IMPAKT BREAST CANCER CONFERENCE

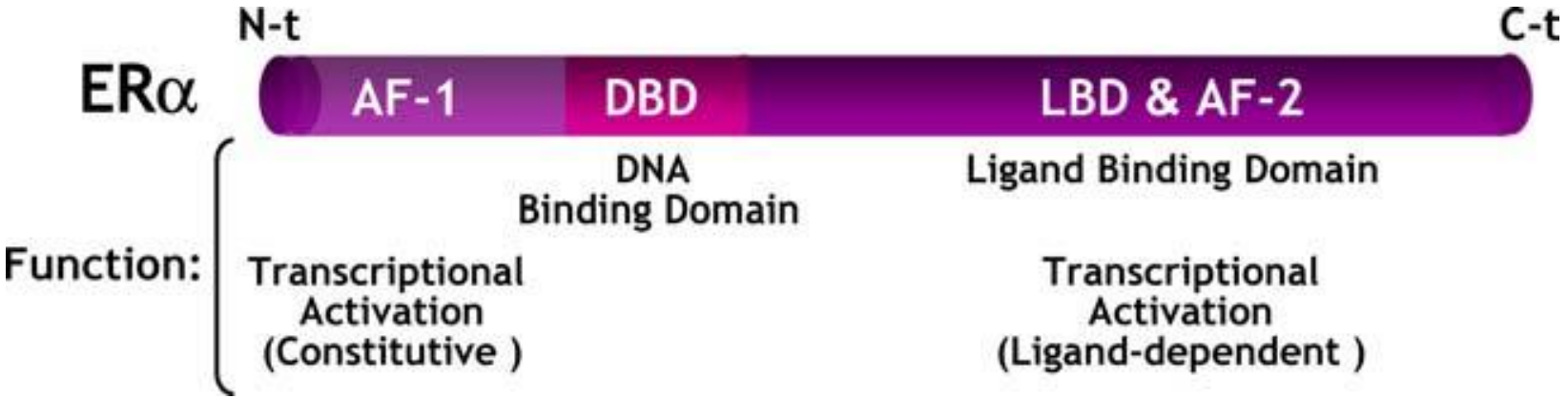
IMProving cAre and Knowledge through Translational research

Hormone Signaling

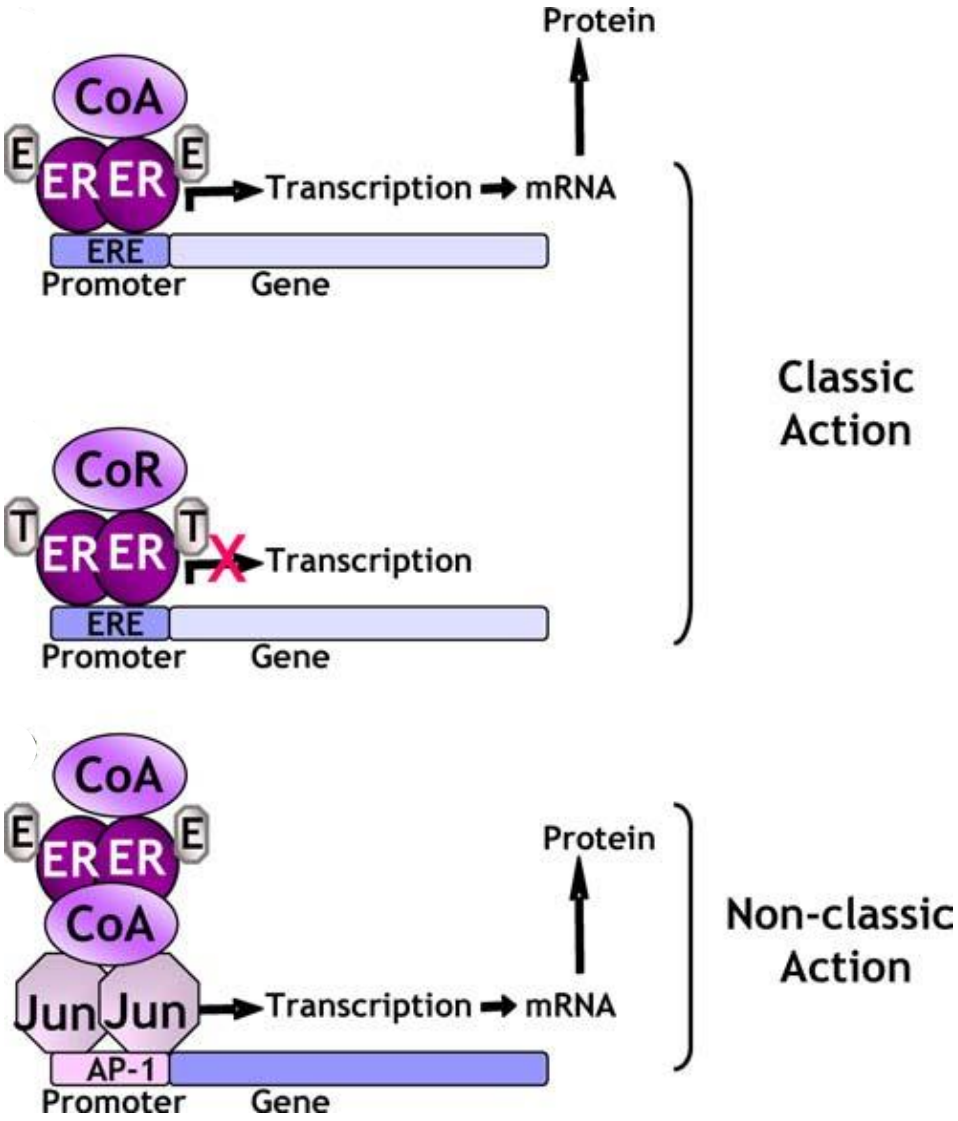
Grazia Arpino

University of Naples Federico II

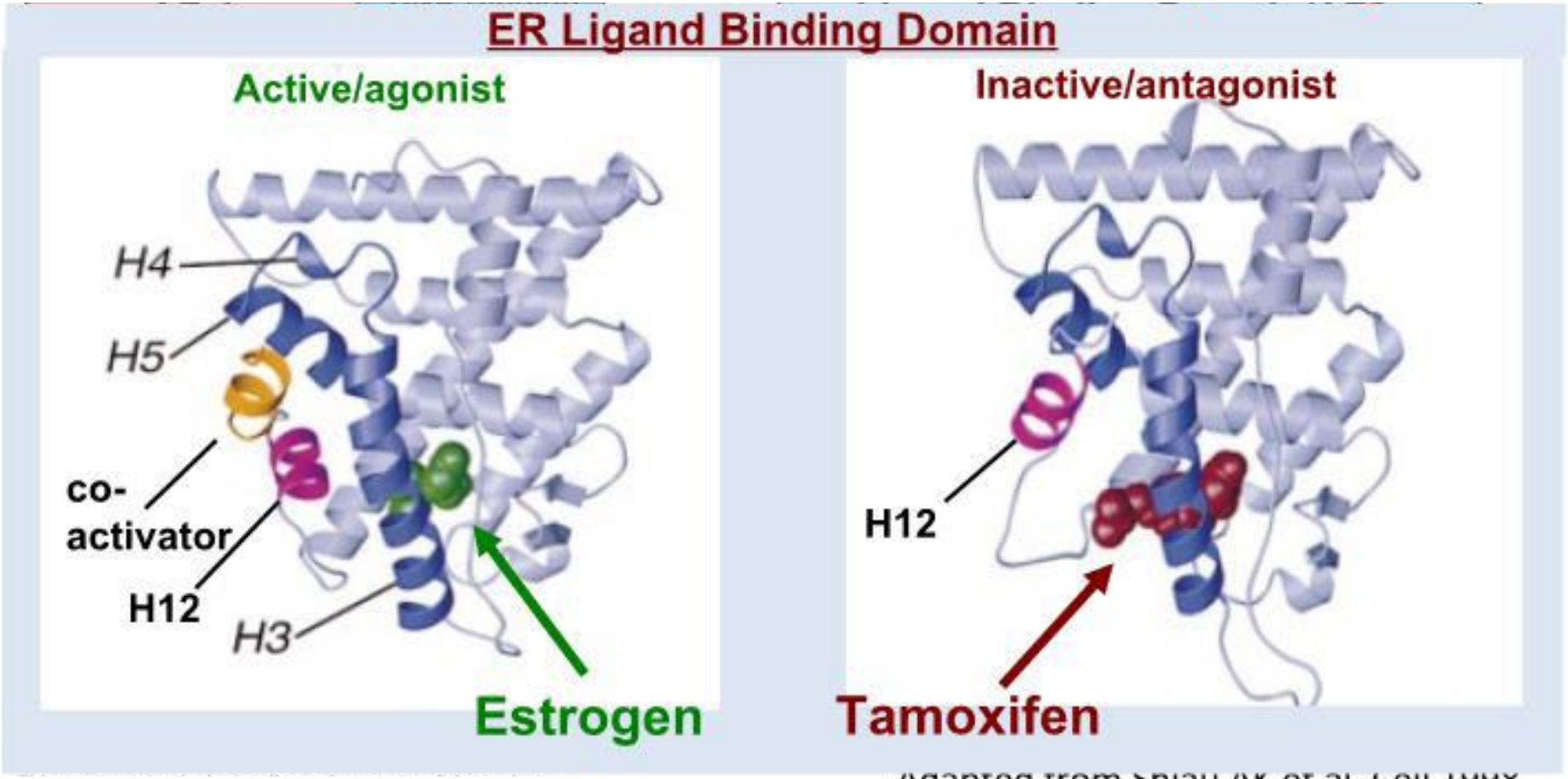
ER structure



ER Nuclear Genomic Activity

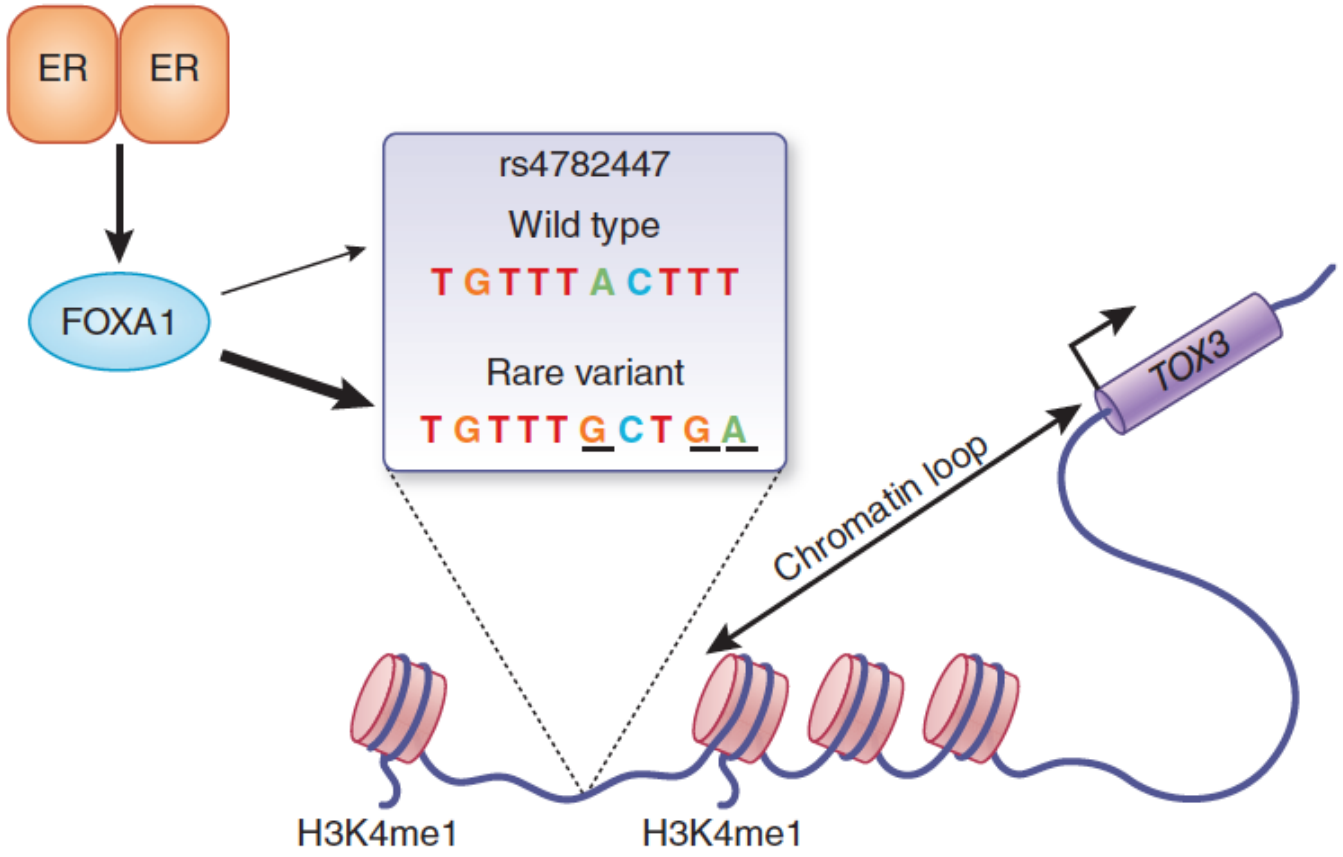


ER Ligand Binding Domain



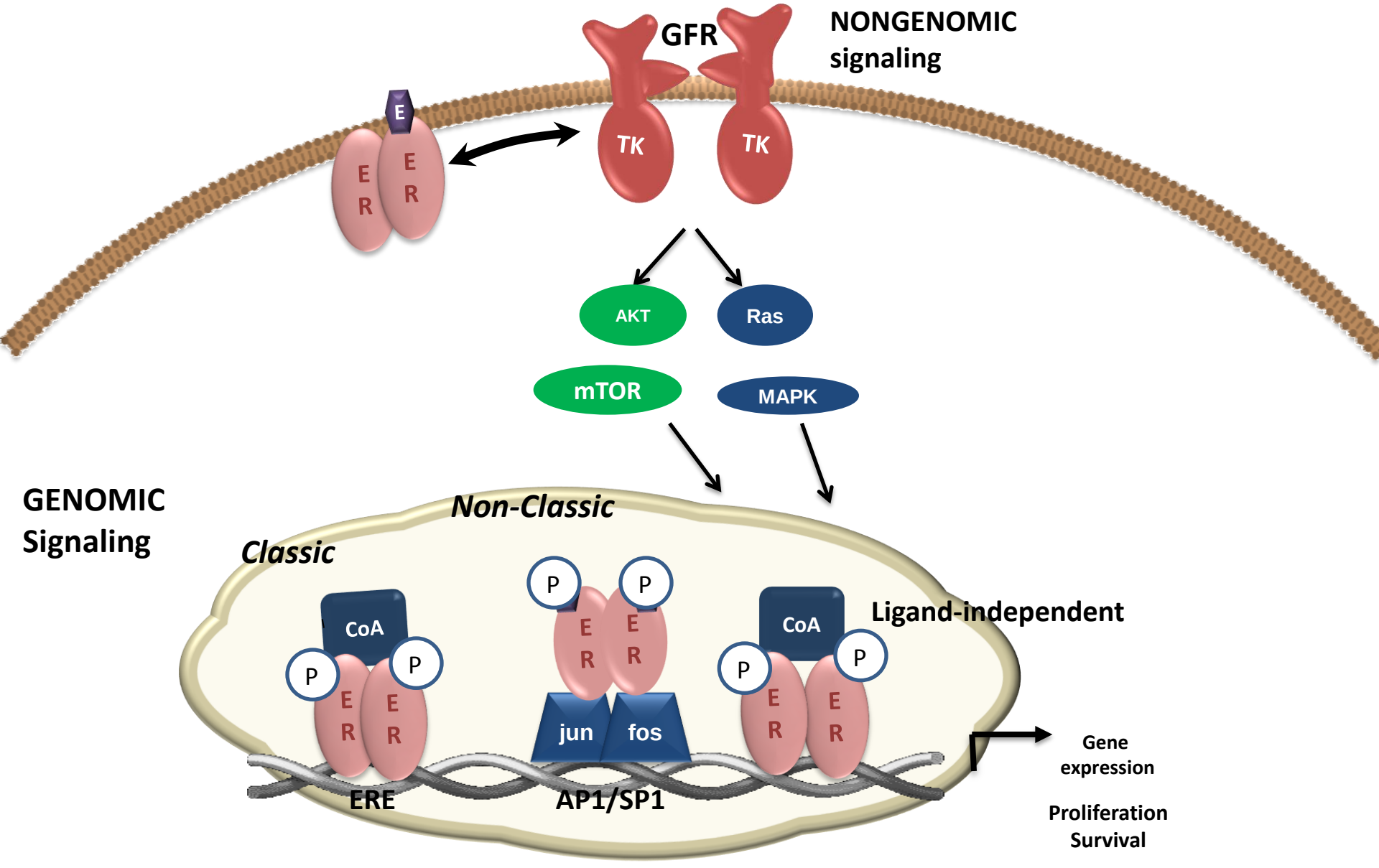
Adapted from Shiau AK et al, Cell 1998

ER Regulates Gene Trascription from a Distance And Needs Fox A1



Ross et al Nature 2012

ER activity is through genomic and non-genomic pathways

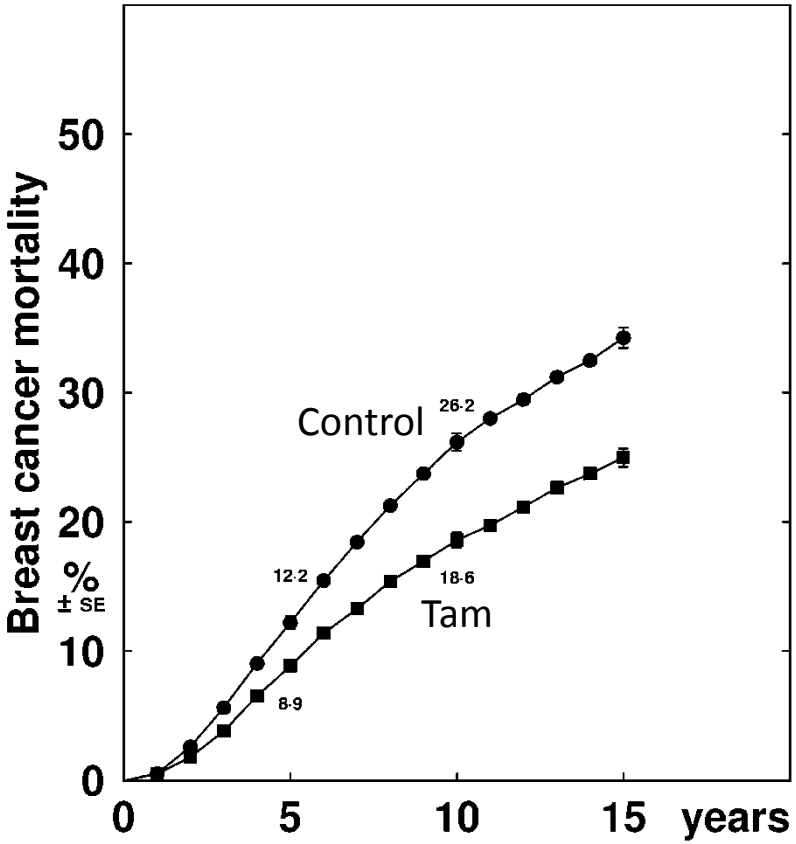


- Estrogen receptor is the target of endocrine treatment and is expressed in ~70% of breast cancers

- Estrogen receptor is the target of endocrine treatment and is expressed in ~70% of breast cancers

- Endocrine treatment, like Tamoxifen is, to date, the most effective targeted therapy developed for cancer

~5 years tamoxifen vs. Not, ER+ only
BREAST CANCER MORTALITY



Death rates (% / year: total rate – rate in women without recurrence) & logrank analyses

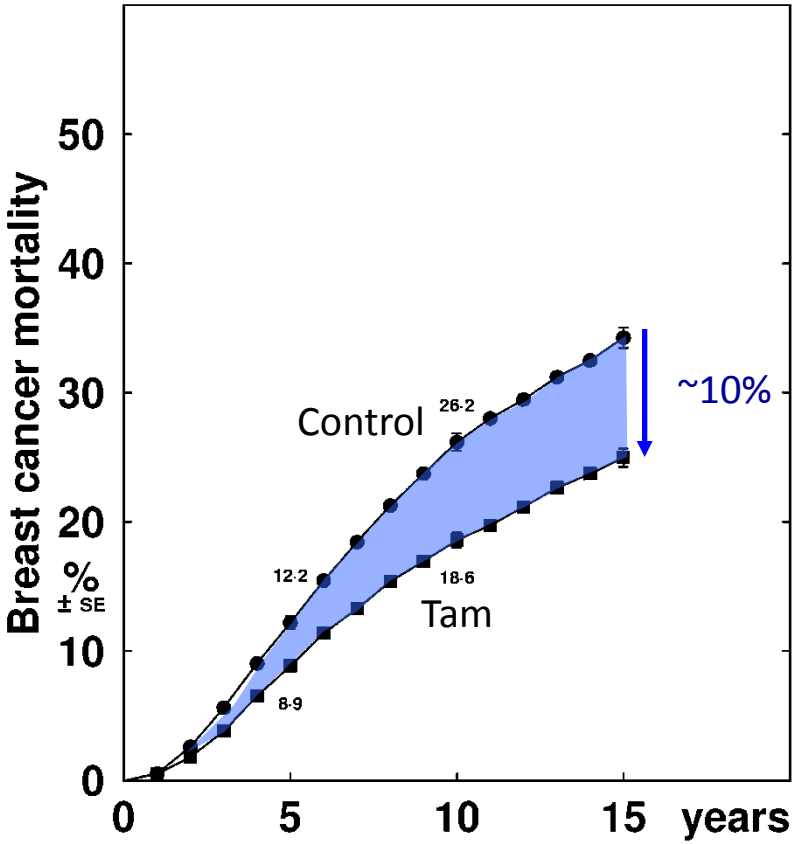
	Years 0 – 4	Years 5 – 9	Years 10 – 14	Year 15+
Tamoxifen	1·84 (429 / 23343)	2·35 (425 / 18107)	1·64 (197 / 12004)	1·55 (94 / 6050)
Control	2·55 (580 / 22754)	3·46 (585 / 16905)	2·35 (252 / 10734)	1·95 (102 / 5239)
Rate ratio, from	0·71 SE 0·06	0·66 SE 0·05	0·70 SE 0·08	0·92 SE 0·14
(O–E) / V	–80·7 / 236·3	–99·3 / 235·9	–36·5 / 103·9	–3·7 / 44·5

- Estrogen receptor is the target of endocrine treatment and is expressed in ~70% of breast cancers

- Endocrine treatment, like Tamoxifen is, to date, the most effective targeted therapy developed for cancer

EBCTGC, Lancet 2011

~5 years tamoxifen vs. Not, ER+ only
BREAST CANCER MORTALITY



Death rates (% / year: total rate – rate in women without recurrence) & logrank analyses

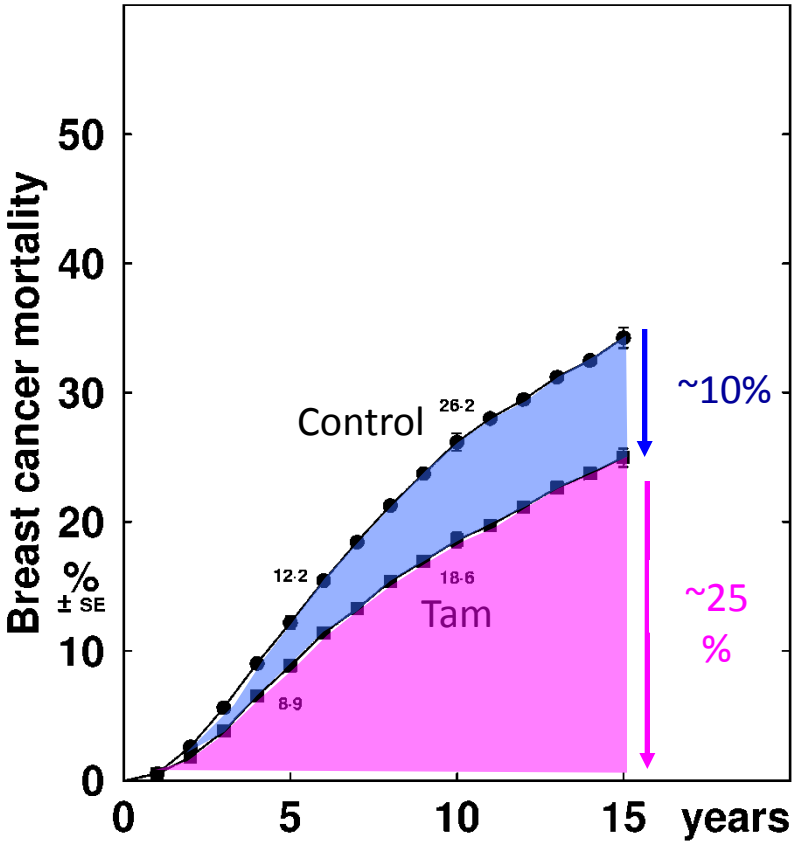
	Years 0 – 4	Years 5 – 9	Years 10 – 14	Year 15+
Tamoxifen	1·84 (429 / 23343)	2·35 (425 / 18107)	1·64 (197 / 12004)	1·55 (94 / 6050)
Control	2·55 (580 / 22754)	3·46 (585 / 16905)	2·35 (252 / 10734)	1·95 (102 / 5239)
Rate ratio, from	0·71 SE 0·06	0·66 SE 0·05	0·70 SE 0·08	0·92 SE 0·14
(O–E) / V	–80·7 / 236·3	–99·3 / 235·9	–36·5 / 103·9	–3·7 / 44·5

- Estrogen receptor is the target of endocrine treatment and is expressed in ~70% of breast cancers

- Endocrine treatment, like Tamoxifen is, to date, the most effective targeted therapy developed for cancer

EBCTGC, Lancet 2011

~5 years tamoxifen vs. Not, ER+ only
BREAST CANCER MORTALITY



Death rates (% / year: total rate – rate in women without recurrence) & logrank analyses

	Years 0 – 4	Years 5 – 9	Years 10 – 14	Year 15+
Tamoxifen	1·84 (429 / 23343)	2·35 (425 / 18107)	1·64 (197 / 12004)	1·55 (94 / 6050)
Control	2·55 (580 / 22754)	3·46 (585 / 16905)	2·35 (252 / 10734)	1·95 (102 / 5239)
Rate ratio, from	0·71 SE 0·06	0·66 SE 0·05	0·70 SE 0·08	0·92 SE 0·14
(O–E) / V	–80·7 / 236·3	–99·3 / 235·9	–36·5 / 103·9	–3·7 / 44·5

- Estrogen receptor is the target of endocrine treatment and is expressed in ~70% of breast cancers

- Endocrine treatment, like Tamoxifen is, to date, the most effective targeted therapy developed for cancer

- However, almost one quarter of the patients develop resistance (i.e. mestatic disease)

EBCTGC, Lancet 2011

Major Problem
(Endocrine Therapy Resistance)

- ESR1 Pathway
- Signal Transduction Pathways
- Rb Pathway

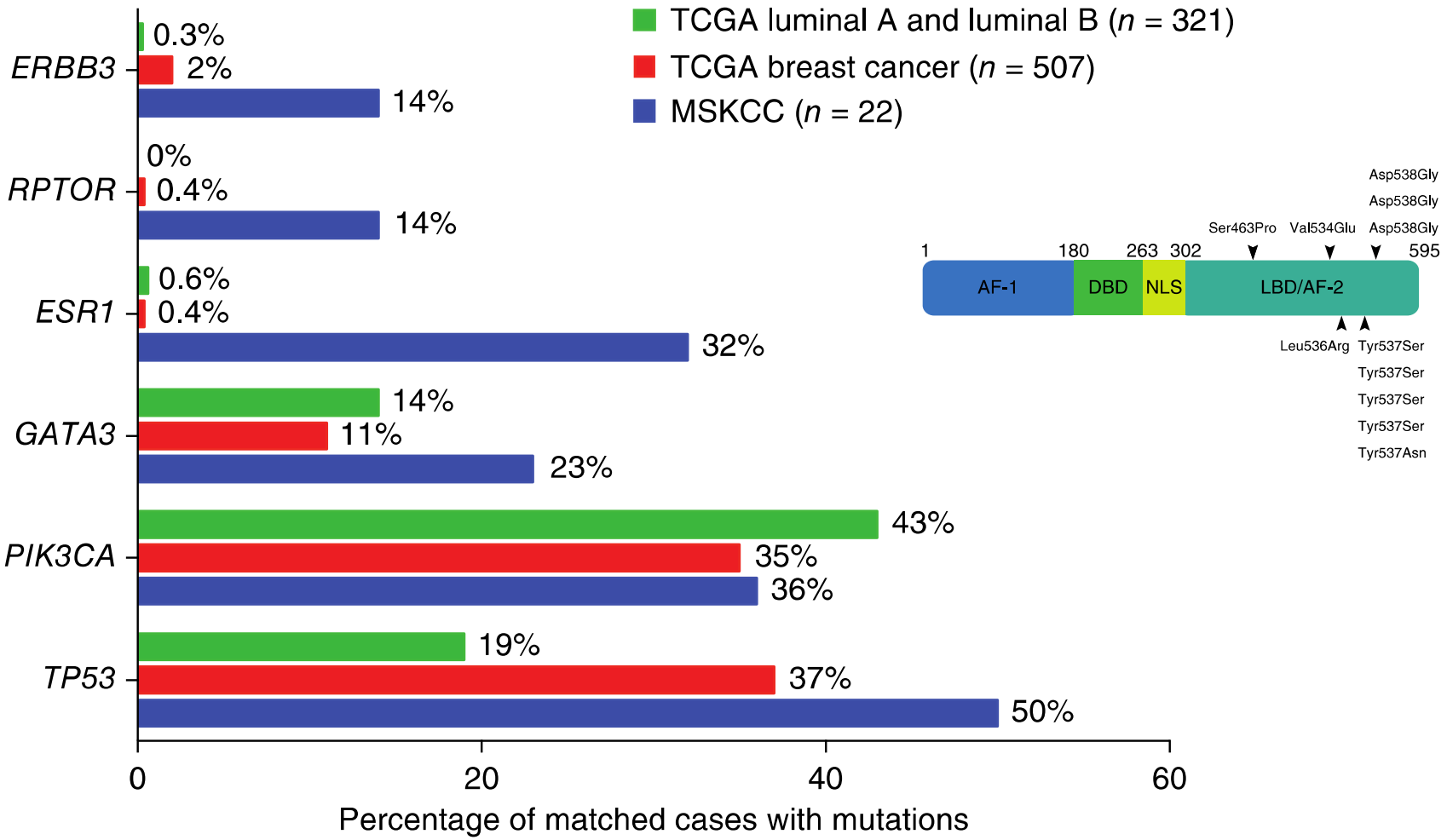
Hormone Receptor Pathways (ESR1)

- Activating ESR1 mutations in hormone resistant breast cancer

Robinson Nature Genetics 2013

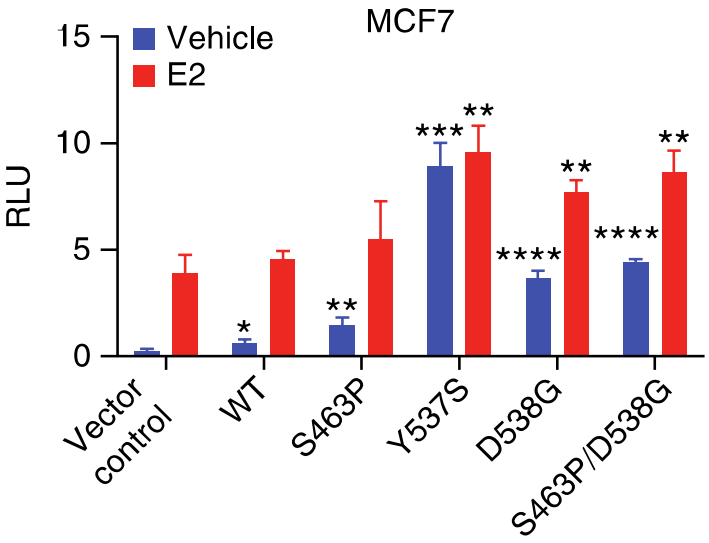
- ESR1 ligand binding domain mutations in hormone resistant breast cancer

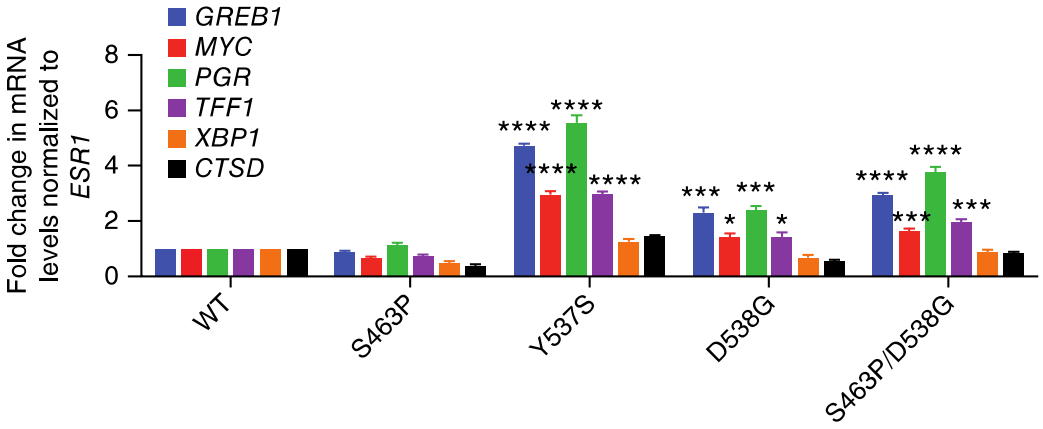
Toy Nature Genetics 2013



Toy et al. Nature Genetics 2013

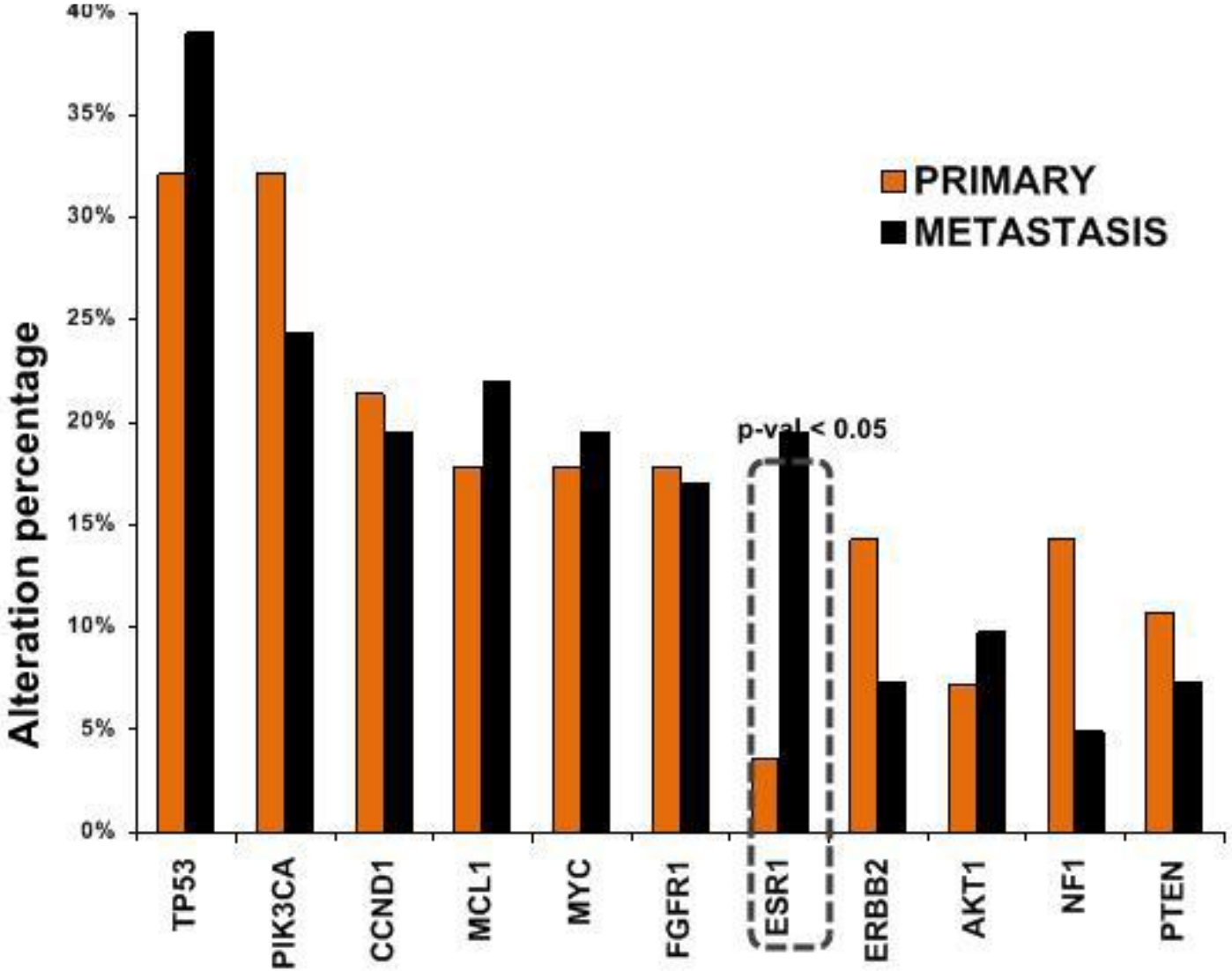
ER LBD Mutants Demonstrate Elevated Activity in the Absence of Hormone Stimulation





Toy et al. Nature Genetics 2013

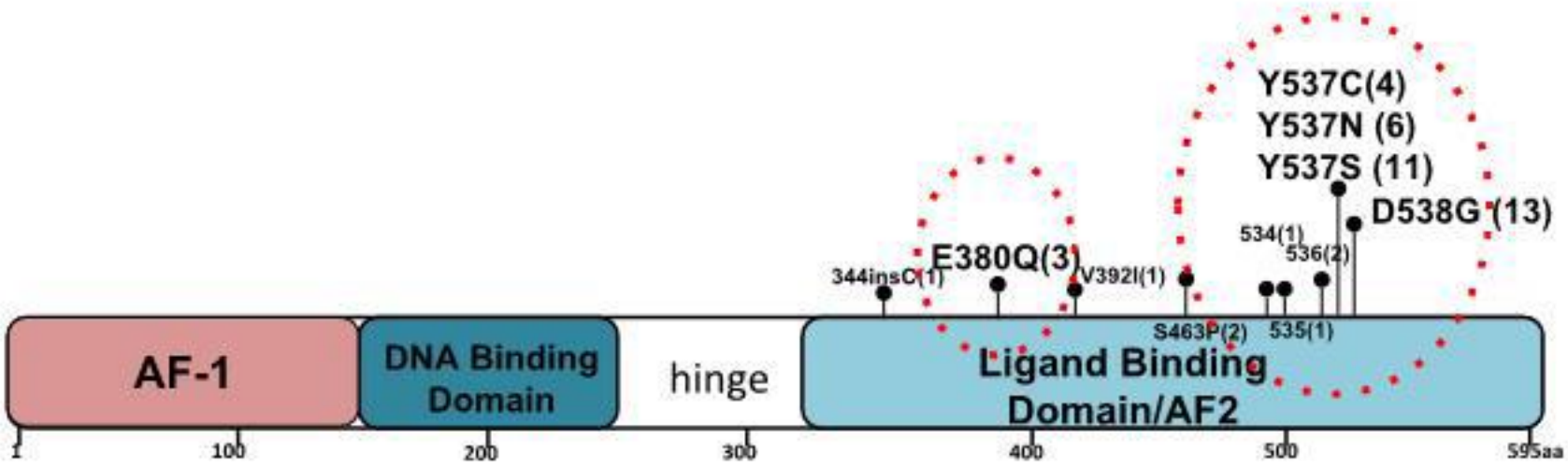
Genomic Alterations in Primary vs. Metastatic ER+ Tumors

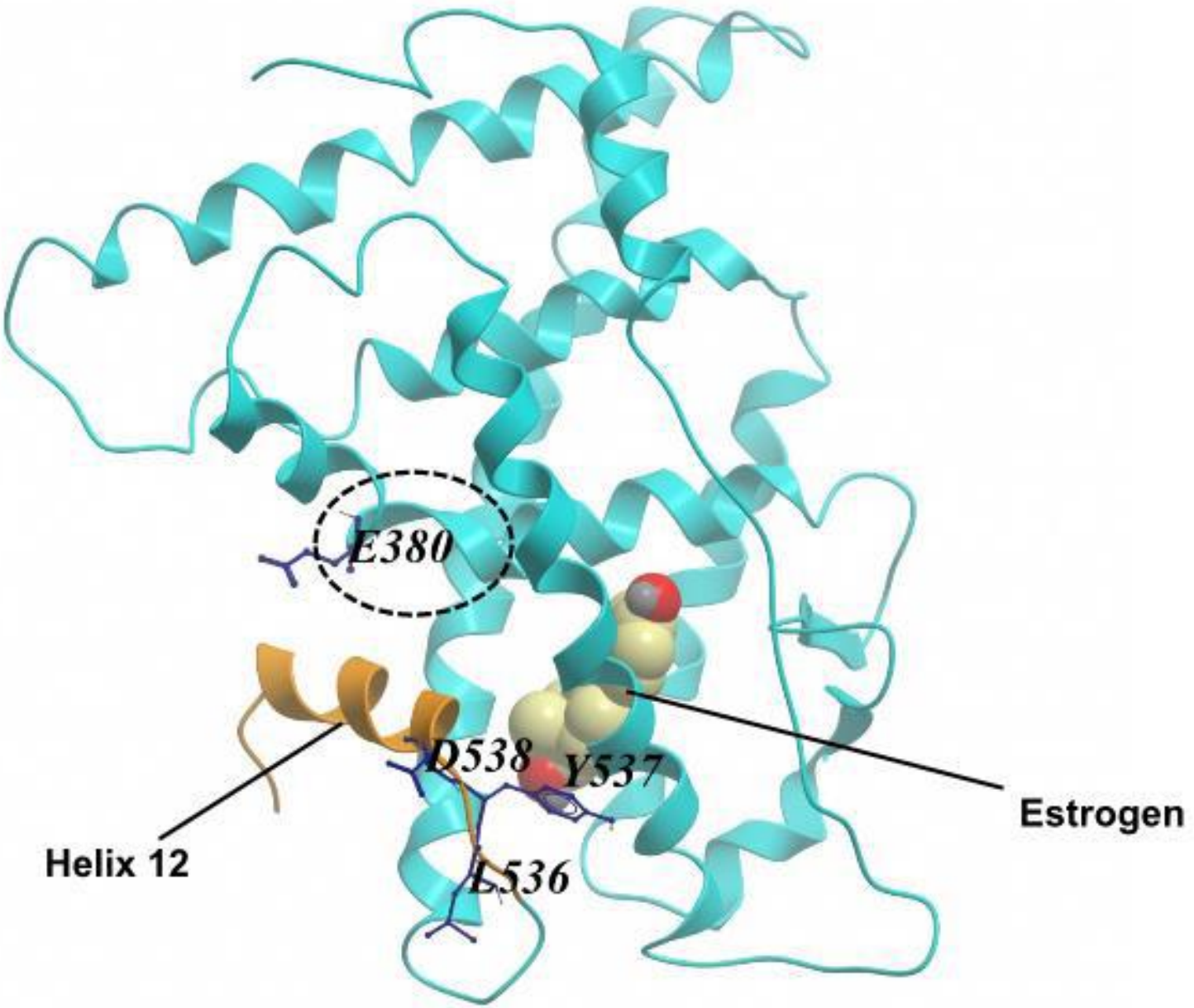


Data from 134 ER-positive: 58 primary breast cancers and 76 metastatic samples.

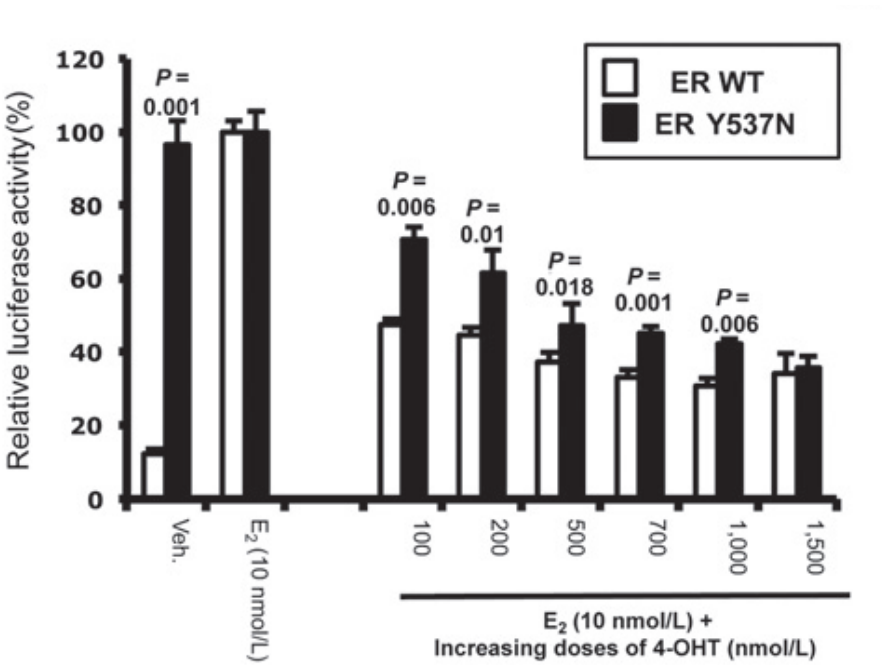
Jeselsohn et al, CCR 2014

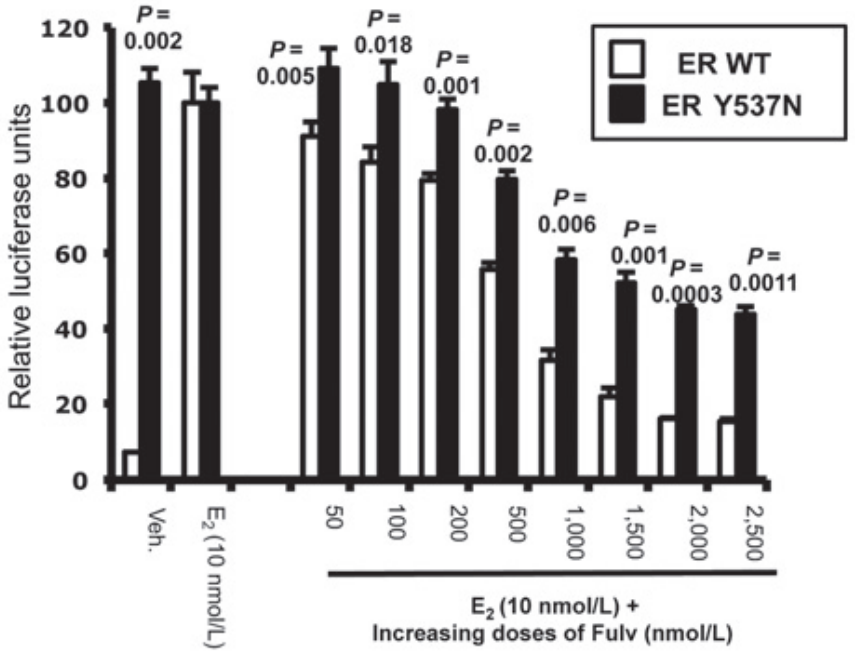
Summary of all ER mutations




E380
D538
Y537
L536
Helix 12
Estrogen

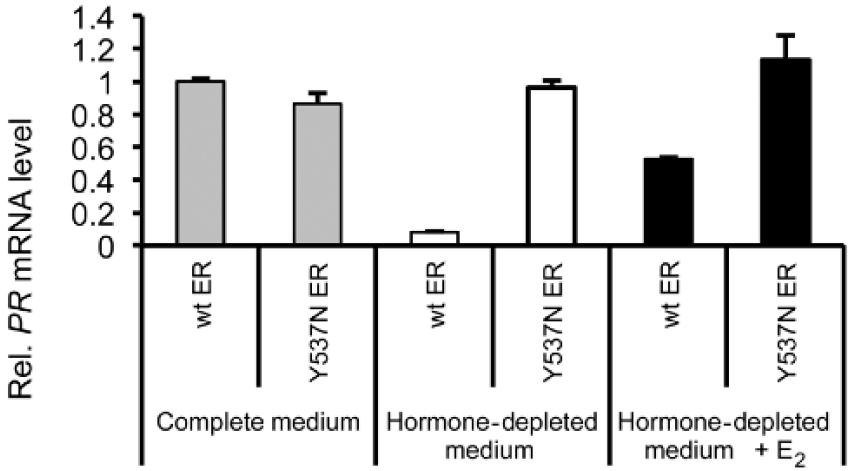
Mutant ER Confers Relative Resistance to Cell Growth Response to Tamoxifen and Fulvestrant

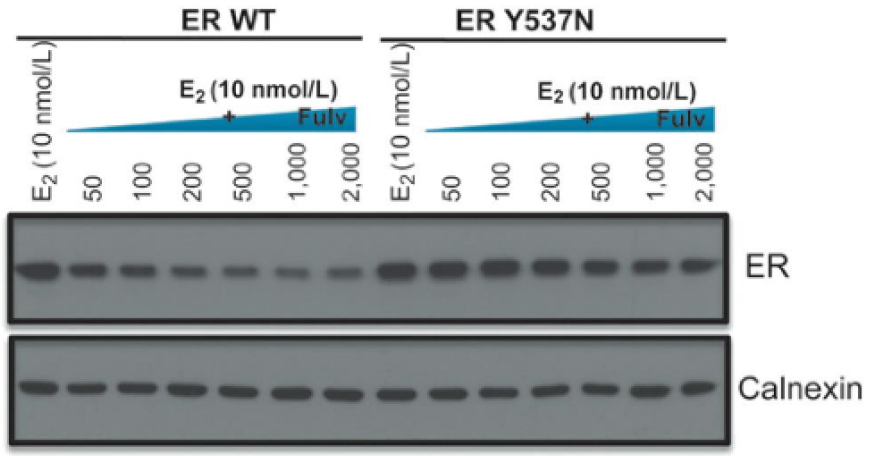




Jeselsohn et al, CCR 2014

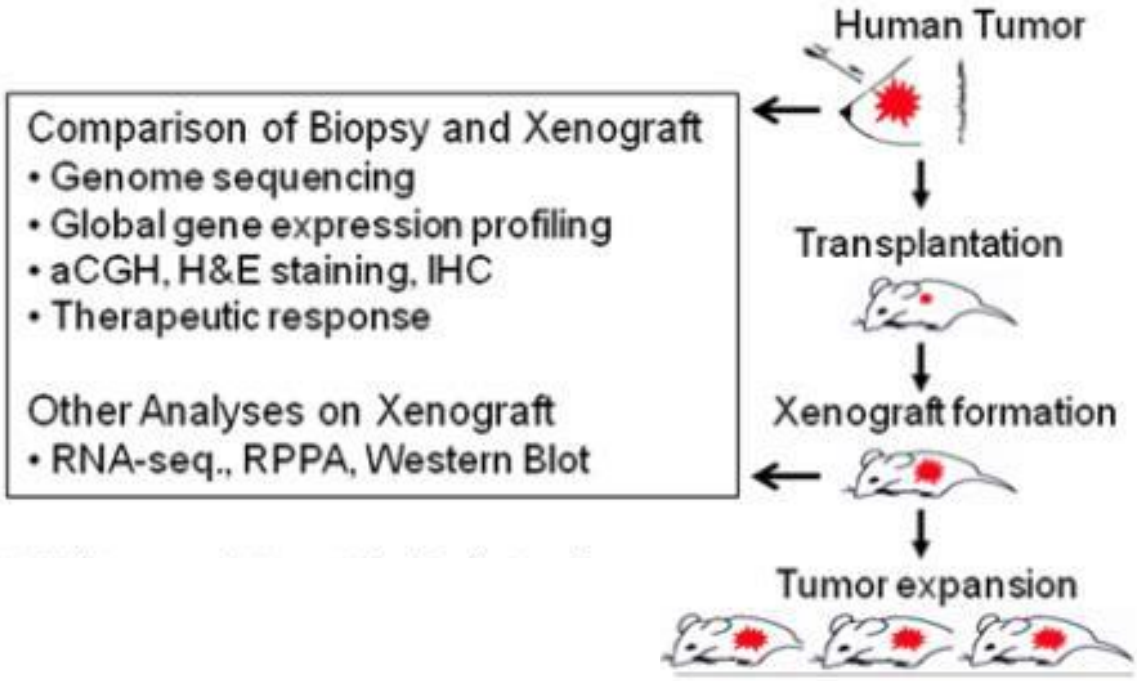
Mutant ER Constitutively Activates the Trascription of Endogenous ER dependent genes and is relatively Resistant to Fulvestrant Induced Degradation





Jeselsohn et al, CCR 2014

Patient-Derived Xenograft (PDX) models



Shao, SABCS 2013

Ligand-binding domain mutations are frequent in aromatase inhibitor-resistant breast cancer

*Y537N: an activating ESR1 mutation described by Zhang et al in 1997

Metastatic samples (**22%**):

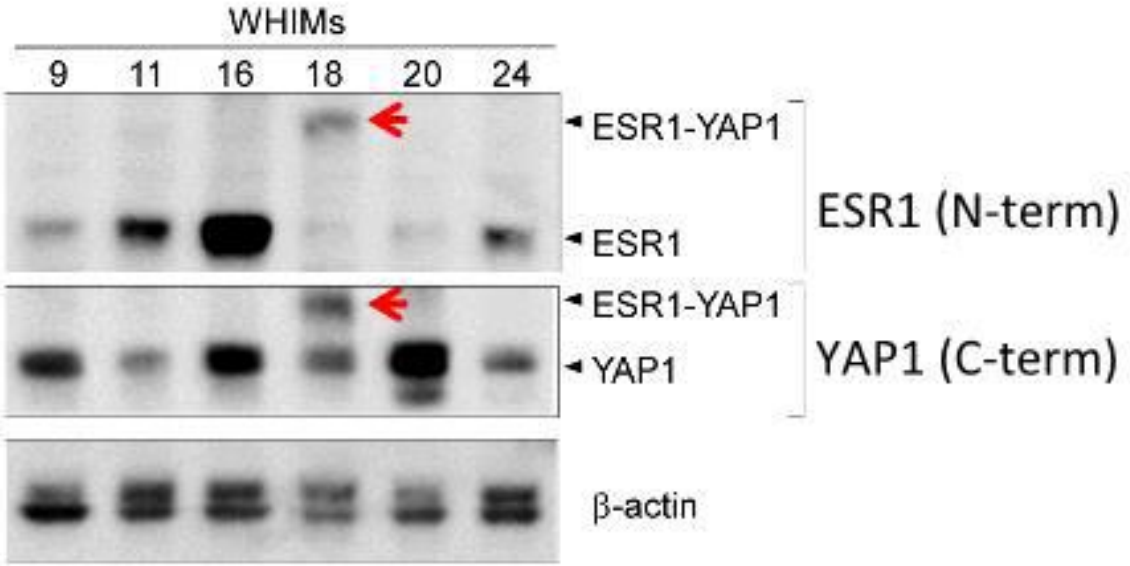
- 6 of 11 (55%) by Robinson et al, 2013
- 9 of 36 (25%) by Toy et al, 2013
- 5 of 44 (11%) in BOLERO Trial, 2013

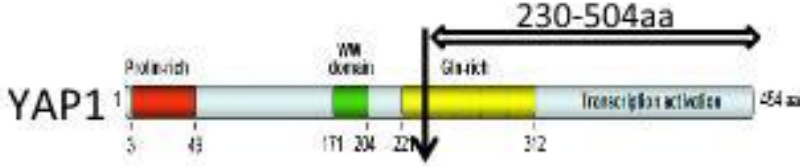
Primary Samples (<1%):

- 6 of 183 (3%) in BOLERO Trial
- 0 of 46 (0%) by Ellis et al., 2012
- 0 of >500 (0%) in TCGA

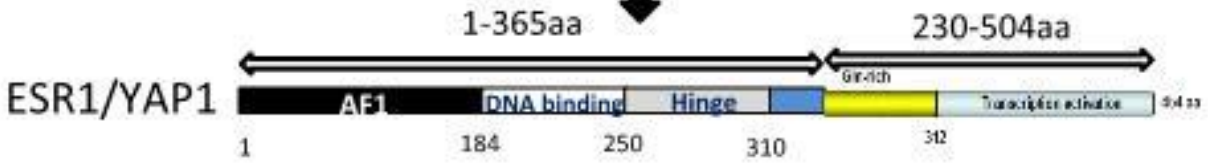
Shao et al SABCS 2013

ESR1/YAP1 fusion



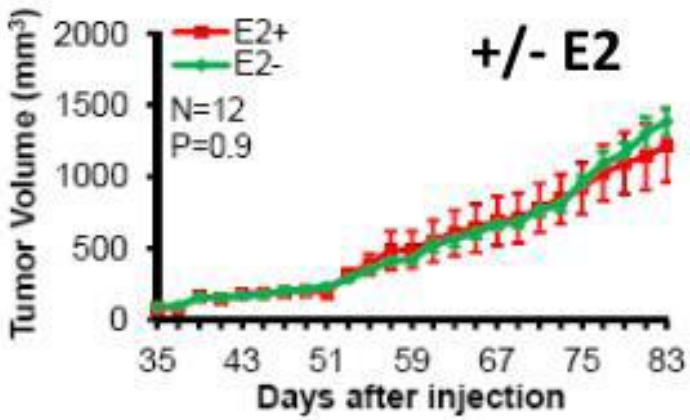


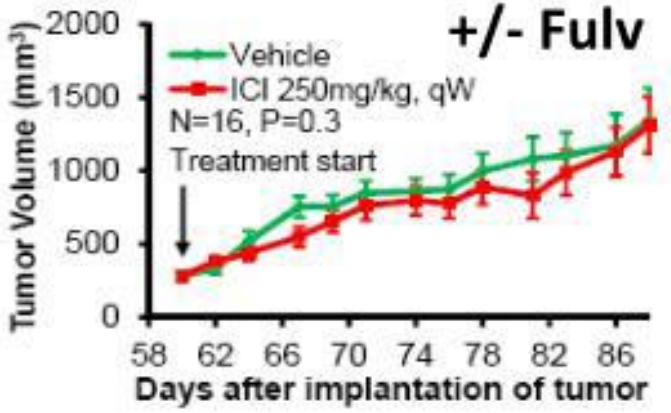
Balanced reciprocal translocation



Li et al. Cell Reports, 2013 Sep 26;4(6):1116-30

ESR1/YAP1 associates with estradiol-independent and fulvestrant-resistant tumor growth





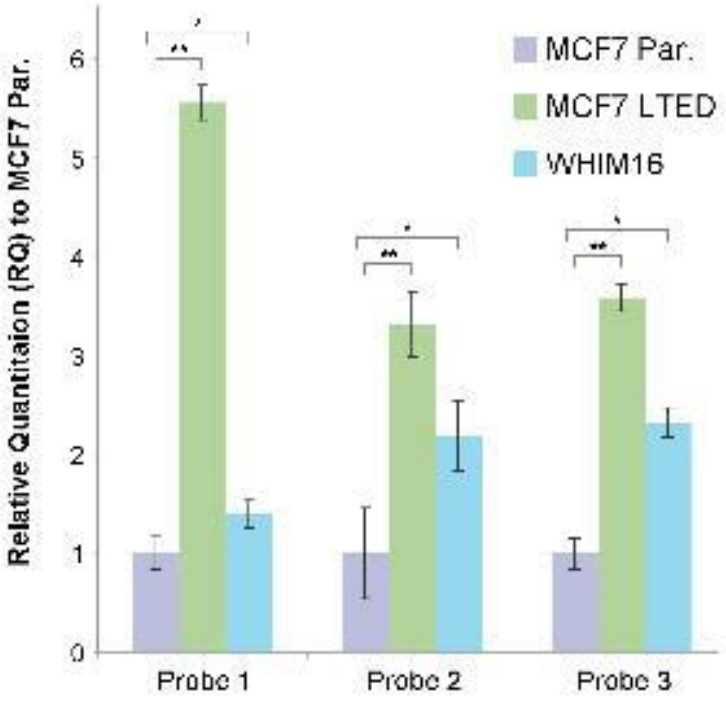
Gene Traslocations cannot be treated with classic endocrine therapies and will require alternative therapies

Li et al. Cell Reports, 2013 Sep 26;4(6):1116-30

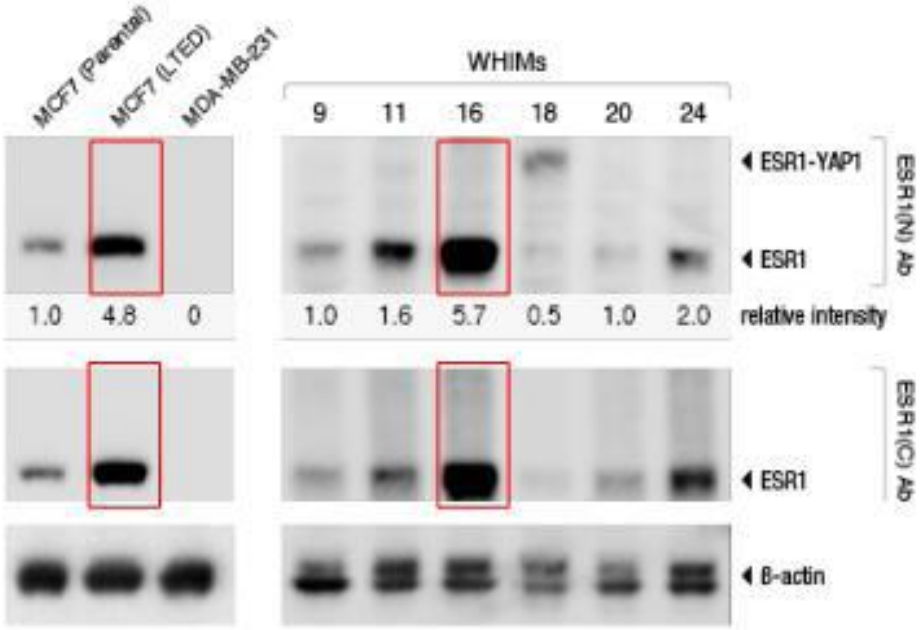
Courtesy of Dr Shao

ESR1 gene amplification causes high-level ESR1 protein expression

Q-PCR on genomic DNA

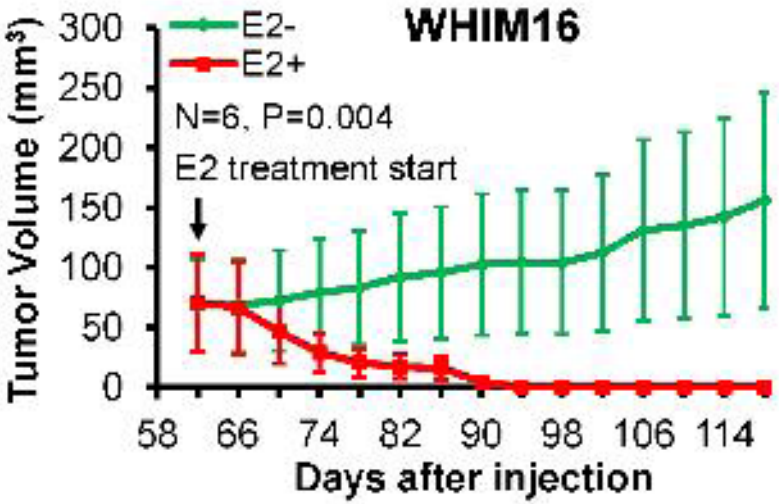


Western blots

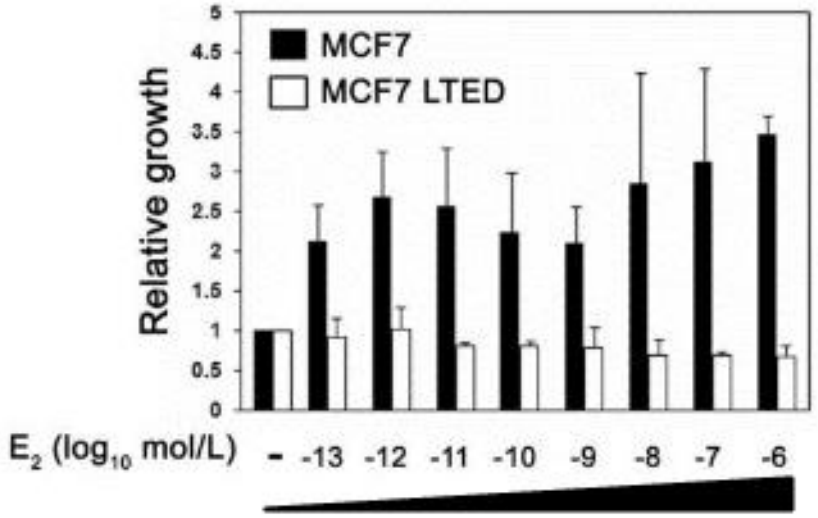


Li et al. Cell Reports, 2013 Sep 26;4(6):1116-30

ESR1 gene amplification is associated with the paradoxical antitumor effect of estradiol



Li et al. Cell Reports , 2013 Sep 26;4(6):1116-30



Sanchez et al., Breast Cancer Res. 2011 Mar 1;13(2):R21

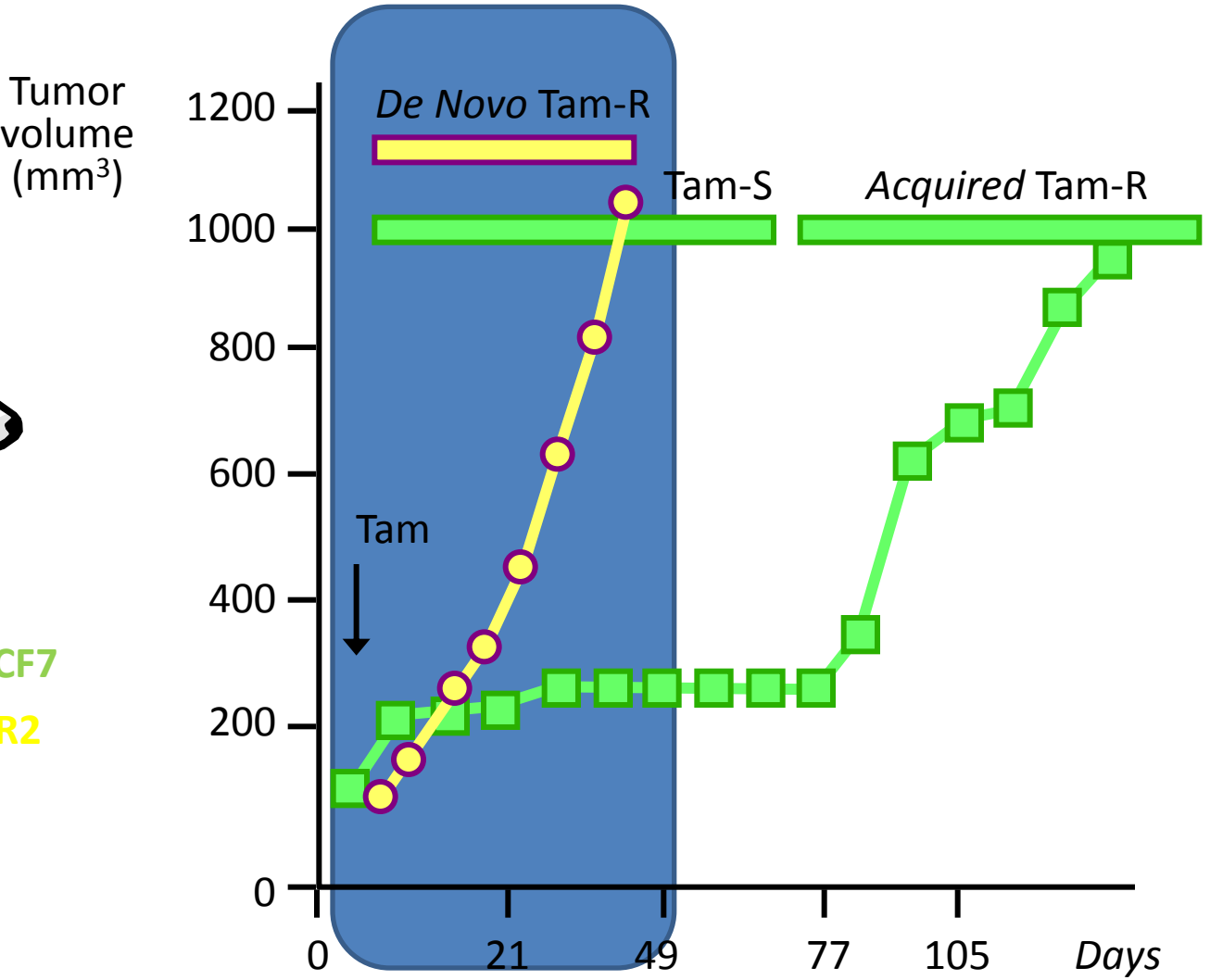
- ESR1 gene amplification may underline the “Haddow’s paradox”: the antitumor effect of estrogenic compounds
- ESR1 gene amplification may be an acquired resistance to long term hormone deprivation
- Both estradiol and anti-estrogens may be effective in treating tumors harboring ESR1 gene amplification

Signal Transduction Pathways

- De Novo Resistance: mostly in HER2-pos/ER-pos BC

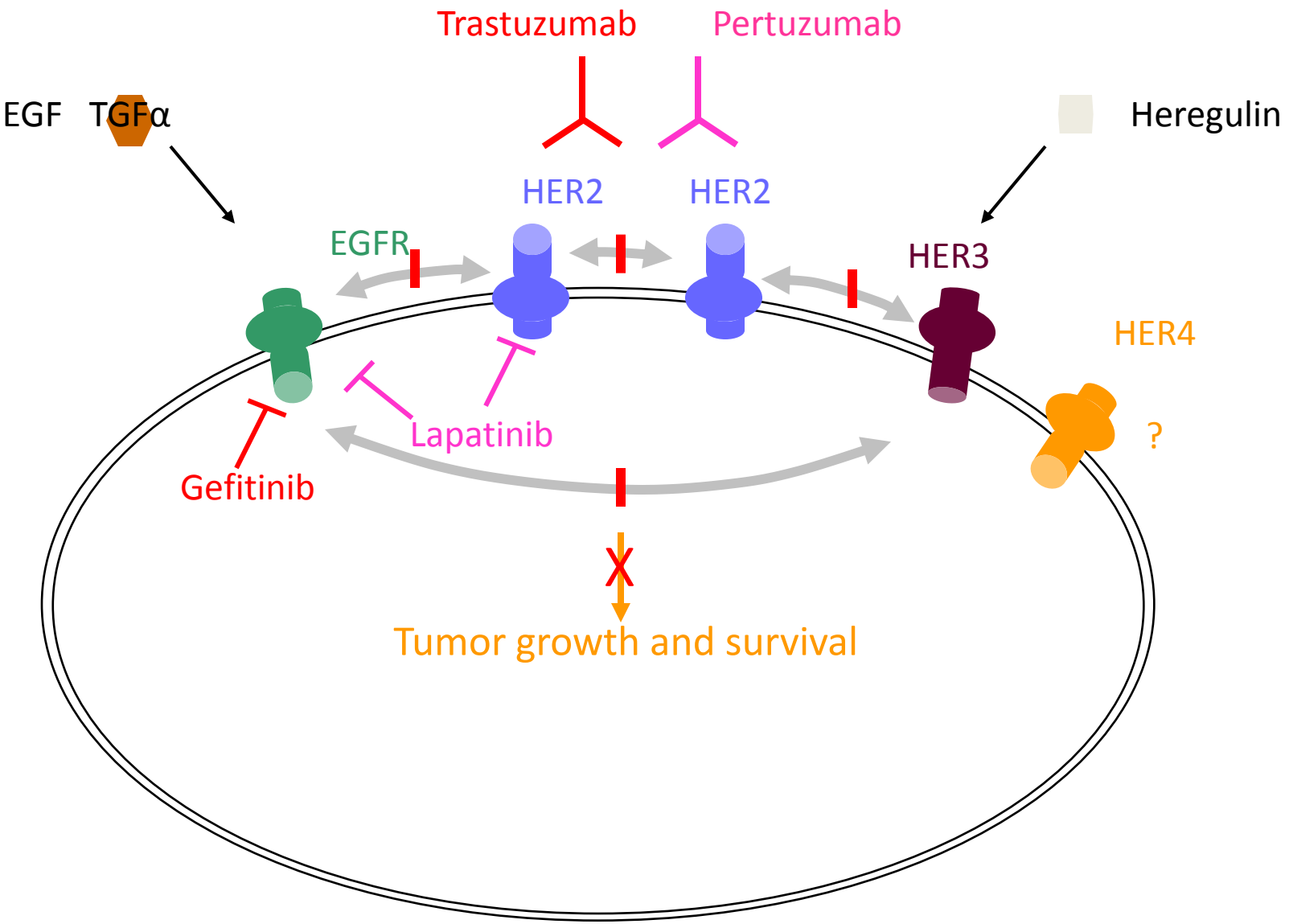
- Acquired Resistance: mostly in HER2-neg/ER-pos BC

In Vivo Model of Tamoxifen Resistance

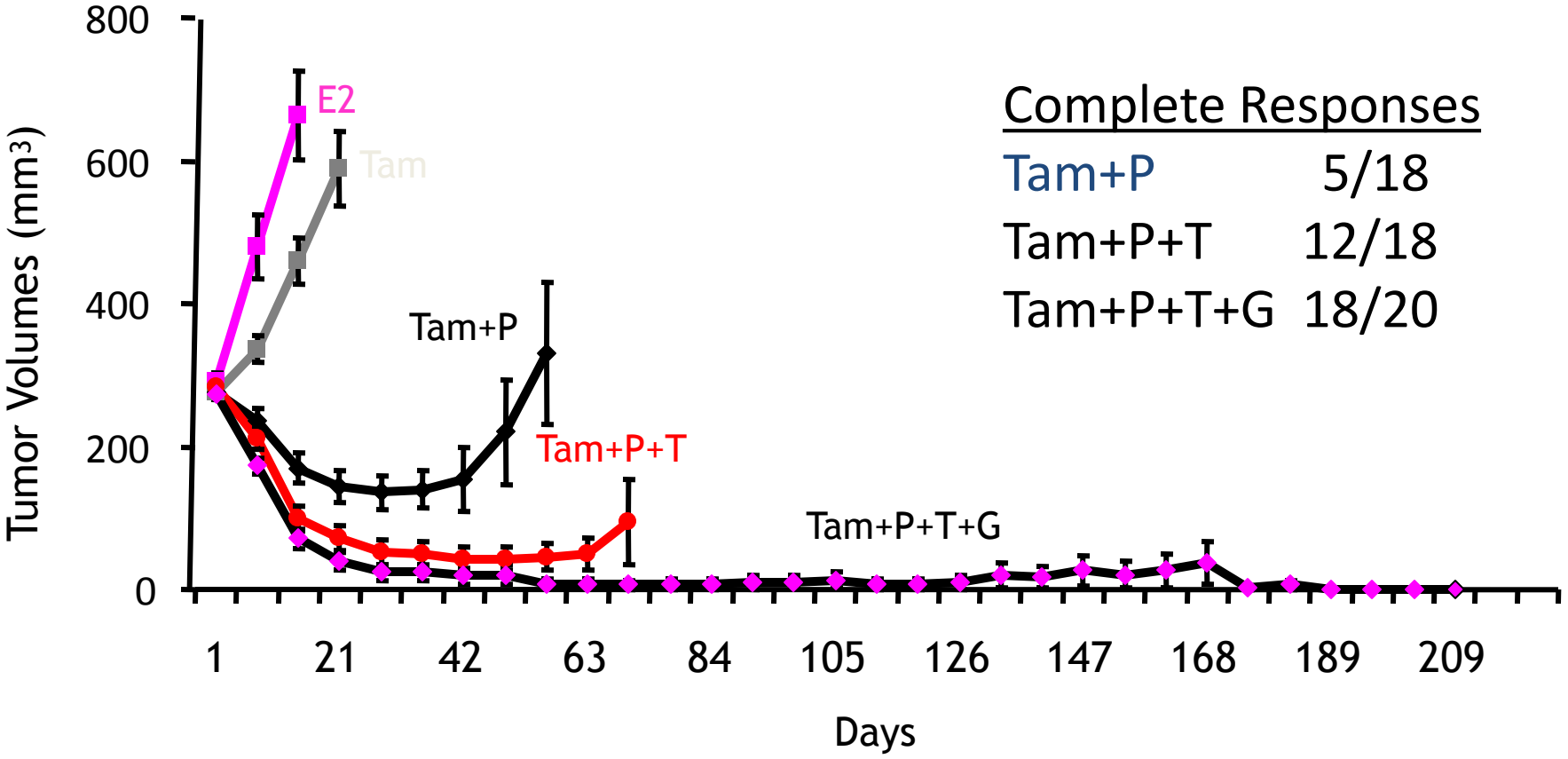


Osborne et al, JNCI 1994

HER Family Inhibitors



Effect of HER Family Inhibitors on Tam-Stimulated Growth

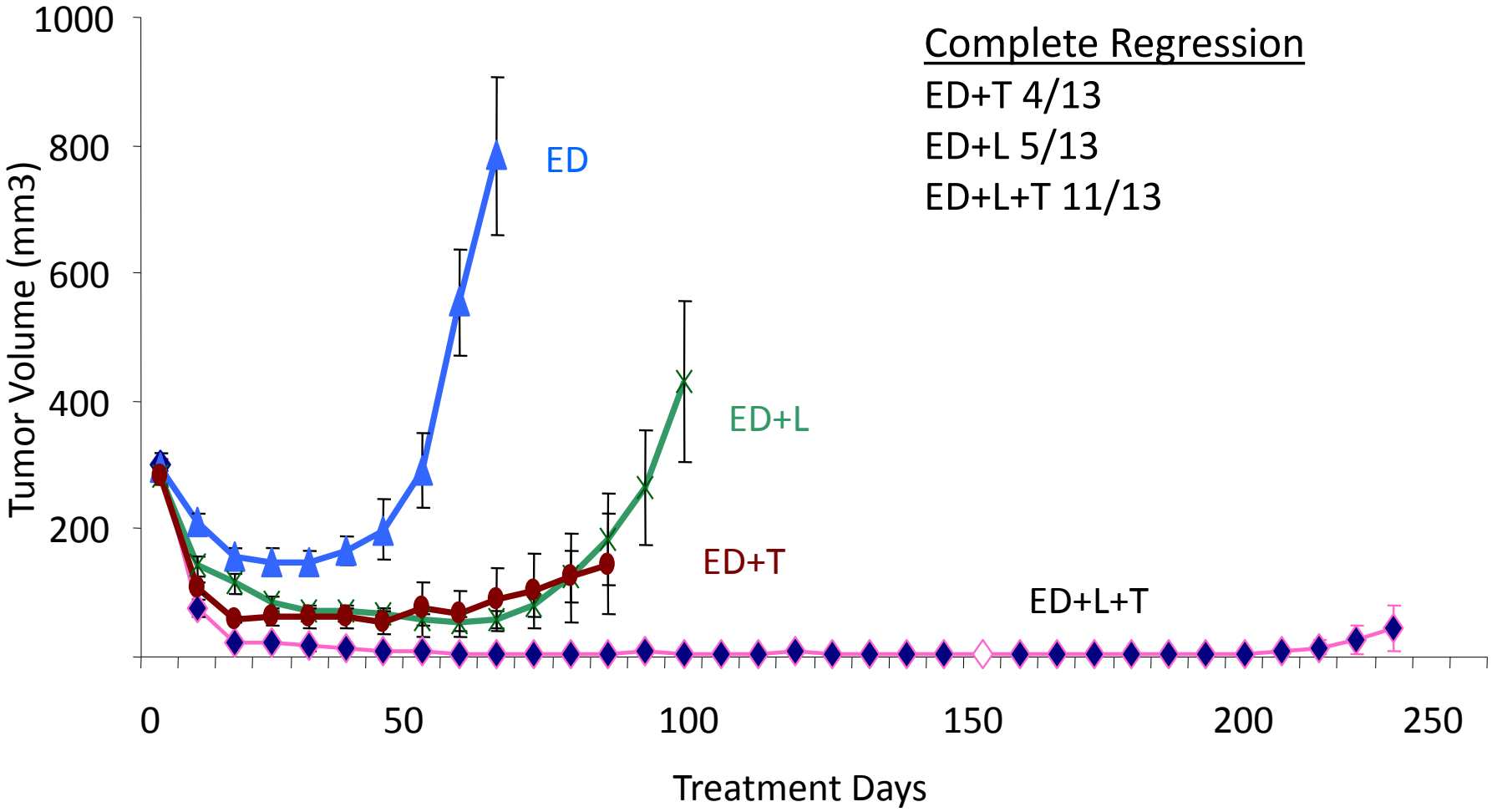


Complete Responses

Tam+P	5/18
Tam+P+T	12/18
Tam+P+T+G	18/20

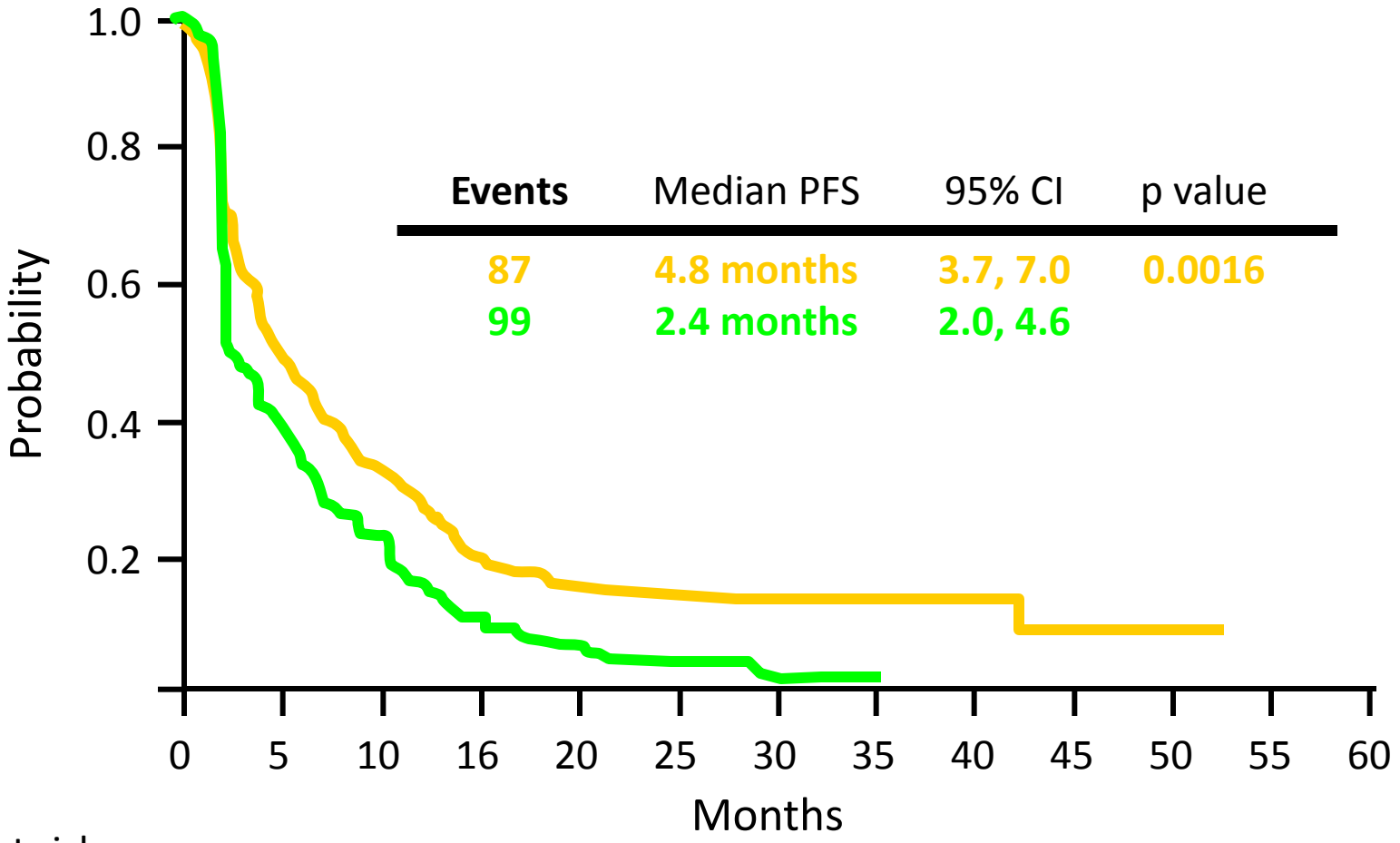
Arpino, JNCI 2007

Effect of HER Family Inhibitors on Estrogen Deprivation



Rimawi CCR 2011

TanDEM Progression-free Survival

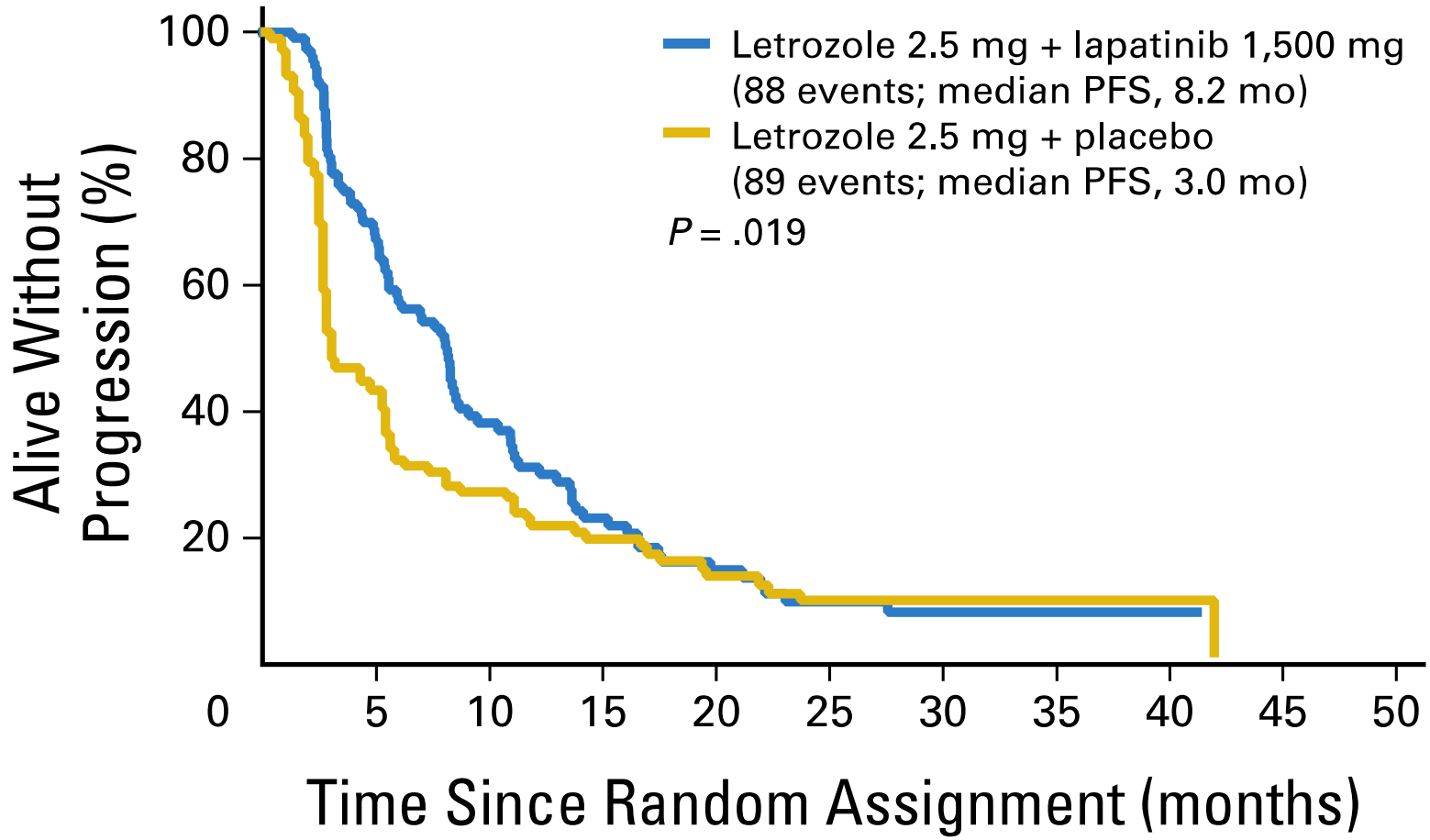


No. at risk

A + H	103	48	31	17	14	13	11	9	4	1	1	0	0
A	104	36	22	9	5	4	2	1	0	0	0	0	0

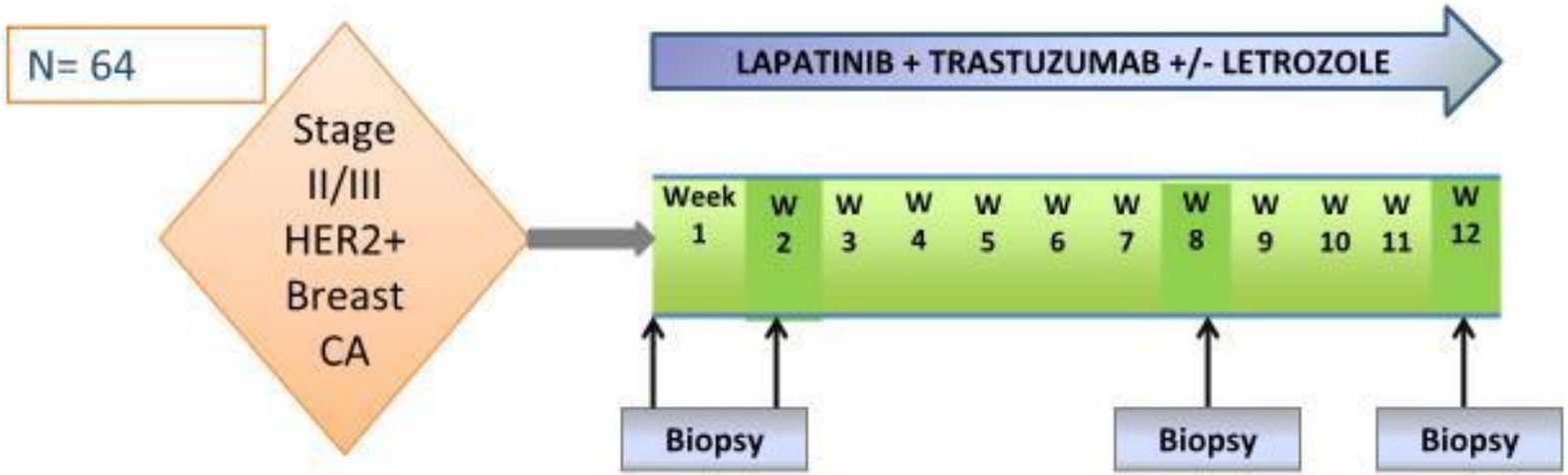
Kaufman et al, JCO 2009

EGF30008 – PFS HER2-positive population



Patients at risk									
Letrozole + lapatinib	111	69	33	20	12	8	4	1	1
Letrozole	108	43	26	18	12	7	5	2	2

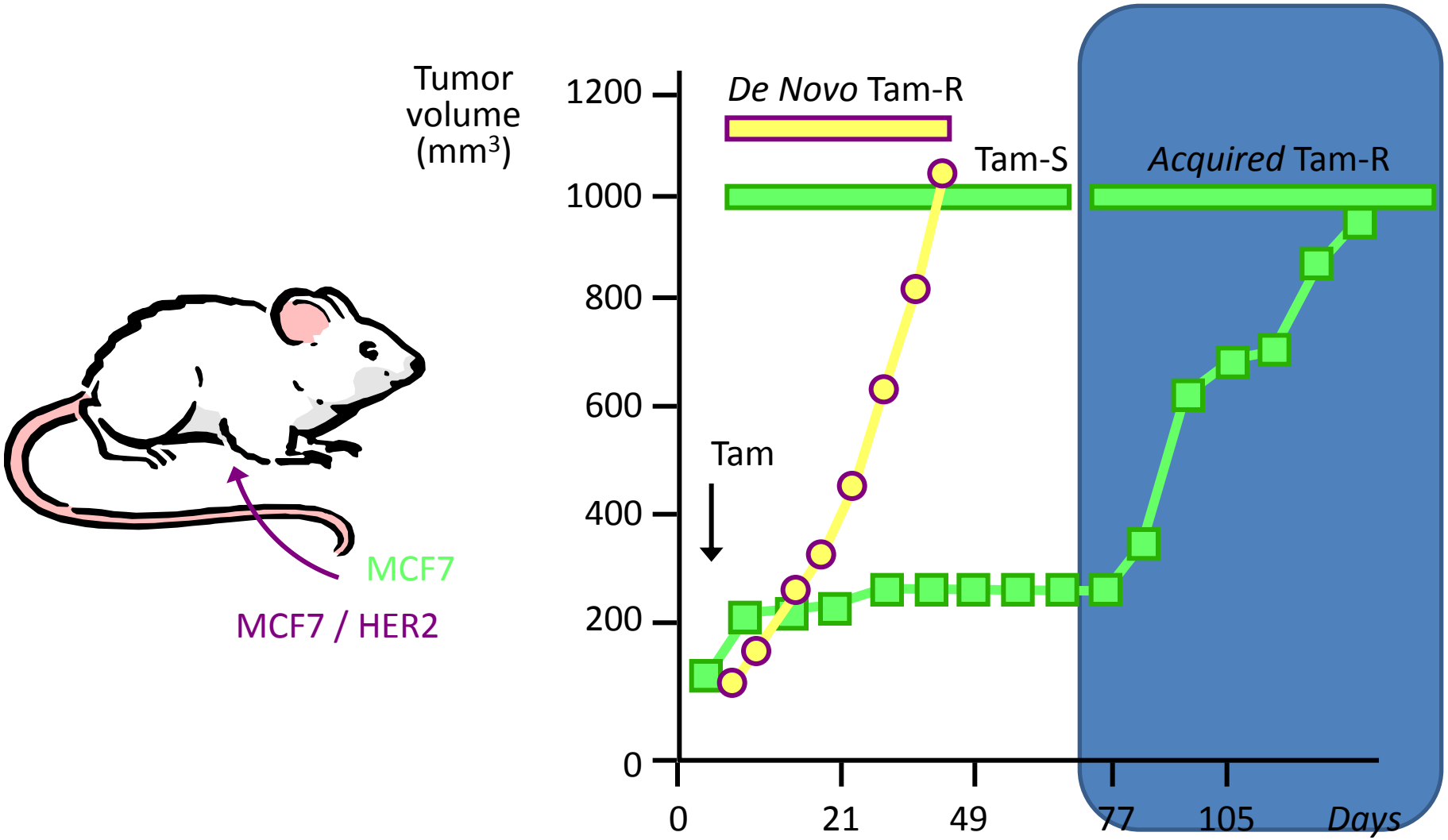
TBCRC 006: Neoadjuvant Lapatinib & Trastuzumab Without Chemotherapy



	Path CR	Near Path CR	Path CR + Near
Overall	27%	22%	49%
ER-	36%	3%	39%
ER+	21%	33%	54%

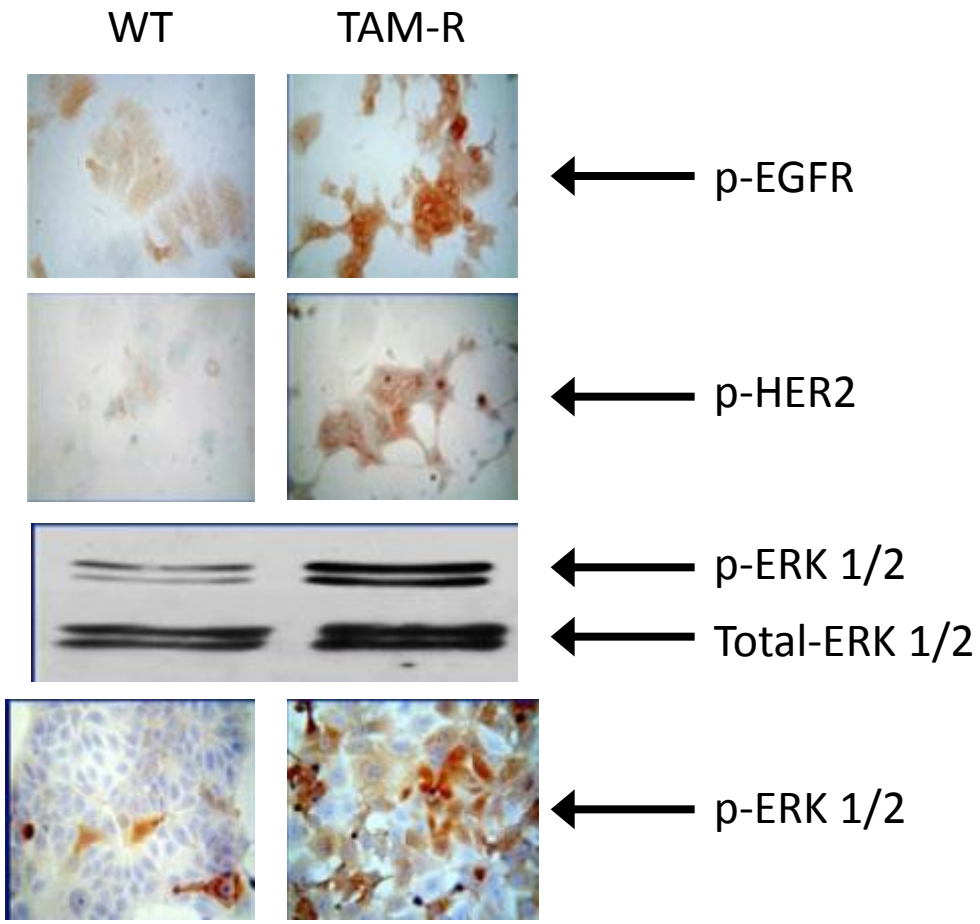
M Rimawi et al,JCO 2013

In Vivo Model of Tamoxifen Resistance



Osborne et al, JNCI 1994

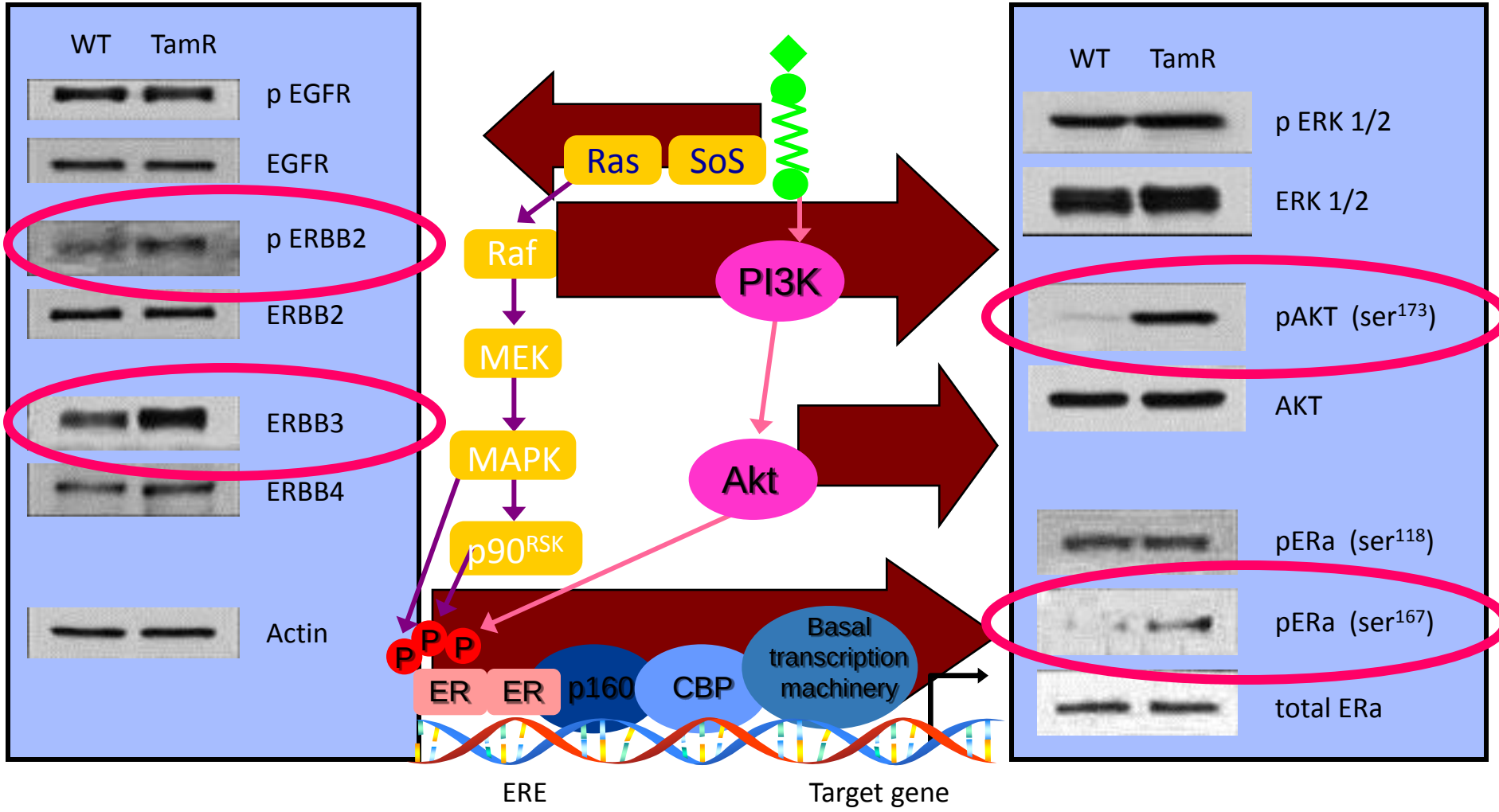
ER+ve Tamoxifen Resistance Cells (TAM-R) show Increased EGFR Signaling



Knowlden et al. Endocrinology 2003

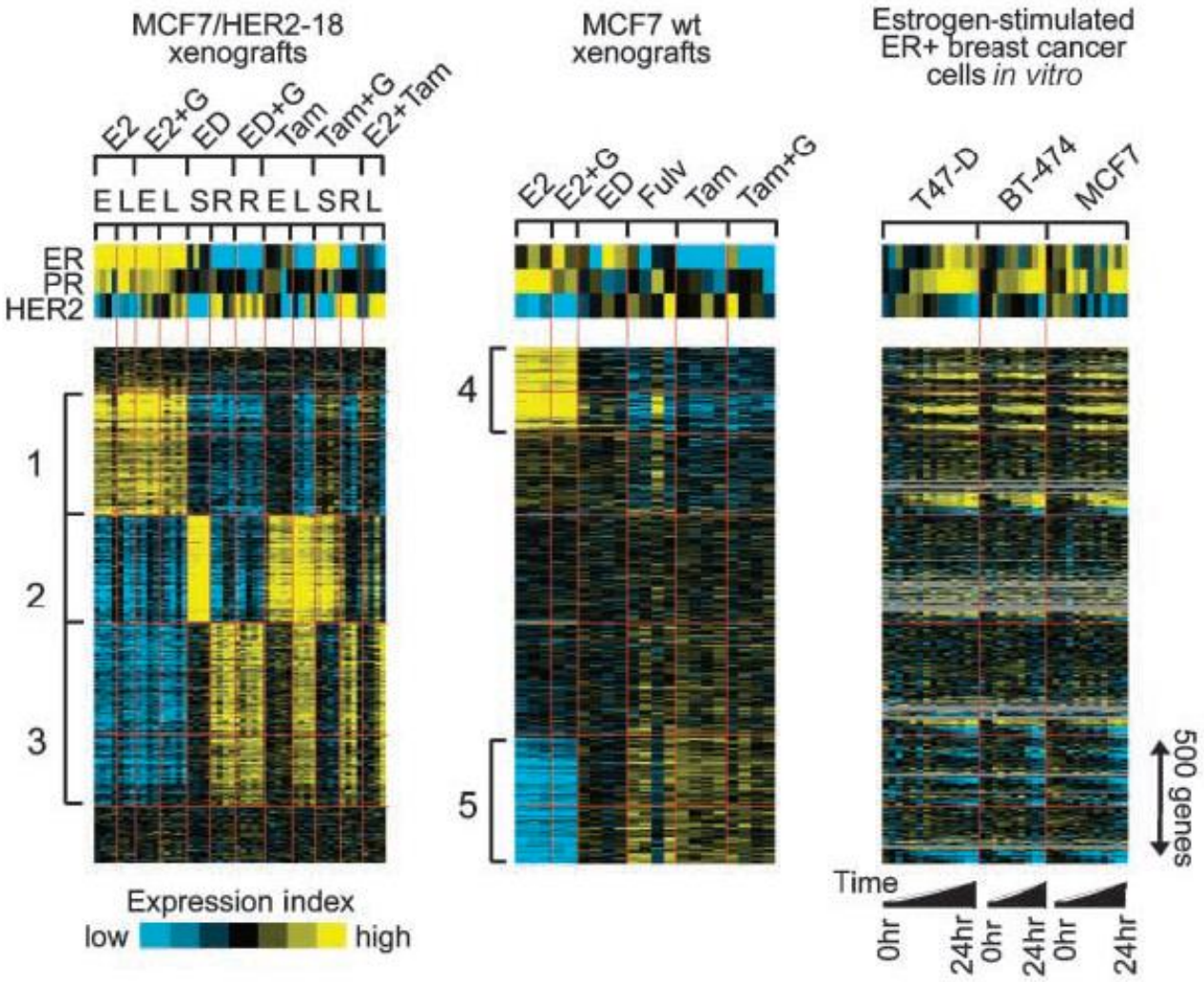
Changes in Growth Factor Receptor Expression and ER Activation in Acquired TamR vs WT cell lines

Type I growth factor receptors
(EGFR, ERBB2, ERBB3, ERBB4)



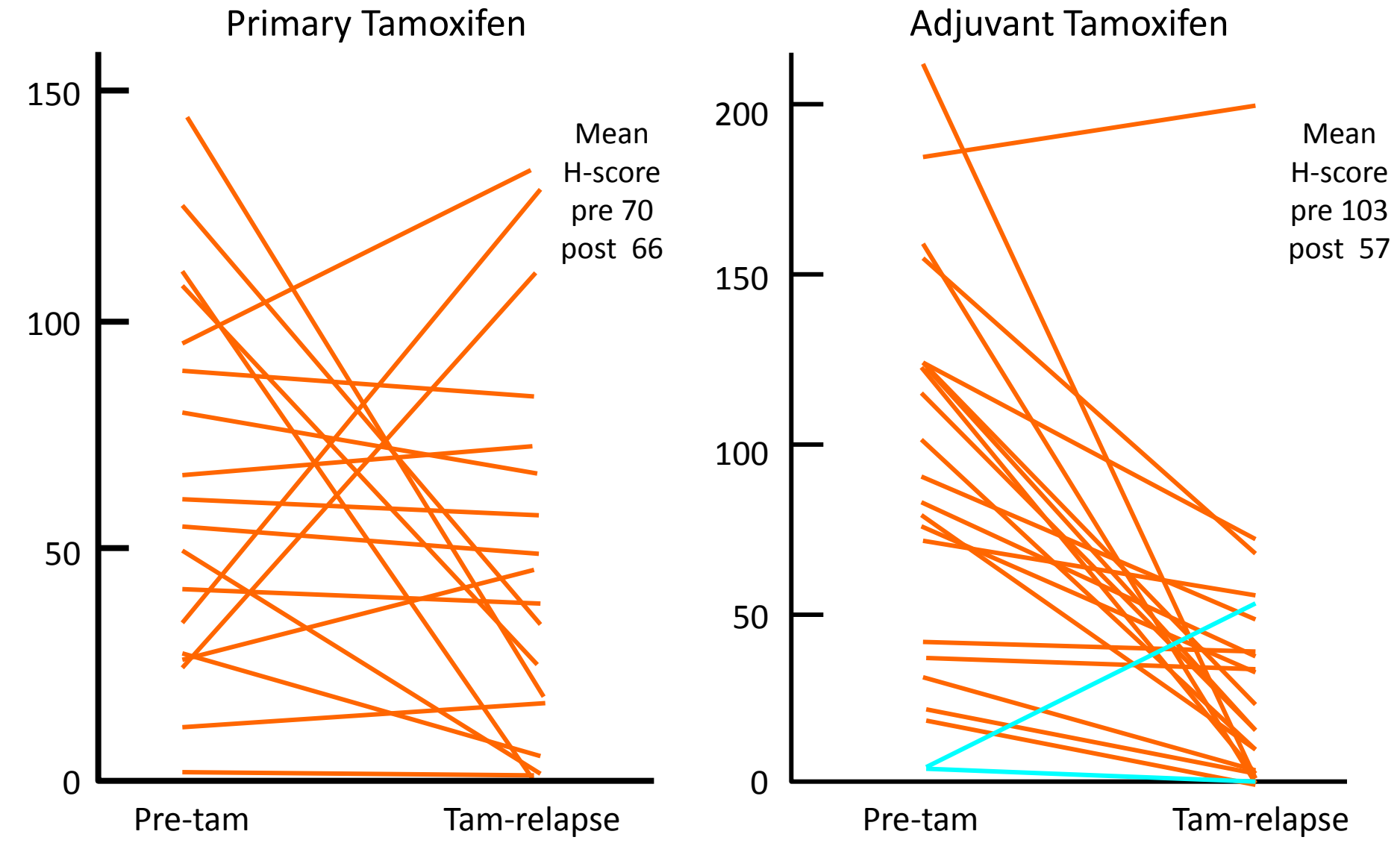
Pancholi et al. Endocr Relat Cancer 2008

Changes in Molecular Profile Subtype at the Development of Endocrine Resistance



Creighton et al Cancer Res. 2009

ER Expression and Acquired Resistance to Tamoxifen



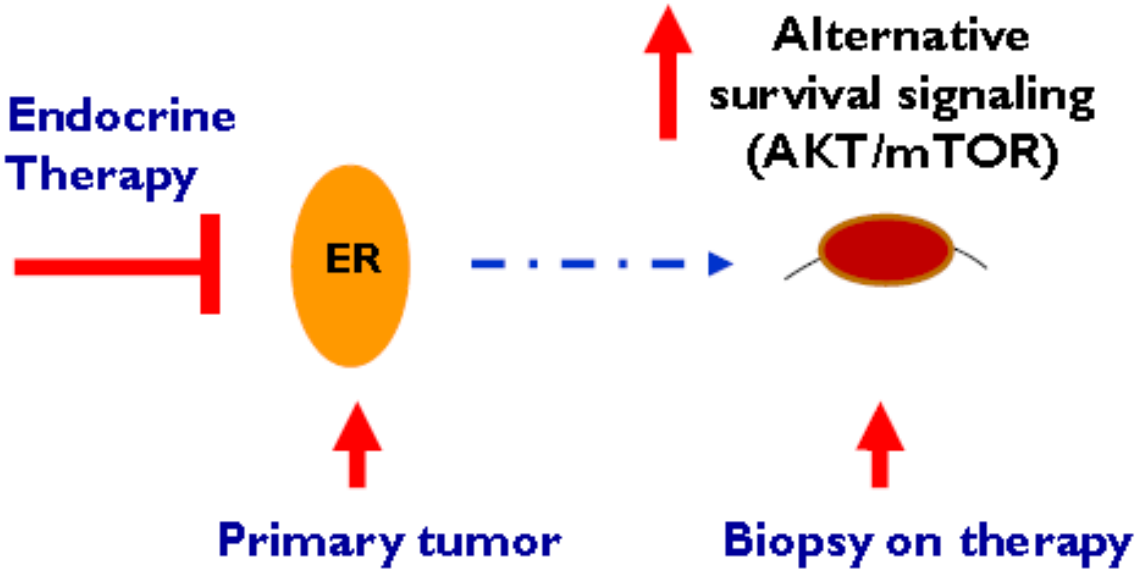
Johnston et al, Cancer Res 1995

Changes of ER and PgR Expression in Primary vs. Subsequent Metestatic Disease

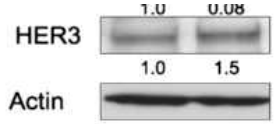
	Liver metastasis		Total
	Negative	Positive	
ER			
Primary tumor			
Negative, *n* (%)	43 (74.1)	15 (25.9)	58 (100)
Positive, *n* (%)	22 (11.2)	175 (88.8)	197 (100)
Total, *n*	65	190	255
Overall discordance rate (95% CI)	14.5 (10.4–19.4)		
PgR			
Primary tumor			
Negative, *n* (%)	73 (80.2)	18 (19.8)	91 (100)
Positive, *n* (%)	106 (64.6)	58 (35.4)	164 (100)
Total, *n*	179	76	255
Overall discordance rate (95% CI)	48.6 (42.3–54.9)		
HER2 status[a]			
Primary tumor			
Negative, *n* (%)	111 (94.1)	7 (5.9)	118 (100)
Positive, *n* (%)	17 (31.5)	37 (68.5)	54 (100)
Total, *n*	128	44	172
Overall discordance rate (95% CI)	13.9 (9.1–20.1)		

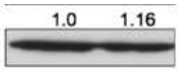
Curigliano et al. Ann. Onc. 2011

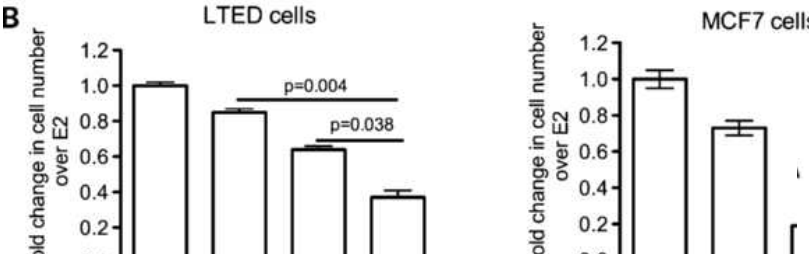
Dynamic ER Signaling:
De-Repression of Resistance Pathways

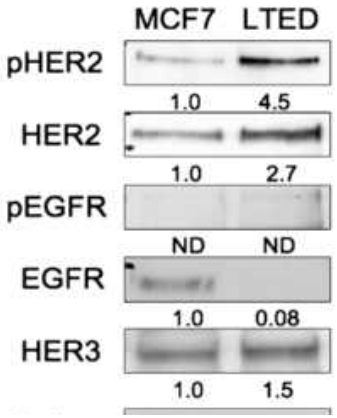


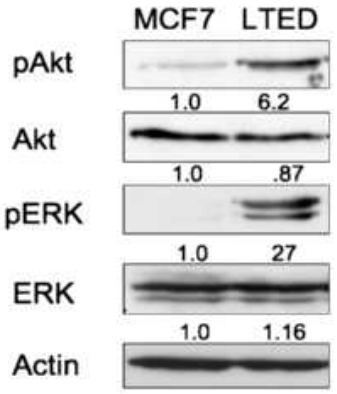
Lapatinib Restores Hormone Sensitivity in HER2-Negative ER-Positive Breast Cancer with Acquired Endocrine Resistance

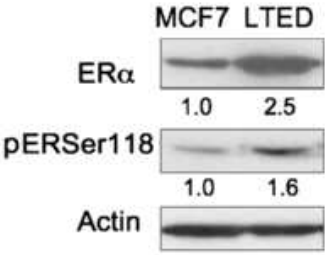


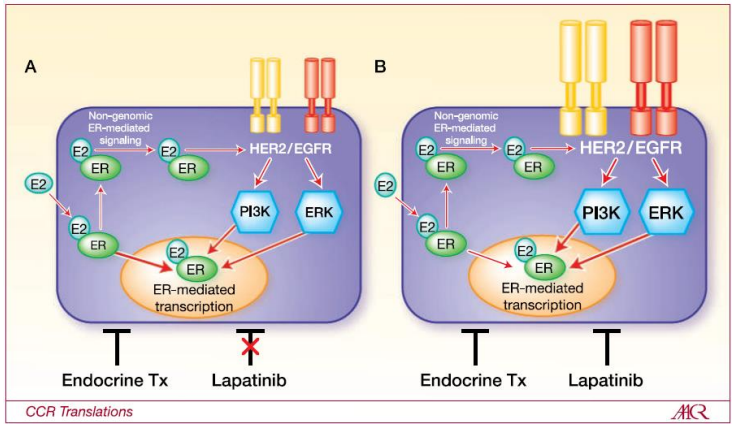






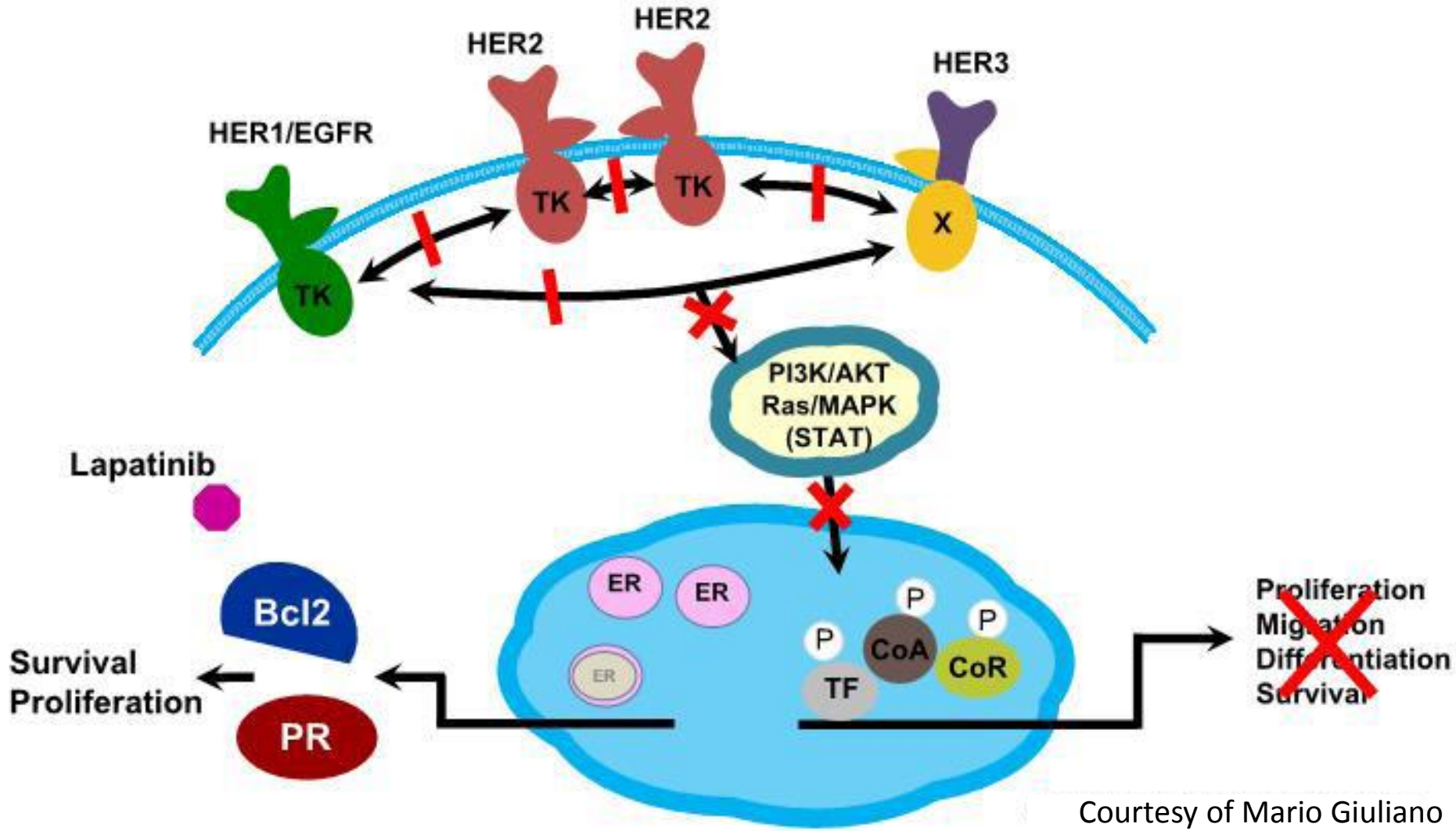






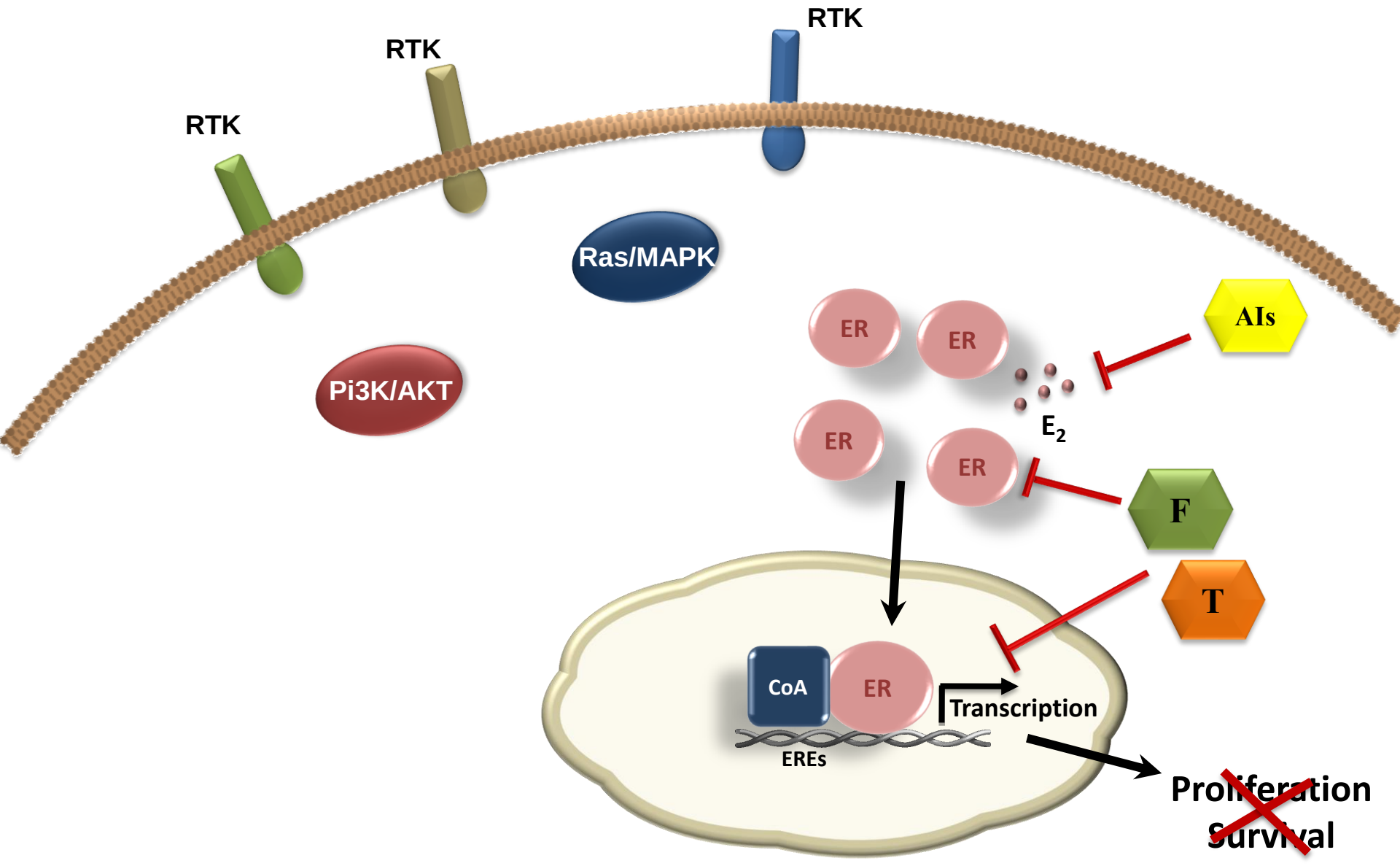
Leary et al CCR 2011

ER Signaling Can Become a Dominant Alternative Driver in HER2-positive Cells Treated With anti HER2 Therapy

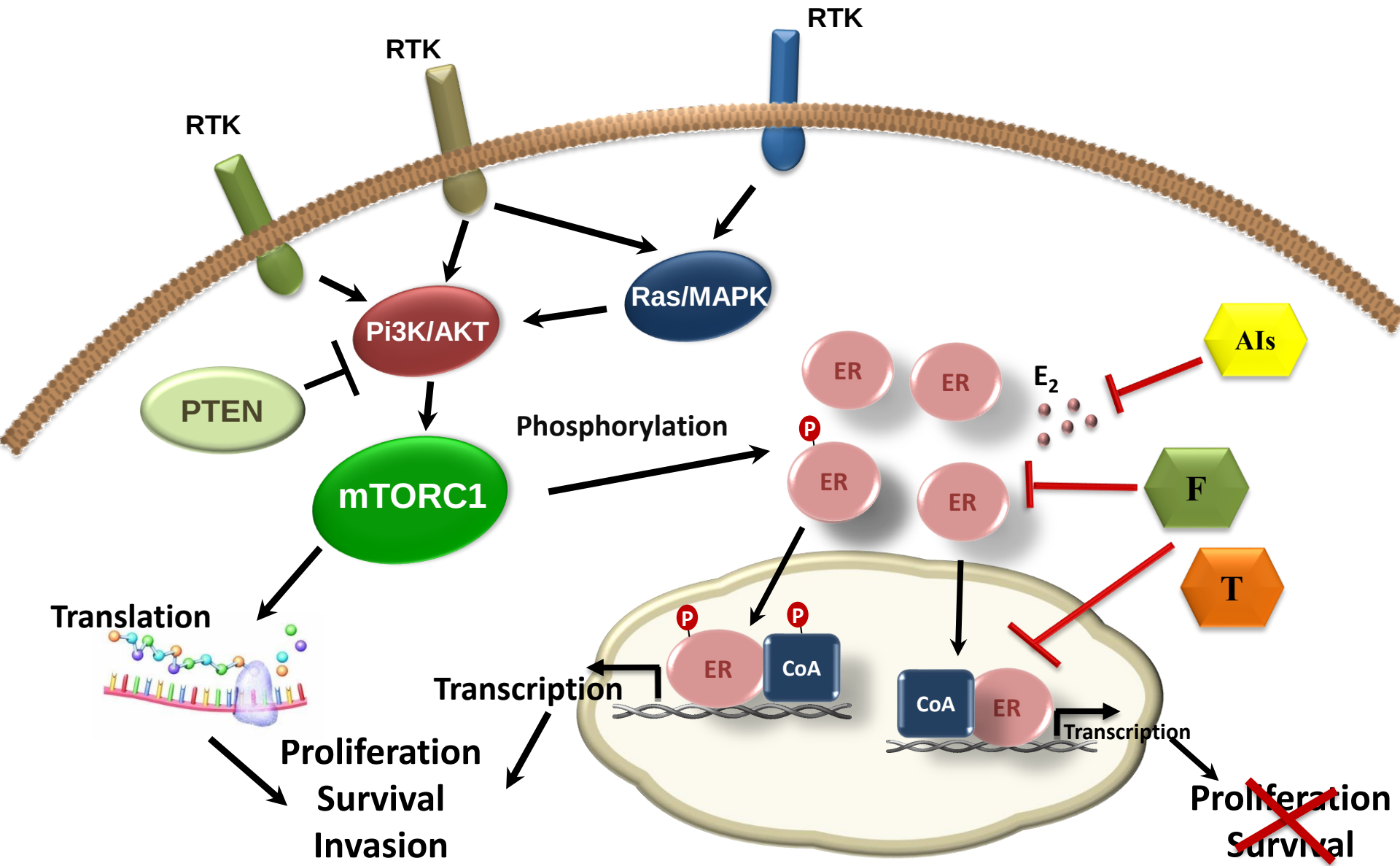


Courtesy of Mario Giuliano

Endocrine responsiveness



Role of PI3K/Akt/mTOR pathway in acquired endocrine resistance

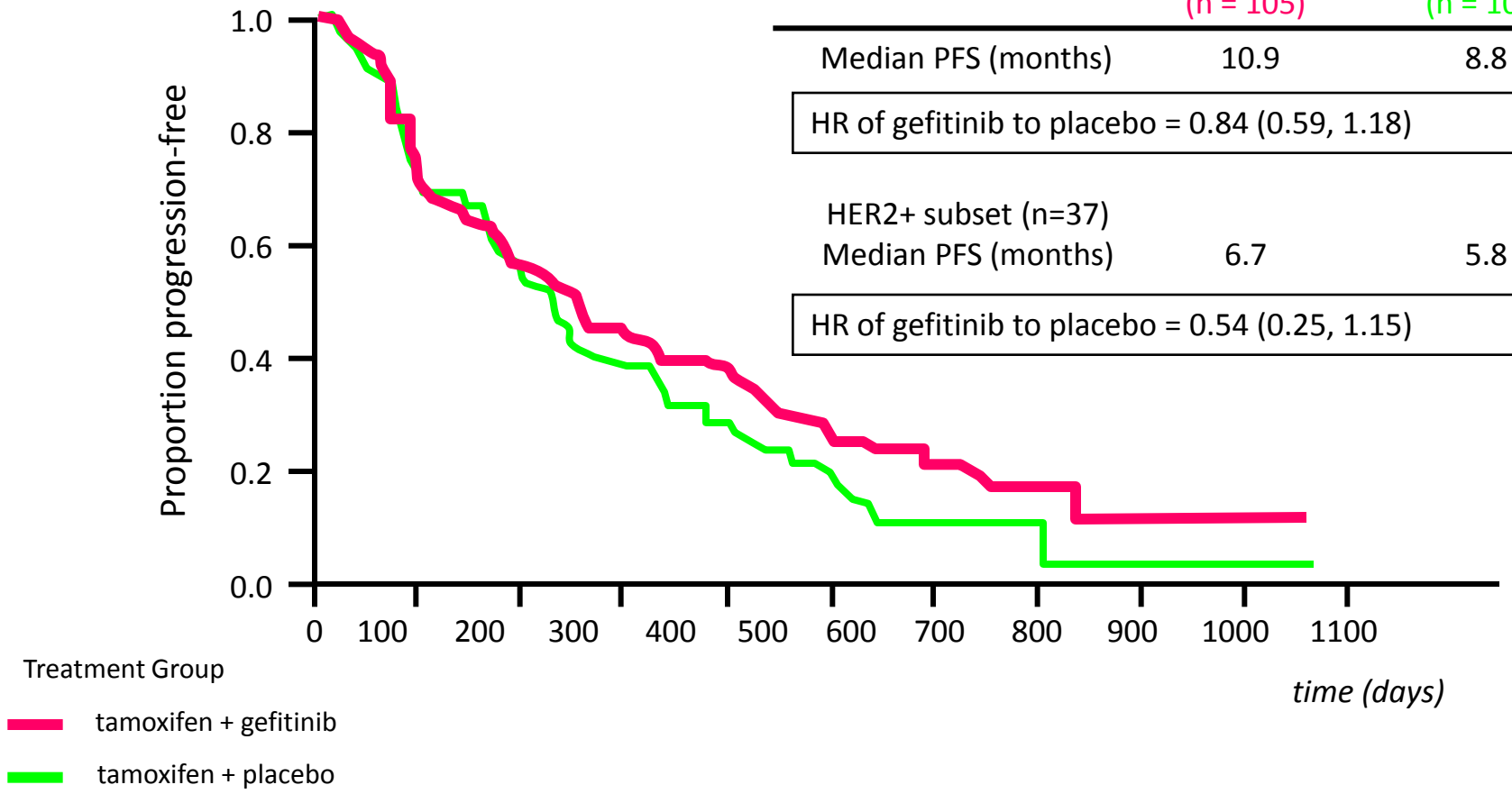


1839IL/0225 – A randomised phase II study of Tamoxifen ± Gefitinib in patients with ER+ve metastatic breast cancer

STRATUM 1: (Endocrine Naive or > 12 m post adjuvant tamoxifen)

Time to Progression

	Tamoxifen + Gefitinib (n = 105)	Tamoxifen + Placebo (n = 101)
Median PFS (months)	10.9	8.8
HR of gefitinib to placebo = 0.84 (0.59, 1.18)		
HER2+ subset (n=37) Median PFS (months)	6.7	5.8
HR of gefitinib to placebo = 0.54 (0.25, 1.15)		

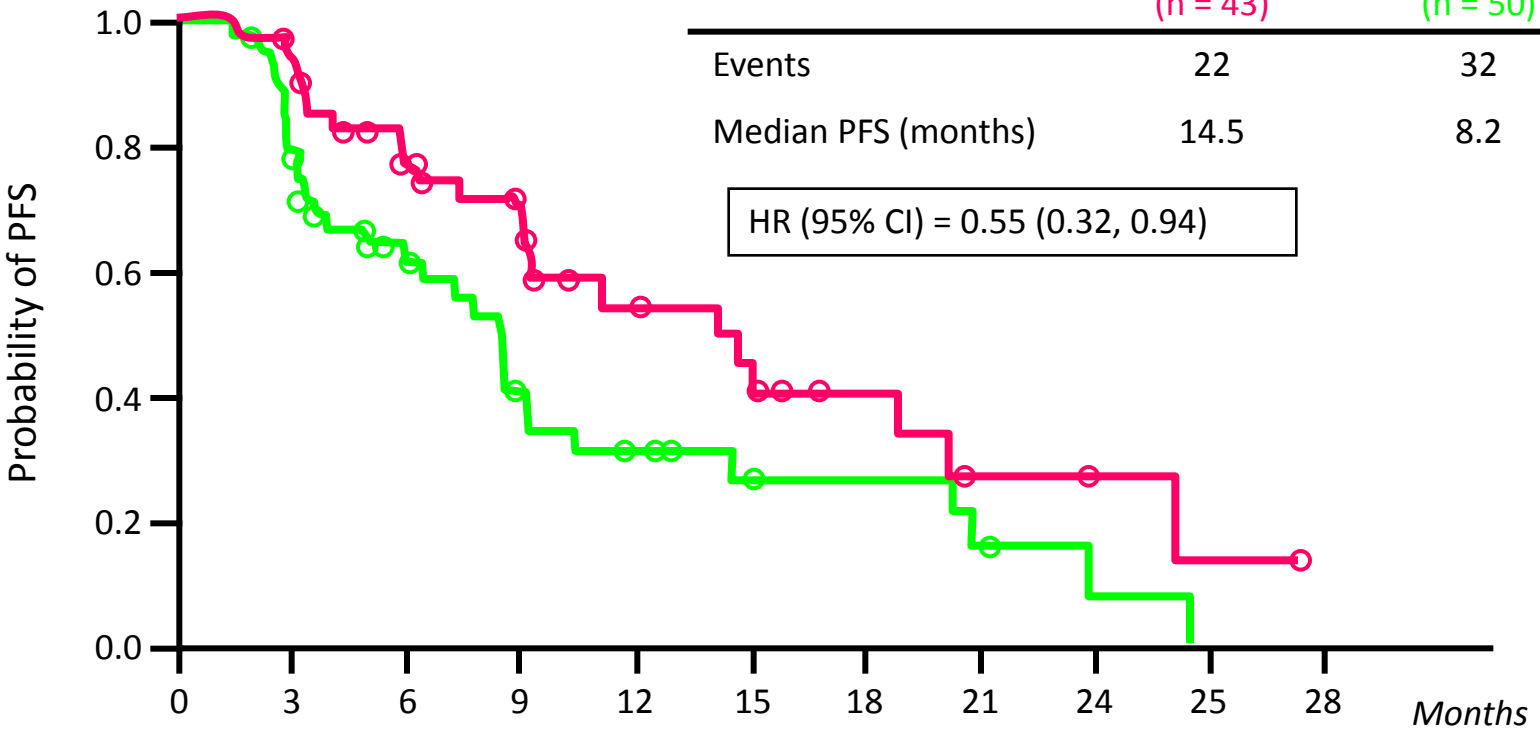


Osborne et al. CCR 2010

Randomised phase II study of Anastrozole ± Gefitinib in patients with ER+ve metastatic breast cancer

	Anastrozole + Gefitinib (n = 43)	Anastrozole + Placebo (n = 50)
Events	22	32
Median PFS (months)	14.5	8.2

HR (95% CI) = 0.55 (0.32, 0.94)



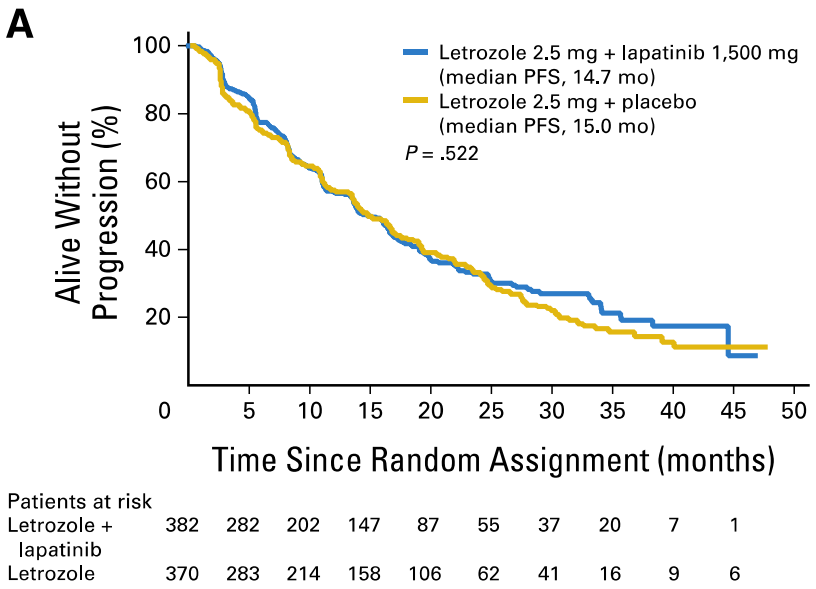
At risk										
Placebo	50	35	23	13	9	6	5	3	1	
Gefitinib	43	40	28	22	13	10	6	3	2	1

Cristofanilli et al. CCR2012

EGF30008 – HER2-ve Patients (N=952)

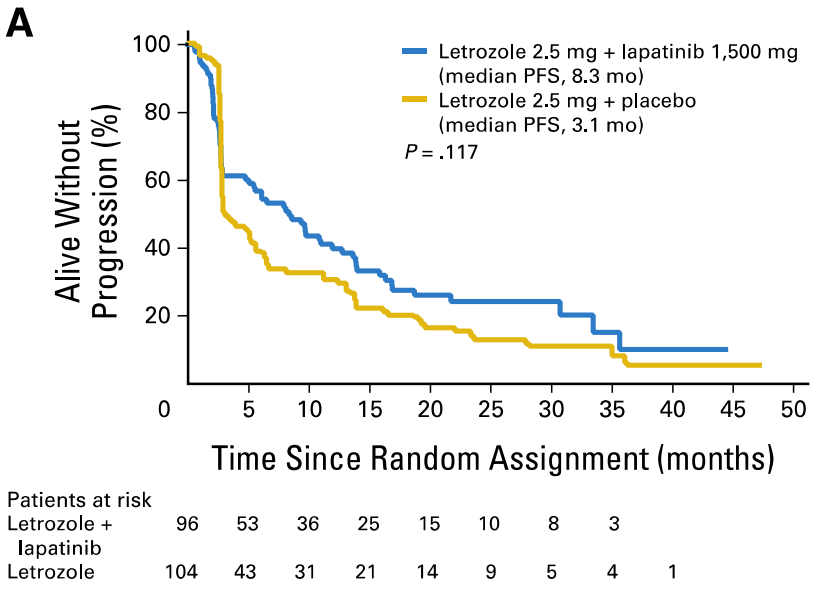
≥ 6 Mo Since D/C of Tam (33%) or No Tam (67%)

- Median tam duration 5 y
- Median time since d/c 3.5 y

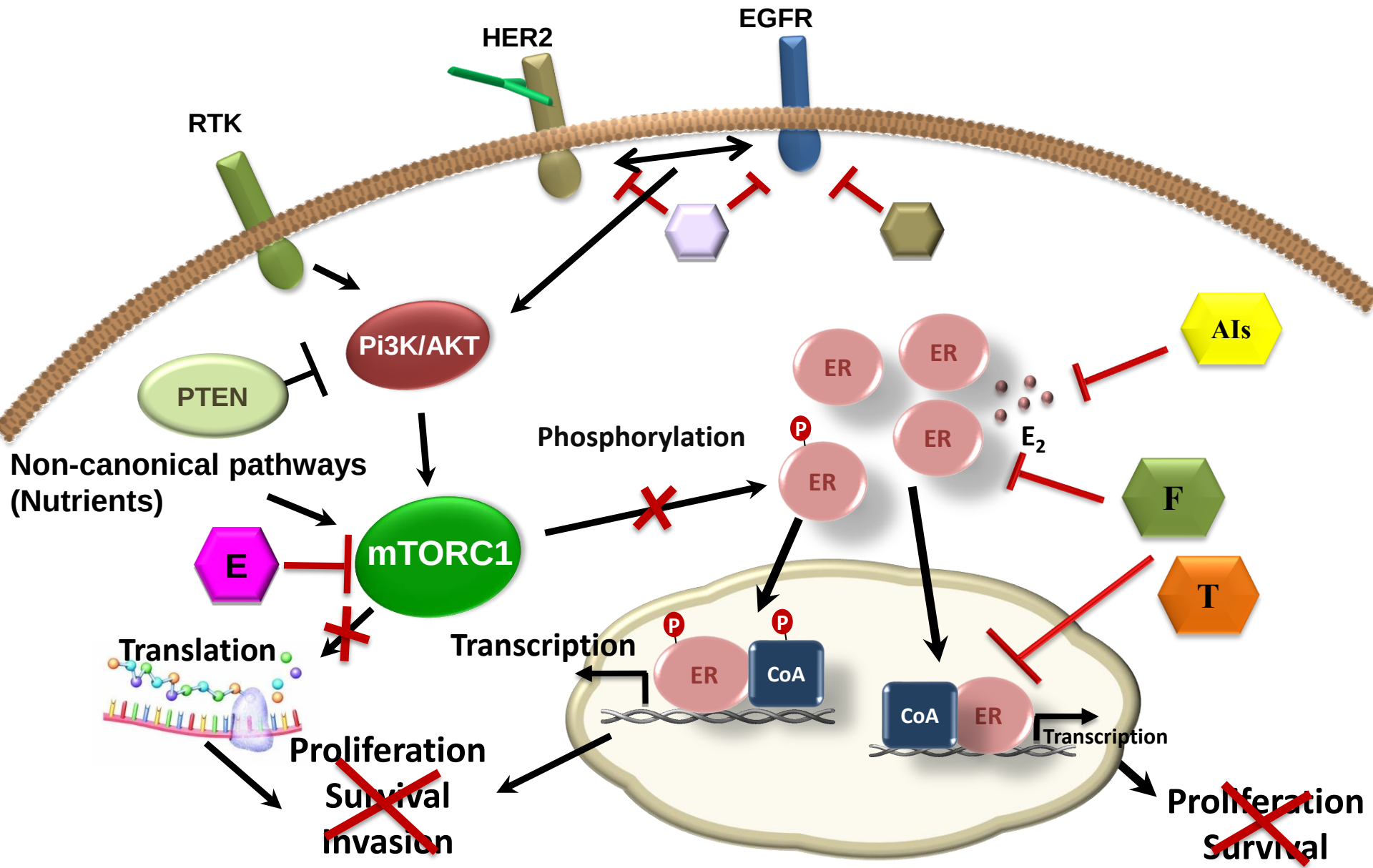


< 6 Mo Since D/C of Tam

- Median tam duration 2.8 y
- Median time since d/c 1 mo

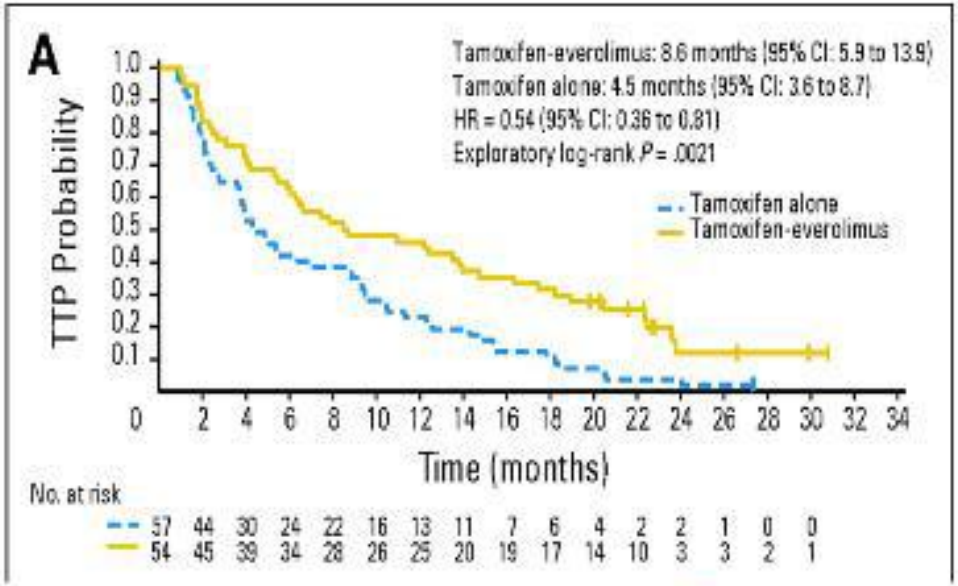


Association of endocrine therapy with Egfr/Her2 inhibitors

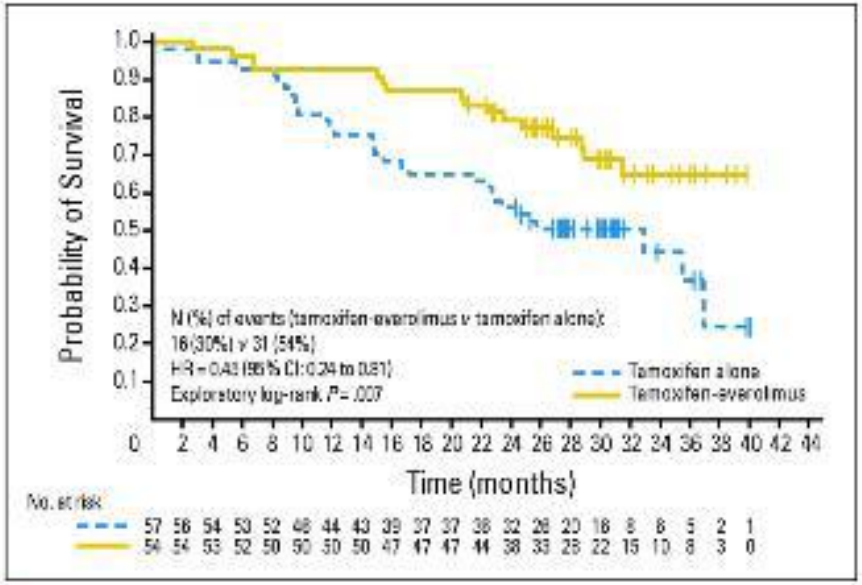


TAMRAD Trial:
Tamoxifen ± everolimus in ER+ HER2- breast cancer with prior AI treatment

PFS



OS

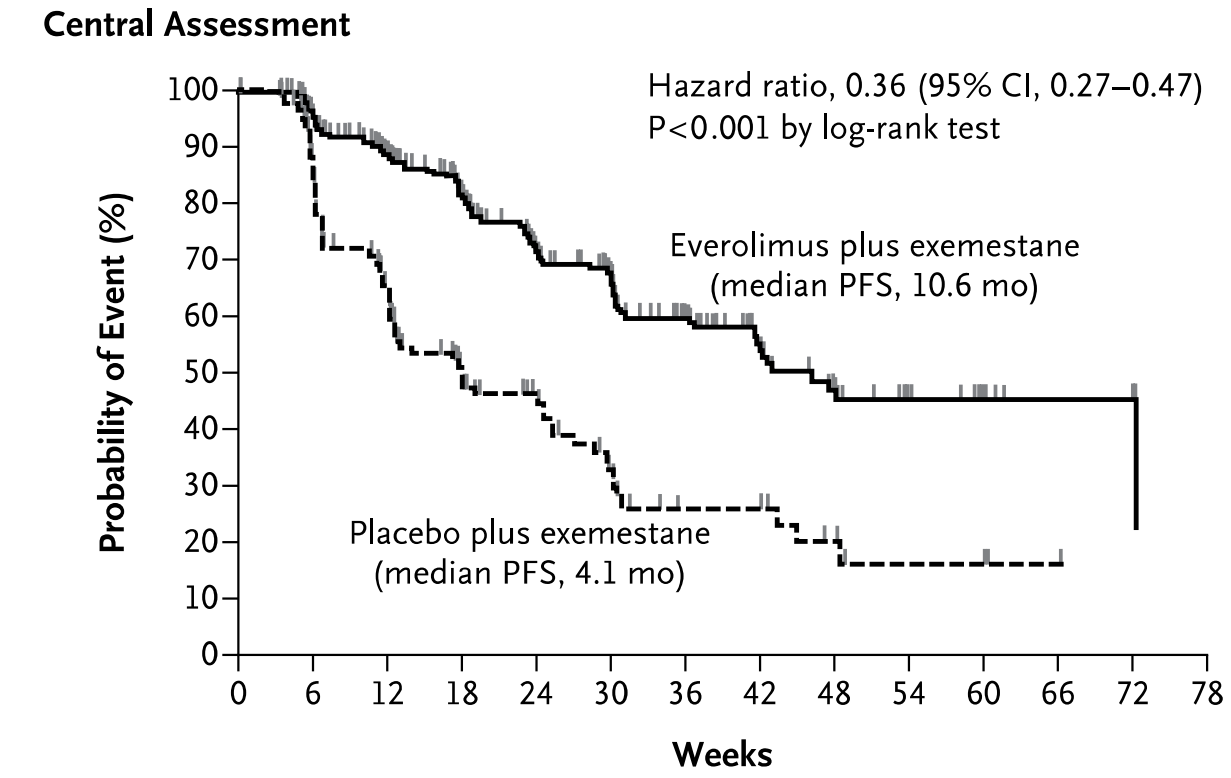


Response rates

Tam	13%
Tam + EVO	14%

Bachelot T et al. JCO 2012;30:2718-2724

Bolero 2-PFS



No. at Risk

Everolimus	485	385	281	201	132	102	67	43	28	18	9	3	2	0
Placebo	239	168	94	55	33	20	11	11	6	3	3	1	0	0

Rb Pathway
Targeting CDKs in ER+ Breast Cancer

- Cyclin dependent kinases (CDK), a group of serine/threonine kinases, play a key role in regulating cell cycle progression by interacting with specific cyclin proteins

Musgrove et al Nat Rev Can 2011

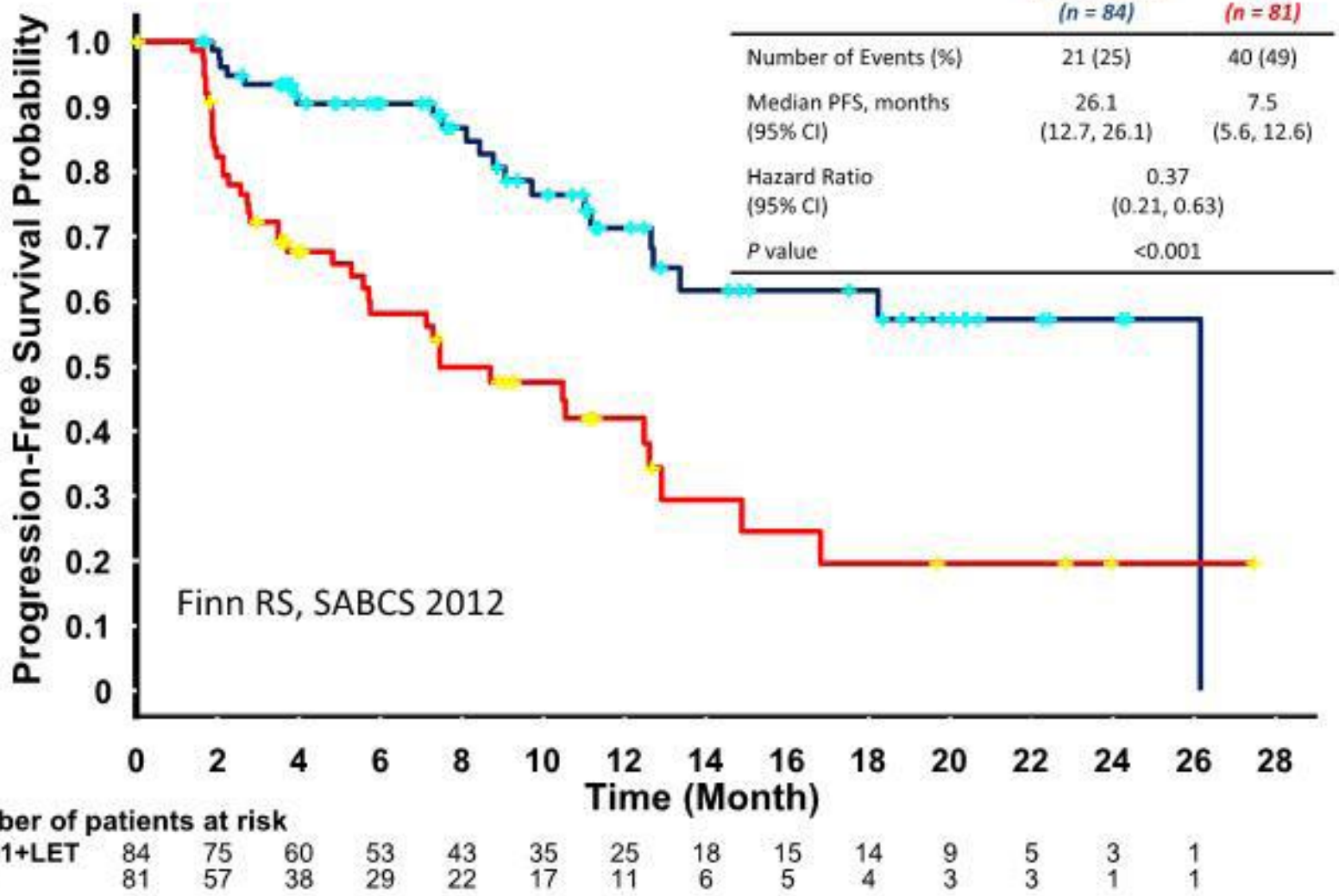
- PD 0332991 (palbociclib) is an oral, highly selective inhibitor of CDK 4/6 kinase

-Prevents cellular DNA sinthesis by prohibiting progression of the cell cycle from G1 to S phase

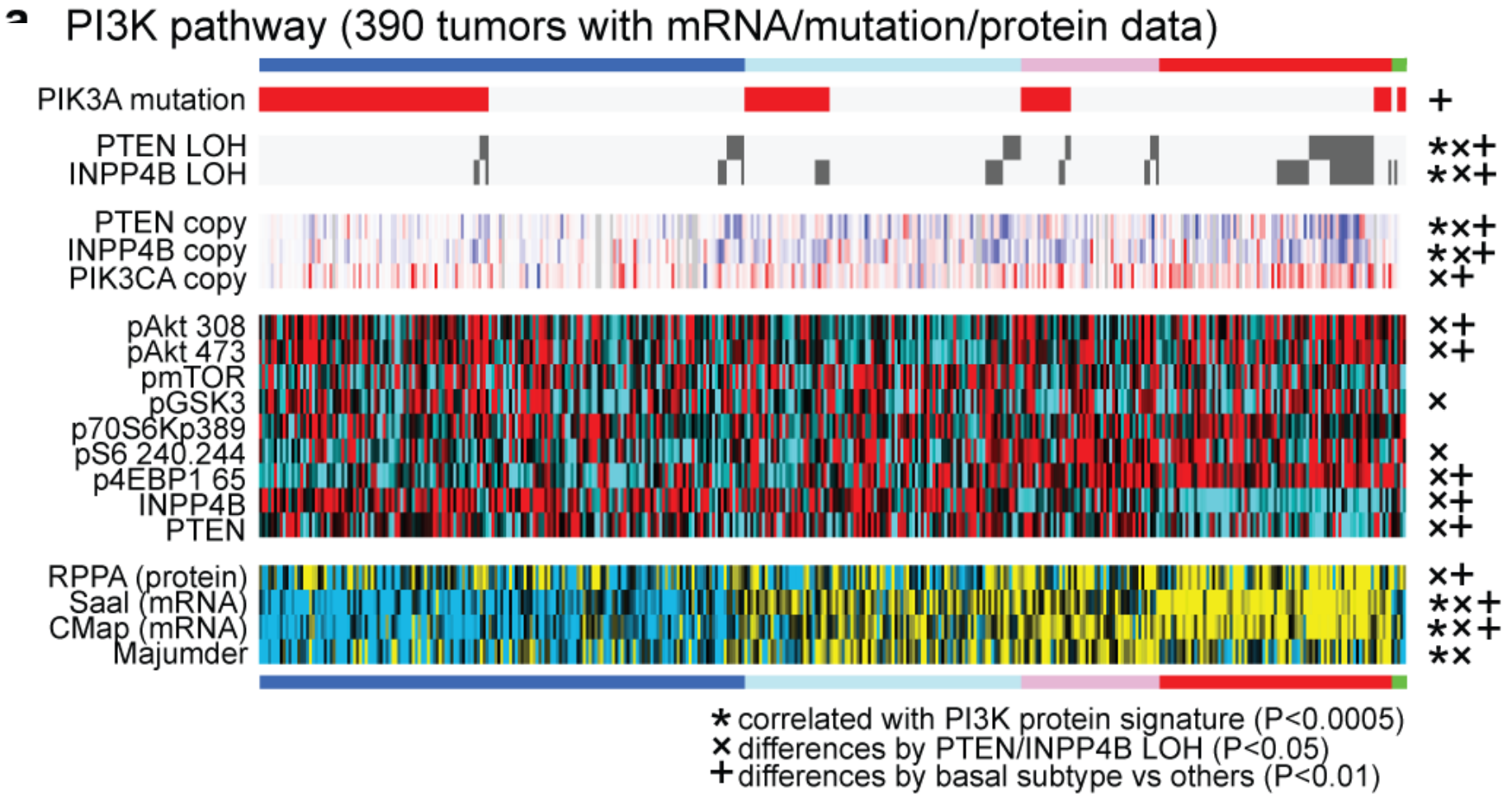
-Synergistic activity also observed in vitro when combined with tamoxifen

Finn et al. BCR 2009

1st line therapy for ER+ MBC
Letrozole ± palbociclib



	PD 991 + LET (n = 84)	LET (n = 81)
Number of Events (%)	21 (25)	40 (49)
Median PFS, months (95% CI)	26.1 (12.7, 26.1)	7.5 (5.6, 12.6)
Hazard Ratio (95% CI)	0.37 (0.21, 0.63)	
P value	<0.001	



Nature. 2012 October 4; 490(7418): 61–70. doi:10.1038/nature11412.

Clinical Implications

1. In breast cancer, HR and GF signaling are the dominant pathways driving tumor growth and survival
2. Alternative pathways may contribute to endocrine resistance development

Identification of the networks driving progression in an individual patient's tumor, and

Completely or nearly completely blocking those pathways,

May lead to tumor eradication in patients.