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Predictive sensitivity and concordance of Machine-Learning Tools for diagnosing DFNA9 in a large series of p.Pro51Ser variant carriers in the *COCH***-gene.**

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Abstract

Objective: In this study we aimed to evaluate the predictive cross-sectional sensitivity and longitudinal concordance of a machine-learning algorithm in a series of genetically confirmed p.P51S variant carriers (DFNA9).

Study Design: Cross-sectional study.

Setting: tertiary and secondary referral center.

Patients: Audiograms of 111 subjects with the p.P51S mutation in the *COCH*-gene were analysed cross-sectionally. A subset of 17 subjects with repeated audiograms were used for longitudinal analysis.

Intervention(s): All audiological thresholds were run through the web-based AudioGene© v4.0 software.

Main Outcome Measure(s): Sensitivity for accurate prediction of DFNA9 for cross-sectional data and concordance of correct prediction for longitudinal auditory data.

Results: DFNA9 was predicted with a sensitivity of 93.7% in a series of 222 cross-sectionally collected audiological thresholds (76.1% as first gene locus). When using the hearing thresholds of the best ear, the sensitivity was 94.6%. The sensitivity was significantly higher in DFNA9 patients aged younger than 40 and aged 60 years or older, compared to the age group of 40 to 59 years, with resp. 97.6% (p< 0.0001) and 98.8% (p<0.0001) accurate predictions. An average concordance of 91.6% was found to show the same response in all successive longitudinal audiometric data per patient.

Conclusions: Audioprofiling software can accurately predict DFNA9 in an area with a high prevalence of confirmed carriers of the p.P51S variant in the *COCH*-gene. This algorithm yields high promises for helping clinicians in directing genetic testing in case of a strong family history of progressive hearing loss, especially for very young and old carriers.

Introduction

Hearing impairment affects approximately 466 million people worldwide and the prevalence increases with age^{1,2}. It significantly impacts quality of life at the individual level and poses a burden on the community level, with substantial economic impact. Causes vary from environmental to genetic factors³. Hereditary hearing loss can be subdivided in syndromic, which is associated with non-otologic anomalies, and non-syndromic, which is associated only with otologic symptoms. Non-syndromic hearing loss is further classified by mode of heritance: autosomal dominant (DFNA), autosomal recessive (DFNB), X-linked (DFNX), or mitochondrial. Subtypes are then numbered sequentially in the order by which they were described e.g. DFNA1, DFNA2 and so on. Most of these cases inherit autosomal recessive with 77%, followed by an autosomal dominant inheritance with $22\%^{3,4}$. Autosomal recessive hearing loss is characterized by severe prelingual hearing loss, whereas autosomal dominant hearing loss typically presents post-lingual and is progressive in nature⁵.

DFNA9 is an autosomal dominant disorder, characterized by progressive sensorineural hearing loss and vestibular deterioration. It is caused by mutations in the Coagulation Factor C Homolog (*COCH*)-gene. It is one of the many causes for mendelian hearing loss that can potentially be detected with algorithm's based on clinical data. In these patients, hearing loss typically starts developing at the high frequencies in the third decade and is accompanied with progressive vestibular deterioration⁶⁻⁹.

When encountering a patient with progressive sensorineural hearing loss, adequate history taking is key to determine a familial predisposition. If an underlying genetic cause is considered, genetic analysis can be requested to confirm clinical suspicion, but if clinical features may help the clinician to select accountable gene mutations, it may help reduce health dispenses⁵.

Based on the study of phenotype-genotype correlations in autosomal dominant non-syndromic hearing loss, a computer algorithm (AudioGene[©]) was developed by Hildebrand et al., with the initial goal to prioritize causative gene loci for Sanger sequencing. Based on audiological thresholds and annual threshold deterioration of patients with suspected autosomal dominant hereditary hearing loss, this program provides a top three prediction of accountable gene loci containing the causative mutation. This is considered a powerful tool which complements the otolaryngologist's daily clinical practice¹⁰⁻¹³.

Our aim was to determine the predictive sensitivity of AudioGene^{\odot} in a series of 111 genetically confirmed c.151 C>T, p.Pro51 Ser (P51S) variant carriers in the *COCH*-gene. Additionally, the average positive concordance, using successive longitudinal audiometric data per patient, was evaluated.

Methods and materials

The study was designed and conducted according to the Declaration of Helsinki (1996) and it was approved by the local ethics committees of the Antwerp University Hospital and the Hasselt Jessa Hospital (B300201630243). (Dale & amp; Salo, 1996) The study was registered in ClinicalTrials.gov (NCT04331015).

Patient enrolment and chart lifting started on January $1st$, 2019 and ended on January $31st$, 2020. All siblings of definite p.P51S variant carriers of at least 18 years of age were eligible for enrolment. The following exclusion criteria were used: all siblings younger than 18 years at the time of investigation, conductive hearing loss (difference of at least 15 dB HL between air and bone conduction measured on at least 3 subsequent frequencies, sensorineural hearing loss due to other concomitant disease, a history of significant occupational noise exposure, vestibular dysfunction due to other causes than DFNA9, previous middle ear surgery, known neurological disorders, known cerebral/cerebellar disorders, intracranial disease/tumours, unwillingness or inability to undergo thorough audiological and vestibular examination and eardrum perforation.

Audiometric analysis was performed by the modified Hughson-Westlake methodology to determine hearing thresholds in decibels hearing level (dB HL) at the following respective frequencies: 0.125, 0.25, 0.5, 1, 2, 3, 4, 6 and 8 kHz¹⁴.

The subjects' age was allocated according to the age at the time of investigation. Subjects were divided into three age groups: ≤ 40 years, 40-60 years and ≥ 60 years.

For those p.P51S carriers of whom more than two audiometry's were measured with at least one year in between each measurement, all audiometric data were collected to evaluate the concordance of AudioGene[©] v4.0 in consequently reproducing the same result after each run. All data were run through the open-source software AudioGene^{\odot} v4.0 (Center for

Bioinformatics and Computational Biology, University of Iowa City, IA, USA)([http://Audiogene.eng.uiowa.edu/\)](http://audiogene.eng.uiowa.edu/) to predict the three most probable genetic loci for autosomal dominant hearing loss.

Hearing thresholds for right ears, left ears, all ears individually and binaural averaged hearing thresholds were run through the algorithm. In addition, a distinction was made per patient between the ear with the best and worst hearing thresholds based on the average of hearing thresholds for all frequencies.

Subjects were divided in the following groups, according to the predictions:

- Group 1: those carriers with accurate DFNA9 prediction as first gene locus.
- Group 2: those with accurate DFNA9 prediction as gene locus at any rank (within top 3).
- Group 3: those with no DFNA9 prediction within the top 3 gene loci.

Differences were assessed by means of the Fisher exact test for categorical data and Mann-Whitney U test for continuous data. Differences between the different age groups were investigated using the chi-square test.

Furthermore, descriptive statistics were used to describe the cross-sectional and longitudinal data and corresponding predictions. Cross-sectional data were analysed based on hearing thresholds of each ear individually, best and worst ear and binaural averaged hearing thresholds. Longitudinal data were analysed based on hearing thresholds of each ear separately, since several measurements were excluded due to conductive hearing loss.

Results

Data of 222 ears from 111 genetically confirmed p.P51S *COCH* variant carriers were included in our database. Age ranged from 18 to 80 years with a mean of 54 years and 51.4% of our study population consisted of females. For the concordance study, longitudinal audiometric data of 17 carriers were collected, totalizing 128 measurements from 28 ears (mean 56 y, range: 29-79). There was an average of four successive measurements for each individual, with a time lapse of at least 1 year in between each measurement. 53.6% of these cases consisted of males.

Univariate analysis showed no significant differences in age, gender and hearing thresholds between the different groups as defined in the methods section (see supplementary digital material).

Based on the algorithm's predictions for each ear individually, DFNA9 was accurately predicted by AudioGene[®] in 76.1% of the cases, 14% by 2nd prediction and 3.6% by 3rd prediction (sensitivity for positive prediction: 93.7%). Both for binaural averaged and assessment of left and right ears separately showed similar results.

The algorithm's sensitivity for hearing thresholds of the best and worst hearing ear were 94.6% and 92.8%, respectively (table 1).

Table 2 summarizes the sensitivity of AudioGene[©] v4.0 in accurately predicting DFNA9 as first gene locus as well as adequate DFNA9 prediction within top 3 gene loci in the three age groups respectively. In the group of DFNA9 prediction within top 3 gene loci, sensitivity was significantly higher for subjects aged under 40 years (97.6%) and above 60 years (98.8%). Similar results were obtained when DFNA9 was predicted as first gene locus.

In figures 1 and 2, the frequency distribution of all different predicted gene loci demonstrated DFNA2 as the second most common locus in the top three predictions as well as in the group of first predictions.

Mean audiometric thresholds and corresponding 95% confidence interval of all 111 crosssectionally acquired audiometric data of group 2 versus group 3 were displayed in figure 3. The average audioprofiles of those measurements associated with adequate DFNA9 prediction by the algorithm tended to show a more typical ski-slope or down-sloping curve compared to those with no DFNA9 prediction within the top 3 gene loci. As demonstrated above, there were no significant differences between the two groups with regard to age, gender and hearing thresholds, however, one can observe slight differences in audioprofile, with the former showing a more typical ski-slope or down-sloping curve compared to the average audioprofile of those carriers with inaccurate prediction. The same results were obtained when analysing the pure tone average (PTA) of different frequencies (0.5-2 kHz; 0.5-4 kHz; 1-4 kHz).

In figure 4, the same comparison of the audioprofiles of longitudinal audiometric data reproduced similar curves, however, with an additional tendency to show lower thresholds at the lower frequencies in group 3 (no DFNA9 prediction within top 3 gene loci). Once more, age, gender and hearing threshold of both group 2 and 3 were not significantly different from each other.

The average positive concordance to accurately predict DFNA9 as $1st$ gene locus was 78.6% and 91.6% when the condition was predicted within top 3 gene loci. Outcome details are available in supplementary digital material.

Discussion

Based on clinical features, of various types of genetic hearing loss such as audioprofiles, artificial intelligence (AI) can potentially predict the gene locus of causative genetic mutation and aid the physician in his clinical decision making in case of a family history of progressive hearing loss¹⁰. In this study we aimed to evaluate the predictive cross-sectional sensitivity and longitudinal concordance of a machine-learning algorithm, AudioGene[©] v4.0, in a large series of confirmed p.P51S variant carriers (DFNA9).

The sensitivity to accurately predict DFNA9 regardless of ranking within top 3 gene loci was 93.7% and 76.1% as first gene locus prediction. The high positive sensitivity was independent of laterality of the audiometric data, whether this was limited to binaural average thresholds, left or right sided and best or worst ear.

Remarkably, this sensitivity was age-dependent, since this was significantly higher for predictions with audioprofiles derived from very young carriers $($ < 40 y) and from older subjects $(\geq 60y)$. This may correspond to the fact that the progression of hearing loss is not linear in DFNA9 and the highest degree of hearing decline is observed between 40 and 59 years in these patients^{7,9}. This might be very difficult to pick up with an algorithm that is based on progression assuming linear deterioration.

Hence, what seems to be a limitation of the program might in fact become its most powerful usefulness in clinical practice. When there is a high suspicion of carrier status of p.P51S variant in *COCH*, causing DFNA9, in a particular individual with a positive history of familial hereditary oto-vestibular impairment, this algorithm might become extremely useful in predicting DFNA9 for those carriers at a young age, when vestibular impairment is not yet present or is not yet debilitating (40 y) or for those aged older than 60 years, when it has

become less evident in drawing up a three-generation family tree that still contains enough living individuals while they have descendants who do not yet have hearing loss.

Furthermore, patients who are eligible for cochlear implants often belong to this age group (\geq 60 y) and many of them also present with combined oto-vestibular deficits, which are not necessarily caused by *COCH* mutations.

Running their audioprofiles through programs such as AudioGene© might help selecting potential carriers of DFNA9 causing variants more accurately. With this perspective, it would then be easier and less costly to select those patients for screening for *COCH*, before applying multi-gene testing.

Figure 3 depicts the superimposed audioprofiles (means and 95% CI) of cases with DFNA9 within top 3 gene loci versus cases with no DFNA9 prediction. The variance of the group without correct prediction was higher than that of the group with correct prediction due to the low number of inclusions, which creates higher standard error values. Even if no significant differences were found between both groups at any frequency, it should be noted that the curve of the group with accurate prediction had a more pronounced down sloping curve and more pronounced selective typical high frequency loss, as it is well-known in early stages of DFNA9. For comparison, those of the group with inaccurate prediction showed lower hearing thresholds at the middle frequencies. The audioprofile of the group with accurate prediction coincided better with the Age-Related Typical Audiograms (ARTA) for the p.P51S variant carries^{7,9}.

The high concordance of the algorithm to reproduce the same prediction on successive runs of all audiometric measurements per individual testifies to the robustness of the program.

DFNA2 was the second most predicted locus, independent of the ranking within the top 3 gene loci. DFNA2 is linked to mutations in the *KCNQ4* gene and is associated with

predominantly high frequency hearing loss with adult onset, however, in contrast to DFNA9, hearing loss is more pronounced in high and medium frequencies and deterioration starts at an earlier age. Furthermore, DFNA2 shows no abnormalities on imaging and is not associated with vestibular deterioration¹⁵⁻²⁰. The somewhat similar audiometric presentation might explain the high ranking of DFNA2 predictions in our series of confirmed p.P51S variant carriers. DFNA2 may also have a high ranking because the algorithm includes considerably more data of this genetic disorder compared to the others 10 .

Despite very high sensitivity for DFNA9 predictions obtained in the largest series of 111 genetically confirmed p.P51S carriers, the program does not yield 100% accuracy in all cases and it does not indicate whether or not the subject actually carries no mutations at all. This may be very misleading. Including normative ISO 7209 values for age and gender for the prediction of non-carriership would further facilitate the applicability of the program in everyday clinical practice¹⁰.

Another reason for caution is that more than 30 different mutations in the *COCH*-gene have been described, each with discrete differences in phenotype²¹. Depending on their location in the gene, several variants may express with late-onset hearing loss and more pronounced vestibular impairment whereas others may limit expression with only early onset hearing loss without vestibular signs²². Furthermore, approximately 67 loci for autosomal dominant nonsyndromic hearing loss have been described in the literature. It should be noted that not all of these have their audioprofiles recorded in AudioGene^{\odot 12,23}. Consequently, the algorithm cannot predict these causes of hearing loss

Limitations of this study are that positive and negative predictive values were not calculated due to absence of any false negatives. Also, since the number of inaccurate predictions (i.e. no DFNA9 within top 3 gene loci) was very limited compared to the number of accurate

predictions, possible pattern differences in audioprofile between accurate and inaccurate predictions could not be computed with sufficient significance.

The high sensitivity and concordance in predicting DFNA9 in the largest series of genetically confirmed p.P51S variant carriers, independently of cross-sectional or longitudinal collected audiometric data, yields high promises for helping the clinician in diagnosing hereditary hearing loss, especially for very young $($ <40 y) and older carriers (≥ 60 years).

However, in order to limit inaccurate predictions, additional information about family history, vestibular impairment, and if available comparison of audioprofiles with ISO 7209 normative value for presbycusis in relation to age and gender may contribute to better performances. The user should always remember the possibility of absence of any carrier status when running this algorithm.

To conclude, we can state that this machine-learning-based algorithm provides a valuable addition in everyday clinical practice to cost-effectively screen for autosomal dominant hearing loss.

Tables & figures

Table 1:

Prediction percentage of automated algorithm for DFNA9.

Table 2:

 X^2 analysis of subjects per age category with correct $1st$ prediction versus no correct prediction in top 3 and correct prediction in top 3 versus no correct prediction in top 3.

Figure 1: Number of predictions per locus in a population of 1st predictions

Figure 3:

Mean hearing thresholds of cross-sectional auditory data in dB HL and corresponding 95% confidence interval of cases with and without correct prediction in top three.

Frequency

Figure 4:

Mean hearing thresholds of longitudinal auditory data in dB HL and corresponding 95% confidence interval of cases with and without correct prediction in top three.

Supplemental digital content

Supplemental table 1:

Univariate analysis comparing cases with correct $1st$ prediction with cases with $2nd$ or 3rd prediction.

Supplemental table 2:

Univariate analysis comparing cases with correct prediction in top 3 with cases without correct prediction in top 3.

Supplemental table 3:

Univariate analysis comparing cases with correct $1st$ prediction with cases without correct prediction in top 3.

Supplemental table 4:

Concordance for successive hearing thresholds per case.

Supplemental table 5:

Descriptive data of DFNA9 cases with corresponding longitudinal audiometry's.

References

- 1. WHO. WHO global estimates on prevalence of https://www.who.int/pbd/deafness/estimates/en/ *.* 2018.
- 2. Nash SD, Cruickshanks KJ, Klein R, et al. The prevalence of hearing impairment and associated risk factors: the Beaver Dam Offspring Study. *Arch Otolaryngol Head Neck Surg.* 2011;137(5):432-439.
- 3. Shearer AE, Hildebrand MS, Smith RJH. Hereditary Hearing Loss and Deafness Overview. In: Adam MP, Ardinger HH, Pagon RA, et al., eds. *GeneReviews((R))*. Seattle (WA)1993.
- 4. Morton NE. Genetic epidemiology of hearing impairment. *Ann N Y Acad Sci.* 1991;630:16-31.
- 5. Chang KW. Genetics of Hearing Loss--Nonsyndromic. *Otolaryngol Clin North Am.* 2015;48(6):1063-1072.
- 6. Robertson NG, Khetarpal U, Gutierrez-Espeleta GA, Bieber FR, Morton CC. Isolation of novel and known genes from a human fetal cochlear cDNA library using subtractive hybridization and differential screening. *Genomics.* 1994;23(1):42-50.
- 7. Bom SJ, Kemperman MH, Huygen PL, Luijendijk MW, Cremers CW. Cross-sectional analysis of hearing threshold in relation to age in a large family with cochleovestibular impairment thoroughly genotyped for DFNA9/COCH. *Ann Otol Rhinol Laryngol.* 2003;112(3):280-286.
- 8. Bischoff AM, Huygen PL, Kemperman MH, et al. Vestibular deterioration precedes hearing deterioration in the P51S COCH mutation (DFNA9): an analysis in 74 mutation carriers. *Otol Neurotol.* 2005;26(5):918-925.
- 9. JanssensdeVarebeke S, Topsakal V, Van Camp G, Van Rompaey V. A systematic review of hearing and vestibular function in carriers of the Pro51Ser mutation in the COCH gene. *Eur Arch Otorhinolaryngol.* 2019;276(5):1251-1262.
- 10. Hildebrand MS, DeLuca AP, Taylor KR, et al. A contemporary review of AudioGene audioprofiling: a machine-based candidate gene prediction tool for autosomal dominant nonsyndromic hearing loss. *Laryngoscope.* 2009;119(11):2211-2215.
- 11. Eppsteiner RW, Shearer AE, Hildebrand MS, et al. Using the phenome and genome to improve genetic diagnosis for deafness. *Otolaryngol Head Neck Surg.* 2012;147(5):975-977.
- 12. Taylor KR, Deluca AP, Shearer AE, et al. AudioGene: predicting hearing loss genotypes from phenotypes to guide genetic screening. *Hum Mutat.* 2013;34(4):539-545.
- 13. Huygen PL, Pennings RJ, Cremers CW. Characterizing and Distinguishing Progressive Phenotypes in Nonsyndromic Autosomal Dominant Hearing Impairment. *Audiological Medicine.* 2003;1(1):37-46.
- 14. Carhart R, & Jerger, J. Preferred method for clinical determination of pure-tone thresholds. *Journal of Speech & Hearing Disorders.* 1959;24, (24):330–345.
- 15. Dominguez LM, Dodson KM. Genetics of hearing loss: focus on DFNA2. *Appl Clin Genet.* 2012;5:97-104.
- 16. Topsakal V, Pennings RJ, te Brinke H, et al. Phenotype determination guides swift genotyping of a DFNA2/KCNQ4 family with a hot spot mutation (W276S). *Otol Neurotol.* 2005;26(1):52-58.
- 17. De Leenheer EM, Ensink RJ, Kunst HP, et al. DFNA2/KCNQ4 and its manifestations. *Adv Otorhinolaryngol.* 2002;61:41-46.
- 18. Coucke PJ, Van Hauwe P, Kelley PM, et al. Mutations in the KCNQ4 gene are responsible for autosomal dominant deafness in four DFNA2 families. *Hum Mol Genet.* 1999;8(7):1321-1328.
- 19. Van Hauwe P, Coucke PJ, Declau F, et al. Deafness linked to DFNA2: one locus but how many genes? *Nat Genet.* 1999;21(3):263.
- 20. Van Camp G, Coucke PJ, Kunst H, et al. Linkage analysis of progressive hearing loss in five extended families maps the DFNA2 gene to a 1.25-Mb region on chromosome 1p. *Genomics.* 1997;41(1):70-74.
- 21. Downie L, Halliday J, Burt R, et al. Exome sequencing in infants with congenital hearing impairment: a population-based cohort study. *Eur J Hum Genet.* 2020;28(5):587-596.
- 22. Bae SH, Robertson NG, Cho HJ, et al. Identification of pathogenic mechanisms of COCH mutations, abolished cochlin secretion, and intracellular aggregate formation: genotype-phenotype correlations in DFNA9 deafness and vestibular disorder. *Hum Mutat.* 2014;35(12):1506-1513.
- 23. Van Camp G SR. Hereditary Hearing Loss Homepage. [https://hereditaryhearingloss.org.](https://hereditaryhearingloss.org/)