Genomic Portrait of Breast Cancer (with an emphasis on clinical utility)

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Conflicts of interest

- Prosigna, PAM50, Nanostring and Bioclassifier LLC – stocks, patents, consulting and company position
- HER2 targeting consulting, Genentech and Puma
- PI3 kinase pathway consulting, Novartis, Genentech
- CDK4/6 inhibitor consulting, Pfizer, Eli Lilly, Novartis.
- MDM2 inhibitor consulting, Sanofi-Aventis
- Fulvestant consulting, AstraZenica

The Cancer Genome Atlas for Breast

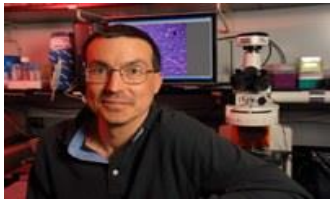
ARTICLE

doi:10.1038/nature11412

Comprehensive molecular portraits of human breast tumours

The Cancer Genome Atlas Network*

We analysed primary breast cancers by genomic DNA copy number arrays, DNA methylation, exome sequencing, messenger RNA arrays, microRNA sequencing and reverse-phase protein arrays. Our ability to integrate information across platforms provided key insights into previously defined gene expression subtypes and demonstrated the existence of four main breast cancer classes when combining data from five platforms, each of which shows significant molecular heterogeneity. Somatic mutations in only three genes (*TP53*, *PIK3CA* and *GATA3*) occurred at >10% incidence across all breast cancers; however, there were numerous subtype-associated and novel gene mutations including the enrichment of specific mutations in *GATA3*, *PIK3CA* and *MAP3K1* with the luminal A subtype. We identified two novel protein-expression-defined subgroups, possibly produced by stromal/microenvironmental elements, and integrated analyses identified specific signalling pathways dominant in each molecular subtype including a HER2/phosphorylated HER2/EGFR/phosphorylated EGFR signature within the HER2-enriched expression subtype. Comparison of basal-like breast tumours with high-grade serous ovarian tumours showed many molecular commonalities, indicating a related aetiology and similar therapeutic opportunities. The biological finding of the four main breast cancer subtypes caused by different subsets of genetic and epigenetic abnormalities raises the hypothesis that much of the clinically observable plasticity and heterogeneity occurs within, and not across, these major biological subtypes of breast cancer.



Charles Perou

TCGA Breast Tumor Significantly Mutated Gene List by Clinical Receptor Status (n=507)

Gene	All Tumors (n=507)			ER+/Her2- (n=330)			Clinical Her2+ (n=75)			Triple-negative (n=86)		
	#Cases	LRT	CT	#Cases	LRT	CT	#Cases	LRT	CT	#Cases	LRT	CT
PIK3CA	180	0	0	145	0	0	23	0	0	9	5.55E-09	3.22E-10
TP53	187	0	0	68	0	0	41	0	0	68	0	0
GATA3	54	0	0	45	0	0	8	0	0	0	NA	NA
MAP3K1	39	0	0	36	0	0	2	NA	NA	0	NA	NA
CDH1	33	0	0	30	0	0	2	NA	NA	1	NA	NA
MLL3	37	0	0	28	0	0	5	NA	NA	3	NA	NA
MAP2K4	21	0	0	19	0	0	1	NA	NA	1	NA	NA
PTEN	17	0	0	16	0	0	0	NA	NA	1	NA	NA
RUNX1	18	0	0	15	0	0	1	NA	NA	0	NA	NA
USH2A	27	2.08E-02	1.42E-03	13	NA	NA	4	NA	NA	9	NA	NA
RYR2	22	4.08E-02	1.09E-02	13	NA	NA	5	NA	NA	3	NA	NA
NCOR1	17	1.25E-02	2.99E-05	13	1.10E-05	4.33E-07	1	NA	NA	1	NA	NA
NF1	14	1.84E-02	4.20E-03	11	1.09E-02	1.39E-02	1	NA	NA	2	NA	NA
TBX3	13	2.77E-13	6.72E-13	11	5.74E-12	4.91E-12	0	NA	NA	1	NA	NA
CTCF	13	4.68E-03	3.74E-05	11	6.46E-04	2.31E-06	1	NA	NA	1	NA	NA
AKT1	12	1.76E-12	6.83E-11	11	2.75E-13	3.94E-12	1	NA	NA	0	NA	NA
PIK3R1	14	3.99E-10	6.34E-11	9	8.44E-07	1.82E-06	4	NA	NA	1	NA	NA
PTPRD	12	3.87E-02	1.69E-02	8	NA	NA	2	NA	NA	2	NA	NA
SF3B1	10	6.26E-04	9.08E-04	7	2.27E-03	1.07E-02	1	NA	NA	0	NA	NA
CBFB	8	9.90E-08	7.72E-08	7	1.32E-07	5.10E-08	0	NA	NA	1	NA	NA
AFF2	13	1.98E-02	3.97E-03	6	NA	NA	3	NA	NA	4	NA	NA
TBL1XR1	8	2.38E-04	9.26E-06	6	6.33E-04	1.83E-05	1	NA	NA	1	NA	NA
ZFP36L1	7	5.89E-04	2.60E-04	6	7.14E-05	1.27E-04	0	NA	NA	1	NA	NA
RPGR	10	1.00E-02	1.59E-03	5	NA	NA	0	NA	NA	3	NA	NA
CDKN1B	5	7.09E-05	1.11E-03	5	4.60E-06	5.94E-05	0	NA	NA	0	NA	NA
DCAF4L2	7	2.36E-02	4.99E-02	4	NA	NA	2	NA	NA	1	NA	NA
GPS2	6	1.87E-05	4.75E-04	4	6.98E-03	3.73E-02	1	NA	NA	1	NA	NA
OR6A2	4	1.71E-02	2.54E-02	4	3.67E-03	4.32E-03	0	NA	NA	0	NA	NA
RB1	9	1.59E-02	1.59E-03	3	NA	NA	1	NA	NA	4	2.77E-02	4.64E-02
PTPN22	7	2.44E-03	8.63E-03	3	NA	NA	4	6.57E-03	3.36E-02	0	NA	NA
SEPT13	4	1.32E-03	4.71E-02	3	NA	NA	0	NA	NA	1	NA	NA
HIST1H2BC	4	2.36E-02	2.58E-02	2	NA	NA	1	NA	NA	1	NA	NA
CCND3	2	5.07E-03	4.11E-02	2	8.48E-04	7.40E-03	0	NA	NA	0	NA	NA
GPR32	5	1.58E-02	3.14E-02	1	NA	NA	1	NA	NA	1	NA	NA

Whole genome sequencing in a clinical trial

Li Ding

ARTICLE

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Whole-genome analysis informs breast cancer response to aromatase inhibition

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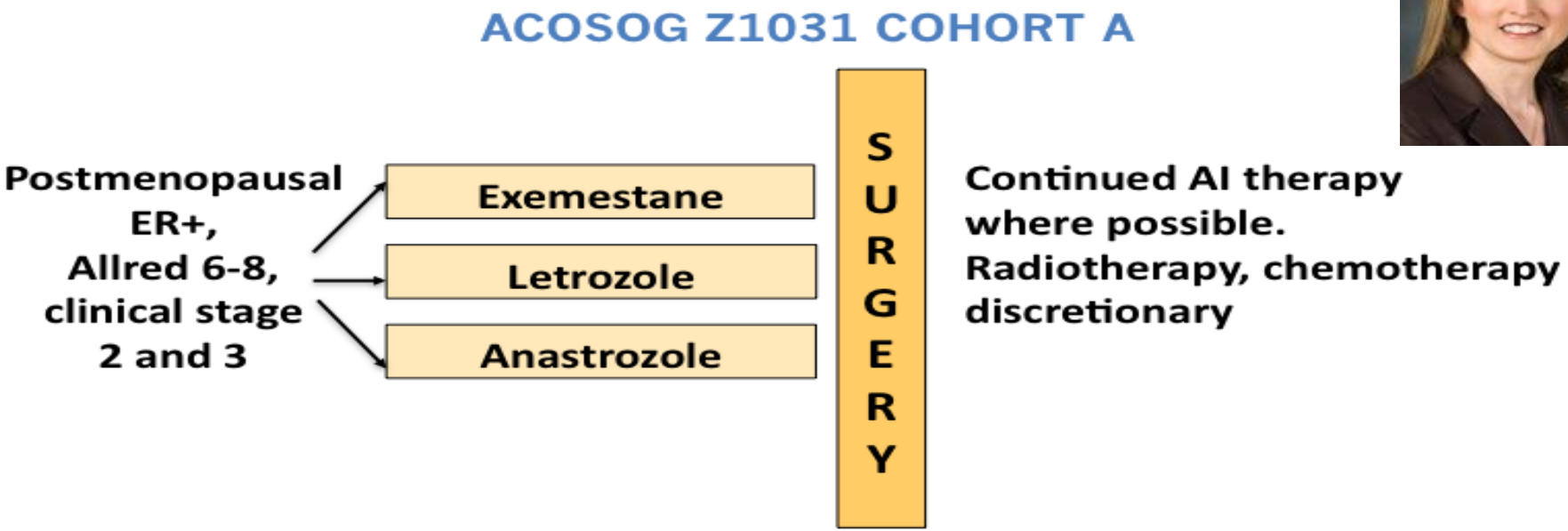
Elaine Mardis

Ellis, M.J., Ding, L., Shen, D., Luo, J., Suman, V.J., Wallis, J.W., Van Tine, B.A., Hoog, J., Goiffon, R.J., Goldstein, T.C., *et al. (2012). Whole-genome analysis informs breast cancer response to aromatase inhibition. Nature 486, 353-360.*

Clinical Trial Design



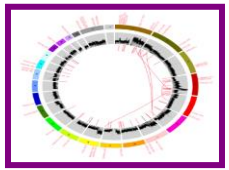
1) 51% of patients requiring mastectomy at baseline could undergo breast conserving surgery after neoadjuvant aromatase inhibitor therapy
2) No clinically significant difference in outcomes between the two arms
3) The three aromatase inhibitors suppressed Ki67 to the same extent, indicating that they are biologically equivalent agents.

Ellis et al JCO 2011: 29, 2342-9

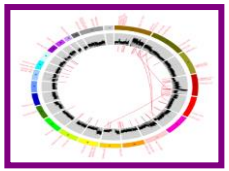
NCI, BCRF, Pfizer, Novartis

Experimental Design For Genomic Discovery

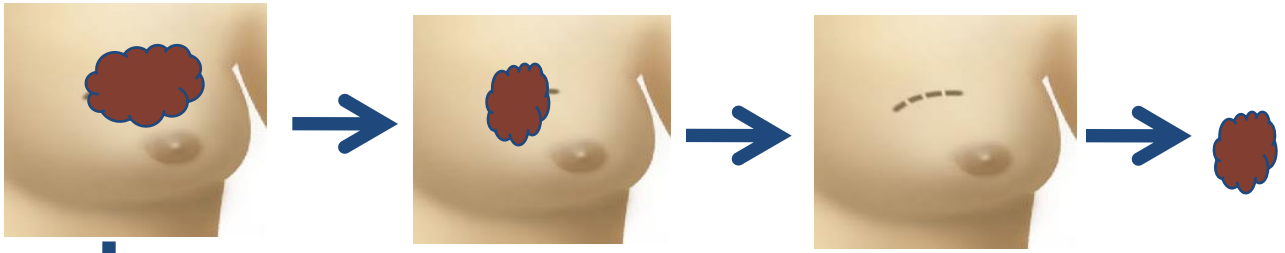
2 baseline frozen cores
70%+ tumor cellularity
DNA extracted



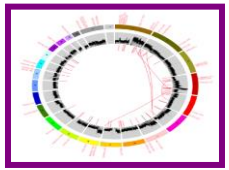
Ki67 in surgical sample
Greater that 10% = Unfavorable



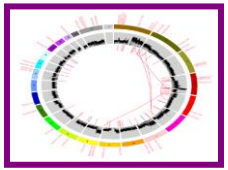
16 to 18 weeks of aromatase inhibition



2 baseline frozen cores
70%+ tumor cellularity
DNA extracted



Ki67 in surgical sample
Less than10% = Favorable



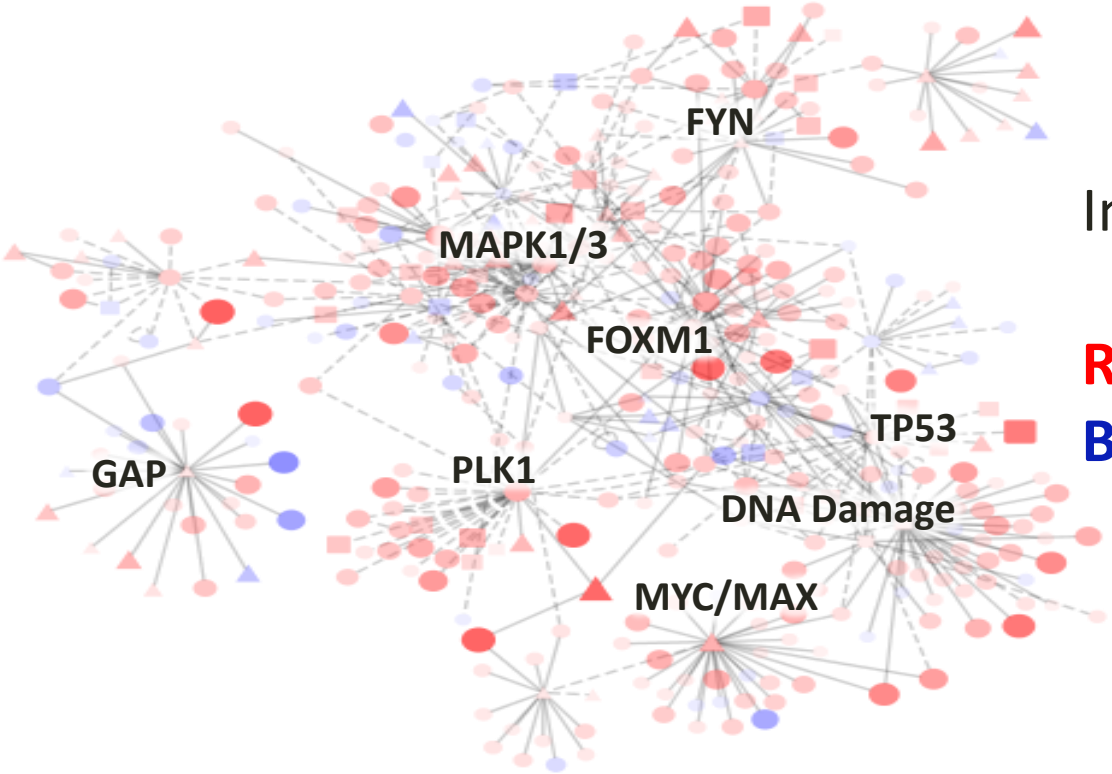
BCRF, NHGRI, NCI

Correlations between mutations, AI responses markers and histology

Gene	Expression/histo-pathology variable		Mutation Frequency*	SET1 P†	SET2 P†	Whole Set FDR P¶
TP53	Luminal subtype	Luminal A	9.3% (13/140)	0.001	0.46	0.041
		Luminal B	21.5% (38/177)			
TP53	Histological grade	I	4.5% (3/66)	0.050	0.067	0.020
		II/III	19.2% (48/250)			
MAP3K1	Luminal subtype	Luminal A	20.0% (28/140)	0.018	0.028	0.005
		Luminal B	6.2% (11/177)			
MAP3K1	Histological grade	I	25.8% (17/66)	0.061	0.011	0.005
		II/III	8.8% (22/250)			
CDH1	Histological Type	Ductal	5.9% (10/169)	0.41‡	2.8E-11	3.9E-10
		Lobular	50.0% (20/40)			

Ellis, M et al Nature DOI: 10.1038/nature11143

Pathways of Aromatase Inhibitor Resistance



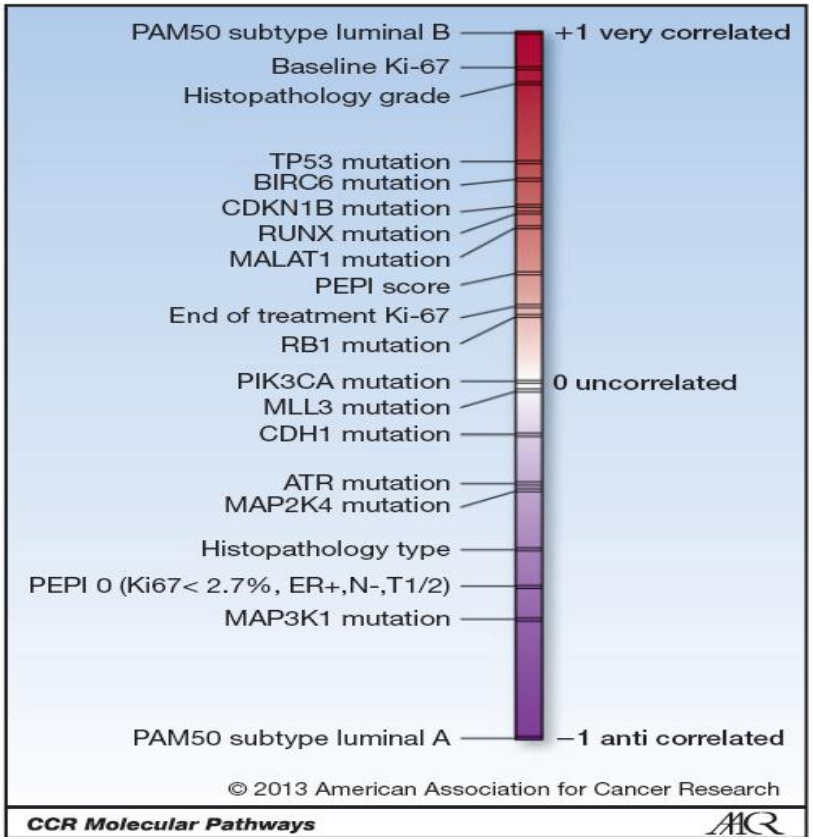
In resistant tumors:

Red = Up-regulation

Blue = Down-regulation

Ellis, M et al Nature DOI: 10.1038/nature11143

Extracting Medical Information from complex genomic data

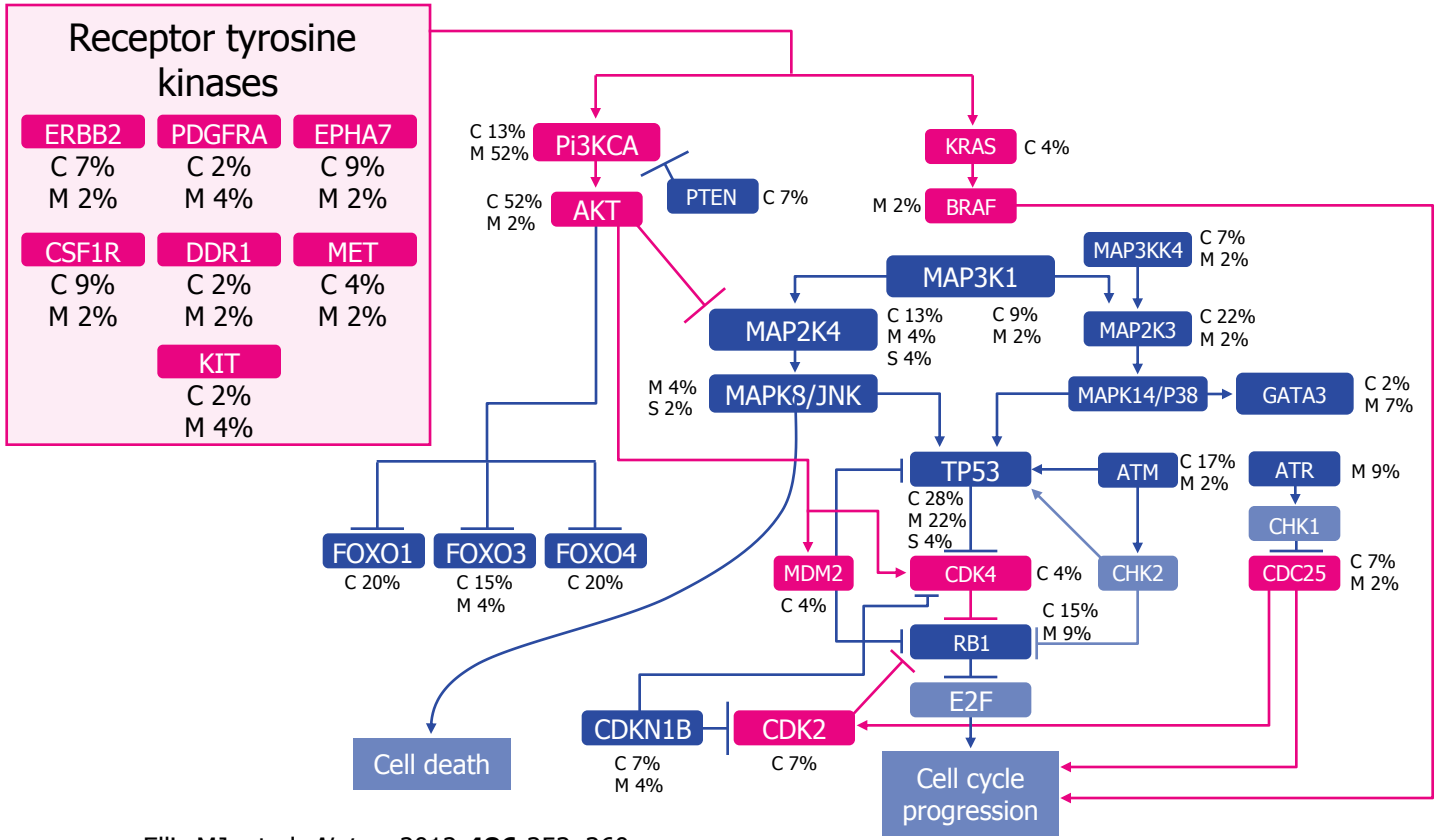


Josh Stuart

Ted Goldstein

Goldstein T PE, Ellis MJ, Stuart JM. Molecular Pathways: Extracting Medical Knowledge from High Throughput Genomic Data. Clin Cancer Res. 2013

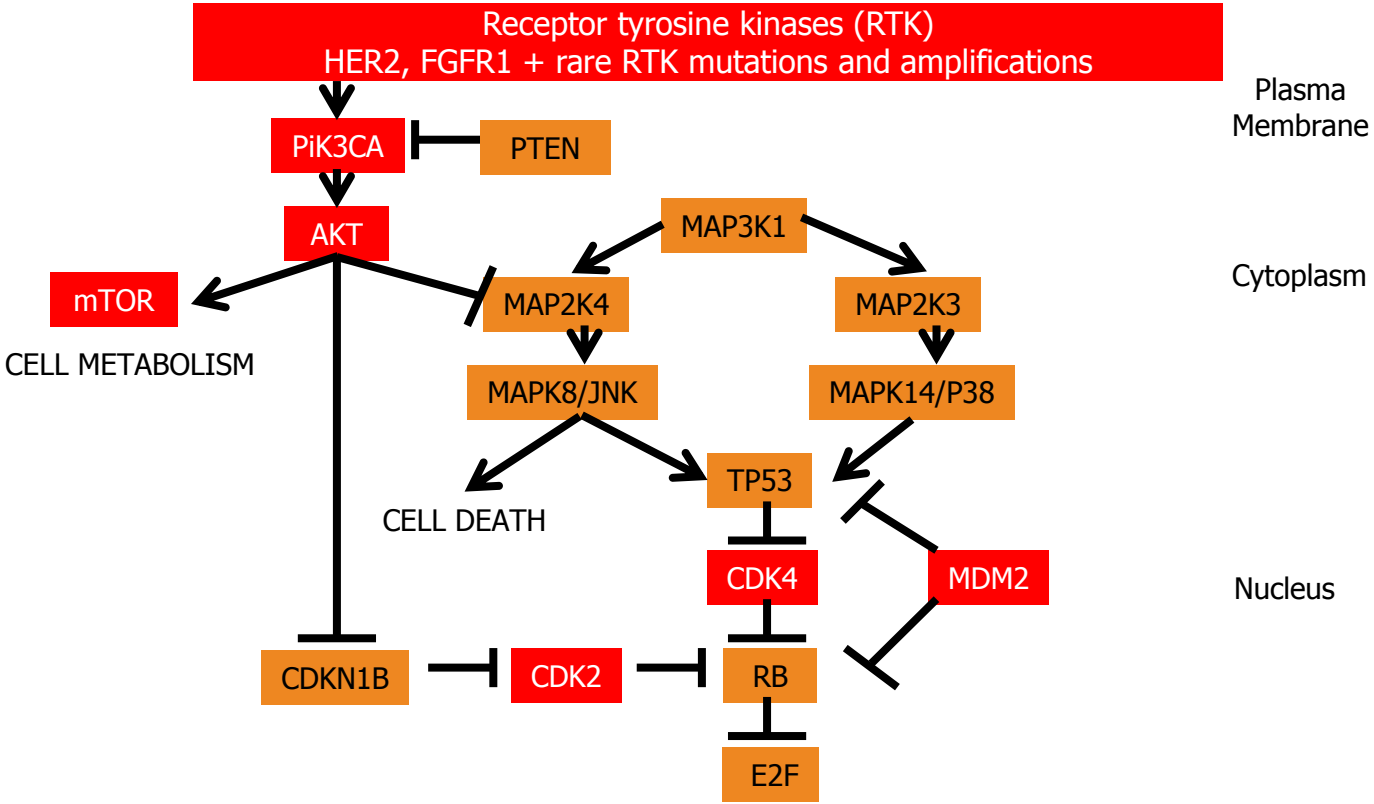
A mutational map of pathways in luminal type breast cancer



Physician Scientist Training Program
Washington University
Brian Van Tine

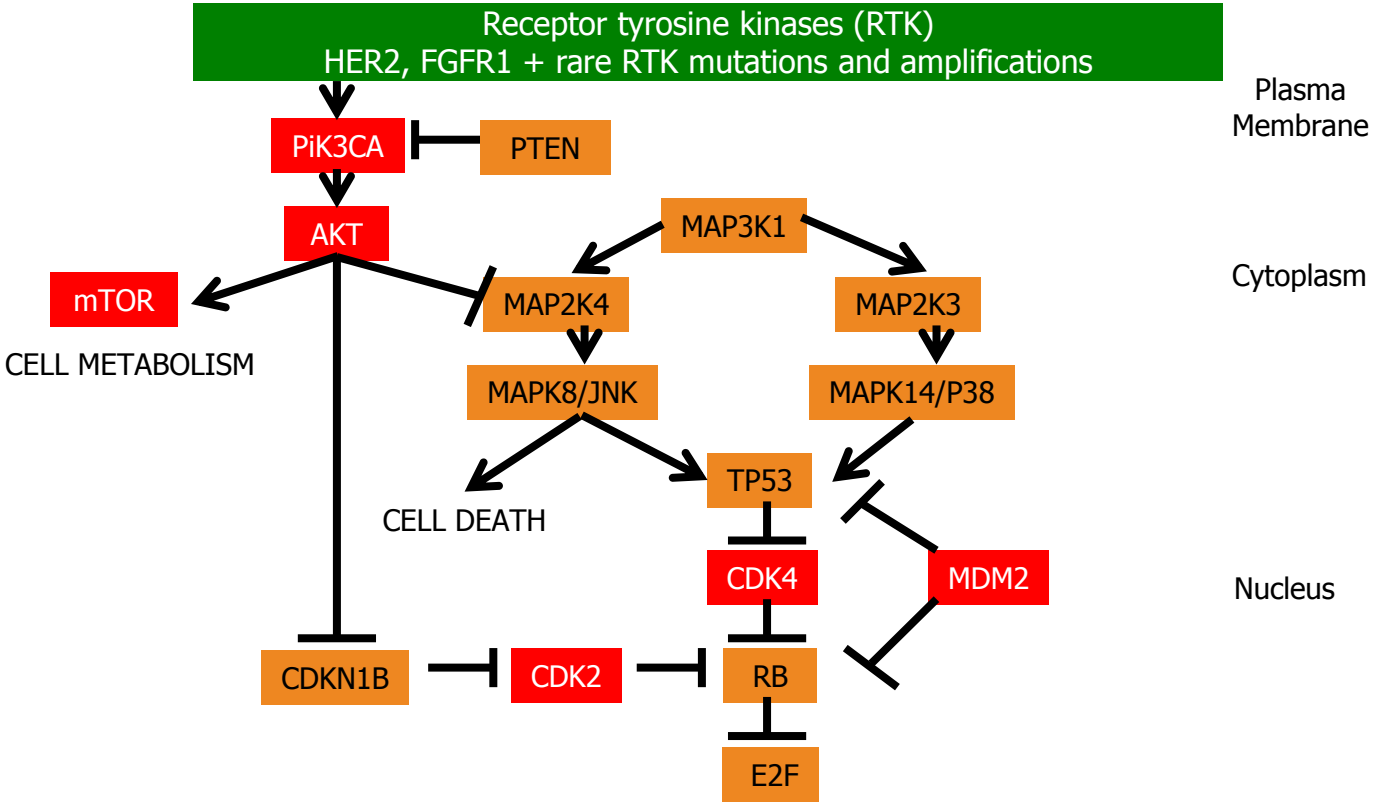
Ellis MJ, et al. *Nature* 2012;**486**:353–360

A therapeutic roadmap for luminal-type breast cancer



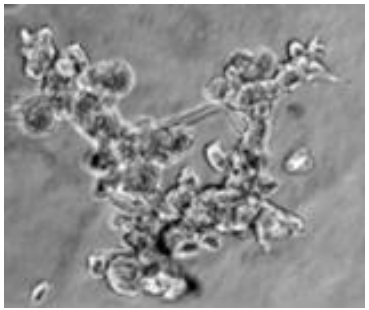
Ellis MJ and Perou CM. *Cancer Discov.* 2013;**3**:27–34

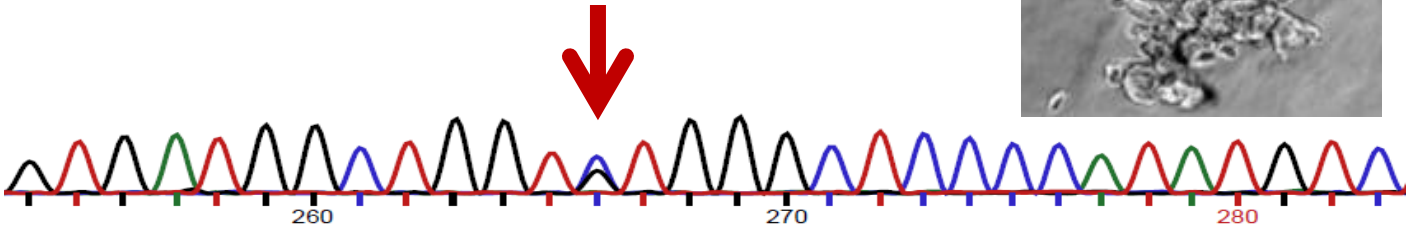
A therapeutic roadmap for luminal-type breast cancer



Ellis MJ and Perou CM. *Cancer Discov.* 2013;**3**:27–34

What do we do with genomic anecdotes?





V777L:

G T G A T G G C T G G T C T G G G C T C C C C A T A T G T C

V M A G L G S P Y V

WT:

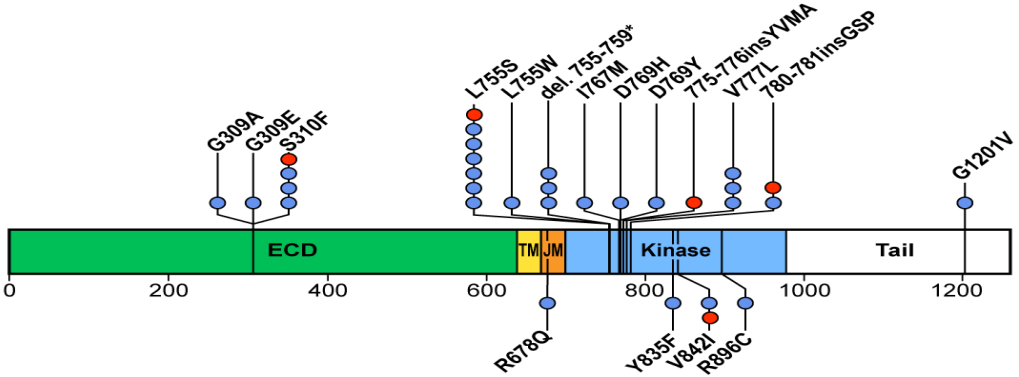
G T G A T G G C T G G T G T G G G C T C C C C A T A T G T C

V M A G V G S P Y V

a.a.773 a.a.777 a.a.782

Recurrent HER2 Somatic Mutations

Shyam Kavuri



Ron Bose

- Blue circle from Bose et al, Red from Ross et al
- From 8 publications with a total of 1,499 patients.
- 20% of patients have mutations at amino acids 309 or 310.
- 68% of patients have mutations at amino acids 755-780.

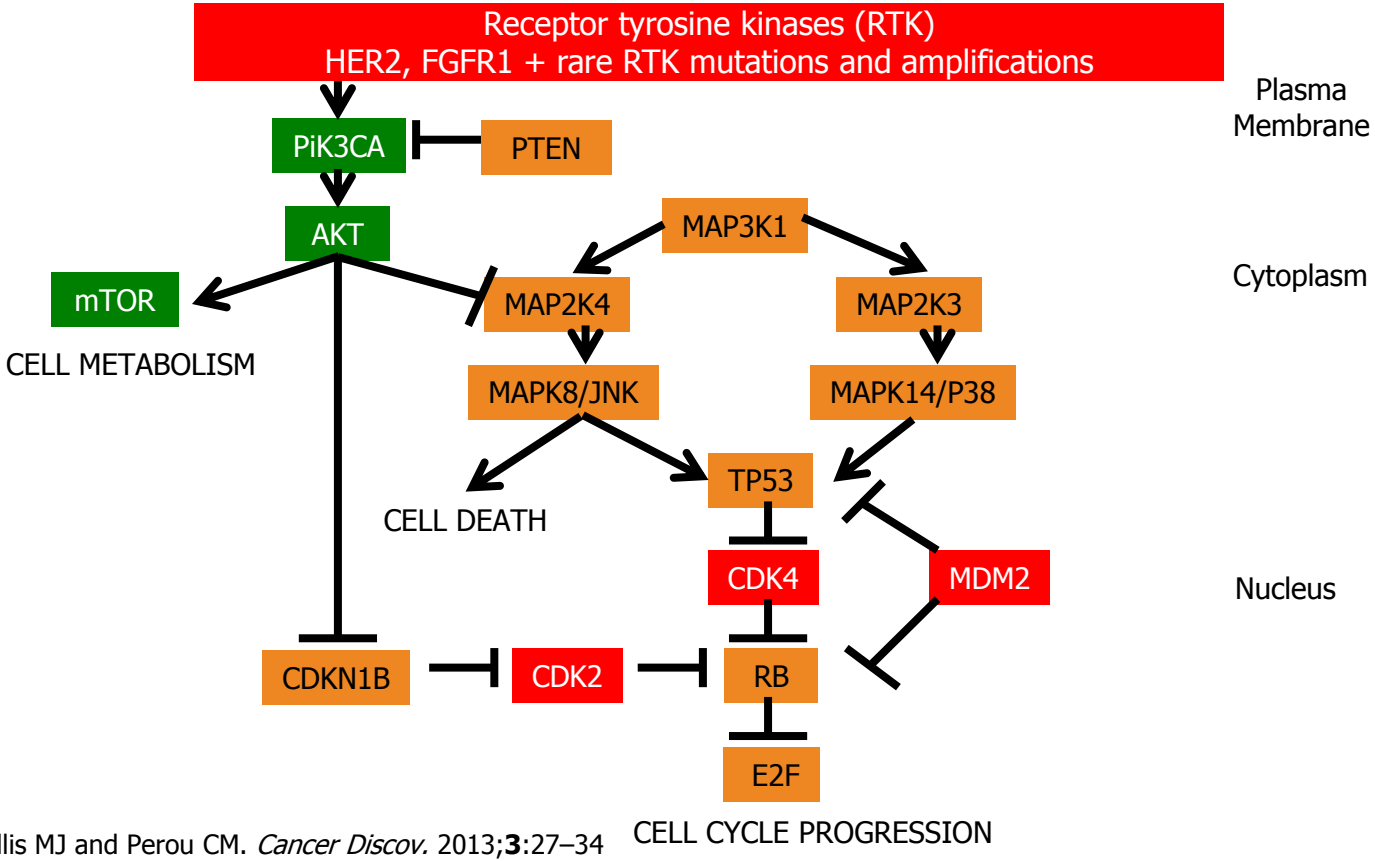
Bose R, et al *Cancer discovery* 3: 224-37, 2013
Ross JS, et al *Clinical cancer research* 19: 2668-76, 2013

dGENE analysis of Luminal Breast Cancers

Kumar RD, et al PLoS One 2013; 8: e67980..

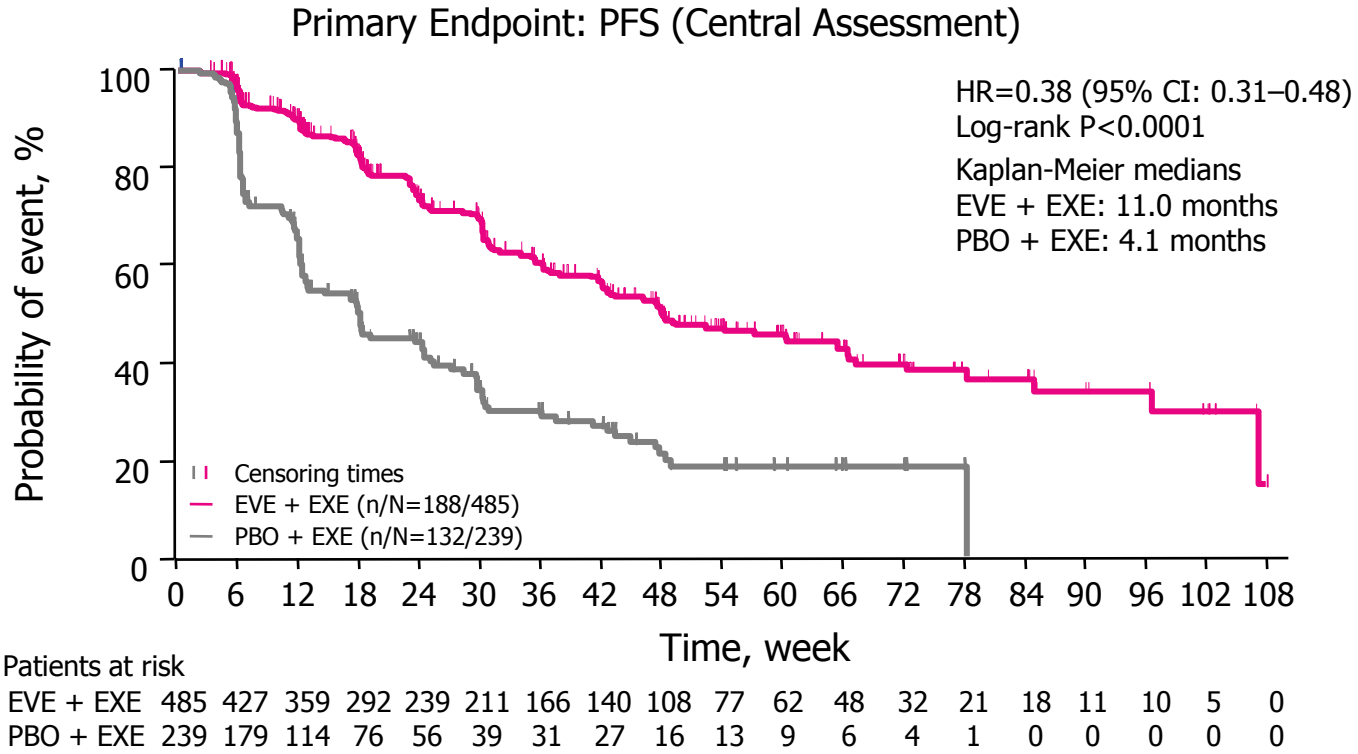
dGene class	NCBI Symbol	Full Name	Patients Affected
ST_KINASE	MAP3K1	mitogen-activated protein kinase kinase kinase 1	9
	ATR	ataxia telangiectasia and Rad3 related	5
	OBSCN	obscurin	3
	SMG1	smg-1 homolog	3
	ALPK2	alpha-kinase 2	2
	BRAF	v-raf murine sarcoma viral oncogene homolog B1	2
	DCLK3	doublecortin-like kinase 3	2
	LRRK2	leucine-rich repeat kinase 2	2
	MAP2K4	mitogen-activated protein kinase kinase 4	2
TYR_KINASE	KIT	c-kit	2
	PDGFRA	platelet-derived growth factor receptor	2
	TEX14	testis expressed 14	2

A therapeutic roadmap for luminal-type breast cancer



Ellis MJ and Perou CM. *Cancer Discov.* 2013;**3**:27–34

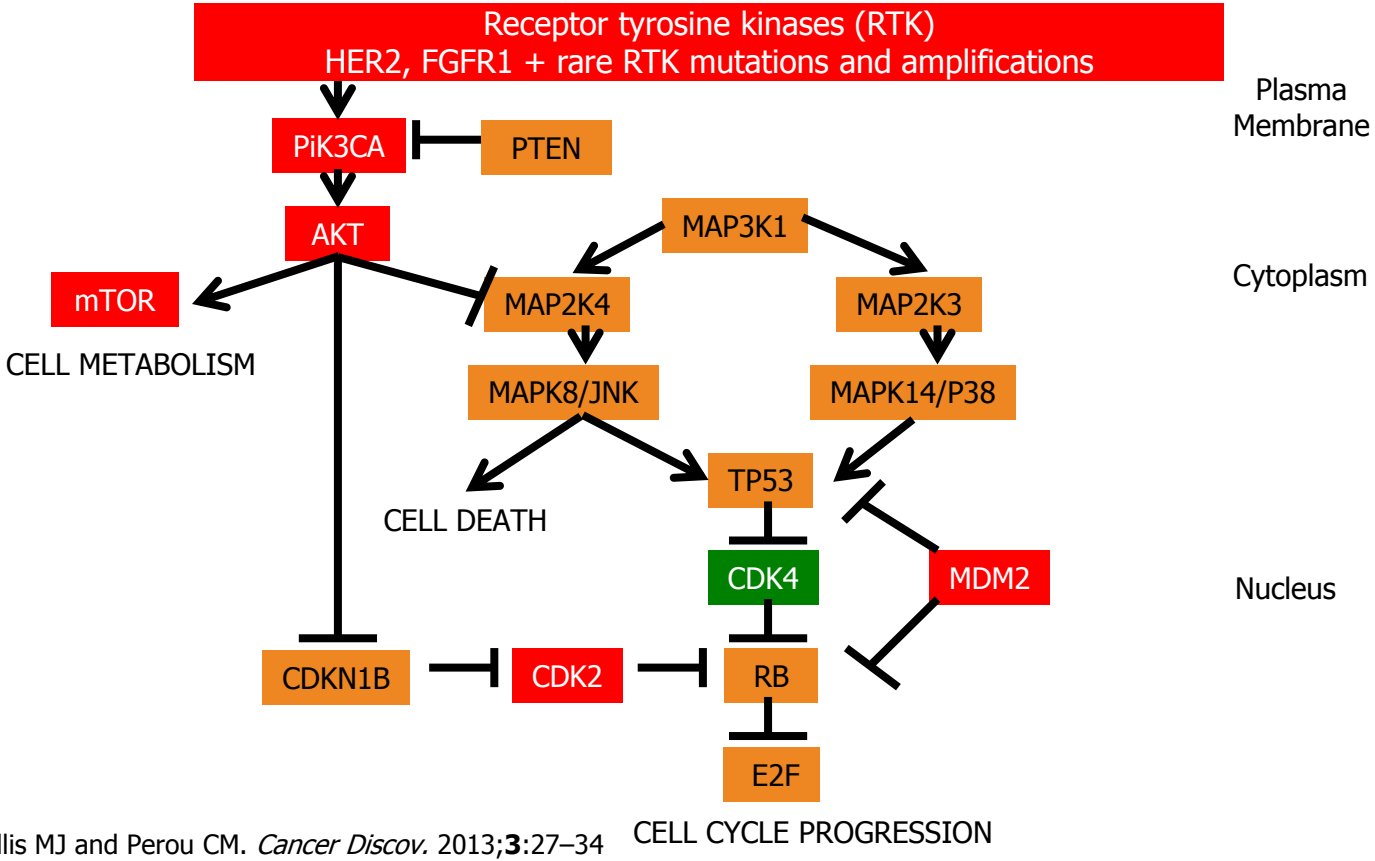
The BOLERO-2 study provides important proof of concept for targeting the mTOR pathway in metastatic breast cancer



CI, confidence interval; EVE, everolimus; EXE, exemestane; HR, hazard ratio; PBO, placebo

Piccart M, et al. Presented at: 2012 San Antonio Breast Cancer Symposium; 4–8 December 2012; Abstract P6-04-02

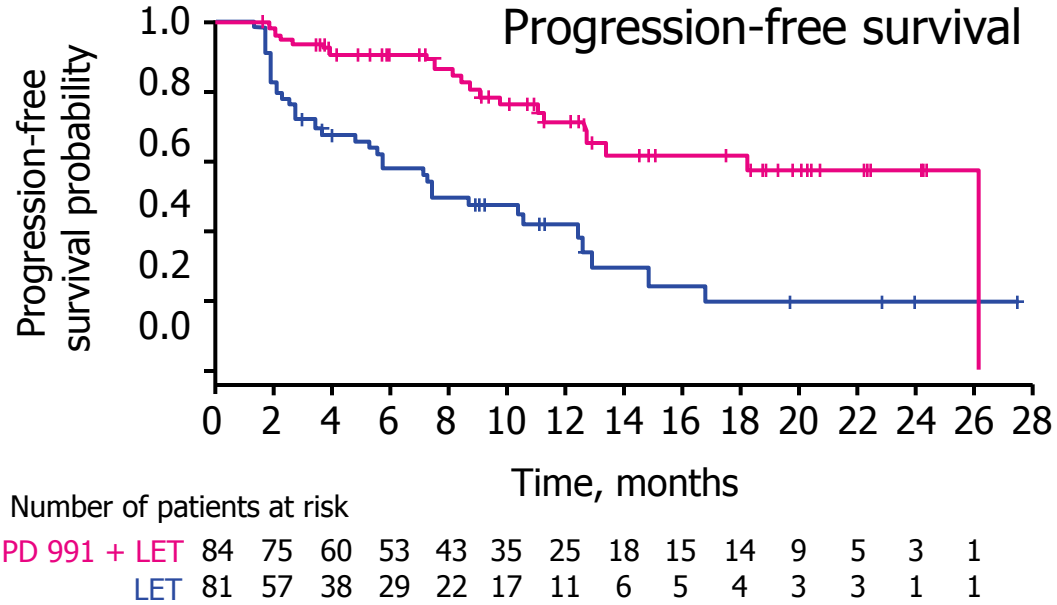
A therapeutic roadmap for luminal-type breast cancer



Ellis MJ and Perou CM. *Cancer Discov.* 2013;**3**:27–34

Inhibition of CDK 4 and 6 kinases for the treatment of ER+ HER– advanced breast cancer

Clinical benefit, including in patients with cyclin D1 amplification and/or p16 loss, using novel compound PD991 in combination with letrozole in a phase II study



	PD 991 + LET (n=84)	LET (n=81)
Number of events (%)	21 (25)	40 (49)
Median PFS, months (95% CI)	26.1 (12.7, 28.1)	7.5 (5.6, 12.6)
Hazard ratio (95% CI)	0.37 (0.21, 0.63)	
P value	<0.001	

Finn RS, et al. *Cancer Res.* 2012;**72**(24 Suppl):5s – Presented at 2012 CTRC-AACR San Antonio Breast Cancer Symposium (Abstract #S1–6)

Primary endpoint: pCR rate

Clinical Stage II or III ER+ (Allred 6-8) HER2- Breast Cancer

BIOPSY

Cycle 0
(days -28 to -1)
Anastrozole

Tumor *PIK3CA* Mutation Analysis

Mutation Present

16 weeks (4 x 28-day Cycle)

AKT inhibitor Trial
MK-2206 PO (Days 1, 8, 15, 22)
+ Anastrozole PO Daily

SURGERY

2-week
Biopsy for Ki67

Ki67 > 10%
Surgery or Chemotherapy at the discretion of treating physician

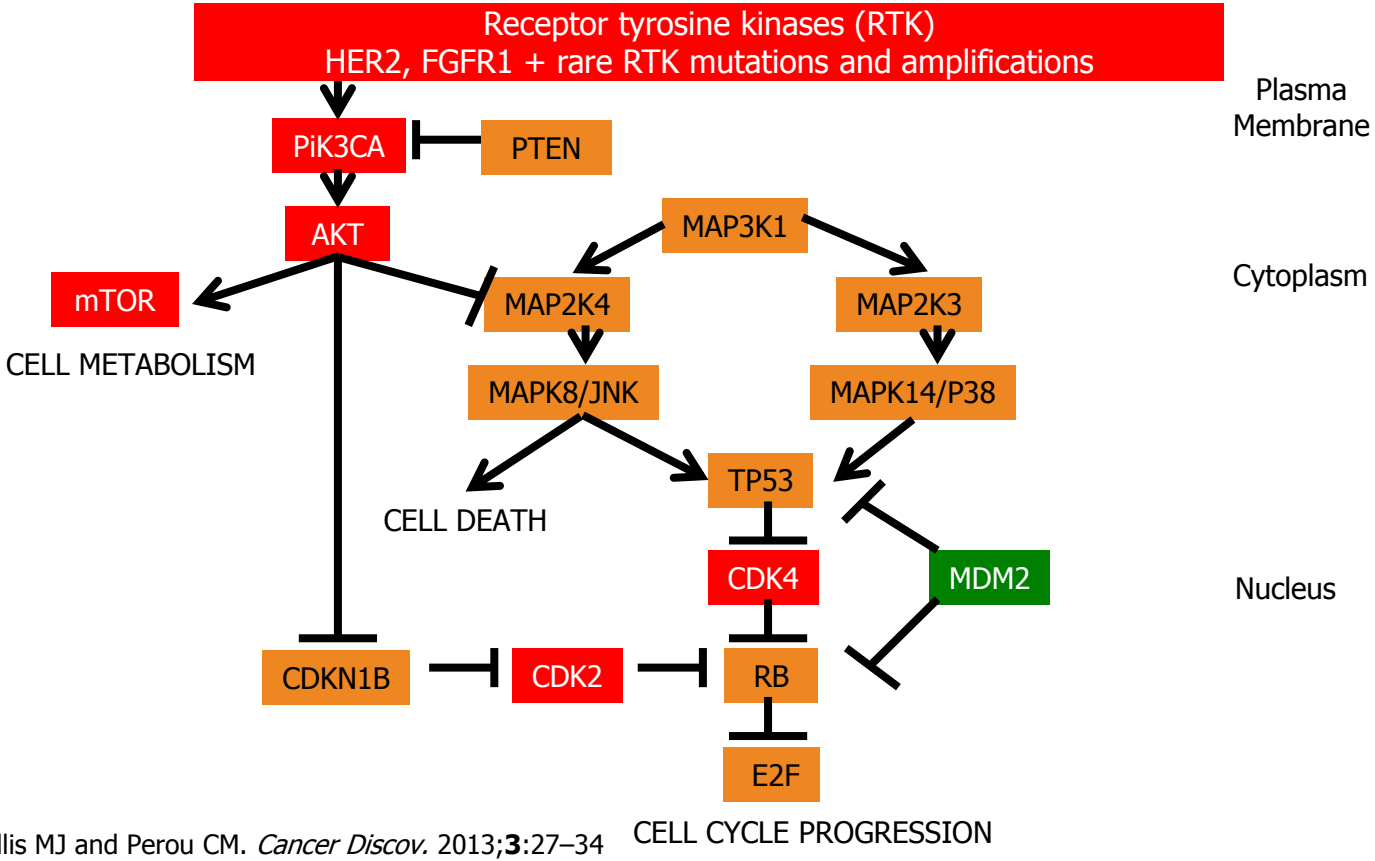
Mutation Absent

Cdk4/6 inhibitor Trial
PD991 PO (Days 1-21) x 4 cycles
+ Anastrozole PO Daily

SURGERY

2 stage design: 1st stage: n=16. 2nd stage: n=13

A therapeutic roadmap for luminal-type breast cancer



Ellis MJ and Perou CM. *Cancer Discov.* 2013;**3**:27–34

Cell PRESS
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Cell Reports
Article

Endocrine-Therapy-Resistant *ESR1* Variants Revealed by Genomic Characterization of Breast-Cancer-Derived Xenografts

Shunqiang Li,[1,2,13] Dong Shen,[3,13] Jieya Shao,[1] Robert Crowder,[1] Wenbin Liu,[4] Aleix Prat,[5,6] Xiaping He,[6] Shuying Liu,[4] Jeremy Hoog,[1] Charles Lu,[3] Li Ding,[2,3,9] Obi L. Griffith,[3] Christopher Miller,[3] Dave Larson,[3] Robert S. Fulton,[3] Michelle Harrison,[3] Tom Mooney,[3] Joshua F. McMichael,[3] Jingqin Luo,[2,7] Yu Tao,[7] Rodrigo Goncalves,[1] Christopher Schlosberg,[8] Jeffrey F. Hiken,[8] Laila Saied,[9] Cesar Sanchez,[10] Therese Giuntoli,[1] Caroline Bumb,[1] Crystal Cooper,[1] Robert T. Kitchens,[1] Austin Lin,[1] Chanpheng Phommaly,[1] Sherri R. Davies,[1] Jin Zhang,[3] Megha Shyam Kavuri,[1] Donna McEachern,[11] Yi Yu Dong,[1] Cynthia Ma,[1,2] Timothy Pluard,[1,2] Michael Naughton,[1,2] Ron Bose,[1,2] Rama Suresh,[1] Reida McDowell,[1] Loren Michel,[1,2] Rebecca Aft,[12] William Gillanders,[12] Katherine DeSchryver,[1] Richard K. Wilson,[2,3] Shaomeng Wang,[11] Gordon B. Mills,[4] Ana Gonzalez-Angulo,[4] John R. Edwards,[8] Christopher Maher,[1,2,3] Charles M. Perou,[6] Elaine R. Mardis,[2,3] and Matthew J. Ellis[1,2,*]

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Jackie Snider
Sherri Davies
Katherine DeSchryver
Jieya Shao
Triparna Sen
Chanpheng Phommaly
Robert Crowder
Rodrigo Goncalves

HAMLET core
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Tom Kitchens