Table S1. Thyroid cancer cell lines and their genetic alterations

Cell line	Tumor origin	Genetic alterations
C643	ATC	*HRAS* (G13R+/-)
Hth7	ATC	*NRAS* (Q61R+/-), *AKT1* copy gain
FTC133	FTC	*PTEN* (R130* +/+)
OCUT1	ATC	*BRAF* (V600E+/-), *PIK3CA* (H1047R+/+)
K1	PTC	*BRAF* (V600E+/-), *PIK3CA* (E542K+/+)
SW1736	ATC	*BRAF* (V600E+/-)
BCPAP	PTC	*BRAF* (V600E+/+), *AKT1* copy gain
TPC1	ATC	*RET/PTC1* Rearrangement
KAT18	ATC	–
Hth74	ATC	–
WRO	FTC	–

+/-, heterozygous mutation; +/+, homozygous mutation

Table S2. Primer sequences used for quantitative RT-PCR analysis of the expression of thyroid iodide-handling genes

Genes	**Forward primer (5'→3')**	**Reverse primer (5'→3')**	**Product length**	**Annealing Temp.**
NIS (NM_000453)	CCTGCTAACGACTCCAGCA	CCAGGGCACCGTAATAGAGA	106bp	60°C
TSHR (NM_000369)	GATATTCAACGCATCCCCAG	AGCTGCTGCAGAGTCACATC	149bp	60°C
TPO (NM_000547)	ACTTGGATCTCCATGTCGCT	GCAGTGTGGATTTAGTGCCA	106bp	60°C
Tg (NM_003235)	CACCAACTCCCAACTTTTCC	CAACTGACCTCCTTTGCCA	123bp	60°C
PAX8 (NM_003466)	TGCCTCACAACTCCATCAGA	CAGGTCTACGATGCGCTG	110bp	60°C
FOXE1 (NM_004473)	GCTGGTTTTCCCTGTCTCTG	AGATGGGGGAGACTGAAGGT	100bp	60°C
TTF1 (NM_003317)	ACCAGGACACCATGAGGAAC	GCTCATGTTCATGCCGCT	116bp	60°C
ß-actin (NM_001101)	GCACAGAGCCTCGCCTT	GTTGTCGACGACGAGCG	93bp	60°C

Figure legends

Figure S1. Induction of *PAX8*, *FOXE1*, and *TTF1* expression in thyroid cancer cells using various inhibitors, individually or in different combinations. **A**) Eleven thyroid cancer cells were treated with inhibitors RDEA119, CCI779 (temsirolimus) and SAHA, individually or in different combinations, to induce the expression of thyroid-specific transcription factors, including *PAX8*, *FOXE1*, and *TTF1*. **B**) Effect of TSH stimulation on *PAX8*, *FOXE1*, and *TTF1*expression induced by inhibitors RDEA119, CCI779 (temsirolimus), perifosine and SAHA, individually or in different combinations. Details are described in the Materials and Methods. Data are presented as the mean of values from three assays for the ratios of specific gene expression presented as Natural Log, with red showing the highest expression, followed by black, and green as the lowest. Green asterisks show that the basal level of gene expression is low. R, RDEA119; C, CCI779 (temsirolimus); P, Perifosine; S, SAHA.

Figure S2. Effects of the Akt inhibitor perifosine on thyroid cancer cells. Various thyroid cancer cell lines, as indicated, were treated with 5 μM perifosine for 30 h. The phosphorylation level of Akt was detected by Western blotting using a specific anti-phosphorylated Akt (p-Akt) antibody. Immunoblotting with an antibody against β-actin was used for quality control.

Figure S3. TSH enhancement of *Tg*, *PAX8*, *FOXE1*, and *TTF1* expression induced by various inhibitors, individually or in different combinations, in thyroid cancer cells. Various thyroid cancer cells were treated with inhibitors RDEA119, CCI779 (temsirolimus), perifosine and SAHA as described in Materials and Methods, individually or in different combinations, supplementing without or with 20 mU/ml TSH. Data are presented as the mean ± SD of values from three assays. R, RDEA119; C, CCI779 (temsirolimus); P, Perifosine; S, SAHA. *, $P < 0.05$; **, $P < 0.01$ for comparison with control.

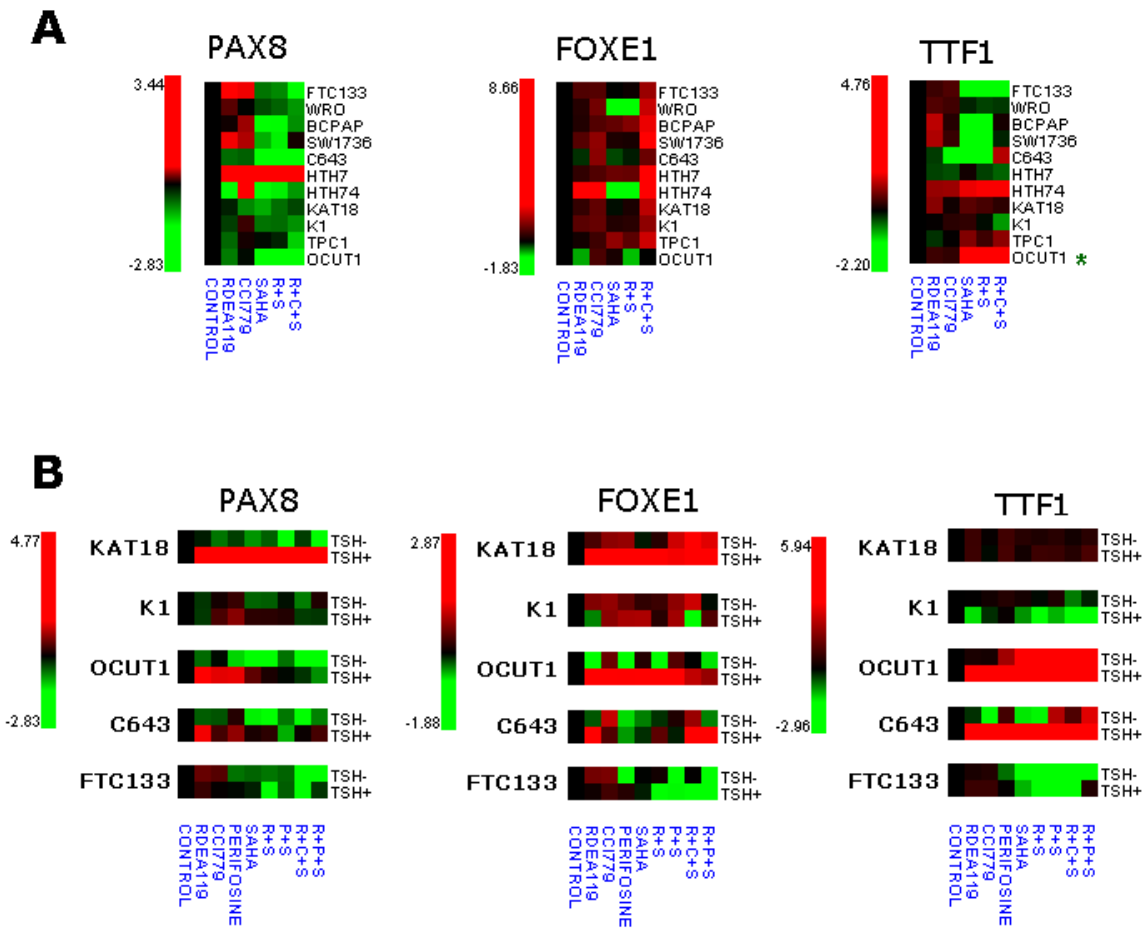

A
PAX8
3.44
-2.83
FTC133
WRO
BCPAP
SW1736
C643
HTH7
HTH74
KAT18
K1
TPC1
OCUT1
CONTROL
RDEA119
CCI779
SAHA
R+S
R+C+S
FOXE1
8.66
-1.83
TTF1
4.76
-2.20
*
B
PAX8
4.77
-2.83
KAT18
K1
OCUT1
C643
FTC133
TSH-
TSH+
FOXE1
2.87
-1.88
TTF1
5.94
-2.96
CONTROL
RDEA119
CCI779
PERIFOSINE
SAHA
R+S
P+S
R+C+S
R+P+S

Figure S1

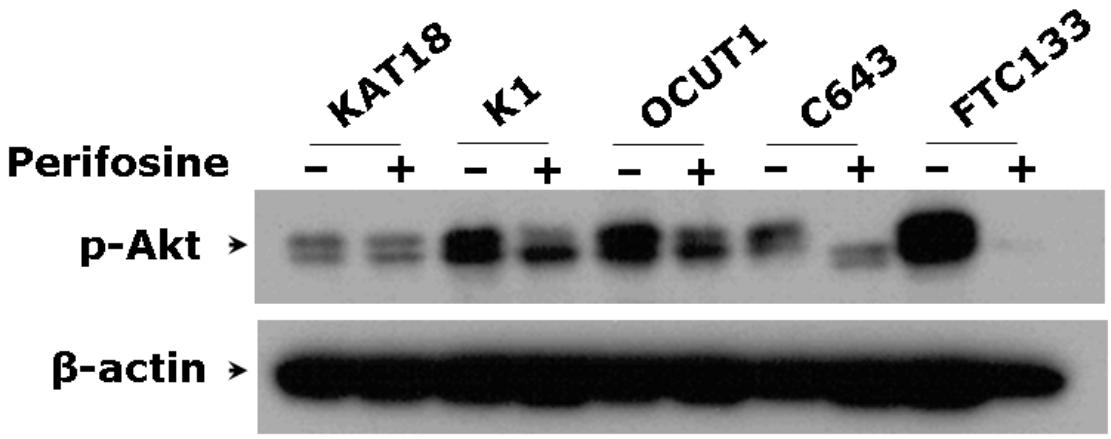


Figure S2

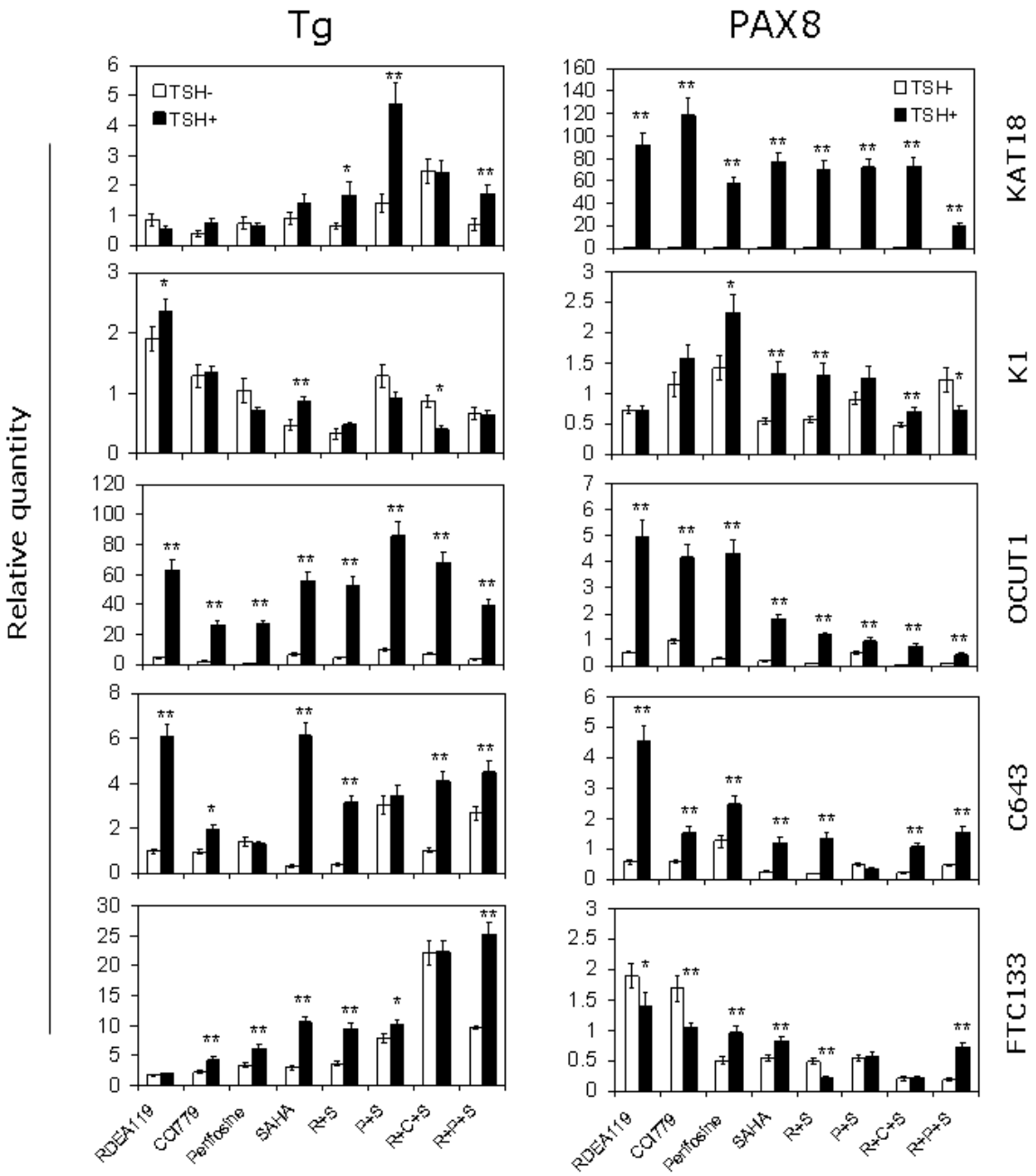
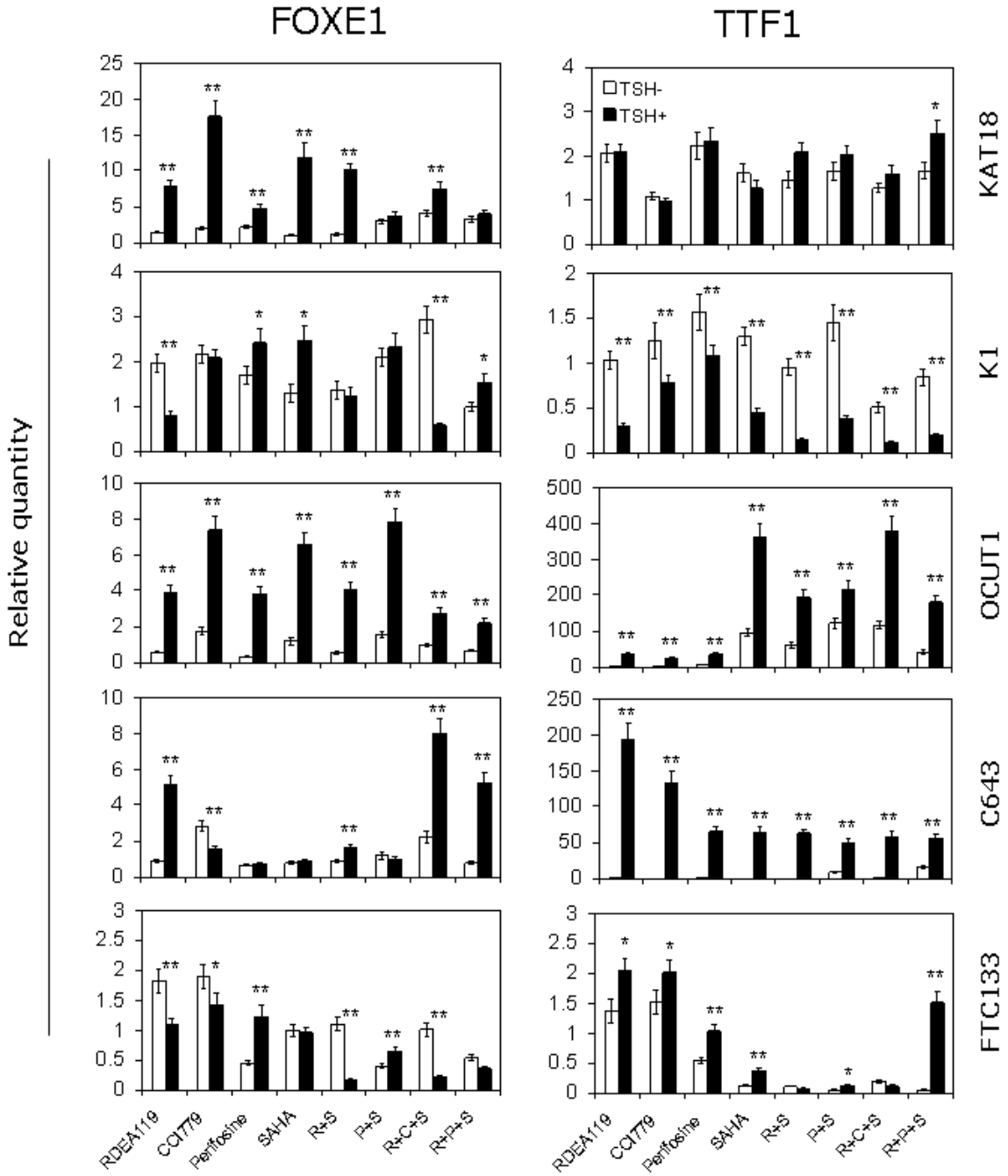

Tg
PAX8
TSH-
TSH+
Relative quantity
KAT18
K1
OCUT1
C643
FTC133
RDEA119
CO1779
Perifosine
SAHA
R+S
P+S
R+C+S
R+P+S

Figure S3

(Continued)


FOXE1
TTF1
TSH-
TSH+
Relative quantity
KAT18
K1
OCUT1
C643
FTC133
RDEA119
CCI779
Perifosine
SAHA
R+S
P+S
R+C+S
R+P+S