LEARNING OBJECTIVES
1. Know the natural history of the two most common forms of hereditary optic neuropathy, Leber hereditary optic neuropathy (LHON) and dominant optic atrophy (DOA).
2. Know the available evidence in the literature on the treatment of the mitochondrial diseases in general and of the hereditary optic neuropathies in particular.
3. Be familiar with the future options for treatment of the hereditary optic neuropathies.
4. To know the range of presentations and course for Leber’s hereditary optic neuropathy (LHON) and dominant optic atrophy (DOA).
5. To understand the possible pathophysiological mechanisms by which LHON and DOA may cause RGC loss.
6. To adequately weigh the evidence for treatment of LHON and DOA.

CME QUESTIONS
1. True or False? Natural history studies have shown that all males who have homoplasmic mtDNA mutations for LHON will have visual loss some time during their lifetime.
2. True or False? The evidence supports treatment of acute LHON with ubiquinone, vitamins and anti-oxidants.
3. True or False? Gene therapy in LHON involves direct insertion of replacement mtDNA into the mitochondria.
4. True or False? All cases of LHON begin with an acute or subacute loss of vision.
5. True or False? Patients with LHON who have visual loss and optic atrophy do not recover any substantial level of visual acuity or visual fields.

KEY WORDS
1. Leber Hereditary Optic Neuropathy
2. Dominant Optic Atrophy
3. Mitochondrial Disease
4. Hereditary optic neuropathy
5. LHON

EVIDENCE PRESENTATION
The past two decades have witnessed remarkable advances in our understanding of the clinical presentation, genetics and even the pathophysiology of the hereditary optic neuropathies, specifically Leber hereditary optic neuropathy (LHON) and dominant optic atrophy (DOA).1-4. We now know that most of the hereditary optic neuropathies, including LHON and DOA, have a pathophysiology reflecting a final common pathway in mitochondrial dysfunction, despite their genetic origin in two different genomes (LHON a result of point mutations in the mitochondrial DNA, and DOA a consequence of mutations in nuclear chromosomes). However, investigations into potential therapies for these and other mitochondrial disorders are still in their nascency. Before reviewing the evidence currently available on the treatment of these disorders, it is important to discuss the natural history of visual loss in these clinical settings, specifically the prognosis for spontaneous visual recovery.

NATURAL HISTORY OF VISUAL LOSS IN LHON AND DOA
LHON
In most patients with LHON, vision loss is devastating and permanent, with visual acuities typically worse than 20/200 in both eyes.5 Approximately 50% of patients with visual loss from LHON will recognize sequential eye symptoms, with intervals between affected eyes ranging from days to months, with a typical interval of 2 to 4 months.5,6 At least 97% of patients with visual loss in one eye will have second eye involvement within 1 year.7 In some patients, visual recovery may occur after acute visual loss, sometimes manifested as a “fenestration” within a visual field defect (the so-called donut or bagel scotoma) or with more diffuse return of central visual acuity and color vision, usually bilaterally.2,8-10 Visual recovery, when
Symptomatic treatments should be considered in all DOA patients.\textsuperscript{18,22-26} The rate of progression varies. Progressive decline in visual acuity occurs in 19-67% of the majority of patients.\textsuperscript{17} LHON, DOA may nevertheless significantly impair quality of life in the majority of patients.\textsuperscript{14-16} An additional positive prognostic feature is an age of onset less than 20 years, and especially less than 10 years.\textsuperscript{2,8,15} It has also been suggested that thicker RNFL and larger optic disc vertical diameter on OCT may be associated with a better visual prognosis.\textsuperscript{14-16}

DOA

Visual loss in DOA is detected between ages 4 and 6 in the majority of patients.\textsuperscript{17} and 58–84% of patients with DOA report visual impairment by age 11.\textsuperscript{18,19} Compared to LHON, vision loss is typically mild in DOA, with a mean visual acuity of 20/80 to 20/120.\textsuperscript{20-22} More than 80% of patients retain vision of 20/200 or better,\textsuperscript{2,23} although visual acuities can range from 20/20 to light perception.\textsuperscript{2,23} Although not as rapid or as devastating as LHON, DOA may nevertheless significantly impair quality of life in the majority of patients.

Progressive decline in visual acuity occurs in 19–67% of DOA patients.\textsuperscript{18,22-26} The rate of progression varies considerably among and within families;\textsuperscript{22} however, in general, disease progression in DOA follows a relatively indolent course, and is independent of visual acuity at diagnosis.\textsuperscript{24} In one long-term follow-up study of 69 patients with a confirmed DOA-causing mutation, of whom 58 (84%) were symptomatic, 43 (62%) had stable visual acuity in at least one eye at 10 year follow-up.\textsuperscript{18} Although 10% of patients in that single study had improvement in their vision, this may reflect improved testing as children age; true substantial spontaneous improvement of vision does not appear to be a feature of DOA. In a more recent study of DOA patients in the north of England, visual function worsened in 29 of 43 patients (67.4%) for whom there was longitudinal follow-up (mean follow-up time of 15 years).\textsuperscript{26}

TREATMENTS FOR LHON AND DOA: REVIEW OF THE LITERATURE

Symptomatic Treatments

Symptomatic treatments should be considered in all patients with vision-impairing optic neuropathies to improve quality of life, in particular to aid with reading, communication, gainful employment, navigation, and self-operation of a motor-vehicle.\textsuperscript{27} Low vision aids may benefit patients with severe vision loss from optic neuropathies.\textsuperscript{28} In particular, patients with LHON and DOA are often young adults with preserved peripheral vision, and make excellent candidates for low vision rehabilitation.

Although avoiding agents that may act as mitochondrial "stressors" is a non-specific recommendation for all patients with disorders with a presumed mitochondrial pathophysiology, there is no study which has shown proven benefit.\textsuperscript{29} One recent epidemiologic study suggested that vision loss does indeed occur more often in individuals at risk for LHON who smoke, and possibly those with heavy alcohol intake.\textsuperscript{30} It may be prudent to caution patients to avoid tobacco use, excessive alcohol intake, cyanide-containing products, medications which may have mitochondrial toxicity, and environmental toxins, especially during the acute phase of visual loss.\textsuperscript{31}

DISEASE-MODIFYING TREATMENTS

Treatments for Mitochondrial Disorders

Therapies for mitochondrial disorders are very limited. A 2006 Cochrane review of 678 abstracts and articles found no evidence supporting any intervention in the management of mitochondrial disease.\textsuperscript{29} General therapies that have been studied in mitochondrial disease fall into four main categories:\textsuperscript{32} 1) vitamins and cofactors (Coenzyme Q10(CoQ10), folic acid, vitamin B12, thiamine, riboflavin, L-carnitine, and creatine); 2) electron acceptors (vitamin C, menadiol); 3) free radical scavengers (CoQ10, idebenone, alpha-lipoic acid, and vitamin E); and 4) inhibitors of toxic metabolites (dichloroacetate(DCA)). Most of these general therapies are harmless at their usual doses, although some may be expensive. In the absence of any other proven therapy in mitochondrial disease, many clinicians resort, on theoretical or anecdotal grounds alone, to “mitochondrial cocktails” — various combinations of these agents — to treat their patients.

Coenzyme Q10 (CoQ10) is a lipophilic molecule found in the mitochondrial membrane that shuttles electrons from complex I and II to complex III. In patients with primary CoQ10 deficiency, OXPHOS is interrupted and ATP synthesis is impaired with consequent mitochondrial encephalomyopathy. In some of these patients, supplementation with exogenous CoQ10 has led to clear improvement in function, and doses of up to 3000mg/d of CoQ10 were tolerated without side-effects in other neurological populations.\textsuperscript{33-38} Because of its therapeutic usefulness in treating primary CoQ10 deficiency, exogenous CoQ10 therapy is frequently used to treat other diseases of the OXPHOS system, including LHON. Doses of greater than 400mg per day are typical.\textsuperscript{32} Similarly, exogenous riboflavin (100mg daily) and L-carnitine (3g daily) supplementation, useful in the treatment of multiple acyl-CoA dehydrogenase deficiency and primary carnitine deficiency, respectively,\textsuperscript{39,40} have had their use...
extended to mitochondrial disorders, (although not usually to the hereditary optic neuropathies) despite the absence of documented deficiency of these cofactors in primary mitochondrial diseases.

Vitamin C (4g daily) and menadiol diphosphate (40mg daily) were used as electron acceptors in patients with severe exercise intolerance and mitochondrial myopathy related to complex III deficiency to facilitate electron transfer from complexes I and II to complex IV.41 One patient had dramatic improvement initially on 31P MRS of muscle, but this effect was not sustained and was not seen in other patients with complex III deficiency.41,42

Because oxidative stress in mitochondrial disorders causes release of free radicals and can lead to apoptosis, free radical scavengers including CoQ10 (400mg daily), idebenone (up to 75mg/kg daily), alpha-lipoic acid (600mg daily), and vitamin E (400 IU daily), are often used in the treatment of mitochondrial disease.43,44 The combination of creatine (3g bid), CoQ10 (120mg bid), and alpha-lipoic acid (300mg bid) was shown to reduce serum lactate and markers of oxidative stress in patients with mitochondrial cytopathies in one randomized double-blind controlled trial, probably through a free-radical-scavenging mechanism.45

Idebenone, a short-chain benzoquinone structurally related to CoQ10, readily enters the brain and localizes to the mitochondria. It both stimulates net ATP formation and acts as a potent free radical scavenger protecting the mitochondrial membrane against lipid peroxidation. Compared to other analogs of coenzyme Q, idebenone is particularly suited for by-passing the functional impairment of mitochondrial complex I. Idebenone has been successfully used in Friedreich ataxia to improve both cardiac and neurological symptoms, especially at high doses.45,46 Neutropenia may be a rare side-effect of idebenone.

DCA, which reduces lactate levels by inhibiting pyruvate dehydrogenase, was recently studied in patients with MELAS in a randomized, placebo-controlled trial.47 This trial was terminated prematurely, however, because of an excessively high incidence of peripheral nerve toxicity, overshadowing any potential benefit in MELAS.

Allogenic stem cell transplantation has shown initial success in two MNGIE patients in partially replacing the deficient enzyme, thymidine phosphorylase, although further clinical followup is necessary.48 L-arginine has been shown in a prospective, unblinded, and unrandomized trial of 24 MELAS patients to reduce the frequency and severity of stroke-like episodes.49

DISEASE-SPECIFIC TREATMENT OF LHON

In light of the possibility for spontaneous recovery in some patients with LHON, any anecdotal reports of treatment efficacy must be considered with caution. The older literature includes attempts to treat or prevent the acute phase of visual loss with systemic steroids, hydroxycobalamin,50 and cyanide antagonists, none of which have proved effective.51-55 In the 1960s, reports from Japan advocated craniotomy with lysis of chiasmal arachnoid adhesions in patients with LHON, with 80% of more than 120 patients reporting visual improvement.56,57 Although the data are impressive, no further reports have followed, and it is difficult to support a surgical therapy logistically removed from the site of ocular neurovascular changes and of presumed primary involvement (the retinal ganglion cells). Optic nerve sheath decompression after progressive visual loss in two LHON patients resulted in no improvement.58,59

Mashima and colleagues60 reported the case of a 10 year old boy homoplasmic for the 11778 mutation who had early improvement in both eyes after 1 year of oral therapy with idebenone, but such an early age of onset certainly could have predisposed this child to spontaneous recovery. Other single case reports have also raised the possibility of a beneficial effect of idebenone on visual and neurologic recovery.61,62 In 2000, Mashima and colleagues63 reported on 28 LHON patients, 14 of whom were treated with idebenone combined with vitamin B2 and vitamin C. There was no significant difference in the number of eyes with visual recovery, although the authors claimed that the treatment seemed to speed recovery when it occurred. Huang and colleagues64 described a 21 year old man with visual loss from the 11778 mutation for 8 months who had substantial improvement of his vision within 4 months of starting CoQ10. Barnils and colleagues65 found no beneficial effects of large doses of idebenone and vitamin C and riboflavin in the prevention of second eye involvement in two LHON patients harboring the 11778 mutation.

Minocycline has been shown to have protective effects in various models of neurodegenerative disorders such as Parkinson’s disease, Huntington’s disease, spinal cord injury and amyotrophic lateral sclerosis.66 In one in vitro study, minocycline had a significant protective effect on the survival of LHON cybrid cells, presumably through anti-oxidant-mediated and megapore-inhibitor anti-apoptotic effects.66 No human LHON studies have been performed with minocycline to date, and this drug’s lack of efficient uptake into the central nervous system and its likely narrow range of useful concentration in the retinal ganglion cells may limit its usefulness in LHON. Cyclosporin A has also been shown to be protective in cell culture analysis of oxidative stress due to the 11778 LHON mutation and of complex I toxin-induced apoptosis in neurons,67,68 as has exogenous glutathione.69

Brimonidine purite is an α-2 agonist used in the treatment of glaucoma which has been shown to have stabilizing effects on retinal ganglion cell survival in animal and human optic neuropathies, presumably partly through the promotion of antiapoptotic cell signals. Because brimonidine’s antiapoptotic properties likely occur through complex I, it seemed an obvious choice of agent to test in LHON, since the three primary mtDNA LHON mutations are located in protein coding genes of complex
I. Brimonidine’s efficacy as a prophylactic agent for second eye visual loss in LHON was evaluated in an open–labeled, non–randomized, multicenter study of nine patients with acute vision loss in one eye from LHON.7 Despite the use of the drug, all patients had deterioration of visual acuity, and seven of eight patients followed for longer than two months had visual acuity in the second eye of 20/200 or worse at the end of the study.

Despite the treatment failure of the brimonidine study, LHON offers a unique “laboratory” for the investigation of new interventions in mitochondrial disease. Since LHON vision loss often occurs in a bilateral sequential fashion, a window of opportunity exists for possible therapeutic intervention after vision loss in the first eye but before second eye involvement.7 LHON has the additional desired property that drugs, adenovirus gene vectors, and other agents may be easily and directly delivered to the tissue at risk, the RGCs and optic nerve, by vitreous injection (see below). Although LHON alone presents this opportunity for experimentation, intervention studies in this “laboratory” have enormous potential for generalization to other mitochondrial diseases, and perhaps to apoptosis–mediated diseases as a whole, including the acquired optic neuropathies.4

Because of the encouraging results of the Friedrich ataxia idebenone study46, centers in Europe and Canada are investigating the use of idebenone at high doses in the treatment of LHON. Unfortunately, the original plan to enroll patients in the acute phase of LHON soon after first eye involvement proved challenging secondary to poor recruitment. However, these investigators have just completed recruitment in a study of idebenone at high doses (900 mg/day) vs. placebo in the treatment of LHON patients (older than 13 and younger than 65) with visual loss for up to 5 years (Patrick Chinnery, personal communication). Eighty–four affected LHON patients with primary mtDNA mutations were included in the study and recruitment is now complete. Although treatment efficacy results are not available at the time of this writing, no serious adverse effects have been reported.

DISEASE-SPECIFIC TREATMENT OF DOA

There are no reports of treatment of DOA patients of which I am aware and no ongoing clinical trials of any agent.

Gene therapy

Gene therapy shows significant promise in the treatment of mitochondrial diseases. Many ingenious strategies have been devised using transfected nuclear and mitochondrial genes to reduce the overall proportion of heteroplasmic mutant mtDNA in vitro, in yeast models, and in animal models — a strategy called “gene shifting”.32 Although it is possible to introduce DNA into the cell nucleus using a variety of vectors, the techniques required to introduce genes directly into mitochondria have yet to be developed.70 Directly targeted repair or replacement of mutated mitochondrial genes is therefore not currently possible. However, “allotopic rescue” is one means of circumventing this barrier.32,71,72 With allotopic rescue, the nuclear genome is transfected by a genetically engineered adenovirus–associated virus (AAV) or other vector, to express a protein usually expressed by the mitochondrial genome. The transfected gene is engineered to attach a mitochondrial targeting polypeptide to the end of the transcribed protein, ensuring the nuclear protein is transported into the mitochondria. The nuclear protein, once in the mitochondria, may replace or complement a protein expressed by mutated mtDNA. This technique of allotopic rescue has been used to replace a mutated ND4 protein in a cybrid cell line homoplasmic for the 11778 LHON mutation, with consequent improvement in biochemical function and ATP synthesis.71,73

Two independent groups have now demonstrated the proof–of–principle that allotopic expression can be effective treatment for LHON, one in a mouse model74,75 and one in a rat model.76 In both studies, an animal model of LHON–like optic neuropathy was induced by intravitreal injection of the human ND4 gene harboring the LHON 11778 mutation. Subsequent intravitreal injection of the wild–type ND4 prevented both retinal ganglion cell loss and impairment of visual function.76

Another gene therapy strategy involves the in vitro transfection of homoplasmic 11778 LHON cells with an AAV vector containing the human mitochondrial superoxide dismutase (SOD2) gene.77 Superoxide dismutase, an antioxidant, is encoded by the nuclear SOD2 gene and detoxifies free radical species within the mitochondrial matrix, thereby acting as an anti–apoptotic agent. Although the SOD2 gene is expressed in LHON cells, superoxide dismutase activity is attenuated in cells homoplasmic for the LHON mutation.78 When LHON cells were transfected with the SOD2–AAV vector, superoxide dismutase was overexpressed, and three–day survival was increased by 89% in transfected LHON cells compared to non–transfected controls.77 This strategy of bolstering antioxidant mechanisms to prolong cell survival was also observed to protect against optic neuropathy in complex I–deficient mice, animals with similar histopathology to human LHON patients.79 These ground–breaking studies clearly open the door to future human clinical studies on patients with LHON.

In heteroplasmic mtDNA diseases, selective destruction of mutant mtDNA with a mutation–specific restriction endonuclease shifts heteroplasmly toward the wild–type state, allowing repopulation of mitochondria with wild–type mtDNA. This strategy has been shown to be effective both in vitro65 and in a murine model of the typically heteroplasmic mtDNA disorder known as neurogenic muscle weakness, ataxia and retinitis pigmentosa (NARP) which results from a point mutation at mtDNA position 8993.81 Unfortunately, LHON is only a heteroplasmic disease in a minority of pedigrees (probably less than 15%)5,82 and the risk of visual loss is reduced among those carriers who are heteroplasmic, making intervention in this manner probably unnecessary.
One form of gene therapy “treatment” for children of mothers with known mtDNA mutations would be the in vitro replacement of the entire mitochondrial genome of an oocyte which could then be fertilized in vitro and implanted for normal embryo development. This technique has been successfully demonstrated in primates, in which the nuclear contents from the mother’s egg is transferred by a technique known as “spindle replacement” to an enucleated, mitochondrial-replete donor cytoplast.81

Although gene therapy holds significant promise in human mitochondrial disease, its clinical use currently faces several challenges.32 Appropriate transfection vectors must be selected, and their delivery to affected tissues must be optimized. The duration of gene therapy effect must be improved, as current transfection methods have not resulted in prolonged and autonomous maintenance of transfected genetic material.84 Finally, patient safety from immunological and oncological side effects and from mtDNA depletion must be guaranteed,32,81 and efficacy must be shown in appropriate animal models before human trials can begin.

Plans for allotropic rescue studies in non–human primates are underway (John Guy, personal communication). In the meantime, identification of patients and carriers with the 11778 LHON mtDNA mutation are ongoing at the University of Miami in order to document feasibility for a clinical gene therapy trial and to develop standardized trial outcome measures (see http://www.bpei.med.miami.edu/site/disease/disease_neuro_LHON.asp#LHON).

Genetic counseling
One crucial aspect of the management of patients with hereditary optic neuropathies is genetic counseling, and knowledge of the basic principles of both nuclear and mitochondrial genetics is essential.

A patient with a DOA mutation has a 50% probability of transmitting the pathogenic allele to each of his or her children. Children with the mutant allele then have a 66–88% chance of developing DOA, in keeping with the known penetrance of the disease,20,85 although penetrance may be nearly complete when the parent manifests DOA themselves.23

Men with the LHON mtDNA mutations should be uniformly reassured that they have no chance of transmitting their mtDNA mutation to their children.86 Women with mtDNA mutations, on the other hand, always have a risk of transmitting mitochondrial disease to their children, and those with the LHON mtDNA mutations are no exception. If the mother is homoplasmic for the LHON mtDNA mutation, then all offspring will be homoplasmic as well. Replicative segregation during embryogenesis complicates matters further, as mutation loads may become magnified or diminished in various tissues of the developing fetus in an unpredictable way.32,86 Even asymptomatic heteroplasmic mothers with very low mutation loads in blood may have children with severe disease from very high mutation loads.89 The risk of transmission of disease with heteroplasmic mtDNA point mutations is therefore impossible to predict accurately. Prenatal testing with amniocentesis or chorionic villus sampling is confounded by heteroplasy as well: amniocytes and chorionic villi may have mutation loads different from other fetal tissues and are unlikely to reflect the child’s ultimate phenotypic outcome, as large shifts in the proportion of mutant mtDNA may occur in developing tissue in utero or after birth as a result of replicative segregation.32

The degree of risk for expression depends on several factors, including the presence or absence of heteroplasmy. The mutation load measured in a woman’s blood cells does not necessarily reflect the mutation load in her other cells, such as oocytes.89 Zygotes may therefore begin embryogenesis with a mutation load quite different from the total mutation load in the mother.

REFERENCES


EXPERT OPINION & COMMENTARY

Natural History of Visual Loss in LHON and DOA

LHON

• 11778
• 3460
• 14484

• Mixed mtDNA mutation and environment
  - Smoking
  - Ethanol
  - Antibiotics
• Men vs. Women – Menopause

DOA

• Classical
• Subclinical
• Plus age

Treatments for Mitochondrial Diseases

1. Vitamins
2. Electron acceptors
3. Free radical scavengers

Clinical Trials

1. Brimonidine
2. Idebenone

On the horizon

1. Gene therapy
2. Drug delivery trickery

CME ANSWERS

1. False
2. False
3. False
4. False
5. False