Peroxisomal Disorders and Retinal Degeneration

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Abstract Peroxisomal disorders are a group of inherited metabolic diseases, which can be incompatible with life in the postnatal period or allow survival into adulthood. Retinopathy is a recurrent feature in both the severely and mildly affected patients, which can be accompanied with other ophthalmological pathologies. Thanks to next-generation sequencing, patients originally identified with other inherited blinding diseases were reclassified as suffering from peroxisomal disorders. In addition, new peroxisomal gene defects or disease presentations exhibiting retinal degeneration were recently identified. The pathogenic mechanisms underlying retinopathy in peroxisomal disorders remain unresolved.

XX.1 Introduction

Peroxisomes are small organelles fulfilling a range of metabolic functions in virtually all eukaryotic cells. As their name suggests, they are involved in redox homeostasis, but they also play essential roles in lipid metabolism (Van Veldhoven 2010; Wanders and Waterham 2006). This includes the breakdown of phytanic acid via α -oxidation and the degradation of very-long-chain (polyunsaturated) fatty acids (VLC(PU)FA), dicarboxylic acids and the branched chain pristanic acid via peroxisomal β -oxidation. Besides catabolism, peroxisomal β -oxidation is essential for the synthesis of docosahexaenoic acid (DHA), which is known to be enriched in photoreceptor outer segments (POS), and for the formation of bile acids. The synthesis of ether lipids also depends on peroxisomal enzymatic steps.

Retinopathy is a frequent pathology in rare inherited diseases caused by peroxisome dysfunction. Peroxisomal disorders can be categorized into peroxisome biogenesis disorders (PBD), in which the whole organelle is dysfunctional due to a mutation in a *PEX* gene, and single enzyme deficiencies (SED), in which one of the metabolic pathways is defective (Wanders and Waterham 2006). In this review, we will focus on the ocular phenotypes in Zellweger spectrum disorders (ZSD) and in peroxisomal β -oxidation disorders, highlighting recent novel insights obtained, thanks to improved genetic diagnosis.

XX.2 Eye Abnormalities Are Recurrent Symptoms in Peroxisome Biogenesis Disorders

To date, mutations in 14 *PEX* genes are known to cause PBD of which the disease severity depends on the residual function of the related peroxin. Peroxins are necessary to assemble and maintain proper functioning peroxisomes (Argyriou et al. 2016). This results in a range of clinical manifestations, described as Zellweger spectrum disorders (ZSD) and rhizomelic chondrodysplasia punctata type I. Patients with the most severe phenotype, Zellweger syndrome, present with cranial dysmorphisms, hypotonia and liver dysfunction, and mostly die within the first year of life, whereas patients with intermediate or mild phenotypes, survive into childhood and sometimes into adulthood (Argyriou et al. 2016). The most common mutations occur in the *PEX1* and *PEX6* genes (~60% and 15%, respectively) (Ebberink et al. 2011).

Already in the 1970s, peroxisomes were described in the retina by using diaminobenzidine histochemistry to detect catalase (Leuenerberger and Novikoff 1975; Robison and Kuwabara 1975). More recently, we (Das et al. 2019) and others (Argyriou et al. 2019; Smith et al. 2016; Zaki et al. 2016) described the peroxisomal distribution in the retina more accurately by using immunohistochemical stainings and immunoblotting for peroxins. Peroxisomes occur in every retinal layer but are particularly abundant in the inner photoreceptor segments, in the RPE and in retinal ganglion cells, indicating that these organelles exert important functions both in the inner and outer retina. This is further supported by abnormal retinal pigmentation, retinal degeneration and impaired electroretinogram (ERG) responses in ZSD. Other frequently found ophthalmological defects include optic atrophy, glaucoma, cataract and nystagmus (Argyriou et al. 2016; Berendse et al. 2016; Courtney and Pennesi 2013; Noguer and Martinez 2010). Only in a few reports, the retinal histopathology was described, showing RPE atrophy, photoreceptor outer and inner segment degeneration, ONL thickness reduction and atrophy of the ganglion cell and nerve fibre layers as well as optic nerve atrophy. Macrophage infiltration was found in the retina, optic nerve and vitreous (Cohen et al. 1983; Courtney and Pennesi 2013; Glasgow et al. 1987).

Interestingly, several patients presenting with a disease phenotype of Leber's congenital amaurosis (Lambert et al. 1989; Majewski et al. 2011; Michelakakis et al. 2004) or Usher's syndrome (Raas-Rothschild et al. 2002; Zaki et al. 2016) have been re-diagnosed as suffering from ZSD, highlighting the need for differential diagnosis. More recently, Heimler syndrome was added to the mildest end of the ZSD. The syndrome was first described in two siblings suffering from sensorineural hearing loss (SNHL), enamel hypoplasia and nail abnormalities (Beau's lines) (Heimler et al. 1991). Through genetic analysis, the disease-causing mutations in Heimler patients were found in the *PEX1* or *PEX6* gene (Ratbi et al. 2015; Ratbi et al. 2016; Smith et al. 2016; Wangtiraumnuay et al. 2018) or in the *PEX26* gene (Neuhaus et al. 2017). The ocular phenotype consisting of a late-onset macular dystrophy and retinal pigmentation was described more recently (Lima et al. 2011) but appears to be variable. According to Ratbi et al. (2015), 7 out of 12 patients suffered from retinal pigmentation, whereas only 2 developed macular dystrophy. Interestingly, the ocular pathology is mainly situated in the posterior part of the eye, and the optic nerve seems to be spared (Wangtiraumnuay et al. 2018).

It remains unresolved why mild mutations in other *PEX* genes such as *PEX2*, *PEX10* and *PEX12* do not give rise to retinal defects but result in cerebellar ataxia (Argyriou et al. 2016). Importantly, a mouse model in which the most common mutation in the *PEX1* gene (G843D) was mimicked also develops retinal degeneration (Argyriou et al. 2019). This paves the way for preclinical studies to test AAV-mediated gene therapy approaches.

XX.3 Selective Defects in Peroxisomal β -Oxidation Cause Retinal Degeneration

The three most prevalent SED causing deficient β -oxidation are X-linked adrenoleukodystrophy (X-ALD), multifunctional protein 2 (MFP-2) and acyl-CoA oxidase 1 (ACOX1) deficiency, in decreasing order of occurrence (Van Veldhoven 2010). Although these proteins are involved in the same metabolic pathway, the ocular phenotypes are markedly different.

X-linked adrenoleukodystrophy (X-ALD) is caused by a mutation in the *ABCD1* gene, necessary for the import of saturated VLCFA in the organelle preceding their degradation via β -oxidation (Kawaguchi and Morita 2016). Visual deterioration due to optic nerve atrophy is frequently found, but the outer retina is not affected (Grainger et al. 2010).

Acyl-CoA oxidase 1 (ACOX1) catalyses the first desaturation step in the peroxisomal β -oxidation pathway (Van Veldhoven 2010). Like ZSD, ACOX1 deficiency is characterized by an array of eye abnormalities: retinal pigmentation and degeneration, optic atrophy, nystagmus, strabismus and fixation problems (Carrozzo et al. 2008; Ferdinandusse et al. 2007; Suzuki et al. 2002; Suzuki et al. 1994; Wang et al. 2014). Interestingly, one patient presented with pigmentary retinopathy accompanied with normal ERG responses (Kurian et al. 2004), whereas another patient displayed normal fundi but flattened ERG responses (Poll-The et al. 1988). Most patients suffer from a severe disease course and do not survive into adolescence. However, two siblings with a milder phenotype were described, surviving into their 50s, and presenting with distinct ocular phenotypes (Ferdinandusse et al. 2010). Fundoscopy in the male patient showed retinitis pigmentosa, but no optic disc abnormalities. He also developed small lens opacities, whereas his sister developed bilateral cataracts with nonvisible fundi.

The second (hydration) and third (dehydrogenation) step are catalysed by multifunctional protein 2 (MFP-2, also called D-bifunctional protein (D-BP)) for most peroxisomal β -oxidation substrates (Van Veldhoven 2010). Ferdinandusse et al. (2006) described the clinical and metabolic presentation of a cohort of 126 patients suffering from MFP-2 deficiency. Severely affected patients are almost indistinguishable from patients suffering from Zellweger syndrome, and their visual defects are also similar to those in ZSD and ACOX1 deficiency. However, several patients with a milder disease course were described, the so-called juvenile-onset MFP-2 deficiencies. Abnormal retinal pigmentation with or without abnormal ERG responses was reported, but visual acuity was normal (Amor et al. 2016; Lines et al. 2014; McMillan et al. 2012). Interestingly, no eye pathologies were reported in a few patients with dysfunction in other enzymes participating in peroxisomal β -oxidation, i.e. ABCD3, ACOX2 and SCPx.

Acyl-CoA binding domain containing protein 5 (ACBD5) is a quite novel peroxisomal membrane protein that was assigned with multiple functions, including the tethering of peroxisomes to the ER, peroxisomal mobility, lipid transfer between the ER and peroxisomes, and assisting in the catabolism of VLCFA (Costello et al. 2017; Ferdinandusse et al. 2017; Hua et al. 2017; Yagita et al. 2017). Remarkably, exome sequencing of a large cohort of retinopathy patients revealed mutations in the *ACBD5* gene (Abu-Safieh et al. 2013).

XX.4 Lack of Mechanistic Insights

In all peroxisomal disorders, the mechanisms linking peroxisomal protein dysfunction with the retinal pathology remain obscure. In mildly affected patients, routine metabolite testing in plasma often does not reveal any or only small increases in VLCFA concentrations. It was hypothesized several decades ago that the lack of DHA in the retina was causative for the retinopathy in ZSD and peroxisomal β -oxidation deficiency (Martinez 1992). However, several D-BP patients with severe retinopathy had normal DHA levels in plasma (Ferdinandusse et al. 2006). In an open trial, supplementation of DHA appeared to stabilize and even improve the visual acuity of ZSD patients (Noguer and Martinez 2010). Conversely, in a double-blind, placebo-controlled study, no clinical benefit of DHA was observed (Paker et al. 2010). Further research will be necessary to elucidate the contribution of peroxisomal metabolism to retinal homeostasis.

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