Supplemental Data

Figure S1. RT-PCR analysis of c-Myc, cyclinD1, MMP-7, Axin2, E-cadherin, vimentin, N-cadherin and slug in CNE1 and HNE1 cells.

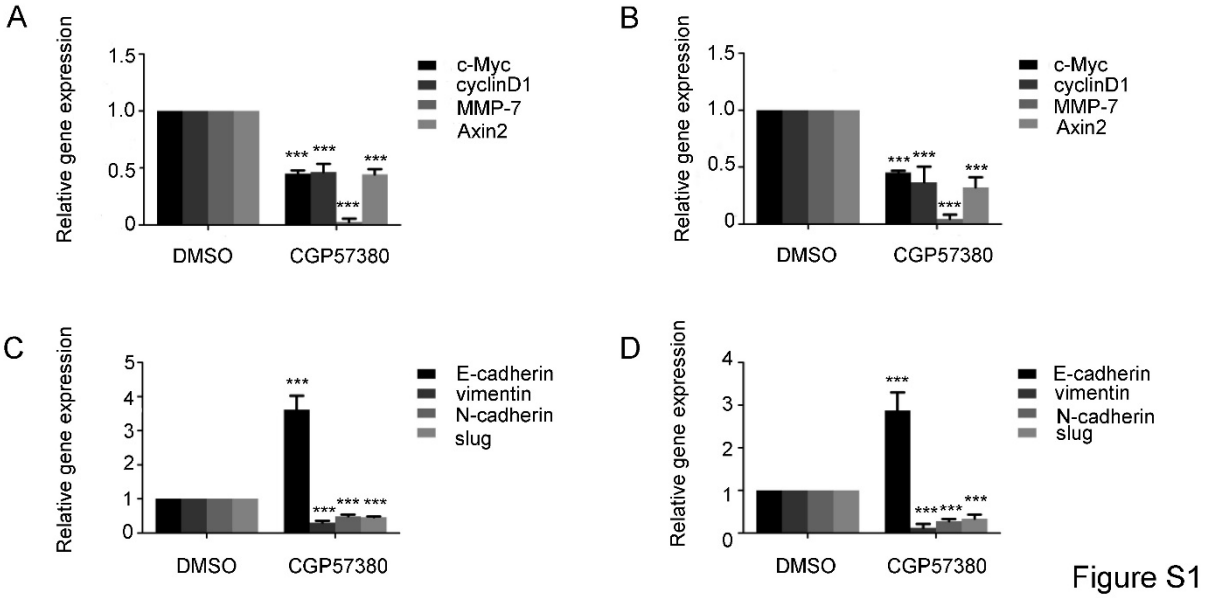


Figure S1

Figure S2. Detection and analysis of apoptosis by flow cytometry in CNE1 and HNE1 cells treated by DMSO or CGP57380.

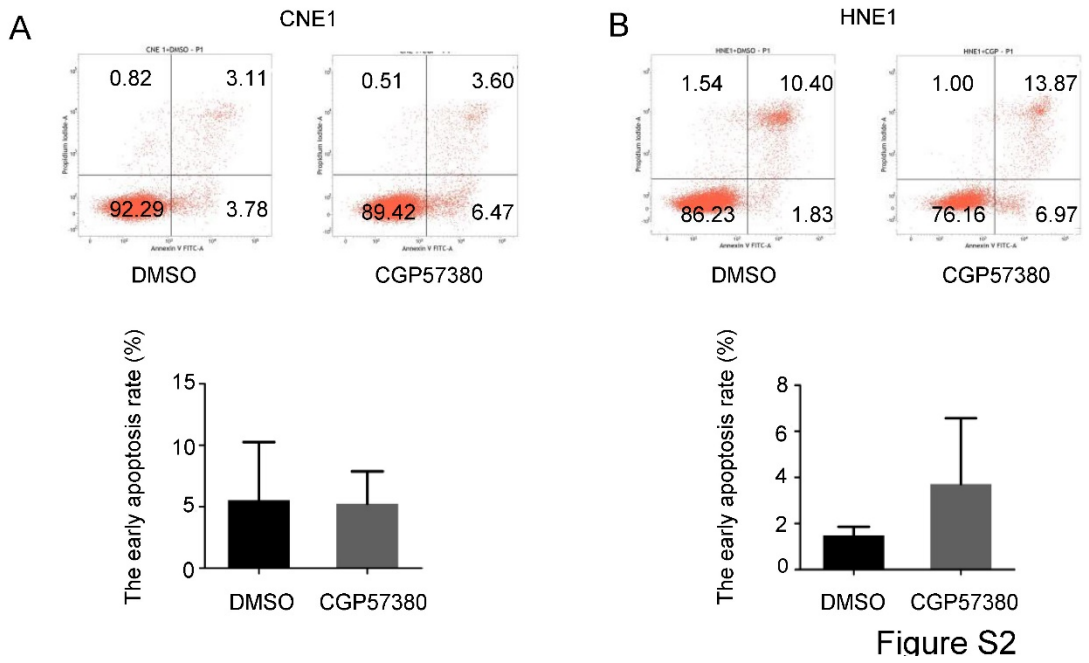


Figure S2

Figure S3. IHC detection of p-eIF4E, cyclinD1, c-Myc and MMP-7 in tumor tissues from CNE1 xenografts.

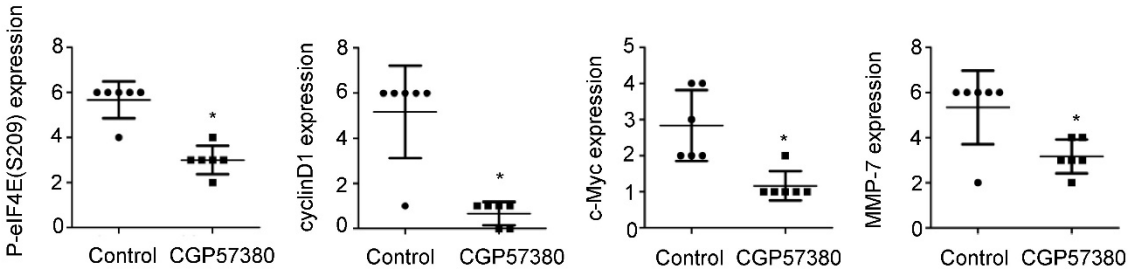
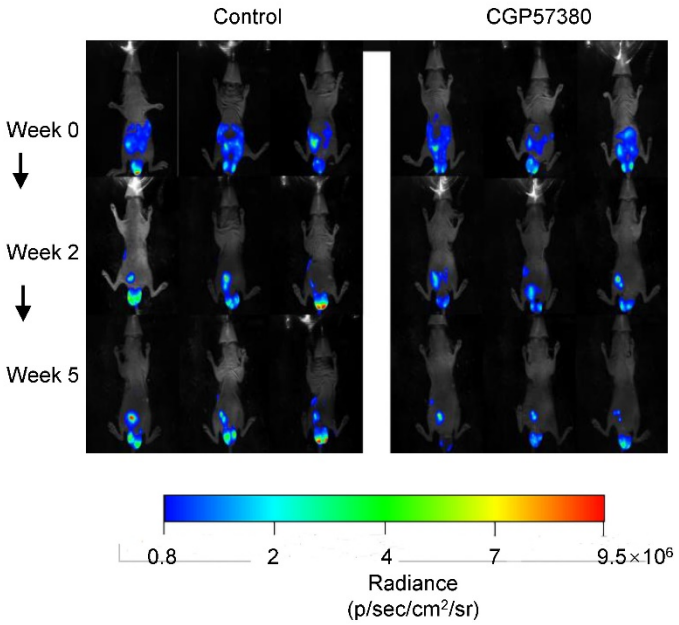


Figure S3

Figure S4. Luminescence signals of intraperitoneal CNE1-Luc tumor xenografts from different treatment groups at the indicated week.



The Intensity Of Radiance

	Control (p/sec/cm²/sr)			CGP57380 (p/sec/cm²/sr)		
Week 0	2320061	2583680	3090454	2650992	2371556	4026934
Week 1	2417273	2147287	2061139	2694774	3806625	3946544
Week 2	4506279	2330429	3564835	2298029	3225885	3050267
Week 3	4556269	3795855	3864579	1992605	2750836	2830549
Week 4	4618231	4230646	4149291	1618487	2408748	2801224
Week 5	6279436	4136284	5797626	1322484	2243071	2625784

Figure S4

Figure S5. Colony formation assay of CNE1 cells with or without CGP57380 was measured 14 days after radiation at a single dose of 0, 2, 4, or 6Gy.

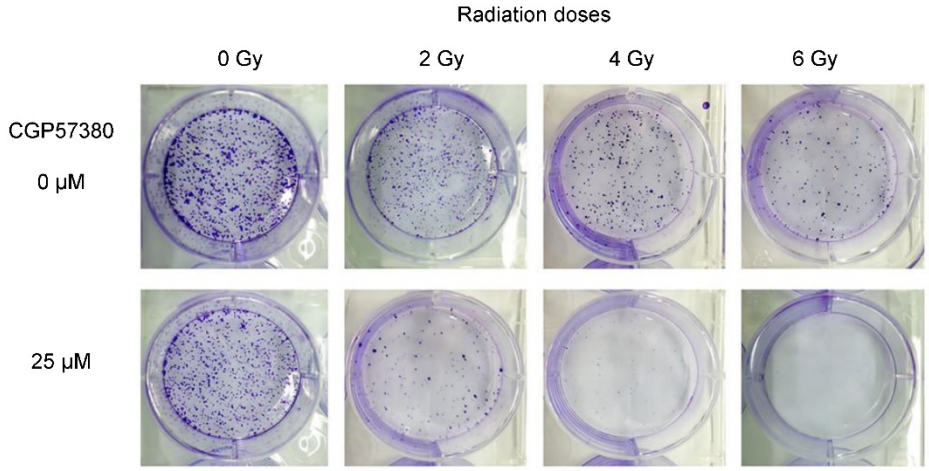


Figure S5

Figure S6. Transwell assay of CNE1 cells treated with DMSO, MNK inhibitor CGP57380, AKT inhibitor MK2206, AKT activator IGF-1, combined treatment with CGP57380 and MK2206, combined treatment with CGP57380 and IGF-1, Wnt inhibitor XAV939, or combined treatment with CGP57380 and XAV939 for 24 h.

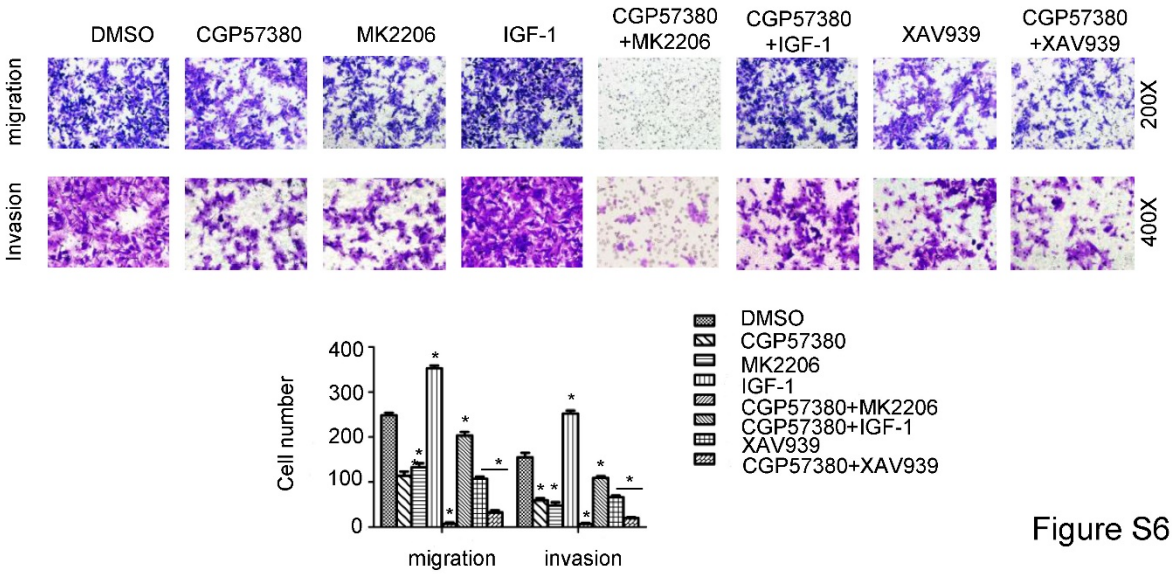


Figure S6

Supplemental MATERIAL AND METHODS:

Antibodies and reagents: A full list of antibodies is included in the following table. Mnk inhibitor CGP57380 (Sigma-Aldrich, USA) was dissolved in DMSO for preparing a working stock solution of 100 mM. Recombinant Human IGF-1 (291-G1) was purchased from R&D company, and diluted in 1 × PBS at the concentration of 25 ng/mL for the treatment. MK2206 (Merch & Co, USA) and XAV939 (Selleck Chemicals) were formulated in DMSO for preparing a stock solution, and diluted in fresh RPMI1640 culture medium at the concentration of 3 μM and 20 μM respectively for the treatment.

Table S1. Antibody list

Antigen	Catalog number	Source	Application&Dilutions
p-Mnk1 (Thr197/202)	2111	Cell signaling	WB (1:1000)
Mnk1	2195	Cell signaling	WB (1:1000)
p-eIF4E(S209)	ab76256	Abcam	WB (1:1000), IHC (1:500)
eIF4E	2067	Cell signaling	WB (1:2000)
p-β-catenin(S33/37/Thr41)	9561S	Cell signaling	WB (1:1000)
p-β-catenin(S552)	5651P	Cell signaling	WB (1:1000)
β-catenin	8480P	Cell signaling	WB (1:1000), IHC (1:500), IF (1:100)
p-GSK-3β(S9)	5558P	Cell signaling	WB (1:1000)
GSK-3β	12456	Cell signaling	WB (1:1000)
p-AKT(S473)	clone 2109Y	Abcam	WB (1:1000)
AKT	4685	Cell signaling	WB (1:1000)
cyclinD1	clone 92G2	Cell signaling	WB (1:1000), IHC (1:500)
c-Myc	5605P	Cell signaling	WB (1:1000), IHC (1:400)
MMP-7	AF907	R&D	WB (1:500), IHC (1:1000)
histone H3	4499P	Cell signaling	WB (1:1000)
CuZnSOD	2770	Cell signaling	WB (1:1000)
E-cadherin	3195	Cell signaling	WB (1:1000), IHC (1:1000)
Vimentin	5741	Cell signaling	WB (1:1000), IHC (1:1000)
PARP	9532	Cell signaling	WB (1:1000)

GAPDH	60004-1-Ig	Proteintech Group	WB (1:1000)

Table S2. Primers and sequences used in this study

gene symbol	**Primer sequence**
c-Myc	Forward: CCTACCCTCTCAACGACAGC
	Reverse: TTCCTCCTCAGAGTCGCTGC
cyclinD1	Forward: CTCCACCTCACCCCCTAAAT
	Reverse: AGAGCCCAAAAGCCATCC
MMP-7	Forward: GGAACAGGCTCAGGACTATCTC
	Reverse: CAACATCTGGCACTCCACA
Axin2	Forward: CTCCTTGGAGGCAAGAGC
	Reverse: GGCCACGCAGCACCGCTG
E-cadherin	Forward: CCTGGGACTCCACCTACAGA
	Reverse: CTGCTTGGATTCCAGAAACG
vimentin	Forward: AGGTGGATCAGCTCACCAATGACA
	Reverse: TCAAGGTCAAGACGTGCCAGAGAA
N-cadherin	Forward: AAATTGAGCCTGAAGCCAAC
	Reverse: GTGGCCACTGTGCTTACTGA
slug	Forward: AGATCTGCCAGACGCGAACT
	Reverse: GCATGCGCCAGGAATGTTCA
β-catenin	Forward: GAGCCTGCCATCTGTGCTCT
	Reverse: ACGCAAAGGTGCATGATTTG
GAPDH	Forward: CCACCCATGGCAAATTCCATGGCA
	Reverse: TCTAGACGGCAGGTCAGGTCCACC