**Supporting Information for**

**Early-Onset Hearing Loss in Mouse Models of Alzheimer’s Disease and increased DNA Damage in the Cochlea.**

Jae-Hyeon Park1, Burcin Duan Sahbaz1, Komal Pekhale1, Xixia Chu1, Mustafa N. Okur1, Mhamed Grati2, Kevin Isgrig2, Wade Chien2,3, Elena Chrysostomou4,Lauren Sullivan4,Deborah L. Croteau1,5, Uri Manor4 and Vilhelm A. Bohr1,6\*

**LIST OF SUPPLEMENTARY MATERIALS**

Supplementary Figure S1

Supplementary Figure S2

Supplementary Figure S3

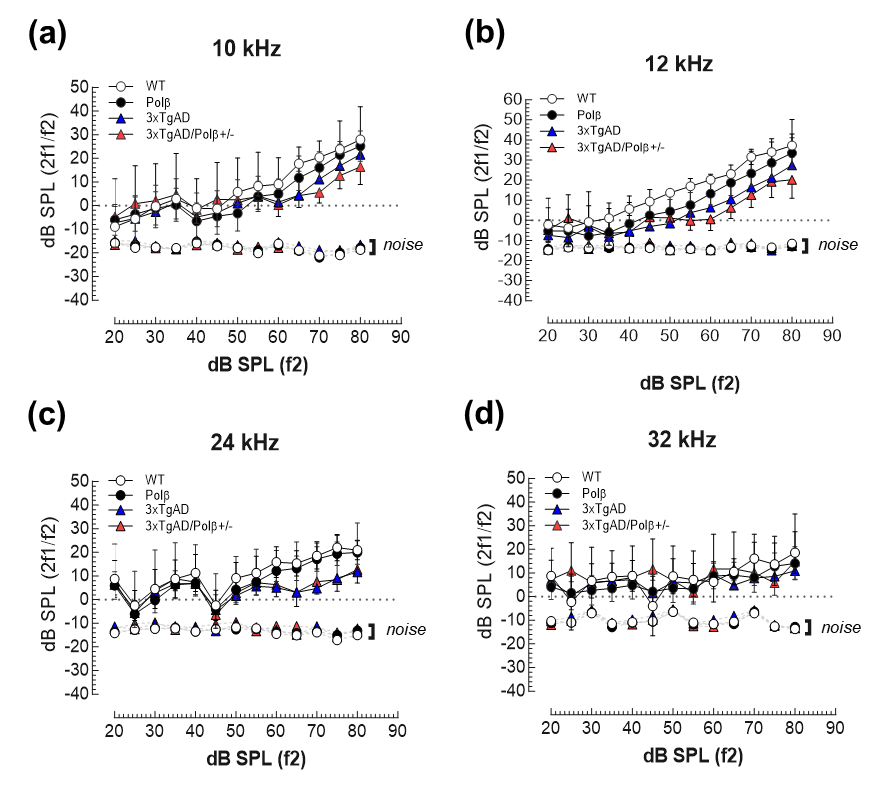
Supplementary Figure S4

Supplementary Figure S5

Supplementary Figure S6

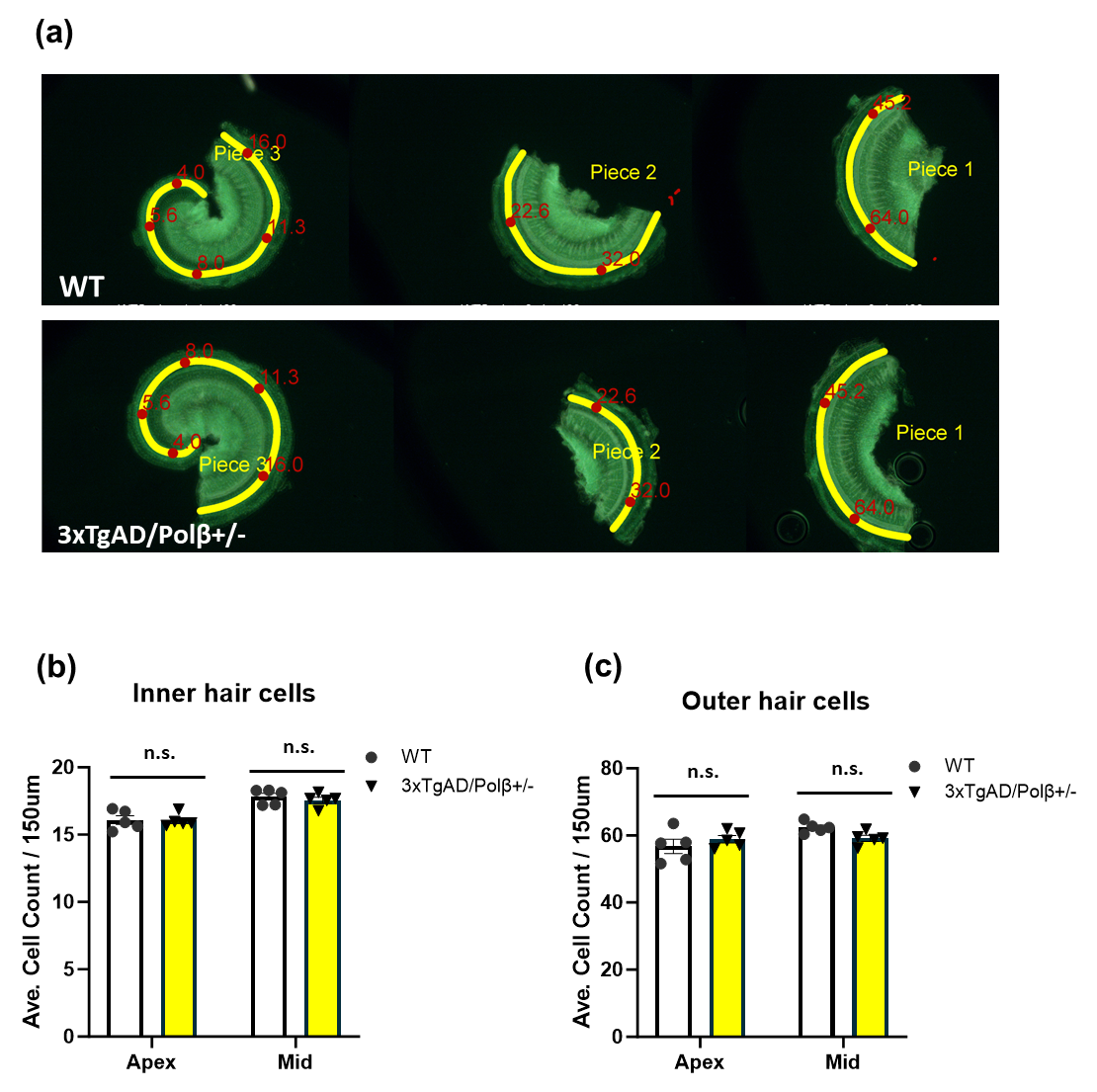
Supplementary Table S1

Fig. S1.



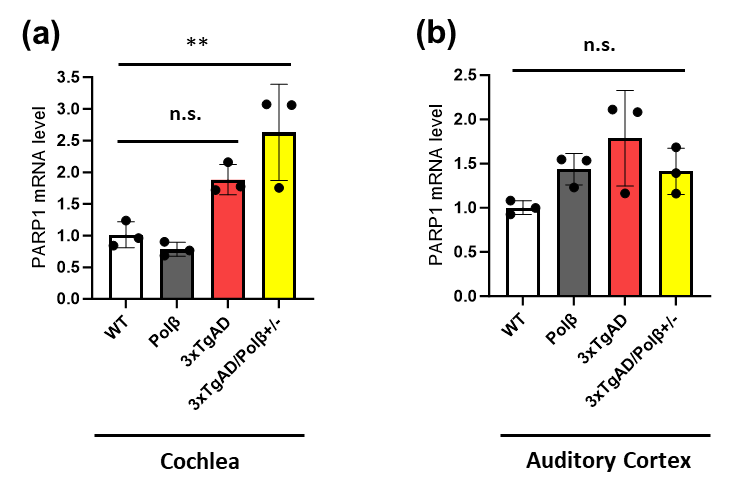
**Supplementary Figure 1.** DPOAE levels were recorded at 4 weeks of age at the multiple frequencies. (a) 10 kHz, (b) 12 kHz, (c) 24 kHz, and (d) 32 kHz. There are no significant reductions in DPOAE signals of WT, Polβ, 3xTgAD, and 3xTgAD/Polβ+/− mice. n = 8-11 mice per group. Two-way ANOVA with Tukey’s multiple comparisons test. Error bars represent the mean ± SD.

Fig. S2.

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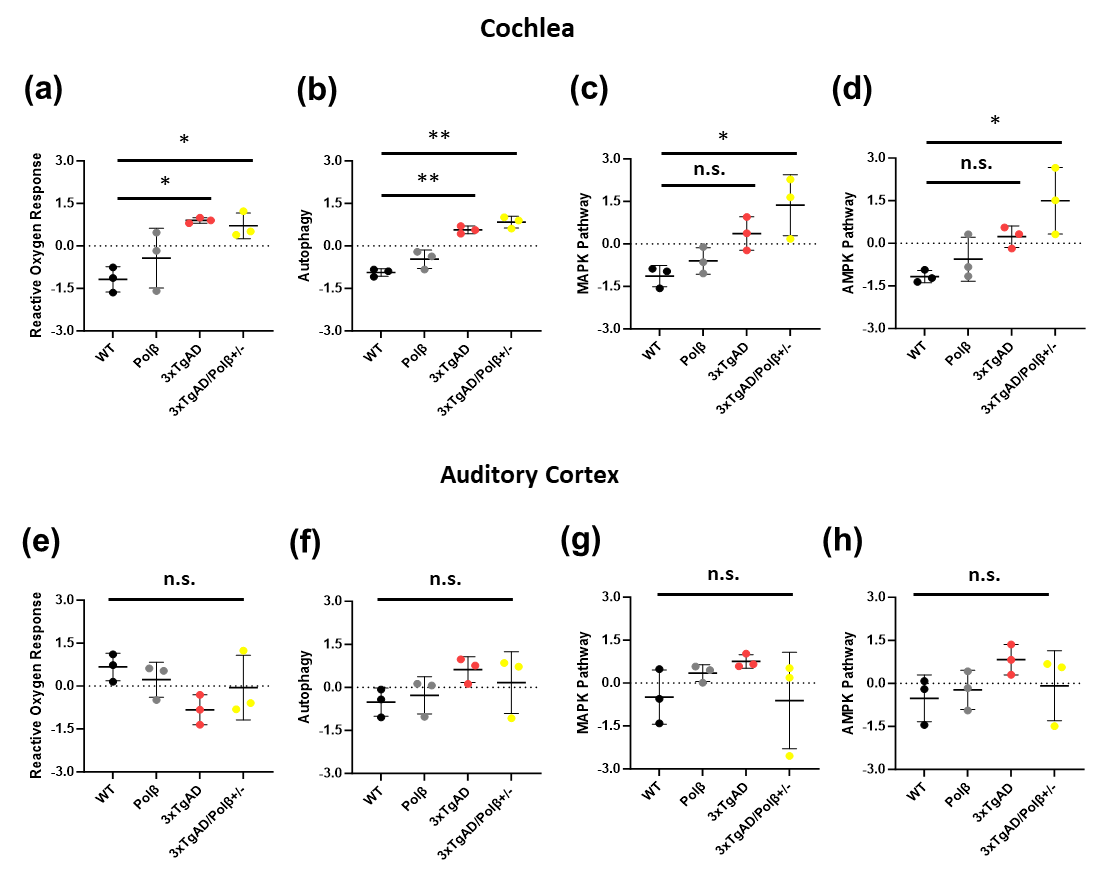
**Supplementary Figure 2.** Quantitative analysis of cochlea inner and outer hair cells in WT 3xTgAD/Polβ+/− male mice. (a) The cochlea frequency map. The average number of inner (b) and outer (c) hair cells per 150 μm in the apex and middle regions of the cochlea in WT and 3xTgAD/Polβ+/− mice at 4 weeks of age. There are no significant changes in the number of inner and outer hair cells from apex and middle regions in 3xTgAD/Polβ+/− mice. n = 5 male mice per group. Two-way ANOVA with Tukey’s multiple comparisons test. Error bars represent the mean ± SD.

Fig. S3.



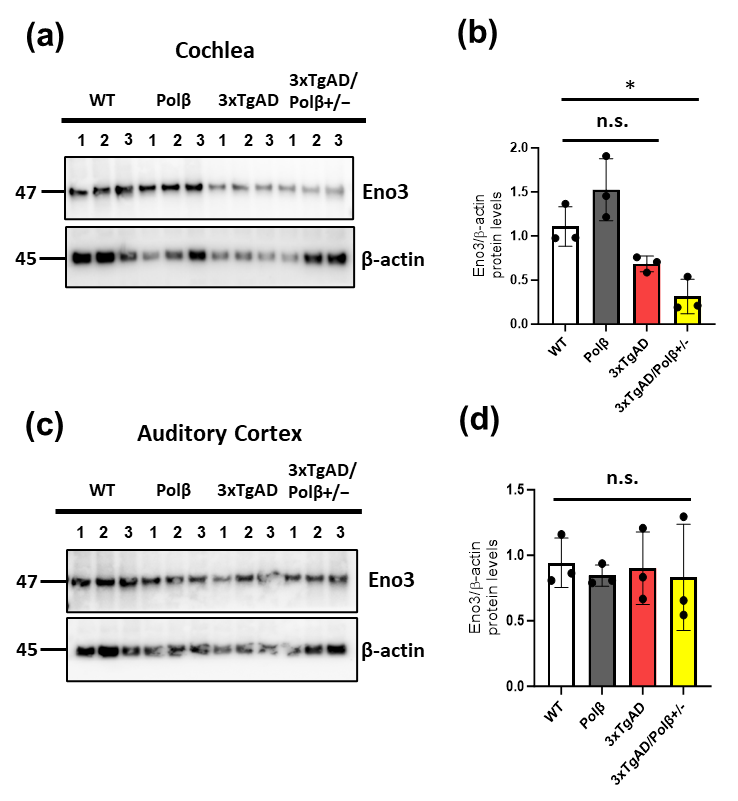
**Supplementary Figure 3.** PARP1 is increased in cochlea of 3xTgAD/Polβ+/− male mice. (a and b) qPCR analysis for the relative gene expression of PAPR1 in the cochlea and auditory cortex. n = 3 male mice per group. \*\*p < 0.01, Two-way ANOVA with Tukey’s multiple comparisons test. Error bars represent the mean ± SD.

Fig. S4.

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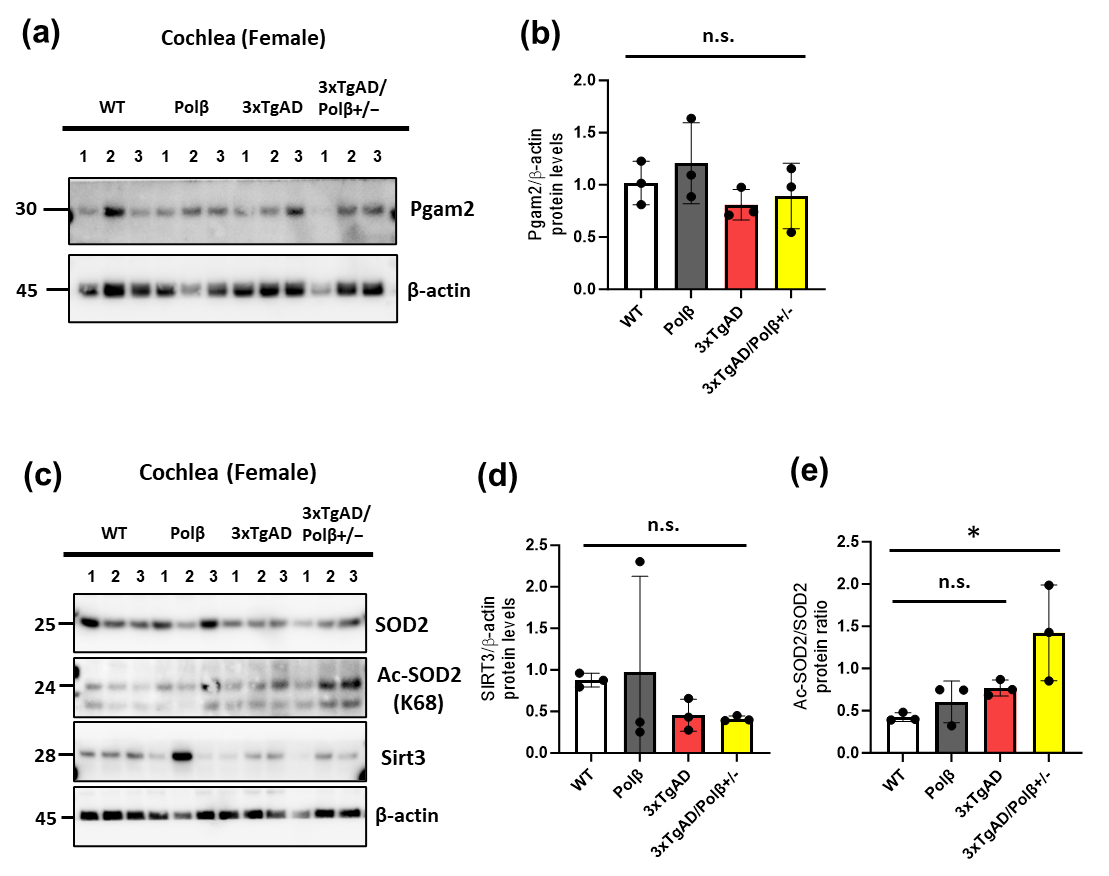
**Supplementary Figure 4.** The pathways associated with cell death & oxidative stress are activated in the cochlea. (a - h) NanoString pathway scores analysis in cochlea and auditory cortex. ROS, autophagy, MAPK, and AMPK signaling pathways were significantly changed in cochlea of 3xTgAD and 3xTgAD/Polβ+/− mice, but there are no significant differences in auditory cortex. n = 3 male mice per group. \*p < 0.05, \*\*p < 0.01, Two-way ANOVA with Tukey’s multiple comparisons test. Error bars represent the mean ± SD.

Fig. S5.



**Supplementary Figure 5.** DNA damage accumulation in cochlea decreased the Eno3 expression in 3xTgAD/Polβ+/− male mice. (a and c) Representative western blots showing Eno3 in cochlea and auditory cortex. (b and d) Quantification of Eno3 protein levels. n = 3 male mice per group. All bands were normalized to loading controls β-actin. \*p < 0.05, Two-way ANOVA with Tukey’s multiple comparisons test. Error bars represent the mean ± SD.

Fig. S6.



**Supplementary Figure 6.** Pgam2 andSIRT3 were not decreased at 4 weeks of age in female mice. (a - c) Representative western blots showing Pgam2, SIRT3, SOD2, and Ac-SOD2 (acetyl K68) in cochlea of female mice. (b - e) Quantification of Pgam2, SIRT3 and Ac-SOD2 (acetyl K68) protein levels. n = 3 female mice per group. Ac-SOD2 band was normalized to total SOD2. The other bands were normalized to loading controls β-actin. \*p < 0.05, Two-way ANOVA with Tukey’s multiple comparisons test. Error bars represent the mean ± SD.

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| **Table. S1.** Antibodies used for western blot (WB). | | |
| **Antibody** | **Source** | **Dilution** |
| PAR (mouse) | Trevigen (4335-MC-100) | 1:1000 (WB) |
| PARP1 (rabbit) | Proteintech (13371-1-AP) | 1:1000 (WB) |
| PGAM2 (rabbit) | Proteintech (15550-1-AP) | 1:1000 (WB |
| TBX2 (mouse) | Santa Cruz (sc-514291) | 1:1000 (WB) |
| ENO3 (rabbit) | Proteintech (55234-1-AP) | 1:1000 (WB) |
| Total H2AX (rabbit) | Cell signaling (7631) | 1:1000 (WB) |
| γH2AX (rabbit) | Cell signaling (9718) | 1:1000 (WB) |
| β-Actin (mouse) | Sigma-Aldrich (A2228) | 1:5000 (WB) |
| SIRT3 (rabbit) | Cell signaling (2627) | 1:1000 (WB) |
| SOD2 (rabbit) | Proteintech (24127-1-AP) | 1:1000 (WB) |
| SOD2 (acetyl K68) (rabbit) | abcam (ab137037) | 1:1000 (WB) |
| Goat anti-mouse HRP | Southern biotech (1010-05) | 1:5000 (WB) |
| Goat anti-rabbit HRP | Southern biotech (4010-05) | 1:5000 (WB) |