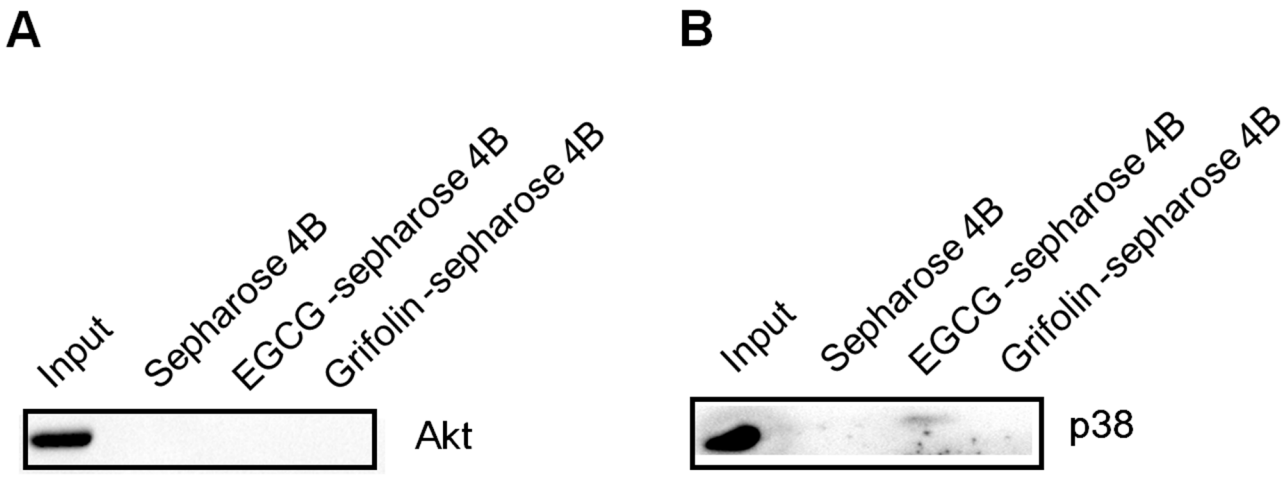
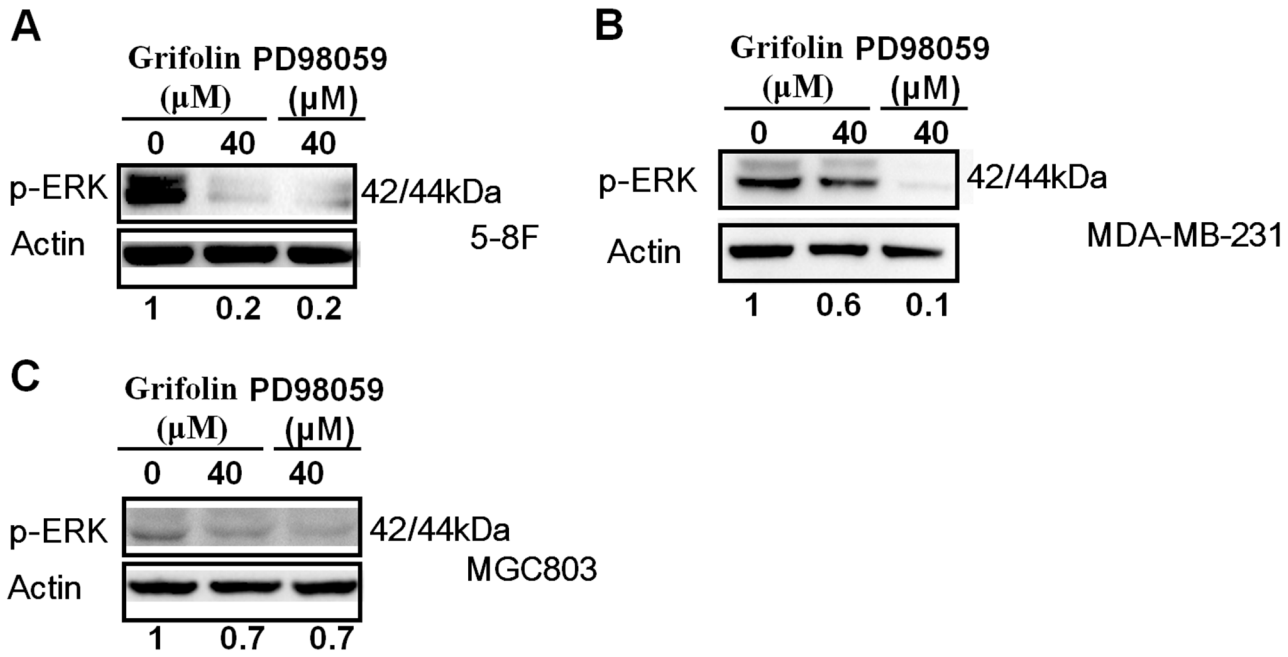
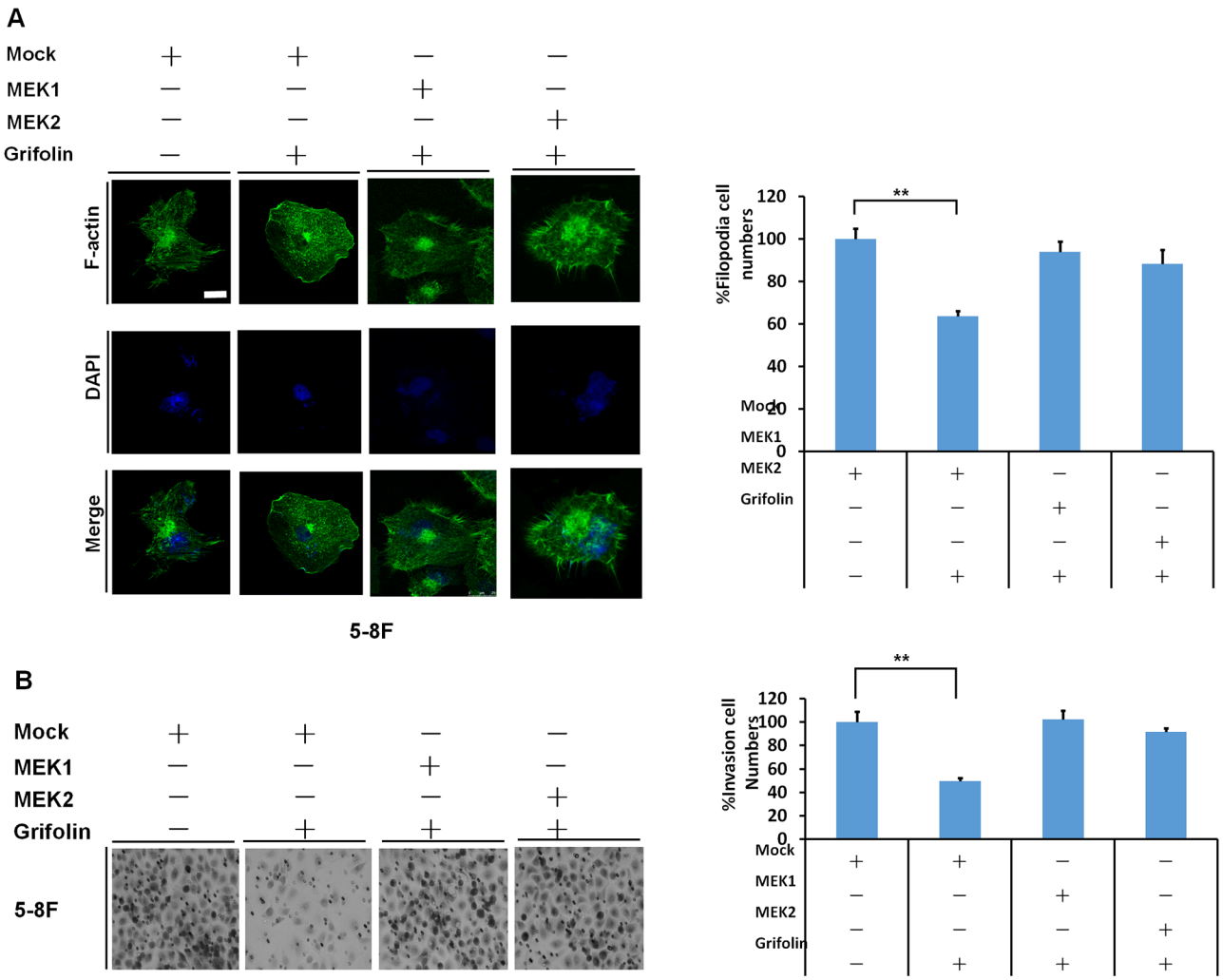
SUPPLEMENTARY FIGURES



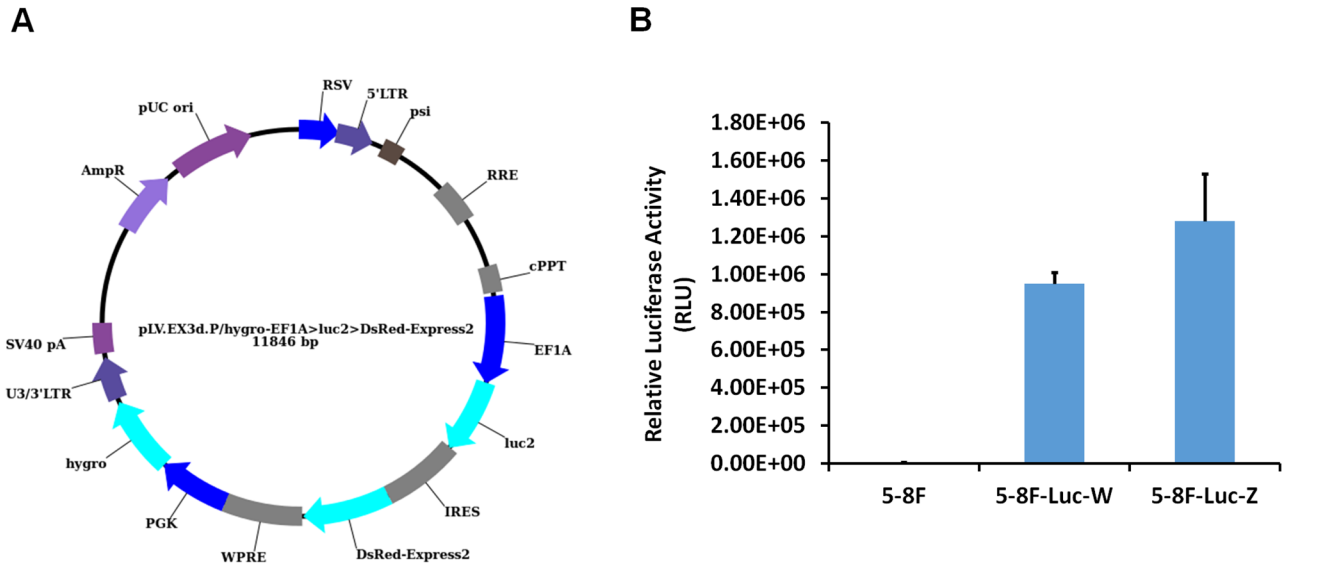
Supplementary Figure S1: The target specificity of grifolin-binding proteins. Immunoblot analysis of whole cell lysate following purification by a Sepharose 4B, grifolin-Sepharose 4B or EGCG-Sepharose affinity column. A. The Akt/grifolin complex or **B.** p38/grifolin complex was detected by Western blot using an antibody against Akt and p38, respectively.



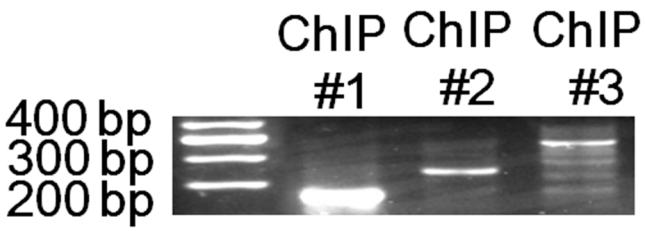
Supplementary Figure S2: Grifolin decreases phosphor-ERK in high-metastatic cancer cells. A. 5–8F, **B.** MDA-MB-231 or **C.** MGC803 cells were treated as designated for 12 h. Cell lysates were prepared and examined by Western blot with an antibody against phosphor-ERK.



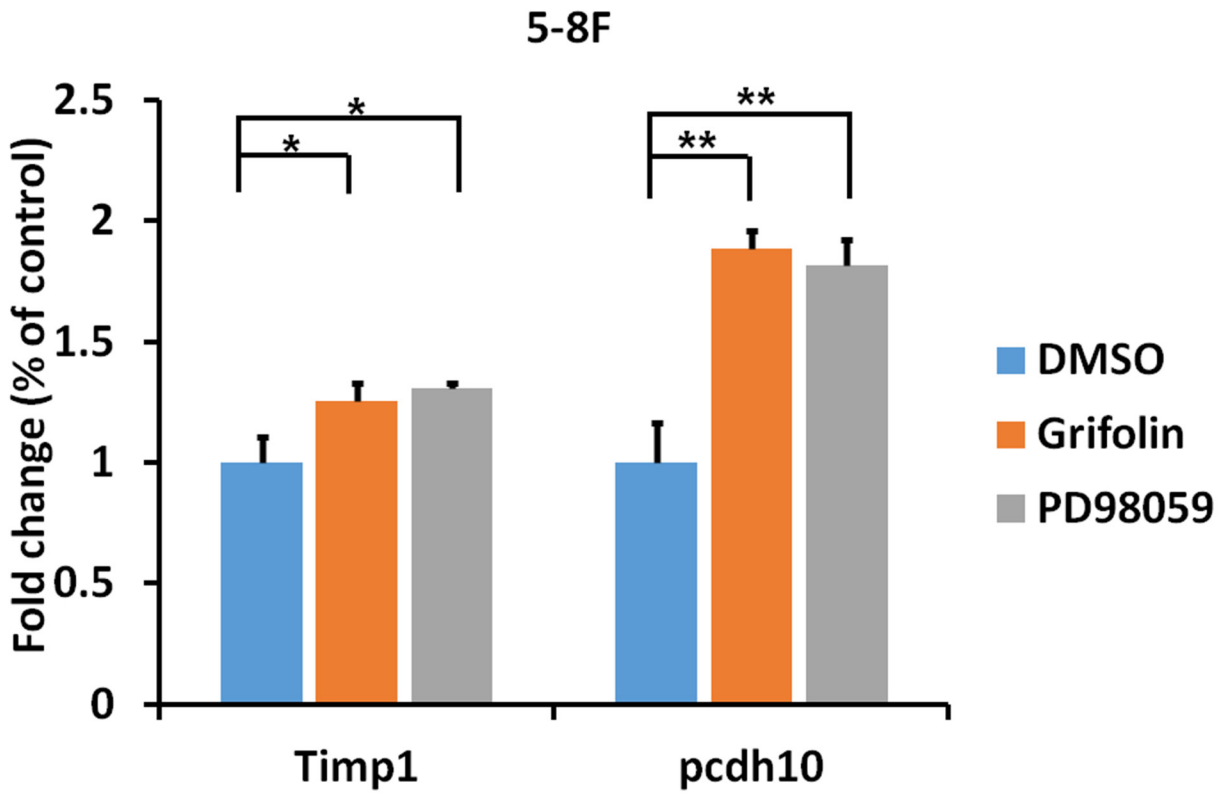
Supplementary Figure S3: Overexpression of constitutively active MEK1/2 rescue filopodia formation and invasion capacity of 5–8F cells suppressed by grifolin treatment. 5–8F cells were transfected with constructs as indicated, followed by grifolin or DMSO (control) treatment for additional 24 h, then cells were applied to (A) immunofluorescence analysis or (B) invasion assay. **A.** Cells were stained with fluorescein isothiocyanate/phalloidin and visualized using Leica TCS SP5 confocal microscopy. Images are representative of at least 3 independent experiments. Scale bar, 25 μm. **B.** Counts were obtained (in triplicate fields of view) from the Mock control (average set at 100% invasion) and were used to calculate percent invasion for all other treatments. Data are shown as mean values ± S.D. of independent, triplicate experiments. The asterisks (**) indicate a significant difference ($P < 0.01$) compared to the Mock control.



Supplementary Figure S4: Establishment of nasopharyngeal carcinoma cell 5–8F-Z stable expression of luciferase. A. The firefly luciferase (luc) gene was inserted into vector pLV.Des3d.P/Hygro by Gateway Recombination Cloning Technology. The construct (pLV.EX3d.P/Hygro-EF1A > luciferase (luc2) > IRES/ DsRed-Express2) was transfected into 5–8F cells to establish two cloned cell line 5–8F-luc-W and 5–8F-luc-Z, which stable expressed luciferase. **B.** The luciferase assay was used to evaluate the luciferase acivity of 5–8F-luc-W and 5–8F-luc-Z, respectively.



Supplementary Figure S5: Evaluation efficiency of different ChIP primers for Elk1 binding to the *dnmt1* promoter. Three pairs of ChIP primers for each potential binding site in the *dnmt1* promoter region were examined for the ability to be enriched by Elk1 immunoprecipitation. ChIP primers were as follows: ChIP 1#, sense 5′-ATCATGG CTCATTGCAGCCTAC-3′; antisense 5′-TCAAGACCAGCCTGAGCAACAT-3′; ChIP 2#, 5′-GCCATG TTG CTC AGGCTGGT -3′; antisense 5′-AACTTAAG GATAGCTATACAGC-3′; ChIP 3#, 5′-GCCATGTTGCTCAGGCTGGT-3′; antisense 5′-AACTATAGTGCTCCTAGAAG-3′. The cross-linked chromatin of 5–8F cell was precipitated with Elk1 antibody. Precipitated DNA was analyzed by PCR using primers that amplified a 158-bp, a 234-bp or a 360-bp sequence corresponding to ChIP 1#, ChIP 2# and ChIP 3#, respectively.



Supplementary Figure S6: Restored mRNA expression of metastasis suppressor gene Timp1 and pcdh10 induced by grifolin. 5–8F cells were treated with DMSO, 40 μM grifolin or 40 μM PD98059 as designated for 24 h. Total RNA was isolated from cells and subjected to real-PCR. PCR primers were as follows: Timp1, sense 5′-CTGCGGATACTTCCACAGGTC-3′; antisense 5′-GCAAGAGTCCATCCTGCAGTT-3′; pcdh10, sense 5′-AGTACGGACACTGAGCACAACC-3′; antisense 5′-CGGCGAGGT CTGTCAACTAGATAG-3′. Data are shown as mean values ±S.D. of independent, triplicate experiments. The asterisks (**) indicate a significant difference ($P < 0.01$) compared to the control.