Supplementary Data

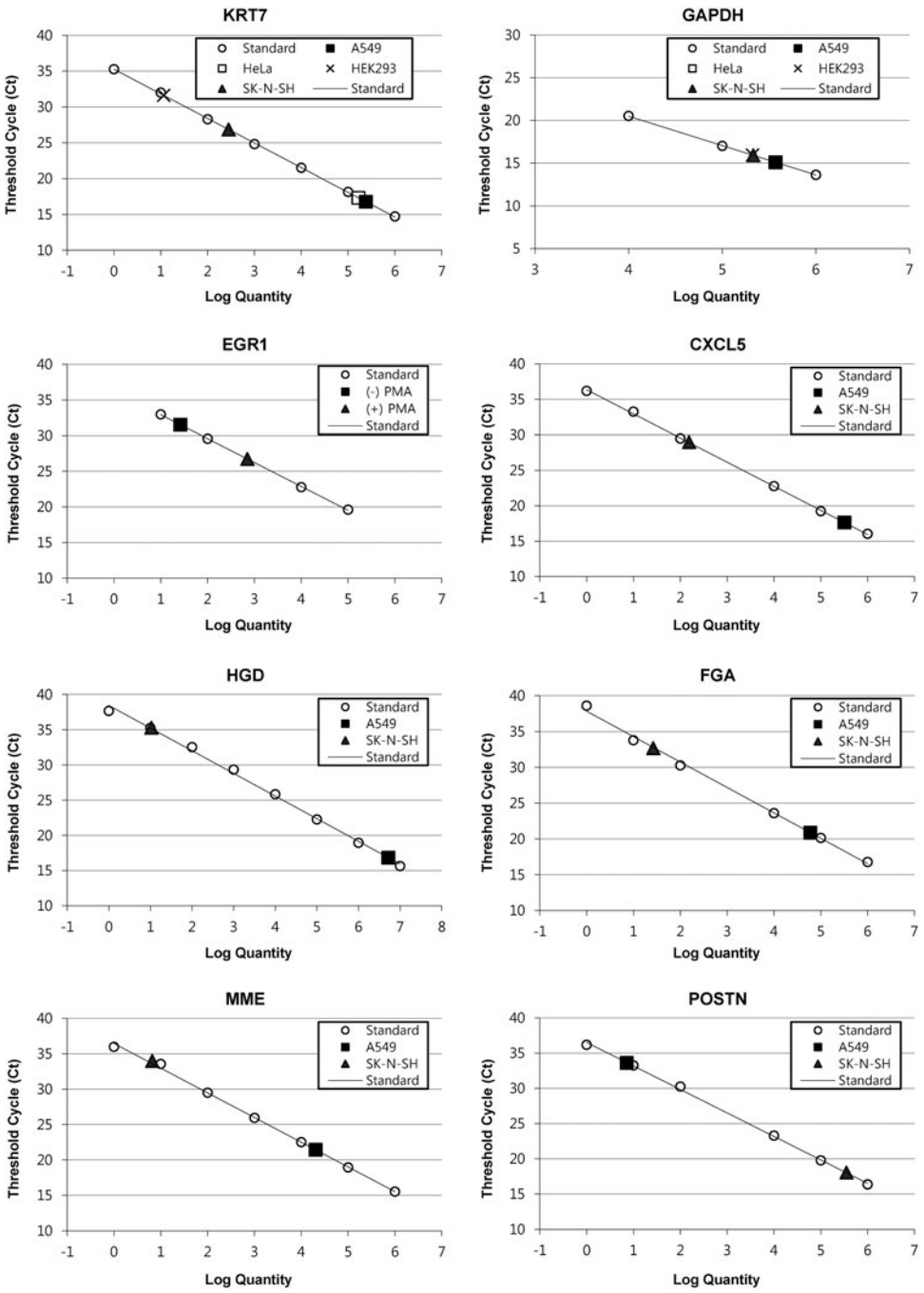
Supplementary Materials and Methods

Quantification of delivered siRNAs

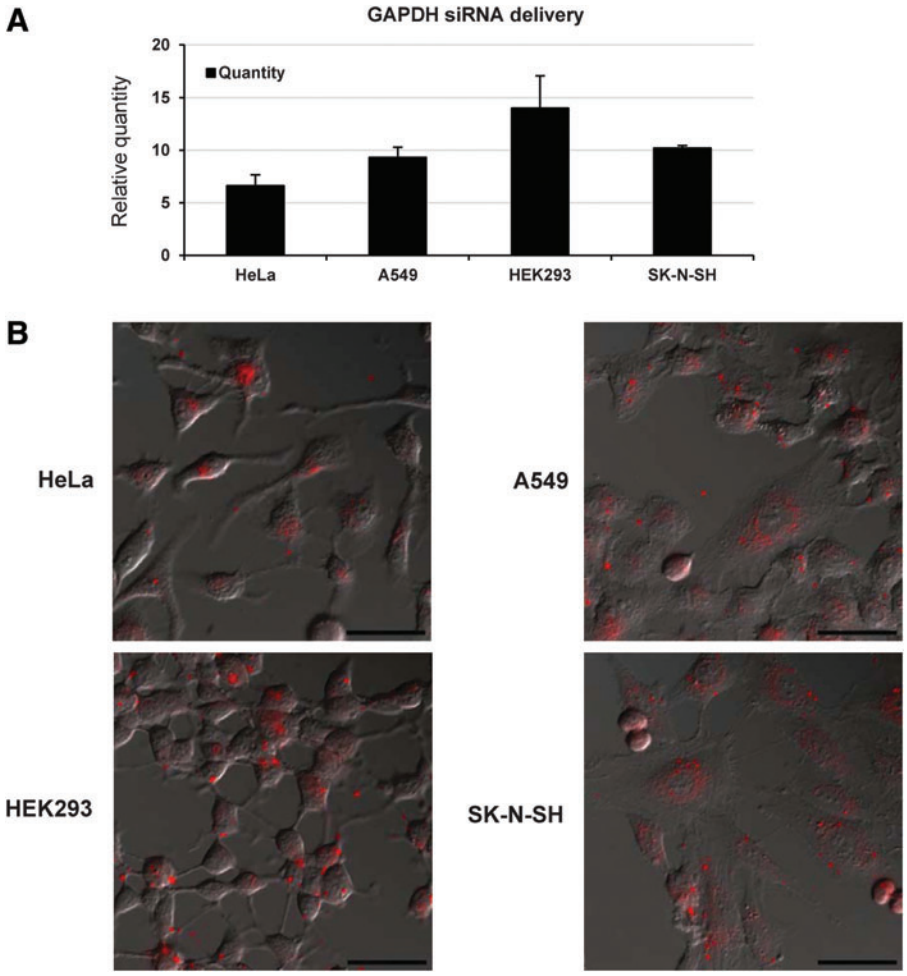
After transfection, cells were washed in three times with phosphate buffered saline and total RNA was extracted using an Iso-RNA Lysis reagent according to manufacturer's instructions. Ten nanograms of total RNA was processed to Custom TaqMan Small RNA Assays (Applied Biosystems, Foster City, CA, USA). According to the manufacturer's protocol, reverse transcription targeting siGAPDH-2 was performed using the custom synthesized stem-loop RT primer specific to the siGAPDH-2 antisense strand and TaqMan MicroRNA Reverse Transcription Kit (Applied Biosystems). For quantitative PCR, TaqMan Small RNA Assay primers set specific to the siGAPDH-2 antisense strand and TaqMan Universal PCR Master Mix II, No UNG (Applied Biosystems) was used. Real-time PCR experiments were done with an StepOne Real-Time PCR System (Applied Biosystems).

Fluorescence microscopy analysis

Cells grown on glass bottom petri dishes were transfected with siGAPDH-2 labeled with Cy3 at the 3′ end of antisense strand (50 nM final) for 4 hours. Following 2 quick washes, images were acquired using fluorescence microscope (Olympus IX-81) and Metaview imaging software at ×400 magnification.



SUPPLEMENTARY FIG. S1. Standard curve plots for the analyzed genes in this study. The standard curve was generated by plotting the cycle threshold (C_T) values obtained against the logarithm of a known quantity of polymerase chain reaction (PCR) targets. Data from each condition [cell lines or phorbol-12-myristate-13-acetate (PMA) non-induction/ induction] are also presented and results show that C_T values are positioned within linear range of the standard curve.



SUPPLEMENTARY FIG. S2. Analysis of small interfering RNS (siRNA) cellular delivery in four cell lines. **(A)** Quantification of the delivered siRNA. siGAPDH-2 was transfected into each cell lines (10 nM final) for 6 hours and delivered siRNA was quantified through the TaqMan quantitative reverse transcription–polymerase chain reaction (qRT-PCR) assays. Data in the graph represent mean ± SD values of three independent experiments. **(B)** Fluorescence visualization of delivered siRNA. Cells were transfected with 3′-Cy-3-labeled siGAPDH-2 (50 nM final) for 4 hours. Images were acquired at ×400 magnification, and overlay of DIC and Cy3 fluorescence images is presented. The scale bar represents 50 μm.

Supplementary Table S1. Small Interfering RNA Information

Gene name	*Accession number*	*siRNA target site*	*RNA oligo name*	*Sequence*
KRT7	NM_005556	645–663	siKRT7-1 sense	5′-UGGAGGACUUCAAGAAUAA(dTdT)-3′
			siKRT7-1 antisense	5′-UUAUUCUUGAAGUCCUCCA(dTdT)-3′
KRT7		927–945	siKRT72 sense	5′-AGUAUGAGGAGAUGGCCAA(dTdT)-3′
			siKRT7-2	5′-UUGGCCAUCUCCUCAUACU(dTdT)-3′
GAPDH	NM_002046	660–678	siGAPDH-1 sense	5′-GGUCAUCCAUGACAACUUU(dTdT)-3′
			siGAPDH-1 antisense	5′-AAAGUUGUCAUGGAUGACC(dTdT)-3′
GAPDH		316–334	siGAPDH-2 sense	5′-CAAUAUGAUUCCACCCAUG(dTdT)-3′
			siGAPDH-2 sense	5′-CAAUAUGAUUCCACCCAUG(dTdT)-3′
EGR1	NM_001964	1536–1554	siEGR1 sense	5′-GGACAAGAAAGCAGACAAA(dTdT)-3′
			siEGR1 antisense	5′-UUUGUCUGCUUUCUUGUCC(dTdT)-3′
CXCL5	NM_002994	398–416	siCXCL5 sense	5′-CUGAAGAACGGGAAGGAAA(dTdT)-3′
			siCXCL5 antisense	5′-UUUCCUUCCCGUUCUUCAG(dTdT)-3′
HGD	NM_000187	817–835	siHGD sense	5′-GUGUGGAGCUGGAGACAUA(dTdT)-3′
			siHGD antisense	5′-UAUGUCUCCAGCUCCACAC(dTdT)-3′
FGA	NM_021871	676–694	siFGA sense	5′-GGUCAUUGCCAAAGACUUA(dTdT)-3′
			siFGA antisense	5′-UAAGUCUUUGGCAAUGACC(dTdT)-3′
MME	NM_000902	1530–1548	siMME sense	5′-AGAAAAGGCCUUAGCAAUU(dTdT)-3′
			siMME antisense	5′-AAUUGCUAAGGCCUUUUCU(dTdT)-3′
POSTN	NM_006475	486–504	siPOSTN sense	5′-GGGAGGAGATCGAGGGAAA(dTdT)-3′
			siPOSTN antisense	5′-UUUCCCUCGTUCUCCUCCC(dTdT)-3′
*Luciferase**	pTRE3G-Luc	1885–1903	siLuc sense	5′-UUGUUUUGGAGCACGGAAA(dTdT)-3′
			siLuc antisense	5′-UUUCCGUGCUCCAAAACAA(dTdT)-3′

*For Luciferase targeting small interfering (siRNA), target site is based on the pTRE3G-Luc (Clontech) vector used in this study.

Supplementary Table S2. Primer Information

Gene	*Name*	*Primer name*	*Sequence*
GAPDH	GAPDH-forward GAPDH-reverse	5′-GAGTCAACGGATTTGGTCGT-3′ 5′-GACAAGCTTCCCGTTCTCAG-3′	184
KRT7	KRT7-forward KRT7-reverse	5′-GAGCCGGTTGGCTGGAGATGGAG-3′ 5′-CAGGGCATTGCTGCCCATGGTTC-3′	123
EGR1	EGR1-forward EGR1-reverse	5′-ACCTGACCGCAGAGTCTTTTC-3′ 5′-GCCAGTATAGGTGATGGGGG-3′	109
CXCL5	CXCL5-forward CXCL5-reverse	5′-CCACTATGAGCCTCCTGTCC-3′ 5′-TGGGATGAACTCCTTGCGTG-3′	185
HGD	HGD-forward HGD-reverse	5′-GACGAAGGCCAAGTCACTCA-3′ 5′-AAGGTATGCAGGCCACTCAC-3′	127
FGA	FGA-forward FGA-reverse	5′-ACGCGTCGTTCATGCTCTAA-3′ 5′-GAAGGCAGCTTCATCAGGGT-3′	188
MME	MME-forward MME-reverse	5′-ATCAAGTTTGGGTCTGTGCTG-3′ 5′-TCCAGCTGTGGCAACAAGAAT-3′	102
POSTN	POSTN-forward POSTN-reverse	5′-TGGAAACCATCGGAGGCAAA-3′ 5′-AATCGCACCGTTTCTCCCTT-3′	109
TUBA1A	TUBA1A-forward TUBA1A-reverse	5′-GACCAAGCGTACCATCCAGT-3′ 5′-CACGTTTGGCATACATCAGG-3′	199