

Multiple cancer pathways regulate telomere protection

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APPENDIX

- **Appendix figure legends**
- ***Appendix Figure S1:* Effect of TRF1 inhibitory compounds on RAP1 and TIN2 protein levels.**
- ***Appendix Figure S2.:*TRF1 inhibitors induce telomere induced foci in lung cancer cells.**
- ***Appendix Figure S3:* Novel TRF1 inhibitors can act independently of the PI3K-AKT pathway.**
- ***Appendix Figure S4:* Cell numbers upon transduction with eGFP-*Trf1* WT or the indicated mutant alleles at day 8**
- ***Appendix Figure S5:* *In vivo* combinatorial studies with the new TRF1 inhibitory compounds using patient-derived GBM xenograft models**
- ***Appendix Table S1:* Chemical compounds tested in a primary screening for identifying novel compounds with the ability to downregulate TRF1 protein levels**
- **Appendix Table S2: P-values**

Appendix Figure S1. Effect of TRF1 inhibitory compounds on RAP1 and TIN2 protein levels. **(A)** TIN2 Western blot images (left) and quantification of TIN2 protein levels (right) of CHA9-3 lung cancer cells treated with the indicated compounds for 24h at 1 μ M. Data are representative of n=8 (DMSO) and n=4 (HSP90i, Aurorai, Docetaxel, PLKi, CDKi, mTORi, Gemcitabine, MEKi, ERKi, RTKi) independent experiments. **(B)** RAP1 Western blot images (left) and quantification of RAP1 protein levels (right) of CHA9-3 lung cancer cells treated with the indicated compounds for 24h at 1 μ M. Data are representative of n=8 (DMSO) and n=4 (HSP90i, Aurorai, Docetaxel, PLKi, CDKi, mTORi, Gemcitabine, MEKi, ERKi, RTKi) independent experiments. Data are represented as mean $\pm$ SEM. Significant differences using unpaired *t*-test are indicated by **P* < 0.05, ***P* < 0.01, ****P* < 0.001.

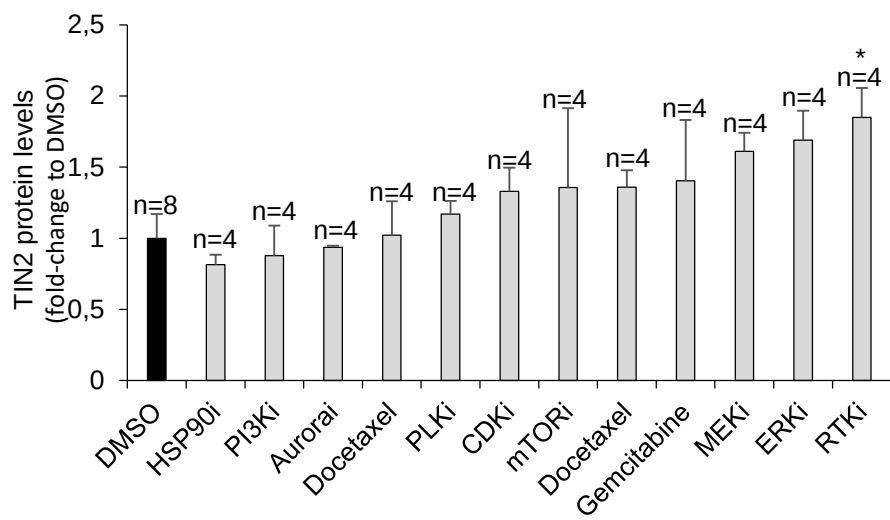
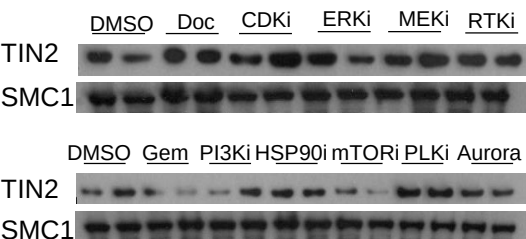
Appendix Figure S2. TRF1 inhibitors induce telomere induced foci in lung cancer cells. (A) Quantification of γ H2AX nuclear intensity upon treatment of CHA9-3 lung cancer cells with the indicated compounds. Data are representative of n=6 (DMSO) and n=3 (PI3Ki, mTORi, RTKi, ERKi, MEKi, HSP90i, Docetaxel, CDKi) independent experiments. **(B)** Quantification of mean number of telomere induced foci (TIF) per nucleus in CHA9-3 lung cancer cells treated with the indicated compounds for 24h at 1 μ M. Data are representative of n=3 independent experiments. **(C)** Mean number of TIFs per nucleus in CHA9.3 lung cancer cells normalized to γ H2AX nuclear intensity. Data are representative of n=3 independent experiments. Data are represented as mean $\pm$ SEM. Significant differences using unpaired *t*-test are indicated by **P* < 0.05, ***P* < 0.01, ****P* < 0.001.

Appendix Figure S3. Novel TRF1 inhibitors can act independently of the PI3K-AKT pathway. (A) Western blot images (up) and p-AKT/AKT protein levels (down) of h676 GSCs treated with the indicated compounds for 24h at 1 μ M. Data are representative of n=2 independent experiments. Data are represented as mean $\pm$ SEM. Significant differences using unpaired *t*-test are indicated by **P* < 0.05, ***P* < 0.01.

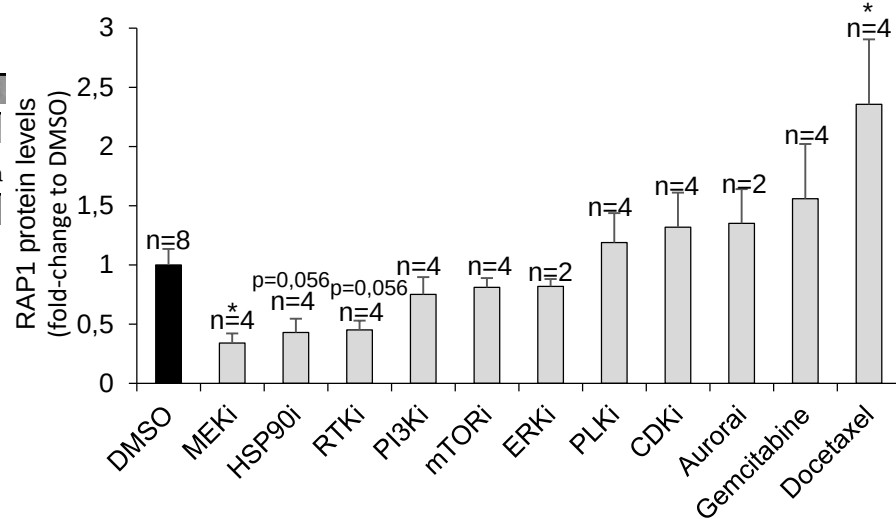
Appendix Figure S4. Cell numbers upon transduction with eGFP-*Trf1* WT or the indicated mutant alleles at day 8. Statistical significance is indicated in the table (*) between the different conditions (down). Data are representative of n=5 independent experiments. Data are represented as mean $\pm$ SEM. Significant differences using ANOVA test Bonferroni are indicated by **P* < 0.05, ***P* < 0.01.

Appendix Figure S5. *In vivo* combinatorial studies with the new TRF1 inhibitory compounds using patient-derived GBM xenograft models. (A) Quantification of the percentage of mice died during treatment with the indicated compounds as single agents or in combination. n represents number of mice: vehicle n=8; PI3K n=17; MEKi, Docetaxel, Gemcitabine, RTKi, ERKi and their combinations with PI3Ki n=4; HSP90i and the combination with PI3K n=2. Significant differences using Chi-Square are indicated by $*P < 0.05$, $**P < 0.01$, $***P < 0.001$. **(B-C)** Longitudinal tumor growth follow-up in mice injected with h676 GSCs and treated with the indicated compounds as single agents or combination. Data are represented as mean $\pm$ SEM. n represents number of tumors: in B: vehicle n=16, PI3Ki n=16, RTKi n=8, Combination n=8; in C: vehicle n=16, PI3Ki n=16, HSP90i n=4, Combination n=4. Significant differences using unpaired *t*-test are indicated by $*P < 0.05$, $**P < 0.01$, $***P < 0.001$.

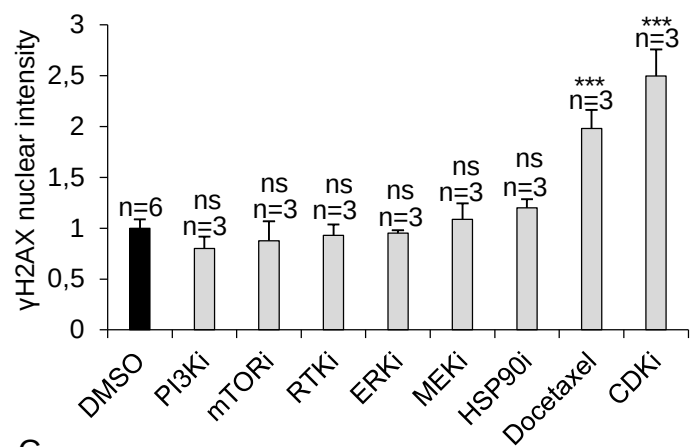
A



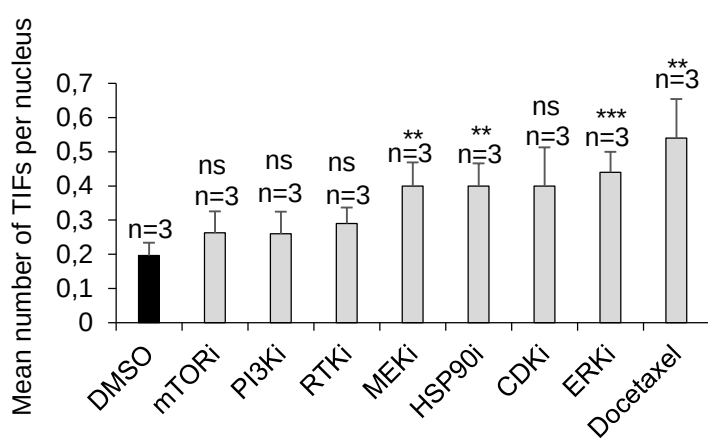
B



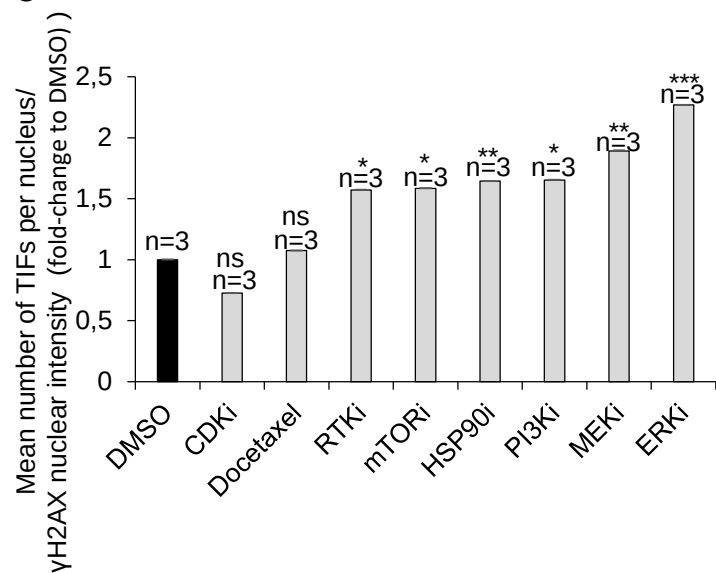
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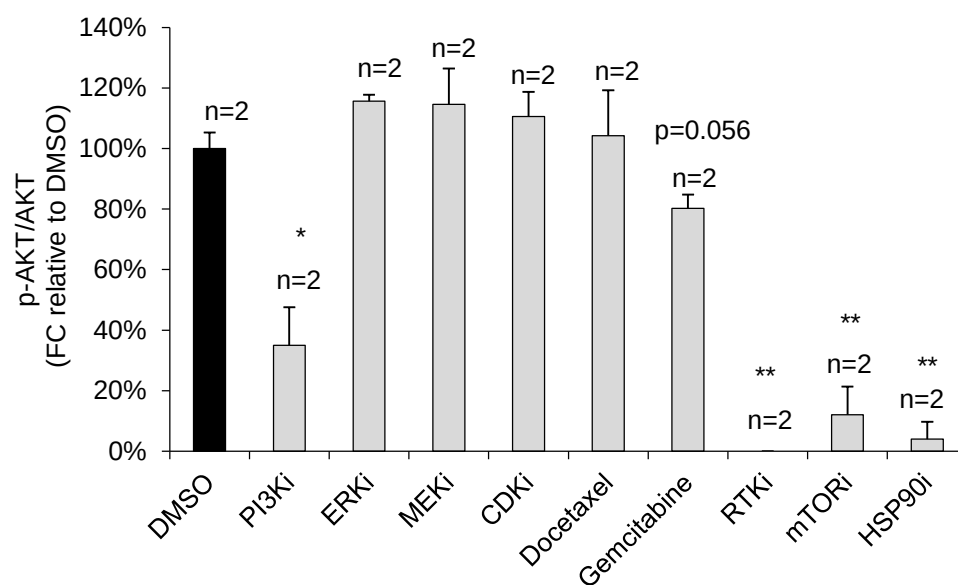
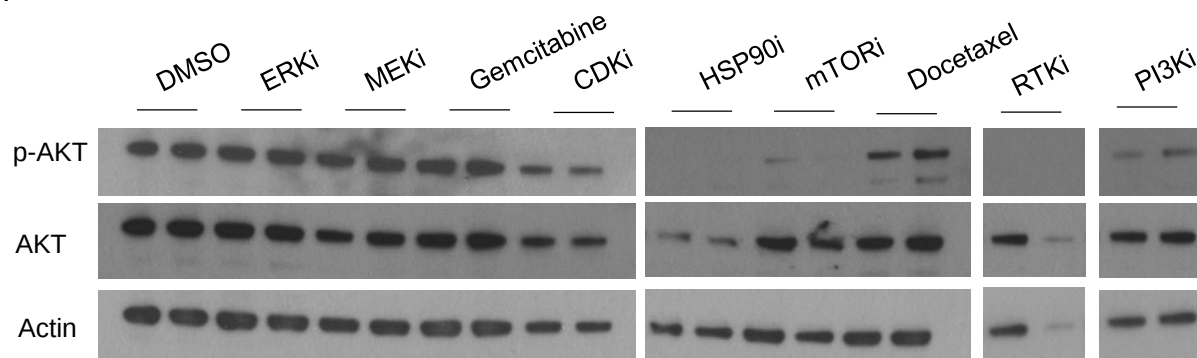
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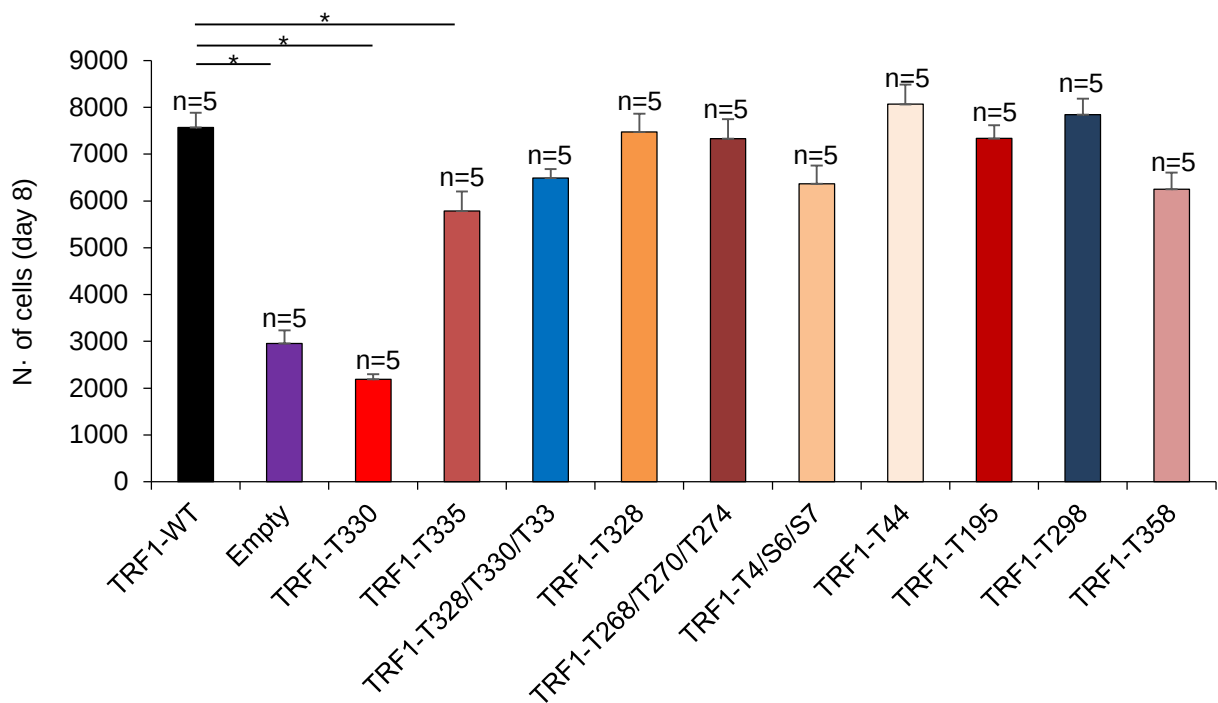


C



A





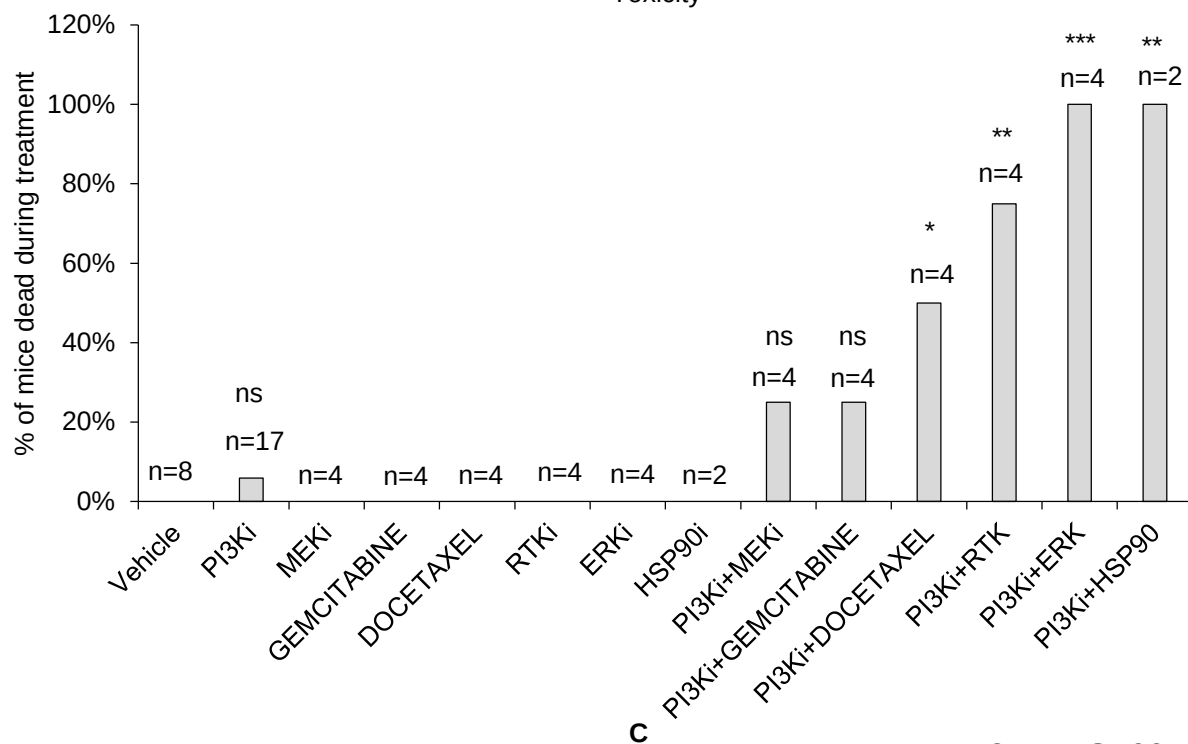
ANOVA test - Bonferroni

	Cre	WT	T44	T195	T298	T328	T330	T335	T4/S6/S7	T358	T268/T270/T274	T328/T330/T33
Cre		*	*	*	*	*		*	*	*	*	*
WT	*						*	*				
T44	*						*	*		*		
T195	*						*					
T298	*						*	*				
T328	*						*					
T330		*	*	*	*	*		*	*	*	*	*
T335	*	*	*		*		*					
T4/S6/S7	*						*					
T358	*		*				*					
T268/T270/T274	*						*					
T328/T330/T33	*						*					

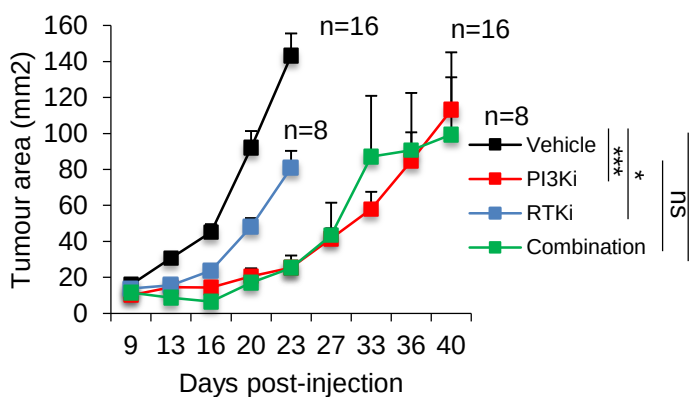
*The mean difference is significance at the 0,05 level

A

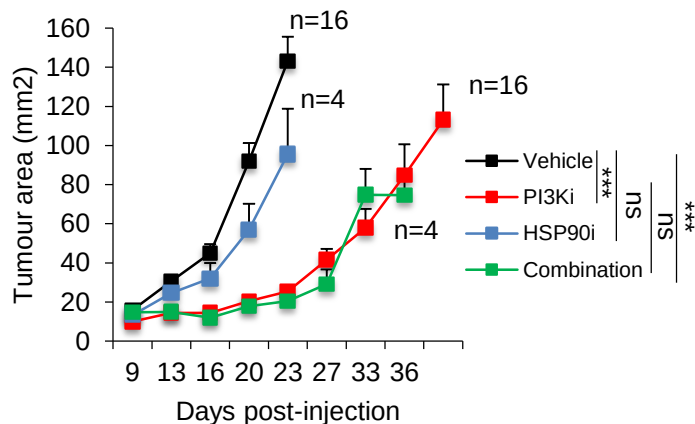
Toxicity

**B**

PI3K + RTK

**C**

PI3K + HSP90



Appendix table S1. Chemical compounds tested in a primary screening for identify novel compounds with the ability to downregulate TRF1 protein levels.
A cut-off above 20% inhibition of TRF1 intensity was established to consider a drug as a hit. Color code indicate those drugs that inhibit the same target. A retest in one representative of each class was done to confirm the hits and consider true hits those with an inhibition or TRF1 levels above 20%.

drug_name	Inhibition sum_int2_area Vs DMSO (%)	sd	Retest by confocal	Inhibition sum_int2_area Vs DMSO (%) in validation assay
GEDATOLISIB	41,765	0,54		
BEZ-235	40,515	6,16		
GSK2126458	39,43	4,62		
CNIO-PI3K-3	40,785	2,67		
GDC-0941	39,5	0,03		
ZSTK474	37,77	0,30	Yes	38,9
BKM120	35,69	2,47		
PI3K-CNIO-1	32,975	3,75		
CNIO-PI3K-2	25	7,07		
KU-0063794	35,24	0,96		
RAPAMYCIN	28,275	5,10		
MK-2206	29,585	0,15		
GSK461364	38,135	0,64	Yes	26,38
SELU METINIB	34,105	0,33	Yes	21,55
TRAMETINIB	32,615	0,80		
PD-0325901	28,07	0,35		
SCH772984	38,89	0,37	Yes	35,58
DASATINIB	32,895	2,27	yes	42,85
TAXOTERE	32,03	1,53	Yes	26,94
VINCRI STINE	23,925	1,61		
PACLITAXEL	27,375	0,04		
CNIO-ATR-1	30,445	1,36	Yes	<5
AZ20	22,99	1,94		
GELDANAMYCIN	30,255	0,05	Yes	45,43
PANO BINOSTAT	23,745	0,19	Yes	<5
5-Fluoracil	21,965	1,03	Yes	<5
ALISERTIB	21,57	2,94	Yes	26,11
VX-680	21,395	2,26		
GEMCITABINE	21,115	1,80	Yes	38,03
CRIZOTINIB	20,125	2,37	Yes	15,29
FLAVOPIRIDOL	46,66	0,38	Yes	86,34
DOVITINIB	19,915	0,86		
BYL-719	19,735	11,29		
CUDC-907	18,39	1,85		
Mitomycin C	17,85	0,27		
SAHA	17,74	1,88		
CUDC-101	17,635	3,10		
PERIFOSINE	16,84	1,57		
SNS-314 MESYLATE	16,54	1,05		
Ixazomib	14,805	1,96		
KU-57788	14,165	4,41		
DABRAFENIB	13,73	3,45		
CARFILZOMIB	13,31	3,35		
PEMETREXED	13,255	2,13		
ETOPOSIDE	12,99	0,92		
GDC-0994	12,24	2,46		
BORTEZOMIB	11,06	1,51		
TX-1123	10,91	5,61		
ABIRATERONE	8,79	5,43		
AT7519	8,61	2,73		
AICAR	7,79	10,49		
TEMOZOLOMIDE	7,05	13,31		
SN-38	5,845	0,23		
OLAPARIB	5,075	5,35		
ELESCLOMOL	5,02	3,44		
BARDOXOLONE METHYL	< 5	2,75		
BAY 61-3606	< 5	4,78		
BAY 87-2243	< 5	9,50		
BIRB 796	< 5	6,52		
CISPLATINUM	< 5	4,21		
CNIO-PIM-1	< 5	6,92		
CYCLOPHOSPHAMIDE	< 5	2,57		
Dibenzazepine	< 5	2,81		

Appendix table S1. Chemical compounds tested in a primary screening for identify novel compounds with the ability to downregulate TRF1 protein levels.

A cut-off above 20% inhibition of TRF1 intensity was established to consider a drug as a hit. Color code indicate those drugs that inhibit the same target. A retest in one representative of each class was done to confirm the hits and consider true hits those with an inhibition or TRF1 levels above 20%.

drug_name	Inhibition sum_int2_area Vs DMSO (%)	sd	Retest by confocal	Inhibition sum_int2_area Vs DMSO (%) in validation assay
EFLORNITHINE	< 5	11,05		
ERLOTINIB	< 5	55,30		
EX-527	< 5	6,77		
Fulvestrant	< 5	6,12		
Galunisertib	< 5	1,53		
GEFITINIB	< 5	7,88		
GENISTEIN	< 5	26,00		
GLEEVEC, IMATINIB	< 5	10,94		
IRINOTECAN	< 5	2,25		
KETOCONAZOLE	< 5	0,11		
LAPATINIB	< 5	3,42		
LETROZOLE	< 5	0,83		
LINIFANIB	< 5	2,57		
LOMUSTINE	< 5	25,63		
LY2801653	< 5	29,07		
METFORMIN	< 5	9,60		
Mifepristone	< 5	7,45		
MK-906	< 5	10,44		
OSI-906	< 5	10,97		
OXALIPLATIN	< 5	44,07		
Palbociclib	< 5	10,76		
Pazopanib	< 5	47,46		
4-PB (SODIUM 4-PHENYLBUTYRATE)	< 5	2,45		
PF 4708671	< 5	27,01		
PFK15	< 5	5,18		
PX-478	< 5	3,81		
QUIZARTINIB	< 5	0,68		
Roscovitine	< 5	7,42		
SB 203580	< 5	19,71		
SB 505124	< 5	10,03		
SEMAGACESTAT	< 5	50,28		
Silmitasertib	< 5	18,57		
SORAFENIB	< 5	40,46		
S-RUXOLITINIB	< 5	1,93		
Suramin	< 5	5,13		
Tamoxifen	< 5	3,52		
TANZISERTIB	< 5	27,20		
Tempol	< 5	33,45		
VALPROIC ACID	< 5	9,46		
Vemurafenib	< 5	25,14		
VISMODEGIB	< 5	15,05		
Zileuton	< 5	18,31		
AZD5363	< 5	18,07		
CAL-101	< 5	16,74		
DISULFIRAM	< 5	0,74		
GSK2636771	< 5	10,34		
PILARALISIB	< 5	18,58		
TGX-221	< 5	24,34		
NVP-BGJ398	> 5	11,62		
Ricolinostat	> 5	0,05		

Appendix Table S2: P Values

FIGURE	SAMPLE	P VALUE
Figure 1C		
DMSO	Aurorai	0,0503
	HSP90i	0,0041
	mTORi	0,0054
	Docetaxel	0,0116
	ERKi	1,77E-05
	MEKi	2,08E-05
	Gemcitabine	0,0007
	CDKi	0,0021
	RTKi	6,50E-05
	PLKi	0,1330
Figure 2A		
DMSO	ERKi	0,0014
	MEKi	0,0169
	RTKi	0,0420
	HSP90i	0,0004
	CDKi	0,0054
	mTORi	0,2388
	PI3Ki	0,0118
	Docetaxel	0,0045
Figure 2B	Sample	PVALUE
DMSO	mTORi	0,0371
	Docetaxel	0,0207
	ERKi	0,0371
	MEKi	0,0199
	RTKi	0,0133
	HSP90i	0,0028
	Gemcitabine	0,0079
	CDKi	0,0003
Figure 2C		
DMSO	mTORi	2,87E-05
	MEKi	0,0033
	RTKi	1,58E-08
Figure 5C	Sample	PVALUE
TRF1-WT	T44	0,4232
	T195	0,9891
	T298	0,2465
	T358	0,7300
	T4 S6 S7	0,1457
	T268 T270 T274	0,5850
	T328 T330 T335	3,68E-05
	T328	0,2960
	T330	0,0600
	T335	0,1576
Figure 5F		
TRF1-WT	T328 T330 T335	0,053
	T328	0,2960
	T330	0,0600
	T335	0,1576

Appendix Table
S2: P Values

Figure 4J	SAMPLE	P VALUE
	T4	0,04
	T298	0,12
	T330	0,02
	T336	0,07
Figure 4M	T4S6S7	0,00001
	T44	0,02
	T195	0,1
	T268T270T274	0,0005
	T298	0,87
	T328T330T335	0,005
	T358	0,17
Figure 4N	T328	0,00001
	T335	0,00001
Figure 4O	T248	0,18
	T330	0,04
	S344	0,003
	T330S344	0,0007
5G	WT+ERKi	0.04
	T328+ERKi	0.045
	T330+ERKi	0.03
	T335+ERKi	0.026
	T328/T330/T335 +ERKi	0.032
5H	Scrb1+ERKi	0.03
	WT+ERKi	0.015
	T328+ERKi	0.024
	T330+ERKi	0.036
	T335+ERKi	0.041
	T328/T330/T335 +ERKi	0.039
6A	Scrb1	0.008
	shERK	0.03
	Scrb1	0.004
	shERK	0.026
6B	ERK1/2 RNAi	0.03
	ERK1/2 RNAi	0.021
6E	shERK	0.03
	shERK TRF1WT	0.034
	shERK T328	0.042
	shERK T330	0.021
	shERK T335	0.03
	shERK T328/T330/T335	0.027

Appendix Table

S2: P Values

Figure 7A	SAMPLE	P VALUE
DMSO	PI3Ki	0,0001
	RTKi	7,34E-05
	Combination	2,09E-09
Combination	PI3Ki	5,50E-09
	RTKi	2,03E-05
Figure 7B		
DMSO	PI3Ki	0,0022
	ERKi	0,1178
	Combination	3,89E-07
Combination	PI3Ki	2,03E-07
	ERKi	1,58E-07
Figure 7C		
DMSO	PI3Ki	0,0011
	MEKi	0,0081
	Combination	1,26E-09
Combination	PI3Ki	3,80E-06
	MEKi	2,40E-06
Figure 7D		
DMSO	PI3Ki	0,0190
	HSP90i	0,0002
	Combination	1,28E-06
Combination	PI3Ki	1,46E-07
	HSP90i	0,0028
Figure 7E		
DMSO	PI3Ki	0,0018
	Gemcitabine	1,30E-05
	Combination	1,28E-07
Combination	PI3Ki	1,32E-09
	Gemcitabine	3,65E-05
Figure 7F		
DMSO	PI3Ki	0,0064
	Docetaxel	1,63E-05
	Combination	1,00E-06
Combination	PI3Ki	7,14E-06
	Docetaxel	0,0034

Appendix Table

S2: P Values

Figure 7G	SAMPLE	P VALUE
DMSO	PI3Ki	0,0083
	RTKi	1,64E-05
	Combination	1,38E-06
Combination	PI3Ki	0,0052
	RTKi	0,0075
Figure 7H		
DMSO	PI3Ki	0,0083
	ERKi	0,0195
	Combination	0,0080
Combination	PI3Ki	0,2902
	ERKi	0,2932
Figure 7I		
DMSO	PI3Ki	0,0083
	MEKi	0,0009
	Combination	8,35E-06
Combination	PI3Ki	0,0047
	MEKi	0,2955
Figure 7J		
DMSO	PI3Ki	0,0083
	HSP90i	0,0048
	Combination	2,03E-05
Combination	PI3Ki	0,0879
	HSP90i	0,6510
Figure 7K		
DMSO	PI3Ki	0,0083
	Gemcitabine	3,17E-05
	Combination	6,83E-07
Combination	PI3Ki	0,0751
	Gemcitabine	0,0048
Figure 7L		
DMSO	PI3Ki	0,0083
	Docetaxel	0,1271
	Combination	0,0035
Combination	PI3Ki	0,5163
	Docetaxel	0,7246

Appendix Table

S2: P Values

Figure 8A		
Vehicle	PI3Ki	1,22E-05
	ERKi	0,0210
	Combination	0,0120
Combination	PI3Ki	0,0507
Figure 8B		
Vehicle	PI3Ki	1,22E-05
	MEKi	0,0131
	Combination	0,0091
Combination	PI3Ki	0,0078
Figure 8C		
Vehicle	PI3Ki	1,22E-05
	Docetaxel	0,2138
	Combination	0,0037
Combination	PI3Ki	0,0390
Figure 8D		
Vehicle	PI3Ki	1,22E-05
	Gemcitabine	0,0292
	Combination	0,0013
Combination	PI3Ki	0,0570
Figure 8E		
Vehicle	PI3Ki	0,4186
	ERKi	0,1623
	Combination	0,0067
PI3Ki	ERKi	0,1812
Combination	PI3Ki	0,0780
	ERKi	0,0546
Figure 8F		
Vehicle	PI3Ki	0,4186
	MEKi	0,2206
	Combination	0,0069
PI3Ki	MEKi	0,1817
Combination	PI3Ki	0,0803
	MEKi	0,2365
Figure 8G		
Vehicle	PI3Ki	0,4186
	Docetaxel	0,0092
	Combination	0,0049
PI3Ki	Docetaxel	0,0868
Combination	PI3Ki	0,0732
	Docetaxel	0,1681
Figure 8H		
Vehicle	PI3Ki	0,4186
	Gemcitabine	0,7881
	Combination	0,9588
PI3Ki	Gemcitabine	0,5674
Combination	PI3Ki	0,4797
	Gemcitabine	0,8582

Appendix Table

S2: P Values

Appendix figure S1A	SAMPLE	PVALUE
DMSO	Docetaxel	0,3239
	CDKi	0,3785
	ERKi	0,0961
	MEKi	0,1092
	RTKi	0,0478
	Gemcitabine	0,4392
	PI3Ki	0,7605
	HSP90i	0,5911
	mTORi	0,5623
	PLKi	0,6030
	Aurora	0,8530
Appendix figure S1B		
DMSO	Docetaxel	0,0276
	CDKi	0,3845
	ERKi	0,5080
	MEKi	0,0280
	RTKi	0,0567
	Gemcitabine	0,2514
	PI3Ki	0,3827
	HSP90i	0,0566
	mTORi	0,5044
	PLKi	0,6370
	Aurora	0,3922
Appendix figure S2A		
DMSO	PI3Ki	0,2223
	CDKi	0,0002
	Docetaxel	0,0008
	ERKi	0,72
	HSP90i	0,1974
	MEKi	0,6095
	mTORi	0,5145
	RTKi	0,6513
Appendix figure S2B		
DMSO	ERKi	0,0003
	MEKi	0,0045
	RTKi	0,1113
	HSP90i	0,0044
	CDKi	0,1614
	PI3Ki	0,3842
	Docetaxel	0,0026
	mTORi	0,3270

Appendix Table
S2: P Values

Appendix figure S2C	SAMPLE	P VALUE
DMSO	ERKi	6,73E-05
	PI3Ki	0,0150
	CDKi	0,1128
	Docetaxel	0,6573
	HSP90i	0,0058
	MEKi	0,0094
	mTORi	0,0410
	RTKi	0,0224
Appendix figure S3A		
DMSO	HSP90i	0,0032
	mTORi	0,0073
	Docetaxel	0,7422
	ERKi	0,0595
	MEKi	0,2516
	Gemcitabine	0,0567
	CDKi	0,2645
	RTKi	0,0014
	PI3Ki	0,0215
Appendix figureS4	P values are represented in the table below	
Appendix figureS5A		
Vehicle	PI3Ki	0,48
	PI3Ki+MEKi	0,33
	PI3Ki+Gemcitabine	0,33
	PI3Ki+Docetaxel	0,01
	PI3Ki+RTKi	0,0047
	PI3Ki+ERKi	0,005
	PI3Ki+HSP90i	0,0016
Expanded View Figure 3C		
For pAKT/AKT: Vehicle	PI3Ki (ETP-47037)	0,8651
For pS6/S6: Vehicle	PI3Ki (ETP-47037)	0,6660
Expanded View Figure 3D		
Vehicle	PI3Ki (ETP-47037)	0,2514
Expanded View Figure 3E		
Vehicle	PI3Ki (ETP-47037)	0,4879
Expanded View Figure 4C		
DMSO	PI3Ki	1,56E-17
	RTKi	5,84E-45
	Combination	4,7E-73
Combination	PI3Ki	1,61E-58
	RTKi	9,74E-21

Appendix Table

S2: P Values

Expanded View Figure 4D		
DMSO	PI3Ki	1,00E-34
	ERKi	7,94E-12
	Combination	1,19E-62
Combination	PI3Ki	3,56E-35
	ERKi	6,86E-50
Expanded View Figure 4E		
DMSO	PI3Ki	0,0003
	MEKi	0,8340
	Combination	4,90E-23
Combination	PI3Ki	7,17E-17
	MEKi	5,23E-25
Expanded View Figure 4F		
DMSO	PI3Ki	2,35E-20
	HSP90i	1,79E-32
	Combination	3,86E-35
Combination	PI3Ki	1,17E-18
	HSP90i	0,0005
Expanded View Figure 4G		
DMSO	PI3Ki	5,45E-07
	Gemcitabine	1,06E-21
	Combination	3,11E-22
Combination	PI3Ki	3,74E-17
	Gemcitabine	0,0009
Expanded View Figure 4H		
DMSO	PI3Ki	1,21E-10
	Docetaxel	1,97E-19
	Combination	1,94E-19
Combination	PI3Ki	1,39E-18
	Docetaxel	0,0002

Appendix Table
S2: P Values

Appendix figure S4								
I) type		(J) type		Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
							Lower Bound	Upper Bound
	0		1	-4612.200*	477.548	.000	-6330.19	-2894.21
			44	-5108.000*	477.548	.000	-6825.99	-3390.01
			195	-4376.800*	477.548	.000	-6094.79	-2658.81
			298	-4885.400*	477.548	.000	-6603.39	-3167.41
			328	-4517.200*	477.548	.000	-6235.19	-2799.21
			330	766.600	477.548	1.000	-951.39	2484.59
			335	-2829.400*	477.548	.000	-4547.39	-1111.41
			467	-3408.400*	477.548	.000	-5126.39	-1690.41
			358	-3292.400*	477.548	.000	-5010.39	-1574.41
			3268	-4375.400*	477.548	.000	-6093.39	-2657.41
			3328	-3533.800*	477.548	.000	-5251.79	-1815.81
	1		0	4612.200*	477.548	.000	2894.21	6330.19
			44	-495.800	477.548	1.000	-2213.79	1222.19
			195	235.400	477.548	1.000	-1482.59	1953.39
			298	-273.200	477.548	1.000	-1991.19	1444.79
			328	95.000	477.548	1.000	-1622.99	1812.99
			330	5378.800*	477.548	.000	3660.81	7096.79
			335	1782.800*	477.548	.033	64.81	3500.79
			467	1203.800	477.548	.996	-514.19	2921.79
			358	1319.800	477.548	.533	-398.19	3037.79
			3268	236.800	477.548	1.000	-1481.19	1954.79
			3328	1078.400	477.548	1.000	-639.59	2796.39
	44		0	5108.000*	477.548	.000	3390.01	6825.99
			1	495.800	477.548	1.000	-1222.19	2213.79
			195	731.200	477.548	1.000	-986.79	2449.19
			298	222.600	477.548	1.000	-1495.39	1940.59
			328	590.800	477.548	1.000	-1127.19	2308.79
			330	5874.600*	477.548	.000	4156.61	7592.59
			335	2278.600*	477.548	.001	560.61	3996.59
			467	1699.600	477.548	.056	-18.39	3417.59
			358	1815.600*	477.548	.027	97.61	3533.59
			3268	732.600	477.548	1.000	-985.39	2450.59
			3328	1574.200	477.548	.122	-143.79	3292.19
	195		0	4376.800*	477.548	.000	2658.81	6094.79
			1	-235.400	477.548	1.000	-1953.39	1482.59
			44	-731.200	477.548	1.000	-2449.19	986.79
			298	-508.600	477.548	1.000	-2226.59	1209.39
			328	-140.400	477.548	1.000	-1858.39	1577.59
			330	5143.400*	477.548	.000	3425.41	6861.39
			335	1547.400	477.548	.143	-170.59	3265.39
			467	968.400	477.548	1.000	-749.59	2686.39
			358	1084.400	477.548	1.000	-633.59	2802.39
			3268	1.400	477.548	1.000	-1716.59	1719.39
			3328	843.000	477.548	1.000	-874.99	2560.99
	298		0	4885.400*	477.548	.000	3167.41	6603.39
			1	273.200	477.548	1.000	-1444.79	1991.19
			44	-222.600	477.548	1.000	-1940.59	1495.39
			195	508.600	477.548	1.000	-1209.39	2226.59
			328	368.200	477.548	1.000	-1349.79	2086.19
			330	5652.000*	477.548	.000	3934.01	7369.99
			335	2056.000*	477.548	.005	338.01	3773.99
			467	1477.000	477.548	.218	-240.99	3194.99
			358	1593.000	477.548	.109	-124.99	3310.99
			3268	510.000	477.548	1.000	-1207.99	2227.99
			3328	1351.600	477.548	.447	-366.39	3069.59

Appendix Table

S2: P Values

328		0	4517.200*	477.548	.000	2799.21	6235.19
		1	-95.000	477.548	1.000	-1812.99	1622.99
		44	-590.800	477.548	1.000	-2308.79	1127.19
		195	140.400	477.548	1.000	-1577.59	1858.39
		298	-368.200	477.548	1.000	-2086.19	1349.79
		330	5283.800*	477.548	.000	3565.81	7001.79
		335	1687.800	477.548	.060	-30.19	3405.79
		467	1108.800	477.548	1.000	-609.19	2826.79
		358	1224.800	477.548	.891	-493.19	2942.79
		3268	141.800	477.548	1.000	-1576.19	1859.79
		3328	983.400	477.548	1.000	-734.59	2701.39
330		0	-766.600	477.548	1.000	-2484.59	951.39
		1	-5378.800*	477.548	.000	-7096.79	-3660.81
		44	-5874.600*	477.548	.000	-7592.59	-4156.61
		195	-5143.400*	477.548	.000	-6861.39	-3425.41
		298	-5652.000*	477.548	.000	-7369.99	-3934.01
		328	-5283.800*	477.548	.000	-7001.79	-3565.81
		335	-3596.000*	477.548	.000	-5313.99	-1878.01
		467	-4175.000*	477.548	.000	-5892.99	-2457.01
		358	-4059.000*	477.548	.000	-5776.99	-2341.01
		3268	-5142.000*	477.548	.000	-6859.99	-3424.01
		3328	-4300.400*	477.548	.000	-6018.39	-2582.41
335		0	2829.400*	477.548	.000	1111.41	4547.39
		1	-1782.800*	477.548	.033	-3500.79	-64.81
		44	-2278.600*	477.548	.001	-3996.59	-560.61
		195	-1547.400	477.548	.143	-3265.39	170.59
		298	-2056.000*	477.548	.005	-3773.99	-338.01
		328	-1687.800	477.548	.060	-3405.79	30.19
		330	3596.000*	477.548	.000	1878.01	5313.99
		467	-579.000	477.548	1.000	-2296.99	1138.99
		358	-463.000	477.548	1.000	-2180.99	1254.99
		3268	-1546.000	477.548	.145	-3263.99	171.99
		3328	-704.400	477.548	1.000	-2422.39	1013.59
4_6_7		0	3408.400*	477.548	.000	1690.41	5126.39
		1	-1203.800	477.548	.996	-2921.79	514.19
		44	-1699.600	477.548	.056	-3417.59	18.39
		195	-968.400	477.548	1.000	-2686.39	749.59
		298	-1477.000	477.548	.218	-3194.99	240.99
		328	-1108.800	477.548	1.000	-2826.79	609.19
		330	4175.000*	477.548	.000	2457.01	5892.99
		335	579.000	477.548	1.000	-1138.99	2296.99
		358	116.000	477.548	1.000	-1601.99	1833.99
		3268	-967.000	477.548	1.000	-2684.99	750.99
		3328	-125.400	477.548	1.000	-1843.39	1592.59
358		0	3292.400*	477.548	.000	1574.41	5010.39
		1	-1319.800	477.548	.533	-3037.79	398.19
		44	-1815.600*	477.548	.027	-3533.59	-97.61
		195	-1084.400	477.548	1.000	-2802.39	633.59
		298	-1593.000	477.548	.109	-3310.99	124.99
		328	-1224.800	477.548	.891	-2942.79	493.19
		330	4059.000*	477.548	.000	2341.01	5776.99
		335	463.000	477.548	1.000	-1254.99	2180.99
		467	-116.000	477.548	1.000	-1833.99	1601.99
		3268	-1083.000	477.548	1.000	-2800.99	634.99
		3328	-241.400	477.548	1.000	-1959.39	1476.59
3268		0	4375.400*	477.548	.000	2657.41	6093.39
		1	-236.800	477.548	1.000	-1954.79	1481.19
		44	-732.600	477.548	1.000	-2450.59	985.39
		195	-1.400	477.548	1.000	-1719.39	1716.59
		298	-510.000	477.548	1.000	-2227.99	1207.99
		328	-141.800	477.548	1.000	-1859.79	1576.19
		330	5142.000*	477.548	.000	3424.01	6859.99
		335	1546.000	477.548	.145	-171.99	3263.99
		356	967.000	477.548	1.000	-750.99	2684.99
		368	1083.000	477.548	1.000	-634.99	2800.99