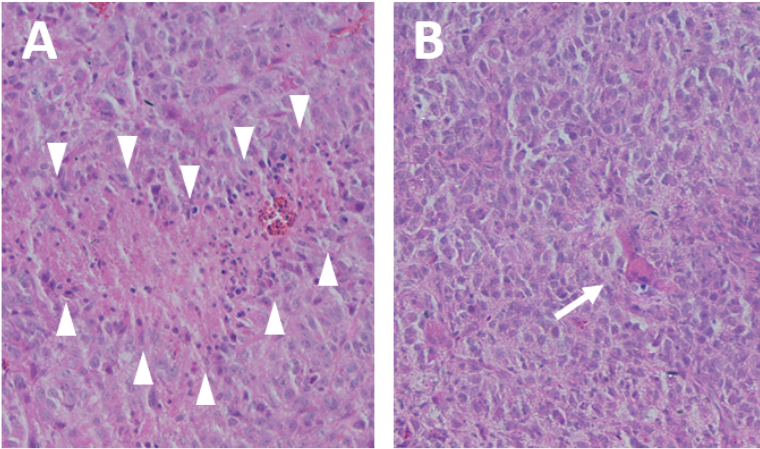


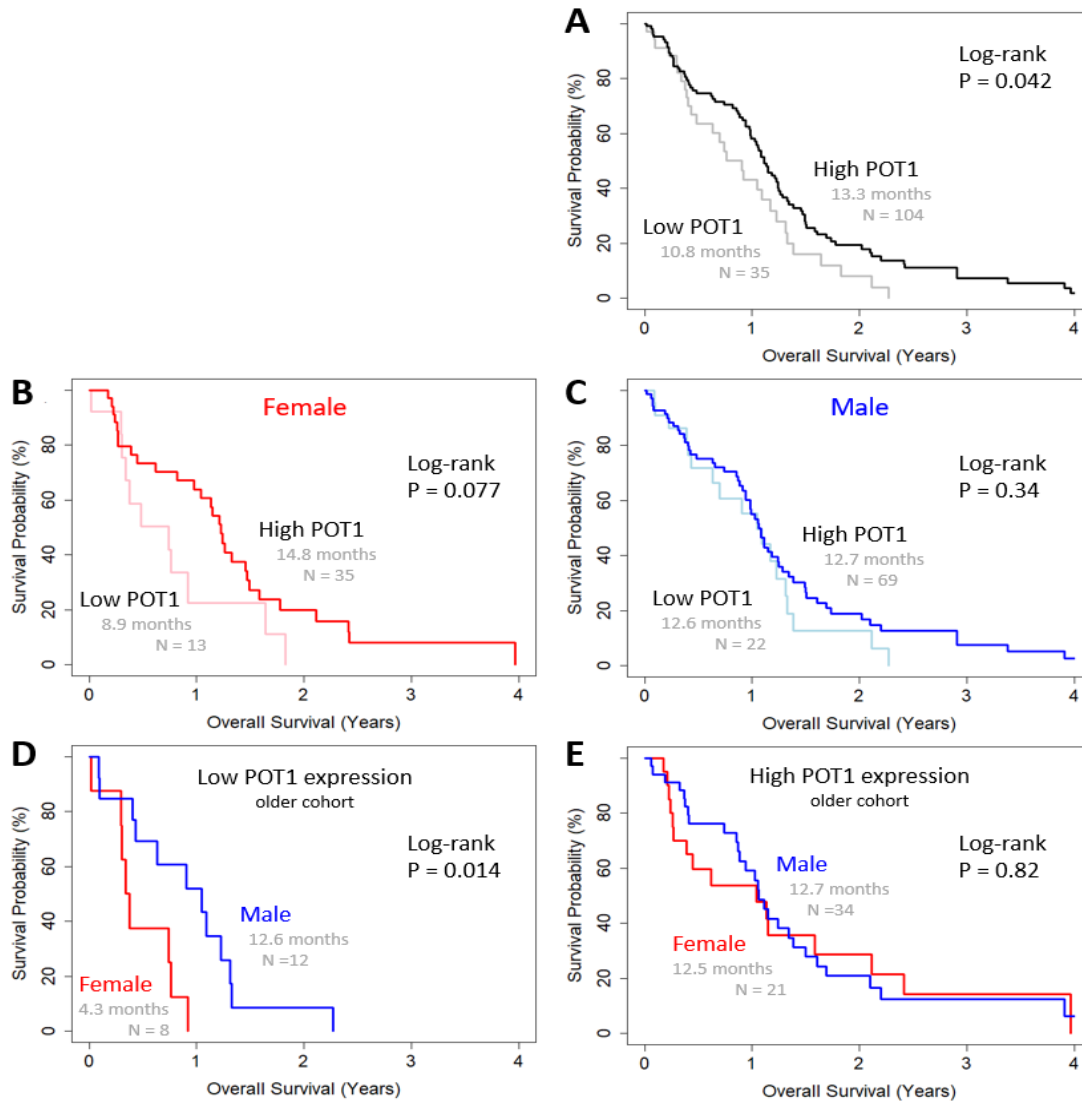
Supplementary Figures

Jalali, *et al.* **POT1 regulates proliferation and confers sexual dimorphism in glioma**



Supplementary Figure 1. C5 glioma tumors share the aggressive histological features of C3 tumors.

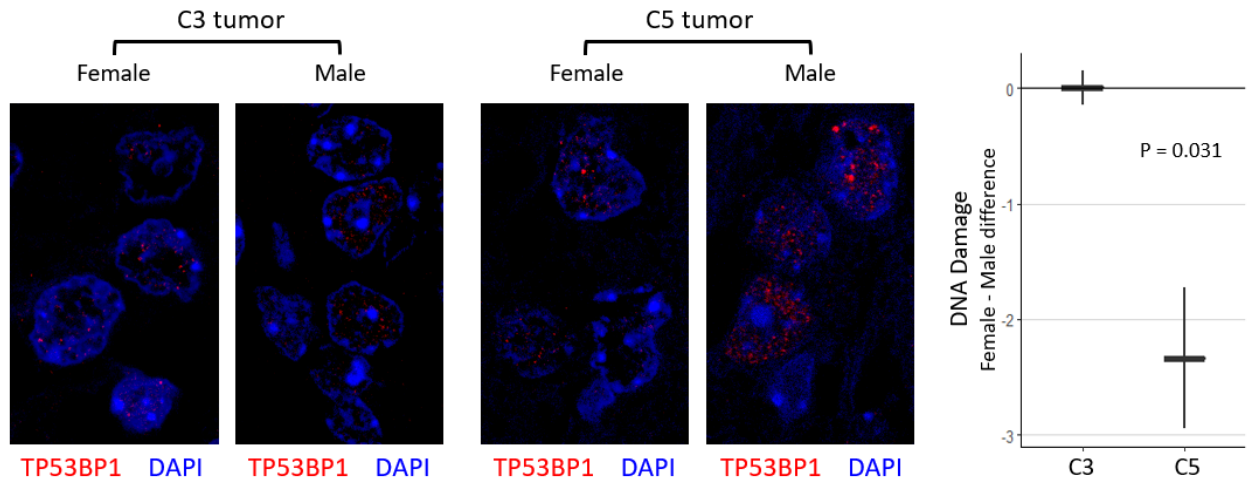
(A) H&E section of C5 tumors showing an area of necrosis surrounded by pseudopalisading tumor cells (arrowheads). (B) H&E section of C5 tumors showing an area of vascular endothelial proliferation (arrow). C3 tumors contain knockouts of Trp53, Nf1, and Pten. C5 tumors contain additional knockouts of Pot1a and Pot1b.



Supplementary Figure 2. Kaplan-Meier Survival Analysis of Grade IV IDH-Wildtype

Glioma (Glioblastoma)

(A) Kaplan-Meier plot showing a lower median survival in glioma patients with low POT1 expression tumors. (B-C) Female patients with low POT1 expression tumors have a more drastic reduction in survival. (D-E) In the older cohort (>61 years old), female patients with low POT1 expressing tumors have a shorter survival than males while the survival of patients with high POT1 expressing tumors is not sex-dependent.



Supplementary Figure 3. Loss of POT1 in mouse glioma has sex-dependent

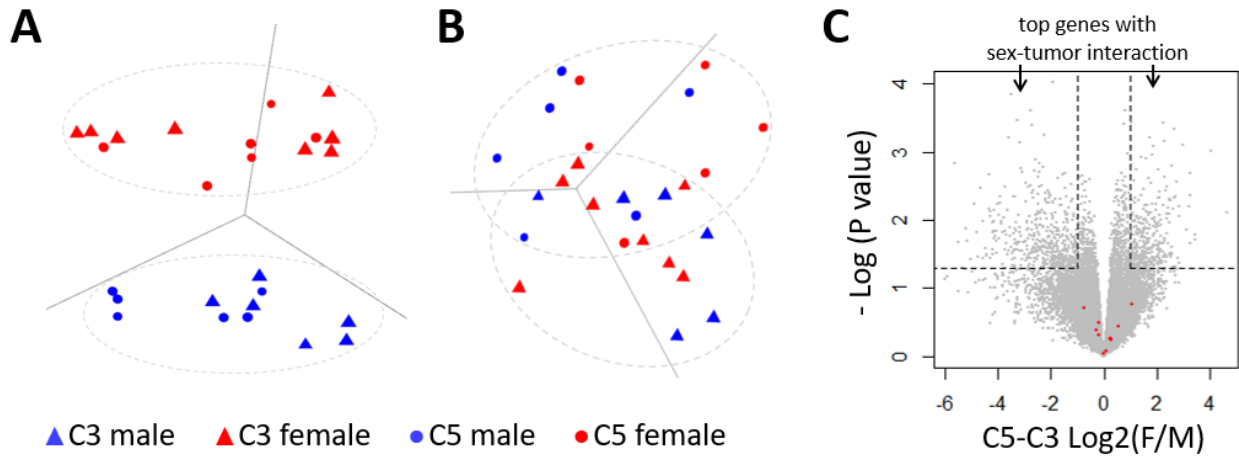
effects on DNA damage. IHC signal from TP53BP1, a marker of DNA double-strand

break, was used as a surrogate for DNA damage. Quantification of the results show that

female-male difference in DNA damage is lower in C5 tumors. Plots show mean \pm

standard error, and units are arbitrary. C3 tumors contain knockouts of Trp53, Nf1, and

Pten. C5 tumors contain additional knockouts of Pot1a and Pot1b.



Supplementary Figure 4. Analysis of RNA Sequencing Transcriptomic Data from

C3 and C5 Tumors (A) RNA sequencing of C3 and C5 tumor samples and 3D Principal

Component Analysis (PCA) using the 300 most variable genes shows clear separation

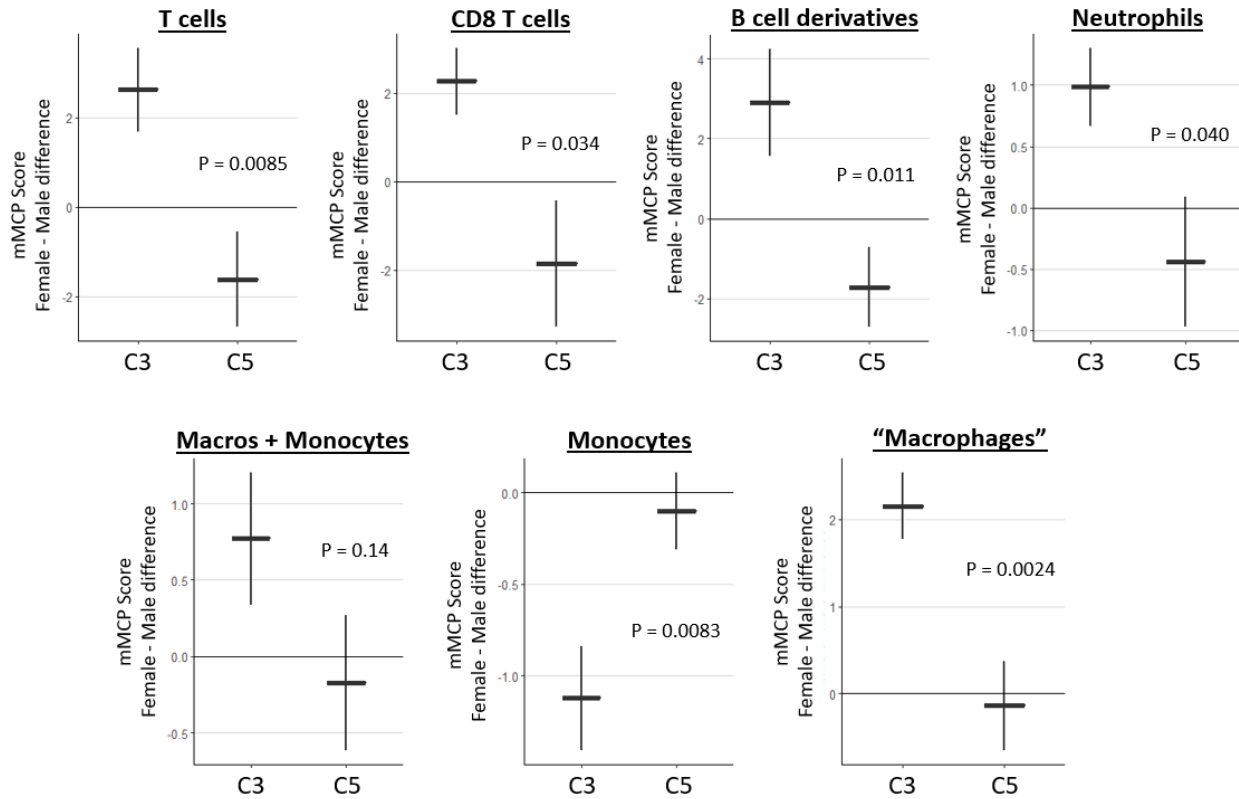
of samples by sex. (B) 3D-PCA using the next 1000 most variable genes shows

overlapping clusters based on tumor type. (C) Volcano plot for sex-tumor interaction.

Log₂(F/M): Log₂(female/male gene expression ratio). Dashed lines demarcate top

genes with sex-tumor interaction. Genes on the Y chromosome, which have a fully sex-
dependent expression, are marked in red, showing no significant sex-tumor interaction.

C3 tumors contain knockouts of Trp53, Nf1, and Pten. C5 tumors contain additional
knockouts of Pot1a and Pot1b.



Supplementary Figure 5. Sex-dependent effects of Pot1a/b loss on different

immune cell lineages mMCP-counter analysis on RNA sequencing data from C3 and

C5 tumors confirms previous immune findings. There is a general decrease in female-

male difference in immune infiltration of tumors as a result of Pot1a/b loss. However,

Monocytes, generally immune-suppressive, show an increase in female-male

difference. "Macrophages" refers to the score difference between

Macrophages/Monocytes and Monocytes. C3 tumors contain knockouts of Trp53, Nf1,

and Pten. C5 tumors contain additional knockouts of Pot1a and Pot1b.