International consensus statement on the clinical and therapeutic management of

Leber's Hereditary Optic Neuropathy

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Abstract

Leber's hereditary optic neuropathy (LHON) is currently estimated as the most frequent mitochondrial disease (1 in 27,000-45,000). Its molecular pathogenesis and natural history is now fairly well understood. Furthermore, LHON is also the first mitochondrial disease for which a therapy has been approved (based on the antioxidant drug idebenone - Raxone®, Santhera Pharmaceuticals) in June 2015 by the European Medicine Agency (EMA), *under exceptional circumstances* due to the rarity and severity of the disease. However, what remains unclear includes the optimal target population, timing, dose and frequency of administration of idebenone in LHON due to lack of accepted definitions, criteria and general guidelines for the clinical management of this mitochondrial disorder. To address these issues, a consensus conference with a panel of experts from Europe and North America was held in Milan (Italy) in 2016. The conference aimed at providing expert consensus statements for the clinical and therapeutic management of LHON based on the currently available evidence, and this report presents the conclusions of this conference, providing the first guidelines ever for clinical and therapeutic management of LHON.

Introduction

Leber's hereditary optic neuropathy (LHON) is a paradigm for mitochondrial diseases in many regards. It was the first disease to be associated with mitochondrial DNA (mtDNA) point mutations (1), and is hence maternally inherited. These mutations affect complex I subunit genes. LHON is the most frequent mitochondrial disorder with a prevalence ranging from 1 in 27,000 in North-East of England (2), to 1 in 45,000 in a meta-analysis of reports on the European population (3).

LHON is a blinding disorder, usually affecting young men and leading to selective degeneration of retinal ganglion cells (RGCs) and optic atrophy within a year after disease onset. The subset of macular RGCs, providing axons for the papillomacular bundle and serving central vision, are affected first and preferentially, resulting in loss of visual acuity, dyschromatopsia, large central scotomas, and temporal pallor of the optic disc (4,5). Recent optical coherence tomography (OCT) and histopathology studies have substantiated the occurrence of a precise pattern in retinal nerve fiber layer (RNFL) loss, disease progression and natural history (6,7). In particular, loss of macular RGCs precedes the clinical disease onset; by approximately four months, maximal loss has occurred (8).

In unaffected carriers of the LHON mutation there may be recognizable changes on fundus examination and OCT measurements including vascular abnormalities (microangiopathy and telangectatic vessels), hyperemia of the optic disc, and fiber swelling (pseudoedema) that is objectively detected by OCT as increased thickness of the retinal fiber layer at the inferior and temporal quadrants (9,10). The conversion to the symptomatic stage is characterized by loss of macular RGCs on OCT (preclinical changes), but with visual acuity and fields still being normal (8). A central scotoma subsequently develops and central visual acuity starts to deteriorate rapidly, at which stage the patient usually seeks medical attention. The evolution in the first weeks/months is

described canonically as acute/subacute, depending on how rapidly the loss of central vision evolves (11). Within four to six months visual acuity stabilizes, but clinical metrics such as visual fields and OCT measurements may still evolve, usually plateauing at one year after onset. At this point, the so-called acute phase ends with a transition into the chronic stage of the disease. Despite the fact that most patients remain stable for the rest of their lives with profoundly impaired vision, a subgroup may experience some degree of spontaneous visual recovery depending on the mutation subtype and the age at onset (12). This usually manifests as a shrinking and/or a fenestration of the scotoma.

LHON is primarily a clinical diagnosis. The features common to all mitochondrial optic neuropathies consist of central or cecocentral scotomas, impaired color vision and ultimately optic nerve head pallor, especially temporally. In this context, particularly in a young adult, a subacute onset and a maternal family history of visual loss can be very useful in determining the diagnosis. A definitive diagnosis of LHON is currently rapidly obtained by the molecular identification of one of the three common mtDNA mutations (m.11778G>A/MT-ND4, m.3460G>A/MT-ND1, m.14484T>C/MT-ND6), accounting for about 90% of cases. If this primary screen is negative and there is a high index of clinical suspicion supported by a maternal mode of inheritance in a patient with a family history, sequencing the entire mtDNA is advisable to identify one of the rare LHON mutations (13).

The literature frequently states that there is no proven therapy for LHON despite many purported treatments being tested (14,15). However, the European Medicine Agency (EMA) approved in June 2015, *under exceptional circumstances* due to the rarity and severity of the disease, the use of idebenone (Raxone®, Santhera Pharmaceuticals) in LHON, recognizing a sufficient amount of clinical evidence for safety and partial efficacy in a subgroup of treated patients (16-20). The approved product label states that idebenone is indicated for the treatment

of visual impairment in adolescents and adult patients with LHON at a dose of 900mg per day in 3 divided doses. However, there remains some controversy on the optimal target population, timing, dose and frequency of administration of idebenone in LHON patients, compounded by the lack of accepted definitions, criteria and general guidelines for the clinical management of this mitochondrial disorder.

To address these issues, a consensus conference with a panel of experts from Europe and North America was held in Milan (Italy). The conference aimed at providing expert consensus statements for the clinical and therapeutic management of LHON based on the currently available evidence.

Methods

In March 2016 (18^{th}-19^{th}) there was an inaugural congress, *"Update on Optic Nerve Degeneration - a European network: 1^{st} international meeting "*, held in Milan, Italy under the patronage of the San Raffaele Scientific Institute, a tertiary reference health care center in Northern Italy. There were approximately 200 attendees and this provided the opportunity for gathering most of the world experts in mitochondrial optic neuropathies. A satellite meeting "Consensus Conference" focused on the "Clinical and Therapeutic management of LHON" was promoted and hosted by the San Raffaele Institute with technical and secretarial support provided by a private and independent editorial and journalistic consultancy agency – Content Ed (www.contentednet.com).

The expert panel consisted of 16 leading investigators in the field from Europe and North America who were chosen on the basis of their international recognition as experts in mitochondrial optic neuropathies and in particular in LHON and contacted six months in advance.

The final consensus jury included 13 experts, neurologists and ophthalmologists, who accepted to participate and were able to attend in person. Three additional experts (AK, CC-V and MV) did not attend in person the consensus conference, but contributed substantially to different phases of the process. All participants were clinicians and investigators who had extensive experience in clinical and genetic management and research, as well as treatment of optic neuropathies and LHON. Three scientific participants (VC, PB and CLM) were appointed to lead the consensus process serving as the scientific committee. They also participated in the voting sessions due to the limited number of LHON experts and their widely recognized expertise in the field. The consensus process was conducted in line with the last updated methodological indications of the Italian Institute of Health (21). It was developed in 4 stages:

1. Development of clinically relevant questions on LHON and definition of preliminary statements on the basis of a non-systematic literature review,

2. Integration of the feedback from the components of the jury,

3. Consensus meeting with discussion and voting of refined statements on the basis of participants' clinical experience and available evidence,

4. Analysis and publication of the final consensus statements.

The scientific committee identified a number of clinically relevant questions about LHON suitable for consensus discussion and formulated a series of statements addressing each question according to their experience and clinical evidence. All relevant scientific literature, as identified by scientific leaders and two methodological experts, was reviewed in advance in order to identify questions and draft preliminary statements relating to each question. To this end, a literature search was undertaken and was last updated on 15th March 2016. The data pack included studies relevant for the questionnaire topics, as identified by a MEDLINE search using "Leber's Hereditary

Optic Neuropathy" or "Leber Hereditary Optic Neuropathy" or "LHON" as query, with no restrictions on publication date, but only studies published in English were considered. Studies were selected for inclusion if they were: (a) randomized, double-blind, placebo-controlled, or uncontrolled trials; (b) observational studies including prospective or retrospective cohort studies, case-control or cross sectional studies; (c) case series; (d) case reports; (e) systematic reviews and meta-analyses; (f) expert opinion-based pieces; and (f) guidelines. 673 papers were identified and made available to all participants, representing the basis for the proposed statements (listed in Supplementary Materials).

Before the meeting, all members of the jury received the questions and the statements elaborated by scientific leaders, for their assessment and possible changes. All changes and suggestions were considered by the three scientific leaders and the statements were then consolidated in a version that served as the basis for the discussion at the consensus meeting covering four main topics in LHON: (I) Staging; (II) Diagnosis and prognosis; (III) Therapeutic management; (IV) Screening family members; with a total of 20 questions.

The meeting, on March 17th at San Raffaele Scientific Institute in Milan, lasted 7 hours during which the scientific leaders presented each of the questions/statements. Each statement was debated, reformulated when it was considered incorrectly posed or misleading and then voted through a 4-point Likert-type scale handled by an electronic voting system ("1 - completely disagree", "2" partially disagree", "3" partially agree", "4" completely agree"). The total maximum possible score for any given statement was then 52 (in the case all 13 voters completely agreed to the proposed statement allocating a score of 13×4). The "consensus" threshold was set at ≥60% of the total possible score, whilst the "strong~~high~~ consensus" threshold was set at ≥75%. Each participant (including scientific leaders) was equally weighted in scoring the statements. If one of

these two thresholds was met, the statement was considered approved. If a consensus was not reached (i.e. a score of < 60%), the statement was considered not approved.

The results that were originally tabulated at the consensus meeting were submitted in Microsoft PowerPoint™ form to all members of jury, for their final approval. The manuscript reporting the outcomes of the consensus conference was drafted by the scientific leaders and circulated to all members of the jury for final editorial additions and approvals.

Results~~The statements~~

The results of the consensus process, with all questions and related statements, are summarized in Table 1. A strong consensus was reached for 16/20 statements and there was a lack of consensus for 3 statements. The jury decided that there was no scientific basis for voting on one statement (*Statement #14* of the consensus).

(I) Staging

Rationale: Historically, LHON has been categorised into three distinct clinical groups: asymptomatic mutation carriers, patients with acute LHON (with disease duration of one year or less) and those with chronic LHON (with disease duration of more than one year) (9, 11). More recently, the assessment by OCT of the different stages of LHON (10, 22), in conjunction with the natural history (6, 8), and established visual parameters, such as best-corrected visual acuity and visual fields, have provided new insights that have led to the sub-classification of the acute stage into "subacute" and "dynamic" stages (*Statement #1* of the consensus). Furthermore, other phenotypic groups have been proposed, namely, childhood- and late-onset LHON (23, 24), and the

contribution of anatomic (25), environmental (26-30) and hormonal factors (31) have emerged as potential disease modifiers.

Question 1: How would you describe the different clinical stages of LHON?

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Clinical stages of LHON can be defined as follows, according to time from onset and clinical investigations:

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0. Asymptomatic (mutation carriers)
1. Subacute (<6 months from onset)
2. Dynamic (6-12 months)
3. Chronic (>12 months)

The following clinical variants can be also considered:

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1. Slowly progressive, defined according to patients' characteristics independently from time since disease onset
2. Childhood disease, i.e. onset in a patient <12 years old
3. Late onset, i.e. onset in a patient >45 years old

NB. The jury emphasized that for the clinical diagnosis the following clinical investigations were important: visual acuity, colour vision, fundus examination, visual field perimetry, and OCT imaging. The participants also noted that for childhood disease, optic nerve head size was important. For late-onset LHON cases, the participants also felt that it was important to consider toxic exposure (smoking, drinking and environmental factors) and hormonal factors (oestrogens).

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To the statement, there was strong consensus.

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(II) Diagnosis and prognosis

Rationale:

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The diagnosis of LHON has been greatly improved by the availability of genetic testing (32). However, LHON remains a clinical diagnosis with easily recognizable classical features and disease evolution (4,5), which is corroborated by the maternal inheritance of the disease (33) when there is a family history, leading to *Statement #2*.

~~3)~~ Question 2: **Are there any clinical characteristics, algorithms or investigations to reach a rapid diagnosis of LHON?**

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Diagnosis of LHON can be usually made based on patient and family clinical history as well as baseline investigations including a formal neuro-ophthalmological examination and mtDNA genetic testing

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NB **There was a strong consensus**

Rationale: Despite the greatly improved recognition of LHON, there can still be considerable diagnostic delay that is compounded by variability in the initial findings and misinterpretation of the clinical picture, in particular with optic neuritis (34,35) or toxic optic neuropathy (36), thus a differential diagnosis remains an important point to consider, as remarked in *Statements #3 and #4.*

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~~4)~~ Question 3: —**Typically, how long does it take from symptom onset to confirmation of diagnosis?**

"It can take months to reach a confirmed diagnosis"

NB **There was a strong consensus**

~~5)~~ Question 4: **What are the main differential diagnoses?**

- *Other optic neuropathies especially optic neuritis (neuromyelitis optica), toxic, metabolic and compressive optic neuropathies*
- *Maculopathies*
- *Non organic visual loss (not a diagnosis of exclusion)*

If further extraocular features are present, consider LHON 'Plus', including LHON/MS-like variant

Consider performing appropriate investigations including brain MRI and laboratory studies

NB **There was strong consensus**

Rationale: Multiple reports on large LHON cohorts from different countries have defined some prognostic factors, which may impact the clinical course (23, 25, 37-39) and possibly the management of the disease, leading to *Statement #5*.

~~6)~~ Question 5: **How would you assess a patient's prognosis? Does this impact on your management?**

In LHON, positive prognostic factors are:

- *Younger age*
- *Type of mutation (14484/ND6)*

However, prognostic factors do not affect management

NB **There was a strong consensus**

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(III) Therapeutic management

Rationale: The standard clinical evaluation of LHON patients includes visual acuity and visual fields. OCT studies have proved a valuable tool in evaluating the clinical course of the disease (6, 8, 10, 22). These considerations lead to *Statement #6.*

~~7)~~ **Question 6: Before initiating ANY treatment, what baseline assessments do you usually perform?**

The following examinations should be performed before starting any treatment:

- *Visual acuity (ETDRS charts)*
- *Automated visual field test*
- *OCT (optic nerve head and RNFL analysis, and macular ganglion cell analysis)*

NB **There was a strong consensus**

Rationale: Idebenone has been used as treatment of patients with LHON since 1992 (40-43). A number of publications have recently evaluated the therapeutic benefit of idebenone in LHON, including a placebo-controlled randomised clinical trial of 85 patients treated for 24 weeks with a dosage of 900 mg/day (16, 19, 20) and a large retrospective case-series of 44 patients treated with variable doses of idebenone over a mean treatment duration of 41 months compared with 59 untreated patients (17). Based on the available literature and the personal experience of the experts, S*tatements #7-16* were formulated to critically evaluate a number of issues, including inclusion criteria for treatment, dosage and duration of treatment, as well as follow-up evaluations for assessing a clinically relevant response to treatment.

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~~8)~~ Question 7: **In subacute/dynamic (<6 months, 6-12 months) patients, at which stage of disease would you ideally start 900 mg/day treatment?**

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"Idebenone should be started as soon as possible at 900 mg/day in patients with disease less than one year"

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NB **There was a strong consensus** with one participant in partial disagreement

~~9)~~ Question 8: **In chronic cases, do you consider treatment?**

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There is not enough evidence to recommend treatment in chronic patients between 1 and 5 years (after the 2nd eye onset), and no evidence to recommend treatment in chronic patients > 5 years (after the 2nd eye onset)

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-NB **There was a strong consensus** but two participants disagreed

Question 9: **What is your suggested frequency of follow-up, would it differ by stage?**

The ideal frequency of follow-up is as follows:

- *Approximately every 3 months for subacute and dynamic cases*
- *Approximately every 6 months for the 2nd year*
- *Once a year thereafter*

NB There was a strong consensus

Question 10: **How would you define a clinically relevant response (recovery of vision) to treatment?**

Response should be defined according to:

- *Improvement of two lines of BCVA on ETDRS charts (or from off-chart to on-chart)*

- *Automated visual field test (MD)*

NB There was strong consensus with one participant in partial disagreement

Question 11: How long would you treat at 900 mg/day in order to assess response?

"In subacute/dynamic patients, treatment at 900 mg/day should be continued for at least one year in order to assess the start of therapeutic response or until a plateau in terms of improvement is reached"

NB There was strong consensus

Question 12: Once you have confirmed a favorable clinically relevant outcome, for how long after plateau would you continue treatment?

"One year"

NB There was strong consensus but two participants partially disagreed and one participant disagreed

Question 13: If you treat a chronic patient for how long you would continue treatment?

"One year"

NB Consensus was not reached. Five participants did not vote.

Question 14: For chronic patients once you have confirmed a favorable clinically relevant outcome, for how long after plateau would you continue treatment?

"One year"

NB Not voted due to lack of consensus on statement #13.

Question 15: What assessments do you perform to evaluate response during maintenance phase?

The following examinations should be performed to assess response during maintenance phase:

- *Visual acuity*
- *Automated visual field test*

NB There was strong consensus.

Question 16: Do you consider a maintenance dose also for non-responders?

"In non-responders, the maintenance dose can be 300 mg/day"

NB There was strong disagreement with one participant in agreement

(IV) Screening of family members

Rationale: The provision of genetic testing (32) of the proband's maternal lineage poses a number of ethical and economical issues, which led to *Statement #17.*

Question 17: Is it necessary to perform genetic screening for LHON mutation in all maternally-related family members?

"No"

NB There was a strong consensus

Rationale: Similarly, the clinical evaluation and prophylactic treatment of unaffected LHON mutation carriers is a particularly relevant issue (9, 10, 25-27, 29, 39, 44-47). This point has been developed in *Statements #18 and #19.*

Question 18: Should all maternally-related relatives be clinically screened?

Yes

Commented [C1]: Reviewer 2 is suggesting to consider factors (such as age, angio-OCT) that should be taken into account in determine which subjects should be screened or not

NB 11 participants disagreed that all maternally-related relatives should be screened**. There was strong disagreement** with two participants in agreement

Question 19: Would you consider treating them?

"Currently treatment is not recommended for relatives of a LHON patient, but lifestyle counseling is recommended"

NB There was a strong consensus

Rationale: There are a few proposed risk factors for disease conversion (25, 27, 29, 39, 48). However, a systematic validation of these factors translated into a scoring system predicting disease risk is still pending. This issue led to the final *Statement #20.*

Question 20: Are you aware of any algorithm or predictive risk factors that could be used to assess risk of becoming symptomatic?

Currently there is no clinical prognostic factor that can be used

NB There was strong consensus with one participant in disagreement.

Commented [C2]: Reviewer 2 wants to know which risk factor is this participant considering a reliable?

The Jury unanimously endorsed the final version of all questions and statements (Table 1).

Discussion

There was strong consensus for 16 statements, whereas for 3 statements there was lack of consensus (#13,#16, and #18). For statements #8, #12 and #20 there was specific disagreement from two, one and one participants, respectively. Specifically, for statement #8 the two participants thought that there was sufficient evidence to justify the use of idebenone for chronic

cases. For statement #12, one participant thought that idebenone treatment could be continued for more than 1 year after plateau in patients who had shown a treatment response. For statement #20, one participant disagreed that there is no reliable predictive risk factor to assess risk of disease conversion in a LHON carrier.

There was strong disagreement for statements #16 and #18. Eleven out of 13 participants fully disagreed that idebenone should be continued with a maintenance dose in non-responder patients. Also, 11 out of 13 participants fully disagreed that it was necessary to clinically screen all maternal relatives including asymptomatic carriers.

Statements #1 to #6 and #9 pertained to the diagnosis and work-up of patients who presented with LHON for which there was strong consensus. The group debated the historical staging (9,11) of the disease and introduced a new distinction. Based on the currently available published evidence (6,8), the first year of the disease can be subdivided into subacute (less than 6 months) and dynamic (6-12 months) phases, based on both functional and structural changes. Visual acuity decreases throughout the subacute phase and then stabilizes at about 6 months. Congruently, the thinning of macular RGC layer measured by OCT is largely completed by 4 to 6 months, highlighting the severe cell loss of the cells originating the papillomacualr bundle (8, 49). Conversely, RNFL thickness evaluated by OCT is characterized in the first 6 months by swelling, followed by progressive quadrant specific thinning (6). Thus, the dynamic stage, 6 to 12 months after onset, is characterized by still ongoing changes in RNFL as opposed to a substantial stability of RGC loss in the macula (8) Visual field defects may still progress in this dynamic stage (50). Although the ideal therapeutic window for LHON remains unknown, it is possible that there is a greater potential for visual recovery in the subacute phase compared with the dynamic phase.

Statements #7, #8 and #10-16 pertained to the use of idebenone in LHON. The group agreed with consensus that idebenone was indicated in subacute and dynamic patients at 900

mg/day dosage (16), starting the treatment as soon as possible (17). There was also consensus about the continuation of the treatment in responders, based on the established outcome measures of best-corrected visual acuity and visual fields, and the criteria for defining a clinically relevant response (17). The group strongly disagreed with the use of Idebenone as a maintenance dose in non-responders who have received treatment for one year. Concerning patients in the chronic stage (> 1 year), it was determined that there was not enough evidence to support treatment for those patients with disease duration of 1-5 years, and no evidence for treatment after 5 years of disease duration. As a consequence, there was no consensus on statement #13 and the Jury unanimously agreed not to vote on statement #14. The group based its judgment strictly on the existing data on clinical administration of idebenone in patients, as derived from the published literature (14-20, 40-43) and the unpublished information publicly available at the EMA website, including the European public assessment report (EPAR; ema.europa.eu/Find medicine/Human medicines/European public assessment reports) and the summary of the opinion of the Committee for Orphan Medicinal Products for Raxone (ema.europa.eu/Find medicine/Human medicines/Rare disease designation). Preclinical research has also been considered, with specific studies in cells derived from LHON patients as well as in animal models (51-55), being aware that the effectiveness and mechanisms of action of idebenone are still debated by the scientific community.

Statements #17-20 pertained to the proband's maternally-related relatives and specifically unaffected carriers. The group agreed that the unaffected mutation carriers did not require routine genetic or clinical investigations. However, as best practice with any genetic disease, genetic counseling is offered to the patient who has the right to refuse any wider family involvement. Furthermore, the group had a strong consensus that treatment is not recommended

for asymptomatic carriers and that there is currently no prognostic measure to predict disease conversion.

Conclusions and relevance for clinical practice

The diagnosis of LHON should be based on a careful history, the evaluation of key structural and functional visual parameters, and on a molecular confirmation of a pathogenic mtDNA mutation. The management of LHON includes the offer of genetic counseling, informing the patient about potentially preventable lifestyle risk factors and, for subacute and dynamic cases, the use of idebenone at the currently approved dose. Idebenone should be discontinued in non-responder patients and is currently not recommended in patients in the chronic stages of the disease. These guidelines and recommendations are based on a consensus developed on the current state of the literature. Further investigations and clinical trials are needed to lead to better disease-modifying treatments and to improve the management of patients with LHON.

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Conflict of Interest Disclosures:

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References

1. **Wallace DC**, Singh G, Lott MT, Hodge JA, Schurr TG, Lezza AM, Elsas LJ 2nd, Nikoskelainen EK. Mitochondrial DNA mutation associated with Leber's hereditary optic neuropathy. Science. 1988;242:1427-30.
2. **Gorman GS**, Schaefer AM, Ng Y, Gomez N, Blakely EL, Alston CL, Feeney C, Horvath R, Yu-Wai-Man P, Chinnery PF, Taylor RW, Turnbull DM, McFarland R. Prevalence of nuclear and mitochondrial DNA mutations related to adult mitochondrial disease. Ann Neurol. 2015;77:753-9.
3. **Mascialino B**, Leinonen M, Meier T. Meta-analysis of the prevalence of Leber hereditary optic neuropathy mtDNA mutations in Europe. Eur J Ophthalmol. 2012;22:461-5.
4. **Carelli V**, Ross-Cisneros FN, Sadun AA. Mitochondrial dysfunction as a cause of optic neuropathies. Prog Retin Eye Res. 2004;23:53-89.
5. **Yu-Wai-Man P**, Griffiths PG, Chinnery PF. Mitochondrial optic neuropathies - disease mechanisms and therapeutic strategies. Prog Retin Eye Res. 2011;30:81-114.
6. **Barboni P**, Carbonelli M, Savini G, Ramos Cdo V, Carta A, Berezovsky A, Salomao SR, Carelli V, Sadun AA. Natural history of Leber's hereditary optic neuropathy: longitudinal analysis of the retinal nerve fiber layer by optical coherence tomography. Ophthalmology 2010;117:623-7.
7. **Pan BX**, Ross-Cisneros FN, Carelli V, Rue KS, Salomao SR, Moraes-Filho MN, Moraes MN, Berezovsky A, Belfort R Jr, Sadun AA. Mathematically modeling the involvement of axons in Leber's hereditary optic neuropathy. Invest Ophthalmol Vis Sci. 2012;53:7608-17.

8. **Balducci N**, Savini G, Cascavilla ML, La Morgia C, Triolo G, Giglio R, Carbonelli M, Parisi V, Sadun AA, Bandello F, Carelli V, Barboni P. Macular nerve fibre and ganglion cell layer changes in acute Leber's hereditary optic neuropathy. Br J Ophthalmol. 2016; 100:1232-7.
9. **Nikoskelainen E**, Hoyt WF, Nummelin K. Ophthalmoscopic findings in Leber's hereditary optic neuropathy. I. Fundus findings in asymptomatic family members. Arch Ophthalmol. 1982;100:1597-602.
10. **Savini G**, Barboni P, Valentino ML, Montagna P, Cortelli P, De Negri AM, Sadun F, Bianchi S, Longanesi L, Zanini M, Carelli V. Retinal nerve fiber layer evaluation by optical coherence tomography in unaffected carriers with Leber's hereditary optic neuropathy mutations. Ophthalmology. 2005;112:127-31.
11. **Nikoskelainen E**, Hoyt WF, Nummelin K. Ophthalmoscopic findings in Leber's hereditary optic neuropathy. II. The fundus findings in the affected family members. Arch Ophthalmol. 1983;101:1059-68.
12. **Stone EM**, Newman NJ, Miller NR, Johns DR, Lott MT, Wallace DC. Visual recovery in patients with Leber's hereditary optic neuropathy and the 11778 mutation. J Clin Neuroophthalmol. 1992;12:10-4.
13. **Achilli A**, Iommarini L, Olivieri A, Pala M, Hooshiar Kashani B, Reynier P, La Morgia C, Valentino ML, Liguori R, Pizza F, Barboni P, Sadun F, De Negri AM, Zeviani M, Dollfus H, Moulignier A, Ducos G, Orssaud C, Bonneau D, Procaccio V, Leo-Kottler B, Fauser S, Wissinger B, Amati-Bonneau P, Torroni A, Carelli V. Rare primary mitochondrial DNA mutations and probable synergistic variants in Leber's hereditary optic neuropathy. PLoS One. 2012;7:e42242.
14. **Newman NJ**. Treatment of hereditary optic neuropathies. Nat Rev Neurol. 2012;8(:545-56.

15. **Yu-Wai-Man P**, Votruba M, Moore AT, Chinnery PF. Treatment strategies for inherited optic neuropathies: past, present and future. Eye (Lond). 2014;28:521-37.
16. **Klopstock T**, Yu-Wai-Man P, Dimitriadis K, Rouleau J, Heck S, Bailie M, Atawan A, Chattopadhyay S, Schubert M, Garip A, Kernt M, Petraki D, Rummey C, Leinonen M, Metz G, Griffiths PG, Meier T, Chinnery PF. A randomized placebo-controlled trial of idebenone in Leber's hereditary optic neuropathy. Brain. 2011;134:2677-86.
17. **Carelli V**, La Morgia C, Valentino ML, Rizzo G, Carbonelli M, De Negri AM, Sadun F, Carta A, Guerriero S, Simonelli F, Sadun AA, Aggarwal D, Liguori R, Avoni P, Baruzzi A, Zeviani M, Montagna P, Barboni P. Idebenone treatment in Leber's hereditary optic neuropathy. Brain. 2011;134:e188.
18. **Newman NJ**. Treatment of Leber hereditary optic neuropathy. Brain. 2011;134:2447-50.
19. **Klopstock T**, Metz G, Yu-Wai-Man P, Büchner B, Gallenmüller C, Bailie M, Nwali N, Griffiths PG, von Livonius B, Reznicek L, Rouleau J, Coppard N, Meier T, Chinnery PF. Persistence of the treatment effect of idebenone in Leber's hereditary optic neuropathy. Brain. 2013;136:e230.
20. **Rudolph G**, Dimitriadis K, Büchner B, Heck S, Al-Tamami J, Seidensticker F, Rummey C, Leinonen M, Meier T, Klopstock T. Effects of idebenone on color vision in patients with leber hereditary optic neuropathy. J Neuroophthalmol. 2013;33:30-6.
21. **Candiani g**, Colombo c, Daghini R et al. (2013) [Methodological handbook – How to Organize a Consensus Conference]. Italian Institute of Health: Rome. Available at http://www.snlg-iss.it/en_CC_methodological_handbook. Last accessed 29/07/2016.
22. **Barboni P**, Savini G, Valentino ML, Montagna P, Cortelli P, De Negri AM, Sadun F, Bianchi S, Longanesi L, Zanini M, de Vivo A, Carelli V. Retinal nerve fiber layer evaluation by optical

coherence tomography in Leber's hereditary optic neuropathy. Ophthalmology. 2005;112:120-6.

23. **Barboni P**, Savini G, Valentino ML, La Morgia C, Bellusci C, De Negri AM, Sadun F, Carta A, Carbonelli M, Sadun AA, Carelli V. Leber's hereditary optic neuropathy with childhood onset. Invest Ophthalmol Vis Sci. 2006;47:5303-9.
24. **Dimitriadis K,** Leonhardt M, Yu-Wai-Man P, Kirkman MA, Korsten A, De Coo IF, Chinnery PF, Klopstock T. Leber's hereditary optic neuropathy with late disease onset: clinical and molecular characteristics of 20 patients. Orphanet J Rare Dis. 2014;9:158.
25. **Ramos Cdo V**, Bellusci C, Savini G, Carbonelli M, Berezovsky A, Tamaki C, Cinoto R, Sacai PY, Moraes-Filho MN, Miura HM, Valentino ML, Iommarini L, De Negri AM, Sadun F, Cortelli P, Montagna P, Salomao SR, Sadun AA, Carelli V, Barboni P. Association of optic disc size with development and prognosis of Leber's hereditary optic neuropathy. Invest Ophthalmol Vis Sci. 2009;50:1666-74.
26. **Sadun AA**, Carelli V, Salomao SR, Berezovsky A, Quiros PA, Sadun F, DeNegri AM, Andrade R, Moraes M, Passos A, Kjaer P, Pereira J, Valentino ML, Schein S, Belfort R. Extensive investigation of a large Brazilian pedigree of 11778/haplogroup J Leber hereditary optic neuropathy. Am J Ophthalmol. 2003;136:231-8.
27. **Kirkman MA**, Yu-Wai-Man P, Korsten A, Leonhardt M, Dimitriadis K, De Coo IF, Klopstock T, Chinnery PF. Gene-environment interactions in Leber hereditary optic neuropathy. Brain. 2009;132:2317-26.
28. **Newman NJ**. Leber hereditary optic neuropathy: bad habits, bad vision? Brain. 2009;132:2306-8.
29. **Carelli V**, d'Adamo P, Valentino ML, La Morgia C, Ross-Cisneros FN, Caporali L, Maresca A, Loguercio Polosa P, Barboni P, De Negri A, Sadun F, Karanjia R, Salomao SR, Berezovsky A,

Chicani F, Moraes M, Moraes Filho M, Belfort R Jr, Sadun AA. Parsing the differences in affected with LHON: genetic versus environmental triggers of disease conversion. Brain. 2016;139:e17.

30. **Yu-Wai-Man P**, Hudson G, Klopstock T, Chinnery PF. Reply: Parsing the differences in affected with LHON: genetic versus environmental triggers of disease conversion. Brain. 2016;139:e18.
31. **Giordano C**, Montopoli M, Perli E, Orlandi M, Fantin M, Ross-Cisneros FN, Caparrotta L, Martinuzzi A, Ragazzi E, Ghelli A, Sadun AA, d'Amati G, Carelli V. Oestrogens ameliorate mitochondrial dysfunction in Leber's hereditary optic neuropathy. Brain. 2011;134:220-34.
32. **Maresca A**, Caporali L, Strobbe D, Zanna C, Malavolta D, La Morgia C, Valentino ML, Carelli V. Genetic Basis of Mitochondrial Optic Neuropathies. Curr Mol Med. 2014; 14: 985-992.
33. **Erickson RP**. Leber's optic atrophy, a possible example of maternal inheritance. Am J Hum Genet. 1972;24:348-9.
34. **McClelland CM**, Van Stavern GP, Tselis AC. Leber hereditary optic neuropathy mimicking neuromyelitis optica. J Neuroophthalmol. 2011;31:265-8.
35. **Hsu TK**, Wang AG, Yen MY, Liu JH. Leber's hereditary optic neuropathy masquerading as optic neuritis with spontaneous visual recovery. Clin Exp Optom. 2014;97:84-6.
36. **Cullom ME**, Heher KL, Miller NR, Savino PJ, Johns DR. Leber's hereditary optic neuropathy masquerading as tobacco-alcohol amblyopia. Arch Ophthalmol. 1993;111:1482-5.
37. **Riordan-Eva P**, Sanders MD, Govan GG, Sweeney MG, Da Costa J, Harding AE. The clinical features of Leber's hereditary optic neuropathy defined by the presence of a pathogenic mitochondrial DNA mutation. Brain. 1995;118:319-37.

38. **Nikoskelainen EK**, Huoponen K, Juvonen V, Lamminen T, Nummelin K, Savontaus ML. Ophthalmologic findings in Leber hereditary optic neuropathy, with special reference to mtDNA mutations. Ophthalmology. 1996;103:504-14.

39. **Giordano C**, Iommarini L, Giordano L, Maresca A, Pisano A, Valentino ML, Caporali L, Liguori R, Deceglie S, Roberti M, Fanelli F, Fracasso F, Ross-Cisneros FN, D'Adamo P, Hudson G, Pyle A, Yu-Wai-Man P, Chinnery PF, Zeviani M, Salomao SR, Berezovsky A, Belfort R Jr, Ventura DF, Moraes M, Moraes Filho M, Barboni P, Sadun F, De Negri A, Sadun AA, Tancredi A, Mancini M, d'Amati G, Loguercio Polosa P, Cantatore P, Carelli V. Efficient mitochondrial biogenesis drives incomplete penetrance in Leber's hereditary optic neuropathy. Brain. 2014;137:335-53.

40. **Mashima Y**, Hiida Y, Oguchi Y. Remission of Leber's hereditary optic neuropathy with idebenone. Lancet. 1992;340:368-9.

41. **Cortelli P**, Montagna P, Pierangeli G, Lodi R, Barboni P, Liguori R, Carelli V, Iotti S, Zaniol P, Lugaresi E, Barbiroli B. Clinical and brain bioenergetics improvement with idebenone in a patient with Leber's hereditary optic neuropathy: a clinical and 31P-MRS study. J Neurol Sci. 1997;148:25-31.

42. **Carelli V**, Barboni P, Zacchini A, Mancini R, Monari L, Cevoli S, Liguori R, Sensi M, Lugaresi E, Montagna P. Leber's Hereditary Optic Neuropathy (LHON) with 14484/ND6 mutation in a North African patient. J Neurol Sci. 1998;160:183-8.

43. **Mashima Y**, Kigasawa K, Wakakura M, Oguchi Y. Do idebenone and vitamin therapy shorten the time to achieve visual recovery in Leber hereditary optic neuropathy? J Neuroophthalmol. 2000;20:166-70.

44. **Quiros PA**, Torres RJ, Salomao S, Berezovsky A, Carelli V, Sherman J, Sadun F, De Negri A, Belfort R, Sadun AA. Colour vision defects in asymptomatic carriers of the Leber's

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hereditary optic neuropathy (LHON) mtDNA 11778 mutation from a large Brazilian LHON pedigree: a case-control study. Br J Ophthalmol. 2006;90:150-3.

45. **Ventura DF**, Gualtieri M, Oliveira AG, Costa MF, Quiros P, Sadun F, de Negri AM, Salomão SR, Berezovsky A, Sherman J, Sadun AA, Carelli V. Male prevalence of acquired color vision defects in asymptomatic carriers of Leber's hereditary optic neuropathy. Invest Ophthalmol Vis Sci. 2007;48:2362-70.
46. **Yee KM**, Ross-Cisneros FN, Lee JG, Da Rosa AB, Salomao SR, Berezovsky A, Belfort R Jr, Chicani F, Moraes-Filho M, Sebag J, Carelli V, Sadun AA. Neuron-specific enolase is elevated in asymptomatic carriers of Leber's hereditary optic neuropathy. Invest Ophthalmol Vis Sci. 2012;53:6389-92.
47. **Barboni P**, Savini G, Feuer WJ, Budenz DL, Carbonelli M, Chicani F, Ramos Cdo V, Salomao SR, Negri AD, Parisi V, Carelli V, Sadun AA. Retinal nerve fiber layer thickness variability in Leber hereditary optic neuropathy carriers. Eur J Ophthalmol. 2012;22:985-91.
48. **Giordano L**, Deceglie S, d'Adamo P, Valentino ML, La Morgia C, Fracasso F, Roberti M, Cappellari M, Petrosillo G, Ciaravolo S, Parente D, Giordano C, Maresca A, Iommarini L, Del Dotto V, Ghelli AM, Salomao SR, Berezovsky A, Belfort R Jr, Sadun AA, Carelli V, Loguercio Polosa P, Cantatore P. Cigarette toxicity triggers Leber's hereditary optic neuropathy by affecting mtDNA copy number, oxidative phosphorylation and ROS detoxification pathways. Cell Death Dis. 2015;6:e2021.
49. **Yu-Wai-Man P**, Votruba M, Burté F, La Morgia C, Barboni P, Carelli V. A neurodegenerative perspective on mitochondrial optic neuropathies. Acta Neuropathol. 2016. 132(6):789-806.
50. **Newman NJ**, Biousse V, Newman SA, Bhatti MT, Hamilton SR, Farris BK, Lesser RL, Turbin RE. Progression of visual field defects in leber hereditary optic neuropathy: experience of the LHON treatment trial. Am J Ophthalmol. 2006;141:1061-1067.

51. **Angebault C**, Gueguen N, Desquiret-Dumas V, Chevrollier A, Guillet V, Verny C, Cassereau J, Ferre M, Milea D, Amati-Bonneau P, Bonneau D, Procaccio V, Reynier P, Loiseau D. Idebenone increases mitochondrial complex I activity in fibroblasts from LHON patients while producing contradictory effects on respiration. BMC Res Notes. 2011;4:557.

52. **Erb M**, Hoffmann-Enger B, Deppe H, Soeberdt M, Haefeli RH, Rummey C, Feurer A, Gueven N. Features of idebenone and related short-chain quinones that rescue ATP levels under conditions of impaired mitochondrial complex I. PLoS One. 2012:e36153.

53. **Giorgio V**, Petronilli V, Ghelli A, Carelli V, Rugolo M, Lenaz G, Bernardi P. The effects of idebenone on mitochondrial bioenergetics. Biochim Biophys Acta. 2012;1817:363-9.

54. **Yu-Wai-Man P**, Soiferman D, Moore DG, Burté F, Saada A. Evaluating the therapeutic potential of idebenone and related quinone analogues in Leber hereditary optic neuropathy. Mitochondrion. 2017. pii: S1567-7249(17)30012-0. [Epub ahead of print]

~~50.~~55. **Heitz FD**, Erb M, Anklin C, Robay D, Pernet V, Gueven N. Idebenone protects against retinal damage and loss of vision in a mouse model of Leber's hereditary optic neuropathy. PLoS One. 2012:e45182.