9th International Symposium on Targeted Anticancer Therapies

STA-9090 (Ganetespib) and AT13387

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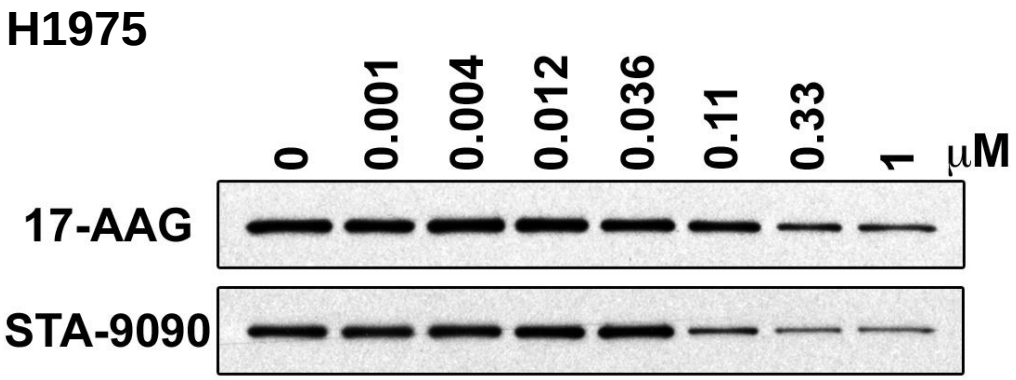
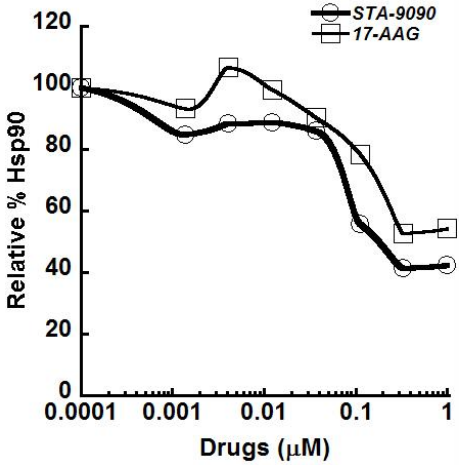
Disclosures

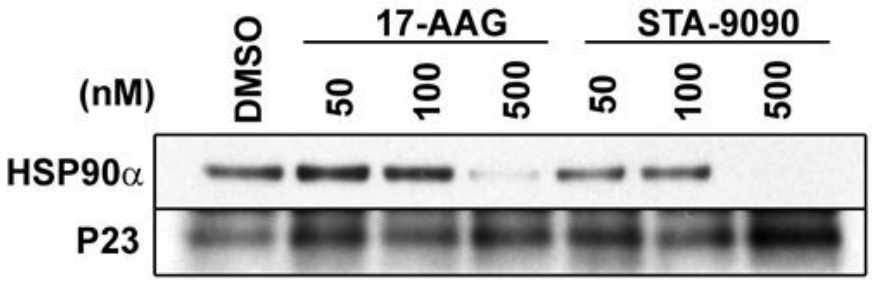
I am an investigator on STA-9090 and AT13387 trials.

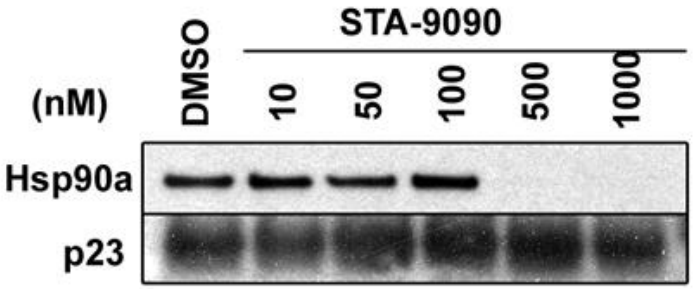
I have no financial relationships to disclose.

STA-9090: Biochemical Assays for Potency of Hsp90 Inhibition









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Non-geldanamycin resorcinol-containing triazole

Comparison of EGFR TKI and HSP90 inhibitor efficacy in Ba/F3 ectopically expressing different EGFR mutations

EGFR Activating Mutations					EGFR Activating Mutations in *cis* with T790M				
EGFR Mutation	Erlotinib IC_{50}	CL-387,785 IC_{50}	17-AAG IC_{50}	STA-9090 IC_{50}	EGFR Mutation	Erlotinib IC_{50}	CL-387,785 IC_{50}	17-AAG IC_{50}	STA-9090 IC_{50}
Del E746_A750	4	1	92	***4***	Del E746_A750/T790M	>10000	294	73	***10***
Del S752_I759	37	<1	180	***12***	Del S752_I759/T790M	>10000	537	88	***7***
Del L747_A750InsP	5	21	67	***4***	Del L747_A750InsP/T790M	>10000	445	38	***4***
Del L747_A753InsS	1	<1	136	***3***	Del L747_A753InsS/T790M	>10000	258	119	***4***
Del E746_S752InsV	25	274	58	***5***	Del E746_S752InsV/T790M	>10000	756	40	***2***
L858R	16	5	246	***7***	L858R/T790M	>10000	950	155	***12***
A767_V769duspASV	>3000	427	2262	***34***					
H773_V774insH	>3000	229	110	***40***					
D770_N771insNPG	>3000	7	12	***1***					

*Units: nmol/L

Takeshi Shimamura, Proc. AACR 2009

Comparative potency of 17-AAG and STA-9090 against NSCLC cell lines

Cell Line	EGFR	ERBB2	KRAS	Others	Erlotinib-Gefitinib	17-AAG IC_{50}*	STA-9090 IC_{50}*
H3255	L858R	Wild-type	Wild-type		Sensitive	58	**22**
HCC827	Del E746_A750	Wild-type	Wild-type		Sensitive	18	**7**
PC9	Del E746_A750	Wild-type	Wild-type		Sensitive	7	**2**
HCC4006	Del L747_E749, A750P	Wild-type	Wild-type		Sensitive	25	**12**
NCI-H1975	L858R/T790M	Wild-type	Wild-type		Resistant	75	**<1**
NCI-H820	Del E746_L751, Ins I/T790M	Wild-type	Wild-type		Resistant	34	**3**
DFCI-LU011	Del L747_E749, A750P	Wild-type	Wild-type		Resistant	111	**2**
NCI-H1650	Del E746_A750	Wild-type	Wild-type		Resistant	7	**7**
NCI-H1781	Wild-type	G776insV_G/C	Wild-type		Resistant	22	**2**
NCI-H1734	Wild-type	Wild-type	G13C		Resistant	96	**12**
A549	Wild-type	Wild-type	G12S		Resistant	75	**22**
NCI-H460	Wild-type	Wild-type	Q61H		Resistant	77	**14**
NCI-H358	Wild-type	Wild-type	G12C		Resistant**	3	**1**
A427	Wild-type	Wild-type	G12D		Resistant	4	**<1**
NCI-H441	Wild-type	Wild-type	G12V		Resistant	111	**26**
NCI-H1299	Wild-type	Wild-type	Wild-type	NRAS (Q61K)	Resistant	36	**6**
NCI-H1666	Wild-type	Wild-type	Wild-type	BRAF (G466V)	Medium	27	**6**
NCI-H1819	Wild-type	Wild-type (Amp)	Wild-type	PIK3CA	Resistant	749	**7**
NCI-H1703	Wild-type	Wild-type	Wild-type	PDGFRA (Amp)	Resistant	3	**3**
NCI-H596	Wild-type	Wild-type	Wild-type	RB Null	Resistant	3,500	**7**
NCI-H522	Wild-type	Wild-type	Wild-type		Resistant	7	**6**
HCC1833	Wild-type	Wild-type	Wild-type		Resistant	4	**<1**
Calu-3	Wild-type (Amp)	Wild-type (Amp)	Wild-type		Resistant**	16	**9**

* Units: nmol/L **Results varies among reports

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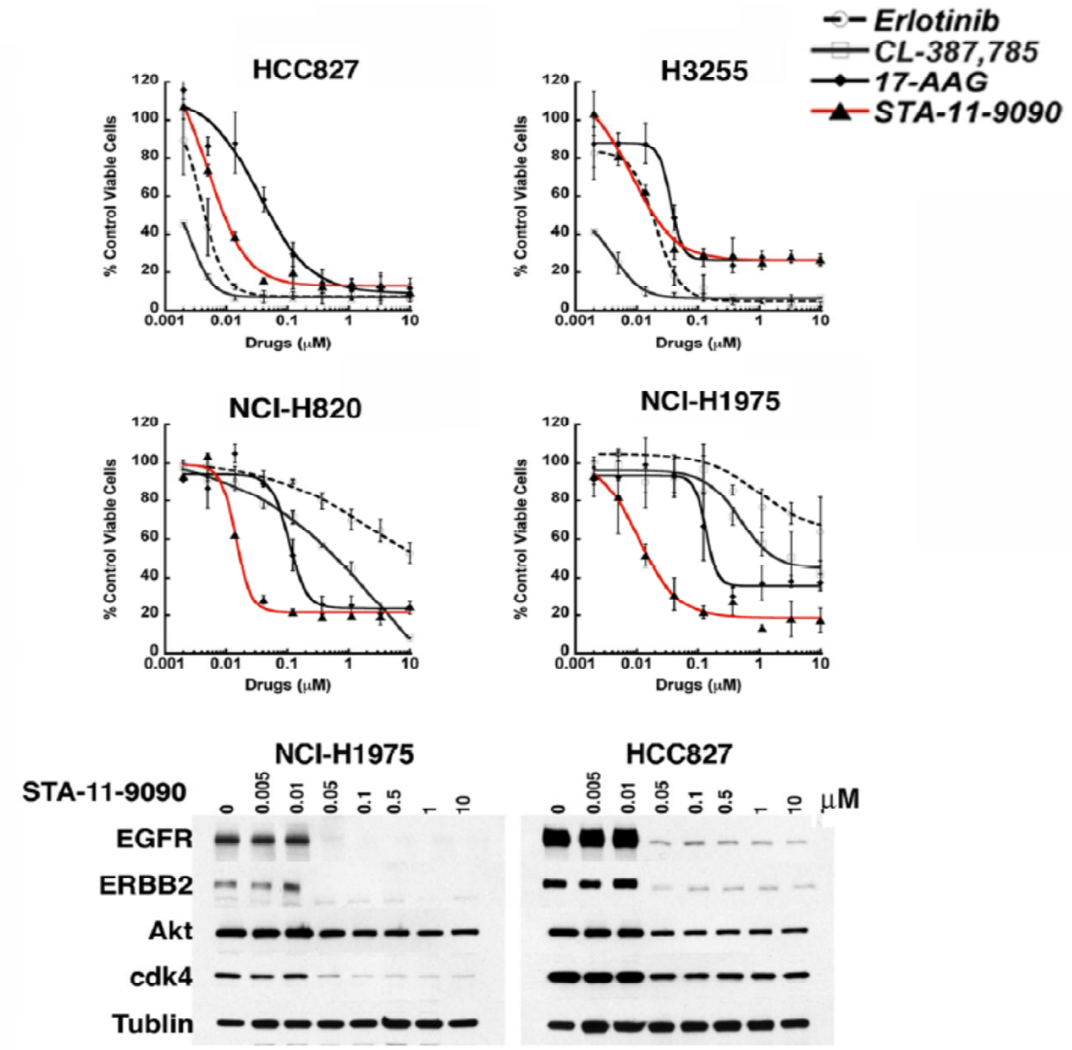
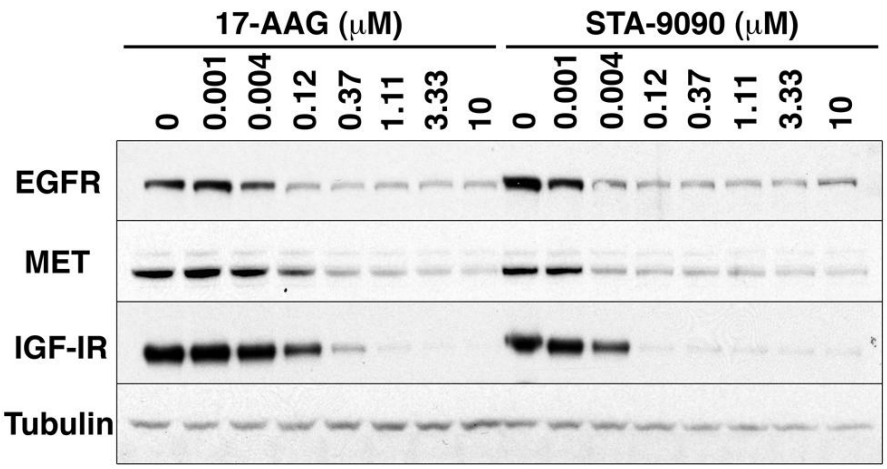
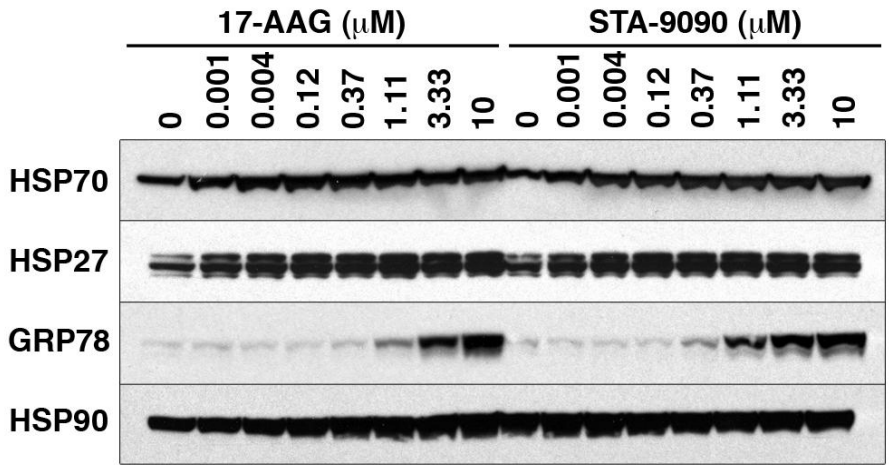
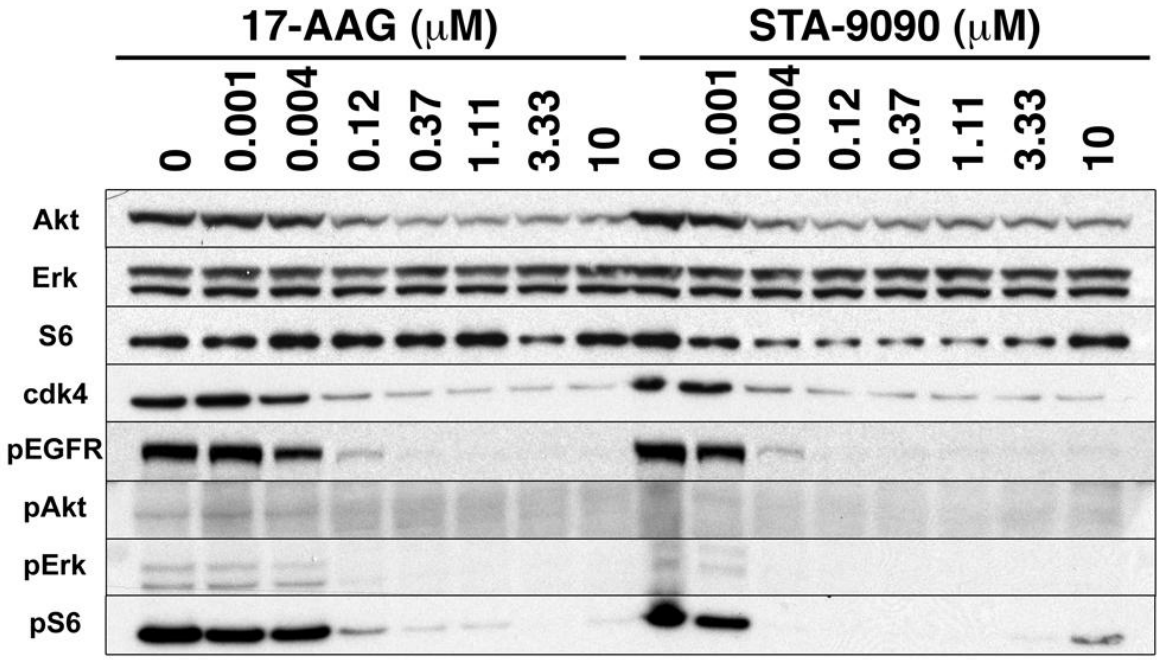
Potency of STA9090 against T790M-expressing NSCLC cell lines



Client Depletion by 17-AAG and STA-9090 in NCI-H1975 cells







NCI-H1975

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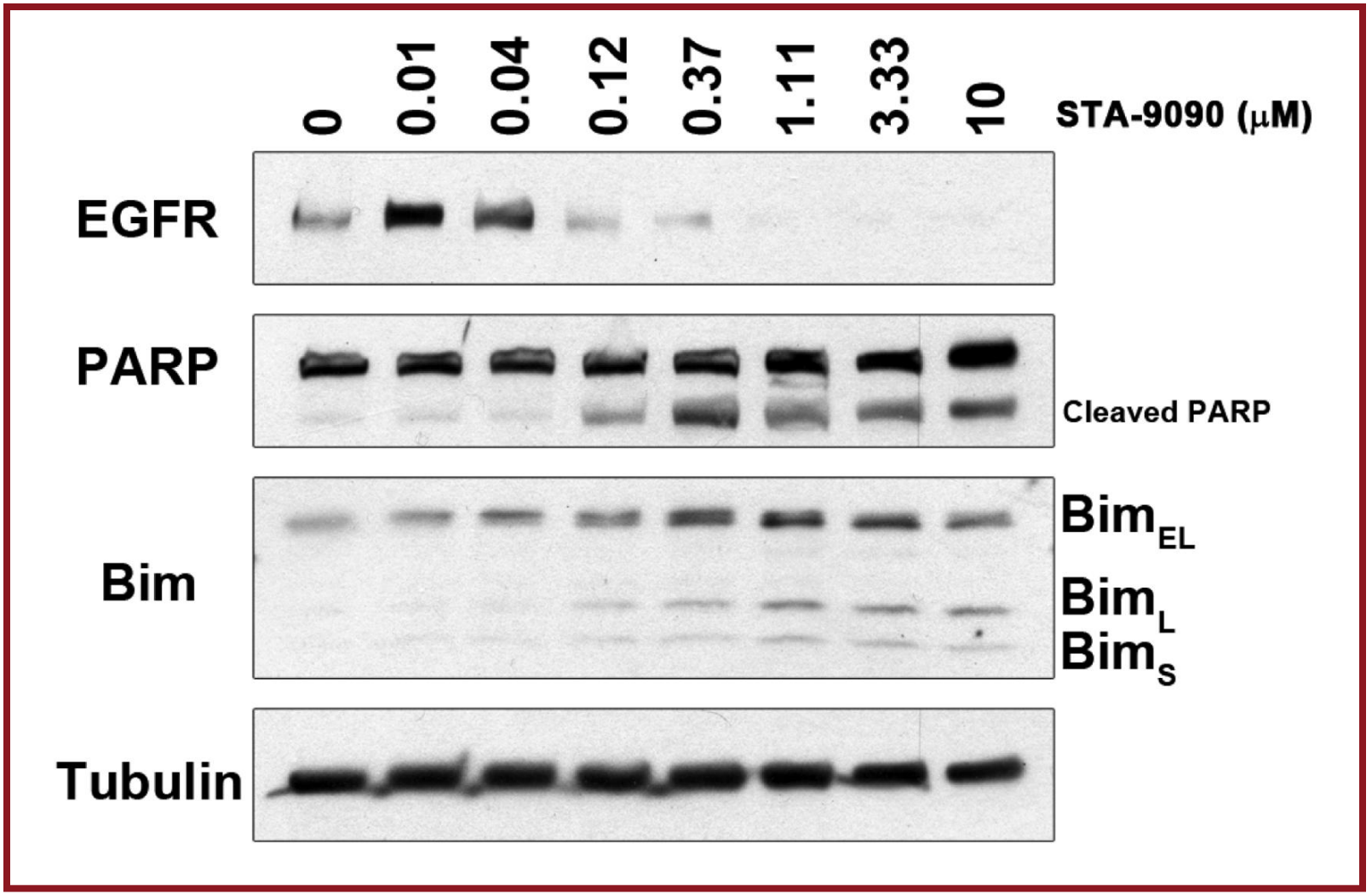
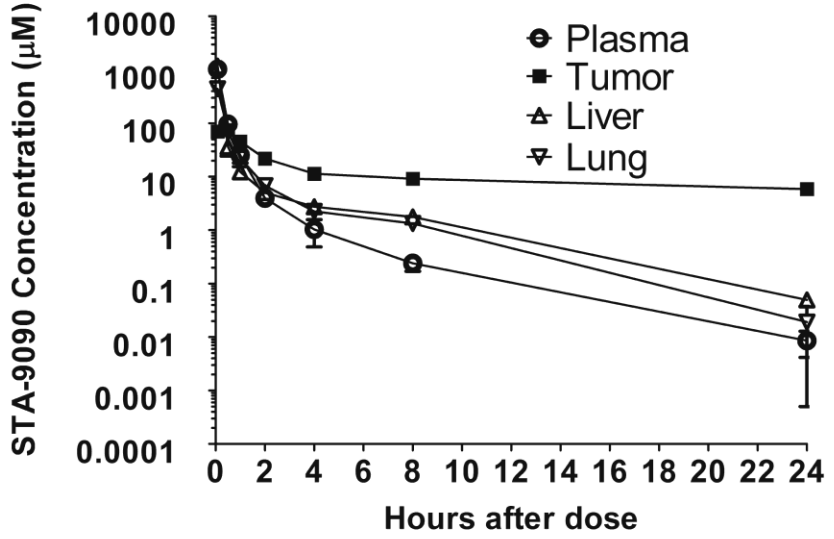
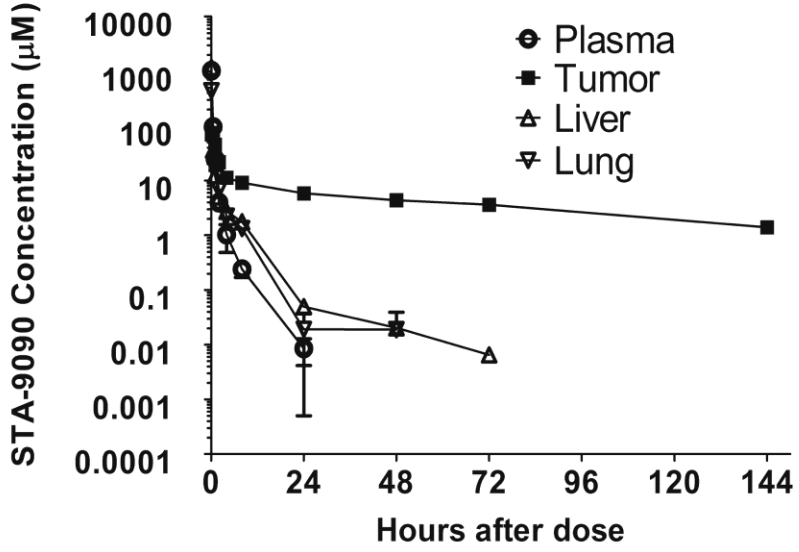
Induction of Bim by STA-9090



STA-9090 Pharmacokinetics following a single dose in mice bearing NCI-H1975 xenografts





Kevin Foley, Synta Pharmaceuticals

STA-9090 Pharmacokinetics in mice bearing NCI-H1975 xenografts

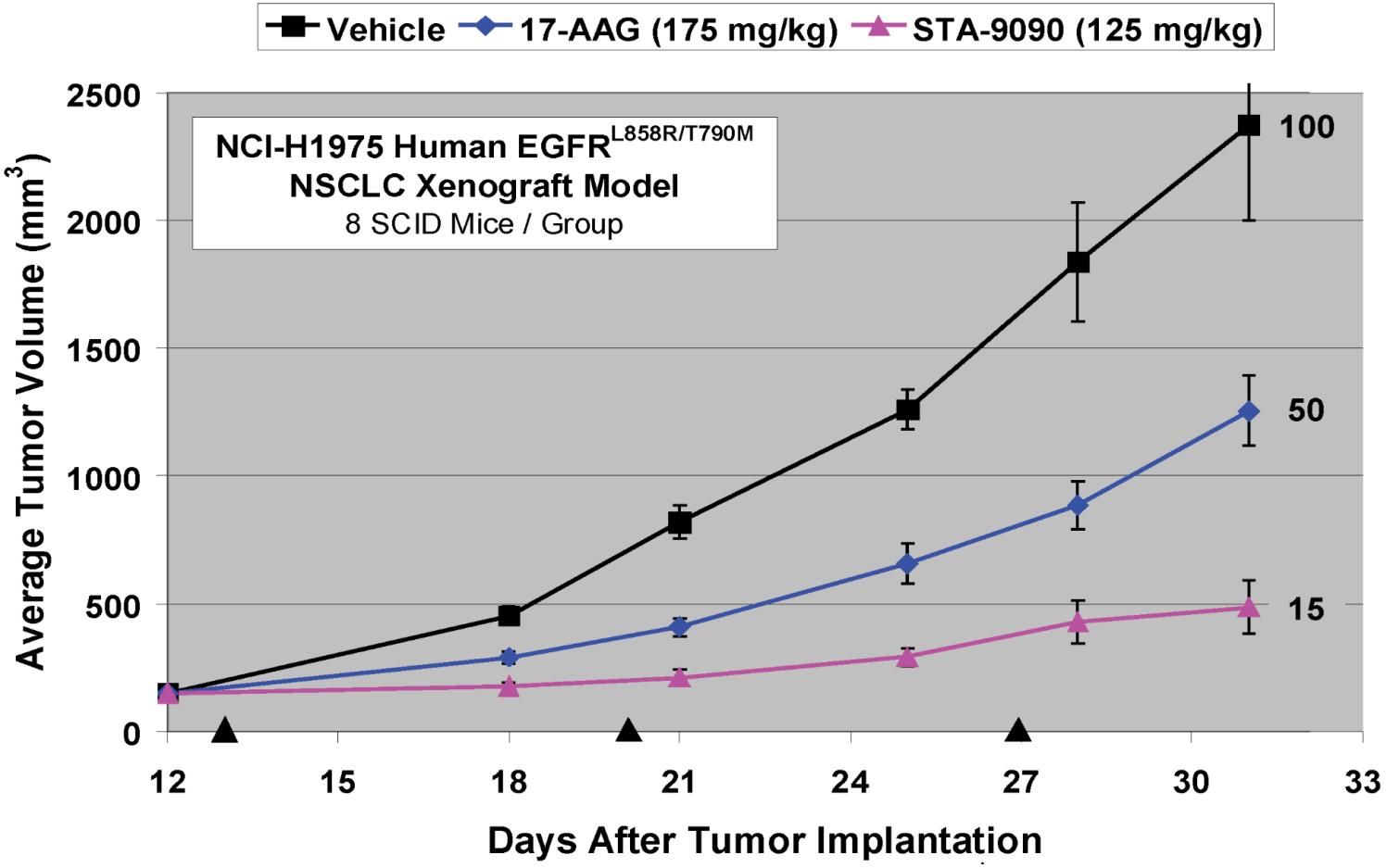
STA-9090 Biodistribution: Pharmacokinetic parameters

Sample Typle	$T_{1/2}$ (h)	Tmax (h)	Cmax (μM)	AUClast (μM•h)	AUCinf (μM•h)	Vss (L/kg)	Cl (L/h/Kg)
Plasma (PS)	3	0.083	1025	399	399	0.23	0.86
Tumor (TM)	58.3	0.5	74.8	691	809	--	--
Liver (LV)	5.6	0.083	1165	352	352	--	--
Lung (LU)	5.4	0.083	441	190	190	--	--

Assuming 1μM = 1μmol/g of tissue

Kevin Foley, Synta Pharmaceuticals

STA-9090 is more efficacious *in vivo* than 17-AAG when dosing 1X/week at HNSTD*s



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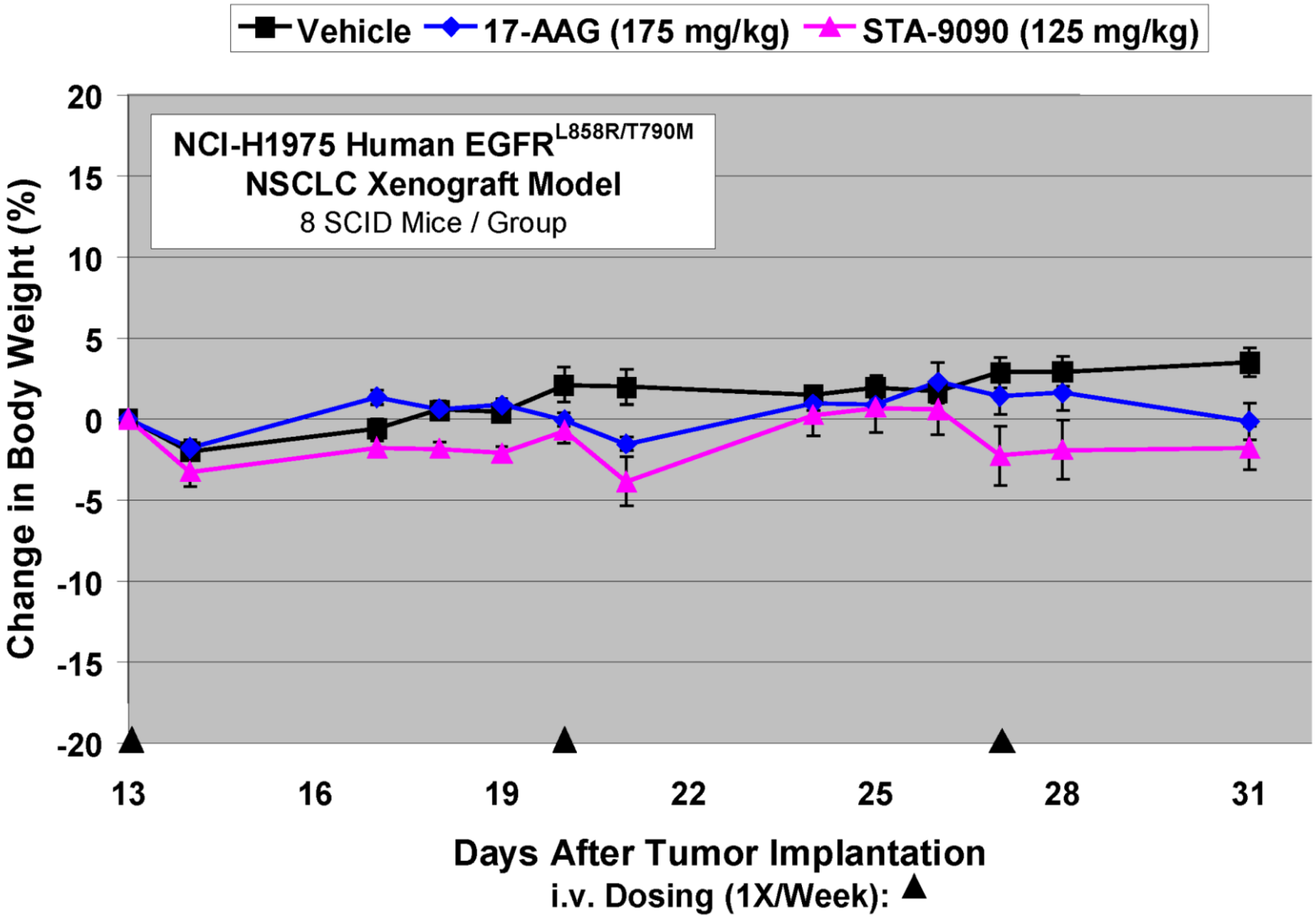
STA-9090 is well tolerated *in vivo*



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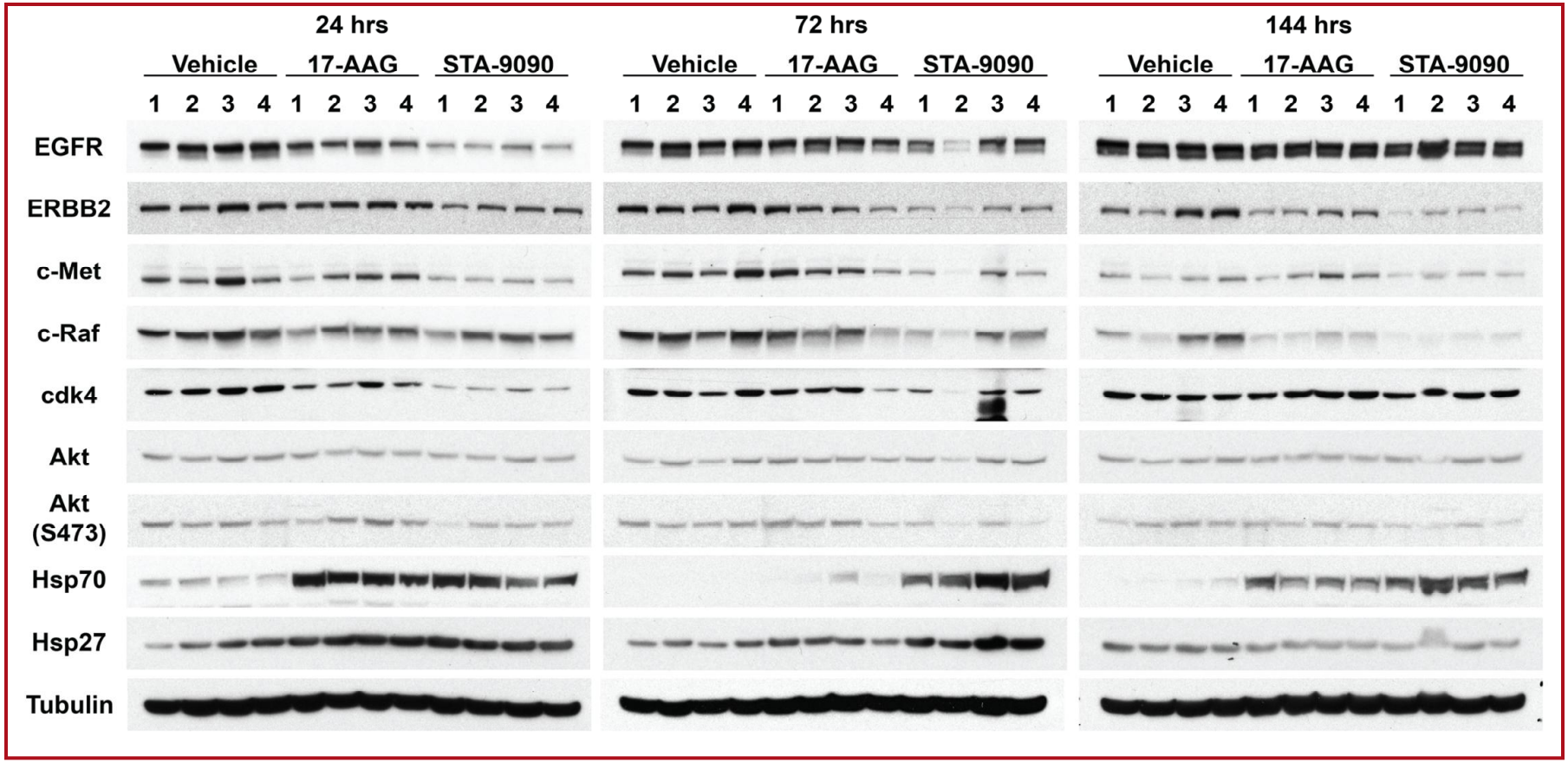
STA-9090 Pharmacodynamics in NCI-H1975 xenografts



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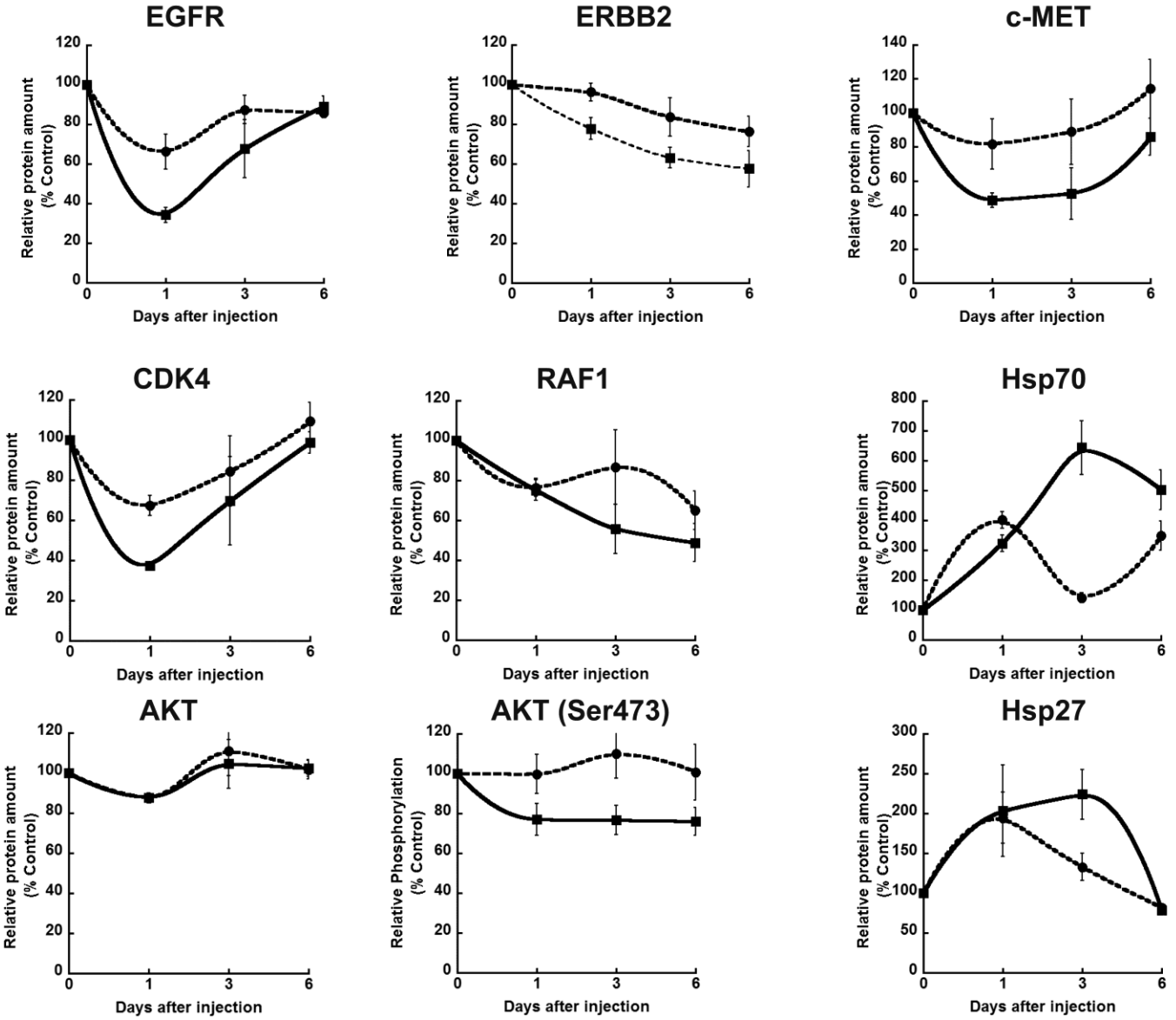
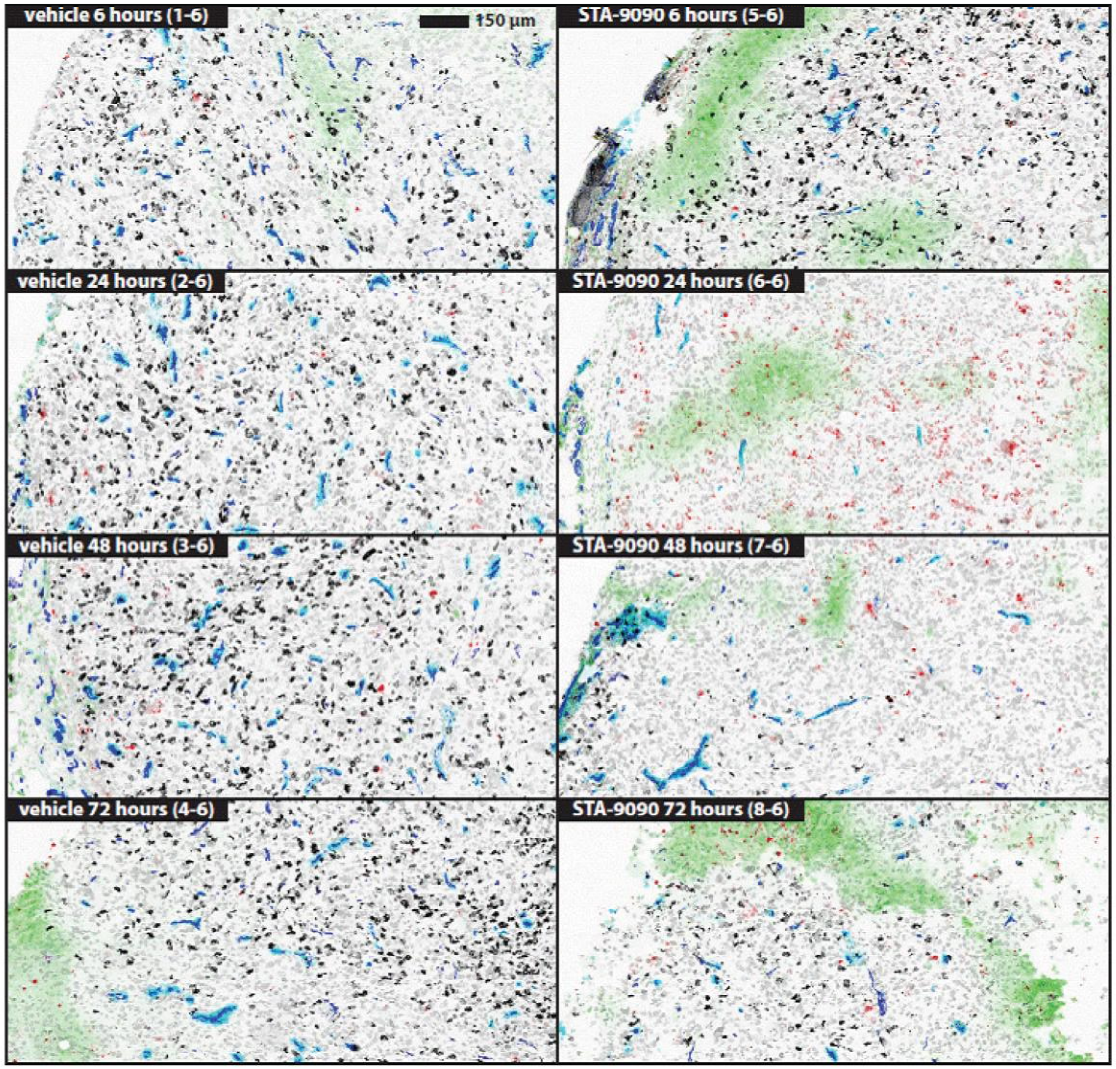
Quantification of Hsp90 clients after 17-AAG or STA-9090 in NCI-H1975 xenografts



STA-9090 Pharmacodynamics in NCI-H1975 xenografts



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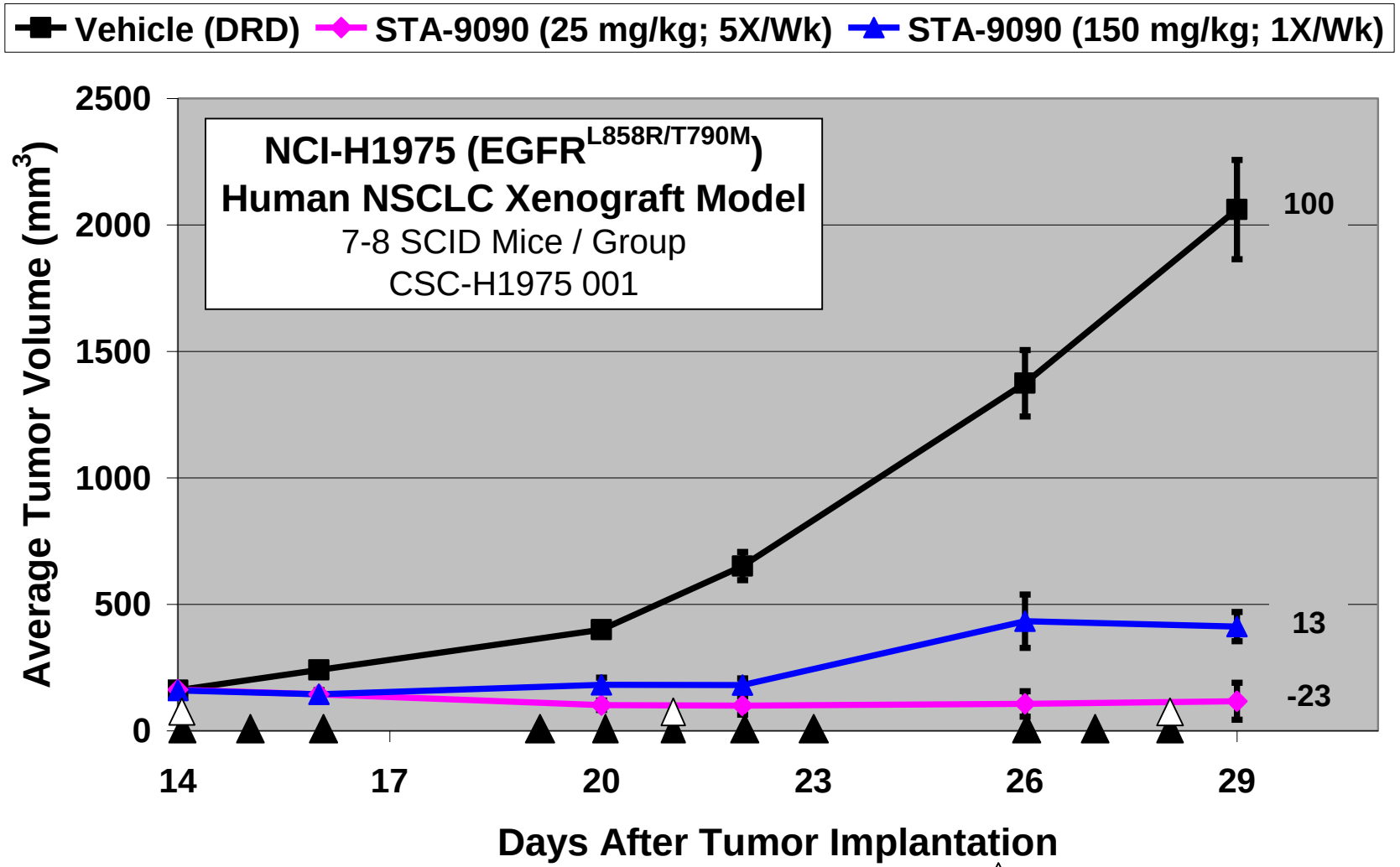
Consecutive Daily Dosing May be Superior to Intermittent Dosing



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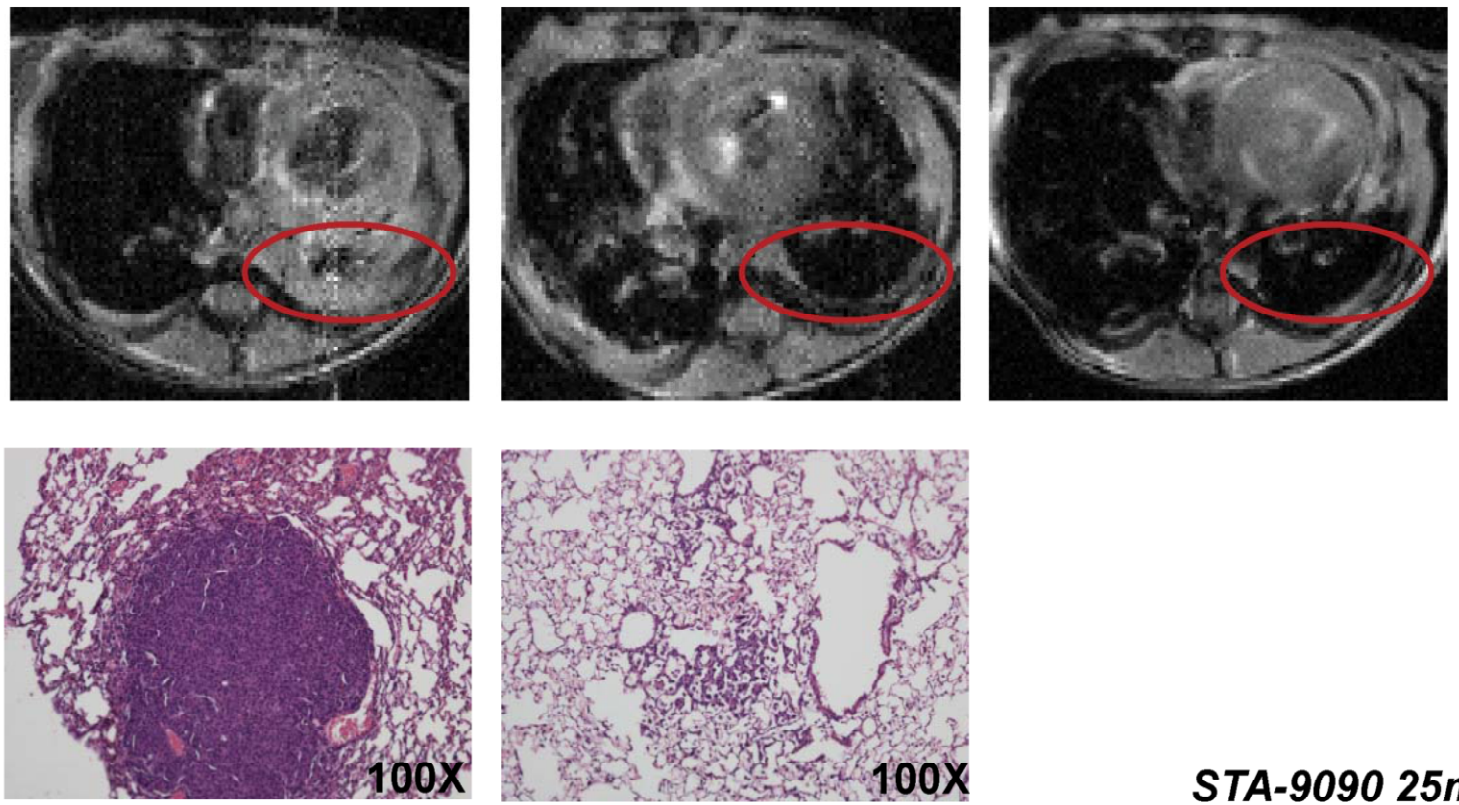
STA-9090 is active in an L858R/T790M murine adenocarcinoma model

Inducible bitransgenic mouse expressing EGFR$^{delE746\text{-}S752/T790M}$ responds to STA-9090 treatment



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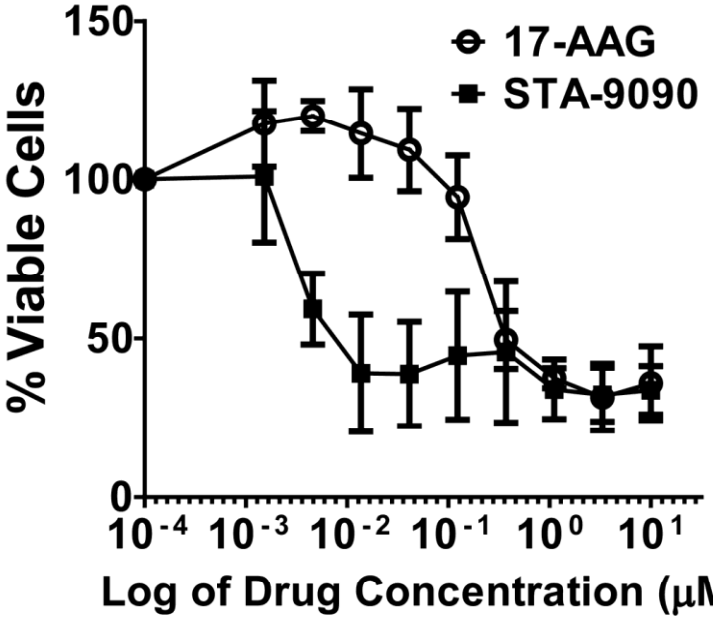
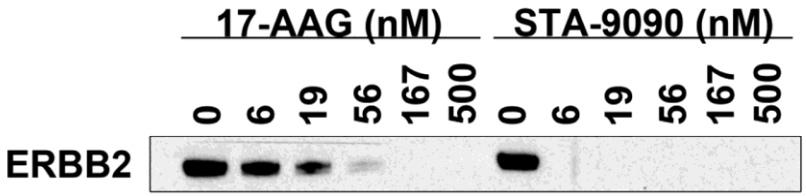
STA-9090 is active against BaF3 cells transformed by mutant ErbB2

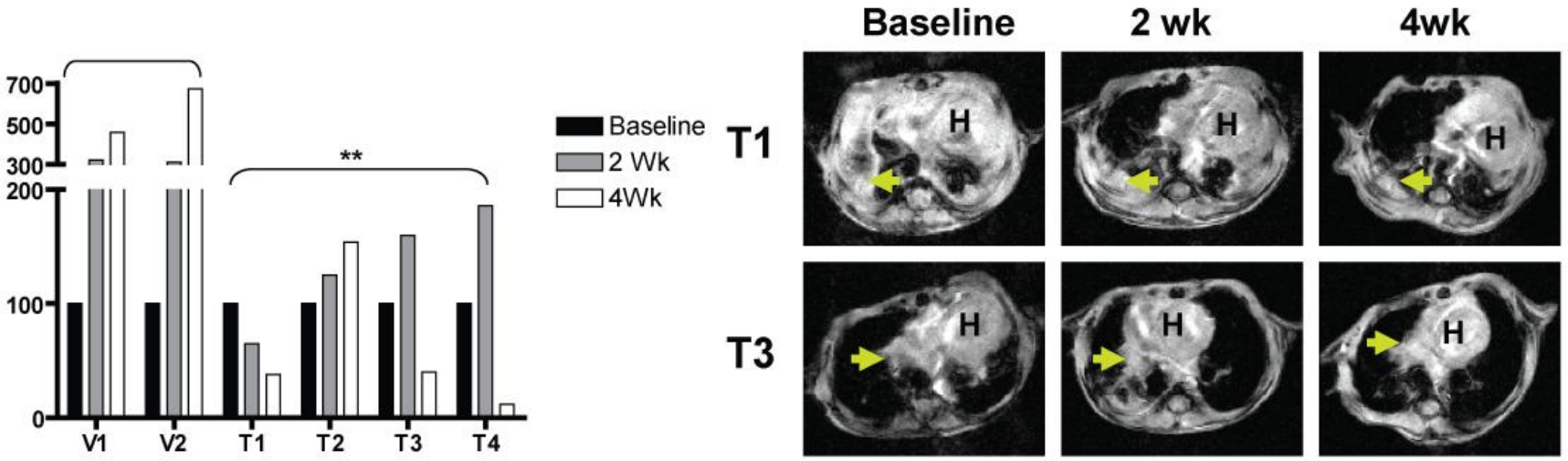




17-AAG IC50: 0.2033μM

STA-9090 IC50: 0.004275μM

STA-9090 against mutant Her2-driven murine adenocarcinoma



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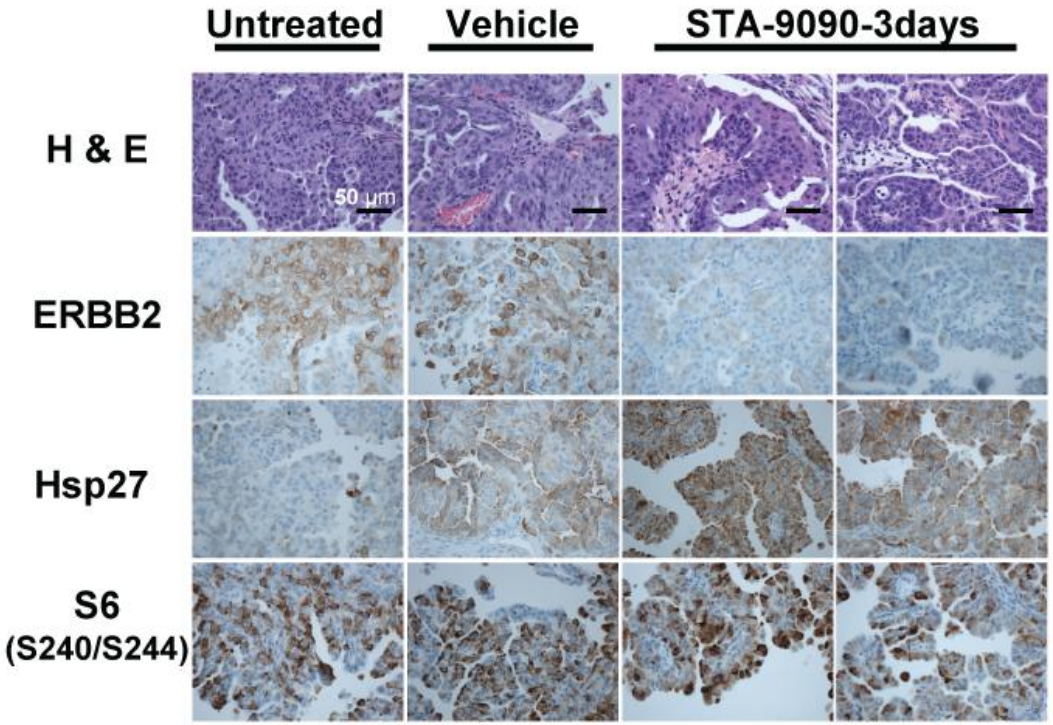
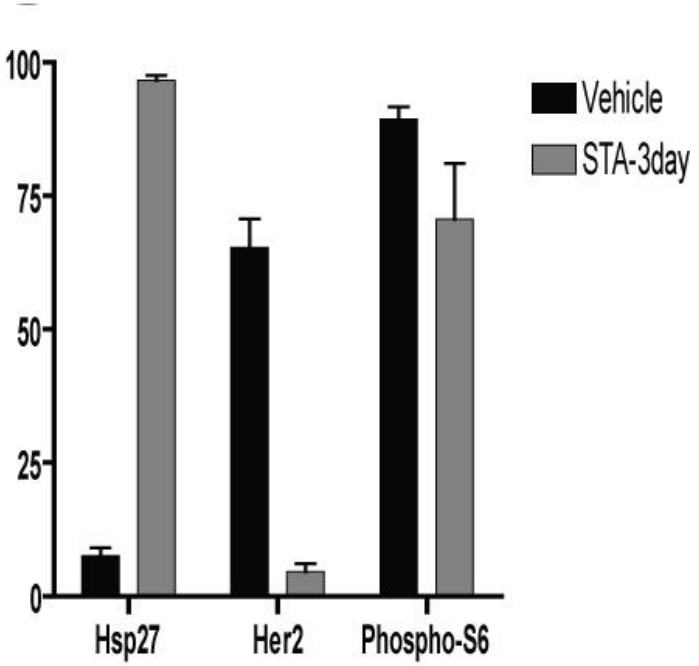
Pharmacodynamics of STA-9090 in mutant ErbB2-driven murine adenocarcinoma



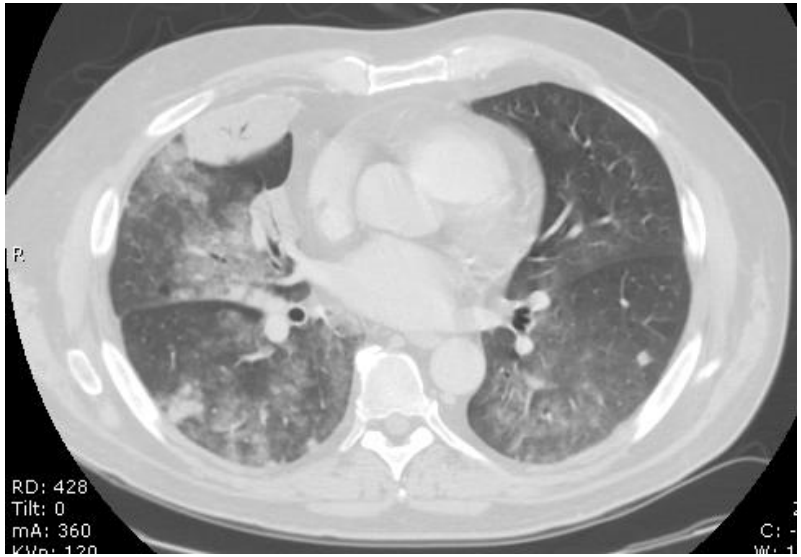


Phase 1 Ganetespib Safety Summary

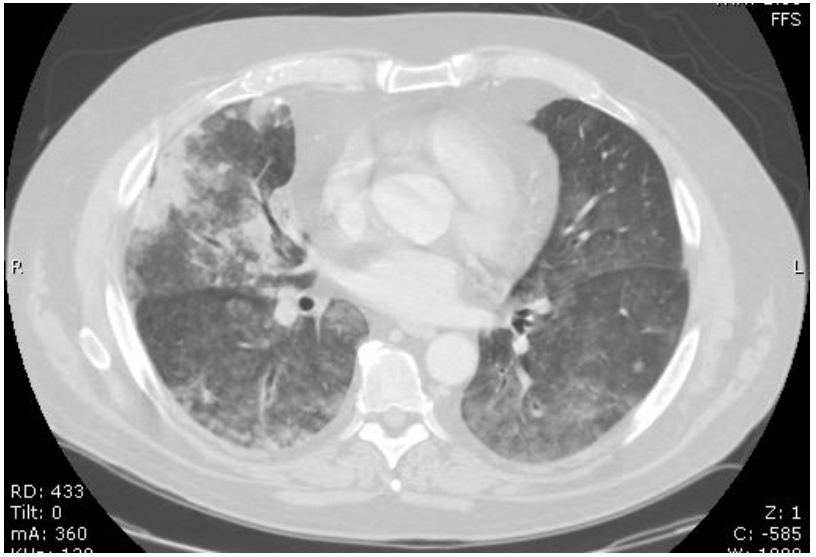
- Once-weekly dosing (3/4) MTD is 216 mg/m^2; recommended Phase 2 single agent dose is 200 mg/m^2
- Twice-weekly schedule (3/4) now evaluating 173 mg/m^2
- DLT is mild to moderate, manageable diarrhea (typical duration 24h post dose)
- Severe liver toxicity not observed
  - AST\ALT NCI CTC Gr ≥3 in 2-3% of patients (N=198)
  - Reversible and manageable with dose delay and reduction
- Only minimal ocular toxicity observed
  - 1 patient (0.5%; N=198) had symptoms consistent with ocular toxicity
  - Several patients (2-3%; N=198) had symptoms that may suggest ocular toxicity
- Bone marrow toxicity not observed

66M, NSCLC

Prior treatments included carboplatin, paclitaxel, bevacizumab, erlotinib, pemetrexed, topotecan, bortezomib, experimental retinoid; longest treatment duration: 2-4 months



Baseline



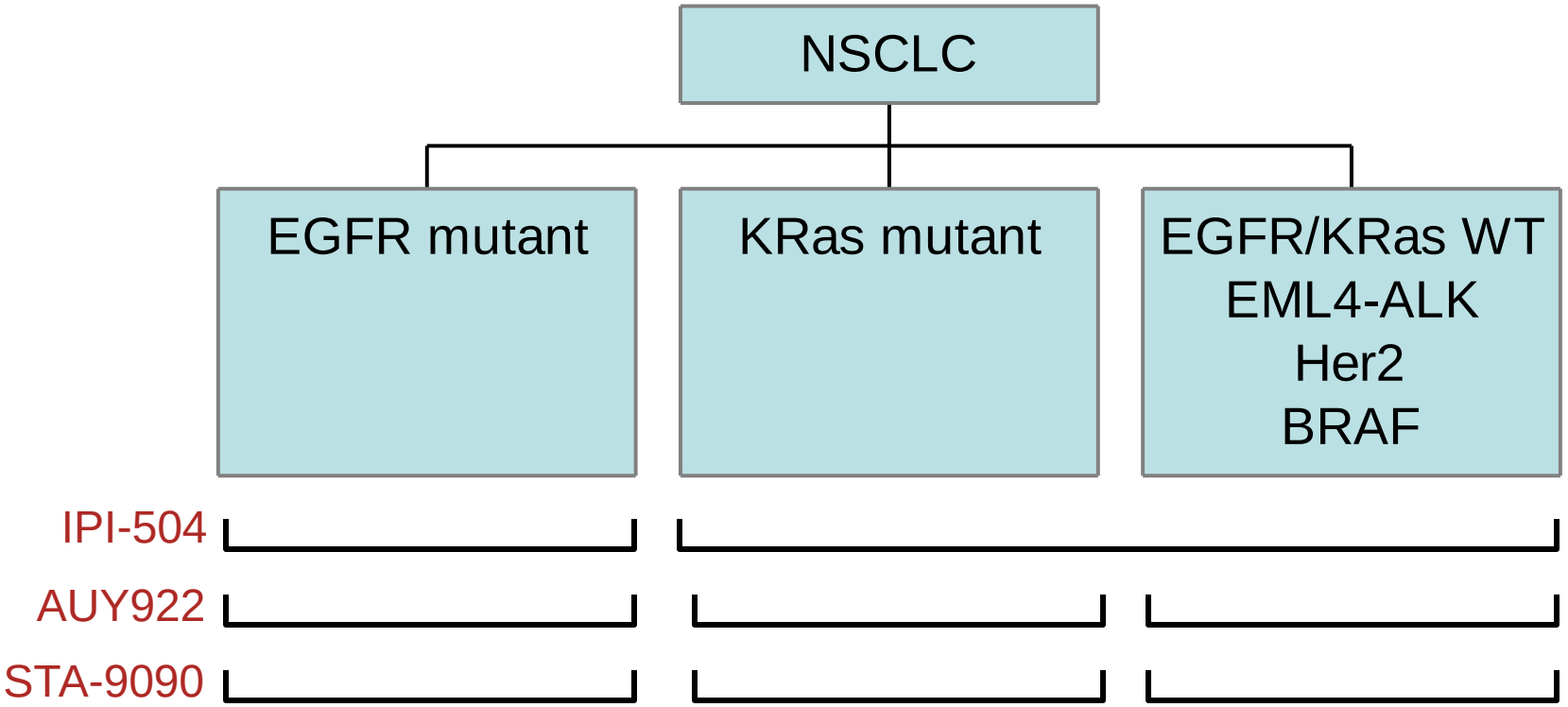
Post 4 cycles

Ganetespib 150 mg/m^2: 26% total TL tumor shrinkage. Duration: 13 months

Additional Durable Responses and Regressions in Phase 1 Studies

- Melanoma (PR)
- Colon (PR)
- RCC (durable regression)
- GIST (durable regression)
- NSCLC (durable regression)
- AML (hematologic improvement)
- CML (hematologic improvement)

Schema for Hsp90 Inhibitor Trials in NSCLC



Phase 2 Study of Ganetespib in NSCLC

Ganetespib at 200 mg/m2 qW for 3 wks, 1 wk off until disease progression

Stage IIIB/IV NSCLC PS 0 and 1 PD at study entry

- **A: mutant EGFR (n=15)**
- **B: mutant K-Ras (n=17)**
- **C: wild type EGFR / wild type K-Ras (n=25)**
- **D: Extension: adenocarcinoma only (n=32)**

- Genotyping required for all patients
- Primary endpoint : PFS at 16 wks
- Two stage design: for cohorts A,B,C: Stage 1 (n=14 pts); if ≥ 2 pts progression-free at week 16, enroll Stage 2 (n = 9 pts)
- Patients who progressed on single agent but had some clinical benefit were allowed to roll over to cohort E: weekly ganetespib + docetaxel (n=5)

Analysis Population: Cohort C & D

Evaluable patients for clinical activity (N = 33)

- Adenocarcinoma patients with wild-type EGFR & K-ras
- Met main inclusion / exclusion criteria
- Received at least one dose of study drug
- Had both baseline and at least one follow-up radiological assessment

Safety population (N = 36)

- All adenocarcinoma patients with wild-type EGFR & K-ras who received at least one dose of study drug

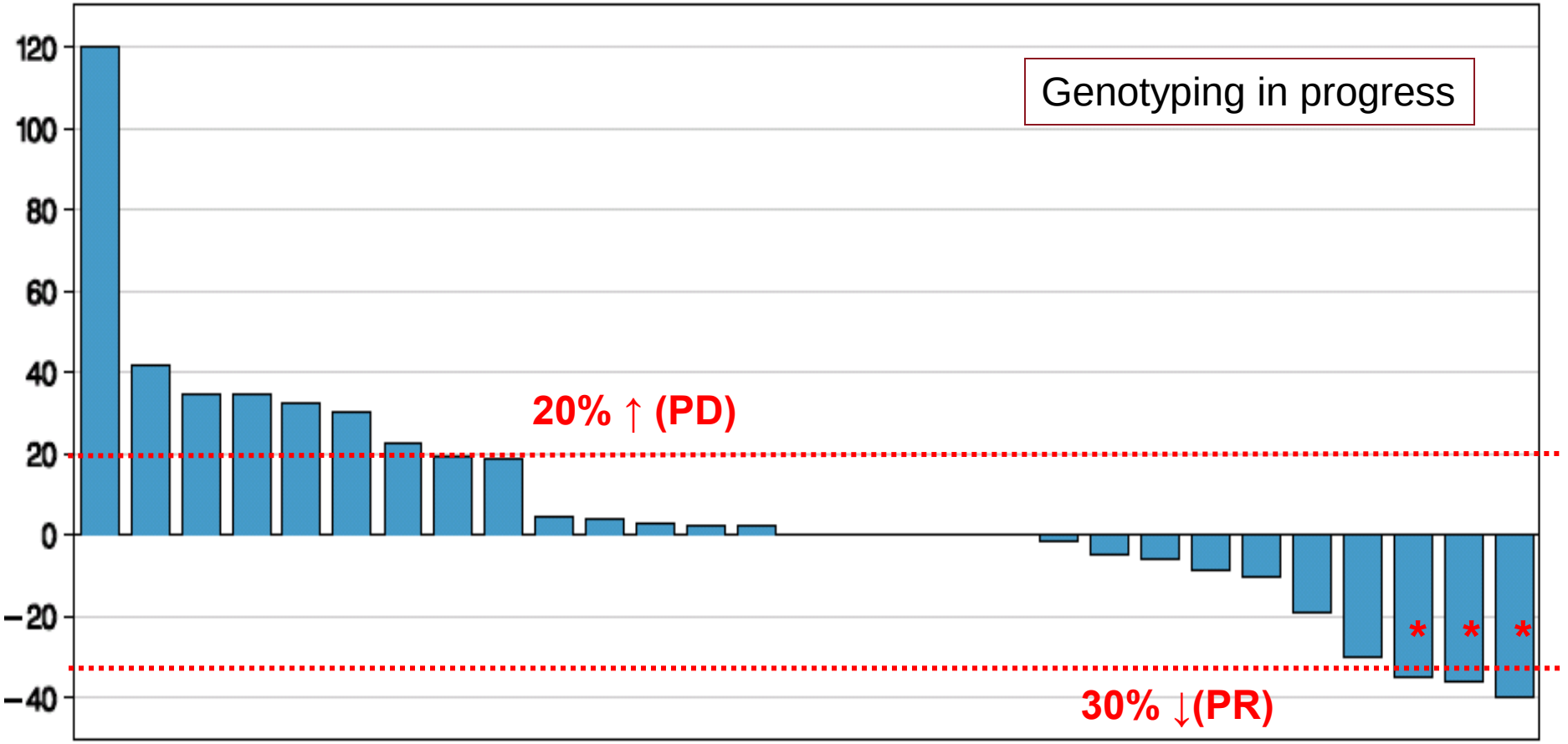
Demographics and Baseline Status

	N=36
Age (yrs)	
Median	59
(Range)	36- 82
Sex (N, %)	
Male	19 (52.8)
Female	17 (47.2)
ECOG Status (N,%)	
0	10 (27.8)
1	24 (66.7)
Unknown	2 (5.5)
# Prior Treatments	
Mean	3
Median	2
(Range)	1 - 10

Treatment-related AEs occurring in ≥ 10 % of patients

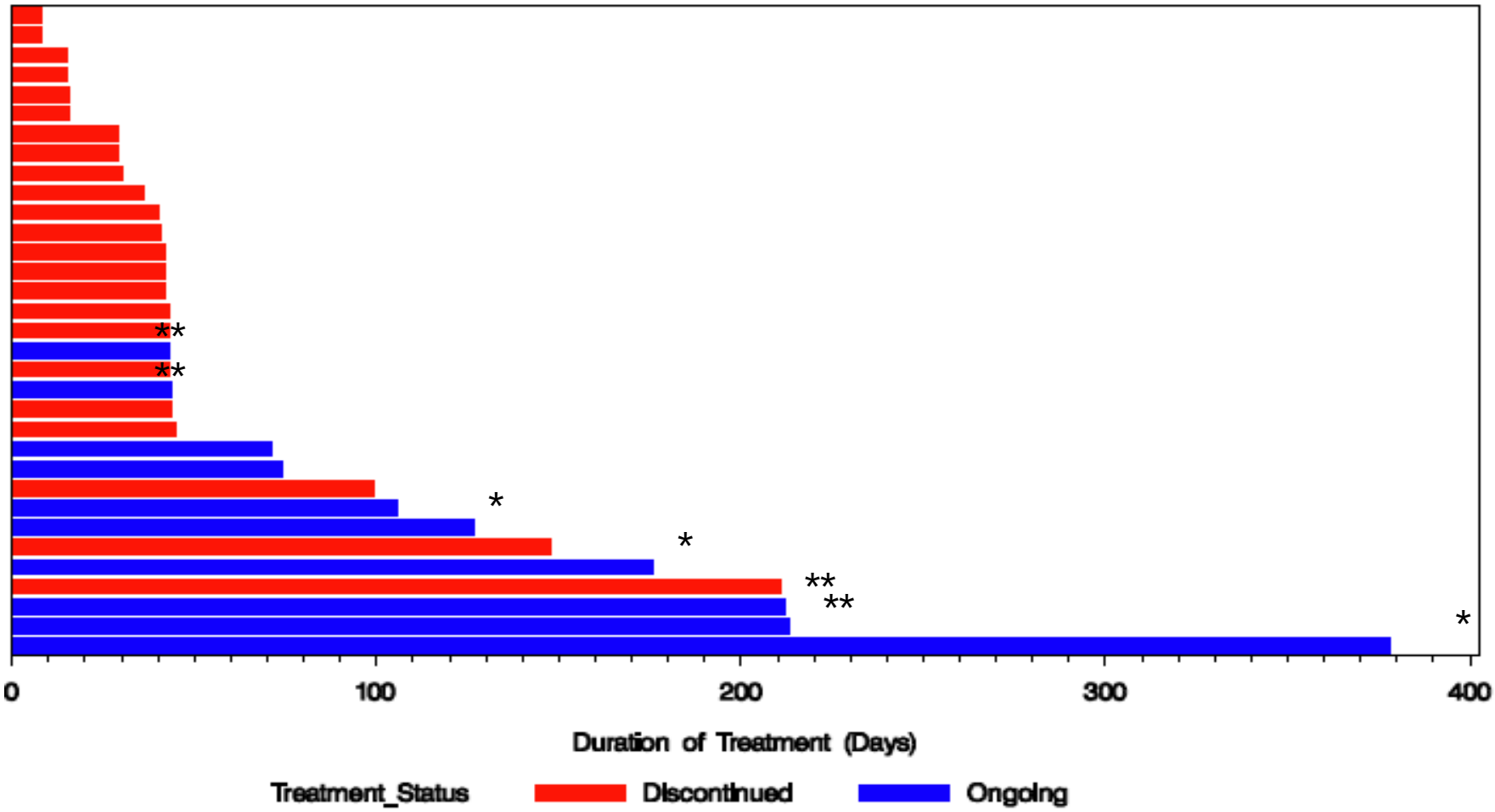
	AEs N (%) N=36	≥ Grade 3 AEs N (%) N=36
Diarrhea	29 (81)	2 (6)
Fatigue	11 (31)	3 (8)
Nausea	11 (31)	0
Insomnia	6 (17)	2 (6)
Increased Alk Phos	5 (14)	0
Decreased appetite	5 (14)	0

Best change sum of longest diameters



22 pts target lesion stabilization (<20%)
10 pts target lesion regression
3 confirmed PRs – durable: 14+ months; 6+ months; 6+ months; all ongoing

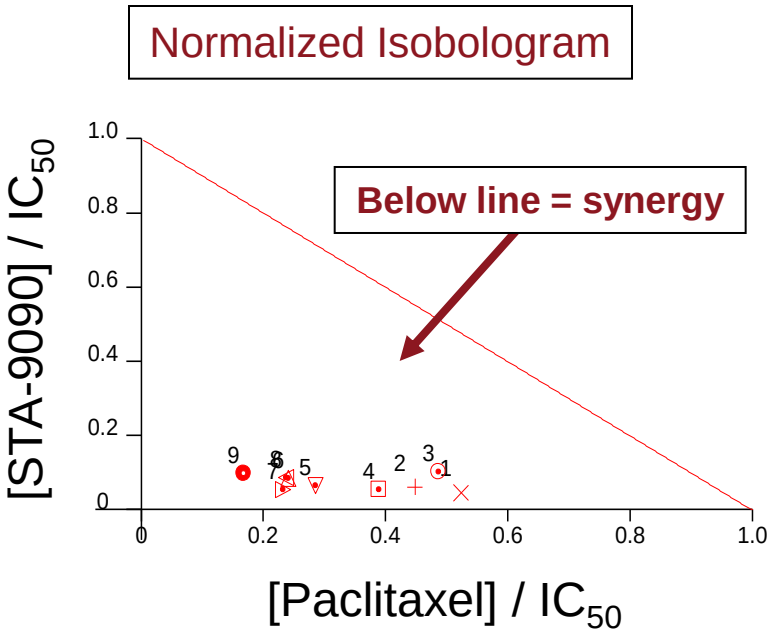
Time on treatment



* Confirmed PRs

** Continued treatment on combination (Cohort E)

Synergy with taxanes: *in vitro*



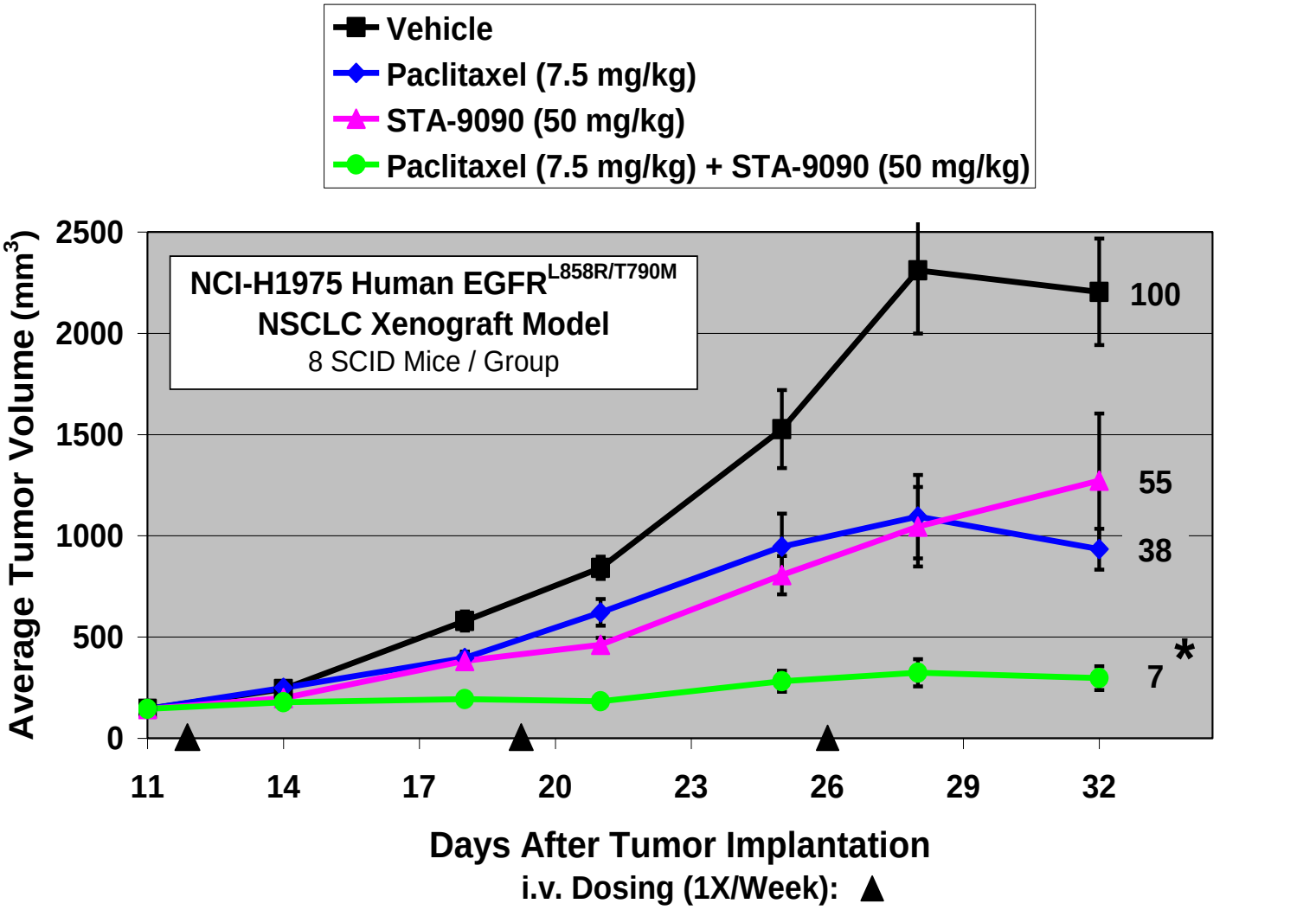
STA-9090 (nM)	Paclitaxel (nM)	Ratio	CI	Combo Effect
2.51	0.69	3.6:1	0.59	Synergy
4.01	0.69	5.8:1	0.33	Synergy
6.42	0.69	9.3:1	0.27	Synergy
2.51	1.11	2.3:1	0.51	Synergy
4.01	1.11	3.6:1	0.35	Synergy
6.42	1.11	5.8:1	0.33	Synergy
2.51	1.78	1.4:1	0.57	Synergy
4.01	1.78	2.3:1	0.45	Synergy
6.42	1.78	3.6:1	0.29	Synergy

CI < 1 = synergy
CI = 1 = additive
CI > 1 = antagonism

NCI-H1975 ($EGFR^{L858R/T790M}$) erlotinib-resistant human NSCLC cells treated concurrently for 72 hrs
Synergy calculated by median-effect method of Chau & Talalay

Foley, AACR-IASLC 2010

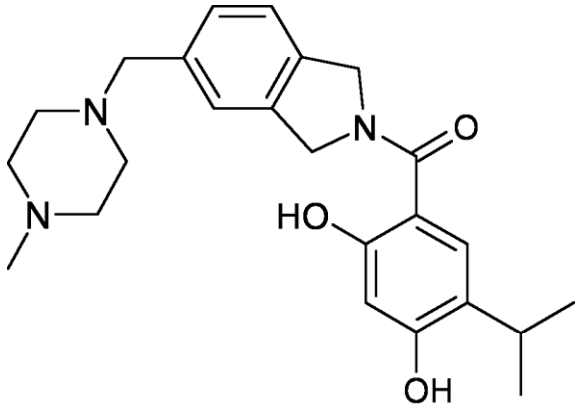
Synergy with taxanes: *in vivo*



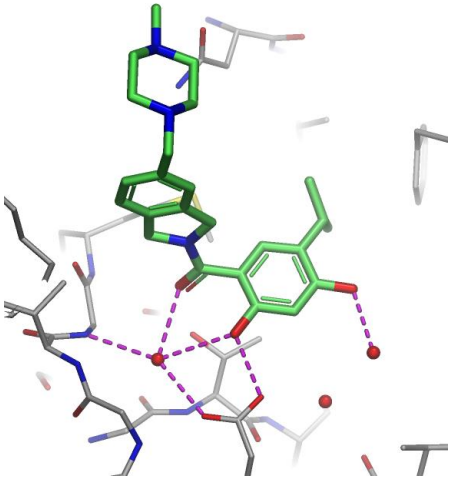
* $P < 0.05$

Foley, AACR-IASLC 2010

AT13387



AT13387
0.00071μM (ITC)
58nM (HCT116)



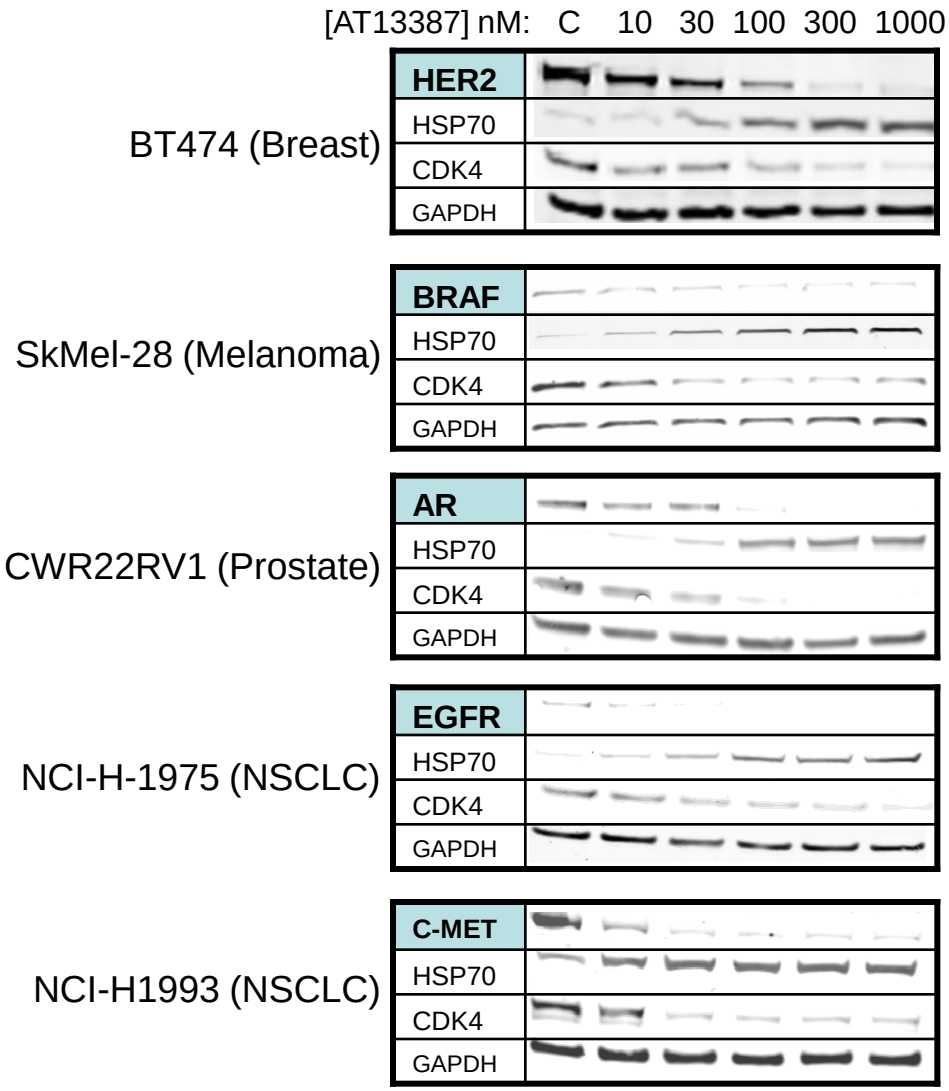
Data courtesy John Lyons and Astex Investigators

AT13387 *In Vitro* Preclinical Data

AT13387 Kd (nM)	
HSP90α	0.6

By ITC competition assay

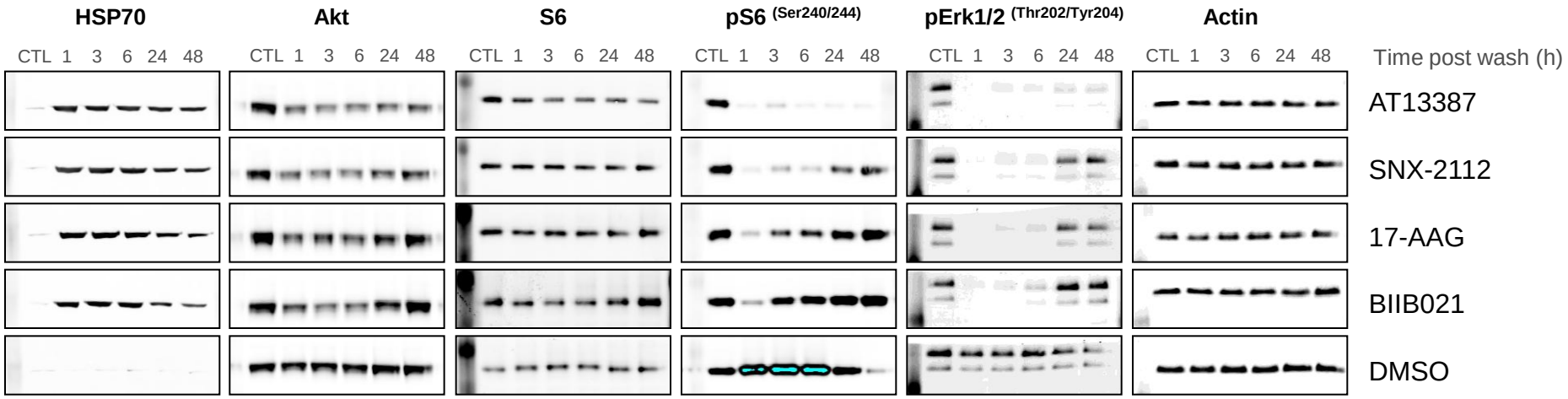
Origin	Cell line	IC_{50}
Colon	HCT116	58
	HT-29	80
Lung	A549	22
	NCI-H1975	14
	NCI-H1703	58
	A431	27
Breast	MCF-7	64
	MDA-MB-468	26
	T47D	30
	BT474	23
Myeloma	U266	70
	RPMI 8226	70
Prostate	LNCaP	78
	22Rv1	46
Melanoma	A375	18
	SkMel 28	46



Cells were treated for 18 hours with the stated doses of AT13387 before harvesting cell lysates and immunoblotting

Duration of AT13387 Action *in vitro:* Comparison with Other Agents

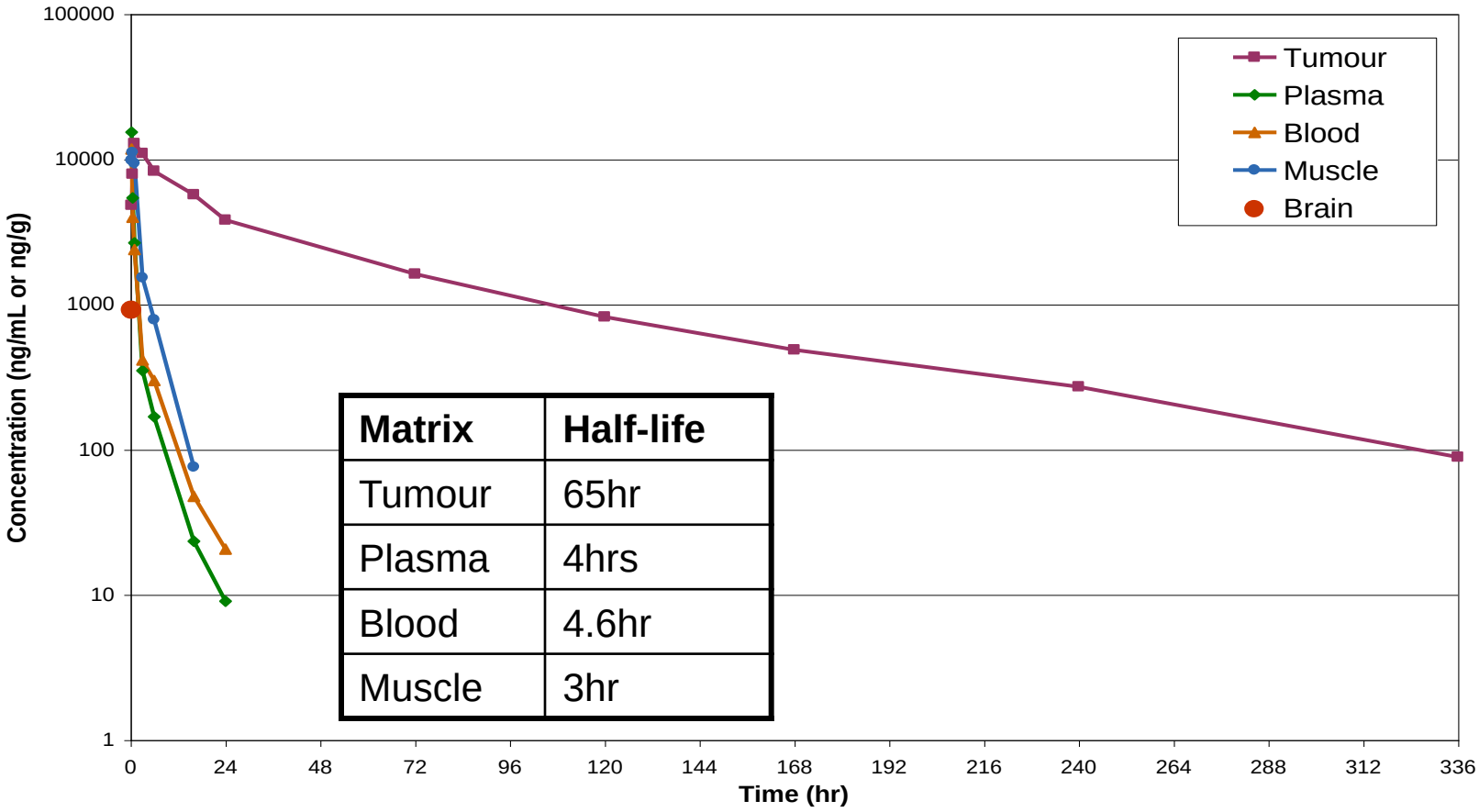
AT13387 suppresses client proteins and phospho-signaling longer than other HSP90 inhibitors



A375 *B-Raf* mutant melanoma cells were treated with 1μM AT13387 for 24h

AT13387 - Prolonged Tumor Half-life

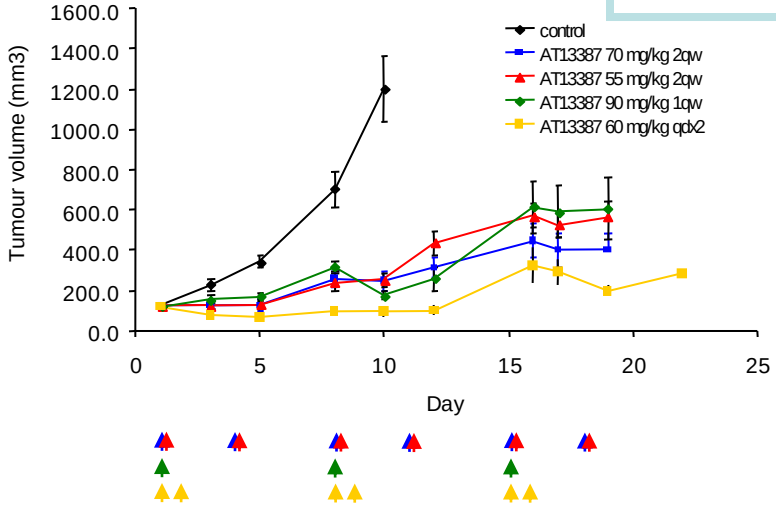
Mouse IP, single dose, 60mg/kg



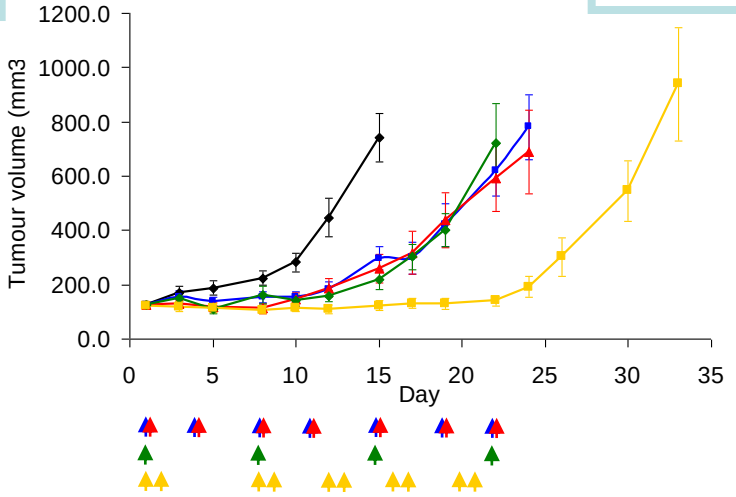
Matrix	Half-life
Tumour	65hr
Plasma	4hrs
Blood	4.6hr
Muscle	3hr

AT13387 – Anti Tumor Activity in Xenografts

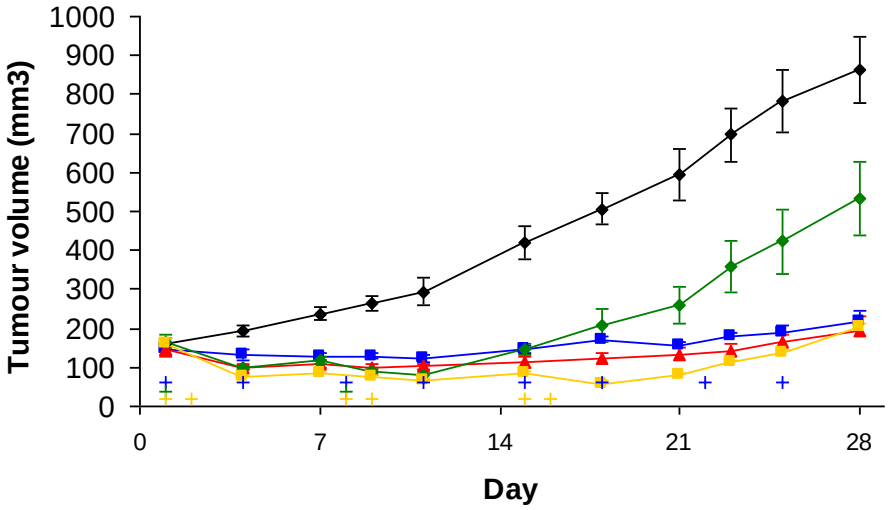
NCI-H1975



A375

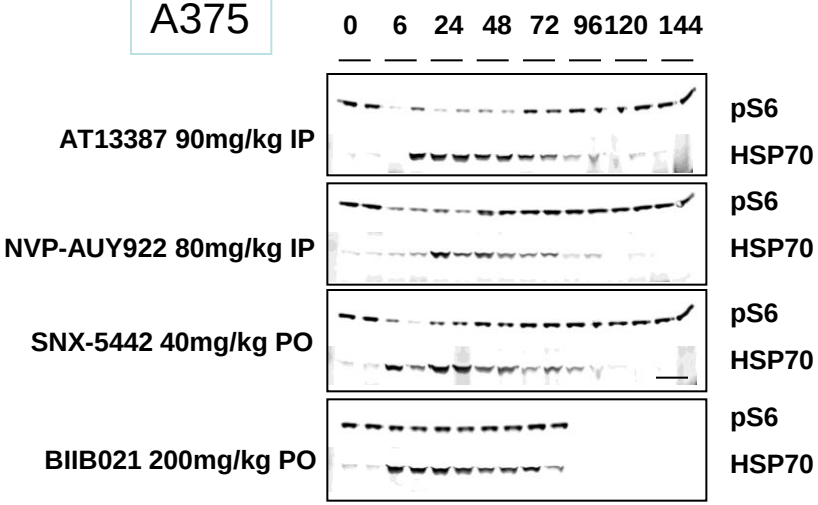


MV4-11



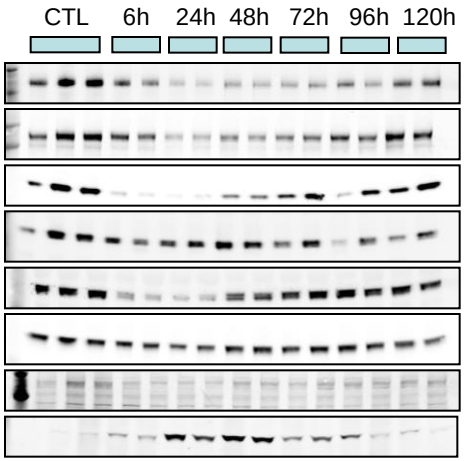
Pharmacodynamic Effects of AT13387 in Xenografts

A375

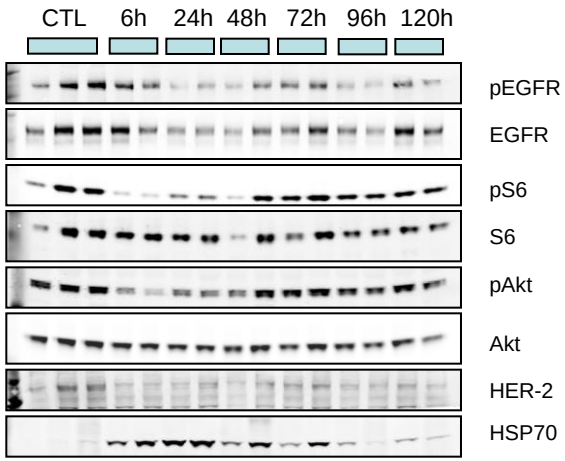


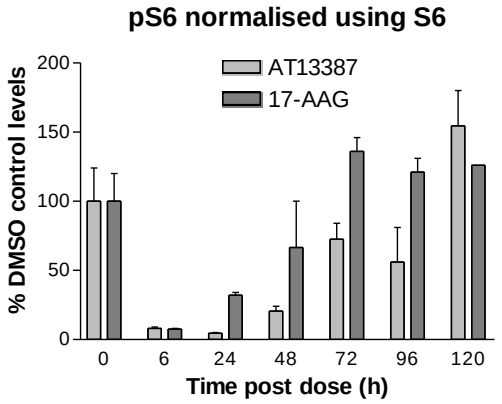
Extended tumor pharmacodynamics are a consequence of 2 properties:
Slowly reversible HSP90 inhibition
Long tumor half life

AT13387 80mg/kg IP



17-AAG 80mg/kg IP





NCI-H1975

AT13387 - Phase I Study

Dose escalation study in patients with refractory solid malignancies

- Enrichment for tumor types whose growth is dependent upon known HSP90 client proteins
  - melanoma, lung, prostate or breast
- Schedules
  - Twice weekly schedule: 1 hour IVI, three weeks out of four (days 1,4,8,11,15,18 q. 28)
  - Once weekly schedule: 1 hour IVI, three weeks out of four (days 1, 8, & 15 q. 28)

AT13387 Twice Weekly Schedule

Dose Level *(total dose per 4 week cycle)*	**Patients Treated (Number)**	**Number of Cycles Received**	**Dose Limiting Toxicities**
10 mg/m^2 *(60 mg/m^2)*	Glioblastoma multiforme (1) Prostate cancer (1) Colorectal adenocarcinoma (2)	1- 3 *(median 2)*	None
20 mg/m^2 *(120 mg/m^2)*	Thyroid cancer (1) Melanoma (1) Colorectal adenocarcinoma (1)	2– 6 *(median 2)*	None
40 mg/m^2 *(240 mg/m$^{2)}$*	Melanoma (2) Squamous cell carcinoma of the oesophagus (1)	2 - 3 *(median 3)*	None
80 mg/m^2 *(Six doses = 480 mg/m$^{2)}$*	Colorectal adenocarcinoma (2) Melanoma (1) Carcinoma of the pituitary gland (1) NSCLC (1)	1 – 8 *(median 2)*	None
120 mg/m^2 *(Six doses = 720 mg/m^2)*	Synovial sarcoma (1) Melanoma (3), NSCLC (3) Pancreatic cancer (1) Colon cancer (1), GIST (1) Small cell bladder cancer (1) Prostate cancer (1), Breast cancer (1)	1 – 4 (*median 2*)	Diarrhea & fatigue (Grade 1 and 2) Reversible visual changes (Grade 1 - 3) Hypotension during dosing (Grade 2) Dry skin/mouth (Grade 1)

120 mg/m^2/day declared as MTD on basis of aggregated toxicities

AT13387 Twice-Weekly Schedule

- 28 patients treated with twice-weekly dosing, 10 mg/m^2 to 120 mg/m^2
- MTD/RP2D: 120 mg/m^2 on the basis of aggregate mild/moderate toxicities (plus 1 DLT – grade 3 visual disturbance)
- Gastrointestinal toxicity (especially transient diarrhoea) and fatigue most commonly reported
- 9/9 patients treated with 120 mg/m^2 twice weekly experienced visual disturbances (13 pts actually treated at this dose)
- 3 AEs of QTc prolongation reported: Grade 1 in two pts (one at 10 mg/m^2 and one at 80 mg/m^2); Grade 2 in one pt at 120 mg/m^2 Review of QT data from Holter monitoring showed no consistent effects; QT prolongation post-dosing only seen in patients with abnormal QT at baseline
- Two patients had SD > 6 months: uveal melanoma; follicular thyroid ca

AT13387 Once Weekly Schedule

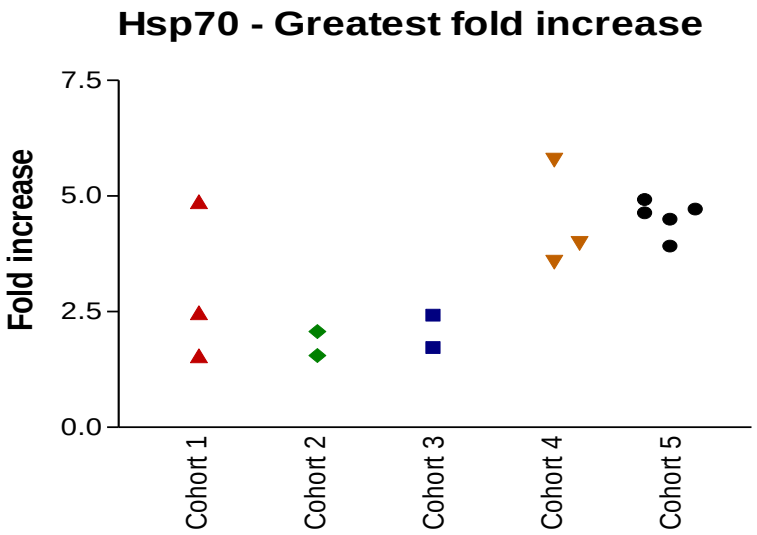
Dose Level *(total dose per 4-week cycle)*	**Patients Treated (Number)**	**Number of Cycles Received**	**Dose Limiting Toxicities/AE's**
150 mg/m^2 *(450 mg/m^2)*	Mesothelioma (1) Pancreatic (1) GIST (1) Melanoma (1)	1- 3 *(all patients off study, median 1 cycle)*	No DLTs; 1/4 visual disturbances (with dry eyes); 2/4 infusion reactions
180 mg/m^2 *(540 mg/m^2)*	GIST (1) Rectal (1) Bronchioloalveolar (1)	2-3 *(all patients off study, median 3 cycles)*	No DLTs; 3/3 with grade 1-2 GI toxicities; 1/3 infusion site reaction 1/3 color vision disturbance
220 mg/m^2 *(660 mg/m^2)*	GIST (1) Colon (1) Melanoma (1) NSCLC (2) Cholangiocarcinoma (1)	Cycle 3 complete (PET response/SD), ongoing Died 1 week after 1st dose (PD?) Cycle 1 complete, ongoing PD after 1 cycle Cycle 1 day 8 – 25th Jan, ongoing Cycle 1 day 15 – 24th Jan, ongoing	No DLTs; All patients had grade 1-2 GI symptoms (nausea, diarrhea). Insomnia, HA, scalp soreness, palatal swelling & pain, fatigue, chills, flushing, & infusion site reactions also seen; Grade 1 visual disturbance in 2 pts (apparent movement of stationary objects; color disturbance)

HSP70 is Up-Regulated in PBMCs in a Dose-Dependent Manner

Cohort 5 (120mg/m2/day) Pt 01 006

Hsp70 is induced in peripheral blood mononuclear cell (PBMC) lysates during infusion

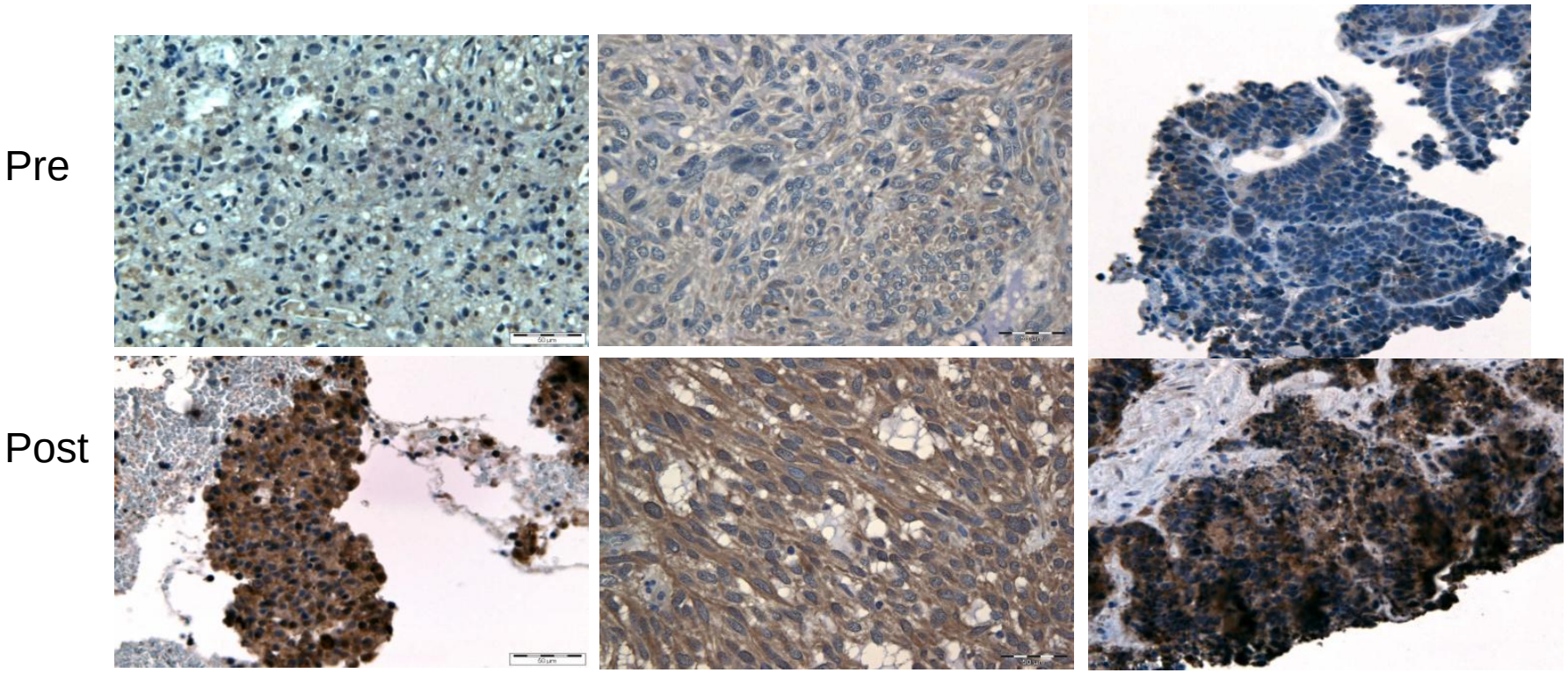
Across cycles in each patient



The greatest fold increase in Hsp70 is plotted for each individual

Trend is for larger fold consistent increases at 80 mg/m^2 and above

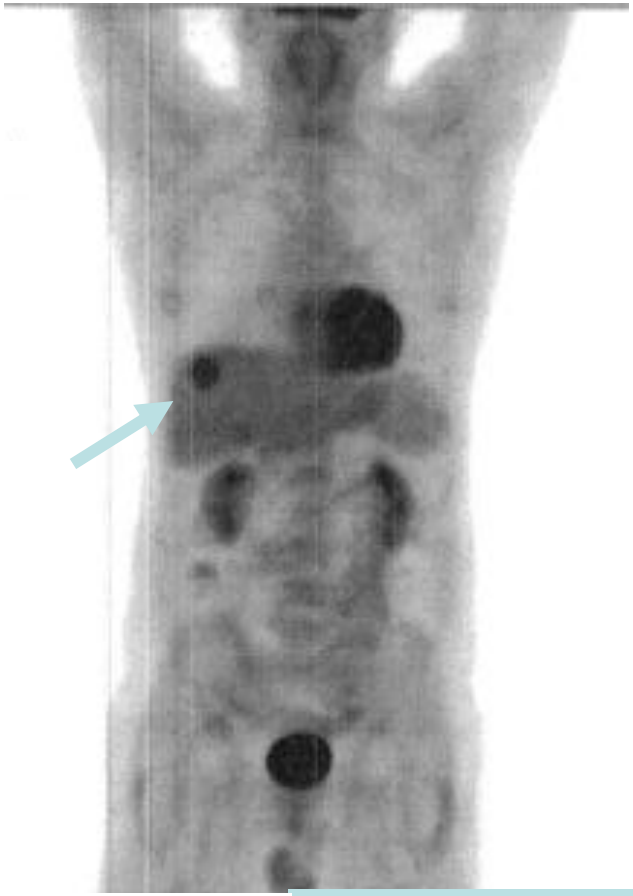
Hsp70 Induction in Tumor Biopsies



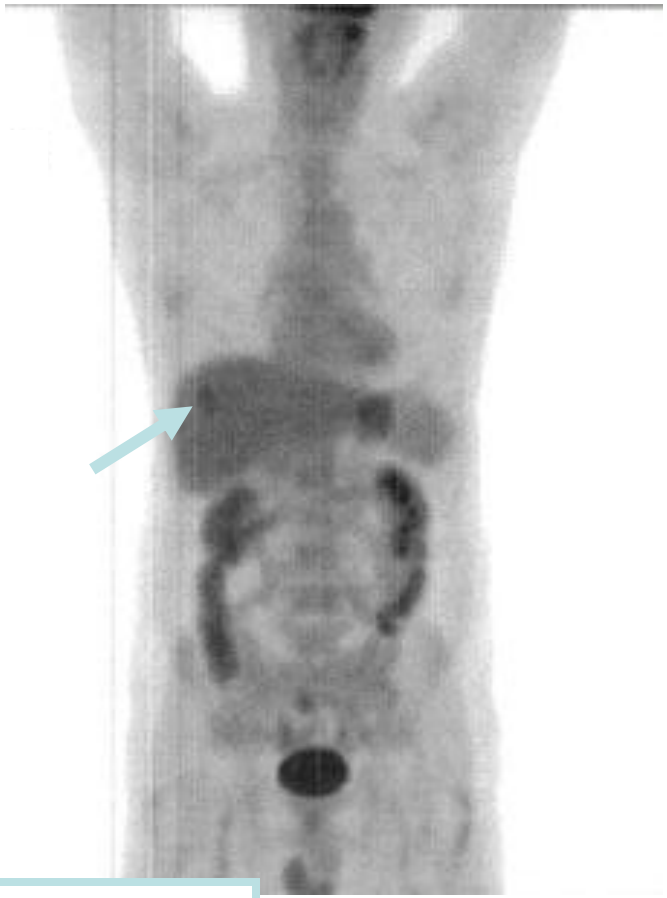
Post-treatment biopsies taken on day 19; 24 hours after 6th dose at 120 mg/m^2

GIST FDG-PET Response

Pre-treatment
(2nd Nov 2010)

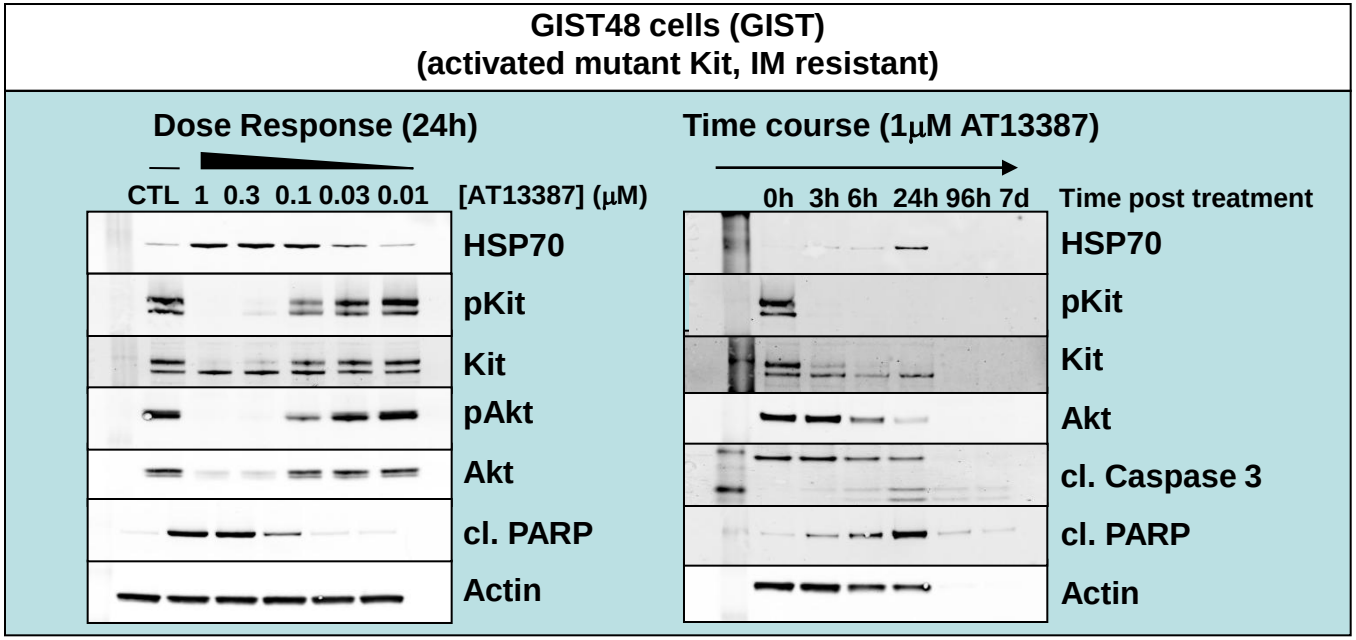


Post-treatment
(23rd Nov 2010)



220 mg/m^2 weekly

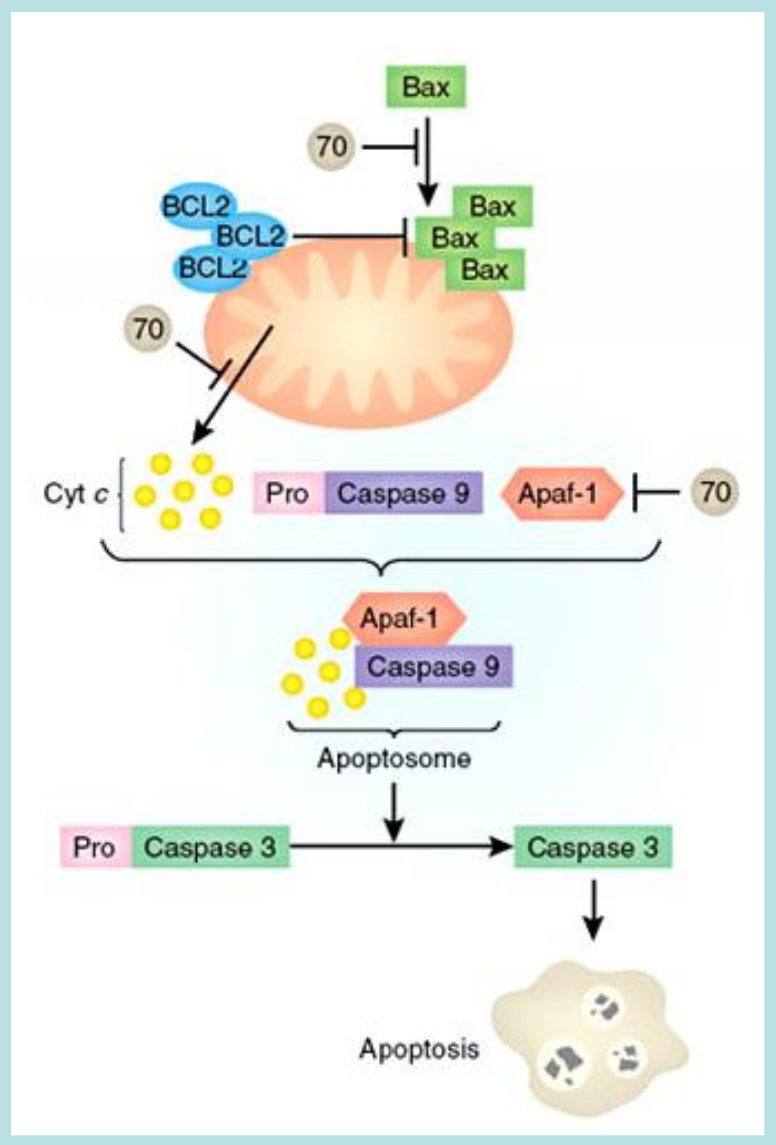
Preclinical Data to Support Phase 2 Trial in GIST

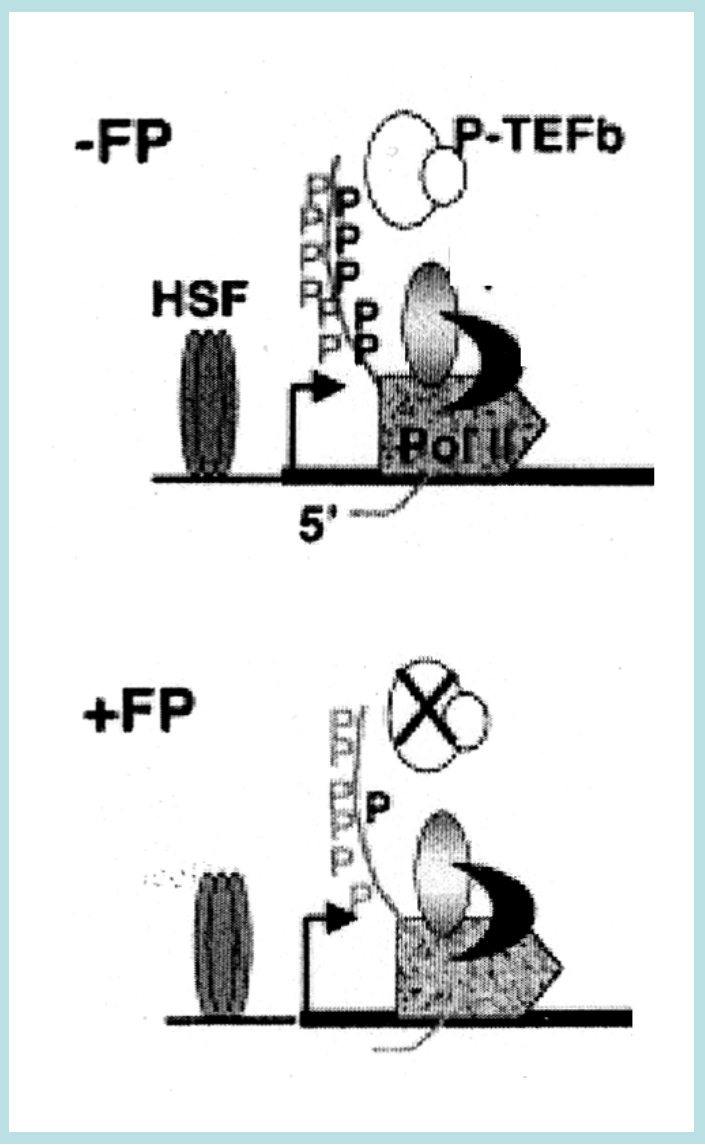


Sensitivity of GIST cell lines to AT13387

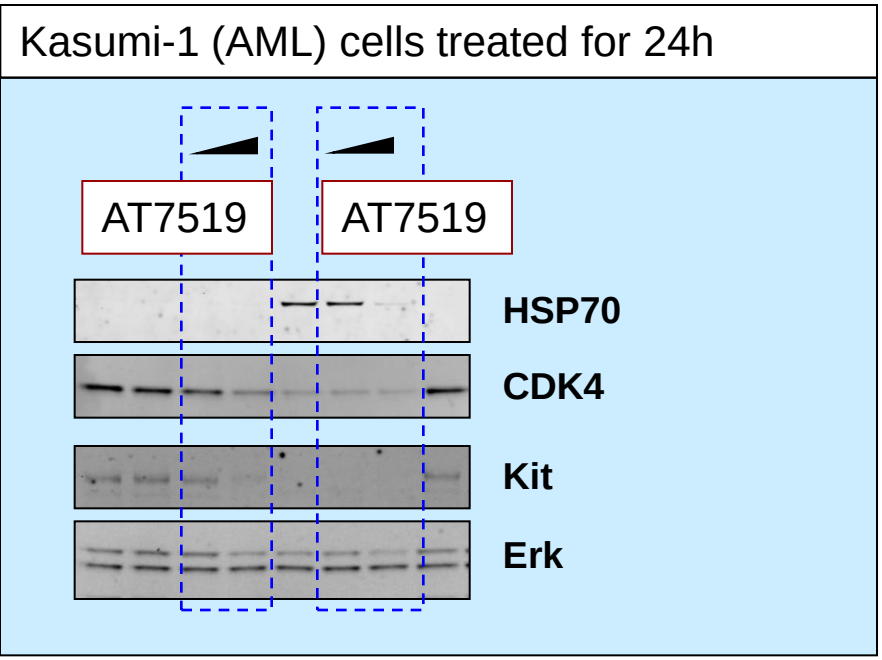
Cell Line (Derivation)	Mutation	Resistance Pharmacology	Antiproliferative IC_{50} (nM)			
			AT13387	17 AAG	Imatinib	Sunitinib
GIST-T1 (Treatment naïve)	KIT Exon 11	Imatinib sensitive	36	29	45	25
GIST-882 (Treatment naïve)	KIT Exon 13	Imatinib sensitive	72	130	180	69
GIST-430 (PD on imatinib)	KIT Exon 11 +V654A	Imatinib resistant	100	285	>1000	285
GIST-48B	KIT independent	Imatinib and sunitinib resistant	310	890	>1000	>1000
GIST 430B (17AAG-resistant)	NQO1 delete	17AAG-resistant	75	>1000	N/D	N/D

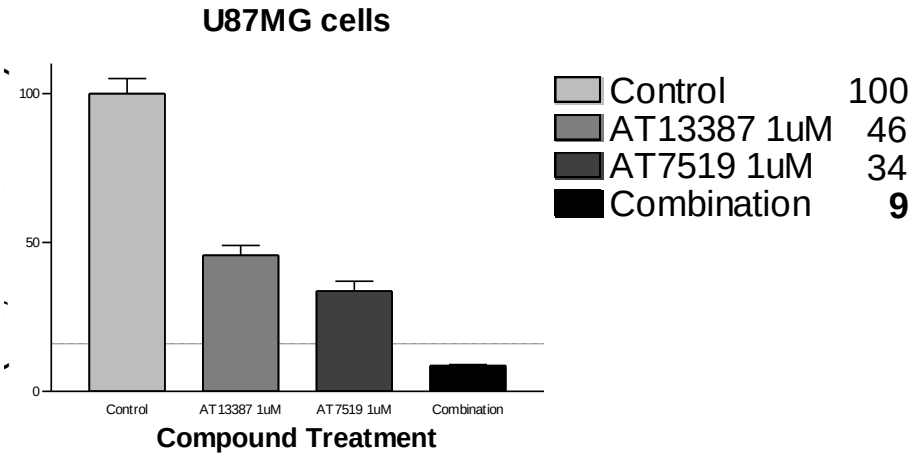
Hsp70 is an antiapoptotic protein; Transcriptional induction can be blocked by inhibition of cdk9





AT13387 and AT7519 combination





Kasumi-1 Proliferation IC_{50} : AT7519 300nM

Courtesy, John Lyons, Astex

AT13387 – Clinical Development Plan

Single agent trials

- Phase I (HER2 enriched) AT13387 twice weekly consecutive day schedule: 2 patients treated
- TKI-refractory NSCLC (weekly schedule planned)
- JAK-positive myeloproliferative disease

Combination trials

- With trastuzumab in HER2 amplified breast cancer
- With bortezomib in multiple myeloma
- With Ara-C in FLT3-driven AML
- With Cdk inhibitor
- With PARP inhibitor
- With MEK inhibitor in ocular melanoma

Conclusions

- Second generation Hsp90 inhibitors have greater *in vitro* potency than 17-AAG and improved *in vivo* efficacy in xenograft models.
- Preclinical PK demonstrate prolonged persistence in the tumor compartment, variable kinetics of client depletion. For some clients, including EGFR, more frequent dosing that once-weekly may be necessary.
- STA-9090 has activity in murine models of lung adenocarcinoma.
- STA-9090 and AT13387 are reasonably well tolerated in once-weekly and twice-weekly schedules.
- GI toxicity (diarrhea) is commone, generally grade 1 -2 and manageable with supportive care. Absence of severe hepatotoxicity. STA-9090 also notable for only minimal ocular toxicity.
- For STA-9090, single agent activity has been observed, especially in NSCLC with wild-type EGFR and wild-type KRas (genotypic analysis ongoing).
- Preclinical synergism with taxanes supports a plan for a Phase 2b/3 NSCLC trial of STA-9090 in combination with docetaxel.
- Preclinical data in GISTmodels support re-examination of this disease type with non-hepatotoxic Hsp90 inhibitors.
- Novel approaches are needed, including strategies that may deplete other chaperone proteins.

Acknowledgements

STA-9090

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AT13387

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