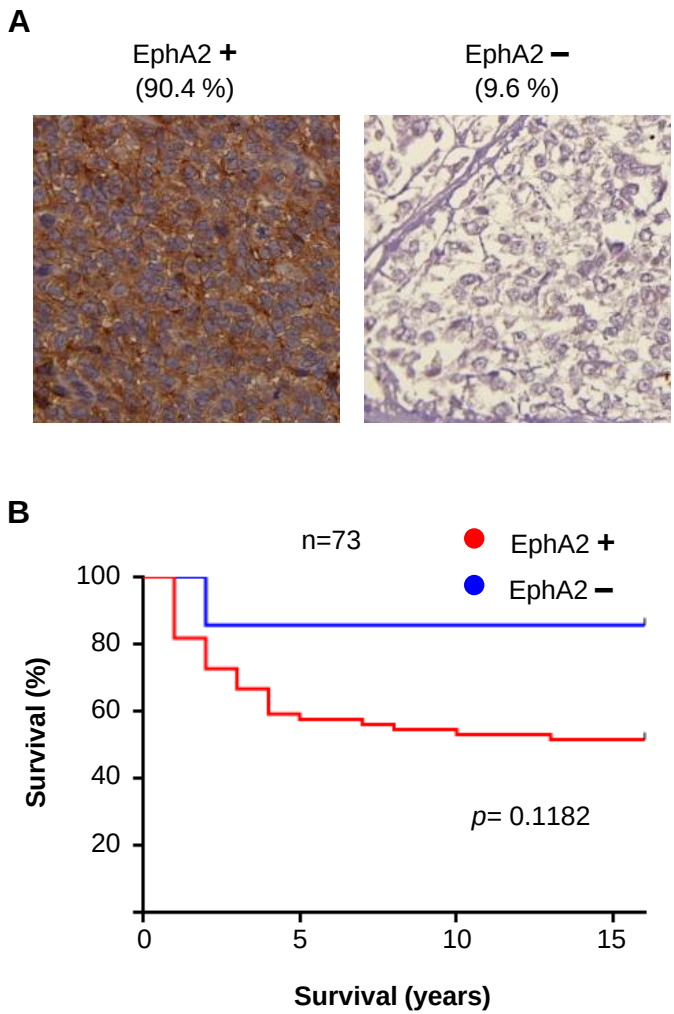


Figure S1: EphA2 phosphorylation, but not its mere presence, is key to aggressiveness in Ewing sarcoma cell lines. **(A)** Sample micrographs from the Ewing sarcoma tissue microarray showing differential expression patterns of EphA2. Magnification: 40× **(B)** Kaplan-Meier curve comparing differential survival of Ewing sarcoma patients in function of EphA2 expression.

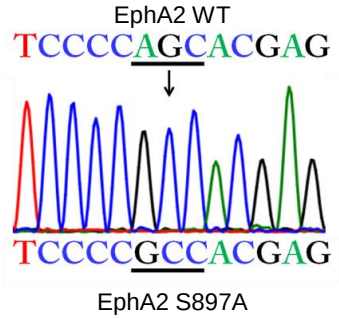


Figure S2: Diagram representing the EphA2 site-directed mutagenesis in S897 residue.

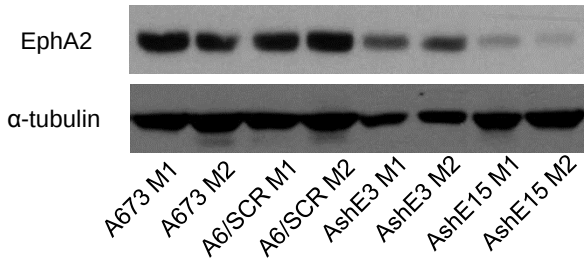
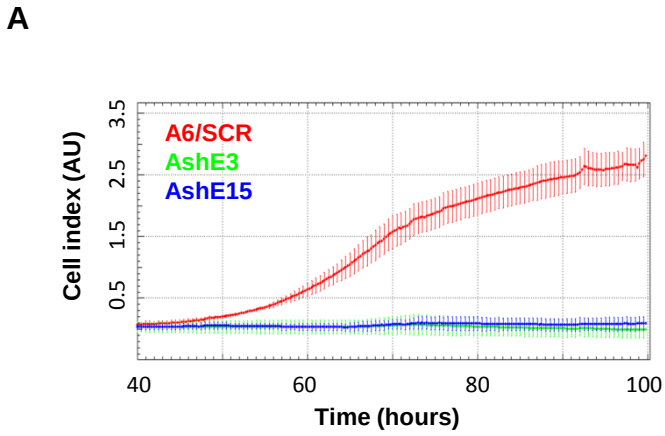
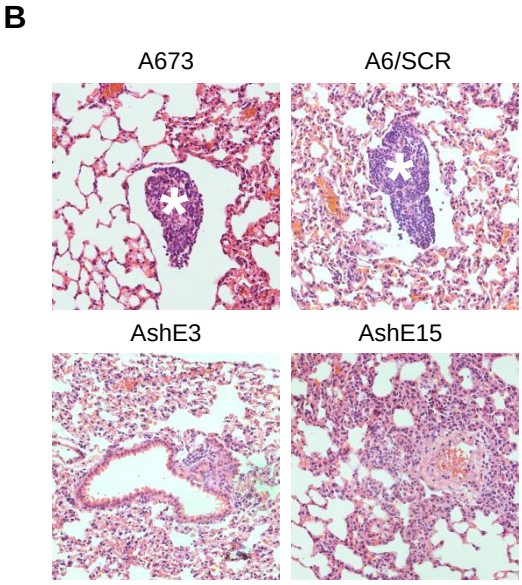


Figure S3: Representative western blot showing EphA2 levels from samples obtained from subcutaneous tumors induced by the A673 EphA2 silencing model. M# refers to the sample mice.





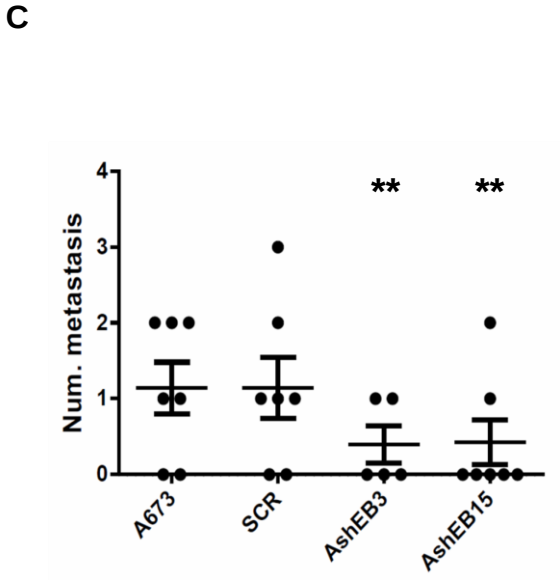


Figure S4: EphA2 silencing reduces long-term invasive capacity in A673 cells. **(A)** Real-Time invasion assay in Matrigel using xCELLigence system. **(B)** Representative images of mice lungs after orthotopic metastasis assay. Magnification 20x. **(C)** Quantification of total number of metastasis per mice after orthotopic metastasis assay. Data are represented as means ±SD. Statistical significance was achieved by the Student's *t* test from at least three different experiments: *$p \leq 0.05$ **$p \leq 0.01$.

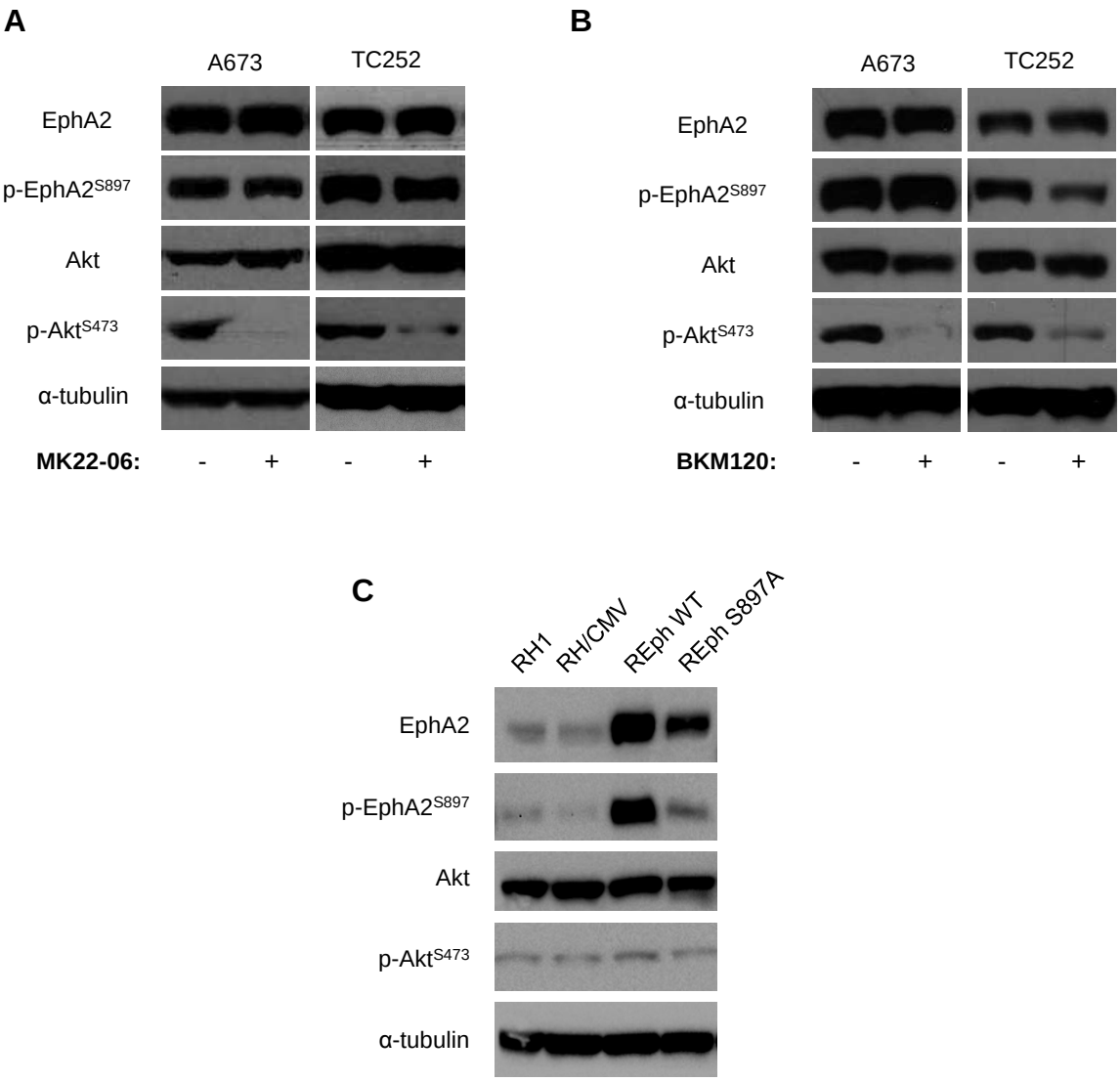


Figure S5: There is no reciprocal regulatory phosphorylation feedback loop between Akt and EphA2, independent of ligand presence. **(A-B)** Representative western blots showing EphA2 and p-EphA2^{S897} levels after MK-2206 **(A)**, and BKM120 **(B)** treatments. Phosphorylation of Akt is shown as control of treatment efficiency. **(C)** Representative western blot showing Akt phosphorylation in RH1 EphA2 reintroduction model.

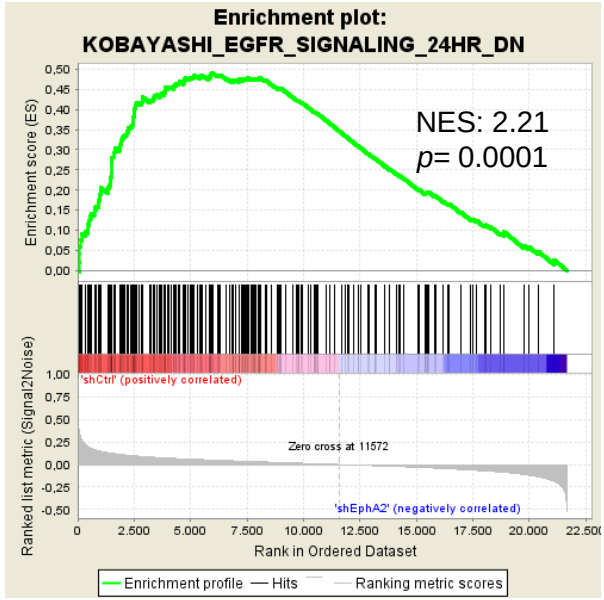


Figure S6: GSEA showing a correlation between genes differentially expressed after EphA2 silencing and after EGFR inhibition. NES = Normalized Enrichment Score.

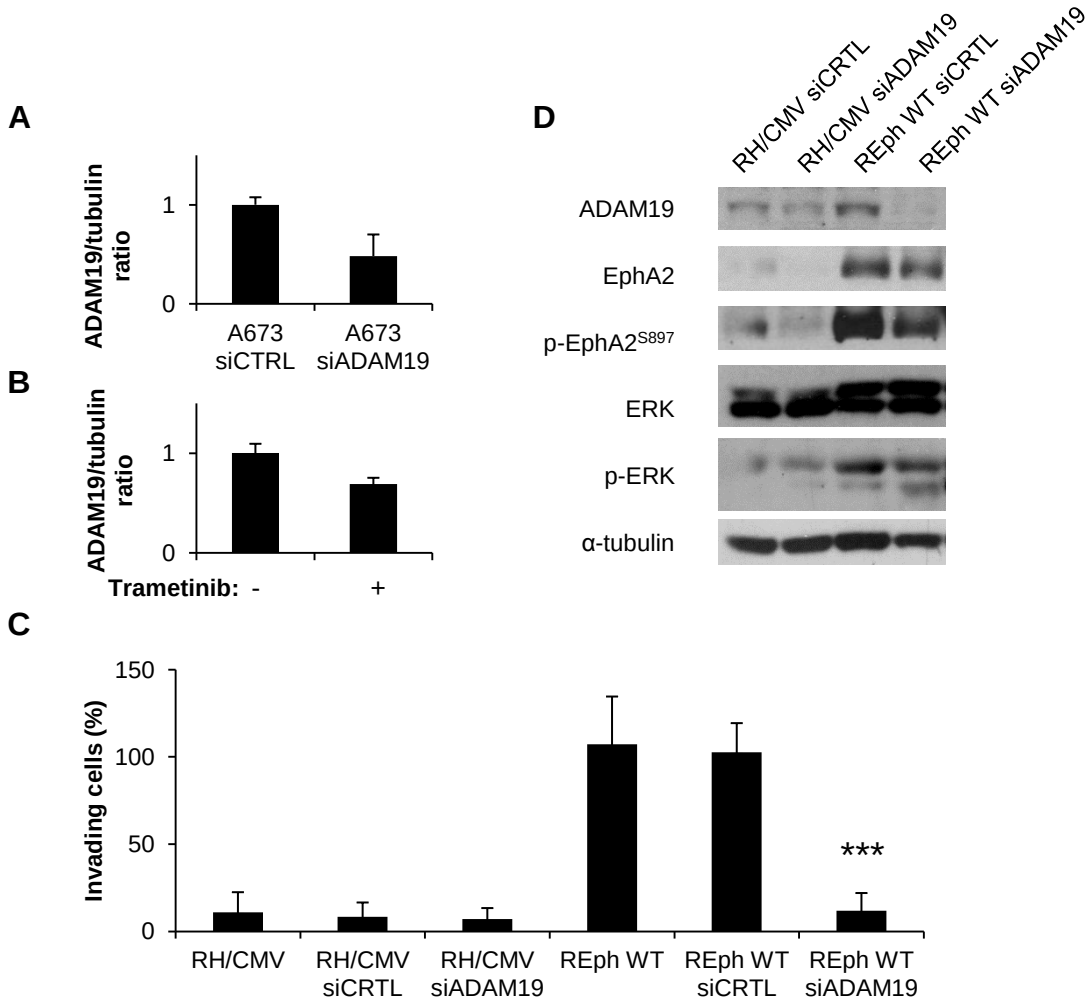


Figure S7: Metalloproteinase disintegrin ADAM19 is key for EphA2-linked aggressive behavior. **(A)** Quantification of ADAM19 expression in A673 after silencing, data from Figure 7D. **(B)** Quantification of ADAM19 expression levels in A673 treated with the MEK inhibitor Trametinib, data from Figure 7E. **(C)** Invasion assay in Matrigel-coated Boyden chambers after ADAM19 silencing in RH1 EphA2 reintroduction model. **(D)** Representative western blot showing ADAM19 expression and phosphorylation of EphA2 and ERK after siRNA silencing in RH1 EphA2 reintroduction model. siCTRL = non-targeting siRNA. Data are represented as means ±SD. Statistical significance was achieved by the Student's *t* test from at least three different experiments: ***$p \leq 0.001$.