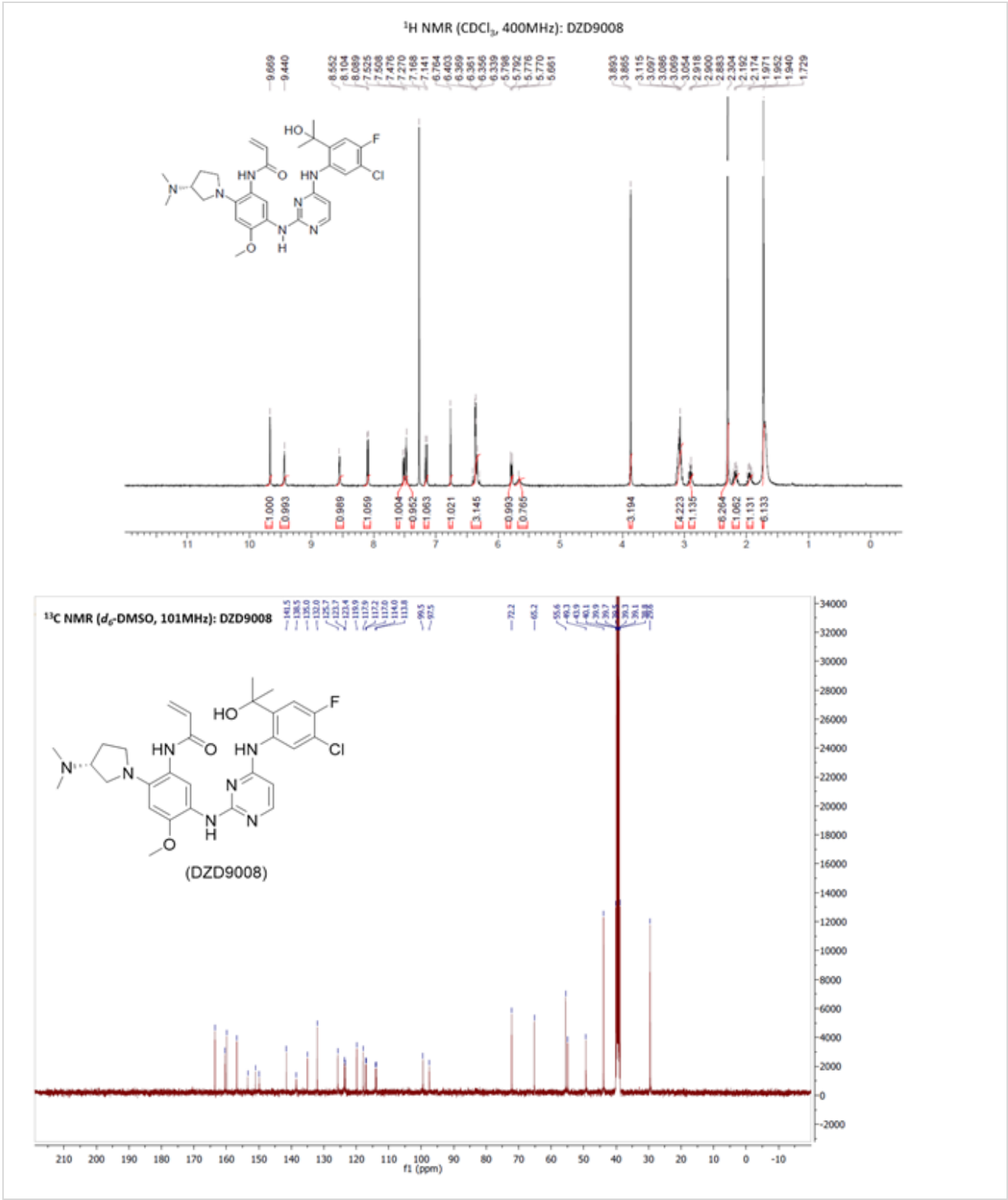
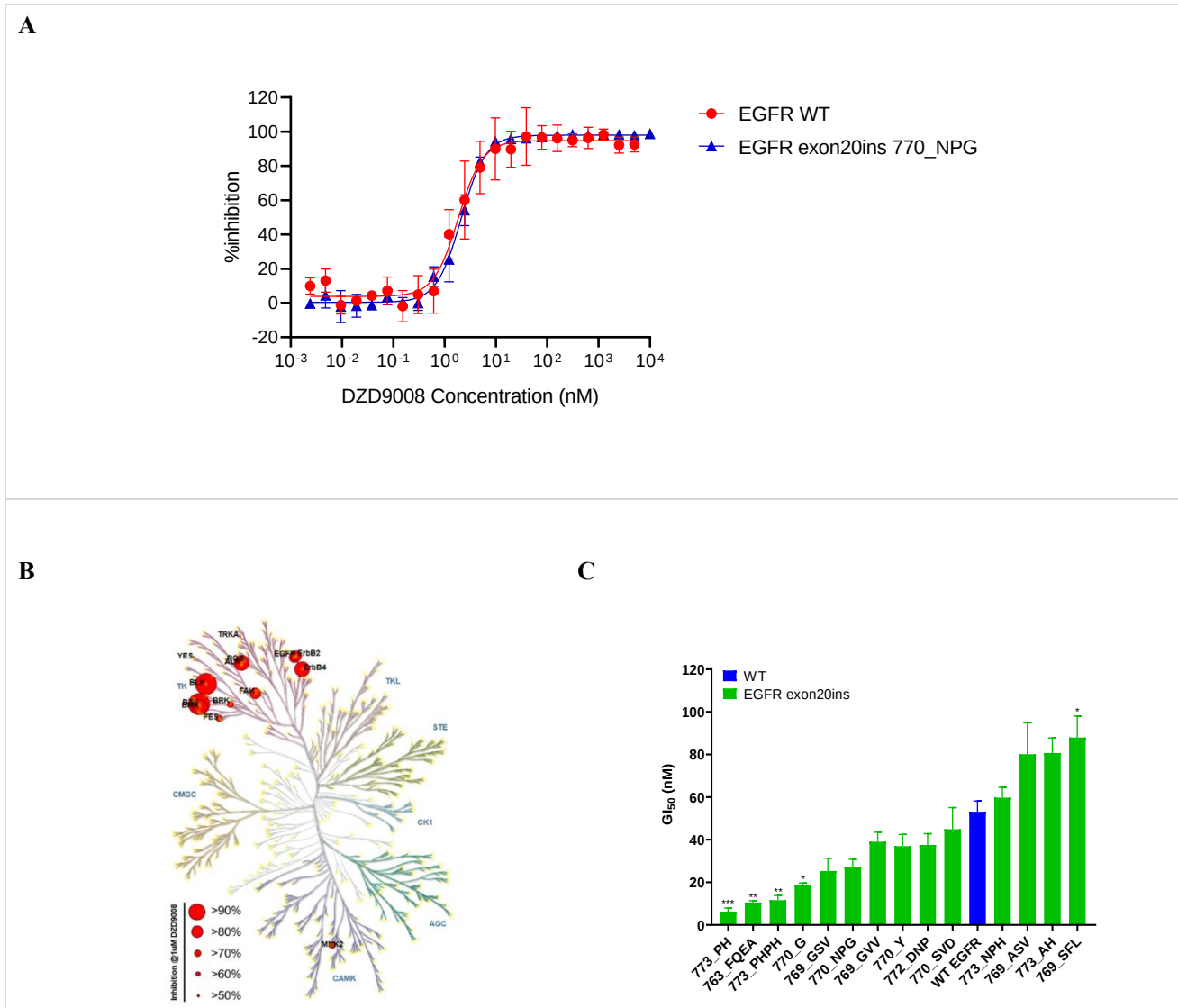
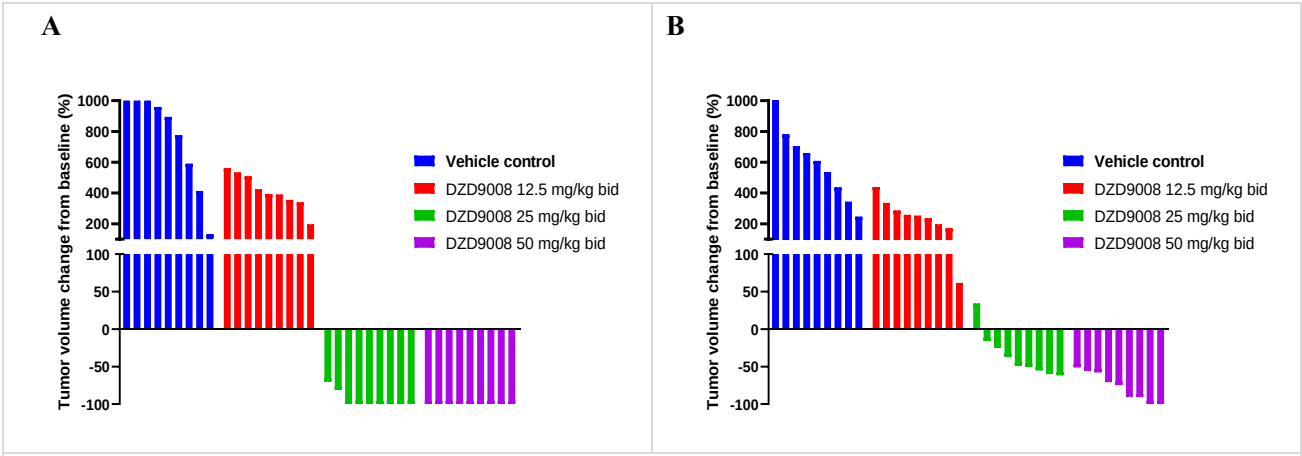
Supplementary Materials



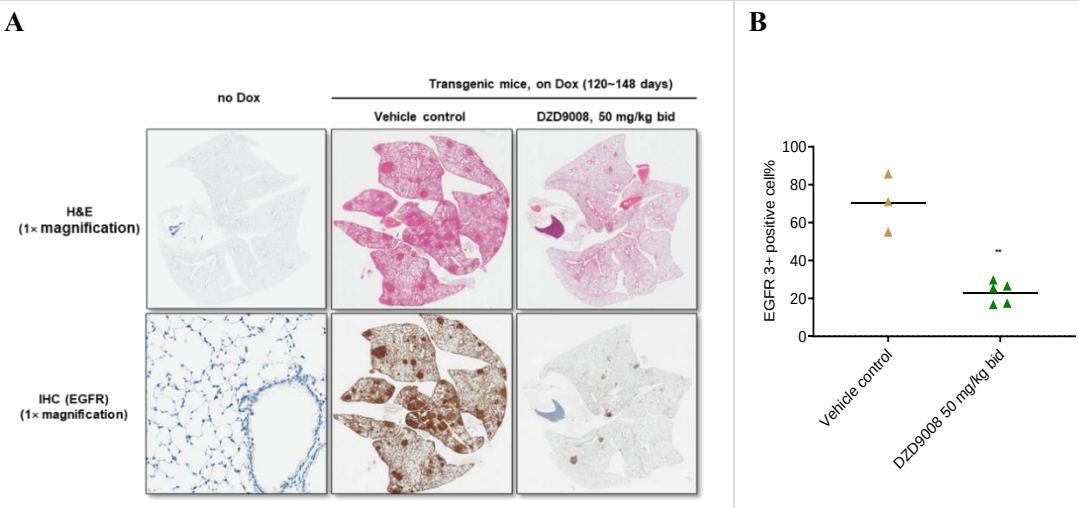
Supplementary Figure S1. ^{1}H and ^{13}C nuclear magnetic resonance spectra of sunvozertinib DZD9008 = sunvozertinib.



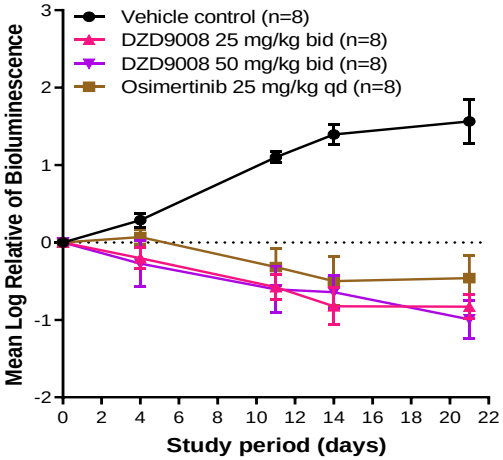
Supplementary Figure S2. Enzymatic activity and anti-proliferation activity of sunvozertinib. A. Enzymatic activity of sunvozertinib on EGFR Exon20ins 770_NPG and wild type. The compound was pre-incubated with recombinant kinases at room temperature for 30 minutes, then the reaction was initiated by adding 2 mM ATP and substrate peptide which could be phosphorylated by kinases in the reaction. After 30 minutes incubation, the reaction was stopped by adding detection reagent mix containing EDTA. **B.** The kinases with > 50% inhibition by 1 uM sunvozertinib at Km ATP were plotted on the human kinome tree (www.kinhub.org). Circle size is proportional to percentage inhibition. **C.** Anti-proliferation activity GI_{50} of sunvozertinib in Ba/F3 cell lines carrying different EGFR exon20ins or A431 carrying wild-type EGFR. Cells were treated with sunvozertinib at a series of concentrations for 72 hours, and then cell viability was analyzed using the CellTiter-Glo viability assay. Three independent experiments were performed. Data were presented as mean±standard error of the mean. One-way ANOVA test was used for comparison with wild-type EGFR. *P<0.05, **P<0.01, ***P<0.001. DZD9008 = sunvozertinib.



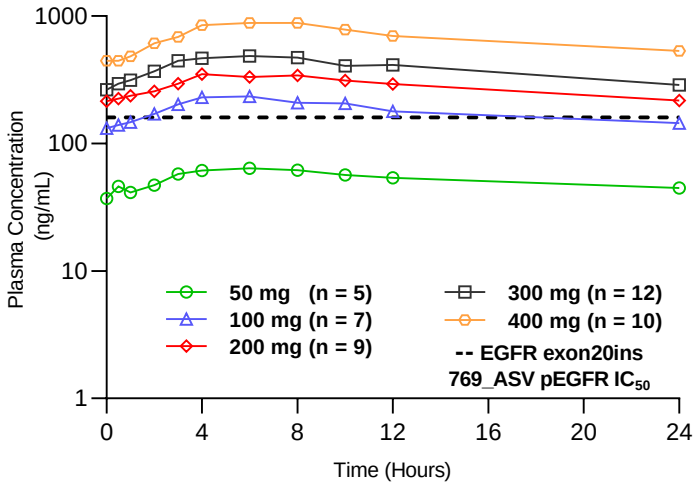
Supplementary Figure S3. Waterfall plots of i*n vivo* antitumor activity of sunvozertinib in EGFR exon20ins xenograft models. A. Antitumor activity of sunvozertinib in PDX model LU0387 carrying EGFR exon20ins 773_NPH. **B.** Antitumor activity of sunvozertinib in A431 xenograft model expressing wild-type EGFR. DZD9008 = sunvozertinib. Bid: twice daily. The tumor volume change shown here were the volume change at the end of treatment in Figure 2.



Supplementary Figure S4. Sunvozertinib induced profound tumor growth inhibition in transgenic mice tumor models expressing EGFR exon20ins 769_ASV. A. Representative H&E (hematoxylin and eosin) and EGFR IHC (immunohistochemistry) staining of lung samples collected from EGFR exon20ins 769_ASV mice after induction by doxycycline at 2 mg/ml for the 120 to 148 days. Then mice were treated with sunvozertinib at 50 mg/kg or vehicle for 30 days (n = 3 to 5 mice per group). **B.** Dot plots of tumor burden before and after 30 days of treatment in EGFR exon20ins 769_ASV transgenic tumor model. The EGFR positivity was used to reflect the tumor burden. Each dot represented one mouse. P value was determined by student t-test using GraphPad Prism. **P < 0.01. DZD9008 = sunvozertinib.



Supplementary Figure S5. Antitumor activity of sunvozertinib in brain metastasis (BM) xenograft model. The BM model was established by intracranial injection of luci-H1975 cell clone expressing EGFR T790M mutation. BM: brain metastasis. bid: twice daily. qd: once daily. DZD9008 = sunvozertinib.



Supplementary Figure S6. Sunvozertinib geometric mean plasma concentration over time after multiple-dose administration at cycle 2 day 1. Pooled analysis of WU-KONG1 and WU-KONG2 studies. Data cut-off date (WU-KONG1: 15 October 2020; WU-KONG2: 9 September 2020). Blank dash line represents the back-converted plasma concentration of sunvozertinib of which free fraction equals to pEGFR IC_{50} of one of the most prevalent EGFR exon20ins subtypes 769_ASV.