**S1 Western blot of cortex samples grouped by age (A-C).** Lamin A/C was used as a loading control. n>3 per group, with technical replicates of each set. 100µg of total protein extract load per sample. (D-F) Graphic representation of Polβ relative to WT protein levels., D-F. *= p<0.05, **=p<0.01.
Microarray analysis of 20 month cortex samples shows that of the BER genes only polβ gene expression is significantly decreased in all samples critically including 3xTgAD. All expression levels are relative to WT. Refer to material and methods and Figure 4.
A circular *in vitro* DNA repair substrate (A) was used to assess BER repair capacity of protein extracts from 20 month brain samples (B). The reduction in Polβ levels conferred reduced insertion activity leading to an overall decrease in repair capacity, B & C. n=3, p>0.05. Refer to materials and methods and Akbari *et al.* (2004) for further information.
A. 6 month cortex results

B. 14 month hippocampal results

S2 No change in APP or tau in the transgenic mice: Representative western blot analysis of human transgenic proteins was conducted on APP and Tau at time points 6 and 14 months respectively (A, B). No differences were seen between the transgenic groups therefore having a DNA repair deficiency does not influence expression of the human transgenes.
S2 (C) (left) Representative full hippocampal western blots of LC3I compared to β-Actin. Shows that the protein level of LC3I is significantly higher in 3xTg/Polβ compared to 3xTgAD. LC3II is not visible at this short exposure time (see figure 2(E)) (n>4). (right) LC3I is significantly higher in the 3xTg/Polβ mice.
S2 (D) The amygdala is associated with anxiety, this behavior is elevated in all 3xTg mouse groups at 6 and 24 months irrespective of polβ status. Polβ animals have elevated anxiety response at 24 months. All groups except WT lose exploratory behavior at 24 months. Error bars on the 6 month Polβ group are an accurate representation of all sets, (n=10-18 per group). (E) Polβ deficient mice spend less time in the open (Z)one compared to residual zone at 24 months. (insert) diagrammatic representation of the residual area (R) versus the zone (Z)
Elevated plus maze results also revealed an anxiety response in the 3xTgAD mouse groups at 6 and 14, but not 24 months. Polβ mice show a high level of anxiety at 24 months correlating with open field data.
S2 (G) Additional images of the of amyloid accumulation in the hippocampus of 14 and 24 month old mice. Refer to Figure 2(F) for further details.
**S2 (H)** Additional images of the amyloid accumulation in amygdala of the 14 month old mice. Refer to Figure 2(G) for further details.
S3 (A) Representative image of full hippocampus showing loss of hippocampal volume is not restricted to the dendate gyrus in 14 month old animals. Refer to Figure 3 for full details.
S3 (B) Morris water maze learning trials showed limited difference between the groups at 14 months of age. Also see Figure 3E.
S3 (C) Probe phase data for memory retention at 14 month shows very similar trends to that done at 20 months. The 3xTg/Polβ mice had poor retention of memory not being able to significantly recall platform location, 4 hours after final learning trial. Quadrant abbreviations: T= Target Quadrant, UR= Upper Right quadrant, LL= lower left, LR= lower right.
S3 (D) Directness of path analysis calculated using 20 month MWM data. Similar trends between mice are seen as in quadrant based analysis (Figure 3F). 72 hours after final training the 3xTgAD/Polβ but not 3xTgAD can only find the platform randomly, (p<0.05). Also see figure S3(C).
Electrophysiology results show no difference at input.

(E) Deficiency Without Affecting Basal Synaptic Transmission at CA1 Hippocampal Synapses. Histogram showing WT mice CA1 neurons exhibit normal basal synaptic transmission and was not altered by Polβ. The amplitude of the fiber volley is plotted against fEPSP slopes. The input/output curves for neurons in slices from WT (n = 10 slices, 4 mice) and Polβ(n = 10 slices, 4 mice) were not significantly different (mean ± SEM, p > 0.05, Student’s t-test).

(F) Paired Pulse Facilitation (PPF) was measured to determine if affects neurotransmitter release from presynaptic terminals at CA1 synapses. PPF is not effected by Pol Beta HT treatment on 3xTgAD. With inter-pulse intervals of 50 ms the values for slices from 3xTgAD and 3xTg/Polβ mice are 1.82 ± 0.05 vs., 1.78 ± 0.06, respectively. (n = 5-6 slices from four pair of mice). Collectively, these findings suggest that Pol Beta HT treatments does not play a major role in basal transmission at CA1 synapses.
### S3, (G)

**Kruskal-Wallis One Way Analysis of Variance on Ranks**

| Group             | N  | Missing | Median | 25%  | 75%  |
|-------------------|----|---------|--------|------|------|
| WT                | 71 | 1       | 165.5  | 165  | 167.125 |
| Polβ              | 71 | 1       | 130    | 128  | 130  |
| 3xTgAD            | 71 | 1       | 127.8  | 121.188 | 128.6   |
| 3xTgAD/Polβ      | 71 | 1       | 108    | 108  | 108  |

\[= 128.373 \text{ with 3 degrees of freedom. (} P = <0.001\)]

The differences in the median values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference \( (P = <0.001) \)

**All pairwise Multiple Comparison Procedures (Tukey Test)**

| Comparison         | Diff of Ranks | q     | P<0.05 |
|--------------------|---------------|-------|--------|
| WT vs. 3xTgAD/Polβ | 10645         | 10645 | Yes    |
| WT vs. 3xTgAD      | 6369          | 6369  | Yes    |
| WT vs. Polβ        | 4562          | 4562  | Yes    |
| Polβ vs. 3xTgAD/Polβ | 6083       | 6083  | Yes    |
| Polβ vs. 3xTgAD    | 1807          | 1807  | No     |
| 3xTgAD vs. 3xTgAD/Polβ | 4276       | 4276  | Yes    |

**S3 (G)** Statistical information to complement LTP results (Figure 3H).
**S4 (A)** Principle component analysis (PCA) of significant genes showed that groups segregated most strongly according to transgenic genotype and then the level of Polβ.
S4 (B) Venn diagram comparison of pathways significantly regulated in the frontal cortex of the groups relative to WT. The 3xTg/Polβ mouse is more similar to the 3xTgAD but is still different from both parental strains.
| Gene      | 3xtg/Polβ | 3xTgAD | Polβ |
|-----------|-----------|--------|------|
| NDUFA1    | -5.81     | -2.652 | -0.42|
| NDUFA10   | -0.707    | -0.324 | -1.054|
| NDUFA2    | -4.698    | -1.647 | -0.673|
| NDUFA3    | -6.054    | -2.533 | -0.615|
| NDUFA4    | -2.606    | -0.117 | -0.364|
| NDUFA5    | -4.171    | -2.204 | -0.696|
| NDUFA6    | -3.572    | -0.583 | 0.085 |
| NDUFA7    | -4.89     | -2.018 | -0.568|
| NDUFA8    | -1.958    | -0.562 | -0.242|
| NDUFA9    | 0.032     | 0.043  | -0.435|
| NDUFA10   | -0.59     | 0.239  | 0.124 |
| NDUFAF1   | 1.457     | -0.47  | 0.846 |
| NDUFB2    | -3.753    | -1.972 | 2.486 |
| NDUFB3    | -4.261    | -2.429 | -0.297|
| NDUFB4    | -2.075    | -2.64  | -1.445|
| NDUFB5    | -1.016    | -1.307 | -0.093|
| NDUFB6    | -2.106    | -3.269 | -0.734|
| NDUFB7    | -3.726    | -2.265 | 0.466 |
| NDUFB8    | -1.71     | 0.605  | -0.484|
| NDUFB9    | -0.665    | -0.995 | 1.442 |
| NDUFC1    | -4.934    | -1.769 | 0.128 |
| NDUFC2    | -2.107    | 0.283  | 0.05  |
| NDUFS1    | 0.627     | 0.684  | 0.502 |
| NDUFS2    | 0.647     | 0.393  | 0.266 |
| NDUFS3    | -0.825    | -0.519 | -0.276|
| NDUFS4    | -1.918    | -1.653 | 0.815 |
| NDUFS6    | -4.333    | -0.833 | -0.549|
| NDUFS7    | -2.739    | -1.365 | -0.339|
| NDUFS8    | -1.389    | -0.241 | 0.354 |
| NDUFV1    | 0.788     | 1.13   | -0.601|
| NDUFV2    | -0.757    | -0.153 | 0.49  |

**COMPLEX 1**
### COMPLEX 3

|          | 3xTg/Polβ | 3xTgAD | Polβ |
|----------|-----------|--------|------|
| UQCRH    | -2.812    | -0.42  | -0.002 |
| UQCRFS1  | -0.036    | -0.88  | -0.706 |
| UQCRC2   | 0.783     | 0.494  | -0.401 |
| UQCRC1   | -0.349    | 0.067  | -0.551 |
| UQCRB    | -3.665    | -3.104 | 0.42  |
| UQCR     | -5.369    | -1.811 | -0.482 |

### COMPLEX 4

|          | 3xTg/Polβ | 3xTgAD | Polβ |
|----------|-----------|--------|------|
| COX10    | 0.351     | 0.736  | 0.693 |
| COX11    | -0.092    | -0.671 | 1.132 |
| COX15    | 0.315     | 0.535  | -0.456 |
| COX17    | -5.399    | -1.934 | -0.703 |
| COX18    | -0.791    | -0.031 | -1.075 |
| COX4I1   | -0.962    | 1.205  | 0.521 |
| COX4I2   | -0.14     | 0.29   | -0.088 |
| COX5A    | -3.067    | -0.667 | -0.157 |
| COX5B    | -3.379    | -0.476 | -0.432 |
| COX6A1   | -3.419    | -0.473 | -0.486 |
| COX6A2   | -6.58     | -3.731 | -0.219 |
| COX6B    | -4.832    | -1.597 | -0.382 |
| COX6C    | -5.286    | -5.038 | -0.356 |
| COX7A1   | -5.248    | -3.905 | -1.513 |
| COX7A2   | -2.982    | -2.521 | 0.12  |
| COX7A2L  | 0.941     | 1.141  | 0.674 |
| COX7B    | -2.455    | -2.044 | -0.991 |
| COX7C    | -4.292    | -1.91  | 0.016 |
| COX8A    | -2.65     | 0.753  | -0.383 |
| COX8B    | -2.412    | -1.141 | 1.344 |
| COX8C    | 0.171     | -0.334 | 0.079 |
S4 (C) Gene expression patterns of complex 1, 3, 4 and 5 show heavy down regulation of principle genes associated with energetic dysfunction. This decrease is most pronounced in the 3xTg/Polβ mice that have a synergistic reduction in these energetic components. Blue arrows correspond to the direction of the gene list compared to the graphs, with the top gene being located closest to the y-axis.
**S4, (D)**

| Pathway                                | Polβ Z-score | 3xTgAD Z-score | 3xTg/Polβ Z-score | p value | p value | p value |
|----------------------------------------|--------------|----------------|------------------|---------|---------|---------|
| REACTOME ELECTRON TRANSPORT CHAIN      | 0            | -5.8538        | -14.8033         | 0.191988| 0.000107| 8.76E-14|
| KEGG OXIDATIVE PHOSPHORYLATION        | -2.3113      | -4.73404       | -14.4702         | 0.002568| 0.001359| 3.79E-12|
| MOOTHA VOXPHOS                         | 0            | -7.17609       | -17.7486         | 0.124994| 4.55E-07| 3.03E-18|
| MOOTHA MITOCHONDRIA                   | 0            | -5.5478        | -8.84446         | 0.068614| 1.70E-06| 7.45E-08|
| REACTOME ELECTRON TRANSPORT CHAIN      | 0            | -5.8538        | -14.8033         | 0.191988| 0.000107| 8.76E-14|

**S4(D)** Table showing both the z score and p-values for pathways associated with oxidative phosphorylation. See Figure 4(D) for graphical representation and further details.
S4 (E) Metabolic studies confirm microarray results showing clear metabolic dysfunction and elevated RER. VO2 (ml/Kg/hour), VCO2 (ml/Kg/hour). Reserve respiratory capacity (RER) is VCO2/VO2 (F) 3xTg/Polβ did not show significant differences in other metabolic parameters with the exception of heat output. All transgenic mice had elevated heat output indicative of metabolic dysfunction, n=4.
We compared most down regulated pathways in the mice and human AD patient pathways and found that out of the 25 pathways in the 3xTg/Polβ (yellow), 20 were also significantly decreased in human AD (purple). When we compare the three mice groups, we clearly see there are different pathways down-regulated in the Polβ (red) and 3xTgAD (blue). These pathways are combined in the 3xTg/Polβ (yellow) making the mice more similar to the human patient array (purple).

| Gene Ontology Term                                      | Human MCI | Human AD | Mouse Polβ | 3xTgAD | 3xTg/Polβ |
|---------------------------------------------------------|-----------|----------|------------|--------|-----------|
| GO0003529 RIBONUCLEOPROTEIN COMPLEX                     | -9.051002288 | -9.82325752 | 0          | -9.302801913 | -17.85645823 |
| GO0003735 STRUCTURAL CONSTITUENT OF RIBOSOME            | -9.27719923 | -11.61301105 | 0          | -7.581829802 | -16.28906262 |
| GO0005840 RIBOSOME                                      | -9.28472801 | -12.09250373 | 0          | -6.908662537 | -15.52512185 |
| GO0008137 NADH DEHYDROGENASE (UBIQUINONE) ACTIVITY      | -3.270447766 | -3.710681966 | 0          | -4.911071043 | -12.57555182 |
| GO0006412 TRANSLATION                                   | -9.09074929 | -8.51186712  | 0          | -5.505080027 | -10.93164686 |
| GO0004129 CYTOCHROME C OXIDASE ACTIVITY                  | 0          | -3.29119567  | 0          | -4.442712111 | -10.5254276  |
| GO0005739 MITOCHONDRIAN                                 | -7.13964338 | 0         | 0          | -4.959024352 | -9.02710416  |
| GO0003954 NADH DEHYDROGENASE ACTIVITY                    | -2.562746565 | -3.73102555 | 0          | -4.439252984 | -7.093906555 |
| GO0002254 RIBOSOME BIOGENESIS AND ASSEMBLY              | -6.249189391 | -6.929502666 | 0          | -8.759042177 | -16.28906262 |
| GO0005743 MITOCHONDRIAL INNER MEMBRANE                  | -5.499601521 | -2.529648974 | 0          | -2.976078803 | -6.654215644 |
| GO0005830 CYTOSOLIC RIBOSOME                             | -6.819570261 | -10.74301106 | 0          | -6.60605106  | -6.520484551 |
| GO0005746 MITOCHONDRIAL RESPIRATORY CHAIN               | -2.90541203 | -2.453604208 | 0          | -3.50811099  | -6.520484551 |
| GO0016071 MRNA METABOLIC PROCESS                         | -1.799058048 | 0         | 0          | -4.3810772   | -6.111818188 |
| GO0045263 PROTON TRANSPORTING ATP SYNTHASE COMPLEX      | -1.697789588 | -2.08200626 | 0          | -1.68779625 | -5.913244367 |
| GO0005843 CYTOSOLIC SMALL RIBOSOMAL SUBUNIT             | -5.006286301 | -6.487637493 | 0          | -5.40329361  | -6.520484551 |
| GO004633 HYDROGEN ION TRANSPORTING ATP SYNTHASE A       | 0          | -4.163160442 | 0          | -2.217204934 | -6.484846428 |
| GO004691 HYDROGEN ION TRANSPORTING ATPASE ACTIVITY      | 0          | -4.473630088 | 0          | -2.135693035 | -5.481804059 |
| GO0016469 PROTON TRANSPORTING TWO SECTOR ATPASE COMPLEX | 0          | -3.872142191 | 0          | -2.043714044 | -4.68566032 |
| GO0016491 OXIDOREDUCTASE ACTIVITY                        | -3.828604201 | 0         | 0          | -4.251825686 | -3.968041945 |
| GO0015986 ATP SYNTHESIS COUPLED PROTON TRANSPORT        | 0          | -3.760498973 | 0          | -3.61794403  | -3.47921232 |
| GO0005389 PROTEASOME CORE COMPLEX                        | -1.636606682 | 0         | 0          | -3.47921232  | -3.47921232 |
| GO0015078 HYDROGEN ION TRANSMEMBRANE TRANSPORTER ACTIVITY | -2.13019159 | -4.966262403 | 0          | -2.42706286 | -3.103386036 |
| GO0016272 PREFOLDIN COMPLEX                              | -1.583410188 | 0         | 0          | -3.054517814 | -2.85874025 |
| GO0015992 PROTON TRANSPORT                              | -2.47751152 | -4.120983703 | 0          | -1.897674323 | -1.874433232 |
| GO0006364 RRNA PROCESSING                               | -1.690056284 | 2.423000173 | 0          | -1.897674323 | -1.874433232 |
| GO0007017 MICROTUBULE BASED PROCESS                      | -2.089744515 | -1.694204162 | 0          | -1.897674323 | -1.874433232 |
S4 (H) The 25 pathways from figure S4G were taken and compared, individual pathway z-scores were analyzed by one-way ANOVA with tukey multiple comparison test. The 3xTg/Polβ was not significantly different from Human AD in these pathways. The 3xTg/Polβ mouse was very different from both 3xTgAD and Polβ. * p<0.05, ** p<0.01, *** p<0.005, **** p<0.001.
S4, (H) Using a second human patient AD array data set (refer to results) derived from human AD fibroblasts we again show that the 3xTg/Polβ mouse have more pathway similar to this human AD data set than the 3xTgAD or Polβ alone.