

Supplementary Figure 1S

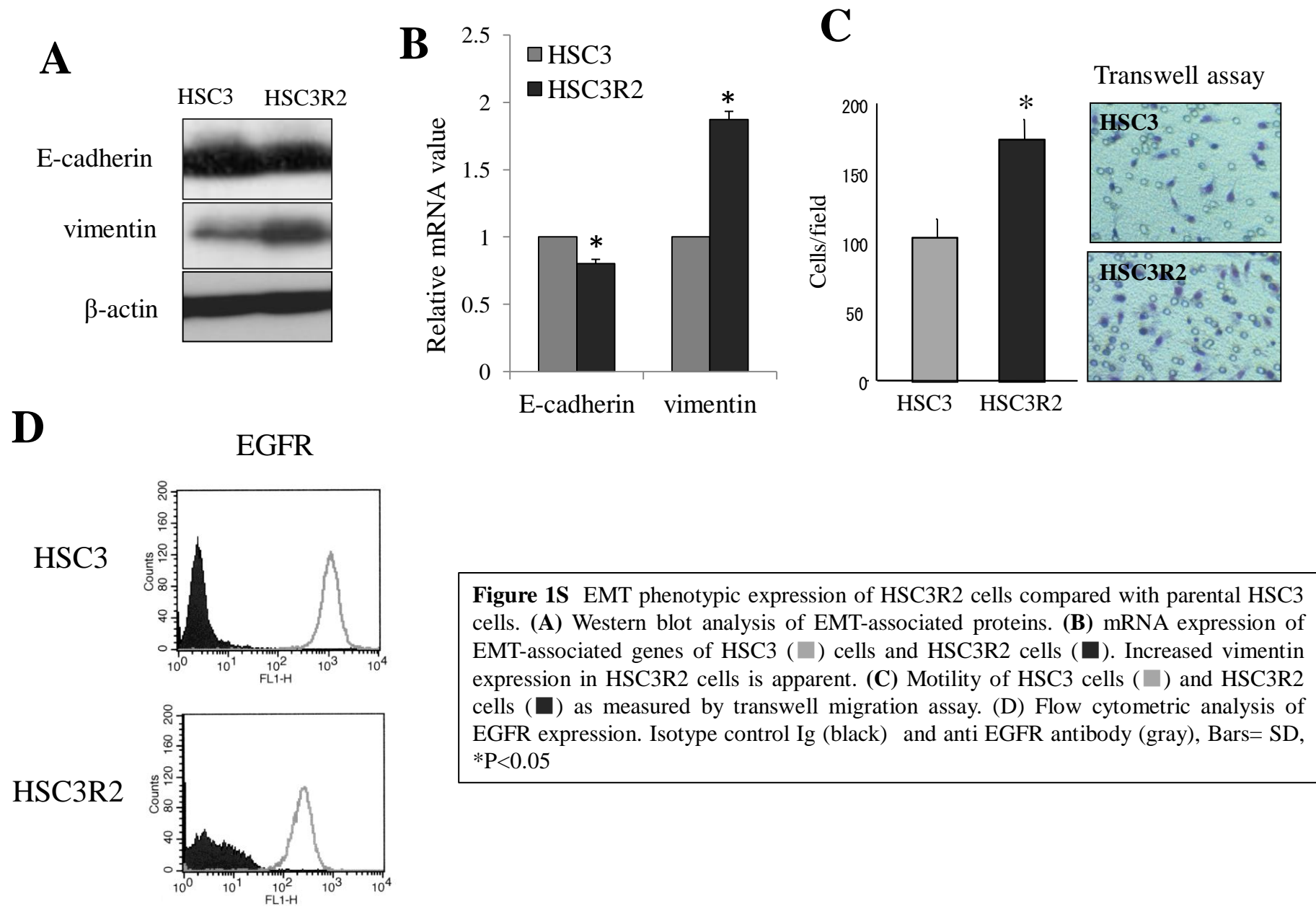


Figure 1S EMT phenotypic expression of HSC3R2 cells compared with parental HSC3 cells. (A) Western blot analysis of EMT-associated proteins. (B) mRNA expression of EMT-associated genes of HSC3 (■) cells and HSC3R2 cells (■). Increased vimentin expression in HSC3R2 cells is apparent. (C) Motility of HSC3 cells (■) and HSC3R2 cells (■) as measured by transwell migration assay. (D) Flow cytometric analysis of EGFR expression. Isotype control Ig (black) and anti EGFR antibody (gray), Bars= SD, *P<0.05

Supplementary Figure 2S

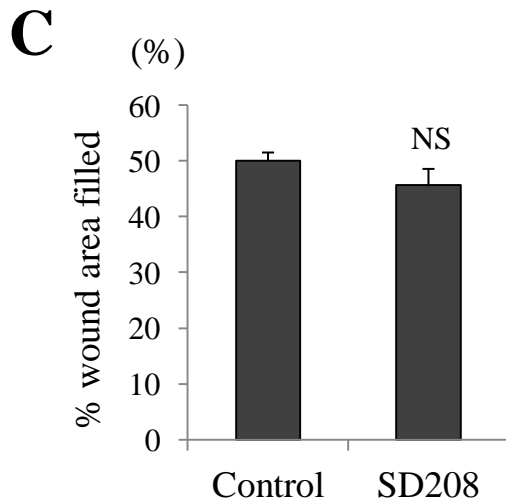
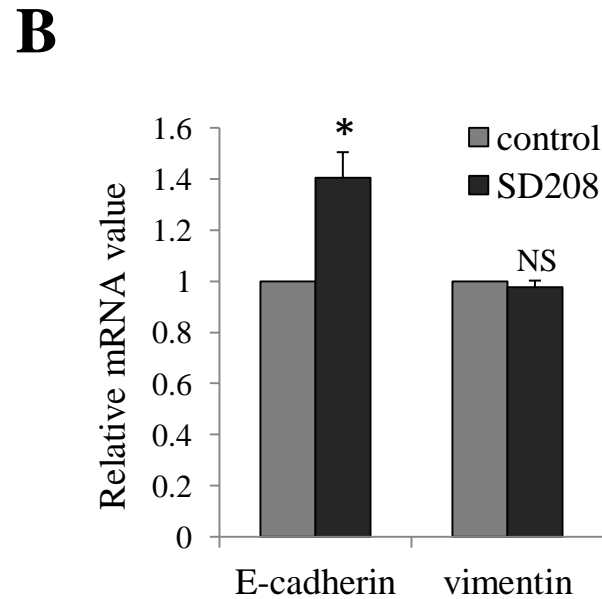
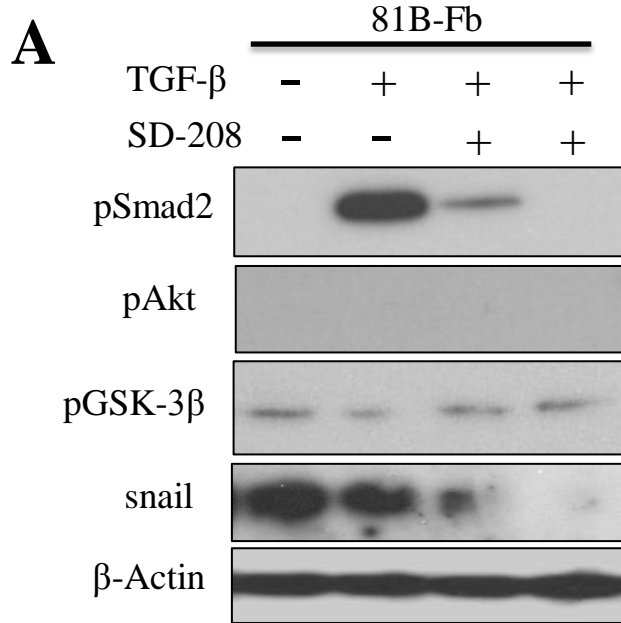


Figure 2S Effects of TGF- β and TGF- β RI inhibitor (SD208) on the EMT phenotype of 81B-Fb cells. (A) Western blot analysis. Cells were starved in serum-free medium for 24 h, and exposed to SD208 at increasing concentrations (1 and 5 μ M) for 2 h at 37°C. Cells were then stimulated with TGF- β (2.5 ng/ml) for 10 min, lysed and subjected to Western blotting. (B) mRNA expression of E-cadherin and vimentin of 81B-Fb cells with or without SD208. (C) Motility of 81B-Fb cells as measured by wound-closure assay. Bars= SD, *P<0.05. NS=not significant.

Supplementary Figure 3S

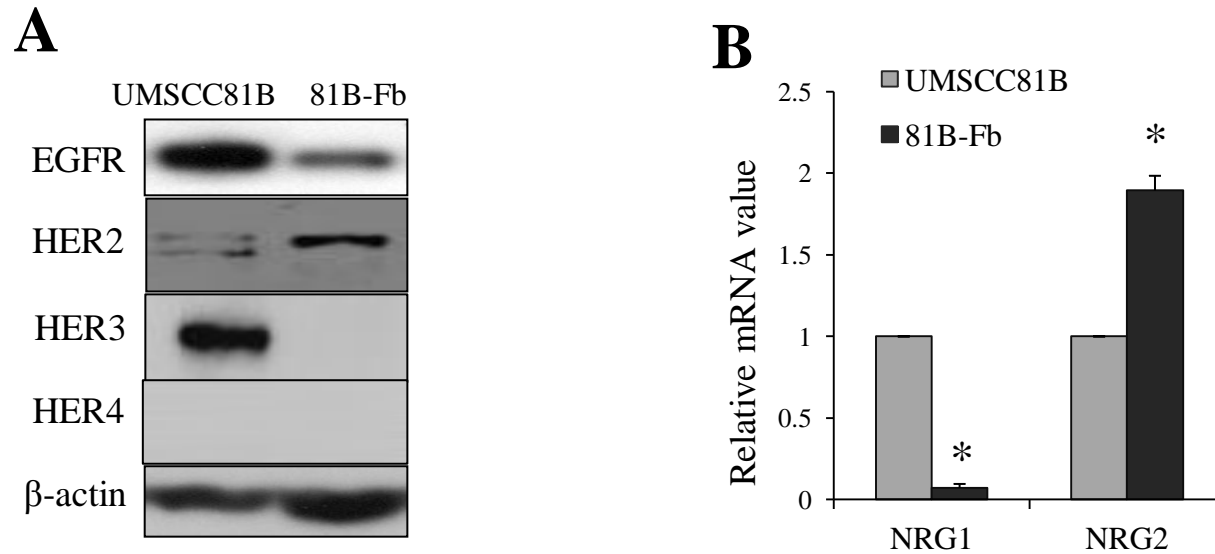
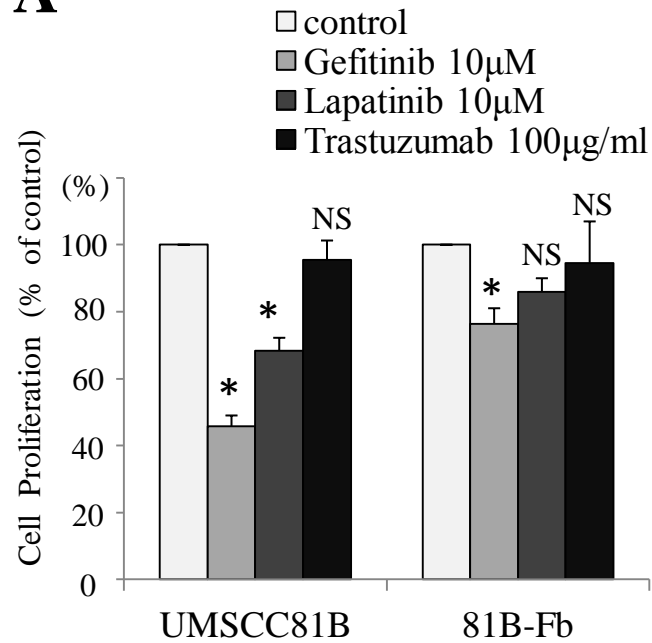


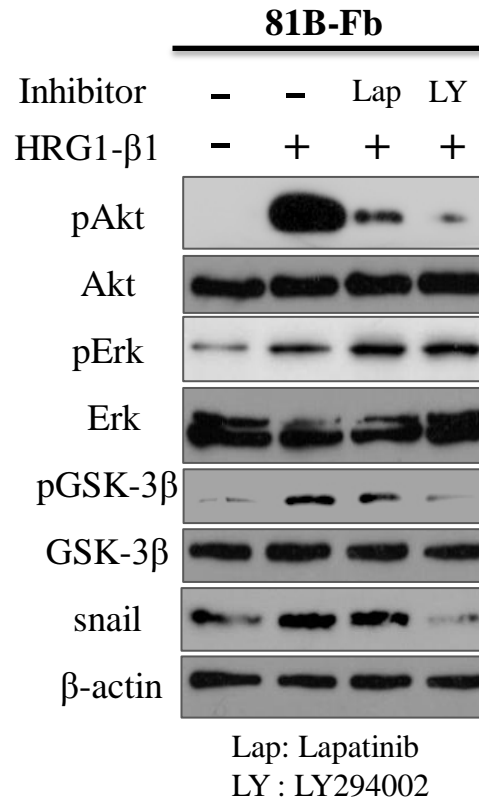
Figure 3S Expression of HER receptor family and neuregulin ligands in UMSCC81B and 81B-Fb cells. **(A)** Western blot analysis of EGFR, HER2, HER3 and HER4 in UMSCC81B and 81B-Fb cells. **(B)** qRT-PCR analysis of NRG1 and NRG2 mRNA expression in UMSCC81B cells (■) and 81B-Fb cells (■). Bars=SD, *P<0.05.

Supplementary Figure 4S

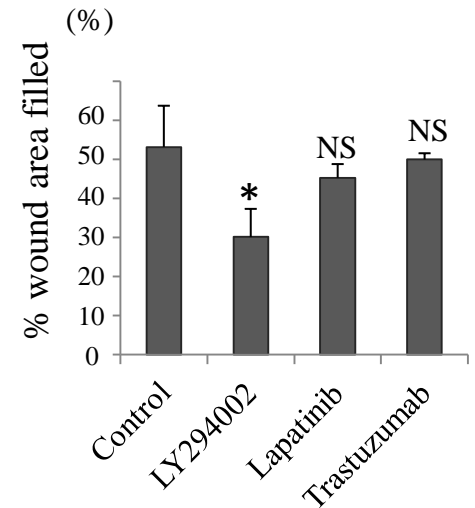
A



B



C



D

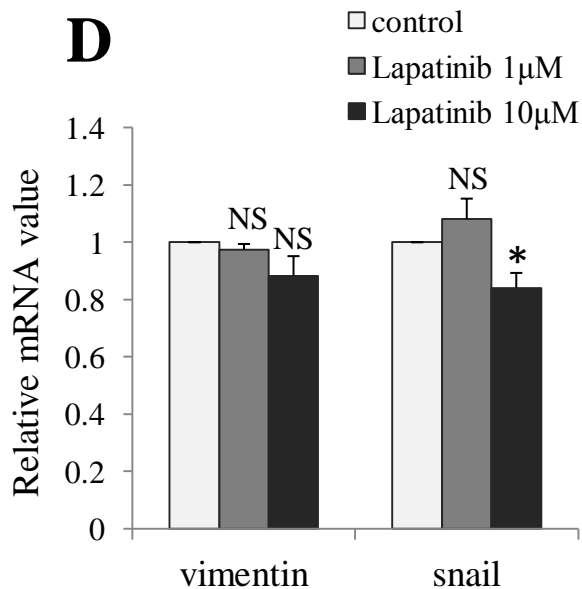


Figure 4S Effects of lapatinib and trastuzumab on the proliferation and EMT phenotype of 81B-Fb cells. (A) Growth inhibition of 81B-Fb cells by gefitinib, lapatinib and trastuzumab. (B) Western blot analysis of Akt, GSK-3 β and snail expression in response to heregulin- β 1 with or without lapatinib (LY294002) treatment. (C) Inhibition of motility of 81B-Fb cells by LY294002, lapatinib and trastuzumab treatment (12 h). (D) Effect of lapatinib treatment (24 h) on the vimentin and snail mRNA expression of 81B-Fb cells. Bars= SD, *P<0.05. NS=not significant.

Supplementary Table 1S

Sequences of primer-pairs for qRT-PCR used in this study

	Forward	Reverse	probe number
E-cadherin	cctggcctcagaagacagaa	ttgtctttgttgatttgatctg	# 44
vimentin	aaagtgtggctgccaagaac	agcctcagagaggtcagcaa	# 16
snail	gctgcaggactctaataccaga	atctccggaggtgggatg	# 11
twist	aaggcatcactatggactttctct	gccagtttgatcccagtatctt	# 6
slug	ttatgttctctataaaacactgactg	tgcagtgagggcaagaaaa	# 7
EGF	aagaatgggggtcaaccagt	tgaagttggttgattgacc	# 27
HB-EGF	tggggcttctcatgttagg	catgcccaacttcactttctc	# 55
amphiregulin	cggagaatgcaaataatagagcac	caccgaaatattcttgctgaca	# 38
EGFR	cgggacatagtcagcagtga	gggacagcttgatcacact	# 44
NRG1	cttgtggctggcatcatgt	cagcttttccgctgtttct	# 49
NRG2	caaatatggcaacggcaga	ggcctcgcagacatactcc	# 39
GAPDH	agccacatcgctcagacac	gcccaatacgaccaaatcc	# 60