Table S1: Patient demographics and pathologic information. 19 HER2(+) (3+ by immunohistochemistry and/or amplified by FISH) and 20 HER2(-) invasive breast carcinomas were reviewed by a board-certified pathologist (D.A.D.). IQGAP1 immunostaining was scored as described in the Methods. Patient age and hormone receptor status are shown. ER, estrogen receptor; PR, progesterone receptor; HR(+), hormone receptor-positive; TNBC, triple-negative breast cancer; AMP, amplified; POS, positive; NEG, negative.

Patient	Diagnosis	Age at Diagnosis (years)	HER2	ER	PR	IQGAP1
1	HER2(+)	34	3+	POS	NEG	3+ (POS)
2	HER2(+)	42	3+	NEG	NEG	3+ (POS)
3	HER2(+)	37	3+	NEG	NEG	3+ (POS)
4	HER2(+)	54	3+	NEG	NEG	3+ (POS)
5	HER2(+)	33	3+	POS	POS	3+ (POS)
6	HER2(+)	53	3+	POS	POS	3+ (POS)
7	HER2(+)	39	3+	POS	POS	3+ (POS)
8	HER2(+)	31	3+	POS	POS	2+ (POS)
9	HER2(+)	52	3+	POS	POS	2+ (POS)

10	HER2(+)	61	3+	NEG	NEG	2+ (POS)
11	HER2(+)	63	AMP	NEG	NEG	3+ (POS)
12	HER2(+)	61	AMP	POS	POS	2+ (POS)
13	HER2(+)	55	AMP	NEG	NEG	2+ (POS)
14	HER2(+)	78	AMP	NEG	NEG	2+ (POS)
15	HER2(+)	49	AMP	NEG	NEG	2+ (POS)
16	HER2(+)	71	AMP	POS	NEG	2+ (POS)
17	HER2(+)	58	AMP	POS	NEG	1+ (NEG)
18	HER2(+)	92	AMP	NEG	NEG	1+ (NEG)
19	HER2(+)	49	AMP	POS	POS	1+ (NEG)

20	HR(+)	59	NEG	POS	POS	2+ (POS)
21	HR(+)	60	NEG	POS	POS	2+ (POS)
22	HR(+)	60	NEG	POS	POS	2+ (POS)
23	HR(+)	63	NEG	POS	POS	2+ (POS)
24	HR(+)	63	NEG	POS	POS	1+ (NEG)
25	HR(+)	35	NEG	POS	POS	1+ (NEG)
26	HR(+)	51	NEG	POS	NEG	1+ (NEG)
27	HR(+)	53	NEG	POS	POS	1+ (NEG)
28	HR(+)	69	NEG	POS	POS	1+ (NEG)
29	HR(+)	51	NEG	POS	POS	1+ (NEG)

30	HR(+)	70	NEG	POS	POS	1+ (NEG)
31	HR(+)	44	NEG	POS	POS	1+ (NEG)
32	HR(+)	50	NEG	POS	NEG	1+ (NEG)
33	HR(+)	66	NEG	POS	POS	1+ (NEG)
34	HR(+)	64	NEG	POS	NEG	1+ (NEG)
35	HR(+)	79	NEG	POS	POS	0 (NEG)
36	HR(+)	66	NEG	POS	POS	0 (NEG)
37	HR(+)	46	NEG	POS	POS	0 (NEG)
38	TNBC	76	NEG	NEG	NEG	1+ (NEG)
39	TNBC	84	NEG	NEG	NEG	1+ (NEG)

Fig. S1: Knockdown of IQGAP1 reduces HER2 half-life. A, SkBR3 cells were transiently transfected with siRNA against renilla luciferase (siRen) or siRNA against IQGAP1 (siIQ12). 36 h after siRNA transfection, cells were serum-starved for 48 h. Total RNA was extracted, DNAse treated and subjected to quantitative RT-PCR analysis to determine HER2 mRNA expression. RT- and H_2O controls were included in all RT-PCR plates to verify the specificity of PCR amplification. HER2 mRNA was normalized to that of GAPDH. The data in the bottom panel represent the mean ± SE (n = 4). B, SkBR3 cells were transiently transfected with siRNA against renilla luciferase (siRen) or siRNA against IQGAP1 (siIQ12). Immediately after siRNA transfection, cells were incubated with 100 μg/ml cyclohexamide (CHX) for 0, 4, 16, 24 or 40 h. Equal amounts of protein were resolved by SDS-PAGE, transferred to PVDF membranes and probed with anti-HER2 and anti-β-Tubulin antibodies. The data are representative of 3 independent experiments. C, the amount of HER2 was quantified by densitometry and corrected for the amount of β-Tubulin in the corresponding lysate. Samples which were transfected with siRen are depicted by black squares and those transfected with siIQ12 are depicted by white squares. The data, expressed relative to the amount of HER2 in control cells transfected with siRen, represent the mean ± SE (n = 3). *, significantly different from control cells transfected with siRen and treated for 0 h ($p < 0.05$); θ, significantly different from cells transfected with siRen ($p < 0.05$).

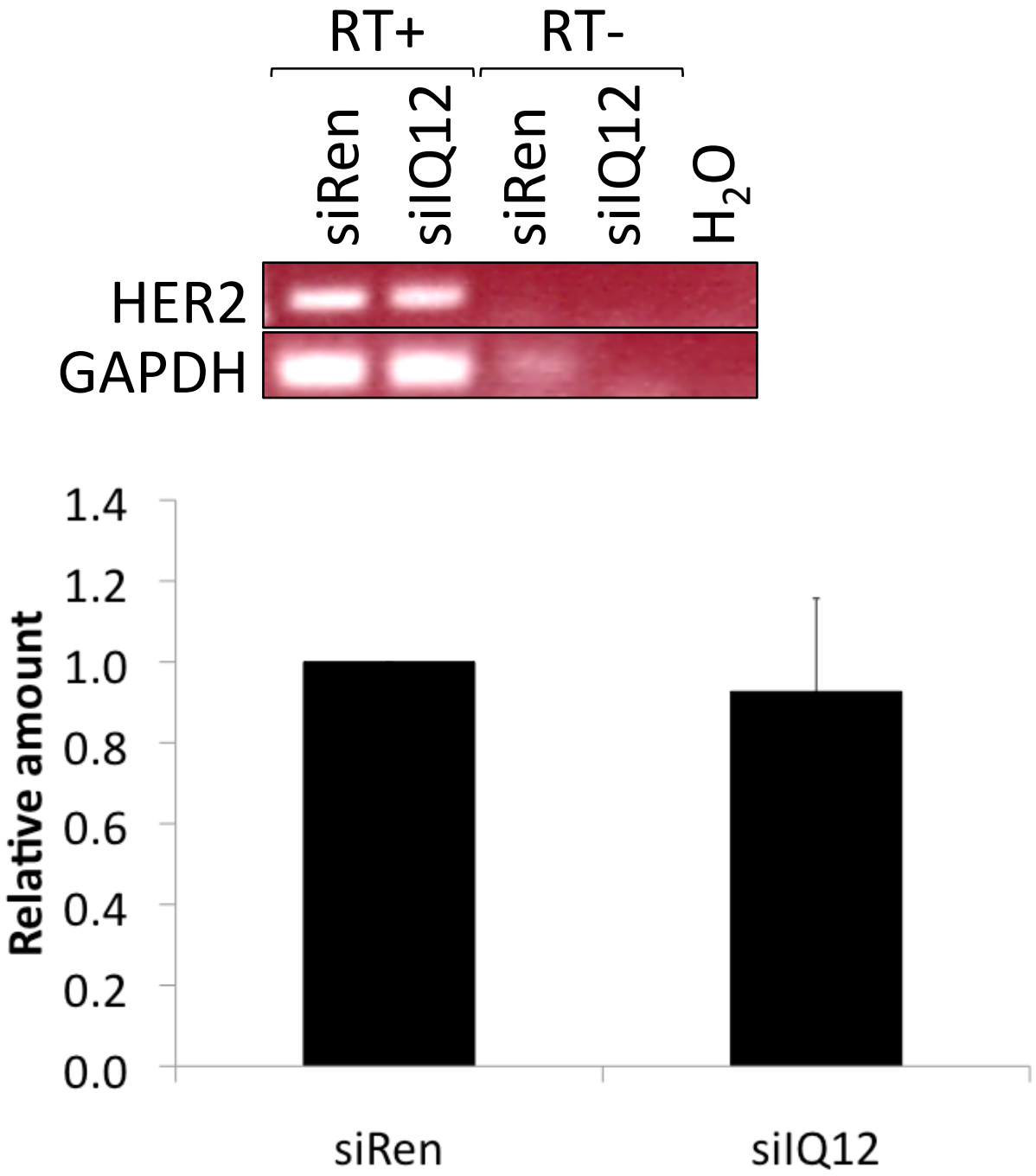
RT+
RT-
siRen
siIQ12
siRen
siIQ12
H2O
HER2
GAPDH
Relative amount
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
siRen
siIQ12

Figure S1A

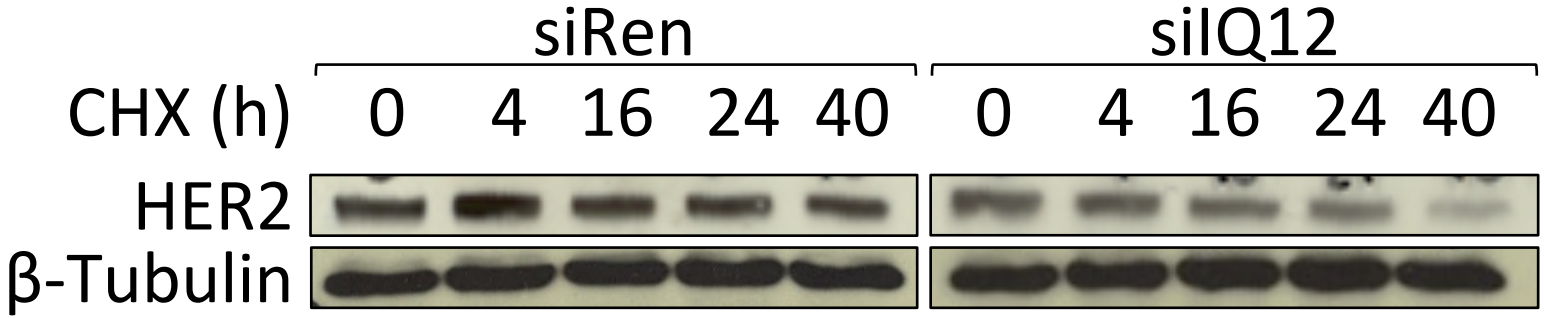

Figure S1B

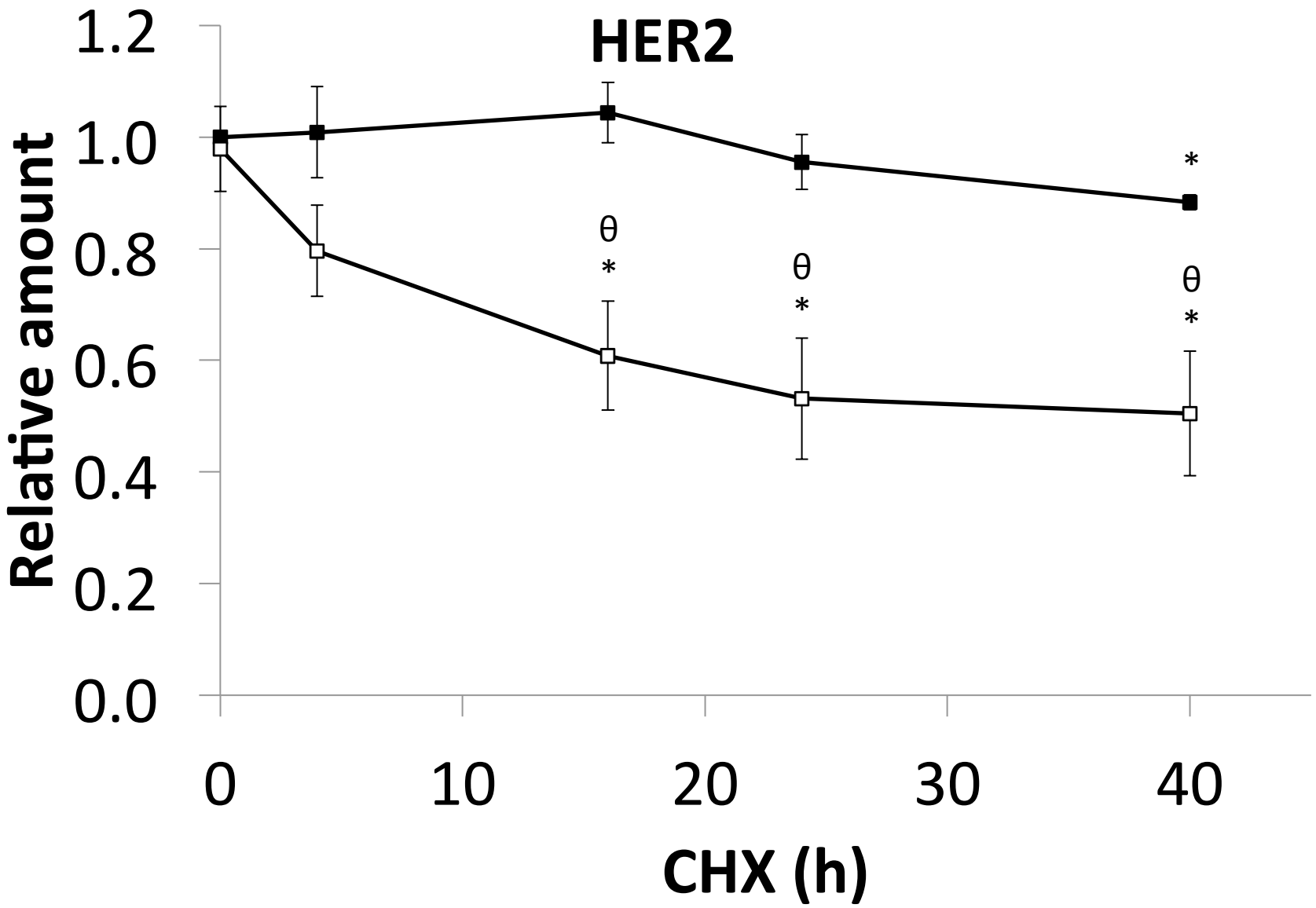

Figure S1C

Fig. S2: Neither knockdown of IQGAP1 nor trastuzumab significantly effects ERK activation. A, the amount of pERK in Fig. 5A was quantified by densitometry and corrected for the amount of total ERK in the corresponding lysate. Samples which were treated with vehicle are depicted by black bars and those treated with trastuzumab are depicted by white bars. The data, expressed relative to the amount of total ERK in control cells transfected with siRen and treated with vehicle, represent the mean ± SE (n = 5). B, the amount of pERK in Fig. 5C was quantified by densitometry and corrected for the amount of total ERK in the corresponding lysate. Samples which were treated with vehicle are depicted by black bars and those treated with trastuzumab are depicted by white bars. The data, expressed relative to the amount of total ERK in control cells transfected with siRen and vector and treated with vehicle, represent the mean ± SE (n = 3). C, the amount of pERK in Fig. 10A was quantified by densitometry and corrected for the amount of total ERK in the corresponding lysate. Samples which were treated with vehicle are depicted by black bars and those treated with trastuzumab are depicted by white bars. The data, expressed relative to the amount of total ERK in control cells transfected with siRen and treated with vehicle, represent the mean ± SE (n = 5).

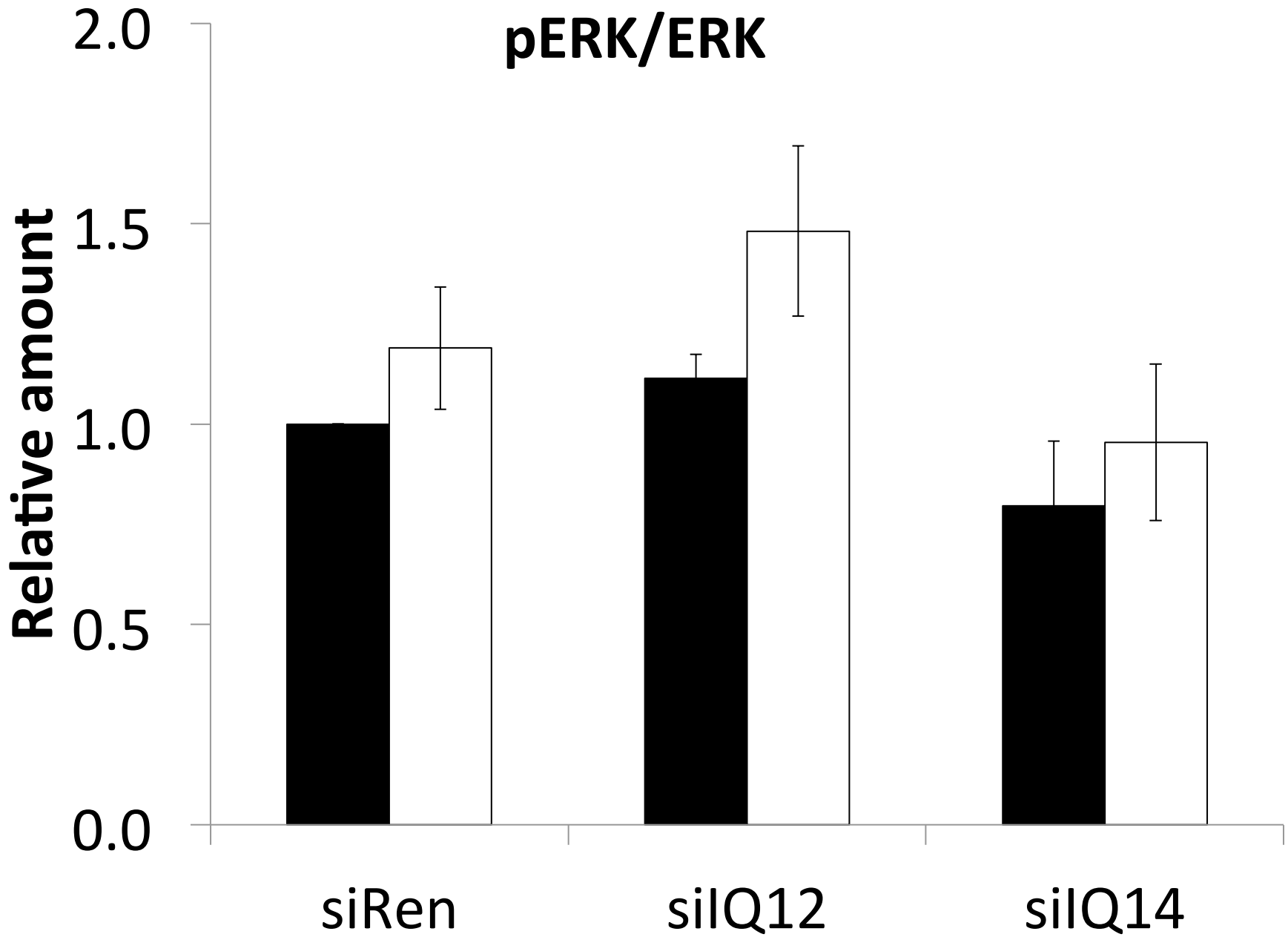

Figure S2A

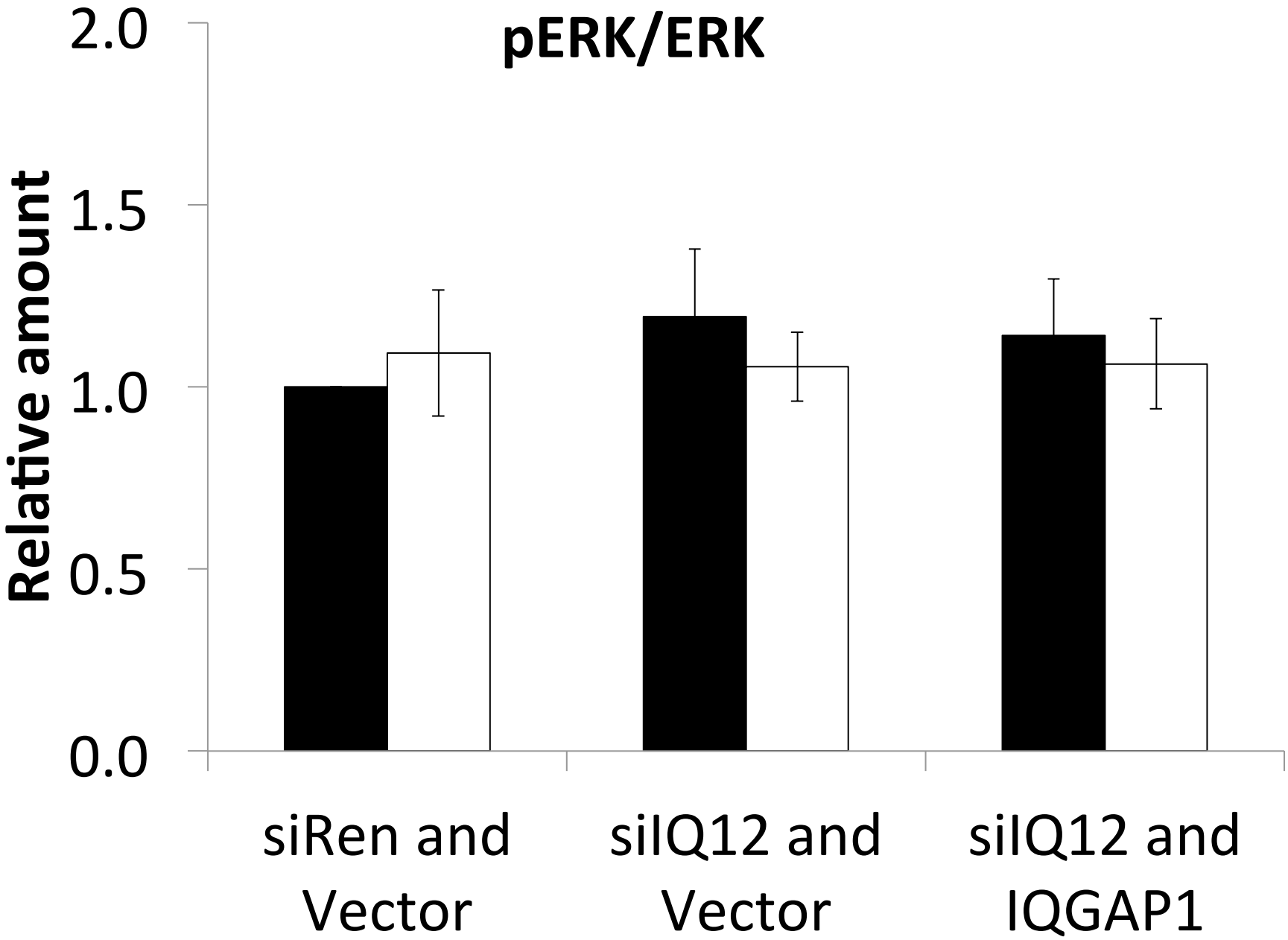
pERK/ERK
Relative amount
2.0
1.5
1.0
0.5
0.0
siRen and Vector
siIQ12 and Vector
siIQ12 and IQGAP1

Figure S2B

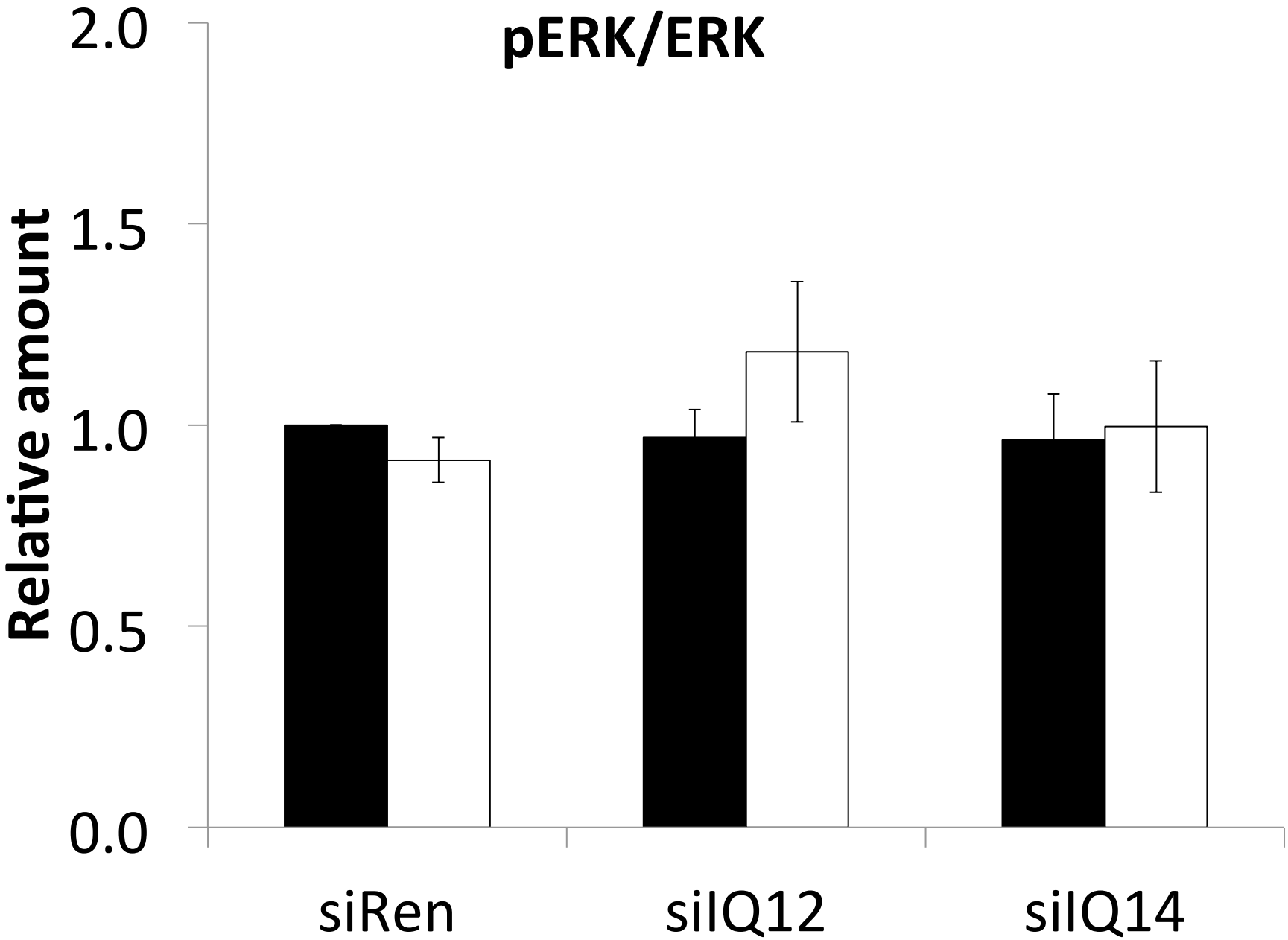

Figure S2C