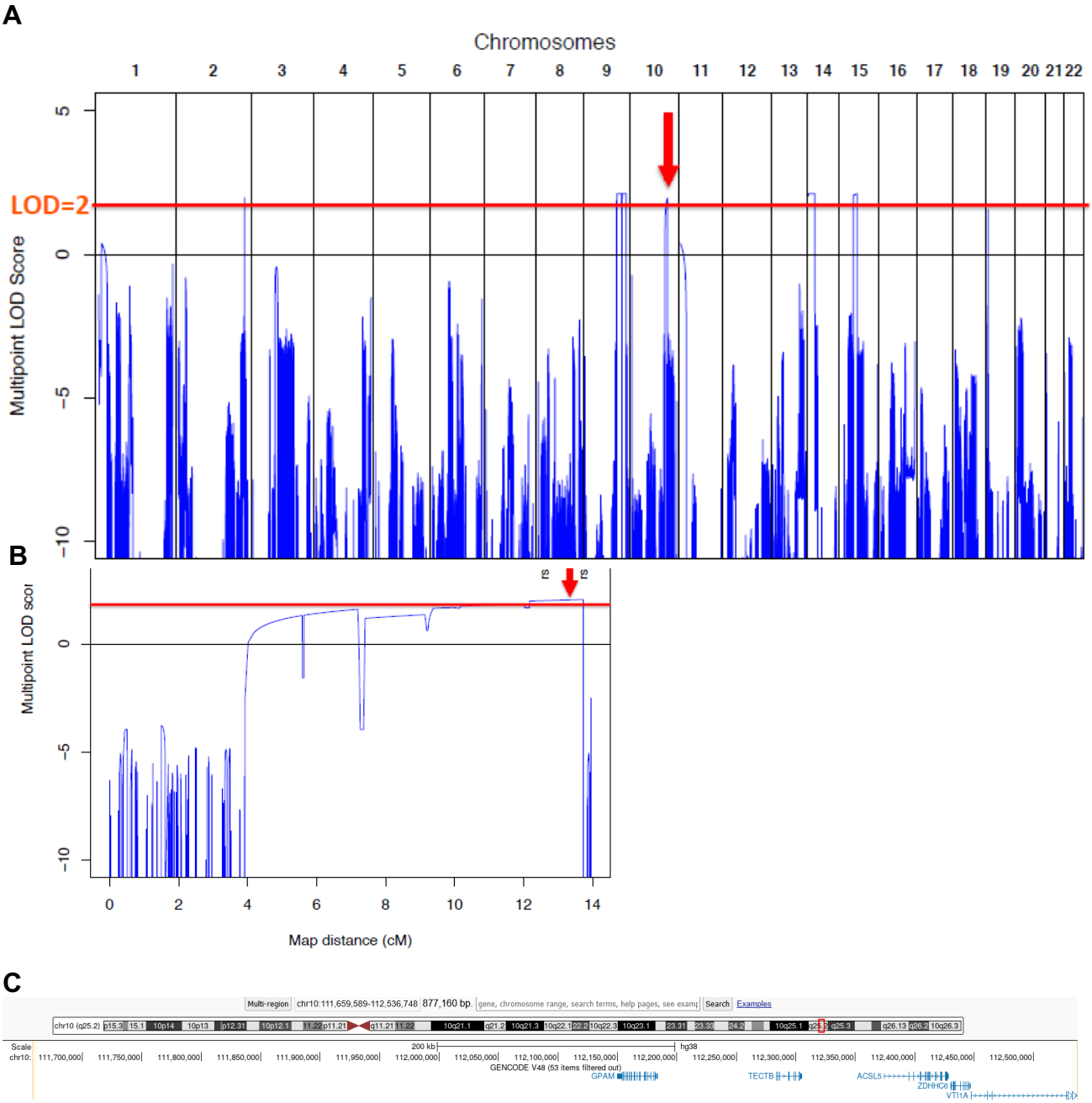
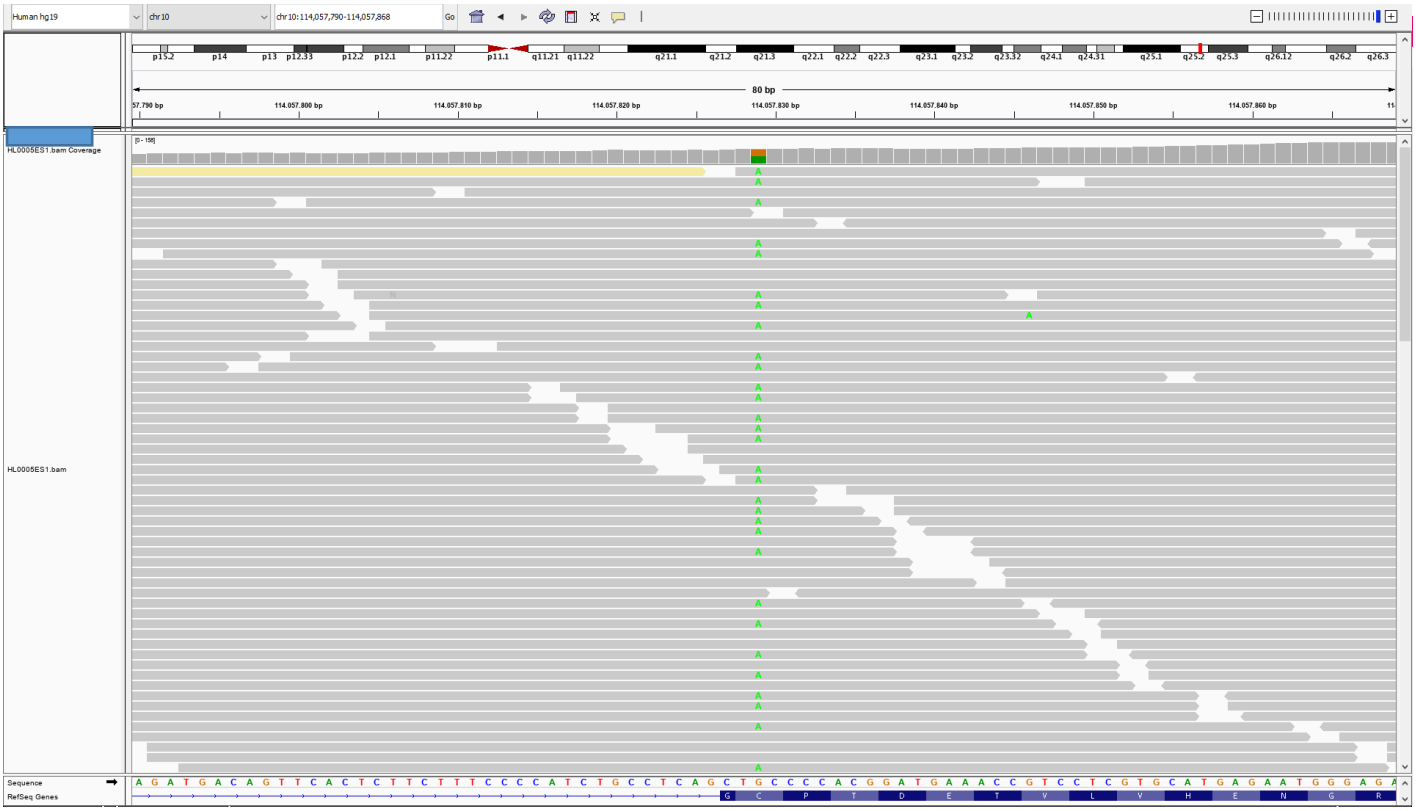
Supplementary Figures

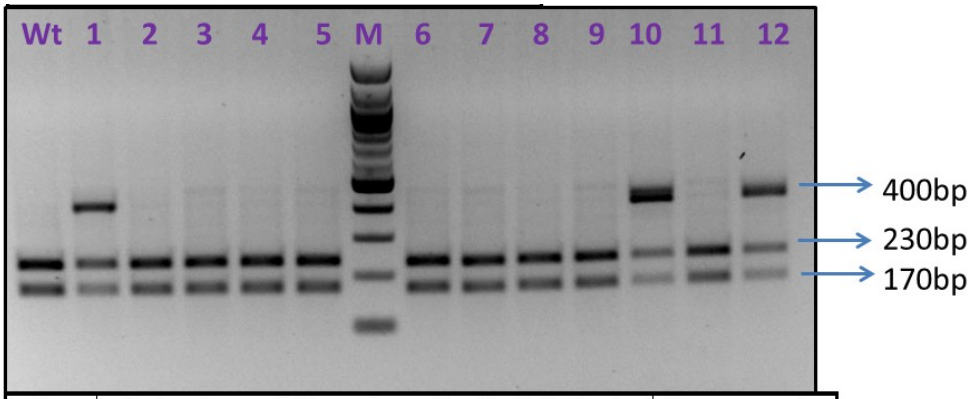


Supplementary Figure 1. Linkage analysis. (**A**) Linkage analysis showing genome-wide linkage intervals reaching a maximum LOD >2 (red line). (**B**) Chromosome 10 linkage map showing the significant interval and location of *TECTB*. (**C**) Gene content of the chromosomal interval on chromosome 10 reaching LOD significance.



Supplementary Figure 2. Exome sequencing visualization of the *TECTB* c.674G>A, p.(Cys225Tyr) variant

A

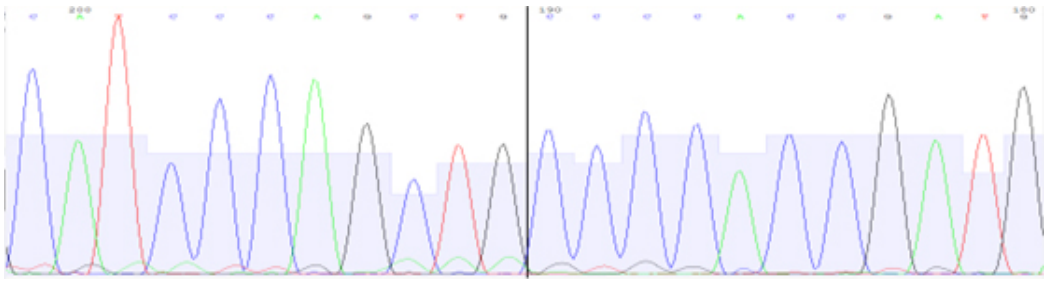


primers	Product size	Wild type after PvuII digestion	Het after PvuII digestion	Homo mutant after PvuII digestion
F1 R1	400	170 + 230	170 + 230 and 400	400

B

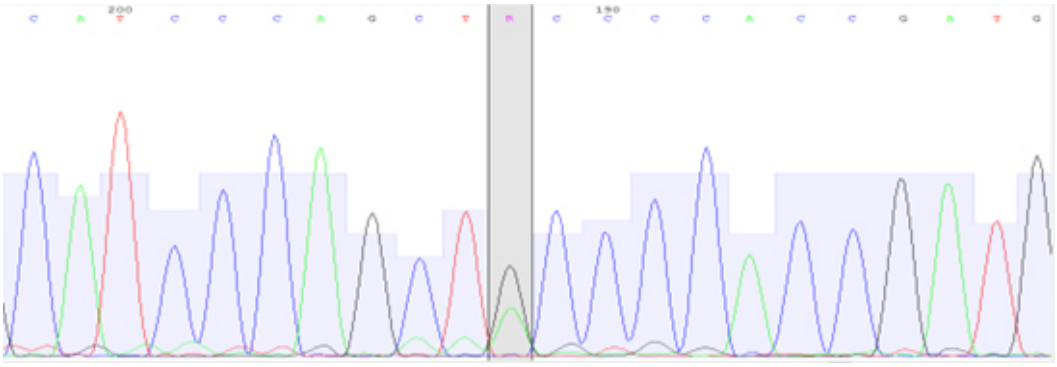
Wild type:

C C C A G C T G C C C C A C C G A T

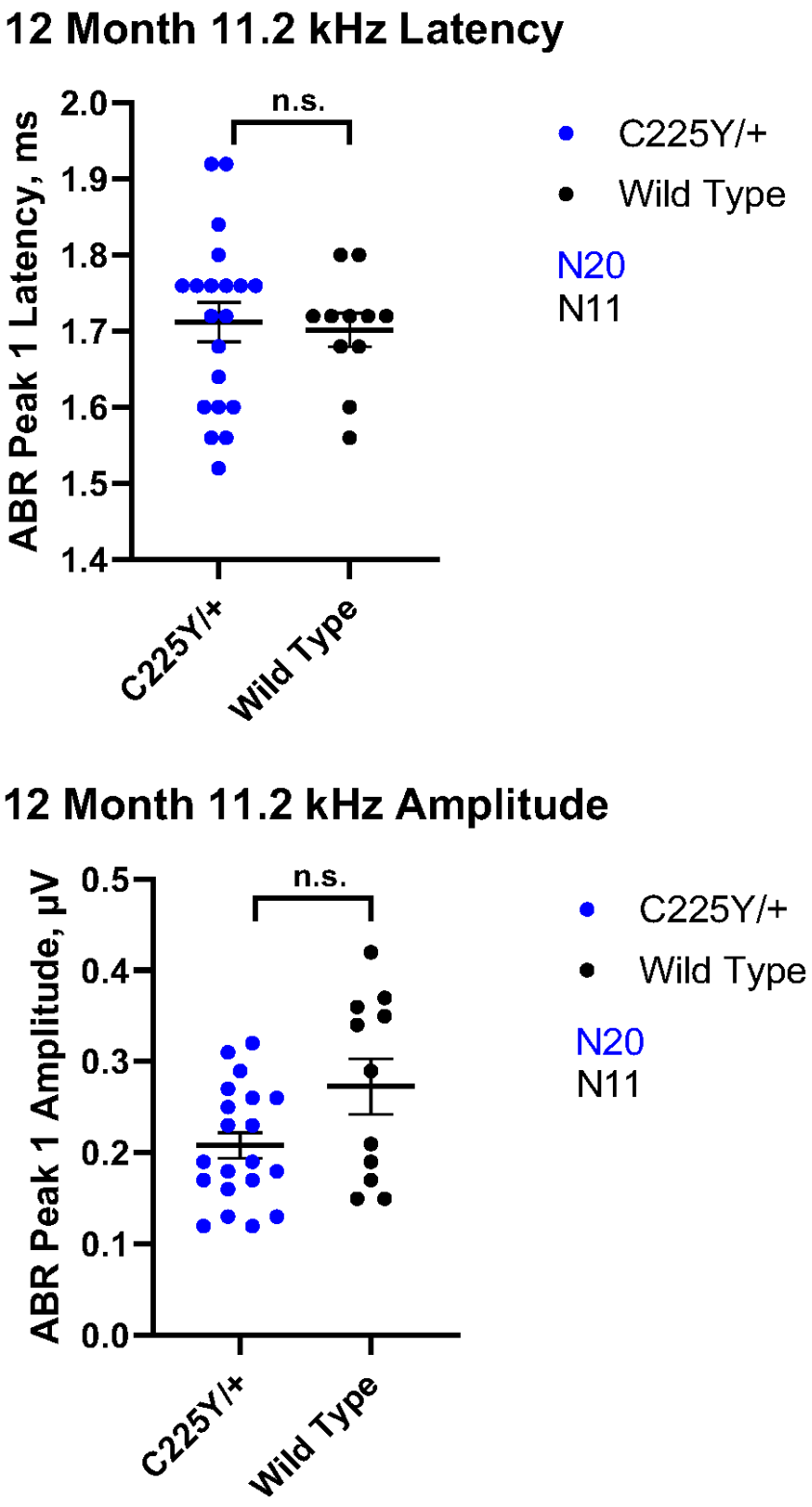


Heterozygous (pup #1):

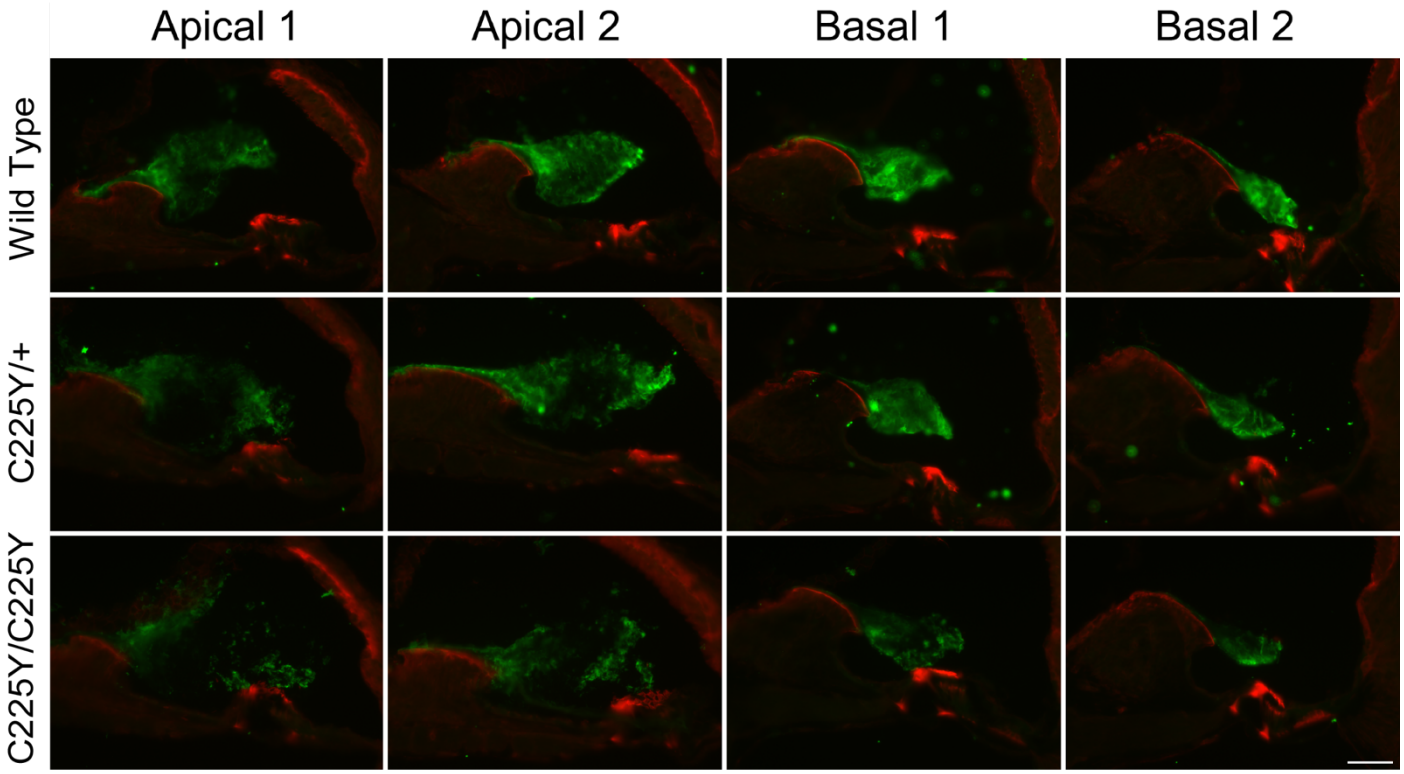
C C C A G C T G/A C C C C A C C G A T



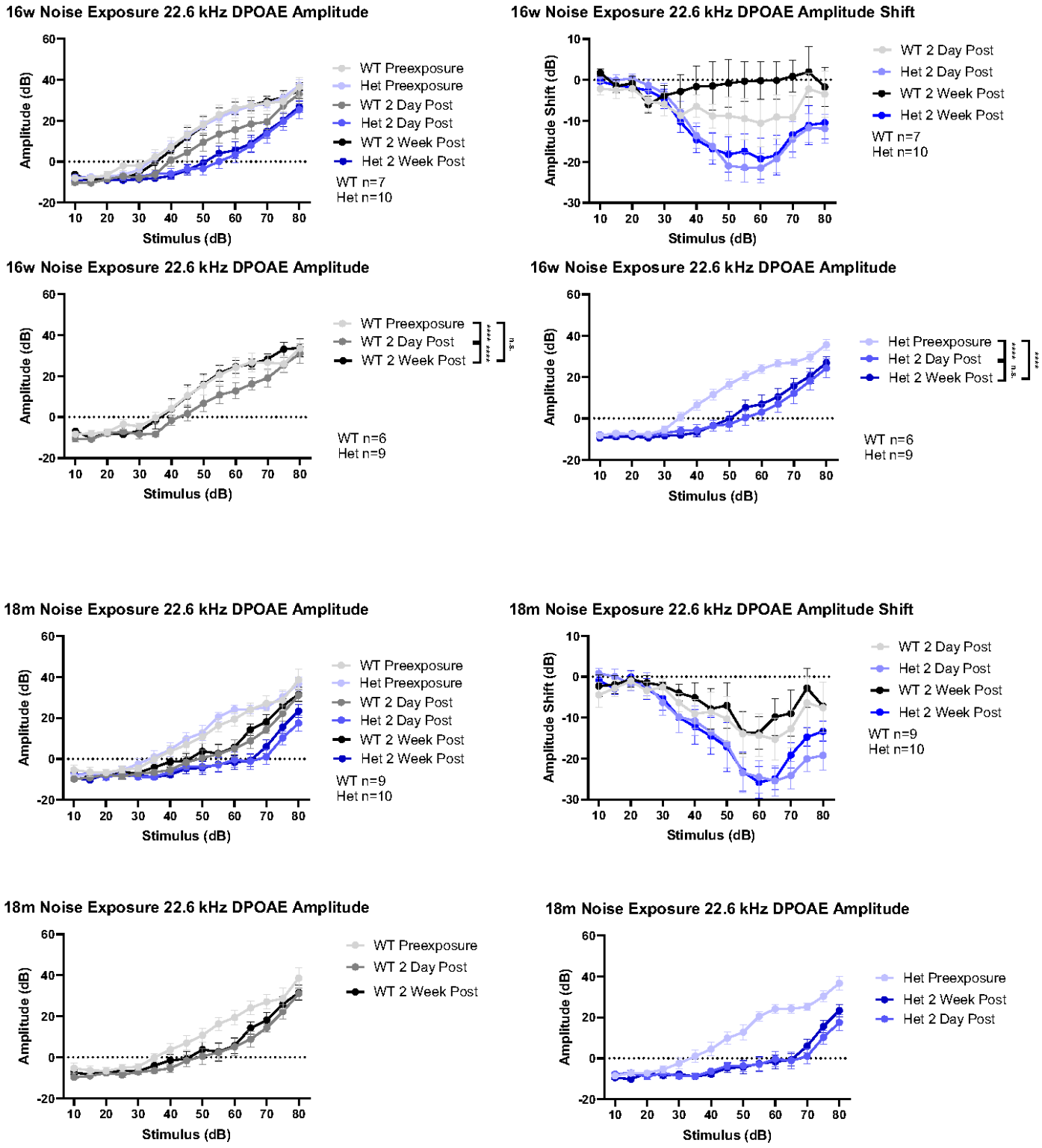
Supplementary Figure 3. (A) Genomic DNA extracted from a wild-type mouse and 12 littermate pups born following CRISPR/Cas-9 mediated genome editing was amplified with primers F1 and R1 and digested with *PvuII* as described in *Material and methods* section. **(B)** Sanger sequencing of F2R2 PCR products from the wild-type and heterozygous (pup #1) littermates shown above in panel *A*. Sequencing confirmed that Pup#1 was correctly modified, while pups #10 and #12 showed evidence of mosaicism.



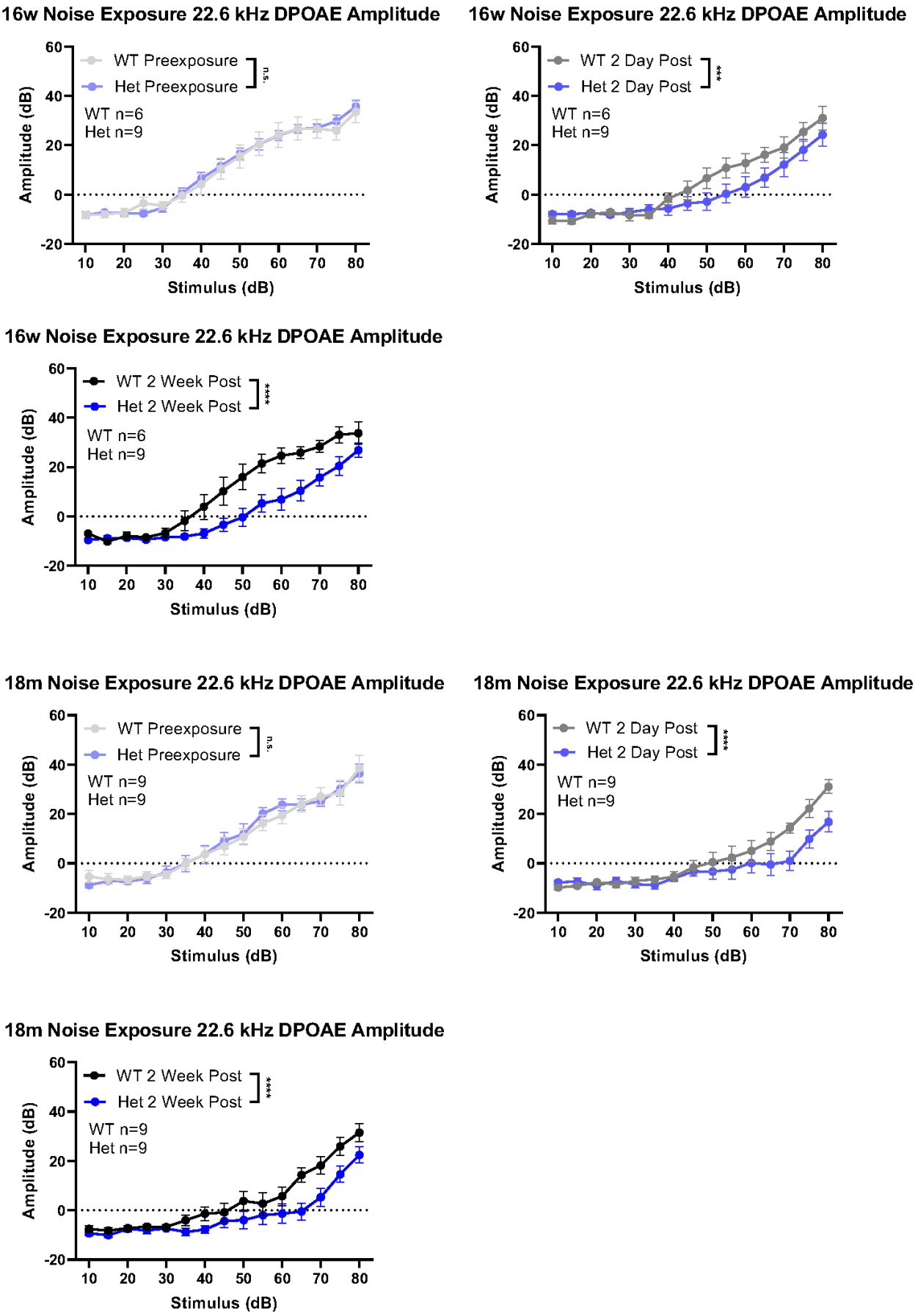
Supplementary Figure 4. ABR peak 1 latency and amplitude measurements in response to 11.2 kHz pure tone stimulation in 12-month-old *Tectb*$^{C225Y/+}$ and wild-type mice. The number of animals per group is indicated within each plot. Data are shown as individual values with the horizontal line representing the mean ± SEM. *Statistical analysis:* Welch's t tests; ns – not significant.



Supplementary Figure 5. TECTB-C225Y protein is detected within the TM. Anti-TECTB (R7) immunolabeling of cochlear cryosections from P80 mice, performed in 4 cochlear locations reveal the presence of anti-TECTB immunoreactivity within the TMs of all three genotypes. These observations indicate that wild-type TECTB and the TECTB-C225Y protein variant are both recognized by the anti-TECTB antibody. *Scale bar* (bottom right), 50 µm.



Supplementary Figure 6. DPOAE peak amplitude growth as a function of sound intensity level for 22.4kHz stimulus measured in WT and *Tectb*$^{C225Y/+}$ mice before noise exposure, 2 days and 2 weeks post-noise. Datasets are grouped to compare the recovery time points. The number of animals per group is indicated within each plot. Data are shown as mean ± SEM. *Statistical analysis:* two-way ANOVA with Tukey's multiple comparisons; ns – not significant, * p<0.05, ** p<0.01, *** p<0.001, **** p<0.0001. For comparison between the genotypes, see *Supplementary Figure 7.*



Supplementary Figure 7. DPOAE peak amplitude growth as a function of sound intensity level for 22.4kHz stimulus measured in WT and *Tectb*$^{C225Y/+}$ mice before noise exposure, 2 days and 2 weeks post-noise, comparing the effect of mouse genotype. The number of animals per group is indicated within each plot. Data are shown as mean ± SEM. *Statistical analysis:* two-way ANOVA with Tukey's multiple comparisons; ns – not significant, * p<0.05, ** p<0.01, *** p<0.001, **** p<0.0001. Same data as in *Supplemental Figure 6*.