

MITOCHONDRIAL GENETIC DISEASES WITH OPHTHALMOLOGICAL MANIFESTATIONS: FROM MOLECULAR PATHOPHYSIOLOGY TO CLINICAL MANAGEMENT

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ABSTRACT

Mitochondrial genetic disorders are the most common group of inherited metabolic diseases, and the eye – a tissue of exceptionally high, heterogeneous energy demand – is affected in more than half of patients, often as the presenting feature. This review synthesises current understanding of the molecular pathophysiology, clinical spectrum, diagnosis, and therapeutic landscape of mitochondrial disease with ophthalmic involvement. We outline how the unique biology of mitochondrial DNA (mtDNA) – maternal inheritance, high mutability, heteroplasmy, and the threshold effect – together with the tissue-specific bioenergetic dependence of photoreceptors, retinal pigment epithelium, and retinal ganglion cells, explains the phenotypic heterogeneity of these conditions. Leber hereditary optic neuropathy is discussed as a paradigm of primary mtDNA complex I disease, alongside large-scale deletion syndromes (chronic progressive external ophthalmoplegia, Kearns-Sayre syndrome, Pearson syndrome), multisystem syndromes with secondary ophthalmic involvement (MELAS, MERRF), and nuclear-encoded disorders such as POLG-related disease and autosomal dominant optic atrophy. We review the diagnostic pathway, integrating clinical red flags, electrophysiology and imaging with dual-genome and long-read sequencing, and discuss current management – idebenone, supportive care, and genetic counselling around maternal mtDNA transmission. We summarise emerging strategies (allotopic and non-viral gene delivery, mitochondrially targeted nucleases, antioxidants, mitophagy modulation, mitochondrial transplantation, and stem-cell approaches) and their limitations, and discuss unresolved challenges around diagnostic cost, variant interpretation, and reproductive counselling. Earlier, more systematic recognition of mitochondrial ophthalmic disease, supported by expanding genomic and therapeutic tools, is likely to reshape clinical outcomes for this historically underdiagnosed group of conditions.

KEYWORDS: mitochondrial DNA; Leber hereditary optic neuropathy; heteroplasmy; retinal ganglion cell; Kearns-Sayre syndrome; mitochondrial gene therapy

1. INTRODUCTION

Mitochondrial diseases are widely regarded as the most frequent group of inherited neurometabolic disorders, with a combined prevalence of roughly 1 in 4,300-5,000 individuals [1,2]. Because oxidative phosphorylation (OXPHOS) underlies the energy economy of virtually every cell, these disorders are multisystemic; yet few organs illustrate this as clearly as the eye. Ophthalmic involvement – ptosis, external ophthalmoplegia, optic neuropathy, and pigmentary retinopathy – occurs in a substantial proportion of patients and not infrequently is the presenting, or even the sole, manifestation for years before a systemic diagnosis is reached [2,3]. This gives eye specialists a disproportionate role in initiating the diagnostic pathway [4]. For the practising ophthalmologist, the message is simple: unexplained optic atrophy, ophthalmoplegia, or pigmentary retinopathy in a young patient, particularly with a maternal family history, should prompt consideration of a mitochondrial cause rather than being labelled idiopathic.

Two features of mitochondrial genetics distinguish this group from classical Mendelian disorders. First, mtDNA is present in hundreds to thousands of copies per cell, so pathogenic variants coexist with wild-type molecules – heteroplasmy – and clinical expression depends on the mutant fraction exceeding a tissue- and mutation-specific threshold [5,6]. Second, mtDNA is inherited exclusively through the maternal line, with implications for genetic counselling that have no equivalent in autosomal disease [7]. Superimposed on this is extensive locus and allelic heterogeneity: overlapping phenotypes can arise from mutations in the mitochondrial genome itself or in any of well over 300 nuclear genes supporting mitochondrial replication, translation, or dynamics [8,9].

This review brings together the molecular pathophysiology of mitochondrial ophthalmic disease, characterises the principal clinical syndromes, and summarises the diagnostic and therapeutic strategies reshaping the outlook for these historically incurable conditions.

2. Molecular Basis of Mitochondrial Dysfunction

2.1 Bioenergetic demands of the eye

Mitochondria generate most cellular ATP through OXPHOS, a five-complex electron transport chain in the inner mitochondrial membrane, and impaired assembly or function of any complex is the final common lesion in mitochondrial disease [10]. The eye is among the most energy-demanding organs in the body, but this demand is distributed unevenly across ocular tissues, which largely explains why particular mutations produce particular ophthalmic phenotypes. Photoreceptors maintain an extraordinarily high, light-dependent energy requirement to support phototransduction, outer-segment renewal, and the "dark current," and their inner segments contain a dense population of mitochondria [11,12]. They were long assumed to rely almost exclusively on aerobic glycolysis, a Warburg-like effect first described in retina in the 1920s; more recent metabolic-flux work indicates instead that intact photoreceptors, supported by a metabolically coupled retinal pigment epithelium (RPE), draw substantially on OXPHOS and fatty-acid β -oxidation, with lactate shuttled to the RPE and Müller glia for further oxidation in a retinal analogue of the Cori cycle [13,12]. Retinal ganglion cells (RGCs), whose unmyelinated intraretinal axons run over long, energy-costly projections through the optic nerve, are similarly OXPHOS-dependent, particularly at the lamina cribrosa where myelination begins [14]. The RPE is not merely a supporting layer: it is densely mitochondrial and performs the daily phagocytosis and lysosomal digestion of shed photoreceptor outer-segment discs, an energetically demanding task that itself depends on RPE mitochondrial ATP output. When photoreceptor fatty-acid oxidation is impaired, unmetabolised lipids accumulate and are shed onto the RPE, where defective mitochondrial processing is thought to contribute to toxic lipid by-products implicated in retinal degeneration [12]. The lens, by contrast, is avascular, relies predominantly on anaerobic glycolysis, and is correspondingly resistant to primary OXPHOS failure [3]. This architecture explains why the optic nerve and retina are disproportionately affected in mitochondrial disease: even a modest reduction in ATP synthesis efficiency can push chronically high-demand, low-reserve RGCs past a critical bioenergetic threshold, triggering apoptosis and axonal degeneration [14].

2.2 Unique features of mitochondrial DNA

Human mtDNA is a 16.6-kilobase, circular, double-stranded molecule encoding 13 OXPHOS subunits, 22 transfer RNAs, and 2 ribosomal RNAs, all essential for mitochondrial protein synthesis [4]. Unlike nuclear DNA, mtDNA lacks a protective histone coat, relies on a limited repair repertoire, and lies close to the electron transport chain, the main cellular source of reactive oxygen species (ROS) [15,16]. These features are thought to explain why the mtDNA mutation rate is roughly 10- to 100-fold higher than that of the nuclear genome [17,16]. Because mitochondria, and the mtDNA within them, are inherited essentially only from the oocyte, all pathogenic mtDNA mutations pass along the maternal lineage [7].

2.3 Heteroplasmy and the threshold effect

Each cell contains hundreds to thousands of mtDNA copies across many mitochondria, so a pathogenic variant typically coexists with wild-type genomes within the same cell – heteroplasmy, as opposed to homoplasmy, where essentially all copies carry the variant [5]. Cells and tissues tolerate substantial heteroplasmic loads without overt consequence because of reserve OXPHOS capacity; only once the mutant fraction exceeds a mutation- and tissue-specific proportion does clinical disease become apparent – the threshold effect [6,18]. This threshold is not fixed: point mutations affecting mitochondrial tRNA genes, such as m.3243A>G, typically require very high heteroplasmy (80-90%) before overt OXPHOS impairment emerges, whereas large-scale deletions become clinically manifest at lower mutant loads, often 60-70% [6,19] (Figure 1). Because heteroplasmy levels differ between tissues, and even between cells of the same tissue, the threshold effect explains much of the intra- and inter-familial phenotypic variability that is a hallmark of mitochondrial disease [5,7]. This has a direct clinical corollary: because large-scale deletions reach their threshold at a lower mutant load, KSS and Pearson syndrome tend to declare themselves earlier and more severely than point-mutation diseases such as LHON, and different organs cross their own threshold at different ages as heteroplasmy drifts with cell division.

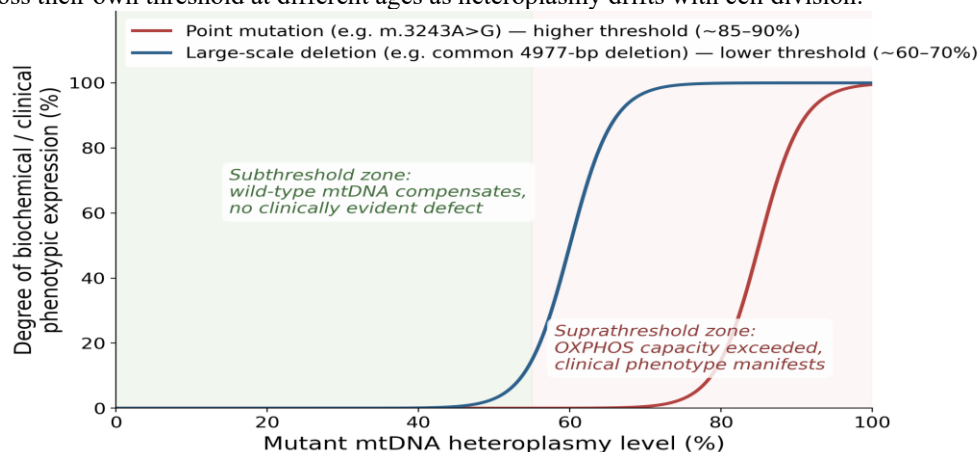


Figure 1. The heteroplasmy threshold effect. Clinical and biochemical expression of a pathogenic mtDNA variant emerges only once the mutant heteroplasmy fraction exceeds a mutation- and tissue-specific threshold.

Large-scale deletions typically manifest at lower mutant loads than pathogenic point mutations affecting mitochondrial tRNA genes.

3. Spectrum of Ophthalmic Manifestations of Mitochondrial Disease

3.1 Optic nerve disease: Leber hereditary optic neuropathy as a paradigm

Leber hereditary optic neuropathy (LHON) is the most frequently encountered mitochondrial optic neuropathy and the classical paradigm of a primary, mtDNA point-mutation disease affecting the eye [20]. It typically presents as painless, subacute central visual loss in one eye, with the fellow eye becoming involved within weeks to months [4]. Roughly 90-95% of confirmed cases are caused by one of three primary point mutations in complex I genes: m.11778G>A in MT-ND4 (about 70% of cases, poorest prognosis for spontaneous recovery), m.14484T>C in MT-ND6 (best chance of recovery, roughly one-third to two-thirds of eyes), and m.3460G>A in MT-ND1 [21,22,23]. These mutations impair the assembly, stability, or catalytic efficiency of complex I, reducing electron flux and ATP output while increasing mitochondrial ROS production [22,24]. In retinal ganglion cells, particularly the small-calibre, unmyelinated axons of the papillomacular bundle, this combination of energy failure and oxidative stress triggers apoptotic cell death and produces the characteristic dense central or cecentral scotoma [14,22].

LHON also departs from a simple monogenic pattern. The primary mutations are usually homoplasmic, yet penetrance is markedly incomplete and skewed by sex, at roughly 50% in male and 10% in female carriers, with 80% or more of affected individuals being male [25,23]. Neither figure is explained by the mtDNA mutation alone; both have prompted a search for nuclear modifiers, including candidate X-linked loci at Xp21 and Xq25-27 [23,26,25], alongside environmental triggers such as tobacco and alcohol and a modulating effect of mitochondrial haplogroup background [27].

3.2 Syndromes with external ophthalmoplegia

A second major clinical axis is progressive impairment of extraocular muscle motility, typically with ptosis. Chronic progressive external ophthalmoplegia (CPEO) can result from primary mtDNA rearrangements (single large-scale deletions or, less commonly, point mutations) or from mutations in nuclear genes maintaining mtDNA replication and repair, most importantly POLG, together with TWNK, RRM2B, POLG2 and TK2 [28,29]. Nuclear POLG mutations alone account for 13-45% of patients presenting with CPEO and multiple mtDNA deletions [30], so a CPEO phenotype does not by itself indicate a primary mtDNA disorder.

Kearns-Sayre syndrome (KSS) is a more severe, syndromic variant, defined by an obligate triad of onset before age 20, progressive external ophthalmoplegia, and pigmentary retinopathy, plus at least one of cardiac conduction block, elevated CSF protein, or cerebellar ataxia [31,32]. The retinopathy classically produces a "salt-and-pepper" fundus, distinct from typical retinitis pigmentosa, and visual acuity is usually preserved unless secondary optic atrophy or severe macular involvement supervenes [33]. KSS is most often caused by a single large-scale mtDNA deletion of roughly 1.1 to 10 kb, with a 4,977-bp "common deletion" identified in a substantial proportion of cases [34,35]. Because these deletions sit at variable heteroplasmic loads across tissues, cardiac, endocrine, and neurological involvement frequently coexist and evolve over time [31,34].

A related but genetically distinct group of "CPEO-plus" phenotypes arises from nuclear genes controlling mitochondrial dynamics, of which autosomal dominant optic atrophy (ADOA) is the most important ophthalmic example. ADOA is caused predominantly by mutations in OPA1, a dynamin-related GTPase governing inner-membrane fusion and cristae architecture that also restrains apoptosis [36,37]. Depending on their location within the GTPase, middle, or effector domains, OPA1 mutations act through haploinsufficiency or a dominant-negative mechanism, producing distinguishable effects that plausibly contribute to variable severity, from isolated optic atrophy to syndromic "ADOA-plus" with deafness and neuropathy [38,37]. Unlike LHON, OXPHOS capacity itself is not consistently impaired in ADOA fibroblasts, pointing to a mechanism rooted in mitochondrial network architecture rather than direct bioenergetic failure [39]. This carries therapeutic weight: LHON is approached mainly through strategies that restore electron flow or buffer oxidative stress, whereas ADOA seems better suited to fusion-promoting or mitophagy-based interventions, since antioxidants alone would not address a defect in network architecture.

3.3 Rare and overlapping mitochondrial syndromes

Pearson syndrome is the infantile counterpart of large-scale mtDNA deletion disease, presenting in the first year of life with transfusion-dependent sideroblastic anaemia, neutropenia, thrombocytopenia, and exocrine pancreatic insufficiency, with substantial infant mortality [40,41]. Children who survive often go on to develop the clinical triad of KSS later in childhood, reflecting a tissue-dependent redistribution of the same deletion over time [42].

MELAS (mitochondrial encephalomyopathy, lactic acidosis, and stroke-like episodes), most commonly caused by the m.3243A>G mutation in MT-TL1, has a broad ophthalmic spectrum despite not being primarily an eye disease [43]. Pigmentary retinopathy, a fine granular macular pattern distinct from the KSS "salt-and-pepper" fundus, is reported in 38-86% of carriers and can serve as a valuable diagnostic clue [44,45]. Patients may also develop progressive external ophthalmoplegia, ptosis, posterior subcapsular cataract, optic atrophy, and, during stroke-like episodes, cortical visual field defects or blindness from occipital lobe involvement [46,47].

MERRF (myoclonic epilepsy with ragged-red fibres), caused in roughly 80% of cases by m.8344A>G in MT-TK, less commonly includes optic atrophy and ptosis among its features, which more typically comprise myoclonus, epilepsy, ataxia, and hearing loss [48,49]. Secondary mitochondrial dysfunction has also been implicated in

Alzheimer and Parkinson disease; OCT measurement of retinal nerve fibre layer and ganglion cell layer thinning is being explored as a non-invasive surrogate marker of central neurodegeneration, potentially detectable before cognitive symptoms appear, though the evidence remains mixed [50]. A symptom-first differential diagnosis, linking each presenting ophthalmic sign to candidate syndromes and their underlying genetic defects, is presented in Figure 2. Table 1 provides a structured overview of the major mitochondrial genetic syndromes with ophthalmic involvement, including their genetic basis, inheritance, key ocular features, and typical age of onset.

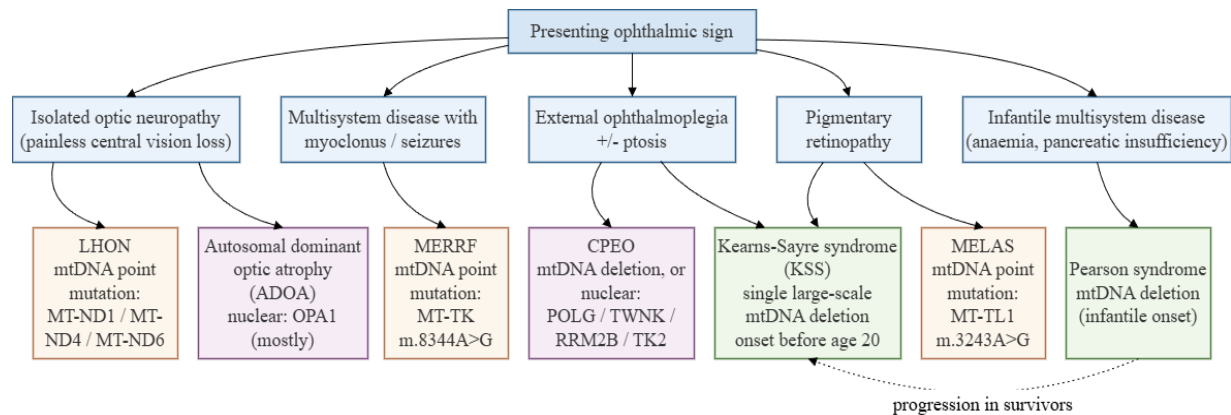


Figure 2. Symptom-first differential diagnosis of mitochondrial genetic disease with ophthalmic involvement: presenting ophthalmic sign, candidate syndromes, and the underlying mtDNA or nuclear gene defect for each.

Table 1. Major mitochondrial genetic syndromes with ophthalmic involvement.

Syndrome	Genetic basis	Inheritance	Key ophthalmic features	Typical onset
LHON	mtDNA point mutations: m.11778G>A (MT-ND4), m.14484T>C (MT-ND6), m.3460G>A (MT-ND1)	Maternal; reduced, male-skewed penetrance	Subacute, painless, sequential bilateral central vision loss; optic atrophy	15-35 y
CPEO	mtDNA single deletions/point mutations, or nuclear POLG, TWNK, RRM2B, TK2	Maternal (mtDNA) or autosomal (nuclear)	Progressive ptosis and external ophthalmoplegia, usually without retinopathy	Adulthood
Kearns-Sayre syndrome	Single large-scale mtDNA deletion (~1.1-10 kb; common 4977-bp deletion)	Sporadic	Ptosis, ophthalmoplegia, “salt-and-pepper” pigmentary retinopathy	<20 y
Pearson syndrome	Same mtDNA deletion spectrum as KSS	Sporadic	Systemic (sideroblastic anaemia, pancreatic insufficiency); ophthalmic KSS triad emerges in survivors	Infancy
MELAS	mtDNA MT-TL1 m.3243A>G	Maternal	Pigmentary retinopathy, ophthalmoplegia, posterior subcapsular cataract, cortical visual loss in stroke-like episodes	Childhood-adulthood
MERRF	mtDNA MT-TK m.8344A>G	Maternal	Optic atrophy, ptosis/ophthalmoparesis (less common features)	Childhood-adulthood
Autosomal dominant optic atrophy	Nuclear OPA1 (most common)	Autosomal dominant	Progressive bilateral optic atrophy, temporal disc pallor; “ADOA-plus” with deafness/neuropathy	Childhood

4. Diagnosis: From Clinical Suspicion to Molecular Confirmation

4.1 Clinical approach

Diagnosing mitochondrial ophthalmic disease requires a systematic, multisystem approach rather than reliance on a single pathognomonic sign. A detailed family history focused on maternal-line transmission, early onset, and a progressive course should raise suspicion, particularly alongside a targeted ophthalmic examination (visual acuity, colour vision, visual fields, fundus examination, and OCT of the retinal nerve fibre layer and ganglion cell complex) and systemic evaluation for cardiac, endocrine, and neurological involvement [4,2]. Visual

electrophysiology gives objective evidence of retinal ganglion cell or outer retinal dysfunction and can detect abnormalities before they are clinically apparent, helping to distinguish optic neuropathy from primary retinal disease and to characterise mixed phenotypes such as maternally inherited diabetes and deafness (MIDD), KSS, and NARP [51,52]. Ptosis, ophthalmoplegia, and pigmentary retinopathy together in a young patient remain one of the most reliable "red flags" for KSS specifically [31].

4.2 Genetic testing

Establishing a precise molecular diagnosis is critical for prognostication, family counselling, and access to mutation-specific therapies [8]. Because pathogenic variants may reside in either genome, a comprehensive strategy increasingly relies on simultaneous NGS of the whole mitochondrial genome together with a nuclear gene panel or exome, rather than sequential single-gene testing [8,53]. Dual-genome NGS panels applied to large cohorts report diagnostic yields around 14-15%, split roughly evenly between nuclear and mitochondrial contributions [53]. Standard short-read NGS struggles to resolve large-scale mtDNA deletions precisely, because short reads cannot easily span deletion breakpoints or reliably quantify their heteroplasmy. Third-generation long-read platforms (Oxford Nanopore, PacBio) sequence the entire mitochondrial genome in single, amplification-free reads and can map breakpoints and quantify heteroplasmy in one assay, emerging as a promising complement to the workflow many laboratories rely on for suspected KSS or CPEO [54,55]. Even so, adult patients commonly endure a prolonged "diagnostic odyssey" before a molecular answer is reached [1,56], compounded by the fact that pathogenic mtDNA variants, particularly large-scale deletions, may be present at low or undetectable heteroplasmy in blood, sometimes necessitating a more invasive tissue such as skeletal muscle [57]. Interpretation is further complicated by allelic and locus heterogeneity, pleiotropy, and the challenge of classifying variants of uncertain significance against a background of variable heteroplasmy [8,9]. The full diagnostic pathway, from initial clinical suspicion through electrophysiology and dual-genome sequencing to long-read sequencing for unresolved rearrangements, is illustrated in Figure 3. Table 2 details the individual diagnostic modalities, what each assesses, and its clinical utility in the workup of suspected mitochondrial ophthalmic disease.

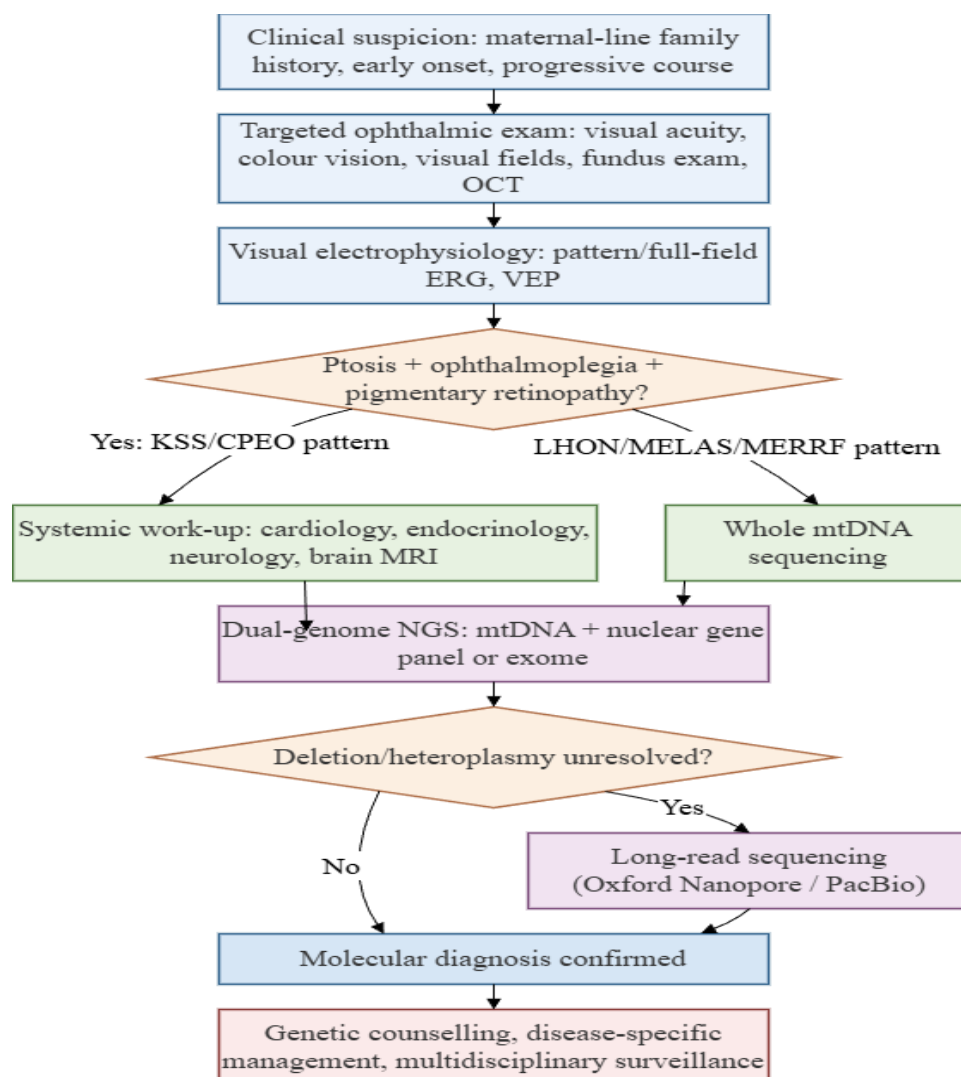


Figure 3. Diagnostic pathway for suspected mitochondrial ophthalmic disease, from clinical suspicion through electrophysiology and dual-genome sequencing to long-read sequencing for unresolved large-scale rearrangements.

Table 2. Diagnostic modalities in mitochondrial ophthalmic disease.

Diagnostic modality	What it assesses	Clinical utility
Clinical examination & family history	Maternal transmission pattern, age of onset, systemic involvement	Initial triage; identifies “red-flag” combinations (e.g. ptosis + ophthalmoplegia + retinopathy suggests KSS)
Visual acuity, colour vision, visual fields	Central/cecocentral visual function	Detects and monitors optic neuropathy over time
OCT (RNFL and macular ganglion cell layer)	Structural retinal ganglion cell and nerve fibre layer loss	Objective, quantifiable; tracks structural progression
Pattern/full-field ERG and VEP	Retinal ganglion cell and outer retinal function; optic nerve conduction	Distinguishes optic neuropathy from primary retinal disease; can precede symptoms
Whole mtDNA sequencing	Point mutations, heteroplasmy level, large-scale deletions	Confirms mtDNA-based diagnoses (LHON, MELAS, MERRF, KSS deletions)
Long-read sequencing (Oxford Nanopore / PacBio)	Precise breakpoints and heteroplasmy of large-scale mtDNA deletions/duplications in a single amplification-free read	Resolves structural rearrangements (e.g. KSS/CPEO deletions) that short-read NGS and standard ddPCR panels can miss
Nuclear gene panel / exome (dual-genome NGS)	POLG, OPA1, TWNK, RRM2B, TK2, KIF5A and related genes	Identifies nuclear-encoded causes; substantially increases overall diagnostic yield
Muscle biopsy (histochemistry, deletion analysis)	Ragged-red fibres, COX deficiency, tissue-specific heteroplasmy	Useful when blood mtDNA testing is uninformative, especially in KSS/CPEO
Systemic work-up (cardiology, endocrinology, neurology, MRI)	Multisystem involvement	Supports syndromic diagnosis and guides ongoing surveillance

5. Therapeutic Approaches and Emerging Directions

5.1 Current therapeutic strategies

Idebenone, a short-chain synthetic analogue of coenzyme Q10, remains the only medication with regulatory approval specifically for LHON in several jurisdictions [23,58]. It acts as an electron shuttle that bypasses a dysfunctional complex I by donating electrons more directly to complex III, partially restoring ATP synthesis while also acting as an antioxidant; it does not correct the underlying genetic defect [58,59]. Efficacy was first evaluated in the randomised, placebo-controlled RHODOS trial (85 patients, idebenone 900 mg/day versus placebo for 24 weeks); the primary endpoint did not reach significance in the intention-to-treat population, but secondary visual-acuity endpoints favoured idebenone, particularly when treatment began within the first year of onset [60]. The subsequent LEROS study and an expanded-access programme extended these findings to the chronic phase, reporting recovery rates comparable to, or exceeding, those in RHODOS and reinforcing that benefit depends on how soon treatment starts [59,61]. This benefit is not uniform across genotypes, however: in subacute eyes carrying m.3460G>A, idebenone has been associated with possible clinical worsening rather than benefit, so treatment decisions should account for the specific primary mutation [62]. Other antioxidant and cofactor strategies, including vatiquinone (EPI-743), coenzyme Q10, creatine, and vitamins C and E, are used mainly as adjunctive support in broader mitochondrial disease, with open-label evidence of benefit in Leigh syndrome but limited placebo-controlled confirmation [63,64,65]. Management otherwise remains largely supportive: surgical correction of ptosis, low-vision rehabilitation, diplopia management, and coordinated surveillance of cardiac, endocrine, and neurological complications [31,4].

5.2 Emerging and experimental therapeutic directions

Gene replacement (allotopic expression) therapy has advanced furthest for LHON caused by m.11778G>A. Lenadogene nolparvovec is a recombinant AAV2/2 vector carrying a codon-optimised, nuclear-targeted copy of wild-type MT-ND4, delivered by intravitreal injection so the transgene product is imported into mitochondria and competes with the mutant protein within complex I [66,67]. Across the pivotal phase III trials (REVERSE, RESCUE) and the confirmatory REFLECT trial, unilateral injection produced a sustained improvement in visual acuity that was, unexpectedly, observed bilaterally, including in sham-injected fellow eyes [68,67]. Non-human primate studies indicate this contralateral benefit is associated with detectable transfer of vector DNA to the untreated eye and optic chiasm; a mechanistic study suggested such transfer may be facilitated by connexin-43-mediated astrocyte networks, but this remains one hypothesis among several, and its contribution relative to systemic immune-mediated mechanisms is unresolved [66,69]. Five-year follow-up (RESTORE) has shown persistence of this benefit with an acceptable safety profile [70,71]. AAV-based delivery nonetheless carries an immunological cost that deserves more attention than it typically receives: across the pivotal trials, intraocular inflammation developed in roughly 75% of treated eyes versus about 10% of sham-injected eyes, and pre-existing or treatment-induced neutralising antibodies to AAV2 can preclude re-dosing or treatment of the fellow eye with

the same serotype. Strategies under investigation include engineered, less immunogenic capsids, alternative serotypes, and peri-injection corticosteroid regimens [72].

A conceptually distinct strategy targets heteroplasmy itself rather than replacing the missing gene product. Mitochondrially targeted restriction endonucleases, zinc-finger nucleases (mtZFNs), TALE nucleases (mitoTALENs), and compact single-protein ARCUS nucleases can be engineered to selectively cleave mutant mtDNA; because linearised mtDNA is rapidly degraded, this shifts the heteroplasmic balance toward wild-type upon replication, without exogenous gene delivery [73,18,74]. This "knockdown" approach has been demonstrated in patient-derived cybrid cell lines – cytoplasmic hybrids in which a patient's mitochondria are introduced into a nuclear background cell line depleted of its own mtDNA (ρ^0 cells) – and in animal models targeting LHON- and MELAS-associated mutations, though clinical translation is still constrained by delivery efficiency and mutation-specific reagent design [73,18]. Because AAV delivery is itself immunogenic and cargo-limited, non-viral carriers, including DQAsome nanovesicles, MITO-Porter liposomes, and peptide- or polymer-conjugated constructs, are being explored as less immunogenic alternatives for importing DNA or RNA into mitochondria, including constructs designed to deliver ND1 sequences relevant to LHON, though none has yet reached clinical testing [75,76].

Mitophagy, the selective autophagic clearance of dysfunctional mitochondria principally through the PINK1/Parkin pathway, has emerged as both a pathogenic modifier and a therapeutic target. Impaired mitophagy has been documented in cellular models of LHON, where it compounds the direct consequences of the primary mutation, while pharmacological or genetic enhancement of mitophagy protects retinal ganglion cells in experimental models of optic nerve injury and glaucoma [22,77,78]. The mitochondria-targeted antioxidant SkQ1, in a small phase 2a trial, improved visual outcomes in LHON patients, mediated partly through PINK1/Parkin-dependent mitophagy alongside direct ROS scavenging [79]. Related compounds such as MitoQ have shown safety and physiological benefit in early-phase human trials in other organ systems, though dedicated ophthalmic trials remain limited [80].

Mitochondrial transplantation, the direct delivery of exogenous, respiration-competent mitochondria to diseased tissue, is an early-stage strategy of particular relevance to the eye given its surgical accessibility. In animal models, intravitreally or subretinally injected isolated mitochondria are internalised by retinal ganglion cells, photoreceptors, and the RPE, and improve electroretinographic responses, oxidative metabolism, and RGC survival after optic nerve crush and ischaemic insult [81,82,83]. Interest extends to ischaemic and glaucomatous optic neuropathy and, prospectively, RPE-mitochondrial dysfunction in age-related macular degeneration, though translation requires resolving donor-mitochondria sourcing, immunogenicity, and delivery durability [81,84].

A related, cell-based strategy – delivery of autologous bone-marrow-derived mesenchymal stem cells intended to supply replacement ganglion-like cells and transfer healthy mitochondria – has already been trialled in patients with LHON and OPA1-related ADOA through the open-label, patient-funded Stem Cell Ophthalmology Treatment Study (SCOTS, SCOTS2; NCT01920867, NCT03011541), with visual acuity gains reported in most treated eyes [20]. Independent reviewers have flagged major design limitations, including the absence of a control arm and substantial out-of-pocket cost, and caution that these results are not yet evidence that stem-cell therapy is ready for clinical translation [58,20].

6. Clinical Management, Genetic Counselling, and the Multidisciplinary Team

Early and accurate diagnosis shortens the path to a molecular answer, allows earlier treatment (particularly idebenone, whose benefit is greatest when started soon after onset), and provides the foundation for informed reproductive counselling [60,1]. Genetic counselling in mitochondrial disease departs substantially from Mendelian paradigms because of maternal transmission, heteroplasmy, the threshold effect, and the mitochondrial genetic bottleneck, which together make recurrence-risk prediction considerably less certain than for nuclear mutations [7,85]. Counselling should be non-directive and delivered over time, addressing recurrence risk for future children, the availability of preimplantation genetic testing (PGT) for heteroplasmic variants, and prenatal diagnosis for nuclear-encoded disease, which follows conventional Mendelian risks [85,7]. For women with very high heteroplasmy or homoplasmic variants, for whom PGT may not identify a sufficiently low-heteroplasmy embryo, mitochondrial donation techniques such as pronuclear transfer have recently been reported in a substantial clinical series, with markedly reduced or undetectable mutant load in the resulting children [85]. The technique nonetheless remains subject to substantial ethical and regulatory debate internationally, since it affects the germline; it is permitted only in a small number of jurisdictions under strict oversight. Given the systemic nature of mitochondrial disease, longitudinal management is best delivered through a multidisciplinary team spanning ophthalmology, neurology, cardiology, endocrinology, and clinical genetics, with surveillance tailored to each syndrome, for example periodic cardiac assessment in KSS [31,42].

7. DISCUSSION

This review points to a field in transition, moving from a largely descriptive, phenotype-first discipline toward one organised around precise molecular diagnosis and, increasingly, genotype-specific therapy. Several points merit emphasis. First, the tissue-specific bioenergetic architecture of the eye explains why mitochondrial mutations produce strikingly selective ophthalmic phenotypes despite ostensibly ubiquitous expression of the underlying defect, and deserves greater integration into how clinicians interpret ocular findings. Second, the

maturation of gene therapy for LHON, culminating in regulatory success for lenadogene nolparvovec in the m.11778G>A subgroup, shows that a genetically precise, single-mutation strategy is achievable even in a disease category historically regarded as intractable. The incompletely understood contralateral benefit, and the substantial rate of intraocular inflammation, are reminders that mechanistic understanding and manageable toxicity both continue to lag behind clinical efficacy. Third, allotopic gene replacement, heteroplasmy-shifting nucleases, non-viral delivery, and mitochondrial or stem-cell transplantation represent distinct, potentially complementary therapeutic logics whose relative durability, specificity, and risk profiles remain to be clarified. Fourth, idebenone remains the only approved pharmacotherapy for LHON, but the evidence, including RHODOS, LEROS, and the m.3460G>A caution discussed above, points to a real yet modest and genotype-dependent effect, underscoring early, mutation-specific diagnosis over drug efficacy alone. Fifth, technological capability has outpaced clinical implementation: dual-genome and long-read sequencing exist, yet many adult patients still endure years of specialist visits before a molecular diagnosis, arguing for greater "red flag" recognition in general ophthalmology and neurology training rather than for further sequencing innovation alone. Some authors caution that enthusiasm for gene- and nuclease-based therapies has outpaced the modest scale and short follow-up of the underlying trials, and that the poorly controlled design of early cell-based studies such as SCOTS should temper, rather than encourage, patient expectations. The ethical and regulatory debate around mitochondrial donation reflects a similarly genuine disagreement that lies beyond the scope of a purely clinical assessment.

7.1 Unresolved questions and future paradigms

Three open questions seem worth flagging. First, within a single population of retinal ganglion cells sharing an ostensibly uniform heteroplasmy level, some cells degenerate while immediate neighbours survive; this variability is not fully explained by measured mtDNA mutant load and may instead reflect local differences in glial support, vascular supply, or mitochondrial biogenesis capacity that current assays do not capture. Second, no single intervention is likely to be sufficient on its own: combining a genotype-specific approach, such as gene replacement or a heteroplasmy-shifting nuclease, with an agent that supports mitochondrial quality control, such as a mitophagy inducer, may prove more durable than either strategy alone, though this has not yet been tested prospectively. Third, mitochondrial donation raises an ethical concern beyond the immediate reproductive decision: because the technique alters the germline, the long-term health of children conceived this way, and of their own descendants, remains unknown, and that uncertainty is likely to constrain wider adoption regardless of near-term technical success.

CONCLUSION

Mitochondrial genetic disease is an important, probably underrecognised cause of ophthalmic pathology, disproportionately affecting the energy-dependent structures of the eye – the optic nerve, retina, and extraocular muscles. Understanding the distinctive genetic features of mtDNA together with the tissue-specific energetic dependence of ocular structures is essential for accurate diagnosis, prognostication, and genetic counselling. Early diagnosis, built on a systematic clinical approach and confirmed by dual-genome and long-read testing, enables timely management and an accurate prognosis, while an expanding therapeutic toolkit offers genuine, if early-stage, promise for conditions historically lacking disease-modifying treatment. Considerable challenges persist: the cost of comprehensive genetic testing, the complexity of variant interpretation in a heteroplasmic background, and the ethical dimensions of reproductive counselling and mitochondrial donation. Large prospective studies and standardised, syndrome-specific guidelines are needed to translate these advances into consistently improved care.

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