Supplementary Table 1: Specific designed primers used for PCR and Sequencing

GENE	EXON	FORWARD PRIMER	REVERSE PRIMER	GC CONTENT	ANNEALING TEMP	PRODUCT LENGTH (bp)
SPNS2	5	CTCAGCACCCCTCTCTCTTC	CTGAGTCCCTCCTTCTGCAG	60	60^{0}C	263
SPNS2	6	CTCAGCACCCCTCTCTCTTC	CCCTGGTGTCCTAACTTCCT	60	58^{o}C	223

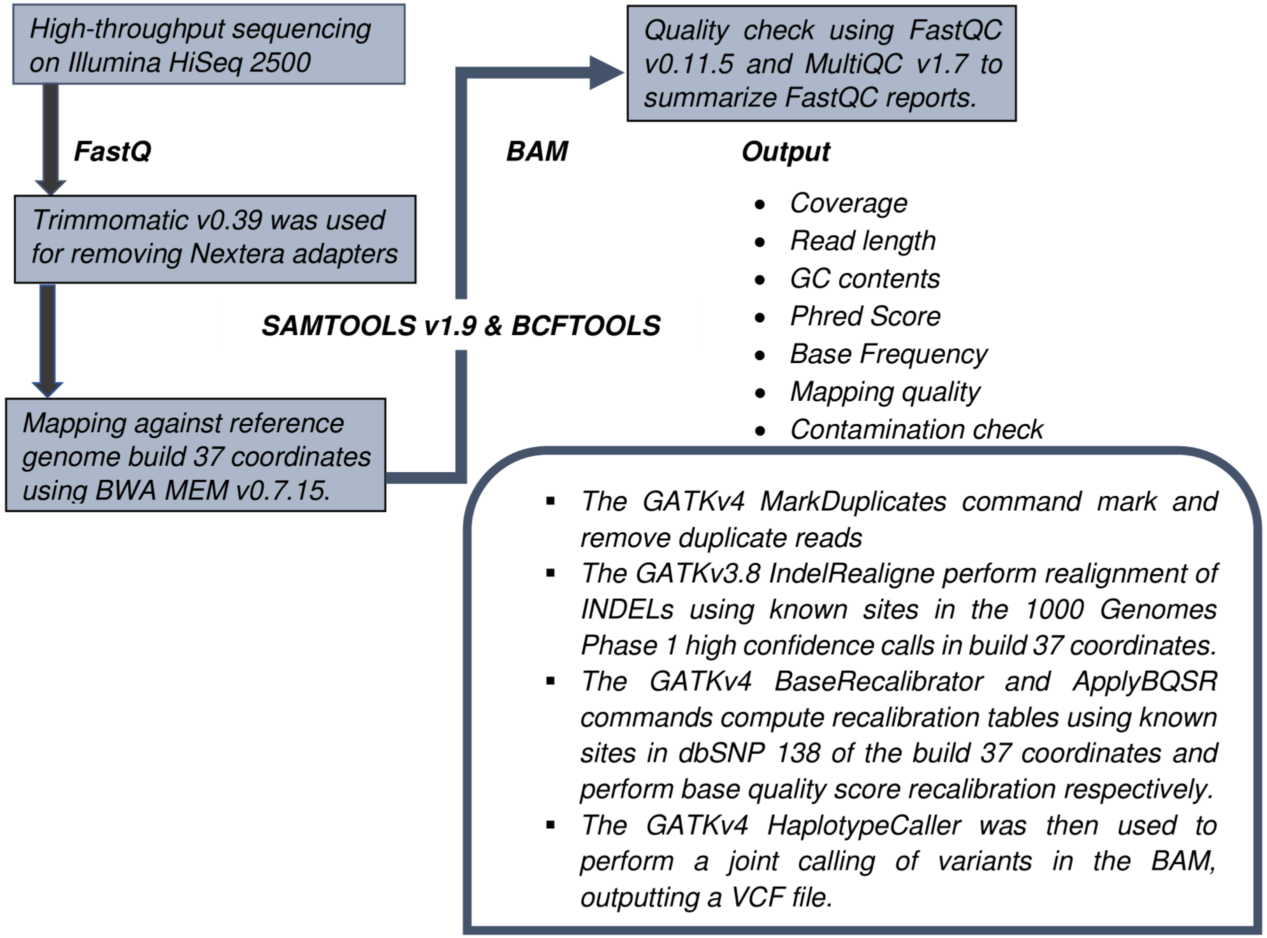
Supplementary Table 2: Summary of WES QC metrics

Sample Name	% Dups	% GC	Total Sequences(millions)
CAM103_S1	14.7%	44%	37.4
CAM104_S1	13.9%	46%	34.7
CAM115_S1	14.6%	47%	33.5
CAM116_S1	12.4%	45%	30.2
CAM117_S1	12.6%	45%	33.4
CAM119_S1	15.3%	47%	36.7
CAM14_S1	12.7%	45%	33.1
CAM167_S1	13.2%	45%	34.6
CAM168_S1	12.8%	46%	30.2
CAM192_S1	14.1%	46%	35.6

CAM28_S1	13.7%	46%	33.7
CAM41_S1	14.3%	45%	36.1
CAM51_S1	15.6%	47%	42.8
CAM56_S1	14.4%	46%	34.1
CAM60_S1	13.2%	46%	36.6
CAM75_S1	14.5%	47%	35.1
CAM76_S1	15.1%	46%	40.7
CAM78_S1	14.6%	45%	38.1

Sample Name	**% Dups**	**% GC**	**Total Sequences(millions)**
17104FL-07-01-SA002_S1	35.5%	50%	105.2
17104FL-07-01-SA007_S1	33.3%	49%	90.5
17104FL-07-01-SA013_S1	32.6%	49%	72.4
17104FL-07-01-SA015_S1	34.4%	50%	87.6
17104FL-07-01-SA016_S1	35.6%	50%	100.8
17104FL-07-01-SA017_S1	36.4%	50%	106.6
17104FL-07-01-SA021_S1	34.1%	50%	90.3
17104FL-07-01-SA022_S1	35.7%	50%	94.9
17104FL-07-01-SA023_S1	31.9%	49%	60.5
17104FL-07-01-SA025_S1	34.0%	50%	80.3
17104FL-07-01-SA026_S1	32.4%	50%	85.4
17104FL-07-02-SA027_S1	33.3%	50%	84.8

17104FL-07-02-SA028_S1	31.6%	49%	64.4
17104FL-07-02-SA029_S1	35.5%	50%	100.6
17104FL-07-02-SA030_S1	35.2%	50%	95.6
17104FL-07-02-SA031_S1	37.5%	50%	122.6
17104FL-07-02-SA033_S1	31.8%	50%	71.1
17104FL-07-02-SA035_S1	32.9%	50%	66.6
17104FL-07-02-SA037_S1	32.3%	50%	72.6
17104FL-07-02-SA038_S1	28.7%	50%	45.1
17104FL-07-02-SA040_S1	33.5%	49%	87.2
17104FL-07-02-SA041_S1	31.9%	50%	67.6



High-throughput sequencing on Illumina HiSeq 2500

Trimmomatic v0.39 was used for removing Nextera adapters

Mapping against reference genome build 37 coordinates using BWA MEM v0.7.15.

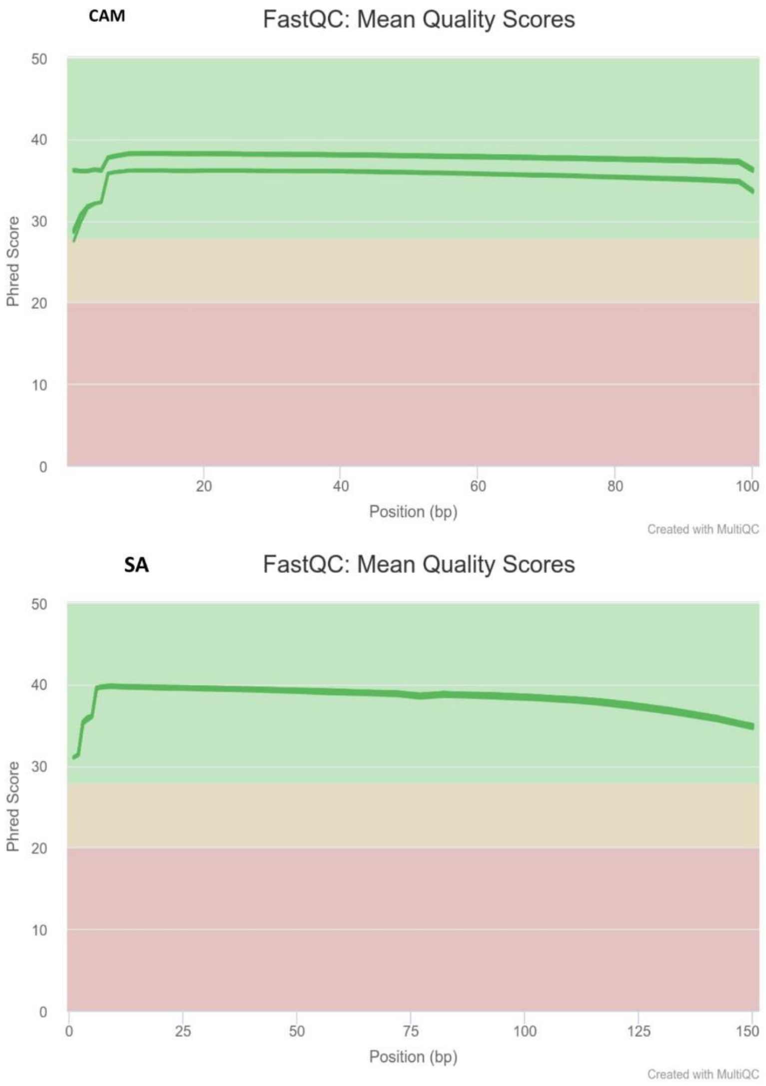
Quality check using FastQC v0.11.5 and MultiQC v1.7 to summarize FastQC reports.

Output

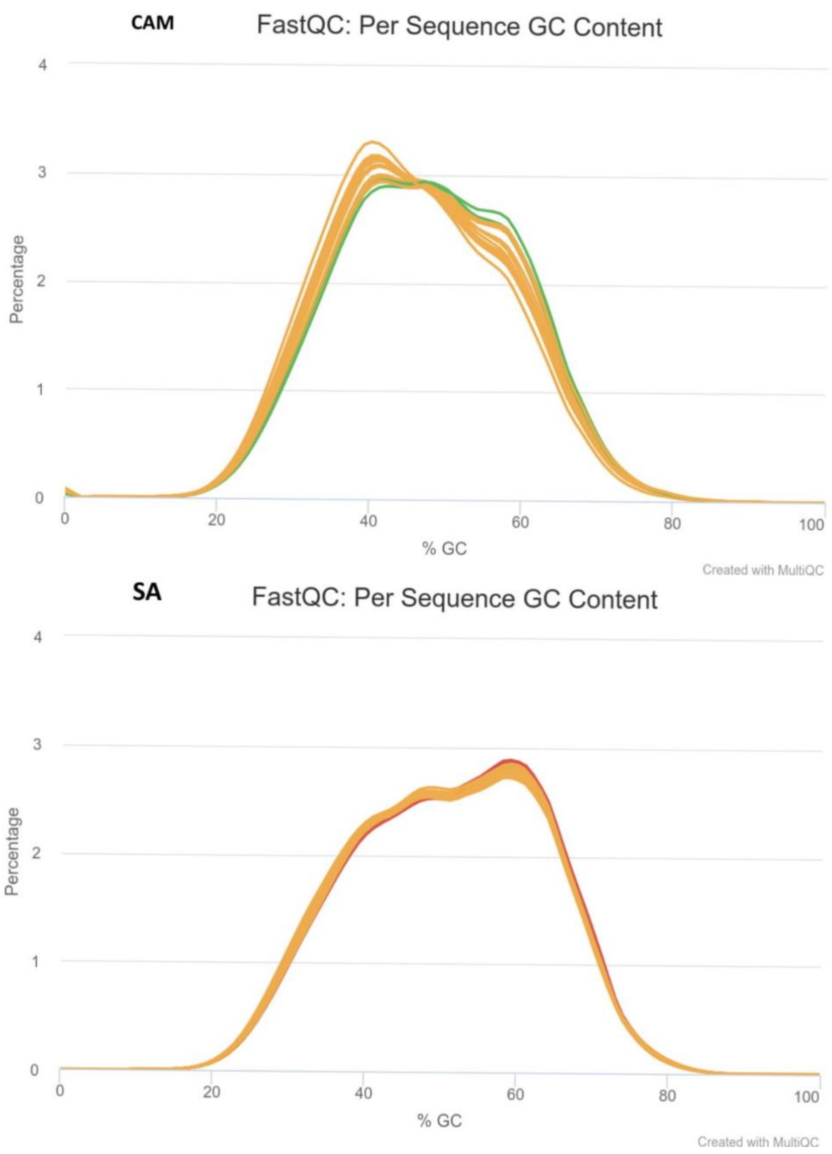
- *Coverage*
- *Read length*
- *GC contents*
- *Phred Score*
- *Base Frequency*
- *Mapping quality*
- *Contamination check*

- *The GATKv4 MarkDuplicates command mark and remove duplicate reads*
- *The GATKv3.8 IndelRealigne perform realignment of INDELs using known sites in the 1000 Genomes Phase 1 high confidence calls in build 37 coordinates.*
- *The GATKv4 BaseRecalibrator and ApplyBQSR commands compute recalibration tables using known sites in dbSNP 138 of the build 37 coordinates and perform base quality score recalibration respectively.*
- *The GATKv4 HaplotypeCaller was then used to perform a joint calling of variants in the BAM, outputting a VCF file.*

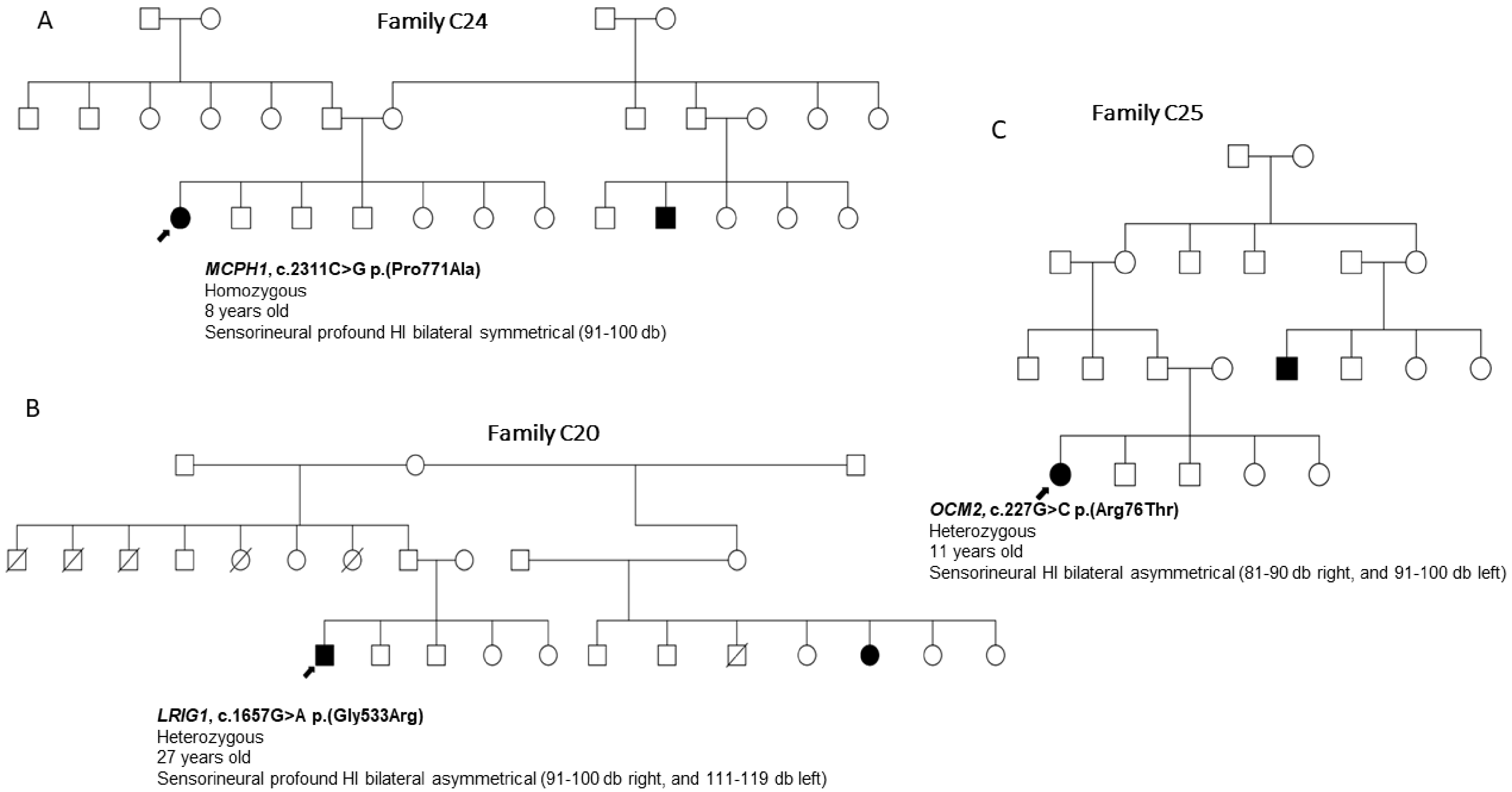
Supplementary Figure 1: Post-sequencing analyses and bioinformatics approaches and tools used in this study.



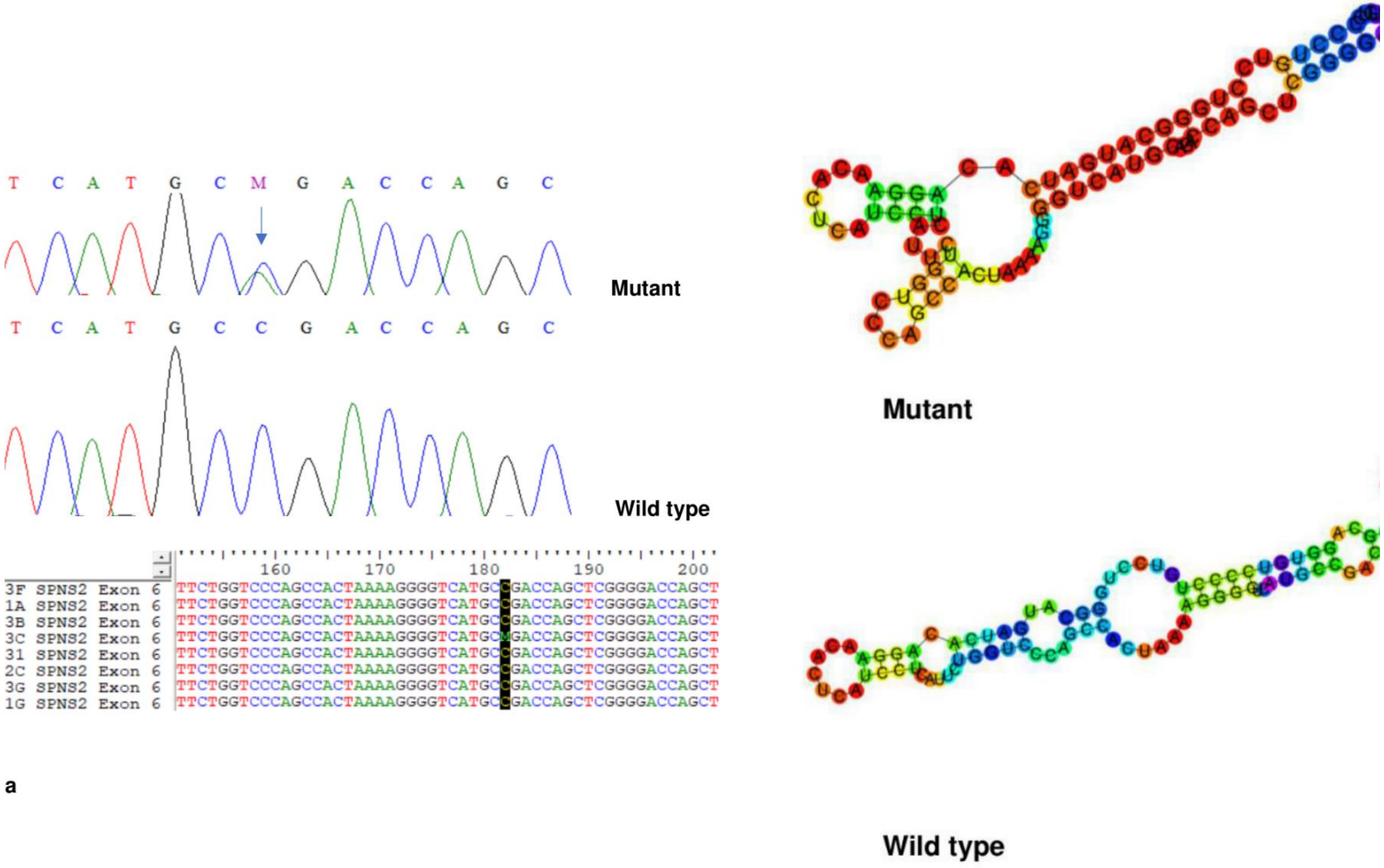
Supplementary Figure 2: Mean sequencing Phred scores of > 35 was achieved in the WES data.



Supplementary Figure 3: High per sequencing GC content obtained from the WES data.



Supplementary Figure 4. Pedigrees of patients' families found with PLP in Human-Mouse Orthologs Genes
Panel A. The patient identified with biallelic PLP variant MCPH1, c.2311C>G p.(Pro771Ala), was a 8-year-old girl, presenting with congenital prelingual bilateral and symmetrical sensorineural profound hearing loss (90-100 db), from an non-consanguineous parents, with a family history compatible with autosomal recessive non-syndromic hearing impairment. The two other Cameroonian patients that were monoallelic with variants in *LRGI1*, c.1657G>A p.(Gly533Arg) and *OCM2*, c.227G>C p.(Arg76Thr), Panel B and C respectively, also presented with congenital non-syndromic sensorineural hearing impairment with families' pedigrees compatible with autosomal recessive inheritance.



a

b

Supplementary Figure 5: [A] Chromatograms' differentiation and sequence alignment of *SPNS2* exon 6 c.C867A p.P289Q. [B] The prediction of the secondary structure of transfer RNA with and without the *SPNS2* P867Q variant. The thermodynamic free energy of mutant is 49.31 kcal/mol while for wild-type is 49.77 kcal/mol, result suggest a possibility to affect intermolecular base-pairing and consequently reduction in RNA functions. Source: http://rna.tbi.univie.ac.at/.