Mitochondrial Retinopathy

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Purpose: To report the retinal phenotype and the associated genetic and systemic findings in patients with mitochondrial disease.

Design: Retrospective case series.

Participants: Twenty-three patients with retinopathy and mitochondrial disease, including chronic progressive external ophthalmoplegia (CPEO), maternally inherited diabetes and deafness (MIDD), mitochondrial encephalomyopathy, lactic acidosis, and stroke-like episodes (MELAS), Kearns-Sayre syndrome, neuropathy, ataxia, and retinitis pigmentosa (NARP) syndrome, and other systemic manifestations.

Methods: Review of case notes, retinal imaging, electrophysiologic assessment, molecular genetic testing including protein modeling, and histologic analysis of muscle biopsy.

Main Outcome Measures: Phenotypic characteristics of mitochondrial retinopathy.

Results: Genetic testing identified sporadic large-scale mitochondrial DNA deletions and variants in MT-TL1, MT-ATP6, MT-TK, MT-RNR1, or RRM2B. Based on retinal imaging, 3 phenotypes could be differentiated: type 1 with mild, focal pigmentary abnormalities; type 2 characterized by multifocal white-yellowish subretinal deposits and pigment changes limited to the posterior pole; and type 3 with widespread granular pigment alterations. Advanced type 2 and 3 retinopathy presented with choriotreital atrophy that typically started in the peripapillary and paracentral areas with foveal sparing. Two patients exhibited a different phenotype: 1 revealed an occult retinopathy, and the patient with RRM2B-associated retinopathy showed no foveal sparing, no severe peripapillary involvement, and substantial photoreceptor atrophy before loss of the retinal pigment epithelium. Two patients with type 1 disease showed additional characteristics of mild macular telangiectasia type 2. Patients with type 1 and mild type 2 or 3 disease demonstrated good visual acuity and no symptoms associated with the retinopathy. In contrast, patients with advanced type 2 or 3 disease often reported vision problems in dim light conditions, reduced visual acuity, or both. Short-wavelength autofluorescence usually revealed a distinct pattern, and near-infrared autofluorescence may be severely reduced in type 3 disease. The retinal phenotype was key to suspecting mitochondrial disease in 11 patients, whereas 12 patients were diagnosed before retinal examination.

Conclusions: Different types of mitochondrial retinopathy show characteristic features. Even in absence of visual symptoms, their recognition may facilitate the often challenging and delayed diagnosis of mitochondrial disease, in particular in patients with mild or nebulous multisystem disease. Ophthalmology Retina 2022;6:65-79 © 2021 by the American Academy of Ophthalmology. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

Supplemental material available at www.ophthalmologyretina.org.

Mitochondria are central for cellular function and survival because they mediate processes such as energy production, metabolism control, and apoptosis.1-3 In 1988, initial reports described an association of variants in the mitochondrial genome with monogenic disease, including Leber hereditary optic neuropathy, Kearns-Sayre syndrome, and mitochondrial myopathies.4-6 Mitochondrial disease may involve 1 or several organs with limited genotype—phenotype correlations and symptoms ranging from mild to severe.1,2

Studies on the retinal phenotype associated with a specific mitochondrial variant (m.3243A>G, MT-TL1 gene) indicate that diagnosis of associated syndromes such as mitochondrial encephalomyopathy with lactic acidosis and stroke-like episodes (MELAS) and maternally inherited diabetes and deafness (MIDD) often may be facilitated by identification of the distinct retinal features.7-10 A grading scheme for m.3243A>G-associated retinal manifestations and a comprehensive characterization of associated systemic features illustrated the variable disease severity across organ systems and among patients.7-10 Less detailed information is available about retinal changes in patients with other mitochondrial diseases, and a comprehensive characterization of retinal phenotypes is lacking.
<table>
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<th>Patient No.</th>
<th>Gender</th>
<th>Referral Diagnosis</th>
<th>Mitochondrial Retinopathy</th>
<th>Age (yrs)</th>
<th>BCVA (OD/OOS)</th>
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<th>Electroretinography</th>
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<th>Heteroplasmy Level (Specimen)</th>
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Table 1. (Continued.)

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AG = atrophy gyrate; AMD = age-related macular degeneration; APMPPE = acute posterior multifocal placoid pigment epitheliopathy; BCVA = best-corrected visual acuity; CF = counting fingers; CPEO = chronic progressive external ophthalmoplegia; DR = diabetic retinopathy; E = female; GA = geography atrophy; HM = hand movements; M = male; MD = macular degeneration or dystrophy; MELAS = mitochondrial encephalomyopathy; NARP = neuropathy, ataxia, and retinitis pigmentosa syndrome; NE = not examined; OD = right eye; OS = left eye; RD = retinal dystrophy; RP = retinitis pigmentosa; — = not available.

*Patients 11 and 12 are related (mother and daughter).

1Assumed amblyopia resulting from ocular trauma in childhood.

References: 15, 16, 21, 22, 23, 24, 25, 26.
With upcoming potential treatment options for mitochondrial disease, a detailed understanding of associated retinal neurodegeneration is required for patient identification and as potential parameters for clinical end points. Here, we investigate the phenotypic spectrum of mitochondrial retinopathy and illustrate novel phenotypic features and genotype—phenotype correlations.

**Methods**

This retrospective study of patients with retinal changes and a diagnosis of mitochondrial disease adhered to the tenets of the Declaration of Helsinki. Institutional review board approval (Ethics Committee, Medical Faculty, University of Bonn) and patient informed consent were obtained.

Best-corrected visual acuity (BCVA) and visual impairment was classified according to World Health Organization (WHO) definitions as mild (<20/40), moderate (<20/60), or severe (<6/60). Clinical assessment included standardized anterior segment and dilated fundus examination and, in selected patients, full-field electroretinography. Multimodal retinal imaging included OCT, autofluorescence (AF), and ultra-widefield imaging (Supplemental Methods, available at www.ophthalmologyretina.org).

A comprehensive general medical history was obtained and a neurologic workup was performed. Muscle biopsy samples were obtained from a proximal limb muscle, and cryostat sections were analyzed histologically and histochemically, including with modified Gomori’s trichrome, cytochrome c oxidase (COX), and succinate dehydrogenase (SDH) stainings. Information on genetic testing may be found in the Supplemental Methods.

**Results**

The study included 23 patients (10 women) from 22 families with an age at first presentation between 17 and 74 years (median, 50 years; interquartile range, 36–62 years; Table 1). The retinal phenotype was key to suspecting mitochondrial disease in 11 patients; 12 patients had been diagnosed with mitochondrial disease before retinal examination (Table S1, available at www.ophthalmologyretina.org).

At first examination, 32 eyes (18 patients) had a BCVA of 20/40 or better, BCVA was ≥20/25 in 28 eyes (15 patients). Mild, moderate, or severe reduction of BCVA was present in 7 eyes (6 patients), 5 eyes (4 patients), or 1 eye, respectively. In 1 eye, BCVA was reduced to counting fingers because of ocular trauma in childhood. Patient-reported symptoms included vision problems in dim light conditions (n = 9), reduced visual acuity (n = 9), glare (n = 2), scotoma (n = 1), and metamorphopsia (n = 1). The most frequent combination of symptoms was problems in dim light conditions and reduced visual acuity (n = 7). Twelve patients reported no vision problems indicative of a retinal disease (Table 1).

**Phenotypic Variability of Mitochondrial Retinopathy**

Retinal changes showed considerable variability, but specific recurrent patterns allowed grouping into the following different types.

Type 1 (Fig 1; Fig S1, available at www.ophthalmologyretina.org). Six patients without vision problems relating to retinal disease (26%; mean BCVA, 20/20; mean age, 52 years) revealed mild, focal pigmentary abnormalities on funduscopy associated with increased and decreased AF and alterations at the interface between photoreceptor outer segments and the retinal pigment epithelium.

**Figure 1.** Mitochondrial retinopathy type 1. Fundus autofluorescence imaging showing focally increased and decreased autofluorescence (left column) corresponding to alterations at the interface between photoreceptor outer segments and the retinal pigment epithelium on OCT (right column): (A) patient 1 and (B) patient 4.
(RPE) on OCT scans. On electroretinography testing (n = 3), scotopic and photopic recordings were within normal limits except for 1 individual who showed slightly reduced photopic amplitudes. Some of these patients also showed mild peripheral pigment retinopathy; however, this was not documented systematically.

Type 2 (Fig 2; Fig S2, available at www.ophthalmology retina.org). Seven patients (30%; mean BCVA, 20/32; mean age, 51 years) revealed a distinct retinal phenotype in which fundus changes are more severe than in type 1, but where these remain limited to the posterior fundus, even in late disease stages with atrophy. These patients showed multifocal faint white-yellowish or hyperpigmented subretinal deposits and pigment changes, and AF imaging showed hyperautofluorescent dot or fleck-like lesions. Sharply demarcated chorioretinal atrophy occurred in the peripapillary and paracentral macular area and involved the fovea in only the most advanced cases. Electroretinography recordings (n = 2; BCVA, 20/20) were within normal limits. Four patients reported no visual symptoms, whereas the other 3 showed reduced visual acuity (n = 3), vision problems in dim light conditions (n = 3), and glare (n = 1).

Figure 2. Mitochondrial retinopathy type 2. Fundus autofluorescence imaging (left column) showing hyperautofluorescent dot or fleck-like lesions and, where present, sharply demarcated dark areas representing chorioretinal atrophy. These atrophic areas also show a sharp demarcation from relatively preserved retina on OCT images (right column): (A) patient 8, (B) patient 9, (C) patient 10, and (D) patient 12.
Type 3 (Fig 3; Fig S3, available at www.ophthalmologyretina.org). Eight patients (35%) revealed widespread (extending beyond the vascular arcade) granular pigmented fundus alterations corresponding to a granular AF pattern. Associated changes on OCT included reflectivity changes primarily in the ellipsoid and interdigitation zone and an increased distance between ellipsoid band and the retinal pigment epithelium layer on OCT (right column): (A, B) patients 15 and 17 without macular atrophy and (C, D) patients 18 and 20 with fovea-sparing macular chorioretinal atrophy.

Figure 3. Mitochondrial retinopathy type 3. Fundus autofluorescence imaging (left column) imaging showing peripapillary atrophy and a widespread granular autofluorescent pattern corresponding to reflectivity changes primarily in the ellipsoid and interdigitation zone and sometimes an increased distance between ellipsoid band and the retinal pigment epithelium layer on OCT (right column): (A, B) patients 15 and 17 without macular atrophy and (C, D) patients 18 and 20 with fovea-sparing macular chorioretinal atrophy.

BCVA, 20/32; mean age, 57 years) showed 1 or more bilateral areas of paracentral and peripapillary chorioretinal atrophy that spared the foveal center, and 2 patients (mean BCVA, 20/63; mean age, 65 years) showed more advanced degeneration that included the fovea. The chorioretinal atrophy corresponded to sharply demarcated areas of decreased AF and, on OCT, atrophy of the photoreceptor layer and RPE. On electroretinography testing (n = 8), photopic and scotopic responses were reduced to a similar extent in all except for the youngest patient.
patients in this group, except the one with normal electroretinography recordings, reported vision problems in dim light conditions.

Two patients showed a different retinal phenotype and could not be classified as types 1, 2, or 3. Patient 22 showed no visual symptoms, but electroretinography recordings revealed reduced cone responses (approximately 50% of the lower limit of that of control participants) and rod responses in the lower normal range. Funduscopy and AF imaging showed no obvious changes (occult retinopathy), but a distinct retinal phenotype was found on OCT with thinning of the outer nuclear layer, mainly toward the peripheral scan. C. In RRM2B-associated retinopathy (patient 23), a patchy area of reduced autofluorescence is surrounded by diffuse, partly spot-shaped hyperautofluorescent lesions. D. OCT imaging showing widespread atrophy of the photoreceptor layer, despite partial presence of the retinal pigment epithelium. Additional information on RRM2B-associated retinopathy can be found in Supplemental Figures 4 and 8.

Figure 4. Occult mitochondrial retinopathy and RRM2B-associated retinopathy. A, B, An occult retinopathy, characterized by normal retinal appearance on funduscopy and (A) autofluorescence imaging, but distinct changes on (B) OCT was identified in patient 22. B, OCT scan showing a fading of the ellipsoid and interdigitation zone and thinning of the outer nuclear layer, mainly toward the peripheral scan. C. In RRM2B-associated retinopathy (patient 23), a patchy area of reduced autofluorescence is surrounded by diffuse, partly spot-shaped hyperautofluorescent lesions. D. OCT imaging showing widespread atrophy of the photoreceptor layer, despite partial presence of the retinal pigment epithelium. Additional information on RRM2B-associated retinopathy can be found in Supplemental Figures 4 and 8.

Longitudinal Observations and Additional Findings on Retinal Imaging

Fundus changes showed overall a high symmetry between eyes (Fig 5; Figs S5 and S6, available at www.ophthalmologyretina.org), and longitudinal observations were available for 15 patients. In patients without chorioretinal atrophy at baseline (n = 8), no atrophy developed and visual acuity remained stable (range, 20/25–20/20; median follow-up, 3 years; range, 1–16 years). In patients with pre-existing chorioretinal atrophy (n = 7; median follow-up, 4 years; range, 3–6 years), atrophy progressed remarkably and included an evolution from multiple separate areas of atrophy to larger and eventually confluent areas of atrophy (Fig S7, available at www.ophthalmologyretina.org). Growth of advanced chorioretinal atrophy resulted in foveal involvement in 3 eyes (2 patients) and an associated decline of mean BCVA from 20/40 to 20/100.

Near-infrared autofluorescence (NIR-AF), which originates from the melanin-containing RPE and choroidal tissue, often was altered considerably. An early loss of RPE-derived NIR-AF caused an unmasking of the choroidal autofluorescent pattern predominantly in patients with type 3 mitochondrial retinopathy, with dark, large choroidal vessels and brighter choroidal stroma in between (Fig 6; Fig S3). Areas of RPE atrophy were not well visible on NIR-AF images because of a lack of contrast with areas of preserved RPE, which revealed a particularly low signal in areas that appeared granular on conventional AF images.

No optic atrophy was observed fundoscopically in patients 1 through 22, and when performed (n = 14), OCT-based measures of the peripapillary retinal nerve fiber layer thickness consistently were within the normal range. In contrast, patient 23 showed partial optic atrophy with mild thinning of the retinal nerve fiber layer (Fig S4).

Two patients showed macular changes reminiscent of macular telangiectasia type 2. Patient 2 showed an asymmetric foveal dip with temporal thinning and a focal wedge-shaped
loss of macular pigment temporally in both eyes (Fig S2). Patient 6 demonstrated similar changes in the right eye. The patient’s left eye showed hyporeflective cavities, substantial loss of macular pigment, increased reflectivity on blue reflectance imaging, and mild vascular leakage mainly in the temporal macula (Fig 7).
Systemic Disease Associations, Genetic Findings, and Muscle Biopsy

Other organ manifestations likely associated with mitochondrial disease were highly variable (Table S1). This included chronic progressive external ophthalmoplegia (CPEO) in 9 patients, of which 3 also had systemic findings (CPEO plus). One patient previously was diagnosed with neuropathy, ataxia, and retinitis pigmentosa (NARP) syndrome. Further frequently observed multisystem mitochondrial features included hearing loss (n = 12), diabetes (n = 8), cardiac abnormalities (n = 6), polyneuropathy (n = 5), and ataxia (n = 5).

Mitochondrial disease was confirmed by genetic testing (Table S1). In patients with type 1 or type 3 mitochondrial retinopathy, a sporadic single large-scale mitochondrial DNA deletion was identified in 9 patients, and previously described pathogenic missense variants of the mitochondrial genome (MT-TLI, MT-TK) were found in 2 patients. Furthermore, a previously described variant of unknown significance was detected in MT-TLI (patient 18), and a novel variant of unknown significance was identified in MT-RNRI and MT-ATP6, respectively (patients 19 and 20). All patients with type 2 mitochondrial retinopathy carried the m.3243G>A variant in the MT-TLI gene. Patient 22 carried a previously described MT-ATP6 variant. In patient 23, a novel homozygous variant in RRM2B was identified (confirmed by segregation analysis, the patient had known consanguineous parents). Structural evaluation of the p.Ala192Val variant and its high polyphen-2 score, 0.964, indicated likely pathogenicity (Fig S8, available at www.ophthalmologyretina.org).

Additional targeted next-generation sequencing (n = 9), containing a broad spectrum of genes associated with (syndromic) retinal dystrophies, did not detect additional variants that would explain the retinal phenotype (Table S2, available at www.ophthalmologyretina.org).

A muscle biopsy was performed in 13 patients and identified distinct characteristics of mitochondrial disease in 11 patients (Table S1; Fig S9, available at www.ophthalmologyretina.org). In addition, myopathic or neurogenic alterations, or both, were observed in 7 patients. In 2 patients (patients 1 and 19), the muscle biopsy results alone were not conclusive to confirm the clinical diagnosis of a mitochondrial disease.

Figure 6. Blue autofluorescence (left column) and near-infrared autofluorescence (NIR-AF) (right column) showing a distinctive pattern in patients with type 3 mitochondrial retinopathy in (A) patient 17 and (B) patient 19 (left eye, rotated). Because of an early loss of retinal pigment epithelium-derived NIR-AF, no distinction between areas with and without RPE atrophy is obvious using NIR-AF.
Exemplary Cases Demonstrating the Value of Retinal Phenotyping

The following 2 case reports exemplify the often multidisciplinary approach to diagnosing mitochondrial disease.

**Patient 17.** A 58-year-old patient with BCVA of 20/20 in both eyes reported vision problems in dim light conditions of approximately 5 years’ duration. He was taking medication for diabetes mellitus and psoriatic arthritis and had sensorineural hearing loss. His deceased mother had received a diagnosis of retinal degeneration and diabetes mellitus. Retinal examination revealed changes consistent with mitochondrial retinopathy type 3: widespread mottled fundus pigmentation and slightly constricted vessels (Fig 8A–C), paracentral atrophic areas of the outer retina and RPE that were associated with paracentral scotomata on visual field testing, and electoretinography examination results showing reduced scotopic and photopic responses. A subsequent neurologic examination revealed mild afferent ataxia with gait abnormalities, mild sensorimotor axonal and demyelinating polyneuropathy, as well as intermittent dysarthria and anomic aphasia. Histologic examination of a muscle biopsy sample with combined SDH and COX staining showed numerous COX-negative and SDH-positive fibers (Fig 8D) indicating mitochondrial disease. Mild neurogenic changes were also present. Genetic analysis identified a pathogenic variant in the MT-TL1 gene (m.3255G>A) previously reported in patients with myoclonic epilepsy with ragged red fibers and Kearns-Sayre syndrome.21,22

**Patient 21.** A 60-year-old patient with BCVA of 20/63 in the right eye and counting fingers in the left eye reported vision problems in dim light conditions and declining visual acuity of approximately 8 years’ duration. He previously had received a diagnosis of central areolar choroidal dystrophy. Funduscopy revealed granular fundus alterations and chorioretinal atrophy that included the peripapillary region, but spared a small foveal island (Fig 9). Photopic and scotopic responses were reduced on electretinography testing. Various multisystem disorders were diagnosed, including sensorineural hearing loss diagnosed at the age of 48 years, dilated cardiomyopathy (New York Heart Association classification II–III), chronic atrial fibrillation,
cardiac arrhythmia, exertional dyspnea, as well as cognitive and psychomotor changes; these alterations were never seen as part of a systemic disease. Because the retinopathy indicated a mitochondrial disease, a muscle biopsy was performed that revealed numerous COX-negative and SDH-positive fibers. Subsequent molecular genetic testing identified a single mitochondrial DNA deletion in skeletal muscle tissue. This patient illustrates the value of recognizing mitochondrial retinopathy in a patient with a previously indeterminate multisystemic disease.

**Discussion**

Mitochondrial disease may present with highly variable severity. For the retina, this was investigated previously in greatest detail for the m.3243G>A variant, for which 4 different severity grades were suggested, ranging from mild pigmentary abnormalities at the level of the RPE to fovea-involving chorioretinal atrophy.7-9 Fundus changes associated with the m.3243G>A variant usually remain...
limited to the posterior pole, with normal responses on full field electroretinography testing. 7–16

Other retinal phenotypes have been reported in patients with various mitochondrial diseases, but terminology and depth of phenotyping have been inconsistent. For instance, Kearns and Sayre described, in 1958, patients with “pigmentary degeneration” on fundoscopy, night vision abnormalities, and some histologic similarities, but also clinical differences, to retinitis pigmentosa (RP). 27 In subsequent publications, the retinal phenotype of Kearns-Sayre syndrome was described as RP, atypical RP, tapetoretinal degeneration, salt-and-pepper retinopathy, and pigmentary retinopathy. 28–30 We believe that most previously described cases match the phenotype classified herein as mitochondrial retinopathy type 3 and that the diagnosis of RP may have been imprecise. The macular involvement seems to follow a similar pattern as that observed in patients with the m.3243G>A variant (mitochondrial retinopathy type 2), but a distinctive feature of type 3 disease is the more widespread retinal involvement, as shown on widefield imaging and fullfield electroretinography testing.

Mitochondrial retinopathy type 1 shows only mild fundus changes and may represent grade 1 changes described previously. 7 Although currently it is unclear if type 1 changes may progress to type 2 or 3 disease, type 1 disease in older patients will be at the mild end of the spectrum of mitochondrial retinopathy.

Overall, the broad view on retinal changes associated with mitochondrial disease allowed a classification into different subtypes (Table S3, available at www.ophthalmologyretina.org) of mitochondrial retinopathy. This classification may be extended in the future if additional consistently associated phenotypes are identified. For instance, the occult retinopathy in patient 22 and the characteristic retinal phenotype observed in patient 23 indicate that rare types of mitochondrial retinopathy exist; however, additional patients with such phenotypes would need to be identified to confirm whether this is indeed the case. Potential overlaps between phenotypes, and if change from one pattern to another may occur, also remain to be investigated.

Pathophysiologic Considerations and Differential Diagnosis

Structural abnormalities identified in this cohort indicate that the primarily affected cell layer is usually the RPE, supporting previous clinical and histologic observations. 36–40 The most extensive and specific changes were observed on autofluorescence imaging that mainly relies on signals originating in the RPE, such as lipofuscin and melanin. 41 Early changes on OCT imaging occur at the interface between photoreceptor cells and the RPE cell layer, where postmortem specimens showed loss of the normal apical microvilli structure of RPE cells in areas with preserved photoreceptors. 36–38 Patients 22 and 23 were exceptions in this cohort because findings on retinal imaging indicated photoreceptor atrophy, whereas the RPE layer remained apparently unaffected or less affected.

Histologic studies of mitochondrial retinopathy reported a depletion of melanin granules from RPE cells. 36,38–40 Although the mechanism for this finding remains unknown, it may explain the lack of normal NIR-AF signal that is associated with a granular pattern on short-wavelength autofluorescence images. Comparable observations on multimodal imaging have been made in patients with choroideremia, indicating similar structural alterations in RPE cells. 32,42 Lack of (physiologic) RPE melanin may lead to decompensation of mechanisms counteracting oxidative stress, hence, eventually leading to cell death. 44

Despite pronounced retinal alterations, many patients have only mild symptoms and preserve good visual acuity because of relative foveal sparing, whereas vision problems in dim light conditions frequently are present. Whether the abnormal RPE—photoreceptor interface may explain the reduced rod function (e.g., resulting from an impaired recovery of photopigment) that is typically associated with widespread changes on autofluorescence images (type 3 disease) remains to be investigated using more refined electroretinography test protocols, because a primary effect of mitochondrial dysfunction on rods cannot be excluded based on this dataset. Ganglion cells, which also have a high energy demand, seem to be resilient to degeneration in most patients with mitochondrial retinopathy. It will be interesting to better understand pathophysiologic differences compared with patients with Leber hereditary optic neuropathy and whether some patients with Leber hereditary optic neuropathy may demonstrate mild retinal alterations.

Neurosensory alterations that may occur independent of RPE changes were observed in 2 patients with features typical for early macular telangiectasia type 2; 45–47 a macular disease with complex pathophysiology that likely involves mitochondrial dysfunction. 48 A recent genome-wide analysis identified susceptibility loci that include genes encoding mitochondrial enzymes, 49 and patients with macular telangiectasia type 2 may show low serine levels and increased levels of deoxyphosphoglycerides, both of which have been linked to disrupted mitochondrial function. 50–52 Advanced mitochondrial retinopathy with macular atrophy would preclude the diagnosis of macular telangiectasia type 2, which is always limited to a specific macular area. 53 Hence, screening of a larger number of patients with nonatrophic mitochondrial retinopathy will be necessary to confirm a true association with macular telangiectasia type 2.

The differential diagnosis may depend on the type of mitochondrial retinopathy and includes age-related macular degeneration, previous central serous chorioretinopathy, or retinal dystrophies such as Stargardt disease. Similar retinal phenotypes also may be observed in patients with Danon disease, rubella retinopathy, and similar postinflammatory conditions 54 as well as toxic retinopathies such as pentosan polysulfate maculopathy. 55 A very rare inherited retinal disease associated with mutations in the RCBTB1 gene recently was described with a similar retinal phenotype as type 3 mitochondrial retinopathy. 56 Such findings may indicate shared pathophysiologic pathways or involvement of the RCBTB1 gene in mitochondrial function.
Value of Retinal Phenotyping for Diagnosing Mitochondrial Disease

The clinical diagnosis of a mitochondrial disease often is challenging, and patients may experience a burdensome diagnostic odyssey. Genetic testing, histologic analysis of a muscle biopsy, or both may confirm the suspected diagnosis of a mitochondrial disease. However, results of both diagnostic approaches may remain inconclusive. Furthermore, targeted next-generation sequencing panels for retinal dystrophies currently include only a few mitochondrial genes. Detailed, non-invasive retinal phenotyping may support or even confirm the diagnosis of a mitochondrial disease, and hence may be seen as an additional useful biomarker. This includes that retinal examination may be supportive in evaluating the pathogenicity of novel identified mitochondrial variants if a characteristic retinal phenotype is present.

The retinal phenotype also may be the first indication of a mitochondrial disease, as exemplified in almost half of the patients presented herein. The characteristic retinal phenotype may lead to the diagnosis of mitochondrial disease in patients with otherwise mild systemic disease, as well as in patients with severe multisystem disease of unknown cause. This is of importance because an early and precise diagnosis is crucial, as it may lead to assessment of (treatable) systemic manifestations, lifestyle adjustments, and avoidance of drugs that may impact mitochondrial function. With various treatment options currently under investigation, a precise diagnosis may become even more important.

Study Limitations

This study is limited by the relatively small number of included patients, an intrinsic challenge of reporting phenotypic findings in rare diseases. The small sample size also may explain why no obvious association between severity of retinopathy and systemic disease manifestations or genotype was identified, although this also may be explained by various degrees of heteroplasmy and tissue segregation effects. The frequency of retinopathy in patients with mitochondrial disease remains to be determined, because the presence of retinal changes was a prerequisite in this cohort.

Conclusions

Mitochondrial retinopathy presents with a spectrum of distinct phenotypes. Specific retinal changes may be a characteristic finding in a variety of mitochondrial diseases and—in some patients—may be crucial for achieving the systemic diagnosis. Mild, asymptomatic mitochondrial retinopathy might be underreported, but severe mitochondrial retinopathy may be associated with considerable vision loss and disease burden. Ophthalmologists may play an important role in the diagnosis of patients with mitochondrial disease.

Footnotes and Disclosures

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References

27. Kearns TP, Sayre GP. Retinitis pigmentosa, external ophthalmoplegia, and complete heart block: unusual syndrome with histologic study in one of two cases. AMA Arch Ophthalmol. 1958;60:280–289.


