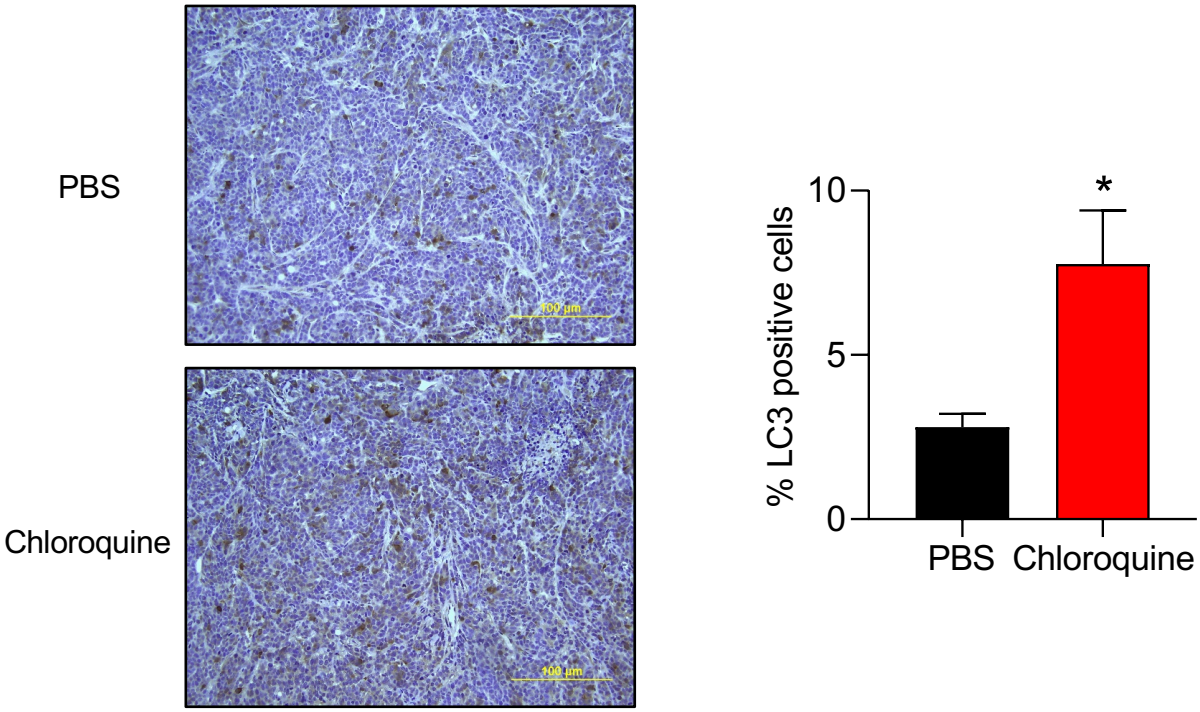
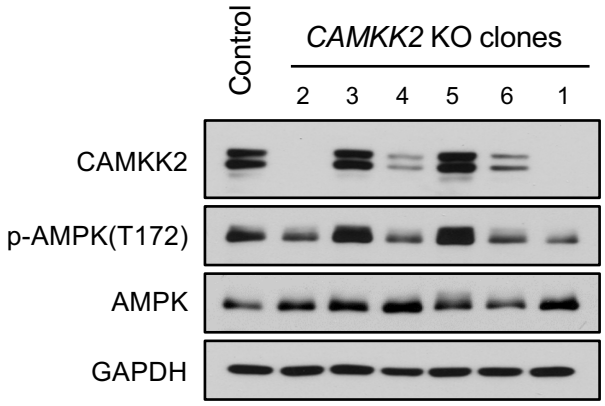
Figure S1



Supplementary Figure S1. LC3 IHC staining (*left*) and quantification (*right*) of 22Rv1-fLuc xenograft mouse tumors from vehicle (PBS) or chloroquine treatment groups. *P* values were calculated using a two-tailed t test. **P* < 0.05.

Figure S2

A

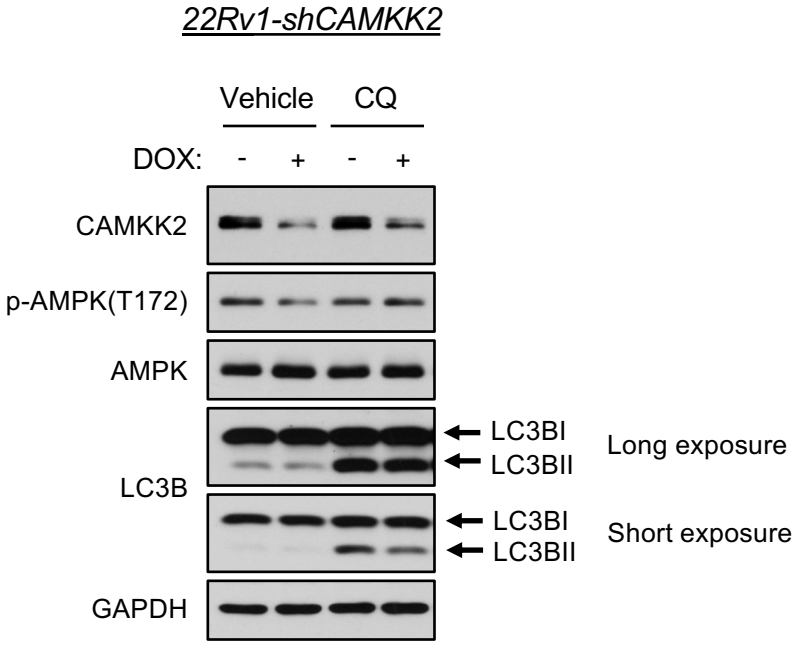


B

C4-2 Cas9 Control (WT *CAMKK2*)
5'-CCGTGACATCAAACCTTCCA-3'
KO clone-1 (Heterozygous)
5'-CCG—ACATCAAACCTTCCA-3' (deletion)
5'-CCGGTGACATCAAACCTTCCA-3' (insertion)
KO clone-2 (Homozygous)
5'-CCGTTGACATCAAACCTTCCA -3' (insertion)

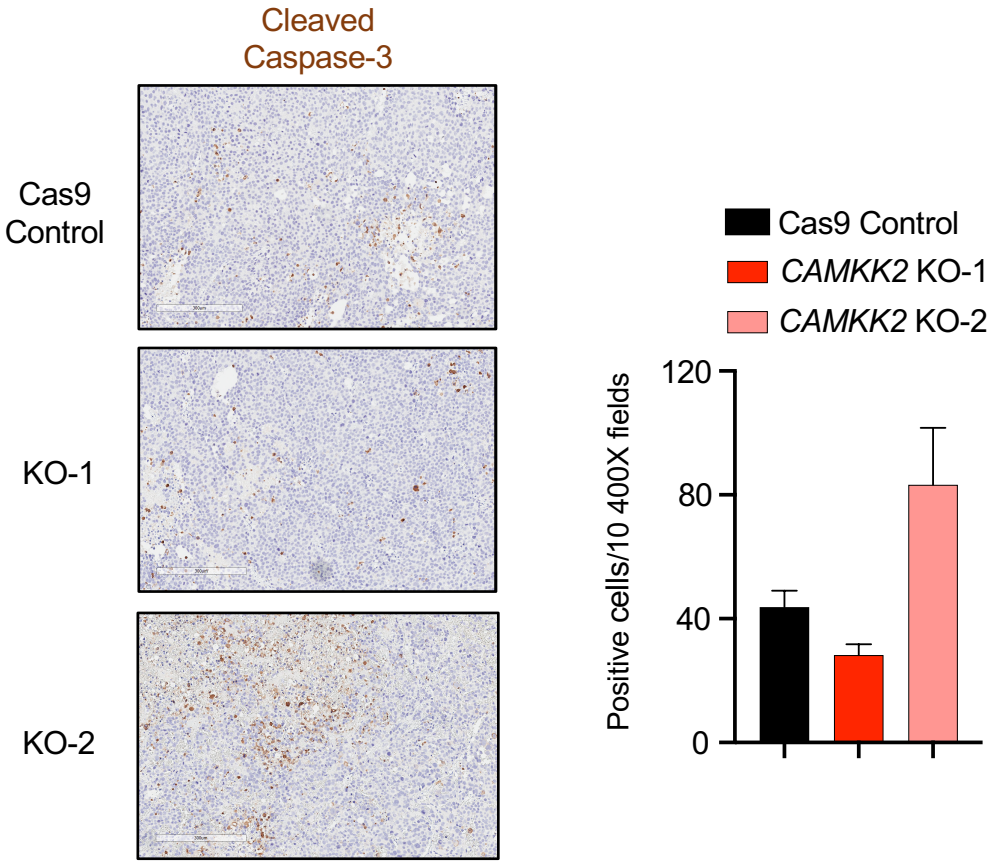
Supplementary Figure S2. (A) sgRNAs targeting *CAMKK2* were expressed in C4-2 inducible Cas9 cells and single cell clones were selected after DOX (200 ng/ml) treatment. Immunoblot analysis of these clones and their parental C4-2 Cas9 cells. Note, the clone numbers here were matched for clarity with the numbering throughout the main text and figures. (B) Sanger sequence of C4-2 Cas9 cells and its derivatives *CAMKK2* KO clone 1 and clone 2. Red dashes and letters indicate the identified mutations.

Figure S3



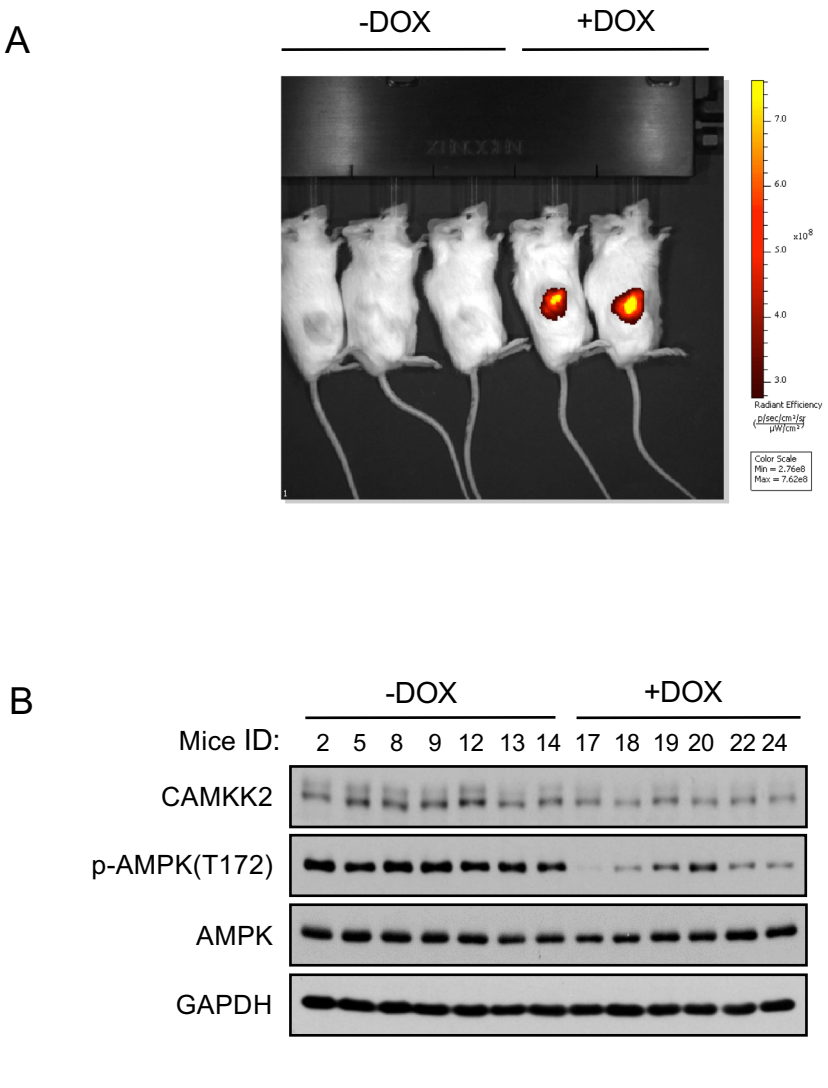
Supplementary Figure S3. Immunoblot analysis of 22Rv1-sh*CAMKK2* cells ± 800 ng/ml DOX ± 20 μM chloroquine treatment for 72 hours.

Figure S4



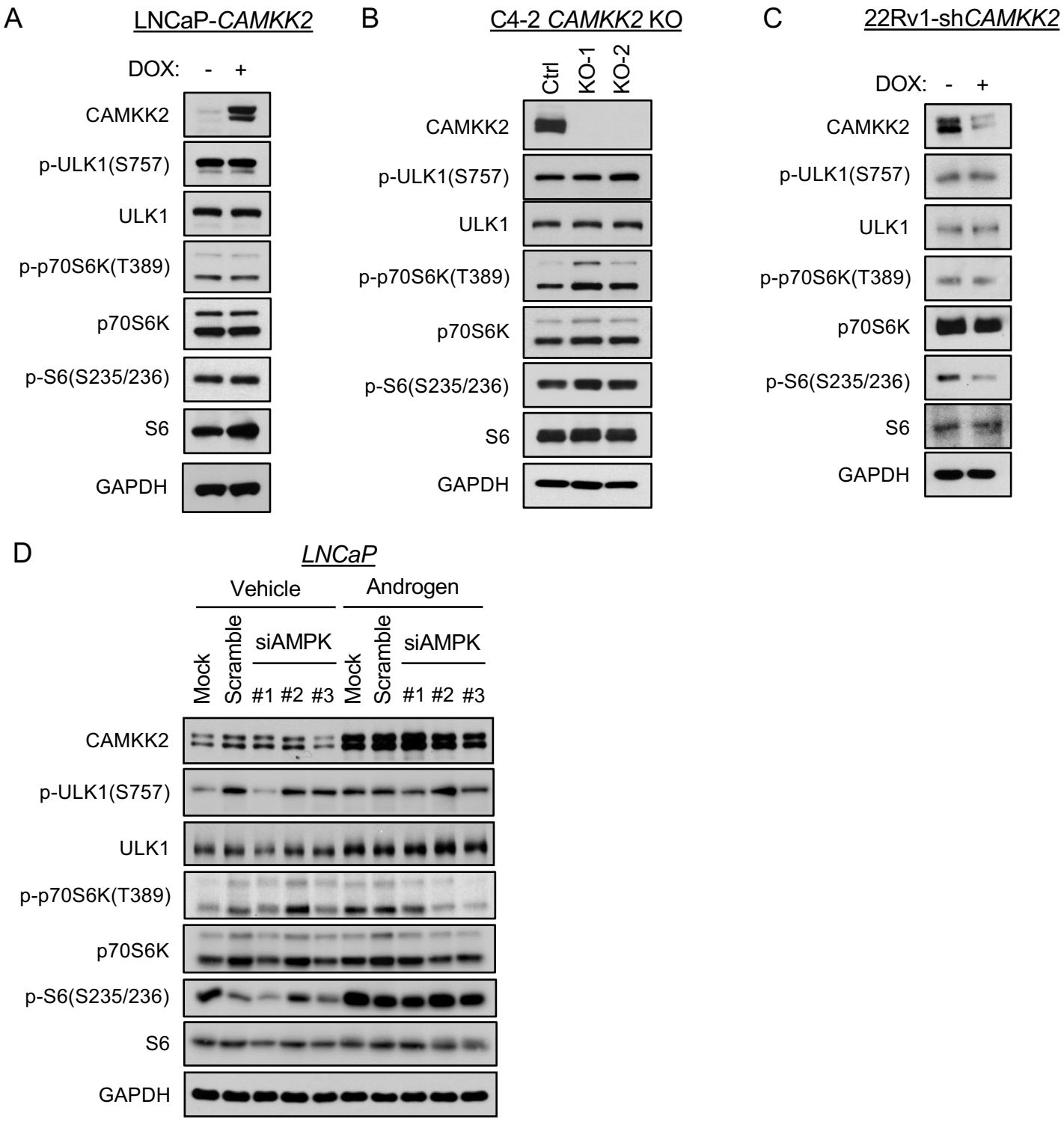
Supplementary Figure S4. Cleaved caspase-3 IHC staining and quantification of C4-2 Cas9 control and *CAMKK2* knockout (KO) xenograft tumors.

Figure S5



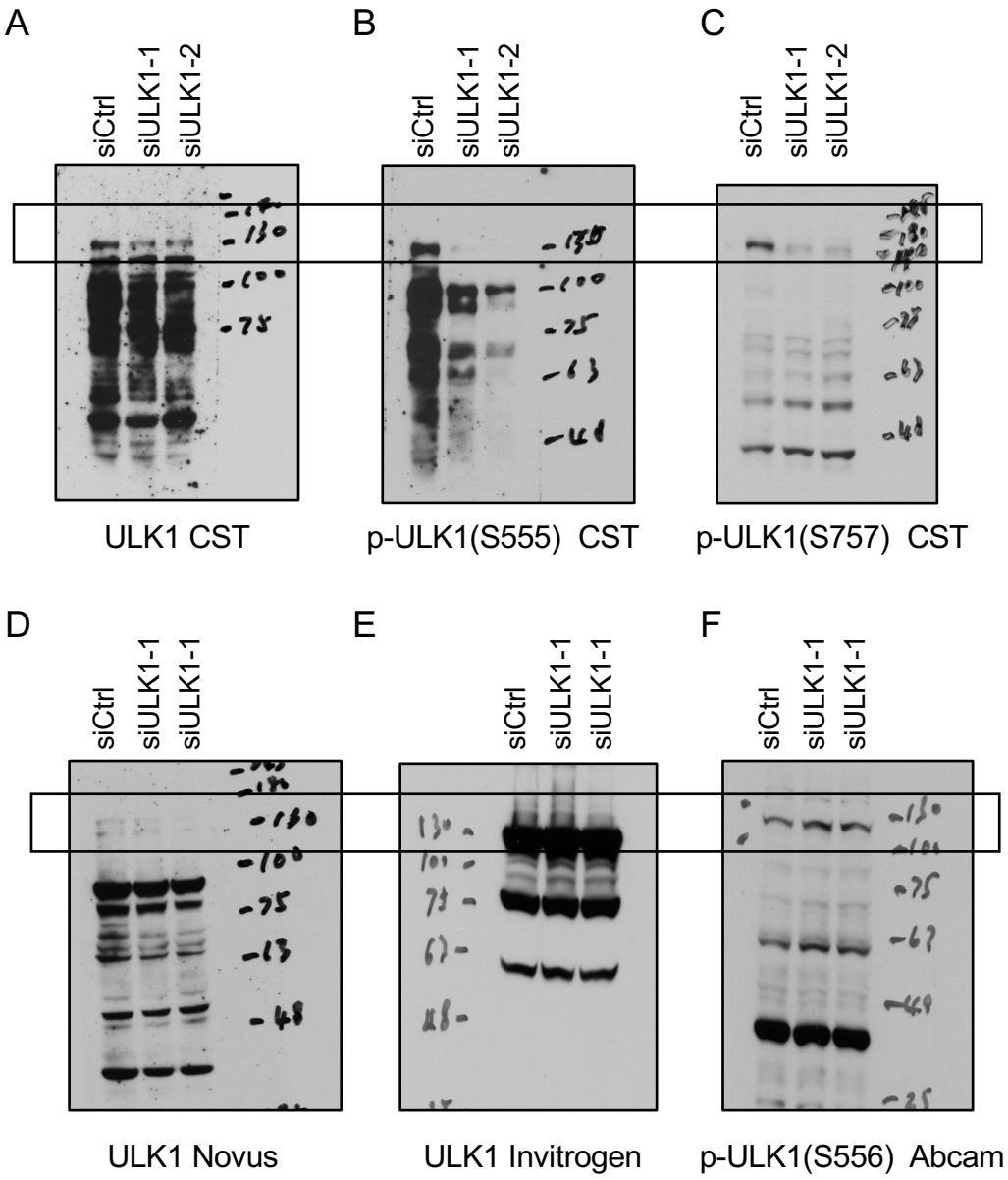
Supplementary Figure S5. (A) Fluorescence imaging of 5 sample mice (3 on normal chow, 2 on DOX-containing chow) confirming the *CAMKK2* shRNA expression. (B) Immunoblot analysis of tumors from 22Rv1-sh*CAMKK2* xenograft mice ± DOX.

Figure S6



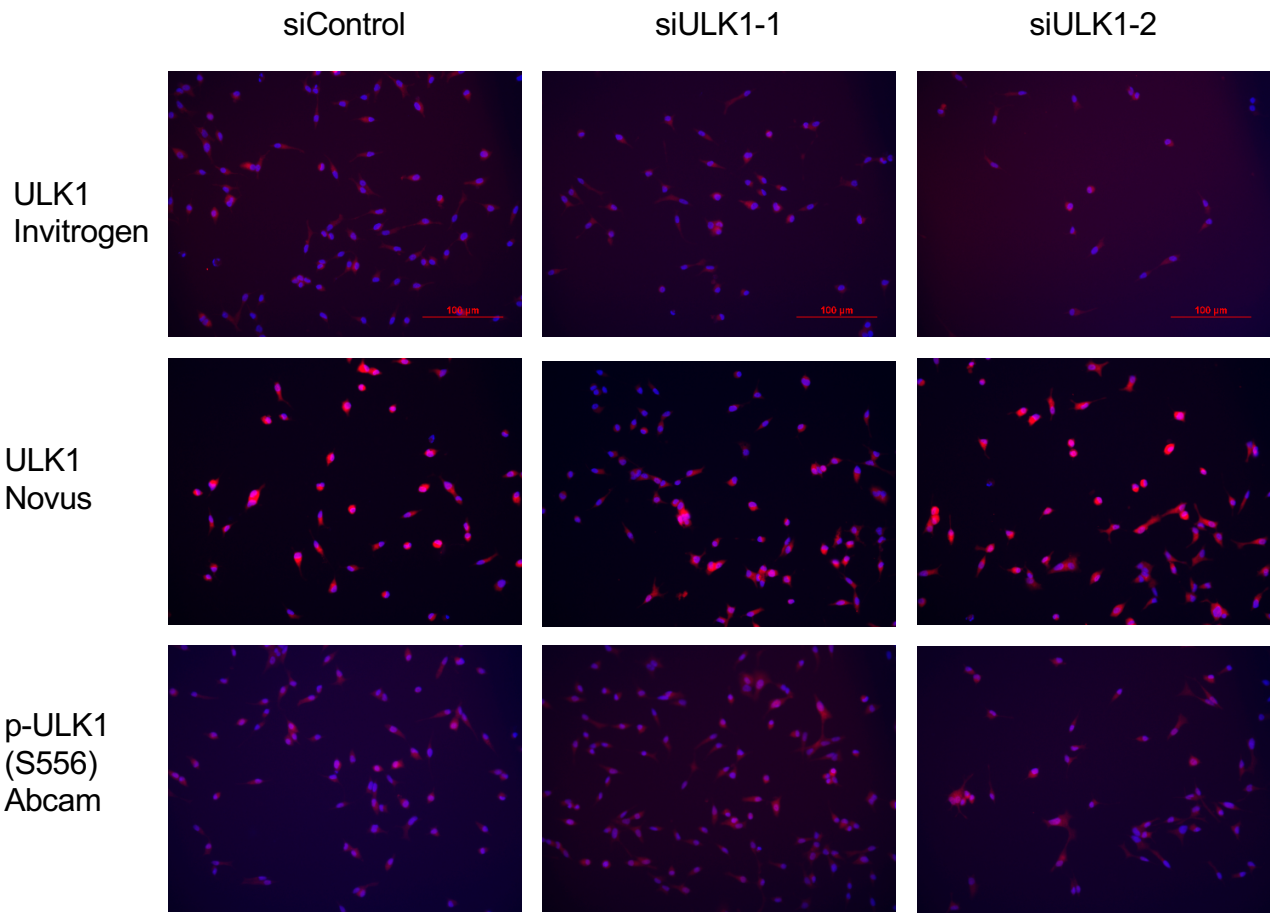
Supplementary Figure S6. (A) LNCaP-*CAMKK2* cells treated ± DOX (50 ng/ml) for 48 hours. Cell lysates were subjected to immunoblot analysis. (B) Immunoblot analysis of C4-2 Cas9 control and *CAMKK2* knockout (KO) cells. (C) 22Rv1-sh*CAMKK2* cells treated ± DOX (800 ng/ml) for 48 hours. Cell lysates were subjected to immunoblot analysis. (D) Immunoblot analysis of parental LNCaP cells treated with siRNAs targeting either scramble or AMPK ± R1881 for 72 hours.

Figure S7



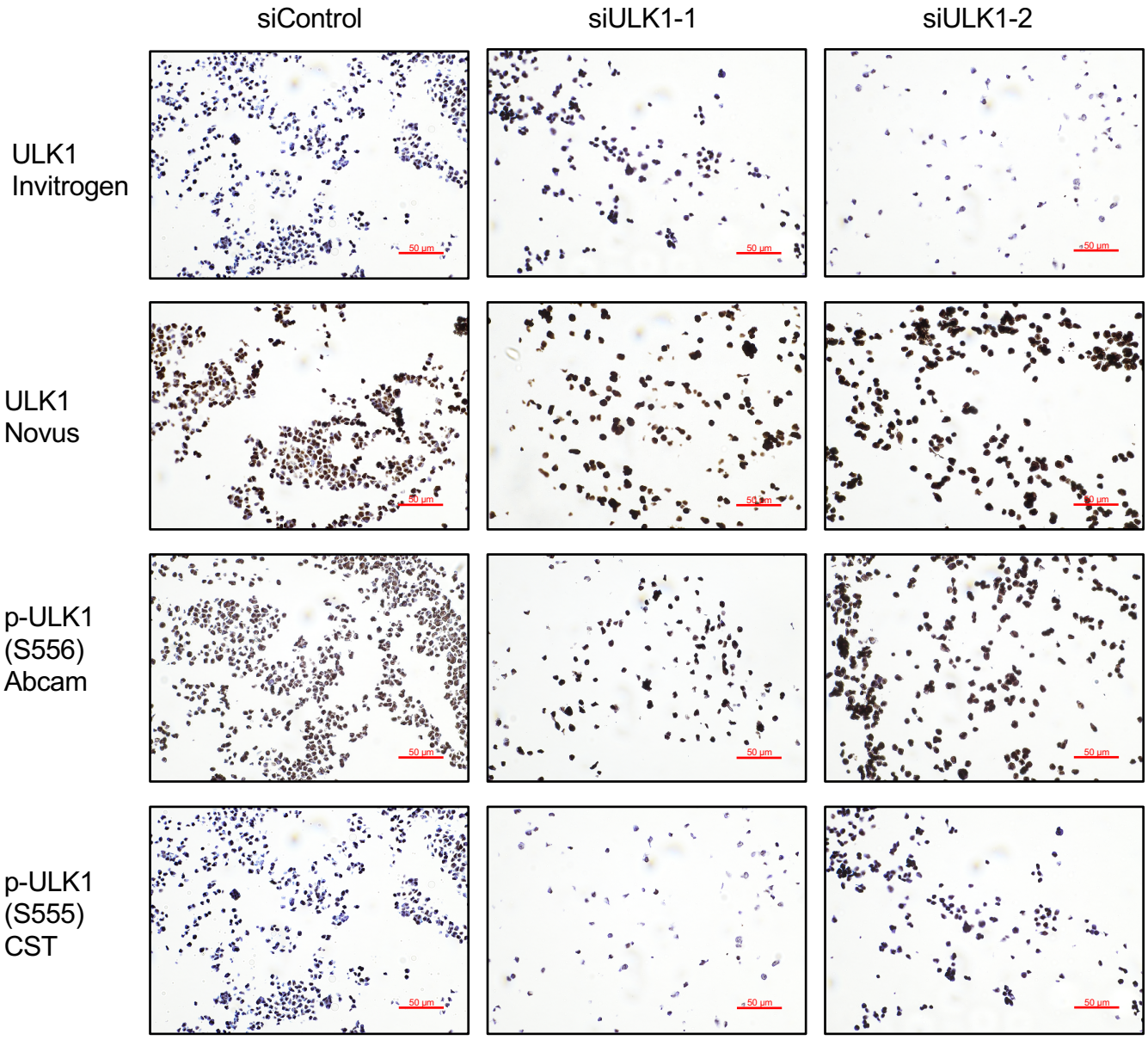
Supplementary Figure S7. LNCaP cells were transfected with siRNAs targeting scramble control or ULK1 for 72 hours and then probed with different ULK1 antibodies. (A) ULK1 (R600, #4773, Cell Signaling Technology), (B) p-ULK1(S555) (D1H4, #5869, Cell Signaling Technology), (C) p-ULK1(S757) (#6888, Cell Signaling Technology), (D) ULK1 (#JA58-36, Novus Biologicals), (E) ULK1 (#MA532699, Invitrogen), (F) p-ULK1(S556) (#ab203207, Abcam).

Figure S8



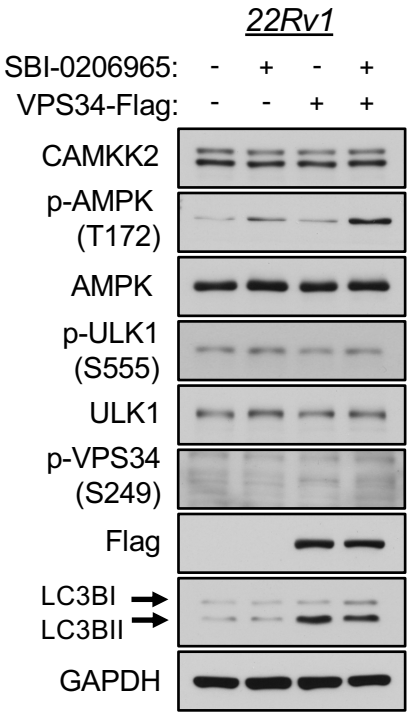
Supplementary Figure S8. LNCaP cells were transfected with siRNAs targeting scramble control or ULK1 for 72 hours and then stained with different ULK1 antibodies (red) and DAPI counterstain (blue) for ICC/IF.

Figure S9



Supplementary Figure S9. LNCaP cells were transfected with siRNAs targeting scramble control or ULK1 for 72 hours and cell plugs were embedded in paraffin and IHC stained with indicated primary antibodies.

Figure S10



Supplementary Figure S10. 22Rv1 cells were transfected ± VPS34-Flag for 48 hours. Immunoblot analysis of transfected cells with 2 hours SBI-0206965 (10 μM) treatment.

Supplemental Table 1. siRNA, shRNA and sgRNA sequences.

	Sequence
siRNA	
AMPK (*PRKAA1*) #1	5'-CACAGAAGGAUUUAAAUAUUGAGGG-3'
AMPK (*PRKAA1*) #2	5'-ACCAUGAUUGAUGAUGAAGCCUUAA-3'
AMPK (*PRKAA1*) #3	5'-UUAAGGCUUCAUCAUCAAUCAUGGU-3'
ULK1 #1	5'-GCAUCGGCACCAUCGUCUtt-3'
ULK1 #2	5'-GCAUGGACUUCGAUGAGUUtt-3'
shRNA	
CAMKK2	5'-GGCATCGAGTACTTACACT-3'
sgRNA	
CAMKK2	5'-TGGAAGGTTTGATGTCACGG-3'