Retinal ciliopathies

Essay
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A ciliopathy is a genetic disorder of the cellular cilia or the cilia anchoring structures, the basal bodies, or of ciliary function. The phenotypic parameters that define a ciliopathy may be used to both recognize the cellular basis of a number of genetic disorders and to facilitate the diagnosis and treatment of some diseases of unknown etiology.

The cilium is a nanomachine composed of perhaps over 600 proteins in molecular complexes, many of which also function independently as nanomachines. Cilia are present in almost every cell in the body "primary cilia". Each cilium is composed of a microtubule backbone, called an axoneme. This axoneme is the central cytoskeletal core of the cilium. This axoneme develops from and is anchored to a specialized centriole called the basal body. The basal body, like centrioles, acts as microtubule organizing centers for the cilia. The primary microtubule organizing center of a cell is the centrosome, which consists of two centrioles surrounded by pericentriolar material. The basal bodies of the cilia consist of nine microtubule triplets. Classically, cilia are divided into two main types according to the geometry of microtubules within the ciliary axonemes:“9+2” (motile) cilia composed of nine microtubule doublets surrounding a central pair, and “9+0” (primary, nonmotile) cilia composed of only the nine microtubule doublets without a central pair. An alternative classification categorizes cilia into three groups: motile, primary, and nodal. The nodal cilia contain a “9+0” morphology as do the primary cilia, but have been localized to the node in gastrulating-stage embryos.

In photoreceptors, the connecting cilium is a slender structure that connects the outer and inner segments. In addition to its structural role, the photoreceptor cilium plays a critical transport function. Because the

outer segments of photoreceptors are incapable of synthesis of the essential proteins and lipids, all photo transduction proteins and disc membrane lipids must be synthesized in the inner segment and then transported to the outer segment. A specialized system termed intraflagellar transport (IFT) is responsible for moving cargo toward the axonemal tip or away from it. This cargo is transported by the microtubule associated proteins (MAPs), namely the kinesins responsible for anterograde transport. 99Actin, which has been detected at the membrane of the entire cilium, may provide the structural basis for the motility of myosin VIIa, an unconventional myosin, which is thought to be involved in the active transport of the visual pigment rhodopsin through the connecting cilium. Other proteins have been mapped to the cilia and basal bodies, suggesting other potential functions of these structures.

The localization of numerous proteins implicated in retinal degenerations to the retinal cilium has incited a tremendous amount of research on this small organelle both in animal models as well as in human subjects. While the functions of many of the proteins located in the cilia are poorly understood, disruption of the function of these proteins may result in a wide variety of phenotypes ranging from isolated retinal degeneration to more pleiotropic phenotypes.

Retinitis pigmentosa-1 (RP1) is a form of RP that is often transmitted in an autosomal dominant manner caused by a mutation in RP1 gene. Mutations in the retinitis pigmentosa 1 (*RP1*) gene are a common cause of retinitis pigmentosa .The four-exon *RP1* gene encodes a 2156 amino acid protein that is expressed exclusively in photoreceptor cells. The RP1 protein is located in the region of the connecting cilium and axoneme of photoreceptor cells. RP1 mutation may cause

photoreceptor dysfunction by disrupting disc formation, may also be involved in transport of rhodopsin from the outer to the inner segment, RP1 may be part of a protein complex that captures the outer segment discs then links and aligns them correctly with the axoneme so disruption of RP1 gene results in loss of the organized perpendicular arrangement of the discs, RP1 gene may be also be involved in microtubule dynamics, and ciliary axoneme length regulation. Clinically patients with RP1 mutations, have the general picture of RP but visual acuity is often preserved at the level of 20/20 to 20/70 until the late decades of life. Visual fields losses, however, are more severe and occur earlier.

The *XlRP3 (*XL retinitis pigmentosa 3) locus encodes at least two major isoforms of the retinitis pigmentosa GTPase regulator (RPGR), $RPGR_{1–19}$ and $RPGR_{ORF15.}$ $RPGR_{1–19}$ is encoded by 19 exons of *XlRP3*, whereas $RPGR_{ORF15}$ is produced from the alternate retention of the purine-rich intron 15 leading to an RPGR isoform with a terminal and extended exon 15. The exact function of RPGR is not fully understood, however through its interaction with various molecules, it is believed that RPGR plays a role in disc morphogenesis and intracellular trafficking as well as nucleocytoplasmic transport and other nuclear activities. The majority of the mutations causing *XlRP3* are found in the C-terminal domain of $RPGR_{ORF15}$ and mutations were never found in the sequence encoding the unique C-terminal domain of $RPGR_{1–19.}$ Mutations in the retinitis pigmentosa GTPase regulator (RPGR) gene result in various diseases include: X-linked RP, macular degeneration, x-linked cone dystrophy, and primary ciliary dyskinesia.

RPGR-IP (retinitis pigmentosa GTPase regulator-interacting protein), also known as RPGR-IP1, is a component of the axoneme of the photoreceptor connecting cilium. RPGRIP was identified through its

interaction with RPGR. Because RPGRIP associated stably with the ciliary axoneme and its localization remained unchanged in photoreceptors lacking RPGR, RPGRIP was proposed to be the primary resident, whereas RPGR depended on RPGRIP for its localization in the connecting cilia, It's function in disc morphogenesis may be due to direct or indirect effect. Mutations in the retinitis pigmentosa GTPase regulator interacting protein (RPGR-IP) are associated with Leber congenital amaurosis and cone-rod dystrophy.

The proteins encoded by the Usher genes are members of protein classes with very different functions. Usher Proteins form an Usher protein scaffold network at the photoreceptor connecting cilium They may also have "non-ciliary" functions as suggested by their localization in the outer plexiform layer to photoreceptor ribbon synapses, which are structurally and functionally specialized for massive and sustained neurotransmitter release. Mutations in the Usher genes result in an autosomal recessive disorder known as Usher syndrome which is characterized by bilateral deafness and RP and is considered the most common cause of combined blindness and deafness is classified into three main categories according to the age of onset and the degree of hearing loss and vestibular involvement.

NPHP (Nephronophthisis) is largely inherited as an autosomal recessive disease with homozygous single gene mutations/deletions or compound heterozygous mutations occurring in a single NPHP gene. This usually allows a molecular diagnosis and accurate genetic counseling to be performed. More than nine nephronophthisis genes have been identified to date, each of which encodes a nephrocystin protein. Nephrocystin proteins have role in formation of functional complexes at the ciliary base, protein interactions and iiliogenesis. Mutations in the

nephronophthisis genes result in autosomal recessive nephronophthisis with or without retinopathy and other associated conditions. Nephronophthisis is a chronic renal disease characterized by progressive "phthisis" or wasting away of the tubules and glomeruli. This results in the formation of corticomedullar cysts in the kidney and ends in renal failure. The most commonly associated syndrome is retinal dystrophy and retinal degeneration leading to blindness (Senior–Loken syndrome). Other associations include Joubert syndrome and related diseases (JSRD). NPHP is genetically and clinically heterogeneous. Traditionally, NPHP has been subdivided into infantile, juvenile and adolescent forms, based on the age of onset of renal failure.

The BBS(Bardet-Biedl Syndrome) proteins are a heterogonous group of proteins that do not all belong to the same functional category, and unlike the numerous NPHP proteins that form a functional group, the BBS proteins may not interact with each other. Mutations in 14 genes are known to be associated with BBS: BBS1, BBS2, ARL6 (BBS3), BBS4, BBS5, MKKS (BBS6), BBS7, TTC8 (BBS8), BBS9, BBS10, TRIM32 (BBS11), BBS12, MKS1 (BBS13), and CEP290 (BBS14), Also mutations in WDPCP (BBS15) and SDCCAG8 (BBS16) may be associated with BBS. About 16 BBS genes have been identified to date. However, the function of these genes remains largely unknown. Of these genes, three (BBS1, BBS2, and BBS7) are believed to have a beta-propellar domain that are known to have a variety of different functions, and two (BBS4 and BB8) are believed to have tetratricopeptide repeat domains that function in protein-protein interactions, The BBS3 gene encodes a small Ras-family protein called ADPribosylation- like protein 6 (ARL6), which is a small GTPase protein with regulatory functions, The BBS10 and BBS12 genes as well as the BBS6 gene (also known as

MKKS for McKusick- Kaufmann Syndrome) encode chaperonin-related proteins that are believed to play a role in protein folding or assembly. The BBS11 gene encodes a TRIM (tripartite motif) family protein that has ubiquitin ligase activity, enabling it to attach ubiquitin onto lysine residues on proteins to mark them for degradation by proteasomes , BBS8 was found to form a stable complex with PCM1 (pericentriolar material 1). PCM1 is believed to be involved in ciliogenesis by recruiting proteins necessary for centrosome replication and in organizing or anchoring microtubules and BBS4 is believed to function as a subunit of the dynein molecular motor, whereby it may recruit PCM1, and assist in intraflagellar transport. Mutation in BBS genes result clinically in Bardet-Biedl Syndrome (BBS) an autosomal recessive disorder that affects numerous tissues and often displays RP. The diagnosis of BBS is clinical and there is a remarkable degree of inter- and intra-familial variability. Therefore, the diagnosis is based on the presence of four of six *primary clinical features.* In cases that have only three of the six primary clinical features, two *secondary features* are required for diagnosis.

The concept of "retinal ciliopathies" brings to attention the importance of further molecular analysis of this organelle in an attempt to identify further candidate genes for retinal degeneration. However, with the known genes involved in the retinal ciliopathies, it remains unclear whether the clinical phenotype results from disturbance of the ciliary function or from disruption of other functions of the gene product, particularly for proteins that have been located to more than one subcellular compartment, such as certain Usher or BBS proteins. Grouping the retinal ciliopathies together may be important in diagnosing disease as diagnostic technologies advance and in developing targeted therapies for these disorders.

Furthermore, understanding many of these conditions as ciliopathies leads the ophthalmologist to have a high degree of suspicion that a systemic finding may be present. As such, it become imperative to look for associated disorders of cilia: neurosensory hearing loss, anosmia, developmental delay, mental retardation, hydrocephalus, neural tube defects, situs-inversus, infertility and other reproductive system disorders, disorders of limb and digit development, oral malformations, obesity, cystic kidney disease and renal failure, liver disease, respiratory disease, and other ciliopathies which are only now being recognized as such.