Pharmacogenomics: Science, Challenges and Promises

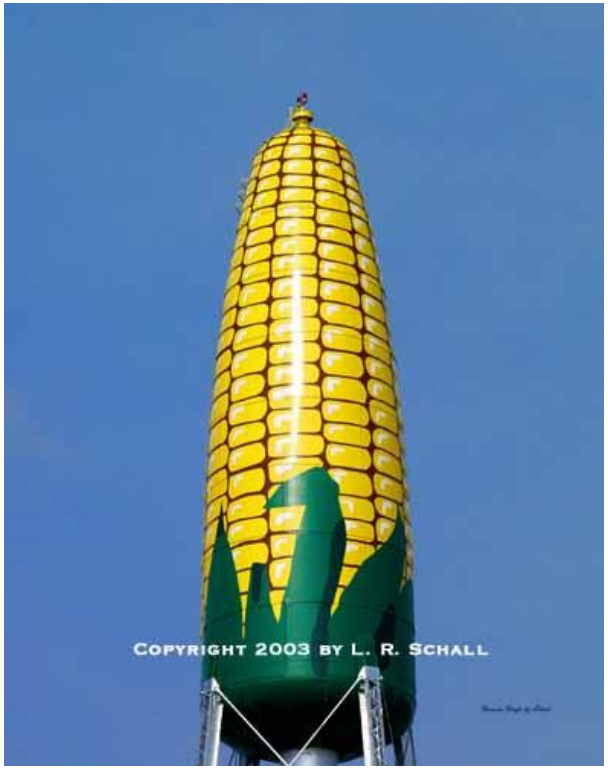
Liewei Wang M.D.,PhD.

Professor,

Department of Molecular Pharmacology and Experimental Therapeutics (MPET)

Mayo Clinic Rochester

From Shanghai to Rochester



Pharmacogenomics

- A study of the role of inheritance in variation of drug response.
- A critical component of precision medicine.
- Both tumor DNA and germline DNA can contribute to response to anticancer therapy.

Research Path

Functional characterization of SNPs in drug metabolizing enzymes

Pharmacogenetics. 2003 Sep;13(9):555-64;
Proc Natl Acad Sci U S A. 2005 Jun 28;102(26):9394-9
J Biol Chem. 2006 Mar 17;281(11):7364-73.
Clin Cancer Res. 2008 Jun 1;14(11):3503-13.

Application of cell line models and patient DNA, together with genome wide association approach and functional studies

Cancer Res. 2008 Sep 1;68(17):7050-8.
Hum Mol Genet. 2008 Oct 15;17(R2):R174-9.
Cancer Cell. 2009 Sep 8;16(3):259-66.
Cell Cycle. 2010 Jan 1;9(1):6-7.
J Clin Oncol. 2010 Nov 1;28(31):4674-82.
Genome Res. 2010 Nov;20(11):1482-92.
N Engl J Med. 2011 Mar 24;364(12):1144-53.
Curr Opin Pharmacol. 2011 Aug;11(4):360-4
PLoS One. 2012;7(5):e36252.
Cancer Discov. 2013 Jul;3(7):812-25.
Genome Res. 2013 Sep;23(9):1363-72.
PLoS One. 2015 Mar 5;10(3):e0119208
Genes Dev. 2015 Nov 1;29(21):2244-57.
Mol Cell. 2016 Feb 18;61(4):614-24

Application of animal models and cell lines, together with high throughput omics and functional studies in PGx of anti-cancer

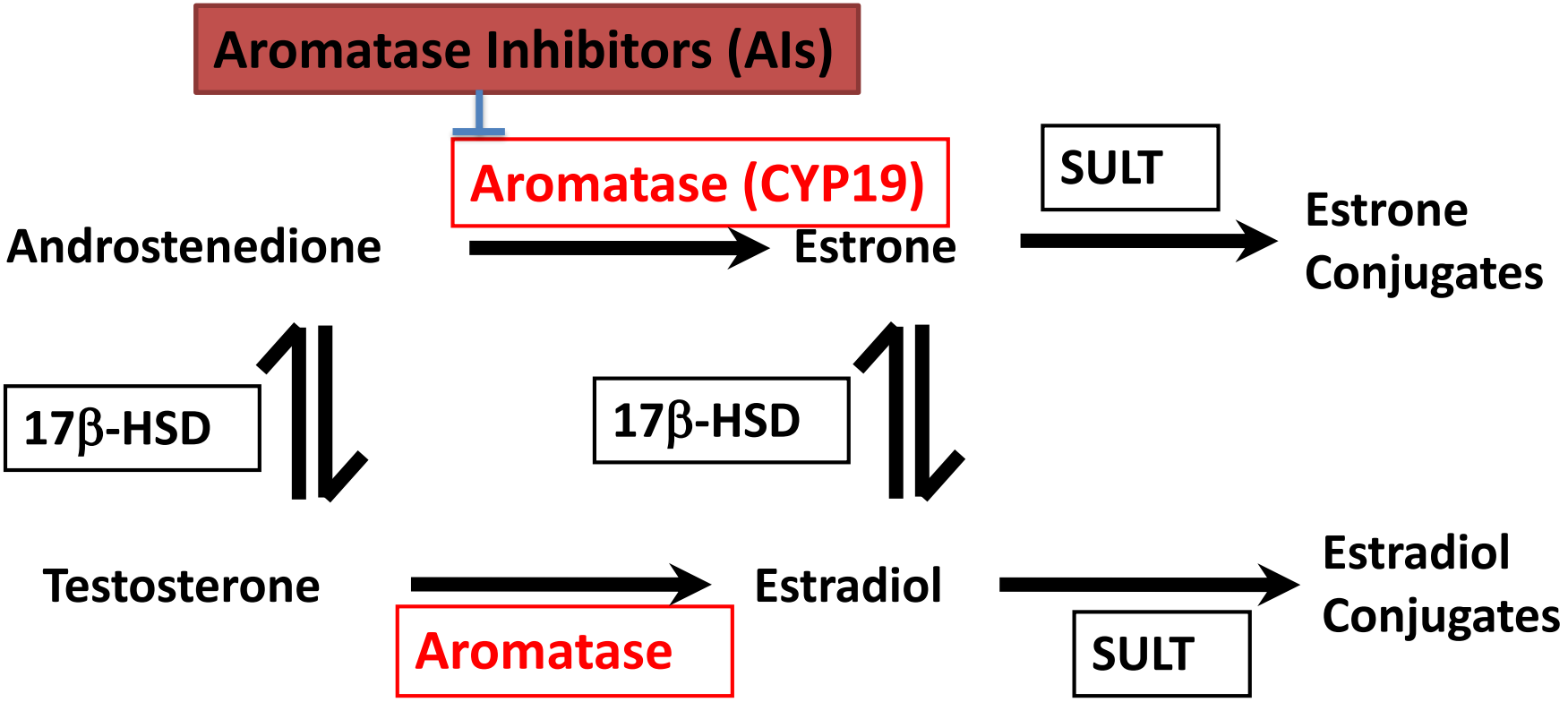
BEAUTY study
PROMOTE study

ER+ Breast Cancer Treatment

- 70-80% of all breast cancer patients are ER+
- Endocrine Therapy – AIs
- Endocrine Therapy – SERMs
- Both can be used in the prevention setting
- Third generation AIs include:
  - Irreversible steroidal inhibitors, such as exemestane
  - Non-steroidal reversible inhibitors, such as anastrozole and letrozole

Aromatase Inhibitors (AIs)

- AIs are the mainstay of treatment of ER+ breast cancer patients, which constitute 70-80% of all breast cancer patients.



Aromatase Inhibitors (AIs)

- **Response to AI varies greatly, ranging from AI-induced ADE to lack of efficacy—disease recurrence.**
- **AI response in women with high BMI is lower relative to tamoxifen and BMI is directly associated with circulating estrogen levels.**

Supports hypothesis that PD effect of AIs is a determinant of efficacy.

Does the germline genome contribute to AI response through its PD effect, i.e., reduction in estrogen level?

Clinical Studies

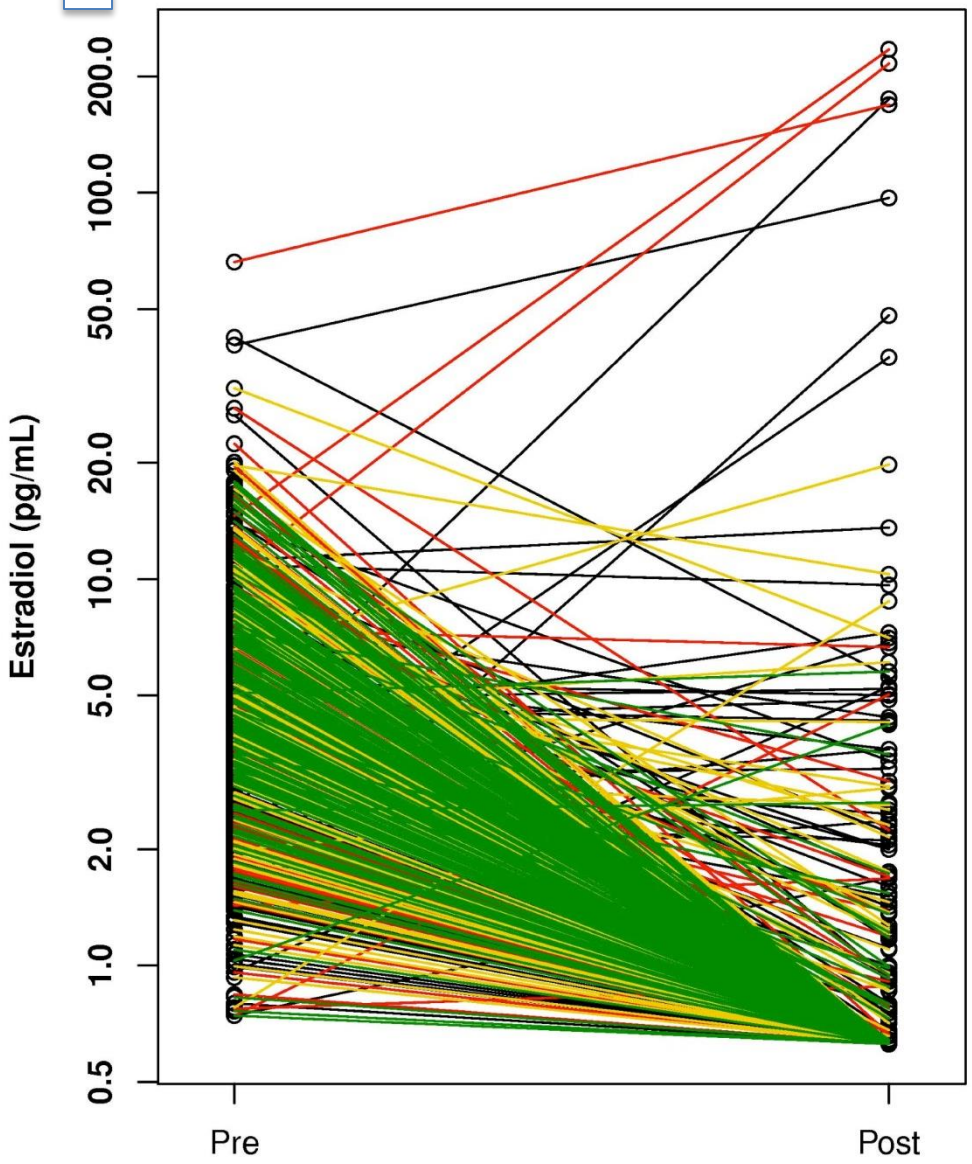
Mayo, MD Anderson and MSK trial: ≈800 women with primary breast cancer treated with anastrozole in the adjuvant setting

- **Obtain plasma for hormone assays and blood for DNA**
- **GWAS genotype with Illumina Human610-Quad BeadChip**

Estradiol: Baseline and on Anastrozole

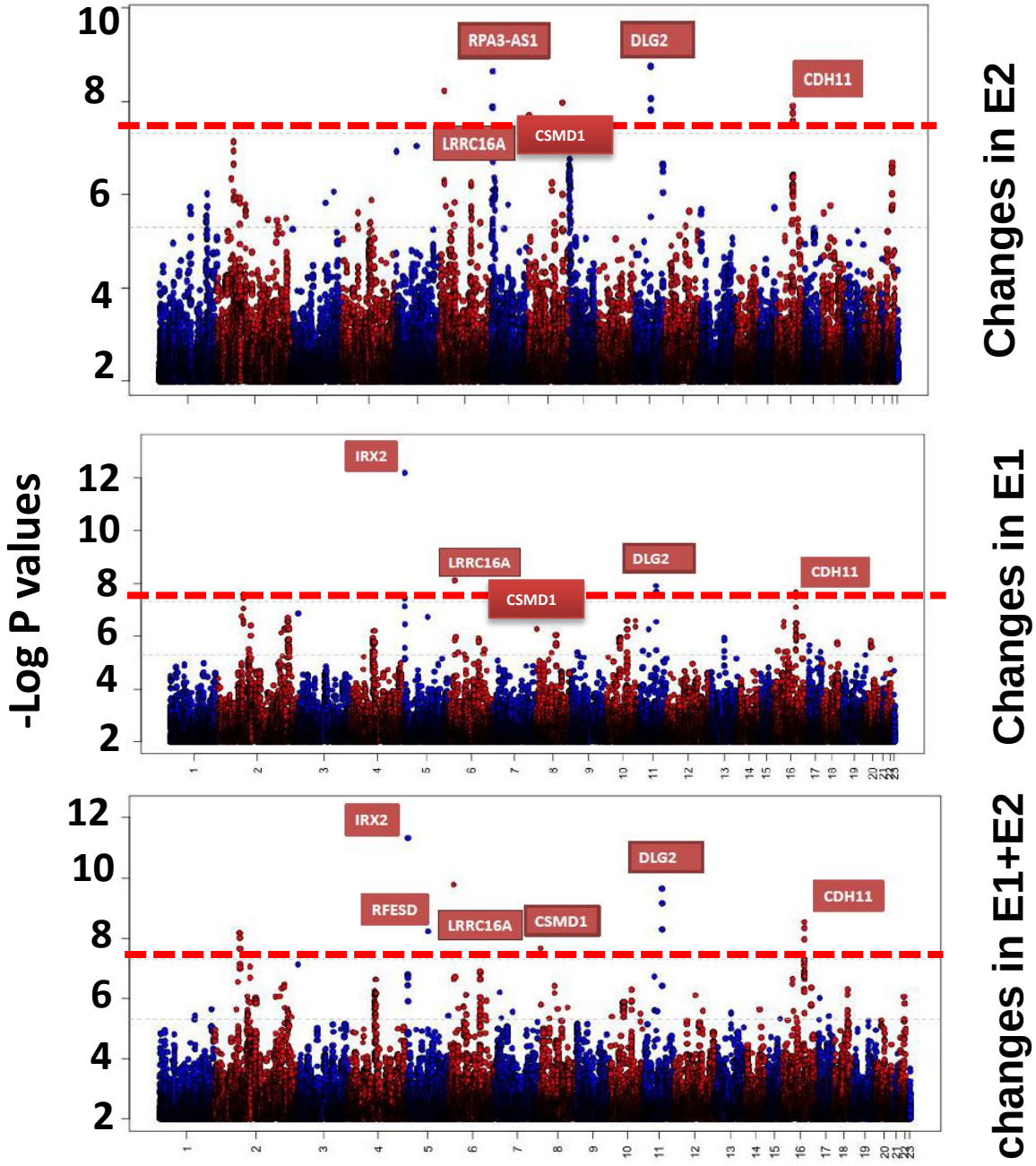
n=643; Lower Limit of Quantitation, 0.625 pg/ml

Anastrozole Concentration
Quartile:
Black: Lowest
Red: Second
Yellow: Third
Green: Highest



Ingle et al. Steroids Epub 8/24/2014

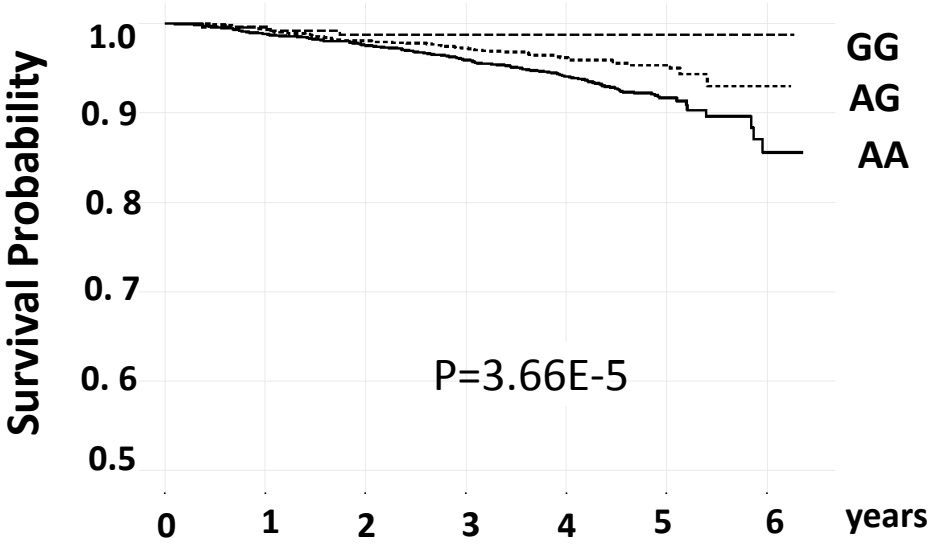
GWAS of changes in estrogen levels pre and post AI treatment



NCIC-CTG MA27

- **Phase III trial comparing exemestane and anastrozole, N=4800.**
- **GWAS with multiple phenotypes including breast events.**

Kaplan-Meier Curve for Time to Breast Event by rs6990851 Genotype



Numbers at risk

AA	2805	2733	2606	2433	1769	580	44
AG	1599	1560	1486	1417	1031	354	21
GG	250	245	230	216	159	45	1

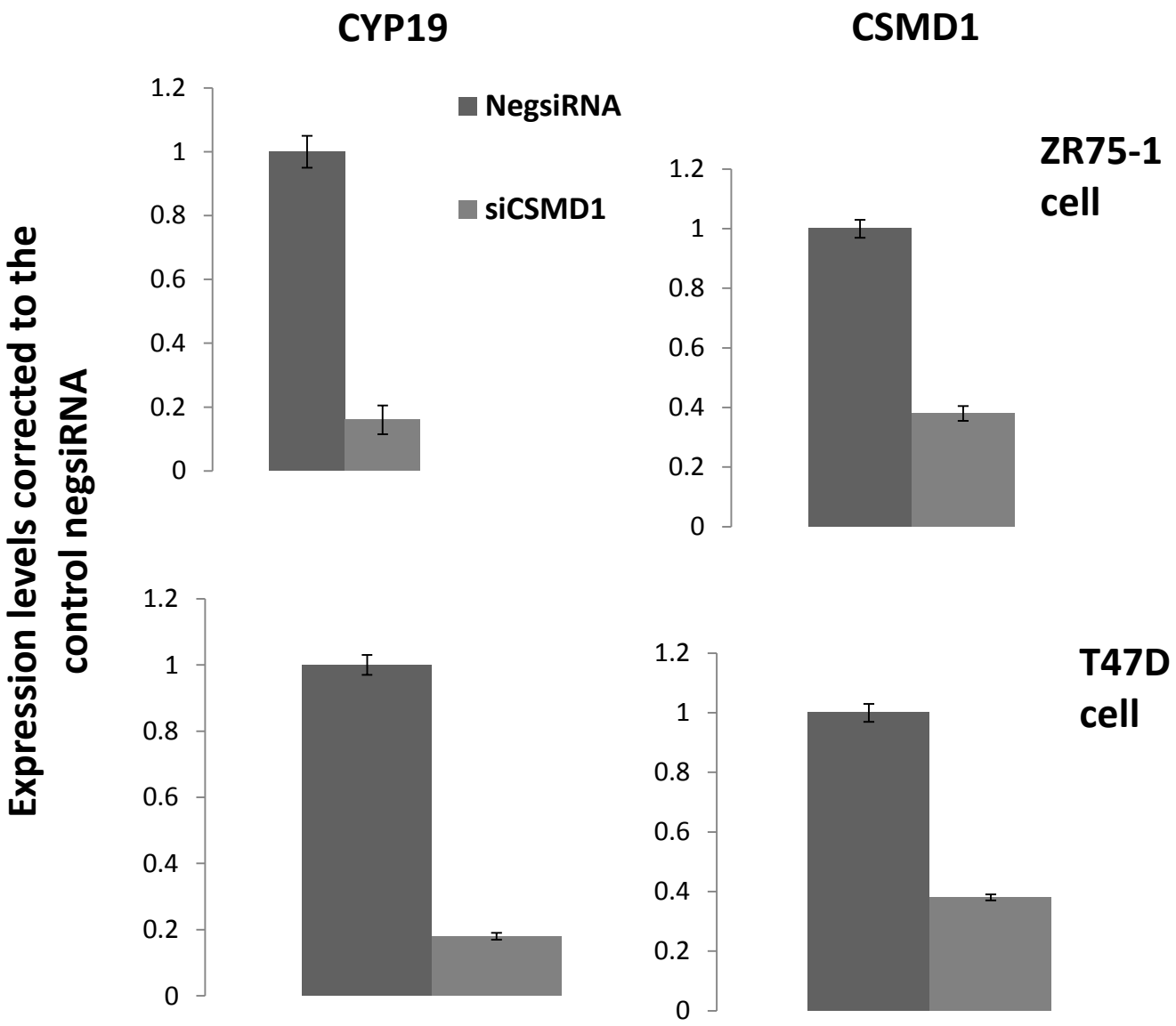
SNP association with time to distant metastasis

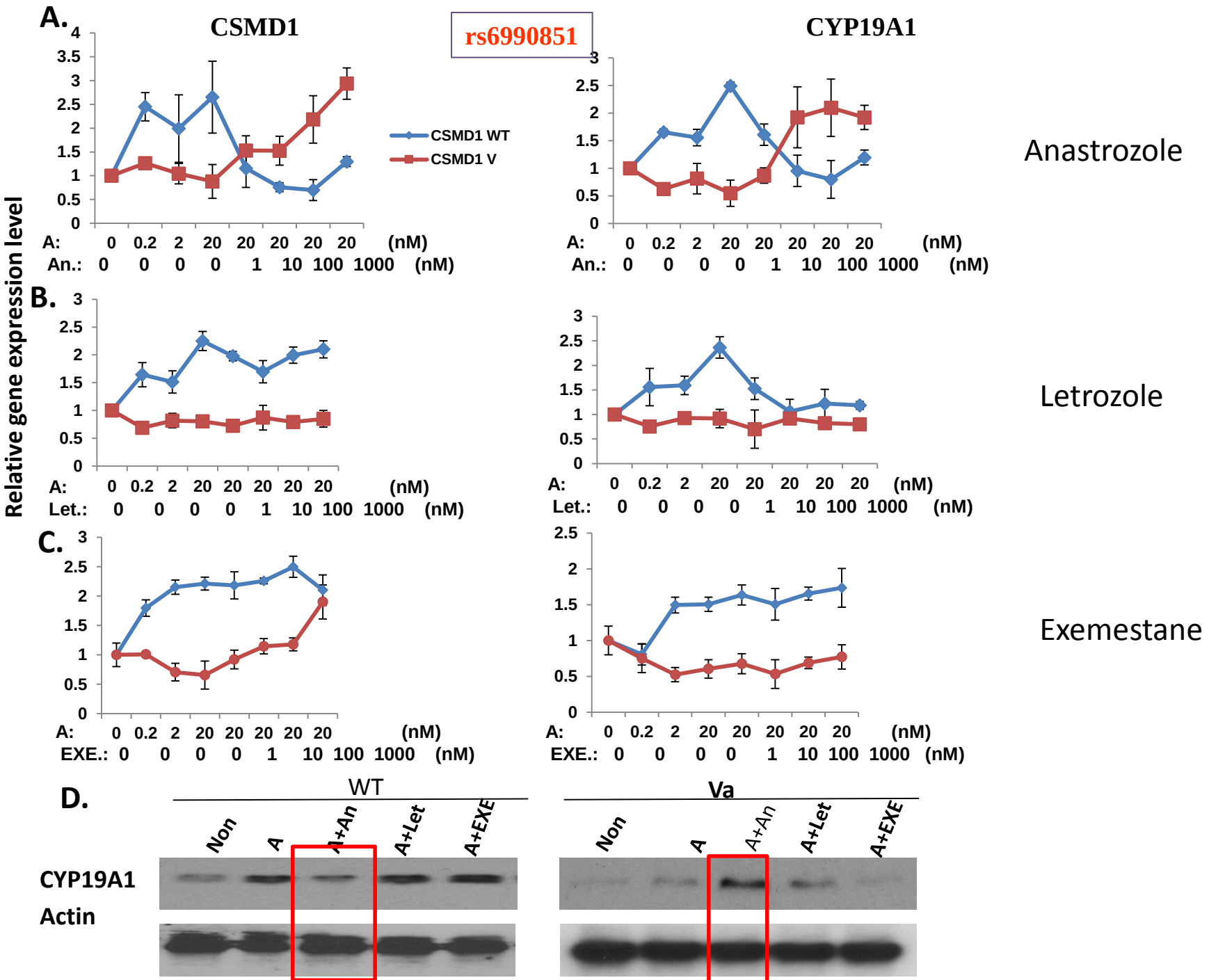
SNP	Minor Allele	HR	l95	U95	P	Sample
rs6990851	G	0.5947	0.436	0.8112	0.00104	All
rs6990851	G	0.4861	0.304	0.7772	0.00259	Anastrozole Arm
rs6990851	G	0.7145	0.4726	1.08	0.111	Exemestane Arm

CSMD1: CUB And Sushi Multiple Domains 1

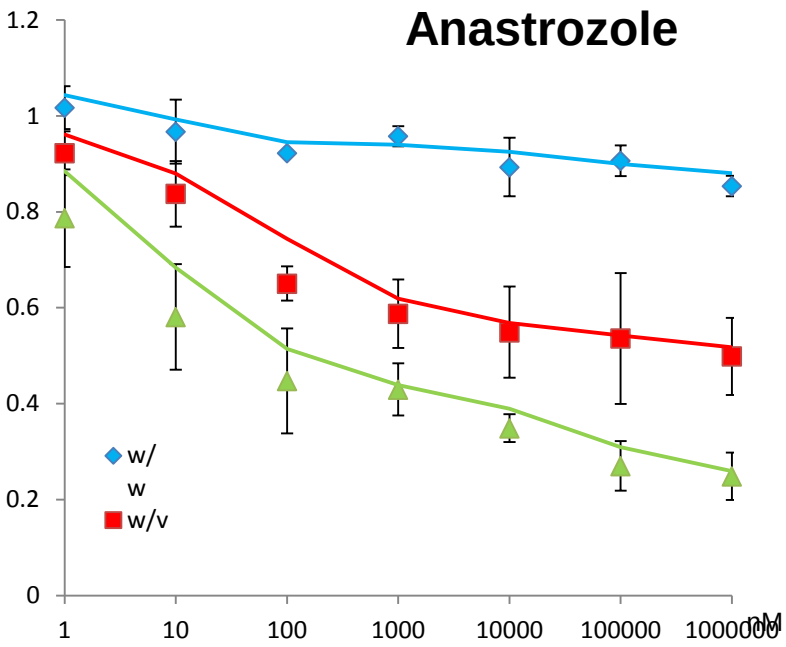
- In breast cancer, loss of CSMD1 is associated with high tumor grade and poor survival, suggesting that CSMD1 is a tumor suppressor gene.
- Promotes degradation of C4b & C3b, inhibits membrane attack complex assembly.
- SNPs within this gene associate with multiple Neuronal pathologic conditions.

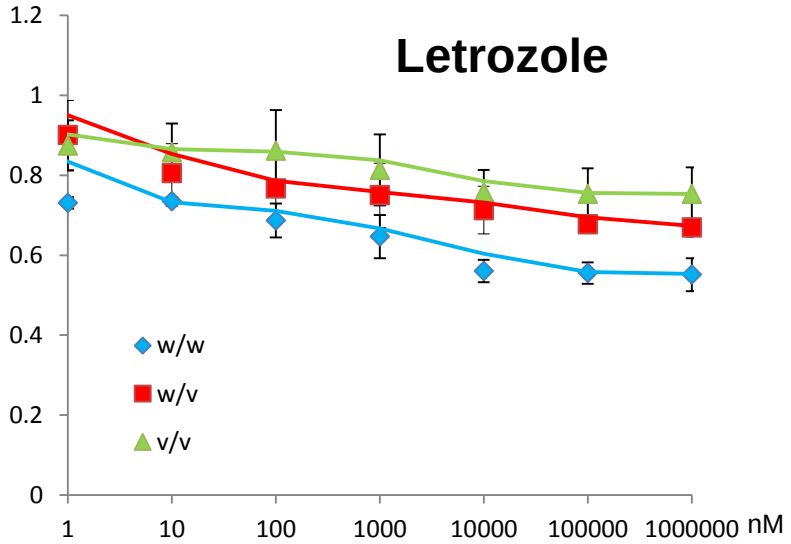
CSMD1 regulates CYP19 expression

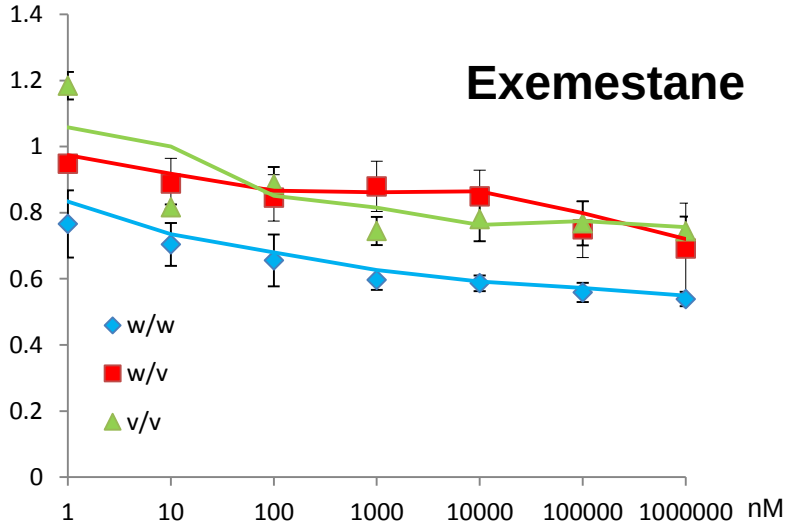



A.
CSMD1
rs6990851
CYP19A1
CSMD1 WT
CSMD1 V
Relative gene expression level
A: 0 0.2 2 20 20 20 20 20 (nM)
An.: 0 0 0 0 1 10 100 1000 (nM)
Anastrozole
B.
Let.: 0 0 0 0 1 10 100 1000 (nM)
Letrozole
C.
EXE.: 0 0 0 0 1 10 100 1000 (nM)
Exemestane
D.
WT
Va
Non
A
A+An
A+Let
A+EXE
CYP19A1
Actin

CSMD1 SNP Dependent Effect on Aromatase Inhibitor Response







Conclusions

- **Response to anastrozole, but not letrozole and exemestane is SNP dependent.**
- **Our finding indicates additional mechanisms associated with anastrozole mechanisms of action.**
- **Germline genetic variation can contribute to the drug response not only by affecting pharmacokinetics genes but also pharmacodynamics genes.**

PGx of Chemotherapy in Breast Cancer : Application of NextGen Sequencing Analysis and PDX Models

- **Systemic chemotherapy reduces the annual odds of disease recurrence, breast cancer mortality, and all-cause mortality by 24%, 15% and 14.9% , respectively.**

- **There is great heterogeneity in response to chemotherapy.**

- **There are no specific biomarkers for anthracycline–taxane-based therapy.**

BEAUTY
(Breast Cancer Genome Guided Therapy)

PIs: Matthew Goetz, Judy Boughey

Lab PI: Liewei Wang

Lab team: Eric Wieben, Dick Weinshilboum, James Ingle
Bowen Gao, Jia Yu, Minetta Liu

Pathology team: Dan Visscher, Ann Moyer

Radiology team: Amy Conners, Katie Jones

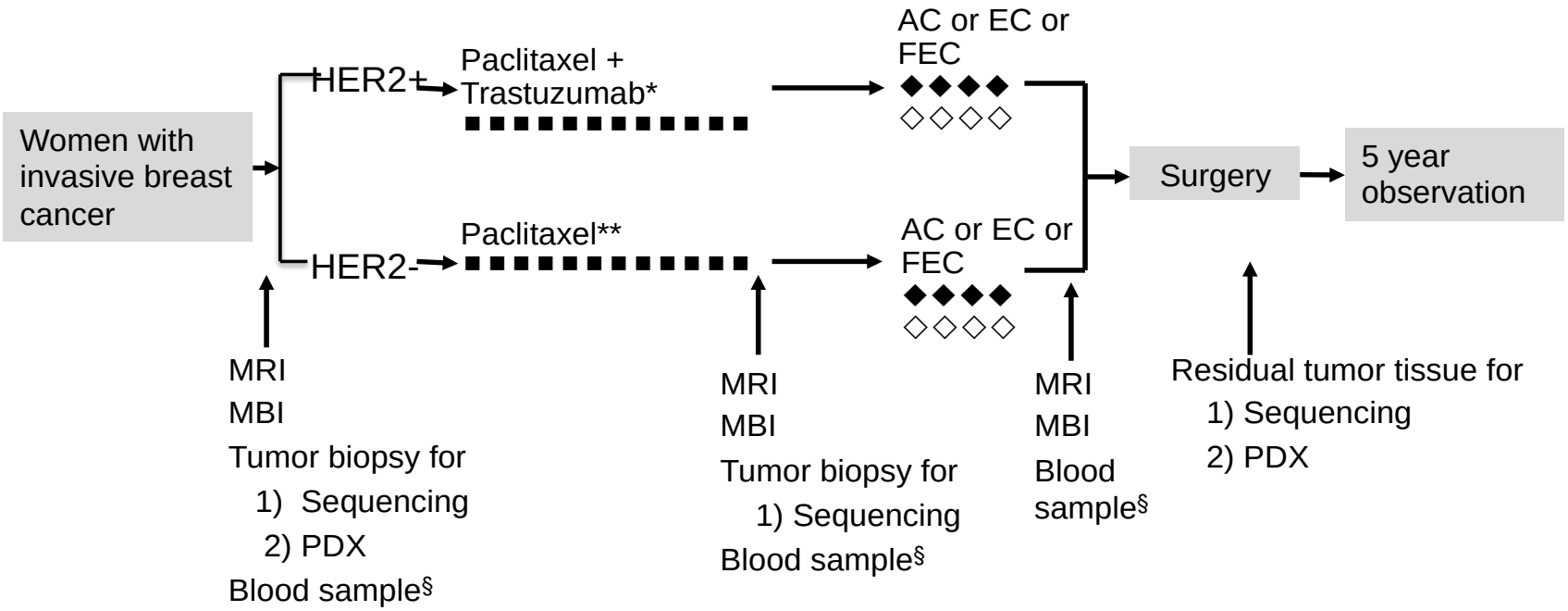
Genetic counselor: Marissa Ellingson

Stats team: Jeanette Eckel Passow, Vera Suman, Travis Dockter, Krishna Kalari, Steve Hart, Hugues Sicottes, Jason Sinnwell

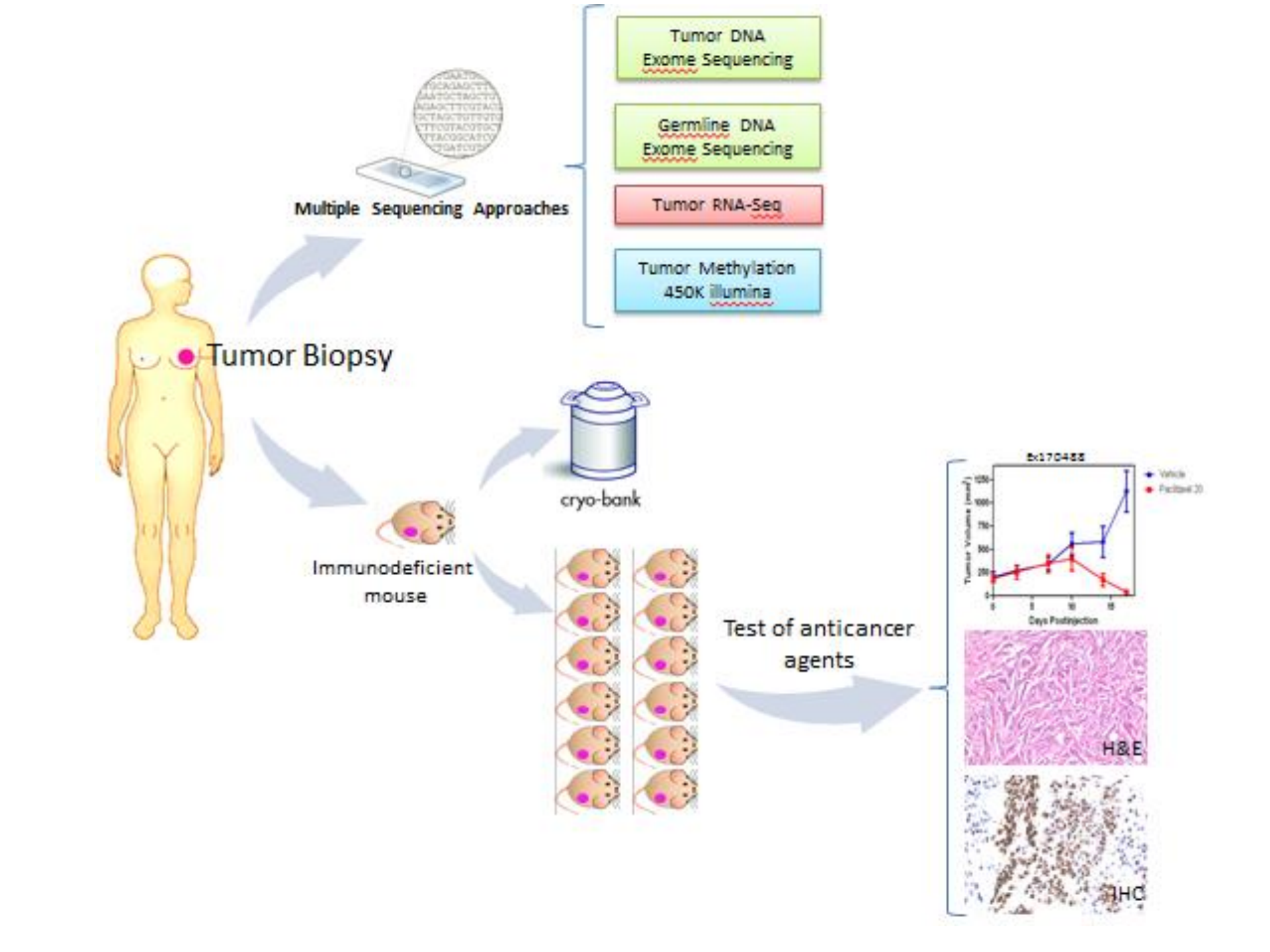
Arizona team: Don Northfelt, Rick Gray

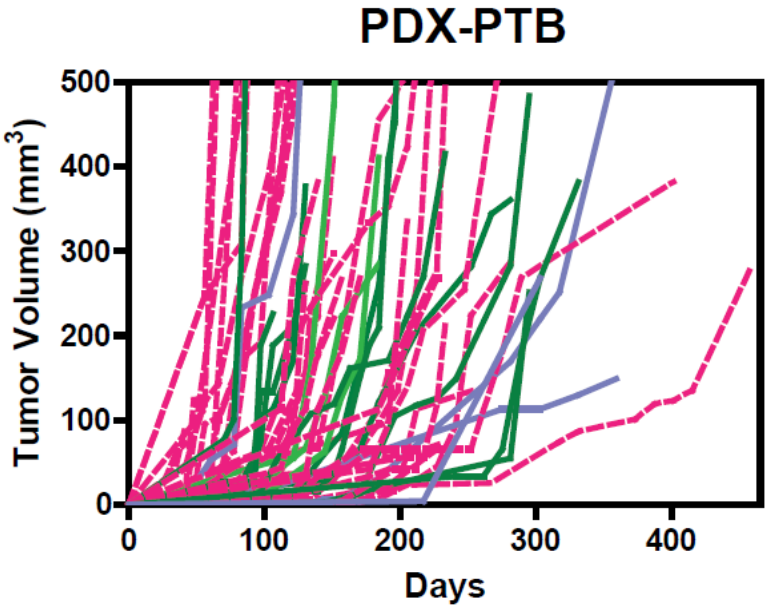
Florida team: Alvaro Moreno, Sarah McLaughlin

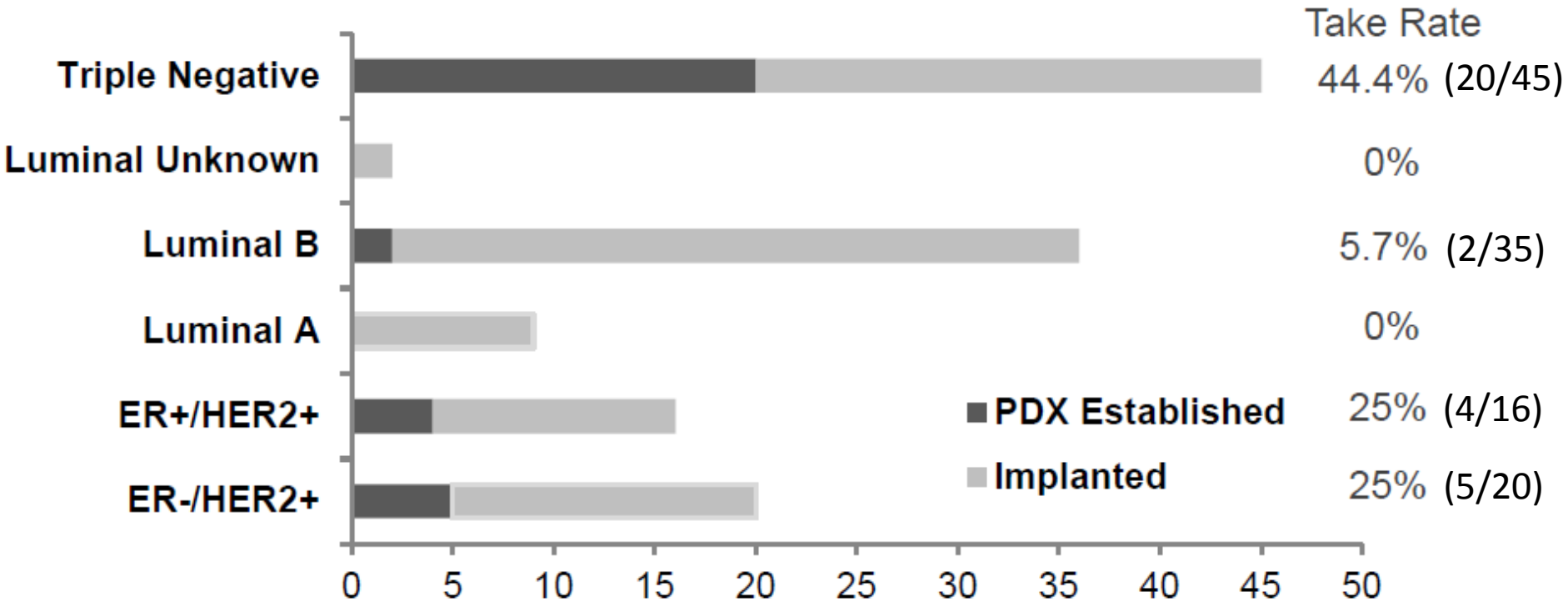
Breast Cancer Genome Guided Therapy Study (BEAUTY)



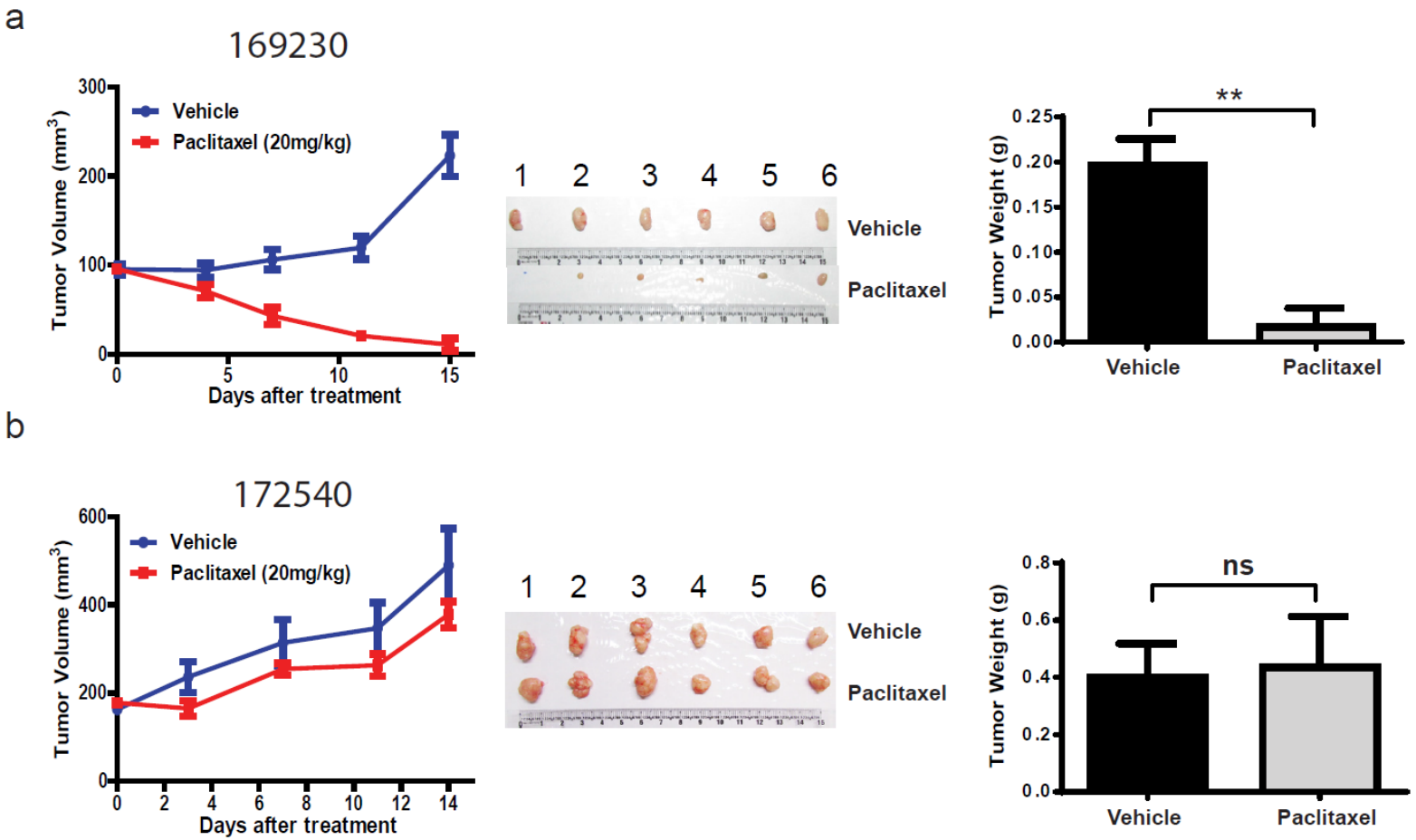
*Pertuzumab allowed for HER2+ disease after 10/18/13
**Carboplatin allowed for TN disease after 3/5/14
§For sequencing, biomarkers, CTCs, and cell-free DNA


Tumor DNA
Exome Sequencing
Germline DNA
Exome Sequencing
Tumor RNA-Seq
Tumor Methylation
450K illumina
Multiple Sequencing Approaches
Tumor Biopsy
cryo-bank
Immunodeficient
mouse
Test of anticancer
agents
H&E
IHC

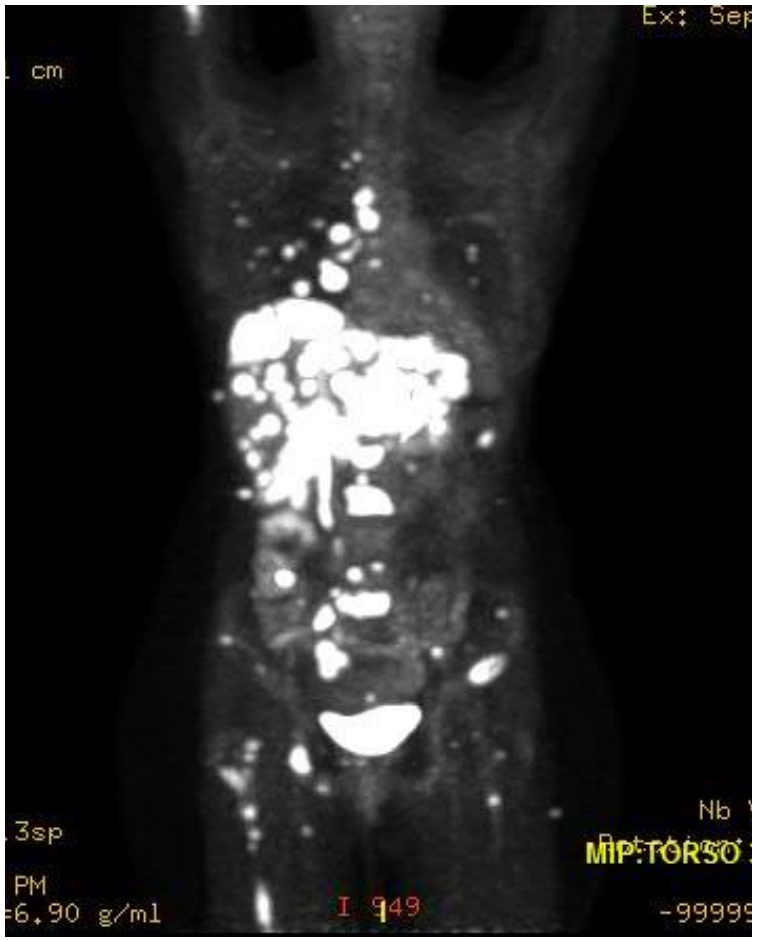

PDX-PTB
Tumor Volume (mm3)
500
400
300
200
100
0
0
100
200
300
400
Days

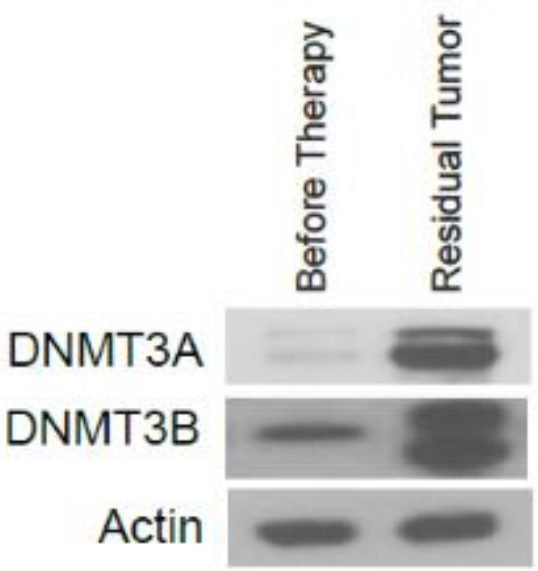

Take Rate
Triple Negative
44.4% (20/45)
Luminal Unknown
0%
Luminal B
5.7% (2/35)
Luminal A
0%
ER+/HER2+
25% (4/16)
ER-/HER2+
25% (5/20)
PDX Established
Implanted
0
5
10
15
20
25
30
35
40
45
50

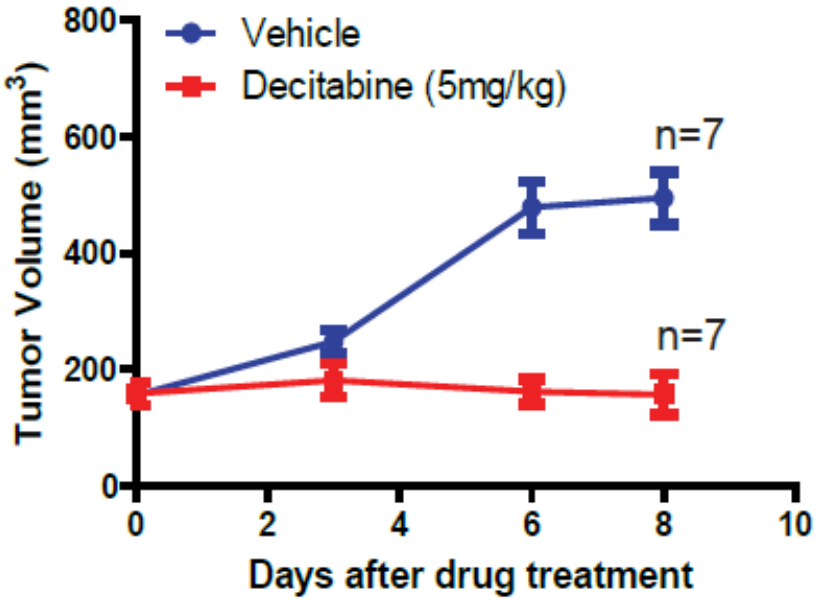
PDX taxane response correlates with original patient response



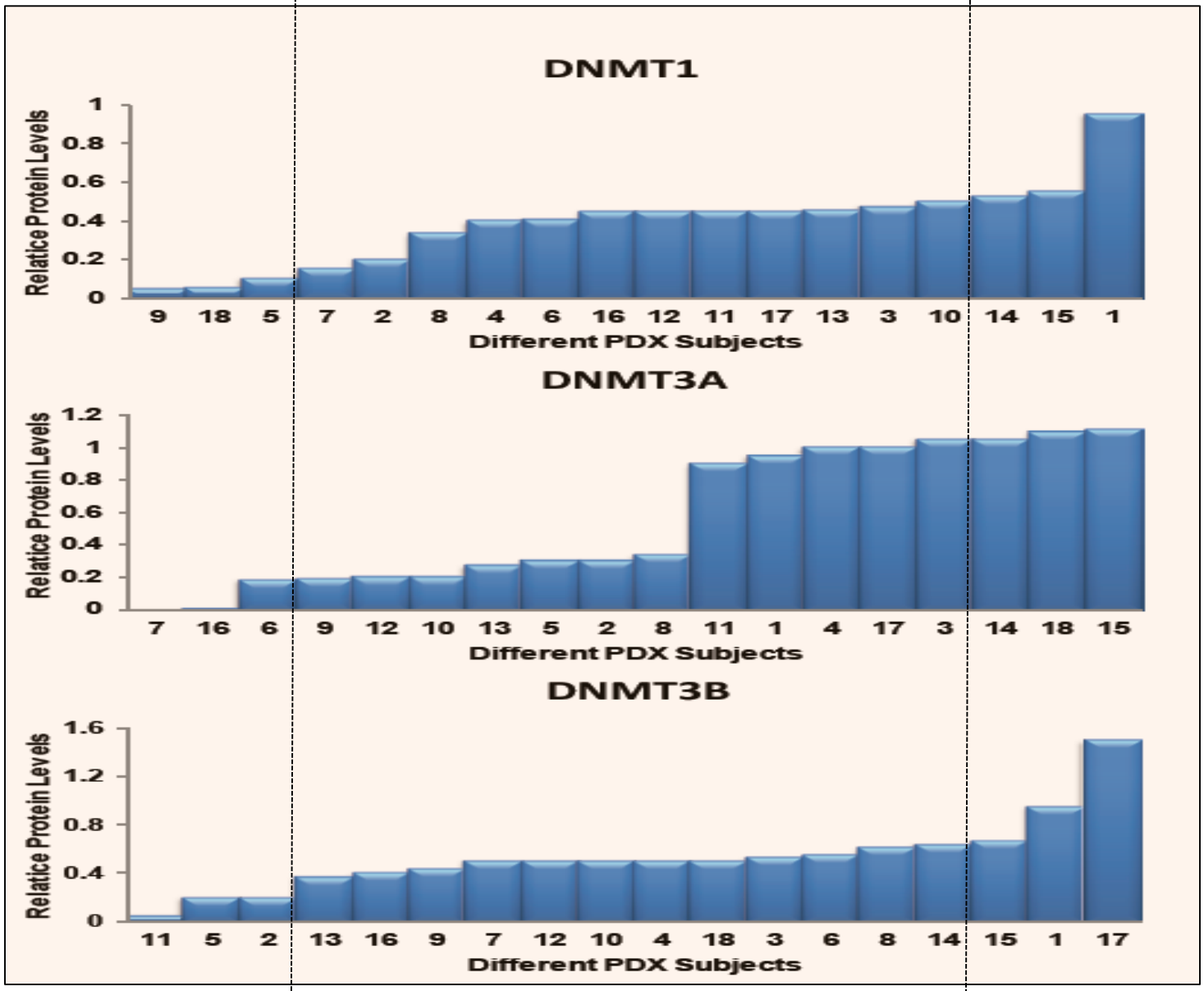
- **4 months following completion of chemotherapy**
- **October 2013**
- **Fatigue, diffuse pain, night sweats, failure to thrive**
- **PET scan**
- **Biopsy of liver: Metastatic breast cancer: ER/PR/HER2 negative**



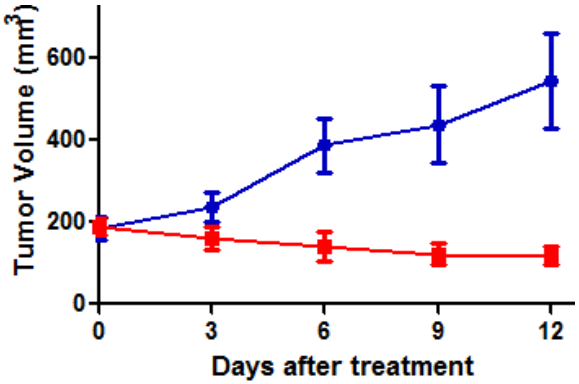

Before Therapy
Residual Tumor
DNMT3A
DNMT3B
Actin

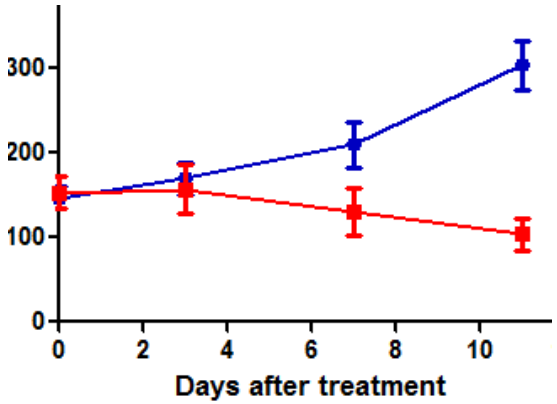

Vehicle
Decitabine (5mg/kg)
n=7
n=7
Tumor Volume (mm3)
800
600
400
200
0
0
2
4
6
8
10
Days after drug treatment

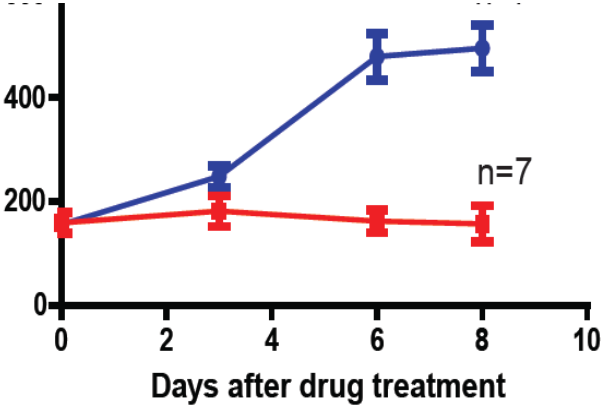
Hypothesis: Decitabine may be effective in the treatment of breast cancer patients, especially those with high levels of DNMTs.


DNMT1
Relatice Protein Levels
1
0.8
0.6
0.4
0.2
0
9 18 5 7 2 8 4 6 16 12 11 17 13 3 10 14 15 1
Different PDX Subjects
DNMT3A
Relatice Protein Levels
1.2
1
0.8
0.6
0.4
0.2
0
7 16 6 9 12 10 13 5 2 8 11 1 4 17 3 14 18 15
Different PDX Subjects
DNMT3B
Relatice Protein Levels
1.6
1.2
0.8
0.4
0
11 5 2 13 16 9 7 12 10 4 18 3 6 8 14 15 1 17
Different PDX Subjects

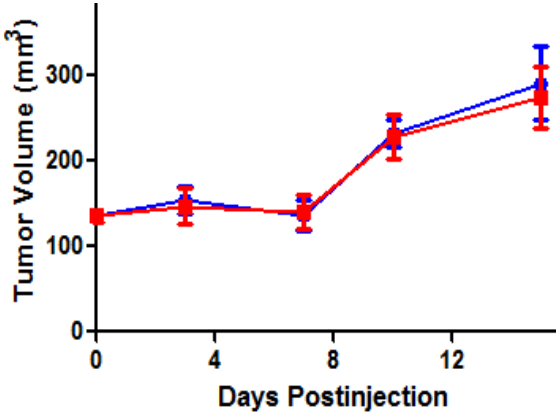
High DNMT Group

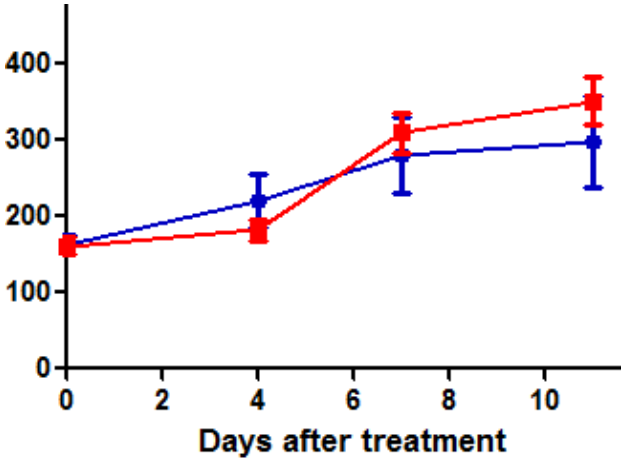


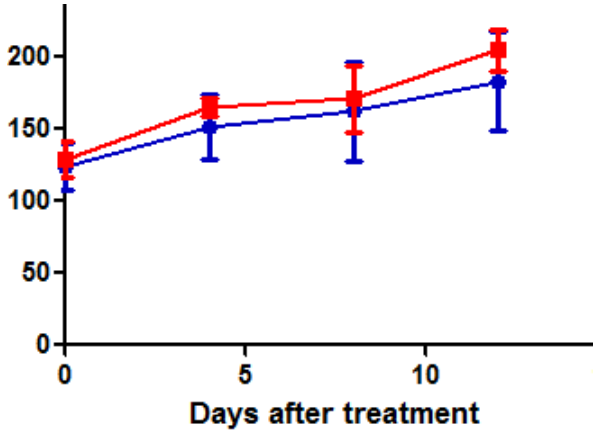




Low DNMT Group







Conclusions

- **Both germline and tumor genome can contribute to drug response.**
- **Integrated omics can help us to understand the drug resistance mechanisms and help us to accelerate drug development.**
- **Functional and mechanistic studies beyond biomarkers are crucial in the era of precision medicine.**
- **Team work is the key to the success.**

Acknowledgements

- **PGRN: Drs. Weinshilboum and Ingle, RIKEN Center for Genomics Science and NCIC-CTG**
- **Functional Genomics: Drs. Cairns and Tanda Dudenkov**
- **BEAUTY PIs: Drs. Goetz and Boughey**

  BEAUT Team
- **PDX and Biomarker Team: Drs. Jia Yu, Bo Qin and Ping Yin**
- **Stat and Bioinformatics Team: Dr. Rani Kalari**
- **Funding: Mayo-NIH Pharmacogenomics Research Network (PGRN), NCI R01 and Mayo Breast SPORE and Center for Individualized Medicine (CIM)**







Dr. Richard Weinshilboum

Dr. Matthew Ames

Dr. Scott Kaufmann

Dr. Rochelle Long

ASCPT