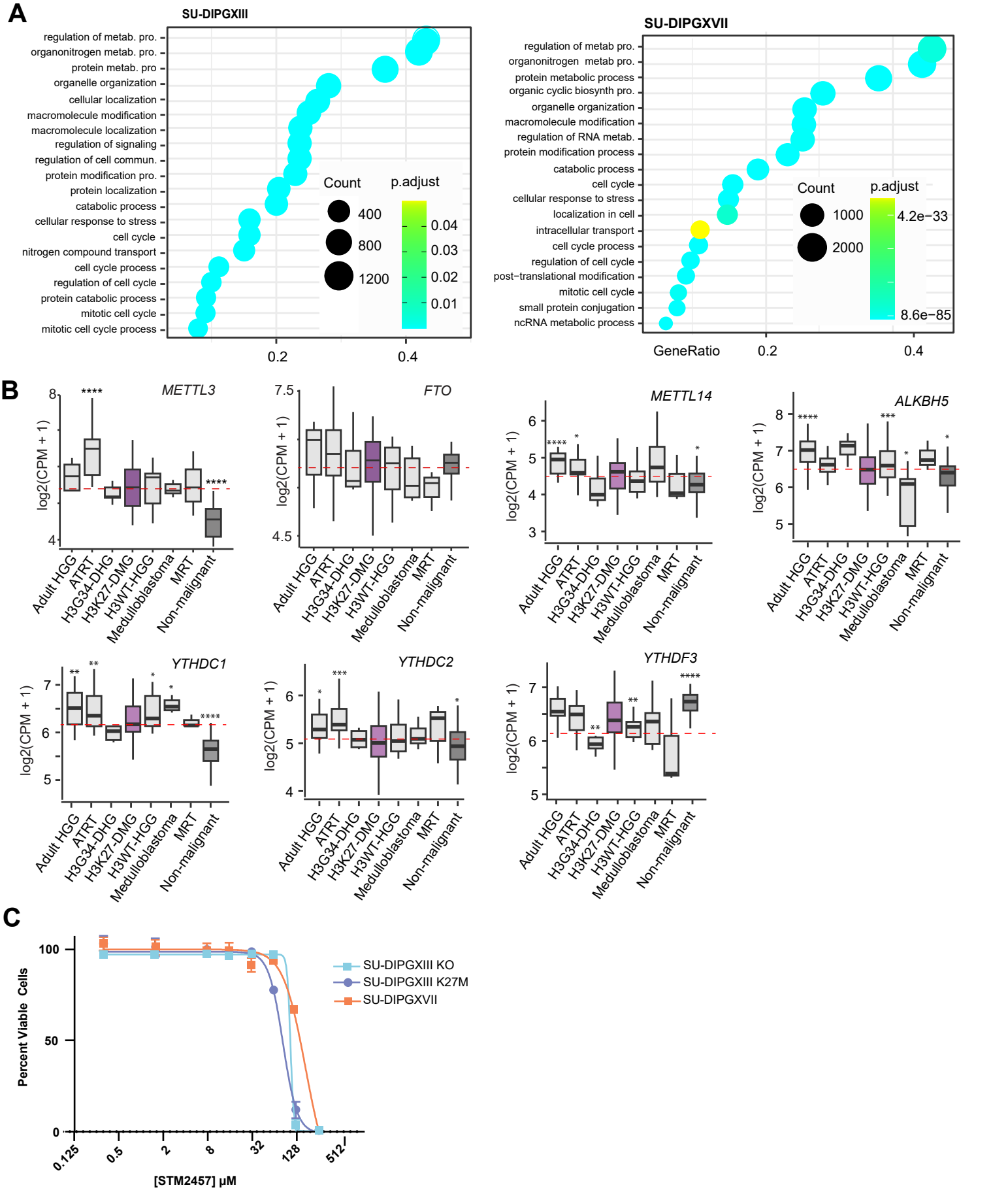
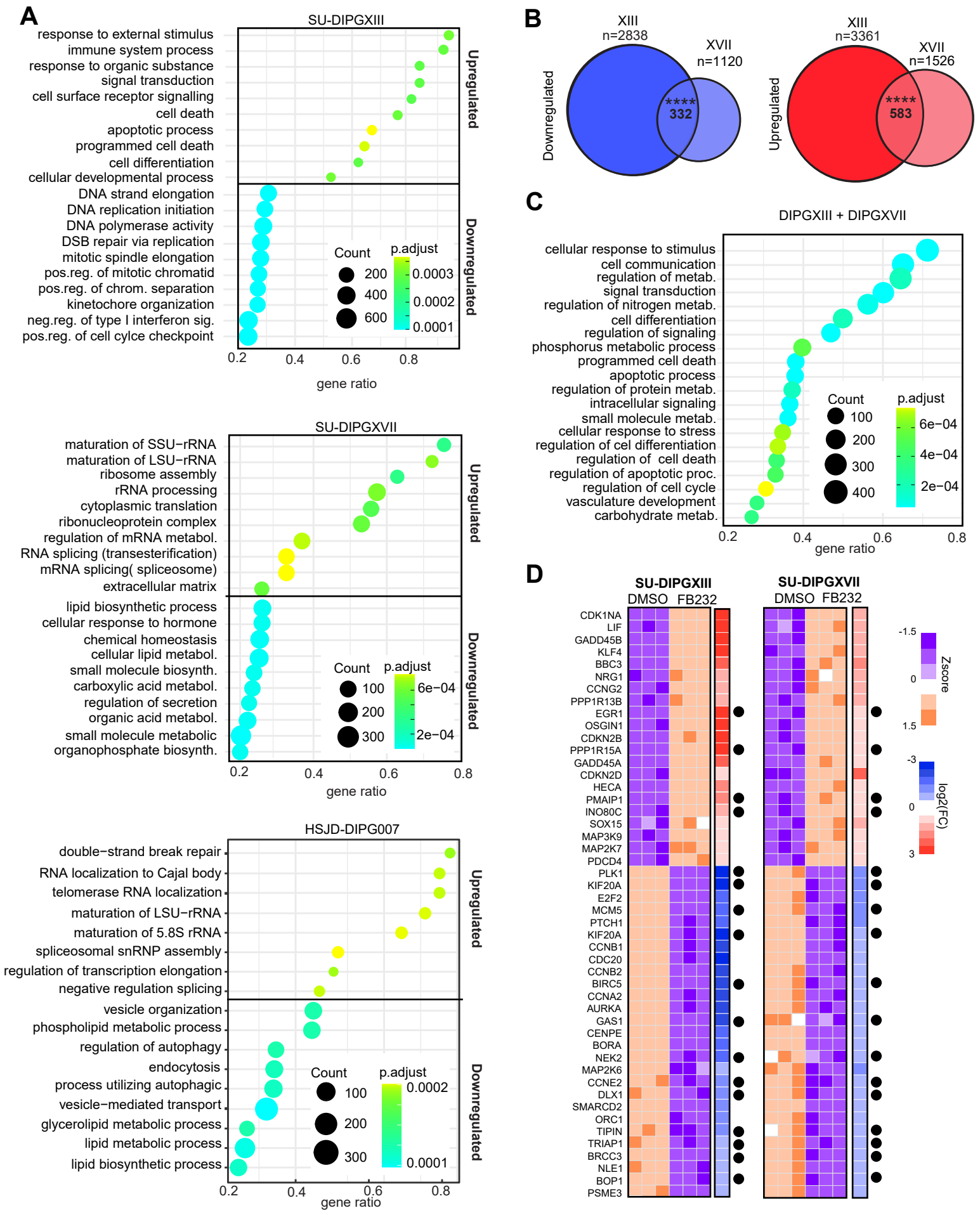
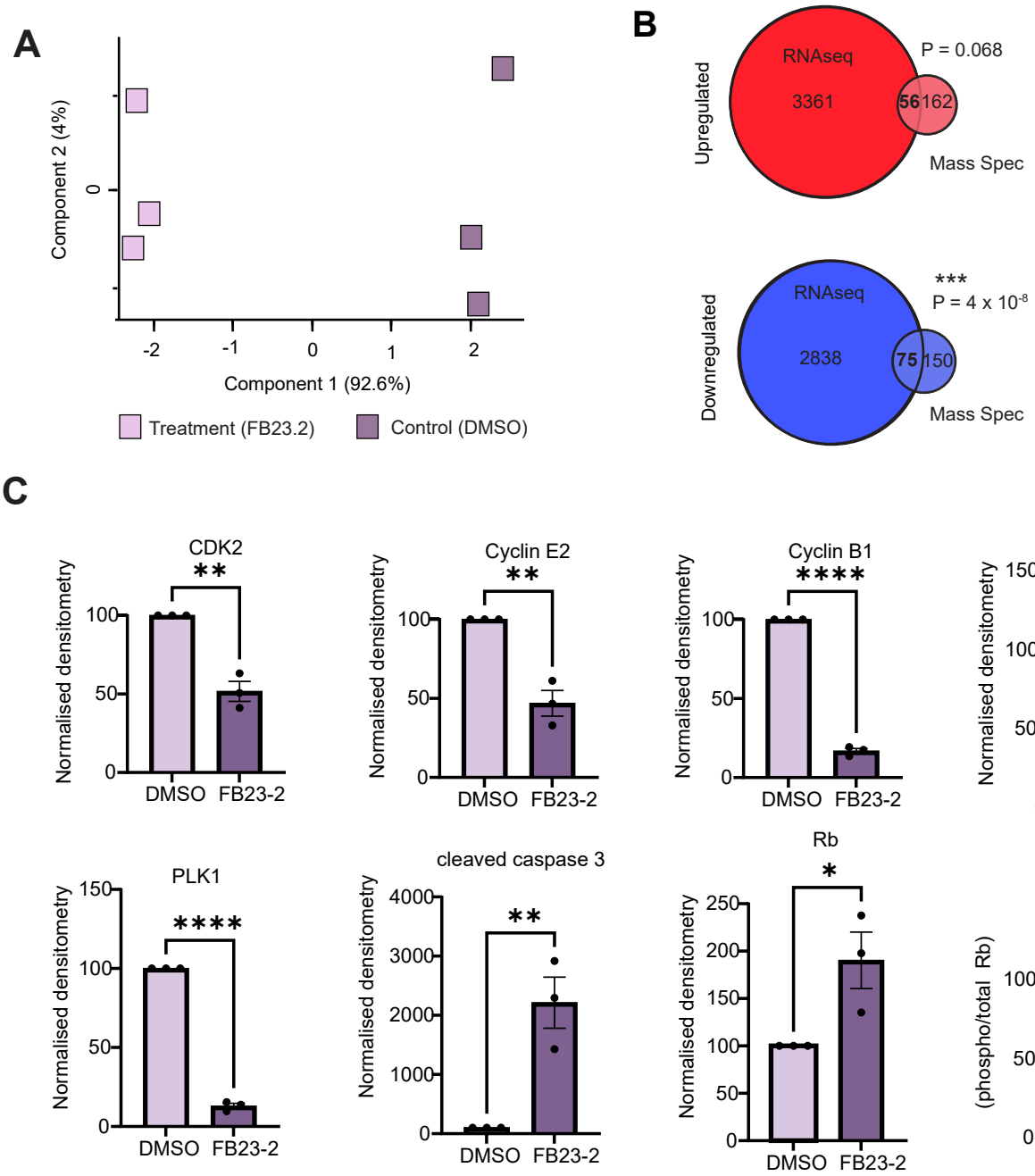


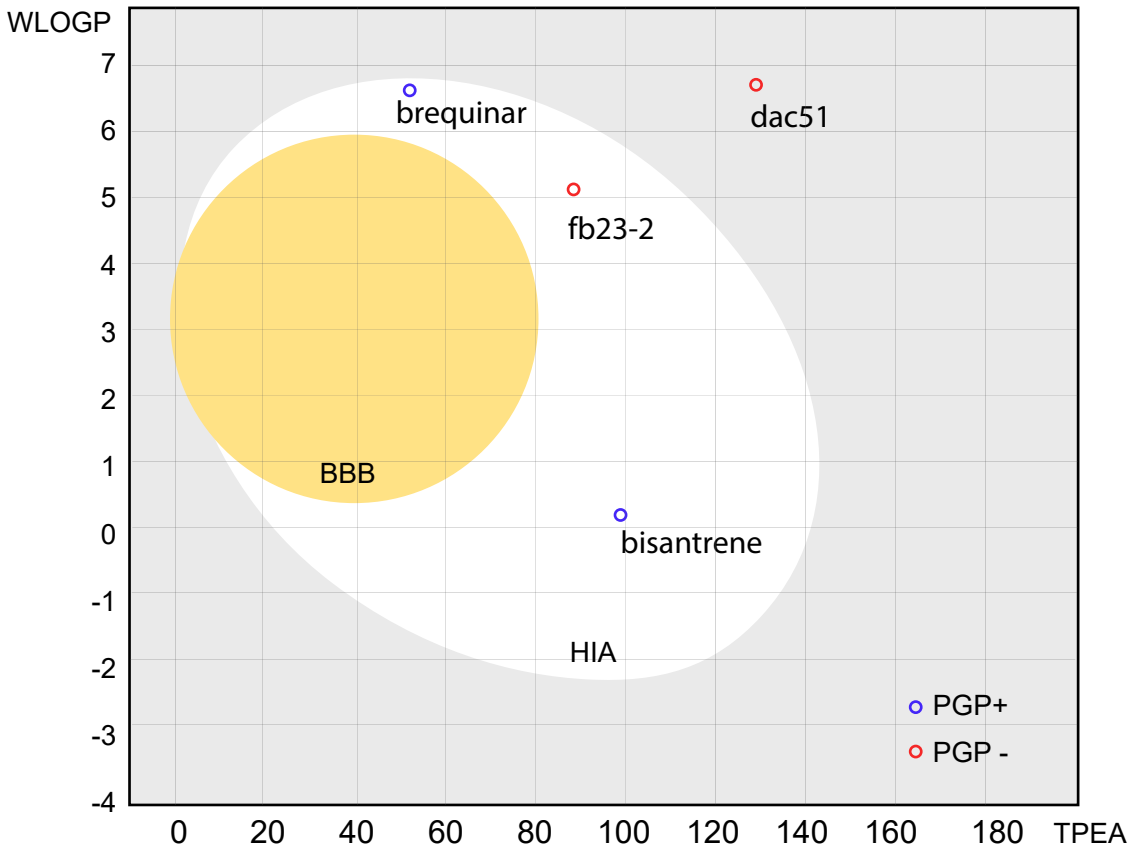

**Supplemental Figure 1. A)** Gene Ontology enrichment analysis of transcripts modified by m6A (>90% probability; >20 reads) in DMG cell lines SU-DIPGXIII (Top) and SU-DIPGXVII (bottom). **B)** Average transcript levels (log2(CPM+1) of m6A readers, writers, erasers in different brain cancers and non-malignant cells. Adult high grade gliomas (Adult HGG; n=16), typical teratoid rhabdoid tumors (ATRT; n=19), H3G34-mutant diffuse hemispheric gliomas (H3G34-DHG; n=6), H3K27-altered diffuse midline gliomas (H3K27-DMG; n=51), H3WT - high grade gliomas (H3WT-HGG; n=19), Medulloblastoma (n=7), malignant rhabdoid tumors (n=7), non-malignant (n=39). The boxplots show the median, first and third quartiles with the whiskers extended to the last point within 1.5X of the interquartile range. Red dotted line indicates cohort median. P-value for significant difference from median shown: * < 0.05, ** < 0.01, *** < 0.001, **** < 0.0001[46]. **C)** Viability of cell lines following 24 hours of STM2457 treatment (0.3 µM, 1.6 µM, 7.8 µM,15.6 µM, 31.2 µM, 62.5 µM, 125 µM, 250 µM). Data is represented as the mean of 3 biological replicates with standard error shown. Patient derived DMG lines: SU-DIPGXVII and SU-DIPGXIII; Isogenic K27M KO: SU-DIPGXIII K27M KO




**Supplemental Figure 2. A)** Gene Ontology enrichment analysis of differentially expressed transcripts after FB23-2 treatment from SU-DIPGXIII (top), SU-DIPGXVII (middle) and HSJD-DIPG007 (bottom). **B)** Overlap between genes differentially upregulated (top) and downregulated (bottom) in DIPGXIII vs DIPGXVII after FB23-2 treatment. ****p<0.001. **C)** Gene Ontology enrichment analysis of genes commonly upregulated and downregulated in SU-DIPGXIII and SU-DIPGXVII after FB23-2 treatment. **D)** Heatmap of log2FC and Zscore values for a subset of genes involved in cell cycle regulation (GO:0051726) and apoptotic regulation (GO: 0042981), which are differentially expressed in both SU-DIPGXIII and SU-DIPGXVII following FB23-2 treatment. Genes identified as having m6A modification sites in DMG are indicated by black circles.




**Supplemental Figure 3. A)** Principal Component Analysis of protein levels in SU-DIPGXIII cells after 24 hour treatment with either DMSO or 8μM FB23-2 treatment. **B)** Overlap between transcripts and proteins differentially upregulated (top) and downregulated (bottom) in SU-DIPGXIII after FB23-2 treatment. **C)** Densitometry analysis of protein levels following 24 hour treatment of 8μM FB23-2 in SU-DIPGXIII. pRB = phosphorylated Rb (retinoblastoma protein); S807/811= serine 807/811. $p<0.05$, $^{**}p<0.01$, $^{***}p<0.001$, $^{****}p<0.001$.




**Supplemental Figure 4.** BOILED-egg plot of potent FTO inhibitor