugs, amiodarone, infliximab, clioquinol, dapsone, quinine, pheniprazine, suramin sodium, or isoniazid; (7) history of epilepsy; (8) pregnancy; (9) severe dermatitis that could be affected by attaching the electrode pad; (10) participation in other clinical studies; 11) other inappropriate cases as judged by the physicians who are responsible for the study. If both eyes met the criteria, we compared the visual function of the eyes and chose the worse eye. This was due to concern that unexpected problems could occur and adversely affect visual function since the ES device is unapproved.

ES protocol

All participants were presented with an outline of this study, including the schedule of examination and how to use the ES equipment in detail. ES treatments were performed by the participants themselves or with the aid of a supportive person every other day, over a 12-week period, at home, using a non approved portable device (Fig. 4) provided by Mayo Corporation (Aichi, Japan). The ES treatment method has been described elsewhere[36]. In brief, two electrode pads were placed above the eyebrow and on the lower eyelid across the eye and connected to the device. Each ES treatment was conducted using a biphasic square wave with the following properties: amplitude, 1 mA; duration, 10 ms; stimulation frequency, 20 Hz; duration, 30 min. To mitigate issues arising from unexpected device troubles or other emergencies, individuals with no visual abnormalities were present to support each treatment. Since ES treatment was performed at home by the patients and their family, we also asked for the patient's adequate cooperation to ensure that the study would be successful. We explained the use of the equipment to the patients and their families during the initial examination and had them practice, and confirmed they used it properly. We also asked them to keep a record of each day they used the machine. The records were brought to each examination day, and the examining physician checked them each time.

ES schedule

The ES treatment and examination schedule are described in Table 4. Visual function was assessed 0, 1, 4, 8, and 12 weeks after ES treatment (treatment period), as well as 1, 4, and 8 weeks after the final ES treatment (observation period).

Fig. 4. Portable device for transcutaneous electrical stimulation.

Sample size

Sample size was determined on the following basis. In our previous study, 10 patients with LHON underwent ES every 2 weeks for 10 weeks (a total of six treatments). The mean LogMAR before ES was 1.70 in 7 patients with a corrected visual acuity of 0.01 or better and the mean LogMAR visual acuity at 1 week after the final ES was 1.60; thus, the mean change was −0.10 (standard deviation 0.10). Consequently, the difference in the mean LogMAR and SD was defined as −0.10 and 0.1, respectively. In addition, the null hypothesis was designed such that the mean difference in LogMAR before and after ES treatment is −0.10. Based on this assumption, a sample size of 10 subjects was calculated to ensure 80% power of the *t*-test at a two-sided significance level of 5%. To account for dropouts, 14 patients were enrolled.

Examination

In our evaluation of efficacy of ES, we collected sex, age, best-corrected visual acuity (BCVA), critical flicker frequency (CFF), visual field measured by a Humphrey field analyzer (HFA) using the 30–2 program and size V stimulation (Carl Zeiss Meditec, Dublin, California, USA). We selected seven areas in HFA-measured visual fields, as shown in Fig. 5: small central (4 points), large central (16 points), total, nasal-upper, nasal-lower, temporal-upper, and temporal-lower. We included the outermost points in each area because patients with LHON have a central scotoma and may have eccentric fixation. We defined the sensitivity of the area as the sum of the sensitivities and converted the sensitivity to apostils (asb) from decibels (dB).

We also collected the thicknesses of the retinal nerve fiber layer (RNFL) and ganglion cell complex (GCC), as measured by spectral-domain optic coherence tomography (Cirrus HD-OCT, Carl Zeiss, Jena, Germany), and the corneal endothelial cell density (CED) as measured by specular microscopy (Konan Medical, Nishinomiya, Japan).

Outcome measure and analysis

The primary outcome was defined as the difference in LogMAR BCVA between the baseline and 1 week after the final ES treatment. Four parameters were evaluated as secondary analyses: (1) difference between LogMAR at 4 and 8 weeks of ES and LogMAR before stimulation; (2) whether the actual measured sensitivity of each point for the visual field measured at optotype size V improved by more than 5 dB exceeded the number of points at which the sensitivity decreased by more than 5 dB; (3) whether the CFF value increased by more than 20% of the initial value at 1, 4, and 8 weeks after the last stimulation; and (4) whether the peripapillary RNFL and GCC thickness decreased by more than 20% of the initial value. To evaluate the safety of ES, we analyzed the frequency of systemic and skin diseases in the electrode-applied area, fluctuations in the number of corneal endothelial cells, and the frequency of eye diseases. In an additional subgroup analysis, patients were stratified according to whether they had participated in our previous study with ES, and LogMAR was analyzed in accordance with the primary and secondary endpoints.

All statistical analyses, including Wilcoxon's signed rank test, the Friedman test, and the linear mixed-effect model, were performed using EZR (ver.1.64, https://www.jichi.ac.jp/saitama-sct/SaitamaHP.files/download.html) software. A p value of < 0.05 was considered to indicate statistical significance.

Item		Screening	Treatment period				Observational period			Drop out
			1	4	8	12	13	16	20	
	Day	−28	7 ± 2	28 ± 4	56 ± 4	84 ± 4	91 ± 2	112 ± 7	140 ± 14	
IC		●								
BCVA		●	●	●	●	●	●	●	●	●
Slit		●	●	●	●	●	●	●	●	●
IOP		●	●	●	●	●	●	●	●	●
FDS		●	●	●	●	●	●	●	●	●
CFF		●	●	●	●	●	●	●	●	●
CED		●	●			●			●	●
VF		●	●	●			●	●	●	●
OCT		●	●			●			●	●
AE			●	●	●	●	●	●	●	●

Table 4. Treatment timing and examination schedule of the study. IC: informed consent, BCVA: best corrected visual acuity, Slit: slit-lamp exam, IOP: intraocular pressure, fds: fundus examination, CFF: critical flicker frequency, CED: corneal endothelial cell density, VF: visual field, OCT: optical coherence tomography, AE: adverse events.

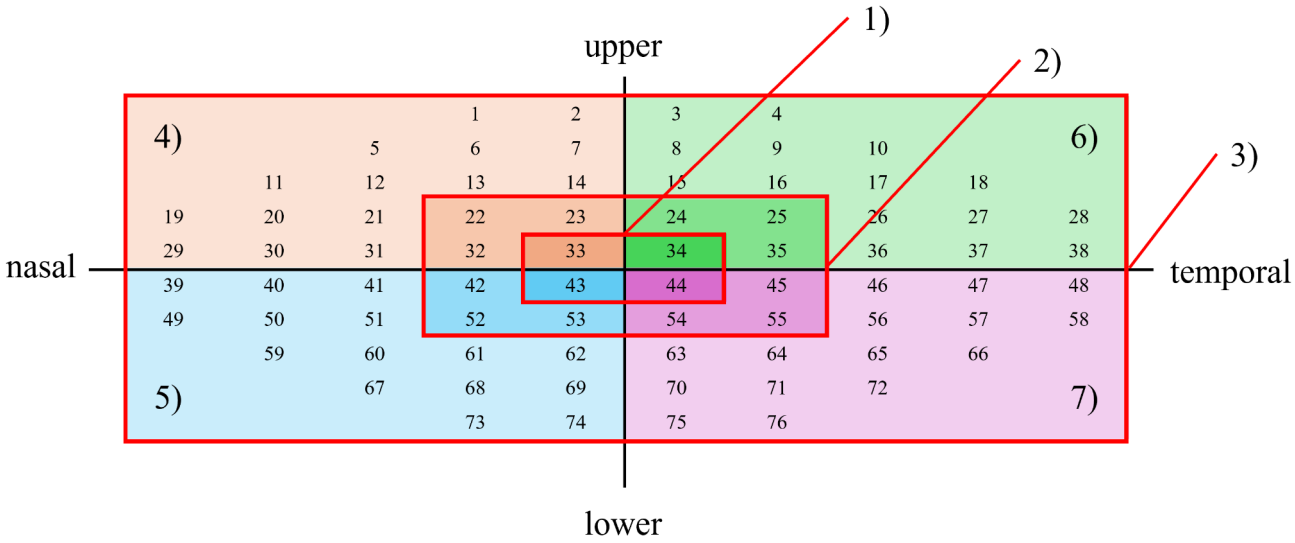

Fig. 5. Schematic image of the visual field analysis (evaluated using Humphrey visual field analyzer, 30 – 2, right eye). The 76 measurement points were divided into seven areas: (1) Small central, 4 points; (2) Large central, 16 points; (3) Total, 76 points; (4) Nasal-upper, 19 points; (5) Nasal-lower, 19 points; (6) Temporal-upper, 19 points; (7) Temporal-lower, 19 points. Owing to the characteristics of LHON, the outermost points were included in the analysis.

Data availability

The datasets used and/or analyzed during the current study available from the corresponding author on reasonable request.

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Author contributions

KU and TK designed this study. FT, KU and TK registered the patients and instructed on how to use the portable ES equipment. FT, KU, MA, TK, YYN, MN examined the patients. TN monitored the study. FT collected the data. FT and KU analysed the result. KU, TK, MN wrote the manuscript. MN supervised the study. All authors read and approved the final manuscript.

Declarations

Statement of Ethics

This study was conducted following the Declaration of Helsinki. Informed consents were obtained from all participants. This clinical trial was approved by the Kobe University Clinical Research Ethical Committee, Japan (No. C190030) and registered with the Japan Registry of Clinical Trials (No. jRCTs052200033, https://jrct.niph.go.jp/). This study was registered on 06/07/2020.

Competing interests

The authors declare no competing interests.

Additional information

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