Prevalence and Characteristics of STRC Gene Mutations (DFNB16): A Systematic Review and Meta-Analysis

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Background: Mutations in the STRC (MIM 606440) gene, inducing DFNB16, are considered a major cause of mild–moderate autosomal recessive non-syndromic hearing loss (ARNSHL). We conducted a systematic review and meta-analysis to determine the global prevalence and characteristics of STRC variations, important information required for genetic counseling.

Methods: PubMed, Google Scholar, Medline, Embase, and Web of Science were searched for relevant articles published before January 2021.

Results: The pooled prevalence of DFNB16 in GJB2-negative patients with hearing loss was 4.08% (95% CI: 0.0289–0.0573), and the proportion of STRC variants in the mild–moderate hearing loss group was 14.36%. Monoallelic mutations of STRC were 4.84% (95% CI: 0.0343–0.0680) in patients with deafness (non-GJB2) and 1.36% (95% CI: 0.0025–0.0696) in people with normal hearing. The DFNB16 prevalence in genetically confirmed patients (non-GJB2) was 11.10% (95% CI: 0.0716–0.1682). Overall pooled prevalence of deafness–infertility syndrome (DIS) was 36.75% (95% CI: 0.2122–0.5563) in DFNB16. The prevalence of biallelic deletions in STRC gene mutations was 70.85% (95% CI: 0.5824–0.8213).

Conclusion: Variants in the STRC gene significantly contribute to mild–moderate hearing impairment. Moreover, biallelic deletions are a major feature of STRC mutations. Copy number variations associated with infertility should be seriously considered when investigating DFNB16.

Keywords: STRC gene, mutation, deafness, prevalence, meta-analysis

INTRODUCTION

According to the World Health Organization, one in five people worldwide lives with hearing loss (HL)—5.5% of the population of the world (World Health Organization, 2021). Hearing problems can have a devastating impact on the mental health of the people and the ability to communicate, study, and even earn a living. Approximately half the cases of deafness have a genetic etiology (Sheffield and Smith, 2019). Although variations in gap junction protein beta 2 (GJB2)
gene are the most common factor for prelingual, recessive deafness (50%), stereocilin (STRC) gene, known as DFNB16, is supposed to be another major contributor to bilateral mild-to-moderate hearing impairment (HI) (Francey et al., 2012; Yokota et al., 2019). Moreover, STRC mutations are considered the second most frequent cause associated with autosomal recessive non-syndromic hearing loss (ARNSHL) (Sloan-Heggen et al., 2016; Plevova et al., 2017; Back et al., 2019; Cada et al., 2019).

The STRC gene is located on chromosome 15q15.3 and named after its encoded protein—stereocilin—an extracellular structural protein expressed in the outer hair cells (OHCs) of the inner ear. Stereocilin was detected in six sensory areas in the inner ears of mice: the organ of Corti, the utricular maculae, the saccular maculae, and the three cristae ampullares of the vestibule (Verpy et al., 2001). Stereocilin was associated with OHCs in two cell-surface specializations interconnecting with the hair bundle in Corti contained with inner hair cells, OHCs, supporting cells, ciliated ends of the hair cells, and the tectorial membrane. The two specializations are the horizontal top connectors of adjacent stereocilia, which have a zipper-like structure, and the attachment links that anchor the tallest stereocilin into the overlying tectorial membrane (Verpy et al., 2008, 2011). In stereocilin null (Strc_/_) mice, both these links were absent, and progressive HI appeared from P15 (Verpy et al., 2008, 2011).

STRC has a tandem structure, and the linkage region includes three other genes: CATSPER2 (MIM 607249), PP1PSK1 (MIM 610979), and CKMT1B (MIM 123290) (Zhang et al., 2007). Causative alterations in the STRC gene include copy number variations (CNVs), single nucleotide variants (SNVs), or small insertions/deletions (indels). Recently, CNVs have been recognized as having an important role in STRC variations (Yokota et al., 2019). The STRC deletions are frequently accompanied by the deletion of the CATSPER2 gene accounting for sperm motility. This genotype, characterized by deletions including both CATSPER2 and STRC, is known as deafness-infertility syndrome (DIS) in both males and females (Hildebrand et al., 2009). STRC is part of a tandem duplication, and the second copy is a pseudogene (pSTRC). The highly homologous (>99%) distal pseudogene makes molecular analysis to detect STRC mutations by next-generation (NGS) and exome sequencing (ES) challenging (Vona et al., 2015; Shi et al., 2019). Reliable screening is especially significant and affordable in recent years due to the development of analytical approaches, such as MPLA, long-range/nested PCR, and droplet digital PCR (Back et al., 2019; Shi et al., 2019).

Many studies have routinely described genetic testing of patients with DFNB16. Compared with other deafness-associated gene variants, STRC has its own unique qualities and may lead to non-syndromic or syndromic deafness. However, to date, there is no publication systematically describing the prevalence and features of DFNB. Against this background, this meta-analysis provides a global and current pooled prevalence of STRC mutations based on available gene detection. Results generated from this paper will widen available options and contribute to genetic counseling for medical workers and individuals affected by HI.

**METHODS**

This search was performed following the guidelines of Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) (Moher et al., 2009) (Supplementary Table 1).

**Search Strategy**

The literature search was conducted by electronic databases (PubMed, Google Scholar, Medline, Embase, and Web of Science) for the English language articles published prior to January 2021. We entered search terms (“STRC” OR “stereocilin” OR “DFNB16”) into each database. Two authors (SH and DZ) separately undertook literature searches and checked the reference lists of all selected articles. When disagreements occurred after the screening, further discussion took place to reach a consensus.

**Eligibility Criteria**

We included researches that met the following criteria: (1) original research, (2) study population with HI and sample sizes with no <10 probands, (3) STRC gene detection, and (4) available full-text papers written in English. We excluded (1) duplicate publications, reviews, studies with overlapping data, mechanisms and/or animals, abstract-only articles, and texts without raw data; (2) fewer than 10 probands reported; and (3) studies published in languages other than English.

**Study Selection and Data Extraction**

Two authors (SH and DZ) independently accomplished the literature selection based on predetermined criteria. The other researchers (YG and ZF) reviewed whether the results were consistent. If disagreements occurred, further discussion took place until a consensus was reached. A standard data extraction diagram is presented in Figure 1.

After relevant publications were selected, the data were collected by two reviewers (SH and DZ) from included papers as follows: first author, year of publication, region, study population, gene detection method, DFNB16, genetically confirmed cases follow: first author, year of publication, region, study population, gene detection method, DFNB16, genetically confirmed cases (non-GJB2), total HI patients (non-GJB2), DIS, carriers in total HI patients (non-GJB2), carriers in the normal-hearing population, and types of mutations in STRC (biallelic CNVs, CNVs + SNVs, or small indels, biallelic SNVs, or small indels). Discrepancies were discussed and resolved by the senior author (GG).

**Quality Assessment**

The risk of bias in each observational study was calculated using a tool developed by Hoy et al. (2012), with total scores ranging from 0 to 10. Bias was judged to be of low risk (9–10 points), moderate risk (6–8 points), or high risk (<6 points).

**Statistical Analysis**

The meta-analysis was conducted using R (version 4.0.4, The R Foundation, Vienna, Austria). To bring the proportion data closer to a normal distribution, the logit transformation was used to solve the proportions (0 or 1) exist (Lipsey and Wilson, 2000). The Shapiro–Wilk normality test was
applied to calculate the normal distribution of the transformed sample data. Assessments and 95% confidence intervals (CIs) of the prevalence of all collected articles were estimated by the random-effects model. Forest plots were used to show percentages of each study, the summary rate, and heterogeneity among publications. Between-study heterogeneity was evaluated by the $I^2$ statistic. Meta-regression was calculated to investigate the potential source of high heterogeneity. The sensitivity analysis was completed by removing low-quality papers (score ≤ 5) and determining whether the results were stable. Funnel plots and Egger's bias test were used to assess the publication bias.

**RESULTS**

A total of 294 publications were extracted from the databases. After duplicates were excluded, titles and abstracts of the remaining articles were screened, and full-text versions of 73 relevant studies were further reviewed. Finally, 37 papers were included in the meta-analysis and were used in the subsets shown in Figure 1. All detailed information extracted from eligible articles is presented in Table 1, Supplementary Table 2. The prevalence of DFNB16 in HI patients (non-GJB2) was 4.08% (95% CI: 0.0289–0.0573), the prevalence of STRC carriers in the HI participants (non-GJB2) was 4.84% (95% CI: 0.0343–0.0680), and those with normal hearing accounted for 1.36% (95% CI: 0.0025–0.0696). The prevalence of DFNB16 in genetically confirmed cases (non-GJB2) was 11.10% (95% CI: 0.0716–0.1682), the prevalence of DIS in DFNB16 patients was 36.75% (95% CI: 0.2122–0.5563), and the prevalence of biallelic deletions in DFNB16 patients was 70.85% (95% CI: 0.5824–0.8213).

**Quality Assessment**

The details of quality assessment scores for each article are available in Supplementary Table 3. None of the included
### TABLE 1 | Proportions extracted for meta-analysis.

| References       | Region      | Study population | Gene detection method\(a\) | Frequency of DFNB16\(b\) | Carrier-frequency in non-GJB2 HI | Carrier-frequency in people with normal hearing | Frequency of DFNB16 in confirmed cases\(c\) | Frequency of DIS in DFNB16 | Types of mutations in STRC |
|------------------|-------------|------------------|-----------------------------|--------------------------|---------------------------------|-----------------------------------------------|---------------------------------|-----------------------------|----------------------------|
|                   |             |                  |                             |                          |                                 |                                               |                                 |                             | CNVs | CNVs+ NVs or indels | SNVs or indels |
| Sheppard et al. (2018) | USA         | HL               | NGS+CNV                     | 2.50%                    | 2.50%                           |                                               | 7.14%                           |                             |                 |                  |                |
| Lebeke et al. (2016)    | Cameroon    | ARNSHL           | NGS+CNV                     | 10.00%                   |                                 |                                               |                                 |                             |                 |                  |                |
| Marková et al. (2018)    | Czech       | NSHL             | NGS+CNV                     | 5.56%                    | 5.56%                           |                                               |                                 | 37.50%                     | 50.00% | 43.75%           | 6.25%         |
| Plevova et al. (2017)     | Czech       | HL               | NGS+CNV                     | 10.20%                   |                                 |                                               | 35.71%                          |                             |                 |                  |                |
| Chang and Choi (2014)     | Korea       | HL               | NGS                        | 0.88%                    |                                 |                                               | 1.56%                           |                             |                 |                  |                |
| Safka Brozkova et al. (2020) | Czech    | NSHL             | NGS+CNV                     | 5.23%                    | 2.61%                           |                                               | 40.74%                          | 50.00%                     | 36.36% | 13.64%           |                |
| Kim et al. (2020)         | Korea       | HL               | NGS+CNV                     | 36.71%                   |                                 |                                               | 60.42%                          | 44.83%                     | 58.62% | 34.48%           | 6.90%         |
| Schrauwen et al. (2013)   | Europe      | ARNSHL           | NGS                        | 8.33%                    |                                 |                                               |                                 |                             |                 |                  |                |
| Kannan-Sundhari et al. (2020) | Iran | HL               | NGS                        | 4.35%                    |                                 |                                               |                                 |                             |                 |                  |                |
| Ito et al. (2019)         | Japan       | NSHL             | NGS+CNV                     | 5.95%                    | 2.38%                           |                                               | 0.93%                           |                             |                 |                  |                |
| Back et al. (2019)        | Germany     | ARNSHL           | NGS+CNV                     | 10.98%                   |                                 |                                               |                                 |                             |                 |                  |                |
| Mehta et al. (2016)       | USA         | NSHL             | NGS+CNV                     | 2.41%                    |                                 |                                               | 37.14%                          |                             |                 |                  |                |
| Morgan et al. (2020)      | Italy       | NSHL             | NGS+CNV                     | 7.53%                    |                                 |                                               | 21.21%                          |                             |                 |                  |                |
| García-García et al. (2020) | Spain    | HL               | NGS+CNV                     | 2.75%                    | 0.92%                           |                                               | 7.89%                           |                             |                 |                  |                |
| Morgan et al. (2018)      | Italy       | NSHL             | NGS+CNV                     | 1.94%                    |                                 |                                               | 6.25%                           |                             |                 |                  |                |
| Francey et al. (2012)     | USA         | NSHL             | NGS+CNV                     | 2.58%                    | 0.52%                           |                                               |                                 | 52.94%                     | 29.41% | 17.66%           |                |
| Gu et al. (2015)          | China       | NSHL             | NGS+CNV                     | 1.59%                    |                                 |                                               |                                 |                             |                 |                  |                |
| Yokota et al. (2019)      | Japan       | NSHL             | NGS+CNV                     | 1.95%                    | 5.28%                           |                                               | 7.05%                           | 88.24%                     | 100%             |                  |                |
| Downie et al. (2020)      | Australia   | HL               | NGS+CNV                     | 4.76%                    |                                 |                                               |                                 | 10.81%                     |                             |                |                  |                |
| Sommen et al. (2016)      | Western-European | ARNSHL       | NGS+CNV                     | 0.76%                    | 8.40%                           |                                               |                                 | 3.45%                      |                             |                 |                  |                |
| Zazo Seco et al. (2017)   | Netherlands | HL               | NGS+CNV                     | 2.09%                    |                                 |                                               |                                 |                             |                 |                  |                |
| Vona et al. (2015)        | Germany     | NSHL             | NGS+CNV                     | 6.38%                    | 5.32%                           |                                               |                                 |                             |                 |                  |                |
| Shearer et al. (2014)     | USA         | HL               | NGS+CNV                     | 10.81%                   |                                 |                                               | 83.78%                          | 16.22%                     |                 |                  |                |
| Budde et al. (2020)       | Egypt       | NSHL             | NGS+CNV                     | 1.75%                    |                                 |                                               | 2.27%                           |                             |                 |                  |                |
| Cabanillas et al. (2018)  | Spain       | HL               | NGS+CNV                     | 4.00%                    | 8.00%                           |                                               | 9.52%                           |                             |                 |                  |                |
| Moteki et al. (2016)      | Japan       | NSHL             | NGS+CNV                     | 1.55%                    |                                 |                                               | 5.77%                           |                             |                 |                  |                |
| Sloan-Heggen et al. (2016) | USA         | HL               | NGS+CNV                     | 6.93%                    |                                 |                                               | 20.58%                          | 23.94%                     | 77.46% | 21.13%           | 1.41%         |
| Mandelker et al. (2014)   | NA          | HL               | NGS+CNV                     | 63.64%                   |                                 |                                               | 36.36%                          |                             |                 |                  |                |

(Continued)
| References        | Region   | Study population | Gene detection method | Frequency of DFNB16 | Carrier-frequency in non-GJB2 HI | Carrier-frequency in people with normal hearing | Frequency of DFNB16 in confirmed cases | Frequency of DIS in DFNB16 | Types of mutations in STRC |
|-------------------|----------|------------------|-----------------------|---------------------|----------------------------------|-----------------------------------------------|---------------------------------|--------------------------|-----------------------------|
| Bademci et al. (2016) | Multiple | ARNSHL           | NGS+CNV               | 0.63%               |                                  | 1.11%                                         |                                 |                          | CNVs                        |
| Baux et al. (2017)  | France   | NSHL             | NGS+CNV               | 5.70%               |                                  | 18.00%                                        |                                 |                          | CNVs+ NVs or SNVs or indels |
| Ji et al. (2014)    | China    | HL               | NGS+CNV               | 11.27%              |                                  |                                               |                                 |                          | SNVs or indels              |
| Costales et al. (2020) | Spain   | HL               | NGS                   | 4.55%               |                                  |                                               |                                 |                          |                             |
| Sloan-Heggen et al. (2015) | Iran  | HL               | NGS+CNV               | 0.33%               |                                  |                                               |                                 |                          |                             |
| Guan et al. (2018)  | USA      | NSHL             | NGS+CNV               | 8.00%               |                                  | 18.18%                                        |                                 |                          |                             |
| Amr et al. (2018)   | NA       | HL               | NGS+CNV               |                     |                                  |                                               | 35.48%                         | 74.19%                   | 22.58%                     |
| Shearer et al. (2013) | USA    | NSHL             | NGS+CNV               | 4.26%               | 1.06%                           |                                               |                                 |                          |                             |
| Brownstein et al. (2020) | Israel | HL               | NGS                   | 2.27%               |                                  |                                               |                                 |                          |                             |

*NGS including: panel, targeted testing, ES, exome sequencing; CES, clinical exome sequencing; WES, whole exome sequencing; TGE, targeted genome enrichment; Sanger also included; CNV including: MLPA, QF-PCR, long-range/nested PCR, microdroplet PCR, droplet digital PCR, array CGH, SNP microarray, CMA, chromosomal microarray analysis; QCF PCR, quantitative comparative fluorescent PCR; MPS, massively parallel sequencing.

**The frequency of STRC is achieved from GJB2-negative patients.

**Genetically confirmed cases except for GJB2-related.
studies received low risk for items 2, 3, and 9, as the cluster sampling method, random selection, census, or the length of the shortest prevalence period were not provided in each survey. Out of the remaining 7 possible points, 4 studies received 7 points, 13 studies obtained 6 points, and 20 studies received 5 points.

The Global Prevalence of DFNB16 in GJB2-Negative Hearing Impaired Patients

The proportion of STRC mutations in GJB2-irrelevant HI patients varied from 0.33 to 36.71% among 33 studies (Table 1), with the highest rate observed in Korea (Kim et al., 2020). The overall pooled prevalence was 4.08% (95% CI: 0.0289–0.0573, $I^2 = 83\%$, $p < 0.01$) using a random-effects model among 6,325 subjects (Figure 2). The $I^2$ and $p$-value indicated substantial heterogeneity.

Meta-regression was used to estimate the source of heterogeneity. We investigated five categorical moderators: region (Europe, Asia, America, or others), study population (HL, NSHL, or ARNSHL), gene detection method (NGS or NGS+CNV), degree of HI (mild–moderate or other HI), and quality assessment grade (score ≤5 or score ≥5). Significant estimates were not found for moderators in region, study population, gene detection method, and quality assessment ($p = 0.9351, 0.8068, 0.6876, 0.8419$). The degree of HI was significantly associated with the overall pooled prevalence ($p = 0.0003$). The $R^2$ (amount of heterogeneity accounted for) was 49.91%, meaning that the degree of HI can explain about 49.91% of heterogeneity in the DFNB16 prevalence among GJB2-negative HI patients.

The prevalence of STRC mutations in GJB2-negative HI patients was further analyzed by subgroup focusing on world region (Supplementary Figure 1A) and the degree of HI (Supplementary Figure 1B). For region, the highest rate estimated was in Europe, which was 5.40% (95% CI: 0.0409–0.0711), followed by the US at 3.94% (95% CI: 0.0222–0.0690), other regions with 3.00% (95% CI: 0.0103–0.0841), and Asia at 2.77% (95% CI: 0.0075–0.0965). The estimated prevalence of mild–moderate HI was 14.36% (95% CI: 0.0365–0.4259), and that of other HI was 3.67% (95% CI: 0.0281–0.0478).

To estimate the stability of outcomes, we conducted sensitivity analyses by assessing the effects of removing low-quality publications (Francey et al., 2012; Chang and Choi, 2014; Gu et al., 2015; Vona et al., 2015; Bademci et al., 2016; Mehta et al., 2016; Sommen et al., 2016; Baux et al., 2017; Zazo Seco et al., 2017; Cabanillas et al., 2018; Sheppard et al., 2018; Back et al., 2019; Budde et al., 2020; Downie et al., 2020; Kim et al., 2020; Safka Brozкова et al., 2020). The summary prevalence of DFNB16 in non-GJB2 HL patients was 3.98% (95% CI: 0.0260–0.0606, $I^2 = 70\%$, $p < 0.01$), which stabilized the findings in the range of that of crude analysis. The $I^2$ and $p$-value also indicated substantial heterogeneity. Results from a funnel plot (Figure 3A) and Egger test ($p = 0.0549$, Figure 3B) indicate an insignificant level of publication bias.

Prevalence of Carrier STRC Mutation in Non-GJB2 Hearing Impaired Participants and Individuals With Normal Hearing

Eleven studies presented monoallelic variants of the STRC gene in non-GJB2 HI participants, with carriers varying from 0.92% in Spain to 11.27% in China (Table 1). The pooled carrier prevalence in HI participants (non-GJB2) assessed by the random-effects model was 4.84% (95% CI: 0.0343–0.0680, $I^2 = 54\%$, Figure 4). Three studies showed the carrier of the STRC mutation among normal-hearing individuals (Table 1). The summary carrier prevalence in people with normal hearing was 1.36% (95% CI: 0.0025–0.0696, $I^2 = 90\%$, Figure 5) using a random-effects model.

Prevalence of DFNB16 Among Genetically Confirmed Cases (Non-GJB2)

Twenty-eight studies described genetically confirmed patients (Supplementary Table 2). Twenty-four were included in the meta-analysis to estimate the prevalence of STRC mutations in genetically diagnosed patients (non-GJB2) because the sample sizes of those with genetic diagnoses in these publications were no <10 cases (Table 1). The prevalence achieved using the random-effects model was 11.10% (95% CI: 0.0716–0.1682, $I^2 = 85\%$, $p < 0.01$) in 1,610 GJB2-negative genetically confirmed cases (Figure 6). $I^2$ and $p$-value indicated substantial heterogeneity. Meta-regression was applied to evaluate the source of heterogeneity, with five categorical moderators: region, study population, gene detection method, degree of HI, and quality assessment. Significant values were not detected for region, study population, gene detection method, and degree of HI, and grade of quality assessment. Significant values were not detected for the random-effects model was 11.10% (95% CI: 0.0716–0.1682, $I^2 = 85\%$, $p < 0.01$) in 1,610 GJB2-negative genetically confirmed cases (Figure 6). $I^2$ and $p$-value indicated substantial heterogeneity. Meta-regression was applied to evaluate the source of heterogeneity, with five categorical moderators: region, study population, gene detection method, degree of HI, and grade of quality assessment. Significant values were not detected for region, study population, gene detection method, and degree of HI, and grade of quality assessment.

To estimate the stability of outcomes, we conducted sensitivity analyses by assessing the effects of deleting low-quality studies (Chang and Choi, 2014; Bademci et al., 2016; Mehta et al., 2016; Sommen et al., 2016; Baux et al., 2017; Zazo Seco et al., 2017; Cabanillas et al., 2018; Guan et al., 2018; Sheppard et al., 2018; Budde et al., 2020; Downie et al., 2020; Kim et al., 2020; Safka Brozкова et al., 2020). The summary prevalence of DFNB16 in genetically confirmed (non-GJB2) HL patients was 9.63% (95% CI: 0.0558–0.1614, $I^2 = 80\%$, $p < 0.01$) in the sensitivity analysis and without apparent fluctuation. The $I^2$ and $p$-value showed substantial heterogeneity.

Prevalence of Deafness–Infertility Syndrome in STRC-Associated Hearing Impairment

Twelve studies provided information about homozygous deletion in the CATSPER2 gene (Supplementary Table 2). Six articles,
FIGURE 2 | The global prevalence of DFNB16 in GJB2-negative hearing impaired (HI) patients.

FIGURE 3 | (A) Funnel plot. (B) Egger test.
including ≥10 DFNB16 patients, were chosen for meta-analysis (Table 1). The overall pooled DIS prevalence in DFNB16 was 36.75% (95% CI: 0.2122–0.5563, $I^2 = 80\%$) using a random-effects model among 201 individuals (Figure 7).

Prevalence of Biallelic Deletions in DFNB16
The types of mutations in STRC were described in 36 studies (Supplementary Table 2). Of these, we selected nine articles with sample sizes not smaller than 10 DFNB16 cases for meta-analysis (Table 1). Because data extracted from Yokota et al. (2019) had proportions equal to 1, we analyzed the raw data with double arcsine transformation in advance. The pooled prevalence of biallelic deletions in DFNB16 was 70.85% (95% CI: 0.5824–0.8213, $I^2 = 74\%$) with a random-effects model (Figure 8).

DISCUSSION
The classical two-step strategy for deafness genetic testing consists of GJB2/6 locus analysis and gene panel based on NGS technologies. Although most panels have the STRC gene to detect SNVs or small indels, it is challenging for the NGS dataset to accurately detect CNVs in STRC, and even more challenging to detect CATSPER2 (Yokota et al., 2019). Considering the high prevalence of DFNB16 in genetically confirmed cases (11.10%), especially in the mild–moderate HI subgroup (50.71%), and biallelic CNVs in DFNB16 (70.85%), we consider that CNV detection of STRC should be accompanied by panel testing in case of misdiagnosis. Furthermore, males with DFNB16 should be advised about CATSPER2 gene sequencing because the DIS prevalence in DFNB16 was 36.75%. This is essential information that should be considered during genetic counseling.

Different multistep strategies have been implemented for genetic exploration of HL. Valuable data were generated and screened in our study, and information should be extracted according to the following three aspects to make outcomes more reliable: First, as far as possible, consanguineous individuals should be merged into families. For instance, Lebeko et al. (2016) included 26 individuals from 10 GJB2-negative families, and the DFNB16 proportion was based on 10 families rather than patients. Although Mehta et al. (Mehta et al., 2016) did not describe consanguinity precisely, we did not omit the study from the analysis because it offered a large sample with the number of families marginally less than individuals. Second, apart from deletions and gene-pseudogene conversions, we also...
identified heterozygous duplications as monoallelic mutations, although there was no evidence of whether the duplications were pathogenic or had any effect on phenotypes (Yokota et al., 2019). At present, no patient has been diagnosed with biallelic duplications. Variations in pseudogenes that mutated into an inactive form and SNVs classified as non-pathogenic or benign were excluded for pooling prevalence. Third, when we estimated the prevalence of DFNB16 in genetically confirmed cases (non-GJB2), DIS in DFNB16, and biallelic deletions in STRC-associated HI patients, we removed studies with probands <10, which accords with the sample size exclusion criteria mentioned. From databases and references, only Cada et al. (2019) and Markova et al. (2020) showed more than 10 DFNB16 sample sizes for clinical features of DFNB16. However, we rejected these studies because it was unclear whether the data presented were for all the original patients or only the cases selected after qualified audiological examination was completed. There were no other eligible articles to add.

When separating by HI degree, the prevalence of DFNB16 in non-GJB2 patients was significantly higher in the mild–moderate group (14.36%) than the other-HI group (3.67%) whose degree of HI was unclear or mixed. In the same way, the prevalence of DFNB16 in genetically confirmed cases (non-GJB2) was significantly higher in the mild–moderate group.
Our results emphasized that STRC is a primary contributor to mild–moderate HI. This conclusion has also been documented in previous research (Francey et al., 2012; Yokota et al., 2019; Kim et al., 2020).

Our data show that diallelic CNVs (mainly deletions) are an extreme factor in STRC gene mutations and are probably accompanied by homozygous deletions in CATSPER2 gene (36.75%) simultaneously. Given that STRC CNVs might be ignored in studies using NGS HL panels, screening techniques that contain CNV detection of STRC and CATSPER2 are recommended for patients before NGS analyses, especially in patients with bilateral mild-to-moderate HI (Plevova et al., 2017). Males with DIS will be deaf and infertile, and this is crucial information that should be realized during genetic counseling (Yokota et al., 2019). Females who inherit homozygous STRC-CATSPER2 deletions will only be deaf (Hildebrand et al., 2009), but CATSPER2 CNVs in women should also be taken seriously, not only to identify the etiology in probands but also to predict and prevent the disability in the next generation.

There are some limitations to this meta-analysis. First, studies of the prevalence of STRC-related patients were available from only 16 countries, and data were not equally distributed. There were insufficient publications to provide adequate information for some continents. For example, in Africa, data were derived exclusively from Cameroon and Egypt. There were only five studies from developing countries, likely due to limited medical recording systems and medical care linked to possible under-recognition and late diagnosis of this disease. Second, the study designs and population covered in the included studies varied from hospital-based studies to national research. Investigation of DFNB16 in different populations and settings is urgently needed. Moreover, sample sizes of research studies need to be expanded, and more funding will be required for large-scale studies. Third, there is no uniform detection method for STRC variations. Information about STRC CNVs, monoallelic mutation, or CATSPER2 deletions was ignored in some studies, affecting our confidence in prevalence assessment. The screening of CNVs could be impacted by different CNV detection methods, such as NGS or SNP array, and the proportion of STRC mutations may be found to be higher if effective strategies are applied (Yokota et al., 2019).

In conclusion, we undertook the first meta-analysis to demonstrate that DFNB16 plays a crucial role in mild-to-moderate ARNSHL. The findings also emphasize the significance of detecting copy number variations of the STRC gene.

**DATA AVAILABILITY STATEMENT**

The original contributions presented in the study are included in the article/Supplementary Material, further inquiries can be directed to the corresponding author.

**AUTHOR CONTRIBUTIONS**

SH conducted literature searches and data extraction and wrote the manuscript. DZ conducted literature searches and data extraction. YG and ZF reviewed the results. GG determined the final outcome, and wrote and checked the manuscript. All authors contributed to the article and approved the submitted version.

**SUPPLEMENTARY MATERIAL**

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fgene.2021.707845/full#supplementary-material

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