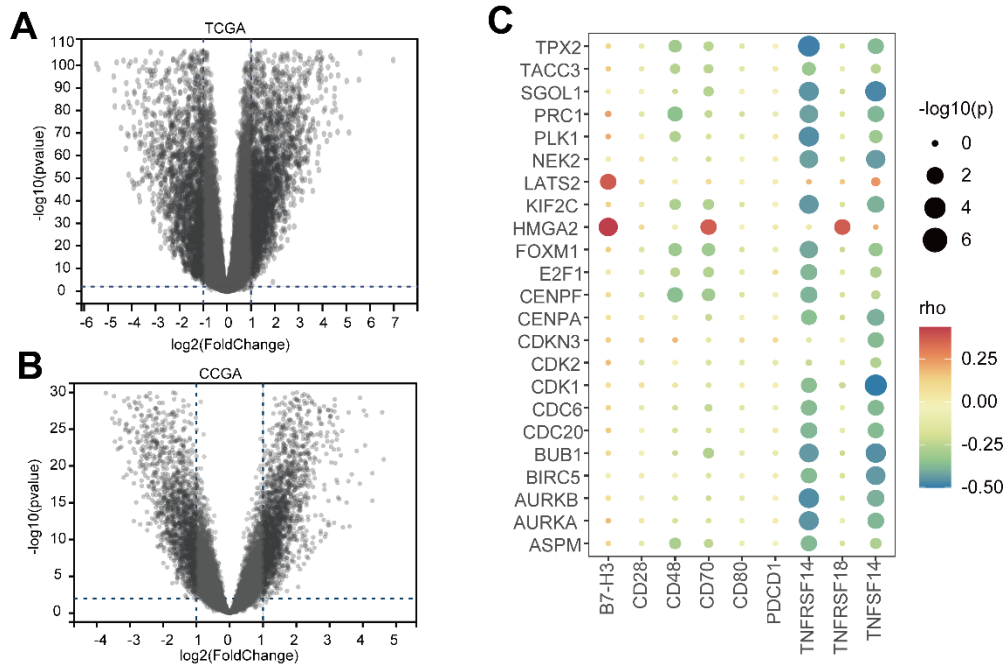


1 Supplementary Figure 1



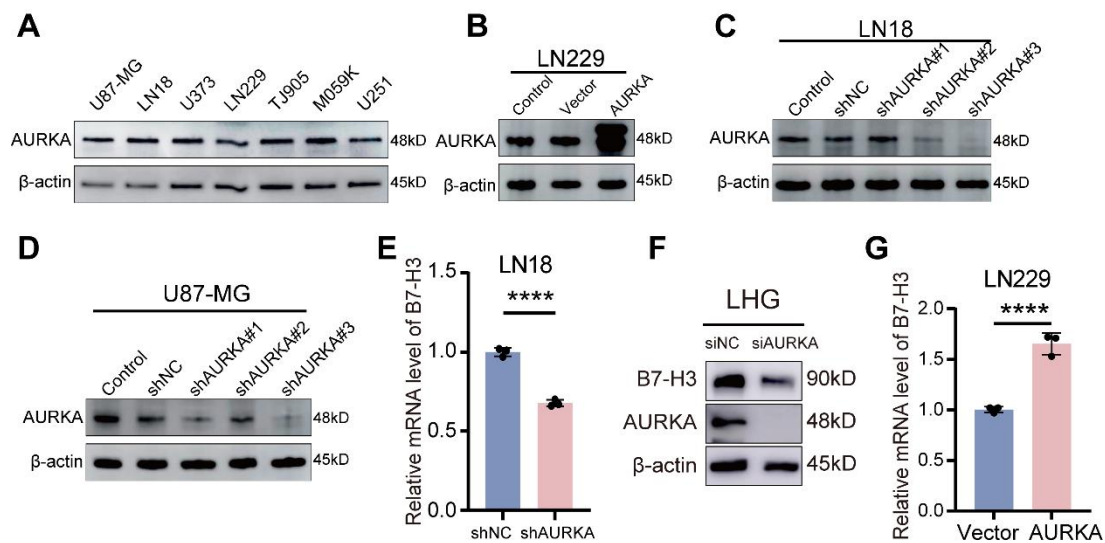
2

3 **Supplementary Figure 1 (A and B)** Volcano plots of the DEGs between  
 4 GBM (WHO-IV and IDH wild type) and LGG (WHO-II and IDH mutant)  
 5 based on the TCGA **(A)** and CGGA **(B)** databases. **(C)** Heatmaps  
 6 displaying the correlations in the TCGA datasets between cell cycle genes  
 7 among the DEGs in both the TCGA and CGGA cohorts and immune  
 8 checkpoint genes among the DEGs in both the TCGA and CGGA cohorts.

9

10

11 **Supplementary Figure 2**



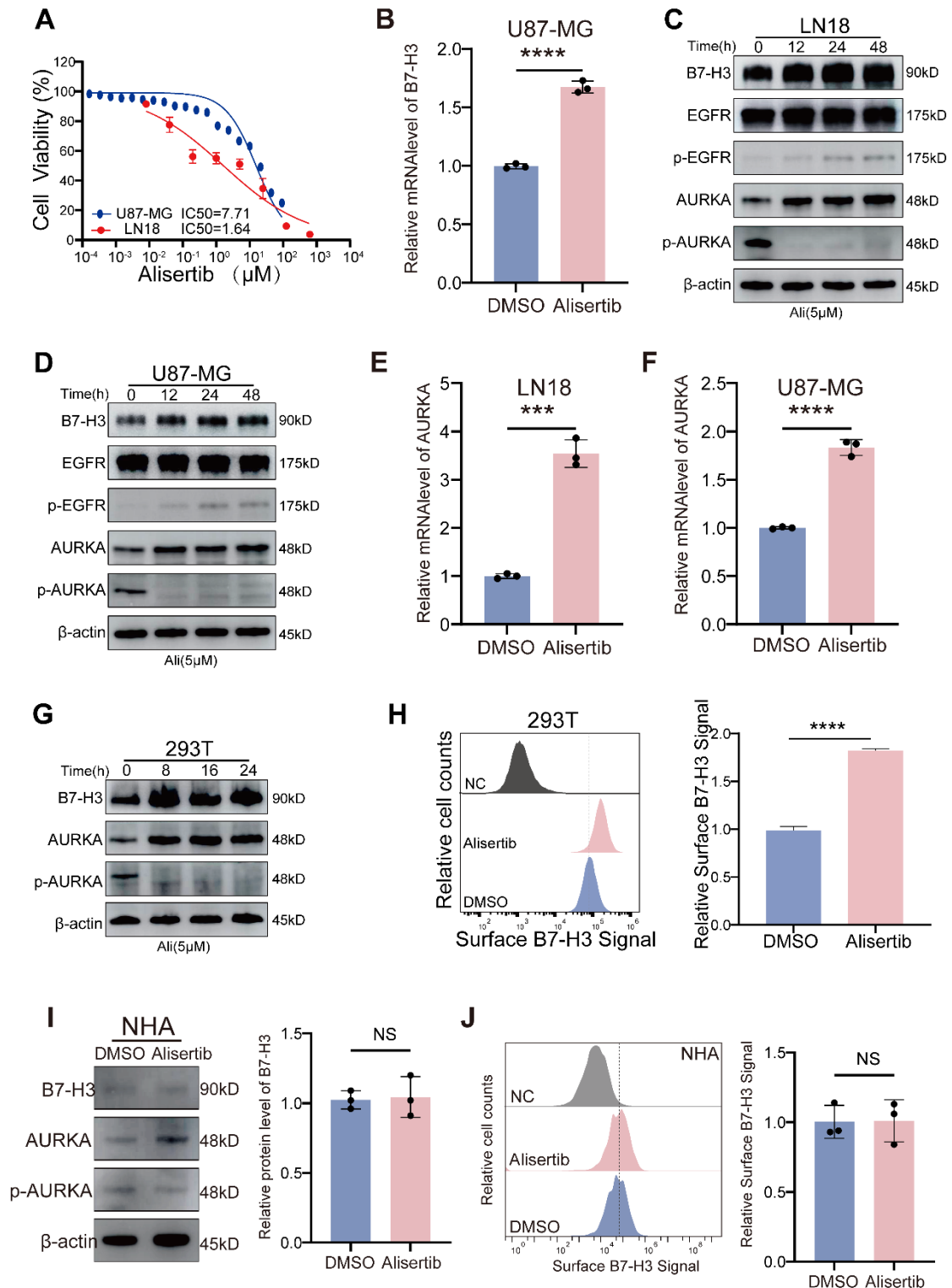
12

13 **Supplementary Figure 2 (A)** The protein expression of AURKA in the  
 14 human glioma cell lines U87-MG, LN18, U373, LN229, TJ905, M059K  
 15 and U251 was detected by Western blotting.  $\beta$ -actin was used as the  
 16 internal control. **(B)** Overexpression of AURKA by AURKA\_cDNA\_Flag  
 17 in LN229 cells. The protein expression of AURKA was detected via  
 18 Western blotting.  $\beta$ -actin was used as the internal control. **(C and D)**  
 19 Silencing of AURKA with specific shRNAs in LN18 and U87-MG cells.  
 20 The protein expression of AURKA was detected via Western blotting.  $\beta$ -  
 21 actin was used as the internal control. **(E)** The mRNA level of B7-H3 in  
 22 LN18 cells expressing shNC or AURKA-shRNA#3. **(F)** The protein  
 23 expression of AURKA and B7-H3 in LHG cells transfected with AURKA-  
 24 siRNA or siNC was detected by Western blotting.  $\beta$ -actin was used as the  
 25 internal control. **(G)** The mRNA level of B7-H3 in LN229 cells expressing  
 26 vector or AURKA\_cDNA\_Flag. Statistical significance was assessed by a  
 27 2-tailed unpaired Student's t test and the data represent the means  $\pm$  SD **(E**

28 **and G).** All samples were biologically independent, and 3 independent  
29 experiments were performed. \*\*\*\* $P < 0.0001$ .

30

31 **Supplementary Figure 3**



32

33 **Supplementary Figure 3 (A)** LN18 (n = 6 independent samples) and U87-

34 MG (n=3 independent samples) cells were treated with increasing

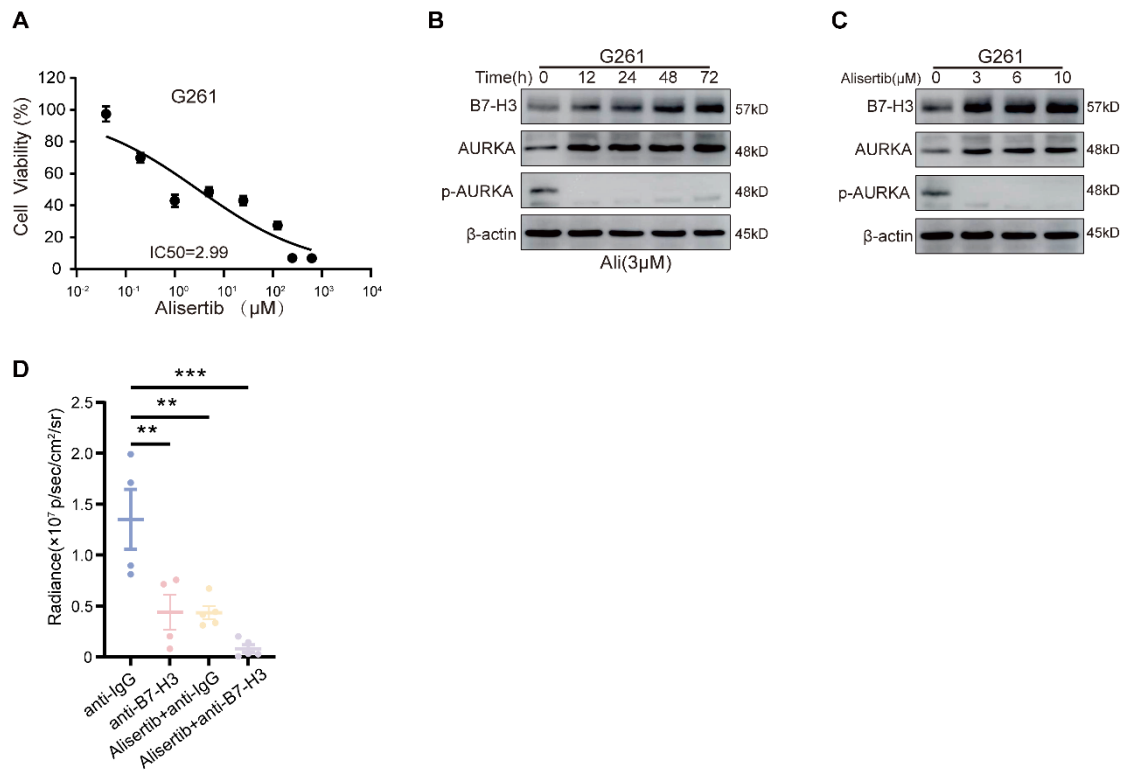
35 concentrations of alisertib for 72 h. The cell viability was analyzed by a

36 CCK-8 assay. **(B)** The mRNA level of B7-H3 in U87-MG cells treated with  
37 alisertib (24 h, 5  $\mu$ M) or DMSO. **(C and D)** The protein expression of B7-  
38 H3, total AURKA, p-AURKA (T288), p-EGFR (Y1068) and total EGFR  
39 in LN18 and U87-MG cells treated with alisertib (5  $\mu$ M) for different  
40 durations (0 h, 12 h, 24 h and 48 h) was detected by Western blotting.  $\beta$ -  
41 actin was used as the internal control. **(E and F)** mRNA level of AURKA  
42 in LN18 and U87-MG cells treated with alisertib (24 h, 5  $\mu$ M) or DMSO.  
43 **(G)** The protein expression of B7-H3, total AURKA and p-AURKA (T288)  
44 in 293T cells treated with alisertib (5  $\mu$ M) for different durations (0 h, 8 h,  
45 16 h and 24 h) was detected via Western blotting.  $\beta$ -actin was used as the  
46 internal control. **(H)** B7-H3 levels on the cell surface of 293T cells treated  
47 with alisertib (24 h, 5  $\mu$ M) or DMSO were quantified via flow cytometry.  
48 Cells that were only stained with isotype control antibodies were used as  
49 the negative control (NC). **(I)** The protein expression of B7-H3, total  
50 AURKA and p-AURKA (T288) in NHA cells treated with alisertib (24 h,  
51 5  $\mu$ M) or DMSO was detected by Western blotting.  $\beta$ -actin was used as the  
52 internal control. **(J)** B7-H3 levels on the cell surface of NHA cells treated  
53 with alisertib (24 h, 5  $\mu$ M) or DMSO were quantified via flow cytometry.  
54 Cells that were only stained with isotype control antibodies were used as  
55 the negative control (NC). Statistical significance was assessed by using a  
56 2-tailed unpaired Student's t test and the data represent the means  $\pm$  SD (B,  
57 E, F, H, I and J). All the samples were biologically independent. Three

58 independent experiments were performed **(B-J)**. \*\*\* $P < 0.001$ , \*\*\*\* $P$   
59  $< 0.0001$ . NS, not significant.

60

61 **Supplementary Figure 4**



62

63 **Supplementary Figure 4 (A)** G261 cells were treated with increasing  
 64 concentrations of alisertib for 72 h, and cell viability was analyzed via a  
 65 CCK-8 assay. (n = 6 independent samples). **(B)** The protein expression of  
 66 B7-H3, total AURKA, and p-AURKA (T288) in G261 cells treated with  
 67 alisertib (3  $\mu\text{M}$ ) for different periods (0 h, 12 h, 24 h, 48 h or 72 h) was  
 68 detected by Western blotting (n = 3/group).  $\beta$ -actin was used as the internal  
 69 control. **(C)** The protein expression of B7-H3, total AURKA, and p-  
 70 AURKA (T288) in G261 cells treated with increasing concentrations of  
 71 alisertib for 24 h was detected by Western blotting (n = 3/group).  $\beta$ -actin  
 72 was used as the internal control. **(D)** On day 26, the tumor volume in the  
 73 alisertib + anti-IgG (n = 5), anti-B7-H3 mAb (n = 4), and combination  
 74 treatment (n = 5) groups was significantly lower than that in the anti-IgG

75 group (n = 4). The tumors in the alisertib + anti-B7-H3 mAb group were  
76 smaller than those in the alisertib + anti-IgG or anti-B7-H3 mAb group, but  
77 the difference was not statistically significant. Statistical significance was  
78 assessed via 1-way ANOVA followed by Tukey's multiple comparisons test  
79 and the data represent the means  $\pm$  SEM (D). \*\* $P < 0.01$ , \*\*\* $P < 0.001$ .