Mutation analysis and audiologic assessment in six Chinese children with primary distal renal tubular acidosis

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Abstract

The objective of this study is to identify ATP6V1B1, ATP6V0A4 and SLC4A1 genes mutations and assess audiologic characteristics in six Chinese children with primary distal renal tubular acidosis from four unrelated families between the ages of 2 and 13 years. Both ATP6V0A4 and ATP6V1B1 genes were preferentially screened in all index cases by direct sequence analysis. If inconclusive then SLC4A1 gene should be analyzed for mutation. Their clinical features, hearing status and inner ear imaging structure were also investigated. Six loss-of-function mutations were identified in six patients. Two novel mutations were identified in either of ATP6V0A4 and ATP6V1B1 genes, respectively. Two probands from different kindreds with mutations in ATP6V1B1 presented early onset profound sensorineural hearing loss (SNHL) and enlarged vestibular aqueduct (EVA). Two from different families carrying ATP6V0A4 mutations manifested early onset moderate mixed HL and moderate SNHL, respectively, the former comorbid with EVA, while the latter not; however, both their elder sisters showed normal hearing and inner ear. These findings expand the spectrum of mutations in the ATP6V0A4 and ATP6V1B1 genes associated with primary dRTA. Our study confirms the association of EVA and mutations in either of these two genes. More studies are necessary to clarify the relationship between dRTA, SNHL, EVA, and gene mutations.

Keywords

Distal renal tubular acidosis, enlarged vestibular aqueduct, gene mutation, hearing loss, ATP6V0A4 gene, ATP6V1B1 gene

Introduction

Distal renal tubular acidosis (dRTA) results from the impaired secretion of hydrogen ions from the distal nephron, causing metabolic acidosis, often with hypokalemia, nephrocalcinosis, and/or nephrolithiasis.¹⁻³

Both autosomal dominant (AD) and autosomal recessive (AR) inheritance patterns have been reported in primary dRTA. Mutations in the ATP6V1B1 and ATP6V0A4 genes, encoding subunits B1 and a4 of apical H⁺-ATPase, cause recessive forms of dRTA.¹⁻⁶ ATP6V1B1 mutations have been associated with early sensorineural hearing loss (SNHL, OMIM 267300), whereas ATP6V0A4 mutations are classically associated with either late-onset SNHL or normal hearing (OMIM 602722). Enlarged vestibular aqueduct (EVA) was described in patients with recessive dRTA and SNHL, and recently, this abnormality has been associated with mutations in the ATP6V1B1 gene.⁷ However, there is more and more evidence that these two recessive forms of dRTA are genetically more heterogeneous in the auditory phenotype than previously assumed. A few cases harbored ATP6V1B1 mutations without SNHL have been described.⁸⁻⁹ Recent genetic analyses have revealed that some individuals with mutations in the ATP6V0A4 gene also have early-onset severe SNHL.⁹ And most recently, Andreucci et al. described a child carrying ATP6V0A4 mutations with early profound SNHL and unexpected EVA.¹⁰ Furthermore, this auditory phenotype of severe hearing impairment and enlarged endolymphatic compartments was verified by an Atp6v0a4 knockout mouse model.¹¹ Mutations in the SLC4A1 gene encoding the human AE1 have been associated with either AD or AR dRTA.¹²,¹³ Additionally, the presence of SNHL in a patient who showed no mutation in ATP6V1B1 and ATP6V0A4 genes do not exclude a priori the possibility of a form of dRTA related to a mutation in SLC4A1 gene, as the development of SNHL may be the consequence of many different causes, such as other genetic factors, ototoxic agents, neonatal distress and electrolyte disequilibrium. Therefore, lack of definite phenotype-genotype correlation asks for the need of genetic analysis in dRTA patients.

Herein, we performed mutation analysis in six Chinese patients from four unrelated kindreds with dRTA. On the other hand, their audiologic phenotype and genotype correlations were also investigated.
Materials and methods

Patients

We studied six children (three females and three males) with dRTA belonging to four unrelated families; their clinical features and representative biochemical data are shown in Table 1. Diagnosis was based on the presence of the following necessary criteria: Metabolic acidosis with a normal anion gap and inappropriate alkaline urine (pH > 6.0) in a context of acidosis (CO₂CP < 18 mmol/L); normal fractional excretion rate of bicarbonate (FEHCO₃, normal value < 5%), the FEHCO₃ is calculated after adequate alkalization; and/or nephrocalcinosis. Additional optional criteria were hypercalciuria, hypokalemia, polyuria, and failure to thrive. Audiological data and inner ear radiologic finding of all patients were collected. Hearing was assessed by pure-tone audiometry (PTA) and/or automated auditory brainstem response (AABR) test. The severity of hearing loss was ranked as mild (20–40 dB), moderate (41–55 dB), moderately severe (56–70 dB), severe (71–90 dB) and profound (91 dB or greater). In addition, medical records were reviewed to document the results of hearing screening tests that children may have received. Screening devices, test parameters, and test results are listed in Table 2. The parents of the patients in families II and III were consanguineous. Patients III-1 and III-2, and patients IV-1 and IV-2 were siblings. The study protocol was approved by the Ethics Committee on Human Studies at the affiliated hospital of medical college Qingdao University. Informed consent was obtained from each patient and his or her parents.

Mutation analysis

Both ATP6V0A4 and ATP6V1B1 genes were preferentially screened in all index cases, if inconclusive (no mutation or only one was identified in either gene) then SLC4A1 gene should be analyzed for mutation. Genomic DNA was extracted from peripheral blood of these six patients, their family members, and 100 normal healthy controls by the GenElute blood genomic DNA kit (Sigma, NA2010, St. Louis, MO). Twenty pairs of oligonucleotide primers were generated to amplify all coding exons (from exon 3 to Table 1. Clinical characteristic at diagnosis of six children with distal renal tubular acidosis.

<table>
<thead>
<tr>
<th>Patient</th>
<th>I-1</th>
<th>II-1</th>
<th>III-1</th>
<th>III-2</th>
<th>IV-1</th>
<th>IV-2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender</td>
<td>F</td>
<td>M</td>
<td>F</td>
<td>M</td>
<td>F</td>
<td>M</td>
</tr>
<tr>
<td>Current age (years)</td>
<td>2</td>
<td>3</td>
<td>11</td>
<td>8</td>
<td>13</td>
<td>11</td>
</tr>
<tr>
<td>Age at diagnosis (weeks)</td>
<td>7</td>
<td>13</td>
<td></td>
<td>22</td>
<td>4</td>
<td>17</td>
</tr>
<tr>
<td>Manifestation</td>
<td>Pneumonia, vomiting</td>
<td>Vomiting, dehydration</td>
<td>Growth retardation</td>
<td>Family study</td>
<td>Dehydration</td>
<td>Family study</td>
</tr>
<tr>
<td>Height (cm) (50th percentile of Chinese children normal reference values for height and weight of different ages and genders at diagnosis)</td>
<td>54.0 (56.0)</td>
<td>59.0 (61.5)</td>
<td>58.0 (64.5)</td>
<td>52.5 (54.5)</td>
<td>62 (62.2)</td>
<td>56.5 (59.0)</td>
</tr>
<tr>
<td>Weight (kg) (50th percentile of Chinese children normal reference values for height and weight of different ages and genders at diagnosis)</td>
<td>4.0 (4.7)</td>
<td>5.4 (6.3)</td>
<td>5.7 (7.1)</td>
<td>4.2 (4.5)</td>
<td>5.8 (6.2)</td>
<td>4.8 (5.5)</td>
</tr>
<tr>
<td>Blood pH</td>
<td>7.32</td>
<td>7.31</td>
<td>7.26</td>
<td>7.20</td>
<td>7.23</td>
<td>7.25</td>
</tr>
<tr>
<td>SHCO₃</td>
<td>13.9</td>
<td>14.2</td>
<td>12.0</td>
<td>9.9</td>
<td>11.0</td>
<td>10.0</td>
</tr>
<tr>
<td>SK</td>
<td>3.3</td>
<td>3.1</td>
<td>2.8</td>
<td>3.1</td>
<td>4.2</td>
<td>2.6</td>
</tr>
<tr>
<td>Ionized Sc (mmol/L, 1.12–1.23)</td>
<td>1.42</td>
<td>1.22</td>
<td>1.20</td>
<td>1.23</td>
<td>1.62</td>
<td>1.19</td>
</tr>
<tr>
<td>GFR (mL/min/1.73 m²) (50th percentile of Chinese children normal reference values for height and weight of different ages and genders at diagnosis)</td>
<td>26.5</td>
<td>26.7</td>
<td>23.5</td>
<td>31.1</td>
<td>27.6</td>
<td>28.1</td>
</tr>
<tr>
<td>UpH</td>
<td>7.5</td>
<td>8.0</td>
<td>8.5</td>
<td>7.5</td>
<td>8.0</td>
<td>8.5</td>
</tr>
<tr>
<td>UCa/Cr (mg/mg) (setting threshold at 35 dB)</td>
<td>0.53</td>
<td>0.68</td>
<td>0.71</td>
<td>0.93</td>
<td>0.77</td>
<td>0.89</td>
</tr>
<tr>
<td>FEHCO₃ (%)</td>
<td>4.0</td>
<td>3.5</td>
<td>NA</td>
<td>4.8</td>
<td>3.7</td>
<td>2.8</td>
</tr>
</tbody>
</table>

Nephrocalcinosis | 2 | 2 | 3 | 1 | 2 |

Notes: S, serum; U, urinary; FEHCO₃, fractional excretion of bicarbonate; NA, not available.

Table 2. Hearing status and inner ear radiologic characteristics of these children with distal renal tubular acidosis.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age at confirmation of hearing loss (months)</th>
<th>SNHL</th>
<th>EVA</th>
<th>Therapy</th>
<th>Screening device</th>
<th>Type of UNHS</th>
<th>Results of UNHS</th>
</tr>
</thead>
<tbody>
<tr>
<td>I-1</td>
<td>9</td>
<td>Bilateral profound</td>
<td>Positive (MRI at 12 months)</td>
<td>Cochlear implants</td>
<td>GSI AUDIO screener</td>
<td>DPOAE</td>
<td>Bilateral pass</td>
</tr>
<tr>
<td>II-1</td>
<td>7</td>
<td>Bilateral profound</td>
<td>Positive (CT at 12 months)</td>
<td>Cochlear implants</td>
<td>GSI AUDIO screener</td>
<td>DPOAE</td>
<td>Unilateral pass</td>
</tr>
<tr>
<td>III-1</td>
<td>37</td>
<td>No</td>
<td>Negative (MRI at 11 years)</td>
<td>Hearing aids</td>
<td>NATUS ALGO 3i</td>
<td>AABR</td>
<td>Bilateral pass</td>
</tr>
<tr>
<td>III-2</td>
<td>37</td>
<td>Bilateral moderate</td>
<td>Positive (MRI &amp; CT at 3 years)</td>
<td>Hearing aids</td>
<td>NATUS ALGO 3i</td>
<td>AABR</td>
<td>Bilateral pass</td>
</tr>
<tr>
<td>IV-1</td>
<td>37</td>
<td>No</td>
<td>Negative (MRI at 7 years)</td>
<td>Hearing aids</td>
<td>ILO v6</td>
<td>TEOAE</td>
<td>Bilateral pass</td>
</tr>
<tr>
<td>IV-2</td>
<td>60</td>
<td>Bilateral moderate</td>
<td>Negative (MRI at 5 years)</td>
<td>Hearing aids</td>
<td>ILO v6</td>
<td>TEOAE</td>
<td>Bilateral pass</td>
</tr>
</tbody>
</table>

Notes: SNHL, sensorineural hearing loss; EVA, enlarged vestibular aqueduct; UNHS, universal newborn hearing screening; MRI, magnetic resonance imaging; CT, computed tomography; DPOAE, distortion product otoacoustic emission; AABR, automated auditory brainstem response; TEOAE, transient evoked otoacoustic emissions.
and flanking intronic regions of the ATP6V0A4 gene (Supplemental Table 1). The coding region and adjacent intronic segments of the ATP6V1B1 and SLC4A1 genes were amplified using previously described primer pairs, respectively.\textsuperscript{2,14} PCRs were performed in 50 \mu L of solution containing 0.2 mM dNTP, 0.03 U/\mu L Taq polymerase (Takara EX Taq Hot start version, DRR006B, Osaka, Japan), 2.0 mM MgCl\(_2\), 2.5 \mu L 10 \times PCR Mg\(^{2+}\)-free buffer (Takara), approximately 50 ng genomic DNA and 1 mM of each primer. PCR was performed with an initial denaturation step at 95°C for 5 min, subsequently followed by 33 cycles with denaturation at 95°C for 45 s, annealing at 52–66°C for 45 s and elongation at 72°C for 45 s. PCR samples were subjected to bidirectional sequencing. The sequence reactions were run on an ABI Prism 3730 DNA Analyzer (Applied Biosystems, Carlsbad, CA).

**Results**

**Clinical findings**

A total of six children from four unrelated kindreds were enrolled in this study. Table 1 summarizes the clinical and biochemical manifestations leading to the diagnosis of dRTA in each patient. As shown in Table 1, all children showed nephrocalcinosis confirmed by abdominal ultrasonography, however, only two of six presented hypercalciuria. In addition, two of six children had hypercalcemia (patient I-1 and IV-1). Of both with hypercalcemia, hyperparathyroidism and vitamin D intoxication were excluded because of normal values of intact parathyroid hormone, 1,25(OH)-vitamin D and 25-OH-vitamin D. The calcium level of patient IV-1 normalized in the first 72 h with correction of metabolic acidosis and dehydration, whereas this similar phenomenon did not occur in patient I-1 who kept a mild elevated calcium level for nearly 1 year.

Alkali therapy and potassium supplement (sodium citrate and potassium citrate) was effective for metabolic acidosis, hypokalemia and delayed growth in all patients, with this therapy improving their growth to almost normal range.

**Mutation analysis**

Three variants including two novel mutations in the ATP6V1B1 gene were identified in two of the four kindreds (Figure 1A, Table 3). Patient I-1 was a compound heterozygote with a recurrent duplication of c.1155dupC (p.Ile386Hisfs*56)\textsuperscript{1,3,5,9} and a novel deletion of c.1356delT. This novel deletion led to a frameshift that resulted in premature termination of the protein at codon 486 (p.Phe452Leufs*35). Patient II-1 showed homozygosity for a novel splicing variant of c.786-1G\textsuperscript{4}C (loss of splice acceptor site of intron 8).

We also identified three distinct mutations including two novel aberrations in ATP6V0A4 gene in the other two families (Figure 1B, Table 3). A novel homozygous nonsense variant c.C580\textsuperscript{4}T (p.R194*) was revealed in both the sister III-1 and the brother III-2. While a novel splicing variant c.2258-1G\textsuperscript{4}C (loss of splice acceptor site of intron 21) and a...
previously reported mutation c.2137delG (p.Glu713fs*53) were identified in the sister IV-1 and the brother IV-2, both parents were heterozygous for the two mutations. Direct sequencing analysis failed to find above-mentioned mutations in 100 unrelated healthy subjects.

We did not perform the SLC4A1 gene analysis since two definite variants were found in each patient.

**Audiological phenotype and genotype correlation**

Table 2 presented the hearing status of all patients. Following the mutation testing, both patient I-1 and II-1 who harbored ATP6V1B1 gene mutations underwent a full confirming test of hearing status at the age of 9 months and 7 months, respectively. No ABR and no emissions could be elicited in both ears of them, and auditory multiple-frequency steady state evoked response (ASSR) test showed that both patients had very little residual hearing restricted to the low frequencies in both ears. And the normal Type ‘‘A’’ tympanogram suggested their normal middle ears function. Based on the above findings, both patient I-1 and II-1 was diagnosed with profound SNHL. The cerebral magnetic resonance imaging (MRI) scan performed at the age of 1 year showed dilation of the endolymphatic sac in patient I-1, while the high-resolution computed tomography (CT) of temporal bone carried out at the same age demonstrated EVA in patient II-1. Subsequently, both patients underwent cochlear implantation which helped to bring the achievement of almost normal spoken language development during the follow-up period. Patient III-1 has had no audio graphic evidence of HL to date; her brother who possesses the same mutation in the ATP6V0A4 gene presented with early-onset bilateral moderate HL however, which was confirmed by ABR at age 3 years. High-resolution CT and MRI of his temporal bones showed bilateral EVA with no other inner and middle ear malformations (Figure 2). Since then, he had begun to use bilateral hearing aids. During the follow-up period, from the age of 5 to the age of 8 years, he experienced four times of attacks of rotatory vertigo, and his hearing decreased gradually with fluctuating exacerbation which was associated with common cold infections. PTA demonstrated that he suffered from bilateral mixed HL characterized by air-bone gaps at lower frequencies, and his right pure-tone averages (PTAs, PTAs is calculated from the audiometric thresholds at the frequencies of 500, 1000, and 2000 Hz) deteriorated from 56.3 dB to 78.3 dB, while his left PTAs worsened from 63.3 dB to 80 dB (Figure 3A and B). Patients IV-1 and IV-2 are also siblings and harbor the same ATP6V0A4 mutations. However, only the little brother (patient IV-2) manifested with early-onset bilateral moderate SNHL which was verified with PTA at age 5 years (Figure 3C), while his 13-year-old sister (IV-1) has had no audiologic evidence of HL up to now. No abnormal middle and inner ear imaging findings was found by the cerebral MRI scan performed at the age of 7 years in sister and at the age of 5 years in brother, respectively. And the bilateral PTAs of patient IV-2 maintained stable throughout the follow-up period of 6 years.

Table 3. Detected mutations in ATP6V1B1 and ATP6V0A4 of the six children with distal renal tubular acidosis.

<table>
<thead>
<tr>
<th>Patient</th>
<th>ATP6V1B1(^a) gene mutation</th>
<th>ATP6V0A4(^b) gene mutation</th>
</tr>
</thead>
<tbody>
<tr>
<td>I-1</td>
<td>c.[1155dupC];[1356delT] (p.[Ile386Hisfs<em>56];[Phe452Leufs</em>35])</td>
<td></td>
</tr>
<tr>
<td>II-1</td>
<td>c.[786-1G&gt;C];[786-1G&gt;C] (r.[sp];[sp])</td>
<td>c.[C580&gt;T];[C580&gt;T] (p.[Arg194*];[Arg194*])</td>
</tr>
<tr>
<td>III-1, III-2</td>
<td>c.[C580-4T];[C580-4T] (p.[Arg194*];[Arg194*])</td>
<td>c.[2258-1G&gt;C];[2137delG] (r.[sp];[p.Glu713fs*53])</td>
</tr>
</tbody>
</table>

Notes: \(^a\)Nucleotides numbered according to the sequence in GenBank NM_001692.

\(^b\)Nucleotides numbered according to the sequence in GenBank NM_020632.
Among the six children, all of them received newborn hearing screening test, five passed hearing screening in both ears, and one passed the screening in only one ear (patient II-1). Among the patients with HL, 50% of patients (1/2) with ATP6V1B1 mutations and 100% of patients (2/2) with ATP6V0A4 variants passed the screening test in both ears at birth.

Discussion
We have identified three ATP6V1B1 mutations (one duplication, one deletion, and one splicing mutation) and three ATP6V0A4 mutations (one deletion, one nonsense, and one splicing mutation) in patients with primary dRTA. All of the six above mentioned mutations may result in translational frame shifting, truncated proteins, or unstable mRNA, causing the abnormal function of H⁺-ATPase and dRTA. These findings expand the spectrum of mutations in the ATP6V0A4 and ATP6V1B1 genes associated with primary dRTA, especially in Chinese populations. It is worth noting that the mutation p.Ile386Hisfs*56 observed in one of our four kindreds has been previously described in 21 families, of which 16 are from North Africa. Our data may provide more evidence and information to confirm that p.Ile386Hisfs*56 is a frequent or potential ‘‘hotspot’’ mutation. However, we could not exclude the possibility of a founder effect for this mutation because it might have transferred to China by population migration via Silk Road from North Africa and West Asia more than 1000 years ago. This study is the first report of ATP6V1B1 mutations in Chinese.

Hypercalciuria is a characteristic biochemical finding in patients with dRTA, however, four of six patients were normocalciuric at the time of diagnosis in this study. In fact, normocalciuria is not uncommon at diagnosis in young dRTA patients (<1 year). As we all know, many factors can contribute to the differences in urinary calcium excretion, such as dietary, calcium absorption and hormonal control, as well as renal handling of calcium. Three (patient I-1, II-1 and IV-1) of the four patients with normocalciuria had evident signs of dehydration resulted by vomiting, fever, or other causes at diagnosis in this study. Extracellular volume contraction is a well-known stimulus for para-cellular calcium reabsorption in the proximal tubules. Therefore, volume contraction may be an important cause for normocalciuria in them. In addition, dietary sodium is an important modulator of urinary calcium excretion and a close correlation exists between urinary sodium and urinary calcium excretion. Low salt intake is associated with decreased urinary calcium excretion. Therefore, we suppose that low dietary salt intake may help to explain their normal urinary calcium excretion. Furthermore, dietary sodium restriction may aggravate volume depletion and augment hypocaliuria.

Undoubtedly, hypercalciuria is a major contributor to nephrocalcinosis in dRTA. It may seem paradoxical that nephrocalcinosis was confirmed in all of our four children with normocalciuria at diagnosis. This may demonstrate that nephrocalcinosis can occur in patients with dRTA without hypercalciuria. However, we would rather believe that normocalciuria may merely stand for a status of patients with dRTA in a dynamic changing process. Tsai HY et al. proposed a dynamic mode of calcium excretion in patients with dRTA which well explained this ambivalence. At the early stage, metabolic acidosis promotes bone demineralization and hypercalciuria. Hypercalciuria, persistently high urine pH and profound hypocitraturia predispose the development of the nephrocalcinosis. With time, polyuria, renal sodium wasting and poor intake result in volume depletion, especially in young babies with low salt intake. Then volume contraction, low salt content in infant formula, and alkaline urine lead to the increased calcium reabsorption and normocalciuria.

Nevertheless, it should not be neglected that ethnicity and genetic polymorphism may be another determinant factor in the difference of urinary calcium excretion. It is well known...
that the normal spot urine calcium/creatinine (UCa/Cr) ratio has a significantly negative correlation with age. Sargent's group proposed the 95th percentile of normal reference values for UCa/Cr was 0.86 mg/mg for Caucasian infants less than 7 months. However, subsequent investigations have shown that the UCa/Cr may vary with ethnicity, geographic area and genetic polymorphism. There is evidence that the UCa/Cr ratio in Chinese adolescents is low when compared with published values for Caucasian children. Regrettably, there are no accurate reference values of UCa/Cr for Chinese infants. So the possibility of that hypercalciuria was inappropriately labeled as normocalciuria could not be completely excluded.

We found that two (patient I-1 and IV-1) of six patients presented with hypercalcemia. It is known that hypercalcemia is associated with dRTA not only in neonates but also in other ages. A combination of increased calcium reabsorption at proximal tubules due to a decrease in glomerular filtration rate (GFR) secondary to severe dehydration, systemic acidosis causing calcium chelation from bones have been postulated as possible mechanisms for hypercalcemia associated with dRTA. The fact that the abnormality of hypercalcemia was rapidly corrected with the rectification of systemic metabolic acidosis and dehydration in patient IV-1 may add weight to this hypothesis. However, even after acid–base disturbance and volume depletion was corrected, the patient I-1 presented sustained hypercalcemia in the absence of excessive vitamin D therapy and primary hyperparathyroidism. The hypercalcemia can be explained by a disequilibrium in which the intake of calcium exceeds the internal buffering capacity of the bone and the renal capacity to excrete calcium. However, further studies will be required to better define the mechanisms and possible etiological factors.

In this study, the association of EVA with ATP6V0A4 was observed for the second time, which in conjunction with the evidence from Kcc4+/− mice confirmed such connection. However, the early onset HL is not always strictly related to EVA in patients with ATP6V0A4 mutations, as can be observed in our patients. The exact pathogenetic mechanism of EVA remain unclear, a recent study demonstrated that the lack of endocochlear potential and disruption of endolymph pH homeostasis due to the dysfunction of H+-ATPases which expressing on the endolymphatic sac epithelium may play an important role. EVA, as an anomaly of the structures of inner ear, which might lead to SNHL or mixed HL, and manifest with normal hearing, congenital total deafness, progressive HL, or fluctuating HL. In addition, air-bone gap in the audiogram, especially at the lower frequencies, is one of the predominant features of EVA. In this investigation, there is no noticeable difference can be seen between the characteristics of EVA in our primary dRTA patients and that of other common causes of EVA. The loss of function of H+-ATPases in the inner ear is undoubtedly the basic reason of EVA and HL; however, EVA may play a modificatory role to the manifestation of HL. More studies are necessary to clarify the relationships between dRTA, SNHL, EVA, and gene mutations.

It is not clear when hearing loss due to an ATP6V1B1 or ATP6V0A4 mutation begins and how much residual hearing remains at birth, because the level of hearing in these patients at birth has not been studied. We found that one out of two HL patients with ATP6V1B1 mutations and both HL patients with ATP6V0A4 variants passed the screening test in both ears at birth. These data suggest that many patients with ATP6V1B1 or ATP6V0A4 mutations have better hearing than the UNHS threshold (~35 dB) at birth. This result is similar to the investigation on newborns with PDS mutations. As to the hereditary HL, the appropriate early intervention service is important to the development of speech and language, especially pre-lingual deafness. We think that it may be wise to implement repetitive screenings at preverbal and preschool ages in children with primary dRTA. However, in the light of our small sample size, investigations based on more subjects are needed to verify our conclusions.

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Declaration of interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the article. This work was supported by a grant from the National Natural Science Foundation (81170653), a Shandong province Science & Technology Development Plan (BS2010YY011), as well as Qingdao Science & Technology Development Plans (12-1-4-2-21-jch &13-1-3-40-nsh).

References


Supplemental material available online

Supplemental Table 1