The epigenetic regulation of sensorineural deafness

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Abstract. Sensorineural deafness is a common defect and related to the structural and functional abnormality of the sensory nerve epithelial cells in the cochlea. A growing number of studies have found that epigenetic mechanisms, such as DNA methylation and histone modification, are also important factors in the occurrence of deafness in addition to genetic factors. This article summarizes recent findings concerning the role of epigenetics in deafness and the epigenetic strategies to restore hearing.

1. Introduction

The sensorineural deafness is a common defect in newborn infants and persons over 80 years of age. Statistics show that 360 million people worldwide have lost their hearing, 32 million of them are children, and about 60 to 70 percent of hearing loss is due to genetic factors [1].

The inner ear of mammals is one of the most complicated microscopic organs in the human body, consisting of the cochlea in charge of the hearing and the vestibule in charge of the balance, the loss of which will respectively cause the hearing loss and disorientation. The cochlear sensory epithelial cell (CSEC), vestibular supporting cell and mechanoreceptor cell are the polarized epithelial cells with cilia on the top surface, so they are called hair cells. Most hearing loss is due to abnormal development or degeneration of Cochlear hair cells. The development of inner ear requires the coordination for expression of genes and regulatory factors [2].

The epigenetic modifications, mainly including DNA methylation and histone modification, may play a significant role in development and cell differentiation. Recent studies show that the epigenetic is associated with deafness.

2. DNA Methylation

DNA methylation, addition of a methyl group on position 5 of cytosine residues of the duster of CpG dinucleotide (CpG Island), is catalyzed by DNA methyltransferases. The CpG island methylation of promoter region is typically associated with transcriptional repression. The methylation process is achieved mainly through recruiting the methyl-CpG-binding protein (MECP) to hinder the binding of transcription factors to promoter regions, or inducing the histone modification and chromatin remodeling to disturb the initiation or extension of the transcription. Meanwhile, the DNA methylation also has the significant effect in maintaining the DNA stability. Abnormal CpG methylation is closely connected with some genetic syndromes inclusive of hearing loss [3].

Stickler syndrome type 1 (STL1) is caused by the type-II collagen gene (COL2A1) mutation. The major representations include the progressive sensorineural hearing loss, progressive muscle disease; blindness resulted from the vitreous degeneration and retinal detachment, premature degenerative
change, epiphyseal dysplasia, osteoarthritis, partially abnormal facial features and cleft palate. Studies have shown that methylated CGA codon is a mutational hotspot in the COL2A1 gene [4].

The autosomal dominant cerebellar ataxia, deafness, and narcolepsy (ADCADN) and hereditary sensory neuropathy type IE (HSN1E) are resulted from the genetic heterozygous mutation of DNA methyltransferase DNMT1 [5]. The ADCADN is the late onset disease (at the age of 30-40), with the symptoms of narcolepsy, cataplexy, nerve deafness, optic atrophy, dystaxia, Alzheimer’s disease, psychosis and etc. The narcolepsy and deafness are the common original symptom, subsequently followed by the dystaxia [6]. The HSN1E patient will suffer from the sensory hearing loss resulted from the abnormality of inner ear, and hearing loss progressive aggravation. The deafness will become more serious with time. Generally, it will become the moderate or severe deafness at the age of 20-45 [7]. The DNMT1 mutation generally will cause the bilateral hearing loss, but the unilateral hearing loss is also reported sometimes.

The facioscapulohumeral muscular dystrophy 1A (FSHMD1A) is related to the abnormal methylation of repeated DNA sequences. The most prominent representation of such syndrome is the myasthenia resulted from the malnutrition. Generally, it starts from the face, shoulder and arm muscles, and finally ends at the buttocks and legs. Such syndrome also includes the sensorineural hearing loss and retinal microvascular abnormalities. In most cases, the patient will have the high frequent (mainly within 4000-6000HZ) hearing loss. The features of FSHMD1A are the low levels of DNA methylation and chromosome 4q35 regional tandem repeat polymorphism. Meanwhile, the severity of FSHMD1A syndrome is related to the methylation level, the patients with severe phenotype and early morbidity show more prominent D4Z4 hypomethylation [8].

3. Histone Modifications

A nucleosome is the basic structural unit of chromatin consisting of the core particle, linker DNA and histone H1. The core particle consists of 147bp of DNA wrapped around a core of eight histone protein (histone H2A-H2B dimers and H3-H4 dimers) [9]. Except for maintaining DNA folding, the histone also plays a significant role in the regulation of gene expression, DNA damage repair and other processes. DNA accessibility is affected by the methylation, acetylation, phosphorylation, ubiquitination and other post-transcriptional modifications of histone [10]. Most histone modifications are dynamic and reversible, and catalyzed by corresponding enzymes. Histone methylation and acetylation are catalyzed by histone methyltransferase (HMTs) and histone acetyltransferase (HATs) respectively, while histone demethylase (HDMs) and histone deacetylase (HADCs) remove the methyl and acetyl groups. Studies have found that the inheritance hearing loss is related to the mutations in genes that encode histone modification enzymes.

The Sotos syndrome is frequently related to conductive hearing loss, which has been shown to be caused by mutations in histone methyltransferase NSD1 [11]. The kleefstra syndrome is developmental disorder characterized by low intelligence, low muscle tone in childhood, unique facial features and sensorineural hearing loss. The heterozygous mutation in euchromatin histone lysine methyltransferase 1 (EHMT1), a component of E2F6 compound, inhibits gene transcription by methylating histone H3K9, causing Kleefstra syndrome [12]. The heterozygous mutation of lysine-specific methyltransferase 2D (KMT2D, namely, MLL2/MLL4) is related to the Kabuki syndrome. The features of Kabuki syndrome are unique facial features, mild to moderate mental retardation, microcephalus, muscular hypertonia, skeletal deformity, cardiac disorders and hearing loss [13]. The deafness is a common symptom of Kabuki syndrome and can be conductive and neurosensory properties or both. About 6% Kabuki syndrome case is resulted from the mutation of lysine specific demethylase 6A (KDM6A). The KDM6A will catalyze the H3K27me3, causing the occurrence of demethylation. Although the KMT2D and KDM6A have effects on different lysine residues, both can activate the transcription [14]. The mutation of lysine acetyltransferase 6B (KAT6B/MYST4) is related to the genitopatellar syndrome (GPS) and Say-Barber-Biesecker variant of Ohdo syndrome. The common phenotype of these KAT6B-related diseases include the global developmental delay/ mental retardation, hypotonia, cryptorchidism, patellar aplasia or other defects,
such as the congenital heart defects, altered dentition, hearing loss, thyroid dysfunction [15]. KAT6B is a histone acetyltransferase with transcriptional activation at the n-terminal and transcriptional inhibition at the c-terminal. The mutation causing the GPS occurs in the proximal portion of the last exon, and the expressed protein has no C-end structural domain. The mutation causing the Say-Barber-Bieseker variant of Ohdo syndrome occurs in the whole gene, mostly in the last exon at the distal end [16]. Chromodomain-helicase-DNA-binding protein 7 (CHD7) also plays a very significant role in the development of inner ear. The CHD7 mutation will cause the CHARGE syndrome. A significant clinical feature of CHARGE syndrome is dysplasia of the semicircular canal in the inner ear, which may enable us to further prevent the formation of the cochlea [17].

4. Epigenetics: Damage and Protection
Noise is one of the most common causes of hearing loss. In addition, many studies have shown that the cochlea sensory nerve epithelial cells are vulnerable to various damage of ototoxic drugs, such as the Aminoglycosides (e.g. Gentamicin), loop diuretic (e.g. Furosemide), Platinum-based emotherapy drugs (e.g. Cisplatin), and some Non-Steroid Anti-Inflammatory Drugs (NSAIDs), and etc. The formation of reactive oxygen species (ROS) is the molecular pathological mechanism of hair cells death caused by aminoglycoside antibiotics and cisplatin [18, 19]. The occurrence of ROS is related to the DNA damage, increased chromosome degradation and DNA methylation changes [20, 21]. Aminoglycosides may also induce histone deacetylase to be recruited to chromatin in mammalian hair cells [22]. How to prevent hair cell loss effectively is an urgent problem in medicine.

Histone methylation, as an important epigenetic modification, is involved in the regulation of gene expression, development and injury response. Hair cell injury is usually associated with a rapid increase in H3K9me2. The specific inhibitor bix01294 and uncle0638 of histone methyltransferase G9a/GLP can reduce the H3K9me2 level, and further prevent from the hair cell death [23]. HDACs inhibitors act as anticancer agents, and some have been FDA approved for use in specific types of cancer treatments. However, Broad-spectrum and specific HDACs inhibitors have a concentration-dependent protective effect on inflammatory, neurodegenerative, and oxidative stress models [24]. In the inner ears, HDACs inhibitors may have protective effects against the aminoglycosides damage.

Cisplatin is a widely used chemotherapeutics drug, but its major side effect is ototoxicity. Epigenetic-related drugs, such as lysine specific demethylase 1 (LSD1) inhibitors, restrain the demethylation of histone H3K4 (primarily H3K4me2) to prevent cisplatin-induced hair cell loss. The research results show that LSD1 inhibitors prevent the spiral ganglion neurons injuries caused by cisplatin, through regulating the demethylation of histone H3K4 and preventing the enhancement of ROS level, which may become a potential strategy for the treatment of cisplatin-induced hearing loss [25]. Down-expression of LSD1 expression inactivates the Wnt/β-catenin and FGF signaling pathway, which will further significantly reduce the hair cell regeneration after loss caused by neomycin.

5. Outlook
The epigenetics is an important mechanism of cell development and reprogramming. The study and application of the epigenome will be of great significance in preventing and treating the deafness. Meanwhile, the epigenetics will also provide a direction for studying how drugs and noises affect the gene regulation network and how to avoid the damage of drugs and noise to hearing. The results of epigenetic studies may enable us to further understand the gene regulation network of the development of sensory nerve epithelial hair cells in the cochlea.

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References

[1] W.E. Nance. The genetics of deafness. Ment Retard Dev Disabil Res Rev, 9, 2: 109–119 (2003)
[2] M.W. Kelley. Hair cell development: commitment through differentiation. Brain Res, 1091, 1: 172–185 (2006)
[3] M.J. Provenzano, F.E. Domann. A role for epigenetics in hearing: establishment and maintenance of auditory specific gene expression patterns. Hear Res, 233, 1-2: 1–13 (2007)
[4] D.J. Wilkin, R. Liberfarb, W.G. Cole, et al. Rapid determination of COL2A1 mutations in individuals with Stickler syndrome: analysis of potential premature termination codons. Am J Med Genet, 94, 2: 141–148 (2000)
[5] P.G. Overveld, L. Enthoven, L. Felicetti, et al. Variable hypomethylation of D4Z4 in facioscapulohumeral muscular dystrophy. Ann Neurol, 58, 4: 569–576 (2005)
[6] Z. Sun, Y. Wu, X. Duan, et al. Aberrant signature methylome by DNMT1 hot spot mutation in hereditary sensory and autonomic neuropathy 1E. Epigenetics, 9, 8: 1184–1193 (2014)
[7] J. Winkelmann, L. Lin, G. Plazzi, et al. Mutations in DNMT1 cause autosomal dominant cerebellar ataxia, deafness and narcolepsy. Hum Mol Genet, 21, 10: 2205–2210 (2012)
[8] C.J. Klein, M.V. Botuyan, S. Hammans, et al. Mutations in DNMT1 cause hereditary sensory neuropathy with dementia and hearing loss. Nat Genet, 43, 6: 595–600 (2011)
[9] K. Luger, A.W. Mäder, T.J. Richmond, et al. Crystal structure of the nucleosome core particle at 2.8 Å resolution. Nature, 389, 6648: 251-260 (1997)
[10] G.E. Zentner, S. Henikoff. Regulation of nucleosome dynamics by histone modifications. Nat Struct Mol Biol, 20, 3: 259-266 (2013)
[11] K. Tatton-Brown, A. Murray, S. Banka, et al. Weaver syndrome and EZH2 mutations: clarifying the clinical phenotype. Am J Med Genet, 161: 2972–2980 (2013)
[12] M. Tachibana, J. Ueda, H. Iwanari, et al. Histone methyltransferases G9a and GLP form heteromeric complexes and are both crucial for methylation of euchromatin at H3–K9. Genes Dev, 19, 7: 815–826 (2005)
[13] S.B. Ng, A.W. Bigham, M.J. Mcmillin, H.I. Gildersleeve, et al. Exome sequencing identifies MLL2 mutations as a cause of Kabuki syndrome. Nat Genet, 42, 9: 790–793 (2010)
[14] J.E. Lee, C. Wang, X. Feng, et al. H3K4monoand di-methyltransferase MLL4 is required for enhancer activation during cell differentiation. Elife, 2: e01503 (2013)
[15] A.C. Campos, F. Molognoni, V. D’almeida, et al. Oxidative stress modulates DNA methylation during melanocyte anchorage blockade associated with malignant transformation. Neoplasia, 9, 12: 1111–1121 (2007)
[16] E.I. Campos, D. Reinberg. Histones: annotating chromatin. Annu Rev Genet, 43: 559–599 (2009)
[17] W.S. Layman, E.A. Hurd, D.M. Martin, et al. Chromodomain proteins in development: lessons from CHARGE syndrome. Clin. Genet, 78, 1: 11–20 (2010)
[18] A.G. Cheng, L.L. Cunningham, E.W. Rubel, et al. Mechanisms of hair cell death and protection. Curr Opin Otolaryngol Head Neck Surg, 13, 6: 343–348 (2005)
[19] J. Schacht, A.E. Talaska, L.P. Rybak, et al. Cisplatin and aminoglycoside antibiotics: hearing loss and its prevention. Anat. Rec. (Hoboken), 295, 11: 1837–1850 (2012)
[20] K.V. Donkena, C.Y. Young, D.J. Tindall, et al. Oxidative stress and DNA methylation in prostate cancer. Obstet Gynecol Int, 2010: 302051 (2010)
[21] D. Ziech, R. Franco, A. Pappa, M.I. Panayiotidis, et al. Reactive oxygen species (ROS)–induced genetic and epigenetic alterations in human carcinogenesis. Mutat Res, 711, 1-2: 167–173 (2011)
[22] F.Q. Chen, J. Schacht, S.H. Sha, et al. Aminoglycoside-induced histone deacetylation and hair cell death in the mouse cochlea. J Neurochem, 108, 5: 1226–1236 (2009)
[23] H. Yu, Q. Lin, H. Li, et al. Inhibition of H3K9 methyltransferases G9a/GLP prevents ototoxicity and ongoing hair cell death. Cell Death Dis, 214: e506 (2013)
[24] C. Robert, F.V. Rassool. HDAC inhibitors: roles of DNA damage and repair. Adv. Cancer Res,
[25] A. Li, Y. He, H. Li, et al. Lysine-specific demethylase 1 inhibitors protect cochlear spiral ganglion neurons against cisplatin-induced damage. Neuroreport, 26, 9: 539-47 (2015)