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REVIEW

Genetics and pathological mechanisms of Usher syndrome

Denise Yan and Xue Z Liu

Usher syndrome (USH) comprises a group of autosomal recessively inherited disorders characterized by a dual sensory impairment of the audiovestibular and visual systems. Three major clinical subtypes (USH type I, USH type II and USH type III) are distinguished on the basis of the severity of the hearing loss, the presence or absence of vestibular dysfunction and the age of onset of retinitis pigmentosa (RP). Since the cloning of the first USH gene (MYO7A) in 1995, there have been remarkable advances in elucidating the genetic basis for this disorder, as evidence for 11 distinct loci have been obtained and genes for 9 of them have been identified. The USH genes encode proteins of different classes and families, including motor proteins, scaffold proteins, cell adhesion molecules and transmembrane receptor proteins. Extensive information has emerged from mouse models and molecular studies regarding pathogenesis of this disorder and the wide phenotypic variation in both audiovestibular and/or visual function. A unifying hypothesis is that the USH proteins are integrated into a protein network that regulates hair bundle morphogenesis in the inner ear. This review addresses genetics and pathological mechanisms of USH. Understanding the molecular basis of phenotypic variation and pathogenesis of USH is important toward discovery of new molecular targets for diagnosis, prevention and treatment of this debilitating disorder.
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INTRODUCTION

Usher syndrome (USH) is a group of recessively inherited disorders characterized by deafness and vision loss. The blindness occurs from a progressive retinal degeneration termed retinitis pigmentosa (RP). Vestibular dysfunction may also be a component. It has been estimated that USH accounts for between 3 and 6% of the congenitally deaf population, up to 8–33% of individuals with RP and 50% of the deaf–blind population.[1,2] Prevalence of USH in different populations ranging from 3.5 to 6.2 per 100 000, and a carrier rate of 1 in 100, have been reported.[1–3] However, the syndrome is more common in regions with small, isolated, often inbred populations such as in Israel (Samarathians), Pakistan (Hutterites), France (Poitou-Charentes region), Northern Sweden, Finland and Accadian population of Louisiana, United States.[2–6] Progress on the molecular genetics and clinical research of USH has revealed broad genetic and clinical heterogeneity. USH was historically divided into three clinical subtypes: USH type I, USH type II and USH type III (USH1, USH2 and USH3). Although this classification of USH remains in clinical use, atypical clinical types have been described that defy this easy classification. Each USH subtype is genetically heterogeneous. Seven USH1 loci (USH1B–USH1H) have been identified so far by linkage analyses of USH1 families. Five of these genes have been isolated (see http://webhost.ua.ac.be/hhh/). Three genetic loci for USH2 (USH2A, USH2C and USH2D) have been reported and the corresponding genes have been determined. To date, only 1 USH3 locus has been described. Although a common phenotype has been described in the different USH types, the identified USH genes encode for proteins from different protein classes and families. Growing evidence suggests that these proteins are organized in a protein 'interactome' in both the inner ear and the retina. This network may be critical for the development and maintenance of the sensorineural cells.[7–13] These progresses on the molecular genetics have greatly advanced our understanding of the genetic causes and the pathogenesis of USH. In this review, we summarize the current developments in the field of USH.

USH IS CLINICALLY AND GENETICALLY HETEROGENEOUS

Traditionally, USH is subdivided into three clinical subclasses. The subtypes are differentiated by the severity and progression of the hearing loss and by the presence or absence of vestibular symptoms, with visual impairment because of RP being common to all three subtypes. USH shows significant genetic heterogeneity, and at least 11 distinct loci have been identified and genes for 9 of them have been cloned.

USH type 1

USH1 patients are congenitally profoundly deaf, and have vestibular dysfunction as well as prepubertal onset of progressive RP.[14,15] A delay

Department of Otolaryngology, Miller School of Medicine, University of Miami, Miami, FL, USA
Correspondence: Dr XZ Liu, Department of Otolaryngology (D-48), University of Miami, 1666 NW 12th Avenue, Miami, FL 33136, USA.
E-mail: xliu@med.Miami.edu
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in motor development is the clinical indication of congenital absence of vestibular function. The audiometric configuration is described as 'residual' with hearing in the low frequencies being generally preserved.[16,17] Thus, children diagnosed with USH1 are ideal cochlear implant candidates.[16–21] To date, seven genetic loci for USH1 (*USH1B–H*) have been mapped on chromosomes 14q32, 11q13.5, 11p15.1, 10q22.1, 21q21 and 10p-22, 17q24-25 (http://webhost.ua.ac.be/hhh/). Five of the corresponding genes have been cloned: the actin-based motor protein myosin VIIa (*Myo7a*, *USH1B*);[22,23] two cadherin-related proteins, otocadherin or cadherin 23 (*Cdh23*, *USH1D*)[24,25] and protocadherin 15 (*Pcdh15*, *USH1F*);[26,27] and two scaffold proteins, harmonin (*USH1C*)[28,29] and sans (*USH1G*).[30,31] USH1 is the most severe form of USH. This form accounts for 30–40% of USH.[15,32,33] We and others have shown that the most common USH1 genetic subtype is USH1B, which accounts for between a third and one half of USH1 in the United Kingdom and the United States.[5,34,35] Mutations in the *CDH23* gene at the USH1D locus are the second most frequent cause of USH1, accounting for between 10 and 35% of the phenotype.[34,35] Defects in *PCDH15* was found to account for 11% of the US and UK cohort of USH1,[35] and may be the most common cause of USH1 among Ashkenazi Jewish families, due to a founder mutation.[36] The R245X mutation of the gene was detected among a large proportion of cases of USH1 in this population.[37] USH1C, identified mainly among the Acadian population of Louisiana,[38,39] has also been detected in diverse ethnic groups.[40] The genetic cause of USH1 generally leads to a typical USH 1 phenotype. However, we have previously shown that mutations in *MYO7A*, the gene responsible for USH1B, can also result in a wide phenotypic spectrum, including atypical USH.[41] *MYO7A* has also been shown to harbor mutations causing both nonsyndromic dominant (DFNA11) and recessive (DFNB2) deafness. DFNA11 is characterized by progressive sensorineural hearing loss with varying degrees of vestibular dysfunction,[42–47] whereas DFNB2 has been reported to cause congenital profound deafness and variable vestibular dysfunction.[48,49] There was no obvious correlation between mutation in the MOY7A gene and the resulting phenotype. Mutations of the gene encoding harmonin have been identified as the primary defect in *USHIC* patients.[28,29] We and other have subsequently reported that they can also result in nonsyndromic recessive deafness DFNB18.[50,51] Most of the reported phenotypic variability in USH1 is associated with mutations in the *CDH23* gene (USH1D). In humans, missense mutations in *CDH23* have also been reported to cause nonsyndromic deafness (DFNB12).[25] In a study of ethnically and geographically diverse USH1D families, Astuto *et al.*[34] reported an atypical clinical presentation of USH1D. Atypical features included moderate to severe hearing loss, progressive and or/asymmetric deafness and normal motor development coupled with either normal or mildly abnormal vestibular function. There is a clear phenotypic/genotypic correlation in patients with mutations in the *CDH23* gene: homozygosity for truncating nonsense, frameshift and splice site mutations have been reported to cause typical USH1D, whereas missense mutations result in either a milder form, which overlaps with clinical types USH2 or 3, or nonsyndromic deafness.[52,53] Atypical USH1 has also been associated with mutations in the *SANS* gene (USH1G).[54] The affected individuals had moderate to profound prelingual deafness. Vision and vestibular were normal.

USH type 2

USH2, which is less severe than type 1, is characterized by congenital moderate to severe deafness, with a high-frequency sloping configuration. The vestibular function is normal and onset of RP is in first or second decade. The onset of the visual symptoms in type 2 occurs usually several years later than for USH1. The mean age at onset of night blindness in type 2 is 15 years, and the mean age of diagnosis of RP is 24 years.[55] Owing to the overlap in the clinical appearances of visual symptoms in types I and II due to considerable variation in age of onset, these symptoms are not considered reliable predictors of USH type in individual cases.[14–16,56] Furthermore, it has been reported that the severity of the visual signs and symptoms does not differ significantly in USH type I and II.[55,57,58]

Three genetic loci have been reported so far in USH2 (USH2A, USH2C and USH2D). The corresponding genes have been cloned. Mutations in the *USH2A* gene on chromosome 1q41, encoding usherin, are the most common accounting for up to 85% of the USH2 cases.[59,60] USH2A was previously described as an extracellular matrix protein.[61,62] A second USH2A isoform (isoform B) containing a transmembrane region and a short cytoplasmic part was subsequently identified.[63] Mutations in *USH2A* have been associated with a wide spectrum of phenotypes, including typical USH2 and atypical USH2, and can also lead to nonsyndromic autosomal recessive RP. Progressive hearing loss was reported in patients who are heterozygous for the most common mutation in the *USH2A* gene, 2299delG, and another frequent mutation of the gene, C759F.[64–66] The C759F mutation in homozygous state was reported to cause nonsyndromic RP.[67–69] The protein encoded by the *VLGRI* (very large G-protein coupled receptor 1) gene at the USH2C locus is a member of the serpentine G-protein coupled receptor superfamily.[70] Defects in the *Whirlin* gene, a PDZ (post-synaptic density, disc-large, Zo-1 protein domains) domain-containing scaffold protein, are responsible for USH2D[71] and nonsyndromic hearing loss (DFNB31).[72] Mutations in USH2C and USH2D are rare.[70,71] There was no clear correlation between the variations in auditory phenotypes found in USH2 and the underlying molecular defects.

USH type 3

USH3 is characterized by variable onset of progressive hearing loss, variable onset of RP, and variable impairment of vestibular function (normal to absent).[73–75] In general, developmental motor milestones are normal in type 3. USH3 is not as common as USH1 and USH2 with a prevalence of 2–4% within all USH cases. However, it is the most prevalent form of USH in Finland and among the Ashkenazi Jewish population, where it accounts for up to 40% of the condition.[76,77] USH3 is caused by mutations in the USH3A (clarin-1) gene, mapped on 3q21-q25.[78,79] The nonsense mutation 528T-G, also known as Finmajor, seemed to be relatively common in Finnish patients with this genetic defect,[80] whereas in Ashkenazi Jewish families, the 144T-G missense mutation is particularly prevalent.[76,78,81] The USH loci, clinical features and related genes and proteins as well as the associated phenotypes are reported in Table 1.

THE USH MOLECULES ARE MEMBERS OF DIFFERENT PROTEINS CLASSES AND FAMILIES

The gene products of nine identified USH genes are members of protein classes with very different functions (Figure 1). The gene MYO7A (USH1B) encodes myosin 7A, an unconventional myosin, with a predicted domain structure consisting of a motor head domain, five calmodulin-binding IQ motifs, two FERM domains, two MyTH4 domains and an Src homology 3 (SH3) domain.[84] Three different classes of isoforms are identified for the USH1C protein, harmonin. All three isoforms contain two PDZ (PSD95, discs large, ZO-1) domains (PDZ1 and 2) and one coiled-coil domain. Class A isoforms

Table 1 USH subtypes, clinical features, loci, genes, proteins and additional phenotypes

		Clinical features							
Clinical USH subtypes	*USH locus*	*Auditory*	*Vestibular*	*Ocular*	*Gene*	*Protein*	*Protein class*	*Location*	*Nonsyndromic deafness locus*
	USH1A[a]								
USH1	USH1B	Profound congenital HL	Absent	Before puberty	*MYO7A*	Myosin VIIa	Motor protein	11q13.5	DFNA11/ DFNB2
USH1	USH1C				*USH1C*	Harmonin	Scaffold protein	11p15.1	DFNB18
USH1	USH1D				*CDH23*	Cadherin 23	Cell–cell adhesion protein	10q22.1	DFNB12
USH1	USH1E				*USH1E*	Unknown	Unknown	21q21	
USH1	USH1F				*PCDH15*	Protocadherin 15	Cell–cell adhesion protein	10q21.1	DFNB23
USH1	USH1G				*USH1G*	SANS	Scaffold protein with ankyrin repeats and SAM domain	17q25.1	
USH1	USH1H[b]				Unknown	Unknown	Unknown	15q22–23	
USH2	USH2A	Prelingual onset of moderate to severe high-frequency sloping HL	Normal	2nd decade	*USH2A*	Usherin	Transmembrane protein	1q41	
USH2	USH2B[a]								
USH2	USH2C				*VLGR1*	VLGR1, Mass1	Transmembrane receptor protein	5q14.3	
USH2	USH2D				*WHRN*	Whirlin	Scaffold protein	9q32–q34	DFNB31
USH3	USH3A	Variable onset of progressive HL	Variable	Variable	*USH3A*	Clarin-1	Transmembrane protein	3q25.1	

Abbreviations: HL, hearing loss; SAM, sterile α-motif; USH, Usher syndrome.
[a]The locus USH1A was withdrawn by Gerber *et al.*[82] who provided evidence that the French families mapped to this locus had mutations in the *MYO7A* gene, and the locus USH2B has been reported by Kremer *et al.*[9] as withdrawn.
[b]The USH1H has recently been reported after genome-wide linkage scanning of two large Pakistani consanguineous families.[83]

contain an additional PDZ domain (PDZ3). The class B isoforms also contain this third PDZ domain, a second coiled-coil domain and a proline, serine, threonine-rich region.[28,29] Otocadherin or cadherin 23 (*Cdh23* and *USH1D*)[24,25] and protocadherin 15 (*Pcdh15* and *USH1F*)[26,27] are two cadherin-related proteins. Cadherins are calcium-dependent cell adhesion proteins. *CDH23* complementary DNA encodes a very large, single-pass transmembrane protein. It has an extracellular domain containing 27 Ca^{2+}-binding extracellular cadherin and a short intracellular domain with a C-terminal class I PDZ-binding motif (PBM;-X[ST]X[VIL]$^{-COOH}$.[85] Similar to *CDH23*, PCDH15 (USH1F) has either 11 (isoform A) or one extracellular cadherin domain (isoform B), a transmembrane domain and a C-terminal class I PBM. The scaffold protein SANS (USH1G) is composed of three ankyrin domains (ANK), a central region (CENT), a sterile α-motif (SAM) and a C-terminal class I PBM.[30,31] The short isoform of the USH2A protein contains an N-terminal thrombospondin/pentaxin/laminin G-like domain, a laminin N-terminal (LamNT) domain, 10 laminin-type EGF-like (EGF Lam) and four fibronection type III (FN3) domains.[62] In addition to these regions, the long isoform contains two laminin G (LamG), 28 FN3, a transmembrane domain and an intracellular domain with a C-terminal class I PBM.[63] Isoform B of the very large G-coupled protein receptor, VLGR1 (USH2C), contains a thrombospondin/pentaxin/laminin G-like domain, 35 Ca^{2+}-binding calcium exchanger β (Calx) domains, seven EAR/EPTP repeats, a seven-transmembrane region and an intracellular domain containing a C-terminal class I PBM.[70] The long form of Whirlin (USH2D/DFNB31) contains a proline-rich domain and three PDZ domains, whereas the short C-terminal form, hereafter referred to as short whirlin, contains only the proline-rich and the third PDZ domain.[72] Clarin-1, the only USH3A protein, has four (isoform A) or one transmembrane (isoform C) domain.[79]

USH1 proteins are expressed in inner hair cells throughout life. Their spatial and temporal subcellular distributions vary dramatically during development until maturation is reached. Myosin VIIA is found expressed in the inner and outer hair cells of the inner ear, particularly in the stereocilia and in the cuticular plate.[23,86,87] Harmonin, CDH23 and PCDH15 are detected in the hair bundle as soon as it emerges from the surface of the sensory cells.[52,88] Harmonin isoform B is found present mainly at the tips of stereocilia during early postnatal stages but its expression decreases by postnatal day 30 in both the cochlea and vestibule.[88] CDH23 is first observed along the entire length of the emerging stereocilia and then becomes progressively confined to the tip region. PCDH15 has been detected uniformly distributed along the growing stereocilia.[52] Both CDH23 and PCDH15 are associated with tip links and kinocilial links.[89,90] During the differentiation of hair cells, CDH23 is localized at ankle and transient lateral links between the membranes of the neighboring

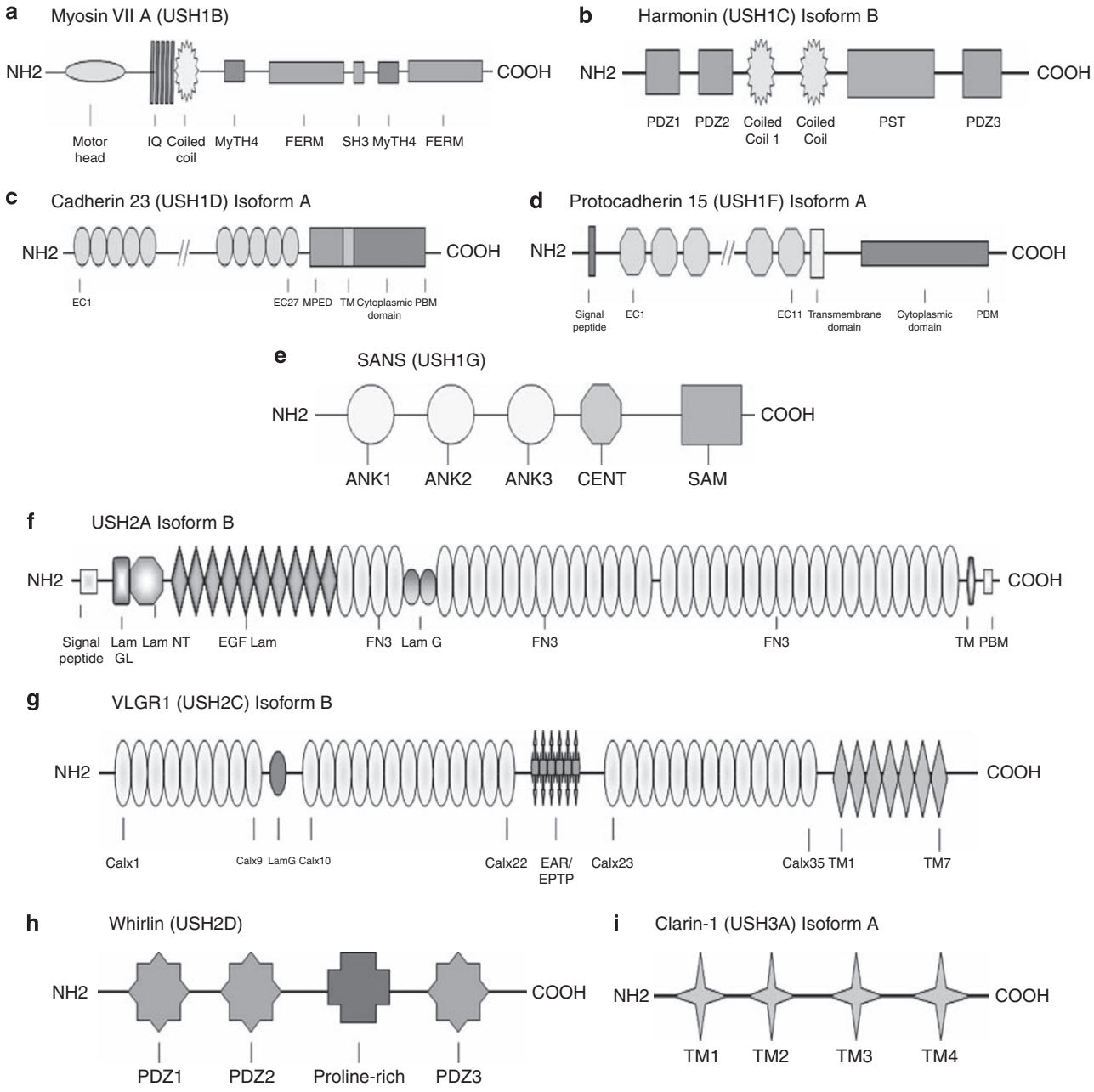

Figure 1 Schematic representation of the Usher proteins and their major isoforms. (**a**) Myosin VIIa consists of a motor head domain, followed by a neck region composed of five IQ (isoleucine-glutamine) motifs. The tail is composed of a coiled-coil domain, two FERM domains, two MyTH4 domains and an Src homology 3 (SH3) domain. (**b**) The USH1C protein, harmonin isoform B contains three PDZ (PSD95, discs large, ZO-1) domains (PDZ1, 2 and 3), two coiled-coil domains and a proline, serine, threonine-rich region (PST). (**c**) The isoform A of CDH23 is composed of 27 extracellular cadherin (EC) repeats (EC1-27), a membrane proximal extracellular cadherin domain (MPED), a transmembrane domain (TM) and intracellular domain with a C-terminal class I PDZ-binding motif (PBM). (**d**) PCDH15 (isoform A) has 11 EC repeats, a transmembrane domain and a C-terminal class I PBM. (**e**) The scaffold protein SANS consists of three ankyrin (ANK)-like repeats, a central region (CENT), a sterile α-motif (SAM) and a C-terminal class I PBM. (**f**) The isoform B of the USH2A protein is composed of an N-terminal thrombospondin/pentaxin/laminin G-like domain (LamGL), a laminin N-terminal (LamNT) domain, 10 laminin-type EGF-like (EGF Lam), four fibronection type III (FN3) domains, two laminin G (LamG), 35 FN3, a transmembrane domain and an intracellular domain with a C-terminal class I PBM. (**g**) The isoform B of the very large G-coupled protein receptor, VLGR1, has a thrombospondin/pentaxin/laminin G-like domain, 35 Ca^{2+}-binding calcium exchanger β (Calx) domains, six EAR/EPTP repeats, a seven-transmembrane region and an intracellular domain containing a C-terminal class I PBM. (**h**) Whirlin is composed of three PDZ domains (PDZ1, 2 and 3) and a praline-rich region. (**i**) Clarin-1, the USH3A protein (isoform A), has four transmembrane (TM) domains.

stereocilia.[91] In mature cochlear hair cells, both CDH23 and PCDH15 are localized at the tip links, in which they can form heteromeric complexes.[92] Interestingly, we have shown that mutations in both *Cdh23* and *Pcdh15* genes can interact to cause hearing loss in humans and mice. Digenic heterozygotes in both species are deaf, whereas single heterozygotes are normal.[93] SANS is highly concentrated below

the cuticular plate region of inner and outer hair cells and is especially abundant below the kinociliar basal body.[11] Both Usherin and VLGR1 are highly expressed in the basal region of the developing hair bundle, at the ankle-link level, during development, and expression disappears thereafter.[11,94] These two proteins are thus thought to be part of the transient ankle-link complex. In the mouse cochlea, whirlin is restricted to the stereocilia of the sensory inner and outer hair cells.[72] Expression pattern analysis localizes transcripts of USH3A to mouse cochlear hair cells and spiral ganglion cells.

THE USH PROTEINS ARE ORGANIZED IN A MUTUAL 'INTERACTOME'

The USH proteins mainly colocalize in the stereocilia and at the synaptic regions of hair cells of the inner ear.[7,8,11,88,95–97] Stereocilia are mechanosensing organelles located at the apical surface of both the auditory and vestibular hair cells. The bending of the hair bundle by a sound wave opens mechanically the gated transduction channels at the tip of the stereocilia, initiating the electrical signal cascade for sound perception.[98,99] Molecular and colocalization analyses in mouse models together have shown many interactions among the USH1 and USH2 proteins. In the inner ear, these interactions are essential for proper development of the hair bundle and may have a role in the mechanoelectrical signal transduction and synaptic function of mature hair cells.

A multiprotein scaffold complex model, with a central role for the PDZ domain containing protein homologs, harmonin and whirlin, and the SAM domain of Sans, has been proposed. There is evidence that harmonin and whirlin can bind all other components of the USH network, including CDH23, PCDH15, Usherin, VLGR1 and myosin VIIA.[11,88] These two proteins bind through one or more of their PDZ domains to either a C-terminal consensus class I PBM[85] or to internal PDZ-binding domains of their interaction partners. Binding of VLGR1,[8,95] USH2A isoform B[7,8,11] and PCDH15[7,8] to the PDZ domains of whirlin and/or harmonin has been shown to be dependent on their C-terminal class I PBM, whereas both a class I C-terminal PBM and an internal PBM with homology to the internal PBM of the adaptor protein RIL[100] are involved in the binding of CDH23.[96] Myosin VIIa does not have a C-terminal PBM, and its binding therefore relies on one or more not yet identified internal PBMs.[88] SANS does contain a conserved C-terminal class I PBM.[11] However, its binding to harmonin and/or whirlin does not seem to be affected by deletion of this binding domain, suggesting that one or more putative internal PBMs may be implicated in the binding. Myosin VIIa has also been found to interact with SANS[11] and PCDH15.[97]

THE USH PROTEINS INTERACT TO CONTROL THE MORPHOGENESIS OF HAIR CELL BUNDLES IN THE INNER EAR

Much of the knowledge regarding the function of USH protein was obtained from the study of USH mouse models. Mice that have defective myosin VIIa (shaker-1),[22] spontaneous mouse mutants[101] and targeted mouse models for harmonin,[102–104] CDH23 (*waltzer*),[105,106] PCDH15 (*Ames waltzer*),[107] Sans (*Jackson shaker*),[30] Whirlin (*whirler*),[72] transgenic *VLGR1*^{del7TM} mice[94] and mouse model for *Ush2A*$^{-/-}$mouse[108] have been reported. All the USH mouse models show severe hearing loss and vestibular dysfunction. Surprisingly, except the *Ush2A*$^{-/-}$ mouse, none shows signs of RP observed in USH patients.[108] Differences in the extent of molecular redundancy may explain why retinal degeneration occurs in humans but not in mice. Electron microscopy scanning analysis of the auditory sensory cells in mutant mice lacking any of the USH proteins revealed abnormalities in the structure and organization and, more specifically, malformations in hair bundle shape, which ultimately lead to degeneration of hair cells.

These findings suggest that the USH proteins form a transmembrane network that regulates hair bundle morphogenesis. Mutation in any of the USH genes will lead to failure of the USH complexes, which likely will cause USH.

In mouse models, the USH proteins have been localized to the developing auditory hair bundle, specifically the growing stereocilia or the kinocilium.[11,52,88,91,109–111] On the basis of the *in vitro* direct interaction between USH proteins and localization studies in mouse models, specific roles for each of the USH proteins have been suggested. In addition to its role in stereocilia bundle development,[112] Myo7a is believed to use long filaments of actin as tracks along which to transport other USH complex molecules.[113] In the USH 'interactome' model, it is proposed that the extracellular interstereocilia links are anchored intracellularly by the scaffold proteins, harmonin and whirlin, through direct binding to the actin cytoskeleton or through other proteins, including myosin VIIa, myosin XV, MYO1c and/or vezatin.[11,88,109,114–116] In addition, we and others have shown that the association of other proteins with the USH complex through harmonin indicates that the complex may be functionally linked to a number of basic cell-biological processes, including cell polarity and cell–cell interactions.[117,118] On the basis of the analysis of the *whirler* mutant and the expression pattern of whirlin in the inner ear, it was hypothesized that whirlin may coordinate F-actin growth.[72] Both CDH23 and PCDH15 are components of transient lateral links, kinociliary links and tip links in the inner ear sensory cells,[89,91,92,119,120] are therefore thought to be crucial for morphogenesis and mechanotransduction. The two proteins have a class I PDZ-binding domains, enabling them to bind harmonin and whirlin and as such anchor themselves to the actin cytoskeleton.[26,96] Interestingly, it has been recently shown that in mouse model for nonsyndromic deafness DFNB12 (*salsa*), a missense mutation in *Cdh23* affects only the mechanotransduction machinery of hair cells without effects on hair bundle development.[121] This finding may provide an explanation for the phenotype variations that are associated with mutations in USH genes. *In vitro* binding assays and colocalization study have shown that SANS physically interacts with harmonin and myosin VIIa; however, no direct binding could be found with Cdh23 and Pcdh15. Because of its high expression below the cuticular plate in hair cells, SANS is not considered as part of any of the interstereocilial link complexes, but rather has a role in trafficking molecules of these complexes.[10] Both usherin and VLGR1 are transiently expressed during early cochlear development at the ankle link that connect the stereocilia of hair cells at their base.[11,94] These two proteins are thus believed to be integral members of the transient ankle-link complex. The absence of ankle links transgenic mice carrying a mutation in *VLGR1 (VLGR1*^{del7TM}) further support involvement of *VLGR1* in a molecular complex associated with the ankle links.[94] A diagram of a hair bundle with location of USH proteins is shown in Figure 2.

CONCLUSION

USH is a group of clinically variable and genetically heterogeneous autosomal recessive syndromes. As USH results in the loss of the two most vital human senses, the burden to patients with this disorder is tremendous. During the past decade, remarkable progress has been made in the identification of the USH genes as well as in the elucidation of the pathogenesis of the syndrome. An important finding that has emerged from the studies is that the USH proteins

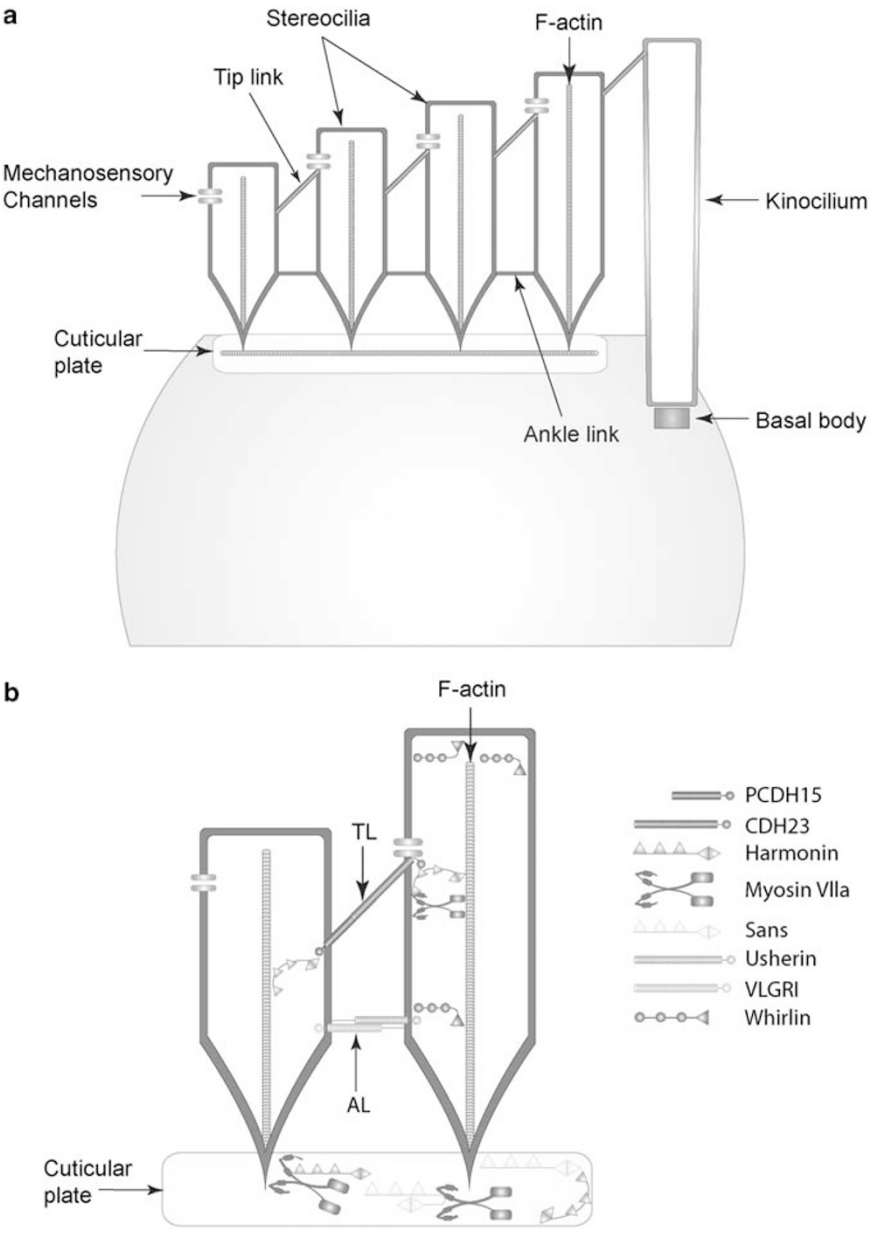

Figure 2 (**a**) Diagram of a developing hair bundle. Stereocilia are held together and to the kinocilium by diverse side-links. Tip links (TL) are thought to gate the mechanoelectrical transduction channel. (**b**) The diagram shows the localization of CDH23 and PCDH15 at tip links. The binding of harmonin B to CDH23, PCDH15 and F-actin could anchor the interstereocilia links to the stereocilia actin core. Myo7a is believed to use long filaments of actin as tracks along which to transport other USH complex molecules. Sans located below the cuticular plate may have a role in trafficking molecules of the USH complex. Both Usherin and VLGR1 are members of the ankle links (AL) that are tethered to the actin stereocilia core through the scaffold proteins whirlin and possibly harmonin B.

have a dual function in hair cell development and mechanotransduction. Interestingly, recent evidence suggests that USH mutations, which lead to nonsyndromic hearing loss, might affect only the mechanotransduction machinery of hair cells without effects on hair bundle development.[121] Understanding the cellular and molecular basis of phenotypic variation and pathogenesis of USH is essential in the progress toward discovery of new molecular targets for diagnosis, prevention and treatment of this debilitating disorder.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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